Chronic kidney disease and an uncertain diagnosis of Fabry disease: Approach to a correct diagnosis

Linda van der Tol a,⁎, Einar Svarstad b,c, Alberto Ortiz d, Camilla Tøndel e, João Paulo Oliveira f, Liffert Vogt g, Stephen Waldek h, Derralynn A. Hughes i, Robin H. Lachmann j, Wim Terryn k, Carla E. Hollak l, Sandrine Florquin m, Marius A. van den Bergh Weerman n, Christoph Wanner m, Michael L. West n, Marieke Biegstraatena, Gabor E. Linthorsta,⁎

a Department of Endocrinology and Metabolism, Academic Medical Center, Amsterdam Lysosome Center ‘Sphinx’, Amsterdam, Netherlands
b Department of Medicine, University of Bergen, Bergen, Norway
c Department of Medicine, Haukeland University Hospital, Bergen, Norway
d Unidad de Diálisis, ISF-Fundación Jimenez Diaz/UAM, IRSIN, Madrid, Spain
e Department of Pediatrics, Haukeland University Hospital, Bergen, Norway
f Medical Genetics, Hospital São João, Faculty of Medicine of University of Porto, Porto, Portugal
g Department of Nephrology, Academic Medical Center, Amsterdam, Netherlands
h Department of Medicine, Negotiation Trust, Ir University College London, UK
i Charles Dent Metabolic Unit, National Hospital for Neurology and Neurosurgery, London, UK
j Department of Internal Medicine, Division of Nephrology, Ghent University Hospital, Ghent, Belgium
k Department of Pathology, Academic Medical Center, Amsterdam, Netherlands
l Department of Medicine, Division of Nephrology, University of Würzburg, Würzburg, Germany
m Department of Nephrology, Dallhaus University, Halifax, Nova Scotia, Canada

A R T I C L E   I N   P R E S S

Molecular Genetics and Metabolism xxx (2014) xxx–xxx

Contents lists available at ScienceDirect

Molecular Genetics and Metabolism

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

ARTICLE INFO

Article history:
Received 3 July 2014
Received in revised form 13 August 2014
Accepted 13 August 2014
Available online xxxx

Keywords:
Fabry disease
Consensus
Diagnosis
Kidney pathology
Chronic kidney disease

ABSTRACT

Background and objectives: Screening for Fabry disease (FD), an X-linked lysosomal storage disorder, reveals a significant number of individuals with a genetic variant of unknown significance without classical FD manifestations; these variants in the α-galactosidase A gene often result in a high residual leukocyte α-galactosidase A and it is unclear whether these individuals suffer from FD. Therefore, a structured diagnostic approach is warranted. We present a diagnostic algorithm on how to approach adults with chronic kidney disease and an uncertain diagnosis of FD nephropathy.

Design, setting, participants, and measurements: A modified Delphi procedure was conducted to reach consensus among 11 FD experts. A systematic review was performed to identify possible criteria that could confirm or exclude FD nephropathy.

Results: The gold standard for FD nephropathy was defined as characteristic storage on electron microscopy (EM) in a kidney biopsy in the absence of medication that may induce similar storage. The suggested criteria to confirm FD nephropathy are as follows: ‘renal cysts’, ‘Maltese cross sign’, ‘immunohistochemical staining of Gb3 in urine’ and ‘high urinary Gb3’; and to exclude FD nephropathy: ‘absence of renal cysts’, ‘small kidneys’ and ‘high protein excretion’ were rejected because of low or uncertain specificity. Urinary Gb3 may be increased in other kidney diseases and there was no agreement on this criterion, although a third of the panel indicated that it is sufficient to diagnose FD nephropathy. The ‘Maltese cross sign’ and ‘high urinary Gb3’ were selected as red flags to suggest the possibility of FD nephropathy, but are not sufficient for a definite diagnosis of FD nephropathy.

Conclusions: In adults with chronic kidney disease, an α-galactosidase A gene variant and an uncertain diagnosis of FD, a kidney biopsy with EM analysis should be performed to confirm or reject the diagnosis of FD nephropathy. Other criteria currently cannot substitute for a biopsy in these cases.

© 2014 Elsevier Inc. All rights reserved.

1. Introduction

Fabry disease (McKusick 301500; FD) is an X-linked, lysosomal storage disease caused by deficient activity of α-galactosidase A (AGAL-A). The estimated birth prevalence has been reported to be between 1:40,000 and 1:170,000 for males [1–3]. More than 670, mostly private,
mutations/variants in the α-galactosidase A (GLA) gene have been described [4,5]. Classical FD is characterized by angiokeratoma, neuropathic pain, cornea verticillata, an- or hypohidrosis, and in males by absent or near absent AGAL-A activity and very high globotriaosylceramide (Gb3) and lysoGb3 in plasma and urine [6,7]. The kidneys, heart and central nervous system are often affected. Females can also be affected although, in general, organ involvement is less severe. 

Since the availability of enzyme replacement therapy (ERT) with recombinant AGAL-A (agalactosidase alpha, Shire HGT and agalsidase beta, Genzyme, a Sanofi company) an increasing number of studies have been performed in which high risk populations (i.e. with a non-specific symptom such as chronic kidney disease or left ventricular hypertrophy) as well as newborns are screened for FD, for a review see [8]. These screening studies as well as individual case finding revealed a higher than expected number of individuals with a GLA gene variant (defined as any alteration of the wild type GLA gene, irrespective of its suspected pathogenicity) and/or a deficiency of the AGAL-A enzyme. Approximately 80% of these identified individuals with a GLA gene variant are lacking characteristic FD features such as cornea verticillata [8]. Males who are identified by screening most often have a higher residual AGAL-A activity and normal or slightly elevated (lyso)Gb3 levels than classically affected males [6,7]. While the pathogenicity of some variants is well described, this is not the case for most of the GLA variants identified by screening. Thus, individuals often have an uncertain diagnosis of FD in the presence of a genetic variant of unknown significance (GVUS) [8,9]. For example, Terryn et al. showed that subjects identified by screening with the GLA variant p.A143T had no characteristic FD features and kidney biopsies of three individuals showed no Gb3 storage [10], while in classically affected males, kidney Gb3 deposits are already present during childhood [11]. In Taiwan the splice site variant IVS4+510C→A has a high prevalence of 1 in 1300 newborns, but adults harboring this variant did not demonstrate a classical FD phenotype. Yet, characteristic storage is reported in some tissue biopsies, but there is to date no systematic evaluation of structural changes in these individuals. Similar clinico-pathological variability has been reported for other GLA variants: e.g. N215S and R112H [12,13].

In the wake of our growing understanding of the spectrum of phenotypes associated with GLA variants, difficult dilemmas have emerged as these individuals may be misdiagnosed with FD [8]. Misdiagnosis of FD can cause distress and inappropriate initiation of costly ERT. Often, individuals with a GLA variant of unknown significance are simply diagnosed as FD patients and treated as such without further consideration of other contributing (cardiovascular) risk factors and the pathogenicity of the GLA variant [8]. Increased awareness for FD will inevitably lead to a higher than expected number of individuals with a genetic variant (designated as the main subject were included. The prevalence of kidney specific features in FD patients and control groups was calculated and specified by gender. Sensitivity and specificity were calculated wherever possible. Unfortunately, data to calculate specificity were often absent, since most studies did not include a control group with non-FD kidney diseases. As a surrogate, specificity was calculated based on prevalence in healthy controls. Candidate criteria to exclude (exit criteria) or confirm (entry criteria) the diagnosis of FD nephropathy were selected by the study team (LT, MB, GEL). We aimed to select features that were compared to other kidney diseases with a high specificity of ~90%. Because the number of criteria that fit this requirement was very limited, all studies were reported to the experts to assess the possible diagnostic criteria. The candidate items that were initially selected as possible entry criteria were subsequently selected as candidate criteria to serve as red flags. A red flag indicates that the presence of a feature makes a diagnosis of FD nephropathy more likely, but it is not sufficient for a definite diagnosis of FD nephropathy by itself. Details on the literature search and criteria are presented in the online supplement.

2.2. Systematic review and pre-selection of voting items

PubMed and Embase were searched for studies involving adults in peer reviewed journals that investigated kidney specific features in FD patients and/or healthy or diseased controls. Clinical trials as well as studies that investigated kidney specific features as the main subject were included. The prevalence of kidney specific features in FD patients and control groups was calculated and specified by gender. Sensitivity and specificity were calculated wherever possible. Unfortunately, data to calculate specificity were often absent, since most studies did not include a control group with non-FD kidney diseases. As a surrogate, specificity was calculated based on prevalence in healthy controls.

Candidate criteria to exclude (exit criteria) or confirm (entry criteria) the diagnosis of FD nephropathy were selected by the study team (LT, MB, GEL). We aimed to select features that were compared to other kidney diseases with a high specificity of ~90%. Because the number of criteria that fit this requirement was very limited, all studies were reported to the experts to assess the possible diagnostic criteria. The candidate items that were initially selected as possible entry criteria were subsequently selected as candidate criteria to serve as red flags. A red flag indicates that the presence of a feature makes a diagnosis of FD nephropathy more likely, but it is not sufficient for a definite diagnosis of FD nephropathy by itself. Details on the literature search and criteria are presented in the online supplement.

2.3. Statistical considerations

As customary for a Delphi procedure, criteria were accepted if ≥75% (n ≥ 9/11 panelists) of the panelists agreed, and no one disagreed [15]. Neutral votes were accepted if the other conditions were met. Data are presented as absolute numbers, or median and range. Cronbach’s α was calculated to assess consensus among experts on a scale of 0–1, where 1 indicated full consensus.

3. Results

3.1. Adopted results

The panel re-emphasized the results of a previous consensus study on general diagnostic criteria for FD. These criteria include biochemical data (residual leukocyte enzyme activity of AGAL-A, plasma Gb3 and lysoGb3) as well as clinical data (see Table 2) [14].

3.2. Systematic literature search and pre-selection of voting items

The literature search revealed 25 articles that studied kidney specific features in FD patients of which 4 also studied a control group with non-Fabry kidney diseases. Four features were selected as possible criteria to
confirm or exclude FD nephropathy for survey round one (Table 1). Details on the literature search are presented in the online supplement.

3.3. Panel

Thirteen FD experts (8 nephrologists and 5 general FD experts) were invited to participate. Two experts supported the initiative, but were unable to participate due to time constraints. The panel consisted of three internists (CH, DH, RL) and eight nephrologists (AO, JO, ES, CT (pediatric nephrologist), WT, CW, MW, SW). All panelists participated in all rounds.

3.4. Delphi procedure and diagnostic criteria

Rounds 1 and 2 were online questionnaires. Because some issues required further clarification, the questionnaire of the second round was repeated in an individual telephonic round (round 3). Overall consensus, measured by Cronbach’s α, increased from 0.85 in round 1 to 0.97 in round 3, indicating excellent consensus.

The panel fully agreed that the current diagnostic algorithm is developed to aid in the diagnosis for individuals with CKD in whom a GLA variant was already found, who do not develop clinical phenotype of classical FD (Table 2) [14]. Thus, these individuals have an uncertain diagnosis of classical FD. Further evaluations are needed, following the diagnostic algorithm.

Initially, criteria for CKD were discussed according to the level of proteinuria and estimated GFR. Based upon feedback from the expert panel, the 2012 ‘Clinical Practice Guideline for the Evaluation and Management of Chronic Kidney Disease’ by the KDIGO working group was adopted to define CKD [16].

Full agreement was reached on the current gold standard, defined as concentric multi-lamellated myelin bodies with a zebra like pattern (zebra bodies) with a periodicity of approximately 5 nm on electron microscopy (EM) [17], in a kidney biopsy, in the absence of medication use that may induce similar storage [18]. The biopsy should be assessed by an expert team on FD pathology. It was emphasized that in case of an uncertain diagnosis, a kidney biopsy should always be considered as it is currently the only assessment that can confirm or exclude the diagnosis in individuals with an uncertain diagnosis of FD nephropathy and CKD.

The preselected diagnostic criteria from the literature in round 1 are depicted in Table 1. The panel commented that renal cysts have a high prevalence, ranging from 5 to 41% in the general population [19–21]. It was suggested that the presence of medullary cysts may imply the presence of congenital kidney disease, while Ries et al. reported a 50% prevalence of parapelvic cysts among 24 FD patients [22]. Because of the non-specific nature and limited data, renal cysts were rejected by all experts as a diagnostic criterion.

The ‘Maltese cross sign’ and ‘imunohistochemical staining of Gb3 in urine’ may be helpful as diagnostic assessments [23], but results may be difficult to interpret. The differentiation between different Maltese cross types is challenging and the Maltese cross sign may also occur in patients with other causes for high urinary lipid content. The ‘Maltese cross sign’ criterion was rejected by 10/11 experts, 1 voted neither agree nor disagree. Staining urine sediment immunohistochemically for Gb3 is hampered by the natural presence of Gb3 in several cell-types and limited data is available on urinary Gb3 in other, non FD, kidney diseases. Following discussion, 9/11 experts rejected ‘imunohistochemical staining of Gb3 in urine’ as a criterion, 1 agreed and 1 voted neither agree nor disagree.

Table 2

Criteria for a definite diagnosis of FD. This definition was made to select those patients in whom there is no doubt that FD is present. If this definition is not met, FD cannot be ruled out, but further evaluation is needed to avoid labeling this individual with the wrong diagnosis. Some may have Fabry disease but others may have another cause for the diseased organ(s).

<table>
<thead>
<tr>
<th>Assessment</th>
<th>Entry criteria</th>
<th>Exit criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Literature</td>
<td>Kidney ultrasound/MRI/CT, Urine light microscopy</td>
<td>Renal cysts, Immunohistochemical staining of Gb3 in urine, Maltese cross sign in urine, High urinary Gb3 (in range of classical males)</td>
</tr>
<tr>
<td>Suggested by panel</td>
<td>Urine mass spectroscopy, Kidney ultrasound/MRI/CT</td>
<td></td>
</tr>
</tbody>
</table>

* Because the panel decided ‘high proteinuria’ is not a valid criterion for diagnostic purpose, no cutoff value was defined.

A variant in the GLA gene

A definite diagnosis of FD (classical phenotype)

Severely decreased or absent leukocyte AGAL activity (<5% of the normal mean) combined with a minimum of 1 of the following criteria:

- Fabry neuropathic pain; cornea verticillata; angiokeratoma; increased plasma lysosGb3 or Gb3 in the range of ‘classical’ FD males
- OR
- an affected family member with a definite diagnosis according to the criteria above

Uncertain FD diagnosis

The individual does not fit the criteria for a definite diagnosis of classical FD. Further evaluations are needed, following the diagnostic algorithm.

Gold standard

The gold standard for a diagnosis of FD in patients with an uncertain FD diagnosis, a CVUS and a non-specific FD sign is the demonstration of characteristic storage of the affected organ (e.g. heart, kidney, aside from skin) by electron microscopy analysis, according to the judgment of an expert pathologist.

Definitions:

- **Fabry neuropathic pain** fits the ‘characteristic clinical criteria’ if there is neuropathic pain in hands and/or feet, starting before age 18 or increasing with heat and/or fever. Quantitative sensory testing (QST) reveals a decreased cold detection threshold and the intradermal nerve fiber density (IENFD) is decreased. There is no other cause for Fabry neuropathic pain.
- **Angiokeratoma** fit the ‘characteristic clinical criteria’ if they are clustered and present in characteristic areas: bathing trunk area, lips, and umbilicus. There is no other cause for angiokeratoma.
- **Cornea verticillata** fits the ‘characteristic clinical criteria’ if there is a whorl like pattern of corneal opacities. There is no other cause (medication induced, among other: amiodarone, chloroquine).

Please cite this article as: L. van der Tol, et al., Chronic kidney disease and an uncertain diagnosis of Fabry disease: Approach to a correct diagnosis, Mol. Genet. Metab. (2014), http://dx.doi.org/10.1016/j.ymgme.2014.08.007
Three additional criteria were suggested (Table 1). First, ‘urinary Gb3’ (in the range of classical males), measured in whole urine, was suggested to confirm FD nephropathy. Others suggested that urinary and/or plasma Gb3 levels may be significantly increased in other diseases than FD [24,25]. Consequently, there was no agreement, although 4/11 of the panelists were convinced that this criterion, in the context of chronic kidney disease and a GLA variant, is sufficient to confirm FD nephropathy. The remaining 7 panelists rejected this criterion.

Second, it was postulated that small kidneys could possibly exclude FD nephropathy. It was discussed that in all causes for CKD, including FD, small kidneys occur at end stage disease. The most frequent cause of small kidneys is hypertension, which may also coexist with FD, although in the experience of the experts classical FD patients often have normal or low blood pressure in early phases of the disease. All panelists rejected ‘small kidneys’ as an exit criterion in the absence of hypertension, while 3/11 experts agreed that in the presence of hypertension, small kidneys exclude FD nephropathy. As a result, the ‘small kidneys’ criterion was not included in the diagnostic algorithm.

Finally, ‘high level of proteinuria’ was suggested to be rarely found in FD patients and to serve as a possible exit criterion. Because this feature was not studied in the literature, we assessed the maximum 24 hour protein excretion in 102 (39 males) Dutch FD patients with a definite diagnosis of classical FD, in accordance with previously defined criteria (Table 2) [14]. Median total protein excretion was 0.31 g/24 h (range 0.07–4.8), 0.62 g/24 h in males (0.13–4.39) and 0.24 g/24 h (0.07–4.80) in females. Few patients had proteinuria in the nephrotic range (>3 g/24 h) (see Fig. 1 and Table 3). An applicable cutoff value was debated and different opinions were shared by the experts. Most experts reported that some FD patients can have very high proteinuria. Proteinuria can also be influenced by concomitant diseases, e.g. hypertension. Because of limited data and absence of a cutoff value, this exit criterion was rejected by most experts (8/11). One voted neither agree nor disagree and 2 experts agreed that high proteinuria could serve as an exit criterion.

The ‘Maltese cross sign’ and ‘high urinary Gb3’ were depicted as red flags, indicating that the presence of these features raises the suspicion of FD nephropathy, but further assessments are mandatory. It was deemed not necessary to perform these 2 assessments in all individuals with an uncertain diagnosis of FD nephropathy.

Ultimately, the diagnostic algorithm was constructed (Fig. 2).

4. Discussion

An international panel of experts agreed on the diagnostic approach for individuals with CKD and an uncertain diagnosis of FD nephropathy. Group consensus was excellent, represented by a Cronbach’s α of 0.97. The experts indicated that a biopsy of the kidney with EM assessment is currently the only available tool that can reliably confirm or exclude FD nephropathy and should be considered in all patients without a classical pattern of disease manifestations, a GLA variant and CKD who have an uncertain diagnosis of FD (predefined criteria, Table 2 [14]).

This group endorses the diagnostic value of a biopsy of an affected organ (i.e. heart, kidney) to assess a true diagnosis of FD, as proposed in a previous study on individuals with LHV and an uncertain diagnosis of FD [14]. However, use of medication that may induce similar storage at any time during the medical history should always be excluded. The importance of morphological evidence in cases where the diagnosis of FD remains uncertain was stressed by the panel. In clinical practice, a nephrologist may not always be involved in the care of FD patients and a kidney biopsy is not routinely performed. However, we strongly recommend that those individuals with an uncertain diagnosis of FD nephropathy are assessed by a nephrologist and a kidney biopsy should be considered.

The storage pattern in FD, with characteristic lysosomal inclusions on EM, is specific for FD in the absence of medication (such as

![Fig. 1. Proteinuria in 102 (39 males) FD patients with a definite diagnosis of FD.](image)

![Fig. 2. Diagnostic algorithm for individuals with an uncertain diagnosis of Fabry disease (FD). *In the absence of medication use that may induce similar storage.](image)

Please cite this article as: L. van der Tol, et al., Chronic kidney disease and an uncertain diagnosis of Fabry disease: Approach to a correct diagnosis, Mol. Genet. Metab. (2014), http://dx.doi.org/10.1016/j.ymgme.2014.08.007
It is crucial that the kidney pathology is assessed by an expert team including a pathologist and nephrologist. Although similar storage to that of FD has been reported in other lysosomal storage diseases [26,27], the clinical presentation is distinct from that of FD. Thus, the specificity is not disputed in the clinical context of symptoms that are compatible with FD. Furthermore, it is encouraged that the conclusion of the clinico-pathological examination should also state if the amount of storage is compatible with the renal involvement of that particular patient. A detailed scoring system of the amount and characteristics of Fabry-specific storage in various kidney cells has recently been developed [28]. Because not all centers routinely perform EM on kidney biopsies, adequate routines and collaboration with specialized pathology departments are essential. The risks of a kidney biopsy are small when general contraindications are respected and complications most often fully resolve [29,30]. The indication and possible risks should be assessed carefully in each case. Clearly, the benefits of a correct diagnosis (either confirming or excluding FD nephropathy) are substantial. This study focused on diagnosis only, neither treatment indications nor the benefit of repeated renal biopsies to evaluate treatment for FD was discussed.

Our study was hampered by very limited and insufficient data on the prevalence of certain parameters in non-FD kidney diseases. Because of this, the specificity of most features could almost never be determined. Furthermore, expertise on some assessments is likely to be present only in few centers (e.g. staining of urinary cells for Gb3, interpretation of Maltese cross signs) and these assessments are therefore not widely applicable. The limited data emphasize the need to investigate the diagnostic accuracy of parameters in FD nephropathy and non-FD kidney disease in more detail, especially those tests that are minimally invasive.

The panel extensively discussed the value of increased levels of urinary Gb3, which is historically seen as a hallmark of FD. Data from previous studies indicate that Gb3 may be increased in plasma and/or urine in other diseases as well [24,25]. As early as 1978, reports on urinary glycosphingolipids in the urine of patients without sphingolipidoses were published [31]. The specificity of the diagnostic criteria that were part of this study and that (in)directly assess Gb3 in urine (‘immunohistochemical staining of Gb3 in urine’, ‘Maltese cross sign in urine’ and ‘high urinary Gb3 (in range of classical males)’) was therefore debated by the panel. It is quite likely that urinary Gb3 levels in the high range as usually seen in classical males can differentiate between classical FD and other diseases. The deacylated form of Gb3, lysoGb3, may be of even more importance as it can clearly differentiate classical patients from non-classical FD patients [6,7,32]. The ‘Maltese cross sign’ and ‘high urinary Gb3 (in the range of classical males)’ were selected as red flags. In individuals with an uncertain diagnosis of FD, who are not suitable for a kidney biopsy, these features may be helpful, but not conclusive, to address the diagnostic dilemma.

The current study does not serve to advocate screening for FD in cohorts or individuals with CKD. It is solely intended to provide the best possible knowledge on the diagnostic process in an individual with CKD and a GVUS in the GLA gene. Screening of individuals to identify FD patients is advocated by some to be able to initiate early treatment [33–35]. It is important to realize that the majority of individuals who are identified through screening do not present with the characteristic pattern of FD and possibly do not suffer from FD [8]. This causes unnecessary burden for the individual and family members. Moreover, for individuals with a non-classical FD phenotype with biopsy proven characteristic storage in the kidney, the natural history is yet unclear, and the effect of ERT has not been studied independently. While ERT may delay complications in patients with classical disease [36], a careful risk/benefit consideration should be made on an individual basis before initiating ERT in non-classical FD. Ideally, this group should be further studied within a well-defined study protocol.

In conclusion, a kidney biopsy with EM analysis should be considered for all individuals with CKD, a variant in the GLA gene and an uncertain diagnosis of FD, as it is currently the only diagnostic procedure to confirm or reject the diagnosis of FD nephropathy. With this approach, the unnecessary burden of inappropriate diagnosis of FD, counseling and treatment with costly ERT can be avoided. Equally important, individuals with true FD can be identified to initiate appropriate individualized counseling, treatment and family screening.

Disclosures

LT has received travel support and reimbursement of expenses from Actelion, Shire HGT or Genzyme.
MB, GL and CH have received travel support, honoraria for consultations and speaker’s fees from Actelion, Genzyme, Shire HGT, Protalix or Amicus. All fees are donated to the Gaucher Stichting or the AMC Medical Research for research support.
DH has received travel support, honoraria for consultations and speaker’s fees from Actelion, Genzyme, Shire HGT, Protalix or Amicus.
RL has received honoraria and consultancy fees from Genzyme, Shire and Actelion.
J0 has received travel support from Genzyme and Shire HGT, speaker’s fees from Genzyme, and research support from Genzyme.
AO has received travel support, reimbursement of expenses, speaker’s fees and honoraria for consultations from Genzyme and speaker’s fees from Shire HGT.
ES has received speaker’s fee and travel support from Genzyme and Shire.
CT has received travel support and speaker’s fee from Shire and Genzyme.
WT has received unrestricted research grants and speaker’s fees from Genzyme and Shire.
CW is a member of the European Board of Advisors to the Fabry Registry and has received travel support and speaker’s fee from Genzyme and Shire. A research grant was given to the institution by Genzyme.
SW has received research grants from Biomarin, Synavega, Shire, Genzyme and Amicus, and received travel support, honoraria for consultations and speaker’s fees from Genzyme, Shire HGT, GSK and Orphan-Europe.
MW received research funds, honoraria and consultant fees from Actelion Pharmaceuticals, Amicus Therapeutics, Genzyme (a Sanofi company), GlaxoSmithKline, Shire Human Genetic Therapies and Sumitomo Pharma.
SF, MBW and LV have none to declare.

Acknowledgments

This study was performed within the framework of the Dutch Top Institute Pharma (TIPharma, project number T6-504: ‘Fabry or not Fabry: valorization of clinical and laboratory tools for improved diagnosis of Fabry disease’). TIPharma is a non-profit organization that catalyzes research by founding partnerships between academia and industry. Partners: Genzyme, a Sanofi company; Academic Medical Center, University of Amsterdam; subsidizing party: Shire HGT.
http://www.tipharma.com/pharmaceutical-research-projects/drug-discovery-development-and-utilisation/hamlet-study.html. The industry partners had no role in the content of this manuscript, or selection of panel members. RHL and DH are supported by the National Institute for Health Research University College London Hospitals Biomedical Research Centre. AO is supported by Intensificatie en Redinren 012/0021 of the Fondo de Investigaciones Sanitarias—ISCII. We want to thank Ben JHM Poorthuis for his valuable advice on the biochemical aspects of Fabry disease and Bouwien E. Smid for her contribution during the preparation of the consensus document.

Abstracts of this study have been presented with a Poster at the ERA-EDTA congress 2014 and the WORLD symposium 2014.