IONIS PHARMACEUTICALS, INC.

ISIS 396443-CS3A

A Study to Assess the Efficacy, Safety, Tolerability, and Pharmacokinetics of Multiple Doses of Nusinersen (ISIS 396443) Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Protocol Amendment 6 – 25 January 2016

Sponsor:

Ionis Pharmaceuticals, Inc.

2855 Gazelle Court

Carlsbad, CA 92010

ISIS 396443-CS3A

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Protocol History:

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Sponsor:

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PPD

ISIS 396443

Ionis Protocol Number ISIS 396443-CS3A

Protocol Amendment 6

Clinical Phase: 2

A Study to Assess the Efficacy, Safety, Tolerability, and Pharmacokinetics of Multiple Doses of Nusinersen (ISIS 396443) Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

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Date: 25 January 2016

Confidentiality Statement

Protocol Signature Page

Protocol Number: ISIS 396443-CS3A

Protocol Title: A Study to Assess the Efficacy, Safety, Tolerability, and Pharmacokinetics of Multiple Doses of Nusinersen (ISIS 396443) Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment: Amendment 6

Date: 25 January 2016

I hereby acknowledge that I have read and understand the attached clinical protocol, entitled "A Study to Assess the Efficacy, Safety, Tolerability, and Pharmacokinetics of Multiple Doses of Nusinersen (ISIS 396443) Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy", dated 25 January 2016, and agree to conduct the study as described herein.

I agree to comply with the International Conference on Harmonization Tripartite Guideline on Good Clinical Practice (E6).

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of Ionis Pharmaceuticals, Inc.

Investigator's Signature

Investigator's Name *(please print)*

Date (DD Month YYYY)

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PROTOCOL AMENDMENT

Protocol Number: ISIS 396443-CS3A

Protocol Title: A Study to Assess the Efficacy, Safety, Tolerability, and Pharmacokinetics of Multiple Doses of Nusinersen (ISIS 396443) Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment: 6

Amendment Date: 25 January 2016

The following modifications to Protocol ISIS 396443-CS3A, have been made.

Minor changes (not included in the list of changes below) have been made throughout the protocol to correct errors and/or to improve the overall clarity of the original protocol but these changes do not impact subject safety, exposure, or the overall study design.

If there are more than 1 or 2 changes, insert the following table.

The following table provides a summary list of major changes to the protocol:

Protocol Section	Description of Change	Rationale
Study Title	Added “Efficacy” and “Nusinersen”	Study title was updated to appropriately reflect the study objectives. Nusinersen is an approved USAN name for ISIS 396443.
Protocol Synopsis: Objectives, Section 1: Objectives	Amended Study Objectives to reflect efficacy assessment as the primary study objective and safety and tolerability as the secondary study objectives.	Study is of sufficient duration now to assess the efficacy of nusinersen in this patient population.
Protocol Synopsis: Criteria for Evaluation Section 10.1: Study Endpoints	Efficacy and Safety endpoints are listed according to the amended study objectives	Efficacy endpoints related to motor function and event-free survival represent the most meaningful endpoints in this patient population.
Section 10.5: Planned Methods of Analysis	Inserted the methods for analysis of efficacy endpoints of the study	Description of the methodology for analysis of the efficacy endpoints related to motor function and event-free survival was included to clarify how these data will be analyzed and presented.

PROTOCOL SYNOPSIS

Protocol Title	A Study to Assess the Efficacy, Safety, Tolerability, and Pharmacokinetics of Multiple Doses of Nusinersen (ISIS 396443) Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy
Study Phase	2
Indication	Spinal Muscular Atrophy (SMA)
Objectives	***Primary Objective:*** To examine the clinical efficacy of multiple doses of ISIS 396443 administered intrathecally to patients with Infantile-Onset SMA. ***Secondary Objectives:*** To examine the safety and tolerability of multiple doses of ISIS 396443 administered intrathecally to patients with Infantile-Onset SMA. To examine the CSF and plasma pharmacokinetics of multiple doses of ISIS 396443 administered intrathecally to patients with Infantile-Onset SMA.
Number of Subjects	Approximately 8-20 subjects may be enrolled into this multiple-dose study of ISIS 396443. The number of subjects may be higher if some subjects must be replaced and/or if the sizes of cohorts are expanded in order to obtain further experience with one or both dose levels.
Study Design and Methodology	This study will test the clinical efficacy, safety, tolerability, and pharmacokinetics of multiple doses of ISIS 396443 administered as intrathecal (IT) injections by lumbar puncture (LP). All subjects will receive ISIS 396443. Two 'loading' dose levels (scaled by infant age to be equivalent to 2 year old doses of 6 mg or 12 mg, based on CSF volume) will be evaluated sequentially. The initial dose level of 6 mg will be studied in a cohort of 4 subjects. The 12 mg dose level will be studied in 4 to approximately 16 subjects. Following this, all subjects will receive 'maintenance' doses of 12 mg equivalent ISIS 396443 on Days 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261. Cohort 1 (n = 4): 'Loading' dosing of 6 mg equivalent ISIS 396443 on Days 1, 15, 85, IT injection; followed by 'maintenance' doses of 12 mg equivalent ISIS 396443 on Days 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261 IT injection. Cohort 2 (n = 4-16): 'Loading' dosing of 12 mg equivalent ISIS 396443 on Days 1, 15, 85, IT injection; followed by 'maintenance' doses of 12 mg equivalent ISIS 396443 on Days 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261 IT injection. After Informed consent is obtained, subjects will undergo a Screening evaluation no greater than 21 days prior to first dose administration at which their eligibility for the study will be examined. Subjects who meet the eligibility criteria will be admitted to the study center on Study Day 1, undergo pre-dose evaluations, and then receive an LP injection of Study Drug (ISIS 396443). Subjects will return to the study center on Days 15, 85, 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261 for follow-up evaluations of clinical efficacy and safety and subsequent injections. Following the LP injection on Day 1, subjects will remain at the study center for at least 24 hours post-injection for safety monitoring. Following LP injections on Days 15, 85, 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261, subjects will remain at the study center for at least 6 hours post-injection for safety monitoring. Safety monitoring visits will occur on Days 16, 29, 86, 92, 169, 254, 337, 442, 568, 694, 820, 946, 1072, 1198, and 1352 (through 13 weeks after the last dose of ISIS 396443). In addition, the study center will monitor the subject's condition through telephone contact on Study Days 8, 43, 57, 71, 106, 127, 134, 148, 189, 197, 218, 239, 274, 295, 316, 358, 380, 400, 421, 463, 484, 506, 526, 547, 589, 610, 632, 652, 673, 715, 736, 758, 778, 799, 841, 862, 884, 904, 925, 967, 988, 1010, 1030, 1051, 1093, 1114, 1136, 1156, 1177, 1219, 1240, 1262, 1282, 1303, and 1324 (i.e., approximately every 3 weeks). A CSF sample will be taken pre-dose on each injection day for safety laboratory evaluation and PK analyses. If a subject terminates early from the study, they will be encouraged to complete assessments per the Day 1352 visit.

Protocol Synopsis *Continued*

Study Population and Main Criteria for Inclusion/ Exclusion	**Inclusion Criteria:** Subjects must meet all of the following criteria at screening to be eligible: 1. Signed informed consent of parent(s) or guardian(s) 2. Genetic documentation of 5q SMA homozygous gene deletion or mutation 3. Onset of clinical signs and symptoms consistent with SMA at ≥ 21 days and ≤ 6 months (180 days) of age 4. Males and females between ≥ 21 days and ≤ 7 months (210 days) of age at Screening 5. At study entry, receiving adequate nutrition and hydration (with or without gastrostomy), in the opinion of the Site Investigator 6. Body weight > 5^{th} percentile for age using CDC guidelines 7. Medical care meets and is expected to continue to meet guidelines set out in the Consensus Statement for Standard of Care in SMA (Wang et al. 2007), in the opinion of the Site Investigator 8. Gestational age of 35 to 42 weeks and gestation body weight ≥ 2 kg 9. Reside within approximately 9 hours ground-travel distance from a participating study center for the duration of the study. Residence > 2 hours ground-travel distance from a study center must obtain clearance from the Site Investigator and the study Medical Monitor 10. Able to complete all study procedures, measurements and visits and parent or guardian/subject has adequately supportive psychosocial circumstances, in the opinion of the Site Investigator **Exclusion Criteria:** Subjects meeting any of the following criteria are not eligible for the study: 1. Hypoxemia (O_2 saturation awake < 96% or O_2 saturation asleep < 96%, without ventilation support) 2. Presence of an untreated or inadequately treated active infection requiring systemic antiviral or antimicrobial therapy at any time during the screening period 3. History of brain or spinal cord disease that would interfere with the LP procedures, CSF circulation, or safety assessments 4. Presence of an implanted shunt for the drainage of CSF or an implanted CNS catheter 5. History of bacterial meningitis 6. Clinically significant abnormalities in hematology or clinical chemistry parameters, as assessed by the Site Investigator, at Screening that would render the subject unsuitable for inclusion 7. Treatment with another investigational drug (e.g., albuterol, riluzole, carnitine, creatine, sodium phenylbutyrate, salbutamol, valproate, hydroxyurea etc.), biological agent, or device within 90 days prior to enrollment or anytime during the study. Any history of gene therapy or cell transplantation 8. The subject's parent(s) or legal guardian(s) is unable to understand the nature, scope, and possible consequences of the study, or does not agree to comply with the protocol defined schedule of assessments 9. Ongoing medical condition that according to the Site Investigator would interfere with the conduct and assessments of the study. Examples are medical disability other than SMA that would interfere with the assessment of safety or would compromise the ability of the subject to undergo study procedures

Protocol Synopsis *Continued*

Study Drug and Administration	ISIS 396443 (20 mg/mL, 2.5 mL, diluted to specified concentration with artificial CSF diluent OR 2.4 mg/mL as 'Ready-to-Use Vial'; see Section 7.1.2. for more details) will be administered as an IT LP injection. The volume of the injection and thus the dose will be adjusted for the subject's age, based on scaling to CSF volume. Thus, younger subjects will be given a lower dose of drug, achieved by injecting a smaller volume that is proportional to estimated CSF volume for age, such that dose volume will be equivalent to 5 mL for age 2 to adult. Dosing instructions and details regarding administration will be provided in the Study Drug Manual.
Criteria for Evaluation	**Efficacy Evaluations:** • Achievement of motor milestones as evaluated by the Hammersmith Infant Neurological Examination (Module 2) • Event-free survival determined by the proportion of subjects who are alive and do not require permanent ventilatory support (defined as ≥ 16 hours ventilation/day continuously for at least 14 days in the absence of an acute reversible illness) • Improvement in muscle strength as measured by the Children's Hospital of Philadelphia Infant Test for Neuromuscular Disease (CHOP INTEND) • Improvement in neuromuscular electrophysiology measured by the Compound Muscle Action Potential (CMAP) of the ulnar and peroneal nerves • Measures of respiratory status (number of respiratory events, respiratory-related hospitalizations, ventilator use) • Growth parameters (weight for age/length, head circumference, chest circumference, head to chest circumference ratio, arm circumference) **Safety/Tolerability Endpoints:** • Adverse events • Vital signs (HR, BP, respiratory rate, pulse oximetry awake) • Neurological examinations • Physical examinations and weight • Clinical laboratory tests • CSF laboratory tests • ECGs • Use of concomitant medications **Pharmacokinetic Measures:** • Plasma levels of ISIS 396443 • CSF levels of ISIS 396443 **Additional Measures:** • SMN2 copy number (if no Athena result existing) • SMN2 gene sequencing (if no Athena result existing)

Protocol Synopsis *Continued*

Safety Monitoring and Dose Escalation	Safety data will be reviewed on an ongoing basis by the Medical Monitor and by the DSMB. At the onset of the study, a Dose-Limiting-Toxicity (DLT) is defined as an adverse event that in the judgment of a Site Investigator is of sufficient significance to be dose limiting, is possibly or definitely related to Study Drug (i.e., the adverse event is substantially less likely to occur in patients not treated with Study Drug), and that is not a known: 1) sign or symptom of SMA disease, or 2) effect of the LP injection procedure, or 3) effect of anesthesia/sedation in SMA patients, if utilized. The progression of the study from the initial dose level (Cohort 1) to Cohort 2 will be determined by the Sponsor and the DSMB and generally be based on the number of DLTs observed in patients treated with ISIS 396443. In general, the occurrence of DLTs in 2 patients in Cohort 1 may result in the dose tested being considered as dose limiting. The occurrence of 1 DLT in 4 subjects in Cohort 1 may result in the cohort being expanded to 6 subjects (2 additional subjects) to further assess the safety within that dose level. After the last patient within Cohort 1 completes the Day 29 visit, safety results for the cohort will be reviewed by the Medical Monitor and the DSMB and a recommendation regarding further enrollment and escalation to Cohort 2 will be made. The Sponsor and DSMB will determine if enrollment of additional patients is required to confirm there is an acceptable toxicity profile prior to its designation as MTD.
Statistical Considerations	The sample size selection is based on prior experience to ensure that the safety, tolerability, and pharmacokinetics will be adequately assessed while minimizing unnecessary patient exposure rather than statistical considerations for the efficacy endpoints. Interim efficacy and safety analyses will be performed to provide content for regulatory submissions and to support ISIS 396443 drug development planning and business activities. Details of the analyses are contained in the Statistical Analysis Plan.
Sponsor	Ionis Pharmaceuticals, Inc.

STUDY GLOSSARY

Abbreviation/Acronym	Definition
AE	Adverse event/experience
ALT	Alanine aminotransferase
aPTT	Activated partial thromboplastin time
ASO	Antisense oligonucleotide
AST	Aspartate aminotransferase
BUN	Blood urea nitrogen
CMAP	Compound muscle action potential
C_{max}	Maximum concentration
CNS	Central nervous system
CPK	Creatinine phosphokinase
CRF	Case report form
CSF	Cerebrospinal fluid
CT	Computerized tomography
DLT	Dose limiting toxicity
DSMB	Data and Safety Monitoring Board
eCRF	Electronic case report form
ECG	Electrocardiogram
FL	Full-length
GCP	Good Clinical Practice
HFMSE	Hammersmith Functional Motor Scale Expanded
hnRNP	Heterogeneous nuclear ribonucleoproteins
ICH	International Conference on Harmonisation
INR	International normalized ratio
IRB	Institutional Review Board
IT	Intrathecal
LP	Lumbar puncture
MAD	Multiple ascending-dose
MedDRA	Medical Dictionary for Regulatory Activities
miRNA	Micro ribonucleic acid (RNA)
MOE	2′-*O*-(2-methoxyethyl)

MRI	Magnetic resonance imaging
mRNA	Messenger ribonucleic acid
MTD	Maximum tolerated dose
NCS	Not clinically significant
NOAEL	No observed adverse effect level
PK	Pharmacokinetic(s)
PT	Prothrombin Time
SAE	Serious adverse event
SMA	Spinal muscular atrophy
SMN	Survival motor neuron
snRNA	Small nuclear ribonucleic acid
SUSARs	Suspected unexpected serious adverse reactions
$t_{½}$	Terminal elimination half-life
TIMPSI	Test for Infant Motor Performance Screening Items
T_{max}	Time to maximal concentration
WMA	World Medical Association

1. OBJECTIVES

The objectives of this study are to evaulate the clinical efficacy, safety, tolerability, and pharmacokinetics (PK) of multiple doses of ISIS 396443 administered intrathecally to patients with infantile-onset Spinal Muscular Atrophy (SMA).

1.1 Primary Objective

To examine the clinical efficacy of multiple doses of ISIS 396443 administered intrathecally to patients with Infantile-Onset SMA.

1.2 Secondary Objectives

To examine the safety and tolerability of multiple doses of ISIS 396443 administered intrathecally to patients with infantile-onset SMA.

To examine the cerebral spinal fluid (CSF) and plasma PK of multiple doses of ISIS 396443 administered intrathecally to patients with infantile-onset SMA.

2. BACKGROUND AND RATIONALE

2.1 Spinal Muscular Atrophy

SMA is an autosomal recessive neuromuscular disease characterized by degeneration of the motor neurons in the anterior horn of the spinal cord, resulting in atrophy of the voluntary muscles of the limbs and trunk. With an incidence of 1:6000 to 1:10,000 live births, it is the most common genetic cause of infant mortality, and a major cause of childhood morbidity due to weakness, in the U.S. The natural history of SMA includes four major phenotypes that are recognized dependent on age of onset and achieved motor abilities. The most severe form, Type I SMA, has a disease onset within the first few months of life; these children are never able to sit or walk and usually die from respiratory failure by the age of 2 years. Type II SMA patients are able to sit but never walk unaided, with symptoms presenting between 6-18 months of age. Type III SMA patients are able to sit and walk but individuals with this form may become severely and increasingly disabled. Type IV or adult-onset SMA patients have an age of onset over 18 years of age and have normal life expectancies.

In 95% of SMA patients, a deletion in the *SMN1* gene on Chromosome 5q11-q13 is found; with the remaining 5% attributable to small mutations in the same gene (Lefebvre et al. 1995; Helmken et al. 2003). *SMN1* lies in the telomeric portion of an inverted duplication of a region of Chromosome 5. The centromeric half of the duplication contains a homologous gene, named *SMN2* that differs from *SMN1* by 11 nucleotides. The open reading frames for both genes encode for proteins with identical amino acid sequences. Survival motor neuron (SMN) gene transcripts, similar to most mammalian transcripts, undergo alternative splicing in which certain exons are either included or excluded from the mature protein coding transcripts (Keren et al. 2010). In particular, Exon 7 of the *SMN1* gene is alternatively spliced with 90 to 95% of the mature messenger ribonucleic acid (mRNA) transcripts derived from the *SMN1* gene containing Exon 7, and 5 to 10% of transcripts missing Exon 7. The transcripts missing Exon 7 (often referred to as Δ7) produce a truncated protein which is defective and unstable (Cho et al. 2010). One (1) of the 11 nucleotide differences between *SMN1* and *SMN2*, a C to T substitution occurs in Exon 7 of the *SMN2* gene resulting in an alternative splicing pattern that

favors skipping of Exon 7. The result is that as much as 90% of the transcripts produced from *SMN2* are missing Exon 7. The remainder, SMN2 transcripts containing Exon 7, produces a full-length (FL) protein product identical to the SMN1 protein, since the C to T substitution is silent. Humans have a variable copy number of the *SMN2* gene (0-8 copies). The number of *SMN2* copies and the resulting amount of FL-SMN protein expressed in SMA patients (10-40% of normal SMN protein levels) correlates with SMA disease severity and thus *SMN2* is a key modifier of disease phenotype (Coovert et al. 1997; Lefebvre et al. 1997; Feldkotter et al. 2002; Prior et al. 2004).

2.2 Therapeutic Rationale

Since the number of *SMN2* gene copies and resulting amount of SMN protein is correlated with disease onset and severity, a therapeutic approach predicted to benefit SMA patients is to increase the levels of full length SMN2 pre-mRNA by restoring the splicing pattern that gives rise to full length SMN2 mRNA. Increasing inclusion of Exon 7 in the SMN2 transcript will increase FL-SMN protein levels and SMN protein activity. A therapeutic strategy for promoting Exon 7 inclusion is through the use of antisense oligonucleotides (ASOs) (see Figure 1).

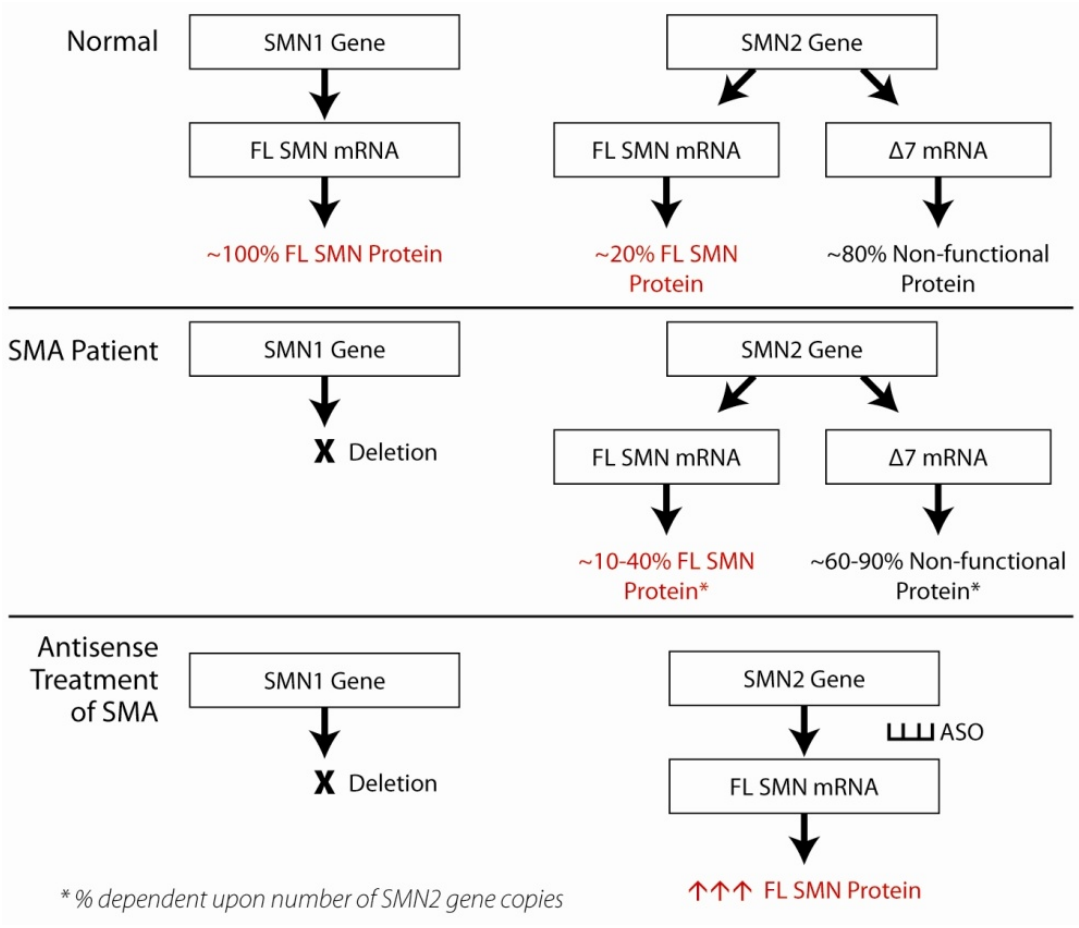

Figure 1 ASO Therapeutic Approach for Treatment of SMA

The known potential risks associated with ISIS 396443 are elaborated on in the Guidance to Investigator section of the Investigator's Brochure. Additional study associated risks relate to the lumbar puncture (LP) procedure are also elaborated on in the Guidance to Investigator section of the Investigator's Brochure.

2.3 ISIS 396443

2.3.1 Mechanism of Action

ISIS 396443 is a fully modified, 2′-*O*-2-methoxyethyl (MOE), ASO drug designed to bind to a specific sequence in the intron downstream of Exon 7 of the SMN2 transcript. The region of the pre-mRNA targeted by ISIS 396443 is normally occupied by heterogeneous nuclear ribonucleoproteins (hnRNP) A1/2 proteins, masking the U1 small nuclear ribonucleic acid (snRNA) binding site at the 5′-exon-intron junction of Exon 7, and is referred to as ISS-N1. U1 snRNA base pairs to the sequences that define the 5′-splice site, which is thought to be one of the first steps that initiates splicing of an intron. ISIS 396443 displaces the hnRNP A1/2 proteins from the pre-mRNA binding site, allowing U1 snRNA to bind to the exon-intron junction and promote assembly of the spliceosomal complex, thus promoting inclusion of Exon 7 into the mRNA which results in production of FL-SMN protein.

2.3.2 Chemistry

Chemically, ISIS 396443 is a synthetic oligomer of 18 nucleotides (i.e., an 18-mer) that are connected sequentially by phosphorothioate linkages. Each of the 17 internucleotide linkages is a 3′-*O* to 5′-*O* phosphorothioate diester. The 18 sugar residues are uniformly modified with 2′-*O*-(2-methoxyethyl) (MOE). These MOE-modified nucleotides confer (1) increased affinity to the target mRNA (McKay et al. 1999), (2) increased resistance to exonucleases and endonucleases (thereby increasing stability in tissue) (Geary et al. 2003), and (3) amelioration of some of the high dose toxicities associated with ASO containing only the phosphorothioate linkages (Henry et al. 2000).

The sequence of ISIS 396443 is written as follows:

5′-^{Me}U^{Me}CA^{Me}C^{Me}U^{Me}U^{Me}U^{Me}CA^{Me}UAA^{Me}UG^{Me}C^{Me}UGG- 3′

Where A and G are 2′-*O*-(2-methoxyethyl)nucleosides, ^{Me}C is 5-methyl-2′-*O*-(2-methoxyethyl)cytidine and ^{Me}U designates 5-methyl-2′-*O*-(2-methoxyethyl)uridine.

2.3.3 Preclinical Experience

Detailed information concerning the preclinical studies conducted with ISIS 396443 can be found in the Investigator's Brochure. A summary is included below.

ISIS 396443 was identified after an extensive screen of greater than 500 2′-MOE oligonucleotides in *in vitro* splicing assays, reporter gene assays and in SMA patient fibroblasts (Hua et al. 2007; Hua et al. 2008). Data have shown that ISIS 396443 promotes a concentration-dependent increase in full-length transcripts (including Exon 7) in patient fibroblast cells, achieving greater than 90% full length SMN2 transcripts and forms nuclear structures, called gems, known to contain SMN protein. In a mild mouse model of SMA, ISIS 396443 promoted inclusion of Exon 7 in the SMN2 transgene in a variety of peripheral

tissues when dosed systemically (Hua et al. 2008) and in central nervous system (CNS) tissue, including spinal cord, when injected into the lateral ventricle. ISIS 396443 produced greater than 90% Exon 7 inclusion in the transgenic mice and increased SMN protein production in motor neurons, resulting in the appearance of gems in motor neurons. These studies were extended to a more severe mouse model of SMA (SMA Δ7) (Le et al. 2005), where the CNS delivery of drug produced a dose-dependent effect on SMN2 Exon 7 inclusion, SMN protein production, and survival. These mice treated with ISIS 396443 demonstrated improved weight gain, improvements in muscle morphology, muscle strength, and motor coordination and improved morphology of the motor neuron junctions (Passini et al. 2011). Further, ISIS 396443 was shown to distribute widely in the CNS following intrathecal (IT) administration in monkey (Passini et al. 2011).

The pharmacokinetics and toxicity of ISIS 396443 were assessed following: 1) single intrathecal (IT) lumbar bolus injections (1 to 7 mg) in adult monkeys 2) following 14 weeks (with a 4-week interim sacrifice) of repeated IT lumbar bolus injections (0.3 to 3 mg/wk or every other week) in juvenile monkeys and 3) following 1-year of repeated IT lumbar bolus injections in juvenile monkeys. Detailed results from these preclinical studies conducted with ISIS 396443 can be found in the Investigator's Brochure.

2.3.4 Clinical Experience

Detailed information concerning the clinical studies conducted with ISIS 396443 can be found in the Investigator's Brochure. A summary is included below.

ISIS 396443 has been evaluated in a completed open-label, single ascending-dose (SAD) Phase 1 study designed to assess the safety, tolerability and pharmacokinetics of ISIS 396443 in patients with SMA (ISIS 396443-CS1). A single-dose of ISIS 396443 was administered by IT injection to SMA patients aged 2 to 14 years of age. Four (4) dose levels (1, 3, 6, and 9 mg) were evaluated sequentially. Each dose level was studied in a cohort of 6 or 10 patients, where all patients received drug. In this study all subjects completed dosing and the follow-up visits per protocol. Overall, ISIS 396443 was well-tolerated and no safety concerns were identified up to the 9.0 mg dose level, given as a single IT injection. No serious adverse events (SAEs) or dose-limiting toxicities (DLTs) were reported in ISIS 396443-CS1. Adverse events (AEs) reported were mild or moderate in severity and there was no relationship with ISIS 396443 dose level. In addition, no ISIS 396443 related adverse changes in neurological exams were reported, despite intensive monitoring during the immediate post-dosing period. CSF and plasma drug concentrations observed were generally consistent with predictions made from nonclinical monkey studies.

ISIS 396443 is also being evaluated in seven ongoing studies: ISIS 396443-CS2, ISIS 396443-CS10, ISIS 396443-CS12, ISIS 396443-CS3B (ENDEAR), ISIS 396443-CS4 (CHERISH), 232SM201 (NURTURE) and 232SM202 (EMBRACE).

ISIS 396443-CS10 is an open-label, single dose, re-dosing study for SMA patients who previously participated in Cohorts 2, 3 and 4 in ISIS 396443-CS1.

ISIS 396443-CS2 is an open-label, multiple ascending-dose (MAD) Phase 1/2a study designed to assess the safety, tolerability and pharmacokinetics of ISIS 396443 in patients with SMA.

Multiple doses of ISIS 396443, ranging from 3 mg to 12 mg, are being administered by intrathecal injection to SMA patients aged 2 to 15 years of age.

ISIS 396443-CS12 is an open-label, multiple-dose study to assess the safety and tolerability of a 12 mg dose level of ISIS 396443 administered intrathecally in patients with spinal muscular atrophy who previously participated in ISIS 396443-CS2 or ISIS 396443-CS10.

ISIS 396443-CS3B (ENDEAR) is a randomized, double-blind, sham-procedure controlled study designed to assess clinical efficacy and safety of ISIS 396443 in patients with infantile-onset SMA. A 12 mg dose equivalent scaled by CSF volume is being evaluated in symptomatic SMA infants ≤ 7 months at the time of consent of age utilizing a loading regimen of 4 intrathecal doses on Days 1, 15, 29 and 64, followed by a q4 months maintenance period.

ISIS 396443-CS4 (CHERISH) is a randomized, double-blind, sham-procedure controlled study designed to assess clinical efficacy and safety of ISIS 396443 in patients with later-onset SMA aged 2-12 years. A 12 mg dose is being evaluated utilizing a loading regimen of 3 intrathecal doses on Days 1, 29 and 85, followed by a q6 months maintenance period.

232SM201 (NURTURE) is an open-label study to assess the efficacy, safety, tolerability and pharmacokinetics of multiple doses of ISIS 396443 delivered intrathecally to subjects with genetically diagnosed and pre-symptomatic SMA.

232SM202 (EMBRACE) is a randomized, double-blind, sham-procedure controlled study to assess safety and tolerability and to explore efficacy of ISIS 396443 administered intrathecally in subjects with SMA who are not eligible to participate in the clinical studies ISIS 396443-CS3B or ISIS 396443-CS4.

2.4 Rationale for Dose and Schedule of Administration

The proposed study will test the clinical efficacy, safety, tolerability, and PK of multiple doses of ISIS 396443 administered as IT injections. Two (2) 'loading' dose levels (scaled by infant age to be equivalent to 2 year old doses of 6 mg or 12 mg, based on CSF volume) will be evaluated sequentially, given during a'loading' dosing period on Study Days 1, 15, and 85. Following that, all subjects will be given 'maintenance' doses of 12 mg equivalent ISIS 396443 on Study Days 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261.

ISIS 396443 dose levels and dose interval for ISIS 396443-CS3A were selected based on preclinical toxicology and pharmacokinetic observations from monkey studies utilizing single-dose and repeat dosing (for 14 weeks) IT administration, consideration of the target tissue concentration anticipated for drug pharmacology, and safety data in the completed and ongoing clinical studies of ISIS 396443 to date. Based upon pharmacology and pharmacokinetic results in SMA transgenic mice, we estimate that the target tissue concentration to produce 50 to 90% SMN2 Exon 7 inclusion is between 1 and 10 μg/g spinal cord tissue. Nonclinical studies in juvenile monkeys receiving IT doses of ISIS 396443 showed a resulting gradient of distribution of ISIS 396443 along the spinal cord, with mean lumbar spinal cord concentrations ranging from 1.6-2.3 fold and 2.0-3.5 fold higher than thoracic and cervical spinal cord levels, respectively. The lowest dose selected for this multiple-dose clinical study (6 mg ISIS 396443) is predicted to achieve spinal cord tissue concentrations at the low end of this range (approximately 6 μg/g lumbar and 2 μg/g cervical concentrations), while the highest dose (12 mg ISIS 396443) is

predicted to achieve levels at the high end of this range (approximately 10 µg/g lumbar and 3 µg/g cervical spinal cord tissue concentrations), following the first dose.

The loading dose interval was selected based on the nonclinical pharmacokinetic and pharmacology data as the dose interval to achieve and maintain ISIS 396443 spinal cord tissue levels that are predicted to be within the upper end of the pharmacologically active range by Day 85 (predicted to be approximately 30 µg/g lumbar and 10 µg/g cervical tissue concentration), while at the same time considering subject safety and convenience for repeated LP intrathecal injections. The maintenance dose interval (once every 4 months) was selected based on the estimated spinal tissue and CSF drug half-life (4-6 months) and was selected to maintain spinal cord tissue levels of ISIS 396443 at a steady-state level within the estimated pharmacologically active range.

Additional details on dose scaling and expected CSF and tissue concentrations are summarized in the Investigator's Brochure.

3. EXPERIMENTAL PLAN

3.1 Study Design

This study will test the clinical efficacy, safety, tolerability, and PK of multiple doses of ISIS 396443 administered as IT injections by LP. Two (2) 'loading' dose levels (scaled by infant age to be equivalent to 2 year old doses of 6 mg or 12 mg, based on CSF volume) will be evaluated sequentially. The initial dose level of 6 mg will be studied in a cohort of 4 subjects. The 12 mg dose level will be studied in 4 to approximately 16 subjects. Following this, all subjects will receive 'maintenance' doses of 12 mg equivalent ISIS 396443 on Days 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261.

Cohort 1 (n = 4): 'Loading' dosing of 6 mg equivalent ISIS 396443 on Days 1, 15, 85, IT injection; 'maintenance' doses of 12 mg equivalent on Days 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261, IT injection.

Cohort 2 (n = 4-16): 'Loading' dosing of 12 mg equivalent ISIS 396443 on Days 1, 15, 85, IT injection, 'maintenance' doses of 12 mg equivalent on Days 253, 379, 505, 631, 757, 883, 1009, 1135, 1261, IT injection.

3.2 Number of Study Centers

This study will be conducted at multiple centers in the United States and Canada.

3.3 Number of Subjects

Approximately 8-20 subjects will be enrolled into this multiple-dose study of ISIS 396443. The number of subjects may be higher if some subjects must be replaced and/or if the sizes of some cohorts are expanded in order to obtain further experience with some dose levels.

3.4 Overall Study Duration and Follow-up

The Study will consist of screening, treatment period including 'loading' dosing and 'maintenance' dosing, and post-treatment follow-up period. The total duration of participation in the study is approximately 3.7 years. Please refer to the Schedule of Procedures in Appendix A.

3.4.1 Screening

After informed consent/assent is obtained, subjects will undergo a Screening evaluation no greater than 21 days prior to first dose administration at which their eligibility for the study will be examined.

3.4.2 Treatment

Subjects who meet the eligibility criteria will be admitted to the study center on Study Day 1, undergo pre-dose evaluations, and then receive an LP injection of Study Drug (ISIS 396443). The subject will remain at the study center for at least 24 hours post-injection for safety monitoring and laboratory specimen collection. Subjects will return to the tudy center on Days 15, 85, 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261 for follow-up evaluations and subsequent injections. Following LP injections on Study Days 15, 85, 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261 subjects will not stay overnight in the hospital but will be monitored for at least 6 hours post-injection before leaving the study facility. A CSF sample will be taken pre-dose on each injection day for safety and Pharmacokinetic (PK) analyses. During the treatment period, the study center will monitor the subject's condition through safety monitoring visits on Study Days 16, 29, 86, 92, 169, 254, 337, 442, 568, 694, 820, 946, 1072, and 1198, and by telephone contact on Study Days 8, 43, 57, 71, 106, 127, 134, 148, 189, 197, 218, 239, 274, 295, 316, 358, 380, 400, 421, 463, 484, 506, 526, 547, 589, 610, 632, 652, 673, 715, 736, 758, 778, 799, 841, 862, 884, 904, 925, 967, 988, 1010, 1030, 1051, 1093, 1114, 1136, 1156, 1177, 1219, 1240, 1262, 1282, 1303, and 1324 (i.e., approximately every 3 weeks).

3.4.3 Post-Treatment Follow-up

During the post-treatment follow-up period, the study center will monitor the subject's condition through telephone contact on Study Days 1282, 1303, and 1324 (i.e., every 3 weeks through 13 weeks after the last dose of ISIS 396443). A safety monitoring visit will occur on Day 1352 (through 13 weeks after the last dose of ISIS 396443).

If a subject terminates early from the study, they will be encouraged to complete all assessments per the Day 1352 visit.

3.5 End of Study

The end of study is last subject, last visit (either in-person visit or telephone contact).

3.6 Safety Monitoring and Dose Escalation

Safety data will be reviewed on an ongoing basis by the Sponsor and the Ionis Medical Monitor. Safety data will also be reviewed on an ongoing basis by an independent Data and Safety Monitoring Board (DSMB).

The progression of the study from the initial dose level (Cohort 1) to Cohort 2 will be determined by the Sponsor and the DSMB and generally be based on the number of DLTs observed in patients treated with ISIS 396443. In general, the occurrence of DLTs in 2 patients in Cohort 1 may result in the dose tested being considered as dose limiting. The occurrence of 1 DLT in 4 subjects in Cohort 1 may result in the cohort being expanded to 6 subjects (2 additional subjects) to further assess the safety within that dose level.

After the last patient within Cohort 1 completes the Day 29 visit, safety results for the cohort will be reviewed by the Sponsor and the DSMB and a recommendation regarding further enrollment and escalation to Cohort 2 will be made. The Sponsor and DSMB will determine if enrollment of additional patients is required to confirm there is an acceptable toxicity profile prior to its designation as MTD.

3.7 Dose Limiting Toxicity

At the onset of the study, a DLT is defined as an adverse event that in the judgment of a Site Investigator is of sufficient significance to be dose limiting, is possibly or definitely related to Study Drug (i.e., the AE is substantially less likely to occur in subjects not treated with Study Drug), and that is not a known: 1) sign or symptom of SMA disease, or 2) effect of the LP injection procedure.

If a suspected DLT occurs during injection of Study Drug, treatment of the subject with Study Drug should be stopped (i.e., the injection should be immediately discontinued). The Site Investigator should contact the Ionis Medical Monitor as soon as possible following any dosing discontinuations to discuss the case. The DSMB will also be informed and will determine if any relevant findings have been observed with other subjects in the study.

Subjects that experience a DLT will discontinue study treatment but will be monitored by follow-up safety visits.

4. SUBJECT ENROLLMENT

4.1 Screening

Before subjects may be enrolled into the study, the Sponsor requires a copy of the Study Center's written Institutional Review Board (IRB) or Research Ethics Board (REB) approval of the protocol, informed consent form, informed assent form (if applicable) and all other subject information and/or recruitment material.

Before a subject's participation in the trial, the Investigator is responsible for obtaining written informed consent from the parent(s) or legal guardian(s). At the time of consent, the subject will be considered enrolled into the study and will be assigned a unique screening number before any study procedures, including screening procedures, are performed. At the time of registration, subjects will be assigned a unique subject identification number. This number will be used to identify the subject throughout the trial and must be used on all study documentation related to that subject. The screening number and subject identification number must remain constant throughout the entire trial. In the event the subject is re-consented and re-screened the subject must be given a new screening number. Screening numbers and subject identification numbers, once assigned, will not be re-used.

4.2 Registration

Subjects will be registered after all screening assessments have been completed and after the Investigator and the Ionis Medical Monitor have verified that they are eligible per criteria in Sections 5.1 and 5.2.

No subject may begin treatment prior to registration and assignment of a unique subject identification number.

4.3 Replacement of Subjects

If a subject withdraws from the trial prior to the administration of Study Drug, the subject will be replaced. If a subject withdraws for reasons other than safety after receiving any amount of Study Drug and prior to the Day 85 visit, the Site Investigator and the Ionis Medical Monitor will discuss the reason(s) for the discontinuation and the appropriateness of enrolling a replacement subject. If a subject withdraws for safety reasons that are potentially drug related after receiving any amount of Study Drug, the DSMB will review the case and provide recommendations regarding replacement of the subject.

5. SUBJECT ELIGIBILITY

5.1 Inclusion Criteria

To be eligible to participate in this study candidates must meet the following eligibility criteria at screening.

1. Signed informed consent of parent(s) or guardian(s)
2. Genetic documentation of 5q SMA homozygous gene deletion or mutation
3. Onset of clinical signs and symptoms consistent with SMA at ≥ 21 days and ≤ 6 months (180 days) of age
4. Males and females between ≥ 21 days and ≤ 7 months (210 days) of age at Screening
5. At study entry, receiving adequate nutrition and hydration (with or without gastrostomy), in the opinion of the Site Investigator
6. Body weight > 5th percentile for age using CDC guidelines
7. Medical care meets and is expected to continue to meet guidelines set out in the Consensus Statement for Standard of Care in SMA (Wang et al. 2007), in the opinion of the Site Investigator
8. Gestational age of 35 to 42 weeks and gestation body weight ≥ 2 kg
9. Reside within approximately 9 hours ground-travel distance from a participating study center for the duration of the study. Residence > 2 hours ground-travel distance from a study center must obtain clearance from the Site Investigator and the study Medical Monitor
10. Able to complete all study procedures, measurements and visits and parent or guardian/subject has adequately supportive psychosocial circumstances, in the opinion of the Site Investigator

5.2 Exclusion Criteria

Subjects meeting any of the following criteria are not eligible for the study:

1. Hypoxemia (O_2 saturation awake < 96% or O_2 saturation asleep < 96%, without ventilation support)
2. Presence of an untreated or inadequately treated active infection requiring systemic antiviral or antimicrobial therapy at any time during the screening period
3. History of brain or spinal cord disease that would interfere with the LP procedures, CSF circulation, or safety assessments
4. Presence of an implanted shunt for the drainage of CSF or an implanted CNS catheter
5. History of bacterial meningitis
6. Clinically significant abnormalities in hematology or clinical chemistry parameters, as assessed by the Site Investigator, at screening that would render the subject unsuitable for inclusion
7. Treatment with another investigational drug (e.g., albuterol, riluzole, carnitine, creatine, sodium phenylbutyrate, salbutamol, valproate, hydroxyurea, etc.), biological agent, or device within 90 days prior to enrollment or anytime during the study. Any history of gene therapy or cell transplantation
8. The subject's parent(s) or legal guardian(s) is unable to understand the nature, scope, and possible consequences of the study, or does not agree to comply with the protocol defined schedule of assessments
9. Ongoing medical condition that according to the Site Investigator would interfere with the conduct and assessments of the study. Examples are medical disability other than SMA that would interfere with the assessment of safety or would compromise the ability of the subject to undergo study procedures

6. STUDY PROCEDURES

6.1 Study Schedule

All required study procedures are outlined in Appendices A, B and C.

6.2 Study Assessments

6.2.1 Collection of CSF

Subjects will have CSF collected pre-dose during the LP procedure on Days 1, 15, 85, 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261 for safety and PK analyses. Approximately 5 mL of CSF will be collected by lumbar puncture, using a 'spinal anesthesia' needle and a standard LP collection kit. A 22G anesthesia needle is recommended. Depending on institutional guidelines, anesthesia or sedation may be used for the procedure. Spinal ultrasound may be used for the LP procedure, if deemed necessary, but is not required. CSF will be used for standard laboratory

measurement of cells, glucose, and protein, and ISIS 396443 pharmacokinetic analyses. Extra CSF may be stored for investigation of possible biomarkers of SMA disease or the pharmacodynamic effects of ISIS 396443 (e.g., CSF mRNA, CSF protein, CSF miRNA panel) or for profiling of drug binding proteins, bioanalytical method validation purposes, stability assessments, metabolite assessments, immunogenicity assessments (including assay development and validation purposes), or to assess other actions of ISIS 396443 with CSF constituents.

6.2.2 Laboratory Analytes

Laboratory measurements of serum chemistry, hematology, urinalysis, coagulation parameters, and plasma antibodies to ISIS 396443 will be performed at the times shown in the Schedule of Procedures (Appendix A). The analytes to be measured are shown in Appendix B.

6.2.3 Neurological Examinations as Assessed by the Hammersmith Infant Neurological Examination (HINE)

Neurological examinations will be performed using the Hammersmith Infant Neurological Examination (HINE). These examinations will include assessment of mental status, level of consciousness, sensory function, motor function, cranial nerve function, and reflexes. Motor milestones will be accessed using Section 2 of the HINE, which is comprised of 8 independent milestone categories: voluntary grasp, ability to kick in supine position, head control, rolling, sitting, crawling, standing, and walking. HINE will be performed at the times/dates according to the schedule as shown in Appendix A (Schedule of Procedures).

6.2.4 Pharmacokinetics Specimen Collection

Plasma and CSF specimens will be collected as shown in Appendix A (Schedule of Procedures) and Appendix C (Pharmacokinetic Sampling Schedule). The following ISIS 396443 plasma PK parameters (though not necessarily limited to) will be derived when appropriate from the individual subject concentration vs. time profiles using noncompartmental-based methods and based on actual sampling times:

- The maximal observed plasma drug concentration (C_{max})
- The time to reach C_{max} in plasma (T_{max})
- The area under the plasma concentrations time curve from the time of the IT dose to the last collected sample
- The apparent terminal elimination half-life ($t_{½}$), if possible

6.2.5 Children's Hospital of Philadelphia Infant Test for Neuromuscular Disease (CHOP INTEND)

Subjects will be evaluated using the CHOP INTEND test according to the schedule as shown in Appendix A (Schedule of Procedures). The CHOP INTEND test was specifically designed to evaluate the motor skills of infants with significant motor weakness, including infants with SMA (Glanzman et al. 2010). The CHOP INTEND test captures neck, trunk, proximal and distal limb strength in 14 elicited and 2 observational items. The CHOP INTEND has been established as a safe and reliable infant motor measure in infantile-onset SMA.

6.2.6 Hammersmith Functional Motor Scale – Expanded

All subjects who have maintained a CHOP INTEND total score of ≥ 50 for 2 consecutive study visits will be evaluated using the Hammersmith Functional Motor Scale Expanded (HFMSE) at the times shown in Appendix A (Schedule of Procedures). This evaluation will be performed in addition to the CHOP INTEND. The HFMSE is a reliable and validated tool to assess motor function in children with SMA. The scale was originally developed with 20 scored activities and was devised for use in children with SMA type 2 and type 3 with limited ambulation to give objective information on motor ability and clinical progression (Main et al. 2003). The expanded scale includes an additional module of 13 items developed to allow for evaluation of ambulatory SMA patients (O'Hagen et al. 2007). The HFMSE has been shown to be highly correlated with other clinical assessments and shows good test-retest reliability. The HFMSE is easy to use and quickly administered.

6.2.7 Compound Muscle Action Potential

Measurements of Compound Muscle Action Potential (CMAP) will be assessed (ulnar and peroneal nerves) according to the schedule as shown in Appendix A (Schedule of Procedures). CMAP is an electrophysiological technique that can be used to determine the approximate number of motor neurons in a muscle or group of muscles. CMAP is a well validated method for tracking disease progression in neuromuscular disorders such as spinal muscular atrophy (SMA) (Swoboda et al. 2005; Lewalt et al. 2010) and amyotrophic lateral sclerosis (Shefner et al. 2011) and has been proposed as a potential biomarker of a therapeutic effect in SMA.

6.2.8 Growth Parameters

Subjects will be assessed by growth measurements according to the schedule as shown in Appendix A (Schedule of Procedures). Growth parameters of body length, head circumference, chest circumference, and arm circumference will be measured. Additional parameters of weight for age, weight for length, and head to chest circumference ratio will be calculated.

6.2.9 Ventilation Use

The subject's ventilator use will be recorded at the times/dates according to the schedule as shown in Appendix A (Schedule of Procedures).

6.2.10 Phone Assessments

Monitoring telephone calls will occur on Study Days 8, 43, 57, 71, 106, 127, 134, 148, 189, 197, 218, 239, 274, 295, 316, 358, 380, 400, 421, 463, 484, 506, 526, 547, 589, 610, 632, 652, 673, 715, 736, 758, 778, 799, 841, 862, 884, 904, 925, 967, 988, 1010, 1030, 1051, 1093, 1114, 1136, 1156, 1177, 1219, 1240, 1262, 1282, 1303, and 1324 (i.e., approximately every 3 weeks between study visits). Changes in concomitant medications and adverse events will be recorded. In addition, information on the subject's ventilator use and SMA disease status will be collected.

6.2.11 Safety Evaluations

Safety will be evaluated by assessment of AEs including SAEs as described in Section 9. Additional safety evaluations include the following parameters:

- Neurological examinations (HINE)

- Vital signs
- Physical examinations and weight
- Clinical laboratory tests (serum chemistry, hematology, urinalysis)
- CSF laboratory tests (cell count, protein, glucose)
- Electrocardiogram (ECG)
- Use of concomitant medications

7. STUDY DRUG

7.1 Study Drug

There are 2 types of Study Drug that may be provided for this study:

a. Study Drug at a concentration of 20 mg/mL which is diluted prior to use; this is described as the 'Two-Vial' configuration. See Section 7.1.1 for more details.

b. Study Drug at a concentration of 2.4 mg/mL; this is described as 'Ready-to-Use Vial' configuration. See Section 7.1.2 for more details.

7.1.1 Two-Vial Configuration (ISIS 396443 and Diluent) Description

ISIS 396443 active drug product and artificial CSF diluent are manufactured by Ionis Pharmaceuticals, Inc., Carlsbad, CA, USA. The active drug product is supplied at a concentration of 20 mg/mL as a 2.5 mL fill volume in a 5 mL clear glass vial. The diluent (artificial CSF) is supplied as a 20 mL fill volume in a 30 mL clear glass vial. These configurations allow for various clinical doses by using different dilution procedures between the active drug product and diluent vials. Study Drug characteristics are described in Table 1. More details are provided in the Investigators Brochure.

Table 1 Study Drug Characteristics

Study Drug	ISIS 396443	Diluent (artificial CSF)
Strength	20 mg/mL	NA
Volume/vial	2.5 mL solution per vial	20 mL solution per vial
Route of Administration	IT injection	IT injection

ISIS 396443 and the diluent (artificial CSF) must be stored securely at 2° C to 8° C.
ISIS 396443 must be protected from light.

7.1.2 Ready-To-Use Vial Configuration Description

In the 'Ready-to-use' vial configuration, the Study Drug is supplied in 6 mL clear glass vials at a concentration of 2.4 mg/mL. The Study Drug and its storage and preparation instructions will be provided by the Sponsor or designee. Study Drug characteristics are listed under Table 2.

Table 2 Study Drug Characteristics: Ready-To-Use Vial Configuration

Study Drug	ISIS 396443 Drug Product
Strength	2.4 mg/mL
Volume/vial	5.0 mL solution per vial
Route of Administration	IT injection

ISIS 396443 must be stored securely at 2° to 8° C and protected from light.

7.2 Packaging and Labeling

Ionis Pharmaceuticals, Inc. will provide the Site Pharmacists with packaged Study Drug labeled in accordance with regulatory requirements.

7.3 Study Drug Accountability

The study staff is required to document receipt, dispensing and return of Study Drug supplies provided by the Sponsor. Drug accountability documentation and all used and unused Study Drug vials (ISIS 396443 and diluent) must be returned to Ionis Pharmaceuticals, Inc. or designee.

8. TREATMENT OF SUBJECTS

8.1 Study Drug Administration

Each subject will receive a single IT bolus (1-3 minute) LP injection of Study Drug using a 'spinal anesthesia' needle and 5 mL syringe. A 22G spinal anesthesia needle is recommended. The target site for needle insertion is the L3/L4 space, but may be 1 segment above or 1-2 segments below this level, if needed. Prior to the injection 5-6 mL of CSF fluid is to be collected for analyses. Depending on institutional guidelines, anesthesia or sedation may be used for the LP procedure, following institutional procedures. Spinal ultrasound may be used for the LP procedure, if deemed necessary, but is not required.

Table 3 outlines the dose equivalent and ISIS 396443 concentration for ISIS 396443 dose cohorts to be delivered during the 'loading' dosing treatment period (i.e., dosing on Days 1, 15 and 85). Study Drug dosing during the 'loading' dosing treatment period will be done using the two-vial configuration.

Table 4 outlines the dose equivalent and ISIS 396443 concentration for ISIS 396443 to be delivered during the 'maintenance' dosing treatment period (i.e., dosing on Days 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261). Study Drug dosing during the 'maintenance' dosing

treatment period will be done using the two-vial configuration and will transition to the ready-to-use vial configuration once the current protocol amendment has been approved.

Table 3 ISIS 396443 Dose Equivalents and Concentration During the 'Loading' Dosing Treatment Period

Cohort	Dose Equivalent (mg)	Concentration (mg/mL)	Injection Volume (mL)
1	6	1.2	See Table 5
2	12	2.4	See Table 5

Table 4 ISIS 396443 Dose Equivalent and Concentration During the 'Maintenance' Dosing Treatment Period

Cohorts	Dose Equivalent (mg)	Concentration (mg/mL)	Injection Volume (mL)
1 and 2	12	2.4	See Table 5

The volume of the injection will be adjusted based on the subject's age. Subjects who are older than 24 months (730 Days) on the day of dosing will receive the full 12 mg dose of the Study Drug (5 mL). For subjects younger than 24 months of age, the volume of the injection will be adjusted based on the subject's age on the day of dosing per Table 5, such that each subject will receive a 12 mg equivalent dose based on CSF volume scaling.

Table 5 ISIS 396443 Dose Volume to be Injected

Age	Estimated CSF Volume*	Injection Volume (mL)
0-3 months (0-90 days)	120 mL	4 mL
3-6 months (91-182 days)	130 mL	4.3 mL
6-12 months (183 – 365 days	135 mL	4.5 mL
12-24 months (366 – 730 days)	140 mL	4.7 mL
> 24 months (> 730 days)	150 mL	5.0 mL

* Matsuzawa et al. 2001

Please refer to the Study Drug Manual provided by the Sponsor for detailed instructions for Study Drug preparation and administration.

8.2 Other Protocol-Required Drugs

There are no other protocol required drugs, but depending on institutional guidelines anesthesia or sedation may be used for the LP procedure, following institutional procedures.

8.3 Other Protocol-Required Procedures

There are no other protocol-required treatment procedures.

8.4 Treatment Precautions

Subjects will be encouraged to lie flat for 1 hour following dosing, if possible.

8.5 Safety Monitoring Rules

Please refer to the Guidance to Investigator section of the Investigator Brochure.

8.6 Stopping Rules

Please refer to Section 8.8. There are no additional specific stopping rules for this study but the Investigator should discuss significant concerns relating to individual subjects with the Ionis Medical Monitor to ensure that it is appropriate for the subject to continue Study Drug.

8.7 Adjustment of Dose and/or Treatment Schedule

No adjustment of dose is permitted. In the event of a concurrent illness that would prevent the dosing procedure from being performed safely, an adjustment in the dose schedule may be permitted, but must be approved by the Sponsor Medical Monitor. In this case, the scheduled Day 15 dosing visit may be delayed until up to Day 70 and the scheduled Day 85 dosing visit may be delayed up to Day 120. The scheduled dosing visits during the 'maintenance' period (i.e., Days 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261) may be delayed by up to 8 weeks.

8.8 Discontinuation of Study Treatment

A subject must permanently discontinue study treatment for any of the following:

- The subject withdraws consent
- The subject experiences an adverse event that necessitates permanent discontinuation of study treatment
- The subject experiences a DLT as defined in Section 3.7

The reason for discontinuation of study treatment must be recorded in the Case Report Form (CRF) and source documentation.

Subjects that discontinue treatment will enter the post-treatment follow-up period unless consent is withdrawn (see Appendix A).

8.9 Withdrawal of Subjects from the Study

Subjects must be withdrawn from the study for any of the following:

- Withdrawal of consent

- The subject is unwilling or unable to comply with the protocol

Other reasons for withdrawal of subjects from the study might include:

- At the discretion of the Investigator for medical reasons
- At the discretion of the Investigator or Sponsor for noncompliance
- Significant protocol deviation
- Administrative decision by the Investigator or Sponsor

All efforts will be made to complete and report the observations as thoroughly as possible up to the date of withdrawal. All information, including the reason for withdrawal from Study, must be recorded in the CRF.

Any subject who withdraws consent to participate in the study will be removed from further treatment and study observation immediately upon the date of request. These subjects should be encouraged to complete the early termination study procedures and observations at the time of withdrawal (Appendix A).

For subjects withdrawn for reasons other than withdrawal of consent every effort should be made to complete the early termination study procedures and observations at the time of withdrawal (see Appendix A).

8.10 Concomitant Therapy and Procedures

The use of concomitant therapies or procedures defined below must be recorded on the subject's CRF. AEs related to administration of these therapies or procedures must also be documented on the appropriate CRF.

8.10.1 Concomitant Therapy

A concomitant therapy is any non-protocol specified drug or substance (including over-the-counter medications, herbal medications and vitamin supplements) administered between screening and last telephone contact.

Subjects should consult with the Site Investigator or qualified designee prior to initiating any new medication, including non-prescription compounds or any other non-drug therapy.

Allowed Concomitant Therapy

Throughout the study, Site Investigators or designated licensed physicians involved in the study may prescribe concomitant medications or treatments deemed necessary for adverse events or to provide adequate supportive care.

Disallowed Concomitant Therapy

Study subjects are prohibited from receiving other experimental agents during the study. This includes marketed agents being used off-label and/or at experimental doses that are being tested

for the treatment of SMA. The following agents are specifically prohibited: valproate, riluzole, carnitine, creatine, sodium phenylbutyrate, hydroxyurea, salbutamol.

8.10.2 Concomitant Procedures

A concomitant procedure is any therapeutic intervention (e.g., surgery/biopsy) or diagnostic assessment (e.g., blood gas measurement, bacterial cultures) performed between screening and last visit.

9. SERIOUS AND NON-SERIOUS ADVERSE EVENT REPORTING

9.1 Sponsor Review of Safety Information

Safety information will be collected, reviewed, and evaluated by the Sponsor in accordance with the Safety Management Plan throughout the conduct of the clinical trial.

9.2 Regulatory Requirements

The Sponsor is responsible for regulatory submissions and reporting to the Investigators of SAEs including suspected unexpected serious adverse reactions (SUSARs) per the International Conference on Harmonization (ICH) guidelines E2A and ICH E6. Country-specific regulatory requirements will be followed in accordance with local country regulations and guidelines.

IRBs/REBs will be notified of any serious adverse event (SAE) according to applicable regulations. The DSMB will be notified of any SAE as specified in the DSMB charter.

The Sponsor will evaluate the available information and decide if there is a reasonable possibility that the Study Drug caused the AE and, therefore, meets the definition of a SUSAR.

For the purpose of regulatory reporting of SUSARs, there are no "expected" AEs in this study population. For Study Drug "expected" AEs, refer to the Investigator Brochure.

9.3 Definitions

9.3.1 Adverse Event

An adverse event is any unfavorable and unintended sign (including a clinically significant abnormal laboratory finding, for example), symptom, or disease temporally associated with the study or use of investigational drug product, whether or not the AE is considered related to the investigational drug product.

9.3.2 Adverse Reaction and Suspected Adverse Reaction

An adverse reaction is any adverse event caused by the Study Drug.

A suspected adverse reaction is any adverse event for which there is a reasonable possibility that the drug caused the adverse event. A suspected adverse reaction implies a lesser degree of certainty about causality than an adverse reaction.

9.3.3 Serious Adverse Event (SAE)

A serious adverse event is any adverse event that in the view of either the Investigator or Sponsor, meets any of the following criteria:

- Results in death
- Is life threatening: that is, poses an immediate risk of death at the time of the event

  An AE or suspected adverse reaction is considered "life-threatening" if, in the view of either the Investigator or Sponsor, its occurrence places the subject at immediate risk of death. It does not include an AE or suspected adverse reaction that, had it occurred in a more severe form, might have caused death.
- Requires inpatient hospitalization or prolongation of existing hospitalization

  Hospitalization is defined as an admission of greater than 24 hours to a medical facility and does not always qualify as an AE
- Results in a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- Results in congenital anomaly or birth defect in the offspring of the subject (whether the subject is male or female)
- Important medical events that may not result in death, are not life-threatening, or do not require hospitalization may also be considered serious when, based upon appropriate medical judgment, they may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

9.4 Monitoring and Recording Adverse Events

Any pre-existing conditions or signs and/or symptoms present in a subject prior to the start of the study (i.e., before informed consent) should be recorded as Medical History and not recorded as AEs unless the pre-existing condition worsened. The Investigator should always group signs and symptoms into a single term that constitutes a **single unifying diagnosis** if possible.

9.4.1 Serious Adverse Events

In the interest of subject safety, and in order to fulfill regulatory requirements**,** all SAEs (regardless of their relationship to Study Drug) should be reported to the Sponsor within 24 hours of the Study Center's first knowledge of the event. The collection of SAEs will begin after the subject signs the informed consent form and stop at the end of the subject's follow-up period which is defined as the subject's last visit. When the Investigator is reporting by telephone, it is important to speak to someone in person versus leaving a message. An Initial Serious Adverse Event Form should be completed and a copy should be faxed to the Sponsor.

Detailed information should be actively sought and included on Follow-Up Serious Adverse Event Forms as soon as additional information becomes available. All SAEs will be followed until resolution. SAEs that remain ongoing past the subject's last protocol-specified follow-up visit will be evaluated by the Investigator and Sponsor. If the Investigator and Sponsor agree the subject's condition is unlikely to resolve, the Investigator and Sponsor will determine the follow-up requirement.

9.4.2 Non-Serious Adverse Events

The recording of non-serious AEs will begin after the subject signs the informed consent form and will stop at the end of the subject's follow-up period, which is defined as subject's last visit. The Investigator will monitor each subject closely and record all observed or volunteered AEs on the Adverse Event Case Report Form.

9.4.3 Evaluation of Adverse Events (Serious and Non-Serious)

The Investigator's opinion of the following should be documented on the Adverse Event Case Report Form:

9.4.3.1 Relationship to the Study Drug

The event's relationship to the Study Drug is characterized by one of the following:

- **Related:** There is clear evidence that the event is related to the use of Study Drug e.g., confirmation by positive re-challenge test
- **Possible:** The event cannot be explained by the subject's medical condition, concomitant therapy, or other causes, and there is a plausible temporal relationship between the event and Study Drug administration
- **Unlikely/Remote:** An event for which an alternative explanation is more likely (e.g., concomitant medications or ongoing medical conditions) or the temporal relationship to Study Drug administration and/or exposure suggests that a causal relationship is unlikely (For reporting purposes, Unlikely/Remote will be grouped together with Not Related)
- **Not Related:** The event can be readily explained by the subject's underlying medical condition, concomitant therapy, or other causes, and therefore, the Investigator believes no relationship exists between the event and Study Drug

9.4.3.2 Severity

The event's severity is characterized by one of the following:

- **Mild:** The event is easily tolerated by the subject and does not affect the subject's usual daily activities
- **Moderate:** The event causes the subject more discomfort and interrupts the subject's usual daily activities
- **Severe:** The event is incapacitating and causes considerable interference with the subject's usual daily activities

If the event is an SAE, then all applicable seriousness criteria must be indicated (criteria listed in Section 9.3.3).

9.4.3.3 Action Taken with Study Drug

Action taken with Study Drug due to the event is characterized by one of the following.

- **None:** No changes were made to Study Drug administration and dose
- **Permanently Discontinued:** Study drug was discontinued and not restarted
- **Temporarily Interrupted, restarted – same dose:** Dosing was temporarily interrupted or delayed due to the AE and restarted at the same dose

9.4.3.4 Treatment Given for Adverse Event

Any treatment (e.g., medications or procedures) given for the AE should be recorded on the Adverse Event Case Report Form (treatment should also be recorded on the concomitant treatment or ancillary procedures CRF as appropriate).

9.4.3.5 Outcome of the Adverse Event

If the event is a non-serious AE then the event's outcome is characterized by one of the following:

- **AE Persists:** Subject terminates from the trial and the AE continues
- **Recovered:** Subject recovered completely from the AE
- **Became Serious:** The event became serious (the date that the event became serious should be recorded as the Resolution Date of that AE and the Onset Date of the corresponding SAE)
- **Change in Severity (if applicable):** AE severity changed

If the event is a SAE then the event's outcome is characterized by one of the following:

- **Ongoing:** SAE continuing
- **Persists (as non-serious AE):** Subject has not fully recovered but the event no longer meets serious criteria and should be captured as an AE on the non-serious AE electronic case report form (eCRF) (the SAE resolution date should be entered as the date of onset of that AE)
- **Recovered:** Subject recovered completely from the SAE (the date of recovery should be entered as the SAE resolution date)
- **Fatal:** Subject died (the date of death should be entered as the SAE resolution date)

9.5 Procedures for Handling Special Situations

9.5.1 Abnormalities of Laboratory Tests

Clinically significant abnormal laboratory test results may, in the opinion of the Investigator, constitute or be associated with an AE. Examples of these include abnormal laboratory results

that are associated with symptoms, or require treatment, e.g., bleeding due to thrombocytopenia, tetany due to hypocalcemia, or cardiac arrhythmias due to hyperkalemia. Whenever possible, the underlying diagnosis should be listed in preference to abnormal laboratory values as AEs. Clinically significant abnormalities will be monitored by the Investigator until the parameter returns to its baseline value or until agreement is reached between the Investigator and Ionis Medical Monitor. Laboratory abnormalities deemed not clinically significant (NCS) by the Investigator should not be reported as AEs. Similarly, laboratory abnormalities reported as AEs by the Investigator should not be deemed NCS on the laboratory sheet.

The Investigator is responsible for reviewing and signing all laboratory reports. The signed clinical laboratory reports will serve as source documents.

9.5.2 Prescheduled or Elective Procedures or Routinely Scheduled Treatments

A prescheduled or elective procedure or a routinely scheduled treatment will not be considered an SAE, even if the subject is hospitalized; the Study Center must document all of the following:

- The prescheduled or elective procedure or routinely scheduled treatment was scheduled (or was on a waiting list to be scheduled) prior to obtaining the subject's consent to participate in the study
- The condition that required the prescheduled or elective procedure or routinely scheduled treatment was present before and did not worsen or progress in the opinion of the Investigator between the subject's consent to participate in the study and the timing of the procedure or treatment
- The prescheduled or elective procedure or routinely scheduled treatment is the sole reason for the intervention or hospital admission

9.5.3 Dosing Errors

Study Drug dosing errors should be documented as Protocol Deviations. A brief description should be provided in the deviation, including whether the subject was symptomatic (list symptoms) or asymptomatic, and the event accidental or intentional.

Dosing details should be captured on the Dosing CRF. If the subject takes a dose of Study Drug that exceeds protocol specifications and the subject is symptomatic, then the symptom(s) should be documented as an AE and be reported per Section 9.4.

Should an overdose occur, the Investigator or designee should refer to the Guidance to Investigator's section of the Investigator's Brochure and contact the Sponsor within 24 hours.

10. STATISTICAL CONSIDERATIONS

10.1 Study Endpoints

10.1.1 Primary Endpoint

- Achievement of motor milestones as evaluated by Module 2 of the Hammersmith Infant Neurological Examination

10.1.2 Secondary Endpoints

The efficacy of multiple doses of ISIS 396443 as assessed by:

- Event-free survival determined by the proportion of subjects who are alive and do not require permanent ventilatory support (defined as tracheostomy or the need for ≥ 16 hours ventilation/day continuously for at least 14 days in the absence of an acute reversible illness)
- Improvement in muscle strength as measured by the Children's Hospital of Philadelphia Infant Test for Neuromuscular Disease (CHOP INTEND)
- Improvement in neuromuscular electrophysiology measured by the Compound Muscle Action Potential (CMAP) of the ulnar and peroneal nerves

The safety and tolerability of multiple doses of ISIS 396443 as assessed by:

- Adverse events
- Neurological examinations
- Vital signs
- Physical examinations and weight
- Clinical laboratory tests (serum chemistry, hematology, urinalysis)
- CSF laboratory tests (cell count, protein, glucose)
- ECGs
- Use of concomitant medications

CSF and plasma PK of ISIS 396443.

10.1.3 Exploratory Endpoints

The following efficacy evaluations will be performed as exploratory endpoints:

- Measures of respiratory status (number of respiratory events, respiratory infections, respiratory-related hospitalizations, ventilator use, O_2 saturation awake)
- Growth parameters (weight for age/length, head circumference, chest circumference, head to chest circumference ratio, arm circumference)

10.2 Sample Size Considerations

There is no statistical rationale for the selected sample size of 4 treated subjects in Cohort 1 and 4 to approximately 16 subjects in Cohort 2. The sample size was selected to ensure that the safety and tolerability of ISIS 396443 will be adequately assessed while minimizing unnecessary subject exposure rather than statistical considerations for the efficacy endpoints.

10.3 Populations

Safety Population: All subjects who are registered and receive at least one dose of Study Drug.

Pharmacokinetic Population: All subjects who are registered and for which there is at least one evaluable post-dose pharmacokinetic sample.

Efficacy Population: all subjects who are registered, receive all scheduled loading doses of Study Drug (i.e., Days 1, 15 and 85), and complete the Day 92 visit.

10.4 Interim Analysis

Interim efficacy and safety analyses will be performed to provide content for regulatory submissions and to support ISIS 396443 drug development planning and business activities. Details of the analyses are contained in the Statistical Analysis Plan.

10.5 Planned Methods of Analysis

10.5.1 Demographic and Baseline Characteristics

Demographic and baseline characteristics will be summarized descriptively for each dose cohort.

10.5.2 Efficacy Analysis

The primary analysis will include the proportion of subjects who achieved improvement in motor milestones at their last visit. It will be tabulated by cohort and overall.

Motor milestones will be accessed using Section 2 of the Hammersmith Infant Neurological Exam (HINE) which is comprised of 8 independent milestone categories: voluntary grasp, ability to kick in supine position, head control, rolling, sitting, crawling, standing, and walking. Within each of these categories, subjects can progress from complete inability to perform a motor task, for example no grasp, no kicking, inability to maintain head upright, all the way to complete mastery of each category, for example achievement of pincer grasp, touching toes while supine, or maintaining their head upright.

Event-free survival will be evaluated by determining a proportion of subjects who are alive and do not require permanent ventilatory support. Permanent ventilation is defined as tracheostomy or the need for ≥ 16 hours ventilation/day continuously for at least 14 days in the absence of an acute reversible illness.

The CHOP INTEND infant motor function scale total score, change, and percent change from baseline will be summarized by cohort and overall, and by visit.

The absolute values for CMAP amplitude and CMAP area, as well as absolute and percent changes from baseline, will be summarized by cohort and overall, and by visit.

Full details of the methodology for analyses described above, as well as all additional analyses, are contained in the Statistical Analysis Plan.

10.5.3 Safety and Pharmacokinetic Analysis

All subjects exposed to ISIS 396443 will be included in the safety analyses.

Treatment duration and amount of Study Drug received will be summarized by dose cohort. Subject incidence rates of all adverse events will be tabulated by Medical Dictionary for Regulatory Activities (MedDRA™) system organ class, and by MedDRA™ term. Tables and/or narratives of "on-study" deaths, serious and significant adverse events, including early withdrawals due to adverse events, will also be provided.

All treatment emergent AEs, all treatment emergent AEs potentially related to Study Drug, all treatment emergent serious AEs, and all treatment emergent serious AEs potentially related to Study Drug will be summarized.

Laboratory tests including chemistry panel, complete blood count with differential, urinalysis, and CSF components will be summarized by study visit for each dose cohort. These safety variables will also be presented over time after Study Drug administration, as appropriate, using results prior to Day 1 dosing as Baseline. Vital sign results will be presented similarly.

Physical and neurological examination findings and results from ECG will be listed for review. As appropriate, results will also be summarized descriptively for each dose cohort. Concomitant medication usage for each subject will be listed for review. Plasma pharmacokinetic parameters and ISIS 396443 concentrations in plasma and CSF for the Pharmacokinetic population will be summarized by dose cohort using descriptive statistics and, where warranted, presented graphically.

11. INVESTIGATOR'S REGULATORY OBLIGATIONS

11.1 Informed Consent

The written informed consent documents should be prepared in the language(s) of the potential subject population, based on an English version provided by the Sponsor and should be easy to understand.

Before a subject's participation in the trial, the Investigator is responsible for obtaining written informed consent from the parent or legal guardian after adequate explanation of the aims, methods, anticipated benefits, and potential hazards of the study and before any protocol-specific screening procedures or any Study Drugs are administered. Sufficient time must be given to consider whether to participate in the study.

The acquisition of informed consent and the parent/legal guardian's agreement or refusal of his/her notification of the primary care physician should be documented in the subject's medical records, and the informed consent form(s) should be signed and personally dated by the parent/legal guardian and by the study person who conducted the informed consent discussion. The original signed informed consent form(s) should be retained in the Study Master File and in any other locations required by institutional policy, and a copy of the signed consent form(s) should be provided to the parent or guardian.

11.2 Ethical Conduct of the Study

The Guidelines of the World Medical Association (WMA) Declaration of Helsinki dated October 2002 the applicable regulations and guidelines of current Good Clinical Practice (GCP) as well as the demands of national drug and data protection laws and other applicable regulatory requirements will be strictly followed.

11.3 Institutional Review Board/Research Ethics Board

A copy of the protocol, proposed informed consent form, other written subject information, and any proposed advertising material must be submitted to the IRB/REB for written approval. A copy of the written approval of the protocol and informed consent form must be received by the Sponsor before recruitment of subjects into the study and shipment of Study Drug. A copy of the written approval of any other items/materials that must be approved by the Study Center or IRB/REB must also be received by the Sponsor before recruitment of subjects into the study and shipment of Study Drug. The Investigator's Brochure must be submitted to the IRB/REB for acknowledgement.

The Investigator must submit to and, where necessary, obtain approval from the IRB/REB for all subsequent protocol amendments and changes to the informed consent documents. The Investigator should notify the IRB/REB of deviations from the protocol in accordance with ICH GCP Section 4.5.2. The Investigator should also notify the IRB/REB of serious adverse events occurring at the Study Center and other adverse event reports received from the Sponsor, in accordance with local procedures.

The Investigator will be responsible for obtaining annual IRB/REB approval/renewal throughout the duration of the study. Copies of the Investigator's reports, all IRB/REB submissions and the IRB/REB continuance of approval must be sent to the Sponsor.

11.4 Subject Confidentiality

The Investigator must ensure that the subject's confidentiality is maintained. On the CRFs or other documents submitted to the Sponsor, subjects should be identified by unique initials and a subject study number only. Documents that are not for submission to the Sponsor (e.g., signed informed consent/assent forms) should be kept in strict confidence by the Investigator.

In compliance with federal and local regulations/ICH GCP Guidelines, it is required that the Investigator and institution permit authorized representatives of the company, of the regulatory agency(s), and the IRB/REB direct access to review the subject's original medical records for verification of study-related procedures and data. Direct access includes examining, analyzing, verifying, and reproducing any records and reports that are important to the evaluation of the study. The Investigator is obligated to inform and obtain the consent of the subject's parent or guardian to permit named representatives to have access to his/her study-related records without violating the confidentiality of the subject.

12. ADMINISTRATIVE AND LEGAL OBLIGATIONS

12.1 Protocol Amendments

Protocol amendments must be made only with the prior approval of the Sponsor. Agreement from the Investigator must be obtained for all protocol amendments and amendments to the informed consent document. The regulatory authority and IRB/REB must be informed of all amendments and give approval for any amendments likely to affect the safety of the subjects or the conduct of the trial. The Investigator **must** send a copy of the approval letter from the IRB/REB to the Sponsor.

12.2 Study Termination

Both the Sponsor and the Investigator reserve the right to terminate the study, according to the terms of the study contract. The Investigator/Sponsor should notify the IRB/REB in writing of the trial's completion or early termination.

12.3 Study Documentation and Storage

The Investigator should ensure that all appropriately qualified persons to whom he/she has delegated trial duties are recorded on a Sponsor-approved Delegation of Site Responsibilities Form.

Source documents are original documents, data, and records from which the subject's CRF data are obtained. These include but are not limited to hospital records, clinical and office charts, laboratory and pharmacy records, imaging, and correspondence. In this study, CRFs may not be used as source documents.

The Investigator and Study Center staff is responsible for maintaining a comprehensive and centralized filing system of all study-related (essential) documentation in accordance with Section 8 of the ICH Guidelines (E6), suitable for inspection at any time by representatives from the Sponsor and/or applicable regulatory authorities. Elements should include:

- Subject files containing completed CRFs, informed consents, and supporting copies of source documentation
- Study files containing the protocol with all amendments, Investigator's Brochure, copies of pre-study documentation and all correspondence to and from the IRB/REB and the Sponsor
- If drug supplies are maintained at the Study Center, proof of receipt, Study Drug Product Accountability Record, Return of Study Drug Product for Destruction, and all drug-related correspondence

In addition, all original source documents supporting entries in the CRFs must be maintained and be readily available.

No study document should be destroyed without prior written agreement between the Sponsor and the Investigator. Should the Investigator wish to assign the study records to another party or move them to another location, he/she must notify the Sponsor.

12.4 Study Monitoring

The Sponsor representative and regulatory authority inspectors are responsible for contacting and visiting the Investigator for the purpose of inspecting the facilities and, upon request, inspecting the various records of the trial (e.g., CRFs and other pertinent data) provided that subject confidentiality is respected.

The Sponsor monitor is responsible for inspecting the CRFs at regular intervals throughout the study to verify adherence to the protocol; completeness, accuracy, and consistency of the data; and adherence to local regulations on the conduct of clinical research. The monitor should have

access to subject medical records and other study-related records needed to verify the entries on the CRFs.

The Investigator agrees to cooperate with the monitor to ensure that any problems detected in the course of these monitoring visits, including delays in completing CRFs, are resolved.

In accordance with ICH GCP and the Sponsor's audit plans, this study may be selected for audit by representatives from the Sponsor's Clinical Quality Assurance Department. Inspection of Study Center facilities (e.g., pharmacy, drug storage areas, laboratories) and review of study-related records will occur to evaluate the trial conduct and compliance with the protocol, ICH GCP, and applicable regulatory requirements.

To ensure the quality of clinical data a clinical data management review will be performed on subject data received by the Sponsor. During this review, subject data will be checked for consistency, omissions, and any apparent discrepancies. In addition, the data will be reviewed for adherence to the protocol and GCP. To resolve any questions arising from the clinical data management review process, data queries and/or Study Center notifications will be sent to the Study Center for completion and return to Sponsor.

The Principal Investigator will sign and date the indicated places on the CRF. These signatures will indicate that the principal Investigator inspected or reviewed the data on the CRF, the data queries, and the Study Center notifications, and agrees with the content.

12.5 Language

CRFs must be completed in English. Generic names for concomitant medications should be recorded in English if possible, unless it is a combination drug, then record the trade name in English.

All written information and other material to be used by subjects and investigative staff must use vocabulary and language that are clearly understood.

12.6 Compensation for Injury

The Sponsor maintains appropriate insurance coverage for clinical trials and will follow applicable local compensation laws. Subjects will be treated and/or compensated for any study-related illness/injury in accordance with the information provided in the Compensation for Injury section of the Informed Consent document.

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14. APPENDICES

Appendix A Schedule of Procedures

Appendix A – Schedule of Procedures for 'Loading' Dosing Period, Visits Only*

*** Note that monitoring telephone calls will also occur on Study Days 8 (±1), 43 (±2), 57 (±2), 71 (±2), 106 (±3), 127 (±3), 134 (±3), 148 (±3), 189 (±3), 197 (±3), 218 (±3), 239 (±3) (i.e., every 2-3 weeks throughout the study)[13]**

Study Period	Screen	Dosing														
Study Day	D -21 to D -1	D1 Injection			D2	D15 (±1D) Injection			D16	D29 (±1D)	D85 (±2D) Injection			D86	D92 (±1D)	D169 (±5D)
		Pre-dose	LP	Post-dose		Pre-dose	LP	Post-dose			Pre-dose	LP	Post-dose			
Study Drug: LP Injection[10]			X				X[9]					X[9]				
In-Patient Stay (24 hours)				X												
Informed Consent	X															
Inclusion/Exclusion Criteria	X															
Medical History	X															
SMN Genetics[1]											X					
Vital Signs[2]	X	X		4X[3]	X[5]	X		4X[3]	X	X	X		4X[3]	X	X	X
Weight	X	X			X				X	X	X			X	X	X
Growth Parameters[4]	X									X					X	X
Physical Examination	X	X				X				X	X				X	X
Ventilator Use	X	X				X				X	X				X	X
Neurological Examination	X	X		2X[6]	X[5]	X		2X[6]	X	X	X		2X[6]	X	X	X
ECG	X									X					X	
Safety Labs[7, 12]	X									X					X	X
Coagulation Labs	X															
Immunogenicity		X									X					X
CSF Safety[12]		X				X					X					
CSF PK[11]		X				X					X					
Plasma PK[11]		X		3X	X						X		X		X	
CHOP INTEND	X[8]									X					X	X
CMAP	X[8]									X					X	X
Con Med Recording	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Adverse Event Collection	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

1 Only for those patients who do not have documented evidence of SMN genetics from Athena Diagnostics

2 Resting blood pressure, pulse, respiratory rate, temperature, and pulse oximetry awake. Pulse oximetry asleep will also be assessed at screening only

3 Vital signs performed 1, 2, 4, 6 hours after dosing

4 Length, weight for age/length, head circumference, chest circumference, head to chest circumference ratio, arm circumference

5 Conducted within 20-24 hours after dosing

6 Neurological exams at 3 and 6 hours after dosing

7 Serum chemistry, hematology, urinalysis panels (see Appendix B for analytes). Cystatin C will not be analyzed at Day 29 due to blood volume limitations

Appendix A – Schedule of Procedures for 'Loading' Dosing Period, Visits Only* *Continued*

Legend Text *Continued*

8 CMAP and CHOP INTEND assessments do not form part of the screening assessment or inclusion/exclusion criteria but are baseline measurements taken during the screening phase of the study
9 Overnight stay is optional on Day 15 and Day 85, if needed due to complications of the LP procedure
10 Injections may not be scheduled within 72 hours after an immunization
11 Refer to Appendix C for PK sampling schedule
12 Refer to Appendix B for analytes
13 At telephone contact, changes in concomitant medications and adverse events will be recorded as well as information on the subject's ventilator use and SMA disease status

Appendix A – Schedule of Procedures for 'Maintenance' Dosing Period, Visits Only*

*** Note that monitoring telephone calls will also occur on Study Days 274 (±3), 295 (±3), 316 (±3), 358 (±3), 400 (±3), 421 (±3), 463 (±3), 484 (±3), 526 (±3), 547 (±3), 589 (±3), 610 (±3), 652(±3), 673 (±3), 715 (±3), 736 (±3), 778 (±3), 799 (±3), 841 (±3), 862 (±3), 904 (±3), 925 (±3), 967 (±3), 988 (±3), 1030 (±3), 1051 (±3), 1093 (±3), 1114 (±3), 1156 (±3), 1177 (±3), 1219 (±3), 1240 (±3), 1282 (±3), 1303 (±3), and 1324 (±3) (i.e., approximately every 3 weeks)[10]**

Study Period	Dosing/Followup										
Study Day	D253 Injection (±7D)			D254	D337 (±7D)	D379, 505, 631, 757, 883, 1009, 1135, and 1261 Injection (±7D)			D380, 506, 632,758, 884, 1010, 1136, 1262	D442, 568, 694, 820, 946, 1072, 1198 (±7D)	D1352 (±7D) or Early Termination (ET)
	Pre-dose	LP	Post-dose			Pre-dose	LP	Post-dose			
Study Drug: LP Injection[7]		X[6]					X[6]				
Vital Signs[1]	X		4X[2]	X	X	X		4X[2]		X	X
Weight	X				X	X				X	X
Growth Parameters[3]	X[11]				X	X[11]				X	X
Physical Examination	X				X	X				X	X
Ventilator Use	X				X	X				X	X
Neurological Examination	X		2X[4]	X	X	X		2X[4]		X	X
ECG					X					X[12]	X
Safety Labs[5, 9]	X[11]				X					X	X
Immunogenicity[13]	X[11]				X					X	X
CSF Safety[9]	X					X					
CSF PK[8]	X					X					
Plasma PK[8]	X[11]					X[11]					X
CHOP INTEND	X[11]				X	X[11]				X	X
HFMSE[14]						X[11]				X	X
CMAP	X[11]				X					X	X
Phone Contact									X		
Con Med Recording	X	X	X	X	X	X	X	X	X	X	X
Adverse Event Collection	X	X	X	X	X	X	X	X	X	X	X

1 Resting blood pressure, pulse, respiratory rate, temperature, and pulse oximetry awake
2 Vital signs performed 1, 2, 4, 6 hours after dosing
3 Length, weight for age/length, head circumference, chest circumference, head to chest circumference ratio, arm circumference
4 Neurological exams at 3 and 6 hours after dosing
5 Serum chemistry, hematology, urinalysis panels (see Appendix B for analytes)
6 Overnight stay is optional on Days 253, 379, 505, 631, 757, 883, 1009, 1135, and 1261 if needed due to complications of the LP procedure
7 Injections may not be scheduled within 72 hours after an immunization
8 Refer to Appendix C for PK sampling schedule
9 Refer to Appendix B for analytes
10 At telephone contact, changes in concomitant medications and adverse events will be recorded as well as information on the subject's ventilator use and SMA disease status
11 These assessments can be performed up to 7 days prior to dosing, if necessary

Appendix A – Schedule of Procedures for 'Maintenance' Dosing Period, Visits Only* *Continued*

Legend Text *Continued*

12 ECG should be performed on Day 568 only

13 Immunogenicity assessments will be performed as deemed necessary on samples collected on other days

14 All subjects who have maintained a CHOP INTEND total score of ≥ 50 for 2 consecutive study visits will be evaluated using the Hammersmith Functional Motor Scale Expanded (HFMSE) in addition to CHOP INTEND

Appendix B Laboratory Analytes

Appendix B Laboratory Analytes

CLINICAL SAFETY ASSESSMENTS *(minimum requirements)*		OTHER ASSESSMENTS
Clinical Chemistry	***Urinalysis***	****PK***
Sodium	*Specific gravity*	*Plasma ISIS 396443 levels*
Potassium	*pH*	*CSF ISIS 396443 levels*
Chloride	*Protein*	
Total protein	*Glucose*	***SMN Genetics***
Albumin	*Ketones*	*SMN2 Copy Number*
Calcium	*Bilirubin*	*SMN Gene Sequencing*
Phosphorus	*Blood*	
Bicarbonate	*Red blood cells*	
Glucose	*White blood cells*	
BUN	*Epithelial cells*	
Creatinine	*Bacteria*	
Cystatin C	*Casts*	
Total serum Bilirubin	*Crystals*	
Alkaline phosphatase		
AST (SGOT)	***Hematology***	
ALT (SGPT)	*Red blood cells*	
CPK	*Hemoglobin*	
	Hematocrit	
Coagulation	*Platelets*	
aPTT	*White blood cells*	
PT	*WBC Differential*	
INR	• *(% and absolute)*	
	• *Neutrophils*	
Immunogenicity Evaluation	• *Eosinophils*	
ISIS 396443 plasma Abs	• *Basophils*	
	• *Lymphocytes*	
CSF Safety (Minimum Requirements)	• *Monocytes*	
Red blood cells		
White blood cells		
Glucose		
Protein		

***** Any of the collected PK plasma and CSF samples from the study patients may also be used for investigation of possible biomarkers of SMA disease or the pharmacodynamic effects of ISIS 396443 (e.g., CSF mRNA, CSF protein, CSF miRNA panel) or for profiling of drug binding proteins, bioanalytical method validation purposes, stability assessments, metabolite assessments, immunogenicity assessments (including assay development and validation purposes), or to assess other actions of ISIS 396443 with plasma and CSF constituents

Appendix C Pharmacokinetic Sampling Schedule

Appendix C Pharmacokinetic Sampling Schedule

Treatment Period	Study Day	Timepoints	Blood Collection	CSF Collection
Multiple Dose: LP Injection	D1	Predose	0.35 mL	0.5 mL
		1 hr	0.35 mL	NA
		2 hr	0.35 mL	NA
		4 hr	0.35 mL	NA
	D2	24 hr	0.35 mL	NA
	D15	Predose	NA	0.5 mL
	D85	Predose	0.35 mL	0.5 mL
		4 hr	0.35 mL	NA
	D92	Anytime	0.35 mL	NA
	D253	Predose	0.35 mL	0.5 mL
	D379	Predose	0.35 mL	0.5 mL
	D505	Predose	0.35 mL	0.5 mL
	D631	Predose	0.35 mL	0.5 mL
	D757	Predose	0.35 mL	0.5 mL
	D883	Predose	0.35 mL	0.5 mL
	D1009	Predose	0.35 mL	0.5 mL
	D1135	Predose	0.35 mL	0.5 mL
	D1261	Predose	0.35 mL	0.5 mL
	D1352/ET	Anytime	0.35 mL	NA

NA Not applicable (No Collection Scheduled)

Details on sampling, preparation, and shipment are included in the study laboratory manual

Any of the collected PK plasma and CSF samples from the study patients may also be used for investigation of possible biomarkers of SMA disease or the pharmacodynamic effects of ISIS 396443 (e.g., CSF mRNA, CSF protein, CSF miRNA panel) or for profiling of drug binding proteins, bioanalytical method validation purposes, stability assessments, metabolite assessments, immunogenicity assessments (including assay development and validation purposes), or to assess other actions of ISIS 396443 with plasma and CSF constituents

ISIS PHARMACEUTICALS, INC.

ISIS 396443-CS3A

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment 5 – 29 May 2015

Sponsor:

Isis Pharmaceuticals, Inc.

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ISIS 396443-CS3A

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment 5 – 29 May 2015

Protocol History:

Original Protocol:	4 March 2013
Amendment 1:	14 August 2013
Amendment 2:	18 January 2014
Amendment 3:	14 March 2014
Amendment 4:	19 May 2014

Sponsor:

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ISIS 396443

Isis Protocol Number ISIS 396443-CS3A

Protocol Amendment 5

Clinical Phase: 2

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

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Date: 29 May 2015

Confidentiality Statement

Protocol Signature Page

Protocol Number:	ISIS 396443-CS3A
Protocol Title:	A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy
Amendment:	Amendment 5
Date:	29 May 2015

I hereby acknowledge that I have read and understand the attached clinical protocol, entitled "A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy", dated 29 May 2015, and agree to conduct the study as described herein.

I agree to comply with the International Conference on Harmonization Tripartite Guideline on Good Clinical Practice (E6).

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of Isis Pharmaceuticals, Inc.

Investigator's Signature

Investigator's Name *(please print)*

Date (DD Month YYYY)

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PROTOCOL AMENDMENT

Protocol Number: ISIS 396443-CS3A

Protocol Title: A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment: 5

Amendment Date: 29 May 2015

The purpose of this amendment is to (1) extend the 'maintenance' dosing treatment period by adding 4 doses of 12 mg equivalent of ISIS 396443 given every 4 months (i.e., on Study Days 883, 1009, 1135, and 1261); (2) add the Hammersmith Functional Motor Scale-Expanded as an outcomes measure for all subjects who have maintained a CHOP INTEND total score of ≥ 50 for two consecutive study visits; (3) add a ready-to-use vial configuration of the Study Drug; (4) add language on the possibility to perform an interim analysis to support potential discussions with regulatory agencies. The following table provides a summary list of changes to the protocol:

Section	Description of Change	Justification
Protocol Synopsis **2.4 Rationale for Dose and Schedule of Administration** **3. Experimental Plan** **6. Study Procedures** **Appendix A** **Appendix C**	Maintenance dosing extended to add 4 additional doses of 12 mg equivalent of ISIS 396443 given once every 4 months. Additional telephone contact assessments (approximately once every 3 weeks) have been added to accommodate the extension of the treatment period.	Due to the severe, life-threatening nature of infantile-onset SMA, and the encouraging risk-benefit profile of ISIS 396443 observed in this patient population in early parts of the study, the treatment duration is being extended (including additional study visit and telephone contact assessments).
2.3.4 Clinical Experience	Update clinical section with respect to Phase 2 study status.	This section was updated to list all ongoing studies.
Protocol Synopsis **6.2 Study Assessments** **10.1.3 Exploratory Endpoints** **13. References** **Appendix A**	Hammersmith Functional Motor Scale Expanded (HFMSE) was added as an exploratory endpoint for subjects who have maintained a CHOP INTEND total score of ≥ 50 for two consecutive study visits. Two (2) references on the HFMSE were added.	The HFMSE is added to allow capturing functional improvements seen in stronger infants who have started to reach the ceiling of the CHOP INTEND.
Protocol Synopsis **7. Study Drug** **8.1 Study Drug Administration**	Added information on the two types of Study Drug supply: (1) a two-vial configuration in which Study Drug is at a concentration of 20 mg/mL which is diluted prior to use, and (2) a ready-to-use vial configuration in which the Study Drug is at a concentration of 2.4 mg/mL.	As the study progresses, the transition will be made from the two-vial configuration to the ready-to-use vial configuration for ease of administration.
Protocol Synopsis **10.4 Interim Analysis**	Added language to allow for an interim analysis.	The possibility to perform an interim analysis was added to support potential discussions with regulatory agencies.

ISIS PHARMACEUTICALS, INC.

ISIS 396443-CS3A

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment 4 – 19 May 2014

Sponsor:

Isis Pharmaceuticals, Inc.

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ISIS 396443-CS3A

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment 4 – 19 May 2014

Protocol History:

Original Protocol:	4 March 2013
Amendment 1:	14 August 2013
Amendment 2:	18 January 2014
Amendment 3:	14 March 2014

Sponsor:

PPD

ISIS 396443

Isis Protocol Number ISIS 396443-CS3A

Protocol Amendment 4

Clinical Phase: 2

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

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Date: 19 May 2014

Confidentiality Statement

Protocol Signature Page

Protocol Number:	ISIS 396443-CS3A
Protocol Title:	A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy
Amendment:	Amendment 4
Date:	19 May 2014

I hereby acknowledge that I have read and understand the attached clinical protocol, entitled "A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy", dated 19 May 2014, and agree to conduct the study as described herein.

I agree to comply with the International Conference on Harmonization Tripartite Guideline on Good Clinical Practice (E6).

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of Isis Pharmaceuticals, Inc.

Investigator's Signature

Investigator's Name *(please print)*

Date (DD Month YYYY)

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PROTOCOL AMENDMENT

Protocol Number: ISIS 396443-CS3A

Protocol Title: A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment: 4

Amendment Date: 19 May 2014

The purpose of this amendment is to extend maintenance dosing with ISIS 396443 by adding 4 additional doses of 12 mg equivalent given every 4 months (i.e., on Study Days 379, 505, 631, and 757).

The following table provides a summary list of changes to the protocol:

Section	Description of Change	Justification
Protocol Synopsis, **3. Experimental Plan**, **6.2 Study Procedures**, **8. Treatment of Subjects** **Appendix A**, **Appendix C**	Maintenance dosing extended to add 4 additional doses of 12 mg equivalent of ISIS 396443 given once every 4 months	Due to the severe, life-threatening nature of infantile-onset SMA, and the encouraging risk-benefit profile of ISIS 396443 observed in this patient population in the early part of the study, the treatment duration is being extended
2.3.3 Preclinical Experience	Detailed results of preclinical studies replaced with a summary statement and the relevant sections of the Investigator Brochure are referenced	Investigator Brochure has been updated with results of the long-term toxicology study in juvenile monkeys
2.4 Rationale for Dose and Schedule of Administration	Rationale for loading and maintenance dose and regimen has been updated	Dose and dosing regimen selection have been justified on the basis of available clinical and preclinical PK data
8.1 Study Drug Administration	Row added for study drug injection volume in subjects >24 months of age	Necessary to accommodate extension of the treatment period

ISIS PHARMACEUTICALS, INC.

ISIS 396443-CS3A

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment 3 – 14 March 2014

Sponsor:

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ISIS 396443-CS3A

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment 3 – 14 March 2014

Protocol History:

Original Protocol: 4 March 2013

Amendment 1: 14 August 2013

Amendment 2: 18 January 2014

Sponsor:

PPD

ISIS 396443

Isis Protocol Number ISIS 396443-CS3A

Protocol Amendment 3

Clinical Phase: 2

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

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Date: 14 March 2014

Confidentiality Statement

Protocol Signature Page

Protocol Number:	ISIS 396443-CS3A
Protocol Title:	A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy
Amendment:	Amendment 3
Date:	14 March 2014

I hereby acknowledge that I have read and understand the attached clinical protocol, entitled "A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy", dated 14March 2014, and agree to conduct the study as described herein.

I agree to comply with the International Conference on Harmonization Tripartite Guideline on Good Clinical Practice (E6).

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of Isis Pharmaceuticals, Inc.

Investigator's Signature

Investigator's Name *(please print)*

Date (DD Month YYYY)

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PROTOCOL AMENDMENT

Protocol Number: ISIS 396443-CS3A

Protocol Title: A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment: 3

Amendment Date: 14 March 2014

The purpose of this amendment is to add permanent ventilation to the combined efficacy endpoint. The definition of permanent ventilation is ≥16 hours ventilation/day continuously for >2 weeks in the absence of an acute reversible illness. This definition is consistent with what was done in Natural History Study data in the infantile SMA patient population (PNCR study, Finkel et al., unpublished data).

The following table provides a summary list of changes to the protocol:

Section	Description of Change
Synopsis; **10.1.3 Exploratory Endpoints**	Changed efficacy endpoint to: Age at death or permanent ventilation (defined as ≥16 hours ventilation/day continuously for >2 weeks in the absence of an acute reversible illness), to be consistent with natural history study data in the infantile SMA population (PNCR study, Finkel et al.).

ISIS PHARMACEUTICALS, INC.

ISIS 396443-CS3A

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment 2 – 18 January 2014

Sponsor:

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ISIS 396443-CS3A

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment 2 – 18 January 2014

Protocol History:

Original Protocol: 4 March 2013

Amendment 1: 14 August 2013

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ISIS 396443

Isis Protocol Number ISIS 396443-CS3A

Protocol Amendment 2

Clinical Phase: 2

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

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Date: 18 January 2014

Confidentiality Statement

Protocol Signature Page

Protocol Number:	ISIS 396443-CS3A
Protocol Title:	A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy
Amendment:	Amendment 2
Date:	18 January 2014

I hereby acknowledge that I have read and understand the attached clinical protocol, entitled "A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy", dated 18 January 2014, and agree to conduct the study as described herein.

I agree to comply with the International Conference on Harmonization Tripartite Guideline on Good Clinical Practice (E6).

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of Isis Pharmaceuticals, Inc.

Investigator's Signature

Investigator's Name *(please print)*

Date (DD Month YYYY)

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PROTOCOL AMENDMENT

Protocol Number: ISIS 396443-CS3A

Protocol Title: A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment: 2

Amendment Date: 18 January 2014

The purpose of this amendment is to extend the study to add one additional 'maintenance' dose of 12 mg equivalent ISIS 396443 on Study Day 253, approximately 6 months after the subjects have received their Day 85 dose. A few typographical errors were also corrected for consistency.

The following table provides a summary list of changes to the protocol:

Section	Description of Change
PROTOCOL SYNOPSIS, Study Design and Methodology	Added 'maintenance' dose treatment and follow-up to the study design
2.4 Rationale for Dose and Schedule of Administration	Added dose rationale for the single 'maintenance' dose
3. EXPERIMENTAL PLAN	Added 'maintenance' dose treatment and follow-up to the study design
6.2 Study Assessments	Added 'maintenance' dose treatment and follow-up to the study assessments
8.1 Study Drug Administration	Added study drug administration instructions for the 'maintenance' dose
8.7 Adjustment of Dose and/or Treatment Schedule	Added instructions for the 'maintenance' dose
APPENDIX A Schedule of Procedures	Added 'maintenance' dose treatment and follow-up to the schedule of procedures
APPENDIX C Pharmacokinetic Schedule	Added pharmacokinetic sample schedule for the 'maintenance' dose period

ISIS PHARMACEUTICALS, INC.

ISIS 396443-CS3A

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment 1 – 14 August 2013

Sponsor:

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ISIS 396443-CS3A

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment 1 – 14 August 2013

Protocol History:

Original Protocol: 4 March 2013

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ISIS 396443

Isis Protocol Number ISIS 396443-CS3A

Protocol Amendment 1

Clinical Phase: 2

A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

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Date: 14 August 2013

Confidentiality Statement

Protocol Signature Page

Protocol Number:	ISIS 396443-CS3A
Protocol Title:	A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy
Amendment:	Amendment 1
Date:	14 August 2013

I hereby acknowledge that I have read and understand the attached clinical protocol, entitled "A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy", dated 14 August 2013, and agree to conduct the study as described herein.

I agree to comply with the International Conference on Harmonization Tripartite Guideline on Good Clinical Practice (E6).

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of Isis Pharmaceuticals, Inc.

Investigator's Signature

Investigator's Name *(please print)*

Date (DD Month YYYY)

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PROTOCOL AMENDMENT

Protocol Number: ISIS 396443-CS3A

Protocol Title: A Study to Assess the Safety, Tolerability, and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Patients with Infantile-Onset Spinal Muscular Atrophy

Amendment: 1

Amendment Date: 14 August 2013

The purpose of this amendment is to: 1) change the dose from 9 to 12 mg ISIS 396443; 2) add additional subjects to the study, such that Cohort 2 now may enroll between 4 to approximately 16 subjects; 3) add age at death and age at death or ≥16 hours ventilation/day for >2 weeks as exploratory endpoints; and 4) update the clinical experience section, specifically regarding the ISIS 396443-CS2 and ISIS 396443-CS12 studies.

The following table provides a summary list of changes to the protocol:

Section	Description of Change
2.3.4 Clinical Experience	Updated information regarding ISIS 396443-CS2 and ISIS 396443-CS12 studies
2.4 Rationale for Dose and Schedule of Administration	Changed “9 mg ISIS 396443” to “12 mg ISIS 396443” throughout the section and added rationale for 12 mg dose
3.3 Number of Subjects	For Cohort 2 changed “4” to “4 to approximately 16” subjects to allow additional subjects to enroll in this study prior to when the anticipated Phase 3 study in Infantile-onset SMA commences
8.1 Study Drug Administration	Updated dose and concentration in Table 2 and Table 3 for 12 mg dosing
10.2 Sample Size Considerations	Changed “sample size of 8” to “sample size of approximately 20”
10.1.3 Exploratory Endpoints	Added age at death and age at death or ≥16 hours ventilation/day for >2 weeks as exploratory endpoints