15 October 2020
EMA/568312/2020
Committee for Medicinal Products for Human Use (CHMP)

Assessment report

Oxlumo

International non-proprietary name: lumasiran

Procedure No. EMEA/H/C/005040/0000

Note

Assessment report as adopted by the CHMP with all information of a commercially confidential nature deleted.

Official address Domenico Scarlattilaan 6 ● 1083 HS Amsterdam ● The Netherlands
Address for visits and deliveries Refer to www.ema.europa.eu/how-to-find-us
Send us a question Go to www.ema.europa.eu/contact **Telephone** +31 (0)88 781 6000 An agency of the European Union

Table of contents

List of abbreviations

AAS	Atomic Absorption Spectrometry
ADA	Anti-drug antibody
ADR	Adverse drug reaction
AE	Adverse event
AGXT	Alanine-glyoxylate aminotransferase gene
AGT	Alanine-glyoxylate aminotransferase
ALT	Alanine aminotransferase
ASGPR	Asialoglycoprotein receptor
AST	Aspartate aminotransferase
AUC	Area under the plasma concentration-time curve
AX-HPLC	Anion exchange HPLC
BIL	Bilirubin
BSA	Body surface area
Cmax	Maximum observed plasma concentration
CHMP	Committee for Medicinal Products for Human use
CL	Clearance
CL/F	Apparent plasma clearance; calculated as dose/AUC0-inf
CLR	Renal clearance
CPG	Controlled pore glass
CPP	Critical process parameter
CQA	Critical Quality Attribute
CSR	Clinical study report
CYP	Cytochrome P450
DB	Double-blind
DDI	Drug-drug interaction
DMC	Data Monitoring Committee
DoE	Design of experiments
DP	Drug product
DS	Drug substance
EAP	Expanded Access Protocol
EC	European Commission
ECG	Electrocardiogram

eGFR	Estimated glomerular filtration rate
EMA	European Medicines Agency
ESRD	End-stage renal disease
FDA	United States Food and Drug Administration
FTIR	Fourrier Transform Infrared Spectroscopy
GalNAc	N-acetylgalactosamine
GalNAc-PS	GalNAc polymer support
GC-FID	Gas chromatography flame ionising detector
GO	Glycolate oxidase
GR	Glyoxylate reductase
HAO1	Hydroxyacid oxidase 1
hERG	Human ether à go go related gene
HLT	High Level Term
HPLC	High performance liquid chromatography
ICH	International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
IPC	In-process control
IPRP-HPLC	Ion-pair reversed phase HPLC
ISR	Injection site reaction
ISS	Integrated Summary of Safety
KF	Karl Fischer titration
LDH	Lactate dehydrogenase
LFT	Liver function test
LLOQ	Lower limit of quantitation
MAD	Multiple-ascending dose
MedDRA	Medical Dictionary for Regulatory Activities
mRNA	Messenger RNA
MS	Mass Spectrometry
NCA	Non-compartmental analysis
NMR	Nuclear Magnetic Resonance
NMT	Not more than
NOR	Normal Operating Range
OD	Orphan drug
OFAT	One factor at a time

OLE	Open-label extension
PACMP	Post approval change management protocol
PAR	Proven Acceptable Range
PD	Pharmacodynamic(s)
PH	Primary hyperoxaluria
PH1	Primary hyperoxaluria type 1
Ph. Eur.	European Pharmacopoeia
PIP	Paediatric Investigation Plan
PK	Pharmacokinetic(s)
PP	Polypropylene
PPQ	Process performance qualification
PRIME	European Medicines Agency Priority Medicines
q3M	Once every 3 months
qM	Once monthly
QTc	Corrected QT interval
QTPP	Quality target product profile
RH	Relative Humidity
RISC	RNA-induced silencing complex
REC	Recommendation
RNAi	RNA interference
RP-HPLC	Reversed phase HPLC
SAD	Single-ascending dose
SAE	Serious adverse event
SC	Subcutaneous
siRNA	Small interfering RNAs
SMQ	Standardised MedDRA Query
SOC	System Organ Class
t½	Elimination half-life; time required for a 50% decrease in the concentration of a drug
tmax	Time to maximum lumasiran plasma concentration
UF	Ultrafiltration
ULN	Upper limit of normal
UV	Ultraviolet

1. Background information on the procedure

1.1. Submission of the dossier

The applicant Alnylam Netherlands B.V. submitted on 31 March 2020 an application for marketing authorisation to the European Medicines Agency (EMA) for Oxlumo, through the centralised procedure falling within the Article 3(1) and point 4 of Annex of Regulation (EC) No 726/2004. The eligibility to the centralised procedure was agreed upon by the EMA/CHMP on 22 March 2018.

Oxlumo, was designated as an orphan medicinal product EU/3/16/1637 on 21 March 2016 in the following condition: treatment of primary hyperoxaluria.

Oxlumo was granted eligibility to PRIME on 22 March 2018 in the following indication: treatment of primary hyperoxaluria Type 1.

Eligibility to PRIME was granted at the time in view of the following:

- Primary hyperoxaluria Type 1 is a serious genetic condition resulting in end-stage renal disease for which there are no effective treatments other than organ transplantation and pyridoxine which could delay disease progression but only in a sub-set of affected patients.
- The mechanism of action of lumasiran provides strong biological plausibility as an effective treatment for the whole target population.
- Despite the limited data available data, both non-clinical and clinical consistently demonstrate a dramatic decrease in urinary oxalate levels. The results show 24-h urinary oxalate excretion close to normalisation at end of observation period in both cohorts of treated patients, meeting the definition of positive response reported with existing therapies.

The applicant applied for the following indication the treatment of primary hyperoxaluria type 1 (PH1) in all age groups.

The legal basis for this application refers to:

Article 8.3 of Directive 2001/83/EC - complete and independent application

The application submitted is composed of administrative information, complete quality data, non-clinical and clinical data based on applicants' own tests and studies and/or bibliographic literature substituting/supporting certain tests or studies.

Information on Paediatric requirements

Pursuant to Article 7 of Regulation (EC) No 1901/2006, the application included an EMA Decision P/0004/2020 on the agreement of a paediatric investigation plan (PIP).

At the time of submission of the application, the PIP EMEA-002079-PIP01-16-M01 was not yet completed as some measures were deferred.

Information relating to orphan market exclusivity

Following the CHMP positive opinion on this marketing authorisation, the Committee for Orphan Medicinal Products (COMP) reviewed the designation of Oxlumo as an orphan medicinal product in the

approved indication. More information on the COMP's review can be found in the Orphan maintenance assessment report published under the 'Assessment history' tab on the Agency's website: ema.europa.eu/en/medicines/human/EPAR/oxlumo.

Similarity

Pursuant to Article 8 of Regulation (EC) No. 141/2000 and Article 3 of Commission Regulation (EC) No 847/2000, the applicant did not submit a critical report addressing the possible similarity with authorised orphan medicinal products because there is no authorised orphan medicinal product for a condition related to the proposed indication.

Applicant's request for consideration

Accelerated assessment

The applicant requested accelerated assessment in accordance to Article 14 (9) of Regulation (EC) No 726/2004.

New active Substance status

The applicant requested the active substance lumasiran contained in the above medicinal product to be considered as a new active substance, as the applicant claims that it is not a constituent of a medicinal product previously authorised within the European Union.

PRIME support

Upon granting of eligibility to PRIME, Martina Weise was appointed by the CHMP as rapporteur.

A kick-off meeting was held on 6 September 2018. The objective of the meeting was to discuss the development programme and regulatory strategy for the product. The applicant was recommended to address the following key issues through relevant regulatory procedures:

- The need for carcinogenicity studies;
- The clinical pharmacology package with a focus on the need for a DDI study and generation of data in patients with moderate to severe hepatic impairment;
- The PK/PD model used for dose selection; an
- The design of Study ALN-GO1-003 (ILLUMINATE-A) in children and adults with PH1 and relatively intact renal function (eGFR >45 mL/min/1.73m^2).

Protocol assistance

The applicant received the following Protocol assistance on the development relevant for the indication, subject to the present application:

Date	Reference	SAWP co-ordinators
28 February 2018	EMEA/H/SA/4014/1/2018/PA/PR/III	Dr Elmer Schabel, Dr Hrefna Gudmundsdottir
19 September 2019	EMEA/H/SA/4014/2/2019/PA/PR/I	Dr Elmer Schabel, Dr Hrefna Gudmundsdottir

The Protocol assistance pertained to the following non-clinical and clinical aspects:

- Adequacy of the planned nonclinical programme to support MAA in adult and paediatric patients with PH1.
- Acceptability and timing of the planned carcinogenicity studies
- Appropriateness of the plans for investigation of drug-drug-interactions
- Need for a hepatic impairment study
- Adequacy of observed PKPD data and the PKPD modelling strategy to support Phase 3 dose selection in patients below and over 6 years of age with either preserved or reduced renal function
- Acceptability of the proposed pivotal Phase 3 study design (ILLUMINATE-A): patient population, efficacy and safety endpoints, placebo-controlled period, potential comparison to historical control group, plans for follow-up, sample size, statistical analysis, safety database
- Adequacy of the envisaged clinical data package (efficacy and safety) to support B/R assessment at the time of MAA

1.2. Steps taken for the assessment of the product

The Rapporteur and Co-Rapporteur appointed by the CHMP were:

Rapporteur: Martina Weise Co-Rapporteur: Fátima Ventura

The application was received by the EMA on	31 March 2020
Accelerated Assessment procedure was agreed-upon by CHMP on	27 February 2020
The procedure started on	23 April 2020
The Rapporteur's first Assessment Report was circulated to all CHMP members on	24 June 2020
The Co-Rapporteur's first Assessment Report was circulated to all CHMP members on	26 June 2020
The PRAC Rapporteur's first Assessment Report was circulated to all PRAC members on	29 June 2020
The PRAC agreed on the PRAC Assessment Overview and Advice to CHMP during the meeting on	9 July 2020
The CHMP agreed on the consolidated List of Questions to be sent to the applicant during the meeting on	21 July 2020
The applicant submitted the responses to the CHMP consolidated List of Questions on	14 August 2020
The Rapporteurs circulated the Joint Assessment Report on the responses to the List of Questions to all CHMP members on	11 September 2020
The CHMP agreed on a list of outstanding issues in writing and/or in an oral explanation to be sent to the applicant on	15 September 2020
The applicant submitted the responses to the CHMP List of Outstanding	22 September 2020

Issues on	
The Rapporteurs circulated the Joint Assessment Report on the responses to the List of Outstanding Issues to all CHMP members on	11 September 2020
The outstanding issues were addressed by the applicant in writing before the CHMP on	12 October 2020
The CHMP, in the light of the overall data submitted and the scientific discussion within the Committee, issued a positive opinion for granting a marketing authorisation to Oxlumo on	15 October 2020

2. Scientific discussion

2.1. Problem statement

2.1.1. Disease or condition

The proposed indication for Oxlumo (lumasiran) is for the treatment of primary hyperoxaluria type 1 (PH1) in all age groups.

PH1 is a rare, progressive, and potentially life-threatening, autosomal recessive inborn error of metabolism, resulting in increased endogenous hepatic production of oxalate, the key toxic metabolite responsible for the clinical manifestations of the disease. In addition to PH1, two other types of primary hyperoxaluria (PH) have been identified that result from different enzymatic defects; PH1 accounts for approximately 80% of PH cases and is the most clinically severe.

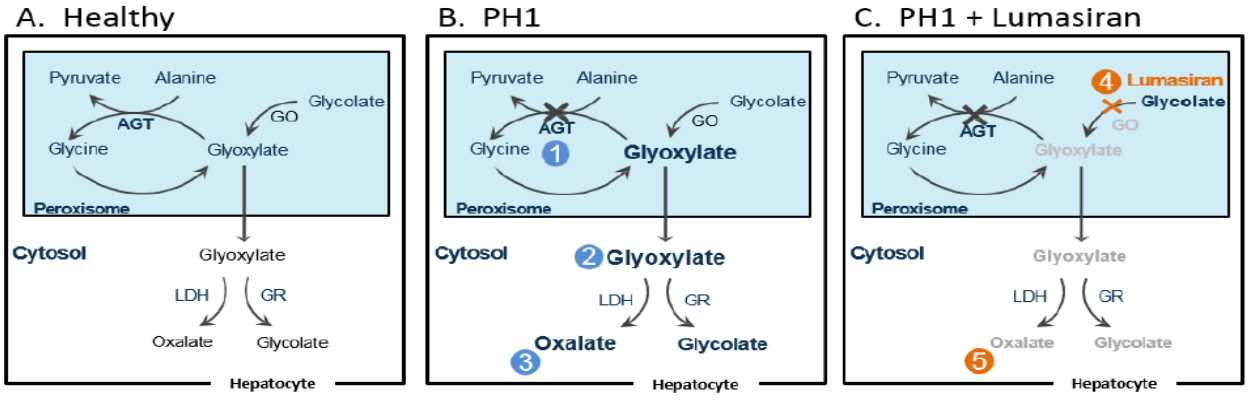
2.1.2. Epidemiology

The incidence of PH1 is estimated to be approximately 1 in 120,000 live births, and the prevalence is 1 to 3 per million in North America and Europe (Cochat and Rumsby 2013; Hopp et.al 2015; Hoppe 2010). At the time of orphan designation in 2017 by the COMP, the decision was based on an estimated prevalence of 0.05 in 10,000 people in the European Union (EU). This was considered equivalent to a total of around 2,600 people (in the EEC region). The disease is more prevalent in areas with founder mutations and where consanguineous marriages are common, including the Middle East and Northern Africa. PH1 usually presents as a paediatric disease, with symptoms first appearing before 6 years of age in more than half of patients. Underdiagnosis is likely common due to the broad phenotypic heterogeneity and known delays in diagnosing patients with PH1.

2.1.3. Biologic features, aetiology and pathogenesis

PH1 results from a mutation of the AGXT (Alanine-glyoxylate aminotransferase) gene leading to low activity with a high variety of the type of underlying mutation, and consequently, also the clinical course of the disease. The reduced or absent AGT activity leads to inability to convert glyoxylate into glycine with the excess substrate then converted into oxalate, leading to a several fold increase in plasma and urine levels of oxalate. **Figure 1** describes the state of a healthy person, the state of patients with PH1, and the proposed Mechanism of Action (MoA) of the compound:

Figure 1: Normal metabolism, defect in PH1, and proposed Mechanism of action of lumasiran.



The deficiency of the liver peroxisomal enzyme AGT, leads to an accumulation of glyoxylate which is converted into oxalate. Hepatic oxalate overproduction, results in calcium crystal formation PH1 frequently have renal stones, nephrocalcinosis, and renal failure that may ultimately result in cardiac, ocular, dermal, and other systemic manifestations of the disease

2.1.4. Clinical presentation, diagnosis and stage/prognosis

Hepatic oxalate overproduction, due to the deficiency of the liver peroxisomal enzyme AGT is converted into oxalate and results in calcium crystal formation, leading to urolithiasis, nephrocalcinosis, renal impairment and end-stage renal disease, as well as systemic oxalosis with manifestations in bone (pain, anaemia, fractures), skin, blood vessels (calcification, pulmonary hypertension), heart (cardiac failure, arrhythmias), eyes, and nerves.

Generally, three types of disease manifestation are described:

An infantile-onset disease with early nephrocalcinosis and rapid progression to renal failure due to increased oxalate load and immature glomerular filtration rate (GFR), a disease type manifesting with recurrent kidney stones in adolescence or early adulthood associated with rapid deterioration of renal function, and a late-onset disease type with only occasional kidney stone passages and, less commonly, ESRD as the first symptom in adulthood.

In addition to these clinical presentations, some patients are diagnosed only after they have progressed to ESRD, undergone kidney transplantation, and rapidly developed oxalate deposition in the transplanted kidney. A minority of patients are identified through familial testing, as full siblings of patients with PH1 each have a 25% risk of also having the disease. Thus, siblings are often screened to detect subclinical or early disease. Regardless of disease manifestation, severity, or age of onset, the pathophysiology of PH1 is the same across the entire patient population.

Published natural history data have shown that, in a cohort of 247 PH1 patients, 24% of patients were in ESRD by age 20 years, 57% by 40 years, and 88% by 60 years (Hopp et.al, 2015).

2.1.5. Management

There is currently no licensed medicinal product available for the treatment of PH1.

The current clinical standard of care uses the following treatments:

- Hyperhydration and inhibitors of crystallization:

Patients with preserved renal function are treated with hyperhydration and crystallization inhibitors order to slow the progression of disease and decrease the incidence of renal stones. The amount of fluid is usually high, and burdensome, especially in the paediatric age. Infants and younger children who are unable to comply may even require a gastrostomy or nasogastric tube for continuous day and night hyperhydration. The treatment with crystallization inhibitors is also not without problems, owing to the taste of these products.

- Pyridoxine

The treatment of pyridoxine (Vit. B6) is based on the fact that pyridoxine is a cofactor of alanine-glyoxylate aminotransferase (AGT), potentially correcting the peroxisome to mitochondrion AGT mistargeting that is associated with some of the mutations in the alanine-glyoxylate aminotransferase (AGXT) gene, which codifies for AGT. Hence, the treatment is only successful in a part of the patients, and the percentage (for successful treatment) as given by the applicant is 5 %.

- Renal Replacement Therapy (RRT) and Transplantation (liver transplantation, or combined kidney and liver transplantation; LTx or combined LTx/KTx)

Patients with deteriorating kidney function regularly require RRT or transplantation. Depending on the overall oxalate level a regular dialysis schedule (3x/week) is often not sufficient, and intensive RRT (6x/week with added peritoneal dialysis) is frequently needed. Some experts also recommend pre-emptive (combined) transplantation therapy, reflecting the serious prognosis of the disease, once kidney function starts to deteriorate. The perioperative mortality in patients undergoing combined KTx/LTx has been estimated to be 17%.

It is acknowledged that the current treatment approaches (e.g. hyperhydration and inhibitors of crystallization) may cause compliance problems, and/or a relevant reduction of quality of life and exposes patients to invasive procedures (gastrostomy and NG-tube placement). The treatment with pyridoxine also concerns only a minority of patients, and clean data on efficacy are not available. RRT and LTx (or LTx/KTx) are of course invasive procedures and reserved for those with end-stage disease. Pre-emptive transplantation exposes patients to a relevant operative risk and subsequent life-long immunosuppression.

About the product

Lumasiran is a double-stranded, small interfering RNA (siRNA) covalently linked to a triantennary N-acetylgalactosamine (GalNAc). Lumasiran uses the RNAi pathway to specifically target the 3' untranslated region of HAO1 mRNA in the liver, thereby preventing the synthesis of the corresponding GO protein. Lumasiran is designed with the GalNAc moiety conjugated to the sense strand of the siRNA to enable selective delivery to the liver via uptake by the asialoglycoprotein receptor (ASGPR). ASGPRs are expressed primarily and abundantly (0.5 to 1 million per cell) on the cell surface of hepatocytes and specifically bind to the glycoproteins with terminal galactose or GalNAc residues.

Binding of the GalNAc ligand of lumasiran and ASGPR triggers receptor-mediated endocytosis of the ligand-receptor complex, resulting in release of the siRNA into the cytoplasm of the hepatocyte. ASGPR is subsequently recycled to the cell surface and is available for successive uptake of circulating GalNAc-conjugated siRNA. The abundance of ASGPR expression on hepatocytes and the ability to mediate multiple rounds of uptake of GalNAc-conjugated siRNA into the hepatocytes makes this a high capacity system that is assumed to be non-saturable at exposures in the therapeutic range. ASGPR capacity is thought to be independent of age and hepatic function.

Upon delivery to the liver, the double-stranded lumasiran siRNA is loaded into the cellular multiprotein enzyme cleavage complex known as the RNA-induced silencing complex (RISC) in the cytosol. Once loaded, the antisense strand (guide strand) of lumasiran binds to the complementary sequence in the HAO1 mRNA. Pairing of HAO1 mRNA with the antisense strand within the RISC/siRNA complex results in specific and highly efficient cleavage of the HAO1 mRNA, thereby preventing the synthesis of the corresponding GO protein, which in turn reduces – by reducing the target substrate of GO, glyoxylate – the production of oxalate in the liver. The underlying enzymatic defect (relating to AGT) is not targeted with the proposed treatment.

Proposed indication

Oxlumo is indicated for the treatment of primary hyperoxaluria type 1 (PH1) in all age groups.

Recommended dose

Oxlumo is administered by subcutaneous injection. The recommended dose of Oxlumo consists of loading doses given once a month for 3 months,

followed by maintenance doses, as shown in **Table 1**. Dosing is based on body weight.

Table 1: Oxlumo weight-based dosing regimen

Body weight	Loading dose	Maintenance dose (the maintenance dose should begin one month after the last loading dose)
less than 10 kg	6 mg/kg once monthly for 3 months	3 mg/kg once monthly
10 kg to less than 20 kg	6 mg/kg once monthly for 3 months	6 mg/kg once every 3 months (quarterly)
20 kg and above	3 mg/kg once monthly for 3 months	3 mg/kg once every 3 months (quarterly)

Type of application and aspects on development

The CHMP agreed to the applicant's request for an accelerated assessment as the product was considered to be of major public health interest. This was based on:

- The disease is a serious and life-threatening condition
- There is an unmet medical need for the treatment of this condition
- Lumasiran is considered suitable to present a major therapeutic innovation for the treatment of the disease and to address the unmet medical need in the condition.

2.2. Quality aspects

2.2.1. Introduction

The finished product is presented as a solution for injection containing 94.5 mg of lumasiran as the active substance. The product contains 100 mg/0.5 ml lumasiran sodium salt equivalent to 94.5 mg/0.5 ml of lumasiran.

Other ingredients are: sodium hydroxide (pH adjustment), phosphoric acid (pH adjustment), water for injections.

The product is available in a glass vial with a fluoropolymer coated rubber stopper and an aluminium overseal with a flip off button as described in section 6.5 of the SmPC. Each vial contains 0.5 mL solution for injection.

2.2.2. Active Substance

General information

Lumasiran is the sodium salt of a chemically synthesised double-stranded oligonucleotide covalently linked to a ligand (referred to as L96) containing three N-acetylgalactosamine (GalNAc) residues. The sense strand (A-131522) and the antisense strand (A-131532) contain 21 and 23 nucleotides, respectively.

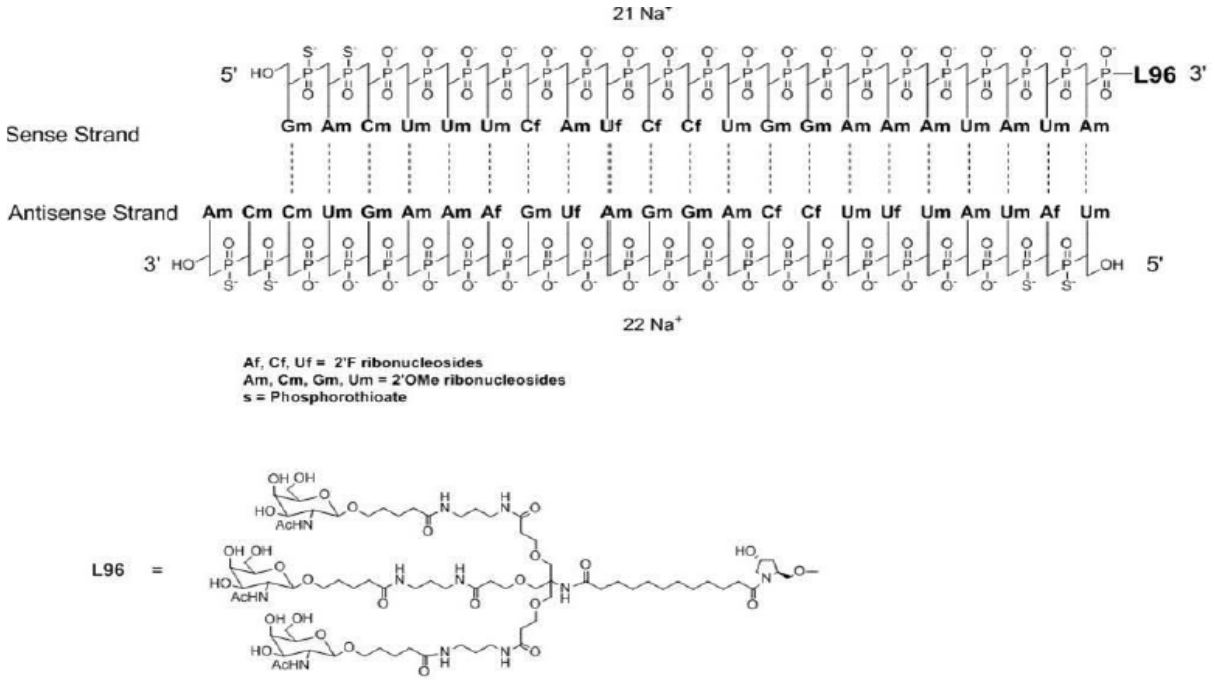
The molecular formula and molecular weight of lumasiran sodium are summarised in **Table 2**.

Table 2: Active substance molecular formula and molecular weight

	Drug Substance	Sense Strand (A-131522)	Antisense Strand (A-131532)
Molecular formula of the free acid	$C_{530}H_{712}F_{10}N_{173}O_{320}P_{43}S_6$	$C_{294}H_{414}F_4N_{87}O_{172}P_{21}S_2$	$C_{236}H_{298}F_6N_{86}O_{148}P_{22}S_4$
Molecular formula of the sodium salt	$C_{530}H_{669}F_{10}N_{173}O_{320}P_{43}S_6Na_{43}$	$C_{294}H_{393}F_4N_{87}O_{172}P_{21}S_2Na_{21}$	$C_{236}H_{276}F_6N_{86}O_{148}P_{22}S_4Na_{22}$
Molecular weight of the free acid	16340.54 Da	8709.49 Da	7631.05 Da
Molecular weight of the sodium salt	17285.76 Da	9171.10 Da	8114.65 Da

The 3'-end of the sense strand is conjugated to the triantennary GalNAc moiety. The sense strand (A-131522) contains two phosphorothioate linkages at the 5' end. The antisense strand (A-131532) contains four phosphorothioate linkages - two at the 3' end and two at the 5' end. The 21 nucleotides of the sense strand hybridise with the complementary 21 nucleotides of the antisense strand, thus forming 21 nucleotide base pairs and a two-base overhang at the 3'-end of the antisense strand. The active substance structure is illustrated in **Figure 2**.

Figure 2. Active substance structure



Abbreviations: Af=adenine 2′-F ribonucleoside; Cf=cytosine 2′-F ribonucleoside; Uf=uracil 2′-F ribonucleoside; Am=adenine 2′-OMe ribonucleoside; Cm=Cytosine 2′-OMe ribonucleoside; Gm=guanine 2′-OMe ribonucleoside; Um=uracil 2′-OMe ribonucleoside

The active substance is a white to pale yellow powder. It is hygroscopic in nature. It has solubility of NLT 387 mg/ml in water which is considered be "freely soluble".

The chemical structure of lumasiran sodium active substance was elucidated and characterised by a combination of orthogonal analytical techniques. Characterisation data are provided on lumasiran active substance and on the single strand (sense and antisense) intermediates.

The structure elucidation data are acceptable to confirm the correct sequences of the single strand intermediates and the duplex structure of the lumasiran active substance.

Lumasiran exhibits stereoisomerism due to the presence of a number of chiral centres.

All the pentose moieties of the nucleotides in the lumasiran active substance are in the naturally occurring D-ribose form. The chirality of the D-ribose is maintained during the synthesis of the modified conformation; RNA molecules adopt the classic A-form as demonstrated by the spectrum of Circular Dichroism.

The phosphodiester (PO) linkages of double-stranded siRNAs require protection against cleavage by exonucleases. This is achieved by replacement of one of the non-bridging oxygen atoms in one or more PO linkages with sulfur. The resulting phosphorothioate group (PS) is chiral, with either R_p or S_p absolute configuration at the phosphorus atom. Thus, the solid-phase synthesis of PS modified oligonucleotides leads to a mixture of R_p/S_p isomers, which results in a population of diastereoisomers.

The antisense strand contains four PS modifications, with two on the 5'end and two at the 3'end, resulting in the formation of sixteen (2^4=16) diastereomers. The sense strand contains two PS modification on the 5'end, corresponding to four (2^2=4) diastereomers.

The applicant provided additional information on the three-dimensional (3D) structure of lumasiran. The 3D structure of the active substance as well as of the individual single strands is the result of the stereochemistry showed by the individual nucleic acid monomers, nucleosides and phosphoramidites as well as of the two chiral raw materials used in the synthesis and the reactions involved in the active substance synthesis. It has been demonstrated that overall the active substance synthesis produce consistently phosphorathioate diesters ratio.

The circular dichroism spectra of lumasiran active substance indicates that exists in solution in an A-form, as expected. The 3D structure of lumasiran active substance was also confirmed. Data shows consistency of the stereochemistry of the lumasiran active independently of the manufacturing site or scale of the synthesis, as expected from a solid-phase based synthesis. This consistency has also been demonstrated in both the sense and antisense strands produced with different scales. The information on the three-dimensional structure and stereochemistry of the active substance is considered adequate.

Manufacture, characterisation and process controls

The lumasiran active substance is manufactured in seven main steps using well defined starting materials with acceptable specifications.

Step 1 to Step 5 consist of: the synthesis of the single-strand oligonucleotide by solid-phase phosphoramidite synthesis (Step 1), cleavage and deprotection (C&D) (Step 2), crude ultrafiltration (UF) (Step 3), purification by anion exchange chromatography (Step 4), and a final UF (Step 5).

Each strand is individually purified and concentrated in Step 3 through Step 5. After the final UF, the two individual strands are annealed (Step 6) to form the duplex, which is then lyophilised and packaged in Step 7 to produce lumasiran sodium active substance.

Protected phosphoramidites are considered suitable starting materials for synthetic oligonucleotides. An appropriate justification for the classification of phosphoramidites as starting materials has been provided. Detailed information on the impurity profiles of the phosphoramidite starting materials has been provided. The proposed phosphoramidite specifications are acceptable.

In summary, the selection of all starting materials has been carried out according to the principles of ICH Q11, i.e. they all have defined physical and chemical properties and structure; they are incorporated as a structural fragment into the structure of the lumasiran; they are purchased from commercial suppliers; and they are controlled with specifications to ensure lumasiran active substance quality. The selection of the starting materials is considered adequately justified taking into account also the manufacturing process which includes numerous cycles and purification steps, which ensure the proper purge of starting materials' potential related and degradation impurities preventing their carry over to the single strand intermediates or even to the final lumasiran active substance. Impurities of starting materials have been identified and the proposed acceptance limits provided are considered acceptable. Suppliers of the starting materials are mentioned in the dossier. It has been confirmed that any addition of alternative suppliers for the starting materials will be made by the submission of a variation.

A list of all reagents, solvents, and auxiliary materials used in the active substance manufacturing process, with relevant specifications, has been provided.

A summary of the relevant quality attributes, their acceptance criteria, and location of controls for lumasiran active substance is provided. Process characterization activities to develop a control strategy for process performance qualification and commercial manufacturing have been sufficient described. Risk assessments were performed utilizing historical process understanding from the applicant's manufacturing platform across multiple Alnylam products. Process parameter target set points, normal operating ranges (NOR) and proven acceptable ranges (PAR) were identified using design of experiments (DOE) and one factor at a time (OFAT) approaches. For individual unit operations, the desired outcomes from characterization studies were minimization of impurities and maximization of purity and yield.

The critical process steps and associated critical process parameters (CPPs) for each unit operation and the non-critical process parameters of the manufacturing process are summarised, along with details of the criticality evaluation of process steps. The defined proven acceptable ranges (PARs) are acceptable and have been justified in the process development studies. The in-process tests performed at each step are described and the proposed limits are considered acceptable. In-process hold times have been sufficiently investigated.

The quality of double-stranded oligonucleotides is pre-determined by the quality of the single strand precursors. Therefore, the control of these intermediates by adequate specifications is essential since some of the impurities can only be controlled at the level of the single strands.

Identity by molecular weight and retention time, and more importantly, by sequencing is performed for the sense and antisense strand. Consequently, the sequence is proven also for the resulting duplex. Purity of the sense and the antisense strand is determined by two techniques, AX-HPLC and IPRP-HPLC. The proposed tests on the single strands are acceptable. The specified impurities for the single strands are grouped based on their retention times and controlled within defined limits for each group. This is acceptable as it is commonly applied for synthetic oligonucleotide products.

The single strand in-process control limits for sense and antisense strands are currently acceptable. The CHMP recommends (and applicant commits) to re-evaluate these limits post-approval when data from additional 10 commercial batches becomes available (REC1).

A three stage lifecycle approach is applied for process validation of the active substance manufacturing process.

The commercial manufacturing process for the active substance was developed in parallel with the clinical development program. Information on batch history has been provided in the dossier. All changes are described in detail and the influence on active substance quality has been in general sufficiently investigated and described.

The characterisation of the active substance and its impurities are in accordance with the EU guideline on chemistry of new active substances. Potential and actual impurities were well discussed with regards to their origin and characterised.

As per ICH Q3A guideline, impurities are classified into organic impurities, inorganic impurities and residual solvents. The organic impurities are further classified into process-related impurities and product-related impurities.

Product-related oligonucleotide impurities are formed during the manufacturing process or during storage, including degradants. These impurities are controlled in the manufacturing process and at long term storage conditions by two orthogonal HPLC techniques (AX-HPLC and IPRP-HPLC) in the single strand intermediates and the final active substance.

Typical impurities are deletion (shortmers) and addition (longmers) impurities, partially deprotected oligonucleotide chains that are not fully deprotected or improperly deprotected during manufacture, phosphodiester (P=O) impurities, where a phosphodiester replaces the thiophosphate (P=S) in the sense and antisense strands, impurities carried over from parent starting material impurities and in particular those associated with the triantennary N-acetyl galactosamine (GalNAc) portion of the sense strand.

In general, a good understanding of the impurity profile in the single strand intermediates and the final active substance has been demonstrated. Numerous impurities have been identified. Degradation pathways and the impact of annealing on degradation have been in general sufficiently investigated and discussed. Impurity monitoring is performed on impurities grouped by adjusted RRT ranges which is acceptable for synthetic oligonucleotides with an extremely complex impurity profile. The qualification of impurities has been sufficiently described. Process related organic impurities are low molecular weight organic impurities such as residual starting material, reagents and by-products from the manufacturing process. These impurities have been discussed and are considered removed due to extensive washing, chromatographic and ultrafiltration steps.

There are genotoxic substances formed during the manufacture of both single strands. The evaluation of the presence of these genotoxic materials in the active substance is performed in accordance with the principles stipulated in the ICH M7 guideline. Residual solvents have been adequately addressed and batch analysis data have been provided. Inorganic impurities have been sufficiently addressed. The active substance is packaged in a gamma irradiated, high density polyethylene (HDPE) bottle with a polypropylene (PP) screw-top and a secondary foil laminate bag. The packaging materials complies with Regulation (EC) No. 1935/2004, Regulation (EC) No. 2023/2006 (GMP) and EC Regulation (EC) No. 10/2011 as amended.

Specification

The active substance specification includes tests for appearance (visual), identity by duplex retention time (IPRP-HPLC-UV), identity by single strand MW (IPRP-HPLC ESI MS), identity by Tm (spectrophotometry), identity of single strands by sequence (MS-MS fragmentation), sodium content (flame AAS), purity (non-denaturing IPRP-HPLC UV), purity

(denaturing IPRP-HPLC UV), assay (UV absorption), pH (Ph. Eur.), water content (KF), elemental impurities (ICP-MS), acetonitrile (headspace GC-FID), bacterial endotoxins (Ph. Eur.) and bioburden (Ph. Eur.).

The chosen specification attributes are appropriate. Three different purity methods are employed to control the purity of the duplex and impurities resulting from the single strands. The concept of grouping of impurities as proposed in the specification is acceptable. Identity of the single strands is determined by sequencing of the single strands as intermediates during the synthesis.

The acceptance criteria in the active substance specification are acceptable. The CHMP recommends (and applicant commits) to re-evaluate the active substance acceptance criteria post-approval when data from additional 10 commercial batches becomes available (REC2).

The analytical methods used have been adequately described and (non-compendial methods) appropriately validated in accordance with the ICH guidelines. Satisfactory information regarding the reference standards used for impurities testing has been presented.

Batch analysis data of the active substance are provided. The results are within the specifications and consistent from batch to batch.

Stability

Stability data from ten batches of active substance from the proposed manufacturer stored in the intended commercial package for up to 48 months under long term conditions (-20°C ± 5°C) and for up to six months under accelerated conditions (25°C ± 2°C / 60 ± 5%) according to the ICH guidelines were provided.

The quality attributes tested under the storage conditions were appearance, assay, water content, purity by non-denaturing IPRP HPLC, purity by denaturing AX HPLC and purity by denaturing IPRP HPLC.

No negative trends have been observed at long term and accelerated conditions with the exception of two supportive batches where increased water content and results for total impurities above the specification limits have been observed at accelerated conditions. However, these supportive batches were by design manufactured with higher impurity levels.

Forced degradation studies have been performed. The stress conditions studied were temperature, acid, base, oxidation and light. Similar degradation was observed. The stability-indicating properties of the three purity methods have been demonstrated.

Based on the available data, a retest period for the active substance of 36 months when stored at the recommended long-term storage condition of -20°C or below is acceptable.

2.2.3. Finished Medicinal Product

Description of the product and Pharmaceutical development

The finished product is presented as solution for injection containing 94.5 mg of lumasiran as the active substance. The product contains 100 mg/0.5 ml lumasiran sodium salt equivalent to 94.5 mg/0.5 ml of lumasiran (i.e. 200 mg/ml lumasiran sodium equivalent to 189 mg/mL lumasiran).

The finished product is a sterile, preservative-free, colorless to yellow solution intended for single use subcutaneous injection. It is supplied in a single-use Type I clear glass vial with a fluoropolymer-coated bromobutyl rubber stopper and an aluminum overseal with a flip-off button. Each vial contains 0.5 ml nominal volume of lumasiran solution (containing 94.5 mg of lumasiran).

The finished product is a solution of the active substance in water for injection with a target pH of 7.0 adjusted with sodium hydroxide or phosphoric acid in a vial. No further excipients are used. All excipients are well known pharmaceutical ingredients and their quality is compliant with Ph. Eur standards. There are no novel excipients used in the finished product formulation. The list of excipients is included in section 6.1 of the SmPC and in paragraph 2.2.1 of this report.

The pharmaceutical development followed a systematic approach whereby the formulation has been developed with respect to concentration to volume ratio, osmolality, viscosity (syringeability) and pH to accommodate subcutaneous application of the finished product. The quality target product profile (QTPP) was presented. The active substance quality attributes that may impact the finished product critical quality attributes were clearly identified. The formulation has been consistent throughout development from non-clinical through to the PPQ lots.

The proposed fill volume has been sufficiently justified. The manufacturing process has also been systematically developed starting with clinical batches. Manufacturing process development followed a risk-based approach combined with process development studies. Critical and non-critical quality attributes are defined. The results form the basis for the overall control strategy. The manufacturing process parameters, associated targets, and NORs/PARs defined based on the development work have been defined in the dossier section P.3.3.

The use of sterile filtration as the sterilization method for the finished product is justified with literature reference, i.e. that dry and steam heat have been demonstrated to impact the impurities profile of chemically modified RNA and that γ-irradiation has been shown to cause oxidative degradation in nucleotides. Furthermore, the applicant explains that temperatures required for thermal sterilisation exceed the melting temperature of siRNA duplex which would result in denaturation of the active substance. The applicant's justification was accepted in line with the principles of the the EMA Guideline on the sterilisation of the medicinal product, active substance, excipient and primary container (EMA/CHMP/CVMP/QWP/850374/2015).

As the product is proposed for use in children, the suitability of the formulation for use in children was addressed as part of the development, in accordance with the guideline on Pharmaceutical development of medicines for paediatric use (EMA/CHMP/QWP/805880/2012 Rev. 2). The information provided on injection volume, dosing accuracy and patient acceptability is considered acceptable from a quality point of view.

The primary container closure system consists of a Type I clear glass vial with a gray bromobutyl rubber stopper containing a fluorinated polymer barrier film on the face/plug and a B2 silicone lubricity coating. The material complies with Ph. Eur. and EC requirements. The suitability of the container closure system was evaluated with respect to protection, safety, and compatibility. Protection was demonstrated by container closure integrity testing. Furthermore, adsorption and delamination of the vials are not issues. Based on simulation test results a toxicological evaluation of the leachables from the container closure system has been presented. The used type I clear glass vial and the bromobutyl rubber stopper 4023/50 with fluoropolymer-coated contact surface are not considered to pose a safety risk. The choice of the container closure system has been validated by stability data and is adequate for the intended use of the product.

Compatibility by in-use studies has been demonstrated for commercial 1 mL and 3 mL polycarbonate or polypropylene syringes and needles of bore sizes 21 G and 30 G. The respective syringe samples were stored at 25°C for 8 hours and at 2°C to 8°C for 48 hours. Purity testing by AX-HPLC UV and assay by UV spectrophotometry revealed no significant differences of the syringe samples to the control vials.

Manufacture of the product and process controls

The finished product is manufactured and primary packaged at Vetter Pharma-Fertigung GmbH & Co. KG, Langenargen, Germany. The manufacturing process consists of four main steps: formulation, bioburden reduction filtration, sterile filtration and aseptic filling. The process is considered to be a non-standard manufacturing process.

The manufacturing process is sufficiently described including process parameters with set-points, NORs and PARs as appropriate, as well as hold and processing times. The in-process controls (IPCs) are adequate for this type of manufacturing process and pharmaceutical form.

Major steps of the manufacturing process have been validated by a number of studies. Process validation data covers the proposed maximum batch size. It has been demonstrated that the manufacturing process is capable of producing the finished product of intended quality in a reproducible manner.

To further guarantee microbiological safety of the finished product the applicant was requested to include requirements for microbial contamination and bacterial endotoxins in the specifications of all excipients. The applicant has adopted the requested change for water for injection specification and justified not including microbial testing for sodium hydroxide and phosphoric acid. The justification is considered acceptable.

Product specification

The finished product specifications include appropriate tests for this kind of dosage form; appearance (Ph. Eur.), identity by duplex retention time (IPRP-HPLC UV), identity by single strand molecular mass (IPRP-HPLC MS), purity (IPRP-HPLC UV, non-denaturing), assay (UV spectrophotometry), purity (AX-HPLC UV denaturing), purity (IPRP-HPLC UV, denaturing), pH (Ph. Eur.), osmolality (Ph. Eur.), particulate matter (Ph. Eur.), bacterial endotoxins (Ph. Eur.), sterility (Ph. Eur), volume in container (Ph. Eur.) and container closure integrity (oxygen headspace).

The finished product is released on the market based on the above release specifications, through traditional final product release testing. The analytical methods used have been adequately described and appropriately validated in accordance with the ICH guidelines. Satisfactory information regarding the reference standards used for and impurities testing has been presented. The reference standard used in the testing finished product is the same as that used for active substance.

The proposed limits for impurities are acceptable. The CHMP recommends (and applicant commits) to re-evaluate and further tighten the finished product specification limits as applicable post-approval when data from additional 10 commercial batches are available (REC3).The potential presence of elemental impurities in the finished product has been assessed on a risk-based approach in line with the ICH Q3D Guideline for Elemental Impurities. Additional controls in the finished product specification are not necessary as the results are consistently below the control thresholds of 30% of the established PDEs.

The applicant originally provided in the active substance documentation a brief statement that no risk of presence of N-nitrosamines impurities is considered in active substance or finished product. The arguments and justification provided were considered insufficient and resulted in a major objection to request a full risk evaluation concerning the presence of nitrosamine impurities also in the finished product and applying the principles outlined in the notice "Information on nitrosamines for marketing authorisation holders (EMA/189634/2019)". The applicant has submitted the requested risk assessment concerning nitrosamine impurities in both active substance and finished product concluding that no risk of the presence of N-nitrosamine impurities was identified in either. The response provided was considered acceptable and the major objection was resolved.

Batch analysis data are provided. All data meet the commercial acceptance criteria and demonstrate batch to batch consistency confirming the consistency of the manufacturing process and its ability to manufacture to the intended product specification.

Stability of the product

Stability data from ten batches of finished product stored for up to 48, 36 and 12 months respectively under long term conditions (2-8°C, 25°C/60%RH, 30°C/75%RH) and for up to 6 months under accelerated conditions (40 ºC / 75% RH) according to the ICH guidelines were provided. The finished product batches are representative of those proposed for marketing and were packed in the primary packaging proposed for marketing.

Further results for supporting stability batches have been included, i.e. for two batches produced with exaggerated impurity profiles for use in nonclinical studies. An additional small-scale batch was used solely for thermal stress, cyclic stress and photostability studies. Results obtained during the assessment of thermal and cyclic stress also show resistance of the finished product to thermal degradation under the parameters studied (i.e., up to 14-days at 60°C and freeze-thaw cycling from -20°C to 60°C with cumulative exposure periods of 12-days under each condition). Direct exposure of the finished product to an ICH Guideline Q1B 1x light exposure equivalent induced only minor change (<5%); further direct exposure to light (up to an 8x exposure equivalent), induced a concomitant decrease in purity as a function of increasing light exposure.

The stability batches were tested in line with the shelf-life specifications The currently available stability results at long-term and at accelerated conditions show no obvious trends and are within specification limits, except for one batch stored at 40°C/75%RH where an out of specification (OOS) result for pH occurred. The test on pH gave results of 8.1 after 3 months, after 4 and 5 months the values found are both at pH 8.0. The specified limits for pH are 6.0-8.0.

Based on available stability data, the proposed shelf-life of 3 years and the storage conditions "Once the vial is opened, the medicinal product should be used immediately", "Do not store above 30°C" and "Keep vial in the outer carton to protect from light" as presented in sections 6.3 and 6.4 of the SmPC are acceptable.

Post approval change management protocols

Two post approval change management protocols (PACMP) are included to introduce an additional active substance manufacturer and an additional finished product manufacturer post approval.

Adventitious agents

No excipients derived from animal or human origin have been used. There is no risk of potential contamination with adventitious agents of viral or non-viral origin associated with the active substance or finished product.

2.2.4. Discussion on chemical, pharmaceutical and biological aspects

Information on development, manufacture and control of the active substance and finished product has been presented in a satisfactory manner.

A major objection raised requesting an updated risk assessment for possible nitrosamines impurities has been resolved (no risk for nitrosamines identified). Two post approval change management protocols (PACMP) are included to introduce an additional active substance manufacturer and an additional finished product manufacturer post approval. There are three quality recommendations to

update single strand in-process control limits, active substance specifications and finished product specifications when further commercial batch data is available.

The results of tests carried out indicate consistency and uniformity of important product quality characteristics, and these in turn lead to the conclusion that the product should have a satisfactory and uniform performance in clinical use.

2.2.5. Conclusions on the chemical, pharmaceutical and biological aspects

The quality of this product is considered to be acceptable when used in accordance with the conditions defined in the SmPC. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way.

2.2.6. Recommendation(s) for future quality development

In the context of the obligation of the MAHs to take due account of technical and scientific progress, the CHMP recommends the following points for investigation:

1. To re-evaluate and tighten the single strand in-process control limits as applicable when data from additional 10 commercial batches are available.
2. To re-evaluate and tighten the active substance specification limits as applicable when data from additional 10 commercial batches are available.
3. To re-evaluate and tighten the finished product specification limits as applicable when data from additional 10 commercial batches are available.

2.3. Non-clinical aspects

2.3.1. Introduction

The pharmacology, safety pharmacology, pharmacokinetics, and toxicology of lumasiran were evaluated in a series of *in vitro* and *in vivo* nonclinical studies. According to the applicant, all pivotal studies were carried out in accordance with good laboratory practice (GLP).

2.3.2. Pharmacology

Primary pharmacodynamic studies

In vitro studies

BIO15032: *In Vitro* Identification of HAO1-GalNAc Candidates in Support of Lead Selection for ALN-GO1 [Study BIO15032; non-GLP]

Forty-nine candidate siRNAs targeting hydroxyacid oxidase 1 (*HAO1)* were identified using custom bioinformatics tools complementary to the human *HAO1* transcript as well as the ortholog mRNA sequences from cynomolgus monkey, mice, and rat. *In vitro* activity of these chemically modified siRNAs with triantennary GalNAc ligands conjugated to the 3' end of the sense strand was evaluated by

by transfection in primary Cynomolgus monkey hepatocytes (PCH) and determination of the extent of inhibition of *HAO1* mRNA following transfection.

In a second round, chemically modified analogues of 2 siRNAs identified in the first round were designed, synthesised and tested for inhibition of *HAO1* mRNA. The chemical modifications introduced were expected to increase siRNA stability (and hence durability). The study resulted in identification of a siRNA (AD-65585) which was later termed lumasiran with an IC_{50} value for inhibition of *HAO1* mRNA of 0.01 nM in primary monkey hepatocytes.

In-vivo studies

Pharmacodynamic Evaluation of ALN-65585 in Wild-Type Mice Following a Single Subcutaneous Injection [Study BIO15016; non-GLP]

Lumasiran pharmacology was evaluated in female WT mice by quantifying liver *HAO1* mRNA and serum glycolate levels. A single SC dose of lumasiran in female WT mice at doses between 0.1 and 10 mg/kg bw resulted in a dose-dependent reduction of liver *HAO1* mRNA with a dose of 10 mg/kg bw resulting in approximately 90% reduction (ED_{90}on Day 10. The effective dose producing a 50% reduction (ED_{50}) in *HAO1* mRNA in the mouse was estimated to be 0.3 mg/kg bw. Serum glycolate levels increased in a dose-dependent manner with a maximum level approximately 4-fold above baseline levels at 3 mg/kg bw.

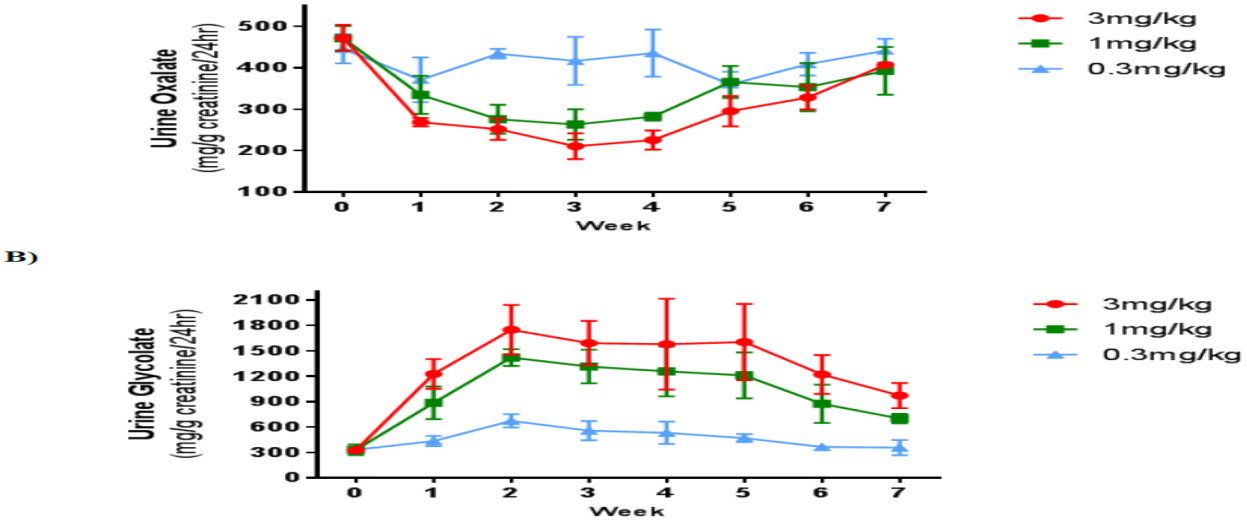
Evaluation of ALN-65585 Duration in Wild-Type Mice Following a Single Subcutaneous Injection [Study BIO15022; non-GLP]

A single dose of 3 mg/kg bw of lumasiran was administered SC to female WT mice. Animals were euthanised between Days 3 and 84, after which liver samples were obtained for evaluation of the duration of *HAO1* reduction. A single SC dose of lumasiran in mice at 3 mg/kg bw resulted in ≥70% mRNA reduction for approximately 6 weeks, after which mRNA levels recovered towards baseline levels by 12 weeks post dose.

Pharmacologic Evaluation of ALN-65585 in a Mouse Model of Primary Hyperoxaluria Type I Following a Single Subcutaneous Injection [Study BIO15028; non-GLP]

Male alanine-glyoxylate aminotransferase deficient mice (AGXT -/-) lacking liver AGXT mRNA and protein received a single SC dose of PBS or lumasiran. Urinary oxalate and glycolate levels showed dose-dependent reductions and increases respectively after a single dose of lumasiran (**Figure 3**).

Figure 3. Effects on urinary oxalate and glycolate in the AGXT deficient mouse after a single dose of lumasiran



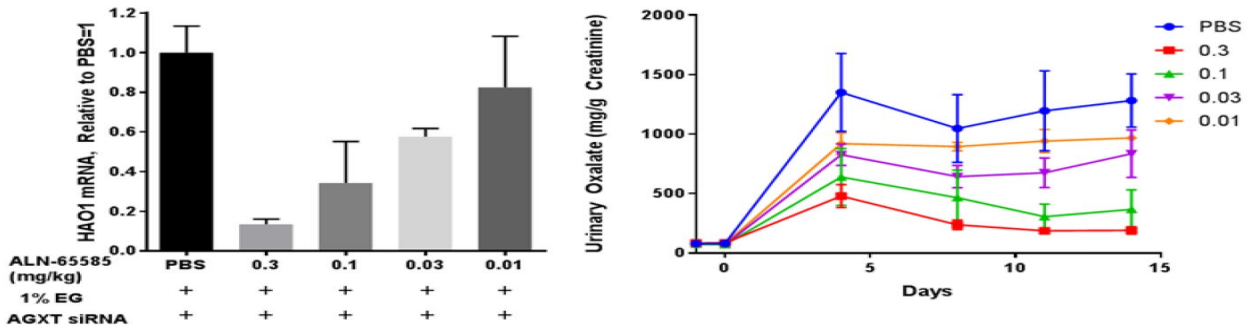
Pharmacodynamic Evaluation of ALN-65585 in Wild-Type Rats Following a Single Subcutaneous Injection [Study BIO15019; non-GLP]

A single SC administration of PBS (control) or lumasiran at doses between 0.1, and 10 mg/kg bw to male WT Sprague Dawley rats resulted in a dose-dependent reduction of *HAO1* mRNA (with doses ≥3 mg/kg bw resulting in ED_{90} reduction. The ED_{50} for *HAO1* in WT rats was estimated to be 0.3 mg/kg bw and the 0.1 mg/kg bw dose had minimal effect on *HAO1* mRNA reduction. Serum glycolate levels increased in a dose-dependent manner with a maximum level approximately 8-fold above baseline after a single 10 mg/kg bw dose of lumasiran.

Pharmacologic Evaluation of ALN-65585 in a Rat Model of Primary Hyperoxaluria Type I Following a Single Subcutaneous Injection [Study BIO15027; non-GLP]

Sprague Dawley rats received weekly 1 mg/kg bw doses of AGXT siRNA in a lipid nanoparticle to decrease the levels of *AGXT* in the liver and mimic the genetic deficiency in patients with PH1. Liver *HAO1* mRNA (obtained at sacrifice at Day 14) and 24-hour urinary oxalate were quantified to determine the degree of *HAO1* reduction required for maximal oxalate reduction (**Figure 4**).

Figure 4. Dose-dependent reductions in HAO1 mRNA and urinary oxalate in a PH1-induced rat model after a single dose of lumasiran



Pharmacologic Evaluation of ALN-65585 in a Rat Model of Primary Hyperoxaluria Type I Following Multiple Subcutaneous Injections [Study BIO15030; non-GLP]

Male wild-type (Sprague Dawley) rats were injected on Day 0, 7, 14, and 21 IV with AGXT siRNA (AD-63102) formulated in an AF-011 lipid nanoparticle and SC with lumasiran at doses of 0.3, 1, or 3 mg/kg bw or PBS.

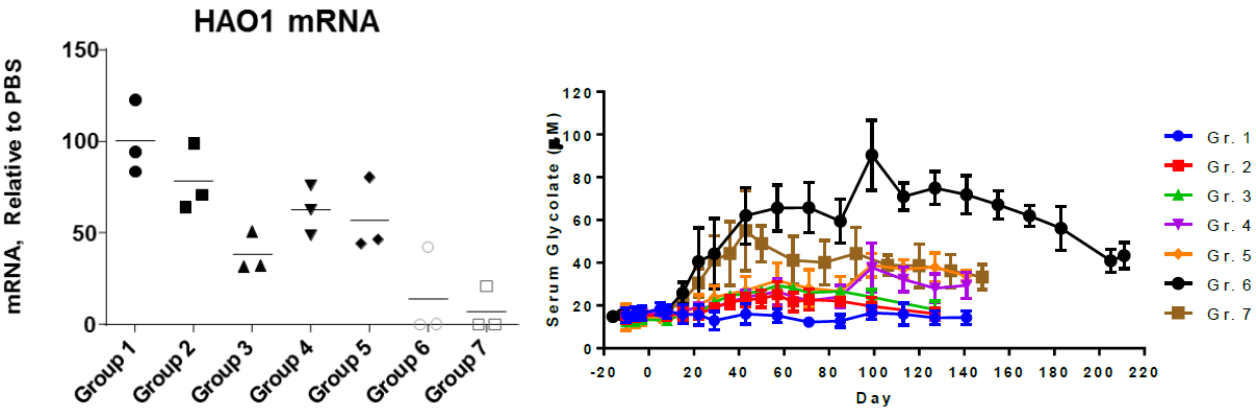
Four weekly SC doses of lumasiran resulted in sustained urinary oxalate reductions in all dose groups. On Day 28, after repeat dosing of lumasiran, all groups showed ≥95% mRNA reduction and >85% urinary oxalate reduction.

Pharmacodynamic Evaluation of AD-65585 Following Subcutaneous Injection of Male Cynomolgus Monkeys [Study BIO15029; non-GLP]

Study animals received 6, once monthly doses of phosphate-buffered saline (PBS) (Group 1), 8 once weekly (QW) doses of 0.25 or 1 mg/kg bw of lumasiran (Groups 2 and 3), 6 once monthly doses of 1, 2, or 4 mg/kg bw of lumasiran (Groups 4, 5, and 6), or 4 once weekly doses of 2 mg/kg bw followed by 5 once monthly doses of 1 mg/kg bw of lumasiran (Group 7).

HAO1 mRNA levels were quantified from liver biopsies. Glycolate levels were quantified in serum and urine, and oxalate levels were quantified in urine.

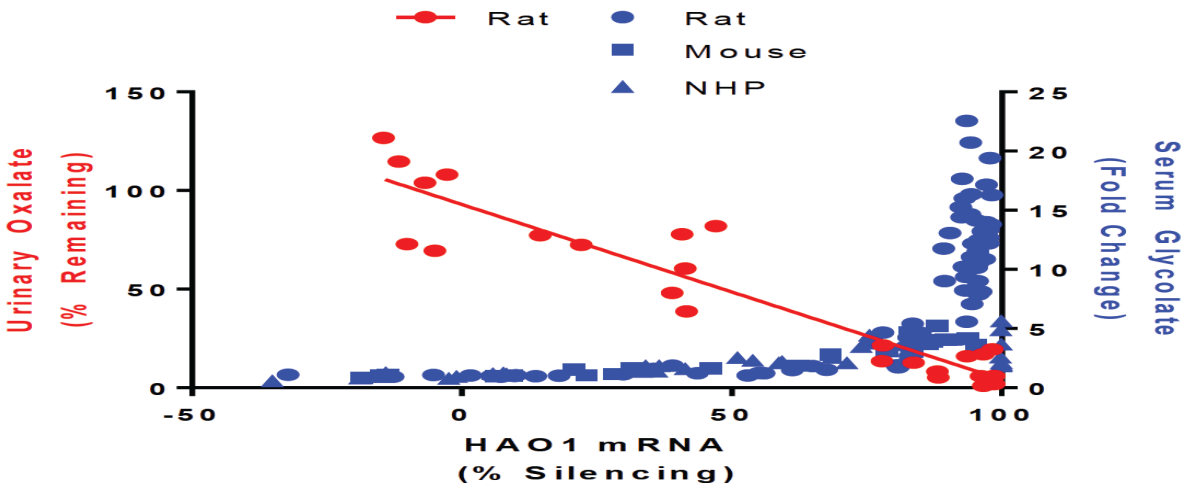
Figure 5. Effects on HAO1 mRNA and serum glycolate in monkeys after repeat dosing of lumasiran



Relationship of HAO1 Silencing to Oxalate Metabolism in Rodents and Nonhuman Primates [Study BIO16013; non-GLP]

The study re-evaluated data generated in studies BIO15016, BIO15019, BIO15027, BIO15029 and BIO15030. Serum glycolate or urine oxalate and liver HAO1 mRNA data was plotted on one graph to show the relationship of HAO1 mRNA silencing to oxalate metabolism (**Figure 6**).

Figure 6. Relationship of HAO1 mRNA reduction to glycolate and oxalate metabolism in rodents and monkeys



Secondary pharmacodynamic studies

Mining of a public dbSNP database (Study BIO15020) demonstrated nearly 100% sequence conservation for the lumasiran target site in the overwhelming majority of humans whose *HAO1* gene has been sequenced to date.

In vitro analysis of mRNA reduction by lumasiran was conducted on the potential off-target transcripts identified from an *in silico* analysis comparing the sequence of only the antisense strand against the human transcriptome (Study BIO15033). Of the 14 transcripts with the greatest potential for off-target effects expression was not detected in Hep3B cells for three of these, consistent with the known lack of liver expression for these transcripts. Of the 11 transcripts that were expressed in these liver cells, no appreciable inhibition of 10 of these target genes was observed after transfection of lumasiran at concentrations up to 10 nM. The one potential off-target gene dimethylglycine dehydrogenase (*DMGDH*, NR104002) that did show modest inhibition at the highest concentration of lumasiran in these cells was then directly compared for potency to *HAO1* inhibition in transfected COS-7 cells, a cell line that abundantly expresses both *HAO1* and *DMGDH*. The results suggest there is a >1000-fold difference between the on-target reduction of *HAO1* by lumasiran and the off-target reduction of any of the predicted off-target transcripts.

Safety pharmacology programme

Cardiovascular and respiratory systems

Telemeterised male Cynomolgus monkeys were given SC injections of the control, 10 or 100 mg/kg bw of lumasiran on Days 1, 8, 15, and 22 in a parallel-dosing design GO1-GLP15-014.

All animals survived until study termination and were returned to the stock colony on Day 25 of the dosing phase. No abnormal clinical observations were attributed to administration of lumasiran. At doses of 10 or 100 mg/kg bw, lumasiran had no effect on qualitative ECG findings, quantitative ECG or hemodynamic parameters, respiration rate, or body temperature following repeat dosing and the NOEL was ≥100 mg/kg bw, the highest dose tested.

An *in vitro* human ether-à-go-go-related gene (hERG) assay was not conducted based on the molecular size (approximately 16 kDa), physical chemical properties of lumasiran and negligible

distribution to the heart. This was confirmed with another GalNAc-conjugated RNAi duplex of similar size and structure to lumasiran. targeting a mRNA sequence unrelated to *HAO1* (data not shown).

Central nervous system

CNS safety was investigated as part of repeated dose general toxicity studies GO1-GLP15-009 (8-week) and GO1-GLP15-036 (36 weeks) in Cynomolgus monkeys. No lumasiran-associated neurological observations were noted up to the highest investigated doses of 100 mg/kg bw and 300 mg/kg bw in the 8- and 36-week repeat-dose GLP toxicity studies.

Pharmacodynamic drug interactions

No dedicated studies to evaluate pharmacodynamic drug interactions were submitted as the potential for pharmacodynamic drug interactions with lumasiran in humans is expected to be low given that there are no other *HAO1* suppressive agents or other agents that affect *HAO1* production.

2.3.3. Pharmacokinetics

Absorption

Absorption was investigated as non-GLP studies in rats and cynomolgus monkeys and in GLP-compliant toxicokinetic studies in mice, rats, rabbits and monkeys (described in the toxicology section of this report).

Non GLP-compliant studies in rats and monkeys.

Study GO1-DSM15-022

Plasma lumasiran levels were determined after single intravenous and subcutaneous and after multiple (once weekly for 8 weeks or once monthly for 3 month) subcutaneous administration in male and female rats by the LC-TOF-MS method.

Following a single lumasiran dose of 5 mg/kg IV bolus administration in rats, the mean C_{max} was 25.0 µg/mL with a mean area under the concentration versus time curve from the time of dosing to the last measurable concentration (AUC_{last}) value of 7.95 h*µg/mL. Elimination was rapid with an estimated $t_{1/2}$ of 0.4 hours. The mean total clearance (CL) and volume of distribution at steady state (V_{ss}) values were 646 mL/h/kg and 332 mL/kg, respectively. The applicant comments that, this indicates a moderate distribution of lumasiran beyond the vasculature. For doses 1 to 10 mg/kg, plasma exposure of lumasiran (C_{max} and AUC) increased approximately dose proportionally over the dose range evaluated. The apparent plasma $t_{1/2}$ was consistent across subcutaneous doses (1.0 hour).

Multiple-once weekly subcutaneous administration of 1 mg/kg for a total of 8 doses and 3 once-monthly doses of 4 mg/kg were also investigated. There was no accumulation following once weekly or once monthly dosing and repeat-dose pharmacokinetics showed no time dependence. Mean plasma exposure and apparent $t_{½}$ values for both schedules were similar to those observed after a single subcutaneous dose.

Study GO1-DSM15-020

Plasma lumasiran levels were determined after single intravenous and subcutaneous and after multiple (once weekly for 8 weeks or once monthly for 3 month) subcutaneous administration in male and female cynomolgus monkeys by the LC-TOF-MS method.

Following a single IV dose of 10 mg/kg, the mean C_{max} was 134 µg/mL with a mean AUC_{last} value of 58.9 h*µg/mL. Elimination from systemic circulation was rapid with an estimated $t_{½}$ of 0.6 hours. The mean CL and V_{ss} values were 189 mL/h/kg and 163 mL/kg, respectively, suggesting moderate distribution of lumasiran beyond the vasculature.

Following a single subcutaneous dose of lumasiran at 0.1, 1, 5, or 10 mg/kg in monkeys, plasma t_{max} was achieved at approximately 2 hours. Plasma C_{max} and AUC_{last} values increased in an approximately dose-proportional manner across the dose range evaluated, with similar $t_{1/2}$ values (approximately 3.5 hours) across doses; an exception was the 1 mg/kg dose group where females showed an extended half-life (12 hours) due to 2 animals in the group with significant plasma levels noted at later time points. Plasma concentrations in the 0.1 mg/kg dose group were sparse with 2 animals having no detectable lumasiran at any time point. The remaining 4 animals had detectable lumasiran concentrations in only 1 to 3 time points creating a profile that is less robust than the 1, 5 or 10 mg/kg dose groups.

Plasma concentrations of lumasiran were measured after multiple subcutaneous doses of 1 mg/kg (total of 8 once weekly doses) and 4 mg/kg (total of 3 once monthly doses). Overall, the dose normalised plasma pharmacokinetic profiles were similar in monkeys following single and multiple dosing indicating no time-dependent changes in the pharmacokinetic of lumasiran. Males had slightly higher AUClast and t½ values (<2-fold) after weekly dosing vs females; exposure and half-life values were equivalent between males and females after monthly dosing. There was minimal accumulation in plasma after multiple dosing.

Distribution

Plasma protein binding of lumasiran in mouse, rat, monkey, and human plasma was measured using an electrophoretic gel mobility shift assay (EMSA) (Study DSM19-021). The results are shown in **Table 3**.

Table 3. Plasma protein binding in rats, monkeys and humans, Study DSM19-021

Concentration (µg/mL)	**Mean±SD Percent Bound**		
	Rats	**Monkeys**	**Humans**
50	35.0±1.94	37.1±2.68	19.6±3.99
25	61.9±1.29	50.9±2.21	29.5±2.08
10	73.3±2.80	52.8±6.14	35.0±0.880
5	84.2±1.03	69.7±1.37	59.9±1.45
1	92.2±2.67	82.5±7.07	76.6±3.02
0.5	95.9±0.509	85.5±0.391	85.0±0.703

Note: n=3 samples per concentration

Distribution in rats

The distribution of lumasiran into liver and kidney was investigated in rats after single dose or 8 once weekly of 1 mg/kg or 3 once monthly subcutaneous administrations of lumasiran (GO1-DSM15-022). The results are presented in **Table 4.**

Table 4. Distribution of lumasiran in rat liver and kidney, Study GO1-DSM15-022

Regimen	Single Dose				QW×8	QM×3
Dose (mg/kg)	0.1	1	5	10	1	4
Day	1	1	1	1	50	57
Liver						
$t_{½}$ (h)	129	191	105	138	132	NC
t_{max} (h)	5.0	4.0	6.0	8.0	14.0	8.0
C_{max} (µg/g)	1.02	11.1	65.7	120	16.6	70.2
AUC_{last} (h*µg/g)	94.1	1210	5820	10700	1570	1320
Kidney						
$t_{½}$ (h)	NC	158	365	236	NC	NC
t_{max} (h)	0.25[a]	16.0	3.0	14.0	24.0	14.0
C_{max} (µg/g)	0.490[a]	0.870	4.35	8.56	4.02	5.27
AUC_{last} (h*µg/g)	113[a]	216	642	1540	406	103

Abbreviations: AUC_{last}=area under the concentration versus time curve from 0 to last quantifiable tissue concentration; C_{max}=maximum observed concentration occurring at t_{max}; NC=not calculated; QM=once monthly; QW=once weekly; $t_{½}$=elimination half-life; t_{max}=time to reach maximum concentration.[a] Value represents mean for females only; parameter was NC for males.

Distribution in rats by quantitative whole-body autoradiography (GO1-DSM18-038)

10 mg/kg [^{14}C] labeled lumasiran was administered subcutaneously to male non-pigmented (SD) rats and distribution and pharmacokinetic parameters in the different tissues were investigated by quantitative whole-body autoradiography.

The greatest exposure was observed for the dose site, followed by the liver and the kidney. Substantially lower concentrations of lumasiran were observed in almost every other tissue, including adrenal, heart, lung, spleen, thyroid, thymus, pancreas, small intestine, and testes. Central nervous system tissues had negligible exposure to radioactivity after SC administration of [14C]-lumasiran.

Distribution in monkeys

The distribution of lumasiran in monkey liver was evaluated after a single, or multiple once weekly or once monthly doses of lumasiran by subcutaneous administration (GO1 DSM15-020).

The kinetic parameters of lumasiran obtained in the liver are shown in **Table 5**.

Table 5. Overall mean liver pharmacokinetics in monkeys after single and multiple dose administration in Study GO1 DSM15-020.

Regimen	Single Dose - IV	Single Dose - SC				QW×8 – SC	QM×3 - SC
Dose (mg/kg)	10	0.1	1	5	10	1	4
Male Liver							
$t_{1/2}$ (h)	296	615	292	371	409	NC	312[a]
t_{max} (h)	88	48	16	16	48	1104	588
C_{max} (µg/g)	76.1	2.01	31.0	90.4	169	70.4	73.9
AUC_{last} (h*µg/g)	20300	1066.5	13485	41600	76700	66300	56850

Abbreviations: IV=intravenous; NC=not calculated due to insufficient data points; QM=once monthly; QW=once weekly; SC=subcutaneous.
Note: Values represent the overall combined (male+female) mean.
[a] Females only.

Metabolism

***In Vitro* Metabolic Stability of Lumasiran in Serum and Liver S9 Fraction (Study BA15014)**

The stability of lumasiran was evaluated in pooled serum of mouse, rats monkey and human with and without human liver S9-fractions.

No significant degradation (< 10% degradation) of lumasiran occurred in plasma of the species evaluated. In liver S9 fraction the anti- sense strand was stable to a similar degree, whereas the sense strand showed a slightly lower stability with a degradation from approximately 15 to 20 %.

Metabolite Profiling of Lumasiran

The metabolic patterns of lumasiran´s antisense and sense strands were investigated in samples from *in vitro* and *in vivo* nonclinical studies (BA15014). *In vitro* metabolite profiles were generated after incubating lumasiran with plasma and liver S9 fractions from mouse, rat, monkey, and human.

Plasma and liver samples were taken from pharmacokinetic studies in rats (GO1-DSM15-022), and monkeys (GO1-DSM15-020). Both species were dosed with 10 mg/kg lumasiran subcutaneously. Human plasma and urine samples were taken from the Phase 1 clinical study (ALN-GO1-001).

Metabolism of the Antisense Strand

The antisense strand of lumasiran was primarily metabolised to form the metabolite AS(N-1)3' lumasiran. There were a total of 12 antisense strand metabolites identified *in vivo* across species that formed from sequential exonuclease activity on the 5' and 3' ends of the strand.

The metabolite profiles of rat and monkey were comparable to the profiles obtained *in vitro* samples. The antisense strand was metabolised to a low extent to AS(N-1)3' lumasiran. The plasma AUC of the metabolite was smaller than or equal to 10% of the full length antisense strand exposure in either the rat or monkey.

In monkey liver, there was also a deaminated antisense strand metabolite (deaminated lumasiran) resulting in terminal adenosine to inosine conversion at the 3’ end. This was not detected as a circulating metabolite in rat or monkey and was not detected in liver S9 for any species. Both metabolites are not major circulating plasma components.

In pooled human plasma and urine samples (taken from clinical study ALN-GO1-001) the antisense of lumasiran was equally metabolised to AS(N-1)3’ lumasiran and the exposure was smaller than 10% of full-length parent compound.

Metabolism of Sense Strand

In vitro and *in vivo* metabolite profiling in rat and monkey serum and plasma (BA15014) demonstrated that the sense strand of lumasiran was minimally metabolised and was stable due to the GalNAc group from the triantennary ligand at the 3'-end, which protected the sense strand from the 3'-end exonuclease metabolism until the loss of the sugar moieties.

In vivo metabolite profiling of human pooled plasma and urine samples (BA15014) sampled for 24 hours following a single 6 mg/kg subcutaneous administration of lumasiran demonstrated that the sense strand was minimally metabolised with the loss of one GalNAc.

In Vitro Hepatocyte Metabolism

Since a deaminated lumasiran was identified in monkey liver samples an *in vitro* human hepatocyte model (HepatoPac®) was used to determine if deaminated lumasiran is likely to be formed in human

liver (Study G01-DSM19-022). The potential for age-dependent differences in ADME properties of lumasiran was also investigated using hepatocytes from 8 donors (aged 1 month to 48 years).

The cells were preloaded for 24 hours with media spiked with lumasiran at 10 µg/mL.

In this model, the metabolite AS(N-1)3' lumasiran was present at levels less than lumasiran whereas deaminated lumasiran was present at higher levels than parent after a 168-hour incubation period, suggesting that humans have the ability to form deaminated lumasiran. No correlation with ASGR level or function observed with age

In Vitro CYP Inhibition

In pooled human liver microsomes, lumasiran did not directly inhibit CYP1A2, CYP2B6, CYP2C9, CYP2C19, CYP2D6, and CYP3A4/5 at concentrations ranging from 0.6 nM to 614 µM (Study 319N-1506A). Lumasiran showed inhibition of CYP2C8 with a calculated IC_{50} value of 461 µM, which is approximately 14000-fold above expected pharmacologic concentrations. Time-dependent inhibitory potential was also evaluated by preincubating liver microsomes with varying concentrations of lumasiran in the presence and in the absence of cofactor, NADPH. Lumasiran did not demonstrate any time-dependent inhibition towards any of the CYP isoforms evaluated.

In Vitro CYP Phenotyping

Lumasiran was incubated with individual recombinant cytochrome P450 (rCYP) enzymes rCYP1A2, rCYP2B6, rCYP2C8, rCYP2C9, rCYP2C19, rCYP2D6, rCYP3A4 and rCYP3A5 to determine if CYPs contribute to the metabolism of lumasiran (Study 319N-1506B).

The percentage of lumasiran remaining after 45 minutes was approximately 100% for all rCYPs tested in the presence of NADPH.

In Vitro CYP Induction

Lumasiran was not tested as an inducer of CYP enzymes *in vitro*. The applicant referred to published *in vitro* DDI data for siRNA GalNAc conjugate molecules that share similar physicochemical properties with lumasiran have been evaluated *in vitro* and were not inducers of major CYPs (Ramsden et al. 2019).

Excretion

Renal excretion of lumasiran was evaluated in rats following a single subcutaneous dose of 5 mg/kg (Study GO1-DSM15-022). Renal clearance was 29 mL/h/kg in males and 17 mL/h/kg in females and accounted for <1% of the total systemic clearance, with the majority of clearance from circulation resulting from liver uptake. Only minimal amounts of total test article administered in rats were recovered in feces with most samples having no detectable lumasiran.

The excretion of radiolabeled lumasiran was investigated after administration of a single subcutaneous dose (10 mg/kg) of [^{14}C]-lumasiran to male intact and bile duct cannulated animals (Study GO1-DSM18-038, **Table 6**).

Table 6. Overview of excretion data in rat from study GO1-DSM18-038

ID Species	Anal.	Urine (% dose)	Faeces (% dose)	Bile (% dose)	Cage residues (% dose)	Carcass (% dose)	Recovery (% dose)	Time (h)

Rat (intact)	[^{14}C]-lumasiran	19.5 ± 1.34	33.9 ±0.693		1.09 ± 0.378	8.88 ± 1.10	63.4 ± 1.32	0-1344
Rat (bile duct cannulated)	[^{14}C]-lumasiran	15.3 ± 2.43	0.666 ± 0.289	28.3 ± 6.52	0.666 ± 0.291	34.0 ± 6.06	78.9 ± 1.73	0-168

INV-DSM16-057 – WIL-268058B

Lumasiran concentrations in seminal fluid were evaluated in male NZW rabbit after single subcutaneous administration of 3 mg/kg and 30 mg/kg. Samples were collected 8, 24, 72, and 216 hours post dose.

Very low levels of lumasiran were detected at all time points post-dose in both dose groups, with Cmax occurring at 8 hours post-dose. These values were less than 1% of the plasma Cmax when compared with plasma toxicokinetic levels observed in pregnant female rabbits that also received doses of 3 and 30 mg/kg.

GO1-DSM19-002

The excretion of radiolabeled lumasiran were investigated after administration of a single subcutaneous dose (10 mg/kg) of [^{14}C]-lumasiran to male monkeys.

The mean cumulative recovery over 90 days was relatively low with 54.6%±8.12%. Approximately 38% of the dose could be found in the urine, 8% in the feces, and 9% in the cage residue. Detectable levels of radioactivity were measured in the excreta of each animal daily through Day 90 but diminished appreciably after Day 2. The largest amount (~30%) was recovered in the first 24 hours.

Measurable levels of radioactivity were observed in the kidney, and to a higher degree in the liver. The highest levels of radioactivity in the kidney and liver were measured at 24 hours post dose, with estimates of 0.33% and 54.7% of the total radioactive dose, respectively. The amount of the radioactive dose remaining in kidneys at 90 days post dose was minimal and consistent at ~0.04%. Radioactivity was still measurable at 90 days post dose in the liver, ranging from ~0.9% to 7.4%. Though the carcasses were not analyzed as part of this study, the excretion profile supports the likelihood that the remaining unrecovered radioactive dose was not retained in the carcass.

2.3.4. Toxicology

Single dose toxicity

A standard single-dose toxicity study with lumasiran was not submitted.

Clinical observations were made following a single dose in the GLP *in vivo* rat erythrocyte micronucleus study (GO1-GLP15-016). In this study, rats were monitored for up to 48 hours after SC administration of a single dose of lumasiran. There were no deaths or adverse clinical signs observed. Lumasiran was well tolerated at up to 2000 mg/kg (the limit dose in this study).

Repeat dose toxicity

The applicant submitted five repeated-dose toxicity studies, summarised in **Table 7**.

Table 7. Overview on repeated dose toxicity studies

Study ID (GLP status)	**Species (sex)**	**Duration**	**Dose (mg/kg) / Frequency of administration**	**NOAEL (mg/kg) Major findings**
GO1-DSM15-001 (non-GLP)	Rat (M)	2 weeks	0, 30, 100 / once a week	Changes in clinical pathology parameters (coagulation), histology (liver, kidney).
GO1-GLP15-008 (GLP)	Rat (M+F)	8 weeks + 13 weeks recovery	0, 5, 15, 50 / once a week and 50 / once a month	50 QW and 50 QM 5mg: Injection site findings; ↓ FIB 15mg: As above, plus prolonged PT; ↑ ALP (M); ↑ CHOL (F); ↑ TRIG (F); ↑ relative liver weights (F); hepatocyte vacuolation 50mg: As above, plus ↑ AST; ↑ GGT (M); ↑ BILI (M); ↑ CHOL (M); ↓ red cell mass (RBC, HGB, HCT) (M); ↑ PLT (M); ↑ RETI (M); ↑ LYM (M); ↑ WBC; ↑ GLOB with corresponding ↓ ALB:GLOB ratio (F); diffuse, pale liver in 1 male; basophilic granules in Kupffer cells; individual hepatocyte necrosis, increased mitoses, and centrilobular hepatocyte hypertrophy; basophilic granules in renal tubular epithelium Recovery Phase Findings: ↑ absolute RETI (M); hepatocellular vacuolation; basophilic granules in cytoplasm of Kupffer cells (M); hepatocellular hypertrophy/ karyomegaly (F); basophilic granules in kidney (F)
GO1-GLP15-038 (GLP)	Rat (M+F)	25 weeks	0, 20, 50 and 200 / once a month	200 QM 20 mg: ↓ FIB; hepatocellular vacuolation; increased pigment containing Kupffer cells; hepatocyte karyomegaly; increased mitotic figures in hepatocytes; basophilic granules in the kidney tubule cell cytoplasm

				50 mg: As above, plus ↓ RBC mass (F); ↓ MCV, MCH, MCHC, and ↑ ABS RETIC (F); ↑ PT (F); ↓ aPTT (M); ↑ CHOL; ↑ GLOB and ↓ ALB:GLOB (F); vacuolation of renal tubule cells (M) 200 mg: As above, plus ↓ RBC mass (M); ↓ WBC (F); ↓ aPTT (F); ↑ ALP (M)
GO1-GLP15-009 (GLP)	Cynomolgus monkey (M+F)	8 weeks + 13 weeks recovery	0, 10, 30 and 100 / once a week and 100/ once a month	100 QW and 100 QM 10 mg: Accumulations of vacuolated macrophages in the sinusoids of lymph nodes 30 mg: As above, plus basophilic granules in Kupffer cells 100 mg: As above, plus ↑ ALP Recovery Phase Findings (Day 92): As above; ↑ ALP not present at recovery
GO1-GLP15-036 (GLP)	Cynomolgus monkey (M+F)	36 weeks	0, 30, 100, 300 / once a month	300 QM 30 mg: ↑ ALP (M); accumulations of vacuolated macrophages in the sinusoids of lymph nodes 100 mg: As above, plus basophilic granules in Kupffer cells 300 mg: As above

ABS RETIC=absolute reticulocytes; ALB=albumin; ALP=alkaline phosphatase; aPTT=activated partial thromboplastin time; AST=aspartate dehydrogenase; AUC=area under the concentration-time curve; AUClast=area under the concentration-time curve to last quantifiable sample; BILI=bilirubin; CHOL=cholesterol; Cmax=Maximum observed (peak) concentration occurring at tmax; F=female; FIB=fibrinogen; GGT=gamma glutamyl transferase; GLOB=globulin; HCT=haematocrit; HGB=hemoglobin; LYM=lymphocytes; M=male; MCV=mean corpuscular volume; MCH=mean corpuscular hemoglobin; MCHC=mean corpuscular hemoglobin concentration; NOAEL=no observed adverse effect level; PLT=platelets; PT=prothrombin time; RBC=red blood cell; RETI=reticulocytes; TRIG=triglycerides; WBC=white blood cells

Genotoxicity

Lumasiran was tested in a standard battery of genotoxicity tests according to ICH S2(R1). Studies conducted and main results are listed in **Table 8**.

Table 8. Overview of genetic toxicity studies with lumasiran

Type of test/study ID/GLP	Test system	Concentrations/ Concentration range/ Metabolising system	Results Positive/negative/equivocal
Gene mutations in bacteria / GO1-GLP15-015 / yes	*Salmonella* strainsTA98, TA100, TA1535, TA1537 and *E. coli* WP2uvrA	+/- S9: 1.6, 5.0, 16.0, 50.0, 160, 500, 1600, and 5000 µg/plate	No bacteriotoxicity, no significant increase in revertants
chromosomal aberrations in mammalian cells / GO1-GLP15-013 / yes	cultured human peripheral blood lymphocytes	3 h +/- S9 and 24 h –S9: 3.39, 4.84, 6.92, 9.89, 14.1, 20.2, 28.8, 41.2, 58.8, 84.0, 120, 172, 245, 350, and 500 µg/mL	No cytotoxicity up to the highest recommended dose, no significant increase in chromosomal aberrations
Chromosomal aberrations in vivo / GO1-GLP15-016 / yes	Sprague Dawley rat 5/sex/group, plus 9/sex/group for TK, micronuclei in bone marrow	application route SC, single dose, harvest 24 and 48 h post dose males: 0, 500, 1000, 2000 mg/kg females: 0, 2000 mg/kg	No toxicity observation and no bone marrow toxicity in PCE/NCE ratio No significant increase in micronucleated PCEs TK results at 2000 mg/kg: males: C_{max} 208 µg/ml, AUC_{48h} 4510 µg*h/ml females: C_{max} 220 µg/ml, AUC_{48h} 3680 µg*h/ml

Carcinogenicity

Carcinogenicity studies are planned in Tg-rasH2 mice for 26 weeks with dose levels of 0, 150, 500, and 1500 mg/kg once a month and in Sprague Dawley rats for 2-years with dose levels of 0, 20,55, and 110 mg/kg once a month. In concordance with the CHMP scientific advice the study results will be submitted post approval.

Reproduction Toxicity

Study designs and major findings are summarised in **Table 9**.

Table 9. Summary table of reproductive toxicity studies performed for lumasiran

Study Type (Study ID) GLP	Species; Number/ Dose Group	Dose (mg/kg) s.c. injection Dosing Period	Major Findings	NOAEL (mg/kg)
Male and female fertility and early embryonic development (treated M/F were paired with untreated F/M) (GO1-GLP18-002) GLP + TK	 SD-Rats M 20/dose F 20/dose TK 3/6/6/6 for M + F	0 (0.9 % NaCl)) 5 15 50 QW M:29 days prior to/during mating, until termination (8 doses total) F: 22 days prior to mating until GD 6 (5 doses total) C-Section: GD 13	↓mean bw gain (M/HD) ↓food consumption (M/LD-HD) M: AUC_{last}= 63.7 µg.h/mL; Cmax= 9.71 µg/mL F: AUC_{last}= 40.6 µg.h/mL; Cmax= 7.20 µg/mL	Paternal and maternal 50 Male and female fertility and reproduction 50

		TK (M D29, F GD 6))		
DRF Female fertility and early embryonic development (GO1-GLP16-006) GLP +TK +Tissue distr. +PD	SD-Rats 22/dose TK + tissue 3/6/6/6	0 (0.9 % NaCl)) 10 (LD) 30 (MD) 100 (HD) D15 / D8 / D1 prior to mating (3 doses) and 0 (0.9% NaCl) 3 (LD) 10 (MD) 30 (HD) on GD 1-17 QD C-section GD21 TK (GD17) Tissue (GD21) PD (D15/ D1 prior to mating, GD21)	↓food consumption (LD-HD) ↓bw (LD-HD) **haematology:** ↓Fib (LD-HD), ↓Red Cell Mass (LD-HD: ↓RBC count, ↓Hb, ↓HC), ↑abs. neutrophils (LD-HD), ↑platelet counts (LD-HD) ↑WBC count (HD: ↑abs. neutrophils,↑lymphocytes, ↑monocytes, ↑LUC) **clinical chemistry** ↓protein (MD,HD), ↓A/G ratio (MD,HD), ↑cholesterol (MD,HD) ↑triglycerides, (MD-HD) **fetal parameters** ↓mean fetal weights (LD-HD)	Maternal 0 (zero)_ Female fertility and embryo-fetal development 100 QW/30 QD mg/kg (AUC_{last}= 26.6 µg.h/mL; Cmax= 5.85 µg/mL)
Embryo-fetal development (GO1-GLP18-001) GLP +TK	SD-Rats 22/dose TK 3/6/6/6	0 (0.9 % NaCl) 3 (LD) 10 (MD) 30 (HD) daily GD6 – GD17 C-section GD18/GD21 TK (GD17)	↓food consumption (HD) **fetal parameters** <u>skeletal variations*:</u> -bipartite ossification of sternebrae (2/2 HD), -misshapen cervical arches (1/1 LD, 2/1 MD, 4/4 HD)	Maternal 30 (AUC_{last}= 17.9 µg.h/mL; Cmax= 3.49 µg/mL) Embryo-fetal development 10 (AUC_{last}= 4.39 µg.h/mL; Cmax= 0.878 µg/mL)
DRF Embryo-fetal development (GO1-GLP16-005) GLP +TK +Tissue distr. +PD	NZW rabbits 8/dose TK 3/dose	0 (0.9 % NaCl) 3 (LD) 10 (MD) 30 (HD) daily GD7 – GD19 C-section: GD29 TK (GD17 / GD19) Tissue (GD29) PD (GD7, GD13, GD29)	**fetal parameters:** <u>visceral malformations*:</u> -dilated aortic arch (1/1 MD, 1/1 HD) -diaphragmatic hernia (2/1 MD) -absent gall bladder (1/1 MD) -cardiomegaly (1/1 LD) -three chambered heart (1/1 LD) -ventricular septum defect (1/1 LD, 1/1 MD) -hepatomegaly (1/1 LD) -abnormal lobulation liver (1/1 MD) -microsplenia (1/1 MD)	Maternal 30 (AUC_{last}= 46.6 µg.h/mL; Cmax= 6.02 µg/mL) Embryo-fetal development 0 (zero)
Embryo-fetal development (GO1-GLP16-20) GLP +TK +Tissue distr. +PD	NZW rabbits 25/dose TK 3/dose	0 (0.9 % NaCl) 3 (LD) 10 (MD) 30 (HD) daily GD7 – GD19 C-section: GD29 GD20 for TK + tissue TK (GD7, GD19)	**clinical signs** few faeces (MD, HD) ↓**body weight gain** GD7-GD20 (LD-HD) ↓**food consumption** GD7-GD29 (LD-HD) **fetal parameters** <u>skeletal malformations*:</u> fused mandible/zygomatic arch (1/1 in ctrl, 3/2 HD)	Maternal: 30 (AUC_{last}= 59.7 µg.h/mL; Cmax= 10.1 µg/mL) Embryo-fetal development 10

		Tissue (GD20) PD (GD7, GD13, GD19, GD29)		(AUC_{last}= 12.2 µg.h/mL; Cmax= 2.4 µg/mL)
Pre- and postnatal development (GO1-GLP18-018) GLP +TK	SD-rats F0: 22/dose TK (F0) 4/4/4/4 F1: 17 – 22 /dose	0 (0.9 % NaCl) 5 (LD) 15 (MD) 50 (HD) every 6 days F0 (GD7, GD13, GD19; LCD6, LCD12, LCD18) †F0: LCD21 F0 TK (GD7, GD13, GD19; LCD6, LCD12, LCD18) †F0 TK (LCD12) fetal plasma: LCD12 (2h after dosing F0) F1 litters on LCD 21: 1/sex for F2 F1 reproductive function C-section: GD13	**F0** ↓bw GD10-GD12 (HD) **F1 (pre-weaning)** ↓live-born pups (HD) ↓pup body weights (LD,MD)	F0 maternal 50 F1 neonatal/ development50 (zero) F1 reproductive function 50
*fetuses/litter abs. : absolute, A/G: albumin/globulin, Fib: fibrinogen, GD: gestation day, Hb: haemoglobin, HC: haematocrit, HD: high dose, LCD: lactation day, LD: low dose, LUC: large unstained cell counts, MD: mid dose, NZW: New Zealand White, RBC: red blood cell, SD: Sprague Dawley, TK: toxicokinetics, QD: daily, QW: weekly				

Placental transfer

Concomitantly with the studies on embryo-fetal development in rats and rabbits, placental, fetal liver and fetal tissue lumasiran concentrations were measured. Lumasiran was not detected in fetal tissue and liver samples. No lumasiran was detected in placentae of the low dose group and only in low concentrations in the higher dose groups of both species.

Milk excretion

No specific study to investigate the excretion of lumasiran into the milk of lactating animals was submitted. However, concomitant with the study on pre-postnatal development in rats lumasiran plasma concentrations were measured in suckling pups of lumasiran treated dams. Lumasiran plasma levels were found to be below the level of quantification indicating that lumasiran concentrations in milk are too low to show up in plasma of suckling pups.

Juvenile toxicity

In a pilot juvenile and toxicity study performed in rats from PND4 to PND33, injection site reactions, decreases in fibrinogen and microscopic findings in the kidneys were the main observations after weekly SC injections of lumasiran (GO1-GLP15-043). Similar effects were also observed in the 8-week repeat-dose study in rats, which were 4-5 weeks of age at start of treatment (GO1-GLP15-008). However, liver (increases in liver enzymes, hepatic necrosis) as well as kidney findings (microscopic changes) were much more pronounced in the older rats of the 8-week study although doses were in general lower. This was probably due to higher cumulative doses resulting from altogether 9 weekly doses in the older compared to 5 weekly doses in neonate/juvenile rats.

Plasma toxicokinetics showed no sex differences in exposure to lumasiran and no accumulation in the plasma of treated juvenile animals. Like in adult animals, liver exposure was much higher than kidney exposure. Increases in glycolate levels and decreases in HAO1 mRNA showed pharmacologic activity of lumasiran on PND32 after QW lumasiran treatment from PND 4 onwards in all dose groups. Concerning age and comparison to humans, a rat at PND 32 equals approximately a 6-8 year old child. Altogether, due to kidney findings, the NOAEL of the pilot juvenile toxicity study is at 30 mg/kg/week with exposure multiples of approximately 2 compared to human therapeutic exposures (based on monthly levels).

Toxicokinetic data

Toxicokinetic (TK) assessments were included as part of the GLP-compliant toxicity studies.

An 8-Week Subcutaneous Injection Toxicity and Toxicokinetic Study in Rats Followed by a 13-Week Recovery Phase (GO1-GLP15-008)

Table 10. Summary of TK parameters of ALN-GO1 in rats in study GO1-GLP15-008 (genders combined)

Day 1				
(mg/kg/dose)	5 QW	15 QW	50 QW	50 QM
t1/2 (h)	ND	0.84	0.96	0.89
tmax (h)	1.0	0.50	1.0	1.0
Cmax (ng/mL)	581	2900	14400	14500
tlast (h)	4.0	4.0	8.0	8.0
AUClast (ng.h/mL)	1220	6020	42600	43100
AUCinf (ng.h/mL)	ND	6310	42800	43200
Day 57				
t1/2 (h)	1.2	1.4	2.2	1.5
tmax (h)	1.0	1.0	1.0	1.0
Cmax (ng/mL)	596	1760	12700	10700
tlast (h)	8.0	8.0	8.0	8.0
AUClast (ng.h/mL)	2090	7710	51800	44600
AUCinf (ng.h/mL)	2110	7890	56500	46000

A 25-Week Subcutaneous Injection Toxicity and Toxicokinetic Study in Rats (GO1-GLP15-038)

Table 11. TK parameters of ALN-GO1 in rats after subcutaneous administration of ALN-GO1 every 4 weeks for 7 doses in Study GO1-GLP15-038; genders combined

	Day 1			Day 169		
ALN-GO1 (mg/kg/day)	20	50	200	20	50	200
Cmax (ng/mL)	3521	12180	37665	1511	6830	29948
tmax (h)	1.0	1.0	2.0	1.0	1.0	0.5
tlast (h)	12	24	24	24	24	24
AUClast (ng.h/mL)	10099	44563	265876	12630	51273	377599
AUCinf (ng.h/mL)	10125	ND	266458	14510	52930	387990
t1/2 (h)	1.4	ND	2.6	9.3	5.0	4.2

An 8-Week Subcutaneous Injection Toxicity and Toxicokinetic Study in Monkeys Followed by a 13-Week Recovery Phase (GO1-GLP15-009)

Table 12. Mean (Male and Female Combined) TK parameters of ALN-GO1 after subcutaneous administration of ALN-GO1 in cynomolgus monkeys once weekly or once every four weeks in study GO1-GLP15-009

Day 1				
ALN-GO1 (mg/kg/dose)	10 QW	30 QW	100 QW	100 QM
Cmax (ng/mL)	2710	8320	20100	23700
tmax (h)	1.7	2.2	4.8	3.1
tlast (h)	9.6	14	24	24
AUClast (ng.h/mL)	13500	64700	312000	336000
AUCinf (ng.h/mL)	16600	96800	333000	339000
t1/2 (h)	2.6	5.3	7.2	6.0
Day 57				
Cmax (ng/mL)	2950	9340	25600	30300
tmax (h)	2.1	2.4	4.4	3.2
tlast (h)	9.6	21	24	24
AUClast (ng.h/mL)	15300	86600	367000	389000
AUCinf (ng.h/mL)	19500	88600	ND	413000
t1/2 (h)	2.7	3.0	ND	5.1

A 36-Week Subcutaneous Injection Toxicity and Toxicokinetic Study in Monkeys (GO1-GLP15-036)

Table 13. Mean TK parameters of ALN-GO1 after subcutaneous administration of ALN-GO1 in Cynomolgus monkeys once every four weeks in Study GO1-GLP15-036

Day 1									
	Males			Females			Genders Combined		
ALN-GO1 (mg/kg/dose)	30	100	300	30	100	300	30	100	300
Cmax (ng/mL)	8840	25600	57000	9360	19400	58200	9100	22500	57600
tmax (h)	3.0	2.5	3.5	2.5	2.5	3.6	2.8	2.5	3.6
tlast (h)	40	34	40	48	44	48	44	39	44
AUClast (ng.h/mL)	91700	321000	1050000	93600	315000	1110000	92600	318000	1080000
AUCinf (ng.h/mL)	98800	324000	1110000	76400	321000	1110000	87600	323000	1110000
t1/2 (h)	3.4	3.7	3.8	7.7	3.0	3.4	5.5	3.4	3.5
Day 253									
Cmax (ng/mL)	11200	37700	85800	8620	26700	67000	9890	31400	76400
tmax (h)	4.0	2.7	5.5	4.0	6.5	6.0	4.0	4.9	5.8
tlast (h)	24	24	24	24	24	24	24	24	24
AUClast (ng.h/mL)	108000	490000	1450000	118000	383000	1120000	113000	429000	1290000
AUCinf (ng.h/mL)	110000	529000	2240000	129000	ND	1600000	119000	529000	1920000
t1/2 (h)	3.1	6.0	14	3.1	ND	14	3.1	6.0	14

- **Interspecies comparison**

The applicant has provided exposure multiples of animal exposure (AUC(0-last)) to human therapeutic exposure for the two pivotal repeated dose studies in rodents (rats) and non-rodents (monkeys), respectively (**Table 14**).

Table 14. Lumasiran safety margins based on HED and $AUC_{0\text{-last}}$

Study	Dose (mg/kg/month)	AUC0-last (h*µg/mL)[a]	Safety Margin for Clinical Dose of 2 mg/kg/month AUClast [b]= 4.96 µg*h/mL	
			Based on AUC0-last (h*µg/mL)	Based on HED[c] (mg/kg)
GO1-GLP15-038: 25-week SC Toxicity and TK Study in **Rats**	20	12.6	**2.5×**	1.6×
	50	51.3	**10.3×**	4.0×
	200	378	**76.1×**	16.1×
GO1-GLP15-036: 36-week SC Toxicity and TK Study in **Monkeys**	30	113	**22.8×**	4.8×
	100	429	**86.4×**	16.1×
	300	1290	**260×**	48.4×

a AUC last was taken from the dose on the last day of lumasiran administration for the animal studies; data are from sexes combined.
b AUC calculated value from aggregate plasma pharmacokinetic parameters of lumasiran after a single dose of lumasiran in healthy adult volunteers and PH1 patients (ALN-GO1-001 Part A & B).
c HED was calculated using the nonclinical dose and dividing by the appropriate species allometric conversion factor (6.2 for rat and 3.1 for monkey) as outlined in the Guidance for Industry: Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers (FDA, July 2005).

Local tolerance

No dedicated local tolerance studies with lumasiran were submitted.

The potential of dermal irritation at the SC injection sites and microscopic evaluation of the injection site tissue was investigated in all repeat-dose toxicity studies.

In the repeat-dose GLP toxicology studies in rats and monkeys, transient, very slight erythema and transient, very slight to slight oedema were observed with once weekly SC administration of lumasiran. Dermal observations following once monthly SC administration to rats for 25 weeks appeared rather infrequently, were also present in control animals, and did not always correlate with the location of the SC injection; therefore, they were not considered adverse or lumasiran related.

In the 36-week monkey study (GO1-GLP15-036), no lumasiran-related dermal irritation was observed. Microscopic findings at the injection site, cellular infiltrates, and/or inflammation were present in both rats and monkeys at all dose levels; increased incidence and severity of subcutis muscle degeneration (minimal to moderate) was seen in rats.

Other toxicity studies

A non-GLP-compliant study (GO1-DSM16-048) was performed to investigate the effects of chronic renal impairment (via the 5/6-nephrectomised rat model) on PK, PD, and toxicity in male Sprague Dawley rats after a single SC dose of lumasiran.

A single administration of 1 or 3 mg/kg lumasiran was not associated with any lumasiran-related effects on serum chemistry, body weights, organ weights, or macroscopic and microscopic findings in the liver and kidney in either sham surgery or nephrectomised animals. Nephrectomy did not substantially affect the extent of rat liver HAO1 mRNA reduction after a single 1-mg/kg SC injection of lumasiran. Renal impairment does not result in plasma exposure differences after administration of a single dose of lumasiran. Kidney concentrations were lower on Day 2 in nephrectomised rats compared with sham surgery animals. Comparing Day 2 with Day 14, these data suggest increased and prolonged lumasiran kidney concentrations and prolonged kidney residence in nephrectomised animals dosed with lumasiran compared with sham-operated animals.

Anti-Drug Antibody (ADA) was evaluated in rat and monkey plasma following weekly administration of lumasiran for 25 and 36 weeks, respectively. A low incidence, 3 out of 36 treated monkeys in the 36-week repeat-dose toxicity study (GO1-GLP15-036) were confirmed to have a low titre of anti-lumasiran antibodies. Plasma exposure of lumasiran was not impacted and there were no toxicological findings attributed to the presence of ADAs in these animals. No ADAs were detected in treated rats.

The impurity profiles of the drug substance batches used in the nonclinical toxicology studies were comparable to the impurity profiles of the material used in clinical investigations and that proposed for use in the marketed product. Additionally, a 13-week repeat-dose toxicity study in rats (GO1-GLP19-002) was conducted to generate toxicology data with lumasiran drug substance that contained potential commercial scale manufacture impurities that were not identified in previous toxicology lots.

Toxicologic findings were injection site reactions, decreased body weight gain, changes in serum liver parameters, serum lipids, coagulation and haematology. These changes were similar to the findings made in the rat repeated-dose studies and do not identify new target organs of toxicity. The histological alterations observed in liver and kidney were also similar to the findings in the repeated-dose studies. Thus, there is no hint that the commercial scale impurities cause toxicity beyond the effects of the active substance lumasiran.

2.3.5. Ecotoxicity/environmental risk assessment

The applicant provided an environmental risk assessment (ERA) in accordance with the Guideline on the environmental risk assessment of medicinal products for human use (EMEA/CHMP/SWP/4447/00 corr 2).

The $PEC_{surface\ water}$ was refined with prevalence data from the Orphan Designation EU/3/16/1637.

Table 15. Summary of main study results

Substance (INN/Invented Name): Lumasiran sodium			
PBT screening		Result	**Conclusion**
Bioaccumulation potential- log K_{ow}	OECD107	< -2.8 at pH 7	Potential PBT N
PBT-assessment			
Parameter	**Result relevant for conclusion**		**Conclusion**
Bioaccumulation	log K_{ow}	< -2.8 at pH 7	not B
PBT-statement:	The compound is not considered as PBT nor vPvB		
Phase I			
Calculation	**Value**	**Unit**	**Conclusion**
$PEC_{surfacewater}$, refined with prevalence	0.0000088	µg/L	> 0.01 threshold N

2.3.6. Discussion on non-clinical aspects

The pharmacology, pharmacokinetics and toxicology of lumasiran were evaluated in a series of *in vitro* and *in vivo* non-clinical studies. Overall, the study program appears to be appropriate and complete.

Pharmacology of lumasiran was demonstrated in WT mice, rats and monkeys and in an induced rat PH1 model.

In all animal models, single or repeat weekly or monthly subcutaneous administration of lumasiran primarily resulted in dose-dependent reductions of *HAO1* mRNA, increases in serum glycolate levels and/or reductions in urinary oxalate.

In WT mice, rats and monkeys, as the percent of *HAO1* mRNA decreased, serum glycolate levels increased but in a nonlinear manner, such that substantial glycolate increases were not seen until approximately 70% to 80% reduction of *HAO1* mRNA. These results are consistent with the expected enzymology of glycolate oxidase in WT animals, where enzyme levels should not be rate limiting in the conversion of substrate (glycolate) until they reach low levels.

In the PH1-induced rat model, as the percent of *HAO1* mRNA decreased, the amount of urinary oxalate decreased in a linear manner (*HAO1* mRNA reduction to oxalate lowering) even at low levels of mRNA silencing. This is consistent with the expected enzymology of glycolate oxidase, where enzyme levels should be rate limiting in product (oxalate) formation at all levels.

The applicant conducted a GLP-compliant cardiovascular safety study with determination of the respiration rate in telemetered conscious male Cynomolgus monkeys. No immediate or delayed effects on clinical observations, qualitative or quantitative ECG parameters, hemodynamic parameters, respiration rate, or body temperature were seen in this study and the NOEL was ≥100 mg/kg bw, the highest dose tested. The safety factor of about 20 between C_{max} at the maximum recommended dose in humans and the exposure in this safety pharmacology study is considered acceptable.

CNS safety was investigated as part of repeated dose general toxicity studies GO1-GLP15-009 and GO1-GLP15-036 in Cynomolgus monkeys. No lumasiran-associated neurological observations were noted but there is presently some discrepancy between the NOEL stated by the applicant and the dosing levels of the studies.

Mining of a public dbSNP database revealed a low frequency of SNPs which strongly suggests that lumasiran would be efficacious in reducing *HAO1* in people of all ethnic and geographical backgrounds.

In vitro analysis of mRNA reduction by lumasiran was conducted on the potential off-target transcripts identified from an *in silico* analysis comparing the sequence of only the antisense strand against the

human transcriptome. The results suggest there is a >1000-fold difference between the on-target reduction of *HAO1* by lumasiran and the off-target reduction of any of the predicted off-target transcripts, confirming the specificity of lumasiran for *HAO1.*

Plasma PK profiles were characterized following multiple SC dosing regimens of lumasiran (8 weekly doses of 1 mg/kg and 3 monthly doses of 4 mg/kg in rats and monkeys). In both species, the multiple-dose plasma PK data were consistent with single-dose plasma PK data. Due to the short half-life of lumasiran compared with dosing frequency, there was no accumulation in rat and minimal accumulation in monkey plasma following repeat once weekly or once monthly dosing at the PK doses tested. Overall, these PK properties of lumasiran in rats and monkeys indicate no time- or dose-dependency following multiple SC doses. The TK properties of lumasiran were evaluated in mice, rats, rabbits, and monkeys to support the toxicological assessment. In general, there was little to no accumulation, no consistent sex related TK differences and the plasma exposure (Cmax and AUC) increased either dose proportionally, or in some cases, increased slightly greater than dose proportionally across the dose range tested.

The applicant also conducted a single-dose study in 5/6 nephrectomised rats to mimic the situation in patients suffering from renal insufficiency due to the underlying disease. No relevant differences to healthy animals were detected in respect to the PK, PD and toxicity parameters tested.

Submitted studies show the preferred uptake of lumasiran into the target organ after subcutaneous administration. Several studies showed a good correlation between lumasiran administration and an increase in plasmatic glycolate concentration, the expected effect in healthy animals. The metabolism of lumasiran was primarily due to nucleoside cleavage by exo- or endonucleases. Investigations on DDI were limited to studies on inhibition. Further studies on DDI were not considered necessary with respect to the publication of Ramsden et al. 2019. Overall, the animal species involved in the non-clinical study program are adequate from the perspective of pharmacokinetics.

Consistent toxicology findings in the repeat-dose studies were histological changes in the liver. In the rat, these consisted of intracellular vacuoles in periportal cells containing lipid-like material as well as basophilic granules in Kupffer cells. Hepatocyte hypertrophy, mitosis and necrosis were also observed. Severity of the changes increased with increasing dose; effects were already seen at the lowest dose tested. The findings were partially reversible. These histological changes were accompanied by changes in serum liver parameters such as increased AST, ALP and GGT. Furthermore, there was consistent decrease in plasma fibrinogen levels, sometimes together with prolonged prothrombin time, and increase in serum lipids (cholesterol and triglycerides). Lipid vacuoles in periportal cells, cell necrosis and regeneration (mitosis) indicate cytotoxicity. Cytotoxic effects could also be responsible for the observed decrease in fibrinogen since a known mechanism via activation of PPARα was ruled out.

In monkeys, basophilic granules were observed in the Kupffer cells of the liver, and an increase in serum ALP. No other histological liver changes or serum chemistry alterations were observed in monkeys.

According to the data presented, the lumasiran level in the liver is similar in rats and monkeys. The reason for this apparent difference in toxicity between species remains unclear.

Mechanistic studies in the published literature have investigated lumasiran among other experimental siRNAs in order to elucidate the mechanism underlying the described hepatic effects (*Janas et al.2018)* and several possible mechanisms of toxicity to hepatocytes have been postulated. These include binding to cellular proteins as e.g. observed with oligonucleotides containing a high amount of phosphorothioate linkages, disturbance of endogenous processes of RNAi involving microRNAs or off-target knock-down of genes other than the desired one (HAO1 in the present case). As the underlying mechanism however is still unknown, these observations are described in the pre-clinical safety section

of the SmPC which also states that the reason for the apparent rodent-specificity is not understood and the relevance for humans is unclear. In addition, hepatic effects have been included in the RMP as an important potential risk. Further information on the effect of lumasiran on the liver in humans will be collected through the ongoing trials but also though the planned long-term real-world safety study in patients with PH1.

In the rat, kidney changes were also observed, consisting of basophilic granules in the tubular cells. In monkeys, vacuolated macrophages were observed in lymph nodes already at the lowest dose.

Lumasiran is designed in such a way that it is selectively taken up through the binding of GalNAc by the asialoglycoprotein receptor (ASGPR) on the surface of liver cells, after which it becomes internalised. At higher doses, this uptake mechanism becomes saturated so that lumasiran then becomes excreted via kidney, leading to increasing tissue concentrations in this organ. Basophilic granules were observed in Kupffer cells in rats and monkeys and in renal tubular cells of rats. It is conceivable that the basophilic material represents lumasiran or some degradation products of it. Kupffer cells resemble macrophages and could therefore be involved in scavenging lumasiran.

In the study of Janas et al. 2018, the basophilic granules in Kupffer and tubular kidney cells were also addressed as well as the finding of vacuolated macrophages in certain lymph nodes. The authors performed immunohistochemistry staining using an antibody directed against a common part of therapeutic siRNA like lumasiran. The results indicate that the basophilic granules in liver and Kidney as well as the vacuoles in the macrophages contain siRNA-related material.

Lumasiran did not show any adverse effects on male and female fertility and pre- and post-natal development in rats. In embryo-foetal development studies in rats and rabbits, skeletal abnormalities were observed, but at high exposure multiples relative to human therapeutic exposures. The NOAELs were approximately 20 to 70 times higher (based on monthly exposures). Animal studies do not indicate direct or indirect harmful effects with respect to reproductive toxicity. The use of this medicinal product could be considered during pregnancy taking into account the expected health benefit for the woman and potential risks to the foetus.

In pre- and postnatal development in rats lumasiran plasma concentrations were measured in suckling pups of lumasiran treated dams. Lumasiran plasma levels were found to be below the level of quantification. It is unknown whether lumasiran is excreted in human milk.

A dose range finding toxicity study in neonate rats did not show increased sensitivity of the developing rat to either the toxicology or pharmacology of lumasiran at exposure multiples of 2 compared to human therapeutic exposures (based on monthly exposures).

Lumasiran is not mutagenic or clastogenic and is not considered to have an immunostimulatory or immunotoxicity potential. There were no findings in these studies or in any published studies investigating the suppression of GO that raise a pharmacologically based cause for concern regarding carcinogenic potential. However, it is recommended that the applicant further investigates the carcinogenic potential of lumasiran following the authorisation of the product.

The applicant conducted a repeated-dose study in rats with a drug substance batch containing commercial scale impurities which were not present in the batches used for the other toxicology studies. The findings in this study were in line with the findings of the other repeated-dose rat studies.

Lumasiran PEC-surface water value is below the action limit of 0.01 µg/L and is not a PBT substance as log Kow does not exceed 4.5. Lumasiran is therefore not expected to pose a risk to the environment.

2.3.7. Conclusion on the non-clinical aspects

There are no objections to the marketing authorization of Oxlumo from a non-clinical perspective.

2.4. Clinical aspects

2.4.1. Introduction

GCP

The Clinical trials were performed in accordance with GCP as claimed by the applicant.

The applicant has provided a statement to the effect that clinical trials conducted outside the Community were carried out in accordance with the ethical standards of Directive 2001/20/EC.

- Tabular overview of clinical studies

Study, Status, Data Cutoff	Study Design / Objectives, Location	Patient Population	Dosage Regimen: Number of Patients
ALN-GO1-001 (Study 001) *Completed:* *23 Jan 2019*	Phase 1/2, randomised, single blind, placebo-controlled **Part A:** SAD study (001A) in healthy subjects to evaluate safety, tolerability, PK, PD and ADA of lumasiran Country: UK **Part B:** MAD study (001B) in patients with PH1, with the addition of 2 open-label extension cohorts, to evaluate safety, tolerability, PK, PD, and ADA of lumasiran Countries: France, Germany, Israel, Netherlands, UK	Healthy volunteers, 18-64 years	Placebo: N=8 Lumasiran: N=24 0.3 mg/kg (N=6) 1.0 mg/kg (N=6) 3.0 mg/kg (N=6) 6.0 mg/kg (N=6)
		Adults and children (6-64 years) with PH1	Placebo: N=3 Lumasiran: N=17 1.0 mg/kg qM x 3 (N=7) 3.0 mg/kg qM x 3 (N=7) 3.0 mg/kg q3M x 2 (N=3) <u>Open-Label Period</u> Lumasiran: N=20 The 3 placebo patients subsequently received open-label Lumasiran: 1.0 mg/kg qM x 3 (N=8) 3.0 mg/kg qM x 3 (N=8) 3.0 mg/kg q3M x 1 (N=4)[a]
ALN-GO1-002 (Study 002) *Ongoing* *Data cutoff:* *30 Jan 2020*	Phase 2, open-label, long-term extension study for up to 54 months to evaluate the long-term safety, efficacy, PK, PD and ADA of lumasiran Countries: France, Germany, Israel, Netherlands, UK	PH1 patients who completed Study 001B	Lumasiran: 1 mg/kg or 3 mg/kg SC, once monthly or quarterly (N=20) 1 mg/kg qM (N=3); all 3 patients transitioned to 3 mg/kg q3M by Month 6 3 mg/kg qM (N=7) 3 mg/kg q3M (N=10)[b]

Study, Status, Data Cutoff	Study Design / Objectives, Location	Patient Population	Dosage Regimen: Number of Patients
ALN-GO1-003 (Study 003) *Ongoing (6-Month double-blind treatment period [Primary Analysis] completed; Extension period ongoing).* *Data cutoff: 01 May 2020*	Phase 3, randomized (2:1), double-blind (DB), placebo-controlled study with an extended dosing period to evaluate the efficacy and safety of lumasiran DB period (Primary Analysis): 6 months 3-Month Blinded Treatment Extension period: Month 6 to Month 9 OLE period: Month 9 to Month 60 Countries: US, France, Germany, Israel, Netherlands, Switzerland, United Arab Emirates, UK	Adults and children ≥6 years with PH1	**DB period (**Primary Analysis)**:** Placebo: N=13 Lumasiran: N=26 Loading dose: 3 mg/kg qM for a total of 3 doses Maintenance dose: 3 mg/kg q3M **Extension period[c]:** Placebo patients cross over to receive lumasiran 3 mg/kg qM for 3 consecutive months; then, 3 mg/kg q3M Lumasiran patients receive lumasiran 3 mg/kg q3M
ALN-GO1-004 (Study 004) *Ongoing* *Data cut-off: 09 March 2020*	Phase 3 open-label single-arm study to evaluate the efficacy, safety, PK, PD and ADA of lumasiran Countries: US, France, Germany, Israel, UK	Infants and children <6 years of age with PH1	Weight-based dosing regimen: N=18 <10 kg: 6 mg/kg qM for 3 months (loading dose) followed by 3 mg/kg qM (maintenance dose) (N=3) 10 to <20 kg: 6 mg/kg qM for 3 months (loading dose) followed by 6 mg/kg q3M (maintenance dose) (N=11) ≥20 kg: 3 mg/kg qM for 3 months (loading dose) followed by 3 mg/kg q3M (maintenance dose) (N=2) Dosing up to 60 months
ALN-GO1-005 (Study 005) *Ongoing* *Data cut-off:14 May 2020*	Phase 3 open-label single-arm study to evaluate the efficacy, safety, PK, and PD of lumasiran	Patients of all ages with advanced kidney disease	N=4 (at time of data cut-off)

2.4.2. Pharmacokinetics

No absolute bioavailability and human absorption, distribution, metabolism, and excretion (ADME) studies have been performed for lumasiran because of the long tissue half-life in liver and the consequent health hazard from prolonged exposure to radioactivity in healthy subjects.

The PK of lumasiran has been evaluated in 4 submitted clinical studies (001, 002, 003 and 004). Preliminary data on PK of lumasiran from study 005 in 4 patients with ESRD requiring haemodialysis were additionally submitted.

Population pharmacokinetic modelling

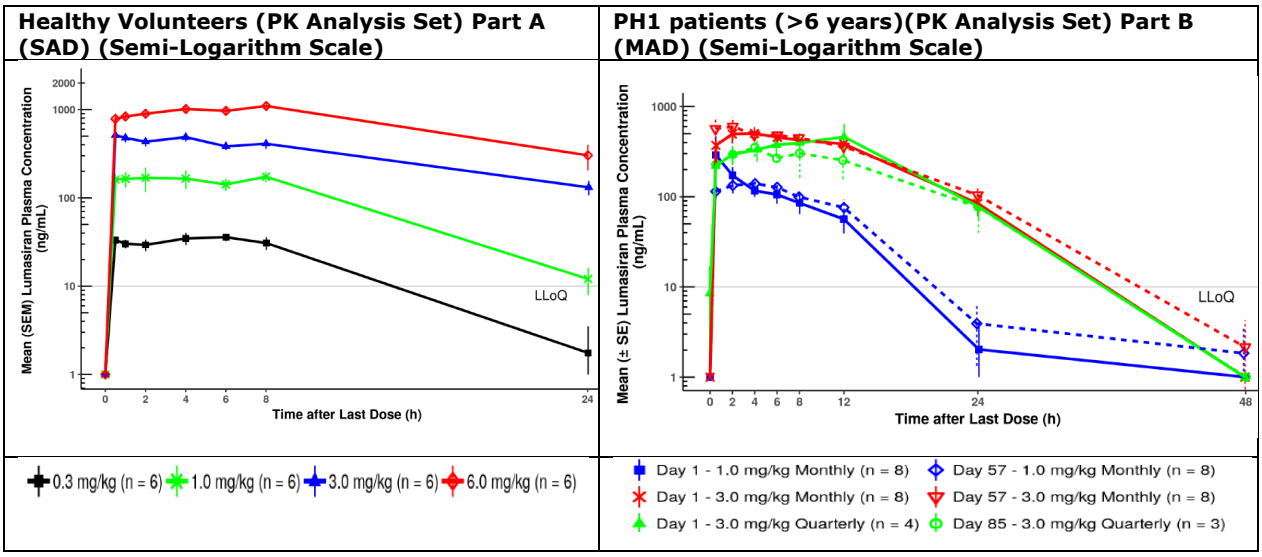
A population pharmacokinetic analysis for plasma lumasiran concentrations was performed with data from a total of 99 subjects, 24 healthy adult volunteers (24.2%) and 75 PH1 patients (75.8%) from Studies 001, 002, 003, and 004. Model development revealed that lumasiran PK was best described by a 2-compartment disposition model with first-order absorption process from the SC injection site into

systemic circulation. Total plasma clearance was a sum of two elimination pathways: hepatic uptake clearance and renal clearance. The covariate effect of body weight on PK parameters was incorporated using fixed physiologically based allometric exponents. Renal clearance for lumasiran was fixed to the patient's baseline renal function. Random effects were added to parameters hepatic clearance and absorption rate constant (Ka) to describe inter-individual variabilities. Inter-occasion variability was added on Ka to explain random PK differences observed over multiple occasions of PK sampling within a subject. Estimation of PK parameters was precise, goodness-of-fit plots did not reveal any model-misspecification and the VPC showed that concentrations were adequately predicted.

Absorption

Absolute bioavailability of lumasiran following subcutaneous (SC) administration has not been determined in humans. Mean plasma concentration-time profiles of lumasiran in healthy volunteers after the administration of a single SC dose and in PH1 patients after initial and multiple administration of different doses in study 001B are presented in **Figure 7**, showing transient plasma exposures.

Figure 7. Study 001: Mean (+/- SEM) plasma lumasiran concentration (ng/mL) versus time profiles



LLOQ=lower limit of quantification; SEM=standard error of the mean;
Concentrations below LLoQ (10 ng/mL)were imputed to 0 ng/mL and are displayed as 1 ng/mL for this scale.

A summary of lumasiran PK parameters across studies is presented in **Table 16**.

Table 16. Summary of lumasiran PK parameters across studies (non-compartmental analysis)

PK Parameters	Study 001A				Study 001B[a]		Study 003	Study 004	
	0.3 mg/kg	1 mg/kg	3 mg/kg	6 mg/kg	1 mg/kg	3 mg/kg[b]	3 mg/kg	3 mg/kg	6 mg/kg
C_{max} (ng/mL)									
Mean (%CV)	39.8 (21.6%)	204 (54.6%)	533 (30.0%)	1180 (17.0%)	324 (151.1%)	532 (48.7%)	513 (60.4%)	1300 (105.7%)	1020 (33.6%)
Median (min-max)	36.0 (31.4-54.9)	167 (134-428)	518 (302-743)	1210 (840-1420)	156 (66.1-1520)	529 (205-1130)	438 (38.5-1500)	1300 (329-2280)	912 (523-1760)
	(n=6)	(n=6)	(n=6)	(n=6)	(n=8)	(n=12)	(n=38)	(n=2)	(n=14)
AUC_{0-24} (ng.h/mL)									
Mean (%CV)	NA	2650 (56.6%)	7200 (15.6%)	16800 (26.0%)	1740 (46.4%)	7120 (40.9%)	NA	NA	9230 (24.9%)
Median (min-max)	429 (429-429)	2230 (1730-5290)	7350 (5620-8560)	16300 (11300-24700)	1690 (855-3030)	8430 (2890-10700)	NA	4370 (4370-4370)	8510 (5920-13300)
	(n=1)	(n=5)	(n=6)	(n=6)	(n=7)	(n=11)	NA	(n=1)	(n=11)
AUC_{last} (ng.h/mL)									
Mean (%CV)	294 (33.0%)	1900 (29.4%)	7210 (15.6%)	16800 (26.1%)	1430 (48.9%)	7050 (39.5%)	NA	5860 (35.8%)	8810 (25.1%)
Median (min-max)	258 (193-430)	990 (945-2510)	7370 (5620-8570)	16300 (11300-24800)	1180 (618-2380)	7400 (2890-10700)	NA	5860 (4370-7340)	7960 (5920-13300)
	(n=6)	(n=6)	(n=6)	(n=6)	(n=8)	(n=12)	NA	(n=2)	(n=14)
Half-life (h)									
Mean (%CV)	7.07 (5.3%)	NA	5.98 (25.5%)	NA	3.27 (46.8%)	5.85 (55.9%)	NA	NA	5.24 (44.6%)
Median (min-max)	7.07 (6.80-7.33)	NA	5.98 (4.90-7.06)	3.47 (3.47-3.47)	3.29 (1.70-4.79)	5.54 (2.08-10.9)	NA	1.42 (1.42-1.42)	4.98 (2.61-10.3)
	(n=2)	NA	(n=2)	(n=1)	(n=4)	(n=6)	NA	(n=1)	(n=10)
t_{max} (h)									
Mean (%CV)	5.34 (30.5%)	3.42 (104.7%)	3.17 (89.7%)	5.76 (52.8%)	3.34 (50.2%)	6.36 (63.1%)	NA	4.53 (80.1%)	3.73 (54.2%)

Median (min-max)	5.02 (4.00-8.02)	1.50 (0.517-8.00)	3.00 (0.500-8.00)	7.00 (0.500-8.07)	3.99 (0.567-5.97)	6.00 (0.533-12.0)	NA	4.53 (1.97-7.10)	3.74 (1.93-8.10)
	(n=6)	(n=6)	(n=6)	(n=6)	(n=8)	(n=12)	NA	(n=2)	(n=14)
PK Parameters	**Study 001A**				**Study 001B[a]**		**Study 003**	**Study 004**	
	0.3 mg/kg	**1 mg/kg**	**3 mg/kg**	**6 mg/kg**	**1 mg/kg**	**3 mg/kg[b]**	**3 mg/kg**	**3 mg/kg**	**6 mg/kg**
Vz/F (L)									
Mean (%CV)	NA	389 (28.3%)	219 (35.7%)	NA	116 (76.6%)	181 (82.3%)	NA	NA	64.0 (59.5%)
Median (min-max)	NA	389 (311-467)	219 (164-274)	139 (139-139)	111 (27.9-213)	143 (32.8-427)	NA	20.2 (20.2-20.2)	54.0 (29.6-152)
	NA	(n=2)	(n=2)	(n=1)	(n=4)	(n=6)	NA	(n=1)	(n=10)
CL/F (L/h)									
Mean (%CV)	NA	37.9 (23.2%)	25.0 (10.6%)	NA	21.6 (45.3%)	18.8 (37.2%)	NA	NA	8.20 (20.4%)
Median (min-max)	NA	37.9 (31.7-44.1)	25.0 (23.1-26.9)	27.8 (27.8-27.8)	20.5 (11.4-34.1)	17.9 (10.9-27.3)	NA	9.89 (9.89-9.89)	8.40 (5.63-10.2)
	NA	(n=2)	(n=2)	(n=1)	(n=4)	(n=6)	NA	(n=1)	(n=10)
Fe_{0-24} (%)									
Mean (%CV)	17.4 (14.0%)	19.1 (20.4%)	21.0 (25.5%)	25.8 (12.6%)	11.1 (33.7%)	9.73 (54.4%)	NA	NA	NA
Median (min-max)	18.2 (12.7-19.4)	19.0 (14.4-24.6)	21.7 (11.7-26.4)	25.6 (21.3-30.2)	9.53 (7.21-17.8)	8.59 (3.18-22.3)	NA	NA	NA
	(n=6)	(n=6)	(n=6)	(n=5)	(n=7)	(n=11)	NA	NA	NA
CL_R (L/h)									
Mean (%CV)	NA	5.49 (37.8%)	5.82 (22.6%)	6.34 (18.2%)	2.26 (52.0%)	2.25 (49.1%)	NA	NA	NA
Median (min-max)	8.78 (8.78-8.78)	5.65 (2.49-8.33)	6.47 (3.56-6.84)	6.50 (4.52-7.57)	1.87 (1.45-4.50)	2.15 (0.547-3.94)	NA	NA	NA
	(n=1)	(n=5)	(n=6)	(n=5)	(n=6)	(n=10)	NA	NA	NA

Abbreviations: AUC0-24= area under the plasma concentration versus time curve from 0 to 24 hours; AUClast=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; CLR=renal clearance; Cmax=maximum plasma concentration; CV=coefficient of variation; Fe0-24=fraction excreted in urine from time 0 to 24 hours; max=maximum; min=minimum; NA=not available/not calculated; PK=pharmacokinetics; qM=every month; q3M=every 3 months; tmax=time to reach maximum plasma concentration; VZ/F=terminal phase extravascular volume of distribution.

Notes: for Study 003 AUC and half-life could not be calculated because of sparse data and 4h post-dose was used as Cmax. All values were based on Day 1 of lumasiran.

a Includes placebo patients who crossed over to lumasiran

- **Influence of food**

No studies were conducted to evaluate drug-food interactions because lumasiran is administered subcutaneously.

Distribution

Plasma protein binding was moderate to high (77 to 85%) at clinically relevant concentrations in plasma from adult healthy donors.

No specific human distribution studies were conducted. Distribution was thus informed based on non-clinical data (See Non-clinical section of this report).

The lumasiran volume of distribution estimate following IV dosing in cynomolgus monkeys from nonclinical study GO1-DSM15-020 was used to determine the body weight based central volume of distribution (V2) for model building in the population PK analysis. The same relationship between volume of distribution and body weight was assumed in man. For a typical 70 kg adult, this yielded a population estimate for the apparent central volume of distribution (Vd/F) of 4.9 L (0.07 L/kg*body weight) for lumasiran.

Elimination

Lumasiran in plasma was below the Lower Limit of Quantitation (LLoQ) within 24 to 48 hours in most patients with PH1 (study 001B) (**Figure 7**).

Following a single oral SC dose of lumasiran, elimination half –life in plasma was evaluable in only five adult healthy subjects where it ranged from 3.47 to 7.33 hours with a mean (%CV) of 5.91 (28.2%) hours (study 001A). Apparent total clearance (CL/F) in these subjects ranged from 23.1 to 44.1 L/h with a mean (%CV) of 30.7 (26.3%) L/h.

In paediatric and adult patients with PH1 (≥6 years), half-life was more variable and ranged from 2.08 to 10.9 hours with a mean (%CV) of 5.16 (55.5%) hours in study 001B (n=9) and similarly from 1.42 to 10.3 hours with a mean of 4.89 (51.1%) hours in paediatric PH1 patients (<6 years) in study 004 (n=11). CL/F ranged lower from 10.9 to 34.1 L/h with a mean (CV%) of 20.9 (36.6%) L/h in study 001B (and from 5.63 to 10.2 L/h with a mean (CV%) of 8.35 (19.9%) L/h in study 004.

In the combined analysis of adult healthy subjects and paediatric and adult PH1 patients across studies, mean half-life (CV%) following a single SC dose of lumasiran was estimated 5.19 (47.0%) hours (n=57). And mean CL/F (CV%) was estimated as 17.3 L/h (60.7%).

Comparable to these observed values, model-estimated t1/2 ranged from 3.7 to 8.8 hours and CL/F ranged from 4.6 to 32.4 L/h across body weight groups in the population PK analysis.

Based on non-clinical studies, the majority of lumasiran is taken up by the liver, the target organ of effect. PK/PD modelling predicted a lumasiran terminal half-life in human liver to be 66.9 days.

- **Excretion**

In humans, excretion of lumasiran into the bile and faeces was not determined.

A mean of 17.4 to 25.8% of the administered dose was excreted in the urine as unchanged lumasiran in healthy subjects over 24 hours (study 001A) **(Table 16)**.

Compared to healthy subjects, the mean fraction of administered lumasiran excreted in urine in patients with PH1 was lower. Mean $Fe_{0\text{-}24}$ (%) ranged from 7.17 to 13.7% following single or multiple SC administration of lumasiran in study 001B. This difference was explained with the lower eGFR values in PH1 patients and was predicted not to influence the PK of lumasiran, since the majority of lumasiran is cleared from plasma by hepatic uptake.

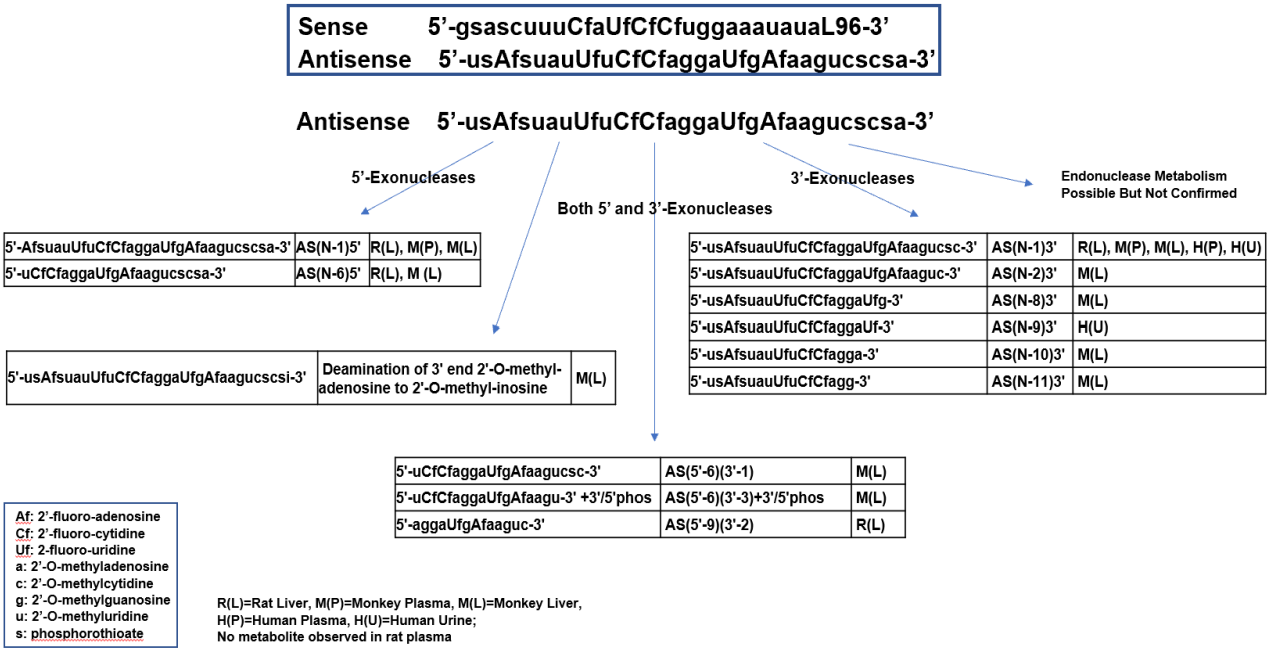
Metabolism

The metabolism of lumasiran was investigated *in vitro* in human, mouse, rat and monkey serum and liver S9 fractions, and *in vivo* in rat and monkey plasma and liver samples obtained from nonclinical PK studies GO1-DSM15-022 and GO1-DSM15-020 and *in vivo* in human plasma and urine samples obtained from the single 6.0 mg/kg dose cohort (highest tested dose) in Study 001 (study BA15014).

Lumasiran is metabolised primarily by nucleoside cleavage by exo- or endonucleases. While the sense strand is metabolised by sequential removal of GalNAc moieties and removal of the 3' nucleotide, the antisense (AS) strand is primarily metabolised to form AS(N-1)3' lumasiran. This pharmacologically active metabolite was <10% of lumasiran in both plasma and urine in healthy subjects on lumasiran. A further deaminated metabolite pharmacologically equipotent to lumasiran may form in human liver, as observed in monkey liver, but was not detected in human plasma or urine. Lumasiran was not a substrate of CYP enzymes *in vitro*.

The proposed biotransformation pathways of the lumasiran AS strand is presented in **Figure 8**.

Figure 8. Proposed biotransformation pathways of lumasiran antisense strand in rats, monkeys, and in humans



Dose proportionality and time dependencies

In the assessment of lumasiran pharmacokinetics (PK), dose proportionality analyses were exploratory in nature. While Cmax and AUC0-inf showed a linear increase, AUClast and AUC0-24 increased slightly

Accumulation ratios were calculated using data from Studies 001B and 003 where patients received repeated doses of lumasiran. Across studies and dosing schedules, the mean and median accumulation ratio ranged from 0.86 to 1.29, confirming that lumasiran does not accumulate in plasma following repeated dosing, consistent with the short mean plasma half-life of 3 to 8 hours relative to monthly and once every 3 month dosing schedules evaluated in clinical studies.

Special populations

Impaired renal function

In the pooled PH1 population, 30, 30, and 11 subjects treated with lumasiran had normal renal function (eGFR ≥90 mL/min), mild renal impairment (eGFR 60 to <90 mL/min), and moderate renal impairment (eGFR 30 to <60 mL/min), respectively, at baseline. There were no patients with severe renal impairment in the lumasiran clinical studies used in this comparison.

Lumasiran Cmax values were available in 11 subjects with moderate renal impairment, while only 2 moderate renal impairment patients contributed AUC values. This is because 9 of the 11 subjects were enrolled in Study 003 where AUC values could not be calculated.

There was no trend in C_{max}; a trend toward higher lumasiran $AUC_{0\text{-last}}$ values in patients with moderate renal impairment was noted however the small sample size for this analysis (n=2) did not allow to draw any robust conclusions.

The population PK analysis, included 44 subjects (44.4%) with normal renal function, 44 subjects (44.4%) with mild renal impairment, and 11 subjects (11.1%) with moderate renal impairment at baseline.

Renal clearance for lumasiran was fixed to the patient's baseline renal function (estimated glomerular filtration rate [eGFR]*body surface area [BSA]/1.73). In patients with normal renal function, the model estimated renal elimination to account for 20% of the CLP with 80% of administered lumasiran available for liver uptake. Renal impairment is predicted to result in lowering of urinary excretion of urine (<20%) leading to transiently higher circulating plasma concentrations and consequently a higher fraction available for liver uptake (>80%).

In addition, lumasiran is being evaluated in patients with end-stage renal disease (ESRD) in Study 005. Preliminary PK data from this study are summarised in **Table 17**.

Table 17*:* Lumasiran plasma PK parameters in the 4 patients enrolled in Study 005 (Pharmacokinetic Analysis Set, Study 005)

Age (years)	Number of Doses	Dose (mg/kg)	t_{max} (h)	t_{last} (h)	C_{max} (ng/mL)	$AUC_{0\text{-last}}$ (h·ng/mL)	$t_{1/2}$ (h)	CL/F (L/h)
3	2	6	2.1	10.1	2970	13191	1.13	3.69
6	2	6	2.0	9.9	1180	6286	3.43	8.06
6	3	6	4.0	10.3	11500	50909	NA	NA
16	2	3	10.0	21.1	1360	16736	NA	NA

Abbreviation: $AUC_{0\text{-last}}$= 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 C_{max}=maximum plasma concentration, NA=not available (parameters could not be calculated), PK=pharmacokinetic, $t_{½}$=elimination half-life, t_{last}=time of last measurable concentration, t_{max}=time to reach maximum plasma concentration.

Impaired hepatic function

To categorise hepatic function, NCI-ODWG liver dysfunction criteria as defined in the following were used: **Normal**: total bilirubin ≤ ULN and AST ≤ ULN; **Mild**: (total bilirubin ≤ ULN and AST > ULN) or (ULN < total bilirubin ≤ 1.5 x ULN); **Moderate**: 1.5 x ULN < total bilirubin ≤ 3 x ULN.

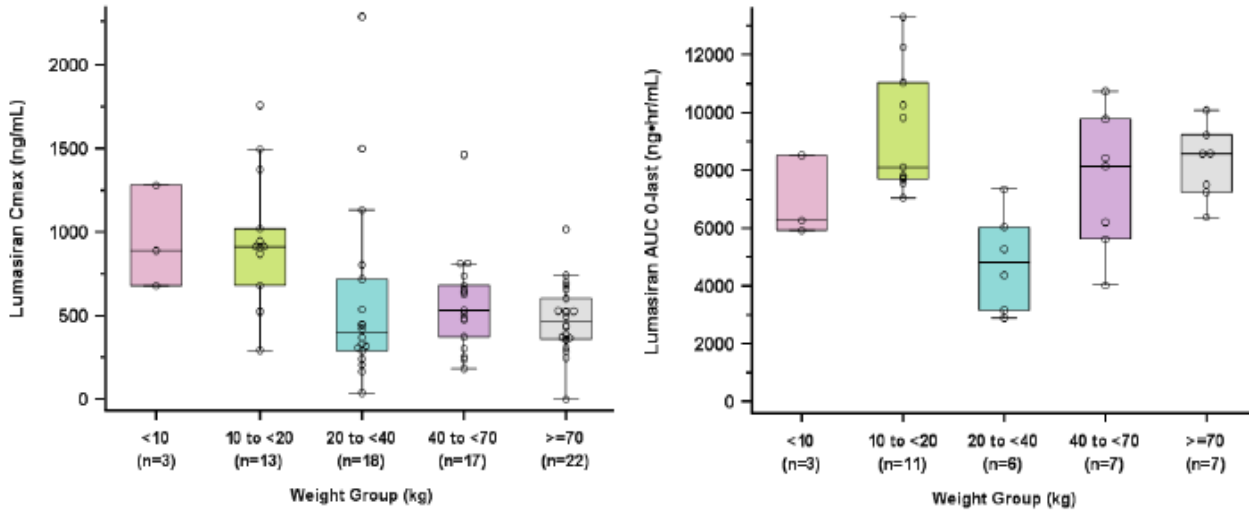
According to this classification, in the pooled population of patients with PH1 treated with the recommended doses of lumasiran, 70 patients had normal hepatic function, 2 patients from the placebo arm of Study 003 had mild hepatic impairment; and one patient from Study 001 had moderate hepatic impairment. Due to sparse sampling in Study 003, AUC data were not available for the 2 patients with mild hepatic impairment

Median steady-state plasma Cmax and AUC0-last values for lumasiran were comparable across hepatic function groups.

Weight

The influence of body weight on Cmax and AUC of lumasiran following treatment with the recommended dose across pre-defined weight groups is summarised in **Figure 9**.

Figure 9. Comparison of lumasiran Cmax and AUC across studies by body weight group in PH1 subjects treated with phase 3 regimens of lumasiran (Pharmacokinetic Analysis Set)



Note: Assessment values were from Baseline/Day 1 visit. Cmax values in Study 003 were lumasiran plasma concentrations at 4 hours post-dose (Baseline/Day 1).

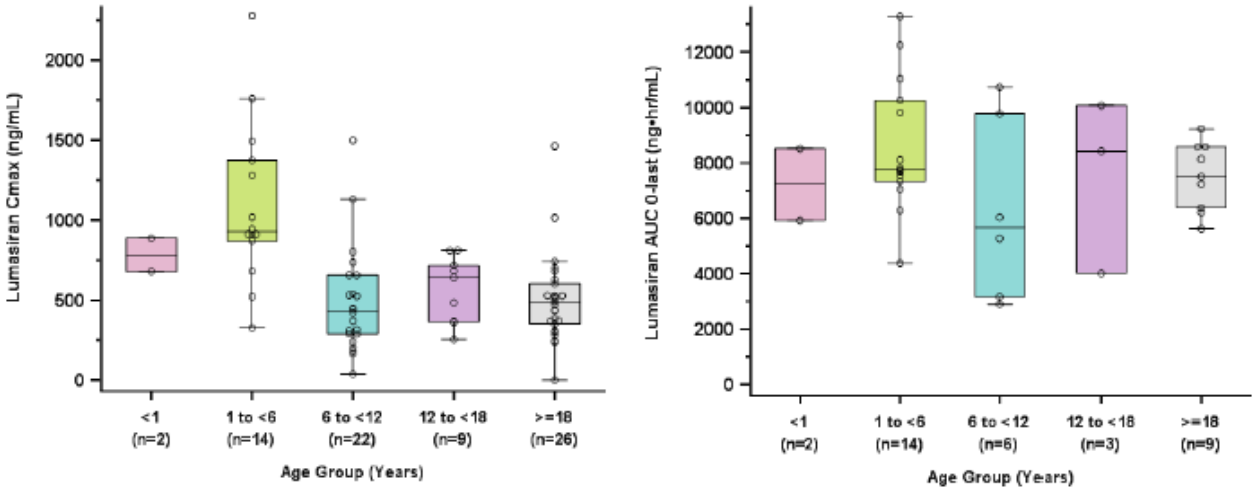
In the database included in the population PK analysis, body weight ranged from 6.2 to 110 kg, with median of 55.0 kg. Population PK analysis revealed that body weight had an impact on hepatic clearance and intercompartmental clearance, central and peripheral volume of distribution and ka. Absorption rate constant decreased with increasing body weight and the other parameters increased with increasing body weight, whereas body weight corrected clearance decreased with increasing body weight.

Age

PK data from 26 adults aged ≥18 years and 47 adolescents and children aged <1 year to <18 years (overall age range: 4 months to 60 years) were available. No elderly patients were included in the development programme.

The effect of age was evaluated by comparing lumasiran PK and PD in patients in predefined age categories (**Figure 10**).

Figure 10: Comparison of lmasiran Cmax and AUC across studies by age in PH1 subjects treated with Phase 3 regimens of lumasiran (Pharmacokinetic Analysis Set)



Note: Assessment values were from Baseline/Day 1 visit. Cmax values in Study 003 were lumasiran plasma concentrations at 4 hours postdose (Baseline/Day 1).

Pharmacokinetic interaction studies

No dedicated *in vivo* DDI studies were submitted.

Concomitant use of pyridoxine (Vitamin B6)

Pyridoxine (Vitamin B6) is a current non-approved therapy to treat PH1. Pyridoxine is primarily metabolised in the liver, with the major metabolite being 4-pyridoxic acid, which is inactive and excreted in urine.
Comparison of the PK of lumasiran in patients with and without concomitant pyridoxine using pooled data from Studies 001A, 001B, 003, and 004 revealed a slight tendency for increased lumasiran exposure with concomitant use of Vitamin B6. In the patients with concomitant use of Vitamin B6, median Cmax and AUC0-last of lumasiran was 623 ng/mL and 8270 h•ng/mL, compared to 486 ng/mL and 7140 h•ng/mL, respectively, without concomitant use of Vitamin B6.

2.4.3. Pharmacodynamic

The applicant has conducted as the main source of dose-finding and dose determination the studies 001 and 002, which have been described above. In addition, dose-finding and dose-determination for the product was also based on an extensive PK (described in the previous section) and PK-PD modelling exercise.

Population PKPD modelling

A PKPD model was developed to quantify the relationship between predicted human liver PK and pharmacodynamic response observed as plasma glycolate elevation in adult healthy volunteers and urinary oxalate reduction in PH1 patients ≥6 years of age. Clinical data available as of 20 March 2018 were used in the development and validation of the PK/PD model. A total of 277 observed plasma glycolate measurements up to 17 months post dose were available at the time of model development.

The population PK/PD model was used to simulate steady state urinary oxalate profiles at 1.0 to 6.0 mg/kg dose levels following continuous qM or q3M SC administrations. Simulation results showed dose dependent oxalate reductions over the dose range studied, with diminishing additional effect predicted with higher doses consistent with an asymptotic rather than linear relationship. A 3.0 mg/kg q3M dose of lumasiran is predicted to suppress UOx to near normal levels with median predicted value of 0.44 (90% PI: 0.32- 0.73) mMol/24-hour/1.73m2 at steady-state. Thus, quarterly doses of at least 3.0 mg/kg are needed to reduce urinary oxalate levels to near or within normal.

When more clinical data were available, two additional PKPD models were developed: a model for 24-hour urinary oxalate corrected for BSA in PH1 patients ≥6 years of age, and a model for spot urinary oxalate:creatinine ratio in PH1 patients across all ages from infants to adults including data from studies 001 Part B, Study 002, Study 003, and Study 004.

For the first model, the significant covariates were the effect of eGFR on baseline 24-hour urinary oxalate corrected for BSA and baseline 24-hour urinary oxalate corrected for BSA on Imax.

For the second model, significant parameter-covariate relationships were time-varying height and baseline eGFR for the baseline spot urinary oxalate:creatinine ratio and baseline spot urinary oxalate:creatinine ratio for Imax.

Mechanism of action

Lumasiran targets the 3’ untranslated region of HAO1 mRNA in the liver, thereby preventing the synthesis of the corresponding GO protein. Lumasiran is designed with a GalNAc moiety conjugated to the sense strand of the siRNA to enable selective delivery to the liver via uptake by the asialoglycoprotein receptor (ASGPR). ASGPRs are expressed primarily and abundantly (0.5 to 1 million per cell) on the cell surface of hepatocytes and specifically bind to the glycoproteins with terminal galactose or GalNAc residues.

Binding of the GalNAc ligand of lumasiran and ASGPR triggers receptor-mediated endocytosis of the ligand-receptor complex, resulting in release of the siRNA into the cytoplasm of the hepatocyte. ASGPR is subsequently recycled to the cell surface and is available for successive uptake of circulating GalNAc-conjugated siRNA.

Upon delivery to the liver, the double-stranded lumasiran siRNA is loaded into the cellular multiprotein enzyme cleavage complex known as the RNA-induced silencing complex (RISC) in the cytosol. Once loaded, the antisense strand (guide strand) of lumasiran binds to the complementary sequence in the HAO1 mRNA. Pairing of HAO1 mRNA with the antisense strand within the RISC/siRNA complex results in cleavage of the HAO1 mRNA, thereby preventing the synthesis of the corresponding GO protein, which in turn reduces – by reducing the target substrate of GO, glyoxylate – the production of oxalate in the liver. The underlying enzymatic defect (relating to AGT) is not targeted with the proposed treatment.

Primary and Secondary pharmacology

PD in healthy volunteers:

32 healthy volunteers were included in study 001A and the evaluation of the plasma glycolate levels showed a clear dose dependent increase over time, with a plateau being reached after about 6 weeks after the single dose administered with stable levels to the end of the administration period. No effects could be seen with the lowest dose of 0.3 mg/kg BW.

PD in patients:

This refers to the Part B of study 001 which was the MAD part of the study in PH1 patients with relatively preserved renal function (defined as estimated glomerular filtration rate [eGFR] >45 mL/min/1.73m2).

In this trial, patients were randomised 3:1 to lumasiran or placebo into the dosing cohorts 1.0 mg/kg once monthly, 3.0 mg/kg once monthly, or 3.0 mg/kg once every 3 months, and all patients received the first dose of lumasiran on Day 1. Treatment was for a primary period of 85 days, with a switch of placebo-treated patients afterwards, and a prolonged observation period for all patients. Long-term observations were evaluated for an observation period of 197 day altogether.

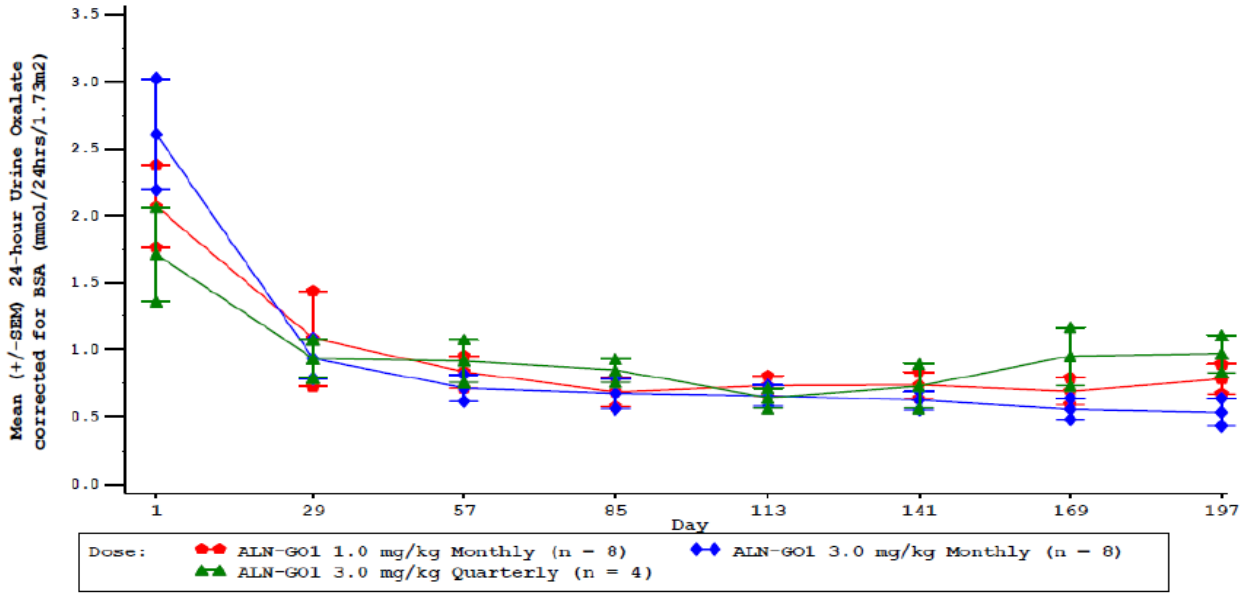
Results on the reduction of urinary oxalate from this study are summarised in **Table 18**.

Table 18. 24-Hour urinary oxalate corrected for BSA (mmol/24h/1.73m2) and percent change from baseline, Part B (Placebo Comparison Analysis Set [Day 1 to Day 85])

Time Point Statistic	Placebo (n=3)	Lumasiran			
		1.0 mg/kg qM (n=7)	3.0 mg/kg qM (n=7)	3.0 mg/kg q3M (n=3)	All Treated (N=17)
Baseline, n	3	7	7	3	17
Mean (SD)	1.96 (0.321)	1.67 (0.734)	1.78 (0.647)	1.35 (0.413)	1.66 (0.636)
Day 29, n	1	6	7	3	16
Mean (SD)	1.95 (NR)	0.91 (0.519)	0.77 (0.366)	0.71 (0.257)	0.81 (0.398)
Mean (SD) % change	-2.4 (NR)	-42.0 (27.00)	-57.5 (10.84)	-47.4 (5.06)	-49.8 (18.62)
Day 57, n	2	6	7	3	16
Mean (SD)	1.37 (1.047)	0.71 (0.293)	0.51 (0.256)	0.65 (0.143)	0.61 (0.258)
Mean (SD) % change	-27.8 (47.11)	-50.5 (21.86)	-71.1 (10.77)	-51.1 (4.99)	-59.6 (17.84)
Day 85, n	1	7	7	2	16
Mean (SD)	2.18 (NR)	0.56 (0.236)	0.56 (0.242)	0.76 (0.134)	0.58 (0.227)
Mean (SD) % change	9.1 (NR)	-64.6 (17.71)	-68.4 (10.60)	-51.9 (3.87)	-64.7 (14.13)

Results for the lumasiran-Treated Analysis Set (Day 1 to Day 197, **Figure 11**) were consistent with the placebo-comparison dataset, indicating maintenance of effect with repeat dosing. The overall mean percent reduction was greatest for the 3.0 mg/kg once monthly dose regimen compared to the 1.0 mg/kg once monthly and the 3.0 mg/kg once every 3 months regimen at each time point from Day 29 to Day 197, indicating a dose dependent response in 24-hour urinary oxalate levels to lumasiran.

Figure 11. Study 001: 24-hour urine oxalate corrected for BSA over time by dose cohort, Part B (Pharmacodynamic Analysis Set: Single-Blind Period)



Reduction in 24-hour urine oxalate:creatinine ratio was evident by Day 29 in lumasiran-treated patients, while no change was observed in placebo-treated patients. A greater reduction in 24-hour urine oxalate:creatinine ratio was observed with 3 mg/kg qM compared to other dose cohorts at each time point. The reduction of 24-hour urine oxalate:creatinine ratios was sustained through Day 197, with some recovery observed for the 1 mg/kg qM and 3 mg/kg quarterly groups on Day 197.

PH1 patients treated with lumasiran showed a decrease in plasma oxalate levels, consistent with the reduction in urine oxalate excretion. Patients treated with lumasiran had an overall mean percent reduction in plasma oxalate of 48.4% (n=15) at Day 57 and 44.0% (n=18) at Day 85. For patients initially treated with lumasiran, the reduction in plasma oxalate observed during the blinded treatment period, Day 1 to Day 85, was maintained through Day 113 for the 3 mg/kg q3M dose group.

By Day 29, all patients in the lumasiran groups had an observable increase in 24-hour urine glycolate:creatinine ratios: 37.3%, 36.8% and 26.7% in the 1 mg/kg qM, 3 mg/kg qM and 3 mg/kg q3M groups, respectively. Patients in the 1 mg/kg qM group had the highest mean increase at Day 57 (79.7%). All dose groups showed a sustained increase in 24-hour urinary glycolate:creatinine ratios through Day 85.

Increases in spot urine glycolate:creatinine ratios were observed by Day 15, with continued increase over the dosing period that ranged from 16.8% (Day 15) to 208.3% (Day 57). In the placebo-treated patients, the glycolate:creatinine ratios remained relatively stable during the 85-day treatment period

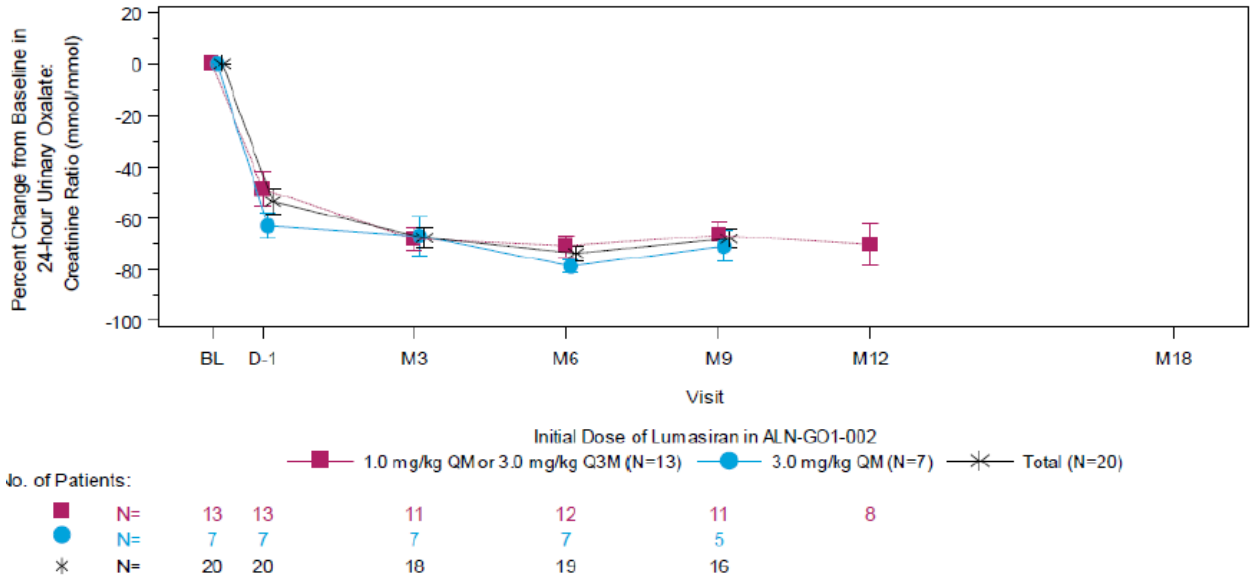
Plasma glycolate levels increased in patients treated with lumasiran. By Day 29, a mean increase in plasma glycolate of 23.6% and 33.0% was observed in the 1 mg/kg qM and 3 mg/kg qM groups, respectively. Data were not available for patients in the 3 mg/kg q3M group since plasma PK samples (which served in this study as samples for plasma glycolate analysis re-testing using a validated analysis [refer to Study 001 CSR addendum 1-erratum 1]) were not drawn for this cohort on Day 29. Overall, the magnitude of plasma glycolate increase was similar to that seen in urine and did not show a dose-response.

PD activity was also shown for the long-term treatment in Study 002. This is an ongoing Phase 2, Multicenter, Open-Label, Extension Study to Evaluate the Long-Term Administration of ALN-GO1 in Patients with Primary Hyperoxaluria Type 1. Eligibility of patients was based on the previous treatment of patients in study 001 within 12 months of study completion of part 001B. The study duration was planned to be 54 months.

Patients were to initiate dosing with SC lumasiran at the same dose and regimen as they received in Study 001B (1 mg/kg once monthly, 3 mg/kg once monthly, or 3 mg/kg once every 3 months). All patients were later switched to either 3.0 mg/kg QM, or 3.0 mg/kg q3M (reporting of the results therefore refer to two treatment groups only). The evaluation of the PD parameters was entirely descriptive. The endpoints used for the trial were similar to those of trial 001B. Baseline for the study was defined as the derived baseline value in Study 001B.

All 20 patients from study 001B were included into the trial. The currently documented drug exposure for this study amounts to a median/mean of 10.4/11.2 months, respectively. **Figure 12** shows the results for the percent change of 24-hour urinary oxalate:creatinine ratio.

Figure 12. Mean percent change (±SEM) from baseline in 24-hour urinary oxalate:creatinine ratio by visit (Pharmacodynamic Analysis Set, Study 002)



At baseline in Study 001B, the mean plasma oxalate concentration for all patients was 15.3 µmol/L. Consistent with the observations in 24-hour urinary oxalate, patients showed stable reductions in plasma oxalate levels during treatment in Study 002, with mean values that approached the lower detection limit of 5.55 µmol/L. The mean maximum percent reduction for all patients was 53.1% (range: 12.5 to 76.4%).

Reductions were also observed in the spot urine oxalate:creatinine ratios during treatment in Study 002 that were consistent in magnitude and durability with the reductions observed in 24-hour urinary oxalate:creatinine ratios. The highest mean percent reduction from baseline was observed at the Month 3 visit (74.8%) and levels were stable thereafter.

Mean plasma glycolate levels were higher at the start of Study 002, due to persistence of the treatment effect from Study 001B. Levels increased after resuming treatment and then stabilised, at

approximately 50% above baseline in Study 001B. Glycolate plasma levels were also monitored in other studies, but were partly considered to be markers of safety (see safety assessment).

Plasma glycolate levels observed in patients enrolled in the lumasiran clinical trials are presented in **Table 19**.

Table 19. Baseline and post-dose plasma glycolate levels from patients enrolled in studies 002, 003, 004, and 005. Data are presented as median (IQR) observed levels (µmol/L) and median (IQR) percent change from baseline.

Study	Renal Status	Baseline Observed Levels	Month 3 Observed Levels	Month 6 Observed Levels	Month 12 Observed Levels
			Percent Change from BL	Percent Change from BL	Percent Change from BL
ALN-GO1-002	eGFR>45 ml/min/1.73m^2	N=17 146.0 (88.6-202.0)	Not Assessed	N=20 189.5 (134.0, 214.0)	N=19 202.0 (119.0-254.0)
				N=17 +26.21% (5.74%-63.73%)	N=17 +30.23% (9.93%-65.57%)
ALN-GO1-003	eGFR≥30 ml/min/1.73m^2	N=39 109.0 (72.6-148.0)	N=36 219.5 (155.5-298.0)	N=37 222.0 (160.0-297.0)	N=19 166.0 (147.0-230.0)
			N=36 +76.14% (39.33%-203.33%)	N=37 +77.86% (45.03%-177.53%)	N=19 +50.15% (32.20%-82.61%)
ALN-GO1-004	If ≥12 months of age: eGFR>45 ml/min/1.73m^2 If <12 months of age: normal serum creatinine	N=18 186.3 (102.5-289.0)	N=18 280.0 (216.0-383.0)	N=18 316.5 (159.0-396.0)	N=6 246.0 (210-215.0)
			N=18 +38.84% (18.05%-100.82%)	N=18 +28.36% (6.57%-145.99%)	N=6 +27.92% (9.26%-323.73%)
ALN-GO1-005	eGFR≤45 ml/min/1.73m^2 [All enrolled patients on dialysis]	N=4 465.0 (320.0-609.25)	N=3 744.0 (622.0-1150.0)	Not Available	Not Available
			N=3 +75.57% (32.03%-127.42%)		

Abbreviations: BL=Baseline; eGFR=estimated glomerular filtration rate; IQR=interquartile range

Mean 24-hour urinary glycolate:creatinine ratios were increased compared to baseline in Study 001B and stable during treatment in Study 002. At the Month 3 visit, a single patient (003-001) treated with 3.0 mg/kg once monthly had a markedly elevated 24-hour urinary glycolate:creatinine ratio of 4.410 mmol/mmol, approximately 10 times the patient's baseline level. The patient's 24-hour urinary glycolate:creatinine ratios at subsequent visits at Months 6 and 9 were consistent with the means for all patients at these timepoints.

Spot urine glycolate:creatinine ratios were increased compared to baseline in Study 001B, consistent with the increases observed in 24-hour urinary glycolate:creatinine ratios, and were then stable during treatment in Study 002. For patient 003-001, who had a marked elevation in 24-hour urinary glycolate:creatinine ratio at the Month 3 visit, the patient's spot urine glycolate:creatinine ratio at Month 3 (0.820 mmol/mmol) was consistent with the mean level for all patients at this timepoint (0.715 mmol/mmol).

No dedicated studies on secondary pharmacology were submitted.

Anti-drug antibodies

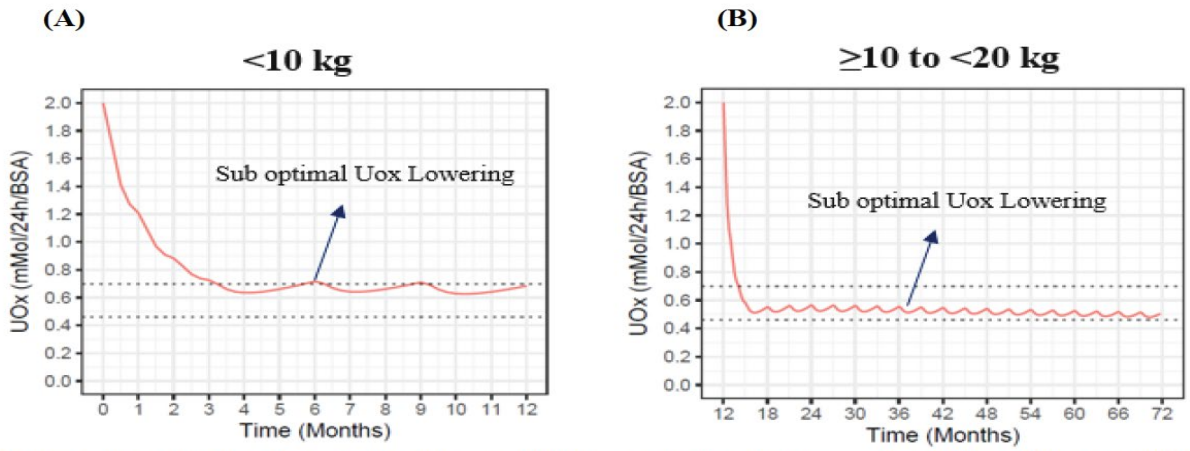
Immunogenicity assessment was conducted using data across lumasiran Studies 001A, 001B, 002, 003, and 004. The incidence of treatment-emergent ADA in lumasiran-treated healthy subjects and patients was 6% with low ADA titers (1:50). The presence of ADA was transient and did not influence the PK and PD of lumasiran.

Dose selection for PH1 patients < 6 years of age

In order to select an optimal lumasiran regimen for children less than 6 years old, the PKPD model was updated to extend the predictive capability of the model to include infants (at birth) up to adulthood. In the integrated PKPD model, the effect of body weight on lumasiran PKPD was estimated by simultaneously modeling PKPD data from monkeys (median weight 3.5 kg), healthy human adults (median weight 75 kg), and PH1 patients 6 years and above (median body weight 35 kg) using allometry.

In the 0 to 1 years, 1 to 6 years, and 6 years old age category, the mean liver weights as a percentage of body weight were 3.5%, 2.9%, and 2.0%, respectively. Hence, liver concentrations of lumasiran are expected to be lower in children. In addition, elimination rate of lumasiran is expected to be faster in children compared to adults, based on the principle of allometry. Renal function matures to 50 and 90% of adult levels by 3 and 12 months of age, respectively. Simulations on different dosing regimen were conducted (**Figure 13**).

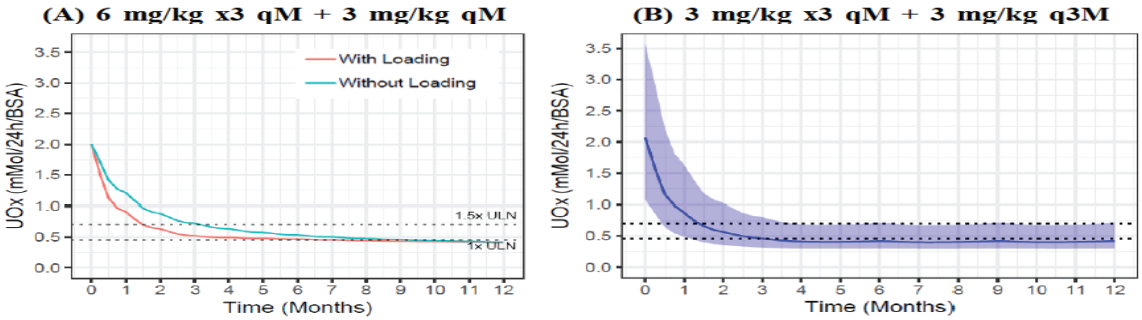
Figure 13. Suboptimal UOx suppression in <20 kg PH1 patients on dose regimen selected for ≥20 kg PH1 patients (3 mg/kg qMx3 Loading and 3 mg/kg q3M Maintenance)



Note: x-axis is indicative of age, treatment begins at birth in panel (A), and at 1 year of age (12 months) in panel (B)

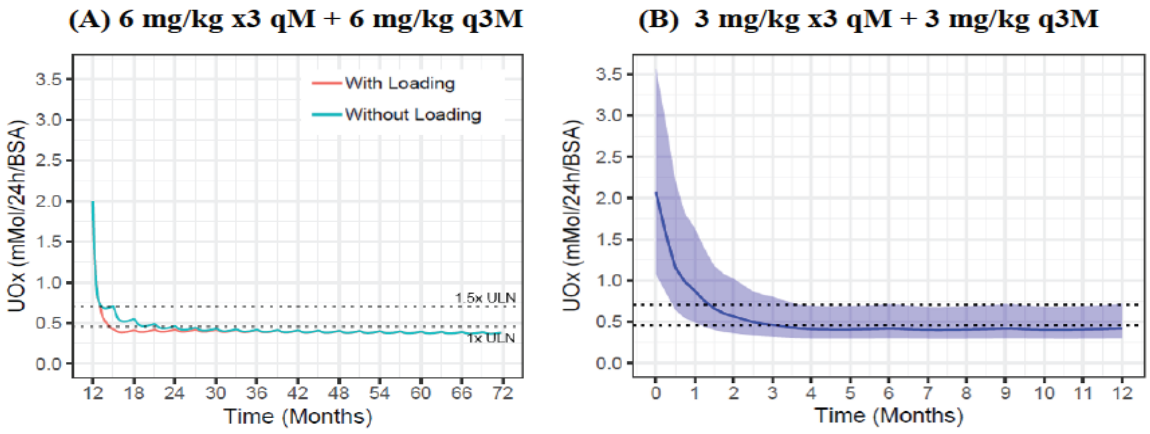
For children below 6 years, limited clinical data are available. A PKPD model for spot urinary oxalate values was developed. Based on this model, simulations were conducted to demonstrate the impact of the proposed lumasiran dosing regimens in different weight groups (**Figures 14, 15**).

Figure 14. Proposed dose regimen for children with <10 kg body weight (A) yields similar oxalate reduction as patients weighing > 20 kg (B)



Solid line=Median; Shaded area=90% prediction interval
Note: In panel (A), x-axis is indicative of age, treatment begins at birth

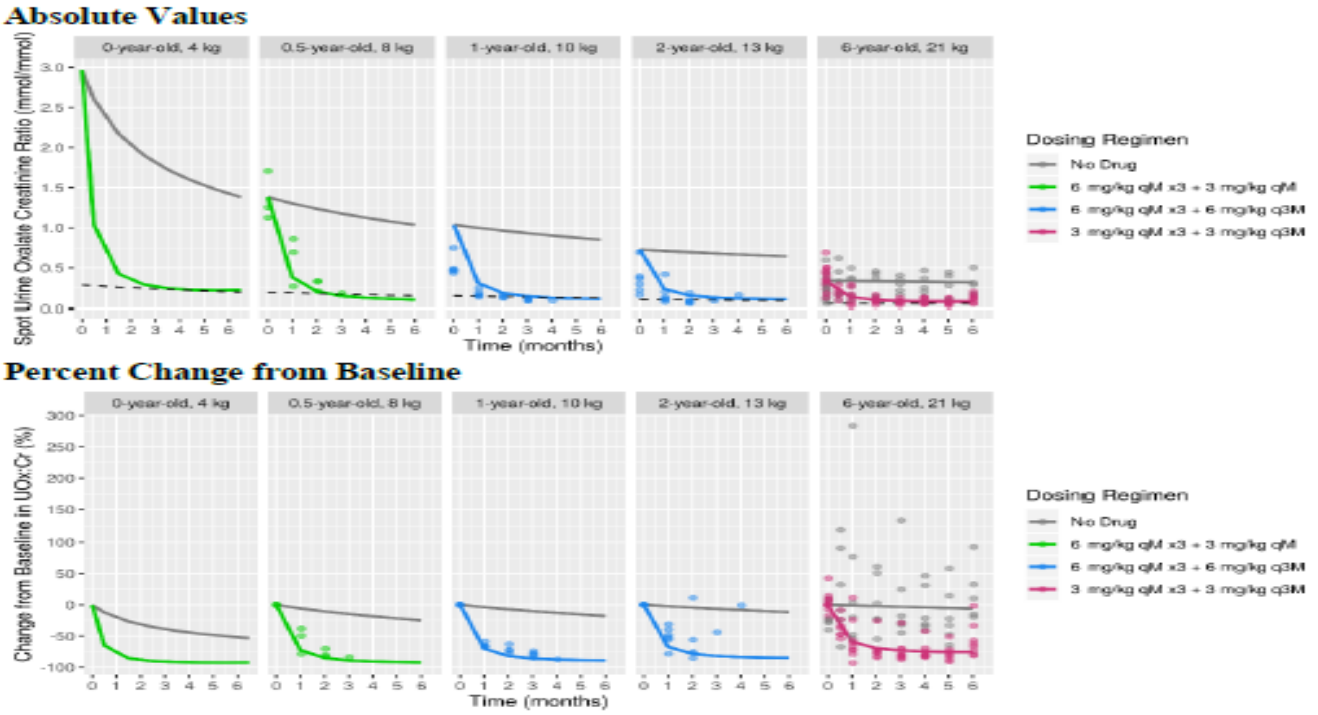
Figure 15. Proposed dose regimen for children weighing 10 to 20 kg (A) yields similar oxalate reduction as the Phase 3 dose in ≥6 years (B)



Solid line=Median; Shaded area=90% prediction interval
Note: In panel (A), x-axis is indicative of age, treatment begins at 1 year (12 months) of age

In the absence of drug, spot urinary oxalate:creatinine ratio is highest at birth and decreases with increasing age (shown as grey line in **Figure 16**).

Figure 16. Model-predicted and observed spot urinary oxalate:creatinine ratio in PH1 patient from birth to 6 years of age.



Abbreviations: CDC=Centers for Disease Control and Prevention; eGFR=estimated glomerular filtration rate; PH1=primary hyperoxaluria type 1; q3M=once every 3 months (quarterly); qM=once monthly; UOx:Cr=spot urinary oxalate:creatinine ratio
Dashed black line is the 95th percentile of UOx:Cr in healthy subjects with the respective age range according to Matos et al [Matos 1999] The difference between the solid grey line and the dashed black line represents the UOx:Cr produced by the liver in subjects with PH1. For the purpose of the estimation of UOx:Cr, subject demographics were the 50th of the CDC Growth Chart, and eGFR was the median of the baseline eGFR in the modeling dataset of 86 ml/min/1.73 m^2.

2.4.4. Discussion on clinical pharmacology

After SC administration, lumasiran is absorbed from the injection site into plasma with median Tmax ranging between 1.5 to 9 hours and is detectable in plasma up to 48 hours post dose. Lumasiran is rapidly cleared from the plasma with a half-life of 3 to 8 hours. While Cmax and $AUC_{0\text{-}inf}$ showed a linear increase, AUC_{last} and $AUC_{0\text{-}24}$ increased slightly more than proportional over the dose range 0.3 mg/kg to 6.0 mg/kg. No accumulation of lumasiran in plasma was observed after repeated once monthly or once every 3 months (quarterly) dosing.

Plasma protein binding was moderate to high (77 to 85%) at clinically relevant concentrations in plasma from adult healthy donors. The population estimate for the apparent central volume of distribution (Vd/F) of lumasiran was 4.9 L (0.07 L/kg) for a typical 70 kg adult. While the sense strand is metabolised by sequential removal of GalNAc moieties and removal of the 3' nucleotide, the antisense (AS) strand is primarily metabolised to form AS(N-1)3' lumasiran. This pharmacologically active metabolite was <10% of lumasiran in both plasma and urine in healthy subjects on lumasiran. A further deaminated metabolite pharmacologically equipotent to lumasiran may form in human liver, as observed in monkey liver, but was not detected in human plasma or urine. Lumasiran was not a substrate of CYP enzymes *in vitro*.

Renal clearance is a minor route of elimination of unchanged lumasiran from human plasma. The population PK model estimated that CLR accounted for <20% of the total clearance for lumasiran in patients with normal renal function (eGFR of 90 mL/min/1.73 m2). In healthy subjects, a mean of 17.4 to 25.8% of the administered dose was excreted in the urine as unchanged lumasiran over 24 hours compared to 7.17 to 13.7% in patients with PH1. This difference was explained with the lower eGFR values in PH1 patients, and was not judged to influence the PK of lumasiran, with the majority of lumasiran cleared from plasma by hepatic uptake.

The effect of intrinsic factors on PK of lumasiran was evaluated using pooled data from a Non-compartmental analysis (NCA) and in the population PK analysis. In patients with renal impairment, median $AUC_{0\text{-}last}$ was 25% higher in patients with moderate renal impairment in NCA, however, due the very limited sample size, robust conclusion could not be reached. In the population PK analysis, simulations indicated that renal impairment has minimal impact (≤20%) on the predicted AUC and Cmax of lumasiran without the need for dose adjustment in renally impaired patients. Interim PK data in patients with end-stage renal disease (ESRD) requiring dialysis show 3- to 7-fold higher Cmax and 2- to 3.5-fold AUC0-last increase relative to non-dialysis patients within the same body weight category in a preliminary comparison. Since the decline in plasma levels was similar as in patients without ESRD, it is expected that the increased plasma levels would not result in relevant extrahepatic distribution of the compound based on the molecular properties.

Hepatic function classification of patients included in the development programme was based on the NCI-ODWG criteria which are not considered suitable to diagnose the presence and define the level of hepatic impairment. Thus, data in patients with hepatic impairment is considered missing. Limited pharmacokinetic data in patients with mild and transient elevations in total bilirubin (total bilirubin >1.0 to 1.5×ULN) showed comparable plasma exposure of lumasiran and similar pharmacodynamics as patients with normal hepatic function. Published literature reports have shown lower expression of the ASGPR in the liver, i.e. the receptors responsible for lumasiran uptake, in patients with hepatic impairment. Non-clinical data suggest that this may not influence liver uptake or pharmacodynamics at therapeutic doses. The clinical relevance of these data is, however, unknown and in patients with moderate or severe hepatic impairment there is a potential for decreased efficacy. Therefore, caution is warranted when treating these patients and efficacy should be carefully monitored as there is a potential for decreased efficacy.

Increased systemic exposure to lumasiran was detected also in the youngest patients below the age of 1 year. Further, limited data are available in this age group. These patients should, therefore, be treated with caution.

No elderly patients were included in the development programme. This was considered acceptable as although patients can be diagnosed with PH1 at any age, most individuals experience their first symptoms in early childhood.

Available *in vitro* data, show that there is a low risk for CYP- related clinical PK interactions as substrate or inhibitor of CYPs at clinically relevant concentrations. Presently available *in vitro* data do not indicate a need for clinical DDI studies.

Based on median AUC, lumasiran exposure appeared to be slightly increased with concomitant use of Vitamin B6, however substantial overlap in exposures was observed. The incidence of treatment-emergent anti-drug antibodies (ADA) due to lumasiran was 6%. Presence of ADA did not appear to impact on the PK of lumasiran.

PK /PD modelling data indicate that plasma PK is not a predictor of PD activity. Liver PK, on the other hand, appears to drive the pharmacological effect.

The parameters chosen for the evaluation of the pharmacodynamic activity, namely urinary oxalate, plasma oxalate, urinary glycolate, and plasma glycolate were considered adequate as a means for assessing and documenting the PD activity of the compound. Of these, the glycolate related parameters are the only PD parameter that can also be used in healthy volunteers. The urinary oxalate related parameters were evaluated based on 24-hour urine collections (and normalised for body surface area), and with spot urine evaluations which were expressed as and related to the creatinine concentration in urine, in order to account for the dilution status of the urine measured. The chosen parameters are considered fully adequate to evaluate the PD activity of the compound.

Submitted results show a clear response following lumasiran administration in the reduction of urinary oxalate excretion and plasma oxalate and the increase in glycolate. The monthly doses appear to induce a more rapid onset of effect, and finally greater absolute and relative difference to baseline and to placebo (for which no relevant changes were observed), as compared to the dose administered q3M. The 3 mg/kg monthly dose demonstrated overall somewhat larger effects than the 1 mg/kg dose. These comparisons however cannot be considered conclusive due to the low number of patients included in the placebo and the three-monthly dose groups.

The long-term analysis revealed a clear maintenance of the effects seen during the initial dosing period, consistently favouring the 3 mg/kg monthly dose over the two other doses at least for the 24-hour urine-based oxalate excretion data. However, it could also be demonstrated that the 3 monthly dosing was also able to maintain the effects observed initially, which was considered as a demonstration that the dosing interval could be appropriate for long-term treatment.

Available data support the PD activity of the compound in the target population of patients from the age of 6 years.

Patients from study 001 could enter a long-term open-label extension study (Study 002) after termination of study 001. No new patients were recruited for this study. This study is still ongoing at the time of submission and reported with an interim report. For this trial, the endpoints reported also include the similar PD endpoints reported in the trial 001, and include a comparison to baseline, which was defined as the baseline before inclusion into study 001. The planned study duration is 54 months, and this trial therefore will also provide data on the long-term safety and efficacy of lumasiran.

In this trial, patients were initially receiving their last dose in trial 001, but patients receiving 1.0 mg kg monthly were later switched to the two 3.0 mg/kg doses (at 3 and 6 months). All patients were

observed for at least 6 months in the submitted interim report and the mean/median exposure was 11.1/10.4 months in total. The pharmacodynamic effects observed in this study were fully consistent with the results achieved in study 002, and a full maintenance of the effects with only minimal fluctuation could be demonstrated.

Body weight-based dosing is recommended for lumasiran based on available clinical data and PK and PK/PD modelling. The latter has some deficiencies, as the correlation between plasma PK and liver PK was not evaluated and modelling assumptions were based on allometric scaling between species. Furthermore, the impact of immature organ function (liver and kidney) in the paediatric population were not fully considered. Nevertheless, the overall model was in good agreement with the spot urinary oxalate:creatinine ratios observed for this population.

With regards to intrinsic factors such as age and body weight, in children, weight-normalised clearance was 1.8 to 2-fold higher in infants below 10 kg and 1.5-fold higher in children 10 to <20 kg compared to adults. Even though weight-normalised clearance was increased 1.5-fold in children between 10 and 20 kg, a 2-fold higher dose is proposed for this body weight category. Tmax occurred earlier in younger children (with lower body weight) and elimination phase was faster. Overall, this resulted in a median observed exposure (AUC) that was higher in the lower body weight categories for the proposed loading doses. So, overall, with the proposed doses, exposure in children below 6 years (<20 kg) is expected to be slightly higher compared to adults. The applicant stated that slightly higher plasma concentrations in paediatric patients were needed in order to account for higher liver size relative to body weight in children, and rapid growth in body weight and liver size in addition to allometric scaling to account for faster clearance in paediatric liver. Lumasiran, at the proposed dosing regimen, is estimated to yield a spot urinary oxalate:creatinine ratio that approaches ULN by Month 3. Oxalate levels at Month 6 are expected to be similar to oxalate levels at Month 3, which would indicate that maximum lowering is achieved by Month 3 using a loading dose regimen and is subsequently maintained with the maintenance regimen.

As there are only limited data, the potential effects of increased lumasiran exposure in the younger population will be further characterised in the ongoing study ALN-GO1-004 and the planned observational study ALN-GO1-007.

The applicant did not submit secondary pharmacology studies. The lack of such studies was considered acceptable, as based on its PK and PD properties, no PD interaction with lumasiran is expected.

Further extended long-term storage stability data still needs to be reported for plasma and urine samples that have not been analysed from study ALN-GO1-002. It is recommended that the applicant submits there results once stability sample testing is complete.

Overall, it can be concluded that the documentation of the pharmacodynamics has adequately documented the activity of the compound adapted to the small numbers/rare disease prevalence, and that the trials conducted for the PK-PD programme formed an adequate basis for the further investigation of the compound.

2.4.5. Conclusions on clinical pharmacology

Pharmacokinetics of lumasiran in the target organ (liver) drives the long duration of PD effect. The majority of lumasiran is cleared from plasma by ASGPR-mediated hepatic uptake, with renal clearance contributing to a lesser extent.

As lumasiran has not been investigated in patients with hepatic impairment, caution is required when treating patients with moderate or severe hepatic impairment Tand efficacy should be monitored in these patients.

Increases in systemic exposures are expected in renal impairment and caution should be used when treating these patients. Weight-based subcutaneous dosing of lumasiran is being proposed based on observed data but also PK and PK/PD modelling.

Data in children under the age of 6 years of age are still relatively limited, however, no safety concerns specific for the young paediatric population have been raised. Additional information in this population will be collected through the ongoing studies as described in the RMP.

2.5. Clinical efficacy

2.5.1. Dose response studies

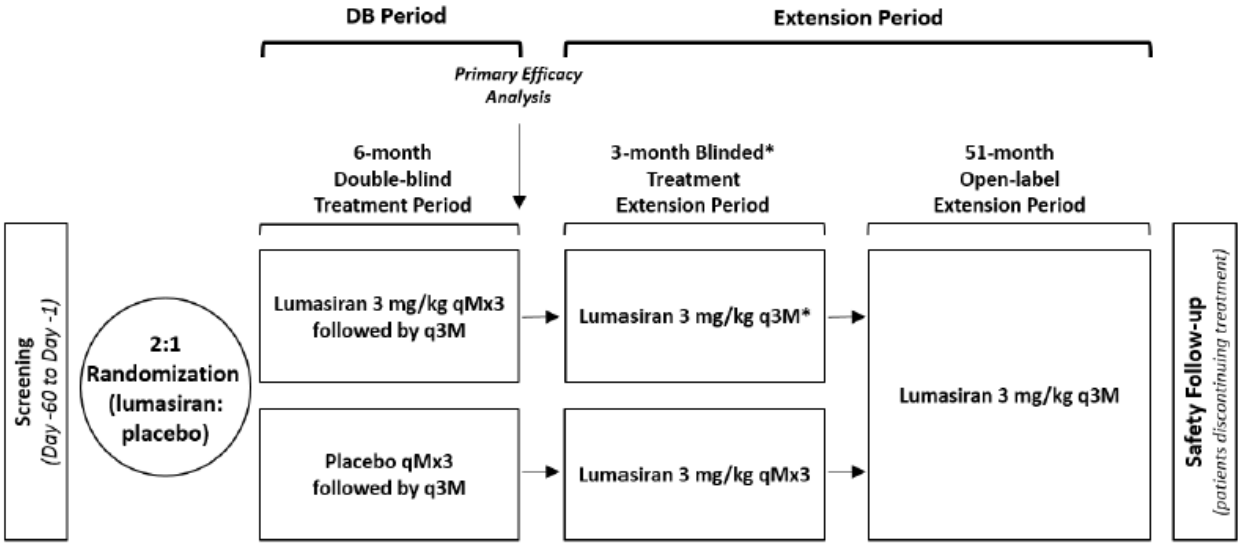
Dose-finding was investigated in studies ALN-G01-001 and ALN-G01-002 with additional supportive information deriving form PK and PK-PD modelling and simulation (See Sections 2.4.2 and 2.4.3 of this report).

2.5.2. Main study

ALN-G01-003 (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.

Methods

Figure 17 provides a schematic of the overall study design.



Abbreviations: DB=double-blind; OLE=open-label extension; q3M=once every 3 months; qM=once monthly; qMx3=once monthly for 3 consecutive months (ie, Day 1, Month 1, Month 2).

Study Participants

Inclusion criteria:

- Age 6 years or older.
- Documentation or confirmation of PH1 as determined by genetic analysis prior to randomisation.
- Mean 24-hour urinary oxalate excretion from the first 2 valid 24-hour urine collections is ≥0.70 mmol/24hr/1.73m2.
- If taking pyridoxine (vitamin B6) for the treatment of PH1, was required to have been on a stable regimen for at least 90 days before randomization, and willing to remain on this stable regimen for 12 months from first study drug administration.
- Patient was able to understand and was willing and able to comply with the study requirements and to provide written informed consent. In the case of patients under the age of legal consent, the legal guardian(s) was required to provide informed consent and the patient should have provided assent per local and national requirements.

Exclusion criteria (selection):

- Medical history included clinical evidence of extrarenal systemic oxalosis, as determined

  by the Investigator.
- Had any of the following laboratory parameter assessments at screening:
  - Alanine aminotransferase (ALT) or aspartate aminotransferase (AST) >2× upper limit of normal (ULN).
  - Total bilirubin >1.5×ULN. Patients with elevated total bilirubin that was secondary to documented Gilbert's syndrome were eligible if the total bilirubin was <2×ULN.
  - International normalised ratio (INR) >1.5 (patients on oral anticoagulant [eg, warfarin] with an INR <3.5 were allowed).
- Had known active human immunodeficiency virus infection; or evidence of current or chronic hepatitis C virus or hepatitis B virus infection.
- Estimated GFR of <30 mL/min/1.73m2 at screening (calculation was based on the Modification of Diet in Renal Disease [MDRD] formula for patients ≥18 years of age and the Schwartz Bedside Formula for patients <18 years of age).
- History of renal or liver transplant.
- Was not willing to comply with the contraceptive requirements during the study period, as described in the protocol.
- Female patient was pregnant, planning a pregnancy, or breast-feeding.

Treatments

Lumasiran was administered as a solution for injection that contains 189 mg/mL lumasiran (equivalent to 200 mg/mL of lumasiran sodium). Placebo was supplied as a sterile, preservative free normal saline 0.9% solution. Study drug was administered SC based on total body weight.

Body weight collected within 3 months prior to the study drug dose or the pre-dose weight collected on the study visit day or dosing day was used for dose calculations.

During the DB Period (Day 1 through Month 6 assessments), patients received loading doses of 3.0 mg/kg lumasiran or an equivalent volume of placebo once monthly for 3 doses (at the Day 1, Month 1, and Month 2 visits), followed by 1 maintenance dose of 3.0 mg/kg lumasiran or placebo administered at the Month 3 visit. All doses were SC injections.

Starting at Month 6 (after the Month 6 assessments), all patients were to receive active drug in the Extension Period; however, to preserve the blind for the prior DB Period, treatments were:

- In patients randomised to placebo: loading doses of lumasiran 3.0 mg/kg once monthly for 3 doses (at the Month 6, 7, and 8 visits).
- In patients randomised to lumasiran: maintenance dose at the Month 6 visit as well as placebo doses at the Month 7 and 8 visits.

At Month 9 (after the Month 9 assessments), all patients received their first open-label maintenance dose of lumasiran.

Objectives

The Primary Objective of the trial was to evaluate the effect of lumasiran on percent reduction in urinary oxalate excretion.

The secondary objectives were the characterisation of the effect of lumasiran on absolute levels of urinary oxalate excretion, the oxalate:creatinine ratios, and plasma oxalate, the effect of lumasiran on renal function, and to evaluate the long-term treatment effect of lumasiran.

Exploratory objectives included the evaluation of quality of life, the changes (in the occurrence of) nephrocalcinosis and renal stones, and several PD and PK parameters.

Outcomes/endpoints

Primary

- Percent change in 24-hour urinary oxalate excretion from baseline to Month 6 corrected for BSA

Secondary

- Absolute change in 24-hour urinary oxalate corrected for BSA from baseline to Month 6.
- Percent change in 24-hour urinary oxalate:creatinine ratio from baseline to Month 6.
- Percent change in plasma oxalate from baseline to Month 6.
- Proportion of patients with 24-hour urinary oxalate level at or below 1.5×ULN at Month 6.
- Proportion of patients with 24-hour urinary oxalate level at or below ULN at Month 6.

- Absolute change in plasma oxalate from baseline to Month 6.
- Change in estimated glomerular filtration rate (eGFR) from baseline to Month 6.
- Extension Period endpoint, which includes several individual endpoints: Change from baseline (percent and absolute) in: 24-hour urinary oxalate excretion, 24-hour urinary oxalate:creatinine ratios, and eGFR; and percentage of time that 24-hour urinary oxalate is ≤ 1.5×ULN; all available Extension Period data are presented as of the cutoff date for this interim CSR.

The exploratory endpoints comprised the following: Change in KDQOL for patients ≥18 years of age at screening, and the PedsQL (generic and ESRD modules) for patients <18 years of age at screening, the change in EQ-5D and EQ-5D VAS; the change in rate of renal stone events, the change in nephrocalcinosis as assessed by renal ultrasound, the change in urinary and plasma glycolate, the change in urinary oxalate:creatinine ratios as assessed in random spot urine collections, the PK profile of lumasiran, the frequency of ADA, the change in patient resource use (eg, work/school attendance, visits to doctor/hospital), the change in patient experiences as evaluated by patient and caregiver experience surveys, and the frequency and seriousness of AEs.

Sample size

Patients were randomised 2:1 to receive lumasiran or placebo, respectively. Assuming a mean percent reduction from baseline to Month 6 in 24-hour urinary oxalate corrected for BSA of 17% in the placebo arm and a standard deviation (SD) in both arms of 25%, 24 patients were calculated to provide 90% power to detect a treatment difference of 37% at a 2-sided 5% significance level (ie, 54% reduction in the lumasiran arm). To account for potential drop-outs, 30 patients were planned to be enrolled in the study.

Randomisation

Using an Interactive Response System (IRS), patients were randomised 2:1 to lumasiran or placebo. Randomization was stratified by mean urinary oxalate level (>1.70 versus ≤1.70 mmol/24hr/1.73m2) calculated using the values obtained from the first 2 valid baseline 24-hour urine collections.

Each patient was uniquely identified in the study by a combination of the site number and patient identification number. Upon signing the ICF, the patient was assigned a patient identification number by the IRS. The Investigator or his/her designee contacted the IRS to randomise the patient after confirming that the patient fulfilled all the inclusion criteria and none of the exclusion criteria.

Blinding (masking)

Because the drug product was different in colour and appearance to placebo (lumasiran: yellow, placebo: colourless), the site pharmacists were involved in ensuring the double-blind administration: All site pharmacists who prepared study drug during the blinded period of the study were to ensure that each syringe was covered in provided yellow transparent film in order to mask the identity of study drug. All other personnel of both sponsor and investigational sites remained blinded until final collection of the month 6 data.

Statistical methods

The primary analysis was performed using a restricted maximum likelihood (REML) based Mixed-Effect Model Repeated Measures (MMRM) approach. The outcome variable was percent change from baseline in 24-hour urinary oxalate corrected for BSA (mmol/24hr/1.73m2) at Months 3, 4, 5, and 6. Analysis included fixed effects of treatment arm (lumasiran versus placebo) and scheduled visits (Months 3, 4, 5, and 6), as well as continuous, fixed covariate of baseline 24-hour urinary oxalate corrected for BSA (mmol/24hr/1.73m2) level and patient as a random factor..

The primary comparison was the least squares (LS) mean treatment difference (lumasiran-placebo) in percent change from baseline of 24-hour urinary oxalate excretion from Months 3 to 6. This LS mean difference was presented along with corresponding standard errors of the mean (SEMs), 95% confidence intervals (CIs), and p-value from the model.

Two sensitivity analyses were conducted to evaluate the sensitivity to the estimated treatment effect by the assumption that the treatment effect reaches steady state at Month 3 and is maintained 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 post-baseline data (including percent change from baseline at Months 1 and 2).

A gatekeeping testing strategy was implemented to control the overall type I error rate. The primary endpoint was compared between treatment arms at a 2-sided significance level of 0.05. If statistically significant, then the secondary endpoints listed below were tested at the same level of 0.05 in the following hierarchical order:

- Absolute change in 24-hour urinary oxalate corrected for BSA from baseline to Month 6
- Change in 24-hour urinary oxalate:creatinine ratio (value/ULN) from baseline to Month 6
- Proportion of patients with 24-hour urinary oxalate level below 1.5xULN [ULN=0.46 mmol/24h/1.73m2] at Month 6
- Proportion of patients with 24-hour urinary oxalate level below ULN [ULN=0.46 mmol/24h/1.73m2] at Month 6.

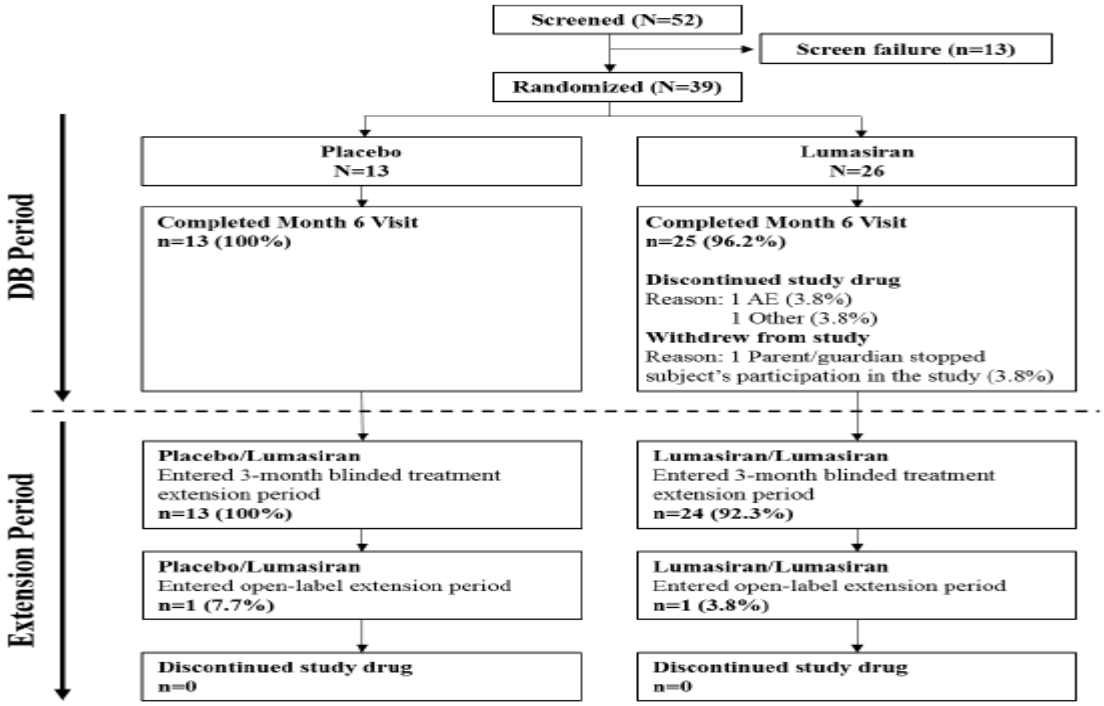
Only if the comparison was significant at a 2-sided significance level of 0.05 was the next endpoint in the hierarchy formally tested; if a given comparison was not significant, the subsequent tests were to be performed, and the results summarised, but statistical significance was not to be inferred.

Continuous endpoints will be analysed using an MMRM approach similar to the one described for the primary endpoint. For binary endpoints, odds ratio and the differences in proportions with corresponding 95% confidence will be summarised.

Results

Participant flow

Figure 18. Disposition of patients in the DB period + extension period in study ALN-G01-003.



Recruitment

Study initiation date: 13 December 2018

Interim data cut-off date: 06 November 2019 (additional interim data cut-off date: 01 May 2020)

Conduct of the study

A total of 5 major protocol deviations were reported among 4 patients:

- One patient had 2 short pauses in pyridoxine use and therefore did not maintain a stable pyridoxine regimen for the first 12 months following the first dose of study drug (one pause was during the DB Period and one pause was during the Extension Period).
- 24-hour urine sample did not meet validity criteria and procedure was not repeated as specified (1 patient; Month 3) or sample was not collected (1 patient; Month 4).
- The appropriate ICF was not signed by the patient/guardian prior to performing study procedures (1 patient). The patient signed consent during a clinic visit but had completed a 24-hour urine collection during screening beforehand.

The original protocol was finalised on 09 July 2018. Since then, there have been 2 major protocol amendments.

Protocol Amendment 1 (23 July 2018): The primary purpose for this protocol amendment was to provide additional clarification about the patient caregiver surveys specific to patients under the legal age of consent, and to make corrections to the open-label extension period Schedule of Assessments, which was missing the Month 27 visit in Year 2 (Study Day 757 ±14 days).

Protocol Amendment 2 (19 March 2019): The primary purpose of Amendment 2 was to broaden the patient population by allowing enrolment of patients with a glomerular filtration rate ≥30 mL/min/1.73 m2 and to align clinical objectives and endpoints across the Phase 3 program.

Baseline data

The baseline demographic data and disease characteristics of the included patients in study are shown in **Tables 20** and **21** respectively.

Table 20. Demographic characteristics for the DB period of study ALN-G01-003 (Safety Analysis Set)

Demographic	Placebo (N=13)	Lumasiran (N=26)	Overall (N=39)
Age at informed consent (years)			
Mean (SD)	17.0 (15.19)	18.7 (11.52)	18.1 (12.68)
Min, Max	6, 60	6, 47	6, 60
Age category (years), n (%)			
6 to <12	7 (53.8)	9 (34.6)	16 (41.0)
12 to <18	1 (7.7)	5 (19.2)	6 (15.4)
18 to <65	5 (38.5)	12 (46.2)	17 (43.6)
Sex, n (%)			
Male	8 (61.5)	18 (69.2)	26 (66.7)
Female	5 (38.5)	8 (30.8)	13 (33.3)
Race, n (%)			
Asian	3 (23.1)	3 (11.5)	6 (15.4)
Japanese ancestry	0	0	0
White	9 (69.2)	21 (80.8)	30 (76.9)
Other	0	2 (7.7)	2 (5.1)
More than one race	1 (7.7)	0	1 (2.6)
Ethnicity, n (%)			
Hispanic or Latino	0	1 (3.8)	1 (2.6)
Not Hispanic or Latino	13 (100.0)	25 (96.2)	38 (97.4)
Region, n (%)			
North America	2 (15.4)	11 (42.3)	13 (33.3)
Europe	8 (61.5)	10 (38.5)	18 (46.2)
Middle East	3 (23.1)	5 (19.2)	8 (20.5)

Table 21. Baseline disease characteristics for the DB period of study ALN-G01-003 (Safety Analysis Set).

Baseline Disease Characteristics	Placebo (N=13)	Lumasiran (N=26)	Overall (N=39)
Age at diagnosis (years)			
Mean (SD)	7.9 (15.84)	9.6 (8.99)	9.0 (11.53)
Min, Max	-1[a], 59	0, 36	-1[a], 59
24-hour urinary oxalate excretion corrected for BSA (mmol/24hr/1.73m^2)			
Mean (SD)	1.794 (0.6836)	1.836 (0.5966)	1.822 (0.6182)
Median	1.683	1.768	1.723
Min, Max	0.68, 2.84	0.76, 3.05	0.68, 3.05
≤1.70, n (%)	7 (53.8)	11 (42.3)	18 (46.2)
>1.70, n (%)	6 (46.2)	15 (57.7)	21 (53.8)
24-hour urinary oxalate:creatinine ratio (mmol/mmol)			
Mean (SD)	0.2369 (0.11029)	0.2089 (0.10117)	0.2182 (0.10370)
Median	0.2338	0.1706	0.1855
Min, Max	0.090, 0.416	0.077, 0.500	0.077, 0.500
Plasma oxalate (µmol/L)			
Mean (SD)	15.49 (7.341)	14.77 (7.628)	15.01 (7.444)
Median	13.10	13.05	13.10
Min, Max	7.8, 28.4	7.0, 43.5	7.0, 43.5
eGFR CKD stage (mL/min/1.73m^2), n (%)			
≥90	4 (30.8)	9 (34.6)	13 (33.3)
60 to <90	6 (46.2)	13 (50.0)	19 (48.7)
45 to <60	1 (7.7)	2 (7.7)	3 (7.7)
30 to <45	2 (15.4)	2 (7.7)	4 (10.3)
Patient-reported history of the following, n (%)			
Symptomatic renal stone events	10 (76.9)	23 (88.5)	33 (84.6)
Lithotripsy/stone removal procedures in the past 12 months prior to consent	3 (23.1)	4 (15.4)	7 (17.9)
Pyridoxine use at baseline	9 (69.2)	13 (50.0)	22 (56.4)
Pyelonephritis	5 (38.5)	5 (19.2)	10 (25.6)
Urinary tract infections	5 (38.5)	11 (42.3)	16 (41.0)
Nephrocalcinosis	9 (69.2)	12 (46.2)	21 (53.8)
Number of renal stone events in the 12 months prior to consent			
0	9 (69.2)	15 (57.7)	24 (61.5)
1 to 5	4 (30.8)	8 (30.8)	12 (30.8)
6 to 10	0	2 (7.7)	2 (5.1)
>10	0	1 (3.8)	1 (2.6)
Presenting symptoms[b], n (%)			
Asymptomatic (familial screening)	3 (23.1)	2 (7.7)	5 (12.8)
Renal stone	7 (53.8)	21 (80.8)	28 (71.8)
Nephrocalcinosis	7 (53.8)	10 (38.5)	17 (43.6)
Other	3 (23.1)	4 (15.4)	7 (17.9)

Numbers analysed

The analysis sets were defined as follows

Full Analysis Set (FAS): All randomised patients who received any amount of study drug. Patients were analysed according to the treatment to which they were randomised.

Safety Analysis Set: All patients who received any amount of study drug. Patients were analysed according to the treatment actually received.

PK Analysis Set: All patients who received any amount of study drug and have at least 1 PK concentration measurement.

All Lumasiran Treated Set: All patients who received any amount of lumasiran, including patients who took lumasiran during the DB Period and patients who initially took placebo during the DB Period and then switched to lumasiran during the Extension Period.

Plasma Oxalate Analysis Set: All patients who received any amount of study drug and have a baseline plasma oxalate level ≥1.5×lower limit of quantitation (LLOQ).

The FAS was the primary population used to evaluate efficacy for the primary, secondary, and exploratory endpoints during the DB Period. Safety was analysed using the Safety Analysis Set.

The PK Set was used to evaluate the PK endpoints.

The Plasma Oxalate Analysis Set was used to evaluate the plasma oxalate endpoints.

The All Lumasiran Treated Set was used to summarise the long-term efficacy and safety of lumasiran; all available long-term data are presented as of the cut-off date for this CSR.

The analysis sets are summarised in **Table 22.**

Table 22. Analysis populations in study ALN-G01-003.

Study Population	No. of Patients (%)		
	Placebo (N=13)	Lumasiran (N=26)	Overall (N=39)
FAS	13 (100.0)	26 (100.0)	39 (100.0)
Safety Analysis Set	13 (100.0)	26 (100.0)	39 (100.0)
All Lumasiran Treated Set	13 (100.0)	26 (100.0)	39 (100.0)
PK Analysis Set	13 (100.0)	26 (100.0)	39 (100.0)
Plasma Oxalate Analysis Set[a]	10 (76.9)	23 (88.5)	33 (84.6)

Outcomes and estimation

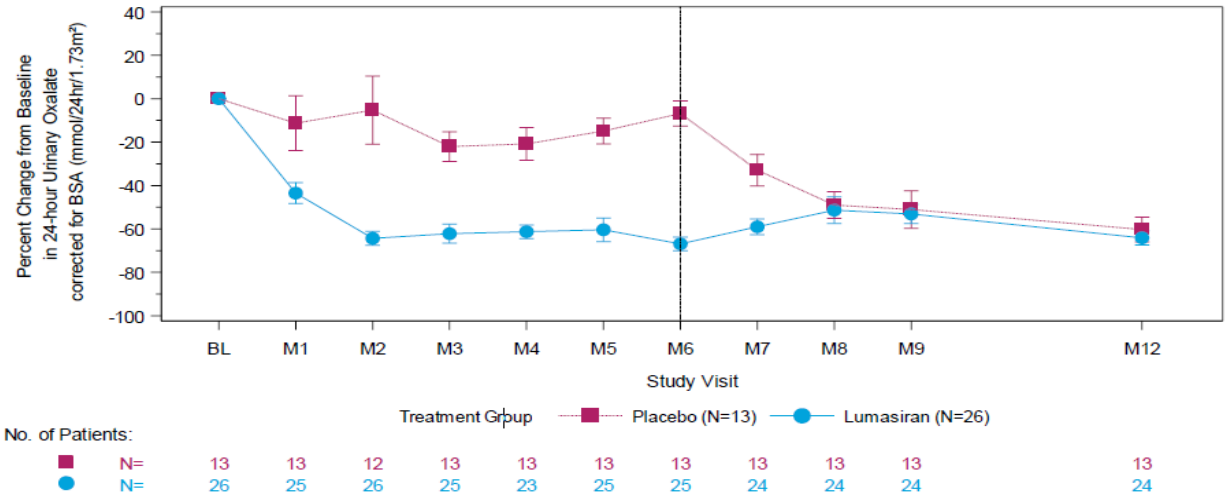
Primary endpoint

The primary endpoint of the percent change in 24-hour urinary oxalate excretion corrected for BSA from baseline to Month 6 is shown in **Table 23** and the time-course of the urinary oxalate excretion including the extension phases is shown in **Figure 19.**

Table 23. Primary endpoint: percent change from baseline in 24-hour urinary oxalate corrected for BSA (mmol/24hr/1.73m2) During the DB period of study ALN-G01-003, MMRM (FAS)

Visit	Statistic[a]	Placebo (N=13)	Lumasiran (N=26)
Baseline[b]	Mean (SEM)	1.794 (0.1896)	1.836 (0.1170)
Percent Change from Baseline to Month 6 (Average of Months 3-6)	LS Mean (SEM)	-11.838 (3.8132)	-65.385 (2.9383)
	95% CI	(-19.531, -4.146)	(-71.317, -59.452)
	Difference in LS Mean (SEM) (lumasiran - placebo)		-53.546 (4.3224)
	95% CI		(-62.314, -44.778)
	p-value		1.685E-14

Figure 19. Percent change (±SEM) from baseline in 24-hour urinary oxalate corrected for BSA (mmol/24hr/1.73m2) During study ALN-G01-003, based on observed values (FAS)



Abbreviations: BL=baseline; BSA=body surface area; DB=double-blind; FAS=full analysis set; M=month; SEM=standard error of the mean.

Continued treatment with lumasiran (lumasiran/lumasiran group) in the Extension Period led to maintenance of this reduction through Month 12, with a mean percent change (SEM) from baseline in 24-hour urinary oxalate corrected for BSA of -66.9% (3.1%) at Month 6 and -64.1% (3.3%) at Month 12. Placebo crossover patients had a mean (SEM) percent change from baseline in 24-hour urinary oxalate corrected for BSA of -6.8% (5.8%) at Month 6. At Month 12 (6 months of lumasiran treatment), the mean (SEM) percent change from baseline in 24-hour urinary oxalate corrected for BSA was -57.3% (4.9%) relative to the first dose of lumasiran in the Extension Period.

Results from the two sensitivity analyses (adding the interaction of visit and treatment to the primary model and including all post-baseline data) are shown in **Tables 24** and **25**.

Table 24. Sensitivity analysis 1: Percent change from baseline in 24-hour urinary oxalate corrected for BSA (mmol/24hr/1.73m2), including treatment and time interaction, during the DB period of study ALN-G01-003, MMRM (FAS)

Visit	Statistic[a]	Placebo (N=13)	Lumasiran (N=26)
Baseline[b]	Mean (SEM)	1.794 (0.1896)	1.836 (0.1170)
Percent Change from Baseline to Month 6 (Average of Months 3-6)	LS Mean (SEM)	-16.423 (4.2856)	-63.034 (3.0736)
	95% CI	(-25.118, -7.728)	(-69.261, -56.807)
	Difference in LS Mean (SEM) (lumasiran - placebo)		-46.611 (5.2747)
	95% CI		(-57.308, -35.915)
	p-value		1.479E-10

Table 25. Sensitivity Analysis 2: Percent change from baseline in 24-hour urinary oxalate corrected for BSA (mmol/24hr/1.73m2), including all visits and treatment and time interaction, during the DB period of study ALN-G01-003, MMRM (FAS)

Visit	Statistic[a]	Placebo (N=13)	Lumasiran (N=26)
Baseline[b]	Mean (SEM)	1.794 (0.1896)	1.836 (0.1170)
Percent Change from Baseline			
Month 1	LS Mean (SEM)	-11.567 (9.2424)	-44.130 (6.6851)
	95% CI	(-30.311, 7.178)	(-57.671, -30.589)
	Difference in LS Mean (SEM) (lumasiran - placebo)		-32.563 (11.4069)
	95% CI		(-55.687, -9.439)

Secondary endpoints

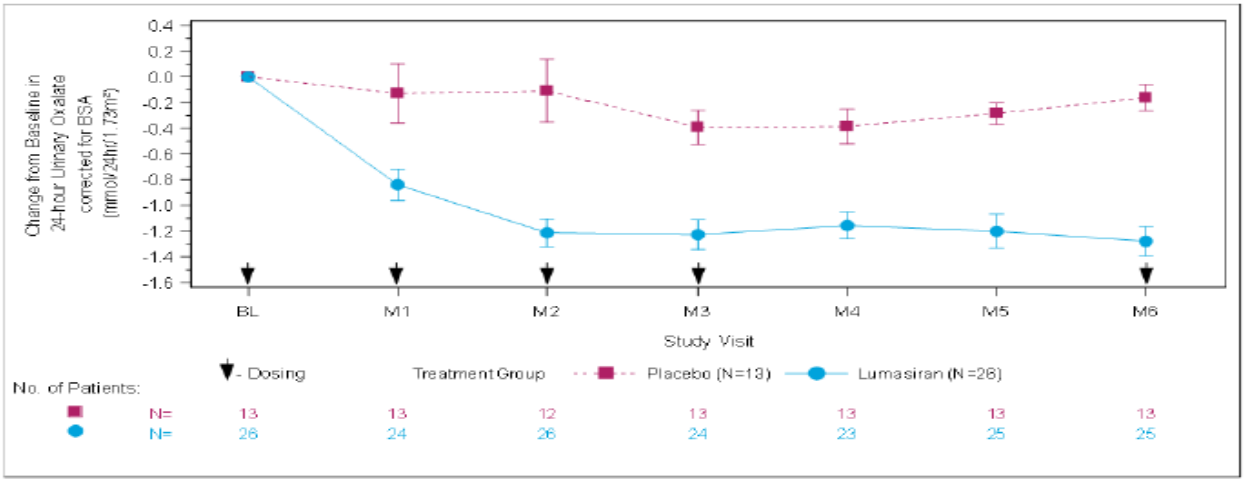
Secondary endpoints and corresponding changes over time are presented in this section.

Absolute change from baseline in 24-hour urinary oxalate vcorrected for BSA

Table 26. Absolute change from baseline in 24-hour urinary oxalate corrected for BSA (mmol/24hr/1.73m2) during the DB period of study ALN-G01-003, MMRM (FAS)

Visit	Statistic[a]	Placebo (N=13)	Lumasiran (N=26)
Baseline[b]	Mean (SEM)	1.794 (0.1896)	1.836 (0.1170)
Absolute Change from Baseline to Month 6 (Average of Months 3-6)	LS Mean (SEM)	-0.267 (0.0831)	-1.242 (0.0610)
	95% CI	(-0.436, -0.099)	(-1.365, -1.118)
	Difference in LS Mean (SEM) (lumasiran - placebo)		-0.975 (0.0998)
	95% CI		(-1.177, -0.772)
	p-value		1.225E-11

Figure 20. Absolute change from baseline in 24-hour urinary oxalate corrected for BSA (mmol/24hr/1.73m2) during the DB period of study ALN-G01-003 (FAS)

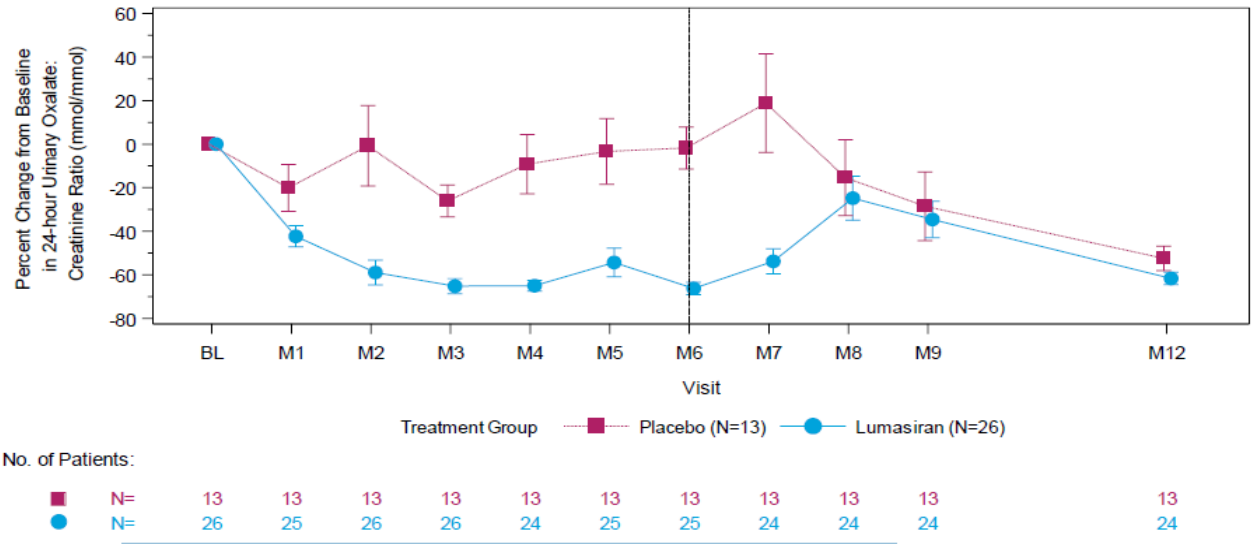


24-hour urinary oxalate:creatinine ratio (percent change)

Table 27. Percent change from baseline in 24-hour urinary oxalate:creatinine ratio (mmol/mmol) during the DB period of study ALN-G01-003, MMRM (FAS)

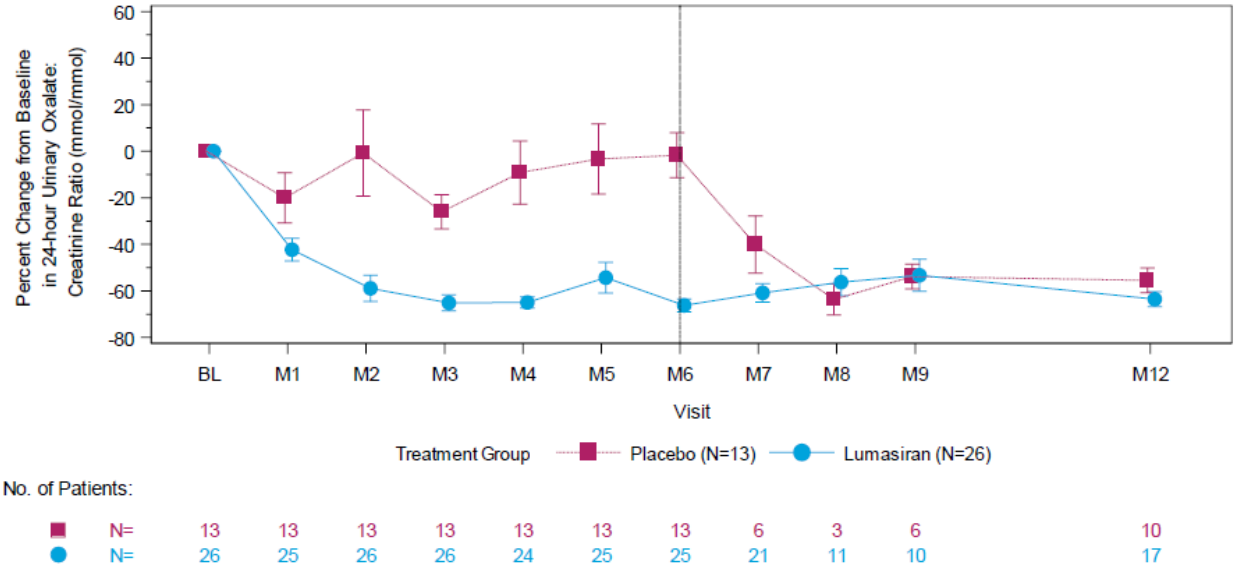
Visit	Statistic[a]	Placebo (N=13)	Lumasiran (N=26)
Baseline[b]	Mean (SEM)	0.2369 (0.03059)	0.2089 (0.01984)
Percent Change from Baseline to Month 6 (Average of Months 3-6)	LS Mean (SEM)	-10.7751 (5.35366)	-62.5469 (4.04475)
	95% CI	(-21.5778, 0.0277)	(-70.7071, -54.3868)
	Difference in LS Mean (SEM) (lumasiran - placebo)		-51.7718 (6.16118)
	95% CI		(-64.2653, -39.2784)
	p-value		5.032E-10

Figure 21. Study 003: Mean (±SEM) of percent change from baseline in 24-hour urinary oxalate:creatinine ratio at each visit (Full Analysis Set, including all values)



An investigation for the unexpected increase in the urinary oxalate:creatinine ratio results in all patients at months 7 and 8, identified inadequate mixing of 24-hour urine creatinine samples at the laboratory by one analyst affecting a total of 65 24-hour urinary creatinine samples obtained in the extension period. Results of a sensitivity analysis conducted excluding these samples are presented in **Figure 22**.

Figure 22: Study 003: Mean (±SEM) of percent change from baseline in 24-hour urinary oxalate:creatinine ratio at each visit, sensitivity analysis with valid urinary creatinine data



Percent change in POx (µmol/l) from baseline

Table 28. Percent Change from Baseline in Plasma Oxalate (µmol/L) During the DB Period of study ALN-G01-003, MMRM (Plasma Oxalate Analysis Set)

Visit	Statistic[a]	Placebo (N=10)	Lumasiran (N=23)
Baseline[b]	Mean (SEM)	17.76 (2.167)	15.73 (1.585)
Percent Change from Baseline to Month 6 (Average of Months 3-6)	LS Mean (SEM)	-0.32 (4.293)	-39.80 (2.938)
	95% CI	(-9.12, 8.48)	(-45.81, -33.80)
	Difference in LS Mean (SEM) (lumasiran - placebo)		-39.48 (5.181)
	95% CI		(-50.10, -28.87)
	p-value		2.862E-08

Continued treatment with lumasiran (lumasiran/lumasiran group) in the Extension Period led to maintenance of this reduction through Month 12, with a mean (SEM) percent change from baseline in plasma oxalate of -36.9% (4.9%) at Month 6 and -35.0% (6.1%) at Month 12.

Placebo crossover patients had a mean (SEM) percent change from baseline in plasma oxalate of 12.4% (6.9%) at Month 6. At Month 12 (6 months of lumasiran treatment), the mean (SEM) percent change from baseline in plasma oxalate was -48.9% (5.1%) relative to the first dose of lumasiran in the Extension Period.

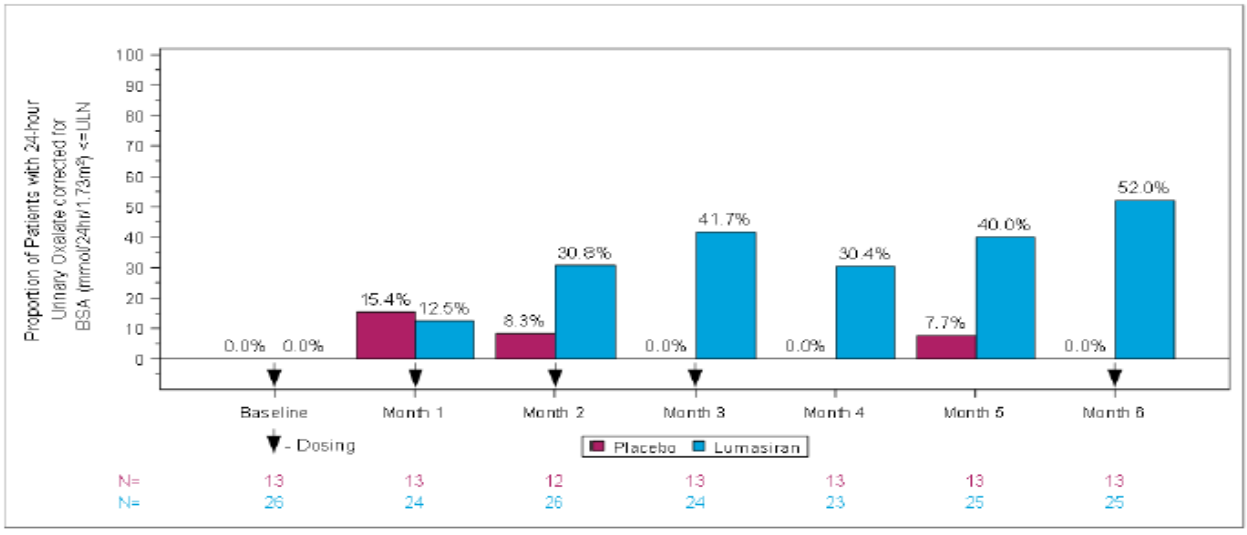
Proportion of patients with 24 UOx corrected for BSA achieving ≤1.5xULN or ≤ULN

Table 29. Proportion of patients with 24-hour urinary oxalate corrected for BSA achieving ≤1.5xULN or ≤ULN ≤, study ALN-G01-003 (FAS)

Threshold	Month 6 (DB Period; FAS)		Month 12 (Extension Period; All Lumasiran Treated Set)	
	Placebo (N=13)	Lumasiran (N=26)	Placebo/ Lumasiran (N=13)	Lumasiran/ Lumasiran (N=26)
≤1.5×ULN	0/13	21/25 (84.0%)	10/13 (76.9%)	21/24 (87.5%)
≤ULN	0/13	13/25 (52.0%)	4/13 (30.8%)	9/24 (37.5%)

Percentage of patients achieving normalisation of 24-Hour Urinary Oxalate Corrected for BSA by study visit during the double-blind period of the study are shown in **Figure 23**.

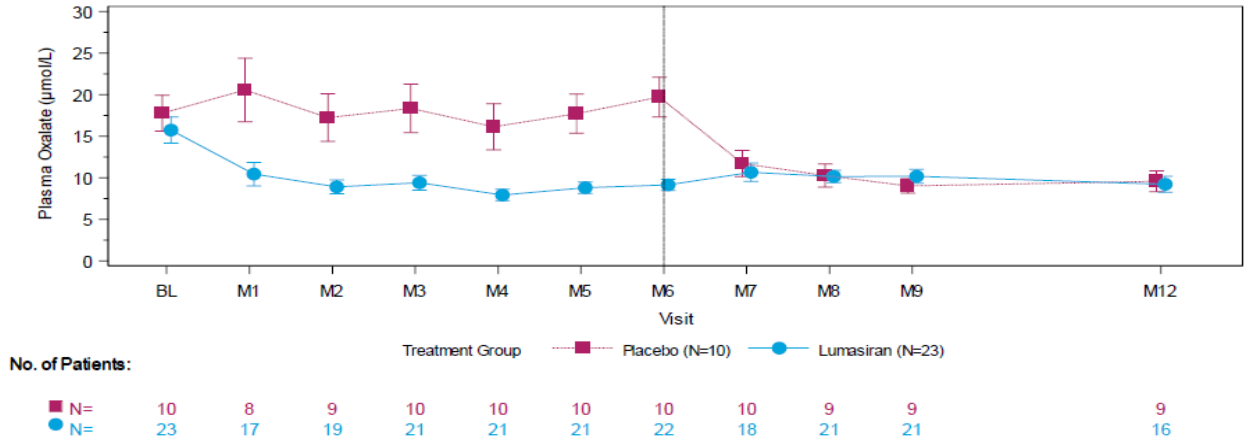
Figure 23. Proportion of patients with 24-hour urinary oxalate corrected for BSA ≤ULN at each visit during the DB period of study ALN-G01-003 (FAS)



Absolute change in Pox (µmol/l) from baseline

Plasma oxalate endpoints were evaluated using the prespecified Plasma Oxalate Analysis Set, which included patients who received study drug and had a baseline plasma oxalate level ≥1.5×LLOQ. In this analysis (**Figure 28**), patients with lower baseline plasma oxalate levels were excluded.

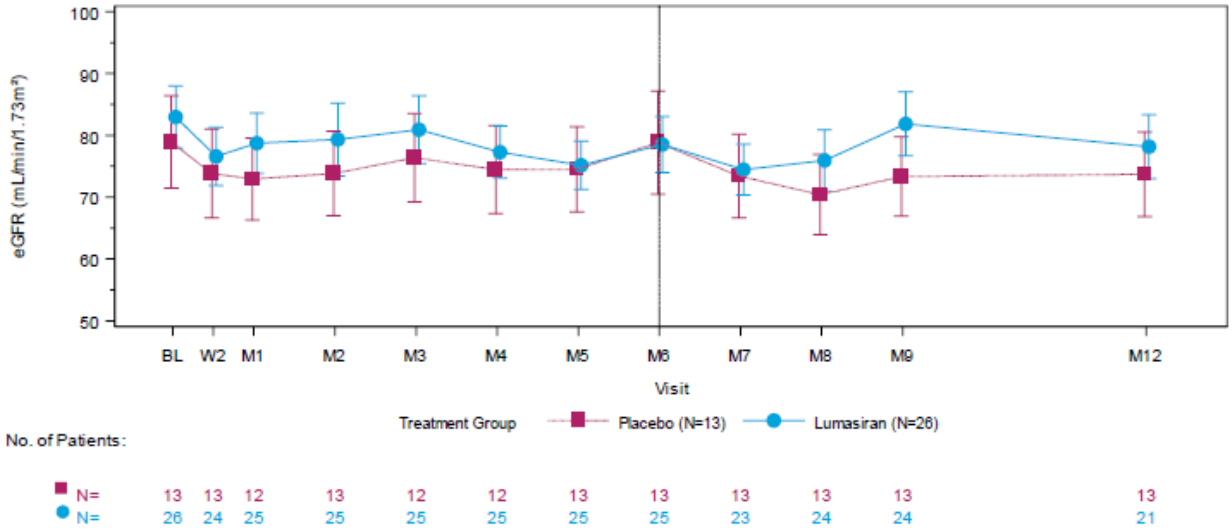
Figure 24. Absolute change from baseline in plasma oxalate (µmol/L) during study ALN-G01-003 (Plasma Oxalate Analysis Set)



Abbreviations: BL=baseline; DB=double-blind; LLOQ=lower limit of quantification; M=month; SEM=standard error of the mean.

Change in estimated glomerular filtration rate (eGFR) from baseline

Figure 25. Actual Values in eGFR (mL/min/1.73m2) at Each Visit study ALN-G01-003 (FAS)



Exploratory endpoints

Renal stone events

The pre- and post-treatment stone event rates are shown separately for the active (**Table 30**) and placebo (**Table 31**)-treated patients.

Table 30. Summary of rate of renal stone events in patients randomised to lumasiran by time period, Study ALN-G01-003 (All Lumasiran Treated Set)

Statistic	12M Period Before Informed Consent	Screening (≤60 days)	DB Period (Day 1 to M6)	Extension Period (>M6 to M12)
Patients randomized to lumasiran, N=26				
Patients with ≥1 postbaseline renal stone event, n (%)	11 (42.3)	5 (19.2)	5 (19.2)	7 (26.9)
Total no. of renal stone events	83	9	13	10
Total no. of person-days	9497	1217	4359	4285
Rate per 100 person-days[a]	**0.87**	**0.74**	**0.30**	**0.23**
95% CI	(0.70, 1.08)	(0.38, 1.42)	(0.17, 0.51)	(0.13, 0.43)

Abbreviations: DB=double blind; CI=confidence interval; M=month.
Note: A renal stone event is defined as a patient-reported event that includes ≥1 of the following: visits to a healthcare provider because of a renal stone; medication for renal colic; stone passage; macroscopic hematuria due to a renal stone.
[a] Rate is calculated as (total number of renal stone events divided by total person-days at risk)*100.

Table 31. Summary of rate of renal stone events in patients randomised to placebo by time period, ALN-G01-003 (All Lumasiran Treated Set)

Statistic	12M Period Before Informed Consent	Screening (≤60 days)	DB Period (Day 1 to M6) (Placebo Treatment)[b]	Extension Period (>M6 to M12) (Lumasiran treatment)
Patients randomized to placebo, N=13				
Patients with ≥1 postbaseline renal stone event, n (%)	4 (30.8)	0	2 (15.4)	1 (7.7)
Total no. of renal stone events	7	0	4	1
Total no. of person-days	4748	628	2211[c]	2200
Rate per 100 person-days[a]	**0.15**	**0.00**	**0.18**	**0.05**
95% CI	(0.07, 0.31)	(0.00, 0.59)	(0.07, 0.48)	(0.01, 0.32)

Abbreviations: DB=double blind; CI=confidence interval; M=month.
Note: A renal stone event is defined as a patient-reported event that includes ≥1 of the following: visit to a healthcare provider because of a renal stone; medication for renal colic; stone passage; macroscopic hematuria due to a renal stone.

Nephrocalcinosis development

Renal ultrasound findings for nephrocalcinosis were graded for each kidney, with a range of 0 to 3, with a higher grade indicating greater severity. For the overall change in grade relative to baseline, "improving" was defined as bilateral improvement or unilateral improvement and no change on the opposite side; "worsening" was defined as bilateral worsening or unilateral worsening and no change on the opposite side; and "indeterminate" was defined as unilateral improvement with worsening on the opposite side. Results for the Double-blind and extension phases of Study ALN-G01-003 are presented in **Table 32**.

Table 32. Nephrocalcinosis grading and change in grade, study ALN-G01-003

	Month 6 (DB Period; FAS)		Month 12 (Extension Period; All Lumasiran Treated Set)	
	Placebo (n/N=12/13)	Lumasiran (n/N=22/26)	Placebo/ Lumasiran (n/N=6/13)	Lumasiran/ Lumasiran (n/N=11/26)
Improvement	0	3	1	5
Unilateral improvement	0	2	1	1
Bilateral improvement	0	1	0	4
Worsening	1	0	0	2
Unilateral worsening	1	0	0	1
Bilateral worsening	0	0	0	1
No change	11	19	5	4

Abbreviations: DB=double-blind; FAS=full analysis set.
n=number of patients who had renal ultrasounds at both baseline and at the time point indicated; N=number of patients in analysis set.

Plasma glycolate levels:

As demonstrated in the studies 001 and 002, glycolate can be considered an indicator of PD activity. In study 003, the changes during the double-blind period are summarised in **Table 33**.

Table 33. Absolute change from baseline in plasma glycolate (µmol/L) during the DB period (FAS)

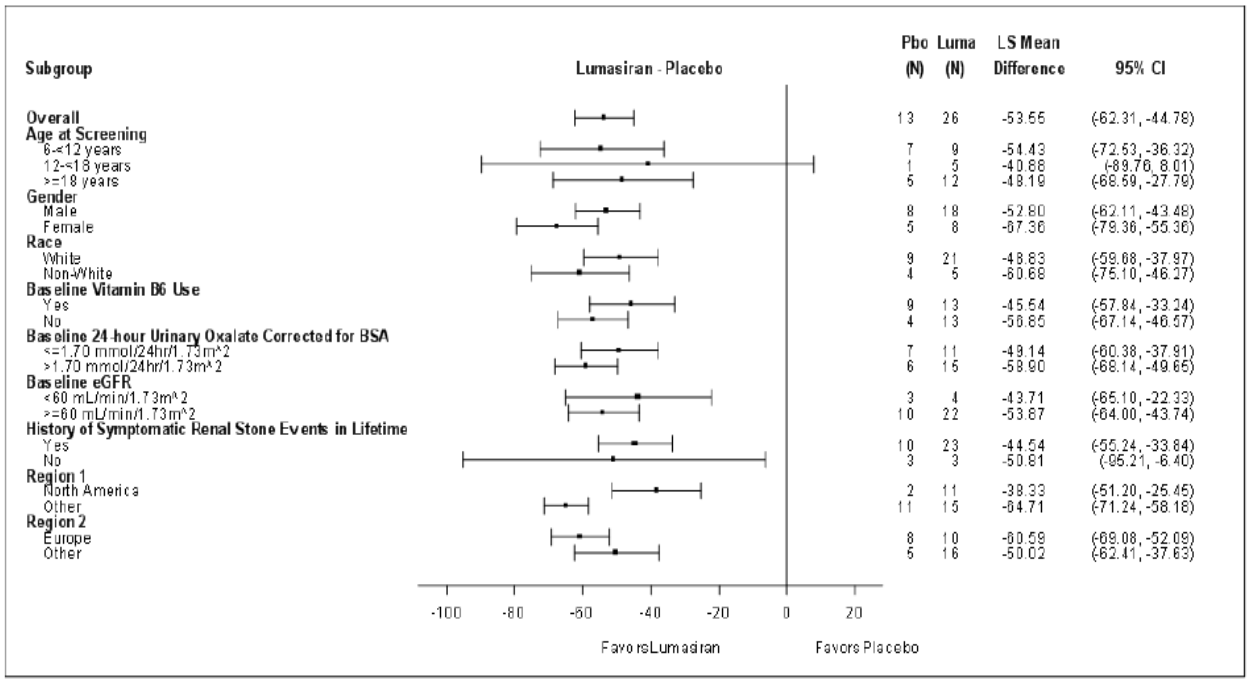
Visit	Statistic	Placebo (N=13)	Lumasiran (N=26)
Baseline[a]	Mean (SEM)	107.4 (13.73)	122.1 (12.35)
Absolute Change from Baseline			
Month 1	Mean (SEM)	32.7 (15.22)	64.0 (14.38)
Month 2	Mean (SEM)	9.5 (13.29)	81.1 (10.89)
Month 3	Mean (SEM)	21.5 (8.51)	98.8 (18.92)
Month 4	Mean (SEM)	17.6 (9.11)	99.7 (20.31)
Month 5	Mean (SEM)	-3.3 (10.81)	80.3 (12.56)
Month 6	Mean (SEM)	13.8 (13.37)	100.4 (12.63)

Ancillary analyses

Subgroup analyses:

Several subgroup analyses were conducted with the primary endpoint, which are displayed in **Figure 26**.

Figure 26. Forest plot of treatment difference in percent change from baseline in 24-hour urinary oxalate corrected for BSA (mmol/24hr/1.73m^2) during the DB period of study ALN-G01-003, MMRM (FAS)



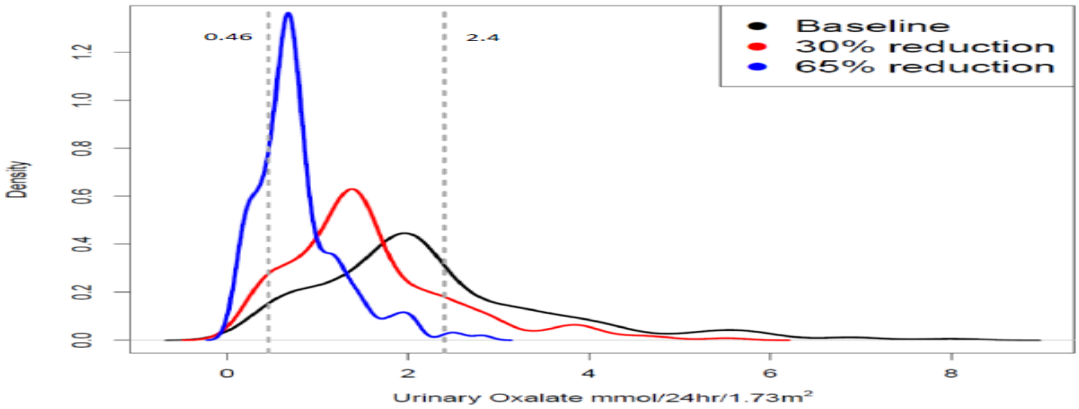
Urinary and Plasma Oxalate as Measures of Clinical Benefit in the Treatment of Primary Hyperoxaluria Type 1

In order to predict the clinical benefit of any given percentage reduction in hepatic oxalate production for PH1 patients with relatively preserved renal function, a 2-part analysis using published data was performed. In Part 1, the effect of reductions in hepatic oxalate production on UOx values in the PH1 population without ESRD was modelled. In Part 2, the UOx distributions to ESRD outcomes was linked.

Modelling Part 1

Recently-published registry data describing 192 patients with PH1 who presented without ESRD and had 24-hour UOx measures available from their time of diagnosis (Zhao et.al, 2016) were used to model the clinical effect (i.e. progression to ESRD) of reduced hepatic oxalate production on UOx in patients with PH1 and relatively preserved renal function (eGFR >45 mL/min/1.73m2), (**Figure 27**).

Figure 27. Model of population distribution of PH1 patients at baseline and two different percentage reductions in urinary oxalate



The y-axis of the histogram represents the relative frequency of each oxalate value in the population. Dashed vertical lines indicate values of urinary oxalate excretion that are 0.46 mmol/24hr/1.73m2 (ULN) and 2.4 mmol/24hr/1.73m2, the latter of which is associated with an increased risk of developing ESRD at 30 years (Figure 13). [Zhao et.al, 2016]

Modelling Part 2

Using extrapolation of data on renal survival generated for all types of PH in the RKSC registry (Zhao et.al, 2016), the number of patients who are prevented from developing ESRD at 30 years follow up given the simulated reductions in UOx that was modelled in Part 1 was estimated (**Table 34**).

Table 34. Model of population distribution of PH1 patients clinical events according to simulated reductions in urinary oxalate from baseline.

	Baseline	Reduction in Urinary Oxalate from Baseline												
		20%	25%	30%	35%	40%	45%	50%	55%	60%	65%	70%	75%	80%
ESRD events per 100 patients	39	33	32	30	28	27	25	25	23	22	22	21	21	21
ESRD events prevented per 100 patients, relative to baseline	-	6	7	9	11	12	14	14	16	17	17	18	18	18
Percent reduction in ESRD events, relative to baseline	-	15%	18%	23%	28%	31%	36%	36%	41%	44%	44%	46%	46%	46%
ESRD events per 100 patients, ***assuming no risk for those with normal urinary oxalate levels***	39	32	31	29	26	24	22	22	20	19	17	15	12	8
ESRD events prevented per 100 patients, relative to baseline, ***assuming no risk for those with normal urinary oxalate levels***	-	7	8	10	13	15	17	17	19	20	22	24	27	31
Percent reduction in ESRD events, relative to baseline, ***assuming no risk for those with normal urinary oxalate levels***	-	18%	21%	26%	33%	38%	44%	44%	49%	51%	56%	62%	69%	79%

Summary of main study

The following tables summarise the efficacy results from the main studies supporting the present application. These summaries should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 35. Summary of efficacy for trial ALN-GO1-003

Title: 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 (ILLUMINATE A)	
Study identifier	ALN-GO1-003 EudraCT Number: 2018-001981-40
Design	Randomised, Double-Blind, multi-centre, Placebo-Controlled Study with an Extended Dosing Period

	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:		6 months double-blind phase No run-in phase Up to 54 months open-label treatment
Hypothesis	Superiority against placebo		
Treatments groups	Lumasiran		Lumasiran 3 mg/kg SC per month for 3 months; 3 mg/kd SC every three months thereafter. In order to keep the blind in the initial open-label phase, monthly injections of placebo were administered at month 7 and 8
	Placebo		Placebo treatment as scheduled for active. Lumasiran active treatment schedule commencing at month 6 as indicated for active treatment group.
Endpoints and definitions	Primary endpoint	Urinary oxalate	% change in 24h UOx corrected for BSA (mmol/24hr/1.73m^2) MMRM (Average of M3-M6)
	Secondary Endpoints:	Urinary oxalate	Absolute change in 24 UOx corrected for BSA (mmol/24hr/1.73m2) MMRM from baseline to M6, MMRM (Average of M3-6)
		Urinary oxalate related to creatinine concentration	Percent change in 24 UOx:creatinine ratio (mmol/mmol/l) from baseline to M6, MMRM (Average of M3-6)
		Plasma oxalate	Percent change in POx (µmol/l) from baseline to M6, MMRM (Average of M3-6)
		Normal or near normal urinary oxalate excretion	Proportion of patients with 24h UOx corrected for BSA ≤1.5xULN at month 6
		Normal oxalate urinary oxalate excretion	Proportion of patients with 24 UOx corrected for BSA≤ULN at month 6.
		Change in Plasma oxalate	Absolute change in POx (µmol/l), from baseline to M 6 MMRM, Average of M3-6.
		Glomerular filtration rate	Change in eGFR from baseline to month 6
Database lock	Interim database lock: 25 October 2019		

<u>Results and Analysis</u>

Analysis description	**Primary Analysis**
Analysis population and time point description	Intent to treat (FULL ANALYSIS SET) Time point: 6 months

Descriptive statistics and estimate variability	Treatment group		Lumasiran	Placebo
	Number of subject		26	13
	PEP: 24 hr UOx % change from BL to M6 (LSM; SEM)		-65.385 (2.9383)	-11.8838 (3.8132)
	Absolute change in 24 UOx corrected for BSA (Mean; SEM)		-1.242 (0.0610)	-0267 (0.0831)
	Percent change in UOx:creatinine ratio (mmol/mmol) from baseline to M6, MMRM (Average of M3-6) (Mean; SEAM)		-62.5469 (4.04475)	-10.7751 (5.35366)
	Percent change in POx (µmol/l) from baseline to M6, MMRM Average of M3-6) (Mean, SEM)		-39.80 (2.938)	-0.32 (4.293)
	Proportion of patients with 24h UOx corrected for BSA ≤1.5xULN at month 6 (n/n; %)		21/25 (84%)	0
	Proportion of patients with 24 UOx corrected for BSA≤ULN at month 6. (n/n; %)		13/25 (52.0%)	0
	Absolute change in Pox(µmol/l) from baseline to M6, MMRM (average of M3-6) (Mean; SEM)		-7.46 (0.766)	1.25 (1.121)
Effect estimate per comparison		Measure	Lumasiran vs. Placebo	
			Statistic	p-value
	Primary endpoint 24 hr UOx % change from BL to M6	Difference in LS Mean (95% CI)	53.546 (62.314, 44.778)	1.685E-14
	Secondary endpoint: Absolute change in 24 UOx corrected for BSA from BL to M6	Difference in LS Mean (95% CI)	-0.975 (-1.177, -0.772)	1.225E-11
	Percent change in 24h UOx:creatinine ratio (mmol/mmol) from baseline to M6	Difference in LS Mean (95% CI)	-51.7718 (64.2653, 39.2784)	5.032E-10
	Percent change in POx (µmol/L), from baseline to M6	Difference in LS Mean (95% CI)	-39.48 (-50.10, -28.87)	2.862E-08

	Proportion of patients with 24h UOx corrected for BSA ≤1.5×ULN at Month 6	Difference in proportion (95%CI)	0.84 (0.55, 0.94)	8.341E-07
	Proportion of patients with 24h UOx corrected for BSA ≤ULN at Month 6	Difference in proportion (95%CI)	0.52 (0.23, 0.70)	0.0010
	Absolute change in POx (µmol/L), from baseline to M6, MMRM (Average of M3-6)	Difference in LS Mean (95% CI)	-8.71 (-11.45, -5.98)	3.893E-07

Supportive studies

Study ALN-G01-004 (ILLUMINATE B)

Study ILLUMINATE B is an ongoing Open-Label Study to Evaluate the Efficacy, Safety, Pharmacokinetics, and Pharmacodynamics of Lumasiran in Infants and Young Children with Primary Hyperoxaluria Type 1. The study was initiated on 22 April 2019 and the most recent interim reports submitted has a data cut-off date of 30 June 2020. This study consists of 2 periods: a 6-month Primary Analysis Period followed by a Long-term Extension Period of 54 months.

Enrolment in the study has been completed. 18 patients have completed the 6-month primary analysis period and entered into the Long-term Extension Period. No patients have discontinued treatment or withdrawn from the study.

This study recruited patients aged at least 37 weeks estimated gestational age but less than 6 years of age, and a documented diagnosis of PH 1. Urinary oxalate:creatinine ratio had to be greater than the upper limit of normal based on age on at least 2 of 3 single-void collections during screening. Other inclusion criteria were resembling those of study 003, e.g. exclusion of relevant other disease, and intake of pyridoxine.

The proposed primary endpoint is the percent change in urinary oxalate excretion from baseline to Month 6, Secondary endpoints include absolute change in urinary oxalate excretion from baseline, the proportion of patients with urinary oxalate excretion ≤ the upper limit of normal (ULN) and ≤ 1.5 x ULN, absolute and percent change of oxalate in plasma, plasma PK parameters of lumasiran, and the change from BL in eGFR.

Ten of 18 enrolled patients (55.6%) were female and 16 were white (88.9%). Median age at consent was 50.1 months (range: 3 to 72 months); 2 patients were <1 year of age, 2 patients were 1 to < 2 years of age and 14 patients were 2 to <6 years of age. Median body weight at first lumasiran dose was 14.5 kg (range: 6.2 to 24.3 kg); 3 patients weighed < 10 kg, 12 patients weighed 10 to<20 kg and 3 patients weighed ≥20 kg.

Baseline spot urinary oxalate:creatinine ratio, plasma oxalate, and plasma glycolate were highest in patients who weighed<10 kg (n=3) and lower in the 10 to< 20 kg (n=12) and ≥20 kg (n=3) initial weight dose groups.

The cumulative median duration of exposure (N=18) to lumasiran was 10.51 months (range: 5.6 to 13.4 months) during the study. The mean number of doses received per patient was 6.8 (range: 5 to 12 doses) and the cumulative number of doses administered was 122; no doses were missed

The results for the primary endpoint of percent change from baseline in spot urinary oxalate:creatinine ratio in the primary interim efficacy analysis set are summarised in **Table 36**.

Table 36. Percent change from baseline in spot urinary oxalate:creatinine ratio in the primary interim efficacy analysis Month 6 vs. baseline of Study ALN-G01-004 (Interim Efficacy Analysis Set)

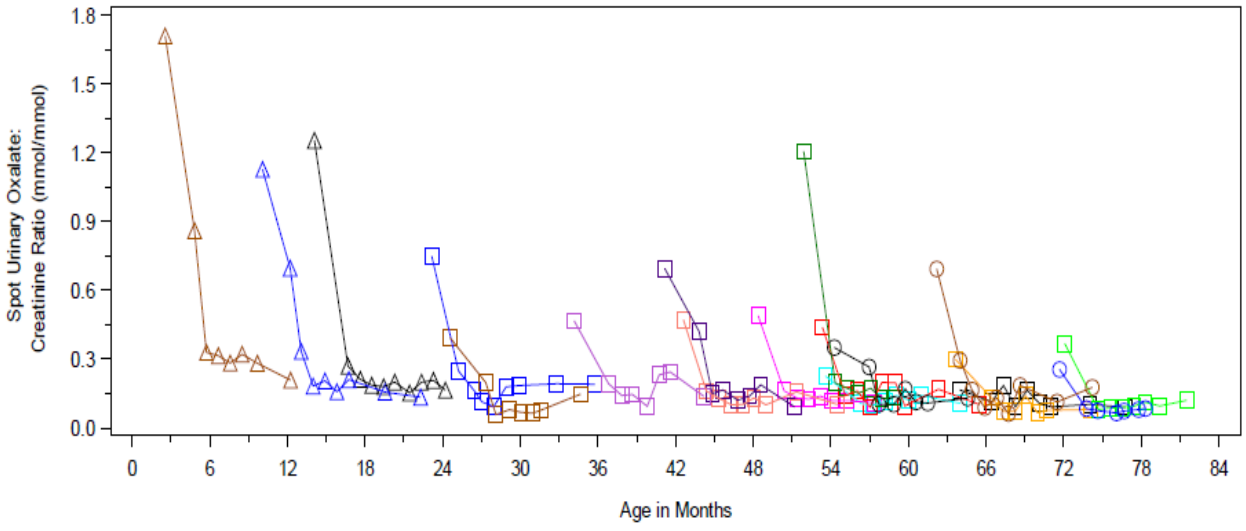
Analysis (N=18)	LS Mean (SEM)	95% CI
Percent change from baseline in spot urinary oxalate:creatinine ratio (mmol/mmol) to Month 6 (average from Month 3 to Month 6)	-71.97 (2.706)	-77.52, -66.42
Sensitivity: Percent change from baseline in ULN* ratio (ratio of spot urinary oxalate:creatinine to ULN) to Month 6 (average from Month 3 to Month 6)	-70.19 (2.619)	-75.56, -64.82

CI=confidence interval; LS=least squares; SEM=standard error of the mean; ULN=upper limit of normal

**Age-dependent ULN based on [Matos 1999]*

A decrease in spot urinary oxalate:creatinine ratio was observed in all age groups (**Figure 28**).

Figure 28. Actual values in spot urinary oxalate:creatinine ratio (mmol/mmol) by age during the primary analysis and extension periods of Study ALN-G01-004 (Primary Interim Efficacy Analysis Set).



The results of the secondary efficacy evaluations are presented in **Table 37**.

Table 37. Secondary interim efficacy results at month 6 of Study ALN-G01-004

Analysis (N=18)	Parameter	Result
Absolute change from baseline in spot urinary oxalate:creatinine ratio (mmol/mmol)	LS Mean (SEM)	-0.4925 (0.01452)
	95% CI	-0.5233, -0.4617
Percent change from baseline in plasma oxalate	LS Mean (SEM)	-31.73 (3.796)
	95% CI	-39.53, -23.94
Percent change from baseline in plasma oxalate for patients with baseline plasma oxalate ≥1.5 × LLOQ (n=13)	LS Mean (SEM)	-39.35 (4.793)
	95% CI	-49.43, -29.28
Absolute change from baseline in plasma oxalate (µmol/L) for patients with baseline plasma oxalate ≥1.5 × LLOQ (n=13)	LS Mean (SEM)	-6.92 (0.670)
	95% CI	-8.32, -5.51
Absolute change from baseline in plasma oxalate (µmol/L)	LS Mean (SEM)	-5.23 (0.487)
	95% CI	-6.23, -4.23
Proportion of patients with spot urinary oxalate excretion	≤ULN	1 (5.6%)
	≤1.5 × ULN	9 (50%)
Change from baseline in eGFR (ml/min/1.73m^2) (n=16)*	Mean (SD) change	-0.263 (15.3845)
	Mean (SD) % change	0.87 (12.048)

CI=confidence interval; eGFR=estimated glomerular filtration rate; LLOQ=lower limit of quantification; LS=least squares; SD=standard deviation; SEM=standard error of the mean
**Change in eGFR can only be evaluated in patients at least 12 months of age at baseline.*

Study ALN-G01-005 ("Study 005"):

Study 005 is an ongoing, multicenter, Phase 3, single-arm study to evaluate the efficacy, safety, PK, and PD of lumasiran in patients with PH1 who have advanced renal disease, as evident by eGFR ≤45 ml/min/1.73m2 (or serum creatinine elevated for age, in patients <12 months of age). No study report has been submitted, but the results of the data collected so far were presented (Data cut-off point 20 May 2020).

The study comprises a 6-month primary analysis period and a long-term Extension Period of up to 54 months and 2 cohorts: Cohort A - patients who do not yet require dialysis, and Cohort B - patients on haemodialysis.

Initial results are available in 4 patients (Cohort B; 2 male and 2 female) with the mean age of 7.8 (range: 3 to 16) years. The overall mean (SD) baseline plasma oxalate measured was 129.6 (32.1) µmol/L. Three patients have received 2 doses of lumasiran, and 1 patient has received 3 doses.

Data showed a reduction in plasma oxalate ≥33% in 3 patients at Month 1, suggesting that lumasiran may be effective in the patients with severe renal impairment receiving haemodialysis.

2.5.3. Discussion on clinical efficacy

Design and conduct of clinical studies

As discussed in the clinical pharmacology section of this report, the applicant did not conduct a dedicated dose-finding study, but the proposed dosing was determined from data from studies 001 and 002, and an extensive modelling exercise.

The pivotal study conducted within the programme was Study 003, which is a randomised double-blind trial with a 6 months double-blind treatment period, and a long-term open-label extension period which is ongoing at the time of submission and projected to last for 54 months for all patients included. The study included patients with an age from 6 and older with a confirmed diagnosis of PH 1 and a 24-hour urinary oxalate excretion rate of more than 0.70 mml/24hr/1.73 m^2. Patients taking Vitamin B-6 could be included if stable conditions could be assured. No requirements on other standard background treatments (e.g. hyperhydration) were imposed. The patients had to have a relatively preserved renal function with at least an eGFR of <30 mL/min/1.73 m^2 at screening.

Overall, the included patient population is acceptable for the claimed indication even though patients appear to represent the mild to moderate spectrum of the disease.

This trial used similar endpoints to the preceding studies 001B and 002, based on urinary oxalate excretion parameters (change in 24-hour urinary oxalate excretion corrected for BSA (both absolute and relative changes), change in 24-hour urinary oxalate:creatinine ratio; change in plasma oxalate, and change in eGFR. Additionally, categorical responder-type analyses were added with the proportion of patients achieving near normal (at or below 1.5xULN) or normal 24-hour urinary oxalate levels.

The applicant did not follow the initial protocol assistance advice to rank the categorical evaluation of normal or near normal oxalate urinary excretion as primary, however the proposed ranking and subsequent control of the type I error was intended to assure the compliance with this request.

The primary analysis was performed using a restricted maximum likelihood (REML) based Mixed-Effect Model Repeated Measures (MMRM) approach, which included two sensitivity analyses with different co-variates and time-points to be imputed. Relevant subgroups according to age, gender, race, and baseline urinary oxalate excretion, as well as history of stone events, vitamin B6 intake, and region were also pre-defined.

The design of the trial is generally considered adequate and suitable to document the efficacy of lumasiran, even though the short duration of the placebo-controlled treatment phase does not allow to draw conclusions on longer term treatment effects. Additional data from this study are expected to better characterise the long-term effects of lumasiran (see efficacy data and additional analysis below).

The lab-based biomarker endpoints are surrogate endpoints for the clinical outcomes events of interest, such as the development of renal function over time (including manifestation of ESRD), the occurrence of renal stone events, and the development of nephrocalcinosis, as well as in late stage of disease, the development of oxalosis of other organs with the consequent impairments of function (e.g. heart with heart failure). The use of these surrogate endpoints is supported by their high theoretical biological plausibility. In addition, the applicant provided a rationale for the choice of these endpoints, primarily based on literature reports on the prognostic value of urinary oxalate excretion in "renal survival" and development and occurrence of kidney stones and nephrocalcinosis (e.g. Zhao et.al, 2016; Hoppe et.al, 2015, Milliner, et.al, 2020).

The applicant has also submitted interim results from an open-label study in children aged under 6 years (study 004). This trial, similar to 003, also includes a 6 months primary analysis period, and a

long-term extension period up to 54 months. Due to the open-label design of this study, only baseline comparisons are included. In addition, due to the young age of the study population only a sub-population was able to provide 24-hour urine collections, which is considered acceptable.

The primary evaluation is urinary oxalate excretion based on the % change in spot urine Urinary oxalate:creatinine Ratio. Additional endpoints reported for this interim report further comprised plasma oxalate, and eGFR changes. The dosing in the young children was a once monthly dose for 3 months similar to older children and adults, which was, however, higher in children with a BW of less than 20 kg. From the fourth dose onwards, the small children were also treated with q3M doses in the weight band above 10 kg BW, with those above 20 kg receiving the adult dose of 3 mg/kg q3M, and those between 10 and 20 kg receiving 6 mg/kg q3M. The children in the weight band under 10 kg were further treated with a monthly dose of 3 mg/kg BW. The inclusion criteria determined that – apart from having a diagnosis of PH1, the patients would need to have an oxalate:creatinine ratio above the upper limit of normal.

Efficacy data and additional analyses

Study 003 included 39 patients randomised 2:1 to active and placebo treatment. There were only two discontinuations of the trial medication during the study. Baseline demographic and disease characteristics data were comparable between the two treatment groups, with some variation in subgroups (e.g. according to age and region) owing to the small sample size. Based on the baseline disease characteristics, the population could be classified of being mildly to moderately affected by PH1 at the time of inclusion, and only a 15-20% of the study population had eGFR below 60 mL/min/1.73m2. There were 5 major protocol deviations during the trial, all of which were not considered to impact on the validity of the data reported. Due to the low number of discontinuations for this initial study period, the number of missing values was limited, and their way of imputation is not considered to be of decisive relevance with regard to overall results.

The primary evaluation of the trial documented a highly significant percent reduction from baseline to month 6 in the 234-hour urinary oxalate corrected for BSA, which was about 65%, as compared to 11 % for the placebo group. The sensitivity analyses were in accordance with the primary evaluation. Similar statistically significant differences were seen in the absolute changes of this parameter. Continued treatment with lumasiran (lumasiran/lumasiran group) in the extension period led to maintenance of this reduction through Month 12, with a mean percent change from baseline in 24-hour urinary oxalate corrected for BSA of 64.1% at Month 12. Similar to patients randomised to lumasiran, placebo crossover patients had a rapid and sustained decrease in the 24-hour urinary oxalate corrected for BSA, with declines seen at 1 month after starting lumasiran and a similar time course and magnitude of decrease as lumasiran patients in the DB Period. In these patients at Month 12 (6 months of lumasiran treatment), the mean percent change from baseline in 24-hour urinary oxalate corrected for BSA was -57.3% relative to the first dose of lumasiran in the extension period.

The evaluation of the 24-hour urine oxalate:creatinine ratio also revealed a just over 60% relative decrease compared with an about 11% decrease in placebo. Plasma oxalate decreased by about 40% for active treatment and remained virtually unchanged in the placebo group. A review of the percent change from baseline in 24-hour urinary oxalate:creatinine ratio at each visit during the extension period showed increases in the lumasiran/lumasiran group at Months 7 and 8, with subsequent decreases observed at Months 9 and 12. Placebo crossover patients also showed an increase in 24-hour urinary oxalate:creatinine ratio at Month 7, 1 month after starting lumasiran treatment. An investigation was conducted into these unexpected results identified inadequate handling of 24-hour urine creatinine samples by an analyst. A sensitivity analysis excluding affected urine creatinine

samples showed a consistent decrease in the in 24-hour urinary oxalate:creatinine ratio, for the duration of the study following initiation of lumasiran treatment.

The categorical evaluation of patients achieving a "near normal" or "normal" urinary oxalate excretion was 84% and 52% respectively in the lumasiran treated patients during the double-blind phase with no patients in the placebo group achieving the thresholds. The evaluation of the absolute change in plasma oxalate was also in favour of the active treatment group, and highly statistically significant. All the secondary endpoints mentioned were included in a hierarchical testing procedure, and due to the significant results in all endpoints, this did not have to be stopped at any point.

Plasma glycolate levels increased in actively treated patients, reflecting the PD activity expected whereas only some fluctuations occurred in placebo patients.

Inconclusive results were reported for the clinical endpoints such as eGFR changes, renal stone events, and the ultrasound-based evaluation of nephrocalcinosis in the DB period of the study, that could be explained with the short duration of this period. Analysis of the 1-year data on renal stone events in the placebo - lumasiran vs. lumasiran – lumasiran group in conjunction with the pre-treatment rates do, however, suggest some trends to improvement with respect to renal events findings on active treatment, when compared to placebo or to baseline. Similar support could be derived from the ultrasound-based evaluation of nephrocalcinosis.

Additional supportive evidence for the surrogate efficacy parameters chosen in the prevention of clinical events can be derived from case reports of patients with liver transplant (Cochat et, al, 1989; Coulthard and Lodge 1993; Galanti and Contreras 2010), as well as from studies on patients responding to Vitamin B6 treatment (van Woerden et.al, 2003).

Finally, the applicant developed a mathematical model, according to which treatment with lumasiran would over the course of 30 years prevent 17-22 ESRD events per 100 patients treated. However, it would be recommended to update this model once data from the recently initiated study 005 in patients with renal impairment become available.

The applicant also presented the pre-planned sub-group analyses according to age, gender, race, Vitamin B6 intake, baseline urinary oxalate, baseline eGFR, history of stone events, and two different evlauations of the region (North America vs. RoW and Europe vs. RoW). All results for the primary evaluation of this subgroup analysis showed consistency with the main outcome, except in the subgroup of adolescents, which is highly likely due to the small number of patients included (1 and 5 in the two groups). The results of this subgroup analysis add further reassurance on the robustness of the overall results.

Available data from study 004 suggest that similar effects as seen in older children and in adults can be achieved in the very young paediatric population.

Lumasiran treatment in this study resulted in a consistent decrease of urinary oxalate excretion, as measured by the spot urine oxalate:creatinine ratio. The decrease was more pronounced in patients with higher baseline oxalate levels. The mean eGFR remained relatively stable during the trial. However, it is noted that the variability is very high in the included 14 patients.

Secondary endpoints other than the eGFR exclude a deterioration in the clinical condition of these patients. However, it is acknowledged that beneficial effects on renal function are unlikely to become visible after a short-term treatment of 6 months. Similar to study 003 for the older population, the ultrasound-based assessment of nephrocalcinosis appears to indicate some improvement across the still limited observation period.

Extrapolation of the adult data lends additional support for use of lumasiran in the young paediatric population as the pathophysiology of the disease, and the mechanism of action of lumasiran are the

same across the different age groups. As similar treatment effect could be expected therefore for adults and paediatric patients. This is confirmed by comparing the parameters used for urinary oxalate excretion in studies 003 and 004, which indeed show a similar magnitude of effect.

The CHMP noted however, that data remain limited particularly in the most vulnerable population of the age of less than 1 or even 2 years, which were represented with 2 and 4 patients respectively and more data are expected in this sub-population through the ongoing studies 004 and 005.

The long-term effects of lumasiran treatment on clinical events cannot be adequately evaluated within the timeframe of this procedure. Open-label extensions of all studies that the applicant has initiated in this condition are ongoing, and will potentially be able to provide more comprehensive data on long-term effects, However, as these data will be collected in a non-comparative fashion the CHMP has recommended that the applicant should further characterise the natural history of the diseases, using existing registries. Information from such a study with untreated (or SOC-treated) patient population could be used as an additional demonstration of the benefit of the lumasiran-treated patients in the ongoing studies.

2.5.4. Conclusions on the clinical efficacy

Overall, efficacy of lumasiran in patients with primary hypeoxaluria type 1 has been demonstrated.

Results from study 003 demonstrate strong and consistently statistically significant effects on the surrogate parameters chosen and show that oxalate production and excretion can be normalised in large percentage of treated patients. These results in conjunction with the high biological plausibility that this will translate to an improvement in clinically relevant events in the long run, and the positive trends in the clinical events observed, is considered a sufficient proof of efficacy.

The observed results in the 18 patients treated for at least 6 months in study 004, further supports the use of lumasiran in all age groups.

Additional data are expected from ongoing studies which will provide information on the long-term treatment effect and in the younger paediatric population.

2.6. Clinical safety

Patient exposure

In total, 105 subjects, including 81 patients with PH1 and 24 healthy volunteers are included in the lumasiran safety database. This includes 4 patients with terminal renal insufficiency from the 005 study (data cut-off point 14 May 2020) and the last 2 enrolled patients form study 004 that at the time of data cut-off had completed the 2-month visit but are not included in the pooled data-set.

Seventy-five patients with PH1, including 54 paediatric patients are included in the overall pooled analysis with data cut-off dates of 30 January, 14 February and 09 March 2020 for studies 002, 003 and 004, respectively. Thirty-nine patients provided placebo-controlled data (26 on lumasiran and 13 on placebo) in study 003 and all of these completed the DB phase at the time of data cut-off.

Information provided in this section refers to this overall pooled safety population of 75 patients unless otherwise stated. The extent of exposure for overall pooled experience is presented in **Table 38**.

Table 38. Exposure to lumasiran in the overall pooled safety analysis set (Data cut-off 09 March 2020)

	Pooled Lumasiran Experience					
	Age ≥6 years				Age <6 years	
	Study 002		Study 003*		Study 004*	
Parameter	1 mg/kg qM or 3 mg/kg q3M (N=13)	3 mg/kg qM (N=7)	3 mg/kg qM for 3 consecutive months; then, 3 mg/kg q3M (N=39)	Total (Age ≥6 years) (N=59)	**<10 kg:** 6 mg/kg qM x 3; then, 3 mg/kg qM (n=3) **10 to <20 kg:** 6 mg/kg qM x 3; then, 6 mg/kg q3M (n=11) **≥20 kg:** 3 mg/kg qM x 3; then, 3 mg/kg q3M (n=2) (N=16)	Total (All Patients) (N=75)
Total duration of drug exposure (months)						
Mean (SD)	12.09 (3.13)	9.55 (2.68)	4.48 (2.82)	6.75 (4.33)	2.63 (1.06)	5.87 (4.22)
Median (min, max)	12.94 (6.8, 17.1)	9.36 (6.6, 14.8)	5.81 (0.1, 9.3)	6.31 (0.1, 17.1)	2.69 (1.3, 5.2)	5.85 (0.1, 17.1)
Number of patients on study drug for, n (%)						
≥1 day	13 (100.0)	7 (100.0)	39 (100.0)	59 (100.0)	16 (100.0)	75 (100.0)
≥3 months	13 (100.0)	7 (100.0)	26 (66.7)	46 (78.0)	4 (25.0)	50 (66.7)
≥6 months	13 (100.0)	7 (100.0)	16 (41.0)	36 (61.0)	0	36 (48.0)
≥9 months	10 (76.9)	5 (71.4)	1 (2.6)	16 (27.1)	0	16 (21.3)
≥12 months	8 (61.5)	1 (14.3)	0	9 (15.3)	0	9 (12.0)
≥15 months	2 (15.4)	0	0	2 (3.4)	0	2 (2.7)

Abbreviations: q3M=once every 3 months; qM=monthly; max=maximum; min=minimum; SD=standard deviation.
*Dose range as proposed in the SmPC.

Adverse events

Overall, frequency of AEs observed in the placebo-controlled setting was higher on lumasiran as compared to placebo (102 in 84.6% patients vs. 18 AEs in 69.2% patients), with clearly more patients on lumasiran having drug-related AEs (42.3% of patients with 33.3% AEs vs. 7.7% of patients with 5.6% AEs). Average number of AEs per patient in the patients with at least 1 AE was higher on lumasiran than on placebo (4.6 vs. 3 AEs per patient). Two patients stopped treatment due to an AE on lumasiran treatment, including 1 drug-related AE that was subsequently updated to not related by the Investigator.

In the pooled data analyses majority of the patients (77.3%) reported at least 1 AE during lumasiran treatment (215 AEs in total and 3.7 AEs per patient with an AE) and cross-study analysis showed some differences between the studies. All patients > 6 years developed an AE on the highest dose tested (3 mg/kg qM; 002 study; 7/7 patients; 25 AEs; 3.6 AEs/patient), whereas, the largest study (003) evaluating the recommended therapeutic dose had the lowest proportion of patients with AEs (66.7%) across the pooled studies. The lowest number of AEs per patient (2.3 AE/patient) was reported on the lowest lumasiran dose (002 study; 1 mg/kg qM, or 3 mg/kg q3M dose arm) tested, whereas the highest average number of AEs per patient was in 003 study (127 AEs in 26 patients with AEs; 4.9 AE/patient). Average proportion of the drug-related AEs was about 26% across the pooled studies (56 DRAEs out of 215 AEs) with the lowest number being reported in the youngest population of <6 years of age (12.5% of the patients and 8.6% of the AEs), and the highest in the 003 study (35.9% of the patients and 34,6% of AEs).

When regarding the AE frequencies in the youngest children (< 6 years old; 004 study), proportion of the patients with AEs (81.3% of the patients), or number of the AEs per patient in this population (2.7 AE/patient) did not seems to reveal major differences to the safety profile observed in the older population (002 and 003 studies; range: 66.7 – 100% of patients with AEs; 2.3 to 4.9 AEs/patient).

Common adverse events (AE) and the common AEs which occurred at least 5% more frequently on lumasiran compared to placebo are summarised in **Tables 39** and **40** respectively.

Table 39. Adverse events reported in ≥10% of patients during the placebo-controlled period in Study 003 (Safety Analysis Set)

Preferred Term	Placebo (N=13) n (%)/No. events	Lumasiran (N=26) n (%)/No. events
At least 1 AE	9 (69.2)/18	22 (84.6)/102
Injection site reaction	0	6 (23.1)/10
Headache	3 (23.1)/3	3 (11.5)/3
Injection site erythema	0	3 (11.5)/3
Injection site pain	0	3 (11.5)/3
Rhinitis	2 (15.4)/2	2 (7.7)/2
Upper respiratory tract infection	2 (15.4)/2	2 (7.7)/2

Table 40. Adverse events Reported in ≥10% of patients treated with lumasiran that occurred at least 5% more frequently than in patients treated with placebo during the double-blind period in study 003 (Safety Analysis Set)

	Placebo (N=13) n (%)/No. events	Lumasiran (N=26) n (%)/No. events	Percentage (%) Difference Between Groups
Injection site reactions	0	9 (34.6)/25	34.6
Abdominal pain[a]	1 (7.7)/1	4 (15.4)/6	7.7
[a] Includes abdominal pain, abdominal pain upper, abdominal pain lower, and abdominal discomfort.			

The most frequently observed AEs in the overall pooled experience (≥10% of patients treated with lumasiran) were injection site reaction (24.7%), pyrexia (14.3%), and headache, rhinitis, upper respiratory tract infection, and vomiting (10.4% each). The most frequently observed AEs (≥10% of patients) considered related to lumasiran by the Investigator were injection site reactions (24.7%). Most AEs were mild or moderate in severity; 1 (1.3%) patient had a severe AE of urosepsis that was considered not related to study drug by the Investigator.

When evaluated by medical concept grouping, injection site reactions were reported in 32.5% of patients during the overall pooled experience.

Injection site reactions appeared to reveal patterns both, in the time-to-start, as well as, in the duration. The absolute majority of these events started on the day of the drug administration (Day 0) and resolved on the same day without treatment (pain, discomfort, erythema, swelling, discoloration, mass, and induration) or within the following 2 days (swelling, induration and mass). None of the events, which started at Day 0 lasted longer than 3 days. Some of the AEs, however, emerged with delay, i.e. 2 - 43 days after the drug administration and lasted considerably longer (3 – 100 days). In the majority of the cases these delayed reactions represented a combination of erythema and pruritus, sometimes accompanied with swelling. Overall, pruritus (7 events) occurred always as a delayed AE and was always accompanied by other AEs (mostly erythema). Pain and discomfort started always on Day 0 and resolved on the same day. Only one case of rash and one case of exfoliation were reported in the pooled analysis and had late start and prolonged duration.

Study 004

As of 30 June 2020, all (100%) patients reported at least 1 adverse event (AE), all of which were mild or moderate in severity. The most common AEs (reported in ≥15% of patients) included pyrexia (38.9%), rhinitis, upper respiratory tract infection, and vomiting (22.2% each), and injection site reaction (16.7%). AEs considered related to lumasiran by the Investigator were injection site reactions (ISRs) in 3 (16.7%) patients and headache in 1 (5.6%) patient. All AEs related to lumasiran were mild in severity.

No AEs mapping to the Drug-related Hepatic Disorders Standard MedDRA Query (SMQ) were reported. There were no clinically relevant changes in haematology or blood chemistry values. No clinically relevant elevations in liver function tests (LFTs) were reported and no patients had worst post-baseline alanine aminotransferase (ALT) or aspartate aminotransferase (AST) values >3 × upper limit of normal (ULN) during the study.

There were no deaths, severe AEs, or AEs leading to treatment discontinuation, interruption, or withdrawal from the study.

Study 005

As of the data extraction date of 17 August 2020, 5 (41.7%) patients had had an AE. One (8.3%) patient had a non-serious AE of injection site haematoma that was mild in severity and was considered related to study drug. No other patients have had ISRs or other AEs considered related to study drug. There have been no treatment-emergent deaths, AEs leading to treatment discontinuation, or AEs leading to treatment interruption.

Serious adverse event/deaths/other significant events

As of 01 June 2020, no deaths have been reported on the lumasiran treatment.

Overall number of serious adverse events (SAEs) reported on lumasiran treatment was low.

In study 001, two patients had SAEs prior to Day 85 (3 mg/kg qM): one had an SAE of vomiting due to an upper ureteric stone that was treated with hydration and another patient had an SAE of nephrolithiasis that was treated with hyperhydration and lithotripsy. Two (2) additional patients had SAEs after Day 85, which included gastroenteritis in 1 patient (1 mg/kg qM) and abdominal pain, vomiting, and pyrexia in another patient (3 mg/kg qM).

In study 002 one patient developed an SAE of renal colic.

In study 003, 1 patient treated with 3 mg/kg q3M lumasiran experienced a craniocerebral injury and bone (rib) contusion as a result of a road traffic accident, and one had an SAE of urosepsis.

In study 004 in the ≥20 kg weight group one patient had an SAE of viral infection.

All events were mild or moderate in intensity and none of them was considered drug related.

Hepatic events

As lumasiran is directed to the liver, the frequency of hepatic events was evaluated by performing an analysis of AEs mapping to the Drug-related Hepatic Disorders SMQ.

During the placebo-controlled experience, no patients in either treatment group had AEs mapping to the Drug-related Hepatic Disorders SMQ.

In the overall pooled experience, 3 (4.0%) patients treated with lumasiran had AEs mapping to the Drug-related Hepatic Disorders SMQ. These include 1 patient with an AE of hepatomegaly and 1 patient with an AE of AST increased reported in the Extension Period of Study 003, and 1 patient with an AE of blood bilirubin (indirect bilirubin) increased reported in Study 002. The first two events were considered related to lumasiran by the investigators. All 3 events were non-serious, mild in severity, and did not result in any change to dose or withdrawal from the study. All 3 patients continue to receive lumasiran. No hepatic AEs have been reported in Study 004.

Renal Events

As patients with PH1 are at risk of recurrent kidney and bladder stones, the frequency of renal and urinary disorders other than renal stone events was evaluated.

During the placebo-controlled experience, 2 (7.7%) patients in the lumasiran group and no patients in the placebo group had AEs within the Renal and Urinary Disorders System Organ Class (SOC). Both AEs, polyuria and worsening renal pain, were considered not related to study treatment by the Investigator. Neither event led to a change in study drug administration.

In the overall pooled experience, no additional patients had renal AEs.

Laboratory findings

No apparent changes were observed in haematology, serum chemistry parameters, vital signs, ECG, etc.

Liver function tests and eGFR data were the only laboratory data pooled for analysis in the overall pooled experience.

On laboratory analysis, there have been no major changes in LFT parameters related to lumasiran treatment and no apparent pattern of imbalances compared to placebo in the placebo-controlled setting.

Pooled analysis showed mostly mild and transient elevations in ALT, AST, total bilirubin and ALP in 10.7%, 4%, 8%, and 4% (worst post-baseline values) in the lumasiran treated, respectively. These frequencies were in the same range as those observed in placebo-controlled experience. No cases of Hy's Law were reported.

Mean and median values for eGFR appeared stable over time in the placebo-controlled experience. No apparent changes were detected in the parameter across various studies.

Safety in special populations

The maximum age of the tested patients was 60 years.

Increased systemic levels of lumasiran were observed in the patients younger than 6 years of age. Limited safety information in these patients do not raise any age-specific safety concerns. However, limited safety data is available in the youngest (< 1 year) population, caution is warranted when using lumasiran in this population.

A dedicated renal impairment study was not conducted for the lumasiran clinical development program based on the minor (<26%) role of the kidneys in the excretion of lumasiran. Approximately half of the patients treated in the overall pooled experience had mild (37.3%) or moderate (17.3%) renal impairment at baseline.

An analysis of AEs was performed based on baseline eGFR values (≥90 mL/min/1.73m^2 [normal renal function], 60 to <90 mL/min/1.73m^2 [mild renal impairment], or 30 to <60 mL/min/1.73m^2 [moderate renal impairment]) and did not reveal any apparent differences in the safety profile between these groups.

Preliminary data in 4 patients with ESRD and on dialysis did not reveal any apparent differences in AE profile compared to the overall treated population. Data on plasma glycolate as measured prior to dialysis showed relative increase of glycolate on treatment with lumasiran similar to that in patients with more preserved or normal renal function (see **Table 19** in section 2.4.3). However, absolute values of glycolate at baseline and on treatment in these patients was several times higher, indicating that safety monitoring is warranted in the patients with severe and ESRD (with or without dialysis), as increased glycolate may cause, or worsen metabolic acidosis in these patients.

No data are available in patients with hepatic impairment. Given the decreased expression of ASGPR in these patients, reduced uptake of lumasiran is possible (see sections 2.4.2 and 2.4.4). Decreased lumasiran uptake in the liver and increased systemic exposure may be the consequence. While increased systemic lumasiran has not been linked to adverse events thus far and do not raise safety concerns at this point in time, decreased efficacy due to the reduced lumasiran uptake in the liver cannot be excluded and requires monitoring.

Influence of intrinsic or extrinsic factors on the lumasiran safety cannot be adequately assessed based on the subgroup analyses provided due to small sample sizes, differences in dosing schedules and treatment duration. Due to the lack of data, lumasiran should not be used in pregnant or lactating women.

Immunological events

In the pivotal Study 003 only 1 lumasiran-treated patient (including those initially randomised to placebo who received lumasiran during the Extension Period) had a post-dose ADA sample during the study that tested positive for ADA. This patient tested positive for ADA at Month 6 with a low titer of 1:50. The presence of low-level ADA in this patient did not affect safety, as no AEs have been reported in this patient.

Overall, across all 4 clinical studies in the lumasiran development program, including the patients with PH1 and healthy volunteers, a total of 6 of 100 (6%) lumasiran-treated individuals with post-dose ADA samples had treatment emergent ADA to lumasiran.

ADA titers were low (1:50) and transient and ADAs do not seem to influence PK/PD or safety of lumasiran.

Safety related to drug-drug interactions and other interactions

No DDI studies have been conducted and safety related to DDI interactions in other clinical studies have not been studied.

Discontinuation due to adverse events

In total, only two lumasiran-treated patients discontinued treatment due to the AEs.

The AEs leading to discontinuation were fatigue and disturbance in attention in one case. Another case concerned a 7-year old, who developed a needle phobia because of venepuncture and the guardian decided to withdraw the child.

None of these events was considered drug related.

Post marketing experience

Not applicable.

2.6.1. Discussion on clinical safety

Safety assessment of lumasiran is primarily based on the placebo-controlled setting of one pivotal study and the pooled analysis of the selected 3 studies (the pivotal study and 2 supportive studies). The safety database is small. This is acceptable given the ultra-orphan status of the disease and the amount of data presented on 6-month treatment can be considered sufficient for assessment of safety in this rare condition with the pre-requisite that further data will be collected and submitted post-approval.

Long-term treatment experience (>1 year) with lumasiran is limited and will be collected from the studies included in the Risk Management Plan (RMP). Safety in the population above 65 years of age is missing. However, this is not seen as a major deficiency, given the typically young age of the targeted

population and since the PK, PD, and respectively, safety of lumasiran are predicted to be comparable in primary hyperoxaluria type 1 patients across age groups including the elderly.

Overall, the studied population is reflective of the target population with the exception of very young children <2 years of age. Use in this population is included in the RMP as missing information with additional data expected from the ongoing trials ALN-GO1-004 and 005 as well as the planned observational PASS.

Limited data with lumasiran in patients with severe/end phase renal impairment have been provided and the definition used in the clinical development program to diagnose hepatic impairment is not accepted. Therefore treatment with lumasiran in patients with hepatic impairment and end-stage renal impairment are considered as missing information in the RMP. These sub-populations will be further characterised from the ongoing trials ALN-GO1-005 and the planned observational study.

Increased levels of glycolate were observed in these patients with end-stage renal impairment compared to the overall treated population. Caution when using lumasiran and monitoring of signs of metabolic acidosis is warranted in the patients with severe/end-stage renal impairment. Decreased expression of ASGPRs is reported in the patients with hepatic impairment, that could potentially lead to limited liver uptake and reduced efficacy of lumasiran. Therefore, monitoring of efficacy in the patients with moderate and severe hepatic impairment is required.

The frequency of total adverse events and AE related to the study drug was higher on lumasiran compared to placebo (84.6% vs. 69.2% and 42.3% of patients with 33.3% AEs vs. 7.7% of patients with 5.6% AEs) in the pivotal study. Similar results were observed in the pooled data analysis, with 89.6% patients experiencing AE and 39% experiencing drug-related AE. Eight patients in the overall population (001-005 studies) had non-drug related SAEs of mild or moderate intensity and no fatal cases were registered on treatment with lumasiran.

Cross-study comparison of AE frequencies is difficult due to small sample sizes, variable patient populations, differences in dosing and especially in exposures. However, no apparent differences were spotted.

Adverse events that were most frequently reported on lumasiran were injection site reactions. Notably, these AEs were not reported on placebo treatment and some of the patients on placebo developed injection site reactions after they were switched to lumasiran treatment, that suggests clear causal relationship of these events to the active drug. Injection site reactions were generally mild, majority resolved within two days, and did not result in interruption or discontinuation of treatment. Some of the events persisted longer, for a period of up-to several months, but generally resolved without treatment, and did not lead to discontinuation of the treatment.

As the injection sites will be alternated, and the injections are to be administered quarterly, the observed events do not raise any safety concerns.

Another commonly reported AE with higher frequency on lumasiran (15.4 vs 7.7%) was abdominal pain. No apparent causal relationship could be established with lumasiran, but neither could this be excluded.

Both injection site reactions and abdominal pain are regarded as ADRs and are included in the product information of lumasiran.

Most of the AEs were mild in severity and none of the AEs in the pooled analysis were severe. Only two AEs reported during lumasiran treatment led to treatment discontinuation suggesting good tolerability to the drug.

As lumasiran is directed to liver, liver-related AEs and laboratory parameters were scrutinised. Generally, number of liver-related AEs was low (3 patients with one AE each, 4% in pooled data set) with no cases reported in the patients younger than 6 years. On laboratory analysis, there have been no major changes in LFT parameters related to lumasiran treatment and no pattern of imbalances compared to placebo in the placebo-controlled setting. No cases of Hy's Law were reported in the patients of any age.

These results do not raise any safety concerns regarding potential impact of lumasiran on liver function. However, in light of the limited data available, experience with other similar drugs (e.g. givosiran), and preclinical findings suggestive of possible off-target effects of lumasiran, hepatic effects are included in the RMP as a potential important risk to allow further characterisation of the liver-related effects of the drug in studies that are ongoing or being planned.

No major findings were reported as AEs related to kidneys or urinary tract, nor were any major changes observed in the renal function parameters. Data do not raise any concerns with regard to the potential negative effects on renal function.

No apparent changes were observed in haematology, serum chemistry parameters, vital signs, ECG, etc. Influence of intrinsic or extrinsic factors on the lumasiran safety cannot be assessed properly based on subgroup analyses due to small sample sizes, differences and exposure/treatment duration.

Due to the lack of data use of lumasiran in pregnant or lactating women is considered as missing information in the RMP. The planned observational study will collect and evaluate information on maternal, foetal and infant outcomes following exposure to lumasiran during pregnancy and breastfeeding

Low titers of ADAs were detected in some of the lumasiran-exposed patients, which appeared transient. No apparent impacts on safety, PK and PD were observed.

No DDI studies have been conducted, however this was considered acceptable considering physicochemical properties, mechanism of action, PK properties of lumasiran, and data from the *in vitro* studies which suggest that such interactions are unlikely.

In general, the safety profile of lumasiran is in accordance with the results from non-clinical studies and known class effects of siRNA molecules targeted to the liver. The limited safety database will be supplemented by information which will be collected through the ongoing clinical studies and the planned observational study.

2.6.2. Conclusions on the clinical safety

Despite the limited size of the safety database, due to the rarity of primary hyperoxaluria type 1, the overall safety profile of lumasiran is considered acceptable. The main safety concerns are injection site reactions, abdominal pain, and the increased risk of metabolic acidosis in patients with severe/end-stage renal impairment, due to the increased levels of glycolate, which are addressed adequately through appropriate routine risk minimisation measures. The potential effects of treatment on the liver will be further characterised through ongoing and planned post-authorisation studies.

2.7. Risk Management Plan

Safety concerns

Summary of safety concerns	
Important identified risks	None
Important potential risks	• Hepatic effects
Missing information	• Longer-term safety (>1 year) • Use in patients with hepatic impairment • Use in patients with severe renal impairment or ESRD, including patients on dialysis • Use in pregnant or lactating women and effects on pregnancy outcomes • Use in patients <2 years of age • Immunogenicity

Abbreviations: ESRD=end-stage renal disease

Pharmacovigilance plan

Ongoing and Planned Additional Pharmacovigilance Activities

Study number **Short name** **Status**	**Summary of objectives**	**Safety concerns addressed**	**Milestones** **(required by regulators)**	**Due dates**
Category 3 - Required additional pharmacovigilance activities				
ALN-GO1-002 Study 002 Phase 2, multicentre, open-label, single-arm, long-term extension study in patients who completed Study ALN-GO1-001 Part B Ongoing	To evaluate the long-term safety and clinical activity of lumasiran in patients with PH1	• Hepatic effects • Longer-term safety (>1 year) • Immunogenicity	Interim report	Interim study report: 13 FEB 2020
			Final report	Final study report: DEC 2023
ALN-GO1-003 (Study 003) Phase 3, randomised, double-blind, placebo-controlled study with an Extended Dosing Period Ongoing	To evaluate the long-term efficacy and safety of lumasiran in patients with PH1	• Hepatic effects • Longer-term safety (>1 year) • Immunogenicity	Interim report	Interim study report: 12 MAR 2020
			Final report	Final study report: AUG 2024

Study number **Short name** **Status**	**Summary of objectives**	**Safety concerns addressed**	**Milestones** **(required by regulators)**	**Due dates**
ALN-GO1-004 Study 004 A Phase 3, multicentre, single-arm study in infants and young children with PH1 Ongoing	To evaluate the efficacy, safety, PK, and PD of lumasiran in paediatric patients with PH1 <6 years of age	• Hepatic effects • Longer-term safety (>1 year) • Use in patients <2 years of age • Immunogenicity	Interim report	Interim study report: 12 MAR 2020
			Interim Report	Interim study report: DEC 2020
			Final report	Final study report: MAR 2025
ALN-GO1-005 Study 005 A Phase 3, multicentre, single-arm study in patients with PH1 Ongoing	To evaluate the efficacy, safety, PK, and PD of lumasiran in patients with PH1 of all ages who have advanced disease with or without haemodialysis	• Use in patients with severe renal impairment or ESRD, including patients on dialysis • Hepatic effects • Longer-term safety (>1 year) • Use in patients <2 years of age • Immunogenicity	Interim report	Interim study report: NOV 2021
			Final report	Final study report: JAN 2026
ALN-GO1-007 Observational PASS Planned	To characterise the long-term real-world safety of lumasiran in patients with PH1 To collect and evaluate information on maternal, fetal and infant outcomes following exposure to lumasiran during pregnancy and breastfeeding	• Hepatic effects • Longer-term safety (>1 year) • Use in patients with hepatic impairment • Use in patients with severe renal impairment or ESRD, including patients on dialysis • Use in pregnant or lactating women and effects on pregnancy outcomes • Use in patients <2 years of age • Immunogenicity	Protocol submission for review	Planned protocol submission: within 3 months after positive EC decision
			Final protocol	Planned final protocol submission date: Q2 2021
			Interim updates	Study progress reports will be provided with each PSUR. Interim analysis (as agreed)

Study number Short name Status	Summary of objectives	Safety concerns addressed	Milestones (required by regulators)	Due dates
			Final report	Final study report planned due date: *TBD*

Abbreviations: EC=European Commission; ESRD=end-stage renal disease; PASS=Post-authorisation safety study; PD=pharmacodynamics; PH1=primary hyperoxaluria type 1; PK=pharmacokinetics; PSUR=periodic safety update report; TBD=to be determined.

Risk minimisation measures

Safety concern	Routine risk minimization activities
Important identified risk: None	Not applicable
Important potential risk: Hepatic effects	Routine risk communication: • Not applicable Routine risk minimization activities recommending specific clinical measures to address the risk: • Not applicable Other routine risk minimization measures beyond the Product Information: • Legal status: Prescription-only medication
Missing information: Longer-term safety (>1 year)	Routine risk communication: • A summary of the safety profile of lumasiran in the clinical development program is provided in the Undesirable effects section (Section 4.8) of the SmPC.
Missing information: Use in patients with hepatic impairment	Routine risk communication: • Information on the absence of data in patients with hepatic impairment is included in the Posology and method of administration section (Section 4.2) and Pharmacokinetic properties section (Section 5.2) of the SmPC. • Information that caution is required when treating patients with moderate or severe hepatic impairment is included in the Posology and method of administration section (Section 4.2) of the SmPC. • In the Warnings and Precautions section (Section 4.4) of the SmPC it is also included that patients with moderate or severe hepatic impairment should be monitored for potential decreased efficacy.
Missing information: Use in patients with severe renal impairment or ESRD,	Routine risk communication: • Information on the limited data in patients with severe renal impairment, ESRD or patients on dialysis is included in the

Safety concern	Routine risk minimization activities
including patients on dialysis	Posology and method of administration section (Section 4.2) and Pharmacokinetic properties section (Section 5.2) of the SmPC. • The following information is also included in the Warnings and Precautions section (Section 4.4) of the SmPC: Treatment with lumasiran increases plasma glycolate levels, which may increase the risk of metabolic acidosis or worsening of pre-existing metabolic acidosis in patients with severe or end-stage renal disease. These patients should therefore be monitored for signs and symptoms of metabolic acidosis.
Missing information: Use in pregnant or lactating women and effects on pregnancy outcomes	Routine risk communication: • Information on the lack of clinical data in pregnant women and in lactating women is included in the Fertility, pregnancy and lactation section (Section 4.6) of the SmPC, with a cross-reference to nonclinical data on embryo-fetal development, lactation, and fertility in the Preclinical safety data section (Section 5.3) of the SmPC. Routine risk minimization activities recommending specific clinical measures to address the risk: • Advice is provided to evaluate the benefits and risks of treatment with lumasiran during pregnancy and breastfeeding for the mother and infant, and the mother's clinical need for lumasiran in the Fertility, pregnancy, and lactation section (Section 4.6) of the SmPC and Section 2 of the Package Leaflet.
Use in patients <2 years of age	Routine risk communication: • Information on safety profile in pediatric population is provided in Section 4.8 and Section 5.2 of the SmPC. Information on limited data in children younger than 1 year of age is included in the Posology section (Section 4.2) and the Pharmacokinetic properties section (Section 5.2) of the SmPC. Routine risk minimization activities recommending specific clinical measures to address the risk: • None
Immunogenicity	Routine risk communication: • Information on immunogenicity is provided in Section 4.8 of the SmPC. Routine risk minimization activities recommending specific clinical measures to address the risk: • None

Conclusion

The CHMP and PRAC considered that the risk management plan version 0.5 is acceptable.

2.8. Pharmacovigilance

Pharmacovigilance system

The CHMP considered that the pharmacovigilance system summary submitted by the applicant fulfils the requirements of Article 8(3) of Directive 2001/83/EC.

Periodic Safety Update Reports submission requirements

The requirements for submission of periodic safety update reports for this medicinal product are set out in the Annex II, Section C of the CHMP Opinion.

The new EURD list entry will use the EBD to determine the forthcoming Data Lock Points.

2.9. New Active Substance

The applicant compared the structure of lumasiran with active substances contained in authorised medicinal products in the European Union and declared that it is not a salt, ester, ether, isomer, mixture of isomers, complex or derivative of any of them.

The CHMP, based on the available data, considers lumasiran to be a new active substance as it is not a constituent of a medicinal product previously authorised within the European Union.

2.10. Product information

2.10.1. User consultation

The results of the user consultation with target patient groups on the package leaflet submitted by the applicant show that the package leaflet meets the criteria for readability as set out in the *Guideline on the readability of the label and package leaflet of medicinal products for human use.*

2.10.2. Additional monitoring

Pursuant to Article 23(1) of Regulation No (EU) 726/2004, Oxlumo (lumasiran) is included in the additional monitoring list as:

- It contains a new active substance which, on 1 January 2011, was not contained in any medicinal product authorised in the EU;

Therefore the summary of product characteristics and the package leaflet includes a statement that this medicinal product is subject to additional monitoring and that this will allow quick identification of new safety information. The statement is preceded by an inverted equilateral black triangle.

3. Benefit-Risk Balance

3.1. Therapeutic Context

3.1.1. Disease or condition

Oxlumo is proposed to treat primary hyperoxaluria type 1. This is an inherited metabolic disorder which leads to overproduction of oxalate in the liver, with a subsequent formation or renal concrements, nephrocalcinosis, deterioration of renal function, and, in late stages, organ oxalosis. The type I of the disease comprises about 80% of the total PH population, with the most severe forms occurring in this subgroup. There is considerable morbidity associated with the condition, already in early years of life, depending on the time of manifestation, which is variable. Data on mortality are not

exactly known, but it can be assumed that the condition is associated with increased mortality overall. The disease is very rare with an estimated prevalence of 0.05 in 10,000 people in the European Union.

Oxlumo treatment is aimed at reducing the production of oxalate in the liver, thus reducing plasma levels and urinary excretion of oxalate. It is assumed that the reduction of the oxalate burden will lead to a reduced frequency of the clinical events.

3.1.2. Available therapies and unmet medical need

Current therapy consists of basic measures such as hyperhydration and medicinal products to increase solubility of oxalate. This is frequently burdensome with especially young children not able to comply fully with the requirements. The only substance used in the condition, but currently not licensed, is pyridoxine (Vitamin B6) which, however, is able to reduce the oxalate burden only in a minority of patients. Depending on the deterioration of renal function, when ESRD has developed, patients will in later stages be treated with dialysis, which frequently has to be applied with a tight schedule. Renal transplantation is then also carried out, and patients have also been treated with (pre-emptive) combined renal and liver transplantation. All of the three latter procedures are of course burdened with increased morbidity and also mortality.

3.1.3. Main clinical studies

The applicant has conducted a small clinical programme owing to the rarity of the disease. The pivotal study submitted is study 003, which was randomised, placebo controlled, multi-national and of 6 months duration with an ongoing open-label extension period with a proposed duration of 54 months. The trial recruited 39 patients aged 6-60 which were randomised 2:1 to active and placebo treatment, respectively. Patients were having a genetically confirmed diagnosis of PH 1, and a mean 24-hour urinary oxalate excretion from the first 2 valid 24-hour urine collections being $\geq$0.70 mmol/24hr/1.73m^2. The dosing of the study medication was 3 mg/kg BW monthly for the first three months, and then 3 mg/kg BW every three months.

The applicant has also submitted a study in patients under the age of 6 (study 004), which is a non-randomised study with a proposed study duration of 60 months, and a primary evaluation for efficacy at 6 months. This study is reported with an interim report for the efficacy results.

Both studies taken together support the treatment of patients of all ages.

3.2. Favourable effects

The favourable effects of the compound are based on the surrogate parameter of reducing the oxalate burden in the body, with a reduction of oxalate in plasma and its excretion in urine. The evaluation of the urine excretion was based on 24-hour urine collections with the endpoints "24-hour urinary oxalate corrected for body surface area (BSA)", which was the primary endpoint (evaluated as percent change from baseline for the primary analysis, and as absolute change as secondary endpoint), "24-hour urinary oxalate:creatinine ratio" (to account for the dilution status of the urine; secondary endpoint), with both being evaluated as relative and absolute changes, and the change in plasma oxalate. Further secondary endpoints comprise the proportion of patients with a 24-hour urinary oxalate level at or below 1.5xULN, and at or below ULN at month 6.

In the pivotal study 003, all these parameters demonstrated statistically significant changes, with a reduction in24-hour urinary oxalate excretion of 65.4% (compared to 11.8 for placebo treated patients). In addition 52% and 84% of lumasiran treated patients achieved respectively normal or

near normal levels of 24 hour urinary oxalate corrected for BSA (≤1.5×ULN). In contrast no placebo treated patient had normal or near normal levels of urinary oxalate excretion at month 6. Of note, the secondary endpoints mentioned were included into a hierarchical testing strategy and are thus under full type-I error control.

These effects were highly consistent across all subgroups analysed (age, weight, sex, geographic region etc.).

The trial in young children evaluated only two of the secondary endpoints and over a shorter period. However, these also indicated favourable changes in this population similar to the magnitude seen in the adult population. Paediatric patients achieved a reduction of 72.0% in spot urinary oxalate: creatinine ratio from baseline (averaged over months 3 through month 6). Furthermore, nine patients achieved near normalisation (≤1.5×ULN), including 1 patient who achieved normalisation ((≤ULN), at month 6 in spot urinary oxalate: creatinine ratio. The 6 months treatment data provided for 18 patients was considered sufficient to draw conclusions on efficacy in this age group. The CHMP therefore concluded that the claim of treating patients of all age ranges was appropriately substantiated.

3.3. Uncertainties and limitations about favourable effects

The demonstration of efficacy, as described above, is currently based on surrogate parameters only. Although high biological plausibility can be attributed to the hypothesis underlying the use of the surrogates, the prognostic value of these, in the context of medical treatment of the condition, is not available, and therefore the surrogate parameters have to be considered to be non-validated. The applicant has tried to support the prognostic value of oxalate reduction with data on patients with liver transplantation and pyridoxine treatment, which indicate improvement of the condition in terms of clinical events (e.g. renal stones). However, these data are based on published literature case reports only.

The results of the trial have also included secondary and exploratory endpoints with the evaluation of the occurrence of renal stone events, the development of nephrocalcinosis (diagnosed by ultrasound), and changes in renal function as measured by eGFR. However, none of these parameters has indicated any improvement in the actively treated group as compared to the placebo group. The failure to demonstrate a clinical benefit is largely attributable to the short (controlled) observation period, as well as the small sample size. Post hoc analysis of 1-year data of the placebo - lumasiran vs. lumasiran – lumasiran group do indeed suggest some improvement in the renal events/ultrasound findings on active treatment. However, longer treatment and observation data are required to confirm this finding.

No efficacy data are available for patients above the age of 65, however, majority of PH1 patients are younger than 65. Further, PK and PD analyses across various ages suggest similar effects. Thus, extrapolation of effects to this population appears justified.

Data from the trial in the small children under the age of 6 is limited. This included only 2 children below the age of 1 and 4 patients were aged less 2 years. The treatment of patients in the very young population is therefore burdened with additional uncertainties. Further data in this population will be collected post-marketing.

3.4. Unfavourable effects

The majority of the patients reported at least 1 AE on lumasiran. The proportion of patients with AEs was higher on lumasiran treatment than with placebo.

Most frequently reported AEs (frequency ≥10%) were AEs grouped under the SMQ terms injection site reactions and abdominal pain. Injection site reactions were the most prominent AEs and were reported only on lumasiran treatment. Some of the AEs occurred after repeated treatment, on the day of drug administration, or as delayed reactions, whereby the delayed reaction could last several months. AEs related to abdominal pain were also reported more frequently on lumasiran than on placebo (15.4 vs 7.7%).

Only mild and moderate AEs were reported including the SAEs and none of the SAEs were considered as drug related.

Hepatic and renal effects are the acknowledged potential risks of siRNAs. AEs related to hepatic disorders and elevations in LFT were reported on lumasiran, however, without marked differences to the placebo group, and no apparent changes in the renal parameter were observed.

3.5. Uncertainties and limitations about unfavourable effects

Limitations of the safety database include small sized, variable population, and limited and variable exposure across trials. Patients across the broad age range were treated with different dosing schedules and for variable treatment duration, mostly for up-to 12 months. Data on long-term safety is limited to a small number of > 6 years old patients with <1 year exposure. Such variability combined with small sample size and short observation times creates uncertainties in interpretation of safety profile.

For the youngest, most vulnerable (i.e. <2 years old) population and in the patients with severe/end-stage renal impairment, data are very limited. Experience of treatment with lumasiran in patients with hepatic impairment (of any degree) is absent.

3.6. Effects Table

Table 41. Effects Table for Oxlumo for the treatment of primary hyperoxaluria type 1 (PH1) in all age groups

Effect	Short Description	Unit	Lumasiran	Placebo	Uncertainties/ Strength of evidence	References
Favourable Effects						
Urinary oxalate reduction	% change in 24h UOx corrected for BSA	%	-65.4	-11.8	Strong evidence, highly statistically significant. All other urinary and plasma related evaluations were in full support	Study 003
Renal stone events	Number	n	5	2	Weak evidence, high uncertainty due to paucity of events. Trend for improvement in favour of lumasiran from longer term data	Study 003

Effect	Short Description	Unit	Lumasiran	Placebo	Uncertainties/ Strength of evidence	References
Spot urinary oxylate:creatinine ratio in children	Percent change in average of Month 3 - 6 vs. baseline	LS Means (SEM) %	-72 (2.7)	N/A	Pronounced effect, similar to the effects in the controlled setting in adults	Study 004
Unfavourable Effects						
Injection site reactions	Percent of patients with at least one ISR	%	33.3	0		Study 003
Abdominal pain	Percent of patients with at least one AE	%	15.4	7.7	The event may be associated with background disease.	Study 003

Abbreviations: UOx – urinary oxalate; BSA-Body surface area; LS-Means-Least Square Means; SEM-Standard error of the mean; ISR-Injection site reaction; AE-Adverse event.

3.7. Benefit-risk assessment and discussion

3.7.1. Importance of favourable and unfavourable effects

With regard to oxalate reduction, the near normalisation or full normalisation of the urinary oxalate excretion is seen as the most clinically relevant parameter. Although still considered a surrogate, the biological plausibility that normalisation of the oxalate burden would lead to a reduced clinical manifestation of the disease appears convincing.

The reduction of oxalate as demonstrated in the study 003 is a strong effect induced by the treatment, which has shown consistency across different disease severities, and in different patient subgroups (according to age, different regions etc). A clinical benefit, e.g. with regard to renal events, has not been demonstrated, which is likely due to the short placebo-controlled treatment phase of 6 months and the small sample size in this orphan indication. In fact, renal stone events were even occurring at a numerically slightly higher rate in actively treated patients but the ultrasound-based evaluations of the progression of nephrocalcinosis (in those patients with pre-existing nephrocalcinosis) showed some (numerical) improvements compared to placebo. However, the assumption that in patients with near normal levels of oxalate excretion (achieved by 84%), or even full normalisation of oxalate excretion (achieved by 52%), a further disease progression with regard to deterioration of renal function, development of nephrocalcinosis, renal stone events, and finally ESRD can be halted or ameliorated appears reasonable. Also, the data of post hoc analysis of renal events over 1 year, which showed trends towards improvement in these events on lumasiran treatment are supportive in this respect.

Adverse events were more frequently reported in the lumasiran treatment group compared to placebo. However, most of the adverse events were mild.

The compound, being an RNA-based injectable, is "naturally" bound to cause fear of injection, and injection pain, which is considered especially relevant for the intended population, mainly being in the paediatric age range. However, high level of compliance and low number of drop outs from the studies, are reassuring regarding compliance in clinical reality. In addition, about 33% of the patients

experienced injection site reactions. The relevance of the injection site reactions is alleviated with the schedule of administration, which is one injection every three months only, and alternating sites of administration. The evaluation of adverse events has also revealed that abdominal pain occurs relatively frequently in connection with the injection. Although no causal relationship to lumasiran could be established, these AEs were qualified as ADRs of lumasiran. The severity of the abdominal pain was mild in most cases, duration transient, and the impact on the further treatment appears to be low.

The potential for ADA formation appears to be low, and there is currently no indication that ADA formation would influence efficacy or safety of the compound.

Hepatic events are an important potential risk and may be a class effect of liver-targeting siRNA products. Events suggestive of lumasiran effects on liver were infrequent. Further evaluation of the possible causal relationship and of clinical relevance of these events is required. No signs of possible negative impact on renal function could be detected based on the available data set.

3.7.2. Balance of benefits and risks

The clinical studies performed have demonstrated strong effects with regard to reduction of oxalate production in the body, measured as the oxalate "load" in plasma and urine, and have shown that about half of the patients can achieve normal levels of oxalate, and more than 80% can achieve near normal levels. Due to the strong biological plausibility, the expectation that the favourable effects demonstrated on the surrogate parameters can translate into a reduced progression of the disease is fully supported. The expected reduction of clinical events, and prevention of ESRD is considered to be of high clinical relevance.

The observed adverse reactions were generally mild in nature and the most commonly reported ADR of injection site reaction relates to the mode of administration of lumasiran. Although these reactions were relatively frequent, the schedule of administration is suitable to alleviate the burden to patients.

Overall, the observed favourable effects are considered to outweigh the unfavourable effects.

3.8. Conclusions

The overall B/R of Oxlumo is positive.

4. Recommendations

Outcome

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considers by consensus that the benefit-risk balance of Oxlumo is favourable in the following indication:

treatment of primary hyperoxaluria type 1 (PH1) in all age groups.

The CHMP therefore recommends the granting of the marketing authorisation subject to the following conditions:

Conditions or restrictions regarding supply and use

Medicinal product subject to restricted medical prescription (see Annex I: Summary of Product

Characteristics, section 4.2).

Other conditions and requirements of the marketing authorisation

Periodic Safety Update Reports

The requirements for submission of periodic safety update reports for this medicinal product are set out in the list of Union reference dates (EURD list) provided for under Article 107c(7) of Directive 2001/83/EC and any subsequent updates published on the European medicines web-portal.

The marketing authorisation holder shall submit the first periodic safety update report for this product within 6 months following authorisation.

Conditions or restrictions with regard to the safe and effective use of the medicinal product

Risk Management Plan (RMP)

The MAH shall perform the required pharmacovigilance activities and interventions detailed in the agreed RMP presented in Module 1.8.2 of the marketing authorisation and any agreed subsequent updates of the RMP.

An updated RMP should be submitted:

- At the request of the European Medicines Agency;
- Whenever the risk management system is modified, especially as the result of new information being received that may lead to a significant change to the benefit/risk profile or as the result of an important (pharmacovigilance or risk minimisation) milestone being reached.

New Active Substance Status

Based on the CHMP review of the available data, the CHMP considers that lumasiran is a new active substance as it is not a constituent of a medicinal product previously authorised within the European Union.