

Title: The Utility of the FIPI score in predicting long-term clinical outcomes in patients with Fabry disease receiving enzyme replacement therapy with agalsidase alfa.

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Abstract

Fabry disease is a rare X-linked lysosomal storage disorder in which there is deficiency of alpha galactosidase A. Enzyme replacement therapy (ERT) is commercially available and has been demonstrated to improve cardiac and renal outcomes. Predictive scores, such as the Fabry International Prognostic Index (FIPI), have been developed to stratify disease severity; however, these have not been validated to predict outcomes in patients receiving ERT. We show that the FIPI score at baseline can predict outcomes in a group of patients on long-term ERT.

Keywords: Fabry disease; alpha galactosidase A; Enzyme replacement therapy; Agalsidase alfa; Long-term effectiveness;

1. Introduction

Fabry disease is a rare X-linked lysosomal storage disorder in which deficiency of the enzyme alpha galactosidase A results in cellular accumulation of globotriaosylceramide. The manifestations of the disease are multiple and diverse, and include chronic pain, gastrointestinal symptoms, a characteristic rash (angiokeratoma), cardiomyopathy and cardiac conduction defects, renal failure, stroke and transient ischaemic attacks (TIAs). Replacement of alpha galactosidase A by intravenous infusion gained marketing approval in the EU in 2001, and is currently commercially available in two formulations: agalsidase alpha (Replagal, Shire Pharmaceuticals) and agalsidase beta (Fabrazyme, Genzyme Corp.)

The clinical manifestations of the condition are heterogeneous and not accurately predicted by genotype alone. Clinical scores have been developed to quantify disease severity e.g. Mainz Severity Score Index (MSSI) (Whybra et al., 2004) and attempts have been made to develop predictive scores to stratify disease severity, e.g. the Fabry International Prognostic Index (FIPI) (Hughes et al., 2012). The FIPI was developed using clinical and pathological data from 1483 patients in the Fabry Outcome Survey (FOS), an international registry of Fabry disease patients. We used a univariate analysis to identify and select candidate prognostic factors which correlated with clinically-relevant outcomes; these were further refined using a multivariate analysis and assigned a weighted score corresponding with their level of statistical significance (Hughes et al., 2012). The overall score utilizes information from clinical history, examination, and routine investigations to predict disease-related outcomes in five domains: serious cardiac events, renal events, neurological events, and all-cause mortality, and a 'composite' endpoint representing any of the above. To date, the use of the FIPI and associated organ system scores in predicting response to ERT has not been explored.

This study analyses the 15 patients that participated in an initial clinical trial of agalsidase alpha (Replagal, Shire Pharmaceuticals) (Hughes et al., 2008). Participants underwent double-blind randomized allocation to either agalsidase alpha (0.2mg/kg by intravenous infusion every two weeks) or placebo for 6 months. Subsequently, the patients were then enrolled in an open-label follow up during which they received agalsidase alpha (0.2mg/kg every two weeks) for a further two years. This cohort of patients has been receiving enzyme replacement therapy and continually followed up for over 16 years, longer than any published to date.

2. Materials and Methods

2.1 Patients

All 15 patients were male hemizygotes with a confirmed diagnosis of Fabry disease by both genotyping and leukocyte alpha galactosidase A activity. Detailed demographics have already been published (Hughes et al., 2008). In terms of mutations, 7 patients had a missense mutation in GLA, 5 patients had a nonsense mutation in GLA, and the remaining 3 patients had deletion mutations resulting in frameshifts. This study reviewed patients involved in the original clinical trial and was not funded by a commercial sponsor. All patients received the best current standard of care funded through the NHS.

2.2 Clinical outcomes

A retrospective case note review was conducted in March 2014 for all 15 patients who participated in the original trial (Hughes et al., 2008). Significant outcomes were categorized into cardiac, renal, neurological, and all-cause mortality. Cardiac endpoints were defined as myocardial infarction, cardiac surgery, major valvular disease, pacemaker or ICD insertion, or reaching New York Heart Association grade III or IV status. Renal endpoints were defined as end-stage renal failure requiring dialysis or renal transplant. Neurological endpoints were defined as stroke, TIA, or prolonged reversible ischaemic neurological event (PRIND). A 'composite' endpoint was defined as any cardiac, renal, or neurological event as previously defined, or mortality.

2.3 Neutralising antibodies

During the double-blinded, placebo-controlled phase of the initial trial, participants were tested for serum anti-Replagal antibodies at baseline, 9 weeks post initiation, 17 weeks post initiation and 24 weeks post initiation. This was continued during the two-year open label extension, with measurements taken at 13, 27, 41, 55, 81 and 107 weeks post initiation of ERT. Specimens were assessed for in-vitro neutralising activity and defined as containing a positive neutralising antibody where percentage of neutralizing activity was greater than 50%. For the purposes of this paper, patients who developed a positive neutralising antibody at any time during these measurement periods were defined as having a positive antibody status.

2.4 Urinary and plasma Gb3

Urinary and plasma Gb3 levels were calculated at baseline (pre-treatment) as described in Hughes et al., 2008.

2.5 Risk stratification

Participants were risk-scored using two scoring systems, the Mainz Severity Score Index (MSSI) and the Fabry International Prognostic Index (FIPI). This was retrospectively calculated for the point of initiation of enzyme replacement therapy using case records from the initial trial.

The MSSI uses information from history, examination, serum creatinine, electrocardiography, echocardiography, urinalysis, and magnetic resonance and computed tomography scans of the brain to stratify patients into three groups of overall disease severity: mild, moderate, and severe (Whybra et al., 2004).

The FIPI uses history, examination, electrocardiography, serum creatinine, urinalysis, pure tone audiometry and echocardiography to calculate risk scores in four domains: cardiac, renal, neurological and a composite score based on outcomes in any of those categories, plus all-cause mortality (Hughes et al. 2012).

Cardiac, renal, and neurological scores may be stratified into three groups according into risk of developing organ-related serious adverse outcomes depending on the calculated score: low (0-2), medium (3-4), and high- (5-7). Composite or FIPI scores are stratified into

two groups according to risk of reaching any organ-specific adverse outcome or mortality: low (0-3) and high (4-7).

2.6 Statistical Analyses

Kaplan-Meier analysis was performed to assess the ability of the MSS1 score, the FIPI scoring system, patient genotypes, antibody status, and urinary and plasma GL3 concentration to predict between-group differences in time to cardiac, renal, neurological, or composite endpoints.

All data were analysed with an intention-to-treat principle. Statistical analysis and graphing were performed using IBM SPSS version 20 for Windows, IBM Corp., Armonk, New York, USA, and GraphPad Prism version 6.00 for Windows, GraphPad Software, La Jolla California USA.

3. Results

3.1 Patient treatment history on ERT

In general, ERT was well-tolerated by this group of patients, and there were no recorded adverse effects on clinical note review. Eleven of the fifteen patients received uninterrupted ERT during the follow-up period. One patient entered a Phase II trial of investigational therapy seven years after initiating ERT and up to the end of the follow-up period. Another patient entered a trial twelve years after initiating ERT, and returned to ERT monotherapy for the rest of the follow-up period. After two years of agalsidase alfa therapy, one patient switched from **agalsidase alfa** to agalsidase beta due to deteriorating renal function on the former; four years after initiation of ERT, the same patient experienced a four-month interruption in ERT due to issues with funding. This was subsequently resolved, and the patient received uninterrupted therapy with agalsidase beta for the rest of the follow-up period. Another patient experienced brief intermittent interruptions in enzyme replacement therapy due to non-compliance.

3.2 Risk stratification

Using the FIPI composite scoring system at baseline prior to ERT initiation, 6/15 patients were classified as low-risk overall and 9/15 patients as high risk overall. At the time of risk score calculation, mean age in the low-risk group was 31.3 (range 23-43) and mean age in the high-risk group was 41.9 (range 34-51). This difference was statistically significant using an unpaired T test ($p=0.014$). Comparison of composite FIPI scores and mutation type is shown in Table 1.

3.3 Urinary and plasma Gb3 concentrations

The median urinary sediment Gb3 concentration at baseline (prior to ERT therapy) was 1725nmol/24hr (range 35 to 4559nmol/24hr). The median plasma Gb3 concentration at baseline was 14.14nmol/ml (range 3.36 to 18.35nmol/ml).

3.4 Significant outcomes

Patient outcomes together with demographic and clinical information are summarized in Table 1. Three deaths occurred in the patient group, which were at 1.0, 10.3 and 13.4 years post-initiation of ERT and were attributable to perioperative complications of renal transplant surgery, a fatal ischaemic stroke, and a fatal myocardial infarction, respectively. Of the surviving patients, the mean length of follow up was 12.9 years from initiation of ERT (range: 8.6-14.3 years) and the current median patient age at follow-up was 52 years (range: 37-65 years).

Seven patients experienced a serious disease-related cardiac event, which included two myocardial infarctions (one of which was fatal), two cases of severe mitral regurgitation, one insertion of a permanent cardiac pacemaker, one insertion of an implantable cardiac defibrillator, and one cardiac bypass operation for ischaemic heart disease. Within the FIPI cardiac subgroups, 1/2 of the "low-risk" FIPI cardiac patients experienced a cardiac event, 2/5 of the "medium-risk" FIPI patients did so, and 4/7 of the "high-risk" FIPI patients experienced a cardiac event.

Four patients developed a serious disease-related renal event: in all cases this was the development of end-stage renal failure. 3/4 patients then received a renal allograft. All of the affected patients scored as "high-risk" on the FIPI renal prognostic score.

Five patients developed a serious disease-related neurological event. Three patients suffered a stroke (one patient died as a result), one patient had a transient ischaemic attack (TIA), and one had a TIA followed two years later by a stroke. Within the neurological score groups, 1/7 of the "low-risk" patients experienced a neurological event, 4/6 of the "medium-risk" patients experienced a neurological event, and 0/2 of the "high-risk" group did so.

Four patients survived without experiencing any cardiac, renal or neurological serious adverse events during the 14 year follow-up period; all of these were rated as "low risk" using the FIPI composite scoring system.

3.5 Endpoint prediction

Kaplan-Meier analyses of the time to serious cardiac, renal or neurological events stratified by the corresponding organ-system predictive score are shown in Figures 1(a), 1(b), and 1(c), respectively; a breakdown of patients numbers in each risk group is shown in Table 2. Comparisons using the log-rank test showed a significant difference was observed between high and low-risk FIPI groups in time to the composite end-point ($p=0.029$ (Figure 1d)). There was no significant difference between the three risk groups for any of these scores ($p=0.78$ for cardiac score; $p=0.36$ for renal score; and $p=0.22$ for neurological score).

No significant difference in time to the composite end-point was observed between MSSl groups ($p=0.19$), different types of mutations ($p=0.90$), baseline urinary Gb3 concentrations above or below the median value ($p=0.63$), plasma Gb3 levels above or below the median value ($p=0.21$) (data not shown), or patients with or without neutralising antibodies ($p=0.99$) (data not shown).

Age of start on ERT	Mutation	MSI	Cardiac risk group	Cardiac events	Renal risk group	Renal events	Neuro. risk group	Neurological events	FIPI group	Other events
23	N215S missense	Mild	Low	-	Low	-	Mild	-	Low	-
25	717del2	Mild	Medium	-	High	-	Mild	-	Low	-
27	A143T missense	Moderate	Medium	Fatal myocardial infarction (40)	High	-	Mild	-	Low	-
30	R301X nonsense	Moderate	Medium	-	Medium	-	Mild	-	Low	Bipolar disorder (38 onwards)
40	R49L missense	Severe	Medium	⚡	Medium	⚡	Mild	⚡	Low	⚡
43	R227X (nonsense)	Moderate	Medium	⚡	Medium	⚡	Mild	Fatal stroke (53)	Low	Chronic severe depression (48), suicide attempt (49)
34	R227X nonsense	Severe	High	-	High	Dialysis-dependent renal failure (37), renal transplant (38)	Moderate	TIA (39)	High	Switched from Replagal to Fabrazyme in 2002
36	W209X nonsense	Moderate	High	Non-ST elevation myocardial infarction (49)	High	-	Severe	-	High	-
37	520delT	Moderate	High	-	High	End stage renal failure (49)	Moderate	-	High	-
38	Del 10bpexon 6	Moderate	High	-	High	End-stage renal failure (39), renal transplant	Mild	-	High	Death from complications of renal transplant surgery (39)
41	P250T missense	Moderate	Low	Severe mitral regurgitation (53)	Low	-	Moderate	Stroke (52)	High	-
43	R342Q missense	Moderate	High	Severe mitral regurgitation (47), mitral valve repair (49), infective endocarditis ⚡(57)	High	-	Moderate	Stroke ⚡(57)	High	-
47	N215S (missense)	Moderate	Medium	Implantable cardiac defibrillator for paroxysmal SVT (61)	High	Dialysis dependent renal failure (51), renal transplant (54)	Severe	-	High	-
50	R342Q missense	Severe	High	Dual chamber cardiac pacemaker (56)	High	-	Moderate	-	High	-
51	R227X nonsense	Moderate	High	Cardiac bypass surgery (58)	High	-	Moderate	TIA (61), stroke (63)	High	-

Table 1. Individual patient details on mutation, risk stratification, and clinical outcomes grouped by organ system. (* temporarily related events)

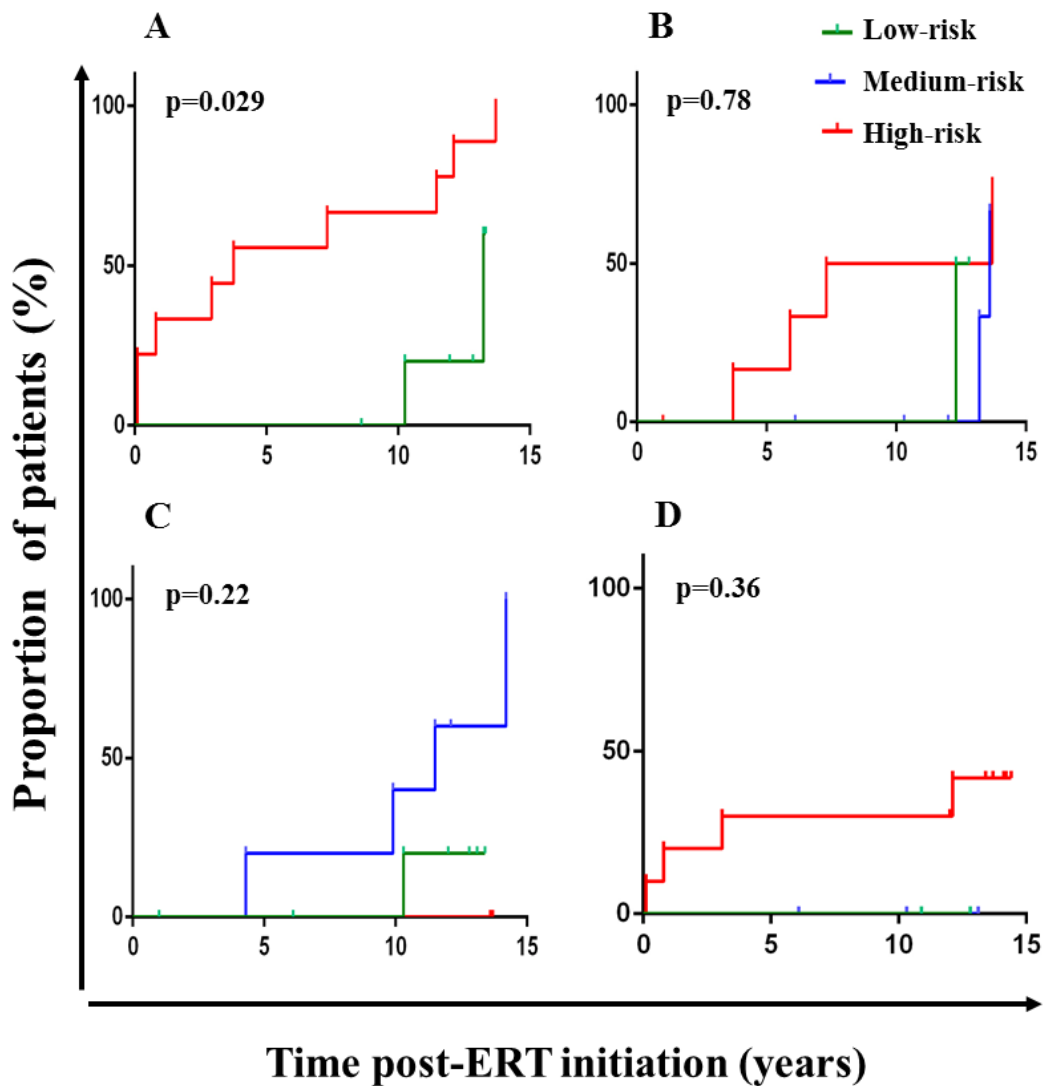


Figure 1. Kaplan-Meier analyses of time in years post-ERT initiation to defined disease-related endpoints of low, medium (expect in composite) or high-risk FIPI groups (n=15) A. Composite (FIPI) endpoints ($p=0.029$). B. Cardiovascular endpoints ($p=0.78$). C. Neurological endpoints ($p=0.22$). D. Renal endpoints ($p=0.36$). A breakdown of patient numbers in each group is given in Table 2.

FIPI risk severity	Composite FIPI (endpoint)	Cardiovascular FIPI (endpoint)	Neurological FIPI (endpoint)	Renal FIPI (endpoint)
Low	6 (2)	2 (1)	7 (1)	2 (0)
Medium	n/a	6 (2)	6 (4)	3 (0)
High	9 (9)	7 (4)	2 (0)	10 (4)

Table 2: The total number of patients in composite, cardiovascular, neurological and renal FIPI groups presented as per risk group with the number reaching a defined endpoint listed in brackets.

4. Discussion

This cohort of FD patients has received ERT for longer than any other, and, in terms of outcomes on therapy, only four patients had not reached any of our defined endpoints after up to 14 years of follow-up. Eleven patients experienced serious Fabry-related cardiac, renal, and/or neurological events, and three patients died as a result of Fabry disease and its complications. This is a small cohort with which to compare published natural history studies. The mean cumulative survival of a cohort of male Fabry patients reported around the start of the observation period of our study was 50 years (Macdermot et al 2001). A comparative statistic cannot be calculated in our cohort as 12/15 patients are alive however current survival is 52 years (range: 37-65 years).

The FIPI score for time to the composite endpoint demonstrated a statistically significant difference between the high and low-risk groups. The high-risk group experienced more Fabry-related complications and did so earlier than patients in the lower-risk group. Therefore, calculation of this score may be beneficial for determining prognosis for patients starting ERT, and its use of readily available clinical parameters makes it easily accessible to physicians treating the disease. Although the cardiac, renal, and neurological scores showed no significant difference between groups, there was a non-significant trend towards poorer organ-specific outcomes in the higher risk cardiac and renal groups and therefore detailed clinical assessment prior to ERT is likely to be as informative as the subscores. The neurological FIPI score did not seem to be able to distinguish risk for stroke which were all in low and medium risk groups with none in the high risk group. This may reflect a cardiological component to stroke in Fabry as the neurological subscore was not derived to include any cardiac parameters.

No single clinical factor was more able to predict the overall outcome. Analyses of MSSI groups, urine and plasma Gb3 concentrations, and the presence or absence of neutralizing antibodies had no predictive value for significant disease-related clinical outcomes in this study. This is in contrast to a previous study where baseline urine Gb3 excretion significantly predicted change from baseline estimated glomerular filtration rate in patients receiving agalsidase alfa for 12 months (Schiffmann et al 2013). Analysis by genotype was also not predictive for example the two patients with the N215S mutations had similar baseline leucocyte alpha galactosidase enzyme activities but fell into different FIPI subgroups and experienced distinct outcomes.

As ERT was not available prior to the trial, many patients had advanced disease. It is possible that clinical outcomes may have been better if earlier initiation of ERT had been possible for these patients and indeed those patients progressing prior to ERT are more likely to perform poorly on ERT (Hopkin et al 2016)

The main strength of this study is the length of follow-up on ERT is greater than any previously published, and hence represents an excellent starting point to validate the FIPI score as a method of discriminating groups of patients with varying response to ERT. Limitations include the small patient cohort, which was heterogeneous in age, genotype and disease severity and that the patients may have experienced more clinical progression prior to therapy since it had not been previously available. To validate the use of the FIPI conclusively for prediction of prognosis on ERT, larger numbers of patients on long-term ERT would be required.

Conflict of interests

DJML and DGJM have no conflicts of interest to declare. ABM has received honoraria, travel support, and unrestricted grants from Actelion, BioMarin, Genzyme, and Shire. DAH has received honoraria for speaking and advisory boards, and support for travel and research from Shire, Genzyme/Sanofi, Amicus, and Protalix.

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Supplementary table

Variable	Score
Cardiac	
eGFR <60 ml/min/1.73 m ²	1.5
Hearing impairment*	1.0
LVM index ≥50 g/m ^{2.7}	1.0
Proteinuria (>300 mg/24 h)	0.5
Presence of vertigo	0.5
Presence of angiokeratomas or telangiectasias	2.0
Renal	
eGFR <60 ml/min/1.73 m ²	2.0

Variable	Score
Gender—male	1.5
Proteinuria (>300 mg/24 h)	1.5
Presence of angiokeratomas or telangiectasias	2.0
Neurological	
Hearing impairment*	2.5
eGFR <60 ml/min/1.73 m ²	2.0
Presence of vertigo	1.5
Presence of anhidrosis/hypohidrosis	1.0
Composite	
eGFR <60 ml/min/1.73 m ²	2.0
Hearing impairment*	2.0
Microalbuminuria	1.0
LVM index ≥50 g/m ²	1.0
Presence of anhidrosis/hypohidrosis	1.0
Total composite	

- ECG, electrocardiogram; eGFR, estimated glomerular filtration rate; LVM, left ventricular mass.
- * Hearing impairment is average pure tone audiometric thresholds at 0.5, 1 and 2 kHz in one or both ears > 25 dB ISO.

Supplementary table. The variables contributing to the, cardiovascular, neurological, renal, and composite FIPI scores.