pegunigalsidase alfa, a novel PEGylated a-Galactosidase-A ERT for Fabry disease

Safety and efficacy of 1-year treatment experience

Derralynn Hughes[1], Simeon Boyd[2], Pilar Giraldo[3], Derlis Gonzales[4], Myrl Holida[5], Ozlem Goker - Alpan[6], Gustavo Maegawa[7] Mohamed Atta[8], Kathy Nicholls[9], Raphael Schiffmann[10], Ahmad Tuffaha[11], Laura Barisoni[12], Robert Colvin[13], J.Charles[14] Sari Alon[15], Jenny Krupko[15], Mali Szlaifer[15], Einat Brill-Almon[15], Raul Chertkoff[15]

Abstract

The current report is a one-year follow-up, specifically a sub-analysis of 10 Fabry disease (FD) patients with a classic phenotypic presentation of the disease, out of 18 treated with pegunigalsidase alfa (PRX-102) recruited to two clinical studies: PB-102-F01 and its extension PB-102-F02. These phase I/II dose-ranging studies (0.2mg/kg; 1 mg/kg; 2mg/kg) evaluate the safety, pharmacokinetics and efficacy parameters on patients treated with pegunigalsidase alfa administered IV every other week.

Fabry disease is an X-linked disorder caused by the loss of function of the lysosomal enzyme α-Galactosidase-A. Phenotypic differences have been described between classically and non-classically affected patients. Pegunigalsidase alfa, a novel, PEGylated, chemically modified α-galactosidase A, which resulted in a more stable homodimer enzyme, expressed in the ProCellEx® system using a plant cell line.

Results: the enzyme has enhanced pharmacokinetic properties including a half-life of approximately 80 hours, and a substantially higher AUC result. Symptomatic FD naïve male and female patients (>18 y.o.) were recruited to the study. The primary outcome consists of safety, adverse events, clinical laboratory, physical examination and ECG. The secondary outcome includes pharmacokinetics and exploratory efficacy parameters: plasma Gb3 and Lyso-Gb3, kidney functions: eGFR and proteinuria, and a BPI questionnaire to assess pain. Additional parameters were kidney Gb3 inclusion bodies (assessed by biopsies at baseline and 6 months), cardiac MRI and MSSI (Mainz Severity Score Index).

After 1 year of treatment, classic FD patients presented a mean total reduction of Gb3 inclusions in PTCs of 84.1 ± 3.3 % (BLISS score); a mean reduction in plasma Gb3 and Lyso-Gb3 of 33.3 ± 7.6% and 57.6 ± 6.8% respectively, stability in annualized eGFR and cardiac MRI, and an improvement in MSSI and BPI.

In summary: pegunigalsidase alfa was found to be available throughout the 2-week infusion intervals, and well tolerated with the majority (98%) of adverse events being mild and moderate. Only 1 of the patients experienced an event of hypersensitivity, and only 3 patients, (~ 19%) developed treatment-induced antibodies which turned to be negative for ADA after 1-year of treatment. Efficacy was demonstrated in various disease parameters including: stability in kidney function, stability in cardiac function and reduction in plasma and kidney biopsy-assessed biomarkers levels.

Study Design

Phase I/II, Open Label, Dose Ranging

General Design

Adult Fabry Patients
Three dose groups:

0.2 mg/kg | 1 mg/kg | 2 mg/kg

Intravenously, every 2 weeks

Main Inclusion Criteria:
- Symptomatic Fabry patients
- ERT naïve or patients who are off ERT in the last 6 months; negative IgG anti PRX-102 antibody
- eGFR ≥ 60 mL/min/1.73m2

Main Exclusion Criteria:
- Chronic kidney disease stages 3-5
- Severe myocardial fibrosis by MRI
- Pregnant or nursing
- Known allergies to ERT

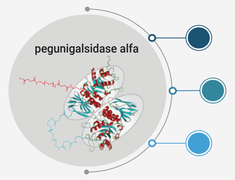
Overall study design

Demographics and baseline enzymatic activity

	0.2 mg/kg (n=6)	1 mg/kg (n=8)*	2 mg/kg (n=4)
Mean age (years) ± SD (range)	30.0 ± 10.8 (21-50)	34 ± 9.7 (17.5-52.5)	40.6 ± 9.5 (21-54)
Male : Female	4:2	6:2	1:3
Ethnicity			
Caucasian	4	4	4
African American	1	2	0
Asian	0	0	0
Other	1	0	0
Mean Enzymatic Activity	**0.2 mg/kg (males=4, females=2)**	**1 mg/kg (males*=6, females=2)**	**2 mg/kg (males=1, females=3)**
In leucocytes (range) (normal 33-134 nmol/hr/mg prt.)	Males: 3.15 (1.6-5) Females: 27.5 (15-40)	Males: 2.67 (0-7.8) Females: 69.5 (67-72)	Male 0.56 Females: 42.66 (33-53)
In plasma (range) (normal 4-21.9 nmol/hr/ml)	Males: 0.22 (0-0.4) Females: 3.15 (2-4.3)	Males: 0.28 (0.05-0.44) Female: 6.8 (5.8-7.8)	Male: 0.4 Females: 4.80 (2.52-7.8)

* one subject discontinued due to AE; one subject discontinued due to non compliance

Drug Design



- PEGylated covalently-linked homodimer composed of two subunits produced in plant cells
- Subunits linked together through the 2000Da **PEG cross-linker** resulting in 114 kDa enzyme
- Contains additional **PEG moieties** bound to only one subunit through a lysine residue

Classic Fabry disease (FD) patients:

- Symptomatic adult FD patients
- Plasma and/or leucocyte alpha galactosidase activity less than 30% mean normal levels
- And one or more of the described characteristic features of FD:
  - Characteristic Facies
  - Neuropathic pain
  - Cornea verticillata
  - Clustered angiokeratoma
  - Diaphoresis
  - Abdominal pain
  - Diarrhea/constipation

Results

Safety

Of the 18 pegunigalsidase alfa treated patients, 17 experienced a total of 223 AEs. All AEs were mild or moderate in intensity except for 4 patients who experienced 4 severe AEs (i.e. pain in extremity, renal hematoma, migraine, bronchospasm). Two of these, migraine and bronchospasm were considered by the investigator to be possibly and definitely related to treatment. Renal hematoma and the bronchospasm were also considered SAEs; bronchospasm led to patient's discontinuation from the study. Eight (8) of 18 treated patients experienced 30 adverse events during or within 2 hours of the infusion; 24 were considered by the investigator to be probably, possibly or definitely related to treatment.

Safety in 416 infusions

223 (100%)
Total AEs in 17/18 subjects

- 219 (98.2%) AEs mild and moderate
- 4 (1.8%) Severe
  - 2 Severe treatment related in 2 pts. (Migraine, Bronchospasm*)
  - 2 Severe not treatment related in 2 other pts.
- 2 (0.9%) Serious AEs
  - Bronchospasm - treatment related*
  - Kidney hemorrhage post biopsy, not treatment related**
- 54 (24.2%) Related, possibly related
- 169 (75.8%) Not related, unlikely related

*52 year old male experienced a Grade 3 serious adverse event of bronchospasm related to the study drug 40 minutes following the first infusion initiation, received a total of 115mg investigational drug. Was treated with inhalations, adrenalin and steroids, and discharged the following day. Discontinued Per Protocol. Anti PRX-102 IgG was negative and anti PRX-102 IgE was positive at baseline. **28 year old male, pre treatment renal hematoma post kidney biopsy- Not related.

8 patients experienced 30 AEs*

ID	IR AEs	Description (MeDRA Preferred Term)	Treatment-induced ADA
0.2 mg/kg			
01-F101	(+)	Headache	(-)
51-F102	(+)	Chest discomfort, Sneezing, Nausea, Hyperhidrosis	(-)
12-F103	(+)	Pruritus generalized	(-)
26-F104	(-)		(-)
17-F105	(-)		(+)
15-F106	(-)		(+)
1.0 mg/kg			
04-F107	(+)	Hypotension, Dizziness, Dyspnea, Rash maculo-papular	(-)
09-F108	(+)	Infusion related reaction, Chest pain, Pruritus, Rash, Dermatitis contact, Nausea, Dizziness	(-)
10-F111	(+)	Bronchospasm	(-)
03-F112	(-)		(-)
07-F113	(-)		(+)
12-F114	(-)		(-)
12-F115	(-)		(-)
2.0 mg/kg			
15-F116	(-)		(-)
15-F117	(+)	Abdominal pain	(-)
17-F118	(+)	Abdominal pain upper	(-)
09-F119	(-)		(-)

*During or within 2 hours of PRX-102 Infusion

Efficacy

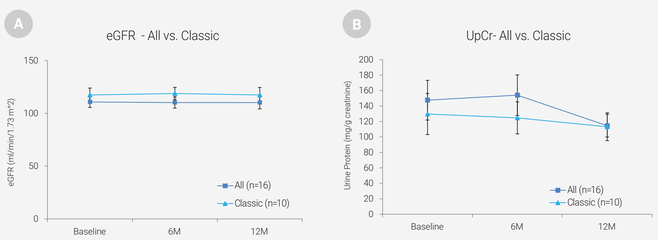
Efficacy analysis presented for:

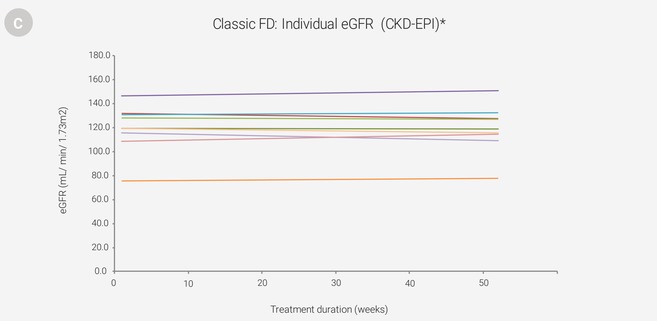
- All patients (n=16; 9M:7F)
- Classic Fabry disease (FD) patients (n=10 ; 9M:1F)

Stable kidney functions

Phase I/II clinical trial results indicate a positive impact on kidney function of Fabry patients. Fabry disease is progressive and based on the rate of accumulation of Gb3, and disease severity increases with increasing age. Increased proteinuria together with kidney failure is one of the dominant clinical manifestations of the disease.

Renal failure is the most common cause of death in male Fabry disease patients (Germain, 2010). All 16 patients participating in the study (Per protocol) exhibited a stabilization of eGFR values (A) and slight reduction in proteinuria (B) throughout a 12-month period; individual eGFR trend lines are shown for the classic Fabry patients (C).

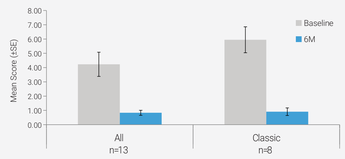




* Excluding 1 male patient treated intermittently with doxycycline throughout the year

Reduction of Gb3 in kidney peritubular capillaries

Quantitative BLISS score



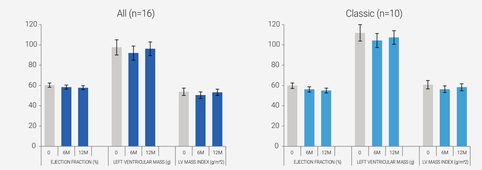
>300 peritubular capillaries (PTC) were scored for Gb3 inclusions in each biopsy. Slides underwent digital imaging before scoring. Images were distributed in a random and blinded manner for annotation by one pathologist, and subsequent scoring by two other pathologists.

	% change ±SE
All patients (n=13)	-67.8 ± 8.9
Classic patients (n=8)	-84.1 ± 3.3

The pharmacodynamics of this enzyme can be seen by the reduction in Gb3 inclusions in PTC. The mean reduction was >80% in the classic FD patients and 68% in the entire cohort.

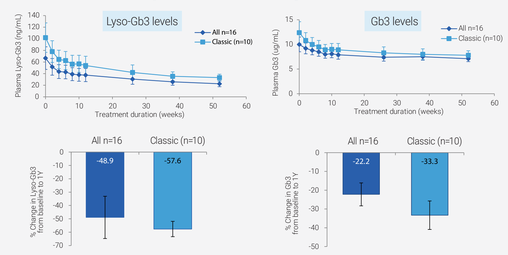
Stable cardiac parameters (by MRI)

Mean LVM, LVMI and EF



Cardiac MRI was performed at baseline, 6 and 12 months of pegunigalsidase alfa treatment. MRI read was done centrally in a blind manner. Cardiac MRI results showed that treated FD patients maintained in the normal ranges throughout the 12 months of study period. No cardiac fibrosis was observed throughout the study period.

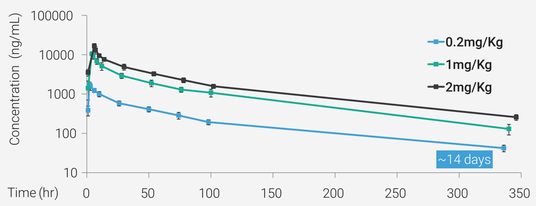
Biomarkers - plasma Gb3/Lyso-Gb3



A progressive reduction in plasma Gb3 and lyso-Gb3 was shown throughout the study, with higher reduction (% change from baseline) of both biomarkers with the classic FD patients.

Pharmacokinetics (PK)

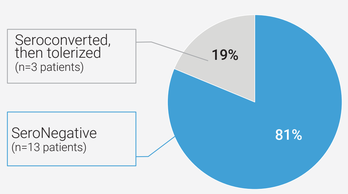
Plasma pegunigalsidase alfa concentration vs. time



Summary of PK observations:
- Pegunigalsidase alfa PK parameters and profile indicate dose dependency
- Plasma half-life of approximately 80 hrs
- Available enzymes throughout the 2-week intervals

Immunogenicity

Treatment-Induced Anti-Drug Antibodies (ADA)



Summary of Immunogenicity observations:
- **Low incidence of ADA with low titers:** 3/16 patients 19%; Max titer 4633
- **Neutralizing Antibodies:** 2/3 positive patients had neutralizing activity in some samples
- **Immune Tolerization:** All 3 ADA positive patients turned to be negative for ADA after 12M of treatment

Overall Conclusions

Pegunigalsidase alfa (PRX-102) – PEGylated covalently-linked recombinant alpha-GAL-A enzyme, stable homodimer, produced in plant cells

- **Pegunigalsidase alfa has a longer half-life and a substantially higher AUC**, available enzyme throughout 2-week infusion intervals
- **Pegunigalsidase alfa is well tolerated:** the majority of adverse events were mild and moderate in severity
- **Limited formation of antibodies,** ADA response was transient and tolerization was observed
- **Effectiveness demonstrated in various disease parameters** including: stable kidney and cardiac function, reduction of Gb3 inclusions in kidney peritubular endothelial cells, reduction of plasma Gb3 and Lyso-Gb3

Next Stage Clinical Development - Balance Study:

- Phase 3, randomized, double blind, active control study
- Evaluate the safety and efficacy of PRX-102 compared to agalsidase beta in patients with FD previously treated with agalsidase beta with rapidly declining renal function
- Classic FD patients with impaired renal function
  - Screening eGFR by CKD-EPI equation 40 to 90 mL/min/1.73 m^2
  - Linear negative slope of eGFR of ≥ 2 mL/min/1.73 m^2
- 2 years treatment duration
- Extension study will be offered to patients at the end of the study

- http://www.fabrynext.com
- ClinicalTrials.gov Identifier:NCT02795676

[1]LSDU, Institute of Immunity and Transplantation, Royal Free London NHS Foundation Trust, London, UK, [2]Department of Pediatrics, Section of Genetics, MIND Institute, UC Davis Medical Center, Sacramento, USA, [3]Hospital de Dia Quiron, Zaragoza, Spain, [4]Instituto Privado De Hematologia Investigacion Clinica, Asunción, Paraguay, [5]Medical Genetics-Lysosomal Storage Disorders, University of Iowa , Iowa City, Iowa, USA, [6]O&O Alpan LLC, Fairfax, Virginia, USA, [7]Department of Pediatrics/Genetics & Metabolism, University of Florida, Gainesville, Florida, USA, [8]Department of Medicine, division of Nephrology, Johns Hopkins University School of Medicine, Baltimore, Maryland, USA, [9]Nephrology Department, the Royal Melbourne Hospital, Melbourne, Australia, [10]Baylor Institute of Metabolic Diseases, Baylor University Medical Center, Dallas, Texas, USA, [11]Kansas University Medical Center, Kansas City, Kansas, USA, [12]University of Miami, Department of Pathology, Miami Florida, USA, [13]Massachusetts General Hospital ,Harvard Medical School, Department of Pathology Boston, Massachusetts, USA, [14]University of North Carolina, Department of Pathology, Chapel Hill, North Carolina, USA, 15Protalix Biotherapeutics, Carmiel, Israel