

Echocardiographic and electrocardiographic alterations in Fabry Disease

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A thesis submitted to fulfil requirements for the degree of

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This is to certify that to the best of my knowledge, the content of this thesis is my own work. This thesis has not been submitted for any degree or other purposes.

I certify that the intellectual content of this thesis is the product of my own work and that all the assistance received in preparing this thesis and sources have been acknowledged.

Matthew Zada

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Preface

Experimental work contained in this thesis was performed by the author at Westmead Hospital Cardiology Department under the supervision of Professor Liza Thomas between 2020 and 2023 to fulfill the requirements for the degree of Master of Philosophy at The University of Sydney.

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In addition to the statements above, in cases where I am not the corresponding author of a published item, permission to include the published material has been granted by the corresponding author.

Matthew Zada

29/6/23

As supervisor for the candidature upon which this thesis is based, I can confirm that the authorship attribution statements above are correct.

Professor Liza Thomas

29/6/23

Ethics

Ethics approval for this project was provided by the Western Sydney local health District ethics committee (HREC No. ETH13628).

Abbreviations List

^{18}F FDG PET = ^{18}F Fluorodeoxyglucose positron emission tomography

$^{99\text{m}}\text{TcDPD}$ = $^{99\text{m}}\text{Tc}$ Technitium-labelled 3,3-diphosphono-1,2-propanodicarboxylic acid

$^{99\text{m}}\text{TcPYP}$ = $^{99\text{m}}\text{Tc}$ Technitium-labelled pyrophosphate

$^{99\text{m}}\text{TcHMDP}$ = $^{99\text{m}}\text{Tc}$ Technitium-labelled hydroxymethylene diphosphonate

AA = secondary amyloidosis

ACEi = angiotensin converting enzyme inhibitors

AF = atrial fibrillation

AICD = automated implantable cardioverter-defibrillator

AL = amyloid light-chain

ARB = angiotensin receptor blockers

ATTRv = variant or mutant transthyretin amyloidosis

ATTRwt = wild-type transthyretin amyloidosis

ECG = electrocardiograph

ERT = enzyme replacement therapy

FD = Fabry Disease

Gb3 = globotriaosylceramide

GLA = alpha galactosidase A

GLS = global longitudinal strain

HCM = hypertrophic cardiomyopathy

HFpEF = heart failure with preserved ejection fraction

LGE = late gadolinium uptake

LS = longitudinal strain

LV = left ventricular

LVH = left ventricular hypertrophy

Lyso-Gb3 = globotriaosylsphingosine

MACE = major adverse cardiovascular event

MRI = magnetic resonance imaging

NSVT = non sustained ventricular tachycardia

NT-proBNP = N terminal pro-brain natriuretic peptide

TTE = transthoracic echocardiogram

TTR = transthyretin

VT = ventricular tachycardia

Chapter 1 – Introduction and Literature Review

1.1 Overview of Infiltrative cardiomyopathy and Introduction

The infiltrative cardiomyopathies are a group of disorders that impact on cardiovascular function as a result of extracellular deposition (as occurs in amyloidosis, sarcoidosis and mucopolysaccharidoses) or intracellular deposition (as occurs in glycogen, lipid or iron storage disorders) (1). Subsequently, there is progressive increase in left ventricular (LV) wall thickness or left ventricular hypertrophy (LVH) and diastolic dysfunction resulting in a poor prognosis.

In the past, these disorders were irreversible with no treatment options available.

However, with medical advancements, treatments have become available for conditions such as Fabry Disease (FD) and Amyloidosis. Generally, these treatments have better outcomes if commenced earlier in the disease process. Therefore, it is imperative that these disorders are identified early. In addition to increasing awareness amongst physicians, advanced cardiac imaging techniques such as cardiac magnetic resonance imaging (MRI) and advanced echocardiographic techniques, play a fundamental and important role in the diagnosis of these disorders.

The purpose of this thesis is to examine if advanced echocardiographic techniques (such as strain analysis) can identify subclinical cardiac involvement in FD (i.e. prior to the development of LVH). Additionally, this thesis will assess if echocardiographic markers of cardiac involvement in FD are associated with Major Adverse Cardiovascular Events

(MACE). Finally, this thesis will explore if there are gender-based differences in electrocardiographic alterations seen in FD, as well as their association to echocardiographic changes.

1.2 Amyloid cardiomyopathy

Amyloidosis is a systemic disorder caused by the extracellular accumulation of misfolded protein fibrils in various organs, including the heart, ultimately resulting in organ dysfunction (2). There are over 30 recognised proteins that can misfold and form amyloid fibrils, however, the majority are rare (3). The three most common types of systemic amyloidosis are amyloid light-chain (or primary) (AL) amyloidosis, transthyretin (TTR) amyloidosis, and amyloidosis associated with chronic inflammation (AA) (4). The two most common forms of systemic amyloidosis are AL and TTR amyloidosis. AL amyloidosis is caused by deposition of a monoclonal immunoglobulin light chain, which can be in the setting of multiple myeloma, with 50-60% of patients having cardiac involvement (5, 6). TTR amyloidosis results in accumulation of a protein derived from the TTR protein which is produced in the liver. This could be 'wildtype' (ATTRwt) or due to mutations in the ATTR gene (variant or ATTRv). AA amyloidosis typically occurs secondary to chronic inflammatory conditions such as rheumatoid arthritis and Familial Mediterranean Fever and rarely (~2%) involves the heart (4, 6).

Cardiac amyloidosis involves the heart with extracellular deposition of amyloid fibrils in cardiac tissue – this can be cardiac specific, or as a result of systemic amyloidosis. Similar to other forms of infiltrative cardiomyopathy, delay in diagnosis of cardiac amyloidosis

occurs often due to non-specific symptoms which are difficult to differentiate from other causes of diastolic dysfunction and LVH. Clinically, cardiac amyloidosis presents with symptoms and signs of heart failure with preserved ejection fraction (HFpEF), including exertional dyspnoea, orthopnoea, and signs of right heart failure such as an elevated jugular venous pressure, peripheral oedema and ascites (7). Amyloid fibril deposition in the vasculature can cause jaw claudication, lower limb claudication and angina (due to small vessel disease) (8). As the disease progresses, a low and fixed cardiac output state is seen which can result in exertional syncope and this has a very poor prognosis (9).

Characteristically, the 12-lead electrocardiograph (ECG) in patients with cardiac amyloidosis usually demonstrates low voltage QRS complexes ($<5\text{mm}$) in the limb leads. This is a result of extracellular amyloid fibril deposition causing “pseudo-hypertrophy” with apparent increase in left ventricular (LV) wall thickness but not of myocardial mass (10). Other common electrical abnormalities seen in cardiac amyloidosis include atrial fibrillation (AF) as well as conduction disease such as atrio-ventricular block and bundle branch block (6).

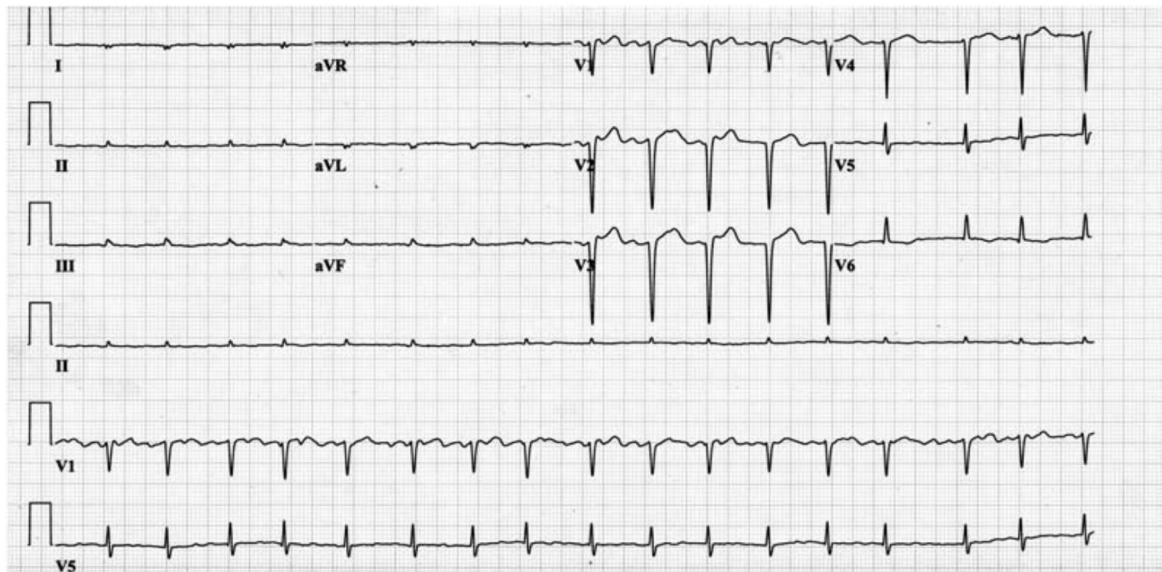


Figure 1. A 12-lead ECG showing low voltage QRS complexes in the limb leads for a patient with cardiac amyloidosis who had a septal LV wall thickness of 16mm and a posterior LV wall thickness of 17mm. From "*Electrocardiographic findings in primary systemic amyloidosis and biopsy-proven cardiac involvement.*" (Murtagh et al. (2005); Am J Cardiol 95(4): 535-537).

The transthoracic echocardiogram (TTE) shows evidence of LVH (measured as increased LV wall thickness) (6). More advanced echocardiographic techniques such as strain analysis can help in differentiating different forms of hypertrophic cardiomyopathy (HCM) (11). Strain is a dimensionless measure of the change in tissue length relative to its initial length i.e. $(\text{final length} - \text{initial length}) / \text{initial length}$ (12). Therefore, contraction is depicted by negative strain values and relaxation by positive strain values.

Echocardiographic strain analysis involves tracking "ultrasonic" speckles to assess regional deformation (13). The best evaluated strain parameter is global longitudinal strain (GLS) which is calculated by averaging the longitudinal deformation of 6 segments in each of 3 apical views (4-chamber, 2-chamber and 3 chamber/long-axis) (14). Cardiac amyloidosis often shows a pattern of relative apical sparing of longitudinal strain (LS), defined as a value of > 1.0 for average apical longitudinal strain (LS)/(average basal LS +

mid-LS) (15). This finding has a sensitivity of 93% and specificity of 82% for detecting cardiac amyloidosis (15).

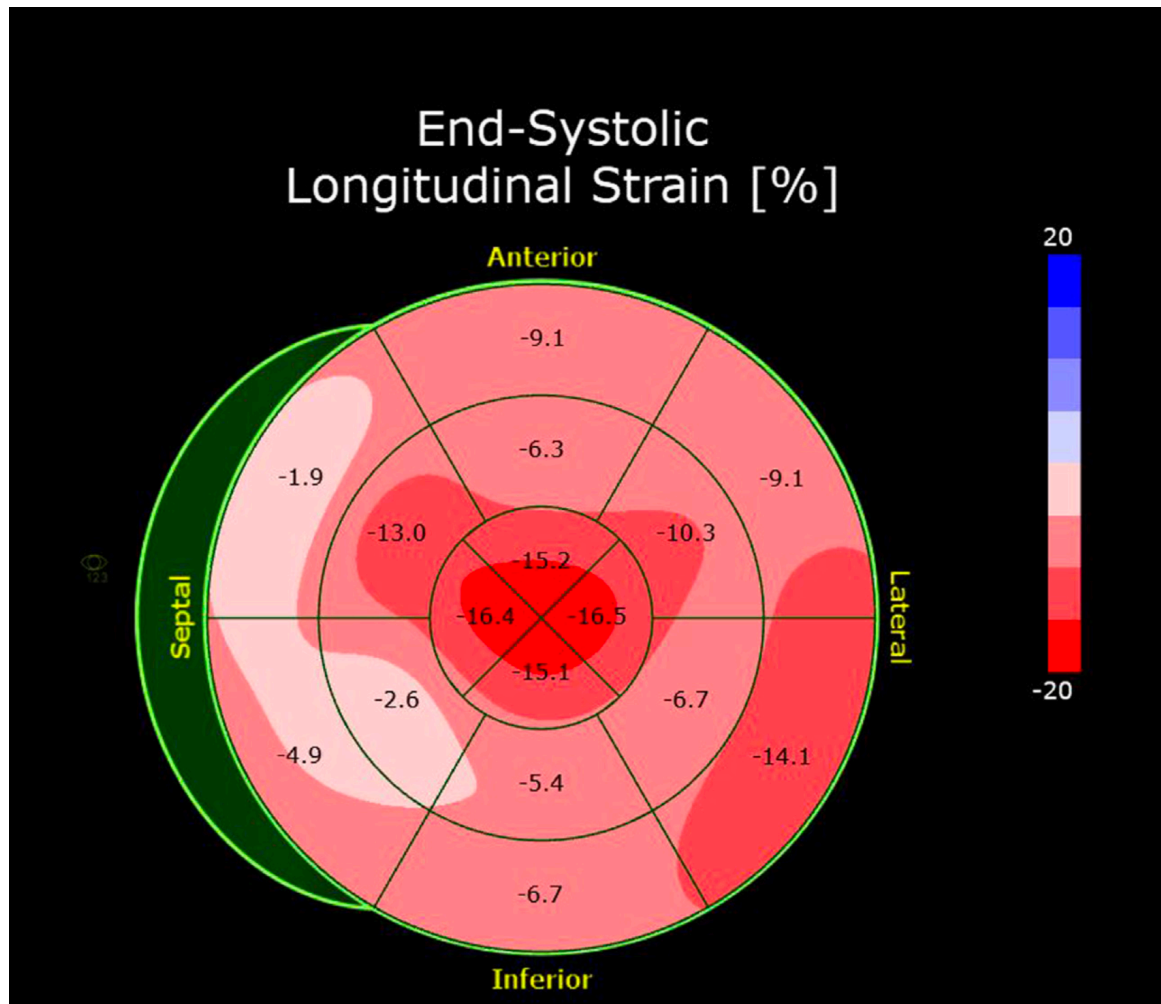


Figure 2. Demonstration of reduced regional strain in the basal and mid segments of the LV (pale colour), with relative sparing of the apical segments (darker colour). From *"Echocardiography in Infiltrative Cardiomyopathy"* (Perry et al; Heart Lung Circ. 2019 May 10; 28(9): 1365-1375).

Cardiac MRI can help in differentiating different forms of HCM, particularly with the use of a contrast agent (gadolinium) – late uptake of gadolinium, or late gadolinium enhancement (LGE), correlates with myocardial fibrosis or scar and in the context of cardiac amyloidosis, this is secondary to extracellular amyloid fibril deposition (16). The

pattern seen on cardiac MRI in cardiac amyloidosis is typically global subendocardial or transmural LGE (typically seen as the disease progresses with more infiltration), and often involves the atrium – which is rarely seen in other cardiac diseases; LGE, if present, correlates with biopsy-proven amyloidosis as well as mortality (16, 17).

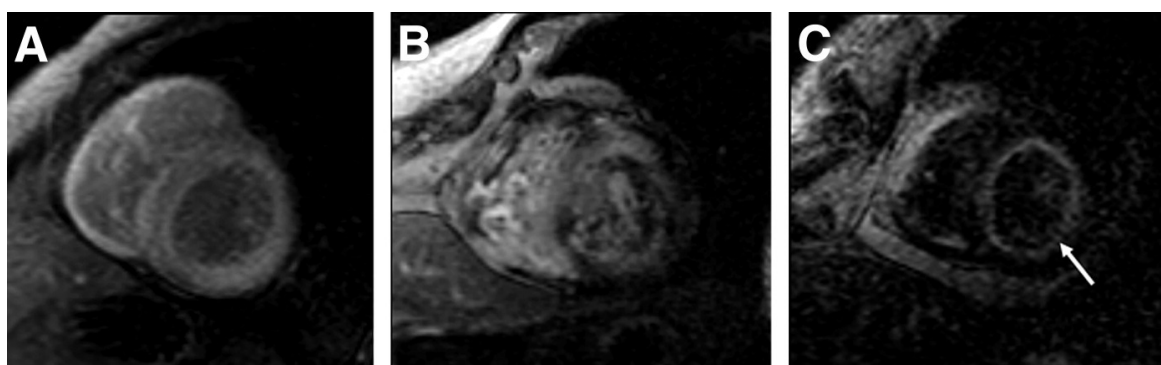


Figure 3. LGE patterns on cardiac MRI seen in cardiac amyloidosis from 3 different patients: (A) global transmural LGE in a **homogenous** pattern in the left and right ventricles; (B) global transmural LGE in a **heterogenous** pattern in the left and right ventricle; (C) global subendocardial LGE (arrow). From "Role of cardiac magnetic resonance imaging in the detection of cardiac amyloidosis." (Syed et al. 2010; JACC Cardiovasc Imaging 3(2): 155-164).

Additionally, T1 time, which is a quantitative measurement of T1 relaxation time either pre- or post-contrast, is prolonged in cardiac amyloidosis (18). Also, the temporal order of nulling on inversion time (TI) scout images can identify cardiac amyloidosis (19). In normal subjects, the nulling pattern shows nulling of the blood pool followed by nulling of the myocardium (which coincides with nulling of the spleen). However, if nulling of the myocardium occurs prior to the blood pool and does not coincide with nulling of the spleen, then this pattern is 70% sensitive and 100% specific for the diagnosis of cardiac amyloidosis (19).

1.2.1 Amyloid Light Chain (AL) Amyloidosis

AL amyloidosis is caused by proliferation of an abnormal monoclonal plasma cell population that causes the over production of lambda light chains (or less commonly kappa light chains) (20). Unlike multiple myeloma, the manifestations of the disease are due to the amyloid fibril deposition in organs secondary to the abnormal circulating free light chains (20). It is approximately one-tenth as common as multiple myeloma with an estimated annual incidence of 10.5 per million person-years (21), most often presenting from the fifth to the seventh decade of life. Various organs can be affected – most commonly the kidneys (causing nephrotic syndrome) and the heart (with more than 50-60% of patients with AL amyloidosis developing cardiac involvement), but other organs can also be involved including the liver and gastrointestinal system, the peripheral and autonomic nervous system, the vascular system and the soft tissues (20).

In addition to the features of heart failure described earlier in cardiac amyloidosis, other clinical features in AL amyloidosis include macroglossia, anasarca (due to nephrotic syndrome) as well as peripheral neuropathy (20). Prompt diagnosis of cardiac involvement in AL amyloidosis is imperative as the condition has a rapidly progressive course with median survival of less than 8 months after the onset of symptoms of cardiac failure (21, 22). With therapy, remission can be achieved, and prognosis can be extended to many years (20).

Standard laboratory tests can be unrevealing, however, if AL amyloidosis is suspected, then serum free light chains (to detect an abnormal kappa/lambda ratio) as well as

serum and urine electrophoresis should be performed (with immunofixation as this increases sensitivity) to detect a monoclonal band (20). Diagnosis is usually confirmed with a bone marrow or abdominal fat pad biopsy (90% sensitivity) which is positive for congo red staining with apple green birefringence under polarized light; but if negative, biopsy of other potentially involved organs can be performed, for example a biopsy of the kidney (23). High sensitivity troponin is often elevated at a chronically low level as well as the heart failure marker N terminal pro-brain natriuretic peptide (NT-proBNP) (20).

TTE findings in AL amyloidosis are similar to those described above for cardiac amyloidosis in general, with increased LV wall thickness, diastolic dysfunction and relative apical sparing of GLS. Additionally, in AL amyloidosis, impairment in basal LS has been shown to be an independent predictor of mortality (24) with GLS worse than -11.78% predicting mortality (25).

There are two goals of treatment for AL amyloidosis: firstly, to treat heart failure; and secondly, chemotherapy targeted at abnormal plasma cells responsible for production of light chains. With regards to heart failure therapy, angiotensin converting enzyme inhibitors (ACEi) and angiotensin receptor blockers (ARB) are poorly tolerated haemodynamically as they can exacerbate hypotension (26). Beta blockers can cause bradycardia but could be considered in the context of AF with rapid ventricular response. Digoxin use should be limited as it can bind to amyloid fibrils and induce digoxin toxicity (27). Pacemakers are indicated as per standard guidelines and are often required due to

atrioventricular block as well as sinus node dysfunction. However, a role for prophylactic automated implantable cardioverter-defibrillators (AICD) has not been established as patients with AL amyloidosis most commonly have cardiac arrest secondary to sudden bradycardia or pulseless electrical activity (28).

Chemotherapy regimens often used involve the proteasome inhibiting agent bortezomib in combination with cyclophosphamide and dexamethasone as well as the novel monoclonal antibody daratumumab which targets CD38+ plasma cells (29, 30). For patients who are eligible and who have a low risk of transplant related mortality, an autologous stem cell transplant can be considered up front as this can often induce a complete haematological response (31). If detected at an advanced stage, median survival can be as short as 4-6 months; however, if detected early with limited organ involvement (particularly cardiac), current therapy can improve prognosis to 5-10 years (32).

1.2.2 Transthyretin (TTR) Amyloidosis

As described earlier, TTR amyloidosis is caused by misfolding of the TTR protein to form amyloid fibrils. The TTR protein is synthesised predominantly in the liver and transports thyroxine and other proteins in the blood stream (33). The more common 'wild type' form, ATTRwt, also described as systemic 'senile' amyloidosis as it predominantly occurs later in life, is caused by an increase in the accumulation of wild-type TTR protein. ATTRwt almost always causes cardiac amyloidosis and it occurs more commonly in males (33). It is increasingly being recognised as a common form of amyloidosis involving

elderly males, as autopsy studies in elderly subjects (85 years and older) have shown evidence of cardiac amyloid deposits in up to 25% of cases, with milder forms commoner in females and more severe forms commoner in male subjects (34). TTR amyloidosis can also be a result of mutant TTR amyloidosis, i.e. ATTRv, also known as variant, familial or hereditary TTR amyloidosis, caused by a mutation in the TTR gene leading to misfolding of the protein (4). Cardiac involvement depends on the mutation (35). There are over 100 known mutations, with the most common being Val50Met (which has both cardiac and neurological manifestations), whilst there are mutations that have predominantly cardiac involvement only (36).

In addition to the features of heart failure seen in all forms of cardiac amyloidosis, other clinical features seen more often in TTR amyloidosis are a predisposition to develop carpal tunnel syndrome (generally a decade prior to the onset of cardiac manifestations) as the TTR protein has a tendency for depositing in tenosynovial tissue (37). In comparison to AL amyloidosis, low-voltage QRS complexes are far less common in TTR amyloidosis.

On cardiac MRI, features which may suggest a diagnosis of TTR amyloidosis more so than AL amyloidosis are that patients with TTR amyloidosis more frequently demonstrate transmural LGE (90% vs 37%), and LV mass is frequently much higher (average of 228g vs 167 g) (38). Despite this, symptoms in TTR amyloidosis seem to be less severe than that seen in AL amyloidosis (39).

Radioactive bone tracers such as ^{99m}Tc -labelled 3,3-diphosphono-1,2-propanodicarboxylic acid ($^{99m}\text{TcDPD}$), ^{99m}Tc -labeled pyrophosphate ($^{99m}\text{TcPYP}$), and ^{99m}Tc -labeled hydroxymethylene diphosphonate ($^{99m}\text{TcHMDP}$) are avidly taken up by hearts infiltrated by TTR, where grade 2 or 3 radiotracer uptake occurs in greater than 99% of patients with TTR amyloidosis (this is in contrast to AL amyloidosis where there is no uptake in the majority but can be minimal/grade 1 uptake in ~20%) (39, 40). Additionally, even though 99% of patients with AL amyloidosis have evidence of a monoclonal protein, up to 25% of patients with TTR amyloidosis may also have an unrelated monoclonal gammopathy of unknown significance (MGUS), which may lead clinicians to pursuing a diagnosis of AL amyloidosis (41, 42). Therefore, patients with no evidence of a monoclonal protein in serum or urine, and who have a positive bone scintigraphy scan, the specificity and positive predictive value can approach 100% for the diagnosis of TTR amyloidosis (40). In such cases, no tissue biopsy is required. However, if there is any uncertainty surrounding the diagnosis, tissue biopsy is vital to confirm the diagnosis (42).

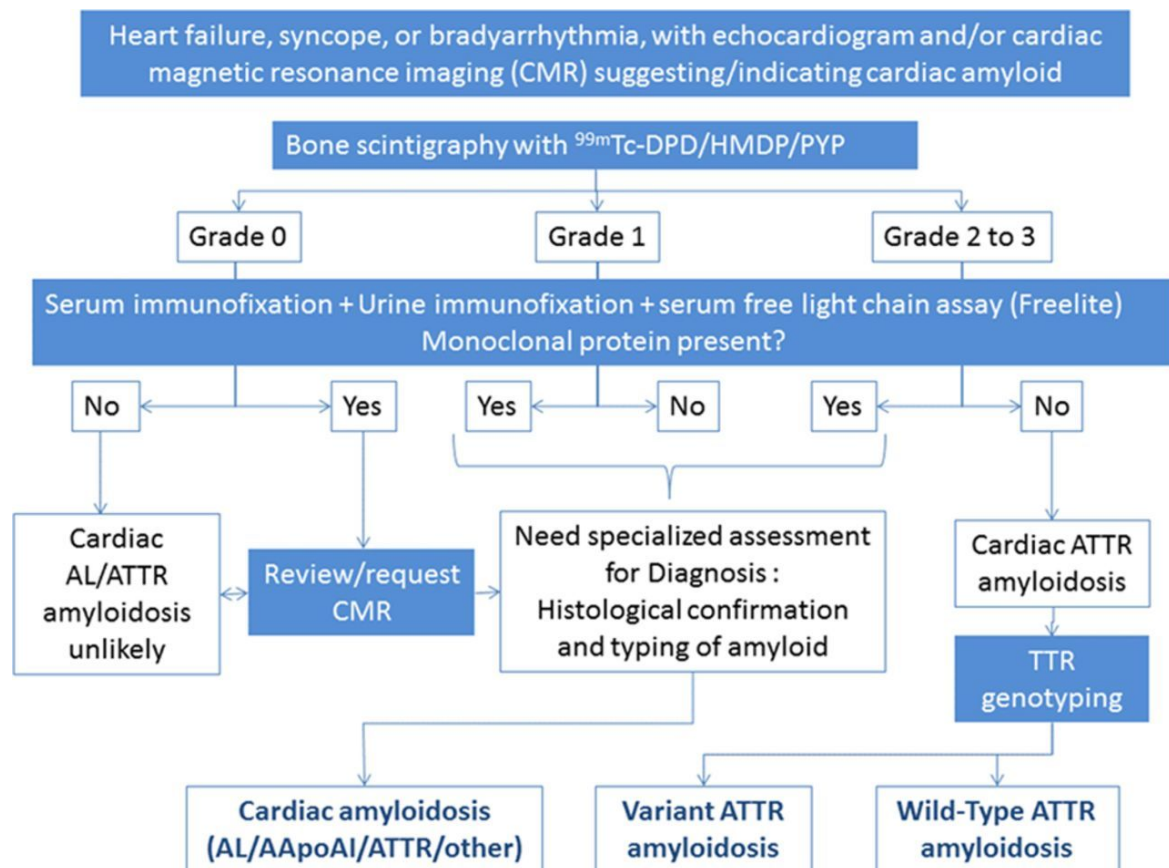


Figure 4. Diagnostic algorithm for patients with suspected cardiac amyloidosis. From “Nonbiopsy Diagnosis of Cardiac Transthyretin Amyloidosis” (Gillmore et al; Circulation. 2016 Jun 14;133(24):2404-12).

In terms of therapy for patients with TTR amyloidosis, the mainstay of treatment for patients with symptoms of heart failure or fluid overload is diuretics including loop diuretics and aldosterone antagonist therapy (33). ACEi, ARB’s and beta blockers are tolerated poorly in TTR amyloidosis, and there is no evidence of prognostic benefit with their use (33).

Several non-specific therapies have been investigated which inhibit TTR amyloid fibril formation or which stabilise the TTR protein in vitro including green tea extract, diflunisal and doxycycline; however, the evidence of clinical benefit is modest (43, 44, 45). More

recently, randomised trials have demonstrated tafamidis, a specific TTR stabiliser, reduces amyloid fibril formation, improves functional capacity and quality of life, but also reduces mortality and hospitalisations (46). There are several other investigational therapies in active trials where cardiovascular outcomes have not yet been established. These therapies, such as patisiran, target TTR ribonucleic acid and thus inhibit TTR synthesis therefore reducing the formation of amyloid fibrils (47).

1.3 Sarcoid cardiomyopathy

Sarcoidosis is a granulomatous disease that can target any organ of the body, including the heart (48). The aetiology is unknown, but granulomas form secondary to a postulated immune response to a yet unknown antigen in a person who is genetically susceptible (49). The typical targeted organ is the lung, where pulmonary sarcoidosis results in decades of fibrotic accumulation and subsequent respiratory failure and death (50). However, if granulomas form in the heart, they can be acutely life-threatening secondary to heart failure, heart block and malignant arrhythmias (51). As such, even though respiratory failure is the most common cause of death in patients with sarcoidosis, the acuity of cardiac sarcoidosis is more concerning. There is variability in how often the heart is involved and ethnicity tends to be a determinant. For example, in Japan, autopsy studies have shown that sarcoidosis can involve the heart in as up to 68% of patients with sarcoidosis, whereas in Northern America, the heart is less commonly affected with only 21% of African Americans and 14% of Caucasians having evidence of cardiac involvement on autopsy (52).

Cardiac sarcoidosis has a multitude of clinical presentations. Firstly, patients with cardiac sarcoidosis can be asymptomatic. Other presentations include heart failure, palpitations presyncope and syncope (48). Ventricular tachycardia (VT) in cardiac sarcoidosis occurs most often secondary to re-entrant scar-based circuits (53). Atrial arrhythmias are common and are caused by granulomatous scar formation as well as diastolic dysfunction and pulmonary hypertension (54). Conduction disturbances such as bundle branch block (both left and right) and complete atrioventricular block are also quite common – this is a result of granuloma-induced scar formation around the conduction system (55). In one study, 25% of patients less than 55 years of age presenting with high grade atrioventricular block, were diagnosed with cardiac sarcoidosis (56).

There is interest in screening patients with extra-cardiac sarcoidosis for cardiac involvement. ECG patterns which may indicate cardiac sarcoidosis include fragmentation of the QRS complexes and bundle branch block (57). Findings on echocardiography which might suggest cardiac involvement include reduction in LV GLS, reduction in LV systolic function, changes in LV wall thickness (including aneurysm formation and thinning of the basal septal wall), as well as other regional wall motion abnormalities (58).

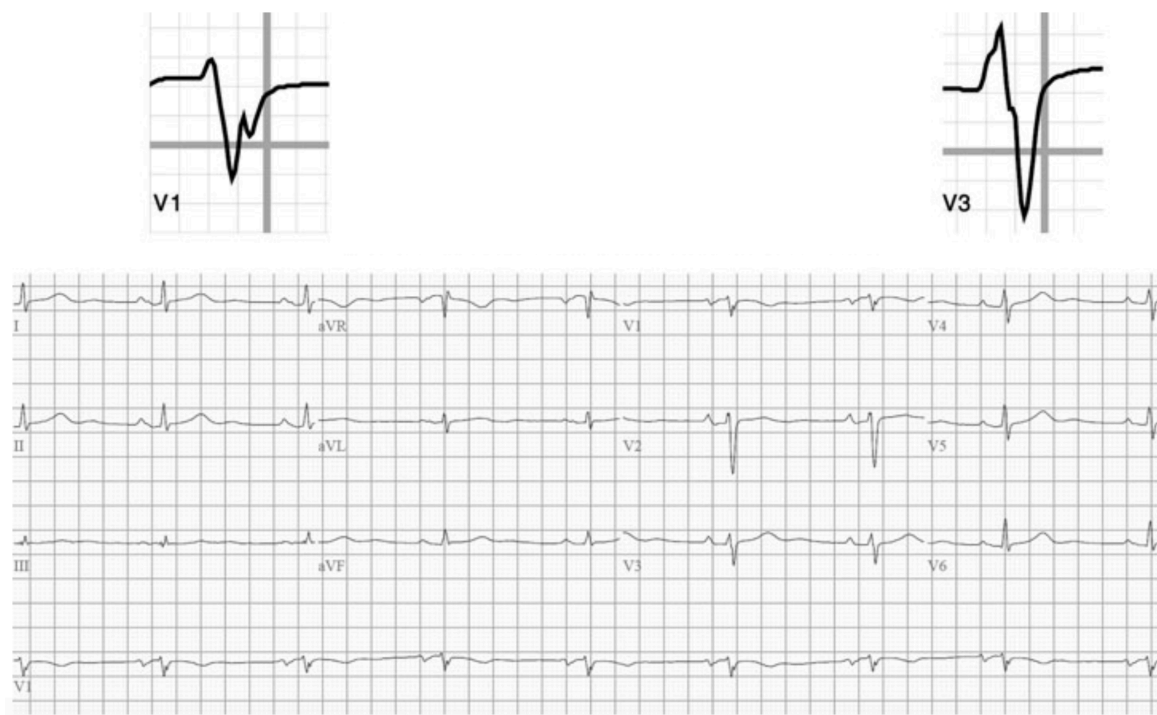


Figure 5. Demonstration of QRS fragmentation which can be seen in patients with sarcoidosis who have cardiac involvement. From Homsí, M., et al. (2009). *"Fragmented QRS complexes on 12-lead ECG: a marker of cardiac sarcoidosis as detected by gadolinium cardiac magnetic resonance imaging."* (Homsí et al. *Ann Noninvasive Electrocardiol* 14(4): 319-326).

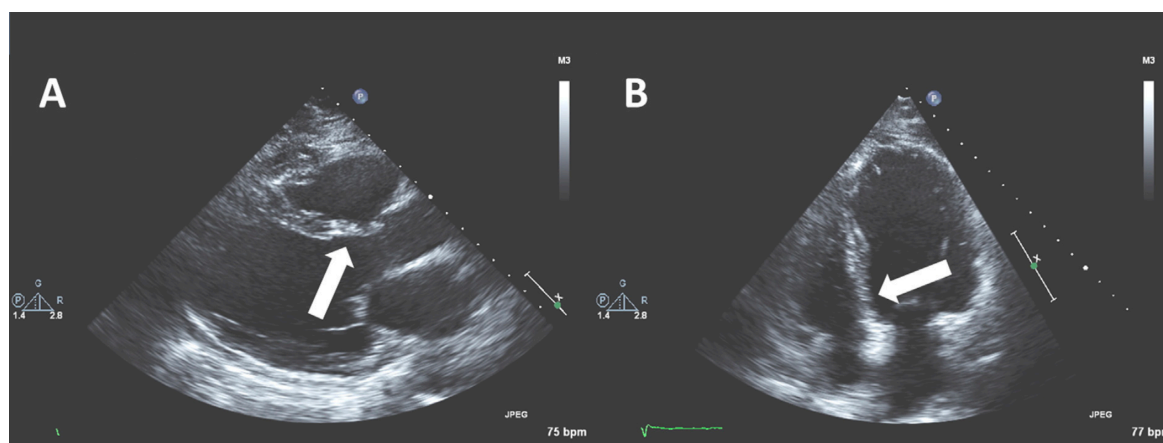


Figure 6. Demonstration of thinning of the basal interventricular septum seen in patients with cardiac sarcoidosis in the parasternal long axis (A) and 4-chamber (B) transthoracic echocardiography views. From *"Echocardiography in Infiltrative Cardiomyopathy"* (Perry et al; *Heart Lung Circ.* 2019 May 10; 28(9): 1365-1375).

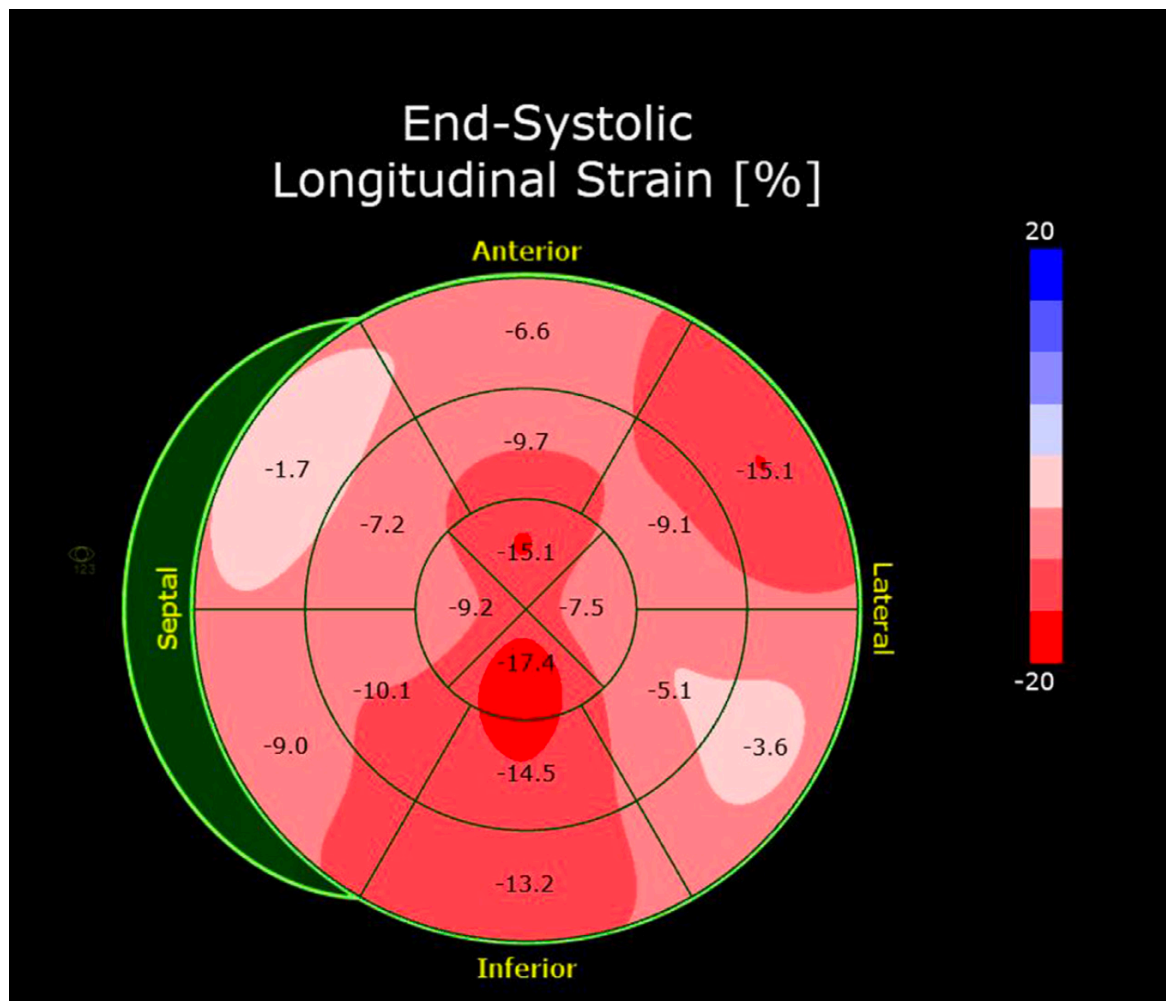


Figure 7. Demonstration of reduced GLS in a patient with cardiac sarcoidosis, with regional variation not corresponding to a coronary artery distribution. From *"Echocardiography in Infiltrative Cardiomyopathy"* (Perry et al; Heart Lung Circ. 2019 May 10; 28(9): 1365-1375).

The definitive diagnostic test for sarcoidosis is a biopsy of the affected organ, however, the diagnostic yield from a cardiac biopsy is quite low given granulomas form in a patchy distribution (59). Therefore, advanced cardiac imaging techniques such as cardiac MRI and ¹⁸Fluorodeoxyglucose positron emission tomography (¹⁸FDG PET) play a crucial role in the diagnosis of cardiac sarcoidosis. On cardiac MRI, a LGE pattern which is not consistent with a coronary artery distribution is suggestive of sarcoidosis (60). The degree of LGE is also predictive of malignant ventricular arrhythmias and death (61).

Active inflammation secondary to granulomatous disease can also be detected on cardiac MRI as T2 hyperenhancement (62). ^{18}F FDG PET makes use of a radioactive labelled glucose to identify active inflammation (63). In sarcoidosis, patterns of focal/patchy uptake (or patchy on diffuse uptake) are suggestive of active granulomatous disease (64).

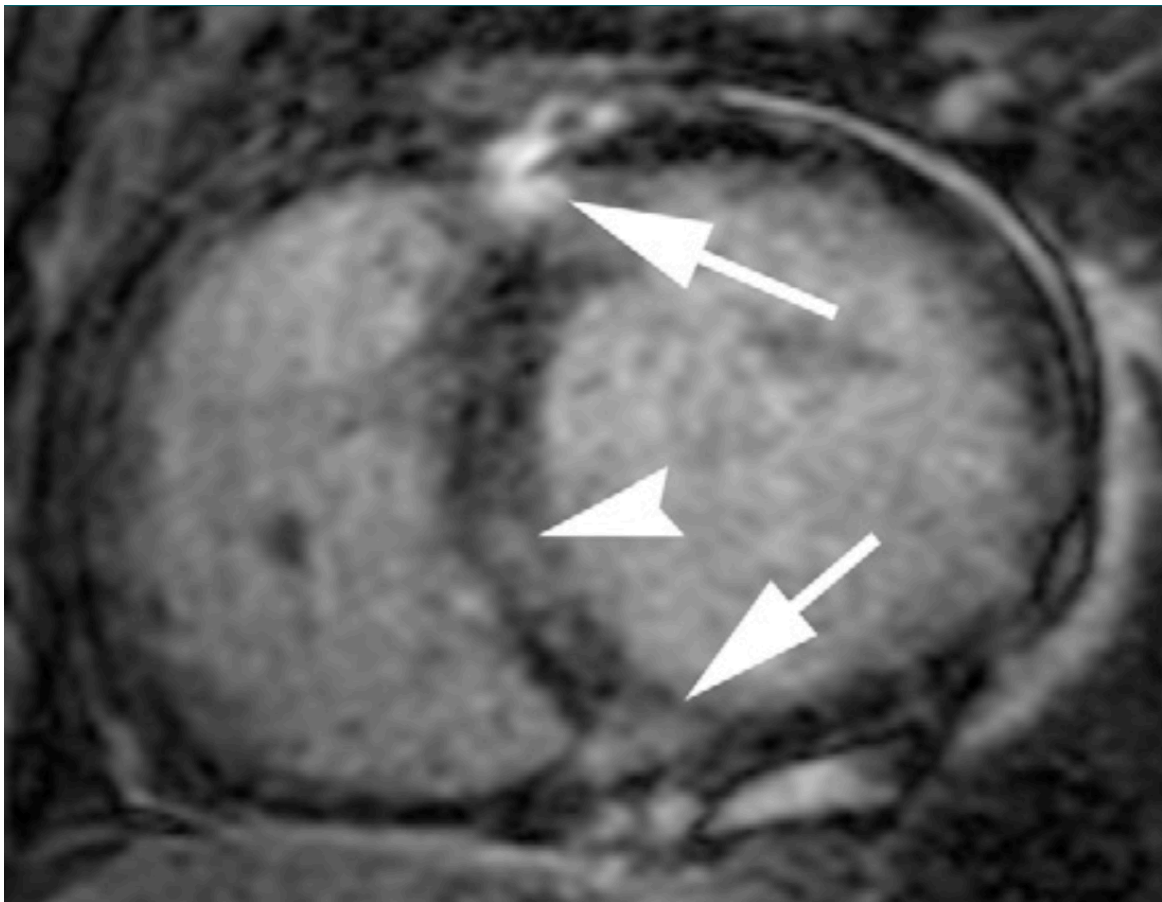


Figure 8. Demonstration of patchy LGE in a patient with cardiac sarcoidosis affecting the anterior and posterior interventricular septum (transmural LGE) as well as the centre of the interventricular septum (mid-wall LGE). From "*Delayed contrast enhancement on MR images of myocardium: past, present, future.*" (Ordovas et al 2011; Radiology 261(2): 358-374).

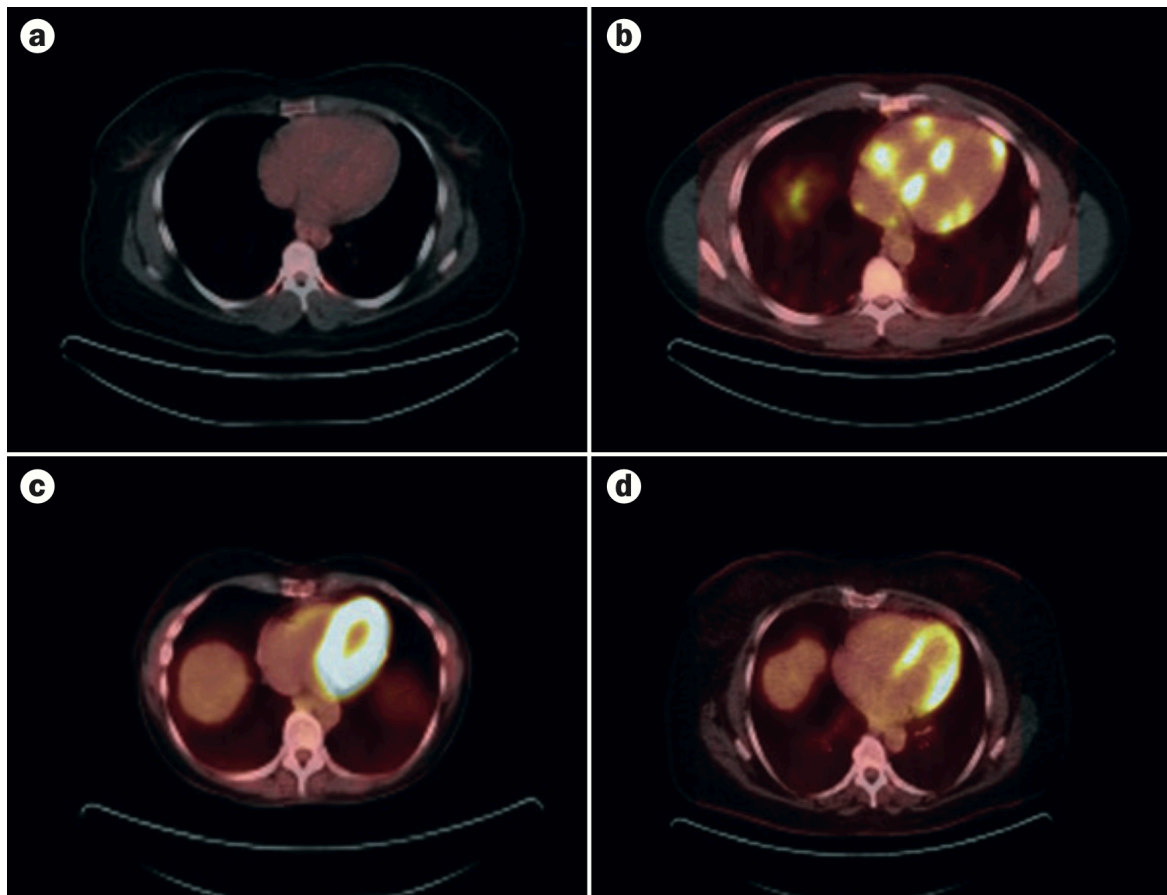


Figure 9. Demonstration of patterns seen on ^{18}F FDG PET: (a) no uptake; (b) patchy uptake; (c) diffuse uptake; (d) patchy on diffuse uptake. Granulomatous disease secondary to cardiac sarcoidosis often show a patchy (b) or patchy on diffuse (d) uptake pattern. From "Pathophysiology and clinical management of cardiac sarcoidosis." (Hamzeh et al 2015; Nat Rev Cardiol 12(5): 278-288).

The Heart Rhythm Society Expert Consensus Statement (65) suggests a diagnosis of *probable* cardiac sarcoidosis for patients with histological evidence of extra-cardiac sarcoidosis with the following cardiac manifestations (so long as other causes of these manifestations have been excluded):

- unexplained heart failure with reduced ejection fraction (less than 40%);
- unexplained sustained VT;
- high degree atrioventricular block (Mobitz type II second degree or third degree);

- cardiac ¹⁸FDG-PET with patchy uptake in a pattern which is consistent with cardiac sarcoidosis;
- cardiac MRI with LGE in a pattern consistent with cardiac sarcoidosis

The treatment of cardiac sarcoidosis is two-fold: firstly, immunosuppressive therapy targets active granulomatous myocarditis to prevent the progression of granulomatous disease into scar tissue; secondly, cardiac complications, such as heart failure, arrhythmias and conduction disease require cardiac specific treatment. There is a lack of guidelines and evidence as to when it is best to institute immunosuppressive therapy, however, expert consensus is that immunosuppressive therapy should be instituted for patients with evidence of active cardiac inflammation detected on ¹⁸FDG PET scan, in whom a diagnosis of definite cardiac sarcoidosis (confirmed histologically with a cardiac biopsy) or probable cardiac sarcoidosis (as per the Heart Rhythm Society Expert Consensus Statement above) is made (65).

For patients with cardiac sarcoidosis who develop advanced atrioventricular block, corticosteroid therapy can revert this to normal sinus rhythm or a lower grade atrioventricular block (66, 67). Patients with normal systolic function have minimal active granulomatous disease and scar, but immunosuppressive therapy can prevent progression of disease and scar formation (49). For patients who have mild-moderate reduction in LV function, immunosuppressive therapy results in an improvement in LV function (68). Patients with severely reduced systolic function have a high burden of scar and are less likely to respond to immunosuppressive therapy (68). Management of LV

systolic dysfunction also involves instating contemporary therapies for heart failure with reduced ejection fraction, such as ACEi, beta blockers and mineralocorticoid receptor antagonists (49).

Patchy myocardial fibrosis forms the substrate for the development of VT resulting in patients requiring AICD. Anti-arrhythmic agents such as amiodarone and sotalol are initiated but are often ineffective for controlling VT. For example, a study demonstrated that 40% of patients with cardiac sarcoidosis on antiarrhythmic therapy for ventricular arrhythmia had breakthrough arrhythmia requiring AICD therapy (69). Therefore, AICD implantation has specific indications in patients with cardiac sarcoidosis. These include: (1) sustained spontaneous ventricular arrhythmias with LV ejection fraction less than 35% despite optimal medical therapy; (2) unexplained or near syncope; or (3) a positive electrophysiological study demonstrating inducible ventricular arrhythmia irrespective of LV ejection fraction (65). VT ablation plays a crucial role in the management of cardiac sarcoidosis. Electrophysiological studies often show evidence of numerous inducible VTs in each patient with cardiac sarcoidosis, and with ablation the VTs can be eliminated or VT burden can be reduced significantly (70, 71).

Finally, for end-stage heart failure or intractable VT, heart transplantation can be considered, however, prognosis is worse than for patients without sarcoidosis undergoing heart transplantation (5 year survival of 52.4 vs 76.2%) (72).

1.4 Lysosomal storage disorders

Lysosomal storage disorders (LSD) are inherited disorders whereby due to mutations, specific lysosomal enzymes, membrane transporters or proteins involved in lysosomal biology are altered resulting in accumulation of specific substrates that cannot be broken down (73). The disorders are classified depending on the accumulated substrate – e.g., glycogen storage diseases (Pompe and Danon disease), mucopolysaccharidoses and the sphingolipidoses (Fabry, Gaucher and Niemann-Pick diseases). These diseases can affect many organs but can importantly affect the heart with varying presentations including HCM, dilated cardiomyopathy, coronary artery disease and valvular disease.

2.1 Overview of Fabry Disease

FD is an X-linked lysosomal storage disorder caused by alpha-galactosidase A (GLA) enzyme deficiency and subsequent accumulation of glycosphingolipids.

2.1.1 Aetiology and pathophysiology of Fabry Disease

The GLA gene that encodes the enzyme is 7 exons in length and is located on the X-chromosome (Xq22.1). Mutations affecting the GLA gene are heterogenous including missense mutations, large gene rearrangements as well as small deletions or insertions, and splice mutations, with over 900 mutations being identified that cause FD (74). The pathology results from the accumulation of glycosphingolipids (globotriaosylceramide i.e. Gb3, which is also abbreviated as GL-3, and its deacylated derivative globotriaosylsphingosine i.e. lyso-Gb3, which is also abbreviated to lyso-GL-3) in various organ tissues including the vascular endothelium (75). As it is inherited in an X-linked pattern, it was initially thought that females were asymptomatic carriers; however, it is now recognised that female patients with FD can develop severe disease, although the disease manifests at a later age and with slower progression compared to males (76).

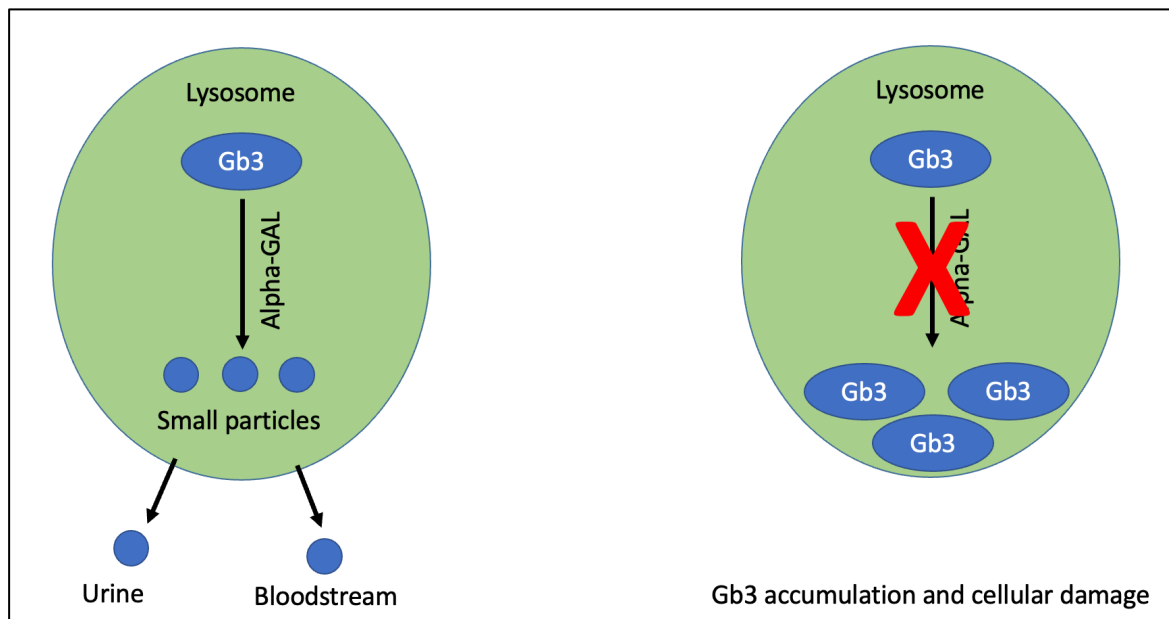


Figure 10. Pathophysiology of Fabry Disease – deficiency in the lysosomal enzyme GLA results in accumulation of its substrate Gb3.

2.1.2 Prevalence of Fabry Disease

The incidence of FD is reported to be in the range of 1 in 40,000-117,000; however, given it is an under-recognised condition, the incidence may be significantly higher (77). Some studies screening for the disease in newborns, estimate the incidence could be higher with rates as high as 1 in 3100 newborns in Italy, and rates as high as 1:1600 male newborns in Taiwan, although these studies included likely benign variants of the GLA gene (78, 79).

2.1.3 Clinical features of Fabry Disease

Because FD is a rare condition (with lack of awareness amongst most physicians) and patients with FD present with signs and symptoms that are non-specific, the mean time to diagnosis of FD is 15 years (80). FD can affect the dermatological, ophthalmological,

neurological, gastrointestinal, renal and cardiac systems, with the heart being the organ most frequently affected (81). Although symptoms consequent to FD can be present in childhood, cardiac involvement is often not detected until the third or fourth decade of life, with males affected earlier due to the X-linked nature of the disease (82).

There are several signs which should alert a clinician to the diagnosis of FD. A family history of nephropathy, cryptogenic stroke and LVH or HCM, inherited in an X-linked pattern should heighten suspicion for FD. As such, screening for FD usually occurs in patients presenting with end stage renal failure, unexplained LVH or cryptogenic stroke. Prevalence of FD in patients presenting with prior diagnosis of HCM can range from 3.9-12.0% (83, 84), is between 1-5% in patients presenting with cryptogenic stroke (85, 86) and can be up to 1% in patients with end stage renal failure (87).

Childhood and adolescent symptoms in male patients with FD can come on as early as 5-10 years of age and include (81, 88):

- acroparesthesia – manifesting as pins-and-needles, burning or numbness in the hands and feet, but particularly the fingers and toes
- angiokeratomas – small, dark raised lumps on the skin surface due to capillary ectasia, more so on the buttocks, groin and lower trunk; angiokeratomas are present in 66% of males and 36% of females with FD and the presence of these skin lesions correlates with the overall disease severity (including renal failure, cardiac disease and cerebrovascular disease) (89)

- hypohidrosis, heat/cold intolerance or exercise intolerance due to autonomic dysfunction
- gastrointestinal symptoms including nausea and diarrhoea

Other clinical features which develop later in the disease course (generally in the 3rd to 4th decade of life, but occasionally earlier) include

- stroke or transient ischaemic attack
- nephropathy and proteinuria
- cardiac manifestations including LVH
- corneal verticillata – fine golden-brown/grey opacities that stem out from a central whirl across the inferior aspect of the cornea
- lymphoedema – occurs in 16% of males and 6% of females and although cardiac failure and renal failure can exacerbate lower limb oedema, lymphoedema has been observed in their absence (89)

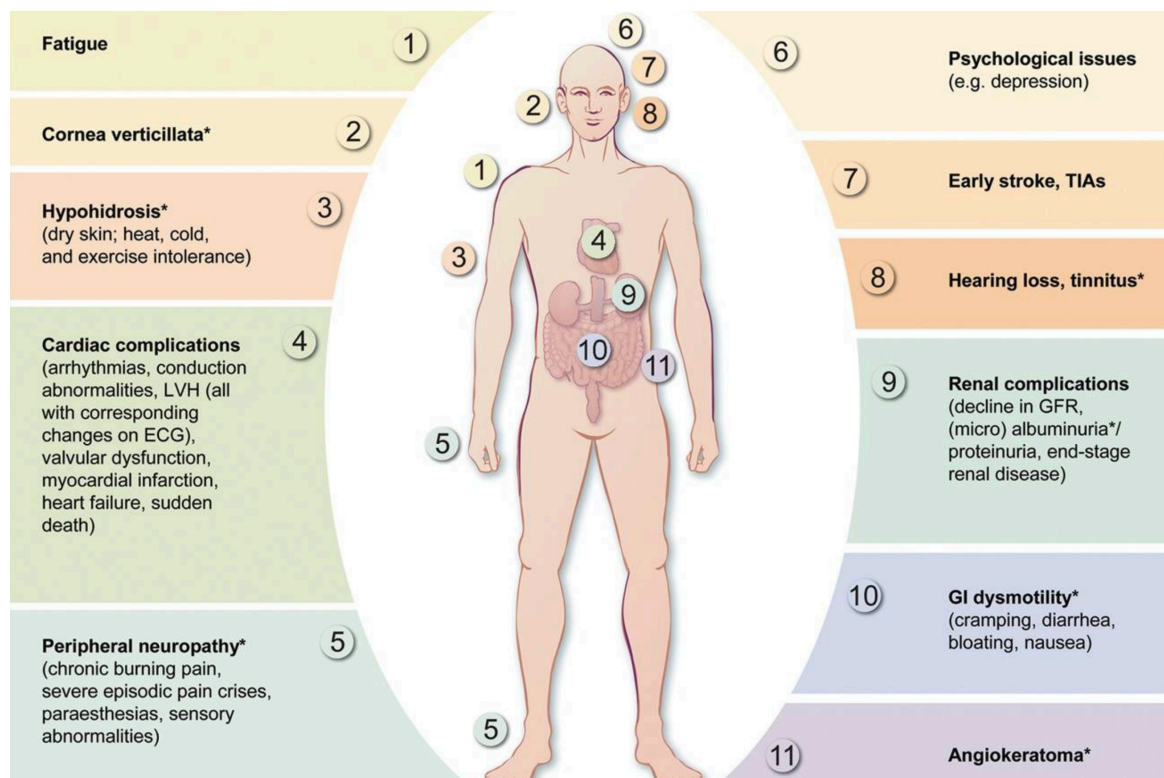


Figure 11. Diagrammatic representation of the clinical features of Fabry Disease. From *"Left ventricular hypertrophy in Fabry disease: a practical approach to diagnosis"* (Yousef et al; Eur Heart J. 2013 Mar 14; 34(11): 802–808).

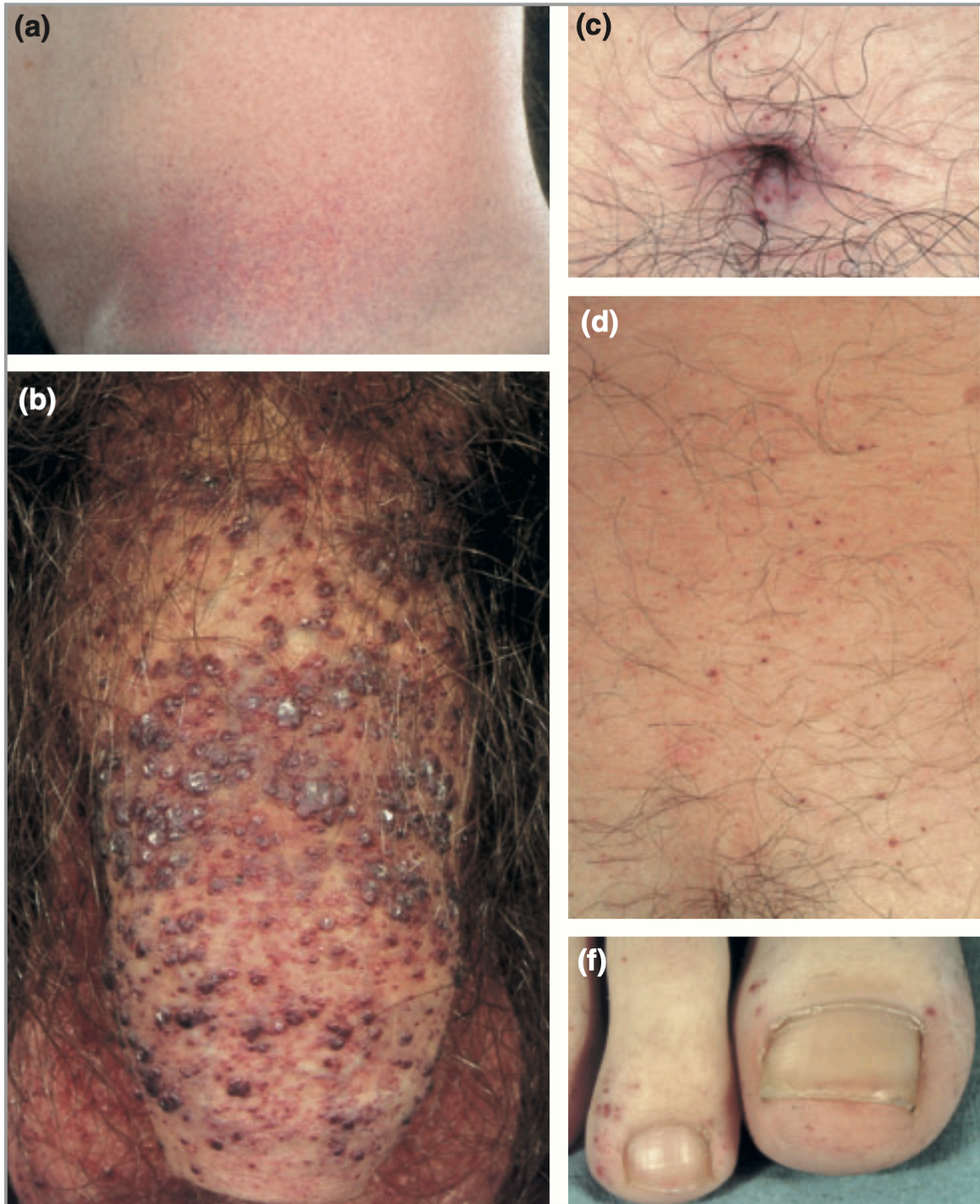


Figure 12. Angiokeratomas seen over the: (a) flank; (b) genitals; (c) umbilicus; (d) lower back; (e) toes. From *"Fabry disease and the skin: data from FOS, the Fabry outcome survey"* (Orteu et al; British Journal of Dermatology, Volume 157, Issue 2, 1 August 2007, Pages 331–337)

With regards to cardiac manifestations, the diagnosis of FD should be considered in the case of unexplained LVH, particularly if the LVH is symmetric, concentric, homogenous or non-obstructive (77). In rare cases, hypertrophy can result in LV outflow tract obstruction (90). Other cardiac manifestations will be discussed later but can include:

- ECG / Holter monitor abnormalities including atrio-ventricular block and arrhythmias
- Echocardiographic abnormalities including reduction in GLS (91) and thinning of the basal inferolateral LV wall, thought to be secondary to fibrosis in advanced disease – this finding is associated with chronic heart failure & cardiac death (92)
- Cardiac MRI findings of postero-lateral LGE, reduction in non-contrast T1 signal and elevation in T2 signal

2.1.4 Diagnosis of Fabry Disease

When FD is suspected, biochemical and genetic testing should be undertaken to confirm the diagnosis. Assays are available to test activity of GLA in dried blood spots or in leukocytes – significantly diminished activity will usually confirm the diagnosis in men, but activity levels may be normal in females (77). Raised levels of the accumulated glycosphingolipids (Gb3 and lyso-Gb3) can also be analysed in plasma or in urinary samples. However, as expected levels are usually lower, but can even be normal in women. Additionally, in patients with certain mutations of the GLA gene, levels may not be elevated (77). Therefore, the diagnosis in suspected cases should be confirmed with genetic analysis of the GLA gene.

2.1.5 Cardiac manifestations of Fabry Disease

In some patients with specific mutations, the heart can be the only organ involved (cardiac variant of FD) (93). Similarly, in female heterozygous carriers with near normal enzymatic activity, often the heart is the only organ affected, albeit with delayed onset and slower progression compared to their male counterparts (94, 95).

Cardiac involvement is characterized by progressively increasing LV wall thickness due to intracellular accumulation of glycosphingolipids, interstitial remodelling and eventually, replacement myocardial fibrosis (90, 96). Therefore, patients with FD most often present with LVH without evidence of associated conditions that cause increased afterload, such as hypertension or aortic stenosis (80).

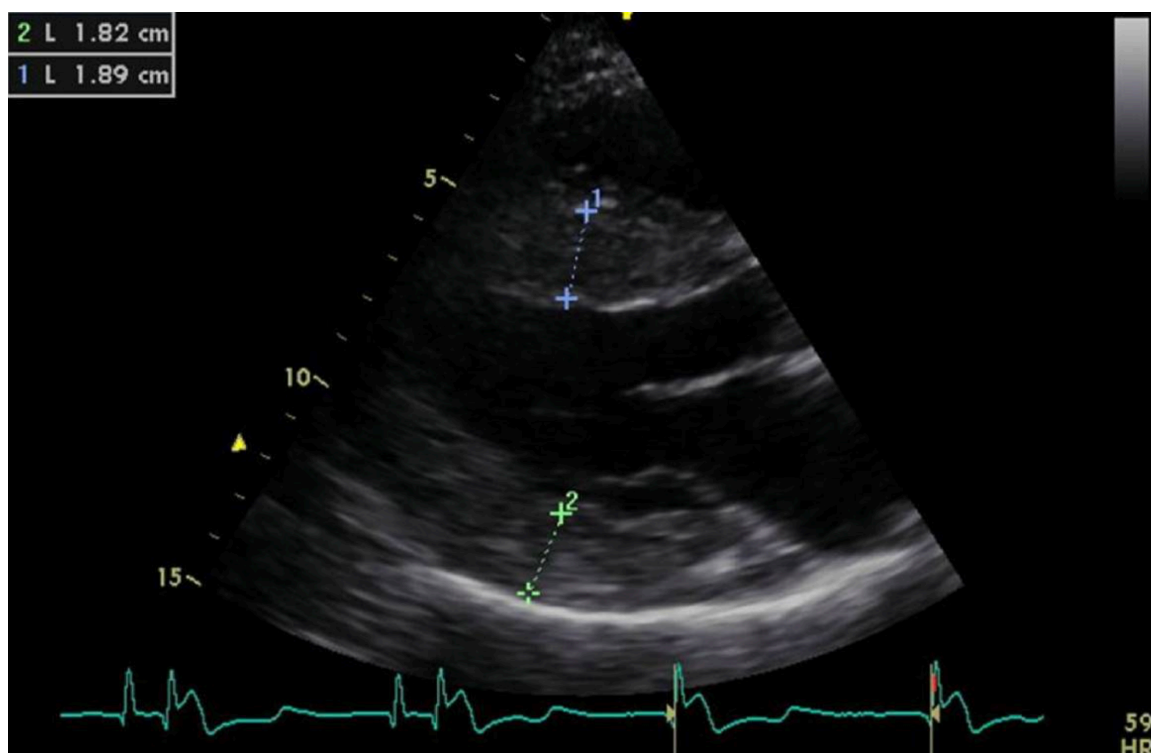


Figure 13. TTE parasternal long axis view of left ventricular hypertrophy in FD From "Fabry disease." (Germain 2010; Orphanet J Rare Dis 5: 30).

Cardiac involvement can lead to the development of significant life-threatening ventricular arrhythmias, AF as well as conduction defects (e.g. sinus node dysfunction, chronotropic incompetence or severe atrioventricular block) (82, 96, 97).

Cardiac symptoms include dyspnoea (due to diastolic heart failure), exertional angina (due to microvascular dysfunction), palpitations (due to arrhythmias such as AF and non-sustained VT) and syncope (due to conduction abnormalities or ventricular arrhythmias) (98).

2.1.6 Risk stratification of Fabry Disease

Cardiovascular complications are the leading cause of death for patients with FD (in one study, 40% of patients died due to cardiovascular disease compared to < 10% due to neurological or renal disease) (99). In a recent systematic review (100) which included 4185 patients with FD with a follow-up period of 1.2 – 10 years, in the 8.3% of patients that died, 75% had cardiovascular causes and 62% were due to sudden cardiac death. Interestingly, in this review, the prevalence of VT was 15.3%. Furthermore, factors that were associated with sudden cardiac death included male sex, increasing age, LVH and evidence of LGE on cardiac MRI.

The presence and degree of LVH has been associated with cardiac morbidity and mortality (101). Other predictive factors for cardiovascular complications include increasing age, end-stage kidney disease, and prolonged QRS duration on ECG (102). Therefore, early diagnosis of FD is imperative as the development of LVH may be prevented or may regress with early treatment with enzyme replacement therapy (ERT), and may therefore reduce the cardiovascular morbidity and mortality associated with the disease (103).

3.1 Cardiac evaluation of Fabry Disease

3.1.1 Electrocardiographic evaluation

In patients with FD, the ECG is seldom normal. Patients with FD demonstrate electrocardiographic changes including a prolonged PQ interval and QRS duration, as well as ECG features of LVH (104, 105, 106). Due to the X-linked inheritance pattern of the disease, ECG alterations are more likely to be present in men with FD as compared to women. Niemann et al evaluated the ECGs of 150 patients with FD (including 63 men and 87 women) and demonstrated that men were more likely to have broadened QRS, higher Sokolow-Lyon Index and Right Bundle Branch Block criteria (107).

3.1.1.1 PQ interval

Interestingly, there have been reports of young patients with early-stage FD demonstrating a shortened PQ interval without evidence of an accessory pathway (104, 105, 108). It is proposed that conduction velocity is increased as a result of accumulation of Gb3 in cardiomyocytes, an interaction between Gb3 and the ion channels involved in propagation of action potentials and due to the increase in cardiomyocyte diameter (104, 105). Furthermore, in glycogen storage disorders other than FD, PQ interval shortening may be related to glycogen storage within proximity to the atrioventricular node (109). With progression of FD, macroscopic changes to the myocardium (left atrial enlargement) results in prolongation of the PQ interval (104).

3.1.1.2 QRS interval

Similar to PQ interval shortening, young patients with early-stage FD also demonstrate QRS interval shortening, most likely secondary to enhanced conduction velocity (104, 105). As FD progresses, the QRS interval prolongs (106). This is thought to be due to the increased time taken for the left ventricle to depolarize as a result of LVH and increased LV mass (as seen in patients with LVH from any cause) (110, 111, 112, 113). As such, prior studies in FD have also demonstrated a correlation between prolonged QRS duration on ECG and LV mass seen on TTE (113) and cardiac MRI (107). The QRS prolongation is secondary to right bundle branch block more commonly than left bundle branch block (107, 114, 115). This may be a result of preferential accumulation of Gb3 in the right bundle branch as evidenced by vacuolization within myocytes and extension into the right bundle branch (116).

3.1.1.3 ECG for detecting Left ventricular hypertrophy

With disease progression in FD, LV wall thickness increases and this is associated with an increased Sokolow-Lyon index on ECG (113). Therefore, most male patients with advanced FD will meet Sokolow-Lyon LVH criteria on ECG.

3.1.1.4 Long duration electrocardiogram monitoring for detection of arrhythmias

Arrhythmias, both supraventricular and ventricular, can occur in FD even prior to patients developing LVH or other clinical signs (80, 117). Patients with FD are also at high risk of developing atrioventricular block, chronotropic incompetence and symptomatic bradycardia, with rates of antibradycardia pacing being more than 25 times that in the

general population (118). Furthermore, hypertrophy and myocardial fibrosis in patients with FD frequently manifests in patients developing VT, ventricular fibrillation and subsequent sudden cardiac death (119).

Therefore, frequent long-duration monitoring (e.g. annual 48 hour Holter monitoring) is recommended in guidelines as abnormalities are frequently detected resulting in altered management decisions (e.g. implantation of permanent pacemaker or AICD, initiation of anticoagulation for AF) (120). Incidence of an implanted cardiac device is as high as 18% with an annual incidence of 1.9% per year (121). Predictors of pacemaker or AICD implantation include severity of disease, myocardial fibrosis and patients with a late diagnosis or late initiation of treatment with ERT (122).

3.1.1.5 Electrophysiological testing

In asymptomatic patients with FD, there is no significant evidence of clinical or prognostic benefit in the routine use of an Electrophysiological Study (EPS) to assess the risk of malignant ventricular arrhythmias or sudden cardiac death (123). Additionally, as discussed previously, a short PR interval can be seen in FD (due to accelerated atrioventricular nodal conduction rather than an accessory pathway) and so in isolation, a short PR interval is not an indication for an EPS (124).

However, an EPS should be considered in patients with persistent or recurrent supraventricular arrhythmias such as atrioventricular nodal re-entry tachycardia,

accessory pathway mediated atrioventricular re-entry tachycardia, focal atrial tachycardia and atrial flutter (125). Similarly, an EPS should also be considered in patients with evidence of pre-excitation (i.e. presence of a delta wave on ECG). In such patients, ablation of the accessory pathway should be considered in the presence of symptoms such as syncope or palpitations (125).

3.1.2 Transthoracic Echocardiographic Evaluation

The key echocardiographic features in FD are increased LV wall thickness, left atrial enlargement, valvular thickening and mitral regurgitation (115).

3.1.2.1 Left Ventricular Size

Generally, LV volumes and diameters are within the normal range for patients with FD (115).

3.1.2.2 Left Ventricular Wall Thickness

LV wall thickness is increased in patients with FD and generally LVH is concentric (115). However, apical or septal asymmetrical hypertrophy with LV outflow tract obstruction have also been described in the literature and these variants may mimic the phenotypical and clinical features of sarcomeric or classical HCM (115, 126, 127). With worsening myocardial fibrosis, the basal infero-lateral LV segments can become thin and hypokinetic (92).

3.1.2.3 Left Ventricular Function

LV ejection fraction is usually normal in patients with FD despite an increase in LV wall thickness (91). Studies in patients with FD have tried to identify other markers of subclinical LV dysfunction. For e.g., Zamorano et al identified a reduction of tissue Doppler velocity (S' and e') at the mitral annulus as a marker of myocardial dysfunction in patients with FD without evidence of LVH (128). A more sensitive marker of subclinical LV dysfunction is myocardial LS which is an assessment of myocardial deformation utilising speckle tracking. FD patients have impairment in myocardial LS, despite having normal LV wall thickness, normal LV ejection fraction and prior to the onset of clinical cardiovascular manifestations (77, 91, 129). Therefore, impaired LS may be a marker of subclinical LV systolic dysfunction. As the disease progresses, with worsening LVH, extensive myocardial fibrosis and co-existent coronary artery disease, a reduction in LV systolic function can be seen (130).

Diastolic function can be normal early in the disease process. But as cardiac involvement worsens, diastolic parameters become abnormal (131). Elevated LV filling pressure (i.e. increased E/e' ratio) is associated with worse prognosis (102).

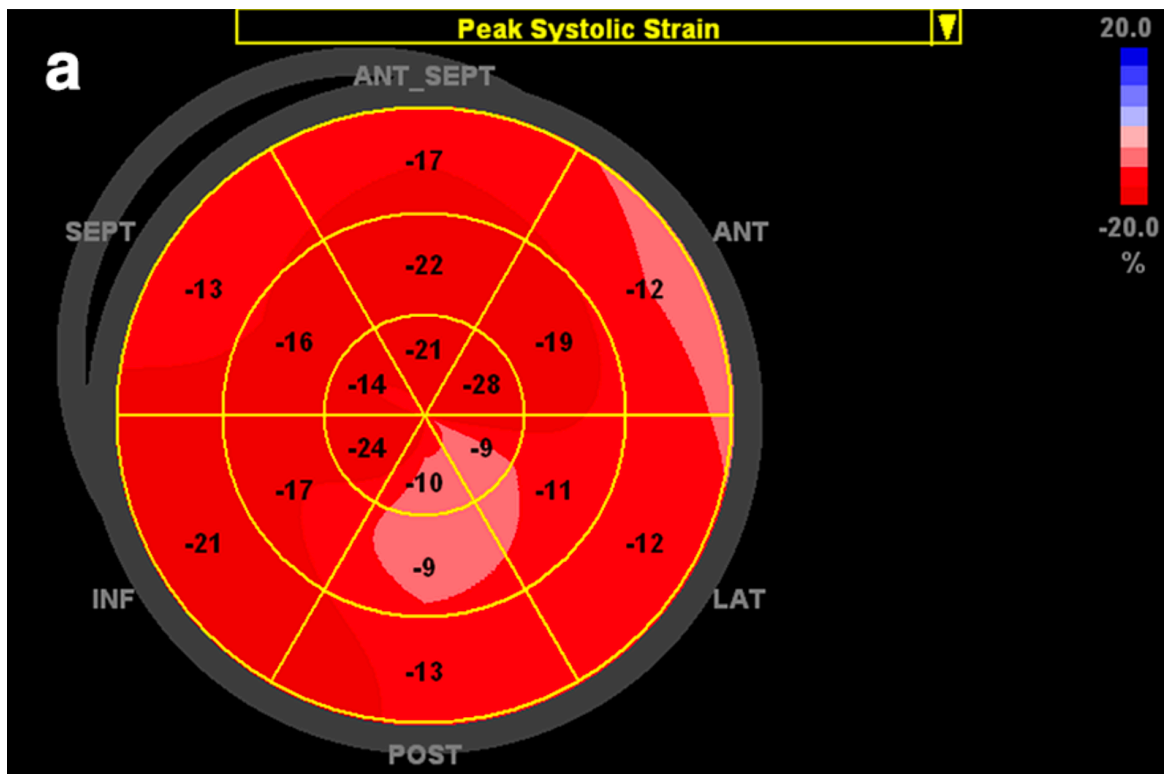


Figure 14. Demonstration of reduced GLS in a patient with FD and LVH. From: "Longitudinal strain bull's eye plot patterns in patients with cardiomyopathy and concentric left ventricular hypertrophy." (Liu et al. 2016; Eur J Med Res. 2016 May 10;21(1):21).

3.1.2.4 Left Atrial Volume

Alterations to the left atrium occur in FD as well. Left atrial strain is reduced prior to the development of left atrial enlargement (132). There is subsequent enlargement of the left atrium associated with reduced left atrial compliance (133). These changes can occur prior to the development of LVH (133).

3.1.2.5 Other echocardiographic features

The right ventricle is also affected in FD (134). In one study of 58% with proven FD, the right ventricle chamber size was normal, but right ventricular hypertrophy was present in 40%. Systolic function was preserved, however diastolic function was impaired. The

prevalence and degree of right ventricular involvement was related to the prevalence and degree of LVH as well as the age of the patient.

With regards to valvular disease in FD, there is often mild-to-moderate aortic and mitral regurgitation with leaflet thickening. In a minority of patients, the mitral regurgitation can be severe and may require surgical correction (135). Patients with FD very rarely have stenotic valvular disease (136).

In advanced FD, there is often mild-to-moderate dilatation of the aortic sinus and ascending aorta, but the risk of aortic dissection is low (137).

3.1.3 Evaluation by Cardiac Magnetic Resonance Imaging

Cardiac MRI, although not readily accessible, can serve several purposes in the assessment of patients with FD. Firstly, it allows for an accurate estimate of LV size and mass. Additionally, with gadolinium contrast, it allows for the assessment of myocardial fibrosis (138). In patients with FD, Moon et al correlated LGE in the mid-myocardial layer of the inferolateral wall on cardiac MRI, with histological evidence of myocardial fibrosis (focal myocardial collagen scarring) (139). The presence of fibrosis is significant as it is associated with an increased risk of malignant arrhythmia and sudden cardiac death (119, 140). Furthermore, patients with FD can develop myocardial fibrosis prior to the development of significant LVH, particularly female patients, due to the X-linked inheritance pattern of the disease (141). Therefore, routine use of cardiac MRI in patients

with FD may assist in identifying patients who have subclinical myocardial involvement as this may have prognostic implications and may influence management of such patients – in one study, cardiac MRI was able to detect cardiac involvement in 21% of FD patients not on ERT (142).

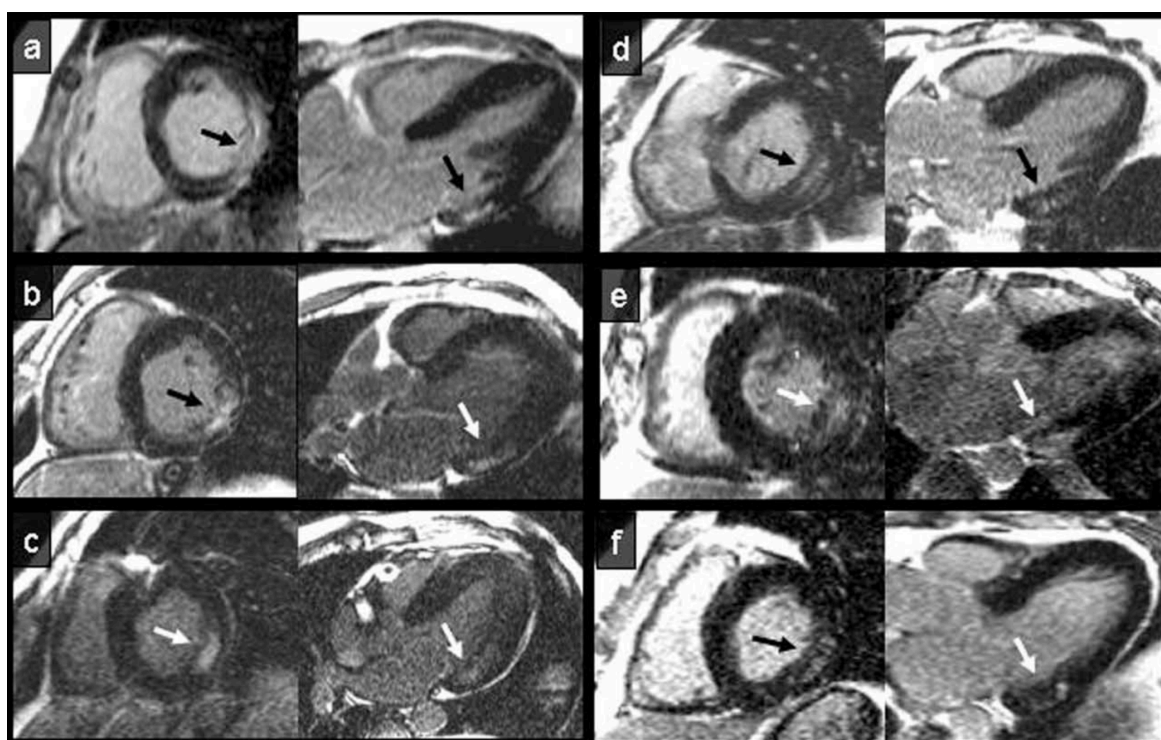


Figure 15. Cardiac MRI of 6 patients with FD showing consistent infero-lateral LGE without wall motion abnormalities in short axis (left column) and long axis (right column) views. From: "*Gadolinium enhanced cardiovascular magnetic resonance in Anderson-Fabry disease. Evidence for a disease specific abnormality of the myocardial interstitium.*" (Moon et al. 2003; Eur Heart J 24(23): 2151-2155).

Through native non-contrast T1 mapping, cardiac MRI can detect myocardial disease involving the myocyte and interstitial space – in patients with FD, quantitative measurements of myocardial T1 signal show that it is reduced (143, 144). The reduction in myocardial T1 signal may be a result of increased myocardial lipid content (145). T1 signal reduction is seen in over 90% of patients with FD who have LVH, but more

importantly, it is also seen in 40% of patients prior to the development of LVH (146). The reduction in T1 signal is seen in the interventricular septum as well as the right ventricular free wall (145, 147). However, in the inferolateral wall (the location of myocardial fibrosis), T1 signal can become 'pseudo-normal' or even increased as the low native T1 signals due to lipid overload are counteracted by the inferolateral replacement fibrosis (148).

Finally, cardiac MRI can be utilised to identify other changes which may not be as readily detected on TTE. These include apical aneurysm (149) as well as papillary muscle hypertrophy (which can be a marker of early cardiac involvement in FD) (150).

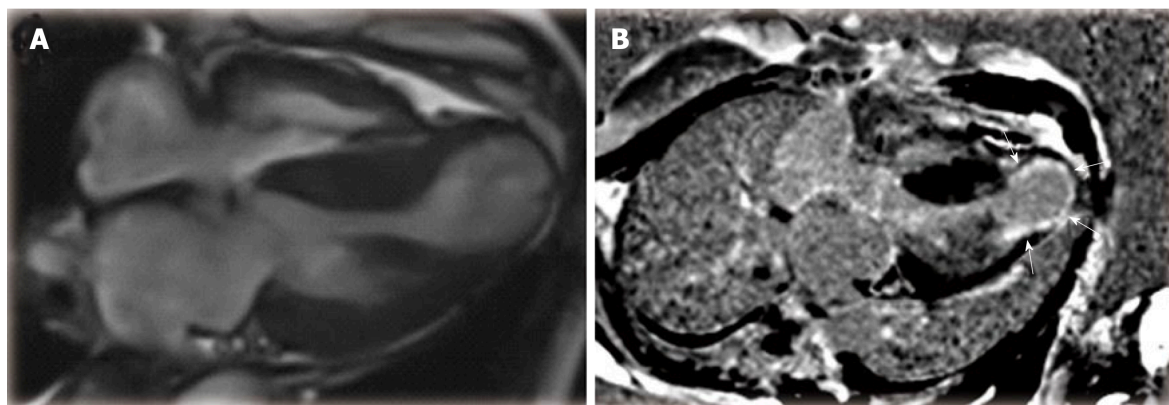


Figure 16. (A) Cardiac MRI with contrast showing concentric LVH with mid-cavity obstruction and large apical aneurysm in long-axis view. (B) LGE at site of LV apical aneurysm (arrows) with extension into the infero-lateral segment. From: "*Advanced Anderson-Fabry disease presenting with left ventricular apical aneurysm and ventricular tachycardia.*" (Poulin et al. 2015; World J Clin Cases 3(6): 519-524).

3.1.4 Laboratory studies

Several studies have tried to establish an association between laboratory tests and prognosis. Elevated levels of high sensitivity troponin correlate with the degree of myocardial fibrosis seen on cardiac MRI (by way of LGE), and thus represents advanced disease with significant cardiac involvement and as such is associated with a worse prognosis (100, 151, 152). Other cardiac biomarkers such as plasma NT-proBNP are also raised in patients with FD who have cardiac involvement. Although NT-proBNP levels can be elevated in patients without LVH on echocardiography, highest concentrations of NT-proBNP are usually seen in patients with LVH, diastolic dysfunction (153) and with evidence of myocardial fibrosis or scar on cardiac MRI (154).

Plasma levels of the accumulated glycosphingolipid lyso-Gb3 are raised in patients with FD. With initiation of ERT or chaperone therapy, a decrease in lyso-Gb3 levels can be seen (155). Therefore, plasma levels of lyso-Gb3 can be monitored to gauge the effectiveness of treatment on reducing levels of glycosphingolipids. Patients on ERT can also develop neutralizing anti-drug antibodies. Presence of these antibodies has been associated with worsening cardiac disease (increased septal wall thickness) and kidney disease (deterioration in kidney function) (156). An increase in lyso-GB3 levels for patients on ERT may indicate the presence of anti-drug antibodies.

Routine evaluation of renal function as well as an assessment of the presence of microalbuminuria or proteinuria is also imperative as renal dysfunction can occur secondary to renal involvement in FD (157). Importantly, there is an association with

severe renal dysfunction and an increased risk of developing cardiac complications in FD (102).

3.1.5 Endomyocardial biopsy

Endomyocardial biopsy should not be routinely performed and instead should be used to confirm or exclude FD as the cause of LVH when the diagnosis is uncertain. For example, endomyocardial biopsy can be considered in patients with LVH who have a genetic mutation in the GLA enzyme with a variance of unknown significance, low lyso-Gb3 levels (as they should be elevated) or high residual enzyme activity (i.e. greater than 10%, as enzyme activity should be depressed) (158, 159).

4.1 Management of Fabry Disease

Treatment in FD comprises of directed therapy for the underlying pathological disease process as well as medical therapy for the sequelae of organ involvement. The directed therapy, which includes ERT and chaperone therapy, aim to increase levels of enzyme activity and thus reduce levels of accumulated substrate.

4.1.1 Enzyme Replacement Therapy and Chaperone Therapy

4.1.1.1 *Enzyme Replacement Therapy*

ERT is indicated in all symptomatic patients at the earliest sign of organ involvement, as ERT can slow Gb3 deposition in organs including the heart. Studies have shown that patients with FD who initiated treatment at a younger age, tended to have better outcomes than those who began treatment at an older age with more advanced disease (160). In early trials of ERT, patients with FD on ERT for 6 months, had significantly reduced LV mass on cardiac MRI and had a mean reduction of 20% in myocardial Gb3 content on endomyocardial biopsy (161). Whereas, patients who had advanced heart disease (e.g. patients with increased LV mass and with fibrosis on cardiac MRI depicted by LGE), did not show regression of LVH during ERT (162). In contrast, mild LVH may partially regress in patients with FD if ERT is commenced early (163). Therefore, in male patients, treatment guidelines generally recommend commencing ERT at the time of diagnosis (as early treatment improves outcomes, particularly prior to the development of myocardial fibrosis and LVH). In female patients however, ERT should be considered for symptomatic patients (neuropathic pain, severe gastrointestinal symptoms, pathological proteinuria, or cardiac involvement which has traditionally focused on

assessing for evidence of LVH) (164). Interestingly, as described previously, female patients can develop myocardial fibrosis (detected on cardiac MRI), prior to the development of LVH. Thus, there has been interest in considering whether female patients will benefit from ERT prior to the development of LVH (141). However, it is important to note that cardiac MRI is an expensive imaging modality and is not as readily available as other imaging modalities such as echocardiography.

Studies on longer term outcomes of ERT have shown mixed results. For instance, a Cochrane meta-analysis reviewing randomised controlled trials highlighted that due to the rarity of the condition and limited population available to study, the long term influence of ERT on morbidity and mortality was not clear (although only 9 trials were analysed with limited long term follow-up data) (165). In contrast, pooled analysis of 77 cohort studies showed that ERT is associated with a lower incidence of renal, cardiovascular and cerebrovascular events and thus this study recommended the use of ERT for preventing major organ complications (166).

ERT is administered as a fortnightly intravenous infusion and there are two types available – agalsidase alfa (Replagal) or agalsidase beta (Fabrazyme or Fabagal).

4.1.1.2 Chaperone Therapy

Chaperone therapy is an alternative to ERT, where an orally administered medication (migalastat) binds to the active site of the GLA enzyme and stabilises the enzyme which

results in allowing appropriate trafficking of the enzyme to lysosomes. Once within the lysosome, the chaperone molecule dissociates from the enzyme allowing the enzyme to perform its function and catabolise substrates (167). Data from several trials has shown that migalastat therapy results in a reduction in LV mass (168, 169). The advantage of migalastat is that it is an oral therapy compared to fortnightly intravenous infusions; however, the disadvantage is that migalastat therapy is reserved for patients who have an amenable specific gene mutation (most commonly a single missense mutation), which is the case for only 30% of patients with FD (79).

4.1.1.3 Novel therapies in development for Fabry Disease

Substrate reduction therapy (SRT) works by inhibiting the synthesis of glycosphingolipids such as Gb3 and thus lower Gb3 levels systemically (170). One such agent, Lucerastat is currently being investigated as a potential therapeutic option (171).

Gene therapy is also a potential future therapeutic option. Studies in FD knockout mice have shown success in introducing a functional GLA gene via an adenovirus or retrovirus vector, with subsequent increase in enzyme activity levels at 6 months (172).

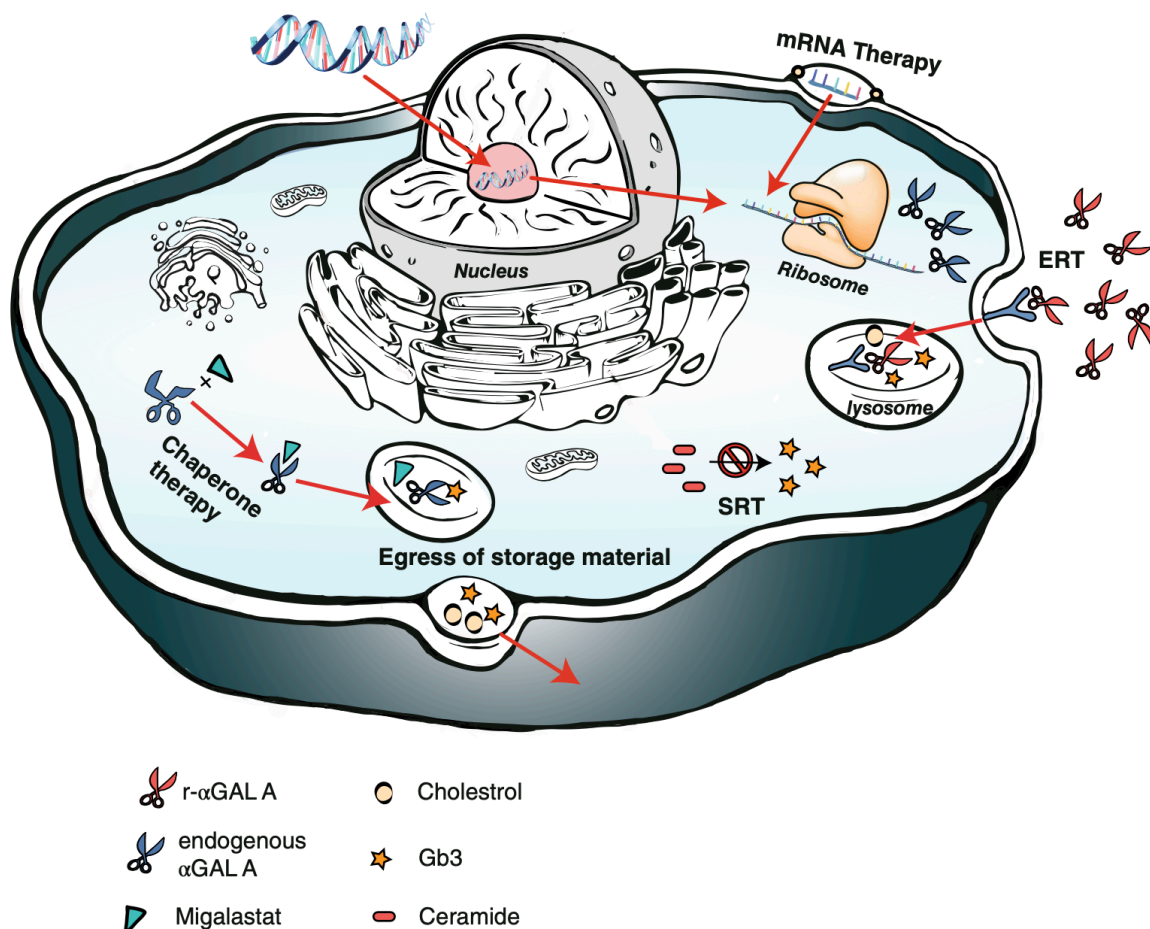


Figure 17. Overview of different approaches in treating FD – ERT aims to replace defective GLA. Chaperone therapy binds to the active site of the unstable GLA enzyme to aid proper folding. Substrate reduction therapy targets glycosphingolipid synthesis with subsequent reduction in formation of Gb3 and its derivatives. Gene therapy aims to correct the underlying genetic defect of FD using a viral vector. MRNA therapy induces transient endogenous production of the GLA enzyme. From: " *Developments in the treatment of Fabry disease*" (Van der Veen et al. 2019; J Inherit Metab Dis 43(5): 908-921).

4.2.1. Assessment, investigation and management of cardiac issues

4.2.1.1 Palpitations and Syncope

Palpitations frequently occur and have been reported by 15% to 38% of patients with FD at some stage of their disease (98). AF is common in patients with FD, therefore,

episodes of palpitations which occur often, or which are prolonged in duration should be investigated with ambulatory ECG monitoring.

Syncope is reported by 2-6% of adult patients with FD, being more common in men (98). Syncope in patients with FD can be due to bradyarrhythmia (due to sinus node dysfunction, atrioventricular block), tachyarrhythmia (due to VT), autonomic dysfunction or structural causes (due to LV outflow tract obstruction). Therefore, it is imperative that patients with syncope are investigated with an ECG, prolonged ambulatory monitoring (e.g. 48 hours), exercise stress test (to assess for chronotropic competence) as well as considering exercise stress echocardiography in patients who have exertional syncope (to look for LV outflow tract obstruction) (173). In patients who a cause for syncope cannot be identified, insertion of an implantable loop recorder should be considered (122, 174).

Symptomatic bradycardia is common in FD – in one study 8% of patients followed over a 5-year period required anti-bradycardia pacing with approximately half of the patient's indication being sinus node dysfunction and the other half being atrioventricular block (118). Similarly, in FD, non-sustained ventricular tachycardia (NSVT) is commonly seen – in one study, up to 8% of patients had NSVT detected on ambulatory ECG monitoring (175). Data on the relationship between NSVT and the risk of sudden cardiac death is limited in FD, however, in myocardial disease processes that cause fibrosis, the extent of fibrosis as well as documented NSVT were associated with sudden cardiac death – this was also seen in one study in patients with FD where sudden cardiac death only occurred in patients with documented VT and with evidence of myocardial fibrosis on cardiac MRI

(176). Therefore, patients with FD who have survived a cardiac arrest (due to VT or ventricular fibrillation) or who have syncope or haemodynamic collapse as a result of VT, should receive AICD therapy as per current guidelines (123). In relation to AICD therapy for primary prevention, the decision should be individualised for each patient taking into account the known risk factors associated with sudden cardiac death (including age, male gender, LVH, LGE on cardiac MRI and the presence of NSVT) (100).

4.2.1.2 Atrial Fibrillation

AF occurs commonly in patients with FD, with approximately 5% of male and 3% of female patients developing AF (108). Similarly, patients with FD have a higher incidence of stroke than the general population (95).

Anticoagulation

Scoring systems for estimating risk of stroke in patients with FD who develop AF are not validated and should not be used (e.g. CHADS₂, CHADS₂VASc). However, anticoagulation should be considered in all patients with AF or atrial flutter (108). Non-vitamin K Oral Anticoagulants (NOACs) may be preferred over warfarin as they have a lower risk of intracranial bleeding (and patients with FD are at increased risk of cerebral microbleeds (177)) – although data on NOAC use is lacking. NOAC use may also slow the progression of renal function deterioration as it reduces the risk of warfarin-induced nephropathy (178). However, as renal function declines in FD, NOACs may be contraindicated, and

other anticoagulation should be considered. In patients where anticoagulation is contraindicated, left atrial appendage closure could be considered (108, 179).

Rhythm control

A rhythm control approach to the management of AF is often difficult in patients with FD due to progressively worsening atrial substrate and as rhythm control pharmacotherapy is often contraindicated (108). Sotalol use can be considered, however, it is contraindicated in decompensated congestive heart failure or with significant renal dysfunction ($\text{CrCl} < 10\text{ml/min}$) (180). Likewise, flecainide is contraindicated with severe renal impairment and should be avoided in patients with structural heart disease as it may be pro-arrhythmogenic and increases cardiac adverse events (181). Long term amiodarone use may induce phospholipidosis, resulting in a decrease in intracellular lysosomal enzyme levels (182), and thus could potentially reduce the effects of ERT (108) – thus, amiodarone use should be limited to acute symptomatic episodes where other medical therapy has been exhausted or is contraindicated. In patients with FD, data on catheter ablation is scarce, but there have been reports of successful restoration of sinus rhythm post pulmonary vein isolation (183) and so an interventional approach to rhythm control should be considered. Alternatively, if control of AF is unable to be achieved, his bundle ablation combined with pacemaker implantation could be considered (77).

Rate control

Patients with FD are prone to developing significant bradycardia and atrioventricular conduction abnormalities. Therefore, if a rate control strategy is being considered, it is essential to perform ambulatory ECG monitoring prior to and during therapy with such agents to prevent the development of significant symptomatic bradycardia (108).

4.2.1.3 Chest pain and coronary artery disease

Patients with FD have evidence of endothelial involvement of coronary arteries. In one study, intravascular ultrasound demonstrated more diffuse and hypoechogenic plaque in FD compared to controls (who had more focal and echogenic plaque), which is most likely a result of higher lipid content in plaque and accumulation of glycosphingolipid in endothelial and smooth muscle cells (184). Despite this, patients with FD do not have an increased incidence of coronary stenosis and acute coronary syndrome compared to the general population. However, patients with FD who have chest pain should still have an assessment for coronary artery disease as a cause of angina. It is important to note that myocardial perfusion imaging via positron emission tomography shows a decrease in coronary flow reserve in patients with FD who have no large epicardial coronary artery stenosis and so this investigation may not be as relevant for clinical evaluation and management (185, 186). It is also important to note that patients with FD often have abnormal baseline ECG changes (i.e. LVH criteria with repolarisation abnormalities) and so exercise stress testing may be of limited value.

In many patients with FD, chest pain is a result of microvascular dysfunction and small vessel disease (186, 187). There is evidence on myocardial perfusion imaging via positron emission tomography, that myocardial blood flow is reduced particularly to the apical region, even in patients who are female and who do not have LVH (186).

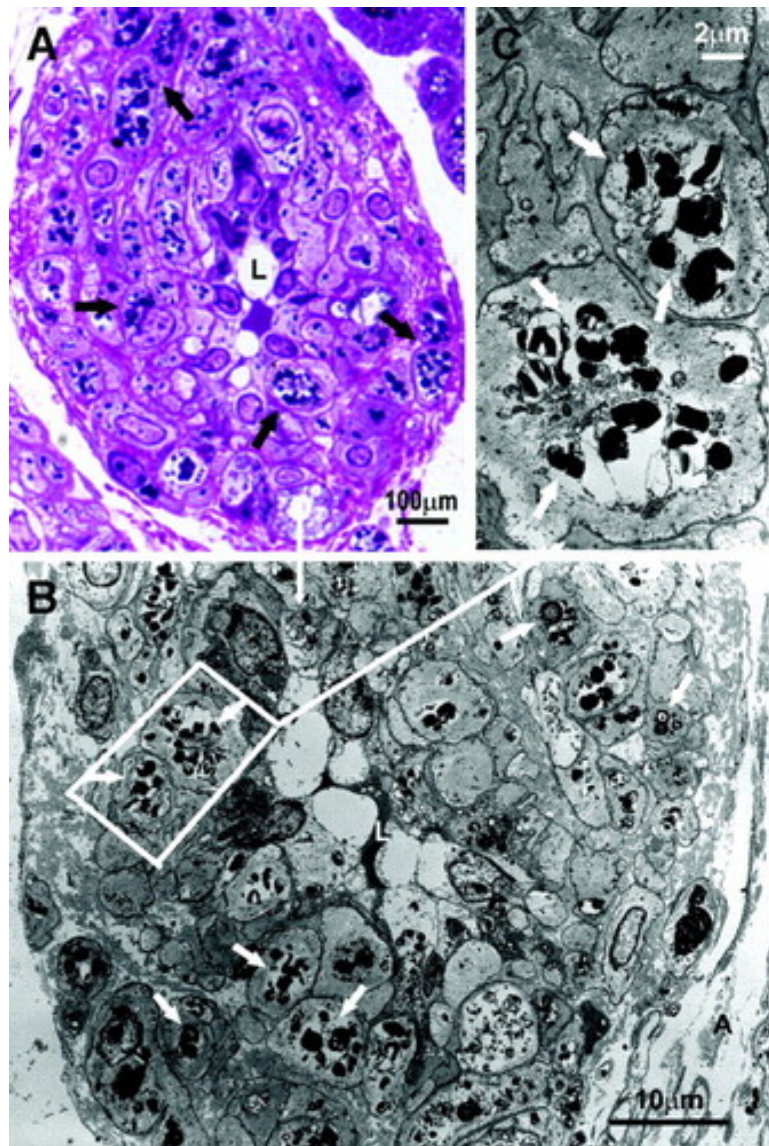


Figure 18. (A) Sections from LV endomyocardial biopsy from a patient with FD showing an intramural coronary artery exhibiting thickened wall and narrowed lumen (L) due to hypertrophy and hyperplasia of smooth muscle cells (A, black arrows); electron microscopy (B and C) shows these smooth muscle cells consist of glycosphingolipids organized in membrane-bound bodies (white arrows). From: "*Angina in fabry disease reflects coronary small vessel disease.*" (Chimenti et al. 2008; Circ Heart Fail 1(3): 161-169).

Once the diagnosis of coronary artery disease is made, patients with FD should be managed as per current guidelines. However, it is important to note that patients who have significant LVH carry a higher peri-operative risk for coronary artery bypass grafting surgery as well as for coronary angiography with percutaneous coronary intervention (108). Additionally, care should be exercised if utilising beta blockers or calcium channel blockers for their anti-anginal properties as their negative chronotropic effects carry increased risk of bradycardia in patients with FD (108).

4.2.1.4 Heart failure

Heart failure symptoms and exertional dyspnoea are common in patients with FD. Peripheral oedema is also common in patients with FD, and although heart failure can contribute, patients with FD also have renal involvement with subsequent nephropathy as well as lymphoedema (188). Heart failure symptoms are likely secondary to diastolic dysfunction and increased LV diastolic pressure given the LV ejection fraction is normal in the majority patients. However, other conditions need to be considered.

As per the ESC guidelines for the management of patients with HCM, It is recommended that all patients with FD with exertional dyspnoea undergo stress echocardiography to assess for exercise-induced LV outflow tract obstruction as well as exercise-induced mitral regurgitation (189). Additionally, patients with FD may also experience exertional dyspnoea secondary to chronotropic incompetence and so there is benefit in performing

exercise stress testing when considering this possibility (190). Finally, patients with FD can also have pulmonary involvement and so spirometry should be performed in all breathless patients to assess for obstructive or restrictive lung disease (191).

The management of heart failure with reduced ejection fraction is as per current guidelines as well. However, caution should be exercised with therapy with beta blockers and ivabradine as patients with FD are at increased risk of symptomatic bradycardia and atrioventricular nodal dysfunction (108). Similarly, caution should be exercised with the use of ACEi, ARBs and spironolactone as patients with FD often have nephropathy with renal involvement and are prone to the development of hyperkalaemia with such therapy (108).

Chapter 2 – Basal segmental longitudinal strain: a marker of subclinical myocardial involvement in Fabry Disease

As discussed above, cardiac involvement in FD is characterised by progressive increase in LV wall thickness. It more commonly affects males due to the X-linked inheritance pattern. Echocardiographic two-dimensional LS analysis has increased sensitivity for the detection of subclinical myocardial involvement in various diseases (14) and is more readily available than cardiac MRI. This chapter reports results of our first study. The aim of this study was to assess global and regional LS in patients with FD. Additionally, this chapter sought to examine the association between echocardiographic parameters (including LS) and MACE.

The findings highlighted that in patients with FD, basal LS was differentially impaired (more so than mid wall and apical LS). Impairment in basal LS was present even in female patients with FD, despite normal LV wall thickness. Finally, impaired basal LS was associated with MACE.

These findings have important clinical implications. Firstly, basal LS analysis should be considered when screening patients with FD for cardiac involvement, as up to 60% of patients with FD, and particularly females, may not have increased LV wall thickness at the time of diagnosis (135). TTE and LS analysis is more readily available than cardiac MRI and as such could be more widely utilised in screening for cardiac involvement. Secondly,

current Australian guidelines recommend instituting ERT for men with FD at diagnosis, whilst for female heterozygotes, the recommendation to start ERT is based on the presence of symptoms or evidence of organ involvement (i.e. increased LV wall thickness). Therefore, by screening female patients with FD for impaired basal LS, this could identify subclinical cardiac involvement, thereby prompting earlier treatment with ERT.

Basal Segmental Longitudinal Strain: A Marker of Subclinical Myocardial Involvement in Anderson-Fabry Disease



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Background: Cardiac involvement in Anderson-Fabry disease (AFD) is associated with increased left ventricular (LV) wall thickness. The aim of this study was to evaluate if two-dimensional global and regional strain in patients with AFD can identify early myocardial involvement (when LV wall thickness and function are normal). Additionally, the association of altered strain with adverse cardiovascular events was evaluated.

Methods: In a retrospective cross-sectional study, 43 patients with AFD, before enzyme replacement therapy (mean age, 44 ± 12 years; 58.1% men), were compared with age- and gender-matched healthy control subjects. The mean follow-up duration among patients with AFD for major adverse cardiovascular events (MACE) was 82 months.

Results: LV ejection fraction was similar between groups (patients with AFD vs control subjects, $61 \pm 8\%$ vs $61 \pm 6\%$; $P = .89$). However, global longitudinal strain (LS) was impaired in patients with AFD compared with control subjects ($-16.5 \pm 3.8\%$ vs $-20.2 \pm 1.7\%$, $P < .001$), with greater impairment in patients with AFD with increased LV wall thickness ($-15.4 \pm 3.9\%$ vs $-18.7 \pm 2.3\%$, $P < .006$). Additionally, LS was most impaired in the basal segments in patients with AFD ($-14.8 \pm 3.7\%$ vs $-20.3 \pm 1.1\%$, $P < .001$). MACE occurred in 19 of 43 patients (four women, 15 men), and Kaplan-Meier analysis demonstrated that MACE were associated with impaired basal LS.

Conclusions: In patients with AFD, altered basal LS is present even in those with normal LV wall thickness and is associated with MACE. Therefore, basal LS should be considered when screening for cardiac involvement in AFD, particularly in female patients with AFD with normal LV wall thickness. (J Am Soc Echocardiogr 2021;34:405-13.)

Keywords: Anderson-Fabry disease, Strain, Basal longitudinal strain, Two-dimensional speckle-tracking, Left ventricular wall thickness, Adverse outcomes

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Anderson-Fabry disease (AFD) is an X-linked lysosomal storage disease, caused by α -galactosidase A enzyme deficiency, with consequent progressive accumulation of globotriaosylceramide in various organ tissues.¹ The clinical manifestations of AFD include dermatologic, ophthalmologic, neurologic, gastrointestinal, renal, and cardiac involvement, with the heart being the organ most frequently involved.² Although many symptoms occur in childhood, cardiac involvement is often not detected until the third or fourth decade of life.³

Cardiac involvement in AFD is characterized by a progressive increase in left ventricular (LV) wall thickness, interstitial remodeling, and eventual replacement myocardial fibrosis.⁴ The heart can be the only organ involved in patients with specific mutations⁵ and in female heterozygous carriers with near normal enzymatic activity.⁶ Traditionally, female heterozygotes were considered disease "carriers"⁷; however, it is now widely accepted that affected women can develop cardiomyopathy and other organ involvement, similar to hemizygous men, albeit with delayed onset and slower progression.⁸ Cardiac

Abbreviations**2D** = Two-dimensional**AFD** = Anderson-Fabry disease**AUC** = Area under the curve**COV** = Coefficient of variation**ERT** = Enzyme replacement therapy**GCS** = Global circumferential strain**GLS** = Global longitudinal strain**LS** = Longitudinal strain**LV** = Left ventricular**LVEF** = Left ventricular ejection fraction**LVMI** = Left ventricular mass index**MACE** = Major adverse cardiovascular event(s)**ROC** = Receiver operating characteristic**SR** = Strain rate**TTE** = Transthoracic echocardiography

morbidity and mortality have been associated with increased wall thickness.⁹

In this study, we evaluated myocardial mechanics in patients with AFD by echocardiographic two-dimensional (2D) strain analysis that has increased sensitivity for the detection of subclinical myocardial involvement in a variety of conditions.¹⁰

Our aim was to assess alterations in global and regional LV longitudinal strain (LS) in an enzyme replacement therapy (ERT)-naïve AFD cohort and to examine the effects of gender and LV wall thickness on basal LS. In addition, we examined for echocardiographic parameters that were associated with major adverse cardiovascular events (MACE).

METHODS

This was a retrospective cross-sectional study of 43 consecutive genetically confirmed patients with AFD (mean age, 44 ± 12 years; 25 men; refer to [Supplemental Table 4](#) for baseline

clinical features) from the Department of Genetic Medicine Clinic at Westmead Hospital (2009-2014). All patients underwent transthoracic echocardiography (TTE) after initial review in the clinic, and no patient had commenced ERT. Patients with AFD were compared with age- and gender-matched healthy control subjects, with no documented cardiovascular disease or risk factors, who were not on any medications, from a departmental database. Ethical approval was obtained from the South West Sydney Health District Human Research Ethics Committee.

Echocardiography

Comprehensive TTE was performed using a Vivid 7 or E9 ultrasound scanner (GE Vingmed Ultrasound, Horten, Norway) with a 2.5-MHz broadband transducer; M-mode, 2D, color, Doppler, and Doppler tissue imaging were obtained according to American Society of Echocardiography guidelines.¹¹ An average of three measurements were used for all parameters. Raw digital images were stored for offline analysis.

Traditional Measurements

LV wall thickness was measured from 2D images from the parasternal long-axis view in end-diastole; patients with AFD were stratified on the basis of American Society of Echocardiography guidelines to have increased wall thickness if ≥ 11 mm for men and ≥ 10 mm for women.¹² Relative wall thickness was also determined.¹² LV end-diastolic and end-systolic volumes were measured using the Simpson biplane method from apical four- and two-chamber views,

and LV ejection fraction (LVEF) was calculated. LV mass was calculated using the Devereux method per American Society of Echocardiography criteria and indexed (LV mass index [LVMI]) to body surface area¹³; gender-based cutoffs for increased LV mass (>115 g/m² for men and >95 g/m² for women) were used. Peak E and peak A velocities were obtained from pulsed Doppler mitral inflow.¹⁴ Tissue Doppler velocities (s' , e' , and a' velocities) were measured from the septal and lateral mitral annulus.¹⁵ The ratio of E velocity to the average of septal and lateral e' velocity was determined (E/e').

Two-Dimensional LV Strain Imaging

Two-dimensional strain analysis was performed retrospectively using the same customized offline computer software for all patients (EchoPAC version 201; GE Vingmed Ultrasound), from zoomed apical LV four-, two-, and three-chamber views, obtained at high frame rates (>55 frames/sec). The endocardial border was manually traced in end-systole, and the width of the region of interest was manually adjusted to track the myocardium. Peak systolic strain (S) in each apical view was obtained from six segments ([Figure 1](#)) and global LS (GLS) as an average from three apical views. Additionally, systolic (S), early diastolic (E), and late diastolic (A) strain rates (SRs) were obtained. Circumferential and radial strain were obtained as averages of six segments from the parasternal short-axis views at the papillary muscle level, per guidelines.^{16,17} Strain from up to two segments could be excluded in one imaging plane; if more than two segments could not be adequately tracked, the strain measurement from this plane was omitted. Additionally, LS from basal, mid, and apical segmental levels was calculated as an average of six segments at each level.

Clinical Outcomes

MACE were assessed after initial TTE, including death, myocardial infarction, cerebrovascular accident, exacerbation of heart failure, arrhythmia, or implantation of a permanent pacemaker or an automatic implantable cardioverter-defibrillator. The mean follow-up duration of patients with AFD for MACE was 82 months.

Statistical Analysis

Continuous variables are expressed as mean $\pm$ SD and categorical variables as percentages. Nonparametric tests (Mann-Whitney and Kruskal-Wallis) were used to evaluate differences between patients with AFD and control subjects. Conditional logistic regression analysis was used in matched case-control sets to evaluate segmental strain in patients with AFD versus matched control subjects. Nonparametric tests were also used to examine differences between male and female patients with AFD and in patients with AFD with and without increased LV wall thickness and LVMI ([Supplemental Tables 1 and 2](#)). Pearson correlation examined the association between basal LS and LV wall thickness and LVMI. Receiver operating characteristic (ROC) curve analysis was performed to evaluate the areas under the curve (AUCs) for GLS, basal LS, and LVEF in identifying cardiac involvement in AFD; the DeLong test was used to determine if they were significantly different. Kaplan-Meier analysis using the log-rank test was performed for GLS, basal LS, and LV wall thickness as predictors of MACE. ROC curve analysis was performed to evaluate the AUCs for basal LS and LV wall thickness in predicting MACE. Statistical analyses were performed using SPSS version 22

HIGHLIGHTS

- In Fabry disease, impaired basal strain demonstrates early myocardial involvement.
- Basal strain is impaired even when LV wall thickness is normal.
- Impaired basal strain is associated with worse clinical outcomes.
- Consider basal strain analysis when screening patients with Fabry disease.

(SPSS, Chicago, Illinois) and MedCalc version 15.8 (MedCalc Software, Ostend, Belgium), with P values $< .05$ considered to indicate statistical significance.

To assess intraobserver variability, 10 randomly selected patients were reviewed by the same investigator 4 weeks apart. Interobserver variability was assessed by a second blinded investigator in the same 10 patients, and intraclass correlation coefficients and coefficients of variation (COVs) were calculated.

RESULTS

Clinical and traditional echocardiographic characteristics of patients with AFD and matched control subjects are presented in [Table 1](#). Despite differences between groups for blood pressure, values were within the normotensive range. Clinical and traditional echocardiographic characteristics of patients with AFD on the basis of gender are presented in [Table 2](#).

Traditional Echocardiographic Parameters

LV wall thickness and LVMI were increased in patients with AFD and were greater in male than female patients with AFD. LVEF and s' velocity were comparable between patients with AFD and control subjects and between male and female patients with AFD ([Tables 1](#) and [2](#), respectively). Diastolic function demonstrated an increase in E velocity, a decrease in e' velocity, and a consequent increase in E/e' ratio in patients with AFD compared with control subjects ([Tables 1](#) and [2](#)).

LV Global and Segmental Strain

Mean GLS in the control group ($-20.2 \pm 1.7\%$) was comparable with normal reported GLS values (-19.7% ; 95% CI, -20.4% to -18.9%).¹⁸ GLS was impaired in patients with AFD compared with control subjects ([Table 3](#)). There was no significant difference in GLS between male and female patients with AFD ([Table 4](#)).

S SR and circumferential and radial strain were also impaired in patients with AFD, with no difference on the basis of gender. Strain measurements from the six basal, mid, and apical segmental levels were averaged ([Table 3](#)). LS was significantly impaired in all three segmental levels in the AFD group ([Figure 2](#)), with the greatest impairment in basal segmental LS ([Figures 3A–C](#) and [Supplemental Table 5](#)).

We performed ROC analysis for identifying cardiac involvement in patients with AFD; an AUC of 0.85 was obtained for GLS and a significantly higher AUC of 0.93 for basal LS by DeLong analysis ($P = .011$; [Figure 4](#)). GLS more impaired than -18.1% had sensitivity of 67% and specificity of 98%, and basal LS more impaired than -18.1% had sensitivity of 81% and specificity of 100%.

Reproducibility of 2D Strain Analysis

The intraclass correlation coefficient for interobserver variability for GLS was 0.96 (95% CI, 0.82–0.99; COV = 18.1%) and for S SR was 0.85 (95% CI, 0.39–0.96; COV = 18.1%), while intraobserver variability was 0.99 (95% CI, 0.96–1.00; COV = 19.8%) and 0.99 (95% CI, 0.94–0.99; COV = 14.7%), respectively. Intraclass correlation coefficients for interobserver variability for apical, mid, and basal segmental LS were 0.89 (95% CI, 0.54–0.97; COV = 12.8%), 0.99 (95% CI, 0.96–0.99; COV = 21.0%), and 0.98 (95% CI, 0.94–0.99; COV = 23.2%), respectively, while intraobserver variability was 0.96 (95% CI, 0.82–0.98; COV = 12.8%), 0.99 (95% CI, 0.98–1.00; COV = 21.0%), and 0.99 (95% CI, 0.98–1.00; COV = 23.3%), respectively.

MACE and Event-Free Survival

The mean follow-up duration for MACE was 82 months. MACE occurred in 19 of 43 patients with AFD (four women, 15 men); 10 had arrhythmias (six atrial fibrillation, four nonsustained ventricular tachycardia requiring implantation of defibrillators), four had cerebrovascular accidents, four had acute myocardial infarctions, and one had admission with heart failure ([Supplemental Table 3](#)). ERT was commenced in 26 of 43 patients after their initial TTE (with 17 of 19 in the subgroup that sustained MACE and nine of 24 in the subgroup that did not sustain MACE).

The prognostic value of LV wall thickness, GLS, and basal LS was evaluated using log-rank analysis, examining variables as tertiles. Log-rank analysis showed that increased LV wall thickness (tertiles: <10.1 , 10.2 to 12.8, and >12.8) and impaired basal LS (basal LS tertiles: $<-13.5\%$, -13.5% to -16.5% , and $>-16.5\%$) were associated with reduced event-free survival ($P = .017$ and $P = .009$, respectively). However, GLS did not demonstrate a significant association ($P = .11$). Additionally, clinical outcomes were worse for patients with AFD who had greater basal segmental LS impairment (two of 14 events for basal LS better than -16.5% , five of 14 events for basal LS -13.5% to -16.5% , and 12 of 15 events for basal LS worse than -13.5% ; $P = .009$). Kaplan-Meier analysis was performed for LV wall thickness, GLS, and basal LS ([Figure 5](#)). ROC curve analysis for predicting MACE demonstrated AUCs of 0.82 for basal LS and 0.79 for LV wall thickness, which were not significantly different by DeLong analysis ($P = .74$; [Figure 6](#)).

Increased LV Wall Thickness and Mass and Strain Analysis

Data for normal values of basal LS are not well established. Therefore, we defined the cutoff for normal basal LS on the basis of our control group (i.e., mean -2 SDs; $-20.3\% + 2.2\% = -18.1\%$). Of the 11 of 18 (61%) female patients with AFD who had normal LV wall thickness, 6 of 11 (55%) had impaired basal LS ([Figure 7](#)). Among the male patients with AFD, only 3 of 25 (12%) had normal LV wall thickness, and 2 of 3 (67%) had impaired basal LS ([Figure 7](#)). Pearson correlation analysis between basal LS and LV wall thickness and between basal LS and LVMI showed inverse correlations ($r = -0.626$ and $r = -0.622$, respectively, $P < .001$ for both; [Supplemental Figures 1A and B](#)).

DISCUSSION

Our results highlight that patients with AFD have altered myocardial mechanics with reduced GLS and global circumferential strain (GCS).

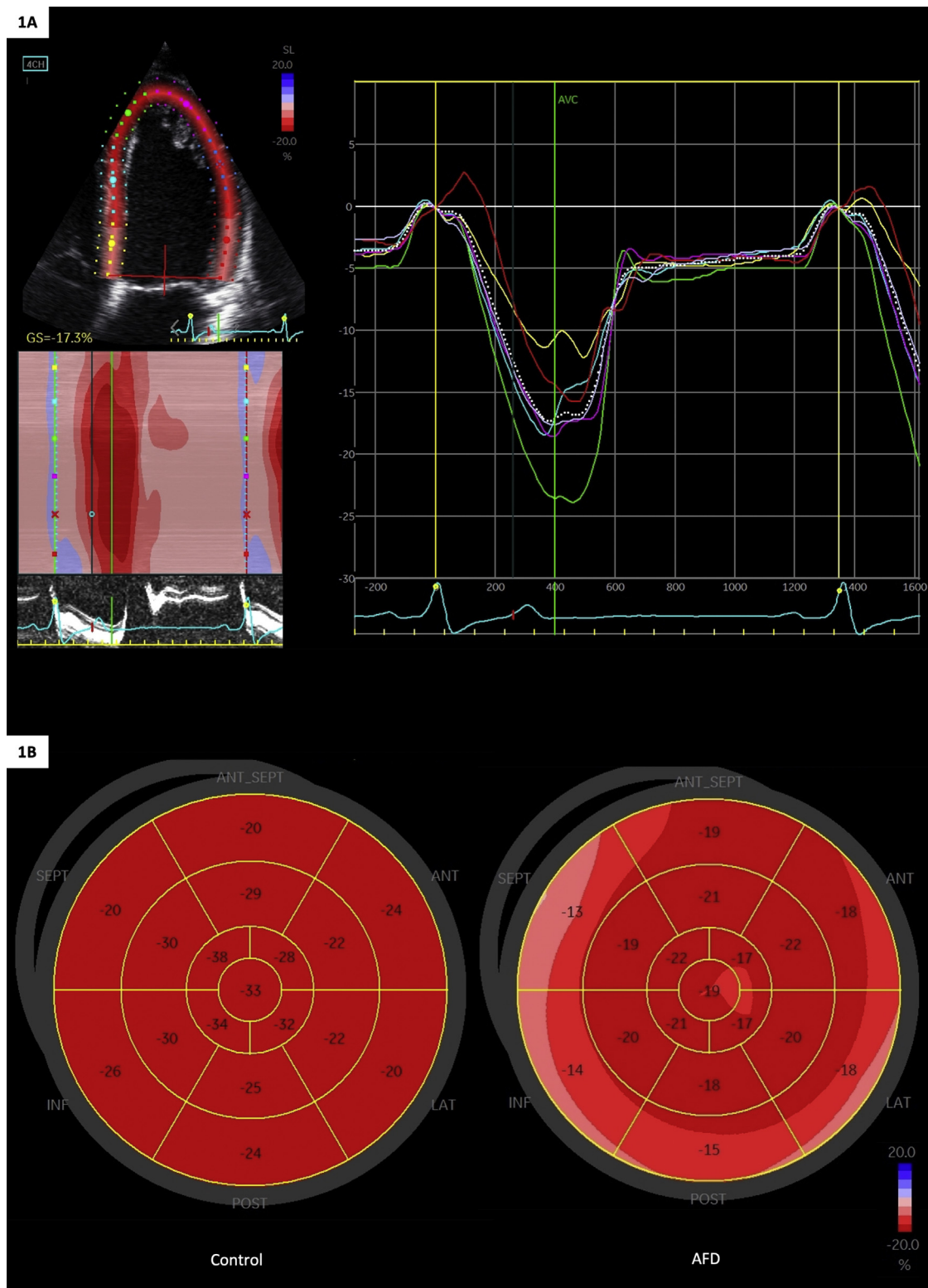


Figure 1 (A) Strain analysis from apical four-chamber view in a patient with AFD with impaired GLS. Strain of basal segments (red and yellow lines) was markedly lower than that of mid and apical segments. (B) Bull's-eye plot of peak systolic strain showing impaired basal segments in a patient with AFD compared with a control subject.

Table 1 Clinical and traditional echocardiographic characteristics of patients with AFD compared with age- and gender-matched normal control subjects

Parameter	Patients with AFD	Control subjects	P
	(n = 43)	(n = 43)	
Age, y	44 ± 12	45 ± 13	.704
Body surface area, m ²	1.85 ± 0.21	1.82 ± 0.19	.583
Systolic BP, mm Hg	122 ± 14	114 ± 9	.004*
Diastolic BP, mm Hg	77 ± 9	71 ± 8	.003*
Heart rate, beats/min	68 ± 13	67 ± 11	.914
IVS, mm	12.5 ± 4.0	8.5 ± 1.9	<.001*
PW, mm	11.6 ± 3.5	8.4 ± 1.8	<.001*
RWT	0.49 ± 0.17	0.36 ± 0.09	<.001*
LVEDD, mm	48.9 ± 5.9	47.1 ± 6.0	.302
LVESD, mm	28.6 ± 6.1	29.2 ± 5.8	.442
LVEF, %	61 ± 8	61 ± 6	.853
Indexed LVM, g/m ²	117 ± 43	67 ± 23	<.001*
Peak E velocity, cm/sec	85 ± 20	76 ± 17	.018
Peak A velocity, cm/sec	67 ± 17	53 ± 13	<.001*
Average E/e' ratio	10.3 ± 3.3	7.3 ± 2.7	<.001*
s' velocity, cm/sec	7.4 ± 1.5	7.7 ± 1.6	.551
e' velocity, cm/sec	8.1 ± 2.4	9.5 ± 2.7	.026*
a' velocity, cm/sec	8.7 ± 2.1	9.2 ± 2.2	.399

BP, Blood pressure; IVS, interventricular septum; LVEDD, LV end-diastolic diameter; LVESD, LV end-systolic diameter; LVM, LV mass; PW, posterior wall; RWT, relative wall thickness.

Data are expressed as mean ± SD.

*P < .05.

The greatest reduction in LS was in the basal segments, and this was evident even when LV wall thickness was normal. Basal LS was a better indicator of subclinical cardiac involvement than GLS and was also associated with MACE. Therefore, using basal LS to identify early myocardial involvement in patients with AFD may prompt early initiation of ERT and could thereby improve clinical outcomes.

Changes in Echocardiographic Parameters in Fabry Disease

AFD is an infiltrative disorder resulting in intracellular accumulation of glycosphingolipids, with men manifesting cardiac involvement earlier.¹⁹ Therefore, it is expected that structural LV parameters such as wall thickness and LVMI are increased, as observed in our AFD cohort, and more so in hemizygous men compared with female patients with AFD, despite being similar in age.

Similar to previous studies,¹⁹⁻²² even though LV systolic function parameters such as LVEF and S' velocity were similar between patients with AFD and control subjects, LV GLS and GCS were impaired in patients with AFD. Gruner *et al.*¹⁹ reported similar subclinical myocardial dysfunction, with reductions in both GLS and GCS, and like our study, theirs demonstrated worsening GLS with increased LV wall thickness. The myocardial architecture of the left ventricle is complex, with subendocardial longitudinal fibers, circumferentially arranged midmyocardial fibers, and subepicardial oblique fibers.²³ GLS is largely attributed to subendocardial fiber function, while GCS is attributed to subepicardial fiber function.²⁴

Table 2 Clinical and traditional echocardiographic characteristics of patients with AFD by gender

Parameter	Men	Women	P
	(n = 25)	(n = 18)	
Age, y	44 ± 11.9	44 ± 11.9	.922
Body surface area, m ²	1.91 ± 0.18	1.77 ± 0.22	.033*
Systolic BP, mm Hg	125 ± 13	118 ± 14	.054
Diastolic BP, mm Hg	79 ± 9	74 ± 8	.170
Heart rate, beats/min	66 ± 13	71 ± 14	.237
IVS, mm	14.5 ± 3.6	9.8 ± 2.9	<.001*
PW, mm	13.1 ± 3.3	9.6 ± 2.6	<.001*
RWT	0.54 ± 0.19	0.40 ± 0.11	.009*
LVEF, %	60.2 ± 9	63 ± 7	.491
Indexed LVM, g/m ²	133 ± 38	95 ± 39	.002*
LVEDD, mm	49.9 ± 6.4	47.5 ± 5.0	.331
LVESD, mm	29.3 ± 6.8	27.7 ± 4.8	.605
Peak E velocity, cm/sec	78 ± 19	95 ± 16	.009*
Peak A velocity, cm/sec	64 ± 16	72 ± 18	.143
Average E/e' ratio	10.1 ± 3.7	10.1 ± 2.9	.588
s' velocity, cm/sec	7.3 ± 1.6	7.6 ± 1.4	.545
e' velocity, cm/sec	8.0 ± 3.3	9.1 ± 2.7	.091*
a' velocity, cm/sec	9.0 ± 2.6	8.3 ± 1.1	.174

BP, Blood pressure; IVS, interventricular septum; LVEDD, LV end-diastolic diameter; LVESD, LV end-systolic diameter; LVM, LV mass; PW, posterior wall; RWT, relative wall thickness.

Data are expressed as mean ± SD.

*P < .05.

Table 3 Comparison of global and segmental LV strain parameters between patients with AFD and age- and gender-matched normal control subjects

Strain parameter	Patients with AFD	Control subjects	P
	(n = 43)	(n = 43)	
LV GLS, %	-16.5 ± 3.8	-20.2 ± 1.7	<.001*
LV S SR, sec ⁻¹	-1.0 ± 0.5	-1.2 ± 0.1	<.001*
LV E SR, sec ⁻¹	1.4 ± 0.4	1.4 ± 0.3	.146
LV A SR, sec ⁻¹	0.9 ± 0.2	0.9 ± 0.2	.983
LV circumferential strain, %	-14.2 ± 4.1	-21.8 ± 1.5	<.001*
LV radial strain, %	46.5 ± 26.1	58.2 ± 6.1	.001*
Apical segment strain, %	-19.4 ± 4.7	-22.7 ± 2.1	<.001*
Mid segment strain, %	-16.5 ± 4.0	-20.7 ± 1.3	<.001*
Basal segment strain, %	-14.8 ± 3.7	-20.3 ± 1.1	<.001*
Apical segment S SR, sec ⁻¹	-1.26 ± 0.3	-1.35 ± 0.2	.088
Mid segment S SR, sec ⁻¹	-0.98 ± 0.2	-1.14 ± 0.1	.001*
Basal segment S SR, sec ⁻¹	-1.01 ± 0.2	-1.16 ± 0.1	<.001*
Apical segment E SR, sec ⁻¹	1.86 ± 0.7	1.67 ± 0.4	.203
Mid segment E SR, sec ⁻¹	1.22 ± 0.4	1.37 ± 0.3	.095
Basal segment E SR, sec ⁻¹	1.13 ± 0.4	1.33 ± 0.3	.015*
Apical segment A SR, sec ⁻¹	0.90 ± 0.4	0.85 ± 0.2	.886
Mid segment A SR, sec ⁻¹	0.83 ± 0.3	0.85 ± 0.2	.488
Basal segment A SR, sec ⁻¹	0.91 ± 0.2	0.88 ± 0.2	.437

Data are expressed as mean ± SD.

*P < .05.

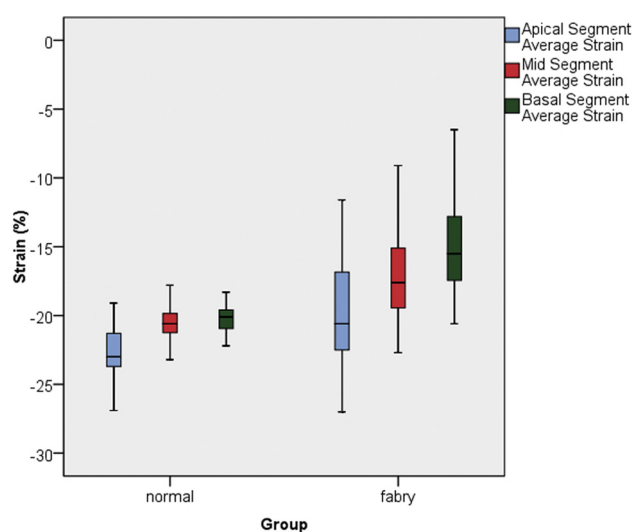
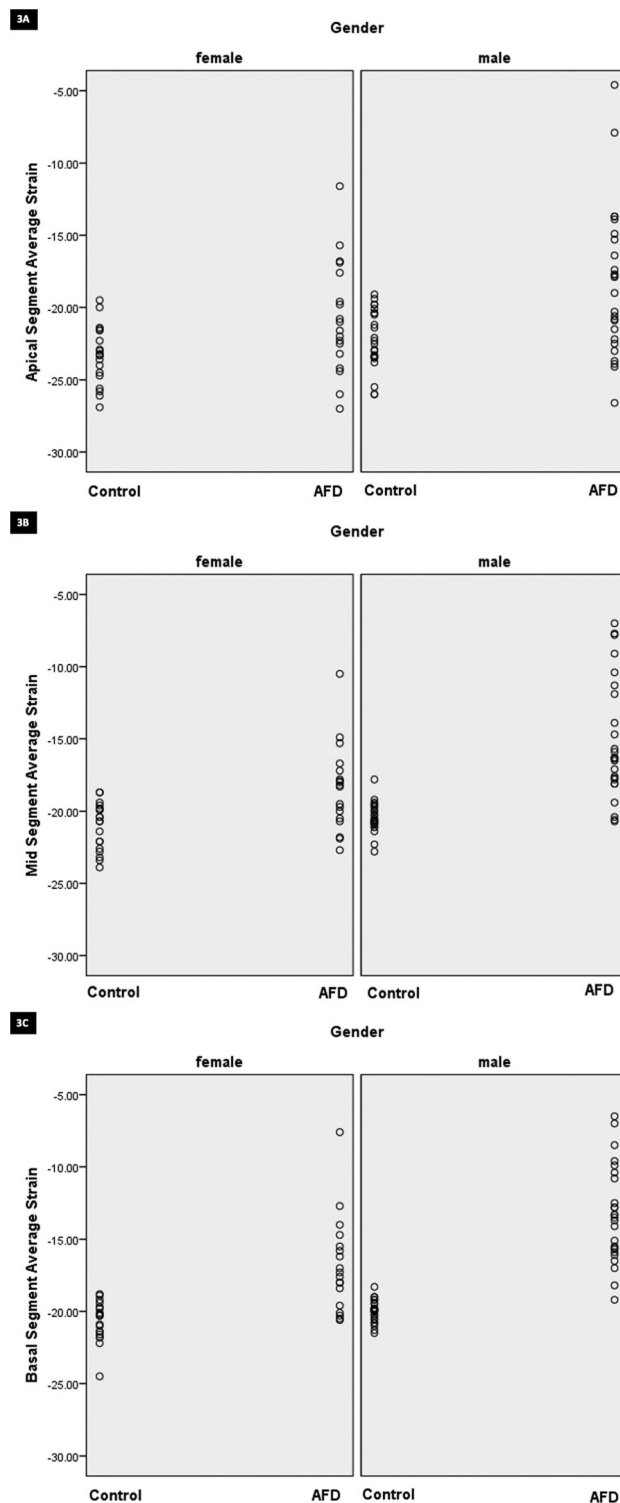
Table 4 Comparison of LV strain parameters between patients with AFD by gender

Strain parameter	Men	Women	P
	(n = 25)	(n = 18)	
LV GLS, %	-15.5 ± 3.9	-17.8 ± 3.2	.092
LV S SR, sec^{-1}	-0.9 ± 0.2	-1.2 ± 0.7	.085
LV E SR, sec^{-1}	1.3 ± 0.4	1.5 ± 0.3	.067
LV A SR, sec^{-1}	0.9 ± 0.3	0.9 ± 0.2	.115
LV circumferential strain, %	-13.9 ± 4.5	-14.6 ± 3.6	.731
LV radial strain, %	49.8 ± 21.4	42.2 ± 31.4	.104

Basal Strain Is Superior to GLS for Identifying Cardiac Involvement in Patients with AFD

Our finding of impaired basal LS is similar to a recent study including a smaller number of patients with AFD.²⁵ Impaired basal LS has been previously reported, corresponding with fibrotic LV segments (basal posterolateral segments) with late gadolinium enhancement on cardiovascular magnetic resonance imaging.²⁶ To our knowledge, no prior studies have analyzed whether basal LS is superior to GLS in identifying cardiac involvement in patients with AFD, as demonstrated by a significantly higher AUC by ROC curve analysis. Moreover, we found that basal LS was associated with MACE, with similar predictive value as increased LV wall thickness.

Subclinical cardiac dysfunction with impaired basal LS occurs in both female and male patients with AFD, even when LV wall thickness is normal. This is of particular importance in female patients with AFD, as current guidelines²⁷ recommend instituting ERT for men with AFD at diagnosis, while for female heterozygotes, the recommendation to start ERT is based on the presence of symptoms or evidence of organ involvement (i.e., increased LV wall thickness).^{28,29} Importantly, because up to 60% of patients with AFD

**Figure 2** Apical, mid, and basal segmental strain in normal cohort and patients with AFD.**Figure 3** (A) Apical segmental strain: comparison between age- and sex-matched normal control subjects and patients with AFD. (B) Midsegmental strain: comparison between age- and sex-matched normal control subjects and patients with AFD. (C) Basal segmental strain: comparison between age- and sex-matched normal control subjects and patients with AFD.

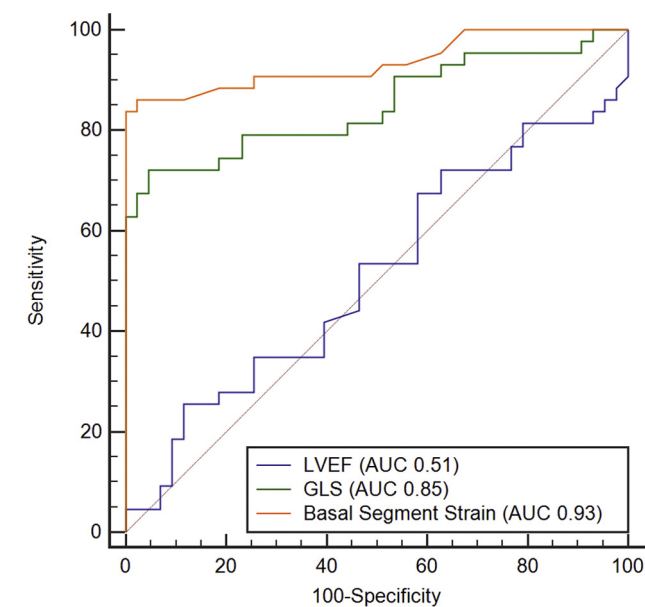


Figure 4 ROC curve analysis evaluating AUCs for LVEF (0.51), GLS (0.85), and basal segmental LS (0.93) in identifying cardiac involvement in patients with AFD.

may not have increased LV wall thickness at the time of diagnosis, conventional echocardiographic parameters have limitations in the assessment of cardiac involvement in early stages of AFD, and in this instance, segmental strain analysis may have particular utility.³⁰ Future studies would need to determine if initiating ERT when LV wall thickness is normal but basal LS is impaired is beneficial and improves clinical outcomes.

Basal LS in Other Phenotypes with LV Hypertrophy: Mechanisms in AFD

Reduction in basal LS has been reported in other conditions with increased LV wall thickness, including noninfiltrative conditions such as hypertension,³¹ aortic stenosis,³² and hypertrophic cardiomyopathy,^{33,34} and other infiltrative conditions such as cardiac amyloidosis.³⁵ In noninfiltrative conditions, reduction in basal LS has been attributed to a larger radius of curvature at the basal segments, with increased wall stress in accordance with Laplace’s law, with a consequent reduction in LS. In infiltrative pathologies, it would suggest earlier deposition in basal segments, with the reduction in basal LS related to the infiltrative process per se, as well as to an associated myocardial inflammatory response, in the early stage of the disease process. Subsequently, with disease progression, replacement fibrosis would occur, as reported by Niemann *et al.*³⁶ in patients with AFD. Thus, although in advanced disease impaired basal LS may be a surrogate of LV fibrosis, because of the lack of contemporaneous CMR data, we are unable to confirm these observations. We demonstrate an inverse correlation between LV wall thickness and basal LS, suggesting that impaired strain measurements may relate to replacement LV fibrosis, more evident in patients with AFD with increased LV wall thickness. This is in keeping with a proposed model of LV cardiomyopathy in AFD whereby patients initially have impaired LS, followed by impaired radial strain that accompanies an increase in LV wall thickness; this eventually progresses to myocardial fibrosis with further deterioration in longitudinal and radial strain.²⁰

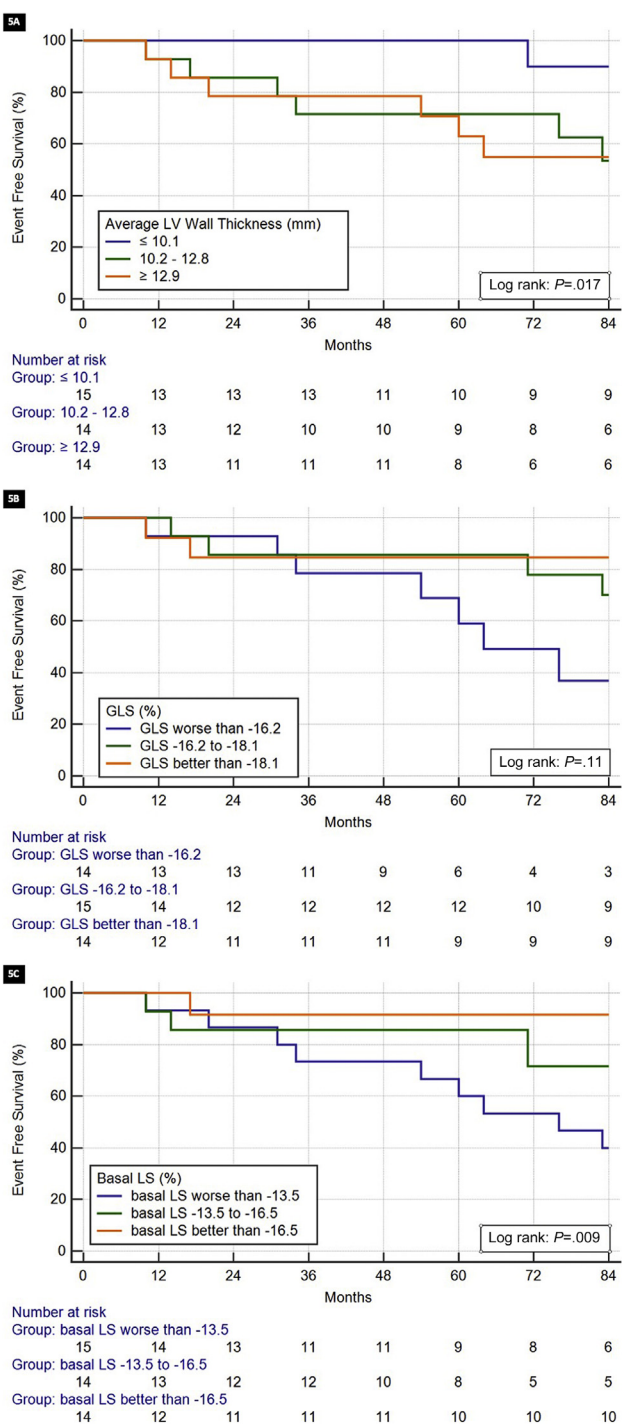


Figure 5 Kaplan-Meier analysis evaluating the effect of LV wall thickness (A), GLS (B), and basal LS (C) on event-free survival.

Limitations

This study had a modest sample size; however, it is inherently difficult to recruit a large number of patients with AFD at a single center, given the low disease prevalence. Despite this we were able to demonstrate differences in basal LS as well as its utility for identification of early cardiac involvement and association with MACE in patients with AFD. We appreciate that a significant number of patients with AFD were

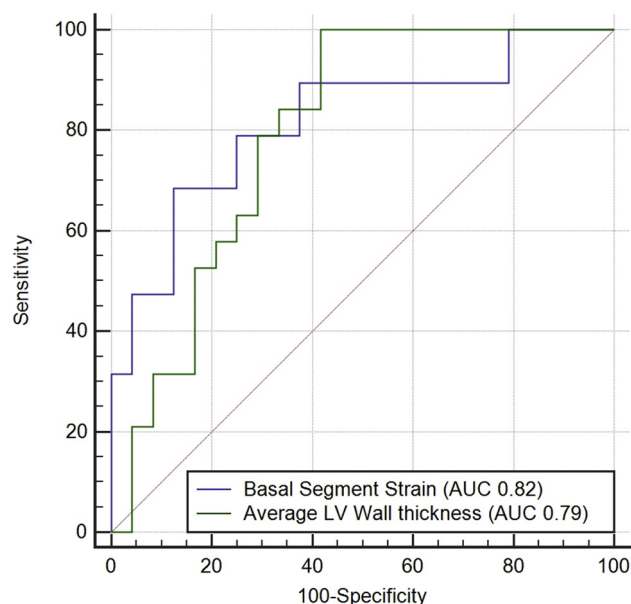


Figure 6 ROC curve analysis evaluating AUCs for LV wall thickness (0.794) and basal LS (0.818) in identifying MACE in patients with AFD.

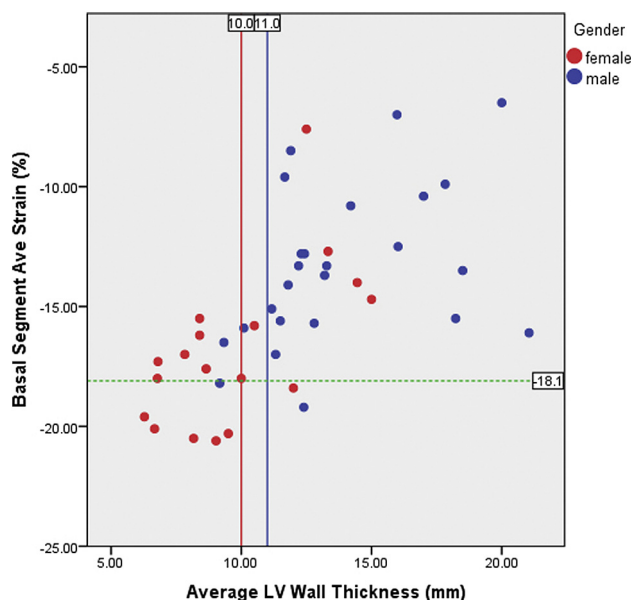


Figure 7 Scatterplot of basal LS versus average LV wall thickness on the basis of gender. Cutoff for increased LV wall thickness of ≥ 10 mm for women and ≥ 11 mm for men. Cutoff for -18.1% for basal LS (mean + 2 SDs). The scatterplot demonstrates that patients with AFD, particularly women with normal wall thickness, have impaired basal LS.

started on ERT after their initial TTE, but all initial transthoracic echocardiographic examinations were performed in ERT-naïve patients. Further studies would be required to establish the effect of ERT on changes in transthoracic echocardiographic markers (e.g., basal strain) and correlate them with clinical outcomes.

Because of the small sample size and low rate of MACE, we were unable to perform multiple comparisons when analyzing echocardiographic markers that predict MACE. Contemporaneous CMR was not available for the identification and correlation of fibrosis. Strain measurements may be affected by loading, and although in the normal range, blood pressure was higher in patients with AFD.

CONCLUSION

In patients with AFD, basal LS was impaired, demonstrating subclinical myocardial dysfunction. Female patients with AFD demonstrated impaired basal LS similar to hemizygous men, and this was present despite normal wall thickness in a significant number. Impaired basal LS was associated with worse clinical outcomes. Therefore, basal LS should be considered when evaluating patients with AFD for cardiac involvement and to evaluate prognosis. Future longitudinal studies are warranted to validate the role of basal LS in monitoring patients with AFD on ERT.

SUPPLEMENTARY DATA

Supplementary data related to this article can be found at <https://doi.org/10.1016/j.echo.2020.11.009>.

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Supplemental Table 1 Echocardiographic characteristics of the AFD patient subgroups on the basis of LV wall thickness

Parameter	Normal wall thickness (n = 14)	Increased wall thickness (n = 29)	P
Age, y	41.5 ± 13.1	45.2 ± 11.1	.218
IVS thickness, mm	8.3 ± 1.3	14.6 ± 3.1	<.001*
PW thickness, mm	8.2 ± 1.1	13.3 ± 3.0	.019*
Average wall thickness, mm	8.2 ± 1.2	14.0 ± 2.9	<.001*
RWT	0.34 ± 0.06	0.55 ± 0.17	<.001*
LVEF, %	60.9 ± 8.4	61.3 ± 8.5	.678
LVMI, g/m ²	75.7 ± 16.7	136.9 ± 36.6	<.001*
LVEDD, mm	48.4 ± 6.6	49.1 ± 5.6	.697
LVESD, mm	29.1 ± 6.0	28.4 ± 6.2	.534
Peak E velocity, cm/sec	95 ± 18	80 ± 19	.038*
Peak A velocity, cm/sec	67 ± 21	67 ± 15	.917
s' velocity, cm/sec	7.9 ± 1.3	7.2 ± 1.6	.189
e' velocity, cm/sec	9.8 ± 2.5	7.8 ± 3.1	.008*
a' velocity, cm/sec	8.0 ± 1.5	9.1 ± 2.2	.063
Average E/e' ratio	8.9 ± 2.5	10.7 ± 3.5	.103
LV GLS, %	-18.7 ± 2.3	-15.4 ± 3.9	.013*
LV S SR, sec ⁻¹	-1.2 ± 0.8	-0.92 ± 0.2	.165
LV E SR, sec ⁻¹	1.5 ± 0.3	1.2 ± 0.4	.015*
LV A SR, sec ⁻¹	0.9 ± 0.2	0.9 ± 0.3	.938
LV circumferential strain, %	-14.1 ± 3.1	-14.3 ± 4.5	.729
LV radial strain, %	40.5 ± 19.1	49.5 ± 28.8	.408

IVS, Interventricular septal; LVEDD, left ventricular end-diastolic diameter; LVESD, left ventricular end-systolic diameter; PW, posterior wall; RWT, relative wall thickness.

Data are expressed as mean ± SD.

*P < .05.

Supplemental Table 2 Echocardiographic characteristics of the AFD patient subgroups on the basis of LVMI

Parameter	Normal LVMI (n = 16)	Increased LVMI (n = 27)	P
Age, y	40.8 ± 12.5	45.9 ± 11.1	.076
IVS thickness, mm	8.7 ± 1.7	14.9 ± 3.1	<.001*
PW thickness, mm	8.5 ± 1.5	13.4 ± 3.0	<.001*
Average wall thickness, mm	8.6 ± 1.6	14.2 ± 2.9	<.001*
RWT	0.36 ± 0.07	0.56 ± 0.17	<.001*
LVEF, %	62.0 ± 9.0	60.7 ± 8.1	.94
LVMI, g/m ²	76.7 ± 15.8	140.8 ± 34.8	<.001*
LVEDD, mm	48.6 ± 6.2	49.0 ± 5.8	.980
LVESD, mm	29.0 ± 5.8	28.4 ± 6.3	.580
Peak E velocity, cm/sec	96 ± 17	79 ± 19	.007*
Peak A velocity, cm/sec	66 ± 21	68 ± 15	.669
s' velocity, cm/sec	7.9 ± 1.2	7.1 ± 1.7	.134
e' velocity, cm/sec	9.8 ± 2.4	7.7 ± 3.1	.002*
a' velocity, cm/sec	7.9 ± 1.6	9.2 ± 2.2	.030*
Average E/e'	9.0 ± 2.4	10.7 ± 3.7	.097
LV GLS, %	−18.6 ± 2.2	−15.2 ± 4.0	.007*
LV S SR, s ^{−1}	−1.2 ± 0.7	−0.9 ± 0.2	.490
LV E SR, sec ^{−1}	1.5 ± 0.3	1.2 ± 0.4	.072
LV A SR, sec ^{−1}	0.8 ± 0.1	0.9 ± 0.3	.497
LV circumferential strain, %	−14.1 ± 2.9	−14.3 ± 4.7	.641
LV radial strain, %	41.2 ± 20.7	49.8 ± 28.8	.407

IVS, Interventricular septal; LVEDD, left ventricular end-diastolic diameter; LVESD, left ventricular end-systolic diameter; PW, posterior wall; RWT, relative wall thickness.

Data are expressed as mean ± SD.

*P < .05.

Supplemental Table 4 Baseline clinical features of patients with AFD at the time of TTE (and prior to ERT)

	Value
Age, y	44 ± 11.9
Gender (male/female)	25/18
How diagnosed?	
Family screening	33
Renal biopsy (proteinuria or end-stage kidney disease)	7
Cardiac biopsy (LVH, syncope)	3
Hypertension	22 (51)
Hypercholesterolemia	24 (56)
Diabetes mellitus	4 (9)
Smoking status (nonsmoker/current smoker/ex-smoker)	27/13/3
Kidney function	
Creatinine	118 ± 135
CKD (stage 1/2/3/4/5)	1/2/5/2/6
NYHA functional class (I/II/III/IV)	39/4/0/0
Clinical features	
Cardiac symptoms (exertional dyspnea/syncope/palpitations)	4/3/3
Neurologic symptoms (acroparesthesia/hearing impairment/vertigo)	5/3/1
Gastrointestinal symptoms (diarrhea/proctitis)	8/1

CKD, Chronic kidney disease; LVH, left ventricular hypertrophy; NYHA, New York Heart Association.

Data are expressed as mean ± SD, number, or number (percentage).

Supplemental Table 3 Breakdown of MACE in Fabry disease

Event	Frequency in men with AFD	Frequency in women with AFD	Total events
Atrial fibrillation	4	2	6
Nonsustained ventricular tachycardia and insertion of AICD	2	2	4
Acute myocardial infarction	4	0	4
Cerebrovascular event	4	0	4
Exacerbation of heart failure	1	0	1
Total	15	4	19

AICD, Automatic implantable cardioverter-defibrillator.

Supplemental Table 5 Basal segmental longitudinal strain analysis of patients with AFD

Basal segment	Mean longitudinal strain, %
Basal inferoseptal	−13.1 ± 4.3
Basal anterolateral	−13.1 ± 6.3
Basal inferior	−16.9 ± 5.6
Basal anterior	−15.8 ± 6.0
Basal inferolateral	−14.7 ± 5.6
Basal anteroseptal	−14.7 ± 4.1

Kruskal-Wallis analysis showed a significant difference between the mean longitudinal strain values for the basal segments (P = .032).

Chapter 3 – Electrocardiographic characteristics and their correlation with echocardiographic alterations in Fabry Disease

In this chapter we report the results of our second study. The aim of this study was to characterise the ECG parameters in FD patients compared to healthy controls and to assess for gender-based differences in ECG parameters. Additionally, ECG alterations were correlated to structural (e.g. LV mass) and functional (e.g. LS) echocardiographic findings.

The findings showed that in contrast to female patients with FD, male patients more commonly had alterations in ECG parameters such as prolonged PQ interval, prolonged QRS duration and evidence of LVH on ECG. However, ECG was not as sensitive at detecting evidence of LVH as was echocardiography (with only 48% of patients with LVH on echocardiography meeting Sokolow-Lyon criteria for LVH on ECG). Patients with FD who had evidence of subclinical cardiac involvement (impaired basal LS) demonstrate LVH criteria on ECG. Whereas patients with FD who had increased LV wall thickness on echocardiography, represent a group with more advanced disease, and demonstrate more severe ECG abnormalities (LVH criteria as well as QRS prolongation).

These findings have clinical implications for screening for cardiac involvement and risk stratification in patients with FD. For instance, if a patient with FD presents with an ECG

abnormality (such as LVH criteria), but LV wall thickness is normal on TTE, this may prompt investigation for impairment in basal LS and if impaired, could indicate cardiac involvement.



Article

Electrocardiographic Characteristics and Their Correlation with Echocardiographic Alterations in Fabry Disease

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Abstract: Fabry disease (FD) is an X-linked disorder with α -galactosidase A deficiency. Males (>30 years) and females (>40 years) often present with cardiac manifestations, predominantly left ventricular hypertrophy (LVH). The aim of this study was to evaluate electrocardiographic (ECG) characteristics within FD patients to identify gender related differences, and to additionally explore the association of ECG parameters with structural and functional alterations on transthoracic echocardiography (TTE). Retrospective cross-sectional analysis of 45 FD patients with contemporaneous ECG and TTE was performed and compared to age and gender matched healthy controls. FD patients demonstrated alterations in several ECG parameters particularly in males, including prolonged P-wave duration (91 vs. 81 ms, $p = 0.022$), prolonged QRS duration (96 vs. 84 ms, $p < 0.001$), increased R-wave amplitude in lead I (8.1 vs. 5.7 mV, $p = 0.047$), increased Sokolow–Lyon index (25 vs. 19 mV, $p = 0.002$) and were more likely to meet LVH criteria (31% vs. 7%, $p = 0.006$). FD patients with impaired basal longitudinal strain (LS) on TTE were more likely to meet LVH criteria (41% vs. 0%, $p = 0.018$). Those with more advanced FD (increased LV wall thickness on TTE) were more likely to meet LVH criteria but additionally demonstrated prolonged ventricular depolarization (QRS duration 101 vs. 88 ms, $p = 0.044$). Therefore, alterations on ECG demonstrating delayed atrial activation, delayed ventricular depolarization and evidence of LVH were more often seen in male FD patients. Impaired basal LS, a TTE marker of early cardiac involvement, correlated with ECG abnormalities. Increased LV wall thickness on TTE, a marker of more advanced FD, was associated with more severe ECG abnormalities.

Keywords: Fabry disease; electrocardiogram; transthoracic echocardiography; LV hypertrophy; global longitudinal strain; basal longitudinal strain



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1. Introduction

Fabry disease (FD) is an X-linked lysosomal storage disorder due to α -galactosidase A enzyme deficiency, with resultant progressive accumulation of globotriaosylceramide (Gb3) in various organ tissues [1]. There are a variety of clinical manifestations including neurological, gastrointestinal, renal and cardiac in affected individuals [2]. Whilst many symptoms occur in childhood, cardiac symptoms usually do not present until the third or fourth decade [3].

FD patients with cardiac involvement develop several manifestations, typically with progressive increase in left ventricular (LV) wall thickness (on transthoracic echocardiography (TTE) or on cardiac magnetic resonance imaging (CMR)), significant life-threatening

arrhythmias or conduction defects, interstitial remodeling and myocardial fibrosis (as demonstrated on myocardial biopsy) [4]. Electrocardiographic (ECG) alterations have been reported including altered conduction, demonstrated by a prolonged PQ interval and QRS duration, as well as ECG features of LVH [5–7]. Prior studies of FD patients have shown a correlation between prolonged QRS duration on ECG and LV mass on TTE [8] and CMR [9]. Early diagnosis of FD is imperative as the evidence suggests that LVH may be prevented or may regress with early treatment with enzyme replacement therapy (ERT); moreover, in advanced FD, the response to ERT is poor [10]. Recently, there has been interest in identifying ECG parameters associated with cardiac involvement prior to the development of LVH [11,12].

As FD is an X-linked condition, traditionally female heterozygotes were considered carriers [13]; however, it is now widely accepted that female FD patients can develop cardiomyopathy, but with delayed onset and slower progression [14]. Variable findings have been reported in relation to ECG changes in females with FD [6].

In this retrospective cross-sectional study, our aim was to characterize ECG parameters in FD patients compared to healthy controls and evaluate gender related differences in ECG parameters. We further sought to examine the relationship between two widely available investigations and evaluate the correlation between ECG parameters with structural (e.g., LV mass) and functional changes (e.g., longitudinal strain) on TTE. In particular, we examined the relationship of ECG parameters with basal longitudinal strain, a marker of early cardiac involvement [15].

2. Materials and Methods

We retrospectively analyzed data from 45 genetically confirmed FD patients (mean age 42 yrs, 26/45 males) from the Department of Genetic Medicine at Westmead Hospital between 2009–2014 (baseline characteristics in Supplementary Table S2). The majority (38) of patients had contemporaneous TTE and ECG performed prior to ERT therapy; whilst only 7 patients had already been commenced on ERT at the time of their TTE and ECG. Subsequently, a further 19 FD patients received ERT based on current guidelines in Australia. FD patients were compared to age and gender matched healthy controls from a departmental database, with no documented cardiovascular disease or risk factors and were not on any medications. Ethical approval was obtained from the South West Sydney Health District Human Research Ethics Committee.

2.1. Electrocardiography

Standard 12-lead ECGs were performed, recorded at 50 mm/s with an amplitude of 1 mV/10 mm. An average of 3 measurements were performed manually with the aid of calipers for all parameters. Figure 1 demonstrates the ECG parameters that were assessed including PQ interval (average of PQ interval in lead II and V1), P-wave duration (average of P-wave duration in lead II and V1), QRS duration (in lead II), R wave amplitude in lead I, Sokolow–Lyon index (sum of voltage of S wave in V1/V2 and R wave in V5/V6) using the previously described criteria and the Modified Cornell Index (measuring R wave amplitude in aVL with patients meeting LVH criteria if >12 mm) [16]. PQ interval and P-wave duration were corrected to a heart rate of 60 by dividing intervals by the heart rate and multiplying by 60. ECGs were assessed for meeting LVH (Sokolow–Lyon criteria) and right bundle branch block (RBBB). ECG parameters were compared between the following groups: FD patients vs. controls (Table 1A); male FD patients vs. male controls (Table 1A); female FD patients vs. female controls (Table 1A); and male FD patients vs. female FD patients (Table 1B). We examined ECG findings in subgroups of FD patients based on TTE criteria: increased LV wall thickness vs. normal LV wall thickness, impaired GLS vs. normal GLS and impaired basal longitudinal strain (LS) vs. normal basal LS (Supplementary Table S1).

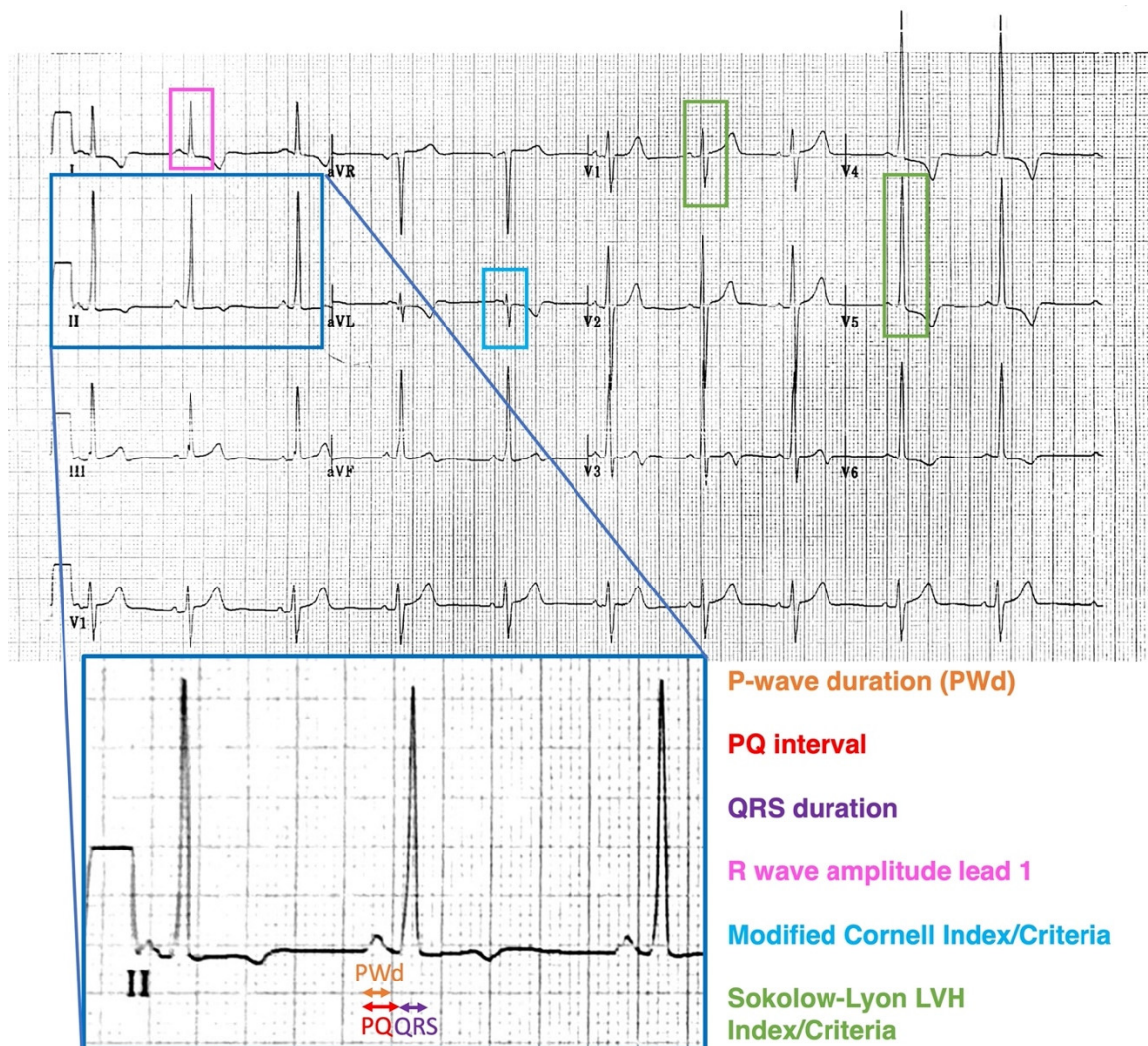


Figure 1. ECG parameters assessed included PQ interval in lead II and lead V1, P wave duration (PWd) in lead II and V1, QRS duration in lead II, R wave amplitude in lead 1, Sokolow–Lyon index and criteria, Modified Cornell index and criteria and complete right bundle branch block criteria.

2.2. Echocardiography

A comprehensive TTE was performed using commercially available ultrasound systems (Vivid 7 or E9 ultrasound scanner; GE Healthcare, Horten, Norway) with a 2.5-MHz transducer; M-mode, 2D, color, Doppler and tissue Doppler imaging were obtained in accordance with American Society of Echocardiography (ASE) guidelines [17]. Digital images were stored for offline analysis. One patient was excluded from analysis due to poor image quality. An average of 3 measurements were used for all echocardiographic parameters. LV wall thickness was measured from 2D images from the parasternal long axis view in end diastole; FD patients were stratified based on ASE guidelines to have increased wall thickness if ≥ 11 mm for males and ≥ 10 mm for females [18]. LV mass was calculated using the Devereux method as per ASE criteria and indexed (LVMI) to body surface area (BSA) [19]; gender-based cutoffs for increased LV mass used were >115 g/m² for males and >95 g/m² for females. LVEF was calculated using Simpson's biplane method by measuring LV end diastolic and end systolic volumes from apical -4 and -2 chamber views. Biplane maximum left atrial volume at end systole was measured from the apical -4 and -2 chamber views and indexed to body surface area (LAVI). Peak E and peak A velocities were obtained from pulsed Doppler mitral inflow. Tissue Doppler velocities (e')

were measured from the septal and lateral mitral annulus. The ratio of E velocity over average of septal and lateral e' velocity was determined (E/e'). (Table 2).

Table 1. (A). Electrocardiographic characteristics of FD patients compared to age and gender matched normal controls. ECG parameters assessed are listed in the first column. Comparison is made between FD patients and matched controls; between male FD patients and matched male controls only; and between female FD patients and matched female controls only. (B) Comparison between the electrocardiographic characteristics of male FD patients and female FD patients. ECG parameters assessed are listed in the first column.

(A)									
	All FD Patients Compared to Matched Controls			Male FD Patients Compared to Matched Male Controls			Female FD Patients Compared to Matched Female Controls		
	FD [n = 45]	Controls [n = 45]	p-Value	Male FD [n = 26]	Male Controls [n = 26]	p-Value	Female FD [n = 19]	Female Controls [n = 19]	p-Value
Age (yrs.)	42	42	0.939	42	42	0.934	42	42	0.84
Corrected PQ Interval (ms)	150	137	0.051	169	148	0.027[†]	126	121	0.729
Corrected PWD (ms)	91	81	0.022[†]	105	88	0.008[†]	74	72	0.644
QRS duration (ms)	96	84	<0.001[†]	103	85	<0.001[†]	87	82	0.271
R wave amplitude lead I (mV)	8.1	5.7	0.047[†]	9.1	5.8	0.014[†]	6.7	5.6	0.885
Sokolow–Lyon index (mV)	25	19	0.002[†]	28.4	19.6	0.008[†]	21.5	17.7	0.065
Sokolow–Lyon LVH criteria	14/45 (31%)	3/45 (7%)	0.006[†]	11/26 (42%)	2/26 (8%)	0.009[†]	3/19 (16%)	1/19 (5%)	0.604
Modified Cornell Index (mV)	4.2	3	0.507	5.3	3.2	0.374	2.7	2.7	0.965
Modified Cornell LVH criteria	Apr-45	0/45	0.117	26-Apr	0/26	0.11	0/19	0/19	-
RBBB criteria	5/45 (11%)	2/45 (4%)	0.434	5/26 (19%)	2/26 (8%)	0.419	0/19	0/19	-
(B)									
ECG Parameters	Male FD [n = 26]			Female FD [n = 19]			p-value		
Age (yrs)	42			42			0.818		
Corrected PQ Interval (ms)	169			126			0.001[†]		
Corrected PWD (ms)	105			74			<0.001[†]		
QRS duration (ms)	103			87			0.004[†]		
R wave amplitude lead I (mV)	9.1			6.7			0.051		
Sokolow–Lyon index (mV)	28.4			21.5			0.043[†]		
Sokolow–Lyon LVH criteria	11/26 (42%)			3/19 (16%)			0.102		
Modified Cornell Index (mV)	5.3			2.7			0.164		
Modified Cornell LVH criteria	4/26			0/19			0.126		
RBBB criteria	5/26 (19%)			0/19			0.063		

Table 1. Cont.

	All FD Patients Compared to Matched Controls	Male FD Patients Compared to Matched Male Controls	Female FD Patients Compared to Matched Female Controls	
LVH on TTE		20/26 (77%)	7/19 (37%)	0.013 [†]
Impaired GLS on TTE		18/26 (69%)	10/18 (55%)	0.525
Impaired Basal LS on TTE		23/26 (89%)	11/18 (61%)	0.064

Intervals and voltage are expressed as Means. Categorical variables are expressed as frequency and percentage of occurrence in each group. [†] $p < 0.05$. PWD = P-wave duration; LVH = Left Ventricular Hypertrophy; RBBB = Right bundle branch block; GLS = global longitudinal strain; LS = longitudinal strain, FD = Fabry disease, ECG = electrocardiograph

2.3. Strain Analysis

2D global longitudinal strain (GLS) analysis was performed using customized offline computer software (EchoPAC, GE, Horten, Norway, Version 201), from zoomed apical LV 4-, 2- and 3-chamber views, obtained at high frame rates (>55 fps). The endocardial border was manually traced in end-systole and the width of the region of interest (ROI) was manually adjusted to track the myocardium. Strain from up to 2 segments could be excluded in one imaging plane; if >2 segments could not be adequately tracked, the strain measurement from this plane was omitted. Impaired GLS was defined as GLS worse than -18.0% [20] and impaired basal longitudinal strain (LS) was defined as basal LS worse than -18.1% [15].

2.4. Inter-Observer and Intra-Observer Variability

To assess intra-observer variability, TTE and ECG of 10 randomly selected patients were reviewed by the same investigator 4 weeks apart. Inter-observer variability was assessed by a second blinded investigator in the same 10 patients.

2.5. Statistical Analysis

Continuous variables are expressed as a mean, and categorical variables as a percentage. Non-parametric tests (Mann-Whitney) were performed to examine differences in means between FD patients and controls and between male and female FD patients. Chi-square analysis was performed to examine differences in categorical variables. Pearson's correlation was performed to evaluate associations between continuous variables. Statistical analyses were performed using Statistical Package for Social Sciences, version 22 (SPSS, Chicago, IL, USA) and MedCalc version 15.8 (MedCalc Software Ltd., Ostend, Belgium) with $p < 0.05$ considered statistically significant. Inter and intra-observer variability were evaluated using intra-class correlation (ICC) and the coefficient of variation (CoV).

Table 2. Comparison of clinical, echocardiographic and electrocardiographic markers of FD patients with normal LV wall thickness and normal basal LS; normal LV wall thickness with impaired basal LS; and increased LV wall thickness and impaired basal LS.

	Normal LV Wall Thickness & Normal Basal LS [<i>n</i> = 8] [Group A]	Normal LV Wall Thickness & Impaired Basal LS [<i>n</i> = 9] [Group B]	Increased LV Wall Thickness & Impaired Basal LS [<i>n</i> = 25] [Group C]
Clinical characteristics			
Age	31	46 *	45
Males	3 (38%)	3 (33%)	20 (80%)
ERT—no. on ERT at time of simultaneous ECG/TTE	0	1 (11%)	6 (24%)
ERT—total no. who were treated with ERT after ECG/TTE	1 (13%)	4 (44%)	21 (84%)
Albuminuria/Chronic kidney disease	0	1 (11%)	12 (48%)
Dyslipidaemia	1 (13%)	5 (56%)	15 (60%)
Diabetes mellitus	0	2 (22%)	1 (4%)
Arterial hypertension	0	2 (22%)	15 (60%)
Transient ischaemic attack / stroke	1 (13%)	2 (22%)	5 (20%)
Ischaemic heart disease	0	1 (11%)	2 (8%)
Echocardiographic parameters			
Average LV wall thickness (mm)	8.3	7.5	14.6 +
Left ventricular mass index (g/m ²)	73.4	76.9	141.9 +
LVEF (%)	61	60	61
Peak E (cm/s)	98	93	78
Peak A (cm/s)	63	64	66
E/A	1.61	1.63	1.26
Lateral e' (cm/s)	14	12	9 +
Septal e' (cm/s)	11	9 *	7
Average E/e'	8.22	9.68	11.0
LAVI (mL/m ²)	44.6	40.7	36.8
Valvular disease	Moderate MR—1 Mitral prolapse—1 (mild)	Mild MR—2 Mild TR—3	Moderate MR—3 Moderate PR—1 Mild MR—1 Mild TR—2 Mild AS—1
ECG parameters			
Corrected PQ Interval (ms)	138	146	160
Corrected PWD (ms)	85	87	98
QRS duration (ms)	90	88	102
R wave amplitude lead I (mV)	5.1	6.0	10.3+
Sokolow–Lyon index (mV)	20.7	22.6	28.0
Sokolow–Lyon LVH criteria	0/8	1/9 (11%)	13/25 (52%) +
Modified Cornell index (mV)	2.3	2.4	5.7
Modified Cornell LVH criteria	0/8	0/9	4/25 (16%)
RBBB criteria	0/8	1/9 (11%)	4/25 (16%)

Intervals and voltage are expressed as Means. Categorical variables are expressed as frequency and percentage of occurrence in each group. PWD = P-wave duration; LVH = Left Ventricular Hypertrophy; RBBB = Right bundle branch block; GLS = Global longitudinal strain; LS = longitudinal strain; LVEF = Left ventricular ejection fraction; LAVI = Left atrial volume index; MR = mitral regurgitation; PR = pulmonary regurgitation; TR—tricuspid regurgitation; AS = aortic stenosis, FD = Fabry disease, ECG = electrocardiograph. * $p < 0.05$ for Group A vs. Group B. + $p < 0.05$ for Group B vs. Group C.

3. Results

3.1. ECG in FD versus Controls

All FD patients were in sinus rhythm. Analysis by gender (Table 1A) showed that corrected PQ interval was longer in male FD patients (169 vs. 148 ms, $p = 0.027$) but not so for female FD patients (126 vs. 121 ms, $p = 0.729$). The corrected P-wave duration was longer in FD patients (91 vs. 81 ms, $p < 0.022$), but similar to the corrected PQ interval, was

significantly longer for male FD patients (105 vs. 88 ms, $p = 0.008$), but not so in female FD patients (74 vs. 72 ms, $p = 0.644$). The QRS duration was significantly longer in FD patients compared to controls (96 vs. 84 ms, $p < 0.001$) and again, QRS duration was significantly longer in male FD patients (103 vs. 85 ms, $p < 0.001$), but not so in female FD patients (87 vs. 82 ms, $p = 0.271$).

Amplitude of the QRS complex was higher in FD patients in lead I (8.1 vs. 5.7 mV, $p = 0.047$) and in the chest leads (25 vs. 19 mV, $p < 0.002$). When analyzed by gender, QRS amplitude was higher in male FD patients both in lead I (9.1 vs. 5.8 mV, $p = 0.014$) and the chest leads (28.4 vs. 19.6 mV, $p = 0.008$), but not for female FD patients (lead I: 6.7 vs. 5.1 mV, $p = 0.885$; chest leads: 21.5 vs. 17.7 mV, $p = 0.065$). FD patients were more likely to meet Sokolow–Lyon criteria for LVH (31% vs. 7%, $p = 0.006$) and male FD patients met LVH criteria more frequently versus controls (42% vs. 8%, $p = 0.009$), though not observed in female FD patients (16% vs. 5%, $p = 0.604$). There was no significant difference in the presence of RBBB in FD patients compared to controls (11% vs. 4%, $p = 0.434$).

3.2. Male FD vs. Female FD: ECG and Echocardiographic Correlates

The corrected PQ interval, P wave duration and QRS duration were significantly longer in male FD patients compared to female FD patients (Table 1B). The average voltage was higher in male FD patients compared to female FD patients in the chest leads (28.4 vs. 21.5 mV, $p = 0.043$) but not in lead I (9.1 vs. 6.7 mV, $p = 0.051$). Male FD patients met LVH criteria more often than females, though not statistically significant (42% vs. 16%, $p = 0.102$). Only male FD patients had RBBB when compared to females (5/26 vs. 0/19). Male FD patients were more likely to have increased LV wall thickness on TTE (77% vs. 37%, $p = 0.013$). Strain analysis demonstrated that male and female FD patients were as likely to have impaired GLS (69% vs. 55%, $p = 0.525$) and impaired basal LS (89% vs. 61%, $p = 0.064$).

3.3. Correlation of Electrocardiographic and Echocardiographic Markers

There was a significant correlation (Figure 2) between LAVI and the corrected PQ interval ($r = 0.42$, $p = 0.005$) as well as the corrected P-wave duration ($r = 0.48$, $p = 0.003$).

There was a significant correlation (Figure 2) between LVMI on TTE with QRS duration ($r = 0.30$, $p = 0.044$) and with Sokolow–Lyon index on ECG ($r = 0.39$, $p = 0.011$). Similarly, there were significant correlations between GLS and QRS duration ($r = 0.47$, $p = 0.001$) and Sokolow–Lyon index ($r = 0.54$, $p < 0.001$), as also between basal LS and QRS duration ($r = 0.59$, $p < 0.001$) and Sokolow–Lyon index ($r = 0.56$, $p = 0.004$).

3.4. ECG Characteristics in FD Patients with Altered TTE Parameters: Increased Wall Thickness, Altered GLS and Basal Segmental LS

Subgroup analysis of FD patients (Supplementary Table S1) showed that those with increased wall thickness on TTE ($n = 27$) had a significantly longer QRS duration (101 vs. 88 ms, $p = 0.044$) and higher QRS complex amplitude in lead I (9.9 vs. 5.4 mV, $p = 0.002$) but not so in the chest leads (27.7 vs. 21.8 mV, $p = 0.112$). As expected, FD patients with increased wall thickness on TTE were also more likely to meet LVH criteria on ECG (48% vs. 6%, $p = 0.003$).

FD patients with impaired GLS ($n = 28$) had higher QRS complex amplitude in lead I (9.7 vs. 5.7 mV, $p = 0.008$) and were more likely to meet LVH criteria on ECG (43% vs. 13%, $p = 0.049$). Similarly, FD patients with impaired basal LS ($n = 34$) had higher QRS complex amplitude in lead I (9.1 vs. 5.1 mV, $p = 0.014$) and were also more likely to meet LVH criteria on ECG (41% vs. 0%, $p = 0.018$).

We examined ‘stages’ of disease progression by dividing the FD patients into three groups: (1) EARLY (Group A): normal LV wall thickness and normal basal segmental LS; (2) INTERMEDIATE (Group B): normal LV wall thickness and impaired basal segmental LS; and (3) LATE (Group C): increased LV wall thickness and impaired basal LS. There were significant differences in ECG and TTE parameters (Table 2), albeit acknowledging

the relatively small numbers in groups 1 and 2. Two female FD patients with hypertension and mildly increased LV wall thickness (likely due to hypertension) with normal basal LS were excluded from this subgroup analysis.

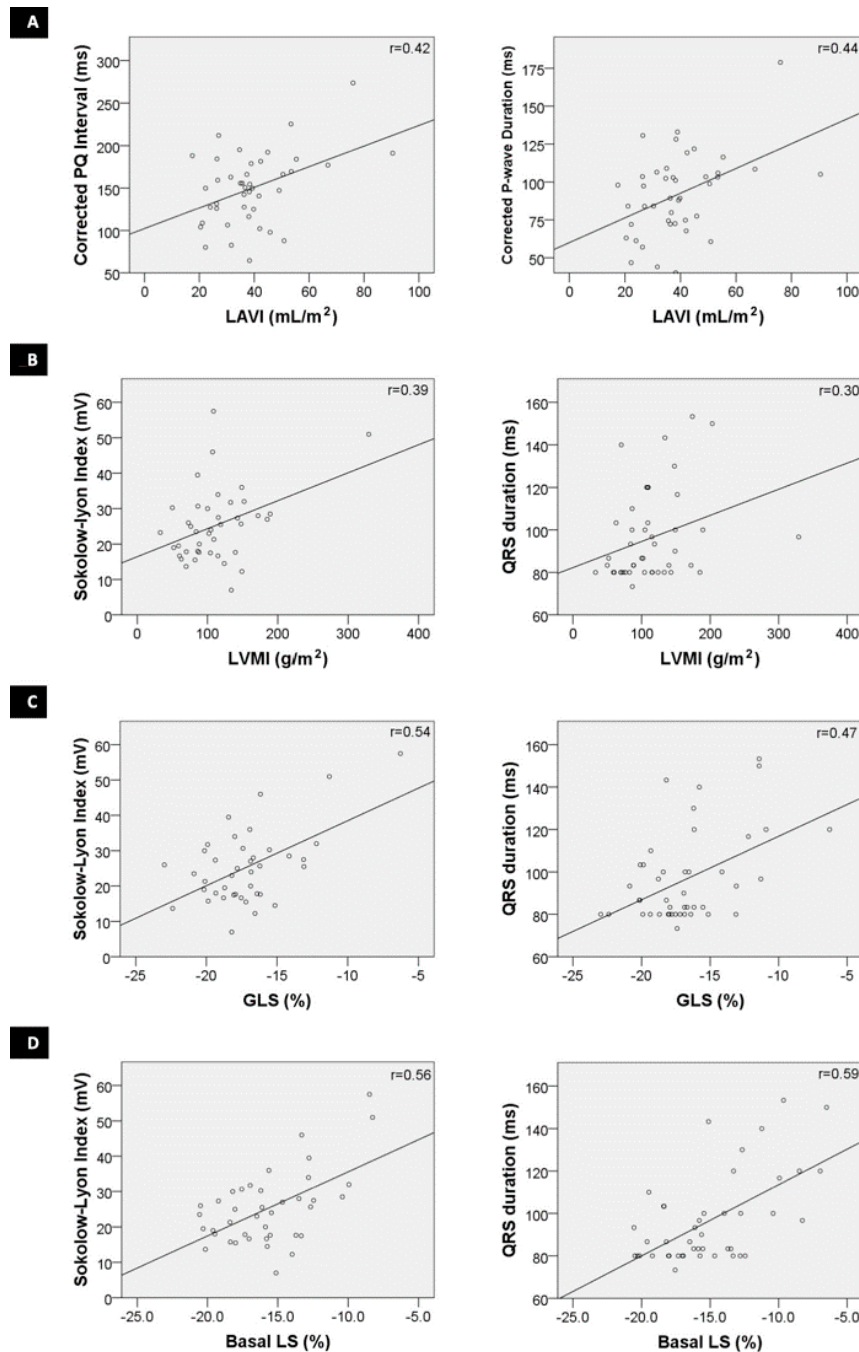


Figure 2. Correlation between electrocardiographic and echocardiographic parameters; (A)—Correlation between corrected PQ interval and maximum left atrial volume indexed to body surface area (LAVI) and between corrected P-wave duration and LAVI; (B)—Correlation between Sokolow–Lyon index and left ventricular mass indexed to body surface area (LVMI) and between QRS duration and LVMI; (C)—Correlation between Sokolow–Lyon index and Global Longitudinal Strain (GLS) and between QRS duration and GLS; and (D)—correlation between Sokolow–Lyon index and Basal Longitudinal Strain (LS) and between QRS duration and Basal LS.

3.5. Reproducibility of ECG Analysis

The ICC for inter-observer variability for the PQ interval was 0.90 (95% confidence interval [CI], 0.62–0.98; coefficient of variation (COV), 6.22%) whilst the intraobserver variability for the PQ interval was 0.92 (95% CI, 0.68–0.98; COV = 5.47%). The ICC for inter-observer variability for the QRS duration was 0.86 (95% CI, 0.45–0.97; COV = 6.32%) whilst the intraobserver variability for the QRS duration was 0.84 (95% CI 0.35–0.96; COV = 5.65%). The ICC for inter-observer variability for LVH Sokolow–Lyon index was 0.97 (95% CI 0.89–0.99; COV = 7.87%) whilst the intraobserver variability for LVH Sokolow–Lyon index was 0.99 (95% CI 0.95–0.99; COV = 5.31%).

We have previously reported inter- and intra-observer variability for TTE parameters GLS and segmental strain [15].

4. Discussion

In this single center cohort of FD patients, males more commonly had alteration in ECG parameters including prolonged corrected PQ interval (i.e., lengthened P-wave duration), prolonged QRS duration and evidence of LVH on ECG. These ECG alterations were not observed as frequently in female FD patients. Prolonged QRS duration and Sokolow–Lyon index correlated with traditional TTE abnormalities (increased LV wall thickness) but had a stronger correlation with novel TTE markers such as GLS and basal LS.

4.1. ECG Parameters in FD Males and Females

Our results support previous observations that significant ECG alterations are more likely to be present in FD males. Despite one study not finding significant differences in the PQ interval and QRS interval between FD males and females [6], the study of Niemann et al. evaluating 150 FD patients (63 males and 87 females; average age of 38 years), demonstrated that FD males were more likely to have prolonged QRS duration, higher Sokolow–Lyon index and RBBB criteria [9]. Additionally, we identified altered PQ interval, P wave duration and QRS duration in FD males. In general, in FD, cardiac involvement occurs earlier and more commonly in males [14]. Therefore, our observations suggest that FD males, albeit at similar or younger age than female FD patients were at a later stage in the disease course, with more males having increased wall thickness on TTE and more ECG alterations.

4.2. Alterations in Conduction: PQ Interval and P-Wave Duration

Interestingly, young patients with early-stage FD have been reported to have PQ interval shortening, as a result of P wave shortening [5,6], without evidence of an accessory pathway [21]; however, none of the patients in our study demonstrated this. Potential mechanisms for enhanced conduction velocity include accumulation of Gb3 in cardiomyocytes, increase in diameter of conducting cells and direct interaction between Gb3 and ion channels involved in action potential propagation [5,6]. In other glycogen storage disorders, it is thought that electrophysiological phenomena such as PQ interval shortening are directly related to glycogen storage in and around the atrioventricular node [22]. ERT normalizes the shortened PQ intervals and shortened P waves [23]. As FD progresses, the extent of infiltration worsens with macroscopic changes to the myocardium, causing PQ interval prolongation (corresponding to left atrial enlargement) [5]. This theory is supported by the correlation of LAVI to PQ interval in our cohort.

4.3. Alterations in Conduction: QRS Interval and Right Bundle Branch Block

Patients in early-stage FD have QRS interval shortening, likely a result of enhanced conduction velocity [5,6]. With disease progression, the QRS interval prolongs [7]. In patients with LVH (from any cause), prolonged QRS interval is often seen and is thought to be due to the increased time taken for left ventricular depolarization [24–26]. As previously shown by Kampmann et al. [8], our cohort also demonstrated prolonged QRS duration on

ECG, likely secondary to increased LV mass, considering almost two-thirds of our cohort demonstrated increased LV wall thickness on TTE (27/45).

Similar to previous studies, our cohort demonstrated right bundle branch block more commonly than left bundle branch block [9,27,28]. We hypothesize that accumulation of Gb3 occurs more often within the right bundle than within the left bundle in patients with FD. There is certainly histological evidence reported in the literature of vacuolization within myocytes with preferential extension into the right bundle [29].

4.4. ECG for Detecting LVH (Sokolow–Lyon Criteria)

With worsening cardiomyopathy in FD, LV wall thickness increases with an increased Sokolow–Lyon index on ECG [7]. FD males in our study showed evidence of LV hypertrophy with several meeting the Sokolow–Lyon criteria on ECG. This is largely attributable to male FD patients being at ‘a later stage’ of their disease, evidenced by 20/26 male FD patients having increased LV wall thickness on TTE.

Although 27/45 patients had increased LV wall thickness on TTE, only 13/27 (48%) met Sokolow–Lyon criteria for LVH on ECG. This is in keeping with previous studies that have shown that the sensitivity of ECG at detecting LVH varies depending on the method used but ranges from 20–55%, with specificity decreasing from 95% to 77% as sensitivity increases [25,26,30]. Therefore, even though ECG is a simple method of screening for LV hypertrophy in FD, it is not as sensitive as TTE.

4.5. Correlating Electrocardiographic and Echocardiographic Parameters

Namdar et al. have reported that PQ interval may be prolonged in FD due to left atrial enlargement [5]. We demonstrated a significant correlation between LAVI and both PQ interval and P-wave duration, supporting the concept that the PQ interval is prolonged due to delayed atrial depolarization secondary to left atrial enlargement.

Kampmann et al. analyzed 166 ECGs and TTEs of hemi- and heterozygous FD patients (44% males; 56% females; average age 35 years) and found a significant correlation between Sokolow–Lyon index and QRS duration with LVMI [8], similar to our findings. GLS identifies subclinical cardiac involvement in a variety of conditions [31]; and in FD, GLS can be impaired even with normal LV wall thickness [15]. Augusto et al. have recently described several phases of FD including a silent ‘accumulation stage’ where LV wall thickness is normal, with evidence of detectable infiltration on CMR with corresponding impairment in GLS on TTE [32]. A previous study that used a binary definition to classify patients into 2 groups—normal versus abnormal ECG (defined as any of the following: prolonged or shortened PQ interval; prolonged QRS duration; the presence of LVH criteria; or t wave inversion), also demonstrated correlation with impaired GLS [33].

We sought to further explore the relationship between ECG parameters and strain parameters. In our cohort of 45 FD patients, 28 (62%) had impaired GLS and 34 (76%) had impaired basal LS; whilst 27 (60%) had increased LV wall thickness on TTE (Supplementary Table S1). This would suggest that strain, in particular basal LS, is more sensitive for detecting cardiac involvement in FD (with strain impaired in FD patients with normal LV wall thickness). FD patients with impaired basal LS demonstrated ECG abnormalities such as Sokolow–Lyon LVH criteria, but not QRS prolongation. Whereas patients with increased LV wall thickness on TTE, represent a group with more advanced disease and more severe ECG abnormalities (Sokolow–Lyon LVH criteria as well as QRS prolongation).

To our knowledge, this is the first instance where ECG abnormalities have been correlated to impaired basal LS in FD patients. This has practical implications for patient screening; if for example there is evidence of LVH on an ECG, but the LV wall thickness is normal on TTE, this may prompt the clinician to investigate for cardiac involvement by assessing basal longitudinal strain. Other investigations including cardiac MRI or myocardial biopsy may be confirmatory.

Therefore, we have proposed a model which highlights these findings and demonstrates corresponding ECG and TTE findings that could be expected with disease pro-

gression in FD (Figure 3): In early-stage FD, LV wall thickness, GLS and basal LS are normal—ECG characteristics may include shortened PQ interval and QRS duration due to enhanced conduction (although this was not seen in our cohort probably due to most of our cohort being at a later stage in the disease process); In intermediate stage FD, LV wall thickness is normal although there may be evidence of impairment in longitudinal strain, particularly basal LS—in such patients the PQ interval and QRS duration lengthen so that they lie within the normal range and the Sokolow–Lyon index may be increased; In late-stage FD, LV wall thickness is increased and GLS as well as basal LS are impaired—ECG characteristics would include evidence of LVH (increased Sokolow–Lyon index) and prolongation of the PQ interval and QRS duration (with a predilection for development of RBBB).

		Early stage FD	Intermediate stage FD	Late stage FD
TTE	LV wall thickness	Normal	Normal	Increased
	Strain	Normal	Impaired basal LS Impaired GLS	Impaired basal LS Impaired GLS
	LAVI	Normal	Normal	Increased
ECG	Sokolow–Lyon index	Normal	Normal / Increased	Increased
	PQ interval P-wave interval	Short	Normal	Prolonged
	QRS duration	Short	Normal	Prolonged
	RBBB			May be present

Figure 3. Expected transthoracic echocardiographic (TTE) and electrocardiographic (ECG) changes with progression of Fabry Disease (FD). LAVI = maximum left atrial volume indexed to body surface area. LS = longitudinal strain. GLS = global longitudinal strain. RBBB = right bundle branch criteria.

4.6. Limitations

This study had a modest sample size; however, it is inherently difficult to recruit large numbers of FD patients at a single center, given the low disease prevalence. Despite this we were able to demonstrate differences in ECG characteristics in FD patients.

CMR was not performed at a contemporaneous time as the ECG and TTE. Hence correlation with CMR findings could not be reported meaningfully. 3D echocardiography that allows more accurate estimation of LV mass and cardiac volumes was not performed as several patients had TTE performed prior to 3D technology being widely available.

Hypertensive heart disease may mimic some of the structural changes observed in FD. 60% of FD patients had a history of hypertension; however, all were managed according to guidelines and blood pressure was well controlled except for 1 female FD patient who was hypertensive at the time of assessment (with a systolic blood pressure more than 140 mmHg). Hence it is likely that changes observed are more attributable to FD than coexistent hypertension.

Our study compared FD patients to healthy controls as opposed to those with hypertrophic phenotype (e.g., hypertensive heart disease). However, a previous publication [16] has reported ECG characteristics in varying types of hypertrophic phenotypes.

5. Conclusions

ECG abnormalities in FD patients are more common in male FD patients. Alterations in conduction present in FD include prolonged PQ interval, prolonged P-wave duration and prolonged QRS duration (particularly RBBB). Impaired longitudinal strain on TTE is an earlier marker of cardiac involvement in FD and is associated with LVH on ECG. However, increased LV wall thickness on TTE occurs at a later stage in the disease process and in such instances demonstrates LVH and prolonged ventricular depolarization on ECG.

Supplementary Materials: The following are available online at <https://www.mdpi.com/article/10.3390/jcdd9010011/s1>, Table S1: Electrocardiographic characteristics of FD patients in comparison to echocardiographic findings. ECG parameters assessed are listed in the first column. Comparison is made between FD patients with increased LV wall thickness and normal LV wall thickness; FD patients with impaired GLS vs. normal GLS; and between FD with impaired basal LS and normal basal LS, Table S2: Baseline clinical and echocardiographic characteristics of FD patients. Table S3: Comparison of clinical, echocardiographic and electrocardiographic markers of FD patients with normal LV wall thickness (cut off 13mm). and normal basal LS; normal LV wall thickness with impaired basal LS; and increased LV wall thickness and impaired basal LS.

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Chapter 4 – Conclusions and future directions

In conclusion, this thesis has identified that advanced echocardiographic techniques such as LS analysis can identify subclinical cardiac involvement in FD, particularly in females with FD even in the absence of associated LV hypertrophy; moreover, impairment in basal LS was associated with MACE. This may have implications for treatment for female FD patients. Further studies would need to assess whether initiating ERT has an effect on echocardiographic markers such as basal LS and if ERT improves clinical outcomes for patients with FD who have normal LV wall thickness but impaired basal LS. Additionally, future studies could correlate these findings contemporaneously with cardiac MRI.

This thesis has also evaluated how ECG parameters correlate to TTE findings in patients with FD at differing stages of the disease process. Future studies could assess whether ERT halts progression of TTE and ECG abnormalities and how they correlate contemporaneously with cardiac MRI.

A limitation of these studies was the lack of 3D echocardiographic data, as many of the echocardiograms did not have this data available. We did not have Gb3 levels in all patients and hence comparisons of ECG and echocardiographic parameters to Gb3 could not be performed.

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