Journal of Internal Medicine 2005; **258**: 225–230

Amyloid heart disease mimicking hypertrophic cardiomyopathy*

S. MÖRNER[1], U. HELLMAN[1,2], O. B. SUHR[1], E. KAZZAM[1,3] & A. WALDENSTRÖM[1]
From the [1]Department of Public Health and Clinical Medicine, Section of Cardiology, Heart Center, University Hospital, Umeå, [2]Department of Medical Biosciences/Medical and Clinical Genetics, University Hospital, Umeå, Sweden, and [3]Department of Internal Medicine, Faculty of Medicine and Health Sciences, United Arab Emirates University, Al-Ain, United Arab Emirates

Abstract. Mörner S, Hellman U, Suhr OB, Kazzam E, Waldenström A (University Hospital, Umeå, Sweden; United Arab Emirates University, Al-Ain, United Arab Emirates). Amyloid heart disease mimicking hypertrophic cardiomyopathy. *J Intern Med* 2005; **258:** 225–230.

Objective. To investigate the importance of transthyretin (TTR) gene mutations in explaining the phenotypic expression in patients diagnosed with hypertrophic cardiomyopathy (HCM) in northern Sweden.

Background. Hypertrophic cardiomyopathy is relatively common and often caused by mutations in sarcomeric protein genes. Mutations in the TTR gene are also common, one of which causes familial amyloid polyneuropathy (FAP), with peripheral polyneuropathy and frequently, cardiac hypertrophy. These circumstances were highlighted by the finding of an index case with amyloidosis, presenting itself as HCM. Initial rectal and fat biopsies did not show amyloid deposits. Later on, the patient was shown to carry a TTR gene mutation, and cardiac amyoloidosis was confirmed by myocardial biopsy. Only then was a repeated fat biopsy positive for amyloid deposits.

Design. Cross-sectional study.

Setting. Cardiology tertiary referral centre.

Subjects. Forty-six unrelated individuals with HCM and the index case were included. Common diagnostic criteria for HCM were used. The 46 patients with HCM were previously analysed for mutations in eight sarcomeric protein genes and the TTR gene was now analysed by denaturing high-performance liquid chromatography and direct sequencing.

Results. One mutation in the TTR gene (Val30Met) was found in three individuals and the index case.

Conclusions. Three of the 46 cases with HCM carried the Val30Met mutation, and were considered likely to have cardiac amyloidosis, like the index case. As a correct diagnosis of cardiac amyloidosis is mandatory for a potentially life-saving treatment, TTR mutation analysis should be considered in cases of HCM not explained by mutations in sarcomeric protein genes.

Keywords: familial amyloid polyneuropathy, genetics, hypertrophic cardiomyopathy, mutation.

Introduction

Hypertrophic cardiomyopathy (HCM) is characterized by left and/or right ventricular hypertrophy, with predominant involvement of the interventricular septum in the absence of other causes of hypertrophy, such as hypertension or valvular heart disease [1]. Typical morphological changes include myocyte hypertrophy and sarcomere disarray surrounding areas of increased connective tissue. The prevalence in the general population is about 0.2% [2]. The disease is genotypically and phenotypically heterogeneous, with a wide variety of clinical manifestations, ranging from asymptomatic individuals to severe symptoms and early death [1]. In approximately 55% of the cases HCM has been suggested to be familial, inherited in an autosomal dominant fashion [3]. More than 150 different

*Presented as a poster (abstract) at the American Heart Association congress in New Orleans, 7–10 November 2004.

mutations in 10 sarcomeric protein genes have been identified in families with HCM [4].

Patients with HCM often exhibit symptoms of dyspnoea, chest pain, palpitations and sometimes syncope. Atrial fibrillation and nonsustained ventricular tachycardia (NSVT) are relatively common arrhythmias. It is an important cause of sudden cardiac death in children and young adults (i.e. sudden death in young athletes). However, in the adult population, HCM has a relatively benign prognosis with an estimated annual mortality of ≈0.7% [5, 6].

Amyloidosis is caused by deposits of misfolded proteins derived from different plasma proteins. It may appear late in life as senile cardiac amyloidosis, where the amyloid deposits are derived from normal transthyretin (TTR) [7]. The heart may also be affected in systemic AL amyloidosis or, more rarely, in AA amyloidosis [8]. Cardiac amyloidosis may also be a feature of hereditary TTR amyloidosis, caused by mutations in the TTR gene. The most common is the TTR Val122Ile mutation, with a prevalence in African-Americans of 3.9% [9]. Several other mutations with predominant heart involvement are known [10]. However, cardiac amyloid deposits may also be noted in TTR mutations in which neuropathy is the predominant symptom, such as familial amyloidotic polyneuropathy (FAP), Portuguese type (TTR Val30Met). Over 1000 patients in close to 500 families with this mutation have been identified in Portugal, constituting the largest FAP patient population worldwide [11]. The second largest clustering is in northern Sweden, where more than 350 patients are known. In northern Sweden, the mean TTR Val30Met carrier frequency is 1.5% ranging from 0.0 to 8.3% in different subpopulations. There is a notable discrepancy between the regional distribution of the TTR Val30Met allele and the morbidity rate for FAP [12]. The typical phenotype of FAP is characterized by a progressive somatic and autonomic polyneuropathy, with complications from several other organ systems such as the gastrointestinal tract, kidneys, eyes and heart [13]. Typical cardiac symptoms are related to conduction defects [14, 15] or low voltage electrocardiogram (ECG) [15, 16], but some patients develop isolated progressive cardiac hypertrophy [16–18]. Recently, we have identified an index case presenting as HCM, without symptoms of polyneuropathy. Genotyping 3 years later revealed the TTR Val30Met mutation and a subsequent biopsy proved cardiac amyloidosis. This led us to further investigate the importance of TTR gene mutations in explaining the phenotypic expression in patients primarily diagnosed with HCM, from northern Sweden.

Material and methods

Index case

A 63-year-old previously healthy male without a history of hypertension, presented with sudden development of dyspnoea. Electrocardiogram showed a third-degree AV-block without signs of low voltage (total height of the QRS complex in the ECG <5 mm in the limb leads and <10 mm in the precordial leads) and prompt symptom relief was achieved after pacemaker implantation. Echocardiography revealed a nondilated left ventricle (LV) with a septal and posterior wall thickness of 21 and 15 mm respectively, consistent with the diagnosis of HCM (Fig. 1). Systolic function was normal and there was no sign of valvular disease. Three years later, symptoms of heart failure gradually developed. A chest X-ray showed cardiac enlargement and pulmonary congestion. Echocardiography showed hypertrophy of the same magnitude as before, with preserved ejection fraction. Cardiac catheterization revealed elevated right atrial, end diastolic left and right ventricular pressures. These data in

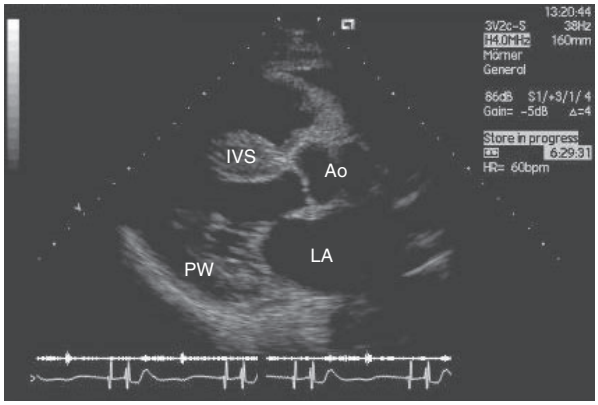

Fig. 1 Echocardiogram from the 68-year-old male index patient. Parasternal long axis view, showing marked septal hypertrophy and also hypertrophy of the left ventricular posterior wall. IVS, interventricular septum; Ao, aorta; PW, posterior wall; LA, left atrium.

combination with the initial presentation with AV-block and the fact that the patient originated from an endemic area for FAP, led to further investigation including genotyping, which was positive for the TTR Val30Met mutation. Still, no polyneuropathy or symptoms from the gastrointestinal tract were present. Electromyographic (EMG) and electroneurographic examinations did not disclose any abnormalities suggestive of an axonal neuropathy. Biopsies from abdominal fat and rectum did not show amyloid and no immunoglobulin light chains were found in the urine. A cardiac biopsy showed small myocytes and rich deposits of amyloid that reacted with an anti-TTR antibody. A second skin/subcutaneous fat biopsy showed amyloid deposits that reacted with an anti-TTR antibody.

Patients and clinical evaluation

The region of northern Sweden has a population of 883 000 inhabitants. Specialized cardiac care is provided by the Heart Center at Umeå University Hospital. The Hospital Discharge Register of the National Board of Health and Welfare in Stockholm was used to identify patients that had been hospitalized with HCM. Moreover, the doctors in charge of cardiology at all 12 hospitals in the region were contacted to obtain information about known patients with HCM. The criterion for the diagnosis of HCM was left ventricular hypertrophy demonstrated by echocardiography, with a wall thickness of ≥15 mm [19]. Exclusion criteria were arterial hypertension or ongoing antihypertensive treatment, significant valvular disease or known systemic disease (e.g. amyloidosis) capable of producing cardiac hypertrophy.

Forty-six unrelated individuals with HCM were included in the study, 25 men and 21 women with a mean age of 61.6 years (range: 26–80). Each individual had at least one living first-degree relative >18 years old (maximum 11 relatives). In 12 cases, all first-degree relatives were studied and in another 28 cases, as many as possible, but not all were studied. In six cases, no relatives were studied, mainly because they were asymptomatic and did not wish to participate. When family members had moved outside the region of northern Sweden, information about the phenotype was sometimes obtained through medical records from other hospitals, but no genotyping was performed. Cardiac amyloidosis was not suspected in any of the cases, or known in the family. In a previous study, of the same 46 cases, eight sarcomeric protein genes known to cause HCM were analysed: the cardiac β-myosin heavy chain; cardiac myosin-binding protein C; cardiac troponin T; α-tropomyosin; cardiac essential and regulatory myosin light chains; cardiac troponin I; and, cardiac α-actin gene. It was found that 13 cases carried a sarcomere protein mutation [20]. In 32 of the remaining 33 cases, no family history of HCM was known and the disease was considered to be sporadic. The patients underwent physical examination including echocardiography (M-mode, two-dimensional and Doppler), 12-lead ECG, 24-h Holter ECG and genotyping. Echocardiographic evaluation was performed with an Acuson xp/10 or Acuson Sequioa ultrasound system (Acuson, Mountain View, CA, USA). Views of the heart were obtained from the parasternal, apical and subcostal positions. All measurements were done according to the standards of the American Society of Echocardiography [21]. Informed consent was obtained from each individual and the protocol was approved by the ethics committee of Umeå University.

Genetic analysis

DNA was extracted from peripheral blood leucocytes by standard protocol. Exons 2, 3 and 4 of the TTR gene were amplified by 'touch down' polymerase chain reaction (PCR) and analysed by denaturing high-performance liquid chromatography (DHPLC) [22]. Previously published intronic sets of oligonucleotide primers were used [23]. The PCR was followed by a heteroduplex formation step, where the PCR products were slowly cooled down from 95 °C to room temperature, at 1.5 °C min^{-1}. Heteroduplexes were resolved from the corresponding homoduplexes using the WAVE system (Transgenomic, San Jose, CA, USA), an automated HPLC with a DNA separation column. The WAVEMAKER™ (Transgenomic, San Jose, CA, USA) software was used to determine the optimal temperature for heteroduplex separation. The nature of the sequence variation was then determined by direct sequencing of the PCR product using both forward and reverse primers on an automated fluorescent DNA sequencer, ABI 377 (PE Applied Biosystems, Foster City, CA, USA).

Statistical analysis

Fisher's exact test was used to compare noncontinuous data expressed as proportions.

Results

One missense mutation was detected in three elderly sporadic HCM cases and in the index case, identical with the mutation found in Swedish patients with FAP (Table 1). A G→A transition in exon 2 of the TTR gene substitutes valine for methionine at residue 30 (TTR Val30Met). Another three individuals were found to have a known polymorphism (Gly6Ser) [24].

None of the patients with the TTR Val30Met mutation had symptoms of polyneuropathy or low voltage in ECG registrations. The index case and two of the other TTR Val30Met-positive cases had conduction disturbances and received a pacemaker, whilst none of the 43 HCM patients without this mutation had severe conduction disturbances ($P = 0.001$, Fisher's exact test).

In one TTR Val30Met carrier, rectal biopsy was negative for amyloidosis. In another case, vitreous opacities of the eye were found, but presence of amyloid deposits elsewhere was not investigated. None of the three TTR Val30Met-positive cases carried any mutation in eight known HCM genes previously studied [20]. Only the index case is still alive, the other three TTR Val30Met carriers having all died, within 8 years of HCM diagnosis. When the medical records of another 59 known patients with FAP in our hospital were reviewed, 11 cases (19%) with varying degree of polyneuropathy were also found to have a marked cardiac hypertrophy on echocardiography (LV wall thickness of ≥15 mm).

Discussion

Hypertrophic cardiomyopathy is relatively common in the general population. Therefore, such patients are expected to be seen by general practitioners as well as cardiologists. Mutations in 10 different sarcomere protein genes are well known causes of the disease. There is an ongoing discussion about the relationship between the specific mutation involved and prognosis. So far, only a few mutations have been recognized to convey a poor prognosis. Sometimes there may be difficulties in distinguishing HCM from other conditions, such as athlete's heart and hypertensive heart disease. In the present study, we demonstrate that cardiac amyloidosis can present itself with a phenotype resembling HCM (the index case). The other three TTR Val30met-positive cases, constituting 7% of the HCM patients in this study, were not biopsy-proven to have cardiac amyloidosis. However, one case had vitreous opacities, two cases had severe conduction disturbances, and none of them carried a sarcomeric gene mutation. Therefore, it seems highly likely that they also suffered from cardiac amyloidosis related to the TTR mutation. Cardiac involvement in AL amyloidosis carries an extremely bad prognosis, but the outcome in patients suffering from hereditary TTR amyloidosis is also bleak, with a survival of <10 years [25]. Therefore, TTR gene mutation analysis can be useful in some cases of HCM, as cardiac amyloidosis is progressive, potentially fatal and can be treated with liver and heart transplantation in certain patients.

The classical cardiac manifestation in amyloidosis is considered to be that of a restrictive cardiomyopathy. The distinction between hypertrophic and restrictive cardiomyopathy is not

Table 1 Clinical characteristics of patients with the transthyretin (TTR) Val30Met mutation

Gender	Age[a] (years)	Age at diagnosis (years)	Familial/sporadic	Symptoms	LA (mm)	IVSD (mm)	LVPWD (mm)	IVSD/LVPWD	LVEDD (mm)
Male[b]	68	63	Sporadic	Dyspnoea	39	21	15	1.4	44
Male	77	72	Sporadic	Dyspnoea	57	23	17	1.4	54
Female	77	70	Sporadic	Palpitations	42	20	10	2.0	45
Female	71	68	Sporadic	Dyspnoea	40	26	24	1.1	31

[a]Age at time of echocardiography.
[b]Index case.
LA, left atrium; IVSD, interventricular septum dimension in end diastole; LVPWD, left ventricular posterior wall dimension in end diastole; LVEDD, left ventricular diameter in end diastole.

always obvious from noninvasive investigations, because different ventricular filling patterns may occur in HCM and cardiac hypertrophy may be present in restrictive cardiomyopathy. None of the patients in our study showed low voltage ECG, which is otherwise found in 30–50% of patients with FAP [15, 16]. Even though our cases were found in an endemic area for FAP, more than 100 different TTR gene mutations are known worldwide, and for the majority, cardiac involvement is predominant [10].

It is interesting to note that amyloid in peripheral tissues in the index case was detected only after repeated biopsies. This issue has previously been addressed by O'Hara and Falk, showing that approximately 15% of patients with cardiac amyloidosis have a negative fat biopsy, suggesting that a biopsy from the clinically affected organ will sometimes be necessary [26]. In our index case, cardiac amyloidosis could be confirmed by myocardial biopsy. Thus, neither a negative skin/subcutaneous fat biopsy nor a negative rectal biopsy excludes systemic amyloidosis. This demonstrates one of few clinical situations where the usefulness of gene mutation analysis can be relevant in the management of patients with a relatively common disease state.

Conclusion

As a correct diagnosis of cardiac amyloidosis is mandatory for a potentially life-saving treatment, TTR mutation analysis should be considered in cases of HCM not explained by mutations in sarcomeric protein genes.

Conflict of interest statement

The authors have no financial or other conflict of interest to declare in relation to this work.

Acknowledgements

This study was supported by The Swedish Heart and Lung Foundation, The Medical Faculty at Umeå University, The Medical Research Council (13045-03A), the patients association FAMY/AMYL, The Heart Foundation of northern Sweden, The Swedish Society of Medicine and The European Society of Cardiology.

References

1 Towbin J, Roberts R. Cardiovascular diseases due to genetic abnormalities. In: Schlant RC, Alexander RW, eds. *Hurst's The Heart: Arteries and Veins*, 8th edn. New York, USA: McGraw-Hill Inc., 1994; 1725–59.
2 Maron BJ, Gardin JM, Flack JM, Gidding SS, Kurosaki TT, Bild DE. Prevalence of hypertrophic cardiomyopathy in a general population of young adults. Echocardiographic analysis of 4111 subjects in the CARDIA Study. Coronary artery risk development in (young) adults [see comments]. *Circulation* 1995; **92:** 785–9.
3 Maron BJ, Nichols PF, III, Pickle LW, Wesley YE, Mulvihill JJ. Patterns of inheritance in hypertrophic cardiomyopathy: assessment by M-mode and two-dimensional echocardiography. *Am J Cardiol* 1984; **53:** 1087–94.
4 Marian AJ, Roberts R. The molecular genetic basis for hypertrophic cardiomyopathy. *J Mol Cell Cardiol* 2001; **33:** 655–70.
5 Cannan CR, Reeder GS, Bailey KR, Melton LJ III, Gersh BJ. Natural history of hypertrophic cardiomyopathy. A population-based study, 1976 through 1990. *Circulation* 1995; **92:** 2488–95.
6 Cecchi F, Olivotto I, Montereggi A, Santoro G, Dolara A, Maron BJ. Hypertrophic cardiomyopathy in Tuscany: clinical course and outcome in an unselected regional population. *J Am Coll Cardiol* 1995; **26:** 1529–36.
7 Westermark P, Sletten K, Johansson B, Cornwell GG III. Fibril in senile systemic amyloidosis is derived from normal transthyretin. *Proc Natl Acad Sci U S A* 1990; **87:** 2843–5.
8 Kyle RA, Spittell PC, Gertz MA *et al.* The premortem recognition of systemic senile amyloidosis with cardiac involvement. *Am J Med* 1996; **101:** 395–400.
9 Jacobson DR, Pastore RD, Yaghoubian R *et al.* Variant-sequence transthyretin (isoleucine 122) in late-onset cardiac amyloidosis in black Americans. *N Engl J Med* 1997; **336:** 466–73.
10 Connors LH, Lim A, Prokaeva T, Roskens VA, Costello CE. Tabulation of human transthyretin (TTR) variants, 2003. *Amyloid* 2003; **10:** 160–84.
11 Sousa A, Coelho T, Barros J, Sequeiros J. Genetic epidemiology of familial amyloidotic polyneuropathy (FAP)-type I in Povoa do Varzim and Vila do Conde (north of Portugal). *Am J Med Genet* 1995; **60:** 512–21.
12 Holmgren G, Costa PM, Andersson C *et al.* Geographical distribution of TTR met30 carriers in northern Sweden: discrepancy between carrier frequency and prevalence rate. *J Med Genet* 1994; **31:** 351–4.
13 Andersson R. Familial amyloidosis with polyneuropathy. A clinical study based on patients living in northern Sweden. *Acta Med Scand Suppl* 1976; **590:** 1–64.
14 Eriksson P, Karp K, Bjerle P, Olofsson BO. Disturbances of cardiac rhythm and conduction in familial amyloidosis with polyneuropathy. *Br Heart J* 1984; **51:** 658–62.
15 de Freitas AF. The heart in Portuguese amyloidosis. *Postgrad Med J* 1986; **62:** 601–5.
16 Eriksson P, Backman C, Eriksson A, Eriksson S, Karp K, Olofsson BO. Differentiation of cardiac amyloidosis and hypertrophic cardiomyopathy. A comparison of familial amyloidosis with polyneuropathy and hypertrophic cardiomyopathy by electrocardiography and echocardiography. *Acta Med Scand* 1987; **221:** 39–46.

17 Hattori T, Takei Y, Koyama J, Nakazato M, Ikeda S. Clinical and pathological studies of cardiac amyloidosis in transthyretin type familial amyloid polyneuropathy. *Amyloid* 2003; **10:** 229–39.
18 Hongo M, Ikeda S. Echocardiographic assessment of the evolution of amyloid heart disease: a study with familial amyloid polyneuropathy. *Circulation* 1986; **73:** 249–56.
19 Richardson P, McKenna W, Bristow M *et al.* Report of the 1995 World Health Organization/International Society and Federation of Cardiology Task Force on the definition and classification of cardiomyopathies. *Circulation* 1996; **93:** 841–2.
20 Morner S, Richard P, Kazzam E *et al.* Identification of the genotypes causing hypertrophic cardiomyopathy in northern Sweden. *J Mol Cell Cardiol* 2003; **35:** 841–9.
21 Sahn DJ, DeMaria A, Kisslo J, Weyman A. Recommendations regarding quantitation in M-mode echocardiography: results of a survey of echocardiographic measurements. *Circulation* 1978; **58:** 1072–83.
22 Xiao W, Oefner PJ. Denaturing high-performance liquid chromatography: a review. *Hum Mutat* 2001; **17:** 439–74.
23 Nichols WC, Benson MD. Hereditary amyloidosis: detection of variant prealbumin genes by restriction enzyme analysis of amplified genomic DNA sequences. *Clin Genet* 1990; **37:** 44–53.
24 Jacobson DR, Alves IL, Saraiva MJ, Thibodeau SN, Buxbaum JN. Transthyretin Ser 6 gene frequency in individuals without amyloidosis. *Hum Genet* 1995; **95:** 308–12.
25 Plante-Bordeneuve V, Lalu T, Misrahi M *et al.* Genotypic-phenotypic variations in a series of 65 patients with familial amyloid polyneuropathy. *Neurology* 1998; **51:** 708–14.
26 O'Hara CJ, Falk RH. The diagnosis and typing of cardiac amyloidosis. *Amyloid* 2003; **10:** 127–9.

Correspondence: Stellan Mörner MD, PhD, Department of Cardiology, Heart Center, Umeå University Hospital, S-901 85 Umeå, Sweden.
(fax: +46(90) 137633; e-mail: stellan.morner@medicin.umu.se).