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Renal Complications of Fabry Disease in Children

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Abstract

Fabry disease is an X-linked α -galactosidase A deficiency, resulting in accumulation of glycosphingolipids, especially globotriaosylceramide, in cells in different organs in the body. Renal failure is a serious complication of this disease. Fabry nephropathy lesions are present and progress in childhood while the disease commonly remains silent by routine clinical measures. Early and timely diagnosis of Fabry nephropathy is crucial since late initiation of enzyme replacement therapy may not halt progressive renal dysfunction. This may be challenging due to difficulties in diagnosis of Fabry disease in children and absence of a sensitive non-invasive biomarker of early Fabry nephropathy. Accurate measurement of glomerular filtration rate and regular assessment for proteinuria and microalbuminuria are useful, though not sensitive enough to detect early lesions in the kidney. Recent studies support the value of renal biopsy in providing histological information relevant to kidney function and prognosis and renal biopsy could potentially be used to guide treatment decisions in young Fabry patients. This review aims to provide an update of the current understanding, challenges and needs to better approach renal complications of Fabry disease in children.

Introduction

Fabry disease, first described by Johannes Fabry and William Anderson in 1898, is an X-linked α -galactosidase A deficiency which results in the failure to breakdown glycosphingolipids particularly globotriaosylceramide (abbreviated GL3 or Gb3). The build up of GL3 in the lysosomes and other cell compartments starts in fetal life [1], and leads to abnormal cellular function which in turn causes the symptoms of the disease. Since the disease is X-linked and, hence, more severe in males, much of the research on Fabry disease has focused on adult males. However, more recent research has clearly demonstrated that Fabry disease symptoms often start in childhood and are seen in both males and females [2]. Among the complications of Fabry disease, renal failure causes significant morbidity and mortality. While in most patients Fabry renal involvement is clinically silent in childhood, once becoming evident as overt proteinuria, Fabry nephropathy leads progressively to renal failure. Patients may not fully benefit from enzyme replacement therapy (ERT) if started when proteinuria is clinically manifest or glomerular filtration rate has already declined. Thus, early diagnosis and treatment of Fabry nephropathy in children may be critical to

preserve renal function. This review provides a general perspective about diagnosis of Fabry disease in children, available methods to evaluate renal function in Fabry patients and their challenges especially in young patients, the emerging role of kidney biopsy in assessment of Fabry nephropathy, and what is known about the effects of ERT on renal lesions in Fabry disease.

Diagnosis and Treatment of Fabry Disease in Children

Early diagnosis of Fabry disease is necessary for considering intervention to optimally prevent or ameliorate complications of the disease. Newborn screening of lysosomal storage diseases, including Fabry disease, is becoming more widespread and will help to identify patients early in life[2]. Currently however, due to the nonspecific nature of the initial symptoms, most children are diagnosed because of family history[2]. Nevertheless, there are also a few more specific features that can lead to diagnosis of Fabry disease, such as swirling pattern of the cornea seen with slit lamp examination which is present early in life in most but not all patients. Angiokeratomas, purple vascular lesions that may occur anywhere on the body, also highly suggestive of Fabry disease, may not occur until adolescence in males and may not be seen in some females. Neuropathic pain, primarily in hands and feet, is often the first symptom of Fabry disease, and may start as early as 2 years of age with a median age of 6 years for boys and 9 years for girls[2]. Other early symptoms include gastrointestinal distress, hypohydrosis, exercise intolerance, and increased heat sensitivity. Patients with non-classical symptoms or late-onset Fabry disease are more difficult to identify. Newborn screening will be especially helpful for early diagnosis of such patients[3].

Organ dysfunction develops later, and includes increased risk for stroke, hypertrophic cardiomyopathy, arrhythmias, and progressive renal disease. Renal disease is the most prominent organ dysfunction, and most male patients and a substantial fraction of female patients will eventually develop nephropathy without enzyme replacement therapy (ERT) [4]. Microalbuminuria is an indicator of renal dysfunction in Fabry patients but its prognostic value is, as yet, unclear[5]. Overt proteinuria may start as early as 10 years of age[6]. Although typically occurring by the third to fifth decade of life in men with Fabry disease[7], ESRD can occur as early as 16 years of age[6]. Patients with kidney failure appear to have concurrent involvement of other major organs [8]. While major organ dysfunction in children is rare, it does occur. Strokes have been noted as early as age 9 years (RJH, personal observation), cardiac arrhythmias and valve dysfunction are sometimes seen in adolescence[2]. While increased left ventricular mass was found to be common in children with Fabry disease compared to control subjects [2] established hypertrophic cardiomyopathy is not common in pediatric patients [9].

The diagnosis of Fabry disease is confirmed either by measurement of α -galactosidase A activity in leukocytes or by molecular studies for mutations in the gene. For females the diagnosis cannot be reliably made or ruled out without molecular studies, as even symptomatic patients often have normal serum α -galactosidase A levels[10]. Less frequently, the diagnosis has been confirmed by renal biopsy findings[11]. Late initiation of enzyme replacement therapy (ERT) may not provide optimal benefit[12]. Treatment at earlier stages of the disease has been suggested to improve prognosis. Early diagnosis is therefore important to ensure both clinical monitoring and early access to treatment. Unfortunately, there are no studies that have defined criteria for initiation of treatment in children. Until recently the monitoring of pediatric patients consisted primarily of following the same renal functional parameters commonly followed in adults with Fabry disease. These did not adequately assess the pain, hypohydrosis, or gastrointestinal symptoms that are the major complaints of pediatric patients. In addition the monitoring for kidney and

cardiac progression was not sensitive enough to be helpful in young patients. A recommended protocol for monitoring pediatric patients with Fabry disease has now been developed but has not yet been clinically validated[13]. This includes measures of quality of life, pain, and other non-specific symptoms as well as formal measures of GFR, proteinuria and cardiac function.

ERT aims to provide enough α -galactosidase A to decrease the symptoms and to delay or prevent major organ dysfunction. This is likely to require initiation of treatment before onset of potentially serious manifestations such as renal insufficiency or proteinuria. Agalsidase alfa (Replagal®, Shire Human genetic Therapies, Inc., Cambridge, MA) and agalsidase beta (Fabrazyme®, Genzyme corporation, Cambridge, MA) are two different forms of ERT currently available, both of which have been shown to be safe and well tolerated in pediatric Fabry patients[14, 15]. The infusions are given intravenously every 2 weeks at 1.0 mg/kg for agalsidase beta and 0.2 mg/kg for agalsidase alfa. While complete clearance of plasma and endothelial cells of the skin or kidney has been demonstrated using 1.0 mg/kg agalsidase beta, 0.2 mg/kg of agalsidase alfa generally achieves partial GL-3 clearance. Nevertheless, ERT studies in children were too small and the outcome measures not sensitive enough to clearly demonstrate improvement of symptoms or long term prognosis[14-16].

Assessment of Renal Function in Children with Fabry Disease

Generally, proteinuria, hypertension, increased serum creatinine and reduced glomerular filtration rate (GFR) are hallmarks of progressive kidney disease, and measurements of proteinuria and serum creatinine are the cornerstones in the assessment and monitoring of kidney function. However, these tests have low sensitivity in detecting early kidney damage and abnormal values are often late signs of kidney disease, already in the setting of at least partially irreversible structural damage. It is well known that renal morphologic damage is present in renal biopsies of Fabry patients many years and usually decades before manifest proteinuria and/or elevated serum creatinine is found[17, 18].

The lack of sensitive and non-invasive diagnostic and prognostic biomarkers of early kidney disease is a general challenge and focus of much research in nephrology, and this issue is especially relevant where therapeutic options like ERT are available and where the goal is to institute adequate intervention before irreversible structural damage prevails. None of the potential non-invasive biomarkers currently used in research programs, such as plasma or urine GL3[19], lyso-GL3 (AKA lyso-Gb3) [20] and the circulating vascular proliferation factor sphingosine-1-phosphate [21] are of established diagnostic or prognostic value[22].

A major challenge in the surveillance of the kidney function in Fabry patients is the correct and timely diagnosis of early decline in GFR, especially since the progression of kidney disease carries a more serious prognosis when ERT is initiated at a later stage; i.e. GFR < 60 ml/min and/or proteinuria > 1 gram/day[23]. Serum creatinine is dependant on muscle mass. It is well known that both serum creatinine and creatinine-based estimated GFR (eGFR) are inaccurate filtration markers, especially in the “creatinine-blind” window corresponding to GFR 60-90 ml/min/1.73 m² or above, i.e. in the earlier phases of decreasing renal function which may represent the “window of opportunity” in terms of stabilizing or potentially reversing the renal functional loss by adequate ERT. Moreover, it has been demonstrated that the median height and weight percentiles were below the US 50th percentile among male patients in the Fabry registry study by Hopkin et al.[2]. As a consequence, creatinine-based kidney function measurements are subject to serious bias, and indeed eGFR has been shown to overestimate true GFR by a mean of 24 ml/min/1.73 m² in a study of 8 young male adults, while no such overestimation was seen in the female patients[24]. Similar overestimation of GFR by about 50 ml/min/1.73 m² was reported in Fabry children using

the “old” Schwartz formula. However, the mean GFR overestimation was reduced to 5.3 ml/min/1.73 m² when the “new abbreviated” Schwartz formula was tested, similar to the slightly different Counahan-Barrett formula[25, 26]. Although different creatinine-based GFR formulas are used in children and adults, one should note that these formulas are validated against patient groups with moderate to severe kidney failure (CKD 3-4), and not against patients with normal function or early GFR decline. It is interesting that overestimation of GFR is a frequent pitfall in both adult and pediatric Fabry patients, and the consequence is a potential failure of timely recognition of early decline in GFR. Although West et al showed hyperfiltration in a fraction of adult Fabry patients using measured GFR[27], reliance on eGFR can lead to overestimation of this phenomenon [27]. Therefore, the concept of hyperfiltration should be used with caution in Fabry patients until validated with more exact GFR methods[26].

Cystatin C has been used for a long time in pediatric GFR measurements[28], but the accuracy of the method and assays as well as the combination with creatinine methods are still debated, and the method is not widely available[25, 29]. Measured GFR (mGFR) methods have been recommended to overcome inaccuracies in estimated GFR measurements, and Cr⁵¹-EDTA, iothalamate and iohexol methods are thought to be equally precise. Iohexol has recently been recommended as a safe and well tolerated method[30], and repeated measurements have been shown to have less variation than estimated GFR in Fabry children[26]. We recommend the use of mGFR at the very least, at some initial point as a baseline measurement before initiation of ERT, and every second year in the surveillance program in stable patients or when clinical deterioration is suspected.

Regular kidney function monitoring includes measurements of both albumin and total protein in the urine. The prevalence of albuminuria or proteinuria in children has been reported to be 10% in a report on Fabry registry data[2]. Generally, proteinuria is the cardinal sign of kidney disease and albuminuria remains the best existing biomarker for most glomerular diseases, especially in the early phases of the disease progression. Fabry disease may be no exception although the relevance of small increases in albuminuria (microalbuminuria) as a biomarker is largely based on validation studies of the earlier stages of renal damage in patients with diabetes and, to a lesser extent, hypertensive renal disease. The specific immunoassay methods for analysis of albuminuria are standardized and precise, and the analytical performance of albumin assays are superior to that of total protein assays, at least at the relevant lower concentrations (microalbuminuria) to detect what may be incipient renal damage[31]. In Fabry disease, one should keep in mind that tubular involvement is common, and subtle increases in albumin loss may either be a result of increased glomerular leakage of albumin due to damage of the filtration barrier or decreased tubular reabsorption of filtered albumin. We routinely measure albumin/creatinine ratios (ACR) and protein/creatinine ratios (PCR) in three consecutive overnight morning urine samples and use the median value as the representative value. This procedure is easily performed by the patients collecting urine samples for the preceding two days stored in a refrigerator and brought together with a fresh sample on the day of consultation. These measurements should be undertaken at the baseline evaluation, and later at regular follow-up intervals, at least on a yearly basis for GFR and perhaps more frequently for ACR and PCR. Urine concentration defects have also been demonstrated in Fabry patients[32].

Adjunctive Renoprotective Therapy

Angiotensin-converting enzyme inhibitors (ACEI) or angiotensin receptor blockers (ARB) have become standard therapies in proteinuric renal diseases, and benefits of such intervention in terms of renal and cardiovascular protection are shown in multiple studies, mostly - but not exclusively - in hypertensive patients with already established renal insufficiency[33, 34]. Tahir and co-workers have demonstrated sustained reduction of

manifest proteinuria in adult Fabry patients treated with agalsidase beta[35], and the results of a randomized trial of these agents are soon expected. The current experience with anti-proteinuric therapy in Fabry disease is confined to adult patients. Feriozzi et al. in a study of Fabry Outcome Survey patients found no significant change in proteinuria after 3 years of ERT with or without ACE or ARB use[36]. However, the number of patients studied was small. The use of ACE/ARB in Fabry patients with advanced nephropathy is challenging since they usually have low blood pressure. Currently, based mostly on uncontrolled observations, the recommended overall goal of ACEI/ARB is to lower urinary protein excretion to < 500 mg/day and stabilize the decline of kidney function to about 1 ml/min/1.73m² per year[37]. Urine albumin and protein reduction by these drugs is documented also in diabetic patients[38].

The precise diagnosis and follow-up of co-existent renal and cardiac disease is one of the major challenges in Fabry disease, and renal and cardiac progression may occur asynchronously in spite of ERT. Recent Fabry Registry data demonstrate that the primary cause of death in both male and female patients was cardiac disease[39]. In a Dutch study, about 30 % of adult patients were found to have concomitant renal and cardiac disease[19]. Ries and co-workers demonstrated that cardiac abnormalities were preceded by proteinuria and about 50 % of 25 male children had increased left ventricular mass index or abnormal electrocardiogram at the age of 15. Furthermore, because the exact time of onset of these cardiac abnormalities usually is unknown, the event-free time may be overestimated[14].

Renal Biopsy in Children with Fabry Disease

The role of the renal biopsy in the management of children with Fabry disease is currently evolving and, thus, not yet well established. It is, therefore, useful to consider this issue in the broader context of renal biopsy indications in pediatric nephrology. Biopsies are generally performed for diagnosis, prognosis and/or for making treatment decisions[40]. In children with known Fabry disease, diagnostic renal issues are uncommon and generally confined to patients with unusual clinical features such as early onset of heavy proteinuria or gross hematuria, as discussed elsewhere in this review. Renal biopsies for prognosis may have different relevance to Fabry boys vs. girls. Based on Fabry registry data, approximately 20% of males by age 50 and 50% of males by age 70 will have advanced renal disease[41]. While the risk for females by age 50 was approximately 5%, the median age at ESRD of about 38 years, was similar for males and females [41]. What is clear from the natural history of Fabry renal disease is that much of the progressive renal structural damage in Fabry disease occurs in clinical silence (see section on Biopsy Findings and Structural functional Relationships). Moreover, although overt proteinuria in males is a predictor of relatively poor treatment response to ERT[42], and a harbinger of progression to ESRD[43], the predictive value of lesser quantities of urinary protein (e.g., microalbuminuria) in Fabry disease, is unknown. Suffice it to say, a higher fraction of women that expected to progress to ESRD appear to have elevated urinary protein levels[41], however, elevated urinary protein levels are far less predictive of subsequent GFR loss in females vs. males with Fabry disease[43]. Moreover, although ERT dosage tends to be uniform, the beneficial effects may be quite variable and using functional parameters for dose adjustment may be too little-too late for preventing advanced renal injury[42]. It should also be pointed out that renal biopsies are used for diagnosis and therapy guidance for diseases such as IgA nephropathy and lupus nephritis [44] where the risk of ESRD is less than for Fabry disease. Finally, the availability of newer biopsy needles and real-time ultrasound guidance have substantially improved the safety of kidney biopsies in children[40, 44]. These points argue for an expanded role for the renal biopsy in Fabry children. In male children, the decision to initiate treatment is generally made on the basis of the good tolerability of ERT therapy, the benefits on symptoms and quality of life, and the high risk of serious end-organ damage

later in life[2], but there are no reports in which renal biopsies have been used to make these decisions. However, baseline biopsies prior to initiation of ERT in Fabry boys could better define the severity of the underlying lesions. Moreover, follow-up biopsies 2-5 years later could help to determine the adequacy of therapy and, perhaps the need for individualization of ERT dosing[23]. Renal biopsy may be particularly helpful in young female Fabry patients where the risk of serious end organ injury and the severity of symptomatology may be even less well correlated than in males[41]. For example, in young women with Fabry disease, quality of life scores may be largely in the normal range and may not be reflective of underlying tissue injury or the deterioration in quality of life yet to come[41]. Thus, the renal biopsy may be particularly helpful in females where questions as to earlier ERT initiation may be more open. In fact, severity of the disease is much more variable in females even if carrying exactly same mutation, perhaps at least partly due to random X-inactivation phenomenon. Renal biopsy by providing information about the extent of mosaicism (unpublished data) could potentially assist decisions of ERT initiation or dosage in females. Again, a pre-ERT biopsy in young females with Fabry disease could serve as a baseline which, together with follow-up biopsies, could allow assessment of the effectiveness of ERT dosing at a time in Fabry disease natural history where renal function tests are not yet informative. In fact, Fabry females at risk of serious renal disease may be even more amenable to treatment with improvement in reduced eGFR and reduction in proteinuria over 4 years of ERT treatment not seen in ERT studies of primarily male patient populations[45, 46]. To put Fabry renal disease in females in some perspective, the overall risk of serious kidney disease is similar to lupus in females. Thus, although much lower than Fabry males, it remains, by any standard, unacceptably high.

Biopsy Findings and Structural-Functional relationships

GL-3 accumulation in Fabry disease occurs in lysosomes and other cellular compartments[47]. Excessive lysosomal GL-3 manifests as cytoplasmic inclusions in concentric lamellar (myelin figures) or periodic band (zebra bodies) configuration, which are easily recognizable on toluidine blue stained sections or using electron microscopy (Figure 1). In the kidney of Fabry patients, these GL-3 inclusions can be found in virtually all cell types, among which podocytes are most prominently involved, followed by distal tubular and parietal epithelial cells.

Similar to other slowly progressive renal diseases, understanding evolution of advanced lesions in Fabry nephropathy requires studying earlier findings, here, in biopsies from children. However, such studies focusing on pathologic findings of the disease in children are few[17, 18, 48]. Gubler et al showed abundant GL-3 inclusions in all glomerular cells and in vessels in 3 children 8 to 12 years old without proteinuria[48]. These authors observed more heterogeneous distribution of inclusions in females, perhaps because of random X-inactivation. Tøndel et al described biopsy findings in 9 children with Fabry disease (age 7-18 years)[17]. All biopsies showed large numbers of GL-3 inclusions in podocytes and distal tubules. Importantly, despite normal GFR values in all patients and slightly elevated urine albumin or protein/creatinine ratios in 2/3 of patients, all biopsies had abnormal findings, including glomerulosclerosis, interstitial fibrosis, arteriopathy, and glomerular hyaline and early segmental sclerosis. On electron microscopy, all biopsies showed segmental widening of foot processes, and podocyte and distal tubular cells showed abundant GL3 inclusions. Mesangial and endothelial GL3 inclusions were present in all cases, but in less numbers, excepting the two boys who were treated with ERT. Similarly, Fogo et al, in a study primarily including adults with average age of 39 [range 16-70] years showed that even in Fabry patients with stage I CKD and minimal proteinuria, 63% of patients had glomerular sclerotic lesion, and about 13% had sclerosis in >20% of glomeruli[49]. Despite prevalent glomerular lesions, interstitial fibrosis was minimal in

CKD I and II patients. In a cohort of 14 Fabry patients younger than age 20 with normal GFR and only two patients with proteinuria above 200 mg/g creatinine, male Fabry patients had increased foot process width compared to control subjects, consistent with podocyte injury[18]. These studies indicate that current clinical modalities for evaluation of early Fabry nephropathy are insensitive, and could potentially delay initiation of ERT[17].

Current knowledge about the relationships between biopsy pathologic findings and renal function is limited. Recently, we showed that in young Fabry patients GL-3 accumulation in podocytes was progressive with age, and correlates with urine protein creatinine ratio (UPCR) and foot process widening (Figure 3), a manifestation of podocyte injury[18]; Mesangial GL-3 accumulation correlated with UPCR, but not with age; and there was no relationship between endothelial GL-3 inclusions and age or UPCR. These results are in contrast with previous studies that did not show a relationship between glomerular cell GL-3 inclusions and renal function or age[17, 48], most likely because we used quantitative morphometric methods rather than a subjective scoring system. Fogo et al showed in adults that more advanced, and non-specific chronic lesions, such as arterial and glomerular scleroses were more severe in Fabry patients with advanced CKD[49]. There was no gender difference in histologic findings of these studies except for arterial hyalinosis which was more severe for females who were, on average, older than males. Germain et al in a study of 58 patients (mean age 31; range 17-62 years) showed that sclerosis in >50% of glomeruli at baseline was associated with higher rate of GFR decline[23]. It is our view that scoring systems [49] and, even more so, morphometric measuring tools for Fabry patient kidney biopsies[18], will expand the usefulness of renal biopsies for the assessment of prognoses and treatment responses in Fabry kidney disease.

Mechanisms of Tissue Injury

Three mechanisms of tissue injury have been proposed for Fabry nephropathy[48, 50]: vascular/ischemic, podocyte, and tubulointerstitial injury. Gubler et al suggested that ischemic lesions are more common in Fabry patients 25 years or older, while these lesions were absent or mild in younger patients. The authors attributed these ischemic lesions to arterial pathologic findings[48], and suggested that arterial fibrinoid deposits, possibly due to smooth muscle cell necrosis secondary to GL3 overload, is the earliest vascular change of Fabry nephropathy. Of note, although none of the biopsies in Tøndel's study had ischemic changes, 4/9 demonstrated the so-called Fabry arteriopathy lesions (Figure 2). Although we did not find a relationship between glomerular endothelial GL-3 accumulation and UPCR or GFR, cytoplasmic fenestration was reduced in Fabry patients (Figure 1), suggestive of endothelial injury[18].

As already noted, the presence of foot process widening in young Fabry patients [18] is consistent with podocyte injury (Figure 1). This finding can be seen even before clinical proteinuria is manifest, perhaps as a result of tubular reabsorption of excess filtered protein. Progressive podocyte injury can lead to podocyte loss, and segmental glomerulosclerosis, which is a common late finding in Fabry nephropathy[48, 49]. Interstitial fibrosis and tubular atrophy are usually late findings[49], although as stated above, early interstitial fibrosis was detectable in 1/3 of pediatric cases studied by Tøndel et al.[17].

Renal biopsy can also be very useful to identify pathologies, other than Fabry nephropathy whenever the clinical picture is atypical for the disease. Concomitant lupus nephritis[51], granulomatous interstitial nephritis[52], crescentic glomerulonephritis[53], and IgA nephropathy [54] have been reported with Fabry nephropathy. Although the concurrence of IgA nephropathy and Fabry disease appears to be more frequent than random, and the two

disease may be related. It is important to recognize these cases since disease-specific interventions may improve outcomes.

Effect of ERT on renal morphologic damage

ERT treatment for up to 54 months has been demonstrated to stabilize kidney function in patients with proteinuria < 1 g/day and GFR > 60 ml/min/1.73 m², [23] and ERT, often in combination with ACEI/ARB, seems to slow the decline of GFR in patients with more advanced renal disease [55]. Although ERT has been available for about 11 years, no studies have shown that progressive renal disease can be effectively prevented or improved in the long run. Notably, there is a conspicuous gap in the knowledge of how early ERT should be undertaken in children to prevent progressive organ damage, and whether there exist an early “window of opportunity” that allows a greater possibility for reversibility of structural damage during adequate ERT, nor is it known whether a beneficial outcome is dependent on an ERT dose threshold. No biomarkers have been validated as a guidance of adequate ERT. Recently, a Dutch study found that plasma concentration of lyso-GL3 increased in patients where the ERT dose was reduced as a consequence of the current enzyme shortage, however, no major clinical events were observed in the relatively short mean observation period of 1.3 years [56].

Initial short-term renal biopsy studies demonstrated virtually complete clearance of GL-3 deposits from glomerular and interstitial capillary endothelium and mesangial cells, but not from podocytes, vascular smooth muscle cells or distal tubular cells [57]. Germain and co-workers demonstrated partial clearance of podocytes in 8 adult patients treated for up to 54 months with agalsidase beta 1.0 mg/kg [23]. However, no larger biopsy studies have been performed that clarify whether complete or partial reversal of podocyte damage during ERT is dose- or time-dependent, or whether early initiation in childhood will improve renal prognosis in the long run [58]. Such a study would be highly informative and would cast light on the potential of various cellular responses to treatment as prognostic markers for the development of differential ERT strategies including individual dose-response effects. It is also noteworthy that the initial FDA approval of agalsidase beta in the USA was based on clearance of endothelial cells from GL-3 inclusions following infusion of this drug (see <http://www.fda.gov/Drugs/DevelopmentApprovalProcess/HowDrugsareDevelopedandApproved/ApprovalApplications/TherapeuticBiologicApplications/ucm128159.htm>), a criteria which still is accepted by FDA and NIH as the gold standard for new drug trials in Fabry disease (see <http://www.amicustherapeutics.com/clinicaltrials/at1001.asp>). However, absence of relationship between endothelial GL-3 accumulation and age or UPCR [18] argues against this parameter being the gold standard of treatment effectiveness in Fabry disease. On the other hand, progressive accumulation of GL-3 in podocytes with age and presence of relationships between podocyte GL-3 density, podocyte injury and UPCR in young Fabry patients (Figure 3) suggests that podocyte injury may be a potential biomarker candidate of Fabry nephropathy progression.

Conclusion

Renal complications of Fabry disease deserve much more attention, especially in younger patients. While challenges of Fabry disease diagnosis in children contribute into late diagnosis, and therefore, late treatment, even in those with established diagnosis, the lack of sensitive non-invasive biomarkers of early Fabry nephropathy, and the lack of widely available evidence based guideline for when to initiate ERT often leads to delayed treatment. Severity of kidney injury is highly variable in Fabry disease, even among those with exact same mutation. Discovery of early non-invasive biomarkers that can predict disease progression and treatment response would be extremely useful to develop such

guidelines. However, in the mean time, cross-sectional and longitudinal renal biopsy and biomarker studies may be required to meet these goals.

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References

1. Tsutsumi O, Sato M, Sato K, Mizuno M, Sakamoto S. Early prenatal diagnosis of inborn error of metabolism: a case report of a fetus affected with Fabry's disease. *Asia Oceania J Obstet Gynaecol*. 1985; 11:39–45. [PubMed: 2990395]
2. Hopkin RJ, Bissler J, Banikazemi M, Clarke L, Eng CM, Germain DP, Lemay R, Tylki-Szymanska A, Wilcox WR. Characterization of Fabry disease in 352 pediatric patients in the Fabry Registry. *Pediatr Res*. 2008; 64:550–555. [PubMed: 18596579]
3. Spada M, Pagliardini S, Yasuda M, Tükel T, Thiagarajan G, Sakuraba H, Ponzzone A, Desnick RJ. High incidence of later-onset fabry disease revealed by newborn screening. *Am J Hum Genet*. 2006; 79:31–40. [PubMed: 16773563]
4. Schiffmann R, Warnock DG, Banikazemi M, Bultas J, Linthorst GE, Packman S, Sorensen SA, Wilcox WR, Desnick RJ. Fabry disease: progression of nephropathy, and prevalence of cardiac and cerebrovascular events before enzyme replacement therapy. *Nephrol Dial Transplant*. 2009; 24:2102–2111. [PubMed: 19218538]
5. Meroni M, Spisni C, Tazzari S, Di Vito R, Stingone A, Bovan I, Torri Tarelli L, Sessa A. Isolated glomerular proteinuria as the only clinical manifestation of Fabry's disease in an adult male. *Nephrol Dial Transplant*. 1997; 12:221–223. [PubMed: 9027807]
6. Sheth KJ, Roth DA, Adams MB. Early renal failure in Fabry's disease. *Am J Kidney Dis*. 1983; 2:651–654. [PubMed: 6405613]
7. Branton MH, Schiffmann R, Sabnis SG, Murray GJ, Quirk JM, Altarescu G, Goldfarb L, Brady RO, Balow JE, Austin HA Iii, Kopp JB. Natural history of Fabry renal disease: influence of alpha-galactosidase A activity and genetic mutations on clinical course. *Medicine (Baltimore)*. 2002; 81:122–138. [PubMed: 11889412]
8. Ortiz A, Cianciaruso B, Cizmarik M, Germain DP, Mignani R, Oliveira JP, Villalobos J, Vujkovic B, Waldek S, Wanner C, Warnock DG. End-stage renal disease in patients with Fabry disease: natural history data from the Fabry Registry. *Nephrol Dial Transplant*. 2010; 25:769–775. [PubMed: 19846394]
9. Kampmann C, Wiethoff CM, Whybra C, Baehner FA, Mengel E, Beck M. Cardiac manifestations of Anderson-Fabry disease in children and adolescents. *Acta Paediatr*. 2008; 97:463–469. [PubMed: 18363956]
10. Zarate YA, Hopkin RJ. Fabry's disease. *Lancet*. 2008; 372:1427–1435. [PubMed: 18940466]
11. Cybulla M, Schaefer E, Wendt S, Ling H, Krober SM, Hovelborn U, Schandlmaier S, Rohrbach R, Neumann HP. Chronic renal failure and proteinuria in adulthood: Fabry disease predominantly affecting the kidneys. *Am J Kidney Dis*. 2005; 45:e82–89. [PubMed: 15861341]
12. Lidove O, West ML, Pintos-Morell G, Reisin R, Nicholls K, Figueroa LE, Parini R, Carvalho LR, Kampmann C, Pastores GM, Mehta A. Effects of enzyme replacement therapy in Fabry disease--a comprehensive review of the medical literature. *Genet Med*. 2010; 12:668–679. [PubMed: 20962662]
13. Fabry Registry Minimum Recommended Schedule of Assessment. 2010. [http://www.fabrazyme.com/global/freg/schedule of assessments.pdf](http://www.fabrazyme.com/global/freg/schedule%20of%20assessments.pdf)
14. Ries M, Clarke JT, Whybra C, Timmons M, Robinson C, Schlaggar BL, Pastores G, Lien YH, Kampmann C, Brady RO, Beck M, Schiffmann R. Enzyme-replacement therapy with agalsidase alfa in children with Fabry disease. *Pediatrics*. 2006; 118:924–932. [PubMed: 16950982]
15. Ramaswami U, Parini R, Pintos-Morell G, Kalkum G, Kampmann C, Beck M. Fabry disease in children and response to enzyme replacement therapy: results from the Fabry Outcome Survey. *Clin Genet*. 2011

16. Wraith JE, Tylki-Szymanska A, Guffon N, Lien YH, Tsimaratos M, Vellodi A, Germain DP. Safety and efficacy of enzyme replacement therapy with agalsidase beta: an international, open-label study in pediatric patients with Fabry disease. *J Pediatr*. 2008; 152:563–570. 570, e561. [PubMed: 18346516]
17. Tondel C, Bostad L, Hirth A, Svarstad E. Renal biopsy findings in children and adolescents with Fabry disease and minimal albuminuria. *Am J Kidney Dis*. 2008; 51:767–776. [PubMed: 18436087]
18. Najafian B, Svarstad E, Bostad L, Gubler MC, Tondel C, Whitley C, Mauer M. Progressive podocyte injury and globotriaosylceramide (GL-3) accumulation in young patients with Fabry disease. *Kidney Int*. 2011; 79:663–670. [PubMed: 21160462]
19. Vedder AC, Linthorst GE, van Breemen MJ, Groener JE, Bemelman FJ, Strijland A, Mannens MM, Aerts JM, Hollak CE. The Dutch Fabry cohort: diversity of clinical manifestations and Gb3 levels. *J Inher Metab Dis*. 2007; 30:68–78. [PubMed: 17206462]
20. Aerts JM, Groener JE, Kuiper S, Donker-Koopman WE, Strijland A, Ottenhoff R, van Roomen C, Mirzaian M, Wijburg FA, Linthorst GE, Vedder AC, Rombach SM, Cox-Brinkman J, Somerharju P, Boot RG, Hollak CE, Brady RO, Poorthuis BJ. Elevated globotriaosylsphingosine is a hallmark of Fabry disease. *Proc Natl Acad Sci U S A*. 2008; 105:2812–2817. [PubMed: 18287059]
21. Brakch N, Dormond O, Bekri S, Golshayan D, Corvejon M, Mazzolai L, Steinmann B, Barbey F. Evidence for a role of sphingosine-1 phosphate in cardiovascular remodelling in Fabry disease. *Eur Heart J*. 2010; 31:67–76. [PubMed: 19773225]
22. Schiffmann R, Waldek S, Benigni A, Auray-Blais C. Biomarkers of Fabry disease nephropathy. *Clin J Am Soc Nephrol*. 2010; 5:360–364. [PubMed: 19965549]
23. Germain DP, Waldek S, Banikazemi M, Bushinsky DA, Charrow J, Desnick RJ, Lee P, Loew T, Vedder AC, Abichandani R, Wilcox WR, Guffon N. Sustained, long-term renal stabilization after 54 months of agalsidase beta therapy in patients with Fabry disease. *J Am Soc Nephrol*. 2007; 18:1547–1557. [PubMed: 17409312]
24. Aakre KM, Tondel C, Brun A, Svarstad E. The MDRD equation may mask decline of glomerular filtration rate in Fabry patients with normal or nearly normal kidney function. *Clin Nephrol*. 2009; 71:118–124. [PubMed: 19203503]
25. Schwartz GJ, Work DF. Measurement and estimation of GFR in children and adolescents. *Clin J Am Soc Nephrol*. 2009; 4:1832–1843. [PubMed: 19820136]
26. Tondel C, Ramaswami U, Aakre KM, Wijburg F, Bouwman M, Svarstad E. Monitoring renal function in children with Fabry disease: comparisons of measured and creatinine-based estimated glomerular filtration rate. *Nephrol Dial Transplant*. 2010; 25:1507–1513. [PubMed: 20038520]
27. West M, Nicholls K, Mehta A, Clarke JT, Steiner R, Beck M, Barshop BA, Rhead W, Mensah R, Ries M, Schiffmann R. Agalsidase alfa and kidney dysfunction in Fabry disease. *J Am Soc Nephrol*. 2009; 20:1132–1139. [PubMed: 19357250]
28. Grubb A, Nyman U, Bjork J, Lindstrom V, Rippe B, Sterner G, Christensson A. Simple cystatin C-based prediction equations for glomerular filtration rate compared with the modification of diet in renal disease prediction equation for adults and the Schwartz and the Counahan-Barratt prediction equations for children. *Clin Chem*. 2005; 51:1420–1431. [PubMed: 15961546]
29. Eriksen BO, Mathisen UD, Melsom T, Ingebretsen OC, Jenssen TG, Njolstad I, Solbu MD, Toft I. Cystatin C is not a better estimator of GFR than plasma creatinine in the general population. *Kidney Int*. 2010; 78:1305–1311. [PubMed: 20844470]
30. Schwartz GJ, Abraham AG, Furth SL, Warady BA, Munoz A. Optimizing iothexol plasma disappearance curves to measure the glomerular filtration rate in children with chronic kidney disease. *Kidney Int*. 2010; 77:65–71. [PubMed: 19847157]
31. Lamb EJ, MacKenzie F, Stevens PE. How should proteinuria be detected and measured? *Ann Clin Biochem*. 2009; 46:205–217. [PubMed: 19389884]
32. Wornell P, Dyack S, Crocker J, Yu W, Acott P. Fabry disease and nephrogenic diabetes insipidus. *Pediatr Nephrol*. 2006; 21:1185–1188. [PubMed: 16721592]
33. Jafar TH, Stark PC, Schmid CH, Landa M, Maschio G, de Jong PE, de Zeeuw D, Shahinfar S, Toto R, Levey AS. Progression of chronic kidney disease: the role of blood pressure control,

- proteinuria, and angiotensin-converting enzyme inhibition: a patient-level meta-analysis. *Ann Intern Med.* 2003; 139:244–252. [PubMed: 12965979]
34. Arnlov J, Evans JC, Meigs JB, Wang TJ, Fox CS, Levy D, Benjamin EJ, D'Agostino RB, Vasan RS. Low-grade albuminuria and incidence of cardiovascular disease events in nonhypertensive and nondiabetic individuals: the Framingham Heart Study. *Circulation.* 2005; 112:969–975. [PubMed: 16087792]
 35. Tahir H, Jackson LL, Warnock DG. Antiproteinuric therapy and fabry nephropathy: sustained reduction of proteinuria in patients receiving enzyme replacement therapy with agalsidase-beta. *J Am Soc Nephrol.* 2007; 18:2609–2617. [PubMed: 17656478]
 36. Feriozzi S, Schwarting A, Sunder-Plassmann G, West M, Cybulla M. Agalsidase alfa slows the decline in renal function in patients with Fabry disease. *Am J Nephrol.* 2009; 29:353–361. [PubMed: 18974635]
 37. Jain G, Warnock DG. Blood pressure, proteinuria and nephropathy in Fabry disease. *Nephron Clin Pract.* 2011; 118:c43–48. [PubMed: 21071972]
 38. Poulsen PL, Ebbeløj E, Nosadini R, Fioretto P, Deferrari G, Crepaldi G, Mogensen CE. Early ACE-i intervention in microalbuminuric patients with type 1 diabetes: effects on albumin excretion, 24 h ambulatory blood pressure, and renal function. *Diabetes Metab.* 2001; 27:123–128. [PubMed: 11353877]
 39. Waldek S, Patel MR, Banikazemi M, Lemay R, Lee P. Life expectancy and cause of death in males and females with Fabry disease: findings from the Fabry Registry. *Genet Med.* 2009; 11:790–796. [PubMed: 19745746]
 40. Bohlin AB, Edstrom S, Almgren B, Jaremko G, Jorulf H. Renal biopsy in children: indications, technique and efficacy in 119 consecutive cases. *Pediatr Nephrol.* 1995; 9:201–203. [PubMed: 7794720]
 41. Wilcox WR, Oliveira JP, Hopkin RJ, Ortiz A, Banikazemi M, Feldt-Rasmussen U, Sims K, Waldek S, Pastores GM, Lee P, Eng CM, Marodi L, Stanford KE, Breunig F, Wanner C, Warnock DG, Lemay RM, Germain DP. Females with Fabry disease frequently have major organ involvement: lessons from the Fabry Registry. *Mol Genet Metab.* 2008; 93:112–128. [PubMed: 18037317]
 42. Schaefer RM, Tylki-Szymanska A, Hilz MJ. Enzyme replacement therapy for Fabry disease: a systematic review of available evidence. *Drugs.* 2009; 69:2179–2205. [PubMed: 19852524]
 43. Wanner C, Oliveira JP, Ortiz A, Mauer M, Germain DP, Linthorst GE, Serra AL, Marodi L, Mignani R, Cianciaruso B, Vujkovic B, Lemay R, Beitner-Johnson D, Waldek S, Warnock DG. Prognostic indicators of renal disease progression in adults with Fabry disease: natural history data from the Fabry Registry. *Clin J Am Soc Nephrol.* 2010; 5:2220–2228. [PubMed: 20813854]
 44. Hussain F, Mallik M, Marks SD, Watson AR. Renal biopsies in children: current practice and audit of outcomes. *Nephrol Dial Transplant.* 2010; 25:485–489. [PubMed: 19729468]
 45. Schiffmann R, Ries M, Timmons M, Flaherty JT, Brady RO. Long-term therapy with agalsidase alfa for Fabry disease: safety and effects on renal function in a home infusion setting. *Nephrol Dial Transplant.* 2006; 21:345–354. [PubMed: 16204287]
 46. Banikazemi M, Bultas J, Waldek S, Wilcox WR, Whitley CB, McDonald M, Finkel R, Packman S, Bichet DG, Warnock DG, Desnick RJ. Agalsidase-beta therapy for advanced Fabry disease: a randomized trial. *Ann Intern Med.* 2007; 146:77–86. [PubMed: 17179052]
 47. Askari H, Kaneski CR, Semino-Mora C, Desai P, Ang A, Kleiner DE, Perlee LT, Quezada M, Spollen LE, Wustman BA, Schiffmann R. Cellular and tissue localization of globotriaosylceramide in Fabry disease. *Virchows Arch.* 2007; 451:823–834. [PubMed: 17674039]
 48. Gubler MC, Lenoir G, Grunfeld JP, Ulmann A, Droz D, Habib R. Early renal changes in hemizygous and heterozygous patients with Fabry's disease. *Kidney Int.* 1978; 13:223–235. [PubMed: 418264]
 49. Fogo AB, Bostad L, Svarstad E, Cook WJ, Moll S, Barbey F, Geldenhuys L, West M, Ferluga D, Vujkovic B, Howie AJ, Burns A, Reeve R, Waldek S, Noel LH, Grunfeld JP, Valbuena C, Oliveira JP, Muller J, Breunig F, Zhang X, Warnock DG. Scoring system for renal pathology in

- Fabry disease: report of the international study group of fabry nephropathy (ISGFN). *Nephrol Dial Transplant*. 2010; 25:2168–2177. [PubMed: 19833663]
50. Alroy J, Sabnis S, Kopp JB. Renal pathology in Fabry disease. *J Am Soc Nephrol*. 2002; 13(Suppl 2):S134–138. [PubMed: 12068025]
 51. Rosenmann E, Kobrin I, Cohen T. Kidney involvement in systemic lupus erythematosus and Fabry's disease. *Nephron*. 1983; 34:180–184. [PubMed: 6410291]
 52. Hiraizumi Y, Kanoh M, Shigematsu H, Yamashina M, Kondo T. A case of Fabry's disease with granulomatous interstitial nephritis. *Nippon Jinzo Gakkai Shi*. 1995; 37:655–661. [PubMed: 8583703]
 53. Singh HK, Nicleleit V, Kriegsmann J, Harris AA, Jennette JC, Mihatsch MJ. Coexistence of Fabry's disease and necrotizing and crescentic glomerulonephritis. *Clin Nephrol*. 2001; 55:73–79. [PubMed: 11200872]
 54. Kawamura O, Sakuraba H, Itoh K, Suzuki Y, Doi M, Kuwabara H, Oshima S, Abe S, Warabi H, Yoshizawa N. Subclinical Fabry's disease occurring in the context of IgA nephropathy. *Clin Nephrol*. 1997; 47:71–75. [PubMed: 9049452]
 55. Breunig F, Weidemann F, Strotmann J, Knoll A, Wanner C. Clinical benefit of enzyme replacement therapy in Fabry disease. *Kidney Int*. 2006; 69:1216–1221. [PubMed: 16609685]
 56. Smid BE, Rombach SM, Aerts JM, Kuiper S, Mirzaian M, Overkleeft HS, Poorthuis BJ, Hollak CE, Groener JE, Linthorst GE. Consequences of a global enzyme shortage of agalsidase beta in adult Dutch Fabry patients. *Orphanet J Rare Dis*. 2011; 6:69. [PubMed: 22041095]
 57. Eng CM, Banikazemi M, Gordon RE, Goldman M, Phelps R, Kim L, Gass A, Winston J, Dikman S, Fallon JT, Brodie S, Stacy CB, Mehta D, Parsons R, Norton K, O'Callaghan M, Desnick RJ. A phase 1/2 clinical trial of enzyme replacement in fabry disease: pharmacokinetic, substrate clearance, and safety studies. *Am J Hum Genet*. 2001; 68:711–722. [PubMed: 11179018]
 58. Tondel C, Bostad L, Svarstad E. 5 years follow-up renal biopsies in paediatric and adult Fabry patients on enzyme replacement therapy. *NDT Plus*. 2010; 3:iii25.

Figure 1.

Electron Micrograph of a glomerulus from an 11-years-old boy with Fabry disease. The patient had normal GFR and a urine protein/creatinine ratio of about 40 mg/g at biopsy. There are abundant GL-3 inclusions (long black arrows) in podocytes (PC). Similar inclusions are found in endothelial cells (white arrow) and mesangial cells (spiral arrow). Short black arrows show widened foot processes over a capillary (Cap) loop. Arrowheads show loss of fenestration in an endothelial cell. Magnification: 35,000 x.

Figure 2.

A. Mild Fabry arteriopathy characterized by focal replacement of smooth muscle cells in the vascular wall by hyaline-like material (arrow) in a kidney biopsy from a 12-years-old girl with Fabry disease, PAS stain, 40x. The patient had normal GFR and a urine protein/creatinine ratio of 90 mg/g at biopsy. **B.** More severe Fabry arteriopathy from an autopsy case. Arrows show that a substantial portion of the muscular wall is replaced by the hyaline-like material, H&E, 20x.

Figure 3.

Relationships between urine protein creatinine ratio (UPCR) and age with foot process width (FPW) and podocyte GL-3 inclusion density [Vv(Inc/PC)] in young Fabry patients. Both Vv(Inc/PC) and FPW increased with age and correlated with UPCR, although these relationships were more prominent in male patients (black circles) compared to females (white circles). (adapted and modified from Najafian et al., Ref. 18).