Official Title:	**A Phase 1/2, Single-Blind, Placebo-Controlled, Single and Multiple Ascending Dose Safety, Tolerability, Pharmacokinetic and Pharmacodynamics Study of Subcutaneously Administered ALN-GO1 in Healthy Adult Subjects, and Patients With Primary Hyperoxaluria Type 1**
NCT Number:	**NCT02706886**
Document Date:	**Clinical Study Protocol, Amendment 5, 14 February 2018**

CLINICAL STUDY PROTOCOL
ALN-GO1-001

Protocol Title:	A Phase 1/2, Single-Blind, Placebo-Controlled, Single- and Multiple-Ascending Dose Safety, Tolerability, Pharmacokinetic and Pharmacodynamics Study of Subcutaneously Administered ALN-GO1 in Healthy Adult Subjects, and Patients with Primary Hyperoxaluria Type 1
Investigational Drug:	ALN-GO1
EudraCT Number:	2015-004407-23
Protocol Date:	Original Protocol 18 December 2015 Amendment 1_01 July 2016 Amendment 2_21 September 2016 Amendment 3_09 December 2016 Amendment 4_27 June 2017 Amendment 5_14 February 2018
Sponsor:	Alnylam Pharmaceuticals, Inc. 300 Third Street Cambridge, MA 02142 USA Telephone: +1-617-551-8200
Sponsor Contact:	[redacted], MD [redacted]

The concepts and information contained in this document or generated during the study are considered proprietary and may not be disclosed in whole or in part without expressed written authorization of Alnylam Pharmaceuticals, Inc.

SPONSOR PROTOCOL APPROVAL

I have read this protocol and I approve the design of this study.

[redacted], MD

14 Feb 2018

Date

INVESTIGATOR'S AGREEMENT

I have read the ALN-GO1-001 protocol and agree to conduct the study as outlined. I agree to maintain the confidentiality of all information received or developed in connection with this protocol.

_____

Printed Name of Investigator

_____

Signature of Investigator

_____

Date

PROTOCOL SYNOPSIS

Protocol Title

A Phase 1/2, Single-Blind, Placebo-controlled, Single- and Multiple-ascending Dose Safety, Tolerability, Pharmacokinetic, and Pharmacodynamics Study of Subcutaneously Administered ALN-GO1 in Healthy Adult Subjects and Patients with Primary Hyperoxaluria Type 1

Product Name

ALN-GO1

Indication

Primary hyperoxaluria type 1 (PH1)

Phase

Phase 1/2

Study center(s)

The single-ascending dose (SAD) part in healthy adult subjects (Part A) will be conducted at 1 clinical study center in the United Kingdom. The multiple-ascending dose (MAD) part in patients with PH1 (Part B) is expected to take place at approximately 12 clinical study centers worldwide.

Objectives

Primary

- Evaluate the safety and tolerability of single- and multiple-ascending doses of ALN-GO1, respectively, in healthy adult subjects and in patients with PH1

Secondary

- Characterize the pharmacokinetics (PK) of ALN-GO1
- Evaluate the pharmacodynamics (PD) of ALN-GO1

Exploratory

- [redacted] exploratory [redacted]
- [redacted]

Study Design

This is a randomized, single-blind, placebo-controlled study of subcutaneously administered ALN-GO1. Subjects and patients will be randomized in a 3:1 ratio to receive either ALN-GO1 or placebo. The study is designed to evaluate the safety, tolerability, PK, and PD of single- and multiple-ascending doses of ALN-GO1 and will be conducted in 2 parts:

- Part A: SAD part in healthy adult subjects
- Part B: MAD part in adult and pediatric patients with PH1

In Part A, subjects in each cohort will be randomized to receive 1 dose of ALN-GO1 or placebo.

In Part B, patients will be enrolled in up to 6 sequential dose cohorts to receive ALN-GO1 or placebo monthly or quarterly. Patients dosed monthly will receive 3 doses of ALN-GO1 or placebo in a blinded fashion. After completion of the blinded portion of the study, patients dosed monthly will be unblinded on or after Day 78. Patients who initially received placebo will then receive 3 doses of open-label ALN-GO1 dosed monthly at the same dose administered to the cohort into which they were initially randomized and will follow the same assessment schedule. Patients dosed quarterly will receive either ALN-GO1 or placebo on Day 1. All patients in quarterly dosing cohorts, including those initially randomized to placebo, will receive open-label ALN-GO1 on Day 85 at the same dose administered to the cohort into which they were initially randomized. Up to 2 expansion cohorts in

Part B may be enrolled based on available safety and PD data; these patients will all receive open-label ALN-GO1, not placebo.

After the dosing period, patients will return to the clinical study center for continued safety, tolerability, PK, and PD monitoring for at least 12 weeks (84 days) following the last dose of study drug. Following completion of the 12-week postdose follow-up period, patients will be invited to participate in an open-label extension study.

For patients who do not enroll in the open-label extension study, safety and PD follow up will continue until 24-hour urinary oxalate is >80% of baseline, and plasma glycolate is <20% above baseline or ≤ the ULN. If an investigator wishes to discontinue follow-up after completion of the postdose follow-up period and prior to oxalate and glycolate recovery, the Safety Review Committee (SRC) must agree based upon consideration of emerging data on the safety of ALN-GO1 knockdown and the individual patient's safety and PD data.

The SRC will perform ongoing reviews of safety, tolerability, and available PD data, with the primary purpose of protecting the safety of subjects/patients participating in this clinical study.

Number of Planned Subjects and Patients

A total of up to 64 participants (including optional and expansion cohorts) are planned to be enrolled in this study as follows:

- Part A: Up to 40 healthy adult subjects
- Part B: Up to 24 adult and pediatric patients with PH1

Diagnosis and Main Eligibility Criteria

Part A will include healthy subjects, aged 18 to 64 years, inclusive. Part B will include patients diagnosed with PH1, aged 6 to 64 years, inclusive, with relatively well-preserved renal function. Diagnosis of PH1 will be based on the presence of alanine glyoxylate aminotransferase (AGXT) mutations or reduced hepatic AGT enzyme activity.

Investigational Product, Dose and Mode of Administration

ALN-GO1 is a synthetic, double-stranded small interfering RNA oligonucleotide directed against hydroxyacid oxidase 1 mRNA that is covalently linked to a ligand containing 3 N-acetylgalactosamine residues. ALN-GO1 will be supplied as a sterile solution for subcutaneous (SC) injection at a targeted concentration of 200 mg/mL. The starting dose for Part A will be 0.3 mg/kg. The starting dose for Part B was determined to be 1.0 mg/kg by the SRC based on review of safety, tolerability, and available PD data, from Part A. Additional cohorts may be enrolled at higher, lower, or intermediate dose levels, but will not exceed the maximum administered dose of 6.0 mg/kg, and will follow the protocol-specified dose escalation criteria.

Reference Therapy, Dose and Mode of Administration

Placebo will be supplied by the clinical study center as a sterile, preservative-free normal saline 0.9% solution for SC injection.

Duration of Treatment and Overall Duration of Study

The duration of treatment is as follows:

- Part A: The estimated total time on study, inclusive of screening and safety and PD follow-up, for each subject is up to 405 days. The duration of treatment is a single dose.
- Part B: For all patients, the duration of screening is up to 45 days and the minimum duration of postdose follow-up is 84 days. The duration of treatment and estimated total time on study, inclusive of screening, for each patient is as follows:
  - For patients dosed monthly: The duration of treatment for patients initially randomized to receive active study drug is 57 days. The estimated total time on study is up to 462 days. Additionally, the duration of treatment for patients initially randomized to receive placebo is 141 days. The estimated total time on study for each patient initially randomized to receive placebo, then active study drug, is up to 546 days.
  - For patients dosed quarterly: The duration of treatment is 85 days for patients randomized to placebo and active study drug. The estimated total time on study is up to 490 days.

The overall duration of the study is estimated to be 4 years, including enrollment.

Endpoints

Primary

- The primary endpoint is the incidence of adverse events. Safety will also be evaluated through vital signs, electrocardiograms, clinical laboratory assessments, and physical examinations.

Secondary

Secondary Endpoints for Part A

- PK parameters (including, but not limited to, maximum plasma concentration [C_{max}], time to reach maximum plasma concentration [t_{max}], area under the plasma concentration versus time curve [AUC], apparent terminal elimination half-life [$t_{½}$], fraction eliminated in urine [f_e/F], and renal clearance [CL_R])
- Plasma and urine glycolate concentrations

Secondary Endpoints for Part B

- PK parameters including, but not limited to, C_{max}, t_{max}, AUC, $t_{½}$, f_e/F, and CL_R
- Urinary oxalate excretion (oxalate concentration in 24-hour urine collection)
- Urinary glycolate excretion (glycolate concentration in 24-hour urine collection)
- Plasma glycolate concentration
- Calculated creatinine clearance

Exploratory

- Plasma oxalate concentration
- [redacted]

[redacted]

  - [redacted]
  - [redacted]
  - [redacted]

Statistical Methods

Statistical analyses will be primarily descriptive. Data from Parts A and B will be analyzed separately. Tabular summaries will be generated by dose level and dosing regimen of ALN-GO1 and placebo (pooled across all cohorts) for Part A and Part B (through Day 85).

Safety and PD data will be summarized for Part B by dose level and dosing regimen of ALN-GO1 compared to placebo for data collected up to, and including, study Day 85. Data collected after Day 85 will be summarized separately for patients in quarterly dosing cohorts who receive a second dose of ALN-GO1 on Day 85. Data from placebo patients from all cohorts will be combined. Additionally, all safety and PD data collected from all patients in Part B during the ALN-GO1 dosing period, regardless of treatment sequence, will be combined and summarized by ALN-GO1 dose level and regimen.

Data collected beyond the designated dosing period will be summarized or presented in data listings.

Descriptive statistics will be presented for continuous variables. Frequencies and percentages will be presented for categorical and ordinal variables. Percentages will be based on the number of non-missing values in a dose group.

Table 1: Schedule of Assessments for Single-ascending Dose Cohorts in Healthy Subjects (Part A)

Study Stage	Screening	Dosing Period			Postdose Follow-up Period					Safety and PD Follow-up[a]
Study Day	**Days -45 to -2**	**Day -1[b]**	**Day 1**	**Day 2[c]**	**Day 8**	**Day 15**	**Day 29**	**Day 43**	**Day 57/EOT[d]**	**Q28D**
Visit Window (days)						(±2)	(±3)	(±3)	(±3)	(-2 to +5)
Informed consent	X									
Demography	X									
Medical history[e]	X	X								
Inclusion/exclusion criteria	X	X								
Full physical examination[f]	X								X	
Symptom-directed physical examination[f]		X	X	X	X	X	X	X		X
Height	X									
Body weight and BMI[g]	X	X							X	X
Vital signs[h]	X	X	X	X	X	X	X	X	X	X
12-lead ECG[i]	X	X	X	X			X		X	X
Pregnancy test/FSH screening[j]	X	X					X		X	X
Clinical laboratory assessments[k]	X	X	X	X	X	X	X	X	X	X
Urine sample for drugs of abuse[l]	X	X								
Randomization			X							
Study drug administration[m]			X							
Blood and urine samples for PK analyses[n]			X	X						
Blood samples for PD analyses[o]	X		X		X	X	X	X	X	X
Blood and urine samples for exploratory [redacted]			X		X		X		X	
Blood sample for ADA analysis			X				X		X	

Table 1: Schedule of Assessments for Single-ascending Dose Cohorts in Healthy Subjects (Part A)

Study Stage	Screening	Dosing Period			Postdose Follow-up Period					Safety and PD Follow-up[a]
Study Day	**Days -45 to -2**	**Day -1[b]**	**Day 1**	**Day 2[c]**	**Day 8**	**Day 15**	**Day 29**	**Day 43**	**Day 57/EOT[d]**	**Q28D**
Visit Window (days)						(±2)	(±3)	(±3)	(±3)	(-2 to +5)
Review/record AEs[p]			X							
Prior and concomitant medications	X									

Abbreviations: ADA=antidrug antibodies; AE=adverse event;; BMI=body mass index; D=Day; ECG=electrocardiogram; EOT=end of treatment; FSH=follicle-stimulating hormone; ICF=informed consent form; LFT=liver function tests; PD=pharmacodynamics; PK=pharmacokinetic; Q=every; SC=subcutaneous.

Notes:

- When scheduled at the same time points, the assessments of vital signs and 12-lead ECGs must be performed first, before the physical examinations, blood sample collections, and urine collections.
- Assessments should be performed predose, where applicable, and unless otherwise noted.

[a] Safety and PD follow-up will continue: 1) for at least 57 days, and 2) until plasma glycolate decreases to a level that is no more than 20% above of baseline or until plasma glycolate is below the upper limit of normal (≤14 nmol/mL).

[b] Subjects will be admitted to the clinical study center for predose assessments.

[c] Subjects will be discharged from the clinical study center following the completion of the assessments.

[d] If a subject chooses to withdraw consent, every effort should be made to conduct the assessments performed at the EOT visit.

[e] Complete medical history will be obtained at screening and any changes will be updated on admission to the clinical study center. Events occurring after signing of the ICF and before study drug administration will be captured as medical history.

[f] See Section 7.5.3 for assessments to be performed during a full physical examination or symptom-directed examination.

[g] Day -1 body weight will be used for calculation of the ALN–GO1 or placebo (study drug) dose to be administered.

[h] Vital signs (blood pressure, pulse rate, body temperature, and respiration rate) will be measured with the subject in the supine position after the subject has rested comfortably for 10 minutes. On Day 1 only, vital signs will be measured within 1 hour predose; and 30 (±5 minutes) and 4 hours (±15 minutes) postdose.

[i] All 12-lead ECGs are triplicate. Triplicate 12-lead ECGs will be measured approximately 5 minutes apart. Recordings will be obtained after the subject has rested comfortably in the supine position for approximately 10 minutes. Subjects should remain supine between ECGs. ECGs should be performed at the same time of day throughout the study ±1 hour. On Day 1 only, ECGs will be measured predose and 1, 2, 3, and 4 hours postdose.

[j] Pregnancy tests will be performed for women of childbearing potential only. A serum pregnancy test will be performed at Screening and urine pregnancy tests will be performed thereafter per the Schedule of Assessments and any time pregnancy is suspected. The results of the pregnancy test must be known before study drug administration. FSH will be measured at Screening only to confirm post-menopausal status.

[k] Biochemistry, hematology, coagulation, LFTs, and urinalysis parameters are described in Section 7.5.6. Day -1 clinical laboratory assessment results must be reviewed before study drug administration. Clinical laboratory tests will be evaluated by a local laboratory. Day 1 predose LFT and creatinine measurements will be locally analyzed and centrally confirmed.

[l] Drugs of abuse are described in Section 7.5.6.3.

[m] Study drug will be administered via SC injection as described in Section 6.2.2.

[n] Blood and urine samples for PK analysis will be collected at the time points listed in Table 10.

[o] Fasting blood samples should be collected. On Day 1, the blood sample for PD analysis must be collected within 1 hour predose. The remaining PD samples should be collected at the same time of day (±1 hour). See Laboratory Manual for instructions on sample processing and aliquoting of samples for storage and PD analyses.

[p] AEs occurring after signing of the ICF and before study drug administration will be captured as medical history, while all AEs that occur after study drug administration, and baseline AEs that worsen after study drug administration, must be recorded and reported as AEs.

Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B – Monthly Dosing)

Study Stage	Screening	Dosing Period												Postdose Follow-up Period[a]						Safety and PD Follow-up
Study Day (D)	-45 to -2	-1	1	2	3	15	28	29	43	56	57/EOT	58	59[b]	84	85	112	113	140	141	169; then, Q28D
Visit Window (D)						±2		±4	±4		±4				±4		±4		±4	±4
Informed consent (and assent for patients under the age of legal consent)	X																			
Demography	X																			
Medical history[c]	X	X																		
Inclusion/exclusion criteria	X	X																		
Full physical examination[d]	X																		X	
Symptom-directed physical examination[d]		X	X	X		X		X	X		X				X		X			X
Height[e]	X										X				X				X	X
Body weight and BMI	X	X[f]									X				X				X	X
Vital signs[g]	X	X	X	X		X		X	X		X				X		X		X	X
12-lead ECG[h]	X	X	X	X				X			X				X		X		X	X
Echo	X														X					X[i]
Pregnancy test/FSH screening[j]	X	X	X					X			X				X		X		X	X
Clinical laboratory assessments[k]	X	X	X	X		X		X	X		X				X		X		X	X
Troponin I[l]	X	X						X			X				X					X
Urine sample for drugs of abuse[m]	X	X																		
Randomization			X[n]																	

Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B – Monthly Dosing)

Study Stage	Screening	Dosing Period												Postdose Follow-up Period[a]						Safety and PD Follow-up
Study Day (D)	**-45 to -2**	**-1**	**1**	**2**	**3**	**15**	**28**	**29**	**43**	**56**	**57/EOT**	**58**	**59[b]**	**84**	**85**	**112**	**113**	**140**	**141**	**169; then, Q28D**
Visit Window (D)						±2		±4	±4		±4				±4		±4		±4	±4
Study drug administration[o]			X					X			X									
Blood and urine samples for PK analyses[p]			X	X	X	X		X			X	X	X		X					
Blood sample for PD analyses[q]	X		X			X		X	X		X				X		X		X	X
24-hour urine collection for PD analyses	X[r]	X[s]					X[s]			X[s]				X[s]		X[s]		X[s]		X[s]
[redacted] PD analysis[t]		X	X	X		X		X	X		X				X		X		X	X
Blood and urine samples for exploratory [redacted]			X					X			X[v]				X		X			
Blood sample for pyridoxine (vitamin B6) levels[w]			X					X			X				X		X		X	
Blood samples for ADA analysis			X					X			X				X				X	X[x]
Review/record AEs[y]			X																	
Prior and concomitant medications	X																			

Abbreviations: ADA=antidrug antibodies; AE=adverse event; BMI=body mass index; D=day; ECG=electrocardiogram; Echo=echocardiography; eGFR=estimated glomerular filtration rate; EOT=end of treatment; FSH=follicle-stimulating hormone; ICF=informed consent form; LFT=liver function test; PD=pharmacodynamics; PK=pharmacokinetic; Q=every; SC=subcutaneous.

Notes:

- When scheduled at the same time points, the assessments of vital signs and 12-lead ECGs must be performed first, before the physical examinations, blood sample collections, and urine collections.
- Assessments should be performed predose, where applicable, unless otherwise noted.

- Grey columns indicate the study day when the 24-hour urine collection for PD analysis should begin. Patients may be admitted to the clinical study center to complete the 24-hour urine collections.
  a. Patients will be invited to participate in an open-label extension study once they complete the postdose follow-up period on Study Day 141. For patients who do not enroll in the open-label extension study, safety and PD follow-up will continue until plasma glycolate is <20% above baseline or ≤ the ULN AND 24-hour urinary oxalate is >80% of baseline, or until the SRC makes a decision per investigator request to discontinue follow-up on a case-by-case basis. The decision cannot be made until after completion of the postdose follow-up period (84 days after last dose).
  b. Patients initially randomized to placebo may continue in the study receiving administration of open-label ALN-GO1 according to the Schedule of Assessments (Table 3). Patients will be unblinded on or after Day 78.
  c. Complete medical history (including documentation or confirmation of PH1) will be obtained at screening and any changes will be updated on Day -1. AEs occurring after signing of the ICF and before study drug administration will be captured as medical history.
  d. See Section 7.5.3 for assessments to be performed during a full physical examination or symptom-directed physical examination.
  e. During screening and at EOT, height will be measured for all patients; thereafter, height will only be measured for patients <18 years of age. Additionally, for patients <18 years of age, at each time point, height will be measured in centimeters, and in triplicate, to facilitate calculation of eGFR using the Schwartz Bedside Formula.
  f. Day -1 body weight will be used for calculation of the ALN–GO1 or placebo (study drug) dose to be administered during this portion of the study.
  g. Vital signs (blood pressure, pulse rate, body temperature, and respiration rate) will be measured in the supine position after the patient has rested comfortably for 10 minutes. On Day 1 only, vital signs will be measured within 2 hours predose; and 30 (±5 minutes) and 4 hours (±15 minutes) postdose.
  h. All 12-lead ECGs are triplicate, using centralized equipment. ECGs will be measured approximately 5 minutes apart. Recordings will be obtained after the patient has rested comfortably in the supine position for approximately 10 minutes. Patients should remain supine between ECGs. On dosing days, ECGs will be measured within 2 hours predose; and at 1 hour (±20 minutes), 2 hours (±20 minutes), and 4 hours (±20 minutes) postdose. On all other days, ECGs should be collected at approximately the same time of day corresponding to the predose collection (±1 hour).
  i. Echo will be performed during the first visit of the Safety and PD Follow-up Period (Day 169), and thereafter, approximately every 168 days corresponding with visits to the clinical study center for the duration of the study.
  j. Pregnancy tests will be performed for women of childbearing potential only. A serum pregnancy test will be performed at Screening or after the onset of menarche if the patient was not of childbearing potential at screening, and urine pregnancy tests will be performed thereafter per the Schedule of Assessments and any time pregnancy is suspected. The results of the pregnancy test must be known before study drug administration. FSH will be measured at Screening only to confirm post-menopausal status.
  k. Biochemistry, hematology, coagulation, LFTs, and urinalysis parameters are described in Section 7.5.6. Clinical laboratory tests will be analyzed by a central laboratory. Within 4 days prior to dosing, LFT measurements will be analyzed by a local laboratory and confirmed by a central laboratory. Local clinical laboratory results for LFT measurements must be available and reviewed by the Investigator before study drug administration.
  l. During Screening and throughout the Dosing Period, abnormal results for troponin I tests should be repeated. During the Dosing Period only, local clinical laboratory results must be drawn within 4 days prior to dosing and available and reviewed by the Investigator before study drug administration. Troponin I levels will be measured on the first day of the Safety and PD Follow-up Period (Day 169), and thereafter, approximately every 168 days for the duration of the study. Troponin I results from Day -1, Day 85, Day 169, and for the remainder of the study will be analyzed by a central laboratory. Central laboratory results are not required before study drug administration.
  m. Drugs of abuse are described in Section 7.5.6.3. Screening for drugs of abuse will be performed for patients who are above the age of legal consent.
  n. Randomization occurs between Day -1 and Day 1.
  o. Study drug will be administered via SC injection as described in Section 6.2.2.
  p. Blood and urine samples for PK analysis will be collected at the time points listed in Table 11.
  q. On Day 1, the blood sample for PD analysis must be collected within 2 hours predose. The remaining PD samples should be collected at approximately the same time of day corresponding to the predose collection (±1 hour), as applicable.
  r. During the screening period, 24-hour urine collections will be completed at 2 separate time points. The first screening 24-hour (±30 minutes) urine collection will be used to assess eligibility. The second screening 18-24-hour urine collection will only be initiated after eligibility is confirmed.

s. Single, 18-24-hour urine sample collections will be performed. The 24-hour urine collection starting on Day -1 must conclude on Day 1 before administration of the first dose of study drug. The 24-hour urine collection should be completed within 4 days prior to the study visit for Day 29, Day 57, Day 85, Day 113, and Day 141, and the Q28D Safety and PD Follow-up visits.
t. On Day -1, collect [redacted] sample before starting the 24-hour urine sample collection. On all other days, when a 24-hour urine collection is scheduled, collect [redacted] sample after the 24-hour urine collection, but before study drug administration, as applicable.
u. See the Laboratory Manual for instructions on sample processing and aliquoting of exploratory [redacted].
v. Blood samples for exploratory [redacted] are optional for pediatric patients who exceed the maximum blood volume collection limits listed in Table 13.
w. Blood sample for pyridoxine (vitamin B6) is required only for patients receiving therapeutic pyridoxine. On days when a blood sample for pyridoxine will be collected, patients should be instructed not to take vitamin B6 before the blood sample is collected and study drug is administered.
x. During the Safety and PD follow-up period, blood sample for ADA should be collected every 56 days after Day 141 (eg, Day 197, Day 253, Day 309) for the duration of the study.
y. AEs occurring after signing of the ICF and before study drug administration will be captured as medical history, while all AEs that occur after study drug administration, and baseline AEs that worsen after study drug administration, must be recorded and reported as AEs.

Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1 – Monthly Dosing

Study Stage		Dosing Period											Postdose Follow-up Period[a]						Safety and PD Follow-up
Study Day (D)	**84**	**85**	**86**	**87**	**99**	**112**	**113**	**127**	**140**	**141/EOT[b]**	**142**	**143**	**168**	**169**	**196**	**197**	**224**	**225**	**253; then, Q28D**
Visit Window (D)					±2		±4	±4		±4				±4		±4		±4	±4
Full physical examination[c]																		X	
Symptom-directed physical examination[c]		X	X		X		X	X		X				X		X			X
Height[d]		X								X				X				X	X
Body weight and BMI		X[e]								X				X				X	X
Vital signs[f]		X	X		X		X	X		X				X		X		X	X
12-lead ECG[g]		X	X				X			X				X		X		X	X
Echo		X												X					X[h]
Pregnancy test[i]		X					X			X				X		X		X	X
Clinical laboratory assessments[j]		X	X		X		X	X		X				X		X		X	X
Troponin I[k]		X					X			X				X					X
Study drug administration[l]		X					X			X									
Blood and urine samples for PK analyses[m]		X	X	X	X		X			X	X	X		X					
Blood sample for PD analyses[n]		X			X		X	X		X				X		X		X	X
24-hour urine collection for PD analyses[o]	X					X			X				X		X		X		X
[redacted] PD analysis[p]		X	X		X		X	X		X				X		X		X	X

Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1 – Monthly Dosing

Study Stage		Dosing Period											Postdose Follow-up Period[a]						Safety and PD Follow-up
Study Day (D)	**84**	**85**	**86**	**87**	**99**	**112**	**113**	**127**	**140**	**141/EOT[b]**	**142**	**143**	**168**	**169**	**196**	**197**	**224**	**225**	**253; then, Q28D**
Visit Window (D)					±2		±4	±4		±4				±4		±4		±4	±4
Blood and urine samples for exploratory [redacted]		X[r]					X			X				X		X			
Blood sample for pyridoxine (vitamin B6) levels[s]		X					X			X				X		X		X	
Blood samples for ADA analysis		X					X			X				X				X	X[t]
Review/record AEs	X																		
Concomitant medications	X																		

Abbreviations: ADA=antidrug antibodies; AE=adverse event; BMI=body mass index; D=day(s); ECG=electrocardiogram; Echo=echocardiography; eGFR=estimated glomerular filtration rate; EOT=end of treatment; PD=pharmacodynamics; PK=pharmacokinetic; Q=every; SC=subcutaneous.

Notes:

- When scheduled at the same time points, the assessments of vital signs and 12-lead ECGs must be performed first, before the physical examinations, blood sample collections, and urine collections.
- Assessments should be performed predose, where applicable, and unless otherwise noted.
- Grey columns indicate the study day when the 24-hour urine collection for PD analysis should begin. Patients may be admitted to the clinical study center to complete the 24-hour urine collections.

a. Patients will be invited to participate in an open-label extension study once they complete the postdose follow-up period on Study Day 225. For patients who do not enroll in the open-label extension study, safety and PD follow-up will continue until plasma glycolate is <20% above baseline or ≤ the ULN AND 24-hour urinary oxalate is >80% of baseline, or until the SRC makes a decision per investigator request to discontinue follow-up on a case-by-case basis. The decision cannot be made until after completion of the postdose follow-up (84 days after last dose).
b. If a patient chooses to withdraw consent, every effort should be made to conduct the assessments performed at the EOT visit.
c. See Section 7.5.3 for assessments to be performed during a full physical examination or symptom-directed physical examination.
d. On Day 85 and at EOT, height will be measured for all patients; thereafter, height will only be measured for patients <18 years of age. Additionally, for patients <18 years of age, at each time point, height will be measured in centimeters, and in triplicate, to facilitate calculation of eGFR using the Schwartz Bedside Formula.
e. The Day 57 body weight will be used for calculation of the ALN–GO1 dose to be administered during this portion of the study.
f. Vital signs (blood pressure, pulse rate, body temperature, and respiration rate) will be measured in the supine position after the patient has rested comfortably for 10 minutes. On Day 85 only, vital signs will be measured within 2 hours predose; and 30 (±5 minutes) and 4 hours (±15 minutes) postdose.

g. All 12-lead ECGs are triplicate, using centralized equipment. ECGs will be measured approximately 5 minutes apart. Recordings will be obtained after the patient has rested comfortably in the supine position for approximately 10 minutes. Patients should remain supine between ECGs. On dosing days, ECGs will be measured within 2 hours predose; and at 1 hour (±20 minutes), 2 hours (±20 minutes), and 4 hours (±20 minutes) postdose. On all other days, ECGs should be collected at approximately the same time of day corresponding to the predose collection (±1 hour).
h. Echo will be performed during the first visit of the Safety and PD Follow-up Period (Day 253), and thereafter, approximately every 168 days corresponding with visits to the clinical study center for the duration of the study.
i. Pregnancy tests will be performed for women of childbearing potential only. A serum pregnancy test will be performed after the onset of menarche if the patient was not of childbearing potential at screening, and a serum or urine pregnancy test will be performed per the Schedule of Assessments and any time pregnancy is suspected. The results of the pregnancy test must be known before ALN-GO1 administration.
j. Biochemistry, hematology, coagulation, LFTs, and urinalysis parameters are described in Section 7.5.6. Clinical laboratory tests will be analyzed by a central laboratory. Within 4 days prior to dosing, LFT measurements will be analyzed by a local laboratory and confirmed by a central laboratory. Local clinical laboratory results for LFT measurements must be available and reviewed by the Investigator before study drug administration.
k. Throughout the Dosing Period, local clinical laboratory results for troponin I must be drawn within 4 days prior to dosing and available and reviewed by the Investigator before study drug administration and abnormal results should be repeated. Troponin I levels will be measured on the first day of the Safety and PD Follow-up Period (Day 253), and thereafter, approximately every 168 days for the duration of the study. Troponin I results from Day 85, Day 169, and for the remainder of the study will be analyzed by a central laboratory.
l. ALN-GO1 will be administered via SC injection as described in Section 6.2.2.
m. Blood and urine samples for PK analysis will be collected at the time points listed in Table 11.
n. On Day 85, the blood sample for PD analysis must be collected within 2 hours predose. The remaining PD samples should be collected at approximately the same time of day corresponding to the predose collection (±1 hour), as applicable.
o. Single, 18-24-hour urine sample collections will be performed. On dosing days, the urine collection period should conclude in the morning before study drug administration. The 24-hour urine collection should be completed within 4 days prior to the study visits for Day 85, Day 113, Day 141, Day 169, and Day 197, Day 225, and the Q28D Safety and PD Follow-up visits.
p. On dosing days, when a 24-hour urine collection is scheduled, collect [redacted] sample after the 24-hour urine collection, but before study drug administration.
q. See the Laboratory Manual for instructions on sample processing and aliquoting of exploratory [redacted]
r. Blood samples for exploratory [redacted] are optional for pediatric patients who exceed the maximum blood volume collection limits listed in Table 13.
s. Blood sample for pyridoxine (vitamin B6) is required only for patients receiving therapeutic pyridoxine. On days when a blood sample for pyridoxine will be collected, patients should be instructed not to take vitamin B6 before the blood sample is collected and study drug is administered.
t. During the Safety and PD follow-up period, blood sample for ADA should be collected every 56 days after Day 225 (eg, Day 281, Day 337, Day 393) for the duration of the study.

Table 4: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B – Quarterly Dosing)

Study Stage	Screening	Dosing Period													Postdose Follow-up Period[a]								Safety and PD Follow-up
Study Day (D)	-45 to -2	-1	1	2	3	15	28	29	43	56	57	84	85/ EOT[b]	86	87	99	112	113	127	140	141	168	169	197; then, Q28D
Visit Window (D)						±2		±4	±4		±4		±4			±2		±4	±4		±4		±4	±4
Informed consent (and assent for patients under the age of legal consent)	X																							
Demography	X																							
Medical history[c]	X	X																						
Inclusion/ exclusion criteria	X	X																						
Full physical examination[d]	X																						X	
Symptom-directed physical examination[d]		X	X	X		X		X	X		X		X	X		X		X	X		X			X
Height[e]	X										X		X					X					X	X
Body weight and BMI	X	X[f]									X		X					X					X	X
Vital signs[g]	X	X	X	X		X		X	X		X		X	X		X		X	X		X		X	X

Table 4: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B – Quarterly Dosing)

Study Stage	Screening	Dosing Period													Postdose Follow-up Period[a]								Safety and PD Follow-up
Study Day (D)	-45 to -2	-1	1	2	3	15	28	29	43	56	57	84	85/ EOT[b]	86	87	99	112	113	127	140	141	168	169	197; then, Q28D
Visit Window (D)						±2		±4	±4		±4		±4			±2		±4	±4		±4		±4	±4
12-lead ECG[h]	X	X	X	X				X			X		X	X				X			X		X	X
Echo	X																	X						X[i]
Pregnancy test/FSH screening[j]	X	X	X					X			X		X					X			X			X
Clinical laboratory assessments[k]	X	X	X	X		X		X	X		X		X	X		X		X	X		X			X
Troponin I[l]	X	X											X					X						X
Urine sample for drugs of abuse[m]	X	X																						
Randomization			X[n]																					
Study drug administration[o]			X										X											
Blood and urine samples for PK analyses[p]			X	X	X	X							X	X	X	X		X						
Blood sample for PD analyses[q]	X		X			X		X	X		X		X			X		X	X		X		X	X

Table 4: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B – Quarterly Dosing)

Study Stage	Screening	Dosing Period														Postdose Follow-up Period[a]								Safety and PD Follow-up
Study Day (D)	**-45 to -2**	**-1**	**1**	**2**	**3**	**15**	**28**	**29**	**43**	**56**	**57**	**84**	**85/ EOT[b]**	**86**	**87**	**99**	**112**	**113**	**127**	**140**	**141**	**168**	**169**	**197; then, Q28D**
Visit Window (D)						±2		±4	±4		±4		±4			±2		±4	±4		±4		±4	±4
24-hour urine collection for PD analyses	X[r]	X[s]					X[s]			X[s]		X[s]					X[s]			X[s]		X[s]		X[s]
[redacted] PD analysis[t]		X	X	X		X		X	X		X		X			X		X	X		X		X	X
Blood and urine samples [redacted]			X					X			X		X[v]					X			X			
Blood sample for pyridoxine (vitamin B6) levels[w]			X					X			X		X					X			X		X	
Blood samples for ADA analysis			X					X			X		X					X			X		X	X[x]
Review/record AEs[y]			X																					
Concomitant medications																								

Abbreviations: ADA=antidrug antibodies; AE=adverse event; BMI=body mass index; D=day; ECG=electrocardiogram; Echo=echocardiography; eGFR=estimated glomerular filtration rate; EOT=end of treatment; FSH=follicle-stimulating hormone; ICF=informed consent form; LFT=liver function test; PD=pharmacodynamics; PK=pharmacokinetic; Q=every; SC=subcutaneous.

Notes:

- When scheduled at the same time points, the assessments of vital signs and 12-lead ECGs must be performed first, before the physical examinations, blood sample collections, and urine collections.
- Assessments should be performed predose, where applicable, unless otherwise noted.
- Grey columns indicate the study day when the 24-hour urine collection for PD analysis should begin. Patients may be admitted to the clinical study center to complete the 24-hour urine collections.
  a. Patients will be invited to participate in an open-label extension study once they complete the postdose follow-up period on Study Day 169. For patients who do not enroll in the open-label extension study, safety and PD follow up will continue until plasma glycolate is <20% above baseline or ≤ the ULN AND 24-hour urinary oxalate is >80% of baseline, or until the SRC makes a decision per investigator request to discontinue follow-up on a case-by-case basis. The decision cannot be made until after completion of the postdose follow-up period (84 days after last dose).
  b. Patients initially randomized to placebo will receive open-label ALN-GO1.
  c. Complete medical history (including documentation or confirmation of PH1) will be obtained at screening and any changes will be updated on Day -1. AEs occurring after signing of the ICF and before study drug administration will be captured as medical history.
  d. See Section 7.5.3 for assessments to be performed during a full physical examination or symptom-directed physical examination.
  e. During screening and at EOT, height will be measured for all patients; thereafter, height will only be measured for patients <18 years of age. Additionally, for patients <18 years of age, at each time point, height will be measured in centimeters, and in triplicate, to facilitate calculation of eGFR using the Schwartz Bedside Formula.
  f. Day -1 body weight will be used for calculation of the ALN–GO1 or placebo (study drug) dose to be administered during the dosing period.
  g. Vital signs (blood pressure, pulse rate, body temperature, and respiration rate) will be measured in the supine position after the patient has rested comfortably for 10 minutes. On Day 1 only, vital signs will be measured within 2 hours predose; and 30 (±5 minutes) and 4 hours (±15 minutes) postdose.
  h. All 12-lead ECGs are triplicate, using centralized equipment. ECGs will be measured approximately 5 minutes apart. Recordings will be obtained after the patient has rested comfortably in the supine position for approximately 10 minutes. Patients should remain supine between ECGs. On dosing days, ECGs will be measured within 2 hours predose; and at 1 hour (±20 minutes), 2 hours (±20 minutes), and 4 hours (±20 minutes) postdose. On all other days, ECGs should be collected at approximately the same time of day corresponding to the predose collection (±1 hour).
  i. Echo will be performed during the first visit of the Safety and PD Follow-up Period (Day 197) and approximately every 168 days corresponding with visits to the clinical study center for the duration of the study.
  j. Pregnancy tests will be performed for women of childbearing potential only. A serum pregnancy test will be performed at Screening or after the onset of menarche if the patient was not of childbearing potential at screening, and urine pregnancy tests will be performed thereafter per the Schedule of Assessments and any time pregnancy is suspected. The results of the pregnancy test must be known before study drug administration. FSH will be measured at Screening only to confirm post-menopausal status.
  k. Biochemistry, hematology, coagulation, LFTs, and urinalysis parameters are described in Section 7.5.6. Clinical laboratory tests will be analyzed by a central laboratory. Within 4 days prior to dosing, LFT measurements will be analyzed by a local laboratory and confirmed by a central laboratory. Local clinical laboratory results for LFT measurements must be available and reviewed by the Investigator before study drug administration.
  l. During Screening and throughout the Dosing Period, abnormal results for troponin I tests should be repeated. During the Dosing Period only, local clinical laboratory results must be drawn within 4 days prior to dosing and available and reviewed by the Investigator before study drug administration. Troponin I levels will be measured on the first day of the Safety and PD Follow-up Period (Day 197), and thereafter, approximately every 168 days for the duration of the study. Troponin I results from Day -1, Day 85, Day 113, Day 197, and for the remainder of the study will be analyzed by a central laboratory. Central laboratory results are not required before study drug administration.
  m. Drugs of abuse are described in Section 7.5.6.3. Screening for drugs of abuse will be performed for patients who are above the age of legal consent.
  n. Randomization occurs between Day -1 and Day 1.
  o. Study drug will be administered via SC injection as described in Section 6.2.2.

p. Blood and urine samples for PK analysis will be collected at the time points listed in Table 12.
q. On Day 1, the blood sample for PD analysis must be collected within 2 hours predose. The remaining PD samples should be collected at approximately the same time of day corresponding to the predose collection (±1 hour), as applicable.
r. During the screening period, 24-hour urine collections will be completed at 2 separate time points. The first screening 24-hour (±30 minutes) urine collection will be used to assess eligibility. The second screening 18-24-hour urine collection will only be initiated after eligibility is confirmed.
s. Single, 18-24-hour urine sample collections will be performed. The 24-hour urine collection starting on Day -1 must conclude on Day 1 before administration of the first dose of study drug. The 24-hour urine collection should be completed within 4 days prior to the study visit for Day 29, Day 57, Day 85, Day 113, Day 141, and Day 169, and the Q28D Safety and PD Follow-up visits.
t. On Day -1, collect [redacted] sample before starting the 24-hour urine sample collection. On all other days, when a 24-hour urine collection is scheduled, collect [redacted] sample after the 24-hour urine collection, but before study drug administration, as applicable.
u. See the Laboratory Manual for instructions on sample processing and aliquoting [redacted].
v. Blood samples [redacted] on Day 85 are optional for pediatric patients who exceed the maximum blood volume collection limits listed in Table 13.
w. Blood sample for pyridoxine (vitamin B6) is required only for patients receiving therapeutic pyridoxine. On days when a blood sample for pyridoxine will be collected, patients should be instructed not to take vitamin B6 before the blood sample is collected and study drug is administered.
x. During the Safety and PD follow-up period, blood sample for ADA should be collected every 56 days after Day 169 (eg, Day 225, Day 281, Day 337) for the duration of the study.
y. AEs occurring after signing of the ICF and before study drug administration will be captured as medical history, while all AEs that occur after study drug administration, and baseline AEs that worsen after study drug administration, must be recorded and reported as AEs.

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LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation or Specialist Term	Explanation
AE	adverse event
ALP	alkaline phosphatase
ALT	alanine transaminase
AST	aspartate aminotransferase
BMI	body mass index
BUN	blood urea nitrogen
CRO	contract research organization
CTCAE	Common Terminology Criteria for Adverse Events
ECG	electrocardiogram
Echo	echocardiography
eCRF	electronic case report form
eGFR	estimated glomerular filtration rate
EOT	End of Treatment
FSH	follicle-stimulating hormone
GalNAc	N-acetylgalactosamine
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
GO	glycolate oxidase
HIV	human immunodeficiency virus
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IRB	Institutional Review Board
ISR	injection site reaction
MAD	multiple-ascending dose
MDRD	modification of diet in renal disease
MedDRA	Medical Dictionary for Regulatory Activities
NHP	non-human primates
OTC	over the counter
PD	pharmacodynamic
PK	pharmacokinetics
QTcB	Bazett-corrected QT interval

Abbreviation or Specialist Term	Explanation
QTcF	Fridericia corrected QT interval
SAD	single-ascending dose
SAE	serious adverse event
SAP	statistical analysis plan
SC	Subcutaneous
siRNA	small interfering RNA
SOC	System Organ Class
SRC	Safety Review Committee
SUSARs	suspected unexpected serious adverse reactions
ULN	upper limit of normal
WHO	World Health Organization
WOCBP	women of child bearing potential

1. INTRODUCTION

1.1. Disease Overview

Alnylam Pharmaceuticals is developing ALN-GO1, a synthetic, small interfering RNA (siRNA) therapeutic directed against hydroxyacid oxidase 1 (HAO1) messenger RNA (mRNA), which is covalently linked to a ligand containing 3, N-acetylgalactosamine residues. The proposed indication for ALN-GO1 is the treatment of Primary Hyperoxaluria Type 1 (PH1).

PH1 is a rare, autosomal recessively inherited disease characterized by excessive production of oxalate and consequent hyperoxaluria. Given the relative insolubility of oxalate, it crystallizes in the urinary tract, primarily as calcium oxalate. This results in recurrent nephrolithiasis and/or nephrocalcinosis, with progressive renal disease leading to renal failure.[1] As renal function declines, calcium oxalate is deposited systemically, with consequent end organ damage. This stage of the disease, called systemic oxalosis, arises when the glomerular filtration rate (GFR) has declined to below 30 to 45 mL/min per 1.73 m^2.[1]

PH1 is caused by mutations in both alleles of the alanine glyoxylate aminotransferase (AGXT) gene, which encodes the liver peroxisomal enzyme alanine-glyoxylate aminotransferase (AGT). Over 150 mutations in AGXT have been described. There are broad genotype–phenotype associations, notably in the responsiveness of disease caused by some mutations, to treatment with pyridoxine (vitamin B6). However, disease phenotype can be highly variable, including the responsiveness to vitamin B6 treatment, even within families.[2]

Given the rarity and heterogeneity of PH1, many patients go undiagnosed for years after the initial clinical manifestations of the disease. The incidence of PH1 is approximately 1 in 120,000 live births, and the prevalence is 1 to 3 per million in North America and Europe.[1-3] The disease is more prevalent in areas where consanguineous marriages are common, especially in the Middle East and Northern Africa.[4, 5]

PH1 is primarily a pediatric disease, with symptoms first appearing in approximately half (48.6%) of the patients in the Rare Kidney Stone Consortium Primary Hyperoxaluria Registry between birth and four years of age. Approximately three-quarters of patients (74.7%) showed signs or symptoms before the age of 15 and 83.5% showed symptoms before 20 years of age. In comparison, only 16.5% of patients first displayed symptoms after age 20. Additionally, nearly one third (29.9%) of patients were diagnosed between birth and four years of age and two-thirds (66%) were diagnosed before 20 years of age. [Rare Kidney Stone Consortium Primary Hyperoxaluria. Available from: http://rarekidneystones.org/hyperoxaluria/, accessed on 25 September 2015] The majority of patients are, therefore, diagnosed as children, and consequently, are more likely to have some preservation of renal function at time of diagnosis.

Most PH1 patients exhibit urinary oxalate excretion greater than 2-fold the upper limit of normal (ULN), although patients with severely compromised renal function may have lower excretion rates. PH1 is definitively diagnosed by gene sequencing to detect pathological mutations in AGXT, or by evaluation of AGT enzymatic activity in liver tissue obtained by biopsy.

Currently, there are no approved therapies for the treatment of PH1. Disease management is based on supportive measures, including high fluid intake, potassium citrate (to increase urinary oxalate solubility), Vitamin B6, and treatment of complications such as urinary tract stones and infections. Dietary modification plays a minor role in treatment since endogenous oxalate

production far exceeds dietary intake. Patients progressing to, or presenting with end-stage renal disease (ESRD) require intense kidney dialysis. However, dialysis, day and night, 6 days per week, may be inadequate to effectively offload accumulating oxalate.[6] Combined liver/kidney transplantation offers potentially curative therapy, but with limited availability, the attendant medical risks, and intense use of health care resources. Therefore, there is a high unmet medical need for additional treatments for patients with PH1. The deterioration of renal function that occurs in PH1 disease indicates the importance of investigating potentially disease-modifying interventions as early as possible after diagnosis.

1.2. ALN-GO1

1.2.1. ALN-GO1 Description

ALN-GO1 (containing siRNA drug substance ALN-65585) is an investigational medicinal product that comprises a synthetic siRNA that specifically targets the mRNA of the HAO1 gene which encodes glycolate oxidase (GO).

1.2.2. Rationale for ALN-GO1 for the Treatment of Primary Hyperoxaluria Type 1

GO, a hepatic enzyme upstream of AGT, mediates the oxidation of glycolate to glyoxylate, which then is further metabolized to oxalate. Suppression of GO by ALN-GO1 is expected to reduce the production of oxalate, while increasing the generation of glycolate. Unlike oxalate, glycolate is highly soluble and readily excreted in the urine. The effective substitution of glycolate for oxalate production is hypothesized to significantly ameliorate the course of PH1 disease. Moreover, the mechanism of action of ALN-GO1 suggests that plasma glycolate could be a biomarker for the inhibition of GO1, serving as a pharmacodynamic (PD) measure of ALN-GO1 activity in healthy subjects.

1.2.3. Nonclinical Data

The pharmacology, safety pharmacology, drug metabolism and pharmacokinetics (PK), and toxicology of ALN-GO1 were evaluated in a series of in vitro and in vivo nonclinical studies.

Nonclinical pharmacology studies include the assessment of in vitro activity of ALN-GO1 in primary hepatocytes from non-human primates (NHP); and in vivo in wild-type (WT) mice deficient in AGXT (a genetic model of PH1), WT rats, rats with inhibited AGXT activity (an induced model of PH1), and WT NHP. When subcutaneously administered, ALN-GO1 demonstrated potent, dose-dependent pharmacologic activity, resulting in reduced liver HAO1 mRNA levels with the expected increases in plasma glycolate levels in WT and diseased animals as well as subsequent reductions in urinary oxalate in diseased animals. Modest increases (approximately 1.5-fold) in plasma glycolate levels were observed in NHP treated with 1 mg/kg of ALN-GO1 subcutaneously administered every 4 weeks, the lowest dose studied with a 4-week dosing interval.

Good Laboratory Practice (GLP) -compliant toxicology studies were performed in rats at 0, 5, 15, or 50 mg/kg weekly or 50 mg/kg monthly and in NHP at 0, 10, 30, and 100 mg/kg weekly or 100 mg/kg monthly, with treatment administered for 8 weeks. The no observed adverse effect levels (NOAEL) were 50 mg/kg in rats and 100 mg/kg in NHP (Table 5).

Genetic toxicity studies (bacterial reverse mutation, human peripheral blood lymphocyte chromosomal aberrations, and rat bone marrow micronucleus assays) were all negative based on industry guidelines.[7]

In summary, nonclinical studies have shown pharmacological activity in NHP at doses as low as 1.0 mg/kg, while doses as high as 50 mg/kg and 100 mg/kg in rat and NHP, respectively, have resulted in no adverse effects after 8 weeks of treatment. These data provide the justification for the starting dose for administration in this clinical study.

1.3. Study Design Rationale

This is a randomized, single-blind, placebo-controlled, multi-center Phase 1/2 single-ascending dose (SAD) and multiple-ascending dose (MAD) study designed to evaluate the safety, tolerability, PK, and PD of subcutaneously administered ALN-GO1 in healthy adult subjects and in adult and pediatric patients with PH1. The primary objective of the study is to evaluate the safety and tolerability of single-ascending doses of ALN-GO1 in healthy adult subjects and multiple-ascending doses of ALN-GO1 in patients with PH1. Secondary and exploratory objectives of the study include the characterization of plasma and urine PK for ALN-GO1 and the evaluation of the PD effect of ALN-GO1, including glycolate and oxalate concentration and excretion.

Part B of this study will be implemented as a delayed start study; all patients will receive active study drug. Patients initially randomized to receive placebo will transition to active study drug; thus, additional safety data will be collected from patients transitioning from placebo to active study drug. The design allows for evaluation of safety and PD data at multiple dose levels in healthy subjects before initiating dosing in patients with PH1. This will permit more efficient dose selection in patients, given the rarity of the disease, and limits exposure of patients to ALN-GO1 as safety is being assessed.

1.4. Dose Rationale

The proposed starting dose in healthy subjects in Part A of this study is 0.3 mg/kg. Data in NHP suggest that this dose will have a low level of pharmacologic activity, while NOAELs determined in rat and NHP 8-week GLP toxicity studies indicate a wide margin of safety. While it is expected that nonclinical PD and toxicity will translate to humans on a body weight (mg/kg) basis, a more conservative approach for determining the starting dose was calculated by converting the NOAEL in the rat and NHP to a human equivalent dose (HED) using body surface area (BSA; mg/m^2). Using BSA conversions, safety margins for the clinical starting dose of 0.3 mg/kg were 26.7-fold and 106.7-fold in the rat and NHP, respectively, whether study drug was administered weekly or every 4 weeks. Collectively, the results of the ALN-GO1 nonclinical and pharmacological studies support the starting dose of 0.3 mg/kg in this first-in-human study. The starting dose in patients will be the lowest dose determined to have a pharmacological effect in healthy subjects that was also considered safe and well-tolerated.

Table 5: No Observed Adverse Effect Level and Starting Dose Safety Margins

	NOAEL (mg/kg)	HED based on BSA (mg/kg)	Safety margin for proposed starting dose of 0.3 mg/kg (Part A)	
			Based on mg/kg	Based on BSA[a]
Rat (QW×9)	50	8	167-fold	26.7
Rat (Q4W×3)	50	8	167-fold	26.7
NHP (QW×9)	100	32	333-fold	106.7
NHP (Q4W×3)	100	32	333-fold	106.7

Abbreviations: BSA=body surface area; HED=human equivalent dose; NHP=nonhuman primates; NOAEL=no observed adverse effect level; Q=every; W=week.

[a] BSA margin calculated using species-scaled conversion assumptions.[8]

Doses were selected for this study in accordance with Committee for Medicinal Products for Human Use (CHMP) guidelines on Risk Identification and Mitigation in first-in-human clinical studies (EMEA/CHMP/SWP/28367/07).

Preliminary data from Part A suggest that ALN-GO1 is well-tolerated and support selection of the starting dose for the first cohort of Part B of study ALN-GO1-001. The Part B starting dose is 1.0 mg/kg, administered every 28 days, the lowest dose determined to have a pharmacological effect in healthy subjects in Part A that was also considered well-tolerated. The starting dose in Part B is based on data derived from Part A and recommended by the Safety Review Committee (SRC). Additional cohorts may be enrolled at higher, lower, or intermediate dose levels, but will not exceed the maximum administered dose of 6 mg/kg, and will follow the protocol-specified dose escalation criteria.

1.5. Benefit-Risk Assessment

ALN-GO1 is designed to reduce hepatic production of oxalate. The potential benefit of this treatment is the amelioration of the clinical course of PH1 in patients across the spectrum of disease, irrespective of age and disease stage; however, patients with PH1 in this study may not receive treatment for a sufficient duration, or at an adequate dose, to experience clinical benefit. There is no benefit for healthy subjects participating in this clinical study of ALN-GO1. The potential risks of ALN-GO1 include pathway- and disease-specific risks, and non-specific, off-target risks.

Reduction of GO is expected to lead to reduction in hepatic oxalate production at the expense of increased glycolate, an organic acid. Therefore, study drug-induced increases in plasma and urine glycolate levels are anticipated in both healthy subjects and patients administered ALN-GO1. Since elevated levels of this organic acid are expected to be readily buffered, and its high solubility is not expected to result in crystallization in the urinary tract, the potential risk of increased glycolate production is considered low. Importantly, no toxicity has been observed in NHP pharmacology and toxicology studies, where profound suppression of hepatic GO, with associated increases in plasma glycolate levels, has been demonstrated. In particular,

maintenance of normal serum bicarbonate levels in these animals indicates that there is no evidence of acidosis. Finally, a recent clinical case study described a child with an incidentally discovered homozygous defect in HAO1, the gene encoding GO. The patient exhibited marked elevations of urine glycolate, but no associated metabolic abnormalities and normal renal and hepatic function.[9]

The potential negative consequences of ALN-GO1 administration to patients with impaired renal function, including those with end stage renal disease (ESRD), are considered low. ALN-GO1, in common with other Sponsor-developed RNAi compounds, is conjugated to N-acetyl galactosamine (GalNAc) to enable rapid and specific uptake by hepatocytes via the asialoglycoprotein receptor. Clinically effective doses of ALN-GO1 are not expected to saturate this receptor-mediated uptake or to result in enhanced extra-hepatic uptake or significant extra-hepatic exposure via accumulation; however, only patients with relatively well-preserved renal function are eligible to participate in this study.

PH1 is primarily a pediatric disease, with predominantly early childhood expression of symptoms, an early age at diagnosis, and a decline in renal function with increasing age in childhood through adulthood. [Rare Kidney Stone Consortium Primary Hyperoxaluria. Available from: http://rarekidneystones.org/hyperoxaluria/, accessed on 25 September 2015] The consequence is that early treatment is required to gain the greatest potential benefit and prevent loss of renal function as these patients age. Systemic oxalosis, resulting from over production combined with insufficient dialytic clearance of oxalate, is the most devastating complication of PH1 and is not prevented or effectively treated by dialysis. Liver/kidney transplantation is considered curative, but has high cost, high morbidity, and relatively low availability. Children with relatively well-preserved renal function are more prevalent than adults and are the primary target population for development of ALN-GO1. For this reason, children are included in the population for evaluation in this study.

This study is designed such that safety data in healthy adult subjects will be evaluated at dose levels higher than proposed for the starting dose in patients, including those as young as 6 years of age. Since the intended lowest dose for administration in patients will have been demonstrated to result in increased plasma glycolate concentration, a marker of the PD effect of ALN-GO1, and adequate safety in healthy subjects, the initial starting dose in patients is expected to be a potentially active dose of ALN-GO1.

Although the study includes a placebo control, all patients initially assigned to placebo will be administered active treatment via a delayed start study design. Study assessments and visits have been reduced as much as possible to minimize the burden of the study on children without risking patient safety. The potential benefit to children enrolled in this study includes possible reduction in oxalate production during the study period, which may have a temporarily ameliorating effect on their disease. In addition, experience in children with this disease under carefully controlled conditions will provide data that may enhance the future development of this therapeutic.

Important potential risks to healthy subjects and patients include injection site reactions (ISRs). Other potential risks include: embryofetal toxicity, coagulation abnormalities, and liver function test abnormalities. These potential risks have been reduced by the specific inclusion and exclusion criteria incorporated into the selection of healthy subjects as well as for patients with PH1 in this clinical study.

No data are available on the use of ALN-GO1 in pregnancy; however, human mutagenicity is not suggested based on the available nonclinical data (see Investigator's Brochure for further information). Embryofetal risk is limited by requiring that women of childbearing potential (WOCBP) must have a negative pregnancy test, cannot be breast feeding, and must be willing to use a highly effective method of contraception as specified in the protocol. Male subjects/patients are not required to use the contraception measures required for female study subjects/patients. No male contraception is considered to be required.

The occurrence of ISRs will be carefully monitored.

Nonclinical studies in rats showed mild to moderate decreases in fibrinogen and occasionally minimal prolonged prothrombin time without accompanying clinical or microscopic evidence of hemorrhage. This effect is likely species-specific as it was not observed in NHPs and will be monitored via clinical laboratory safety assessments.

As ALN-GO1 is a hepatically-targeted therapeutic, liver function tests will be carefully monitored. Nonclinical data suggest that there is a wide margin between the proposed clinical doses and any hepatic findings in toxicology studies and that any findings can be monitored. The potential non-specific, off-target risk of ALN-GO1 administration is considered to be low, based on nonclinical toxicological assessment of this molecule and accumulating experience with this RNA interference (RNAi) therapeutics platform.

During the course of this study, safety will be monitored by study Investigators, the Sponsor Medical Monitor, and a SRC. The initial dose level for administration in the MAD part of the study will have been previously shown to be safe, tolerable, and pharmacologically active based on SRC review of safety and PD data from the SAD part of the study. Stopping rules have also been designed to protect study participants.

Overall, in this clinical study of ALN-GO1, the risk to healthy subjects is considered low and the benefit/risk assessment is favorable in patients with PH1. Moreover, since evaluation of safety and PD effects in children will be important in the design of future studies, it is considered important and appropriate to enroll children in the current study.

The complete summary of the clinical and nonclinical data relevant to the investigational drug and its study in human subjects is provided in the Investigator's Brochure.

2. OBJECTIVES

2.1. Primary Objective

- Evaluate the safety and tolerability of single- and multiple-ascending doses of ALN-GO1, respectively, in healthy adult subjects and in patients with PH1

2.2. Secondary Objectives

- Characterize the PK of ALN-GO1
- Evaluate the PD of ALN-GO1

2.3. Exploratory Objectives

- [redacted] exploratory [redacted]
- [redacted]

3. ENDPOINTS

3.1. Primary Endpoint

- The primary endpoint is the incidence of adverse events (AEs). Safety will also be evaluated through vital signs, electrocardiograms (ECGs), clinical laboratory assessments, and physical examinations.

3.2. Secondary Endpoints

3.2.1. Secondary Endpoints for the Single-ascending Dose Part in Healthy Adult Subjects (Part A)

- PK parameters including, but not limited to, maximum plasma concentration [C_{max}], time to reach maximum plasma concentration [t_{max}], area under the plasma concentration versus time curve [AUC], apparent terminal elimination half-life [$t_{½}$], fraction eliminated in urine [f_e/F], and renal clearance [CL_R]
- Plasma and urine glycolate concentration

3.2.2. Secondary Endpoints for the Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

- PK parameters including, but not limited to, C_{max}, t_{max}, AUC, $t_{½}$, f_e/F, and CL_R
- Urinary oxalate excretion (oxalate concentration in 24-hour urine collection)
- Urinary glycolate excretion (glycolate concentration in 24-hour urine collection)
- Plasma glycolate concentration
- Calculated creatinine clearance

3.3. Exploratory Endpoints

- Plasma oxalate concentration
- [redacted]

3.3.1. Exploratory Endpoints for the Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

- [redacted]
- [redacted]
- [redacted]

4. INVESTIGATIONAL PLAN

4.1. Summary of Study Design

This is a randomized, single-blind, placebo-controlled study of subcutaneously administered ALN-GO1. The study is designed to evaluate the safety, tolerability, PK, and PD of single- and multiple-ascending doses of ALN-GO1 and will be conducted in 2 parts:

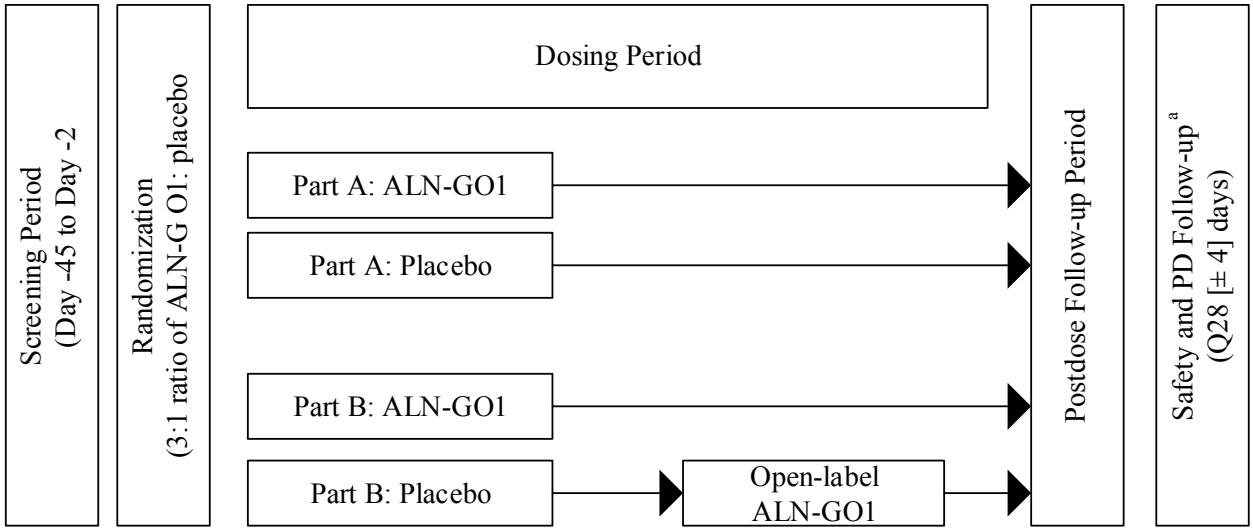
- Part A: single-ascending dose (SAD) part in healthy adult subjects
- Part B: multiple-ascending dose (MAD) part in adult and pediatric patients with PH1

The study will be conducted in a single-blind manner, with the Investigators, SRC, and Sponsor unblinded to permit ongoing unblinded review of safety, tolerability, PK, and PD data.

Part A will be conducted at 1 clinical study center in the UK. Part B is expected to take place at approximately 12 clinical study centers worldwide.

The study design is summarized in Figure 1.

Figure 1: Study Design



[a] Patients in Part B will be invited to participate in an open-label extension study once they complete the postdose follow-up period. For patients who do not enter the open-label extension study, safety and PD follow-up will continue until they meet the criteria described in Section 4.1.2, or until the SRC makes a decision per investigator request to discontinue follow-up on a case-by-case basis. The decision cannot be made until after completion of the last postdose follow-up visit (84 days after last dose).

4.1.1. Single-ascending Dose Part in Healthy Adult Subjects (Part A)

Part A is the single-ascending dose part of the study in healthy adult subjects. Subjects will be enrolled in 1 of 3 ascending dose cohorts, with the possibility for 2 additional optional cohorts. Each cohort will be comprised of 8 subjects randomized 3:1 to ALN-GO1 or placebo (study drug).

Subjects will be screened from -45 to -2 days before study drug administration. Subjects will be admitted to the clinical study center on Day -1 to determine continued eligibility and for predose assessments. Subjects in each cohort will be randomized on Day 1 and will receive 1 SC dose of

study drug. Subjects will be discharged from the clinical study center on Day 2 after completing the 24-hour postdose follow-up assessments.

Subjects will return to the clinical study center on an outpatient basis for safety, tolerability, PK, and PD monitoring at time points specified in the Schedule of Assessments through the last postdose follow-up visit (Day 57). Safety and PD follow-up will continue: 1) for at least 57 days, and 2) until plasma glycolate decreases to a level that is no more than 20% above of baseline or until plasma glycolate is below the upper limit of normal (≤14 nmol/mL).

4.1.2. Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

Part B is the multiple-ascending dose part of the study in up to 24 adult and pediatric patients with PH1 with relatively well-preserved renal function. Two ascending dose cohorts will be enrolled, with the possibility to also enroll up to 3 additional optional cohorts to further explore the optimal dose or regimen. Each cohort will be comprised of 4 patients, randomized 3:1 to ALN-GO1 or placebo. Up to 2 cohorts in Part B may be expanded by up to 4 additional patients per cohort (these patients will all receive ALN-GO1, not placebo).

Patients will be screened within 45 days prior to study drug administration. Baseline urinary oxalate excretion and creatinine clearance will be assessed through 24-hour urine collections. Patients will be randomized between Day -1 and Day 1 and will receive the first dose of ALN-GO1 or placebo on Day 1. The 24- and 48-hour postdose follow up assessments will take place on Day 2 and Day 3. Patients who receive study drug monthly will return to the clinical study center for safety, tolerability, PK, and PD monitoring at time points specified in the Schedule of Assessments (Table 2) for the remaining 2 single-blind doses of study drug (through Day 57). After completion of the blinded portion of the study, patients dosed monthly will be unblinded (on or after Day 78). Patients who initially received placebo will then receive ALN-GO1 at the same dose administered to the cohort into which they were initially randomized and will follow the same assessment schedule as indicated in Table 3.

Patients who receive study drug quarterly will return to the clinical study center for safety, tolerability, PK, and PD monitoring at time points specified in the Schedule of Assessments (Table 4) and will receive a 2nd dose of study drug at Day 85. Patients who initially received placebo will receive a single dose of ALN-GO1 on Day 85 at the same dose administered to the cohort into which they were initially randomized. Patients dosed quarterly will be unblinded to initial treatment assignment following completion of the postdose follow-up period.

After the dosing period, patients will return to the clinical study center for continued safety, tolerability, PK, and PD monitoring for at least 12 weeks (84 days) following the last dose of study drug. Following completion of the 12-week postdose follow-up period, patients will be invited to participate in an open-label extension study.

For patients who do not enroll in the open-label extension study, safety and PD follow up will continue until 24-hour urinary oxalate is >80% of baseline, and plasma glycolate is <20% above baseline or ≤ the ULN. If an investigator wishes to discontinue follow up after completion of the postdose follow-up period and prior to oxalate and glycolate recovery, the SRC must agree based upon consideration of emerging data on the safety of ALN-GO1 knockdown and the individual patient's safety and PD data.

4.2. Duration of Treatment and Overall Duration of Study

The duration of treatment is as follows:

- Part A: The estimated total time on study, inclusive of screening, for each subject is up to 405 days. The duration of treatment is a single dose.
- Part B: For all patients, the duration of screening is up to 45 days and the minimum duration of postdose follow-up is 84 days. The duration of treatment and estimated total time on study, inclusive of screening, for each patient is as follows:
  - For patients dosed monthly: The duration of treatment for patients initially randomized to receive active study drug is 57 days. The estimated total time on study is up to 462 days. Additionally, the duration of treatment for patients initially randomized to receive placebo is 141 days. The estimated total time on study for each patient initially randomized to receive placebo, then active study drug, is up to 546 days.
  - For patients dosed quarterly: The duration of treatment is 85 days for patients randomized to placebo and active study drug. The estimated total time on study is up to 490 days.

The overall duration of the study is estimated to be 4 years, including enrollment.

4.3. Number of Subjects and Patients

A total of up to 64 participants (including optional and expansion cohorts) are planned to be enrolled in this study as follows:

- Part A: Up to 40 healthy adult subjects
- Part B: Up to 24 adult and pediatric patients with PH1

4.4. Method of Assigning Patients to Treatment Groups

This is a randomized, single-blind, placebo-controlled study. In Parts A and B, after confirmation of eligibility, during screening, and upon admission to the clinical study center, subjects/patients will be assigned to a dose level cohort and randomized in a 3:1 ratio (ALN-GO1:placebo). No subject/patient will be a member of more than 1 cohort. A unique subject/patient identification number, incorporating the clinical study center number, will be assigned sequentially to the subject/patient.

The clinical study center pharmacy staff will randomize the subject/patient in accordance to a cohort-specific randomization list generated by the biostatistician at the Contract Research Organization (CRO), for appropriate dispensation of the study drug.

The information furnished to the Sponsor for subject/patient identification will be the assigned subject/patient identification number, the randomization number of subjects/patients admitted to the study, the age, and sex. The subject's/patient's identification number will appear on all documents relating to that subject/patient and will be cross-referenced by the randomization number for the enrolled subjects/patients. The Sponsor may receive a copy of the randomization list.

4.5. Blinding

This is a single-blind, placebo-controlled study; therefore, only the study subjects/patients will be blinded to treatment assignment. Patients in Part B dosed monthly will be unblinded on or after Day 78 in order for patients and their families to be better prepared for the transition to receive ALN-GO1 if initially randomized to placebo. Patients dosed quarterly will be unblinded to initial treatment assignment following completion of the postdose follow-up period. The Investigators, Medical Monitors at the Sponsor and CRO, clinical study center personnel, pharmacokineticist, and members of the SRC will have knowledge of the treatment assignment. The clinical study center pharmacy staff will maintain the single-blind according to clinical study center-specific procedures and the Pharmacy Manual. Syringes containing dispensed study drug will be masked in the pharmacy before transfer to the clinic.

During the blinded period, if the subject/patient becomes seriously ill during the study, and the treating physician determines that the clinical management of the subject requires that the subject know the study drug assignment, the Investigator may break the blind, as necessary. If time permits, clinical study center personnel should contact the Medical Monitor before unblinding to discuss the need to unblind the subject/patient. A record of when the blind was broken, who broke the blind, and why it was broken, will be maintained in the Trial Master File.

See the Pharmacy Manual for additional details.

4.6. Safety Review Committee

A SRC will perform ongoing reviews of safety, tolerability, and available study data collected in all study parts (Parts A and B) with the primary purpose of protecting the safety of subjects/patients participating in this clinical study. The SRC will undertake safety data review before initiation of dosing in a new cohort in Part A and Part B, and before initiation of dosing in Part B. After initiation of dosing in Part B, safety reviews will be conducted in accordance with the SRC charter at a minimum of every 3 months for the duration of the study. Part A and Part B of the study will incorporate adaptive study design features, which will allow for the continuing modification of dosing options and/or regimen as new data emerge, and in accordance with recommendations of the SRC (Table 9). Protocol-defined stopping rules will be used as criteria for stopping cohorts (Table 6) or individual patients (Table 7). Additionally, the SRC may recommend discontinuation of the study at its discretion. The investigator may also request that the SRC determine on a case-by-case basis whether study follow-up will be discontinued in patients in Part B who do not enroll in the open-label extension study who have completed the postdose-follow up period, but have not yet met the recovery criteria for plasma glycolate and urinary oxalate specified in Section 4.1.2. The decision will be informed by emerging data on the safety of ALN-GO1 knockdown and the individual patient's safety and PD data.

The SRC will be comprised of the Sponsor Medical Monitor, a Medical Monitor from the CRO, the Principal Investigator from the clinical study center conducting Part A of the study, an independent pediatrician experienced in clinical investigation who is not a study Investigator, and 3 Investigators at the clinical study sites who will be selected by the Sponsor.

The SRC will be governed by a charter that will be signed prior to enrollment of the first subject.

4.7. Study Drug Dosing, Study Progression, and Dose Escalation

For the purpose of this study, progression rules are based on toxicity. The Common Terminology Criteria for Adverse Events (CTCAE; Version 4.0, or higher) will be used to grade AEs. The decision to enroll an optional cohort or extend an existing cohort will be made by the SRC based on available safety, tolerability, and PD data, if further elucidation of dose response is considered necessary to better understand dose response and/or safety and tolerability.

4.7.1. Study Drug Dosing, Study Progression, and Dose Escalation in Part A

The following are the planned dose levels for Part A; however, the actual dose administered may be modified based on SRC review of emerging safety, tolerability, and available PD data in previous cohorts. The initial starting dose level is expected to have an adequate safety margin and a low level of pharmacologic activity based on GLP toxicology study results and nonclinical pharmacology.

- Cohort 1: 0.3 mg/kg
- Cohort 2: 1.0 mg/kg
- Cohort 3: 3.0 mg/kg
- Cohort (optional)
- Cohort (optional)

The SRC will review a minimum of 48 hours of safety data from at least 6 subjects before escalation to the next dose level. The SRC will review AEs and clinical laboratory evaluation data in order to proceed to the next cohort.

Based on SRC review of accumulated safety and PD data, 2 additional optional subject cohorts may be enrolled and dosed according to the same eligibility criteria and randomization scheme as Cohorts 1 to 3. The additional cohorts may be enrolled at higher, lower, or intermediate dose levels, but will not exceed the maximum administered dose of 6.0 mg/kg.

4.7.2. Study Drug Dosing, Study Progression, and Dose Escalation in Part B

Preliminary safety and PD data from Part A was evaluated by the SRC. The initial dose level of 1.0 mg/kg for administration in Part B was shown to be well-tolerated, and the lowest pharmacologically active dose. In Part B, all patients in a cohort dosed monthly will receive the same dose for each of the 3 study drug doses. Patients initially receiving 3 doses of placebo will receive 3 doses of ALN-GO1, with all 3 doses being the same for each of the 3 administrations of ALN-GO1. Patients in a cohort dosed quarterly who are randomized to ALN-GO1 will receive the same dose for each of the 2 study drug doses. Patients initially receiving 1 dose of placebo on Day 1 will receive 1 dose of ALN-GO1 on study Day 85. The maximum dose administered will not exceed 6.0 mg/kg.

Dose levels and/or dosing regimen in each cohort in Part B may be modified by the SRC. The next higher dose cohort can be enrolled after at least 3 patients in the previous cohort receive their first and second dose and have been followed for at least 14 days following the second dose of study drug. The SRC must review accumulating safety data from both single- and multiple-ascending dose cohorts to confirm the dose level and permit dosing in the next MAD cohort. In

Part B, patients randomized to placebo will be administered active ALN-GO1 in an open-label manner according to the same dose and assessment schedule for the cohort to which they were initially assigned.

Based on SRC review of accumulated safety, tolerability, and PD data, 3 additional cohorts may be enrolled and dosed according to the same eligibility criteria in Part B to better define safety or PD effects. Additional cohorts may be enrolled at higher, lower, or intermediate dose levels, but will not exceed the maximum administered dose of 6.0 mg/kg, and will follow the protocol-specified dose escalation criteria.

Based on SRC review of accumulated safety, tolerability, and available PD data, up to 2 cohorts in Part B may be extended by up to 4 additional patients per cohort based on study progression and stopping rules. These patients will all receive active drug, not placebo.

4.7.3. Dose Suspension and Stopping Rules

For the purpose of this study, dose suspension and stopping rules are based on toxicity. Standard toxicity grading according to the CTCAE will be used to grade AEs. The term 'suspension' means that no further study drug will be administered at the dose level and that further dose escalation/progression will be suspended. If a suspension/stopping rule is met, there will be no further enrollment or dosing in the current cohort, or escalation to another cohort, in that part of the study and an ad hoc SRC meeting will be held. Following SRC review and recommendations, dosing may be resumed at the same or higher dose level following approval from the concerned Regulatory Authority and the independent ethics committee (IEC)/institutional review board (IRB), if required by local regulations. However, de-escalation to a lower dose or intermediate may be allowed without prior Regulatory Authority or IEC/IRB approval.

4.7.3.1. Cohort Progression/Escalation and Suspension/Stopping Rules in Part A

Study cohort progression/escalation and suspension/stopping rules for AEs considered possibly or definitely related to study drug for Part A are described in Table 6. If a subject meets a stopping rule, the subject will be managed as clinically indicated and asked to complete all safety assessments according to the protocol.

Table 6: Cohort Progression/Escalation and Suspension/Stopping Rules for Part A

CTCAE Grade[a]	Severity/ Seriousness	Reversibility	Number of Subjects Affected	Action	Effect on Dose Progression/Escalation
I	Mild	N/A	N/A	Next dose determined by SRC	N/A
II	Moderate	Showing signs of reversibility (ie, AE which shows signs of improvement in the judgment of Investigator)	≤2 subjects in different SOC		
			≤2 subjects in same SOC OR 3 subjects in different SOC[b]	Dose level may continue OR be extended AND dose escalation on hold until results of continuation or extension are available	Following continuation or extension, dose escalation may proceed as per clinical study protocol
			≥3 subjects in same SOC OR ≥4 subjects in different SOC[b]	Dose level suspended	A lower (intermediate) dose level may be administered in the next cohort AND dose continuation, extension, or escalation requires Regulatory Agency, IEC/IRB, and SRC approval
		Showing no signs of reversibility	≥2 subjects[b]		

Table 6: Cohort Progression/Escalation and Suspension/Stopping Rules for Part A

<table>
<tr><th>CTCAE Grade[a]</th><th>Severity/ Seriousness</th><th>Reversibility</th><th>Number of Subjects Affected</th><th>Action</th><th>Effect on Dose Progression/Escalation</th></tr>
<tr><td rowspan="4">III</td><td rowspan="3">Severe, not serious</td><td rowspan="2">Showing signs of reversibility (ie, AE which shows signs of improvement in the judgment of Investigator)</td><td>1 subject[b]</td><td>Dose level may continue
OR
be extended
AND
dose escalation on-hold until results of continuation or extension are available</td><td>Following continuation or expansion, dose escalation may proceed as per the clinical study protocol</td></tr>
<tr><td>≥2 subjects[b]</td><td rowspan="3">Dose level suspended</td><td rowspan="3">A lower (intermediate) dose level may be administered in the next cohort
AND
dose continuation, extension or escalation requires Regulatory Agency, IEC/IRB, and SRC approval</td></tr>
<tr><td>Showing no signs of reversibility</td><td rowspan="2">≥1 subject</td></tr>
<tr><td>Severe, serious</td><td>N/A</td></tr>
<tr><td>IV/V</td><td>Life-threatening/ Fatal</td><td>N/A</td><td>≥1 subject</td><td>Study suspended</td><td>Study continuation requires Regulatory Agency, IEC/IRB, and SRC approval</td></tr>
</table>

Abbreviations: AEs=adverse events; IEC=Independent Ethics Committee; IRB=Institutional Review Board; N/A=not applicable; SOC=System, Organ, Class; SRC=Safety Review Committee.

a Common Terminology Criteria for Adverse Events, Version 4.0 or higher, will be used to grade AEs.

b SRC review of Part A cohorts for escalation to the next dose level will occur following at least 48 hours of safety monitoring after the prior cohort has been dosed.

4.7.3.2. Individual Patient Progression/Escalation and Suspension/Stopping Rules in Part B

Individual progression/escalation and suspension/stopping rules applying only to individual patients enrolled in Part B are presented in Table 7. If an individual patient meets a suspension/stopping rule, the SRC will meet to determine whether dosing of the remaining

patients in the cohort can continue. Table 7 applies to AEs considered likely to be related to the underlying disease, or AEs considered possibly or definitely related to study drug.

CTCAE criteria will be used to grade AEs. For patients with abnormal renal function consistent with the underlying disease at baseline, evidenced by elevated serum creatinine, the change in this parameter from baseline should be used in grading AEs, rather than laboratory normal values.

Table 7: Individual Patient Progression/Escalation and Suspension/Stopping Rules for Part B

CTCAE Grade[a]	Severity/Seriousness	Action
I	Mild	No action required.
II	Moderate	Dose administration will be discontinued unless the AE is considered unrelated to study drug or reversible and continued dosing in the study is considered safe by the SRC.
III	Severe, not serious	
	Severe, serious	Dose administration will be discontinued.
IV/V	Life-threatening/Fatal	Dose administration will be discontinued.

Abbreviations: AEs=adverse events; SRC=Safety Review Committee.

a Common Terminology Criteria for Adverse Events will be used to grade AEs.

4.7.3.3. Cohort Progression/Escalation and Suspension/Stopping Rules in Part B

Study cohort progression/escalation and suspension/stopping rules for AEs considered likely to be related to the underlying disease and for AEs considered possibly or definitely related to study drug for Part B are described in Table 8. If a patient meets a stopping rule, the patient will be managed as clinically indicated and asked to complete all safety assessments according to the protocol. For patients with abnormal renal function consistent with their underlying disease at baseline, evidenced by elevated serum creatinine, the change in this parameter from baseline should be used in grading AEs, rather than laboratory normal values.

Table 8: Cohort Progression/Escalation and Suspension/Stopping Rules for Part B

Adverse Event(s)	Action
If ≥1 patient in a cohort experiences a severe AE and/or SAE, judged to be possibly or definitely related to study drug	Dosing within that cohort, and in any cohorts receiving a higher dose, will be suspended until SRC review. Following data review, the SRC may permit continuation of any ongoing Part B cohort. Any further dose escalation in a subsequent cohort will be suspended. Resumption of dose escalation will require a substantial amendment, approved by the regulatory authority and relevant ethics committee, if required by local regulations. However, de-escalation to a lower dose, or intermediate but lower dose, may be allowed by the SRC without a protocol amendment.
If ≥1 patient in Part B experiences a Grade 4 or 5 AE[a] judged to be possibly or definitely related to study drug	Dosing of all patients in Part B will be suspended until SRC review. Resumption of the study will require a substantial amendment, approved by the regulatory authority and relevant ethics committee, if required by local regulations.

Abbreviations: AE=adverse event; SAE=serious adverse event; SRC=Safety Review Committee.

a Common Terminology Criteria for Adverse Events will be used to grade AEs.

4.8. Adaptive Study Design Features

The use of adaptive study design features will allow for the continuing development of the study as new data emerge; therefore, the study can be adjusted in accordance with the prespecified areas, features, and limits listed in Table 9.

Table 9: Adaptive Study Design Areas, Features, and Limits

Areas	Features	Limits
A. Dosing regimen	1. Dosing regimens may be determined or adapted in accordance with safety, tolerability, and available PD data collected up to the decision making time point. 2. The term "dosing regimen" includes: (1) the dose level administered, (2) the frequency of dosing, and (3) the duration of dosing, ie, number of doses administered. Accordingly, these can be adjusted individually or in combination.	I. The starting dose for subjects in Part A will be 0.3 mg/kg. II. The starting dose for subjects in Part B of the study will not exceed doses already evaluated and considered safe and tolerable in Part A. III. In Part A, provided no safety or tolerability concerns have occurred, the maximum single dose administered will be 6 mg/kg. IV. The interval between doses in Part B may be increased or decreased, but will not exceed 3 doses over a 57 day period.

Table 9: Adaptive Study Design Areas, Features, and Limits

Areas	Features	Limits
		V. Duration of dosing: no more than 1 dose will be administered in Part A; dosing will not extend beyond 3 doses over a 57 day period in Part B. VI. The maximum dose level in Part B will not exceed a dose found to be safe and well-tolerated in Part A.
B. Overlap	1. Dosing regimens may overlap.	I. Protocol-specific minimum study progression/escalation requirements for the SRC must be met before dose progression/escalation (Section 4.7).
C. Cohort Sizes	1. Withdrawn subjects (Part A) and patients (Part B) can be replaced at the discretion of the Sponsor and Investigator if not withdrawn for safety reasons (Section 5.3). 2. Replacement subjects may be enrolled in an ongoing cohort, or dosed together as a group or separately.	I. Protocol-specific minimum requirements must be met before dose escalation (Section 4.7). II. Provided suspension or stopping rules have not been met, the SRC may permit up to 2 cohorts in Part B to be extended by up to 4 additional patients per cohort receiving active treatment to further characterize the safety, tolerability, and PD of the study drug.
D. Optional Cohorts	1. Optional cohorts may be included to explore additional dosing regimens	I. Up to 2 optional cohorts may be included in Part A. II. Up to 3 optional cohorts may be included in Part B. III. Cohort sizes will not exceed 8 subjects for each optional cohort in Part A, and 8 patients for each optional cohort in Part B. IV. To further explore safety, tolerability, PD, and/or PK, study drug dose level may be escalated or de-escalated to a lower and/or to intermediate dose levels. Dose escalation in Part B will not exceed a dose level found to be safe and well-tolerated in Part A. V. Dosing in optional cohorts will follow the same randomization list as that in planned cohorts in the respective parts of the study. VI. The total number of study subjects/patients indicated in Section 4.3 cannot be exceeded.
E. Samples and assessments	1. If applicable, the inpatient period may be prolonged for subjects/patients in all dose levels in all cohorts if considered by the Investigator to be necessary for clinical reasons or for expeditious	I. If applicable, a maximum extended inpatient period due to clinical concerns cannot be defined as the extension will be as long as necessary to ensure the safety of the subjects/patients. II. If applicable, an unplanned or extended inpatient stay because of a suspected adverse

Table 9: Adaptive Study Design Areas, Features, and Limits

Areas	Features	Limits
	conduct of the study. Prolonged inpatient periods will be recommended on a subject/patient, case-by-case basis and/or if the SRC considers it necessary for determining safety and tolerability for a future dose level cohort.	event will constitute a serious adverse event and will be considered in the context of the study stopping rules (Section 4.7).
	2. Specialist referrals may be made if considered clinically necessary by the Investigator, delegate, Sponsor, or SRC for subjects/patients on a case-by-case basis.	I. Specialist referrals for subjects/patients will be determined on a case-by-case basis, as necessary to ensure the safety of subjects/patients, so a maximum number of referrals cannot be defined.
	3. In Part A and B, timing of PK and/or PD samples (blood and/or urine) may be adjusted in accordance with evolving data and dosing schedule/regimen. 4. In Part A and B, additional or fewer PK and/or PD samples (blood and/or urine) may be obtained in accordance with evolving data and dosing schedule.	I. Maximum blood volume for any subject/patient is based on age and weight guidelines; the maximum blood volume will not be exceeded. (Section 7). II. Optional PK and/or PD analysis can be performed at any stage during or after the study to facilitate decision-making and/or to increase understanding of the compound (ie, samples will be obtained, but not necessarily analyzed; applicable to Features 3 and 4). III. The optional analysis is limited to the protocol-specified purpose (applicable to Features 3 and 4).
Abbreviations: PD=pharmacokinetic; PK=pharmacokinetic; SRC=Safety Review Committee.		

5. SELECTION AND WITHDRAWAL OF PATIENTS

5.1. Inclusion Criteria

Each subject/patient must meet all of the following inclusion criteria to be eligible for enrollment in the study.

5.1.1. Inclusion Criteria for Parts A and B

1. Male and female subjects aged 18 to 64 years (or age of legal consent, whichever is older), inclusive (Part A) and 6 to 64 years, inclusive (Part B).
2. Women of child-bearing potential must have a negative pregnancy test, cannot be breast feeding, and must be willing to use a highly effective method of contraception 14 days before first dose, throughout study participation until the completion of the follow-up periods (see Section 6.4).

3. Willing to comply with protocol-required visit schedule and visit requirements; and able to provide written informed consent and assent in the case of patients under the age of legal consent.

5.1.2. Additional Inclusion Criteria for Part A

4. Body Mass Index (BMI) of 18-30 kg/m^2, inclusive at screening and Day -1.

5.1.3. Additional Inclusion Criteria for Part B

5. Documentation or confirmation of PH1 as determined by genetic analysis and biochemical criteria and definite diagnosis based on the presence of AGXT mutations or reduced hepatic AGT enzyme activity that is considered evidence of the disease state (medical history)
6. 24-hour urinary oxalate excretion of >0.7 mmol/1.73m^2/day
7. Estimated GFR of >45 mL/min/1.73m^2 (calculation will be based on the MDRD formula for patients ≥18 years of age and the Schwartz Bedside Formula for patients <18 years of age; Section 11.2 in the Appendix)
8. If taking Vitamin B6 (pyridoxine), must have been on stable regimen for at least 90 days before study entry, and willing to remain on this stable regimen for the study duration.

5.2. Exclusion Criteria

Each subject/patient must not meet any of the following exclusion criteria to be eligible for enrollment in the study.

5.2.1. Exclusion Criteria for Parts A and B

1. Any uncontrolled or serious disease, or any medical or surgical condition (with the exception of PH1 for patients in Part B) that may either interfere with participation in the clinical study, and/or put the subject significant risk (according to the Investigator's judgment) if he/she participates in the clinical study
2. An underlying known disease or surgical or medical condition (with the exception of PH1 for patients in Part B) that in the opinion of the investigator might interfere with the interpretation of the clinical study results
3. Triplicate 12-lead ECG with clinically significant abnormalities
4. Active serious mental illness or psychiatric disorder including, but not limited to, schizophrenia, bipolar disorder, or severe depression requiring concurrent pharmacologic intervention
5. Clinically significant illness (with the exception of PH1 for patients in Part B) within 7 days before the first dose of study drug
6. Male and female subjects/patients aged 18 to 64 years (or age of legal consent, whichever is older), systolic blood pressure >140 mm Hg and/or a diastolic blood pressure >90 mmHg after 10 minutes of rest at screening
7. Abnormal for AST/ALT and any other clinical safety laboratory result considered clinically significant and unacceptable by the Investigator at screening or Day -1

8. Received an investigational agent within 90 days or 5 half-lives of the first dose of ALN-GO1 study drug, whichever is longer, or in the active follow-up phase of another clinical trial involving interventional treatment
9. Clinical laboratory evidence or a clinical diagnosis of viral hepatitis or human immunodeficiency virus (HIV) infection
10. Consume more than 14 (female) or 21 (male) units of alcohol per week (1 unit=1 glass of wine (125 mL), 1 measure of spirits or ½ pint of beer (subjects/patients above the age of legal consent). Alcohol abuse is defined as regular weekly intake of more than 21 units for males and 14 units for females (using alcohol tracker at http://www.nhs.uk/Tools/Pages/NHSAlcoholtracker.aspx).
11. History or clinical evidence of drug abuse, within the 12 months before screening. Drug abuse is defined as compulsive, repetitive and/or chronic use of drugs or other substances with or without problems related to their use and/or where stopping or a reduction in dose will lead to withdrawal symptoms (subjects/patients above the age of legal consent).
12. Positive for drugs of abuse at screening (subjects/patients above the age of legal consent)
13. Heavy smokers and users of nicotine (defined as the equivalent of ≥10 cigarettes per day).
14. Known history of allergic reaction to an oligonucleotide or GalNAc
15. History of intolerance to SC injection or relevant abdominal scarring (eg, surgical, burns)
16. Legal incapacity or limited legal capacity at screening of patient, parent, or legal guardian
17. Any conditions which, in the opinion of the Investigator would make the subject/patient unsuitable for enrollment or could interfere with the subject's/patient's participation in, or completion of, the study.
18. Donation of more than 500 mL of blood for adults (>18 years of age) within 90 days before the first dose of study drug (it is expected that children [6 to18 years of age] will not have donated blood).
19. Women who are pregnant or breastfeeding

5.2.2. Additional Exclusion Criteria for Part B

20. Clinical evidence of systemic oxalosis, including but not limited to, myocardial dysfunction, bone marrow, bone, or eye infiltration with oxalate as determined by the Investigator
21. For patients <18 years old, diastolic and/or systolic blood pressure equal to or greater than the 95th percentile for age, gender, and height (see Appendix 11.4 Table 14 and Table 15) after 10 minutes of rest at screening.
22. Echocardiography (Echo) assessment of abnormal left ventricular systolic function, defined as left ventricular ejection fraction <55% at screening
23. Troponin I greater than the upper limit of normal (ULN) at screening (abnormal results should be repeated)

5.3. Removal from Therapy or Assessment

Subjects/Patients are free to discontinue treatment or withdraw from the study at any time and for any reason, without penalty to their continuing medical care. Any discontinuation of treatment or withdrawal from the study must be fully documented in the electronic case report form (eCRF), and should be followed up by the Investigator. The Investigator may withdraw a subject/patient at any time if this is considered to be in the subject's/patient's best interest.

Discontinuation of study drug and withdrawal from the study are described in Section 5.3.1 and Section 5.3.2, respectively.

5.3.1. Discontinuation of Study Drug

The Investigator or designee may discontinue dosing in a subject/patient if the subject/patient:

- Is in violation of the protocol
- Experiences a serious or intolerable AE
- Requires a prohibited medication
- Becomes pregnant
- Is found to be noncompliant with the protocol-requirements in a manner that is considered by the Investigator to compromise patient safety and/or the integrity of the study

The Investigator will confer with the Sponsor, or Medical Monitor, before discontinuing dosing. Subjects/Patients who are pregnant will be discontinued from dosing immediately (see Section 7.5.7.6 for reporting and follow-up of pregnancy). Subjects/patients who discontinue study drug may be replaced (see Section 5.3.3). Subjects/patients who discontinue study drug dosing for any reason will be encouraged to complete the remaining assessments through the remaining scheduled study visits so that their experience is captured in the study analyses. Subjects/Patients who discontinue study drug but who agree to attend the remaining scheduled study visits will not be considered withdrawn from study. Such subjects/patients may receive local standard of care treatment for their disease, as applicable.

5.3.2. Withdrawal From Study

A subject/patient may withdraw from the study at any time. However, study integrity and interpretation is optimal if all randomized patients continue study assessments and follow-up. Subjects/Patients considering withdrawing from the study should be informed that they can alternatively discontinue study treatment and complete the remaining scheduled study visits or agree to alternative follow-up processes (as described in Section 5.3.1).

If a subject/patient still chooses to withdraw consent/assent, every effort should be made to conduct the assessments performed at the EOT visit. When a subject/patient withdraws from the study, the primary reason for withdrawal must be recorded in the appropriate section of the eCRF and all efforts will be made to complete and report the observations as thoroughly as possible. If a subject/patient withdraws due to a serious adverse event (SAE), the SAE should be followed as described in Section 7.5.7.

5.3.3. Replacement of Subjects or Patients

Replacement subjects/patients may be enrolled to ensure that the minimum data requirements for SRC dose escalation decisions and study progression are met, as described in Section 4.6 and Section 4.7. Subjects/patients who are discontinued from treatment for reasons other than experiencing an AE may be replaced following discussion between the Sponsor and Investigator.

The replacement subject/patient will be assigned a unique study identification number and will receive the same study drug assignment and dose level as the subject/patient who is being replaced.

6. TREATMENTS

6.1. Treatments Administered

The treatments administered in this study are ALN-GO1 and placebo. ALN-GO1 is a synthetic, double-stranded small interfering RNA oligonucleotide directed against hydroxyacid oxidase 1 mRNA that is covalently linked to a ligand containing 3 N-acetylgalactosamine residues.

Study drug supplied for this study must not be used for any purpose other than the present study and must not be administered to any person not enrolled in the study. Study drug that has been dispensed to a subject/patient and returned unused must not be re-dispensed to a different subject/patient.

6.2. Investigational Study Drug

Detailed information describing the preparation, administration, and storage of ALN-GO1 is provided in the Pharmacy Manual.

6.2.1. Description

ALN-GO1 Solution for Injection (SC use) is a clear, colorless to pale yellow solution essentially free of particulates. ALN-GO1 will be supplied by the Sponsor as a sterile solution for SC injection at a targeted concentration of 200 mg/mL. Placebo will be supplied by the clinical study center as a sterile, preservative-free normal saline 0.9% solution for SC injection.

6.2.2. Dose and Administration

Dose cohorts for this study are described in Section 4.7.1 (Part A) and Section 4.7.2 (Part B). Subjects/patients will be administered study drug by SC injection(s). Study drug will be administered by qualified clinical study center staff under the supervision of the Investigator, or designee, and the injection site will be marked and mapped for later observation. The preferred site of injection is the abdomen. Optional additional sites are the upper arms and thighs. If a local reaction around the injection site occurs, photographs may be obtained. Detailed instructions for study drug administration are in the Pharmacy Manual.

6.2.3. Dose Modifications

The dose modifications permitted in this study are described in Section 4.7.3.

6.2.4. Preparation, Handling, and Storage

Study drug may be dispensed only by the Investigator, by a staff member specifically authorized by the Investigator, or by pharmacy staff, as appropriate.

Each clinical study center will be responsible for assembly and labeling of injection syringe(s) according to procedures detailed in the Pharmacy Manual. The pharmacy staff will prepare the study drug using an aseptic technique. The amount (in mg) of study drug to be administered will be determined based on the assigned dose level for the cohort and the Day -1 body weight for healthy subjects and patients with PH1. On dosing days, the pharmacist, or designee, will withdraw the required amount of study drug into 1 or more syringes to be administered to the subject/patient on that day. The procedure for preparing study drug and the volume to be loaded into each syringe is provided in the Pharmacy Manual.

No special procedures for the safe handling of ALN-GO1 are required. Study drug will be stored upright and refrigerated at approximately 2 to 8°C protected from light in the storage area of the clinical study center pharmacy, in a secure, temperature-controlled, locked environment with restricted access. Any deviation from the recommended storage conditions should be reported to the Sponsor and use of the study drug halted until authorization for its continued use has been provided by the Sponsor or designee. Additional storage details are provided in the Pharmacy Manual.

A Sponsor representative or designee will be permitted, upon request, to audit the supplies, storage, dispensing procedures, and records.

6.2.5. Packaging and Labeling

ALN-GO1 (solution for SC injection) is packaged in 2-mL glass vials with a fill volume of no less than 0.55 mL to allow for complete withdrawal of 0.5 mL of drug product at the pharmacy. The container closure system consists of a Type I glass vial, a Teflon-faced 13-mm stopper, and a flip-off aluminum seal.

6.2.6. Accountability

The Investigator or designee will maintain accurate records of receipt and the condition of the study drug supplied for this study, including dates of receipt. In addition, accurate records will be kept of when and how much study drug is dispensed and administered to each patient in the study. Any reasons for departure from the protocol dispensing regimen must also be recorded.

At the completion of the study, there will be a final reconciliation of all study drugs.

Further instructions about drug accountability are detailed in the Pharmacy Manual.

6.3. Concomitant Medications

All concomitant medications must be recorded in the eCRF. Concomitant medications will be coded using the WHO Drug Dictionary.

Any concomitant medication that is required for the patient's welfare may be administered by the Investigator. However, it is the responsibility of the Investigator to ensure that details regarding the medication are recorded on the eCRF.

6.3.1. Permitted Concomitant Medications

For Part A and Part B, the following medications/treatments are permitted:

- hormone replacement therapy
- oral contraceptives, injectable progesterone, and subdermal implants are permitted for contraception
- acetaminophen (maximum 2 g daily) for treatment of AEs
- at the discretion of the Investigator, prescription or nonprescription medications may be permitted when necessary to treat an AE; before the subject uses any prescription or nonprescription medications, the Investigator or delegate must be consulted and justify their use

For Part B, the following medication/treatment is also permitted:

- Vitamin B6 (pyridoxine): If taking vitamin B6, must have been on stable regimen for at least 90 days before study entry, and willing to remain on this stable regimen for the study duration.
- Sodium or potassium citrate

6.3.2. Prohibited Concomitant Medications

For Part A, the following medications/treatments are not permitted:

- Any OTC medications, except routine vitamins from 7 days before the first dose of study drug, unless considered not clinically relevant by the Investigator and Sponsor
- Prescription medications not specified in Section 6.3.1 from 14 days or 5 half-lives (whichever is longer) before the first dose of study drug.

For Part B, there are no prohibited medications; however, all concomitant medications must be reviewed by the Investigator.

6.4. Contraceptive Requirements

Women of child-bearing potential (WOCBP) must be willing to use a highly effective method of contraception 14 days before first dose, and throughout study participation until the completion of the follow-up periods. Highly effective methods of birth control result in a low failure rate (ie, less than 1% per year). Birth control methods, which may be considered as highly effective, include:

- Established use of oral (except low-dose gestagens [eg, lynestrenol and norethisterone]), implantable, injectable, or transdermal hormonal methods of conception
- Placement of an intrauterine device
- Placement of an intrauterine hormone-releasing system
- Bilateral tubal occlusion

- Surgical sterilization of male partner (with the appropriate post-vasectomy documentation of the absence of sperm in the ejaculate; for female patients on the study, the vasectomized male partner should be the sole partner for that patient);
- True sexual abstinence, when in line with the preferred and usual lifestyle of the patient (and for adolescents who are not sexually active). Periodic abstinence (eg, calendar, ovulation, symptothermal, post-ovulation methods) and withdrawal are not acceptable methods of contraception. Abstinent subjects have to agree to use 1 of the abovementioned contraceptive methods, if they start sexual relationships during the study until the completion of the follow-up periods.

WOCBP includes any female patient who has experienced menarche and who is not postmenopausal or permanently sterilized (eg, bilateral tubal occlusion, hysterectomy, or bilateral salpingectomy). Postmenopausal state is defined as no menses for 12 months without an alternative medical cause, confirmed by follicle stimulating hormone (FSH) level within the postmenopausal range.

No male contraception is considered to be required.

6.5. Treatment Compliance

All doses will be administered by qualified clinical study center personnel and any missed doses will be reported and recorded on the eCRF.

7. STUDY ASSESSMENTS

The schedules of assessments are provided in Table 1 for Part A; and in Table 2, Table 3, and Table 4 for Part B.

7.1. Screening/Baseline Assessments

Subject/patient demographic data and complete medical history (including documentation or confirmation of PH1) will be obtained at Screening. Screening safety assessments are included in the Schedule of Assessments, with additional details provided in Section 7.5, as applicable.

7.1.1. Renal Function in Part B

7.1.1.1. Urinary Oxalate Excretion and Creatinine Clearance

During the screening period, 24-hour urine collections will be completed at 2 separate time points. The first screening 24-hour urine collection will be used to assess eligibility. The second screening 24-hour urine collection will only be initiated after eligibility is confirmed. Blood samples for serum creatinine will be obtained throughout the study as part of clinical laboratory assessments (see Section 7.5.6) for the calculation of creatinine clearance.

Patients will be instructed to collect urine samples for the full 24-hour period; however, after confirming eligibility, for patients unable to collect urine samples for the full 24-hour period, urinary oxalate excretion may be calculated from samples collected over an 18 to 24 hour period.

Patients will have the option to bring the 24-hour urine collections to the clinical study center at specified follow-up visits, courier samples to the clinical study center or to the vendor performing analyses, or elect to have the 24-hour urine collected during an inpatient stay at the clinical study center for other assessments.

7.1.1.2. Estimated Glomerular Filtration Rate

eGFR (mL/min/1.73m^2) will be calculated to confirm eligibility and to assess renal function during the study. The calculation will be based on the Modification of Diet in Renal Disease (MDRD) formula for patients ≥18 years of age and the Schwartz Bedside Formula for patients <18 years of age (Appendix 11.2).[10, 11]

7.2. Pharmacodynamic Assessments

Urine and blood samples will be collected for assessment of PD parameters (oxalate and glycolate concentrations) at the time points in the Schedule of Assessments.

In Part B, single, 24-hour urine collections [redacted] will be collected for the analysis of urinary glycolate and oxalate excretion, and creatinine clearance. The 24-hour urine collection starting on Day -1 must conclude on Day 1 before administration of the first dose of study drug. Blood samples will also be collected for the analysis of plasma glycolate concentration.

Options for providing 24-hour urine collections to the clinical study center are in Section 7.1.1.1.

Details regarding the processing and aliquoting of samples for storage and PD analyses will be provided in the Laboratory Manual.

7.3. Pharmacokinetic Assessments

Blood and urine samples will be collected for assessment of ALN-GO1 PK parameters in Part A and Part B of the study at the time points in the Schedule of Assessments. In Part B, samples may also be analyzed for the [redacted] A detailed schedule of time points for the collection of blood and urine samples for PK analysis is in Table 10 for Part A and in Table 11 and Table 12 for Part B in Appendix 11.1.

Options for providing urine collections to the clinical study center are in Section 7.1.1.1.

The concentration of ALN-GO1 will be determined using a validated assay. Details regarding the processing, shipping, and analysis of the samples will be provided in the Laboratory Manual.

7.4. Exploratory Assessments

Blood and urine samples will be collected for exploratory analyses of plasma oxalate concentration[redacted]

Details regarding the collection, processing, storage, and shipping of samples will be in the Laboratory Manual.

7.5. Safety Assessments

The assessment of safety during the course of the study will consist of the surveillance and recording of AEs, including serious adverse events (SAEs), recording of concomitant medication and measurements of vital signs, weight, physical examination and ECG findings, and clinical laboratory assessments; Echo assessments will also be evaluated in Part B only.

Safety will be monitored over the course of the study by an SRC as described in Section 4.6.

7.5.1. Vital Signs

Vital sign measurements include blood pressure, heart rate, body temperature, and respiratory rate. Vital signs will be measured in the supine position (should be consistent for each patient), after the subject/patient has rested comfortably for 10 minutes. Body temperature will be measured using a tympanic or oral thermometer. A patient's body temperature should be measured using the same method throughout the course of the study.

7.5.2. Weight and Height

Body weight will be measured in kilograms. Body mass index will be calculated from the height and weight. For patients <18 years of age, height will be measured in centimeters, and in triplicate, to facilitate calculation of eGFR using the Schwartz Bedside Formula.

7.5.3. Physical Examination

A full physical examination will include general appearance, eyes, ears, nose and throat, chest/respiratory, heart/cardiovascular, gastrointestinal/liver, musculoskeletal/extremities, dermatological/skin, thyroid/neck, lymph nodes, and neurological/psychiatric assessments.

A symptom-directed physical examination will include the evaluation of changes in symptoms, or the onset of new symptoms, since the last visit.

7.5.4. Electrocardiogram

Triplicate 12-lead ECGs will be measured approximately 5 minutes apart. Recordings will be obtained after the subject/patient has rested comfortably in the supine position for approximately 10 minutes. Subjects/Patients should remain supine between ECGs. ECGs should be performed at approximately the same time of day throughout the study.

Additional ECGs may be collected at the discretion of the Investigator. The electrophysiological parameters assessed will include, but are not limited to, rhythm, ventricular rate, PR interval, QRS duration, QT interval, ST and T waves, Bazett-corrected QT interval (QTcB) and Fridericia corrected QT interval (QTcF).

The Investigator or designee is responsible for reviewing the ECGs to assess whether the results have changed since the Screening/Baseline visit and to determine the clinical significance of the results. For any clinically significant changes from the Screening/Baseline visit (eg, ischemic ECG changes, wave/interval changes, or arrhythmia), the Investigator must contact the Medical Monitor to discuss continued participation of the subject/patient in the study.

7.5.5. Echocardiography (Part B only)

Echo assessments will be performed according to instructions provided in a study manual. The Investigator or designee is responsible for reviewing the Echos to assess whether the results have changed since the Screening/Baseline visit and to determine the clinical significance of the results. For any clinically significant changes from the Screening/Baseline visit, the Investigator must contact the Medical Monitor.

7.5.6. Clinical Laboratory Assessments

Clinical laboratory tests will be evaluated as noted in the Schedule of Assessments. In the event of an unexplained clinically relevant abnormal laboratory test occurring after study drug administration, the test should be repeated and followed up at the discretion of the Investigator until it has returned to the normal range or stabilized, and/or a diagnosis is made to adequately explain the abnormality.

Hematology	
Complete blood count with differential	
Serum Chemistry	
Sodium	Potassium
BUN	Phosphate
Creatinine and eGFR calculation (using the MDRD or Schwartz formula depending on age)	Albumin
Uric acid	Calcium
Total protein	Carbon dioxide/bicarbonate
Glucose	Chloride
Cardiac Enzyme	
Troponin I	
Liver Function Tests	
AST	ALP
ALT	Bilirubin (total and direct)
Coagulation Panel	
Prothrombin time	International Normalized Ratio
Activated partial thromboplastin time	

Urinalysis	
Visual inspection for appearance and color	Bilirubin
pH (dipstick)	Nitrite
Specific gravity	RBCs
Ketones	Urobilinogen
Albumin (optional)	Leukocytes
Glucose	Microscopy (if clinically indicated)
Protein	
Immunogenicity	
Antidrug Antibodies	
Pregnancy Testing (WOCBP only)	
β-human chorionic gonadotropin	

Abbreviations: AST=aspartate transaminase; ALP=alkaline phosphatase; ALT=alanine transaminase; BUN=blood urea nitrogen; eGFR=estimated glomerular filtration rate; MDRD=modification of diet in renal disease; WOCBP=women of child bearing potential.

7.5.6.1. Immunogenicity

Blood samples will be collected to evaluate antidrug antibodies (ADA). Blood samples for ADA testing that are collected on the day of dosing must be collected before study drug administration. Blood samples to evaluate ADAs will be collected throughout the treatment and follow-up periods as detailed in the Schedule of Assessments. Confirmed positive ADA samples will be tested for cross-reactivity with DNA and nucleic acids.

Details regarding the processing, shipping, and analysis of the samples will be provided in the Laboratory Manual.

7.5.6.2. Pregnancy Testing

A pregnancy test will be performed for WOCBP only. A serum pregnancy test will be performed at Screening or after the onset of menarche if the patient was not of childbearing potential at screening, and urine pregnancy tests will be performed thereafter per the Schedule of Assessments and any time pregnancy is suspected. The results of the pregnancy test must be known before study drug administration. Women who are pregnant are not eligible for study participation. Any woman with a positive pregnancy test during the study will be discontinued from study drug, but will continue to be followed for safety. Women determined to be pregnant while on study will be followed until the pregnancy outcome is known (see Section 7.5.7.6 for follow-up instructions).

7.5.6.3. Drugs of Abuse

At Screening, urine will be tested for the following drugs of abuse: amphetamines, barbiturates, benzodiazepines, cannabinoids, opiates, cocaine, and methadone. If a subject/patient fails the screening, they will be excluded from the study, unless, in Part B only, the positive result is due to prescribed medications for patients. A repeat drug screen may only be performed when

methodological reasons are believed to have led to a false positive. Borderline positive results, unless covered by the preceding condition, are to be considered as positive and the subject/patient will be excluded from the study. If subjects/patients are found to be positive due to short term medication use, eg, antitussives medication, they may undergo a repeat drug screen to determine if they continue to meet study requirements.

All subjects/patients should refrain from consuming poppy seeds 48 hours before screening and then from 48 hours before study drug administration until follow-up has been completed. Poppy seed ingestion may cause a positive result for opiates in urine drug screen.

7.5.6.4. Maximum Blood Volume

The maximum blood volume for subjects in Part A will not exceed 500 mL over the course of the study.

In Part B, the maximum blood volume, which will be collected from pediatric patients over the course of the study, will be based on age and weight and will not exceed those specified in Table 13 from the Feinstein Institute for Medical Research Human Subject Protection Program Guidance Document (Section 11.3 in the Appendix).[12] The maximum blood volume for adult and pediatric patients initially randomized to ALN-GO1 is not expected to exceed 400 mL over the course of the study. The maximum blood volume for adult and pediatric patients initially randomized to placebo is not expected to exceed 450 mL over the course of the study. The blood volume limits for patients in Part B are based on those for 6 year old girls in the 5th percentile for weight, the smallest patients who may be enrolled in the study.

7.5.7. Adverse Events

7.5.7.1. Definitions

Adverse Event

According to the International Conference on Harmonization (ICH) E2A guideline Definitions and Standards for Expedited Reporting, and 21 CFR 312.32, IND Safety Reporting, an AE is any untoward medical occurrence in a patient or clinical investigational subject administered a medicinal product and which does not necessarily have a causal relationship with this treatment.

Serious Adverse Event

A serious adverse event (SAE) is any untoward medical occurrence that at any dose:

- Results in death
- Is life-threatening (an event which places the patient at immediate risk of death from the event as it occurred. It does not include an event that had it occurred in a more severe form might have caused death)
- Requires in-patient hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability or incapacity
- Is a congenital anomaly or birth defect

- Is an important medical event that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient and may require intervention to prevent 1 of the other outcomes listed in the definition above (eg, events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias, convulsions, or the development of drug dependency or abuse).

Adverse Event Severity

AE severity should be graded using the Common Terminology Criteria for Adverse Events (CTCAE). Refer to CTCAE for unique clinical descriptions of severity (Grades I through IV) for each AE based on the following general guideline:

Grade I:	Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated
Grade II:	Moderate; minimal, local or noninvasive intervention indicated; limiting age appropriate instrumental activity of daily living (eg, preparing meals, shopping for groceries or clothes, using the telephone, managing money)
Grade III:	Severe or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care activity of daily living (ie, bathing, dressing and undressing, feeding self, using the toilet, taking medications, and not bedridden)
Grade IV:	Life-threatening consequences; urgent intervention indicated
Grade V:	Death related to an adverse event

Changes in severity should be documented in the medical record to allow assessment of the duration of the event at each level of severity. Adverse events characterized as intermittent require documentation of the start and stop of each incidence. When changes in the severity of an AE occur more frequently than once a day, the maximum severity for the experience that day should be noted. If the severity category changes over a number of days, then those changes should be recorded separately (with distinct onset dates).

AE severity and seriousness are assessed independently. 'Severity' characterizes the intensity of an AE. 'Serious' is a regulatory definition and serves as a guide to the Sponsor for defining regulatory reporting obligations (see definition for Serious Adverse Event).

Relationship of the Adverse Event to Study Treatment

The relationship of each AE to study treatment should be evaluated by the Investigator using the following criteria:

Definitely related:	A clinical event, including laboratory test abnormality, occurring in a plausible time relationship to the medication administration, and which cannot be explained by concurrent disease or other drugs or chemicals. The response to withdrawal of the drug should be clinically plausible.

Possibly related:	A clinical event, including laboratory test abnormality, with a reasonable time sequence to the medication administration, but which could also be explained by concurrent disease or other drugs or chemicals. Information on the drug withdrawal may be lacking or unclear.
Unlikely related:	A clinical event, including laboratory test abnormality, with little or no temporal relationship to medication administration, and which other drugs, chemicals, or underlying disease provide plausible explanations.
Not related:	A clinical event, including laboratory test abnormality that has no temporal relationship to the medication or has more likely alternative etiology.

7.5.7.2. Eliciting and Recording Adverse Events

Eliciting Adverse Events

The patient should be asked about medically relevant changes in his/her health since the last visit. The patient should also be asked if he/she has been hospitalized, had any accidents, used any new medications, or changed concomitant medication routines (both prescription and OTC). In addition to patient observations, AEs will be documented from any clinically relevant laboratory findings, physical examination findings, ECG changes, or other findings that are relevant to patient safety.

Recording Adverse Events

The Investigator is responsible for recording non-serious AEs that are observed or reported by the patient after administration of the first dose of study drug regardless of their relationship to study drug through the end of study. Non-serious AEs will be followed until the end of study.

The Investigator is responsible for recording SAEs that are observed or reported by the patient after the time when the informed consent is signed regardless of their relationship to study drug through the end of study. SAEs will be followed until satisfactory resolution, until baseline level is reached, or until the SAE is considered by the Investigator to be chronic or the patient is stable, as appropriate.

All AEs must be recorded in the source records for the clinical study center and in the eCRF for the patient, whether or not they are considered to be drug-related. Each AE must be described in detail: onset time and date, description of event, severity, relationship to investigational drug, action taken, and outcome (including time and date of resolution, if applicable).

For purposes of this study, ISRs are considered to be Adverse Events of Clinical Interest. These AEs will be recorded both on a supplemental eCRF and on an Adverse Event of Clinical Interest form. Refer to the eCRF completion guidelines for details on these forms.

For SAEs, record the event(s) in the eCRF. If the EDC system is unavailable, complete the back-up SAE form.

7.5.7.3. Serious Adverse Events Require Immediate Reporting to Sponsor/Designee

An assessment of the seriousness of each AE will be made by the Investigator. Any AE and laboratory abnormality that meets the SAE criteria in Section 7.5.7.1 must be reported to the Sponsor or designee within 24 hours from the time that clinical study center staff first learns of the event. All SAEs must be reported regardless of the relationship to study drug.

The initial report should include at least the following information:

- Subject/Patient's study number
- Description and date of onset of the event
- Criterion for serious
- Preliminary assignment of relationship to study drug, and
- Investigator/site information

To report the SAE, complete the eCRF. If the EDC system is unavailable, complete the back-up SAE form. Within 24 hours of receipt of follow-up information, the Investigator must update the SAE form. SAEs must be reported using the contact information provided in the Study Reference Guide.

Appropriate remedial measures should be taken by the Investigator using his/her best medical judgment to treat the SAE. These measures and the patient's response to these measures should be recorded. All SAEs, regardless of relationship to study drug, will be followed by the Investigator until satisfactory resolution or the Investigator deems the SAE to be chronic or stable. Clinical, laboratory, and diagnostic measures should be employed by the Investigator as needed to adequately determine the etiology of the event.

7.5.7.4. Sponsor Safety Reporting to Regulatory Authorities

The Sponsor or its representative is required to report certain study events in an expedited manner to the Food and Drug Administration, the European Medicines Agency's EudraVigilance electronic system according to Directive 2001/20/EC, and to all country Regulatory Authorities where the study is being conducted, according to local applicable regulations.

The following describes the safety reporting timeline requirements for suspected unexpected serious adverse reactions (SUSARs) and other reportable events:

Immediately and within 7 calendar days

- Any suspected adverse reaction that is associated with the use of the study drug, unexpected, and fatal or life threatening. Follow-up information must be reported in the following 8 days.

Immediately and within 15 calendar days

- Any suspected adverse reaction that is associated with the use of the study drug, unexpected, and serious, but not fatal or life threatening, and there is evidence to suggest a causal relationship between the study drug and the reaction.
- Any finding from tests in laboratory animals that suggest a significant risk for human patients including reports of mutagenicity, teratogenicity, or carcinogenicity.
- Any event in connection with the conduct of the study or the development of the study drug that may affect the safety of the trial patients.

In addition, periodic safety reporting to regulatory authorities will be performed by the Sponsor or its representative according to national and local regulations.

7.5.7.5. Serious Adverse Event Notification to the Institutional Review Board/Independent Ethics Committee

SUSARs will be reported to the IRB/IEC per their institutional policy by the Investigator or Sponsor (or Sponsor designee) according to country requirements. Copies of each report and documentation of IRB/IEC notification and acknowledgement of receipt will be kept in the Investigator's study file.

7.5.7.6. Pregnancy Reporting

If a female patient becomes pregnant during the course of this study, the Investigator must report the pregnancy to the Sponsor or designee within 24 hours of being notified of the pregnancy. Details of the pregnancy will be recorded on the pregnancy reporting form. The patient should receive any necessary counseling regarding the risks of continuing the pregnancy and the possible effects on the fetus.

The pregnancy should be followed by the Investigator until completion. At the completion of the pregnancy, the Investigator will document the outcome of the pregnancy. If the outcome of the pregnancy results in a postpartum complication, spontaneous abortion, stillbirth, neonatal death, or congenital anomaly, then the Investigator should follow the procedures for reporting an SAE as outlined in Section 7.5.7.3.

7.5.7.7. Overdose Reporting

An overdose is defined as any dose administered to or taken by a patient (accidentally or intentionally) that exceeds the highest daily dose, or is at a higher frequency, than included in the protocol. It is up to the investigator to decide whether a dose is to be considered an overdose, in consultation with the Sponsor. Overdose must be recorded in the eCRF.

All reports of overdose (with or without an AE) must be reported within 24 hours to the Sponsor or designee.

8. STATISTICS

This is a randomized, single-blind, placebo-controlled study of ALN-GO1 administered subcutaneously to healthy adult subjects (Part A) and to adult and pediatric patients with PH1 disease (Part B). The study is designed to evaluate the safety, tolerability, PK, and PD of single- and multiple-ascending doses of ALN-GO1.

8.1. Determination of Sample Size

The sample size was based on clinical considerations rather than power calculations. Up to 64 participants (40 subjects and 24 patients) are planned to be enrolled in this study, including optional and expansion cohorts (see Table 9).

8.2. Statistical Methodology

A Statistical Analysis Plan (SAP) will be finalized before database lock. The plan will detail the implementation of all planned statistical analyses in accordance with the principal features stated

in the protocol. Any changes to the methods described in the final SAP will be described and justified as needed in the clinical study report.

Statistical analyses will be primarily descriptive. Analyses will be performed using SAS® (Version 9.2, or higher). Data from Parts A and B will be analyzed separately. Tabular summaries will be generated by dose level and dosing regimen of ALN-GO1 and placebo (pooled across all cohorts) for Part A and Part B (through Day 85).

Safety and PD data will be summarized by dose level and dosing regimen of ALN-GO1 compared to placebo for data collected up to, and including, study Day 85. Data from placebo patients from all cohorts will be combined. Data collected after Day 85 will be summarized separately for patients in quarterly dosing cohorts who receive a second dose of ALN-GO1 on Day 85. Additionally, all safety and PD data collected from all patients in Part B during the ALN-GO1 dosing period, regardless of treatment sequence, will be combined and summarized by ALN-GO1 dose level and regimen.

Data collected beyond the designated dosing period will be summarized or presented in data listings.

Descriptive statistics will be presented for continuous variables. Frequencies and percentages will be presented for categorical and ordinal variables. Percentages will be based on the number of non-missing values in a dose group.

8.2.1. Populations to be Analyzed

The populations (analysis sets) are defined as follows:

- Safety Analysis Set: All subjects/patients who receive at least 1 dose of study drug.
- PK Analysis Set: All subjects/patients who receive at least 1 dose of study drug and have at least 1 postdose sample for PK parameters and who have evaluable PK data.
- PD Analysis Set: All subjects/patients who receive at least 1 dose of study drug and who have at least 1 postdose blood and/or urine sample evaluable for PD parameters.

8.2.2. Examination of Subgroups

Subgroup analyses may be conducted for selected endpoints. Detailed methodology will be provided in the SAP.

8.2.3. Handling of Missing Data

Unrecorded values will be treated as missing. The appropriateness of the method(s) described for handling missing data may be reassessed and documented in the SAP prior to database lock. Depending on the extent of missing values, further investigation may be made into the sensitivity of the analysis results to the method(s) specified.

8.2.4. Baseline Evaluations

Demographics and other baseline characteristics will be summarized using the Safety Analysis Set by dose level groups and placebo. If the PK and PD Analysis Sets contain a different set of subjects/patients compared to those included in the Safety Analysis Set, then demographic information will be summarized separately for those analysis sets. Descriptive statistics for age,

race, ethnicity, gender, height, weight, and BMI will be provided. Demographic information for patients in Part B may include additional disease-specific information collected at baseline.

8.2.5. Pharmacodynamic Analyses

For Part A, PD analysis will be the comparison of change from baseline in plasma glycolate concentration for each subject in ALN-GO1 dose groups and placebo.

For Part B, PD analysis will be the comparison of the change from baseline in urinary oxalate excretion (oxalate concentration in 24-hour urine collection), with additional analyses of the change from baseline in urinary glycolate excretion (glycolate concentration in 24-hour urine collection), plasma glycolate concentration, and calculated creatinine clearance, for each patient in active dose groups and placebo.

The PD parameters will be summarized using descriptive statistics for actual results and relative to baseline for each follow-up time points.

8.2.6. Pharmacokinetic Analyses

Pharmacokinetic analyses will be conducted using noncompartmental methods.

Pharmacokinetic parameters include, but are not limited to, C_{max}, t_{max}, AUC, $t_{½}$, f_e/F, and CL_R. Other parameters may be calculated, if necessary.

8.2.7. Safety Analyses

The primary safety parameter is the incidence of AEs. Safety will also be evaluated through vital signs, ECGs, clinical laboratory assessments, and physical examinations; Echo assessments will be evaluated in Part B only. ADAs will also be analyzed.

AEs will be summarized by the MedDRA System Organ Class and Preferred Term (Version 16, or higher). Prior and concomitant medications will be classified according to the World Health Organization (WHO) drug dictionary.

Separate tabulations of the incidence of treatment emergent adverse events (TEAEs), TEAEs by maximum severity, treatment-related AEs, SAEs, and discontinuation due to AEs will be provided. By-subject listings will also be provided for any deaths, SAEs and AEs leading to discontinuation.

Descriptive statistics will be provided for clinical laboratory data and vital signs data, presented as both actual values and changes from baseline over time. Clinical laboratory assessment shift tables from baseline to worst post-values will be presented. Abnormal physical examination findings, ECG, and Echo (Part B only) data will be presented in a by-patient data listing.

Descriptive statistics will be provided for ECG interval data and presented as both actual values and changes from baseline relative to each on-study evaluation and to the last evaluation on study. Details of any abnormalities will be included in patient listings.

ADA results will be provided in a by-patient data listing.

8.2.8. Interim Analysis

There is no formal interim analysis planned for this study.

9. STUDY ADMINISTRATION

9.1. Ethical and Regulatory Considerations

This study will be conducted in accordance with the protocol, all applicable regulatory requirements, and the guidelines of Good Clinical Practice (GCP). Compliance with GCP provides public assurance that the rights, safety, and well-being of study subjects/patients are protected consistent with the principles that have their origin in the Declaration of Helsinki.

9.1.1. Informed Consent

The Investigator will ensure that the subject/patient/legal guardian is given full and adequate oral and written information about the nature, purpose, possible risk and benefit of the study. Subjects/Patients/Legal guardians must also be notified that they are free to discontinue from the study at any time. The subject/patient/legal guardian should be given the opportunity to ask questions and allowed time to consider the information provided. In the case of patients under the age of legal consent, legal guardian(s) must provide informed consent and the patient should provide assent per local regulations and institutional standards.

The subject's/patient's/legal guardian's signed and dated informed consent (or assent, if applicable) must be obtained before conducting any study procedures.

The Investigator must maintain the original, signed Informed Consent Form (or assent, if applicable). A copy of the signed Informed Consent Form (or assent, if applicable) must be given to the subject/patient/legal guardian.

9.1.2. Ethical Review

The final study protocol, including the final version of the ICF, must be approved or given a favorable opinion in writing by an IRB or IEC, as appropriate. The Investigator must submit written approval before he or she can enroll any subject/patient into the study.

The Investigator is responsible for informing the IRB or IEC of any amendment to the protocol in accordance with local requirements. In addition, the IRB or IEC must approve all advertising used to recruit subjects/patients for the study. The protocol must be reapproved by the IRB or IEC upon receipt of amendments and annually, as local regulations require.

Initial IRB approval of the protocol, and all materials approved by the IRB for this study including the subject/patient consent form (and assent form, as applicable per institutional standards) and recruitment materials must be maintained by the Investigator and made available for inspection.

The Investigator will submit reports of SAEs as outlined in Section 7.5.7. In addition, the Investigator agrees to submit progress reports to the IRB or IEC per their local reporting requirements, or at least annually and at the conclusion of the study. The reports will be made available to the Sponsor or designee.

Any communications from regulatory agencies in regard to inspections, other studies that impact this protocol or the qualifications of study personnel should be promptly reported to the Sponsor or its designee.

The Investigator is also responsible for providing the IRB with reports of any reportable serious adverse drug reactions from any other study conducted with the investigational drug. The Sponsor or designee will provide this information to the Investigator.

Major changes in this research activity, except those to remove an apparent immediate hazard to the subject/patient, must be reviewed and approved by the Sponsor and the IRB or IEC that approved the study. Amendments to the protocol must be submitted in writing to the Investigator's IRB or IEC and the Regulatory Authority for approval before subjects/patients are enrolled under the amended protocol.

9.1.3. Study Documentation, Confidentiality, and Records Retention

All documentation relating to the study should be retained for the period of time required by applicable local law. If it becomes necessary for the Sponsor, the Sponsor's designee, applicable IRB/IEC, or applicable regulatory authorities to review or audit any documentation relating to the study, the Investigator must permit direct access to all source documents/data. Records will not be destroyed without informing the Sponsor in writing and giving the Sponsor the opportunity to store the records for a longer period of time at the Sponsor's expense.

The Investigator must ensure that the subject's/patients' anonymity will be maintained. On the eCRFs or other documents submitted to the Sponsor or designees, subjects/patients should not be identified by their names, but by the assigned subjects/patient number and initials. If subjects/patient names are included on copies of documents submitted to the Sponsor or designees, the names (except for initials) will be obliterated and the assigned subjects/patient number added to the document. Documents not for submission to the Sponsor (eg, signed ICFs) should be maintained by the Investigator in strict confidence.

The Investigator must treat all of the information related to the study and the compiled data as confidential, whose use is for the purpose of conducting the study. The Sponsor must approve any transfer of information not directly involved in the study.

In compliance with local and/or regional regulations, this clinical study may be registered and study results may be posted on public registries, such as ClinicalTrials.gov.

9.1.4. End of the Study

The end of the study is defined as last patient last visit.

9.1.5. Discontinuation of the Clinical Study

The Sponsor reserves the right to discontinue the study for clinical or administrative reasons at any time. If the site does not recruit at a reasonable rate, the study may be discontinued at that site. Should the study be terminated and/or the site closed for whatever reason, all documentation and study drug pertaining to the study must be returned to the Sponsor or its representative, and the Investigators, IEC/IRB and Regulatory Authorities will be promptly informed of the termination and the reason for the decision. The Investigator should promptly inform the subjects/patients and assure appropriate therapy and follow-up. Subjects/Patients should then be withdrawn from the study.

9.2. Data Quality Control and Quality Assurance

9.2.1. Data Handling

Study data must be recorded on case report forms (paper and/or electronic) provided by the Sponsor or designee on behalf of the Sponsor. Case report forms must be completed only by persons designated by the Investigator. If eCRFs are used, study data must be entered by trained site personnel with access to a valid and secure eCRF system. All data entered into the eCRF must also be available in the source documents. Corrections on paper CRFs must be made so as to not obliterate the original data and must be initialed and dated by the person who made the correction.

9.2.2. Study Monitoring

The clinical monitor, as a representative of the Sponsor, has an obligation to closely follow the study conduct at the site. The monitor will visit the Investigator and clinical study center periodically and will maintain frequent telephone and written contact. The monitor will maintain current personal knowledge of the study through observation, review of study records and source documentation, and discussion of the conduct of the study with the Investigator and staff.

The monitor will review source documents, systems and CRFs to ensure overall quality and completeness of the data and to confirm study procedures are complied with the requirements in the study protocol accurately. The Sponsor, or its designee, will be allowed to conduct site visits to the investigation facilities for the purpose of monitoring any aspect of the study. The Investigator agrees to allow the monitor to inspect the drug storage area, study drug stocks, drug accountability records, subject/patient charts and study source documents, and other records relative to study conduct.

9.2.3. Audits and Inspections

Periodically, the Sponsor or its authorized representatives audit clinical investigative sites as an independent review of core trial processes and documents to determine whether these activities were conducted, and data were recorded, analyzed, and accurately reported according to the protocol, GCP guidelines of the ICH, and any applicable regulatory requirements. A regulatory authority, an IEC or an IRB may visit the site to perform audits or inspections, including source data verification. The Investigator should contact the Sponsor, or its designee, immediately if contacted by a regulatory agency about an inspection.

9.3. Publication Policy

It is intended that after completion of the study, the data are to be submitted for publication in a scientific journal and/or for reporting at a scientific meeting. A copy of any proposed manuscript must be provided and confirmed received at the Sponsor at least 30 days before its submission, and according to any additional publication details in the Investigator Agreement.

10. LIST OF REFERENCES

1. Cochat, P. and G. Rumsby, *Primary hyperoxaluria.* N Engl J Med, 2013. **369**(7): p. 649-58.

2. Hopp, K., et al., *Phenotype-Genotype Correlations and Estimated Carrier Frequencies of Primary Hyperoxaluria.* J Am Soc Nephrol, 2015.

3. Hoppe, B., *Evidence of true genotype-phenotype correlation in primary hyperoxaluria type 1.* Kidney Int, 2010. **77**(5): p. 383-5.

4. Al-Eisa, A.A., M. Samhan, and M. Naseef, *End-stage renal disease in Kuwaiti children: an 8-year experience.* Transplant Proc, 2004. **36**(6): p. 1788-91.

5. Kamoun, A. and R. Lakhoua, *End-stage renal disease of the Tunisian child: epidemiology, etiologies, and outcome.* Pediatr Nephrol, 1996. **10**(4): p. 479-82.

6. Cochat, P., et al., *Primary hyperoxaluria Type 1: indications for screening and guidance for diagnosis and treatment.* Nephrol Dial Transplant, 2012. **27**(5): p. 1729-36.

7. *ICH Harmonized Tripartite Guideline on Genotoxicity Testing and Data Interpretation for Pharmaceuticals Intended for Human Use S2(R1)*.

8. *Guidance for Industry: Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers.* U.S. Department of Health and Human Services Food and Drug Administration Center for Drug Evaluation and Research (CDER) July 2005.

9. Frishberg, Y., et al., *Mutations in HAO1 encoding glycolate oxidase cause isolated glycolic aciduria.* J Med Genet, 2014. **51**(8): p. 526-9.

10. Levey, A.S., et al., *A new equation to estimate glomerular filtration rate.* Ann Intern Med, 2009. **150**(9): p. 604-12.

11. Schwartz, G.J., et al., *New equations to estimate GFR in children with CKD.* J Am Soc Nephrol, 2009. **20**(3): p. 629-37.

12. *The Feinstein Institute for Medical Research. Human Subject Protection Program Guidance Document: Maximum Blood Draw Limits.* http://www.feinsteininstitute.org/wp-content/uploads/2013/02/Maximum-Blood-Draw-Limits.pdf. 2013.

13. Flynn, J.T., et al., *Clinical Practice Guideline for Screening and Management of High Blood Pressure in Children and Adolescents.* Pediatrics, 2017. **140**(3).

11. APPENDICES

11.1. Pharmacokinetic Assessment Time Points

Table 10 contains a detailed schedule for the collection of blood and urine samples for PK analysis for Part A.

Table 10: Pharmacokinetic Time Points for Single-ascending Dose Cohorts in Healthy Subjects (Part A)

Study Day	Protocol Time (hh:mm)	Blood PK	Urine PK	Pooled Urine
Day 1	Predose (within 1 hour of dosing)	X	X	
	00:00 (dose)			X (00:00-04:00)
	00:30 (±2 min)	X		
	01:00 (±5 min)	X		
	02:00 (±5 min)	X		
	04:00 (±10 min)	X		
	04:01 (±10 min)			X (04:01-08:00)
	06:00 (±15 min)	X		
	08:00 (±15 min)	X		
	08:01 (±15 min)			X (08:01-24:00)
Day 2	24:00 (±30 min)	X		

Table 11 contains a detailed schedule for the collection of blood and urine samples for PK analysis for patients dosed monthly in Part B.

Table 11: Pharmacokinetic Time Points for Multiple-ascending Dose Cohorts in Patients with PH1 (Part B – Monthly Dosing)

Study Day	Protocol Time (hh:mm)	Blood PK	Urine PK	Pooled Urine
Day 1	Predose (within 2 hours of dosing)	X	X	
	00:00 (dose)			X (00:00-04:00)
	00:30 (±2 min)	X		
	02:00 (±5 min)	X		
	04:00 (±10 min)	X		
	04:01 (±10 min)			X (04:01-08:00)
	06:00 (±15 min)	X		
	08:00 (±15 min)	X		
	08:01 (±15 min)			X (08:01-12:00)
	12:00 (±15 min)	X		
	12:01 (±15 min)			X (12:01-24:00)
Day 2	24:00 (±30 min)	X		
Day 3	48:00 (±30 min)	X		
Day 15	Anytime during visit	X		
Day 29	Predose (within 2 hours of dosing)	X	X	
	00:00 (dose)			
	02:00 (±5 min)	X		
Day 57	Predose (within 2 hours of dosing)	X	X	
	00:00 (dose)			X (00:00-04:00)
	00:30 (±2 min)	X		
	02:00 (±5 min)	X		
	04:00 (±10 min)	X		
	04:01 (±10 min)			X (04:01-08:00)
	06:00 (±15 min)	X		
	08:00 (±15 min)	X		
	08:01 (±15 min)			X (08:01-12:00)
	12:00 (±15 min)	X		

Table 11: Pharmacokinetic Time Points for Multiple-ascending Dose Cohorts in Patients with PH1 (Part B – Monthly Dosing)

Study Day	Protocol Time (hh:mm)	Blood PK	Urine PK	Pooled Urine
	12:01 (±15 min)			X (12:01-24:00)
Day 58	24:00 (±30 min)	X		
Day 59	48:00 (±30 min)	X		
Day 85[a]	Anytime during visit	X		
Day 85[b]	Predose (within 2 hours of dosing)	X	X	
	00:00 (dose)			X (00:00-04:00)
	00:30 (±2 min)	X		
	02:00 (±5 min)	X		
	04:00 (±10 min)	X		
	04:01 (±10 min)			X (04:01-08:00)
	06:00 (±15 min)	X		
	08:00 (±15 min)	X		
	08:01 (±15 min)			X (08:01-12:00)
	12:00 (±15 min)	X		
	12:01 (±15 min)			X (12:01-24:00)
Day 86[b]	24:00 (±30 min)	X		
Day 87[b]	48:00 (±30 min)	X		
Day 99[b]	Anytime during visit	X		
Day 113[b]	Predose (within 2 hours of dosing)	X	X	
	00:00 (dose)			
	02:00 (±5 min)	X		
Day 141[b]	Predose (within 2 hours of dosing)	X	X	
	00:00 (dose)			X (00:00-04:00)
	00:30 (±2 min)	X		
	02:00 (±5 min)	X		
	04:00 (±10 min)	X		
	04:01 (±10 min)			X (04:01-08:00)
	06:00 (±15 min)	X		
	08:00 (±15 min)	X		

Table 11: Pharmacokinetic Time Points for Multiple-ascending Dose Cohorts in Patients with PH1 (Part B – Monthly Dosing)

Study Day	Protocol Time (hh:mm)	Blood PK	Urine PK	Pooled Urine
	08:01 (±15 min)			X (08:01-12:00)
	12:00 (±15 min)	X		
	12:01 (±15 min)			X (12:01-24:00)
Day 142[b]	24:00 (±30 min)	X		
Day 143[b]	48:00 (±30 min)	X		
Day 169[b]	Anytime during visit	X		

a. For subjects initially randomized to ALN-GO1
b. Collect blood and urine samples from patients who previously received placebo and continue on-study to receive open-label ALN-GO1.

Table 12 contains a detailed schedule for the collection of blood and urine samples for PK analysis for patients dosed quarterly in Part B.

Table 12: Pharmacokinetic Time Points for Patients with PH1 (Part B – Quarterly Dosing)

Study Day	Protocol Time (hh:mm)	Blood PK	Urine PK	Pooled Urine
Day 1	Predose (within 2 hours of dosing)	X	X	
	00:00 (dose)			X (00:00-04:00)
	00:30 (±2 min)	X		
	02:00 (±5 min)	X		
	04:00 (±10 min)	X		
	04:01 (±10 min)			X (04:01-08:00)
	06:00 (±15 min)	X		
	08:00 (±15 min)	X		
	08:01 (±15 min)			X (08:01-12:00)
	12:00 (±15 min)	X		
	12:01 (±15 min)			X (12:01-24:00)
Day 2	24:00 (±30 min)	X		
Day 3	48:00 (±30 min)	X		
Day 15	Anytime during visit	X		
Day 85	Predose (within 2 hours of dosing)	X	X	
	00:00 (dose)			X (00:00-04:00)
	00:30 (±2 min)	X		
	02:00 (±5 min)	X		
	04:00 (±10 min)	X		
	04:01 (±10 min)			X (04:01-08:00)
	06:00 (±15 min)	X		
	08:00 (±15 min)	X		
	08:01 (±15 min)			X (08:01-12:00)
	12:00 (±15 min)	X		
	12:01 (±15 min)			X (12:01-24:00)
Day 86	24:00 (±30 min)	X		
Day 87	48:00 (±30 min)	X		

Table 12: Pharmacokinetic Time Points for Patients with PH1 (Part B – Quarterly Dosing)

Study Day	Protocol Time (hh:mm)	Blood PK	Urine PK	Pooled Urine
Day 99	Anytime during visit	X		
Day 113	Anytime during visit	X		

11.2. Formulae for Estimated Glomerular Filtration Rate Calculation

Estimated glomerular filtration rate (eGFR; in mL/min/1.73m^2) will be calculated from serum creatinine (SCr) based on the Modification of Diet in Renal Disease formula for patients ≥18 years of age and the Schwartz Bedside Formula for patients <18 years of age.

Modification of Diet in Renal Disease Formula [10]

- Conventional units
  - eGFR (mL/min/1.73m^2) = 175 × (S_{Cr} [mg/dL])$^{-1.154}$ × (age)$^{-0.203}$ × (0.742, if female), or × (1.212, if African American)
- SI units
  - eGFR (mL/min/1.73m^2) = 175 × (SCr [µmol/L]/88.4)$^{-1.154}$ × (age)$^{-0.203}$ × (0.742, if female), or × (1.212, if African American)

Schwartz Bedside Formula [11]

- Conventional units
  - eGFR (mL/min/1.73 m^2) = (0.413 × height [cm])/Scr (mg/dL)
- SI units
  - eGFR (mL/min/1.73 m^2) = (36.2 × height [cm])/Scr (µmol/L)

11.3. Blood Volume Limits in Pediatric Patients (Part B)

The maximum blood volume, which will be collected from pediatric patients in Part B over the course of the study, will be based on age and weight and will not exceed those specified in Table 13, which was adapted from the Human Subject Protection Program Guidance Document.

Table 13: Maximum Allowable Total Blood Collection Volumes Chart (Part B)

Body Weight	Body Weight	Total blood volume	Maximum allowable volume in a 24 hour period		Total volume collected in a 30-day period	
			2.5% of total blood volume	3% of total blood volume	5% of total blood volume	10% of total blood volume
(kg)	(lbs)	(mL)	(mL)	(mL)	(mL)	(mL)
1	2.2	100	2.5	3	5	10
2	4.4	200	5	6	10	20
3	6.6	240	6	7.2	12	24
4	8.8	320	8	9.6	16	32
5	11	400	10	12	20	40
6	13.2	480	12	14.4	24	48
7	15.4	560	14	16.8	28	56
8	17.6	640	16	19.2	32	64
9	19.8	720	18	21.6	36	72
10	22	800	20	24	40	80
11-15	24-33	880-1200	22-30	26.4-36	44-60	88-120
16-20	35-44	1280-1600	32-40	38.4-48	64-80	128-160
21-25	46-55	1680-2000	42-50	50.4-60	64-100	168-200
26-30	57-66	2080-2400	52-60	62.4-72	104-120	208-240
31-35	68-77	2480-2800	62-70	74.4-84	124-140	248-280
36-40	79-88	2880-3200	72-80	86.4-96	144-160	288-320
41-45	90-99	3280-3600	82-90	98.4-108	164-180	328-3600
46-50	101-110	3680-4000	92-100	110.4-120	184-200	368-400
51-55	112-121	4080-4400	102-110	122.4-132	204-220	408-440
56-60	123-132	4480-4800	112-120	134.4-144	224-240	448-480
61-65	134-143	4880-5200	122-130	146.4-156	244-260	488-520
66-70	145-154	5280-5600	132-140	158.4-168	264-280	528-560
71-75	156-165	5680-6000	142-150	170.4-180	284-300	568-600
76-80	167-176	6080-6400	152-160	182.4-192	304-360	608-640
81-85	178-187	6480-6800	162-170	194.4-204	324-340	648-680
86-90	189-198	6880-7200	172-180	206.4-216	344-360	688-720
91-95	200-209	7280-7600	182-190	218.4-228	364-380	728-760
96-100	211-220	7680-8000	192-200	230.4-240	384-400	768-800

Adapted from http://www.feinsteininstitute.org/wp-content/uploads/2013/03/Recruitment-Methods-for-Clinical-Research-Studies.pdf.[12]

11.4. Normative Pediatric Blood Pressure Tables

For pediatric patients <18 years old, study entry criteria for blood pressure will be determined according to normative blood pressure tables based on normal-weight children included in published guidelines issued by the American Academy of Pediatrics [13] (see Table 14 and Table 15). Patients will be excluded if diastolic and/or systolic blood pressure is equal to or greater than the 95th percentile for sex, age, and height (mean of screening height measurements).

Table 14: Blood Pressure Levels for Girls by Age and Height Percentile

Age (y)	BP Percentile	SBP (mm Hg)							DBP (mm Hg)						
		Height Percentile or Measured Height							Height Percentile or Measured Height						
		5%	10%	25%	50%	75%	90%	95%	5%	10%	25%	50%	75%	90%	95%
1	Height (in)	29.7	30.2	30.9	31.8	32.7	33.4	33.9	29.7	30.2	30.9	31.8	32.7	33.4	33.9
	Height (cm)	75.4	76.6	78.6	80.8	83	84.9	86.1	75.4	76.6	78.6	80.8	83	84.9	86.1
	50th	84	85	86	86	87	88	88	41	42	42	43	44	45	46
	90th	98	99	99	100	101	102	102	54	55	56	56	57	58	58
	95th	101	102	102	103	104	105	105	59	59	60	60	61	62	62
	95th + 12 mm Hg	113	114	114	115	116	117	117	71	71	72	72	73	74	74
2	Height (in)	33.4	34	34.9	35.9	36.9	37.8	38.4	33.4	34	34.9	35.9	36.9	37.8	38.4
	Height (cm)	84.9	86.3	88.6	91.1	93.7	96	97.4	84.9	86.3	88.6	91.1	93.7	96	97.4
	50th	87	87	88	89	90	91	91	45	46	47	48	49	50	51
	90th	101	101	102	103	104	105	106	58	58	59	60	61	62	62
	95th	104	105	106	106	107	108	109	62	63	63	64	65	66	66
	95th + 12 mm Hg	116	117	118	118	119	120	121	74	75	75	76	77	78	78
3	Height (in)	35.8	36.4	37.3	38.4	39.6	40.6	41.2	35.8	36.4	37.3	38.4	39.6	40.6	41.2
	Height (cm)	91	92.4	94.9	97.6	100.5	103.1	104.6	91	92.4	94.9	97.6	100.5	103.1	104.6
	50th	88	89	89	90	91	92	93	48	48	49	50	51	53	53
	90th	102	103	104	104	105	106	107	60	61	61	62	63	64	65
	95th	106	106	107	108	109	110	110	64	65	65	66	67	68	69
	95th + 12 mm Hg	118	118	119	120	121	122	122	76	77	77	78	79	80	81
4	Height (in)	38.3	38.9	39.9	41.1	42.4	43.5	44.2	38.3	38.9	39.9	41.1	42.4	43.5	44.2
	Height (cm)	97.2	98.8	101.4	104.5	107.6	110.5	112.2	97.2	98.8	101.4	104.5	107.6	110.5	112.2
	50th	89	90	91	92	93	94	94	50	51	51	53	54	55	55
	90th	103	104	105	106	107	108	108	62	63	64	65	66	67	67
	95th	107	108	109	109	110	111	112	66	67	68	69	70	70	71
	95th + 12 mm Hg	119	120	121	121	122	123	124	78	79	80	81	82	82	83
5	Height (in)	40.8	41.5	42.6	43.9	45.2	46.5	47.3	40.8	41.5	42.6	43.9	45.2	46.5	47.3
	Height (cm)	103.6	105.3	108.2	111.5	114.9	118.1	120	103.6	105.3	108.2	111.5	114.9	118.1	120
	50th	90	91	92	93	94	95	96	52	52	53	55	56	57	57
	90th	104	105	106	107	108	109	110	64	65	66	67	68	69	70
	95th	108	109	109	110	111	112	113	68	69	70	71	72	73	73
	95th + 12 mm Hg	120	121	121	122	123	124	125	80	81	82	83	84	85	85
6	Height (in)	43.3	44	45.2	46.6	48.1	49.4	50.3	43.3	44	45.2	46.6	48.1	49.4	50.3
	Height (cm)	110	111.8	114.9	118.4	122.1	125.6	127.7	110	111.8	114.9	118.4	122.1	125.6	127.7
	50th	92	92	93	94	96	97	97	54	54	55	56	57	58	59
	90th	105	106	107	108	109	110	111	67	67	68	69	70	71	71
	95th	109	109	110	111	112	113	114	70	71	72	72	73	74	74
	95th + 12 mm Hg	121	121	122	123	124	125	126	82	83	84	84	85	86	86
7	Height (in)	45.6	46.4	47.7	49.2	50.7	52.1	53	45.6	46.4	47.7	49.2	50.7	52.1	53
	Height (cm)	115.9	117.8	121.1	124.9	128.8	132.5	134.7	115.9	117.8	121.1	124.9	128.8	132.5	134.7
	50th	92	93	94	95	97	98	99	55	55	56	57	58	59	60
	90th	106	106	107	109	110	111	112	68	68	69	70	71	72	72
	95th	109	110	111	112	113	114	115	72	72	73	73	74	74	75
	95th + 12 mm Hg	121	122	123	124	125	126	127	84	84	85	85	86	86	87

Age (y)	BP Percentile	SBP (mm Hg)							DBP (mm Hg)						
		Height Percentile or Measured Height							Height Percentile or Measured Height						
		5%	10%	25%	50%	75%	90%	95%	5%	10%	25%	50%	75%	90%	95%
8	Height (in)	47.6	48.4	49.8	51.4	53	54.5	55.5	47.6	48.4	49.8	51.4	53	54.5	55.5
	Height (cm)	121	123	126.5	130.6	134.7	138.5	140.9	121	123	126.5	130.6	134.7	138.5	140.9
	50th	93	94	95	97	98	99	100	56	56	57	59	60	61	61
	90th	107	107	108	110	111	112	113	69	70	71	72	72	73	73
	95th	110	111	112	113	115	116	117	72	73	74	74	75	75	75
	95th + 12 mm Hg	122	123	124	125	127	128	129	84	85	86	86	87	87	87
9	Height (in)	49.3	50.2	51.7	53.4	55.1	56.7	57.7	49.3	50.2	51.7	53.4	55.1	56.7	57.7
	Height (cm)	125.3	127.6	131.3	135.6	140.1	144.1	146.6	125.3	127.6	131.3	135.6	140.1	144.1	146.6
	50th	95	95	97	98	99	100	101	57	58	59	60	60	61	61
	90th	108	108	109	111	112	113	114	71	71	72	73	73	73	73
	95th	112	112	113	114	116	117	118	74	74	75	75	75	75	75
	95th + 12 mm Hg	124	124	125	126	128	129	130	86	86	87	87	87	87	87
10	Height (in)	51.1	52	53.7	55.5	57.4	59.1	60.2	51.1	52	53.7	55.5	57.4	59.1	60.2
	Height (cm)	129.7	132.2	136.3	141	145.8	150.2	152.8	129.7	132.2	136.3	141	145.8	150.2	152.8
	50th	96	97	98	99	101	102	103	58	59	59	60	61	61	62
	90th	109	110	111	112	113	115	116	72	73	73	73	73	73	73
	95th	113	114	114	116	117	119	120	75	75	76	76	76	76	76
	95th + 12 mm Hg	125	126	126	128	129	131	132	87	87	88	88	88	88	88
11	Height (in)	53.4	54.5	56.2	58.2	60.2	61.9	63	53.4	54.5	56.2	58.2	60.2	61.9	63
	Height (cm)	135.6	138.3	142.8	147.8	152.8	157.3	160	135.6	138.3	142.8	147.8	152.8	157.3	160
	50th	98	99	101	102	104	105	106	60	60	60	61	62	63	64
	90th	111	112	113	114	116	118	120	74	74	74	74	74	75	75
	95th	115	116	117	118	120	123	124	76	77	77	77	77	77	77
	95th + 12 mm Hg	127	128	129	130	132	135	136	88	89	89	89	89	89	89
12	Height (in)	56.2	57.3	59	60.9	62.8	64.5	65.5	56.2	57.3	59	60.9	62.8	64.5	65.5
	Height (cm)	142.8	145.5	149.9	154.8	159.6	163.8	166.4	142.8	145.5	149.9	154.8	159.6	163.8	166.4
	50th	102	102	104	105	107	108	108	61	61	61	62	64	65	65
	90th	114	115	116	118	120	122	122	75	75	75	75	76	76	76
	95th	118	119	120	122	124	125	126	78	78	78	78	79	79	79
	95th + 12 mm Hg	130	131	132	134	136	137	138	90	90	90	90	91	91	91
13	Height (in)	58.3	59.3	60.9	62.7	64.5	66.1	67	58.3	59.3	60.9	62.7	64.5	66.1	67
	Height (cm)	148.1	150.6	154.7	159.2	163.7	167.8	170.2	148.1	150.6	154.7	159.2	163.7	167.8	170.2
	50th	104	105	106	107	108	108	109	62	62	63	64	65	65	66
	90th	116	117	119	121	122	123	123	75	75	75	76	76	76	76
	95th	121	122	123	124	126	126	127	79	79	79	79	80	80	81
	95th + 12 mm Hg	133	134	135	136	138	138	139	91	91	91	91	92	92	93
14	Height (in)	59.3	60.2	61.8	63.5	65.2	66.8	67.7	59.3	60.2	61.8	63.5	65.2	66.8	67.7
	Height (cm)	150.6	153	156.9	161.3	165.7	169.7	172.1	150.6	153	156.9	161.3	165.7	169.7	172.1
	50th	105	106	107	108	109	109	109	63	63	64	65	66	66	66
	90th	118	118	120	122	123	123	123	76	76	76	76	77	77	77
	95th	123	123	124	125	126	127	127	80	80	80	80	81	81	82
	95th + 12 mm Hg	135	135	136	137	138	139	139	92	92	92	92	93	93	94

Age (y)	BP Percentile	SBP (mm Hg)							DBP (mm Hg)						
		Height Percentile or Measured Height							Height Percentile or Measured Height						
		5%	10%	25%	50%	75%	90%	95%	5%	10%	25%	50%	75%	90%	95%
15	Height (in)	59.7	60.6	62.2	63.9	65.6	67.2	68.1	59.7	60.6	62.2	63.9	65.6	67.2	68.1
	Height (cm)	151.7	154	157.9	162.3	166.7	170.6	173	151.7	154	157.9	162.3	166.7	170.6	173
	50th	105	106	107	108	109	109	109	64	64	64	65	66	67	67
	90th	118	119	121	122	123	123	124	76	76	76	77	77	78	78
	95th	124	124	125	126	127	127	128	80	80	80	81	82	82	82
	95th + 12 mm Hg	136	136	137	138	139	139	140	92	92	92	93	94	94	94
16	Height (in)	59.9	60.8	62.4	64.1	65.8	67.3	68.3	59.9	60.8	62.4	64.1	65.8	67.3	68.3
	Height (cm)	152.1	154.5	158.4	162.8	167.1	171.1	173.4	152.1	154.5	158.4	162.8	167.1	171.1	173.4
	50th	106	107	108	109	109	110	110	64	64	65	66	66	67	67
	90th	119	120	122	123	124	124	124	76	76	76	77	78	78	78
	95th	124	125	125	127	127	128	128	80	80	80	81	82	82	82
	95th + 12 mm Hg	136	137	137	139	139	140	140	92	92	92	93	94	94	94
17	Height (in)	60.0	60.9	62.5	64.2	65.9	67.4	68.4	60.0	60.9	62.5	64.2	65.9	67.4	68.4
	Height (cm)	152.4	154.7	158.7	163.0	167.4	171.3	173.7	152.4	154.7	158.7	163.0	167.4	171.3	173.7
	50th	107	108	109	110	110	110	111	64	64	65	66	66	66	67
	90th	120	121	123	124	124	125	125	76	76	77	77	78	78	78
	95th	125	125	126	127	128	128	128	80	80	80	81	82	82	82
	95th + 12 mm Hg	137	137	138	139	140	140	140	92	92	92	93	94	94	94

Adapted from Flynn et al. 2017 [13]

Table 15: Blood Pressure Levels for Boys by Age and Height Percentile

Age (y)	BP Percentile	SBP (mm Hg)							DBP (mm Hg)						
		Height Percentile or Measured Height							Height Percentile or Measured Height						
		5%	10%	25%	50%	75%	90%	95%	5%	10%	25%	50%	75%	90%	95%
1	Height (in)	30.4	30.8	31.6	32.4	33.3	34.1	34.6	30.4	30.8	31.6	32.4	33.3	34.1	34.6
	Height (cm)	77.2	78.3	80.2	82.4	84.6	86.7	87.9	77.2	78.3	80.2	82.4	84.6	86.7	87.9
	50th	85	85	86	86	87	88	88	40	40	40	41	41	42	42
	90th	98	99	99	100	100	101	101	52	52	53	53	54	54	54
	95th	102	102	103	103	104	105	105	54	54	55	55	56	57	57
	95th + 12 mm Hg	114	114	115	115	116	117	117	66	66	67	67	68	69	69
2	Height (in)	33.9	34.4	35.3	36.3	37.3	38.2	38.8	33.9	34.4	35.3	36.3	37.3	38.2	38.8
	Height (cm)	86.1	87.4	89.6	92.1	94.7	97.1	98.5	86.1	87.4	89.6	92.1	94.7	97.1	98.5
	50th	87	87	88	89	89	90	91	43	43	44	44	45	46	46
	90th	100	100	101	102	103	103	104	55	55	56	56	57	58	58
	95th	104	105	105	106	107	107	108	57	58	58	59	60	61	61
	95th + 12 mm Hg	116	117	117	118	119	119	120	69	70	70	71	72	73	73
3	Height (in)	36.4	37	37.9	39	40.1	41.1	41.7	36.4	37	37.9	39	40.1	41.1	41.7
	Height (cm)	92.5	93.9	96.3	99	101.8	104.3	105.8	92.5	93.9	96.3	99	101.8	104.3	105.8
	50th	88	89	89	90	91	92	92	45	46	46	47	48	49	49
	90th	101	102	102	103	104	105	105	58	58	59	59	60	61	61
	95th	106	106	107	107	108	109	109	60	61	61	62	63	64	64
	95th + 12 mm Hg	118	118	119	119	120	121	121	72	73	73	74	75	76	76
4	Height (in)	38.8	39.4	40.5	41.7	42.9	43.9	44.5	38.8	39.4	40.5	41.7	42.9	43.9	44.5
	Height (cm)	98.5	100.2	102.9	105.9	108.9	111.5	113.2	98.5	100.2	102.9	105.9	108.9	111.5	113.2
	50th	90	90	91	92	93	94	94	48	49	49	50	51	52	52
	90th	102	103	104	105	105	106	107	60	61	62	62	63	64	64
	95th	107	107	108	108	109	110	110	63	64	65	66	67	67	68
	95th + 12 mm Hg	119	119	120	120	121	122	122	75	76	77	78	79	79	80
5	Height (in)	41.1	41.8	43.0	44.3	45.5	46.7	47.4	41.1	41.8	43.0	44.3	45.5	46.7	47.4
	Height (cm)	104.4	106.2	109.1	112.4	115.7	118.6	120.3	104.4	106.2	109.1	112.4	115.7	118.6	120.3
	50th	91	92	93	94	95	96	96	51	51	52	53	54	55	55
	90th	103	104	105	106	107	108	108	63	64	65	65	66	67	67
	95th	107	108	109	109	110	111	112	66	67	68	69	70	70	71
	95th + 12 mm Hg	119	120	121	121	122	123	124	78	79	80	81	82	82	83
6	Height (in)	43.4	44.2	45.4	46.8	48.2	49.4	50.2	43.4	44.2	45.4	46.8	48.2	49.4	50.2
	Height (cm)	110.3	112.2	115.3	118.9	122.4	125.6	127.5	110.3	112.2	115.3	118.9	122.4	125.6	127.5
	50th	93	93	94	95	96	97	98	54	54	55	56	57	57	58
	90th	105	105	106	107	109	110	110	66	66	67	68	68	69	69
	95th	108	109	110	111	112	113	114	69	70	70	71	72	72	73
	95th + 12 mm Hg	120	121	122	123	124	125	126	81	82	82	83	84	84	85
7	Height (in)	45.7	46.5	47.8	49.3	50.8	52.1	52.9	45.7	46.5	47.8	49.3	50.8	52.1	52.9
	Height (cm)	116.1	118	121.4	125.1	128.9	132.4	134.5	116.1	118	121.4	125.1	128.9	132.4	134.5
	50th	94	94	95	97	98	98	99	56	56	57	58	58	59	59
	90th	106	107	108	109	110	111	111	68	68	69	70	70	71	71
	95th	110	110	111	112	114	115	116	71	71	72	73	73	74	74
	95th + 12 mm Hg	122	122	123	124	126	127	128	83	83	84	85	85	86	86

Age (y)	BP Percentile	SBP (mm Hg)							DBP (mm Hg)						
		Height Percentile or Measured Height							Height Percentile or Measured Height						
		5%	10%	25%	50%	75%	90%	95%	5%	10%	25%	50%	75%	90%	95%
8	Height (in)	47.8	48.6	50	51.6	53.2	54.6	55.5	47.8	48.6	50	51.6	53.2	54.6	55.5
	Height (cm)	121.4	123.5	127	131	135.1	138.8	141	121.4	123.5	127	131	135.1	138.8	141
	50th	95	96	97	98	99	99	100	57	57	58	59	59	60	60
	90th	107	108	109	110	111	112	112	69	70	70	71	72	72	73
	95th	111	112	112	114	115	116	117	72	73	73	74	75	75	75
	95th + 12 mm Hg	123	124	124	126	127	128	129	84	85	85	86	87	87	87
9	Height (in)	49.6	50.5	52	53.7	55.4	56.9	57.9	49.6	50.5	52	53.7	55.4	56.9	57.9
	Height (cm)	126	128.3	132.1	136.3	140.7	144.7	147.1	126	128.3	132.1	136.3	140.7	144.7	147.1
	50th	96	97	98	99	100	101	101	57	58	59	60	61	62	62
	90th	107	108	109	110	112	113	114	70	71	72	73	74	74	74
	95th	112	112	113	115	116	118	119	74	74	75	76	76	77	77
	95th + 12 mm Hg	124	124	125	127	128	130	131	86	86	87	88	88	89	89
10	Height (in)	51.3	52.2	53.8	55.6	57.4	59.1	60.1	51.3	52.2	53.8	55.6	57.4	59.1	60.1
	Height (cm)	130.2	132.7	136.7	141.3	145.9	150.1	152.7	130.2	132.7	136.7	141.3	145.9	150.1	152.7
	50th	97	98	99	100	101	102	103	59	60	61	62	63	63	64
	90th	108	109	111	112	113	115	116	72	73	74	74	75	75	76
	95th	112	113	114	116	118	120	121	76	76	77	77	78	78	78
	95th + 12 mm Hg	124	125	126	128	130	132	133	88	88	89	89	90	90	90
11	Height (in)	53	54	55.7	57.6	59.6	61.3	62.4	53	54	55.7	57.6	59.6	61.3	62.4
	Height (cm)	134.7	137.3	141.5	146.4	151.3	155.8	158.6	134.7	137.3	141.5	146.4	151.3	155.8	158.6
	50th	99	99	101	102	103	104	106	61	61	62	63	63	63	63
	90th	110	111	112	114	116	117	118	74	74	75	75	75	76	76
	95th	114	114	116	118	120	123	124	77	78	78	78	78	78	78
	95th + 12 mm Hg	126	126	128	130	132	135	136	89	90	90	90	90	90	90
12	Height (in)	55.2	56.3	58.1	60.1	62.2	64	65.2	55.2	56.3	58.1	60.1	62.2	64	65.2
	Height (cm)	140.3	143	147.5	152.7	157.9	162.6	165.5	140.3	143	147.5	152.7	157.9	162.6	165.5
	50th	101	101	102	104	106	108	109	61	62	62	62	62	63	63
	90th	113	114	115	117	119	121	122	75	75	75	75	75	76	76
	95th	116	117	118	121	124	126	128	78	78	78	78	78	79	79
	95th + 12 mm Hg	128	129	130	133	136	138	140	90	90	90	90	90	91	91
13	Height (in)	57.9	59.1	61	63.1	65.2	67.1	68.3	57.9	59.1	61	63.1	65.2	67.1	68.3
	Height (cm)	147	150	154.9	160.3	165.7	170.5	173.4	147	150	154.9	160.3	165.7	170.5	173.4
	50th	103	104	105	108	110	111	112	61	60	61	62	63	64	65
	90th	115	116	118	121	124	126	126	74	74	74	75	76	77	77
	95th	119	120	122	125	128	130	131	78	78	78	78	80	81	81
	95th + 12 mm Hg	131	132	134	137	140	142	143	90	90	90	90	92	93	93
14	Height (in)	60.6	61.8	63.8	65.9	68.0	69.8	70.9	60.6	61.8	63.8	65.9	68.0	69.8	70.9
	Height (cm)	153.8	156.9	162	167.5	172.7	177.4	180.1	153.8	156.9	162	167.5	172.7	177.4	180.1
	50th	105	106	109	111	112	113	113	60	60	62	64	65	66	67
	90th	119	120	123	126	127	128	129	74	74	75	77	78	79	80
	95th	123	125	127	130	132	133	134	77	78	79	81	82	83	84
	95th + 12 mm Hg	135	137	139	142	144	145	146	89	90	91	93	94	95	96

Age (y)	BP Percentile	SBP (mm Hg)							DBP (mm Hg)						
		Height Percentile or Measured Height							Height Percentile or Measured Height						
		5%	10%	25%	50%	75%	90%	95%	5%	10%	25%	50%	75%	90%	95%
15	Height (in)	62.6	63.8	65.7	67.8	69.8	71.5	72.5	62.6	63.8	65.7	67.8	69.8	71.5	72.5
	Height (cm)	159	162	166.9	172.2	177.2	181.6	184.2	159	162	166.9	172.2	177.2	181.6	184.2
	50th	108	110	112	113	114	114	114	61	62	64	65	66	67	68
	90th	123	124	126	128	129	130	130	75	76	78	79	80	81	81
	95th	127	129	131	132	134	135	135	78	79	81	83	84	85	85
	95th + 12 mm Hg	139	141	143	144	146	147	147	90	91	93	95	96	97	97
16	Height (in)	63.8	64.9	66.8	68.8	70.7	72.4	73.4	63.8	64.9	66.8	68.8	70.7	72.4	73.4
	Height (cm)	162.1	165	169.6	174.6	179.5	183.8	186.4	162.1	165	169.6	174.6	179.5	183.8	186.4
	50th	111	112	114	115	115	116	116	63	64	66	67	68	69	69
	90th	126	127	128	129	131	131	132	77	78	79	80	81	82	82
	95th	130	131	133	134	135	136	137	80	81	83	84	85	86	86
	95th + 12 mm Hg	142	143	145	146	147	148	149	92	93	95	96	97	98	98
17	Height (in)	64.5	65.5	67.3	69.2	71.1	72.8	73.8	64.5	65.5	67.3	69.2	71.1	72.8	73.8
	Height (cm)	163.8	166.5	170.9	175.8	180.7	184.9	187.5	163.8	166.5	170.9	175.8	180.7	184.9	187.5
	50th	114	115	116	117	117	118	118	65	66	67	68	69	70	70
	90th	128	129	130	131	132	133	134	78	79	80	81	82	82	83
	95th	132	133	134	135	137	138	138	81	82	84	85	86	86	87
	95th + 12 mm Hg	144	145	146	147	149	150	150	93	94	96	97	98	98	99

Adapted from Flynn et al. 2017 [13]

ALN-GO1-001 Protocol Amendment 1

Summary of Changes dated 01 July 2016

Compared to the Original Protocol dated 18 December 2015

A Phase 1/2, Single-Blind, Placebo-Controlled, Single- and Multiple-Ascending Dose Safety, Tolerability, Pharmacokinetic and Pharmacodynamics Study of Subcutaneously Administered ALN-GO1 in Healthy Adult Subjects, and Patients with Primary Hyperoxaluria Type 1

Rationale for Protocol Amendment

The primary purpose of this amendment is to provide the starting dose of ALN-GO1 for administration to patients with primary hyperoxaluria type 1 in Part B of the study, which is based on preliminary data from Part A of the study in healthy subjects. The Part B starting dose is 1.0 mg/kg, administered every 28 days, the lowest dose determined to have a pharmacological effect in healthy subjects in Part A that was also considered well-tolerated.

Further, in response to feedback from Regulatory Agencies, study cohort progression/escalation and suspension/stopping rules have been included for Part B. These rules apply to adverse events (AEs) considered likely to be related to the underlying disease and for AEs considered possibly or definitely related to study drug. Common Terminology Criteria for Adverse Events criteria will continue to be used to grade AEs. The revised dose escalation rules will be specified to take into consideration the severity of the AEs observed and the frequency of the occurrence of these toxicities within a given cohort in order to authorize dosing in the next cohort (as outlined in Table 1 below). These rules will guide Part B dose escalation and stopping decisions for the cohort by the Safety Review Committee.

The following changes have been made in Part B to streamline and simplify blood sample collections and study evaluations:

- Clarified that the exploratory endpoint [redacted]
- Clarified symptom-directed exam specifications and that symptom-directed physical examinations occur during the study, except during Screening and on the last postdose follow-up visit when a full physical examination occurs
- [redacted] clarified that eligibility requires a 24-hour urine sample collection [redacted]
- Removed requirement for an inpatient stay at the clinical study center for 24-hour urine sample collection, added a 6 hour window for sample collection after study eligibility is confirmed, and clarified the start day for sample collections
- Removed blood and urine sample collection for pharmacokinetic (PK) analyses on Day 141 for patients initially randomized to ALN-GO1 as it is not expected that there will be measureable drug concentrations at this time point postdose

- Removed the Day 2 blood sample collection for pharmacodynamic (PD) analyses as it is not expected that there will be an evident PD effect this soon postdosing
- Added blood and urine sample collections for exploratory [redacted] for patients initially randomized to placebo
- Changed the day on which patients will be unblinded to Day 78, rather than Day 85, in order for patients and their families to be better prepared for the transition to receive ALN-GO1 if initially randomized to placebo
- Specified the screening blood pressure criteria for eligibility for patients 6 to18 years, inclusive, accounting for blood pressure cut-off values for hypertension in children
- Specified that clinical laboratory assessments of liver function tests and serum creatinine will be reviewed locally before study drug administration
- Specified that on days when a blood sample for vitamin B6 should be collected, patients should be instructed to not take vitamin B6 before the blood sample is collected and the study drug is administered
- Removed the 28 day window for the Day 85 study visit indicating that this visit must occur on the scheduled day
- Clarified that additional cohorts may be enrolled at higher, lower, or intermediate dose levels, but will not exceed the maximum administered dose of 6.0 mg/kg
- Updated the procedure for recording serious adverse events (SAEs) in the case report form, with an SAE form as back-up

The following changes have been applied to Part A and Part B to simplify the study:

- Specified that plasma samples, in addition to urine samples, will be collected for assessment of PD parameters
- Increased the overall study duration from 1 year and 8 months to 2 years
- Clarified that the relationship of AEs to study drug will be classified as definitely, possible, unlikely, or not related to study drug; AEs will not be classified as probably related

Additional administrative changes include the following: aligned footnotes in the Schedules of Assessments for Part B (Table 2 and Table 3); moved the blood sample for antidrug antibody analysis from Day -1 to Day 1, and added a blood sample for antidrug antibody analysis at Day 57, for consistency with Part A; added windows for postdose electrocardiograms in Part B; and corrected the time range for pooled urine sample collection for PK analysis on Day 113 and Day 141 from 4-6 hours to 0-4 hours in Part B.

A detailed summary of changes is provided in Table 1. Corrections to typographical errors, punctuation, grammar, abbreviations, and formatting are not detailed. Additionally, updates to the protocol as described in administrative change letters, dated 05 May 2016; 22 February 2016; and 24 February 2016, are not detailed.

Table 1: Protocol Amendment 1 Detailed Summary of Changes

The primary section(s) of the protocol affected by the changes in the amendment are indicated. The corresponding text has been revised throughout the protocol. Deleted text is indicated by ~~strikeout~~; added text is indicated by **bold** font.

Purpose: Provided the starting dose of ALN-GO1 with dose rationale for Part B of the study and that additional cohorts may be enrolled at higher, lower, or intermediate dose levels, but will not exceed the maximum administered dose of 6.0 mg/kg

The primary change occurs in Section 1.4 Dose Rationale

Added: **Preliminary data from Part A suggest that ALN-GO1 is well-tolerated and support selection of the starting dose for the first cohort of Part B of study ALN-GO1-001. The Part B starting dose is 1.0 mg/kg, administered every 28 days, the lowest dose determined to have a pharmacological effect in healthy subjects in Part A that was also considered well-tolerated. The starting dose in Part B is based on data derived from Part A and recommended by the Safety Review Committee (SRC). Additional cohorts may be enrolled at higher, lower, or intermediate dose levels, but will not exceed the maximum administered dose of 6 mg/kg, and will follow the protocol-specified dose escalation criteria.**

Section(s) also containing this change:

- Synopsis
- Section 4.7.2 Study Drug Dosing, Study Progression, and Dose Escalation in Part B

Purpose: Specified that plasma samples, in addition to urine samples, will be collected for assessment of PD parameters

The primary change occurs in Section 3.2, Secondary Endpoints

Now reads:
- **Plasma and** urine glycolate concentration

Section(s) also containing this change:

- Synopsis
- Section 1.3, Study Design Rationale
- Section 7.2, Pharmacodynamic Assessments

Purpose: Clarified that the exploratory endpoint [redacted]

- [redacted]

Section(s) also containing this change:

- Synopsis
- Section 7.2, Pharmacodynamic Assessments

Purpose: Increased the overall study duration from 1 year and 8 months to 2 years

The primary change occurs in Section 4.2, Duration of Treatment and Overall Duration of Study

Now reads: The overall duration of the study is estimated to be ~~1 year and 8 months~~ **2 years**, including enrollment.

Section(s) also containing this change:

- Synopsis

Purpose: Changed the day on which patients will be unblinded to Day 78 (from Day 85)

The primary change occurs in Section 4.5 Blinding

Now reads: This is a single-blind, placebo-controlled study; therefore, only the study subjects/patients will be blinded to treatment assignment~~ (up to Day 85 for Part B)~~**. Patients in Part B will be unblinded on or after Day 78 in order for patients and their families to be better prepared for the transition to receive ALN-GO1 if initially randomized to placebo**.

Section(s) also containing this change:

- Synopsis
- Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)
- Section 4.1.2 Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

Purpose: Clarified that the relationship of AEs to study drug will be classified as definitely, possible, unlikely, or not related to study drug

The primary change occurs in Section 4.7.3.1 Cohort Progression/Escalation and Suspension/Stopping Rules in Part A

Now reads: Study cohort progression/escalation and suspension/stopping rules for AEs considered possibly~~, probably,~~ or definitely related to study drug for Part A are described in Table 5. If a subject meets a stopping rule, the subject will be managed as clinically indicated and asked to complete all safety assessments according to the protocol.

Section(s) also containing this change:

- Section 4.7.3.2 Individual Patient Progression/Escalation and Suspension/Stopping Rules in Part B

Purpose: Provided study cohort progression/escalation and suspension/stopping rules for Part B have been included in response to feedback from Regulatory Agencies

The primary change occurs in Section 4.7.3.3 Cohort Progression/Escalation and Suspension/Stopping Rules in Part B

Added: **Study cohort progression/escalation and suspension/stopping rules for AEs considered likely to be related to the underlying disease and for AEs considered possibly or definitely related to study drug for Part B are described in Table 7. If a patient meets a stopping rule, the patient will be managed as clinically indicated and asked to complete all safety assessments according to the protocol. For patients with abnormal renal function consistent with their underlying disease at baseline, evidenced by elevated serum creatinine, the change in this parameter from baseline should be used in grading AEs, rather than laboratory normal values.**

Table 7: Cohort Progression/Escalation and Suspension/Stopping Rules for Part B

Adverse Event(s)	Action
If ≥1 patient in a cohort experiences a severe AE and/or SAE, judged to be possibly or definitely related to study drug	**Dosing within that cohort, and in any cohorts receiving a higher dose, will be suspended until SRC review. Following data review, the SRC may permit continuation of any ongoing Part B cohort.** **Any further dose escalation in a subsequent cohort will be suspended. Resumption of dose escalation will require a substantial amendment, approved by the regulatory authority and relevant ethics committee. However, de-escalation to a lower dose, or intermediate but lower dose, may be allowed by the SRC without a protocol amendment.**
If ≥1 patient in Part B experiences a Grade 4 or 5 AE[a] judged to be possibly or definitely related to study drug	**Dosing of all patients in Part B will be suspended until SRC review. Resumption of the study will require a substantial amendment, approved by the regulatory authority and relevant ethics committee.**

Abbreviations: AE=adverse event; SAE=serious adverse event; SRC=Safety Review Committee.
a Common Terminology Criteria for Adverse Events will be used to grade AEs.

Purpose: Specified the screening blood pressure criteria for eligibility for patients 6 to17 years, inclusive

The primary change occurs in Section 5.2.2 Additional Exclusion Criteria for Part B

Added: **For patients aged 6 to 11 years, inclusive, males with systolic blood pressure >115 mmHg and/or a diastolic blood pressure >75 mmHg and females with systolic blood pressure >110 mmHg and/or a diastolic blood pressure >75 mmHg after 10 minutes of rest at screening. For male and female patients aged 12 to 17 years, inclusive, systolic blood pressure >120 mmHg and/or a diastolic blood pressure >80 mmHg after 10 minutes of rest at screening.**

Section(s) also containing this change:

- Section 5.2.1 Exclusion Criteria for Parts A and B

Purpose: [redacted] *clarified that eligibility requires a 24-hour urine sample collection* [redacted]

The primary change occurs in Section 7.1.1.1, Urinary Oxalate Excretion and Creatinine Clearance

Now reads:

~~Urinary oxalate excretion and creatinine clearance will be measured over a 24-hour urine collection period.~~ During the screening period ~~only~~, 24-hour urine collections will be completed ~~in triplicate to assess baseline urinary oxalate excretion and creatinine clearance. The first 2 urine collections may be completed over 2 consecutive 24-hour periods, or separated in time for the convenience of patients. The excretion of oxalate in~~ **at 2 separate time points.** The first **screening 24-hour** urine collection will be used to assess eligibility. ~~Patients will be admitted to the clinical study center on the evening of Day -2 to permit the third,~~**The second screening** 24-hour urine collection ~~of the screening period, which will be completed before dosing on Day 1.Evaluation of oxalate excretion is generally performed on a 24-hour urine collection; however, for patients unable to provide 24-hour urine collection~~ [redacted] ~~may be used to calculate~~**will only be initiated after eligibility is confirmed. A blood sample for serum creatinine must be obtained after each 24-hour urine collection for PD analysis for the calculation of creatinine clearance.**

Patients will be instructed to collect urine samples for the full 24-hour period; however, after confirming eligibility, for patients unable to collect urine samples for the full 24-hour period, urinary oxalate excretion may be calculated from samples collected over an 18 to 24 hour period.

~~After the screening period, assessments are single, 24-hour urine collections (Section Section 7.2)~~**Patients will have the option to bring the 24-hour urine collections to the clinical study center at specified follow-up visits, courier samples to the clinical study center or to the vendor performing analyses, or elect to have the 24-hour urine collected during an inpatient stay at the clinical study center for other assessments.**

Purpose: Removed requirement for an inpatient stay at the clinical study center for 24-hour urine sample collection, added a 6-hour window for sample collection after study eligibility is confirmed, and clarified the start day for sample collections

The primary change occurs in Section 7.2 Pharmacodynamic Assessments

Now reads:

In Part B, **single,** 24-hour urine collections [redacted] will be collected for the analysis of urinary glycolate and oxalate excretion, and creatinine clearance. **The 24-hour urine collection starting on Day -1 must conclude on Day 1 before administration of the first dose of study drug.** Blood samples will **also** be collected for the analysis of

plasma glycolate concentration.

Options for ~~the~~**providing** 24-hour urine collections~~, patients will have the option to bring the urine collection~~ to the clinical study center ~~at specified PD follow-up visits, courier samples to the clinical study center or vendor performing analyses, or have the 24-hour urine collected during an inpatient stay at the clinical study center for other assessments.~~ **are in Section 7.1.1.1.**

Section(s) also containing this change:

- Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)
- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and Will Receive Open-Label ALN-GO1
- Table 8: Adaptive Study Design Areas, Features, and Limits

Purpose: Clarified symptom-directed exam specifications and that symptom directed physical examinations occur during the study, except during Screening and on the last postdose follow-up visit when a full physical examination occurs

The primary change occurs in Section 7.5.3, Physical Examination

Now reads: A symptom-directed physical examination will include ~~chest/respiratory, heart/cardiovascular, dermatological/skin, gastrointestinal/liver, and musculoskeletal/extremities assessments~~**the evaluation of changes in symptoms, or the onset of new symptoms, since the last visit.**

Section(s) also containing this change:

- Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)
- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and Will Receive Open-Label ALN-GO1

Purpose: Removed requirement for an alcohol screening (protocol administrative change letter, dated 24 February 2016)

The primary change occurs in Section 7.5.5.3, Drugs of Abuse and Alcohol Screening

Now reads: ~~An alcohol test (urine and/or breathalyzer) will be performed according to local clinical study center policy at the time points listed in the Schedule of Assessments. If a subject/patient tests positive, they will be excluded from the study.~~

Purpose: Updated the procedure for recording serious adverse events in the case report form, with an SAE form as back-up

The primary change occurs in Section 7.5.6.3 Serious Adverse Events Require Immediate Reporting to Sponsor/Designee

Now reads: To report the SAE, complete the **eCRF. If the EDC system is unavailable, complete the back-up** SAE form. Within 24 hours of receipt of follow-up information, the Investigator must update the SAE form. SAEs must be reported using

the contact information provided in the Study Manual

Section(s) also containing this change:

- Section 7.5.6.2, Eliciting and Recording Adverse Events

Purpose: Removed blood and urine sample collection for pharmacokinetic analyses on Day 141 for patients initially randomized to ALN-GO1

The primary change occurs in Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)

Removed: The "X" from the Day 141 column for the row titled "Blood and urine samples for PK analyses" was deleted.

Purpose: Removed the Day 2 blood sample collection for pharmacodynamic analyses

The primary change occurs in Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)

Removed: The "X" from the Day 2 column for the row titled "Blood sample for PD analyses" was deleted.

Section(s) also containing this change:

- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and Will Receive Open-Label ALN-GO1

Purpose: Specified that clinical laboratory assessments of liver function tests and serum creatinine will be reviewed locally before study drug administration

The primary change occurs in Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)

Now reads: Biochemistry, hematology, coagulation, LFTs, and urinalysis parameters are described in Section 7.5.5. ~~Day -1~~**On Day -1, and on days when study drug is administered, clinical laboratory tests will be analyzed by a local laboratory and confirmed by a central laboratory. Local** clinical laboratory assessment results **for LFT measurements** must be **available and** reviewed **by the Investigator** before study drug administration. **On all other days, clinical laboratory tests will be analyzed by a central laboratory. A blood sample for** serum creatinine must be obtained after ~~the third 24-hour urine collection for PD analysis on Day 1; and after each 24-~~**each 24-**hour urine collection for PD analysis for the calculation of creatinine clearance~~. Clinical laboratory tests will be evaluated by a central laboratory, except for tests performed on Day -1, and all predose LFT and creatinine measurements, which will be locally analyzed and centrally confirmed.~~ **(and before study drug administration, as applicable)**

Section(s) also containing this change:

- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and Will Receive Open-Label ALN-GO1

Purpose: Specified that on days when a blood sample for vitamin B6 should be collected, patients should be instructed to not take vitamin B6 before the blood sample is collected and the study drug is administered

The primary change occurs in Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)

Now reads: **On days when a blood sample for pyridoxine (vitamin B6) will be collected, patients should be instructed not to take vitamin B6 before the blood sample is collected and study drug is administered.**

Section(s) also containing this change:

- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and Will Receive Open-Label ALN-GO1

Purpose: Removed the 28-day window for the Day 85 study visit

The primary change occurs in Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and Will Receive Open-Label ALN-GO1

Removed: The "28-day window" from the Day 85 column for the row titled "Visit Window (D)" was deleted.

Purpose: Added blood and urine sample collections for exploratory [redacted] *for patients initially randomized to placebo*

The primary change occurs in Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and Will Receive Open-Label ALN-GO1

Added: An "X" was added to the row titled [redacted]

Purpose: Corrected typographical errors, punctuation, grammar, abbreviations, and formatting, and incorporated administrative change letters

These changes are not listed individually.

ALN-GO1-001 Protocol Amendment 2

Summary of Changes (dated 21 September 2016) compared to Protocol Amendment 1 (dated 01 July 2016)

A Phase 1/2, Single-Blind, Placebo-Controlled, Single and Multiple Ascending Dose Safety, Tolerability, Pharmacokinetic and Pharmacodynamics Study of Subcutaneously Administered ALN-GO1 in Healthy Adult Subjects, and Patients with Primary Hyperoxaluria Type 1

Rationale for Protocol Amendment

This purpose of this protocol amendment is to address feedback from Regulatory Agencies. Additionally, preliminary nonclinical embryofetal toxicity results (as described in a nonclinical safety report) were recently received, which will be further elucidated in a planned definitive study. These data showed a low incidence of cardiac malformation in low- and mid-dose treated groups but not in high-dose or control groups. Based on the lack of dose response and absence of drug exposure in any fetal tissues, the relationship to test article is uncertain. However, the possibility of a pharmacologically-mediated effect cannot be ruled out, and thus, we have extended the follow-up period and contraception requirements.

Based on these considerations, the following changes are being implemented:

- All patients/subjects will be followed until pharmacodynamic recovery occurs; there will no longer be a maximum follow-up of 180 days. Contraceptive requirements mirror this change such that women of childbearing potential are required to use approved methods of contraception through the end of the follow-up periods until PD recovery occurs. As a result of the extended follow-up periods, estimates of total time on study and overall duration of study have been updated.
- For Part B, blood samples for anti-drug antibody analysis have been added at the final dosing visit/end of treatment visit, at 28 days after the final dose of ALN-GO1, and every 56 days for the remainder of the follow-up periods.
- Language has been added to indicate that confirmed positive ADA samples will be tested for cross-reactivity with DNA and nucleic acids.
- For Part B, pharmacokinetic (PK) sampling times have been added to ensure capture of the full PK profile. In order to reduce burden on patients and reduce total amount of blood sampled, several PK time points have been removed, specifically:
  - Following the first ALN-GO1 dose, the 1-hr time point has been removed and 12-hour, 48-hour, and 15-day time points have been added for blood PK.
  - Following the second ALN-GO1 dose, 24-hour pooled urine will no longer be collected, and all blood PK time points have been removed except for predose and 2-hour postdose.

  - Following the third/final ALN-GO1 dose, the 1 hr time point has been removed and 12-hour, 24-hour, 48-hour, and 15-day time points have been added for blood PK. Pooled urine following the third/final ALN-GO1 dose has been extended such that it will be collected over a 24-hour (rather than 8 hour) period.

- In order to accommodate the additional time points for blood and urine PK in Part B, clinic visits have been added at Day 3, Day 58 and Day 59. For subjects initially receiving placebo, clinic visits have also been added at Day 87, Day 142, and Day 143.
- Text has been modified to clarify that prior to the administration of each dose LFTs (rather than all clinical laboratory tests) will be reviewed locally prior to central laboratory confirmation.
- Text has been added to clarify that patients should begin pregnancy tests after the onset of menarche, if menarche occurs after the screening period during the course of the study.
- An erroneous reference to antiviral efficacy data has been removed.

A detailed summary of changes is provided in Table 1. Corrections to typographical errors, punctuation, grammar, abbreviations, and formatting are not detailed, nor are updates to the protocol described in administrative change letters dated 15 July 2016, 04 August 2016, and 19 September 2016.

Table 1: Protocol Amendment 2 Detailed Summary of Changes

The primary section(s) of the protocol affected by the changes in the protocol amendment are indicated. The corresponding text has been revised throughout the protocol. Deleted text is indicated by ~~strikeout~~; added text is indicated by **bold** font.

Purpose: Follow-up periods have been extended to continue until pharmacodynamic recovery occurs.

The primary change occurs in Table 2 (Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)), footnote a

Now reads: Safety and PD follow-up will continue ~~for~~ **until recovery of both plasma glycolate and urinary oxalate occurs. Plasma glycolate must decrease to a level that is no more than 20% above baseline or to below the upper limit of normal (14 nmol/mL). Urinary oxalate must increase to a level that is above 80% of baseline.**~~up to 180 days after administration of the last dose of ALN-GO1 or placebo (study drug), or until plasma glycolate is within 20% of baseline, whichever duration is shorter~~.

Section(s) also containing this change:

- Synopsis, Duration of Treatment and Overall Duration of Study
- Table 1, footnote a
- Table 2, footnote a
- Table 3, footnote a
- Section 4.1.1 Single-ascending Dose Part in Healthy Adult Subjects (Part A)
- Section 4.1.2 Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)
- Section 4.2 Duration of Treatment and Overall Duration of Study

Purpose: Contraception requirements have been extended to continue through the entire study period.

The primary change occurs in Section 5.1.1 Inclusion Criteria for Parts A and B

Now reads: Women of child-bearing potential must have a negative pregnancy test, cannot be breast feeding, and must be willing to use a highly effective method of contraception 14 days before first dose, **and** throughout study participation~~, and for 90 days after last dose administration~~ **until the completion of the follow-up periods**.

Section(s) also containing this change:

- Section 6.4 Contraceptive Requirements
- Section 7.5.6.6 Pregnancy Reporting

Purpose: ADA collection time points have been added at the final dosing visit/end of treatment visit and during the follow-up period.

The primary change occurs in Table 2 (Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B))

Now reads: Adjustments to Table 2 and 3 have been made to indicate that blood samples for ADA analysis will occur at the final dosing visit/end of treatment visit, at 28 days after the final dose of ALN-GO1, and every 56 days for the remainder of the follow-up periods.

Section(s) also containing this change:

- Table 2 footnote t
- Table 3 footnote q
- Section 7.5.5.1 Immunogenicity

Purpose: To clarify the pregnancy test schedule for patients experiencing menarche after the screening period

The primary change occurs in Table 2 (Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)), footnote i

Added text: Pregnancy tests will be performed for women of childbearing potential only. A serum pregnancy test will be performed at Screening **or after the onset of menarche if the patient was not of childbearing potential at screening**, and urine pregnancy tests will be performed thereafter per the Schedule of Assessments and any time pregnancy is suspected. The results of the pregnancy test must be known before study drug administration. FSH will be measured at Screening only to confirm post-menopausal status.

Section(s) also containing this change:

- Table 3, footnote h
- Section 7.5.5.2 Pregnancy Testing

Purpose: Addition has been made to indicate that cross-reactivity testing will occur for confirmed positive ADA samples

The primary change occurs in Section 7.5.5.1 Immunogenicity

Added text: **Confirmed positive ADA samples will be tested for cross-reactivity with DNA and nucleic acids.**

Purpose: Changed time points for blood and urine PK sampling in Part B

The primary change occurs in Table 10 (Pharmacokinetic Time Points for Multiple-ascending Dose Cohorts in Patients with PH1 (Part B))

Now reads: Adjustments to Table 10 have been made to reflect modifications to blood and urine PK time points.

Section(s) also containing this change:

- Table 2
- Table 3
- Section 7.3 Pharmacokinetic Assessments

Purpose: Added clinic visits in Part B to accommodate additional blood and urine PK sampling

The primary change occurs in Table 2 (Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)) and Table 3 (Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1)

Added text: Columns have been added to Table 2 and Table 3 indicating clinic visits will occur on Day 3, Day 58, Day 59, Day 87, Day 142, and Day 143. "X"s are included in these columns corresponding to the following assessments: symptom-directed physical examination, vital signs, and blood and urine samples for PK analyses.

Section(s) also containing this change:

- Section 4.1.2 Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

Purpose: Updated maximum blood volume estimates for Part B to accommodate additional PK and ADA sampling, and extended follow-up period

The primary change occurs in Section 7.5.5.4 Maximum Blood Volume

Now reads: The maximum blood volume for adult and pediatric patients initially randomized to ALN-GO1 ~~will~~ **is** not **expected to** exceed ~~250~~ **260** mL over the ~~6 month~~ course of the study. The maximum blood volume for adult and pediatric patients initially randomized to placebo ~~will not~~**is not expected to** exceed ~~350~~ **375** mL over the ~~9 month~~ course of the study.

Purpose: The end of study definition has been added.

Section 9.1.4 End of Study has been added to the protocol. Subsequent sections under 9.1 have been consequently renumbered.

Added text: **9.1.4. End of the Study**
The end of the study is defined as last patient last visit.

Purpose: Clarified that prior to the administration of each dose, LFTs (rather than all clinical laboratory tests) will be reviewed locally prior to central laboratory confirmation.

The primary change occurs in Footnote j of Table 2 (Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B))

Now reads: Biochemistry, hematology, coagulation, LFTs, and urinalysis parameters are described in Section 7.5.5. **Clinical laboratory tests will be performed by a central laboratory.** On Day -1, **Day 29, and Day 57,** ~~and on days~~

~~when study drug is administered,~~ ~~clinical laboratory tests~~ **LFTs** will be analyzed by a local laboratory and confirmed by ~~a~~the central laboratory. Local clinical laboratory assessment results for LFT measurements must be available and reviewed by the Investigator before study drug administration. ~~On all other days, clinical laboratory tests will be analyzed by a central laboratory.~~

Section(s) also containing this change:

- Footnote i of Table 3

Purpose: Removed erroneous reference to antiviral efficacy data.

The primary change occurs in Section 4.7 Study Drug Dosing, Study Progression, and Dose Escalation

Now reads: The decision to enroll an optional cohort, extend an existing cohort, and (in the US) to enroll patients under 12 years of age, will be made by the SRC based on available safety, tolerability, and ~~antiviral efficacy~~ **PD** data, if further elucidation of dose response is considered necessary to better understand dose response and/or safety and tolerability.

Purpose: Correct typographical errors, punctuation, grammar, abbreviations, and formatting, and incorporated administrative change letters.

These changes are not listed individually.

ALN-GO1-001 Protocol Amendment 3

Summary of Changes dated 09 December 2016

A Phase 1/2, Single-Blind, Placebo-Controlled, Single and Multiple Ascending Dose Safety, Tolerability, Pharmacokinetic and Pharmacodynamics Study of Subcutaneously Administered ALN GO1 in Healthy Adult Subjects, and Patients with Primary Hyperoxaluria Type 1

Rationale for Protocol Amendment

The primary purpose of this amendment is to further define the cardiac function requirements and monitoring for adult and pediatric patient population being considered for the multiple-ascending dose part (Part B) of study ALN-GO1-001. These updates are based on recommendations from the Safety Review Committee (SRC) and further discussion with primary hyperoxaluria type 1 (PH1) experts on the cardiovascular manifestations of this disease, in particular in patients with severe disease. Preliminary data from the single-ascending dose part (Part A) of this study in healthy adult subjects suggest that ALN-GO1 is well-tolerated at all doses tested; no signs or symptoms of cardiac abnormalities have been reported. The benefit:risk profile for ALN-GO1 remains unchanged.

Alnylam Pharmaceuticals is developing RNA interference (RNAi) therapeutics for multiple clinical indications. Recently, ENDEAVOUR, a Phase 3 study with revusiran, an RNAi therapeutic for the treatment of amyloidosis-related cardiomyopathy in hereditary transthyretin-mediated amyloidosis, was stopped due to an imbalance in mortality between the treatment arm relative to the placebo arm. The cause of death in this study was primarily cardiac in origin, which is consistent with the underlying natural history in this patient population with advanced heart failure (New York Heart Association Class II and III). The root cause of this mortality imbalance remains under investigation.

Following discontinuation of the revusiran program, the SRC for ALN-GO1-001 was convened. The SRC noted that patients with PH1 with systemic oxalosis can also develop infiltrative cardiomyopathy in later stages of the disease process. Since this initial study with ALN-GO1 is restricted to patients without evidence of systemic oxalosis, patients are not expected to have decreased ejection fraction; however, the SRC recommended a cautious approach by adding screening cardiac assessments to exclude patients with compromised cardiac function. The SRC also recommended additional cardiac monitoring for patients enrolled in this study.

The following changes are being implemented in Part B as outlined below:

- Added exclusion criteria for ECHO assessment of left ventricular ejection fraction (LVEF) <55% and troponin I greater than the upper limit of normal (ULN) at screening
- Added ECHO and troponin I assessments
- Specified that electrocardiograms (ECGs) will be read using centralized equipment

- Increased maximum blood volume to align with cardiac monitoring evaluations
- Aligned wording describing the resumption of dosing requirements after a dose suspension rule has been met

A detailed summary of changes is provided in Table 1. Corrections to typographical errors, punctuation, grammar, abbreviations, and formatting are not detailed.

Table 1: Protocol Amendment 3 Detailed Summary of Changes

The primary section(s) of the protocol affected by the changes in the protocol amendment are indicated. The corresponding text has been revised throughout the protocol. Deleted text is indicated by ~~strikeout~~; added text is indicated by **bold** font.

Purpose: Added ECHO assessments

The primary change occurs in Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)

Added text: Added a row for ECHO assessments in the table with 'X' at Screening, Day 85 during the Postdose Follow-up Period, and approximately every 168 days during Safety and PD Follow-up

Section(s) also containing this change:

- Footnote 'i' in Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)
- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1
- Footnote 'h' in Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1
- Section 7.5, Safety Assessments
- Section 7.5.5, Echocardiography (Part B only)
- Section 8.2.7, Safety Analyses

Purpose: Added troponin I assessments

The primary change occurs in Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)

Added text: Added a row for troponin I assessments in the table with 'X' at Screening, Day -1, 29, and 57/EOT during the Dosing Period, and Day 85 during the Postdose Follow-up Period, and approximately every 168 days during Safety and PD Follow-up

Section(s) also containing this change:

- Footnote 'l' in Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)
- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1
- Footnote 'h' in Table 3

- Section 7.5.6, Clinical Laboratory Assessments

Purpose: Specified that ECGs will be read using centralized equipment

The primary change occurs in Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)

Added text: h. All 12-lead ECGs are triplicate**, using centralized equipment.** ~~Triplicate 12-lead~~ ECGs will be measured 5 minutes apart. Recordings will be obtained after the patient has rested comfortably in the supine position for approximately 10 minutes. Patients should remain supine between ECGs. On dosing days, ECGs will be measured within 1 hour predose; and at 1 hour (±20 minutes), 2 hours (±20 minutes), and 4 hours (±20 minutes) postdose. On all other days, ECGs should be collected at approximately the same time of day corresponding to the predose collection (±1 hour).

Section(s) also containing this change:

- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1
- Footnote 'g' in Table 3
- Section 7.5.4, Electrocardiogram

Purpose: Aligned wording describing the resumption of dosing requirements after a dose suspension rule has been met

The primary change occurs in Section 4.7.3, Dose Suspension and Stopping Rules

Now reads: For the purpose of this study, dose suspension and stopping rules are based on toxicity. Standard toxicity grading according to the CTCAE will be used to grade AEs. The term 'suspension' means that no further study drug will be administered at the dose level and that further dose escalation/progression will be suspended. If a suspension/stopping rule is met, there will be no further enrollment or dosing in the current cohort, or escalation to another cohort, in that part of the study and an ad hoc SRC meeting will be held. Following SRC review, dosing may be resumed at the same or higher dose level following approval from the concerned Regulatory Authority and the independent ethics committee (IEC)/institutional review board (IRB) ~~in accordance with applicable requirements~~**, if required by local regulations**. However, de-escalation to a lower dose, or intermediate may be allowed without prior Regulatory Authority or IEC/IRB approval.

Section(s) also containing this change:

- Table 7: Cohort Progression/Escalation and Suspension/Stopping Rules for Part B

Purpose: Added exclusion criteria for ECHO assessment of LVEF and troponin I at screening

The primary change occurs in Section 5.2.2, Additional Exclusion Criteria for Part B

Added text: **22. Echocardiography (ECHO) assessment of abnormal left ventricular systolic function, defined as left ventricular ejection fraction <55% at screening**

23. Troponin I greater than the upper limit of normal (ULN) at screening (abnormal results should be repeated)

Purpose: Increased maximum blood volume to align with additional cardiac monitoring evaluations

The primary change occurs in Section 7.5.6.4, Maximum Blood Volume

Added text: In Part B, the maximum blood volume, which will be collected from pediatric patients over the course of the study, will be based on age and weight and will not exceed those specified in Table 11 from the Feinstein Institute for Medical Research Human Subject Protection Program Guidance Document (Section 11.3 in the Appendix).[13] The maximum blood volume for adult and pediatric patients initially randomized to ALN-GO1 is not expected to exceed ~~260 mL~~ **350 mL** over the course of the study. The maximum blood volume for adult and pediatric patients initially randomized to placebo is not expected to exceed ~~375 mL~~ **450 mL** over the course of the study. The blood volume limits for patients in Part B are based on those for 6 year old girls in the 5th percentile for weight, the smallest patients who may be enrolled in the study.

Purpose: Correct typographical errors, punctuation, grammar, abbreviations, and formatting

These changes are not listed individually.

ALN-GO1-001 Protocol Amendment 4

Summary of Changes (dated 27 June 2017) compared to Protocol Amendment 3 (dated 09 December 2016)

A Phase 1/2, Single-Blind, Placebo-Controlled, Single- and Multiple-Ascending Dose Safety, Tolerability, Pharmacokinetic and Pharmacodynamics Study of Subcutaneously Administered ALN-GO1 in Healthy Adult Subjects, and Patients with Primary Hyperoxaluria Type 1

Rationale for Protocol Amendment

The protocol is being amended to revise the study follow-up period for Part B. In Part B of this study, adults or pediatric patients are randomized to receive multiple doses of ALN-GO1 or placebo. Prior to this amendment, the protocol required that following completion of dosing, patients continue safety and pharmacodynamic (PD) follow-up until urinary oxalate is >80% of baseline and plasma glycolate is <20% above baseline or to below the upper limit of normal.

To allow patients the opportunity to continue to receive potentially beneficial treatment for PH1, a chronic disease with no approved therapies, we are amending the protocol to potentially shorten the duration of follow-up to allow patients in Part B to transition to an open-label extension study earlier, provided that urinary oxalate is above the upper limit of normal and patients meet at least 1 of the following criteria:

- One 24 hour urinary oxalate value is >80% of baseline.
- Two 24 hour urinary oxalate values are above the midpoint between their baseline and nadir 24 hour urinary oxalate values. The nadir must be from a valid collection after all doses are administered.
- At least 12 months have elapsed from time of final dose administration.

Relaxing the threshold for oxalate recovery and capping the duration of safety and PD follow-up potentially avoids a long lapse in treatment administration for patients who wish to continue to receive ALN-GO1 in an open-label extension study. Glycolate recovery is not considered to be required for patients who enroll in the open-label extension study since plasma glycolate and safety data will continue to be monitored in the extension study.

Other changes being introduced in this amendment include an increase in the number of optional cohorts and sample size in Part B in order to allow for further exploration of the optimal dose and dosage regimen to support later stage clinical development. The current protocol allows the option to enroll 2 additional cohorts or to expand a cohort by up to 4 additional patients in Part B. The protocol is being amended to allow for up to 3 optional cohorts in Part B. In addition, this amendment allows the Safety Review Committee (SRC) to permit up to 2 cohorts in Part B to be extended by up to 4 additional patients. The total number of patients in Part B has been increased to up to 24 patients.

A Schedule of Assessments for quarterly dosing in Part B has also been added in this amendment. The current protocol allows the dosing regimen to be determined or adapted in accordance with safety, tolerability and PD data; however, the only schedules of assessments included were for monthly dosing. Preliminary data from Part A in healthy subjects informed the potential option for quarterly dosing in Part B. This amendment specifies the schedule and planned procedures for quarterly dosing in Part B, if implemented.

In addition to these changes, the following has been updated for this study:

- The information in the Benefit-Risk Assessment, which remains positive, was updated to align with the information in the Investigator's Brochure (Edition 2).
- The Statistical Methods section has been updated to account for a quarterly dosing regimen, if implemented.
- Section 4.6 Safety Review Committee was amended to align with recent changes to the SRC charter regarding the frequency of safety data reviews during the study. The SRC charter stipulates that after initiation of dosing in Part B, the SRC will review data approximately every 4 weeks during the dosing and post-dose follow-up periods, or as further decided by the SRC until all subjects and patients have completed their participation in the study as per protocol.
- Clarified that the PK population (analysis set) includes subjects/patients with any evaluable postdose PK data.
- Clarified that Part B is expected to take place at approximately 12 clinical study centers worldwide.
- Clarified that for certain study visits, blood sample collection for exploratory [redacted] is optional for pediatric patients who exceed the maximum blood volume collection limits.

A detailed summary of changes is provided in Table 1. Corrections to typographical errors, punctuation, grammar, abbreviations, and formatting (including administrative changes noted in Protocol Administrative Change #8 (dated 26 April 2017) are not detailed.

Table 1: Protocol Amendment 4 Detailed Summary of Changes

The primary section(s) of the protocol affected by the changes in the protocol amendment are indicated. The corresponding text has been revised throughout the protocol. Deleted text is indicated by ~~strikeout~~; added text is indicated by **bold** font.

Purpose: Add Schedule of Assessments for Quarterly Dosing and associated Pharmocokinetic Time Points table

The primary change occurs in Table 4: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) – Quarterly Dosing, Appendix 11.1 text, and Table 12: Pharmacokinetic Time Points for Patients with PH1 (Part B – Quarterly Dosing)

Section(s) also containing this change:

- Section 7 Study Assessments
- Section 7.3 Pharmacokinetic Assessments

Purpose: Update table titles to clarify monthly vs quarterly dosing schedules

Now reads:

- Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 **(Part B – Monthly Dosing)** ~~(PartB)~~
- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1 **– Monthly Dosing**
- Table 11: Pharmacokinetic Time Points for Multiple-ascending Dose Cohorts in Patients with PH1 (Part B **– Monthly Dosing**)

Purpose: Amend study design to describe quarterly dosing procedures

The primary change occurs in Section 4.1.2. Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

Now reads: Patients will be randomized ~~on~~ **between Day -1 and Day 1** and will receive the first dose of ALN-GO1 or placebo **on Day 1**. The **24- and** 48-hour postdose follow up assessments will take place on Day 2 and Day 3. Patients **who receive study drug monthly** will return to the clinical study center ~~on an outpatient basis~~ for safety, tolerability, PK, and PD monitoring at time points specified in the Schedule of Assessments (**Table 2**) for the remaining 2 single-blind doses of study drug (through Day 57). After completion of the blinded portion of the study, patients **dosed monthly** will be unblinded (on or after Day 78). Patients who initially received placebo will then receive ALN-GO1 at the same dose administered to the cohort into which they were initially randomized and will follow the same assessment schedule ~~with the first dose administered on the new Day 1 (corresponding to study Day 85)~~ **as indicated in Table 3.**

Patients who receive study drug quarterly will return to the clinical study center for safety, tolerability, PK, and PD monitoring at time points specified in the Schedule of Assessments (Table 4) and will receive a 2nd dose of study drug at Day 85. Patients who initially received placebo will receive a single dose of ALN-GO1 on Day 85 at the same dose administered to the cohort into which they were initially randomized. Patients dosed quarterly will be unblinded to initial treatment assignment following completion of the postdose follow-up period

Section(s) also containing this change:

- Synopsis

Purpose: Clarify blinding plan for quarterly dosing cohort(s), if enrolled

The primary change occurs in Section 4.5 Blinding

Added text: This is a single-blind, placebo-controlled study; therefore, only the study subjects/patients will be blinded to treatment assignment. Patients in Part B **dosed monthly** will be unblinded on or after Day 78 in order for patients and their families to be better prepared for the transition to receive ALN-GO1 if initially randomized to placebo. **Patients dosed quarterly will be unblinded to initial treatment assignment following completion of the postdose follow-up period.** The Investigators, Medical Monitors at the Sponsor and CRO, clinical study center personnel, pharmacokineticist, and members of the SRC will have knowledge of the treatment assignment. The clinical study center pharmacy staff will maintain the single-blind according to clinical study center-specific procedures and the Pharmacy Manual. Syringes containing dispensed study drug will be masked in the pharmacy before transfer to the clinic.

Purpose: Update Benefit-Risk Assessment to align with the Investigator's Brochure

The primary change occurs in Section 1.5 Benefit-Risk Assessment

Now reads: ~~Non-specific potential risks~~ **Important potential risks** to healthy subjects and patients include **injection site reactions (ISRs). Other potential risks include: embryofetal** ~~risk~~ **toxicity**, **coagulation abnormalities**, and liver function test abnormalities.

These potential risks have been reduced by the specific inclusion and exclusion criteria incorporated into the selection of healthy subjects as well as for patients with PH1 in this clinical study.

No data are available on the use of ALN-GO1 in pregnancy; however, ~~there is no suspicion of~~ human ~~teratogenicity based on class effects or genotoxic potential~~ **mutagenicity is not suggested based on the available nonclinical data (see Investigator's Brochure for further information)**. Embryofetal risk is limited by requiring that women of childbearing potential (WOCBP) must have a negative pregnancy test, cannot be breast feeding, and must be willing to use a highly effective method of contraception as specified in the protocol. Male subjects/patients are not required to use the contraception measures required for female study subjects/patients. No male contraception is considered to be required.

The occurrence of ISRs will be carefully monitored.

Nonclinical studies in rats showed mild to moderate decreases in fibrinogen and occasionally minimal prolonged prothrombin time without accompanying clinical or microscopic evidence of hemorrhage. This effect is likely species-specific as it was not observed in NHPs and will be monitored via clinical laboratory safety assessments.

Purpose: Increase the number of patients in the study due to the option to enroll up to 1 additional optional cohort or to expand a cohort in Part B

The primary change occurs in Section 8.1 Determination of Sample Size

Now reads: The sample size was based on clinical considerations rather than power calculations. Up to 6~~0~~**4** participants (40 subjects and 2~~0~~**4** patients) are planned to be enrolled in this study, including optional **and expansion** cohorts (see Table 9).

Section(s) also containing this change:

- Synopsis
- Section 4.3 Number of Subjects and Patients

Purpose: Specify criteria for patients who wish to enroll in an open-label extension study following completion of the postdose follow-up period and clarified duration of safety and PD follow up for patients who do not enroll in an open-label extension study.

The primary change occurs in Section 4.1.2. Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

Added text:

After the dosing period, patients will return to the clinical study center for continued safety, tolerability, PK, and PD monitoring through the last postdose follow-up visit. **Following completion of the postdose follow-up period, patients will be invited to participate in an open-label extension study provided that:**

- **Urinary oxalate is above the ULN and patients meet at least 1 of the following criteria:**
  - **One 24-hour urinary oxalate value is >80% of baseline.**
  - **Two 24-hour urinary oxalate values are above the midpoint between their baseline and nadir 24-hour urinary oxalate values. The nadir must be from a valid collection after all doses are administered.**
  - **At least 12 months have elapsed from time of final dose administration.**

For patients who do not enroll in the open-label extension study, safety and PD follow up will continue until:

- **24-hour urinary oxalate is >80% of baseline, AND**
- **Plasma glycolate is <20% above baseline or ≤ the ULN.**

Section(s) also containing this change:

- Synopsis
- Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) – Monthly Dosing
- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1 – Monthly Dosing

Purpose: Increase the number of optional cohorts and expansion cohorts in Part B to allow for further exploration of the optimal dose or dosing regimen.

The primary change occurs in Section 4.1.2 Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

Now reads: Part B is the multiple-ascending dose part of the study in **up to 24** adult and pediatric patients with PH1 with relatively well-preserved renal function. ~~Patients will be enrolled in 1 of 2 ascending dose cohorts.~~ **Two ascending dose cohorts will be enrolled, with the possibility to also enroll up to 3** ~~Two~~ additional optional cohorts ~~may also be enrolled~~ **to further explore the optimal dose or regimen.** Each cohort will be comprised of 4 patients, randomized 3:1 to ALN-GO1 or placebo. ~~Expansion of a cohort in Part B by up to 4 additional patients may also occur~~ **Up to 2 cohorts in Part B may be expanded by up to 4 additional patients** (these patients will all receive ALN-GO1, not placebo).

Section(s) also containing this change:

- Table 9: Adaptive Study Design Areas, Features, and Limits
- Section 4.7.2 Study Drug Dosing, Study Progression, and Dose Escalation in Part B

Purpose: Differentiate monthly vs quarterly drug dosing procedures

The primary change occurs in 4.7.2 Study Drug Dosing, Study Progression, and Dose Escalation in Part B

Now reads: In Part B, all ~~subjects~~ **patients** in a cohort **dosed monthly** will receive the same dose for each of the 3 study drug doses. ~~Subjects~~ **Patients** initially receiving 3 doses of placebo will receive 3 doses of ALN-GO1, with all 3 doses being the same for each of the 3 administrations of ALN-GO1. **Patients in a cohort dosed quarterly who are randomized to ALN-GO1 will receive the same dose for each of the 2 study drug doses. Patients initially receiving 1 dose of placebo on Day 1 will receive 1 dose of ALN-GO1 on study Day 85.** The maximum dose administered will not exceed 6.0 mg/kg.

Dose levels **and/or dosing regimen** in each cohort in Part B may be modified by the SRC. The next higher dose cohort can be enrolled after at least 3 patients in the previous cohort receive their first and second dose and have been followed for at least 14 days following the second dose of study drug. The SRC must review accumulating safety data from both single- and multiple-ascending dose cohorts to confirm the dose level and permit dosing in the next MAD cohort. In Part B, ~~after unblinding,~~ patients randomized to placebo will be administered active ALN-GO1 in an open-label manner according to the same dose and assessment schedule for the cohort to which they were initially assigned.

Purpose: Revise statistical methods to account for quarterly dosing, if implemented

The primary change occurs in Section 8.2 Statistical Methodology

Now reads:

Data from Parts A and B will be analyzed separately. Tabular summaries will be generated by dose level **and dosing regimen** of ALN-GO1 and placebo (pooled across all cohorts) for Part A and Part B (through Day 85).

~~For the blinded portion of Part B, s~~Safety and PD data will be summarized ~~for each cohort~~ by dose level **and dosing regimen** of ALN-GO1 compared to placebo for data collected up to, and including, study Day 85. ~~After receiving 3 single-blind doses of study drug (and completing assessments for that portion of the study), patients initially randomized to placebo will be unblinded and administered 3 doses of active ALN-GO1 in an open-label manner according to the same dose and assessment schedule. Data from the blinded and open-label portion of the study will also be combined to summarize the safety and PD effect of ALN-GO1 at each dose level.~~ **Data from placebo patients from all cohorts will be combined. Data collected after Day 85 will be summarized separately for patients in quarterly dosing cohorts who receive a second dose of ALN-GO1 on Day 85. Additionally, all safety and PD data collected from all patients in Part B during the ALN-GO1 dosing period, regardless of treatment sequence, will be combined and summarized by ALN-GO1 dose level and regimen.**

Data collected beyond the designated dosing period will be summarized or presented in data listings.

Section(s) also containing this change:

- Synopsis

Purpose: Clarify the PK population (analysis set) includes patients/subjects with any evaluable postdose PK data.

The primary change occurs in Section 8.2.1. Populations to be Analyzed

Now reads:

PK Analysis Set: All subjects/patients who receive at least 1 dose of study drug and have at least 1 postdose ~~blood~~ sample **for PK parameters and who have** evaluable ~~for~~ PK ~~parameters~~ **data**.

Purpose: Clarify that Part B is expected to take place at approximately 12 clinical study center worldwide

The primary change occurs in Section 4.1 Summary of Study Design

Now reads:

Part B is expected to take place at ~~up to~~ **approximately** 12 clinical study centers worldwide.

Purpose: Update treatment duration and overall study duration to account for the option for quarterly dosing Part B and also to account for the option to include an additional optional cohort or expansion cohort in PartB.

The primary change occurs in Section 4.2 Duration of Treatment and Overall Duration of Study

Now reads: The duration of treatment is as follows:

- Part A: The estimated total time on study, inclusive of screening, for each subject is up to 405 days. The duration of treatment is a single dose.
- Part B: ~~The estimated total time on study, inclusive of screening and safety and PD follow-up, for each patient initially randomized to receive active study drug is 462 days. The duration of treatment is 57 days. Additionally, the estimated total time on study, inclusive of screening and safety and PD follow-up, for each patient initially randomized to receive placebo, then active study drug, is 546 days. The duration of treatment is 141 days~~
  - **For patients dosed monthly: The duration of treatment for patients initially randomized to receive active study drug is 57 days. The estimated total time on study, inclusive of screening, for each patient is up to 462 days. Additionally, the duration of treatment for patients initially randomized to receive placebo is 141 days. The estimated total time on study, inclusive of screening, for each patient initially randomized to receive placebo, then active study drug, is up to 546 days.**
  - **For patients dosed quarterly: The duration of treatment is 85 days for patients randomized to placebo and active study drug. The estimated total time on study, inclusive of screening, is up to 490 days.**

The overall duration of the study is estimated to be ~~3~~**4** years, including enrollment.

Section(s) also containing this change:

- Synopsis

Purpose: Clarify that triplicate12-lead ECGs will be measured approximately 5 minutes apart.

The primary change occurs in Section 7.5.4 Electrocardiogram

Added text: Triplicate 12-lead ECGs will be measured **approximately** 5 minutes apart.

Section(s) also containing this change:

- Table 1: Schedule of Assessments for Single-ascending Dose Cohorts in Healthy Subjects (Part A)
- Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B – Monthly Dosing)
- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1 – Monthly Dosing

Purpose: Delete text in Schedule of Assessment footnotes providing examples of days when echocardiograms are performed and days when troponin I is measured.

The primary change occurs in footnote i (echocardiogram) and footnote l (troponin I) in Table 2 Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)

Now reads:

i. Echo will be performed during the first visit of the Safety and PD Follow-up Period (Day 169), and thereafter, approximately every 168 days corresponding with visits to the clinical study center ~~(eg, Day 337, Day 505, Day 673)~~ for the duration of the study.

l. During Screening and throughout the Dosing Period, abnormal results for troponin I tests should be repeated. During the Dosing Period only, local clinical laboratory results must be drawn within 4 days prior to dosing and available and reviewed by the Investigator before study drug administration. Troponin I levels will be measured on the first day of the Safety and PD Follow-up Period (Day 169), and thereafter, approximately every 168 days ~~(eg, Day 337, Day 505, Day 673)~~ for the duration of the study.

Section(s) also containing this change:

- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1 footnote h (echocardiogram) and footnote k (troponin I)

Purpose: Clarify that for certain visits, blood samples for exploratory [redacted] are optional for pediatric patients who exceed the maximum blood volume collection limits.

The primary change occurs in Table 2 Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) and Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1

Added text: Footnote **v.** (Table 2) **Blood samples for exploratory[redacted] on Day 57 are optional for pediatric patients who exceed the maximum blood volume collection limits listed in Table 13.**

Footnote **r.** (Table 3) **Blood samples for exploratory[redacted]s on Day 85 are optional for pediatric patients who exceed the maximum blood volume collection limits listed in Table 13.**

Purpose: Amend text describing the frequency of SRC data reviews to align with recent changes to the SRC charter.

The primary change occurs in Section 4.6 Safety Review Committee

Now reads The SRC will undertake safety data review before initiation of dosing in a new cohort in Part A and Part B, and before initiation of dosing in Part B. After initiation of dosing in Part B, ~~the SRC will review data approximately every 4 weeks after administration of the last dose of ALN-GO1 until recovery of both plasma glycolate and urinary oxalate occurs~~ **safety reviews will be conducted in accordance with the SRC charter at a minimum of every 3 months for the duration of the study.**

Purpose: Increase the maximum blood volume over the course of the study in patients initially randomized to ALN-GO1 to account for the quarterly dosing schedule, if implemented

The primary change occurs in Section 7.5.6.4 Maximum Blood Volume

Now reads: The maximum blood volume for adult and pediatric patients initially randomized to ALN-GO1 is not expected to exceed ~~350~~ **400** mL over the course of the study.

Purpose: Remove reference to outpatient visits in Part B since outpatient visits are not required (sites have the option to admit patients)

The primary change occurs in Section 4.1.2 Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

Now reads Patients will return to the clinical study center ~~on an outpatient basis~~ for safety, tolerability, PK, and PD monitoring at time points specified in the Schedule of Assessments for the remaining 2 single-blind doses of study drug (through Day 57).

After the dosing period, patients will return to the clinical study center ~~on an outpatient basis~~ for continued safety, tolerability, PK, and PD monitoring through the last postdose follow-up visit.

Section(s) also containing this change:

- Synopsis

Purpose: Clarify in Table 3 that the calculation of the ALN–GO1 dose to be administered during the open label portion of the study will be based on the body weight obtained on Day 57, not Day 85

The primary change occurs in Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1 footnote e

Now reads: e. The Day ~~85~~ **57** body weight will be used for calculation of the ALN–GO1 dose to be administered during this portion of the study.

Purpose: Simplify text describing study design period

The primary change occurs in Section 4.1.2. Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

Now reads: Patients will be screened **within** ~~from -~~45 ~~to -2~~ days ~~before~~ **prior to** study drug administration. Baseline urinary oxalate excretion and creatinine clearance will be assessed through 24-hour urine collections. ~~The remaining screening and predose assessments will take place on Day -1.~~

Purpose: Add footnote to Figure 1 to indicate that patients in Part B will be invited to participate in an open-label extension study following the postdose follow-up period

The primary change occurs in Figure 1: Study Design

Added text: [a] **Patients in Part B will be invited to participate in an open-label extension study provided they meet the criteria described in Section 4.1.2.**

Purpose: Correct typographical errors, punctuation, grammar, abbreviations, and formatting

These changes are not listed individually.

ALN-GO1-001 Protocol Amendment 5

Summary of Changes (dated 14 February 2018) compared to Protocol Amendment 4 (dated 27 June 2017)

A Phase 1/2, Single-Blind, Placebo-Controlled, Single- and Multiple-Ascending Dose Safety, Tolerability, Pharmacokinetic and Pharmacodynamics Study of Subcutaneously Administered ALN-GO1 in Healthy Adult Subjects, and Patients with Primary Hyperoxaluria Type 1

Rationale for Protocol Amendment

The protocol is being amended to allow patients with primary hyperoxaluria type 1 (PH1) in Part B to more rapidly rollover to an open-label extension study to continue to receive ALN-GO1. This amendment shortens the required follow-up period from up to 1 year to 12 weeks after their last dose of ALN-GO1, without requiring the protocol-defined thresholds for urinary oxalate levels. Data to date indicate that ALN-GO1 has a favorable impact on suppressing urinary oxalate production, the cause of morbidity and mortality in patients with PH1. In addition, the available data have not identified a safety concern of ALN-GO1 nor ALN-GO1-mediated GO1 knockdown. Patients who rollover to the open-label extension study will continue to be monitored for safety and ongoing reviews of safety, tolerability and available study data will be performed by the same Safety Review Committee (SRC) used in this study.

This amendment will also permit the investigator to discontinue safety and pharmacodynamic (PD) follow up for patients who do not enroll in the open-label extension study, and who have not yet met the protocol-specified recovery criteria. This decision must not be made until completion of the required 12-week follow-up period and must be endorsed by the SRC based upon the patient's pharmacodynamic and safety data, as well as emerging data on the safety of ALN-GO1 knockdown. This change will potentially reduce the total burden of study follow-up for some patients while ensuring patient safety.

Another change being introduced in this amendment includes revision of the blood pressure exclusion criteria for pediatric patients in Part B. The definition for uncontrolled hypertension in patients <18 years of age is being redefined from a set cutoff by age to using the American Academy of Pediatric guidelines which account for age, gender and height. This will prevent pediatric patients who are normotensive by these accepted guidelines from being excluded from the study.

In addition to these changes, the following has been updated for this study:

- Extended the allowable time window (from 1 to 2 hours) to conduct predose assessments (ie, vital signs, 12-lead ECGs, physical examinations, blood and urine sample collections) when scheduled at the same time points for Part B.
- Clarified that blood samples for pyridoxine (vitamin B6) are required only for patients receiving therapeutic pyridoxine since they are being measured to monitor adherence to the baseline regimen.

A detailed summary of changes is provided in Table 1. Corrections to typographical errors, punctuation, grammar, abbreviations, and formatting are not detailed.

Table 1: Protocol Amendment 5 Detailed Summary of Changes

The primary section(s) of the protocol affected by the changes in the protocol amendment are indicated. The corresponding text has been revised throughout the protocol. Deleted text is indicated by ~~strikeout~~; added text is indicated by **bold** font.

Purpose: Clarify that blood samples for pyridoxine (vitamin B6) are required only for patients receiving therapeutic pyridoxine.

The primary change occurs in Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B – Monthly Dosing

Now reads: **Blood sample for pyridoxine (vitamin B6) is required only for patients receiving therapeutic pyridoxine.** On days when a blood sample for pyridoxine ~~vitamin B6~~ will be collected, patients should be instructed not to take vitamin B6 before the blood sample is collected and study drug is administered.

Section(s) also containing this change:

- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1 – Monthly Dosing, footnote s
- Table 4: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B – Quarterly Dosing), footnote w

Purpose: Shorten the study follow up criteria for patients in Part B who wish to enroll in an open-label extension study.

The primary change occurs in Section 4.1.2 Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

Now reads: After the dosing period, patients will return to the clinical study center for continued safety, tolerability, PK, and PD monitoring ~~through~~ **for at least 12 weeks (84 days) following the last** ~~postdose follow-up visit~~ **dose of study drug.** Following completion of the **12-week** postdose follow-up period, patients will be invited to participate in an open-label extension study ~~provided that:~~

~~Urinary oxalate is above the ULN and patients meet at least 1 of the following criteria:~~

- ~~One 24-hour urinary oxalate value is >80% of baseline.~~
- ~~Two 24-hour urinary oxalate values are above the midpoint between their baseline and nadir 24-hour urinary oxalate values. The nadir must be from a valid collection after all doses are administered.~~
- ~~At least 12 months have elapsed from time of final dose administration.~~

Section(s) also containing this change:

- Synopsis

Purpose: Clarify that up to 2 cohorts in Part B may be extended by up to 4 patients per cohort.

The primary change occurs in Section 4.1.2 Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (PartB)

Now reads: Based on SRC review of accumulated safety, tolerability, and available PD data, up to 2 cohorts in Part B may be extended by up to 4 additional patients **per cohort** (these patients will all receive active drug, not placebo).

Section(s) also containing this change:

- 4.7.2 Study Drug Dosing, Study Progression, and Dose Escalation in Part B
- Table 9: Adaptive Study Design Areas, Features, and Limits

Purpose: Clarify the duration of screening and minimum duration of postdose follow up

The primary change occurs in Section 4.2 Duration of Treatment and Overall Duration of Study

Now reads:

- Part A: The estimated total time on study, inclusive of screening, for each subject is up to 405 days. The duration of treatment is a single dose.
- Part B: **For all patients, the duration of screening is up to 45 days and the minimum duration of postdose follow-up is 84 days. The duration of treatment and estimated total time on study, inclusive of screening, for each patient is as follows:**
  - For patients dosed monthly: The duration of treatment for patients initially randomized to receive active study drug is 57 days. The estimated total time on study~~, inclusive of screening for each patient~~ is up to 462 days. Additionally, the duration of treatment for patients initially randomized to receive placebo is 141 days. The estimated total time on study~~, inclusive of screening,~~ for each patient initially randomized to receive placebo, then active study drug, is up to 546 days.
  - For patients dosed quarterly: The duration of treatment is 85 days for patients randomized to placebo and active study drug. The estimated total time on study~~, inclusive of screening,~~ is up to 490 days.

Section(s) also containing this chage:

- Synopsis

Purpose: Add an alternative threshold for discontinuation from study follow-up for patients in Part B who do not wish to enroll in an open-label extension study.

The primary change occurs in Section 4.1.2 Multiple-ascending Dose Part in Patients with Primary Hyperoxaluria Type 1 (Part B)

Now reads:

For patients who do not enroll in the open-label extension study, safety and PD follow up will continue until 24-hour urinary oxalate is >80% of baseline, **and** ~~AND P~~ **p**lasma glycolate is <20% above baseline or ≤ the ULN. **If an investigator wishes to discontinue follow up after completion of the postdose follow-up period and prior to oxalate and glycolate recovery, the SRC must agree based upon consideration of emerging data on the safety of ALN-GO1 knockdown and the individual patient's safety and PD data.**

Section(s) also containing this change:

- Synopsis
- Section 4.6 Safety Review Committee

Purpose: Revise blood pressure eligibility criteria for pediatric patients in Part B and provide corresponding age and weight-based blood pressure tables

The primary change occurs in 5.2.2 Additional Exclusion Criteria for Part B

Revised text:

21. For patients **<18 years old,** ~~aged 6 to 11 years, inclusive, males with systolic blood pressure >115 mmHg and/or a diastolic blood pressure >75 mmHg and females with systolic blood pressure >110 mmHg and/or a diastolic blood pressure >75 mmHg after 10 minutes of rest at screening. For male and female patients aged 12 to 17 years, inclusive, systolic blood pressure >120 mmHg and/or a diastolic blood pressure >80 mmHg~~ **diastolic and/or systolic blood pressure equal to or greater than the 95th percentile for age, gender, and height (see Appendix 11.4 Table 14 and Table 15)** after 10 minutes of rest at screening.

Purpose: Add Appendix to include normative pediatric blood pressure tables and corresponding reference

The primary change occurs in Appendix 11.4 Nortmative Pediatric Blood Pressure Tables

Added text: **For pediatric patients <18 years old, study entry criteria for blood pressure will be determined according to normative blood pressure tables based on normal-weight children included in published guidelines issued by the American Academy of Pediatrics [13] (see Table 14 and Table 15). Patients will be excluded if diastolic and/or systolic blood pressure is equal to or greater than the 95th percentile for sex, age, and height (mean of screening height measurements).**

Added tables:

- Table 14: Blood Pressure Levels for Girls by Age and Height Percentile
- Table 15: Blood Pressure Levels for Boys by Age and Height Percentile

Section(s) also containing this change:

- References: Flynn, J.T., et al., *Clinical Practice Guideline for Screening and Management of High Blood Pressure in Children and Adolescents.* Pediatrics, 2017. **140**(3).

Purpose: Extend the allowable time window to conduct predose assessments (ie, vital signs, 12-lead ECGs, physical examinations, blood and urine sample collections) when scheduled at the same time points for Part B

The primary change occurs in footnote g (vital signs), footnote h (ECG), footnote q (blood sample for PD analysis) in Table 2 Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B)

Now reads:

g. Vital signs (blood pressure, pulse rate, body temperature, and respiration rate) will be measured in the supine position after the patient has rested comfortably for 10 minutes. On Day 1 only, vital signs will be measured within **2** ~~1~~ hour**s** predose; and 30 (±5 minutes) and 4 hours (±15 minutes) postdose.

h. All 12-lead ECGs are triplicate, using centralized equipment. ECGs will be measured approximately 5 minutes apart. Recordings will be obtained after the patient has rested comfortably in the supine position for approximately 10 minutes. Patients should remain supine between ECGs. On dosing days, ECGs will be measured within **2** ~~1~~ hour**s** predose; and at 1 hour (±20 minutes), 2 hours (±20 minutes), and 4 hours (±20 minutes) postdose. On all other days, ECGs should be collected at approximately the same time of day corresponding to the predose collection (±1 hour).

q. On Day 1, the blood sample for PD analysis must be collected within **2** ~~1~~ hour**s** predose. The remaining PD samples should be collected at approximately the same time of day corresponding to the predose collection (±1 hour), as applicable.

Section(s) also containing this change:

- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1 – Monthly Dosing, footnote f (vital signs), footnote g (ECG), and footnote n (blood sample for PD analysis)
- Table 4: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B – Quarterly Dosing), footnote g (vital signs), footnote h (ECG), and footnote q (blood sample for PD analysis)
- Table 11: Pharmacokinetic Time Points for Multiple-ascending Dose Cohorts in Patients with PH1 (Part B – Monthly Dosing), Day 1 Predose, Day 29 Predose, Day 57 Predose, Day 85 Predose, Day 113 Predose, and Day 141 Predose
- Table 12: Pharmacokinetic Time Points for Patients with PH1 (Part B – Quarterly Dosing), Day 1 Predose and Day 85 Predose

Purpose: Clarify that patients in Part B are enrolled in sequential dose cohorts and align study design language between the body of the protocol and synopsis

The primary change occurs in the synopsis

Now reads: In Part B, patients will be **enrolled in up to 6 sequential dose cohorts** ~~randomized~~ to receive ALN-GO1 or placebo monthly or quarterly. Patients dosed monthly will receive 3 doses of ALN-GO1 or placebo **in a blinded fashion**. After completion of the blinded portion of the study, patients dosed monthly will be unblinded on or after Day 78. Patients who initially received placebo will then receive 3 doses of open-label ALN-GO1 dosed monthly at the same dose administered to the cohort into which they were initially randomized and will follow the same assessment schedule. Patients dosed quarterly will receive either ALN-GO1 or placebo on Day 1. All patients in quarterly dosing cohorts, including those initially randomized to placebo, will receive open-label ALN-GO1 on Day 85 at the same dose administered to the cohort into which they were initially randomized. **Up to 2 expansion cohorts in Part B may be enrolled based on available safety and PD data; these patients will all receive open-label ALN-GO1, not placebo.**

Purpose: Modify footnotes in Schedule of Assessments and Figure 1 to indicate the shortened duration of study follow-up in patients who enroll in the open-label extension study and added an alternative threshold for discontinuation from study follow-up for patients in Part B who do not wish to enroll in an open-label extension study.

The primary change occurs in Figure 1: Study Design

Revised text: [a] Patients in Part B will be invited to participate in an open-label extension study **once they complete the postdose follow-up period. For patients who do not enter the open-label extension study, safety and PD follow-up will continue until they meet the criteria** ~~provided they meet the criteria~~ described in Section **Error! Reference source not found., or until the SRC makes a decision per investigator request to discontinue follow-up on a case-by-case basis. The decision cannot be made until after completion of the last postdose follow-up visit (84 days after last dose).**

Section(s) also containing this change:

- Table 2: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) – Monthly Dosing, footnote a
- Table 3: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B) who Previously Received Placebo and will Receive Open-Label ALN-GO1 – Monthly Dosing, footnote a
- Table 4: Schedule of Assessments for Multiple-ascending Dose Cohorts in Patients with Primary Hyperoxaluria Type 1 (Part B – Quarterly Dosing), footnote a

Purpose: Correct typographical errors, punctuation, grammar, abbreviations, and formatting

These changes are not listed individually.