Protocol A6281323

A PHASE 3 MULTICENTER, OPEN LABEL, MULTI COHORT STUDY TO EVALUATE THE EFFICACY AND SAFETY OF SOMATROPIN IN JAPANESE PARTICIPANTS WITH PRADER-WILLI SYNDROME (PWS)

Statistical Analysis Plan
(SAP)

Version: 3

Date: 16 Dec 2022

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APPENDICES

1. VERSION HISTORY

Table 1. Summary of Changes

Version/ Date	Associated Protocol Amendment	Rationale	Specific Changes
3 16 Dec 2022	Amendment 1 13 Dec 2021	Some summaries were added.	6.6.1. Adverse Events Added the summaries for TEAEs of special interest. Overall: Minor edits for the description maintenance.
2 13 May 2022	Amendment 1 13 Dec 2021	Some clarifications were added.	Sections 3: Added the calculation expressions. Section 3.5.2: Added the table footer according to the protocol amendment. Section 4: Updated to the accurate description. Sections 6.5.3: Added the analysis for medication errors. Appendix 2: Applied the visit windows to safety analyses and re-calculated the target days and the day windows to weekly basis according to the protocol amendment. Overall: Minor edits for the description maintenance.
1 21 Oct 2020	Original 21 Aug 2020	N/A	N/A

2. INTRODUCTION

This statistical analysis plan (SAP) provides the detailed methodology for summary and statistical analyses of the data collected in Study A6281323. This document may modify the plans outlined in the protocol; however, any major modifications of the primary endpoint definition or its analysis will also be reflected in a protocol amendment.

This version of SAP specifically covers the analyses of data in screening and treatment period. Extension period is out of scope. For extension period, the analyses will be documented in another SAP or updated in this SAP.

2.1. Study Objectives, Endpoints, and Estimands

Objectives	Estimands	Endpoints
Primary:	**Primary:**	**Primary:**
• To evaluate the efficacy of somatropin in participants with PWS.	• Population: Japanese participants with PWS in each cohort; • Variable: Change from baseline to Month 12 in lean body mass (%) measured by DEXA; • Intercurrent events: All data after an intercurrent event (major deviation of diet/exercise, discontinuation of treatment, discontinuation of study, etc.), if collected, will not be considered; • Population level summary: Mean [Growth Hormone (GH) naïve pediatric and GH treated pediatric cohort] or LS mean (adult cohort).	• Change from baseline to Month 12 in lean body mass (%) measured by DEXA.
Secondary:	**Secondary:**	**Secondary:**
• To evaluate the effects of somatropin on other body composition paramaters.	• Population: Japanese participants with PWS in each cohort; • Variable: Each secondary endpoint; • Intercurrent events: All data after an intercurrent event (major deviation of diet/exercise, discontinuation of treatment, discontinuation of study, etc.), if collected, will not be considered; • Population level summary: Mean (GH naïve pediatric and GH treated pediatric cohort) or mean/LS mean (adult cohort).	• [KEY SECONDARY] Change from baseline to Month 12 in lean body mass (%) measured by BIA; • Change from baseline to Month 12 in body fat (%) measured by DEXA; • Change from baseline to Month 12 in adipose tissue distribution measured by abdominal CT; • Change from baseline to Month 6 in lean body mass (%) measured by DEXA (adult cohort only).
• To evaluate the safety and tolerability of somatropin.	• N/A.	• AE.

Objectives	Estimands	Endpoints
		• SAE. • Clinical laboratory values. • Bone maturation.
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[redacted]	[redacted]	[redacted]

2.1.1. Primary Estimand(s)

The estimand for the primary endpoint is the hypothetical estimand, which estimates the effect if all participants maintain their treatment and adhere to the protocol. It includes the following 4 attributes:

- Population: Japanese participants with PWS in each cohort;
- Variable: Change from baseline to Month 12 in lean body mass (%) measured by DEXA;
- Intercurrent events: All data after an intercurrent event (major deviation of diet/exercise, discontinuation of treatment, discontinuation of study, etc.), if collected, will not be considered;

- Population level summary: Mean (GH naïve pediatric and GH treated pediatric cohort) or LS mean (adult cohort).

2.1.2. Secondary Estimand(s)

The estimand for secondary endpoints is the hypothetical estimand, which estimates the effect if all participants maintain their treatment and adhere to the protocol. It includes the following 4 attributes:

- Population: Japanese participants with PWS in each cohort;
- Variable: Each secondary endpoint;
- Intercurrent events: All data after an intercurrent event (major deviation of diet/exercise, discontinuation of treatment, discontinuation of study, etc.), if collected, will not be considered;
- Population level summary: Mean (for GH naïve pediatric and GH treated pediatric cohort) or mean/LS mean (for adult cohort).

2.1.3. Additional Estimand(s)

Not Applicable

2.2. Study Design

This is a multicenter, open label, multi cohort study to evaluate the efficacy and safety of somatropin in Japanese participants with PWS. This study has 3 cohorts (GH naïve pediatric cohort, GH treated pediatric cohort and adult cohort) and all participants will receive somatropin.

The durations of the study periods are listed below:

- Screening Period: up to 28 days;
- Treatment Period: 12 months;
- Extension Period: 36 months or until regulatory approval, whichever is sooner.

The planned somatropin doses in this study are as below (the weekly dose should be divided into 6 or 7 subcutaneous injections):

- GH naïve pediatric cohort: 0.245 mg/kg/week.
- GH treated pediatric cohort: 0.084 mg/kg/week. The dosage may be adjusted according to participants' symptoms and serum IGF-1 levels but should not exceed 1.6 mg/day.

- Adult cohort: starting with 0.042 mg/kg/week, then titrated up to 0.084 mg/kg/week from Month 1. The dosage may be adjusted according to participants' symptoms and serum IGF-1 levels but should not exceed 1.6 mg/day.

A participant is considered to have completed the study if the individual participant has completed all phases of the study, including the last visit.

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

3. ENDPOINTS AND BASELINE VARIABLES: DEFINITIONS AND CONVENTIONS

Lean body mass (%) and body fat (%) will be calculated as the followings.

lean body mass (%) = lean body mass (kg) / [lean body mass (kg) + body fat (kg)]

body fat (%) = body fat (kg) / [lean body mass (kg) + body fat (kg)]

3.1. Primary Endpoint(s)

The primary endpoint for the study is change from baseline to Month 12 in lean body mass (%) measured by DEXA.

3.2. Secondary Endpoint(s)

The secondary endpoints for the study are as follows.

- [KEY SECONDARY] Change from baseline to Month 12 in lean body mass (%) measured by BIA.
- Change from baseline to Month 12 in body fat (%) measured by DEXA.
- Change from baseline to Month 12 in adipose tissue distribution measured by abdominal CT.
- Change from baseline to Month 6 in lean body mass (%) measured by DEXA (adult cohort only).

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[redacted]

IGF-1 SDS will be determined from the age and gender standards listed in the article by Isojima T. *et al.*[1].

3.4. Baseline Variables

Demographic and baseline characteristics include:

- Age.
- Sex.
- Ethnicity.
- Race.
- Duration of disease.
- Lean body mass (%) measured by DEXA and BIA.
- Body fat (%) measured by DEXA and BIA.
- Adipose tissue distribution measured by abdominal CT.
- IGF-1 SDS.
- BMI.
- Waist circumference.
- Vital Signs.
- Height, Height SDS, and Weight.

Height SDS will be determined from the age and gender standards listed in the national survey in year 2000[2] [based on data of the Ministry of Health, Labor and Welfare's Infant and Children's Growth Survey Report (0 to 6 years old) and the Ministry of Education, Culture, Sports, Science and Health Statistics Report (6 to 17 years old)].

3.5. Safety Endpoints

3.5.1. Adverse Events

Tretment-Emergent Adverse events (TEAEs) and Serious Adverse Events (SAEs) will be assessed.

3.5.2. Laboratory Data

The following safety laboratory tests will be collected.

Table 2. Protocol-Required Safety Laboratory Assessments

Hematology	Chemistry	Endocrinology
WBC count	**General condition**	**Pregnancy test**
Hemoglobin	Total protein	β-hCG[a]
Hematocrit	Albumin	
RBC count		**Growth hormone**
MCV	**Lipid**	IGF-1
MCH	Total cholesterol	
MCHC	Triglyceride	**Adrenal glands function**
Platelet count	HDL cholesterol	Cortisol
	LDL cholesterol	
		Thyroid function
	Liver function	TSH
	AST	FT4
	ALT	
	ALP	**Gonadal function**
	Total bilirubin	LH
		FSH
	Renal function	E2
	BUN	Testosterone
	Creatinine	
	Electrolyte	
	Calcium	
	Sodium	
	Potassium	
	Chloride	
	Glucose metabolism	
	HbA1c	
	Casual blood glucose[b]	

a. Both local serum and urine testing will be acceptable. A urine pregnancy test should have sensitivity of at least 25 mIU/mL. A serum hCG testing can be selected as an alternative to a serum β-hCG when clinically appropriate at the discretion of the investigator.
b. For participants with preexisting diabetes.

3.5.3. Other Safety Endpoints

- Vital Signs

  Vital signs will include temperature, systolic and diastolic blood pressure, and pulse rate.

- Bone Age

  Bone age will be assessed only in pediatric participants by an X-ray picture of the left hand and wrist. If the participant reaches bone age of 17 for boys and 15 for girls, the assessment is not necessary after that.

- Physical Examinations, including Behavioral and Mental Health Status

A physical examination will include, at a minimum, assessments of the cardiovascular, respiratory, gastrointestinal, and neurological systems. Assessments of the behavioral/mental health status will also be included.

- Height, Height SDS, and Weight

  Height and weight will also be measured and recorded. Height SDS will be calculated.

4. ANALYSIS SETS (POPULATIONS FOR ANALYSIS)

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

Participant Analysis Set	Description
Full Analysis Set (FAS)	All participants assigned to study intervention and who take at least one dose of study intervention.
Efficacy Evaluable Set	All participants assigned to study intervention and who take at least one dose of study intervention and have at least one efficacy evaluation.

Defined Analysis Set	Description
Adherence to protocol regimen	For participants who experience major deviation of diet/exercise or other major protocol deviation and/or discontinuation of treatment or study withdrawal, all data after the major deviation and discontinuation will not be considered.

5. GENERAL METHODOLOGY AND CONVENTIONS

The complete analyses will occur when all participants have completed the treatment period.

5.1. Hypotheses and Decision Rules

In this study, no formal hypothesis testing will be conducted.

The efficacy will be evaluated comprehensively based on individual data and summary statistics in each cohort for the primary, secondary, CCI [redacted] In addition, it will be confirmed in adult cohort that the point estimate of the least squares (LS) mean of the primary endpoint is above 0.

5.2. General Methods

5.2.1. Analyses for Binary Endpoints

The number, percentage and 95% confidence interval (CI) will be presented.

5.2.2. Analyses for Continuous Endpoints

Descriptive summary statistics (number, mean, standard deviation, median, minimum, and maximum) will be presented.

Least squares (LS) mean and its 95% CI of change from baseline will be estimated based on a Mixed-effect Model Repeated Measures (MMRM). Visit, baseline value, and baseline value-by-visit interaction are fixed effects in the model.

The restricted maximum likelihood (REML) estimation method will be used with an unstructured covariance matrix to describe the covariance matrix. The Kenward-Roger degrees of freedom will be used.

If there are convergence issues with the model, the default Newton-Raphson algorithm will be changed to the Fisher scoring algorithm in the initial value. In the case where there are still model convergence issues, the Toeplitz covariance matrix structures will be tried when repeated measures are 2-time points. When those are more than 2-time points, the following variance-covariance matrix structures will be tried in this particular order until convergence is obtained: Heterogeneous Toeplitz [TOEPH], Heterogeneous Autoregressive (1) [ARH(1)], Toeplitz [TOEP], Autoregressive (1) [AR(1)], and Compound Symmetry [CS].

5.2.3. Analyses for Categorical Endpoints

The frequency and percentage for each category will be presented.

5.3. Methods to Manage Missing Data

No imputation will be made for missing data.

6. ANALYSES AND SUMMARIES

Unless otherwise specified, data will be summarized by each cohort.

For change from baseline endpoints, it would require that a participant have a baseline value and at least 1 post baseline value to be included in the analysis for that endpoint.

6.1. Primary Endpoint(s)

6.1.1. Lean body mass (%) measured by DEXA at Month 12

6.1.1.1. Main Analysis

- Estimand strategy: Hypothetical (section 2.1.1).
- Analysis set: Efficacy Evaluable Set (Adherence to protocol regimen) (section 4).
- Analysis methodology: In adult cohort, change from baseline will be analyzed using a MMRM including data from Month 6, and 12 with visit, baseline value, and baseline value-by-visit interaction as fixed effects. In GH naïve pediatric and GH treated pediatric cohort, observed value and change from baseline will be summarized descriptively (section 5.2.2).

- Intercurrent events and missing data: All data after an intercurrent event (major deviation of diet/exercise, discontinuation of treatment, discontinuation of study, etc.), if collected, will not be considered. No imputation will be made for missing data.

- In adult cohort, the LS mean and its 95% CI will be presented. In GH naïve pediatric and GH treated pediatric cohort, the descriptive summary statistics will be presented.

6.1.1.2. Sensitivity/Supplementary Analyses

In adult cohort, observed value and change from baseline will be summarized descriptively.

6.2. Secondary Endpoint(s)

6.2.1. Lean body mass (%) measured by BIA at Month 12 [KEY SECONDARY]

6.2.1.1. Main Analysis

- Estimand strategy: Hypothetical (section 2.1.2).

- Analysis set: Efficacy Evaluable Set (Adherence to protocol regimen) (section 4).

- Analysis methodology: In adult cohort, change from baseline will be analyzed using a MMRM including data from Month 1, 3, 6, 9, and 12 with visit, baseline value, and baseline value-by-visit interaction as fixed effects. In GH naïve pediatric and GH treated pediatric cohort, observed value and change from baseline will be summarized descriptively (section 5.2.2).

- Intercurrent events and missing data: All data after an intercurrent event (major deviation of diet/exercise, discontinuation of treatment, discontinuation of study, etc.), if collected, will not be considered. No imputation will be made for missing data.

- In adult cohort, the LS mean and its 95% CI will be presented. In GH naïve pediatric and GH treated pediatric cohort, the descriptive summary statistics will be presented.

6.2.1.2. Sensitivity/Supplementary Analysis

In adult cohort, observed value and change from baseline will be summarized descriptively.

6.2.2. Body fat (%) measured by DEXA at Month 12

6.2.2.1. Main Analysis

- Estimand strategy: Hypothetical (section 2.1.2).

- Analysis set: Efficacy Evaluable Set (Adherence to protocol regimen) (section 4).

- Analysis methodology: In adult cohort, change from baseline will be analyzed using a MMRM including data from Month 6, and 12 with visit, baseline value, and baseline value-by-visit interaction as fixed effects. In GH naïve pediatric and GH

treated pediatric cohort, observed value and change from baseline will be summarized descriptively (section 5.2.2).

- Intercurrent events and missing data: All data after an intercurrent event (major deviation of diet/exercise, discontinuation of treatment, discontinuation of study, etc.), if collected, will not be considered. No imputation will be made for missing data.

- In adult cohort, the LS mean and its 95% CI will be presented. In GH naïve pediatric and GH treated pediatric cohort, the descriptive summary statistics will be presented.

6.2.2.2. Sensitivity/Supplementary Analysis

In adult cohort, observed value and change from baseline will be summarized descriptively.

6.2.3. Adipose tissue distribution measured by abdominal CT at Month 12

- Estimand strategy: Hypothetical (section 2.1.2).

- Analysis set: Efficacy Evaluable Set (Adherence to protocol regimen) (section 4).

- Analysis methodology: Observed value and change from baseline will be summarized descriptively (section 5.2.2).

- Intercurrent events and missing data: All data after an intercurrent event (major deviation of diet/exercise, discontinuation of treatment, discontinuation of study, etc.), if collected, will not be considered. No imputation will be made for missing data.

- The descriptive summary statistics will be presented.

6.2.4. Lean body mass (%) measured by DEXA at Month 6 (adult cohort only)

6.2.4.1. Main Analysis

- Estimand strategy: Hypothetical (section 2.1.2).

- Analysis set: Efficacy Evaluable Set (Adherence to protocol regimen) (section 4).

- Analysis methodology: Change from baseline will be analyzed using a MMRM including data from Month 6, and 12 with visit, baseline value, and baseline value-by-visit interaction as fixed effects (section 5.2.2).

- Intercurrent events and missing data: All data after an intercurrent event (major deviation of diet/exercise, discontinuation of treatment, discontinuation of study, etc.), if collected, will not be considered. No imputation will be made for missing data.

- The LS mean and its 95% CI will be presented.

6.2.4.2. Sensitivity/Supplementary Analysis

Observed value and change from baseline will be summarized descriptively.

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- CCI [redacted]

6.4. Subset Analyses

Not applicable

6.5. Baseline and Other Summaries and Analyses

6.5.1. Baseline Summaries

Demographics and baseline characteristics listed in section 3.4 will be summarized according to CDISC and Pfizer standards (CaPS) for FAS.

6.5.2. Study Conduct and Participant Disposition

Participants evaluation, disposition, discontinuation will be summarized according to CaPS.

6.5.3. Study Treatment Exposure

The number of dosing days and actual administered dosage will be summarized according to CaPS.

Medication error data will be reported in a list.

6.5.4. Concomitant Medications and Nondrug Treatments

Prior drug and non-drug treatment, concomitant drug and non-drug treatment will be summarized according to CaPS.

6.5.5. Correlation between DEXA and BIA

For lean body mass (%) and body fat (%), scatter plots of observed values measured by DEXA and BIA at the same visit will be presented. All cohorts and visits will be plotted in the same figure in different symbols. The correlation coefficient will be calculated.

6.6. Safety Summaries and Analyses

All analyses will be performed for FAS.

6.6.1. Adverse Events

Incidence of TEAEs and SAEs will be summarized according to CaPS.

Incidence of all-causality and treatment-related TEAEs of special interest will be summarized by SOCs and PTs. AEs of special interest will be defined in the safety narrative plan.

6.6.2. Laboratory Data

Laboratory data will be summarized in accordance with CaPS.

6.6.3. Other Safety Endpoints

- Vital Signs

  Vital sign data will be summarized in accordance with CaPS.

- Bone Age, Bone maturation

  Bone age and bone maturation will be reported in a list. Bone maturation will be calculated as bone age / chronological age.

- Physical Examinations, including Behavioral and Mental Health Status

  Physical Examinations will be reported in a list.

- Height, Height SDS, and Weight

  Observed value and change from baseline will be summarized according to CaPS.

7. INTERIM ANALYSES

7.1. Introduction

No formal interim analysis will be conducted for this study. As this is an open-label study, the sponsor may conduct unblinded reviews of the data during the course of the study for the purpose of safety assessment and/or supporting clinical development.

7.2. Interim Analyses and Summaries

Not applicable

8. REFERENCES

1. Standardized central curves and reference intervals of serum insulin-like growth factor-I (IGF-I) levels in a normal Japanese population using the LMS method. Endocrine Journal 2012, 59 (9), 771-780.

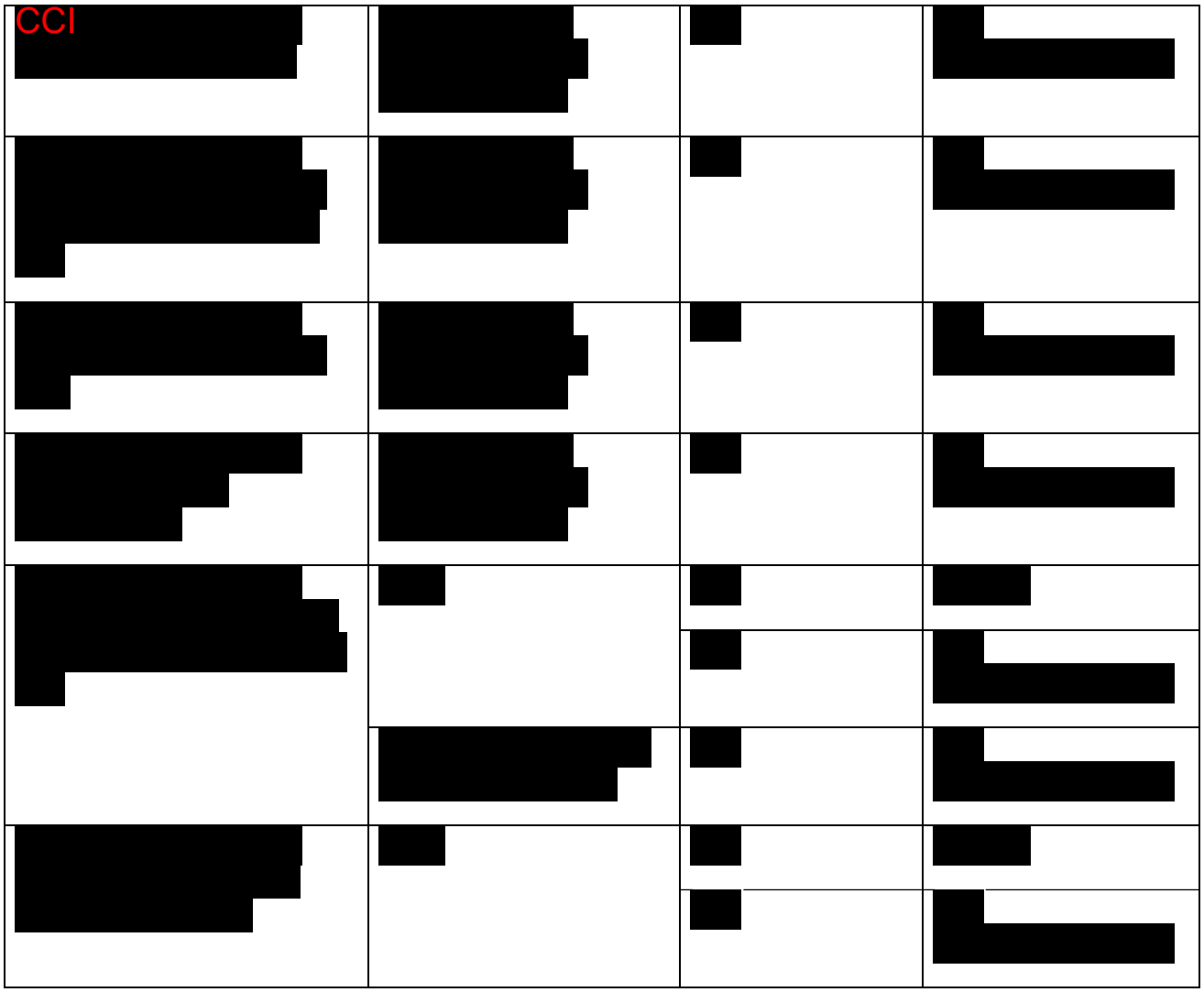
2. http://jspe.umin.jp/medical/files/fuhyo1.pdf (Last Access Date: 21 Oct 2020)

9. APPENDICES

Appendix 1. Summary of Efficacy Analyses

The table below summarizes analyses of efficacy endpoints. The efficacy evaluable set with adherence to protocol regimen will be used for all efficacy analyses.

Endpoint	Cohort	Analysis Type	Analysis Model
Primary			
Change from baseline to Month 12 in lean body mass (%) measured by DEXA	Adult	Main analysis	MMRM
		Sensitivity/supplementary analysis	N/A (Summary statistics)
	GH naïve pediatric and GH treated pediatric	Main analysis	N/A (Summary statistics)
Secondary			
Change from baseline to Month 12 in lean body mass (%) measured by BIA	Adult	Main analysis	MMRM
		Sensitivity/supplementary analysis	N/A (Summary statistics)
	GH naïve pediatric and GH treated pediatric	Main analysis	N/A (Summary statistics)
Change from baseline to Month 12 in body fat (%) measured by DEXA	Adult	Main analysis	MMRM
		Sensitivity/supplementary analysis	N/A (Summary statistics)
	GH naïve pediatric and GH treated pediatric	Main analysis	N/A (Summary statistics)
Change from baseline to Month 12 in adipose tissue distribution measured by abdominal CT	Adult, GH naïve pediatric, and GH treated pediatric	Main analysis	N/A (Summary statistics)
Change from baseline to Month 6 in lean body mass (%) measured by DEXA	Adult	Main analysis	MMRM
		Sensitivity/supplementary analysis	N/A (Summary statistics)
CCI			



Appendix 2. Definition and Use of Visit Windows in Reporting

The following visit windows will be used for summary analyses by visit labels.

Visit Label	Target Day	Definition [Day window]
Screening		Days -28 to -1
Day 1	1	Baseline
Month 1	28	Days 14 to 41
Month 2	56	Days 42 to 69
Month 3	84	Days 70 to 97
Month 6	182	Days 137 to 227
Month 9	273	Days 228 to 318
Month 12	364	Days 319 to 409

If two or more visits fall into the same window, keep the one closest to the Target Day. If two visits are equaled distant from the Target Day in absolute value, the later visit should be used.

Appendix 3. List of Abbreviations

Abbreviation	Term
AE	adverse event
ALP	alkaline phosphatase
ALT	alanine aminotransferase
AST	aspartate aminotransferase
β-hCG	beta-human chorionic gonadotropin
BIA	bioelectrical impedance analysis
BUN	blood urea nitrogen
CaPS	CDISC and Pfizer standards
CDISC	Clinical Data Interchange Standards Consortium
CI	confidence interval
CT	computed tomography
DEXA	dual energy X-ray absorptiometry
E2	estradiol
FAS	full analysis set
FSH	follicle-stimulating hormone
FT4	free thyroxine
GH	growth hormone
HbA1c	hemoglobin A1c
HDL	high-density lipoprotein
IGF-1	Insulin-like growth factor 1
LDL	low-density lipoproteins
LH	luteinizing hormone
LS	least-squares
MCH	mean corpuscular hemoglobin
MCHC	mean corpuscular hemoglobin concentration
MCV	mean corpuscular volume
MMRM	mixed-effect model repeated measures
PWS	Prader-Willi syndrome
RBC	red blood cell
REML	restricted maximum likelihood
SAE	serious adverse event
SAP	statistical analysis plan
SDS	standard deviation score
TEAE	treatment-emergent adverse event
TSH	thyroid stimulating hormone
WBC	white blood cell