Risk stratification for sudden cardiac death in childhood hypertrophic cardiomyopathy

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Declaration

I, Gabrielle Norrish, confirm that the work presented in this thesis is my own. Where information has been derived from other sources, I confirm that this has been indicated in the thesis.

Acknowledgements

I would like to thank my supervisor, Dr Juan Pablo Kaski, for introducing me to the world of inherited cardiac diseases and his endless enthusiasm and encouragement throughout the last 4 years. You have inspired me and provided an exemplary example of how research can be integrated with excellent clinical care.

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This work would not have been possible without the collaborative efforts of a large number of individuals at each of the International Paediatric Hypertrophic Cardiomyopathy Consortium sites. Thank you for sharing your knowledge and time so generously.

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Abstract

Sudden cardiac death (SCD) is the most common cause of mortality in childhood onset hypertrophic cardiomyopathy (HCM). Despite this, our understanding of risk factors and ability to identify those at highest risk is limited. The aims of this thesis were to perform the first systematic investigation of SCD in childhood HCM and develop a novel paediatric-specific risk model.

Using a large, retrospective, international multi-centre cohort of children with HCM this thesis firstly describes the natural history of childhood onset disease. The second part of the thesis presents the results of the first systematic review and meta-analysis of clinical risk factors for SCD in childhood HCM identifying four major clinical risk factors; previous malignant arrhythmic event, unexplained syncope, non-sustained ventricular tachycardia (NSVT) and extreme left ventricular hypertrophy. In the third part of this thesis, the current risk stratification guidelines are shown to have only moderate discriminatory ability (C statistic 0.62 (95% CI 0.55-0.70)) with a corresponding 5-year positive and negative predictive value of 9.0% and 94.5%. Thus, a novel pediatric model is developed using pre-selected clinical variables from the meta-analysis (unexplained syncope, NSVT, left atrial diameter, maximal wall thickness and left ventricular outflow tract gradient). The model's ability to predict risk at 5 years was validated; C statistic 0.69 (95% CI, 0.66-0.72) and calibration slope 0.98 (95% CI, 0.59-1.38). In the final part of this thesis the role of additional novel risk factors, including 12-lead electrocardiograph and genotype, are explored.

In summary, this thesis is the first systematic and comprehensive investigation of risk factors for SCD in childhood HCM. Important differences between adult and childhood disease are described and the first validated risk model for use in paediatric clinical care is presented.

Impact statement

Hypertrophic cardiomyopathy is the second commonest cardiomyopathy presenting during childhood and is associated with significant morbidity and mortality. The most common cause of death in this young population is sudden cardiac, occurring at a rate of 1-2% per year. Implantable Cardiac Defibrillators (ICDs) can treat malignant heart rhythms and be life-saving, but identifying which patients will benefit from this treatment remains a significant challenge for clinicians.

The research underlying this thesis represents the first systematic assessment of risk factors and risk stratification methods for sudden death in childhood hypertrophic cardiomyopathy. I have shown that important differences exist between childhood and adult disease with the implication that paediatric specific risk stratification tools are required. I established a large, multi-centre retrospective cohort of over 1000 children worldwide with HCM to overcome the inherent challenges of studying a relatively rare event, such as SCD, in a rare disease. Current risk stratification guidelines had only modest discriminatory ability to identify those at highest risk leading to unnecessary ICD implantation in many. This thesis presents the first validated risk stratification model for SCD in childhood disease allowing clinicians for the first time to calculate individualised estimates of 5-year sudden death risk.

This work has been disseminated within the scientific community by publishing in high impact journals and presentation at both paediatric and adult conferences. The developed HCM Risk-Kids model is widely available at www.hcmriskkids.org.
The results of this research have direct implications for patient care and adoption of the HCM Risk-Kids model in clinical practice will assist shared-decision making between doctors, patients and their families.

As part of this work, data on the largest number of children with HCM in the world has been collected in The International Paediatric Hypertrophic Cardiomyopathy Consortium (IPHCC). In addition to developing important collaborative links with 45 expert centres worldwide, this consortium represents a unique resource that can be utilised by researchers in future studies investigating childhood HCM.

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List of Abbreviations

2D	2 dimensional
4C	Four Chamber
ABPRE	Abnormal Blood Pressure Response to Exercise
ACMG	American College of Medical Genetics
AD	Autosomal Dominant
AE	Adverse Event
AHA/ACCF	American Heart Association/American College of Cardiology Foundation
AP	Anteroposterior
AR	Autosomal Recessive
ASA	Alcohol Septal Ablation
ASH	Asymmetric septal hypertrophy
AV	Atrioventricular
BP	Blood Pressure
BSA	Body Surface Area
BVH	Biventricular Hypertrophy
CCF	Congestive Cardiac Failure
CFC	Cardiofaciocutaneous syndrome
CI	Confidence Interval
CMRI	Cardiac Magnetic Resonance Imaging
CS	Costello Syndrome
CV	Cardiovascular
CVA	Cerebrovascular Accident
CVD	Cardiovascular Death
DCM	Dilated Cardiomyopathy
DMD	Duchenne Muscular Dystrophy
DNA	Deoxyribonucleic acid
ECG	Electrocardiogram
EPS	Electrophysiology Study
ESC	European Society of Cardiology
FA	Friedreich's ataxia
FHx	Family History
FS	Fractional Shortening
FU	Follow Up
GSD	Glycogen Storage Disease
HCM	Hypertrophic Cardiomyopathy
HR	Heart Rate
ICD	Implantable Cardioverter Defibrillator
IEM	Inborn error of metabolism
IQR	Interquartile range
IVST	Interventricular Septal Thickness
LA	Left atrium
LBBB	Left Bundle Branch Block

LGE	Late Gadolinium Enhancement
LV	Left Ventricle
LVH	Left Ventricular Hypertrophy
LVOT	Left Ventricular Outflow Tract
LVPWT	Left Ventricular Posterior Wall Thickness
MACE	Major Arrhythmic Cardiac Event
MAPK	Mitogen-Activated Protein Kinase-dependent signalling pathway
MELAS	Mitochondrial Encephalomyopathy, Lactic Acidosis, and Stroke-like episodes
MERRF	Myoclonic Epilepsy with Ragged Red Fibres
MI	Multiple Imputation
MLVWT	Maximal Left ventricular Wall Thickness
MPS	Mucopolysaccharidoses
MV	Multivariate
NACCS	National Australian Childhood Cardiomyopathy Study
NGS	Next Generation Sequencing
NHS	National Health Service
NPV	Negative Predictive Value
NS	Noonan Syndrome
NSML	Noonan Syndrome with Multiple Lentigines
NSVT	Non-Sustained Ventricular Tachycardia
NYHA	New York Heart Association
OR	Odds Ratio
P/LP	Pathogenic/Likely Pathogenic
PCMR	Pediatric Cardiomyopathy Registry
PPM	Permanent Pacemaker
PPV	Positive Predictive Value
PS	Pulmonary Stenosis
PSAX	Parasternal Short Axis
PSLAX	Parasternal Long Axis
RBBB	Right Bundle Branch Block
RV	Right Ventricle
RVOTO	Right Ventricular Outflow Tract Obstruction
SCD	Sudden Cardiac Death
TTE	Transthoracic Echocardiogram
TX	Transplant
UK	United Kingdom
UV	Univariate
VF	Ventricular Fibrillation
VLCADD	Very long-chain acyl-coenzyme A dehydrogenase deficiency
VT	Ventricular Tachycardia
VUS	Variant of Unknown Significance

Personal contributions

This project was conceptualised by Dr Juan Pablo Kaski and Professor Perry M. Elliott. I was responsible for the subsequent conduct of all aspects of the study including; obtaining ethical approval, recruitment to the International Paediatric Hypertrophic Cardiomyopathy Consortium, data collection and data analysis. I would like to acknowledge and thank the following individuals for their contributions:

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Aims of thesis

This is the first comprehensive systematic investigation of sudden cardiac death in childhood hypertrophic cardiomyopathy. The specific aims were to:

- Develop an international multi-centre cohort of children (under the age of 16 years at presentation) with hypertrophic cardiomyopathy to allow the description of the natural history of childhood disease and investigation of paediatric specific risk factors for sudden cardiac death
- Perform a systematic review and meta-analysis of risk factors for sudden cardiac death in childhood hypertrophic cardiomyopathy
- Validate current risk stratification guidelines for sudden cardiac death in childhood hypertrophic cardiomyopathy
- Develop and validate the first paediatric specific risk model for sudden cardiac death in childhood hypertrophic cardiomyopathy

Chapter 1: Introduction

1.1 Overview of Paediatric Hypertrophic Cardiomyopathy (HCM)

Definition

The first descriptions of Hypertrophic Cardiomyopathy (HCM) as we now recognise it are attributed to a pathologist, Donald Teare, who 60 years ago compared it to 'a tumour of the heart'[1]. Early descriptions highlighted not only the pathological histological hallmark of myocyte disarray, but also identified it as a familial disease linked to early sudden death. Over time the histological description has expanded to include a classical triad of micro- and macroscopic findings which include; myocyte disarray, small vessel disease and fibrosis [2, 3].

In clinical practice, HCM is defined as increased left ventricular wall thickness (on transthoracic echocardiogram (TTE)) that is not explained by abnormal loading conditions [4, 5]. During childhood, the definition takes into account somatic growth and uses body surface area corrected normal values for left ventricular wall thickness. A diagnosis of HCM during childhood is therefore made if maximal left ventricular wall thickness (MLVWT) is greater than 2 standard deviations (≥2 Z scores) above the population mean in the absence of abnormal loading conditions [4, 5].

1.1.1 Incidence and epidemiology

Although initially considered a rare disease, adult community echocardiographic screening studies in diverse geographic cohorts have reported a prevalence of HCM of 1 in 500 (0.2%)[6-9]. Indeed, with advances in clinical practice, including genetic testing and the availability of high resolution cardiac magnetic resonance imaging (CMRI), one recent study has suggested that the true prevalence may be as high as

1 in 200[10]. These studies include individuals who are asymptomatic and without a clinical diagnosis. The prevalence of clinically diagnosed HCM is estimated to be much lower (0.03% in a recent North American study [11]) and likely varies depending on the underlying health system and local screening practices.

The true prevalence of HCM in childhood is unknown. Population epidemiological studies from Finland, Australia and North America have reported an annual incidence of between 0.24-0.47/100,000[12-14] making it the second most common cardiomyopathy (after dilated cardiomyopathy (DCM)) presenting during childhood. Up to 60% of childhood disease presents in infancy with the age of presentation being at least partly determined by the underlying aetiology (Section 1.3 below). Adult and childhood population studies have both reported a male preponderance, which in childhood is estimated to be between 60-75%[13, 15, 16]. This has been hypothesised to be explained by the modifier effect of sex hormones[17], however its presence in pre-adolescent cohorts suggests other genetic or epigenetic factors contribute.

1.1.2 Aetiology and pathogenesis

The European Society of Cardiology (ESC) recommends dividing the underlying aetiology of cardiomyopathies into familial (genetic) and non-familial (non-genetic) forms[18]. In common with adult disease, the underlying aetiology in childhood is most commonly genetic; however it is recognised to be more heterogeneous and includes inborn errors of metabolism (IEM), RASopathy syndromes, neuromuscular disease as well as sarcomeric protein disease (Table 1 below). Aetiology has been shown to be an important determinant of the both the clinical features and long-term outcomes of childhood disease (Section 1.4 below).

Sarcomeric protein disease

Up to 60% of adults with HCM have a disease-causing variant in a sarcomeric protein gene identified on genetic testing, which is inherited as an autosomal dominant trait. The majority of variants (75-80%) are found in myosin-binding protein C (MYBPC3) or β-myosin heavy chain (MYH7). Approximately 15-20% of patients have variants in other sarcomeric proteins including; cardiac troponin T (TNNT2), cardiac troponin I (TNNI3), α-tropomyosin (TPM1), α-cardiac actin (ACTC), essential myosin light chain (MYL3), regulatory myosin light chain (MYL2), cardiac troponin C (TNNC1) and α-myosin heavy chain (MYH6) [19, 20]. A small number of patients (<1%) are recognised to have disease-causing variants in z-disk proteins, such as muscle LIM protein (CSRP3), or genes involved in calcium handling, such as phospholamban.

Sarcomeric protein variants exhibit variable and age-related penetrance which is incompletely understood[21, 22]. Historically it was believed that sarcomeric disease was uncommon in childhood and that left ventricular (LV) hypertrophy did not develop until adolescence or early adulthood. This was mainly based on the observation that in a small cohort (n=39) of patients with familial disease, progression of LV hypertrophy was most commonly seen in later childhood[23]. As a result, current clinical guidelines recommend commencing family screening for first degree child relatives from the age of 10 years[4, 5]. However, two separate studies have now shown that over 50% of disease in childhood is caused by variants in sarcomeric protein genes[24, 25] with the most common variants being found in MYBPC3 or MYH7. A recent study assessing the yield of clinical screening in childhood reported that 5% of first degree relatives met diagnostic criteria for HCM, the majority of which (72%) were under the age of 12 years at diagnosis with a significant proportion (11%) under the age of 10 [26, 27]. Therefore, whilst the

aetiology of childhood HCM is more variable, there is now recognition that in common with adults the majority of disease, even in infants and young children, is secondary to sarcomeric protein variants.

RASopathies

The RASopathies are a group of heterogeneous and overlapping clinical syndromes caused by variants in the mitogen-activated protein kinase-dependent signalling pathway (MAPK), which regulates cell proliferation, differentiation, survival and apoptosis (Table 1 below) [28]. The most common RASopathy syndrome is Noonan syndrome (NS), which has an estimated incidence of 1:1,000 – 1:2,500 live births. A disease-causing variant in the MAPK pathway is identified in 70-80% of patients with clinical features of RASopathies, most commonly Protein Tyrosine Phosphatase Non-Receptor Type 11 (PTPN11), RAF1, Son of Sevenless (SOS1) or Serine/threonine-protein kinase B-raf (BRAF). This group of conditions is characterised by extra-cardiac features including facial dysmorphia, skeletal deformities, skin and hearing abnormalities, and intellectual disability. Up to 80% have cardiac involvement, which includes valvar pulmonary stenosis (PS) (≈60%), HCM (≈30%), septal defects (≈5%) and atrioventricular (AV) canal abnormalities (≈4%)[29, 30].

Overall this group of syndromes account for less than 10% of all paediatric HCM[12, 14-16], however up to 20% of those with infantile disease may have an underlying RASopathy syndrome[16, 31]. Population studies have reported that one third of patients with a RASopathy syndrome have HCM[29], however the prevalence varies depending on the underlying gene involved (eg RAF1 >75%, RIT1≈50-60%, KRAS ≈30%, PTPN11≈ 10, SOS1≈ 10%)[32-36].

Neuromuscular disorders

Neuromuscular diseases account for less than 10% of all paediatric HCM. Friedreich's Ataxia (FA) is the most commonly associated diagnosis and is characterised by a progressive ataxia, sensory neuropathy, areflexia and LV hypertrophy. Although neurological symptoms precede cardiac involvement in the majority, up to 90% have cardiac involvement under the age of 18 years[37]. Presentation with HCM in infancy is rare and the diagnosis is typically made in adolescence [15, 16].

Inborn errors of metabolism

IEM incorporates a large number of heterogeneous conditions (Table 1 below), which are individually rare but have a combined incidence approaching 1 in 800 live births[38]. Most IEM are multi-system diseases characterised by dysmorphic features, neurological involvement, myopathy, hepatosplenomegaly and developmental delay. As a group, they account for up to 10% of all paediatric HCM [12, 14-16]. The majority of IEM present in infancy apart from notable exceptions, for example Anderson Fabry disease.

	Aetiology			Inheritance pattern	Proportion of childhood HCM
Familial/genetic	Sarcomeric protein gene variants			AD	>50%
	RASopathy syndromes	Noonan Syndrome (NS)		AD	<10%
		Costello Syndrome (CS)		AD	
		Noonan Syndrome with Multiple Lentigines (NSML)		AD	
		Cardiofaciocutaneous syndrome (CFC)		AD	
	Inborn errors or metabolism	Disorders of glycogen metabolism	Glycogen storage diseases (eg Pompe disease, GSD III)	AR	<10%
			AMP kinase disease	AD	
			Danon disease	X-linked	
		Lysosomal storage disease	MPS I, II, III, VII	AD/AR/X-linked	
		Disorders of fatty acid metabolism	Carnitine transport defects	AR	
			Fatty acid oxidation defects eg VLCADD	AR	
		Mitochondrial disorders	Respiratory chain complex deficiencies	AR	
			Mitochondrial DNA abnormalities (eg MELAS, MERRF, Leigh syndrome, Kearns Sayre Syndrome)	Matrilineal	
			methylglutaconic aciduria (eg Barth syndrome, Sengers syndrome)	X-linked, AR	
	Neuromuscular disease	Friedreich's Ataxia		AR	<10%
Non-familial/ Non-genetic	Obesity			NA	Negligible
	Infants of diabetic mothers			NA	
	Amyloid			NA	
	Mimics – Steroid induced LVH, Athletic training			NA	

Table 1 Aetiology of childhood hypertrophic cardiomyopathy

AD (Autosomal dominant), AR (Autosomal recessive), GSD (Glycogen storage disorder), MPS (Mucopolysaccharidoses), VLCADD (Very-long-chain acyl-CoA dehydrogenase deficiency), DNA (deoxyribonucleic acid), MELAS (Mitochondrial Encephalopathy, Lactic acidosis, and Stroke-like episodes), MERRF (Myoclonic epilepsy with ragged red fibred)

1.1.3 Clinical characteristics

Our current understanding of the clinical characteristics of childhood HCM is primarily derived from North American [16] and Australian registry studies[15], which described 855 and 80 patients diagnosed with HCM under the age of 18 years respectively. They have provided valuable information on the wide spectrum of childhood disease, including phenotypic and clinical differences characteristic of particular aetiologies (Table 2 below). Detailed descriptions of phenotype progression, natural history other than mortality, and the impact of current screening practices on outcomes is currently poorly described.

Mode of presentation

Whilst the diagnosis of HCM is imaging based, the reason for presentation is varied and partly determined by the underlying aetiology. Although sarcomeric disease can present at any age, those with an underlying RASopathy or IEM often present in infancy, and FA commonly presents in later childhood [15, 16, 31, 39, 40]. In infancy, the diagnosis is often made following presentation with heart failure symptoms, including dyspnoea, poor feeding and failure to thrive, or during evaluation for a heart murmur [15, 39, 41, 42]. Older children may be referred for symptoms (such as chest pain, palpitations, syncope, dyspnoea), ECG abnormalities or through family screening[15, 41, 43]. A small proportion (3-4%) are diagnosed following a resuscitated cardiac arrest[13, 41]. As many syndromic causes of childhood disease are associated with extra-cardiac abnormalities, their co-existence may suggest a particular aetiology (Table 2 below).

Clinical features

Childhood is a time of significant somatic growth which is reflected in a rapidly changing cardiac phenotype. Population studies have provided useful descriptions of

the cardiac phenotype at baseline, but our knowledge of how this changes during childhood remains limited.

Left ventricular hypertrophy

Left ventricular hypertrophy (LVH), as defined by a maximal LV wall thickness greater than 2 standard deviations (Z scores) above body-surface area corrected mean, is a pre-requisite for the diagnosis of HCM [4, 5] yet the pattern of hypertrophy varies. Asymmetric septal hypertrophy (ASH) is present in the majority (≈70%)[15, 16, 41], whilst concentric hypertrophy is more common in those with syndromic (RASopathy, IEM or FA) disease [15, 16, 31, 44]. Up to one quarter have concomitant right ventricular hypertrophy [15], which is frequently associated with concentric LVH and an underlying RASopathy [30, 31].

Our understanding of the progression of LVH during childhood is incompletely understood. Maron et al [23]described the progression of LVH in 39 patients with familial childhood disease reporting that increases in hypertrophy were seen more frequently in adolescence. This paradigm is reflected in current screening guidelines. Of note, this study only contained 10 patients under the age of 12 years, 40% of whom had hypertrophy at baseline. No large studies have systematically described the changing phenotype of familial childhood disease to date. However, recent studies describing the follow up of either childhood relatives[26, 27] or genotype positive children[45, 46], have demonstrated that in familial disease, once present, LVH increases throughout childhood and in some cases may start to regress in early adulthood. No study has described the progression of hypertrophy in childhood RASopathy patients and little is known about individual IEM. The exception to this being Danon disease in which male patients are known to develop early and

progressive severe concentric HCM necessitating cardiac transplantation at an average age of 17.9 years[47, 48].

Left ventricular outflow tract obstruction

Left ventricular outflow tract obstruction (LVOT obstruction) was historically an essential part of the diagnosis of HCM as reflected by diagnostic terms including idiopathic hypertrophic subaortic stenosis or muscular subaortic stenosis [49, 50]. It is defined as a maximal LVOT gradient, as measured using Doppler echocardiography, above 30mmHg at rest or during provoking manoeuvres that alter LV loading conditions (such as Valsalva or exercise) [4]. However, haemodynamic effects are typically only seen above 50 mmHg[51]. Approximately one third of adults have LVOT obstruction at rest, and a gradient is elicited in a further third during exercise [52-54]. The reported prevalence of LVOT obstruction in childhood varies widely (22-60%)[41, 55-60], likely reflecting the heterogeneous nature of childhood disease and differing definitions of LVOT obstruction in published cohorts. Indeed, children with RASopathy syndromes are more likely to have obstructive disease compared to 'idiopathic' cohorts [29, 61]. Similar to adult cohorts, a provocable gradient has been shown in 50-60% of symptomatic children with no gradient at rest [62]. The mechanisms of LVOT obstruction are often multifactorial and include an anatomically narrowed LVOT, basal anteroseptal hypertrophy and systolic anterior motion of the mitral valve. Importantly, children with an underlying RASopathy syndrome frequently have anatomically complex LVOT obstruction due to a polyvalvulopathy with abnormal chordal attachments of the mitral valve[29]. Concomitant right ventricular outflow tract obstruction (RVOTO) secondary to hypertrophied RV outflow tract bundles or pulmonary stenosis is also commonly reported in this group of patients[30, 31].

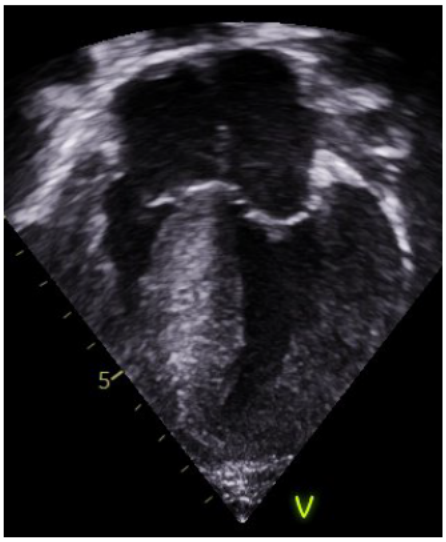
LVOT obstruction causes symptoms of chest pain, exertional dyspnoea and syncope secondary to reduced cardiac output causing myocardial ischaemia and changes in LV filling pressures. Management is typically focused on relieving symptoms and treating asymptomatic obstruction during childhood is controversial due to conflicting reports regarding its effect on long-term prognosis. Accepted medical therapy includes the use of beta-blockers[63], disopyramide[64] and calcium channel blockers[65, 66]. For those with refractory symptoms or fixed obstruction, surgical myectomy has been shown to be effective at reducing the gradient and providing symptomatic relief with low operative mortality or morbidity in experienced centres[67-69]. A recent single centre experience of surgical relief during infancy (n=12), two thirds of which had a RASopathy syndrome, reported no significant difference in early or late mortality compared to those operated on later in childhood but with a trend to requiring re-operation in the infant group[70]. Other invasive gradient reduction therapies used in adult populations, such as alcohol septal ablation (ASA) therapy, are not recommended in childhood as the long-term effect of such therapies are unknown [71].

Left ventricular function

Systolic function in HCM is typically described as hyper-dynamic with preserved global measures of LV function (eg ejection fraction). However, children with an underlying diagnosis of an IEM (particularly mitochondrial disease) may present with, or develop, progressively impaired LV function [72, 73]. In a minority of adult patients (≈5%) evolution to a dilated-hypokinetic phase with LV dilatation, systolic dysfunction and LV wall thinning has been described [74]. Although this is the most common cause for heart transplantation in children with HCM [75], this is exceedingly rare in non-syndromic disease.

Abnormal diastolic parameters have been observed to precede the development of hypertrophy in sarcomere mutation carriers and contribute to a ‘pre-clinical’ phenotype[76]. Diastolic dysfunction in established disease causes elevated LV end-diastolic pressures and an increase in the left atrial volume. In adult patients, left atrial size is an important predictor of atrial fibrillation and thromboembolic risk[77, 78], both of which are uncommon during childhood. However, impaired diastolic function is an important cause of morbidity in childhood disease causing symptoms of dyspnoea and atypical chest pain.

A)



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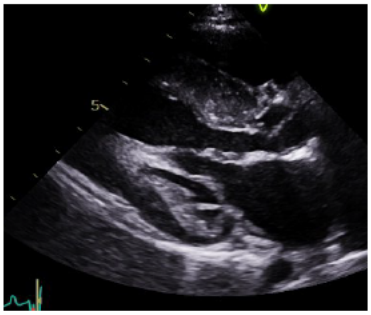

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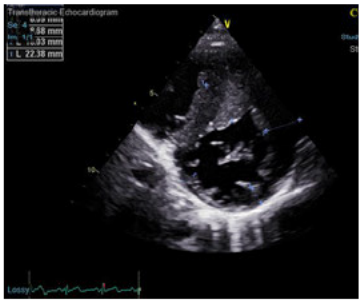


Aetiology	Presenting age	Extra-cardiac features	Cardiac phenotype	
			ECG features	Echo features
Sarcomeric	Throughout childhood	Nil		ASH, LVOTO (20-60%), preserved systolic function, impaired diastolic function
RASopathy syndrome	Infancy	Facial dysmorphia, Learning difficulties, skeletal abnormalities	Superior axis, BVH	Concentric/BVH, valvulopathy complex LVOTO +/- RVOTO. Preserved systolic function, impaired diastolic function
IEM	Infancy	Learning difficulties, neuromuscular symptoms, deafness, encephalopathy, metabolic abnormalities (eg hypoglycaemia, metabolic acidosis)	Pre-excitation, BVH, AV conduction disease	Concentric/BVH, valvulopathy (MPS), impaired systolic function
Neuromuscular	Adolescence	Ataxic gait, sensory neuropathy, areflexia	Small voltages	Concentric, LVOTO uncommon

Table 2 Clinical characteristics of childhood hypertrophic cardiomyopathy by aetiology

Figures A) Concentric BVH in a patient with mitochondrial disease, B) Concentric LVH with dysplastic thickened mitral valve leaflets in a patient with Noonan syndrome, C) ASH in a patient with a pathogenic MYH7 variant

1.1.4 Long-term outcomes and mortality

Early publications from small highly selected tertiary centre populations reported that the long-term prognosis of childhood HCM was poor with annual mortality rates up to 7% [79, 80]. Over time, reflecting what has been seen in the adult literature, reported mortality rates from larger unselected populations have fallen. The most recent, large scale population based studies have reported an overall annual mortality rate of under 3%[15, 16]. However, significant variability in outcomes exists, which can be partly explained by the underlying aetiology and age of presentation. Publications from the Pediatric Cardiomyopathy Registry (PCMR) have described that patients with 'idiopathic' or non-syndromic disease have a relatively good prognosis with estimated 1 and 5 year survival of 94.4% (95% 92.4-96.4%) and 82.2% (95% CI 76.2-88.2%) [16]. In comparison, those with an underlying IEM or RASopathy have repeatedly been shown to have a worse prognosis with a 1 year survival of 53.6 % (95% CI 41.3-66) and 82.4% (95% CI 73.0-91.9) respectively[16, 31]. Early presentation, in infancy, has also been shown to be associated with worse prognosis. This might partly be explained by the high proportion of syndromic disease in this population, but is also likely to also be a reflection of disease severity as one year mortality rates for non-syndromic infantile disease is high (19% (95% CI 14.0-24.0)). [15, 16, 39, 40, 42, 66, 81]. Importantly, although infantile disease is associated with worse overall prognosis, early PCMR publications described that for those children that survived to 1 year, mortality plateaued and long-term survival was similar to those presenting later in childhood[16]. More recently, this observation has been confirmed in the published 15 year follow up for 80 patients in the National Australian Childhood Cardiomyopathy Study (NACCS) registry (of whom 61% presented in infancy)[81]. They described significant mortality and morbidity within

the first year of diagnosis, with 14% of patients reaching a composite outcome of death or cardiac transplantation. Yet, for patients that survived 1 year, long term outcomes were good with an annual mortality or cardiac transplantation rate of 0.4%. This compares favourably to estimates of cardiovascular mortality of 1-2% per year in adult patients[82].

The cause of death in childhood HCM has been shown to differ depending on the age of presentation. Heart failure-related deaths are responsible for the majority of early deaths, particularly in infancy[15, 81]. The underlying pathogenesis of heart failure likely differs by aetiology. Patients with an IEM have been described to develop a dilated, hypokinetic phenotype [72, 73], however progression to an end-stage hypokinetic phase during childhood is rare in familial non-syndromic disease[74]. Heart failure in these patients is therefore most likely to be secondary to diastolic failure with preserved systolic function although this has not been well described [15]. Outside of infancy, sudden cardiac death (SCD) is the most common cause of mortality [43, 81] and is discussed in detail below. Other types of cardiovascular deaths reported in adult populations, such as following thromboembolic events, are rarely seen during childhood.

Reflecting the difference in cardiac and non-cardiac disease phenotype, the cause of death also varies according to underlying aetiology. Heart failure related deaths are more common in patients with syndromic disease (IEM or RASopathy) whilst non-cardiac deaths are seen most frequently in patients with IEM who frequently have multi-systemic involvement [15, 16, 31, 40, 81].

Predicting the outcome of children with HCM is challenging because of the significant variability in age, aetiology, cardiac phenotype and natural history. Whilst aetiology and age of presentation have been shown to partly explain the variability in

outcomes, retrospective registry studies have provided valuable insights into additional or additive clinical risk factors. Multiple studies have shown that presenting with symptoms of congestive cardiac failure is associated with a higher cardiovascular mortality over follow up[16, 40, 42]. Certain phenotypic features have also been linked to a worse prognosis including concentric LVH, biventricular involvement, severe left ventricular hypertrophy and impaired systolic function [15, 40, 81]. As many of these clinical risk factors are associated with non-syndromic disease, it is likely that both the cardiac phenotype and aetiology are important for long-term outcome. A detailed exploration of the risk factors for sudden cardiac death can be found in Section 1.2.

Cardiac transplantation

Cardiomyopathies are the most common indication for heart transplantation listing during childhood, yet HCM constitutes only a small proportion (≈6%) of this cohort[83, 84]. Indication for transplant listing includes refractory heart failure, which in infants is typically due to massive hypertrophy and small cavity size[85], and arrhythmias[83]. Compared to other cardiomyopathies, HCM have a higher waitlist mortality [84] and are a high-risk population for mechanical support [86]. Long term survival following transplant in childhood is poorly described and limited to small cohorts (n=49) but may be lower than reported for other cardiomyopathies [83] .

1.2 Sudden cardiac death in HCM

1.2.1 Incidence

From its early descriptions, HCM was synonymous with poor prognosis and sudden unexpected cardiac death often occurring in young patients[50, 87]. High profile deaths of young athletes portrayed in the media have further served to highlight HCM as a malignant diagnosis[88]. Indeed, HCM is recognised to be an important cause of SCD in young athletes even in the context of pre-participation screening [89, 90].

In adult cohorts, early reports described incidence rates of SCD of up to 6% per year [87, 91, 92]. Yet, reflecting initial bias from small tertiary centre populations, the published incidence rate in adults has fallen over time and is now estimated to be less than 1% per year [82, 93]. Similarly, initial reports from small, highly selective paediatric cohorts described SCD incidence rates of up to 7% per year [41, 79, 80] but more recent, large population studies have reported an annual SCD rate of 1-2% [16, 41, 60, 81](Figure 1 below). Outside of infancy, SCD is the most common cause of mortality, and is a risk that is present throughout childhood [60, 81]. A single study has described a higher incidence of SCD in the pre- and early adolescent years (aged 9-14yrs) of up to 7.2% per year [43] but this has not been confirmed in other populations. Overall, the incidence of SCD in childhood is higher than that described in adults. The underlying reasons for this observation have not been fully explored but may reflect early presentation of severe disease.

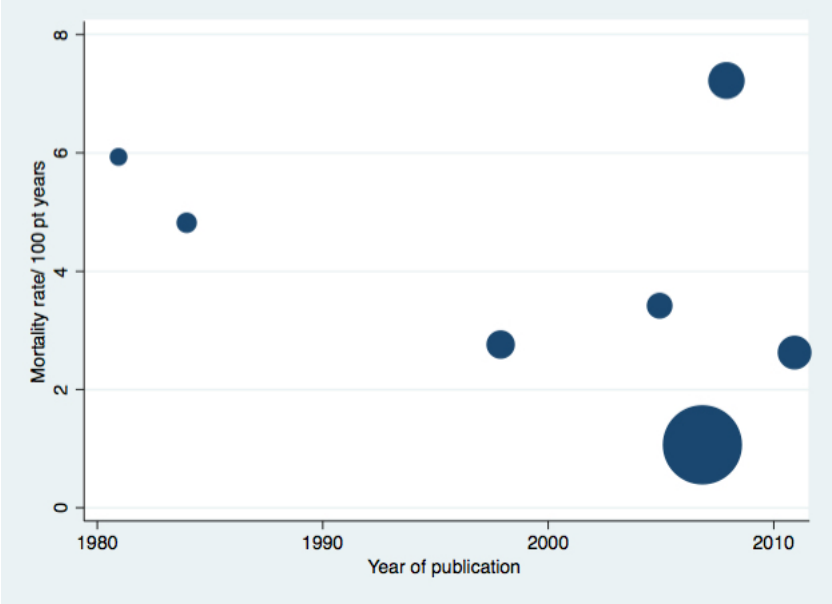


Figure 1 Published rates of sudden cardiac death childhood Hypertrophic Cardiomyopathy over time
Study size is represented by the size of marker[15, 16, 40, 59, 79, 80, 94]

1.2.2 Pathophysiology

The hallmark macroscopic and microscopic histological features of HCM include myocyte disarray, fibrosis and small vessel disease [2]. In a post-mortem study of 75 patients with HCM Varnava et al [95]reported that SCD was associated with the extent of myocyte disarray as well as clinical features of myocardial ischaemia before death (e.g. chest pain and ST depression on exercise). Myocardial fibrosis was more common in those with heart failure deaths but also associated with clinically detected ventricular arrhythmias.

Whilst is not surprising that this triad of pathological findings creates a pro-arrhythmic substrate, the mechanism by which it translates into an increased risk of ventricular arrhythmias is incompletely understood. Electrophysiological studies have described increased fractionation of RV paced beats, creating a substrate for re-entry tachycardias [96]. LV hypertrophy likely causes dispersion of repolarisation, myocardial disarray disrupting cell alignment, and areas of fibrosis creating a localised conduction block. In addition, mice models of HCM have shown altered

calcium homeostasis which reduces the effective refractory period and causes transmural dispersion of repolarisation predisposing to ventricular arrhythmias [97]. Follow up studies in human induced pluripotent stem cell cardiomyocytes (iPSC) with troponin T mutations have showed similar increased calcium sensitivity suggesting that this mechanism is relevant for human disease [98]. The observation that the overall incidence of SCD in HCM is low despite these universal underlying structural and biochemical abnormalities, suggests that transient electrical or structural changes (such as ischaemia, or dynamic outflow tract obstruction) in the context of a pro-arrhythmic substrate are the trigger for ventricular arrhythmias.

The primary underlying arrhythmia resulting in SCD is thought to be ventricular fibrillation (VF) or ventricular tachycardia (VT) in the majority of cases. This is predominantly based on the observation that patients implanted with implantable cardioverter defibrillators (ICDs) receive therapies for ventricular arrhythmias associated with haemodynamic compromise [99, 100]. Monomorphic ventricular tachycardia (VT) is described to be a particular feature of adult patients with LV apical aneurysms [101]. Yet recent studies analysing intracardiac ICD electrograms found that monomorphic VT was the presenting rhythm for the majority of appropriately delivered therapies, suggesting it may be more common than previously thought [102, 103]. The trigger for ventricular arrhythmias is not clearly understood but premature ventricular ectopics [103] and supraventricular tachycardias ([102, 104], including sinus tachycardia, have been implicated. Atrioventricular conduction disease[105] and asystole[106] have also been linked to SCD but their contribution is likely to be small.

1.2.3 Risk factors for SCD in adult HCM

Identifying patients at high risk of malignant ventricular arrhythmias who might benefit from preventative therapies is an important part of clinical care. Clinical risk factors in adult patients have been extensively investigated and described in multiple observational, retrospective, longitudinal cohort studies as summarised below. Of note, as SCD itself is a rare event, a composite end-point of SCD or equivalent events (including appropriate ICD therapies for tachyarrhythmias and resuscitated cardiac arrest) is widely accepted and used in the literature. Although this is a pragmatic approach, certain limitations including the true equivalence of ICD therapies and SCD must be recognised. The seven major risk factors recognised to be associated with an increased risk of SCD in adult disease are described below.

Previous malignant arrhythmia (VF/VT)

The most predictive clinical risk factor for SCD is having experienced and survived a previous malignant ventricular arrhythmia. Patients with previously documented VF or sustained VT (defined as VT lasting for longer than 30 seconds[4]) are at high risk of further arrhythmias with a 5-year event-free survival of approximately 60% described in small cohort studies [107, 108]. More recently, large longitudinal cohort studies of HCM patients with an ICD have estimated an annual incidence of appropriate ICD therapies between 4-10% compared to 2-3.6% in those without a malignant arrhythmic history [99, 100]. Reflecting the high-risk profile of this group of patients, clinical guidelines recommend a secondary prevention ICD in those who have previously experienced VF/VT [4, 5].

Maximum left ventricular wall thickness (MLVWT)

MLVWT, as determined by transthoracic echocardiography (TTE), has been shown in multiple studies to be associated with SCD [52, 82, 109-111] [112] and it is a well-accepted clinical risk factor. However, controversies about how it should be used for risk stratification remain as reflected in the current clinical guidelines (See section 2.6.1) [38]. Patients with extreme LVH (MLVWT ≥ 30mm) have been described to be at particular risk of arrhythmic events in some studies [112, 113] with a recent meta-analysis reporting a hazard ratio 3.1 (95% CI 1.8-4.4) [114]. This has led to the suggestion that, even in the absence of other clinical risk factors, the presence of extreme LVH could justify consideration for primary prevention ICD implantation [110]. This view has been challenged by others who report important additive effects of co-existing risk factors [82, 109] and highlight the small number of patients with extreme hypertrophy reported in the literature. In support of this contrasting view, a recent large multi-centre study demonstrated an inverted-U shaped relationship between MLVWT and SCD risk meaning that those with the most extreme hypertrophy (MLVWT≥35mm) did not have an increased risk of SCD compared to those with mild hypertrophy [115].

Left ventricular outflow tract obstruction (LVOT obstruction)

LVOT obstruction has been shown to be a risk factor for SCD in several large population studies [52, 53, 82, 116, 117]. The measured LVOT gradient is recognised to be dynamic and highly variable depending on the loading conditions and it is unclear what role provocable gradients, present in one third of adult HCM patients, play in risk stratification. As LVOT gradient is a potentially modifiable risk factor, there is interest in whether gradient reduction therapies (pharmacotherapy or surgical) could modify the risk of SCD. In one small single-centre study of HCM

patients with an ICD, those who underwent myectomy had a lower rate of appropriate therapies compared to those without surgery [118]. Two subsequent meta-analyses have shown that overall mortality following gradient reduction therapy (ASA or myectomy) equals that of HCM patients without LVOT obstruction, however the annual risk of SCD appears to be lower (0.4-0.5%) [119, 120]. Surgical gradient reduction strategies are not currently recommended for asymptomatic patients [4], but further studies are required to investigate the effect that these treatments have on long-term risk of SCD.

LA dilatation

Left atrial (LA) diameter has not been as extensively investigated as other echocardiographic measures, yet it has been found to be independently associated with SCD [82, 87, 116, 121, 122] and is included in current guidelines. LA diameter is also recognised to be a risk factor for atrial fibrillation [78].

Non-Sustained Ventricular Tachycardia (NSVT)

NSVT is defined as ≥3 consecutive ventricular beats occurring at a rate greater than 120bpm lasting less than 30 seconds [4]. It is detected in approximately 20-30% of patients with HCM on ambulatory electrocardiograph (ECG) monitoring [111, 123] and has been shown to be associated with SCD [52, 82, 111, 113]. A recent meta-analysis reported an overall hazard ratio of 2.89 (95% CI 2.21-3.58) [114] but NSVT appears to be particularly important when detected in younger patients (<30 years) [111]. There is no evidence that the frequency, duration or rate of NSVT provides further useful information for risk stratification [111]. Exercise induced arrhythmias (VF or NSVT) are rare, occurring in < 2% of patients who often have other features

of a severe phenotype (eg severe hypertrophy or dilated LA)[124]. When present they are associated with an additional increased risk of SCD occurring during follow up (HR 3.1 [95%CI 1.29-7.61], p value 0.01)[124].

Unexplained syncope

Syncope is a commonly reported symptom (15-20%) [125, 126] in HCM with a variety of underlying mechanisms which include haemodynamic (LVOT obstruction, reduced preload in context of diastolic dysfunction and vasovagal) or arrhythmic (bradyarrhythmias, supraventricular or ventricular arrhythmias) [127]. Identifying the underlying cause is challenging yet important as unexplained syncope, which is presumed to be secondary to ventricular arrhythmias, is associated with SCD [52, 82, 113, 114, 122, 124, 128]. The timing of syncope has also been shown to be important; recent syncope (within 6 months) is accompanied by a 5-fold increased risk of SCD whereas historical syncope is not associated with an increased risk [122].

Family history of sudden cardiac death

Initial descriptions of HCM included individuals with highly malignant family histories in which one or more members had died suddenly[50]. A family history of SCD has therefore historically been recognised as a risk factor for SCD and this has been confirmed in retrospective cohort studies [82, 110, 116, 125] leading to its inclusion as a major risk factor in clinical guidelines. Its interpretation and use as a risk factor has been complicated by differing definitions in published studies [4, 5]. Logically, multiple deaths in a family could be interpreted as conferring a higher risk for an

individual patient, however neither the number of deaths nor the proximity of relationship to an individual has been explored in studies to date.

Exercise blood pressure response

A normal blood pressure (BP) response to exercise is defined an increase in the systolic BP (≥20mmHg) during exercise with a subsequent fall to baseline in recovery[4]. An abnormal blood pressure response to exercise (ABPRE) has been described in up to one third of HCM patients consisting of either hypotension (fall in systolic BP>20mmHg) or failure to increase systolic BP (≥ 20mmHg) [129-131]. An ABPRE has been reported to be associated with increased cardiovascular mortality and SCD risk in younger patients (<50 years) on univariable analysis [130, 131] but this has not been shown in multivariable analyses [52, 111, 124]. Conversely, a normal BP response has been shown to have a high negative predictive power for low-risk patients (97%)[130]. The significance of BP response to exercise in older patients has not been assessed.

Age

Young age has been reported to be associated with a higher risk of SCD [82] [132] but this finding has not been replicated in all studies ([53, 116, 126]. However, individual risk factors including syncope and NSVT appear to be particularly important in young patients [111, 122].

Additional proposed risk factors not currently accepted as major clinical risk factors are described in Table 3 below

Clinical risk factor	Comment
Late gadolinium enhancement (LGE) on CMRI	LGE can identify areas of myocardial fibrosis and has been shown to be associated with other clinical risk factors for SCD. It is unclear if either the presence or amount of LGE is an independent predictor for SCD [133-137]
Genetic testing	There are conflicting reports regarding the prognostic significance of genetic testing in the literature. Proposed risk factors for SCD include the presence of any sarcomeric variant[138, 139], specific genotype-phenotype correlations[140-144] and multiple disease causing variants[145, 146]. There is currently insufficient evidence to support the use of genotype in risk stratification.
LV apical aneurysm	A subset of HCM patients at high risk of arrhythmic (particularly monomorphic VT) and thromboembolic events[101].
End-stage disease (impaired systolic function)	A subset of HCM patients at high risk of SCD[147-149].
12-lead ECG	Abnormalities on the 12 lead ECG are common and present in over 90% of patients[150]. There are conflicting reports on the role of ECG features such as patterns of repolarisation[150, 151] [152, 153], QT dispersion[154, 155], and measures of LVH[150, 156] in risk stratification.

Table 3 Additional clinical risk factors for sudden cardiac death in adult populations

Clinical risk factors not currently included in risk stratification guidelines that may act as risk modifiers.

CMRI = cardiac magnetic resonance imaging, LGE = late gadolinium enhancement, SCD = sudden cardiac death, LV = left ventricular, VT = ventricular tachycardia, ECG = electrocardiogram

1.2.4 Risk factors for SCD in childhood HCM

Whilst risk factors for SCD have been extensively investigated and described in adult disease, the role of conventional risk factors have not been systematically evaluated in childhood. Moreover, suggested novel risk factors for SCD in childhood disease have not been evaluated in adequately powered studies. Four clinical variables are currently categorised as major risk factors as described below.

Previous malignant arrhythmia (VF/VT)

In agreement with adult studies, children who have survived VF/VT are at high risk of further malignant arrhythmias [41, 157]. One study has reported a hazard ratio of 5.45 (95% CI 3.67-8.10) on univariable analysis[41]. This sub-group of patients are recommended for secondary prevention ICD implantation [4, 5].

Extreme left ventricular hypertrophy

Left ventricular hypertrophy has been reported to be associated with SCD in childhood disease[56, 57] and extreme left ventricular hypertrophy (MLVWT ≥ 30mm/ Z-score ≥ 6) is considered a major risk factor [4, 5, 158]. Measures of hypertrophy in published studies vary and include interventricular septal (IVS) thickness, LV posterior wall thickness (LVPWT), LV wall thickness: cavity ratio, body-surface area corrected (Z-score) maximal wall thickness, and extreme LVH. The most appropriate measure of hypertrophy to predict risk is unknown.

Non-sustained ventricular tachycardia (NSVT)

The true prevalence of NSVT in childhood HCM is unknown with estimates from small retrospective cohorts of between 15-30% [41, 59, 158, 159]. NSVT has been

described as a risk factor for SCD in some published studies [41, 59], yet others have failed to find a significant association [55, 158, 159]. Its role in risk stratification and the magnitude of its effect is therefore unknown. No study has assessed the importance of frequency, rate or length of NSVT detected on ambulatory ECG in childhood. The significance of exercise induced arrhythmias during childhood is also unknown.

Unexplained syncope

Unexplained syncope is considered a major risk factor for SCD in current guidelines. A significant association with SCD has been reported in some published studies [159, 160], however the temporal association between events has not been explored.

Family history of SCD

A family history of SCD was found to be significantly associated with SCD in a mixed adult/paediatric cohort[125], however these findings have not been replicated in other published studies[57, 59]. Its use as a risk factor in childhood disease is largely extrapolated from adult studies.

Additional proposed risk factors for SCD in childhood disease not currently included in the guidelines are described in Table 4 below.

Clinical risk factor	Comment
Age	SCD risk has been reported to be increased in pre-adolescent years (9-14yrs)[43]. Children presenting in infancy are believed to be at lower risk of SCD [15, 81]
12 lead ECG	There have been few paediatric studies with conflicting results. Proposed 12 lead ECG features include; measures of LV hypertrophy[94] and abnormal repolarisation[161]. An ECG risk score has been developed by Ostman-Smith et al[161] but this has not been independently validated.
Heart rate variability	Heart rate variability has been linked to SCD in one study[162]
Myocardial bridging	Myocardial bridging on coronary angiogram was a risk factor for SCD in one small study[163]. This invasive investigation is not part of routine care.
LGE on CMRI	LGE has been shown to increase during childhood and is associated with left ventricular hypertrophy[46]. It is unclear if LGE is an independent risk factor for SCD[164, 165].
Inducible VT on electrophysiology study (EPS)	Inducible VT has been reported to be associated with SCD[159], however its invasive nature limits its use in risk stratification.
Restrictive physiology	Restrictive physiology, as assessed by TTE, was associated with a 3.8-fold increase in the risk of SCD in one study[166]
Genetic testing	The majority of studies investigating the role of genotype and risk exclude paediatric patients. Of those that include younger patients, survival time is often calculated from time of transition to adult care with an inherent survival bias. Single case reports have described severe disease associated with compound heterozygote/homozygote sarcomeric variants[167, 168]. The role of genotype in SCD risk during childhood is unknown.
Gender	Higher mortality rates reported in female patients in one study [40]
Symptoms	Symptoms of heart failure have been associated with heart-failure related deaths [41, 169]. The contribution of symptoms, including chest pain and palpitations, to SCD is unclear ([43, 59, 166]
ABPRE	ABPRE (as defined in adults) has not been shown to be associated with adverse outcome during childhood [41, 57, 157].
24 hour BP monitoring	Morning abnormal blood pressure ratio was associated with SCD in one small study[170].
LVOT obstruction	The definition of LVOT obstruction varies in the literature. Increasing LVOT gradient has been linked to SCD [41] but the relationship is not clearly described.
Left atrial size	Left atrial size is not included as a major risk factor but a significant association has been reported in two studies [41, 166].

Aetiology	With the exception of individual specific metabolic aetiologies (eg Danon disease [48]), patients with syndromic disease are considered to be at lower risk of arrhythmias[16, 31]. As these patients are often excluded from population studies, their SCD risk and clinical risk factors are largely unknown.

Table 4 Proposed additional risk factors for sudden cardiac death in childhood population

SCD=sudden cardiac death, ECG = electrocardiograph, LV = left ventricular, LGE = late gadolinium enhancement, CMRI = cardiac magnetic resonance imaging, VT = ventricular tachycardia, EPS = electrophysiology study, TTE = transthoracic echocardiogram, ABPRE= abnormal blood pressure response to exercise, BP = blood pressure, LVOT = left ventricular outflow tract

1.2.5 Preventing SCD in childhood HCM

Exercise avoidance

In supervised settings, exercise induced arrhythmias are thought to be rare[124], yet early reports identifying HCM as the most common cause of SCD in young athletes has led many clinicians to recommend exercise limitation. This view has been challenged more recently by a meta-analysis that reported the most common finding at autopsy in young victims of SCD is actually a structurally normal heart (27%). HCM accounted for only 10% of cases[171]. Simultaneously, recent population studies have found that the majority of sudden deaths in HCM or healthy young people occur at rest or during normal activities rather than whilst exercising [172, 173]. However, sinus tachycardia has been implicated as a possible triggering event for malignant arrhythmias in HCM patients with ICDs [102, 103] providing a possible mechanism for exercise induced arrhythmias. The question of whether it is safe for young people with an ICD to train at elite level has been investigated in a multi-centre registry of young competitive athletes (23% HCM) [174]. Of 18 (14%) individuals who received at least one appropriate therapy, 4 occurred during exercise at a rate of 1.5 appropriate shocks during sport/100 pt years. Therefore, in this small heterogeneous cohort of patients, ventricular arrhythmias occurred at a similar rate in athletes compared to unselected paediatric HCM populations. Two trials in adult HCM patients have shown that moderate exercise can improve exercise capacity with no adverse malignant arrhythmic events although the studies were not powered to assess safety of exercise[175, 176]. High quality evidence of the safety of high intensity exercise in HCM is currently lacking. Current guidelines, based largely on expert opinion, recommend that patients with HCM should be discouraged from

participating in intense physical activity particularly if they have risk factors for SCD[4, 5].

Pharmacotherapy

There are no randomised controlled trials or high quality studies to support the use of anti-arrhythmic medication as preventative therapy for SCD in HCM. Although early reports suggested that amiodarone[177] could reduce the risk of malignant arrhythmias, subsequent studies have failed to show a benefit[178]. Amiodarone has also been shown to increase the defibrillation threshold, which could result in harm[179]. One group has advocated the use of beta-blockers during childhood, reporting a reduction in the risk of SCD with high dose therapy (up to 23mg/kg/day) [63]. The results of this study have not been independently confirmed in adult or paediatric populations. Disopyramide has not been shown to have a significant effect on SCD risk[180].

Implantable Cardioverter Defibrillators (ICDs)

Arguably the greatest therapeutic advance in HCM to date has been the introduction of the ICD over the last 20 years. ICDs were first shown to be effective at treating malignant ventricular arrhythmias in adult cohorts. Contemporary estimates of the annual rate of appropriate shocks for primary or secondary prevention devices are between 2-5% and 4-11% respectively [99, 100, 181]. Despite advances in technology, device related complications and inappropriate shocks continue to be responsible for significant morbidity in this population, highlighting the need to appropriately select patients who are likely to benefit from device implantation. However, population studies have suggested that the introduction of the ICD has

changed the natural history of the disease with heart failure, rather than SCD, becoming the leading cause of mortality in adults [100].

ICDs have also been shown to be effective at terminating malignant ventricular arrhythmias in paediatric HCM patients [158, 182] but at the expense of a higher rate of complications and inappropriate therapies. Retrospective population studies have reported that, over a relatively short follow up time (mean 5-7 years), ICD related complications occur in up to 30% of childhood cohorts whilst inappropriate therapies are seen in 17-32%[157, 158, 182, 183]. As this younger group of patients will have ongoing lifetime exposure to these risks the balance between benefit and harm is particularly important.

1.2.6 Risk stratification for SCD in HCM

Whilst ICDs have been shown to be effective at terminating malignant ventricular arrhythmias[99, 100, 182], device-related complications necessitate that clinicians select patients most likely to benefit from device implantation. Little controversy exists for patients who have experienced previous VT/VF who are widely accepted to be at high risk of further ventricular arrhythmias. This group of patients are recommended for implantation of a secondary prevention device in all paediatric and adult guidelines providing life expectancy is longer than 1 year [4, 5]. However, selection of patients for implantation of a primary prevention device remains challenging and is subject to ongoing controversy.

Adult HCM

The first risk stratification guideline for adult disease was jointly published by the American College of Cardiology (ACC) and the European Society of Cardiology (ESC) in 2003. This recommended the use of 5 conventional clinical risk factors

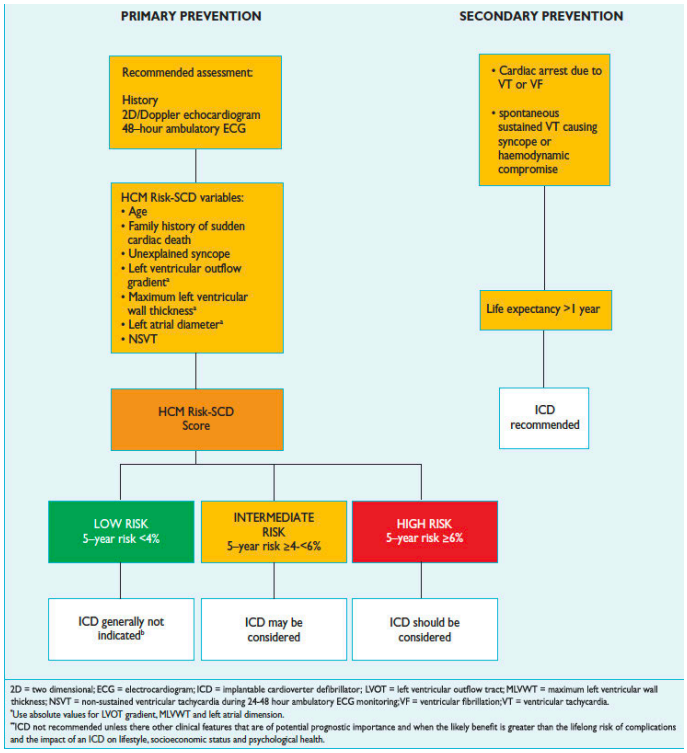
(NSVT, unexplained syncope, ABPRE, family history SCD and extreme LVH) to assess risk. Reflecting the finding that coexistence of multiple risk factors was associated with an increased risk of SCD [109, 125], ICD implantation was recommended if 2 or more risk factors were present. Following the publication of this joint guideline a divergence of the approach to risk stratification in North America and Europe was seen that continues today.

From 2006 onwards, North American guidelines have recommended that ICD implantation is reasonable in the presence of a single risk factor. This is largely based on the observation that there was a poor correlation between number of risk factors and ICD therapies in a cohort of patients *a priori* identified as high-risk by clinicians [99]. In its most recent iteration, North American guidelines (2011 ACCF/AHA)[5] recommend that a single risk factor is sufficient for ICD implantation in those with extreme LVH, unexplained syncope or a family history of SCD. Whereas an ICD should be considered in patients with NSVT and ABPRE if additional risk factors or modifiers are present (Figure 2 below).

In contrast, until 2014, the ESC continued to recommend ICD implantation in the presence of 2 or more risk factors. Although they differed in specifics, the approach to risk stratification in both guidelines provided relative, rather than absolute, estimates of risk between non-homogenous groups and did not account for differing weighting of each risk factor. They also necessitate the conversion of MWT into a binary variable for the purpose of risk stratification creating an artificial step in risk prediction. The coding of MWT as a binary predictor may not accurately account for the relationship between MWT as discussed in section 2.3 above. A large observational cohort study published in 2013 reported that whilst annual SCD incidence increased with the incremental number of risk factors, the threshold of ≥1

or ≥2 risk factor had modest discriminatory power to identify those at high risk (5-year C- statistic 0.63 and 0.64 respectively). This was at the expense of ICD implantation in a significant number of patients who did not experience an event (PPV 23.3%)[184].

In response to this finding in 2014 the first validated risk prediction model for SCD in HCM was developed in a large, multi-centre retrospective cohort using 6 pre-selected variables reported to be associated with SCD in published multi-variable analyses (age, MLVWT, LA diameter, Maximal LVOT gradient, Unexplained syncope, Family history SCD and NSVT)[82]. The HCM Risk-SCD model enabled clinicians for the first time to calculate individualised estimates of 5-year SCD risk to guide ICD implantation decisions. External validation studies have confirmed that this model has superior discriminatory ability (C-index 0.69) compared to the 2011 ACCF/AHA guidelines [185-187] and it has been endorsed by and incorporated into the ESC guidelines. In its most recent iteration the ESC guidelines categorise patients into 3 groups depending on their estimated 5-year risk; Low risk <4%, Intermediate risk 4-6% and High risk ≥ 6%. (Figure 2 below). This model has been criticised for incorrectly classifying some patients who experience events as low risk [188] and has not been adopted by North American guidelines.



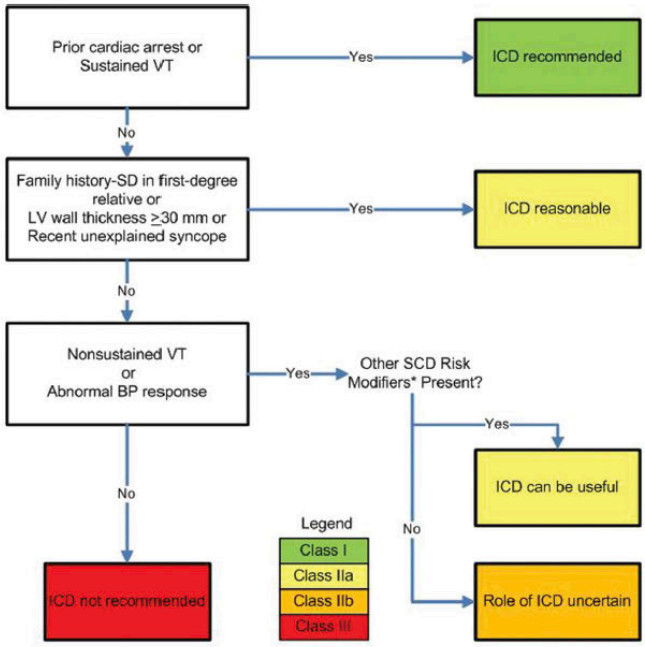


Figure 2 Risk stratification guidelines for preventing sudden cardiac death in adult patients

A) European Society of Cardiology (2014) and (B) American College of Cardiology Foundation/American Heart Association (2011)

Paediatric HCM

Not surprisingly given the sparsity of evidence for clinical risk factors in childhood disease, the current European and North American guidelines each contain only short sections on risk stratification for primary prevention in childhood. Both recommend the use of four major risk factors extrapolated from adult studies to assess risk; extreme LV hypertrophy (MLVWT ≥30mm or Z score ≥ 6), unexplained syncope, NSVT and family history of SCD. The guidelines reflect the assumption that a summative effect on risk exists in the presence of multiple risk factors. However, the threshold for recommending ICD implantation differs. An ICD implantation is recommended in those with ≥1 risk factor in the ACCF/AHA 2011 guidelines or ≥2 risk factors in the ESC 2014 guidelines (Figure 3 below). This approach to risk stratification has not been validated in childhood disease although it is widely used. The HCM Risk-SCD model endorsed by ESC adult guidelines is not validated for use in patients under the age of 16 years as they were excluded from model development. Therefore, although younger patients appear to be at higher risk of both malignant arrhythmias and ICD related complications, the evidence supporting current risk stratification approaches is sparse.

Recommendations	Class	Level
ICD implantation is recommended in children who have survived a cardiac arrest or experienced documented sustained ventricular tachycardia.	I	B
ICD implantation should be considered in children with two or more major paediatric risk factors* after appropriate counselling and when an assessment of the lifelong risk of complications and the impact of an ICD on lifestyle and psychological health suggests a net benefit from ICD therapy.	IIa	C
ICD implantation may be considered in children with a single major paediatric risk factor* after appropriate counselling and when an assessment of the lifelong risk of complications and the impact of an ICD on lifestyle and psychological health suggests a net benefit from ICD therapy.	IIb	C

It is reasonable to recommend an ICD for high-risk children with HCM, based on unexplained syncope, massive LV hypertrophy, or family history of SCD, after taking into account the relatively high complication rate of long-term ICD implantation. ***(Level of Evidence: C)***

Figure 3 Risk stratification guidelines for preventing sudden cardiac death in childhood patients

A) European Society of Cardiology (2014) and (B) American College of Cardiology Foundation/American Heart Association (2011)

Summary

In summary, childhood HCM is a rare heterogeneous disease with variable long-term outcomes. The most common cause of death is SCD, which appears to occur at a higher rate than seen in adult patients. Despite significant advances in risk stratification for adults with HCM, our current understanding of the risk factors for SCD in childhood disease is limited and current approaches to risk stratification have not been validated This has important implications for patient care.

Chapter 2: Methods

2.1 Study population: International Paediatric Hypertrophic Cardiomyopathy Consortium

In 2016, we formed the International Paediatric Hypertrophic Cardiomyopathy consortium with the purpose of systematically describing the natural history, outcomes and risk factors for SCD in childhood disease. The consortium was initially made up of thirty-nine centres worldwide with an additional ten centres recruited in 2019 for an external validation cohort (Figure 4 below). A list of contributing centres can be found in the Appendix. I was responsible for facilitating the study set-up at each collaborating centre, communicating all study information and coordinating data collection. Consecutively evaluated patients from participating centres were eligible for inclusion if they first met diagnostic criteria for HCM between 1-16 years of age. HCM was defined as the presence of a maximal left ventricular wall thickness greater than 2 standard deviations (z-score) above the population mean in the absence of abnormal loading conditions[4]. Patients with known phenocopies of sarcomeric disease, including inborn errors of metabolism, RASopathies, or neuromuscular disease, were not eligible for inclusion as they are recognised to be a heterogeneous group with distinct arrhythmic risk. Patients diagnosed in infancy (under 1 year) were also excluded as they are recognised to have a worse prognosis with death primarily secondary to heart failure. Finally, patients identified through screening as genotype positive but phenotype negative were also excluded as arrhythmic events are not expected to occur in this group of patients[189].
Patients meeting inclusion criteria were identified separately using multiple sources at each collaborating centre by the local lead investigator. The method of patient

identification differed by site and included; clinic lists, searches of pre-existing systematic research or clinical databases and searches of the electronic patient record (EPR). For UK sites (n=13), I travelled to each centre individually to identify patients, and prevent patient duplication if seen in more than one UK centre, in conjunction with the local lead investigator.

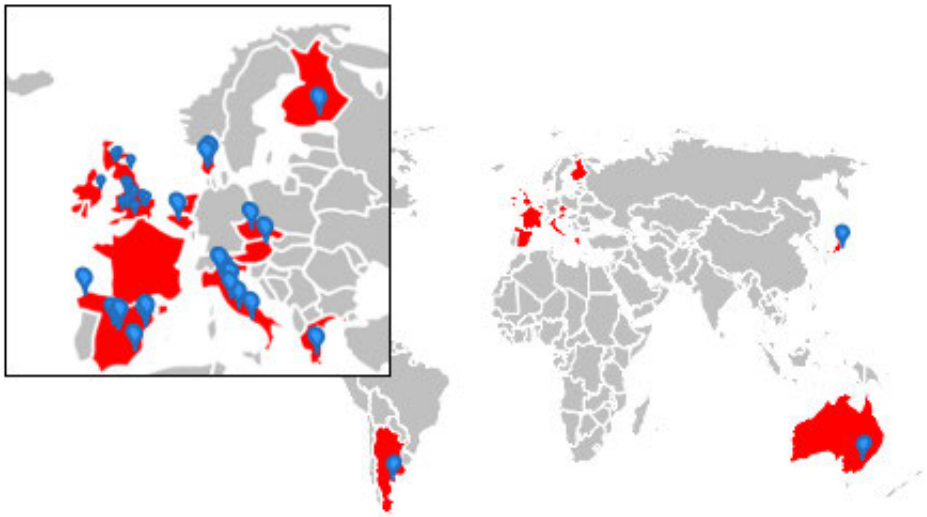


Figure 4 Paediatric Hypertrophic Cardiomyopathy Consortium: Location of participating centres in development cohort

2.2 Patient assessment and data collection

Anonymised non-invasive clinical data from baseline and follow up were collected retrospectively, including; demographics, aetiology, symptoms, pedigree analysis, date and results of genetic testing, medical therapy, resting and ambulatory 12-lead electrocardiograph, exercise testing, and 2-dimensional Doppler and colour transthoracic echocardiogram. Aetiology was classified as non-syndromic in the absence of a diagnosis of a RASopathy syndrome (Noonan or other malformation syndrome), neuromuscular disease (including Friedreich's ataxia) or inborn error of

metabolism. All patients had planned clinical reviews every 6-18 months as determined by the local clinician. Data was collected independently at each participating site by the lead investigator with the exception of sites in the United Kingdom (UK). For UK sites (n=13), I travelled to each centre individually to identify and collect data in conjunction with the local lead investigator. Data was entered into a RedCap research database which I designed for this study.

Heart failure symptoms were assessed using the Ross criteria (for symptom assessment below 5 years of age) or the New York Heart Association (NYHA) functional classification (for those over 5 years of age)[190]. Patients were defined as having heart failure symptoms if they had a score of 2 or more. Unexplained syncope was defined as a transient loss of consciousness with no identifiable cause at or prior to first evaluation[4]. A family history of sudden cardiac death (SCD) was defined as a history of SCD in 1 or more first degree relative under 40 years of age or SCD in a first degree relative with confirmed HCM at any age[4].

2.3 Clinical investigations and outcomes

Echocardiographic data collection:

Echocardiographic measurements were made according to current guidelines[4, 5]. Specifically, maximal left ventricular wall thickness (MLVWT) in millimetres is the greatest thickness measured in the parasternal long or short axis views (2D or M-Mode) at end-diastole. For short-axis views, wall thickness is measured in 4 places at the level of the mitral valve and papillary muscles (anterior, posterior septum, lateral and posterior wall) and in 2 places at the apex (anterior and posterior septum). According to current guidelines, extreme left ventricular hypertrophy was defined as a MLVWT ≥ 30mm or Z-score ≥6. Left atrial diameter in millimetres was

measured in the parasternal long axis view (2D or M-Mode) at end-systole. Peak instantaneous left ventricular outflow tract gradient was calculated at rest or with Valsalva manoeuvres using peak Doppler velocity with application of the Bernoulli equation (gradient = $4V^2$, where V is the peak aortic outflow velocity). Left ventricular tract outflow (LVOT) obstruction was defined as an instantaneous peak Doppler LVOT pressure gradient ≥30mmHg. A haemodynamically significant gradient was considered to be an instantaneous peak Doppler gradient ≥50mmHg[51]. LVOT obstruction was defined as mild (30-50mmHg), moderate (50-90mmHg) or severe (≥90mmHgs). Impaired left ventricular systolic function was defined as a fractional shortening (FS) ≤28% or ejection fraction ≤55%[191].

Electrocardiographic data collection:

Age-specific normal values for ECG parameters were used[192]. The following parameters were measured (average of 3 beats) from lead II, or V5 if quality of trace was poor: P wave amplitude (mV) and duration (ms), PR interval (ms), QRS axis, QRS duration (ms), QRS amplitude (mV), limb-lead QRS amplitude sum (mV), 12-lead amplitude-duration product[156] (mV/s), QT interval (ms), corrected QT interval (ms) (Bazett's formula), Sokolow–Lyon score (SV1 or SV2 + RV5 or RV6 ≥35mm)[193]. The presence of the following parameters were described: left or right atrial enlargement, dominant S wave in V4, pathological Q waves, pathological T wave inversion (>1mm beyond V1 aged ≥14 years, or beyond V3 aged <14 years), giant negative T waves (≥10mm), giant positive T waves (≥10mm), ST segment depression (≥2mm in any lead), ST segment elevation (≥2mm in leads V1-V3, or ≥1mm in all other leads), left bundle branch block (LBBB) or right bundle branch block (RBBB) and three specific ECG patterns ("pseudo-necrosis", "pseudo-ST elevation myocardial infarction [pseudo-STEMI]" and "low voltages")[150]. The ECG

risk score, based on 8 parameters (deviation in QRS axis, pathological T wave inversion in limb or precordial leads, ST-segment depression, dominant S wave in V4, limb-lead amplitude sum, 12-lead amplitude duration product and QTc), was calculated as described by Ostman-Smith et al[156].

Non-sustained ventricular tachycardia (NSVT) was defined as three or more consecutive ventricular beats at a rate of greater than 120 beats/min with a duration of less than 30 seconds on ambulatory ECG monitoring[4].

Exercise testing data collection:

An abnormal blood pressure response to exercise was defined as the failure of systolic blood pressure (BP) to rise by ≥20mmHg (flat BP response) or a fall in BP during exercise (hypotensive BP response)[4, 130].

Genetic data collection:

Genetic testing was performed at the treating clinicians' discretion as part of usual clinical care. I collected data for all individuals in whom genetic testing had been performed including: date of testing, genetic testing strategy (predictive or panel testing) and variants identified. Genetic testing data was not available for 6 centres. Before 2011, targeted testing of HCM genes (4-10 genes) was performed by direct Sanger sequencing. Next generation sequencing (NGS) were available from 2011 onwards. For the purpose of analysis, NGS panels were described as small (≤21 genes) or expanded (≥21 genes). The genes included in panels varied depending on the year and clinical laboratory conducting the testing. The pathogenicity of all reported variants was re-classified through a collaboration with Dr Lorenzo Monserrat (Health in Code) according to the American College of Medical Genetic Classification (ACMG)[194]. Variants were described as pathogenic (P), likely

pathogenic (LP), variant of unknown significance (VUS), likely benign or benign. Genotyped individuals were described as SARC+ (at least 1 disease causing P/LP variant in sarcomere genes present), SARC VUS (sarcomere VUS present) or SARC – (no disease-causing P/LP or VUS variant in sarcomere gene present)

Clinical outcomes

The primary study end point was a composite outcome of SCD or an equivalent event (aborted cardiac arrest, appropriate ICD therapy, or sustained VT associated with haemodynamic compromise)[82]. SCD was defined as a witnessed sudden death with or without documented cardiac failure, death within 1 hour of new symptoms, or nocturnal death with no antecedent history of worsening symptoms[4]. Cardiac rhythm at the time of ICD therapy was adjudicated by each implanting centre. Therapies were considered appropriate when triggered by a ventricular tachyarrhythmia (VF or sustained VT) and inappropriate in the setting of expert-adjudicated sinus tachycardia, supraventricular tachycardia or device/lead malfunction. Secondary outcomes included all-cause mortality, cardiac transplantation, ICD or pacemaker implantation and surgical left ventricular myectomy. Patients with no clinical review within 3 years of the study end-date were classified as lost to follow up. Outcomes were ascertained by the treating cardiologist at each participating centre.

2.4 General statistical methods

Normally distributed continuous variables are described as mean +/- standard deviation with two or three group comparisons conducted using Student *t* test and ANOVA, respectively. Skewed data are described as median (interquartile range) with two or three group comparisons performed using Wilcoxon rank sum and Kruskal Wallis tests, respectively. The distributions of categorical variables were compared using the Chi-Square test or Fisher's exact test. Two sample proportion tests were performed to compare groups within variables of interest. A significance level of 0.05 was used for all comparisons.

Estimates of survival were obtained using the Kaplan-Meier product limit method. Group differences in survival were assessed using the log rank test. Statistical analysis was performed using StataCorp. 2015. Stata Statistical Software: Release 14. College Station, TX: StataCorp LP or R software (version 3.6.2)

Z scores

MLVWT and LA diameter measurements are expressed in millimetres and as z-scores relative to the population body-surface area corrected mean. Body surface area was calculated from weight[195]. I chose the largest published reference populations for both measurements following a review of the literature (Table 5 below.) Published z scores for LVWT differ in terms of the anatomical location (eg IVST or LVPWT), echocardiographic window (eg parasternal short axis or parasternal long axis], and echocardiographic mode (eg 2D or M-Mode measurement). The pattern of hypertrophy in HCM is highly variable; and includes eccentric or apical disease, which may not be detected in classical echocardiographic views. The application of traditional Z scores in HCM therefore

has inherent limitations. As it was important to correct absolute measurements for body size in childhood I pragmatically chose to use published z-score for IVST for all LVMWT measurements.

The equations used to calculate Z-scores are described below

LA diameter (Neilan et al[196])

For male patients:

$$\frac{\left(\frac{LA\ diameter\ (mm)}{10.665} \times Bodyweight\ (Kg)^{0.225}\right) - 1}{0.118}$$

For female patients:

$$\frac{\left(\frac{LA\ diameter\ (mm)}{10.74} \times Bodyweight\ (Kg)^{0.246}\right) - 1}{0.124}$$

MLVWT (Lopez et al[197])

$$\frac{\left(\frac{MWT\ (cm)}{BSA^{0.4}}\right) - 0.58}{0.09}$$

Published study	Country	Number patients	Inclusion criteria	Method of measurement
Left atrial diameter Z score				
Kampmann et al 2000[198]	Italy	2036	Age: 0-18 years BSA (Dubois): 0-2.0	AP diameter M mode PSLAX
Cantinotti et al 2014[199]	Italy	1091	Age: 0-17 years BSA (Hancock): 0.12-0.67	AP/LL diameter 2D, 4Ch
Pettersen et al 2008[200]	USA	782	Age: 0-18 years BSA (Dubois): 0-20	AP diameter 2D PSLAX
Neilan et al 2009[196]	USA	4109	Age: 1-17 years Weight (8.2-91.4)	AP diameter 2D PSLAX
Huwez et al 1994[201]	UK	127	Age: 0-19 years BSA (Dubois): 0.27-1.603	AP diameter M Mode PSLAX
Oran et al 2014[202]	Turkey	2000	Age: 0-17 years	AP diameter M Mode PSLAX
Nidorf et al 1992[203]	USA	196	Age: 0-18 years	AP diameter PSLAX
Gokhroo et al 2017[204]	India	595	Age: 4-15 years BSA (Haycock)	AP/LL diameter 2D, 4Ch
Interventricular septal thickness Z score				
Pettersen et al 2008[200]	USA	782	Age: 0-18 years BSA (Dubois): 0-20	M Mode PSAX (tip of papillary muscles)
Kampmann et al 2000[198]	Italy	2036	Age: 0-18 years BSA (Dubois): 0-2.0	M Mode PSLAX
Oran et al 2014[202]	Turkey	2000	Age: 0-17 years	M Mode PSLAX
Huwez et al 1994[201]	UK	127	Age: 0-19 years BSA (Dubois): 0.27-1.603	M Mode PSLAX
Gokhroo et al 2017[204]	India	595	Age: 4-15 years BSA (Haycock)	M Mode PSAX (tip of papillary muscles)
Lopez et al 2017[197]	USA	3215	Age: 0-18 years (Haycock + Gehan and George)	M Mode PSAX (tip of papillary muscles)

Table 5 Summary of published Z–scores for left atrial diameter and interventricular septal thickness echocardiographic measurements in childhood

LL= longest length

Ethical approval

NHS wide Health Research Authority (HRA) approval was given for collecting anonymised, non-invasive clinical data in UK with waiver of informed consent. Local ethical committee approval was obtained for all other participating sites.

Chapter 3: The clinical presentation and outcomes of childhood HCM

3.1 Introduction

Childhood Hypertrophic cardiomyopathy (HCM) is a rare disease with an estimated incidence of 0.24-0.47 per 100,000[12-14]. Understanding the spectrum of disease, symptom burden and outcomes is essential for the management of children with HCM, however our current understanding is incomplete as described in Chapter 2. Retrospective population registry studies from North America[16] and Australia[15], containing 855 and 80 patients respectively, have provided valuable insights into the epidemiology, clinical presentation, and survival of childhood HCM. These studies have described a disease with a heterogeneous underlying aetiology, which includes inborn errors of metabolism, neuromuscular disorders and malformation syndromes. However, they lacked detailed descriptions of individual underlying syndromes or diagnoses meaning our understanding of the aetiology of disease is incomplete. Additionally, in recent years there has been increased recognition of the importance of family screening following demonstration that the majority of disease, even in childhood, is secondary to pathogenic sarcomeric protein variants (6, 7). The effect of changing screening practices on both the age of presentation and aetiology in a large childhood cohort has not been assessed.

Long-term survival for the majority of children with HCM is reported to be good [81]. Aetiology and age of presentation have been shown to be at least partly responsible for differences in long-term outcomes[15, 16, 31] but disease-specific causes of mortality and outcomes other than mortality have been poorly described. Since the early descriptions of HCM there have been many changes in medical management most notably the introduction of implantable cardioverter defibrillators (ICD's), which

have been shown to be effective at terminating malignant ventricular arrhythmias[158, 182]. Family screening now also offers the opportunity for intervention at an earlier stage. It is currently unknown if these advances have translated into improved long-term outcomes for childhood disease. An improved understanding of the effect of changing medical practices on the demographics, clinical characteristics and outcomes in childhood HCM would help identify current gaps in knowledge and understanding. This work was published in the European Heart Journal in 2019[205].

3.2 Aim

The aims of this chapter were to;

1) Describe the demographics, clinical characteristics and outcomes of childhood HCM
2) Investigate the era effect and impact of changing medical practices on the childhood HCM population.

3.3 Methods

The Paediatric Hypertrophic Cardiomyopathy Consortium includes 1029 patients with childhood HCM as described in Chapter 2 Methods. I performed a detailed description of the aetiology, clinical characteristics and outcomes of childhood disease with respect to era using a subset of this population from the United Kingdom. The rationale for using this smaller cohort is that I am able to verify that it is well-characterised as the data was collected by myself during visits to 13 out of 14 UK paediatric cardiac centres. The UK cohort additionally includes patients with infant-onset and syndromic disease which were excluded from the original international paediatric hypertrophic cardiomyopathy consortium.

For this sub-study patients were eligible for inclusion if they were diagnosed with HCM in the UK under the age of 16 years between 1980 and 2017. Eligible patients were identified by the principle investigator at each site using multiple sources, including medical databases and echocardiography log books.

Details of data collection and clinical investigations are described in Chapter 2.

The primary patient outcomes, taken from last clinic appointment, were SCD or an equivalent event (as defined in Chapter 2); heart-failure related death; cardiac transplantation; or non-cardiac death. Patients were classified as lost to follow up if last clinical review was more than 3 years ago. In order to investigate possible era effects, patients were grouped according to the decade in which they were first diagnosed (1980-1989; 1990-1999; 2000-2009; 2010-2017).

Statistical analysis:

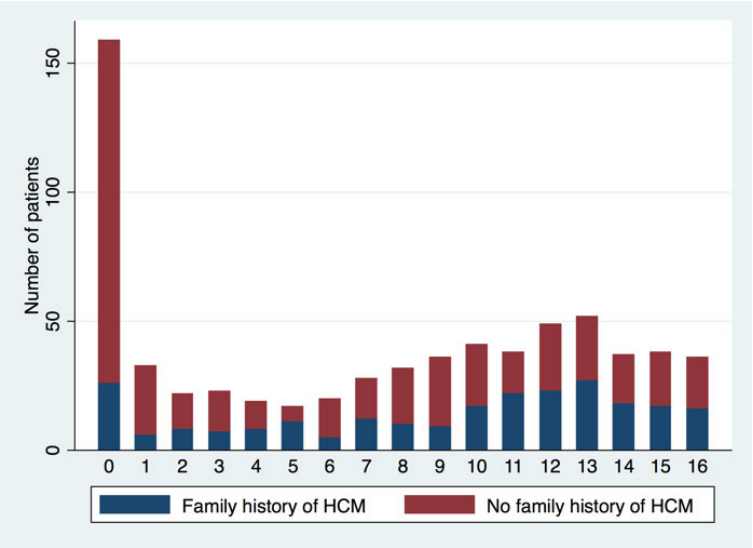
General statistical measures are described in chapter 2.

3.4 Results

I identified 687 patients diagnosed with HCM aged 16 years or younger in the UK. Median age at presentation was 5.2 years (IQR 2.3-9.9 years; range 0-16 years). One hundred and fifty-nine patients (23%) presented under the age of 1 year and 314 (46%) during pre-adolescent years (1- 12 years old) (Figure 5 below). 434 patients (63%) were male. Baseline clinical and demographics characteristics by age of diagnosis are described inTable 6. below.

Aetiology

Aetiology varied; 433 patients (63%) had non-syndromic HCM, 126 patients (18.3%) a RASopathy syndrome, 64 (9.3%) an IEM, and 59 (8.6%) Friedreich's ataxia. The baseline demographics by underlying aetiology are described in Table 7 below. The underlying diagnosis for those with syndromic disease is described in Table 8 below. The number of patients with an underlying IEM or Friedreich's ataxia increased over time (Table 9 below). Patients with Friedreich's ataxia presented later in childhood (mean age of 10.4 years (range 4 - 16)), although four patients were diagnosed below the age of 7 years. (Figure 5 below).



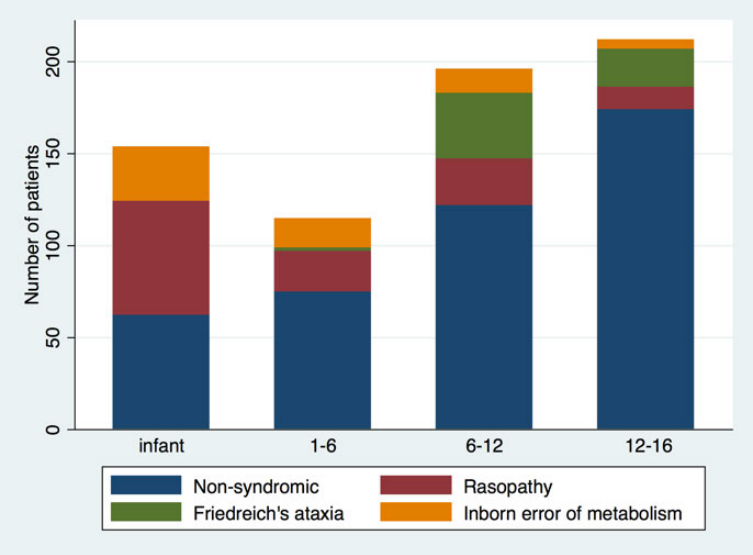


Figure 5 Bar chart showing age of presentation of childhood HCM
a) by family history b) by underlying aetiology

		Whole cohort	< 1 year	> 1 year	P value
Gender	**Male**	434 (63.2%)	102 (64.2%)	332 (62.9%)	0.770*
Aetiology	**Non-syndromic**	433 (63%)	62 (39%)	371 (70.3%)	<0.001
	RASopathy syndrome	126 (18.3%)	67 (42%)	59 (11.2%)	
	Friedreich's ataxia	59 (8.6%)		59 (11.2%)	
	Other neuromuscular syndrome	5 (0.7%)		5 (1%)	
	Inborn error of metabolism	64 (9.3%)	30 (18.9%)	34 (6.4%)	<0.001
Family history of HCM (n = 680)		242 (35.6%)	26 (16.4%)	216 (41.5%)	<0.001*
Family history of SCD (n = 682)		50 (7.3%)	3 (1.9%)	47 (9%)	0.039*
NYHA/ Ross at presentation (n = 684)	**I**	516 (75.4%)	94 (59.5%)	422 (80.2%)	<0.001
	II	133 (19.4%)	42 (26.6%)	91 (17.3%)	0.120
	III	29 (4.2%)	17 (10.8%)	12 (2.3%)	<0.001
	IV	6 (0.9%)	5 (3.2%)	1 (0.2%)	0.267
Cause of mortality (n=75)	**SCD**	20 (2.9%)	2 (1.3%)	18 (3.4%)	<0.001
	CCF	12 (1.7%)	10 (6.3%)	2 (0.4%)	
	Other-CV	12 (1.7%)	8 (5%)	4 (0.8%)	
	Non-CV	17 (2.5%)	12 (7.5%)	5 (0.9%)	
	Unknown	14 (2%)	3 (1.9%)	11 (2.1%)	

Table 6 Baseline clinical and demographic characteristics by age of diagnosis.
Data expressed as number (%). Total number of patients is 687 unless otherwise stated. All comparisons are made using a two-sample proportion test unless otherwise stated. *Comparisons were made using Chi square test.

		Non-syndromic (n=433)	Noonan syndrome (or RASopathy) (n=126)	Friedreich's ataxia (n=59)	Inborn error of metabolism (n=64)	P value
Gender	**Male**	286 (66%)	83 (66%)	28 (47%)	33 (52%)	0.008
Age at presentation (years)	**<1**	62 (14%)	67 (53%)	0	30 (47%)	<0.001
	1-5	75 (17%)	22 (17%)	2 (3%)	16 (25%)	
	6-11	122 (28%)	25 (20%)	36 (61%)	13 (20%)	
	12-16	174 (40%)	12 (10%)	21 (36%)	5 (8%)	
Family history of HCM (n = 680)		214 (50%)	8 (6%)	9 (15%)	11 (17%)	<0.001
Family history of SCD (n = 682)		45 (10.5%)	1 (1%)	2 (4%)	2 (3%)	0.001
NYHA/Ross at presentation (n = 684)	**I**	345 (80%)	77 (61%)	48 (83%)	41 (65%)	<0.001
	II	71 (16%)	41 (33%)	9 (15%)	12 (19%)	
	III	13 (3%)	7 (6%)	0	9 (14%)	
	IV	3 (1%)	1 (<1%)	1 (2%)	1 (2%)	
Myectomy		38 (8.8)	25(20%)	0	0	<0.001
Cause of mortality (n=75)	**SCD**	18 (4.2%)	2 (1.6%)	0	0	0.013
	CCF	4 (1%)	4 (3.2%)	0	4 (6.3%)	
	Other CV	7 (1.6%)	4 (3.2%)	0	1 (1.6%)	
	Non-CV	3 (0.7%)	5 (4%)	1 (1.7%)	8 (12.5%)	
	Unknown	8 (1.8%)	2 (1.6%)	0	3 (6.3%)	

Table 7 Baseline clinical and demographic characteristics by underlying diagnosis
Data expressed as number (%). Total number of patients is 687 unless otherwise stated. CCF = congestive cardiac failure, CV = cardiovascular

Family history:

242 patients (35.6%) had a first-degree relative with HCM; of these, 214 (88%) had non-syndromic HCM. The median age of presentation was higher compared to those without a family history [11 years (5-13) vs 6 years (0-12), p<0.0001] and they were less likely to present during infancy (Table 6, Figure 5 above.). 141 patients (58%) with a family history presented in pre-adolescent years (<12 years). 50 patients (7.3%) had a family history of SCD in a first-degree relative.

Symptoms at presentation:

516 patients (75.4%) were asymptomatic for heart failure symptoms at presentation (New York Heart Association (NYHA)/Ross functional class 1). Thirty-five patients (5.1%) had symptoms of congestive cardiac failure (NYHA/Ross>2). Heart failure symptoms were more common in patients presenting in infancy (Table 6) or with an underlying diagnosis of a RASopathy syndrome or IEM (Table 7). Thirty-eight patients (5.6%) had a history of unexplained syncope at presentation. Twenty-four patients (3.5%) presented following an aborted SCD or out of hospital arrest. 195 (28.4%) were started on cardiac medications at presentation; B-blockers (n=159, 23.1%), amiodarone (n=5, 0.7%), other (n=31, 4.5%).

Inborn error of metabolism (n=64)	**Storage disorders**	GSD III	9
		GSD IIb (Danon disease)	6
		GSD II (Pompe disease)	9
		Mucopolysaccharide storage disorder	12
	Mitochondrial disorders	MELAS	2
		Leigh syndrome	1
		Sengers syndrome	2
		Congenital lactic acidosis	3
		Mitochondrial cytopathy	7
		Mitochondrial respiratory chain disorders	1
	Disorders of fatty acid metabolism	Carnitine transporter deficiency	2
		VLCADD	2
	Disorder of glycosylation	Carbohydrate glycoprotein deficiency	2
	Disorder of amino acid metabolism	Glutaric acidemia type 3	1
	Other	Molybdenum cofactor deficiency	2
		Purine nucleoside phosphorylase deficiency	1
		Aicardi-goutieres syndrome	1
		Kabuki Syndrome	1
Neuromuscular disease (n=64)	**Friedreich's Ataxia**	Friedreich’s Ataxia	59
	Other neuromuscular disease (n=5)	Becker muscular dystrophy	1
		Emery Dreifuss muscular dystrophy	1
		Myofibrillar myopathy	1
		Downs syndrome	1
		Unknown	1
RASopathy (n= 126)	**Noonan Syndrome**	Noonan syndrome	110
	Other RASopathy (n=16)	Costello syndrome	5
		Noonan syndrome with multiple lentigines	10
		Cardiofaciocutaneous syndrome	1

Table 8 Specific diagnosis for patients with syndromic hypertrophic cardiomyopathy

GSD = Glycogen Storage Disorder, MELAS= Mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes, VLCADD= Very long-chain acyl-coenzyme A dehydrogenase deficiency

		1980-1989 (n=11)	1990-1999 (n=117)	2000-2009 (n=254)	2010-2017 (n=305)	P value
Gender	**Male**	6 (55%)	67 (57%)	161 (63%)	200 (66%)	+ 0.412
Syndrome	**Idiopathic/non-syndromic**	5 (45%)	88 (75%)	157 (62%)	183 (60%)	+ 0.025
	RASopathy	4 (36%)	19 (16%)	48 (19%)	55 (18%)	
	Friedreich ataxia	1 (10%)	9 (8%)	26 (10%)	23 (8%)	
	Inborn error of metabolism	1 (10%)	1 (1%)	21 (8%)	41 (13%)	
Mortality rate (per 100 person years at risk)		2.27 (95% CI 0.852 - 6.05)	1.38 (95% CI 0.890-2.14)	1.71 (95% CI 0.123-2.373)	2.54 (95% CI 1.672 - 3.858)	* 0.735
SCD rate (per 100 person years at risk)		1.13 (95% CI 0.284 - 4.537)	1.17 (95% CI 0.729-1.886)	1.14 (95% CI 0.765-1.703)	1.73 (95% CI 1.044 - 2.872)	* 0.876

Table 9 Clinical characteristics and survival rates by era of presentation.
Data expressed as number (%). Total number of patients is 687 unless otherwise stated. + Indicates comparisons were made using Chi square test. * Indicates comparison made using Log Rank Test. SCD = sudden cardiac death

Investigations at baseline:

MLVWT on 2D echocardiography at baseline ranged from 4-48mm (median 13mm, IQR 11-18mm, n=645), with median Z score +10.1 (IQR +3.6 to +34.5). Pattern of hypertrophy varied; asymmetric (n=413, 60.1%), concentric (n=154, 22.4%), biventricular (n=41, 6.0%, eccentric (n=4, 0.6%), apical (n=5, 0.7%), and unknown (n=66, 9.6%). LVOT gradient at baseline was documented in 604 (88%) patients with a median gradient 9mmHg (IQR 6,35). One hundred and sixty patients (26.5%) had LVOT obstruction and 32 patients (5.3%) had a gradient >90mmHg. Thirty-one patients (4.5%) had impaired LV systolic function. Phenotype varied by aetiology (Table 10).

Of 483 patients (71%) with a 24-hour ambulatory ECG at baseline, NSVT was detected in seven (1.4%). An exercise test was performed in 220 out of 412 patients (53%) aged 6 years and above. Blood pressure response to exercise was classified as abnormal in 84 patients (38%), of which 23 (27%) had LVOT obstruction at rest and 37 (44%) were on beta-blocker therapy. No patient had documented arrhythmias during exercise.

			Whole cohort	Non-syndromic	RASopathy	IEM	P value
Echocardiographic data at diagnosis (n=677)	**MLVWT (mm) [median (IQR)]**		13.0 (11.0-18.0)	15.0 (12.0-21.0)	11.0 (9.0-13.0)	11.0 (9.0-13.0)	0.001
	MLVWT (Z score) [median (IQR)]		10.1 (3.6 – 37)	4.2 (3.0-5.6)	4.1 (3.1-5.3)	3.4 (2.5-4.7)	0.001
	Pattern of hypertrophy	**ASH**	431 (60.1%)	343 (85.5%)	52 (47.3%)	8 (14.6%)	<0.001
		Concentric	154 (22.4%)	37 (9.2%)	34 (30.9%)	39 (70.9%)	
		BVH	41 (6.0%)	10 (2.5%)	23 (20.9%)	8 (14.6%)	
	Left atrial diameter (mm) [mean (SD)] [n = 352]		29 (9.2)	31.6 (8.5)	23.4 (10.4)	20.7 (7.3)	0.001
	Left ventricular outflow gradient at rest (mmHg) [n=604]	**< 30**	444 (64.6%)	287 (73.2%)	58 (55.2%)	49 (89.1%)	<0.001
		30-50	47 (6.8%)	27 (6.9%)	17 (16.2%)	2 (3.6%)	
		50-90	81 (11.8%)	55 (14.0%)	22 (21.0%)	4 (7.3%)	
		>90	32 (12.1%)	23 (5.9%)	8 (7.6%)	0	
	Impaired systolic function		31 (4.5%)	20 (4.6%)	1 (0.8%)	8 (12.5%)	0.016
Ambulatory ECG performed (n=483)	**Yes**	**NSVT**	7 (1.4%)				
		No NSVT	476 (98.6%)				
	No		204 (29%)				
Blood pressure response to exercise (n=220)	**Normal (>20mmHg rise in systolic BP)**		126 (57%)				
	Flat (<20mmHg rise in systolic BP)		68 (31%)				
	Hypotensive (Fall in systolic BP)		16 (7%)				

Table 10: Results of baseline investigations by underlying diagnosis

Data expressed as number (%). Total number of patients is 687 unless otherwise stated. SD = standard deviation * Exercise test in patients ≥6 years

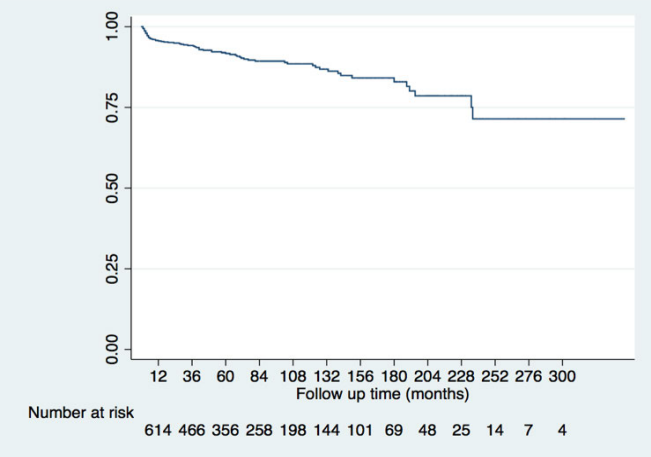
Outcomes:

Median length of follow up was 5.2 years (IQR 2.3-9.9 years). 529 (77%) patients remained under the age of 18 years at last follow up. 98 patients (14.3%) were classified as lost to follow up, of which 41 (42%) had been transitioned to adult care. 135 patients (20%) underwent ICD implantation for primary (n=108, 80%) or secondary (n=27, 10%) prevention. Sixty-three (9.2%) patients had a myectomy, which was more frequently performed in those with a RASopathy syndrome ($p<0.001$). A pacemaker was implanted for sinoatrial disease (n=6), AV node disease (n=12) or LVOT obstruction (n=8).

Mortality:

612 patients (89.1%) were alive at last clinical follow up. Seventy-five patients (10.9%) died: SCD in 20 (2.9%); non-cardiovascular (CV) in 17 (2.5%); congestive cardiac failure (CCF) in 12 (1.8%); other CV causes in 12 (1.8%); and unknown in 14 (2.3%). 16 patients (80%) who died suddenly were under 18 years of age at the time of death. Ten patients (1.5%) underwent cardiac transplantation. Survival free from death or transplantation was 95.6% (95% CI 93.7-96.9) at 12 months and 86.3% (95% CI 82.6 - 89.2) at 10 years (Figure 6). No difference in survival was seen by era of presentation (Figure 6). Children diagnosed during infancy or with an IEM had a worse prognosis, with survival at 12 months of 83.3% (95% CI 76.5 - 88.3%, $p<0.0001$) or 82.2% (95% CI 70.2 - 89.8%) respectively (Figure 6*,*Table 11*)* Cause of death differed by underlying aetiology (Table 7) and age of diagnosis (Table 6 *&* Figure 7*).*

A



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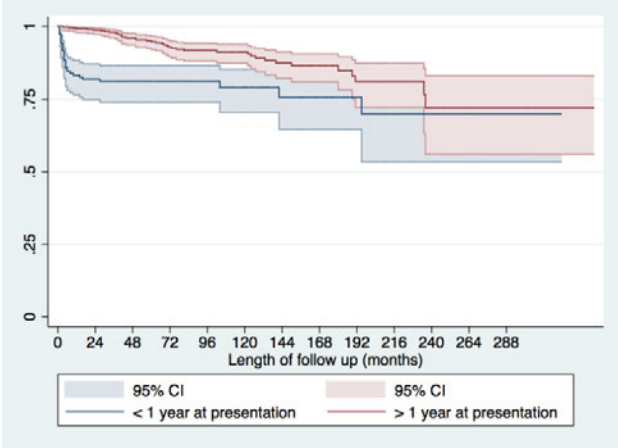

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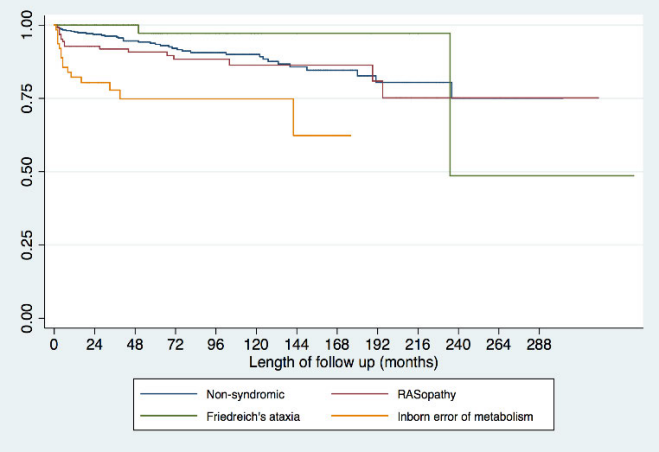

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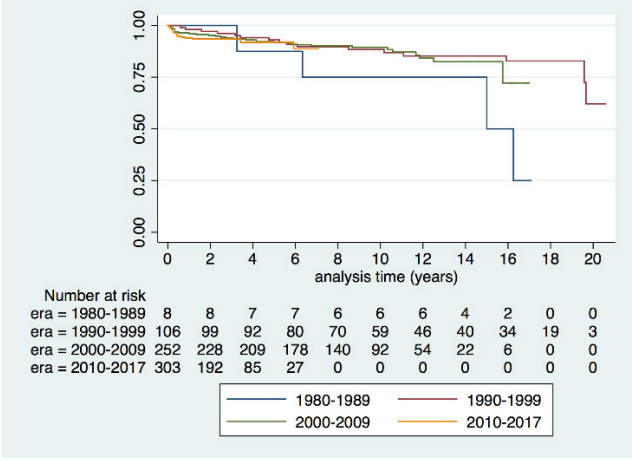


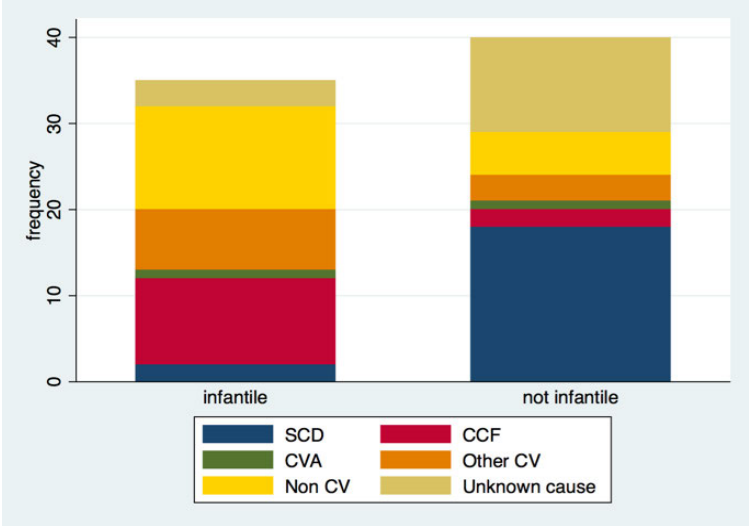
Figure 6 Kaplan-Meier curves showing survival free estimates from all-cause mortality or cardiac transplantation

a) Whole cohort; b) Stratified by age of presentation, Log rank test p<0.0001. 95% CI are shown. c) Stratified by aetiology, Log rank test <0.0001. d) Stratified by era, Log rant test P =0.735

		Survival after HCM diagnosis, % (95% confidence interval)			
		1 year	**5 year**	**10 year**	**Log rank analysis**
Whole cohort		95.6% (93.7 - 96.9)	90.6% (87.9 - 92.7)	86.3% (82.6 - 89.2)	
Age at presentation	**< 1 year (n= 159)**	83.3% (76.5 - 88.3)	80.5% (73.2 - 85.9)	78.3% (69.7 - 84.7)	<0.0001
	> 1 year (n=528)	99.2% (97.9 - 99.7)	93.6% (90.6 - 95.6)	88.6% (84.3 - 91.7)	
Aetiology	**Non-syndromic (n=433)**	97.6% (95.7 - 98.7)	92.7% (89.4 - 95)	87.5% (82.8 - 91.1)	<0.0001
	RASopathy (n=126)	92.5% (86.1 - 96)	90.5% (83.4 - 95.6)	85.9% (76.7 - 91.7)	
	Inborn error of metabolism (n=64)	82.2% (70.2 - 89.8)	66.4% (50.2 - 78.3)	66.4% (50.2 - 78.3)	
	Friedreich's ataxia (n=59)	100	97.1% (80.9 - 99.6)	97.1% (80.9 - 99.6)	

Table 11 Survival rates from time of presentation to death or heart transplant

Survival rates, using the Kaplan-Meier method, with subgroup analysis based on age at diagnosis and aetiology. Patient numbers are indicated (n=)



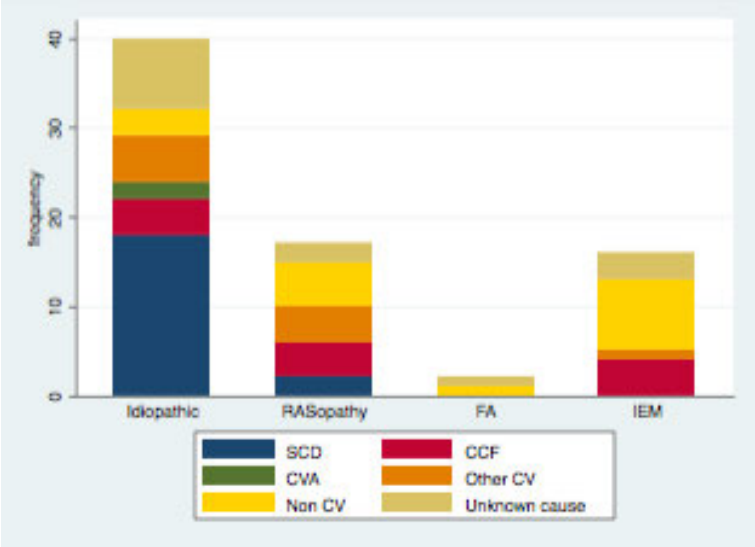


Figure 7 Stacked bar chart showing the mode of death
a) by age of presentation and b) underlying aetiology

Major arrhythmic events:

Fifty-eight patients (8.4%) had an arrhythmic event (SCD in 20 (34%); resuscitated cardiac arrest in 11 (19%); sustained VT in 8 (14%); appropriate ICD discharge in 19 (33%)) during follow up, with an event rate of 1.2 per 100 patient years at risk. Freedom from arrhythmic event was 98% (95% CI 97.3-99.3) at 1 year, 92.9% (95% CI 90.3 - 94.9) at 5 years and 89.6% (95% CI 86.1 - 92.2%) at 10 years. Two events (3.4%) occurred in infancy and 16 (27.6%) in pre-adolescence (< 13 years). Arrhythmic events occurred in 51 patients (88%) with non-syndromic HCM, 5 (9%) with a RASopathy and 2 (3%) with an IEM (Table 12). No patients with neuromuscular disease (e.g. Friedreich's ataxia) had an arrhythmic event. 9 patients (7.6%) who had been transitioned had arrhythmic events (SCD n=4, appropriate ICD discharge n=5).

	Diagnosis	Age at presentation	Symptoms at presentation	Phenotype	History of arrhythmias	Interventions	SCD or equivalent event
Patient 1	Danon's disease	13 years	Unexplained syncope	Concentric MWT 30mm LVOT 10mmHg	Nil	ICD implanted aged 14 yrs	Sustained VT aged 16yrs
Patient 2	Glycogen Storage Disease	3 months	NYHA 1	Concentric MWT 9mm LVOT 6 mmHg	Nil	Reveal device implanted aged 14 (unexplained syncope aged 13)	SCD - reveal device documented VT + fine VF aged 14 years
Patient 3	Noonan's	13 years	NYHA II	ASH MWT 18mm LVOT 58mmHg	NSVT aged 16 years	Nil	SCD aged 18 years
Patient 4	Noonan's	11 years	NYHA 1	Concentric MWT 18mm LVOT 23mmHg	NSVT aged 15 years	Primary prevention ICD implanted aged 16 years	Appropriate ICD therapy aged 16 years
Patient 5	Noonan's	9 days	NYHA II	BVH MWT 14mm LVOT 32mmHg	Nil	Myectomy aged 5 years	SCD aged 8 years
Patient 6	Other RASopathy	2 years	NYHA 1	ASH MWT 18mm LVOT 16mmHg	NSVT aged 9 years	Nil	Aborted cardiac arrest aged 10 years
Patient 7	Noonan's	3 months	NYHA III	BVH MWT 12mm LVOT 73 mmHg RVOTO	Nil	Nil	Sustained VT aged 14 months

Table 12 Clinical characteristic of patients with syndromic disease and arrhythmic events

3.5 Discussion and limitations

In this chapter I have described the clinical characteristics and outcomes of a national cohort of 687 childhood HCM patients. This cohort represents one of the largest studies in paediatric HCM, and the largest systematic study from Europe.

Aetiology and screening

Our current understanding of the aetiology and clinical characteristics of childhood HCM is primarily derived from a North American registry, which described 855 patients under the age of 18 years[16]. The cohort of patients that I describe has a comparable length of follow up [although recruited from a wider time period (1980 – 2017 vs 1990-2009)], which provides a useful comparison of the clinical presentations and outcomes of childhood HCM in different healthcare systems. In agreement with the North American study, I found that the underlying aetiology of childhood HCM was heterogeneous. The most common aetiology overall was non-syndromic but 37% had syndromic disease, which was more common in infantile disease. In contrast to previously published studies, I have reported specific diagnoses highlighting that the 'syndromic HCM' group includes patients with a wide variety of underlying diagnoses, each with its own characteristic disease phenotype and natural history.

An increasing number of HCM patients were identified over time with a higher proportion of patients with inborn errors of metabolism included in the most recent era (2010 onwards, n=41) compared to earlier time periods (1990-1999, n=1). This is likely to be due to systematic cardiac screening of patients with metabolic disease as well as more widespread family screening for non-syndromic disease.

Current guidelines recommend that routine family clinical screening for non-syndromic disease is not started before the age of 10 [4] or 12 years [5] as conventional consensus opinion is that the development of a phenotype in sarcomeric disease rarely occurs before adolescence. This is largely based upon a single historical study in which progression of hypertrophy was seen during adolescent years[23]. In my analysis, almost half (n=214, 49%) of patients with non-syndromic disease had a family history of HCM, over 50% of which were diagnosed before adolescence. I am not able to determine if these patients were diagnosed through screening or in response to symptoms, but it is noteworthy that the majority were asymptomatic at presentation. Patients with a family history of HCM were older at diagnosis compared to those without a family history. The age of presentation for familial disease is determined both by disease specific and healthcare system factors, such as the age at which family screening started. Patients with more severe disease may therefore present at a younger age with symptoms whereas screening may identify children with milder subclinical disease. The PCMR registry reported a much lower proportion of familial disease (n=109, 17%) and did not describe the age of presentation[16]. However, the results from my analysis suggest that the age-related penetrance of HCM is highly variable and that consideration should be given to commencing screening at an earlier age. In a separate analysis of the clinical yield of screening for HCM in first-degree child relatives seen at a Great Ormond Street Hospital published in 2019, I showed that almost 5% of individuals (8% of families) met diagnostic criteria during childhood, the majority of which (72%) presented in pre-adolescent years [26]. These results have subsequently been replicated by other groups [27] and suggest that a paradigm shift in the approach to family screening for

HCM in childhood is required. Nonetheless, it is apparent that current screening practices are changing the demographic of childhood HCM.

Phenotype of childhood HCM

Consistent with previous reports, I found that the HCM phenotype was varied and associated with underlying aetiology and age of presentation. The majority of patients were asymptomatic at presentation, however those presenting in infancy were more likely have heart failure symptoms (40% vs 18%, p value <0.001). The pattern of hypertrophy was strongly determined by aetiology; ASH was most common in those with non-syndromic disease (86%), whilst most patients with an IEM had concentric hypertrophy (71%). Impaired LV systolic function was uncommon overall (4.5%), but seen in one-eighth of patients with an IEM. Systolic function is typically hyperdynamic in HCM and whilst evolution to a dilated-hypokinetic phase has been described to occur in a minority (≈5%) of adult HCM patients[74], this is rarely reported in childhood. My findings therefore emphasise that the finding of impaired systolic function should prompt clinicians to look for an underlying syndromic aetiology. One third of patients had obstructive disease, which is comparable to adult cohorts[52]. However, 1 in 2 patients with a RASopathy syndrome had a LVOT gradient above 30mmHg, confirming previous reports that this group of patients have a higher prevalence of obstructive disease[29]. Non-sustained VT is detected in 20-30% of adult HCM patients [111, 113] but was observed in only 1.4% (n=7) of this cohort. This could suggest that NSVT occurs less frequently in childhood disease although importantly one third of the cohort did not have an ambulatory ECG within 6 months of diagnosis. Interestingly, a significant proportion of patients had an abnormal BP response to exercise (38%). The clinical

significance of this observation is not known as the definition of ABPRE used in adult practice[4] has not been validated in childhood.

Survival

My analysis confirmed that most children with HCM have a relatively good prognosis, with overall mortality or transplantation rates of 1.5 per 100 patient years at risk. Furthermore, this data provides further proof that that underlying aetiology has a significant impact on outcomes [15, 16]. Five-year survival was much lower for patients with an IEM (66%), compared to those with a diagnosis of a RASopathy syndrome (90.5%) or Friedreich's ataxia (97%). These survival rates are higher than those previously reported in the North American registry[16], which likely reflects the variability in phenotype and outcomes that are seen. The cause of death differed depending on the underlying aetiology, non-cardiac deaths were more common in patients with an IEM (n=8, 12.5%) or RASopathy (n=5, 4%). This highlights the multi-systemic nature of such conditions where cardiac involvement is only one aspect of the clinical phenotype and may not determine long-term outcomes. Also in agreement with previous reports[15, 16], I found that presentation at a young age was associated with worse outcomes. This finding may be partly explained by age-related difference in aetiology. However, it may also be a surrogate marker for disease severity as patients with more severe disease may be expected to present earlier in life and with worse outcomes.

No era effect was seen on survival, the explanation for which is likely multifactorial. The medical management of children with HCM has not significantly changed over the study period. However, ICDs have become widely used and shown to be effective at terminating malignant arrhythmias[158, 182, 206]. In this cohort, over

time the proportion of patients experiencing SCD decreased whilst those with an aborted SCD/appropriate ICD therapy increased. As the most common cause of death outside of infancy was SCD, one might therefore have expected improved survival over time. This apparent contradiction could be explained by the higher number of infants with an inborn error of metabolism or RASopathy, who are known to have a worse overall prognosis, in recent eras.

Arrhythmic events

Arrhythmic events occurred at a rate of 1.2 per 100 patient years at risk. While this is significantly lower than initial reports [43, 79, 80], it remains higher than in adult cohorts (0.81% per year) [93, 184]. The majority of events occurred in adolescent years (≥12 years) but one third (28%) occurred in pre-adolescence, including 2 during infancy. This highlights the importance of risk stratification for SCD at all ages. The finding that arrhythmic events occurred after transition to adult care confirms recent reports[60] that arrhythmic risk is not limited to childhood and adolescence and ongoing surveillance is required. Interestingly, 7 of the patients who died suddenly had phenocopies of sarcomeric HCM, groups of patients that are traditionally regarded as having a low risk for an arrhythmic event. One of these patients had Danon syndrome, which is recognized to have a high risk of SCD[47], however five had a diagnosis of a RASopathy syndrome and one had a glycogen storage disorder. This highlights the need for a systematic assessment of arrhythmic risk in all patients with HCM, regardless of the underlying aetiology. It is noteworthy that there were no arrhythmic events in the 59 children with Friedreich's ataxia, suggesting that patients with Friedreich's ataxia may be at lower risk of ventricular arrhythmias. Challenges remain in identifying patients at highest risk of malignant

arrhythmias as evidenced by patients experiencing SCD/aborted arrest (n=31) whilst under clinical follow up and a low number of appropriate ICD therapies (14%) in those with a primary prevention ICD.

Limitations

This analysis is limited by inherent problems of retrospective studies, in particular missing or incomplete data. As no prospective database exists in the UK, all patients diagnosed with HCM over the study period (1980-2016) may not have been included. The number of patients by calendar year of diagnosis increased over time, which may be due to more complete identification of affected patients. However, it may also represent a change in clinical practice, with earlier screening of affected families and routine cardiac assessment for patients with multi-systemic syndromes or diagnoses. 14% of patients were classified as lost to follow up (last clinical review more than 3 years ago) of whom 42% had been transitioned to adult care. Although the mortality rate is unlikely to be affected by this missing data due to nationally recorded death data in the UK National Health Service (NHS), other outcomes, such as arrhythmic events, could be underestimated. As patients were recruited from a number of centres, variations in clinical assessment and management between sites are inevitable. However, this is also a strength of the analysis, as it accurately reflects the clinical management of patients in the UK, both historically and in the current era.

3.6 Conclusions

In this chapter I describe the first European multi-centre investigation of paediatric HCM. The results show that, during childhood, HCM is a heterogeneous disease in terms of its age of presentation, aetiology and outcomes. Important novel findings include the detection of a phenotype in pre-adolescents with a family history of HCM and a change in the prevalence of syndromic HCM, particularly inborn errors of metabolism, over time. This demonstrates that changes in screening practices are changing the demographics of childhood HCM. Although the majority of children with HCM had good outcomes (freedom from death or transplantation was 90.6% at 5 years), SCD is the most common cause of mortality, occurring at a rate of 1.2 per 100 patient years at risk. SCD rates in childhood therefore remain higher than reported for adult cohorts. Patients diagnosed in infancy or with an inborn error of metabolism have a worse prognosis, most commonly secondary to CCF or non-cardiovascular death, but arrhythmic events are also seen in these patient groups. Although ICDs were implanted in one fifth of patients, the majority of these did not receive appropriate therapies and other patients were unprotected from lethal arrhythmic events during follow up. An improved understanding of the risk factors for SCD in childhood disease is needed to improve outcomes in this diverse patient group.

Chapter 4: Risk factors for SCD in childhood HCM: A systematic review and meta-analysis

4.1 Introduction

Sudden cardiac death (SCD) is the most common cause of mortality outside of infancy in childhood hypertrophic cardiomyopathy (HCM)[15, 81] with an estimated annual incidence between 1-2%[15, 16, 41]. Implantable Cardioverter Defibrillators (ICDs) have been shown to be effective at terminating malignant ventricular arrhythmias[159, 182], however identifying which patients are most likely to benefit remains challenging as shown in chapter 3. Risk factors for SCD in adult populations are well-described and current risk stratification guidelines for childhood disease are largely extrapolated from adult practice[4, 5]. A large number of potential paediatric specific risk factors for SCD during childhood have been reported in the literature over the last 30 years. However, to date, no study has investigated the strength of evidence supporting individual risk factors for SCD in childhood disease. The following work was published in the *European Journal of Preventative Cardiology* in 2017 [207].

4.2 Aims

The aim of this chapter was to perform a systematic review and meta-analysis of published literature to identify clinical risk factors for SCD in non-syndromic childhood HCM.

4.3 Methods

Study Selection

I searched the online MEDLINE and Pubmed database using the following Medical Subject Headings (MeSH) terms: "((hypertrophic cardiomyopathy) AND (death OR sudden death OR cardiac death OR outcome OR prognosis OR risk factors) AND (children OR childhood OR young OR paediatric OR pediatric)). Searches were limited to; original articles written in English, patients aged < 18 years, and publishing date 1963 – December 2015. I supplemented the search strategy by a manual search of the reference list for included papers and recent review articles.

Inclusion criteria

Studies were eligible for inclusion if reporting on a cohort of childhood HCM patients with a primary or secondary end point of SCD, SCD-equivalent event (aborted cardiac arrest or appropriate ICD therapy) or cardiovascular death (CVD). Studies with an end point of CVD secondary to heart failure alone were excluded. Studies not reporting estimates of association (including case-reports and letters) were also excluded. My population of interest was children (aged 1-18 years) diagnosed with non-syndromic HCM, studies were therefore excluded if the study cohort was; entirely infants (< 1 years), mixed adult/paediatric with < 75% under the age of 18 years without separate analysis of paediatric subjects, or limited to rarer phenocopies of sarcomeric HCM including Noonan syndrome or other RASopathies. Studies reporting on the use of genotyping or invasive clinical markers to predict SCD were not included in this review as the aim was to identify readily available clinical risk factors.

Data collection

I reviewed the titles and abstracts of all studies identified by the study selection strategy to determine eligibility. Dr Nicoletta Cantarutti independently reviewed the

same. All eligible studies were read in full and I extracted the following data; number of subjects, patient demographics (age, gender, aetiology), study design, clinical risk factor definitions, length of follow up and end-point (SCD, SCD-equivalent event or CVD). For all studies, event count data were extracted to calculate odds ratios (ORs). For studies reporting survival analyses, hazard ratios from univariate or multivariate Cox regression analyses were collected. In accordance with the Quality of Reporting of Meta-analyses (QUORUM) statement, an assessment of quality was performed for each included study[208].

Statistics

To account for heterogeneity in effect estimates between included studies, I performed a random-effects meta-analysis. I investigated the outcome of interest (SCD or SCD-equivalent event, CVD) as both a dichotomous, and time-dependent outcome. For survival analyses, reported hazard ratios from Cox's proportional hazard regression analysis were combined using the generic inverse variance method to create a pooled estimate of effect with 95% confidence intervals. The pooled estimate of effect reported is a weighted average of effect from the included studies where weight is calculated using estimates of precision (inverse of variance of the estimate). For studies considering SCD or SCD-equivalent event/CVD as a dichotomous outcome, I extracted event and risk factor counts to calculate the study odds ratio (OR). Pooled estimates of ORs with 95% confidence interval were generated using the Maentel-Haenszel (M-H) method. The measure of heterogeneity between included studies is described using I^2 where 0% represents no heterogeneity and 75% represent high heterogeneity. A significance level of 5% (P value < 0.05) was used for all analysis. The analysis was performed using Review Manager (version 5.3).

I received support from Dr Elena Pissaridou and Dr Deborah Ridout (Population policy and practice programme, Institute of Child health, UC, UK) to complete the analysis.

4.4 Results

The search strategy is summarised in Figure 5 below. 820 studies were identified on the initial literature search of which 699 were excluded based on the title and a further 88 on reading the abstract. The full text article was obtained for 33 studies resulting in the exclusion of a further 8 studies. 25 studies were included in the meta-analysis. Reasons for exclusion are described in Figure 8.

A description of included studies is shown in Table 13 below. Types of studies varied; retrospective cohort n=19 (76%), case control n=5 (20%), prospective cohort n=1 (4%). All but one study was retrospective. The end-point measured was SCD in 10 (40%), CVD in 7 (28%) and both SCD and CVD in 8 (32%).

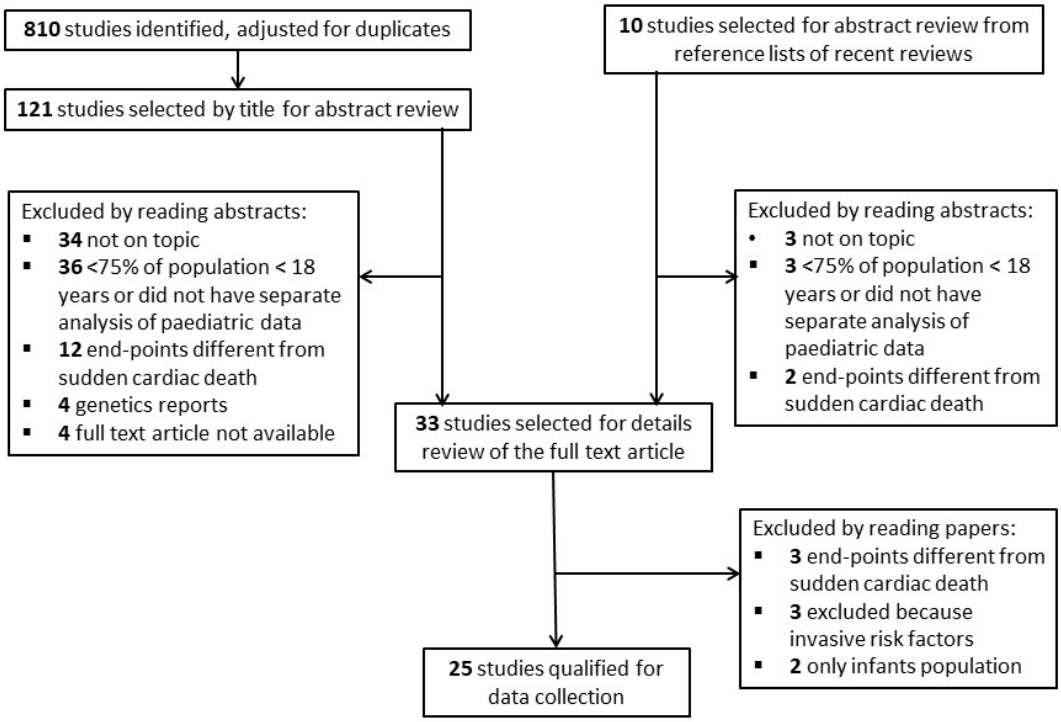


Figure 8 Consort flow diagram showing meta-analysis study selection process

References	Type	No.patients	Age (years)		Male (%)	Follow-up (years)		Endpoint	
			Mean +/-2SD	Median (IQR)		Mean +/-2SD	Median (IQR)	SCD	CD
Bharucha et al. 2015[169]	Case control	80	ND	0.45 (0.12 -2.48)	ND	ND	14 (10.7 -17.1)	Yes	Yes
Ziolkowska et al. 2015[41]	Cohort retrospective	112	ND	14.1 (7.8-16.6)	60	ND	6.5 (2.9-9.6)	Yes	Yes
Windram et al. 2015 [164]	Cohort retrospective	38	12.7+/-3.3	ND	79	ND	ND	Yes	
El-Saiedi et al. 2014[56]	Cohort retrospective	128	5.9 +/- 3.4	ND	63.3	7 (tot FU)	ND		Yes
Smith et al. 2014[165]	Cohort retrospective	30	14.1 +/- 3.2	ND	57	ND	2.2 (0.57-2.6)	Yes	
Smith et al. 2014[209]	Case control	30	14.1 +/- 3.2	ND	57	ND	2.3 (0.35-2.8)	Yes	Yes
Chaowu et al. 2013[210]	Case control	71	12.8 +/- 4.1	ND	65	2.4 +/- 1.6	ND	Yes	
Lipshultz et al. 2013[40]	Cohort retrospective	788	6 +/- 6.3	ND	68	ND	ND		Yes
Kamp et al. 2013[157]	Cohort retrospective	73	14.8 +/- 4.9	ND	59	ND	2.4 (0.1–12.9)	Yes	
Maron et al. 2013[158]	Cohort retrospective	224	14.5 +/- 3.6	ND	67	4.3 +/- 3.3	ND	Yes	
Hickey et al. 2012[58]	Cohort retrospective	120	9.2 & 6.8	ND	ND	8.2	ND		Yes
Maskatia et al. 2012[166]	Cohort retrospective	119	ND	12.7 & 9.9	79	ND	4.4 (2.1-6.6)	Yes	Yes
Moak et al. 2011[159]	Cohort retrospective	144	ND	14.4 (3-20)	70	ND	6.4 (3-9.6)	Yes	
Decker et al. 2009[57]	Cohort retrospective	96	10.6 +/- 5.4	12.2 (7.8-14.8)	69	6.4 +/- 5.2	5 (0.08-24)		Yes
Ostman-Smith et al. 2008[43]	Cohort retrospective	150	4.6	ND	58	7	ND	Yes	
Colan et al. 2007[16]	Cohort retrospective	634	ND	7.07 (0.28-13.17)	74	ND	2.1 (6.2m-12.3)		Yes

Nugent et al. 2005[15]	Cohort retrospective	80	ND	0.5 (0.12-2.5)	69	ND	5.25 (2.42-8.54)		Yes
Ostman-Smith at al. 2005[94]	Cohort retrospective	128	5.7 +/- 0.5	3.6	ND	11 +/- 0.8	ND	Yes	Yes
Mcmahon et al. 2004[211]	Cohort prospective	86	ND	12	65	ND	2.2 (0.2-4.25)	Yes	
Yanagi et al. 2004[170]	Case control	20	13.4 +/- 3.1	ND	70	7.2 +/- 4.4	ND	Yes	Yes
Butera et al. 2003[212]	Case control	17	10.3 +/- 5.8	ND	53	ND	ND	Yes	Yes
Yetman et al. 1998[59]	Cohort retrospective	99	ND	5 (1d-17)	72	ND	4.8	Yes	
Romeo et al. 1990[213]	Cohort retrospective	37	7 +/- 4	ND	65	9.2 +/- 5.1	ND		Yes
Mckenna et al. 1988[160]	Cohort retrospective	53	20+/1 & 13+/-7	ND	57	ND	3 (1w-7)	Yes	
McKenna et al. 1984[80]	Cohort retrospective	37	9	ND	ND	12	ND	Yes	Yes

Table 13 Description of included studies in the meta-analysis of risk factors for sudden cardiac death in childhood hypertrophic cardiomyopathy

(n=25) ND, not described; m, months; w, weeks; d, day

Twenty-three clinical risk factors were investigated. The definition of risk factors varied between studies and is described in Table 14 below.

Risk factors	Definition
Adverse cardiac event	Previous aborted cardiac event or appropriate ICD shock [57, 157] or spontaneous sustained VT [41, 159].
Extreme LVH	Maximum LV wall thickness ≥ 30mm[41] [157, 158] or z-score ≥ 6 [56, 57]
Syncope	Unexplained transient loss of consciousness at or prior to first evaluation [41]
NSVT	≥3 consecutive ventricular beats at rate ≥100 beats/min[159] or ≥120beats/min [41, 158] lasting < 30 seconds, evident on ambulatory ECG recordings.
Family history of SCD	≥1 first-degree relatives died suddenly, aged <40 years with or without diagnosis of HCM or when SCD occurred in a first-degree relative at any age diagnosed with HCM[41].
Left ventricular outflow tract obstruction	Peak gradient ≥16mmHg [15, 56, 57] or ≥ 20mmHg [59] or ≥ 30 mmHg [41] [58] by Doppler echocardiography in resting conditions.
Left atrial enlargement	Maximum anteroposterior linear diameter Z-score>2 measured at end-systole from the parasternal long-axis view [41] or LA–to–aortic root ratio ≥1.5[166] .
Abnormal blood pressure response to exercise	Failure to increase systolic BP by more than 20mmHg from rest to peak exercise [41, 57] or a fall greater than 20mmHg from peak pressure[159]

Table 14 Definition of clinical risk factors for sudden cardiac death used in the included studies

LVH = left ventricular hypertrophy, NSVT = Non-sustained Ventricular Tachycardia, ICD = Implantable Cardioverter Defibrillator, ECG = Electrocardiograph, VT = Ventricular Tachycardia, SCD = Sudden Cardiac Death, LA = left atrium, BP = blood pressure

For the purpose of analysis, and to provide a measure of the strength of evidence supporting individual risk factors, risk factors were defined as either probable/major risk factors (defined as being investigated in four or more studies and significantly associated with the end point in at least two univariate or multivariate analyses) or possible/minor risk factors (defined as being significantly associated with the end point in one or more univariate analyses but investigated in 3 or less studies).

Major clinical risk factors

Previous adverse cardiac event (aborted cardiac arrest or sustained ventricular tachycardia)

Five studies investigated the role of previous adverse cardiac event using an end-point of either SCD [41, 59, 157, 159] or all-cause CVD[57]. A significant association was reported in two univariate analyses [41, 159] and one multivariate analysis[41]. In a multivariable analysis of a small cohort of children with an ICD, Kamp et al [157]reported that a history of resuscitated arrest showed a trend towards an association but did not reach statistical significance (HR 4.5 (95% CI 0.8-26.1, p value 0.09)). Decker et al[57] did not find an association with the end point of CVD, of which only 3 (27%) were SCD, however risk factors for SCD were not separately investigated. The pooled HR and OR for previous adverse event was 5.4 (95% CI 3.67-7.95, P value <0.001) and 5.06 (95% 2.11-12.17, P value <0.001) respectively (Figure 9 below) No inter-study heterogeneity was observed (I^2=0).

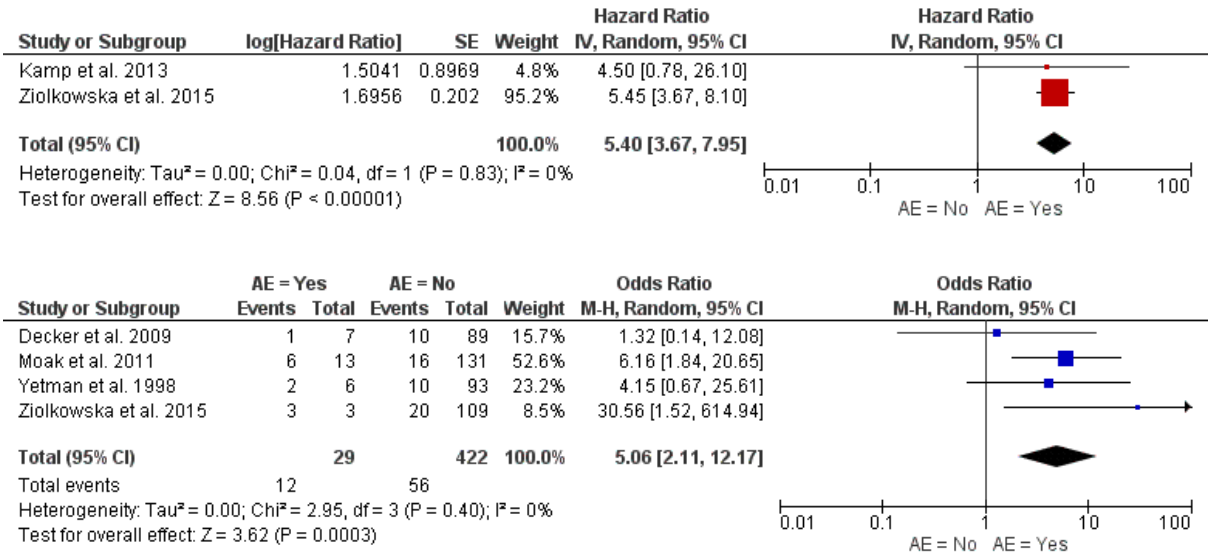


Figure 9 Pooled estimates for previous adverse cardiac event (VF/VT) and risk of sudden cardiac death

a) Hazard ratios for sudden cardiac death or cardiovascular death for previous adverse event (AE). The size of the square corresponds with the number of patients in the study. The bars represent the upper and lower 95% CI. Hazard ratios with CI >1 indicate a significant association with sudden cardiac death; b) Odds ratios for sudden cardiac death or cardiovascular death for previous adverse event. The size of the square corresponds with the number of patients in the study. The bars represent the upper and lower 95% CI. Odds ratios with CI >1 indicate a significant association with sudden cardiac death

Unexplained syncope

Seven studies assessed unexplained syncope as a risk factor for either SCD[41, 59, 80, 157, 159, 213] or CVD [41, 57, 80]. A significant association with SCD was reported in 3 univariate [80, 159, 213]and 1 multivariate analysis[213]. No study found a relationship between unexplained syncope and CVD. The temporal association of syncope and events has not been assessed in childhood. The pooled hazard ratio for unexplained syncope was 1.89 (0.69-5.16, p value 0.22) and pooled odds ratio 2.64 (1.21-5.79, p value 0.02) (Figure 10 below). Significant inter-study heterogeneity ($I^2$40-46%) was observed.

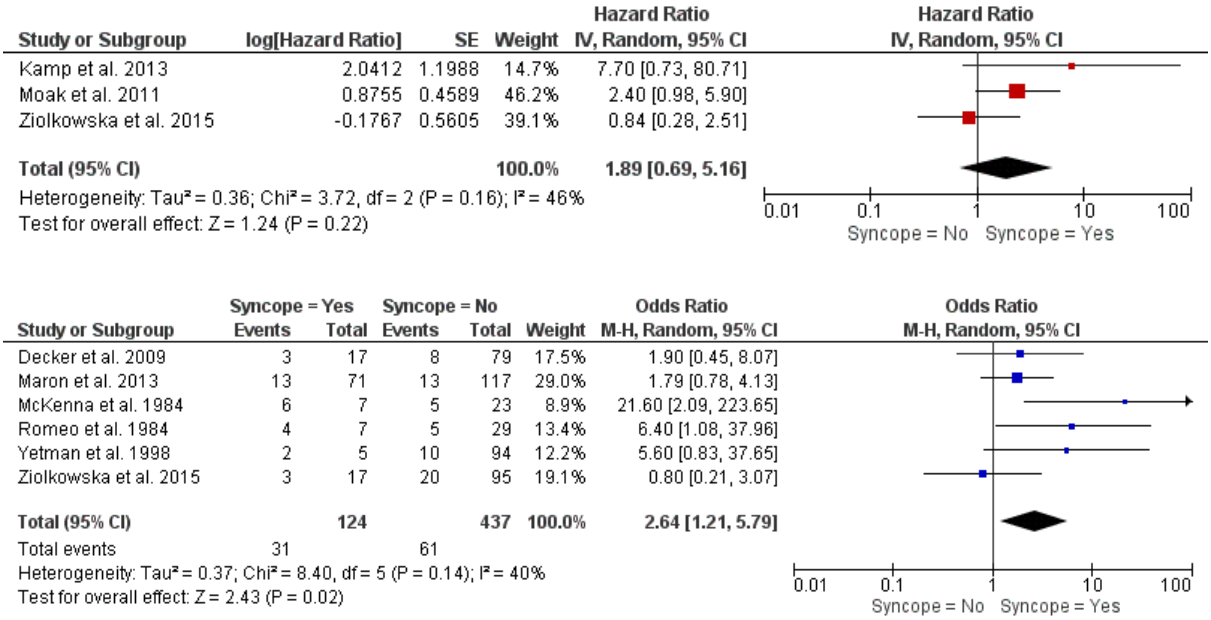


Figure 10 Pooled estimates for unexplained syncope and risk of sudden cardiac death

a) Hazard ratios for sudden cardiac death or cardiovascular death for unexplained syncope
b) Odds ratio for sudden cardiac death or cardiovascular death for unexplained syncope

Non-sustained ventricular tachycardia (NSVT)

Six studies investigated the association of NSVT detected on ambulatory ECG with SCD[41, 59, 158, 159] or CVD [56, 57]during follow up. Two studies reported a significant association with SCD on univariable analysis [59, 158]. Of two multivariable analyses with an end point of SCD, only one trended towards significance (P value 0.078)[41]. Moak et al [159]found no association of NSVT on ambulatory ECG recordings but a significant association with inducible VT on electrophysiology study. No studies with an end-point of CVD described a significant association with NSVT. No study explored the importance of rate, length or frequency of NSVT on the risk of SCD. The pooled HR and OR for NSVT was 2.13 (95% CI 1.21-3.74, p value 0.0009) and 2.05 (96% CI 0.98-4.28, p value 0.06) (Figure 11) . Mild heterogeneity between studies was seen ($I^2$11-19%).

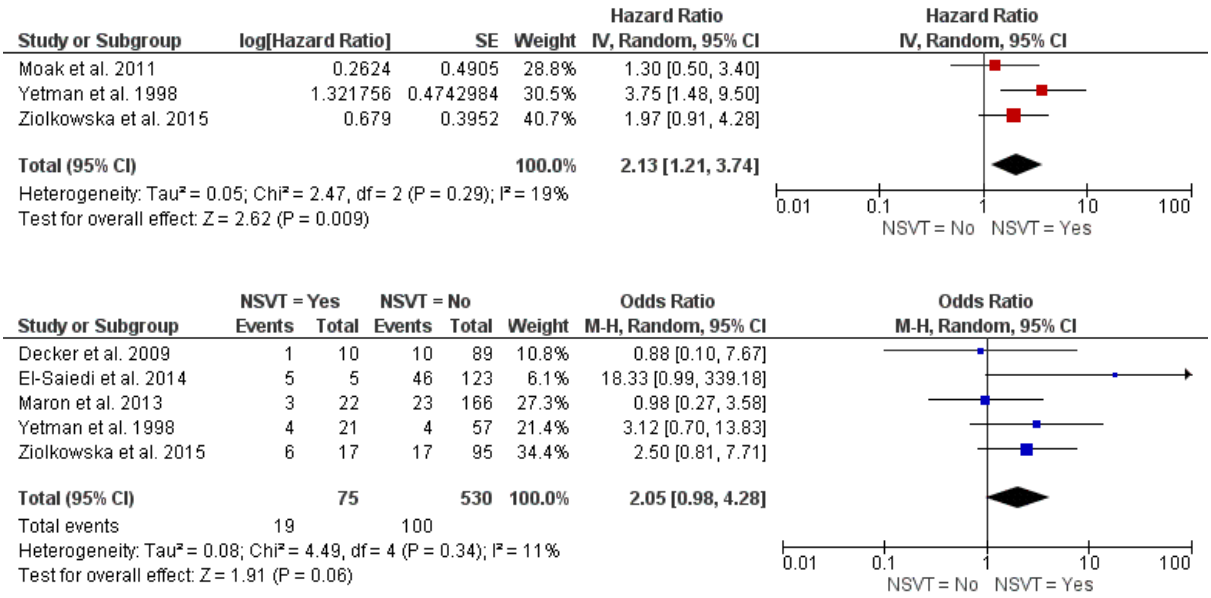


Figure 11 Pooled estimates for Non-Sustained Ventricular Tachycardia on ambulatory ECG and risk of sudden cardiac death
a) Hazard ratios for sudden cardiac death or cardiovascular death for NSVT b) Odds ratio for sudden cardiac death or cardiovascular death for NSVT

Left ventricular hypertrophy

Measures of left ventricular hypertrophy were investigated as a risk factor for SCD[41, 59, 94, 157-159, 166, 169, 211] or CVD[15, 40, 41, 56, 57, 94, 166, 169] in thirteen studies making it the most frequently assessed clinical risk factor. Studies assessing LVH are summarised in Table 15 below. The measure of hypertrophy varied widely; interventricular septal thickness (n=9), LV posterior wall thickness (n=6), LV wall thickness/cavity ratio (n=2) and extreme LVH (n=5). Six studies reported a significant association between LVH and SCD on univariable analysis[41, 94, 158, 159, 169, 211] (extreme LVH n=2, IVST n=2, LVPWT n=1, LVWT: Cavity n=1). Three studies with an end-point of SCD reported a multivariable analysis of which one reported a significant association between increasing IVST and SCD[94]. Measures of LVH were associated with an end-point of CVD in 6 studies on univariable analysis[15, 40, 41, 56, 57, 94]] (extreme LVH n=2, IVST n=1, LVPWT

n=3, LVPWT:cavity n=1) and 2 studies on multivariable analysis[56, 94] (extreme LVH n=1, IVST n=1, LVPWT:cavity ratio n=1). Extreme LVH (as defined in Table 14) was associated with SCD in half of the studies using this measure of hypertrophy[158, 211] and both studies with an end-point of CVD[56, 57]. The pooled hazard ratio for extreme LVH was 1.8 (95% CI 0.75-4.32, p value 0.19). The pooled odds ratio for extreme LVH was 1.70 (95% CI 0.85-3.40, p value 0.13) Figure 12 below. Moderate inter-study heterogeneity was observed ($I^2$21-31%).

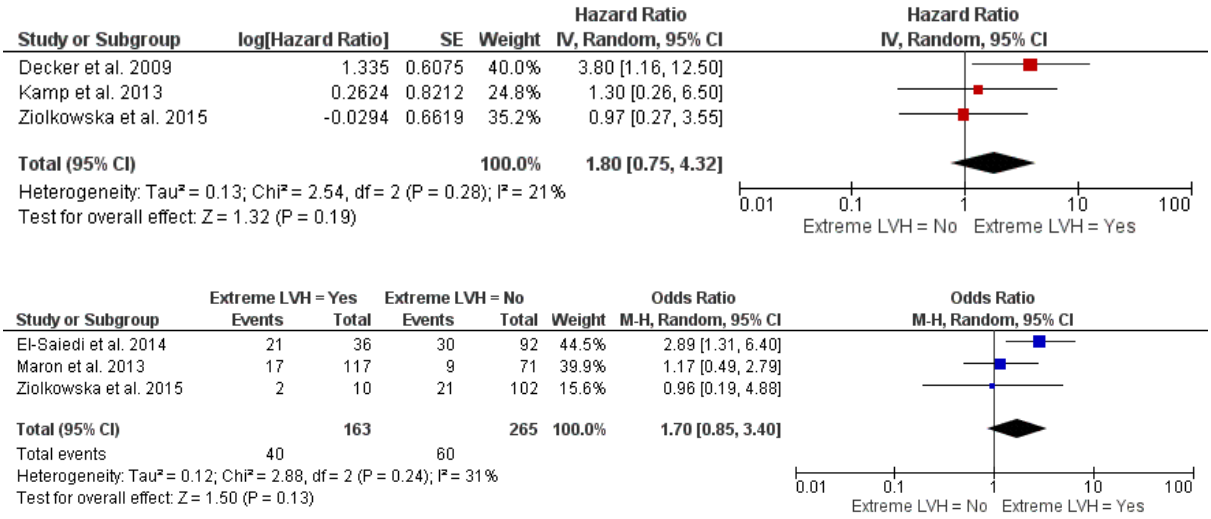


Figure 12 Pooled estimates for extreme left ventricular hypertrophy and risk of sudden cardiac death

a) Hazard ratios for sudden cardiac death or cardiovascular death for extreme LVH b) Odds ratio for sudden cardiac death or cardiovascular death for extreme LVH

References	N of patients	Follow-up (years)		SCD Endpoint			CVD end-point		
		Mean +/-2SD	Median (IQR)	SCD	UV (p value)	MV (p value)	CVD	UV (p value)	MV (p value)
Bharucha et al. 2015	80	ND	14 (10.7 - 17.1)	Yes	0.81 (2) **0.02 (3)**				
Ziolkowska et al. 2015	112	ND	6.5 (2.9-9.6)	Yes	0.71 (1), 0.39 (2), 0.068 (3), **0.03 (4)**		Yes	**0.01 (3)** 0.052 (4)	0.258 (3)
El-Saiedi et al. 2014	128	7 (tot FU)	ND				Yes	**0.07(1), 0.04 (2), 0.0001 (3)**	
Lipshultz et al. 2013	788	ND	ND				Yes	0.58 (2), **0.02 (3)**	
Maron et al. 2013	224	4.3 +/- 3.3	ND	Yes	**0.026 (1)**				
Maskatia et al. 2012	119	ND	4.4 (2.1-6.6)	Yes		0.37 (2)	Yes		
Moak et al. 2011	144	ND	6.4 (3-9.6)	Yes	**0.01 (2)**	0.13 (2)			
Decker et al. 2009	96	6.4 +/- 5.2	5 (0.08-24)				Yes	**<0.04 (1)**	
Nugent et al. 2005	80	ND	5.25 (2.42-8.54)				Yes	**0.02 (3)**	0.91 (3)
Ostman-Smith at al. 2005	128	11 +/- 0.8	ND	Yes	**0.001 (2)**	**0.036 (2)**	Yes	**0.003 (4)**	**0.005 (4)**
Mcmahon et al. 2004	86	ND	2.2 (0.2-4.25)	Yes	**0.002 (1),** 0.11 (2), 0.81 (3)				
Yetman et al. 1998	99	ND	4.8	Yes	0.11 (2)				
Kamp et al. 2013	73	ND	2.4 (0.1–12.9)	Yes	0.78 (1)				

Table 15 Summary of studies investigating measures of left ventricular hypertrophy and the risk of sudden cardiac death
Extreme left ventricular hypertrophy (LVH), (2) Interventricular Septal Thickness (IVST), (3) Left Ventricular Posterior Wall Thickness (LVLVPWT), (4) Left ventricular wall thickness (LVWT) to cavity ratio.
ND = not described. SCD = sudden cardiac death, CVD = cardiovascular death, SD = standard deviation, IQR = interquartile range

Minor clinical risk factors

Family History of sudden cardiac death (SCD)

A family history of SCD was investigated as a risk factor for SCD in 7 studies[40, 41, 56, 57, 59, 157, 159]. Kamp et al reported a significant association in a small cohort (n=73) of children with an ICD[157]. However, all other studies found no significant association. No study investigated the role of either the number of affected family members or degree of relativity in predicting risk.

Left ventricular outflow tract obstruction (LVOT obstruction)

Of six studies[15, 41, 56-59] investigating LVOT obstruction as a risk factor for SCD only 1 found a significant association[56], however the definition of LVOT obstruction varied as described in Table 14 above. El Saeidi et al reported an increased risk of CVD in the presence of a LVOT gradient ≥ 16mmHg. Both studies using the guideline endorsed threshold for LVOT obstruction of 30mmHg [41, 58]found no significant association between obstructive disease and SCD during follow up. One study described LVOT gradient as a continuous variable, and reported a higher risk of SCD with increasing gradient[41].

Left atrial size

Three studies explored the role of left atrial diameter as a clinical risk factor for either SCD [41, 166, 211] or CVD [41, 211]. Increasing LA size was associated with an increased risk of SCD in two out of three studies reporting a hazard ratio of 3.125 (95% CI 1.45-6.74, p value 0.001)[41] and 3.4 (95% CI 1.1-11.2, p value 0.049)[166] respectively.

Abnormal blood pressure response to exercise (ABPRE)

An ABPRE as defined in adult studies (Table 14 above) was not associated with the end point of SCD in any included study[41, 57, 157, 159]. Decker et al [57]reported an increased prevalence of CVD associated with ABPRE in a small cohort (n=68) of patients who underwent exercise testing.

Twenty-four hour blood pressure monitoring

One study reported an association between abnormal morning blood pressure ratio and the risk of SCD[170]. No other studies investigating blood pressure monitoring were identified.

Electrocardiograph (ECG) changes

Five studies investigated the role of different ECG abnormalities in predicting SCD risk[41, 59, 80, 94, 159]. QTc dispersion was found to be statistically associated with SCD in two studies on univariable or multivariable analysis[41, 59]. Corrected QT interval (ms), RS sum (mV) and ECG features of right ventricular hypertrophy were each associated with the end point of SCD in a single study. Heart rate variability on ambulatory ECG was investigated in two studies which both reported a correlation with SCD risk[170, 212], however this only reached statistical significance in one[212].

Late gadolinium enhancement (LGE) on cardiac magnetic resonance (CMR) imaging

Three studies looked at the predictive value of LGE on CMR as a risk factor for SCD in childhood HCM[164, 165, 210]. The presence of LGE was statistically associated

with LV wall thickness/mass, however although LGE was associated with an increased incidence of death, it was not an independent risk factor for SCD.

Gender

Gender was considered as a risk factor for mortality in five studies[40, 56, 94, 157, 169] of which only one reported a significant association. Lipshultz et al [40]described an increased risk of CVD for female patients on univariable analysis with a corresponding HR of 10.6 (95% CI 1.2-90.2, p value 0.03).

Symptoms

Cardiac symptoms other than syncope were investigated in eight studies[15, 40, 41, 57, 59, 94, 166, 213], however the definitions of 'symptomatic' patients were highly variable making interpretation challenging. The presence of heart failure symptoms was significantly associated with CVD in 3 out of 4 studies[40, 41, 213]. An end-point of SCD was used in 3 studies, of which only one found a significant association between the presence of symptoms and risk of SCD[166]. The definition of a patient being symptomatic in this study included a previous adverse event, syncope, chest pain, dizziness and palpitations. As this composite measure includes variables known to have an independent association with SCD, it is unclear if being symptomatic per se has an independent role in risk stratification.

Age

Age at presentation or diagnosis was considered as a possible risk factor in almost half of included studies[15, 16, 40, 41, 43, 56, 59, 94, 159, 166, 169]. Presentation in infancy was associated with an increased risk of CVD likely secondary to heart

failure deaths[15, 16, 56]. No study reported an association between presentation in infancy and SCD. Outside of infancy the majority of studies reported no significant association between age and outcomes but individual studies described increased risk of mortality at particular times in childhood including early childhood [94] or adolescence [43, 159].

4.5 Discussion and limitations

This chapter describes the results of the first systematic review of risk factors for SCD in childhood HCM which identified four 'major' clinical risk factors; unexplained syncope, NSVT, extreme left ventricular hypertrophy and previous VF/VT. Whilst some of the risk factors in childhood are the same as conventionally reported adult risk factors, important differences between risk stratification in children and adults are seen.

Conventional risk factors

Current guidelines for risk stratification recommend the use of four clinical risk factors for SCD in childhood [4, 5]. These are largely extrapolated from adult practice and include extreme left ventricular hypertrophy (defined as MWT ≥ 30mm or Z score ≥ 6), NSVT, unexplained syncope and a family history of SCD. A primary prevention device is recommended if a single risk factor is present in the North American guidelines, or if two or more risk factors are present in the European guidelines. However, whilst a large number of clinical risk factors for childhood disease have been reported in the literature, the absence of well-designed, large population studies means that the evidence supporting individual risk factors is not robust.

My meta-analysis supports the use of NSVT and unexplained syncope as a clinical risk factor for childhood disease with a pooled hazard ratio estimate of 2.13 (95% CI 1.21-3.74, p value 0.0009) and 2.64 (1.21-5.79, p value 0.02) respectively. Left ventricular hypertrophy was investigated as a clinical risk factor in a large number of studies and was classified as a major risk factor, however the most useful measure and threshold for assessing risk remains unclear. Of note, extreme hypertrophy was

only significantly associated with SCD in half of the studies using this as an end-point and the pooled HR and OR's did not reach statistical significance (HR 1.8 (95% CI 0.75-4.32, p value 0.19), OR 1.70 (95% CI 0.85-3.40, p value 0.13)). Recent studies in adults have described a non-linear relationship between MWT and risk meaning those with the most severe hypertrophy (>35mm) do not have the highest risk of arrhythmic events[115]. Additionally, the most recent adult European guidelines use MWT as a continuous variable in their risk algorithm[82]. The relationship between LVH and SCD risk in childhood is unknown and future studies are needed to determine the most clinically useful measure of LVH for risk stratification in paediatric practice.

In contrast, my meta-analysis did not find evidence to support the use of family history of SCD as a risk factor for childhood disease. Although it was investigated in 7 studies, only 1 found an association in a small cohort of patients with an ICD who had *a priori* been determined to be high risk for malignant arrhythmias by the treating clinician[157]. Its inclusion in the guidelines is supported by a single study which was excluded from this meta-analysis as it excluded patients under the age of 14 years and only 11% of the cohort was under the age of 20 years[125]. There is robust evidence to support the use of family history in adult patients, however there is currently insufficient evidence to support its use during childhood. Possible explanations for this include a higher prevalence of de novo variants in childhood HCM, low proportion of sarcomeric disease in the included cohorts or insufficient reporting of family history.

Additional risk factors for childhood disease

Adult risk stratification guidelines describe additional clinical risk factors, including left atrial diameter, LVOT gradient and ABPRE, which can modify an individuals' risk.

Although only 4 major risk factors were identified in this review, our data suggest that additional risk factors could similarly be useful in paediatric practice. Left atrial diameter was only investigated in 3 studies, but was significantly associated with SCD in 2. Therefore, whilst it did not meet the criteria for a 'major' risk factor, it is likely to be important and future studies are needed to confirm this. LVOT obstruction was investigated in 6 studies, however the definition of 'obstruction' varied and the guideline endorsed definition (≥30mmHg) was used in only 2, both of which reported no association with SCD. This limited our ability to assess its role as a risk factor in childhood disease but suggests it may differ from that previously reported in adults. Childhood is also a time of considerable somatic growth meaning that the cardiac phenotype may change considerably over the course of follow up. This may have important implications for risk stratification but it has not yet been explored.

Limitations

The number of studies included in this meta-analysis was small and all but one was retrospective. It is therefore limited by problems inherent to retrospective studies including missing data and incomplete information. Childhood HCM is a rare disease and included studies reported small, often heterogeneous, patient cohorts (all but 3 had less than 150 participants). Historically, population studies have reported cohorts from specialised tertiary centres that care for highly symptomatic patients. The implication of this is that included patients may have more severe disease and not be representative of the wider population.

As the incidence of SCD in childhood disease is low and many patients considered high risk have undergone ICD implantation, a composite end-point of SCD or equivalent event (resuscitated cardiac arrest, appropriate ICD therapy, sustained VT)

was used in some studies. Although this is accepted practice, it should be recognised that these events, particularly appropriate ICD therapy or sustained VT, are not truly equivalent to SCD. Pooled hazard and odds ratios for SCD are reported, however the precision of estimates is low (as reflected in the width of the confidence intervals) due to the small number of studies, low event rate and heterogeneity of included studies.

Finally, comparisons between studies was complicated by differences in study design and the definition of individual risk factors. This was a particular problem for LVH where the measure of hypertrophy (eg ISVT, LVPWT, cavity ratio, extreme hypertrophy) differed as well as the choice of z-score calculator, each of which is derived from a different normal population and correlate poorly with each other.

4.6 Conclusions

In this meta-analysis, I have identified four 'major' clinical risk factors for SCD in childhood HCM; unexplained syncope, NSVT, extreme left ventricular hypertrophy and previous VF/VT. Important differences exist between risk stratification in children and adults. Well-designed, large population studies are required to systematically investigate paediatric specific risk factors.

4.7 Update to meta-analysis

Since the publication of this meta-analysis a further 5 studies have explored clinical risk factors for SCD in childhood HCM as summarised in Table 16 below. The results largely support the findings and provide further evidence supporting the use of clinical risk factors such as LVH, unexplained syncope, LA diameter and NSVT in risk prediction during childhood. Meanwhile other risk factors, including a family

history of SCD, ABPRE and LGE on CMR, were not found to be associated with arrhythmic events reinforcing differences between adults and children with the disease. Of note, whilst one further study reported an association between LVOT obstruction (≥20mmHg)[161] and SCD, two large studies described an inverse relationship between LVOT gradient and risk in childhood[55, 214].

Candidate predictor	No. studies using CVD as end point	No. studies using SCD as end point	No. Studies showing association with SCD on UV	No. Studies showing association with SCD on MV	No. studies showing association with CVD on UV	No. Studies showing association with CVD on MV
Left ventricular hypertrophy	1 [81]	4 [161] [214] [55] [60]	2 [161] [55]	2 [214] [55]	1 [81]	1 [81]
Male sex	1 [81]	1 [161]				
LVOT obstruction		2 [161, 214]	1 [161]			
NSVT		4 [161] [214] [55, 60]	1 [161]	1 [214]		
FHx of SCD		3 [161] [214] [55]				
Syncope		3 [161] [214] [60]		1 [214]		
ECG		1 [161]	1 [161]	1 [161]		
Age		2 [214] [60]		1 [214]		
Left atrial diameter		1 [214]		1 [214]		
Abnormal BP response to exercise		1 [55]				
Late gadolinium enhancement on CMR		1 [46]				

Table 16 Summary of studies investigating clinical risk factors for sudden cardiac death from 2016 onwards
Measures of LVH; IVST [55, 161, 214], LVPWT [81] [214] [55, 161], extreme hypertrophy [60]. CVD = Cardiovascular Death, SCD = Sudden Cardiac Death, UV = univariable, MV = multivariable, CMR = cardiac magnetic resonance, BP = blood pressure, ECG = electrocardiograph

Chapter 5: Validation of the current European and North American guidelines for risk stratification in childhood

5.1 Introduction

Sudden cardiac death continues to be the most common cause of mortality in childhood HCM outside of infancy, and as shown in chapter 3, appears to occur at a higher rate compared to adults with the disease [81, 205]. Risk stratification for arrhythmic events is therefore an important part of clinical management. As described in chapter 4, clinical risk factors for SCD used in childhood disease are largely extrapolated from adult practice and the evidence supporting individual risk factors is not robust[207]. Nevertheless, current guidelines recommend the use of four traditional clinical risk factors for the purposes of risk stratification; unexplained syncope, NSVT on ambulatory ECG monitoring, family history of SCD and extreme LVH (defined as a MLVWT ≥ 30mm or Z-score ≥6) [4, 5]. The threshold for recommending ICD implantation varies between the European and North American guidelines. One risk factor is sufficient to consider a device in the North American guidelines, whereas two or more risk factors are needed in the European guidelines. Whilst these guidelines are widely used, the efficacy of this approach to risk stratification in childhood HCM has not previously been validated. This chapter is based on work published in Europace in 2019 [215].

5.2 Aims

The aim of this chapter was to perform the first external validation study of risk stratification guidelines in an international, multi-centre cohort of childhood HCM.

5.3 Methods

The study population has been previously described in Chapter 2 [216]. 1029 patients with childhood-onset non-syndromic HCM were identified from the International Paediatric Hypertrophic Cardiomyopathy Consortium. Patients with a secondary prevention indication for ICD implantation (eg history of documented sustained ventricular arrhythmia or resuscitated cardiac arrest) were excluded from this analysis.

Data collection

Data collection and clinical investigations are described in Chapter 2. In accordance with the 2014 European Society of Cardiology (ESC) [4] and 2011 American College of Cardiology Foundation/American Heart Association (ACCF/AHA) [5] guidelines, risk factors for an arrhythmic event were assessed at baseline and comprised; NSVT on ambulatory ECG monitoring, unexplained syncope, extreme LVH (MWT≥30mm or Z score ≥6), and a family history of SCD. Both the ESC and ACCF/AHA guidelines assess equal weight to each clinical risk factor so that the overall clinical risk profile is calculated as the sum of risk factors present. Patients were classified as 'high' or 'low' risk for an arrhythmic event as per guideline recommendations (ESC guidelines: 'high risk' if 2 or more risk factors present; ACCF/AHA guidelines: 'high risk' if a single risk factor present [extreme LVH, Unexplained syncope, family history of SCD]).

Study outcome and follow up

As described in Chapter 2 the primary patient outcome, taken from last clinical review, was a composite outcome of SCD or an equivalent event (resuscitated

cardiac arrest, appropriate ICD therapy or sustained VT with haemodynamic compromise) defined in this chapter as a Major Arrhythmic Cardiac Event (MACE).

Statistical analysis

General statistical methods are described in Chapter 2.

Due to small patient numbers, patients with two or more risk factors were combined and group differences in survival were assessed using the log-rank test. For the purposes of assessing the discriminatory ability of the guidelines, I coded missing data as absent, reflecting the use of the guidelines in clinical practice. I considered the risk factor profile to be a continuous score ranging from 0 (when no risk factors were present) to 4 (when all clinical risk factors were present) and the C-statistic at 1 and 5 years was estimated. I constructed the receiver operating curve by plotting the sensitivity against (1-specificity) for all possible prognostic values (eg. ≥0, ≥1, ≥2, ≥3 or ≥4 risk factors) and calculated the area under the curve. The positive and negative predictive value of the guideline threshold was expressed as a percentage and calculated as shown below.

Positive predictive value:

$$\frac{sensitivity \times prevalence}{((sensitivity \times prevalence) + (1 - specificity) \times (1 - prevalence))}$$

Negative predictive value:

$$\frac{sensitivity \times (1 - prevalence)}{((1 - sensitivity \times prevalence) + (specificity) \times (1 - prevalence))}$$

5.4 Results

Baseline demographics

1029 patients (male n=702, 68.2%) presented with HCM at a median age of 11 years (IQR 7- 14). Baseline clinical characteristics are shown in Table 17 and Figure 13 below

Baseline clinical characteristic		**N (%)**
Age (median, IQR)		11 (7,14)
Male gender		702 (68.2%)
Family history HCM (n=1011)		537 (53.1%)
Family history SCD (n=1025)		131 (12.8%)
Unexplained Syncope (n=1028)		102 (9.9%)
NYHA/Ross (n=1011)	**1**	788 (77.9%)
	2	191 (19%)
	3	29 (2.9%)
	4	3 (0.3%)
Medical therapy at baseline (n= 1026)	**None**	599 (58.4%)
	B-Blockers	412 (40.2%)
	Amiodarone	9 (0.9%)
	Other	6 (0.6%)
NSVT on ambulatory ECG (n= 856)		55 (6.4%)
MWT (mm) [mean, +/- SD] (n=997)		17.1 (7.4)
Z score MWT [mean, +/- SD] (n=906)		11.1 (7.1)
Extreme LVH (MWT ≥ 30mm/ Z score ≥ 6)		290 (28.2%)
LA diameter (mm) [mean, +/- SD] (n=712)		33.4 (8.5)
Z score LA diameter [mean, +/- SD] (n=675*)		1.9 (2.3)
LVOTg max (mmHg) [median, IQR] (n=871)		9 (6, 22)
LVOT obstruction (LVOTg ≥30mmHg)		189 (18.4%)

Table 17 Baseline clinical characteristics of non-syndromic international paediatric hypertrophic cardiomyopathy consortium cohort

N=1029 unless otherwise indicated. HCM=hypertrophic cardiomyopathy, SCD=sudden cardiac death, IQR= interquartile range, NYHA = New York Heart Association, NSVT = Non-sustained ventricular tachycardia, MWT = maximal wall thickness, LVH = left ventricular hypertrophy, LA = left atrial, SD = standard deviation, LVOTg = left ventricular outflow tract gradient

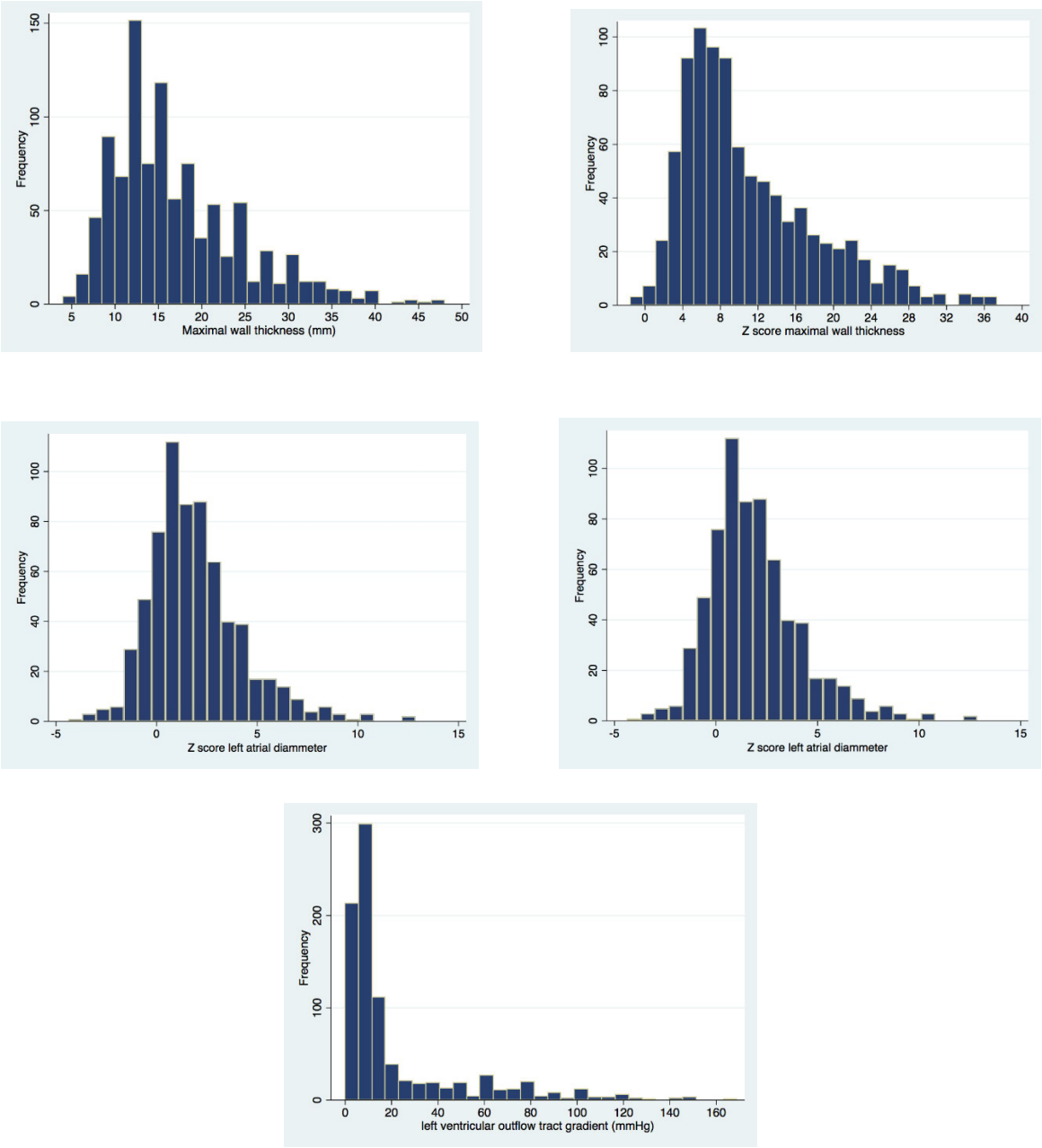


Figure 13 Histograms showing clinical phenotype at baseline
a) Maximal LV wall thickness (mm) b) Maximal LV wall thickness Z score c) left atrial diameter (mm) d) left atrial diameter Z score e) left ventricular outflow tract gradient (mmHg)

Prevalence of clinical risk factors

The number of traditional clinical risk factors at baseline varied; 0 (n=599, 58.2%), 1 (n=339, 32.9%), 2 (n=75, 7.3%), 3 (n=15, 1.5%), 4 (n=1, 0.1%). Complete data was available for 813 patients (79%), data were missing for 1 or 2 traditional risk factors in 214 (20.8%) and 2 (0.2%) patients respectively.

Outcomes

Over a median follow up 5.5 years (IQR 2.6-10.2), 66 (6.4%) patients died (SCD n=45 (4.4%), CCF n=6 (0.6%), CVA n=2 (0.2%), other-CV n=4 (0.4%), non-CV n=2 (0.2%), unknown cause n=7 (0.7%)) and 25 (2.4%) had a cardiac transplant. Overall annual mortality incidence was 0.90 per 100 patient years (95% CI 0.71-1.15). Age at the time of death and estimates of survival are shown in Figure 14 below. Seventy-seven (7.5%) required a LV myectomy and 35 (3.5%) underwent permanent pacemaker implantation (sinoatrial disease n=7, atrioventricular (AV) node disease n=10, LVOT obstruction n=11, unknown indication n=7). An implantable cardioverter defibrillator (ICD) was implanted in 268 (26.2%) patients for primary (n=246) or secondary (n=22) prevention at a median age 15.2 years (IQR 13.0 – 17.3).

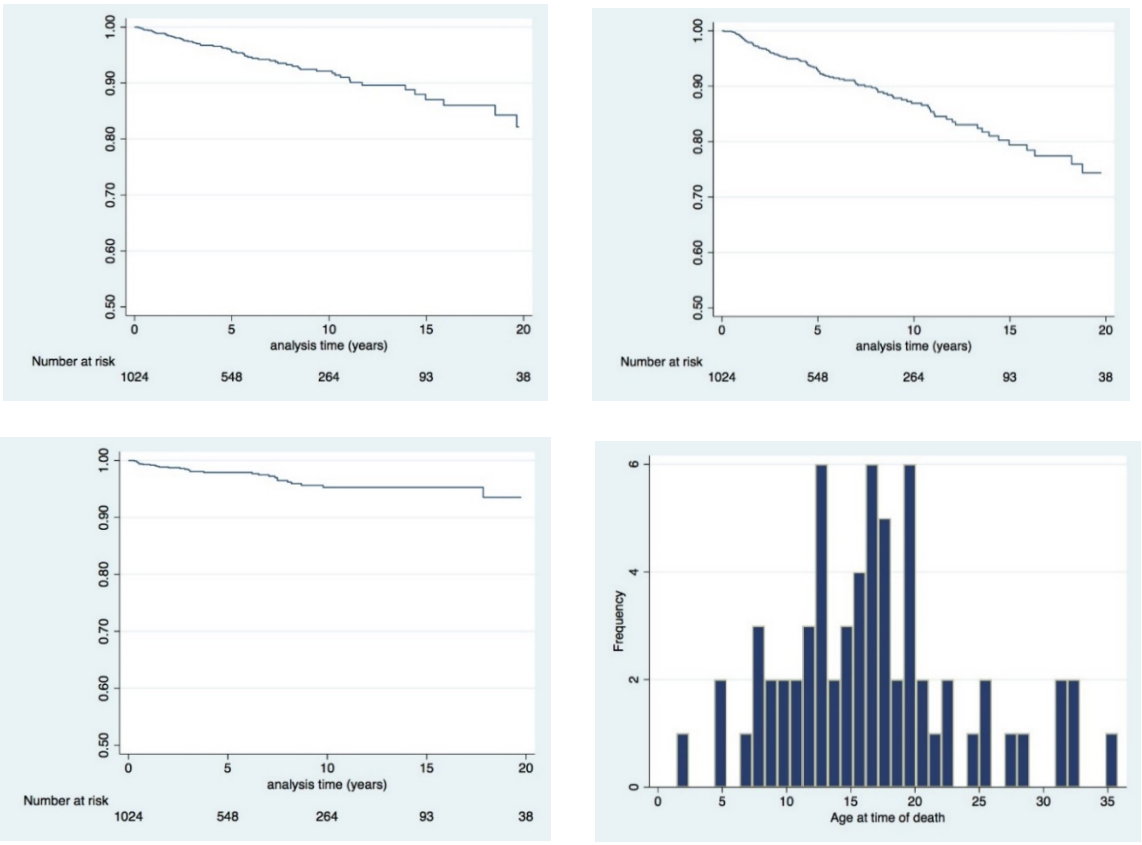
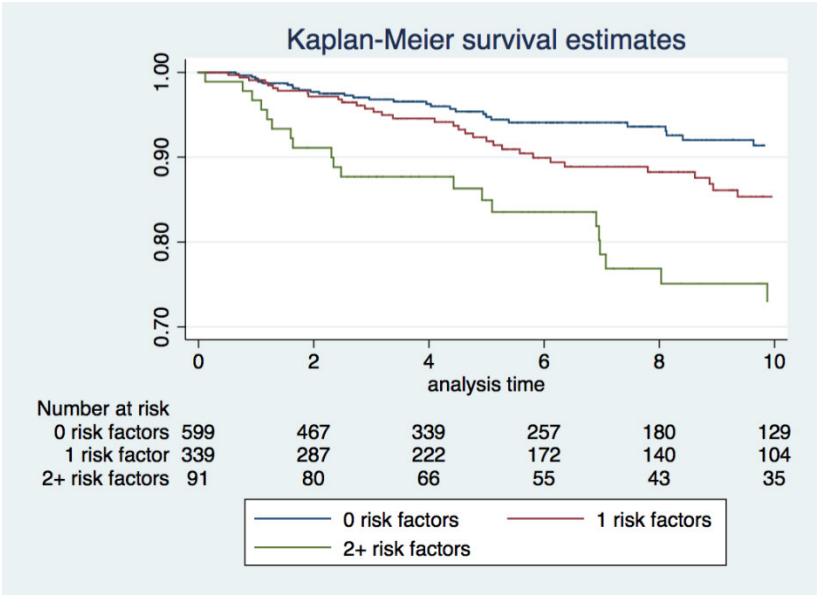


Figure 14 Mortality and arrhythmic events in childhood hypertrophic cardiomyopathy

Kaplan Meier survival curve showing event free survival estimates from a) all-cause mortality b) SCD or equivalent event c) Heart failure deaths or cardiac transplantation d) age at time of death

Arrhythmic events and clinical risk profile

One hundred and four patients (10.1%) had a MACE (SCD n=45 (4.4%), resuscitated cardiac arrest n=16 (1.6%), appropriate ICD therapy n=32 (31%) or sustained VT n=11 (1.1%)) with an overall annual incidence rate of 1.4 per 100 patient years (95% CI 1.18-1.73). Fifty-eight events occurred within the first 5 years of follow up. MACE occurred in 38 patients (6.3%) with no clinical risk factors, 41 (12.1%) with 1 risk factor, 20 (26.7%) with 2 risk factors, and 5 (50%) of those with 3 risk factors. A single patient with 4 clinical risk factors was followed up for 5.7 years with no MACE. Annual incidence rate per 100 patient years of MACE for patients with 0, 1 or more than 2 risk factors was 1.0 (95% CI 0.75 – 1.42), 1.53 (95% CI 0.11-2.08) and 2.67 (95% CI 1.80 – 3.95) respectively. Event free survival free estimates for patients with different clinical risk profiles is shown in Figure 15 below. Of 339 patients with a single clinical risk factor; 178 (52.5%) had extreme LVH, 82 (24.2%) family history of SCD, 54 (15.9%) history of unexplained syncope, and 25 (7.4%) NSVT detected on ambulatory ECG. Annual MACE incidence rate per 100 patient years for patients with isolated NSVT, unexplained syncope, extreme LVH or a family history of SCD was 1.80 (95% CI 0.74-4.32), 1.09 (95% CI 0.45-2.61), 1.7 (95% CI 1.13-2.56) and 1.36 (95% CI 0.68-2.71) respectively. Event free survival estimates for patients with a single clinical risk factor is shown in Figure 15 below.



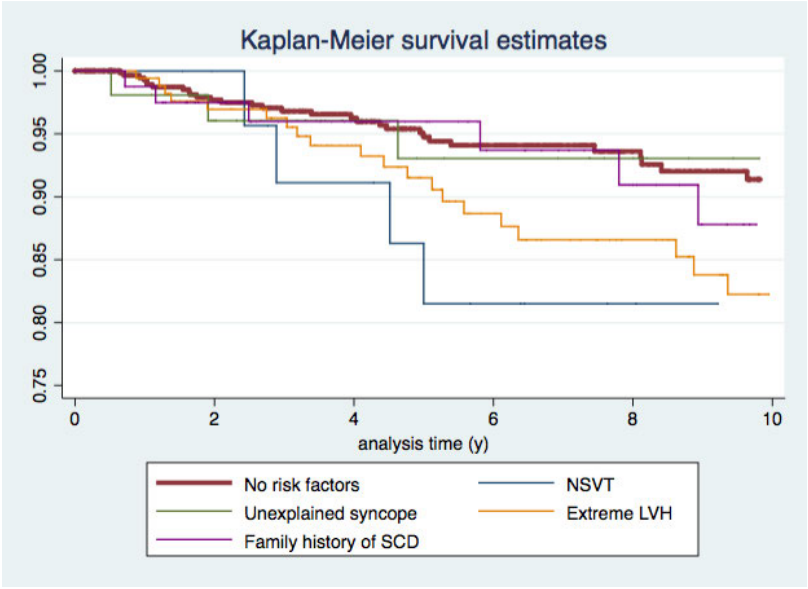


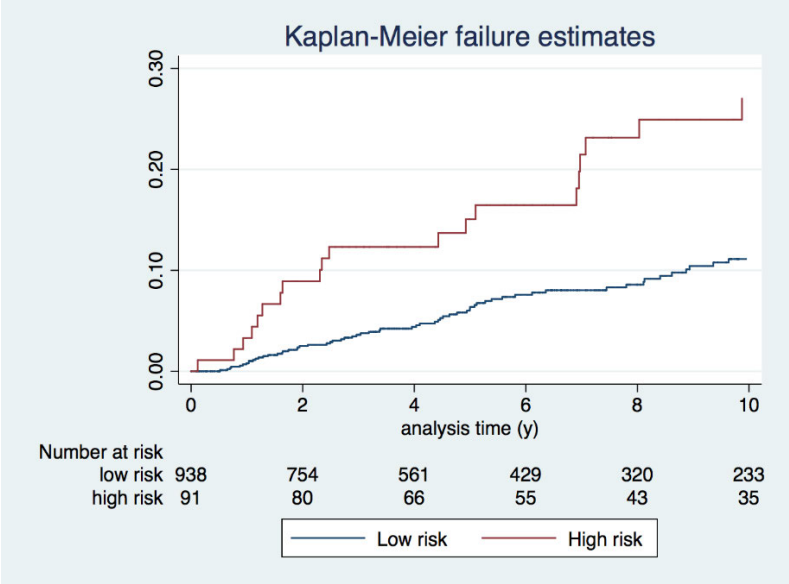
Figure 15 Kaplan Meier Survival curve showing estimates of event-free survival from a major arrhythmic clinical event
A) patients with different ESC clinical risk profile*s [log-rank test 0.8993] and B)* patients with a single clinical risk factor

Discrimination performance of European Society of Cardiology risk stratification guideline

Patients classified as 'high risk' according to the ESC guidelines (≥2 RF) were more likely to experience a MACE during follow up compared to those classified as 'low risk' (<2 RF) [hazard ratio 2.28 (95%CI 1.44-3.60), p 0.0011] Figure 16 below. The C-statistic at 1 and 5 year follow up was 0.63 (95% CI 0.44-0.82) and 0.62 (95% CI 0.55-0.70) respectively. The corresponding 5 year PPV and NPV of using the ESC treatment thresholds to guide ICD implantation was 9.0% and 94.5%.

Discrimination performance of American Heart Association risk stratification guideline

Patients classified as 'high risk' according to the AHA guidelines (≥1 RF) were more likely to experience a MACE during follow up [hazard ratio 1.70 (95% CI 1.14-2.52), p value <0.001] Figure 16 below. The C-statistic at 1 and 5 year follow up was 0.65 (95% CI 0.46-0.84) and 0.60 (95% 0.53-0.68) respectively. Five year PPV and NPV of using the AHA treatment threshold to guide ICD implantation was 7.7% and 94.4%.



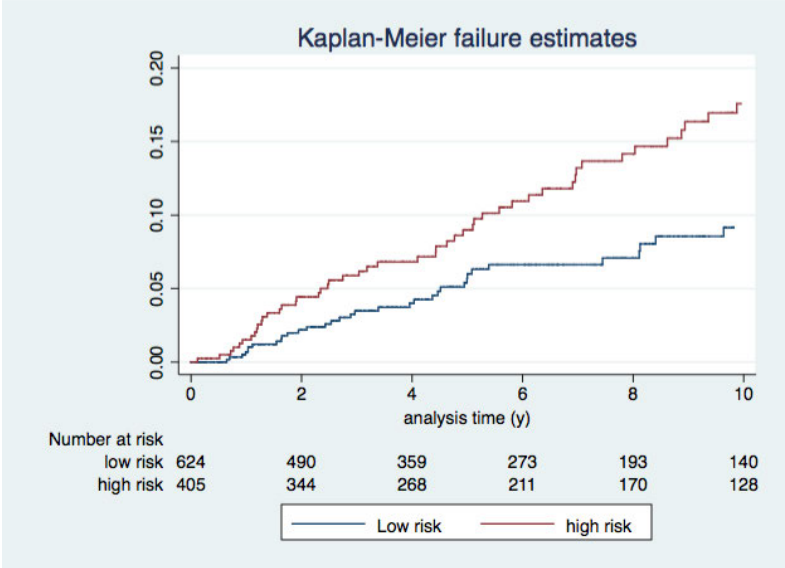


Figure 16 Cumulative incidence of major arrhythmic cardiac events for patients classified as high risk by current guidelines
A)European society of cardiology: low risk (<2 risk factors) vs high risk (≥2 risk factors, B) American heart Association: low risk (0 risk factors) vs high risk (≥1 risk factor)

5.5 Discussion and limitations

In this chapter I have described the results of the first external validation of current risk stratification guidelines for SCD in childhood HCM[215] showing that they have only a modest ability to discriminate between patients at high and low risk for a future arrhythmic event (C statistic 0.62 and 0.60 at 5 years) leading to unnecessary ICD implantation in many.

Prevalence of risk factors for arrhythmic events in childhood disease

As discussed in chapter 4 the evidence supporting the use of traditional risk factors in childhood disease, particularly for extreme LVH and family history of SCD, is limited[207]. Nonetheless, both the European [4] and North American [5] guidelines recommend their use in risk stratification. In this large cohort of patients with childhood HCM, the majority (58%) had no conventional risk factors for SCD and only a small number (n= 91, 8.8%) met the ESC threshold for ICD implantation. The most common risk factor was extreme LVH, which was present in one third of patients. NSVT was detected infrequently (6%). Despite this, a quarter of patients underwent ICD implantation during follow up, the majority of which were for primary prevention. Although an individual's phenotype could be expected to change during childhood, this discrepancy suggests that clinicians are taking into account other clinical parameters not included in the guidelines when assessing risk. Additionally, although only a small number of patients had multiple clinical risk factors for SCD, the overall rate of events was higher in this study compared to similar sized adult cohorts (1.4 vs 0.8/100 pt years)[185]. This suggests that additional or alternative risk factors, not currently included in the guidelines, may be important for childhood disease.

A caveat to this is that one fifth of patients had missing data for one or more clinical risk factors meaning their risk profile was incomplete. Missing data was most common for NSVT (17%), which could reflect difficulties in obtaining certain investigations in the very young. Although this approach to analysis has been used in previous adult cohort studies [184], it may have led to an under-estimation of the prevalence of individual risk factors. However, this approach likely reflects real world clinical practice where absence of a result may be taken to be equivalent to absence of risk meaning that it is likely to be a true representation of how the guidelines perform.

Discriminatory power of current risk stratification guidelines

My analysis shows that the incidence of an arrhythmic event increased incrementally in the presence of additional risk factors, which is perhaps not surprising given that each traditional risk factor is a marker of more severe disease. 'High risk' patients, as determined by a threshold of 1 or 2 risk factors, had a higher incidence rate of arrhythmic events occurring during follow up. However, one third of events occurred in patients with no risk factors and the majority of events (76%) were in patients classified as 'low risk' by the ESC guidelines. The ability of the guidelines to discriminate between low and high risk patients is therefore limited.

Children are known to be at an increased risk of long-term complications following ICD implantation[159, 182], yet one quarter of this cohort received a device. Of these, only 12% received an appropriate ICD therapy during the follow up of the study highlighting the difficulties in identifying those at increased risk who may benefit from device implantation. Indeed, 42% of MACE occurred in patients not judged to be at high risk by clinicians who therefore did not undergo ICD implantation. Both guidelines had a low PPV (7-9%) meaning that if the guidelines

were applied, by far the majority of patients classified as high risk would not experience an event but would be exposed to the potential complications of a device. Whilst both guidelines have a limited ability to correctly identify those at high risk of an event resulting in unnecessary ICD implantation, the main difference lies in the number of patients exposed to risk of ICD complications. The North American guidelines [5] recommend considering an ICD in the presence of a single risk factor meaning that a larger number of patients (two-fifth) are classified as high risk. A solitary risk factor was indeed associated with an increased incidence of an arrhythmic event in this cohort. NSVT, although infrequently detected, appeared to have the greatest effect although confidence intervals are wide limiting my ability to make meaningful comparisons. However, whilst a solitary risk factor was associated with an increased incidence of an arrhythmic event, the discriminatory power of this threshold was low with no change in the negative predictive value (94%) compared to the ESC threshold for intervention. This means that more patients are exposed to the risk of an ICD with no significant improvement in the discrimination between high and low risk patients. Large multi-centre collaborative studies are needed to investigate the relationship between individual risk factors and SCD.

My findings are in agreement with published adult validation studies of the current adult North American guidelines that use a similar approach to risk stratification [184]. This approach provides relative risks for non-homogenous groups rather than individualised estimates, and converts continuous variables (eg MWT) into binary variables for the purposes of risk stratification. As described in chapter 4, whilst the degree of LV hypertrophy has been shown to be associated with SCD risk in childhood, only half of studies using a threshold of ≥30mm/≥Z score 6 reported a significant association[207]. Additionally, adult cohort studies have described an

inverted relationship between MWT[115] and risk meaning those with the most severe hypertrophy are not at the highest risk of arrhythmic events. The clinical validity of transforming data in this way for the purpose of risk stratification is therefore unclear.

Limitations

This analysis was limited by inherent problems of retrospective data, in particular missing or incomplete data. As described in Section 6.5 above, this could have led to under-estimating the prevalence of traditional risk factors in childhood disease. As childhood HCM is a rare disease[12, 14] and SCD a relatively rare outcome[15, 81, 205], despite this cohort containing the largest population of non-syndromic HCM to date, there were small numbers of events and small numbers of patients classified as ‘high risk’ according to the ESC guidelines. This reduced my power to detect statistically significant differences and as a result the reported confidence intervals are wide.

Childhood HCM is recognised to be a heterogeneous disease[15, 16, 205] but current risk stratification approaches do not account for this heterogeneity. Although the ESC and AHA guidelines do not specifically exclude patients with syndromic disease, it is generally accepted that they are clinically distinct from sarcomeric disease and should be treated as a separate entity. However, it is increasingly recognised that these patients do experience arrhythmic events[31, 47, 205] and continue to be exposed to this risk throughout childhood if they survive outside of infancy[81]. My analysis excluded patients with syndromic and infantile-onset disease as intended by the guidelines, but large multi-centre studies are needed to

systematically assess the clinical risk factors for arrhythmic events in these patient groups.

5.6 Conclusions

In this part of my thesis I have performed the first validation of the current risk stratification guidelines in childhood HCM. I found that although the incidence of an arrhythmic event increased with additional risk factors, the guidelines have a limited ability to distinguish between high- and low-risk patients leading to unnecessary ICD implantation in many patients.

Chapter 6: Developing the first validated risk prediction model for sudden cardiac death in childhood hypertrophic cardiomyopathy

6.1 Introduction

In 2014 O'Mahony et al developed the first validated risk prediction model for SCD in adult hypertrophic cardiomyopathy [82]. HCM Risk-SCD uses readily available clinical risk factors to calculate an individualised estimate of 5-year SCD risk and has been shown to have an improved discriminatory ability compared to traditional risk stratification tools, which provide relative rather than absolute estimates of risk [185-187]. This model has been adopted by the adult European Society of Cardiology guidelines with 3 categories of risk defined; < 4% ICD not generally indicated, 4-6% ICD may be considered, and ≥ 6% ICD should be considered [4]. However, this model cannot be used in paediatric practice as patients under the age of 16 years were excluded from model development and it has not been validated for use in a childhood population.

In contrast to the progress made in adult HCM risk stratification, risk stratification approaches in paediatric practice have remained fundamentally unchanged for over two decades and are largely extrapolated from adult experience [4, 5]. In chapter 5, I showed that this approach to risk stratification has a limited ability to discriminate between children at high and low risk with unnecessary ICD implantation in many [215]. Whilst the evidence-base supporting or refuting individual risk factors in childhood disease may be limited, important differences between adult and childhood disease exist [207]and paediatric specific models that incorporate paediatric specific risk factors are needed. Part of the work described in this chapter was published in *JAMA cardiology* in 2020[216].

6.2 Aim

The aims of this chapter were;

1) To perform the first validation study of the HCM-Risk SCD model in childhood HCM
2) To develop and validate the first risk prediction model for SCD in childhood HCM

6.3 Methods

Study population

The study population was identified from the International Paediatric Hypertrophy Consortium as described in Chapter 2. Patients were excluded from model development if they had a history of prior VF or sustained VT (n=10) or less than 1 month follow up (n=29). The final model development cohort consisted of 1024 patients.

Patient assessment, data collection and clinical outcomes have been previously described in chapter 2.

The model development process is summarised in Figure 17 below

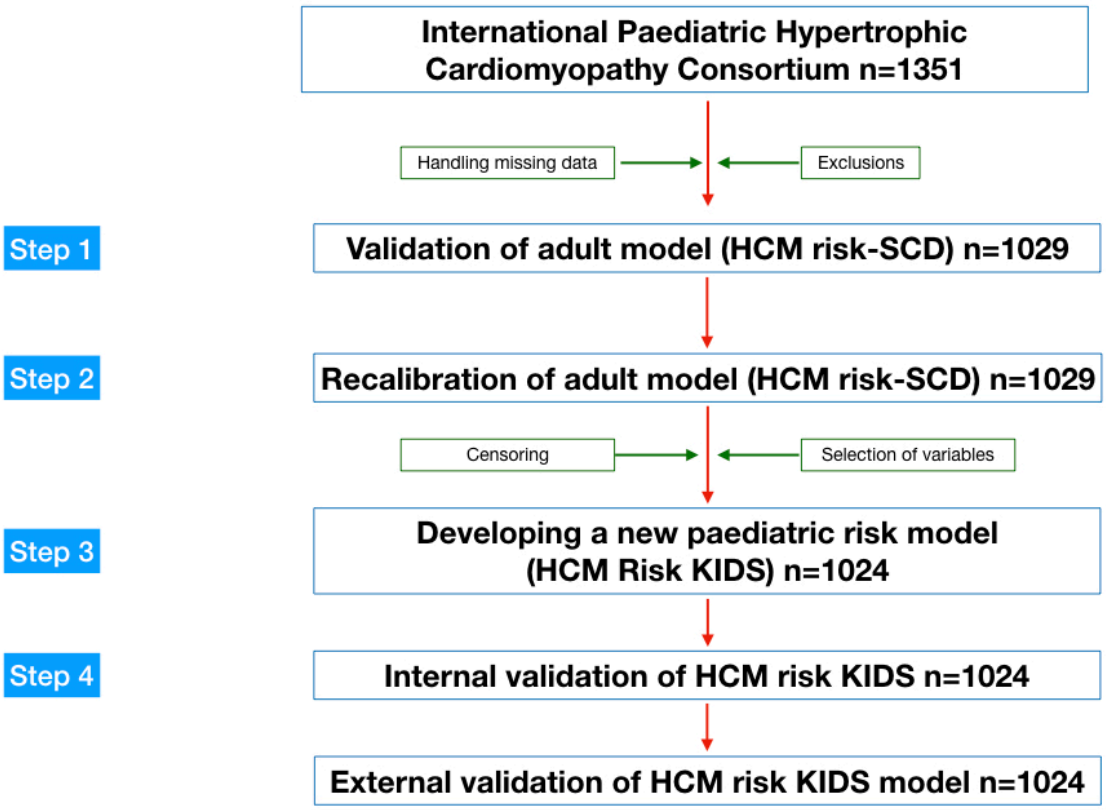


Figure 17 Model development process
*Five additional patients were excluded from the development of the new paediatric risk model owing to more than 50% missing data for paediatric variables.

General statistical methods

General statistical methods as described in Chapter 2.

I censored the follow up of patients at age 22 years as the aim of the model was to predict 5-year risk in childhood disease (≤ 16 years). The follow up time for all patients was therefore calculated from the date of first evaluation to the date of reaching the study end-point, death from another cause, or the date of most recent evaluation prior to the end of study period (December 2017 or reaching age 22 years).

Handling of missing data

Patients with more than 50% of missing predictors were excluded from model development. I compared the characteristics of those with complete and missing data and used logistic regression to identify predictors of missingness. Data was assumed to be missing at random (MAR) and values for the missing predictors were imputed using multiple imputation (MI) techniques based on chained equations[217, 218] for normally distributed variables, and predictive mean matching (k=5) for non-normally distributed variables. All predictors of missingness, the outcome, predictor variables and estimate of the cumulative hazard function were included in the MI model. A total of 49 imputed data sets were generated and estimates obtained from the imputed datasets were combined using Rubin's rule[219]. MI was performed in collaboration with Professor Rumana Omar and Dr Ting Ding, department of statistical sciences, University College London.

6.3.1 Validation of adult SCD model (HCM-Risk SCD[82]) in paediatric HCM

I performed the first validation of the adult model (HCM-Risk SCD[82]) in childhood disease in collaboration with Professor Rumana Omar and Dr Ting Ding, department of statistical sciences, University College London.

The 5-year risk of SCD was calculated using the HCM-Risk SCD formula [82]

$$Probability\ of\ SCD\ at\ 5\ years = 1 - 0.998^{\exp(prognostic\ index)}$$

Where PI is the prognostic index = 0.15939858 MWT - 0.00294271*MWT^2 + 0.0259082*LAd + 0.00446131* LVOTgmax + 0.4583082* FHSCD + 0.82639195*NSVT + 0.71650361* Unexplained syncope - 0.01799934*Age*

Patients were grouped into 3 categories of 5-year SCD risk as proposed by 2014 ESC guidelines [4]; < 4% ICD not generally indicated, 4-6% ICD may be considered and ≥ 6% ICD should be considered.

We assessed the discriminative ability of the model to distinguish between individuals at high and low risk of SCD events by calculating Uno's C-index (C-Uno)[220, 221]. A value of 1 indicates perfect discrimination, whilst a value of 0.5 indicates no discrimination. The calibration slope was calculated to assess the degree of agreement between observed and predicted hazards of SCD where a value close to 1 suggests good overall agreement [222]. I compared observed and predicted risk of SCD at 5 years by risk groups (0-<2%, 2-<4%, 4-<6%, ≥6%) in one imputed data graphically.

6.3.2 Recalibration of HCM-Risk SCD in paediatric HCM

I re-calibrated the adult HCM-Risk SCD model in a childhood cohort in collaboration with Professor Rumana Omar and Dr Ting Ding, department of statistical sciences, University College London. We re-calculated the model co-efficients for the adult predictor variables (Age (yrs), unexplained syncope, NSVT, LA diameter (mm), MWT

(mm), MWT^2 (mm), LVOT gradient (mmHg) and family history of SCD) in our paediatric population using a Cox proportional hazards regression model. Uno's C-index and the calibration slope were used to assess the discriminatory ability and degree of agreement of the recalibrated model as described in section 7.3.4 above.

6.3.3 Development of a paediatric specific risk model

Selection of predictor variables

I pre-selected candidate clinical risk factors for inclusion as predictor variables based on the results of my systematic review of the literature performed in 2015 as described in chapter 4 [207] and summarised in Table 18 below. Selection of predictor variables was not limited to studies with multivariable survival analyses due to the limited evidence base in childhood HCM. Clinical risk factors were selected as predictor variables if they had been examined in more than 2 published studies and independently associated with SCD in 2 or more univariable or multivariable survival analyses. I identified five pre-specified predictors meeting selection criteria; unexplained syncope, NSVT on ambulatory ECG recordings, LVMWT Z score, left atrial Z score and maximal LVOT gradient. The definition and coding of candidate predictors is shown in Table 19 below.

Candidate predictor	Total no. of studies using UV/MV survival analysis with CVD as an end-point	Total no. of studies using UV/MV survival analysis with SCD as an end-point	No. of studies showing significant independent association with CVD UV survival analysis	No. of studies showing significant independent association with SCD UV survival analysis	No. of studies showing significant independent association with CVD in MV survival analysis	No. of studies showing significant independent association with SCD in MV survival analysis
Age	11[15, 16, 40, 41, 43, 56, 59, 94, 159, 166, 169]	6 [41, 43, 59, 166, 169, 223]	3[43, 94, 159]	2 [43, 159]	0	0
Gender	5 [40, 56, 94, 157, 169]	2 [157, 169]	1[40]	0	1[40]	0
NYHA/Ross	4[15, 40, 41, 166, 213]	2 [41, 94]	2 [41, 213]	1 [41]	1 [40]	0
Unexplained syncope	7 [41, 57, 59, 80, 157, 159, 213]	6 [41, 59, 80, 157, 159, 213]	3 [80, 159, 213]	3 [80, 159, 213]	1 [213]	1 [213]
Family history of SCD	7 [40, 41, 56, 57, 59, 157, 159]	4 [41, 59, 157, 159]	0	0	1[157]	1[157]
Non-sustained VT on ambulatory ECG	6 [41, 56, 57, 59, 158, 159]	4 [41, 59, 158, 159]	2 [59, 158]	2 [59, 158]	0	0
Left ventricular hypertrophy+	13 [15, 40, 41, 56, 57, 59, 94, 157-159, 166, 169, 211]	9 [41, 59, 94, 157-159, 166, 169, 211]	9 [15, 40, 56, 57, 94, 158, 159, 169, 211]	6 [41, 94, 158, 159, 169, 211]	2 [56, 94]	1 [94]
Left atrial diameter	3 [41, 166, 211]	3 [41, 166, 211]	1 [41]	1 [41]	1[166]	1[166]
Left ventricular outflow tract obstruction*	6 [15, 41, 56-59]	2 [41, 59]	1 [56]	1 [41]	0	0
ABPRE	4 [41, 57, 157, 159]	3 [41, 157, 159]	1 [57]	0	0	0

Table 18 Summary of candidate predictors following systematic review of literature

The following search strategy was used to identify studies: MEDLINE search: MeSH terms “((hypertrophic cardiomyopathy) AND (death OR sudden death OR cardiac death OR outcome OR prognosis OR risk factors) AND (children OR childhood OR young OR paediatric)). Search was limited to: original articles written in English; patients aged < 18 years; published 1963 to December 2015. Initial search strategy was supplemented with manual searches

+ Measure of left ventricular hypertrophy varied between studies: extreme left ventricular hypertrophy (maximal wall thickness >30mm/Z score >6)[41, 56, 57, 157, 158, 211]; interventricular septal wall thickness (IVST) [40, 41, 56, 59, 94, 159, 166, 169, 211]; left ventricular posterior wall thickness (LVLVPWT) [15, 40, 56, 169, 211]; LVLVPWT:LV cavity [41, 94]

*Measure of LVOT obstruction varied between studies: LVOT gradient, mmHg [41], peak LVOT obstruction gradient >20mmHg [56], peak LVOT obstruction gradient >16mmHg [57]

Candidate predictor variable	Definition	Coding
NYHA/Ross functional class	New York Heart Association functional classification[224]/Modified Ross heart failure classification for children [190] at baseline evaluation	Binary (NYHA/Ross 1 = 0, NYHA/Ross ≥2 = 1)
Unexplained syncope	Defined as a transient loss of consciousness with no identifiable cause at or prior to first evaluation [16, 41, 82, 159].	Binary (No = 0, Yes = 1)
Family history of SCD	History of SCD in 1 or more first degree relative under 40 years of age or SCD in a first degree relative with confirmed HCM at any age[41, 59, 82]	Binary (No = 0, Yes = 1)
Non-sustained ventricular tachycardia (NSVT)	≥3 consecutive ventricular beats at a rate of ≥120 beats/minute lasting <30 seconds on ambulatory ECG monitoring (minimum duration 24 hours) at or prior to first evaluation [111, 158].	Binary (No = 0, Yes = 1)
Maximal wall thickness Z score	Defined as the number of standard deviations away from the population mean [197]the 2D measurement of maximal MWT (mm)* is at baseline evaluation	Continuous (Z score)
Left atrial diameter Z score	Defined as the number of standard deviations away from the population mean[196] the 2D measurement of maximal left atrial diameter (mm)^ is at baseline evaluation.	Continuous (Z score)
Maximal left ventricular outflow tract gradient	The maximum LV outflow gradient at rest or with Valsalva provocation using continuous wave Doppler from the apical 3- or 5-chamber views.+ [225]	Continuous (mmHg)

Table 19 Definition of pre-selected candidate predictor variables

*Maximal wall thickness is the greatest thickness as measured by 2D echocardiography in the parasternal short-axis views of LV in four places at the level of the mitral valve and papillary muscles (anterior and posterior septum, lateral and posterior wall) and in two places at apical level (anterior and posterior septum)[4].^Left atrial diameter is determined by M mode or 2D echocardiography in the parasternal long axis plane+ Peak outflow tract gradient is determined using the modified Bernoulli equation: Gradient = $4V^2$ (where V is the peak aortic outflow velocity)

Model development

I developed the paediatric model in collaboration with Professor Rumana Omar and Dr Ting Ding, department of statistical sciences, University College London.

All continuous predictors were centred around their mean value to avoid extreme values of the 5-year baseline survivor function and to reduce the correlation between linear and quadratic interactive terms. For each continuous variable, Cox proportional hazard regression models were used to test the assumption of linearity with the outcome. Where non-linearity was found, quadratic terms were included. The final model was therefore developed using a Cox proportional hazards regression model including 5 pre-specified predictors (Table 19 above) and quadratic terms for MWT Z score. It has previously been estimated that a minimum of 10 SCD or equivalent events are required per model coefficient to allow regression coefficients to be estimated with adequate precision[226]. Additional events are required if non-linear terms for continuous predictors are used. The model was developed using the entire follow up data and all events that occurred during follow up (eg 1024 patients and 89 events), with enough events to estimate regression coefficients for the 6 predictors selected. The proportional hazards assumption was investigated using Schoenfeld residuals[227]. As the number of patients and events in each centre was small, all regression models were fitted using robust standard errors to account for clustering by centre[228].

The probability of SCD at *t* years for an individual patient can be calculated using the following equation.

$$Probability\ of\ SCD\ at\ 5\ years = 1 - S_0(t)^{\exp(prognostic\ index)}$$

Where $S_0\,(t)$ is the average survival probability at time *t* and the *prognostic index* is the sum of the products of the predictors and their coefficients.

Internal validation of the paediatric model

We internally validated the performance of the developed model using bootstrapping as it allows all aspects of model development to be validated and is therefore the most efficient validation procedure[229]. 200 bootstrap samples were generated from each imputed data set (n=49) and estimates were combined using Rubin's rule[219]. As the aim of the model was to predict SCD risk at 5 years, patient follow up was censored at 5 years from the date of baseline evaluation for validation. Uno's C-index and the calibration slope were used to assess model discrimination and degree of agreement as described in section 7.3.1 above. The C-index and calibration results presented are an average of the bootstrap samples. The comparison between observed and predicted risk of SCD at 5 years by risk groups (0-<2%, 2-<4%, 4-<6%, ≥6%) in one imputed data set is shown graphically

6.4 Results

Baseline clinical characteristics

The baseline characteristics of the cohort are described in Chapter 5 and Table 20 below.

		Divided by era				
	Whole cohort	**Pre-1990 (n=29)**	**1990-1999 (n=129)**	**2000-2009 (n=416)**	**2010 onwards (n=450)**	**P-value**
Male gender	699 (68.3%)	18 (62.1%)	79 (61.2%)	289 (69.5%)	313 (69.6%)	0.253
Age (yrs)	11 (7,14)	8 (5,13)	11 (5,13)	11 (7,14)	11 (7,14)	0.11
Family history of HCM	534 (53.1%)	11 (38%)	70 (54.7%)	220 (53.5%)	233 (53.2%)	0.417
Family history SCD	130 (12.8%)	4 (13.8%)	23 (18%)	54 (13%)	49 (11%)	0.215
Unexplained syncope	102 (9.9%)	5 (17.2%)	18 (14%)	44 (10.6%)	35 (7.8%)	0.086
NYHA/Ross >1	223 (22.2%)	11 (40.3%)	36 (28.4%)	95 (23.6%)	81 (18.1%)	0.006
NSVT	55 (6.4%)	11 (40.7%)	6 (5.3%)	28 (7.9%)	10 (2.8%)	<0.001
MWT (mm)	17.1 (+/-7.4)	22 (+/-8.9)	19.4 (+/-8.1)	17.4 (+/-7.5)	16 (+/-6.7)	0.013
MWT z-score	11.1 (+/-7.1)	16.2 (+/-8.5)	14.3 (+/-8.0)	11.4 (+/-7.3)	9.8 (+/-6.4)	0.003
LA diameter (mm)	33.4 (+/-8.5)	33.6 (+/-6.1)	34.1 (+/-8.2)	35.8 (+/-8.8)	31.3 (+/-7.8)	0.133
LA z-score	1.9 (+/-2.3)	2.8 (+/-2.5)	2.4 (+/-2.1)	2.6 (+/-2.6)	1.3 (+/-1.9)	<0.001
LVOT (mmHg)	9 (6, 22)	10 (5,10)	12 (6, 46)	10 (6, 23)	8 (5, 16)	0.010

Table 20 Baseline demographics of development cohort and clinical characteristics by era of presentation

Clinical outcomes and survival

Over a total follow up period of 5984 patient years (median 5.4 yrs, IQR 2.6-8,3); 77 (7.7%) underwent a myectomy, 43 (4.2%) required a PPM, and 267 (26.1%) an ICD for primary (n=244, 91.4%) or secondary (n=23, 8.6%) prevention of SCD. Fifty-three (5.2%) patients died and 21 (2.1%) underwent cardiac transplantation. Cause of mortality was SCD (n=30, 56.6%), heart failure (n=9, 17.0%), other cardiovascular (n=3, 5.7%) or non-cardiovascular (n=2, 3.8%). In 3 patients (5.7%), cause of death was unknown. Overall annual mortality rate was 0.89/100 patient years (95% CI 0.68-1.16).

Eighty-nine patients reached the SCD or equivalent end-point by the end of the study; SCD n=39, 43.8%; resuscitated cardiac arrest n=16, 18.0%; appropriate ICD discharge n=24, 27.0%; and sustained VT n=10, 11.2%. Age at time of event is shown in Figure 18 below. Overall annual SCD event rate was 1.49/100 patient years (95% CI 1.15 – 1.92).

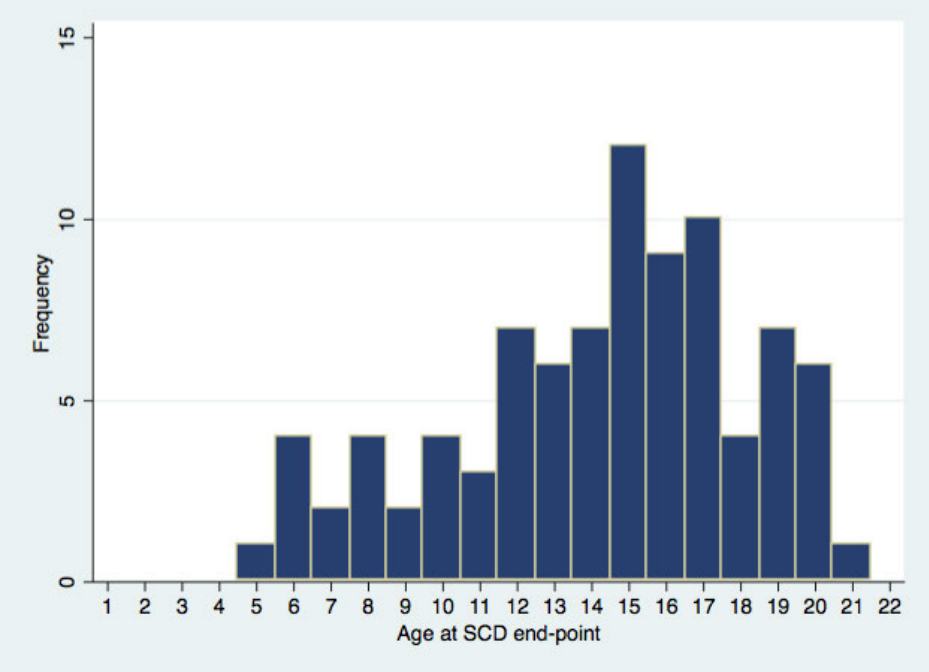


Figure 18 Bar chart showing age at the time of arrhythmic event

The clinical characteristics of those with and without the SCD end point are described in Table 21 below. Patients with a SCD event were more likely to have heart failure symptoms (HR 1.70, 95% CI 1.08-2.65, p value 0.02), unexplained syncope (HR 2.06, 95% CI 1.20-3.54, p=0.009), NSVT on ambulatory ECG monitoring (HR 1.93, 95% CI 1.03-3.61, p value 0.04), severe hypertrophy (HR 1.05 for each Z score increase 95% CI 1.02-1.07, p value <0.001) and left atrial dilatation (HR 1.19 for each Z score increase 95% CI 1.08-1.30, p value <0.001).

	Whole cohort n=1029	Patients with SCD end-points (n=89)	Patients without SCD end-points (n=938)	Hazard ratio	95% CI	P value
Age (median IQR)	11 (7,14)	10 (6, 13)	11 (7, 14)	1.051	0.997-1.107	0.06
Male	699 (68.3%)	24 (27%)	301 (32.2%)	0.734	0.495-1.172	0.20
NYHA >1	223 (22.2%)	28 (31.8%)	195 (21.2%)	1.689	1.078-2.645	0.02
Family history SCD	130 (12.8%)	12 (13.5%)	118 (12.7%)	1.008	0.548-1.851	0.98
Family history HCM	534 (53.1%)	42 (48.3%)	492 (53.5%)	0.833	0.547-1.270	0.39
Unexplained syncope	102 (9.9%)	16 (18%)	86 9.2%)	2.059	1.198-3.540	0.009
NSVT	55 (6.4%)	12 (16.4%)	43 (5.5%)	1.932	1.034-3.610	0.04
MWT (mm), mean (+/-SD)	17.1 (+/- 7.4)	20 (+/- 7.5)	16.9 (+/-7.3)	1.047	1.022-1.073	<0.001
Z score MWT, mean (+/-SD)	11.1 (+/- 7.1)	15 (+/- 7.5)	10.7 (+/- 7)	1.051	1.024-1.079	<0.001
MWT ≥ 30mm	81 (7.9%)	25 (31.7%)	164 (20.7%)	2.351	1.325-4.176	0.004
LA diameter (mm), mean (+/-SD)	33.4 (8.5%)	36.5 (+/- 9.3)	33.0 (+/- 8.3)	1.047	1.019-1.075	0.001
Z score LA diameter, mean (+/-SD)	1.9 (+/- 2.3)	3.2 (+/- 2.6)	1.8 (+/- 2.2)	1.185	1.081-1.298	<0.001
LVOTg max (mmHg)	9 (6, 22)	12 (6-36)	9 (5.9-20)	1.000	0.993-1.008	0.83
LVOT ≥30mmHg	189 (18.4%)	25 (31.7%)	164 (20.7%)	1.476	0.916-2.380	0.11

Table 21 Clinical characteristics of patients with and without sudden cardiac death endpoint and univariable Cox regression models

NYHA = New York Heart Association, SCD = sudden cardiac death, MWT=maximal wall thickness, LA = left atrium, LVOTg max = Maximal left ventricular outflow tract gradient. NSVT = non-sustained ventricular tachycardia, ECG = electrocardiograph

Missing data

Data was missing in 1, 2 or 3 predictor variables in 252 (24.6%), 176 (17.2%) and 69 (6.7%) patients respectively. Predictor variables with the highest proportion of missing data were LA Z score diameter (n=349, 34.1%), NSVT on ambulatory ECG (n=168, 16.4%) and maximal LVOT gradient (n=153, 14.9%). Predictors of missingness were the absence of heart failure symptoms (NYHA/Ross classification <2) and degree of hypertrophy (Z score MWT). Table 22 below describes clinical predictor variables before and after missing imputation

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		Missing data (n, %)	Original dataset (n=1024)	MI dataset (n=49388)
Age (mean, 95% CI)		NA	10.4 (10.2-10.7)	10.4 (10.2-10.7)
Male gender		NA	699 (68.3%)	68.3%
Family history HCM		18 (1.7%)	534 (53.1%)	53.1%
Family history SCD		4 (0.4%)	130 (12.8%)	12.8%
Unexplained Syncope		1 (0.1%)	102 (9.9%)	9.5%
NYHA/Ross	**1**	18 (1.7%)	783 (77.8%)	78. 3%
	2+		223 (22.2%)	21.7%
NSVT on ambulatory ECG		168 (16.4%)	55 (6.4%)	6.2%
MWT (mm) [mean, 95% CI]		32 (3.1%)	17.1 (16.7-17.6)	17.9 (17.3-18.4)
Z score MWT [mean, 95% CI]		118 (11.5%)	11.1 (10.6 – 11.6)	11.5 (10.9-12.0)
LA diameter (mm) [mean, 95% CI]		313 (30.6%)	33.4 (32.8 – 34.0)	33.8 (33.1 - 34.4)
Z score LA diameter [mean, 95% CI]		349 (34.1%)	1.92 (1.7 – 2.1)	1.96 (1.8 - 2.1)
LVOTg max [median, IQR]		153 (14.9%)	21.8 (19.9-23.7)	21.3 (19.1 - 23.4)

Table 22 Summary of missing data and comparison of distribution of predictor variables before and after multiple imputation

Validation of adult HCM-Risk SCD model in paediatric HCM

I assessed the performance of the adult model in 1029 patients with 89 events. The model had a moderate ability to discriminate between individuals at high and low risk (C-index 0.67 (95% CI 0.65-0.69)) but risk predictions were not accurate (calibration slope 0.79 (95% CI 0.43-1.15)). The risk of SCD appeared to be underestimated for all risk groups (Figure 19 below).

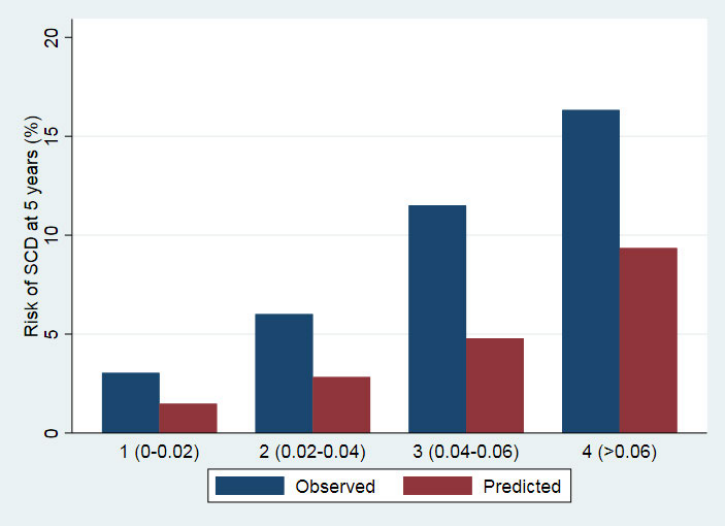


Figure 19 Comparison of observed and predicted risk by clinical risk group for external validation of adult HCM-Risk SCD model.
Vertical bars represent observed (blue) and model-based predicted (red) probability of SCD by 5 years using one imputed data set.

Recalibration of adult HCM-Risk SCD model in paediatric HCM

As the published adult HCM-Risk SCD model did not accurately predict risk in the paediatric population, I recalibrated the model predictor coefficients using the paediatric dataset. Following recalibration, the risk of SCD at 5 years for can be calculated from the following equation.

$$Probability\ of\ SCD\ at\ 5\ years = 1 - 0.9244^{\exp(prognostic\ index)}$$

where Prognostic Index = 0.1548734(MWT-17.13) - 0.0026427*(MWT^2-348.04) + 0.0428425*(LA-33.39) + 0.3746369*Unexplained Syncope + 0.1738467*NSVT - 0.0042133*(LVOT-21.8) - 0.0208741*Age – 0.1556381*Family history of SCD.*

The estimates of the hazard ratios for the recalibrated model are described in Table 23 below. The performance of the recalibrated adult model was assessed and showed improved agreement between predicted and observed risk (calibration slope 0.99, 95% CI 0.52-1.46), but no change in the discriminatory ability (C-index 0.66, 95% CI 0.64 – 0.69)

Predictor variable	Hazard ratio (95% CI)	P-value
Age (years)	0.98 (0.92-1.04)	0.513
Unexplained syncope	1.45 (0.82-2.56)	0.195
NSVT	1.19 (0.62-2.30)	0.604
LA diameter (mm)	1.04 (1.01-1.08)	0.011
MWT (mm)	1.17 (1.01-1.35)	0.034
MWT^2 (mm)	0.997 (0.994-1.001)	0.100
LVOT gradient (mmHg)	0.996 (0.988-1.004)	0.301
Family history SCD	0.86 (0.46-1.60)	0.625

Table 23 Univariable Cox proportional hazards regression model of recalibrated HCM-Risk SCD model in paediatric HCM

Development of a paediatric specific risk model (HCM Risk-Kids)

As neither the original or recalibrated adult HCM-Risk SCD model could accurately predict risk in the paediatric population, a paediatric risk model was developed using the pre-specified predictor variables described in Table 19 above (unexplained syncope, NSVT, Z score LA diameter, Z score MWT thickness and LVOT gradient). The estimates of the hazard ratio derived from the model are shown in Table 24 below.

	HCM risk-Kids SCD risk prediction model		**Sensitivity analysis: model with predictor of missingness**	
Predictor variable	**Hazard ratio (95% CI)**	**P-value**	**Hazard ratio (95% CI)**	**P-value**
NSVT	1.20 (0.62-2.34)	0.582	1.16 (0.59-2.29)	0.663
LA diameter Z score	1.14 (1.03-1.26)	0.012	1.13 (1.02-1.26)	0.017
MWT Z score	1.24 (1.10-1.41)	0.001	1.24 (1.10-1.41)	0.001
MWT Z score^2	0.995 (0.992-0.999)	0.011	0.995 (0.992-0.999)	0.011
LVOT gradient	0.993 (0.985-1.001)	0.121	0.993 (0.985-1.002)	0.114
Unexplained syncope	1.54 (0.88-2.69)	0.133	1.52 (0.86-2.66)	0.147
NYHA			1.15 (0.70-1.88)	0.589
Uno's C statistic	0.69 (0.66-0.72)		0.69 (0.66-0.72)	
Calibration slope	0.98 (0.59-1.38)		0.96 (0.56-1.36)	

Table 24 Paediatric sudden cardiac death risk prediction model and sensitivity analyses for predictor of missingness

The risk of SCD at 5 years for SCD in childhood HCM can be calculated from the following equation (HCM risk KIDS)

$$Probability\ of\ SCD\ at\ 5\ years = 1 - 0.9494^{\exp(prognostic\ index)}$$

where Prognostic Index = 0.2171364(MWT z score-11.09) - 0.0047562*(MWT Z score^2-174.12) + 0.130365*(LA diameter Z score-1.92) + 0.429624*Unexplained Syncope + 0.1861694*NSVT - 0.0065555*(maximal LVOT gradient-21.8)*

We performed a sensitivity analysis including predictors of missingness (NYHA). However, this did not change the hazard ratio estimates for individual predictors or the performance of the model (Table 24 above).

Internal validation of HCM risk KIDS model

We assessed the performance of the newly developed paediatric model using 1024 patients with 58 events (follow-up was censored at 5 years). The C-index was 0.69 (95% CI 0.66-0.72) and the calibration slope was 0.98 (95% CI 0.66-0.72). A comparison between observed and predicted risk is shown graphically in Figure 20 below.

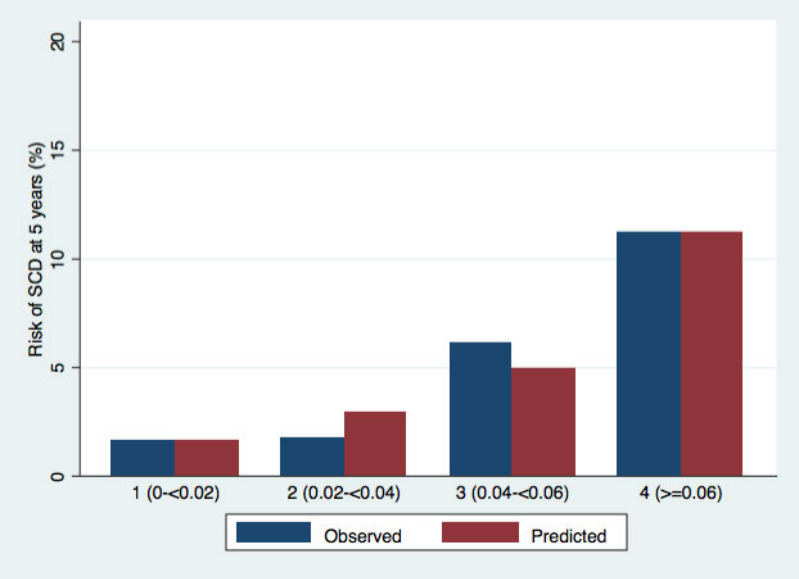


Figure 20 Comparison of observed and predicted risk by clinical risk group for the paediatric sudden cardiac death risk model

Vertical bars represent observed (blue) and model-based predicted (red) probability of SCD by 5 years using one imputed data set

Clinical implications of the HCM risk-Kids model

I investigated the clinical implications of retrospectively applying the HCM risk-KIDS model in 527 patients who had complete data to allow me to estimate 5-year risk of SCD, of whom 34 met the SCD end point. The number of patients with an estimated 5-year risk of <4%, 4-6% and ≥6% was 178 (33.8%), 100 (19.0%) and 249 (47.2%) respectively. The SCD end-point was reached by 3 patients (1.7%) with a predicted risk of <4%, 5 patients (5.0%) with a predicted risk of 4-6% and 26 (10.4%) with a predicted risk of ≥6%. The cumulative probability of SCD by risk group is shown in the Kaplan Meier curves in Figure 21 below.

Using a 5-year estimated risk threshold of ≥6% would identify 26 of 34 SCD end points (76.5%). An ICD would be implanted in 223 of 493 patients (45.2%) not reaching the end point within 5 years. For every 10 ICDs implanted in patients with a risk of ≥6%, 1 patient may potentially be saved from SCD at 5 years. Using a 5-year estimated risk threshold of ≥4% would identify 31 of 34 SCD end-points (92.2%). An ICD would be implanted in 318 of 493 patients (64.5%) not reaching SCD end-points within 5 years. For every 12 ICDs implanted in patients with a risk of ≥4%, 1 patient may potentially be saved from SCD at 5 years.

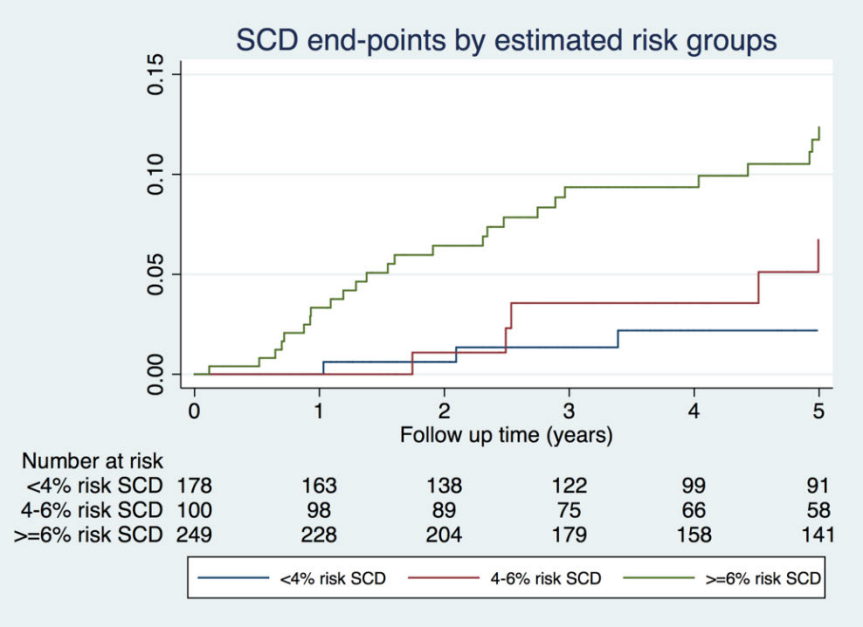


Figure 21 Kaplan-Meier curve showing cumulative probability of sudden cardiac death end points within 5 years of baseline evaluation by clinical risk groups calculated by the paediatric SCD risk model.
Patients with complete data for the calculation of 5-year SCD risk estimates (n = 527) were classified into 3 risk groups (<4%, 4%-<6%, and ≥ 6%)

6.4.1 Investigating the relationship of maximal wall thickness and estimated SCD risk

The baseline clinical characteristics by severity of left ventricular hypertrophy are described in Table 25 below. Those with higher MLVWT z scores were more likely to have heart failure symptoms (NYHA>1), LVOT obstruction, a dilated LA or NSVT. The estimated risk of SCD at 5 years had a non-linear, inverted U-shaped relationship with body surface area corrected MLVWT (Figure 22 below). Estimated risk plateaued at a MLVWT z score of 22.8; further increases were not associated with an additional increase in the estimated risk of SCD. This relationship was not altered by the presence or absence of other clinical risk factors.

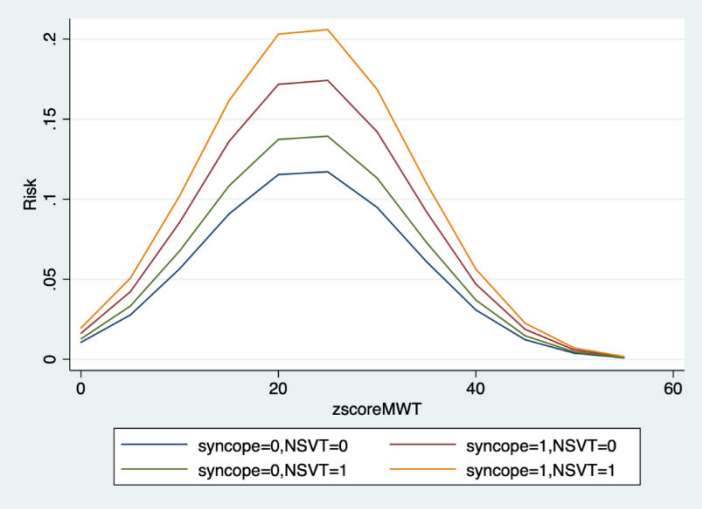


Figure 22 Relationship of estimated 5-year risk of sudden cardiac death to maximal wall thickness.
The risk of SCD at 5 years was calculated using HCM Risk-Kids with the following predictors kept constant to the cohort mean: maximal left ventricular outflow tract gradient, 21.79741 mm Hg; left atrial diameter Z score, 1.918261. The 4 curves represent the estimated risk with all possible combinations of non-sustained ventricular tachycardia (NSVT) and unexplained syncope. In all cases, the risk of SCD increases up to a point, and once a plateau is reached, the risk declines.

	MWT z score			P value
	<10 (n=506)	10-<20 (n=278)	≥20 (n=122)	
Male sex	348 (68.8%)	183 (65.8%)	88 (72%)	0.435
Age at baseline (y)	9.9 (+/-4.4)	10.3 (+/-4.4)	10.9 (+/-4.3)	0.096
B Blocker therapy	173 (34.2%)	130 (46.8%)	75 (61.5%)	<0.001
NYHA class >1 (n=974)	79 (15.8%)	79 (29.4%)	47 (39.2%)	< 0.001
Family history SCD	65 (12.9	33 (11.9%)	14 (14.9%)	0.661
NSVT (n=825)	16 (3.9%)	21 (9.2%)	13 (11.8%)	0.002
Unexplained syncope (n=991)	37 (7.3%)	39 (14.0%)	18 (14.8%)	0.003
Maximal LVOT gradient (mmHg) (n=858)	8 (5,14)	11 (6, 36)	20 (8,49)	<0.001
LVOT gradient ≥30mmHg	51 (11.8%)	72 (29.3%)	45 (39.8%)	<0.001
Z score LA diameter (mean +/- SD)	1.6 (+/-2.2)	2.3 (+/-2.4)	2.3 (+/-2.2)	<0.001
Septal myectomy during follow up	13 (2.6%)	28 (10.1%)	20 (16.4%)	< 0.001
MACE during follow up	27 (5.3%)	39 (14.0%)	23 (18.9%)	<0.001
incidence of MACE per 100 pt years (95% CI)	0.77 (0.50-1.19)	1.98 (1.40 – 2.79)	2.74 (1.79 – 4.20)	<0.001

Table 25 Clinical characteristics by degree of left ventricular hypertrophy

NYHA = New York Heart Association, SCD = Sudden Cardiac Death, NSVT = Non-sustained Ventricular Tachycardia, LVOT = Left Ventricular Outflow Tract, LA = Left atrial, MACE = Major Arrhythmic Cardiac Event, MWT = maximal wall thickness, pt=patient, y=years

6.4.2 Investigating the relationship of LVOT gradient and estimated SCD risk

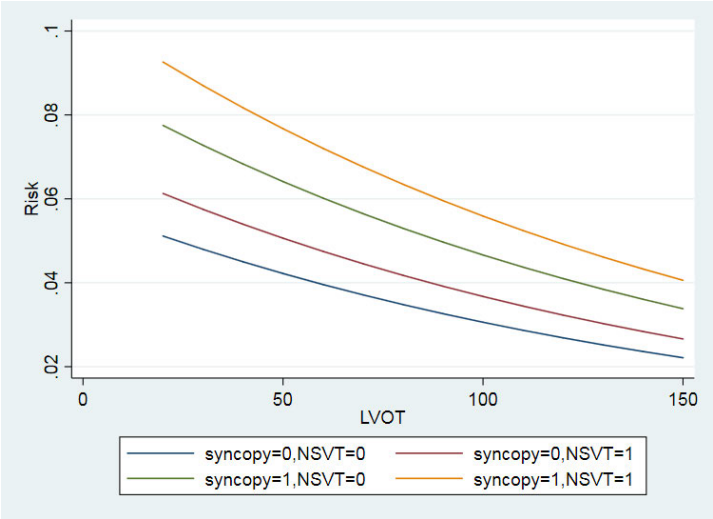
The baseline characteristics by LVOT gradient are described in Table 26 below. Those with obstructive disease were more likely to have symptoms of heart failure (NYHA>1), higher maximal wall thickness, larger LA diameter and have undergone a LV septal myectomy during follow up.

	<30mmHg (n=682)	**30-50mmHg (n=58)**	**50-100mmHg (n=98)**	**>=100mmHg (n=33)**	**P value**
Male gender	461 (67.6%)	39 (67.2%)	68 (69.4%)	24 (72.7%)	0.921
Age at baseline (mean, +/-SD)	10.5 (+/-4.3)	8.9 (+/-4.9)	9.1 (+/-4.7)	9.4 (+/-4.4)	0.0018
B Blocker therapy	288 (42.3%)	30 (51.7%)	50 (51.6%)	19 (57.6%)	0.085
NYHA>1	137 (20.5%)	17 (29.3%)	37 (37.8%)	14 (43.8%)	<0.001
FHx SCD	93 (13.7%)	8 (13.8%)	10 (10.4%)	1 (3.0%)	0.287
NSVT	39 (6.8%)	5 (10.4%)	7 (8.5%)	2 (8.7%)	0.771
Unexplained syncope	70 (10.3%)	9 (15.5%)	6 (6.1%)	4 (12.1%)	0.296
Z score MWT (mean, +/-SD)	10.3 (+/-6.6)	15.1 (+/-7.6)	15 (+/-8.1)	16.6 (+/-8.3)	<0.001
Z score LA (mean, +/-SD)	1.7 (+/-2.3)	2.8 (+/-2.4)	3 (+/-2.4)	3.4 (+/-2.5)	<0.001
Length of follow up	7.2 (+/-6.4)	6.1 (+/-5.4)	7.7 (+/-6.5)	7.8 (+/-5.1)	0.448
ICD implanted during follow up	164 (24.2%)	21 (36.3%)	30 (31.3%)	9 (27.3%)	0.127
Myectomy during follow up	20 (2.9%)	9 (15.5%)	29 (29.9%)	12 (36.4%)	<0.001
MACE event	54 (7.9%)	11 (18.9%)	13 (13.2%)	1 (3.0%)	0.009
MACE rate/ 100 pt years (95% CI)	1.40 (1.06-1.80)	3.5 (1.90-6.30)	2.08 (1.21-3.58)	0.42 (0.06-2.98)	0.219
All-cause mortality	54 (7.9%)	9 (15.5%)	13 (13.3%)	3 (9.1%)	0.106
Mortality rate /100 pt years (95% CI)	1.10 (0.84-1.43)	2.54 (1.32-4.88)	1.17 (1.00-2.97)	1.16 (0.37-3.60)	0.065

Table 26 Baseline clinical characteristics and outcomes of patients with and without left ventricular outflow tract obstruction

NYHA = New York Heart Association, FHx = family history, SCD = sudden cardiac death, NSVT = Non-Sustained Ventricular Tachycardia, MWT = maximal wall thickness, LA = left atrial, ICD = implantable cardioverter defibrillator, MACE= Major Arrhythmic Cardiac Event

Compared to those with non-obstructive disease, patients with mild, moderate or severe LVOT obstruction had a hazard of SCD of 1.75 (95% CI 0.89-3.44), 1.04 (95% 0.55-1.98), and 0.70 (0.36-1.35), respectively (global p value 0.204). Corresponding incidence rates are described in Table 26 above. An apparent inverse relationship existed between LVOT gradient and estimated risk (Figure 23 below).



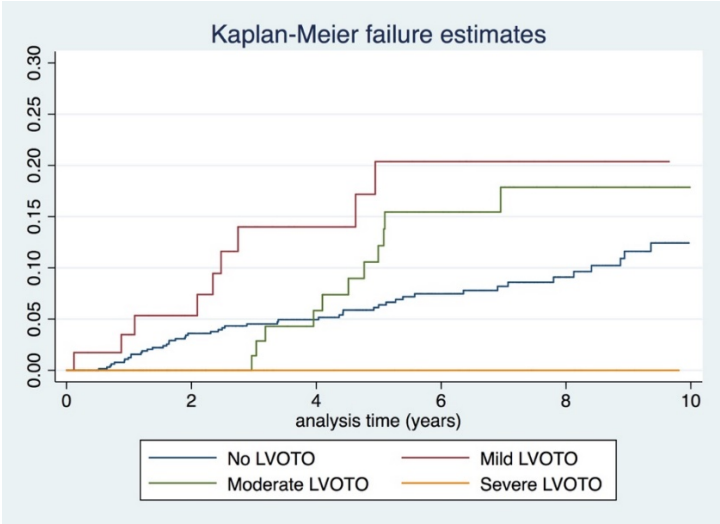


Figure 23 Relationship of left ventricular outflow tract gradient and SCD
A) Kaplan Meier curve showing the incidence of SCD events by degree of left ventricular outflow tract obstruction. LVOT obstruction defined by maximal LVOT gradient (mmHg); No LVOT obstruction < 30mmHg, Mild LVOT obstruction 30-50mmHg, Moderate LVOT obstruction 50-100mmHg, Severe LVOT obstruction >100mmHg B) Relationship of the estimated SCD risk and maximal LVOT gradient. The risk of SCD at 5 years was calculated with the following predictors kept constant to the cohort mean: z score left atrial diameter 1.92, z score MWT 11.09. The 4 curves represent the estimated risk of all possible combinations of non-sustained VT (NSVT) and syncope.

Including possible confounders (myectomy or B blocker therapy) in the HCM Risk-Kids model reduced the significance of LVOT gradient as a variable but did not change the estimated hazard ratio (Table 27 below).

	Including myectomy in HCM risk kids model			**Including B blocker therapy in HCM risk kids model**			**Original model**		
	Hazard ratio	**P value**	**95% CI**	**Hazard ratio**	**P value**	**95% CI**	**Hazard ratio**	**P value**	**95% CI**
NSVT	1.24	0.535	0.63-2.42	1.43	0.319	0.71-2.88	1.2	0.582	0.62-2.33
Unexplained syncope	1.42	0.253	0.78-2.60	1.53	0.174	0.83-2.81	1.54	0.133	0.88-2.69
Z score LA	1.16	0.007	1.04-1.29	1.15	0.008	1.04-1.28	1.14	0.012	1.03-1.26
MWT z score	1.28	<0.001	1.12-1.48	1.25	0.002	1.08-1.44	1.24	0.001	1.1-1.41
MWT z score^2	0.994	0.007	0.990-0.998	0.995	0.029	0.991-1.00	0.995	0.001	0.992-0.999
LVOT	0.997	0.453	0.877-1.01	0.994	0.190	0.99-1.00	0.993	0.121	0.99-1.00
Myectomy	0.439	0.057	0.19-1.02						
B Blocker				0.70	0.147	0.43-1.14			

Table 27 HCM Risk Kids prediction model sensitivity analysis adjusted for left ventricular outflow tract gradient confounders

6.5 Discussion and limitations

In this part of my thesis I have developed the first validated paediatric risk model for SCD in childhood HCM and shown it to have an improved ability to discriminate between high and low risk patients compared to the current paediatric guidelines.

Comparing risk stratification in adult and paediatric populations

The ability to calculate individualised estimates of risk for adult HCM patients using the HCM risk-SCD [82]model arguably represents a significant advance for both clinicians and patients, allowing meaningful discussions around the risks and benefits of prophylactic ICD implantation to take place. In contrast, risk stratification approaches for SCD in childhood have remained largely unchanged for 2 decades relying on the assessment of a small number of risk factors to guide ICD implantation, an approach that I have shown in chapter 5 to have limited discriminatory power[215]. Children with HCM are known to have a higher risk of SCD compared to adults and are additionally at increased risk of complications following ICD implantation[158, 182]. The importance of correctly identifying those most likely to benefit from preventative therapy is therefore paramount. The results of the first external validation of the adult HCM-risk SCD model in a childhood cohort show that although the model's discriminatory ability is superior to current paediatric guidelines (C statistic 0.67 vs 0.62), the estimates of risk generated are not accurate, with risk underestimated for all risk groups. It is therefore not appropriate to use the original HCM-risk SCD proposed by O'Mahony et al to risk stratify paediatric patients. The calibration of the model's estimates was improved following recalibration of the predictor variable coefficients, yet this did not significantly improve the model's discriminatory power. This is likely explained by the inclusion of

variables in the model with insufficient evidence in childhood disease including family history of sudden cardiac death and age.

A new paediatric model for risk stratification in HCM

The new paediatric model that I have developed uses 5 clinical variables to estimate risk (unexplained syncope, NSVT, LA diameter, MLVWT and LVOT gradient) and shows better discrimination between high- and low-risk patients than both the current paediatric guidelines and recalibrated adult risk model, with good calibration between the expected and observed risk. The performance is similar to that reported in adult cohorts for the adult model (C-Index 0.69 vs 0.70)[185]. Unexplained syncope, degree of hypertrophy, LA diameter, and NSVT showed the strongest association with the study outcome, although this finding was not significant at the 15% level for NSVT. Interestingly, LVOT gradient appears to be inversely associated with the risk of SCD in this population, a finding that is discussed in further detail below. Age was not included as a predictor variable as, outside of infancy, its role in prognosis remains unclear [16, 230]. However, the effect of age may have been mitigated by accounting for somatic growth using body surface area corrected, rather than absolute 2D, echocardiographic measurements. Family history of SCD was not chosen as a predictor variable as it was only associated with SCD in one out of seven published studies[207] as described in chapter 4. The lack of evidence for a family history of SCD in the current literature could be explained by a higher prevalence of de novo mutations in childhood, incomplete reporting of family history or failure to adjust for family linkage.

My complete case analysis suggests that, using a threshold of ≥6, identifies the majority of patients at risk of a SCD event during the follow-up period. The

identification of patients at risk of SCD was at the expense of ICD implantation in 45% of patients not yet reaching the end point during follow-up. However, as previous studies have demonstrated variable latency between ICD implantation and first appropriate therapy [158, 182], these young patients may yet benefit from the decision to implant a device. Importantly, the threshold of 6% for this analysis was extrapolated from the adult guidelines and may not be appropriate for childhood disease. Consensus opinion of experts will be required to determine whether absolute thresholds for ICD recommendations are needed and, if so, where those thresholds should be set. Whilst this model represents a significant improvement on current risk stratification methods, it remains imperfect meaning that additional risk factors may be important to refine the model. In addition, because childhood is a time of significant somatic growth, the phenotype of a patient may evolve rapidly with a resulting change in the arrhythmic risk profile. Future studies exploring the changing role of individual clinical risk factors during childhood and use of serial clinical investigations in predicting risk would be valuable.

The relationship between continuous risk factors and estimated risk

In adult patients with HCM, the severity of left ventricular hypertrophy has been shown to have an inverted U-shaped relationship with the risk of SCD [115]. An association between LVH and the risk of SCD has frequently been reported in childhood disease and current guidelines recommend using a threshold for maximal wall thickness of ≥30mm or z score ≥6 [4, 5] when assessing risk. The evidence for this particular threshold is limited, yet the implication is that risk increases in a linear fashion with increasing hypertrophy. In this large childhood HCM cohort, I show that, unsurprisingly, severe LVH was associated with other phenotypic features of severe

disease, including LVOT obstruction, left atrial dilatation and NSVT. However, an inverted U-shaped relationship between the degree of hypertrophy and estimated risk of SCD at 5 years was seen, showing that, beyond a threshold, further increases in hypertrophy were not associated with additional risk. This finding is in agreement with other recent reports from independent large paediatric cohorts[55, 214] and suggests that, whilst MLVWT is important for risk stratification, it should not be used either as a binary variable or in isolation to guide ICD implantation decisions in children with HCM as is recommended in current guidelines [5].

Obstructive disease (LVOT gradient ≥ 30mHg) is a well-established risk factor for SCD in adult HCM patients. However, as I discussed in Chapter 4, previous paediatric studies have reported conflicting results on its role in risk stratification for childhood disease. In this large cohort of children with non-syndromic HCM I unexpectedly found an apparent inverse relationship between LVOT gradient and predicted risk, meaning that those with the most severe obstruction did not appear to have a significantly higher risk of arrhythmic events. LVOT obstruction is generally defined as a peak instantaneous Doppler gradient ≥30mmHg [4] but a LVOT gradient only becomes haemodynamically significant above 50mmHg [51]. Applying these two thresholds to the current dataset confirms an increased proportion of patients with LVOT obstruction and SCD events (13.2% vs 7.9% and 10.7% vs 8.8%), but this hides the complexity of the relationship. Although the overall effect size is small (with confidence intervals for the estimated hazard ratio including 1), similar findings have been reported in two large independent paediatric HCM cohorts [55, 214] and have been interpreted as LVOT obstruction having a potentially protective effect in childhood. There may be unmeasured confounders contributing to the observed relationship between LVOT obstruction and SCD risk in this sample.

For example, it is possible that some of the patients included may have undiagnosed RAS-MAPK syndromes, which are known to have a higher prevalence of obstructive disease and lower risk of arrhythmic events [29]. Additionally, the importance of latent or provocable obstruction has not been explored. However, whilst there is no clear pathophysiological explanation for this observation, it is clear that the relationship between LVOT obstruction and SCD risk in childhood is complex and requires further exploration. This is particularly important for patient care as LVOT gradient is potentially modifiable, unlike most other traditional risk factors [63, 118, 231]. Including myectomy in the HCM-Risk-Kids model in an exploratory analysis did not change the estimated hazard ratio but did reduce its significance in a multivariable model, raising the possibility that gradient reduction therapy could modify SCD risk. Further longitudinal studies are needed to explore the relationship of LVOT gradient and SCD risk in childhood.

Limitations

As childhood HCM is a rare disease and SCD is an uncommon event, a multicentre, retrospective, longitudinal design was necessary to develop a paediatric-specific model. This study is therefore limited by inherent problems of retrospective studies, in particular, missing data. The higher proportion of patients with at least 1 missing predictor compared with the adult development cohort (48.5% in the present study vs 21.7%[82]) may be explained by difficulties obtaining certain investigations in young patients (eg, ambulatory electrocardiogram) and the use of contemporaneously written echocardiographic reports. Missing data were associated with milder hypertrophy and the absence of heart failure symptoms, meaning the complete case analysis is inherently biased towards patients

with more severe disease. This bias may explain the predominance of patients with a calculated 5-year risk score of 4% or greater (n = 349/527) and also suggests that clinicians are more likely to investigate thoroughly in the presence of severe disease. As the cohort was recruited longitudinally, the length of follow-up for individual patients varied, with a median length of follow-up of 5.3 years. The longevity of an ICD device is reported to be between 5 and 9 years [232, 233] although children are known to be at increased risk for lead-related complications necessitating revision [159, 182]. The finite battery life and need for repeated device replacements, along with the lifetime burden of complications, needs to be carefully considered by clinicians when counselling patients and their parents on ICD implantation. Although this study includes data collected across a wide time period, medical management of children with HCM has not changed significantly over this time, and I have shown in Chapter 3 no era effect on survival. However, patients presenting in the earliest era (pre1990) were more likely to be symptomatic for heart failure symptoms and have NSVT detected. Patients presenting in more recent years had lower absolute MWT and corresponding z scores, but this difference did not reach statistical significance. The difference in MWT and corresponding z score may be the result of patients being diagnosed at an earlier time point in disease expression, possibly through family screening, although not at a younger age.

Inherent to the study design, a survival bias is unavoidable as patients not surviving an out-of-hospital arrest are not represented. Future studies comparing those surviving an out of-hospital arrest and those identified post-mortem would be useful but was beyond the scope of this work. The model I have developed should only be used in patients with similar clinical characteristics to the study cohort. In particular, it

should not be used in patients presenting in infancy or with syndromic disease. Future studies exploring the risk of SCD in these subgroups are required. Although preliminary analysis suggests that LVOT obstruction may have a 'protective' effect, the absence of serial clinical data and small numbers of patients with obstructive disease limited my ability to investigate the relationship between LVOT gradient and risk. Well-designed multi-centre studies are required to address the question as to whether gradient reduction therapies could play a role in modifying a patients SCD risk profile.

6.6 Conclusions

This part of my thesis has developed the first validated risk stratification model for SCD in childhood HCM (HCM Risk-Kids) in a large, international cohort using readily collected clinical risk factors. The individualized estimates of risk could help clinicians to identify patients at highest risk and balance the risk of an arrhythmic event with prophylactic ICD implantation. External validation studies are now required to demonstrate the accuracy of this model's predictions in diverse patient populations.

Chapter 7: Exploring novel risk factors for SCD in childhood hypertrophic cardiomyopathy: 12 lead electrocardiogram

7.1 Introduction

HCM Risk-Kids is the first validated paediatric specific model for risk prediction in childhood HCM. However, in common with similar adult risk prediction models based on traditional clinical risk factors, its performance remains imperfect meaning additional predictors may be important for prognosis[214, 216]. The 12-lead electrocardiogram (ECG) is a routine, low cost clinical investigation in HCM that provides qualitative and quantitative information about the phenotype. The ECG phenotype of childhood HCM has not previously been systematically described, but abnormalities are seen in over 90% of adult patients[150, 234]. Studies in adults have reported conflicting findings about the association of individual ECG abnormalities (such as measures of left ventricular hypertrophy[156, 235] and abnormal [150, 236] or prolonged repolarisation pattern[152, 153, 237]) and SCD. However, to date, only a single group has investigated the role of ECG phenotype in risk stratification during childhood[94, 156]. They have proposed an ECG risk score to predict arrhythmic events in HCM independently of traditional clinical risk factors, but this approach has not been independently validated in children[94, 161].

7.2 Aim

The aims of this chapter were to

1) Describe the ECG phenotype of childhood HCM in a large, international, multi-centre cohort
2) To investigate the role of the ECG phenotype in risk prediction for arrhythmic events.

7.3 Methods

Study population:

A multi-centre, retrospective cohort of patients aged 16 years or younger fulfilling diagnostic criteria for HCM, with an available baseline resting 12-lead ECG were identified from the International Paediatric Hypertrophic Cardiomyopathy Consortium[216] described in chapter 2. Patients with a previous history of resuscitated cardiac arrest or sustained ventricular tachycardia, were excluded. Patient assessment, data collection and clinical outcomes have been previously described in chapter 2.

Resting 12-lead ECGs from baseline evaluation were analysed using electronic callipers independently by myself and 3 other reviewers (Miss Ella Field, Mr Cristian Topriceanu and Miss Helen Walsh, Centre for Inherited Cardiovascular diseases, Great Ormond Street Hospital) unaware of the clinical details of the patients. ECGs were excluded if they were not within 6 months of baseline evaluation or if ECG trace quality was poor. Inter-observer reliability was quantified using the one way intraclass coefficient (ICC) and discrepancies were reviewed by a senior supervisor (Dr Juan Pablo Kaski, Centre for Inherited Cardiovascular diseases, Great Ormond Street Hospital).

Statistical analysis:

General statistical methods are described in Chapter 2.

Initially, univariable Cox regression models were used to screen a list of pre-specified variables (ECG and clinical) not included in the HCM Risk-Kids model or ECG risk score based on a significance level of 50% and complete case analysis. A multivariate model was then fitted with all the variables that had a P-value of <0.5 in

the univariate screening together with all variables in the HCM Risk-Kids model and ECG risk score. The missing values for these variables were imputed using multiple imputation techniques based on chained equation [218]. The imputation model included all explanatory variables, the Nelson Aalen estimator of the cumulative hazard function and the MACE outcome. A total of 40 imputed datasets were created. Penalised multivariable regression with least absolute shrinkage and selection operator (LASSO) was used to select the final variables for the multivariable model. Penalised regression was performed in collaboration with Dr Chen Qu and Professor Rumana Omar at Institute of statistical sciences, University College London.

Validation of the ECG risk score in paediatric HCM

The ECG risk score [156]was calculated for each patient and a threshold score of >5 was used to define high risk as described previously in Chapter 2. I assessed the discriminatory performance of this threshold for MACE occurring within 5 years or end of follow up using Harrell's C-index. A value of 1 indicates perfect discrimination and a value of 0.5 indicates no discrimination. The positive predictive value (PPV) of a threshold of >5 was calculated by dividing (sensitivity x prevalence) by ((sensitivity x prevalence) + (1-specificicity) x (1-prevalence)) and expressed as a percentage. The negative predictive value (NPV) was calculated by dividing (specificity x (1-prevalence)) by ((1-sensitivity x prevalence) + (specificity (1-prevalence)) and expressed as a percentage.

7.4 Results

Baseline characteristics:

356 patients with available resting ECG data from 28 centres were identified from the HCM-risk KIDS cohort (n=1029). I compared the baseline demographic and clinical characteristics of the ECG subgroup and total HCM risk cohort in Table 28 below. The ECG cohort had marginally lower MLVWT Z scores (mean MWT Z score 10.1 +/-6.5 vs 11.1 +/-7.2, p value 0.041) and more recent year of presentation (pre-2010 n=125, 35.1% vs n=578, 56.2%, P value < 0.001).

Variable	HCM Risk-Kids (n=1029)		ECG cohort (n=356)		
		Missing data, n		Missing data, n	p-value
Male	702 (68.2%)	0	245 (68.9%)	0	0.834
Pre-2000	161 (15.6%)	0	13 (3.7%)	0	**<0.001**
2000-2010	417 (40.5%)		112 (31.4%)		
2010 onwards	451 (43.8%)		190 (53.4%)		
Age at baseline, years (mean +/- SD)	10.0 ± 4.5	0	10.1 ± 4.5	0	0.747
NYHA >1	223 (21.8)	18	84 (23.6)		0.507
FHx SCD	131 (12.7)	4	36 (10.1)	0	0.184
Unexplained syncope	98 (9.5)	60	37 (10.4)	0	0.882
NSVT	61 (5.9)	191	22 (6.3)	45	0.919
MLVWT (mm)	17.1 ± 7.4	86	16.4 ± 7	5	0.101
MLVWT z-score (mean +/- SD)	11.1 ± 7.2	123	10.1 ± 6.5	12	**0.041**
LA diameter (mm) (mean +/-SD)	32.7 ± 9.4	303	33.0 ± 8.8	65	0.726
LA diameter z-score (mean +/- SD)	1.9 ± 2.3	354	1.82 ± 2.3	68	0.420
Maximal LVOT gradient (mmHg) (median, IQR)	9 (6 - 21.5)	158	9.3 (6 - 20)	51	0.845

Table 28 Baseline demographic and clinical characteristics of HCM Risk-Kids cohort and ECG cohort.
ECG, electrocardiography; SD, standard deviation; NYHA, New York Heart Association; NSVT, non-sustained ventricular tachycardia; MWT, maximal wall thickness; LA, left atrium; LVOT, left ventricular outflow tract; ICD, implantable cardiac defibrillator; MACE, major arrhythmic cardiac event

Prevalence of electrocardiographic characteristics:

I describe the prevalence of individual ECG abnormalities in table 2. Nine patients (2.5%) had no ECG abnormalities at baseline. Two hundred and seventy-seven (77.8%) had one or more repolarisation abnormalities. A pseudo-STEMI pattern was present in 93 patients (26.3%) [38 had ST elevation, 10 had giant positive T waves and 45 had both], a pseudonecrosis pattern in 86 patients (24.4%) and a low QRS voltages pattern in 1 (0.3%). Figure 24 below shows examples of common ECG abnormalities. One hundred and forty-five patients (40.7%) had an ECG risk score of > 5.

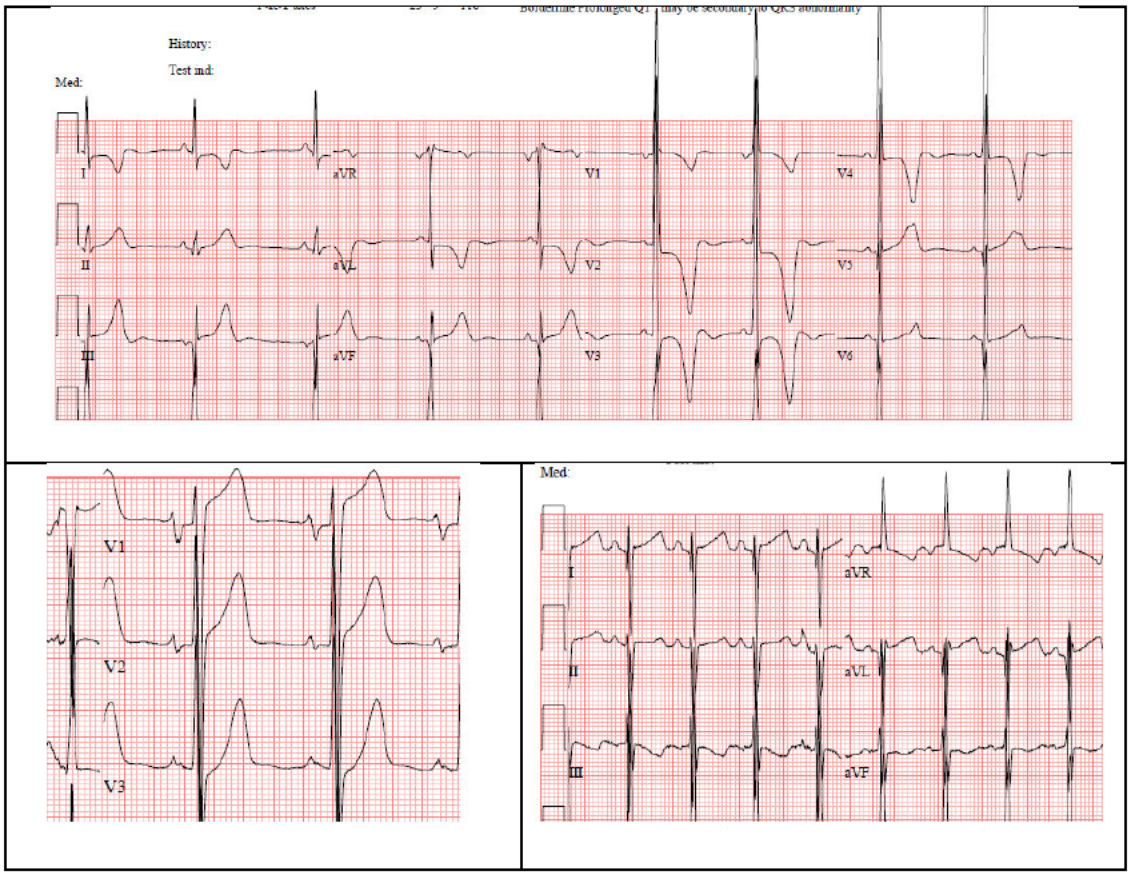


Figure 24 Examples of common electrocardiograph patterns
a)Left ventricular hypertrophy, pseudonecrosis and T wave abnormalities; b) Pseudo-STEMI pattern; c) Bi-atrial enlargement, Superior QRS axis and prolonged QTc

ECG variable		Whole cohort (n=356)	MACE (n=25)	No MACE (n=331)	P value
QRS axis abnormal		126 (35.4%)	10 (40.0%)	116 (35.1%)	0.777
	Left	87 (24.4%)			
	Right	27 (7.6%)			
	Extreme right/left	12 (3.4%)			
Pre-excitation		16 (4.5%)	1 (4.0%)	15 (4.5%)	>0.999
Left atrial enlargement (n=354)		41 (11.6%)	2 (8.0%)	39 (11.8%)	0.416
Right atrial enlargement (n=351)		70 (19.9%)	4 (16.0%)	66 (19.9%)	0.362
Pathological Q-waves (n=352)		167 (47.4%)	11 (44.0%)	156 (47.1%)	0.881
	Inferior	84			
	Lateral	14			
	Inferolateral	66			
	Anterior	3			
Giant inverted T-waves (n=352)		28 (7.9%)	4 (16.0%)	24 (7.3%)	0.132
	Inferior	2			
	Lateral	3			
	Inferolateral	5			
	Anterior	17			
Giant positive T-waves (n=353)		82 (23.2%)	9 (36.0%)	73 (25.0%)	0.186
	Inferior	3			
	Lateral	22			
	Inferolateral	1			
	Anterior	53			
Pathological T-wave inversion-any lead		196 (55.1%)	18 (72.0%)	178 (53.8%)	0.077
	Limb leads (n=355)	172 (48.5%)	17 (68.0%)	155 (46.8%)	0.069
	Precordial leads (n=353)	124 (35.1%)	14 (56.0%)	110 (33.2%)	**0.040**
ST-segment depression >2mm		59 (16.6%)	6 (24.0%)	53 (16.0%)	0.398
	Inferior	14			
	Lateral	9			
	Inferolateral	20			
	Anterior	14			
ST elevation		122 (34.3%)	13 (52.0%)	109 (32.9%)	0.086
	Inferior	25			
	Lateral	16			
	Inferolateral	15			
	Anterior	62			
Dominant S wave in V4		152 (42.7%)	12 (48.0%)	140 (42.3%)	0.729

Sokolow-Lyon, mm (n=355)		46.4 ±29.7	44.64 ±20.20	46.58 ± 30.33	0.895
LV hypertrophy (SLS ≥35mm)		240 (67.6%)	17 (68.0%)	223 (67.6%)	0.965
Mean QRS duration, ms		96.4 (+/- 40.3)	100 +/- 20	100 +/- 40	0.321
QRS duration >120ms		33 (9.3%)	3 (12%)	30 (9.1%)	0.625
QTc >440 ms		121 (34.0%)	12 (48.0%)	109 (32.9%)	0.189
Left bundle branch block		11 (3.1%)	1 (4.0%)	10 (3.0%)	>0.999
Right bundle branch block (n=355)		9 (2.5%)	1 (4.0%)	8 (2.4%)	>0.999
ECG patterns	Low QRS voltages (n=355)	1 (0.3%)	0 (0.0%)	1 (0.3%)	>0.999
	Pseudonecrosis (n=353)	86 (24.4%)	6 (24.0%)	80 (24.2%)	>0.999
	Pseudo-STEMI (n=353)	93 (26.3%)	11 (44.0%)	82 (24.8%)	0.065
Total ECG Risk score		4.9 +/-3.24	6.1 ± 3.40	4.9 ± 3.22	0.063
	Risk score >5	145 (40.7%)	15 (60.0%)	130 (39.3%)	0.068

Table 29 Baseline electrocardiographic characteristics of the study population

Arrhythmic events

Over a median follow up of 3.9 years (IQR 2.0-7.7), 5 patients (1.4%) underwent cardiac transplantation and 14 (3.9%) died: SCD (n=9, 2.5%), heart failure (n=4, 1.1%) and thrombo-embolic event (n=1, 0.3%). Overall annual mortality rate was 0.77 per 100 patient years (95% CI 0.46-1.30). Twenty-five patients had a MACE: appropriate ICD therapy n=12 (3.4%), SCD n=9 (2.5%), resuscitated cardiac arrest n=3 (0.8%) and sustained VT with haemodynamic compromise n=2 (0.6%). Overall annual MACE rate was 1.38 per 100 patient years (95% CI 0.93-2.04). ECG characteristics associated with MACE on univariable analysis are described in Table 29 above.

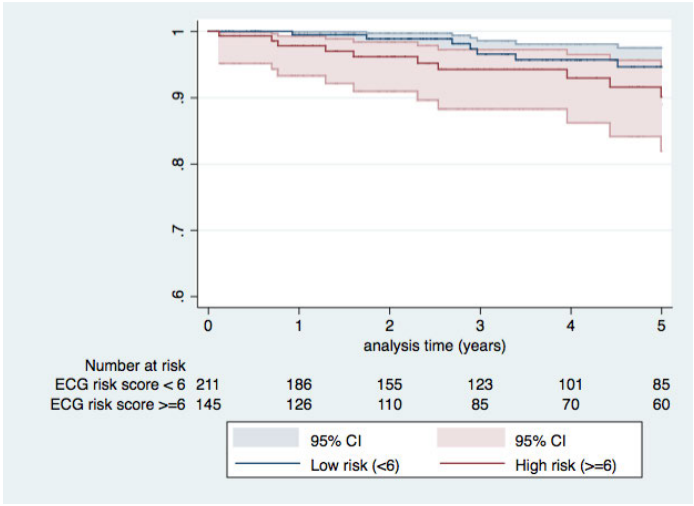
Role of the ECG in predicting 5-year arrhythmic events

In 17 patients, the MACE end-point was reached within 5 years of follow up. The clinical characteristics associated with 5-year MACE on Cox univariate regression analysis were measures of LV hypertrophy (MLVWT HR 1.09 (95% CI 1.03-1.15, p value 0.002), MLVWT Z score HR 1.07 (95% CI 1.01 – 1.13, p value 0.002) and LA diameter (HR 1.05 (95% CI 1.00-1.10, p value 0.067). No ECG variables were associated with the end-point (Table 30 below). On multivariable analysis, only MLVWT, LA diameter and LVOT gradient were associated with MACE. MLVWT and LVOT gradient were selected in all imputed datasets by Lasso regression and LA diameter was selected in 60% of imputed datasets.

	Univariate Cox regression		Multivariate penalized regression
Variable	**HR (95% CI)**	**p-value**	**Lasso estimates**
Clinical risk factors			
Heart failure (NYHA >1)	0.62 (0.18-2.14)	0.446	
FHx of SCD	1.08 (0.25- 4.73)	0.918	
Unexplained syncope	2.33 (0.76 – 7.14	0.140	
NSVT	2.71 (0.77-9.53)	0.120	
MLVWT (mm)	1.09 (1.03-1.15)	**0.002**	1.063
MLVWT z-score	1.07 (1.01 – 1.13)	**0.002**	
LA diameter (mm)	1.05 (1.00 – 1.10)	0.067	1.014
LA diameter z-score	1.03 (0.84 – 1.26)	0.788	
Maximal LVOT gradient (mmHg)	1.00 (0.99 – 1.02)	0.519	1.001
ECG risk factors			
Pathological Q-waves	0.65 (0.24 – 1.75)	0.394	
Giant inverted T-waves	0.58 (0.77 -4.39)	0.600	
Giant positive T-waves	1.60 (0.59 – 4.33)	0.354	
Pathological T-wave inversion-any lead	1.85 (0.65 – 5.26)	0.247	
Pathological T-wave inversion limb leads	2.42 (0.85 – 6.88)	0.097	
Pathological T-wave inversion precordial leads	1.51 (0.58 – 3.90)	0.400	
ST-segment depression >2mm	1.18 (0.67 – 2.07)	0.562	
ST elevation	2.06 (0.79 – 5.34)	0.138	
Dominant S wave in V4	1.46 (0.56 – 3.79)	0.436	
Limb-lead QRS-sum, mV	1.05 (0.97 – 1.14)	0.198	
Chest-lead QRS-sum, mV	1.00 (0.98 – 1.02)	0.950	
12-lead QRS-sum, mV	1.00 (0.99 – 1.01)	0.849	
12-lead product, mV	1.03 (0.94 – 1.13)	0.576	
Sokolow-Lyon, mm	1.00 (0.98-1.01)	0.939	
Mean QRS duration, ms	4.38 (0.00-77)	0.698	
QTc >440 ms	1.75(0.67-5.54)	0.250	
Left bundle branch block	1.73 (0.23 – 13.10)	0.595	
Right bundle branch block	1.96 (0.25-14.87)	0.515	
Low QRS voltages	0.00 (0, ∞)	0.998	
Pseudonecrosis	0.89 (0.29 – 2.75	0.846	
Pseudo-STEMI	2.37 (0.91 – 6.14)	0.076	

Total Risk score	1.12 (0.97 – 1.29)	0.121
Risk score > 5	1.70 (0.78 – 5.37)	0.147

Table 30 Cox regression analysis for arrhythmic outcome within 5 years



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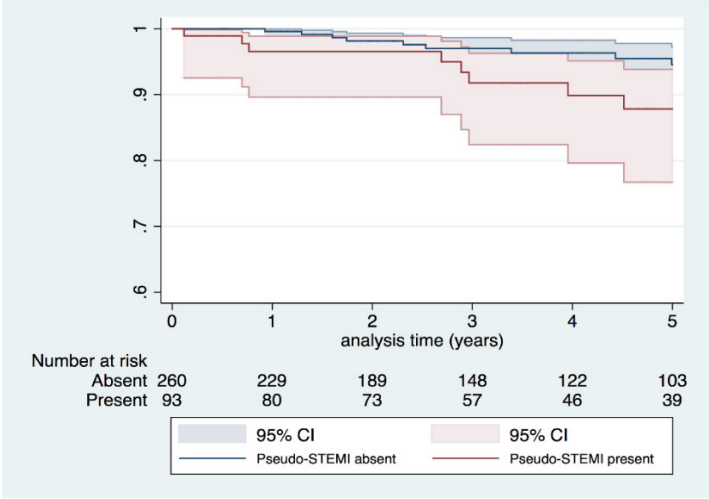

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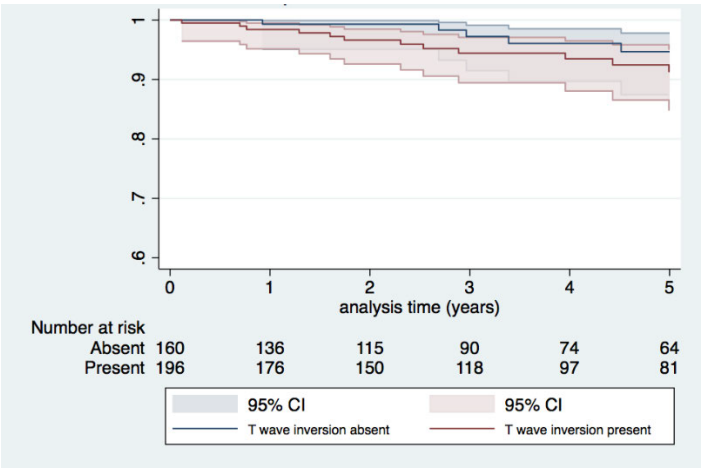

C

Figure 25 Kaplan Meier survival curve showing estimates of event free survival from arrhythmic events by ECG pattern

a) ECG risk score b) the presence of a pseudo-STEMI pattern c) the presence of pathological T wave inversion

Performance of ECG risk model in predicting 5-year arrhythmic event:

Of 145 patients with an ECG score >5; 135 (93.1%) did not have a MACE within 5 years. Harrell's C-index, which represents the probability of correctly distinguishing between high and low risk patients using an ECG risk score threshold of >5 was 0.606 (95% CI 0.584-0.630) at 5 years. The corresponding positive and negative predictive values were 6.9% (95% CI 4.7-10.1%) and 96.7% (95% CI 94.3-98.1%).

Performance of ECG risk model in predicting end of follow up arrhythmic event:

Of 145 patients with an ECG score >5; 130 (89.7%) did not have an event by the end of follow up. Harrell's C-index, which represents the probability of correctly distinguishing between high and low risk patients using an ECG risk score threshold of >5 was 0.560 (95% CI 0.560-0.585) at 5 years. The corresponding positive and negative predictive values were 10.3% (95% CI 7.5-14.0%) and 95.3% (95% CI 92.5-97.0%).

7.5 Discussion and Limitations

In this part of my thesis I have performed the largest description of the ECG phenotype of childhood HCM to date and shown a high prevalence of ECG abnormalities. One third of patients did not meet ECG criteria for LVH, suggesting it is not a sensitive measure of morphological LVH. No single ECG parameter was associated with risk. The ECG risk score had moderate discriminatory ability but with a low positive predictive value. This suggests that the role of the baseline ECG phenotype in improving clinical risk stratification in childhood HCM is limited.

Prevalence of ECG abnormalities in childhood HCM

Less than 3% of the cohort had a normal resting 12 lead ECG, which is comparable to previous reports of 4-6% in adult patients[150, 234]. The most common ECG findings were repolarisation abnormalities, LV hypertrophy, QRS axis abnormalities and QT prolongation. Of note, one third of patients did not meet ECG criteria for LVH, suggesting that this is not a sensitive measure of hypertrophy in childhood HCM as previously described in adult cohorts[235]. One third of patients had QT prolongation, which has previously been reported to be predictive of both all cause[152, 238] and arrhythmic mortality in adult HCM cohorts[153, 237, 239] but this does not appear to be the case in children. A small number of patients had ECG abnormalities typically associated with syndromic or metabolic disease (eg superior QRS axis and pre-excitation). As genotype information was not available, this raises the possibility of undiagnosed non-sarcomeric disease.

Although the most common ECG abnormalities are similar to those reported in adult patients[150, 152, 235], age-specific differences were seen. Biagini et al[150] have previously described three distinct ECG patterns in adult HCM patients: pseudo-necrosis, low QRS voltages and pseudo-STEMI, which was more common in younger patients. In keeping with this, a higher proportion of this cohort had a pseudo-STEMI pattern (26% vs 17%) whilst only one patient had low QRS voltages. The finding of low voltages in a small proportion (3%) of adult HCM patients did not appear to be related to end-stage disease and it is possible that differences in body habitus between paediatric and adult patients could explain the failure to observe this pattern during childhood. The proportion of patients with electrocardiographic evidence of left atrial enlargement was also lower than in adults (14 vs 34%)[150]. The cardiac phenotype is recognised to evolve rapidly during childhood and early

adulthood, and it is likely that these differences reflects age-related progression of both the cardiac and electrocardiographic phenotype. Future studies correlating the evolving ECG and cardiac phenotype in childhood HCM are required.

The association of ECG abnormalities with arrhythmic events

Children are known to have a higher overall risk of arrhythmic events compared to adult patients (1.1 vs 0.8)[40, 81, 185, 216] despite a lower proportion of traditional clinical risk factors, suggesting that additional risk factors may be important in childhood HCM. Previous studies, all but one of which have included mainly adult patients, have reported conflicting findings regarding the association of individual ECG abnormalities and arrhythmic events[150, 152, 153, 156, 235]. The only previous paediatric study [94, 161] reported a significant association between an arrhythmic event and individual ECG abnormalities (such as LV hypertrophy (sum R/S waves, Sokolow-Lyon index and QRS amplitude duration product) and ST segment depression). In contrast, in my analysis, although a higher proportion of patients with T wave inversion experienced a MACE, no individual ECG abnormalities at baseline were statistically associated with 5-year risk of an arrhythmic event on time-dependent univariable or multivariable analysis. As previous studies have shown that ECG patterns may correlate only weakly with a patient's clinical phenotype[235], it is perhaps not surprising that ECG variables on their own have a limited ability to predict risk. On multi-variable analysis, only clinical risk factors previously recognised to be associated with SCD (eg measures of LV hypertrophy, left atrial dilatation and left ventricular outflow tract obstruction) and included in current risk stratification guidelines, were associated with the arrhythmic end-point. My results suggest that individual ECG parameters may not improve current risk prediction models.

The performance of the ECG risk score

The ECG risk score developed by Östman -Smith et al is composed of 8 ECG variables; deviation in QRS axis, pathological T wave inversion in limb or precordial leads, ST-segment depression, dominant S wave in V4, limb-lead amplitude sum, 12-lead amplitude duration product and QTc. Although initially developed in a small cohort of adult patients, it has been reported to be a strong predictor of SCD in children with HCM[156, 161] with a score of 5 or more having a PPV and NPV of 45% and 99% for an arrhythmic event respectively[161]. I have performed the first external validation of the ECG risk score in childhood and have shown it to have only a modest ability to discriminate between high and low risk patients over 5 years follow up (C-statistic 0.6026). This performance is comparable to the current paediatric ESC[4]/AHA guidelines[5] but lower than the newly developed paediatric risk model HCM Risk-Kids described in Chapter 6 [214, 216]. This supports the hypothesis that, although individual ECG parameters may not be predictive of events, a composite score could be useful. The high NPV of the ECG risk score means that patients with a low score can indeed be reassured. However, the low PPV (<10%) suggests that, if used on its own to guide ICD implantation, the majority of patients classified as high risk would not experience an appropriate therapy but would be exposed to the risk of long-term device-related complications[158]. Indeed, two fifths of this cohort had an ECG score of more than 5 and would have been defined as high risk leading to ICD implantation. Importantly, the baseline ECG score was not associated with arrhythmic events when traditional clinical risk factors were accounted for suggesting that its role in improving risk stratification may be limited. The poorer performance of the ECG risk score in this cohort may in part be explained by the timing of ECG recordings or length of follow up. Although the ECG

risk score was developed using ECGs from baseline assessment, in the previous paediatric validation study, the last available ECG before an event or end of follow up was used meaning the calculated ECG risk score was temporally related to the event in question. This likely reflects an evolving cardiac phenotype during childhood and future studies exploring the use of serial clinical investigations for predicting risk should include ECG parameters.

The outcome of 5-year MACE was chosen to assess the ability of the ECG risk score to predict events that could be treated by ICD implantation during childhood as the reported longevity of an ICD device is between 5 and 7 years [232]. Nonetheless, the performance of the model did not improve when I considered whole follow up. Additionally, although the cohort of patients reported by Ostman-Smith et al is derived from a national cohort, it had a higher prevalence of traditional risk factors (eg NSVT (29%), family history SCD (29%), LVOT obstruction (45%)) compared to other population based cohort studies[15, 16, 216] and one fifth of patients had syndromic disease known to have a different arrhythmic risk profile. It may not therefore be truly representative of the wider sarcomeric childhood HCM population. Of note, my findings agree with the only external validation of the ECG risk score in an adult HCM cohort[155].

Limitations

HCM is a rare disease in childhood and thus my analysis was limited by small numbers of patients and events, despite being recruited from a large international consortium of expert centres (n=39). Due to its retrospective design, only one-third of the HCM risk SCD cohort had an available 12-lead ECG meeting inclusion criteria, and this differed by era. A larger number of historic patients (pre-2010) did not meet

inclusion criteria but the mean length of follow up in the ECG or whole cohort did not differ. Patients in this analysis had a lower mean MLVWT, which could be explained by a higher proportion of screening patients included in more recent era, but did not otherwise differ in terms of baseline clinical characteristics or incidence of arrhythmic events to other large population studies. This suggests that the results of this study are representative and applicable to a wider childhood HCM population. Nonetheless, the small number of events may have limited our ability to detect statistically significant differences. This cohort of patients did not include those presenting in infancy or with syndromic disease who are known to have a worse prognosis and may differ in ECG phenotype. Future studies describing and investigating the role of the 12-lead ECG in these patient groups are needed.

7.6 Conclusions

In this chapter I have described that in a large, international, multi-centre cohort of childhood HCM, ECG abnormalities are common and varied, occurring in over 95% of patients. Despite a high prevalence of abnormalities, no individual ECG findings were associated with an arrhythmic event. The ECG risk score had a modest ability to discriminate between high and low risk patients but with a low PPV. This suggests that the role of baseline ECG phenotype in improving risk stratification in childhood HCM is limited.

Chapter 8: Exploring novel risk factors for SCD in childhood hypertrophic cardiomyopathy: genotype

8.1 Introduction

In common with adult-onset disease, the majority of disease presenting during childhood is caused by variants in sarcomeric protein genes[24, 25]. Current guidelines recommend the use of genetic testing to facilitate family screening[4, 5], yet there is interest in whether genotype could be used to predict long-term outcomes. Whilst genotype-phenotype correlations have been reported[144], such as minimal hypertrophy associated with *Troponin T* disease[141, 142, 240], studies in adults describing associations between genotype and arrhythmic risk have reported conflicting findings[139, 230, 240-242]. To date only small single-centre studies have investigated the role of genotype for risk stratification in childhood disease [60, 243].

8.2 Aim

The aims of this chapter were to

1) Describe the genetic spectrum of non-syndromic childhood HCM in a large, international, multi-centre cohort
2) To investigate the role of genotype in risk prediction for arrhythmic events during childhood

8.3 Methods

Study population

I formed a multi-centre, retrospective cohort of patients aged 16 years or younger fulfilling diagnostic criteria for HCM who had undergone genetic testing from the International Paediatric Hypertrophic Cardiomyopathy Consortium[216] described in chapter 2. Patients with a previous history of resuscitated cardiac arrest or sustained ventricular tachycardia, were excluded.

Patient assessment, data collection and clinical outcomes have been previously described in chapter 2.

Reporting of genetic variants

The American College of Medical Genetics and Genomics (ACMG) guidelines[194] provide standards for assessing variant pathogenicity using different sources of evidence. Reported variants are classified as Pathogenic (P), Likely Pathogenic (LP), Variant of Unknown Significance (VUS), Likely benign (LB) and Benign (B) (Figure 26 below). Sources of evidence include;

- Population databases – to obtain frequencies of variants in large population studies (eg dbSNP[244], gnomAD[245])
- Disease databases – list variants identified in patients with a particular disease (eg ClinVar[246], OMIM (https://omim.org), Health in Code Inc mutation database (https://mutaciones.healthincode.com)
- *In silico* predictive data tools to predict the effect of the variant on protein structure or splicing (eg PolyPhen[247], SIFT[248]).
- Functional studies to predict the effect of the variant on protein structure or splicing
- Segregation of the reported variant with disease

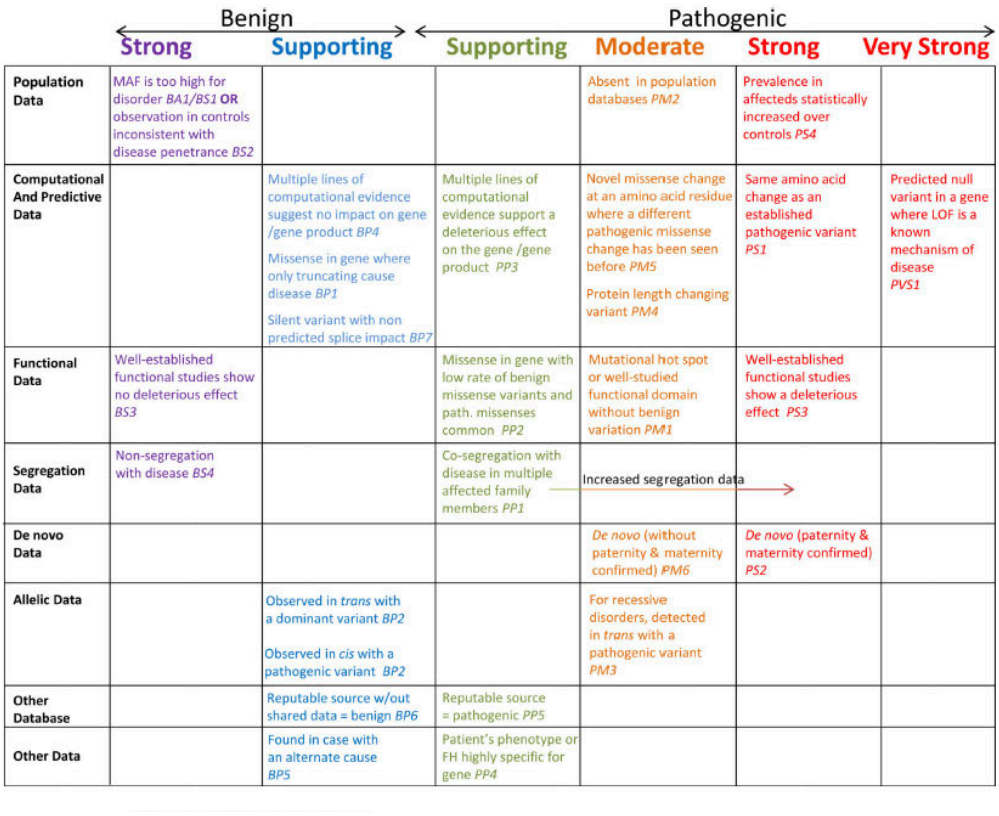

Figure 26 Evidence framework for classification of genetic variant. Reproduced from Richards et al[194]

BS = benign strong; BP = benign supporting; FH= family history; LOF = loss-of-function; MAF = minor allele frequency; path = pathogenic; PM = pathogenic moderate; PP = pathogenic supporting; PS = pathogenic strong; PVS = pathogenic very strong

All variants reported by collaborating centres were reclassified according to ACMG guidelines by Dr Lorenzo Monserrat from *Health In Code (Ltd*) as pathogenic, likely pathogenic, variant of unknown significance, likely benign and benign. Additional information on major and supporting criteria used by *Health In Code* is provided in Appendix 2.

Variants identified in genes not associated with HCM (non-HCM) were excluded from this analysis.

Statistical analysis:

General statistical methods are described in Chapter 2.

I used univariable Cox proportional hazards regression models to test the assumption of linearity for each genetic predictor using the end point of SCD or an equivalent event occurring during follow up. I performed multivariable Cox regression analysis with entry set at a significance level of 0.2. Variable selection was performed using a backward selection approach retaining variables with a significance of less than 0.1%. For each variable, the hazard ratio and corresponding 95% confidence intervals are reported.

8.4 Results

Data on genetic testing was available for 828 patients (80.9%) from 33 collaborating centres, of which 505 (61%) patients had undergone genetic testing. Year of genetic testing was known in 305 (61.0%) patients; pre-2010 n=56 (18.4%), 2010-2015 n=101 (33.1%), 2015 onwards n=148 (48.5%) (Figure 26 below). Size of genetic panel varied; predictive testing (n=65, 12.9%), small panel (n=123, 24.4%), expanded panel 115 (22.8%) and unknown (n=202, 40.0%). Genetic testing strategy changed by era with increasing use of expanded NGS panels since 2011 (Figure 26 below). Patients who underwent genetic testing were more likely to have; presented in recent eras (49.7% vs 34.1%, p value <0.001), a family history of HCM (56.6% vs 51.7%, p value 0.035) and an absence of heart failure symptoms (18.3% vs 26.0%, p value 0.003). Other baseline characteristics were comparable as described in Table 31 below.

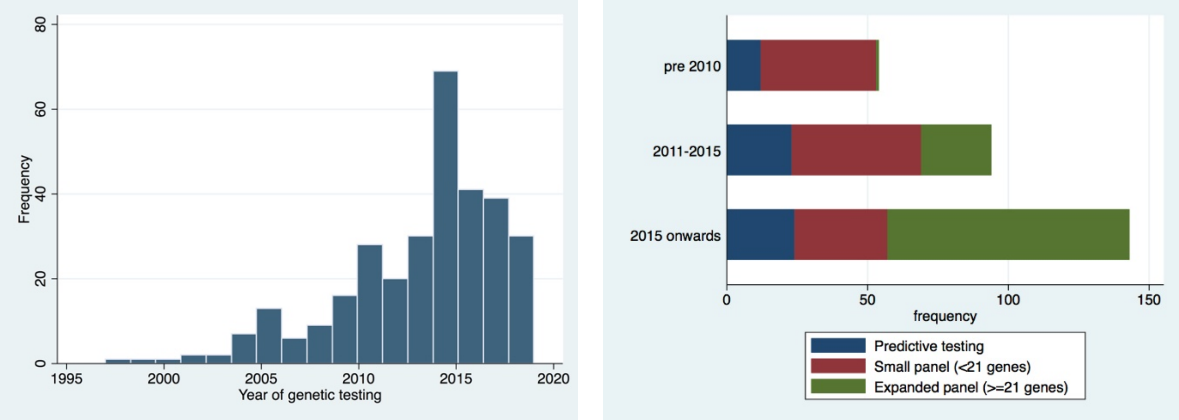

Figure 27 Genetic testing strategy by era
a)Number of patients undergoing genetic testing over time b) Size of genetic panel by era

		Whole cohort (n=1024)	Genotyped (n=505)	Not genotyped (n=518)	P value
Age at baseline (median, IQR)		11 (7-14)	11.2 (6.9-14.0)	11.7 (7.6-14.3)	0.111
Era of presentation	Pre-2000	160 (16.1%)	77 (15.4%)	83 (16.8%)	**<0.001**
	2000-2005	155 (15.6%)	63 (12.6%)	92 (18.6%)	
	2005-2010	262 (26.4%)	111 (22.2%)	151 (30.6%)	
	2010-2015	330 (33.2%)	192 (38.5%)	138 (28.0%)	
	>2015	86 (8.7%)	56 (11.22%)	30 (6.1%)	
Male sex		699 (68.3%)	350 (69.3%)	348 (67.2%)	0.465
FHx HCM (n-1011)		537 (53.1%)	269 (56.6%)	268 (51.7%)	**0.035**
FHx SCD (n=1020)		130 (12.8%)	73 (14.5%)	57 (11.1%)	0.102
Unexplained syncope (n=1023)		102 (9.9%)	51 (10.1%)	51 (9.9%)	0.901
NYHA/Ross >1 (n=1006)		223 (22.2%)	90 (18.3%)	133 (26.0%)	**0.003**
B Blockers at baseline (n=1021)		410 (44.2%)	212 (42.1%)	198 (38.4%)	0.527
NSVT on ambulatory ECG (n=856)		55 (6.4%)	28 (6.6%)	27 (6.2%)	0.812
MLVWT (n=997)		17.1 (7.4)	17.2 (7.4)	17.0 (7.4)	0.6822
Z Score MLVWT (n=906)		11.1 (7.4)	11.0 (6.9)	11.1 (7.4)	0.8064
LA diameter (n=712)		33.4 (8.5)	32.0 (7.6)	35.0 (9.1)	**<0.001**
Z score LA diameter (n=675)		1.9 (2.3)	1.5 (1.9)	2.3 (2.6)	**<0.001**
Maximal LVOT gradient (n=871)		9 (6-22)	9 (5.4-20)	9.4 (6-23)	0.6426
SCD during follow up		104 (10.2%)	46 (9.1%)	58 (11.2%)	0.269
SCD during childhood (<21 years)		89 (8.7%)	38 (7.5%)	51 (9.9%)	0.188
Incidence of SCD/ 100 pt yrs (95% CI)		1.44 (1.19-1.75)	1.25 (0.94-1.67)	1.65 (1.27-2.13)	0.181
Mean length of follow up yrs (+/- SD)		7.0 (6.1)	7.3 (6.9)	6.8 (5.1)	0.2087

Table 31 Baseline demographic and clinical characteristics in HCM Risk-Kids cohort and genetic testing cohort

Outcome of genetic testing

Of 505 patients undergoing genetic testing, a variant was identified in 391 (72.2%) patients. Following ACMG reclassification (n=369 (73.1%)), variants were classified as pathogenic (P) (n=229), likely pathogenic (LP) (n=60), variant of unknown significance (VUS) (n=132), likely benign (n=38) or benign (n=5). Two hundred and fifty-five patients had a single P/LP variant identified in a thick filament sarcomeric protein (*MYBPC3* (n=100) or *MYH7* (n=101)), thin filament sarcomeric proteins (n=40) or non-sarcomeric protein (n=14). Seventeen patients had two P/LP variants identified. A VUS in a HCM gene was identified in 35 patients with one or more P/LP variant and 89 with no P/LP variants. The results of genetic testing are described in Table 32 below and Figure 27 below. The proportion of patients with a P/LP variant did not vary by era of testing (n=45 (80.4%) vs n=75 (73.3%) vs n=104 (70.3%), p value 0.349) Figure 27 below. The proportion of patients with a P/LP variant identified was highest for those undergoing predictive (n=57, 87.7%) or small panel (n=94, 76.4%) testing (Figure 27 below). A VUS was most commonly identified in patients with expanded panel testing (small 36, 29.3% vs expanded n=51, 44.4%, p value < 0.001).

Patients with one or more P/LP sarcomeric variants (SARC+) were more likely to have a family history of HCM and higher MLVWT compared to those with no sarcomeric variant (SARC-) or a VUS only (Table 33 below).

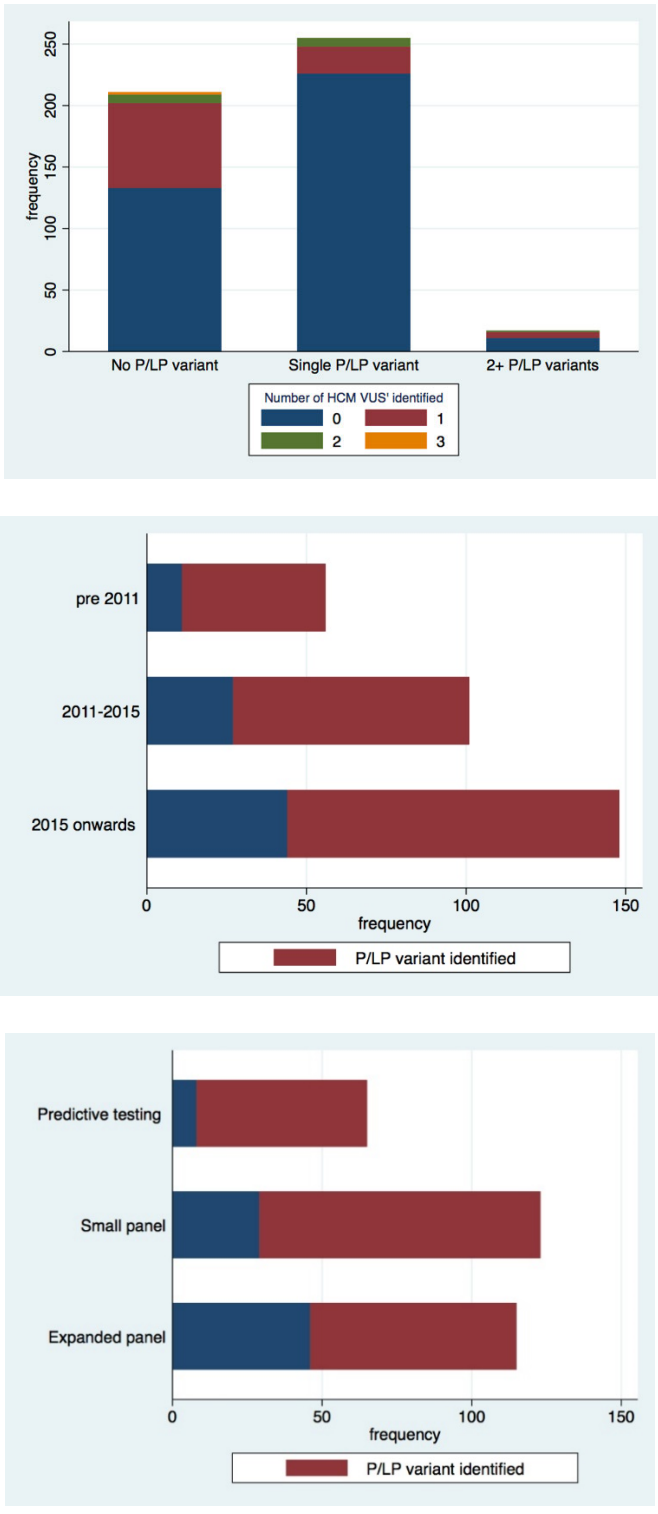

Figure 28 Outcome of genetic testing
A)Presence of VUS by number of P/LP variants identified; B)Proportion of patients with a P/LP variant identified by era of testing; C) Proportion of patients with a P/LP variant identified by genetic testing strategy

ACMG Pathogenic or likely pathogenic variants	**Single variant (n=255)**	Thick filament proteins (n=201)	*MYBPC3*	**100**
			MYH7	101
		Thin filament proteins (n=40)	*ACTC*	7
			MYL2	7
			MYL3	1
			TPM1	9
			TNNI3	8
			TNNC1	1
			TNNT2	7
		Non-sarcomeric proteins (n=14)	*JPH2*	3
			KRAS	1
			LAMP2	2
			PRKAG2	3
			RAF1	1
			DES	3
			FHOD3	1
		Single Pathogenic variant + VUS (n=31)		
		VUS in HCM gene (n=29)	*MYBPC3*	19
			MYH7	7
			TNNI3	1
			TPM1	1
			TTN	1
	Two variants (n=17)	*MYBPC3 + TPM1*		1
		MYBPC3 + MYH7 (LP)		4
		GLA (LP) + SCN5a (LP)		1
		MYBPC3 + MYBPC3		2
		MYH7 + MYH7 (LP)		1
		DES + MYH7 (LP)		1
		MYBPC3 + MYBPC3 (LP)		1
		MYBPC3 + TNNT2		1
		MYH7 + TNNT2		2
		MYBPC3 + MYH7		1
		MYBPC3 + DMD (LP)		1
		MYH7 (LP) + MYH7 (LP)		1
		VUS in HCM gene (6)		
			MYBPC3	4
			GLA	1
			MYH7	1
ACMG VUS	**Single VUS**	69		
	2 or more VUS	9		
		VUS in HCM gene		
			ACTC1	3
			ACTN2	2
			CSPR3	7
			FLNC	2
			HRAS	1
			LZRT1	1
			MYBPC3	10
			MYH6	1
			MYH7	31

	MYL2	2
	MYL3	1
	MYON	1
	PRKAG2	3
	SOS1	4
	TNNT2	5
	TPM1	3
	TTN	5
	VCL	2

Table 32 Results of genetic testing following ACMG reclassification of reported variants

	Genetic testing (n=483)	SARC + (n=272)	SARC – (n=133)	SARC VUS (n=78)	Global P value	SARC + Vs SARC -
Age at baseline (median, IQR)	11.2 (6.9-14.0)	10.1 (4.6)	10.2 (4.6)	10.6 (1.3)	0.602	0.873
Male sex	350 (69.3%)	180 (66.2%)	98 (73.7%)	55 (70.5%)	0.293	0.126
FHx HCM (n=475)	269 (56.6%)	183 (68.0%)	46 (35.4%)	40 (52.6%)	**<0.001**	**<0.001**
FHx SCD (n=482)	71 (14.7%)	45 (16.5%)	16 (12.0%)	10 (13.0%)	0.434	0.233
Unexplained syncope (n=483)	47 (9.7%)	27 (9.9%)	14 (10.5%)	6 (7.7%)	0.785	0.851
NYHA/Ross >1 (n=471)	85 (18.1%)	46 (17.3%)	24 (18.3%)	15 (20.3%)	0.837	0.801
NSVT on ambulatory ECG (n=406)	26 (6.4%)	14 (6.1%)	9 (8.3%)	3 (4.4%)	0.560	0.444
MLVWT (n=475)	17.2 (7.4)	18.2 (7.8)	15.8 (6.2)	16.4 (6.1)	**0.002**	**0.003**
Z Score MLVWT (n=446)	11.0 (6.9)	11.9 (7.2)	10.0 (6.3)	9.8 (5.7)	**0.029**	0.079
LA diameter (n=360)	32.0 (7.6)	32.0 (7.6)	30.9 (7.0)	33.1 (7.9)	0.542	0.340
Z score LA diameter (n=342)	1.5 (1.9)	1.55 (1.9)	1.34 (1.8)	1.6 (2.2)	0.357	0.674
LVOTg max (n=419)	9 (5.4-20)	9 (5,18)	10 (5,25)	10 (6.3,30)	0.474	0.589
SCD during follow up	42 (8.7%)	25 (9.2%)	11 (8.3%)	6 (7.7%)	0.899	0.760
SCD during childhood (<21 years)	34 (7.0%)	20 (7.4%)	10 (7.5%)	4 (5.2%)	0.770	0.952
Incidence of SCD/ 100 pt years (95% CI)	1.25 (0.94-1.67)	1.24 (0.84-1.83)	1.09 (0.61-1.97)	1.13 (0.51 – 2.52)	0.925	0.695
Mortality or transplant	22 (4.6%)	10 (3.7%)	7 (5.3%)	5 (6.4%)	0.534	0.455

Table 33 Table comparing the baseline demographics and clinical characteristics by the presence of a pathogenic sarcomere mutation

Relationship of the outcome of genetic testing and arrhythmic events

Over a mean follow up of 7.3 years (SD +/- 6.9), 46 patients met the end point of SCD or an equivalent event with an overall event rate of 1.28 per 100 patient years (95% CI 0.94 – 1.67). Univariate and multivariate analysis for SCD event is shown in Table 34 below. The presence or number of P/LP variants was not associated with an arrhythmic event occurring during follow up Figure 28 below. Although a higher proportion of those with an arrhythmic event had a P/LP variant in a thin filament sarcomeric protein (n= 6, 14.3% vs n=37, 8.4%, P value 0.116), this did not reach statistical significance. A variant in MYBPC3 or MYH7 was not associated with arrhythmic events. On multivariable analysis, only Z score MLVWT and unexplained syncope were associated with the outcome.

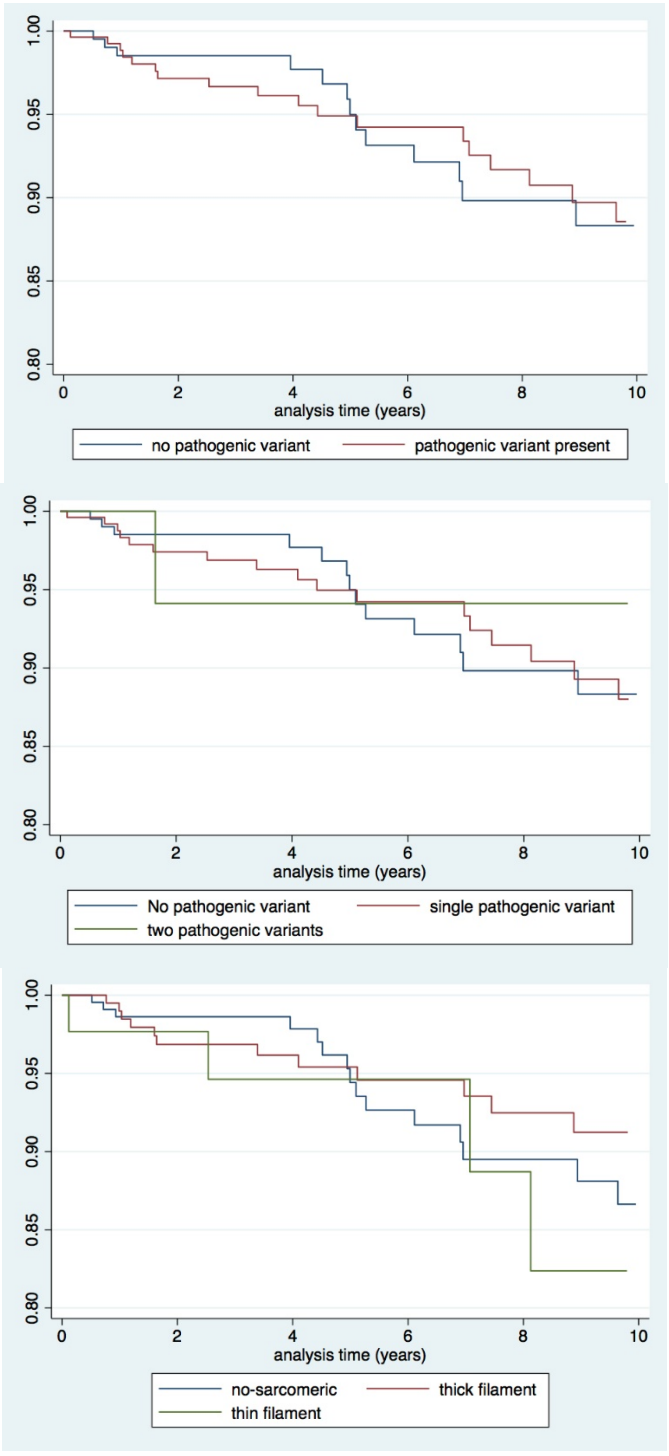

Figure 29 Kaplan Meier survival curve showing event free survival estimates by results of genetic testing

a) presence of a pathogenic/likely pathogenic HCM causing variant b) number of pathogenic/likely pathogenic variant c) location of pathogenic/likely pathogenic variant

		Whole cohort (n=483)	SCD event (n=42)	No SCD event (n=441)	Univariate Hazard ratio (95% CI)	P value	Multivariate hazard ratio (95% CI)	P value
Age at baseline (median, IQR)		11.2 (6.9-14.0)	9.2 (6.9-13.6)	11.2 (7.0-14.0)	1.01 (0.95.108)	0.738		
Male sex		333 (68.9%)	32 (76.2%)	301 (68.3%)	0.69 (0.34 – 1.40)	0.306		
FHx SCD (n=482)		71 (14.7%)	9 (21.4%)	62 (14.1%)	1.45 (0.69-3.03)	0.324		
Unexplained syncope		47 (9.7%)	1 (26.2%)	36 (8.2%)	2.33 (1.16-4.70)	**0.018**	2.42 (1.16-5.05)	**0.0018**
NYHA/Ross >1 (n=471)		85 (18.1%)	10 (24.4%)	75 (17.4%)	1.37 (0.67 – 2.83)	0.391		
B Blockers at baseline (n=482)		202 (41.9%)	21 (51.2%)	181 (41.0%)	1.13 (0.61 – 2.11)	0.690		
NSVT on ambulatory ECG (n=406)		26 (6.4%)	7 (20.0%)	19 (5.1%)	1.63 (0.68-3.88)	0.274		
MLVWT (n=476)		17.2 (7.2)	20.4 (6.6)	16.9 (7.3)	1.03 (1.00 – 1.07)	**0.075**		
Z Score MLVWT (n=476)		11.0 (6.8)	15.7 (7.2)	10.6 (6.6)	1.06 (1.02 – 1.10)	**0.005**	1.05 (1.02-1.09)	**0.006**
LA diameter (n=361)		31.9 (7.5)	33.9 (8.2)	31.7 (7.4)	1.03 (0.98-1.08)	0.255		
Z score LA diameter (n=343)		1.5 (1.9)	2.4 (2.1)	1.4 (1.9)	1.15 (0.97 – 1.37)	**0.112**		
LVOTg max (n=419)		9 (5.4-20)	10 (6-30)	9 (5-20)	1.00 (0.99-1.01)	0.473		
Any HCM variant (VUS/P/LP)		350 (72.5%)	31 (73.8%)	319 72.3%)	1.04 (0.75-1.44)	0.822		
1+ HCM variant (VUS/P/LP)	1	294 (84.3%)	28 (90.3%)	267 83.7%)	0.53 (0.16 – 1.73)	0.291		
	2 or more	55 (15.7%)	3 (9.7%)	52 (16.3%)				
P/LP variant		272 (56.3%)	25 (59.5)	247 (56.0%)	1.12 (0.51 – 2.08)	0.716		
No. P/LP variants	1	255 (52.8%)	24 (57.1%)	231 (50.3%)	1.22 (0.66-2.24)	0.530		
	2	17 (3.5%)	1 (2.4%)	16 (3.6%)	0.52 (0.072-3.81)	0.523		
Protein type*	Thick filament	217 (44.9%)	17 (40.5%)	200 (45.4%)	0.75 (0.40-1.39)	0.360		
	Thin filament	43 (8.9%)	6 (14.3%)	37 (8.4%)	2.01 (0.84 – 4.80)	**0.116**		
Protein *	*MYBPC3*	112 (23.2%)	8 (19.1%)	104 23.6%)	0.77 (0.36-1.67)	0.514		
	MYH7	109 (22.6%)	9 (21.4%)	100 22.7%)	0.79 (0.38 – 1.65)	0.524		

Table 34 Univariable and multivariable cox regression analysis for arrhythmic events and genetic testing *For pathogenic/likely pathogenic variants

8.5 Discussion and limitations

Uptake of genetic testing in childhood HCM

This large, unselected longitudinal cohort offers a unique opportunity to describe the current use of genetic testing in childhood HCM. Not surprisingly, genetic testing has increased over time with a higher proportion of patients undergoing testing in the most recent era (2015 onwards) compared to more historic patients (48% vs 65%, p value 0.010). The clinical characteristics of those with and without genetic testing was similar apart from a lower proportion of patients with heart failure symptoms or left atrial dilatation in those who had undergone testing. One possible explanation for this finding is over-representation of patients who were diagnosed with HCM through family screening who may be more likely to undergo genetic testing and have a milder cardiac phenotype at baseline. In support of this, clinicians were more likely to perform genetic testing if there was a family history of HCM.

Results of genetic testing in childhood HCM

Previous adult studies have reported that a disease-causing sarcomeric variant is identified in 40-60% of HCM patients [139, 230, 241, 249], most commonly in *MYBPC3* or *MYH7* gene proteins[20]. There are comparably few paediatric specific studies, but a disease-causing variant has been identified in 60-80% of patients in small cohort studies[60, 243]. It is beyond the scope of this work to determine the yield of genetic testing in childhood HCM as genetic testing was performed at the treating clinicians' discretion. However, just over half of patients had a pathogenic or likely pathogenic variant, with a VUS identified in a further 18%. Only 17 (3.5%) were compound heterozygote or homozygote for pathogenic sarcomeric variants. This confirms previous findings that the majority of non-syndromic disease, even in young

patients, is caused by single sarcomeric protein variants [24, 25]and suggests that the yield of genetic testing in childhood disease is similar to adult cohorts. The majority of disease-causing variants were identified in *MYBPC3* or *MYH7* genes, with a smaller proportion (15%) in thin filament proteins. Genetic testing additionally identified variants in proteins associated with syndromic or metabolic HCM (eg LAMP2, RAF1 or PRKAG2) in a small number of patients. This could suggest undiagnosed non-sarcomeric disease, but may also support the role of non-sarcomeric variants contributing to disease phenotype in children with clinically non-syndromic disease[139, 250]

Role of genotype in risk stratification

In a large cohort (n=2763) of patients derived from the SHaRE consortium, Ho et al demonstrated that the presence of any disease-causing sarcomeric variant was associated with earlier disease onset (<40 years), more severe hypertrophy and a higher cumulative life time risk of adverse events (heart failure, atrial fibrillation, NYHA ≥3, stroke, SCD, cardiac transplantation, SCD or all cause death)[230, 251]. Although outcomes were modelled from birth, only 9% of the cohort was under the age of 18 years at diagnosis and the majority were recruited from adult centres, meaning an inherent childhood survival bias exists. Nonetheless, these findings replicate what has previously been reported in smaller adult cohort studies[139, 241] with a recent meta-analysis[252] suggesting that the presence of a sarcomere mutation is important for prognosis. In this chapter, I found that sarcomere positive individuals were more likely to have a family history of HCM and more severe hypertrophy. However, there was no association between the presence of a sarcomere variant and an arrhythmic event. The absence of an effect could be secondary to low numbers of events, which reduced my ability to detect statistically

significant effects. However, it could also partly be explained by the timing of arrhythmic events as the majority of events in the SHaRE registry occurred after childhood whilst three quarters of the HCM-Risk Kids cohort remain under the age of 21 years at last follow up. The lifetime risk of malignant arrhythmias is therefore not accurately represented in this dataset. Of note, a previous small paediatric study described worse outcomes for sarcomeric positive patients (including those with a VUS) for a composite outcome of ICD implantation, myectomy, cardiac arrest, cardiac transplantation or death[243]. As this outcome includes multiple, at times clinician determined, outcomes this finding is difficult to compare to the results above. Therefore, whilst the presence of a sarcomere variant is important for long-term prognosis in HCM, it is not clear that is has a significant role to play in predicting arrhythmic risk during childhood.

In small adult cohort studies, the presence of multiple disease causing sarcomeric variants has been linked with a more severe disease phenotype[20, 146, 167, 252] and increased risk of SCD[145]. Paediatric specific studies are sparse, yet case reports have described particularly poor prognosis for compound MYBPC3 heterozygotes/homozygotes presenting during childhood[168, 253]. In this analysis, I confirm that compound heterozygotes/homozygotes are rare, even in childhood disease. Only 1 arrhythmic event occurred in a patient heterozygous for two MYBPC3 variants aged 13 years. Three of the patients with two variants had variants in one or more non-sarcomeric genes (eg DMD, GLA, DES) whose contribution to disease phenotype is unclear. However, possessing two pathogenic variants was not statistically associated with an increased arrhythmic risk on univariable regression analysis.

With advances in genetic testing and larger panel sizes, an increasing number of variants of unknown significance are being identified[139]. Indeed, in this cohort, one or more VUS' were identified in over two fifths of expanded panel tests compared to one third of smaller, more limited panels. Whilst VUS' are often disregarded, Ho et al described an intermediate risk of experiencing an adverse event in patients with a VUS[230] but this did not reach significance for the arrhythmic end-point. The only previously published paediatric study found an increased risk of arrhythmic events in the presence of multiple non-benign sarcomeric variants (including VUS')[243]. Together this suggests that some of these VUS' are pathogenic and that variant burden itself may be important for prognosis. In my analysis, in agreement with the large SHaRE consortium, no significant relationship between arrhythmic events and the presence of single or multiple non-benign HCM-variants was found. Nonetheless, careful interpretation and revisiting of reported variants may be important for long-term prognosis in childhood disease.

Efforts to explore genotype-phenotype correlations in HCM have been limited by significant genetic heterogeneity and variable or incomplete age-related penetrance. Whilst initial small, highly-selected population studies reported an increased risk of arrhythmic events with particular genes (such as troponin T[140, 142]), these results have not been replicated in larger cohort studies[141, 240]. Indeed, in this cohort whilst the presence of a thin filament variant trended towards an increased risk on univariable Cox regression analysis, this effect was not seen when other traditional clinical risk factors were accounted for. The presence of a disease-causing variant in either MYBPC3 or MYH7 was similarly not associated with arrhythmic events during follow up. It is likely that this is because investigating genotype-phenotype correlations at the level of the gene ignores the significant heterogeneity that is seen

between variants in different regions of a gene. Furthermore, although specific areas of genes (eg converter region of MYH7) have been described to be associated with a worse prognosis[242], more recent studies have shown that the natural history may vary considerably even within a gene region[143]. Insufficient evidence therefore currently exists to support the use of genotype at the level of the gene or gene region in risk stratification. A variant specific approach is likely needed but this may be limited by small numbers of patients with individual variants.

Limitations

This analysis is subject to the same limitations as other retrospective cohort studies, including missing data and survivor bias in recruited patients. As genetic testing was performed at the treating clinicians' discretion, and not systematically in all patients, there is the potential for enrichment bias in the sample. In particular, genetic testing was more likely to be performed in those with a family history of HCM and without symptoms of heart failure, which could be a proxy for more severe disease. Previous, mainly adult, studies have reported that predictors for identifying a disease-causing variant on genetic testing include younger age, family history of HCM and degree or pattern of hypertrophy[254-256]. Although only 13% of the cohort were documented to have had predictive genetic testing, proband status was not determined in this cohort so separate analysis by proband status was not possible. When considered together, these features mean that this cohort of patients likely does not truly represent the overall genetic burden in childhood HCM and future, prospective studies are required to investigate the yield of genetic testing in childhood HCM.

There was significant variability in sequencing methods and panel size reported in this cohort of patients. The ability to detect a pathogenic variant is of course dependent on that gene being included in a sequencing panel. However, reassuringly almost 90% of pathogenic variants were in *MYBPC3* or *MYH7*, which would be included in all testing panels regardless of the panel size. The size of the panel is likely to be more important for the interpretation of the role of VUS' in prognosis as I have shown that a VUS was more likely to be identified if an expanded panel is performed. A prospective, multi-centre registry is needed to overcome these challenges.

8.6 Conclusions

In this chapter I have confirmed a high prevalence of sarcomeric disease in non-syndromic childhood HCM and described changing patterns of genetic testing over time. Pathogenic variants were most commonly identified in *MYH7* or *MYBPC3* proteins and compound hetero- or homozygous variants were uncommon. Arrhythmic risk was not predicted by the presence, number or protein location of sarcomeric variants. Large-scale, multi-centre registries are needed to explore the impact that individual variants or gene-regions have on long-term prognosis for childhood onset disease.

Chapter 9: Conclusions, overall limitations and future work

In this thesis, using a unique international cohort of children with HCM, I have for the first time, systematically investigated risk factors for SCD in childhood disease and developed a paediatric specific approach to risk stratification.

9.1 Summary of findings

In Chapter 3, I described the clinical presentation and outcomes of childhood HCM over time in a national UK cohort[205]. I assessed the impact of changing screening practices and showed an increase in the detection of both syndromic and familial sarcomeric disease. For the majority of patients, long term outcomes were good, but aetiology and age of presentation had a significant impact on both survival and cause of mortality. I showed that SCD remains the most common mode of death, outside of infancy, and occurs at a higher rate than in adult cohorts. Despite this, identifying patients at the highest risk remains challenging as evidenced by the majority of events occurring in patients without a primary prevention ICD device.
In chapter 4, I performed the first systematic review and meta-analysis of clinical risk factors for SCD in childhood HCM[207]. The evidence base supporting individual risk factors was not strong with the majority of studies reporting small, heterogeneous, highly selected populations. Four ‘major’ clinical risk factors likely to be important for risk stratification in childhood disease were identified; unexplained syncope, NSVT, left ventricular hypertrophy and previous VF/VT. Additional risk factors that have to date been infrequently investigated in childhood, including LA diameter, are also likely to be important. Some risk factors with significant supporting evidence in adult disease, such as family history of SCD, appear to be less important in childhood

disease. The extrapolation of adult practice to childhood patients may therefore be inappropriate.

Using data from the International Paediatric Hypertrophic Cardiomyopathy consortium, in Chapter 5, I performed the first external validation study of the current European and North American risk stratification guidelines in childhood HCM[215]. Current guidelines recommend the use of four traditional clinical risk factors for risk stratification; unexplained syncope, NSVT on ambulatory ECG monitoring, family history of SCD and extreme LVH. A primary prevention ICD is recommended in the presence of one or two risk factors for the North American or European guidelines respectively. I showed that the guidelines have only a modest ability to discriminate between patients at high and low risk for a future arrhythmic event (C statistic 0.62 and 0.60 at 5 years) leading to unnecessary ICD implantation in many.

As current guidelines have limited discriminatory power, in Chapter 6 I developed the first validated model for SCD risk stratification in childhood HCM. As the first step in model development I performed an external validation of the adult HCM-Risk SCD model in a childhood cohort. I showed that although the model's discriminatory ability was superior to current guidelines (C-statistic 0.67 vs 0.62), risk estimates were inaccurate meaning that risk was underestimated for the majority of patients. I subsequently developed a new paediatric-specific model using 5 pre-selected clinical variables (unexplained syncope, NSVT, LA diameter Z-score, MWT Z-score and LVOT gradient), which for the first time provides clinicians and patients with individualised estimates of 5-year risk[216]. The new model showed superior discrimination between high- and low-risk patients compared to current paediatric guidelines (C-statistic 0.69 vs 0.62), with good calibration between expected and observed risk.

Whilst this model represents a significant improvement on current risk stratification methods, it remains imperfect meaning that additional risk factors may be important to refine the model. In Chapter 7 and Chapter 8 I explored two such candidate risk factors, ECG phenotype and genotype. In the first systematic description of the ECG phenotype in childhood HCM I showed that ECG abnormalities were common, and varied, occurring in over 95% of patients. Despite a high prevalence of ECG abnormalities, no individual findings, or the previously proposed ECG risk score, were associated with an arrhythmic event. This suggests that the ECG phenotype has a limited role to play in improving risk stratification. In Chapter 8, I confirmed that the majority of non-syndromic disease in childhood is secondary to variants in sarcomeric muscle proteins, most commonly MYH7 or MYBPC3. Although limited by small numbers of events, arrhythmic risk was not predicted by the presence, number or protein location of sarcomeric variants. Exploring a variant specific approach to risk stratification could be useful, yet there is currently insufficient evidence to support the use of genotype in childhood risk stratification.

9.2 Overall limitations

As childhood HCM is a rare disease and SCD a rare outcome, a retrospective, longitudinal multi-centre study design was required, which has inherent limitations. The use of retrospective data spanning several decades could mean that findings are not generalizable to current or future patient cohorts. Additionally, as the International Paediatric HCM consortium recruited patients from multiple centres in various geographical locations, differences in the assessment and clinical investigation of patients is inevitable. However, whilst changes in the patient demographics were seen over time in the UK cohort there was no significant change

in outcomes or SCD event rates. This likely reflects limited changes in treatment and patient management over time and suggests that the results are applicable to current medical practice. A higher proportion of patients had at least 1 missing predictor compared to the adult development cohort (48.5% vs 21.7%)[82]. This could be explained by centre-specific or age-related difficulties in obtaining certain investigations such as ambulatory ECG monitoring. Missing data was dealt with using established statistical techniques yet it is an intrinsic limitation to the analysis. The collection of data from multiple geographical sites is additionally a strength of this study as, in contrast to previous studies from single or similar geographical regions, its results are generalizable to a wider childhood HCM population. Although local collaborators were asked to recruit consecutive patients meeting the diagnostic criteria for HCM, it is possible that not all eligible patients were identified if a contemporary database of HCM patients was not kept. The majority of centres included are tertiary referral centres, which may lead to an enrichment bias towards those with more severe disease. However, reassuringly, the characteristics of the patient population are similar to previously published cohorts[15, 16, 214]. Finally, there is an inherent survival bias in the patient cohort as those presenting with but not surviving an out of hospital arrest are not represented.

Despite this study reporting the largest cohort of children with non-syndromic disease in the world to date, the number of SCD-events remained small. This reduced our power to detect statistically significant differences. SCD is a rare event in HCM and therefore a composite end-point of SCD or an equivalent event was chosen in line with previous publications in this field to increase the effective sample size[82, 257]. For patients experiencing ICD therapies, independent review of the presenting cardiac rhythm on intracardiac electrograms (EGM) was performed by

each local investigator to ensure ICD therapy was appropriate (ie for a ventricular tachyarrhythmia). However, although ICD's have been shown to successfully treat ventricular tachyarrhythmias, it is important to recognise that ICD therapies are not truly equivalent to SCD as ventricular arrhythmias may self-terminate.

One criticism of the adult HCM risk SCD model is that although traditional methods of risk stratification may recommend unnecessary ICD implantation in patients that do not experience an event, the risk calculator leaves some low-intermediate risk patients with single risk factors unprotected. In Maron et al's external validation of the HCM risk SCD model, 60% of patients who experienced an event with an estimated risk <4% would have been recommended an ICD according to the ACCF/AHA guidelines[188]. This demonstrates a problem shared with all risk models; it is never possible to identify every patient who has an event. However, as ICD implantation is not a complication-free intervention, particularly in childhood[157, 182], it is important to balance the risk of intervening with that of closely monitoring. Of note, in the HCM Risk-Kids development cohort, only 9% of the patients were deemed as high risk according to the ESC guidelines, of which one third had an event. This means that three quarters of events occurred in patients considered to be at low risk. In comparison, in those with complete data, 47% of patients had a 5-year risk of ≥6% using the newly developed HCM Risk-SCD model, of which 10% had an event. Less than a quarter of events occurred in those with lower 5-year estimates of risk. Although this comparison is limited by missing data, meaning that the simulation is biased towards those with more severe disease, it appears that the new model will predict, and therefore protect, more patients from events but may lead to more ICD implantations. Independent prospective clinical validation studies are needed to analyse the effect of using the model in clinical practice and to

determine if a 5-year risk threshold is appropriate for recommending ICD implantation in childhood HCM. As the risk of mortality from competing causes is lower during childhood, ICD implantation may be appropriate in young patients with lower estimated SCD risks.

An important caveat to using any risk model to predict SCD events is the recognition that risk is dynamic and likely affected by multiple factors that may be disease-specific (eg phenotype, genotype), or external to the disease (eg environmental) and not accounted for within the risk model. The calculated risk estimate is only valid at the time it is calculated, which has important implications for its evaluation in external research populations or in real-world clinical scenarios. This may be particularly relevant for childhood disease due to a rapidly changing clinical phenotype. Future work to incorporate serial phenotypic information could help negate this concern and is discussed below.

Finally, the model I have developed should only be used in patients with similar clinical characteristics to the study cohort. As patients with infant-onset and syndromic (IEM, RASOpathy or neuromuscular) HCM were excluded from model development, the HCM-Risk Kids model cannot be applied in these patient cohorts. Future studies are needed to explore the arrhythmic risk of these patient groups.

9.3 Alternative risk models

In 2020, following the publication of our HCM Risk-Kids model, an alternative paediatric risk model was developed and published by Miron et al in North America[214]. The model was developed on a similar premise to HCM Risk-Kids, using a retrospective multi-centre cohort (n=572 patients) and readily available clinical risk factors to calculate individualised estimates of risk. Whilst there are some

similarities in the model development process there are also important differences in the patient population, predictor variables and methodology. The inclusion criteria for the two studies were identical except for the upper age limit, which was 16 years for HCM Risk-Kids and 18 years for the North American cohort. The ESC adult HCM risk SCD model is validated for use in patients aged 16-18 years and therefore an upper age limit of 16 years was chosen for our paediatric cohort to prevent overlapping cohorts and aid clinical utility of the guidelines. Despite this, the North American cohort was younger at presentation (median age 9.8 (IQR 2.1, 13.9) vs 11 (IQR 7,14)) and had a shorter mean follow up time (mean follow up 3.7 years vs 5.5 years). In terms of baseline clinical characteristics, there was a lower proportion of patients with NSVT (3% vs 6%) and unexplained syncope (3% vs 10%), but a similar proportion with obstructive disease (16% vs 18%). As different Z scores were used to correct for body-surface area in the North American study, and absolute values of MLVWT or LA dilatation were not described, it is not possible to compare the distributions of these variables between the two populations. Clinical risk predictors were included in the model if associated with the end point on multivariable analyses rather than pre-selected based on previous literature. I chose to pre-select variables without studying the predictor-outcome relationship in the development data to prevent a reduction in statistical power and possible sample bias. One important limitation to this approach is the possibility for publication bias with non-significant variables less likely to be reported. Nonetheless, the variables in the two models are identical, with the exception that Miron et al also included age and used two measures of left ventricular hypertrophy (interventricular septal thickness and posterior wall thickness). Age was not included in our model as there is limited evidence that age is important for risk stratification outside of infancy. Additionally,

including age as predictor did not improve our model performance on sensitivity analyses. Using two measurements of LV wall thickness may provide an indication of the distribution of hypertrophy, however it is possible that this approach will lead to an under-estimation of maximal wall thickness for patients with eccentric or apical disease and that collinearity between the predictors could reduce the reliability of the calculated regression coefficients.

Miron et al used machine based learning techniques to develop a model that predicted risk at 5 years. The incidence of SCD in this cohort was similar to the HCM risk-Kids cohort suggesting that both samples are representative of the wider childhood population. The developed model was externally validated in a small cohort of 285 patients with 22 events from the SHaRe consortium. The performance of the model was superior to current guidelines (C-statistic 0.707) and similar to our HCM risk-Kids model. Independent external validation studies of these competing models are needed to compare model performance in clinical practice. However, as both models contain the same clinical risk variables it is plausible that the performance of them will be similar.

9.4 Future work

The work presented in this thesis is the first systematic investigation of SCD risk in childhood HCM and provides valuable and novel insights on the natural history of the disease as well as tangible advances in patient care. However, our understanding of risk stratification remains incomplete and important unanswered questions remain. The key areas that I plan to focus on in the future are discussed and described below.

External validation of HCM Risk-Kids model

A prediction model should ideally provide valid predictions of outcomes for patients in settings distinct from that where the model was developed. To this end, work is currently underway to externally validate the HCM Risk-Kids model in a new independent population to confirm its generalisability. Additional centres have been recruited to the International Paediatric Hypertrophic Cardiomyopathy Consortium and new collaborations have been formed with The Sarcomeric Human Cardiomyopathy Registry (SHaRe) (Appendix).

Investigation of alternative clinical risk factors for SCD in childhood HCM

Quantifying fibrosis on cardiac magnetic resonance imaging (CMRI)

Myocardial fibrosis is implicated in the pathogenesis of HCM and has been proposed to act as a substrate for malignant arrhythmias. Focal replacement fibrosis can be visualised using LGE on CMRI and the presence or extent of LGE has been proposed as a possible independent risk factor for SCD. In adult patients, LGE is detected in approximately 60% of patients [135, 258], typically in the mid-myocardial region, and is associated with the degree of LV hypertrophy. LGE has been assessed as a risk factor for SCD in adult populations with conflicting results. In the only prospective study, Ismail et al found that the extent but not the presence of LGE was associated with arrhythmic events on univariable analysis[258]. However, this effect was not seen when systolic function was adjusted for. In contrast, in several retrospective studies LGE presence was associated with SCD events [135, 259-261], and Chan et al described a proportional relationship between the proportion of LGE and SCD event risk[135]. All studies were limited by small numbers of events and a bias towards those with less severe disease as patients judged to be at high risk who had undergone ICD implantation were excluded from analysis due to technical limitations. In 2015 a meta-analysis, which included 5 independent studies,

was published showing that the presence of LGE was a risk factor for SCD even after adjusting for traditional clinical risk factors [133]. Risk additionally appeared to increase incrementally with proportion of LGE. Since the HCM Risk-SCD model [82] was published and endorsed by the ESC, several studies have hypothesised that LGE on CMRI could improve model performance [137, 262] particularly for low or intermediate risk patients [263]. Maron et al has also suggested that the addition of LGE to ACCF/AHA risk stratification guidelines improves its performance [257]. LGE on CMRI is therefore likely to be important for prognosis in adult disease, yet the extent to which it improves risk stratification beyond traditional markers is not yet clear.

The estimated proportion of paediatric HCM patients with LGE on CMRI ranges from 18-72% in published series [46, 164, 165, 210, 264]. As childhood is a time of disease development and progression, it is likely that these varying estimates reflect a changing phenotype over time. Indeed, in the largest multi-centre study to date, Axelsson-Raja et al described an increase in the presence and proportion of LGE over follow up [46]. In childhood disease, LGE has been shown to be associated with LV hypertrophy but has not been shown to be independently associated with SCD events [46, 164, 210, 264], however these studies have been underpowered to detect a difference. An additional challenge for paediatric practice is that it is unlikely that children under the age of 7 years would tolerate the scan without the need for general anaesthesia. It is therefore likely that, even if LGE is an important risk factor for childhood SCD, it would only be clinically useful in the older childhood cohort. I did not investigate the role of LGE in risk stratification in the HCM Risk-Kids cohort as CMRI is not systematically performed in all centres looking after childhood patients and therefore it is likely that the sample size would have been

underpowered to detect a difference in outcomes. The investigation of LGE as a risk factor to-date has been limited by the largely retrospective design of studies with patient cohorts derived from single, or a small number, of specialist centres in whom CMRI has not been systematically performed or analysed. In 2019, an NIHR Hypertrophic Cardiomyopathy Registry (HCMR) was established, which hopes to overcome some of these challenges, however of note patients under the age of 18 year are excluded from the registry [265]. Large-scale systematic paediatric-specific registries are still therefore needed to address this important clinical question.
While LGE is able to detect focal areas of fibrosis, it is unable to detect diffuse fibrosis given that it relies on the comparison of tissue characteristics with normal myocardium. Newer tissue mapping techniques circumnavigate this problem allowing either native or post-contrast tissue relaxation times to be quantified[266]. To date there is limited literature on the use of T1 mapping in risk prediction but one adult study has shown an association between relaxation times and NSVT or resuscitated cardiac arrest[267]. No study has previously assessed the role of T1 mapping and risk stratification in childhood HCM[268, 269].

Using serial clinical data to predict SCD risk in childhood HCM

Childhood is a time of significant somatic growth, which is likely to be accompanied by rapidly changing disease phenotype. The model I have developed is designed to be used at baseline assessment to predict the risk of an arrhythmic event occurring over the next 5 years. However, in clinical practice, a clinician is likely to wish to reassess a patient's risk during follow up taking into account the changing cardiac phenotype. At the current time, our understanding of phenotype progression during childhood is limited, and it is unknown if individual risk factors have age-specific

effects on prognosis. Future studies exploring the changing role of individual clinical risk factors during childhood and use of serial clinical investigations in predicting risk would be valuable.

9.5 Conclusions

The work contained in this thesis represents the first systematic and comprehensive investigation of SCD in Childhood HCM and confirms that children are at a higher risk of arrhythmic events compared to their adult counterparts. Although risk stratification has traditionally been extrapolated from adult practice guidelines, this thesis highlights the importance of a paediatric specific approach to risk stratification. Thus, a paediatric specific risk model has been developed for use in childhood HCM which for the first time allows clinicians to calculate individualised estimates of SCD risk. Ongoing work is needed to validate this model and investigate the role of additional or serial clinical risk factors.

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Appendix

Appendix 1: International Paediatric Hypertrophic Cardiomyopathy Consortium

	Centre	Number of patients enrolled	% of cohort
Development cohort			
1	Great Ormond Street Hospital, London, UK	210	20.5
2	Children's Memorial Health Institute, Warsaw, Poland	101	9.9
3	Careggi University Hopsital, Florence, Italy	79	7.7
4	Monaldi Hospital, Naples, Italy	52	5.1
5	Onassis Cardiac Surgery Centre, Athens, Greece	45	4.4
6	The Royal Children's Hospital, Melbourne, Australia	44	4.3
7	S. Orsola-Malpighi Hospital, Bologna, Italy	41	4
8	Our Lady's Children's Hospital, Dublin, Ireland	36	3.5
9	Royal Hospital for Children, Glasgow, UK	34	3.3
10	Favaloro Foundation University Hospital, Buenos Aires, Argentina	32	3.1
11	Leiden University Medical Center, Leiden, Netherlands	30	2.9
12	Bambino Gesu Hospital, Rome, Italy	27	2.6
13	University Hospital Motol, Prague, Czech Republic	23	2.3
14	Royal Brompton and Harefield NHS Trust, London, UK	22	2.2
15	Hospital Sant Joan de Deu, Barcelona, Spain	20	2
16	Papa Giovanni XXIII hospital, Bergamo, Italy	19	1.9
17	Birmingham Children's Hospital, Birmingham, UK	15	1.5
18	Hospital General Universitario Gregorio Maranon, Madrid, Spain	15	1.5
19	University Hospital of Wales, Cardiff, UK	14	1.4
20	Leeds General Infirmary, Leeds, UK	14	1.4
21	Val d'Hebron University Hospital, Barcelona, Spain	14	1.4

22	University Hospital Virgen de la Arrixaca, Murcia, Spain	12	1.2
23	Bristol Royal hospital for Children, Bristol, UK	12	1.8
24	Niguarda Hospital, Milan, Italy	12	1.2
25	Complexo Hospitalario Universitario A Coruña, Spain	11	1.1
26	University Hospital La Paz, Madrid, Spain	10	1
27	John Radcliffe Hospital, Oxford, UK	10	1
28	Glenfield Hospital, Leicester, UK	9	0.9
29	Southampton General Hospital, Southampton, UK	9	0.9
30	Hospital Universitario Puerta de Hierro Majadahonda Madrid, Spain	7	0.7
31	Aarhus University Hospital, Aarhus, Denmark	7	0.7
32	University Hospitals Parma, Italy	7	0.7
33	Alder Hey Children's hospital, Liverpool, UK	6	0.6
34	Ghent University Hospital, Belgium	6	0.6
35	Kochi Medical School Hospital, Kochi University, Japan	4	0.4
36	Mater Dei Hospital, Malta	4	0.4
37	Odense University Hospital, Odense, Denmark	3	0.3
38	Evelina Children's Hospital, London, UK	3	0.3
39	Freeman Hospital, Newcastle, UK	2	0.2
External validation cohort			
40	SHaRE consortium	269	
41	Necker –Enfants Malades hospital, Paris, France	63	
42	Heart muscle disease registry Trieste,	13	
43	Hospital Saint Joseph, Marseille, France	1	
44	Charite – Universitatsmedizin Berlin, Germany	17	
45	Rio Horetga University Hospital, Valladolidm Spain	3	
46	Helsinki University Hospital	16	
47	Hokkaido University Hospital, Sapporo, Japan	12	
48	Fondazione Toscana G Monasterio, Massa-Pisa, Italy	12	
49	Children's Hospital ' Louis Turcanu', Timisoara, Romania	4	
50	Great Ormond Street Hospital, London, UK	30	

VARIANT CLASSIFICATION CRITERIA AND CLINICAL UTILITY

Classification	Major Criteria	Supporting Criteria	Clinical Utility
PATHOGENIC OR DISEASE-CAUSING [+ + +]	[1] Widely reported variant with conclusive evidence of a genotype-phenotype association and with consensus about its pathogenicity. [2] Demonstrated cosegregation with a phenotype (>10 meioses). [3] Cosegregation in at least 2 families (≤10 meioses), or present in at least 5 probands with the same phenotype, and meeting at least 2 supporting criteria:	A Protein-truncating variant in a gene where loss of function is a proven pathogenic mechanism. B Functional studies that support pathogenicity. C *De novo* presentation in the setting of a novel disease in the family (maternity and paternity confirmed). D Missense variant that generates the same amino-acid change as a previously reported pathogenic variant. E Variant with very low frequency/absent in the control population (MAF <0.001%).	• Clinical predictive value. • Clinical information. • Genetic counseling. • Familial screening recommended.
VERY LIKELY TO BE PATHOGENIC OR DISEASE-CAUSING [+ +]	[1] Protein-truncating variant in a gene where loss of function is a proven pathogenic mechanism that explains the patient's phenotype, and that meets at least 1 supporting criterion: [2] Missense variant/in-frame insertion or deletion in a non-repetitive region of a gene with demonstrated genotype-phenotype association that explains the patient's disease, and that meets at least 2 supporting criteria:	A Functional studies that support pathogenicity. B *De novo* presentation in the setting of a novel disease in the family (maternity and paternity confirmed). C Affecting a residue in which other pathogenic variants were previously identified. (mutational hot spot); or variant located in a relevant functional domain or region of the protein. D Variant with very low allelic frequency/absent in the control population (MAF <0.001%). E Probable cosegregation in at least one family, or various index cases, but that does not meet criteria for being considered pathogenic.	• Clinical predictive value. • Genetic counseling. (Incomplete information on penetrance and expressivity). • Familial screening recommended.
LIKELY TO BE PATHOGENIC OR DISEASE-CAUSING [+ ?]	[1] Protein-truncating variant with very low frequency/absent in the control population (MAF <0.001%) that affects a gene where loss of function is not an established pathogenic mechanism or that does not meet criteria to be considered pathogenic. [2] Intronic variant outside the consensus region of the gene for which the bioinformatics predictors agree that it would affect the splicing. [3] Missense variant/in-frame insertion or deletion in a non-repetitive region of a gene which does not meet criteria to be considered pathogenic/very likely to be pathogenic, but that meets at least 3 supporting criteria:	A Variant with very low allelic frequency/absent in the control population (MAF <0.001%). B *De novo* presentation in the setting of a novel disease in the family (maternity and paternity unconfirmed). C Patient's phenotype or family history suggests that disease could be explained by mutations in the gene (gene with well-established phenotype-genotype association). D Bioinformatics predictors agree that it would be deleterious. E Located in a mutational hot-spot, functional domain, or relevant region of the codified protein. F Reported in at least 2 unrelated individuals that presented the same phenotype.	• Currently WITHOUT clinical predictive value. • Evaluation of cosegregation can be useful to define pathogenicity.
UNKNOWN CLINICAL SIGNIFICANCE [?]	[1] Variants with contradictory information about their pathogenicity. [2] Variants that do not meet criteria for being included in another classification category.		• WITHOUT clinical predictive value. • Evaluation of cosegregation upon physicians request (research only).
UNLIKELY TO BE PATHOGENIC OR DISEASE-CAUSING [- ?]	[1] Variant allele frequency in control populations is higher than the expected for disease or has a MAF >0.05%. [2] Absence of variant cosegregation with the phenotype in at least 1 family. [3] Meeting at least 2 supporting criteria:	A Missense variant in a gene where only variants causing protein truncation have shown association with disease. B Functional study showing that the variant does not affect the structure or function of the encoded protein. C Bioinformatics predictors agree that the variant would not alter the function of the protein (including splicing variants outside the consensus region of the gene). D In-frame insertions/deletions in a repetitive gene region without a known function. E Presence of the variant in homozygosis in control population.	• WITHOUT clinical predictive value. • Familial screening NOT recommended (research only).
NON-PATHOGENIC (NOT DISEASE-CAUSING) [- -]	[1] MAF >5% in any of the control population databases. [2] Previously reported in the literature with well-established evidence of consensus about its non-disease-causing classification, and with no contradictory data. [3] Absence of cosegregation with the disease in at least 2 reported families. [4] Meeting at least 2 supporting criteria:	A Variant allele frequency in control populations is higher than expected for disease or has a MAF >0.05%. B Absence of cosegregation of the variant with the phenotype in at least 1 family. C Functional study showing that the variant does not affect the structure or function of the encoded protein. D Presence of the variant in healthy unaffected subjects at an age at which the disease should be fully penetrant (variant must be in homozygosis in recessively inherited diseases, or in hemizygosis in X-linked diseases).	• BENIGN • SHOULD NOT be included in familial screening.

Academic output during PhD

Peer-reviewed publications

Norrish G, Chubb H, Field E, Mcleod K, Ilina M, Spentzou G et al. Clinical outcomes and programming strategies of implantable cardioverter defibrillator (ICD) devices in paediatric hypertrophic cardiomyopathy: A UK national cohort strategy. Europace 2020 Nov 22; euaa307. doi: 10.1093/europace/euaa307. Online ahead of print.

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Book chapters

Norrish G, Kaski JP. Hypertrophic Cardiomyopathy. In Diagnosis and Management of Adult Congenital Heart Disese. 4th edition Gaztoulis, Webb, Daubeney and Brida. *In Print*

Norrish G, Akhtar MM, Kaski JP, Elliott PM. Metabolic disease and neuromuscular disorders. In Elliott PM, Lambiase PD, Kumar D (Eds.). Oxford Handbook of Inherited Cardiac Disease 2nd Edition, Oxford University Press (2019)

Norrish G, Kaski JP. Cardiomyopathy in childhood. ESC textbook of cardiovascular medicine. Published 2018

Norrish G, Kaski JP. Hypertrophic Cardiomyopathy. In Diagnosis and Management of Adult Congenital Heart Disese. 3rd edition 615-621. Gaztoulis, Webb, Daubeney and Brida.

Norrish G, Kaski JP. Inherited Arrythmias: LQTS. Challenging concepts in Congenital and Acquired Heart Diseases in the young. Rigby & Jivanji (in print).

Prizes

Young Investigator award, Association of European Paediatric Cardiology 2018

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