International Journal of Cardiology 177 (2014) 400–408

Contents lists available at ScienceDirect

International Journal of Cardiology

journal homepage: www.elsevier.com/locate/ijcard

Uncertain diagnosis of Fabry disease: Consensus recommendation on diagnosis in adults with left ventricular hypertrophy and genetic variants of unknown significance ☆

B.E. Smid [a], L. van der Tol [a], F. Cecchi [e], P.M. Elliott [f], D.A. Hughes [g], G.E. Linthorst [a], J. Timmermans [h], F. Weidemann [i], M.L. West [j], M. Biegstraaten [a], R.H. Lekanne Deprez [b], S. Florquin [c], P.G. Postema [d], B. Tomberli [e], A.C. van der Wal [c], M.A. van den Bergh Weerman [c], C.E. Hollak [a,*]

[a] Department of Endocrinology and Metabolism, Academic Medical Centre, University of Amsterdam, Amsterdam, The Netherlands
[b] Clinical Genetics, Academic Medical Centre, University of Amsterdam, Amsterdam, The Netherlands
[c] Department of Pathology, Academic Medical Centre, University of Amsterdam, Amsterdam, The Netherlands
[d] Department of Cardiology, Academic Medical Centre, University of Amsterdam, Amsterdam, The Netherlands
[e] Department of Clinical and Experimental Medicine, University of Florence, Italy
[f] Department of Cardiology, Heart Hospital, London, UK
[g] Department of Haematology, Royal Free & University College Medical School, London, UK
[h] Department of Cardiology, RadboudUMC, Nijmegen, The Netherlands
[i] Department of Cardiology, Comprehensive Heart Failure Centre, University Hospital Würzburg, Germany
[j] Department of Medicine, Dalhousie University, Halifax, Nova Scotia, Canada

ARTICLE INFO

Article history:
Received 29 August 2014
Accepted 14 September 2014
Available online 20 September 2014

Keywords:
Fabry disease
Left ventricular hypertrophy
Hypertrophic cardiomyopathy
Consensus
Diagnosis

ABSTRACT

Background: Screening in subjects with left ventricular hypertrophy (LVH) reveals a high prevalence of Fabry disease (FD). Often, a diagnosis is uncertain because characteristic clinical features are absent and genetic variants of unknown significance (GVUS) in the α-galactosidase A (GLA) gene are identified. This carries a risk of misdiagnosis, inappropriate counselling and extremely expensive treatment. We developed a diagnostic algorithm for adults with LVH (maximal wall thickness (MWT) of >12 mm), GLA GVUS and an uncertain diagnosis of FD.
Methods: A Delphi method was used to reach a consensus between FD experts. We performed a systematic review selecting criteria on electrocardiogram, MRI and echocardiography to confirm or exclude FD. Criteria for a definite or uncertain diagnosis and a gold standard were defined.
Results: A definite diagnosis of FD was defined as follows: a GLA mutation with ≤5% GLA activity (leucocytes, mean of reference value, males only) with ≥1 characteristic FD symptom or sign (neuropathic pain, cornea verticillata, angiokeratoma) or increased plasma (lyso)Gb3 (classical male range) or family members with definite FD. Subjects with LVH failing these criteria have a GVUS and an uncertain diagnosis. The gold standard was defined as characteristic storage in an endomyocardial biopsy on electron microscopy. Abnormally low voltages on ECG and severe LVH (MWT > 15 mm) <20 years exclude FD. Other criteria were rejected due to insufficient evidence.
Conclusions: In adults with unexplained LVH and a GLA GVUS, severe LVH at young age and low voltages on ECG exclude FD. If absent, an endomyocardial biopsy with electron microscopy should be performed.

1. Introduction

Fabry disease (FD; OMIM 301500 [2]) is an X-linked lysosomal storage disorder caused by a deficiency of α-galactosidase A (AGAL-A). Estimated birth prevalence range between 1:40.000 and 110.000 [3–5]. Over 670 mutations in the α-galactosidase A (GLA) gene have been described [6], mostly appearing in single families. Since the availability of enzyme replacement therapy (ERT) screening in newborns, high risk populations, as well as individual case finding is increasing [7–16]. These screening studies report a surprisingly high prevalence of FD in subjects with left ventricular hypertrophy (LVH) (range, 0–12%). However, while the pathogenicity of some GLA mutations is well described, the subjects identified through screening often have a GLA genetic variant/mutation of unknown significance (GVUS) [17–19].

Interestingly, most males with such GVUS demonstrate significant residual AGAL-A enzyme activity, in contrast to the absent or near

Abbreviations: FD, Fabry disease; LVH, left ventricular hypertrophy; HCM, hypertrophic cardiomyopathy; AGAL-A, alpha-galactosidase A; GLA, alpha-galactosidase A gene; MWT, maximal wall thickness; GVUS, genetic variant of unknown significance; ERT, enzyme replacement therapy; lysoGb3, globotriaosylsphingosine.

☆ All authors take responsibility for all aspects of the reliability and freedom from bias of the data presented and their discussed interpretation.

* Corresponding author. Tel.: +31 20 5666071; fax: +31 20 6917682.
E-mail address: c.e.hollak@amc.uva.nl (C.E. Hollak).

http://dx.doi.org/10.1016/j.ijcard.2014.09.001
0167-5273/

absent enzyme activity in classically affected males [5]. Moreover, most subjects identified through screening are lacking characteristic classical Fabry signs or symptoms such as neuropathic pain, angiokeratoma or cornea verticillata, but present with a single, non-specific Fabry sign such as cryptogenic stroke, proteinuria or LVH, all associated with other more common diseases [20]. Because these subjects have symptoms restricted to single organs, they were coined as cardiac, renal or late onset variants of the disease [21,22]. In addition, while classically affected males invariably have significant elevations in plasma globotriaosylsphingosine (lysoGb3), non-classical FD patients and subjects with a non-pathogenic GLA mutation, such as p.D313Y, have low or normal levels [23–25]. While some still consider the p.D313Y mutation pathogenic [26,27], it has been shown that this mutation results in a pseudo-deficiency of AGAL-A in plasma, with only minimally reduced enzyme activity in cell expression models [28]. Another example of advancing insight concerns the p.A143T mutation, which is frequently identified through screening studies. However, in subjects with this variant presenting with LVH or kidney failure, no characteristic Gb3 deposits were found in biopsies [17]. These examples show that the lack of unequivocal definitions for a definite FD diagnosis leads to difficult clinical dilemmas with a risk of misdiagnosis. Early diagnosis of a true FD patient is of great importance to offer adequate support, but prompt identification of those without FD is of equal importance to avoid distress in families and inappropriate initiation of ERT, an invasive and extremely expensive treatment.

As part of the Hamlet study [1], designed to address the uncertainties related to diagnosing FD, we aimed to gain international consensus on a diagnostic algorithm for adult subjects presenting with LVH (maximal wall thickness in diastole (MWTd) of >12 mm) with an uncertain diagnosis of FD, harbouring a GVUS in the GLA gene.

2. Methods

2.1. Delphi participants

We used a modified Delphi procedure [29] to gain a consensus. The voting panel consisted of internists with expertise in the diagnosis and general management of FD and cardiologists with expertise in FD cardiomyopathy.

2.2. Pre-selection of voting items

A proposal was made for definitions of a definite and uncertain diagnosis of FD, and the gold standard (by MB, CH, BS, and LT). A systematic review was performed to find criteria on electrocardiogram (ECG), cardiac magnetic resonance imaging (CMR) or echocardiography that could be used to either exclude FD (exit criteria) or confirm a diagnosis of FD (entry criteria). PubMed and EMBASE were searched from 1980 till October 2012 with the following search terms: Fabry disease, heart, cardiac, cardiomyopathy, cardiac hypertrophy, LVH, ECG, ultrasound and CMR, including synonyms and MesH terms. Included were peer reviewed English written studies in adult human subjects. Titles and abstracts were screened and cross-referencing was performed. Corresponding authors were contacted if additional clarification was required. Criteria qualified when they were directly compared to other subtypes of hypertrophic cardiomyopathies (HCM) [30] and when sensitivity and specificity could be calculated. We accepted an entry criterion for a diagnosis of FD only if there was a specificity of >90% (i.e. the presence of this criterion confirms a diagnosis of FD; there are no or only very few false positives) and an exit criterion only if the prevalence of this criterion was <10% in FD (i.e. the presence of this criterion in FD is very unlikely, and is specific for other subtypes of HCM).

2.3. Validation of pre-selected criteria

Validation of the pre-selected criteria in patients similar to those identified through screening for LVH is of importance, since the selected criteria from the literature were primarily based upon patients with classical FD versus controls. Criteria that are specific or sensitive in a classically affected group may not necessarily have similar diagnostic accuracy in non-classical FD patients. To determine specificity and sensitivity, the pre-selected criteria were applied to Dutch patients presenting with LVH only (LVH defined as interventricular septal wall thickness of ≥12 mm and/or left ventricular mass of ≥48 g/height in $m^{2.7}$ for females, and ≥51 g/height in $m^{2.7}$ for males [31]). These patients were divided into two groups. A 'positive group' consisted of patients presenting with LVH only and histological evidence of a specific storage pattern, or with a definite (classical) diagnosis based upon the following predefined criteria: a GLA mutation (defined as any abnormality found in the GLA gene) and ≤5% GLA activity (of the mean reference value in leucocytes, males only) with ≥1 characteristic FD sign or symptom (neuropathic pain, cornea verticillata, clustered angiokeratoma) or increased plasma (lyso)Gb3 (in the classical male range) or a family member with a definite diagnosis of FD carrying the same GLA mutation. A 'negative group' consisted of patients with unexplained LVH who did not fulfil the criteria of a definite diagnosis of FD and therefore have a GVUS in the GLA gene (defined as a variant/mutation in the GLA gene of unknown clinical significance) in whom a biopsy of an affected organ excluded FD, or expression studies showed AGAL-A pseudo deficiency (p.D313Y) in index patients. All data were gathered with (written) informed consent. Pre-treatment ECGs were retrospectively assessed by a single investigator (PP) using digitized ECGs and on-screen callipers with the ImageJ program (http://rsb.info.nih.gov/ij/). Data on the following parameters were retrieved from 3 consecutive sinus beats: heart rate, P wave duration, PQ-interval, QRS-duration and QT-interval, QTc [32], Sokolow–Lyon index to assess left ventricular hypertrophy and the sum of the QRS amplitudes in lead I + II + III < 1.5 mV [33,34] as well as a Sokolow–Lyon index of <1.5 mV [35] to assess low voltages. All available echocardiography and CMR reports (baseline and treatment) were retrospectively scored for the presence of pericardial effusion, left ventricular outflow tract obstruction (LVOTO) and late enhancement by gadolinium on CMR.

2.4. Delphi voting rounds

The procedure consisted of two voting rounds and a face-to-face meeting. During the first voting round panellists received the results of the systematic review and validation cohort. Through an online anonymous survey (Survey monkey) they could criticize the validity of the pre-selected criteria [36]. Comments and new criteria could be added. Results of the first round were reviewed, items were adapted or added and the results were provided during the second round. During the face-to-face meeting each criterion was discussed and adapted when necessary. We admitted the possibility for supplementary analyses in panellist's cohorts in case the panel would conclude that the level of evidence of the pre-selected criteria is insufficient.

2.5 Statistical considerations: selection of final items in diagnostic algorithm

In keeping with previous studies [37] we decided to accept criteria in the diagnostic algorithm only when at least 75% of the panel agreed, and none of the panellists disagreed (i.e. only two neutral votes were acceptable). To assess overall consensus, Cronbach's α was calculated [38], with 0 indicating no consensus and 1 full consensus. The following recommendations by Bland and Altman were applied: Cronbach's α should be above 0.9, but preferably above 0.95 for clinical applications [38]. SPSS version 19 was used for statistical analyses.

3. Results

3.1. Delphi procedure and participants

Nine FD experts were invited to participate; seven FD experts (FC, PE, DH, JT, GL, FW and MW) completed all three rounds. At the face-to-face meeting, five experts were present and two were involved by telephone.

3.2. Pre-selection of voting items: systematic review

To preselect voting items proposed to the panel, a systematic review was performed. Our search retrieved 140 articles of which 88 were excluded (Supplementary Fig. 1). From the remaining 52 articles, 9 entry or exit criteria were pre-selected (Table 1). A summary of all articles reviewed and the results of the Dutch validation cohort were presented to the panel (online supplementary data [39-47,72-77,89-126]).

3.3. Voting items

Overall consensus on all voting items, measured by Cronbach's α, increased from 0.87 in round 1 to 0.97 and 0.99 in rounds 2 and 3, respectively.

3.3.1. Definitions of a definite and uncertain diagnosis of FD

There was 100% agreement that a diagnosis of FD in patients presenting with LVH only (defined as a MWT > 12 mm), cannot always be made by biochemical (AGAL-A activity) and/or GLA mutation analysis alone. To determine to whom the cardiac diagnostic algorithm would apply (i.e. the patients with an uncertain FD diagnosis) definitions of a definite and uncertain FD diagnosis were made (see Table 2).

A definite diagnosis of FD (i.e. classical FD) was defined as follows: a GLA mutation with ≤5% AGAL-A activity (of the mean of reference value in leucocytes [48], in males only) with either ≥1 characteristic FD symptom or sign or increased plasma (lyso)Gb3 (in the classical male range) or a family member with a definite diagnosis of FD carrying the