Appendix G. Impact of enzyme replacement therapy dose in the treatment of patients with Fabry disease

The approved doses of agalsidase beta and agalsidase alfa are 1.0 mg/kg and 0.2 mg/kg body weight every 2 weeks (EOW), respectively.\textsuperscript{1,2} The glycosylation patterns differ with agalsidase beta having higher mannose-6-phosphate content and higher sialylation, important for lysosomal uptake and minimizing liver uptake, respectively.\textsuperscript{3,4} The difference in dose resulted in a nine-fold difference in the intracellular agalsidase activity area under the curve over 2 weeks in male Fabry patients, being higher for agalsidase beta than for agalsidase alfa (3,709 [95% CI 2,517–4,900] h [nmol/h/mg] vs 396 [299–493] h [nmol/h/mg], respectively).\textsuperscript{5} Currently, there are no adequately powered head-to-head studies that have compared the long-term effectiveness of the two enzyme preparations on clinical outcomes in well-characterized, phenotypically homogenous populations. It is unlikely that such trials with sufficiently large sample sizes that enable the demonstration of statistical significance in the rare clinical setting of classic or later-onset Fabry disease will be forthcoming.

Few clinical studies have compared agalsidase alfa and agalsidase beta. A randomized, controlled, open-label trial of 34 patients (18 males, 16 females) has compared the efficacy of agalsidase alfa and agalsidase beta, both administered at a dose of 0.2 mg/kg EOW (i.e. for agalsidase beta, one-fifth of the approved dose).\textsuperscript{6} The observed ERT effects on left ventricular mass (LVM, primary endpoint) after 24 months were similar for both treatments. Also, no differences in eGFR, pain, and the reduction of plasma and urinary GL-3 levels were found. Both treatment groups had a high rate of treatment failure (defined as progression of renal disease [33% increase in serum creatinine, need for dialysis, or transplantation], progression of cardiac disease, or occurrence of a new cerebrovascular accident), approaching 25% of patients at 24 months of treatment and, after inclusion of patients who were treated longer, approximately a third of the patients (12/34 patients). The occurrence of treatment failures was considered frequent by the authors and did not differ between the two treatment groups.\textsuperscript{6}

The Canadian Fabry Disease Initiative (CFDI) randomized 92 patients (37 males, 55 females; 1:1 randomization stratified by gender) to either agalsidase alfa (n=62) or agalsidase beta (n=30) at the approved doses.\textsuperscript{7} An interim 5-year report on the primary endpoint (composite clinical endpoint consisting of renal, cardiovascular, and cerebrovascular events, or death) observed rates of serious clinical events of 19.4% for patients receiving agalsidase alfa 0.2 mg/kg EOW and 13.3% for patients receiving agalsidase beta 1.0 mg/kg EOW. The difference (31%) was not significant (p=0.57), but the authors acknowledged the limitations in statistical power.\textsuperscript{7} Use of concomitant medications was frequently reported at baseline (acetylsalicylic acid 67.8%, ACEI and/or ARBs 55.6%, statins 38.9% of patients).\textsuperscript{7} The latest update in the public domain, an 8-year follow-up, was presented in 2016.\textsuperscript{8} At that time, 117 patients had been randomized to agalsidase alfa (69 [60%]) and agalsidase beta (46 [40%]). Baseline clinical parameters and concomitant drug use did not differ except for the Mainz Severity Score Index (MSSI), which was lower (less severe baseline disease) in patients randomized to agalsidase alfa than in those randomized to agalsidase beta (23.2±8.7 vs 27.6±10.1; p=0.02). There was no difference between different ERTs in terms of renal and cardiac outcomes and Kaplan-Meier analysis of time free of first clinical event (p=0.66). However, there were about two times more clinical events per patient on agalsidase alfa (45 events in 69 patients, 0.65 events per patient) than on agalsidase beta (15 events in 46 patients, 0.33 events per patient).\textsuperscript{8} Unfortunately, none of the interim reports provided sample-size
calculations to allow interpretation of the obtained p values. However, the current estimated enrollment of the CDFI study is 600 patients (ClinicalTrials.gov Identifier: NCT00455104) rendering the interim reports grossly underpowered to detect statistically significant differences. The study is expected to continue up to the completion of 10 years.

Indirect evidence suggests that a higher agalsidase dose may be of clinical benefit in reducing the burden of storage within podocytes. In a renal biopsy study in young patients with classic Fabry disease (n=12; median age 16.5 years, range 7–33 years), the cumulative dose of ERT (agalsidase alfa and/or beta) received over 5 years was shown to correlate with the reduction of podocyte globotriaosylceramide (GL-3) deposits. This correlation has been confirmed in a study in a larger group of patients (n=20) with a higher median age (21 years), wider age range (7–62 years), and longer treatment duration (9.4 years). In one clinical trial (21 adult males), a reduced dose of agalsidase beta (0.3 mg/kg EOW) maintained the GL-3 reduction or clearance in various renal cell types previously obtained with the full dose (1.0 mg/kg) in most, but not all, patients at 18-months follow-up.

A temporary shortage of agalsidase beta from June 2009 to February 2012 necessitated a switch of ERT formulation or a dose reduction in most Fabry patients on treatment. A range of publications have reported outcomes after changes in treatment regimens. In a study of 89 patients (37% females overall; 12.5% females in the agalsidase beta 1.0 mg/kg EOW group), patients receiving agalsidase beta 1.0 mg/kg EOW had a stable disease course, whereas those with dose reduction and those with switch to agalsidase alfa 0.2 mg/kg EOW had an impairment of renal function. In contrast to patients in the agalsidase beta 1.0 mg/kg EOW group, patients receiving lower agalsidase beta doses showed significant increases in MSSI scores after 2 years. In a study of patients who were started on (n=29; 52% females) or switched to (n=71; 44% females) agalsidase alfa 0.2 mg/kg EOW, eGFR, proteinuria, and LVMI remained stable over 24 months of treatment. Smaller studies (n=11 [4 males, 7 females] and n=10 [7 males, 3 females]), reported no deterioration of outcomes within 3 years after switching from agalsidase beta to agalsidase alfa. All of these studies were included in a systematic review and meta-analysis (176 patients) evaluating clinical parameters after patients were switched from agalsidase beta to agalsidase alfa. The use of agalsidase alfa was associated with steady levels of GFR and minor significant improvements in cardiac function. The sex of the patients was not reported and there was no information on the severity of GLA mutations. The heterogeneity of the data and the short follow-up period in patients with slowly progressive Fabry disease did not enable the authors to determine whether clinical stability and efficacy are maintained in the long term. In a small study (15 males), a switch from agalsidase alfa 0.2 mg/kg EOW to agalsidase beta 1.0 mg/kg EOW significantly reduced plasma lyso-GL-3 and GL-3 levels with mean percentages reduction from baseline of 39.5% and 17.9%, respectively. This is consistent with another short-term study (n=35; 17 males, 18 females) on the impact of switching from agalsidase beta 1.0 mg/kg EOW to agalsidase alfa 0.2 mg/kg EOW; it did not show a significant increase in Fabry disease-related clinical events. However, higher disease activity indicated by increased plasma globotriaosylphosphoglycerine (lyso-GL-3) levels was observed in nearly all patients evaluated.

A Cochrane review of clinical trials with ERT indicated that, compared with placebo, ERT significantly improves microvascular endothelial deposits of GL-3 and pain-related quality of life although the optimal dose regimen could not be determined. The long-term influence of ERT on the risk of morbidity and mortality related to Fabry disease could not be established. This Cochrane review
was marred by the paucity of long-term clinical trials that allow assessing the impact of ERT on clinical events. However, long-term experience has been accumulated through the analysis of data from the registries that have been established as a result of health-care authorities' recommendations for post-marketing evaluation of the drugs. A complementary overview of the Cochrane publication through a linear regression and a pooled analysis of proportions from cohort studies has recently been published.\(^{20}\) The authors concluded that agalsidase beta is associated with a significantly lower incidence of renal (pooled proportions for renal complications: agalsidase alfa 15.3\% [95\% CI 0.048–0.303\%]; agalsidase beta 6\% [95\% CI 0.04–0.07\%]; untreated patients 21.4\% [95\% CI 0.151–0.2835\%]) and cardiovascular (agalsidase alfa 28\% [95\% CI 0.07–0.55\%]; agalsidase beta 7\% [95\% CI 0.05–0.08\%]; untreated patients 26.2\% [95\% CI 0.149–0.394\%]) events compared with untreated patients and with a significantly lower incidence of cerebrovascular events (agalsidase alfa 11.1\% [95\% CI 0.058–0.179\%]; agalsidase beta 3.5\% [95\% CI 0.024–0.046\%]; and untreated patients 17.8\% [95\% CI 0.123–0.240\%]) compared with untreated patients and agalsidase alfa.\(^{20}\)

The above findings suggest a differential impact of therapy based on the dose of enzyme administered.

**Antibody formation**

The main risk factor for the development of anti-ERT antibodies in lysosomal storage diseases is a genetic mutation that abrogates the production of the enzyme, thus preventing the immune system from recognizing the missing enzyme as self during exposure (cross-reactive immunologic material [CRIM]-negative patients).\(^{21}\) Hence, antibodies or serum-mediated inhibition of agalsidase are uncommonly identified in female Fabry patients, who express [residual ] $\alpha$-Gal A activity and a lower titer compared with male patients.\(^{22}\) Furthermore, antibodies detected in males are cross-reactive to both enzyme preparations.\(^{13,23,24}\)

The lack of a standardized assay further complicates matters as different methods have different sensitivity, leading to different results. For example, in a study that assessed IgG antibodies to agalsidase beta (at baseline and at follow-up) or agalsidase alfa (only at baseline), the antibody titer was calculated as the reciprocal of the last dilution above the assay cutoff point.\(^{17}\) In another study investigating the presence of antibodies against agalsidase beta and agalsidase alfa, the titer was defined as the maximum dilution at which the post-exposure serum had an absorbance at least twice that of the baseline sample of that patient at the same dilution.\(^{24}\) Only studies using the same technique to assess antibodies in patients on different drug regimens provide significant insights into the matter. Unfortunately, the presence of CRIM has not been determined in studies assessing antibody formation in patients receiving different agalsidase preparations.\(^{6,13,25}\) Thus, the impact of the underlying mutation on the development of antibodies to agalsidase may depend on the presence of nonsense mutations.

A large study (n=168; 68 males) supports the concept that the severity of the mutation may be a key risk factor for antibody development in Fabry disease.\(^{13}\) In males, the main risk factor for the development of serum-mediated inhibition was having a nonsense mutation; nonsense mutations were present in 6/23 (26.1\%) patients without serum-mediated inhibition and in 13/18 (72.2\%) patients with serum-mediated inhibition (p<0.01).\(^{13}\) Inhibition did not depend on the initial ERT formulation used, in line with the reported cross-reactivity of antibodies.\(^{13}\) However, serum-mediated inhibition was associated with higher LVM, lower eGFR, and more Fabry disease symptoms.\(^{13}\)
In a randomized, controlled, open-label trial of 34 patients (18 males, 16 females) comparing agalsidase alfa and agalsidase beta both administered at a dose of 0.2 mg/kg EOW (for agalsidase beta one-fifth of the approved dose), IgG anti-agalsidase antibodies developed in 10/16 males (4/8 patients [50%] on agalsidase alfa and 6/8 patients [75%] on agalsidase beta; p=0.3).

Another study analyzed the influence of agalsidase dose on antibody formation and the levels of plasma and urinary GL-3. Thirty-one Fabry patients received either agalsidase alfa (n=18) or agalsidase beta (n=13) at 0.2 mg/kg EOW for ≥12 months (most patients also participated in the study described above). In addition, 21 patients who had received agalsidase beta at 1.0 mg/kg EOW for ≥12 months were included. IgG antibodies were detected in 18 males (4/10 [40%] agalsidase alfa 0.2 mg/kg, 6/8 [75%] agalsidase beta 0.2 mg/kg, and 8/10 [80%] agalsidase beta 1.0 mg/kg EOW) and had neutralizing activity in 17/18 (94%) males. The difference in frequency of antibody formation between the agalsidase beta 1.0 mg/kg group and agalsidase alfa 0.2 mg/kg group was reported to be statistically significant (p=0.005), but there was no difference in frequency between the two groups treated with 0.2 mg/kg EOW (p=0.157), or between the two groups treated with agalsidase beta (p=0.453). The interaction between ERT and mutation (nonsense vs missense) was not explored. Plasma GL-3 levels decreased in all treatment groups. Urinary GL-3, however, was effectively cleared in antibody-negative patients across treatment groups. In antibody-positive patients, treatment with 0.2 mg/kg resulted in an increase of urinary GL-3, while the agalsidase beta 1.0 mg/kg EOW group had significantly reduced levels. No change in renal function was observed in any of the treatment groups, but there was a more robust decrease in LVM in both antibody-negative and antibody-positive patients receiving agalsidase beta at 1.0 mg/kg EOW.

Results of plasma lyso-GL-3 assessments in a subset of patients enrolled in the above-mentioned study have been reported separately. A marked reduction in mean lyso-GL-3 levels within 3 months was found in all treatment groups. However, in antibody-positive patients, only treatment with agalsidase beta 1.0 mg/kg EOW led to a similar reduction in lyso-GL-3 as observed in antibody-negative patients.

References


