CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER:

214103Orig1s000

INTEGRATED REVIEW

Integrated Review

Table 1. Administrative Application Information

Category	Application Information
Application type	NDA
Application number(s)	214103
Priority or standard	Priority
Submit date(s)	4/3/2020
Received date(s)	4/3/2020
PDUFA goal date	12/3/2020
Division/office	Division of Cardiology and Nephrology (DCN)
Review completion date	11/18/2020
Established/proper name	Lumasiran
(Proposed) proprietary name	Oxlumo
Pharmacologic class	Small interfering RNA
Code name	AD-65585, ALN-65585, ALN-GO1
Applicant	Alnylam Pharmaceuticals
Dosage form(s)/formulation(s)	Injectable 94.5 mg/0.5 mL single-dose vial
Dosing regimen	Subcutaneous injection based on body weight.
Applicant proposed indication(s)/ population(s)	Treatment of primary hyperoxaluria type 1 (PH1) in pediatric and adult patients.
Proposed SNOMED indication	65520001 Primary hyperoxaluria, type 1 (disorder)
Regulatory action	Approval
Approved dosage	Body Weight <10 kg: 6 mg/kg monthly for 3 doses then 3 mg/kg monthly; 10 kg to <20 kg: 6 mg/kg monthly for 3 doses then 6 mg/kg every 3 months; ≥ 20 kg: 3 mg/kg monthly for 3 doses then 3 mg/kg every 3 months
Approved indication	For the treatment of primary hyperoxaluria type 1 (PH1) to lower urinary oxalate levels in pediatric and adult patients
Approved SNOMED term for indication	65520001 Primary hyperoxaluria, type 1 (disorder)

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Glossary

ADA	antidrug antibodies
ADME	absorption, distribution, metabolism, excretion
AE	adverse event
AGT	alanine-glyoxylate aminotransferase gene
AGXT	alanine-glyoxylate aminotransferase protein
ALT	alanine aminotransferase
ASGR	asialoglycoprotein receptors
AST	aspartate aminotransferase
AUC	area under the curve
BLA	biologics license application
BW	body weight
CDER	Center for Drug Evaluation and Research
CL_H	hepatic clearance
CI	confidence interval
CL_p	plasma clearance
CL_R	renal clearance
C_{max}	maximum plasma concentration
CYP	cytochrome P450
DMC	data monitoring committee
ECG	electrocardiogram
EFD	embryo-fetal development
eGFR	estimated glomerular filtration rate
E-R	exposure-response
FDA	Food and Drug Administration
FMQ	Food and Drug Administration Medical Dictionary for Regulatory Activities query
GalNAc	N-acetylgalactosamine
GD	gestation day
GLP	good laboratory practice
GO	glycolate oxidase
HAO1	hydroxyacid oxidase 1
IC_{50}	half maximal inhibitory concentration
IND	investigational new drug
K_a	absorption rate
LS	least-squares
MedDRA	Medical Dictionary for Regulatory Activities
MMRM	mixed-effect model repeated measures
MRHD	maximum recommended human dosage
mRNA	messenger RNA
NDA	New Drug Application
NOAEL	no observed adverse effect level
NOEL	no observed effect level
OPQ	Office of Pharmaceutical Quality

PD	pharmacodynamic
PH1	primary hyperoxaluria type 1
PK	pharmacokinetic
PMC	postmarketing commitment
PMR	postmarketing requirement
PT	preferred term
qM	monthly
q3M	every three months
SAE	serious adverse event
SAP	statistical analysis plan
SC	subcutaneous
SD	standard deviation
siRNA	small interfering ribonucleic acid
SOC	system organ class
SPA	special protocol assessment
TEAE	treatment-emergent adverse event
TK	toxicokinetic
ULN	upper limit of normal
VPC	visual predictive check

I. Executive Summary

1. Summary of Regulatory Action

On April 3, 2020, Alnylam Pharmaceuticals submitted an original New Drug Application for Oxlumo® (lumasiran) for the treatment of primary hyperoxaluria type 1 (PH1) in pediatric and adult patients. Lumasiran is a subcutaneously administered, synthetic, double-stranded small interfering ribonucleic acid (siRNA) that inhibits the messenger RNA of the hydroxyacid oxidase 1 (HAO1) gene.

Overview of PH1 and the Development Program

PH1 is a rare, serious disease that often manifests in childhood or adolescence. It is caused by an inherited deficiency of the liver enzyme alanine-glyoxylate aminotransferase (AGT) that leads to overproduction of oxalate by the liver. Early in the disease, excess oxalate is cleared by the kidneys but complexes with calcium in the urine to form kidney stones or deposits in the kidney, leading to a loss of kidney function. As kidney function declines, oxalate accumulates in the body and deposits in other tissues, leading to systemic manifestations, including arrhythmias and cardiac arrest, gangrene, bone/joint pain and fractures, and blindness.

Although the manifestations of PH1 can be serious, calcium oxalate stones are slow to form and pass and loss of kidney function is generally gradual. Given the rarity of the disease, it would be challenging to detect effects on clinical events over the course of a feasible development program. As such, the development program was designed to assess effects on urinary oxalate excretion. Support for the use of urinary oxalate as a surrogate for clinical outcomes in PH1 was based primarily on 1) knowledge of the pathophysiology of the disease and the causal role of urinary oxalate in kidney stone formation, nephrocalcinosis, and loss of kidney function; 2) epidemiologic data showing an association between urinary oxalate and loss of kidney function, particularly in patients with high levels of urinary oxalate; and 3) observational data from patients treated with pyridoxine or liver transplant showing associations between reductions in urinary oxalate and preservation of kidney function. During discussions of the design of the trial, the Agency expressed a willingness to accept a substantial change in urinary oxalate (i.e., near normalization) in patients with high baseline levels as a basis for full approval.

Design of ILLUMINATE-A and -B

In support of the proposed indication, the applicant conducted two studies in patients with PH1 and relatively preserved kidney function: ILLUMINATE-A in patients ≥6 years of age and ILLUMINATE-B in patients <6 years of age. In brief, ILLUMINATE-A was a double-blind, randomized, placebo-controlled trial in which 39 patients were randomized 2:1 to lumasiran or placebo for 6 months then were eligible to receive lumasiran in an extension. The primary endpoint for ILLUMINATE-A was the percent change in 24-hour urinary oxalate excretion from baseline to Month 6. ILLUMINATE-B was a single-arm study in which all patients were treated with lumasiran. The primary endpoint for ILLUMINATE-B was the percent change in spot urinary oxalate-to-creatinine ratio from baseline to Month 6. Although ILLUMINATE-B lacked

a concurrent control, the Agency believed the results of the trial were likely to be interpretable, assuming the reduction in urinary oxalate and time course were generally consistent with the findings in ILLUMINATE-A.

Efficacy

ILLUMINATE-A met its primary endpoint with a least-squares (LS) mean percent change in 24-hour urinary oxalate from baseline to Month 6 (LS means averaged from Months 3 to 6) of -65 (3) in the lumasiran group and -12 (4) in the placebo group (between-group difference -54 [4]; 95% CI -62, -45; $p<0.0001$). Mean baseline urinary oxalate was 1.8 mmol/24 hr/1.73 m^2, well above the upper limit of normal of 0.51. Just over half of patients (21 of 39) had a baseline urinary oxalate over 1.7 mmol/24 hr/1.73 m^2, and the effect size was similar in this group. In addition, approximately half of the patients treated with lumasiran, but none treated with placebo, achieved a 24-hour urinary oxalate in the normal range by Month 6, including 5 of 14 patients with higher baseline values. Thirty-three patients (85%) reported a history of kidney stone events before enrollment, with seven (18%) requiring lithotripsy or stone extraction in the preceding year. There was no treatment effect on kidney stone events or kidney function; however, as previously noted, this was not expected in a trial only 6 months in duration.

Patients in ILLUMINATE-B demonstrated a LS mean percent change in spot urinary oxalate-to-creatinine ratio from baseline to Month 6 (LS means averaged from Months 3 to 6) of -71 (SEM 3; 95% CI -77, -65). The magnitude of the reduction in urinary oxalate and the time course were consistent with the findings in ILLUMINATE-A. The youngest patient was 4 months old.

In conclusion, placebo-controlled data from ILLUMINATE-A provide evidence of a large treatment effect of lumasiran on urinary oxalate in patients ≥6 years of age with PH1, including normalization of urinary oxalate excretion in approximately half of patients. In addition, baseline-controlled data from ILLUMINATE-B provide evidence of a treatment effect in patients <6 years of age that is consistent with the treatment effect observed in ILLUMINATE-A and supports extension of the indication down to birth.

Safety

The safety database is limited as would be expected given the size of the affected population. The most common adverse reactions were injection site reactions, which were generally mild, resolved within one day, and did not lead to treatment discontinuation. Abdominal pain was more common in patients receiving lumasiran (15%) than placebo (8%), but the events were generally mild and did not lead to study drug discontinuation. There is no obvious mechanistic basis for such an effect; hence, it is unclear whether this difference is causally related to drug. Some patients developed low-titer antidrug antibodies (ADA), but there was no apparent effect on PK, PD, or safety. Hepatic toxicity is a theoretical risk given that the drug targets the liver. Although there was no evidence of hepatic toxicity based on either adverse events or laboratory tests, the safety database is limited.

Conclusion

The findings in ILLUMINATE-A and -B are mutually substantiating and provide substantial evidence of lumasiran's effectiveness in lowering urinary oxalate. Although it is difficult to translate the observed decreases in urinary oxalate to clinical benefit based on the available data, given the size of the treatment effect and knowledge of the pathophysiology of the disease, we believe the findings are likely to translate to a benefit on important clinical manifestations of the disease, including nephrolithiasis and related complications, and loss of kidney function. The safety database is limited because of the size of affected population but does not raise significant concerns. As such, we believe the data provide compelling evidence of a benefit that outweighs the risks, and the application warrants approval. The approval will include postmarketing requirements to complete preclinical carcinogenicity studies.

2. Benefit-Risk Assessment

2.1. Benefit-Risk Framework

Table 2. Benefit-Risk Framework

Dimension	Evidence and Uncertainties	Conclusions and Reasons
Analysis of Condition	• Primary hyperoxaluria type 1 (PH1) is a rare, serious disease that often manifests in childhood or adolescence. • PH1 is caused by an inherited deficiency of the liver enzyme alanine-glyoxylate aminotransferase (AGT) that leads to overproduction of oxalate by the liver. • Patients with PH1 have recurrent kidney stones and can have deposition of calcium in the kidneys (nephrocalcinosis), leading to a loss of kidney function. As kidney function declines, oxalate accumulates in the body and deposits in other tissues, leading to systemic manifestations, including arrhythmias and cardiac arrest, gangrene, bone/joint pain and fractures, and blindness.	PH1 is a rare, serious genetic disease that usually presents in childhood with recurrent kidney stones. Manifestations of more advanced disease include kidney failure, bone and joint abnormalities, cardiac arrhythmias and arrest, and blindness.
Current Treatment Options	• There are no approved therapies for PH1. • Pyridoxine may reduce hepatic oxalate production in patients with certain genetic mutations. • Patients are treated with high fluid intake and medications such as citrate and magnesium to reduce calcium oxalate crystallization, though many progress despite these interventions. • Liver transplant replaces the missing AGT enzyme and is essentially curative but is associated with substantial morbidity.	There is unmet medical need for therapies to treat PH1.

Dimension	Evidence and Uncertainties	Conclusions and Reasons
Benefit	• In ILLUMINATE-A in patients ≥6 years of age, lumasiran demonstrated a significant percent reduction in 24-hour urinary oxalate from baseline to Month 6 of -65 (3) in the lumasiran group and -12 (4) in the placebo group ($p<0.0001$). A similar effect was seen in patients with high baseline urinary oxalate values, and approximately half of patients receiving lumasiran achieved normal urinary oxalate values by Month 6. • In ILLUMINATE-B in patients <6 years of age, lumsiran demonstrated a percent reduction in spot urinary oxalate-to-creatinine ratio from baseline of -71 (95% CI -77, -65). The magnitude of the reduction and the time course were consistent with the findings in ILLUMINATE-A. • There were no treatment effects on kidney function or kidney stone events; however, such effects were not expected in trial only 6 months in duration.	Lumasiran reduces urinary oxalate, with over half of patients achieving normal levels by Month 6. Given the pathophysiology of PH1 and the available epidemiologic data, the effects are likely to translate to benefits on kidney stone events and kidney function, although such effects were not observed in the trials. Given that calcium oxalate stones are slow to form and pass and loss of kidney function is generally gradual, such effects were not expected given the relatively short duration of follow-up.
Risk and Risk Management	• Although lumasiran was generally well-tolerated in ILLUMINATE-A and -B, the safety database was small and limited in duration. • The most common adverse events (AEs) were injection site reactions, which occurred throughout the study period. These were generally mild, resolved within one day, and did not lead to treatment discontinuation. • Abdominal pain occurred more frequently in patients receiving lumasiran (15%) than placebo (8%) in ILLUMINATE-A. Episodes were generally mild and did not lead to treatment discontinuation. There is no obvious mechanistic basis for such an effect; hence, it is unclear whether this difference is drug-related. • Low-titer antidrug antibodies were detected in one patient in ILLUMINATE-A and three patients in ILLUMINATE-B. Most were transient, and there was no apparent impact on PK, PD, or safety.	Lumasiran was well-tolerated. Risks were generally mild and included injection site reactions, immunogenicity of unknown significance, and possibly abdominal pain of a non-serious nature. There were no apparent significant or irreversible toxicities.

2.2. Conclusions Regarding Benefit-Risk

Together, ILLUMINATE-A and -B demonstrate that lumasiran causes a meaningful decrease urinary oxalate excretion in patients with PH1. Although it is difficult to translate the observed treatment effect to clinical benefit based on the available data, given the size of the treatment effect and knowledge of the pathophysiology of the disease, we believe the findings are likely to translate to a benefit on important clinical manifestations of the disease, including nephrolithiasis and related complications and loss of kidney function. In light of the age range of participants in the studies, approval is supported for all ages, down to birth. Injection site reactions were the main safety findings, but were generally mild, transient, and did not lead to treatment discontinuation. There were no other apparent significant or irreversible toxicities. As such, the benefits outweigh the risks.

II. Interdisciplinary Assessment

3. Introduction

Disease Background

PH1 is the most common and severe type of primary hyperoxaluria, a group of rare genetic inborn errors of hepatic glyoxalate metabolism characterized by overproduction of oxalate by the liver. To date, three types of PH have been described, defined by the affected gene. PH1 accounts for 70-80% of cases and is caused by recessively inherited mutations in the alanine-glyoxylate aminotransferase (AGXT) gene, which lead to a deficiency of the hepatic peroxisomal enzyme AGT. Under normal circumstances, AGT catalyzes the conversion of glyoxylate to glycine. AGT deficiency results in accumulation of glyoxylate, which is rapidly metabolized to oxalate, a poorly soluble organic acid normally excreted by the kidneys. Oxalate binds to calcium, saturates the urine with calcium oxalate, and deposits in various parts of the kidneys, leading to the primary clinical manifestations of PH1. Patients present, usually in childhood, with symptoms and sequelae of calcium oxalate crystalluria, recurrent urolithiasis, urinary tract infections, nephrocalcinosis, kidney scarring and eventual kidney failure. As kidney function declines, urinary excretion of oxalate decreases and oxalate accumulates in the body, leading to systemic oxalosis, a debilitating condition that can cause arrhythmias, cardiac arrest, gangrene, bone and joint pain, reduced mobility, fractures, and blindness.

Current therapies aim to inhibit calcium oxalate crystalluria and urolithiasis with excessive fluid intake to maintain brisk flow of dilute urine and increase urinary oxalate solubility with citrate and magnesium supplements. There are no approved pharmacologic treatments for PH1. Pyridoxine, a coenzyme of AGT, is partially effective in some genotypes that result in mislocalization of AGT within the hepatocyte. Liver transplant is curative but is associated with significant morbidity. When moderate to severe kidney failure develops, patients require combined or sequential liver and kidney transplant, often preceded by a period on intensive dialysis.

Lumasiran

Lumasiran is a subcutaneously administered, synthetic, double-stranded siRNA conjugated to an N-acetyl-D-galactosamine (GalNAc) ligand. The ligand is designed to [redacted]. The siRNA inhibits the messenger RNA of the hepatic hydroxyacid oxidase 1 (HAO1) gene, which encodes glycolate oxidase, the enzyme that oxidizes glycolate to glyoxylate. Glyoxylate is further metabolized to oxalate. Suppression of glycolate oxidase should, therefore, reduce hepatic oxalate production, in turn reducing oxaluria and its sequela. Glycolate is water-soluble, and in excess is not predicted to have deleterious effects (see Section 6.2). Alnylam Pharmaceuticals is developing lumasiran for the treatment of PH1. Two studies to support the proposed indication, ILLUMINATE-A and ILLUMINATE-B, are the focus of this review.

3.1. Review Issue List

The review team has determined there are no review issues related to the evaluation of risk or benefit that warrant discussion in this section. Discussion of considerations related to the design of the phase 3 studies including the use of urinary oxalate as a surrogate endpoint may be found above in Section 1.

3.2. Approach to the Review

This was a joint review. Kirtida Mistry and Dali Zhou focused on the data supporting efficacy and Kirtida Mistry and the Clinical Data Scientist team, Ling Cao and Jinzhong Liu, focused on the data supporting safety. Philip Gatti reviewed the nonclinical toxicology studies, and Harisudhan Thanukrishnan reviewed the clinical pharmacology data.

The review focused on the two phase 3 trials shown in Table 3 that provide primary support for the proposed indication in patients 6 years of age and older (ILLUMINATE-A) and in patients <6 years of age (ILLUMINATE-B).

Table 3. Clinical Trials Submitted in Support of Efficacy and Safety Determinations[1] for Lumasiran

Trial Identifier	Trial Population	Trial Design	Dose, Number Treated, Duration	Primary Endpoint	Number of Subjects Randomized	Number of Trial Sites
ALN-GO1-003 (ILLUMINATE-A) NCT03681184	Children ≥6 years of age and adults with primary hyperoxaluria type 1 (PH1)	Randomized, double-blind, placebo-controlled	Dose: 3 mg/kg qM x 3 followed by q3M Number treated: 26 Treatment duration: mean 5.5 months	Percent change in urinary oxalate excretion	Planned: 30 Actual: 39	Centers: 16 Countries: 8
ALN-GO1-004 (ILLUMINATE-B) NCT03905694	Infants and children <6 years of age with PH1	Single-arm, baseline-controlled	Dose: <10 kg: 6 mg/kg qM x 3 followed by 3 mg/kg qM; 10 kg-20 kg: 6 mg/kg qM x 3 followed by 6 mg/kg q3M; ≥20 kg: 3 mg/kg qM x 3 followed by 3 mg/kg q3M Number treated (efficacy population): 16 Duration: mean 5.6 months	Percent change in urinary oxalate excretion	Planned: 8 (revised to 20) Actual (safety population): 18	Centers: 9 Countries: 5

Source: Clinical Study Report and adsl.xpt
[1] Includes all submitted clinical trials, even if not reviewed in depth, except for phase 1 and pharmacokinetic studies.
Abbreviations: qM = once monthly; q3M = every three months

4. Patient Experience Data

The Applicant collected information about patient quality of life and patient and caregiver experience living with PH1 (Table 4). The data were not intended to support labeling claims and were not reviewed in detail. The review team also considered the experience and perspectives shared by patients and caregivers during an Externally-led Patient Focused Drug Development meeting hosted by the Oxalosis and Hyperoxaluria Foundation on October 5, 2020 and the publications listed in Table 4 to inform the benefit-risk assessment.

Table 4. Patient Experience Data Submitted or Considered

Data Submitted in the Application		
Check if Submitted	**Type of Data**	**Section Where Discussed, if Applicable**
Clinical outcome assessment data submitted in the application		N/A
☒	Patient-reported outcome	
☒	Observer-reported outcome	
☐	Clinician-reported outcome	
☐	Performance outcome	
Other patient experience data submitted in the application		
☐	Patient-focused drug development meeting summary	
☐	Qualitative studies (e.g., individual patient/caregiver interviews, focus group interviews, expert interviews, Delphi Panel)	
☐	Observational survey studies	
☐	Natural history studies	
☐	Patient preference studies	
☐	Other: (please specify)	
☐	**If no patient experience data were submitted by Applicant, indicate here.**	
Data Considered in the Assessment (But Not Submitted by Applicant)		
Check if Considered	**Type of Data**	**Section Where Discussed, if Applicable**
☒	Perspectives shared at patient stakeholder meeting	1, 2
☐	Patient-focused drug development meeting summary report	
☐	Other stakeholder meeting summary report	
☐	Observational survey studies	
☒	Other: Publications: 1. Lawrence J, Wattenberg DJ: Primary hyperoxaluria: The patient and caregiver perspective. Clin J Am Soc Nephrol 15: 1056-1065, 2020. PMID 32165441 2. Milliner DS, McGregor TL, Thompson A, et al: Endpoints for Clinical Trials in Primary Hyperoxaluria. Clin J Am Soc Nephrol 15: 909-911, 2020. PMID 32165440	1, 2

5. Pharmacologic Activity, Pharmacokinetics, and Clinical Pharmacology

The target site of lumasiran in the HAO1 mRNA is conserved across mammalian species and both rodent and monkey are pharmacologically relevant species. In vitro and in vivo pharmacology studies demonstrated the pharmacodynamic (PD) effect of lumasiran. Transfection of lumasiran into cynomolgus monkey primary hepatocytes resulted in a dose-dependent inhibition of HAO1 mRNA level, with a half maximal inhibitory concentration (IC_{50}) of 10pM. In vivo pharmacological studies in wild type animals as well as animal models of PH1 showed that lumasiran administration significantly lowered urinary oxalate levels in a dose-dependent fashion through the silencing of HAO1 mRNA and secondarily increased serum glycolate (a substrate of GO) levels.

A bioinformatic analysis was conducted for potential off-target binding and revealed no human transcripts (other than HAO1) with fewer than five nucleotide mismatches with either the sense or antisense strand of lumasiran. Follow-up cell transfection assays demonstrated a >1000-fold difference between the “on-target” suppression of HAO1 by lumasiran and the off-target suppression of any of the predicted off-target transcripts, confirming the specificity of the siRNA for HAO1 mRNA.

Table 5. Summary of General Clinical Pharmacology and Pharmacokinetics

Characteristic	Drug Information
	Pharmacologic Activity
Established pharmacologic class (EPC)	Small interfering ribonucleic acid (siRNA)
Mechanism of action	Lumasiran is a double-stranded small interfering ribonucleic acid (siRNA) that reduces levels of glycolate oxidase (GO) enzyme by targeting the hydroxyacid oxidase 1 (HAO1) messenger ribonucleic acid (mRNA) in hepatocytes through RNA interference.
Active moieties	Lumasiran
QT prolongation	Safety pharmacology studies in monkeys did not demonstrate prolongation in QT or QTc (using the individual animal correction factor) after four subcutaneous doses of 10 and 100 mg/kg administered in a 3-week period.
	General Information
Bioanalysis	Lumasiran concentrations in plasma and urine were measured using a validated LC-TOF-MS method with a quantification range of 10 to 10,000 ng/mL. Urinary and plasma oxalate were measured using a validated LC-MS/MS method with a quantification range of 5 to 250 µg/mL (0.055 to 2.78mM) and 0.5 to 50 µg/mL (0.005 to 0.55mM), respectively. A dilution method was later validated to include a lower limit of quantification for urinary oxalate from 0.83 µg/mL (0.0093mM). The performance of the methods was acceptable.
Healthy subjects versus patients	Plasma PK of lumasiran was similar in healthy subjects (n=32, mean age of 29 years, mean BW of 71 kg) and PH1 patients (n=20, mean age of 15 years, mean BW of 50 kg).
Drug exposure at steady state following the therapeutic dosing regimen (or single dosage, if more relevant for the drug)	**Parameter*** **Mean (%CV)** AUC (ng/h/mL) 7960 (21.7) C_{max} (ng/mL) 701 (73.0) *Values are after 3 monthly doses of 3 mg/kg.
Range of effective dosage(s) or exposure	After a lumasiran dose of 3 mg/kg once monthly (qM) for 3 months, 24-hour urinary oxalate levels were reduced by an average of 72% and 61% in patients ≥6 years of age with PH1 in studies ALN-GO1-001 and ILLUMINATE-A, respectively. In study ILLUMINATE-B, a 6 mg/kg qM dose for 3 months reduced spot urinary oxalate-to-creatinine ratio by an average of 72% and 84% in children 10 to <20 kg and <10 kg, respectively.
Maximally tolerated dosage or exposure	A maximum tolerated dose was not identified for lumasiran. A maximum single dose of 6 mg/kg was evaluated in healthy volunteers (C_{max} up to 1420 ng/mL) in study ALN-GO1-001 and after multiple qM doses in PH1 patients weighing <20 kg (Day 1 concentrations 4 hours post-dose up to 1760 ng/mL) in ILLUMINATE-B.
Dosage proportionality	Lumasiran exhibited linear PK in the range of doses evaluated. Mean plasma C_{max} and $AUC_{0\text{-last}}$ increased in an approximately dose proportional manner following a single SC dose of 0.3 to 6 mg/kg or multiple SC doses of 1 to 3 mg/kg.
Accumulation	No accumulation in the plasma occurred after repeated monthly or quarterly dosing, as the plasma half-life was short (1.5 to 9 hours) compared with the dosing interval.

Characteristic	Drug Information
Time to achieve steady-state	Exposure in plasma after a single dose is expected to be similar to exposure at steady-state. The concentrations of lumasiran in the target site (liver) were predicted to reach steady-state by month 3, after administration of the third loading dose. In the absence loading doses, time to achieve steady-state lumasiran liver concentrations and oxalate suppression was estimated to be 12 months.
	Absorption
Bioavailability	The absolute bioavailability of lumasiran after SC dosing was not determined. However, based on near complete absorption of lumasiran from the SC injection site in the rat mass balance study, close to 100% bioavailability of lumasiran is expected after SC dosing in humans.
T_{max} Median (min-max)	4.1 (0.5, 12) hours
	Distribution
Volume of distribution	The population estimate for the apparent central V_d/F in a 70 kg person was 4.9 L, which was close to the total blood volume. Following, subcutaneous dosing, lumasiran is expected to primarily be taken up via the asialoglycoprotein receptors (ASGR) into the liver.
Plasma protein binding	Protein binding was concentration-dependent and lumasiran was 77 to 85% bound at clinically relevant concentrations (0.5 to 1 µg/mL).
Drug as substrate of transporters	No substrate studies were done for lumasiran on drug transporters.
	Elimination
Mass balance results	A mass balance study in humans was not performed due to the potential health hazard from concentrated and prolonged exposure to radioactivity in the liver
Clearance	Body weight-normalized total plasma clearance increased with decreasing body weight: values were 0.64, 0.54 and 0.38 L/h/kg in 10, 20 and 70 kg patients, respectively. The population estimate for the apparent total plasma clearance in a 70 kg patient was 26.5 L/h, with CL_H representing 80% of total clearance (mean: 21.1 L/h) and CL_R representing the remaining 20% (mean: 5.4 L/h, in patients having normal renal function with eGFR of 90 mL/min/1.73 m^2).
Half-life	Mean terminal half-life in plasma was 5.2 hours (range: 1.5 to 9 hours).
Metabolic pathway(s)	Lumasiran was not found to be a substrate for CYP1A2, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, CYP3A4, or CYP3A5 enzymes. Lumasiran is metabolized primarily by nucleoside cleavage by exo- or endo-nucleases in the serum or liver into AS (N-1)3', which was less than 10% of parent circulating in the plasma and urine (Source: clinical study ALN-GO1-001).
Primary excretion pathways (% dosage)	The majority of the administered dose of lumasiran is rapidly taken up and internalized in the liver within 24-48 hours and further metabolized by the exo- or endo- nucleases in the liver. The fraction of the dose excreted in urine depended on kidney function and ranged from 17 to 26% in healthy volunteers and 7 to 14% in PH1 patients.

Characteristic	Drug Information
	Intrinsic Factors and Specific Populations
Body weight	Body weight-based dosing is recommended consisting of appropriate loading and maintenance doses. **Body Weight** / **Loading Dose** / **Maintenance Dose (begin 1-month after the last loading dose)** <10 kg / 6 mg/kg once monthly for 3 doses / 3 mg/kg once monthly 10 to <20 kg / 6 mg/kg once monthly for 3 doses / 6 mg/kg once every 3 months ≥20 kg / 3 mg/kg once monthly for 3 doses / 3 mg/kg once every 3 months
Age	PK data with the body weight-based dosing regimen showed similar $AUC_{0\text{-last}}$ across the evaluated age range (4 months to 60 years). The C_{max} in children <6 years of age was slightly higher, possibly due to faster absorption and the nominally higher dose given to the lower body weight patients. No age-based dose adjustment is needed.
Renal impairment	Across studies, C_{max} of lumasiran was similar, and $AUC_{0\text{-last}}$ was 25% higher (only in moderate impairment) and reduction in urinary oxalate was similar in mild and moderate renal impairment patients compared to patients with normal renal function. Dose adjustment is not needed in patients with renal impairment.
Hepatic impairment	No dose adjustment is required for patients with mild or moderate hepatic impairment. Based on the limited clinical experience and available literature, liver uptake of lumasiran and urinary oxalate reduction is not expected to be affected in mild and moderate hepatic impairment. Lumasiran has not been studied in patients with severe hepatic impairment.
	Drug Interaction Liability (drug as perpetrator)
Inhibition/induction of metabolism	Lumasiran did not show any direct or time-dependent inhibition of any of the major CYP enzymes in vitro at clinically relevant concentrations. CYP induction is not expected with lumasiran, and no studies have been conducted.
Inhibition/induction of transporter systems	No inhibitor or inducer studies were done for lumasiran on drug transporters.
	Immunogenicity (if applicable)
Bioanalysis	Immunogenicity was evaluated by measuring immunoglobin (Ig)G/IgM antibodies against lumasiran in human serum using a validated enzyme linked immunosorbent assay (ELISA) with a sensitivity of 65.6 ng/mL. A neutralizing assay for ADA was not developed because there was no apparent impact on efficacy either in PH1 patients (a potential absence of reduction in oxalate levels) or in healthy volunteers (a potential decrease in glycolate levels).
Incidence	ADA titers were low and transiently observed in 6 of 100 (6%) PH1 patients treated with lumasiran.
Clinical impact	No clinically significant differences in the safety, pharmacokinetic, or pharmacodynamic profiles of lumasiran were observed in patients who tested positive for anti-lumasiran antibody.

5.1. Nonclinical Assessment of Potential Effectiveness

The nonclinical data support the efficacy of lumasiran to decrease levels of HAO1 mRNA, GO enzyme levels and the production of oxalate, based on the following data:

- In vitro study: Transfection assays in primary cynomolgus monkey hepatocyte cells resulted in dose-dependent inhibition of endogenous HAO1 mRNA levels with an IC_{50} of 10pM.
- In vivo study in wild-type mice: A single dose of lumasiran led to dose-dependent suppression of HAO1 mRNA levels in wild-type mice with approximately 91% suppression at the highest dose of 10 mg/kg on Day 10, with an ED50 of 0.3 mg/kg. Single doses of 0.1, 0.3, 1 and 3 and mg/kg lumasiran led to 13%, 43%, 77%, and 85% suppression of HAO1 mRNA expression, respectively. Conversely, serum glycolate concentrations increased from 51 μM in the PBS-treated controls to approximately 217μM at the highest dose of 10 mg/kg on Day 10.
- In vivo study in wild-type rats: A single SC dose of lumasiran led to dose-dependent suppression of HAO1 mRNA in wild-type Sprague-Dawley rats with approximately 94% HAO1 mRNA suppression at the highest dose of 10 mg/kg on Day 10. Single doses of 0.1, 0.3, 1 and 3 mg/kg lumasiran led to 15%, 52%, 69% and 90% HAO mRNA suppression, respectively. Conversely, serum glycolate concentrations increased approximately 8-fold from 1 μM in the PBS-treated controls to approximately 71 μM at the highest dose of 10 mg/kg on Day 10.
- In vivo study in a genetic PH1 mouse model (AGXT-/-) (single dose): Single doses of 0.3, 1 and 3 mg/kg lumasiran resulted in a dose-dependent reduction of urinary oxalate and an increase in urinary glycolate. A single 3 mg/kg dose of lumasiran resulted in a reduction of urinary oxalate by approximately 50%.
- In vivo study in an induced PH1 rat model (single dose): A single dose of lumasiran resulted in a dose-responsive suppression of HAO1 mRNA levels in a rat model of PH1, with approximately 87% inhibition at the highest dose of 0.3 mg/kg on Day 14. Single doses of 0.01, 0.03 and 0.1 mg/kg lumasiran led to 18%, 42% and 66 % inhibition, respectively. In parallel, there was a dose-dependent reduction of urinary oxalate levels in this rat model, with approximately 90% reduction at the highest dose of 0.3 mg/kg on Day 14. Lumasiran doses of 0.01, 0.03 and 0.1 mg/kg, led to 26%, 37% and 75% reduction of urinary oxalate, respectively, in the rats.
- In vivo study in an induced PH1 rat model (repeated dose): Repeated dosing of lumasiran resulted in a potent inhibition of HAO1 mRNA in a rat model of PH1 with approximately 98% HAO1 mRNA silencing after multiple doses of 1 or 3 mg/kg, and 95% HAO1 mRNA silencing after multiple doses of 0.3 mg/kg on Day 28. Repeated dosing of lumasiran led to a potent reduction of urinary oxalate in this rat model that was maintained for 3 weeks. On Day 28, urinary oxalate was reduced up to 98% after the

administration of four doses of lumasiran at 3 mg/kg. After four weekly doses of 0.3 or 1 mg/kg, urinary oxalate was reduced to about 87% and 88% respectively.

- In vivo study in wild-type cynomolgus monkeys (repeated dose): Repeated-dose studies in monkeys showed dose-responsive silencing of HAO1 mRNA, with up to 99% mRNA silencing when dosed with 4 mg/kg lumasiran monthly or 2 mg/kg weekly.

6. Assessment of Effectiveness

6.1. Dose and Dose Responsiveness

The Applicant proposed body weight (BW)-based loading and maintenance dosing regimens as shown in Table 6.

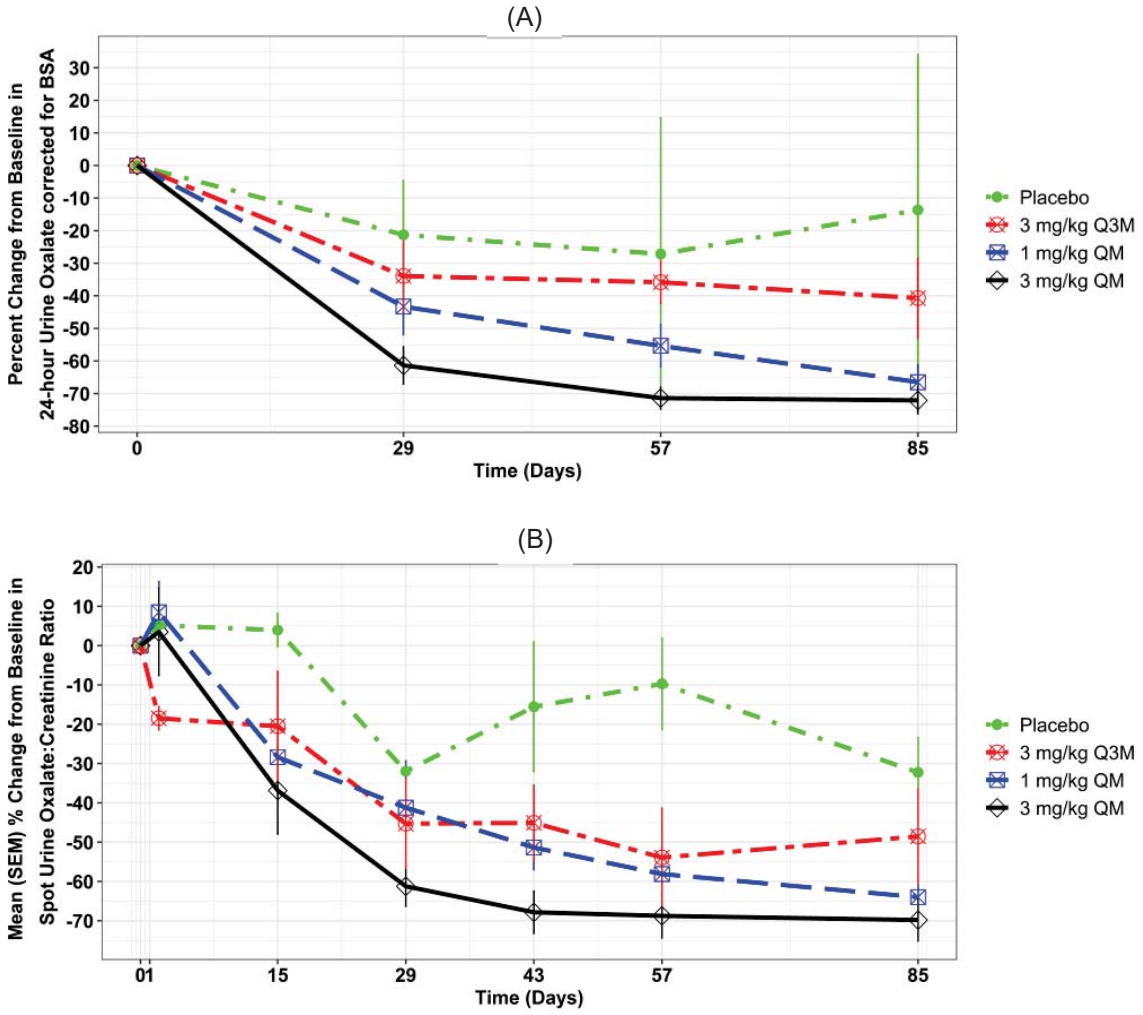
Table 6. Proposed Body Weight-based Loading and Maintenance Dosing Regimens for Lumasiran

Body Weight	Loading Dose	Maintenance Dose (begin 1 month after the last loading dose)
Less than 10 kg	6 mg/kg once monthly for 3 doses	3 mg/kg once monthly
10 kg to less than 20 kg	6 mg/kg once monthly for 3 doses	6 mg/kg once every 3 months
20 kg and above	3 mg/kg once monthly for 3 doses	3 mg/kg once every 3 months

Source: Applicant's study protocol.

The selection of the loading dosing regimen of 3 mg/kg once monthly (qM) for patients weighing ≥20 kg was based on pharmacokinetic (PK), PD and efficacy data from the dose finding trial ALN-GO1-001B (Trial 001B). The results (Figure 1) showed that the 3 mg/kg qM dosing regimen had a faster and greater reduction in 24-hour urinary oxalate corrected for BSA ($UOxBSA_{24h}$) and spot urinary oxalate-to-creatinine ratio (Ox/Cr) than 1 mg/kg qM and 3 mg/kg every three months (q3M).

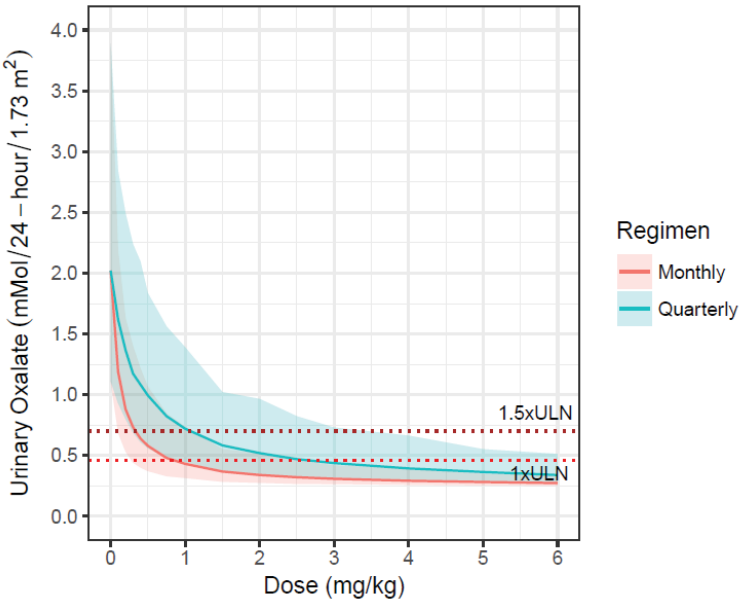
Figure 1. Observed Dose-Response Relationships for (A) Urinary Oxalate Corrected for BSA and (B) Spot Urinary Oxalate-to-Creatinine Ratio in Trial 001B



Source: Applicant's Population PK/PD Modeling Report (Phase 3 data) Figure 13.

The maintenance dosing regimen of 3 mg/kg q3M for patients weighing ≥20 kg was supported by the Applicant's semi-mechanistic allometric PK/PD model. Refer to Section 14.2.2 for more information regarding the Applicant's population PK/PD analysis. The PK/PD model predicted human liver and RNA-induced silencing complex (RISC)-loaded lumasiran PK based on liver and RISC-loaded concentrations in rats and liver concentrations in monkeys using an allometric scaling approach. This approach was implemented because liver PK ($t_{1/2}$= 66.9 days) rather than plasma PK ($t_{1/2}$= 6.0 hours) is believed to be responsible for the PD effect of lumasiran. The PK/PD modeling and simulation analysis showed that 3 mg/kg q3M reached the plateau of the dose-response relationship and was predicted to suppress $UOxBSA_{24h}$ to near normal levels at steady state similar to the 3 mg/kg qM dosing regimen (Figure 2).

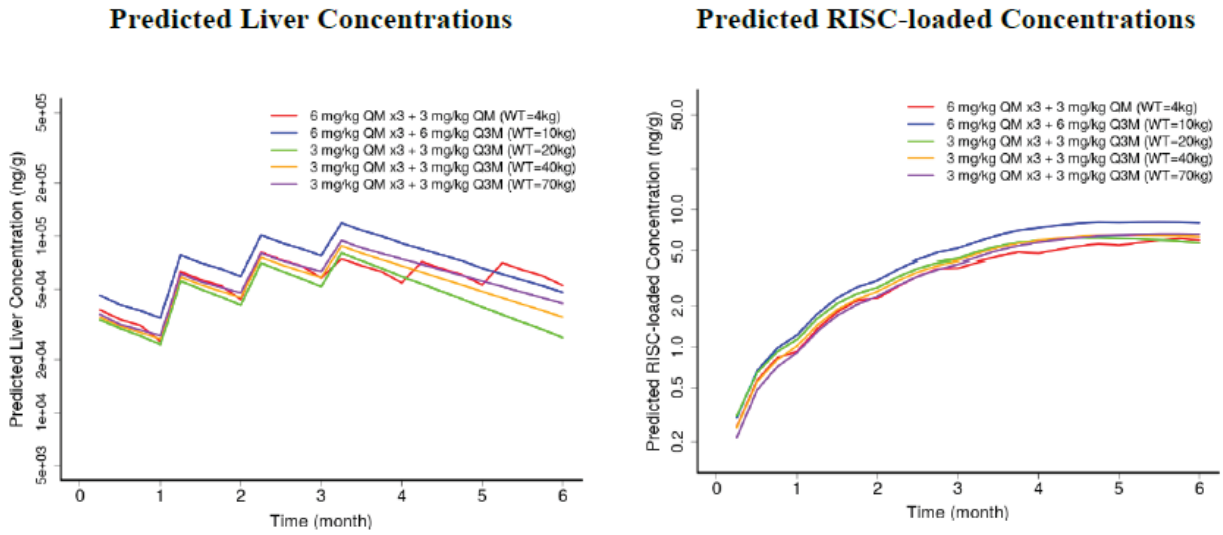
Figure 2. Steady State 24-Hour Urinary Oxalate Levels Following qM and q3M Lumasiran Doses



Source: Applicant's Population PK/PD Modeling Report (Phase 1 data) Figure 14.

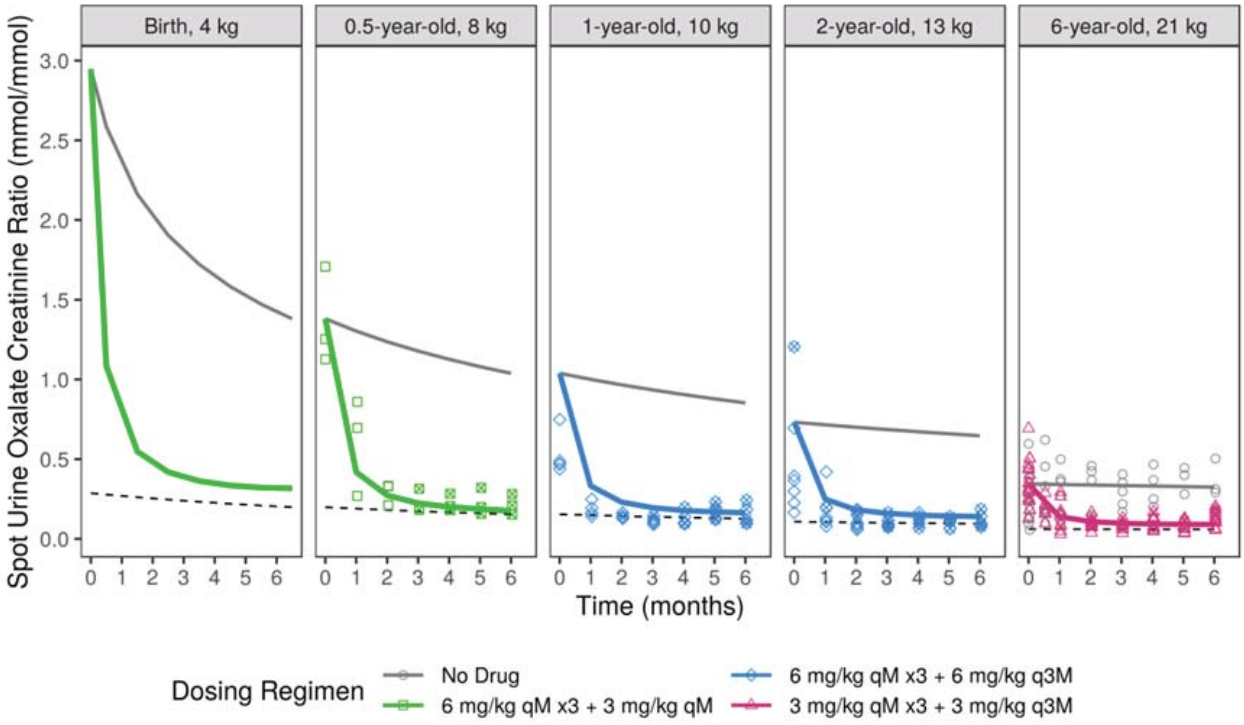
The higher loading dose of 6 mg/kg qM and maintenance dose of 6 mg/kg q3M for patients weighing 10 to <20 kg and 3 mg/kg qM for patients weighing <10 kg were selected because of: 1) a higher BW-normalized lumasiran clearance of 0.60 (%CV: 26%) L/h/kg in patients weighing <20 kg compared to 0.53 (45%) L/h/kg in patients weighing ≥20 kg, and 2) a larger relative liver size (2.9% to 3.5%) in patients <6 years of age compared to patients ≥6 years of age (2.1% to 2.5%) (Noda et al; PMID 9126583). The 3 mg/kg qM maintenance dosing regimen was proposed for infants <10 kg to keep pace with rapid growth in body weight and liver size. The Applicant's PK/PD analysis also showed that the higher lumasiran loading and maintenance dosing regimens for patients <20 kg were expected to achieve comparable liver and RISC-loaded lumasiran PK (Figure 3) and yield Ox/Cr ratio values (Figure 4) that are close to normal levels similar to patients weighing ≥20 kg.

Figure 3. Predicted Concentration-Time Profiles of Lumasiran in Liver and RISC-Loaded Lumasiran With Proposed Dosing Regimens in Patients With PH1



Source: Applicant's Population PK/PD Modeling Report (Phase 3 Data) Figure 25.

Figure 4. Model-Predicted and Observed Spot Urinary Oxalate: Creatinine Ratio in PH1 Patients up to 6 Years of Age



Source: Applicant's Population PK/PD Modeling Report (Additional ALN-GO1-004 Data) Figure 6.

The proposed BW-based dosing regimens for lumasiran were evaluated in ILLUMINATE-A in patients ≥20 kg and in ILLUMINATE-B in patients <20 kg. The efficacy data from these two

trials showed generally similar reductions in the Ox/Cr ratio and $UoxBSA_{24h}$ across different weight groups (Table 7), which justifies the Applicant's proposed BW-based dosing regimens.

Table 7. Comparison of Efficacy Outcomes From ILLUMINATE-A and -B, by Body Weight Group

Trial	Body Weight	Observed % Reduction in Ox/Cr Ratio	Observed % Reduction in $UoxBSA_{24h}$
ALN-GO1-003	20 kg and above	70% (N=26)	65% (N=26)
ALN-GO1-004	Less than 10 kg	84% (N=3)	N.A.
	10 kg to less than 20 kg	67% (N=11)	63% (N=2)
	20 kg and above	71% (N=2)	N.A.

Source: FDA's analysis.

In summary, the proposed BW-based loading and maintenance dosing regimens are justified and approvable.

6.2. Clinical Trials Intended to Demonstrate Efficacy

6.2.1. Results of Analyses, ILLUMINATE-A and -B

The review team did not conduct integrated analyses of efficacy because of differences in the design of the pivotal trials, ILLUMINATE-A and -B. See Sections 6.2.2 and 6.2.3 for analyses of individual trial results.

6.2.2. Trial ALN-GO1-003 (ILLUMINATE-A)

Overview of Study

In support of the indication, the Applicant provided data from the initial 6-month placebo-controlled portion of a phase 3 trial titled "ILLUMINATE-A: A Phase 3 Randomized, Double-Blind, Placebo-Controlled Study with an Extended Dosing Period to Evaluate the Efficacy and Safety of Lumasiran in Children and Adults with Primary Hyperoxaluria Type 1." The trial is being conducted at 16 sites in eight countries, including four sites in the US, five in Europe, and two in the Middle East.

Refer to Section 15 for further details on important trial dates and the design of ILLUMINATE-A.

Initial Protocol and Amendments

The original protocol (Amendment 1) was submitted on July 31, 2018 and amended once (Amendment 2) on March 19, 2019. The amendment allowed enrollment of patients with a lower estimated glomerular filtration rate (eGFR), clarified the primary endpoint, and changed some of the exploratory endpoints to secondary. The trial had enrolled five patients at the time of the amendment. The overview provided in this section is based on the original protocol with amendments noted.

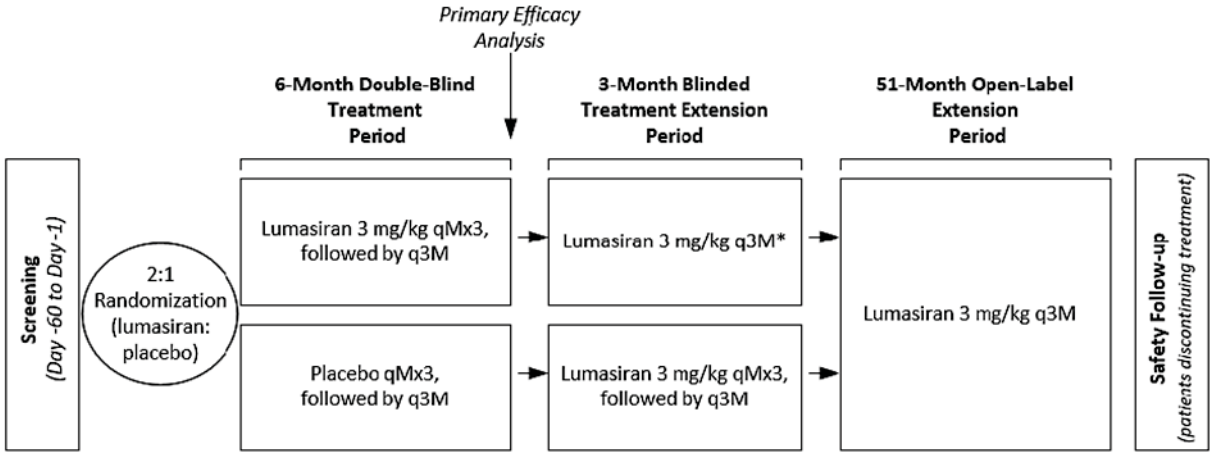
6.2.2.1. Design, ILLUMINATE-A

ILLUMINATE-A is an ongoing phase 3 trial to evaluate the efficacy and safety of lumasiran in adults and children ≥6 years of age with PH1, a confirmed AGXT mutation, urinary oxalate

excretion ≥0.70 mmol/24 hr/1.73 m^2 and an eGFR ≥30 mL/min/1.73 m^2 (reduced from >45 mL/min/1.73 m^2 with Amendment 2). Patients first entered a screening period of up to 60-days. During screening, the diagnosis of PH1 was established by confirmation of a genetic mutation in AGXT and the patients completed three 24-hour urine collections to confirm a mean (changed to median with Amendment 2) 24-hour urinary oxalate excretion ≥0.70 mmol/24 hr/1.73 m^2.

The trial is being conducted in two phases: a 6-month placebo-controlled, double-blind period that has been completed, and an ongoing 54-month extension period. Notable exclusions were history of systemic oxalosis, and prior kidney or liver transplantation. As shown in Figure 5, patients were randomized 2:1 to lumasiran 3 mg/kg or matching placebo monthly for three months then every three months for the first 6 months. The placebo-controlled treatment period ended with completion of the Month 6 visit assessments. Immediately after completion of the treatment period, all patients transitioned to lumasiran in a 54-month extension period. Investigators and patients have remained blinded to initial treatment assignment for the first 3 months of the extension.

Figure 5. ILLUMINATE-A Study Design



Abbreviations: q3M=once every 3 months; qM=once monthly; qMx3=once monthly for 3 consecutive months
* Patients randomized to lumasiran will also receive 2 qM doses of placebo during the 3-month blinded treatment extension period.

Source: Applicant Clinical Study Report.

Patients taking pyridoxine were to be on a stable regimen for at least 90 days before randomization and for the first 12 months of treatment. Patients continued their standard of care regimens, including hyperhydration and crystallization inhibitors.

Refer to Section 15 for further details on the design of ILLUMINATE-A.

Study Objectives

The primary objective was to evaluate the effect of lumasiran on percent reduction in urinary oxalate excretion.

The secondary objectives were to evaluate:

- The effect of lumasiran on absolute levels of urinary oxalate excretion, Ox/Cr ratios, and plasma oxalate (Amendment 2 changed the effect of lumasiran on plasma oxalate from an exploratory to secondary objective)
- The effect of lumasiran on kidney function
- The long-term treatment effect of lumasiran (Amendment 2 changed this from an exploratory to secondary objective)

Study Endpoints

The primary endpoint was the percent change in 24-hour urinary oxalate excretion corrected for BSA from baseline to Month 6 (LS means derived from mixed-effect model repeated measures [MMRM] model averaged across Months 3 to 6; changed from Months 1 to 6 with Amendment 2, to reflect when the treatment effect was expected to have reached steady state). Triplicate (minimum duplicate) 24-hour urine collections for oxalate were obtained during screening prior to the first dose of study drug (baseline) and at Month 6, and the median value from valid collections was used in the analysis (changed from mean with Amendment 2). A single 24-hour urine collection was obtained at other monthly visits. 24-hour urine collections were considered valid if the duration of the collection was 22 to 26 hours, no voids were missed, and urine creatinine was at least 10 mg/kg/day.

The following secondary endpoints were included in plans to control the overall type 1 error rate:

- Absolute change in 24-hour urinary oxalate corrected for BSA from baseline to Month 6
- Percent change in 24-hour urinary Ox/Cr ratio from baseline to Month 6
- Percent change in plasma oxalate from baseline to Month 6 (changed from exploratory to secondary endpoint in Amendment 2)
- Proportion of patients with 24-hour urinary oxalate level at or below 1.5x upper limit of normal (ULN) at Month 6 (changed from below to at or below 1.5×ULN in Amendment 2)
- Proportion of patients with 24-hour urinary oxalate level at or below ULN at Month 6 (changed from below to at or below ULN in Amendment 2)
- Absolute change in plasma oxalate from baseline to Month 6 (changed from exploratory to secondary endpoint in Amendment 2)

The Agency previously advised the Applicant that secondary endpoints that analyze urinary oxalate data in different ways (b) (4) did not provide substantial additional information beyond that provided by the primary endpoint. In addition, plasma oxalate values are generally not markedly elevated in a population with relatively preserved kidney function, and it is not clear what size of a treatment effect would be clinically meaningful. As such, this review is limited to the secondary endpoint of proportion of patients with 24-hour urinary oxalate levels ≤ULN at Month 6, because the clinical review team believes it is a clinically meaningful endpoint that should be included in labeling.

6.2.2.2. Eligibility Criteria, ILLUMINATE-A

The study included adults and children ≥6 years of age with confirmed PH1 as documented by 24-hour urinary oxalate excretion ≥0.70 mmol/24 hr/1.73 m^2 and confirmed alanine-glyoxylate aminotransferase (AGXT) mutations, and an eGFR ≥30 mL/min/1.73 m^2 at screening (lowered from >45 mL/min/1.73 m^2 with Amendment 2).

The study excluded patients with systemic oxalosis (as determined by the investigator), liver or kidney transplant.

6.2.2.3. Statistical Analysis Plan, ILLUMINATE-A

Initial Statistical Analysis Plan and Amendments

The statistical analysis plan (SAP) for ILLUMINATE-A was originally submitted on March 19, 2019. An updated version was submitted on November 27, 2019 after the trial was fully enrolled to align secondary endpoints with protocol Amendment 2 and change sensitivity analyses.

Sample Size Calculation

The planned sample size was 30, which was expected to result in 90% power to detect a treatment difference of 37% at a 2-sided 5% significance level assuming a 17% and a 54% mean percent reduction from baseline to Month 6 in 24-hour urinary oxalate corrected for BSA in the placebo and lumasiran arms, respectively, with a common standard deviation (SD) of 25%. No sample size re-estimation was planned or performed.

Analysis Datasets

The primary efficacy analysis population was the full analysis set, defined as all randomized patients who received any amount of study drug. The full analysis set was also used for secondary endpoints based on urinary oxalate.

Primary Efficacy Analysis

The primary endpoint was the percent change from baseline to Month 6 in 24-hour urinary oxalate corrected for BSA. The primary comparison between the treatment arms was based on the percent changes at Months 3, 4, 5, and 6 from baseline, using a restricted maximum likelihood based MMRM approach. The primary estimate was the LS mean of the primary variable averaged over Months 3 to 6. The fixed effects in the model were treatment arm, and scheduled visits (Months 3, 4, 5, and 6). The random factor was variability between patients. The continuous, fixed covariate was baseline 24-hour urinary oxalate level corrected for BSA. The MMRM approach handles missing data based on missing at random assumption, which assumes that patients who dropout would respond similarly to the remaining patients in the same treatment group.

Two sensitivity analyses were conducted by adding the interaction term of visit and treatment to the primary MMRM model, to evaluate the sensitivity to the estimated treatment effect on the assumption that lumasiran reaches steady state of the treatment effect at Month 3 and maintains the effect through Month 6.

- Sensitivity Analysis 1 estimates the treatment effect of the primary endpoint without assuming equal treatment effect from Month 3 through Month 6. The analysis adds the interaction of visit and treatment to the primary MMRM model, when Month 3 through Month 6 data are used.
- Sensitivity Analysis 2, similar to Sensitivity Analysis 1, estimates the treatment effect of the primary endpoint without assuming equal treatment effect from Month 3 through Month 6, but includes all postbaseline data (including percent change from baseline at Months 1 and 2).

Subgroup Analyses

Analyses of the primary endpoint during the placebo-controlled period were prespecified for the following subgroups: age (6 to 11, 12 to 17 versus ≥18 years of age at screening), gender (male versus female), race (white versus non-white), baseline 24-hour urinary oxalate corrected for BSA (≤1.70 mmol/24 h/1.73 m^2 versus >1.70 mmol/24 h/1.73 m^2), baseline eGFR (≤60 versus >60 mL/min/1.73 m^2), history of renal stones (yes versus no), baseline vitamin B6 use (yes versus no), region 1 (North America [including United States and Canada] versus Other [outside of North America]), and region 2 (Europe versus Other [outside of Europe]).

Secondary Efficacy Analyses

Key secondary endpoints were tested in a hierarchical order at a 2-sided 0.05 significance level. As noted above, this review is limited to the secondary endpoint of proportion of patients with 24-hour urinary oxalate levels ≤ULN at Month 6. The proportions were tested using a CMH test, stratified by baseline urinary oxalate levels (≤1.70 versus >1.70 mmol/24 hr/1.73 m^2).

6.2.2.4. Results of Analyses, ILLUMINATE-A

Baseline Characteristics

Given the size of the trial, baseline demographics were reasonably well-balanced between the treatment arms (Table 8). Patients <18 years of age accounted for 56% of enrollment. Two-thirds of patients were male. Overall, 77% of patients were white, and 15% Asian. A third of patients were enrolled in the United States.

Table 8. Baseline Demographic Characteristics, Safety Population, ILLUMINATE-A

Characteristic	Lumasiran N=26	Placebo N=13
Sex, n (%)		
F	8 (30.8)	5 (38.5)
M	18 (69.2)	8 (61.5)
Age, years		
Mean (SD)	18.7 (11.5)	17.0 (15.2)
Median (min, max)	16.5 (6.0, 47.0)	11.0 (6.0, 60.0)
Age group (years), n (%)		
≥6 to <11	9 (34.6)	7 (53.8)
≥12 to <17	5 (19.2)	1 (7.7)
≥18 to <64	12 (46.2)	5 (38.5)
Race, n (%)		
Asian	3 (11.5)	3 (23.1)
Other	2 (7.7)	0
White	21 (80.8)	9 (69.2)
Multiple	0	1 (7.7)
Country of participation, n (%)		
Switzerland	1 (3.8)	0
Germany	1 (3.8)	0
France	1 (3.8)	3 (23.1)
Great Britain	4 (15.4)	3 (23.1)
Israel	5 (19.2)	2 (15.4)
Netherlands	3 (11.5)	2 (15.4)
United States	11 (42.3)	2 (15.4)
Arab Emirates	0	1 (7.7)

Source: adsl.xpt; Software: Python.
Abbreviations: N = number of subjects in treatment group; n = number of subjects with given characteristic; SD = standard deviation.

As shown in Table 9, baseline disease characteristics were reasonably well balanced between the treatment arms. The Applicant stratified randomization by mean 24-hour urine oxalate (≤1.7 versus >1.7 mmol/24 h/1.73 m^2). At baseline, the mean (SD) 24-hour urinary oxalate excretion was 1.82 (0.62) mmol/24 hr/1.73 m^2 in both arms with 15 patients (58%) in the lumasiran group and 6 in placebo (46%) having a mean 24-hour urinary oxalate level >1.70 mmol/24 hr/1.73 m^2. The ULN is 0.514 mmol/24 hr/1.73 m^2.

Overall, 32 patients (82%) had an eGFR ≥60 mL/min/1.73 m^2 and 7 (18%) had an eGFR 30 to <60 mL/min/1.73 m^2.

At baseline, 22 patients (56%) reported pyridoxine use.

Table 9. Baseline Disease Characteristics, Safety Population, ILLUMINATE-A

Characteristic	Lumasiran N=26	Placebo N=13
24 hr urinary oxalate (mmol/24 hr/1.73 m^2)		
Mean (SD)	1.8 (0.6)	1.8 (0.7)
Median (min, max)	1.8 (0.8, 3)	1.7 (0.7, 2.8)
Mean 24 hr urinary oxalate (mmol/24 hr/1.73m^2) n (%)		
≤1.70	11 (42.3)	7 (53.8)
>1.70	15 (57.7)	6 (46.2)
Plasma oxalate (µmol/L)		
Mean (SD)	14.8 (7.6)	15.5 (7.3)
Median (min, max)	13.1 (7, 43.5)	13.1 (7.8, 28.4)
eGFR (mL/min/1.73 m^2)		
Mean (SD)	83 (25.3)	78.9 (27.9)
Median (min, max)	84.7 (31.7, 131.4)	76.1 (36.3, 125.6)
eGFR (mL/min/1.73 m^2), n (%)		
≥90	9 (34.6)	4 (30.8)
60 to <90	13 (50)	6 (46.2)
45 to <60	2 (7.7)	1 (7.7)
30 to <45	2 (7.7)	2 (15.4)
Pyridoxine use at baseline, n (%)		
Yes	13 (50.0)	9 (69.2)
No	13 (50.0)	4 (30.8)
Patient-reported stone events, n (%)		
Symptomatic stone events	23 (88.5)	10 (76.9)
Lithotripsy or stone removal procedures in the 12 months before consent	4 (15.4)	3 (23.1)

Source: adsl.xpt; Software: Python.
Abbreviations: N = number of subjects in treatment group; n = number of subjects with given characteristic; SD = standard deviation.

Disposition

Screening Period

A total of 52 patients were screened (Table 10). Of those, 11 patients did not meet eligibility criteria: seven did not meet the minimum threshold for urinary oxalate, one did not have confirmation of a diagnosis of PH1, one was excluded based on liver test abnormalities, two had an eGFR ≤45 mL/min/1.73 m^2 before the minimum threshold was lowered to <30 mL/min/1.73 m^2), and two chose not to participate.

Table 10. Patient Screening and Randomization, ILLUMINATE-A

Screening Disposition	ILLUMINATE-A
Patients screened	52
Screening failures	13
Patients randomized	39

Source: adds.xpt and Clinical Study Report; Software: R.

Randomized Period

A total of 39 patients were randomized, 26 to lumasiran and 13 to placebo. All randomized patients received at least one dose of study drug. One patient in the lumasiran group ([redacted] (b) (6)) prematurely discontinued study drug after the first dose because of anxiety and needle phobia

and withdrew from the study after Month 3 because of inability to comply with protocol-specified testing. This patient's data through Month 3 were included in efficacy analyses and later data were considered missing at random, a reasonable assumption given the reason for discontinuation. One patient in the lumasiran group ([(b) (6)]) completed the placebo-controlled period but chose not to enter the extension period. The remaining 37 patients entered the extension period.

Adequacy of 24-hour Urine Collections

24-hour urine collections were considered valid if the duration of collection was 22 to 26 hours, no voids were missed, and urine creatinine was at least 10 mg/kg/day. Only values from valid urine collections were included in the analyses. Invalid urine collections that could not be validated with a repeat collection were considered missing.

Missing data were rare. Thirty-five patients (90%) had no missing data at baseline and Months 3 to 6. All 39 enrolled patients (100%) had valid data at baseline and at least one valid assessment between Months 3 and 6 included in the MMRM model for the primary efficacy analysis. Four patients in the lumasiran group had missing data for at least one month during Months 3 to 6 ([(b) (6)] discontinued the study after Month 3 and did not have data for Months 4 to 6; [(b) (6)] had missing data at Month 3; [(b) (6)] had missing data at Month 4; [(b) (6)] had missing data at Months 3 and 4).

Analysis of Primary Endpoint

The primary endpoint was percent change in 24-hour urinary oxalate excretion corrected for BSA from baseline to Month 6 (LS means derived from MMRM model averaged across Months 3 to 6). As shown in Table 11, there was a statistically significant decrease in 24-hour urinary oxalate excretion corrected for BSA in the lumasiran group compared with placebo (LS mean difference -53.6%; $p < 0.0001$). The findings were consistent in prespecified sensitivity analyses that did not assume the treatment effect was at steady state from Months 3 to 6 (Sensitivity Analysis 1) and included data from Months 1 to 6 (Sensitivity Analysis 2).

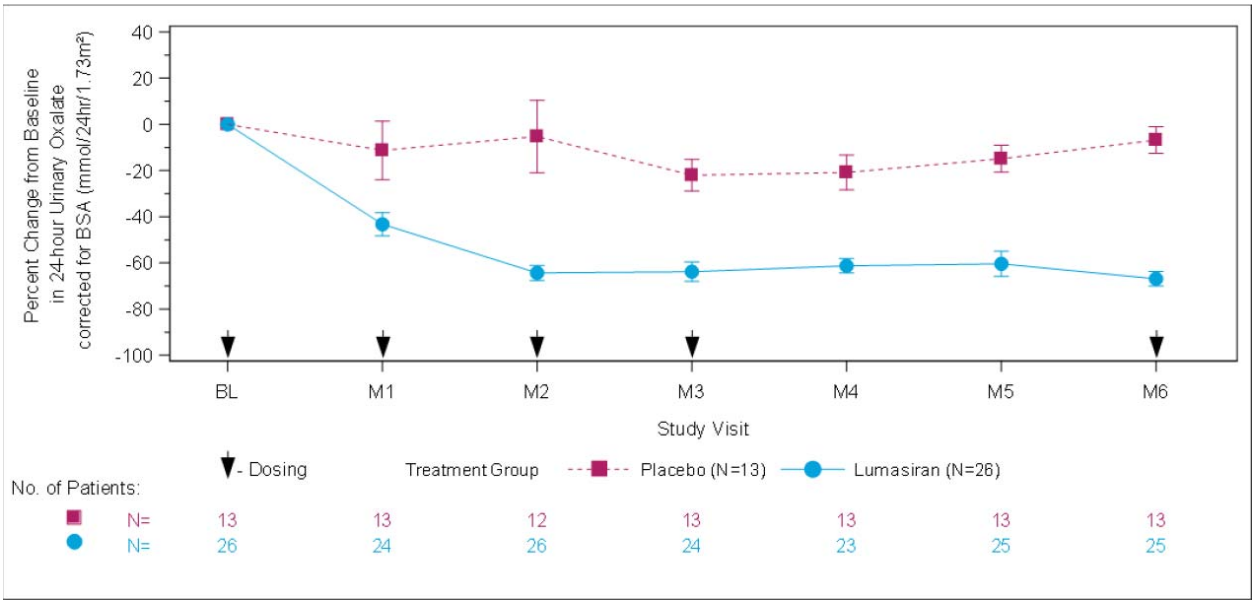
Table 11. Primary Efficacy Analyses: Percent Change From Baseline in 24-Hour Urinary Oxalate Corrected for BSA (mmol/24 hr/1.73 m^2) During the Placebo-Controlled Period, MMRM (Full Analysis Set), ILLUMINATE-A

Analysis	Statistic	Placebo N=13	Lumasiran N=26
Baseline	24-hour urinary oxalate (mmol/24 hr/1.73 m^2) mean (SEM)	1.79 (0.19)	1.84 (0.12)
Primary analysis	Percent change from baseline LS Mean (SEM)	-11.8 (3.8)	-65.4 (2.9)
	95% CI	(-19.5, -4.2)	(-71.3, -59.5)
	Difference in LS mean (SEM) (lumasiran-placebo)	-53.6 (4.3)	
	95% CI	(-62.3, -44.8)	
	p-value	<0.0001	
Sensitivity analysis 1	Percent change from baseline LS Mean (SEM)	-16.4 (4.3)	-63.0 (3.1)
	95% CI	(-25.1, -7.7)	(-69.3, -56.8)
	Difference in LS mean (SEM) (lumasiran-placebo)	-46.6 (5.3)	
	95% CI	(-57.3, -35.9)	
	p-value	<0.0001	
Sensitivity analysis 2	Percent change from baseline LS Mean (SEM)	-16.4 (4.3)	-63.1 (3.1)
	95% CI	(-25.1, -7.8)	(-69.3, -56.9)
	Difference in LS mean (SEM) (lumasiran-placebo)	-46.6 (5.3)	
	95% CI	(-57.3, -36.0)	
	p-value	<0.0001	

Source: Sponsor's table verified by statistical reviewer.
Abbreviations: CI = confidence interval; LS = least squares; N = number of subjects in treatment group; n = number of subjects with given characteristic; SEM = standard error of mean.

Figure 6 shows percent changes in 24-hour urinary oxalate at each study visit during the placebo-controlled period. For patients treated with lumasiran, urinary oxalate decreased through Month 2, and the effect persisted for the duration of the treatment period.

Figure 6. Percent Change From Baseline in 24-Hour Urinary Oxalate Corrected for BSA (mmol/24 hr/1.73 m^2) During the Placebo-Controlled Period, Based on the Observed Values (FAS), ILLUMINATE-A

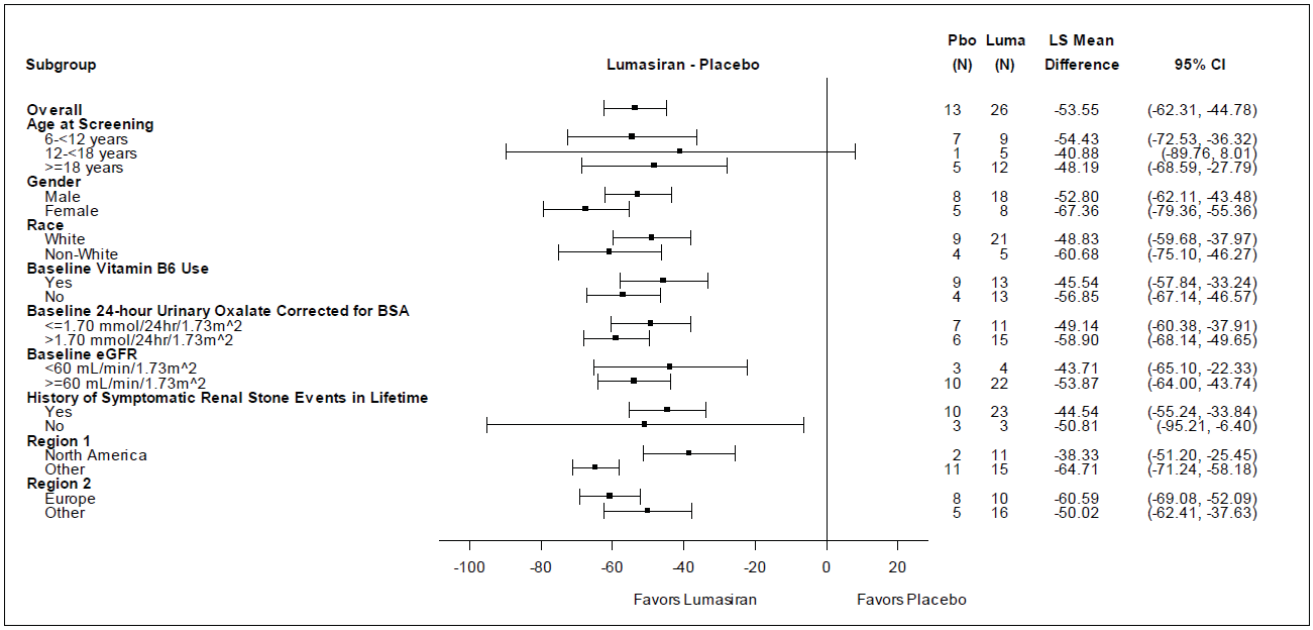


Source: Sponsor's figure verified by statistical reviewer.
Abbreviations: BL = baseline; BSA = body surface area; DB = double-blind; FAS = Full Analysis Set; M = month; SEM = standard error of mean.
BL represents the baseline value; median of all valid assessments collected prior to the first dose of study drug (lumasiran or placebo) in the DB Period without any non-protocol-related sample issues.
Results are plotted as mean (±SEM) of percent change from baseline.
Arrows indicate dosing. Patients received loading doses of 3.0 mg/kg lumasiran or placebo monthly for 3 doses (at baseline [Day 1] and Months 1 and 2) followed by the first maintenance dose of 3.0 mg/kg lumasiran or placebo at Month 3. The Month 6 dose defines the beginning of the Extension Period.

Subgroup Analyses of Primary Endpoint

The primary endpoint findings were generally consistent across the prespecified subgroups (Figure 7). Results were similar for patients taking pyridoxine (vitamin B6) compared with those not taking pyridoxine and across categories based on baseline kidney function and baseline 24-hour urinary oxalate.

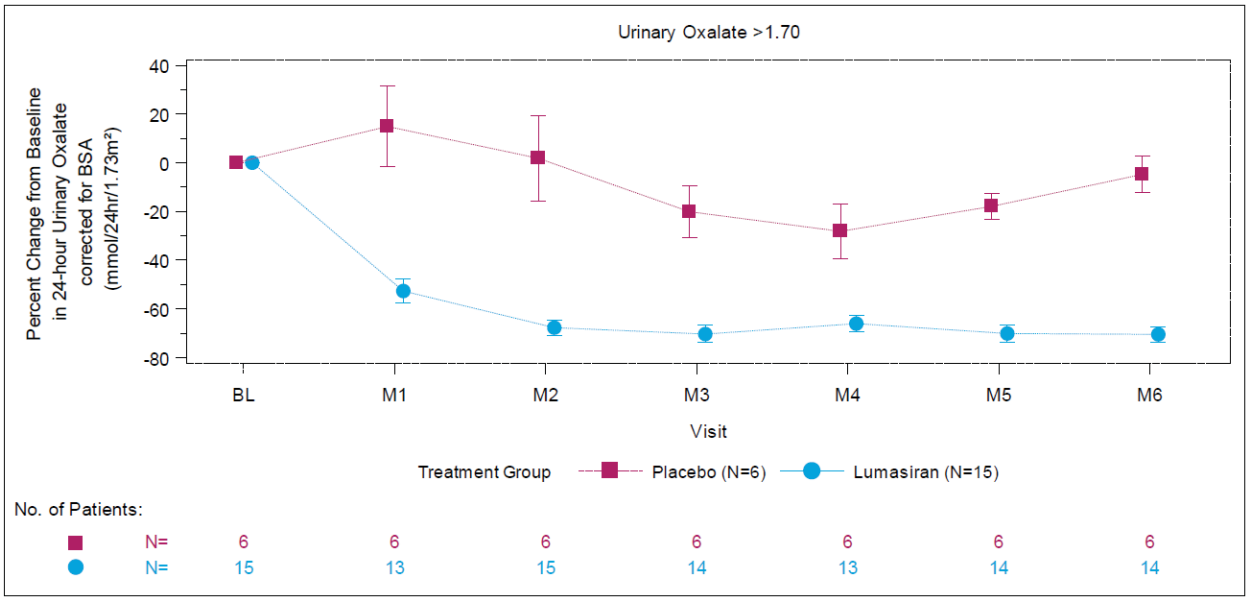
Figure 7. Subgroup Analysis: Difference Between Lumasiran and Placebo in Percent Change From Baseline in 24-Hour Urinary Oxalate (mmol/24 hr/1.73 m^2), ILLUMINATE-A



Source: Sponsor's figure verified by statistical reviewer.
Abbreviations: BSA = body surface area; eGFR = estimated glomerular filtration rate; LS = least squares; N = number of subjects in treatment group.

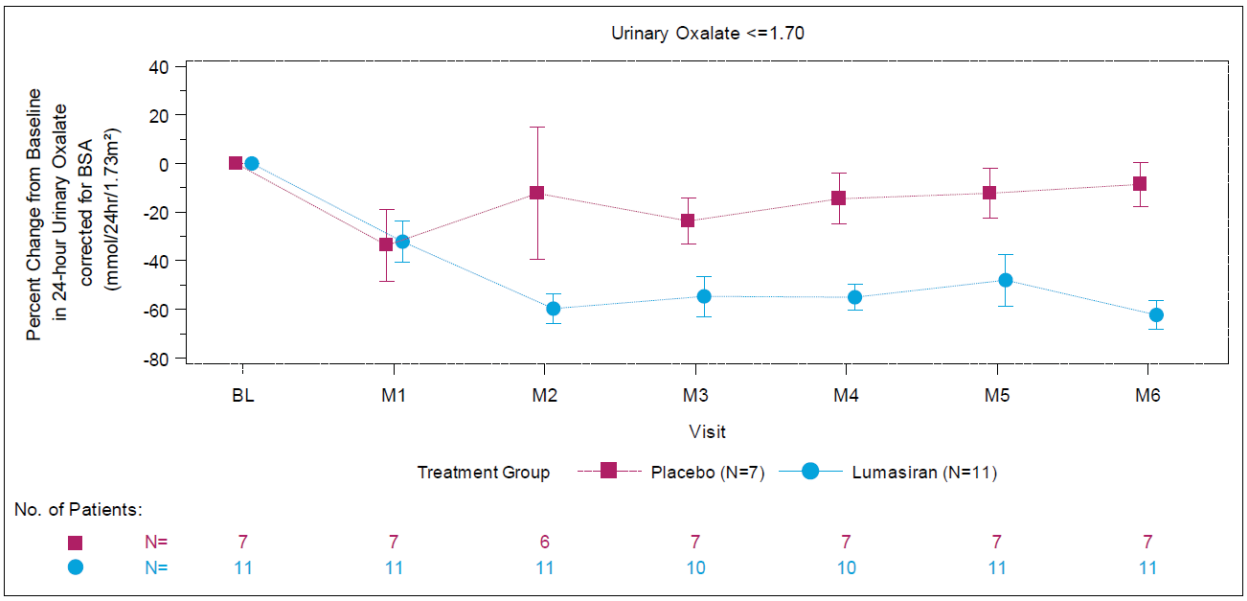
Figure 8 and Figure 9 show change in 24-hour urinary oxalate over time by baseline urinary oxalate category (>1.70 versus ≤1.70 mmol/24 hr/1.73 m^2). Of note, patients with higher baseline urinary oxalate values in the lumasiran group had a LS mean (SEM) percent decrease from baseline to Month 3 to 6 in 24-hour urinary oxalate of 72% (3%) compared with 13% (4%) for patients receiving placebo, a between-group difference of 59% (4%).

Figure 8. Percent Change From Baseline in 24-Hour Urinary Oxalate Corrected for BSA in Patients With Baseline Urinary Oxalate >1.7 mmol/24 hr/1.73 m^2 During the Placebo-Controlled Period, Based on the Observed Values (FAS), ILLUMINATE-A



Source: Sponsor's figure verified by statistical reviewer.
Abbreviations: BL = baseline; BSA = body surface area; DB = double-blind; FAS = Full Analysis Set; M = month; SEM = standard error of mean.
BL represents the baseline value; median of all valid assessments collected prior to the first dose of study drug (lumasiran or placebo) in the DB Period without any nonprotocol-related sample issues.
Results are plotted as mean (±SEM) of percent change from baseline.
Arrows indicate dosing. Patients received loading doses of 3.0 mg/kg lumasiran or placebo monthly for 3 doses (at baseline [Day 1] and Months 1 and 2) followed by the first maintenance dose of 3.0 mg/kg lumasiran or placebo at Month 3. The Month 6 dose defines the beginning of the Extension Period.

Figure 9. Percent Change From Baseline in 24-Hour Urinary Oxalate Corrected for BSA in Patients With Baseline Urinary Oxalate ≤1.7 mmol/24 hr/1.73 m^2 During the Placebo-Controlled Period, Based on the Observed Values (FAS), ILLUMINATE-A



Source: Sponsor's figure verified by statistical reviewer.
Abbreviations: BL = baseline; BSA = body surface area; DB = double-blind; FAS = Full Analysis Set; M = month; SEM = standard error of mean.
BL represents the baseline value; median of all valid assessments collected prior to the first dose of study drug (lumasiran or placebo) in the DB Period without any nonprotocol-related sample issues.
Results are plotted as mean (±SEM) of percent change from baseline.
Arrows indicate dosing. Patients received loading doses of 3.0 mg/kg lumasiran or placebo monthly for 3 doses (at baseline [Day 1] and Months 1 and 2) followed by the first maintenance dose of 3.0 mg/kg lumasiran or placebo at Month 3. The Month 6 dose defines the beginning of the Extension Period.

Analyses of Secondary Endpoints

Because the primary endpoint was successful, the secondary endpoints were tested hierarchically at a 2-sided alpha of 0.05. We limited our review to the secondary endpoint of proportion of patients with 24-hour urinary oxalate levels ≤ULN at Month 6, because we believe it is a clinically meaningful endpoint that should be included in labeling. As shown in Table 12, 13 of 25 (52%) lumasiran-treated and no placebo-treated patients achieved a 24-hour urinary oxalate level ≤ULN at Month 6 (95% CI 0.23, 0.70; p=0.001[1]). Of note, 5 of 14 (36%) patients with a baseline 24-hour urinary oxalate of >1.70 mmol/24 hr/1.73 m^2 achieved a value ≤ULN by Month 6.

[1] Given the 0 count in the placebo group, the p-value for the common odds ratio was calculated using exact statistics. The exact confidence interval for the risk difference was also calculated in SAS (0.19, 0.72), which is similar to the confidence interval reported in Table 12.

Table 12. 24-Hour Urinary Oxalate ≤ULN, ILLUMINATE-A

Statistic	Placebo	Lumasiran
Sample size, n	13	26
≤ULN at Month 6, n (%)		
Overall	0/13 (0)	13/25 (52.0)
By baseline 24-hour urinary oxalate corrected for BSA (mmol/24 hr/1.73 m^2)		
≤1.70	0/7 (0)	8/11 (72.7)
>1.70	0/6 (0)	5/14 (35.7)
Proportion of patients with urinary oxalate ≤ULN at Month 6	0	0.52
95% CI [a]	(0.00, 0.25)	(0.31, 0.72)
Difference in proportions (lumasiran-placebo)	0.52	
95% CI [b]	(0.23, 0.70)	
CMH test p-value [c]	0.001	

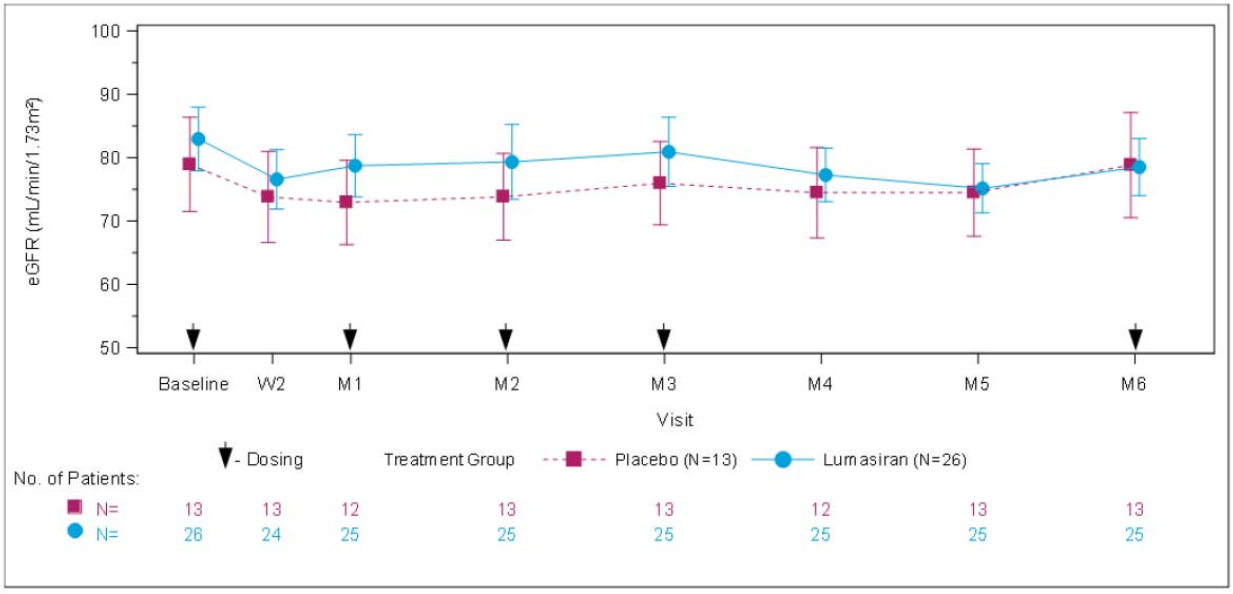
Source: Sponsor's table verified by statistical reviewer.
Abbreviations: BSA = body surface area; CI = confidence interval; CHM = Cochran-Mantel-Haenszel; N = number of subjects in treatment group; n = number of subjects with given characteristic; ULN = upper limit of normal.
[a] Clopper Pearson Exact confidence interval.
[b] Newcombe Method based on the Wilson Score.
[c] P-value is based on Cochran–Mantel–Haenszel test stratified by baseline 24-hour urinary oxalate corrected for BSA (≤1.70 vs. >1.70 mmol/24 hr/1.73 m^2).

Treatment Effects on Loss of Kidney Function and Kidney Stone Events, ILLUMINATE-A

Loss of Kidney Function

Kidney function remained stable in both treatment groups during the trial (Figure 10). Given that the decline in kidney function seen in patients with PH1 is generally slow, evidence of a treatment effect on kidney function was not expected.

Figure 10. eGFR (mL/min/1.73m^2) by Visit during the Placebo-Controlled Period, FAS



Source: Applicant's table (Clinical Study Report ALN-GO1-003 CSR1), verified by statistical reviewer.

Kidney Stone Events

Kidney stones events were identified by investigators based a visit to a healthcare provider (e.g., outpatient clinic, urgent care, or emergency department visit or procedure) for a kidney stone, medication for "renal colic," stone passage, or macroscopic hematuria due to a kidney stone.

Overall, eight patients (31%) in the lumasiran group and three (23%) in the placebo group experienced stone events during the 6-month placebo-controlled period (13 versus 4 stone events, respectively) (Table 13). Evidence of a treatment effect on kidney stone events was not expected given that calcium oxalate stones are slow to form and pass.

Table 13. Kidney Stone Events, Placebo-Controlled Period by Subgroup (Prior Kidney Stone Event Versus No Prior Stone Events), Full Analysis Set, ILLUMINATE-A

Stone events	Placebo N=13	Lumasiran N=26
Patients with Stone Events Overall, n/N (%)	3/13 (23)	8/26 (31)
In patients without event in preceding year, n/N (%)	1/9 (11)	1/15 (7)
In patients with event in preceding year, n/N (%)	2/4 (50)	7/11 (64)
Number of Stone Events	4	13

Source: Applicant's table, Clinical Study Report ALN-GO1-003 CSR1
Abbreviations: N = number of subjects in treatment group; n = number of subjects with given characteristic

6.2.3. ALN-GO1-004 (ILLUMINATE-B)

Overview of Study

In support of the indication in patients <6 years of age, the Applicant provided data from the initial 6-month portion of a single arm phase 3 trial titled "ILLUMINATE-B: An Open-Label Study to Evaluate the Efficacy, Safety, Pharmacokinetics, and Pharmacodynamics of Lumasiran in Infants and Young Children with Primary Hyperoxaluria Type 1." The trial is being conducted at nine sites in five countries, including two sites in the United States, four in Europe and three in the Middle East.

Initial Protocol and Amendments

The original protocol was submitted on December 20, 2018 and amended once (Amendment 1) on August 9, 2019. The amendment increased the sample size from eight to 20 based on a low screen failure rate, added effects on plasma oxalate as a secondary endpoint, and updated the statistical section to specify an MMRM analysis and define the "Efficacy Analysis Set" as all patients who received any amount of lumasiran and had at least one valid Ox/Cr value at baseline and Month 3 to 6. The trial had enrolled more than the originally specified sample size at the time of the amendment; however, no patients were excluded from efficacy analyses based on the revised "Efficacy Analysis Set" and the statistical model is similar to that used in ILLUMINATE-A. The overview provided in this section is based on the original protocol with amendments noted.

6.2.3.1. Design, ILLUMINATE-B

ILLUMINATE-B is an ongoing phase 3, single-arm study to evaluate the efficacy, safety, PK and PD of lumasiran in children birth to <6 years of age with PH1, a confirmed AGXT mutation, Ox/Cr in two out of three single-void collections during screening >ULN for age, and an eGFR >45 mL/min/1.73 m^2 in patients ≥12 months of age or serum creatinine ≤ULN for age at screening in patients <12 months of age. Patient eligibility was confirmed during a 60-day screening period, similar to ILLUMINATE-A. For patients taking pyridoxine (vitamin B6), doses had to be stable for at least 90 days before screening and remain stable until Month 6. The dose of pyridoxine could be adjusted for interval weight gain. Patients continued their standard of care regimens, including hyperhydration and crystallization inhibitors. Notable exclusions were similar to ILLUMINATE-A.

All patients received lumasiran monthly for three months then patients ≥10 kg switched to dosing every three months while patients <10 kg continued monthly dosing (Table 14). Patients who crossed weight thresholds followed the new dosing regimen. The study was divided into a 6-month primary analysis period followed by an extension period of up to 54 months.

Table 14. Weight-Based Lumasiran Dosing

Weight (kg)	Loading Dose (Day 1, Month 1, Month 2)	Maintenance Dose (Month 3 and Beyond)
<10	6 mg/kg	3 mg/kg monthly
≥10 to <20	6 mg/kg	6 mg/kg every 3 months
≥20	3 mg/kg	3 mg/kg every 3 months

Source: Sponsor's study protocol.

Study Objectives

As for ILLUMINATE-A, the primary objective was to evaluate the effect of lumasiran on percent reduction in urinary oxalate excretion.

The secondary objectives were to:

- Evaluate the effect of lumasiran on other measures of urinary oxalate excretion and plasma oxalate (Amendment 1 changed the evaluation of the effect of lumasiran on plasma oxalate from an exploratory to secondary objective)
- Characterize the PK of lumasiran
- Evaluate the effect of lumasiran on renal function

Study Endpoints

The primary endpoint was percent change in urinary oxalate excretion, measured by Ox/Cr, from baseline to Month 6 (LS means derived from MMRM model averaged across Months 3 to 6; Amendment 1 added a MMRM approach to the original descriptive primary analysis and changed the efficacy analyses from the Safety to the Efficacy Analysis Set). Triplicate single-void urine samples, preferably first morning, were obtained for oxalate during screening (baseline) and monthly within seven days before lumasiran dosing. The mean value of the triplicate samples was used in the analyses.

All secondary endpoints were either descriptive or other analyses of urinary oxalate excretion and were not intended to support labeling claims; therefore, they will not be addressed further in this review.

6.2.3.2. Eligibility Criteria, ILLUMINATE-B

The study included children from birth (estimated gestational age at least 37 weeks) to <6 years of age with confirmed PH1 as documented by urinary oxalate-to-creatinine ratio greater than the ULN for age on at least two of three single-void collections during screening and a confirmed alanine glyoxylate aminotransferase (AGXT) mutation.

The study excluded patients with systemic oxalosis (as determined by the investigator), prior liver or kidney transplant, eGFR ≤45 mL/min/1.73 m^2 in patients ≥12 months of age, or serum creatinine >ULN for age at screening in patients <12 months of age.

6.2.3.3. Statistical Analysis Plan, ILLUMINATE-B

Initial Statistical Analysis Plan and Amendments

The original SAP (Version 1.0) was dated May 20, 2019 and was amended once (Version 2.0) on November 12, 2019 to increase sample size from 8 to 20 and add a "Primary Interim Efficacy Analysis" to support the NDA submission.

Sample Size Justification

The sample size was based on feasibility considerations, not power calculations.

Analysis Datasets

The primary interim efficacy analysis was performed on the Primary Interim Efficacy Analysis Set, defined as all the patients among the first 16 enrolled patients who received any amount of lumasiran and had at least one valid Ox/Cr value at baseline and at least one valid Ox/Cr value between Months 3 and 6.

The descriptive statistics of baseline demographics and clinical characteristics were based on the Safety Analysis Set, defined as all patients who received any amount of lumasiran during the study.

Primary Efficacy Analysis

A primary interim analysis was planned to be conducted when the first 16 enrolled patients completed their Month 6 assessment or discontinued treatment (if applicable). The analysis was similar to the primary efficacy analysis but was based on the Primary Interim Efficacy Analysis Set. The primary endpoint was percent change in urinary oxalate excretion from baseline to Month 6 (LS means derived from MMRM model averaged across Months 3 to 6), measured by the percent change from baseline in Ox/Cr (mmol/mmol). The primary analysis was performed using a restricted maximum likelihood based MMRM approach in which the fixed effects were scheduled visits and baseline Ox/Cr value (mmol/mmol), and the random factor was patient. Given the small sample size of the study, an autoregressive covariance structure was used to

model the within-patient error. The primary estimate was the Least Square mean of the primary variable averaged over Month 3 to 6.

The Applicant also conducted an analysis of change from baseline in ULN ratio (ratio of measured Ox/Cr to ULN) from Month 3 to 6 to adjust for the age-related decrease in Ox/Cr that occurs in the first few years of life as kidney function matures.

6.2.3.4. Results of Analyses, ILLUMINATE B

Baseline Characteristics

Of the 18 enrolled patients, 10 (56%) were female, and 16 (89%) were white (Table 15). The median age at consent was 50 months (range 3 to 72 months); two patients (11%) were 0 to <1 year of age, and two patients (11%) were 1 to <2 years of age. Three patients (17%) were <10 kg and two thirds (12 patients) were 10 to <20 kg. Two patients (11%) enrolled in the United States.

Table 15. Baseline Demographic Characteristics, Safety Population, ILLUMINATE-B

Characteristic	Lumasiran (Age <2 yr) N=4	Lumasiran (Age ≥2 to <6 yr) N=14	Lumasiran (Overall) N=18
Sex, n (%)			
F	1 (25.0)	9 (64.3)	10 (55.6)
M	3 (75.0)	5 (35.7)	8 (44.4)
Age, years			
Mean (SD)	1.0 (0.7)	3.9 (1.1)	3.3 (1.6)
Median (min, max)	1.0 (0.2, 1.9)	4.0 (2.0, 5.0)	4.0 (0.2, 5.0)
Age group			
0 to <1 yrs	2 (50.0)	0	2 (11.1)
1 to <2 yrs	2 (50.0)	0	2 (11.1)
2 to <6 yrs	0	14 (100.0)	14 (77.8)
Weight, kg n (%)			
<10	3 (75.0)	0	3 (16.7)
10 to <20	1 (25.0)	11 (78.6)	12 (66.7)
≥20	0	3 (21.4)	3 (16.7)
Race, n (%)			
Other	2 (50.0)	0	2 (11.1)
White	2 (50.0)	14 (100.0)	16 (88.9)
Country			
Germany	0	1 (7.1)	1 (5.6)
France	1 (25.0)	4 (28.6)	5 (27.8)
Great Britain	2 (50.0)	0	2 (11.1)
Israel	1 (25.0)	7 (50.0)	8 (44.4)
United States	0	2 (14.3)	2 (11.1)

Source: adsl.xpt; Software: Python
Abbreviations: N = number of subjects in treatment group; n = number of subjects with given characteristic; SD = standard deviation.

The mean (SD) baseline Ox/Cr was 0.63 (0.43) mmol/mmol (Table 16), with higher values in younger (1.2 [0.4] mmol/mmol) compared with older patients (0.5 [0.3] mmol/mmol). The ULN declines with age, ranging from 0.22 mmol/mmol in patients 1 to 6 months of age to 0.07 in

patients 5 to 6 years of age.[2] In patients ≥1 year of age for whom eGFR could be calculated, the mean (SD) eGFR was 113 (28) mL/min/1.73 m^2. Eleven patients (61%) reported pyridoxine use.

Table 16. Baseline Disease Characteristics, Safety Population, ILLUMINATE-B

Characteristic	Lumasiran (Age <2 yr) N=4	Lumasiran (Age ≥2 to <6 yr) N=14	Lumasiran (Overall) N=18
Spot urine oxalate: creatinine ratio (mmol/mmol)			
Mean (SD)	1.2 (0.4)	0.5 (0.3)	0.6 (0.4)
Median (min, max)	1.2 (0.7, 1.7)	0.4 (0.2, 1.2)	0.5 (0.2, 1.7)
Plasma oxalate (µmol/L)			
Mean (SD)	19.3 (9.8)	11.5 (4.3)	13.2 (6.5)
Median (min, max)	19.7 (7.1, 30.5)	10.5 (6.6, 19.9)	11.5 (6.6, 30.5)
eGFR (mL/min/1.73 m^2)			
Mean (SD)	118.4 (22.8)	112 (28.9)	112.8 (27.6)
Median (min, max)	118.4 (102.3, 134.5)	110.7 (64.7, 174.1)	110.7 (64.7, 174.1)
eGFR (mL/min/1.73 m^2), n (%)			
≥90	2 (50)	12 (85.7)	14 (77.8)
60 to <90	0	2 (14.3)	2 (11.1)
30 to <60	0	0	0
Missing*	2 (50)	0	2 (11.1)
Pyridoxine use at baseline, n (%)			
Yes	3 (75)	8 (57.1)	11 (61.1)
No	1 (25)	6 (42.9)	7 (38.9)

Source: adsl.xpt; Software: Python
Abbreviations: N = number of subjects in treatment group; n = number of subjects with given characteristic; SD = standard deviation.
*eGFR was not calculated in two patients <1 year of age.

Disposition

There were no screen failures. As of the data cutoff, 18 patients had enrolled and been treated with lumasiran, and 16 (89%) had completed the Month 6 visit and entered the Extension Period. The remaining two patients had completed the Month 2 visit. No patient had discontinued treatment or withdrew from the study (Table 17).

[2] ULN spot urinary oxalate-to-creatinine ratios (mmol/mmol): 1-6 months 0.22, 6-12 months 0.17, 1-2 years 0.13, 2-3 years 0.10, 3-5 years 0.08, 5-6 years 0.07 (Source: Applicant's SAP).

Table 17. Patient Disposition, Safety Analysis Set, ILLUMINATE-B

Disposition Category	Lumasiran N=18 n (%)
Patients enrolled	18 (100)
Safety analysis set	18 (100)
Primary interim efficacy analysis set	16 (89)
Discontinued study drug	0 (0)
Discontinued study	0 (0)

Source: Generated by statistical reviewer

Primary Interim Efficacy Results

As shown in Table 18, the primary interim efficacy analysis showed a LS mean percent change from baseline to Month 6 (average from Month 3 to 6) in Ox/Cr of -71% (SEM 3%; 95% CI -77, -65). The results were similar for the prespecified supplementary analysis intended to account for the natural decline in Ox/Cr with age. In this supplementary analysis, percent change from baseline in ULN ratio (defined as ratio of Ox/Cr to age-dependent ULN based on [Matos 1999]) was analyzed using a similar MMRM model. Results of the supplementary analysis showed a similar magnitude of reduction in percent change (LS mean of -69%) in ULN ratio (ratio of Ox/Cr to ULN) from baseline to Month 6 (average from Month 3 to 6) indicating that the impact of age-associated decline in urinary oxalate-to-creatinine ratio was minimal.

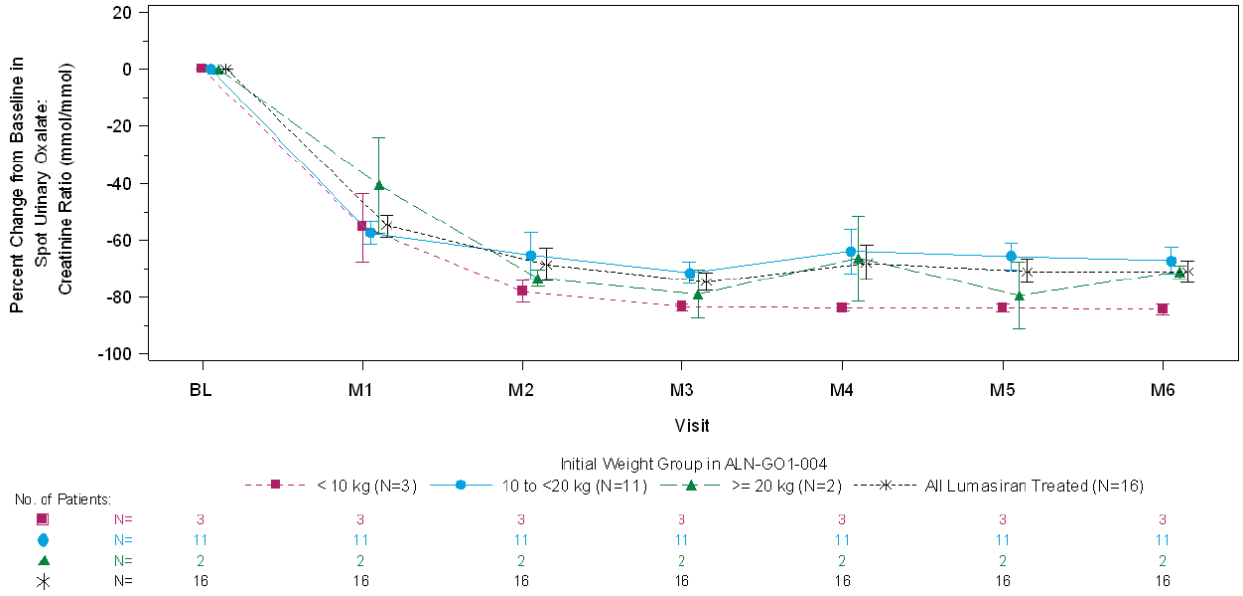
Table 18. Primary Interim Efficacy Results, Primary Interim Supplementary Analysis Set, ILLUMINATE-B

Analysis (N=16)	LS Mean (SEM)	95% CI
Percent change from baseline in spot urinary oxalate: creatinine ratio (mmol/mmol) to Month 6 (average from month 3 to month 6)	-71.1 (3.0)	(-77.2, -65.0)
Supplementary analysis: percent change from baseline in ULN ratio* to month 6 (average from month 3 to month 6)	-69.1 (2.7)	(-74.7, -63.5)

Source: Sponsor's table verified by statistical reviewer
Abbreviations: LS= least squares; SEM = standard error of the mean; ULN = upper limit of normal
*Ratio of spot urinary oxalate: creatinine to age-dependent ULN based on Matos et al. PMID 10430995

As shown in Figure 6, the results were consistent across subgroups based on weight. No additional subgroup analyses were conducted given the small sample size.

Figure 6. Mean (±SEM) Percent Change from Baseline in Spot Urinary Oxalate-to-Creatinine Ratio at Each Visit During the Primary Analysis Period (Primary Interim Efficacy Analysis Set), ILLUMINATE-B



Source: Sponsor's figure verified by statistical reviewer.
Abbreviation: M=month

6.3. Key Review Issues Relevant to Evaluation of Benefit

The review team has determined there are no review issues related to the evaluation of benefit that warrant discussion in this section. Discussion of considerations related to the design of the phase 3 studies including the use of urinary oxalate as a surrogate endpoint may be found in Section 1.

7. Risk and Risk Management

7.1. Potential Risks or Safety Concerns Based on Nonclinical Data

Overview

Lumasiran nonclinical safety was evaluated in the following studies during development: a safety pharmacology studies in monkeys; a single-dose toxicity study in rats; repeat-dose toxicity studies in rats and monkeys for durations up to 25 and 36 weeks, respectively; a full battery of genotoxicity studies (Ames, in vitro chromosomal aberration and in vivo rat micronucleus assays); a full battery of reproductive toxicity studies in the rat and rabbit; and a juvenile toxicity study in the rat. There were no nonclinical safety issues of concern as assessed by these studies. All pertinent studies and findings are summarized in the following section. Full reviews for all nonclinical studies are located in Section 13.1.

Overall, the nonclinical safety assessment for lumasiran was considered acceptable to support marketing approval from a pharmacology/toxicology perspective.

Safety Pharmacology and Pharmacokinetics/ADME

In a standalone cardiovascular safety pharmacology study in the monkey, the administration of lumasiran at 10 and 100 mg/kg weekly for a total of four doses had no immediate or delayed effects on clinical observations, qualitative or quantitative electrocardiogram parameters, hemodynamic parameters, respiratory rate, or body temperature. The no observed effect level dose was 100 mg/kg/week, the highest dose tested. In an 8-week repeated-dose toxicity study in the monkey, no lumasiran-related neurological observations were noted, and the no observed effect level was 100 mg/kg/week.

The plasma PK and toxicokinetic (TK) profiles of lumasiran were evaluated in rats and monkeys following single intravenous and SC administrations or repeat SC administrations given at pharmacologic doses ranging from 0.1 to 10 mg/kg. Lumasiran was rapidly eliminated from the systemic circulation after IV dosing with a clearance and plasma elimination half-life ($t_{1/2}$) of 646 mL/h/kg and 0.4 hours, respectively, in rats and 189 mL/h/kg and 0.6 hours, respectively, in monkeys.

Following a single SC administration in rats and monkeys, lumasiran was rapidly absorbed with a time to reach maximum concentration of 0.65 hours in rats and 2.3 hours in monkeys, with a dose-proportional increase in maximum observed plasma concentration (C_{max}) from 0.0218 to 2.320 µg/mL in both species. Plasma $t_{1/2}$ was shorter in rats (1.0 hour) compared with monkeys (4.7 hours) after SC administration. In general, there were no sex-related differences in the PK properties of lumasiran in rats and monkeys. Repeat-dose plasma PK parameter estimates of lumasiran following 8 once-weekly SC doses of 1 mg/kg and 3 once-monthly doses of 4 mg/kg in rats and monkeys were consistent with single dose PK parameters, indicating a linear PK. Due to the short plasma half-life of lumasiran compared with dosing frequency, there was no accumulation in rat plasma and minimal accumulation in monkey plasma (<2 fold increase) following repeat once weekly or once monthly dosing at the PK doses tested. Overall, the PK properties of lumasiran in rats and monkeys indicate no time- or dose-dependency following repeated SC dosing.

The metabolism of lumasiran was primarily due to nucleoside cleavage by exo- or endonucleases and was generally similar in vitro in both serum and liver S9 fraction obtained from mouse, rat, monkey, and human. The antisense strand was metabolized to metabolite AS(N-1)3' (loss of 1 nucleotide from the 3′ end of the antisense strand) lumasiran while the sense strand was metabolized by sequential removal of GalNAc moieties and removal of the 3' nucleotide.

The in vivo metabolism of lumasiran was assessed by analyzing plasma and liver samples after administration of lumasiran in rats and monkeys, as well as human plasma and urine in pooled samples (obtained from clinical study ALN-GO1-001). The in vivo metabolite profiling of plasma samples was similar across species, comparable to the in vitro serum samples with the antisense strand metabolized to AS(N-1)3' lumasiran while the sense strand was metabolized by removal of GalNAc sugars and removal of the 3' terminal nucleotide. The metabolite profiling of human plasma and urine in pooled samples showed primarily that the antisense strand of lumasiran was metabolized to AS(N-1)3' lumasiran but the concentration was <10% of lumasiran in both plasma and urine. In monkeys, a deaminated metabolite (resulting in terminal adenosine

to inosine conversion at the 3' end of the antisense strand) was observed in liver but was not observed circulating in plasma. Deaminated lumasiran was not detected in the plasma or liver of rats and was not observed in liver S9 homogenates in any species. However, deaminated lumasiran was detected in an in vitro human hepatocyte model (HepatoPac®), suggesting humans can form deaminated lumasiran. Both the AS(N-1)3' lumasiran metabolite and the deaminated metabolite are equipotent pharmacologically to lumasiran but are not major circulating plasma components in any species. In general, the data demonstrated that lumasiran was stable in plasma and was similarly metabolized to AS(N-1)3′ lumasiran in rats, monkeys, and humans, which was the primary circulating metabolite but was <10% of lumasiran exposure.

Renal excretion of lumasiran following administration of a single SC dose of 5 mg/kg was <1% of the administered dose in rats and approximately 20% in monkeys. The rat mass balance study with radiolabeled [^{14}C]-lumasiran administered at 10 mg/kg showed that the primary route of elimination of radioactivity after a single SC dose was in the feces, accounting for 33.9% of the dose through 1344 hours (56 days) post-dose. Urinary elimination in rats was 19.5% of the dose. Relatively high tissue concentrations (in descending order) were observed in the liver, kidney cortex, kidney medulla, cecum, salivary gland, bone marrow, thyroid and pancreas. Tissue concentrations were below the limit of quantification at all times for brain tissues (cerebrum, cerebellum and medulla) and spinal cord. In the monkey [^{14}C]-lumasiran mass balance study, approximately 38% of the dose was eliminated in the urine and 8% of the dose was eliminated in the feces. At the tissue level, only the liver and kidney samples were collected in the monkey mass balance study for radioactivity (drug distribution) analysis and showed that considerably higher levels of radioactivity were measured in the liver sample aliquots compared to the kidney aliquots at each timepoint post-dose. The highest concentration was observed at 24 hour post-dose, accounting for 54.7% of the radioactive dose. The terminal phase elimination half-life of total radioactivity in the liver was extensive (585 h).

General Toxicology (Pivotal Studies)

Chronic administration of lumasiran was assessed in good laboratory practice (GLP) repeat-dose toxicology studies up to 25- and 36-weeks in rats and monkeys, respectively. No target organs of toxicity were identified at the highest doses tested in these studies, but non-adverse, secondary findings were noted in the liver, kidney, coagulation system, site of injection, and lymph nodes, consistent with oligonucleotide class effects. The no observed adverse effect levels (NOAELs) in the chronic repeat-dose toxicity studies with once monthly dosing were the highest doses tested, 200 and 300 mg/kg for rats and monkeys, respectively. See Table 20 for the safety margins by exposure (area under the concentration-time curve [AUC]) and Table 21 for the safety margins by body surface area (BSA)-based dose scaling.

Table 19. Safety Margin by Plasma Exposure

Study	NOAEL (mg/kg)	Nonclinical Exposure at NOAEL (µg•hr/mL)	Safety Margins[a] (Multiples)
25-week rat	200	378	44 X
36-week monkey	300	1290	150 X

Source: FDA's analysis.
Abbreviations: NOAEL = no observed adverse effect level
[a] Exposure multiples were based on population PK analysis from phase 3 trials, where the maximum clinical dose resulted in systemic geometric mean exposures of $AUC_{0\text{-}24hr}$ of 8.56 µg•hr/mL for individuals weighing ≥70 kg, the body weight category in adults with the highest median exposure compared with other body weight categories supported by these pivotal tox studies.

Table 20. Safety Margin by Dose (BSA-based Scaling)

Study	NOAEL (mg/kg)	HED at NOAEL (mg/kg)	Safety Margins[a] (Multiples)
25-week rat	200	32	11 X
36-week monkey	300	97	32 X

Source: FDA's analysis.
Abbreviations: NOAEL = no observed adverse effect level
[a] Safety margins were based on the dose regimen in adult at 3 mg/kg, which is given less frequently in humans than in nonclinical studies.

Lumasiran, like other siRNAs, is quickly cleared from the plasma and taken up by the tissues. The tissue residence time seems very long (elimination half-life in the liver is 24 days), which is longer than the AUC distribution phase. Therefore, the distribution-phase plasma exposure levels provide a poor reflection of the tissue levels. For this reason, it is more appropriate to use BSA-based dose scaling for calculation of margins, which is also a more conservative estimate.

Juvenile Toxicology Study

Juvenile rats were dosed with lumasiran by weekly SC injection, beginning PND 4 to PND 32 (human equivalent age from newborn to approximately 6 years of age), at 0, 10, 30 and 100 mg/kg, for five doses total. This weekly SC administration of lumasiran was generally well tolerated. The only dose-dependent, statistically significant finding was decreases in body weight gain in female rats, with less than 8 (-7%) or 11 g below (-10%), respectively, those in concurrent controls. However, this is not considered adverse because over the course of the study female juvenile body weights continued to trend up towards controls levels and there were no histopathology or clinical pathology correlates.

Other findings include injection site reactions across all dose levels, minimal microscopic kidney and injection site findings across all dose levels, minimally to mildly decreased fibrinogen concentrations across all dose levels, and mildly increased glucose concentration in females administered ≥30 mg/kg. These findings were considered non-adverse given the minimal nature.

In general, the postdose liver and kidney concentrations indicated minimal or slight accumulation upon repeated weekly dosing with no apparent sex-related effects. Liver exposure increased in a less than dose-proportional manner and were higher than kidney exposure, while kidney exposure generally increased in a higher than dose-proportional manner from 10 to 100 mg/kg, possibly indicating saturation of uptake by the ASGR in hepatocytes. A dose-independent, non-sex specific liver HAO1 mRNA silencing (relative to baseline) of approximately 90% was observed on PND 33. Peak concentration C_{max} and AUC from 0 to 24 hours postdose ($AUC_{0\text{-}24}$) values achieved on PND 32 in animals administered 100 mg/kg were 23300 ng/mL and 74800 ng·hr/mL, respectively. This provides a safety margin of 7.4X based on mean AUC in children <6 years old between 10 and 20 kg (the body weight category with the highest exposure).

Table 21. Juvenile Toxicity Safety Margins

Study	Nonclinical Data	NOAEL (mg/kg)	Nonclinical Exposure (µg•hr/mL)	Safety Margins* (Multiples)
Juvenile rat	No mortalities and no dose-limiting toxicities. Female rats showed dose-dependent, mild to moderate body weight gain decreases at ≥30 mg/kg/week. Some oligo class effects (e.g., injection site reaction) were observed, but minimal in nature and not considered adverse.	100	74.8	9 X

Source: FDA's analysis.
Abbreviations: NOAEL = no observed adverse effect level
*Exposure multiples were based onpharmacokinetics analysis where a 6 mg q3M clinical dose resulted in a mean systemic exposure of $AUC_{0\text{-}24hr}$ = 8.3 µg•hr/mL in patients <6 years of age.

Genetic Toxicology and Carcinogenicity Study

Lumasiran was not genotoxic in an in vitro bacterial reverse mutation (Ames) assay, in vitro chromosomal aberration assay in cultured human peripheral blood lymphocytes, or in vivo micronucleus assay in rats. Two-year rat and 6-month transgenic mouse carcinogenicity studies are underway. The available data do not raise concern for a risk of carcinogenicity that should delay approval given the apparent clinical benefit and unmet need. As such, these studies will be postmarketing requirements.

Reproductive Toxicology

Reproductive toxicology studies did not reveal any serious fertility or teratogenic effects of lumasiran. A rat fertility and early embryonic development study showed no adverse effects with a safety margin of 6X. A rat embryo-fetal development (EFD) study had a NOAEL of 30 mg/kg with a safety margin of two. A rabbit EFD study had a NOAEL of 30 mg/kg with a safety margin of 8X. A pre- and postnatal development study had a NOAEL of 50 mg/kg with a safety margin of >2X. All the above safety margins were calculated based on body surface area scaling.

These studies are outlined in more detail in Section 8.

7.2. Potential Risks or Safety Concerns Based on Drug Class or Other Drug-Specific Factors

Potential Risks with GalNAC-conjugated siRNAs

AEs or toxicities associated with GalNAC-conjugated siRNA products include injection site reactions, ALT and AST elevations, mild increases in serum creatinine and decreases in eGFR, positive ADA, and hypersensitivity (including anaphylactic reactions).

Potential Risks Due to Drug-Specific Factors

Lumasiran inhibits GO, leading to accumulation of the precursor glycolate. Effects of chronic elevations in plasma or urine glycolate are unknown; however, the available data suggest these

elevations are likely to be benign. Glycolate is highly water soluble and readily excreted in urine. A case report was published in 2014 of an 8-year-old male with a homozygous splice site mutation in HAO1. He had marked elevations of urinary glycolate, no kidney stones, and normal kidney and liver function.[3] Similarly, GO-deficient mice (Hao1−/−) have asymptomatic marked urinary glycolate elevation without phenotypic differences in urinary sediment, kidney crystal deposition, kidney histology, growth, or development compared with Hao1+/+ mice.[4]

7.3. Potential Safety Concerns Identified Through Postmarket Experience

Lumasiran is not approved in the United States or any foreign market.

7.4. FDA Approach to the Safety Review

Data from the double-blind, placebo-controlled period of the ILLUMINATE-A trial and updated data from the single-arm ILLUMINATE-B study submitted June 23, 2020 were used to support the safety of lumasiran. In addition, the Safety Update Report submitted July 16, 2020 was reviewed for new findings with longer-term follow-up. Safety data from the two trials were not pooled given their differences in design. Analyses used the Safety Population, defined as all patients who received any amount of study drug.

The safety review focused on known and potential toxicities of lumasiran based on its mechanism of action, preclinical findings, and the experience with other GalNAC-conjugated siRNA products, including potential hepatic and kidney toxicity. In addition, we also considered AEs that were possibly drug-related, such as injection site reactions and hypersensitivity reactions, or resulted in study drug discontinuation. Adverse events (AE) were analyzed by Medical Dictionary for Regulatory Activities (MedDRA) preferred term and by pooling similar AEs (referred to as the FDA MedDRA Query [FMQ]). The FMQ analysis is similar to a customized MedDRA query. Adverse events of interest specified by the Applicant included severe or serious injection site reactions and liver toxicity (ALT or AST >3×ULN). A treatment-emergent adverse event (TEAE) was defined as an AE that occurred or worsened on or after the first dose of study drug, and any AE that was subsequently considered related to study drug.

7.5. Adequacy of Clinical Safety Database

The clinical safety database is limited but is considered adequate given the size of the affected population and the unmet need for a specific treatment. During the placebo-controlled period of ILLUMINATE-A, 26 patients ≥6 years of age received lumasiran for a mean duration of 5.5 months (Table 22).

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[3] Frishberg Y, Zeharia A, Lyakhovetsky R, Bargal R, Belostotsky R. Mutations in HAO1 encoding glycolate oxidase cause isolated glycolic aciduria. J Med Genet. 2014;51 (8):526-529.

[4] Martin-Higueras C, Luis-Lima S, Salido E. Glycolate Oxidase Is a Safe and Efficient Target for Substrate Reduction Therapy in a Mouse Model of Primary Hyperoxaluria Type I. Mol Ther. 2016;24 (4):719-725.

Table 22. Duration of Exposure, Safety Population, ILLUMINATE-A

Variable	Lumasiran N=26	Placebo N=13
Duration of treatment (months)		
Mean (SD)	5.5 (0.6)	5.6 (0.1)
Median (min, max)	5.6 (2.8-5.9)	5.6 (5.3-5.8)
Subjects treated, by duration, n (%)		
<3 months	1 (3.8)	0
≥3 to 6 months	25 (96.2)	13 (100)

Source: adex.xpt; Software: Python
Abbreviations: N = number of subjects in treatment arm; n = number of subjects with given treatment duration; SD = standard deviation.

After the 6-month placebo-controlled period of ILLUMINATE-A, 24 of 26 patients originally randomized to lumasiran and all patients randomized to placebo received lumasiran. According to the Safety Update Report, the mean (SD) and median (min, max) durations of exposure as of the February 14, 2020 cutoff date were 7.6 (3) months and 9.1 (2.8, 12.6) months, respectively, and 23 patients (59%) have been on study drug for 9 months or longer.

In the single-arm study, ILLUMINATE-B, 18 patients <6 years of age received lumasiran for a mean (SD) duration of 5.6 (1.6) months (Table 23). To date, 16 patients (89%) have completed the 6-month primary analysis period, and, of those, four patients (22%) have continued treatment for more than 6 months. The mean (SD) duration of exposure in the four patients <2 years of age is 6.8 (1.2) months, with three (75%) receiving lumasiran for >6 months.

Table 23. Duration of Exposure, Safety Population, ILLUMINATE-B

Variable	Lumasiran (Age<2 yr) N=4	Lumasiran (Age ≥2 to <6 yr) N=14	Lumasiran (Overall) N=18
Duration of treatment (months)			
Mean (SD)	6.8 (1.2)	5.3 (1.6)	5.6 (1.6)
Median (min, max)	6.6 (5.6, 8.4)	5.6 (1.9, 8.4)	5.6 (1.9, 8.4)
Subjects treated, by duration, n (%)			
<3 months	0	2 (14.3)	2 (11.1)
≥3 to 6 months	1 (25.0)	11 (78.6)	12 (66.7)
>6 months	3 (75.0)	1 (7.1)	4 (22.2)

Source: adex.xpt; Software: Python
Abbreviations: N = number of subjects in treatment arm; n = number of subjects with given treatment duration; SD = standard deviation.

7.6. Safety Findings and Concerns Based on Review of Clinical Safety Database

7.6.1. Safety Findings and Concerns, ILLUMINATE-A and -B

The review team did not conduct integrated analyses of safety because of differences in the design of the pivotal trials, ILLUMINATE-A and -B. See Sections 7.6.2 and 7.6.3 for analyses of individual trial results.

7.6.2. Safety Findings and Concerns, ILLUMINATE-A

7.6.2.1. Overall Treatment-Emergent Adverse Event Summary, ILLUMINATE-A

More patients had TEAEs in the lumasiran group compared with placebo (Table 24). All were mild or moderate in severity. There were no serious adverse events (SAE) or deaths. One patient in the lumasiran group had AEs leading to a missed dose.

Table 24. Overview of Adverse Events, Placebo-Controlled Period, Safety Population, ILLUMINATE-A

Event Category	Lumasiran (Overall) N=26 n (%)	Placebo (Overall) N=13 n (%)	Risk Difference (95% CI)[1]
Any AE	22 (84.6)	9 (69.2)	15.4 (-13.3, 44.1)
Mild AEs	15 (57.7)	7 (53.8)	3.9 (-29.2, 36.9)
Moderate AEs	7 (26.9)	2 (15.4)	11.5 (-14.4, 37.5)
Severe AEs	0	0	0.0 (0.0, 0.0)
Any SAE	0	0	0.0 (0.0, 0.0)
SAE with fatal outcome	0	0	0.0 (0.0, 0.0)
AE leading to discontinuation of study drug	0	0	0.0 (0.0, 0.0)
AE leading to interruption of study drug	1 (3.8)	0	3.8 (-3.5, 11.2)

Source: adae.xpt; Software: Python
Abbreviations: AE = adverse event; CI = confidence interval; N = number of subjects in treatment arm; n = number of subjects with at least one event; SAE = serious adverse event.
Treatment-emergent adverse events defined as Treatment-emergent adverse events defined as an AE that occurs or worsens on or after the first dose date/time of study drug though 84 days after the last dose of study drug. In addition, any AE that worsened in intensity or was subsequently considered related to study drug was considered treatment-emergent. Only includes DB period.
Grading Scale: AEs that occurred during the study were assessed by the Investigator for severity (i.e., mild, moderate, or severe).
[1] Risk difference column shows absolute difference (with 95% confidence interval) between total treatment and comparator.

7.6.2.2. Deaths, ILLUMINATE-A

There were no deaths in ILLUMINATE-A.

7.6.2.3. Serious Adverse Events, ILLUMINATE-A

There were no SAEs in ILLUMINATE-A.

7.6.2.4. Dropouts and/or Discontinuations Due to Adverse Events, ILLUMINATE-A

In general, lumasiran was well-tolerated. One patient missed a dose of lumasiran because of an AE but completed the double-blind treatment period ((b) (6)). The patient chose not to continue treatment in the open-label extension. One patient permanently discontinued lumasiran after the first dose ((b) (6)).

- (b) (6): 35-year-old male with PH1 and no other significant past medical history taking concomitant potassium citrate and pyridoxine. The patient-reported mild fatigue 9 hours after the first dose of lumasiran (Day 1), which resolved the next day. The patient reported moderate severity fatigue, disturbance of attention, and irritability 6 hours after the second dose of lumasiran (Month 1), which persisted for 27 days and led him to miss the Month 2 dose. Thyroid hormone, vitamin B12, cortisol, and CRP levels and testing for infectious diseases, including lyme, CMV, and tickborne encephalitis, were unrevealing. The patient restarted lumasiran (Month 3) and developed fatigue and disturbance of attention 26 days later. The symptoms resolved after 24 days but recurred 19 days later. As a result, the patient elected not to continue treatment in the open-label extension period but continued safety follow-up. The fatigue and disturbance of attention had resolved by study Day 189, 13 days after the Month 6 visit.

Although the events initially appeared to be temporally associated with lumasiran dosing, later events were not. No other patient had similar AEs. In the nonclinical program, there was no evidence that lumasiran crosses the blood-brain barrier. Additionally, a CNS safety pharmacology study in monkeys found no CNS effects such as behavioral, movement, or reflex abnormalities. It seems unlikely that these AEs were drug-related.

- (b) (6): 7-year-old female with PH1 and no other significant past medical history. The patient received one dose of lumasiran before discontinuing treatment permanently because of fear of injections. The parent/guardian withdrew the patient from the study after Month 3 because of inability to comply with study procedures.

7.6.2.5. Treatment-Emergent Adverse Events, ILLUMINATE-A

All TEAEs that occurred in two or more patients and with >5% risk difference in the lumasiran group compared with placebo fell within the FMQs reported in Table 25. The most common TEAEs were local administration reactions (injection site reactions) followed by abdominal pain and pneumonia. It is not clear how lumasiran could have caused pneumonia and, given the small numbers of events, it is likely the imbalance was due to chance; therefore, we will not discuss those events further. On review of all the TEAEs by System Organ Class and FMQ, we identified a hypersensitivity reaction as one additional AE of interest (Section 17, Table 71).

Table 25. Adverse Events by FDA Medical Query (Narrow) and Preferred Terms Occurring in Two or More Patients and With >5% Risk Difference in the Lumasiran Group Versus Placebo, Safety Population, ILLUMINATE-A

FMQ (Narrow) Preferred Term	Lumasiran (Overall) N=26 n (%)	Placebo (Overall) N=13 n (%)	Risk Difference (95% CI)[1]
Local administration reactions (narrow FMQ)	9 (34.6)*	0	34.6 (16.3, 52.9)
Injection site reaction	6 (23.1)	0	23.1 (6.9, 39.3)
Injection site erythema	3 (11.5)	0	11.5 (-0.7, 23.8)
Injection site pain	3 (11.5)	0	11.5 (-0.7, 23.8)
Injection site discomfort	1 (3.8)	0	3.8 (-3.5, 11.2)
Abdominal pain (narrow FMQ)	4 (15.4)	1 (7.7)	7.7 (-12.4, 27.7)
Abdominal pain	2 (7.7)	0	7.7 (-2.6, 17.9)
Abdominal pain upper	2 (7.7)	0	7.7 (-2.6, 17.9)
Abdominal pain lower	1 (3.8)	0	3.8 (-3.5, 11.2)
Abdominal discomfort	1 (3.8)	1 (7.7)	-3.8 (-20.1, 12.4)
Pneumonia (narrow FMQ)	2 (7.7)	0	7.7 (-2.6, 17.9)
Pneumonia	2 (7.7)	0	7.7 (-2.6, 17.9)

Source: adae.xpt; Software: Python
Abbreviations: CI = confidence interval; FMQ = FDA medical query; N = number of subjects in treatment arm; n = number of subjects with adverse event; SOC = system organ class.
For specific preferred terms under each FMQ, see the table "Adverse Events by FDA Medical Query (Narrow) and Preferred Term"
[1] Risk difference column shows absolute difference (with 95% confidence interval) between total treatment and comparator.
*During labeling discussions, the Applicant added one additional patient (b) (6)) in the lumasiran group (n=10, 38%) who experienced an injection site reaction during the placebo-controlled period. They noted "A new event of ISR was added post database lock." This patient was not included in our analyses. The injection site reaction (erythema, pruritus, rash) was mild, occurred with the third dose of lumasiran, lasted up to one week, and resolved spontaneously (submitted in SN0030 in response to an information request).
Treatment-emergent adverse events defined as Treatment-emergent adverse events defined as an AE that occurs or worsens on or after the first dose date/time of study drug though 84 days after the last dose of study drug. In addition, any AE that worsened in intensity or was subsequently considered related to study drug was considered treatment-emergent. Only includes DB period.

Local Administration Reactions (Injection Site Reactions)

Local administration reactions occurred more frequently in the lumasiran group (nine patients, 35%) compared with placebo (zero patients). Patients could receive up to four injections during the 6-month placebo-controlled period. Five of the nine patients (56%) experienced a reaction with only one dose, and two (22%) had a reaction with all four doses. Local administration reactions occurred with equal frequency throughout the study period with 5 (19%), 5 (20%), 3 (13%), and 5 (20%) patients experiencing reactions on Day 1, Months 1, 2, and 3, respectively. During the study, a total of 25 reactions were reported, of which 21 (84%) resolved within one day of the injection.

Abdominal Pain

During the placebo-controlled period, four patients (15%) in the lumasiran group and one (8%) in placebo reported AEs of abdominal pain or discomfort. All reported one event except patient (b) (6), who reported three. All were mild, did not lead to study drug interruption or discontinuation, and three of the four events with a reported duration lasted ≤1 day. There was no clear temporal relationship with lumasiran dosing or associated laboratory abnormalities, including AST, ALT, bilirubin, or alkaline phosphatase.

One additional patient ([(b) (6)]), reported abdominal pain during the extension period:

- [(b) (6)]: 22-year-old male with a BMI of 26 kg/m^2 on concomitant potassium citrate and paracetamol, originally randomized to lumasiran, reported two episodes of mild abdominal pain during the extension period. The patient was found to have hepatomegaly on ultrasound consistent with hepatic "steatosis." AST, ALT, and bilirubin were normal. The AE did not result in treatment interruption or discontinuation. The relationship of the patient's abdominal pain and hepatomegaly to lumasiran is unclear.

Hypersensitivity

One patient ([(b) (6)]), a 17-year-old male randomized to lumasiran, reported an AE of "allergic reaction unknown cause" (verbatim term; PT hypersensitivity) during the placebo-controlled period.

- [(b) (6)]: 17-year-old male with history of asthma, environmental and food allergies, penicillin allergy, and eosinophilia on concomitant diphenhydramine, salbutamol sulfate inhaler, and epinephrine-pen (as needed for food allergies) reported a single episode of "allergic reaction of unknown cause" on Day 32 one day prior to the second dose of lumasiran. Symptoms included stomach pain, throat pain, chest and back ache, and sneezing/difficulty breathing. An AE of upper respiratory tract infection was reported on Day 33. The patient received his second dose of lumasiran on Day 33. He completed the placebo-controlled period and remains in the extension period. The patient-reported AEs of injection site reactions (injection site pain [stinging at the injection site]) with onset at the time of lumasiran administration and resolution within 15 to 45 minutes. No AEs of allergic reactions were reported with or immediately after lumasiran injections. The patient's ADA assessments have been negative throughout the study. Given the patient's history of allergies, symptoms reported with the hypersensitivity AE, concomitant upper respiratory tract infection, and lack of temporal relationship with the lumasiran injection, the hypersensitivity AE was unlikely to have been related to lumasiran.

Hepatic Adverse Events

No AEs mapped to the Drug-related Hepatic Disorders SMQ during the placebo-controlled period. During the extension period, two patients ([(b) (6)] and [(b) (6)]) had AEs mapping to the Drug-related Hepatic Disorders SMQ.

- [(b) (6)]: See the narrative under the Abdominal Pain section above for details.
- [(b) (6)]: 25-year-old male originally randomized to placebo had an AE of moderate severity of increased AST (2.9×ULN) at the Month 6 visit before the first dose of lumasiran.

Kidney Adverse Events

During the placebo-controlled period, two patients (8%) in the lumasiran group reported AEs that mapped to the Renal and Urinary Disorders System Organ Class. One patient-reported renal pain and the other polyuria (Section 17, Table 71). These symptoms are common in patients with PH1 and are unlikely to be related to lumasiran. No AEs of glomerulonephritis or acute kidney

injury were reported (Renal and Urinary Disorders System Organ Class) during the placebo-controlled or extension periods.

7.6.2.6. Laboratory Findings, ILLUMINATE-A

Liver Tests

Most patients in both groups maintained ALT, AST, and total bilirubin values within the normal range during the trial, and there was no imbalance in the number with values exceeding normal (Table 26). No patient had an AST or ALT value >3×ULN, or total bilirubin >2×ULN.

Table 26. Worst Post-Baseline Liver Tests, Placebo-Controlled Period, Safety Population, ILLUMINATE-A

Laboratory Test		Lumasiran (Overall) N=26 n (%)	Placebo (Overall) N=13 n (%)
ALT	>ULN	4 (15.4)	2 (15.4)
	>3xULN	0	0
AST	>ULN	0	2
	>3xULN	0	0
Total bilirubin	>ULN	2 (7.7)	1 (7.7)
	>2xULN	0	0

Source: Applicant's Table 37 (Clinical Study Report ALN-GO1-003 CSR1). Verified by clinical reviewer.
Abbreviations: N = number of subjects with relevant laboratory data; n = number of subjects with abnormality.

Kidney Tests

There was no change in eGFR over time or between the treatment groups during the placebo-controlled period (Table 27). There were no significant differences in urinalysis noted (hematuria or proteinuria) by treatment arm (data not shown).

Table 27. Mean eGFR (SEM) Over Time, Placebo-Controlled Period, ILLUMINATE-A

Visit	Lumasiran N=26	Placebo N=13
Baseline	83.0 (5.0)	79.0 (7.4)
Week 2	76.6 (4.7)	73.8 (7.2)
Month 1	78.7 (4.9)	73.0 (6.7)
Month 2	79.3 (5.9)	73.8 (6.8)
Month 3	80.9 (5.5)	76.0 (6.6)
Month 4	77.3 (4.2)	74.5 (7.1)
Month 5	75.2 (3.9)	74.5 (6.9)
Month 6	78.5 (4.5)	78.8 (8.3)

Source: Sponsor's table verified by statistical reviewer.
Abbreviation: SEM = standard error of mean.

Immunogenicity

The Office of Biotechnology Products reviewed the Applicant's ADA assay, and noted it is suitable to detect ADAs against lumasiran.

Treatment-emergent ADAs to lumasiran were reported for one of 26 lumasiran-treated patients (4%) during the placebo-controlled period. The patient ([(b) (6)]) tested positive with a low titer (1:50) ADA at a single time point (Month 6). The presence of ADAs did not impact lumasiran PK or efficacy, and no AEs were reported for the patient. According to the Applicant, as of data

cutoff for the safety update report, the patient's follow-up ADA was negative, and no additional patients in the study had positive ADA.

Other

Review of other laboratory data did not reveal unexpected findings during the study.

7.6.3. Safety Findings and Concerns, ILLUMINATE-B

7.6.3.1. Overall Treatment-Emergent Adverse Event Summary, ILLUMINATE-B

All 18 enrolled patients (100%) had at least one TEAE during the study (Table 28). All were mild or moderate in severity. There was one SAE. There were no deaths, and no dose interruptions or discontinuations because of AEs.

Table 28. Overview of Adverse Events, Safety Population, ILLUMINATE-B

Event Category	Lumasiran (Age <2 yr) N=4 n (%)	Lumasiran (Age ≥2 to <6 yr) N=14 n (%)	Lumasiran (Overall) N=18 n (%)
Any AE	4 (100.0)	14 (100.0)	18 (100.0)
Mild AEs	4 (100.0)	14 (100.0)	18 (100.0)
Moderate AEs	2 (50.0)	3 (21.4)	5 (27.8)
Severe AEs	0	0	0
Any SAE	0	1 (7.1)	1 (5.6)
SAE with fatal outcome	0	0	0
AE leading to discontinuation of study drug	0	0	0
AE leading to dose modification of study drug	0	0	0
AE leading to interruption of study drug	0	0	0
AE leading to reduction of study drug	0	0	0
AE leading to delay of study drug	0	0	0

Source: adae.xpt; Software: Python
Abbreviations: AE = adverse event; N = number of subjects in treatment arm; n = number of subjects with at least one event; SAE = serious adverse event
Treatment-emergent adverse events defined as an AE that occurs or worsens on or after the first dose date/time of study drug though 84 days after the last dose of study drug. In addition, any AE that worsened in intensity or was subsequently considered related to study drug was considered treatment-emergent.
Grading Scale: Adverse events that occurred during the study were assessed by the Investigator for severity (i.e., mild, moderate, or severe).

7.6.3.2. Deaths, ILLUMINATE-B

There were no deaths in ILLUMINATE-B.

7.6.3.3. Serious Adverse Events, ILLUMINATE-B

There was one SAE with a preferred term of "viral infection" during the study:

- (b) (6): 4-year-old male with history of PH1, pseudocroup, stoma site ulcer, and nephrocalcinosis, on concomitant potassium citrate, magnesium citrate, macrogol, potassium chloride, sodium bicarbonate, xylometazoline and dexpanthenol, oxymetazoline hydrochloride, ibuprofen, and prednisolone. The patient received his fourth dose of lumasiran on Day 86. He was hospitalized on Day 150 with a several-day history of croup-like breathing, nasal congestion, vomiting, and anuria. He was treated with oxygen, antibiotics, intravenous hydration, and sodium bicarbonate. He was also found to have *Haemophilus influenzae* conjunctivitis. He recovered by Day 154 and was discharged from the hospital. There was no change in lumasiran dosing, and the patient remains on lumasiran in the study. Of note, after enrollment, and prior to the hospitalization, the patient had several similar nonserious AEs of upper respiratory tract infection, nasopharyngitis, bronchitis, pneumonia, and croup, which did not require hospitalization.

7.6.3.4. Dropouts and/or Discontinuations Due to Adverse Events, ILLUMINATE-B

There were no dropouts or discontinuations due to adverse events in ILLUMINATE-B.

7.6.3.5. Treatment-Emergent Adverse Events, ILLUMINATE-B

TEAEs that occurred in more than two patients are shown in Table 29. Most are common events in children (pyrexia, rhinitis, vomiting, and upper respiratory infection) and are unlikely to be drug-related. Injection site reactions occurred in three patients (17%); all were mild, and none led to discontinuation or interruption of lumasiran. There were no liver-related AEs.

Table 29. Treatment-Emergent Adverse Events Occurring in More Than Two Patients, Safety Population, ILLUMINATE-B

Preferred Term	Lumasiran (Age <2 yr) N=4 n (%)	Lumasiran (Age ≥2 to <6 yr) N=14 n (%)	Lumasiran (Overall) N=18 n (%)
Pyrexia	2 (50.0)	4 (28.6)	6 (33.3)
Rhinitis	2 (50.0)	2 (14.3)	4 (22.2)
Injection site reaction	1 (25.0)	2 (14.3)	3 (16.7)
Vomiting	1 (25.0)	2 (14.3)	3 (16.7)
Upper respiratory tract infection	0	3 (21.4)	3 (16.7)

Source: adae.xpt; Software: Python
Abbreviations: AE = adverse event; N = number of subjects in treatment arm; n = number of subjects with adverse event
Treatment-emergent adverse events defined as an AE that occurs or worsens on or after the first dose date/time of study drug though 84 days after the last dose of study drug. In addition, any AE that worsened in intensity or was subsequently considered related to study drug was considered treatment-emergent.

No additional TEAEs of interest were identified by reviewing AEs by system organ class and FMQ in the safety population as shown in Table 73 and Table 74 in Section 17.

7.6.3.6. Laboratory Findings, ILLUMINATE-B

Liver Tests

Most patients had ALT, AST, and total bilirubin values within the normal range during the trial, and no patient had an ALT or AST value >3×ULN or a total bilirubin >2×ULN (Table 30).

Table 30. Worst Post-Baseline Liver Tests, Safety Population, ILLUMINATE-B

Laboratory Test		Lumasiran (Age <2 yr) N=4 n (%)	Lumasiran (Age ≥2 to <6 yr) N=14 n (%)	Lumasiran (Overall) N=18 n (%)
ALT	>ULN	3 (75)	0	3 (17)
	>3xULN	0	0	0
AST	>ULN	3 (75)	5 (36)	8 (44)
	>3xULN	0	0	0
Total bilirubin	>ULN	0	1 (7)	1 (6)
	>2xULN	0	0	0

Source: adsl.xpt, lb.xpt; Software JMP Clinical
Abbreviations: N, number of subjects with relevant laboratory data; n, number of subjects with abnormality.
Clinical reviewer analyses.

Kidney Tests

In patients 1 year of age and older in whom eGFR could be calculated, there was no change in eGFR over time (Table 31).

Table 31. Mean eGFR mL/min/1.73 m^2 (SEM), ILLUMINATE-B

Visit	Lumasiran (Overall) N=17
Baseline	113.6 (7.3)
Month 1	105.8 (4.9)
Month 2	108.1 (5.0)
Month 3	104.4 (4.5)
Month 4	105.8 (4.9)
Month 5	110.8 (4.2)
Month 6	115.2 (5.6)

Source: adlb.xpt; Software JMP
Abbreviation: SEM=standard error of mean
Clinical reviewer analyses.

Immunogenicity

Three of 18 patients (17%) developed low-titer (1:50) ADAs. The ADAs were transient in two patients ((b) (6) and (b) (6)); the third ((b) (6)) developed ADA at the last visit before the data cutoff and has not had a follow-up measurement. ADAs did not affect safety, PK or PD, and all patients remain on lumasiran.

Other

Review of other laboratory data did not reveal unexpected findings during the study.

7.7. Key Review Issues Relevant to Evaluation of Risk

The review team has determined there are no review issues related to the evaluation of benefit that warrant discussion in this section. An overview of the clinical safety findings can be found in Section 1.

8. Therapeutic Individualization

8.1. Intrinsic Factors

Hepatic Impairment

The Applicant proposed the following language for use of lumasiran in patients with hepatic impairment (HI):

(b) (4)

No dedicated HI study in otherwise healthy subjects was conducted for lumasiran. Even if such a study were to be conducted, however, it would not provide information on liver exposures of lumasiran, which is the primary determinant of lumasiran's pharmacologic effect. The clinical experience with lumasiran in patients with PH1 and HI is limited to three patients randomized to lumasiran with reported urinary oxalate data, two with mild HI and one with moderate HI. Hence, the team reviewed existing literature to understand the impact of HI on ASGR-mediated uptake of GalNAc conjugated siRNAs and to evaluate Applicant's proposed dosing in patients with HI.

Lumasiran is a GalNAc conjugated siRNA that is preferentially taken up by the liver via ASGR receptors; therefore, any impact of HI on the expression and functional activity of ASGR could reduce liver uptake and the pharmacological effect. Sugahara et al. (PMID 14647051) evaluated the whole liver uptake of ASGR ligands in subjects with different degrees of HI. The study found whole liver uptake ratios in subjects with chronic hepatitis, Child-Pugh A, Child-Pugh B, and Child-Pugh C of 86%, 74%, 51%, and 33%, respectively, relative to normal controls, indicating that uptake was relatively preserved in those with mild HI (Child-Pugh A) (Table 32). The approved label for a similar GalNAc conjugated siRNA givosiran (Givlaari; NDA 212194) notes that there were no clinically meaningful differences in plasma PK or PD effects in patients with mild HI compared to patients with normal hepatic function. The two patients with PH1 and mild HI treated with lumasiran had reductions in urinary oxalate similar to patients with normal hepatic function, suggesting preserved hepatic uptake. Therefore, the review team agrees with Applicant that patients with mild HI do not require dose adjustment.

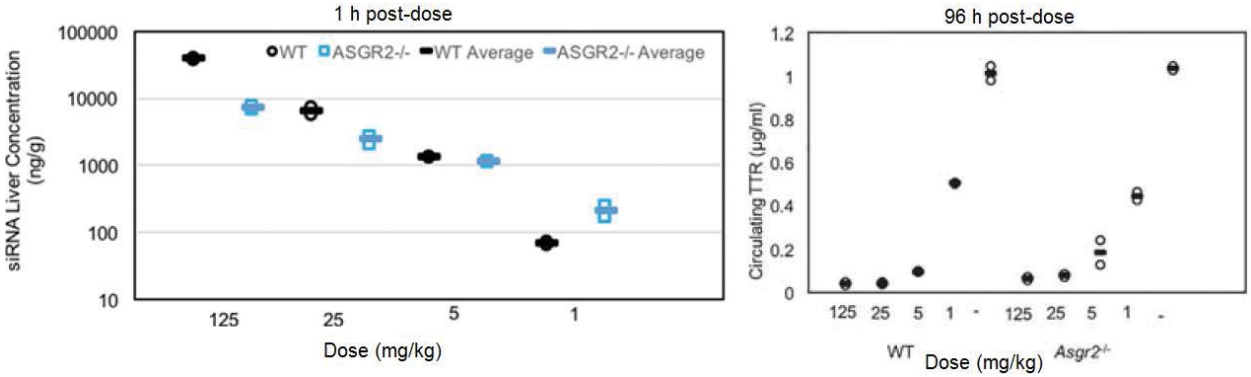
Table 32. Comparison of Mean Whole Liver ASGR-mediated Ligand Uptake Ratios Among Subjects with Varying Degrees of Hepatic Impairment Relative to Healthy Controls

Population	Uptake Ratio
Healthy controls (normal hepatic function)	100%
Chronic hepatitis	86%
Mild (Child-Pugh A)	74%
Moderate (Child-Pugh B)	51%
Severe (Child-Pugh C)	33%

Source: Sugahara et al., PMID:14647051

Considering the rarity and severity of PH1 and the unmet need, the team further explored the available literature to evaluate whether providing dosing recommendations to patients with moderate HI could be supported. Willoughby et al. (PMID 28988716) investigated the potential impact of ASGR receptor expression on the hepatic uptake and PD effects of GalNAc-conjugated siRNAs (targeting TTR and Apo B proteins) using preclinical models of reduced receptor expression. The study showed that ASGR knockdown by up to 60% in *Asgr2* knockout mice did not affect hepatocyte concentrations or the pharmacological activity of a GalNAc-conjugated siRNA, when administered at pharmacologically relevant doses up to 5 mg/kg. At higher doses (25 and 125 mg/kg), saturation in ASGR uptake was observed leading to lower hepatocyte concentrations in the *Asgr2* knockout mice; however, there was no corresponding decrease in efficacy (circulating target protein level) compared with the lower dose. If the siRNA was potent enough to achieve complete inhibition of target mRNA at lower doses (lower hepatocyte concentrations), then saturation of ASGR uptake was unlikely to impact the efficacy, as shown in Figure 11.

Figure 11. Comparison of the GalNAc-siRNA Concentration in the Liver (Left Panel) and Circulating Transthyretin Levels (PD) in Asgr2 Knockout and Wild Type Mice



Source: Willoughby et al., (PMID 28988716)

The recommended dose for lumasiran of 3 to 6 mg/kg based on body weight is in a similar range as the drug shown not to be affected by a moderate level of ASGR knockdown expected in patients with moderate HI. The patient with PH1 and moderate HI treated with lumasiran had a reduction in urinary oxalate similar to patients with normal hepatic function. Patients with moderate HI may have an increase in the plasma PK of lumasiran because of slower or lower uptake by the liver; however, the increase will be transient (a few hours) given the much faster clearance of lumasiran from the plasma compared to the liver. There is also a 44-fold safety margin for plasma lumasiran exposures between what is observed in patients with PH1 at the proposed clinical dose and exposures at the NOAEL in preclinical species. Finally, the review team noted that hepatic impairment is not a manifestation of PH1, and further studies in patients

with HI may not be feasible for a rare disease that manifests in childhood. As such, the review team recommends expanding the dosing recommendations to include patients with moderate HI.

As shown in Table 32, there is a marked decrease in liver uptake of GalNAc conjugated ligands in patients with severe HI, which leads to greater uncertainty regarding liver uptake and efficacy. Therefore, the review team does not believe dosing recommendations can be provided for patients with severe HI.

Renal Impairment

The impact of impaired renal function was determined by comparing the PK and efficacy of lumasiran in patients with PH1 and mild (eGFR 60 to <90 mL/min/1.73 m^2) or moderate (eGFR 30 to <60 mL/min/1.73 m^2) RI at baseline to patients with normal renal function (eGFR ≥90 mL/min/1.73 m^2). Across studies, the C_{max} of lumasiran was similar in patients with mild and moderate RI compared to patients with normal renal function. The reduction in urinary oxalate excretion was similar in patients with normal renal function and those with mild and moderate RI (Section 14.2.3.2). Further, in healthy subjects and patients with PH1 (eGFR >45 mL/min/1.73 m^2, Study 001B), renal excretion accounted for 7% to 26% of the administered dose, indicating a minor role for renal excretion in lumasiran clearance.

The review team agrees that a dose adjustment is not needed for patients with mild or moderate renal impairment (≥30 mL/min/1.73 m^2). Lumasiran has not been studied in patients with an eGFR <30 mL/min/1.73 m^2 or in patients with kidney failure.

Body Weight

The recommended dosing regimen includes actual body weight-based loading and maintenance doses as shown below. For an overview of the results supporting the proposed dosing regimen see Section 14.2.3.3. The review team agrees with the proposed dosing regimen.

Table 33. Lumasiran Actual Body Weight-Based Dosing Regimen for Pediatric and Adult Patients with PH1

Body Weight	Loading Dose	Maintenance Dose (Begin 1 Month After the Last Loading Dose)
Less than 10 kg	6 mg/kg once monthly for 3 doses	3 mg/kg once monthly
10 kg to less than 20 kg	6 mg/kg once monthly for 3 doses	6 mg/kg once every 3 months
20 kg and above	3 mg/kg once monthly for 3 doses	3 mg/kg once every 3 months

Source: Applicant's study protocol.

Other Intrinsic Factors

The covariate effect analysis in population PK/PD models indicated that age, race, sex, and BSA did not have a significant impact on the exposure-response (E-R) relationship for lumasiran (see Section 14.2.3.4). Hence, dose adjustment is not needed for these intrinsic factors. There is no clinical experience in patients over 60 years of age.

8.2. Drug Interactions

The drug-drug interaction potential of lumasiran involving cytochrome P450 (CYP) enzymes was investigated in vitro, and lumasiran is neither a substrate nor an inhibitor of CYP enzymes (see Section 14.1). No in vitro investigations on the CYP induction potential or substrate/inhibitor potential for transporters were conducted for lumasiran. Lumasiran is metabolized by exo- or endo- nucleases in the liver and, because the drug interaction potential for lumasiran is considered minimal, no further clinical DDI studies were conducted.

8.3. Plans for Pediatric Drug Development

FDA granted Alnylam orphan drug designation (designation request number 15-5053) for lumasiran for the treatment of PH1 on February 8, 2016; therefore, the product is exempt from Pediatric Research Equity Act requirements for the indication. ILLUMINATE-A enrolled patients down to 6 years of age, and ILLUMINATE-B enrolled patients <6 years of age. The initial approval of lumasiran for the treatment of PH1 will not be limited by age. Juvenile toxicology data can be found in Section 7.1.

8.4. Pregnancy and Lactation

Animal Data

A complete battery of developmental and reproductive toxicity studies was completed (Table 34). Safety margins from these studies are provided in Table 35.

Table 34. Nonclinical Data Supporting Labeling on Fertility, Pregnancy, and Lactation

Labeling Section	Nonclinical Data
8.1 Pregnancy	• In an embryo-fetal development study in pregnant rats, lumasiran was administered subcutaneously at doses of 3, 10, and 30 mg/kg/day during organogenesis (gestational days 6-17). Administration of lumasiran resulted in no effects on embryo-fetal survival or fetal body weights. No lumasiran-related fetal malformations were observed at 30 mg/kg/day, which is 45 times the MRHD, based on body surface area. • In an embryo-fetal development study in female rabbits, lumasiran was administered subcutaneously at doses of 3, 10, and 30 mg/kg/day during organogenesis (gestational days 7-19). There were decreases in maternal food consumption and decreases in maternal body weight gains at ≥3 mg/kg/day, with some recovery noted during the non-dosing period. There were no lumasiran-related fetal findings identified at doses up to 30 mg/kg/day (90 times the normalized MRHD based on body surface area). • In a pre- and postnatal development study, lumasiran administered subcutaneously to pregnant female rats on gestational days 7, 13, 19 and on lactation days 6, 12, and 18 through weaning at doses up to 50 mg/kg did not produce maternal toxicity or developmental effects in the offspring.
8.2 Lactation	No data
8.3 Females and males of reproductive potential	In a fertility and early embryonic development study, lumasiran administered subcutaneously once weekly at doses up to 50 mg/kg in male and female rats prior to and during mating, and once on gestation day 6 in females, resulted in no adverse effects on fertility or reproductive function in male or female animals.
13.1 Carcinogenesis, mutagenesis, impairment of fertility	• Long-term studies to assess carcinogenic risk of lumasiran have not been conducted. • Lumasiran was not genotoxic in an in vitro bacterial reverse mutation (Ames) assay, in the in vitro chromosomal aberration assay in cultured human peripheral blood lymphocytes, or the in vivo micronucleus assay in rats.

Table 35. Reproductive Toxicity Safety Margins

Study	NOAEL (mg/kg)	Nonclinical Exposure (µg•hr/mL)	Safety Margins by Exposure[a] (Multiples)	Safety Margins by BSA[b]
Fertility (male rat)	50	63.7	9X	11X
Fertility (female rat)	50	40.6	6X	11X
EFD rat	30	17.9	2.6	45X
EFD rabbit	30	59.7	8.7	90X
PPND rat	50	ND	>2.6	13X

Source: FDA's analysis.
Abbreviations: EFD = embryo-fetal development; NOAEL = effect level; PPND = peri- and postnatal; ND=not determined.
[a]Exposure multiples were based on population pharmacokinetics analysis where a 6 mg q3M clinical dose resulted in mean systemic exposures of $AUC_{0\text{-last}}$ = 6.8 µg•hr/mL.
[b]The safety margins were based on the maximum recommended human dose (MRHD) of 3 mg/kg/month normalized to 0.1 mg/kg/day, per body surface area.

Clinical Experience

There are no human data regarding the use of lumasiran in pregnant or lactating women. According to the Division of Pediatric and Maternal Health's consultation by Dr. Miriam Dinatale, based on the drug's large molecular size and short half-life, it is unlikely that lumasiran will accumulate in human milk.

9. Product Quality

The Office of Pharmaceutical Quality review team has assessed NDA 214103 with respect to Chemistry, Manufacturing, and Controls and has determined that it meets all applicable standards to support the identity, strength, quality, and purity that it purports. As such the Office of Pharmaceutical Quality recommends approval of this NDA from a quality perspective.

9.1. Device or Combination Product Considerations

Lumasiran is administered by subcutaneous injection using commercially available syringes and will not be marketed as a combination product.

10. Human Subjects Protections / Clinical Site and Other Good Clinical Practice Inspections / Financial Disclosure

FDA Inspections: The FDA did not request inspections of clinical investigator sites based on review of financial disclosures and consistency of the primary endpoint findings after excluding sites with significant disclosures. Investigators at four sites had financial disclosures exceeding $25,000. Collectively, these sites enrolled 14 patients (36% of ILLUMINATE-A); however, the treatment effect did not change after excluding individual sites from the primary endpoint analysis.

Site Audits and Data Quality Assurance: The Applicant's clinical monitor and the Contract Research Organization monitored the study and conducted investigator site audits to ensure the accuracy and reliability of clinical study data. According to the ILLUMINATE-A and -B Clinical Study Reports, electronic case report forms (eCRFs) were completed from source documents by trained persons and were source-document verified for accuracy and completeness by the clinical monitor.

Financial Disclosures: The Applicant has adequately disclosed financial arrangements with clinical investigators in ILLUMINATE-A and -B (see Section 23 for details). None of the investigators were full- or part-time employees of Alnylam Pharmaceuticals. Four investigators reported disclosable financial interests of >$25,000, including payments for consulting services, service on advisory boards, speaker fees, honoraria, research support, and payment for laboratory audits (see FDA Inspections above). None reported disclosable stock interest in Alnylam Pharmaceuticals.

Protocol Deviations: Major protocol deviations were defined as those that may significantly impact the completeness, accuracy, and/or reliability of the study data; or that may significantly affect a patient's rights, safety, and well-being. Deviations not classified as major were assigned as minor. Major protocol deviations were reviewed and approved by the Applicant prior to the interim database lock for the primary analysis of the placebo-controlled period of ILLUMINATE-A.

Five major protocol deviations were reported in four patients in ILLUMINATE-A:

- One patient ((b) (6)) randomized to placebo did not maintain a stable pyridoxine regimen. There were two pauses in pyridoxine dosing: (1) for five days, starting on Day 134, during the placebo-controlled period, and (2) a 12-day pause, starting on Day 187, during the extension period.
- Two patients ((b) (6) and (b) (6)) randomized to lumasiran had invalid 24-hour urine samples that were not repeated as specified in the protocol (Patient (b) (6); Month 3) or not collected (Patient (b) (6); Months 3 and 4).
- An ICF was not signed by one patient prior to performing study procedures (Patient (b) (6)). The patient signed the consent during a subsequent clinic visit but had already completed a 24-hour urine collection during screening.

One other protocol deviation was reported by the Applicant in five patients in ILLUMINATE-A:

- The first five patients were screened for eligibility and stratified by mean 24-hour urinary oxalate levels (≤1.70 or >1.70 mmol/24 hr/1.73 m^2) using a clinical assay performed at the (b) (4). Thereafter, a validated assay performed at (b) (4) was used. Urinary oxalate values from the first five patients were remeasured using the validated (b) (4) assay. All still met inclusion criteria, but three patients (two in the lumasiran group and one in placebo) who were stratified to lower urinary oxalate category would have been stratified to the higher urinary oxalate

category using the (b) (4) assay. The stratification discrepancies did not lead to an imbalance between the groups. Efficacy analyses used results from the (b) (4) assay.

There was one major protocol deviation in ILLUMINATE-B:

- Written informed consent was not obtained from both parents of Patient (b) (6) prior to performing study screening procedures. One parent provided written consent, but the other had provided verbal telephone consent.

All other protocol deviations were minor. It is unlikely that the protocol deviations affected patient safety or the interpretation of study results.

11. Advisory Committee Summary

The application does not raise significant issues regarding the safety or effectiveness of the drug; hence, no Advisory Committee Meeting was held.

III. Appendices

12. Summary of Regulatory History

U.S. Regulatory Actions and Marketing History

Lumasiran is not marketed in the United States, and there have been no previous regulatory actions.

Summary of Presubmission/ Submission Regulatory Activity

There were several interactions with the Agency over the course of development; a summary of key regulatory milestones, agreements, and advice is provided in Table 36.

Table 36. Key Regulatory Milestones, Agreements, and Advice

Date/Source	Advice From Agency/Regulatory Action
February 8, 2016 Orphan Drug Designation	Agency granted Alnylam orphan drug designation for lumasiran for the treatment of PH1 (ODD 15-5053).
February 9, 2016 Pre-IND meeting (written responses)	Sponsor requested the meeting to discuss their overall development plan, including a proposed phase 1/2 trial design.
February 23, 2018 Breakthrough Therapy Designation	Agency granted lumasiran Breakthrough Therapy designation for the treatment of PH1 based on marked and sustained reductions in urinary oxalate levels in patients with PH1 treated with lumasiran in a phase 2 study.
April 26, 2018 End of Phase 2 Meeting	Agency was willing to accept a substantial change in urinary oxalate in patients with high baseline levels in a placebo-controlled trial in patients ≥6 years of age as a basis for full approval of lumasiran for the treatment of PH1.
March 8, 2019 Advice/Information Request Letter	Agency agreed to use of spot urinary oxalate-to-creatinine ratio in lieu of 24-hour oxalate excretion for primary efficacy assessments in ILLUMINATE-B based on data provided by the Applicant to support agreement between oxalate excretion measured by spot urine samples and 24-hour urine samples, and the Applicant's plan to obtain supportive data from ILLUMINATE-A and -B).
October 10, 2019 PreNDA Written Responses (format and content)	Applicant requested discussion of the format and content of the NDA submission including plans to provide interim data from ILLUMINATE-B to support an indication in patients <6 years of age.
December 20, 2019 Rare Pediatric Disease designation	Agency granted the Applicant Rare Pediatric Disease designation (RPD-2019-246) for lumasiran for the treatment of PH1.
February 24, 2020 Pre-NDA meeting (topline results)	Agency agreed with the Applicant's plan to submit results from the 6-month placebo-controlled, double-blind phase of Study ALN-GO1-003 and available data from open-label Study ALN-GO1-004 in patients <6 years of age. The Applicant proposed to submit additional efficacy and safety data from Study ALN-GO1-004 during the review cycle by May 2020; the Agency noted it would determine whether these data are sufficient for labeling in this population during the review cycle.

Source: Table generated by clinical reviewer from review of Agency communications with the Applicant.

13. Pharmacology Toxicology: Additional Information and Assessment

13.1. Summary Review of Studies Submitted Under the IND

13.1.1. Primary Pharmacology

The drug substance (lumasiran) is a synthetic double stranded siRNA that has been modified by its conjugation with a carbohydrate ligand, GalNAc. Unmodified siRNAs are rapidly eliminated and do not achieve significant tissue distribution upon systemic administration. The conjugation of the drug substance to GalNAc is to (b) (4) In the hepatocytes, lumasiran interferes with the mRNA of the HAO1 gene, which encodes the hepatic peroxisomal enzyme, GO. The mechanism of action of lumasiran is to suppress GO, inhibit glyoxylate synthesis, and therefore, oxalate production, while causing the buildup of glycolate, which is soluble and readily excreted

in the urine. Lumasiran is pharmacologically active in both rodents, lagomorphs and nonhuman primates. Table 37 summarizes the in vitro (monkey hepatocytes) and in vivo proof-of-concept pharmacological studies that have been performed.

Table 37. In Vitro and In Vivo Pharmacological Studies of Lumasiran

Study Title/Study No.	Major Findings
In Vitro Study	
In Vitro Identification of HAO1-GalNAc Candidates in Support of Lead Selection for lumasiran/(b) (4) 15032	Lumasiran was selected as the lead siRNA with sequence complementarity to mouse, monkey and human HAO1, with an IC_{50} of 10pM in primary monkey hepatocytes following transfection.
In Vivo Studies	
Evaluation of Lumasiran in Wild-type Mice Following a Single Subcutaneous Injection/(b) (4) 15016	Lumasiran exhibited dose-dependent reduction of HAO1 mRNA in liver (91% maximum reduction at 10 mg/kg on Day 10) and dose-dependent increases in serum glycolate (4-fold maximum increase at 10 mg/kg on Day 10).
Evaluation of Lumasiran in a Mouse Model of Primary Hyperoxaluria Type 1 Following a Single Subcutaneous Injection/(b) (4) 15028	Lumasiran exhibited dose-dependent and durable reduction of urinary oxalate (50% maximum reduction for≥3 weeks at 3 mg/kg) and urinary glycolate increases (5-fold maximum increase for ≥4 weeks) in a mouse model of PH1 (AGXT -/- mice).
Evaluation of Lumasiran in Wild-Type Rats Following a Single Subcutaneous Injection /(b) (4) 15019	Lumasiran exhibited dose-dependent reduction of HAO1 mRNA in liver (94% maximum reduction at 10 mg/kg on Day 10) and dose-dependent increases in serum glycolate (8-fold maximum increase at 10 mg/kg on Day 10).
Evaluation of Lumasiran in a Rat Model of Primary Hyperoxaluria Type 1 Following Multiple Subcutaneous Injections / (b) (4) 15030	After 4-weekly doses, lumasiran exhibited dose-dependent reduction of HAO1 mRNA in liver (98% maximum reduction at 1 or 3 mg/kg) and dose-dependent urinary oxalate lowering (98% maximum oxalate lowering at 3 mg/kg) in a rat model of PH1 (generated by IV injection of alanine glyoxylate aminotransferase (AGXT) siRNA)
Evaluation of Lumasiran Following Subcutaneous Injection of Male Cynomolgus Monkeys /(b) (4) 15029 ((b) (4) 8321213)	Multidose administration of lumasiran results in potent and dose-responsive HAO1 mRNA silencing up to 99% after a single 4 mg/kg dose or four monthly doses of 2 mg/kg, or eight weekly doses of 1 mg/kg.

13.1.2. Secondary Pharmacology

Secondary pharmacology screening of lumasiran included bioinformatic analysis to identify a set of transcripts that may potentially be inhibited by A-131532, the antisense strand of lumasiran based on sequence homology. Potential off-target transcripts were then assessed for their response to lumasiran in human liver carcinoma cell line (Hep3B) cells. Data are summarized below in Table 38.

Table 38. Secondary Pharmacological Studies of Lumasiran

Study Title/Study No.	Major Findings
Bioinformatic Analysis	
Analysis of Human Hydroxyacid Oxidase (Glycolate Oxidase) 1 Polymorphism Variations in Support of ALN-GO1/(b) (4) 15020	Three single polymorphisms were identified from the NCBI dbSNP database, one of which is within the lumasiran targeting region at position 21 with respect to the 5' start of the antisense strand; however, this position is not considered important for mRNA recognition and cleavage. The other two SNPs had frequencies too low for reliable estimation.
In Vitro "Off-target" Analysis of AD-65585 the *HAO1* Targeting siRNA Component of ALN-GO1 / (b) (4) 15033	Lumasiran produced no off-target reductions at concentrations >1000 fold exceeding the IC_{50} for *HAO1* inhibition, confirming the specificity of lumasiran for *HAO1*.
Safety Pharmacology	
hERG assay	Not performed, given the physiochemical property of lumasiran and targeted liver delivery, this was determined to be acceptable. No special cause for concern for hERG channel interaction.
Cardiovascular safety pharmacology evaluation following subcutaneous injections to male telemetry-instrumented conscious nonhuman primate/Study No. 8322209. Species/strain: cynomolgus monkeys; Number/sex/group: 3/sex/group Dose: 0, 10 and 100 mg/kg Route of administration and dosing Frequency: s.c., days 1, 8, 15 and 22 (4 doses total)	All animals survived until study termination and there was no abnormal clinical observation attributed to lumasiran administration. The administration of ALN-GO1 at 10 and 100 mg/kg had no immediate or delayed effects on clinical observations, qualitative or quantitative ECG parameters, hemodynamic parameters, respiratory rate, or body temperature. The NOEL dose was 100 mg/kg, the highest dose tested.
ALN-GO1: 8-Week subcutaneous toxicity and toxicokinetic study with Neurological examination in Cynomolgus Monkeys/Study No. 8322208 Species/strain: Cynomolgus monkey Number/sex/group: 5/sex/group Dose: 0, 10, 30, 100 mg/kg/week 100 mg/kg/month (for 2 months) Route of administration: s.c.	No lumasiran-related neurological observations were noted.

Study Title/Study No.	Major Findings
General ADME/PK Studies	
The Pharmacokinetics, Bioavailability, Tissue distribution and Pharmacodynamics of GO1 siRNA (AD-65585) after a single Intravenous, or single or multiple subcutaneous doses to Sprague-Dawley Rats Study No. 8322988	• There were no sex-related differences observed in plasma or liver PK, or plasma PD for glycolate after lumasiran administration. After a single 5 mg/kg IV dose in rats, the mean C_{max} was 18,300 ng/ml in females and 31,800 ng/ml in males, observed within 5 minutes of dosing (first sampling time point), and then declining in a bi-exponential manner, with an apparent terminal half-life of 0.4 hours and total clearance (CL) of 646 ml/h/kg. A mean terminal volume of distribution (V_z) approximately 6-fold the blood volume in rats suggests the distribution of lumasiran outside the vasculature. • After a single SC dosing of the test article at 1, 5, and 10 mg/kg, the mean plasma C_{max} and AUC_t did not show any sex differences, and increased in a dose-proportional manner, with the exception of (0.1 mg/kg dose group) the lowest dose, where concentrations at most time points were below the limit of quantitation (BLQ). The terminal elimination half-life (t1/2β) was 1 hour and the mean total clearance after extravascular administration (CL/F) of a single dose ranged from 1850 to 2280 ml/h/kg and did not change in relation to dose. The CL/F values after repeat dosing ranged from 1440 to 2080 ml/h/kg, and were similar between the weekly and monthly dose groups. The mean absolute systemic bioavailability of lumasiran after SC administration (when compared to IV exposure) was 30%. • After a single SC dose, lumasiran rapidly distributed to the tissues, with the highest exposure found in the liver, with no sex-related differences in its distribution. • Liver and kidney exposures to repeat dosing of lumasiran were dose proportional although the test article did not accumulate in the liver; in contrast, there was a 2-fold accumulation in the kidney with weekly dosing. Kidney accumulation was not observed with monthly dosing. The liver exposure to lumasiran was consistently higher than plasma exposure. Quantitation of renal and fecal elimination of ALN-GO1 in pooled samples collected over 48 hours after a single 10 mg/kg dose showed that they are both minor routes of elimination for lumasiran. Renal clearance was 17 ml/h/kg in female rats and 29 ml/h/kg in male rats, but renal clearance for ALN-GO1 in rats only accounted for <1% of the total systemic clearance in rats, with the majority of clearance from circulation by liver uptake. • Plasma glycolate increased in proportion to dose, with a maximum response of 729% increase versus baseline, after a single 10 mg/kg dose. There were no apparent sex-related differences in plasma glycolate area under the effect curve (AUEC), correlated with measures of lumasiran plasma and liver PK.

The Pharmacokinetics, Bioavailability, Tissue distribution and Pharmacodynamics of GO1 siRNA (AD-65585) after a single Intravenous, or single or multiple subcutaneous doses to Cynomolgus Monkeys Study No: 8323135	• After a single 10 mg/kg IV dose of lumasiran in cynomolgus monkeys, plasma levels decreased in a biphasic manner, with a mean C_{max} of 134 µg/ml and an AUC_t of 58.9 h•µg/ml. Lumasiran was rapidly distributed, with a mean apparent volume of distribution at the terminal phase (V_z) of 163 ml/kg, indicating elimination from systemic circulation or distribution outside the vasculature. The mean total CL and elimination half-life of lumasiran were 189 ml/h/kg and 0.6 hours, respectively. • After a single SC dosing of the test article at 0.1, 1, 5 or 10 mg/kg in monkeys, the plasma C_{max} and AUC_t did not show any sex differences, and increased in a dose dependent manner, with the exception of the 1 mg/kg group, where females showed an extended elimination half-life (>11 hours) because of 2 animals with late, detectable plasma levels in the group. The mean total clearance (CL/F) ranged from 357 to 991 ml/h/kg across sexes in the dose groups, with a mean bioavailability of 25%. • After multiple SC dosing of lumasiran in monkeys at doses of 1 mg/kg (8-weekly doses) and 4 mg/kg (3-monthly doses), PK parameters were calculated after the first, fourth, and last doses for the weekly schedule and after the first, second and third doses for the monthly schedule. There were no differences observed between multiple or single SC dose administration, indicating that there is no time-dependent changes in the PK of lumasiran. The mean total clearance ranged from 757 to 1790 ml/h/kg, with a trend of lower values with increasing dose. However, the mean differences were within 2-fold between the 1 and 4 mg/kg dose groups. The plasma concentrations of lumasiran declined in a multiphasic manner after peaking at t_{max}, and $t_{1/2}\beta$ varied across the dose groups on Days 1 and 57, with mean values for males and females from 2.9 to 4.4 hours on Day 1, and from 2.9 to 3.4 hours on Day 57 after 8 weeks of dosing. Mean half-life was approximately 2 to 6 hours. Males had slightly higher AUC_t and $t_{1/2}\beta$ (<2-fold) after weekly dosing, when compared to females. However, exposure and half-life values were similar between males and females after monthly dosing. • The distribution of lumasiran in male and female monkey liver was estimated after a single IV or SC dose, or repeat SC doses. Slight differences exist between males and females in the distribution of lumasiran, with females having 1.2 to 2.7-fold higher exposures in the liver after a single dose. Mean $t_{1/2}$ after a single dose was 296, 615, 292, 371 and 409 hours for the 0.1, 1, 5, and 10 mg/kg groups. The mean liver AUC and C_{max} increased dose proportionally from 1070 to 76,700 h•µg/g and from 2 to 169 µg/g, respectively, across the single-dose SC range from 0.1 to 10 g/kg. After repeat dosing, liver exposures were equivalent in males and females. Liver exposures were higher than plasma exposures by 1335 to 19,816-fold. Liver exposure generally increased in direct

Study Title/Study No.	Major Findings
	proportion to plasma in males, but increased in a less than dose proportional manner to plasma exposure in females. • Quantitation of renal and fecal elimination of lumasiran in pooled samples collected over 168 hours after a single 10 mg/kg dose showed a renal clearance of 145 ml/h/kg in females and 166 ml/h/kg in males, approximating the glomerular filtration rate (GFR) of 178 ml/h/kg in monkeys. Renal clearance is only 20% of the total systemic clearance in monkeys, with the 80% remainder attributed to liver uptake. Lumasiran levels in feces accounted for only minimal amounts of the total test article administered in monkeys. • After a single SC administration of lumasiran (0.1, 5 and 10 mg/kg), a dose dependent increase in plasma glycolate was observed, with only the 5 and 10 mg/kg groups showing a sustained elevation (lasting 672 hours); whereas, with a single IV administration of 10 mg/kg, no response in plasma glycolate was observed, except at 336 hours postdose, where it was elevated at 15.3% compared to baseline. • After a total of 9-weekly SC administrations of 1 mg/kg lumasiran, the mean maximum elevation of plasma glycolate was 174% on Day 85 at 672 hours, and, after a total of 3-monthly SC administrations of 4 mg/kg lumasiran, the mean maximum elevation of plasma glycolate was 134% on Day 64 at 168 hours. There were no apparent sex-related differences in plasma glycolate elevation after SC administration, with the exception of the 5 mg/kg males which did not follow the dose-response curve.
Metabolism Studies	
Drug metabolism and pharmacokinetics nonclinical study report Study No: BA15012	The mean percentage (±SD) of plasma protein binding in mouse, rat, monkey, and human plasma at lumasiran concentrations of 5 to 100 µg/ml ranged from 5.21±1.58% to 40.33±2.67%, respectively.

Additional metabolism studies: Lumasiran is not a substrate (did not inh bit) of CYP1A2, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, CYP3A4 or CYP3A5. In contrast, positive control incubations showed complete parent depletion for enzymes rCYP2C9, rCYP2D6 and rCYP3A4/5. The percentage remaining for the other rCYPs tested ranged from 13.7 to 73.6% after 30 minutes. The percentage of ALN-GO1 remaining after 45 minutes was approximately 100% for all rCYPs tested in the presence of NADPH.
The in vivo and in vitro metabolic studies indicate that LUMASIRAN exhibited similar metabolic profiles across species in both lower and higher order species. No gender differences were observed in the rat, and monkey in vivo plasma and liver metabolic profile of LUMASIRAN.

13.1.3. Toxicology

13.1.3.1. General Toxicology (Pivotal)

Study Number/ Title: 8331914/ ALN-GO1: 25-Week Subcutaneous Toxicity and Toxicokinetic Study in Sprague-Dawley Rats

Key Study Findings

Subcutaneous (SC) administration of lumasiran once monthly to rats for 25 weeks, for a total of 7 doses, was well tolerated. Lumasiran-related findings (body weight effects and pathology

changes) were minor with no impact on the health and well-being of animals; these effects were not considered adverse. The NOAEL was considered to be 200 mg/kg, the highest dose evaluated, which corresponds to a sex-combined maximum plasma concentration (C_{max}) value of 29.9 µg/mL and AUC_{last} value of 378 h*µg/mL on Day 169.

Table 39. Study 8331914/ALN-GO1

Study Features and Methods	Details
GLP Compliance:	Yes
Dose and frequency of dosing:	0, 20, 50 and 200 mg/kg Once/month for 25 weeks
Route of administration:	SC
Formulation/vehicle:	Saline
Species/strain:	Sprague-Dawley Rats
Number/sex/group:	20/sex/group
Age:	6-7 weeks old
Satellite groups/unique design:	TK group (3 control; 9/dose for API)
Deviation from study protocol affecting interpretation of results:	None

Parameters	Major Findings
Mortality	One (incidental)
Clinical signs	None
Body weights	The mean body weight of animals administered 200 mg/kg was up to 10% lower, compared with the controls at the end of the dosing phase. This appears to be the dose-limiting toxicity in males.
Ophthalmoscopy	No effects
ECG	Not measured
Hematology	Minor lumasiran-related effects on hematology included the following: • Minimally lower red blood cell count (-4%), hemoglobin concentration (-5%), and hematocrit (-5%) in males administered 200 mg/kg • Minimally lower hemoglobin concentration (-5 to -6%), hematocrit (-4%), mean corpuscular volume (-3 to -4%), mean corpuscular hemoglobin (-5 to -6%), and mean corpuscular hemoglobin concentration (-2%) and minimally higher absolute reticulocyte count (+20 to +23%) in females administered ≥50 mg/kg • Minimally lower white blood cell count (-27%) due to minimally lower absolute lymphocyte count (-28%) in females administered 200 mg/kg Lumasiran-related effects on coagulation test results were dose-related and included the following: • Mildly to moderately lower fibrinogen concentration in males and females (-24 to -35% and -16 to -44%, respectively) administered ≥20 mg/kg • Minimally prolonged prothrombin time in females (+6 to +10%) administered ≥50 mg/kg and minimally shortened aPTT in males (-6 to -9%) administered ≥50 mg/kg and females (-10%) administered 200 mg/kg No clinical evidence of coagulopathy was noted, and no microscopic correlates to the coagulation test results were

Parameters	Major Findings
	identified. Thus, a specific mechanism for the lower fibrinogen concentration and altered prothrombin and aPTT was undetermined. However, the prolonged prothrombin time may have been related to the lower fibrinogen concentration.
Clinical chemistry	Minor ALN-GO1-related effects on clinical chemistry test results included the following: • Mildly higher cholesterol concentration in males and females (+41 to +47% and +51 to +62%, respectively) administered ≥50 mg/kg • Mildly higher globulin concentration (+13 to +17%) and mildly lower albumin:globulin ratio (-17%) in females administered ≥50 mg/kg • Minimally higher (+62%) ALP activity in males administered 200 mg/kg, which may have been associated with microscopic hepatocellular alterations With the exception of the higher ALP activity in males, no microscopic correlates to the other clinical chemistry findings were identified.
Urinalysis	No effects
Gross pathology	No effects
Organ weights	No effects
Histopathology Adequate battery: Yes Peer reviewed: Yes	Lumasiran-related microscopic observations at the terminal sacrifice were noted in the liver and kidney. Microscopic findings in animals administered ≥20 mg/kg consisted of minimal to marked hepatocellular vacuolation, minimal pigment containing Kupffer cells, minimal increased hepatocyte mitosis, and/or minimal to moderate hepatocyte karyomegaly in the liver, and minimal to slight basophilic granules and/or minimal tubule cell vacuolation in the kidney. Minimal to marked hepatocellular vacuolation in the liver of animals administered ≥20 mg/kg was characterized by microvesicular (numerous small vacuoles without nuclear displacement) and/or macrovesicular (one large vacuole that displaces the nucleus) types. Additional lumasiran-related findings in the liver consisted of minimally increased pigment containing Kupffer cells, and minimal to moderate hepatocyte karyomegaly in animals administered ≥20 mg/kg. Karyomegaly likely represented increased cell turnover and was characterized by increased variability in the size of hepatocyte nuclei and overall cell size, increased binucleated hepatocytes, and some individualization of hepatocytes. Minimal increased mitotic figures in hepatocytes was noted predominately in females administered 200 mg/kg, one female administered 50 mg/kg, and one male administered 20 mg/kg. Lumasiran-related microscopic findings in the kidney consisted of minimal to slight basophilic granules in the kidney tubule cell cytoplasm of animals administered ≥20 mg/kg, and minimal vacuolation of renal tubule cells in males administered ≥50 mg/kg and characterized by sporadic tubules with slightly basophilic cytoplasmic vacuoles without evidence of degeneration. All microscopic findings are considered non-adverse.

Toxicokinetics	Plasma No consistent sex differences in Lumasiran exposure were noted in C_{max} and AUC values following administration of lumasiran once every 4 weeks via SC injection for seven doses. On Day 1, exposure in males was 0.949- to 1.514-times the exposure in females and on Day 169, exposure in males was 0.989- to 2.255-times the exposure in females. Both sexes had similar exposure to Lumasiran; thus, the toxicokinetic parameters of both sexes are combined and discussed in this report. Lumasiran was absorbed in a similar manner across the dose range, with T_{max} values ranging from 1.0 to 2.0 hours postdose on Day 1, and from 0.5 to 1.0 hours postdose on Day 169. Lumasiran C_{max} values on Day 1, following SC dosing of 20, 50 or 200 mg/kg lumasiran, on Day 1 were 3521, 12,180, and 37,665 ng/mL, respectively, and 1511, 6830, and 29,948 ng/mL on Day 169, respectively, and AUC_{last} values were 10,099, 44,563, and 265,876 ng·h/mL on Day 1, respectively, and 12,630, 51,273, and 377,599 ng·h/mL on Day 169, respectively. Lumasiran plasma concentrations declined in a multiphasic manner after peaking, and $t_{1/2}$, where determined, ranged from 1.4 to 2.6 hours on Day 1, and from 4.2 to 9.3 hours on Day 169. Lumasiran exposure (C_{max} and AUC) generally increased in a greater than dose-proportional manner with increases in dose over the dose range studied on Days 1 and 169. For the 2.5- and 4.0-fold increases in lumasiran dose from 20 to 50 mg/kg and from 50 to 200 mg/kg on Day 1, corresponding 3.460- and 3.092-fold increases in C_{max}, respectively, and 4.413- and 5.966-fold increases in AUC_{last}, respectively, were noted. The AUC from time zero extrapolated to infinity (AUC_{inf}) was not determined on Day 1. For the same 2.5-and 4.0-fold increases in dose on Day 169, corresponding 4.521- and 4.385-fold increases in C_{max}, respectively, 4.060- and 7.364-fold increases in AUC_{last}, respectively, and 3.648- and 7.330-fold increases in AUC_{inf} were noted. Little to no accumulation of lumasiran was noted in the plasma with repeated SC dosing of lumasiran every 4 weeks, with Day 169 C_{max} values which were 0.429- to 0.795-times the Day 1 C_{max} values, AUC_{last} values which were 1.151- to 1.420-times the Day 1 values, and AUC_{inf} values which were 1.433- to 1.456-times the Day 1 values. Tissue Following administration of lumasiran once every 4 weeks via SC injection for seven doses, lumasiran was detectable in liver and kidney tissue samples collected at 4, 12, and 24 hours after the last dose (Day 169). No apparent sex differences were observed in liver and kidney concentrations of lumasiran across the dose range evaluated in the study, except for a higher (~2.7-fold) mean kidney C_{max} observed in males administered 20 mg/kg. ALN-GO1 kidney C_{max} values increased in a dose-proportional manner for the 20 to 200 mg/kg dose groups. Lumasiran liver C_{max} values increased in a dose-proportional manner for the 20 and 50 mg/kg dose groups, and increased in less than dose-proportional manner

Parameters	Major Findings
	for the 50 to 200 mg/kg dose groups. Mean lumasiran liver C_{max} was higher than kidney C_{max} across all of the tested dose groups.

Study Number/ Title: 8331915/ ALN-GO1: 36-Week Subcutaneous Toxicity and Toxicokinetic Study in Cynomolgus Monkeys

Key Study Findings

Male and female monkeys were administered vehicle control article/diluent or a dose level of 30, 100, or 300 mg/kg lumasiran via SC injection once every 4 weeks for a total of 10 doses. One male administered 100 mg/kg was found dead with a distended (large) gastrointestinal tract suggestive of bloat with a perforation in the jejunum; bloat (distended gastrointestinal tract) was considered to be the cause of death, which was unrelated to lumasiran. The only lumasiran-related clinical pathology change was minimally increased alkaline phosphatase in males administered ≥30 mg/kg. Lumasiran-related microscopic findings were noted in the liver for animals administered 100 or 300 mg/kg (minimal basophilic granules in the Kupffer cells) and the mandibular and mesenteric lymph nodes of animals administered ≥30 mg/kg (minimal to slight accumulations of vacuolated macrophages in the sinusoids). There were comparable increases from baseline in plasma glycolate concentrations in all dose levels, observed on Day 29 (549, 532, and 590% increase for the 30, 100, and 300 mg/kg lumasiran dose groups, respectively) and Day 253 (592, 581, and 626% increase for the 30, 100, and 300 mg/kg lumasiran dose groups, respectively). These effects were consistent with the known pharmacological effects of lumasiran. Because of the mild severity of findings and the lack of impact on the health and wellbeing of animals, the clinical pathology and microscopic findings were considered not adverse. Thus, the NOAEL is 300 mg/kg, the highest dose evaluated, which corresponds to a sex-combined C_{max} value of 76400 ng/mL and AUC_{last} value of 1290000 ng·h/mL on Day 253.

Study Features and Methods	Details
GLP compliance:	Yes
Dose and frequency of dosing:	30, 100, or 300 mg/kg lumasiran once every 4 weeks for a total of 10 doses
Route of administration:	SC
Formulation/vehicle:	saline
Species/strain:	cynomolgus monkeys
Number/sex/group:	4/sex/group
Age:	29-47 months old
Satellite groups/unique design:	none
Deviation from study protocol affecting interpretation of results:	none

Study Features and Methods	Details
Parameters	**Major Findings**
Mortality	One male (Animal I12489) administered 100 mg/kg was found dead on Day 235. No lumasiran-related clinical observations were noted for this animal during the dosing phase. The animal was found dead with a distended (large) gastrointestinal (GI) tract suggestive of bloat with a perforation in the jejunum. Bloat (distended GI tract) was considered to be the cause of death, which was not considered lumasiran-related. All remaining animals survived to their scheduled sacrifice.
Clinical signs	No lumasiran-related clinical observations or alterations in food consumption were noted. Low qualitative food consumption values, noted sporadically for animals administered lumasiran, were incidental and within the normal variation. Other remarkable observations noted during the dosing phase included prolapsed rectum, excessive tartar in teeth, emesis, thin appearance, hypoactive behavior, fecal painting, discharge, vomitus, abnormal feces (non-formed, liquid, and/or discolored), and skin and pelage findings (alopecia, broken skin, discolored skin discolored hair coat, dry skin, raised area, sores, scabs, scaly skin, and/or thinning hair coat). These appeared rather infrequently, were transient, or were with comparable incidences as controls; therefore, they were considered not test article related.
Body weights	No lumasiran-related alterations in body weight or body weight gain were noted for animals administered up to 300 mg/kg.
Ophthalmoscopy	No effects
ECG	Not measured
Hematology	No effects
Clinical chemistry	Potentially lumasiran-related minimal increases in alkaline phosphatase activity (up to +94% relative to baseline) were noted on Day 254 in males administered ≥30 mg/kg. These were small in magnitude, not noted in females, had no correlative changes in other enzyme activities, and had no microscopic correlates.
Urinalysis	No effects
Gross pathology	No effects
Organ weights	No effects
Histopathology Adequate battery: Yes Peer reviewed: Yes	No adverse lumasiran related microscopic findings were present. The non-adverse lumasiran-related microscopic findings at the terminal sacrifice consisted of minimal basophilic granules in the Kupffer cells in the liver of animals administered 100 or 300 mg/kg and minimal to slight accumulations of vacuolated macrophages in the sinusoids of lymph nodes (mandibular, mesenteric) of animals administered ≥30 mg/kg. The macrophages were characterized by accumulations of foamy cytoplasmic vacuoles, with faintly basophilic staining, often distending the cytoplasm.
Toxicokinetics	Plasma No apparent sex differences were observed in mean glycolate concentrations on Days 1, 29 or 253. The mean

Study Features and Methods	Details
	baseline (Day 1) plasma glycolate concentrations were similar across the animals administered vehicle control or 30, 100 and 300 mg/kg lumasiran and ranged from 61.0 to 65.5 ng/mL. On Day 29, mean plasma glycolate concentrations were >2.7-fold higher for animals administered 30, 100 or 300 mg/kg lumasiran, compared to vehicle control. Mean glycolate concentrations did not increase further with an increase in dose from 30 to 300 mg/kg. On Day 253, mean plasma glycolate concentrations were >3.5-fold higher for animals administered 30, 100 and 300 mg/kg lumasiran, compared to vehicle control. Mean glycolate concentrations were similar with an increase in dose from 30 to 300 mg/kg. Across all dose levels, comparable increase from baseline in plasma glycolate concentrations were observed on Day 29 (549, 532 and 590% increase for the 30, 100 and 300 mg/kg lumasiran dose groups, respectively) and Day 253 (592, 581 and 626% increase for the 30, 100 and 300 mg/kg lumasiran dose groups, respectively). Tissue Lumasiran was detected in liver and kidney samples collected from all animals administered 30, 100 and 300 mg/kg lumasiran. No sex related differences in lumasiran concentrations in either liver or kidney were observed. Overall mean liver lumasiran concentrations were 926± 382, 2390± 629 and 3890± 1170 µg/g for the 30, 100 and 300 mg/kg lumasiran dose groups, respectively. Overall mean kidney lumasiran concentrations were 10.4± 3.54, 55.9± 7.99 and 181± 77.2 µg/g for the 30, 100 and 300 mg/kg lumasiran dose groups, respectively. Mean liver and kidney lumasiran concentrations increased in an approximate dose-proportional manner across the lumasiran dose range evaluated in the present study. A 3.3-fold increase in lumasiran dose (from 30 to 100 mg/kg), resulted in a 2.5-fold increase in liver lumasiran concentrations, a further 3-fold increase in lumasiran dose (from 100 to 300 mg/kg), resulted in a 1.6-fold increase in liver lumasiran concentrations. A 3.3-fold increase in lumasiran dose (from 30 to 100 mg/kg), resulted in a 5.3-fold increase in kidney lumasiran concentrations, a further 3-fold increase in lumasiran dose (from 100 to 300 mg/kg), resulted in a 3.2-fold increase in kidney lumasiran concentrations. Overall mean (combined male and female) liver lumasiran concentrations were 21.5 to 89.0-fold higher than kidney lumasiran concentrations across the lumasiran dose range evaluated in the present study. Liver to kidney concentration ratios in both sexes decreased with an increase in dose from 30 to 300 mg/kg.

Genetic Toxicology

Table 40. Genetic Toxicology

Study No./ Study Title	Key Study Findings
Study No.:8322204 In vitro reverse mutation assay in bacterial cells GLP compliance: Y Study is valid: Y	Lumasiran at a maximum concentration of 5000 µg/plate did not produce any mutagenic effect in any of the 5 bacterial strains tested, with and without metabolic activation.
Study No.: 8322203 Assay for chromosomal aberrations in vitro in Chinese hamster ovary cells GLP compliance: Y Study is valid: Y	Lumasiran at a maximum concentration of 500 µg/ml, in the presence and absence of metabolic activation did not induce chromosomal or chromatid damage in human lymphocytes.
Study No.: 8322202 Assay for Micronucleus Induction in rat bone marrow from a 2-Week oral toxicity study GLP compliance: Y Study is valid: Y	Lumasiran at SC doses of 500, 1,000 or 2,000 mg/kg did not induce micronuclei in polychromatic erythrocytes (PCEs) in the bone marrow of SD male and female rats.

13.1.3.2. Reproductive Toxicology

Study Number/ Title: 20149791/ A Subcutaneous Injection Embryo-Fetal Development Study in Sprague Dawley Rats

GLP Compliance: Yes

Key Study Findings

Lumasiran administered SC once daily to pregnant rats from DG 6 to 17 at 3, 10, and 30 mg/kg did not result in mortality, clinical signs, or effects on maternal body weights or maternal body weight gains. There were non-adverse reductions in food consumption at 30 mg/kg. As a result, the maternal NOAEL for lumasiran was considered to be 30 mg/kg (corresponding to a C_{max} of 3.49 µg/mL and AUC_{last} of 17.9 h*µg/mL on DG 17).

Administration of lumasiran did not result in any effects on embryo-fetal survival or fetal body weights. There were no lumasiran-related fetal malformations; the only lumasiran-related fetal skeletal abnormalities were bipartite ossification of the sternebrae and misshapen cervical arches at 30 mg/kg. These changes were not considered adverse; therefore, the developmental NOAEL was considered to be 30 mg/kg.

Table 41. Methods of Oral Embryo-Fetal Developmental Study in Rats

Parameter	Method Details
Dose and frequency of dosing:	0, 3, 10 and 30 mg/kg/day during DG 6 to 17
Route of administration:	SC
Formulation/vehicle:	saline

Parameter	Method Details
Species/strain:	Sprague-Dawley rat
Number/sex/group:	22 females/group
Satellite groups:	TK: 3 for control; 6 for API
Study design:	Female Sprague Dawley rats were administered lumasiran or control (0.9% NaCl) by SC injection once daily on DG 6 through 17 (N=22 per group) at 0, 3, 10, and 30 mg/kg. The dose volume was 5 mL/kg. Toxicokinetics was assessed in satellite groups (N=3 in the control group and N=6 per lumasiran dose group). The rats assigned to the main study and TK phase were euthanized on DG 21 and DG 18, respectively.
Deviation from study protocol affecting interpretation of results:	none

Table 42. Observations and Results

Parameters	Major Findings
Mortality	none
Clinical signs	No effects seen
Body weights	Transient changes in mean maternal body weights were observed on DG 21 were -0.9%, +0.7%, and -2.5% from controls in the 3, 10, and 30m g/kg dose groups, respectively. Mean maternal body weight gains for the overall interval of DG 6 to 21 were +0.6%, +3.0%, and -2.6% from controls in the 3, 10, and 30 mg/kg dose groups, respectively.
Necropsy findings Cesarean section data	All rats were pregnant and had ovarian and uterine examinations performed on DG 21. There were no lumasiran-related effects on any ovarian or uterine parameter at any dose level.
Necropsy findings Offspring	Low dose: None Mid dose: None High dose: The pancreas was gelatinous in two dams in the 30 mg/kg dose group.

Study Number/ Title: 20158688 / A Subcutaneous Injection Developmental and Perinatal/Postnatal Reproduction Study in Rats, Including a Postnatal Behavioral/Functional Evaluation

GLP Compliance: Yes

Key Study Findings

Lumasiran did not cause any adverse lumasiran-related effects from maternal exposure to the F0 generation or potential in utero and/or exposure through milk. Therefore, the maternal and reproductive NOAEL for lumasiran is considered to be 50 mg/kg, the highest dose tested. The NOAEL for viability and growth in the offspring for lumasiran is also considered to be 50 mg/kg.

Table 43. Methods of Subcutaneous Peri/Post-natal Development Study in Rats

Parameter	Method Details
Dose and frequency of dosing:	0, 5, 15 and 50 mg/kg/day (DG 7, 13 and 19) (Lactation days 6, 12 and 18) Six total doses per animal.
Route of administration:	SC
Formulation/vehicle:	saline
Species/strain:	Sprague-Dawley Rat
Number/sex/group:	22/group
Satellite groups:	4 animals/group for TK

Parameter	Method Details
Study design:	Timed-mated female Sprague Dawley rats (N=22/group) were administered lumasiran or the control article (0.9% NaCl) by SC injection approximately every 6 days (on DG 7, 13, and 19, and on lactation days [LD] 6, 12, and 18) for a total of 6 doses at 0, 5, 15, and 50 mg/kg. The dose volume was 5 mL/kg. Concentrations of lumasiran in plasma was assessed in satellite groups (N=4/group). The rats assigned to the main study (F0) and bioanalysis groups were euthanized on LD 21 and LD 12, respectively. F0 generation females were monitored for viability, clinical signs, body weights, food consumption, parturition, lactation, and maternal behavior. F0 generation females were allowed to deliver naturally and were monitored to weaning (LD 21), at which time F1 generation offspring (1/sex/litter) were selected for continuation on study. Before weaning, all F1 generation rats were assessed for viability, clinical signs, and body weight. During the postweaning period, all F1 generation rats selected for continuation on study were monitored for viability, clinical signs, body weights, food consumption, sexual maturation, behavior (as evaluated by passive avoidance and Morris Watermaze testing), macroscopic observations, and reproductive organ weights. Adult F1 generation males and females were cohabited for the assessment of reproductive function. The ovaries and uteri of F1 generation females were examined for the number and distribution of corpora lutea, implantations, and live and dead embryos. Additionally, blood samples were collected on LD 12 from the F0 generation dams assigned to the bioanalysis study. On postnatal day (PND) 12, blood samples were collected via cardiac puncture (after euthanasia) from F1 generation pups (3 pups/sex/litter) from female rats assigned to the bioanalytical groups at approximately 2 hours post maternal dose. Blood samples were analyzed for lumasiran concentrations.
Deviation from study protocol affecting interpretation of results:	None
Mortality	None
Clinical signs	None
Body weights	There were no lumasiran-related effects on mean maternal body weights or mean maternal body weight gains in the F0 generation during the gestation period. Mean maternal body weight gains were significantly reduced ($p \leq 0.05$) in the 50 mg/kg dose group for the interval of GD 10 to 12 compared to controls (-26% from controls). This reduction was not considered adverse because the effect was transient, and the magnitude of the effect was minor (4.3 g over 2 days). There were no lumasiran-related effects on mean maternal body weights or mean maternal body weight gains in the F0 generation during the lactation period. All changes in mean maternal body weights or mean maternal body weight gains during the lactation period, including those of statistical significance, were considered unrelate to lumasiran because: 1) the change was transient in nature; 2) the change was not dose dependent; and/or 3) there was no effect on the overall interval of LD 1 to 21. There were no lumasiran-related effects on mean maternal food consumption in the F0 generation during the gestation or lactation periods.
Necropsy findings Cesarean section data	There were no lumasiran-related observations noted at macroscopic necropsy examinations in the F0 generation rats. There were no lumasiran-related effects on terminal body weights, absolute ovary and uterine weights, or the ratio of ovaries and uterus weights to terminal body weight in the F0 generation rats.

Parameter	Method Details
	All rats were pregnant and delivered their litters. There were no lumasiran-related effects on any natural delivery parameter. The values for the number of dams delivering litters, the duration of gestation, implantation sites per delivered litter, the gestation index, the number of dams with stillborn pups, and the number of dams with all pups dying were similar among the four dose groups. There was a statistically significant decrease ($p \leq 0.01$) in the 50 mg/kg dose group in the percentage of liveborn pups (95.1% versus 99.2% in controls). This decrease was primarily due to a single dam (2069) that had one liveborn pup, 2 stillborn pups, and 4 pups with undetermined viability due to cannibalization. When this dam is excluded from summarization, the percentage of liveborn pups in the 50 mg/kg dose group is 97.5%, which is comparable to controls and within the historical control range of the Testing Facility. Therefore, the decrease in the percentage of liveborn pups at 50 mg/kg was not considered lumasiran-related. F1 Generation There were no lumasiran-related effects on any litter parameter. The values for the lactation index (percentage of pups born that survived to weaning), surviving pups per litter, the sex ratio, live litter size at weighing, and pup weights per litter were similar among the four dose groups. There were reductions or statistically significant reductions in pup weights in the 5 and 15 mg/kg dose groups at each recorded interval from PND 1 to 21 compared to controls (-7% to -12% from controls). There were no effects on pup weights in the 50 mg/kg dose group; therefore, the reduced pup weights in the 5 and 15 mg/kg dose groups were considered unrelated to lumasiran because they were not dose dependent. There was a statistically significant increase ($p \leq 0.05$) in the 50 mg/kg dose group in the viability index compared to controls (100% versus 97.0% in controls). This increase is not considered toxicologically relevant because an increase in viability index is a positive outcome. There were no lumasiran-related clinical signs in the F1 generation. There were no lumasiran-related macroscopic observations in the F1 generation pups. There were no lumasiran-related effects on mean body weights or mean body weight gains in the F1 males and females during the postweaning or gestation periods. There were no lumasiran-related effects on mean food consumption in the F1 males or females during the postweaning and gestation periods. There were no lumasiran-related effects on sexual maturation in the F1 generation males or females. The mean day on which sexual maturation was achieved ranged from 44.4 to 45.2 days for male balano-preputial separation and 33.0 to 33.3 days for female vaginal patency. The mean body weight recorded on the day of sexual maturation for F1 males and females was similar among the four dose groups. There were no lumasiran-related effects on learning and memory in the F1 generation males or females when evaluated beginning on Day 24 postpartum (±1-day) using the passive avoidance test.

Parameter	Method Details
	There were no lumasiran-related effects on learning and memory in the males or females when evaluated beginning on PND 70 through PND 90 using the Morris water maze test at any dose. There were no lumasiran-related effects on the days needed for mating (2.2 to 3.0 days), mating index (86.4% to 100%), or fertility index (95.0% to 100%) in the F1 generation rats. All values were similar among the four dose groups. There were no lumasiran-related effects noted at macroscopic necropsy examination in the F1 generation males or females. There were no lumasiran-related effects on terminal body weights, organ weights, or the ratio of organ weights to terminal body weights. There were no lumasiran-related effects on any ovarian or uterine parameter at any dose. Pregnancy was confirmed in 21 (95.4%), 22 (100%), 21 (95.4%), and 20 (90.9%) F1 generation females in the 0, 5, 15, and 50 mg/kg dose groups, respectively. **F2 Generation** The concentration of lumasiran was below the lower level of quantification in the plasma of all pups on PND 12. **Toxicokinetics and Lactation (exposures)** The maternal plasma concentration of lumasiran at 2 hours postdose on LD 12 increased at an approximate dose proportional rate.

13.1.3.3. Impurities/Degradants

A dedicated 13-week repeat-dose toxicology study (Study# GO1-GLP19-002, test article batch 18D004) was performed in Sprague-Dawley rats in order to qualify the impurities that are found in batches of lumasiran. Doses of 0, 20, 50 and 200 mg/kg of lumasiran were injected subcutaneously once per month for a total of 4 doses.

The qualification threshold for individual (b) (4)-specific impurities, is set at (b) (4). The Applicant's basis for using NLT (b) (4) as qualification threshold is that (b) (4) Based on molecular mass, the molar amount of a (b) (4) impurity present at (b) (4) level is similar to a small molecule impurity present at 0.10%, the qualification threshold for a small molecule drug substance with a maximum daily dose of <2 g/day (per ICH-Q3A). Based on this assumption, individual impurity peaks are evaluated for their relation to identification and safety qualification thresholds (impurity peaks > (b) (4)% are identified and > (b) (4)% are qualified).

Besides the designated 13-week impurity qualification study, the data from the 25-week repeat-dose toxicity study (#GO1-GLP15-038, test article batch 15D008) was also utilized to qualify these impurities. A NOAEL dose for impurity qualification was identified from both studies as (b) (4) mg/kg/month.

Qualified levels for each lumasiran DS impurity group were calculated from the percentage of the impurity group present in the lumasiran drug substance used in the repeat-dose toxicity studies multiplied by the dosing ratio of NOAEL dose ((b) (4) mg/kg/month) relative to the

maximum recommended human dosage of 6 mg/kg/month (see Applicant's table below). The Applicant's justification for the use of body weight, rather than body surface area (BSA), for dose scaling in this calculation is that the drug is mostly indicated for pediatric population. A comparison based on BSA is not considered appropriate for pediatric population, especially for pediatric patients <20 kg. Based on the data from these two studies, the impurity specification levels proposed for the drug substance were considered qualified from the pharmacology and toxicology perspective, relative to the maximum recommended human dose.

Table 44. Qualified Levels of Specified Impurity RRT Groups of Lumasiran Drug Substance for MRHD of 6.0 mg/kg/month

Impurity Group	RRT	Impurity Level in Tox Batch (Peak Area%)	Toxicity Reference Dose[a] (mg/kg/month)	Qualified Level of MRHD (%)	Toxicity Study Number/Batch
Denaturing AX-HPLC UV					
[redacted]				(b) (4)	GO1-GLP15-038/ 15D008
					GO1-GLP15-038/ 15D008
					GO1-GLP19-002/ 18D004
					GO1-GLP15-038/ 15D008
					GO1-GLP15-038/ 15D008
					GO1-GLP15-038/ 15D008
Denaturing IPRP-HPLC UV					
[redacted]				(b) (4)	GO1-GLP15-038/ 15D008
					GO1-GLP15-038/ 15D008
					GO1-GLP15-038/ 15D008
					GO1-GLP15-038/ 15D008
					GO1-GLP15-038/ 15D008
					GO1-GLP19-002/ 18D004

[a] Nonclinical Dose used for qualification was the NOAEL of (b) (4) mg/kg/month determined in studies GO1-GLP15-038 and GO1-GLP19-002

Source: Applicant's table

13.2. Individual Reviews of Studies Submitted to the NDA

13.2.1. ALN-GO1: A Subcutaneous Injection Fertility and Early Embryonic Development Study in Sprague Dawley Rats

Table 45. Study 20149792/ALN-GO1

Study no.:	20149792
Study report location:	EDR

Conducting laboratory and location:	(b) (4)
GLP compliance:	Yes
Drug, lot #, and % purity:	Lumasiran, lot # 5004024, 99.8%
Methods	Details
Doses:	0, 5, 15, 50 mg/kg/week
Frequency of dosing:	Males: once/ week starting 29 days before cohabitation (8 total doses) Females: once/week starting 22 days before cohabitation and once on presumed Day 6 of gestation
Number/sex/group:	20 animals/sex/dose; 3/dose for TK in controls;6/dose for TK in treated
Dose volume:	5 ml/kg
Formulation/vehicle:	0.9% saline
Route of administration:	SUBCUTANEOUS
Species:	Rat
Strain:	Sprague-Dawley
Comment on study and conduct:	valid
Dosing solution analysis:	All study samples analyzed had mean concentrations within or equal to the acceptance criteria of ± 10% (individual values within or equal to ± 15%) of their theoretical concentrations. For homogeneity, the RSD of concentrations for all samples in each group tested was within th acceptance criteria of ≤ 5%.

Methods	**Details**
Doses:	0, 5, 15, 50 mg/kg/week
Frequency of dosing:	Males: once/ week starting 29 days before cohabitation (8 total doses) Females: once/week starting 22 days before cohabitation and once on presumed Day 6 of gestation
Number/sex/group:	20 animals/sex/dose; 3/dose for TK in controls;6/dose for TK in treated
Dose volume:	5 ml/kg
Formulation/vehicle:	0.9% saline
Route of administration:	SUBCUTANEOUS
Species:	Rat
Strain:	Sprague-Dawley
Comment on study and conduct:	valid
Dosing solution analysis:	All study samples analyzed had mean concentrations within or equal to the acceptance criteria of ± 10% (individual values within or equal to ± 15%) of their theoretical concentrations. For homogeneity, the RSD of concentrations for all samples in each group tested was within th acceptance criteria of ≤ 5%.

Key Study Findings

Administration of lumasiran to male rats did not result in any effects on clinical observations, absolute body weights, mating, fertility, reproductive organ weights, or sperm parameters. Based on these data, the NOAEL for general toxicity parameters and reproduction in treated male rats

was concluded to be 50 mg/kg (corresponding to a C_{max} of 9710 ng/mL and AUC_{last} of 63700 ng*h/mL on Day 29 of Study (DS 29). No drug-related effects were observed on reproductive parameters in untreated females mated with treated males.

Administration of lumasiran to female rats did not result in any effects on clinical observations, body weights, body weight gains, or food consumption. The NOAEL for females for maternal toxicity was considered to be 50 mg/kg (corresponding to a C_{max} of 7200 ng/mL and AUC_{last} of 40600 ng*h/mL on Day 6 of presumed gestation (DG 6). There were no effects on estrous cycling, mating, fertility, or ovarian and uterine parameters in the females. As a result, the reproductive and development NOAEL for females was considered to be 50 mg/kg.

Observations and Results

Males

Mortality

All male rats survived to scheduled euthanasia.

Clinical Observations

There were no ALN-GO1-related clinical signs observed in the males at any dose level.

Body Weights and Body Weight Gains

There were no ALN-GO1-related effects on mean body weights at any dose level, and there were no ALN-GO1-related effects on mean body weight gains in the 5 and 15 mg/kg dose groups.

Mean body weight gains were significantly reduced ($p \leq 0.01$) in the 50 mg/kg dose group for the intervals of Day of Study (DS) 8 to 15, DS 29 to 36, and DS 36 to 43 compared to controls (-24%, -85%, and -32% from controls, respectively). As a result, mean body weight gains were reduced in the 50 mg/kg dose group for the overall interval of DS 1 to 49 (-10% from controls).

There were no significant effects on mean body weights.

Food Consumption

Mean food consumption was reduced or statistically significantly reduced ($p \leq 0.01$ or $p \leq 0.05$) at all dose levels for the intervals of DS 8 to 15 (-54% to -64% from controls) and DS 15 to 22 (-13% to -19% from controls) compared to controls. As a result, mean food consumption was significantly reduced ($p \leq 0.01$) at all dose levels for the overall interval of DS 1 to 29 compared to controls (-19%, -20%, and -24% in the 5, 15, and 50 mg/kg dose groups, respectively).

Mating and Fertility

There were no ALN-GO1-related effects on days needed for mating (2.4 to 2.9 days), mating index (100%), or fertility index (85% to 100%) in the males mated to untreated females.

Macroscopic Pathology

There were no ALN-GO1-related abnormalities detected at necropsy examination at any dose level.

Terminal Body Weights, Organ Weights, and Ratios (%) of Organ Weight to Terminal Body Weight

There were no ALN-GO1-related effects on terminal body weights, organ weights, or the ratios of organ weights to terminal body weights at any dose level.

Sperm Evaluation

There were no ALN-GO1-related changes in sperm parameters (i.e., vas deferens sperm motility or caudal epididymal sperm density).

Untreated Female Rats

All untreated female rats survived to scheduled euthanasia.

Pregnancy was confirmed in 19 (95.0%), 20 (100%), 20 (100%), and 17 (85%) untreated females that were mated with the 20 treated males in each of the 0, 5, 15, and 50 mg/kg dose groups, respectively.

There were no ALN-GO1-related effects on any ovarian or uterine parameter in the untreated females with confirmed pregnancies that were mated with treated males. The litter means for implantations, percentage of preimplantation loss, viable and nonviable embryos, and percentage of postimplantation loss were comparable among the four dose groups. No dam had a litter consisting of only nonviable embryos. No placentae examined had any detectable abnormalities.

The total number of corpora lutea was statistically significantly increased ($p \leq 0.05$) in the females mated with the males in the 50 mg/kg dose group.

Treated Female Rats

Mortality

There was no ALN-GO1-related mortality at any dose level.

Clinical Observations

There were no ALN-GO1-related clinical signs observed in the females at any dose level.

Body Weights

There were no ALN-GO1-related effects on mean body weights or mean body weight gains in the females during the premating period or during the gestation period at any dose level.

Food Consumption

There were no ALN-GO1-related effects on mean food consumption in the females during the premating or gestation periods at any dose level.

Mating and Fertility

The number of estrous stages per 15 days was similar among the ALN-GO1 dose groups and controls before the start of administration and during the precohabitation dose period.

There were no ALN-GO1-related effects on days cohabitated prior to mating (2.5 to 3.3 days), mating index (95% to 100%), or fertility index (85% to 100%).

Macroscopic Pathology

There were no abnormalities detected at necropsy examination at any dose level.

Ovarian and Uterine Examinations

Pregnancy was confirmed in 19 (95%), 20 (100%), 17 (85%), and 18 (90%) treated females in the 0, 5, 15, and 50 mg/kg dose groups, respectively. As a result of the dam that delivered its litter in the 50 mg/kg dose group, ovarian and uterine examinations on DG 13 were performed on 19, 20, 17, and 17 females in the 0, 5, 15, and 50 mg/kg dose groups, respectively.

The litter averages for corpora lutea, implantations, the percentage of preimplantation loss, viable and nonviable embryos, and the percentage of postimplantation loss were similar among the four dose groups. No dam had a litter consisting of only nonviable embryos. No placentae examined had any detectable abnormalities.

There was a statistically significant reduction ($p \leq 0.01$) in the number of corpora lutea on the left ovary in the 50 mg/kg dose group (7.1 corpora lutea versus 8.8 corpora lutea in controls). But this was not considered adverse since there were more corpora lutea on the right overy in the same dose group and there was no effect on the total number of corpora lutea.

Toxicokinetic Evaluations

Maximum concentrations (C_{max}) of ALN-GO1 in male rats reached 327, 1410, and 9710 ng/mL 1 or 2 hours postdose on Study Day 29 following weekly subcutaneous dosing of 5, 15 or 50 mg/kg ALN-GO1, with associated AUC_{last} of 1700, 7870, and 63700 ng·h/mL, respectively.

ALN-GO1 C_{max} in pregnant female rats reached 382, 1070, and 7200 ng/mL 2 hours postdose on gestation day (GD) 6 following weekly subcutaneous dosing of 5, 15 or 50 mg/kg ALN-GO1, with associated AUC_{last} of 1750, 5090 and 40600 ng·h/mL, respectively. Increases in ALN-GO1 dose from 5 to 15 mg/kg and from 15 to 50 mg/kg resulted in greater than dose-proportional increases in C_{max} and AUC_{last} in male rats. The 3-fold increase in ALN-GO1 dose from 5 to 15 mg/kg resulted in 4.31- and 4.63-fold increases in C_{max} and AUC_{last}, and the 3.3-fold increase in ALN-GO1 dose from 15 to 50 mg/kg resulted in respective 6.89- and 8.09-fold increases in C_{max} and AUC_{last}.

In pregnant female rats, increases in ALN-GO1 dose from 5 to 15 mg/kg resulted in approximately dose-proportional increases in exposure and the increase in dose from 15 to 50 mg/kg resulted in greater than dose-proportional increases in C_{max} and AUC_{last}. The 3-fold increase in ALN-GO1 dose from 5 to 15 mg/kg resulted in 2.80- and 2.91-fold increases in C_{max} and AUC_{last}, and the 3.3-fold increase in ALN-GO1 dose from 15 to 50 mg/kg resulted in respective 6.73- and 7.98-fold increases in C_{max} and AUC_{last}.

Table 46. Toxicokinetic Parameters in Male Rates on Study Day 29 and Pregnant Female Rats on Gestations Day 6 Following Weekly SC Dosing of ALN-GO1 at 5, 15, or 50 mg/kg

	Males Study Day 29			Pregnant Females Gestation Day 6		
Dose (mg/kg)	**5**	**15**	**50**	**5**	**15**	**50**
C_{max} (ng/mL)	327	1410	9710	382	1070	7200
t_{max} (h)	2	1	2	2	2	2
AUC_{last} (ng·h/mL)	1700	7870	63700	1750	5090	40600
C_{max}/Dose [(ng/mL)/(mg/kg)]	65.5	94.2	194	76.3	71.3	144
AUC_{last}/Dose [(ng·h/mL)/(mg/kg)]	340	525	1270	350	339	812

CONCLUSION

Administration of ALN-GO1 to Sprague Dawley male rats via SC injection once weekly beginning 29 days before cohabitation (total of 8 doses) at 5, 15, and 50 mg/kg did not result in any effects on clinical observations, body weights, mating, fertility, reproductive organ weights, or sperm parameters in the treated males or on ovarian and uterine parameters in the untreated females. There were reduced body weight gains at 50 mg/kg and reduced food consumption at all dose levels that were not considered adverse because they were not accompanied by reduced absolute body weights. Based on these results, the no-observed-adverse-effect level (NOAEL) for general toxicity parameters and reproduction was concluded to be 50 mg/kg (corresponding to a C_{max} of 9710 ng/mL and AUC_{last} of 63700 ng*h/mL on DS 29).

Administration of ALN-GO1 to female Sprague Dawley rats via SC injection once weekly beginning 22 days before cohabitation and once on DG 6 did not result in any effects on clinical observations, body weights, body weight gains, or food consumption. On the basis of these data, the NOAEL for females for maternal toxicity was concluded to be 50 mg/kg (corresponding to a C_{max} of 7200 ng/mL and AUC_{last} of 40600 ng*h/mL). There were no effects on estrous cycling, mating, fertility or ovarian and uterine parameters in the females. As a result, the reproductive and development NOAEL was concluded to be 50 mg/kg.

13.2.2. Embryo-Fetal Developmental Toxicity and Toxicokinetic Study With ALN-GO1 in Rabbits

Table 47. Embryo-Fetal Developmental Toxicity and Toxicokinetic Study of ALN-GO1 in Rabbits

Study no.:	8340641
Study report location:	EDR
Conducting laboratory and location:	(b) (4)
GLP compliance:	Yes
Drug, lot #, and % purity:	Lumasiran, lot # 5004024, 99.5%

Methods	Details
Doses:	0, 3, 10, 30 mg/kg/day
Frequency of dosing:	q.d. during G.D.7-G.D. 19
Number/sex/group:	25 females/dose; 3/dose for TK
Dose volume:	5 ml/kg
Formulation/vehicle:	0.9% saline
Route of administration:	SUBCUTANEOUS
Species:	Rabbit
Strain:	Hra:(NZW) SPF rabbits
Comment on study design and conduct:	None
Dosing solution analysis:	All concentration verification results met acceptance criteria. The mean % of target results was between 90 and 110%, and the relative standard deviation (%RSD) was not more than 5% and met specifications for concentration verification. No significant peaks were detected in control samples.

Key Study Findings

Administration of lumasiran to pregnant rabbits during organogenesis (GD 7-19) resulted in maternal toxicity as shown by reduced body weight gain at all dose level tested (a body weight gain decrease of 9%, 14%, and 17% at 3, 10, or 30 mg/kg/day, respectively, compared with concurrent controls). During the recovery phase (GD 20 to 29), a partial recovery of body weight gain occurred, but still resulted in a significant decrease in body weight gain at ≥10 mg/kg/day after the recovery phase.

There were no lumasiran-related fetal malformations identified at doses up to 30 mg/kg/day, which was determined to be the developmental NOAEL (corresponding to a GD 19 C_{max} of 10100 ng/mL and a GD 19 $AUC_{(0-24)}$ of 59700 ng·h/mL.)

Observations and Results

F_0 Dams

Mortality

All animals survived to scheduled euthanasia.

Clinical Signs

The only ALN-GO1-related clinical observation was few feces. This finding was primarily correlated with reduced food consumption for animals across all groups, including controls.

Body Weight

Throughout the dosing phase (GD 7 to 20); all treatment groups exhibited reduced body weight gain compared with concurrent controls, with some recovery noted during the non-dosing period (GD 20 through 29). During the dosing phase (GD 7 to 20), control animals gained 252.7 grams (7%), compared with animals administered 3, 10, or 30 mg/kg, which gained 228.8 (7%), 217.1 (6%), or 210.8 grams (6%), respectively, across this same time interval. This represented a body weight gain decrease of 9%, 14%, and 17% for those administered 3, 10, or 30 mg/kg, respectively, compared with concurrent controls. However, during the recovery phase (GD 20 to

29), control animals gained 157.7 grams (4%), compared with animals administered 3, 10, or 30 mg/kg, which gained 176.4 (5%), 143.5 (4%), or 155.4 grams (4%), respectively, across this same time interval. This represented a body weight increase of 12%, decrease of 9%, and decrease of 1%, respectively during recovery, for those administered 3, 10, or 30 mg/kg, compared with concurrent controls, reflecting a partial recovery. At the end of the recovery period, there was still a reduction in body weight gain of 12% and 11% at 10 and 30 mg/kg respectively, compared with the concurrent control group.

Feed Consumption

Animals in all ALN-GO1 treated groups ate somewhat less than concurrent controls, with a transient decrease noted for all treatment groups from approximately GD 12 to 17 and some recovery from GD 18 to 23. Compared with controls, which ate 1834.2 grams from GD 7 to 20, animals administered 3, 10, or 30 mg/kg ALN-GO1 ate 5%, 8%, and 7% less than controls, respectively. Some recovery occurred during the postdose period for ALN-GO1-treated animals, with decreased food consumption values increasing compared to the dosing period. Compared with controls, which ate 1067.7 grams from GD 20 to 29, animals administered 3, 10, or 30 mg/kg ate 2%, 5%, and 1% less than control animals, respectively.

Cesarean Section Data

No notable difference in any C-section parameter, including adjusted fetal body weights (which varied less than 1% from control values), occurred.

Necropsy/ Histopathology

At necropsy, three macroscopic observations were noted. One control animal (B0024) had a mass on its left kidney and an enlarged placenta for Fetus R1; one animal administered 3 mg/kg (B0105) was missing its gall bladder. These findings were observed in a control animal or individually, with no dose dependence; therefore, they were considered not ALN-GO1 related.

Toxicokinetics

Following daily SC dosing of ALN-GO1 in pregnant rabbits from GD 7-19, ALN-GO1 distributed in a similar manner across the dose groups with a time to reach maximum concentration values ranging from 1.0 to 2.0 h postdose on GDs 7 and 19. After daily doses of 3, 10, and 30 mg/kg/day, ALN-GO1 was detected in plasma out to 2, 8, and 8 h postdose on GD 7, respectively, and out to 8, 24, and 24 h postdose on GD 19, respectively.

Mean ALN-GO1 C_{max} values following daily SC dosing of ALN-GO1 at 3, 10, and 30 mg/kg/day were 684, 3580, and 16500 ng/mL on GD 7, respectively, and 457, 2400, and 10100 ng/mL on GD 19, respectively. Mean C_{max} values on GD 19 were 0.640- to 0.672-fold compared to C_{max} on GD 7. Mean ALN-GO1 AUC_{last} values following daily SC doses of ALN-GO1 at 3, 10, and 30 mg/kg/day were 978, 15200, and 75700 ng·h/mL on GD 7, and 2000, 12200, and 59700 ng·h/mL on GD 19 in the 3, 10, and 30 mg/kg/day ALN-GO1 dose groups, respectively. Mean AUC_{last} values on GD 19 were 0.802- to 2.05-fold compared to GD 7. Half-life was not reported due to insufficient data in the terminal phase.

Increases in ALN-GO1 dose from 3 to 10 mg/kg/day and from 10 to 30 mg/kg/day resulted in greater than dose-proportional increases in C_{max} and AUC_{last} on GDs 7 and 19.

The 3.33-fold increase in ALN-GO1 dose from 3 to 10 mg/kg/day resulted in 5.23- and 5.25-fold increases in C_{max} on GD 7 and GD 19, and 15.5- and 6.10-fold increases in AUC_{last}. The 3.00-fold

increase in ALN-GO1 dose from 10 to 30 mg/kg/day on GD 7 and GD 19 resulted in respective 4.61- and 4.21-fold increases in C_{max}, and 4.98- and 4.89-fold increases in AUC_{last}.

Analysis of plasma samples collected from control animals in Group 1 showed no quantifiable ALN-GO1 on GD 7 or GD 19.

Tissue TK: ALN-GO1 was measurable in maternal liver and kidney samples at all ALN-GO1 dose levels on GD 20. Mean maternal liver concentrations of ALN-GO1 increased in an approximately dose-proportional manner from 3 to 10 mg/kg and in a less than dose-proportional manner from 10 to 30 mg/kg after 13 once-daily SC administrations.

Mean maternal kidney concentrations increased in a less than dose-proportional manner from 3 to 10 mg/kg, but increased in a greater than dose-proportional manner over the dose range of 10 to 30 mg/kg.

Maternal liver concentrations were higher (by 14.1-, 21.7-, and 4.9-fold at 3, 10, and 30 mg/kg, respectively) than maternal kidney concentrations across all dose groups.

ALN-GO1 was not measurable in any of the placental tissues from the 3 mg/kg dose group. Low levels of ALN-GO1 were measurable in placental tissues with mean ± SD concentrations of 0.286±0.0682 and 0.47±0.159 µg/g, respectively for the 10 and 30 mg/kg dose groups. ALN-GO1 mean maternal liver concentrations were 4196- and 5192- fold higher than those in placenta at 10 and 30 mg/kg, respectively.

ALN-GO1 was not measurable in any of the fetal liver or fetal tissues.

F1 Offspring

Terminal Observations

Fetal weights were no affected by test agent at any dose level.

Fetal Malformations/ Variations (External, Visceral, Skeletal)

There were no ALN-GO1-related external malformations or variations detected at any dose level.

Pilot Neonate and Juvenile Toxicity and Toxicokinetic Study with ALN-GO1 in Sprague-Dawley Rats When Administered by Subcutaneous Injection.

Table 48. Study 8335749

Study no.:	8335749
Study report location:	EDR
Conducting laboratory and location:	(b) (4)
Duration:	29
Duration Units:	Day (PND 4 through 32)
GLP compliance:	Yes
Drug, lot #, and % purity:	Lumasiran; 982169; 98.5%

Scientific Justification for Study

Lumasiran's Indication and Usage includes pediatric population (from infancy to 17 years) since primary hyperoxaluria -1 is a genetic disease present at birth (inborn error).

At the end-of-phase 2 meeting, the Sponsor requested a waiver for the definitive neonate and juvenile toxicity study based on the lack of significant adverse findings in the GLP pilot study. The Division agreed provided that the pilot neonate-juvenile study included a toxicokinetic evaluation and full toxicological histopathological evaluations of a standard battery of tissues.

Toxicity Specific to Juvenile Animal Studies

Sporadic injection site reactions across all dose levels, mild body weight gain decreases for females given ≥30 mg/kg, minimal microscopic kidney and injection site findings across all dose levels, minimally to mildly decreased fibrinogen concentrations across all dose levels, and mildly increased glucose concentration in females administered ≥30 mg/kg.

Table 49. Juvenile Animal Studies

Methods	Details
Doses:	0, 10, 30, 100 mg/kg Six litters/group; Each group with 5M/5F TK: 3 litters/group; 5M/5F
Route of administration:	SUBCUTANEOUS
Species:	Rat
Strain:	Sprague-Dawley: Crl:CD
Age at start of experiment:	PND 4
Period of development studied:	PND 4-32
Comment on study design and conduct:	Drug administered PND 4 and every week until PND 32 (5 doses total)
Parameters and key endpoints evaluated:	Injection site; liver function; body weight gain
Dosing solution analysis:	The mean % of target results were within 90%-110% and the % RSD was not more than 5%, and met specifications for concentration verification. The control samples from the last preparation did not pass specifications as the absorbance was greater than 0.004 AU. An investigation was conducted into the cause of the failing results. A review of laboratory procedures was performed which yielded no apparent evidence of an error. The failing results were attributable to an artificially low-set AU detection limit (0.004) outside of the manufacturer's recommended lower limit of 0.03 AU. Because all vehicle control samples met the acceptance criteria once the appropriate AU lower limit is considered, the failing result is not expected to impact study integrity or the interpretation of results.

Observations and Results

Mortality

All animals survived to their scheduled sacrifice.

Clinical Signs

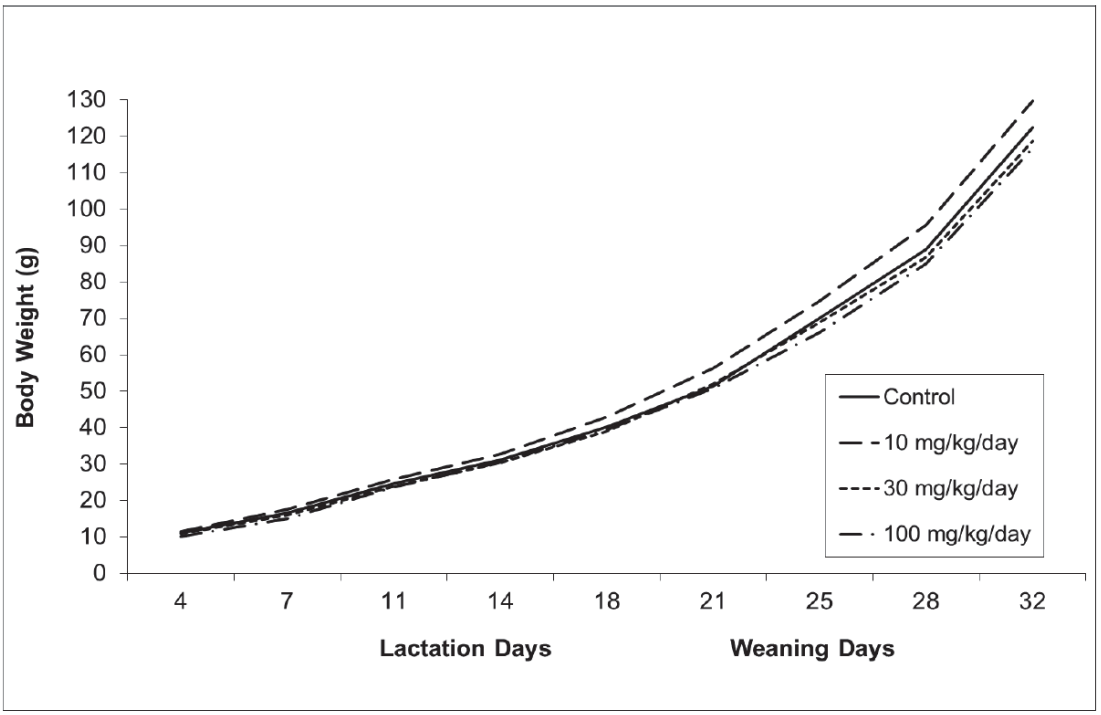
Few clinical observations were noted. Of these, the vast majority observed were dose site reactions including discolored skin (black) and sore or scab, on the dorsal cervical, thoracic, or lumbar area(s). These observations were noted in only a few juveniles/treatment, were also found in controls, and primarily resolved during the duration of the study.

Body Weight

Male juveniles gained weight comparably with concurrent controls, with only minimal body weight gain decreases noted. From PNDs 4 to 32, body weight gains for males administered 10 mg/kg were more than controls (+6%), and male body weight gains for those administered 30 or 100 mg/kg were less than 4 (-3%) or 5 grams below (-4%), respectively, concurrent controls (see male pup body weight chart below).

For male juveniles administered 10 mg/kg ALN-GO1, a transient statistically significant increase in the absolute body weight was noted on PND 21, when these juveniles exhibited a 10% increase from concurrent controls. For male juveniles administered 30 mg/kg ALN-GO1, a transient statistically significant decrease in the absolute body weight was noted on PND 7, when these juveniles exhibited a 4% decrease from concurrent controls. Male juveniles administered 100 mg/kg ALN-GO1 weighed less than controls (-1%) on PND 4, with concomitant statistical significance. Although the decrease in body weight gain seems ALN-GO1 treatment related, yet these decreases were minimal in magnitude and were not considered adverse.

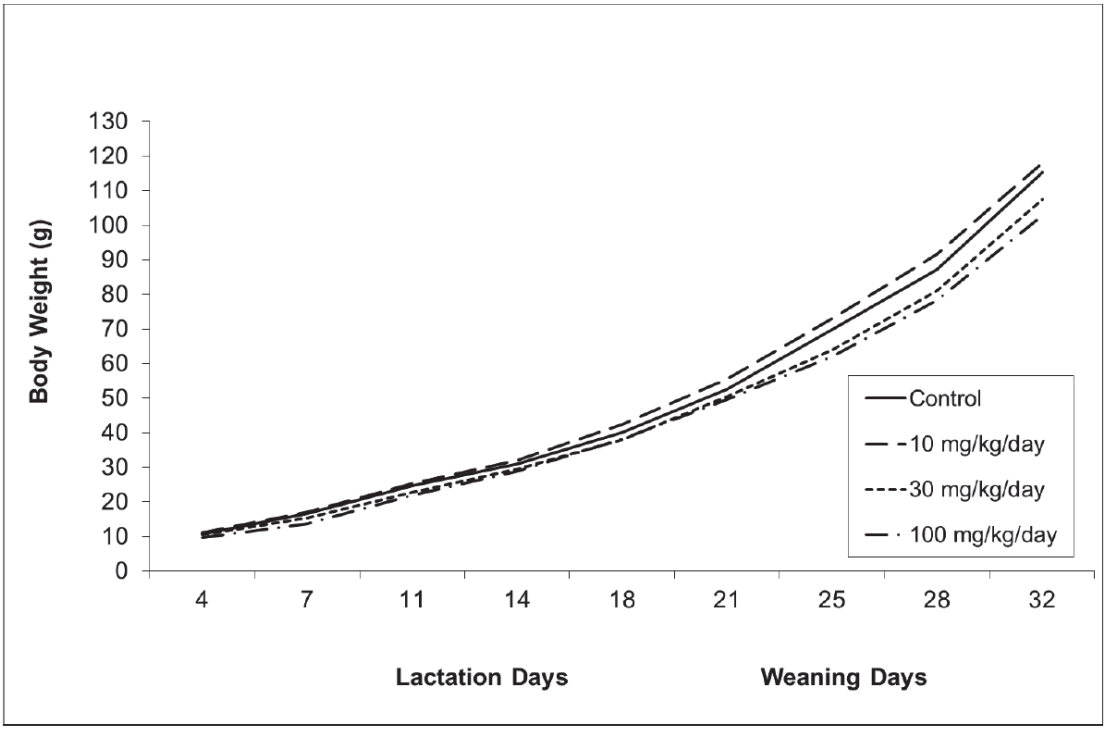
Figure 12. Mean Male Pup Body Weight



Female juveniles gained weight comparable with concurrent controls, with mild to moderate body weight gain decreases noted. From PND 4-32, body weight gains for female juveniles administered 10 mg/kg were greater than controls (+2%), and female juvenile body weight gains for those administered 30 or 100 mg/kg were less than 8 (-7%) or 11 grams below (-10%), respectively, those in concurrent controls.

For female juveniles administered 30 mg/kg, a statistically significant decrease in the absolute body weight was noted on PNDs 7.11, 25, 28, and 32 (-7%, -7%, -9%, -7%, and -7%, respectively) compared to concurrent controls. Female juveniles administered 100 mg/kg weighed less than controls (-9%) on PND 4, with concomitant statistical significance. Female juveniles administered 100 mg/kg weighed less than concurrent controls at each weighing interval, with statistical significance noted on PND 7, 11, 25, 28, and 32, when these juveniles exhibited -17%, -11%, -11%, -7%, and -7%, respectively, compared to controls (see female pup body weight chart below). Although there were mild to moderate dose-dependent effect on body weight gain in female pups, but over the course of the study, female pup body weights continued to trend upwards towards controls levels.

Figure 13. Mean Female Pup Body Weight



Feed Consumption

Not reported.

Ophthalmoscopy

Not reported.

Hematology

ALN-GO1 administration had no effect on hematology test results.

Clinical Chemistry

ALN-GO1 administration had a minor effect on clinical chemistry test results; this consisted of mildly increased glucose concentration (+35 to +52% from control mean) for females administered ≥30 mg/kg. This change was considered non-adverse because it was of small magnitude, limited to one sex, and had no clinical or microscopic correlates.

Urinalysis

Lumasiran did not affect any values.

Sexual Maturation

Not reported.

Reproductive Capacity

Not reported.

CNS/ Neurobehavioral Assessment

Not assessed.

Bone Evaluation

Not evaluated.

Gross Pathology

All macroscopic findings were considered spontaneous and/or incidental because they occurred at a low incidence and were randomly distributed across groups; therefore, they were considered not ALN-GO1 related.

Organ Weights

No ALN-GO1-related changes in organ weight parameters were noted.

Histopathology

Table 50. Incidence and Severity of ALN-GO1-Related Microscopic Findings

	ALN-GO1							
Sex	Males				Females			
Dose Level (mg/kg)	0	10	30	100	0	10	30	100
Kidney								
Number Examined	9	9	9	9	9	9	9	9
Basophilic granules, tubule cell								
Minimal	0	0	0	7	0	0	0	1
Subcutaneous Injection Site								
Number Examined	9	9	9	9	9	9	9	9
Vacuolation, macrophages								
Minimal	0	1	0	2	0	0	1	0

Histopathology evaluation included a standard battery of tissues. Following completion of the primary microscopic evaluation, a peer review evaluation was performed by a second pathologist.

Minimal basophilic granules were present in the proximal tubule epithelial cells of the kidney in animals administered 100 mg/kg. Minimal vacuolation of the macrophages was observed at the SC injection site of one male (Animal B31845) administered 10 mg/kg, two males (Animals B31904 and B31905) administered 100 mg/kg, and one female (Animal B31974) administered 30 mg/kg ALN-GO1. An increased incidence and severity of mixed inflammatory cell infiltrates at the injection site was observed in some females administered 10 or 30 mg/kg ALN-GO1, however this was observed in the absence of a dose response. None of these findings were considered adverse.

Toxicokinetics

No apparent sex differences were observed on PND 4 for either liver or kidney ALN-GO1 TK parameters across all ALN-GO1 dose groups. Maximal ALN-GO1 liver concentrations were observed at 8 h postdose for the 10 and 30 mg/kg ALN-GO1 dose groups and gradually declined up to 24 h postdose. Maximum ALN-GO1 liver concentrations were observed at 24 h postdose for the 100 mg/kg ALN-GO1 dose group (last time point collected in the study). Maximal ALNGO1 kidney concentrations were observed at 2 h postdose for the 10 mg/kg ALN-GO1 dose

group and at 4 h postdose for the 30 and 100 mg/kg ALN-GO1 dose groups. In general, ALNGO1 kidney concentrations remained stable up to 24 h postdose (the last time point collected in the study).

The 24 h postdose ALN-GO1 liver concentrations were generally similar at both PND 4 and PND 32 across the 10 to 100 mg/kg ALN-GO1 dose groups indicating absence of accumulation upon repeat weekly dosing.

The 24 h postdose ALN-GO1 kidney concentrations were generally similar at both PND 4 and PND 32 for the 30 to 100 mg/kg ALN-GO1 dose groups, indicating no accumulation upon repeat weekly administration of ALN-GO1 at these doses. However, a slight (~2-fold) accumulation of ALN-GO1 was observed in the kidneys upon weekly dosing of ALN-GO1 at 10 mg/kg.

On PND 4 and PND 32, ALN-GO1 liver exposures increased in a less than dose-proportional manner across the 10 to 100 mg/kg ALN-GO1 dose range, indicating possible saturation of uptake by the ASGR on hepatocytes. On PND 4 and PND 32, ALN-GO1 kidney exposures generally increased in a higher than dose-proportional manner across the 10 to 100 mg/kg ALN-GO1 dose range.

ALN-GO1 liver exposures were higher, compared to ALN-GO1 kidney exposures on both PND 4 and PND 32, across the 10 to 100 mg/kg ALN-GO1 dose groups. PND 4 ALN-GO1 liver to kidney C_{max} ratios were 12.2, 6.82 and 2.11 for the 10, 30 and 100 mg/kg ALN-GO1 dose groups respectively. PND 4 ALN-GO1 liver to kidney AUC_{last} ratios were 17.9, 6.09 and 2.5 for the 10, 30 and 100 mg/kg dose groups, respectively.

PND 32 ALN-GO1 liver to kidney 24 h postdose concentration ratios were 9.49, 5.33 and 2.29, respectively. The magnitude of ALN-GO1 liver to kidney exposures decreased with an increase in ALN-GO1 dose on both PND 4 and PND 32.

ALN-GO1 liver to kidney exposure ratios decreased with an increase in ALN-GO1 dose on both PND 4 and PND 32.

14. Clinical Pharmacology: Additional Information and Assessment

14.1. In Vitro Studies

Lumasiran is primarily metabolized by exo- or endo- nucleases and the in vitro metabolic stability in pooled serum and liver S9 fractions for 24 h, indicated minimal degradation (>80-90 % remaining) of the sense and antisense strands, across the species (mouse, rat, monkey and human).

In vitro assays also revealed that lumasiran was neither a substrate nor an inhibitor for any of the cytochrome P450 (CYP) enzymes. Incubation for 45 min with recombinant CYP enzymes showed complete stability (~100 % remaining) of lumasiran and hence it was not a substrate for CYP1A2, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, CYP3A4 or CYP3A5. At clinically relevant concentrations, lumasiran did not cause a direct or time-dependent inhibition towards any of the above CYP enzymes, as was shown by an absence of impact on the in vitro metabolism of specific marker substrates for each of the enzyme.

In vitro CYP induction and transporter substrate/inhibition studies have not been conducted with lumasiran, owing to a low potential for such interactions, based on an aggregate analysis of data from similar siRNA GalNAc conjugates (PMID 31270142).

Taken together, the drug interaction liability of lumasiran is low.

14.2. In Vivo Studies

14.2.1. Population PK Analysis

The Applicant preformed the population pharmacokinetic (PK) analysis for lumasiran using 763 plasma PK data from a total of 99 subjects, 24 healthy adult subjects and 75 patients with PH1 from Trials 001, 002, 003 and 004. Descriptive statistics of baseline demographics for continuous demographic data are presented in Table 51 and descriptive statistics of categorical demographic data are presented in Table 52.

Table 51. Baseline Characteristics of PK Population by Study (Continuous Covariates)

Baseline Characteristics	Study 001 Part A (N=24)	Study 001 Part B / Study 002 (N=20)	Study 003 (N=39)	Study 004 (N=16)	Overall (N=99)
Age at consent (years)					
Mean (SD)	28.8 (5.93)	14.9 (10.2)	18.1 (12.7)	3.13 (1.61)	17.6 (12.6)
Median [min, max]	27.5 [21.0, 42.0]	11.5 [6.00, 43.0]	14.0 [6.00, 60.0]	3.50 [0.167, 5.00]	15.0 [0.167, 60.0]
Age at first dose in Study 004 (years)[a]					
Mean (SD)				3.58 (1.771)	
Median [min, max]				3.88 [0.3, 6.1]	
Weight (kg)					
Mean (SD)	72.4 (12.3)	49.9 (27.2)	58.2 (27.6)	14.5 (4.87)	52.9 (28.7)
Median [min, max]	73.6 [45.0, 97.5]	42.8 [21.3, 110]	59.6 [17.3, 107]	13.6 [6.20, 24.3]	55.0 [6.20, 110]
BSA (m^2)					
Mean (SD)	1.86 (0.201)	1.41 (0.474)	1.56 (0.495)	0.619 (0.154)	1.45 (0.557)
Median [min, max]	1.87 [1.36, 2.26]	1.34 [0.852, 2.40]	1.70 [0.753, 2.33]	0.586 [0.317, 0.878]	1.60 [0.317, 2.40]
eGFR (mL/min/1.73 m^2)					
Mean (SD)	90.6 (12.7)	78.5 (21.0)	81.6 (25.7)	113 (28.1)	88.3 (25.3)
Median [min, max]	89.8 [64.6, 117]	74.0 [50.9, 131]	83.4 [31.7, 131]	114 [64.7, 174]	87.0 [31.7, 174]

Abbreviations: BMI=body mass index; BSA=body surface area; eGFR=estimated glomerular filtration rate; max=maximum; min=minimum; PK=pharmacokinetic; SD=standard deviation.

Source: Applicant's Population PK Analysis Report Table 6.

Table 52. Baseline Characteristics of PK/PD Population (Categorical Covariates)

Baseline Characteristics	Count (%)				
	Study 001 Part A (N=24)	Study 001 Part B / Study 002 (N=20)	Study 003 (N=39)	Study 004 (N=16)	Overall (N=99)
Sex					
Male	13 (54.2%)	7 (35.0%)	26 (66.7%)	7 (43.8%)	53 (53.5%)
Female	11 (45.8%)	13 (65.0%)	13 (33.3%)	9 (56.2%)	46 (46.5%)
Race					
White	18 (75.0%)	15 (75.0%)	30 (76.9%)	14 (87.5%)	77 (77.8%)
Black or African American	2 (8.3%)	0	0	0	2 (2.0%)
Asian	1 (4.2%)	4 (20.0%)	6 (15.4%)	0	11 (11.1%)
Other[a]	3 (12.5%)	1 (5.0%)	3 (7.7%)	2 (12.5%)	9 (9.1%)
Renal Function[b]					
Normal	12 (50.0%)	6 (30.0%)	13 (33.3%)	13 (81.2%)	44 (44.4%)
Mild RI	12 (50.0%)	10 (50.0%)	19 (48.7%)	3 (18.8%)	44 (44.4%)
Moderate RI	0	4 (20.0%)	7 (17.9%)	0	11 (11.1%)
Hepatic Function[b,c]					
Normal	24 (100%)	17 (85.0%)	37 (94.9%)	16 (100%)	94 (94.9%)
Mild HI	0	2 (10.0%)	2 (5.1%)	0	4 (4.0%)
Moderate HI	0	1 (5.00%)	0	0	1 (1.0%)
Age category					
≥18 years	24 (100%)	4 (20.0%)	17 (43.6%)	0	45 (45.5%)
12 to <18 years	0	6 (30.0%)	6 (15.4%)	0	12 (12.1%)
6 to <12 years	0	10 (50.0%)	16 (41.0%)	0	26 (26.3%)
1 to <6 years	0	0	0	14 (87.5%)	14 (14.1%)
<1 year	0	0	0	2 (12.5%)	2 (2.0%)
Body weight category					
>70 kg	15 (62.5%)	4 (20.0%)	15 (38.5%)	0	34 (34.3%)
40 to <70 kg	9 (37.5%)	8 (40.0%)	10 (25.6%)	0	27 (27.3%)
20 to <40 kg	0	8 (40.0%)	12 (30.8%)	2 (12.5%)	22 (22.2%)
10 to <20 kg	0	0	2 (5.1%)	11 (68.8%)	13 (13.1%)
<10 kg	0	0	0	3 (18.8%)	3 (3.0%)
ADA[d]					
Absence (Negative)	24 (100%)	18 (90.0%)	28 (71.8%)	14 (87.5%)	84 (84.8%)
Presence (Positive)	0	2 (10.0%)	1 (2.6%)	2 (12.5%)	5 (5.1%)
Not available at data cut-off	0	0	10 (25.6%)	0	10 (10.1%)

Abbreviations: ADA=anti-drug antibody; DB=double-blind; FDA=(US) Food and Drug Administration; HI=hepatic impairment; NCI-ODWG=National Cancer Institute Organ Dysfunction Working Group; PH1=primary hyperoxaluria type 1; PK=pharmacokinetic; RI=renal impairment; ULN=upper limit of normal; US=United States.

Source: Applicant's Population PK Analysis Report Table 7.

Lumasiran PK was best described by a 2-compartment disposition model with first-order absorption process from the SC injection site into systemic circulation. Total plasma clearance (CL_P) was a sum of two elimination pathways: hepatic uptake clearance (CL_H) and renal clearance (CL_R). CL_R for lumasiran was fixed to the patient's baseline renal function (eGFR* BSA/1.73). The volume of central compartment (V_2) was fixed to 0.07 L/kg*body weight based on nonclinical study results. The effect of dose (mg/kg) on CL_H was described using a power function with a dose of 3 mg/kg for the typical subject. The covariate effect of body weight on PK parameters was incorporated using allometric exponents (fixed to 0.75 for clearances and 1.0

for volumes) with a body weight of 70 kg for the typical subject. Inter-individual variabilities were added to parameters CL_H and first-order rate constant of absorption (K_a). Inter-occasion variability was added on K_a to explain random PK differences observed over multiple occasions of PK sampling within a subject. Random residual error was described using a proportional error structure.

The parameter estimates and the bootstrap results for the final PK model are presented in Table 53. The median and 95% CI of the parameter estimates were largely overlapping with bootstrap derived estimates, which supported the robustness of the final model. The goodness-of-plots in Figure 14, visual predictive check (VPC) in Figure 15 and prediction-corrected VPC in

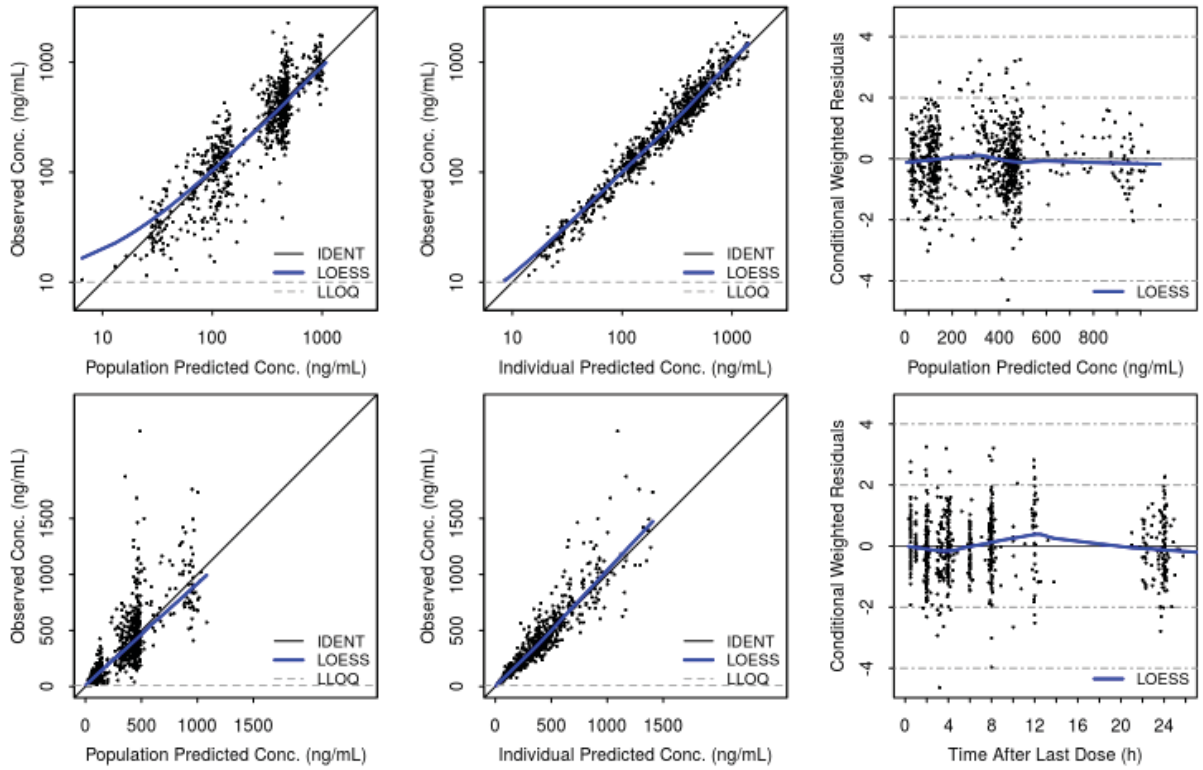
Figure 16 showed that the final PK model adequately described the plasma concentration-time profile of lumasiran.

Table 53. Parameter Estimates and Bootstrap Results for the Final Lumasiran Population PK Model

PK Parameters	Population Estimates	RSE (%)	Shrinkage (%)	Paramertric 95% CI	Bootstrap Median	Bootstrap 95% CI
K_a (h^{-1})	0.101	2.61	17.3	0.090, 0.11	0.10	0.089, 0.12
Weight on K_a	-0.312	24.6	NA	-0.46, -0.16	-0.32	-0.47, -0.16
CL_H (L/h)	21.1	1.09	20.7	19.8, 22.5	21.1	19.8, 22.5
Dose on CL_H	-0.166	23.9	NA	-0.24, -0.088	-0.17	-0.25, -0.083
Q (L/h)	45.9	3.19	NA	36.2, 58.3	46.5	35.3, 59.7
V_3 (L)	58.0	3.23	NA	44.8, 74.9	58.5	43.6, 77.1
IIV on CL_P (%)	21.1	11.1	NA	15.9, 25.3	20.7	15.7, 25.4
IIV on K_a (%)	38.7	13.0	NA	27.1, 47.6	37.6	22.7, 49.8
IOV on K_a (%)	31.0	9.50	NA	24.6, 36.4	30.7	23.7, 36.9
Residual error	26.6%	3.87	NA	24.6, 28.6	26.4	24.6, 28.5

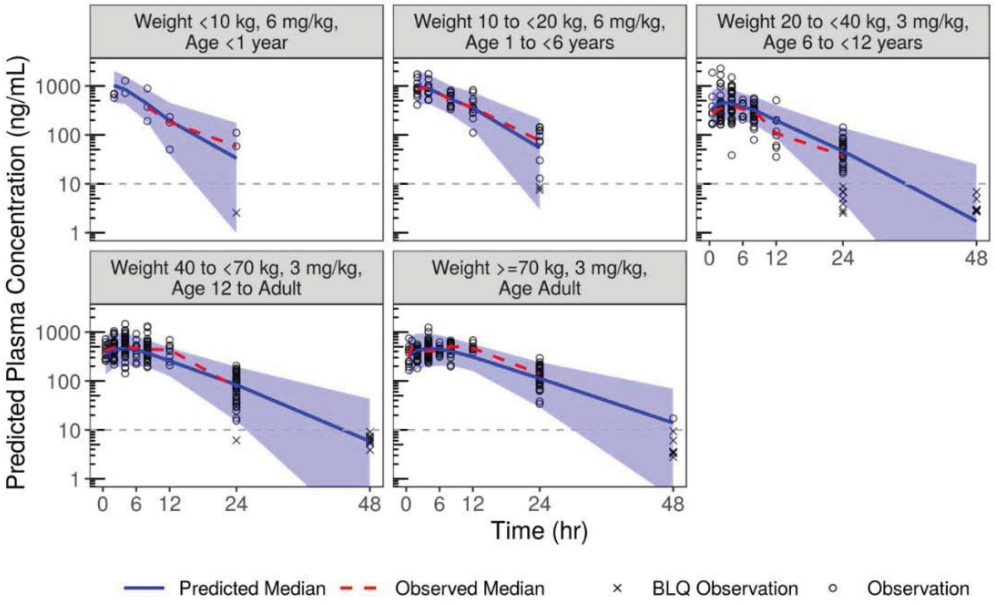
Abbreviations: CI=confidence interval; CL_H=hepatic uptake clearance; IIV=inter-individual variability; IOV=inter-occasion variability; K_a=first-order absorption rate constant; NA=not applicable; PK=pharmacokinetic; Q=apparent inter-compartmental clearance between central and peripheral compartment; RSE=relative standard error; V_2=volume of distribution in central compartment, V_3=volume of distribution in peripheral compartment.
Note: Dose is weight based (mg/kg). Weight is time-varying body weight in kg.
Source: Applicant's Population PK Analysis Report Table 11 and Section 9.33.

Figure 14. Goodness-of-Fit Plots for the Final PK Model



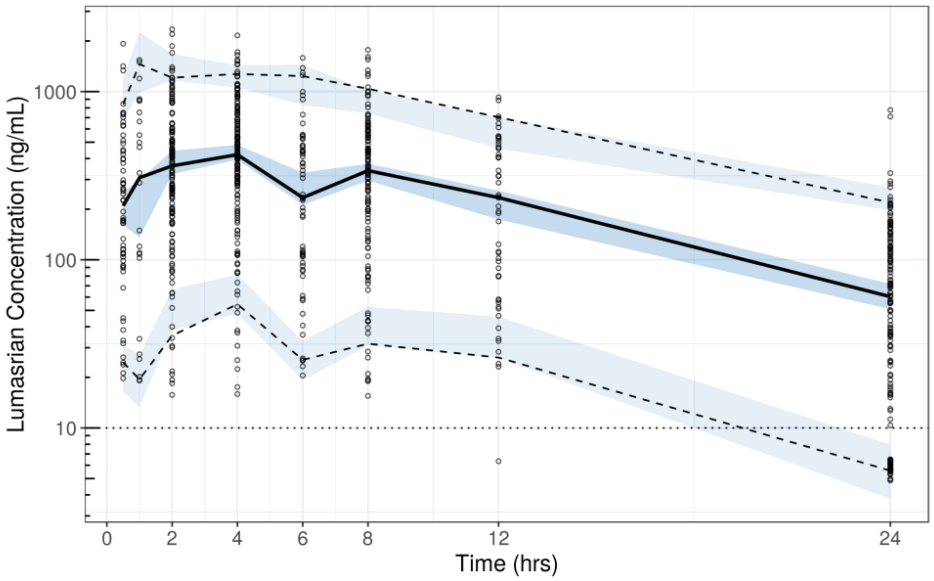
Source: Applicant's Population PK Analysis Report Figure 12.

Figure 15. Visual Predictive Checks for the Final PK Model by Body Weight Categories at the Recommended Dose



Source: Applicant's Population PK Analysis Report Figure 13.

Figure 16. Prediction Corrected Visual Predictive Checks for the Final PK Model



Source: Applicant's Population PK Analysis Report Figure 14.

Predicted PK parameters in patients with PH1 across the range of body weights (<10 to ≥70 kg) are listed in Table 54. Both CL_P and $t_{1/2}$ values increase with body weight, which justified the BW-based dosing regimens in different weight groups.

Table 54. Model Predicted Lumasiran PK Parameters Clearance and Half-Life in Patients With PH1 Across Range of Body Weights

Plasma PK Parameter	Body Weight Category / Proposed Loading or Maintenance Dose					
	<10 kg/ 3 mg/kg	<10 kg/ 6 mg/kg	10 to <20 kg 6 mg/kg	20 to <40 kg/ 3 mg/kg	40 to <70 kg/ 3 mg/kg	≥70 kg/ 3 mg/kg
CL (L/h)						
Mean (SD)	5.10 (1.40)	4.57 (1.28)	7.98 (2.14)	14.4 (3.75)	23.5 (5.16)	32.4 (8.28)
Median [5th-95th]	5.02 [2.94-7.47]	4.43 [2.72-6.69]	7.84 [4.94-11.9]	14.0 [9.09-21.8]	22.8 [16.2-32.8]	31.1 [20.9-47.2]
Weight-Normalized CL (L/h/kg)						
Mean (SD)	0.724 (0.162)	0.650 (0.156)	0.560 (0.126)	0.509 (0.116)	0.419 (0.0859)	0.365 (0.0830)
Median [5th-95th]	0.703 [0.496-1.02]	0.627 [0.425-0.943]	0.547 [0.376-0.787]	0.495 [0.334-0.695]	0.410 [0.297-0.575]	0.358 [0.247-0.512]
$t_{1/2}$ (h)						
Mean (SD)	3.67 (1.86)	3.95 (2.26)	4.76 (2.33)	5.86 (3.12)	7.28 (3.51)	8.81 (4.57)
Median [5th-95th]	3.20 [1.61-7.13]	3.48 [1.58-8.05]	4.25 [1.98-8.95]	5.16 [2.46-11.8]	6.46 [2.98-13.7]	7.63 [3.65-17.0]

Abbreviations: CL=clearance; NHANES=National Health and Nutrition Examination Survey; PK=pharmacokinetic; SD=standard deviation; $t_{1/2}$=terminal elimination half-life.

Source: Applicant's Population PK Analysis Report Table 13.

Renal function was incorporated into the structural model as CL_R fixed to baseline renal function. Renal impairment is predicted to result in lowering of urinary excretion of urine (<30%) leading to transiently higher circulating plasma concentrations and consequently a higher fraction available for liver uptake (>70%) in adult and pediatric patients across different weight groups (Table 55). Renal impairment is expected to have minimal impact on bioavailable fraction of dose to the liver and thus, no dose adjustment is required in patients with renal impairment.

Table 55. Model Derived PK Parameters and Fraction of Lumasiran Dose Available in Liver by Body Weight and eGFR

Age (years)	Body Weight (kg)	BSA (m^2) [a]	CL_H (L/h)	CL_R (L/h)	CL_P (L/h)	Ka (h^{-1})	Half-life (h)	eGFR (mL/min/1.73m^2)			
								90	60	30	15
								Fraction available to Liver (%)			
Adult	110	2.27	29.57	7.09	36.66	0.0878	7.9	80.7%	86.2%	92.6%	96.2%
Adult	70	1.73	21.07	5.40	26.47	0.1011	6.9	79.6%	85.4%	92.1%	95.9%
12	40[c, d]	1.29	13.85	4.03	17.88	0.1204	5.8	77.5%	83.8%	91.2%	95.4%
6	20[c, d]	0.81	8.23	2.53	10.76	0.1495	4.6	76.5%	83.0%	90.7%	95.1%
2	12.7 [c]	0.55	5.86	1.72	7.58	0.1722	4.0	77.3%	83.7%	91.1%	95.3%
1	10[b, d]	0.47	4.90	1.47	6.37	0.1855	3.7	76.9%	83.4%	90.9%	95.2%
0.5	6.2[b]	0.39	3.42	1.22	4.64	0.2154	3.2	73.7%	80.8%	89.4%	94.4%

Abbreviations: BSA=body surface area; CDC=Centers for Disease Control and Prevention; eGFR=estimated glomerular filtration rate; PH1=primary hyperoxaluria type 1.
[a] BSA was calculated using the Mosteller formula: BSA=square root (height (cm)*weight (kg)/3600)
[b] CDC. Birth to 36 months: Boys Length-for-age and Weight-for-age percentiles. Growth Charts. 2000 Section 9.15
[c] CDC. 2 to 20 years: Boys Stature-for-age and Weight-for-age percentiles. Growth Charts. 2000.Section 9.16
[d] approximate body weight
Note: CLH = 21.07*(WT/70)^0.75; CLR = eGFR*60/1000*(BSA/1.73); CLP=CLH+CLR; Ka=0.1011 * (WT/70)^-0.312; Half life=0.693/Ka; %Liver = CLH/(CLH+CLR)*100 based on final popPK model.

Source: Applicant's Population PK Analysis Report Section 9.34.

Covariate analysis showed that the effects of age, sex, race (white versus non-white), disease status (healthy adult subjects versus patients with PH1), presence of ADA, and mild hepatic impairment did not significantly impact lumasiran PK.

Reviewer's comments:

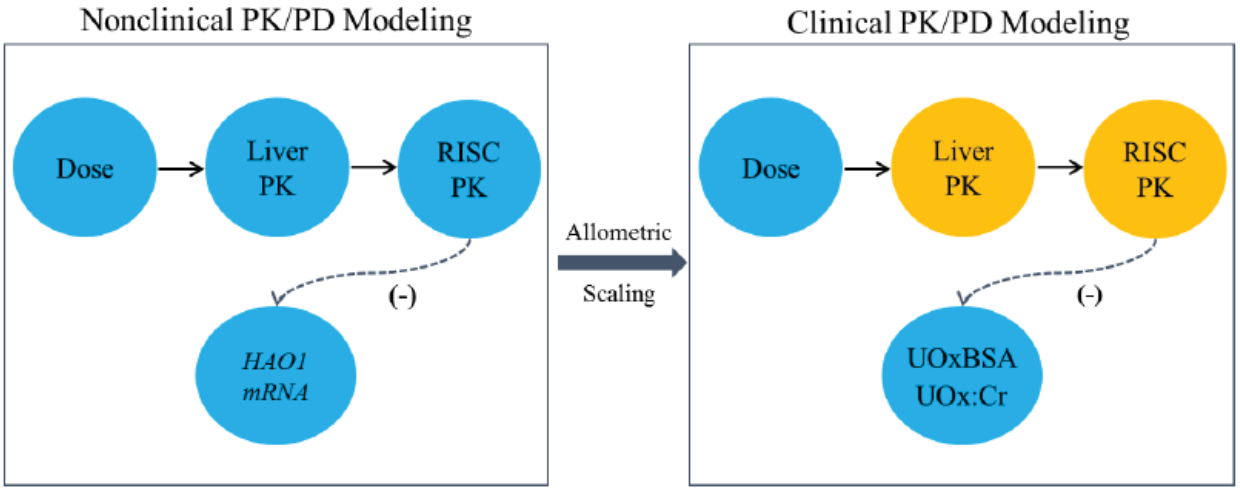
The Applicant's population PK model appears to capture the central tendency of the lumasiran plasma PK time profile. The PK parameters were estimated generally precisely with relatively low shrinkage values for the key PK parameters. Therefore, the population PK model is acceptable and supports the USPI Sections 8.4, 8.6, 8.7, and 12.3 regarding the descriptive PK properties for lumasiran and no dose adjustment for age (0 to 60 years), sex, race, presence of ADA, mild hepatic impairment (total bilirubin ≤ULN and AST >ULN; or total bilirubin >1.0 to 1.5×ULN) and mild to moderate renal impairment (eGFR 30 to <90 mL/min/1.73 m^2).

14.2.2. Population PK/PD Analyses

14.2.2.1. Applicant's Modeling Strategy

The Applicant conducted the population PK/pharmacodynamic (PD) analyses to characterize the E-R relationships between human lumasiran RISC-loaded PK and two PD endpoints: $UOxBSA_{24h}$ and Ox/Cr. The human lumasiran RISC-loaded PK concentrations were derived from concentrations projected in the liver based on allometry and originating from a nonclinical PK/PD model developed with rat and monkey data. Their approach to scaling this model is shown in Figure 17 with the blue shaded variables indicating observed data and orange shaded variables indicating predicted data based on allometric scaling.

Figure 17. Schematic Representation of Lumasiran Clinical PK/PD Models Leveraging Nonclinical PK/PD Information



Abbreviations: BSA=body surface area; *HAO1=hydroxyacid oxidase 1*; mRNA=messenger ribonucleic acid; PD=pharmacodynamic; PK=pharmacokinetic, RISC=RNA-induced silencing complex; UOx:Cr=spot urinary oxalate:creatinine ratio; UOxBSA=24-hour urinary oxalate corrected for BSA.

Source: Applicant's Population PK/PD Analysis Report Figure 3.

In brief, the Applicant took the following steps for the development of mechanistic PK/PD models:

- A nonclinical PK/PD model was developed to describe the relationship between observed liver and RISC-loaded concentrations of lumasiran and changes in HAO1 mRNA following the administration of single SC doses of lumasiran at 1, 5, or 10 mg/kg in Sprague-Dawley rats in Study GO1-DSM19-023. A maximum inhibitory (I_{max}) model best described the relationship between RISC-loaded lumasiran and increased degradation rate of HAO1 mRNA, enabling estimation of an half maximal inhibitory concentration (IC_{50}) value.
- All the lumasiran PK parameters except clearance from the rat liver PK model and clearance from the monkey liver PK model were allometrically scaled to predict lumasiran concentrations in human liver.
- Human RISC-loaded concentrations of lumasiran (i.e., effect compartment concentrations) were predicted using scaled liver PK from rats and monkeys (from Step 2 above) and observed PD (urinary oxalate levels) from the clinical studies.
- The relationship between predicted RISC-loaded lumasiran concentrations and decrease in urinary oxalate measures was described as an I_{max} model using the $UOxBSA_{24h}$ and Ox/Cr ratio data from 75 patients with PH1 in Trial 001B, 002, 003 and 004 across a wide range of dose levels (1 to 6 mg/kg) and varying dose frequencies (qM, q3M and their combinations).

14.2.2.2. Description of Baseline Demographics

Descriptive statistics of the continuous and categorical demographic data included in the PK/PD modeling are summarized in Table 56 and Table 57, respectively.

Table 56. Baseline Characteristics of PK/PD Population by Study (Continuous Covariates)

Baseline Characteristics	Study 001B/002[a]	Study 003	Study 004	Total
	N=20	N=39	N=16	N=75
Age at consent (years)				
Mean (SD)	14.9 (10.2)	18.1 (12.7)	3.13 (1.61)	14.1 (12.0)
Median (min, max)	11.5 [6.00, 43.0]	14.0 [6.00, 60.0]	3.50 [0.167, 5.00]	10.0 [0.167, 60.0]
Age at first dose in Study 004 (years)[b]				
Mean (SD)			3.58 (1.771)	
Median (min, max)			3.88 [0.3, 6.1]	
Body Weight (kg)				
Mean (SD)	49.9 (27.2)	58.2 (27.6)	14.5 (4.87)	46.6 (29.7)
Median (min, max)	42.8 [21.3, 110]	59.6 [17.3, 107]	13.6 [6.20, 24.3]	38.8 [6.20, 110]
Height (cm)				
Mean (SD)	148 (21.3)	156 (23.3)	96.3 (16.6)	141 (31.9)
Median (min, max)	148 [120, 188]	163 [112, 195]	96.3 [58.4, 116]	147 [58.4, 195]
BSA (m^2)				
Mean (SD)	1.41 (0.474)	1.56 (0.495)	0.619 (0.154)	1.32 (0.572)
Median (min, max)	1.34 [0.852, 2.40]	1.70 [0.753, 2.33]	0.586 [0.317, 0.878]	1.27 [0.317, 2.40]
eGFR (mL/min/1.73 m^2)				
Mean (SD)	78.5 (21.0)	81.6 (25.7)	113 (28.1)	87.5 (28.2)
Median (min, max)	74.0 [50.9, 131]	83.4 [31.7, 131]	114 [64.7, 174]	85.9 [31.7, 174]
Baseline UOxBSA (mmol/24hour/1.73 m^2)				
Mean (SD)	2.32 (0.974)	1.82 (0.618)	1.97 (0.763)[c]	1.98 (0.774)
Median (min, max)	2.29 [0.941, 5.18]	1.72 [0.677, 3.05]	1.98 [1.03, 2.89][c]	1.87 [0.677, 5.18]
Baseline UOx:Cr (mmol/mmol)				
Mean (SD)	0.271 (0.144)	0.229 (0.119)	0.618 (0.418)	0.323 (0.269)
Median (min, max)	0.222 [0.116, 0.631]	0.213 [0.0715, 0.596]	0.469 [0.166, 1.71]	0.259 [0.0715, 1.71]
Baseline Plasma Oxalate (µmol/L)				
Mean (SD)	15.1 (6.37)	15.0 (7.44)	12.9 (6.69)	14.6 (6.99)
Median (min, max)	13.6 [6.34, 28.7]	13.1 [6.99, 43.5]	10.5 [6.58, 30.6]	13.1 [6.34, 43.5]

Source: Applicant's Population PK/PD Analysis Report Table 7.

Table 57. Baseline Characteristics of PK/PD Population (Categorical Covariates)

Baseline Characteristics[a]	Study 001B/002[b]	Study 003	Study 004	Total
	N=20	N=39	N=16	N=75
Sex, n (%)				
Female	13 (65.0%)	13 (33.3%)	9 (56.2%)	35 (46.7%)
Male	7 (35.0%)	26 (66.7%)	7 (43.8%)	40 (53.3%)
Race, n (%)				
White	15 (75.0%)	30 (76.9%)	14 (87.5%)	59 (78.7%)
Black or African American	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Asian	4 (20.0%)	6 (15.4%)	0 (0%)	10 (13.3%)
Other	1 (5.0%)	3 (7.7%)	2 (12.5%)	6 (8.0%)
Age categories, n (%)				
<1 year	0 (0%)	0 (0%)	2 (12.5%)	2 (2.7%)
1 to <2 years	0 (0%)	0 (0%)	2 (12.5%)	2 (2.7%)
2 to <6 years	0 (0%)	0 (0%)	12 (75.0%)	12 (16.0%)
6 to <12 years	10 (50.0%)	16 (41.0%)	0 (0%)	26 (34.7%)
12 to <18 years	6 (30.0%)	6 (15.4%)	0 (0%)	12 (16.0%)
≥18 years	4 (20.0%)	17 (43.6%)	0 (0%)	21 (28.0%)
Body weight categories, n (%)				
<10 kg	0 (0%)	0 (0%)	3 (18.8%)	3 (4.0%)
10 to <20 kg	0 (0%)	2 (5.1%)[d]	11 (68.8%)	13 (17.3%)
20 to <40 kg	8 (40.0%)	12 (30.8%)	2 (12.5%)	22 (29.3%)
40 to <70 kg	8 (40.0%)	10 (25.6%)	0 (0%)	18 (24.0%)
≥70 kg	4 (20.0%)	15 (38.5%)	0 (0%)	19 (25.3%)
Renal function, n (%)				
Normal	6 (30.0%)	13 (33.3%)	13 (81.2%)	32 (42.7%)
Mild	10 (50.0%)	19 (48.7%)	3 (18.8%)	32 (42.7%)
Moderate	4 (20.0%)	7 (17.9%)	0 (0%)	11 (14.7%)
Severe	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Hepatic function, n (%)				
Normal[c]	17 (85.0%)	38 (97.4%)	16 (100%)	71 (94.7%)
Mild	2 (10.0%)	1 (2.6%)	0 (0%)	3 (4.0%)
Moderate	1 (5.0%)	0 (0%)	0 (0%)	1 (1.3%)
***AGXT* genotype, n (%)[e]**				
M/M	9 (45.0%)	8 (20.5%)	7 (43.8%)	24 (32.0%)
N/N	8 (40.0%)	12 (30.8%)	5 (31.2%)	25 (33.3%)
M/N	1 (5.0%)	2 (5.1%)	1 (6.2%)	4 (5.3%)
PR/*	2 (10.0%)	17 (43.6%)	3 (18.8%)	22 (29.3%)

Baseline Characteristics[a]	Study 001B/002[b]	Study 003	Study 004	Total
	N=20	N=39	N=16	N=75
Concomitant pyridoxine, n (%)				
Yes	13 (65.0%)	22 (56.4%)	10 (62.5%)	45 (60.0%)
No	7 (35.0%)	17 (43.6%)	6 (37.5%)	30 (40.0%)

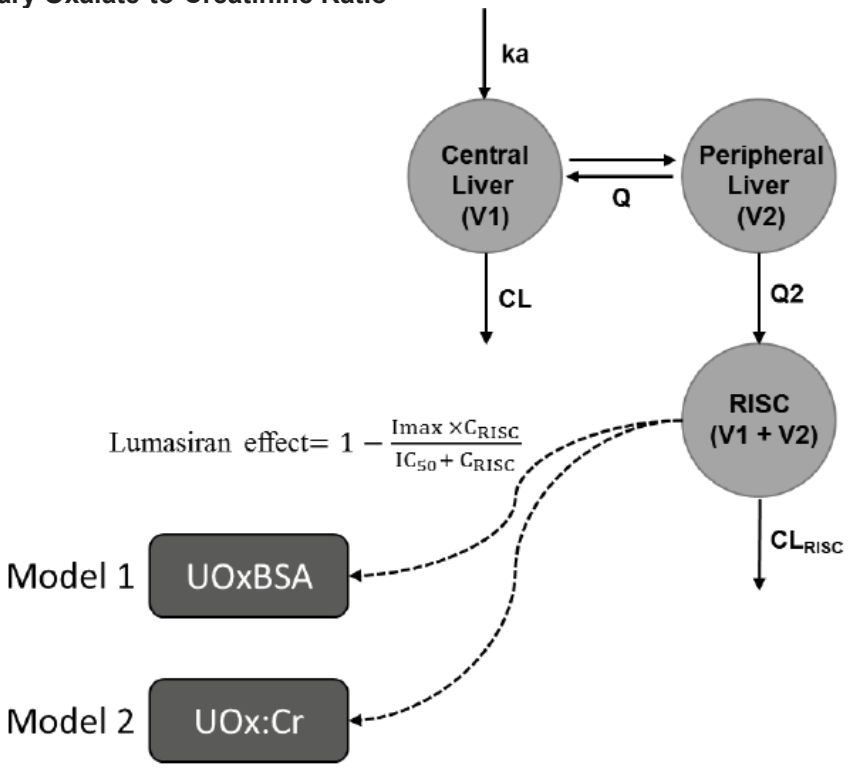
Source: Applicant's Population PK/PD Analysis Report Table 8.

14.2.2.3. Base Model Structure

The schematic of population PK/PD Model is shown in Figure 18, which was based on the mechanistic knowledge of lumasiran PK/PD from nonclinical data and oxalate kinetics. The PK/PD models for $UOxBSA_{24h}$ and Ox/Cr ratio were developed separately:

- **Model 1** for $UOxBSA_{24h}$ in patients with PH1 ≥6 years of age, as this measure was available for subjects only in this age range
- **Model 2** for Ox/Cr ratio in patients with PH1 of all ages, infants to adults

Figure 18. Schematic of Population PK/PD Models for 24-hour Urinary Oxalate Corrected for BSA and Spot Urinary Oxalate-to-Creatinine Ratio



Abbreviations: BSA=body surface area; CL=clearance; C_{RISC}=RISC-loaded concentration; CL_{RISC}=clearance from RISC compartment; ka=absorption rate constant of lumasiran in liver; IC_{50}=RISC-loaded concentration of lumasiran producing 50% of the maximal effect; I_{max}=maximum inhibition; PD=pharmacodynamic; PK=pharmacokinetic; Q=inter-compartmental liver clearance; Q2=rate of uptake into RISC compartment; UOxBSA=24-hour urinary oxalate corrected for BSA; UOx:Cr=spot urinary oxalate:creatinine ratio; V1=volume of central liver compartment; V2=volume of peripheral liver compartment.
Source: Applicant's Population PK/PD Analysis Report Figure 16.

14.2.2.4. Final PK/PD Model for $UOxBSA_{24h}$

The parameter estimates of the final population PK/PD model for $UOxBSA_{24h}$ are presented in Table 58. The final PK/PD model converged with low condition number (<100), parameters were estimated with reasonable precision (<30%), and acceptable shrinkage of random effects (<40%).

Table 58. Parameter Estimates of Final PK/PD Model for 24-Hour Urinary Oxalate Corrected for BSA

PK/PD Parameters	Estimates (Log-transformed)	Estimates (Untransformed)	RSE	IIV (RSE)	Lower 95% CI	Upper 95% CI
ka (h^{-1})		0.119 (fixed)[a]	NA			
CL (g/h)		$2.364\times(WT/70)^{0.75}$ (fixed)[a]				
Q (g/h)		$24.368\times(WT/70)^{0.75}$ (fixed)[a]				
V1 (g)		$1392\times(WT/70)$ (fixed)[a]				
V2 (g)		$3808\times(WT/70)$ (fixed)[a]				
Q2 (g/h)		$0.000254\times(WT/70)^{0.75}$ (fixed)[b]				
CL_{RISC} (g/h)		$1.91\times(WT/70)^{0.75}$	13.8%		1.393	2.427
I_{max}	-0.345	0.708	6.35%	7.93% (22.3%)	0.678	0.739
IC_{50} (ng/g)	-8.19	0.277	2.38%		0.189	0.406
Baseline UOxBSA (mmol/24hr/1.73m^2)		1.89	5.10%	27.8% (11.8%)	1.701	2.079
Baseline UOxBSA on I_{max}[c]		0.220	25.1%		0.112	0.328
eGFR on baseline UOxBSA[d]		-0.557	19.6%		-0.771	-0.343
AGXT genotype of PR/* on baseline UOxBSA[d]		-0.294	20.2%		-0.410	-0.178
Residual Error		29.8%	5.81%		26.4%	33.2%
OFV	-863.168					

Abbreviations: *AGXT*=alanine-glyoxylate aminotransferase gene; CI=confidence interval; CL=clearance; CL_{RISC}=clearance of RISC-loaded drug; IC_{50}=lumasiran concentration in RISC required to reach 50% of maximum inhibition; IIV=inter-individual variability; I_{max} =maximum inhibitory effect of lumasiran; ka=uptake rate constant to liver; OFV=objective function value ; PD=pharmacodynamic; PK=pharmacokinetic; PR/*=pyridoxine-responsive allele with any other allele; Q=intercompartmental clearance; Q2= turnover rate of lumasiran from peripheral liver compartment into RISC; RSE=relative standard error; UOxBSA=24-hour urinary oxalate corrected for BSA; V1=volume of distribution of central liver compartment; V2=volume of distribution of peripheral liver compartment; WT=body weight.

[a] Liver PK parameters were fixed to allometrically scaled values from the rat or monkey liver PK model

[b] Q2 was fixed to the estimate from the previous PK/PD model (run006)

[c] Covariate-I_{max} relationship: $Imax = \text{TVImax} \times \left(\frac{Baseline\ UOxBSA}{1.85}\right)^{0.220}$

[d] Covariates-baseline relationship: $\text{Baseline} = \text{TVBaseline} \times \left(\frac{eGFR}{80}\right)^{-0.557} \times (1 + \text{GenotypePR} \times (-0.294))$

Parameter estimates are for a typical PH1 subject in UOxBSA modeling dataset who is 13 years old, body weight of 52.3 kg, eGFR of 80 mL/min/1.73m^2 and with *AGXT* genotype of M/M or M/N or N/N.

Source: Applicant's Population PK/PD Analysis Report Table 11.

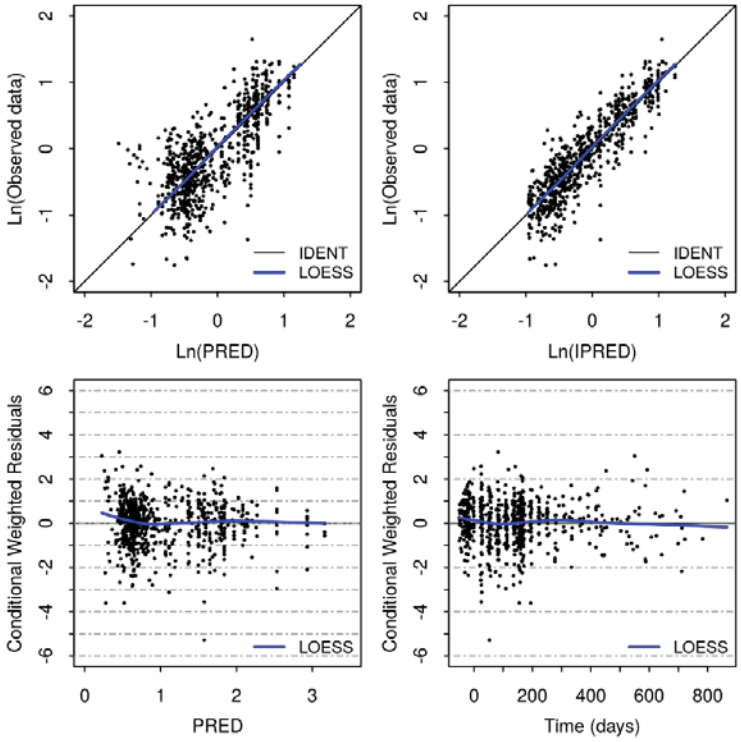
The covariate analysis identified three statistically significant covariates in the final population PK/PD model for $UOxBSA_{24h}$: baseline eGFR and AGXT genotype of PR/* on baseline $UOxBSA_{24h}$ parameter, as well as baseline $UOxBSA_{24h}$ on the maximum effect of lumasiran (I_{max}) parameter.

- The model indicated that renal impairment (i.e., low eGFR) was associated with higher baseline $UOxBSA_{24h}$ (47.1% higher for patient with eGFR of 45 mL/min/m^2 and 84.4% higher for patient with eGFR of 30 mL/min/m^2 relative to normal renal function). The association of lower eGFR with higher baseline $UOxBSA_{24h}$ can be attributed to PH1 patients with a higher baseline having more severe disease and a higher likelihood of progression to renal impairment.
- PH1 patients with AGXT genotype PR/* had 29.4% lower baseline $UOxBSA_{24h}$.
- Baseline $UOxBSA_{24h}$ was found to be a significant covariate on I_{max} such that higher percent reduction from baseline is predicted for patients with higher baseline $UOxBSA_{24h}$: percent reduction in patients with baseline values of 1.85 (observed median) and 5.18 (observed maximum) mmol/24 h/1.73 m^2 were predicted to be 70.8 and 88.8%, respectively. Patients with lower baseline oxalate levels have lower I_{max} due to floor effect.
- Other covariates such as age, sex, race, BSA, time since PH1 diagnosis, mild hepatic impairment and pyridoxine use were not significant and were not included in the final model.

The model-estimated IC_{50} of RISC-loaded lumasiran was 0.277 ng/g in patients with PH1. Predicted RISC-loaded lumasiran concentrations in the hepatocytes of patients with PH1 weighing ≥20 kg receiving 3 mg/kg quarterly doses ranged from 5 to 8 ng/g (>10-fold higher than the estimated IC_{50} of 0.277 ng/g), indicating that the 3 mg/kg quarterly dose of lumasiran is at the asymptote of the E-R curve.

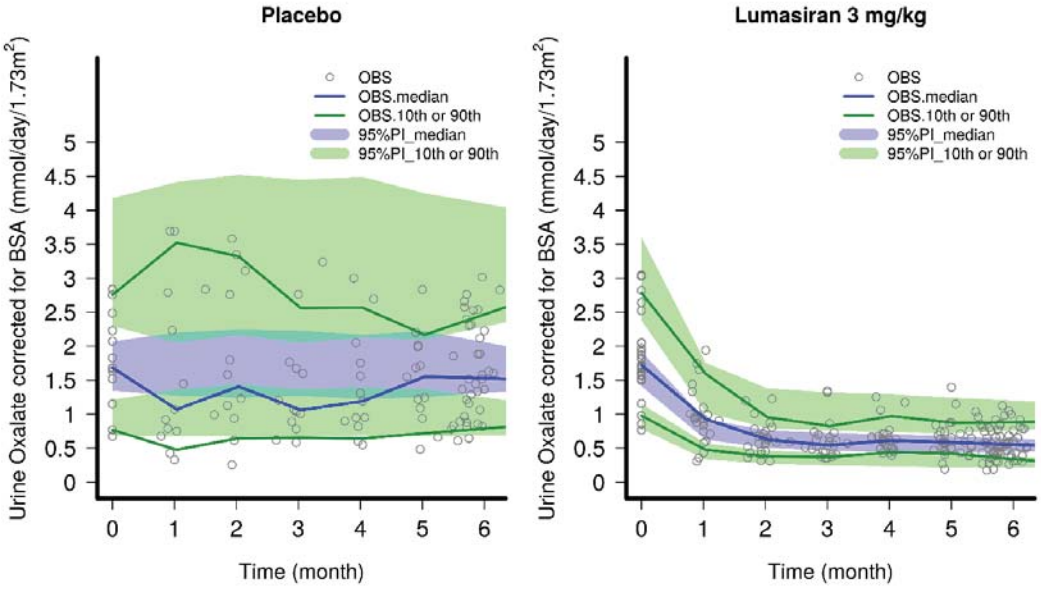
The goodness-of-fit plots (Figure 19) and VPC (Figure 20) showed that the final PK/PD model adequately fit the observed time profiles of $UOxBSA_{24h}$ in patients with PH1.

Figure 19. Goodness-of-Fit Plots of Final PK/PD Model for 24-Hour Urinary Oxalate Corrected for BSA



Source: Applicant's Population PK/PD Analysis Report Figure 17.

Figure 20. Visual Predictive Check for 24-hour Urinary Oxalate Corrected for BSA in Trial 003 (Double-Blind Period)



Source: Applicant's Population PK/PD Analysis Report Figure 18.

14.2.2.5. Final PK/PD Model for Ox/Cr Ratio

The parameter estimates of the final population PK/PD model for Ox/Cr ratio are presented in Table 58. The final PK/PD model converged with low condition number (<100), parameters were estimated with reasonable precision (<30%), and acceptable shrinkage of random effects (<40%).

Table 59. Parameter Estimates of Final PK/PD Model for Spot Urinary Oxalate-to-Creatinine Ratio

PK/PD Parameters	Estimates (Log-transformed)	Estimates (Untransformed)	RSE	IIV[f] (RSE, %)	Lower 95% CI	Upper 95% CI
ka (h^{-1})	-	0.119 (fixed)[a]	NA			
V2 (g) (WT ≥20 kg)	-	2.36 × (WT/70) (fixed)[a]				
V2 (g) (WT <20 kg)	-	2.36 × $(WT/70)^{0.78}$ (fixed)[a]				
V3 (g) (WT ≥20 kg)	-	24.37 × (WT/70) (fixed)[a]				
V3 (g) (WT <20 kg)	-	24.37 × $(WT/70)^{0.78}$ (fixed)[a]				
CL (g/h)	-	1392 × $(WT/70)^{0.75}$ (fixed)[a]				
Q (g/h)	-	3808 × $(WT/70)^{0.75}$ |(fixed)[a]				
Q2 (g/h)	-	0. 000190 × $(WT/70)^{0.75}$ (fixed)[b]				
CL_{RISC} (g/h)	-	1.47 × $(WT/70)^{0.75}$	22.2	-	0.828	2.10
Adult baseline UOx:Cr (mmol/mmol)	-	0.200	5.61	30.4 (25.7)	0.178	0.222
Time-varying height effect on baseline (age <6 years)[c]	-	2.50	6.88	-	2.16	2.84
Time-varying height effect on baseline (age ≥6 years)[c]	-	2.21	12.4	-	1.67	2.75
Baseline eGFR effect on baseline[c]	-	-0.664	16.8	-	-0.882	-0.446
I_{max}[e]	-	0.729	2.30	7.21 (95.6)	0.696	0.762
Baseline UOx:Cr effect on I_{max}[d]	-	0.126	20.2	-	0.0761	0.176
IC_{50} (ng/g)	-8.15	0.288	1.91	73.8 (42.2)	0.212	0.391
Covariance between I_{max} and IC_{50}	-	0.0297	96.1	-	-0.0262	0.0856
Proportional Residual Error (%)	-	33.1	5.23	-	29.7	36.5

Abbreviations: CI=confidence interval; CL=clearance; CL_{RISC}= clearance of RISC loaded drug; Cr=creatinine; eGFR=estimated glomerular filtration rate; IC_{50}=lumasiran concentration in RISC required to reach 50% of maximum inhibition; IIV=inter-individual variability; I_{max}=maximum inhibitory effect of lumasiran; ka=uptake rate constant to liver; Ox=oxalate; PD=pharmacodynamic; PK=pharmacokinetic; Q=intercompartmental clearance; Q2=turnover rate of lumasiran from peripheral liver compartment into RISC; RSE=relative standard error; UOx:Cr=spot urinary oxalate:creatinine ratio; V2=volume of distribution of central liver compartment; V3=volume of distribution of peripheral liver compartment.; WT=body weight.

[a] Liver PK parameters were fixed to allometrically scaled values from the rat liver PK model

[b] Q2 was fixed to the estimate from previous PK/PD model (Run128)

[c] Covariates-baseline relationship: Baseline $\times \left[\left(\frac{147}{\text{Height}}\right)^{-2.30} (\text{if age} < 6 \text{ years old})\right] \times \left[\left(\frac{147}{\text{Height}}\right)^{-2.21} (\text{if age} \geq 6 \text{ years old})\right] \times \left(\frac{eGFR}{86}\right)^{-0.664}$

[d] Covariate-Imax relationship: $I_{max} \times \left(\frac{Baseline\ Urine\ Ox:Cr}{0.259}\right)^{0.126}$

[e] Because in the covariate-I_{max} relationship, effect was centered at the median baseline value of the analysis dataset (0.259 mmol/mmol); here, we recalculated Imax value for a patient with a 0.200 mmol/mmol baseline UOx:Cr

[f] IIV estimate is given as coefficient of variation, and RSE is given in variance the domain

Note: Parameter estimates are for a typical/average PH1 patient in the UOx:Cr modeling dataset who was 10 years of age, with weight 39 kg, height 147 cm, eGFR of 86 mL/min/1.73 m^2 and baseline UOx:Cr of 0.200 mmol/mmol (median baseline values of the modeling dataset).

Source: Applicant's Population PK/PD Analysis Report Table 13.

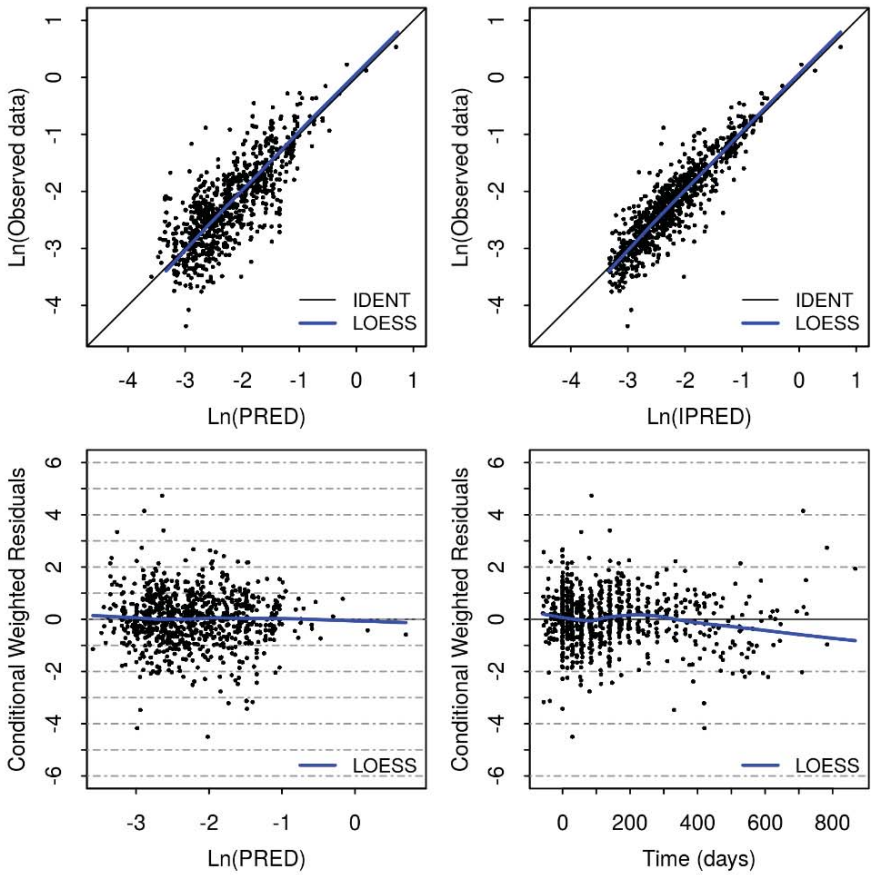
The covariate analysis identified three statistically significant covariates in the final population PK/PD model for Ox/Cr ratio: baseline eGFR and time-varying height on baseline Ox/Cr ratio parameter, as well as baseline Ox/Cr ratio on I_{max} parameter.

- The effect of eGFR on baseline Ox/Cr ratio can be attributed to renal function as a surrogate for the severity of PH1. The model indicated that, on average, PH1 patients with renal impairment (i.e., low eGFR) had higher baseline Ox/Cr ratio (0.194, 0.254, 0.307 and 0.402 mmol/mmol for eGFR of 90, 60, 45 and 30 ml/min/1.73 m^2, respectively).
- Other covariates such as age, sex, race, time since PH1 diagnosis, pyridoxine use, and AGXT genotype were not significant and therefore not included in the final model.

The model-estimated IC_{50} of RISC-loaded lumasiran was 0.289 ng/g in patients with PH1. Predicted RISC-loaded lumasiran concentrations in the hepatocytes of PH1 patients receiving the proposed dosing regimens based on body weight categories (<10 kg, 10 to <20 kg, and ≥20 kg) were similar and ranged from 5 to 8 ng/g. These steady-state RISC concentrations were at the asymptote of the E-R curve since they are >10-fold higher than the estimated IC_{50} of 0.289 ng/g.

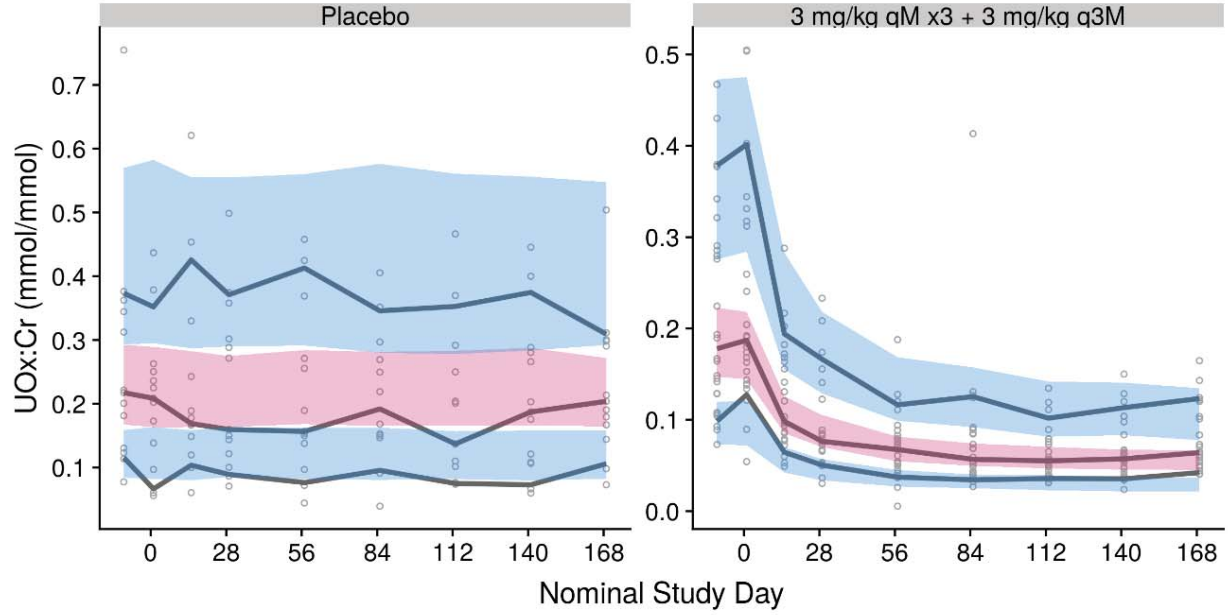
The goodness-of-fit plots (Figure 21) and VPC (Figure 22) showed that the final PK/PD model adequately fit the observed time profiles of Ox/Cr ratio in patients with PH1.

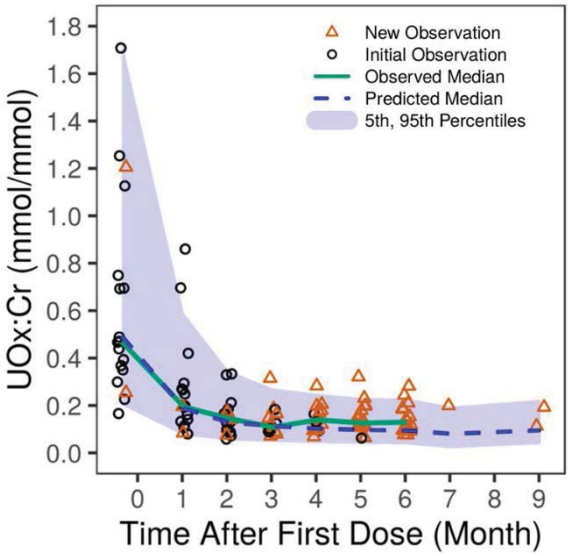
Figure 21. Goodness-of-fit Plots of Final PK/PD Model for Spot Urinary Oxalate-to-Creatinine Ratio



Source: Applicant's Population PK/PD Analysis Report Figure 22.

Figure 22. Visual Predictive Check for Spot Urinary Oxalate-to-Creatinine Ratio in Trials 003 and 004





Source: Applicant's Population PK/PD Analysis Report (Phase 3 Data) Figure 23 and (Additional ALN-GO1-004 Data) Figure 3.

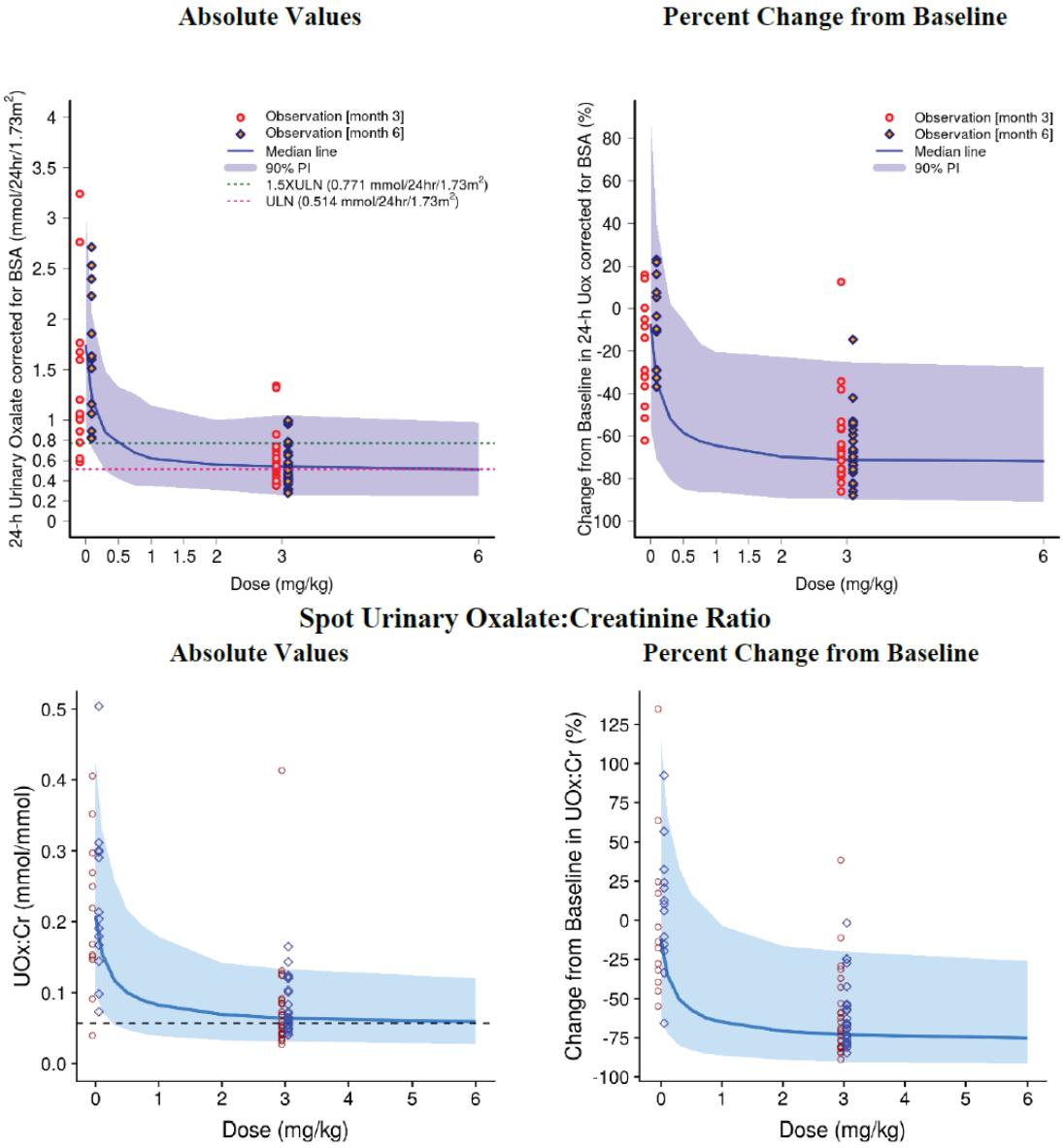
14.2.2.6. Model Applications: Justification of the Proposed Dosing Regimen

Dose Justification for PH1 Patients ≥6 Years of Age

Dose-Response Relationship of Quarterly Regimen

- Both models predicted a dose-dependent reduction in steady-state urinary oxalate levels approaching the ULN with increasing doses.
- At steady state, 3 mg/kg q3M is in the plateau portion of the dose-response curve for both absolute and percent reduction in urinary oxalate.
- There was no significant increase in urinary oxalate lowering with an increased dosing regimen beyond 3 mg/kg q3M, whereas a lower 1 mg/kg q3M dosing regimen resulted in suboptimal urinary oxalate lowering.
- Both models predicted steady-state urinary oxalate levels would be achieved at Month 12. The observed Month 3 urinary oxalate levels obtained with the loading dose regimen in Trial 003 was superimposable on steady state urinary oxalate predictions. These results further support the use of the proposed loading regimen to achieve steady-state levels by 3 months.

Figure 23. Steady-State Dose-Response Relationship for Urinary Oxalate After Quarterly Doses of Lumasiran



Abbreviations: BSA=body surface area; PI=prediction interval; ULN=upper limit of normal; UOx:Cr=spot urinary oxalate:creatinine ratio.
Note: All observed data from Study 003. Red circles=observed data at Month 3; blue diamonds=observed data at Month 6.
Dashed black line in lower panels is the 95th percentile of UOx:Cr in healthy subjects with the respective age range according to Matos et al [Matos 1999].
Source: ALN-GO1_Submission_PKPD/Analysis/UoxBSA/FinalModel/Final4/SIM/sim3/Postprocess/Plot/
Source: Applicant's Population PK/PD Analysis Report Figure 24.

Impact of Loading Regimen on Urinary Oxalate Lowering

Lumasiran 3 mg/kg q3M dosing regimen, without a loading dose regimen, was predicted to result in 22.9% and 42.8% of patients reaching ULN at Month 3 and Month 12, respectively. By comparison, with a loading dose regimen, the proportion of PH1 patients reaching ULN at

Month 3 increased to 40.6% and was stably maintained with a quarterly dosing regimen (Table 60).

Table 60. Comparison of Loading Versus No-loading Doses in Lumasiran Quarterly Doses: 24-Hour Urinary Oxalate Corrected for BSA

Month	UOxBSA[a] (mmol/24 h/1.73 m^2)		Proportion of Patients Below ULN (%)		Proportion of Subjects below 1.5×ULN (%)	
	Loading	No-loading	Loading	No-loading	Loading	No-loading
1	0.755 (0.443, 1.36)	0.762 (0.439, 1.33)	13.3	12.1	52.3	52.4
2	0.606 (0.331, 1.12)	0.679 (0.384, 1.18)	32.2	20.6	76.3	64.9
3	0.561 (0.295, 1.05)	0.656 (0.370, 1.15)	40.6	22.9	82.2	68
6	0.535 (0.273, 1.01)	0.583 (0.314, 1.04)	46.0	37.5	85.0	79.2
9	0.536 (0.272, 1.01)	0.560 (0.295, 1.01)	46.0	41.1	85.0	82.2
12	0.540 (0.273, 1.01)	0.551 (0.288, 0.995)	45.2	42.8	85.0	83.3

Abbreviations: PI=prediction interval: ULN=upper limit of normal; UOxBSA=24-hour urinary oxalate corrected for body surface area.
[a] UOxBSA values are median and 90% PI.
Source: Applicant's Population PK/PD Analysis Report Table 16.

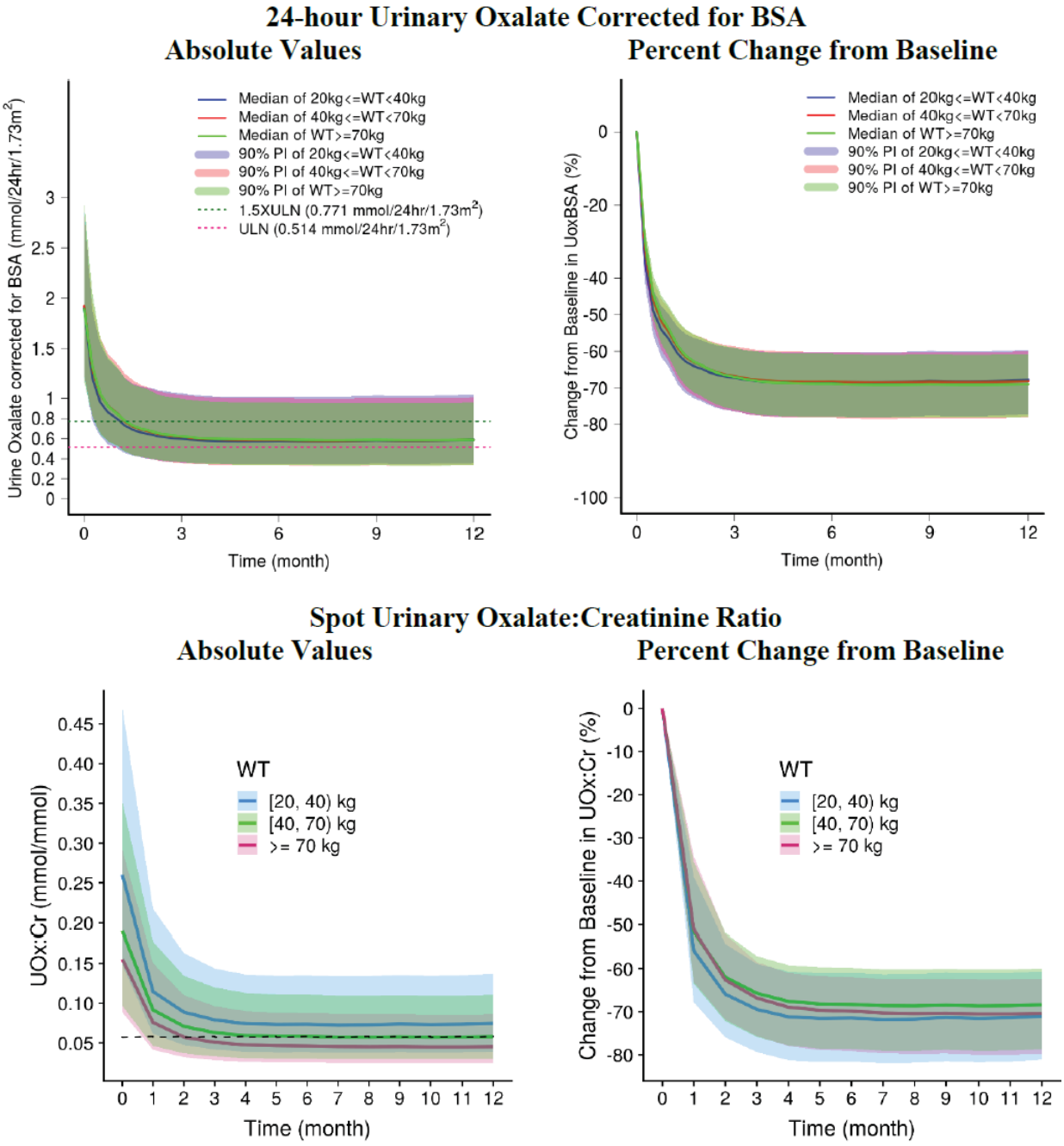
Predicted Liver and RISC PK Exposures in PH1 Patients at Proposed Dosing Regimen Across Body Weights

At proposed dosing regimens, the predicted concentration-time profiles of lumasiran in liver and RISC-loaded lumasiran levels were generally comparable across the range of body weights in patients with PH1. The model predicts that RISC-loaded lumasiran concentrations at Month 3 are >10-fold higher than estimated IC_{50} (0.277 ng/g from the $UOxBSA_{24h}$ model and 0.289 ng/g from Ox/Cr ratio model) resulting in near maximal PD lowering across the range of body weights (Figure 3).

Lumasiran Effect Across Various Body Weights

Simulations from the model indicated similar urinary oxalate lowering for different body weight groups (20≤BW<40 kg, 40≤ BW<70 kg, and BW≥70 kg) as shown in Figure 24. Baseline Ox/Cr ratio is higher in younger children due to the lower creatine production associated with low muscle mass.

Figure 24. Model-Predicted Values of Urinary Oxalate Across Body Weight Groups



Abbreviations: BSA=body surface area; PI=prediction interval; ULN=upper limit of normal; UOxBSA=24-hour urinary oxalate corrected for BSA; UOx:Cr=spot urinary oxalate:creatinine ratio; WT=body weight.
Source: Applicant's Population PK/PD Analysis Report Figure 26.

Dose Justification for PH1 Patients <6 Years of Age

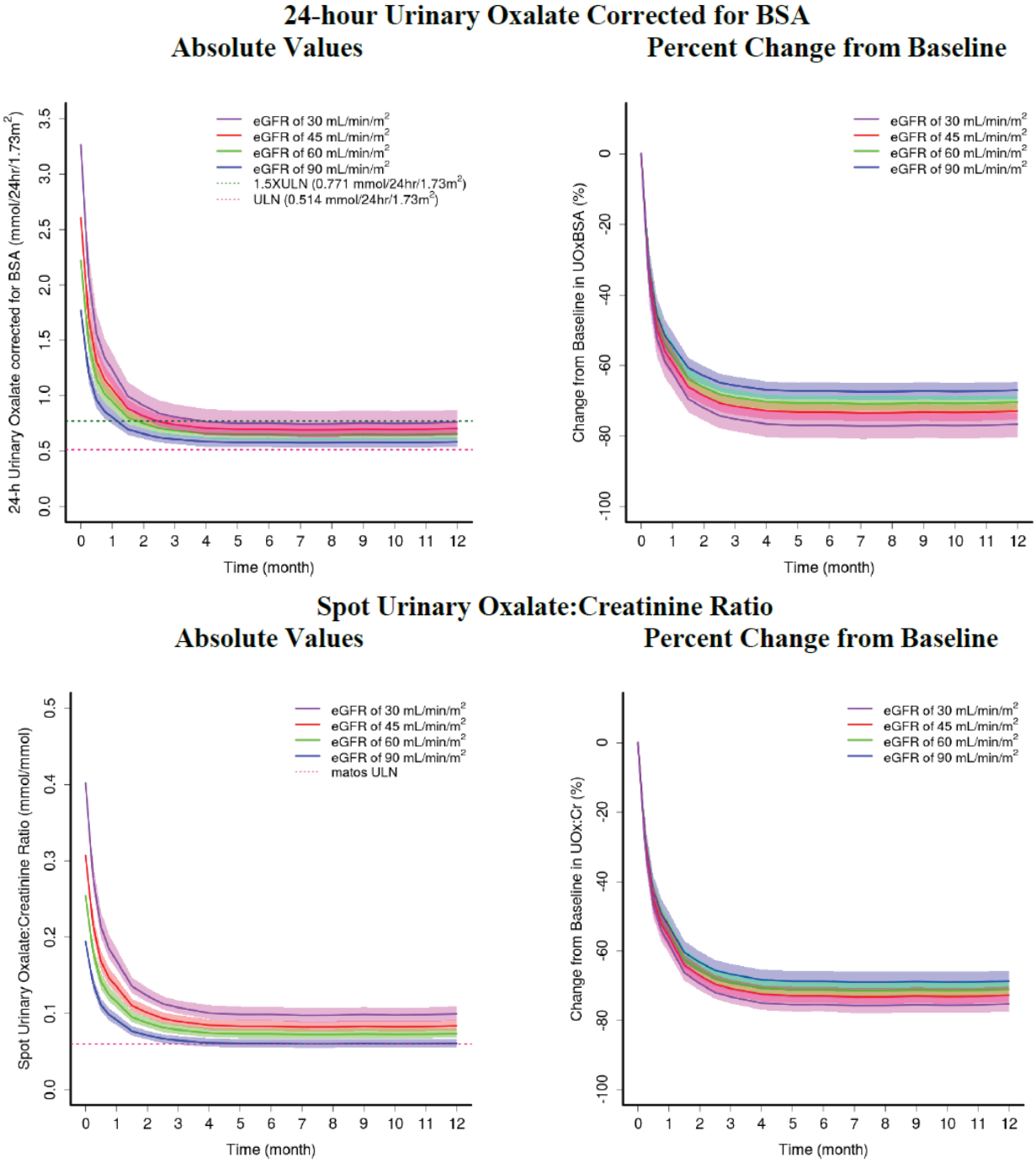
Impact of Baseline Oxalate Levels and Body Weight

Figure 4 shows the impact of the proposed lumasiran dosing regimens in different age groups: birth (4 kg), 6 months (8 kg), 1 year (10 kg), 2 years (13 kg) and 6 years of age (21 kg). In the absence of drug, Ox/Cr ratio is highest at birth and decreases with increasing age. Lumasiran, at the proposed dosing regimen, was estimated to yield an Ox/Cr ratio that approaches ULN by Month 3. Oxalate levels at Month 6 are expected to be similar to oxalate levels at Month 3, indicating that maximum lowering is achieved by Month 3 using a loading dose regimen and is subsequently maintained with the maintenance regimen. These results support the adequacy of lumasiran regimens for the different ages and body weight ranges in patients with PH1 <6 years of age.

Lumasiran Effect Across Various Levels of Renal Function

The impact of lumasiran on urinary oxalate levels across renal impairment categories is shown in Figure 25. At baseline, patients with lower eGFR had higher urinary oxalate levels, indicating greater disease severity. eGFR was not a significant covariate on I_{max} parameter. All patients were predicted to show similar response with lumasiran irrespective of their baseline renal function. Thus, patients with PH1 on lumasiran treatment are expected to approach ULN of respective urinary oxalate measures, regardless of their baseline renal function.

Figure 25. Model-Predicted Urinary Oxalate Lowering in Patients with PH1 Across Renal Function



Abbreviations: BSA=body surface area; eGFR=estimated glomerular filtration rate; ULN=upper limit of normal.
Note: Simulations shown above are for PH1 patients ≥ 6 years; matos ULN=95th percentile of spot urinary oxalate:creatinine ratio in healthy subjects with the respective age range [Matos 1999]
Source: Applicant's Population PK/PD Analysis Report Figure 28.

Reviewer's comments:

The Applicant's population PK/PD analyses: 1) predicted the human liver and RISC-loaded lumasiran PK based on rat and monkey liver PK data using the allometric scaling principles,

and 2) established the relationships between human RISC-loaded PK and urinary oxalate levels ($UOxBSA_{24h}$ and Ox/Cr ratio). The PK/PD analyses do appear to capture the central tendency of $UOxBSA_{24h}$ and Ox/Cr ratio time profiles, which make it reasonable for descriptive purposes. However, to some degree, the accuracy of the PK/PD models for the prediction of human PK in the liver remains uncertain, since the liver PK cannot be measured directly in humans. Ideally, the Applicant should have established the correlations among lumasiran plasma PK, liver PK and RISC-loaded PK in rats and monkeys at individual level so that the human plasma PK could be integrated in the population PK/PD models for the better prediction of human liver PK. However, lumasiran plasma PK, liver PK and RISC-loaded PK were collected from different rats, which makes it infeasible to establish the correlations.

Overall, despite some caveats, the Applicant's population PK/PD modeling and simulations is a reasonable approach to inform the proposed BW-based dosing regimens for general patient population and to support no dose adjustment for patients with renal impairment, as the pivotal trials generally confirmed the effectiveness of the proposed dosing regimens in patients with PH1 across different body weight groups.

14.2.3. Intrinsic Factors

14.2.3.1. Hepatic Impairment

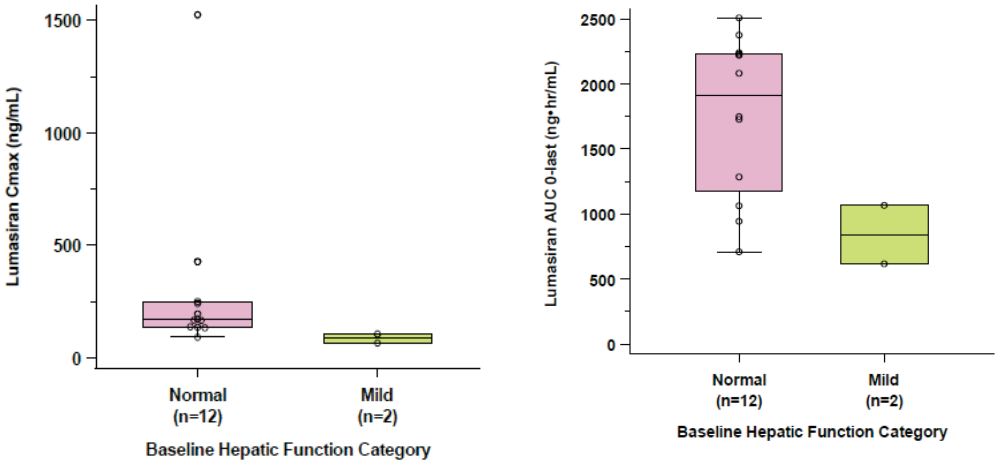
A dedicated hepatic impairment (HI) study was not conducted. The clinical experience of lumasiran in PH1 patients with hepatic impairment was limited. Across the studies enrolling 75 patients with PH1, only five patients had hepatic impairment, out of which only three patients from Study 001B (mild HI=2; moderate HI=1) had both lumasiran plasma PK and urinary oxalate reduction data reported. The impact of impaired hepatic function (National Cancer Institute organ dysfunction working group-NCI ODWG criteria) was determined by comparing lumasiran plasma PK and urinary oxalate reduction in patients with mild (BIL≤ULN and AST >ULN; or ULN<BIL≤1.5×ULN) or moderate HI (1.5 ULN <BIL ≤3×ULN) relative to patients with normal hepatic function (BIL≤ULN and AST≤ULN).

The median values of lumasiran C_{max} in plasma following phase 3 dosing regimen were 528 ng/mL, 339 ng/mL and 450 ng/mL for patients with normal hepatic function, mild and moderate HI, respectively. The median values for $AUC_{0\text{-}last}$ within the respective dosing regimens as well as the reduction in urinary oxalate, were also comparable between patients with mild, moderate HI and patients with normal hepatic function.

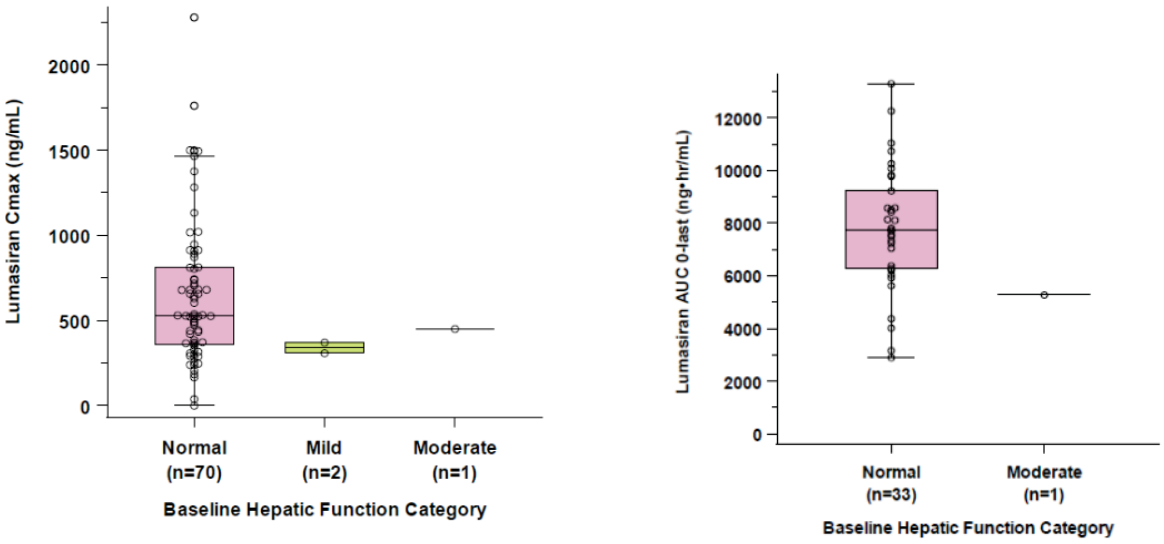
Overall, on comparison of data from PH1 patients with mild and moderate HI, values for the C_{max}, AUC_{last} or efficacy of lumasiran were within the range observed for subjects with normal hepatic function.

Figure 26. Comparison of C_{max} and AUC_{last} Across Studies by Hepatic Function Category in PH1 Patients Treated With Lumasiran

Study 001B
(1 mg/kg qM)

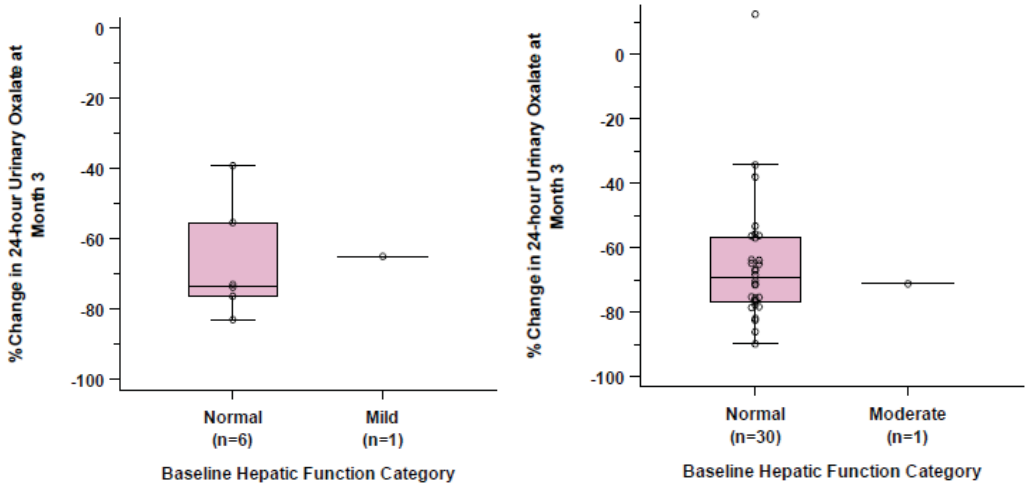


Studies 001B/003
(Phase 3 dosing regimen*)



* Due to sparse sampling in ILLUMINATE-A, AUC data are not available for all patients who received the Phase 3 dosing regimen; the C_{max} values in ILLUMINATE-A were lumasiran plasma concentrations at 4 hours post-dose.
Source: Applicant's summary of clinical pharmacology studies Figure 5 and Figure 13.

Figure 27. Comparison of Percent Change From Baseline in the 24-Hour Urinary Oxalate Corrected for BSA by Hepatic Function Category in PH1 Patients of Study 001B Treated With 1 mg/kg (Mild HI; n=1*) and 3 mg/kg (Moderate HI; n=1) of Lumasiran



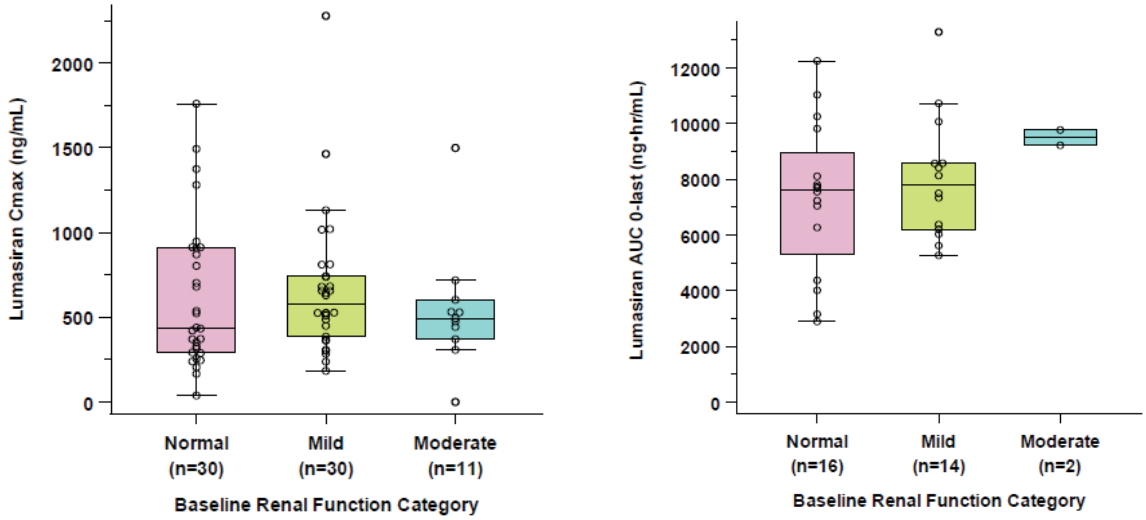
* The second patient with mild HI had % change in spot urinary oxalate assessment which was similar to the first patient with mild HI and other patients with normal hepatic function.
Source: Applicant's summary of clinical pharmacology studies Figure 68b and Figure 32b.

14.2.3.2. Renal Impairment

In general, patients with PH1 had lower median eGFR in comparison to healthy adults (PH1: 79 [51 to 131 mL/min/1.73 m^2]; healthy adults: 91 [65-117 mL/min/1.73 m^2], Study 001A). Approximately 7 to 14% of the administered lumasiran was excreted in urine in PH1 patients, which was lower than the 17 to 26% in healthy adults. A dedicated renal impairment (RI) study was not conducted. The impact of impaired renal function was determined by comparing the PK and efficacy of lumasiran in PH1 patients with mild (eGFR 60 to <90 mL/min/1.73 m^2) or moderate (eGFR 30 to <60 mL/min/1.73 m^2) RI at baseline to patients with normal renal function (eGFR ≥90 mL/min/1.73 m^2). In the pooled PH1 population (N=75; across Studies 001B and 003), 32, 32, and 11 patients treated with lumasiran, had normal renal function, mild, and moderate RI, respectively, at baseline. Patients with severe renal impairment were not enrolled in Studies 001, 002, 003 or 004.

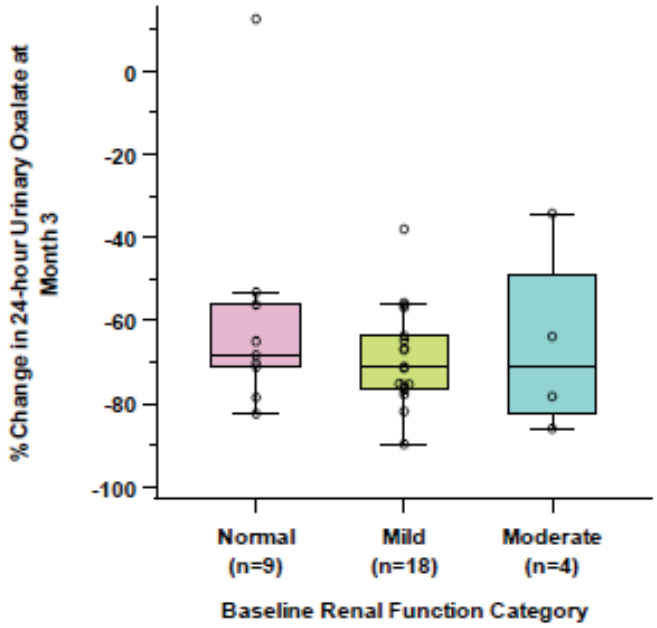
The plasma C_{max} of lumasiran was similar in mild or moderate renal impairment in comparison to patients with normal renal function; however, $AUC_{0\text{-last}}$ was 25% higher in patients with moderate renal impairment.

Figure 28. Comparison of C_{max} and AUC_{last}* Across Studies by Renal Function Category in PH1 Patients Treated With Lumasiran



* Due to sparse sampling in ILLUMINATE-A, $AUC_{0\text{-}last}$ data are not available for all patients; the C_{max} values in ILLUMINATE-A were lumasiran plasma concentrations at 4 hours postdose.
The percent reduction from baseline in 24-hour urinary oxalate corrected for BSA (and spot urinary oxalate-to-creatinine ratio) was similar in patients in the 3 renal function groups, showing a lack of effect of renal impairment on urinary oxalate reduction (efficacy), despite the higher trend in baseline values observed in patients with moderate RI.
Source: Applicant's summary of clinical pharmacology studies Figure 3 and Figure 11.

Figure 29. Comparison of Percent Change From Baseline in 24-Hour Urinary Oxalate Corrected for BSA Across Studies by Renal Function Category in PH1 Patients Treated With Lumasiran



Source: Applicant's summary of clinical pharmacology studies Figure 30b.

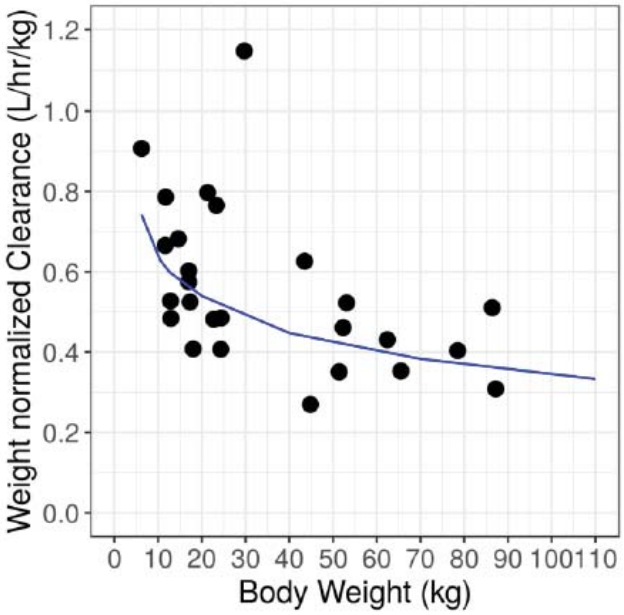
Based on the observed plasma PK and reduction in urinary oxalate data, no dose adjustment is required in renally impaired patients with an eGFR of ≥ 30 mL/min/1.73 m^2.

14.2.3.3. Body Weight

Across studies, the actual body weight of enrolled PH1 patients ranged between 6.2 to 110 kg. Although plasma PK is not the primary determinant of urinary oxalate reduction, plasma PK was a metric measured in the clinical studies to confirm systemic exposure and used for comparison. Body weight normalized plasma clearance was higher in patients <20 kg compared to ≥20 kg, consistent with allometric principles (larger liver to body weight ratio; 3.5 versus 2% of body weight in <10 kgs versus ≥20 kg), as shown in the figure below. This suggested that a higher dose (mg/kg) is required in patients weighing <20 kg in order to achieve similar exposures in patients ≥20 kg, thus supportive of the 6 mg/kg dose qM and q3M in body weight categories of <10 kg and 10 to 20 kg, respectively, as compared to 3 mg/kg in patients >20 kg.

The population PKPD modeling was further used to predict the steady-state liver exposure (RISC-loaded lumasiran concentrations) based on body weight-based dosing regimen. A consistent (>10-fold higher than IC_{50}) exposure predicted to result in near-maximal urinary oxalate lowering (approaching ULN) was estimated across the entire body weight range of PH1 patients, as shown in the table below.

Figure 30. Weight Normalized Plasma Drug Clearance for Lumasiran Versus Baseline Weight From Studies 001 and 004



Note: Black dots are non-compartmental analysis-derived clearance values from intensive sampling obtained in adult and pediatric PH1 patients in Studies 001 and 004, respectively. The blue line is model predicted plasma clearance for lumasiran.

Source: Applicant's Population PK Analysis Report Figure 16.

Table 61. Predicted Liver and RISC PK Exposures for Lumasiran in PH1 Patients After the Proposed Body Weight-Based Dosing Regimen

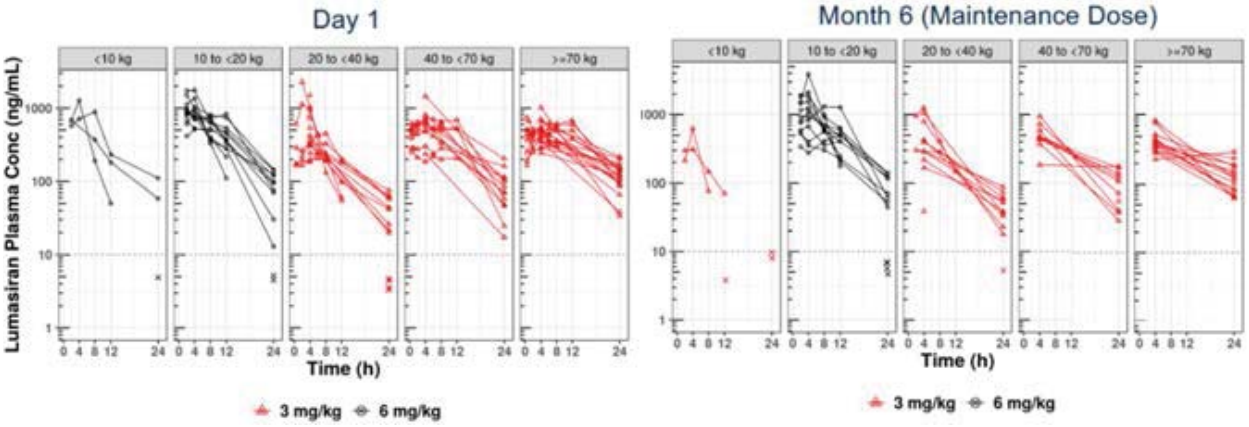
Compartment	PK Parameters	Body Weight				
		4 kg	10 kg	20 kg	40 kg	70 kg
Liver	$C_{max,ss}$ (µg/g)	70.7	65.9	39.7	48.7	55.7
	$C_{trough,ss}$ (µg/g)	52.6	48.1	26.6	34.9	41.7
	AUC_{ss} (µg/g*hr)	41618	38117	22067	27829	32525
RISC	$C_{max,ss}$ (ng/g)	6.15	8.13	6.16	6.52	6.61
	$C_{trough,ss}$ (ng/g)	5.98	7.99	5.68	6.35	6.58
	AUC_{ss} (µg/g*hr)	3.96	5.44	4.02	4.35	4.42

Abbreviations: AUC_{ss}=area under the concentration-time curve at steady state; $C_{max,ss}$=maximum concentration at steady state; $C_{trough,ss}$=minimum concentration at steady state; PK=pharmacokinetic; RISC=RNA-induced silencing complex.

Source: Applicant's Population PK/PD Modeling Report Table 17.

As shown in the figure, the pooled PK analysis across studies showed comparable PK profiles between the body weight categories with the lumasiran dosing regimen on Day 1 and Month 6.

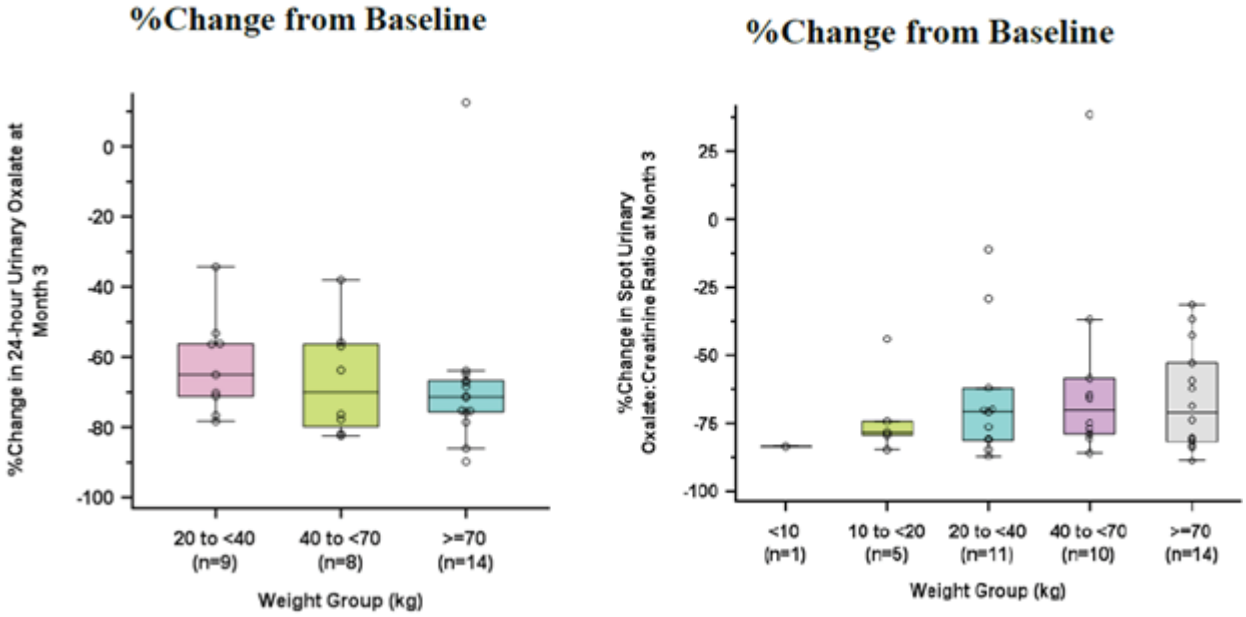
Figure 31. Lumasiran PK Profiles (Pooled) by Weight Category on Day1 and Month 6



Source: Applicant's ALN-GO1 Data Update Report 1 Study 004/Pooled PK/PD Analysis Figure 3.

Following the body weight-based dosing for lumasiran, the median percent reduction from baseline at month 3 ranged between 65 to 71% and 71 to 84%, for the 24-hour urinary oxalate (available only in patients ≥20 kg) and the spot oxalate-to-creatinine ratios (across all weight range), respectively.

Figure 32. Comparison of the Percent Change From Baseline in 24-Hour Urinary Oxalate Corrected for BSA and Spot Oxalate-to-Creatinine Ratio by Body Weight Groups in PH1 Patients Treated With Body Weight-Based Lumasiran Dosing



Source: Applicant's summary of clinical pharmacology studies Figure 33b and Figure 43b.

The body weight-adjusted dosing regimen resulted in similar plasma PK and percent reduction in urinary oxalate.

14.2.3.4. Other Intrinsic Factors

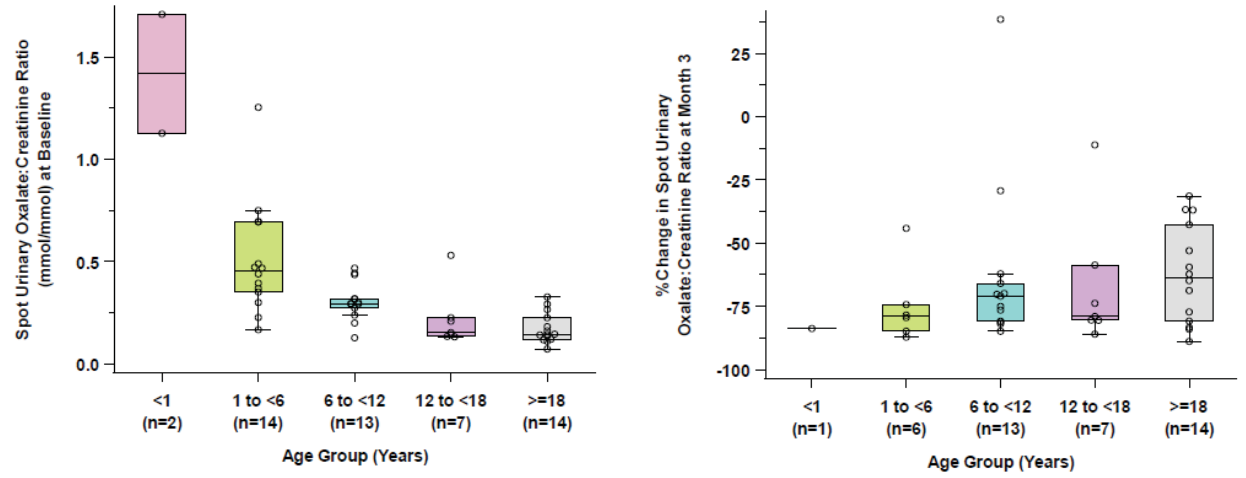
Age

The effect of age was evaluated by comparing lumasiran plasma PK and urinary oxalate reduction in patients in predefined age categories: <1 year, 1 to <6 years, 6 to <12 years, 12 to <18 years, and ≥18 years (overall age range across clinical studies was 4 months to 60 years). In patients <6 years of age, the Ox/Cr was measured as it was not feasible to obtain 24-hour urinary oxalate.

The median C_{max} was slightly higher in the <1 year and 1 to <6 year age groups as children weighing < 20 kgs received a nominally higher dose of lumasiran (6 mg/kg). Population PK analysis also indicated a faster absorption of lumasiran was likely in lower body weight patients. However, the median $AUC_{0\text{-last}}$ values were comparable across the age range.

The values for Ox/Cr (shown to be comparable to 24-hour urinary oxalate normalized to BSA in >6-year-old patient data) was used for the age group comparison below. The high baseline spot urine oxalate-to-creatinine ratio is attributed to the low urinary creatinine level in younger children, since children have lower muscle mass and reduced renal function relative to adolescents and adults. At Month 3, median percent reduction from baseline in Ox/Cr was generally similar across all age groups (range of median percent reduction from baseline: 64 to 84%).

Figure 33. Comparison of Baseline and Percent Change From Baseline in Spot Urinary Oxalate-to-Creatinine Ratio (mmol/mmol) Across Studies by Age Group in PH1 Patients Treated With Lumasiran



Source: Applicant's summary of clinical pharmacology studies Figure 41b1 and Figure 41b.

Results of the covariate model indicated that the effect of age on PK or PD of lumasiran was not significant with the body weight-based dosing regimen, indicating that the PK and efficacy of lumasiran are predicted to be comparable in infants, children, and adult PH1 patients.

Sex

The plasma $AUC_{0\text{-last}}$ and C_{max} were comparable between male and female PH1 patients. Median values for $AUC_{0\text{-last}}$ were 7620 and 7900 h.ng/mL, C_{max} were 508 and 643 ng/mL in male and female patients, respectively. At Month 3, median percent reduction from baseline in 24-hour urinary oxalate corrected for BSA was similar between male and female patients (69.8% and 70.2%, respectively.

Race

The majority of lumasiran study participants with PK data were White (n=56) followed by Asians (n=9), Other (n=7), and Black (n=1). As majority of the study participants were white, comparison was made between White and non-White participants. The median C_{max} and $AUC_{0\text{-last}}$ were similar in males across the different race categories. Modeling did not identify race as a significant covariate in the population PK/PD analysis.

Body Surface Area

The effect of BSA on the PK and PD parameters of lumasiran was evaluated in the population PK/PD analysis. As body weight and BSA are highly correlated, after accounting for body weight, BSA did not show any significant covariate effect in the model.

Conclusion

Based on the comparisons and covariate analyses on the data from PH1 patients, no additional dose adjustment is needed beyond the recommended body weight-based regimen.

14.2.4. Summary of Bioanalytical Method Validation and Performance

Pharmacokinetic Samples: Lumasiran in Plasma and Urine

Lumasiran is a siRNA duplex, composed of single antisense (A-131532) and sense (A-131522) strands. A validated liquid chromatographic time-of-flight mass spectrometry (LC-TOF-MS) assay was used to quantify the concentrations of lumasiran by detecting the antisense strand and sense strand portions of the duplex. Similarly, the internal standard (IS) (b) (4), was also composed of single antisense (b) (4)) and sense ((b) (4)) strands. The peak area ratios of the respective analyte-antisense/IS-antisense and analyte-sense/IS-sense were used to construct two separate standard curves and two distinct sets of data representing the duplex. Final concentrations of lumasiran were reported based on the antisense concentrations.

In brief, the human plasma samples (K_2EDTA) were spiked with IS, processed by solid phase extraction, and analyzed using reversed-phase UHPLC with Turbo Ion Spray TOF-MS detection. Accurate mass of ten ions for each strand of the analyte and IS were monitored in the negative ion mode. The peak area for the analyte or IS was the sum of the response from the respective ten ions. The peak area ratios of the respective strands of analyte/IS were used to build the calibration curve as well as to quantify the QCs and unknown samples.

Table 62. Parameters of Method Validation for Analysis of Lumasiran in Human Plasma

Report Title	Validation of a Method for the Determination of ALN-GO1 in Human Plasma by LC-TOF-MS
Study Number	(b) (4) 319-1505
Analyte Name	ALN-GO1
Internal Standard (IS)	(b) (4)
Analytical Method Type	LC-TOF-MS
Extraction Method	Solid-phase
Sample Volume	100 µL
QC Concentrations	10, 30, 400, 4000, and 8000 ng/mL
Standard Curve Concentrations	10, 20, 100, 300, 1000, 3000, 9000, and 10000 ng/mL
Lower Limit Of Quantitation	10 ng/mL
Upper Limit Of Quantitation	10000 ng/mL
Average Recovery of Analyte (%)	95.2 (by antisense A-131532)
	92.3 (by sense A-131522)
Average Recovery of Internal Standard (%)	94.2 (by antisense (b) (4))
	90.5 (by sense (b) (4))
LLOQ QC Intraday Precision Range (%CV)	5.7 to 6.7 (by antisense A-131532)
	4.2 to 10.5 (by sense A-131522)
LLOQ QC Intraday Accuracy Range (%RE)	5.1 to 14.4 (by antisense A-131532)
	12.4 to 18.1 (by sense A-131522)
Analytical QC Intraday Precision Range (%CV)	1.3 to 5.6 (by antisense A-131532)
	1.9 to 9.9 (by sense A-131522)
Analytical QC Intraday Accuracy Range (%RE)	-9.2 to 14.4 (by antisense A-131532)
	-11.2 to 13.5 (by sense A-131522)
LLOQ QC Interday Precision (%CV)	7.0 (by antisense A-131532)
	8.3 (by sense A-131522)
LLOQ QC Interday Accuracy (%RE)	9.1 (by antisense A-131532)
	14.5 (by sense A-131522)
Analytical QC Interday Precision Range (%CV)	3.3 to 4.7 (by antisense A-131532)
	4.1 to 11.2 (by sense A-131522)
Analytical QC Interday Accuracy Range (%RE)	-6.3 to 11.2 (by antisense A-131532)
	-7.5 to 11.1 (by sense A-131522)
Stock Solution Stability in Water	76 Days at -20°C[a] 16 Hours at Ambient Temperature[a]
Processed Sample Stability	120 Hours at Ambient Temperature
Benchtop Stability in Plasma	24 Hours at Ambient Temperature
Freeze/Thaw Stability in Plasma	5 Cycles at -20°C and -70°C
Benchtop Stability in Whole Blood	2 Hours at 4°C When Centrifuged at 4°C 2 Hours at 4°C When Centrifuged at Ambient Temperature 2 Hours at Ambient Temperature When Centrifuged at Ambient Temperature
Long-term Storage Stability in Plasma	363 Days at -20°C and -70°C
Dilution Integrity	100000 ng/mL diluted 200-fold
Blank Selectivity	≤20.0% LLOQ for analyte; ≤5.0% for IS
2% Hemolyzed Plasma Test	No impact on assay performance
Lipemic Plasma Test	No impact on assay performance

Source: Applicant's Bioanalytical method validation report (b) (4) Study Number: 319-1505 (Validation summary page 13).

For analysis of lumasiran in urine (Studies 001, 002 only), the pooled human urine sample treated with 0.1% CHAPS (3-[(3-cholamidopropyl) dimethylammonio]-1-propanesulfonate) was used for further processing similar to the plasma samples. Pooled human urine treated with 0.1% CHAPS was used as the blank matrix for validation.

Table 63. Parameters of Method Validation for Analysis of Lumasiran in Human Urine

Report Title	Validation of a Method for the Determination of ALN-GO1 in Human Urine by LC-TOF-MS
Study Number	(b) (4) 319-1508
Analyte Name	ALN-GO1
Internal Standard (IS)	(b) (4)
Analytical Method Type	LC-TOF-MS
Extraction Method	Solid-phase
Sample Volume	100 µL
QC Concentrations	10, 30, 400, 4000, and 8000 ng/mL
Standard Curve Concentrations	10, 20, 100, 300, 1000, 3000, 9000, and 10000 ng/mL
Lower Limit Of Quantitation	10 ng/mL
Upper Limit Of Quantitation	10000 ng/mL
Average Recovery of Analyte (%)	92.1 (by antisense A-131532)
	91.7 (by sense A-131522)
Average Recovery of Internal Standard (%)	95.9 (by antisense (b) (4)
	91.1 (by sense (b) (4)
LLOQ QC Intraday Precision Range (%CV)	4.5 to 10.0 (by antisense A-131532)
	4.2 to 7.7 (by sense A-131522)
LLOQ QC Intraday Accuracy Range (%RE)	0.2 to 2.6 (by antisense A-131532)
	8.0 to 15.9 (by sense A-131522)
Analytical QC Intraday Precision Range (%CV)	2.2 to 5.7 (by antisense A-131532)
	1.3 to 9.9 (by sense A-131522)
Analytical QC Intraday Accuracy Range (%RE)	-8.0 to 11.1 (by antisense A-131532)
	-9.5 to 12.7 (by sense A-131522)
LLOQ QC Interday Precision (%CV)	8.0 (by antisense A-131532)
	6.4 (by sense A-131522)
LLOQ QC Interday Accuracy (%RE)	1.1 (by antisense A-131532)
	11.7 (by sense A-131522)
Analytical QC Interday Precision Range (%CV)	3.1 to 5.8 (by antisense A-131532)
	3.6 to 7.5 (by sense A-131522)
Analytical QC Interday Accuracy Range (%RE)	-6.9 to 9.3 (by antisense A-131532)
	-6.5 to 10.3 (by sense A-131522)
Stock Solution Stability in Water	76 Days at -20°C[a] 16 Hours at Ambient Temperature[a]
Processed Sample Stability	150 Hours at Ambient Temperature
Benchtop Stability in Urine Treated with 0.1% CHAPS or 1.0% CHAPS	24 Hours at Ambient Temperature
Benchtop Stability in Urine without CHAPS	24 Hours at Ambient Temperature
Freeze/Thaw Stability in Urine Treated with 0.1% CHAPS or 1.0% CHAPS	6 Cycles at -20°C and -70°C
Freeze/Thaw Stability in Urine without CHAPS	5 Cycles at -20°C and -70°C
Long-term Storage Stability in Urine Treated with 0.1% CHAPS or 1.0% CHAPS	367 Days at -20°C and -70°C
Long-term Storage Stability in Urine without CHAPS	365 Days at -20°C and -70°C
Dilution Integrity	50000 ng/mL diluted 100-fold
Blank Selectivity	≤ 20.0% LLOQ for analyte; ≤ 5.0% for IS
Assay Equivalency	Human urine samples treated with 1.0% CHAPS are comparable to human urine samples treated with 0.1% CHAPS Human urine samples without CHAPS are comparable to human urine samples treated with 0.1% CHAPS

Source: Applicant's Bioanalytical method validation report (b) (4) Study Number: 319-1508 (Validation summary page 15).

The concentrations of calibration standards and QCs for the single sense (A-131522) and antisense (A-131532) strands of lumasiran were qualified to be within the acceptable limits during analysis of plasma and urine samples across the applicable clinical studies.

Biomarker Samples: Urinary Oxalate

Table 64. Parameters of Method Validation for Analysis of Total Oxalate in Human Urine

Report Title	Validation of a method for the determination of total oxalate in human urine by LC-MS/MS
Study Number	(b) (4)319-1816
Analyte Name	Oxalate
Internal Standard (IS)	Oxalate-$^{13}C_2$
Analytical Method Type	LC-MS/MS
Extraction Method	None; samples were diluted 48-fold
Sample Volume	50 µL
QC Concentrations	5 (LLOQ QC) and 15 (LQC) µg/mL in water; endogenous (QC0), endogenous + 30 (LMQC), endogenous + 100 (HMQC), and endogenous + 200 (HQC) µg/mL
Standard Curve Concentrations	5, 10, 20, 40, 80, 120, 225, and 250 µg/mL
Lower Limit Of Quantitation	5 µg/mL
Upper Limit Of Quantitation	250 µg/mL
Variation of Matrix Effect from Six Lots of Matrix (%CV)	≤ 7.4%
LLOQ QC Intraday Precision Range (%CV)	2.8 to 4.3
LLOQ QC Intraday Accuracy Range (%RE)	-7.2 to -2.6
Analytical QC Intraday Precision Range (%CV)	1.3 to 4.3
Analytical QC Intraday Accuracy Range (%RE)	-11.0 to 2.1
LLOQ QC Interday Precision (%CV)	3.8
LLOQ QC Interday Accuracy (%RE)	-5.4
Analytical QC Interday Precision Range (%CV)	3.0 to 4.8
Analytical QC Interday Accuracy Range (%RE)	-7.3 to 0.0
Stock Solution Stability in Water	68 days at -20°C 19 hours at ambient temperature
Processed Sample Stability	126 Hours at 4°C
Benchtop Stability in Human Urine	At least 24 hours at ambient temperature At least 168.5 hours at 4°C
Freeze/Thaw Stability in Human Urine	5 cycles at -20°C 9 cycles at -70°C
Long-term Storage Stability in Human Urine	560 days at -20°C/-70°C
Dilution Integrity	Endogenous + 500 µg/mL diluted 20-fold with water
BQL dilution	6-fold lesser dilution during sample processing
Spike-in Selectivity	Meets acceptance criteria
Interference Test for Ascorbic Acid, Glycolate, and Citrate at 1/1/1 mg/mL	No interference from tested compounds to total oxalate
Parallelism (water and human urine)	Meets acceptance criteria
Batch Size Test	192 injections
Carryover	<20% LLOQ (oxalate) <5% Carryover IS (oxalate-$^{13}C_2$)

Source: Applicant's Bioanalytical method validation report (b) (4) Study Number: 319-1816 (Validation summary page 16).

The concentration of total oxalate in human urine (24 hour and spot) samples were determined using a validated LC-MS/MS assay as a marker for screening and efficacy in PH1 patients. Due

to the presence of endogenous urinary oxalate, validation study assessed urine matrix and surrogate matrix (water) to demonstrate suitability to use surrogate matrix for clinical urine sample analysis. Surrogate or authentic matrix samples were spiked with IS (oxalate-$^{13}C_2$), processed by simple dilution, and analyzed using anion exchange HPLC with Turbo Ion Spray® MS/MS detection using negative ion mode. The LC-MS/MS method was also used to retrospectively analyze urine samples from Phase 1/2 trials, Study 001/002, that were initially analyzed using a different colorimetric assay. In addition, to analyze samples from pediatric patients who have higher fluid intake, a 6-fold dilution step was omitted in order to re-assay sample at a more concentrated level.

Concentrations of oxalate in human urine samples were considered acceptable as the calibration and QC standards met the acceptability criterion across all the studies.

Creatinine:

Validation was done for the use of a commercial assay kit for the colorimetric determination of creatinine in human urine from 0.100 to 2.000mM (used to normalize oxalate levels in spot samples).

Table 65. Parameters of Method Validation for Analysis of Creatinine in Human Urine

Report Title	Determination of Creatinine in Human Urine by Colorimetric Assay
Report Number	(b) (4) 42-0508
Analyte Name and synonym	Creatinine
Sample Volume:	50 µL
Analytical Method Type	Colorimetric
Sample Processing Method	None
Calibration Range:	0.100 – 2.00 mmol/L
LLOQ QC	2.00 mmol/L (0.100 mmol/L at 20X dilution)
ULOQ QC	40.0 mmol/L (2.00 mmol/L at 20X dilution)
Matrix QC Concentrations	6.00 mmol/L (0.3 mmol/L at 20X dilution) 21.0 mmol/L (1.05 mmol/L at 20X dilution) 30.0 mmol/L (1.5 mmol/L at 20X dilution)
QC Intra-batch Precision (%CV)	1.2% to 2.8%
QC Intra-batch Accuracy (%RE)	-2.9% to 1.7%
QC Inter-batch Precision (%CV)	2.1% to 6.7%
QC Inter-batch Accuracy (%RE)	-1.5% to 1.2%
Benchtop Stability in Human Urine	8 Hours at Room Temperature
Freeze/thaw Stability in Human Urine	6 Cycles at -70°C
Long-term Storage Stability in Human Urine	At least [a]311 days stability at -70°C
Parallelism (5X, 10X, 20X, 40X, and 60X) 1X dilution	5.1 mmol/L diluted 1X, 5X, and 20X 24.5 mmol/L diluted 20X, 40X, and 60X Acceptable for 5X to 60X dilution [b]1X dilution validated for hyper-hydrated urine that tested BQL at 5X dilution.
Selectivity (6 lots at 2.00 and 21.0 mmol/L)	100% lots tested were within 100±25% Recovery (2 lots were BQL at 2.00 mmol/L)
Interference of indomethacin at 6 mmol/L and 30 mmol/L creatinine in Human Urine	Creatinine was within 100±20% at 10 µg/mL and 20 µg/mL indomethacin concentration

Source: Applicant's Bioanalytical method validation report (b) (4) Study Number: 42-0508.

Conclusion:

The methods for quantifying lumasiran levels in the human plasma/urine and oxalate, creatinine in the human urine satisfied the method validation criterion in accordance with the FDA guidance and showed an acceptable performance during the sample analysis, as applicable, across the clinical studies 001, 002, 003 and 004.

14.2.5. Immunogenicity

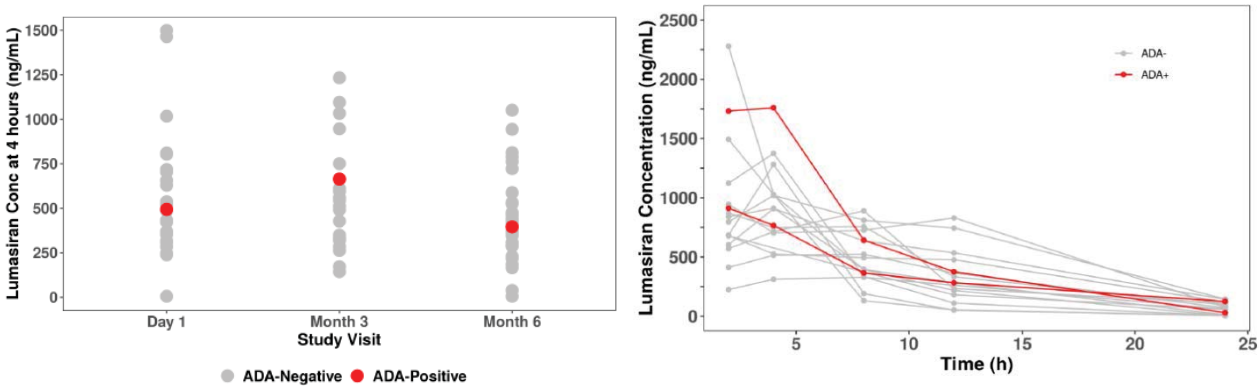
Immunogenicity was evaluated by detecting serum immunoglobulin G/IgM ADA against lumasiran using a validated ELISA. The assay detects ADA against the lumasiran drug substance, the double-stranded siRNA including the N-acetylgalactosamine (GalNAc) moiety and the linker. ADA titer was determined only in samples confirmed ADA-positive in the

screening assay. A neutralizing assay for ADA was not developed but comparison of the urinary (and plasma) oxalate levels between the ADA-positive and ADA-negative PH1 patients and evaluation of the plasma glycolate levels in healthy subjects was used to assess the impact of the ADA for neutralizing activity.

Across all clinical studies in the lumasiran development program, including patients with PH1 and healthy volunteers, 6 of 100 (6%) lumasiran-treated individuals tested positive for ADA, as early as from Day 29.

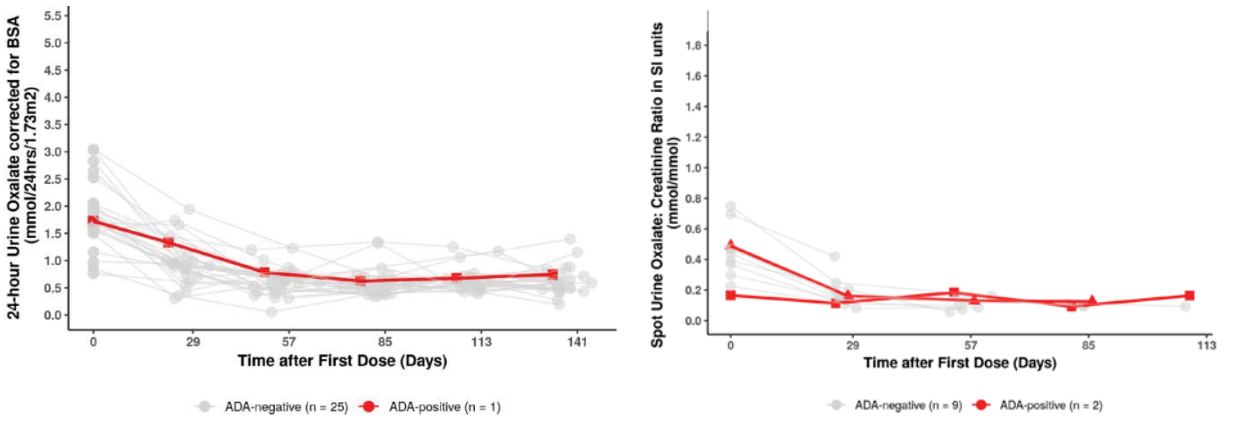
In Study 001B/002, three patients were reported positive for ADA, one on Day 57 and the others on Month 6 and Month 6/Day 225, but subsequently testing negative for ADA at all other time points. In the pivotal study 003, a single patient tested positive at Month 6. In Study 004, two patients tested positive for ADA, one at Day 29 and the other at Month 3.

Figure 34. Comparison of Lumasiran Concentrations Between ADA-Positive and ADA-Negative Patients (ILLUMINATE A, Left Panel/ ILLUMINATE-B, Right Panel)



*ADA was first identified on Month 6. The C_4 lumasiran concentrations for ADA-positive patient is highlighted even at prior visits
Source: Applicant's summary of clinical pharmacology studies Figures, in page 21 and 23.

Figure 35. Comparison of 24-Hour Urine Oxalate Corrected for BSA (ILLUMINATE-A) or Spot Urine Oxalate-to-Creatinine Ratio (ILLUMINATE-B) Between ADA-Positive and ADA-Negative Patients



Source: Applicant's summary of clinical pharmacology studies, Figures in page 316 and 317.

Conclusion:

No clinically significant differences in the safety, pharmacokinetic, or PD profiles of lumasiran were observed in patients who tested positive for anti-lumasiran antibody.

14.2.6. Pharmacokinetic Assessments of Lumasiran

Study 001-Part A-Single Dose Study in Healthy Volunteers

The Part A of Study 001 was a randomized, placebo-controlled, single-blind, single ascending dose study to evaluate the safety, tolerability, PK, PD (plasma glycolate) and ADA of lumasiran in 18-64-year-old healthy volunteers. Participants (n=32) were assigned to receive a single dose of 0.3, 1, 3 or 6 mg/kg of lumasiran by subcutaneous injection (active: placebo:3:1). Plasma samples were obtained at pre-dose and at 30 min, and 1, 2, 4, 6, 8 h on Day 1 and 24 h on Day 2. Pooled 24-hour urine samples were also obtained on Day 1.

Table 66. PK Parameters (Mean, %CV) After a Single SC Dose of Lumasiran in Healthy Volunteers (Study 001, Part A)

	Lumasiran			
PK Parameter	**0.3 mg/kg (n=6)**	**1 mg/kg (n=6)**	**3 mg/kg (n=6)**	**6 mg/kg (n=6)**
t_{max} (h)[a]	5.02 (4.00, 8.02)	1.50 (0.52, 8.00)	3.00 (0.50, 8.00)	7.00 (0.50, 8.07)
C_{max} (ng/mL)	39.8 (21.6)	204 (54.6)	534 (30.0)	1180 (17.0)
$AUC_{0\text{-}last}$ (h•ng/mL)	294 (33.0)	1900 (29.4)	7210 (15.6)	16800 (26.1)
$t_{½}$ (h)[b]	NC	7.07 (5.29)	5.98 (25.5)	3.47 (NC)
V_Z/F (L)[b]	NC	389 (28.3)	219 (35.7)	139 (NC)
CL/F (L/h)[b]	NC	37.9 (23.2)	25.0 (10.6)	27.8 (NC)
$Fe_{0\text{-}24}$ (%)	17.4 (14.0)	19.1 (20.4)	21.1 (25.5)	25.8 (12.6)
CL_R (L/h)	8.78 (NC)	5.49 (37.8)	5.82 (22.6)	6.34 (18.2)

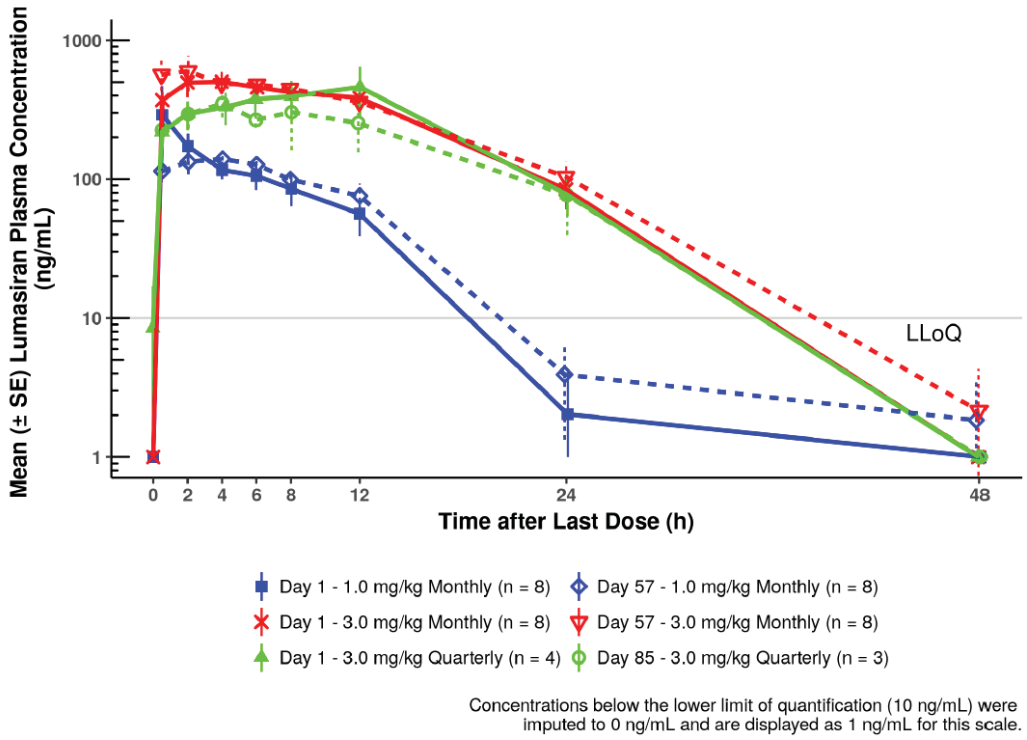
Abbreviations: $AUC_{0\text{-}last}$=area under the plasma concentration versus time curve from 0 to the last measurable concentration; CL/F=apparent total clearance of the drug from plasma; CL_R=renal clearance; C_{max}=maximum plasma concentration; %CV=percent coefficient of variation; $Fe_{0\text{-}24}$=fraction excreted in urine from time 0 to 24 hours; NC=not calculated; PK=pharmacokinetics; SC=subcutaneous; $t_{½}$=elimination half-life; t_{max}=time to reach maximum plasma concentration; V_Z/F=terminal phase extravascular volume of distribution
a. t_{max} is presented as median (min, max).
b. Only estimated for healthy volunteers with a defined elimination phase.

Source: Applicant's Clinical Study Report ALN-GO1-001 Table 21.

Study 001-Part B- Multiple Dose Study in PH1 Patients >6 Years Old

Part B of Study 001 was a randomized placebo-controlled (3:1), double-blind, MAD study to evaluate the safety, tolerability, PK, PD and ADA of lumasiran in 6-64-year-old PH1 patients. Participants (n=12) were assigned to receive a 1 mg/kg once monthly (qM) x 3 doses, 3 mg/kg qM x 3 doses, or 3 mg/kg q3M x 2 doses of lumasiran by subcutaneous injection. Patients who were initially randomized to placebo crossed over to lumasiran on Day 85 (n=3). Two additional expansion cohorts (n=8) were added (1 mg/kg qM, 3 mg/kg qM) as open-label. Intensive plasma PK samples were obtained on Days 1 and 57. Pooled 24-hour urine samples were obtained on Days 1, 57, 85 and 141.

Figure 36. Plasma Pharmacokinetic Profiles (Mean, SE) by Cohort and Day (Semi-Logarithm Scale) After SC Doses of Lumasiran in PH1 Patients (Study 001, Part B)



Source: Applicant's summary of clinical pharmacology studies, Figure in page 304.

Table 67. PK Parameters (Mean, %CV) After Multiple SC Doses of Lumasiran in PH1 Patients (Study 001, Part B)

	1.0 mg/kg qM		3.0 mg/kg qM		3.0 mg/kg q3M	
PK Parameter	Day 1 (n=8)	Day 57 (n=8)	Day 1 (n=8)	Day 57 (n=8)	Day 1 (n=4)	Day 85 (n=4)
t_{max} (h)[a]	3.99 (0.57, 5.97)	3.04 (0.50, 6.00)	4.99 (0.53, 12.00)	2.98 (0.53, 12.00)	9.00 (5.78, 12.02)	5.98 (4.05, 7.95)
C_{max} (ng/mL)	324 (151)	148 (46.0)	582 (45.8)	701 (73.0)	432 (56.7)	412 (42.5)
$AUC_{0\text{-last}}$ (h•ng/mL)	1430 (48.9)	1610 (44.1)	7400 (31.5)	7960 (21.7)	6340 (60.6)	5140 (53.7)
$t_{½}$ (h)[b]	3.27 (46.8)	7.81 (57.9)	5.46 (64.0)	5.84 (53.5)	7.80 (NC)	4.67 (NC)
V_Z/F (L)[b]	116 (76.6)	195 (70.5)	177 (93.9)	187 (79.6)	201 (NC)	165 (NC)
CL/F (L/h)[b]	21.6 (45.3)	18.1 (56.1)	19.1 (41.0)	18.9 (50.5)	17.8 (NC)	24.5 (NC)
$Fe_{0\text{-}24}$ (%)	11.1 (33.7)	9.47 (44.6)	11.2 (54.3)	12.5 (32.3)	7.17 (33.1)	13.7 (26.3)
CL_R (L/h)	2.26 (52.0)	1.96 (56.7)	2.38 (47.5)	2.52 (32.0)	2.06 (58.6)	3.37 (35.2)

Abbreviations: $AUC_{0\text{-last}}$=area under the plasma concentration versus time curve from 0 to the last measurable concentration; CL/F=apparent total clearance of the drug from plasma; CL_R=renal clearance; C_{max}=maximum plasma concentration; CV=coefficient of variation; $Fe_{0\text{-}24}$=fraction excreted in urine from time 0 to 24 hours; max=maximum; min=minimum; NC=not calculated; PK=pharmacokinetics; $t_{½}$=elimination half-life; t_{max}=time to reach maximum plasma concentration; $t_{½}$=elimination half-life; V_Z/F=terminal phase extravascular volume of distribution.

a. t_{max} is presented as median (min, max).

b. Only estimated for patients with a defined elimination phase.

Source: Applicant's Clinical Study Report ALN-GO1-001 Table 22.

The increase in lumasiran plasma exposures were approximately dose proportional over the dose range of 0.3 to 6 mg/kg in healthy volunteers and PH1 patients.

Study 004- Multiple Dose PK/PD Study in PH1 Patients <6 Years Old

The Phase 3 study was an open-label, single-arm study to evaluate the efficacy, safety, PK and ADA of lumasiran in infants and young children <6 years of age with PH1. The weight-based dosing regimen was administered by SC route. The study included a 6-month primary analysis period followed by a long-term extension period of up to 54 months. The Day 1 loading dose was 6 mg/kg in patients <20 kg and 3 mg/kg in patients ≥20 kg.

Table 68. PK Parameters (Mean, %CV) After the First SC Dose of Lumasiran in PH1 Patients <6 Years Old (Study 004)

PK Parameter	Lumasiran Dose Group by Initial Body Weight		
	<10 kg (n=3)	**10 to <20 kg (n=11)**	**≥20 kg (n=2)**
t_{max} (h)[a]	4.22 (2, 8.1)	3.68 (1.93, 7.83)	1.97, 7.1
C_{max} (ng/mL)	950 (32.2)	1040 (35.1)	4370, 7340
$AUC_{0\text{-}last}$ (h•ng/mL)[a]	6270 (5920, 8510)	8110 (7050, 13300)	4370, 7340
$t_{½}$ (h)[b]	5.46	5.22 (47.5)	1.42
V_Z/F (L)[b]	44.3	66.2 (60.0)	20.2
CL/F (L/h)[b]	5.63	8.48 (17.5)	9.89
CL/F/WT (L/h/kg)[b]	0.908	0.584 (19.6)	0.407

Abbreviations: $AUC_{0\text{-}last}$=area under the plasma concentration versus time curve from 0 to the last measurable concentration; CL/F=apparent total clearance of the drug from plasma; CL/F/WT=weight normalized CL/F, calculated as CL/F divided by the weight at the given visit; C_{max}=maximum plasma concentration; PK=pharmacokinetics; $t_{½}$=elimination half-life; t_{max}=time to reach maximum plasma concentration; V_Z/F=terminal phase extravascular volume of distribution

[a] Presented as median (min, max)

[b] n=1 for <10 kg and ≥20 kg dose groups

Source: Applicant's Clinical Study Report ALN-GO1-004 Table 10 and Table 14.2.2.9.

Following Day 1 of the body weight-adjusted dosing regimen, the PK parameters were similar across the different body weight groups.

15. Trial Design: Additional Information and Assessment

ILLUMINATE-A

Important Trial Dates

The study was initiated on December 13, 2018. The last patient visit and the data cutoff for the 6-month placebo-controlled period was November 6, 2019. The database was locked and unblinded for the interim analysis on December 9, 2019.

Trial Administrative Structure

Data Monitoring Committee (DMC)

An independent DMC monitored study progress and safety data on a regular basis and met at least quarterly. DMC membership and responsibilities were defined by a written charter. The DMC chair could, via an Alnylam DMC liaison who could attend open but not closed session meetings, recommend to the Applicant changes to the trial or trial conduct to address any concerns regarding study conduct or subject safety.

Open sessions included DMC members, the Independent Statistician, other members of the Independent Statistical Data Analysis Center (ISDAC), and project team members from Alnylam. Closed sessions included only individuals from the DMC, the Independent Statistician, and other members of the ISDAC. Data for the closed sessions were presented in a semi-blinded manner for ILLUMINATE-A (Group A and B), but the DMC could request and receive the identities of the groups. The information covered in the closed sessions were briefly described in the closed session minutes.

The Applicant submitted the meeting minutes for the open and closed sessions. Review of the minutes did not reveal any significant concerns about study conduct, efficacy, or safety.

Independent Statistical Data Analysis Center (ISDAC)

An independent biostatistics group performed analyses and generated reports for the DMC.

Study Assessments

After the baseline visit (Day 1), patients were to return for study visits at Days 15, 29 and every 28 days thereafter during the 6-month placebo-controlled treatment period. A full physical examination was performed at baseline and Month 6, and a symptom-directed physical examination was conducted at every visit. Vital signs and body weight were obtained at every visit; height was obtained at screening, baseline, and Month 6.

Clinical laboratory assessments were obtained at every visit. These included serum chemistry parameters (sodium, potassium, chloride, bicarbonate, blood urea nitrogen, creatinine, albumin, calcium, phosphate, glucose, uric acid, and total protein), liver tests (aspartate aminotransferase, alanine aminotransferase, total and direct bilirubin, and alkaline phosphatase), dipstick urinalysis

(pH, specific gravity, ketones, albumin, protein, glucose, bilirubin, nitrite, heme, urobilinogen and leucocytes), complete blood count with differential, and coagulation parameters (prothrombin time, internationalized normalized ratio and partial thromboplastin time). Urine microscopy was performed if clinically indicated. Pyridoxine levels (six hours or longer after the last pyridoxine dose) for patients on pyridoxine were obtained at screening, baseline, and monthly.

A spot urine sample, preferably first morning, for oxalate-to-creatinine ratio was obtained at every visit. A blood sample for ADA was obtained at baseline and at Months 1, 3 and 6.

Measurement of Urinary Oxalate

Urinary oxalate excretion was determined from 24-hour urine collections completed at screening, and monthly from Months 1 to 6. A urine collection was considered valid if each of the following criteria were met:

- The duration of the collection was between 22-26 hours between the initial discarded void and the last void or attempt to void.
- No voids were missed between the start and end time of the collection as indicated by the patient's urine collection diary.
- The 24-hour urine creatinine content was at least 10 mg/kg as assessed by the central laboratory.

At screening and Month 6, a minimum of two of three separate urine collections had to be valid, and all Month 6 collections had to be within 14 days before the Month 6 dose of study drug. At all other timepoints, a single 24-hour urine collection was obtained within seven days before dosing.

The Applicant trained study staff, and sites instructed patients on urine collection procedures and the use of urine diaries. De-identified urine diaries were reviewed for correct collection by the Applicant. The duration of collection and urine volume was recorded in the eCRF. Urinary creatinine was measured in an aliquot of the 24-hour urine collection. Invalid collections were repeated and had to be supervised by a healthcare professional during an inpatient visit or with close contact with study staff in an outpatient setting. Patients were to avoid high dose vitamin C preparations within 4 days before oxalate assessments.

Urinary oxalate was analyzed by a central laboratory. The first five patients were screened for eligibility and stratified by mean 24-hour urinary oxalate levels (≤1.70 or >1.70 mmol/24 hr/1.73 m^2) using a clinical assay performed at the (b) (4). Thereafter, a validated assay ((b) (4)) was used.

Key Equations

BSA was calculated using the Mosteller formula: BSA=square root (mean height (cm)*weight (kg)/3600).

eGFR (mL/min/1.73 m^2) was calculated from serum creatinine (SCr) as follows:

- Modification of Diet in Renal Disease (MDRD) equation in patients ≥18 years of age: eGFR = 175 × (SCr [mg/dL])$^{-1.154}$ × (age)$^{-0.203}$ × (0.742, if female), or × (1.212, if African American)
- Schwartz Bedside Formula for patients <18 years of age: eGFR = (0.413 × height [cm])/SCr (mg/dL)

Study Procedures

Randomization

Randomization was managed centrally using an interactive response system. The randomization codes were generated by an independent organization (Medpace) and were not accessible to blinded study personnel (unless required during a medical emergency) during the study period.

Patients were stratified by mean 24-hour urinary oxalate levels (≤1.70 or >1.70 mmol/24 hr/1.73 m^2) calculated using the values obtained from the first two valid baseline 24-hour urine collections.

Blinding

Patients received either lumasiran or matching placebo (preservative-free 0.9% saline solution). Both were provided in 2-mL glass vials containing 0.5 mL of a clear, colorless to pale yellow, sterile solution. The Applicant provided samples of lumasiran and placebo as was provided to investigational sites for ILLUMINATE-A for Agency review. The lumasiran and placebo samples were identical in appearance.

Dosing

Dosing Regimen

The dosing regimen was 3 mg/kg administered by subcutaneous injection (abdomen, upper arms or thighs) once monthly for three consecutive months (loading doses, administered on Day 1, Months 1 and 2) followed by 3 mg/kg once every three months (maintenance doses, which start one month after the last loading dose, at Month 3). Body weight measured within three months prior to dosing or the pre-dose weight at the study visit or dosing day was used for dose calculations.

Individual Subject Dose Modification or Interruption

Dose modifications were not allowed. If dosing was interrupted because of an adverse event, study drug could be restarted at the same dose as per protocol, at the discretion of the investigator.

Individual Subject Dose Stopping Criteria

Treatment was to be discontinued for the following:

- A serious or intolerable AE
- An ALT or AST elevation >3×ULN without alternative cause and accompanied by clinical symptoms consistent with liver injury (e.g., nausea, right upper quadrant abdominal pain, jaundice) or total bilirubin ≥2×ULN or INR ≥1.5
- Significant protocol violations
- Non-adherence to the treatment regimen
- Pregnancy
- Loss to follow-up

Compliance

Study staff administered study drug in clinic during the placebo-controlled period.

Follow-Up

Patients who discontinued study medication prematurely were expected to attend all subsequent study visits and assessments until the end of the study.

Eligibility Criteria

Key Inclusion Criteria

1. Diagnosis of PH1 confirmed with alanine glyoxylate aminotransferase (AGXT) mutations
2. Adults and children ≥6 years of age
3. Mean (changed to median with Amendment 2) 24-hour urinary oxalate excretion ≥0.70 mmol/24 hr/1.73 m^2 from the first two valid 24-hour urine collections at screening

Key Exclusion Criteria

1. History of extrarenal systemic oxalosis, as determined by the investigator
2. History of kidney or liver transplant
3. eGFR ≤45 mL/min/1.73 m^2 (changed to <30 mL/min/1.73 m^2 with Amendment 2) at screening (eGFR calculated using the bedside 2009 Schwartz equation in patients <18 years of age, and Modification of Diet in Renal Disease [MDRD] equation in patients ≥18 years of age)
4. Screening ALT or AST >2×ULN, total bilirubin >1.5×ULN (except for patients with Gilbert's syndrome who were eligible if the total bilirubin was <2×ULN), or INR >1.5 or ≥3.5 if on oral anticoagulation therapy
5. Known active human immunodeficiency virus (HIV) infection; or evidence of current or chronic hepatitis C virus (HCV) or hepatitis B virus (HBV) infection
6. History of intolerance to subcutaneous injections
7. Not willing to comply with the contraceptive requirements during the study period. Women of child-bearing potential and post-menarchal pediatric female patients had to be

willing to use acceptable methods of contraception from 14 days before first dose, throughout study participation, and for 90 days after last dose or until study completion.
8. Female patient who is pregnant, planning a pregnancy, or breast-feeding.
9. History of alcohol abuse within 12 months of screening or unwilling or unable to limit alcohol consumption throughout the course of the study.

ILLUMINATE-B

Important Trial Dates

The study was initiated on April 22, 2019 and is ongoing. The data cutoff for the interim analysis provided with the original NDA submission was October 25, 2019. A data update report with a data cutoff of March 9, 2020 was provided during the review cycle on June 23, 2020.

Trial Administrative Structure

The trial administrative structure was the same as for ILLUMINATE-A. The Applicant submitted the meeting minutes for the open and closed sessions. Review of the minutes did not reveal any significant concerns about study conduct, efficacy, or safety.

Study Assessments

After the baseline visit (Day 1), patients were to return for study visits at Days 15, 29 and every 28 days thereafter for the first 6 months. Height and weight were obtained at all but the Day 15 visit. Triplicate 24-hour urine collections for urinary oxalate were obtained at baseline and Month 6, within 14 days before dosing, in those able to comply. The validity criteria for 24-hour collections were similar to ILLUMINATE-A with modification to the duration of collection (between 18-26 hours between the initial discarded void and the last void or attempt to void), and 24-hour creatinine content (5 mg/kg), which is appropriate for the younger population. The timing of other assessments, including physical examinations, vital signs, clinical laboratory assessments, and blood samples for ADA, was similar to ILLUMINATE-A.

Collection of Primary Endpoint Assessments

Three preferably first-morning, single-void spot urine collections were collected at screening and monthly from Months 1 to 6 up to seven days before dosing. Urinary oxalate was analyzed in a central laboratory using a validated assay ([(b) (4)]).

Study Procedures

Dosing

Dosing Regimen

The dosing regimen was based on weight obtained within seven days before dosing, as shown in Table 14. Monthly doses had to be at least 21 days apart. Patients who crossed weight thresholds followed the new dosing regimen and did not switch back to the lower-weight dosing schedule if their body weight subsequently decreased.

Individual Subject Dose Modification or Interruption

Like ILLUMINATE-A, dose modifications were not allowed. If dosing was interrupted because of an adverse event, study drug could be restarted at the same dose as per protocol, at the discretion of the investigator.

Individual Subject Dose Stopping Criteria

Same as ILLUMINATE-A.

Compliance

Study staff or trained home healthcare professionals observed and verified study drug administration.

Follow-Up

Patients who discontinued study medication prematurely were expected to attend all subsequent study visits and assessments until the end of the study.

Eligibility Criteria

Key Inclusion Criteria

- Diagnosis of PH1 confirmed with alanine glyoxylate aminotransferase (AGXT) mutations
- Children ≥37 weeks estimated gestational age but <6 years of age at consent
- Urinary oxalate-to-creatinine ratio >ULN for age in two of three single-void collections during screening
- If taking therapeutic pyridoxine, must have been on stable regimen for at least 90 days before screening, and able to remain the stable regimen until the Month 6 visit

Key Exclusion Criteria

- For patients ≥12 months of age at screening, eGFR ≤45 mL/min/1.73 m^2 (calculated using the bedside 2009 Schwartz equation). For patients <12 months of age, serum creatinine value per the central laboratory above the ULN for age at screening
- History of kidney or liver transplant, or anticipated liver transplant within 6 months after the first dose of lumasiran

All other key exclusion criteria were similar to ILLUMINATE-A.

16. Efficacy: Additional Information and Assessment

24-Hour Urinary Oxalate (Absolute Change)

The LS mean difference in 24-hour urinary oxalate from baseline to Month 6 (average of Months 3 to 6) was -0.98 mmol/24hr/1.73m^2 (p <0.0001; Table 69).

Table 69. 24-Hour Urinary Oxalate (Absolute Change)

Statistic	Placebo	Lumasiran
Sample size, n	13	26
Baseline mean (SEM)	1.8 (0.19)	1.84 (0.12)
LS mean (SEM)	-0.27 (0.08)	-1.24 (0.06)
95% CI	(-0.44, -0.10)	(-1.37, -1.12)
Difference in LS mean (SEM) (lumasiran-placebo)	-0.98 (0.10)	
95% CI	(-1.18, -0.78)	
p-value	<0.0001	

Abbreviations: CI=confidence interval; LS=least squares; N=number of subjects in treatment group; n=number of subjects with given characteristic; SEM=standard error of mean.
Source: sponsor's table verified by statistical reviewer.

Plasma Oxalate (Absolute Change)

The LS mean difference in plasma oxalate from baseline to Month 6 in patients with a baseline level ≥1.5×LLQ was -8.71 µmol/L (p <0.0001; Table 70). Plasma oxalate levels are generally not markedly elevated in a population with relatively preserved kidney function, and it is not clear what size of a treatment effect would be clinically meaningful; therefore, these findings were not included in labeling. These data, however, are supportive of an effect of lumasiran on urinary oxalate that could not be explained by a loss of kidney function, which would be expected to lead to an increase in plasma oxalate.

Table 70. Plasma Oxalate (Absolute Change)

Statistic	Placebo	Lumasiran
Sample size, n	10	23
Baseline mean (SEM)	17.76 (2.17)	15.73 (1.59)
LS mean (SEM)	1.25 (1.12)	-7.46 (0.77)
95% CI	(-1.04, 3.54)	(-9.03, -5.90)
Difference in LS mean (SEM) (lumasiran-placebo)	-8.71 (1.338)	
95% CI	(-11.45, -5.98)	
p-value	<0.0001	

Abbreviations: CI=confidence interval; LS=least squares; N=number of subjects in treatment group; n=number of subjects with given characteristic; SEM=standard error of mean.
Source: Sponsor's table verified by statistical reviewer.

17. Clinical Safety: Additional Information and Assessment

ILLUMINATE-A

Table 71. Adverse Events by System Organ Class and Preferred Term, Safety Population, ILLUMINATE-A

System Organ Class Preferred Term	Lumasiran (Overall) N=26 n (%)	Placebo (Overall) N=13 n (%)	Risk Difference (95% CI)[1]
General disorders and administration site conditions (SOC)	11 (42.3)	0	42.3 (23.3, 61.3)
Injection site reaction	6 (23.1)	0	23.1 (6.9, 39.3)
Injection site erythema	3 (11.5)	0	11.5 (-0.7, 23.8)
Injection site pain	3 (11.5)	0	11.5 (-0.7, 23.8)
Chest pain	1 (3.8)	0	3.8 (-3.5, 11.2)
Fatigue	1 (3.8)	0	3.8 (-3.5, 11.2)
Injection site discomfort	1 (3.8)	0	3.8 (-3.5, 11.2)
Psychiatric disorders (SOC)	3 (11.5)	0	11.5 (-0.7, 23.8)
Anxiety	1 (3.8)	0	3.8 (-3.5, 11.2)
Fear of injection	1 (3.8)	0	3.8 (-3.5, 11.2)
Irritability	1 (3.8)	0	3.8 (-3.5, 11.2)
Skin and subcutaneous tissue disorders (SOC)	3 (11.5)	0	11.5 (-0.7, 23.8)
Alopecia	1 (3.8)	0	3.8 (-3.5, 11.2)
Eczema	1 (3.8)	0	3.8 (-3.5, 11.2)
Erythema	1 (3.8)	0	3.8 (-3.5, 11.2)
Pruritus	1 (3.8)	0	3.8 (-3.5, 11.2)
Rash erythematous	1 (3.8)	0	3.8 (-3.5, 11.2)
Gastrointestinal disorders (SOC)	4 (15.4)	1 (7.7)	7.7 (-12.4, 27.7)
Abdominal pain	2 (7.7)	0	7.7 (-2.6, 17.9)
Abdominal pain upper	2 (7.7)	0	7.7 (-2.6, 17.9)
Abdominal pain lower	1 (3.8)	0	3.8 (-3.5, 11.2)
Constipation	1 (3.8)	0	3.8 (-3.5, 11.2)
Nausea	1 (3.8)	0	3.8 (-3.5, 11.2)
Abdominal discomfort	1 (3.8)	1 (7.7)	-3.8 (-20.1, 12.4)
Renal and urinary disorders (SOC)	2 (7.7)	0	7.7 (-2.6, 17.9)
Polyuria	1 (3.8)	0	3.8 (-3.5, 11.2)
Renal pain	1 (3.8)	0	3.8 (-3.5, 11.2)

System Organ Class Preferred Term	Lumasiran (Overall) N=26 n (%)	Placebo (Overall) N=13 n (%)	Risk Difference (95% CI)[1]
Infections and infestations (SOC)	11 (42.3)	5 (38.5)	3.8 (-28.7, 36.4)
Pneumonia	2 (7.7)	0	7.7 (-2.6, 17.9)
Urinary tract infection	2 (7.7)	0	7.7 (-2.6, 17.9)
Fungal skin infection	1 (3.8)	0	3.8 (-3.5, 11.2)
Infected bite	1 (3.8)	0	3.8 (-3.5, 11.2)
Kidney infection	1 (3.8)	0	3.8 (-3.5, 11.2)
Nasopharyngitis	1 (3.8)	0	3.8 (-3.5, 11.2)
Pharyngitis	1 (3.8)	0	3.8 (-3.5, 11.2)
Tonsillitis	1 (3.8)	0	3.8 (-3.5, 11.2)
Rhinitis	2 (7.7)	2 (15.4)	-7.7 (-29.8, 14.4)
Upper respiratory tract infection	2 (7.7)	2 (15.4)	-7.7 (-29.8, 14.4)
Otitis media acute	0	1 (7.7)	-7.7 (-22.2, 6.8)
Tooth infection	0	1 (7.7)	-7.7 (-22.2, 6.8)
Nervous system disorders (SOC)	7 (26.9)	3 (23.1)	3.8 (-24.7, 32.4)
Disturbance in attention	1 (3.8)	0	3.8 (-3.5, 11.2)
Dizziness	1 (3.8)	0	3.8 (-3.5, 11.2)
Hypoaesthesia	1 (3.8)	0	3.8 (-3.5, 11.2)
Restless legs syndrome	1 (3.8)	0	3.8 (-3.5, 11.2)
Headache	3 (11.5)	3 (23.1)	-11.5 (-37.5, 14.4)
Musculoskeletal and connective tissue disorders (SOC)	5 (19.2)	2 (15.4)	3.8 (-20.9, 28.6)
Flank pain	1 (3.8)	0	3.8 (-3.5, 11.2)
Groin pain	1 (3.8)	0	3.8 (-3.5, 11.2)
Musculoskeletal chest pain	1 (3.8)	0	3.8 (-3.5, 11.2)
Musculoskeletal pain	1 (3.8)	0	3.8 (-3.5, 11.2)
Back pain	2 (7.7)	1 (7.7)	0.0 (-17.7, 17.7)
Pain in extremity	0	1 (7.7)	-7.7 (-22.2, 6.8)
Blood and lymphatic system disorders (SOC)	1 (3.8)	0	3.8 (-3.5, 11.2)
Iron deficiency anaemia	1 (3.8)	0	3.8 (-3.5, 11.2)
Congenital, familial and genetic disorders (SOC)	1 (3.8)	0	3.8 (-3.5, 11.2)
Thalassaemia beta	1 (3.8)	0	3.8 (-3.5, 11.2)
Ear and labyrinth disorders (SOC)	1 (3.8)	0	3.8 (-3.5, 11.2)
Ear pain	1 (3.8)	0	3.8 (-3.5, 11.2)
Eye disorders (SOC)	1 (3.8)	0	3.8 (-3.5, 11.2)
Vision blurred	1 (3.8)	0	3.8 (-3.5, 11.2)
Immune system disorders (SOC)	1 (3.8)	0	3.8 (-3.5, 11.2)
Hypersensitivity	1 (3.8)	0	3.8 (-3.5, 11.2)

System Organ Class Preferred Term	Lumasiran (Overall) N=26 n (%)	Placebo (Overall) N=13 n (%)	Risk Difference (95% CI)[1]
Vascular disorders (SOC)	1 (3.8)	0	3.8 (-3.5, 11.2)
Hypertension	1 (3.8)	0	3.8 (-3.5, 11.2)
Metabolism and nutrition disorders (SOC)	1 (3.8)	1 (7.7)	-3.8 (-20.1, 12.4)
Vitamin D deficiency	1 (3.8)	0	3.8 (-3.5, 11.2)
Iron deficiency	0	1 (7.7)	-7.7 (-22.2, 6.8)
Injury, poisoning and procedural complications (SOC)	2 (7.7)	2 (15.4)	-7.7 (-29.8, 14.4)
Foot fracture	1 (3.8)	0	3.8 (-3.5, 11.2)
Tibia fracture	1 (3.8)	0	3.8 (-3.5, 11.2)
Contusion	0	1 (7.7)	-7.7 (-22.2, 6.8)
Gastrostomy tube site complication	0	1 (7.7)	-7.7 (-22.2, 6.8)
Respiratory, thoracic and mediastinal disorders (SOC)	2 (7.7)	2 (15.4)	-7.7 (-29.8, 14.4)
Cough	1 (3.8)	0	3.8 (-3.5, 11.2)
Nasal congestion	1 (3.8)	1 (7.7)	-3.8 (-20.1, 12.4)
Oropharyngeal pain	1 (3.8)	1 (7.7)	-3.8 (-20.1, 12.4)

Source: adae.xpt; Software: Python
Treatment-emergent adverse events defined as Treatment-emergent adverse events defined as an AE that occurs or worsens on or after the first dose date/time of study drug though 84 days after the last dose of study drug. In addition, any AE that worsened in intensity or was subsequently considered related to study drug was considered treatment-emergent. Only includes DB period.
1 Risk difference column shows absolute difference (with 95% confidence interval) between total treatment and comparator.
Abbreviations: CI, confidence interval; N, number of subjects in treatment arm; n, number of subjects with adverse event; SOC, system organ class

Table 72. Adverse Events by FDA Medical Query (Narrow) and Preferred Term, Safety Population, ILLUMINATE-A

FMQ (Narrow) Preferred Term	Lumasiran (Overall) N=26 n (%)	Placebo (Overall) N=13 n (%)	Risk Difference (95% CI)[1]
Local administration reactions (narrow FMQ)	9 (34.6)	0	34.6 (16.3, 52.9)
Injection site reaction	6 (23.1)	0	23.1 (6.9, 39.3)
Injection site erythema	3 (11.5)	0	11.5 (-0.7, 23.8)
Injection site pain	3 (11.5)	0	11.5 (-0.7, 23.8)
Injection site discomfort	1 (3.8)	0	3.8 (-3.5, 11.2)
Erythema (narrow FMQ)	4 (15.4)	0	15.4 (1.5, 29.3)
Injection site erythema	3 (11.5)	0	11.5 (-0.7, 23.8)
Erythema	1 (3.8)	0	3.8 (-3.5, 11.2)
Rash erythematous	1 (3.8)	0	3.8 (-3.5, 11.2)
Abdominal pain (narrow FMQ)	4 (15.4)	1 (7.7)	7.7 (-12.4, 27.7)
Abdominal pain	2 (7.7)	0	7.7 (-2.6, 17.9)
Abdominal pain upper	2 (7.7)	0	7.7 (-2.6, 17.9)
Abdominal pain lower	1 (3.8)	0	3.8 (-3.5, 11.2)
Abdominal discomfort	1 (3.8)	1 (7.7)	-3.8 (-20.1, 12.4)
Dyspepsia (narrow FMQ)	2 (7.7)	0	7.7 (-2.6, 17.9)
Abdominal pain upper	2 (7.7)	0	7.7 (-2.6, 17.9)
Pneumonia (narrow FMQ)	2 (7.7)	0	7.7 (-2.6, 17.9)
Pneumonia	2 (7.7)	0	7.7 (-2.6, 17.9)
Anxiety (narrow FMQ)	2 (7.7)	0	7.7 (-2.6, 17.9)
Anxiety	1 (3.8)	0	3.8 (-3.5, 11.2)
Fear of injection	1 (3.8)	0	3.8 (-3.5, 11.2)
Back pain (narrow FMQ)	3 (11.5)	1 (7.7)	3.8 (-15.1, 22.8)
Flank pain	1 (3.8)	0	3.8 (-3.5, 11.2)
Back pain	2 (7.7)	1 (7.7)	0.0 (-17.7, 17.7)
Anaemia (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Iron deficiency anaemia	1 (3.8)	0	3.8 (-3.5, 11.2)
Systemic hypertension (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Hypertension	1 (3.8)	0	3.8 (-3.5, 11.2)
Constipation (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Constipation	1 (3.8)	0	3.8 (-3.5, 11.2)
Nausea (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Nausea	1 (3.8)	0	3.8 (-3.5, 11.2)
Fatigue (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Fatigue	1 (3.8)	0	3.8 (-3.5, 11.2)
Dizziness (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Dizziness	1 (3.8)	0	3.8 (-3.5, 11.2)
Paraesthesia (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Hypoaesthesia	1 (3.8)	0	3.8 (-3.5, 11.2)
Irritability (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Irritability	1 (3.8)	0	3.8 (-3.5, 11.2)
Cough (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Cough	1 (3.8)	0	3.8 (-3.5, 11.2)
Alopecia (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Alopecia	1 (3.8)	0	3.8 (-3.5, 11.2)
Pruritus (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Pruritus	1 (3.8)	0	3.8 (-3.5, 11.2)

FMQ (Narrow) Preferred Term	Lumasiran (Overall) N=26 n (%)	Placebo (Overall) N=13 n (%)	Risk Difference (95% CI)[1]
Rash (narrow FMQ)	1 (3.8)	0	3.8 (-3.5, 11.2)
Rash erythematous	1 (3.8)	0	3.8 (-3.5, 11.2)
Nasopharyngitis (narrow FMQ)	4 (15.4)	2 (15.4)	0.0 (-24.0, 24.0)
Nasopharyngitis	1 (3.8)	0	3.8 (-3.5, 11.2)
Pharyngitis	1 (3.8)	0	3.8 (-3.5, 11.2)
Rhinitis	2 (7.7)	2 (15.4)	-7.7 (-29.8, 14.4)
Haemorrhage (narrow FMQ)	0	1 (7.7)	-7.7 (-22.2, 6.8)
Contusion	0	1 (7.7)	-7.7 (-22.2, 6.8)
Headache (narrow FMQ)	3 (11.5)	3 (23.1)	-11.5 (-37.5, 14.4)
Headache	3 (11.5)	3 (23.1)	-11.5 (-37.5, 14.4)

Source: adae.xpt; Software: Python
Treatment-emergent adverse events defined as Treatment-emergent adverse events defined as an AE that occurs or worsens on or after the first dose date/time of study drug though 84 days after the last dose of study drug. In addition, any AE that worsened in intensity or was subsequently considered related to study drug was considered treatment-emergent. Only includes DB period.
1 Risk difference column shows absolute difference (with 95% confidence interval) between total treatment and comparator.
Abbreviations: CI, confidence interval; FMQ, FDA medical query; N, number of subjects in treatment arm; n, number of subjects with adverse event.

ILLUMINATE-B

Table 73. Adverse Events by System Organ Class and Preferred Term, Safety Population, ILLUMINATE-B

System Organ Class Preferred Term	Lumasiran (Overall) N=18 n (%)
Infections and infestations (SOC)	15 (83.3)
Rhinitis	4 (22.2)
Upper respiratory tract infection	3 (16.7)
Gastroenteritis	2 (11.1)
Nasopharyngitis	2 (11.1)
Bronchitis	2 (11.1)
Tonsillitis	1 (5.6)
Asymptomatic bacteriuria	1 (5.6)
Conjunctivitis bacterial	1 (5.6)
Croup infectious	1 (5.6)
Ear infection	1 (5.6)
Influenza	1 (5.6)
Oral herpes	1 (5.6)
Pharyngitis	1 (5.6)
Pneumonia	1 (5.6)
Urinary tract infection	1 (5.6)
Viral infection	1 (5.6)
Viral pharyngitis	1 (5.6)
General disorders and administration site conditions (SOC)	9 (50.0)
Pyrexia	6 (33.3)
Injection site reaction	3 (16.7)
Influenza like illness	1 (5.6)
Gastrointestinal disorders (SOC)	7 (38.9)
Vomiting	3 (16.7)
Teething	2 (11.1)
Diarrhoea	2 (11.1)

System Organ Class Preferred Term	Lumasiran (Overall) N=18 n (%)
Abdominal pain	1 (5.6)
Aphthous ulcer	1 (5.6)
Mouth ulceration	1 (5.6)
Nausea	1 (5.6)
Respiratory, thoracic and mediastinal disorders (SOC)	6 (33.3)
Cough	2 (11.1)
Oropharyngeal pain	2 (11.1)
Rhinorrhoea	1 (5.6)
Nasal congestion	1 (5.6)
Injury, poisoning and procedural complications (SOC)	3 (16.7)
Arthropod bite	1 (5.6)
Fall	1 (5.6)
Arthropod sting	1 (5.6)
Psychiatric disorders (SOC)	2 (11.1)
Irritability	1 (5.6)
Abnormal behaviour	1 (5.6)
Investigations (SOC)	2 (11.1)
Blood creatinine increased	1 (5.6)
Urine analysis abnormal	1 (5.6)
Skin and subcutaneous tissue disorders (SOC)	2 (11.1)
Rash	1 (5.6)
Rash generalised	1 (5.6)
Rash maculo-papular	1 (5.6)
Blood and lymphatic system disorders (SOC)	1 (5.6)
Iron deficiency anaemia	1 (5.6)
Congenital, familial and genetic disorders (SOC)	1 (5.6)
Factor xii deficiency	1 (5.6)
Ear and labyrinth disorders (SOC)	1 (5.6)
Ear pain	1 (5.6)
Metabolism and nutrition disorders (SOC)	1 (5.6)
Iron deficiency	1 (5.6)
Nervous system disorders (SOC)	1 (5.6)
Headache	1 (5.6)
Renal and urinary disorders (SOC)	1 (5.6)
Haematuria	1 (5.6)
Reproductive system and breast disorders (SOC)	1 (5.6)
Gynaecomastia	1 (5.6)

Source: adae.xpt; Software: Python
Treatment-emergent adverse events defined as an AE that occurs or worsens on or after the first dose date/time of study drug though 84 days after the last dose of study drug. In addition, any AE that worsened in intensity or was subsequently considered related to study drug was considered treatment-emergent.
Abbreviations: N, number of subjects in treatment arm; n, number of subjects with adverse event; SOC, system organ class.

Table 74. Adverse Events by FDA Medical Query (Narrow) and Preferred Term, Safety Population, ILLUMINATE-B

FMQ (Narrow) Preferred Term	Lumasiran (Overall) N=18 n (%)
Nasopharyngitis (narrow FMQ)	6 (33.3)
Rhinitis	4 (22.2)
Nasopharyngitis	2 (11.1)
Pharyngitis	1 (5.6)
Viral pharyngitis	1 (5.6)
Pyrexia (narrow FMQ)	6 (33.3)
Pyrexia	6 (33.3)
Local administration reactions (narrow FMQ)	3 (16.7)
Injection site reaction	3 (16.7)
Vomiting (narrow FMQ)	3 (16.7)
Vomiting	3 (16.7)
Cough (narrow FMQ)	2 (11.1)
Cough	2 (11.1)
Diarrhoea (narrow FMQ)	2 (11.1)
Diarrhoea	2 (11.1)
Rash (narrow FMQ)	2 (11.1)
Rash	1 (5.6)
Rash generalised	1 (5.6)
Rash maculo-papular	1 (5.6)
Anaemia (narrow FMQ)	1 (5.6)
Iron deficiency anaemia	1 (5.6)
Irritability (narrow FMQ)	1 (5.6)
Irritability	1 (5.6)
Abdominal pain (narrow FMQ)	1 (5.6)
Abdominal pain	1 (5.6)
Gynaecomastia (narrow FMQ)	1 (5.6)
Gynaecomastia	1 (5.6)
Haemorrhage (narrow FMQ)	1 (5.6)
Haematuria	1 (5.6)
Headache (narrow FMQ)	1 (5.6)
Headache	1 (5.6)
Nausea (narrow FMQ)	1 (5.6)
Nausea	1 (5.6)
Pneumonia (narrow FMQ)	1 (5.6)
Pneumonia	1 (5.6)

Source: adae.xpt; Software: Python
Treatment-emergent adverse events defined as an AE that occurs or worsens on or after the first dose date/time of study drug though 84 days after the last dose of study drug. In addition, any AE that worsened in intensity or was subsequently considered related to study drug was considered treatment-emergent.
Abbreviations: FMQ, FDA medical query; N, number of subjects in treatment arm; n, number of subjects with adverse event.

18. Mechanism of Action/Drug Resistance: Additional Information and Assessment

Insert text here.

19. Other Drug Development Considerations: Additional Information and Assessment

Insert text here.

20. Data Integrity-Related Consults (Office of Scientific Investigations, Other Inspections)

The Office of Scientific Investigations participated in discussions regarding the need for clinical site inspections (see Section 10).

21. Labeling Summary of Considerations and Key Additional Information

Numerous organizational and editorial revisions have been made to the Applicant's originally proposed draft prescribing information. Substantive revisions were proposed and agreed to with the Applicant and are detailed below. For final labeling, refer to the Approval letter.

Indications and Usage

The indication statement has been modified to state that OXLUMO is indicated '…for the treatment of PH1 to lower urinary oxalate levels...'

Use in Specific Populations

In Section 8.6, Hepatic Impairment, language has been modified to indicated that no dosage adjustment is necessary in patients with mild and moderate hepatic impairment.

Clinical Pharmacology

In Section 12.1, Mechanism of Action, language has been added to clarify that as a result of lumasiran's mechanism of action, it is not expected to be effective in the treatment of primary hyperoxaluria type 2 or type 3.

Clinical Studies

In Section 14.1, ILLUMINATE-A, claims related to (b) (4) have been removed because the available data are not adequate to support inclusion.

Throughout Labeling

Claims and descriptions regarding (b) (4) have been removed from labeling. These data are not adequate (b) (4)

(b) (4)

22. Postmarketing Requirements and Commitments

These are the postmarketing requirements:

- Complete the 26-week GLP carcinogenicity study of lumasiran by subcutaneous injection in TgRasH2 mice, per agreed SPA issued on November 13, 2019
- Complete the 2-year GLP carcinogenicity study of lumasiran by subcutaneous injection in Sprague Dawley rats, per agreed SPA issued on November 13, 2019
- Submit the final audited study reports of the above studies and labeling update (for Section 13.1) for Agency review.

23. Financial Disclosure

Table 75. Covered Clinical Studies: ILLUMINATE-A, ILLUMINATE-B, ALN-GO1-001 and ALN-GO1-002

Was a list of clinical investigators provided:	Yes ☒	No ☐ (Request list from Applicant)
Total number of investigators identified: 124		
Number of investigators who are Sponsor employees (including both full-time and part-time employees): 0		
Number of investigators with disclosable financial interests/arrangements (Form FDA 3455): 4		
If there are investigators with disclosable financial interests/arrangements, identify the number of investigators with interests/arrangements in each category (as defined in 21 CFR 54.2(a), (b), (c), and (f)): Compensation to the investigator for conducting the study where the value could be influenced by the outcome of the study: 0 Significant payments of other sorts: 4 (payments for consulting services, service on advisory boards, speaker fees, honoraria, research support, and payment for laboratory audits) Proprietary interest in the product tested held by investigator: 0 Significant equity interest held by investigator: 0 Sponsor of covered study: 0		
Is an attachment provided with details of the disclosable financial interests/arrangements:	Yes ☒	No ☐ (Request details from Applicant)
Is a description of the steps taken to minimize potential bias provided:	Yes ☒	No ☐ (Request information from Applicant)
Number of investigators with certification of due diligence (Form FDA 3454, box 3): 0		
Is an attachment provided with the reason: N/A	Yes ☐	No ☐ (Request explanation from Applicant)

24. References

Matos V, Van Melle G, Werner D, Bardy D, Guignard JP. Urinary oxalate and urate to creatinine ratios in a healthy pediatric population. Am J Kidney Dis. 1999 Aug;34(2):e1. PMID: 10430995.

Noda T, Todani T, Watanabe Y, Yamamoto S. Liver volume in children measured by computed tomography. Pediatr Radiol. 1997 Mar;27 (3):250-2. PMID: 9126583

Sugahara K, Togashi H, Takahashi K, et al. Separate analysis of asialoglycoprotein receptors in the right and left hepatic lobes using Tc-GSA SPECT. Hepatology. 2003 Dec;38(6):1401-9. PMID: 14647051.

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25. Review Team

Table 76. Reviewers of Integrated Assessment

Role	Name(s)
Regulatory Project Manager	Brian Proctor, RAC
Nonclinical Reviewer	Phil Gatti, PhD
Nonclinical Team Leader	Xuan Chi, MD, PhD
Office of Clinical Pharmacology Reviewer(s)	Harisudhan Thanukrishnan, PhD; Liang Li, PhD (Pharmacometrics)
Office of Clinical Pharmacology Team Leader(s)	Sudharshan Hariharan, PhD; Justin Earp, PhD (Pharmacometrics); Girish Bende, PhD (QT)
Clinical Reviewer	Kirtida Mistry, MBBCh, DCH, MRCPCH
Clinical Team Leader	Kimberly Smith, MD, MS
Statistical Reviewer	Dali Zhou, PhD
Statistical Team Leader	Jialu Zhang, PhD
Cross-Disciplinary Team Leader	Kimberly Smith, MD, MS
Division Director (Pharm/Tox)	Todd Bourcier, PhD
Deputy Division Director (OCP)	Doanh Tran, PhD
Division Director (OCP)	Shirley Seo, PhD
Division Director (OB)	Mark Rothmann, PhD
Associate Director for Labeling (DCN)	Michael Monteleone, MS
Deputy Director for Safety (DCN)	Mary Ross Southworth, PharmD
Deputy Director (DCN)	Aliza Thompson, MD, MS
Division Director (DCN)	Norman Stockbridge, MD, PhD
Office Director	Ellis Unger, MD

OCP=Office of Clinical Pharmacology
OB=Office of Biostatistics,
DCN=Division of Cardiology and Nephrology

Table 77. Additional Reviewers of Application

Office or Discipline	Name(s)
CMC Reviewers	Monica Cooper, PhD; Dhanalakshmi Kasi, PhD; Kumar Janoria, PhD; Poonam Delvadia; Elizabeth Bearr, PhD, Kris Raman, PhD
CMC Lead and ATL	Mohan Sapru, PhD
OPDP	Puja Shah, PharmD
OSI	NA
OSE/DEPI	Marie Bradley PhD, MScPH, MPharm (SE) and Efe Eworuke, PhD (TL)
OSE/DMEPA	Marie Aidoo, PharmD, MPH (SE) and Hina Mehta, PharmD (TL)
OBP/DBRRI	Primary Reviewer: Brian Janelsins, PhD Secondary Reviewer: Rachel Novak, PhD
OSE/DRM	Mona Patel, PharmD (SE) and Laura Zendel, PharmD (TL)
DPMH	Miriam Dinatale, DO

CMC=Chemistry, Manufacturing, and Controls
OPDP=Office of Prescription Drug Promotion
OSI=Office of Scientific Investigations
OSE=Office of Surveillance and Epidemiology
DEPI=Division of Epidemiology
DMEPA=Division of Medication Error Prevention and Analysis

DRM=Division of Risk Management
DPMH=Division of Pediatrics and Maternal Health

Table 78. Signatures of Reviewers

Discipline and Title or Role	Reviewer Name	Office/Division	Sections Authored/ Acknowledged/ Approved
Cross-Disciplinary	Brian Proctor	OND/DCN	☐ Authored ☒ Approved
Project Manager	**Signature:** Brian Proctor -S Digitally signed by Brian Proctor -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, cn=Brian Proctor -S, 0.9.2342.19200300.100.1.1=2001765333 Date: 2020.11.18 20:03:08 -05'00'		
Pharmacology/Toxicology	Phil Gatti	OND/DPTCHEN	5.1, 7.1, 8.4, Appendix 13.1, 13.2, 22 ☒ Authored ☐ Approved
Reviewer	**Signature:** Philip J. Gatti -S Digitally signed by Philip J. Gatti -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, 0.9.2342.19200300.100.1.1=2000323344, cn=Philip J. Gatti -S Date: 2020.11.19 08 52:29 -05'00'		
Pharmacology/Toxicology	Xuan Chi	OND/DPTCHEN	5.1, 7.1, 8.4, Appendix 13.1, 13.2, 22 ☒ Authored ☐ Approved
Team Leader	**Signature:** Xuan Chi -S Digitally signed by Xuan Chi -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, cn=Xuan Chi -S, 0.9.2342.19200300.100.1.1=0014066742 Date: 2020.11.19 14:39:52 -05'00'		
Pharmacology/Toxicology	Todd Bourcier	OND/DPTCHEN	5.1, 7.1, 8.4, Appendix 13.1, 13.2, 22 ☐ Authored ☒ Approved
Supervisor	**Signature:** Todd M. Bourcier -S Digitally signed by Todd M. Bourcier -S DN: c=US, o=U S. Government, ou=HHS, ou=FDA, ou=People, 0.9.2342.19200300.100.1.1=1300235462, cn=Todd M. Bourcier -S Date: 2020.11.19 14:57:54 -05'00'		
Clinical Pharmacology	Harisudhan Thanukrishnan	OTS/OCP/DCEP	5, 8.1, 8.2, Appendix 14.1, 14.2.3, 14.2.4, 14.2.5, 14.2.6 ☒ Authored ☐ Approved
Reviewer	**Signature:** Harisudhan Thanukrishnan -S Digitally signed by Harisudhan Thanukrishnan S DN: c=US o=U S Government ou=HHS ou=FDA ou=People 0 9 2342 19200300 100 1 1=2002874409 cn=Harisudhan Thanukrishnan S Date: 2020 11 19 07:49:44 05 00'		

Discipline and Title or Role	Reviewer Name	Office/Division	Sections Authored/ Acknowledged/ Approved
Clinical Pharmacology/Pharmacometrics	Liang Li	OTS/OCP/DPM	6.1, Appendix 14.2.1, 14.2.2 ☒ Authored ☐ Approved
Reviewer	**Signature:** Liang Li -S Digitally signed by Liang Li -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, cn=Liang Li -S, 0.9.2342.19200300.100.1.1=2001459144 Date: 2020.11.18 21:52:31 -05'00'		
Clinical Pharmacology	Sudharshan Hariharan	OTS/OCP/DCEP	5, 8.1, 8.2, Appendix 14.1, 14.2.3, 14.2.4, 14.2.5, 14.2.6 ☒ Authored ☐ Approved
Team Leader	**Signature:** Sudharshan Hariharan -S Digitally signed by Sudharshan Hariharan -S DN: c=US o=U S Government ou=HHS ou=FDA ou=People 0 9 2342 19200300 100 1 1=2000394743 cn=Sudharshan Hariharan S Date: 2020 11 19 07:03:27 05 00'		
Clinical Pharmacology/Pharmacometrics	Justin Earp	OTS/OCP/DPM	6.1, Appendix 14.2.1, 14.2.2 ☒ Authored ☐ Approved
Team Leader	**Signature:** Justin C. Earp -S Digitally signed by Justin C. Earp -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, cn=Justin C. Earp -S, 0.9.2342.19200300.100.1.1=1300436664 Date: 2020.11.18 20:15:16 -05'00'		
Clinical Pharmacology	Doanh Tran	OTS/OCP/DCEP	5, 6.1, 8.1, 8.2, Appendix 14.1, 14.2.1, 14.2.2, 14.2.3, 14.2.4, 14.2.5, 14.2.6 ☐ Authored ☒ Approved
Deputy Director	**Signature:** Doanh C. Tran -S Digitally signed by Doanh C. Tran -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, cn=Doanh C. Tran -S, 0.9.2342.19200300.100.1.1=1300378169 Date: 2020.11.19 12:21:03 -05'00'		
Biometrics	Dali Zhou	OB/DB2	6.2.2.3, 6.2.2.4, 6.2.3.3, 6.2.3.4, Appendix 16 ☒ Authored ☐ Approved
Reviewer	**Signature:** Dali Zhou -S Digitally signed by Dali Zhou -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, cn=Dali Zhou -S, 0.9.2342.19200300.100.1.1=2002462232 Date: 2020.11.18 20:53:56 -05'00'		

Discipline and Title or Role	Reviewer Name	Office/Division	Sections Authored/ Acknowledged/ Approved
Biometrics	Jialu Zhang	OB/DB2	6.2.2.3, 6.2.2.4, 6.2.3.3, 6.2.3.4, Appendix 16 ☒ Authored ☐ Approved
Team Leader	**Signature:** Jialu Zhang -S Digitally signed by Jialu Zhang -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, cn=Jialu Zhang -S, 0.9.2342.19200300.100.1.1=1300369843 Date: 2020.11.18 22:05:46 -05'00'		
Biometrics	Mark Rothmann	OB/DB2	6.2.2.3, 6.2.2.4, 6.2.3.3, 6.2.3.4, Appendix 16 ☐ Authored ☒ Approved
Division Director	**Signature:** Mark D. Rothmann -S Digitally signed by Mark D. Rothmann -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, 0.9.2342.19200300.100.1.1=1300144907, cn=Mark D. Rothmann -S Date: 2020.11.19 09:54:26 -05'00'		
Clinical	Kirtida Mistry	OND/DCN	2, 3, 4, 6.2, 7.2-7.6, 8.3, 8.4, 9.1, 10, 11; Appendix 12, 15-17, 20, 25 ☒ Authored ☐ Approved
Reviewer	**Signature:** Kirtida Mistry -S Digitally signed by Kirtida Mistry -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, cn=Kirtida Mistry -S, 0.9.2342.19200300.100.1.1=2002814821 Date: 2020.11.19 06:19:43 -05'00'		
Clinical	Kimberly Smith	OND/DCN	1-4, 6.2, 7.2-7.6, 8.3, 8.4, 9-11; Appendix 12, 15-17, 20, 25 ☒ Authored ☐ Approved
Cross-Disciplinary Team Lead	**Signature:** Kimberly Smith -S5 Digitally signed by Kimberly Smith -S5 DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, 0.9.2342.19200300.100.1.1=2000650159, cn=Kimberly Smith -S5 Date: 2020.11.19 07:06:21 -05'00'		
Clinical	Michael Monteleone	OND/DCN	21 ☒ Authored ☐ Approved
Associate Director for Labeling	**Signature:** Michael Monteleone -S Digitally signed by Michael Monteleone -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, 0.9.2342.19200300.100.1.1=2000336759, cn=Michael Monteleone -S Date: 2020.11.19 14:43:34 -05'00'		
Clinical	Mary Ross Southworth	OND/DCN	☐ Authored ☒ Approved
Team Leader	**Signature:** Mary R. Southworth -S Digitally signed by Mary R Southworth S DN: c=US o=U S Government ou=HHS ou=FDA ou=People 0.9.2342 19200300 100 1 1=1300234574 cn=Mary R Southworth S Date: 2020 11 19 07:20 41 05'00'		

Discipline and Title or Role	Reviewer Name	Office/Division	Sections Authored/ Acknowledged/ Approved
Clinical	Aliza Thompson	OND/DCN	☐ Authored ☒ Approved
Deputy Director	**Signature:** Aliza M. Thompson -S D gital y signed by Aliza M Thompson S DN: c=US o=U S Government ou=HHS ou=FDA ou=People 0 9 2342 19200300 100 1 1=1300423038 cn=Al za M Thompson S Date: 2020 11 19 07 53:21 05'00'		
Clinical	Norman Stockbridge	OND/DCN	☐ Authored ☒ Approved
Division Director	**Signature:** Norman L. Stockbridge -S Digitally signed by Norman L Stockbridge -S DN: c=US, o=U.S. Government, ou=HHS, ou=FDA, ou=People, 0.9.2342.19200300.100.1.1=1300068764, cn=Norman L. Stockbridge -S Date: 2020.11.19 12:14:32 -05'00'		

OND=Office of New Drugs
DPTCHEN=Division of Pharm/Tox for Cardiology, Hematology, Endocrinology, and Nephrology
OTS=Office of Translational Sciences
OCP=Office of Clinical Pharmacology
DCEP=Division of Cardiometabolic and Endocrine Pharmacology
DPM=Division of Pharmacometrics
OB=Office of Biostatistics
DB2=Division of Biometrics 2
DCN=Division of Cardiology and Nephrology

This is a representation of an electronic record that was signed electronically. Following this are manifestations of any and all electronic signatures for this electronic record.

/s/

KIMBERLY A SMITH
11/19/2020 04:29:25 PM

ELLIS F UNGER
11/19/2020 11:54:25 PM