TITLE PAGE

Protocol Title:

Phase III, Open-label, Switch Over Trial of the Efficacy and Safety of Agalsidase Beta Biosidus (AGA BETA BS) in Fabry Disease Patients Previously Stabilized with Fabrazyme®.

Protocol Number: BIO-AGA-Fase III-001

Amendment Number: 5

Compound: Recombinant human α-galactosidase A (agalsidase beta)

Brief Title: Switch over study of biosimilar AGA for Fabry Disease (SMILE)

Study Phase: III

Sponsor Name: Biosidus S.A.U.

Legal Registered Address: Biosidus, Constitución 4234 (1254), Buenos Aires

Regulatory Agency Identifier Number(s): Not applicable

Version: April 22, 2024

Sponsor Signatory

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Manager of Clinical Investigation	

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1.0 PROTOCOL SUMMARY

1.1 Synopsis

Protocol Title:

Phase III, Open-label, Switch Over Trial of the Efficacy and Safety of Agalsidase Beta Biosidus (AGA BETA BS) in Fabry Disease Patients Previously Stabilized with Fabrazyme®.

Rationale:

Biosidus' Agalsidase Beta is being developed as a proposed biosimilar product to Fabrazyme, a recombinant human α-galactosidase A used for enzyme replacement therapy in patients with Fabry disease (FD). AGA BETA BS already have shown biosimilarity with Fabrazyme in 7 nonclinical studies involving pharmacodynamics (PD), pharmacokinetics (PK) and toxicit, and in a phase I clinical study.

The latter was a comparative study of pharmacokinetics and pharmacodynamics between AGA BETA BS and Fabrazyme® in healthy volunteers carried out between 2021 and 2022 in the Province of Buenos Aires. It was a sequential, open-label, randomized, parallel two-arm study. Each participant received a single dose of 1 mg/kg body weight of the drug assigned at randomization, at a 1:1 ratio. Participants were followed for 35 days after infusion for safety assessment. Pharmacokinetics, pharmacodynamics and safety results are detailed below: (1)From the average values obtained for pharmacokinetics parameters (**Cmax, AUC0-12h and AUC0-∞),** the biosimilarity between formulations was analyzed by calculating the ratio of Test/Reference formulations and its 90% confidence interval. All three parameters were found to be within the accepted biosimilarity range of 0.80 - 1.25. (2) The average enzyme activity at the end of infusion (5h) for the Biosidus Agalsidase product was 78.38 ± 20.89 mU/ml and for Fabrazyme® 80.81 ± 28.60 mU/ml. These results showed an enzyme activity ratio at 5h Test/Referent of 0.97. No significant differences were found between the two treatment groups (p= 0.8210). (3) Regarding the safety profile, AGA BETA BS showed a similar behavior to the reference agalsidase, both in the number of adverse events and in the category and degree of severity. There were no serious adverse events. All adverse events were classified as mild by the principal investigator. Those classified as "possible" in terms of causality relationship with the investigational drug were expected and were described in the investigator's brochure and the reference product package insert. No neutralizing anti-agalsidase antibodies were detected in any volunteer, neither in the pre-dose samples, nor at 12h post start of infusion nor at 35 days post administration.

In line with the ANMAT Provision 7729/2011, it is proposed to carry out this clinical trial to evaluate the similarity of AGA BETA BS, in terms of safety and efficacy, with the innovator product. Using the same reference product as in previous studies. This is a comparative study, under the same conditions as the authorized use of the innovator product (IV infusion every two weeks, at a dose of 1 mg/kg of body mass), in the indication approved for the innovator product (Fabry disease) adapted to the recommendations accepted in other regulatory agencies for low prevalence diseases.

Objectives and Endpoints:

Objectives	Endpoints
Primary	
• The primary objective of the study is to evaluate the equivalence in efficacy between AGA BETA BS and Fabrazyme after 6 months of treatment in participants with Fabry disease previously stabilized with Fabrazyme, by measuring disease biomarker.	• Mean serum Lyso-Gb3 marker ratio after 26 weeks of treatment, defined as plasma level of the marker Lyso-Gb3 after 26 weeks (6 months) divided by plasma level of the marker Lyso-Gb3 at baseline.
Secondary	
• To evaluate the difference in efficacy between AGA BETA BS and Fabrazyme after 1 year of treatment in participants with Fabry disease previously stabilized with Fabrazyme, by measuring disease biomarker.	• Mean serum Lyso-Gb3 marker ratio after 54 weeks of treatment, defined as plasma level of the marker Lyso-Gb3 after 54 weeks (12 months) divided by plasma level of the marker Lyso-Gb3 at baseline.
• To compare the pain severity before and after the treatment with AGA BETA BS in participants with Fabry disease previously stabilized with Fabrazyme, as measured by the BPI-short form.	• Change from baseline in pain severity as assessed by BPI-short form pain severity items scores after 26 and 54 weeks of treatment.
• To compare the impact of pain on daily functions before and after the treatment with AGA BETA BS in participants with Fabry disease previously stabilized with Fabrazyme, as measured by the BPI-short form.	• Change from baseline in pain interference as assessed by BPI-short form pain interference items scores after 26 and 54 weeks of treatment.
• To compare the participants' perception of their own health before and after the treatment with AGA BETA BS in participants with Fabry disease previously stabilized with Fabrazyme, as measured by the SF-36.	• Change from baseline in SF-36 scores after 26 and 54 weeks of treatment.
Safety	
• To characterize the safety of AGA BETA BS treatment in participants with Fabry disease previously stabilized with Fabrazyme.	• Evaluation of general health based on the analysis of laboratory values for hematology and clinical chemistry from blood samples collected at baseline and after 14, 26, and 54 weeks of treatment. • Evaluation of renal function based on the analysis of laboratory values for BUN, eGFR, urine albumin-creatinine ratio, electrolytes, and phosphate, from blood and urine samples collected at baseline and after 14, 26, and 54 weeks of treatment. • Evaluation of cardiac function based on the analysis of electrocardiogram and bidimensional echocardiogram exams performed at baseline and after 26 and 54 weeks of treatment.

Objectives	Endpoints
	• Evaluation of immunogenicity based on the analysis of anti-AGA levels from blood samples collected at baseline and after 14, 26, and 54 weeks of treatment. • Analysis of data obtained from clinical and physical assessments, and from reported adverse events and IARs throughout the clinical trial.

Abbreviations: AGA BETA BS=Agalsidase Beta Biosidus; Anti-AGA=anti-agalsidase antibodies; BPI=Brief Pain Inventory; BUN=blood urea nitrogen; eGFR=estimated glomerular filtration rate; IARs=infusion-associated reactions; Lyso-Gb3=globotriaosylsphingosine; SF-36=36-Item Short Form Health Survey.

Overall Design:

BIO-AGA-Fase III-001 is a Phase III, prospective, multicenter, open-label, single-group, baseline-controlled, switch over clinical trial to evaluate the efficacy and safety of AGA BETA BS in patients with FD already treated and previously stabilized with Fabrazyme®.

The study will be conducted in 2 parts: a 5-week Lead-in period (period 1) and 54-week treatment period (period 2). During period 1 all participants will receive 2 intravenous (IV) infusions of Fabrazyme®, provided by Biosidus. After that, in period 2 all participants will switch treatment to AGA BETA BS.

The design of this study is based on that used for the only biosimilar approved in an ICH member country (Japan). In both, a "patient's own historical control" is used as a control, in which the efficacy of AGA BETA BS is measured by comparing the value of the surrogate marker (Lyso-Gb3) at baseline - after at least 6 months of treatment with Fabrazyme® - compared to the value at 6 and 12 months of treatment with AGA BETA BS. This will be a paired comparison that maximizes the ability to detect variations in treatment efficacy.

According to the FDA, in its document: "Rare Diseases: Common Issues in Drug Development Guidance for Industry 2019 Feb.", a historical control, is acceptable in two situations: (1) diseases where the course of the disease can be objectively verified and measured, and (2) in cases where concurrent controls are impractical.

We consider that this study is framed in both situations for the following reasons::

- Since FD is a low prevalence disease and considered rare (orphan), two-arm clinical trials are more complex, as it is very difficult to achieve a sufficient sample size (n).
- Lyso-Gb3 is a verifiable and objective marker of disease course, widely accepted as a short-term follow-up parameter.[17]
- Lyso-Gb3 also meets FDA surrogate marker criteria (*1992 FDA's Accelerated Approval regulations*).
- This single arm design has already been used and approved by the regulatory agency in Japan[16], where a formulation of agalsidase beta was shown to be biosimilar to the original formulation (also using a biomarker as an efficacy criterion). This biosimilar

was approved by the regulatory agency at the end of the study, and is currently commercialized.
- This design has also been used in the comparative study of agalsidase beta conducted in South Korea that led to the approval and commercialization of FABAGAL®[18].

Number of Participants:

Around 20 participants are planned for the study. Participants will have previous diagnosis of FD, will be aged at least 16 years and no more than 60 years, will have received Fabrazyme® for at least 6 months, with treatment compliance of at least 80% of the prescribed dose during the last 3 months, and with disease status considered clinically stabilized, at the Investigators' discretion.. Male with classic FD phenotype, female with classic FD and men with late onset may be included. Participants with chronic kidney disease in stage 3b, 4 or 5, who have suffered a clinical cardiovascular or cerebrovascular event in the last 6 months, who have acute kidney injury in the last 12 months or who have clinically significant unstable cardiac disease will be excluded from the trial.

Intervention Groups and Duration:

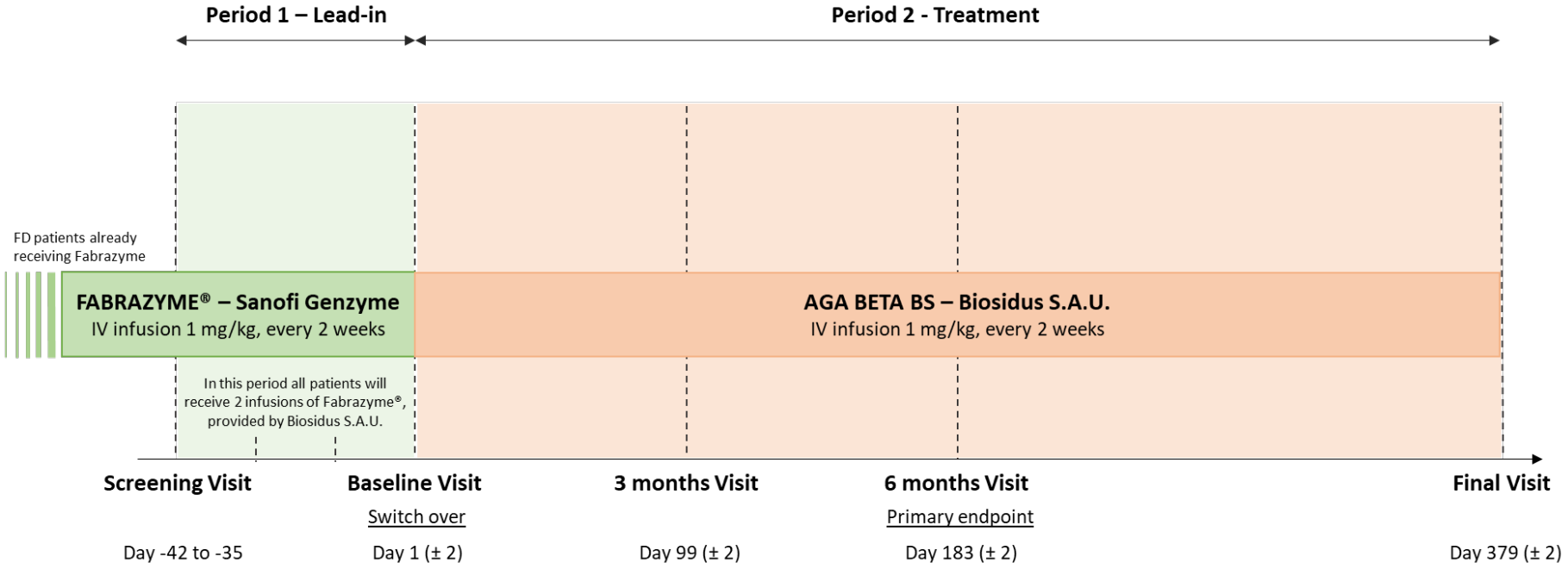
All participants will receive 2 doses of Fabrazyme® first, administered as an IV infusion at a dose of 1.0 mg/kg of body weight with 14 days between doses. After that, all participants will receive AGA BETA BS, administered at dose of 1 mg/kg of body weight, infused every 2 weeks as an IV infusion, for up to 54 weeks (27 infusions).

1. Non-investigational prophylactic pretreatment with antihistamine will be administered at the Investigator's discretion to avoid infusion-associated reactions during the study, except during the first 2 infusions in period 2 in which administration will be compulsory. The antihistamine will be loratadine 10 mg, 1 to 2 hours before infusion, oral route. Furthermore, patients must remain in observation at the study center for at least 1 hour after each infusion during the first four AGA BETA BS infusions. In subsequent infusions, the center staff will contact each patient within 24 hours after infusion to check their clinical condition.

Data Monitoring/Other Committee: No

1.2 Schema

Figure 1 Study Schema



AGA BETA BS=Agalsidase Beta Biosidus; FD=Fabry disease; IV=intravenous.

1.3 Schedule of Activities

Table 1 Schedule of Activities

Procedure / Study Period	Period 1 - Lead-in		Period 2 - Treatment				
	Screening Visit	**Fabrazyme® Infusion Appointments**	**Baseline Visit**	**AGA BETA BS infusion Appointments**	**3 months Visit (After 14 weeks of treatment)**	**6 months Visit (After 26 weeks of treatment)**	**Final Visit (After 54 weeks of treatment)**
Visit day (window)	***Day -42 to -35***	***Day -28 (±2) Day -14 (±2)[1]***	***Day 1 (±2)[1]***	***Every 14 days (±2)[1]***	***Day 99 (±2)[1]***	***Day 183 (±2)[1]***	***Day 379 (±2)[1]***
Informed consent	●						
Inclusion and exclusion criteria	●	●[2]					
Demography and height	●						
Weight	●	●	●	●	●	●	●
Full physical examination	●						●
Medical history (including documented confirmed diagnosis of FD)	●						
Past and current medical conditions	●						
Urine pregnancy test (WOCBP only)[3]	←						→
Laboratory tests[4]	●[5]		●		●[6]	●	●
12-lead ECG	●[5]					●	●
Echocardiogram	●[5]					●	●
Vital signs[7]	●	●	●	●	●	●	●
SF-36			●			●	●
BPI-short form			●			●	●

Procedure / Study Period	Period 1 - Lead-in		Period 2 - Treatment				
	Screening Visit	**Fabrazyme® Infusion Appointments**	**Baseline Visit**	**AGA BETA BS infusion Appointments**	**3 months Visit (After 14 weeks of treatment)**	**6 months Visit (After 26 weeks of treatment)**	**Final Visit (After 54 weeks of treatment)**
Visit day (window)	***Day -42 to -35***	***Day -28 (±2) Day -14 (±2)[1]***	***Day 1 (±2)[1]***	***Every 14 days (±2)[1]***	***Day 99 (±2)[1]***	***Day 183 (±2)[1]***	***Day 379 (±2)[1]***
Phone contact[8]		●	●	●	●	●	●
Fabrazyme® infusions		●					
AGA BETA BS infusions			●	●	●	●	
Anti-AGA[9]			●		●	●	●
Lyso-Gb3 (serum)[9]			●		●	●	●
Delivery of the diary to the participant	●						
Participant diary review[10]		●	●	●	●	●	●
IARs and AE assessment		←	—	—	—	—	→
Concomitant medication review		←	—	—	—	—	→

Abbreviations: AE=adverse event; AGA BETA BS= Agalsidase Beta Biosidus; anti-AGA=antiagalsidase antibodies; BPI=Brief Pain Inventory; COVID-19=coronavirus disease 2019; ECG=electrocardiogram; FD=Fabry disease; HDL=high-density lipoprotein; IARs=infusion-associated reactions; IV=intravenous; LDL=low-density lipoprotein; Lyso-Gb3=globotriaosylsphingosine; SF-36= 36-Item Short Form Survey; WOCBP=women of childbearing potential.

1. Infusions will take place at infusion appointments and at all visits after the Screening visit, except the final visit. If the participant needs to use the +2 days in a visit, to comply with the 14 ± 2-day gap between infusions, he or she will not be allowed to use the whole visit window in the next visit. In this trial, the participants can use the visit windows if they observe the time between infusions of 14 ±2 days. Patients must remain in observation at the study center for at least 1 hour after each infusion during the first four AGA BETA BS infusions. In subsequent infusions, the center staff will contact each patient within 24 hours after infusion to check their clinical condition.
2. For participants whose blood and urine samples are collected for laboratory tests at the Screening visit, the inclusion and exclusion criteria must be reassessed as soon as the results are available, prior to the first Fabrazyme® infusion. Both Fabrazyme® infusions will be included in the time calculation for the assessment of dose stability (participants must be receiving ≥80% of labeled dose/kg for at least last 3 months - inclusion criterion 9).

3. Pregnancy test should be done every 4 weeks in WOCBP. If there is no protocol visit scheduled in that period, the urine pregnancy test will be arranged in the closest infusion appointment.
4. Laboratory tests include complete hematology, blood urea nitrogen, electrolytes, phosphate, creatinine (with eGFR calculation), cholesterol (total, LDL and HDL), triglycerides, glycemia, bilirubin (total and direct), aspartate and alanine aminotransferases, alkaline phosphatase, total proteins, albumin, gamma glutamyl transferase, first morning urine, and urine albumin-creatinine ratio. The blood and urine samples should be collected before the start of the infusion, if applicable.
5. For participants who present results of exams performed in the last 90 days, these exams do not need to be performed again. The results from exams performed in this visit must be available during the first Fabrazyme® infusion appointment for confirmation of compliance with inclusion/exclusion criteria.
6. In this visit the exams urine albumin-creatinine ratio and creatinine will not be performed.
7. Vital signs include heart rate, respiratory rate, body temperature, and blood pressure. Vital signs will be checked before each infusion and if the investigator deems it necessary, hourly during the infusions, if applicable.
8. Phone calls to the participant must be conducted by the study site personnel 24 to 48 hours before each study appointment or visit to check on symptoms compatible with COVID-19 or other infection. To receive the infusions, participants experiencing any infection must wait for clinical recovery, ie, 48 hours being afebrile. If clinical recovery is not established when an infusion is scheduled, the dose must be delayed.
9. Blood sample for this dosage must be collected before the infusion starts.
10. The diary will be given to the participant for them to record information about AEs and concomitant medications. The participant must be instructed to take the diary with them to each medical visit and each infusion appointment for evaluation.

Table 2 Schedule of Activities in the Event of Discontinuation

Procedure \ Study Period	Time of Intervention Discontinuation	Early Discontinuation Visit (participant withdrawn)
Full physical examination	●	●
Urine pregnancy test (WOCBP only)	●	●
Laboratory tests[1] (hematology and biochemistry)		●
12-lead ECG	●	●
Echocardiogram	●	●
Vital signs[2]	●	●
SF-36	●	●
BPI-short form	●	●
Phone contact – follow-up	●[3]	
Anti-AGA antibodies	●	●
Lyso-Gb3 (serum)	●	●
IARs and AE assessment	●	●
Concomitant medication review	●	●

Abbreviations: AE=adverse event; anti-AGA=anti-agalsidase; BPI=Brief Pain Inventory; COVID-19=coronavirus disease 2019; ECG=electrocardiogram; HDL=high-density lipoprotein; IARs=infusion-associated reactions; LDL=low-density lipoprotein; Lyso-Gb3=globotriaosylsphingosine; SF-36=36-Item Short Form Survey; WOCBP=Women of childbearing potential.

1. Laboratory tests include complete hematology, blood urea nitrogen, electrolytes, phosphate, creatinine (with eGFR calculation), cholesterol (total, LDL and HDL), triglycerides, glycemia, bilirubin (total and direct), aspartate and alanine aminotransferases, alkaline phosphatase, total proteins, albumin, gamma glutamyl transferase, first morning urine and urine albumin-creatinine ratio.
2. Vital signs include heart rate, respiratory rate, body temperature, and blood pressure.
3. After intervention discontinuation, instead of following sequential procedures planned in the study, the participant should receive telephone follow-up at same timepoints of all following planned visits to evaluate general health condition.

2.0 INTRODUCTION

Biosidus' Agalsidase Beta is being developed as a proposed biosimilar product to Fabrazyme® which is planned to meet the need for alternatives to high-priced biologic agents in enzyme replacement therapy (ERT) treatments for Fabry disease (FD).

2.1 Study Rationale

This Phase III trial will evaluate the efficacy and safety of Agalsidase Beta Biosidus (AGA BETA BS) in a switch over study with participants with FD previously stabilized with Fabrazyme®.

The AGA BETA BS clinical development follows the currently understood concepts from regulation 7729/2011 of the Argentine National Administration of Drugs, Food and Medical Devices (ANMAT) for biosimilar development. The approach is to demonstrate a similar behavior in terms of identity, potency and purity profile compared with the reference drug, and a comparable profile of safety and efficacy.

Biosidus' Agalsidase Beta is being developed as a proposed biosimilar product to Fabrazyme, a recombinant human α-galactosidase A used for enzyme replacement therapy in patients with Fabry disease (FD). AGA BETA BS already have shown biosimilarity with Fabrazyme in 7 nonclinical studies involving pharmacodynamics (PD), pharmacokinetics (PK) and toxicit, and in a phase I clinical study.

The latter was a comparative study of pharmacokinetics and pharmacodynamics between AGA BETA BS and Fabrazyme® in healthy volunteers carried out between 2021 and 2022 in the Province of Buenos Aires. It was a sequential, open-label, randomized, parallel two-arm study. Each participant received a single dose of 1 mg/kg body weight of the drug assigned at randomization, at a 1:1 ratio. Participants were followed for 35 days after infusion for safety assessment. Pharmacokinetics, pharmacodynamics and safety results are detailed below: (1)From the average values obtained for pharmacokinetics parameters (Cmax, AUC0-12h and AUC0-∞), the biosimilarity between formulations was analyzed by calculating the ratio of Test/Reference formulations and its 90% confidence interval. All three parameters were found to be within the accepted biosimilarity range of 0.80 - 1.25. (2) The average enzyme activity at the end of infusion (5h) for the Biosidus Agalsidase product was 78.38 ± 20.89 mU/ml and for Fabrazyme® 80.81 ± 28.60 mU/ml. These results showed an enzyme activity ratio at 5h Test/Referent of 0.97. No significant differences were observed between the two treatment groups (p= 0.8210). (3) Regarding the safety profile, AGA BETA BS showed a similar behavior to the comparator, both in the number of adverse events and in the category and degree of severity. There were no serious adverse events. All adverse events were classified as mild by the principal investigator. Those classified as "possible" in terms of causality relationship with the investigational drug were expected and were described in the investigator's brochure and the reference product package insert. No neutralizing anti-

agalsidase antibodies were detected in any volunteer, neither in the pre-dose samples, nor at 12h post start of infusion nor at 35 days post administration.

In line with the ANMAT Provision 7729/2011, it is proposed to carry out this clinical trial to evaluate the similarity of AGA BETA BS, in terms of safety and efficacy, with the innovator product. Using the same reference product as in previous studies. This is a comparative study, under the same conditions as the authorized use of the innovator product (IV infusion every two weeks, at a dose of 1 mg/kg of body mass), in the indication approved for the innovator product (Fabry disease) adapted to the recommendations accepted in other regulatory agencies for low prevalence diseases.

2.2 Background

Fabry disease is a rare X-chromosomal-linked lysosomal storage disorder due to various pathogenic mutations in the *GLA* gene, responsible for codifying the lysosomal enzyme α-galactosidase A (α-Gal A). The reduction or complete lack of activity of this enzyme results in slow and progressive accumulation of glycosphingolipids, mainly globotriaocylceramide (Gb3 or GL3) and its deacylated derivative globotriaosylsphingosine (Lyso-GL3, also called Lyso-Gb3) in several cell types throughout the body, affecting the heart, kidneys, skin, eyes, central nervous system, and gastrointestinal system, ultimately generating severe, potentially life-threatening target-organ complications.[1,2,3] Together, the numerous signs and symptoms of FD significantly reduce quality of life and shorten life expectancy by 10 to 20 years.4

Fabry disease encompasses a spectrum of phenotypes ranging from the more severe classic phenotype to atypical late onset forms. The classic manifestation of FD occurs predominantly in men that generally show less than 1% α-Gal A enzyme activity and it usually has its onset in childhood or adolescence.[1,2,5] The nonclassical or atypical phenotype of FD involves residual enzyme activity that leads to generally milder symptoms, with a later onset of 40 to 60 years of age.6 The clinical presentation of FD in women is highly variable, ranging from asymptomatic patients to patients with symptoms as severe as those observed in males with the classic phenotype.1

Until a few decades ago, there was no effective treatment available for inherited lysosomal storage diseases such as FD. The management consisted only of supportive care and pharmacologic management for symptoms control.5 Today, even if it is not a cure, patients with FD have 2 therapeutic modalities available: ERT and oral pharmacological chaperones (PCT).4 Enzyme replacement therapy, which consists of systemic α-Gal A infusion and was the first approved treatment for FD,4 increases the enzyme levels in the body, while PCT have been shown to promote the correct folding of amenable mutated glycosidases and retrieve residual activity levels.5 Other strategies, such as substrate reduction therapy, messenger RNA-based therapy, and gene therapy are in development.4

There are 2 available pharmaceutical preparations of recombinant human α-Gal A approved and used worldwide: agalsidase alfa (Replagal® by Shire) and agalsidase beta (Fabrazyme® by Sanofi Genzyme). While agalsidase alfa is produced by overexpression in human fibroblasts, agalsidase beta is produced by overexpression in chinese hamster ovary cells.4 Both preparations are administered intravenously every 14 days and have similar glycosylation patterns, specific activities, enzyme kinetics,4 and both have been shown to be clinically equally efficacious,1,2 decreasing the accumulation of GL3 in kidney, heart, and dermal cells, improving pain and maintaining kidney functions. Both were approved in 2001 by the European Agency for Evaluation of Medical Products; only Fabrazyme® was approved by the Food and Drugs Administration (FDA) for use in the United States (US).2

Although ERT (Enzyme Replacement Therapy) consists in an efficacious treatment with good tolerance, the treatment leads to high costs, with an average of 250,000 €/year per patient (considering an adult weighing 70 kg).7 Therefore, other treatment options in addition to those already approved are under study.

2.3 Benefit/Risk Assessment

Participants enrolled into this equivalence trial are expected to derive similar benefit and risk from the treatment already experienced before the enrollment, with Fabrazyme®. This is based on the similarity observed during PD, PK, and toxicological testing between the investigational medicinal product and Fabrazyme®. In addition, the safety profile of AGA BETA BS observed in the Phase I trial was similar to that of Fabrazyme®, therefore, in this clinical trial, adverse events (AEs) beyond those already known are not expected.

Based on PD, PK, and toxicological testing carried out prior to initiation of this trial, and the Phase I clinical trial's data described above, AGA BETA BS, as a proposed biosimilar product, may be seen to provide similar efficacy, safety, and immunogenicity to Fabrazyme® in patients with FD.

More detailed information about the known and expected benefits and risks and reasonably expected AEs of agalsidase beta may be found in the current Investigator's Brochure.

The Sponsor will immediately notify the Principal Investigator if any additional safety or toxicology information becomes available during the study.

This study will be performed in compliance with the protocol, International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) Good Clinical Practice (GCP), European Union (EU) Clinical Trial Regulation, and applicable regulatory requirements.

2.3.1 Risk Assessment

The risk arising from the treatment is based on the occurrence of adverse reactions. It is expected that many patients will develop Immunoglobulin G antibodies upon treatment with agalsidase beta, and patients with antibodies have a higher risk of infusion-associated reactions (IARs). In clinical trials with Fabrazyme®, IARs were assessed as mild or moderate in intensity and were the most frequently reported related AEs, including chills, fever, temperature change sensation, hypertension, nausea, vomiting, flushing, paresthesia, fatigue, pain, headache, chest pain, and pruritus.[8] Life-threatening anaphylactic and severe allergic reactions are uncommon.[9]

The IARs have been successfully managed using standard medical practices, such as reduction in infusion rate and/or premedication with, or additional administration of nonsteroidal anti-inflammatory drugs, antipyretics, antihistamines and/or corticosteroids.8

As most IARs occur in the first few months of treatment,[9] all participants will have their infusions at the study center, with direct physician supervision, so as to manage and treat any adverse reactions that could be presented, even if they already are receiving Fabrazyme® for a considerable amount of time, where the IAR's frequency is low or null due to the continued use of agalsidase beta and.[8] Additionally, antihistamine will be administered to avoid IARs obligatorily in the first 2 infusions with the experimental drug.

Additionaly, patients must remain in observation at the study center for at least 1 hour after each infusion during the first four AGA BETA BS infusions. In subsequent infusions, the center staff will contact each patient within 24 hours after infusion to check their clinical condition.

Participant risk will be minimized by implementing conservative eligibility criteria, regular and long-term safety monitoring, including immunogenicity testing.

2.3.2 Benefit Assessment

Despite still having aspects to be explored and understood, ERT is a well-established treatment that has been shown to delay the progression of a multisystemic disease and organ involvement and represents the current standard-of-care therapeutic option for FD.[10]

The response to ERT varies considerably and appears to depend on gender, genotype (classic or later onset/nonclassic), stage of disease or age and agalsidase inhibition by antiagalsidase antibodies (anti-AGA).[10]

The benefit of ERT for patients with FD has been documented in the clinical literature. A Cochrane review of randomized controlled trials showed that ERT reduces microvascular endothelial GL3 deposits and improves pain-related quality of life compared with placebo.[11] This review was complemented by a pooled analysis of cohort studies that showed that

patients taking agalsidase beta had a significantly lower incidence of renal, cardiovascular, and cerebrovascular events than patients not taking ERT.[12] A meta-analysis of double-blind and randomized clinical trials suggests that ERT with agalsidase alfa or beta reduces the severity of neuropathic pain, improves pain-related quality of life, brings about minor changes in left ventricular mass, reduces serum, urine and tissue levels of GL3, and is able to slow down the progression of FD.[13] Additionally, a 10-year study of ERT efficacy in adults reported a survival rate of 94% and most patients (81%) had no severe clinical events during this time. Patients who initiated treatment at a younger age and with less kidney involvement benefited the most from therapy, suggesting that early initiation of ERT could slow or even prevent renal disease progression.[14] The results of Fabrazyme®' published randomized controlled clinical trials and their extension studies showed clearance of GL3 in renal cells and cardiac endothelial cells after 6 months of treatment, significant risk reduction (-61%) of renal, cardiac, cerebrovascular complications and death, significant improvement in pain scores and quality of life, as well as significant reductions in school absences due to sickness and gastrointestinal symptoms.[8,15]

Therefore, participants of this trial will have the benefit of receiving a potentially effective Fabrazyme®'s biosimilar drug free of charge, in addition to being able to contribute to the process of developing a therapy as effective as the current treatment and at a lower cost. In addition, during the study the participant will receive medical evaluations and laboratory tests free of charge as part of the trial.

2.3.3 Overall Benefit Risk Conclusion

Considering the measures taken to minimize risk to participants participating in this study, the potential risks identified in association with study intervention are justified by the anticipated benefits that may be afforded to participants with FD.

3.0 OBJECTIVES AND ENDPOINTS

Table 3 presents the primary, secondary, and safety objectives and corresponding endpoints.

Table 3 Study Objectives and Corresponding Endpoints

Objectives	Endpoints
Primary	
• The primary objective of the study is to evaluate the equivalence in efficacy between AGA BETA BS and Fabrazyme® after 6 months of treatment in participants with Fabry disease previously stabilized with Fabrazyme, by measuring disease biomarker.	• Mean serum Lyso-Gb3 marker ratio after 26 weeks of treatment, defined as plasma level of the marker Lyso-Gb3 after 26 weeks (6 months) divided by plasma level of the marker Lyso-Gb3 at baseline.
Secondary	
• To evaluate the difference in efficacy between AGA BETA BS and Fabrazyme® after 1 year of treatment in participants with Fabry disease previously stabilized with Fabrazyme®, by measuring disease biomarker.	• Mean serum Lyso-Gb3 marker ratio after 54 weeks of treatment, defined as plasma level of the marker Lyso-Gb3 after 54 weeks (12 months) divided by plasma level of the marker Lyso-Gb3 at baseline.
• To compare the pain severity before and after the treatment with AGA BETA BS in participants with Fabry disease previously stabilized with Fabrazyme®, as measured by the BPI-short form.	• Change from baseline in pain severity as assessed by BPI-short form pain severity items scores after 26 and 54 weeks of treatment.
• To compare the impact of pain on daily functions before and after the treatment with AGA BETA BS in participants with Fabry disease previously stabilized with Fabrazyme®, as measured by the BPI-short form.	• Change from baseline in pain interference as assessed by BPI-short form pain interference items scores after 26 and 54 weeks of treatment.
• To compare the participants' perception of their own health before and after the treatment with AGA BETA BS in participants with Fabry disease previously stabilized with Fabrazyme®, as measured by the SF-36.	• Change from baseline in SF-36 scores after 26 and 54 weeks of treatment.
Safety	
• To characterize the safety of AGA BETA BS treatment in participants with Fabry disease previously stabilized with Fabrazyme®.	• Evaluation of general health based on the analysis of laboratory values for hematology and clinical chemistry from blood samples collected at baseline and after 14, 26, and 54 weeks of treatment. • Evaluation of renal function based on the analysis of laboratory values for BUN, eGFR, urine albumin-creatinine ratio, electrolytes, and phosphate, from blood and urine samples collected at baseline and after 14, 26, and 54 weeks of treatment. • Evaluation of cardiac function based on the analysis of electrocardiogram and bidimensional

Objectives	Endpoints
	echocardiogram exams performed at baseline and after 26 and 54 weeks of treatment. • Evaluation of immunogenicity based on the analysis of anti-AGA levels from blood samples collected at baseline and after 14, 26, and 54 weeks of treatment. • Analysis of data obtained from clinical and physical assessments, and from reported adverse events and infusion-related reactions throughout the clinical trial.

Abbreviations: Anti-AGA=antiagalsidase antibodies; AGA BETA BS=Agalsidase Beta Biosidus; BPI=Brief Pain Inventory; BUN=blood urea nitrogen; eGFR=estimated glomerular filtration rate; Lyso-Gb3=globotriaosylsphingosine; SF-36=36-Item Short Form Health Survey.

4.0 STUDY DESIGN

4.1 Overall Design

This is a Phase III, prospective, multicenter, open-label, single-group, baseline-controlled, switch over clinical trial to evaluate the efficacy and safety of AGA BETA BS treatment in patients with FD already treated and previously stabilized with Fabrazyme.

The planned clinical trial aims to establish the equivalence of AGA BETA BS to Fabrazyme®.

A schema of the trial design is shown in Section 1.2.

Approximately 20 participants aged ≥16 years and ≤60 years with FD who are receiving Fabrazyme® for at least 6 months, have received at least 80% of the prescribed dose during the last 3 months, and with disease status considered clinically stabilized at the Investigators' discretion, will be enrolled in this trial.

Screening will be performed between 35 to 42 days prior to the first dose of study experimental treatment. After all protocol-required safety laboratory tests (refer to Section 10.4), eligible participants will enter the study that comprises 2 periods:

1. Lead-in period: all participants will receive 2 intravenous (IV) infusions of Fabrazyme®, provided by Biosidus, at a dose of 1 mg/kg of body mass, with 14 days between doses. The participant's baseline status will be assessed at the end of this period.
2. Treatment period: all participants will switch treatment to AGA BETA BS, administered at dose of 1 mg/kg of body mass, infused every 2 weeks as an IV infusion, for up to 54 weeks. Participants will receive a total of 27 infusions.

During each infusion, participants will be monitored constantly to ensure prompt treatment for eventual IARs that could be presented. Prophylactic pretreatment with antihistamines to avoid IARs will be at the Investigator's discretion during the study, except during the first 2 infusions in period 2 in which administration will be compulsory. In addition, patients must remain in observation at the study center for at least 1 hour after each infusion during the first four AGA BETA BS infusions. In subsequent infusions, the center staff will contact each patient within 24 hours after infusion to check their clinical condition (refer to Section 6.8).

Preferably, all infusions should be performed at the study sites. The population to be evaluated in this study is already being treated with Fabrazyme® and, in accordance with the clinical

practice for patients receiving ERT[9], in most cases it is at home. To allow full participation in the study and to avoid a significant number of drop-outs, infusions may be administered at the participant's home after 4 infusions of AGA BETA BS at the site, at the discretion of the Investigator. Before making this decision, the Investigator must ensure that AGA BETA BS infusions can be safely, effectively and reliably administered at the participant's home. Among other requirements that the Investigator may consider, it is recommended that the participant meet at least the following main criteria:

- The participant is considered medically stable. A comprehensive assessment must be completed prior to deciding on transfer of therapy.
- The participant must have a proven pattern of well-tolerated infusions.
- The participant must have a history of compliance with the prescribed infusion schedule.

The study site personnel are required to contact the participant by phone call or message texts in 2 situations: (1) before each study appointment or visit, for all participants, and (2) within 24 hours after infusion from the fifth infusion of AGA BETA BS. Refer to Section 8.1.5 for detailed information.

The trial will last approximately 60 weeks (including a 5-week Lead-in period and a treatment period of up to 54 weeks) and will have 5 key medical visits. Participants will undergo visits and trial procedures as shown in Schedule of Activities (SoA) in Section 1.3.

Refer to Section 10.3.5 for details on informed consent and to Section 10.3.11 for a definition of study start and a definition of “enrollment”.

4.2 Scientific Rationale for Study Design

An open-label design was selected as all participants will switch Fabrazyme® for AGA BETA BS treatment.

Standard statistical, clinical, and laboratory procedures will be utilized in this study. All efficacy and safety-related measurements in this study are standard for assessing disease activity in participants with FD. All clinical and laboratory procedures in this study are standard and generally accepted.

This historical-controlled switch over design is the basis for rare disease trials due to the very limited pool of eligible patients to be studied, and follows that of 3 clinical trials using ERT treatment in participants with FD (NCT03180840, NCT03018730, and specially the Japanese Phase II/III bioequivalence trial for the first Fabrazyme® biosimilar drug –JR-051[16]– approved in a highly regulated market, by Japan’s Ministry of Health, Labor, and Welfare in September 2018).The efficacy of AGA BETA BS will be measured by comparing the value of

the surrogate marker (Lyso-Gb3) at baseline - after at least 6 months of treatment with Fabrazyme® - compared to the value at 6 and 12 months of treatment with AGA BETA BS. This will be a paired comparison that maximizes the ability to detect variations in treatment efficacy.

According to the FDA, in its document: "Rare Diseases: Common Issues in Drug Development Guidance for Industry 2019 Feb.", a historical control, is acceptable in two situations: (1) diseases where the course of the disease can be objectively verified and measured, and (2) in cases where concurrent controls are impractical.

We consider that this study is framed in both situations for the following reasons:

- Since FD is a low prevalence disease and considered rare (orphan), two-arm clinical trials are more complex, as it is very difficult to achieve a sufficient sample size (n).
- Lyso-Gb3 is a verifiable and objective marker of disease course, widely accepted as a short-term follow-up parameter.[17]
- Lyso-Gb3 also meets FDA surrogate marker criteria (*1992 FDA's Accelerated Approval regulations*).
- This single arm design has already been used and approved by the regulatory agency in Japan[16], where a formulation of agalsidase beta was shown to be biosimilar to the original formulation (also using a biomarker as an efficacy criterion). This biosimilar was approved by the regulatory agency at the end of the study, and is currently commercialized.
- This design has also been used in the comparative study of agalsidase beta conducted in South Korea that led to the approval and commercialization of FABAGAL®[18].

Serum Lyso-Gb3 is a standard worldwide biomarker useful for both diagnosis and therapeutic evaluation of patients during ERT treatment, and therefore, comprises the primary endpoint in this trial. This primary endpoint is proposed as a surrogate endpoint that is reasonably likely to predict clinical benefit. A clinical benefit is a positive therapeutic effect that is clinically meaningful in the context of a given disease. For FD, a clinical benefit could be demonstrated by evaluation of the core disease progression manifestation (eg, renal failure, cardiovascular disease, cerebrovascular disease) but that may take many years, and, therefore, this type of evaluation may not be feasible in a clinical trial setting. In addition, it is unknown which phase of the disease may be most amenable to demonstrating a clinical impact of treatment, and so it is unknown if the most sensitive portion of the disease population is being studied in this trial.

In fact, for rare diseases such as FD, the FDA recognizes that it can sometimes take many years to learn whether a drug actually provides a real effect on how a patient survives, feels, or functions. Mindful of the fact that it may take an extended period of time to measure a drug's intended clinical benefit, in 1992 FDA instituted the Accelerated Approval regulations. These

regulations allowed drugs for serious conditions that filled an unmet medical need to be approved based on a surrogate endpoint.

Furthermore, AGA BETA BS is being developed as a biosimilar product to Fabrazyme®. Fabrazyme® has already demonstrated acceptable clinical benefit and was approved by the most recognized regulatory agencies worldwide. Therefore, it would be scientifically unnecessary and unethical to submit participants to invasive tests (such as kidney biopsies) or require a long stay in a clinical trial setting.

Although it is not recognized as a direct measure of clinical benefit, but rather a surrogate endpoint, the scientific literature has been suggesting that there is a direct correlation between high levels of Lyso-Gb3 and worse clinical outcomes.[17]

4.3 Justification for Dose

In Argentina, EU, US, and many other countries, Fabrazyme® has received health authority approval for long-term ERT in patients with a confirmed diagnosis of FD. As a biosimilar product, the dose of AGA BETA BS selected for this trial is based on the clinically effective dose of the currently available dosage form of Fabrazyme®.

The primary focus of this trial is to demonstrate equivalence of AGA BETA BS (proposed biosimilar to Fabrazyme®) with Argentina-licensed Fabrazyme®. The recommended dose of Fabrazyme®, administered as an IV infusion, is 1.0 mg/kg of body weight given once every 2 weeks.[8] Therefore, in this study, participants enrolled will receive AGA BETA BS 1.0 mg/kg once every 2 weeks.

4.4 End of Study Definition

The end of the study is defined as the date of the last visit of the last participant in the study.

A participant is considered to have completed the study if he/she has completed all periods of the study including the last visit.

4.5 Study Stopping Criteria

4.5.1 Stopping Criteria for Individual Participants

Criteria for discontinuing of study intervention for an individual participant and the criteria for discontinuing the study for an individual participant are presented in Section 7.0.

4.5.2 Criteria for Stopping the Study

The Sponsor may terminate this study prematurely, either in its entirety or at any site. Refer to Section 10.3.11 for more information.

The Investigator may also stop the study at his/her site if he/she has safety concerns. If the Sponsor terminates the study for safety reasons, the Sponsor will promptly notify the Investigators, the Independent Ethics Committees (IECs)/Institutional Review Boards (IRBs), the regulatory authorities, and any contract research organization(s) used in the study of the reason for termination or suspension, as specified by the applicable regulatory requirements.

5.0 STUDY POPULATION

Prospective approval of protocol deviations to recruitment and enrollment criteria, also known as protocol waivers or exemptions, is not permitted.

5.1 Inclusion Criteria

Participants are eligible to be included in the study only if all the following criteria apply:

Sex and Age

1. Male or female participant with ≥16 and ≤60 years of age at the time of signing the informed consent form (ICF).

Reproduction

2. Female participants who are not pregnant, breastfeeding, donating eggs (ova, oocytes), or considering becoming pregnant during the study and for 3 months after the last dose of study treatment.
3. All women of childbearing potential (WOCBP) must have a negative urine pregnancy test at the Screening visit and at Baseline visit (prior to the first dose of experimental intervention).
4. WOCBP must use one highly effective form of birth control contraception through the study and for 3 months after the last dose of study treatment (refer to Appendix 1 in Section 10.1).
5. Male participants who are not considering fathering a child during the study and for 3 months after the last dose of study treatment.
6. Male sexually active participant with female partner(s) of childbearing potential must agree to use male condoms during the study and for 3 months after the last dose of study treatment or have documented successful surgical sterilization.

Informed Consent

7. Capable of giving signed informed consent which includes compliance with the requirements and restrictions listed in the ICF and in this protocol.

Type of Participant and Characteristics

8. Confirmed previous diagnosis of FD.
    a) Women: preferably present genetic testing showing pathogenic *GLA* mutation consistent with FD at screening.

b) Men: preferably present leukocyte α-Gal A activity below normal range and/ or pathogenic *GLA* mutation consistent with FD at screening.
c) Male with classic FD phenotype, female with classic FD and men with late onset may be included.

9. Participants who have been on stable Fabrazyme® treatment for at least 6 months prior to Baseline visit.

10. Patients that in the last 3 months before the baseline visit have been receiving ≥80% of Fabrazyme®'s labeled dose/kg, this calculation includes both infusions provided by Biosidus during the Lead-in period.

11. Disease status considered clinically stabilized, at Investigators' discretion.

12. Estimated glomerular filtration rate (eGFR) ≥45 mL/minute/1.73 m^2 by CKD-EPI equation at Screening visit.

13. If receiving pain killers, angiotensin-converting enzyme (ACE) inhibitors or angiotensin II receptor blockers (ARBs), participants must be in a stable dose for ≥ 4 weeks.

5.2 Exclusion Criteria

Participants are excluded from the study if any of the following criteria apply:

Medical Conditions

1. Chronic kidney disease in stage 3b, 4, or 5.

2. History of dialysis, kidney transplant or participants who are on the waiting list for a kidney transplant.

3. Proteinuria ≥1 g/day at screening.

4. Participants who have suffered a clinical cardiovascular event (such as but not limited to myocardial infarction, transient ischemic attack) within 6 months prior to Screening visit.

5. Participants who have clinically significant unstable cardiac disease (such as but not limited to uncontrolled symptomatic arrhythmia, unstable angina, congestive heart failure New York Heart Association class III or IV).

6. Participants who have suffered a clinical cerebrovascular event (such as but not limited to stroke, transient ischemic attack) within 6 months prior to Screening visit.

7. History of anaphylaxis or other type I hypersensitivity reactions to agalsidase beta.

8. History of acute kidney injury in the 12 months prior to Screening visit (such as but not limited to acute interstitial nephritis, acute renal failure of glomerular origin or caused by vasculitis).

9. Presence of any medical, emotional, behavioral, or psychological condition that, according to the Investigator, would interfere with the participant's compliance with the requirements of the study.

Prior/Concomitant Therapy

10. Treatment initiation or change of dose of ACE inhibitors or ARBs in the 4 weeks before the screening.

Prior/Concurrent Clinical Trial Experience

11. Current participation in an interventional study, in which the participant received any drug within 90 days before the Screening visit.

5.3 Lifestyle Considerations

No lifestyle restrictions are required during any of the study periods.

5.4 Screen Failures

A screen failure occurs when a participant who consents to participate in the clinical study is not subsequently entered in the study. A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants to meet the Consolidated Standards of Reporting Trials publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details, eligibility criteria, and any serious adverse event (SAE).

Individuals who do not meet the criteria for participation in this study (screen failure) may be rescreened, at Investigator’s discretion. Rescreened participants should be assigned a new identification number for every screening/rescreening event. For rescreened participants, the same previously signed ICF should be used unless more than 30 days have passed since the first ICF was signed. In these cases, another ICF must be signed.

6.0 STUDY INTERVENTION(S) AND CONCOMITANT THERAPY

Study intervention is defined as any investigational intervention(s), marketed product(s), placebo, or medical device(s) intended to be administered to a study participant according to the study protocol.

6.1 Study Intervention(s) Administered

The study has a single-arm. First, all participants will receive 2 doses of Fabrazyme® with approximately 14 days between them, and afterwards all participants will switch treatment and receive AGA BETA BS for 54 weeks. Details of both medications are provided in Table 4.

Table 4 Study Intervention(s) Administered

Intervention Label	Fabrazyme®	AGA BETA BS
Intervention Name	Recombinant human alpha-galactosidase A (agalsidase beta)	Recombinant human alpha-galactosidase A (agalsidase beta)
Intervention Description	Agalsidase beta from Sanofi Genzyme	Agalsidase beta from Biosidus
Type	Biological	Biological
Dose Formulation	Lyophilized powder for reconstitution	Lyophilized powder for reconstitution
Unit Dose Strength(s)	35 mg and/or 5 mg	35 mg and/or 5 mg
Dosage Level(s)	1 mg/kg of body mass every 2 weeks	1 mg/kg of body mass every 2 weeks
Route of Administration	IV infusion	IV infusion
Use	Lead-in intervention/background intervention (study period 1)	Experimental intervention (study period 2)
IMP and NIMP/AMP	IMP	IMP
Sourcing	Provided centrally by the Sponsor	Provided centrally by the Sponsor
Packaging and Labeling	Study intervention will be provided in cartons containing 1, 5, or 10 single-use glass vials with 35 mg or 5 mg of agalsidase beta. Each carton will be labeled as required per country requirement.	Study intervention will be provided in cartons containing 1, 5, or 10 single-use glass vials with 35 mg or 5 mg of agalsidase beta. Each carton will be labeled as required per country requirement.

Abbreviations: AMP=auxiliary medicinal products; IMP=investigational medical product; IV=intravenous; NIMP=noninvestigational medical product.

6.2 Preparation, Handling, Storage, Administration and Accountability

6.2.1 Packaging and Labeling

The study interventions will be provided by the Sponsor in sufficient quantity to supply the treatment of all participants included in the trial.

Each carton of study intervention will be labeled per local requirements and this label must remain affixed to the carton. Upon receipt, study treatment should be stored as specified on the label and kept in a secure location. The two investigational drugs (Fabrazyme® and AGA BETA BS) will be distinguishable. Study intervention will only be used for the conduct of this study.

6.2.2 Storage and Accountability

Both investigational drugs must be stored at 2°C to 8°C (36°F to 46°F). All investigational product provided by the Sponsor must be stored according to the storage conditions on the clinical label. The investigational products are for investigational use only and are to be used only within the context of this study. The study treatment supplied for this study must be maintained under adequate security and stored under the conditions specified on the label until dispensed for participant use or returned to the Sponsor, as appropriate. The Investigator or designee must confirm appropriate temperature conditions have been maintained during transit for all study treatment received, and any discrepancies are reported and resolved before use of the study treatment.

Only participants enrolled in the study may receive study intervention, and only authorized study site staff may supply or administer study intervention. All study intervention must be stored in a secure, environmentally controlled, and monitored (manual or automated) area in accordance with the labeled storage conditions with access limited to the Investigator and authorized study site staff.

The Investigator, institution, or the head of the medical institution (where applicable) is responsible for study intervention accountability, reconciliation, and record maintenance (ie, receipt, reconciliation, and final disposition records).

The Investigator, a member of the study site staff, or the institution's pharmacist must maintain an adequate record of the receipt and distribution of all study intervention using the Drug Accountability Form. These forms must be available for inspection at any time.

All unused study interventions must be returned to the Sponsor at the end of the trial in accordance with current legislation.

6.2.3 Reconstitution and Administration

Guidance about reconstitution and dose preparation for Fabrazyme® and AGA BETA BS and the necessary materials for the infusion are provided in Appendix 2 (Section 10.2).

For both investigational drugs, the initial IV infusion rate will be no more than 0.25 mg/minute (15 mg/hour). The infusion rate may be slowed in the event of IARs. After participant tolerance to the infusion is well established, the infusion rate may be increased gradually in increments of 0.05 to 0.08 mg/minute (increments of 3 to 5 mg/hour) with each

subsequent infusion. For participants weighing ≥30 kg, the administration duration should not be less than 1.5 hours (based on individual participant tolerability).

6.3 Measures to Minimize Bias: Randomization and Blinding

This is a nonrandomized, open-label study. There are no stratification factors in this study.

At the Screening visit, all participants who consent will be assigned a unique identification number using a trial supply management system. For participants who do not meet the study selection criteria, the site personnel must log on to the system and identify the participant as a screen failure. Each participant will be assigned only one identification number. Identification numbers must not be reused for different participants. Rescreened participants should be assigned a new identification number for every screening/rescreening event.

Participants who are enrolled will retain their identification numbers assigned at the Screening visit throughout the study. Upon receipt of study treatment, the site will acknowledge receipt in the system.

6.4 Study Intervention Compliance

Participants will be dosed at the study site, directly from the Investigator or designee. The date and time of start of each infusion, as well as the dose and total volume infused, will be recorded in the source documents.

The prescribed dosage, timing, and mode of administration may not be changed. Any departures from the intended regimen must be recorded in the electronic case report form (eCRF).

Noncompliance is defined as taking less than 80% of study treatment during any evaluation period (visit to visit).

6.5 Participants showing poor compliance as assessed by appointments and/or visits attendance for their drug infusions must be counseled on the importance of good compliance to the trial dosing regimen.Dose Modification

Dose modifications are not planned or allowed in this study.

Any dose deviation must be recorded in the eCRF.

In the event of a life-threatening reaction, including anaphylactic or severe allergic reactions, AGA BETA BS will be discontinued, and necessary emergency treatment will be initiated.

6.6 Continued Access to Study Intervention After the End of the Study

Following ANMAT Disposition Nº6677/10, the Sponsor will grant participants who have completed all the study visits and interventions, including all the AGA BETA BS IV infusions, access to the intervention that has been beneficial and medical follow-up, according to individual medical judgment, or to an alternative intervention until access to it is guaranteed by any other means.

6.7 Treatment of Overdose

The Sponsor does not recommend specific treatment for an overdose. There have been no reports of overdose with agalsidase beta. In clinical trials, participants received doses up to 3.0 mg/kg body weight.

In the event of an overdose, the Investigator should:

- Contact Biosidus' pharmacovigilance department immediately.
- Evaluate the participant to determine, in consultation with the Biosidus' pharmacovigilance department, whether study intervention should be interrupted or whether the dose should be reduced.
- Closely monitor the participant for any AE/SAE and laboratory abnormalities until study intervention can no longer be detected systemically (at least 12 hours).
- Document the quantity of the excess dose as well as the duration of the overdose.

6.8 Concomitant Therapy

Any medication or vaccine (including over-the-counter or prescription medicines, recreational drugs, vitamins, and/or herbal supplements) that the participant is receiving at the time of enrollment or receives during the study must be recorded on the eCRF along with:

- Reason for use
- Dates of administration including start and end dates
- Dosage information including dose and frequency
- For vaccines (if applicable) include brand name and manufacturer (plus lot number, if available)

The participant will receive a diary to record any concomitant medication that he or she may use during the study. The delivery of the diary to the participant and its collection for evaluation is described in the SoA (Section 1.3). The participant must be instructed to take the diary with them to each medical visit and each infusion appointment for evaluation. All information collected in the diary must be recorded on the eCRF.

Any questions regarding concomitant or prior therapy should be raised to the Sponsor's emergency contact.

Non-investigational pretreatment with antihistamine will be administered to avoid IARs at the Investigator's discretion in period 1 but will be compulsory during the first 2 infusions in period 2. The antihistamine pretreatment will also be at Investigator's discretion on the following infusions. This medication will be provided by Biosidus to study sites and is described in Table 5. When used, the antihistamine medication must be recorded as concomitant medication.

Table 5 Noninvestigational Antihistaminic Pretreatment

Drug Name	Loratadine
Study Treatment Name:	Loratadine
Type	Drug
Dosage Formulation:	Tablet
Unit Dose Strength(s)/Dosage Level(s):	10 mg
Route of Administration	Oral
Dosing Instructions:	1 tablet 1-2 hours before infusions
Packaging and Labeling	As required per country requirement
Manufacturer	Any approved and commercially available formulation will be used

Other medications such as antipyretic and/or corticosteroid may be administered at the Investigator's discretion to avoid or manage IARs in participants who had experienced them during agalsidase beta treatment or those who developed them during the trial. The selection of pretreatment medication and dose should be based on the participant's age, weight, and severity of the reaction. The time of administration should be based on the onset of action of the medication selected. All medications administered as pretreatment or to manage IARs must be recorded as concomitant medication on the eCRF.

6.8.1 Prohibited and Allowed Concomitant Medications

There are no prohibited medications during the trial. However, use of drugs that increase thrombosis risk, such as contraceptives, should be made with special caution and after a risk benefit assessment performed by the Investigator in each case, since they may theoretically additionally increase the risk of transient ischemic attack/cerebrovascular accident in females with FD.

Vaccination with live vaccines while on study is prohibited. Administration of inactivated vaccines is allowed (eg, inactivated influenza vaccines or severe acute respiratory syndrome coronavirus 2 vaccines).

Antiproteinuric drugs and pain killers are allowed as long as they haven't been started or the dose changed in the 4 weeks before screening (refer to inclusion criterion number 12 and

exclusion criteria number 10). Dose adjustments in these medications must be reported to the Investigator and recorded in the eCRF.

6.8.2 Rescue Medicine

There is no rescue medication planned for this study.

If the participant is presenting a failure of expected pharmacological action and the disease progression or signs or symptoms of disease progression are more severe than expected for the participant's condition under treatment, will be left to the Investigator's discretion whether the participant should be discontinued from the clinical trial and go back to his or her former treatment.

In the presence of clinically significant changes or biochemical modifications of the Lyso-Gb3 biomarker, each investigator must distinguish if the observed changes are still related to the natural history of disease or to an eventual lack of efficacy. In such case, the investigator will decide whether it is pertinent to withdraw the patient from the study

7.0 DISCONTINUATION OF STUDY INTERVENTION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

Discontinuation of specific study sites or of the study as a whole are detailed in Section 10.3.11.

7.1 Discontinuation of Study Intervention

It may be necessary for a participant to permanently discontinue study intervention. If study intervention is permanently discontinued, the participant will remain in the study but instead of following the sequential procedures established in the SoA, he/she will only receive telephone follow-up, if he/she agrees, at the same planned timepoints to evaluate his or her general condition. See the SoA in Section 1.3 for data to be collected at the time of discontinuation of study intervention and follow-up and for any further evaluations that need to be completed.

Participants will discontinue study intervention if any of the following occur:

- Eligibility criteria violation was noted after the participant started study treatment. In that case, the Sponsor, or Sponsor designee, must be contacted. An exception may be granted in rare circumstances for which there is a compelling safety reason to allow the participant to continue with study intervention. In these rare cases, the Investigator must obtain documented approval from the Sponsor, or Sponsor designee, to allow the participant to continue with study intervention.
- Participant is significantly noncompliant with study procedures which would put the participant at risk for continued participation in the study.
- Any AE, laboratory abnormality, or intercurrent illness which, in the judgment of the Investigator, presents a substantial clinical risk to the participant with continued study treatment.
- Clinically significant changes or biochemical modifications of the Lyso-Gb3 biomarker, that, at the Investigator's discretion, represent a lack of efficacy of the investigational product.

Participants who discontinue study intervention might be replaced.

7.1.1 Temporary Discontinuation

Temporary discontinuations (ie, dose delays or interruptions) are not allowed in this study unless in case of clinical signs of infection (coronavirus disease 2019 [COVID-19] or not), in which the participant must wait for clinical recovery (ie, 48 hours being afebrile) to receive the infusions. If clinical recovery is not established when an infusion is scheduled, the dose must be delayed, and the infusion delay and its reason must be recorded in the eCRF.

7.2 Participant Discontinuation/Withdrawal from the Study

A participant may withdraw from the study at any time at his/her own request or may be withdrawn at any time at the discretion of the Investigator for safety, lack of efficacy, behavioral, noncompliance, or administrative reasons. This is expected to be uncommon.

At the time of discontinuing from the study, if possible, an early discontinuation visit should be conducted, as shown in the SoA (Section 1.3). See SoA for data to be collected at the time of study discontinuation and follow-up and for any further evaluations that need to be completed. The participant will be permanently discontinued from the study intervention and the study at that time.

Participants who become pregnant during the study will be discontinued from the study and will be followed as described in Section 8.4.5.

If the participant withdraws consent for disclosure of future information, the Sponsor may retain and continue to use any data collected before such a withdrawal of consent.

If a participant withdraws from the study, he/she may request destruction of any samples taken and not tested, and the Investigator must document this in the study site study records.

Should a participant request or decide to withdraw from the study, all efforts must be made to complete and report the observations as thoroughly as possible up to the date of withdrawal. Participants withdrawing due to an AE should be followed up as provided in Section 8.4.3.

Participants who voluntarily withdraw are termed dropouts. Dropouts and participants withdrawn due to protocol violations may be replaced following discussion with the Investigator and Sponsor.

Participants withdrawn due to an AE might be replaced. Data collected from the replaced participants will be used.

7.3 Lost to Follow-up

A participant will be considered lost to follow-up if he/she repeatedly fails to return for scheduled visits and is unable to be contacted by the study site.

The following actions must be taken if a participant fails to return to the clinic for a required study visit or appointment:

- The study site must attempt to contact the participant and reschedule the missed visit/appointment as soon as possible, counsel the participant on the importance of maintaining the assigned visit schedule and ascertain whether the participant wishes to and/or should continue in the study.

- Before a participant is deemed lost to follow-up, the Investigator or designee must make every effort to regain contact with the participant (where possible, 3 telephone calls, message texts, and if necessary, a certified letter to the participant's last known mailing address or local equivalent methods). These contact attempts should be documented in the participant's medical record.
- Should the participant continue to be unreachable, he/she will be considered to have withdrawn from the study.
- Study site personnel, or an independent third party, will attempt to collect the vital status of the participant within legal and ethical boundaries for all participants randomized, including those who did not get study intervention. Public sources may be searched for vital status information. If vital status is determined as deceased, this will be documented and the participant will not be considered lost to follow-up. Sponsor personnel will not be involved in any attempts to collect vital status information.

8.0 STUDY ASSESSMENTS AND PROCEDURES

Study procedures and their timing are summarized in the SoA (Section 1.3). Protocol waivers or exemptions are not allowed.

Immediate safety concerns should be discussed with the Sponsor immediately upon occurrence or awareness to determine if the participant should continue or discontinue study intervention.

Adherence to the study design requirements, including those specified in the SoA (Section 1.3), is essential and required for study conduct.

All screening evaluations must be completed and reviewed to confirm that potential participants meet all eligibility criteria. The Investigator will maintain a screening log to record details of all participants screened and to confirm eligibility or record reasons for screening failure, as applicable.

Procedures conducted as part of the participant's routine clinical management (eg, blood count) and obtained before signing of the ICF may be utilized for screening purposes provided the procedures met the protocol-specified criteria and were performed within the timeframe defined in the SoA (Section 1.3).

The maximum amount of blood collected from each participant over the duration of the study, including any extra assessments that may be required, will not exceed 240 mL.

Repeat or unscheduled samples may be taken for safety reasons or for technical issues with the samples.

The order of each procedure will be determined by the study site. However, when multiple procedures are scheduled at the same day, they all need to be performed before the infusion takes place.

8.1 General Assessments

8.1.1 Inclusion and Exclusion Criteria

All inclusion and exclusion criteria will be reviewed by the Investigator or qualified designee at Screening visit.

For participants whose blood and urine samples are collected for laboratory tests at the Screening visit, the inclusion and exclusion criteria must be reassessed as soon as the results are available, prior to the first Fabrazyme® infusion.

At Baseline visit, all inclusion and exclusion criteria will be reviewed to ensure that the participant qualifies for the study, especially the inclusion criterion 10 where the 2

Fabrazyme® infusions administrated on the Lead-in period will be included in the time calculation for the assessment of dose stability.

8.1.2 Assignment of an Identification Number

All participants who consent will be given a unique identification number that will be used to identify the participant for all procedures that occur during the study. Each participant will be assigned only 1 identification number. Identification numbers must not be reused for different participants.

8.1.3 Demographics

Demographics and baseline characteristics will be evaluated at Screening visit and include age, gender, race, ethnicity (Hispanic or Latin origin), body weight, height, BMI, and general medical history.

8.1.4 Medical History

A complete medical history, including history of endocrine, neurological, cardiovascular, renal, pulmonary, immunologic and dermatology disorders, along with a history of illicit drugs, alcohol, and tobacco use will be obtained by the Investigator or qualified designee. Special attention should be given to the medical-related inclusion and exclusion criteria (Section 5.1 and 5.2).

The documented confirmed diagnosis of FD, if available, will be reviewed by the Investigator or qualified designee for all participants and recorded in the eCRF.

8.1.5 Phone Calls or Text Messages

The study site personnel will contact the participant by phone call or text message in 2 situations:

- For all participants, 24 to 48 hours before each study appointment or visit to check on symptoms compatible with COVID-19 or other infection. If suspected or positive infection, to receive the infusions the participants must wait for clinical recovery, ie, 48 hours being afebrile. Refer to Section 7.1.1.
- From the fifth infusion of AGA BETA BS onwards, within 24 hours after infusion to check the patient's clinical condition.

All contact attempts, whether calls or text messages, must be recorded in the source documents.

8.2 Efficacy Assessments

The following assessments will be made at the time points indicated in the SoA (see Section 1.3).

8.2.1 Serum Lyso-Gb3 Marker

Serum Lyso-Gb3 levels will be assessed by a central laboratory using standardized and validated methods in addition to applicable calibrated equipment.

8.2.2 Brief Pain Inventory-Short Form

Pain severity and impact of pain on daily functions will be assessed by Brief Pain Inventory (BPI)-short form questionnaire.

The BPI-short form is a self-report measure that has, over time, become a standard for the assessment of pain and its impact. The BPI-short form uses a 24-hour recall period for 9 questions:

- Questions number 1 and 2 are about existence and location of pain.
- Questions number 3 to 6 correspond to pain severity items (pain rated on 0 - 10 scales at its “worst”, “least”, “average”, and “now”).
- Questions number 7 and 8 are about medications and amount of pain relief.
- Question 9 is divided into 7 others (from A to G), which correspond to pain interference items (rated on 0 - 10 scales how much pain has interfered with general activity, walking ability, normal work, mood, enjoyment of life, relations with others, and sleep).

Despite the participant answering the questionnaire in full, the questions 1, 2, 7, and 8 will not be evaluated as an endpoint in this trial.

Sufficient copies of the questionnaire in the Spanish language will be provided to the study sites by the Sponsor. Participants should be instructed to complete the entire questionnaire at the site, when applicable. The Investigator must present the questionnaire to the participant, explain its purpose, give the participant time to read the questionnaire, and ensure that the participant understands how to respond. Immediately after filled, the questionnaire must then be reviewed by study site personnel to ensure that all questions have been properly answered. If partial completion is detected, study site personnel must return the same questionnaire to the participant to have it fully completed.

8.2.3 36-Item Short Form Health Survey

The 36-Item Short Form Health Survey (SF-36) is an often-used, well-researched, self-reported measure of a person's quality of life. It comprises 36 questions which cover 8 domains of health:

1. Limitations in physical activities because of health problems
2. Limitations in social activities because of physical or emotional problems
3. Limitations in usual role activities because of physical health problems
4. Bodily pain

5. General mental health (psychological distress and well-being)
6. Limitations in usual role activities because of emotional problems
7. Vitality (energy and fatigue)
8. General health perceptions

Sufficient copies of the questionnaire in the Spanish language will be provided to the study sites by the Sponsor. Participants should be instructed to complete the entire questionnaire at the site, when applicable. The Investigator must present the questionnaire to the participant, explain its purpose, give the participant time to read the questionnaire, and ensure that the participant understands how to respond. Immediately after filled, the questionnaire must then be reviewed by study site personnel to ensure that all questions have been properly answered. If partial completion is detected, study site personnel must return the same questionnaire to the participant to have it fully completed.

8.3 Safety Assessments

8.3.1 Physical Examinations

Physical examinations will be performed at the time points indicated in the SoA (see Section 1.3). In addition, a symptom-directed physical examination can be performed at any time as deemed necessary by the Investigator.

A complete physical examination will include, at a minimum, assessments of the cardiovascular, respiratory, gastrointestinal, and neurological systems. Height will be measured and recorded at Screening visit only. Weight will be measured and recorded at each visit and infusion appointment.

Investigators should pay special attention to clinical signs related to previous serious illnesses. Physical examination abnormalities noted at the Screening visit prior to the first dose of study intervention should be recorded in the participant's medical history. Any significant physical examination findings after the first dose of study intervention will be recorded as AEs.

8.3.2 Vital Signs

Vital signs measurements will be performed at time points indicated in the SoA (see Section 1.3).

Body temperature, heart rate, respiratory rate, and blood pressure (systolic and diastolic) will be assessed.

For all measurements including before and hourly during the infusions, blood pressure, heart rate, and respiratory rate will be assessed in a sitting position after 5 minutes rest for the participant in a quiet setting without distractions (eg, television, cell phones), with a device previously calibrated or as determined by the manufacturer.

Three readings of blood pressure and heart rate will be taken. The first reading should be rejected. The second and third readings should be averaged to give the measurement to be recorded.

8.3.3 Electrocardiograms

Single 12-lead electrocardiogram (ECG) will be obtained as outlined in the SoA (see Section 1.3) using an ECG machine that automatically calculates the heart rate and measures PR, QRS, QT, and QTc intervals. In case there are no automatic electrocardiographs in the study center, calculations may be performed manually. The ECG will be performed as part of routine cardiac safety monitoring of participants.

ECGs will be acquired after the participant has been in either the supine or semi supine position (the same position will be consistently used during ECG collections) for at least 5 minutes. Participants will be instructed to remain completely stationary (no talking, laughing, deep breathing, sleeping, or swallowing) during the ECG recording. While ECGs are being acquired, electrical devices (such as cellular telephones, fans, heaters, etc) that emit electrical interference in the room must be turned off or removed from the room to the extent that is possible.

This exam will be taken at the study sites. The original ECG traces and variables must be stored in the participants' medical records as source data. The Investigator or designee will evaluate the ECG from a clinical perspective and the result (whether the ECG result is normal or abnormal) will be recorded on the appropriate section of the eCRF and on the ECG trace signed and dated by Investigator or designee. If the result is abnormal, the Investigator or designee will provide further information (eg, sinus bradycardia, arrhythmia).

8.3.4 Echocardiograms

Two-dimensional transthoracic echocardiogram will be obtained as outlined in the SoA (see Section 1.3), to estimate the thickness of the left ventricular wall, left ventricular mass, and left ventricular ejection fraction. The echocardiogram will be performed as part of routine cardiac safety monitoring of participants.

This exam will be taken and evaluated at the study sites, if available, or at a local diagnostic laboratory by an appropriately trained physician, preferably a cardiologist.

In case the echocardiogram be performed outside of study site, the exam will have a window of ±2 days to be performed, except for the test scheduled for the Screening visit that only a +2 days will be allowed. For example, for a participant with a 3-month study visit scheduled for Day 100, the echocardiogram needs to be performed from Day 98 to Day 102 (even if the visit timepoint is Day 99 (±2)).

The Investigator or designee will evaluate the echocardiogram from a clinical perspective and the result (whether the result is normal or abnormal) will be recorded on the appropriate section of the eCRF. If the result is abnormal, the Investigator or designee will provide further information. The original echocardiographic report must be stored in the participants' medical records as source data.

8.3.5 Clinical Safety Laboratory Tests

Clinical laboratory assessments will be performed at time points indicated in the SoA (Section 1.3). Section 10.4 presents the list of clinical laboratory tests to be performed.

Local laboratories will be utilized to process and provide results for clinical laboratory tests. The Investigator or designee must review the laboratory report, document this review, and record any clinically significant changes occurring during the study as an AE. The laboratory reports must be filed with the source documents.

Abnormal laboratory findings associated with the underlying disease are not considered clinically significant unless judged by the Investigator to be more severe than expected for the participant's condition.

All laboratory tests with values considered clinically significantly abnormal during participation in the study should be repeated until the values return to normal or baseline or are no longer considered clinically significant by the Investigator.

If clinically significant values do not return to normal/baseline within a period of time judged reasonable by the Investigator, the etiology should be identified, and the Sponsor notified.

All protocol-required laboratory tests, as defined in Section 10.4, must be conducted in accordance with the laboratory manual and the SoA (Section 1.3).

If laboratory values from nonprotocol-specified laboratory tests performed at the institution's local laboratory require a change in participant management or are considered clinically significant by the Investigator (eg, SAE or AE or dose modification), then the results must be recorded.

8.3.6 Anti-algasidase Antibodies

Antibodies to AGA BETA BS will be evaluated in serum samples collected from all participants according to the SoA (Section 1.3), by a central laboratory. Additionally, serum samples should also be collected at the final visit from participants who discontinued study intervention or were withdrawn from the study.

Serum samples will be screened for antibodies binding to AGA BETA BS and the titer of confirmed positive samples will be reported. Other analyses may be performed to verify the stability of anti-AGA and/or further characterize the immunogenicity of AGA BETA BS.

The detection and characterization of anti-AGA will be performed using a validated assay method by a central laboratory. Samples may be stored for the maximum period of time according to local regulations following the last participant's last visit for the study at a facility selected by the Sponsor to enable further analysis of immune responses to AGA BETA BS.

8.3.7 Pregnancy Testing

Women of childbearing potential should only be included after a negative highly sensitive urine pregnancy test at Screening visit and at Baseline visit (prior to the first dose of experimental treatment).

Additional pregnancy testing should be performed every 4 weeks in WOCBP. If there is no protocol visit scheduled in that period, the urine pregnancy test will be arranged in the closest infusion appointment.

Also, pregnancy testing will be performed whenever a menstrual cycle is missed or when pregnancy is otherwise suspected.

Pregnancy testing will be performed using the test kit approved by the Sponsor and in accordance with instructions provided in its package insert.

8.4 Adverse Events, Serious Adverse Events, and Other Safety Reporting

The definitions of AEs and SAEs can be found in Section 10.5.

Adverse events will be reported by the participant (or, when appropriate, by a caregiver, surrogate, or the participant's legally authorized representative) to the Investigator or delegate, and by the Investigator or delegate to the Sponsor.

The Investigator and any qualified designees are responsible for detecting, documenting, and recording events that meet the definition of an AE or SAE and remain responsible for following up all AEs.

The method of recording, evaluating, and assessing causality of AEs and SAEs and the procedures for completing and transmitting SAE reports are provided in Section 10.5.

8.4.1 Time Period and Frequency for Collecting Adverse Events and Serious Adverse Event Information

All AEs and SAEs will be collected from the signing of the ICF until the last visit, at the time points specified in the SoA (Section 1.3).

The participant will receive a diary to record any AE that may occur during the study. The delivery of the diary to the participant and its collection for evaluation is described in the SoA (Section 1.3). The participant must be instructed to take the diary with them to each medical

visit and each infusion appointment for evaluation. All information collected in the diary must be recorded on the eCRF.

Medical occurrences that begin before the start of study intervention but after obtaining informed consent will be recorded as medical history/current medical conditions, not as AEs.

All SAEs will be recorded and reported to the Sponsor or designee immediately and under no circumstance should this exceed 24 hours of the Investigator's awareness of the event, as indicated in Section 10.5. The Investigator will submit any updated SAE data to the Sponsor or designee within 24 hours of their awareness of the updated information.

Investigators are not obligated to actively seek information on AEs or SAEs after conclusion of the study participation. However, if the Investigator learns of any SAE, including a death, at any time after a participant has been discharged from the study, and he/she considers the event/cause of death to be reasonably related to the study intervention or study participation, the Investigator must promptly notify the Sponsor or designee.

The method of recording, evaluating, and assessing causality of AE and SAE and the procedures for completing and transmitting safety reports are provided in Section 10.5.

8.4.2 Method of Detecting AEs and SAEs

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and nonleading verbal questioning of the participant is the preferred method to inquire about AE occurrences.

8.4.3 Follow-up of AEs and SAEs

After the initial AE/SAE report, the Investigator is required to proactively follow each participant at subsequent visits/contacts. All AEs and SAEs will be followed until resolution, stabilization, the event is otherwise explained, or the participant is lost to follow-up (as defined in Section 7.3). Further information on follow-up procedures is provided in Section 10.5.

8.4.4 Regulatory Reporting Requirements for SAEs

Prompt notification by the Investigator to the Sponsor of an SAE is essential so that legal obligations and ethical responsibilities toward the safety of participants and the safety of a study intervention under clinical investigation are met.

The Sponsor has a legal responsibility to notify both the local regulatory authority and other regulatory agencies about the safety of a study intervention under clinical investigation. The Sponsor will comply with country-specific regulatory requirements relating to safety reporting to the regulatory authority, Institutional Review Board (IRBs)/IECs, and Investigators.

An Investigator who receives an Investigator safety report describing an SAE or other specific safety information (eg, summary or listing of SAEs) from the Sponsor will review and then file it along with the Investigator's Brochure and will notify the IRB/IEC, if appropriate according to local requirements.

Investigator safety reports must be prepared for suspected unexpected serious adverse reactions according to local regulatory requirements and Sponsor policy and forwarded to Investigators as necessary.

8.4.5 Pregnancy

Details of all pregnancies in female participants and, if indicated, female partners of male participants will be collected after the start of study intervention and until 3 months after last dose of study intervention.

If a pregnancy is reported, the Investigator will record pregnancy information on the appropriate form and submit it to the Sponsor or designee within 24 hours of learning of the female participant or female partner of male participant (after obtaining the necessary signed informed consent from the female partner) pregnancy and should follow the procedures outlined in Section 10.5.

While pregnancy itself is not considered to be an AE or SAE, any pregnancy complication (or elective termination) for medical reasons will be reported as an AE or SAE.

If the outcome of the pregnancy meets the criteria for immediate classification as an SAE (ie, spontaneous abortion, stillbirth, neonatal death, or congenital anomaly [including that in an aborted fetus, stillbirth, or neonatal death]) the Investigator will report according to the SAE reporting procedures described in Section 10.5.

The participant/pregnant female partner will be followed to determine the outcome of the pregnancy until delivery of baby. The Investigator will collect follow-up information on the participant/pregnant female partner and the neonate, and the information will be forwarded to the Sponsor or designee. In the event of pregnancy occurring in a participant's partner during the study, written informed consent from the partner must be obtained prior to collection of any such information. The Sponsor will provide a separate consent form for this purpose.

Any poststudy pregnancy-related SAE considered reasonably related to the study intervention by the Investigator will be reported to the Sponsor as described in Section 8.4.4. While the Investigator is not obligated to actively seek this information in former study participants/pregnant female partner, he or she may learn of an SAE through spontaneous reporting.

Any female participant who becomes pregnant while participating in the study will be withdrawn from the study.

8.4.6 Infusion-associated Reactions

In the event of a suspected IAR, the infusion should be stopped, close observation should be initiated, management should be initiated at the Investigator's discretion, and institutional standard support care should also be administered.

Premedications such as nonsteroidal anti-inflammatory drugs, antipyretics, antihistamines and/or corticosteroids may be used to manage an IAR, at the Investigator's discretion. A decrease in infusion rate should also be considered upon reinitiation or subsequent infusions. If the infusion proceeds without incident, consideration may be given to increasing infusion rates in a stepwise manner and to reducing premedication.

All IAR will be recorded as an AE in the eCRF. An IAR can be serious or nonserious and will be reported as detailed in Section 10.5.

8.4.7 Adverse Events of Special Interest

Adverse events will be monitored throughout the study to identify any of interest that may indicate a trend or risk to participants. There is no adverse event of special interest defined for this study.

8.5 Pharmacokinetics

Pharmacokinetics parameters are not evaluated in this study.

8.6 Pharmacodynamics

Pharmacodynamic parameters are not evaluated in this study.

8.7 Genetics

Genetics are not evaluated in this study.

8.8 Biomarkers

Biomarkers other than the primary endpoint (serum Lyso-Gb3, Section 8.2.1) are not evaluated in this study.

8.9 Immunogenicity Assessments

Antibodies other than the safety endpoint (anti-AGA, Section 8.3.6) are not evaluated in this study.

8.10 Health Economics

Health economics parameters are not evaluated in this study.

9.0 STATISTICAL CONSIDERATIONS

The statistical analysis plan (SAP) will be finalized prior database lock and it will include a more technical and detailed description of the statistical analyses described in this section. This section is a summary of the planned statistical analyses of the most important endpoints including primary and key secondary endpoints.

9.1 Statistical Hypotheses

The null hypotheses and alternative hypotheses to be tested in the primary analysis are:

H_0: Ratio of Plasma level of Lyso-Gb3 marker at week 26 and plasma level of the marker LysoGb3 at baseline is less than 80% or greater than 125%. This means that there is a difference between AGA BETA BS (Test) and Fabrazyme® (Reference) after 6 months.

H_1: Ratio of Plasma level of Lyso-Gb3 marker at week 26 and plasma level of the marker Lyso-Gb3 at baseline is in between 80% and 125%. This means that there is no difference between AGA BETA BS (Test) and Fabrazyme® (Reference) after 6 months.

9.1.1 Multiplicity Adjustment

Not applicable.

9.2 Analysis Sets

For the purposes of analysis, the following analysis sets are defined:

Intent To Treat (ITT) set: Intent To Treat will include all participants who are part of the Lead-in period. The ITT population will be used for efficacy analyses.

Safety Set: Safety population will include all participants who receive at least one dose of study treatment (AGA BETA BS). The Safety population will be used for all safety analyses.

Per Protocol (PP) set: Per Protocol set will include all participants who satisfactorily complete the study and comply with the requirements of the protocol. It is a subset of the ITT population without any major protocol deviations. The PP population will be used for exploratory efficacy analyses. Final determinations of the PP population will be made at the data review meeting before database lock.

9.2.1 General Considerations

All data listings, summaries, and analyses will be performed under the guidance and approval of the Sponsor. The SAP will be developed and finalized before database lock and will describe the participant analysis sets to be included in the analyses, and procedures for accounting for missing, unused, and spurious data.

The analysis of complete data for the study will be performed when all the participants have either completed the study or discontinued early from the study, all data from the study are in the database, and the database is locked.

All data collected in the clinical database will be presented in participant data listings. Detailed methodology about summary and statistical analysis of the data collected in this study will be given in the SAP which will be finalized prior to the database lock.

All analyses, summaries, and listings will be performed using SAS® software (version 9.4 or higher) unless otherwise noted. All details regarding the statistical analysis and the preparation of tables, listings and figures will be described in the SAP. The following descriptive statistics will be used as applicable to summarize the study data unless otherwise specified:

- Continuous variables: number of observations sample size (n), mean, standard deviation (SD), median, minimum, and maximum.
- Categorical variables: frequencies (number) and percentages (the percentage of participant in each category relative to the total number of participants in the relevant analysis set or relative to the total number of participants in the relevant analysis set, with assessments available [where appropriate]) in each category will be the default summary presentation.

Baseline definition: Unless otherwise specified, baseline is defined as the last observed value of the parameter of interest prior to the first intake of AGA BETA BS (this includes unscheduled visits).

Any assessment collected prior to intake of AGA BETA BS will be part of Lead-in period.

9.2.2 Primary Endpoint(s) Analysis

The primary efficacy analysis will aim to demonstrate the equivalence in efficacy based on the mean serum Lyso-Gb3 marker ratio after 26 weeks of treatment, considering plasma level of the marker Lyso-Gb3 after 26 weeks divided by plasma level of the marker Lyso-Gb3 at baseline.

A paired t-test will be fit to the log-transformed change from baseline to after 26 weeks of treatment mean plasma Lyso-Gb3 marker.

Mean change from baseline of serum Lyso-Gb3 for after 26 weeks of treatment will be calculated and a 95% confidence interval (CI) for the difference will be computed. The mean change from baseline and its CI will be exponentiated to obtain the ratio of the means between after 26 weeks of treatment and baseline and its CI. Equivalence will be determined if the 95% CI for the postbaseline and baseline ratio lies completely within the range 0.80 to 1.25.

Analysis will be performed based on both ITT and PP populations. Descriptive statistics will be presented overall.

Table 6 Efficacy Analyses

Endpoint	Statistical Analysis Methods
Primary	• The primary efficacy variable will be analyzed using a paired t-test. • Mean change from baseline of serum Lyso-Gb3 after 26 weeks of treatment will be calculated and a 95% CI for the difference will be computed. • Equivalence will be declared if the 95% CI for the postbaseline and baseline ratio is entirely contained within the prespecified equivalence margin of 80% to 125%. • Analysis will be performed based on ITT and PP.
Secondary	• Mean serum Lyso-Gb3 marker ratio after 54 weeks of treatment will be analyzed in an analogous way to the primary endpoint. • For the other continuous secondary efficacy variables: the pain severity score and the pain interference score - change from baseline at each scheduled assessment will be presented in a descriptive statistics manner. • Also, the change in health perception before and after treatment with AGA BETA BS in participants with Fabry disease previously stabilized with Fabrazyme® will be measured by the SF-36 at each scheduled assessment.
Exploratory	Will be described in the SAP finalized before database lock.

Abbreviations: CI=confidence interval; Lyso-Gb3=globotriaosylsphingosine; ITT=Intent to Treat; PP=Per Protocol; SF-36=36-Item Short Form Health Survey.

9.2.3 Secondary Endpoint(s) Analysis

Mean serum Lyso-Gb3 marker ratio after 54 weeks of treatment will be analyzed in an analogous way to the primary endpoint. Analysis will be based on both ITT and PP populations.

Change from baseline in pain severity and pain interference will be assessed through BPI-short form pain severity and pain interference items scores respectively. The change from baseline value will be described after 26 weeks and 54 weeks of treatment, using the n, mean, SD, median, minimum value, and maximum value based on the ITT population.

The change in health perception before and after treatment with AGA BETA BS in participants with FD previously stabilized with Fabrazyme® will be measured by the SF-36. For this, change from baseline will be calculated and described using descriptive statistics (n, mean, SD, median, minimum value, and maximum value) at week 26 and week 54.

9.2.4 Pharmacokinetic/Pharmacodynamic Analyses

Not applicable.

9.2.5 Safety Analyses

For laboratory parameters and other safety variables, both the actual value and the change from the baseline value will be summarized at each visit using the n, mean, SD, median, minimum value, and maximum value.

Change from baseline, frequency counts and percentage as applicable after 14, 26, and 54 weeks of treatment will be summarized for laboratory parameters including hematology, clinical chemistry, blood urea nitrogen, eGFR, urine albumin-creatinine ratio, electrolytes, and phosphate, from blood and urine samples collected.

Change from baseline analyses after 26 weeks and 54 weeks of treatment will be summarized for cardiac function tests including electrocardiogram and bidimensional echocardiogram exams.

For the immunogenicity variable, anti-AGA levels from blood samples will be analyzed at baseline, after 14, 26, and 54 weeks of treatment, and will be summarized by counts and percentages.

Physical assessments, AEs and IARs will be summarized by frequency counts and percentage.

Duration of exposure to investigational product and number of injections will be summarized descriptively by treatment group.

For all safety and immunogenicity variables, no formal statistical analysis will be performed.

All safety analyses will be performed on the Safety Analysis Set.

9.2.6 Other Analyses

Not applicable.

9.3 Interim Analysis

No interim analysis is planned.

9.3.1 Sample Size Reevaluation

A sample size re-estimation is planned to be carried out when 50% of the total number of participants is reached. The objective is to recalculate the sample size according to the pooled variability (% coefficient of variation) observed so far.

9.4 Sample Size Determination

For the sample size calculation, the coefficient of variation of GL3 was estimated as 0.2, and the equivalence acceptance range was 20% (80% to 125%). Based on these values, the required sample size to detect equivalence with 95% confidence intervals, with a type I error of 5% and a power of 80%, was calculated as approximately 16 participants. Accounting for possible early termination, we therefore set a dropout rate of 20%, elevating the sample size to 20 participants.

The sample size of 16 is considered appropriate based on the prevalence of the disease under study and it is like previous clinical trial that evaluated a Fabrazyme® biosimilar drug.[16]

Nevertheless, a sample size re-estimation is planned to be carried out as described in Section 9.3.1.

10.0 SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

10.1 Appendix 1: Contraceptive and Barrier Guidance and Collection of Pregnancy Information

10.1.1 Definitions

Woman of Childbearing Potential (WOCBP)

A woman is considered fertile following menarche and until becoming postmenopausal unless permanently sterile (see below).

Women in the following categories are not considered WOCBP

1. Premenarchal
2. Premenopausal female with 1 of the following:
   a) Documented hysterectomy.
   b) Documented bilateral salpingectomy.
   c) Documented bilateral oophorectomy.

NOTE: Documentation can come from the study site personnel's: review of the participant's medical records, medical examination, or medical history interview.

3. Postmenopausal female:
   a) A postmenopausal state is defined as no menses for 12 months without an alternative medical cause. A high follicle-stimulating hormone (FSH) level in the postmenopausal range may be used to confirm a postmenopausal state in women not using hormonal contraception or hormonal replacement therapy (HRT). However, in the absence of 12 months of amenorrhea, a single FSH measurement is insufficient.
   b) Females on HRT and whose menopausal status is in doubt will be required to use 1 of the non estrogen hormonal highly effective contraception methods if they wish to continue their HRT during the study. Otherwise, they must discontinue HRT to allow confirmation of postmenopausal status before study enrollment.

10.1.2 Contraception Guidance

Male participants

- Male participants with female partners of childbearing potential are eligible to participate if they agree to **one** of the following during the study:
  - Are abstinent from penile-vaginal intercourse as their usual and preferred lifestyle (abstinent on a long-term and persistent basis) and agree to remain abstinent for the duration of the study and for 3 months after study last dose.

- Agree to use a male condom and have their partner use a contraceptive method with a failure rate of <1% per year as described in Table 7 when having penile-vaginal intercourse with a WOCBP who is not currently pregnant.

- Male participants with a pregnant or breastfeeding partner must agree to remain abstinent from penile-vaginal intercourse or use a male condom during each episode of penile penetration during the study.

Female participants

Female participants of childbearing potential are eligible to participate if they agree to use a highly effective method of contraception consistently and correctly as described in the Table 7.

Table 7 Highly Effective Contraceptive Methods

Highly Effective Contraceptive Methods That Are User Dependent[a] *Failure rate of <1% per year when used consistently and correctly.*
Combined (estrogen and progestogen containing) hormonal contraception associated with inhibition of ovulation • Oral. • Intravaginal. • Transdermal. • Injectable.
Progestogen only hormonal contraception associated with inhibition of ovulation • Oral. • Injectable. • Implantable.
Highly Effective Methods That Are User Independent[a]
Implantable progestogen only hormonal contraception associated with inhibition of ovulation • Intrauterine device. • Intrauterine hormone-releasing system. • Bilateral tubal occlusion.
Vasectomized partner *A vasectomized partner is a highly effective birth control method provided that the partner is the sole male sexual partner of the WOCBP and the absence of sperm has been confirmed. If not, an additional highly effective method of contraception should be used.*
Sexual abstinence *Sexual abstinence is considered a highly effective method only if defined as refraining from heterosexual intercourse during the entire period of risk associated with the study intervention. The reliability of sexual abstinence needs to be evaluated in relation to the duration of the study and the preferred and usual lifestyle of the participant.*
NOTES:

[a] Typical use failure rates may differ from those when used consistently and correctly. Use should be consistent with local regulations regarding the use of contraceptive methods for participants participating in clinical studies.

Pregnancy testing:

- Woman of childbearing potential should only be included after a negative highly sensitive urine pregnancy test.
- Additional pregnancy testing should be performed at times specified in the SoA (Section 1.3) and as required locally.
- Pregnancy testing will be performed whenever a menstrual cycle is missed or when pregnancy is otherwise suspected.
- Pregnancy testing will be performed using the test kit approved by the Sponsor and in accordance with instructions provided in its package insert.

Collection of pregnancy information

Male participants with partners who become pregnant

- The Investigator will attempt to collect pregnancy information on any male participant's female partner who becomes pregnant while the male participant is in this study. This applies only to male participants who receive study intervention.
- After obtaining the necessary signed informed consent from the pregnant female partner directly, the Investigator will record pregnancy information on the appropriate form and submit it to the Sponsor or designee within 24 hours of learning of the partner's pregnancy. The female partner will also be followed to determine the outcome of the pregnancy. Information on the status of the mother and child will be forwarded to the Sponsor. Generally, the follow-up will be no longer than 6 to 8 weeks following the estimated delivery date. Any termination of the pregnancy will be reported regardless of fetal status (presence or absence of anomalies) or indication for the procedure.

Female participants who become pregnant

- The Investigator will collect pregnancy information on any female participant who becomes pregnant while participating in this study. Information will be recorded on the appropriate form and submitted to the Sponsor or designee within 24 hours of learning of a participant's pregnancy. The participant will be followed to determine the outcome of the pregnancy. The Investigator will collect follow-up information on the participant and the neonate and the information will be forwarded to the Sponsor or designee. Generally, follow-up will not be required for longer than 6 to 8 weeks beyond the estimated delivery date. Any termination of pregnancy will be reported, regardless of fetal status (presence or absence of anomalies) or indication for the procedure.
- While pregnancy itself is not considered to be an AE or SAE, any pregnancy complication will be reported as an AE or SAE. If the outcome of the pregnancy meets the criteria for

immediate classification as an SAE (ie, spontaneous abortion, stillbirth, neonatal death, or congenital anomaly [including that in an aborted fetus, stillbirth, or neonatal death]), the Investigator will report according to the SAE reporting procedures described in Appendix 5 (Section 10.5).

- Any poststudy pregnancy-related SAE considered reasonably related to the study treatment by the Investigator will be reported to the Sponsor or designee as described in Section 8.4.4.While the Investigator is not obligated to actively seek this information in former participants, he or she may learn of an SAE through spontaneous reporting.
- Any female participant who becomes pregnant while participating in the study will be withdrawn from the study and be followed in accordance with country's current regulations.

10.2 Appendix 2: Reconstitution and Dose Preparation for Fabrazyme or AGA BETA BS

Instructions

- Vials are for single use only.
- Excessive agitation of this product should be avoided. Do not use filter needles during the preparation of the infusion.
- Prolonged exposure of Fabrazyme® or AGA BETA BS to the air/liquid interface, either through time or by agitation, may cause the formation of protein particles.

The following items are suggested for the reconstitution and administration of Fabrazyme® or AGA BETA BS:

- Fabrazyme® or AGA BETA BS (Vials)
- Sterile Water for Injection, USP
- 0.9% Sodium Chloride Injection, USP (Normal Saline)
- Tape
- Two syringes for reconstitution and dilution
- Two needles
- In-line low protein binding particulate filter (0.2 μm)
- Administration set with flow-regulating device or IV infusion pump and tubing
- IV kit
- Anaphylaxis kit
- Angiocatheter
- Gloves
- Alcohol wipes
- Arm board
- Medication label

Reconstitution and dilution (using aseptic technique):

1. Fabrazyme® or AGA BETA BS vials and diluent should be allowed to reach room temperature prior to reconstitution (approximately 30 minutes). The number of vials needed is based on the participant's body weight (kg) and the recommended dose of 1.0 mg/kg. Select the appropriate number of vials so that the total number of mg is equal to or greater than the participant's number of kg of body weight.
2. Reconstitution

    a) Reconstitute each **35 mg vial** of Fabrazyme® or AGA BETA BS by **slowly** injecting **7.2 mL** of Sterile Water for Injection, USP down the inside wall of each vial and not directly onto the lyophilized cake. Roll and tilt each vial gently. Do not invert, swirl, or shake the vial. Each vial will yield a 5.0 mg/mL clear, colorless solution (total extractable dose per vial is 35 mg, 7.0 mL).

b) Reconstitute each **5 mg vial** of Fabrazyme® or AGA BETA BS by **slowly** injecting **1.1 mL** of Sterile Water for Injection, USP down the inside wall of each vial and not directly onto the lyophilized cake. Roll and tilt each vial gently. Do not invert, swirl, or shake the vial. Each vial will yield a 5.0 mg/mL clear, colorless solution (total extractable dose per vial is 5 mg, 1.0 mL).

3. Visually inspect the reconstituted vials for particulate matter and discoloration. Do not use vials exhibiting particulate matter or discoloration. Report lot number for vials exhibiting particulate matter or discoloration.
4. After reconstitution, it is recommended to promptly dilute the vials. Failure to promptly dilute the vials could result in particle formation.
5. Slowly withdraw the reconstituted solution from each vial and further dilute with 0.9% Sodium Chloride Injection, USP to a **total volume based on participant weight specified in Table 8 below**. To minimize the air/liquid interface, remove the airspace within the infusion bag prior to adding the reconstituted Fabrazyme® or AGA BETA BS. Be sure to inject the reconstituted Fabrazyme® or AGA BETA BS solution directly into the sodium chloride solution. Total infusion volumes as low as 50 mL have been used in a clinical trial. Discard any vial with unused reconstituted solution.

Table 8 Minimum Total Volume According to the Participant's Weight

Participant Weight (kg)	Minimum Total Volume (mL)
≤35	50
35.1 – 70	100
70.1 – 100	250
>100	500

6. Gently invert infusion bag to mix the solution, avoiding vigorous shaking and agitation. Use immediately.
7. Fabrazyme® or AGA BETA BS should not be infused in the same intravenous line with other products.
8. The diluted solution should be filtered through an in-line low protein binding 0.2 µm filter during administration.

Reconstituted and diluted solutions should be used immediately. The products contain no preservatives. If immediate use is not possible, the diluted solution may be stored for up to 24 hours at 2 to 8°C (36 to 46°F).

10.3 Appendix 3: Regulatory, Ethical, and Study Oversight Considerations

10.3.1 Regulatory and Ethical Considerations

This study will be conducted in accordance with the protocol and with the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences international ethical guidelines
- Applicable ICH GCP guidelines
- Applicable laws and regulations

The protocol, protocol amendments, ICF, Investigator's Brochure, and other relevant documents (eg, advertisements) must be submitted to an IRB/IEC by the Investigator and reviewed and approved by the IRB/IEC before the study is initiated.

Any amendments to the protocol will require IRB/IEC approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study participants.

Protocols and any substantial amendments to the protocol will require health authority approval prior to initiation except for changes necessary to eliminate an immediate hazard to study participants.

The Investigator will be responsible for the following:

- Providing written summaries of the status of the study to the IRB/IEC annually or more frequently in accordance with the requirements, policies, and procedures established by the IRB/IEC
- Notifying the IRB/IEC of SAEs, or other significant safety findings as required by IRB/IEC procedures
- Providing oversight of the conduct of the study at the study site and adherence to requirements of 21 Code of Federal Regulations (CFR), ICH guidelines, the IRB/IEC, and all other applicable local regulations

After reading the protocol, each Investigator will sign the protocol signature page and send a copy of the signed page to the Sponsor or representative. The study will not start at any study site at which the Investigator has not signed the protocol.

10.3.2 Adequate Resources

The Investigator is responsible for supervising any individual or party to whom the Investigator delegates study-related duties and functions conducted at the study site.

If the Investigator/institution retains the services of any individual or party to perform study-related duties and functions, the Investigator/institution should ensure this individual or party is qualified to perform those study-related duties and functions and should implement procedures to ensure the integrity of the study-related duties and functions performed and any data generated.

10.3.3 Financial Disclosure

Investigators and subinvestigators will provide the Sponsor with sufficient, accurate financial information as requested to allow the Sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate regulatory authorities. Investigators are responsible for providing information on financial interests during the course of the study and after completion of the study.

10.3.4 Insurance

Sponsor will provide insurance in accordance with local guidelines and requirements as a minimum for the participants in this study, valid throughout the trial and 1 year after last participant finishes the trial. The terms of the insurance will be kept in the study files.

10.3.5 Informed Consent Process

The Investigator or his/her representative will explain the nature of the study, including the risks and benefits, to the participant or their legally authorized representative and answer all questions regarding the study.

Participants must be informed that their participation is voluntary. Participants or their legally authorized representatives will be required to sign a statement of informed consent that meets the requirements of 21 CFR 50, local regulations, ICH guidelines, privacy, and data protection requirements, where applicable, and the IRB/IEC or study site.

The medical record must include a statement that written informed consent was obtained before the participant was enrolled in the study and the date the written consent was obtained. The authorized person obtaining the informed consent must also sign the ICF.

Participants must be reconsented to the most current version of the ICF(s) during their participation in the study.

A copy of the ICF(s) must be provided to the participant or their legally authorized representative.

A participant who is rescreened is not required to sign another ICF if the rescreening occurs within 30 days from the previous ICF signature date.

10.3.6 Data Protection

- Participants will be assigned a unique identifier by the Sponsor. Any participant records or datasets that are transferred to the Sponsor will contain the identifier only; participant names or any information which would make the participant identifiable will not be transferred.
- The participant must be informed that his/her personal study-related data will be used by the Sponsor in accordance with local data protection law. The level of disclosure must also be explained to the participant who will be required to give consent for their data to be used as described in the informed consent.
- The participant must be informed that his/her medical records may be examined by Clinical Quality Assurance auditors or other authorized personnel appointed by the Sponsor, by appropriate IRB/IEC members, and by inspectors from regulatory authorities.

10.3.7 Committees Structure

This study does not have an independent safety monitoring committee.

10.3.8 Dissemination of Clinical Study Data

The results of the study should be reported within 1 year from the end of the clinical study. Irrespective of the outcome, the Sponsor will submit to any relevant database a summary of the results of the clinical study within 1 year from the end of the clinical study. It shall be accompanied by a summary written in a manner that is understandable to laypersons.

10.3.9 Data Quality Assurance

- All participant data relating to the study will be recorded on printed or eCRFs unless transmitted to the Sponsor or designee electronically (eg, laboratory data). The Investigator is responsible for verifying that data entries are accurate and correct by physically or electronically signing the eCRF.
- Guidance on completion of eCRFs will be provided in a separated document.
- The Investigator must permit study-related monitoring, audits, IRB/IEC review, and regulatory agency inspections and provide direct access to source data documents.
- Monitoring details describing strategy, including definition of study critical data items and processes (eg, risk-based initiatives in operations and quality such as risk management and mitigation strategies and analytical risk-based monitoring), methods, responsibilities, and requirements, including handling of noncompliance issues and monitoring techniques (central, remote, or on-site monitoring) are provided in the Study Monitoring Plan.
- Details of study monitoring, including possible action required due to COVID-19, will be included in the Study Monitoring Plan.

- The Sponsor or designee is responsible for the data management of this study, including quality checking of the data.
- The Sponsor assumes accountability for actions delegated to other individuals (eg, contract research organizations).
- Records and documents, including signed ICFs, pertaining to the conduct of this study must be retained by the Investigator for 10 years after study completion unless local regulations or institutional policies require a longer retention period. No records may be destroyed during the retention period without the written approval of the Sponsor. No records may be transferred to another location or party without written notification to the Sponsor.

10.3.10 Source Documents

According to the ICH E6 (R2) definition, source data is all information in original records and certified copies of original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial. Source data are contained in source documents (original records or certified copies).

The Investigator/institution should maintain adequate and accurate source documents and study records that include all pertinent observations on each of the study site's participants. Source data should be attributable, legible, contemporaneous, original, accurate, and complete. Changes to source data should be traceable, should not obscure the original entry, and should be explained if necessary (eg, via an audit trail).

Source documents provide evidence for the existence of the participant and substantiate the integrity of the data collected. Source documents are filed at the Investigator's site.

Data reported on the eCRF that are transcribed from source documents must be consistent with the source documents or the discrepancies must be explained. The Investigator may need to request previous medical records or transfer records, depending on the study. Also, current medical records must be available.

The Investigator must maintain accurate documentation (source data) that supports the information entered in the eCRF.

Study monitors will perform ongoing source data verification to confirm that data entered into the eCRF by authorized study site personnel are accurate, complete, and verifiable from source documents; that the safety and rights of participants are being protected; and that the study is being conducted in accordance with the currently approved protocol and any other study agreements, ICH GCP, and all applicable regulatory requirements.

10.3.11 Study and Site Start and Closure

First Act of Recruitment

The study start date is the date on which the clinical study will be open for recruitment of participants.

The first act of recruitment is the first study site open and will be the study start date.

Definitions of a subject's status are as follows:

- Entered: a subject who has signed the ICF to be screened.
- Enrolled: a subject who has met all eligibility criteria.

Study/Site Termination

The Sponsor or designee reserves the right to close the study site or terminate the study at any time for any reason at the sole discretion of the Sponsor. Study sites will be closed upon study completion. A study site is considered closed when all required documents and study supplies have been collected and a study site closure visit has been performed.

The Investigator may initiate study site closure at any time, provided there is reasonable cause and sufficient notice is given in advance of the intended termination.

Reasons for the early closure of a study site by the Sponsor or Investigator may include but are not limited to:

For study termination:

- Discontinuation of further study intervention development.

For study site termination:

- Failure of the Investigator to comply with the protocol, the requirements of the IRB/IEC or local health authorities, the Sponsor's procedures, or GCP guidelines
- Inadequate or no recruitment (evaluated after a reasonable amount of time) of participants by the Investigator
- Total number of participants included earlier than expected.

If the study is prematurely terminated or suspended, the Sponsor shall promptly inform the Investigators, the IECs/IRBs, the regulatory authorities, and any contract research organization(s) used in the study of the reason for termination or suspension, as specified by the applicable regulatory requirements. The Investigator shall promptly inform the participant and should assure appropriate participant therapy and/or follow-up.

10.3.12 Publication Policy

The data generated by this study are confidential information of the Sponsor. The Sponsor will make the results of the study publicly available. The publication policy with respect to the Investigator and study site will be set forth in the Clinical Trial Agreement.

The results of this study may be published or presented at scientific meetings. If this is foreseen, the Investigator agrees to submit all manuscripts or abstracts to the Sponsor before submission. This allows the Sponsor to protect proprietary information and to provide comments.

The Sponsor will comply with the requirements for publication of study results. In accordance with standard editorial and ethical practice, the Sponsor will generally support publication of multicenter studies only in their entirety and not as individual study site data. In this case, a coordinating Investigator will be designated by mutual agreement.

Authorship will be determined by mutual agreement and in line with International Committee of Medical Journal Editors authorship requirements.

10.4 Appendix 4: Clinical Laboratory Tests

The tests detailed in Table 9 will be performed by the local laboratory and the tests detailed in Table 10 will be performed by a central laboratory. For all tests, participant's preparation should follow each laboratory's standard procedure.

Protocol-specific requirements for inclusion or exclusion of participants are detailed in Section 5.0 of the protocol.

Additional tests may be performed at any time during the study as determined necessary by the Investigator or required by local regulations.

Investigators must document their review of each laboratory safety report.

Table 9 Protocol-required Safety Laboratory Tests – Local Laboratory

Laboratory Assessments	Parameters	
Hematology	Complete hemogram:	%Reticulocytes
	Platelet count	White blood cell count with differential:
	Red blood cell count	Neutrophils
	Hemoglobin	Lymphocytes
	Hematocrit	Monocytes
	Red blood cell indices:	Eosinophils
	Mean corpuscular volume	Basophils
	Mean corpuscular hemoglobin	
	Mean cell hemoglobin concentration	
Clinical Chemistry	Blood urea nitrogen	Electrolytes:
	Phosphate	Sodium
	Creatinine[a]	Potassium
	Total cholesterol	Chlorine
	Cholesterol LDL	Bicarbonate
	Cholesterol HDL	Magnesium
	Triglycerides	Calcium
	Glycemia	
	Total bilirubin	Alkaline phosphatase
	Direct bilirubin	Total proteins
	Aspartate aminotransferase	Albumin
	Alanine aminotransferase	Gamma glutamyl transferase
Urinalysis	First morning urine[b]	Urine albumin-creatinine ratio
Other tests	Highly sensitive urine human chorionic gonadotropin pregnancy test (as needed for WOCBP)[c].	

NOTES:
The results of each test must be entered into the eCRF.

[a] The eGFR should be calculated using CKD-EPI equation.

[b] This exam should include visual exam, dipstick test and microscopic exam (color, clarity, pH, specific gravity, glycose, blood, ketones, protein, urobilinogen, bilirubin, leukocyte esterase, nitrite, blood, white blood cells, red blood cells, epithelial cells, crystals, and casts).

[c] Local urine testing will be standard for the protocol unless serum testing is required by local regulation or IRB/IEC.

eCRF=electronic case report form; IEC=Independent Ethics Committee; IRB=Institutional Review Board; WOCBP=women of childbearing potential.

Table 10 Protocol-required Safety Laboratory Tests – Central Laboratory

Laboratory Assessments	Parameters
Biomarker	Serum Lyso-Gb3
Immunology	Anti-AGA

Anti-AGA=anti-agalsidase antibodies; Lyso-Gb3=globotriaosylsphingosine.

10.5 Appendix 5: AEs and SAEs: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting for Study Intervention

10.5.1 Definition of Adverse Event

AE Definition
• An AE is any untoward medical occurrence in a clinical study participant administered a medicinal product and which does not necessarily have a causal relationship with that product. • NOTE: An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease (new or exacerbated) temporally associated with the use of study intervention/treatment, whether or not considered related to the study intervention/treatment.

Events Meeting the AE Definition
• Any abnormal laboratory test results (hematology, clinical chemistry, or urinalysis) or other safety assessments (eg, ECG, echocardiogram, vital signs measurements), including those that worsen from baseline, considered clinically significant in the medical and scientific judgment of the Investigator (ie, not related to progression of underlying disease). • Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition. • New condition detected or diagnosed after study intervention administration even though it may have been present before the start of the study. • Signs, symptoms, or the clinical sequelae of a suspected intervention-intervention interaction. • Lack of efficacy or failure of expected pharmacological action per se will not be reported as an AE or SAE. Such instances will be captured in the efficacy assessments. However, the signs, symptoms, and/or clinical sequelae resulting from lack of efficacy will be reported as AE or SAE if they fulfill the definition of an AE or SAE.

Events <u>NOT</u> Meeting the AE Definition
• Any clinically significant abnormal laboratory findings or other abnormal safety assessments that are associated with the underlying disease, unless judged by the Investigator to be more severe than expected for the participant's condition. • The disease/disorder being studied or expected progression, signs, or symptoms of the disease/disorder being studied, unless more severe than expected for the participant's condition. • Medical or surgical procedure (eg, endoscopy, appendectomy): the condition that leads to the procedure is the AE. • An elective surgery/procedure scheduled to occur during a study will not be considered an AE if the surgery/procedure is being performed for a preexisting condition and the surgery/procedure has been preplanned prior to study entry. However, if the preexisting condition deteriorates unexpectedly during the study (eg, surgery performed earlier than planned), then the

deterioration of the condition for which the elective surgery/procedure is being done will be considered an AE.

- Anticipated day-to-day fluctuations of preexisting disease(s) or condition(s) present or detected at the start of the study that do not worsen.

10.5.2 Definition of Serious Adverse Event

An SAE is defined as any untoward medical occurrence that, at any dose, meets one or more of the criteria listed:

a. Results in death

- For SAEs with the outcome of death, the date and cause of death will be recorded on the appropriate case report form.

b. Is life-threatening

- The term *life-threatening* in the definition of *serious* refers to an event in which the participant was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.

c. Requires inpatient hospitalization or prolongation of existing hospitalization

- In general, hospitalization signifies that the participant has been admitted (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization or fulfills any other serious criteria, the event is serious. When in doubt as to whether hospitalization occurred or was necessary, the AE should be considered serious.
- Hospitalization for elective treatment of a preexisting condition that did not worsen from baseline is not considered an AE.

d. Results in persistent or significant disability/incapacity

- The term disability means a substantial disruption of a person's ability to conduct normal life functions.
- This definition is not intended to include experiences of relatively minor medical significance such as uncomplicated headache, nausea, vomiting, diarrhea, influenza, and accidental trauma (eg, sprained ankle) that may interfere with or prevent everyday life functions but do not constitute a substantial disruption.

e. Is a congenital anomaly/birth defect

- The term congenital anomaly/birth defect means there is suspect that exposure to a medical product prior to conception or during pregnancy may have resulted in an adverse outcome in the child.

f. Other situations:

- Medical or scientific judgment should be exercised by the Investigator in deciding whether SAE reporting is appropriate in other situations such as significant medical events that may jeopardize the participant or may require medical or surgical intervention to prevent one of the other outcomes listed in the above definition. These events should usually be considered serious.

- Examples of such events include invasive or malignant cancers, intensive treatment for allergic bronchospasm, blood dyscrasias, convulsions or development of intervention dependency or intervention abuse.

10.5.3 Recording and Follow-Up of Adverse Event and/or Serious Adverse Event

AE and SAE Recording

- When an AE/SAE occurs, it is the responsibility of the Investigator to review all documentation (eg, hospital progress notes, laboratory reports, and diagnostics reports) related to the event.
- The Investigator will then record all relevant AE/SAE information.
- It is **not** acceptable for the Investigator to send photocopies of the participant's medical records to the Sponsor or designee in lieu of completion of the applicable/required report form.
- There may be instances when copies of medical records for certain cases are requested by the Sponsor or designee. In this case, all participant identifiers, with the exception of the participant number, will be redacted on the copies of the medical records before submission to the Sponsor or designee.
- The Investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE.

Assessment of Intensity

The intensity of an AE is an estimate of the relative severity of the event made by the Investigator based on his or her clinical experience and familiarity with the literature. The following definitions are to be used to rate the severity of an AE:

- Mild: Asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
- Moderate: Minimal, local, or noninvasive intervention indicated; limiting age-appropriate instrumental Activities of Daily Living (ADL). Instrumental ADL refers to preparing meals, shopping for groceries or clothes, using the telephone, managing money, etc.
- Severe: Severe or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling, limiting self-care ADL. Self-care ADL refers to bathing, dressing and undressing, feeding self, using the toilet, taking medications, and not bedridden.

Assessment of Causality

- The Investigator is obligated to assess the relationship between study intervention and each occurrence of each AE/SAE. The Investigator will use clinical judgment to determine the relationship.
- A *reasonable possibility* of a relationship conveys that there are facts, evidence, and/or arguments to suggest a causal relationship, rather than a relationship cannot be ruled out.
- Alternative causes, such as underlying disease(s), concomitant therapy, and other risk factors, as well as the temporal relationship of the event to study intervention administration, will be considered and investigated.

- For causality assessments, events assessed as having a reasonable possibility of being related to study intervention will be considered "related." Events assessed as having no reasonable possibility of being related to study intervention will be considered "unrelated."
- The Investigator will also consult the Investigator's Brochure and/or product information, for marketed products, in his/her assessment.
- For each AE/SAE, the Investigator **must** document in the medical notes that he/she has reviewed the AE/SAE and has provided an assessment of causality.
- There may be situations in which an SAE has occurred and the Investigator has minimal information to include in the initial report to the Sponsor or designee. However, it is very important that the Investigator always makes an assessment of causality for every event before the initial transmission of the SAE data to the Sponsor or designee.
- The Investigator may change his/her opinion of causality in light of follow-up information and send an SAE follow-up report with the updated causality assessment.
- The causality assessment is one of the criteria used when determining regulatory reporting requirements.

Follow-up of AEs and SAEs

- The Investigator is obligated to perform or arrange for the conduct of supplemental measurements and/or evaluations as medically indicated or as requested by the Sponsor or designee to elucidate the nature and/or causality of the AE or SAE as fully as possible. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals.
- If a participant dies during participation in the study or during a recognized follow-up period, the Investigator will provide the Sponsor or designee with a copy of any postmortem findings including histopathology.
- New or updated information will be recorded in the originally submitted documents.
- The Investigator will submit any updated SAE data to the Sponsor or designee within 24 hours of the Investigator's awareness of the information.

10.5.4 Reporting of SAEs

SAE Reporting to the Sponsor or Designee via an Electronic Data Collection System

- The primary mechanism for reporting an SAE to the Sponsor or designee will be the electronic data collection system. The study site will enter the event into the electronic data collection system within 24 hours of the Investigator's awareness of the event.
- If the electronic system is unavailable, then the study site will use the paper SAE report form (see next section) to report the event and will enter the event into the electronic data collection system as soon as the system becomes available.
- After the study is completed at a given study site, the electronic data collection system will be taken offline to prevent the entry of new data or changes to existing data.
- If a study site receives a report of a new SAE from a study participant or receives updated data on a previously reported SAE after the electronic data collection system has been taken

offline, then the study site can report this information on a paper SAE report form (see next section) to the Sponsor or designee.

Contacts for SAE reporting:

- Emails: i.clinica@biosidus.com.ar, farmacovigilancia@biosidus.com.ar, and f.amato@biosidus.com.ar
- Contact telephone numbers: 0800 666 2527 or +54 11 6324 4707

SAE Reporting to the Sponsor or Designee via Paper SAE Report Form

- The primary mechanism for reporting an SAE to the Sponsor or designee will be the paper SAE report form. The study site will submit the SAE report form, via email, within 24 hours of the Investigator's awareness of the event. Facsimile transmission may be utilized as an alternative mode of submission, if necessary.
- Notification of SAE information via telephone does not replace the need for the Investigator to complete, sign and submit the paper SAE report form to the Sponsor or designee within 24 hours of the Investigator's awareness of the event.

Contacts for SAE reporting:

- Emails: i.clinica@biosidus.com.ar, farmacovigilancia@biosidus.com.ar, and f.amato@biosidus.com.ar
- Contact telephone numbers: 0800 666 2527 or +54 11 6324 4707

10.6 Appendix 6: Abbreviations and Definitions

Abbreviation	Definition
ACE	Angiotensin-converting enzyme
ADL	Activities of Daily Living
AE	Adverse event
AGA BETA BS	Agalsidase Beta Biosidus
ANMAT	The National Administration of Drugs, Foods and Medical Devices (Argentina)
Anti-AGA	Anti-agalsidase antibodies
ARBs	Angiotensin II receptor blockers
BPI-short form	Brief Pain Inventory short form
CFR	Code of Federal Regulations
CI	Confidence interval
COVID-19	Coronavirus disease 2019
ECG	Electrocardiogram
eCRF	Electronic case report form
eGFR	Estimated glomerular filtration rate
ERT	Enzyme replacement therapy
EU	European Union
FD	Fabry disease
FSH	Follicle-stimulating hormone
GCP	Good Clinical Practice
GL3 or Gb3	Globotriaocylceramide
HRT	Hormone replacement therapy
IARs	Infusion-associated reactions
ICF	Informed Consent Form
ICH	International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use
IEC	Independent Ethics Committee
IRB	Institutional Review Board
ITT	Intent To Treat
IV	Intravenous
Lyso-Gb3	Globotriaosylsphingosine

Abbreviation	Definition
PCT	Pharmacological chaperones
PD	Pharmacodynamics
PK	Pharmacokinetic
PP	Per Protocol
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SD	Standard deviation
SF-36	36-Item Short Form Health Survey
SoA	Schedule of activities
US	United States
WOCBP	Women of childbearing potential
α-Gal A	α-galactosidase A

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Signature of Investigator

PROTOCOL TITLE: Phase III, Open-label, Switch Over Trial of the Efficacy and Safety of Agalsidase Beta Biosidus (AGA BETA BS) in Fabry Disease Patients Previously Stabilized with Fabrazyme®.

PROTOCOL NO: BIO-AGA-Fase III-001

VERSION: 5.0

This protocol is a confidential communication of the Sponsor. I confirm that I have read this protocol, I understand it, and I will work according to this protocol. I will also work consistently with the ethical principles that have their origin in the Declaration of Helsinki and that are consistent with Good Clinical Practices and the applicable laws and regulations. Acceptance of this document constitutes my agreement that no unpublished information contained herein will be published or disclosed without prior written approval from the Sponsor.

Instructions to the Investigator: Please SIGN and DATE this signature page. PRINT your name, title, and the name of the study center in which the study will be conducted. Return the signed copy to Sponsor or CRO.

I have read this protocol in its entirety and agree to conduct the study accordingly:

Signature of Investigator: _____ Date: _____

Printed Name: _____

Investigator Title: _____

Name/Address of Center: _____

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