A Long-Term Follow-up Study to Evaluate the Safety and Efficacy of Adeno-Associated Virus (AAV) Serotype 8 (AAV8)-Mediated Gene Transfer of Glucose-6-Phosphatase (G6Pase) in Adults with Glycogen Storage Disease Type Ia (GSDIa)

Protocol Number: 401GSDIA02

Original Protocol: 30 JAN 2019
Amendment 1: 17 FEB 2020
Amendment 2: 23 MAR 2021
Amendment 3: 06 OCT 2021
Amendment 4: 03 APR 2023

Brief Title:	Study of Long-Term Safety and Efficacy of Gene Therapy in Glycogen Storage Disease Type Ia
Product number:	DTX401
Indication:	Glycogen storage disease type Ia
IND Number:	17965
EudraCT Number:	2018-004473-27
EU CT Number:	2023-504004-29-00
Sponsor:	Ultragenyx Pharmaceutical Inc. 60 Leveroni Ct Novato, CA 94949 USA
Medical Monitor:	PPD PPD Telephone Number: PPD Fax Number: PPD

This study will be performed in compliance with the protocol, Good Clinical Practice (GCP), and all applicable regulatory requirements and guidelines.

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STATEMENT OF COMPLIANCE

Protocol Title: A Long-Term Follow-up Study to Evaluate the Safety and Efficacy of Adeno-Associated Virus (AAV) Serotype 8 (AAV8)-Mediated Gene Transfer of Glucose-6-Phosphatase (G6Pase) in Adults with Glycogen Storage Disease Type Ia (GSDIa)

Protocol Number: 401GSDIA02, Amendment 4

INVESTIGATOR SIGNATURE:

I have read Protocol 401GSDIA02, Amendment 4. I agree to conduct the study as detailed in this protocol and in compliance with the Declaration of Helsinki, Good Clinical Practice (GCP), and all applicable regulatory requirements and guidelines.

Investigator Signature Date

Printed Name:

SPONSOR SIGNATURE:

As the Sponsor representative, I confirm that Ultragenyx Pharmaceutical Inc. will comply with all Sponsor obligations as detailed in this protocol and in compliance with the Declaration of Helsinki, Good Clinical Practice (GCP), and all applicable regulation requirements and guidelines. I will ensure that the Investigator is informed of all relevant information that becomes available during the conduct of this study.

PPD MD Date

PPD Global Clinical Development
Ultragenyx Pharmaceutical Inc.

PROTOCOL AMENDMENT HISTORY AND SUMMARY OF CHANGES

The following overview summarizes significant changes to the protocol.

Version	Date	Summary of Changes
Version 1.0	30 Jan 2019	Original Protocol
Version 2.0 (Amendment 1)	17 Feb 2020	An overview of changes includes the following: • Assessments for vector shedding was added only for subjects that did not demonstrated clearance in Study 401GSDIA01 (Section 11.3.11, Table 1, and Table 2) • Section 6.2.3 was updated to reflect the number of ongoing studies with DTX401 as of December 2019 • A complete physical examination was added to the Week 260 (or end of study) and Early withdrawal visit (Table 1 and Figure 2) • The recommended target carbohydrate range of the prefasting challenge dinner meal was decreased from 20-30 g to 15-20 g, and the prefasting challenge cornstarch dose was decreased from 35 g to 5 g, to minimize a potential insulin spike that could result in abrupt hypoglycemia and to normalize baseline glucose levels (Section 11.4.1 and Table 1) • Blood sample collection for measurement of glucose and lactate levels was added at the beginning of the controlled fasting challenge to provide baseline levels (Sections 11.3.7.1 and 11.4.1, and Table 1) • Instructions for glucose and lactate samples collected during the fasting challenge were corrected to state that samples should be prepared and transported in accordance with the site's standard procedures (Sections 11.3.7.1 and 11.4.1, and Table 1) • Continuous glucose monitoring was added to collect supplemental information on glucose level trends throughout the study (Sections 11.4.2 and 13.5.3, and Table 1) • The exploratory study objective and endpoint related to morning glucose level were modified to glucose level due to the addition of continuous glucose monitoring (Sections 7, 13.5.4, and 13.1.3, and Figure 2) • Collection of blood for measurement of lipid levels was added on the morning of hospital admission for the fasting challenge (Section 11.4.1 and Table 1) • Collection of blood for measurement of cortisol, fatty acid, glucagon, insulin, and ketone levels was added at the beginning and end of the fasting challenge, or more frequently at the Investigator's discretion (Sections 11.3.7.2 and 11.4.1, and Table 1) • Details regarding the collection of morning glucose levels were clarified, including instances in which a subject is unable to use the assigned continuous glucose monitor (Section 11.4.2 and Table 1) • An enzyme-linked immunospot (ELISPOT) assay was added to assess any long-term potential cell-mediated immune responses to DTX401 (Sections 8.1, 11.3, 11.3.9.1, 13.6, Figure 2, and Table 1) • Text was added to clarify that the reporting period for hypoglycemic events is throughout the study (Sections 11.3.6 and 13.4.2) • The table of Clinical Laboratory Parameters was updated for completeness (Table 2)

Version	Date	Summary of Changes
		• New health-related quality of life assessments (Patient Global Impression of Severity and Patient Global Impression of Change) were added to guide further development of these assessments (Section 11.6.1 and Table 1) • Language was added to clarify that a clinically significant change or abnormal vital sign measurement will be recorded as an AE or SAE if it is felt to be clinically significant in the medical and scientific judgment of the Investigator (Section 11.3.2) • Language was added to clarify that nonemergent hospitalization for cornstarch management will not be considered an SAE (Section 12.1.1) • Language was added to clarify that Investigators should rate AE intensity based on medical judgment and that the current version of the National Cancer Institute Common Terminology Criteria for Adverse Events can be used to guide the rating of an adverse event (Section 12.1.2) • Language was added to indicate that additional interim analyses may be conducted at the Sponsor's discretion (Section 13.7) • Language was added to further clarify the minimum report requirements for concomitant therapies, including any changes that occur in concomitant medication use, and to guidance on recording concomitant procedures (Section 10.5) • The type of sample (eg, blood) to be collected was specified (Sections 11.3.9, 11.3.8, and 11.3.10) • The frequency of assessing prescribed diet and diet intake was increased to monthly for the duration of the study (Section 11.8.2 and Table 1) • The frequency of assessing prescribed cornstarch (or equivalent) and cornstarch (or equivalent) intake was increased to monthly for the duration of the study. It was also noted that follow-up phone calls may occur regularly (approximately once a month) to further discuss if adjustments are for a subject's cornstarch use (Section 11.8.3 and 13.1.3, and Table 1) • Measurement of weight was added to all study visits for the duration of the study (Figure 2 and Table 1) • Magnetic resonance imaging was moved from safety assessments to efficacy assessments (Sections 8.1, 11.3, and 11.4.4) • A liver ultrasound assessment was added to visits occurring in between visits with magnetic resonance imaging assessments, specifically at Weeks 78, 130, 182, and 234 (Section 11.4.5, Figure 2, and Table 1) • Measuring the change from baseline in liver ultrasound results over time to 260 weeks following IV administration of DTX401, by dose level was added as an exploratory objective and endpoint (Sections 7 and 13.1.3) • Language regarding management of GSDIa was clarified (Section 6.1) • Language was added to further clarify the process of record retention, including specifying record retention for 25 years to align with regulatory requirements (Section 14.4.3) • The sample size was increased from between 6 and 12 subjects to approximately up to 18 subjects due to an expansion in the sample size of Study 401GSDIA01 (Sections 8.1.2 and 13.1.4)

Version	Date	Summary of Changes
		• Additional time points were added for the 24-hour urine collection, reflecting a collection at all inpatient visits or visits with a controlled fasting challenge (Figure 2 and Table 1) • The Schedule of Events table was updated to include a row header indicating which visits are inpatient and which visits are outpatient (Table 1) • The protocol was reformatted, and numbering of sections, figures, and tables was updated
Version 3.0 (Amendment 2)	23 Mar 2021	Changes include the following: • The Sponsor's address has been revised (Title Page). • The Coordinating Investigator for the study has been removed (Title Page and Section 13.2). • The Sponsor Contact was changed from PPD MD to PPD MD (Sponsor Signature). • The sample size was decreased from "approximately up to 18 subjects" to "approximately up to 12 subjects" based on current plans to end enrollment in Study 401GSDIA01 after Cohort 4 (Sections 2, 7.1.2, and 12.1.4). • The following changes were made to the controlled fasting challenge assessment (Table 1, Table 2, Sections 10.3.7.1, 10.3.7.2, 10.4.1, and 12.5.1): o The definition of euglycemia for the end of the controlled fasting challenge changed from ≥ 60 mg/dL (≥ 3.33 mmol/L) to ≥ 54 mg/dL (≥ 3.0 mmol/L). o The composition of the dinner meal was updated from a prespecified carbohydrate range (15–20 g) to a personalized meal for each subject, including a target carbohydrate range and overall composition in protein, fats and dietary fiber that is as close as possible to the subject's most current dinner prescription but not higher than the carbohydrate content of the dinner prescription recorded at their baseline visit in Study 401GSDIA01. o The prefasting cornstarch dose was updated from 5 g to match the respective subject's most recent cornstarch prescription and timing post dinner, but not higher than the prescribed amount recorded at the baseline visit in Study 401GSDIA01. o Blood samples on the morning of the controlled fasting challenge were updated to include clinical chemistry (including lipid levels), hematology, coagulation panel, and urinalysis, to be sent to the central laboratory, and samples for AST and ALT levels to be sent to the local laboratory for STAT analysis. o Blood sample assessments at the beginning and end of the controlled fasting challenge were updated to include cortisol, ACTH, free fatty acids, glucagon, insulin, C-peptide, growth hormone, IGFBP1, alanine and ketone levels (3-hydroxy butyrate), and the instructions regarding laboratory sample collection before and during the controlled fasting challenge have been revised for clarity. o A final sample for glucose, lactate, growth hormone, IGFBP1, ACTH, and cortisol measurement has been added 30 minutes after the end of the controlled fasting challenge.

Version	Date	Summary of Changes
		○ Capillary glucose measurement has been added, to be performed at the same time points as blood collection for STAT analysis. • Language was added to specify that liver MRI will be conducted after the controlled fasting challenge is complete (Table 1; Figure 2; Section 10.4.4). • Text was added to clarify the duration of MRI image retention and the potential for MRI images to be analyzed to better understand the effect of DTX401 on GSDIa or other metabolic deficiencies (Section 10.4.4). • The contact information for SAE and pregnancy reporting was updated (Sections 11.1.4.2 and 11.4). • A new section was added to address changes to the study schedule and/or study operations due to the COVID-19 pandemic and the availability of remote visits to be performed by Firma Clinical Research is described (Section 7.3). • A telephone interview (Year 2 – Exit Interview) to better quantify the subject experience in the study has been added at Week 104 (or early withdrawal if the subject withdraws prior to Week 104), and added to the study endpoints (Table 1; Figure 2; Sections 2, 10.6.2, and 12.1.3). • Vector shedding sample collection requirements were clarified to include blood samples (Table 1; Sections 10.3.7 and 10.3.11) • The option for study visits at Weeks 65, 91, 130, 182, and 234 to be conducted via home healthcare and/or telemedicine has been added, if extenuating circumstances prevent the subject from attending an in-person clinical site visit. (Table 1; Section 7.1). • The option for a subject to ship the CGM receiver to the clinical site has been added, if a study visit is conducted remotely or skipped entirely for any reason (Table 1; Section 10.4.2). • Text referring to regulatory guidance has been revised to accommodate current guidance and future revisions (Section 13.1). • The Patient Global Impression of Change (PGI-C) has been added to the health-related quality of life assessments (Table 1; Sections 4, 10.6.1, and Appendix 5). • The numbering of sections was updated as necessary, and the in-text reference citations and list of references were reformatted to align with current Sponsor document styles.
Version 4.0 (Amendment 3)	06 Oct 2021	Changes include the following: • Addition of newly identified risk, based on available published literature (Sections 5.2.2 and 5.3) ○ Dorsal root ganglion and peripheral nerve pathologies have been added as a potential risk with DTX401, based on reports from nonclinical studies of other rAAV gene therapy products. Subjects will be monitored for this potential risk during targeted physical examinations and through collection of adverse event data. • Addition of peripheral and central nervous systems examination to the targeted examination (Table 1 and Section 10.3.4).
Version 5.0 (Amendment 4)	03 Apr 2023	Changes include the following:

Version	Date	Summary of Changes
		• The PPD Medical Monitor was changed from PPD MD to PPD MD (Title Page). • The Sponsor Contact was changed from PPD MD to PPD MD (Sponsor Signature). • Removed unnecessary planned study period detail from synopsis. • Additional detail around completed Study 401GSDIA01 has been added (Synopsis and Section 5.4). • Renamed 'exploratory' endpoints and objectives to be 'tertiary' endpoints and objectives (Synopsis, Section 6, and Section 12) to align with company standards. • Revised tertiary objectives to be more descriptive rather than repetitions of the tertiary endpoints (Synopsis, Section 6, and Section 12). • Updated primary, secondary, and tertiary endpoints so the assessments are analyzed by cohort and overall rather than dose level (Synopsis, Section 6, and Section 12). • Revised tertiary endpoint "urine albumin" to be "24-hour urine protein" (Synopsis, Section 6, and Section 12) for scientific accuracy. • Revised tertiary endpoint "change from baseline in MRI results of the liver" to "change from baseline in liver size and fat fraction (by MRI)" to specify what is being examined by MRI (Synopsis, Section 6, and Section 12). • Revised tertiary endpoint "average monthly use of cornstarch" to be "average daily use of cornstarch" (Synopsis, Section 6, and Section 12) to allow for more precise data collection. • Removed "the change from baseline in liver ultrasound results over time to 260 weeks following IV administration of DTX401" from the tertiary endpoint list (Synopsis, Section 6, and Section 12). • Updated the Week 130, 182, and 234 visits to be "OP/HH" instead of "InP/HH" (Table 1). • Added footnote limiting HRQoL assessments to PGI-S and PGI-C at the Week 156, 182, 208, 234, 260, 286, and 312 visits to reduce subject burden (Table 1). • Corrected a clerical error in Table 1: moved the Week 130 visit from under the "Follow-up: Year 2" column and into the "Follow-up Years 3-6 column." • Added Week 286 and 312 visits and explanation that subjects can roll into the DTX401 disease monitoring program (DMP) upon completion of Week 208 in Study 401GSDIA02 with Sponsor approval (Synopsis, Table 1, Section 6, Section 7.1.3.1, Section 7.1.3.5, Section 7.2, Section 10.3.1, Section 11.1.4.1, and Section 11.1.4.2) to ensure subject access to safety follow up ahead of the DMP's availability. • Added detail around integrating the analysis from Studies 401GSDIA01 and 401GSDIA02 (Synopsis and Section 12.3.1) for clarity on statistical analysis. • Added brief title and EU CT Number (Title Page) and new sections (Section 7.1.3.2, Section 7.1.3.3, Section 7.1.3.6, Section 7.1.4, and Section 13.1.4) to address EU CTR requirements. • Removed the controlled fasting challenge assessment (and associated assessments [24-hour urine collection, lipid levels, cortisol, ACTH, free

Version	Date	Summary of Changes
		fatty acids, glucagon, insulin, C-peptide, growth hormone, IGFBP1, alanine and ketone levels (3-hydroxy butyrate), dinner before controlled fasting challenge, cornstarch dose after dinner but before starting controlled fasting challenge, and glucose and lactate (venous) sample]) from Week 130, 182, and 234 visits (Table 1) to reduce subject burden. • Removed the liver MRI assessment from Weeks 104, 156, and 208 visits (Table 1 and Figure 2) to reduce subject burden. • Shifted liver ultrasound collection from Weeks 130, 182, and 234 to Weeks 104, 156, 208, 260, and 312 (Table 1 and Figure 2) to reduce subject burden. • Updated tertiary objectives and endpoints to include assessment of lactate levels (Synopsis, Section 6, and Section 12). • Updated Section 5.2.3 to reflect completed and ongoing clinical studies. • Updated Section 10.4.2 to include additional instruction around the pairing of CGM sensors and the study-issued receiver and the uploading of data from the receivers.

2. PROTOCOL SYNOPSIS

Name of Sponsor: Ultragenyx Pharmaceutical Inc. (Ultragenyx)

Ultragenyx Product: DTX401

Title: A Long-Term Follow-up Study to Evaluate the Safety and Efficacy of Adeno-Associated Virus (AAV) Serotype 8 (AAV8)-Mediated Gene Transfer of Glucose-6-Phosphatase (G6Pase) in Adults with Glycogen Storage Disease Type Ia (GSDIa)

Study Phase: Phase 1/2

Study Rationale:

The design of this study is consistent with global regulatory guidelines for protocol design. Study 401GSDIA02 is a long-term follow-up study to evaluate the safety and efficacy of adenoassociated virus (AAV) serotype 8 (AAV8)-mediated gene transfer of glucose-6-phosphatase (G6Pase) in adults with glycogen storage disease type Ia (GSDIa). Only subjects who received DTX401 in Study 401GSDIA01 are eligible to participate in Study 401GSDIA02. Study 401GSDIA01 was a Phase 1/2, open-label safety and dose-finding study of AAV8-mediated gene transfer of G6Pase in adults with GSDIa, during which subjects received a single intravenous (IV) dose of DTX401 with steroids (prednisone) to manage alanine aminotransferase (ALT) elevation. Overall, 12 patients were enrolled in following cohorts of 3 subjects each:

- Cohort 1: Dose 1 (2.0×10^{12} genome copies [GC]/kg) with a reactive steroid regimen (prednisone starting dose of 40 mg/day)
- Cohort 2: Dose 2 (6.0×10^{12} GC/kg) with a reactive steroid regimen (prednisone starting dose of 40 mg/day)
- Cohort 3: Dose 2 (6.0×10^{12} GC/kg) with an optimized reactive steroid regimen (prednisone starting dose of 60 mg/day)
- Cohort 4: Dose 2 (6.0×10^{12} GC/kg) with a prophylactic steroid regimen

No investigational product (IP) will be administered during Study 401GSDIA02.

Study Design:

In Study 401GSDIA01, subjects were followed for 1 year after dosing with DTX401. In Study 401GSDIA02, subjects will be followed for at least 3 additional years (and up to 5 additional years) for a total of 4-6 years of follow-up after administration of DTX401. Subjects will visit the study site approximately every 13 weeks during the first year of Study 401GSDIA02 and then approximately every 26 weeks through the end of the study at Week 312 (Year 6) for safety and efficacy evaluations. A long-term disease monitoring program (DMP) DTX401-CL401 is planned. Once the DMP is available to enroll, and after subjects reach the Week 208 visit in Study 401GSDIA02, subjects will transition to the DMP with Sponsor approval. The Schedule of Events is presented in Table 1.

Objectives:

Primary Objective

The primary objective of this study is as follows:

- To determine the long-term safety of DTX401 following a single IV dose in adults with GSDIa

Secondary Objective

The secondary objective of this study is as follows:

- To evaluate the long-term effect of DTX401 on symptom-free euglycemia in a setting of a controlled fasting challenge (CFC)

Tertiary Objectives

The tertiary objectives for this study are as follows:

- To evaluate the long-term effect of DTX401 on lipid profiles (total cholesterol, low-density lipoprotein, and triglycerides)
- To evaluate the long-term effect of DTX401 on uric acid
- To evaluate the long-term effect of DTX401 on proteinuria
- To evaluate the long-term effect of DTX401 on liver size and fat content
- To evaluate the long-term effect of DTX401 on hepatic adenoma progression
- To assess the long-term impact of DTX401 on cornstarch requirements
- To assess the long-term impact of DTX401 on glucose homeostasis
- To assess the long-term impact of DTX401 on the subject's health-related quality of life (HRQoL) and sleep quality
- To describe the long-term immune response to AAV8 capsid proteins after IV administration of DTX401
- To describe the long-term immune response to G6Pase after IV administration of DTX401
- To evaluate the long-term effect of DTX401 on lactate levels
- To evaluate the long-term effects of DTX401 on the endocrine and metabolic responses to fasting

Study Endpoints:

The primary endpoint is as follows:

- The incidence of AEs and SAEs for each cohort and overall assessed by severity and relationship to IP

The secondary endpoint is as follows:

- The change from Day 0 (Study 401GSDIA01) in time to first hypoglycemic event during a controlled fasting challenge over time by cohort and overall, following IV administration of DTX401

The tertiary endpoints are as follows:

- Total cholesterol, low-density lipoprotein, and triglycerides, by time point, cohort, and overall
- Uric acid, by time point, cohort, and overall
- 24-hour urine protein, by time point, cohort, and overall
- The change from baseline in liver size and fat fraction (by MRI) over time following IV administration of DTX401, by cohort, and overall
- The average daily use of cornstarch (or equivalent) over time, by cohort, and overall
- Assessment of serum glucose levels over time, summarized by cohort
- Subject responses to HRQoL and sleep quality assessments over time following IV administration of DTX401, by cohort and overall
- Year 2 – Exit Interviews
- The development of neutralizing antibodies to AAV8 (as determined by a cell-based assay) over time, by cohort, and overall
- The development of anti-AAV8 binding antibodies (as determined by ELISA) over time, by cohort, and overall
- The development of anti-G6Pase antibodies over time, by cohort, and overall
- Assessment of lactate levels over time, summarized by cohort and overall

Number of Sites: Up to 12 global study sites

Number of Subjects: Approximately 12 subjects

Study Population:

Individuals eligible to participate in this study must meet all of the following criteria:

1. Received DTX401 in Study 401GSDIA01
2. Willing and able to provide written informed consent after the nature of the study has been explained, and prior to any research-related procedures being performed
3. Willing and able to comply with all scheduled study visits, procedures, and requirements

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Planned or current participation in any other interventional clinical study that may confound the safety or efficacy evaluation of DTX401 during this study
2. Presence or history of any condition that, in the view of the Investigator, poses a risk to subject safety or places the subject at high risk of poor compliance or not completing the study or that would significantly affect the interpretation of study results

Investigational Product, Dosage, and Administration: Not applicable.

DTX401:

No IP will be administered during Study 401GSDIA02. All subjects enrolled in Study 401GSDIA02 will have received a single IV dose of DTX401 during their participation in Study 401GSDIA01.

Reference Therapy: Not applicable.

Duration of Study:

In Study 401GSDIA01, subjects were followed for 1 year after dosing with DTX401. In Study 401GSDIA02, subjects will be followed for at least 3 additional years (and up to 5 additional years) for a total of 4-6 years of follow-up after administration of DTX401. The duration of Study 401GSDIA02 for each subject is defined as the date the subject provides written informed consent through the Week 312 (Year 6) visit. Once the DMP is available to enroll, and after subjects reach the Week 208 visit in Study 401GSDIA02, subjects will transition to the DMP with Sponsor approval.

Procedures and Assessments:

Safety will be assessed based on adverse events (AEs), serious adverse events (SAEs), vital sign measurements, body weight, physical examination findings, ECG findings, documented symptomatic hypoglycemic events, clinical laboratory assessments (clinical chemistry, hematology, coagulation panel, and urinalysis), measurement of neutralizing antibody titer to AAV8, measurement of AAV8 binding antibody immunoglobulin G, measurement of cell-mediated immune response to AAV8 and G6Pase, measurement of antiG6Pase antibodies, and concomitant medications.

Efficacy will be assessed for each cohort and overall based on the change from Day 0 (Study 401GSDIA01) measurements in time to first hypoglycemic event during a controlled fasting challenge over time following administration of DTX401. Other efficacy assessments include average daily cornstarch use, glucose levels, liver imaging results, and HRQoL and sleep assessments. No control group will be enrolled in Study 401GSDIA02.

Statistical Considerations:

The study is expected to enroll approximately up to 12 subjects. This is dependent on the number of subjects who enrolled in Study 401GSDIA01 and who meet the eligibility criteria for Study 401GSDIA02. The sample size is not based on a power calculation.

No formal hypothesis will be tested. All statistical evaluations will be descriptive. An integrated analysis using data from both Study 401GSDIA01 and Study 401GSDIA02 will be performed to summarize and assess the long-term safety and efficacy of DTX401; there will not be a standalone analysis of data from only Study 401GSDIA02.

<u>Interim Analysis</u>

Administrative analyses may be performed during the study to support registrational activities, respond to regulatory inquiries, or at the Sponsor's discretion.

<u>Safety Analyses</u>

The incidence of AEs will be summarized for each cohort and overall by system organ class and preferred term. Additionally, AEs may be summarized for each cohort and overall by severity

and relationship to IP, if applicable. Serious AEs will be presented for each cohort and overall by relationship to IP. Summary tables will present the total numbers of AEs as well as the number of subjects with AE incidence by system organ class and preferred term. For incidence of relationship to IP, subjects will be counted only once, in the category of the strongest relationship to IP within each system organ class/preferred term. All other safety analyses will be summarized.

The safety database for Study 401GSDIA02 will include assessment results from Study 401GSDIA02 only.

Efficacy Analyses

Symptom-free euglycemia will be measured using the duration of controlled fasting challenge. The duration of the controlled fasting challenge is defined as time to any of the following three conditions: 1) glucose <54 mg/dL (<3.0 mmol/L); 2) signs/symptoms of hypoglycemia; 3) 15 hours fasting without hypoglycemia. The change from Baseline will be calculated using Day 0 in Study 401GSDIA01 as Baseline. All other efficacy analyses will be summarized, at minimum.

Table 1: Schedule of Events

Procedure	Period	–	Follow-up: Year 2[1]				Follow-up: Years 3-6[2]								Early Withdrawal
	Week	Visit 1 (Week 52)[3]	Week 65	Week 78	Week 91	Week 104	Week 130	Week 156	Week 182	Week 208	Week 234	Week 260	Week 286[4]	Week 312[4]	
	Visit Window (Days)	±7	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	N/A
	Visit Type	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP	OP	InP
Informed consent		X													
Eligibility criteria		X													
Medical history[6]		X													
Prior/concomitant medications, therapies, and procedures		X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X[7]
Vital sign measurements (heart rate, BP, RR)		X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X[7]
Body weight measurement		X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X[7]
24-hour urine collection		X		X		X		X		X		X			X
Clinical chemistry, hematology, coagulation panel, urinalysis		X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X[7]
Lipid levels		X[8]		X[8]		X[8]		X[8]		X[8]		X[8]		X[8]	X[8]
Cortisol, ACTH, free fatty acids, glucagon, insulin, C-peptide, growth hormone, IGFBP1, alanine and ketone levels (3-hydroxy butyrate)		X[9]		X[9]		X[9]		X[9]		X[9]		X[9]			X[9]
AAV8 neutralizing antibody testing (cell-based assay)		X[7]				X[7]		X[7]		X[7]		X[7]			X[7]

Procedure	Period	–	Follow-up: Year 2[1]				Follow-up: Years 3-6[2]								Early Withdrawal
	Week	Visit 1 (Week 52)[3]	Week 65	Week 78	Week 91	Week 104	Week 130	Week 156	Week 182	Week 208	Week 234	Week 260	Week 286[4]	Week 312[4]	
	Visit Window (Days)	±7	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	N/A
	Visit Type	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP	OP	InP
AAV8 binding antibody IgG testing (ELISA)		X[7]				X[7]		X[7]		X[7]		X[7]			X[7]
Cell-mediated immune response to AAV8 and G6Pase (ELISPOT)		X[7]				X[7]		X[7]		X[7]		X[7]			X[7]
Anti-G6Pase antibody assay		X[7]				X[7]		X[7]		X[7]		X[7]			X[7]
Complete physical examination		X[7]										X[7,10]	X[7,10]	X[7]	X[7]
Targeted physical examination[11]			X	X[7]	X	X[7]	(X)	X[7]	(X)	X[7]	(X)	X[7,10]	X[7,10]		
12-Lead ECG		X[7]													
Liver MRI[12]		X										X			X
Liver ultrasound				X[7]		X[7]		X[7]		X[7]		X[7]		X[7]	
AE/SAE monitoring		X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X	X[7]
Assessment of symptomatic hypoglycemic events		X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X[7]
Assessment of prescribed diet and dietary intake[13]		X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X[7]
Assessment of cornstarch (or equivalent) use[13]		X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X	X[7]	X[7]
CGM[14]		X	X	X	X	X	X	X	X	X	X	X	X	X	X

Procedure	Period	–	Follow-up: Year 2[1]				Follow-up: Years 3-6[2]								Early Withdrawal
	Week	Visit 1 (Week 52)[3]	Week 65	Week 78	Week 91	Week 104	Week 130	Week 156	Week 182	Week 208	Week 234	Week 260	Week 286[4]	Week 312[4]	
	Visit Window (Days)	±7	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	N/A
	Visit Type	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP	OP	InP
Collect and re-issue Morning Glucose Level monitoring worksheet[15]		X	X	X	X	X	X	X	X	X	X	X	X	X	X
Assessment of morning glucose levels[16]		X	X	X	X	X	X	X	X	X	X	X	X	X	X
Dinner before controlled fasting challenge[17]		X		X		X		X		X		X			X
Cornstarch dose after dinner but before starting controlled fasting challenge[18]		X		X		X		X		X		X			X
Controlled fasting challenge[19]		X		X		X		X		X		X			X
Glucose and lactate (venous) sample (local laboratory [STAT sample])[20]		X		X		X		X		X		X		X	X
Blood, saliva, urine, and stool for vector shedding (qPCR)[21]		X		X		X	X	X	X	X	X	X			X

Procedure	Period	–	Follow-up: Year 2[1]				Follow-up: Years 3-6[2]								Early Withdrawal
	Week	Visit 1 (Week 52)[3]	Week 65	Week 78	Week 91	Week 104	Week 130	Week 156	Week 182	Week 208	Week 234	Week 260	Week 286[4]	Week 312[4]	
	Visit Window (Days)	±7	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	±14	N/A
	Visit Type	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP/ HH[5]	InP	OP	OP	InP
HRQoL (PROMIS-29, PROMIS Social Isolation, PGI-S, PGI-C), and sleep quality (Pittsburgh Sleep Quality Index) assessments		X	X	X	X	X	X	X[22]	X[22]	X[22]	X[22]	X[22]	X[22]	X[22]	X
Endpoint Outcomes Interview (Year 2 - Exit Interview)[23]						X									X

Abbreviations: AAV8 = adeno-associated virus serotype 8; ACTH = adrenocorticotropic hormone ; AE = adverse event; BP = blood pressure; CGM = continuous glucose monitoring; ECG = electrocardiogram; ELISA = enzyme-linked immunosorbent assay; ELISPOT = enzyme-linked immunospot assay; EOS = end of study; EW = early withdrawal; G6Pase = glucose-6-phosphatase; HH = home healthcare visit; HRQoL = health-related quality of life; IGFBP1 = insulin-like growth factor-binding protein 1; IgG = immunoglobulin G; InP = inpatient; MRI = magnetic resonance imaging; N/A = not applicable; OP = outpatient; PGI-C = Patient Global Impression of Change; PGI-S = Patient Global Impression of Severity; RR = respiratory rate; SAE = serious adverse event.

Note: The periods and weeks in Study 401GSDIA02 begin at Year 2 and Week 52, respectively, and are relative to a time zero from the administration of DTX401 in Study 401GSDIA01. The study duration for Study 401GSDIA02 is up to 5 years. The end of study visit occurs at Week 208 or any visit thereafter upon availability of the DMP to enroll.

1 During the first year of Study 401GSDIA02, visits will occur approximately every 13 weeks.

2 After the first year of Study 401GSDIA02, visits will occur approximately every 26 weeks.

3 The first visit for Study 401GSDIA02 may coincide with the Week 52/EOS/EW visit for Study 401GSDIA01. Assessments required for Study 401GSDIA02 that are the same as those performed during the Week 52/EOS/EW visit for Study 401GSDIA01 do not need to be repeated if written informed consent for Study 401GSDIA02 is obtained before the Week 52/EOS/EW assessments are performed for Study 401GSDIA01. Assessments may be performed on more than 1 day provided that all assessments are completed, and results are available within the 7-day window.

4 Visit will only be conducted if a subject has not yet entered the DTX401 DMP. Visit can be outpatient but should be in-clinic.

5 At Weeks 65, 91, 130, 182, and 234, every effort will be made to conduct an on-site assessment. However, under extenuating subject circumstances that, in the Investigator's opinion, make an on-site visit not feasible, and after all reasonable measures to enable the subject's onsite visit have been exhausted, a home healthcare visit and/or telemedicine assessment may be conducted. At these time points, the assessments in parentheses "(X)" will not be performed if the visit is changed to a home healthcare visit.

[6] Any AEs that are ongoing at the completion of Study 401GSDIA01 will be considered medical history in Study 401GSDIA02.

[7] Assessments to be performed prior to the controlled fasting challenge.

[8] A blood sample for measurement of lipid levels will be collected on the morning of hospital admission for each controlled fasting challenge at approximately the same time starting at Day 0 (Study 401GSDIA01). The sample should be collected at least 2 to 4 hours after the subject's last meal.

[9] If possible, a blood sample for measurement of cortisol, ACTH, free fatty acids, glucagon, insulin, C-peptide, growth hormone, IGFBP1, alanine and ketone levels (3-hydroxy butyrate) will be collected at the beginning and end of the controlled fasting challenge, or more frequently at the investigator's discretion.

[10] A targeted physical examination will occur if a subject is not completing an end of study visit and a complete physical examination will occur if a subject is having an end of study visit.

[11] A targeted physical examination will include assessment of the skin, the abdomen with documentation of liver and spleen size, and the respiratory, cardiovascular, gastrointestinal, and peripheral and central nervous systems.

[12] An MRI of the liver will be performed to quantify glycogen and fatty liver content. The MRI will be performed after the controlled fasting challenge is complete for the Week 52 (Visit 1) only to avoid having to replace the CGM sensor prior to the fasting challenge.

[13] Assessment of daily prescribed diet and dietary intake as well as assessment of cornstarch (or equivalent) use will be reviewed at each study visit, and if possible, on a monthly basis through the final study visit. Follow-up phone calls may occur regularly (approximately once a month) to further discuss if adjustments are needed for a subject's cornstarch use.

[14] Supplemental information on glucose level trends will be collected using a CGM device. If subjects do not already have a study-issued CGM device (as these are provided in Study 401GSDIA01), a study-issued CGM will be provided. Subjects should bring their CGM device to each visit, containing CGM data from the 28 days prior to visit). If a study visit is conducted remotely (home healthcare visit or telemedicine), or if a study visit is skipped entirely for any reason, arrangements will be made for the subject to ship the CGM receiver (ideally, containing CGM data from the 28 days prior to the remote/missed visit) to the clinical site. Data from the subject's CGM device will be transferred to the Sponsor throughout the study, including a final data transfer at the end of the Week 312 visit or early withdrawal.

[15] If a subject is unable to use the assigned CGM, the subject should collect morning glucose levels at least 2 mornings per week throughout the study and record the values on the Morning Glucose Level monitoring worksheet provided at Visit 1. Subjects should measure their morning glucose level using their own glucose monitoring device before taking their morning dose of cornstarch. Subjects will be instructed to bring the completed worksheet to each study visit.

[16] If the subject was unable to use the assigned CGM but was able to complete the Morning Glucose Level monitoring worksheet, the subject's worksheet will be collected and reviewed at the time points specified in Table 1, and a new worksheet will be provided to the subject.

[17] Before the controlled fasting challenge begins, the study site will provide dinner for the subject. The dinner meal will be personalized for each subject, and will include a target carbohydrate range and overall composition in protein, fats and dietary fibers that is as close as possible to the subject's most current dinner prescription, but not higher in carbohydrate content than their dinner prescription recorded at their baseline visit in Study 401GSDIA01.

[18] After dinner, subjects will be given an oral dose of cornstarch equivalent per their most recent cornstarch prescription, at approximately the same time that they typically take it, but no later than 3 hours post dinner. The amount of cornstarch administered should not be higher than the prescribed amount recorded at their baseline visit in Study 401GSDIA01.

[19] In addition to the specified time points, if clinically indicated, the controlled fasting challenge can be performed at an unscheduled visit.

[20] During the controlled fasting challenge, blood samples for glucose and lactate (venous) will be collected at the beginning of the controlled fasting challenge and approximately every 60 minutes (±5 minutes) until glucose levels are at or below 70 mg/dL (≤3.9 mmol/L). Once glucose levels are at or below 70 mg/dL (≤3.9 mmol/L), blood samples for glucose and lactate (venous) will be collected approximately every 30 minutes (±5 minutes) until the subject's glucose level drops below 54 mg/dL (<3.0 mmol/L), the subject experiences symptoms of hypoglycemia per the Investigator's discretion, or the fast reaches 15 hours without hypoglycemia, whichever occurs first. Samples should be sent to the local laboratory (STAT sample) and results should be available within 30 minutes or less of the blood sample collection for subject safety. Samples should be prepared and transported to the local laboratory in accordance with the

site's standard procedures. Capillary glucose will be performed at the same time points as blood samples for STAT analysis of glucose and lactate. Results of the capillary glucose and readings from CGM at corresponding time points will be noted on a controlled fasting challenge results sheet. At Week 312 visit a STAT lactate will be collected. A STAT glucose will not be collected at the Week 312 visit.

[21] Blood, saliva, urine, and stool for vector shedding will only be collected for subjects who did not demonstrate clearance in Study 401GSDIA01. Samples for vector shedding analysis will be collected until at least 3 consecutive negative results are obtained for each sample matrix. '

[22] Only PGI-S and PGI-C will be collected at the visit.

[23] A 30-minute scripted telephone interview will be conducted by a third-party vendor, Endpoint Outcomes, to better quantify the subject experience in the study. The interview will be conducted at Week 104, or at the Early Withdrawal visit if the subject withdraws prior to Week 104.

3. TABLE OF CONTENTS

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4. LIST OF ABBREVIATIONS AND DEFINITION OF TERMS

Abbreviation	Definition or Explanation
AAV	adeno-associated virus
AAV8	adeno-associated virus serotype 8
ACTH	adrenocorticotropic hormone
AE	adverse event
ALT	alanine aminotransferase
AST	aspartate aminotransferase
BP	blood pressure
CFR	Code of Federal Regulations
CGM	continuous glucose monitoring
DMP	disease monitoring program
DRG	dorsal root ganglion
ECG	electrocardiogram
eCRF	electronic case report form
EDC	electronic data capture
ELISA	enzyme-linked immunosorbent assay
ELISPOT	enzyme-linked immunospot assay
EOS	end of study
EudraCT	European Union Drug Regulating Authorities Clinical Trials
EW	early withdrawal
FDA	Food and Drug Administration
G6P	glucose-6-phosphate
G6Pase	glucose-6-phosphatase (protein)
G6PC	glucose-6-phosphatase (gene)
G6pc$^{-/-}$	glucose-6-phosphatase knockout
GC	genome copies
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
GSDIa	glycogen storage disease type Ia
HCA	hepatocellular adenoma
HCC	hepatocellular carcinoma
HRQoL	health-related quality of life
ICF	informed consent form
ICH	International Council for Harmonisation

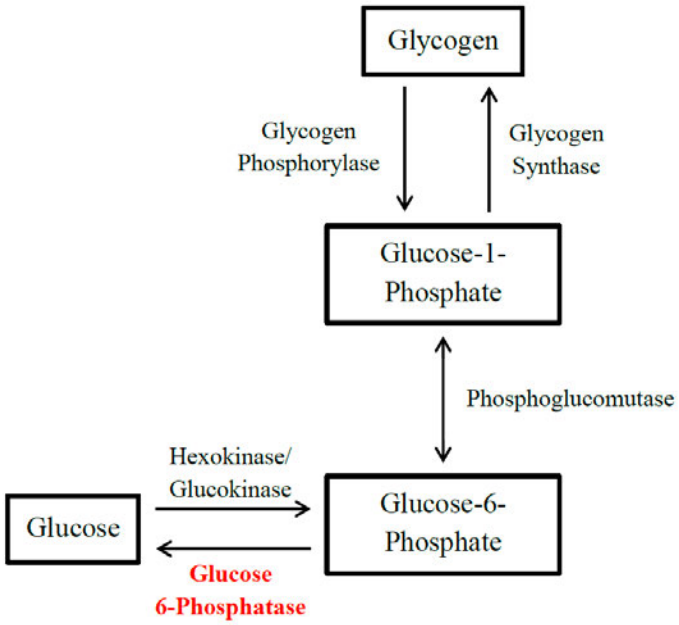
Abbreviation	Definition or Explanation
IEC	Independent Ethics Committee
IGFBP1	insulin-like growth factor-binding protein 1
IgG	immunoglobulin G
IND	Investigational New Drug (application)
InP	inpatient
IP	investigational product
IRB	Institutional Review Board
IV	intravenous
LFT	liver function test
MRI	magnetic resonance imaging
OP	outpatient
PGI-S	Patient Global Impression of Severity
PGI-C	Patient Global Impression of Change
RR	respiratory rate
SAE	serious adverse event
SAP	statistical analysis plan
SUSAR	suspected unexpected serious adverse reaction

5. BACKGROUND

5.1. Overview of GSDIa

Glucose-6-phosphatase is a glucose-6-phosphate (G6P) hydrolase located in the endoplasmic reticulum that is primarily expressed in the liver, kidneys, and intestine. It is a highly hydrophobic protein with 9 transmembrane domains (Pan et al., 1998). The amino and carboxyl termini of the glucose-6-phosphatase (G6Pase) protein are located on opposite sides of the endoplasmic reticulum membrane, with the active site of the enzyme facing the endoplasmic reticulum lumen (Lei et al., 1993; Lei et al., 1995b; Lei et al., 1995c). Glucose-6-phosphatase is responsible for catalyzing the terminal step in hepatic and renal glycogenolysis and gluconeogenesis (Figure 1): the hydrolysis of G6P to glucose and inorganic phosphate provides a significant amount of circulating glucose (Foster and Nordlie, 2002). In patients with glycogen storage disease type Ia (GSDIa), free glucose molecules are not generated for export into the body's circulation, and as a result, glucose from glycogen stores is unavailable during periods of fasting, resulting in severe hypoglycemia. The excess G6P molecules are shunted toward the production of triglycerides (eventually stored as fat) and the pentose-phosphate pathway (leading to increased uric acid production). Additionally, during periods of prolonged hypoglycemia, an increased rate of glycolysis and catabolism of muscle and fats results in a build-up of lactate (Chou and Mansfield, 1999; Chou, 2001; van Schaftingen and Gerin, 2002).

Figure 1: Overview of Glycogenesis and Glycogenolysis



Glycogen storage disease type Ia, also known as von Gierke disease, is a severe autosomal recessive inborn error of metabolism that is caused by mutations of the glucose-6-phosphatase (*G6PC*) gene located on chromosome 17q21 (Lei et al., 1993; Chou, 2001). Patients affected by GSDIa present with severe hypoglycemia and seizures at a few months of age. Like many other inherited disorders, GSDIa can be readily detected by screening a bloodspot sample for one of several common mutations (Lei et al., 1995a). The importance of diagnosing and treating GSDIa early is emphasized by the severe complications that arise when the disease is left untreated,

including life-threatening hypoglycemia, severe lactic acidosis, growth failure, renal Fanconi syndrome, and pancreatitis. Acute complications of GSDIa, such as hypoglycemia, severe metabolic acidosis, and renal tubular dysfunction, respond favorably to and can be prevented with strict dietary restrictions and frequent consumption of uncooked cornstarch. However, these restrictions have significant limitations and require constant vigilance to avoid acute metabolic decompensation that is accompanied by life-threatening hypoglycemia and lactic acidosis. In addition, dietary restrictions are often not an effective means of preventing the long-term clinical complications associated with GSDIa, including proteinuria (that may progress to renal failure), growth retardation, osteopenia, and the formation of hepatocellular adenomas (HCAs) and hepatocellular carcinomas (HCCs) (Chen et al., 1988; Wolfsdorf and Crigler, 1997; Wolfsdorf et al., 1997; Wolfsdorf et al., 1999).

Complications, such as hepatomegaly (resulting from the pathological accumulation of glycogen in the liver), an increase in the number and size of HCAs, and the development of HCCs become more common as individuals with GSDIa age (Franco et al., 2005). For example, HCAs are present in 70% to 80% of GSDIa patients over the age of 25 years, with up to 10% of HCAs developing into life-threatening HCCs (Franco et al., 2005; Reddy et al., 2007; Lee et al., 2012). Orthotopic liver transplantation has become a treatment option for patients and has been shown to correct hypoglycemia and other biochemical abnormalities associated with GSDIa, as well as result in an increase in body height (Matern et al., 1999; Boers et al., 2014). These results provide evidence for the efficacy and benefit of restoring *G6PC* expression in the liver of patients with GSDIa. However, given that liver transplantation is limited by donor availability and is associated with significant risk of morbidity and mortality including a very high rate of renal failure (Davis and Weinstein, 2008; Boers et al., 2014), there is a significant unmet medical need for a treatment that allows for sustained euglycemia and prevention of the long-term complications and life-threatening hypoglycemia that are associated with GSDIa. In nonclinical studies, adeno-associated virus (AAV) serotype 8 (AAV8)-mediated delivery of *G6PC* in mice and dogs with GSDIa has restored G6Pase activity and ameliorated disease sequelae (Koeberl et al., 2009; Koeberl et al., 2008; Weinstein et al., 2010; Chou and Mansfield, 2011), providing evidence that gene transfer could have significant benefit in patients with GSDIa.

5.2. Overview of DTX401 Development

A brief overview of existing information on DTX401 is provided below; a comprehensive review of available data is contained in the Investigator's Brochure (IB) provided by Ultragenyx, which should be reviewed prior to initiating the study.

5.2.1. Brief Description of DTX401

DTX401 is a nonreplicating, recombinant AAV8 vector that contains a codon-optimized, wild-type human *G6PC* coding sequence. DTX401 demonstrates thermal stability, which is a general property of AAV and parvoviruses. DTX401 is supplied as a slightly hyperosmotic buffered formulation solution of approximately 400 milliosmole at pH 8.0. DTX401 is a homogeneous, monodisperse solution that is clear and colorless without visible particulates.

5.2.2. Nonclinical Studies

Ultragenyx has conducted a comprehensive nonclinical program to support the single intravenous (IV) administration of DTX401. Studies of potential clinical significance and relevance to this protocol are summarized below.

The safety and tolerability of DTX401 were evaluated in a non-Good Laboratory Practice (GLP) nonclinical study. Doses up to 1.0×10^{13} genome copies (GC)/kg were evaluated in the neonatal glucose-6-phosphatase knockout (*G6pc*$^{-/-}$) mouse model. Results demonstrated the ability of the candidate transgene constructs to prolong survival, improve biochemical markers of GSDIa disease, and drive persistent expression of G6Pase in the liver. The potential toxicity and biodistribution of DTX401 were evaluated in a GLP nonclinical study. Doses up to 6.5×10^{13} GC/kg were evaluated in C57BL/6 mice. In this study, adverse histopathological findings were observed in the spleen and mandibular and mesenteric lymph nodes (lymphoid hyperplasia) and liver (single cell necrosis/apoptosis, Kupffer cell hypertrophy/hyperplasia), at Day 29 at the highest dose (6.5×10^{13} GC/kg); accompanied by increases in clinical chemistry (including alanine aminotransferase [ALT] and aspartate aminotransferase [AST]) and hematology parameters. These findings are potentially due to an immune response to the administration of a high dose of a foreign protein, as a persistent interferon-γ T-cell response was observed to the AAV8 capsid. The adverse findings were at the high dose only, and completely resolved by Day 92 with no additional intervention. Based on these findings, the no observed adverse effect level of DTX401 in this study was determined to be 1×10^{13} GC/kg.

Dorsal root ganglion (DRG) and peripheral nerve pathologies have been reported in nonhuman primates, piglets, and mice administered rAAV gene therapy products (Bolt et al., 2021; Van Alstyne et al., 2021; Hordeaux et al., 2020a; Hordeaux et al., 2020b; Hinderer et al., 2018; Schuster et al., 2014). The clinical relevance and translation of these nonclinical DRG and nerve pathologies is not known. The main parameters demonstrated to affect severity of the DRG findings was route of administration, where IV dosing was shown to be more resistant to DRG lesions compared to CNS routes of delivery, and vector dose ($> 1 \times 10^{13}$ GC/kg). DTX401 is designed to be hepatotropic and show little transgene expression in tissues outside the liver.

The totality of the available literature information and DTX401 nonclinical data, combined with the used clinical dose (6.0×10^{12} GC/kg), use of a liver-specific promoter, and IV route of administration, suggests that the risk of DRG and peripheral nerve-related toxicities occurring with DTX401 treatment is low. Despite the low risk with DTX401, DRG pathology is currently being assessed as part of the 92-day GLP toxicology study in mice as a follow up to these recent publications.

5.2.3. Previous and Ongoing Clinical Studies

Study 401GSDIA01 was completed in November 2022 and two clinical studies with DTX401 (Studies 401GSDIA02 and DTX401-CL301) are ongoing.

5.3. Summary of Overall Risks and Potential Benefits

Adeno-associated virus vectors represent a class of viral vectors that are nonreplicating, nonintegrating, and present a low risk for gene therapy-related delayed adverse reactions.

Recommended long term safety monitoring for subjects enrolled in AAV gene therapy clinical trials is a minimum of 5 years (CBER, 2006; CHMP, 2009).

Subjects enrolled into Study 401GSDIA02 will be followed for long-term safety and efficacy of the single IV infusion of DTX401 administered in Study 401GSDIA01. Subjects will not receive any additional doses of DTX401. Despite the low risk of DRG and peripheral nerve-related toxicities as mentioned in Section 5.2.2, subjects will be regularly assessed for clinically meaningful signs of sensory neuropathy throughout the 401GSDIA02 study, and if this event occurs, they will be further evaluated and closely monitored. The overall risk and potential benefits for subjects enrolled into Study 401GSDIA02 are considered acceptable for a follow-up study.

5.4. Study Rationale

Study 401GSDIA02 is a long-term follow-up study to evaluate the safety and efficacy of AAV8-mediated gene transfer of G6Pase in adults with GSDIa. Only subjects who received DTX401 in Study 401GSDIA01 are eligible to participate in Study 401GSDIA02. Study 401GSDIA01 was a Phase 1/2, open-label safety and dose-finding study of AAV8-mediated gene transfer of G6Pase in adults with GSDIa, during which subjects received a single IV dose of DTX401 with steroids (prednisone) to manage alanine aminotransferase (ALT) elevation. Overall, 12 patients were enrolled in following cohorts of 3 subjects each:

- Cohort 1: Dose 1 (2.0×10^{12} genome copies [GC]/kg) with a reactive steroid regimen (prednisone starting dose of 40 mg/day)
- Cohort 2: Dose 2 (6.0×10^{12} GC/kg) with a reactive steroid regimen (prednisone starting dose of 40 mg/day)
- Cohort 3: Dose 2 (6.0×10^{12} GC/kg) with an optimized reactive steroid regimen (prednisone starting dose of 60 mg/day)
- Cohort 4: Dose 2 (6.0×10^{12} GC/kg) with a prophylactic steroid regimen

No investigational product (IP) will be administered during Study 401GSDIA02.

6. STUDY OBJECTIVES

Primary Objective
The primary objective of this study is as follows:

- To determine the long-term safety of DTX401 following a single IV dose in adults with GSDIa

Secondary Objective
The secondary objective of this study is as follows:

- To evaluate the long-term effect of DTX401 on symptom-free euglycemia in a setting of a controlled fasting challenge

Tertiary Objectives
The tertiary objectives of this study are as follows:

- To evaluate the long-term effect of DTX401 on lipid profiles (total cholesterol, low-density lipoprotein, and triglycerides)
- To evaluate the long-term effect of DTX401 on uric acid
- To evaluate the long-term effect of DTX401 on proteinuria
- To evaluate the long-term effect of DTX401 on liver size and fat content
- To evaluate the long-term effect of DTX401 on hepatic adenoma progression
- To assess the long-term impact of DTX401 on cornstarch requirements
- To assess the long-term impact of DTX401 on glucose homeostasis
- To assess the long-term impact of DTX401 on the subject's health-related quality of life (HRQoL) and sleep quality
- To describe the long-term immune response to AAV8 capsid proteins after IV administration of DTX401
- To describe the long-term immune response to G6Pase after IV administration of DTX401
- To evaluate the long-term effect of DTX401 on lactate levels
- To evaluate the long-term effects of DTX401 on the endocrine and metabolic responses to fasting

7. INVESTIGATIONAL PLAN

7.1. Overall Study Design and Plan

Study 401GSDIA02 is a long-term follow-up study to evaluate the safety and efficacy of AAV8-mediated gene transfer of G6Pase in adults with GSDIa. In Study 401GSDIA01, subjects were followed for 1 year after dosing with DTX401. In Study 401GSDIA02, subjects will be followed for at least 3 additional years (and up to 5 additional years) for a total of 4-6 years of follow-up after administration of DTX401. Subjects will visit the study site approximately every 13 weeks during the first year of Study 401GSDIA02 and then approximately every 26 weeks through the end of the study at Week 312 (Year 6) for safety and efficacy evaluations. A long-term disease monitoring program (DMP) DTX401-CL401 is planned. Once the DMP is available to enroll, and after subjects reach the Week 208 visit in Study 401GSDIA02, subjects will transition to the DMP with Sponsor approval.

Safety will be assessed based on adverse events (AEs), serious adverse events (SAEs), vital sign measurements, body weight, physical examination findings, ECG findings, documented symptomatic hypoglycemic events, clinical laboratory assessments (clinical chemistry, hematology, coagulation panel, and urinalysis), measurement of neutralizing antibody titer to AAV8, measurement of AAV8 binding antibody immunoglobulin G (IgG), measurement of cell-mediated immune response to AAV8 and G6Pase, measurement of anti-G6Pase antibodies, and concomitant medication.

Efficacy will be assessed for each cohort and overall based on the change from Day 0 (Study 401GSDIA01) measurements in time to first hypoglycemic event during a controlled fasting challenge over time following administration of DTX401. Other efficacy assessments include average daily cornstarch use, glucose levels, liver imaging results, and HRQoL and sleep assessments. No control group will be enrolled in Study 401GSDIA02.

All study visits and the timing of assessments are detailed in Table 1. A schematic of the study design is provided in Figure 2.

Study visits will be inpatient visits to accommodate the controlled fasting challenge, or outpatient visits when the controlled fasting challenge is not performed per protocol.

At Weeks 65, 91, 130, 182, and 234, every effort will be made to conduct an on-site assessment. However, under extenuating subject circumstances that, in the Investigator’s opinion, make an on-site visit not feasible, and after all reasonable measures to enable the subject's onsite visit have been exhausted, a home healthcare visit and/or telemedicine assessment may be conducted at these time points.

Additional unscheduled visits, either on site or home healthcare/telemedicine visits, may be performed to monitor subject safety, especially after any changes to the subject’s diet and cornstarch regimen.

Figure 2: Study Design

Visit 1 (Week 52)
• Visit 1 (Week 52) assessments may coincide with Week 52/EOS/EW visit in Study 401GSDIA01. • End of study or EW assessments for Study 401GSDIA01 do not need to be repeated for this study if written informed consent for Study 401GSDIA02 is obtained before the Week 52/EOS or EW assessments are performed for Study 401GSDIA01. • Assessments may be performed on more than 1 day provided that all assessments are completed and results are available within the 7 day window.
Follow-up: Year 2 (~Every 13 weeks: Weeks 65, 78, 91, 104)
• Concomitant medications, AEs, SAEs, targeted physical examination, vital sign measurements, body weight, clinical laboratory assessments • Measurement of neutralizing and IgG antibodies as well as cell-mediated immune response to AAV8 and anti-G6Pase antibodies (Week 104) • Perform liver ultrasound (Week 78 and 104) • Assess symptomatic hypoglycemic events • Assess prescribed diet/dietary intake and cornstarch (or equivalent) use • Assessment of glucose levels • Complete controlled fasting challenge and 24-hour urine collection (both at Weeks 78 and 104) • Administer HRQoL and sleep quality assessments • Endpoint Outcomes telephone interview (Week 104 only)
Follow-up: Years 3-6 (~Every 26 weeks: Weeks 130, 156, 182, 208, 234, 260, 286, EOS [312])
• Concomitant medications, AEs, SAEs, targeted and complete (Week 312 or EW) physical examination, vital sign measurements, body weight, clinical laboratory assessments • Measurement of neutralizing antibodies and IgG antibodies as well as cell-mediated immune response to AAV8 and anti-G6Pase antibodies (Weeks 156, 208, 260) • Perform liver ultrasound (Weeks 156, 208, 260, 312) • Assess symptomatic hypoglycemic events • Assess prescribed diet/dietary intake and cornstarch (or equivalent) use • Assessment of glucose levels • Complete controlled fasting challenge and 24-hour urine collection (both at visits at Weeks 156, 208, 260) • Perform liver MRI (Week 260, after the controlled fasting challenge is complete) • Administer HRQoL and sleep quality assessments
Early Withdrawal
• Concomitant medications, AEs, SAEs, complete physical examination, vital sign measurements, body weight, clinical laboratory assessments • Measurement of neutralizing and IgG antibodies as well as cell-mediated immune response to AAV8 and anti-G6Pase antibodies • Assess symptomatic hypoglycemic events • Assess prescribed diet/dietary intake and cornstarch (or equivalent) use • Assessment of glucose levels • Complete controlled fasting challenge and 24-hour urine collection

- Perform liver MRI (after the controlled fasting challenge is complete)
- Administer HRQoL and sleep quality assessments
- Endpoint Outcomes telephone interview (only if subject withdraws prior to Week 104)

Abbreviations: AAV8 = adeno-associated virus serotype 8; AE = adverse event; EOS = end of study; EW = early withdrawal; G6Pase = glucose-6-phosphatase; HRQoL = health-related quality of life; IgG = immunoglobulin G; MRI = magnetic resonance imaging; SAE = serious adverse event.

Note: Specified visits that include a controlled fasting challenge will require an approximate 24-hour inpatient stay (Visit 1, Weeks 78, 104, 156, 208, 260, and EW visit, as applicable). All other visits (Weeks 65, 91, 130, 182, 234, 286, and 312) will be conducted on an outpatient basis (approximately 1.5 to 2 hours).

7.1.1. Number of Study Sites

This study will include up to 12 global study sites.

7.1.2. Number of Subjects

This study will include approximately 12 subjects.

7.1.3. Estimated Study Duration

7.1.3.1. Study Duration for Subjects

The duration of the study for each subject is defined as the date the subject provides written informed consent for Study 401GSDIA02 through the Week 312 (Year 6) visit. Subjects will be followed in Study 401GSDIA02 for at least 3 additional years (and up to 5 additional years) for a total of 4-6 years after administration of DTX401 in Study 401GSDIA01. Once the DMP is available to enroll, and after subjects reach the Week 208 visit in Study 401GSDIA02, subjects will transition to the DMP with Sponsor approval.

Specified visits (Visit 1, Weeks 78, 104, 156, 208, 260, and EW visit, as applicable) will include a controlled fasting challenge and will require an approximate 24-hour inpatient stay. All other visits (Weeks 65, 91, 130, 182, 234, 286, and 312) will be conducted on an outpatient basis (approximately 1.5 to 2 hours).

7.1.3.2. Selection of Study Population

Study 401GSDIA02 is an extension study of 401GSDIA01 and all subjects who received DTX401 in that study were eligible to participate in 401GSDIA02.

7.1.3.3. Start of Study Definition

The start of the study is defined as the first signed informed consent.

7.1.3.4. Eligibility Determination Period (Visit 1 [Week 52])

After each subject has provided written informed consent, the Investigator or other qualified study site personnel will determine whether the subject is eligible for the study. This will be accomplished by reviewing the inclusion and exclusion criteria and completing all of the Visit 1 (Week 52/EOS) assessments outlined in the Schedule of Events (Table 1). Visit 1 (of Study 401GSDIA02) assessments may be performed on more than 1 day, provided that all of the assessments are completed and the results are available within the permitted 7-day window. NOTE: Assessments required for Study 401GSDIA02 that are the same as those performed

during the Week 52 or EW visit in Study 401GSDIA01 do not need to be repeated if written informed consent for Study 401GSDIA02 is obtained before the Week 52 or EW assessments are performed for Study 401GSDIA01.

In the event a subject withdraws from Study 401GSDIA01 before the Week 52 visit and then decides to enroll into Study 401GSDIA02, the Schedule of Events for Study 401GSDIA02 will be adjusted to allow for a total of 5 years of safety and efficacy follow-up after DTX401 administration.

7.1.3.5. Study Completion

The end of the study will occur when the last subject completes the Week 312 follow-up visit (or or Week 208 if the last subject enrolls into the DMP).

7.1.3.6. Continued Access to Study Intervention after the End of Study

DTX401 was only administered in Study 401GSDIA01. Additional safety follow up will occur in the DTX401 DMP.

7.1.4. Subject Input into Design

Assessments evaluated in Study 401GSDIA02 have been streamlined and modified by protocol amendment following feedback from patients and families collected by sites.

7.2. Discussion of Study Design, Including Choice of Control Group

This long-term follow-up study will evaluate the safety and efficacy of DTX401 administered during Study 401GSDIA01. Subjects will visit the study site approximately every 13 weeks during the first year of Study 401GSDIA02 and then approximately every 26 weeks through the end of the study at Week 312 (Year 6) (or Week 208 if the last subject enrolls into the DMP) for safety and efficacy evaluations.

No control group will be enrolled in Study 401GSDIA02.

7.3. Changes to the Protocol due to COVID-19

Changes to the protocol or Investigational Plan to minimize or eliminate immediate hazards or to protect the life and well-being of research participants and/or study staff may be implemented without IRB/EC approval or before filing an amendment to the competent authority but are required to be reported afterward. Investigators should contact Ultragenyx or designee to determine an appropriate course of action, which may include but is not limited to remote visits, home health visits, and out of window visits or assessments. Any changes to the protocol or investigational plan must be documented. The Investigator will work with Ultragenyx or Designee and IRB/EC to prioritize reporting of protocol deviations that impact safety of trial participants.

In response to COVID-19 travel restrictions, patients may be monitored remotely in place of a scheduled in-clinic visit or an unscheduled safety visit between study visit time points. To limit unnecessary and potentially unsafe travel to the clinical site, Ultragenyx will provide the means for the Investigator to arrange home visits for collection of lab samples as necessary. A qualified healthcare professional from Firma Clinical Research, trained in the study and its procedures,

will be dispatched to collect samples on behalf of the Investigator. These home health visits will be documented as an unscheduled safety visit in the subject's source and recorded as such in the electronic case report form (eCRF). The Investigator may arrange home visits for collection of lab samples as clinically indicated or in response to COVID-19 related travel restrictions. Any missed scheduled clinic visits or assessments caused by COVID-19 travel restrictions will be entered as COVID-19 Protocol Deviations.

8. STUDY POPULATION

All subjects enrolled in Study 401GSDIA02 will have received a single IV dose of DTX401 during their participation in Study 401GSDIA01.

Information about the study population size is provided in Section 7.1.2.

8.1. Inclusion Criteria

Individuals eligible to participate in this study must meet all of the following criteria:

1. Received DTX401 in Study 401GSDIA01
2. Willing and able to provide written informed consent after the nature of the study has been explained, and prior to any research-related procedures being performed
3. Willing and able to comply with all scheduled study visits, procedures, and requirements

8.2. Exclusion Criteria

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Planned or current participation in any other interventional clinical study that may confound the safety or efficacy evaluation of DTX401 during this study
2. Presence or history of any condition that, in the view of the Investigator, poses a risk to subject safety or places the subject at high risk of poor compliance or not completing the study or that would significantly affect the interpretation of study results

8.3. Removal of Subjects from Therapy or Assessment

In accordance with the Declaration of Helsinki, subjects have the right to withdraw from the study at any time for any reason. The Investigator and Ultragenyx also have the right to remove subjects from the study. Ultragenyx must be notified of all subject withdrawals as soon as possible.

Subjects may be removed from the study for the following reasons:

- A condition or illness that, in the judgment of the Investigator, might place the subject at risk or interfere with study participation
- Subject withdrawal of consent
- Lost to follow up
- Death

Any subject who experiences an unexpected and DTX401-related SAE (Sections 11.1.1 and 11.1.3) that represents a change in the severity or an increase in frequency of the event from their prior medical history or known GSDIa-related medical issues will be evaluated as to whether the subject will continue on the study.

If the reason for removal of a subject from the study is an AE, the AE and any related test or procedure results will be recorded in the source documents and transcribed onto the eCRF. Each

related clinically significant abnormal laboratory value or other clinically meaningful abnormality should be followed until the abnormality resolves or until a decision is made that it is not likely to resolve. All unscheduled tests must be reported to Ultragenyx immediately.

If a subject discontinues from the study prematurely, every reasonable effort should be made to perform the EW Visit procedures within 1 week of discontinuation.

9. TREATMENTS

9.1. Investigational Product

No IP will be administered during this study.

9.2. Reference Therapy

Not applicable.

9.3. Dosage Administration

Not applicable.

9.4. Dosage Rationale

Not applicable.

9.5. Concomitant Medications, Therapies, and Procedures

Subjects may receive concomitant medications, therapies, and procedures as required, with the exception of those identified in Section 9.6 as prohibited. Medications (investigational, prescription, over-the-counter, and herbal) and nutritional supplements taken during the 30 days prior to enrollment or Visit 1 will be reviewed and recorded at enrollment or Visit 1. At Visit 1 in Study 401GSDIA02, current medications will be recorded. At each visit, any concomitant medications added or discontinued during the study should be recorded on the eCRF.

The site personnel should record the following in the eCRF: date the medication was taken, the name of the medication, medication dosage, and the reason the medication was taken. This includes all prescription drugs, herbal products, vitamins, minerals, and over the counter medications. Any changes in concomitant medication use will also be recorded in the subject's eCRF. The minimum requirement for therapies and procedures is to record the name and date of the therapy or procedure and the reason it was performed.

9.6. Restricted or Prohibited Medications, Devices, and Procedures

The following are specifically prohibited during study participation:

- Any IP
- Any gene therapy product

9.7. Treatment Compliance

Not applicable.

9.8. Treatment Group Allocation

9.8.1. Method of Assigning Subjects to Treatment Groups

As no IP is administered in this study, there will be no assignment to treatment groups. Subject numbers will remain the same as those assigned in Study 401GSDIA01.

9.8.2. Blinding

Not applicable.

10. STUDY PROCEDURES

10.1. Study Schedule

Parameters to be assessed and the timing of assessments are provided in Table 1.

10.2. Informed Consent and Eligibility Requirements

Written informed consent for participation in the study must be obtained before performing any study specific tests or evaluations.

Informed consent forms (ICFs) for enrolled subjects and for subjects who are not subsequently enrolled will be maintained at the study site.

The Investigator will maintain a screening log to record details of all subjects screened and to confirm eligibility or record reasons for screening failure, as applicable.

10.3. Safety Assessments

Safety will be evaluated by the incidence, frequency, and severity of AEs and SAEs, and by vital sign measurements, body weight, complete and targeted physical examination findings, electrocardiogram (ECG) results, documented hypoglycemia events, clinical laboratory evaluations (clinical chemistry including liver function tests [LFTs], hematology, coagulation panel, and urinalysis), measurement of neutralizing antibodies to AAV8, measurement of AAV8 binding antibody IgG, measurement of anti-G6Pase antibodies, measurement of cell-mediated immune response to AAV8 and G6Pase, and concomitant medications.

10.3.1. Adverse Events

All AEs will be recorded from the time the subject signs the ICF and up to the end of study (EOS; Week 312 or Week 208 if the last subject enrolls into the DMP) or Early Withdrawal visit. Serious AEs will be recorded for up to 30 days after the EOS or Early Withdrawal visit. The determination, evaluation, reporting, and follow up of AEs will be performed as outlined in Section 11. At each visit, subjects will be asked about any new or ongoing AEs since the previous visit. Assessments of AEs will occur at each study visit.

Clinically significant changes from Visit 1 will be recorded as AEs or SAEs, if appropriate.

10.3.2. Vital Sign Measurements

Vital signs to be measured will include heart rate in beats per minute, seated systolic blood pressure and diastolic blood pressure measured in millimeters of mercury (mm Hg), and respiration rate in breaths per minute. Vital signs measurements will be performed at every visit before any additional assessments are completed.

Vital sign measurements will be recorded on the appropriate page of the eCRF. The medical monitor should be notified of any clinically significant changes or abnormal value in vital sign measurements (Section 11.1). If, in the medical and scientific judgment of the Investigator, a clinically significant change or abnormal vital sign measurement is observed, it should be recorded as an AE or SAE, as defined in Section 11.1, on the appropriate pages of the eCRF.

10.3.3. Body Weight

Body weight will be assessed at the time points specified in the Schedule of Events (Table 1).

10.3.4. Physical Examinations

A complete or targeted physical examination will be performed at the time points specified in the Schedule of Events (Table 1).

A complete physical examination will include assessments of the head, eyes, ears, nose, and throat; skin; abdomen with documentation of liver and spleen size; and the endocrine, metabolic, neurological, respiratory, cardiovascular, gastrointestinal, and musculoskeletal systems.

A targeted physical examination will include assessment of the skin, the abdomen with documentation of liver and spleen size, and the respiratory, cardiovascular, gastrointestinal, and peripheral and central nervous systems.

Physical examination findings will be captured on the appropriate page of the eCRF.

10.3.5. Electrocardiograms

An ECG will be performed at Visit 1 (Table 1). A single 12-lead ECG will be obtained using an ECG machine that automatically calculates the heart rate and measures PR, QRS, QT, and corrected QT intervals.

Twelve-lead ECGs should be measured with the subject having rested for at least 5 minutes beforehand. It is preferred that the measurement be late with the subject rested, rather than on time with the subject not sufficiently rested. If the subject is not sufficiently rested, this needs to be stated in the source documents. Electrocardiogram results will be recorded on the appropriate page of the eCRF.

10.3.6. Hypoglycemic Events

At each visit, the study site will instruct subjects on proper glucose monitoring at home, the signs and symptoms of hypoglycemia, and supplemental glucose treatment if needed. During each visit, the study site will record the number of symptomatic hypoglycemia events in the eCRF.

10.3.7. Clinical Laboratory Tests

The clinical laboratory evaluations to be performed in this study include clinical chemistry (including LFTs), hematology, coagulation panel, and urinalysis. Clinical laboratory testing will be performed at the time points specified in the Schedule of Events (Table 1). Fasting is not required.

Laboratory tests, including ALT, will be closely monitored throughout the duration of the study. Investigators will receive flagged notification of any laboratory values that are outside of the normal range. Any abnormal laboratory test results, including those that worsen from Visit 1 or are considered to be clinically significant in the medical and scientific judgment of the Investigator, are to be recorded as AEs or SAEs (Section 11.1).

Blood, saliva, urine, and stool samples for vector shedding will only be collected for subjects who did not demonstrate clearance in Study 401GSDIA01. Samples for vector shedding analysis will be collected until at least 3 consecutive negative results are obtained for each sample matrix.

However, any clinically significant safety assessments that are associated with GSDIa are **not** to be reported as AEs, **unless** they are judged by the Investigator to be more severe than expected for the subject's condition.

All laboratory tests with results that are significantly abnormal during participation in the study should be repeated until the values return to normal or Visit 1 values. If such values do not return to normal within a period judged reasonable by the Investigator, the etiology should be identified and the Sponsor notified.

The clinical laboratory evaluations to be performed in this study are listed in Table 2. In addition to these clinical laboratory evaluations, glucose and lactate venous blood samples will be obtained as described in Sections 10.4.1 and 10.3.7.1, respectively.

Table 2: Clinical Laboratory Parameters

Clinical chemistry:	Lipid panel, uric acid, sodium, potassium, chloride, carbon dioxide, blood urea nitrogen, creatinine, glucose, calcium, phosphate, magnesium, albumin, total protein, creatine kinase, bilirubin (total, direct, and indirect), ALT, AST, ALP, gammaglutamyl transferase, and lactate dehydrogenase
Clinical chemistry (other):[1]	Glucose and lactate
Hematology:	Complete blood count with differential
Urinalysis:	Specific gravity, pH, glucose, protein, blood (by dipstick), ketones (by dipstick), and microscopic examination (if blood or protein is found)
Coagulation panel:	PT/INR and aPTT
Other:	Cortisol, ACTH, free fatty acids, glucagon, insulin, C-peptide, growth hormone, IGFBP1, alanine and ketone levels (3-hydroxy butyrate), AAV8 neutralizing antibody, AAV8 binding antibody IgG, , cell-mediated immune response to AAV8 and G6Pase, anti-G6Pase antibody, vector genome, vector shedding[2]

Abbreviations: AAV = adeno-associated virus; ACTH = adrenocorticotropic hormone; ALP = alkaline phosphatase; ALT = alanine aminotransferase; aPTT = activated partial thromboplastin time; AST = aspartate aminotransferase; G6Pase, glucose6phosphatase; IGFBP1 = insulin-like growth factor-binding protein 1; PT/INR = prothrombin time/international normalized ratio.

[1] Samples collected during the controlled fasting challenge should be sent to the local laboratory (STAT samples) for analysis.

[2] Samples for vector shedding will only be collected for subjects who do not show clearance in Study 401GSDIA01

Details for the preparation and shipment of samples will be provided by the central or specialty laboratory. Reference ranges for all parameters will be provided to the study site by the central or specialty laboratory.

Subjects who experience an SAE or other AE of concern may, at the discretion of the Investigator (and/or medical monitor), have additional blood samples taken for safety laboratory tests.

10.3.7.1. Lactate

Lactate levels will be measured during the controlled fasting challenge (Section 10.4.1) at the time points specified in the Schedule of Events (Table 1). A blood sample for venous lactate will be collected through an indwelling catheter (central or peripheral) at the beginning of the controlled fasting challenge and approximately every 60 minutes (±5 minutes) until glucose

levels are at or below 70 mg/dL (≤3.9 mmol/L). Once glucose levels are at or below 70 mg/dL (≤3.9 mmol/L), a blood sample for lactate will be collected approximately every 30 minutes (±5 minutes) until the subject's glucose level drops below 54 mg/dL (<3.0 mmol/L), the subject experiences symptoms of hypoglycemia per the Investigator's discretion, or the fast reaches 15 hours without hypoglycemia, whichever occurs first.

Lactate samples should be sent to the local laboratory (STAT sample) and results should be available within 30 minutes or less of the blood collection. Samples should be prepared and transported to the local laboratory in accordance with the site's standard procedures.

10.3.7.2. Other Controlled Fasting Challenge Laboratory Assessments

Blood samples for measurement of cortisol, ACTH, free fatty acids, glucagon, insulin, C-peptide, growth hormone, IGFBP1, alanine and ketone levels (3-hydroxy butyrate) will be collected at the beginning and end of the controlled fasting challenge, or more frequently at the Investigator's discretion, if possible (Section 10.4.1). Details for the preparation and shipment of samples are included in the laboratory manual.

10.3.7.3. 24-Hour Urine Collection

The excretion of total protein, microalbumin, and creatinine in the urine will be determined over a 24-hour period. Timed 24-hour complete urine samples will be collected at the time points specified in the Schedule of Events (Table 1).

10.3.8. Adeno-Associated Virus Serotype 8 Binding Antibody Immunoglobulin G Assay

Blood samples for the AAV8 binding antibody IgG assay will be collected at the time points specified in the Schedule of Events (Table 1) to monitor for circulating anti-AAV8 antibodies. The assay will be performed using a research method (enzyme-linked immunosorbent assay [ELISA]).

10.3.9. Neutralizing Antibodies to AAV8

Blood samples for determination of neutralizing antibodies to AAV8 will be collected at the time points specified in the Schedule of Events (Table 1) to monitor for a humoral immune response to AAV8. The assay will be performed using a research method (cell-based assay).

10.3.9.1. Cell-Mediated Immune Response

The presence of T cells specific for AAV8 and G6Pase will be determined by an enzyme-linked immunospot (ELISPOT) assay. A blood sample will be collected at the time points specified in the Schedule of Events (Table 1).

10.3.10. Anti-Glucose-6-Phosphatase Antibody Assay

Blood samples for the anti-G6Pase antibody assay will be collected at the time points specified in the Schedule of Events (Table 1) to monitor for circulating anti-G6Pase antibodies. The assay will be performed using a research method.

10.3.11. Vector Shedding

In the event that clearance was not observed for vector shedding for a subject in Study 401GSDIA01, then blood, saliva, urine, and stool samples will be collected at the time points specified in the Schedule of Events (Table 1) to monitor for vector shedding. The presence of DTX401 will be determined by quantitative polymerase chain reaction (qPCR). Samples for vector shedding analysis will be collected until at least 3 consecutive negative results are obtained for each sample matrix.

10.3.12. Pregnancy Testing and Contraception Requirements

Pregnancy testing and contraception use are not required for this study.

Refer to Section 11.1.4.3 for pregnancy reporting requirements.

10.4. Disease-Specific Clinical Assessments

10.4.1. Symptom-Free Euglycemia (Controlled Fasting Challenge)

The subject's ability to maintain symptom-free euglycemia during a controlled fasting challenge will be assessed at specified time points (Table 1). However, if clinically indicated, the controlled fasting challenge can be performed at an unscheduled visit.

The controlled fasting challenge requires a 24-hour inpatient stay in a hospital or research facility and will be performed after all other study visit assessments (except MRI, if applicable) have been completed and the required laboratory assessments (as specified below) have been reviewed by the Investigator (or a subinvestigator assigned to review results and make clinical decisions per the site delegation log). Subjects should wear their continuous glucose monitoring (CGM) device throughout the fasting challenge, and should not change the sensor prior to the assessment.

Before the controlled fasting challenge begins, the study site will provide dinner for the subject. The dinner meal will be personalized for each subject, and will include a target carbohydrate range and overall composition in protein, fats and dietary fibers that is as close as possible to the subject’s most current dinner prescription, but not higher in carbohydrate content than the dinner prescription recorded at their baseline visit in Study 401GSDIA01. The start and stop time of the dinner meal and its full dietary composition including amount of carbohydrates consumed will be recorded. After dinner, subjects will be given an oral dose of cornstarch equivalent per their most recent cornstarch prescription, at approximately the same time that they typically take it, but no later than 3 hours post dinner. The amount of cornstarch administered should not be higher than the prescribed amount recorded at their baseline visit in Study 401GSDIA01. The time of ingestion of prefasting cornstarch and the amount consumed will be recorded. After the cornstarch is ingested, the controlled fasting challenge will begin, and the start time will be recorded accordingly. If a subject does not receive cornstarch before going to bed at the time of the CFC, the fasting challenge will start after they complete their dinner. Subjects will be instructed to minimize activity after they finish eating dinner until the end of the controlled fasting challenge.

During the fast, subjects may not ingest any food or drink other than water. The controlled fasting challenge will end when the subject’s glucose level decreases to < 54 mg/dL

(< 3.0 mmol/L) or when the subjects experience signs and symptoms of hypoglycemia, or when the fast reaches 15 hours without hypoglycemia, whichever occurs first. The stop time of the controlled fasting challenge will be recorded. At the end of the controlled fasting challenge, the subject's hypoglycemia should be treated per the institution's standard procedure, which may include providing the subject with a meal, cornstarch, or IV glucose, if medically indicated.

In cases where the controlled fasting challenge is terminated prematurely before the subject reaches a glucose level of < 54mg/dL or without the subject experiencing signs and symptoms of hypoglycemia, or due to any deviation in the fasting challenge protocol, an ad hoc fasting challenge may be scheduled within 6 weeks from the scheduled fasting challenge.

Laboratory sample collection before and during the controlled fasting challenge

On the morning of inpatient visit for the controlled fasting challenge, after vital signs and other non-interventional assessments have been performed, samples will be collected for standard clinical chemistry (including lipid levels), hematology, coagulation panel, and urinalysis. These samples should be collected at least 2 to 4 hours after the subject's last meal and sent to the central lab. A sample for STAT AST and ALT level analysis will also be collected at this time and sent to the local laboratory; the Investigator or delegated subinvestigator should review the results before starting the controlled fasting challenge.

Total urine voided over the 24-hour period starting at the time of inpatient admission will be collected during the controlled fasting challenge.

Blood samples for measurement of cortisol, ACTH, free fatty acids, glucagon, insulin, C-peptide, growth hormone, IGFBP1, alanine and ketone levels (3-hydroxy butyrate) will be collected at the beginning (ie, immediately after the post-dinner dose of cornstarch, or immediately after dinner for subjects who do not take a post-dinner dose of cornstarch) and end of the controlled fasting challenge, or more frequently at the Investigator's discretion. A final sample for glucose, lactate, growth hormone, IGFBP1, ACTH and cortisol measurement will be collected 30 minutes after the end of the controlled fasting challenge. Samples should be prepared and transported to the local laboratory in accordance with the site's standard procedures.

Blood samples for STAT analysis of glucose and lactate will be collected through an indwelling catheter (central or peripheral) at the following time points:

- At the beginning of the fast (ie, immediately after the post-dinner dose of cornstarch, or immediately after dinner for subjects who do not take a post-dinner dose of cornstarch)
- Approximately every 60 minutes (± 5 minutes) until the glucose level decreases to ≤ 70 mg/dL (≤ 3.9 mmol/L)
- Approximately every 30 minutes (± 5 minutes) until the glucose level decreases to < 54 mg/dL (< 3.0 mmol/L) or when the subjects experience signs and symptoms of hypoglycemia, or when the fast reaches 15 hours without hypoglycemia, whichever occurs first

Samples should be prepared and transported to the local laboratory in accordance with the site's standard procedures. To ensure the safety of the study subjects, results from these tests should be available within 30 minutes after the blood sample collection. Capillary glucose will be performed at the same time points as blood samples for STAT analysis of glucose and lactate.

Results of the capillary glucose and readings from CGM at corresponding time points will be noted on a controlled fasting challenge results sheet.

10.4.2. Continuous Glucose Monitoring

Supplemental information on glucose level trends will be collected using a CGM device. If subjects do not already have a study-issued CGM device (as these are provided in Study 401GSDIA01), a study-issued CGM will be provided. Subjects should bring their CGM device to each visit, containing CGM data from the 28 days prior to the visit (Table 1). If a study visit is conducted remotely (home healthcare visit or telemedicine), or if a study visit is skipped entirely for any reason, arrangements will be made for the subject to ship the CGM receiver (ideally, containing CGM data from the 28 days prior to the remote/missed visit) to the clinical site.

Data from the subject's CGM device will be transferred to the Sponsor throughout the study, including a final data transfer at the end of the Week 312 visit or early withdrawal. It is recommended that all CGM sensors should be paired with the study-issued receiver, regardless of whether the subject is also paring the sensor with their own personal device (i.e. smartphone). The receivers should be maintained in bluetooth range of the sensors, whenever possible, and the data from the receivers should be uploaded on the study-issued laptop on a weekly basis. Additional details regarding the care and use of the CGM device and sensors and data transfer to the Sponsor are provided in the Manual of Operations.

10.4.3. Morning Glucose Assessments

If a subject is unable to use the assigned CGM, the subject should collect morning glucose levels at least 2 mornings per week throughout the study (Table 1). Subjects should adhere to their prescribed overnight cornstarch diet. Morning glucose levels should be measured using the subject's own glucose monitoring device before taking their morning dose of cornstarch. The collection time, glucose level, and prior day's overnight cornstarch use should be collected on the Morning Glucose Level monitoring worksheet provided by the site. Subjects should bring the completed worksheets to each study visit. Data collected on the worksheet will be captured on the appropriate page of the eCRF.

10.4.4. Magnetic Resonance Imaging

An MRI of the liver will be conducted at the time points specified in the Schedule of Events (Table 1). The MRI should be completed after the controlled fasting challenge to avoid having to replace the CGM sensor prior to the fasting challenge. Magnetic resonance imaging will be conducted at the study site; however, all results will be transmitted to a central radiologist for interpretation to ensure consistent assessment across study sites. Instructions for transmitting MRI results will be provided separately. Magnetic resonance imaging results will be analyzed for hepatic fat fraction, liver volume, and the relative change in each from Study 401GSDIA01 Baseline (Day 0).

Completion of the MRI will be recorded on the appropriate page of the eCRF. At the end of the study, images may be stored for up to 15 years and analyzed to better understand the effect of DTX401 on GSDIa or other metabolic deficiencies. The choice to allow retention and future analysis will be optional.

10.4.5. Liver Ultrasound

An ultrasound of the liver will be conducted at the time points specified in the Schedule of Events (Table 1). The ultrasound should be completed prior to the controlled fasting glucose challenge. The liver ultrasound will be conducted at the study site and analyzed for liver volume.

10.5. Biomarker Assessments

Not applicable.

10.6. Patient-Reported and Clinician-Reported Outcomes

10.6.1. Health-Related Quality of Life and Sleep Quality Assessments

Subjects will be asked to complete the following HRQoL and sleep quality assessments (Table 1).

- Patient-Reported Outcomes Measurement Information System (PROMIS®)-29 (Appendix 1)
- PROMIS Social Isolation (Appendix 2)
- Pittsburgh Sleep Quality Index (Appendix 3)
- Patient Global Impression of Severity (Appendix 4)
- Patient Global Impression of Change (Appendix 5)

Subjects will complete the HRQoL and sleep quality assessments prior to invasive assessments being conducted. Subjects may pause to rest during administration of the HRQoL and sleep quality assessments, but the entire set of assessments should be completed in 1 session. Study staff should review the completed forms for completeness once the subject returns the forms to staff.

Data collected on the HRQoL and sleep quality assessments will be captured on the appropriate page of the eCRF.

10.6.2. Endpoint Outcomes Interview (Year 2 – Exit Interview)

A 30-minute scripted telephone interview will be conducted at Week 104 (or at Early Withdrawal, if the subject withdraws prior to Week 104) to better quantify the subject experience in the study. The interview will be conducted by a third-party vendor, Endpoint Outcomes, specializing in the capture of caregiver and subject-reported data. Endpoint Outcome uses a script as an interview guide to serve as a basis for the discussion. The interview will be conducted by phone.

The subject has consented to the interview per the study's main ICF. However, prior to the start of the interview, Endpoint Outcomes will verbally ask for the subject's consent for the interview to be audio-recorded. With the subject's verbal consent, the interview will be audio-recorded, and the interviewer will proceed through a script. If the subject does not give permission for the session to be audio-recorded, the interview will not proceed. Subjects will be asked about their experience in the study. They may refuse to answer any question in the interview and may stop at any time.

Site staff will facilitate the phone call so that the subject's contact information remains anonymous. Personal information (including subject's name and telephone number) will remain anonymous and will not be provided to the Sponsor or Endpoint Outcomes. Subjects will be identified only by their study specific subject IDs.

All digital recordings will be stored by Endpoint Outcomes in a secure place where only Endpoints Outcome personnel directly related to this study will be able to access it. The audio interviews will be transcribed by a transcription company under contract and a confidentiality agreement, who will provide the written transcripts that will be further analyzed by Endpoint Outcomes. The transcription company is also contracted to ensure that transcripts will not contain any personal identifying information (eg, reference of first names) that may have been revealed during the interview process; personal identifying information will be removed and will not appear in the final transcript. Data from the interview will be shared with the Sponsor only after all identifying information has been removed.

10.7. Pharmacokinetic Assessments

Not applicable.

10.8. Other Assessments

10.8.1. Demographic and Medical History Assessments

As allowed by local laws and regulations, the following demographic data may be captured on the appropriate page in the eCRF: date of birth, sex, race, and ethnicity.

Medical history will be assessed at Visit 1 (Table 1). Any AEs that are ongoing at the completion of Study 401GSDIA01 will be considered medical history in Study 401GSDIA02.

10.8.2. Assessment of Prescribed Diet and Dietary Intake

The subject's prescribed diet will be reviewed at each study visit, and if possible, on a monthly basis through the final study visit (Table 1). Content, quantity, and frequency of diet intake (including daily carbohydrate intake) will be recorded on the appropriate page of the eCRF.

10.8.3. Assessment of Cornstarch (or Equivalent) Use

The subject's average daily use of cornstarch (or equivalent) will be reviewed at each study visit, and if possible, on a monthly basis through the final study visit (Table 1). Follow-up phone calls may occur regularly (approximately once a month) to further discuss if adjustments are needed for a subject's cornstarch use. Quantity and frequency of intake will be recorded on the appropriate page of the eCRF.

10.9. Appropriateness of Measurements

All measurements will be performed using standard methods that are generally recognized as reliable, accurate, and relevant.

11. SAFETY DATA COLLECTION, RECORDING, AND REPORTING

11.1. Reporting and Follow-up of Adverse Events

11.1.1. Adverse Event Terminology

An **AE** is defined as any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) products.

An **SAE** is an AE that at any dose, in the view of either the Investigator or Sponsor, results in any of the following serious outcomes:

- Death
- A life-threatening AE
- *A life-threatening AE is an event that, in the view of either the Investigator or Sponsor, places the patient or subject at immediate risk of death. It does not include an AE or suspected adverse reaction that, had it occurred in a more severe form, might have caused death.*
- Inpatient hospitalization or prolongation of existing hospitalization
- *Note that hospitalizations planned prior to study enrollment (eg, for elective surgeries) are not considered SAEs; however, an event that prolongs this type of hospitalization would be considered an SAE. Hospitalizations that occur for pre-existing conditions that are scheduled after study enrollment are considered SAEs.*
- Persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly/birth defect
- Important medical events that may not result in death, be immediately life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in the definition. 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 inpatient hospitalization, or the development of drug dependency or drug abuse

Nonemergent hospitalization for cornstarch management will not be considered an SAE.

A **suspected adverse reaction** is any AE for which there is a reasonable possibility that the drug caused the AE. For the purposes of expedited safety reporting, “reasonable possibility” means there is evidence to suggest a causal relationship between the drug and the AE. Suspected

adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any AE caused by a drug.

As there is limited prior clinical experience with DTX401, reference safety information for assessing whether an AE is a suspected unexpected serious adverse reaction (SUSAR) is currently not available. Therefore, any SAE considered related to DTX401 will be considered a **SUSAR** and reported immediately. The SAE should be treated with appropriate supportive and medical care deemed necessary for the well-being of the subject.

11.1.2. Severity of Adverse Events

The severity/toxicity, or intensity, of an AE refers to the extent to which an AE affects the subject's daily activities. The Investigator should rate the intensity of an AE as Grade 1, 2, 3, 4, or 5 based on their medical judgment. The most current version of the National Cancer Institute Common Terminology Criteria for Adverse Events (NCI, 2018) can be used to guide the rating of an AE.

In the event that an AE occurs during the study that is not captured by the CTCAE, the AE should be graded according to the general guidelines outlined in Table 3.

Table 3: General Guidelines for Grading Events Not Captured by the CTCAE

Grade	Criteria
Grade 1	Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
Grade 2	Moderate; minimal, local, or noninvasive intervention indicated; limiting age-appropriate instrumental ADL.[1]
Grade 3	Severe or medically significant but not immediately life threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL.[2]
Grade 4	Life-threatening consequences; urgent intervention indicated.
Grade 5	Death related to AE.

Abbreviations: ADL = activities of daily living; AE = adverse event; CTCAE = Common Terminology Criteria for Adverse Events.

[1] Instrumental ADL refers to preparing meals, shopping for groceries or clothes, using the telephone, managing money, etc.

[2] Self-care ADL refers to bathing, dressing and undressing, feeding self, using the toilet, taking medications, and not bedridden.

Changes in the severity/toxicity of an AE should be documented to allow an assessment of the duration of the event at each level of intensity to be performed. Adverse events characterized as intermittent do not require documentation of onset and duration of each episode.

Terminology Clarification: The term "severe" is often used to describe the intensity (severity) of a specific event (as in mild, moderate, or severe myocardial infarction); the event itself, however, may be of relatively minor medical significance (such as severe headache). The severity is not the same as "serious" which is based on subject/event outcome or action criteria and is usually associated with events that pose a threat to a subject's life or functioning (Section 11.1.1). Seriousness (not severity) serves as a guide for defining regulatory reporting obligations (Section 11.1.4.2).

11.1.3. Relationship of Adverse Events to Investigational Product

The Investigator will assess the potential relationship of the AE to IP using the following descriptions.

Categories of attributions for "unrelated" events:

- ***Unrelated:*** This category applies to an AE that ***is clearly not related*** to the investigational agent/procedure.

Categories of attributions for "related" events:

- ***Possibly Related:*** This category applies to an AE that ***may be related*** to the investigational agent/procedure.
- ***Probably Related:*** This category applies to an AE that ***is likely related*** to the investigational agent/procedure.
- ***Definitely Related:*** This category applies to an AE that ***is clearly related*** to the investigational agent/procedure.

For the purposes of reporting to regulatory agencies, AEs deemed as definitely, probably or possibly related will be considered related.

11.1.4. Adverse Event Reporting

11.1.4.1. General

All AEs (ie, any new condition or worsening in severity or frequency of a preexisting condition) with onset after the subject signs consent for study participation must be promptly documented on the eCRF. The Investigator is responsible for evaluating all AEs, obtaining supporting documents, and ensuring documentation of the events is adequate. Details of the AEs must include severity, relationship to IP, duration, and outcome.

All AEs will be collected from the time the subject signs the ICF and up to the EOS (Week 312 or Week 208 if the last subject enrolls into the DMP) or Early Withdrawal visit. In addition, the Investigator should report any AE they are made aware of that occurs after this time period and that is believed to have a reasonable possibility of being associated with IP. Serious AEs will be recorded for up to 30 days after the EOS or Early Withdrawal visit.

Any AEs ongoing at 30 days following the EOS visit or Early Withdrawal visit should have a comment in the source document by the Investigator that the event has recovered, recovered with sequelae, or stabilized.

11.1.4.2. Requirements for Immediate Reporting of Serious Adverse Events and Serious Adverse Drug Reactions

Ultragenyx or its designee must be notified of the occurrence of any SAE that occurs during the reporting period within 24 hours of the Investigator, designee, or site personnel's knowledge of the event. The study site should record all SAE information on the SAE page in the eCRF and submit the report via the electronic data capture (EDC) system. In the event of any fatal or life-threatening SAE, the Investigator must immediately inform Ultragenyx or its designee by telephone and report the SAE in the EDC system. If, for any reason, it is not possible to report

the SAE in the EDC system (eg, the EDC system is unavailable), the study site should record the SAE on the paper SAE Reporting Form and submit it by fax or email. As soon as it is possible, any SAE reported via fax must be entered into the EDC system. See Section 11.4 for contact information for SAE reporting.

Follow-up SAE information must be submitted in a timely manner as additional information becomes available. All SAEs regardless of relationship to IP must be followed to resolution or stabilization if improvement is not expected.

All deaths, regardless of causality, occurring from the time the subject signs the ICF until 30 days following the EOS (Week 312 or Week 208 if the last subject enrolls into the DMP) or Early Withdrawal visit are to be reported as SAEs to Ultragenyx or its designee within 24 hours of knowledge.

11.1.4.3. Requirements for Reporting of Pregnancy in Subject or Partner

Ultragenyx or its designee must be notified of the occurrence of any pregnancy in a subject or subject's partner that occurs during the reporting period within 2 weeks of the Investigator, designee, or site personnel's knowledge of the event. Pregnancies must be reported by completing and submitting the paper Pregnancy Report Form to Ultragenyx or designee by fax. See Section 11.4 for contact information for pregnancy reporting.

Any reported pregnancy of a subject or a subject's partner that occurs during participation in the study will be monitored for the full duration of the study and/or followed until the outcome of the pregnancy is known. In the event of a pregnancy in the partner of a subject, the Investigator should make every effort to obtain the female partner's consent for release of protected health information.

Ultragenyx or its designee must be notified of the outcome of the pregnancy within 2 weeks of the Investigator, designee, or site personnel's knowledge of the outcome. Pregnancy outcomes must be reported by completing and submitting the paper Pregnancy Report Form to Ultragenyx or designee by fax. All neonatal deaths that occur within 28 days of birth should be reported as an SAE.

Pregnancy-associated SAEs will be processed and submitted, as necessary, per the suspected unexpected serious adverse reaction (SUSAR) reporting process indicated in Section 11.1.5.1.

11.1.5. Safety Communication Plan

11.1.5.1. Serious Adverse Drug Reaction Reporting

Ultragenyx or its designee will submit SUSARs to appropriate Regulatory Authorities (including Competent Authorities in all Member States concerned), Institutional Review Boards (IRBs)/Independent Ethics Committees (IECs), and Investigators as per local laws and regulations. Fatal and life-threatening SUSARs will be submitted no later than 7 calendar days of first knowledge of the event and follow-up information submitted within an additional 8 days. All other SUSARs will be submitted within 15 calendar days of first knowledge of the event.

The Investigator will notify the IRBs/IECs of SAEs, in accordance with IRB/IEC requirements and local laws and regulations. A copy of this notification must be provided to Ultragenyx or its designee.

11.1.5.2. Urgent Safety Matters and Non-SUSAR Reporting

Investigators are required to report any urgent safety matters to Ultragenyx or its designee within 24 hours. Ultragenyx or its designee will inform the Regulatory Authorities, IRBs/IECs, and other Investigators of any events (eg, change to the safety profile of DTX401, major safety findings) that may occur during the clinical study that do not fall within the definition of a SUSAR but may affect the safety of subjects participating in the clinical study, as required, in accordance with applicable laws and regulations. The reporting period for urgent safety issues is the period from the signing of the ICF through 30 days following the EOS or Early Withdrawal visit.

The Investigator will notify the IRBs/IECs of urgent safety matters, in accordance with IRB/IEC requirements and local laws and regulations. A copy of this notification must be provided to Ultragenyx or its designee.

Non-SUSARs will be maintained in the PPD safety database and provided in annual safety reports and/or other aggregate periodic summary reports to Regulatory Authorities and IRBs/IECs per local laws and regulations.

11.2. Safety Data Monitoring

There will not be a Data Monitoring Committee in this study.

Potential safety signals identified during routine monitoring by the PPD Medical Monitor and Ultragenyx during the conduct of the study will be escalated to the appropriate internal Ultragenyx safety governing bodies. Any action indicated by Ultragenyx safety governing bodies will be communicated accordingly to all stakeholders, eg, Regulatory Authorities, IRBs/IECs, and Investigators.

11.3. Study Termination or Suspension Due to Safety Events

Should an unexpected and IP-related SAE occur, the IRB/IEC and appropriate regulatory authorities will be informed per local laws and regulations (Section 11.1.5.1). A full evaluation of the event will be performed to make a decision regarding what actions to take, including whether to recommend stopping the study.

Regulatory Authorities, as well as IRBs/IECs, will be informed if the study is paused or stopped. If the Sponsor deems it appropriate to restart the study following an internal safety review, this will be done only following approval by Regulatory Authorities.

11.4. Safety Contact Information

Table 4: Ultragenyx Contact Information for SAE Reporting

Ultragenyx	**Email:** Ultragenyx@primevigilance.com
	Fax: +1 415.930.4033

12. STATISTICAL METHODS

12.1. Endpoints and Determination of Sample Size

A full description of the analysis details will be provided in the statistical analysis plan (SAP).

12.1.1. Primary Endpoint

The primary endpoint of this study is as follows:

- The incidence of AEs and SAEs for each cohort and overall assessed by severity and relationship to IP

12.1.2. Secondary Endpoint

The secondary endpoint of this study is as follows:

- The change from Day 0 (Study 401GSDIA01) in time to first hypoglycemic event during a controlled fasting challenge over time by cohort and overall, following IV administration of DTX401

12.1.3. Tertiary Endpoints

The tertiary endpoints of this study are as follows:

- Total cholesterol, low-density lipoprotein, and triglycerides, by time point, cohort, and overall
- Uric acid, by time point, cohort, and overall
- 24-hour urine protein, by time point, cohort, and overall
- The change from baseline in liver size and fat fraction (by MRI) over time following IV administration of DTX401, by cohort, and overall
- The average daily use of cornstarch (or equivalent) over time, by cohort, and overall
- Assessment of serum glucose levels over time, summarized by cohort
- Subject responses to HRQoL and sleep quality assessments over time following IV administration of DTX401, by cohort and overall
- Year 2 – Exit Interviews
- The development of neutralizing antibodies to AAV8 (as determined by a cellbased assay) over time, by cohort, and overall
- The development of anti-AAV8 binding antibodies (as determined by ELISA) over time, by cohort, and overall
- The development of anti-G6Pase antibodies over time, by cohort, and overall
- Assessment of lactate levels over time, summarized by cohort and overall

12.1.4. Sample Size Considerations

The study is expected to enroll approximately up to 12 subjects. This is dependent on the number of subjects who enrolled in Study 401GSDIA01 and who meet the eligibility criteria for Study 401GSDIA02. The sample size is not based on a power calculation.

12.2. Analysis Population

12.2.1. Full Analysis Set

The full analysis set will consist of all enrolled subjects who received the dose of DTX401. Unless specified otherwise, the full analysis set will be used for all the analyses.

12.3. Planned Methods of Analysis

12.3.1. General Principles

All subjects who provide written informed consent and are enrolled in Study 401GSDIA02 will be included in the analyses.

All statistical evaluations in the study will be descriptive. No formal hypothesis will be tested. Continuous variables will be summarized by number of subjects and mean, standard deviation, standard error, median, Q1, Q3, minimum, and maximum values. Categorical variables will be summarized by number and percentage of subjects.

An integrated analysis using data from both Study 401GSDIA01 and Study 401GSDIA02 will be performed to summarize and assess the long-term safety and efficacy of DTX401; there will not be a standalone analysis of data from only Study 401GSDIA02.

Full details of the analysis will be provided in the SAP.

12.4. Safety Analyses

12.4.1. Adverse Events

All AEs will be coded according to the Medical Dictionary for Regulatory Activities. The incidence of AEs will be summarized for each cohort and overall by system organ class and preferred term. Additionally, AEs may be summarized for each cohort and overall by severity and relationship to IP, if applicable. Serious AEs will be presented for each cohort and overall by relationship to IP. Summary tables will present the total numbers of AEs as well as the number of subjects with AE incidence by system organ class and preferred term. For incidence of relationship to IP, subjects will be counted only once, in the category of the strongest relationship to IP within each system organ class/preferred term.

12.4.2. Hypoglycemic Events

The number of symptomatic hypoglycemic events occurring during the study reported by the subjects will be summarized. Details of the symptomatic hypoglycemic events will be provided in a listing.

12.4.3. MRI Results of the Liver

Magnetic resonance imaging results will be analyzed for hepatic fat fraction, liver volume, and the relative change in each from Study 401GSDIA01 Baseline (Day 0). Descriptive statistics for MRI results and changes from baseline values will be summarized by study visit.

12.4.4. Clinical Laboratory Assessments

For all clinical laboratory parameters with continuous results, absolute values and changes from baseline will be summarized by study visit. For laboratory parameters with categorical results, shifts from baseline will be summarized by study visit. Laboratory values from the central laboratory and local laboratories will be analyzed.

12.5. Efficacy Analyses

12.5.1. Symptom-Free Euglycemia

Symptom-free euglycemia will be measured using the duration of controlled fasting challenge. Duration of controlled fasting challenge is defined as time to any of the following 3 conditions: 1) glucose <54 mg/dL (<3.0 mmol/L); 2) signs/symptoms of hypoglycemia; or 3) 15 hours fasting without hypoglycemia. Change from Baseline will be provided where Day 0 in Study 401GSDIA01 is considered as baseline.

12.5.2. Use of Cornstarch (or Equivalent)

Average daily use (both quantity and frequency) of cornstarch over time will be summarized in a table and provided in the listings.

12.5.3. Continuous Glucose Monitoring

Continuous glucose monitoring device data will be summarized.

12.5.4. Morning Glucose Levels

Glucose levels will be summarized in a table and provided in the listings.

12.5.5. Health-Related Quality of Life and Sleep Quality

Health-related quality of life and sleep quality over time will be summarized.

12.6. Immunogenicity Analysis

The development of neutralizing antibodies to AAV8 (as determined by a cell-based assay), AAV8 binding antibody IgG (as determined by ELISA), cell-mediated immune response to AAV8 and G6Pase, and anti-G6Pase antibodies over time will be summarized.

12.7. Interim Analyses

Administrative analyses may be performed during the study to support registrational activities, respond to regulatory inquiries, or at the Sponsor’s discretion.

12.8. Data Monitoring Committee

Not applicable.

13. STUDY ETHICS AND CONDUCT

13.1. Ethics

13.1.1. Institutional Review Board or Independent Ethics Committee and Competent Health Authority

The IRB/IEC must be a properly constituted board or committee operating in accordance with International Council for Harmonisation (ICH) Good Clinical Practice (GCP) guidelines and any local ethical and regulatory requirements (eg, 21 CFR Part 56, "Institutional Review Boards," EU Clinical Trials Directive 2001/20/EC, or later guidance). This protocol, any protocol amendments, and the associated ICFs must be reviewed and approved by the IRB/IEC and Competent Health Authority before screening of any subject.

IRB/IEC and Competent Health Authority approval of any protocol amendments must be received before any of the changes outlined in the amendments are put into effect, except when the amendment has been enacted to protect subject safety. In such cases, the chair of the IRB/IEC should be notified immediately and the amendment forwarded to the IRB/IEC and Competent Health Authority for review and approval.

13.1.2. Ethical Conduct of Study

This protocol is written in accordance with the principles established by the 18th World Medical Assembly General Assembly (the Declaration of Helsinki, 1964) and subsequent amendments and clarifications adopted by the General Assemblies. The Sponsor and Investigator will make every effort that the study described in this protocol is conducted in full conformance with those principles, ICH GCP, and applicable local law, eg, Food and Drug Administration (FDA) regulations, European Union (EU) regulations, and any other local ethical and regulatory requirements ("Applicable Study Conduct Obligations") and principles, current Food and Drug Administration (FDA) regulations, EU Clinical Trials Directive 2001/20/EC or later, ICH GCP guidelines, and local ethical and regulatory requirements. Should a conflict arise, the Sponsor and Investigator will follow whichever law or guideline affords the greater protection to the individual subject.

It is the Investigator's responsibility to inform the Sponsor of any serious breach of the protocol (ie, a breach likely to affect to a significant degree the safety and rights of a subject or the reliability and robustness of the data generated in the clinical study) within 24 hours of identification of the breach. The Sponsor will inform the regulatory authorities of the serious breach as required by applicable law or regulation.

13.1.3. Subject Information and Consent

Appropriate forms for documenting written informed consent will be provided by the Investigator and reviewed and approved by Ultragenyx or its designee before submission to the IRB/IEC.

It is the Investigator's responsibility to obtain written informed consent from each potential study subject prior to the conduct of any study procedures. This written informed consent will be obtained after the methods, objectives, requirements, and potential risks of the study have been

fully explained to each potential subject. The Investigator must explain to each subject that the subject is completely free to refuse to enter the study or to withdraw from it at any time.

The method of obtaining and documenting informed consent and the contents of the ICF will comply International Council for Harmonisation (ICH) Good Clinical Practice (GCP) guidelines and any local ethical and regulatory requirements (eg, 21 CFR Part 56, "Institutional Review Boards," EU Clinical Trials Directive 2001/20/EC, or later guidance). Subjects will be given a copy of the signed ICF and will be provided any new information during the course of the study that might affect their continued participation in the study. The Investigator or a qualified designee will be available to answer each subject's questions throughout the study, and all of the subject's questions must be answered to the subject's satisfaction. If the protocol is amended and the ICF is revised, each subject will be required to provide written informed consent again using the revised ICF.

The date of written informed consent will be documented in each potential subject's eCRF. The signed ICF will remain in each subject's study file and must be available to the study monitor(s) at all times.

13.1.4. Data Protection, Anonymization and Security

Any study data transferred to Ultragenyx will be coded and will not contain names or any other information that would make the subject identifiable.

Study information is not to be disclosed to any third party (except employees or agents directly involved in the conduct of the study or as required by law) without prior written consent from Ultragenyx.

The subject must be informed of his/her rights under applicable data protection laws regarding how his/her personal information is being treated. Additionally, the subject must be informed, and where possible, be provided with the details and references, that certain persons or entities may have access to his/her medical records for various, legitimate reasons and purposes, including Ultragenyx auditors or other authorized personnel appointed by Ultragenyx may examine his/her medical records, appropriate IRB/IEC members, and by inspectors from regulatory authorities. The subject must also be provided with the applicable contact information for Ultragenyx, site, and/or the country's/regional's Data Protection Authority in order to inquire about their rights, raise concerns, or issue complaints regarding the treatment of his/her personal information by Ultragenyx.

Ultragenyx complies with applicable data protection laws and treats personal information in accordance with standard security measures. Ultragenyx has implemented policies and procedures to prevent security issues, including data breaches, and avoid/minimize any negative consequences. All computerized systems to be used during the study have been selected following a strict process that encompassed a System Level Risk Assessment (SLRA), including system identification, GxP, General Data Protection Regulation (GDPR), Sarbanes and Oxley Act, and 21 CFR Part 11 applicability. As far GDPR compliance is concerned, a Data Classification Assessment which includes the Data Privacy Impact Assessment (DPIA) was also conducted on all systems, and their results are memorialized in DPIA assessments stored in the Ultragenyx Document Management System. Ultragenyx imposes the same principles to all vendors supporting the study, which translates into (i) selecting such vendors and their systems

against equally stringent security protocols and standards; (ii) ensuring that they comply with applicable data protection laws, and (iii) agreeing to specific data protection terms that include, among others, a data breach response plan and an obligation to collaborate with Ultragenyx in such circumstances timely. These measures exist so Ultragenyx can mitigate any security occurrence and meet their obligations under applicable data protection laws. Therefore, in case of a data breach, Article 33 GDPR will be followed. Additionally, in order to maintain/restore a GxP-compliant environment, an audit will be conducted, and the necessary remedial actions implemented in a timely fashion (including, but not limited to, termination of any relevant third party from the research) to guarantee the integrity of the concerned data and avoid any future repetition.

13.1.5. Future Use of Stored Study Samples

Not applicable.

13.2. Investigators and Study Administrative Structure

Each Investigator must provide Ultragenyx and/or its designee a completed and signed Form FDA 1572, or equivalent, if applicable based on local requirements, and a Financial Disclosure Form. All subinvestigators must be listed on Form FDA 1572, or equivalent, and Financial Disclosure Forms must be completed for all subinvestigators listed on Form FDA 1572.

13.2.1. Investigator Documentation

Prior to beginning the study, the Investigator will be asked to comply with ICH E6 (R2) 8.2 and Title 21 of the CFR by providing the following essential documents, including but not limited to:

- IRB/IEC/IBC approvals
- Original Investigator-signed Investigator agreement page of the protocol
- Form FDA 1572 (or equivalent), fully executed, and all updates on a new fully executed Form FDA 1572 (or equivalent)
- Curriculum vitae for the Investigator and each subinvestigator
- Financial disclosure information to allow the sponsor to submit complete and accurate certification or disclosure statements required under 21 CFR §54. In addition, the Investigators must provide to the sponsor a commitment to promptly update this information if any relevant changes occur during the course of the investigation and for 1 year after the completion of the study
- IRB/IEC/IBC-approved ICF, samples of study site advertisements for recruitment for this study, and any other written information regarding this study that is to be provided to the subject or their legal guardian
- Laboratory certifications and normal ranges for any local laboratories used by the study site, in accordance with 42 CFR §493

13.3. Investigational Product Accountability

Not applicable.

13.4. Data Handling and Record Keeping

13.4.1. Case Report Forms and Source Documents

The Investigator is required to initiate and maintain, for each subject, an adequate and accurate case history that records all observations and other data related to the study for that subject. A validated EDC system will be used for entry of the data into eCRFs. Data must be recorded on eCRFs approved by Ultragenyx or its designee. All information recorded on eCRFs for this study must be consistent with the subject's source documentation.

Initial data entry and any changes to the data will be made only by Ultragenyx-authorized users, and data entries and changes will be captured in an electronic audit trail. An explanation of any data change should be recorded in the eCRF. All data entered in to the eCRF must be verifiable; therefore, eCRFs will be routinely checked for accuracy, completeness, and clarity and will be cross-checked for consistency with source documents, including laboratory test reports and other subject records by Ultragenyx or its designee. The Investigator must allow Ultragenyx or its designee direct access to all source documents.

13.4.2. Data Quality Assurance

Monitoring and auditing procedures developed by Ultragenyx and/or its designee will be implemented to ensure compliance with FDA and ICH GCP guidelines. The Ultragenyx designated representative (the monitor) will contact the Investigator and conduct regular visits to the study site, according to the monitoring plan. The monitor will also be responsible for confirming adherence to the study protocol, inspecting eCRFs and source documents, and ensuring the integrity of the data. Instances of missing or uninterpretable data will be resolved in coordination with the Investigator.

The monitor will also investigate any questions concerning adherence to regulatory requirements. Any administrative concerns will be clarified and followed. The monitor will maintain contact with the site through direct communications with the study site by e-mail, telephone, facsimile, and/or mail. The Investigator and all other site personnel must agree to cooperate fully with the monitor and will work in good faith with the monitor to resolve any and all questions raised and any and all issues identified by the monitor.

The Investigator should understand that regulatory authorities, the IRB/EC, and/or Ultragenyx or its designees have the right to access all eCRFs, source documents, and other study documentation for on-site audit or inspection and will retain this right from the start of the study to at least 2 years after the last approval of a marketing application or for at least 2 years after clinical development of the IP for the indication being studied has been discontinued. The Investigator is required to guarantee access to these documents and to cooperate with and support such audits and inspections.

13.4.3. Record Retention

All study records must be retained in strict accordance with the Clinical Trial Agreement executed between Ultragenyx and the Institution and/or Investigator. For study monitoring, audit, or inspection, the IRB/IEC and Sponsor or its designees have the right to access all CRFs, source documents, and other study documentation. The Investigator or study site will retain such

documents from the start of the study to at least 25 years after the close of the study, guarantee access to these documents, and cooperate with and support such audits and inspections.

13.5. Financing and Insurance

Financing and insurance for this clinical study will be addressed in the Clinical Trial Agreement between Ultragenyx and the Institution and/or Investigator.

13.6. Publication Policy

Any publication or presentation by the Investigator and/or the Institution associated with the study will be in strict accordance with the Clinical Trial Agreement executed between Ultragenyx and the Institution and/or Investigator.

14. REFERENCES

Boers SJ, Visser G, Smit PG, et al. (2014) Liver transplantation in glycogen storage disease type I. *Orphanet J Rare Dis* 9: 47.

Bolt MW, Brady JT, Whiteley LO, et al. (2021) Development challenges associated with rAAV-based gene therapies. *J Toxicol Sci* 46(2): 57-68.

CBER (2006) *Food and Drug Administration, Department of Health and Human Services (US). Guidance for industry: Gene therapy clinical trials – observing subjects for delayed adverse events. November 2006. [cited 2018 Feb 05] [25 screens]. Available from: https://www.fda.gov/downloads/BiologicsBloodVaccines/GuidanceComplianceRegulatoryInformation/Guidances/CellularandGeneTherapy/ucm078719.pdf*.

Chen YT, Coleman RA, Scheinman JI, et al. (1988) Renal disease in type I glycogen storage disease. *N Engl J Med* 318(1): 7-11.

CHMP (2009) Guideline on follow-up of patients administered with gene therapy medicinal products. EMEA Doc Ref. EMEA/CHMP/GTWP/60436/2007. London: European Medicines Agency.

Chou JY (2001) The molecular basis of type 1 glycogen storage diseases. *Curr Mol Med* 1(1): 25-44.

Chou JY and Mansfield BC (1999) Molecular Genetics of Type 1 Glycogen Storage Diseases. *Trends Endocrinol Metab* 10(3): 104-113.

Chou JY and Mansfield BC (2011) Recombinant AAV-directed gene therapy for type I glycogen storage diseases. *Expert Opin Biol Ther* 11(8): 1011-1024.

Davis MK and Weinstein DA (2008) Liver transplantation in children with glycogen storage disease: controversies and evaluation of the risk/benefit of this procedure. *Pediatr Transplant* 12(2): 137-145.

Foster JD and Nordlie RC (2002) The biochemistry and molecular biology of the glucose-6-phosphatase system. *Exp Biol Med (Maywood)* 227(8): 601-608.

Franco LM, Krishnamurthy V, Bali D, et al. (2005) Hepatocellular carcinoma in glycogen storage disease type Ia: a case series. *J Inherit Metab Dis* 28(2): 153-162.

Hinderer C, Katz N, Buza EL, et al. (2018) Severe toxicity in nonhuman primates and piglets following high-dose intravenous administration of an AAV vector expressing human SMN. *Hum Gene Ther* 29(3): 285-298.

Hordeaux J, Buza EL, Dyer C, et al. (2020a) Adeno-Associated Virus-Induced Dorsal Root Ganglion Pathology. *Hum Gene Ther* 31(15-16): 808-818.

Hordeaux J, Buza EL, Jeffrey B, et al. (2020b) MicroRNA-mediated inhibition of transgene expression reduces dorsal root ganglion toxicity by AAV vectors in primates. *Sci Transl Med* 12(569).

Koeberl DD, Pinto C, Brown T, et al. (2009) Gene therapy for inhereted metabolic disorders in companion animals. *ILAR J* 50(2): 122-127.

Koeberl DD, Pinto C, Sun B, et al. (2008) AAV vector-mediated reversal of hypoglycemia in canine and murine glycogen storage disease type Ia. *Mol Ther* 16(4): 665-672.

Lee YM, Jun HS, Pan CJ, et al. (2012) Prevention of hepatocellular adenoma and correction of metabolic abnormalities in murine glycogen storage disease type Ia by gene therapy. *Hepatology* 56(5): 1719-1729.

Lei KJ, Chen YT, Chen H, et al. (1995a) Genetic basis of glycogen storage disease type 1a: prevalent mutations at the glucose-6-phosphatase locus. *Am J Hum Genet* 57(4): 766-771.

Lei KJ, Pan CJ, Liu JL, et al. (1995b) Structure-function analysis of human glucose-6-phosphatase, the enzyme deficient in glycogen storage disease type 1a. *J Biol Chem* 270(20): 11882-11886.

Lei KJ, Shelly LL, Lin B, et al. (1995c) Mutations in the glucose-6-phosphatase gene are associated with glycogen storage disease types 1a and 1aSP but not 1b and 1c. *J Clin Invest* 95(1): 234-240.

Lei KJ, Shelly LL, Pan CJ, et al. (1993) Mutations in the glucose-6-phosphatase gene that cause glycogen storage disease type 1a. *Science* 262(5133): 580-583.

Matern D, Starzl TE, Arnaout W, et al. (1999) Liver transplantation for glycogen storage disease types I, III, and IV. *Eur J Pediatr* 158 Suppl 2: S43-48.

NCI (2018) National Cancer Institute Division of Cancer Treatment and Diagnosis (DCTD), National Cancer Institute (NCI), National Institutes of Health (NIH), Department of Health and Human Services (DHHS). Common Terminology Criteria for Adverse Events V4.3 (CTCAE). Available at:
https://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm#ctc_50.

Pan CJ, Lei KJ, Annabi B, et al. (1998) Transmembrane topology of glucose-6-phosphatase. *J Biol Chem* 273(11): 6144-6148.

Reddy SK, Kishnani PS, Sullivan JA, et al. (2007) Resection of hepatocellular adenoma in patients with glycogen storage disease type Ia. *J Hepatol* 47(5): 658-663.

Schuster DJ, Dykstra JA, Riedl MS, et al. (2014) Biodistribution of adeno-associated virus serotype 9 (AAV9) vector after intrathecal and intravenous delivery in mouse. *Front Neuroanat* 8: 42.

Van Alstyne M, Tattoli I, Delestree N, et al. (2021) Gain of toxic function by long-term AAV9-mediated SMN overexpression in the sensorimotor circuit. *Nat Neurosci*. Epub ahead of print 2021/04/03. DOI: 10.1038/s41593-021-00827-3.

van Schaftingen E and Gerin I (2002) The glucose-6-phosphatase system. *Biochem J* 362(Pt 3): 513-532.

Weinstein DA, Correia CE, Conlon T, et al. (2010) Adeno-associated virus-mediated correction of a canine model of glycogen storage disease type Ia. *Hum Gene Ther* 21(7): 903-910.

Wolfsdorf JI and Crigler JF, Jr. (1997) Cornstarch regimens for nocturnal treatment of young adults with type I glycogen storage disease. *Am J Clin Nutr* 65(5): 1507-1511.

Wolfsdorf JI, Holm IA and Weinstein DA (1999) Glycogen storage diseases. Phenotypic, genetic, and biochemical characteristics, and therapy. *Endocrinol Metab Clin North Am* 28(4): 801-823.

Wolfsdorf JI, Laffel LM and Crigler JF, Jr. (1997) Metabolic control and renal dysfunction in type I glycogen storage disease. *J Inherit Metab Dis* 20(4): 559-568.

15. APPENDICES

APPENDIX 1. PROMIS-29

APPENDIX 2. PROMIS SOCIAL ISOLATION – SHORT FORM 4A

APPENDIX 3. PITTSBURGH SLEEP QUALITY INDEX (PSQI)

APPENDIX 4. PATIENT GLOBAL IMPRESSION OF SEVERITY (PGI-S)

APPENDIX 5. PATIENT GLOBAL IMPRESSION OF CHANGE (PGI-C)