Official Title:	An Open-Label, Fixed-Sequence, Ascending-Dose, First-in-Human Study to Assess the Safety, Tolerability, Pharmacokinetics, Pharmacodynamics, and Efficacy of Intravenous Infusions of ATB200 Co-Administered with Oral AT2221 in Adult Subjects With Pompe Disease
NCT Number:	NCT02675465
Document Date:	Amendment 2: 10 January 2019

CLINICAL STUDY PROTOCOL

AN OPEN-LABEL, FIXED-SEQUENCE, ASCENDING-DOSE, FIRST-IN-HUMAN STUDY TO ASSESS THE SAFETY, TOLERABILITY, PHARMACOKINETICS, PHARMACODYNAMICS, AND EFFICACY OF INTRAVENOUS INFUSIONS OF ATB200 CO-ADMINISTERED WITH ORAL AT2221 IN ADULT SUBJECTS WITH POMPE DISEASE

Protocol Number: ATB200-02

ORIGINAL PROTOCOL: Date: 6 November 2015

AMENDMENT 01: Date: 16 December 2015

AMENDMENT 02: Date: 11 February 2016

AMENDMENT 03: Date: 19 July 2016

AMENDMENT 04: Date: 16 December 2016

AMENDMENT 05: Date: 16 February 2018

AMENDMENT 06: Date: 30 April 2018

AMENDMENT 07: Date: 03 October 2018

AMENDMENT 08: Date: 10 January 2019

EudraCT Number: 2015-004798-34
US IND Number: 127,387

Compounds: ATB200 and AT2221

Sponsor

Amicus Therapeutics, Inc.
1 Cedar Brook Drive
Cranbury, NJ 08512
Phone: 609-662-2000

Serious Adverse Event Contact Information

Role	Contact Information
Reporting of serious adverse events	**Primary method of contact:** **Safety FAX number:** + 1 732-289-6060 If fax is unsuccessful, please use: Safety Email address: saereporting_pompe@amicusrx.com
Inquiries related to serious adverse event reporting or designee	Amicus Therapeutics, Inc. 1 Cedar Brook Drive Cranbury, NJ 08512, USA [redacted], Global Drug Safety & Pharmacovigilance Work number: + 1 609-366-1164 Mobile number: [redacted]

1. DECLARATIONS OF SPONSOR AND INVESTIGATOR

1.1. Declaration of Sponsor

This clinical study protocol is subject to critical review and has been approved by Amicus Therapeutics, Inc.

The information it contains is consistent with:

- The current benefit-risk evaluation of ATB200 co-administered with AT2221
- The moral, ethical, and scientific principles governing clinical research, as set out in the current version of Declaration of Helsinki and the principles of Good Clinical Practice (GCP) described in the United States Code of Federal Regulations (US CFR) Parts 50, 54, 56, and 312 and in the International Conference on Harmonisation (ICH) GCP E6 guidelines

The investigator will be supplied with details of any significant or new findings related to treatment with ATB200 co-administered with A[redacted]

Date: 11 Jan 2019 Signature: [redacted]

[redacted], Clinical Research
Amicus Therapeutics, Inc.

1.2. Declaration of Investigator

I confirm that I have read this clinical study protocol. I understand it, and I will work according to the moral, ethical, and scientific principles governing clinical research, as set out in the current version of Declaration of Helsinki and the principles of GCP described in the US CFR Parts 50, 54, 56, and 312 and in the ICH GCP E6 guidelines. I will also work in accordance with applicable local requirements.

Investigator

Date: _____ Signature: _____

Printed Name: _____

2. SUMMARY OF CHANGES TO THE PROTOCOL

Protocol Number	ATB200-02
Protocol Title	An Open-Label, Fixed-Sequence, Ascending-Dose, First-in-Human Study to Assess the Safety, Tolerability, Pharmacokinetics, Pharmacodynamics, and Efficacy of Intravenous Infusions of ATB200 Co-administered with Oral AT2221 in Adult Subjects with Pompe Disease

The previous version of this protocol (Amendment 7.0, 03 October 2018) was amended to create the current version (Amendment 8.0, 10 January 2019). For definitions of abbreviations used, see Section 5.

Protocol History	
Version and Date of Protocol	**Comments**
Original Protocol, 06 November 2015	Original version
Amendment 1.0, 16 December 2015	Addition of the following text: Subjects will be monitored for at least 48 hours from the start of each infusion during Stage 1 (ie, ERT dose-escalation stage). Since the target infusion duration in the study is 4 hours, all subjects will be closely monitored for the first 4 hours during their infusion, and then observed in hospital for an additional 44 hours prior to discharge. Subjects will be admitted and dosed at either an infusion center or a hospital and thereafter hospitalized for the remainder of the observation period. In Stage 2, when subjects have reached their maximum per protocol ATB200 dose, all subjects will be monitored for at least an additional 2 to 4 hours from the end of their infusion.
Amendment 2.0, 11 February 2016	The protocol was amended to include changes relevant for the: • Addition of a 24-month, open-label extension phase, making this a study conducted in 3 stages, with a total duration of 2 years and 18 weeks • Roll-over enrollment for subjects who complete Stages 1 and 2 (Cohort 1) to the extension stage (Stage 3)

Protocol History	
Version and Date of Protocol	**Comments**
Amendment 2.0, 11 February 2016 (Continued)	• Introduction of new cohorts into the study – Cohort 1 subjects; defined as adults with Pompe disease who have been on ERT for 2 to 6 years prior to enrollment and are able to walk at least 200 meters in the 6MWT – Cohort 2 subjects; defined as adults with Pompe disease who are wheelchair bound, unable to walk unassisted, and have been on ERT (alglucosidase alfa) for ≥ 2 years prior to enrollment – Cohort 3 subjects; defined as adults with Pompe disease who have never received treatment with ERT and are able to walk at least 200 meters in the 6MWT • Enrollment of 4 to 6 ERT-experienced nonambulatory subjects (Cohort 2) and 4 to 6 ERT-naïve subjects (Cohort 3) in Stage 3, who will receive treatment with 20 mg/kg ATB200 co-administered with 260 mg AT2221 • Separate enrollment criteria specified for Cohorts 2 and 3 • Sentinel dosing specified for Cohorts 2 and 3 • Additional analyses planned for Cohorts 2 and 3 • Additional objectives for: – Assessment of long-term efficacy and safety – PK parameters for plasma rhGAA activity and total protein and plasma AT2221 in ERT-naïve subjects • Additional study assessments extending through the end of Stage 3 for all cohorts, including functional assessments to be performed every 6 months • Change made to the values for $AUC_{0-\infty}$ in predicted exposure for single dose administration of AT2221 in humans; corrected to 130 mg dose, 12.35 µg·hr/L and 260 mg dose, 24.71 µg·hr/L (see Section 6.4.2, Table 4) • Addition of a total GAA protein concentration assessment when a blood sample for IgE is collected in all stages for all cohorts

Protocol History	
Version and Date of Protocol	**Comments**
Amendment 3.0, 19 July 2016	The protocol was amended to include typographical and administrative changes to improve the clarity of the document. As well as changes relevant to: • Removal of Table 1, information supplied in a separate document. • Clarifications to Inclusion Criterion 5, 6, 14, 15. • Clarifications to Exclusion Criterion 1, 9, 11, 12, 13, 19, 21, 22, 23, 31, 32. • Clarification to prohibited medications: – Myozyme/Lumizyme after treatment with ATB200 has begun. – β_2-receptor agonists and non-selective β-blockers (eg, propranolol, nadalol and carvedilol). • Clarification to Interim Analysis of up to 4 interim analysis will be performed in the study. • Acceptable methods of contraception for male and female subjects enrolled in the study now include abstinence and the use of a diaphragm combined with a condom. • Schedule of Assessments (Table 9, Table 10, Table 11, and Table 12) was updated with various changes to certain visits due to redundancy of assessments. • Information previously only contained in the Schedule of Assessments table footnotes was also added to the text in Section 10.2. • Visit windows were added toTable 10, Table 11, and Table 12. • Vital signs during PK visits have been adjusted to: Hours 0, 1, 4, 6, 12, and 24. • PK blood sampling Table 13 has been adjusted.

Protocol History	
Version and Date of Protocol	**Comments**
Amendment 3.0, 19 July 2016 (Continued)	• The following text was added to Section 10.1.10 and Section 10.2.12: Upon study completion or subject discontinuation, subjects who are confirmed positive for anti-rhGAA antibodies will complete follow-up immunological testing for up to 6 months (ie, at 1-, 3-, and 6-months follow-up). If anti-rhGAA antibodies are not confirmed positive at follow-up time points earlier than 6 months, no further testing is required. • Addition of Section 10.4, definition of high sustained antibody titer. • Addition of the following text in Section 11.1.1: Subjects experiencing adverse events should be followed until their health has returned to baseline status or stabilized or have otherwise been explained. • Addition of Appendix 7 to include the PGIC and SGIC scales.
Amendment 04, 16 December 2016	The protocol was amended to include the following changes: • Collection of available historical antibody information. • Addition of patient reported outcomes and functional assessments at the End of PK visit for Cohort 1 subjects. • Addition of functional assessments at Months 15 and 21 of Stage 3 for Cohort 1 and to Months 3, 9, 15, and 21 for Cohorts 2 and 3, which will change the frequency of these assessments to every 3 months for the duration of Stage 3 for all cohorts. • Addition of Patient Reported Outcomes at Months 15 and 21 of Stage 3 for Cohort 1 and Months 3, 9, 15, and 21 for Cohorts 2 and 3, which will change the frequency of these assessments to every 3 months for the duration of Stage 3 for all cohorts.

Protocol History	
Version and Date of Protocol	**Comments**
Amendment 04, 16 December 2016 (Continued)	• Addition of Global Impression of Change assessments at Months 6, 12, 15, 18, and 24 of Stage 3 for Cohort 1; Months 3, 9, 15, 18, and 21 for Cohorts 2 and 3, which will change the frequency of these assessments to every 3 months for the duration of Stage 3 for all cohorts. • Elimination of the requirement for a 6-hour fast prior to clinical laboratory testing. • Elimination of abstinence as an allowable form of birth control. • Correction of statistical comparisons for PK data. • Clarification of non-assessment visits (ie, Infusion Only visits). • Clarification of testing procedures at unscheduled visits. • Subject's Global Impression of Change form has been revised (Appendix 7). • Other clarification to improve the readability and understanding of the document.
Amendment 05, 16 February 2018	The protocol was amended to include changes relevant to the: • Implementation of Home Infusion: ERT infusion, for eligible subjects who so desire, at the subject's residence, outside the hospital or clinic setting, may be considered for subjects, where allowed by Principal Investigator, regulatory authorities and/or local ethics committees • Introduction of a new cohort into the study – Additional subjects will be recruited/enrolled to Cohort 4; defined as adults with Pompe disease who have been on ERT for ≥ 7 years and are able to walk at least 200 meters in the 6MWT. Subjects are assigned to Multiple Doses Co-administration of 20 mg/kg ATB200 + 260 mg AT2221 as their study treatment

Protocol History	
Version and Date of Protocol	**Comments**
Amendment 05, 16 February 2018 (Continued)	• Introduction of a new stage – Stage 4; defined Stage 4, treatment period will begin at the end of Stage 3 and will continue as open-label extension until commercialization, study discontinuation or subject withdrawal, with functional assessments every 6 months • Clarification of Study Design: Stage 3 • Addition of rationale for Retrospective Data collection prior to Baseline • Updated protocol with preliminary safety PK efficacy data • Clarification: Changes made to PK sample collection and analysis for Cohorts 1 and 3 during the study • Addition of sparse blood sampling for plasma total GAA protein concentrations from Cohorts 1 and 3 patients • Addition of SAE contact information as page 2 of the protocol • Post-infusion monitoring time changed • Addition of follow-up safety visit assessment • Clarification: PFTs and/or assessments will be performed at the Baseline Visit and every 3 months in Stage 3 for all ambulatory subjects and for nonambulatory subjects without invasive ventilatory support • Clarification: Reporting under dosing during the study • Revision: interim analyses will be performed during the study as needed • Revision: number of subjects (approximately 21-32 planned) • Revision: statistical methods for non-PK-specific parameters section • Addition of retrospective data collection • Table 9 (Treatment Assignment for Stages 1, 2, 3, and 4) was updated

Protocol History	
Version and Date of Protocol	**Comments**
Amendment 05, 16 February 2018 (Continued)	• Schedules of Assessments (Table 9, Table 10, Table 11, and Table 12) were updated with various changes to certain visits due to Stage 4 assessments • Visit windows were added to Table 9, Table 10, and Table 12 • New abbreviated terms and correction of omissions • Typographical and administrative changes were also made to improve the readability and understanding of the document
Amendment 06, 30 April 2018	The protocol was amended to include changes relevant to the: • Clarification of the description, reporting, and management of infusion-associated reactions (IARs) • Updates to the acceptable methods of contraception for male and female subjects enrolled in the study. Removed use of an IUD alone as an acceptable method. • Removal of blood sample collection for measurement of pro-inflammatory cytokines and/or other biomarkers of immune system activation for Stage 4 (all Cohorts) and for Stage 3, Cohort 4 • Administrative changes to improve the readability and understanding of the document
Amendment 07, 03 October 2018	The protocol was amended to include changes relevant to the: • Increase in number of subjects in Cohort 4 to 'approximately 10 subjects' • Deletion of "with alglucosidase alfa (Myozyme/Lumizyme)" in inclusion criterion #29 for Cohort 4 • Increase in upper limit for the 6MWT to 600 meters and decrease in lower limit for the 6MWT to 75 meters in inclusion criterion #32 for Cohort 4 • Increase in upper limit of upright FVC to 85% in inclusion criterion #33 for Cohort 4

Protocol History	
Version and Date of Protocol	**Comments**
Amendment 07, 03 October 2018 (Continued)	• Addition of a UK-specific section to update the acceptable methods of contraception for male and female subjects enrolled in the study to remove the use of double barrier methods as highly effective methods of contraception • Addition of PROMIS® instruments to the Patient-reported Outcomes • Addition of activity monitoring to the exploratory assessments for Cohort 4 in Stage 3 • Creation of a separate Schedule of Assessments for Cohort 4 • Increase in sample size from approximately 18 to 24 subjects to approximately 18 to 34 subjects • Update to Section 15: Study Conduct Considerations
Amendment 08 10 January 2019	The protocol was amended to include changes relevant to the: • Decrease in number of subjects for Cohort 4 from approximately 10 subjects to 6 to 8 subjects • Update to eligibility for home infusion from 12 months to 6 months • For subjects who are confirmed to have a positive result for anti-rhGAA antibodies upon study completion or discontinuation, update to follow-up immunological testing from 6 months to 12 months after the last dose of study drug or until they begin treatment with another ERT or investigational therapy • Removal of activity monitoring, actigraph from Cohort 4 assessments • Update to Table 6 and Table 7 to include data through Month 18 • Update to Table 10 and Table 12 to reflect that height measurement is taken only at screening • Addition of Section 8.8: Criteria for Termination of the Study

3. SYNOPSIS

Name of Sponsor/Company: Amicus Therapeutics, Inc.

Names of Investigational Products: ATB200 and AT2221

Names of Active Ingredients: Recombinant human acid α-glucosidase (rhGAA) and miglustat

Title of Study: An Open-Label, Fixed-Sequence, Ascending-Dose, First-in-Human Study to Assess the Safety, Tolerability, Pharmacokinetics, Pharmacodynamics, and Efficacy of Intravenous Infusions of ATB200 Co-administered with Oral AT2221 in Adult Subjects with Pompe Disease

Study Center(s): Multicenter, international

Protocol Number: ATB200-02

Phase of Development: Phase 1/2

Objectives:

Primary

- To evaluate the safety and tolerability of single-ascending doses of intravenously (IV) infused ATB200
- To evaluate the safety and tolerability of single-ascending doses of IV infused ATB200 as a fixed dose, co-administered with ascending oral doses of AT2221
- To characterize the pharmacokinetics (PK) of single-ascending doses of IV infused ATB200
- To characterize the single- and multiple-dose PK of IV infused 20 mg/kg ATB200 when co-administered with oral 130 mg or 260 mg AT2221
- To characterize the PK of single- and multiple-oral doses of 130 mg or 260 mg AT2221 when co-administered with IV infused ATB200

Secondary

- To evaluate the long-term efficacy of 20 mg/kg of IV infused ATB200 as a fixed dose co-administered with oral 260 mg AT2221 in all subjects from Stage 3
- To evaluate the long-term safety and tolerability of 20 mg/kg of IV infused ATB200 as a fixed dose co-administered with oral 260 mg AT2221 in all subjects from Stage 3
- To characterize single- and multiple-dose PK of plasma rhGAA activity and total rhGAA protein following IV infused 20 mg/kg ATB200 as a fixed dose co-administered with oral 260 mg AT2221 in enzyme replacement therapy (ERT)-naïve subjects

- To characterize the single- and multiple-dose PK of plasma AT2221 following 20 mg/kg of IV infused ATB200 co-administered with oral 260 mg AT2221 in ERT-naïve subjects

Exploratory

To evaluate:

- Anti-rhGAA antibodies (total and neutralizing)
- Cross-reactivity of anti-rhGAA antibodies to alglucosidase alfa
- Pro-inflammatory cytokines and other biomarkers of immune system activation
- Pharmacodynamic (PD) markers (urine hexose tetrasaccharide [Hex4] and serum creatine phosphokinase [CK])
- Impact of anti-rhGAA antibodies on plasma GAA total protein exposures

Study Design:

This is an open-label, fixed-sequence, single- and multiple-ascending dose, first-in-human (FIH) study to evaluate the safety, tolerability, PK, PD, and efficacy of intravenous ATB200 alone and when co-administered with oral AT2221. The study will be conducted in 4 stages.

Approximately 10 to 12 ERT-experienced (alglucosidase alfa) ambulatory subjects (Cohort 1) will be enrolled in Stages 1 and 2. In Stage 1, safety, tolerability, and PK will be evaluated following sequential single-ascending doses of 5 mg/kg, 10 mg/kg, and 20 mg/kg of IV infused ATB200 administered 2 weeks apart. In Stage 2, safety, tolerability, and PK will be evaluated following single- and multiple-ascending dose combinations: 20 mg/kg of IV infused ATB200 co-administered with 130 mg of AT2221 administered orally every 14 days (± 3 days) for 3 doses, followed by 20 mg/kg of IV infused ATB200 co-administered with 260 mg of AT2221 administered orally for 3 doses. ERT-experienced ambulatory subjects who complete Stages 1 and 2 will enter into a long-term extension stage of the study, hereinafter referred to as Stage 3, and will continue to be assessed for safety, tolerability, and efficacy of extended treatment with 20 mg/kg of IV ATB200 co-administered with 260 mg of AT2221 administered orally. In addition, disease-relevant functional assessments will be performed at regular intervals.

In Stage 3, approximately, 12 to 18 additional subjects will enroll, of whom approximately 4 to 6 will be ERT-experienced nonambulatory (Cohort 2), 5 will be ERT-naïve ambulatory subjects (Cohort 3), and approximately 6 to 8 will be ERT-experienced ambulatory subjects who have completed at least 7 years of ERT (Cohort 4). These subjects will be treated with 20 mg/kg of IV infused ATB200 co-administered with 260 mg of AT2221 administered orally every 2 weeks and evaluated for safety, tolerability, PD, and efficacy. Duration of Stage 3 will be 2 years.

Stage 4 treatment period will begin at the end of Stage 3 and continue as an open-label extension until subject withdrawal, regulatory approval or marketing authorization and/or commercialization in the participating subject's country, or study termination by the sponsor, Amicus Therapeutics, Inc. (Amicus).

ERT-naïve subjects will also undergo blood sample collection for single- and multiple-dose PK assessments. Based on having previous exposure to ERT, PK assessments are expected to be similar in ERT-experienced nonambulatory and ERT-experienced ambulatory subjects. In addition, the intensive PK sampling in this study is likely to present an undue burden to nonambulatory subjects. For these reasons, no PK assessments will be conducted in ERT-experienced nonambulatory subjects.

ERT-experienced ambulatory subjects are defined as adults diagnosed with Pompe disease who have been on ERT for 2 to 6 years (Cohort 1) or ≥ 7 years (Cohort 4), prior to enrollment, and who are able to walk at least 200 meters in the 6-Minute Walk Test (6MWT).

ERT-experienced nonambulatory subjects (Cohort 2), are defined as adults diagnosed with Pompe disease who are wheelchair bound and unable to walk unassisted, and have been on ERT for ≥ 2 years prior to enrollment.

ERT-naïve ambulatory subjects (Cohort 3), are defined as adults with Pompe disease who have never received treatment with ERT and who are able to walk at least 200 meters in the 6MWT.

Table 1: Treatment Assignment for Stages 1, 2, 3, and 4

Cohorts	Stage 1 (6 weeks)			Stage 2 (12 weeks)		Stage 3 (2 years)	Stage 4 (Until Approval)
	Period 1 Single-Dose	Period 2 Single-Dose	Period 3 Single-Dose	Period 4 3 Multiple Doses Co-administration	Period 5 3 Multiple Doses Co-administration	Multiple Dose, 24 months, Extension Co-administration	Multiple Dose, Long-term, Extension Co-administration
Cohort 1 (n = 11)	5 mg/kg ATB200	10 mg/kg ATB200	20 mg/kg ATB200	20 mg/kg ATB200 + 130 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221
Cohort 2 (n = 4-6)	NA					20 mg/kg ATB200 + 260 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221
Cohort 3 (n = 5)	NA					20 mg/kg ATB200 + 260 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221
Cohort 4 (n = 6-8)	NA					20 mg/kg ATB200 + 260 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221

Abbreviations: NA = not applicable

Note: At least 1 of the 2 sentinel subjects will complete Stage 2, Period 5 and the safety data will be reviewed by the Safety Steering Committee (SSC) before dosing any newly enrolled subjects from Cohorts 2 and 3. The first 2 subjects from Cohorts 2 and 3 will also serve as sentinel subjects for their respective cohorts.

Study Procedures

Subjects in Cohort 1 will participate in Screening, Baseline, Stage 1 (3-period, fixed-sequence, single-ascending dose of ATB200 alone), Stage 2 (2-period, fixed-sequence, multiple-dose of 20 mg/kg ATB200 co-administered with multiple-ascending doses of AT2221), and Stage 3 (a 24-month extension stage with continued dosing of 20 mg/kg ATB200 co-administered with 260 mg AT2221) of the study. Stage 4 treatment period will begin at the end of Stage 3 and continue as an open-label extension until subject withdrawal, regulatory approval or marketing authorization and/or commercialization in the participating subject's country, or study termination by Amicus.

Subjects in Cohorts 2, 3, and 4 will participate in Screening and Baseline for Stage 3 of the study, as well as Stage 3 dosing-visits. Additionally, subjects in Cohort 3 will participate in serial 24-hour PK visits. Subjects in Cohort 2 and 4 will not participate in PK analyses. The procedures and assessments performed are as follows:

Screening:

- At Screening for either Stage 1 or Stage 3, all subjects will provide informed consent and undergo review of eligibility criteria. Assessments for all subjects include medical history (including forced vital capacity [FVC] and 6MWT, CK, alanine aminotransferase [ALT], aspartate aminotransferase [AST], and urine Hex4 lab results) and history of falls; review of prior and concomitant medications and nondrug therapies; vital signs (heart rate [HR], respiration rate [RR], blood pressure [BP], and temperature); height; weight; comprehensive physical examination (PE); 12-lead electrocardiogram (ECG); clinical safety laboratory assessments (serum chemistry, hematology, and urinalysis); urine pregnancy test; and urine sample for Hex4. Blood samples will also be obtained for immunological assessments (total and neutralizing antibodies, exploratory pro-inflammatory cytokines/other biomarkers of immune system activation, cross-reactivity to alglucosidase alfa, and immunoglobulin E [IgE]), exploratory biomarkers of Pompe disease, and immunogenicity. For subjects in Cohorts 1, 2, and 4, a history of prior infusion-associated reactions (IARs) will also be obtained, and gene that encodes human acid α-glucosidase (*GAA)* genotyping will be performed for those who are unable to provide *GAA* genotyping report at screening. A subject who meets all of the inclusion criteria and none of the exclusion criteria will be assigned to Stage 1 as described above in Table 1.

Baseline:

- Baseline assessments for all subjects include review of eligibility criteria; medical history and history of falls, adverse event (AE) and serious AE (SAE) inquiry, review of prior and concomitant medications and nondrug therapies; vital signs (HR, RR, BP, and temperature); weight; brief PE; ECG; Rasch-built Pompe-specific activity (R-PAct) Scale; Rotterdam Handicap Scale (RHS); Fatigue Severity Scale (FSS); Patient-reported Outcomes Measurement Information System (PROMIS®) for dyspnea, fatigue, physical functioning, and upper extremity; pulmonary function tests (PFTs); motor function tests; and

muscle strength tests for all subjects. Additionally, a history of IARs will be obtained for subjects in Cohorts 1, 2, and 4.

- Retrospective data for patients from Cohorts 1, 2, and 3: All available data for 6MWT, pulmonary function tests, other motor function tests such as 10-meter walk, 4-stair climb, and Gower's, quantitative (dynamometry) and qualitative muscle strength tests, Gait, Stairs, Gower, and Chair maneuver (GSGC) score, and patient-reported outcomes (PROs), if available should be recorded in the Electronic Data Capture (EDC).
- Retrospective data will be gathered from subjects in Cohort 4: At least three 6MWT results, each at least 6 months apart must be available and entered in the EDC. At least two 6MWT results should be within the past three years. Additionally, pulmonary function tests, other motor function tests such as 10-meter walk, 4-stair climb, and Gower's, quantitative (dynamometry) and qualitative muscle strength tests, GSGC score, and PROs, if available should be recorded in the EDC. In addition to the above requirement, all available data will be collected.

Stage 1, Periods 1, 2, and 3:

- Safety: review of AEs, including SAEs and IARs; review of concomitant medications and nondrug therapies; vital signs (HR, RR, BP, and temperature); brief PE; ECG; clinical safety laboratory assessments (serum chemistry, hematology, and urinalysis); and urine pregnancy test.
- PD: urinary Hex4 and serum CK.
- Immunological: blood samples for anti-rhGAA antibodies (total, neutralizing, and cross-reactive with alglucosidase alfa) and blood samples for measurement of pro-inflammatory cytokines and other biomarkers of immune system activation. If needed, IgE and total GAA protein concentration will also be measured.
- Serial 24-hour PK: During Period 1 (Visit 3, Day 1), Period 2 (Visit 4, Day 15), and Period 3 (Visit 5, Day 29), blood sampling for plasma human acid α-glucosidase (GAA) activity levels and total GAA protein concentrations will be taken for all subjects.

Stage 2, Periods 4 and 5:

- Safety: review of AEs, including SAEs and IARs; review of concomitant medications and nondrug therapies; vital signs (HR, RR, BP, and temperature); weight; PE; ECG; clinical safety laboratory assessments (serum chemistry, hematology, and urinalysis); and urine pregnancy test.
- PD: urinary Hex4 and serum CK.
- Immunological: blood samples for anti-rhGAA antibodies (total, neutralizing, and cross-reactive with alglucosidase alfa) and blood samples for measurement of pro-inflammatory cytokines and other biomarkers of immune system activation. If needed, IgE and total GAA protein concentration will also be measured.

- Serial 24-hour PK: During Period 4 (Visit 6, Day 43 and Visit 8, Day 71) and Period 5 (Visit 9, Day 85 and Visit 11, Day 113), blood sampling for plasma GAA activity levels, total GAA protein concentrations, and AT2221 concentrations will be taken for all subjects.

End of PK:

- Safety: review of AEs, including SAEs and IARs; review of concomitant medications and nondrug therapies; vital signs (HR, RR, BP, and temperature); weight; PRO; PFT; motor function; muscle strength tests; PE; ECG; clinical safety laboratory assessments (serum chemistry, hematology, and urinalysis); and urine pregnancy test.
- PD: urinary Hex4 and serum CK.
- Immunological: blood samples for anti-rhGAA antibodies (total, neutralizing, and cross-reactive with alglucosidase alfa) and blood samples for measurement of pro-inflammatory cytokines and other biomarkers of immune system activation. If needed, IgE and total GAA protein will also be measured.

Stage 3:

- Safety: review of AEs, including SAEs, as well as IARs; review of concomitant medications and nondrug therapies; vital signs (HR, RR, BP, and temperature); weight; height; PRO; PE; ECG; clinical safety laboratory assessments (serum chemistry, hematology, and urinalysis); and urine pregnancy test.
- PD: urinary Hex4 and serum CK.
- Immunological: blood samples for anti-rhGAA antibodies (total, neutralizing, and cross-reactive with alglucosidase alfa) and blood samples for measurement of pro-inflammatory cytokines and other biomarkers of immune system activation. If needed, IgE and total GAA protein concentration will also be measured.
- Serial 24-hour PK for subjects in Cohort 3 only: single-dose (first administration of 20 mg/kg ATB200 and 260 mg AT2221), Day 1, and multiple-dose (third administration of 20 mg/kg ATB200 and 260 mg AT2221), Week 4 blood samples for plasma GAA activity levels, total GAA protein concentrations, and AT2221 concentrations will be collected.
- Follow-up sparse blood sampling for plasma total GAA protein will be performed after at least 18 months of ATB200/AT2221 treatment. If the subject has completed Stage 3, sparse PK sampling will be performed in Stage 4.
- Global impression of change (Physician Global Impression of Change [PGIC] and Subject Global Impression of Change [SGIC]); PFTs; motor function tests; muscle strength tests.

Stage 4:

- Safety: review of AEs, including SAEs, as well as IARs; review of concomitant medications and nondrug therapies; vital signs (HR, RR, BP, and temperature); weight; PE; ECG; clinical safety laboratory assessments (serum chemistry, hematology, and urinalysis); and urine pregnancy test.
- PD: urinary Hex4 and serum CK.
- Immunological: blood samples for anti-rhGAA antibodies (total, neutralizing, and cross-reactive with alglucosidase alfa) and blood samples for measurement of pro-inflammatory cytokines and other biomarkers of immune system activation. If needed, IgE and total GAA protein concentration will also be measured.
- Global impression of change (PGIC and SGIC); PFTs; motor function tests; and muscle strength tests.

Subjects who prematurely withdraw from the study will come in for an Early Termination (ET) visit and will undergo all of the assessments that are to be performed at the ET/End of Study visit. No study treatment will be administered. If any of the sentinel subjects withdraw prematurely from the study, that subject will be replaced by the next subject enrolled in the study (eg, if Subject 1 withdraws, Subject 3 will replace that subject as a sentinel subject).

Upon study completion or subject discontinuation, subjects who are confirmed positive for anti-rhGAA antibodies will complete follow-up immunological testing as long as they have a positive result for up to 12 months after the last dose of study drug (ie, at 1-, 3-, 6-, 9-, and 12-months follow-up) or until they begin treatment with another ERT or investigational therapy. If anti-rhGAA antibodies are not confirmed positive at follow-up time points earlier than 6 months, no further testing is required.

Safety Monitoring

Safety will be monitored by the Amicus Medical Monitor and the investigators on a continuous basis and on a regular basis by a Safety Steering Committee (SSC). The details of the membership, meeting schedules, procedures, roles, and responsibilities of the SSC are detailed in the SSC Charter.

Subjects will be monitored in a hospital or infusion center for 48 hours from the start of each infusion during Stage 1 (ie, ERT dose-escalation stage). Since the target infusion duration in the study is 4 hours, all subjects will be closely monitored for the first 4 hours during their infusion and then observed in hospital for an additional 44 hours prior to discharge. Subjects will be admitted and dosed at either an infusion center or a hospital and thereafter hospitalized for the remainder of the observation period.

In Stages 2 and 3, all subjects will be monitored at a hospital or infusion center for an additional 2 to 4 hours from the end of their infusion. Once a subject has completed 1 year in Stage 3, the subjects who have not experienced IARs for the prior 6 months, can be monitored for 1 hour from the end of their infusion, at the PI's discretion. Subjects who switch to home infusion, must be monitored for 2 to 4 hours after the end of their infusions for a period of at least 3 months. If no IARs or other AEs occur during the 3 months, the monitoring can be reduced to 1 hour post infusion at the PIs discretion.

When at least 1 of the 2 Cohort 1 sentinel subjects has completed Stage 2, Period 5 sentinel dosing and the safety data has been reviewed by the SSC, subjects from Cohorts 2 and 3 can begin enrollment into Stage 3. The first 2 subjects from Cohorts 2 and 3 will undergo sentinel dosing in the same manner as subjects from Cohort 1 during Stage 2.

Sentinel Dosing

The first 2 subjects in Cohort 1 of this study will be the sentinel subjects for the study and will be the first 2 subjects dosed in each period of the study (Periods 1 to 5). The first 2 subjects dosed in each cohort will be the sentinel subjects for that cohort. In the event that a sentinel subject is prematurely withdrawn from the study, he/she will be replaced by the next enrolled subject in that cohort.

Note: At least one of the 2 sentinel subjects will complete Period 5, Stage 2 dosing, and all available safety data from Cohort 1 subjects will be reviewed by the SSC, before dosing of subjects in Cohorts 2 and 3 can be started. The first 2 subjects in Cohorts 2 and 3 will also serve as sentinel subjects for their respective cohorts.

In Stage 1 (Periods 1, 2, and 3), subjects will be dosed with single-ascending doses of ATB200 (5 mg/kg [Period 1], 10 mg/kg [Period 2], and 20 mg/kg [Period 3]).

Following the dosing of the 2 sentinel subjects for each study period in Stage 1, an evaluation of the available safety data (PE, vital signs, AEs, infusion reactions, ECG, and available locally performed laboratory tests) will be performed within 48 hours by the Amicus Medical Monitor and the investigators. The SSC will convene for a formal safety review when central safety laboratory data are available for both sentinel subjects at each dose level. If the SSC determines that there are no safety concerns that preclude dosing at the dose assigned for that period, up to 10 additional Cohort 1 subjects will be enrolled and dosed.

Note: Vital signs will be monitored as detailed in the Schedule of Assessments tables. For PK Visits 3, 4, 5, 6, and 9, vital signs will be monitored at hours 0, 1, 4, 6, 12, and 24 after the start of infusion. During the 48-hour observation period, vital signs will be monitored, at a minimum, every 6 to 8 hours until the end of the 48-hour observation period.

The SSC will also convene for a safety review when safety data (including central laboratory safety data) for all Cohort 1 subjects at all three Stage 1 dose levels are available.

Stage 2 (after Visits 6 and 9), safety data from 2 sentinel subjects will be reviewed by the SSC, and safety will be assessed after the first dose as for each period in Stage 1. If the SSC determines that there are no safety concerns that preclude additional dosing at 20 mg/kg ATB200 co-administered with 130 mg AT2221 (Visit 6) or 20 mg/kg ATB200 co-administered with 260 mg AT2221 (Visit 9), up to 10 additional subjects will receive 3 biweekly (every other week) doses at the dose assigned for that period.

The SSC will reconvene when all safety data (including central safety laboratory data) are available for all Cohort 1 subjects at the end of Stage 2.

Stage 3, sentinel dosing will be performed for the first 2 subjects in Cohort 2 and the first 2 subjects in Cohort 3. If the SSC determines that there are no safety concerns that preclude the additional dosing of 20 mg/kg of ATB200 co-administered with 260 mg of AT2221, the

remaining 2 to 4 additional subjects in Cohort 2 and the remaining 2 to 4 additional subjects in Cohort 3 will be dosed.

During the long term extension (Stage 3 and Stage 4) the SSC will convene periodically - at least 2 times a year. The SSC will also convene ad hoc in case of an SAE/suspected unexpected serious adverse reaction (SUSAR) or an identified safety concern.

The SSC may recommend any of the following review:

- Continue the study without modifications
- Continue the study with modifications (amendment)
- Temporarily halt dosing
- Permanently stop dosing

If in the opinion of the SSC there are no AEs or safety concerns in the sentinel subjects that might preclude continued study dosing, dosing will continue for all remaining subjects at that dose level.

Subject safety will continue to be closely monitored by the Amicus Medical Monitor and study investigators on an ongoing basis, and at regular intervals by the SSC.

Home Infusion

Subjects participating in the study will be considered eligible for administration of ATB200/AT2221 at their home after 6 months of ATB200/AT2221 infusions in the study and without an IAR for a period of 6 months prior to last infusion. Subjects with history of recurrent severe or life-threatening IARs including anaphylaxis are not eligible for home infusions. Eligible subjects may request participation in the Home Infusion Program with requests granted on a case-by-case basis following discussion between the Amicus Medical Monitor and Principal Investigator with prior approvals from Institutional Review Boards (IRBs)/Ethics Committees (ECs), and Regulatory Authorities (RAs). Administration of ATB200/AT2221 will be performed at home for eligible subjects by a trained home infusion nurse provided by an Amicus designated home infusion service provider. All subjects participating in the Home Infusion, will be required to complete all functional assessments at the investigational site as defined in the protocol.

Number of Subjects (Planned): Approximately 18 to 34 subjects will enroll in the study. Approximately 10 to 12 subjects in Cohort 1, 4 to 6 subjects in Cohort 2, 5 subjects in Cohort 3, and approximately 6 to 8 subjects in Cohort 4.

Diagnosis and Eligibility Criteria: At the Screening Visit for Stages 1 and 2, subjects from Cohort 1 will be evaluated using the eligibility criteria outlined below. At the Screening Visit for Stage 3, subjects from Cohorts 2, 3, and 4 will be evaluated with the eligibility criteria outlined below as well. Each subject must meet all of the inclusion criteria and none of the exclusion criteria. Waivers of inclusion/exclusion criteria are not permitted.

Inclusion/Exclusion Criteria

Cohort 1

Inclusion Criteria

1. Male and female subjects between 18 and 65 years of age, inclusive
2. Subject must provide signed informed consent prior to any study-related procedures
3. Subjects of childbearing potential must agree to use medically accepted methods of contraception during the study and for 90 days after last co-administration of ATB200 and AT2221
4. Subject has a diagnosis of Pompe disease based on documented deficiency of GAA enzyme activity or by *GAA* genotyping
5. Subject has received ERT with alglucosidase alfa (Myozyme/Lumizyme) for the previous 2 to 6 years, inclusive
6. Subject is currently receiving alglucosidase alfa (Myozyme/Lumizyme), at a frequency of once every other week
7. Subject has received and completed the last 2 infusions without a drug-related adverse event resulting in dose interruption
8. Subject must be able to walk between 200 and 500 meters on the 6MWT
9. Upright FVC must be 30% to 80% of predicted normal value

Exclusion Criteria

1. Subject has received any investigational therapy including adjunctive therapy for Pompe disease, other than alglucosidase alfa within 30 days or 5 half-lives of the therapy or treatment, whichever is longer, prior to the Baseline Visit, or anticipates doing so during the study
2. Subject has received treatment with prohibited medications within 30 days of the Baseline Visit
3. Subject, if female, is pregnant or breastfeeding at screening
4. Subject, whether male or female, is planning to conceive a child during the study
5. Subject requires invasive ventilatory support
6. Subject uses noninvasive ventilatory support ≥ 6 hours a day while awake
7. Subject has a medical or any other extenuating condition or circumstance that may, in the opinion of the investigator or the Medical Monitor, pose an undue safety risk to the subject or compromise his/her ability to comply with protocol requirements
8. Subject has a history of anaphylaxis to alglucosidase alfa
9. Subject has a history of high sustained anti-rhGAA antibodies
10. Subject has a history of allergy or sensitivity to miglustat or other iminosugars

11. Subjects with active systemic autoimmune disease such as lupus, scleroderma, or rheumatoid arthritis; all subjects with autoimmune disease must be discussed with the Amicus Medical Monitor
12. Subjects with active bronchial asthma; all subjects with bronchial asthma must be discussed with the Amicus Medical Monitor

Cohort 2

Inclusion Criteria

10. Male and female subjects between 18 and 65 years of age, inclusive
11. Subject must provide signed informed consent prior to any study-related procedures
12. Subjects of childbearing potential must agree to use medically accepted methods of contraception during the study and for 90 days after last co-administration of ATB200 and AT2221
13. Subject has a diagnosis of Pompe disease based on documented deficiency of GAA enzyme activity or by *GAA* genotyping
14. Subject has received ERT with alglucosidase alfa (Myozyme/Lumizyme) for ≥ 2 years
15. Subject is currently receiving alglucosidase alfa (Myozyme/Lumizyme), at a regular or set frequency
16. Subject has received and completed the last 2 infusions without a drug-related adverse event resulting in dose interruption
17. Subject must be wheelchair-bound and unable to walk unassisted

Exclusion Criteria

13. Subject has received any investigational therapy, including adjunctive therapy for Pompe disease, other than alglucosidase alfa within 30 days or 5 half-lives of the therapy or treatment, whichever is longer, prior to the Baseline Visit, or anticipates to do so during the study
14. Subject has received treatment with prohibited medications within 30 days of the Baseline Visit
15. Subject, if female, is pregnant or breastfeeding at screening
16. Subject, whether male or female, is planning to conceive a child during the study
17. Subject has a medical or any other extenuating condition or circumstance that may, in the opinion of the investigator or the Medical Monitor, pose an undue safety risk to the subject or compromise his/her ability to comply with protocol requirements
18. Subject has a history of anaphylaxis to alglucosidase alfa
19. Subject has a history of high sustained anti-rhGAA antibodies
20. Subject has a history of allergy or sensitivity to miglustat or other iminosugars

21. Subjects with active systemic autoimmune disease such as lupus, scleroderma, or rheumatoid arthritis; all subjects with autoimmune disease must be discussed with the Amicus Medical Monitor
22. Subjects with active bronchial asthma; all subjects with bronchial asthma must be discussed with the Amicus Medical Monitor

Cohort 3

Inclusion Criteria

18. Male and female subjects between 18 and 65 years of age, inclusive
19. Subject must provide signed informed consent prior to any study-related procedures
20. Subjects of childbearing potential must agree to use medically accepted methods of contraception during the study and for 90 days after last co-administration of ATB200 and AT2221
21. Subject has a diagnosis of Pompe disease based on documented deficiency of GAA enzyme activity or by *GAA* genotyping
22. Subject must be able to walk between 200 to 500 meters on the 6MWT
23. Upright FVC must be 30% to 80% of predicted normal value

Exclusion Criteria

23. Subject has received any ERT, including alglucosidase alfa at any time, or any investigational therapy for Pompe disease within 30 days or 5 half-lives of the therapy or treatment, whichever is longer, prior to the Baseline Visit, or anticipates doing so during the study
24. Subject has received treatment with prohibited medications within 30 days of the Baseline Visit
25. Subject, if female, is pregnant or breastfeeding at screening
26. Subject, whether male or female, is planning to conceive a child during the study
27. Subject requires invasive ventilatory support
28. Subject uses noninvasive ventilatory support $\geq$ 6 hours a day while awake
29. Subject has a medical or any other extenuating condition or circumstance that may, in the opinion of the investigator or the Medical Monitor, pose an undue safety risk to the subject or compromise his/her ability to comply with protocol requirements
30. Subject has a history of allergy or sensitivity to miglustat or other iminosugars
31. Subjects with active systemic autoimmune disease such as lupus, scleroderma, or rheumatoid arthritis; all subjects with autoimmune disease must be discussed with the Amicus Medical Monitor
32. Subjects with active bronchial asthma; all subjects with bronchial asthma must be discussed with the Amicus Medical Monitor

Cohort 4

Inclusion Criteria

24. Male and female subjects between 18 and 75 years of age, inclusive
25. Subject must provide signed informed consent prior to any study-related procedures
26. Subject has documented 6MWT on three separate occasions, each at least six months apart with at least two values in the past three years
27. Subjects of childbearing potential must agree to use medically accepted methods of contraception during the study and for 90 days after last co-administration of ATB200 and AT2221
28. Subject has a diagnosis of Pompe disease based on documented deficiency of GAA enzyme activity or by *GAA* genotyping
29. Subject has received ERT for the previous ≥ 7 years
30. Subject is currently receiving alglucosidase alfa (Myozyme/Lumizyme) at a frequency of once every other week
31. Subject has received and completed the last 2 infusions without a drug-related AE resulting in dose interruption
32. Subject must be able to walk between 75 and 600 meters on the 6MWT
33. Upright FVC must be 30% to 85% of predicted normal value

Exclusion Criteria

33. Subject has received any investigational therapy including adjunctive therapy for Pompe disease, other than alglucosidase alfa within 30 days or 5 half-lives of the therapy or treatment, whichever is longer, prior to the Baseline Visit, or anticipates doing so during the study
34. Subject has received treatment with prohibited medications within 30 days of the Baseline Visit
35. Subject, if female, is pregnant or breastfeeding at screening
36. Subject, whether male or female, is planning to conceive a child during the study
37. Subject requires invasive ventilatory support
38. Subject uses noninvasive ventilatory support ≥ 6 hours a day while awake
39. Subject has a medical or any other extenuating condition or circumstance that may, in the opinion of the investigator or the Medical Monitor, pose an undue safety risk to the subject or compromise his/her ability to comply with protocol requirements
40. Subject has a history of anaphylaxis to alglucosidase alfa
41. Subject has a history of high sustained anti-rhGAA antibodies
42. Subject has a history of allergy or sensitivity to miglustat or other iminosugars

43. Subjects with active systemic autoimmune disease such as lupus, scleroderma, or rheumatoid arthritis; all subjects with autoimmune disease must be discussed with the Amicus Medical Monitor
44. Subjects with active bronchial asthma; all subjects with bronchial asthma must be discussed with the Amicus Medical Monitor

Investigational Product, Dosage, and Mode of Administration:

Stage 1 (consists of 3 dosing periods 2 weeks apart for Cohort 1)

- Period 1: a single-dose IV infusion of 5 mg/kg ATB200
- Period 2: a single-dose IV infusion of 10 mg/kg ATB200 to all subjects who have completed Period 1
- Period 3: a single-dose IV infusion of 20 mg/kg ATB200 to all subjects who have completed Period 2

Stage 2 (consists of 2 dosing periods, each comprising 3 study treatment doses, 2 weeks apart for Cohort 1)

- Period 4: 130 mg of AT2221 will be administered orally 1 hour before a single-dose IV infusion of 20 mg/kg ATB200 to all subjects who have completed Period 3 (repeated every 2 weeks for a total of 3 administrations)
- Period 5: 260 mg of AT2221 will be administered orally 1 hour before a single-dose IV infusion of 20 mg/kg ATB200 to all subjects who have completed Period 4 (repeated every 2 weeks for a total of 3 administrations)

Stage 3 (consists of multiple study-dosing, administered every 2 weeks apart for a duration of 24 months)

Stage 4 (consists of multiple study-dosing, administered every 2 weeks apart until study treatment approval or discontinuation of the program)

Study treatment for all subjects in Stage 3 will consist of AT2221 administered orally at 260 mg followed approximately 1 hour later by a single-dose IV infusion of ATB200 at 20 mg/kg (repeated every 2 weeks for up to 24 months).

Note: Subjects are required to fast at least 2 hours before and 2 hours after administration of oral AT2221.

Total Duration of Study:

- Screening: up to 4 weeks
- Stage 1 + Stage 2: 18 weeks
- Stage 3: 24 months
- Stage 4: Until completion or drug approval

Criteria for Evaluation:

Primary:

Safety assessments:

- PEs
- Vital signs, including body temperature, RR, HR, and BP
- AEs, including IARs
- 12-lead ECG
- Clinical safety laboratory assessments: serum chemistry, hematology, and urinalysis

PK of plasma ATB200 and AT2221:

- Plasma GAA activity levels and total GAA protein concentrations PK parameters: maximum observed plasma concentration (C_{max}), time to reach the maximum observed plasma concentration (t_{max}), area under the plasma-drug concentration time curve from Time 0 to the time of last measurable concentration (AUC_{0-t}), area under the plasma-drug concentration time curve from Time 0 extrapolated to infinity ($AUC_{0-\infty}$), alpha- and beta-phase half-life ($t_{½}$), total clearance following IV administration (CL_T), and steady state volume of distribution (V_{ss})
- Ratios of plasma GAA activity levels and total GAA protein concentration C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ for all dose regimens
- Plasma AT2221 PK parameters: C_{max}, t_{max}, AUC_{0-t}, $AUC_{0-\infty}$, and $t_{½}$, apparent total clearance of drug following oral administration (CL_T/F), and terminal phase volume of distribution following oral administration (V_z/F) for each dose level
- Ratio of plasma AT2221 PK parameters: C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ for each dose level
- Exposure estimates from sparse plasma total GAA protein concentrations after at least 18 months on ATB200/AT2221 treatment in Cohorts 1 and 3 patients will be compared to exposures after the first and third doses of 20 mg/kg ATB200 + 260 mg AT2221.

Functional Assessments (performed at Baseline and every 3 months in Stage 3):

For Ambulatory Subjects:

- Motor Function Tests
  - 6MWT
  - 10-Meter Walk Test (10MWT)
  - Gait, Stairs, Gower, and Chair maneuver (GSGC)
  - Timed Up and Go (TUG)

- Muscle Strength Test
  - medical research criteria (MRC) and hand-held dynamometer for both upper and lower limbs
- PFTs
  - FVC, maximum inspiratory pressure (MIP), maximum expiratory pressure (MEP), and Sniff Nasal Inspiratory Pressure (SNIP)

For Nonambulatory Subjects:

- Muscle Strength Test - Upper Limbs Only
  - MRC and hand-held dynamometer performed for upper limbs only
- PFTs - in subjects without invasive ventilatory support only
  - FVC, MIP, MEP, and SNIP

Patient-reported Outcomes (performed at Baseline and every 3 months in Stage 3):

- FSS
- RHS
- R-PAct Scale
- PROMIS instruments for dyspnea, fatigue, physical functioning, and upper extremity

Exploratory:

- Anti-rhGAA antibodies (total and neutralizing)
- Cross-reactivity of anti-rhGAA antibodies to alglucosidase alfa
- Pro-inflammatory cytokines and other biomarkers of immune system activation
- PD biomarkers: Hex4 and serum CK

Methods of Analysis:

Statistical Methods for PK Parameters:

Details of the analysis methods related to PK parameters will be provided in a stand-alone PK Statistical Analysis Plan (PK SAP). In general, basic descriptive statistics will be provided for all PK parameters.

Cohort 1

- Dose proportionality assessment on GAA activity levels and total GAA protein concentration exposure (C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$) ratios of 5, 10, and 20 mg/kg ATB200 alone.
- Analysis of variance (ANOVA) on GAA activity levels and total GAA protein concentration exposure (C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$) ratios of 20 mg/kg ATB200

alone versus 20 mg/kg ATB200 + 130 mg AT2221, and versus 20 mg/kg ATB200 + 260 mg AT2221.

- Dose proportionality assessment for exposure ratios (C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$) between 130 mg and 260 mg AT2221.
- The effect of immunogenicity results on PK, PD, and safety will be evaluated. Further details will be provided in the Statistical Analysis Plan (SAP).

Cohort 3

- ANOVA on GAA activity levels and total GAA protein concentration exposure (C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$) ratios of 20 mg/kg ATB200 + 260 mg AT2221 in subjects from Cohort 3 versus 20 mg/kg ATB200 alone in subjects from Cohort 1, versus 20 mg/kg ATB200 + 130 mg AT2221 in subjects from Cohort 1, and versus 20 mg/kg ATB200 + 260 mg AT2221 in subjects from Cohort 1.
- ANOVA on AT2221 exposure ratios (which will have the same PK parameters) for 260 mg AT2221 in subjects from Cohort 3 versus 130 mg AT2221 and 260 mg AT2221 in subjects from Cohort 1.

Statistical Methods for Non-PK-Specific Parameters:

Details of the analysis methods related to all non-PK-specific parameters will be provided in a separate SAP for the study.

In general, descriptive statistics (n, mean, standard deviation [SD], median, minimum, and maximum) will be provided for all efficacy continuous variables. Each cohort will have its own baseline, and analysis will be performed by cohort and overall. In addition, summary analyses will be performed by combining Cohort 1, 3, and 4 for ambulatory subjects, or by combining Cohort 1 and 4 for ERT-experienced subjects. For Cohort 1, the analysis will also be provided by treatment dosing regimen. The visit values and the change (and percent change) from baseline to the post-baseline assessment will be analyzed by scheduled visit. The 95% confidence interval for the mean (and percent mean) change from baseline by scheduled visit will be provided for summary purposes only. In addition, the visit values and derived values for each subject will be provided in data listings.

Safety and other variables that are continuous (including vital signs, laboratory assessments, and immunogenicity values) will similarly be analyzed using descriptive statistics. Shift tables will be generated for safety laboratory parameters.

Summary statistics will be provided for all variables that are not PK parameters.

Adverse events and other variables that are categorical will be summarized using counts and percentage. In addition, categorical variables (excluding AEs) will be summarized by scheduled visits.

Interim Analyses: Interim analyses will be performed during the study as needed. Details for the interim analysis will be included in the SAP.

4. TABLE OF CONTENTS, LIST OF TABLES, AND LIST OF FIGURES

TABLE OF CONTENTS

LIST OF TABLES

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5. LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

The following abbreviations and specialized terms are used in this study protocol.

Table 2: List of Abbreviations and Specialized Terms

Term or Abbreviation	Definition
4-MU-α-Glc	4-methylumbelliferyl-α-D-glucopyranoside
6MWT	6-Minute Walk Test
10MWT	10-Meter Walk Test
AE	adverse event
Alglucosidase alfa	active ingredient drug substance name for Lumizyme® and Myozyme®; aglucosidase alfa is produced by recombinant DNA technology in a Chinese hamster ovary cell line
ALT	alanine aminotransferase
ANOVA	analysis of variance
AST	aspartate aminotransferase
AT2221	*N*-butyldeoxynojirimycin; iminosugar which is used as a pharmacological chaperone to ATB200 (a recombinant human acid α-glucosidase)
ATB200	recombinant human acid α-glucosidase (rhGAA) enzyme with optimized carbohydrate structures, including mannose 6-phosphate (M6P), to enhance uptake and delivery of active ATB200 to lysosomes
ATB200/AT2221	ATB200 co-administered with AT2221
AUC	area under the plasma drug concentration time curve
AUC_{0-t}	area under the plasma drug concentration time curve from time zero to observed time (t)
$AUC_{0-\infty}$	area under the plasma drug concentration-time curve from time zero extrapolated to infinite time
BLQ	below the limit of quantification
BP	blood pressure
CFR	Code of Federal Regulations
CI	confidence interval
CI-MPR	cation-independent mannose 6-phosphate receptor
CL	clearance
CL_T	total clearance following IV administration

Table 2: List of Abbreviations and Specialized Terms (Continued)

Term or Abbreviation	Definition
CL_T/F	total clearance following oral administration
CK	creatine phosphokinase
C_{max}	maximum observed plasma concentration
CRO	contract research organization
CS	clinically significant
ECG	electrocardiogram
eCRF	electronic case report form
EDC	electronic data capture
ERT	enzyme replacement therapy
ERT-experienced	for the purposes of this study, this refers to subjects previously treated with alglucosidase alfa for at least 2 years prior to enrollment
ERT-naïve	For the purposes of this study, this refers to subjects who have not been previously treated with alglucosidase alfa
ET	early termination
EU	European Union
FSS	Fatigue Severity Scale; a patient-reported outcomes scale to assess level of fatigue and impact of quality of life related to disease or illness
FIH	first-in-human
FVC	forced vital capacity
Gaa	gene encoding (non-human) acid α-glucosidase
GAA	human acid α-glucosidase, may be specified as either GAA enzyme activity or GAA protein
GAA	gene that encodes human acid α-glucosidase
GAA activity	GAA activity refers to the active enzyme activity measured using 4-MU-α-Glc substrate; represents both endogenous GAA and exogenous rhGAA
GCP	Good Clinical Practice
GSGC	Gait, Stairs, Gower, and Chair maneuver; a clinical outcome assessment scoring system to assess motor function in Pompe disease

Table 2: List of Abbreviations and Specialized Terms (Continued)

Term or Abbreviation	Definition
Hex4	hexose tetrasaccharide; commonly designated as Glc4, representing a biochemical entity (Glc-a-1-6 Glc-a1-4 Glc-a-1-4Glc) determined in urine or plasma as a marker of active glycogen metabolism
HR	heart rate
IAR	infusion-associated reaction
ICH	International Conference on Harmonisation
IEC	International Ethics Committee
IgE	immunoglobulin E
IOPD	infantile-onset Pompe disease
IRB	Institutional Review Board
IV	intravenous(ly)
KO	knock-out
LAMP1	lysosome-associated membrane protein 1
ln	natural log
LOPD	late-onset Pompe disease
Lumizyme®	commercially available rhGAA (also referred to as alglucosidase alfa)
M6P	mannose 6-phosphate
MEP	maximum expiratory pressure; a measure of the strength of respiratory muscles, obtained by having the patient exhale as strongly as possible against a mouthpiece
MIP	maximum inspiratory pressure; the inspiratory pressure generated against a completely occluded airway. It is used to evaluate inspiratory respiratory muscle strength especially diaphragm
MRC	medical research criteria
Myozyme®	commercially available rhGAA; also referred to as alglucosidase alfa
NA	not applicable
NHP	non-human primate
NOAEL	no-observed-adverse-effect-level
PAS	Periodic Acid Schiff
PD	pharmacodynamic

Table 2: List of Abbreviations and Specialized Terms (Continued)

Term or Abbreviation	Definition
PE	physical examination
PFT	pulmonary function test
PGIC	Physician Global Impression of Change
PK	pharmacokinetic
PRO	patient-reported outcome
PROMIS®	Patient-reported Outcomes Measurement Information System
rhGAA	recombinant human acid α-glucosidase
R-PAct scale	Rasch-built Pompe-specific Activity scale
RR	respiration rate
SAE	serious adverse event
SAP	statistical analysis plan
SD	standard deviation
SGIC	Subject Global Impression of Change; patient response scale rating overall improvement ranging from very much improved to very much worse
SNIP	Sniff Nasal Inspiratory Pressure; a non-invasive assessment of inspiratory muscle (diaphragm) strength
SSC	Safety Steering Committee
SUSAR	suspected unexpected serious adverse reaction
$t_{½}$	terminal half-life
TEAE	treatment-emergent adverse event
t_{max}	time to reach the maximum observed concentration
TUG	Timed Up and Go; a timed test used to assess mobility
ULN	upper limit of normal
US	United States
V_z/F	terminal phase volume of distribution following oral administration
V_{ss}	steady state volume of distribution
WHO	World Health Organization

Table 2: List of Abbreviations and Specialized Terms (Continued)

Term or Abbreviation	Definition
Specialized Terms	
Cohort 1	ERT-experienced ambulatory subjects (Defined as adults with Pompe disease who have been on ERT for 2 to 6 years prior to enrollment, and are able to walk at least 200 meters in the 6MWT.)
Cohort 2	ERT-experienced nonambulatory subjects (Defined as adults with Pompe disease who are wheelchair bound and unable to walk unassisted, and have been on ERT (alglucosidase alfa) for ≥ 2 years prior to enrollment.)
Cohort 3	ERT-naïve ambulatory subjects (Defined as adults with Pompe disease who have never received treatment with ERT and are able to walk at least 200 meters in the 6MWT.)
Cohort 4	ERT-experienced ambulatory subjects (Defined as adults with Pompe disease who have been on ERT for at least 7 years prior to enrollment, and are able to walk at least 75 meters in the 6MWT.)

6. INTRODUCTION AND STUDY RATIONALE

6.1. Pompe Disease

Pompe disease, also known as acid maltase deficiency or glycogen storage disease type II, is an autosomal recessive genetic disorder caused by mutations in the gene that encodes acid α-glucosidase (*GAA*), an enzyme that catalyzes the breakdown of lysosomal glycogen. Pompe disease is also classified as a neuromuscular disease or a metabolic myopathy. It has an estimated global incidence of 1:40000 (Martiniuk, Chen et al. 1998; Ausems, Verbiest et al. 1999; Hagemans, Winkel et al. 2005a; Kemper, Hwu et al. 2007). Deficiency of GAA leads to the accumulation of glycogen in various tissues, particularly in striated muscles resulting in a broad spectrum of clinical manifestations that may present during infancy (infantile-onset Pompe disease [IOPD]) or in childhood, adolescence, or adulthood (late-onset Pompe disease [LOPD]). Earlier onset of disease and lower enzymatic activity are generally associated with a more severe clinical course (Kishnani, Steiner et al. 2006). The clinical features of the disease include progressive muscle weakness, respiratory insufficiency, and, in the case of IOPD, marked cardiomegaly, hepatomegaly, and cardiac failure (Hirschhorn and Reuser 2001; van der Ploeg and Reuser 2008). Late-onset Pompe disease can present at any age older than 12 months and is characterized by a lack of cardiac involvement and better short-term prognosis (Hirschhorn and Reuser 2001; Muller-Felber, Horvath et al. 2007; van der Ploeg and Reuser 2008). The symptoms are related to progressive skeletal muscle dysfunction. Proximal lower limb and paraspinal trunk muscles are usually affected first, followed by involvement of the diaphragm and accessory muscles of respiration. In some cases, diaphragmatic weakness may be evident before any other significant weakness is noted (Hagemans, Janssens et al. 2004; Hagemans, Winkel et al. 2005b). Most subjects with Pompe disease eventually progress to physical debilitation requiring the use of a wheelchair and assisted ventilation, with premature death often occurring due to respiratory failure (Hagemans, Janssens et al. 2004; Hagemans, Winkel et al. 2005a).

Management of Pompe disease includes enzyme replacement therapy (ERT) with recombinant human α-glucosidase (rhGAA), cardiopulmonary and gastrointestinal support, musculoskeletal and functional rehabilitation, and dietary therapy. Alglucosidase alfa is the only ERT approved for the treatment of Pompe disease. In subjects with IOPD, treatment with alglucosidase alfa has been shown to significantly improve survival compared to historical controls (Myozyme Package Insert 2010). In LOPD, alglucosidase alfa has been shown to have a statistically significant, albeit modest, effect on the 6-Minute Walk Test (6MWT) and forced vital capacity (FVC) compared to placebo (Myozyme Package Insert 2010). However, the majority of subjects either remain stable or continue to deteriorate while on alglucosidase alfa. The reason for the apparent sub-optimal effect of ERT is unclear, but could be partly due to the poor tissue targeting of the current ERT, development of anti-rhGAA neutralizing antibodies, or the progressive nature of underlying muscle pathology. The effect of alglucosidase alfa is less clear for subjects who are already nonambulatory or receiving ventilatory support. The United States (US) product label includes a black box warning with information on the potential risk of hypersensitivity reaction. Life-threatening anaphylactic reactions, including anaphylactic shock, have been observed in subjects treated with alglucosidase alfa.

This study is designed to evaluate the effect of a more efficiently targeted rhGAA (ATB200) with optimized glycosylation co-administered with a chaperone (AT2221) that has been shown to increase the stability of ERT during circulation. Additionally, the study is designed to evaluate safety, tolerability, pharmacokinetics (PK), pharmacodynamics (PD), and immunogenicity of ATB200 co-administered with AT2221 in adult subjects with Pompe disease.

6.2. Pharmacokinetics of Miglustat

AT2221 (miglustat) capsules contain the same active ingredient in Zavesca® (Actelion Pharmaceuticals US Inc.), which is approved for the treatment of Type 1 Gaucher disease, and in that disease, it functions as a competitive and reversible inhibitor of the enzyme glucosylceramide synthase and it reduces the rate of glycosphingolipid biosynthesis (Zavesca Package Insert 2008). The PK of miglustat has been well characterized in subjects with Gaucher disease. In this study, AT2221 acts as a pharmacological chaperone to enhance the activity of ATB200. AT2221 is administered at a lesser frequency (1 dose every 2 weeks) in contrast to dosing with miglustat for Gaucher disease (3 times daily). Treatment assignments for this study are outlined in Table 8.

Absorption: After a 100 mg oral dose of miglustat, the time maximum observed plasma concentration of miglustat (t_{max}) ranged from 2 to 2.5 hours in Gaucher patients. Plasma concentrations show a bi-exponential decline, characterized by a short distribution phase and a longer elimination phase. The effective half-life of miglustat is approximately 6 to 7 hours, which predicts that steady-state will be achieved by 1.5 to 2 days following the start of three times daily dosing.

Miglustat, dosed at 50 and 100 mg three times daily in Gaucher patients, exhibits dose proportional pharmacokinetics. The pharmacokinetics of miglustat were not altered after repeated dosing three times daily for up to 12 months. In healthy subjects, co-administration of Zavesca with food results in a decrease in the rate of absorption of miglustat (maximum plasma concentration [C_{max}] was decreased by 36% and t_{max} delayed 2 hours) but had no statistically significant effect on the extent of absorption of miglustat (area-under-the-plasma-concentration time curve [AUC] was decreased by 14%). The mean oral bioavailability of a 100 mg miglustat capsule is about 97% relative to an oral solution administered under fasting conditions. The pharmacokinetics of miglustat were similar between adult type 1 Gaucher disease patients and healthy subjects after a single dose administration of miglustat 100 mg (Zavesca Labeling-Package Insert 2014). The absolute bioavailability of miglustat was approximately 80% (Wraith and Imrie 2009).

Distribution: Miglustat does not bind to plasma proteins. Mean apparent volume of distribution of miglustat is 83 to 105 liters in Gaucher patients. At steady state, the concentration of miglustat in cerebrospinal fluid of six non-Gaucher patients was 31.4% to 67.2% of that in plasma, indicating that miglustat crosses the blood brain barrier (Zavesca Labeling-Package Insert 2014).

Metabolism and Excretion: The major route of excretion of miglustat is via kidney. Following administration of a single dose of 100 mg ^{14}C-miglustat to healthy volunteers, 83% of the radioactivity was recovered in urine and 12% in feces. In healthy subjects, 67% of the administered dose was excreted unchanged in urine over 72 hours. The most abundant metabolite in urine was miglustat glucuronide accounting for 5% of the dose. The terminal half-life ($t_{½}$) of radioactivity in plasma was 150 hours, suggesting the presence of one or more metabolites with a

prolonged half-life. The metabolite accounting for this observation has not been identified, but may accumulate and reach concentrations exceeding those of miglustat at steady state. Miglustat did not inhibit or induce substrates of cytochrome P450 enzymes (Zavesca Labeling-Package Insert 2014).

6.2.1. Use of Miglustat as a Pharmacological Chaperone

Miglustat, the active ingredient in AT2221, is a small molecule acting as a pharmacological chaperone and has been shown to bind and stabilize GAA to improve its pharmacological properties. Two clinical studies (Section 6.2.1.2) evaluating the co-administration of miglustat with the current approved ERT (alglucosidase alfa) in Pompe disease have been reported in literature.

6.2.1.1. Effect of Miglustat on Plasma GAA Activity of Alglucosidase Alfa

In a clinical trial conducted in 13 subjects with Pompe disease (3 IOPD and 10 LOPD) at 4 treatment centers in Italy, 20 to 40 mg/kg alglucosidase alfa was administered alone and then co-administered with 4 doses of 80 mg/m^2 miglustat. The results of the study showed a mean 6.8-fold increase in GAA activity exposure (measured in terms of AUC) for co-administration compared to alglucosidase alfa alone (Parenti, Andria et al. 2015).

6.2.1.2. Characterization of Plasma Miglustat With Alglucosidase Alfa in Subjects with Pompe Disease

A study conducted at the University of Florida evaluated the PK of plasma miglustat when co-administered with intravenous (IV) infusion of alglucosidase alfa to subjects with Pompe disease. Preliminary plasma miglustat PK data following the third dose of 100 mg miglustat administered 3 times on the day of alglucosidase alfa infusion and 200 mg miglustat administered as a single-dose on the day of alglucosidase alfa infusion are available. Generally, the plasma PK of miglustat in subjects with Pompe disease was similar to that reported in subjects with Gaucher disease. Plasma miglustat exposures were approximately dose proportional. C_{max} values following 100 mg and 200 mg were 1120 ng/mL and 2450 ng/mL, respectively, and area under the plasma concentration time curve from Time 0 to the time of last measurable concentration (t) (AUC_{0-t}) values were 8587 ng·hr/mL and 16092 ng·hr/mL, respectively. Miglustat reached peak concentrations at approximately 3 hours after single-dose administration, and the $t_{1/2}$ ranged from 4.5 to 6.0 hours. Volume of distribution ranged from 78 to 94 L (Doerfler, Kelley et al. 2014).

6.3. ATB200

6.3.1. Nonclinical Summary of ATB200

ATB200, which is considered a next-generation rhGAA and is expressed in a new Chinese hamster ovary cell line, is manufactured using a process that yields significantly higher mannose 6-phosphate (M6P) content compared to the current standard of care for Pompe disease, alglucosidase alfa.

While alglucosidase alfa provides some clinical benefits, the infused enzyme shows insufficient uptake in key disease-relevant muscles, which is likely due to an inadequate glycosylation of

alglucosidase alfa with M6P, a carbohydrate that binds the cation-independent mannose 6-phosphate receptors (CI-MPR) at the cell surface for enzyme internalization and lysosome targeting. Studies show that ATB200 has optimized glycosylation with considerable amounts of Bis-M6P N-glycans for high-affinity binding to the CI-MPR and targeting to the lysosome compared to alglucosidase alfa. The functional benefit of improved affinity of ATB200 for CI-MPR is shown by its improved cellular uptake in fibroblasts derived from subjects with Pompe disease and rat skeletal muscle myoblasts. These studies show that high alglucosidase alfa concentrations are required for effective cellular uptake of the exogenous ERT in Pompe fibroblasts while substantially lower ATB200 concentrations are needed for efficient cellular uptake.

As previously demonstrated, the tolerability and exposure of alglucosidase alfa ERT was improved with co-administration of the pharmacological chaperone miglustat (designated as AT2221 in current study) (Doerfler, Kelley et al. 2014; Parenti, Moracci et al. 2014). These data suggest that ATB200 may also be improved by co-administering with AT2221. Recombinant human acid α-glucosidase enzymes, including ATB200, are not stable at neutral pH and can be irreversibly inactivated. Addition of AT2221 prevents this irreversible inactivation of rhGAA. *In vitro* data suggest that AT2221 binds and physically stabilizes ATB200, which prevents denaturation and irreversible inactivation of the exogenous enzyme, thus improving the exposure and efficacy of ATB200.

The effect of AT2221 on ATB200 exposure was evaluated in a 13-week combination toxicity study with AT2221 (175 mg/kg, administered orally) and ATB200 (100 mg/kg, administered via 2-hour infusion) administered to non-human primates (NHPs). AT2221 co-administration 30 minutes prior to ATB200 infusion showed an approximate 2-fold increase in ATB200 exposure (AUC) and an approximate 2-fold increase in the effective $t_{1/2}$ of ATB200 as compared to IV administration of 100 mg/kg of ATB200 alone.

Several efficacy studies were conducted in gene encoding (non-human) acid α-glucosidase (*Gaa)* knock-out (KO) mice (the Pompe mouse model) to determine the ability of ATB200 to reduce glycogen levels in skeletal muscles compared to alglucosidase alfa and also to determine the effects of co-administration of AT2221 on ATB200-mediated glycogen reduction. For this purpose, 14- to 16-week-old male *Gaa* KO mice were administered 2 biweekly (every other week) IV bolus injections of 20 mg/kg ATB200. Groups of mice were orally administered various doses of AT2221 (1, 3, 5, 10, 20, and 30 mg/kg) 30 minutes before each IV bolus injection of ATB200. As a comparator, another group of mice was administered 20 mg/kg alglucosidase alfa. Significantly greater glycogen reduction was seen with 20 mg/kg ATB200 compared to alglucosidase alfa at the same dose (confirming *in vitro* results of improved M6P content in ATB200 leading to improved uptake into target cells). Importantly, co-administration with AT2221 further improved (up to 2-fold) the ATB200-mediated glycogen reduction in a dose-related manner. Of all the AT2221 doses tested in *Gaa* KO mice, co-administration of 10 mg/kg AT2221 with 20 mg/kg ATB200 was the most optimal combination that led to greatest glycogen reduction compared to ATB200 alone.

The targeted delivery of ATB200 into the lysosomes was further confirmed histologically by Periodic Acid Schiff (PAS) staining and transmission electron microscopic. The PAS staining of skeletal muscles demonstrated the presence of glycogen in the form of intense, magenta punctates that were abundant in every muscle fiber of untreated mice. Unlike alglucosidase alfa

(20 mg/kg), which showed only a mild improvement of reduction of signal (indicative of reduced glycogen), ATB200 (20 mg/kg) alone led to a significant decrease in PAS signals and co-administration with AT2221 (10 mg/kg) resulted in a further reduction in substrate, as evidenced by the clearance of PAS signals in most muscle fibers. The transmission electron microscopic examination of Epon-embedded quadriceps sections also reconfirmed PAS observations indicating maximal reduction with co-administered ATB200 and AT2221, as not only was the number and size of substrate-containing lysosomes reduced, but also the electron density of remaining lysosomes were reduced. Finally, the lysosomal proliferation that is considered one of the hallmarks of Pompe disease was also tested by immunohistochemistry staining and indicated that, in the muscles examined (quadriceps, heart, and diaphragm), the number and size of lysosome-associated membrane protein 1 (LAMP1)-positive vesicles (ie, lysosomes) were greatly increased in untreated *Gaa* KO mice compared to age-matched wild-type animals. Notably, a marked decrease in the LAMP1 immunohistochemistry signal generally was observed following administration of ATB200 but not with alglucosidase alfa. Co-administration with AT2221 led to a further reduction in lysosome proliferation, whereby the level and pattern of LAMP1 in the majority of muscle fibers returned to those observed in wild-type animals. In general, the effect of co-administration of AT2221 with ATB200 on LAMP1 appears to closely mimic the positive effects that were noted for glycogen levels using PAS staining. Overall, the histology data are in alignment with the biochemical assessment of glycogen reduction.

Taken together, these data demonstrate that the higher M6P content of ATB200 enables better lysosomal targeting, which translates to greater glycogen reduction, and this effect can be further improved by the co-administration of ATB200 with AT2221, thus warranting clinical investigation of this next-generation treatment for Pompe disease.

6.4. Dose Rationale

6.4.1. Toxicology and Toxicokinetic Support for Proposed Doses

To support the safety of ATB200, two 6-month Good Laboratory Practice (GLP) repeat-dose toxicology studies were conducted with ATB200 by IV administration, one in rats and one in NHPs. To support the safety of ATB200 co-administered with AT2221, a 3-month GLP co-administration toxicity study was conducted in NHPs with ATB200 administered IV and AT2221 administered orally by nasogastric gavage.

Across all 3 studies, no gender differences in exposure were noted. For all tested doses, plasma ATB200 concentration was measurable to ≥ 24 hours after dosing, while plasma AT2221 concentration was measureable to 74.5 hours after dosing (evaluated only in the 13-week co-administration study). For ATB200, as expected, exposure was lower when determined according to active protein levels, compared to the exposure based on total protein; however, across all studies, exposure increased as the dose increased. Little to no accumulation of either ATB200 or AT2221 was observed upon repeat (once every other week) administration. Finally, in animals administered ATB200, high anti-drug antibodies were noted, and with a certain animals developing neutralizing antibodies. Importantly, anti-drug antibodies/neutralizing antibodies did not affect ATB200 exposure or ATB200 toxicokinetic parameters. Of note, co-administration of ATB200/AT2221 in NHPs resulted in decreased ATB200 clearance (CL) and increased plasma exposure by approximately 2-fold relative to ATB200 monotherapy.

No treatment-related overt signs of toxicity were noted in any of the 3 studies, nor were any adverse, treatment-related hematological or clinical chemistry findings observed. Slight changes in several clinical pathology parameters noted in the rat study (ie, increased white blood cells, lymphocytes, basophils, and calcium, and decreased glucose) for the mid and high dose animals that may have been related to treatment, but the changes were not considered adverse or toxicologically significant based on their slight magnitude, the lack of any corresponding effect on the animals' general condition, and the absence of correlated changes in histopathology. No treatment-related effects on the safety pharmacology parameters, organ weights, or gross and microscopic findings were observed in any of these studies. The no-observed-adverse-effects level (NOAEL) for the 2 repeat dose studies was 200 mg/kg, the highest dose tested. In rats, the $AUC_{0\text{-}t}$ and C_{max} (combined genders) were 21,150 µg•hr/mL and 5,490 µg/mL, respectively, on Day 169 (averaged total protein). In NHPs, the $AUC_{0\text{-}t}$ and C_{max} (combined genders) were 25,000 µg·hr/mL and 4,205 µg/mL, respectively, on Day 169 (averaged total protein). The NOAEL in the co-administration toxicity study in NHPs was 100 mg/kg for ATB200 and 175 mg/kg for AT2221, the highest doses tested. At the highest tested doses of ATB200/AT2221, the $AUC_{0\text{-}t}$ and C_{max} (combined genders) were 13,850 µg·hr/mL and 2,270 µg/mL, respectively, for ATB200 (Day 85; averaged total protein) and 216 µg·hr/mL and 22 µg/mL, respectively, for AT2221 (Day 85).

Two *in vitro* assays were conducted using the final clinical formulation of ATB200, an *in vitro* hemolysis assay and an *in vitro* flocculation assay, both using human whole blood. The assays assessed the hemolytic potential of ATB200 in human whole blood (hemolysis assay) and its compatibility with human serum and plasma (flocculation assay). The dose range tested in both assays spanned the expected C_{max} values for clinical IV administration 2 and 4 hours in duration. No evidence of hemolysis or flocculation was noted up to the highest concentration tested of 1,000 µg/mL, which is more than 2-fold higher than the anticipated C_{max} at the predicted therapeutic dose of 20 mg/kg.

6.4.2. Human Dosing Rationale

To predict ATB200 exposure in humans, a PK model assessing the concentration versus time profile of ATB200 activity was developed based on PK data collected in *Gaa* KO mice and NHPs (Section 6.4.1). The resulting 2-compartment model with linear elimination adequately characterized the concentration versus time profile of ATB200 activity for all dose levels across multiple animal species. The model included a theoretical allometric component accounting for species-specific differences in CL and the volume of distribution (V_d) to predict PK parameters and ATB200 exposure in humans. The PK model also integrated data for rhGAA activity following IV administration of 20 mg/kg alglucosidase alfa. Using the PK model, the predicted CL and V_d of ATB200 in a typical 70 kg human subject are 0.768 L/hr and 2.41 L, respectively. The CL of ATB200 is expected to be approximately 30% faster than the CL of alglucosidase alfa in humans. The faster CL of ATB200 likely reflects its preferential distribution and greater uptake at the site of efficacy resulting from the higher amount of M6P on ATB200, which optimizes drug targeting.

The predicted PK in humans following administration of a single 20 mg/kg IV dose of ATB200 over a 4-hour infusion is shown in Table 3.

Table 3: Predicted PK of Single-dose Administration of ATB200 in Humans

Parameter	Predicted Value
$AUC_{0-\infty}$ (mg·hr/L)	1822
C_{max} (mg/L)	423
t_{max} (hr)	4.0
$t_{1/2}$ (hr)	2.17

Abbreviations: $AUC_{0-\infty}$ = area under the plasma-drug concentration time curve from Time 0 extrapolated to infinity; C_{max} = maximum observed plasma concentration; IV = intravenous; PK = pharmacokinetic(s); t_{max} = time to reach the maximum observed plasma concentration; $t_{1/2}$ = half-life
Note: PK was predicted for 20 mg/kg of ATB200 administered as a single IV dose over a 4-hour infusion and reflects active protein levels.

The AUC_{0-t} and C_{max} values in humans based on total protein are expected to be similar to those based on rhGAA activity given the toxicokinetic results from the 6-month chronic toxicity study in NHPs. In this study, at the lowest dose tested of 30 mg/kg, similar to the anticipated therapeutic dose of 20 mg/kg, the AUC_{0-t} and C_{max} values for active and total protein were virtually identical (approximately 800 µg·hr/mL for AUC_{0-t} based on active and total protein and approximately 350 µg/mL for C_{max} based on active and total protein).

Predicted exposure in humans for single-dose administration of AT2221 is summarized in Table 4.

Table 4: Predicted Exposure for Single-dose Administration of AT2221 in Humans

Parameter	AT2221 Dose	
	130 mg	**260 mg**
$AUC_{0-\infty}$ (µg•hr/mL)	12.35	24.71
C_{max} (µg/mL)	1	2

Abbreviations: $AUC_{0-\infty}$ = area under the plasma concentration time curve from Time 0 extrapolated to infinity; C_{max} = maximum observed plasma concentration
Source: Zavesca (miglustat): New Drug Application 21-348: Clinical Pharmacology and Biopharmaceutics Review(s) 2003

6.4.2.1. Dose Selection for Co-administered ATB200/AT2221

The concentration versus response relationship in *Gaa* KO mice (Section 6.3.1) suggested that co-administration of 20 mg/kg ATB200 with 10 or 20 mg/kg of AT2221 both optimizes the stability of ATB200 activity in plasma and maximizes glycogen reduction in quadriceps. The observed AT2221/ATB200 ratio of 0.01159 (ie, 10 mg/kg AT2221) is expected to correspond to an approximate 260 mg dose of AT2221 in a typical 70 kg human subject based on the predicted mean concentrations of ATB200 and AT2221 in humans (as determined from the nonclinical PK model described in Section 6.4.2. Targeted ratios of 0.01 and 0.02 are expected to correspond to AT2221 doses of 233 and 466 mg, respectively, in a typical 70 kg human subject.

Based on an *in vitro* inhibition assay evaluating the inhibitory effect of AT2221 on ATB200 activity, an approximate 260 mg dose of AT2221 is expected to bind to and stabilize ATB200 in plasma for up to 18 hours, and inhibition of ATB200 activity in the lysosome is only expected to

last approximately 4 hours. The exact timing of AT2221 administration relative to the end of infusion of ATB200 can be determined based on the C_{max} of AT2221 of approximately 1.5 hours. Overall, the simulations suggest that 20 mg/kg of ATB200 co-administered with approximately 260 mg of AT2221 is likely to produce optimal pharmacological outcomes in subjects with Pompe disease.

A 14-day dosing interval is considered an adequate washout time and is greater than 5 times the predicted half-life for ATB200 in the circulation (2.17 hours, approximately 11 hours in total residence time in blood). The estimated half-life of ATB200 in lysosomes is greater than 7 to 10 days. Additionally, the known half-life of miglustat is approximately 6 to 7 hours in plasma. Therefore, it would be anticipated that AT2221 would be cleared from muscle tissues to concentrations below pharmacologically active levels after approximately 35 hours and well before the end of the ATB200/AT2221 dosing interval of 14 days (Zavesca Package Insert 2008).

6.4.2.2. Predicted Safety Margins for Co-administered ATB200/AT2221

The doses selected for the planned clinical studies are 5, 10, and 20 mg/kg ATB200 administered alone and 20 mg/kg ATB200 co-administered with 130 or 260 mg AT2221. Based on modeling and simulation, 20 mg/kg ATB200 is anticipated to result in a C_{max} of 423 µg/mL and an AUC of 1,822 µg·hr/mL (Pharsight 2015). Assuming linear PK, an initial dose of 5 mg ATB200 is anticipated to result in a C_{max} of approximately 100 µg/mL and an AUC of approximately 450 µg·hr/mL. Exposure (area under the plasma drug concentration-time curve from time zero extrapolated to infinite time; $AUC_{0-\infty}$) to AT2221 is anticipated to be approximately 12.35 µg·hr/mL following administration of 130 mg orally and 24.71 µg·hr/mL following administration of 260 mg orally, while C_{max} values are anticipated to be 1 and 2 µg/mL, respectively.

Predicted safety margins in humans for co-administered ATB200/AT2221 are summarized in Table 5.

Table 5: Predicted Safety Margins for Co-administered ATB200/AT2221 in Humans

Parameter	Safety Margin			
	ATB200 (5 mg/kg)	**ATB200 (20 mg/kg)**	**AT2221 (130 mg)**	**AT2221 (260 mg)**
AUC_{0-t} (µg·hr/mL) (fold increase)	31 to 55	8 to 14	20	10
C_{max} (µg/mL) (fold increase)	23 to 55	5 to 13	20	10

Abbreviations: AUC_{0-t} = area under the concentration time curve from Time 0 to the last measurable time point, t; C_{max} = maximum observed concentration
Note: Predicted safety margins were based on TK data determined from long-term toxicity studies in rats and NHPs and a 3-month co-administration ATB200/AT2221 study in NHPs (Section 6.4.1). Data from the 3-month co-administration ATB200/AT2221 study in NHPs were derived from the 100 mg/kg ATB200 with 175 mg/kg AT2221 dose group.

The predicted safety margins for ATB200 administered as a single agent and co-administered with AT2221 support the first-in-human (FIH) clinical study using the proposed clinical doses of 5, 10, and 20 mg/kg ATB200, and 130 and 260 mg AT2221.

Please refer to the ATB200 Investigator's Brochure for additional detail.

6.4.2.3. PK Data from the ATB200-02 Study

Preliminary data from ATB200-02, the current protocol regarding PK of ascending doses of ATB200, effect of AT2221 on the PK of ATB200 and PK of low and high dose of AT2221 are presented below.

6.4.2.4. GAA Protein Using Signature Peptide(s) T09/T50

Total GAA protein in plasma was measured by signature peptides T09 (primary) and T50. Generally, the PK of signature peptide T50 was similar to T09, and therefore confirmed the results of the assay.

In Cohort 1 (ERT-experienced LOPD subjects), total GAA protein by signature peptide T09 exposures (C_{max} and AUC) demonstrated greater than dose-proportional increases during Stage 1 (single-ascending doses of 5, 10, and 20 mg/kg ATB200). Mean AUC for the 20 mg/kg ATB200 alone dose level was similar to translational modeling in the *Gaa* KO mouse to human AUC at the 20 mg/kg dose level (1,822 µg·h/mL). Co-administration with 130 mg AT2221 resulted in a 19.6% increase in total GAA protein mean AUC. Co-administration with 260 mg AT2221 increased total GAA protein AUCs by 26.3%. These increases were mainly observed during the early terminal phase, as shown by observed increases in the alpha-half-life. Clearance of plasma total GAA protein decreased with dose escalation and remained stable when co-administered with AT2221.

Generally, the PK of 20 mg/kg ATB200 based on plasma total GAA protein was similar between single and multiple doses when co-administered with 130 mg AT2221, and between single and multiple doses when co-administered with 260 mg AT2221.

The plasma total GAA protein PK profile of Cohort 3 (ERT-naïve subjects with LOPD) was generally similar to Cohort 1.

Similar to total GAA protein, in Cohort 1 (ERT-experienced LOPD subjects), total GAA activity exposures (C_{max} and AUC) in plasma demonstrated greater than dose proportional increases during Stage 1 (single-ascending doses of 5, 10, and 20 mg/kg ATB200). However, co-administration with either the low or high dose of AT2221 did not result in any relevant change in total GAA activity mean AUC. Clearance of plasma total GAA activity decreased with dose escalation and remained stable when co-administered with AT2221. Steady-state volume of distribution was consistent across all treatments from approximately 4.5 to 5.9 L.

Generally, the PK of 20 mg/kg ATB200 based on plasma total GAA activity was similar between single and multiple doses when co-administered with 130 mg AT2221, and between single and multiple doses when co-administered with 260 mg AT2221.

The GAA activity PK profile of Cohort 3 (ERT-naïve subjects with LOPD) was generally similar to Cohort 1.

Plasma AT2221 exposures (C_{max} and AUC) are approximately dose proportional. The rate of absorption (t_{max}) values support dosing with AT2221 one hour before start of ATB200 infusion, so that peak AT2221 concentrations are attained mid-way into infusion. Terminal elimination half-life was consistent at approximately 5 to 6 hours, as was plasma clearance (approximately 10 L/h) and terminal volume of distribution (approximately 85 L).

Overall, the preliminary exposures of ATB200 and AT2221 from approximately 10 subjects with LOPD are well-within the limits of the established safety margins.

6.4.2.5. ATB200 Exposure Stopping Criteria

Toxicokinetic evaluations from studies conducted in the cynomolgus monkey and rat included both total GAA protein concentrations and GAA activity levels in plasma. Generally, total GAA protein concentrations were either similar to or greater than GAA activity levels. Safety margins were calculated from these toxicity studies based on total protein levels as toxicity and/or immunotoxicity can be manifested by both active and inactive protein. Therefore, total GAA protein was selected to establish ATB200 exposure limits for the clinical study. Three toxicology studies were conducted: two 6-month dose-escalation studies with ATB200 alone to a maximum dose of 200 mg/kg, one in the cynomolgus monkey and one in the rat, and one 13-week co-administration study in the cynomolgus monkey to a maximum dose of 100 mg/kg ATB200 co-administered with 175 mg AT2221. Exposures obtained at the highest dose levels utilized in these studies were NOAELs. Since the current study evaluates both ATB200 alone and co-administration with AT2221 (which may increase total GAA protein exposure in plasma relative to the same ATB200 dose alone), the 100 mg/kg ATB200 co-administered with 175 mg AT2221 C_{max} and AUC_{0-t} parameters were selected as the upper exposure limit for the clinical study. Additionally, the cynomolgus monkey is a more closely related species to humans than the rat and the only appropriate species to extrapolate potential effects of a biologic, such as ATB200, to humans. The mean Day 85 exposure at this co-administration dose level for ATB200 was 2,270 µg/mL (C_{max}), and 13,850 µg·hr/mL (AUC_{0-t}). In the unlikely event a subject achieves one or both of these exposure levels, dosing with ATB200 alone or when co-administered with AT2221 will be stopped.

6.4.2.6. ATB200 Preliminary Efficacy Data

The fourth interim analysis was conducted in September 2017 with data cut-off point in August 2017.

Adverse events (AEs) were generally mild and transient. Most common AEs reported as treatment related were nausea (3/20), tremor (3/20), headache (3/20), fatigue (3/20), diarrhea (2/20), muscle spasm (2/20), and joint swelling (2/20). Three incidents of infusion-associated reactions (IARs) in 400^{+} infusions which were controlled by standard premedication. One IAR event occurred in one nonambulatory ERT-switch patient (skin discoloration). Two IAR events in an ERT-naïve patient (localized pruritus, erythema and burning sensation). Longest duration of treatment was 72 weeks.

Improvement in biomarkers of muscle damage (creatine phosphokinase [CK], alanine aminotransferase [ALT], and aspartate aminotransferase [AST]) and disease substrate (hexose tetrasaccharide [Hex4]) was observed for up to 58 Weeks on ATB200/AT2221 in all three cohorts.

Six-minute walk distance improved in Cohorts 1 and 3 as shown in Table 6. Qualitative and quantitative upper extremity muscle strength improved in Cohort 2 subjects. Forced vital capacity was stable in Cohort 1 and improved in Cohort 3 as shown in Table 7.

Table 6: Change in 6MWT Distance in Meters in Cohorts 1 and 3 Subjects Through Month 18

Cohort	Mean (SD)	Mean (SD) Change from Baseline			
	Baseline	Month 6	Month 9	Month 12	Month 18
1[a] ERT-experienced (2 to 6 years)	(n = 10) **397.2** (96.8)	(n = 10) **+23.9** (52.2)	(n = 10) **+24.5** (40.8)	(n = 10) **+42.2** (46.5)	(n = 9) **+51.7** (45.9)
3 ERT-naïve	(n = 5) **399.5** (83.5)	(n = 5) **+41.8** (29.4)	(n = 5) **+63.5** (23.1)	(n = 5) **+63.1** (29.1)	(n = 5) **+49.0** (28.3)

Abbreviations: 6MWT = 6-minute walk test; ERT = enzyme replacement therapy; n = number of patients; SD = standard deviation

[a] For the purposes of assigning nominal time points to data from Cohort 1, the 6-week duration of Stage 1 (single ascending doses of miglustat) and the 12-week duration of Stage 2 (multiple ascending doses of miglustat [130 mg for 6 weeks, followed by 260 mg for 6 weeks]) were included. Therefore, the Month 6, 9, 12, and 18 time points reflect cumulative data to Stage 3 Month 3, Stage 3 Month 6, Stage 3 Month 9, and Stage 3 Month 15, respectively.

Source: Interim Analysis 6 (data cut-off date of August 2018), Table 14.2.1.3

Table 7: Change in Percentage Predicted Sitting FVC (%) in Cohorts 1 and 3 Subjects Through Month 18

Cohort	Mean (SD)	Mean (SD) Change from Baseline			
	Baseline	Month 6	Month 9	Month 12	Month 18
1[a] ERT-experienced (2 to 6 years)	(n = 9) **52.6** (14.7)	(n = 9) **–1.3** (4.1)	(n = 9) **–1.7** (3.9)	(n = 9) **–3.3** (6.1)	(n = 8) **–4.0** (7.0)
3 ERT-naïve	(n = 5) **53.4** (20.3)	(n = 5) **+4.2** (5.6)	(n = 5) **+6.2** (5.3)	(n = 5) **+4.4** (8.7)	(n = 5) **+6.8** (2.5)

Abbreviations: ERT = enzyme replacement therapy; FVC = forced vital capacity; n = number of patients; SD = standard deviation

[a] For the purposes of assigning nominal time points to data from Cohort 1, the 6-week duration of Stage 1 (single ascending doses of miglustat) and the 12-week duration of Stage 2 (multiple ascending doses of miglustat [130 mg for 6 weeks, followed by 260 mg for 6 weeks]) were included. Therefore, the Month 6, 9, 12, and 18 time points reflect cumulative data to Stage 3 Month 3, Stage 3 Month 6, Stage 3 Month 9, and Stage 3 Month 15, respectively.

Source: Interim Analysis 6 (data cut-off date of 03 August 2018), Table 14.2.1.1

7. OBJECTIVES AND PURPOSE

7.1. Primary Objectives

- To evaluate the safety and tolerability of single-ascending doses of IV infused ATB200
- To evaluate the safety and tolerability of single-ascending doses of IV infused ATB200 as a fixed dose, co-administered with ascending oral doses of AT2221
- To characterize the PK of single-ascending doses of IV infused ATB200
- To characterize the single- and multiple-dose PK of IV infused 20 mg/kg ATB200 when co-administered with oral 130 mg or 260 mg AT2221
- To characterize the PK of single and multiple oral doses of 130 mg or 260 mg AT2221 when co-administered with IV infused ATB200

7.2. Secondary Objectives

- To evaluate the long-term efficacy of 20 mg/kg of IV infused ATB200 as a fixed dose co-administered with oral 260 mg AT2221 in all subjects from Stage 3
- To evaluate the long-term safety and tolerability of 20 mg/kg of IV infused ATB200 as a fixed dose co-administered with oral 260 mg AT2221 in all subjects from Stage 3
- To characterize single- and multiple-dose PK of plasma rhGAA activity and total rhGAA protein following IV infused 20 mg/kg ATB200 as a fixed dose co-administered with oral 260 mg AT2221 in enzyme replacement therapy (ERT)-naïve subjects
- To characterize the single- and multiple-dose PK of plasma AT2221 following 20 mg/kg of IV infused ATB200 co-administered with oral 260 mg AT2221 in ERT-naïve subjects

7.3. Exploratory Objectives

To evaluate:

- Anti-rhGAA antibodies (total and neutralizing)
- Cross-reactivity of anti-rhGAA antibodies to alglucosidase alfa
- Pro-inflammatory cytokines and other biomarkers of immune system activation
- PD biomarkers (urine hexose tetrasaccharide [Hex4] and serum creatine phosphokinase [CK])
- Impact of anti-rhGAA antibodies on plasma GAA total protein exposures

8. INVESTIGATIONAL PLAN

8.1. Study Design

This is an open-label, fixed-sequence, single- and multiple-ascending dose, FIH study to evaluate the safety, tolerability, PK, PD, and efficacy of IV ATB200 alone and when co-administered with oral AT2221. The study will be conducted in 4 stages.

Stages 1 and 2: Approximately 10 to 12 ERT-experienced ambulatory subjects (Cohort 1) will be enrolled. In Stage 1, safety, tolerability, and PK will be evaluated following sequential single-ascending doses of intravenously infused ATB200 for 3 dosing periods at 5, 10, and 20 mg/kg.

In Stage 2, safety, tolerability, and PK will be evaluated following single- and multiple-ascending dose combinations: 20 mg/kg of IV ATB200 co-administered with 130 mg of AT2221 administered orally every 14 (± 3 days) for 3 doses, followed by 20 mg/kg of IV ATB200 co-administered with 260 mg of AT2221 administered orally for 3 doses, as outlined in Table 8. The dosing interval of 14 days, based on the current standard of care, is estimated to provide adequate duration for complete washout (greater than 5 half-lives) of ATB200 (predicted half-life of 2.17 hours) and AT2221 (half-life of 6 to 7 hours). Cohort 1 subjects who have completed Stages 1 and 2 will enter into a long-term extension stage of the study, hereinafter referred to as Stage 3, and will continue to be assessed for safety, tolerability, and efficacy of long-term co-administration of ATB200 and AT2221. In addition, functional assessments relevant to Pompe disease will be performed at regular intervals per the Schedule of Assessments (Table 9, Table 10, Table 11, Table 12, and Table 13).

Stage 3: Approximately 12 to 18 additional subjects will enroll, approximately 4 to 6 of whom will be ERT-experienced nonambulatory subjects (Cohort 2), approximately 5 of whom will be ERT-naïve ambulatory subjects (Cohort 3) and approximately 6 to 8 of whom will be ambulatory subjects with ERT experience of at least 7 years (Cohort 4). Single- and multiple-dose serial PK sampling will be conducted for Cohort 3 subjects but not for Cohort 2 or 4 subjects. Many nonambulatory subjects lack contralateral venous access as a result of sclerosis within peripheral veins. In addition, the intensive PK sampling in this study is likely to present an undue burden to nonambulatory subjects. For these reasons, no PK assessments will be conducted in nonambulatory subjects. PK analysis will not be conducted for Cohort 4 due to the similarity of patient populations in Cohorts 1 and 4, and the well-characterized PK in Cohort 1. Stage 3 will continue for a period of two years for Cohorts 1, 3 and 4.

Stage 4: Subjects who complete Stage 3 will enter into Stage 4. Stage 4 treatment period will be continued as open-label extension and will continue until the date of regulatory approval or marketing authorization and/or commercialization in the participating subject's country, or study termination by the sponsor, Amicus Therapeutics, Inc. (Amicus).

In all stages, every effort should be made to maintain the duration of infusion of ATB200 at 4-hours (± 15 min). Changes in the duration of ATB200 infusion due to safety or tolerability issues will be documented.

In this study, ERT-experienced ambulatory subjects are defined as adults with Pompe disease who have been on ERT for 2 to 6 years (Cohort 1) and ≥ 7 years (Cohort 4) prior to enrollment and who are able to walk at least 200 meters in the 6MWT.

ERT-experienced nonambulatory subjects (Cohort 2), are defined as adults with Pompe disease who are wheelchair bound and unable to walk unassisted, have been on ERT (alglucosidase alfa) for ≥ 2 years prior to enrollment.

ERT-naïve ambulatory subjects (Cohort 3), are defined as adults with Pompe disease who have never received treatment with ERT for Pompe disease and who are able to walk at least 200 meters in the 6MWT.

If a subject prematurely withdraws from the study, the subject may be replaced with a subject who qualifies for any cohort. If any of the sentinel subjects withdraw prematurely from the study, that subject will be replaced by the next subject enrolled in that cohort (eg, if Subject 1 withdraws, Subject 3 will replace that subject as a sentinel subject).

The study is composed of a Screening Visit, a Baseline Visit, Stage 1 (3-period, fixed-sequence, single-ascending dose PK study of ATB200 alone), Stage 2 (2-period, fixed-sequence, single- and multiple-dose PK study of 20 mg/kg ATB200 co-administered with multiple-ascending doses of AT2221), and Stage 3 and Stage 4 treatment period will be continued as open-label extension and will continue until the date of regulatory approval or marketing authorization and/or commercialization in the participating subject's country, or study termination by Amicus. Additional serial 24-hour PK visits are included in Stage 3 for subjects in Cohort 3. If a subject is withdrawing from the study, they will return for an Early Termination (ET) visit within 30 days of their last visit.

The assessments to be performed at each study visit are described in Table 9 for all subjects in Stages 1 and 2, and Table 10, Table 11, Table 12, and Table 13 for all subjects in Stage 3 and 4.

8.2. Safety Monitoring and Sentinel Dosing

Safety will be monitored by the Amicus Medical Monitor and the investigators on a continuous basis and on a regular basis by a Safety Steering Committee (SSC). The details of the membership, meeting schedules, procedures, roles, and responsibilities of the SSC are detailed in the SSC Charter.

Study treatment may be interrupted or permanently stopped by the Amicus Medical Monitor and the SSC, if warranted by the safety and tolerability data. Subjects will be monitored in a hospital or infusion center for 48 hours from the start of each infusion during Stage 1 (ie, ERT dose-escalation stage). Since the target infusion duration in the study is 4 hours, all subjects will be closely monitored for the first 4 hours during their infusion and then observed in hospital for an additional 44 hours prior to discharge. Subjects will be admitted and dosed at either an infusion center or a hospital and thereafter hospitalized for the remainder of the observation period.

In Stages 2 and 3, all subjects will be monitored at a hospital or infusion center for an additional 2 to 4 hours from the end of their infusion. Once a subject has completed one year in Stage 3, the subjects, who have not experienced IARs for prior 6 months, can be monitored for one hour post infusion at the PI's discretion. Subjects who switch to home infusion, must be monitored for 2 to

4 hours for a period of at least 3 months. If no IARs occur, the monitoring can be reduced to one hour post infusion at the PIs discretion. Refer to Section 11.1.3 for further information on IARs, including their reporting and management.

When at least one of the 2 Cohort 1 sentinel subjects has completed Period 5 dosing, and the safety data has been reviewed by the SSC, subjects from Cohorts 2 and 3 can begin enrollment into Stage 3. The first 2 subjects from Cohorts 2 and 3 will undergo sentinel dosing in the same manner as subjects from Cohort 1 during Stage 2.

Sentinel Dosing

The first 2 subjects in Cohort 1 of this study will be the sentinel subjects for the study and will be the first 2 subjects dosed in each period of the study (Periods 1 to 5). The first 2 subjects dosed in each cohort will be the sentinel subjects for that cohort. In the event that a sentinel subject is prematurely withdrawn from the study, he/she will be replaced by another subject.

Note: At least one of the 2 sentinel subjects will complete Period 5, Stage 2 dosing, and the safety data will be reviewed by the SSC, before any newly enrolled subjects in Cohorts 2 and 3 can be dosed. The first 2 subjects in Cohorts 2 and 3 will also serve as sentinel subjects for their respective cohorts.

In Stage 1 (Periods 1, 2, and 3), subjects will be dosed with single-ascending doses of ATB200 (5 mg/kg [Period 1], 10 mg/kg [Period 2], and 20 mg/kg [Period 3]).

Following the dosing of the 2 sentinel subjects for each study period in Stage 1, an evaluation of the available safety data (physical examination [PE], vital signs, adverse events [AEs], infusion reactions, electrocardiogram [ECG], and available locally performed laboratory tests) will be performed within 24 to 48 hours by the Amicus Medical Monitor and the investigators. The SSC will convene for a formal safety review when central safety laboratory data are available for both sentinel subjects at each dose level. If the SSC determines that there are no safety concerns that preclude dosing at the dose assigned for that period, 10 additional subjects will be enrolled and dosed.

Note: Vital signs will be monitored as detailed in Table 9, Table 10, Table 11, Table 12, and Table 13. For PK Visits 3, 4, 5, 6, and 9, vital signs will be monitored at hours 0, 1, 4, 6, 12, and 24 after the start of infusion. During the 48-hour observation period, vital signs will be monitored, at a minimum, every 6 to 8 hours until the end of the 48-hour observation period.

The SSC will also convene for a safety review when safety data (including central laboratory safety data) for all subjects at all three Stage 1 dose levels are available.

In Stage 2 (after Visits 6 and 9), safety data from the 2 sentinel subjects will be reviewed by the SSC, and safety will be assessed after the first dose as for each period in Stage 1. If the SSC determines that there are no safety concerns that preclude additional dosing at 20 mg/kg ATB200 co-administered with 130 mg AT2221 (Visit 6) or 20 mg/kg ATB200 co-administered with 260 mg AT2221 (Visit 9), the remaining subjects in Cohort 1 will receive Stage 2 treatment.

Stage 3 sentinel dosing will be performed for the first 2 subjects in Cohort 2 and the first 2 subjects in Cohort 3. If the SSC determines that there are no safety concerns that preclude the additional dosing of 20 mg/kg of ATB200 co-administered with 260 mg of AT2221, the remaining 2 to 4 additional subjects in Cohort 2 and the remaining 2 to 4 additional subjects in Cohort 3 will also be dosed.

Subjects from Cohorts 2 and 3 will not begin dosing until at least one of the 2 sentinel subjects in Cohort 1 have completed Period 5, Stage 2 dosing and the review of safety data has been completed by the SSC.

The SSC will reconvene when all safety data (including central safety laboratory data) are available for all subjects at the end of Stage 2.

The SSC will also convene ad hoc in case of a Suspected Unexpected Serious Adverse Reaction (SUSAR) or an identified safety concern.

The SSC may recommend any of the following review:

- Continue the study without modifications
- Continue the study with modifications (amendment)
- Temporarily halt dosing
- Permanently stop dosing

If in the opinion of the SSC there are no AEs or safety concerns in the sentinel subjects that might preclude continued study dosing, dosing will continue for all remaining subjects at that dose level.

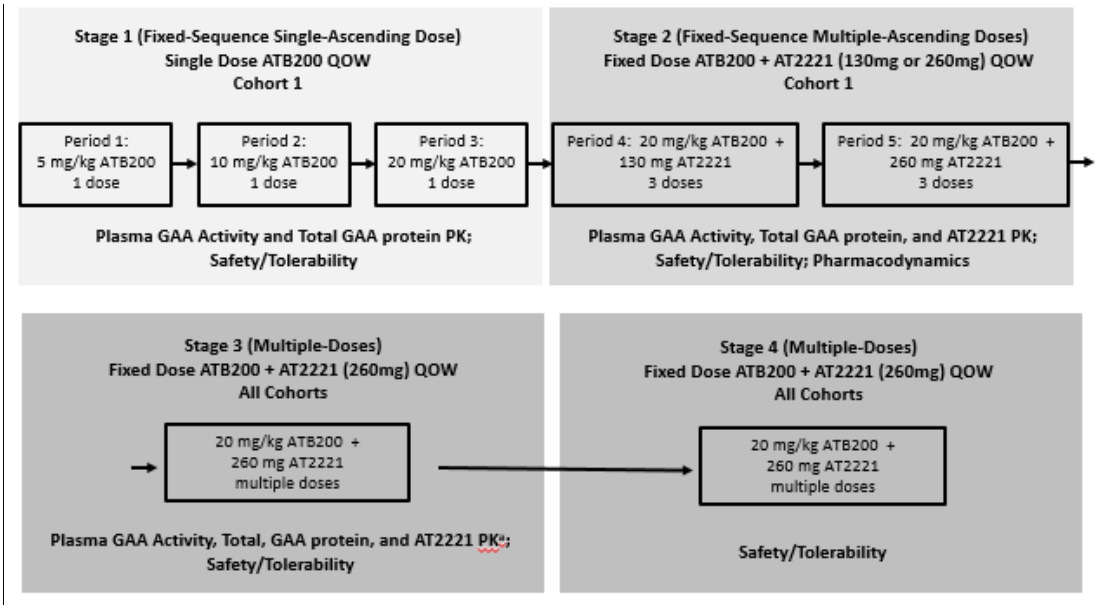
Subject safety will continue to be closely monitored by the Amicus Medical Monitor and study investigators on an ongoing basis, and at regular intervals by the SSC.

8.3. Pharmacokinetic Sampling Details

The PK assessments in this study include blood sampling for plasma GAA activity levels, plasma total GAA protein concentrations, and plasma AT2221 concentrations.

PK assessments will be performed as described in Table 9, Table 12, Table 14, and Table 15.

Figure 1: Study Design Schematic



Abbreviations: GAA = acid α-glucosidase; PK = pharmacokinetics; QOW = every other week
[a] PK assessments for GAA activity, total GAA protein and AT2221 in Stage 3 will only be performed for the first and third doses for Cohort 3. Sparse blood sampling for total GAA protein will be performed after at least 18 months of treatment in Cohorts 1 and 3. If the subject has completed Stage 3, sparse PK sampling will be performed in Stage 4.

8.4. Details of Study Treatment

Cohort 1 subjects will be administered ATB200 as a single agent and ATB200 co-administered with AT2221 according to the treatment assignment in Table 8. Cohorts 2, 3, and 4 will only receive ATB200 co-administered with AT2221 according to the treatment assignment in Table 8.

Table 8: Treatment Assignment for Stages 1, 2, 3, and 4

Cohorts	Stage 1 (6 weeks)			Stage 2 (12 weeks)		Stage 3 (2 years)	Stage 4 (Until Approval)
	Period 1 Single-Dose	Period 2 Single-Dose	Period 3 Single-Dose	Period 4 3 Multiple Doses Co-administration	Period 5 3 Multiple Doses Co-administration	Multiple Dose, 24 months, Extension Co-administration	Multiple Dose, Long-term, Extension Co-administration
Cohort 1 (n = 11)	5 mg/kg ATB200	10 mg/kg ATB200	20 mg/kg ATB200	20 mg/kg ATB200 + 130 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221
Cohort 2 (n = 4-6)	NA					20 mg/kg ATB200 + 260 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221

Table 8: Treatment Assignment for Stages 1, 2, 3, and 4 (Continued)

Cohorts	Stage 1 (6 weeks)			Stage 2 (12 weeks)		Stage 3 (2 years)	Stage 4 (Until Approval)
	Period 1 Single-Dose	Period 2 Single-Dose	Period 3 Single-Dose	Period 4 3 Multiple Doses Co-administration	Period 5 3 Multiple Doses Co-administration	Multiple Dose, 24 months, Extension Co-administration	Multiple Dose, Long-term, Extension Co-administration
Cohort 3 (n = 5)	NA					20 mg/kg ATB200 + 260 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221
Cohort 4 (n = 6-8)	NA					20 mg/kg ATB200 + 260 mg AT2221	20 mg/kg ATB200 + 260 mg AT2221

Abbreviations: NA = not applicable; SSC = Safety Steering Committee
Note: At least one of the 2 sentinel subjects will complete Period 5, Stage 2 dosing, and the safety data will be reviewed by the SSC, before any newly enrolled subjects in Cohorts 2 and 3 can be dosed. The first 2 subjects in Cohorts 2 and 3 will also serve as sentinel subjects for their respective cohorts.

8.5. Stopping Criteria

Subjects may be discontinued from study medication and withdrawn from the study if they meet any of the following stopping criteria:

- Any moderate or severe AE that is at least probably related to study treatment and that poses a significant safety risk to the subject, in the opinion of any one of the following: the principal investigator, Amicus Medical Monitor, or SSC
- A serious adverse event (SAE) that is deemed by the investigator or Amicus Medical Monitor to be at least probably related to study treatment
- Intolerable AE, in the opinion of the investigator

Subjects must discontinue study medication and be withdrawn from the study if they meet any of the following stopping criteria:

- Anaphylactic reaction to study treatment, as defined by the Sampson Criteria (Appendix 1)
- SSC decision based on safety data review
- Any of the following abnormal liver function tests in the absence of a reason other than study treatment, to explain the abnormalities
  - Alanine aminotransferase (ALT) or aspartate aminotransferase (AST) > 8 × upper limit of normal (ULN)
  - ALT or AST > 5 × ULN for more than 2 weeks
  - ALT or AST > 3 × ULN and (total bilirubin > 2 × ULN)

  - ALT or AST > 3 × ULN with the appearance of fatigue, nausea, vomiting, right upper quadrant pain or tenderness, fever, rash, and/or eosinophilia (> 5%)
- Pregnancy

Adverse events that meet the stopping criteria and are reported as serious or severe (per the intensity scale used in Section 11.3), will also be graded using the version of the Common Terminology Criteria for Adverse Events (CTCAE) that is current at study initiation.

Adverse events that meet the stopping criteria will be considered stopping events if they are:

- Deemed related to study treatment
- Classified as Grade ≥ 3 based on the current version of the CTCAE
- In any of the following System Organ Classes:
  - Hepatobiliary Disorders
  - Injury, Poisoning and Procedural Complications
  - Blood and Lymphatic System Disorders
  - Cardiac Disorders
  - Renal and Urinary Disorders
  - Nervous System Disorders

8.6. Concomitant Medications and Nondrug Therapies

Concomitant medications taken within 4 weeks prior to the Screening Visit or at any time throughout the study must be recorded in the electronic case report forms (eCRFs).

Concomitant medications and nondrug therapies (eg, procedures, surgery, physical therapy, assistive devices, etc) specifically associated with an AE or IAR should be entered into the corresponding eCRFs.

8.7. Prohibited Medications

All prohibited medications must be discontinued at least 30 days before the Baseline Visit.

Use of the following medications by all subjects is prohibited during this study:

- miglitol (eg, Glyset®)
- non-study miglustat (eg, Zavesca)
- Myozyme®/Lumizyme® after treatment with ATB200 has begun
- acarbose (eg, Precose®, Glucobay®)
- voglibose (eg, Volix®, Vocarb®, and Volibo®)

Note: None of these medications have a half-life that, when multiplied by 5, is longer than 30 days.

- oral β_2-receptor agonists and non-selective β-blockers (eg, propranolol, nadalol and carvedilol). In situations in which the subject has been on a stable dose of a non-selective β-blocker or has a medical need to be started on that medication, and it is deemed the most appropriate medication for the subject by the PI or by the primary treating physician, subject may be treated with that medication. This decision will be made by the PI in discussion with Amicus Medical Monitor.
- any investigational/experimental drug

8.8. Criteria for Termination of the Study

The study may be terminated by the sponsor for any of the following reasons:

- evidence suggesting that safety risks associated with ATB200/AT2221 treatment outweigh the potential benefits or at the recommendation of the Safety Steering Committee (SSC) (Section 14.3)
- lack of efficacy
- inability to enroll the targeted number of subjects

9. SELECTION AND WITHDRAWAL OF SUBJECTS

9.1. Number of Subjects

Approximately 18 to 34 subjects will enroll in the study. Approximately 10 to 12 subjects, are planned to be enrolled in Cohort 1 of this study, and these subjects will participate in Stage 1 and 2. Stage 3 of the study will continue to treat subjects from Cohort 1 and will also enroll another 12 to 18 subjects approximately; 4 to 6 subjects from Cohort 2; 5 subjects from Cohort 3; and approximately 6 to 8 from Cohort 4.

Subjects who withdraw prematurely from this study may be replaced by subjects eligible for any cohort. If one of the 2 sentinel subjects withdraws prematurely from the study, that subject will be replaced by the next subject enrolled in the study (eg, if Subject 1 withdraws, Subject 3 will replace that subject as a sentinel subject).

9.2. Eligibility Criteria

During the Screening Visits, subjects with Pompe disease will be evaluated against the eligibility criteria given below, as appropriate. Separate sets of criteria exist for ERT-experienced or ERT-naïve ambulatory subjects and ERT-experienced nonambulatory subjects. Each subject must meet all of the inclusion criteria and none of the exclusion criteria. Waivers of inclusion/exclusion criteria are not permitted. Subjects who initially screen fail may be rescreened.

9.2.1. Inclusion and Exclusion Criteria

Cohort 1

Inclusion Criteria

1. Male and female subjects between 18 and 65 years of age, inclusive
2. Subject must provide signed informed consent prior to any study-related procedures
3. Subjects of childbearing potential must agree to use medically accepted methods of contraception during the study and for 90 days after last co-administration of ATB200 and AT2221
4. Subject has a diagnosis of Pompe disease based on documented deficiency of GAA enzyme activity or by *GAA* genotyping
5. Subject has received ERT with alglucosidase alfa (Myozyme/Lumizyme) for the previous 2 to 6 years, inclusive
6. Subject is currently receiving alglucosidase alfa (Myozyme/Lumizyme) at a frequency of once every other week
7. Subject has received and completed the last 2 infusions without a drug-related adverse event resulting in dose interruption
8. Subject must be able to walk between 200 and 500 meters on the 6MWT
9. Upright FVC must be 30% to 80% of predicted normal value

Exclusion Criteria

1. Subject has received any investigational therapy including adjunctive therapy for Pompe disease, other than alglucosidase alfa within 30 days or 5 half-lives of the therapy or treatment, whichever is longer, prior to the Baseline Visit, or anticipates doing so during the study
2. Subject has received treatment with prohibited medications (see Section 8.7) within 30 days of the Baseline Visit
3. Subject, if female, is pregnant or breastfeeding at screening
4. Subject, whether male or female, is planning to conceive a child during the study
5. Subject requires invasive ventilatory support
6. Subject uses noninvasive ventilatory support ≥ 6 hours a day while awake
7. Subject has a medical or any other extenuating condition or circumstance that may, in the opinion of the investigator or the Medical Monitor, pose an undue safety risk to the subject or compromise his/her ability to comply with protocol requirements
8. Subject has a history of anaphylaxis to alglucosidase alfa
9. Subject has a history of high sustained anti-rhGAA antibodies (see Section 10.4)
10. Subject has a history of allergy or sensitivity to miglustat or other iminosugars
11. Subjects with active systemic autoimmune disease such as lupus, scleroderma, or rheumatoid arthritis; all subjects with autoimmune disease must be discussed with the Amicus Medical Monitor
12. Subjects with active bronchial asthma; all subjects with bronchial asthma must be discussed with the Amicus Medical Monitor

Cohort 2

Inclusion Criteria

10. Male and female subjects between 18 and 65 years of age, inclusive
11. Subject must provide signed informed consent prior to any study-related procedures
12. Subjects of childbearing potential must agree to use medically accepted methods of contraception during the study and for 90 days after last co-administration of ATB200 and AT2221
13. Subject has a diagnosis of Pompe disease based on documented deficiency of GAA enzyme activity or by *GAA* genotyping
14. Subject has received ERT with alglucosidase alfa (Myozyme/Lumizyme) for ≥ 2 years
15. Subject is currently receiving alglucosidase alfa (Myozyme/Lumizyme) at a regular or set frequency
16. Subject has received and completed the last 2 infusions without a drug-related adverse event resulting in dose interruption

17. Subject must be wheelchair-bound and unable to walk unassisted

Exclusion Criteria

13. Subject has received any investigational therapy including adjunctive therapy for Pompe disease, other than alglucosidase alfa within 30 days or 5 half-lives of the therapy or treatment, whichever is longer, prior to the Baseline Visit, or anticipates to do so during the study
14. Subject has received treatment with prohibited medications (see Section 8.7) within 30 days of the Baseline Visit
15. Subject, if female, is pregnant or breastfeeding at screening
16. Subject, whether male or female, is planning to conceive a child during the study
17. Subject has a medical or any other extenuating condition or circumstance that may, in the opinion of the investigator or the Medical Monitor, pose an undue safety risk to the subject or compromise his/her ability to comply with protocol requirements
18. Subject has a history of anaphylaxis to alglucosidase alfa
19. Subject has a history of high sustained anti-rhGAA antibodies (see Section 10.4)
20. Subject has a history of allergy or sensitivity to miglustat or other iminosugars
21. Subjects with active systemic autoimmune disease such as lupus, scleroderma, or rheumatoid arthritis; all subjects with autoimmune disease must be discussed with the Amicus Medical Monitor
22. Subjects with active bronchial asthma; all subjects with bronchial asthma must be discussed with the Amicus Medical Monitor

Cohort 3

Inclusion Criteria

18. Male and female subjects between 18 and 65 years of age, inclusive
19. Subject must provide signed informed consent prior to any study-related procedures
20. Subjects of childbearing potential must agree to use medically accepted methods of contraception during the study and for 90 days after last co-administration of ATB200 and AT2221
21. Subject has a diagnosis of Pompe disease based on documented deficiency of GAA enzyme activity or by *GAA* genotyping
22. Subject must be able to walk between 200 to 500 meters on the 6MWT
23. Upright FVC must be 30% to 80% of predicted normal value

Exclusion Criteria

23. Subject has received any ERT, including alglucosidase alfa at any time, or any investigational therapy for Pompe disease within 30 days or 5 half-lives of the therapy or treatment, whichever is longer, prior to the Baseline Visit, or anticipates doing so during the study

24. Subject has received treatment with prohibited medications (see Section 8.7) within 30 days of the Baseline Visit
25. Subject, if female, is pregnant or breastfeeding at screening
26. Subject, whether male or female, is planning to conceive a child during the study
27. Subject requires invasive ventilatory support
28. Subject uses noninvasive ventilatory support ≥ 6 hours a day while awake
29. Subject has a medical or any other extenuating condition or circumstance that may, in the opinion of the investigator or the Medical Monitor, pose an undue safety risk to the subject or compromise his/her ability to comply with protocol requirements
30. Subject has a history of allergy or sensitivity to miglustat or other iminosugars
31. Subjects with active systemic autoimmune disease such as lupus, scleroderma, or rheumatoid arthritis; all subjects with autoimmune disease must be discussed with the Amicus Medical Monitor
32. Subjects with active bronchial asthma; all subjects with bronchial asthma must be discussed with the Amicus Medical Monitor

Cohort 4

Inclusion Criteria

24. Male and female subjects between 18 and 75 years of age, inclusive
25. Subject must provide signed informed consent prior to any study-related procedures
26. Subject has documented 6MWT on three separate occasions, each at least six months apart with at least two values in the past three years
27. Subjects of childbearing potential must agree to use medically accepted methods of contraception during the study and for 90 days after last co-administration of ATB200 and AT2221
28. Subject has a diagnosis of Pompe disease based on documented deficiency of GAA enzyme activity or by *GAA* genotyping
29. Subject has received ERT for the previous ≥ 7 years
30. Subject is currently receiving alglucosidase alfa (Myozyme/Lumizyme) at a frequency of once every other week
31. Subject has received and completed the last 2 infusions without a drug-related AE resulting in dose interruption
32. Subject must be able to walk between 75 and 600 meters on the 6MWT
33. Upright FVC must be 30% to 85% of predicted normal value

Exclusion Criteria

33. Subject has received any investigational therapy including adjunctive therapy for Pompe disease, other than alglucosidase alfa within 30 days, or 5 half-lives of the therapy or

treatment, whichever is longer, prior to the Baseline Visit, or anticipates doing so during the study

34. Subject has received treatment with prohibited medications (see Section 8.7) within 30 days of the Baseline Visit
35. Subject, if female, is pregnant or breastfeeding at screening
36. Subject, whether male or female, is planning to conceive a child during the study
37. Subject requires invasive ventilatory support
38. Subject uses noninvasive ventilatory support ≥ 6 hours a day while awake
39. Subject has a medical or any other extenuating condition or circumstance that may, in the opinion of the investigator or the Medical Monitor, pose an undue safety risk to the subject or compromise his/her ability to comply with protocol requirements
40. Subject has a history of anaphylaxis to alglucosidase alfa
41. Subject has a history of high sustained anti-rhGAA antibodies (see Section 10.4)
42. Subject has a history of allergy or sensitivity to miglustat or other iminosugars
43. Subjects with active systemic autoimmune disease such as lupus, scleroderma, or rheumatoid arthritis; all subjects with autoimmune disease must be discussed with the Amicus Medical Monitor
44. Subjects with active bronchial asthma; all subjects with bronchial asthma must be discussed with the Amicus Medical Monitor

9.3. Withdrawal Criteria

Subjects may discontinue study treatment, withdraw from the study, or be withdrawn from the study for any reason including, but not limited to, the following:

- at their own request or at the request of their legally authorized representative*
- if, in the investigator's opinion, continuation in the study would be detrimental to the subject's well-being
- occurrence of an intolerable treatment-emergent adverse event (TEAE) as determined by the investigator and/or the subject
- failure of the subject to return to the study site for scheduled visits
- persistent noncompliance
- sponsor decision to terminate the study

*"Legally authorized representative" means an individual or judicial or other body authorized under applicable law to consent on behalf of a prospective subject to the subject's participation in the procedure(s) involved in the research.

Pregnancy

Any subject who becomes pregnant during the study **must be withdrawn** from study treatment and will be followed through the outcome of the pregnancy.

In all cases, the reason for and date of withdrawal must be recorded in the eCRF and in the subject's medical records. The subject must be followed up to establish whether the reason was an AE and, if so, the AE must be reported as described in Section 11.4. All subjects who discontinue study treatment will be encouraged to return for an ET Visit.

In case the subject wishes to discontinue treatment, the investigator must inquire and document in the medical record whether:

- the subject only wants to discontinue study treatment but agrees to the follow-up procedures as outlined in the protocol
- the subject wants to discontinue study treatment and all follow-up procedures
- the subject wants to revoke the consent to collect and use further data

Note: In the US, the authorization to use and disclose data for research can only be revoked in writing by the subject.

The investigator must make every effort to contact subjects who discontinue study treatment or visits or are lost to follow-up and schedule the ET assessments. Attempts to contact subjects who are lost to follow-up (eg, times and dates of attempted telephone contact, receipt for sending a registered letter) must be documented in the subject's source document.

9.4. Replacement of Subjects

Subjects who withdraw or are withdrawn from the study may be replaced. If one of the 2 sentinel subjects withdraws prematurely from the study, that subject will be replaced by the next subject enrolled in the study (eg, if Subject 1 withdraws, Subject 3 will replace that subject as a sentinel subject).

Replacement of a subject may only occur after consultation with, and written approval of, the Amicus Medical Monitor.

9.5. Subjects of Reproductive Potential

The allowed methods of contraception described in the following text are only effective when used consistently, correctly, and in accordance with the product label. The investigator is responsible for ensuring subjects understand how to properly use these methods of contraception. A highly effective birth control is defined as one that results in a low failure rate (ie, < 1% per year) when used consistently and correctly.

Subjects must be willing to apply highly effective contraception during the study and through the duration as defined below and for 90 days after the final dose of study treatment.

Woman of childbearing potential: defined as all women physiologically capable of becoming pregnant, must use highly effective contraception during the study and through 90 days after the study treatment. Highly effective contraception is defined as either:

- Total abstinence: When this is in line with the preferred and usual lifestyle of the subject. Periodic abstinence (eg, calendar, ovulation, symptothermal, post-ovulation methods) and withdrawal are not acceptable methods of contraception.
- Sterilization: Subject has had surgical bilateral oophorectomy (with or without hysterectomy) or tubal ligation at least 6 weeks before taking study treatment. In case of oophorectomy alone, only when the reproductive status of the woman has been confirmed by follow up hormone level assessment and she is not of childbearing potential.
- Male partner sterilization (with the appropriate post-vasectomy documentation of the absence of sperm in the ejaculate). For female study subjects, male partner(s) must have undergone sterilization.
- Use a combination of 2 of the following (eg, both a+b) (all countries, with the exception of the UK):
    a. placement of a nonhormonal intrauterine device or nonhormonal intrauterine system
    b. barrier method of contraception: condom or occlusive cap (diaphragm or cervical vault caps) with spermicidal foam/gel/film/cream/vaginal suppository (not applicable in Australia)
    c. hormonal contraception methods (eg, oral, injected, implanted)
- Use of a combination of the placement of a nonhormonal intrauterine device/system and a hormonal contraception method (eg, oral, injected, implanted) (UK only)

Note: Women are considered post-menopausal and not of childbearing potential if they have had 12 months of natural (spontaneous) amenorrhea with an appropriate clinical profile (eg, age appropriate, history of vasomotor symptoms) or 6 months of spontaneous amenorrhea with serum FSH levels > 40 mIU/mL and estradiol < 20 pg/mL, or have had surgical bilateral oophorectomy, only when the reproductive status of the woman has been confirmed by follow-up hormone level assessment and she is considered not of childbearing potential.

Male subjects and their partners must use highly effective methods of contraception (double barrier, eg, spermicidal gel plus condom [all countries, except the UK]) for the entire duration of the study and continue to use contraception and refrain from fathering a child for 90 days following the study treatment.

In the event of pregnancy in a female subject, study treatment must be discontinued. Pregnancies occurring during study participation (female subject or a female partner of a male subject) must be reported to Amicus and followed as described in Section 11.6.1.

10. STUDY ASSESSMENTS AND PROCEDURES

The schedule of assessments and procedures display each study assessment and procedure along with the scheduled time of occurrence and can be found in Table 9 for subjects from Cohort 1 through the end of Stage 2, Table 10 for subjects from Cohort 1 who continue to Stage 3 and 4, Table 11 for subjects from Cohort 2 who begin treatment in Stage 3, Table 12 for subjects from Cohort 3 who begin treatment in Stage 3, and Table 13 for subjects from Cohort 4 who begin treatment in Stage 3. Serial blood sampling time points for PK and immunological assessments are provided in Table 14. Sparse blood sampling time points for plasma total GAA protein for subjects from Cohorts 1 and 3 in Stage 3 are provided in Table 15. All study assessments should be conducted by the investigator and/or a suitably qualified designee. Information will be recorded in the source documents and, where appropriate, the eCRF.

Assessments that occur throughout the study, where applicable and to the extent possible, should be completed during the designated visit, preferably in the order specified in the study procedure manual. Once established, every effort should be made to maintain the order of procedures at each study visit throughout the study.

Table 9: Schedule of Assessments for Stages 1 and 2, Cohort 1 Subjects

Assessments or Procedures[a]	Screening	Baseline	Stage 1[b]			Stage 2[b]						Stage 3
			Period 1	Period 2	Period 3	Period 4			Period 5			End of PK
	Visit 1 Days -28 to -1	Visit 2 Day 0	Visit 3 Day 1 (±3 d)	Visit 4 Day 15 (±3 d)	Visit 5 Day 29 (±3 d)	Visit 6 Day 43 (±3 d)	Visit 7 Day 57 (±3 d)	Visit 8 Day 71 (±3 d)	Visit 9 Day 85 (±3 d)	Visit 10 Day 99 (±3 d)	Visit 11 Day 113 (±3 d)	Visit 12 Day 127 (±3 d)
Informed consent	X											
Assign subject number	X											
Demographics	X											
Eligibility criteria	X	X										
IXRS Registration	X											
Medical history, fall history, and prior medications	X	X										
AEs, SAEs, IARs, and falls	X	X	X	X	X	X	X	X	X	X	X	X
Concomitant medications and nondrug therapies	X	X	X	X	X	X	X	X	X	X	X	X
Vital signs[c, d]	X	X	X	X	X	X	X	X	X	X	X	X
Weight	X	X				X			X			X
Height	X											
Physical examination[e]	X	X	X	X	X	X	X	X	X	X	X	X
12-lead ECG[d]	X	X	X	X	X	X	X	X	X	X	X	X

Table 9: Schedule of Assessments for Stages 1 and 2, Cohort 1 Subjects (Continued)

Assessments or Procedures[a]	Screening	Baseline	Stage 1[b]			Stage 2[b]						Stage 3
			Period 1	Period 2	Period 3	Period 4			Period 5			End of PK
	Visit 1 Days -28 to -1	Visit 2 Day 0	Visit 3 Day 1 (±3 d)	Visit 4 Day 15 (±3 d)	Visit 5 Day 29 (±3 d)	Visit 6 Day 43 (±3 d)	Visit 7 Day 57 (±3 d)	Visit 8 Day 71 (±3 d)	Visit 9 Day 85 (±3 d)	Visit 10 Day 99 (±3 d)	Visit 11 Day 113 (±3 d)	Visit 12 Day 127 (±3 d)
Patient-reported Outcomes												
R-PAct scale		X										X
RHS		X										X
FSS		X										X
PROMIS® measurements for dyspnea, fatigue, physical functioning, and upper extremity[f]		X										X
Clinical Safety Laboratory Assessments, Immunological Assessments, and Urine Collections												
Serum chemistry	X		X	X	X	X	X	X	X	X	X	X
Hematology	X		X	X	X	X	X	X	X	X	X	X
Urinalysis	X		X	X	X	X	X	X	X	X	X	X
Blood sample for anti-rhGAA antibodies[g]	X		X	X	X	X	X	X	X	X	X	X
Blood sample for assessments of neutralizing antibodies[g]	X		X	X	X	X	X	X	X	X	X	X

Table 9: Schedule of Assessments for Stages 1 and 2, Cohort 1 Subjects (Continued)

Assessments or Procedures[a]	Screening	Baseline	Stage 1[b]			Stage 2[b]						Stage 3
			Period 1	Period 2	Period 3	Period 4			Period 5			End of PK
	Visit 1 Days -28 to -1	Visit 2 Day 0	Visit 3 Day 1 (±3 d)	Visit 4 Day 15 (±3 d)	Visit 5 Day 29 (±3 d)	Visit 6 Day 43 (±3 d)	Visit 7 Day 57 (±3 d)	Visit 8 Day 71 (±3 d)	Visit 9 Day 85 (±3 d)	Visit 10 Day 99 (±3 d)	Visit 11 Day 113 (±3 d)	Visit 12 Day 127 (±3 d)
Blood sample for measurement of pro-inflammatory cytokines and/or other biomarkers of immune system activation[h]	X		X	X	X	X	X	X	X	X	X	X
Blood sample for exploratory biomarkers of Pompe disease and immunogenicity[g]	X		X	X	X	X	X	X	X	X	X	X
IgE measurement[i]	X	***If needed***										
Confirm Pompe diagnosis: blood sample for *GAA* genotyping if needed	X											
PK Assessments (also see Table 14 for times of blood sampling)[j]												
Serial blood samples for plasma GAA activity[k]			X	X	X	X	X[l]	X	X	X[l]	X	

Table 9: Schedule of Assessments for Stages 1 and 2, Cohort 1 Subjects (Continued)

Assessments or Procedures[a]	Screening	Baseline	Stage 1[b]			Stage 2[b]						Stage 3
			Period 1	Period 2	Period 3	Period 4			Period 5			End of PK
	Visit 1 Days -28 to -1	Visit 2 Day 0	Visit 3 Day 1 (±3 d)	Visit 4 Day 15 (±3 d)	Visit 5 Day 29 (±3 d)	Visit 6 Day 43 (±3 d)	Visit 7 Day 57 (±3 d)	Visit 8 Day 71 (±3 d)	Visit 9 Day 85 (±3 d)	Visit 10 Day 99 (±3 d)	Visit 11 Day 113 (±3 d)	Visit 12 Day 127 (±3 d)
Serial blood samples for plasma total GAA protein concentration[k]			X	X	X	X	X[l]	X	X	X[l]	X	
Serial blood samples for plasma AT2221 concentration[m]						X	X[l]	X	X	X[l]	X	
Miscellaneous Tests												
Collect aliquot of infusion solution[n]			X	X	X	X	X	X	X	X	X	X
Urine pregnancy[o]	X		X	X	X	X	X	X	X	X	X	X
Urine sample for Hex4	X		X	X	X	X	X	X	X	X	X	X
Pulmonary Function Tests												
SNIP		X										X
MIP		X										X
MEP		X										X
FVC		X										X

Table 9: Schedule of Assessments for Stages 1 and 2, Cohort 1 Subjects (Continued)

Assessments or Procedures[a]	Screening	Baseline	Stage 1[b]			Stage 2[b]						Stage 3
			Period 1	Period 2	Period 3	Period 4			Period 5			End of PK
	Visit 1 Days -28 to -1	Visit 2 Day 0	Visit 3 Day 1 (±3 d)	Visit 4 Day 15 (±3 d)	Visit 5 Day 29 (±3 d)	Visit 6 Day 43 (±3 d)	Visit 7 Day 57 (±3 d)	Visit 8 Day 71 (±3 d)	Visit 9 Day 85 (±3 d)	Visit 10 Day 99 (±3 d)	Visit 11 Day 113 (±3 d)	Visit 12 Day 127 (±3 d)
Motor Function Tests[p]												
Gower's maneuver		X										X
TUG		X										X
10MWT		X										X
4-stair climb		X										X
GSGC score		X										X
6MWT		X										X
Muscle Strength Tests												
Manual muscle strength by MRC		X										X
Quantitative muscle strength by hand-held dynamometer		X										X
Study Dosing												
AT2221 (mg)			N/A	N/A	N/A	130	130	130	260	260	260	260[q]
ATB200 (mg/kg)			5	10	20	20	20	20	20	20	20	20[q]

Abbreviations: 6MWT = 6-Minute Walk Test; 10MWT = 10-Meter Walk Test; AE = adverse event; anti-rhGAA antibodies = antibodies that recognize and can bind to rhGAA; ECG = electrocardiogram; ET = Early Termination (visit); FSS = Fatigue Severity Scale; FVC = forced vital capacity; GAA = human acid α-glucosidase; *GAA* = gene that encodes human acid α-glucosidase (GAA protein); GSGC = gait, stairs, Gower, chair maneuver; Hex4 = hexose tetrasaccharide; IAR = infusion-associated reaction; IgE = immunoglobulin E; IXRS = interactive response technology trial management system; MRC = medical research criteria; MEP = maximum expiratory pressure; MIP = maximum inspiratory pressure; N/A = not applicable; PK = pharmacokinetics; PROMIS = Patient-reported Outcomes Measurement Information System;

rhGAA = recombinant human acid α-glucosidase; RHS = Rotterdam Handicap Scale; R-PAct = Rasch-built Pompe-specific activity; SAE = serious adverse event; SNIP = Sniff Nasal Inspiratory Pressure; TUG = Timed Up and Go

[a] All study assessments will be performed on the day indicated for each period. Twenty-four-hour PK assessments will take place on the second day of each visit (see Table 14).

[b] Sentinel dosing for the first 2 sentinel subjects during all periods and in all stages will be followed by safety review by investigators and Amicus Medical Monitor before dosing all the other subjects.

[c] Sitting vital sign assessments will include measurement of body temperature, rate of respiration, heart rate, and systolic and diastolic blood pressures.

[d] Vital signs will be monitored at hours 0, 1, 4, 6, 12, and 24 (Stage 1) and 0, 1, 4, 6, 11, and 25 (Stage 2). Vital signs should be measured after the blood draws for PK assessments if possible. A standard 12-lead ECG will be performed after infusion at all study visits and at the beginning and end of the infusion (approximately 4 hours after the start of infusion) for all PK visits.

[e] Comprehensive physical examination at Screening and Visit 12; brief physical examination will be performed at other visits where indicated.

[f] PROMIS assessments will be administered if a validated instrument is available for the country.

[g] Blood sampling is to be performed before the infusion commences or AT2221 is dosed. The blood draw can be taken at the same time as clinical safety sample, which is described in the laboratory flowchart. Total GAA protein concentration may be measured as well, as assay sensitivity can be affected by GAA protein.

[h] Three blood samples will be taken: one just prior to dosing of AT2221 (if applicable), one just prior to the start of infusion (preferably within 5 to 10 minutes prior to the start of infusion), and another approximately 2 hours or halfway through the infusion. For Screening and Baseline visits, the blood draws are to be spaced apart at time points similar to PK visit.

[i] At Screening, a sample will be obtained. At all other visits, a blood sample will be obtained for IgE determination if a subject has signs of infusion-related reaction or anaphylaxis (see Section 11.1.3). The sample should be obtained 6 to 8 hours after the onset of the reaction and at 24 hours for IgE measurement. A third sample is to be drawn immediately before the next administration of study drug.Total GAA protein concentration will be measured as well, as assay sensitivity can be affected by GAA protein.

[j] The 24-hour PK sample for each PK assessment will be collected on the second day of each visit.

[k] Blood samples for plasma GAA activity levels and total GAA protein concentrations are to be taken just prior to initiation of infusion (Time 0) and at times indicated in Table 14.

[l] Blood sample for plasma GAA activity, total GAA protein, and AT2221 will be collected prior to administration of the oral dose of AT2221. There will be no subsequent serial PK blood draws.

[m] Blood samples for plasma AT2221 concentrations are to be taken just prior to AT2221 oral administration (Time 0) and at times indicated in Table 14.

[n] Infusion solution aliquots will be stored for future analysis as needed.

[o] For female subjects of childbearing potential, if applicable.

[p] Motor function tests can be performed between Day -7 and Day 0 for baseline measures.

[q] Study treatment will not be dispensed to subjects who complete the ET Visit. No infusion solution will be collected.

Table 10: Schedule of Assessments for Stages 3 and 4, Cohort 1 Subjects

Assessments or Procedures[a]	Stage 3 Cohort 1 Subjects[b]								Stage 4
	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d) every 6 months
AEs, SAEs, IARs, and falls	X	X	X	X	X	X	X	X	X
Concomitant medications and nondrug therapies	X	X	X	X	X	X	X	X	X
Vital signs[c]	X	X	X	X	X	X	X	X	X
Weight	X	X	X	X	X	X	X	X	X
Physical examination[d]	X	X	X	X	X	X	X	X	X
12-lead ECG	X	X	X	X	X	X	X	X	X
Patient-reported Outcomes									
R-PAct scale	X	X	X	X	X	X	X	X	X
RHS	X	X	X	X	X	X	X	X	X
FSS	X	X	X	X	X	X	X	X	X
PROMIS® measurements for dyspnea, fatigue, physical functioning, and upper extremity[e]	X	X	X	X	X	X	X	X	X

Table 10: Schedule of Assessments for Stages 3 and 4, Cohort 1 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 1 Subjects[b]								Stage 4
	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d) every 6 months
Global Impression of Change									
PGIC	X	X	X	X	X	X	X	X	X
SGIC	X	X	X	X	X	X	X	X	X
Clinical Safety Laboratory Assessments, Immunological Assessments, and Urine Collections									
Serum chemistry	X	X	X	X	X	X	X	X	X
Hematology	X	X	X	X	X	X	X	X	X
Urinalysis	X	X	X	X	X	X	X	X	X
Blood sample for anti-rhGAA antibodies[f]	X	X	X	X	X	X	X	X	X
Blood sample for assessments of neutralizing antibodies[f]	X	X	X	X	X	X	X	X	X
Blood sample for measurement of pro-inflammatory cytokines and/or other biomarkers of immune system activation[g]	X	X	X	X	X	X	X	X	
Blood sample for exploratory biomarkers of Pompe disease and immunogenicity[f]	X	X	X	X	X	X	X	X	X
Blood sample for plasma GAA total protein[h]						X			

Table 10: Schedule of Assessments for Stages 3 and 4, Cohort 1 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 1 Subjects[b]								Stage 4
	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d) every 6 months
IgE measurement[i]	***If needed***								
Miscellaneous Tests									
Urine pregnancy[j]	X	X	X	X	X	X	X	X	X
Urine sample for Hex4	X	X	X	X	X	X	X	X	X
Pulmonary Function Tests									
SNIP	X	X	X	X	X	X	X	X	X
MIP	X	X	X	X	X	X	X	X	X
MEP	X	X	X	X	X	X	X	X	X
FVC	X	X	X	X	X	X	X	X	X
Motor Function Tests									
Gower's maneuver	X	X	X	X	X	X	X	X	X
TUG	X	X	X	X	X	X	X	X	X
10MWT	X	X	X	X	X	X	X	X	X
4-stair climb	X	X	X	X	X	X	X	X	X
GSGC score	X	X	X	X	X	X	X	X	X
6MWT	X	X	X	X	X	X	X	X	X
Muscle Strength Tests									
Manual muscle strength by MRC	X	X	X	X	X	X	X	X	X

Table 10: Schedule of Assessments for Stages 3 and 4, Cohort 1 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 1 Subjects[b]								Stage 4
	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d) every 6 months
Quantitative muscle strength by hand-held dynamometer	X	X	X	X	X	X	X	X	X
Study Dosing									
AT2221 (260 mg)	X	***Every other week oral co-administration***							
ATB200 (20 mg/kg)	X	***Every other week infusion co-administration***							

Abbreviations: 6MWT = 6-Minute Walk Test; 10MWT = 10-Meter Walk Test; AE = adverse event; anti-rhGAA antibodies = antibodies that recognize and can bind to rhGAA; ECG = electrocardiogram; ET = Early Termination (visit); FSS = Fatigue Severity Scale; FVC = forced vital capacity; GAA = human acid α-glucosidase; GAA = gene that encodes human acid α-glucosidase (GAA protein); GSGC = Gait, Stairs, Gower, Chair maneuver; Hex4 = hexose tetrasaccharide; IAR = infusion-associated reaction; IgE = immunoglobulin E; MRC = medical research criteria; MEP = maximum expiratory pressure; MIP = maximum inspiratory pressure; PGIC = Physician Global Impression of Change; PROMIS = Patient-reported Outcomes Measurement Information System; rhGAA = recombinant human acid α-glucosidase; RHS = Rotterdam Handicap Scale; R-PAct = Rasch-built Pompe-specific activity; SAE = serious adverse event; SGIC = Subject Global Impression of Change; SNIP = Sniff Nasal Inspiratory Pressure; TUG = Timed Up and Go

[a] All study assessments will be performed in the month indicated.

[b] Sentinel dosing for the first 2 sentinel subjects during all periods and in all stages will be followed by safety review by investigators and Amicus Medical Monitor before dosing all the other subjects.

[c] Sitting vital sign assessments will include measurement of body temperature, rate of respiration, heart rate, and systolic and diastolic blood pressures.

[d] Comprehensive physical examinations will be performed during the visits for Month 12, Month 24, and every assessment visit in Stage 4. Brief physical examinations will be performed at all other visits.

[e] PROMIS assessments will be administered if a validated instrument is available for the country.

[f] Blood sampling is to be performed before the infusion commences or AT2221 is dosed. The blood draw can be taken at the same time as clinical safety sample, which is described in the laboratory flowchart. Total GAA protein concentration may be measured as well, as assay sensitivity can be affected by GAA protein.

[g] Three blood samples will be taken: one just prior to dosing of AT2221 (if applicable), one just prior to the start of infusion (preferably within 5 to 10 minutes prior to the start of infusion), and another approximately 2 hours or halfway through the infusion. For Screening and Baseline visits, the blood draws are to be spaced apart at time points similar to PK visit.

[h] Sparse blood samples for plasma total GAA protein will be taken at any one visit after at least 18 months of ATB200/AT2221 treatment at times indicated in Table 15.

[i] A blood sample will be obtained for IgE determination if a subject experiences anaphylaxis as defined in Appendix 1 or a moderate to severe infusion reaction (in the opinion of the investigator). A blood draw is to be performed 6 to 8 hours after the start of the infusion reaction and at 24 hours for IgE measurement. A third sample is to be drawn immediately before the next administration of study drug. Total GAA protein concentration will be measured as well, as assay sensitivity can be affected by GAA protein.

[j] For female subjects of childbearing potential, if applicable.

Table 11: Schedule of Assessments for Stages 3 and 4, Cohort 2 Subjects

Assessments or Procedures[a]	Stage 3 Cohort 2 Subjects[b]													Stage 4
	Screening Days -28 to -1	Baseline Day 0 (±1 d)	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d) every 6 months
Informed consent	X													
Assign subject number	X													
Demographics	X													
Eligibility criteria	X	X												
IXRS Registration	X													
Medical history, fall history, and prior medications	X	X	X											
AEs, SAEs, IARs, and falls	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Concomitant medications and nondrug therapies	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital signs[c]	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight	X	X				X	X	X	X	X	X	X	X	X
Height	X													
Physical examination[d]	X	X	X	X	X	X	X	X	X	X	X	X	X	X
12-lead ECG	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Table 11: Schedule of Assessments for Stages 3 and 4, Cohort 2 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 2 Subjects[b]												Stage 4
	Screening Days -28 to -1	Baseline Day 0 (±1 d)	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d) every 6 months
Patient-reported Outcomes														
R-PAct scale		X				X	X	X	X	X	X	X	X	X
RHS		X				X	X	X	X	X	X	X	X	X
FSS		X				X	X	X	X	X	X	X	X	X
PROMIS® measurements for dyspnea, fatigue, physical functioning, and upper extremity[e]		X				X	X	X	X	X	X	X	X	X
Global Impression of Change														
PGIC						X	X	X	X	X	X	X	X	X
SGIC						X	X	X	X	X	X	X	X	X
Clinical Safety Laboratory Assessments, Immunological Assessments, and Urine Collections														
Serum chemistry	X		X	X	X	X	X	X	X	X	X	X	X	X
Hematology	X		X	X	X	X	X	X	X	X	X	X	X	X
Urinalysis	X		X	X	X	X	X	X	X	X	X	X	X	X
Blood sample for anti-rhGAA antibodies[f]	X		X			X	X	X	X	X	X	X	X	X

Table 11: Schedule of Assessments for Stages 3 and 4, Cohort 2 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 2 Subjects[b]													Stage 4
	Screening Days -28 to -1	Baseline Day 0 (±1 d)	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d) every 6 months
Blood sample for assessments of neutralizing antibodies[f]	X		X			X	X	X	X	X	X	X	X	X
Blood sample for measurement of pro-inflammatory cytokines and/or other biomarkers of immune system activation[g]	X		X			X	X	X	X	X	X	X	X	
Blood sample for exploratory biomarkers of Pompe disease and immunogenicity[f]	X		X			X	X	X	X	X	X	X	X	X
IgE measurement[h]	X	***If needed***												
Confirm Pompe diagnosis: blood sample for *GAA* genotyping if needed	X													
Miscellaneous Tests														
Collect aliquot of infusion solution			X	X	X									
Urine pregnancy[i]	X		X	X	X	X	X	X	X	X	X	X	X	X

Table 11: Schedule of Assessments for Stages 3 and 4, Cohort 2 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 2 Subjects[b]												Stage 4
	Screening Days -28 to -1	Baseline Day 0 (±1 d)	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d) every 6 months
Urine sample for Hex4	X		X	X	X	X	X	X	X	X	X	X	X	X
Pulmonary Function Tests[j]														
SNIP		X				X	X	X	X	X	X	X	X	X
MIP		X				X	X	X	X	X	X	X	X	X
MEP		X				X	X	X	X	X	X	X	X	X
FVC		X				X	X	X	X	X	X	X	X	X
Muscle Strength Tests[k]														
Manual muscle strength by MRC		X				X	X	X	X	X	X	X	X	X
Quantitative muscle strength by hand-held dynamometer		X				X	X	X	X	X	X	X	X	X

Table 11: Schedule of Assessments for Stages 3 and 4, Cohort 2 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 2 Subjects[b]												Stage 4
	Screening Days -28 to -1	Baseline Day 0 (±1 d)	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d) every 6 months
Study Dosing														
AT2221 (260 mg)	***Every other week oral co-administration***													
ATB200 (20 mg/kg)	***Every other week infusion co-administration***													

Abbreviations: AE = adverse event; anti-rhGAA antibodies = antibodies that recognize and can bind to rhGAA; ECG = electrocardiogram; ET = Early Termination (visit); FSS = Fatigue Severity Scale; FVC = forced vital capacity; GAA = human acid α-glucosidase; *GAA* = gene that encodes human acid α-glucosidase (GAA protein); Hex4 = hexose tetrasaccharide; IAR = infusion-associated reaction; IgE = immunoglobulin E; IXRS = interactive response technology trial management system; MRC = medical research criteria; MEP = maximum expiratory pressure; MIP = maximum inspiratory pressure; PGIC = Physician Global Impression of Change; PROMIS = Patient-reported Outcomes Measurement Information System; rhGAA = recombinant human acid α-glucosidase; RHS = Rotterdam Handicap Scale; R-PAct = Rasch-built Pompe-specific activity; SAE = serious adverse event; SGIC = Subject Global Impression of Change; SNIP = Sniff Nasal Inspiratory Pressure

[a] All study assessments will be performed on the day indicated for each month.

[b] Sentinel dosing for the first 2 sentinel subjects during all periods and in all stages will be followed by safety review by investigators and Amicus Medical Monitor before dosing all the other subjects.

[c] Sitting vital sign assessments will include measurement of body temperature, rate of respiration, heart rate, and systolic and diastolic blood pressures.

[d] Comprehensive physical examinations will be performed at the first visit, and during the visits for Month 12, Month 24, and every assessment visit in Stage 4. Brief physical examinations will be performed at all other visits.

[e] PROMIS assessments will be administered if a validated instrument is available for the country.

[f] Blood sampling is to be performed before the infusion commences or AT2221 is dosed. The blood draw can be taken at the same time as clinical safety sample, which is described in the laboratory flowchart. Total GAA protein concentration may be measured as well, as assay sensitivity can be affected by GAA protein.

[g] Three blood samples will be taken: one just prior to dosing of AT2221 (if applicable), one just prior to the start of infusion (preferably within 5 to 10 minutes prior to the start of infusion), and another approximately 2 hours or halfway through the infusion. For Screening and Baseline visits, the blood draws are to be spaced apart at time points similar to PK visit.

[h] At Screening, a sample will be obtained. At all other visits, if a subject experiences anaphylaxis as defined in Appendix 1 or a moderate to severe infusion reaction (in the opinion of the investigator), a blood draw is to be performed 6 to 8 hours after the start of the infusion reaction and at 24 hours for IgE measurement. A third sample is to be drawn immediately before the next administration of study drug. Total GAA protein concentration will be measured as well, as assay sensitivity can be affected by GAA protein.

[i] For female subjects of childbearing potential, if applicable.

[j] Pulmonary function tests and/or assessments will be performed for subjects without invasive ventilatory support only.

[k] Muscle strength tests will be performed for upper limbs only.

Table 12: Schedule of Assessments for Stages 3 and 4, Cohort 3 Subjects

Assessments or Procedures[a]	Stage 3 Cohort 3 Subjects[b]													Stage 4[c]
	Screening Days -28 to -1	Baseline Day 0	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d)
Informed consent	X													
Assign subject number	X													
Demographics	X													
Eligibility criteria	X	X												
IXRS Registration	X													
Medical history, fall history, and prior medications	X	X												
AEs, SAEs, IARs, and falls	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Concomitant medications and nondrug therapies	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital signs[d, e]	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight	X	X				X	X	X	X	X	X	X	X	X
Height	X													
Physical examination[f]	X	X	X	X	X	X	X	X	X	X	X	X	X	X
12-lead ECG	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Table 12: Schedule of Assessments for Stages 3 and 4, Cohort 3 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 3 Subjects[b]													Stage 4[c]
	Screening Days -28 to -1	Baseline Day 0	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d)
Patient-reported Outcomes														
R-PAct scale		X				X	X	X	X	X	X	X	X	X
RHS		X				X	X	X	X	X	X	X	X	X
FSS		X				X	X	X	X	X	X	X	X	X
PROMIS® measurements for dyspnea, fatigue, physical functioning, and upper extremity[g]		X				X	X	X	X	X	X	X	X	X
Global Impression of Change														
PGIC						X	X	X	X	X	X	X	X	X
SGIC						X	X	X	X	X	X	X	X	X
Clinical Safety Laboratory Assessments, Immunological Assessments, and Urine Collections														
Serum chemistry	X		X	X	X	X	X	X	X	X	X	X	X	X
Hematology	X		X	X	X	X	X	X	X	X	X	X	X	X
Urinalysis	X		X	X	X	X	X	X	X	X	X	X	X	X
Blood sample for anti-rhGAA antibodies[h]	X		X	X	X	X	X	X	X	X	X	X	X	X
Blood sample for assessments of neutralizing antibodies[h]	X		X	X	X	X	X	X	X	X	X	X	X	X

Table 12: Schedule of Assessments for Stages 3 and 4, Cohort 3 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 3 Subjects[b]													Stage 4[c]
	Screening Days -28 to -1	Baseline Day 0	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d)
Blood sample for measurement of pro-inflammatory cytokines and/or other biomarkers of immune system activation[i]	X		X	X	X	X	X	X	X	X	X	X	X	
Blood sample for exploratory biomarkers of Pompe disease and immunogenicity[h]	X		X	X	X	X	X	X	X	X	X	X	X	X
Blood samples for plasma total GAA protein[j]												X		
IgE measurement[k]	X	***If needed***												
Confirm Pompe diagnosis: blood sample for *GAA* genotyping if needed	X													
PK Assessments (see also Table 14 for time of blood samples)[l]														
Serial blood samples for plasma GAA activity			X	X[m]	X									
Serial blood samples for plasma total GAA protein concentration			X	X[m]	X									

Table 12: Schedule of Assessments for Stages 3 and 4, Cohort 3 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 3 Subjects[b]												Stage 4[c]
	Screening Days -28 to -1	Baseline Day 0	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d)
Serial blood samples for plasma AT2221 concentration			X	X[m]	X									
Miscellaneous Tests														
Collect aliquot of infusion solution[n]			X	X	X									
Urine pregnancy[o]	X		X	X	X	X	X	X	X	X	X	X	X	X
Urine sample for Hex4	X		X	X	X	X	X	X	X	X	X	X	X	X
Pulmonary Function Tests														
SNIP		X				X	X	X	X	X	X	X	X	X
MIP		X				X	X	X	X	X	X	X	X	X
MEP		X				X	X	X	X	X	X	X	X	X
FVC		X				X	X	X	X	X	X	X	X	X
Motor Function Tests														
Gower's maneuver		X				X	X	X	X	X	X	X	X	X
TUG		X				X	X	X	X	X	X	X	X	X
10MWT		X				X	X	X	X	X	X	X	X	X
4-stair climb		X				X	X	X	X	X	X	X	X	X
GSGC score		X				X	X	X	X	X	X	X	X	X
6MWT		X				X	X	X	X	X	X	X	X	X

Table 12: Schedule of Assessments for Stages 3 and 4, Cohort 3 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 3 Subjects[b]													Stage 4[c]
	Screening Days -28 to -1	Baseline Day 0	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d)
Muscle Strength Tests														
Manual muscle strength by MRC		X				X	X	X	X	X	X	X	X	X
Quantitative muscle strength by hand-held dynamometer		X				X	X	X	X	X	X	X	X	X
Study Dosing														
AT2221 (260 mg)			X	***Every other week oral co-administration***										
ATB200 (20 mg/kg)			X	***Every other week infusion co-administration***										

Abbreviations: 6MWT = 6-Minute Walk Test; 10MWT = 10-Meter Walk Test; AE = adverse event; Anti-rhGAA antibodies = antibodies that recognize and can bind to rhGAA; ECG = electrocardiogram; ET = Early Termination (visit); FSS = Fatigue Severity Scale; FVC = forced vital capacity; GAA = human acid α-glucosidase; *GAA* = gene that encodes human acid α-glucosidase (GAA protein); GSGC = Gait, Stairs, Gower, Chair maneuver; Hex4 = hexose tetrasaccharide; IAR = infusion-associated reaction; IgE = immunoglobulin E; IXRS = interactive response technology trial management system; MRC = medical research criteria; MEP = maximum expiratory pressure; MIP = maximum inspiratory pressure; PGIC = Physician Global Impression of Change; PK = pharmacokinetics; PROMIS = Patient-reported Outcomes Measurement Information System; rhGAA = recombinant human acid α-glucosidase; RHS = Rotterdam Handicap Scale; R-PAct = Rasch-built Pompe-specific activity; SAE = serious adverse event; SGIC = Subject Global Impression of Change; SNIP = Sniff Nasal Inspiratory Pressure; TUG = Timed Up and Go

[a] All study assessments will be performed on the day indicated for each visit.

[b] Sentinel dosing for the first 2 sentinel subjects during all periods and in all stages will be followed by safety review by investigators and Amicus Medical Monitor before dosing all the other subjects.

[c] Stage 4 begins after 2 years in Stage 3. Frequency of assessments is every 6 months.

[d] Sitting vital sign assessments will include measurement of body temperature, rate of respiration, heart rate, and systolic and diastolic blood pressures.

[e] Vital signs will be monitored at hours 0, 1, 4, 6, 12, and 24 for all PK visits. A standard 12-lead ECG will be performed after infusion at all study visits and at the beginning and end of the infusion (approximately 4 hours after the start of infusion) for all PK visits.

[f] Comprehensive physical examinations will be performed at the first visit, and during the visits for Month 12, Month 24, and every assessment visit in Stage 4. Brief physical examinations will be performed at all other visits.

[g] PROMIS assessments will be administered if a validated instrument is available for the country.

[h] Blood sampling is to be performed before the infusion commences or AT2221 is dosed. The blood draw can be taken at the same time as clinical safety sample, which is described in the laboratory flowchart. Total GAA protein concentration may be measured as well, as assay sensitivity can be affected by GAA protein.

[i] Three blood samples will be taken: one just prior to dosing of AT2221 (if applicable), one just prior to the start of infusion (preferably within 5 to 10 minutes prior to the start of infusion), and another approximately 2 hours or halfway through the infusion. For Screening and Baseline visits, the blood draws are to be spaced apart at time points similar to PK visit.

[j] Sparse blood sampling for plasma total GAA protein will be performed for Cohort 1 and Cohort 3 patients at any one visit after at least 18 months but no more than 24 months of ATB200/AT2221 treatment at times indicated in Table 15.

[k] At Screening, a sample will be obtained. At all other visits, if a subject experiences anaphylaxis as defined in Appendix 1 or a moderate to severe infusion reaction (in the opinion of the investigator), a blood draw is to be performed 6 to 8 hours after the start of the infusion reaction and at 24 hours for IgE measurement. A third sample is to be drawn immediately before the next administration of study drug. Total GAA protein concentration will be measured as well, as assay sensitivity can be affected by GAA protein.

[l] The 24-hour PK sample for each PK assessment will be collected on the second day of each visit (see Table 14).

[m] Blood sample for plasma GAA activity, total GAA protein, and AT2221 will be collected prior to administration of the oral dose of AT2221. There will be no subsequent serial PK blood draws.

[n] Infusion solution aliquots will be stored for future analysis as needed.

[o] For female subjects of childbearing potential, if applicable.

Table 13: Schedule of Assessments for Stages 3 and 4, Cohort 4 Subjects

Assessments or Procedures[a]	Stage 3 Cohort 4 Subjects[b]													Stage 4[c]
	Screening Days -28 to -1	Baseline Day 0	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d)
Informed consent	X													
Assign subject number	X													
Demographics	X													
Eligibility criteria	X	X												
IXRS Registration	X													
Medical history, fall history, and prior medications	X	X												
AEs, SAEs, IARs, and falls	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Concomitant medications and nondrug therapies	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital signs[d]	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight	X	X				X	X	X	X	X	X	X	X	X
Height	X													
Physical examination[e]	X	X	X	X	X	X	X	X	X	X	X	X	X	X
12-lead ECG	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Table 13: Schedule of Assessments for Stages 3 and 4, Cohort 4 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 4 Subjects[b]												Stage 4[c]
	Screening Days -28 to -1	Baseline Day 0	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d)
Patient-reported Outcomes														
R-PAct scale		X				X	X	X	X	X	X	X	X	X
RHS		X				X	X	X	X	X	X	X	X	X
FSS		X				X	X	X	X	X	X	X	X	X
PROMIS® measurements for dyspnea, fatigue, physical functioning, and upper extremity[f]		X				X	X	X	X	X	X	X	X	X
Global Impression of Change														
PGIC						X	X	X	X	X	X	X	X	X
SGIC						X	X	X	X	X	X	X	X	X
Clinical Safety Laboratory Assessments, Immunological Assessments, and Urine Collections														
Serum chemistry	X		X	X	X	X	X	X	X	X	X	X	X	X
Hematology	X		X	X	X	X	X	X	X	X	X	X	X	X
Urinalysis	X		X	X	X	X	X	X	X	X	X	X	X	X
Blood sample for anti-rhGAA antibodies[g]	X		X	X	X	X	X	X	X	X	X	X	X	X
Blood sample for assessments of neutralizing antibodies[g]	X		X	X	X	X	X	X	X	X	X	X	X	X

Table 13: Schedule of Assessments for Stages 3 and 4, Cohort 4 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 4 Subjects[b]												Stage 4[c]
	Screening Days -28 to -1	Baseline Day 0	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d)
Blood sample for exploratory biomarkers of Pompe disease and immunogenicity[g]	X		X	X	X	X	X	X	X	X	X	X	X	X
IgE measurement[h]	X	***If needed***												
Confirm Pompe diagnosis: blood sample for *GAA* genotyping if needed	X													
Miscellaneous Tests														
Collect aliquot of infusion solution[i]			X	X	X									
Urine pregnancy[j]	X		X	X	X	X	X	X	X	X	X	X	X	X
Urine sample for Hex4	X		X	X	X	X	X	X	X	X	X	X	X	X
Pulmonary Function Tests														
SNIP		X				X	X	X	X	X	X	X	X	X
MIP		X				X	X	X	X	X	X	X	X	X
MEP		X				X	X	X	X	X	X	X	X	X
FVC		X				X	X	X	X	X	X	X	X	X

Table 13: Schedule of Assessments for Stages 3 and 4, Cohort 4 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 4 Subjects[b]												Stage 4[c]
	Screening Days -28 to -1	Baseline Day 0	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d)
Motor Function Tests														
Gower's maneuver		X				X	X	X	X	X	X	X	X	X
TUG		X				X	X	X	X	X	X	X	X	X
10MWT		X				X	X	X	X	X	X	X	X	X
4-stair climb		X				X	X	X	X	X	X	X	X	X
GSGC score		X				X	X	X	X	X	X	X	X	X
6MWT		X				X	X	X	X	X	X	X	X	X
Muscle Strength Tests														
Manual muscle strength by MRC		X				X	X	X	X	X	X	X	X	X
Quantitative muscle strength by hand-held dynamometer		X				X	X	X	X	X	X	X	X	X

Table 13: Schedule of Assessments for Stages 3 and 4, Cohort 4 Subjects (Continued)

Assessments or Procedures[a]	Stage 3 Cohort 4 Subjects[b]												Stage 4[c]
	Screening Days -28 to -1	Baseline Day 0	Day 1 (±3 d)	Week 2 (±3 d)	Week 4 (±3 d)	Month 3 (±3 d)	Month 6 (±3 d)	Month 9 (±3 d)	Month 12 (±3 d)	Month 15 (±3 d)	Month 18 (±3 d)	Month 21 (±3 d)	Month 24/ET (±3 d)	Month 30/ET (±7 d)
Study Dosing														
AT2221 (260 mg)			X	***Every other week oral co-administration***										
ATB200 (20 mg/kg)			X	***Every other week infusion co-administration***										

Abbreviations: 6MWT = 6-Minute Walk Test; 10MWT = 10-Meter Walk Test; AE = adverse event; Anti-rhGAA antibodies = antibodies that recognize and can bind to rhGAA; ECG = electrocardiogram; ET = Early Termination (visit); FSS = Fatigue Severity Scale; FVC = forced vital capacity; GAA = human acid α-glucosidase; *GAA* = gene that encodes human acid α-glucosidase (GAA protein); GSGC = Gait, Stairs, Gower, Chair maneuver; Hex4 = hexose tetrasaccharide; IAR = infusion-associated reaction; IgE = immunoglobulin E; IXRS = interactive response technology trial management system; MRC = medical research criteria; MEP = maximum expiratory pressure; MIP = maximum inspiratory pressure; PGIC = Physician Global Impression of Change; PROMIS = Patient-reported Outcomes Measurement Information System; rhGAA = recombinant human acid α-glucosidase; RHS = Rotterdam Handicap Scale; R-PAct = Rasch-built Pompe-specific activity; SAE = serious adverse event; SGIC = Subject Global Impression of Change; SNIP = Sniff Nasal Inspiratory Pressure; TUG = Timed Up and Go

[a] All study assessments will be performed on the day indicated for each visit.

[b] Sentinel dosing for the first 2 sentinel subjects during all periods and in all stages will be followed by safety review by investigators and Amicus Medical Monitor before dosing all the other subjects.

[c] Stage 4 begins after 2 years in Stage 3. Frequency of assessments is every 6 months.

[d] Sitting vital sign assessments will include measurement of body temperature, rate of respiration, heart rate, and systolic and diastolic blood pressures.

[e] Comprehensive physical examinations will be performed at the first visit, and during the visits for Month 12, Month 24, and every assessment visit in Stage 4. Brief physical examinations will be performed at all other visits.

[f] PROMIS assessments will be administered if a validated instrument is available for the country.

[g] Blood sampling is to be performed before the infusion commences or AT2221 is dosed. The blood draw can be taken at the same time as clinical safety sample, which is described in the laboratory flowchart. Total GAA protein concentration may be measured as well, as assay sensitivity can be affected by GAA protein.

[h] At Screening, a sample will be obtained. At all other visits, if a subject experiences anaphylaxis as defined in Appendix 1 or a moderate to severe infusion reaction (in the opinion of the investigator), a blood draw is to be performed 6 to 8 hours after the start of the infusion reaction and at 24 hours for IgE measurement. A third sample is to be drawn immediately before the next administration of study drug. Total GAA protein concentration will be measured as well, as assay sensitivity can be affected by GAA protein.

[i] Infusion solution aliquots will be stored for future analysis as needed.

[j] For female subjects of childbearing potential, if applicable.

Table 14: Pharmacokinetic Blood Sampling (Cohorts 1 and 3)

Stage 1: Periods 1, 2, and 3 Cohort 1														
Time from Start of ATB200 IV Infusion (hr)														
	-1	**0**	**1**	**2**	**3**	**3.5**	**4**	**4.5**	**5**	**6**	**8**	**10**	**12**	**24**
PK Assessments[a]														
Plasma GAA activity		X	X	X	X	X	X	X	X	X	X	X	X	X
Plasma total GAA protein concentration		X	X	X	X	X	X	X	X	X	X	X	X	X
Immunological Assessments														
Anti-rhGAA neutralizing antibodies	X													
Neutralizing antibodies	X													
Pro-inflammatory cytokines[b]	X	X		X										
IgE measurement[c]	***If needed***													

Stage 2: Periods 4 (Visits 6 and 8) and 5 (Visits 9 and 11) Cohort 1 and Stage 3 Cohort 3 - (Day 1 and Week 4 Visits)																			
Time from Start of AT2221 Oral Administration (hr)																			
	0	**1**	**1.5**	**2**	**2.5**	**3**	**3.5**	**4**	**4.5**	**5**	**5.5**	**6**	**7**	**9**	**11**	**12**	**13**	**24**	**25**
PK Assessments[a]																			
Plasma GAA activity		X		X		X		X	X	X	X	X	X	X	X		X		X
Plasma GAA total protein		X		X		X		X	X	X	X	X	X	X	X		X		X

Table 14: Pharmacokinetic Blood Sampling (Cohorts 1 and 3) (Continued)

Stage 2: Periods 4 (Visits 6 and 8) and 5 (Visits 9 and 11) Cohort 1 and Stage 3 Cohort 3 - (Day 1 and Week 4 Visits)																			
Time from Start of AT2221 Oral Administration (hr)																			
	0	**1**	**1.5**	**2**	**2.5**	**3**	**3.5**	**4**	**4.5**	**5**	**5.5**	**6**	**7**	**9**	**11**	**12**	**13**	**24**	**25**
Plasma AT2221 concentration[d]	X	X	X	X	X	X		X		X		X		X	X				X
Immunological Assessments																			
Anti-rhGAA antibodies	X																		
Neutralizing antibodies	X																		
Pro-inflammatory cytokines[b]	X	X				X													
IgE measurement[c]	***If needed***																		

Abbreviations: anti-rhGAA antibodies = antibodies that recognize and can bind to rhGAA; GAA = human acid α-glucosidase; IgE = immunoglobulin E; IV = intravenous; PK = pharmacokinetics

[a] Blood samples for PK assessments from Time 0 to 13 hours should be completed ± 5 minutes from the indicated target time. The 24- and 25-hour time points should be completed ± 1 hour of the indicated target time.

[b] Three blood samples will be taken: one just prior to dosing of AT2221 (if applicable), one just prior to the start of infusion (preferably within 5 to 10 minutes prior to the start of infusion), and another approximately 2 hours or halfway through the infusion.

[c] A blood sample will be obtained for IgE determination if a subject experiences anaphylaxis as defined in Appendix 1 or a moderate to severe infusion reaction (in the opinion of the investigator). A blood draw is to be performed 6 to 8 hours after the start of the infusion reaction and at 24 hours for IgE measurement. A third sample is to be drawn immediately before the next administration of study drug.Total GAA protein concentration will be measured as well, as assay sensitivity can be affected by GAA protein levels.

[d] Blood samples for plasma AT2221 concentrations are to be taken just prior to AT2221 oral administration (Time 0) and at times indicated.

Table 15: Sparse Pharmacokinetic Blood Sampling

Stage 3: Cohorts 1 and 3[a] - (any one visit after at least 18 months of ATB200/AT2221 treatment)																
Time from Start of AT2221 Administration[b] (hr)																
	1	**2**	**2.5**	**3**	**3.5**	**4**	**4.5**	**5**	**5.5**	**6**	**6.5**	**7**	**8**	**9**	**12**	**13**
PK Assessments[b]																
Plasma GAA total protein	X							X		X		X		X		X

[a] If the subject has completed Stage 3, sparse PK sampling will be performed in Stage 4.

[b]Sampling times will be shifted by 1 hour earlier for PK analysis and provided this way in the final PK and clinical databases.

10.1. Description of Study Visits

Please refer to the schedules of assessments in Table 9, Table 12, and Table 14 for the listing and timing of all PK assessments for all visits during the study.

10.1.1. Screening Visit

All subjects will complete the 1-day Screening Visit up to 4 weeks (or 28 days) before starting study treatment. Subjects from Cohorts 2, 3, and 4 who are newly enrolled in the study will complete a Screening Visit in Stage 3.

Assessments may be repeated if requested by the Amicus Medical Monitor.

The Screening Visit must be registered in the interactive response technology trial management system (IXRS) to trigger drug shipment to the site. Refer and comply with detailed guidelines in the IXRS manual.

10.1.2. Baseline

All subjects will complete a Baseline Visit. The Baseline Visit will be Day 0. Functional assessments for baseline measures can be performed between Day -7 to Day 0, as it may not be possible for a subject to complete them all in a single day. If an assessment is not performed during the Baseline Visit, the Screening value may be used as the baseline value.

Retrospective data for patients from Cohorts 1, 2, and 3: All available data for 6MWT, pulmonary function tests, other motor function tests such as 10-meter walk, 4-stair climb, and Gower's, quantitative (dynamometry) and qualitative muscle strength tests, Gait, Stairs, Gower, and Chair maneuver (GSGC) score, and patient-reported outcomes (PROs), if available should be recorded in the Electronic Data Capture (EDC).

Retrospective data will be gathered from subjects in Cohort 4: At least three 6MWT results, each at least 6 months apart must be available and entered in the EDC. At least two 6MWT results should be within the past three years. Additionally, pulmonary function tests, other motor function tests such as 10-meter walk, 4-stair climb, and Gower's, quantitative (dynamometry) and qualitative muscle strength tests, GSGC score, and PROs, if available should be recorded in the EDC. In addition to the above requirement, all available data will be collected.

10.1.3. Stage 1, Period 1 (Visit 3, Day 1), Period 2 (Visit 4, Day 15), and Period 3 (Visit 5, Day 29) (Cohort 1 Subjects Only)

During Period 1, sentinel dosing will be initiated for the first 2 subjects who receive 5 mg/kg ATB200. The site may proceed with dosing the remainder of the subjects receiving 5 mg/kg ATB200, as described in Section 8.2. The same process will be followed for Periods 2 and 3.

Dosing for Periods 1, 2, and 3 is as follows:

- Period 1: Each subject will receive a single IV infusion of 5 mg/kg ATB200
- Period 2: Each subject who has completed Period 1 will receive a single IV infusion of 10 mg/kg ATB200

- Period 3: Each subject who has completed Period 2 will receive a single IV infusion of 20 mg/kg ATB200

10.1.4. Stage 2, Period 4: Visit 6 (Day 43), Visit 7 (Day 57), and Visit 8 (Day 71) (Cohort 1 Subjects Only)

For Period 4 (Visit 6 and Visit 8), the same assessments indicated for Period 1, Period 2, and Period 3 will be performed for all subjects who have completed Period 3 (refer to Section 10.1.3 and Table 9).

At Visit 7, only a pre-dose blood sample will be taken for plasma GAA activity levels, total GAA protein concentration, and AT2221 concentration. No post-dose PK blood samples will be taken.

Note: Subjects are required to fast at least 2 hours before and 2 hours after administration of oral AT2221.

10.1.5. Stage 2, Period 5: Visit 9 (Day 85), Visit 10 (Day 99), and Visit 11 (Day 113) (Cohort 1 Subjects Only)

For Period 5 (Visit 9 and Visit 11), the same assessments indicated for Period 1, Period 2, and Period 3 will be performed for all subjects who have completed Period 4 (refer to Section 10.1.3 and Table 9).

At Visit 10, only a pre-dose blood sample will be taken for plasma GAA activity level, total GAA protein concentration, and AT2221 concentration. No post-dose PK blood samples will be taken.

Note: Subjects are required to fast at least 2 hours before and 2 hours after administration of oral AT2221.

10.1.6. End of PK: Visit 12 (Day 127)

Assessments to be performed at the End of PK Visit are outlined in Table 9. The End of PK Visit will be considered Day 1 of Stage 3 for the subjects in Cohort 1. Subjects will receive 20 mg/kg ATB200 and 260 mg AT2221 at this visit.

Note: Subjects are required to fast at least 2 hours before and 2 hours after administration of oral AT2221.

10.1.7. Stage 3: PK Sampling for Cohort 3 Subjects

All subjects will receive 20 mg/kg ATB200 and 260 mg AT2221 on a biweekly (every other week) basis in Stage 3. For Cohort 3 subjects, for the first and third doses of 20 mg/kg ATB200 and 260 mg AT2221 at Day 1 and Week 4, serial blood samples for plasma GAA activity levels, total protein, and AT2221 will be performed according to Table 9. Cohort 4 will have no PK sampling.

At Visit 2, a blood sample for plasma GAA activity levels, total GAA protein concentration, and AT2221 will be collected prior to administration of the oral dose of AT2221. There will be no subsequent serial PK blood draws in Stage 3.

Note: Subjects are required to fast at least 2 hours before and 2 hours after administration of oral AT2221.

10.1.8. Stage 3: Sparse PK Sampling for Cohorts 1 and 3 Subjects

After at least 18 months of ATB200/AT2221 treatment in Stage 3, all Cohort 1 and 3 subjects will have sparse blood sampling for plasma total GAA protein taken for estimation of AUC and total plasma clearance (CL_T) by compartmental or population PK modeling as indicated in Table 15. If the subject has completed Stage 3, the assessment will be performed in Stage 4.

10.1.9. Stage 4: Open-label Extension

All subjects will receive 20 mg/kg ATB200 and 260 mg AT2221 on a biweekly basis in Stage 4. Functional assessments will be performed every 6 months.

Note: Subjects are required to fast at least 2 hours before and 2 hours after administration of oral AT2221.

10.1.10. Early Termination Visit/End of Study

Subjects who discontinue early from the study will no longer receive study treatment, regardless of study stage. They will undergo the same assessments as for the Month 24 Visit. Assessments are outlined in Table 9, Table 10, Table 11, Table 12, and Table 13.

Upon study completion or subject discontinuation, subjects who are confirmed positive for anti-rhGAA antibodies will complete follow-up immunological testing as long as they have a positive result for up to 12 months after the last dose of study drug (ie, at 1-, 3-, 6-, 9-, and 12-months follow-up) or until they begin treatment with another ERT or investigational therapy. If anti-rhGAA antibodies are not confirmed positive at follow-up time points earlier than 6 months, no further testing is required.

10.1.11. Follow-up Period

The follow-up period has one visit or telephone call. This visit will occur within 1 month after the last treatment visit unless the subject enters a separate study protocol or other program depending on local regulations. At the follow-up visit, the following assessments and procedures will be performed: weight, vital signs, physical examination, serum chemistry, hematology, urine pregnancy test (all females of childbearing potential), urinalysis, ECG, and recording of concomitant medications and adverse events. If the patient does not return for the follow-up visit, information will be collected about concomitant medications and adverse events via telephone call.

10.1.12. Unscheduled Visits

Unscheduled visits for medical reasons such as evaluation of AEs and additional laboratory tests and/or other investigations to further clarify an abnormal test/assessment result can be performed at any time at the investigator's discretion or if requested by the Amicus Medical Monitor. The date and reason for the visit, in addition to information collected from procedures performed, should be captured in the subject's medical record and/or on appropriate eCRFs.

10.2. Description of Study Assessments

The following assessments or procedures are described in order of their appearance in the Schedule of Assessments and Procedures (Table 9, Table 10, Table 11, Table 12, and Table 13). Additional information on collection, processing, and shipping procedures for all samples will be provided in a supporting laboratory flowchart. Details of functional assessments are provided in the corresponding study functional assessment manual.

Any study procedures that involve supine positions may be deferred if the subject is unable to complete them in a supine position.

Due to the amount of time required to perform the functional assessments (ie, 6MWT, motor function tests, muscle strength tests, and pulmonary function tests), these tests may be scheduled either the day before or day after the subject's infusion date if the combination of the functional assessments and the infusion procedure is too burdensome for the subject if performed on the same day.

If a subject is ill or injured at a scheduled visit, the functional assessments may be deferred to when the subject's condition has improved, in consultation with the Medical Monitor.

10.2.1. Informed Consent

Please refer to Section 15.2.1.

10.2.2. Demographics

Demographic data to be collected at Screening and includes date of birth and sex. In the US, race and ethnicity will also be recorded.

10.2.3. Medical History, Fall History, and Prior Medications

Medical history (including FVC and 6MWT, plasma CK, ALT, AST, and urine Hex4 lab results) including information on medications and procedures used to treat/manage each medical condition will be collected at the Screening Visit. This will include a history of falls over the last year. Study staff should complete a structured review of prior medications that are prohibited (Section 8.7) to ensure eligibility and should review again both the medical history (as listed above) and prior medications at Baseline Visit. Details are to be recorded for any past IARs (including intensity, start date, stop date, and outcome), medication use to manage symptoms (including dosage, frequency of administration, start date, and stop date), and any laboratory tests that were obtained as part of the assessment of IARs. Refer to Section 11.1.3 for further information on IARs. In addition, all available information on prior Ab titers and neutralizing antibodies associated with prior ERT use, including the assay technology and the facility where the assay(s) were performed, will be collected in the source documentation for subjects in Cohorts 1 and 2.

10.2.4. Adverse Events and/or Serious Adverse Events

Throughout the study, subjects will be given an opportunity to report AEs. The definitions, reporting, and follow-up of AEs and SAEs are described in Section 11.1.1.

Sentinel dosing for the first 2 sentinel subjects during all periods and in all stages will be followed for safety review by investigators and Amicus Medical Monitor before dosing all the other subjects.

10.2.5. Concomitant Medications and Nondrug Therapies

At each visit, subjects will be asked to report any medications (dosage, frequency, start date, and stop date) and procedures unreported since last visit. All prescription and nonprescription medications will be recorded. In addition, any nondrug therapies (including physiotherapy and occupational therapy) that a subject is undergoing are to be recorded. Concomitant medications will be coded using the World Health Organization Medical Dictionary (WHOMD).

10.2.6. Vital Signs, Weight, and Height

Sitting vital sign assessments will include measurement of body temperature, respiration, heart rate, and systolic and diastolic blood pressures at times noted in Table 9, Table 10, Table 11, Table 12, and Table 13. During blood sample collection for PK assessments for Cohorts 1 and 3, vital signs will be taken after selected blood samples.

During Stage 3, the subject weight will be measured at 3 month intervals in all cohorts. This weight will be used to revise the dose calculation of ATB200 if there has been a change since the previous 3 month measurement.

During Stage 4, the subject weight will be measured at 6 month intervals in all cohorts. This weight will be used to revise the dose calculation of ATB200 if there has been a change since the previous 6 month measurement.

Note: For visits where a subject is administered a new ATB200 dose level for the first time, vital signs will be monitored for the first 12 hours after the start of infusion. During the 48 hour observations period vital signs will be monitored, at a minimum, every 6 to 8 hours until the end of the 48-hour observation period.

10.2.7. Physical Examination

Subjects in Cohort 1 will undergo comprehensive or brief PEs according to the Schedule of Assessments tables. Any clinically significant (CS) abnormal observation, newly emerged, or worsened, during the study should be reported as an AE.

10.2.8. Electrocardiogram

A standard 12-lead ECG will be performed at visits indicated in the Schedule of Assessments tables. For PK visits, ECGs are performed at 2 time points: before dosing and at the end of the infusion (approximately 4 hours after the start of the infusion). Subjects will rest for approximately 5 minutes before the ECG recording begins and will be in the supine position throughout the ECG evaluation. Significant findings not present prior to or worsening after the start of treatment (ie, Baseline), which meet the definition of an AE, must be recorded in the eCRF.

10.2.9. Patient-reported Outcomes (Including Pompe-associated Scales)

Patient-reported outcomes (PROs) to be assessed during this study include R-PAct Scale, Rotterdam Handicap Scale (RHS), Fatigue Severity Scale (FSS), and Patient-reported Outcomes Measurement Information System (PROMIS®) instruments (for dyspnea, fatigue, physical functioning, and upper extremity), and will be administered at Baseline, at the end of Stage 2 for Cohort 1, every 3 months in Stage 3 and every 6 months in Stage 4 for all subjects. The subject should be given the questionnaires to complete at the scheduled visit before any study related procedures take place.

Subjects who have already initiated study drug treatment in this study should complete the PROMIS instruments from the perspective of their experience immediately prior to study entry. For these subjects, this retrospective recall will be done only once. All other instances will reflect current experience.

The PRO tools are provided in Appendix 3, Appendix 4, Appendix 5, and Appendix 6. Countries in which the PRO tools are validated are noted in Appendix 7.

10.2.10. Global Impression of Change

The Subject Global Impression of Change (SGIC) and Physician Global Impression of Change (PGIC) will only be administered in Stage 3 and 4 as indicated in the Schedule of Assessments tables. The administrator of the SGIC will read the questions to the subject.

The Global Impression of Change tools are provided in Appendix 8.

10.2.11. Clinical Safety Laboratory Tests

The collection, processing, and shipment of clinical laboratory samples will be fully described in the study laboratory flowchart. The parameters measured in the serum chemistry, hematology, and urinalysis are summarized in Table 16, Table 17, and Table 18, respectively. Blood and urine samples for clinical laboratory evaluations will be taken before study treatment administration in Stages 1, 2, 3, and 4 and will be collected according to the Schedule of Assessments tables.

The investigator (or his/her designee) will review each laboratory report from the central laboratory and assess any out of range laboratory results as “not clinically significant” or “clinically significant”. Any results which are considered CS should be confirmed in a repeat test at the investigator’s discretion. The investigator should consider repeat testing of persistent CS results until the analyte returns to normal, or until an etiology is determined. The investigator (or his/her designee) will sign and date all laboratory reports.

All CS laboratory results will be reported as an AE or SAE, as appropriate.

If any results demonstrate, in the investigator’s opinion, a CS abnormality that the subject has not previously experienced, then the subject should be asked to return as soon as possible for an unscheduled visit, so that repeat specimens can be obtained.

Clinically significant values are provided in Appendix 2.

10.2.11.1. Serum Chemistry

The serum chemistry parameters are described in Table 16.

Table 16: Serum Chemistry Parameters

ALT	Creatinine
Alkaline phosphatase	Gamma–glutamyltransferase
AST	Glucose
Albumin	Lactate dehydrogenase (LDH)
Bilirubin, total	Magnesium
Blood urea nitrogen	Phosphorous
Calcium, total	Potassium
Carbon dioxide, total (bicarbonate)	Protein, total
Chloride	Sodium
Creatine phosphokinase (CK)	Uric acid

10.2.11.2. Hematology

The hematology parameters are described in Table 17.

Table 17: Hematology Parameters

Platelet count	Automated WBC Differential
Red blood cell count	Neutrophils
WBC count (absolute)	Lymphocytes
Hematocrit	Monocytes
Hemoglobin	Eosinophils
MCV	Basophils

10.2.11.3. Urinalysis

Urinalysis parameters are described in Table 18.

Table 18: Urinalysis Parameters

Color	Ketones
Appearance	Blood
Specific gravity	WBC
pH	Nitrite
Protein	Bilirubin
Glucose	Microscopy of sediment

10.2.12. Immunogenicity Assessments

Blood samples for measurement of anti-rhGAA antibodies (total, neutralizing, and cross-reactive) will be collected at visits indicated in the tables of assessments and collection is to be performed before the infusion commences or AT2221 is dosed. The blood draw can be taken at the same time as clinical safety sample. Sample collection and processing instructions will also be provided in the study laboratory flowchart.

Anti-rhGAA total antibodies and antibodies cross-reactive to alglucosidase alfa will be determined. Upon study completion or subject discontinuation, subjects who are confirmed positive for anti-rhGAA antibodies will complete follow-up immunological testing as long as they have a positive result for up to 12 months after the last dose of study drug (ie, at 1-, 3-, 6-, 9-, and 12-months follow-up) or until they begin treatment with another ERT or investigational therapy. If anti-rhGAA antibodies are not confirmed positive at follow-up time points earlier than 6 months, no further testing is required**.**

Neutralizing antibody assays may include:

- Inhibition of ATB200-mediated hydrolysis of 4-methylumbelliferyl-α-D-glucopyranoside (4-MU-α Glc)
- Inhibition of ATB200-mediated hydrolysis of glycogen
- Inhibition of ATB200 binding to CI-MPR

At Screening, a blood sample will be collected for immunoglobulin E (IgE) for all subjects. If a subject experiences anaphylaxis as defined in Appendix 1 or a moderate to severe infusion reaction (in the opinion of the investigator), 3 blood draws will be required for measurement of IgE levels. The first blood draw should be obtained 6 to 8 hours after onset of the infusion reaction, the second blood draw at 24 hours, and the third immediately before the next administration of study drug. Total GAA protein concentration will be measured from the same blood draw sample as assay sensitivity can be affected by GAA protein levels.

10.2.13. Pro-inflammatory Cytokines and Other Biomarkers of Immune System Activation

Blood samples for pro-inflammatory cytokines and other biomarkers of immune system activation will be drawn at study visits as indicated in Table 14 (PK Blood Sampling). These blood samples will not be drawn for any cohorts in Stage 4 nor for Cohort 4 in Stage 3.

10.2.14. *GAA* Genotyping

At Screening, a blood sample will be collected from subjects for *GAA* gene sequencing to identify *GAA* genotypes that may be associated with Pompe disease (for subjects unable to provide GAA genotyping report during Screening).

10.2.15. Pharmacokinetic Blood Sampling

Serial blood samples for plasma GAA activity levels, total GAA protein concentration, and plasma AT2221 concentrations in Stages 1 and 2 for subjects from Cohort 1 and in Stage 3 for subjects from Cohort 3, will be performed at selected time points detailed in PK Blood Sampling (Table 14) and indicated in Table 9 and Table 12. Total GAA activity levels for 5, 10, and

20 mg/kg ATB200 in plasma will be determined by a validated enzymatic assays using 4-MU-α Glc substrate. Total GAA protein concentrations in plasma for 5, 10, and 20 mg/kg ATB200 will be determined by a validated liquid chromatography–tandem mass spectroscopy (LC-MS/MS) quantification of rhGAA-specific "signature" peptide(s). Plasma AT2221 concentrations for 130 mg and 260 mg AT2221 will be determined by a validated LC-MS/MS assay.

Blood samples for plasma GAA activity, total GAA protein, and AT2221 determinations will be taken as follows:

- Stage 1, a blood sample will be collected from subjects who are enrolled in Cohort 1 for plasma GAA activity levels and total GAA protein only, just prior to initiation of infusion (Time 0) and post-dose at time points detailed in Table 9.
- A blood sample will be collected from subjects who are enrolled in Cohort 1 in Stage 2 and Cohort 3 in Stage 3 for plasma GAA activity levels, total plasma GAA protein, and plasma AT2221 concentration just prior to AT2221 administration (Time 0) and post-dose at time points detailed in Table 14 and indicated in Table 9 and Table 12.
- Blood samples for PK assessments from Time 0 to 12 hours should be completed ± 5 minutes from the indicated target time. The 25-hour time point should be completed ±1 hour of the indicated target time. The 24 and 25-hour PK samples will be collected on the second day of each visit.

Additional information on the sample collection, sample handling, and sample analysis are provided in a separate study laboratory flowchart.

10.2.16. Aliquot of Infusion Solution

A 0.5 to 1 mL sample of the prepared ATB200 infusion solution will be collected from the infusion bag immediately before and immediately after the ATB200 infusions (before flushing the line) at visits indicated in the Schedule of Assessments. This sample may be analyzed for total ATB200 protein or aggregation. The time and date of each preparation of ATB200 infusion solution must be recorded.

10.2.17. Pregnancy Test

All female subjects of childbearing potential (as defined in Section 11.6.1) will have a urine pregnancy test performed at every study visit, except baseline.

10.2.18. Urinary Hex4

Urine samples for Hex4 determination will be collected at every scheduled visit (except Baseline) in Stages 1 and 2, every 3 months in Stage 3, and every 6 months in Stage 4 (see Table 9, Table 10, Table 11, Table 12, and Table 13). Hexose tetrasaccharide concentrations are to be determined by stable isotope dilution-electrospray ionization-tandem mass spectrometry with multiple reaction monitoring. Sample handling and shipment instructions will be outlined in the study laboratory flowchart.

10.2.19. Pulmonary Function Tests (PFTs)/Spirometry

The following pulmonary function tests (PFTs) and/or assessments will be performed at the Baseline, every 3 months in Stage 3 and every 6 months in Stage 4 for all ambulatory and nonambulatory subjects without invasive ventilatory support and at the end of Stage 2 for Cohort 1. Detailed instructions for the PFT assessments will be provided in the corresponding section of a separate study procedure manual.

Maximum inspiratory pressure (MIP), maximum expiratory pressure (MEP), and Sniff Nasal Inspiratory Pressure (SNIP) will be assessed using the Carefusion MicroRPM manometer. The subjects will be seated comfortably with back and feet supported (feet on floor).

Forced vital capacity (FVC) will be assessed in sitting and supine positions using the nSpire KoKo® PFT Spirometer.

10.2.20. Motor Function Tests – Ambulatory Subjects Only

Motor function tests will be performed at Baseline for all subjects, at the end of Stage 2 for Cohort 1, every 3 months in Stage 3 and every 6 months in Stage 4 for all subjects in the order indicated in Table 9, Table 10, Table 11, Table 12, and Table 13. Motor function tests can be performed between Day -7 and Day 0 for baseline measures. Detailed instructions for these tests/assessments will be provided in a separate study procedure manual(s).

10.2.20.1. 6-Minute Walk Test (6MWT)

The objective of the 6MWT is to walk (not run or jog) as far as possible for 6 minutes. The timed walk test should be performed with walking shoes on a flat surface.

In the event of an invalid test, the subject may be offered the opportunity to repeat the 6MWT. The retest may occur immediately after the completion of the invalid 6MWT (following a rest period – minimum of 10 minutes) or the next day, based upon the judgment of the physical therapist and the investigator.

10.2.20.2. Gower's Maneuver

This test involves measuring the time it takes a subject lying supine on the floor to get to a standing position. Subjects may use the floor or their legs, but the test must be performed without the assistance of any furniture.

10.2.20.3. Timed Up and Go (TUG)

The general information regarding Timed Up and Go; a timed test used to assess mobility (TUG) has been derived from Podsiadlo and Richardson (1991). The test involves a seated subject getting up from a standard armchair, walking to a line that is 3 meters (9.8 feet) long, turning around at the line, walking back to the chair, and sitting back down. The test begins when the subject's buttocks begins to lift from the seat and ends when the subject's buttocks touches the seat.

10.2.20.4. 10-Meter Walk Test (10MWT)

This timed walk test should be performed with walking shoes on a flat surface. The purpose of this test is to traverse the distance of 10 meters as fast and safely as possible.

10.2.20.5. 4-Stair Climb

This test is performed with the use of "4 standard stairs." This test will document the time to climb 4 standard stairs and will assist in assignment of the appropriate lower extremity functional grade for those individuals who are ambulatory.

10.2.20.6. Gait, Stairs, Gower, and Chair Maneuver (GSGC) Score

The GSGC score is obtained by adding the scores attributed to each functional test (gait by walking 10 meters, climbing 4 stairs, Gower's maneuver, and rising from a chair) according to how the subject performed them and might vary from a minimum of 4 (normal performance) to a maximum of 27 (worst score).

10.2.21. Muscle Strength Tests – All Subjects

Manual muscle strength will be tested by medical research criteria (MRC) scale. All efforts should be made to maintain the same rater and method throughout the subject's participation in the study. Muscle strength will also be measured using a hand-held dynamometer. Muscle groups tested include shoulder abductors, shoulder adductors, hip flexors, hip abductors, hip adductors (for hip adductors, hand held dynamometer only), knee extensors, knee flexors, elbow extensors, and elbow flexors.

Nonambulatory subjects will perform muscle strength tests for upper limbs only.

These assessments will be performed at baseline and Stage 3 and 4 as indicated in Table 9, Table 10, Table 11, Table 12, and Table 13.

Detailed instructions for these tests will be provided in a separate study procedure manual.

10.3. Biological Specimen

Any biological specimens remaining at the end of the study may be used for future exploratory analyses to improve the understanding of Pompe disease and its management. The retention of samples is optional; subjects will have the option to agree or refuse to have their samples retained. Future genetic testing will not be conducted for retained samples.

10.4. High Sustained Antibody Titer

High sustained antibody titer is defined as subjects with anti-GAA antibody titer of 1:51,200 or higher from the 2 most recent tests using Genzyme's or comparable method prior to screening. If a comparable method is used, it should be discussed with the Amicus Medical Monitor.

10.5. Infusion Only Visits

In Stage 3 and 4, ATB200 is to be administered every 2 weeks as an IV infusion in combination with oral AT2221. Changes in the protocol prescribed duration of ATB200 infusion due to safety or tolerability issues will be documented in the eCRF. During these Infusion Only Visits, in addition to receiving the study treatment, AE/SAE will be collected. Each site should follow their facility guidelines for performing infusions to monitor vital signs or other safety processes that may be in place. These Infusion Only Visits are not outlined in the Schedule of Assessments.

10.6. Home Infusion

Subjects participating in the study will be considered eligible for administration of ATB200/AT2221 on location at their home or residence after having received ATB200/AT2221 administrations in the study for a period of at least 6 months and not having experienced an IAR for a period of 6 months prior to last infusion. Subjects with history of severe IARs are not eligible for home infusions; subjects with a history including a life-threatening IAR are not eligible for home infusions. Eligible subjects may request participation in the Home Infusion Program with requests granted on a case-by-case basis following discussion between the Medical Monitor and PI with prior approvals granted by Institutional Review Boards (IRBs)/Ethics Committees (ECs), and Regulatory Authorities (RAs). Administration of ATB200/AT2221 will be performed at home for eligible subjects by a trained research home infusion nurse provided by an Amicus designated service provider. All subjects participating in the Home Infusion Program, will be required to complete all functional assessments at their primary site as defined in the protocol. Safety monitoring at home including reporting of AEs, SAEs, and IARs is performed by home infusion nurse. Refer to Section 11.1.3 for further information on IARs, including their reporting and management.The home infusion nurse will document and report the findings direct to sites after every infusion as detailed in the manual for home infusion.

Pre-requisites Criteria:

Inclusion Criteria:

- Received ATB200/AT2221 treatment for a minimum of one year
- Meets following requirements regarding feasibility of the home site for ERT infusion:
  - Availability of utilities and equipment required for storage of Drug Product upon receipt
  - Accessibility for delivery of Drug Product and associated supplies
  - Accessibility of trained home infusion nurse for preparation/administration
  - Preparation of ATB200 lyophilized drug product for reconstitution
  - Infusion of large volume parenteral product
  - Disposal of waste and discarding of used medical supplies
- Amicus/designee deems the home site to be appropriate for home infusion following home assessment
- PI agrees to collaborate with the home infusion agency/home infusion nurse with respect to providing advice and support as needed by the home infusion nurse

Exclusion Criteria:

- Any IAR in the past 6 months or recurrent severe or life-threatening IAR including anaphylaxis at any time.

11. ADVERSE EVENTS AND SERIOUS ADVERSE EVENTS

The definition of an AE is presented in Section 11.1.1. For each subject, reporting of AEs begins at the time written informed consent is provided. Investigators are responsible for reporting AE information to Institutional Review Boards (IRBs), International Ethics Committees (IECs), or Research Ethics Boards (REBs) in accordance with the requirements of their institutions.

11.1. Definitions

11.1.1. Adverse Events

An AE is any untoward medical occurrence in a subject administered a pharmaceutical product, biologic (at any dose), or medical device, which does not necessarily have a causal relationship with the treatment. Therefore, an AE can be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medical product, whether or not considered related to the medical product. Adverse events may include the onset of new illness and the exacerbation of pre-existing conditions.

The routine evolution of the disease condition under treatment according to the protocol will be evaluated as part of the disease symptoms assessments. Changes in Pompe disease symptoms must be reviewed by the investigator or a medically qualified sub-investigator and be recorded as "Clinically Significant" or "Not Clinically Significant" in subjects' source records. Changes in the disease condition may not qualify as AEs. However, if there is a clinically relevant worsening of a sign or symptom of the condition under treatment and the outcome fulfills the definition of an AE, it must be reported as directed in the protocol.

Any AE that begins after the first dose of study medication (ATB200 and/or AT2221) will be considered a TEAE.

Subjects experiencing AEs should be followed until their health has returned to baseline status or stabilized or have otherwise been explained.

11.1.2. Serious Adverse Events

An SAE is any AE occurring at any dose that results in any of the following outcomes:

- Death.
- Life-threatening, as assessed by either the investigator or Amicus (places the subject at immediate risk of death from the AE as it occurs; it does not refer to an event which hypothetically might have caused death if it were more severe).
- Persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions.
- Requires in-patient hospitalization (ie, admission to hospital, regardless of duration) or prolongs existing hospitalization.
  - Complications arising during hospitalization are AEs or SAEs, depending on nature of the event, and must be reported as described in this protocol.
  - Medications administered during hospitalization must be recorded in the Concomitant Medications eCRF module.

- Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline does not have to be reported as an SAE provided the planned treatment is documented in Screening Visit source records. If not documented in the Screening Visit source records, an SAE report is required. Outpatient procedures performed in a hospital do not qualify as an SAE.
- Is a congenital anomaly/birth defect.

An important medical event that may not result in one of the above serious outcomes may be considered an SAE when, based upon appropriate medical judgment, it may jeopardize the subject and may require medical or surgical intervention to prevent one of the serious outcomes. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias, or convulsions that do not result in in-patient hospitalization, or development of drug dependency or drug abuse.

For all subjects, AEs and SAEs will be reported from time of informed consent signing, until 30 days after end of study participation.

11.1.3. Infusion-associated Reactions

An IAR is a disorder characterized by 1 or more adverse reaction(s) to the infusion of pharmacological or biological substances. These reactions are classified into 2 major subtypes, immediate and late, according to the time interval between the infusion and the onset of an infusion-related AE.

The reactions are divided into 5 severity grades as follows:

1. Mild (requires observation only); for example, transient flushing or rash
2. Moderate (minimal, usually oral, intervention suffices); for example, urticaria/myalgia/drug fever
3. Severe (vital organ involved yet not in a life-threatening manner; usually requires parenteral medication); for example, bronchospasm/angioedema/hypotension
4. Life-threatening (multisystem involvement of vital organs, urgent and critical care required); for example, systemic anaphylactic reaction
5. Death

Immediate-type IARs commonly involve 1 or more of the following systems: skin (urticaria and erythema), eyes (conjunctivitis), nasopharynx (rhinorrhea, rhinitis), bronchopulmonary tissues (wheezing, bronchospasm), and gastrointestinal tract (diarrhea, vomiting).

Late-type reactions first manifest more than 24 hours post infusion, usually 1 to 3 days post infusion. The most common symptoms of late-type infusion reactions are pruritic skin eruptions, fever, malaise, and polyarthralgia.

Any symptom or sign occurring during or within 2 hours of completion of the infusion should be reported as an immediate-type IAR and deemed drug-related AE, unless an alternative obvious explanation exists, eg, mechanical fall. Any symptom or sign occurring between 2 and 24 hours will be recorded as an IAR if it fits within the description of an IAR in the investigator's opinion. It would be reported as immediate-type IAR and deemed a drug-related AE.

Any symptom or sign occurring between 24 and 96 hours after completion of the infusion that, in the investigator's opinion, fits the clinical description of a symptom or sign associated with an IAR should be reported as a late-type IAR and deemed a drug-related AE.

A symptom or sign that occurs 24 hours or later and in the investigator's opinion is possibly, probably, or likely an IAR should be reported as such and deemed drug-related AE.

Note: The list of IAR symptoms provided in Table 19 is not comprehensive. It is meant to serve as a guideline to the investigators to report and manage IAR as AE. Table 19 also provides a guideline for management of IARs and anaphylaxis (see also Appendix 1). It is at the investigator's discretion to follow the guidelines or implement treatment protocol that their center is accustomed to following.

In the event that a subject experiences an IAR, the subject should be premedicated for subsequent infusions with antihistamines, steroids, and/or acetaminophen. The premedications should be continued for a period of up to 1 year. Decision regarding discontinuation of premedications should occur only after discussion with Amicus Medical Monitor.

Table 19: Guidelines for Management of Immediate Infusion-associated Reactions

Severity	Symptoms (such as, but not restricted to)	Management Guidelines
Mild reaction	Flushing Nausea Mild headache Tachycardia (pulse < 100 bpm) Abdominal pain	REDUCE INFUSION RATE BY 50% Contact investigator. Give oral antihistamine/acetaminophen/paracetamol and/or IV antihistamine. Prepare emergency equipment for any subsequent study treatment escalation. Subject must be observed at site or medical facility for 2-4 hours after resolution of the symptoms. Record all details of times, concomitant medications, and infusion rates. Draw IgE and total GAA sample 6 to 8 and 24 hours after the start of the infusion reaction (2 samples). IF SYMPTOMS PERSIST: STOP THE INFUSION After 30 minutes, if subject has improved, increase to previous infusion rate on direction of investigator/designee. If subject has not improved after 60 minutes, consult with investigator as to whether additional medications should be administered. If subject reacts for second time after return to the prior infusion rate, reduce rate by 50% and contact investigator/designee. IF IN DOUBT, STOP INFUSION

Table 19: Guidelines for Management of Immediate Infusion-associated Reactions (Continued)

Severity	Symptoms (such as, but not restricted to)	Management Guidelines
Moderate reaction	Localized itchiness, and/or raised Urticarial rash (hives) Severe headache Fever or shivering Tachycardia (pulse > 100 bpm) Tachypnea Flushing Nausea Irritability Headache Vomiting Diarrhea Abdominal cramps Myalgia	** STOP INFUSION ** Immediately contact investigator. Administer high-flow oxygen if respiratory symptoms/ distress. Give IM or IV antihistamines (eg, promethazine 25 to 50 mg). Give IV fluid bolus if warranted. Give IV steroids (hydrocortisone 100 mg) on direction of PI/designee. Continue management as directed by the PI/designee. Watch for at least 1 hour after resolution of symptoms. If deemed appropriate by PI/investigator, resume study treatment infusion at reduced rate and prepare emergency equipment. If deemed appropriate by PI/investigator, infusion can be suspended and reinitiated 48 hours later with premedications and at the infusion rate that was last tolerated. Observe subject for a minimum of 6 hours after resolution of the event. Record all details. Report SAE if criteria for SAE reporting are met. Draw IgE and total GAA sample 6 to 8 and 24 hours after the start of the infusion reaction (2 samples).

Table 19: Guidelines for Management of Immediate Infusion-associated Reactions (Continued)

Severity	Symptoms (such as, but not restricted to)	Management Guidelines
Severe reaction, anaphylaxis, or anaphylac-toid reactions	Chest pain Generalized urticarial/ angioedema Symptomatic hypotension(subject feeling faint, paleness) Hoarseness of voice/laryngeal edema Bronchospasm	** STOP INFUSION ** Activate emergency response and immediately contact investigator. For chest pain and symptomatic hypotension, initiate emergency procedures. Administer high-flow oxygen or intubate and mechanically ventilate as appropriate. Give IM adrenaline 300 mcg EpiPen into thigh (avoid buttock). If no response, repeat dose in 5 minutes. Prepare infusion of adrenaline 3 mg/50 mL NS (60 µg/mL), if needed. Give IV fluids 10- to 20-mL/kg fluid bolus on direction of PI/designee. Give IV antihistamines (promethazine 25 to 50 mg in 10 mL WFI over 2 to 3 minutes). Give IV steroids (hydrocortisone 100- to 500-mg bolus IV injection; dilute in 10 mL NS for doses between 100 and 500 mg) and infuse over 2 minutes. Give nebulized beta-2 agonists with high-flow oxygen on direction of PI/designee. Administer other treatments and/or transfer patient to hospital/ ICU as recommended by investigator/ designee. Observe subject for a minimum of 24 hours after resolution of the event. If deemed appropriate by PI/investigator, study treatment infusion can be re-initiated 7 to 14 days with premedications and at the 4-hour duration. Draw IgE and total GAA sample 6 to 8 and 24 hours after the start of the infusion reaction (2 samples). Report SAE if criteria for SAE reporting are met.

Abbreviations: GAA = human acid α-glucosidase; IgE = immunoglobulin E; IM = intramuscular; IV = intravenous; NS = normal saline; PI = Principal Investigator; SAE = serious adverse event; WFI = water for injection
Sources: Vogel 2010; Sampson et al 2006

11.2. Relationship to Study Treatment

The investigator or a medically qualified sub-investigator will review each event and assess its relationship to study treatment based on available information according to the following guidelines:

- Definite: A reaction that follows a distinct temporal relationship from administration of study treatment; that follows a known reaction to the agent or chemical group of

the study treatments; and that cannot be explained by the subject's clinical state or other factors.

- Probable: A reaction that follows a reasonable temporal sequence from administration of study treatment; that follows a known or expected response pattern to the suspected study treatment; and that could not be reasonably explained by the known characteristics of that subject/subject's clinical state.
- Possible: A reaction that follows a reasonable temporal sequence from administration of study treatment; that follows a known or expected response pattern to the suspected study treatment; but that could readily have been produced by a number of other factors.
- Unlikely: A reaction that does not follow a reasonable temporal sequence from administration of study treatment. However, causality from study treatment cannot be ruled out.
- Unrelated: A reaction for which sufficient data exist to indicate that the etiology is unrelated to study treatment.

For the purpose of reporting SAEs to regulatory authorities and IRBs/ECs, any AE assessed by the investigator as definitely, probably, or possibly related to ATB200 and/or AT2221 will be considered "related" to study treatment (ie, associated with the use of study treatment). Any AE assessed as unlikely or unrelated will be considered "not related" to study treatment (ie, not associated with the use of study treatment).

11.3. Assessment of Intensity

The following definitions for rating intensity will be used for all AEs:

- Mild: Awareness of sign, symptom, or event, but the AE is easily tolerated and does not interfere with daily activity.
- Moderate: Discomfort enough to cause interference with usual activity and may warrant intervention, but the subject is still able to function.
- Severe: Incapacitating with inability to do usual activities or significantly affects clinical status, and requires medical intervention.

When the determination of AE intensity rests on medical judgment, the determination of intensity must be made with the appropriate involvement of the investigator or a medically qualified sub-investigator.

It is important to distinguish the difference between SAEs and events that are deemed as severe in intensity. Adverse events of severe intensity are not serious if one of the definitions in Section 11.1.2 is not met; AEs of any intensity must be reported as SAEs if one of the definitions in Section 11.1.2 is met.

11.4. Reporting Adverse Events

Information regarding AEs is to be obtained by questioning or examining subjects.

At each visit, beginning from the time written informed consent is provided until 30 days after the last treatment visit, all new complaints and symptoms (ie, those not existing before subjects provide informed consent) must be recorded as AEs in the eCRF and in subjects' source records. Findings documented on Screening ECGs that are unknown to subjects (ie, asymptomatic subjects) should be recorded as medical history (inclusive of FVC and 6MWT, plasma CK, ALT, AST, and urine Hex4 lab results).

- Pre-existing complaints or symptoms that worsen (eg, increase in intensity, frequency) after subjects provide informed consent must be entered in the AE eCRF module.
- CS laboratory abnormalities must be reported by the investigator as an AE or SAE as appropriate.
- CS changes to Screening ECG results must be reported by the investigator as an AE or SAE as appropriate. CS values will be provided in Appendix 2.
- AEs related to a protocol-required procedure.

For each reported AE, the date the event started and ended, action taken, outcome (resolved, resolved with sequelae, ongoing, or fatal), intensity, and relationship to study treatment must be noted. Every effort should be made to report the start and end time AEs occurred.

All subjects who have AEs, whether or not events are considered associated with the use of ATB200 administered as a single agent or the use of ATB200 co-administered with AT2221, must be monitored to determine the outcome. The clinical course of AEs will be followed according to accepted standards of medical practice, even after the end of the study, until their health has returned to baseline status or stabilized or have otherwise been explained. Should an AE result in death, a full autopsy report should be supplied, if possible. For all AEs that require or result in subject discontinuation from the study, relevant clinical assessments and laboratory tests will be repeated as clinically appropriate, until final resolution or stabilization of the event(s).

11.4.1. Reporting Serious Adverse Events

If an AE is serious (as defined in Section 11.1.2), the investigator must submit a SAE report form at the time the SAE is identified. All SAEs must be immediately reported by the investigator to Amicus or Amicus' representative, but no later than 24 hours after any study site personnel is aware of the event. Serious adverse event report forms must be faxed to the designated Safety Fax number (+ 1 732-289-6060). All supporting documentation available at the time of reporting, with all subject identifiers redacted in accordance with local requirements, must be included in the fax along with the SAE report form. FAX reporting is preferred, but in case of no access to a fax machine, send documentation to the following email address: saereporting_pompe@amicusrx.com.

The initial report must be as complete as possible. All known details of the SAE, and an assessment of the causal relationship between the event and ATB200 administered as a single agent, the use of ATB200 co-administered with AT2221, or study procedure should be included in the initial report. All information not available at the time of the initial report (eg, event end date, discharge summary, results of diagnostic procedures/evaluations related to the event, etc)

must be provided in a follow-up report. Follow-up information must be reported using the designated fax number within 24 hours of receipt at study sites.

Every attempt should be made to describe events in terms of a diagnosis. If a clear diagnosis has been made, individual signs and symptoms should not be recorded unless they represent atypical or extreme manifestations of the diagnosis, in which case they should be reported as separate events. If a clear diagnosis cannot be established, each sign and symptom must be recorded individually.

Study endpoints that meet SAE criteria (as per Section 11.1.2) must also be immediately reported by the investigator to Amicus or Amicus' representative but no later than 24 hours after any study site personnel is aware of the event.

If a nonserious event becomes serious, the event must be reported as described above.

If the investigator becomes aware of an SAE in a subject or receives an unsolicited report of an SAE from a subject more than 30 days after the last treatment, and considers the event possibly, probably, or definitely related to previous study treatment, she/he should contact the Amicus Medical Monitor to determine how the SAE should be documented and reported.

The Amicus Medical Monitor will determine the expectedness of AEs according to current safety reference document (eg, current investigator brochure or other safety-related information as available).

11.4.2. Additional Reporting Requirements for Suspected Unexpected Serious Adverse Reaction

Any AE that is serious, unexpected, and associated with the use of study treatment (SUSAR, also referred to as Expedited Safety Report, Investigational New Drug Safety Report), has additional reporting requirements. All investigators conducting clinical trials with the study treatment will be notified of such events and must inform their IRBs/ECs as required in accordance with local law. Amicus will ensure SUSARs are reported to regulatory agencies in accordance with local law in each country where study sites are located.

- If the SUSAR is fatal or life-threatening, associated with the use of study treatment, and unexpected, regulatory authorities and IRBs/ECs will be notified within 7 calendar days after Amicus learns of the event.
- If the SUSAR is not fatal or life-threatening but is otherwise serious, associated with the use of study treatment, and unexpected, regulatory authorities and IRBs/ECs will be notified within 15 calendar days after Amicus learns of the event.

Safety updates will be provided periodically to the regulatory authorities and IRBs/ECs responsible for the study according to the rules in effect in each country where study sites are located. These updates will include information on SUSARs and other relevant safety findings.

11.4.3. Additional Reporting Requirements for Serious Adverse Reaction

For subjects in this study that participate at more than one study site, Amicus drug safety personnel will forward a copy of the initial SAE report to any alternate site(s) attended by the subject. When all information related to the SAE is available, a MedWatch form will be sent to all sites at which the subject participates.

If additional information such as medical history (inclusive of FVC and 6MWT, plasma CK, ALT, AST, and urine Hex4 lab results) is obtained from the report or from a hospital discharge summary, the data should be reconciled by the primary site (site conducting Functional Assessment) and recorded in the eCRF accordingly.

11.5. Study Treatment Interruptions/Discontinuation Due to an Adverse Event

Investigators may choose to discontinue subjects from the study in the case of an AE or for administrative reasons. All discontinuations must be documented in the eCRF with the appropriate source documents and communicated to the Amicus Medical Monitor immediately.

If the reason for discontinuation of the study is an abnormal assessment (eg, ECG finding) or a laboratory test abnormality, the information must be recorded as an AE in the eCRF and source records.

11.6. Other Reporting Situations

11.6.1. Pregnancy

Pregnancy in and of itself is not regarded as an AE; however, pregnancy information on female subjects and female partner of male subjects, participating in this study, is collected by Amicus.

If a female subject becomes pregnant during the course of this study, or if the female partner of a male subject becomes pregnant during the subject's participation in the study, must be reported using Amicus' pregnancy notification form (subject to receipt of any data privacy release approvals where required under local privacy laws) within 5 working days of the investigator or study staff becoming aware of the pregnancy. If an SAE occurs in conjunction with the pregnancy, the same SAE reporting requirements described in Section 11.4.1 apply. Instructions regarding collection of pregnancy and outcome information will be provided by Amicus.

11.6.2. Overdose/Underdose

Any event associated with, or observed in conjunction with, a product overdose/under dose (whether accidental or intentional) is considered by Amicus to be an AE and must be reported as such. If a subject experiences an overdose (defined as ≥ 20% higher than the assigned dose of study treatment for that period in the protocol) or an underdose (defined as ≥ 20% lower than the assigned dose of study treatment for that period in the protocol) during the course of the study (whether symptomatic or not), Amicus Medical Monitor must be notified within 5 working days of the investigator or study staff first becoming aware of the overdose. Follow-up information must be forwarded on the outcome as applicable. If an SAE occurs in conjunction with the overdose, the same SAE reporting requirements described in Section 11.4.1 apply.

If a subject is unable to schedule a treatment (infusion) up to 7 days after the actual scheduled date, this is considered as a missed dose. The subject will receive the next dose per his/her schedule. In this case, the interval between 2 doses will be approximately 4 weeks. This is recorded as a protocol deviation and an adverse event. Amicus Medical Monitor must be notified within 5 working days of the investigator or study personnel first becoming aware of the missed dose. Follow-up information must be forwarded on the outcome as applicable. If an SAE occurs

in conjunction with the underdose, the same SAE reporting requirements described in Section 11.4.1 apply.

12. DATA ANALYSIS AND STATISTICAL CONSIDERATIONS

This section outlines the planned statistical analyses. Further details of the data handling rule and statistical calculations will be provided in a separate statistical analysis plan (SAP).

12.1. Endpoints

12.1.1. Primary Endpoints

Safety:

- Identification and counts of TEAEs, treatment-emergent SAEs, including IARs
- Changes from baseline in 12-lead ECG
- Changes from baseline in clinical safety laboratory evaluations: serum chemistry, hematology, and urinalysis
- Changes from baseline in PEs
- Changes from baseline in vital signs
- Changes from baseline in serum CK

PK for subjects from Cohort 1:

- Plasma GAA activity levels and total GAA protein concentration PK parameters: C_{max}, t_{max}, AUC_{0-t}, $AUC_{0-\infty}$, $t_{½}$, steady state volume of distribution (V_{ss}), and CL_T for each dose level
- Stage 1: Ratios of plasma GAA activity levels and total GAA protein concentration C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ for 20 mg/kg versus 5 mg/kg, 20 mg/kg versus 10 mg/kg, and 10 mg/kg versus 5 mg/kg
- Stage 2: Ratios of plasma GAA activity levels and total GAA protein concentration C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ for 20 mg/kg ATB200 (from Stage 1) versus single-dose 20 mg/kg ATB200 + 130 mg AT2221, and versus single-dose 20 mg/kg ATB200 + 260 mg AT2221
- Stage 2: Ratios of plasma GAA activity levels and total GAA protein concentration C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ for 20 mg/kg ATB200 + 130 mg AT2221 single-dose versus multiple-dose, and 20 mg/kg ATB200 + 260 mg AT2221 single-dose versus multiple-dose
- Plasma AT2221 PK parameters: C_{max}, t_{max}, AUC_{0-t}, $AUC_{0-\infty}$, $t_{½}$, total clearance following oral administration (CL_T/F), and terminal phase volume of distribution following oral administration (V_z/F) for each dose level
- Ratios of plasma AT2221 C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ for 260 mg single-dose versus 130 mg single-dose, and for 260 mg multiple-dose versus 130 mg multiple-dose
- Ratios of plasma AT2221 C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ for 130 mg multiple-dose versus 130 mg single-dose, and 260 mg multiple-dose versus 260 mg single-dose

12.1.2. Secondary Endpoints

Functional:

- For ambulatory subjects: change and percent change from Baseline in Gower's Maneuver, 4-stair-climb, 6MWT, 10-Meter Walk Test (10MWT), GSGC score, TUG, muscle strength tests (MRC and hand-held dynamometer [for both upper and lower limbs]), and pulmonary function tests (FVC, MIP, MEP, and SNIP [for subjects without invasive ventilatory support])
- For nonambulatory subjects: change and percent change from Baseline in muscle strength tests (MRC and hand-held dynamometer [upper limbs only]) and pulmonary function tests (FVC, MIP, MEP, and SNIP [for subjects without invasive ventilatory support])

Patient-reported Outcomes:

- Change and percent change from Baseline in FSS, RHS, and R-PAct Scale

Global Impression of Change:

- PGIC and SGIC

PK parameters for subjects from Cohorts 1 and 3:

- Plasma GAA activity levels and total GAA protein concentration PK parameters: C_{max}, t_{max}, AUC_{0-t}, $AUC_{0-\infty}$, $t_{½}$, CL_T and V_{ss}
- Plasma GAA activity levels and total GAA protein concentration ratios for C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ for single-dose versus multiple-dose
- Plasma AT2221 PK parameters: C_{max}, t_{max}, AUC_{0-t}, $AUC_{0-\infty}$, $t_{½}$, CL_T/F, and V_z/F
- Plasma AT2221 C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ ratios for single-dose versus multiple-dose
- Single-dose plasma GAA activity levels, total protein concentration and AT2221 C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ ratios for subjects from Cohort 3 versus subjects from Cohort 1 following 20 mg/kg ATB200 alone, 20 mg/kg ATB200 + 130 mg AT2221 single-dose, and 20 mg/kg ATB200 + 260 mg AT2221 single-dose

12.1.3. Exploratory Endpoints

- Anti-rhGAA antibodies (total and neutralizing)
- Cross-reactivity of anti-rhGAA antibodies to alglucosidase alfa
- Pro-inflammatory cytokines and other biomarkers of immune system activation
- PD biomarkers: urinary Hex4 and serum CK
- Evaluation of anti-rhGAA antibody impact on AUC and CL_T after at least 18 months of ATB200/AT2221 treatment as compared to AUC and CL_T after the first and third doses of 20 mg/kg ATB200 + 260 mg AT2221

12.2. Hypotheses

No formal statistical hypotheses are specified for this study.

12.3. Sample Size Considerations

No formal sample size calculation was performed. The sample size between 18 to 34 subjects is considered adequate for the purpose of this study.

12.4. Data Analysis Considerations

12.4.1. Analysis Populations

- Safety Population: this includes all enrolled subjects who have been exposed to at least 1 dose of study treatment (ATB200 and/or AT2221).
  - The Safety population will be used in the analysis of all safety parameters (including AEs, vital signs, laboratory assessments, and biomarkers).
- PK Analysis Population: includes enrolled subjects who have been exposed to at least 1 dose of study treatment (ATB200 and/or AT2221) and have completed at least 1 PK period.
  - The PK population will be used for PK analyses.
- Intent-to-Treat Population: includes all enrolled subjects.
- Other populations may be defined in the SAP.

12.4.2. Interim Analyses

Interim analyses will be performed in the study as needed. Full details for the interim analysis will be presented in the SAP.

12.4.3. PK/PD Analysis

12.4.3.1. Drug Concentration Data

Blood samples will be collected at serial time points specified in Table 14 for plasma GAA activity, plasma total GAA protein concentration, and plasma AT2221 concentration for all subjects. Plasma GAA activity levels, total GAA protein, and AT2221 concentrations will be used to compute PK parameters defined in Section 12.4.3.2.

12.4.3.2. PK Parameters

Plasma PK parameters will be computed by noncompartmental analysis with Phoenix® WinNonlin® version 6.4. For the purpose of calculating PK parameters, values below the limit of quantification (BLQ) will be set to 0 when a BLQ occurs before the first measurable concentration. If a BLQ occurs after a measurable concentration in a profile and is followed by a value above the lower limit of quantification, then the BLQ will be treated as “missing”. If a BLQ value occurs at the end of the blood sampling interval (after the last quantifiable concentration) it will also be treated as “missing”. For descriptive statistics of plasma activity

and concentration data, all BLQs will be set to 0 and included in designated statistical calculations. PK analyses performed on preliminary quality checked activity or concentration data will use nominal time points. PK analyses performed on final activity or concentration data will use actual time points. PK endpoints may include the following:

Plasma GAA activity levels and total GAA protein concentration PK parameters will be calculated as follows:

- C_{max}: Maximum observed activity level or concentration obtained directly from the activity or concentration profile
- t_{max}: Time of the first occurrence of the maximum observed activity level or concentration obtained directly from the activity or concentration profile
- K_{el} or λ_z: The apparent terminal phase elimination rate constant will be estimated by linear regression of logarithmically transformed concentration (or activity) versus time data. Only those data points which are judged to describe the terminal log-linear decline will be used in the regression.
- AUC_{0-t}: The area under the plasma drug concentration-time curve from 0 time (predose) to the time of last quantifiable concentration (t) will be calculated with the linear up/log-down trapezoidal method
- $AUC_{0-\infty}$: The area under the plasma drug concentration-time curve from 0 time (predose) extrapolated to infinite time will be calculated as follows:

  $$AUC_{0-\infty} = AUC_{0-t} + C_t/+\lambda_z$$

  Where C_t is the last observed quantifiable concentration
- $t_{½\beta}$: The beta-phase terminal elimination half-life, calculated as natural log (ln)[2]/λ_z from the last 3 or more quantifiable concentrations from the beta phase of elimination
- $t_{½\alpha}$: The alpha-phase terminal half-life, calculated as ln(2) / λ_z from the first 3 or more quantifiable concentrations after C_{max}. Estimated for plasma GAA activity levels and total GAA protein concentration only
- CL_T: total plasma clearance after IV administration, calculated as Dose/$AUC_{0-\infty}$
- V_{ss}: Steady-state volume of distribution from an IV administration, calculated as Dose•AUMC/$(AUC)^2$, where AUMC is area under the first moment curve

Plasma AT2221 PK parameters will include the parameters listed above in addition to the following:

- CL_T/F: Total plasma clearance from an oral administration, calculated as Dose/$AUC_{0-\infty}$
- V_z/F: Volume of distribution based on the terminal elimination phase from an oral administration, calculated as Dose/($AUC_{0-\infty} \cdot \lambda_z$)

12.4.4. Statistical Methods

12.4.4.1. Statistical Plan for PK Data

For the purpose of summarizing drug concentration/activity data, all BLQs will be set to zero (0) values. Drug concentration/activity data will be summarized with descriptive statistics including arithmetic means, standard deviations (SDs), coefficients of variation, median, minimum, and maximum, standard error of the mean, and 5^{th} and 95^{th} percentiles. Pharmacokinetic parameters will be summarized with descriptive statistics including N (number of subjects in the treatment population), arithmetic means, SDs, coefficients of variation, median, minimum, and maximum. Geometric means will be calculated for AUC_{0-t}, $AUC_{0-\infty}$, and C_{max}.

The following statistical analysis of variance (ANOVA) comparisons will be performed to assess the relative bioavailability on PK parameters C_{max}, AUC_{t-0}, and $AUC_{0-\infty}$ for plasma GAA activity levels and total GAA protein concentrations:

Cohort 1 Subjects:

- 20 mg/kg ATB200 + 130 mg AT2221 (single dose) versus 20 mg/kg ATB200 alone
- 20 mg/kg ATB200 + 260 mg AT2221 (single dose) versus 20 mg/kg ATB200 alone
- 20 mg/kg ATB200 + 130 mg AT2221 (single dose) versus 20 mg/kg ATB200 + 260 mg AT2221 (single dose)
- 20 mg/kg ATB200 + 130 mg AT2221 (multiple dose) versus 20 mg/kg ATB200 + 260 mg AT2221 (multiple dose)
- 20 mg/kg ATB200 + 130 mg AT2221 (single dose) versus 20 mg/kg ATB200 + 130 mg AT2221 (multiple dose)
- 20 mg/kg ATB200 + 260 mg AT2221 (single dose) versus 20 mg/kg ATB200 + 260 mg AT2221 (multiple dose)

Cohort 3 Subjects:

- 20 mg/kg ATB200 + 260 mg AT2221 (single dose) versus 20 mg/kg ATB200 + 260 mg AT2221 (multiple dose)

Cohort 3 versus Cohort 1 Subjects:

- 20 mg/kg ATB200 + 260 mg AT2221 (multiple dose) versus 20 mg/kg ATB200 + 260 mg AT2221 (multiple dose)
- 20 mg/kg ATB200 + 260 mg AT2221 (single dose) versus 20 mg/kg ATB200 + 260 mg AT2221 (single dose)
- 20 mg/kg ATB200 + 260 mg AT2221 (multiple dose) versus 20 mg/kg ATB200 alone
- 20 mg/kg ATB200 + 260 mg AT2221 (single dose) versus 20 mg/kg ATB200 alone

Following ln-transformation, plasma GAA activity levels and total GAA protein concentration C_{max}, $AUC_{0-\infty}$, and AUC_{0-t}, will be separately analyzed using a fixed effects model with fixed effect terms for treatment or subject group. Point estimates, and their associated 90% confidence

intervals (CIs) will be constructed for the differences, test treatment or subject group, and reference treatment or subject group. The point estimates and their associated 90% CIs will then be back-transformed to provide point estimates and 2-sided 90% CIs for the ratios of test/reference, on the original scale.

A dose proportionality assessment will be performed using the power method for 5, 10, and 20 mg/kg ATB200 in Stage 1 and for 130 and 260 mg AT2221 in Stage 2.

For the dose proportionality analysis, in Stage 1, the reference treatment will be 5 mg/kg ATB200, and the test treatments will be 10 mg/kg and 20 mg/kg ATB200 and, in Stage 2, the reference treatment will be 130 mg AT2221, and the test treatment will be 260 mg AT2221. For the ANOVA comparisons within Stage 2, the reference treatments will be single-dose, and the test treatments will be multiple-dose, and between Stages 1 and 2, the reference treatment will be 20 mg/kg ATB200 alone, and the test treatments will be 20 mg/kg ATB200 + 130 mg AT2221 and 20 mg/kg ATB200 + 260 mg AT2221. For between subject comparisons, the reference group will be Cohort 1 subjects, and test group will be Cohort 3 subjects.

For effect of anti-rhGAA antibody on total GAA protein exposures and plasma clearance, statistical comparisons between after at least 18 months of treatment in Stage 3 versus the first and third doses of treatment with 20 mg/kg ATB200 + 260 mg AT2221 will be performed on estimated plasma total GAA protein AUCs, plasma clearance, and 4-, 6-, 8-, and 12-hour plasma total GAA protein concentrations. PK data from both cohorts will be pooled for the statistical comparisons.

12.4.4.2. Efficacy Analyses

Details of the analysis methodology for all non-PK endpoints will be provided in the main SAP for the study.

In general, descriptive statistics will be provided for all efficacy endpoints. Continuous variables will be summarized using the number of observations (n), mean, standard deviation, median, minimum, and maximum, while categorical variables will be summarized using frequency and percentages. The absolute and percent changes from baseline for the continuous efficacy variables will be similarly summarized.

Each cohort will have its own baseline, and analysis will be performed by cohort and overall. In addition, summary analyses will be performed by combining Cohort 1, 3, and 4 for ambulatory subjects, or by combining Cohort 1 and 4 for ERT-experienced subjects. For Cohort 1, the analysis will also be provided by treatment dosing regimen. The visit values and the change from baseline to the post-baseline assessment will be analyzed by scheduled visit. The 95% CI (for the mean change from baseline and/or median difference) by scheduled visit will be provided for summary purposes only.

12.4.4.3. Safety Analyses

Adverse events and concomitant medications terms will be coded using Medical Dictionary for Regulatory Activities (MedDRA) version 16.1 or higher and WHO Drug Dictionary.

In general, TEAEs will be grouped by MedDRA System Organ Classes and by Preferred Terms. Summary statistics will be provided by cohort for AEs, ECG shift changes, vital signs shift changes, and clinical laboratory shift data based on the potentially CS values defined in

Appendix 2. In addition, summary statistics (n, mean, median, SD, minimum, and maximum) will be provided for values at each visit and changes from baseline at each visit for ECG, vital signs, and clinical laboratory assessments by cohort.

Prior and concomitant medications will also be summarized by cohort. A by-subject listing of protocol deviations will be provided.

12.4.4.4. Immunological Analyses

The effect of immunogenicity results on PK, PD, efficacy, and safety will be explored. Further details will be provided in the SAP. Assessments may include the following:

- Baseline anti-rhGAA antibody-positive (total and neutralizing) subjects as a percentage of the total number of subjects whose baseline samples were tested for anti-rhGAA antibodies
- Antibody range (median and interquartile range [IQR]) of the baseline anti-rhGAA antibody-positive samples (total and neutralizing)
- Percentage of baseline anti-drug antibody-positive subjects with significant increases in anti-rhGAA antibodies (total and neutralizing) after biologic drug administration (ie, when any one sample taken after the initial drug administration has an anti-rhGAA titer that is greater than the baseline titer by a scientifically reasonable margin)
- anti-rhGAA antibody-positive subjects at every visit as a percentage of the total number of subjects whose baseline samples were tested for anti-rhGAA (total and neutralizing)
- anti-rhGAA antibody-positive subjects at every visit as a percentage of the total number of subjects whose samples were tested at that visit for anti-rhGAA (total and neutralizing)
- Antibody range (median and IQR) of the anti-rhGAA antibody-positive samples at every visit (total and neutralizing)
- Kinetics and duration of anti-rhGAA antibodies over time in the study (total and neutralizing)
- Evaluation of high, mid, low, or no antibody groups (total and neutralizing) on plasma GAA activity levels and total GAA protein concentration C_{max} AUC, and CL over time in the study
- Evaluation of high, mid, low, or no antibody (total and neutralizing) groups on PD assessments (Hex4 and CK) over time in the study
- Cytokine levels and changes over time in the study

12.4.4.5. Baseline Demographic and Subject Characteristics

Baseline demographics and subject characteristics will be presented for all subjects included in the statistical analysis. Analyses will also be performed separately for subjects in Cohorts 1, 2, 3, and 4.

Functional assessments at baseline will be summarized for all subjects and will include the following:

- For subjects in Cohorts 1, 3, and 4: Motor function tests (Gower's Maneuver, 6MWT, 10MWT, GSGC, TUG, 4-stair climb), muscle strength tests (MRC and hand-held dynamometer [for both upper and lower limbs]), and PFTs (FVC, MIP, MEP, and SNIP)
- For subjects in Cohort 2: muscle strength tests (MRC and hand-held dynamometer [upper limbs only]) and PFTs (FVC, MIP, MEP, and SNIP [in subjects with noninvasive ventilatory support only])
- For all cohorts: PROs (FSS, RHS, and R-PAct Scale)

12.4.4.6. Subject Disposition

A summary table will be provided for subjects enrolled, included in all statistical analyses, completed each visit, discontinued the study, and reasons for discontinuation.

13. STUDY TREATMENTS

13.1. Description of Study Treatments

ATB200 (recombinant human acid α-glucosidase [rhGAA]) functions as ERT.

AT2221 (*N*-butyldeoxynojirimycin) is an iminosugar that functions as a selective pharmacological chaperone of GAA or rhGAA.

Table 20: Investigational Product

	Investigational Product	
Product Name	ATB200	AT2221
Dosage Form	lyophilized powder for intravenous infusion	hard gelatin capsule
Unit Dose	15 mg/mL	65 mg
Route of Administration	intravenous infusion	oral
Physical Description	sterile, nonpyrogenic, white to off-white lyophilized cake or powder supplied in single-use, clear 20 mL (cc) glass vials	White, size 2, hard gelatin capsules supplied in 40 cc HDPE bottles
Manufacturer	WuXi AppTec Biopharmaceuticals Co., Ltd 108 Meiliang Road Mashan, Binhu District, WuXi, China	AAI Pharma 1726 N. 23rd Street Wilmington, NC 28405 USA

13.2. Packaging and Labeling

AT2221 (65 mg) will be supplied by Amicus as hard gelatin capsules in plastic bottles and will be administered orally.

ATB200 (105 mg/vial) will be supplied by Amicus as a lyophilized powder in glass vials and will be administered by IV infusion upon reconstitution with Sterile Water for Injection and followed by dilution with 0.9% Sodium Chloride for Injection prior to administration. Sterile Water for Injection and 0.9% Sodium Chloride for Injection will be supplied by the study sites.

Each container will be labeled in conformance to regulatory requirements and, where applicable, local laws. All labels will be printed with the following information at a minimum: study identifier, identity of drug and dosage, sponsor name and contact details (and/or details of a local designee contact), dosing instructions, storage information, and other applicable local law statements. All labels will comply with legal requirements of each country.

13.3. Study Treatment Administration

13.3.1. ATB200

The doses of ATB200 are either 5, 10, or 20 mg/kg body weight.

ATB200 does not contain any preservatives. Vials are single-use only. Reconstituted vials should be retained at the site until the study monitor checks accountability.

In all stages, ATB200 is to be administered every 2 weeks as an approximate 4-hour IV infusion (± 15 minutes). Changes in the duration of ATB200 infusion due to safety or tolerability issues will be documented. The total volume of infusion is determined by the subject's body weight. Instructions for preparation for infusion, and the volume and rate of infusions will be provided in the Pharmacy manual. Intravenous administration of ATB200 (during all stages) should be performed by qualified study personnel. Any delegation of this responsibility must follow Section 15.4. Rate and duration of infusion must be constant for each infusion. Changes in the rate and/or duration of infusion due to safety or tolerability issues will be documented.

13.3.2. AT2221

The doses of AT2221 are either 130 mg or 260 mg administered orally, approximately 1 hour before ATB200 infusion. For a 130 mg dose, take 2 capsules. For a 260 mg dose, take 4 capsules.

At each visit in Stage 2, the administration of AT2221 oral capsule(s) 1 hour before the IV infusion of ATB200 should be supervised by the investigator or a qualified designee. Subjects should fast for at least 2 hours before and 2 hours after administration of AT2221.

13.4. Study Treatment Accountability

In accordance with local regulatory requirements, the investigator, designated site staff, or head of the medical institution (where applicable) must document the amount of investigational product dispensed and/or administered to study subjects, the amount returned by study subjects, and the amount received from and returned to Amicus (or designee), when applicable. Product accountability records must be maintained throughout the course of the study.

13.5. Handling, Storage, Return, and Disposal of Study Treatment

Sites will be instructed to store AT2221 65 mg oral capsules according to the conditions identified on the labels of each study treatment, at room temperature (15°C to 25°C/59°F to 77°F) with excursions permitted to (30°C/86°F) in a secure area, free from environmental extremes, and with restricted access.

Sites will be instructed to store ATB200 lyophilized powder vials according to the conditions identified on the label, at cold temperature (2°C to 8°C/36°F to 46°F) with short excursions permitted to (25°C/77°F) in a secure area, free from environmental extremes, and with restricted access.

A temperature log must be maintained for the duration of the study, and the temperature of the storage room (or cabinet) in which study treatment is stored must be recorded for each working day of the week that pharmacy staff (or designated study staff) is available. Temperature is required to be recorded using a standard Min-Max thermometer that has calibration records available (details usually contained within the device leaflet) and is maintained according to institutions standard maintenance policy.

Study treatment is to be stored only at the site(s) listed on the Form FDA 1572. Study treatment is to be dispensed only to subjects who have provided written informed consent, have met all entry criteria, and are assigned subject numbers.

The investigator, or appropriately assigned designee, will inventory and acknowledge receipt of all shipments of study treatments. The investigator must keep an accurate record/log of the quantities of study treatment dispensed and administered to each subject. The study monitor will periodically check the supplies of study treatment s held at the site to verify accountability of all study treatment s used and to verify the study treatment accountability logs are completed and maintained in the investigator study file. When instructed by the study monitor, the investigator will return all original containers of study treatments, whether empty, or containing used or unused study treatments to Amicus or their designee for destruction. Sites may not destroy study treatments on site unless Amicus has provided prior written approval.

14. STUDY MANAGEMENT

14.1. Documentation of Protocol-required Information and Study Findings

14.1.1. Electronic Case Report Form Completion

This protocol will use eCRFs with remote electronic data capture provided through a qualified third party vendor. The data will be entered on the eCRFs in a timely manner on an ongoing basis as defined in the Data Management Plan. The investigator, or designated representative, should complete the eCRF pages as soon as possible after information is collected, preferably on the same day that the subject is seen for an examination, treatment, or any other study procedure. Any outstanding entries must be completed immediately after the final examination. An explanation should be given for all missing data.

Details of eCRFs completion and correction will be explained to the investigator. The investigator is responsible for ensuring that data are properly recorded on each subjects eCRFs and related documents. If the investigator authorizes other persons to make entries into the eCRF pages, the names, positions, signatures, and initials of these persons must be supplied to Amicus.

The completed eCRF must be reviewed and electronically signed by the investigator who signed the study protocol signature page to ensure that the observations and findings are recorded on the eCRFs correctly and completely.

At the end of the study, eCRFs (including queries and audit trails) will be retained by Amicus and copies will be sent to the investigator to maintain as the investigator's copy.

14.2. Data Management

Only data specified in the protocol will be collected as part of this study. All eCRF data will be entered into the Electronic Data Capture (EDC) system managed by the contract research organization (CRO). Additional protocol specified data, such as laboratory and ECG data, may be collected through third party vendors and integrated with eCRF data by the CRO to create complete datasets for analysis. All protocol-specified data will be transmitted electronically to Amicus (or the CRO).

Management of clinical data will be performed in accordance with applicable Amicus standards and data cleaning procedures as defined in the Data Management Plan to ensure the integrity of the data (eg, determining errors and inconsistencies in the data, and ensuring data are corrected by site personnel or designees). Adverse events and concomitant medications terms will be coded using MedDRA version 16.1 or higher and WHOMD.

After database lock, each site will receive a compact disc (CD) containing all of their site-specific eCRF data as entered into the EDC system for the study, including full discrepancy and audit history. Additionally, a CD copy of all of the study center's data from the study will be retained by Amicus or designee for storage.

14.3. Safety Steering Committee (SSC)

An SSC is a committee consisting of medical experts from Amicus (the Amicus Medical Monitor), study investigators, and additional medical experts (as needed) formed to evaluate the safety data from sentinel dosing. Subsequent dosing in the study will proceed if the SSC identifies no significant safety issues or concerns that would preclude further treatment and assesses ATB200 co-administered with AT2221 sentinel dose as safe and well tolerated.

The formation and use of an SSC will be determined prior to the beginning of this study, or if, during the study, deemed necessary by Amicus. Specific details pertaining to the membership, roles, responsibilities, and procedures of the SSC will be documented in the SSC Charter. An SSC Charter will include operational and logistical procedures for the SSC.

14.4. Study Monitoring, Source Data Verification, and Onsite Audits

Monitoring and auditing procedures developed or endorsed by Amicus will be followed, in compliance with Good Clinical Practice (GCP) guidelines. Direct access to the onsite study documentation and medical records must be ensured by the investigator.

Monitoring will be done by personal visits from an Amicus representative (ie, study monitor) who will check the eCRFs for completeness and clarity, and crosscheck them with source documents. In addition to the monitoring visits, frequent communications (eg, letter, e-mail, telephone, and fax) by the study monitor will ensure that the study is conducted according to the protocol design and regulatory requirements.

Domestic and foreign regulatory authorities, the IEC/IRB, an auditor authorized by Amicus may request access to all source documents, eCRFs, and other study-related documentations for onsite audit or inspection. Direct access to these documents must be guaranteed by the investigator, who must provide support at all times for these activities. Medical records and other study documents may be copied during audit or inspection provided that the subject names are obliterated on the copies to ensure confidentiality.

The investigator should contact Amicus immediately if contacted by a regulatory authority regarding an inspection or audit.

14.5. Records Retention

The investigator must obtain approval in writing from Amicus before destruction of any records.

Essential documents should be retained until at least 2 years after the last approval of a marketing application in an International Conference on Harmonisation (ICH) region and until there are no pending or contemplated marketing applications in an ICH region, or at least 2 years have elapsed since the formal discontinuation of clinical development of ATB200 administered as a single agent and co-administered with AT2221. However, because of international regulatory requirements or country-specific requirements, Amicus may request retention for a longer period.

Prior to any decision regarding the disposal or destruction of study documents, the investigator should contact Amicus. Amicus may request that the site take alternative actions other than disposal or destruction of study documents.

Essential documents include:

- signed informed consent documents for all subjects
- subject identification code list*, screening log (if applicable), and enrollment log
- composition of the IEC/IRB (or other applicable statement) and record of all communications between the investigator and IEC/IRB as well as between the investigator and Amicus or CRO
- list of sub-investigators and other appropriately qualified persons to whom the investigator has delegated significant study-related duties, together with their roles in the study and their signatures
- copies of eCRFs and documentation of corrections for all subjects
- investigational product accountability records
- record of any body fluids or tissue samples retained
- all other source documents (eg, subject medical records, hospital records, laboratory records, etc)
- all other documents as listed in Section 8 of the ICH GCP E6 guidelines (ie, Essential Documents for the Conduct of a Clinical Trial)

Normally, these records will be held in the investigator's archives. If the investigator is unable to meet this obligation, he or she must ask Amicus for permission to make alternative arrangements. Details of these arrangements must be documented.

14.6. Use of Study Findings

All information concerning ATB200 administered as a single agent and co-administered with oral AT2221 as well as any matter concerning the operation of Amicus, such as clinical indications for ATB200 administered as a single agent and co-administered with oral AT2221, its formula, methods of manufacture, and other scientific data relating to it, that have been provided by Amicus and are unpublished, are confidential, and must remain the sole property of Amicus. The investigator will agree to use the information only for the purpose of carrying out this study and for no other purpose, unless prior written permission from Amicus is obtained.

Amicus has full ownership of the data collected as part of the study.

By signing the clinical study protocol and the confidentiality agreement, the investigator agrees that the results of the study may be used for the purposes of national and international registration, publication, and information for medical and pharmaceutical professionals. The authorities will be notified of the investigator's name, address, qualifications, and extent of involvement.

*EU legislation requires this list to be maintained for a minimum of 15 years.

Amicus will ensure that a final report on the study is prepared. Amicus will ensure that the study findings are reported in a manner that complies with applicable requirements for reporting clinical study results.

As required by local regulation or by the IEC/IRB, a summary of the study will be submitted by Amicus to the regulatory authorities and by the investigator to the IEC/IRB.

14.7. Study Close-out

The end of study is defined as date of database lock.

The study must be closed at the site on completion. Study close-out will be performed by the study monitor upon closure of the study.

Completion or premature termination of the study will be reported by Amicus to the regulatory agency and by Amicus or by the investigator to the IEC/IRB as required by local regulation or by the IEC/IRB.

Furthermore, Amicus or the investigator has the right to close any study site at any time. As much as possible, premature closure would occur after mutual consultation.

Study materials must be returned, disposed of, or retained as directed by Amicus (Section 13.5 and Section 14.5).

15. STUDY CONDUCT CONSIDERATIONS

This global study will include both Investigational New Drug (IND) (US) and non-IND (foreign) sites. All investigators will be required to certify their compliance with both ICH E6 GCP and their respective country's applicable laws and regulations. Both IND and non-IND sites will be operating under a single protocol (ie, there will not be a separate protocol for non-IND sites). The sponsor will ensure the conduct, monitoring, auditing, recording, analysis, and reporting of clinical trial results are in accordance with ICH GCP, providing assurance that the data and reported results are credible and accurate and that the rights, safety, and well-being of trial subjects are protected.

15.1. Posting of Information on Publicly Available Clinical Trial Registers

Amicus will be responsible for registering this study in a public registry that meets the requirements specified by the International Council of Medical Journal Editors, such as www.clinicaltrials.gov.

15.2. Ethical and Legal Aspects

15.2.1. Subject Information and Informed Consent

Signed written informed consent is to be obtained from each subject prior to enrollment into the study. The document must be in a language understandable to the subject and must specify who informed the subject. Where required by local law, the person who informs the subject must be a physician.

The subject (subject's legally authorized representative) must be given the opportunity to read the informed consent document and have all their questions and concerns addressed before giving consent in writing. If the subject or subject's legally authorized subject representative is unable to read, oral presentation and explanation of the written informed consent form and information to be supplied to the subject must take place in the presence of an impartial witness. Consent must be confirmed at the time of consent orally and by the personally-dated signature of the subject or by a local legally-recognized alternative (eg, the subject's thumbprint or mark). Details about why oral presentation was used, how the information was presented, and how the subject provided consent must be described in the medical records.

The subject's consent must be confirmed at the time of consent by the personally dated signature (or thumbprint or mark) of the subject or subject's legally authorized representative or guardian and by the personally-dated signature of the person conducting the informed consent discussions. A copy of the signed informed consent will be given to the subject and the original will be retained by the site. An entry must be made in the subject's dated source documents to confirm that informed consent was obtained prior to any study-related procedures and that the subject received a copy of the signed informed consent.

The witness and the person conducting the informed consent discussions must also sign and personally date the consent document. Until a signed written consent has been obtained, the investigator will not undertake any measures specifically required for this study.

The investigator may inform the subject's primary physician about the subject's participation in the study if the subject has a primary physician and if the subject agrees to the primary physician being informed.

15.2.2. Ongoing Information for IEC/IRB

The information listed below must be submitted to IECs/IRBs according to timelines specified by individual IEC/IRB documented submission policies and procedures, or by local law. Submissions may be made by Amicus (or designee) or by the investigator. The parties responsible for submissions will be identified and documented prior to shipment of study treatments.

- AE information
- expedited safety reports
- periodic reports on the progress of the study

15.2.3. Compliance With Good Clinical Practice (GCP)

This study is to be conducted according to globally accepted standards of GCP (as defined in the ICH GCP E6 guidelines), in agreement with the current version of Declaration of Helsinki, and in keeping with local regulations.

15.2.3.1. Quality Control

In accordance with applicable regulations, GCP, and Amicus' procedures, Amicus or its designee will contact the site prior to the start of the study to review with the study staff the protocol, study requirements, and their responsibilities to satisfy regulatory, ethical, and Amicus' or its designee's requirements. When reviewing data collection procedures, the discussion will include identification, agreement, and documentation of data items for which the eCRF will serve as the source document.

Amicus or its designee will monitor the study to ensure:

- data are authentic, accurate, and complete
- safety and rights of subjects are being protected
- study is conducted in accordance with the currently approved protocol and any other study agreements, GCP, and all applicable regulatory requirements

The investigator and the head of the medical institution (where applicable) agrees to allow the monitor direct access to all relevant documents.

15.2.3.2. Quality Assurance

To ensure compliance with GCP and all applicable regulatory requirements, Amicus may conduct a quality assurance assessment and/or audit of the site records, and the regulatory agencies may conduct a regulatory inspection at any time during or after completion of the study. In the event of an assessment, audit or inspection, the investigator (and medical institution) must agree to grant the advisor(s), auditor(s), and inspector(s) direct access to all relevant documents and to allocate their time and the time of their staff to discuss the conduct of the study, any

findings/relevant issues, and to implement any corrective and/or preventative actions to address any findings/issues identified during the regulatory audit or inspection.

15.2.4. Subject Data Protection

Subject names will not be supplied to Amicus. A unique subject number will be recorded in the case report form, and if the subject name appears on any other document (eg, laboratory report), it must be obliterated on the copy of the document to be supplied to Amicus. Study findings stored on a computer will be stored in accordance with local data protection laws (eg, European Union [EU] Directive 95/46/EC, EU Directive 94/45/EC, and the General Data Protection Regulation (GDPR) (EU) 2016/679]). The subject will be informed that representatives of Amicus, IEC/IRB, or regulatory authorities may inspect their medical records to verify the information collected, and that all personal information made available for inspection will be handled in strictest confidence and in accordance with local data protection laws.

The investigator will maintain a personal subject identification list (subject numbers with the corresponding subject names) to enable records to be identified.

15.3. Clinical Study Protocols and Amendments

Any nonadministrative changes to the protocol, initiated either by Amicus or by the investigator, will require a formal amendment procedure. Approval of all amendments must be obtained from Amicus, relevant IEC/IRB, and regulatory authorities (in accordance with local requirements) prior to implementation. Changes to the administrative aspects of the study will not require formal protocol amendments or IEC/IRB approval, but can be treated as administrative amendments. However, the IEC/IRB should be kept informed of such changes. Changes in study staffing or contact information are examples of administrative changes not requiring formal protocol amendments.

Protocol deviations to eligibility criteria, addition or deletion of tests, dosing, duration of treatment, and/or any other aspect of the study design that may significantly impact subject safety or scientific integrity, are not permitted under GCP or by Amicus, unless necessary to eliminate an immediate hazard to the subject(s).

Where Amicus and/or investigator must take urgent safety measures to protect the subjects from an immediate hazard, a protocol deviation may be allowed prior to obtaining approval from the relevant IEC/IRB (and/or regulatory authorities) according to 21 Code of Federal Regulations (CFR) 312.30(b) (2). In such cases, Amicus and IEC/IRB must be notified within 1 business day.

Amicus and the relevant IEC/IRB, where required by local law, must be informed of all protocol deviations and violations, and the investigators shall document such protocol deviations and violations in subject source document and eCRF.

15.4. Delegation of Investigator Duties

The investigator should ensure that all persons assisting with this study are adequately qualified; informed about their study-related duties and functions, the protocol and any amendments to the protocol, and the study treatment.

The investigator should maintain a list of sub-investigators and other appropriately qualified persons to whom he or she delegates significant study-related duties.

Should the investigator delegate the supervision of the administration of study treatment to a designated person, the designee should have the appropriate medical qualifications to effectively conduct or supervise any potential resuscitation procedures.

15.5. Liability and Insurance

Liability and insurance provisions for this study are given in separate agreements.

15.6. Financial Disclosure

Before the start of the study, the investigator will disclose to Amicus any proprietary or financial interests he or she might hold in the investigational products, ATB200 and AT2221, or Amicus as outlined in the financial disclosure form provided by Amicus. The investigator agrees to update this information in case of significant changes during the study or within 1 year of its completion. The investigator also agrees that, where required by law or regulation, Amicus may submit this financial information to domestic or foreign regulatory authorities in applications for marketing authorizations.

Where required by regulation, the investigator, or Amicus on behalf of the investigator, will also disclose these financial interests to the IEC/IRB and the investigator will disclose his/her financial interests to the subjects in the informed consent information.

Where required by regulation, Amicus or investigator will also submit the financial arrangements for the study to the regulatory authorities or to the IEC/IRB.

Financial disclosures will be provided by each sub-investigator to whom the investigator delegates significant study-related responsibilities.

15.7. Protocol Adherence

Adherence to the protocol is required. Any significant changes to the clinical trial will be incorporated into a protocol amendment and submitted to the IEC/IRB (and, where applicable, regulatory authorities) for approval before implementation of the change(s).

Protocol deviations to inclusion/exclusion criteria, addition or deletion of tests, dosing and/or duration of treatment, or any other aspect of the study design that may significantly impact subject safety or erode data/scientific integrity, are not permitted under GCP or by Amicus unless necessary to eliminate an immediate hazard to the subject (or subjects). Where Amicus and/or investigator must take urgent safety measures to protect subjects from an immediate hazard, a protocol deviation may be allowed before obtaining approval from the relevant IEC/IRB (and/or regulatory authorities). In such cases, the IEC/IRB (and/or regulatory authorities) shall be notified of the need for the urgent protocol deviation, the measures taken, and the plan for further action. In all cases, reporting to IEC/IRBs and regulatory authorities will comply with applicable local requirements.

Amicus and IEC/IRB must be informed of protocol violations and the investigators shall document such protocol violations in subject source records and case report forms.

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APPENDIX 1. SAMPSON CRITERIA FOR ANAPHYLAXIS

Anaphylaxis is highly likely when any one of the following 3 criteria are fulfilled:

1. Acute onset of an illness (minutes to several hours) with involvement of the skin, mucosal tissue, or both (eg, generalized hives, pruritus or flushing, swollen lips-tongue-uvula)
   AND AT LEAST ONE OF THE FOLLOWING
   a. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced PEF, hypoxemia)
   b. Reduced BP or associated symptoms of end-organ dysfunction (eg, hypotonia [collapse], syncope, incontinence)
2. Two or more of the following that occur rapidly after exposure *to a likely allergen for that patient* (minutes to several hours):
   a. Involvement of the skin-mucosal tissue (eg, generalized hives, itch-flush, swollen lips-tongue-uvula)
   b. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced PEF, hypoxemia)
   c. Reduced BP or associated symptoms (eg, hypotonia [collapse], syncope, incontinence)
   d. Persistent gastrointestinal symptoms (eg, crampy abdominal pain, vomiting)
3. Reduced BP after exposure to *known allergen for that patient* (minutes to several hours):
   a. Infants and children: low systolic BP (age specific) or greater than 30% decrease in systolic BP*
   b. Adults: systolic BP of less than 90 mm Hg or greater than 30% decrease from that person's baseline

PEF, Peak expiratory flow; *BP*, blood pressure.
*Low systolic blood pressure for children is defined as less than 70 mm Hg from 1 month to 1 year, less than (70 mm Hg + [2 × age]) from 1 to 10 years, and less than 90 mm Hg from 11 to 17 years.

Source: Sampson, Muñoz-Furlong et al. 2006

APPENDIX 2. POTENTIALLY CLINICALLY SIGNIFICANT GUIDELINES

Criteria for identifying abnormalities as potentially CS are based on the Guidelines for the Division of Neuropharmacological Drug Products, US Food and Drug Administration (revised on April 2, 1987). The laboratory values identified below are not inclusive of all the safety laboratory analytes that will be assessed during this clinical trial. Safety laboratory analytes which are known to be elevated in some adults with Pompe disease (eg, AST, ALT, LDH) and safety laboratory analytes which are not listed below will be reviewed to determine if they are to be noted as potentially CS based on the magnitude of the out of range value as compared to the normal range and the subject's baseline value, and whether the out of range value is coded as CS by the principal investigator.

Laboratory Values			
Variable		**Criterion Values**	
		Standard Units	**SI Units**
Chemistry			
SGOT (AST)		≥ 3 x Upper Limit Normal	
SGPT (ALT)		≥ 3 x Upper Limit Normal	
Alkaline Phosphatase		≥ 3 x Upper Limit Normal	
LDH		≥ 3 x Upper Limit Normal	
BUN		≥ 30 mg/dL	≥ 10.7 μM
Creatinine		≥ 2.0 mg/dL	≥ 176.8 μM
Uric Acid	Male	≥ 10.5 mg/dL	≥ 624.6 μM
	Female	≥ 8.5 mg/dL	≥ 505.6 μM
Bilirubin (Total)		≥ 2.0 mg/dL	≥ 34.2 μM
Hematology			
Hematocrit	Male	≤ 37%	
	Female	≤ 32%	
Hemoglobin	Male	≤ 11.5 g/dL	
	Female	≤ 9.5 g/dL	
Platelets		≤ 75,000/mm^3 or ≥ 700,000/mm^3	≤ 75 x 10^9/L or ≥ 700 x 10^9/L
Leukocytes		≤ 2,800/mm^3 or ≥ 16,000/mm^3	≤ 2.8 x 10^9/L or ≥ 16 x 10^9/L
Eosinophils		≥ 10%	
Neutrophils		≤ 15%	
Urinalysis			
Protein		Increase of ≥ 2 units	
Glucose		Increase of ≥ 2 units	
Casts		Increase of ≥ 2 units	

Abbreviations: BUN = blood urea nitrogen; LDH = Lactate dehydrogenase; SGOT (AST) = serum glutamic oxaloacetic transaminase (aspartate aminotransferase); SGPT (ALT) = serum glutamic pyruvic transaminase (alanine aminotransferase)

Vital Sign Values			
Variable	**Criteria**		**Change Relative to Baseline**
Heart Rate	≥ 120 bpm	and an	increase of ≥ 15 bpm
	≤ 50 bpm	and a	decrease of ≥ 15 bpm
Systolic Blood Pressure	≥ 180 mmHg	and an	increase of ≥ 20 mmHg
	≤ 90 mmHg	and a	decrease of ≥ 20 mmHg
Diastolic Blood Pressure	≥ 105 mmHg	and an	increase of ≥ 15 mmHg
	≤ 50 mmHg	and a	decrease of ≥ 15 mmHg
Weight			change of ≥ 7% body weight

ECG Values		
ECG parameter	**Low**	**High**
PR interval (ms)	< 120 ms	≥ 210 ms
QRS duration (ms)	≤ 50 ms	> 120 ms
QTcB interval (ms)	Not applicable	> 450 ms for males and > 470 ms for females and QTcB increases from baseline > 60 ms (males and females)
Ventricular rate (bpm)	A decrease from reference[a] ≥ 15 bpm, and an absolute value < 50 bpm	An increase from reference[a] ≥ 15 bpm, and an absolute value > 120 bpm

[a] Reference value: baseline or initial visit value, as appropriate.

APPENDIX 3. RASCH-BUILT POMPE-SPECIFIC ACTIVITY (R-PACT) SCALE

Study ATB200-02

Final R-PAct questionnaire Site ID | | | | | Subject ID | | | | | Visit Date D D M M M Y Y Y Y

	Are you able to:	No (0)	Yes, but with difficulty (1)	Yes, without difficulty (2)
1	Comb your hair?	☐	☐	☐
2	Eat (swallow, chew)?	☐	☐	☐
3	Pull on a pair of trousers (without closures)?	☐	☐	☐
4	Prepare a meal?	☐	☐	☐
5	Take a shower?	☐	☐	☐
6	Reach for and grasp an object above your head?	☐	☐	☐
7	Step over a threshold or negotiate obstacles in your path?	☐	☐	☐
8	Turn over in bed?	☐	☐	☐
9	Walk on an uneven surface?	☐	☐	☐
10	Stand up from a seated position?	☐	☐	☐
11	Walk more than 1km?	☐	☐	☐
12	Walk up and down a complete set of stairs?	☐	☐	☐
13	Bend over to pick something up off the ground and then stand up again?	☐	☐	☐
14	Walk at a rapid rate?	☐	☐	☐
15	Garden or carry out tasks in and around your yard?	☐	☐	☐
16	Practice a sport?	☐	☐	☐
17	Bend at the knee (squat) and then stand up again?	☐	☐	☐
18	Run (for example to catch a train)?	☐	☐	☐

APPENDIX 4. ROTTERDAM HANDICAP SCALE (RHS)

Studienummer:

Site ID Subject ID

Baseline

IPA/ Erasmus MC Pompe Survey

Visit Date

Rotterdam 9-items handicap scale

For each question, please tick the answer that describes your current situation best.

Regarding items 1 and 2: moving from room to room or outdoors does not necessarily mean that you have the ability to walk. For example, you can also move from room to room in a wheelchair.

1. Mobility indoors
Are you able to move from room to room, negotiating doors, carpets and polished surfaces?
0 = not applicable
1 = unable to move between rooms
2 = move between rooms mostly with help of another person
3 = move between rooms most of the time independent; sometimes needing help of another person
4 = move between rooms totally independent

2. Mobility outdoors
Are you able to move outdoors from one place to another, negotiating kerbs and uneven grounds?
0 = not applicable
1 = unable to move outdoors
2 = move outdoors mostly with help of another person
3 = move outdoors most of the time independent; sometimes needing help of another person
4 = move outdoors totally independent

3. Kitchen tasks
Are you able to fulfil tasks like making a pot of tea/ coffee, and serving it; are you able to collect items from a high and low cupboard, refrigerator, etcetera? (other kitchen tasks are also applicable)
0 = not applicable
1 = unable to fulfil any kitchen task
2 = able to fulfil only a minimum of these tasks; mostly needing help of another person
3 = able to fulfil the vast majority of these tasks independently; sometimes needing help of another person
4 = able to fulfil all kitchen tasks independently

4. Domestic tasks (indoors)
Are you able to fulfil house-cleaning tasks, such as vacuum cleaning, dishwashing, doing the laundry, dusting, etcetera?
0 = not applicable
1 = unable to fulfil any domestic tasks indoors
2 = able to fulfil only a minimum of these tasks; mostly needing help of another person
3 = able to fulfil the vast majority of these tasks independently; sometimes needing help of another person
4 = able to fulfil all domestic tasks indoors independently

5. Domestic tasks (outdoors)
Are you able to do the shopping, managing the garden, cleaning the car, etcetera?
0 = not applicable
1 = unable to fulfil any domestic tasks outdoors
2 = able to fulfil only a minimum of these tasks; mostly needing help of another person
3 = able to fulfil the vast majority of these tasks independently; sometimes needing help of another person
4 = able to fulfil all domestic tasks outdoors independently

Pagina 1

Studienummer:

Baseline
IPA/ Erasmus MC Pompe Survey

Site ID | Subject ID

Visit Date (D D M M Y Y Y Y)

6. Leisure activities (indoors)
Are you able to read a newspaper/magazine or a book, use the telephone, fulfil a hobby (other than sporting)?
0 = not applicable
1 = unable to fulfil these activities
2 = able to fulfil only a minimum of these activities; mostly needing help of another person
3 = able to fulfil the vast majority of these activities independently; sometimes needing help of another person
4 = able to fulfil all these activities independently

7. Leisure activities (outdoors)
Are you able to go to a party, theatre, movies, concerts, museums, meetings, participate in sport?
0 = not applicable
1 = unable to fulfil these activities
2 = able to fulfil only a minimum of these activities; mostly needing help of another person
3 = able to fulfil the vast majority of these activities independently; sometimes needing help of another person
4 = able to fulfil all these activities independently

Regarding item 8: For example, if you don't have a driving license, you can consider this part of the question as 'being fulfilled', unless it is clear that driving would be absolutely impossible due to your illness.

8. Drive a car/ go by bus/ ride a bicycle
Are you able to drive a car, go on a bus/ subway, or ride a bicycle?
0 = not applicable
1 = unable to fulfil any of these tasks
2 = able to fulfil only one of these tasks (if needed with help of another person)
3 = able to fulfil two of these tasks (if needed with help of another person)
4 = able to fulfil all these tasks independently

9. Work/ study
Are you able to fulfil your prior (before becoming ill) job/ study?
0 = not applicable
1 = unable to fulfil prior job/ study
2 = able to fulfil (partly) adapted job/ study
3 = able to fulfil partly the prior job/ study
4 = able to fulfil completely prior job/ study

Reference:
Merkies IS, Schmitz PI, Van Der Meché FG, Samijn JP, Van Doorn PA.
Psychometric evaluation of a new handicap scale in immune-mediated polyneuropathies.
Muscle Nerve. 2002 Mar;25(3):370-7.

Pagina 2

APPENDIX 5. FATIGUE SEVERITY SCALE (FSS)

Protocol ATB200-02

Site ID | Subject ID | Visit Date (D D M M M Y Y Y Y)

Fatigue Severity Scale

Below are a series of statements regarding your fatigue. By fatigue we mean a sense of tiredness, lack of energy or total body give-out. Please read each statement and choose a number from 1 to 7, where # 1 indicates you completely disagree with the statement and # 7 indicates you completely agree. Please answer these questions as they apply to the past **TWO WEEKS**.

	Completely Disagree						Completely Agree
1. My motivation is lower when I am fatigued	1	2	3	4	5	6	7
2. Exercise brings on my fatigue	1	2	3	4	5	6	7
3. I am easily fatigued	1	2	3	4	5	6	7
4. Fatigue interferes with my physical functioning	1	2	3	4	5	6	7
5. Fatigue causes frequent problems for me	1	2	3	4	5	6	7
6. My fatigue prevents sustained physical functioning	1	2	3	4	5	6	7
7. Fatigue interferes with carrying out certain duties and responsibilities	1	2	3	4	5	6	7
8. Fatigue is among my 3 most disabling symptoms	1	2	3	4	5	6	7
9. Fatigue interferes with my work, family, or social life	1	2	3	4	5	6	7

APPENDIX 6. PROMIS INSTRUMENTS

PROMIS Item Bank v1.0 – Dyspnea Severity – Short Form 10a

Dyspnea Severity – Short Form 10a

Please respond to each question or statement by marking one box per row.

	Over the past 7 days, how short of breath did you get with each of these activities?…	**No shortness of breath**	**Mildly short of breath**	**Moderately short of breath**	**Severely short of breath**	**I did not do this in the past 7 days**
DYSSV001	Dressing yourself without help	☐ 0	☐ 1	☐ 2	☐ 3	☐ X
DYSSV002	Walking 50 steps/paces on flat ground at a normal speed without stopping.....	☐ 0	☐ 1	☐ 2	☐ 3	☐ X
DYSSV003	Walking up 20 stairs (2 flights) without stopping.....	☐ 0	☐ 1	☐ 2	☐ 3	☐ X
DYSSV004	Preparing meals.....	☐ 0	☐ 1	☐ 2	☐ 3	☐ X
DYSSV005	Washing dishes	☐ 0	☐ 1	☐ 2	☐ 3	☐ X
DYSSV006	Sweeping or mopping	☐ 0	☐ 1	☐ 2	☐ 3	☐ X
DYSSV007	Making a bed.....	☐ 0	☐ 1	☐ 2	☐ 3	☐ X
DYSSV008	Lifting something weighing 10-20 lbs (about 4.5-9kg, like a large bag of groceries).....	☐ 0	☐ 1	☐ 2	☐ 3	☐ X
DYSSV009	Carrying something weighing 10-20 lbs (about 4.5-9kg, like a large bag of groceries) from one room to another.....	☐ 0	☐ 1	☐ 2	☐ 3	☐ X
DYSSV010	Walking (faster than your usual speed) for ½ mile (almost 1 km) without stopping.....	☐ 0	☐ 1	☐ 2	☐ 3	☐ X

Last Updated: 1 August 2016

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PROMIS Item Bank v1.0 – Fatigue – Short Form 8a

Fatigue – Short Form 8a

Please respond to each question or statement by marking one box per row.

	During the past 7 days…	**Not at all**	**A little bit**	**Somewhat**	**Quite a bit**	**Very much**
HI7	I feel fatigued	☐ 1	☐ 2	☐ 3	☐ 4	☐ 5
AN3	I have trouble starting things because I am tired.....	☐ 1	☐ 2	☐ 3	☐ 4	☐ 5
	In the past 7 days…					
FATEXP41	How run-down did you feel on average? ...	☐ 1	☐ 2	☐ 3	☐ 4	☐ 5
FATEXP40	How fatigued were you on average?.....	☐ 1	☐ 2	☐ 3	☐ 4	☐ 5
FATEXP35	How much were you bothered by your fatigue on average?.....	☐ 1	☐ 2	☐ 3	☐ 4	☐ 5
FATIMP49	To what degree did your fatigue interfere with your physical functioning?.....	☐ 1	☐ 2	☐ 3	☐ 4	☐ 5
	In the past 7 days…	**Never**	**Rarely**	**Sometimes**	**Often**	**Always**
FATIMP3	How often did you have to push yourself to get things done because of your fatigue?.....	☐ 1	☐ 2	☐ 3	☐ 4	☐ 5
FATIMP16	How often did you have trouble finishing things because of your fatigue?.....	☐ 1	☐ 2	☐ 3	☐ 4	☐ 5

02 Mar 2017

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PROMIS® Item Bank v2.0 – Physical Function – Short Form 20a

Physical Function – Short Form 20a

Please respond to each question or statement by marking one box per row.

		Without any difficulty	With a little difficulty	With some difficulty	With much difficulty	Unable to do
PFA11	Are you able to do chores such as vacuuming or yard work?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFA12	Are you able to push open a heavy door?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFA16r1	Are you able to dress yourself, including tying shoelaces and buttoning your clothes?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFA34	Are you able to wash your back?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFA38	Are you able to dry your back with a towel?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFA51	Are you able to sit on the edge of a bed?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFA55	Are you able to wash and dry your body?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFA56	Are you able to get in and out of a car?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFB19r1	Are you able to squeeze a new tube of toothpaste?	☐ 4	☐ 3	☐ 2	☐ 1	☐ 1
PFB22	Are you able to hold a plate full of food?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFB24	Are you able to run a short distance, such as to catch a bus?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFB26	Are you able to shampoo your hair?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1

29 November 2016

 Page 1 of 2

PROMIS® Item Bank v2.0 – Physical Function – Short Form 7a

Upper Extremity – Short Form 7a

Please respond to each question or statement by marking one box per row.

		Without any difficulty	With a little difficulty	With some difficulty	With much difficulty	Unable to do
PFA14r1	Are you able to carry a heavy object (over 10 pounds /5 kg)?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFA34	Are you able to wash your back?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFA36	Are you able to put on and take off a coat or jacket?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFB13	Are you able to carry a shopping bag or briefcase?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFB28r1	Are you able to lift 10 pounds (5 kg) above your shoulder?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFB34	Are you able to change a light bulb overhead?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1
PFM16	Are you able to pass a 20-pound (10 kg) turkey or ham to other people at the table?	☐ 5	☐ 4	☐ 3	☐ 2	☐ 1

11 July 2018

APPENDIX 7. PATIENT-REPORTED OUTCOMES: AVAILABLE LANGUAGES

Rasch-built Pompe-specific Activity (R-PAct) Scale	**Rotterdam Handicap Scale (RHS)**	**Fatigue Severity Scale (FSS)**	**PROMIS Dyspnea**	**PROMIS Fatigue**	**PROMIS Physical Functioning**	**PROMIS Upper Extremity**
English German Dutch	English Dutch German	English Dutch German	English Dutch German	English Dutch German	English Dutch German	English Dutch German (pending)

APPENDIX 8. PHYSICIAN GLOBAL IMPRESSION OF CHANGE (PGIC) AND SUBJECT GLOBAL IMPRESSION OF CHANGE (SGIC)

Physician (Clinician) Global Impression (PGIC)

Overall, taking into account the subject's symptoms and other neuromuscular functions, how would you rate his/her overall status today, relative to their Baseline Visit?

Please consider Pompe disease-related changes only, without respect to other factors.

❑ 1	❑ 2	❑ 3	❑ 4	❑ 5	❑ 6	❑ 7
Very Much Worse	Much Worse	Somewhat Worse	No Change	Somewhat Improved	Much Improved	Very Much Improved

Please **briefly** explain any indication of change, if possible:

_____

_____

_____

_____

Clinician's Signature:_____Date:_____

Site Number:_____Subject Number:_____

Subject Global Impression of Change (SGIC)

Note: The subject should be read the questions below by the person administering the questionnaire and then record the subjects' responses in the appropriate space below. At the end of the questionnaire both the subject and the administrator will sign the document.

We want to find out how you feel about the effects of the study drug in eight areas of life since you first started taking the medication and then sign in the space provided at the end. Please answer each of the next 8 questions.

1- Since you began taking the study drug, how would you describe your **overall physical wellbeing**? Please circle the number below that matches your current physical wellbeing compared to when you began study treatment.

❑ 1	❑ 2	❑ 3	❑ 4	❑ 5	❑ 6	❑ 7
Very Much Worse	Much Worse	Somewhat Worse	No Change	Somewhat Improved	Much Improved	Very Much Improved

Briefly explain your response in your own words: (Please limit your response to 4 lines).

_____

_____

_____

_____

2- Since you began taking the study drug, how would you describe your **effort of breathing**? Please circle the number below that matches your effort of breathing compared to when you began study treatment.

❑ 1	❑ 2	❑ 3	❑ 4	❑ 5	❑ 6	❑ 7
Very Much Worse	Much Worse	Somewhat Worse	No Change	Somewhat Improved	Much Improved	Very Much Improved

Briefly explain your response in your own words: (Please limit your response to 4 lines).

_____

_____

_____

_____

3- Since you began taking the study drug, how would you describe your **muscle strength**? Please circle the number below that matches your muscle strength compared to when you began study treatment.

❑ 1	❑ 2	❑ 3	❑ 4	❑ 5	❑ 6	❑ 7
Very Much Worse	Much Worse	Somewhat Worse	No Change	Somewhat Improved	Much Improved	Very Much Improved

Briefly explain your response in your own words: (Please limit your response to 4 lines).

_____

_____

_____

_____

4- Since you began taking the study drug, how would you describe your **muscle function**? Please circle the number below that matches your current physical muscle function compared to when you began study treatment.

❑ 1	❑ 2	❑ 3	❑ 4	❑ 5	❑ 6	❑ 7
Very Much Worse	Much Worse	Somewhat Worse	No Change	Somewhat Improved	Much Improved	Very Much Improved

Briefly explain your response in your own words: (Please limit your response to 4 lines).

_____

_____

_____

_____

5- Since you began taking the study drug, how would you describe your **ability to move around**? Please circle the number below that matches your current ability to move around compared to when you began study treatment.

❑ 1	❑ 2	❑ 3	❑ 4	❑ 5	❑ 6	❑ 7
Very Much Worse	Much Worse	Somewhat Worse	No Change	Somewhat Improved	Much Improved	Very Much Improved

Briefly explain your response in your own words: (Please limit your response to 4 lines).

_____

_____

_____

_____

6- Since you began taking the study drug, how would you describe your **activities of daily living** (for example, eating, dressing, bathing, etc.)? Please circle the number below that matches your current activities of daily living compared to when you began study treatment.

❑ 1	❑ 2	❑ 3	❑ 4	❑ 5	❑ 6	❑ 7
Very Much Worse	Much Worse	Somewhat Worse	No Change	Somewhat Improved	Much Improved	Very Much Improved

Briefly explain your response in your own words: (Please limit your response to 4 lines).

_____

_____

_____

_____

7- Since you began taking the study drug, how would you describe your **energy level**? Please circle the number below that matches your current energy level compared to when you began study treatment.

❑ 1	❑ 2	❑ 3	❑ 4	❑ 5	❑ 6	❑ 7
Very Much Worse	Much Worse	Somewhat Worse	No Change	Somewhat Improved	Much Improved	Very Much Improved

Briefly explain your response in your own words: (Please limit your response to 4 lines).

_____

_____

_____

_____

8- Since you began taking the study drug, how would you describe your **level of muscular pain?** Please circle the number below that matches your current level of muscular pain compared to when you began study treatment.

❑ 1	❑ 2	❑ 3	❑ 4	❑ 5	❑ 6	❑ 7
Very Much Worse	Much Worse	Somewhat Worse	No Change	Somewhat Improved	Much Improved	Very Much Improved

Briefly explain your response in your own words: (Please limit your response to 4 lines).

_____

_____

_____

_____

Date Administered:_____ Site Number:_____

Signature of Administrator:_____ Signature of Subject:_____

Name of Administrator:_____ Subject Number:_____