Statistical Analysis Plan

ISIS 420915-CS2

A Phase 2/3 Randomized, Double-Blind, Placebo-Controlled Study to Assess the Efficacy and Safety of ISIS 420915 in Patients with Familial Amyloid Polyneuropathy

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Statistical Analysis Plan Signature Page

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Protocol:	ISIS 420915-CS2
Study Title:	A Phase 2/3 Randomized, Double-Blind, Placebo-Controlled Study to Assess the Efficacy and Safety of ISIS 420915 in Patients with Familial Amyloid Polyneuropathy
Issue Date:	May 13, 2016 (Amendment 9)

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Abbreviations

Abbreviation/Acronym	Definition
+7	Sum 7 test. Includes measurements of nerve conduction, vibration threshold, and heart rate to deep breathing
PPD	PPD
ADA	Anti-ISIS 420915 antibodies
AE	Adverse event
AESI	Adverse event of special interest
ANCOVA	Analysis of covariance
ASO	Antisense oligonucleotide
AUC	Area under the curve
BMI	Body mass index
C_{max}	Maximum observed plasma concentration; obtained directly from the plasma concentration-time profile
CM-ECHO	Cardiomyopathy-ECHO
CI	Confidence interval
CICL	Cardiac Imaging Core Laboratory
CIR	Copy Increment from Reference
CL	Plasma Clearance
CRF	Case report form
CRO	Contract research organization
C-SSRS	Columbia Suicide Severity Rating Scale
CV %	Coefficient of variation, expressed as a percent
DIC	Disseminated intravascular coagulation
DSMB	Data and safety monitoring board
ECG	Electrocardiogram
ECHO	Echocardiogram
ECHO Subgroup	The subgroup of patients having additional echocardiogram assessments
eCRF	Electronic case report form
EDC	Electronic data capture
EOS	End of study
EOT	End of treatment
ERG	Electroretinogram
FAC	Familial amyloid cardiomyopathy

FAP	Familial amyloid polyneuropathy
FAS	Full Analysis Set
GCP	Good Clinical Practice
GLS	Global longitudinal strain
HLT	Higher level term
hATTR-CM	Hereditary transthyretin amyloidosis with cardiomyopathy
hATTR-PN	Hereditary transthyretin amyloidosis with polyneuropathy
HP	Heat Pain
HRDB	Heart Rate to Deep Breathing
ICH	International conference on harmonization
IEC	Independent ethics committee
IM	Immunogenicity
IRB	Institutional review board
IVRS	Interactive voice-response system
J2R	Jump to Reference
LCRIS	Local cutaneous reaction at the injection site
LLQ	Lower limit of quantification
MAR	Missing at Random
mBMI	Modified body mass index (requires determination of serum albumin levels; mBMI = BMI x serum albumin g/L)
PPD	PPD
MedDRA	Medical Dictionary for Regulatory Activities
MMRM	Mixed effects model with repeated measures
mNIS+7	Modified neuropathy impairment Score +7. Standard NIS but with modifications made to the +7 component
Modified +7	+7 test with modifications made to the sensory and nerve conduction testing
MRT	Mean residence time
NCT	Nerve Conduction Test
NIS	Neuropathy impairment score
NIS+7	Neuropathy impairment score +7
NIS-C	Neuropathy impairment score – Cranial Nerves
NIS-R	Neuropathy impairment score – Reflexes
NIS-S	Neuropathy impairment score – Sensation
NIS-W	Neuropathy impairment score – Muscle Weakness

Norfolk QOL-DN	Norfolk quality of life questionnaire-diabetic neuropathy
NSC Score	Neuropathy symptoms and change score
NT-proBNP	N-terminal prohormone of brain natriuretic peptide
OLE	Open label extension
P25	25^{th} percentile
P75	75^{th} percentile
PD	Pharmacodynamic
PK	Pharmacokinetic
PKS	Pharmacokinetic set
PND Score	Polyneuropathy disability score
PPS	Per Protocol Set
QTc	QT interval corrected for heart rate
QTcF	QTc interval calculated using Fridericia's formula
RBP4	Retinol binding protein 4
SAE	Serious adverse event
SAP	Statistical analysis plan
SC	Subcutaneous
SF-36	Short Form (36) Health Survey
SMQ	Standardized MedDRA query
SOC	System Organ Class
SS	Safety set
Study Day 1	Defined as the first day Study Drug is administered to the patient
Study Drug	ISIS 420915 or placebo
SUSARs	Suspected unexpected serious adverse reactions
TEAE	Treatment emergent adverse event
T_{max}	Time to reach C_{max}; obtained directly from the plasma concentration-time profile
TP	Touch Pressure
TTP	Thrombotic thrombocytopic purpura
TTR	Transthyretin
ULN	Upper limit of normal
VDT	Vibration Detection Threshold
WMA	World Medical Association

1 Introduction

This statistical analysis plan describes the final reporting for study ISIS 420915-CS2 as per protocol amendment #9. It also describes the interim reporting to support the Data Safety Monitoring Board (DSMB) reviews and the interim analysis. Any changes from these planned analyses will be stated in the clinical study report.

1.1 Study Overview

Transthyretin (TTR) is synthesized primarily in the liver and is secreted into the plasma as a 55 KD protein composed of four identical subunits of 14 KD each. Transthyretin amyloidosis is a rare hereditary disease caused by mutations in the TTR protein. The disease-causing mutations destabilize the normal tetrameric structure of TTR causing it to aggregate and deposit as insoluble fibril deposits in multiple tissues. These deposits result in local damage to cells leading to a peripheral polyneuropathy (called Familial Amyloid Polyneuropathy or FAP) and a cardiomyopathy (called Familial Amyloid Cardiomyopathy or FAC). In the literature FAP has been referred to as hereditary transthyretin amyloidosis with polyneuropathy (hATTR-PN) and FAC as hereditary transthyretin amyloidosis with cardiomyopathy (hATTR-CM).

The main clinical manifestations of FAP are progressive peripheral sensorimotor and autonomic neuropathy. Death, on average, occurs within 10 years from symptom onset and is primarily due to malnutrition and cachexia, cardiac disease, sudden death or renal failure (Coelho et al. 2008). FAP can be classified into 3 stages of disease based on ambulatory status: Stage 1 – do not require assistance with ambulation; Stage 2 – require assistance with ambulation; Stage 3 – wheelchair or bed bound (Coutinho et al. 1980). The total worldwide prevalence of FAP has been estimated at approximately 10,000 patients (Coelho et al. 2008).

ISIS 420915 is an antisense drug targeted to human TTR mRNA and its hybridization to the cognate TTR mRNA results in the RNase H-mediated degradation of the TTR mRNA, thus preventing production of the TTR protein.

ISIS 420915-CS2 is a randomized, double-blind, placebo-controlled, multicenter study of approximately 15 months treatment duration and 6 months follow-up. Approximately 135 patients will be randomized in a 2:1 ratio (90 ISIS 420915 and 45 placebo) to receive 300 mg ISIS 420915 or placebo. Study Drug (ISIS 420915 or placebo) will be administered three times on alternate days during Week 1 (Days 1, 3 and 5), and then once weekly during Weeks 2–65 (for a total of 67 doses). The end-of-treatment (EOT) efficacy assessment is conducted at Week 66. Following treatment and the EOT efficacy assessment, eligible patients (including patients who received placebo) may elect to enroll in an open-label extension (OLE) study pending study approval by the IRB/IEC and the appropriate regulatory authority. All participating patients in the OLE study will receive 300 mg ISIS 420915 once weekly. Otherwise, patients will enter the 6 month post-treatment evaluation portion of the study.

Patients in the study will be Stage 1 (approximately 50%) and Stage 2 (approximately 50%) FAP patients with the following characteristics:

a. Neuropathy impairment score (NIS) score ≥ 10 and ≤ 130

b. Documented transthyretin variant by genotyping

c. Documented amyloid deposition by biopsy

Interim safety data will be reviewed regularly by the ISIS 420915-CS2 DSMB.

A pharmacodynamic interim analysis of reduction in plasma TTR level will be performed by an independent statistician and reviewed by the DSMB after approximately 45 patients have completed the Week 13 visit. This interim will be a futility analysis and will result in a decision to continue the study as planned or to stop the study.

1.2 Objectives

1.2.1 Primary Objective

The primary objective of this study is to evaluate the efficacy of ISIS 420915 as compared to placebo, given for 65 weeks, as measured by the change from baseline in the modified Neuropathy Impairment Score +7 (mNIS+7) and in the Norfolk Quality of Life-Diabetic Neuropathy (Norfolk QOL-DN) questionnaire total score in patients with Familial Amyloid Polyneuropathy.

1.2.2 Secondary Objectives

To evaluate the efficacy of ISIS 420915 as compared to placebo based on the change from baseline in the following measures:

- Norfolk QOL-DN questionnaire symptoms domain score in Stage 1 patients and Norfolk QOL-DN questionnaire physical functioning / large fiber neuropathy domain score in Stage 2 patients
- Modified body mass index (mBMI) and body mass index (BMI)
- Neuropathy impairment score (NIS) and modified +7
- Neuropathy impairment score +7 (NIS+7)
- Global longitudinal strain (GLS) by echocardiogram (ECHO) in the ECHO subgroup and in the Cardiomyopathy-ECHO (CM-ECHO) Set

To evaluate the pharmacodynamic (PD) effect of ISIS 420915 as compared to placebo, based on the change from baseline in TTR and retinol binding protein 4 (RBP4)

To evaluate the safety and tolerability of ISIS 420915

To evaluate the plasma trough levels of ISIS 420915 in all patients and to evaluate the plasma pharmacokinetic (PK) parameters of ISIS 420915 in a subset of patients.

1.2.3 Tertiary Objectives

The tertiary objectives are to evaluate the change from baseline, as compared to placebo, in the following measures:

- SF-36 questionnaire
- Individual components of NIS, modified +7, and +7
- +7
- Individual domain scores of the Norfolk QOL-DN.

1.2.4 Exploratory Objectives

The exploratory objectives are to evaluate the change from baseline, as compared to placebo, in the following exploratory biomarkers:

- ECHO parameters (except GLS)
- Plasma N-terminal prohormone of brain natriuretic peptide (NT-proBNP)
- Polyneuropathy disability score (PND)
- Neuropathy symptoms and change (NSC) score.

1.3 Hypotheses

The strategy of treating FAP patients with ISIS 420915 is to reduce the levels of mutated and wild-type TTR protein secreted by the liver, a primary organ for antisense oligonucleotide (ASO) distribution after systemic delivery. It is predicted that decreasing the amount of liver-derived TTR protein circulating in the plasma by treatment with ISIS 420915 will result in a decrease in the formation of TTR amyloid fibril deposits, and thus slow or halt disease progression (as measured by the mNIS+7) and maintain or improve quality of life (as measured by the Norfolk QOL-DN).

The two primary endpoints (mNIS+7 and Norfolk QOL-DN questionnaire total score) will be tested using a ranking strategy with the mNIS+7 tested first and the Norfolk QOL-DN tested second. The null hypothesis is that there is no difference between ISIS 420915 and placebo in the change from baseline to Week 66 on the mNIS+7, as evaluated by a repeated measures mixed model analysis. Since the Norfolk QOL-DN will not be tested unless the mNIS+7 is significant, the Norfolk QOL-DN is not involved in formulating the null hypothesis (since under the null, it will not be tested). Should the null hypothesis for the mNIS+7 be rejected, then the null hypothesis for the Norfolk QOL-DN questionnaire total score will be tested. However, if the null hypothesis for the mNIS+7 is not rejected, testing for the Norfolk QOL-DN questionnaire total score will be considered exploratory. No adjustment will be made for multiple testing; both endpoints will be tested at an alpha of 0.05. The primary study hypotheses will be formally evaluated at the time of the end of treatment lock (EOT analysis), occurring after all patients have completed the EOT assessments, the database is locked and treatment code is unblinded.

1.4 Endpoints

The primary efficacy endpoints are the change from baseline to Week 66 in the modified Neuropathy Impairment Score +7 (mNIS+7) and in the Norfolk Quality of Life-Diabetic Neuropathy (Norfolk QOL-DN) questionnaire total score. Details describing the mNIS+7 endpoint and scoring can be found in the mNIS+7 Quality Manual. Other study endpoints are described in Sections 3.4 through 3.8.

2 Procedures

2.1 General Overview of Procedures

The study consists of the following periods:

- <6 week screening and baseline assessment period
- 65 week treatment period
- 1 week EOT efficacy assessment period
- 6 month post-treatment evaluation period (unless patients elect to enroll in an OLE study in which case they will not participate in the post-treatment evaluation period)

Screening assessments include safety and specialty labs, physical exam, vital signs, electrocardiogram (ECG), and body weight. A NIS assessment is also performed that is used solely for patient eligibility purposes.

The primary endpoints (mNIS+7 and Norfolk QOL-DN questionnaire) are measured at baseline, Week 35, and Week 66 (EOT efficacy assessment). Patients who do not enroll in the OLE will also have efficacy assessments measured at Week 91 in the post-treatment evaluation period. The mNIS+7 assessments at baseline and Week 66 are performed twice and the duplicates averaged. The duplicates cannot be performed on the same day. In addition, patients who terminate treatment early for any reason will have the mNIS+7 (2x) and Norfolk QOL-DN questionnaire conducted within 14 days from the last dose of Study Drug.

Other secondary and exploratory efficacy measurements (including the pharmacodynamic measures TTR and RBP4, BMI, mBMI, and NT-proBNP) are measured at baseline, at the end of treatment (Week 65) and periodically throughout the treatment period. The SF-36 questionnaire and PND score are measured at baseline, Week 35, and Week 65.

In addition to ECHOs conducted at Baseline, early termination, and Week 65 for all patients, patients who qualify and consent to participate in the ECHO subgroup will have an additional ECHO conducted during the treatment period at Week 41, which can be done at Week 47 if the patient elects to have a Home Healthcare visit at Week 41. All ECHO assessments have a window of ± 2 weeks.

Patients participating in the PK subgroup will have additional blood draws on Day 1, Week 35, and Week 65 to evaluate pharmacokinetic and other safety parameters. PK subgroup patients will also have additional visits to collect 24 hr, 3 day, and 7 day post-dose blood draws.

Safety labs, adverse events (AEs), concomitant medications, ECGs, electroretinograms (ERGs), Ophthalmology examination, physical examination, vital signs, and specialty labs, etc. are collected periodically throughout the baseline, treatment, and post-treatment evaluation periods.

2.2 Randomization & Treatment Allocation

Almac Clinical Technologies is responsible for providing the randomization for this study. Patients will be randomized after all screening assessments have been completed and after the Investigator has verified that they are eligible. No patient may begin treatment prior to randomization and assignment of a unique subject identification number.

Using an Interactive Voice-Response System (IVRS), eligible patients will be randomized 2:1 to receive ISIS 420915 or placebo, respectively. There will be 2 separate and independent randomizations, one for patients in the PK subgroup (approximately 20) and one for patients who are not in the PK subgroup (approximately 115). Within each subgroup, randomization will use a permuted block schedule using the 8 combinations of the following 3 binary stratification variables:

- Previous treatment with Vyndaqel® or Diflunisal versus no known previous treatment
- Stage 1 versus Stage 2 disease
- V30M TTR mutation versus non-V30M TTR mutation

A description of the randomization process, including block size and the procedure to follow will be provided in a separate document prepared by Almac Clinical Technologies.

The randomization list will be generated prior to enrollment of any patients. The Sponsor Quality Assurance department or designee will hold a copy of the randomization lists generated by the IVRS vendor.

2.3 Conduct

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

2.4 Data Monitoring

2.4.1 Safety Data Monitoring

Ionis Pharmaceuticals, Inc. (or designee) is responsible for processing all reported AEs. All serious adverse events (SAEs), reported to Ionis Pharmaceuticals, Inc. (or designee), are reviewed according to standard operating procedures. The medical monitor will review all AEs and SAEs on an ongoing

basis throughout the study. Ionis Pharmaceuticals, Inc. (or designee) will prepare and submit safety reports to the health authorities worldwide in accordance with local requirements. If it becomes necessary to communicate new safety information, Ionis Pharmaceuticals, Inc. (or designee) will also prepare a safety notification letter and transmit it to all applicable study sites.

2.4.2 Data and Safety Monitoring Board

A Data and Safety Monitoring Board (DSMB) will be convened for the following purposes:

- To review safety and tolerability data collected during the trial (frequency of review is dependent on patient enrollment, information accumulated, and safety event rates but will occur no less than approximately every 6 months); and
- To review results of the predetermined TTR interim analysis (details of the analysis and controlled access to data are outlined in Sections 5.1 and 6.0).

Data summaries and listings will present by treatment group in an unblinded fashion and will be prepared by an independent statistican. No statistical comparisons across treatment groups will be provided, and no hypothesis testing will be done.

Further detail on the DSMB meeting schedule, assessments to be reviewed, flow diagram for the interim analysis, and controlled access to data are outlined in the DSMB Charter.

2.5 Data Management

2.5.1 Case Report Form Data

PPD is responsible for creating the Electronic Data Capture (EDC) data entry screens, database and edit checks using definitions developed by Ionis Pharmaceuticals, Inc.. Ionis Pharmaceuticals, Inc. is responsible for the review, data management querying and locking of the database.

Data are single-entered into the EDC system by the investigator site staff. Programmed edit checks (computer logic that checks the validity of the data entered and also prompts for missing data that are expected to be entered) are run and automatic queries are generated. Ionis Pharmaceuticals, Inc. reviews all data for accuracy and validity and generates additional queries in the EDC system when necessary. Exceptions are the mNIS+7, Norfolk QOL-DN, SF-36, NSC, PND, and body weight information that is entered into the EDC but firewalled from review by Ionis Pharmaceuticals, Inc. The data are corrected or an explanation concerning the query is provided in the EDC system. For the purpose of pre-programming and data cleaning, Parexel (the contract research organization (CRO) performing the statistical analysis) and a firewalled data manager working at the CRO contracted to perform data management (PPD) will have access to the post-baseline mNIS+7, NSC, Norfolk-QOL-DN, SF-36, PND and body weight. After all data are entered, reviewed, and queried, the database is closed and sent to the statistics group for review and for identification of protocol deviations. After any further queries that arose from this review are resolved, the database is locked. Database closing and locking will be done after all patients have completed the EOT assessments (the primary analysis) and again after all subjects complete the

study, referred to as end of study (EOS). The study will be unblinded after the first lock (EOT analysis). Details can be found in the Data Management Plan.

2.5.2 Laboratory Data

Ionis Pharmaceuticals, Inc. is responsible for the format of the laboratory electronic data transfers and the transfer schedule. Ionis Pharmaceuticals, Inc. is responsible for the review of the clinical laboratory data. Central laboratory data are not stored in the EDC system. Investigator sites have access to the data via lab reports sent directly from the laboratory or through the laboratory's web portal (in which case Investigators only have access to data from their site). In order to ensure maintenance of the study blind, post-treatment TTR (also called pre-albumin), RBP4, retinol (also called vitamin A), hsCRP, and NT-proBNP values will not be available to the Sponsor, monitors, Investigators, Study Center Personnel, or the patients until after EOT data base lock and the blind is broken.

2.5.3 Pharmacokinetics Data

Ionis Pharmaceuticals, Inc. is responsible for the management and review of the PK data. This process involves reviewing the patient and visit identifiers with the clinical data collected in the EDC system. The PK data are not stored in the EDC system. Prior to unblinding of the final analysis, any ISIS 420915 concentration data sets provided to Ionis by the bioanalytical lab will be provided without reference to actual patient identifiers to avoid inadvertent or accidental unblinding. The bioanalytical lab may provide Ionis with data sets containing false patient identifiers unrelated to the actual identifiers to allow review of the PK data.

2.5.4 Echocardiogram (ECHO) Data

The ECHO data will be collected, analyzed and stored in a secure database by an independent CRO (PPD The sites will upload the ECHO data on a secure web-portal for analysis by PP D. For the purpose of pre-programming and data cleaning, Parexel and a firewalled data manager working at the CRO contracted to perform data management (PPD) will have access to the post-baseline ECHO data.

2.5.5 Electrocardiogram (ECG) Data

All ECG data (machine read) are entered into the PPD EDC by the sites. The PPD data will be used for ECG summary and analysis.

All triplicate ECG waveforms will be collected and stored in a secure database by an independent CRO (PPD). Selected ECGs will be analyzed by PP D and the resulting data captured in the PP D database. The PP D data is used to support medical monitoring and will not be used for the ECG summary and analysis.

2.5.6 mNIS+7 and Neuropathy Symptoms and Change Score (NSC)

The primary efficacy assessment, mNIS+7 scores, will be collected and stored by an independent contract research group, PPD that is under the direction of PPD . The NSC score is obtained during the NIS assessment procedure and is also collected and stored by PPD . The mNIS+7 results from each site will be faxed to PPD for processing and quality assurance. Faxed copies will be maintained in secure rooms within locked cabinets. The NIS and NSC data are stored in the PPD (a database). The other components of the mNIS+7 (nerve conduction, sensory testing and heart rate to deep breathing) are entered by the PPD into a firewalled portion of the study EDC system. PPD only has access to this portion of the EDC system and the Sponsor and clinical sites do not have access to the PPD portion of the EDC. The mNIS+7 summated score will not be shared with the sites. Up until the EOT database lock and unblinding, the Sponsor will only have access to patient baseline values of the mNIS+7.

2.6 Blinding

The SAP will be finalized prior to unblinding. The EOT analysis will be performed after all patients have completed the EOT assessments, the database is locked and treatment code is unblinded. All of the safety and efficacy outputs will be produced for this final analysis. However, at the end of the study after all of the patients completed their post-treatment evaluation period or enrolled into open label extension study, all data listings and applicable outputs will be updated.

For the purpose of pre-programming and data cleaning, PPD and a firewalled data manager working at the CRO contracted to perform data management (PPD) will have access to the post-baseline mNIS+7, NSC, Norfolk-QOL-DN, SF-36, PND, ECHO and body weight. All staff at PPD will be blinded to treatment assignment. At PPD a small team responsible for providing the DSMB unblinded summary statistics for efficacy endpoints were unblinded to treatment assignments. Details of the unblinded analysis of efficacy endpoints including safeguards to ensure study integrity are detailed in Section 5.2.

3 Analytical Plan

3.1 Statistical Design Summary

This Statistical Analysis Plan (SAP) specifies the study endpoints to be analyzed, the study populations, and the methods of analysis. Section 3.2 provides general guidelines to be followed for all analyses, and covers the analysis populations, handling of missing data, and other general topics. Analyses of baseline characteristics are also covered in this section. Section 3.3 discusses the primary efficacy analysis and the sensitivity analyses to be conducted on the primary efficacy endpoint. Section 3.4 specifies the secondary efficacy endpoints and methods of analysis for these endpoints. Sections 3.5 through 3.8 cover tertiary efficacy endpoints, pharmacodynamic endpoints, pharmacokinetic analyses, and analysis of safety endpoints, respectively.

Section 4 provides the sample size justification. Section 5 presents details on the interim analysis to be conducted for this study: an early review of the reduction in TTR levels with possible stopping for futility.

The SAP concludes with a references section and appendix.

3.2 General Overview of Analyses

This analysis plan describes the reporting of data at the end of the study (unless specified otherwise). Additionally, some of these data will also be reported for the purposes of the DSMB and/or the TTR interim analysis.

3.2.1 Analysis Conventions

All reporting will be performed in SAS version 9.1.3 or higher.

Efficacy results will be summarized under the treatment to which patients were randomized. Safety and PK results will be summarized under the treatment which patients actually received. Should there be any cases after unblinding in which a patient received treatment other than what was randomized, such cases will be discussed in the study report and noted in footnotes where applicable.

All electronic case report form (eCRF) data, lab data transfers, echocardiogram, ECG, and mNIS+7 score data, as well as any outcomes derived from the data, will be provided in the patient data listings. Patient data listings will be presented for all patients enrolled into the study, and will be sorted by treatment group, subject ID, visit, and time point (where appropriate).

Central laboratory assessments will be included in the summary tables, local laboratory assessments will only be included in listings. Exceptions to the use of local laboratory assessment in summary tables is made for platelets, where summary tables will use both local and central laboratory results

All tables will present the population frequencies in each treatment group and/or subgroup (where appropriate) and will indicate the number of subjects with non-missing data and the denominators for percentages.

Descriptive summary statistics including n, mean, percentiles (e.g., median, P25, P75), standard deviation, and range (minimum, maximum) for continuous variables, and counts and percentages for categorical variables, will be used to summarize data by treatment group. Summaries of pharmacokinetic parameters will also present geometric mean, standard deviation of log-transformed data, and CV%. Where appropriate, p-values will be reported. All statistical tests will be conducted using 2-sided tests with a 5% Type I error rate unless otherwise stated.

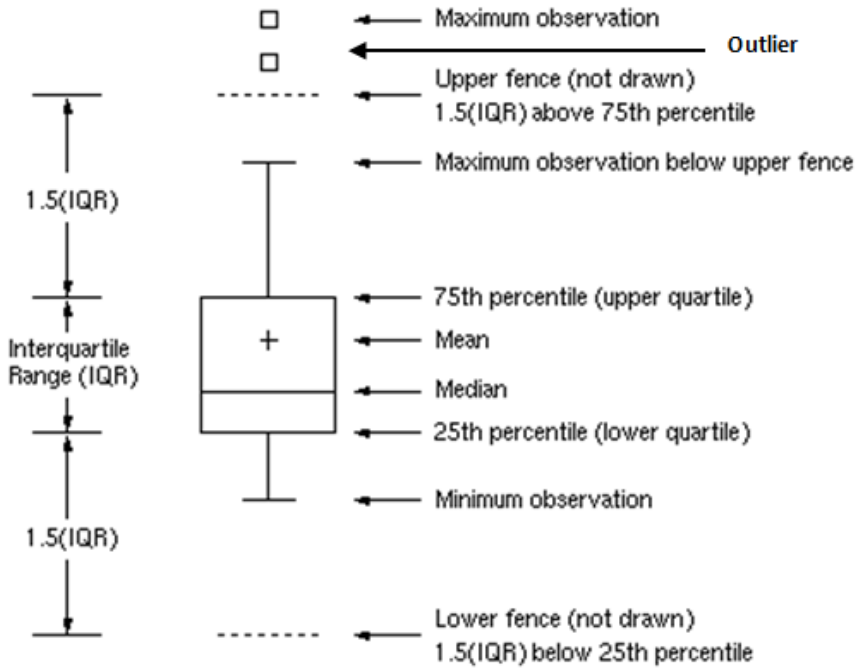
Age will be presented in years. Weight will be presented in kg. Height will be presented in cm. Temperature will be presented in °C. Laboratory values will be summarized using standard units.

Precision for displays will use the following conventions. Means, percentiles (e.g., median, P25, and P75), least squares means, difference in least squares means will be displayed to one more decimal place than measured values. Standard deviations and standard errors will be displayed to two more

decimal places than measured values. The minimum, maximum and confidence intervals will be displayed using the same number of decimal places as the recorded values. All raw values presented in listings will be displayed to the measured precision. Percentages will be displayed to one decimal place. P-values will be displayed to 3 decimal places. Confidence intervals (CIs) will be presented using a comma separator rather than a dash.

All primary and secondary efficacy and pharmacodynamic endpoints, except GLS will be assessed on the Full Analysis Set and Per-Protocol Set, with the former being the basis for the primary efficacy analysis. Tertiary efficacy endpoints will be assessed only on the Full Analysis Set (Note that the study protocol specified these endpoints would additionally be assessed in the Per-Protocol Set). All safety assessments will be performed on the Safety Set. PK endpoints will be assessed in the PK Set as applicable. ECHO endpoints including GLS will be assessed in the All Randomized Set, the ECHO subgroup and in the CM-ECHO Set. All analyses will take place after all patients have completed treatment and their EOT efficacy assessments and the database has been locked.

As shown in the schematic below, boxplots will display values from minimum to maximum within the upper and lower fences, the likely range of variation from P25 to P75 and the median. A symbol inside the box will indicate the mean. A segment inside the box will show the median and "whiskers" above and below the box will show the locations of the minimum and maximum observations within the upper and lower fences (see definition below). Individual observations outside the upper or lower fences will also be marked.



Should the randomization stratum recorded from IVRS be different from the actual data recorded in eDC, the randomization stratum recorded from IVRS will be used in the analysis.

Treatment effects for the primary, secondary, teritiary, and select exploratory endpoints will be evaluated based on a 2-sided significance level of 0.05. All other endpoints will be evaluated in an exploratory manner.

All tables and figures containing efficacy and pharmacodynamic endpoints will indicate whether they present raw data or adjusted results from the statistical model, with footnotes indicating the model used and covariates included in the model.

3.2.1.1 Definitions and Computational Formulas

- Day 1 will refer to the day of the first dose of Study Drug.
- The baseline for efficacy, PD and safety assessments will be defined as follows:
    - Baseline mNIS+7 and its individual components will be defined as the average of two assessments taken within 60 days prior to the first dose of Study Drug. If only one assessment has been done, the single assessment will be used in place of the average. Rarely, for patient convenience, the baseline mNIS+7 assessment(s) (or a subset of this assessment) will have been completed early in the treatment period rather than pre-treatment. These will be considered protocol deviations. These assessments will be included in the analysis as valid baseline assessments provided they are taken within one week after the first dose. The rationale for this is that the pharmacology of the drug indicates that the drug will have no effect on mNIS+7 this early in treatment, and including these values as the baseline assessments will allow these patient's data to be included in the primary analysis. A sensitivity analysis will be conducted excluding these assessments. Note that the NIS screening assessment can be used as one of the two assessments provided it was done within 60 days of first dose of Study Drug.
    - Baseline NSC and individual components will be defined as the average of two assessments taken within 60 days prior to the first dose of Study Drug. If only one assessment has been done, the single assessment will be used in place of the average. Because NSC score is collected during the NIS assessment procedure, it is possible it could be completed early in the treatment period rather than pre-treatment. These will be considered protocol deviations. These assessments will be included in the analysis as valid baseline assessments provided they are taken within one week after the first dose.
    - The baseline ECG will be defined as the average of the triplicate taken on Day 1 Pre-dose. If only one or two assessments are available, the single assessment or average of the two assessments will be used. If the case that Day 1 Pre-dose ECG is missing, screening visit results will be used as baseline.
    - The protocol permitted baseline ERG and ophthalmology examinations can be done up to one week after Study Day 1. Assessments done within one week of first dose will be included in the analysis as valid baseline assessments.
    - The baseline laboratory assessment including PD will be defined as the average of all non-missing pre-dose assessments.

- The baseline for all other assessments will be defined as the last non-missing value prior to the first dose of Study Drug.

- For efficacy endpoints except for BMI and mBMI:
    - The efficacy on-treatment period spans the time during which the study treatment is administered until 52 days after the last dose of medication.
    - The efficacy post-treatment period starts on the day after the efficacy on-treatment period and ends on the day of the patients's last contact date within the study.
- For BMI, mBMI and PD endpoints:
    - The on-treatment period spans the time during which the study treatment is administered until 28 days after the last dose of medication.
    - The on-post-treatment period starts on the day after the on-treatment period and ends on the day of the patient's last contact date within the study.
- For safety endpoints except ERG:
    - The safety on-treatment period spans the time during which the study treatment is administered until 7 days after the last dose of medication.
    - The safety post-treatment period starts on the day after the safety treatment period and ends on the day of the patient's last contact date within the study.
    - The safety on-study period spans the time drug is first administered until the day of the patient's last contact date within the study.
- For ERG:
    - The on-treatment period spans the time during which the study treatment is administered until 28 days after the last dose of medication.
    - The post-treatment period starts on the day after the on-treatment period and ends on the day of the patient's last contact date within the study.

- The PK and Immunogenicity (IM) on-study period spans the time drug is administered until the day of the patient's last contact date within the study.
- Note: Assessments for mNIS+7, NSC, ERG, and ophthalmology done early in the treatment period that are used for baseline cannot also be used as an on-treatment assessment.
- Body mass index (BMI) will be computed using the formula:

    BMI = (weight in kilograms) / [height in cm / 100]^2
- Modified BMI (mBMI) will be computed from BMI and serum albumin levels by:

    mBMI = BMI * serum albumin (g/L).

- Duration of study drug exposure will be derived as the difference between the date of the last dose of study drug and the date of the first dose plus one.
- Study drug will be administered as 1.5 mL subcutaneous (SC) injections. For patients receiving ISIS 420915, the dose for each administration will be 300 mg. The total dose of study drug during the treatment period will be computed for subjects receiving ISIS 420915 by summing the administered dose between the first dose of study drug on Day 1 and the EOT date or date of premature termination. Total dose will be summarized in mg.

3.2.1.2 Scoring of Assessment Instruments

mNIS+7

The mNIS+7 consists of two composite scores: the NIS composite score (maximum of 244 points) and the modified +7 composite score (maximum of 102.32 points).

NIS

The NIS composite score consists of 4 components:

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

Questions that make up each composite are listed in the table below.

NIS composite	Question number
CCI	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]

CCI [redacted]

Modified +7

The modified +7 composite score consists of 4 components:

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

- CCI

Due to the testing algorithm and the MC Core data entry conventions, raw modified + 7 data need to under go a pre-processing step before the scores can be derived. The different pre-processing steps are summarized in the table below.

Component	Data entry convention /Comment	Pre-processing step
CCI		

	CCI	

calculated by multiplying the sum of the 10 averaged subcomponent scores (defined below) by 2.

NIS+7

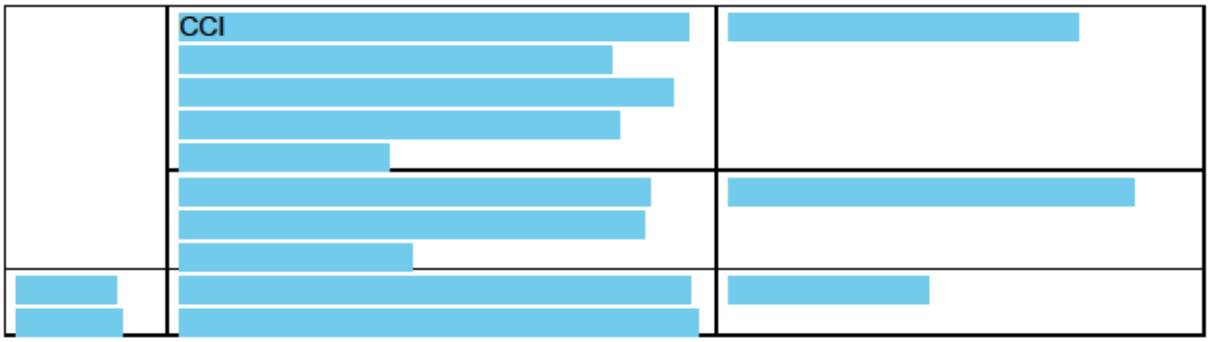
The NIS+7 consists of two composite scores: the NIS composite score (above) and the +7 composite score (maximum of 26.04 points).

+7

The +7 composite score consists of 3 components:

- CCI

Component	Data entry convention /Comment	Pre-processing step
CCI		



Norfolk QOL-DN

The Norfolk QOL-DN (CCI) consists of one composite score (Total QOL) and five sub-domain scores (physical functioning/large fiber neuropathy, activities of daily living, symptoms, small fiber neuropathy, and autonomic neuropathy). The scoring of the Norfolk QOL-DN will be conducted according to the scoring manual developed at the Eastern Virginia Medical School and is summarized below.

Questions that make up each domain are listed in the table below

Norfolk QOL-DN Domain	Question number
CCI	

All symptoms (Questions 1 to 7) are a simple inventory of symptoms of neuropathy. Each of these questions are assessed for the feet, legs, hands and arms, with presence of a symptom scored as a 1 and absence as a 0. The Question Score is calculated by summing the scores for the individual sites for the Question, with the Question Score ranging from 0 (no symptoms) to 4 (symptoms in the feet, legs, hands, and arms). The other Questions, except Questions 31 and 32, are scaled on a 5 point Likert scale, ranging from 0 ("Not a problem") to 4 ("Severe Problem"). In Question 31, "Good" is scored as 0, "Very Good" is scored as -1, "Excellent" is scored as -2 , "Fair" is scored as 1, and "Poor" is scored as 2. In Question 32, "About the same" is scored a 0, "Somewhat better" is scored a -1, "Much better" is scored a -2, "Somewhat worse" is scored a 1, and "Much worse" is scored a 2.

SF-36

The SF-36 (version 2) consists of two composite scores, the Physical Component Summary score and the Mental Component Summary score, as well as eight domain scores (physical function, role-physical, bodily pain, general health, vitality, social functioning, role-emotional, and mental health). The scoring of the SF-36 will be conducted using Quality Metrics Certified Scoring Software v 4.5.

NSC

The NSC (version: 04Jan2009) questionnaire consists of one total score and five domains (muscle weakness, sensory (hypo/loss of sensation), sensory (paresthesia, hyper sensation), autonomic (gastro intestinal (GI) and urinary incontinence), and autonomic (non-GI/non-urinary incontinence)).

The muscle weakness domain is divided in to four sub-domains (head and neck, chest, upper limbs, and lower limbs). The scoring of the NSC questionnaire is described in the Appendix.

Columbia Suicide Severity Rating Scale (C-SSRS)

Scoring of the C-SSRS questionnaire will be done following the scoring and data analysis guidelines provided by the Center for Suicide Risk Assessment at Columbia University Medical Center.

3.2.1.3 Handling of Missing or Replicated Data, Unscheduled Visits, and Early Termination Visits

Composite score: NIS score, +7 score, NIS+7 score, modified +7 score, or mNIS+7 score

Missing data imputation strategies for missing visit level data

If a patient misses a visit (or the visit is performed outside the analysis visit window), or the entire mNIS+7/NIS+7 assessment is not conducted at a visit, then the mNIS+7/NIS+7 and the composite, components and subcomponents will be considered to be missing at that visit. In a Mixed Model for Repeated Measures (MMRM) , missing data are not explicitly imputed. Instead, all available post-baseline assessments (within the scheduled visit windows) of the endpoint during the treatment period are utilized and via modelling of the within subject correlation structure, the endpoint treatment differences (which are adjusted to take account of missing data) are derived. In addition several 'Missing not at random' methods are described in Section 3.3.3.2 of the statistical analysis plan which will be used as sensitivity analyses to impute missing visit level data.

Missing data imputation strategies for missing assessment level data

Two independent assessments of the primary efficacy endpoint, mNIS+7, are planned at the baseline visit and the Week 66 visit, and the early termination visit (for patients that terminate treatment early). A single mNIS+7 assessment is also planned at the Week 35 visit. The mean of the two replicate assessments within visit will be used for analysis of both the baseline and Week 66 visits (provided both visits fall in the visit window and are within 52 days of the last dose of medication) . Subcomponent scores will be averaged first. These will be referred to as the averaged subcomponent scores.

At baseline and Week 66, in the event that only one subcomponent has been performed, the single subcomponent will be used in place of the mean value for that visit for the averaged subcomponent score. If both of the subcomponent values are missing, the averaged subcomponent score is missing. At Week 35, only one assessment is performed, therefore the single subcomponent will be used as the averaged subcomponent score for that visit. These values will be used in the summary and analysis of averaged subcomponent scores.

Two independent assessments of mNIS+7 are planned at early termination visits. If both assessments are within the same visit window the mean of the two will be used. At early terminations, in the event that only one subcomponent has been performed, the single subcomponent will be used in place of the mean value for that visit for the averaged subcomponent score.

The component scores will be computed by summing the averaged subcomponent scores and the composite scores will be computed by summing the component scores.

Imputation of missing averaged subcomponents

If a patient has completed at least part of the mNIS+7/NIS+7 at a visit then the following imputation method will be used to impute this missing assessment level data for the purposes of determining component scores for summary and analysis.

The following missing data imputation steps will be considered and will be used as described below for Groups A, B, and C:

- Step 1: If at least 50% of averaged subcomponent scores within a component are available, the missing averaged subcomponent scores will be set to equal to the mean of the patient's other non-missing averaged subcomponent scores in that component. The component score is then calculated.
- Step 2 (baseline): In the unlikely event that there are more than 50% of the averaged subcomponents scores within a component that are missing at baseline, the missing averaged subcomponent scores will be set to equal to mean baseline averaged subcomponent score from the parent study Randomized Set (across treatment groups). The component score is then calculated.
- Step 3 (post-baseline visits): For certain components, and only under certain conditions which will be listed below, the missing averaged subcomponent scores at that visit within that component only will be set equal to the mean averaged subcomponent score among the subjects randomized to placebo in the Randomized Set at that visit. The component score is then calculated.

  If a post-baseline assessment does not fall into the scheduled analysis windows, there is no obvious visit on which the mean subcomponent scores in the placebo group can be derived. In order to apply step 3, the following visits will be used to derive the mean scores in the placebo group:

Timing of Visit	Imputation rule
Before Week 35	Randomized Set mean at baseline
Between Week 35 and Week 66	Week 35 placebo mean
After Week 66	Week 66 placebo mean

Note: when imputing missing subcomponents based on placebo mean at a specific visit, the placebo mean is derived from on-treatment assessments only (i.e., those that were done within 52 days of last dose).

The components of the mNIS+7 and NIS+7 are grouped into A, B and C based on the imputation step used, as follows. A detailed list of components by group can be found in Table 1– Table 3 in Appendix 1:

- Group A: For components with multiple subcomponents except the NCT component of +7, imputation steps 1 and 2 will be applied.

  If, after applying step 1 for post-baseline visits, 6 out of the 7 components of the mNIS+7 composite score (NIS-C, NIS-R, NIS-S, NIS-W, HP, TP or NCT) are available and only one is missing at that visit, then step 3 will be applied for the missing component.
  If, after applying step 1 for post-baseline visits, 4 out of the 5 components of the NIS+7 composite score (NIS-C, NIS-R, NIS-S, NIS-W, or NCT) are available and only one is missing at that visit, then step 3 will be applied for that the missing component.
  Mean averaged subcomponent score used in the imputation described in step 3 are derived from the averaged subcomponent scores before step 1 imputation among the group of patients who were randomized to placebo.

  If, after applying steps 1-3 as appropriate, there are still missing subcomponent scores, the component score will be set to missing.
- Group B: For the NCT component of +7, it should be noted that the following 3 of the 5 subcomponents of this component may be "not evaluable" as an additional category to missing: 1) fibular nerve motor conduction velocity (PMCVK), 2) fibular nerve distal latency (PMLA), 3) tibial nerve distal latency (TMLA) (see Table 3 in Appendix 1). These nerve conduction attributes are not evaluable when the tibial or fibular nerve amplitude is 0, therefore, these "not evaluable" results are considered informative missing results and a slightly different imputation method is applied here. The following imputation rule will be used for Nerve Conduction Tests component score of +7: The normal deviate score for PMCVK, PMLA and TMLA will be respectively set to 3.72 (the worse response) if the recorded response was classified "not evaluable." After this, imputation step 1 and 2 will be applied.

  If, after applying step 1 for post-baseline visits, 4 out of the 5 components of the NIS+7 composite score (NIS-C, NIS-R, NIS-S, NIS-W, or NCT) are available and only one is missing at that visit, then step 3 will be applied for that the missing component.

  Mean averaged subcomponent score used in the imputation described in step 3 are derived from the averaged subcomponent scores before step 1 imputation among the group of patients who were randomized to placebo in the Randomized Set at that visit.

  Note that these components are *not* used in the nerve conduction component of the *modified* +7, therefore Group B only includes the NCT component of the NIS+7 and not the NCT component of the mNIS+7.

- Group C: The two components, HRDB and vibration tests, have only one subcomponent. Missing data for these averaged subcomponents score will be imputed as follows:
    - For baseline, the missing averaged subcomponent scores will be set to equal to the mean baseline averaged subcomponent score from the Randomized Set (across treatment groups).
    - For post-baseline visits, the missing averaged subcomponent scores at that visit will be set equal to the mean averaged subcomponent score among the subjects randomized to placebo in the Randomized Set at that visit. If a post-baseline assessment does not fall into the scheduled analysis windows, there is no obvious visit on which the mean subcomponent scores in the placebo group can be derived. In order to derive the mean scores in the placebo group, the following visits will be used:

Timing of Visit	Imputation rule
Before Week 35	Randomized Set mean at baseline
Between Week 35 and Week 66	Week 35 placebo mean
After Week 66	Week 66 placebo mean

Composite Score

The composite scores of mNIS+7, NIS+7, modified +7, +7, and NIS will each be calculated by summing the imputed component scores. If any of the component scores after imputation are still missing within a composite, the composite score will be set as missing.

Norfolk QOL-DN Domain and Total score

- For each patient at a specific visit (defined by the analysis visit window), if at least 50% of the questions for a domain (physical functioning/large fiber neuropathy, activities of daily living, symptoms, small fiber neuropathy) are not missing or if at least one question is not missing for autonomic domain, the missing questions are imputed as follows: If any question is missing at baseline, the mean value for this question at baseline from the study population (across all treatment groups) will be used to impute the missing baseline question value. For post-baseline visits during the treatment period, any missing question values will be imputed using the last observed or imputed question value (including baseline value). For the symptom domain, in the case that a patient responded on a particular question (Questions 1- 7) as not a having the symptom but also marked presence of the symptom in their feet, legs, hands, or arm, the question will be set to missing and the imputation rules will be followed.
- Otherwise, the total for that domain will be set to missing

The Norfolk QOL-DN total score will be calculated by summing the imputed domain scores. If any domain score after imputation is still missing, then the Norfolk QOL-DN total score will be set to missing.

The Norfolk QOL-DN individual domain and total scores will only be calculated for visits where the patient had a Norfolk QOL-DN assessment. The individual domain and total scores are set to be missing if a patient misses the visit or does not have a Norfolk QOL-DN assessment at that visit.

NSC Domain and Total score

Two independent assessments of NSC are planned at the baseline visit and the Week 66 visit. A single NSC assessment is planned at the Week 35 visit. The mean of the two replicate assessments within a visit will be used for analysis of both the baseline and Week 66 visits (provided both visits fall in the visit window and are within 52 days of the last dose of medication). The individual questions scores will be averaged first. These will be referred to as the averaged question scores.

At baseline and Week 66, in the event that only one NSC assessment has been performed, the single NSC assessment will be used in place of the mean value for that visit for the averaged question score. If both of the NSC assessments are missing, the averaged question score is missing. At Week 35, only one assessment is performed, therefore the single NSC assessment will be used as the averaged question score for that visit. The sub-domain and domain scores will be computed using the averaged question scores.

Imputing Missing Assessment Averaged Question Scores

The following imputation rule will be applied at the domain level for the sensory (hypo / loss of sensation), sensory (paresthesia, hyper sensation), autonomic (GI and urinary incontinence) and the autonomic (other than GI/urinary incontinence) domains

- If at least 50% of the averaged question scores in the domain are available, the missing questions will be set to be equal to the mean of the non-missing averaged question scores in the domain. The total domain score is then calculated from the sum of the non-missing and imputed averaged question scores in the domain.
- Otherwise, the domain score will be considered to be missing

Note for the imputation of the autonomic (other than GI/urinary incontinence) component in women, questions 35 and 36 are ignored and are not included in the imputation procedure

The following imputation rule will be applied at the sub-domain level for the four muscle weakness sub-domains (head and neck, chest, upper limb, lower limb):

- If at least 50% of the averaged question scores in the sub-domain are available, the missing averaged question scores will be set to be equal to the mean of the non-missing questions in the sub-domain. The total sub-domain score is then calculated from the sum of the non-missing and imputed averaged question scores in the sub-domain
- Otherwise, the sub-domain score will be considered to be missing

The total value for the muscle weakness domain score is calculated by summing the four sub-domain scores. If one or more of the sub-domain scores is missing, the muscle weakness domain score will be missing.

The total NSC score is calculated by summing the 5 domain scores. If one or more of the domain scores is missing the total NSC score will be missing

Pattern of missing data For mNIS+7 and Norfolk QOL-DN composite scores, the pattern of missing data will be explored (see section 3.3.3.1). Multiple imputation methods will be used to impute missing data in the sensitivity analyses of the primary efficacy endpoint. Details of these methods are presented in Section 3.3.3.2.

Imputation of Missing/Partial Dates The imputation of partial or missing dates for adverse events and prior/concomitant medications are detailed in Section 3.9.1. The imputation of partial or missing dates for duration of disease from diagnosis and duration from onset of symptoms for FAP/FAC are detailed in Section 3.2.3.

Replicated Data, Unscheduled Visits, and Early Termination Visits

When change from baseline is assessed, unless otherwise specified, only patients with both baseline and post-baseline measurements will be included in the analyses. If baseline or post-baseline value is missing, then the change from baseline will be set to missing.

For patients who withdraw, all data will be reported prior to the point of withdrawal in line with the population definitions and the specified analysis.

For data that are scheduled to be measured in duplicate or triplicate, the mean will be presented in tables and figures, while all measured values will appear in the listings.

If more than the scheduled number of measurements is taken, the mean will include all replicate measurements.

Analysis Visit Windows

The efficacy and PD data will be assigned to a visit according to the visit windows in the table below. Efficacy assessments that occurred more than 52 days after the last dose of Study Drug will not be included in the efficacy analyses/summaries during the efficacy on-treatment period, even if they occured within one of the visit windows. PD assessments, as well as body weight, BMI, and mBMI, that occurred more than 28 days after the last dose of Study Drug will not be included in the PD analyses/summaries during the PD on-treatment period, even if they occured within one of the visit windows. For patients who have multiple visits within a window, the visit nearest the target day will be used unless two visits are equally near, in which case the average will be used. Note that if there are multiple visits within a window with some being from the post-treatment evaluation period of the study, the visits from the post-treatment evaluation period will not be used. For mNIS+7 the assignment of assessments to a visit is done subcomponent by subcomponent according to the date the component was assessed. As long as the component is completed within the analysis window and within 52 days of last dose it is eligible to be used for the efficacy analyses/summaries during the efficacy on-treatment period. If, after subcomponents have been assigned to visit windows, there are two or more subcomponents of the same type within a window, the subcomponent that was assessed closer to the target day will be used (or the average of the two, if they are equally close). For baseline and Week 66 the two assessments are averaged (provided both assessments are within the visit window and are within 52 days of the last dose of medication). In case of

averaged subcomponents, for the purpose of determining proximity to the visit window target day, the date of the second assessment will be used.

Efficacy/PD measure	Nominal Visit (Target Day)	Analysis Visit Window (Day)
mNIS+7 and individual components, NSC, Norfolk QOL-DN and individual components	CCI	
SF-36 Questionnaire, PND Score	CCI	
BMI and mBMI	CCI	
PD Panel (TTR and RBP4)	CCI	
NT-proBNP	CCI	

Efficacy and PD data collected during the post-treatment period will be summarized with respect to the elapsed time from last dose. Assessments will be slotted into visit windows according to elapsed time from last dose based on the scheduled assessments during the post-treatment evaluation period of the study. The planned windows for this investigation are shown in the table below.

Efficacy/PD measure	Weeks from last dose (Days from last dose)	Analysis Visit Window (Days from last dose)
mNIS+7 and individual components, NSC, Norfolk QOL-DN and individual components	CCI	
SF-36 Questionnaire, PND Score	CCI	
BMI and mBMI	CCI	
PD Panel (TTR and RBP4)	CCI	
NT-proBNP	CCI	

Visit based safety assessments (scheduled and unscheduled) will be assigned to a visit according to analysis visit windows in the table below, even if they occured within one of the visit windows. Except for platelet assessments from the local laboratory, local laboratory assessment will not be assigned to a visit. Safety assessments that occurred during the safety post-treatment period (i.e., more than 7 days after the last dose of study drug) will not be included in the visit based safety analyses during the safety treatment period. If there are multiple visits within a window, the visit nearest the target day will be used unless two visits are equally near, in which case the average will be used. Note that safety assessments that are not utilized in the visit based summary tables will appear in listings and be used in the determination of treatment-emergent shifts or abnormalities.

Safety endpoint	Nominal Visit (Target Day)	Analysis Visit Window (Day)
Vital Signs	CCI	

Safety endpoint	Nominal Visit (Target Day)	Analysis Visit Window (Day)
	CCI	
ECG	CCI	
Retinol and Retinyl palmitate	CCI	
Chemistry Panel	CCI	
Serum Creatinine	CCI	

Safety endpoint	Nominal Visit (Target Day)	Analysis Visit Window (Day)
	CCI	
Hematology	CCI	

Safety endpoint	Nominal Visit (Target Day)	Analysis Visit Window (Day)
	CCI	
Platelets (weekly)	CCI	

Safety endpoint	Nominal Visit (Target Day)	Analysis Visit Window (Day)
	CCI	

Safety endpoint	Nominal Visit (Target Day)	Analysis Visit Window (Day)
Urinalysis	CCI	
Thyroid Panel	CCI	
Inflammatory Panel	CCI	
PT, aPTT, INR	CCI	
Complement	CCI	

Safety endpoint	Nominal Visit (Target Day)	Analysis Visit Window (Day)
	CCI	
Immunogenicity	CCI	
ERG	CCI	
Ophthalmology	CCI	
C-SSRS	CCI	
Body Weight	CCI	

To assess the impact of treatment discontinuation on safety parameters, data from visit based safety assessment may also be summarized with respect to the elapsed time from last dose. Assessment will be slotted into visit windows according to elapsed time from last dose based on the scheduled assessments during the post-treatment evaluation period of the study. The planned windows for this investigation are shown in the table below. After 6 weeks of follow-up in the post-treatment evaluation platelets will be collected as part of the hematology panel.

Safety endpoint	Weeks from last dose (Days from last dose)	Analysis Visit Window (Days from last dose)
Vital Signs	CCI	
Body Weight	CCI	
ECG	CCI	
Retinol and Retinyl palmitate	CCI	

Safety endpoint	Weeks from last dose (Days from last dose)	Analysis Visit Window (Days from last dose)
	CCI	
Chemistry Panel	CCI	
Serum Creatinine	CCI	
Hematology	CCI	
Platelets	CCI	
Urinalysis	CCI	
Thyroid Panel	CCI	
Inflammatory Panel	CCI	
PT, aPTT, INR	CCI	
Complement	CCI	
Immunogenicity	CCI	

Safety endpoint	Weeks from last dose (Days from last dose)	Analysis Visit Window (Days from last dose)
	CCI	
C-SSRS	CCI	

3.2.1.4 Data Summary Plan

The table below details what data will be included in the summary and analysis output by endpoint type, and what output is planned for the EOT and what output will be updated for the EOS. Data summarized/analyzed at EOT will be based on data collected up to the date of the data cut. It is possible that the on-treatment set of data available at EOS will be different to the one that was available at EOT because data collected after the EOT data cut may slot into the on-treatment period. On-treatment output provided at the EOT will be updated at EOS if the data in the on-treatment period has changed from EOT. In select instances the data for a type of endpoint that is summarized or analyzed may not be consistent with the rules in the Table; such exceptions will be noted in this document. Note: assessments for mNIS+7, NSC, ERG, and ophthalmology done early in the treatment period that are used for baseline cannot also be used as an on-treatment assessment.

Endpoint type	Provide at EOT	Update at EOS	Comment/definition
Efficacy	-Listings -On-treatment summary/analysis tables -Post-treatment summary tables	-Listings -On-treatment summary/analysis tables -Post-treatment summary tables	-On-treatment: First dose date ≤ Date of assessment ≤ Last dose date + 52 days -Post-treatment: Date of assessment > last dose date + 52 days -Visit defined by analysis visit window -The primary inference for mNIS+7 and Norfolk QOL-DN will be made at EOT, even if data slotted in the on-treatment period has changed by EOS
PD, BMI and mBMI, ERG	-Listings -On-treatment summary/analysis tables -Post-treatment summary tables	-Listings -On-treatment summary/analysis tables -Post-treatment summary tables	-On-treatment: First dose date ≤ Date of assessment ≤ Last dose date + 28 days -Post-treatment: Date of assessment > last dose date + 28 days -Visit defined by analysis visit window
ECG, Lab, vital signs (including body weight),	-Listings -On-treatment summary tables -Post-treatment summary tables	-Listings -On-treatment summary tables -Post-treatment summary tables	-On-treatment: First dose date ≤ Date of assessment ≤ Last dose date + 7 days -Post-treatment: Date of assessment > last dose date + 7 days -Visit defined by analysis visit window -Abnormal lab analyses described in Sections 3.9.4.1 to 3.9.4.3 include post-baseline data

Endpoint type	Provide at EOT	Update at EOS	Comment/definition
			collected up to the day of the patient's last contact date within the study
AE	-Listings -On-treatment summary tables -On-study summary tables	-Listings -On-treatment summary tables -On-study summary tables	-On-treatment summary tables include TEAE with onset dates up to the last dose date + 7 days -On-study summary tables includes all TEAE in the study, including those with onset dates > 7 days after last dose
Concomitant/ Prior medications	-Listings -On-treatment summary tables -On-study summary table	-Listings -On-treatment summary tables -On-study summary table	-On-treatment summary tables include medications taken on or after the study drug first dose date and up to the study drug last dose date +7 days - On study summary tables include medications taken after the study drug first dose date - Medications taken before the first dose date will be considered prior.
Immunogenicity	-Listings -On-study summary/ analysis tables	-Listings -On-study summary/ analysis tables	-On-study summary/analysis tables include post-baseline data collected up to the day of the patient's last contact date within the study -Visit defined by analysis visit window
PK	-Listings -On-study summary/ analysis tables	-Listings -On-study summary/ analysis tables	-Summary/analysis tables include post-baseline data collected include data collected up to the day of the patient's last contact date within the study -PK parameters will be derived based data collected up to the patient's last contact date within the study -PK concentration will be summarized based on nominal scheduled visits -All PK parameters will be recalculated using all available data at EOS

3.2.1.5 Multicenter Studies

This study expects to enroll approximately 135 patients from 30 to 35 investigative sites. For analyses that include investigative site, the analysis will use pooled sites. All sites with fewer than two randomized patients per treatment group with non-missing baseline and Week 66 mNIS+7 composite scores will be pooled together within country and considered a single site for analysis. If

this results in any site with fewer than two randomized patients per pooled site, the smallest site will be pooled with the next smallest site within the same geographic region (North America, Europe, or South America/Australasia). If there are no other sites in the region, no further pooling will be conducted.

3.2.2 Subject Populations Analyzed

The following analysis populations are defined for this study:

- Screened patients will be defined as those patients who signed an informed consent form.
- The Randomized Set will be defined as those screened patients who received a randomization assignment. Results will be summarized under the treatment to which patients were randomized.
- The Full Analysis Set (FAS) will include all randomized patients who received at least 1 injection of Study Drug (ISIS 420915 or placebo) and who have a baseline and at least one post-baseline efficacy assessment for the mNIS+7 score or Norfolk QOL-DN questionnaire total score. This will be the primary population for analysis of efficacy and PD outcomes. Results will be summarized under the treatment to which patients were randomized.
- The Safety Set (SS) will include all randomized patients who received at least 1 injection of Study Drug. This population will be used for analyses of all safety measures. Results will be summarized under the treatment which the patients received.
- The Per-Protocol Set (PPS) will include the subset of the Full Analysis Set who have received at least 80% of the prescribed doses of Study Drug and who have no major protocol violations that would be expected to affect efficacy assessments. Prescribed doses of Study Drug mentioned above is defined as the total assigned dose in mg from the Week 1 to the last dose within this study, i.e. 300mg*67doses = 20100mg. This will be a secondary population for efficacy and PD analyses and will be used for sensitivity purposes. The detailed criteria and definitions for major protocol violations will be specified and finalized prior to unblinding; individual subjects will be identified as meeting the violation criteria or not after locking the database and prior to unblinding. Results will be summarized under the treatment which the patients received.
- PK Set will include all subjects who are randomized and receive at least one dose of active Study Drug (ISIS 420915) and have at least one evaluable PK sample collected and analyzed with reportable result. Results will be summarized under the treatment which the patients received.
- The Pharmacokinetic subgroup (PKS) will include the patients that participated in the PK subgroup who have at least one evaluable PK result. This population will be used for pharmacokinetic analyses. Results will be summarized under the treatment which the patients received.

- The ECHO subgroup will include the subset of the Randomized Set that and qualified and consented for the ECHO substudy. Results will be summarized under the treatment to which patients were randomized.
- The CM-ECHO Set will include the subset of the Randomized Set that meet at least one of the following criteria, 1) diagnosis of TTR cardiomyopathy at study entry, or 2) eligible to participate in the ECHO subgroup (whether consented or not). Results will be summarized under the treatment to which patients were randomized.
- The TTR subgroup will consist of the subset of the Safety Set who have completed their Week 13 visit and are included in the interim analysis of TTR. Results will be summarized under the treatment to which patients were randomized.

All primary, secondary (except GLS), and PD endpoints will be assessed on the Full Analysis Set and Per-Protocol Set, with the former being the basis for the primary efficacy analysis. All safety assessments will be performed on the Safety Set. PK endpoints will be assessed in the PK Set as applicable.

3.2.3 Patient Characteristics

Patient disposition, including reasons for premature discontinuation of treatment or follow-up, number of patients in each analysis population (FAS, PPS, SS, PKS, ECHO Set, CM-ECHO Set), number and percent of randomized patients within each of the 3 randomization strata (previous treatment with Vyndaqel® or Diflunisal; disease stage; V30M TTR mutation) will be summarized by treatment group and overall for all randomized patients.

Patient allocation by investigative site will be tabulated by treatment group and overall for the all screened patients.

Protocol deviations will be listed and summarized by deviation category.

Patient demographic characteristics, including age, sex, race, ethnicity, TTR genotype, duration of disease from FAP diagnosis (months), and duration from onset of FAP symptoms (months) will be summarized by treatment group and overall for the SS, FAS, PPS, and CM-ECHO Set and for the PK and ECHO subgroups.

Baseline severity of illness, including the NIS score, mNIS+7, PND score, mBMI, BMI, weight, height, and NT-proBNP will be summarized by treatment group and overall for the SS, FAS, and PPS populations. Measures of quality of life and level of functioning, including the baseline Norfolk QOL-DN total score and pre-dose SF-36 will also be included. Information on familial amyloid cardiomyopathy (FAC) will be summarized, including FAC diagnosis (Y/N), duration of disease from FAC diagnosis (months), duration from onset of FAC symptoms (months), and clinical or laboratory criteria used to document the diagnosis of TTR cardiomyopathy.

Missing/partial dates (month and year) for the duration of disease from diagnosis for FAP/FAC and duration from onset of symptoms for FAP/FAC will be imputed as follows. Note that the day for these variables will not be imputed as this was not collected on the CRF. If year is missing no

imputation will be performed. If month is missing and the recorded year is before the year in informed consent date, the missing month will be imputed to be December, otherwise it will be imputed as the month from the informed consent date.

Exposure to Study Drug will be summarized for the SS by treatment duration (weeks), total volume (mL) and total dose administered (in milligrams, for the ISIS 420915 group only). The reason for dose pause will be tabulated. A listing summarizing for each subject the number of dose reductions (frequency of administered dose < protocol defined dose) and missed doses (injection not givens) will be provided. Doses that were were not given because the patient discontinued will not be summarized.

Medical history and baseline physical examination findings will be listed.

Medications will be coded using WHO Drug Dictionary. The final version used will be designated in the clinical study report. Prior medications, concomitant medications used while on-treatment and during the study will be summarized by treatment group for the SS and the CM-ECHO Set.

Prior and concomitant medications will be defined using the start and stop dates, and ongoing fields recorded in the CRFs relative to the first and last dose dates of randomised investigational product. A prior medication is defined as any medication taken up to, but not including the start date of investigational product. A concomitant medication is defined as any medication taken whilst investigational product is being taken. A concomitant medication is started during the post-treatment period (more than 7 days after last dose) will be considered an on-study medication but not on-treatment.

Note that if either of the start dates or stop dates of prior/concomitant medication are missing, the worst, or most conservative, case will be considered when slotting medications (i.e. the medications should slot into all possible phases). If a medication is administered pre-treatment or after first dose of investigation produce and no stop date/time is recorded then usage will be assumed to be ongoing for the remainder of the data collection periods. If a medication is stopped on-treatment or or after first dose and no start date/time is recorded it will be assumed that the medication was ongoing from prior to the start of investigational product. If a medication has no start or stop date it will be assumed that the medication was ongoing from prior to the start of investigational product.

If a partial date is recorded in the CRF, the following convention will be used to assign the medication:

- if the partial date is a start date, a '01' will be used for the day and 'Jan' will be used for the month
- if the partial date is a stop date, a '28/29/30/31' will be used for the day (dependent on the month and year) and 'Dec' will be used for the month.

The recorded partial date will be displayed in listings.

The definitions for prior, and on-treatment and on-study concomitant medication defined relative to use of the investigational product are shown schematically in the diagram below.

		On-Study			CM	
Scenario	Pre-treatment treatment	On-treatment	Post-	PM	On-trt	On-study
1	x----------x			Y	N	N
2	x----------x			Y	Y	Y
3	x----------x			Y	Y	Y
4		x----------x		N	Y	Y
5		x----------x		N	Y	Y
6			x-----x	N	N	Y
7	?----------x			Y	N	N
8		?----------x		Y	Y	Y
9	?----------x			Y	Y	Y
10	? ----------?			Y	Y	Y
11	x----------?			Y	Y	Y
12		x----------?		N	Y	Y
13			x------?	N	N	Y

PM = prior medication; CM – concomitant medication
x = start/stop date/time
? = missing date/time

3.3 Primary Analysis

3.3.1 Primary Endpoint Definition

There are two primary endpoints. The primary efficacy endpoints are the change from baseline to Week 66 in the mNIS+7 score and in the Norfolk QOL-DN questionnaire total score.

The mNIS+7 composite score has a maximum of 346.32 points. It can be broken into two components: the NIS component (maximum of 244 points) and the modified +7 component (maximum of 102.32 points). It can be further divided into the following 8 components: cranial nerves, muscle weakness, reflexes, sensory, heart rate deep breathing, nerve conduction, touch-pressure sensory, and heat-pain sensory.

The normal deviates (nds) from heart rate deep breathing and nerve conduction will be used in calculation of mNIS+7 for the primary analysis.

The mNIS+7 assessment is conducted at baseline (2 times), Week 35, Week 66 (2 times), and Week 91 (post-treatment). For visits where repeated assessments are done, the average will be used for analysis. Subcomponent scores will be averaged first, and the average composite score will be computed by summing the averaged subcomponent scores. Visits occurred after 52 days from the last dose of study medication are considered part of the post-treatment evaluation period, the data collected in the those visits will not be included in the efficacy analysis.

There are two scheduled visits for each of the two primary endpoints: Week 35 and Week 66. The data will be assigned to Week 35 or Week 66 according to the visit windows defined in section 3.2.1.3.

3.3.2 Primary Efficacy Analysis

The two primary endpoints, the mNIS+7 and the Norfolk QOL-DN questionnaire total score, will be tested using a ranking strategy with the mNIS+7 tested first and the Norfolk QOL-DN tested second. The primary efficacy analyses will be the contrast between the ISIS 420915 300 mg group and the placebo group at Week 66 from a Mixed Effects Model with Repeated Measures (MMRM) of change from baseline in each of these two endpoints during the treatment period. The primary analysis will be conducted using the Full Analysis Set (FAS). Interpretation will be made in a stepwise approach; should the null hypothesis, based on the mNIS+7, be rejected at a 2-sided 0.05 significance level, then the Norfolk QOL-DN questionnaire total score will be tested. However, if the null hypothesis for the mNIS+7 is not rejected, testing for the Norfolk QOL-DN questionnaire total score will be considered exploratory. No adjustment is therefore required for multiple hypothesis testing. This sequential testing procedure for the two primary objectives controls the overall Type I error rate at a 2-sided 0.05 level and is based on the methodology developed in Dmitrienko, Tamhane and Wiens (2008). The primary study hypotheses will be formally evaluated at the time of the end of treatment lock, occurring after all patients have completed the EOT assessments, the database is locked and treatment code is unblinded.

The MMRM method will include fixed categorical effects for treatment (two levels), time (two levels), treatment-by-time interaction, and each of the 3 randomization stratification factors (each with two levels). The baseline value of the endpoint and the baseline-by-time interaction will be included as fixed covariates in the model. Model parameters and treatment effects could be estimated by the following example SAS code:

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In this model, missing data are not explicitly imputed (except for the assessment level data imputation specified in Section 3.2.1.3). Instead, all available post-baseline assessments of the endpoint during the treatment period (which fall in the Week 35 and Week 66 visit windows) for subjects in the FAS are utilized and via modelling of the within subject correlation structure, the endpoint treatment differences (which are adjusted to take account of missing data) are derived. The estimation of treatment differences is based upon the assumption that the missing data follows a missing at random mechanism (i.e. the missingness of the observations may be dependent on the observed outcomes or covariates, but not on unobserved outcomes).

The unstructured (2x2) covariance model will be used to model the within patient errors, shared across treatments and small sample adjustments to standard errors and tests will be made following

the Kenward-Roger approach (Kenward, M. G. and Roger, J. H. (1997). In the unlikely circumstance that there are convergence problems with the repeated measures mixed model, this will be explored. For example the SCORING=4 option could be used in the PROC MIXED statement in SAS. This makes SAS use Fisher scoring for the first 4 iterations. If the convergence problem cannot be resolved the unstructured covariance matrix will be replaced by the restricted unstructured covariance matrix (Type=UNR in SAS). This also specifies an unstructured covariance matrix but with a different parameterization.

The treatment contrast for both Week 35 and Week 66 will be estimated by the model, with the Week 66 estimate considered primary. The normality assumptions for the MMRM models will be formally tested using a Shapiro-Wilks test at the 0.01 significance level and assessed by inspection of the following plots:

- Histogram of marginal studentized residuals derived from the MMRM model.
- Normal probability plot.

If the Shapiro-Wilks test assessing normality of the MMRM residuals from week 66 is statistically significant at the 0.01 level, formal hypothesis testing for that endpoint will be done at the 0.025 one-sided significance level using a non-parameteric rerandomization test. Thus, the null hypothesis for the endpoint will only be tested using the results from the MMRM if the p-value from the Sharpio-Wilks test is > 0.01.

Steps for the rerandomization test are:

1. The MMRM detailed above will be fit using the randomized treatment and the estimated treatment effect (difference in least squares means between the treatment arms) at week 66 from this analysis will be stored and will be denoted by t^*.
2. Assignment to ISIS 420915 and Placebo is randomly shuffled within each of the eight study strata. Note that the shuffling will be done for all patients in the FAS irrespective of whether their post-baseline data falls within an analysis window. The MMRM is refit but now uses the shuffled treatment assignment (instead of the actual assigned treatment) and the estimated treatment effect at week 66 from the analysis is stored. This step will be repeated in the event that the MMRM does not converge.
3. Step 2 is repeated 50,000 times, where the collection of the estimated treatment effects under the shuffled treatment assignment defines the randomization distribution. Denote the treatment effect from the ith permutation of the data by t_i.
4. The p-value or the probability of observing a treatment effect as extreme as or more extreme than t^* under the null hypothesis of no treatment difference is estimated by

$$\frac{1+\sum_{i=1}^{50,000} I(t_i \leq t^*)}{1+50,000},$$

where $I(A)$ is an indicator function that equals one if argument A is true, and 0 otherwise. If this one-side p-value is less than 0.025, the null hypothesis of no treatment difference will be rejected. The number of shuffled datasets may be increased depending if the simulation error in the estimated p-value is considered large.

If the rerandomization test is implemented, 95% CIs obtained by bootsrapping the data will be presented, with the resampling being done at the patient and not at the assessment level. A total of 10,000 bootstrap samples will be drawn, with the CI being defined as the 2.5^{th} and 97.5^{th} percentile from the empirical distribution of the treatment effect.

3.3.3 Additional Analyses of Primary Endpoints

In addition to the primary efficacy analysis, the following sensitivity analyses will be conducted on the FAS except where noted for each of the two primary efficacy endpoints:

- **Sensitivity Analysis 1** (Non-Parametric Analysis) – The non-parametric Van-Elteren test will also be performed for the two primary study endpoints as the sensitivity analysis. Hodges-Lehmann estimates of the differences between ISIS 420915 300 mg group and the placebo group as well as distribution-free CIs based on the Wilcoxon Rank Sum Test will also be provided.
- **Sensitivity Analysis 2** (Conservative Assessment Level Imputation) – To examine whether the primary analysis results are robust to the strategy for imputing missing assessment level data, an alternative strategy that results in a conservative estimate of the treatment effect will be implemented. Patients without an assessment at a visit will not have their score imputed for that visit. Imputation of missing baseline assessment level data will follow the approach detailed in Section 3.2.1.3. For patients with at least one non-missing post-baseline subcomponent score missing data will be imputed as follows. Missing post-baseline assessment level data will be imputed for the placebo group using their observed or imputed baseline value. Missing post-baseline assessment level data will be imputed for the ISIS 420915 group using the placebo mean in the Randomized population for that subcomponent at that visit (done after the placebo imputation).
- **Senstivity Analysis 3** (Excluding Assessments done at Early Termination Visits) – In order to examine the robustness of the primary analysis to the inclusion of premature termination data, the primary efficacy analysis will be repeated excluding data collected at early termination visits which are included in the primary analysis.
- Sensitivity analyses will be performed to investigate the impact of alternative missing data assumptions (see sections 3.3.3.2 and 3.3.3.3). These analyses will be done on the Safety Set and will be labeled as:
    - **Sensitivity Analysis 4** – Multiple Imputation assuming Missing at Random
    - **Sensitivity Analysis 5** – Multiple Imputation assuming Copy Increments from Reference
    - **Sensitivity Analysis 6** – Multiple Imputation assuming Jump to Reference
    - **Sensitivity Analysis 7** – Data at Withdrawal Visit Included
- **Sensitivity Analysis 8** (Per Protocol Set) – The primary efficacy analysis will be repeated, using the PPS population.

- **Sensitivity Analysis 9** (Adjustment for Pooled Site) – The primary efficacy analysis will be repeated, but with the addition of pooled investigative site as a fixed categorical effect in the model. In the event that there are model convergence issues, results from this analysis will not be provided and will be noted in the study report. The interaction between treatment and pooled site will also be examined if model convergence permits.
- **Sensitivity Analysis 10** (Responder Analysis) – A responder analysis based on the change in mNIS +7 score will be conducted to examine whether improvement in response is consistent over a range of response thresholds. A responder is defined as a patient whose mNIS +7 score change from baseline to Week 66 is less than or equal to the threshold value. The relationship between response rate and thresolds will be summarized using a cumulative distribution plot. In addition, comparison of response rates for specific threshold values will be done. Thresholds that will be tested will include 0, 2, 4, 6, 8, 10, 15, 20, 30 points above the baseline value. For each of these specific response thresholds, the response rates at Week 66 for both the treated group and the placebo group will be calculated and plotted against the response threshold. Patients that terminate treatment early irrespective of the reason or had missing Week 66 data will be considered a non-responder. The difference in the estimated response rate and associated 95% CI based on the Mantel-Haenszel approach will be presented for each threshold.
- **Sensitivity Analysis 11** (HRDB and Nerve Conductions Scored Using Points and NIS-Sensation Excluded) – Additional sensitivity analysis on mNIS+7 composite score will be performed using the points (instead of Normal Dev) from heart rate deep breathing and nerve conduction and also removing the sensation part of the NIS.
- **Sensitivity Analysis 12** (HRDB Component Excluded) – Additional sensitivity analysis on mNIS+7 composite score will be performed, with HRDB not included as a component. HRDB will also be descriptively summarized for the group that has least one non-imputed HRDB score during efficacy treatment period and a non-imputed score at baseline.
- **Sensitivity Analysis 13** (Modified mNIS+7 Baseline Definition) – As outlined in Section 3.2.1.1, the primary efficacy analysis for mNIS+7 will be repeated using a modified baseline definition, defined as the average of two assessments taken within 60 days prior to the first dose of Study Drug.

Descriptive statistics will be provided for absolute values and change from baseline for Norfolk QOL-DN questionnaire total score by study visit. For mNIS+7 descriptive statistics by visit will be provided for absolute values, change from baseline and percent change percent from baseline. Data collected outside the analysis windows will be summarized.

3.3.3.1 Description of the Missing Data

To examine the nature of missing data, cohorts of subjects will be defined based on the scheduled assessments that were completed.

For each of the primary endpoints there will be 3 cohorts of based on the FAS (based on the post-baseline data included in the primary analysis):

1. Subjects who have a Week 35 assessment only
2. Subjects who have a Week 66 assessment only
3. Subjects who have Weeks 35 and 66 assessments

For each endpoint plots will be produced, illustrating the mean change from baseline over time in each of the cohorts. These will be based on the observed data for each of the cohorts. The trends over time will be visually compared across cohorts to explore patterns of missing data. Of particular interest is the group that has complete follow-up data available (i.e. have Weeks 35 and 66 assessments) and the group with a Week 35 assessment only. This can help indicate if subjects show any sign of decline in their efficacy prior to dropping out from the study.

3.3.3.2 Multiple Imputation Methodology

For each of the primary endpoints, sensitivity analyses will be performed in the Safety Set using multiple imputation methods based on pattern mixture models. First, a repeated measures Gaussian model will be fitted to the data using a Bayesian approach, with non-informative priors for the mean and variance-covariance matrix to provide a joint posterior for the parameters in this model. The repeated measures Gaussian model will include separate mean profiles for each treatment group and the same covariates as those in the primary MMRM analysis.

Independent samples will then be drawn from the posterior distributions for the mean and variance-covariance matrix to provide inputs into an imputation model. For each subject with missing data, these sampled values of the parameters for mean vectors and the variance- covariance matrices specify a joint distribution for their observed and unobserved outcome data.

The post-withdrawal part of each pattern-specific distribution will be modelled using three different approaches discussed below. This imputation model will have the same covariates as those in the primary MMRM analysis.

Based on this imputation model, a single set of data will be sampled for the missing data based on the distribution for the subject's missing data conditional upon their observed data.

The post-withdrawal part of each pattern-specific distribution will be modelled using these three approaches:

1. Missing at Random (MAR) approach.

   The means and variance-covariances following withdrawal are chosen to reflect the subject's own treatment group. This approach should provide similar numerical results to the primary analysis.

2. Copy Increment from Reference (CIR) approach.

   The CIR approach is detailed in Carpenter et al. (2013) and addresses a potential pattern of informative missingness where the assumption is that the active Study Drug halts or slow

disease progression, and the disease progresses after treatment is discontinued. In CIR missing data in the placebo group will be imputed under a within treatment arm MAR assumption. For a patient in the active treatment group their mean profile (i.e. mean increments) will track that of the placebo group, but starting from the benefit obtained.

3. Jump to reference (J2R) approach.

   The J2R approach is detailed in Carpenter et al. (2013) and is an extemely conservative imputation approach that assumes that a patient receiving active Study Drug does not sustain benefit after discontinuing study treatment. In J2R missing data in the placebo group will be imputed under a within treatment arm MAR assumption. For a patient with missing data in the active treatment group their mean response distribution is set to equal that of the placebo group.

For each imputation method used, at least 5000 imputed datasets will be generated. The imputed observations in each dataset will be checked to ensure they are within the possible change from baseline range for the particualr subject they belong to. If a dataset contains out-of-range values, it will be discarded and a new dataset will be generated until there are 5000 datasets which contain no out-of-range values. Each of 5000 imputed data set will then be analyzed using simple analysis of covariance (ANCOVA) model at Week 66 and the resulting treatment differences and their standard errors will be combined using Rubin's rules. The ANCOVA model will include fixed effects for treatment (two levels), and each of the 3 randomization stratification factors (each with two levels) with baseline value of the endpoint as the covariate. Note that in these analyses efficacy assessment that are within the analysis window but more than 52 days after last dose will be included. This is different from the primary analysis, where data after 52 days from last dose is excluded.The number of imputed datasets may be increased after review of results if the simulation error is considered large.

A random seed to be used by a random number generator with value of 2855 will be used to initiate data imputation for all three methods.

3.3.3.3 Sensitivity Analysis Using Data at Withdrawal Visit

The primary outcome variable, modified Neuropathy Impairment Score +7 (mNIS+7) score, is measured on treatment at baseline, Week 35 and Week 66. The primary analysis is the difference between the active treatment and placebo at week 66 based on a repeated measures model for Week 35 and Week 66 data using treatment-by-time 2*2 factorial, baseline-by-time and strata covariate fixed effects, and an unstructured 2*2 covariance matrix shared across treatments. The analyzed response is difference from baseline in mNIS+7, but the treatment difference will be the same as if the absolute value of mNIS+7 were analyzed instead, since baseline-by-time covariate is included in the model.

In order to include the values of mNIS+7 at withdrawal visit in the analysis the following model is fitted as a sensitivity analysis in the Safety Set. Care needs to be taken with its interpretation as these withdrawal visit measurements may be extreme. The absolute value of mNIS+7, and not change from baseline, will be used as response. This is done for simplicity of expression of the

model and makes no difference to the results. The following model is equivalent to the previous primary analysis model when there are no withdrawal visit data. The only difference will come from the inclusion of the additional data from exit visits. If strata had been crossed with time or strata excluded from the primary analysis then they would be algebraically identical. The model is simply a re-expression of the primary analysis model that allows one to include data measured at times other than Weeks 35 and 66.

To implement this model, 3 time parameters should be defined as follows:

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The other primary endpoint, Norfolk QOL-DN, will also be analyzed using the methods described in this section.

3.3.4 Subgroup Analyses of Primary Endpoints

Subgroup analyses will be conducted for each of the two primary efficacy endpoints in the FAS. Subgroups defined by the following variables will be evaluated:

- V30M TTR mutation (Yes, No)
- Age (< 65 years old, ≥ 65 years old)
- Race (White, non-White)
- Sex (Male, Female)
- Region (North America, Europe, South America/Australasia)
- Previous treatment with Vyndaqel® or Diflunisal (Yes, No)
- Disease stage (Stage 1, Stage 2)
- CM-ECHO Set (Included, Not included)

The MMRM for the change from baseline with include fixed categorical effects for treatment (two levels), time (two levels), each of the 3 randomization stratification factors (each with two levels), treatment-by-time interaction, treatment-by-subgroup interaction, and treatment-by-time-by-subgroup interaction. The baseline value of the endpoint and the baseline-by-time interaction will be included as covariates in the model. The treatment-by-subgroup interaction at each timepoint will be tested at the significance level of 0.10. Treatment group differences will be evaluated within each category of the subgroup, regardless of whether the interaction is statistically significant.

3.4 Secondary Analyses

Secondary analyses include the analysis of secondary endpoints and pharmacodynamic analyses.

3.4.1 Secondary Efficacy Endpoint Definitions

Secondary efficacy endpoints are:

- Change from baseline to Week 66 in the Norfolk QOL-DN questionnaire symptoms domain score (Stage 1 patients only) and the Norfolk QOL-DN questionnaire physical functioning/large fiber neuropathy domain score (Stage 2 patients only)
- Change from baseline to Week 65 in the Modified Body Mass Index (mBMI)
- Change from baseline to Week 65 in the Body Mass Index (BMI)
- Change from baseline to Week 66 in the NIS
- Change from baseline to Week 66 in the modified +7

- Change from baseline to Week 66 in the NIS+7
- Change in GLS by ECHO from baseline to Week 65 in the Randomized Set, ECHO subgroup and in the CM-ECHO Set

Treatment group differences will be evaluated using the same method as the primary efficacy analysis (MMRM described in Section 3.3.2). These analyses will be conducted on both the FAS and the PPS populations. The analysis for GLS will be specified in the ECHO SAP.

The normality assumptions for the MMRM will be assessed for each of the secondary endpoints by inspecting the following plots:

- Histogram of marginal studentized residuals derived from the MMRM model.
- Normal probability plot.

The non-parametric Van-Elteren test may be performed as a sensitivity analysis, if deemed necessary. Hodges-Lehmann estimates of the differences between ISIS 420915 300 mg group and the placebo group as well as distribution-free confidence intervals based on the Wilcoxon Rank Sum Test will also be provided.

3.4.2 Pharmacodynamic Endpoint Definitions

Pharmacodynamic endpoints are:

- Change and percent change from baseline to Week 65 in TTR level
- Change and percent change from baseline to Week 65 in RBP4 level.

Analyses of the pharmacodynamic endpoints will be evaluated using the same method as the primary efficacy analysis (MMRM described in Section 3.3.2). All pharmacodynamic analyses will be conducted on both the FAS and the PPS.

In addition the proportion of subjects with percentage decrease from baseline in plasma transthyretin (TTR) $\geq$ 60% will be summarized by treatment group at each visit.

3.5 Tertiary Analyses

The tertiary efficacy analyses will assess whether ISIS 420915 is superior to placebo as measured by the following endpoints:

- Change from baseline to Week 65 in the SF-36 in the Physical Component Summary score, the Mental Component Summary score, and mental health domain score.
- Change from baseline to Week 66 in the individual components of the NIS score (cranial nerves, muscle weakness, reflexes, and sensory)
- Change from baseline to Week 66 in individual components of the modified +7 score (heart rate to deep breathing, nerve conduction, heat-pain sensory, and touch-pressure sensory)

- Change from baseline to Week 66 in individual components of the +7 score (heart rate to deep breathing, nerve conduction and vibration detection threshold)
- Change from baseline to Week 66 in the +7 score
- Change from baseline to Week 66 in individual domain scores of the Norfolk QOL-DN (physical functioning/large fiber neuropathy, symptoms, activities of daily living, small fiber neuropathy, and autonomic neuropathy)

Analyses of the tertiary endpoints will utilize the MMRM described in Section 3.3.2. All tertiary analyses will be conducted on the FAS (Note that the study protocol specified these endpoints would additionally be assessed in the PPS).

Similar to the primary endpoints, by-visit descriptive statistics will be provided for the individual mNIS+7 components, Norfolk QOL-DN questionnaire domain scores, PD assessments, and SF-36 domain scores. At the end of study, the summaries will be updated to include the Post-Treatment follow-up visits if supported by adequate data.

3.6 Exploratory Analyses

Exploratory endpoints include the following:

- Change from baseline in echocardiogram parameters (except GLS) in the Randomized Set, ECHO subgroup and in the CM-ECHO Set
- Change and percent change from baseline in NT-proBNP in the FAS, ECHO subgroup and in the CM-ECHO Set
- Change from baseline in the log-transformed NT-proBNP values in the FAS, ECHO subgroup and in the CM-ECHO Set
- Shift table from baseline in PND score in the FAS and in the PPS
- Change from baseline in NSC total score in the FAS and in the PPS
- Change from baseline in the NSC individual domain scores (muscle weakness, sensory [hypo/loss of sensation], sensory [paresthesia, hyper sensation], autonomic [GI/urinary incontinence], and autonomic [other than GI//urinary incontinence]) in the FAS and in the PPS
- Proportion of patients with at least 60% reduction in TTR in the FAS and in the PPS

Echocardiogram parameters, NSC (total and individual domains), and NT-proBNP (log-transformed) will be summarized and statistically analyzed using the MMRM described in Section 3.3.2. Summary statistics will be used to summarize the other exploratory endpoints.

Note that the 7 point change score (-3, -2, -1, 0, 1, 2, 3) that is recorded on the NSC questionnaire will be listed but not summarised or analysed. Similarly for questions 20-29 the location is only listed. Questions 31-34 are of particular intereste and therefore the number and percentage of patients responding 0 (No), 1 (slight +), 2 (moderate ++) or 3 (severe +++) will be presented.

The ECHO data analysis will be conducted by CICL.

Note: retinol and retinyl palmitate are specified as exploratory endpoints in the protocol. These endpoints are considered safety endpoints and therefore will be summarized with safety laboratory paratemers.

3.7 Analysis of the PD and Primary Efficacy Endpoints Relationship

The relationship between TTR and both primary endpoints will be explored in the FAS and the PPS among the subset with an assessment within the Week 66 analysis window. For each of the primary endpoints, a scatterplot will be provided with change from baseline to Week 66 in the primary endpoint on the y-axis and change from baseline in TTR on the x-axis. Change from baseline in TTR will be represented by the area under the curve (AUC), and calculated using the following formula

$$\sum_{v=1}^{V}[(y_v + y_{v-1})\times(t_v - t_{v-1})\times 0.5] - y_0\times(t_V - t_0)$$

where y_v is the TTR concentration from baseline to visit v on day $t_{v,}$, $v = 0, \ldots V$, with $v = 0$ corresponding to baseline.

A four parameter Emax model will be fit to this data to characterize the relationship. If the Emax model does not converge, other models will be explored.

An additional investigation will be performed based on the categorization of the AUC TTR data. There will be a total of five categories. The first category will include patients in the placebo group. The other categories is for the ISIS 420915 group, where categories are defined based on quartiles of the AUC for change from baseline in TTR. Change for baseline to Week 66 for both primary endpoints will be summarized by TTR category. An analysis of covariance model will be fit with the change from baseline in the efficacy endpoint as the dependent variable, and TTR category, baseline value of the primary endpoint, and randomization factor as explanatory variables. The estimated LS difference between the separate ISIS 420915 TTR categories and the placebo category will be presented.

3.8 Pharmacokinetic (PK) and Immunogenicity (IM) Analysis

Pharmacokinetic endpoints include the following:

- To evaluate the plasma trough levels of ISIS 420915 in all evaluable patients
- To evaluate the plasma pharmacokinetic parameters of ISIS 420915 in a subset of evaluable patients (PK subgroup).

Immunogenicity endpoints include the following:

- To evaluate sample IM status [confirmed positive, negative or unevaluable and, when applicable, titer of anti-ISIS 420915 antibodies (ADA)] before, during, and after treatment with ISIS 420915 or placebo in all evaluable patients

- To evaluate subject IM status (positive, negative or 'unknown') and its characteristics if applicable (onset, duration, peak titer, etc.)

Plasma samples will be collected at protocol designated times for ISIS 420915 plasma pharmacokinetic and immunogenicity assessments from patients receiving either ISIS 420915 300 mg or placebo treatment.

3.8.1 Plasma Pharmacokinetics

Pharmacokinetic analysis will be performed on two separation occasions: after all patients have completed treatment of ISIS 420915 (EOT) and at the end of the study (EOS).

3.8.1.1 Plasma Concentration Data

Plasma concentrations of ISIS 420915, along with the scheduled (nominal) and actual sampling times (i.e., time from SC dosing) will be listed for each evaluable patient by treatment, actual dose, gender, subject immunogenicity (IM) status, and study day. Plasma concentrations below the lower limit of quantification (LLOQ) will be indicated by "BLQ". Percent differences between nominal and actual dose, as well as between scheduled and actual sampling times will also be listed for all patients.

For all patients who receive ISIS 420915 treatment, ISIS 420915 plasma trough and post-treatment concentrations will be summarized using descriptive statistics by dose, study day, and scheduled time point, without and with stratification by IM status (see Section 3.8.2). ISIS 420915 plasma concentrations from the PK subgroup will also be similarly summarized. For the purpose of calculating typical summary descriptive statistics (n, mean, standard deviation (SD), standard error (SE), %CV, geometric mean, geometric %CV, median, minimum, and maximum) for plasma concentrations, all BLQ values will be set to zero. Mean plasma concentrations that are below the LLOQ will be presented as BLQ, and the SD, SE, and %CV will be reported as not applicable. Other stratifications may also be performed if deemed warranted to properly interpret the pharmacokinetic analysis. Samples will be excluded from descriptive statistics if there are large deviations between scheduled and actual sampling times (percent difference between scheduled and actual sampling time greater than 30%), or large deviations between actual dose and nominal dose (percent difference between nominal and actual dose greater than 30%). Any samples excluded from the summary descriptive statistics, if deemed necessary, will be listed separately along with the reason for exclusion.

For all evaluable patients, ISIS 420915 plasma trough (predose) and post-treatment concentrations versus time (actual) profiles from Study Day 1 to 631 for each individual patient, as well as corresponding mean (±SD) plasma concentration versus time (scheduled) profiles will be presented graphically on linear and semilogarithmic scales, without and with stratification by subject immunogenicity status (see Section 3.8.2). For the PK subgroup only, ISIS 420915 plasma concentration versus time (actual) profiles from Study Day 1 to 3, from Study Day 240 to 247, and from Study Day 449 to 456, for each patient, as well as corresponding mean (±SD) plasma concentration versus time (scheduled) profiles (by treatment), will be presented graphically on

linear and semilogarithmic scales, without and with stratification by subject immunogenicity status (see Section 3.8.2). Other stratifications may also be performed if deemed warranted. At the discretion of the pharmacokineticist and/or biostatistician, samples may be excluded from the mean plots if there are large deviations between scheduled and actual sampling times, or large deviations between actual dose and nominal dose.

3.8.1.2 Plasma Pharmacokinetic Parameters

For ISIS 420915 treated patients only, non-compartmental pharmacokinetic analysis of ISIS 420915 will be carried out on each evaluable individual patient data using Phoenix WinNonlin version 6.3 or higher (Pharsight Corporation, Mountain View, CA) to determine plasma pharmacokinetic parameters. For calculation of PK parameters, all BLQ values will be set to zero.

For the PK subgroup (ISIS 420915 treated) only, the following plasma PK parameters will be calculated (when applicable and possible) based on actual sampling times:

- C_{max} (µg/mL): the maximum observed drug concentration in plasma. Calculated for dosing on Study Days 1, 240, and 449.
- T_{max} (hr): the time at which C_{max} occurs. Calculated for dosing on Study Days 1, 240, and 449.
- $AUC_{0\text{-}24hr}$ (µg•hr/mL): areas under the plasma concentration-time curve from zero time (pre-dose) to 24 hours after the SC administration will be calculated using the linear trapezoidal rule for dosing on Study Days 1, 240, and 449.
- $AUC_{0\text{-}48hr}$ (µg•hr/mL): areas under the plasma concentration-time curve from zero time (pre-dose) to 48 hours after the SC administration will be calculated using the linear trapezoidal rule for dosing on Study Day 1 only.
- $AUC_{0\text{-}72hr}$ (µg•hr/mL): areas under the plasma concentration-time curve from zero time (pre-dose) to 72 hours after the SC administration will be calculated using the linear trapezoidal rule for dosing on Study Days 240 and 449 only.
- $AUC_{0\text{-}168hr}$ ($AUC_{0\text{-}\tau}$) (µg•hr/mL): partial AUC from time zero to 168 hr (dosing interval) will be calculated using the linear trapezoidal rule. This parameter will only be calculated for dosing on Study Days 240 and 449 only.
- $MRT_{0\text{-}24hr}$ (hr): mean residence time (MRT) from time zero to 24 hours after the SC administration will be calculated from the equation, $MRT_{0\text{-}24hr} = AUMC_{0\text{-}24hr} / AUC_{0\text{-}24hr}$ where $AUMC_{0\text{-}24hr}$ is the area under the first moment plasma concentration-time curve from time zero to 24 hours after the SC administration. This parameter will be calculated for dosing on Study Day 1, 240, and 449. This parameter will be calculated and reported mainly as an index measure to reflect the expected initial rapid disposition of ISIS 420915 from plasma to tissues shortly after dosing.
- $CL_{0\text{-}24hr}$ /F (L/hr): Plasma clearance will be calculated from $CL_{0\text{-}24hr}/F$= Actual Dose/$AUC_{0\text{-}24hr}$ for dosing on Study Day 1, 240, and 449.

- CL_{ss}/F (L/hr): Plasma clearance at steady-state will be calculated from CL_{ss}/F = Actual Dose/$AUC_{0\text{-}168hr}$ for dosing on Study Days 240 and 449 only.
- Vz/F (L): Apparent volume of distribution in the terminal phase will be calculated from Vz/F = $CL_{ss}/F/\lambda z$ for dosing on Study Day 449 only. Note: calculation of this parameter may not be possible in any ISIS 420915 treated patients rolling into an open label extension study.

For all evaluable patients (including subjects participating in the PK subgroup) receiving active study drug (ISIS 420915), the following plasma PK parameters may be calculated (when applicable and determinable) based on actual sampling times at the discretion of the pharmacokineticist:

- $t_{½\lambda z}$ (day): the plasma disposition half-life associated with the apparent terminal elimination phase may be calculated from the equation, $t_{1/2\lambda z} = 0.693/\lambda z$, based on any evaluable post-treatment data with scheduled collections on Days 491, 533, and 631. A minimum of three data points in the elimination phase will be used to define λ_z, and the correlation of determination values (r^2) have to be at or greater than 0.8 for the estimate to be accepted. Note: if quantifiable data is only available from just 2 time points or $r^2 < 0.8$, this parameter may still be determined at the discretion of the pharmacokineticist and will be flagged as such in the clinical study report. Note: calculation of this parameter may not be possible in any ISIS 420915 treated patients rolling into an open label extension study.
- $C_{trough\ ave,\ Day\ 29\text{-}85}$ (ng/mL): average plasma trough concentrations between Days 29 to 85 (Weeks 5 to 13).
- $C_{trough\ ave,\ Day\ 155\text{-}240}$ (ng/mL): average plasma trough concentrations between Days 155 to 240 (Weeks 23 to 35).
- $C_{trough\ ave,\ Day\ 365\text{-}449}$ (ng/mL): average plasma trough concentrations between Days 365 to 449 (Weeks 53 to 65).

Plasma pharmacokinetic parameters will be listed by dose, subject ID, gender, subject IM status, and study day; and appropriately summarized (separately for the PK subgroup and all evaluable ISIS 420915 treated patients) using descriptive statistics (n, mean, SD, SE, %CV, geometric mean, geometric %CV, median, minimum, and maximum) by study day. Additionally, subject IM status stratified (see Section 3.8.2) plasma pharmacokinetic parameters will be similarly summarized (again separately for the PK Subgroup and all evaluable ISIS 420915 treated patients). Other stratifications may also be performed if deemed warranted at the discretion of the pharmacokineticist and/or biostatistician.

Exposure-response relationships between selected pharmacodynamic (including but not limited to TTR level, mNIS+7 and Norfolk QOL-DN score) and pharmacokinetic measures (including but not limited to plasma trough concentrations) may also be explored (including with and without stratification by IM status), where appropriate.

3.8.2 Immunogenicity (IM) Analyses

Samples collected at predose on Days 1, 29, 85, 197, 323, and 449 and anytime on Day 631, including early termination and unscheduled samples for IM assessment will be analyzed for anti-ISIS 420915 antibodies (ADA). However, plasma samples collected at other time points (for PK

purposes) may also be potentially evaluated if deemed of further interest and warranted by the pharmacokinetic scientist. An evaluable sample will be designated 'IM positive' based on both positive screening and confirmation assay results (i.e., confirmed positive result), and otherwise will be deemed 'IM negative'. Sample IM results (screen positive/negative, confirmed positive/negative or unevaluable, and when applicable, titer of anti-ISIS 420915 antibodies) before, during, and after treatment with study drug (ISIS 420915 or placebo) (sample IM status) will be listed by treatment and dose.

Study subjects will be given 'IM positive' status if they have at least one confirmed positive sample result at any time during the treatment or post-treatment evaluation periods. Study subjects will be given 'IM negative' status if all evaluated IM sample results during the treatment and post-treatment evaluation periods are IM negative and they have at least one evaluable IM result collected post study drug treatment. Otherwise, a study patient will be given 'unknown' IM status. Subject IM results will be listed by treatment and dose for all evaluable patients, which will include but may not be limited to: subject IM status (positive, negative or unknown), the study day associated with the first positive IM status emerged (T_{first}, i.e., onset of ADA development), the last positive IM status observed (T_{last}), the time of last evaluable IM sample collected ($T_{last\ sampling}$), peak titer, and time to reach peak titer. The onset of ADA and time to reach peak titer will be calculated by:

- Onset in days = The date of first sample has "positive" sample IM status - first dose date +1;
- Time to reach peak titer in days = The date of first peak titer observed- first dose date +1;

Other immunogenicity data analysis (e.g. classification as transient or persistent status for IM positive subjects) if there is sufficient number of patients with transient IM status. Transient and Persistent ADA definitions are defined below and based on Shankar et al. (2014).

Transient ADA response will be defined as:

- Treatment-induced ADA detected only at one sampling time point during the treatment or follow-up observation period (excluding the last sampling time point, which will be considered persistent unless shown to be undetectable at a later time) or
- Treatment-induced ADA detected at two or more sampling time points during the treatment (including follow-up period if any), where the first and last ADA-positive samples (irrespective of any negative samples in between) are separated by a period less than 16 weeks, and the subject's last sampling time point is ADA-negative.

Persistent ADA response will be defined as:

- Treatment-induced ADA detected at two or more sampling time points during the treatment (including follow-up period if any), where the first and last ADA-positive samples (irrespective of any negative samples in between) are separated by a period of 16 weeks or longer or
- Treatment-induced ADA detected only at the last sampling time point of the study treatment period or at a sampling time point with less than 16 weeks before an ADA-negative last sample.

The sample IM incidence (number) and incidence rate (percent) at each evaluated study time point, and for the overall treatment and post-treatment evaluation period, as well as subject IM incidence and incidence rate, will be determined and appropriately summarized by treatment, as the total number of and percentage of evaluated subjects with IM negative, positive, and unknown status. Subjects with positive IM status may further be classified as transient or persistent status if applicable, and incidence and incidence rate for being transient or persistent will be appropriately summarized. Furthermore, onset, titer over time, and peak titer of the ADA response, if applicable, will be also appropriately summarized (using descriptive statistics) as median, quartiles (25% and 75%), and range and presented graphically, if deemed appropriate, by treatment at the discretion of the designated study pharmacokineticist and/or statistician (e.g., summarized at each evaluated study time point and overall; summarized by observed peak titer values from the individual IM positive subjects; etc.).

In addition to PK assessments (Section 3.8.1), selected efficacy (Sections 3.3 to 3.6) and safety (Section 3.8) assessments will also be further stratified by subject IM status (i.e., subject IM status being positive , negative or unknown) and presented in tables and/or graphically, as deemed appropriate or warranted by the designated study pharmacokineticist, medical monitor, and/or biostatistician. Other stratifications (e.g., based on antibody titer, onset of ADA, etc.) of selected PK, efficacy and safety assessments may also be performed if deemed warranted at the discretion of the pharmacokineticist, medical monitor, and/or biostatistician.

Efficacy measures to be stratified by subject IM status will include but may not be limited to TTR level, mNIS+7 and Norfolk QOL-DN score. Safety measures to be stratified by subject IM status will include but may not be limited to AEs, and lab tests for hematology and kidney functions.

3.9 Safety Analyses

All safety analyses will use the Safety Set unless otherwise specified.

Safety endpoints include the following:

- Adverse events
- Vital signs and weight
- Physical examination
- Clinical laboratory tests
- Urinalysis
- ECG
- Ophthalmology and ERG examinations
- Columbia Suicide Severity Rating Scale (C-SSRS)
- Thyroid panel

- Inflammatory panel
- Coagulation
- Complement
- Immunogenicity.

For the analysis that will be performed after all patients have completed their EOT assessment, AE tables will include events with onset date up to seven days after last doset. An additional summary table of events while on-study ("summary of all TEAEs by PT and SOC") will be created including all AEs recorded, even if onset date is more than 7 days after last dose. Unless noted otherwise, denominators for all tables will be number of subjects in the SS.

For visit based safety endpoints including laboratory assessments and vital signs, the analyses will use analysis visit windows.

3.9.1 Imputation of Missing/Partial Dates for Adverse Event

The following imputation rules will be applied to impute AE start dates under conservative principles. If the month, year, and day are missing, the adverse event start date will be imputed as the treatment start date. If month and day are missing and year is available and is the same year as in treatment start date, the month and day from the treatment start date will be used to impute the missing month and day for the adverse event start date. Otherwise, missing month and day will be imputed as January 01. If day is missing and month and year is available and the month and year are the same month and year in treatment start date, the day from the treatment start date will be used to impute missing day for the adverse event start date. Otherwise, missing day will be imputed as 01.

3.9.2 Adverse Events

Treatment-emergent adverse events (TEAEs) are defined as adverse events that first occurred or worsened after the first dose of Study Drug. An AE with a completely missing start date will be assumed to be treatment emergent. Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA). The final version used will be designated in the clinical study report. For each TEAE, the severity level of the event (mild, moderate, or severe) will be determined by physician opinion. If the maximum severity of the adverse event is greater than the baseline severity, or if the onset date/time is the same as or after the date/time of the first dose of Study Drug, then the event is considered to be treatment emergent.

In the situation where change in severity (but no change in seriousness) occurs for an adverse event, study sites are instructed to enter an end date and start a new record for the adverse event. In the new record, the changed severity is to be recorded and the start date will be set to be the end date of the previous record. Data linking those records are collected in the database. Consider three scenarios:

- The first AE record occurs prior to the first dose, the second AE record occurs on or after the first dose, and the severity increases: Only the second record will be counted as treatment-emergent.
- The first AE record occurs prior to the first dose, the second AE record occurs on or after the first dose, and the severity decreases: Neither record will be counted as treatment-emergent.
- Both records occur on or after the first dose: If the AE severity on the second record is worse than the severity on the first record, then count both records as treatment-emergent. But, if the severity improves, then only count the first record as treatment-emergent.

Of note, when counting the total number of treatment-emergent events, events linked through change in severity will still be counted as separate events.

The following categories of AEs will be summarized by treatment group: all TEAEs; TEAEs leading to early discontuation from Study Drug or from study; and serious adverse events. The percentages of patients reporting the adverse event will be summarized by MedDRA System Organ Class (SOC) and Preferred Term (PT), sorted by SOC alphabetically and then by decreasing frequency of PT within SOC.

Summaries of related TEAEs, TEAEs by maximum severity, TEAEs during the safety treatment period, and TEAEs grouped by time from first dose to event onset (< 6 months, ≥ 6 months to < 12 months, ≥ 12 months) will also be provided.

All TEAEs will be summarized for subgroups defined by the following variables: Age, Sex, Race, Region, CM-ECHO Set, V30M TTR mutation, previous treatment with Vyndaqel® or Diflunisal, disease stage. Results will not be provided for a variable if the overwhelming majority of patients are within one level of the subgroup. Non-TEAEs reported will be flagged in the all AE listing.

A listing will be provided stating which subjects reported each preferred term. Adverse events in the following categories will be summarized separately:

- Adverse events of special interest
- Other adverse events of interest

Summary tables of the number and percentage of subjects with these adverse events will be displayed split by AE category and treatment group. TEAEs of special interest and other TEAE of interest will also be summarized for patients in the CM-ECHO Set. Statistical comparison between treatment groups will be done for AEs of special interest and other AEs of interest using the risk difference (ISIS 420915 – Placebo) and associated 95% CI.

3.9.2.1 Adverse Events of Special Interest (AESI)

Adverse events of special interest (AESI) are those that require special collection and/or reporting. They are not necessarily those expected to be related to study drug or those most frequently occurring.

The following adverse events have been identified as an important medical risk:

AESI	Definition
Ocular adverse events related to vitamin A deficiency	AE with HLT: Fat soluble vitamin deficiencies and disorders; or AE with PT: Vitamin A decreased; or AE with PT: Vitamin A abnormal; or AE within the SMQ: Optic nerve disorders; or AE within the SMQ: Corneal disorders; or AE within the SMQ: Retinal disorders
Thrombocytopenia	AE with HLT: Thrombocytopenias; or AE with HLT: Platelet analyses
Renal Impairment	AE within the SMQ: Acute renal failure

3.9.2.2 Other Adverse Events of Interest

The following list includes other adverse events that have been identified to be of interest:

Other AEs of interest	Definition
Coagulation abnormalities	AE with HLT: Coagulopathies
Abnormal liver function	AE within the SMQ: Drug related hepatic disorders – comprehensive search
Adverse events at the injection site	AE with HLT: Injection site reaction; or AE with HLT: Administration site reaction NEC
Flu-like symptoms	• AE with PT Influenza like illness; or • AE with PT Pyrexia (or Feeling hot or Body temperature increased) plus at least one of the following symptoms: 1. Chills 2. Myalgia 3. Arthralgia 4. Malaise 5. Fatigue 6. Headache 7. Nausea Note that only events that start on the day of the injection or the day after injection will be included. AEs with partial or missing dates will be handled as follows. First establish whether the event is treatment emergent. If yes, then establish if the preferred terms match the ones listed in the flu-like symptoms definition. If yes, check whether the event onset dates are within the specified time frame. If at this stage, onset date is missing or partially missing, the event will be considered to have started within the required time frame.
CNS disorders	AE with SOC: Nervous system disorders
Haemorrhages	AE within the SMQ: Heamorrhages
Complement activation	AE within SMQ: Hypersensitivity

Other AEs of interest	Definition
Reduced thryroxine	AE within SMQ: Hypothyroidism

For haemorrhages results will be further classified by type of event, using the following categories: actual bleeds, hematomas/subdermal bleeds, investigations SOC (test result), and DIC, TTP. Events classified as actual bleeds or hematomas/subdermal bleeds will be further categorized by whether the event occurred at injection site or not at injection site.

3.9.2.3 Local Cutaneous Reactions at Injection Site (LCRIS)

The following MedDRA preferred terms are determined by the Sponsor's Pharmacovigilance personnel to represent the local cutaneous reaction at the injection site:

- Injection site erythema
- Injection site swelling
- Injection site pruritus
- Injection site pain
- Injection site tenderness

Only events that start on the day of injection and persist for at least two days, i.e. event onset date on the day of injection and resolution date not on the day of injection or the day after the injection, will be included. Events with onset date on the day of injection and missing resolution date will also be included. AEs with partial or missing dates will be handled as follows. First establish whether the event is treatment emergent. If yes, then establish if the preferred terms match the ones listed in the LCRIS definition. If yes, check whether the event onset dates are within the specified time frame. If at this stage, onset date is missing or partially missing, the event will be considered to have started within the required time frame. The number and percent of patients in each treatment group experiencing LCRIS will be tabulated.

Percentage of injections leading to local cutaneous reactions at the injection site will also be summarized. Percentage of injections leading to local cutaneous reactions will be calculated for each patient as (A/B)*100, where A is the number of injections with a local cutaneous reaction at the injection site, and B is the total number of injections.

3.9.2.4 Flu-Like Reactions

The following MedDRA preferred terms are determined by the Sponsor's Pharmacovigilance personnel to be the flu-like reactions:

- Influenza like illness
- Pyrexia (or Feeling hot or Body temperature increased) plus at least two of the following symptoms:
    1. Chills
    2. Myalgia
    3. Arthralgia

Only events that start on the day of injection or the day after injection will be included. AEs with partial or missing dates will be handled as follows. First establish whether the event is treatment emergent. If yes, then establish if the preferred terms match the ones listed in the flu-like reactions definition. If yes, check whether the event onset dates are within the specified time frame. If at this stage, onset date is missing or partially missing, the event will be considered to have started within the required time frame.

The number and percent of patients in each treatment experiencing flu-like reactions will be tabulated.

Percentage of injections leading to flu-like reactions will also be summarized. Percentage of injections leading to flu-like reactions will be calculated for each patient as (A/B)*100, where A is the number of injections associated with a flu-like reaction, and B is the total number of injections.

3.9.3 Vital Signs, Weight, and Physical Examination Findings

Vital signs include systolic and diastolic blood pressure, pulse, respiratory rate, and temperature. Weight will also be analyzed. Absolute values at each visit, change and percent change from baseline to each visit in vital signs and weight will be summarized by treatment group. No statistical comparisons between treatments will be performed for this summary.

All vital signs and weight will be listed. Physical examination findings will also be listed.

3.9.4 Laboratory Measurements

Laboratory tests to ensure patient safety include chemistry panel, complete blood count with differential, thyroid panel, coagulation panel, immunogenicity, inflammatory panel, complement, and urinalysis. Absolute values, change and percent change from baseline will be summarized by visit and treatment group for continuous laboratory tests. No statistical comparisons between treatments will be conducted for these summaries. The mean value (and associated standard error) will be plotted by treatment group over visits for the following laboratory parameters: platelets, eGFR, urine A/C ratio, and urine P/C ratio.

All laboratory test results will be listed. Separate listings will be provided for local and central labs. The central lab listing will be based on the ADaM dataset and will include all central lab records. The local lab listing will be based on the SDTM dataset. A separate listing for platelets will be provided that includes assessment from both the local and central labs. A separate listing will contain only values outside of normal ranges. The number and percent of patients that stopped treatment because they met a protocol-defined stopping rules for liver function, renal function, or platelet counts will be tabulated by treatment group.

Additional investiagation of heptobiliary laboratory assessments, platelet counts and renal parameters will be performed, and are detailed in the sections below. In this investigation confirmed laboratory values will also be summarized.

A confirmed laboratory value is based on consecutive lab values within 7 days. If that value is in the same or worse category the initial value is confirmed. If the consecutive value is in a better category then the initial value is confirmed using consecutive value category. If there is no retest within 7 days then the initial value is presumed confirmed. If there are multiple results on the same day (no matter from the same lab vendor or different lab vendors), then the worst value will be utilized in the analysis.

3.9.4.1 Hepatobiliary Laboratory abnormalities

The number and percentage of patients falling in each of the following categories based on post-baseline assessements will be provided:

- ALT ≥ 3xULN and total bilirubin ≥ 2xULN
- Confirmed ALT ≥ 3xULN and confirmed total bilirubin ≥ 2xULN
- ALT ≥ 3xULN and international normalized ratio > 1.5
- ALT ≥ 3xULN and total bilirubin ≥ 2xULN and ALP < 2xULN
- Hepatocellular injury
- Hepatocellular injury and total bilirubin ≥ 2xULN
- ALT ≥ 3xULN
- ALT ≥ 5xULN
- ALT ≥ 8xULN
- ALT ≥ 10xULN
- ALT ≥ 20xULN
- Confirmed ALT ≥ 3xULN
- Confirmed ALT ≥ 5xULN
- Confirmed ALT ≥ 8xULN
- Confirmed ALT ≥ 10xULN
- Confirmed ALT ≥ 20xULN
- ALT ≥ 3xULN - < 5xULN
- ALT ≥ 5xULN - < 10xULN
- ALT ≥ 10xULN - < 20xULN
- Total bilirubin ≥ 2xULN
- ALP ≥ 2xULN and baseline ALP < 2xULN or baseline ALP missing
- ALT ≥ 3x ULN or ALT ≥ 2 x baseline
- Confirmed ALT ≥ 3x ULN or confirmed ALT ≥ 2 x baseline

For patients that had ALT elevation ≥ 3 xULN, the time from first dose to first ALT elevation ≥ 3 xULN will be summarized using the following descriptive statistics: mean, standard deviation, median, P25, P75, and minimum and maximum.

Shift tables from baseline for ALT, AST, and total bilirubin based on peak (maximum) and confirmed peak category will also be provided. Categories for the ALT and AST shift tables will be < 3 x ULN, ≥ 3

x ULN to < 5 x ULN, ≥ 5 x ULN to < 8 x ULN, and ≥ 8 x ULN. Categories for the total bilirubin shift table will be < 2 x ULN and ≥ 2 x ULN.

A heptocellular injury event is defined as (ALT/ALT ULN)/(ALP/ALP ULN) ≥ 5 and ALT ≥ 3xULN, with the ALT and ALP assessments done on the same day. The categories listed above will also be analyzed using AST in place of ALT. For these analyses the definition of hepatocellular injury will not be changed to depend on AST.

3.9.4.2 Platelets

The number and percentage of patients falling in each of the following categories (using available central and local laboratory assessments) based on post-baseline assessements will be provided:

- Platelet count decrease - Grade 1a [≥ 100 10^9/L to < 140 x 10^9/L]
- Platelet count decrease- Grade 1b [≥ 75 10^9/L to < 100 x 10^9/L]
- Platelet count decrease- Grade 2 [≥ 50 10^9/L to < 75 x 10^9/L]
- Platelet count decrease- Grade 3 [≥ 25 10^9/L to < 50 x 10^9/L]
- Platelet count decrease- Grade 4 [< 25 x 10^9/L]
- Maximum toxicity grade (Grade 1a , Grade 1b, Grade 2, Grade 3, Grade 4)
- Confirmed maximum toxicity grade (Grade 1a , Grade 1b, Grade 2, Grade 3, Grade 4)
- Value < 140 x 10^9/L
- Confirmed ≥ 30% decrease from baseline
- Confirmed ≥ 50% decrease from baseline
- Confirmed value < 140 x 10^9/L
- Confirmed value < 100 x 10^9/L
- Confirmed value < 75 x 10^9/L
- Confirmed value < 50 x 10^9/L
- Confirmed value < 25 x 10^9/L

Note that a platelet value of 140 x 10^9/L is the lower limit of normal for the central laboratory. Furthermore, the platelet counts that define platelet count decrease grades are based on interactions with a regulatory authority, and align with the definitions from the the National Cancer Institute Common Terminology Criteria for Adverse Events version 4.03 (except that the CTCAE does not subdivide Grade 1 into Grade 1a and 1b).

Shift tables from baseline will be provided using the nadir value and the confirmed nadir category. Categories for the shift table will be ≥ 140 x 10^9/L, ≥ 100 x 10^9/L to < 140 x 10^9/L, ≥ 75 x 10^9/L to < 100 x 10^9/L, ≥ 50 x 10^9/L to < 75 x 10^9/L, ≥ 25 x 10^9/L to < 50 x 10^9/L, and < 25 x 10^9/L.

For each of the platelet categories investigated, time from first dose to the onset will be summarized for the patients that met the criterion using the following descriptive statistics: mean, standard deviation, median, P25, P75, and minimum and maximum. Kaplan-Meier plots for time to first event will be provided for value < 140 x10^9/L and value < 100 x10^9/L.

The post-baseline nadir (absolute value, change from baseline, and percent change from baseline) of platelet count will be summarized by treatment group.

Duration of platelet counts below the 140 x 10^9/L will be summarized, based on the duration (in weeks) each subject was below the 140 x 10^9/L .

The above analyses will be repeated for the subgroups defined by CM-ECHO Set.

3.9.4.3 Renal parameters

The number and percentage of patients falling in each of the following categories (using available central laboratory assessments) based on post-baseline assessements will be provided:

- Creatinine clearance by CKD-EPI < 90 mL/min/1.73m^2
- Creatinine clearance by CKD-EPI < 60 mL/min/1.73m^2
- Creatinine clearance by CKD-EPI < 30 mL/min/1.73m^2
- Creatinine clearance by CKD-EPI < 15 mL/min/1.73m^2
- Creatinine clearance by CKD-EPI ≥ 25% decrease from baseline
- Creatinine clearance by CKD-EPI ≥ 50% decrease from baseline
- Urine Alb/C ratio > 5 x ULN
- Urine P/C ratio > 5 x ULN
- Serum creatinine increase > 44.2 umol/l (0.5 mg/dL) from baseline
- Confirmed creatinine clearance by CKD-EPI < 90 mL/min/1.73m^2
- Confirmed creatinine clearance by CKD-EPI < 60 mL/min/1.73m^2
- Confirmed creatinine clearance by CKD-EPI < 30 mL/min/1.73m^2
- Confirmed creatinine clearance by CKD-EPI < 15 mL/min/1.73m^2
- Confirmed creatinine clearance by CKD-EPI ≥ 25% decrease from baseline
- Confirmed creatinine clearance by CKD-EPI ≥ 50% decrease from baseline
- Confirmed urine Alb/C ratio > 5 x ULN
- Confirmed urine P/C ratio > 5 x ULN
- Confirmed serum creatinine increase > 44.2 umol (0.5 mg/dL) from baseline

Shift tables from baseline for creatinine clearance by CKD-EPI will be provided usng the nadir value and the confirmed nadir category. Categories for the shift table will be ≥ 90 mL/min/1.73m^2, ≥ 60 mL/min/1.73m^2 to < 90 mL/min/1.73m^2, ≥ 30 mL/min/1.73m^2 to < 60 mL/min/1.73m^2, ≥ 15 mL/min/1.73m^2 to < 30 mL/min/1.73m^2, and < 15 mL/min/1.73m^2.

The above analyses will be repeated for the subgroups defined by CM-ECHO Set.

3.9.5 Electrocardiograms

Absolute values, change and percent change from baseline in ventricular rate and ECG intervals (PR, QRS, QT, QTcF, QTcB) will be presented by treatment and visit. Shift table from baseline to the worst (highest) post-baseline value by treatment group will be used to assess the change in the QTcF

interval. The categories for the shift table will be: ≤450 msec, >450 msec to ≤ 480 msec, >480 msec to ≤ 500 msec, and > 500 msec. The number and percent of patients experiencing an increase from baseline in QTcF interval of greater than 30 msec or 60 msec at any time post-baseline will be summarized by treatment group. This analysis will be presented overall and also by the subgroup whose QTcF was normal at baseline. Normal QTcF will be defined as ≤450 msec for males or ≤470 msec for females. The number and percent of patients with overall qualitative ECG abnormalities will also be summarized. These analyses will be repeated for patients in the CM-ECHO Set. No statistical comparisons between treatments will be performed for these summaries.

Ventricular rate and ECG intervals (PR, QRS, QT, QTcF, QTcB), as well as treatment-emergent abnormalities, will be listed. The number and percent of patients that stopped treatment because they met a protocol-defined stopping rules for QTc prolongation will be tabulated by treatment group. This tabulation will be repeated for patients in the CM-ECHO Set.

The ECG over-read data are used to support medical monitoring and will not be used for the ECG summary and analysis. A by-patient listing of ECG over-read data will be provided.

3.9.6 Electroretinograms (ERG) and Ophthalmology Exam

The change from baseline in ERG results will be summarized by treatment group.

ERG results will be listed. Ophthalmology exam findings will be listed. The number and percent of patients that stopped treatment because they met a protocol defined stopping rules for ocular effects will be tabulated by treatment group.

3.9.7 Columbia Suicide Severity Rating Scale (C-SSRS)

The C-SSRS collects binary responses to 11 categories: five subtypes of suicidal ideation, five subtypes of suicidal behavior, and self-injurious behavior without suicidal intent. Specifically, the following outcomes are C-SSRS categories and have binary (Yes/No) responses. (The categories have been re-ordered from the actual scale to facilitate the definitions of the composite endpoints and to enable clarity in the presentation of the results.)

Suicidal Ideation:

Category 1 – Wish to Be Dead

Category 2 – Non-specific Active Suicidal Thoughts

Category 3 – Active Suicidal Ideation with Any Methods (Not Plan) without Intent to Act

Category 4 – Active Suicidal Ideation with Some Intent to Act, without Specific Plan

Category 5 – Active Suicidal Ideation with Specific Plan and Intent

Suicidal Behavior:

Category 6 – Preparatory Acts or Behavior

Category 7 – Aborted Attempt

Category 8 – Interrupted Attempt

Category 9 – Actual Attempt (non-fatal)

Category 10 – Completed Suicide

Other

Category 11 – Non-suicidal Self-injurious Behavior

In addition, a numerical score, the Suicidal Ideation Score, will be defined as the highest suicide ideation category (1–5) at which the patient responded "Yes" for the given visit. If the patient did not respond "Yes" to any of these categories, the score will be set to zero.

For each of the 11 categories above, the number and percent of patients with a "Yes" response at any time post-baseline (regardless of baseline response) will be summarized by treatment group.

In addition, treatment-emergent suicidal ideation or behavior will be summarized. Treatment emergence will be identified when the event was reported at any post-baseline visit but was not present at any baseline visit. The binary categories above and the Suicidal Ideation Score will be used to identify the following 8 composite endpoints:

- **Suicidal Ideation**: A "Yes" answer at any time post-baseline to any one of the five suicidal ideation questions (Categories 1–5), regardless of the baseline response
- **Suicidal behavior**: A "Yes" answer at any time post-baseline to any one of the five suicidal behavior questions (Categories 6–10), regardless of the baseline response
- **Suicidal Ideation or Behavior**: A "Yes" answer at any time post-baseline to any one of the ten suicidal ideation or behavior questions (Categories 1–10), regardless of the baseline response
- **Treatment-Emergent Suicidal Ideation** compared to recent history: An increase in the maximum suicidal ideation score post-baseline from the baseline suicidal ideation score
- **Treatment-Emergent Serious Suicidal Ideation** compared to recent history: An increase in the maximum suicidal ideation score to 4 or 5 post-baseline from not having serious suicidal ideation (scores of 0–3) at baseline. Only patients with a baseline score of 0–3 will be considered evaluable for this outcome.
- **Emergence of Serious Suicidal Ideation** compared to recent history: An increase in the maximum suicidal ideation score to 4 or 5 post-baseline from no suicidal ideation (score of 0) at baseline. Only patients with a baseline score of 0 will be considered evaluable for this outcome.
- **Improvement in Suicidal Ideation** compared to baseline: A decrease in the suicidal ideation score at the patient's last C-SSRS assessment compared to the baseline assessment.

- **Emergence of Suicidal Behavior** compared to all prior history: The occurrence of suicidal behavior (a "Yes" response to one or more of Categories 6–10) post-baseline from not having suicidal behavior at baseline.

Each of the composite endpoints will be summarized by treatment group. For each treatment-emergent outcome listed, only those patients with the specified baseline condition will be considered evaluable. In addition, patients who discontinue from the study with no post-baseline C-SSRS assessment will be considered unevaluable for analyses of suicidality. Percents will be based on the number of evaluable patients for each outcome.

C-SSRS results will be listed.

4 Sample Size

The planned sample size for this study was revised from 195 to 135 using information published from the placebo controlled Phase 3 Diflunisal trial (Berk et al. 2013), a retrospective, multinational natural history study in 283 FAP patients (Adams et al. 2015), and uncontrolled data using another TTR mRNA targeted therapeutic (Adams 2015). It is estimated that the placebo group will have a 16 point increase in the mNIS+7 score from baseline to Month 15, and the treated group will have a 6.4 point increase in mNIS+7. The standard deviation of the change from baseline in each treatment group is estimated to be 14. With 135 patients (2:1 allocation ratio) there would be at least 90% power to detect a 9.6 point difference in the change from baseline in the mNIS+7 score between the 2 groups, with a two-sided t-test of 5% alpha, assuming that the dropout rate is approximately 25%. For the Norfolk QOL-DN questionnaire total score, it is estimated that the placebo group will have a 13.3 point change from baseline to Month 15, the treated group will have a 2.6 point change from baseline and the standard deviation of the change from baseline in each treatment group will be 18. With 135 patients, there would be at least 80% power to detect a 10.7 point difference in the change from baseline in the Norfolk QOL total score between the 2 groups, with a two-sided 5% alpha level, assuming that the dropout rate is approximately 25%.

5 Interim Analyses

5.1 Interim Analysis of TTR

An unblinded interim analysis of reduction in plasma transthyretin (TTR) level is planned after approximately the first 45 patients (total drug and placebo treated) have completed the Week 13 visit. The purpose of this analysis is futility as measured by effect on plasma TTR levels, so there will be no statistical penalty. The DSMB will inform the Sponsor whether at least 50% of patients treated with ISIS 420915 achieved either a 60% reduction in plasma TTR level or plasma TTR level below the limit of quantification (BLQ) following 13-weeks of treatment. Only a binary answer (Yes or No, this is the futility criteria) will be communicated to the Sponsor Primary Contact. Independent of the answer being communicated to the Sponsor, a separate evaluation will be performed.

At the request of the Sponsor's ISIS 420915 development partner, PPD , an unblinded evaluation of efficacy and safety will be performed by the independent statistician and reviewed by the DSMB and a small group of firewalled staff at PPD (PPD). The evaluation will include unblinded plasma TTR data following 13 weeks of treatment and cumulative safety and efficacy data on all patients currently enrolled at the time of the analysis. This data will be used by PPD for business decision making. This data will not be communicated to any other GSK employees nor to any member of the Sponsor company. Strict measures have been put in place to maintain confidentiality of the DSMB and PPD (see Section 6.0 and DSMB Charter).

Details of controlled access to the unblinded data are outlined in the DSMB Charter.

5.2 Unblinded efficacy analysis requested by the DSMB

While the study was ongoing the DSMB requested that Ionis Pharmaceuticals provide unblinded summary statistics for the primary efficacy data so that they could better evaluate benefit:risk. The request was fulfilled by PPD , a contract research organization (CRO) that is responsible for pre-programming datasets and TLFs for the Study. Strict measures were put in place to ensure this analysis did not compromise the integrity of the ongoing Study, including the formation of two PPD teams fulfil this request (unblinded and blinded team). Details about the analysis and safeguards to ensure the study integrity was not compromised are detailed in the SAP generated to support the DSMB request, availiable in Section 8.3. The DSMB package included only descriptive statistics. As no formal comparisons of efficacy endpoints were performed no adjustment to the overall study-wise type I error is needed.

6 Study Conduct to Minimize Bias

The sponsor recognizes the importance of confidentiality of interim results. To minimize any potential damage to the integrity of the clinical trial, the method by which the Sponsor will conduct the interim analysis is through the ISIS 420915-CS2 DSMB (which includes a statistician member) and an independent statistician who is distinct from the DSMB.

The independent statistician who supports DSMB and first interim analysis will be an employee of a CRO PPD , one that is independent from a second CRO conducting the trial (PPD) and a third CRO PPD who will be performing the final statistical analysis. InVentiv will also provide programming support for the DSMB, handle all DSMB related analyses, independent of the Sponsor and independent of PPD and PPD The Independent Statistician from InVentiv will maintain secure custody of blinded and unblinded data to ensure the integrity of the data. Additionally, the independent statistician will maintain all unblinded data in electronic form in a secure area. All transfer of data/reports by the independent statistician to the DSMB, which includes patient data will be by secure, trackable courier and/or secure electronic means. The independent statistician will provide all of the unblinded data to the DSMB as outlined in the DSMB Charter. Unblinded summary statistics for efficacy endpoints was requested from the DSMB while the study was ongoing (see above). PPD was responsible for producing the output, which was distributed to

Inventiv for distribution to the DSMB. Section 5.2. details the strict measures in place to ensure fulfilling the DSMB request did not compromise the integrity of the study.

All DSMB members will sign a Confidentiality Agreement with the Sponsor. In addition, all DSMB members will treat as confidential the reports, meeting discussions, minutes and recommendations of the DSMB. Strict rules for DSMB communication with the Sponsor and PPD have been put in place for the purpose of minimizing the potential for bias, as outlined in the DSMB Charter. All written communication described in the DSMB Charter will be by secure, trackable courier and/or secure electronic means. This includes dissemination of DSMB recommendations, and review of minutes of open and closed sessions between the DSMB and, as appropriate, other DSMB members, the Sponsor contacts (primary, CMO and statistics) and/or PPD.

The PPD is limited to up to three PPD employees with the seniority to make business decisions contingent on the study data. The PPD will not discuss the unblinded data/reports with other PPD staff and the Sponsor Study Team until after final database lock.

No Sponsor staff will normally participate in teleconferences between the DSMB/Independent Statistician and PPD. However, as outlined in the DSMB Charter, such participation may occur in the following circumstances: (1) if the DSMB determines that adequate discussion of the issues(s) concerned requires input from the Sponsor then the Sponsor CMO (who is the Sponsor contact for discussions involving unblinded data) may be asked by the DSMB Chair to participate in a teleconference with Partnered Firewalled Staff and (2) if it has been necessary for the DSMB to share data with the Sponsor CMO then the Sponsor CMO may convene a teleconference between the DSMB and Partner Firewalled Staff.

During the conduct of the study, the Sponsor will not have access to any efficacy, pharmacodynamic or exploratory data, except for baseline values. The efficacy data will be collected and stored by independent CROs as detailed below. All CROs will maintain secure custody of their databases. The CROs will not transfer any efficacy data other than baseline data to the Sponsor until after all patients have completed the treatment period and the database has been locked and the study is unblinded. The efficacy data includes the following: mNIS+7, NIS+7, Norfolk QOL-DN, BMI/mBMI, plasma retinol, plasma retinyl palmitate, plasma TTR, plasma RBP4, plasma NT-proBNP, SF-36 questionnaire, ECHO efficacy parameters, NSC and PND scores.

For the purpose of pre-programming and data cleaning, Parexel and an independent data manager at Trennic Data Services will receive post-baseline data for the following endpoints: mNIS+7, NIS+7, Norfolk QOL-DN, BMI/mBMI, SF-36 questionnaire, ECHO efficacy parameters, NSC and PND scores.

The primary efficacy assessment, mNIS+7 scores, will be collected and stored by an independent contract research group, PPD that is under the direction of PPD. The NSC score is obtained during the NIS assessment procedure and is also collected and stored by PPD. The mNIS+7 results from each site will be faxed to PPD for processing and quality assurance. Faxed copies will be maintained in secure rooms within locked cabinets. The NIS and NSC data are stored in the PPD (a database). The

other components of the mNIS+7 (nerve conduction, sensory testing and heart rate to deep breathing) are entered by the PPD into a firewalled portion of the study EDC system. PPD only has access to this portion of the EDC system and the Sponsor and clinical sites do not have access to the PPD portion of the EDC. The mNIS+7 summated score will not be shared with the sites. Up until the database lock, the Sponsor will only have access to patient baseline values of the mNIS+7.

The Norfolk QOL-DN, body weight (needed to calculate BMI/mBMI), SF-36 questionnaire and PND scores will be entered into the EDC system by each site. The independent CRO PPD is contracted to develop and maintain secure custody of the EDC database. During the conduct of the study the Sponsor will receive regular data transfers from PPD but without the above mentioned efficacy data included (except for baseline values).

The plasma retinol, plasma retinyl palmitate, plasma TTR, plasma RBP4, hsCRP, and plasma NT-proBNP samples will be assayed at PPD, the central laboratory contracted for this study. The results will be maintained in PPD's secure database. Neither the Sponsor nor the sites will receive the results from the above mentioned tests (except for baseline values). During the conduct of the study the Sponsor will receive regular lab data transfers from PPD to perform safety assessments, but the above mentioned data will not be included in the transfers.

The ECHO data will be collected, analyzed and stored in a secure database by an independent CRO (PPD). The sites will upload the ECHO data on a secure web-portal for analysis by PPD. Up until the database lock, the Sponsor will only have access to patient baseline ECHO values. ECHOs conducted for safety assessment purposes will be available to the DSMB, Sponsor, and Investigator.

Prior to unblinding of the final analysis, any ISIS 420915 concentration data sets provided to Ionis by the bioanalytical lab will be provided without reference to actual patient identifiers to avoid inadvertent or accidental unblinding. The bioanalytical lab may provide Ionis with data sets containing false patient identifiers unrelated to the actual identifiers to allow review of the PK data.

In conclusion, the unblinding process for the interim analysis and periodic safety reviews has been clearly defined and detailed roles and responsibilities to the independent statistician, DSMB, PPD and Sponsor, such that the potential for bias to enter into the conduct of the study will be minimized. Additional steps to ensure the data for the primary and secondary endpoints remain blinded, such as housing the primary and secondary efficacy data in databases to which the Sponsor has no access, further safeguards the potential for Sponsor bias.

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8 Appendix

8.1 Appendix 1 Components and Subcomponents of the mNIS+7, and NIS+7

Figure 2 Components and Subcomponents of the NIS+7

Table 1: Neuropathy Impairment Score

Component	Subcomponent	Right Side	Left Side	Max Score	Max Sub-Totals	Missing value imputation for Component Score
[redacted]	[redacted] C [redacted]	[redacted]	[redacted]	[redacted]		[redacted]
	[redacted] CI [redacted]	[redacted]	[redacted]	[redacted]		
	[redacted]	[redacted]	[redacted]	[redacted]		
	[redacted]	[redacted]	[redacted]	[redacted]		
	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]		[redacted]
	[redacted]	[redacted]	[redacted]	[redacted]		
	[redacted]	[redacted]	[redacted]	[redacted]		
	[redacted]	[redacted]	[redacted]	[redacted]		
	[redacted]	[redacted]	[redacted]	[redacted]		
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	[redacted]	[redacted]	[redacted]	[redacted]		
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	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]		[redacted]
	CCI [redacted]	[redacted]	[redacted]	[redacted]		
	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	

Component	Subcomponent	Right Side	Left Side	Max Score	Max Sub-Totals	Missing value imputation for Component Score
	CCI					

Table 2: Modified +7 Score

Component	Subcomponent	Max Score	Sub-Totals	Missing value imputation for Component Score
CCI [redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]		[redacted]
	[redacted]	[redacted]		
	[redacted]	[redacted]		
	[redacted]	[redacted]		
	[redacted]	[redacted]	[redacted]	
[redacted]	[redacted]	[redacted]		[redacted]
	[redacted]	[redacted]		
	[redacted]	[redacted]		
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	[redacted]	[redacted]		
	[redacted]	[redacted]	[redacted]	
[redacted]			[redacted]	

Table 3: +7 Score

Component	Subcomponent	Max Score	Max Sub-Totals	Missing value imputation for Component Score
CCI [redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]		[redacted]
	[redacted]	[redacted]		
	[redacted]	[redacted]		
	[redacted]	[redacted]		
	[redacted]	[redacted]	[redacted]	
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]			[redacted]	

8.2 Appendix 2 Scoring of Assessment Instruments

Neuropathy Symptoms and Change (NSC)

The NSC questionnaire consists of 38 questions asking about different symptoms.

A symptom is marked as present if in the judgment of the examining neurologist, it occurs more frequently or more severely than in healthy persons of the same age and gender and is due to neuropathy. If a symptom is present its severity is graded as 1 (slight +), 2 (moderate ++) or 3 (severe +++). If the symptom is not present it is given a score of 0.

Two questions (35 and 36) are only answered by men. These are not included in the score for women.

The questions in the NSC can be divided into the following domains

- Muscle weakness: Questions 1-19
- Sensory (hypo / loss of sensation): Questions 20-22
- Sensory (paresthesia, hyper sensation): Questions 23 - 29
- Autonomic (GI/urinary incontinence): Questions 31, 32, 33, 34
- Autonomic (other than GI/urinary incontinence): Questions 30, 35-38 for men
  Questions 30, 37-38 for women

The Muscle weakness domain is also divided into 4 sub-domains:

- Head and Neck: Questions 1-6
- Chest: Questions 7-9
- Upper Limbs: Questions 10-15
- Lower Limbs: Questions 16-19

For each sub-domain and domain the total score is obtained by summing the relevant questions. The maximum score is therefore 57 for muscle weakness, 9 for Sensory (hypo / loss of sensation), 21 for Sensory (paresthesia, hyper sensation), 12 for Autonomic (GI/urinary incontinence), 15 for Autonomic (other than GI/urinary incontinence) in men and 9 for Autonomic (other than GI/urinary incontinence) in women . The minimum score is zero for each domain.

The NSC total score is the sum of the scores across all 5 domains. The minimum NSC total score is therefore 0 and the maximum NSC total score is 114 for men and 108 for women.

For questions 20-29 the location affected is also collected on the questionnaire but this information is not used in the calculation of the domain or total scores.

The NSC questionnaire also records a change score for the change in symptoms compared to the week before study onset. These are scored as follows

-3	=worse ---
-2	= worse --
-1	= worse -
0	= no change
+1	= better +
+2	= better ++
+3	= better +++

8.3 DSMB SAP

Statistical analysis plan for the unblinded analysis of efficacy endpoints - A request from the DSMB

ISIS 420915-CS2

A Phase 2/3 Randomized, Double-Blind, Placebo-Controlled Study to Assess the Efficacy and Safety of ISIS 420915 in Patients with Familial Amyloid Polyneuropathy

Date: August 31, 2016

Version: 1.0

Statistical Analysis Plan Signature Page

Ionis Pharmaceuticals, Inc.

2855 Gazelle Court

Carlsbad, CA 92010

Protocol:	ISIS 420915-CS2
Study Title:	A Phase 2/3 Randomized, Double-Blind, Placebo-Controlled Study to Assess the Efficacy and Safety of ISIS 420915 in Patients with Familial Amyloid Polyneuropathy
Issue Date:	May 13, 2016 (Amendment 9)

Signature: _____ Date: _____

PPD , Dr.P.H.

CCI , Biometrics

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Signature: _____ Date: _____

CCI , Ph.D.

CCI , Biometrics

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Signature: _____ Date: _____

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1 Introduction

1.1 Overview

This document describes the analysis plan for fulfilling the DSMB's request for an unblinded analysis of the primary efficacy endpoints. The analysis plan covers the two primary efficacy endpoints (Norfolk QOL-DN Total score and mNIS+7 Composite score) and NIS Composite score, a secondary efficacy endpoint that was not requested by the DSMB. NIS Composite score is provided because the Sponsor believes it can aide in the evaluation of ISIS 420915; additional considerations for this endpoint are provided below.

This analysis plan is aligned with the current working version of the ISIS 420915 CS2 statistical analysis plan (SAP); deviations from the SAP and protocol are noted.

1.2 Endpoints

Norfolk QOL-DN, mNIS+7 and NIS are measured at baseline, Week 35, and Week 66 (EOT efficacy assessment). Patients who do not enroll in the OLE will also have efficacy assessments measured at Week 91 in the post-treatment evaluation period. The mNIS+7 assessments at baseline and Week 66 are performed twice and the duplicates averaged. The duplicates cannot be performed on the same day.

NIS is provided to aide in the evaluation of potential the benefit of ISIS 420915 in patients with FAP. The Sponsor has reservation about the mNIS+7 data because the m+7 component relies on a complicated scoring algorithm and currently the dataset is not complete. For this reason the Sponsor is providing results for NIS Composite score, the other major component in the mNIS+7 Composite score. The Sponsor believes this data is more complete based on our evaluation of the baseline data, and can inform the assessment of the benefit of ISIS 420915.

2 Procedures

2.1 Analysis and distribution of the output

The Sponsor is committed to ensure study integrity will not be compromised in fulfilling the DSMB request for an unblinded analysis of efficacy data from an ongoing study.

Analysis of the unblinded efficacy data will be done by PPD, a contract research organization (CRO) that is responsible for pre-programming datasets and TLFs for the Study. To limit the possibility that PPD compromises the study integrity, two separate PPD teams will be involved in fulfilling the DSMB request. One team (Unblinded Team) will only be responsible for analyzing the efficacy data using the real randomization codes. This team will consist of an unblinded statistician and at least one unblinded programmer. The other team (Blinded Team), which is responsible for

supporting the ongoing study and has routine contact with the Sponsor, will remain blinded and will prepare the endpoints for the Unblinded Team to analyze using dummy treatment codes. The Unblinded Team ensures blinding is maintained and prevents unauthorized access of unblinded data to the Blinded Team until the Study is unblinded. Unblinding information is to be stored and accessible in a restricted area of the PPD network, with access restricted to the Unblinded Team.

Real treatment codes will be sent from the Sponsor to the unblinded statistician at PPD by Ionis QA/C, and will be done in accordance with Ionis SOPs.

Output generated from the Unblinded Team will be sent to Inventiv, a CRO that works with the DSMB, who will then distribute it to the DSMB for review. As part of the QC process, the Blinded Team will send baseline composite scores for mNIS+7, Norfolk QOL-DN and NIS to the Sponsor to verify to scoring was implemented correctly. Post-baseline composite scores are not to be sent to the Sponsor for verification since the Sponsor is to be firewalled from post-baseline efficacy data.

2.2 Data management

The Sponsor is firewalled from post-baseline efficacy and PD data, including Norfolk QOL-DN and mNIS+7. Up until the database lock and unblinding for the end of treatment analysis, the Sponsor will only have access to patient baseline values. Efficacy data was transferred from the different study vendors to PPD. Information on the efficacy dataset is detailed in the table below.

	Vendor	Date PXL received data	Most current date in database
NIS data	CCI		
m+7 data	CCI		
Norfolk QOL-DN	CCI		

3 Analytical Plan

3.1 Analysis conventions

3.1.1 Baseline definition

Baseline will be defined as follows:

- Norfolk QOL-DN: Last non-missing value prior to the first dose of Study Drug
- mNIS+7 and NIS: Defined as the average of two assessments taken within 45 days prior to the first dose of Study Drug. If only one assessment has been done, the single assessment will be used in place of the average. Rarely, the baseline treatment mNIS+7 assessment(s) (or a subset of this assessment) will have been completed early in the treatment period rather than pre-treatment. These assessments will be included in the analysis as valid

baseline assessments provided they are taken within one week after the first dose. The rationale for this is that the pharmacology of the drug indicates that the drug will have no effect on mNIS+7 this early in treatment, and including these values as the baseline assessments will allow these patient's data to be included in the analysis.

3.1.2 Analysis population

Unblinded efficacy data will be analyzed for the full analysis set (FAS), defined as all randomized patients who received at least 1 injection of Study Drug (ISIS 420915 or placebo) and who have a baseline and at least one post-baseline efficacy assessment for mNIS+7 or Norfolk QOL-DN. Results will be summarized under the treatment to which patients were randomized.

3.1.3 Analysis windows

Data will be assigned to a nominal visit according to the visit windows in the table below. Because mNIS+7 may be implemented over several days, window definitions are based on the study day the assessment was initiated. Assessments that occurred more than 52 days after the last dose of medication will not be summarized.

Nominal Visit (Target Day)	Window (Day)
Week 35 (Day 239)	209-269
Week 66 (Day 456)	411-501

3.1.4 Scoring of assessment instruments

Norfolk QOL-DN: Norfolk QOL-DN consists of one composite score (Total QOL) and five sub-domain scores (physical functioning/large fiber neuropathy, activities of daily living, symptoms, small fiber neuropathy, and autonomic neuropathy). The scoring of the Norfolk QOL-DN will be conducted according to the scoring manual developed at the Eastern Virginia Medical School. The Norfolk QOL-DN Total score will be calculated by summing the domain scores.

The range for Norfolk QOL-DN Total score is -4 to 124, with large values being less favorable than smaller values.

NIS and mNIS+7: mNIS+7 consists of two composite scores: CCI

CCI

CCI

CCI

3.2 Analysis

3.2.1 Considerations for the interpretation of unblinded efficacy results

The following should be noted:

- Output is based on data (baseline and post-baseline) that have not been cleaned and will be incomplete for some patients. For example, some patients have not yet reached the Week 66 assessment, and some patients will have missing data that will ultimately be entered during the normal database cleaning process. This is particularly relevant to the mNIS+7 endpoint. PPD does not enter any data into the database until all queries have been resolved. Thus, there is a significant amount of mNIS+7 data that has been received by PPD but not yet entered and therefore will be missing for this analysis. There are also circumstances where the test was done by the site, but not yet sent to PPD. Conclusions drawn from the data should therefore be done with appropriate caution.
- There will be fewer subjects with Week 66 data included in the unblinded efficacy analysis than what is anticipated when the end of treatment analysis is performed, meaning that there will be greater degree of uncertainty in characterizing the benefit of ISIS 420915.
- Because the Sponsor is firewalled from post-baseline efficacy data, the Sponsor cannot attest to the accuracy of post-baseline composite scores including change and percent change from baseline.

3.2.2 Endpoints

Endpoints that will be summarized in the FAS include:

- Change and percent change from baseline to Weeks 35 and 66 in Norfolk QOL-DN Total score
- Change and percent change from baseline to Weeks 35 and 66 in mNIS+7 Composite score
- Change and percent change from baseline to Weeks 35 and 66 in NIS Composite score

These endpoints will be summarized descriptively by sample size, mean, standard deviation, data quartiles (25^{th}, 50^{th} and 75^{th}), and min and max values. Neither the MMRM that is prespecified for the primary efficacy analysis nor other inferential statistical methods will be fit to the interim data.

3.2.3 Sensitivity analysis of the endpoints

Because the Study is ongoing and some patients have not been in the Study long enough to have a Week 66 efficacy assessment (the primary study visit), efficacy data will be summarized for the subset of the FAS that are Week 66 evaluable. A patient in the FAS is Week 66 evaluable if they:

1. Had a Week 66 assessment on mNIS+7, NIS or Norfolk QOL-DN; or
2. Did not have a Week 66 assessment on mNIS+7, NIS or Norfolk QOL-DN but either:

a. Discontinued from the Study and had their first dose of Study Drug on or before 28Jun2015; or
b. Had their first dose of Study Drug on or before 30Mar2015.

The 28Jun2015 and 30Mar2015 dates are based on the Week 66 analysis window, and the most current date in the efficacy dataset analyzed (12AUG2016).

3.2.4 Handling of missing assessment level data

3.2.4.1 Norfolk QOL-DN

Missing assessment level data will be imputed according to the following rules:

- For each patient at a specific visit, if at least 50% of the questions for a domain (physical functioning/large fiber neuropathy, activities of daily living, symptoms, small fiber neuropathy) are not missing or if at least one question is not missing for autonomic domain, the missing questions are imputed as follows: If any question is missing at baseline, the mean value for this question at baseline from the study population (across all treatment groups) will be used to impute the missing baseline question value. For post-baseline visits during the treatment period, any missing question values will be imputed using the last observed or imputed question value (including baseline value). For the symptom domain, in the case that a patient responded on a particular question (Questions 1- 7) as not a having the symptom but also marked presence of the symptom in their feet, legs, hands, or arm, the question will be set to missing and the imputation rules will be followed.
- Otherwise, the total for that domain will be set to missing

The Norfolk QOL-DN Total score will be calculated by summing the imputed domain scores. If any domain score after imputation is still missing, then the Norfolk QOL-DN Total score will be set to missing.

The Norfolk QOL-DN individual domain and Total score will only be calculated for visits where the patient had a Norfolk QOL-DN assessment. The individual domain and Total score are set to be missing if a patient misses the visit or does not have a Norfolk QOL-DN assessment at that visit.

3.2.4.2 mNIS+7 and NIS

- **Missing data imputation strategies for missing assessment level data**

  Two independent assessments of the primary efficacy endpoint, mNIS+7, are planned at the baseline visit and the Week 66 visit. A single mNIS+7 assessment is also planned at the Week 35 visit. The mean of the two replicate assessments within visit will be used for analysis of both the baseline and Week 66 visits (provided both visits fall in the visit window and are within 52 days of the last dose of medication). Sub-component scores will be averaged first. These will be referred to as the Averaged Sub-component scores.

  At baseline and week 66, in the event that only one sub-component has been performed, the single sub-component will be used in place of the mean value for that visit for the Averaged Sub-component score. If both of the sub-component values are missing, the

Averaged Sub-component score is missing. At week 35, only one assessment is performed, therefore the single sub-component will be used as the Averaged Sub-component score for that visit. These values will be used in the summary and analysis of Averaged Sub-component scores.

The component scores will be computed by summing the Averaged Sub-component scores and the composite scores will be computed by summing the component scores.

- **Imputation of missing averaged sub-components**

If a patient has completed at least part of the mNIS+7 at a visit then the following imputation method will be used to impute this missing assessment level data for the purposes of determining component scores for summary and analysis.

  - If at least 50% of Averaged Sub-component scores within a component are available, the missing Averaged Sub-component scores will be set to equal to the mean of the patient's other non-missing Averaged Sub-component scores in the component. The component score is then calculated.
  - Otherwise, the component will be considered to be missing.

- **Composite Score**

The composite scores of mNIS+7 and NIS will each be calculated by summing the imputed component scores. If any of the component scores after imputation are still missing within a composite, the composite score will be set as missing.