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Statistical Analysis Plan

Protocol No.: 1VIT15042

Randomized, placebo-controlled, multi-center trial of ferric carboxymaltose in Restless Legs Syndrome patients with iron deficiency anemia

NCT Number: 02826681

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STATISTICAL ANALYSIS PLAN APPROVAL

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LIST OF ABBREVIATIONS AND DEFINITIONS

AST	Aspartate Aminotransferase Test
ALT	Alanine Transaminase Test
ATC	Anatomical Therapeutic Chemical
ANCOVA	Analysis of Covariance
BP	Blood Pressure
BUN	Blood Urea Nitrogen
CBC	Complete Blood Count
CGI-I	Clinical Global Impression- Improvement
CHDQ	Cambridge-Hopkins Dianostic Questionnaire
CRF	Case Report Form
CI	Confidence Interval
DSMB	Data and Safety Monitoring Board
FCM	Ferric Carboxymaltose
Hct	Hematocrit Test
Hgb	Hemoglobin
HTDI	Hopkins-Hening Telephone Diagnostic Interview
HRSQ	Hopkins RLS-Sleep Quality Questionnaire
IDA	Iron-deficiency aAnemia
IRLSS	International Restless Legs Syndrome Severity Scale
IRT	Interactive Response Technology
IV	Intravenous
LDH	Lactate Dehydrogenase
MCH	Mean Cell Hemoglobin
MCHC	Mean Corpuscular Hemoglobin Concentration
MCV	Mean Corpuscular Volume
MedDRA	Medical Dictionary for Regulatory Activities
mg	Milligrams
mL	Milliliter
MOS	Medical Outcomes Survey for Sleep
MRI	Magnetic Resonance Imaging
PCS	Potentially Clinically Significant
PE	Physical Examination
PGI-I	Patient Global Impression-Improvement
PT	Preferred Term
QSM	Quantitative Susceptibility Mapping
RBC	Red Blood Cells
RDW	Red Cell Distribution Width Test
RLS	Restless Legs Syndrome
SAE	Serious Adverse Event
SAP	Statistical Anlaysis Plan
SOC	System Organ Class
SD	Standard Deviation
TEAE	Treament Emergent Adverse Event

TIBC	Total Iron Binding Capcacity
TSAT	% Transferrin Saturation
VS	Vital Sign
WBC	White Blood Cells
WHO	World Health Organization

1. INTRODUCTION

The purpose of this statistical analysis plan (SAP) is to describe the procedures and the statistical methods that will be used to analyze and report results for Protocol 1VIT15042 (Date: 4 September 2019). This SAP should be read in conjunction with the study protocol and case report forms (CRFs).

1.1 STUDY OBJECTIVES

1.1.1 Primary Objective(s)

Treatment Phases I and II primary objective is to evaluate the efficacy and safety of FCM (750 mg dose x 2) for treatment of Restless Legs Syndrome (RLS) in patients with iron-deficiency anemia (IDA).

Long-term extension Phase III primary objective is to evaluate the duration of effect of prior FCM treatment and to determine the effectiveness of further iron repletion with FCM when RLS symptoms worsen or reoccur.

1.2 STUDY ENDPOINTS

1.2.1 Primary Efficacy Endpoint(s)

Primary efficacy endpoint in **Treatment Phase I** is to determine the effectiveness of FCM in improving RLS severity associated with IDA. Effectiveness will be evaluated by:

- IRLSS score change from baseline to Day 42 with comparison between FCM vs. Placebo.
- CGI-I at Day 42.

Primary efficacy endpoint in **Treatment Phase II** is to determine the effectiveness of an additional unblinded single FCM treatment in improving RLS severity in non-responders from Treatment Phase I. Effectiveness will be evaluated by:

- IRLSS score change from baseline (For Phase II the "baseline" is established on Day 42 of Phase I) compared to Day 42 of Treatment Phase II
- CGI-I at Day 42 in Treatment Phase II.

Primary efficacy endpoint in **Long-Term Extension Phase III** is to evaluate the duration of effect of FCM as defined as the number of weeks from the initial treatment without need for further treatment. Initial treatment is defined as Day 0 of Phase I for Responders in Phase I and Day 0 of Phase II for Responders in Phase II.

1.2.2 Secondary Efficacy Endpoint(s)

Treatment Phase I and Phase II:

- Change in IRLSS score from Baseline to Day 42. (Treatment Phase I and II)
- IRLSS score change from Baseline (For Phase II the "Baseline" is established on Day 42 of Phase I) compared to Day 42 of Treatment Phase II.
- CGI-I and PGI-I at each visit and percentage of subjects who are much or very much improved at Day 42. (Treatment Phase I and II)
- Change in sleep quality (MOS Sleep) from Baseline to Day 42. (Treatment Phase I and II)
- Estimate the proportion of treatment non-responders in Phase I who respond to FCM in Phase II for each randomized treatment group. (Phase II only)
- Change in substantia nigra iron index from Baseline to Day 42. The index will be determined using measures of MRI relaxation rate (R2*) or Quantitative Susceptibility Mapping (QSM). (Treatment Phase I and II)
- Change in iron index (R2* or QSM) for thalamus and motor cortex from Baseline to Day 42. (Treatment Phase I and II).

Treatment Phase III:

- Change in IRLSS total score from baseline to each scheduled visit.
- Number (and percentage) of subjects who remain in the study to Week 52.

1.2.3 Safety Endpoints

Safety endpoints include:

- Adverse events
- Laboratory assessments, including hematology (Hgb, Hct, RBC, WBC, MCV, MCH, MCHC, RDW, platelets, differential count), iron indices (serum iron, serum ferritin, and total iron binding capacity (TIBC), percentage serum transferrin saturation (TSAT)), clinical chemistry (sodium, potassium, chloride, BUN, creatinine, albumin, alkaline phosphatase, total bilirubin, gamma-GGT, AST, ALT, LDH, calcium, phosphorus, glucose, bicarbonate)
- Vital signs

1.3 SUMMARY OF THE STUDY DESIGN

1.3.1 General Study Design and Plan

This is a Phase II, randomized, placebo-controlled multi-center study to evaluate the efficacy and safety of Injectafer® (FCM Injection) for treatment of Restless Leg Syndrome (RLS) with iron-deficiency anemia (IDA).

FCM is a stable Type I polynuclear iron (III)-hydroxide carbohydrate complex developed as an intravenous (IV) iron replacement therapy for the treatment of IDA[1]. All subjects who meet the inclusion requirements and no exclusion criteria will qualify to enter the screening phase. The study will include 3 treatment phases, i.e. Phase I (42 days), II (42 days) and III (46 weeks long-term extension), and will enroll 70 subjects to be randomized into Treatment Phase I.

In **Treatment Phase I**, all eligible subjects will be randomized in a 1:1 ratio to receive a blinded dose of either FCM 750 mg undiluted slow IV push at 100 mg/minute or a Placebo (15 ml of Normal Saline) IV push at 2 ml/minute on Days 0 and 7. At the end of treatment Phase I (i.e. Day 42), all subjects will be defined as either treatment responders or non-responders defined as:

> Responder: IRLSS total score ≤10, or IRLSS total score >10 with a CGI-I score of much or very much improvement and the patient does not request further treatment for RLS.
>
> Non-Responder: if neither of these above criteria are met.

In **Treatment Phase II**, Treatment Phase I Non-Responders who have met the necessary laboratory criteria (ferritin <300 ng/mL and a TSAT <45%) will be consented for enrollment in Treatment Phase II. Day 0 of treatment Phase II will occur within 7 days of the completing Treatment Phase I Day 42 visit. These subjects will receive an unblinded 750 mg dose of FCM, treatments will be given as an undiluted slow IV push at 100 mg/minute on Days 0 and 7 of Phase II consequently. Treatment Phase I Non-Responders who do not meet the laboratory criteria for additional dosing will be discontinued from the study and treated for RLS as deemed appropriate by the referring physician. Subjects completing Treatment Phase II will be re-evaluated on Day 42 and defined as either a Responder or Non-Responder, using the same criteria described above. Treatment Phase II Non-Responders will be discontinued from the study after final assessments are complete.

In **Treatment Phase III**, responders from either Treatment Phase I or Treatment Phase II will continue through into this 46-week long-term extension phase of the study, and be monitored and assessed by phone for RLS symptoms (IRLSS and HRSQ) and adverse events on Weeks 15, 25, 34, 43 and 52 (final follow-up visit) from treatment Phase I Day 0. During Phase III, subjects may receive additional unblinded treatments (i.e. a single FCM 750 mg undiluted slow IV push at 100 mg/minute) if at any time the subject reports worsening of RLS symptoms (an increase >4 points on the IRLSS compared to the last evaluation captured for that subject) and laboratory criteria are met (ferritin <300 ng/mL and a TSAT <45%). No additional treatment will be allowed after the Week 46. A final face-to-face study visit will occur on Week 52. If a clinic visit is not possible, final evaluation will be completed by phone.

The Schedule of Events for Phases I, II and III are presented as Table 1.1 and Table 1.2

Table 1.1 Schedule of Events Treatment Phase I and Treatment Phase II

	SCREENING	Treatment Phase I and Treatment Phase II				
STUDY VISIT PROCEDURES	**-14**	**DAY 0**	**DAY 7+1**	**DAY 14+3**	**DAY 28+3 (PHONE CONTACT)**	**DAY 42+3**
Informed Consent	X					X**
Eligibility	X	X				X
Medical History, including prior iron therapy	X	X				
RLS History	X	X				
D/C oral iron products	X					
Site contact IRT	X					
Physical exam	X					X
Vital Signs	X	X	X			X
Height and Weight without shoes	X					
CHDQ	X					
HTDI	X					
IRLSS	X	X			X	X
HRSQ		X				X
CGI-I						X
PGI-I						X
MOS Sleep Scale		X				X
Hematology	X	X	X	X		X
Iron Indices	X	X	X	X		X
Chemistry	X	X	X	X		X
Vitamin D		X	X	X		X
Erythropoietin		X				X
Trace Element		X				
Urine Pregnancy test	X					
Study Drug		X	X			
Brain MRI	X					X
Concomitant Meds	X	X	X		X	X
Adverse Events		X	X		X	X

**** All Subject will be consented to participate in Treatment Phase II prior to any study procedures being completed**

Table 1.2 Schedule of Events Long-Term Extension Phase III

STUDY VISIT PROCEDURES	**Week 15 Phone Contact**	**Week 25 Phone Contact**	**Week 34 Phone Contact**	**Week 43 Phone Contact**	**Week 52 EOS**
IRLSS	X	X	X	X	X
HRSQ	X	X	X	X	X
MOS / PGI-I					X
Concomitant Meds	X	X	X	X	X
Adverse Events	X	X	X	X	X
Physical Exam / VS	**See protocol Section 6.6 for procedures for additional FCM dosing**				
Iron Indices / Chemistries / Hematology					X
FCM Dosing					

1.3.2 Randomization and Blinding

On Day 0 of **treatment Phase I**, all eligible subjects will be randomized via IRT system in a 1:1 ratio to receive a blinded dose of either FCM 750 mg undiluted slow IV push at 100 mg/minute or a Placebo (15 ml of Normal Saline) IV push at 2 ml/minute.

Other than designated randomization personnel, all subjects, investigators, and study conduct personnel are blinded to study drug assignment. The blinded investigators are also blinded to the IRLSS total score and PGI-I score for each subject after randomization. All study personnel will be blinded to the post treatment iron indices and phosphorous (after randomization) throughout the study.

1.3.3 Sample Size and Statistical Power Considerations

Enrollment was planned for 70 randomized subjects (35 per treatment group in Phase I). Based on Study 1VIT05009 in patients without IDA, approximately 50% of patients in the FCM group and 15% of patients in the placebo group are anticipated to have much or very much improvement on the CGI-I. The proposed sample size provides >85% power to detect this difference with the chi-square test. The mean treatment difference (and standard deviation) for change from baseline to Day 42 for IRLSS is anticipated to be 5.0 (7.3). The proposed sample size provides >80% power to detect this difference with a two-sided t-test with alpha=0.05.

2. STATISTICAL METHODS

2.1 GENERAL CONSIDERATIONS

Analysis datasets will be produced according to CDISC standards. All study-collected data will be summarized in tables or graphs. Listing will be produced when appropriate and all ICH-required listings will be produced. In general, continuous variables will be summarized by number of subjects, mean, standard deviation (SD), median, minimum, and maximum values. Categorical variables will be summarized by number and percentage of subjects in each category

In general, summary tables will present data by treatment group (for Phase I) and treatment phase.

Baseline definitions:

- Phase I: baseline (other than iron index) is the last non-missing value prior to receiving randomized study medication.
- Phase II: phase II baseline (other than iron index) is the last non-missing value prior to receiving unblinded FCM in Phase II.
- Phase I and Phase II: baseline for iron index measured by MRI is measured in screening.
- Phase III: phase III baseline is defined as Day 0 of Phase I for responders in Phase I and Day 0 of Phase II for responders in Phase II.

2.1.1 Reporting Precision

Summary statistics will be presented to the following degree of precision, unless otherwise specified:

Statistics	Degree of Precision
Mean, Median, Quartiles,	One decimal place more than the raw data.
Standard deviation	Two decimal places more than the raw data.
Minimum, Maximum	The same as the raw data.
Percentage	One decimal place. A percentage of 100% will be reported as 100%. Percentages of zero will be reported as 0.

For weight and height, one decimal place will be used for summary statistics. Fractional numeric values will be presented with a zero to the left of the decimal point (for example, 0.12 – 0.30).

2.2 DEFINITIONS OF ANALYSIS POPULATIONS (ANALYSIS SETS)

2.2.1 ITT Population

ITT population includes all the subjects who were randomized.

Summaries for ITT population will be based on the treatment group to which they were randomized.

2.2.2 Safety Population

Safety population includes all the subjects who were randomized and received at least one dose of study medication.

Summaries for safety population will be based on the actual treatment they received.

2.3 TIME WINDOWS FOR ANALYSIS

No visit window will be constructed. For analyses, only scheduled visits will be summarized in tables.

2.4 HANDLING OF MISSING DATA

Missing data will not be imputed for descriptive statistical summaries in safety or efficacy analyses.

For the PGI-I score, missing data at Day 42 will be considered as failure in analysis.

Dates missing the day or both day and month of the year will adhere to the following conventions to classify TEAE and to classify prior/concomitant medications.

- A medication with a completely missing start date will be considered a prior medication. A medication with a completely missing stop date will be considered a concomitant medication.
- If complete AE onset date is missing and AE end date is on or after the first dose date then it will be counted as a treatment emergent AE (TEAE) for the study. If complete AE onset date is missing and AE end date is before the first dose date then it will be counted as a not treatment emergent AE (TEAE) for the study.
- If an AE or a medication has a partial missing start or stop date, the following rules will be used for imputation:
    - If year is present but month and day are missing, impute start date as January 1 of that year or first dose date if the year is the same as the year of first dose date and impute stop date as December 31 of that year.
    - If year and day are present but month is missing, impute start month as January or the month of the first dose date if the year and day is the same as the year and day of first dose date and impute stop month as December.
    - If year and month are present but day is missing, impute start date as first day of that month or first dose date if the year and month are the same as the year and month of first dose date and impute stop date as last day of that month.

2.5 ANALYSIS SOFTWARE

All summaries and statistical analyses will be generated using SAS® version 9.4 or higher.

3. STUDY SUBJECTS

3.1 DISPOSITION OF SUBJECTS

Disposition will be summarized for all randomized subjects.

The disposition will include the following:

- Subjects who are randomized (for Phase I only)
- Subjects who are in Safety Population
- Subjects who are in ITT population
- Subjects who complete the study
- Subjects who discontinue the study

The number and percent of subjects will be summarized for each reason for premature discontinuation.
A listing of dispositions will be provided for all subjects.

3.2 PROTOCOL DEVIATIONS

The clinical team will identify deviations and the deviations will be identified and classified into types in the database. The number and percent of subjects will be presented. Summaries will include type of deviation by treatment phase, treatment group (for Phase I only).

A subject data listing of protocol deviations will be provided.

4. DEMOGRAPHICS AND BASELINE CHARACTERISTICS

4.1 DEMOGRAPHICS AND BASELINE CHARACTERISTICS

Baseline characteristics (e.g., sex, age, race, ethnicity, height and weight) will be summarized with descriptive statistics or frequency counts by treatment phase, treatment group (for Phase I only) and overall for the Safety population.

4.2 MEDICAL HISTORY

Medical history will be collected prior to the treatment Phase I. Data will be coded with the Medical Dictionary for Regulatory Activities (MedDRA) Terminology. Events will be coded to System Organ Class (SOC) and preferred term (PT) using MedDRA Version 24.0 or higher. The number and percent of subjects with clinically significant medical history at screening will be summarized by system organ class (SOC) and preferred term (PT) by treatment phase, treatment group (for phase I only) and overall for safety Population.

5. STUDY DRUG AND EXPOSURE

5.1 TREATMENT COMPLIANCE AND EXTENT OF EXPOSURE

Treatment compliance will not be summarized, as each subject will be treated in an inpatient setting to receive a single dose of study drug on Day 0 and Day 7 of Phase I or II or a single dose during any time before week 46 of Phase III.

The number (percentage) of subjects who received IV injections of study drug will be summarized by treatment phase, treatment group (for Phase I only) and overall for safety population. The number (percentage) of subjects who received additional IV injection of study drug in Phase III will be summarized for Phase III safety population.

5.2 PRIOR AND CONCOMITANT THERAPY

Prior only medications are defined as medications that stopped prior to the first dose of study drug in Phase I. Concomitant only medications are defined as medications (other than the study drug) taken on or after the first dose of the study drug in Phase I during the study. Medications started before the first dose of study drug in Phase I and continuing at the time of the first dose of study drug are considered as both prior and concomitant medication.

All investigator terms for medications recorded on the CRF will be coded using the World Health Organization (WHO) Anatomical Therapeutic Chemical (ATC) drug dictionary (Version WHO Drug Dictionary Enhanced, WHO DDE March 2021 or higher). The number (percentage) of subjects who took prior medications will be summarized by treatment group in Phase I and overall for Treatment Phase I safety population. Prior general medications and iron/RLS therapies will be summarized separately. The number (percentage) of subjects who took concomitant medications will be summarized by treatment phase, treatment group (for Phase I only) and overall for safety population.

6. EFFICACY ANALYSES

All efficacy analyses will be based on the Safety Population for each phase accordingly.

Statistical testing for treatment effect:

- Phase I, II and III: no formal statistical testing will be performed. Data will be reported via descriptive statistics only.

6.1 PRIMARY EFFICACY ANALYSIS

6.1.1 Phase I and Phase II: Change in IRLSS total score from baseline to Day 42

The actual value of IRLSS total score on baseline and Day 42, and change from baseline to Day 42 will be summarized via descriptive statistics by treatment group for each treatment phase.

A subject data listing of IRLSS total score for each visit will be provided.

6.1.2 Phase I and Phase II: Percentage of subjects who are much or very much improved at Day 42 as measured by the CGI-I

The number (percentage) of subjects who are rated as much or very much improved with the CGI-I on Day 42 will be summarized by treatment group (for Phase I only) for each treatment phase. The denominator should be the Phase I Safety population for each treatment group in Phase I. For Phase II, the denominator is the Safety population in Phase II.

6.1.3 Phase III: Time from initial FCM treatment to further FCM treatment

Time (weeks) from the initial FCM treatment to further FCM treatment received in Phase III will be calculated as the integer of (date of the further FCM treatment in Phase III - date of the initial FCM treatment)/7 + 1. The time duration will be summarized via descriptive statistics including number of subjects, median, minimum (min) and maximum (max). Initial treatment is defined as Day 0 of Phase I for responders in Phase I and Day 0 of Phase II for responders in Phase II. At any time before week 46 subject could receive a single dose of unblinded FCM treatment (i.e. the further FCM treatment) if RLS symptom is worsened.

6.2 SECONDARY EFFICACY ANALYSES

6.2.1 Phase I and II: Change in IRLSS total score from baseline to Day 42 (Non-responders in Phase I Placebo group)

The change is defined as IRLSS total score at Day 42 minus IRLSS total score at baseline in each phase. The number of subjects, mean, median, standard deviation, minimum (min) and maximum (max) for the observed values, and change from baseline will be reported in summary table.

6.2.2 Phase I and II: Change in IRLSS total score from baseline to Day 42 (Non-responders in Phase I FCM group)

IRLSS total score from baseline to Day 42 between Phase I and Phase II will be summarized. The change is defined as IRLSS total score at Day 42 minus IRLSS total score at baseline in each phase. The number of subjects, mean, median, standard deviation, minimum (min) and maximum (max) for the observed values, and change from baseline will be reported in summary table.

6.2.3 Phase I and Phase II: CGI-I and PGI-I score at Day 42

The number (percentage) of subjects of each category will be summarized for CGI-I and PGI-I, separately.

6.2.4 Phase I and Phase II: Percentage of subjects who are much or very much improved at Day 42 as measured by the PGI-I

The number (percentage) of subjects who are rated as much or very much improved with the PGI-I on Day 42 will be summarized by treatment group (for Phase I only) for each treatment phase.

6.2.5 Phase I and Phase II: Change in sleep quality measured by MOS from baseline to Day 42

The actual value of MOS score on baseline and Day 42, and change from baseline to Day 42 will be summarized via descriptive statistics by treatment group (for Phase I only) for each treatment phase.

6.2.6 Phase I and Phase II: Number (percentage) of treatment non-responders at Day 42 of Phase I who respond to FCM in Phase II

The number (percentage) of treatment non-responders at Day 42 in Phase I who are turned into responders in Phase II, and the treatment responders of each phase, will be summarized by treatment group, separately. For Phase I, treatment differences in percentage will be assessed with the chi-square test, p-values will be presented. The subjects who missing responder/non-responder classification will be considered as non-responder.

6.2.7 Phase I and Phase II: Number (percentage) of treatment responders at Day 42

The number (percentage) of treatment responders at Day 42 of Phase I and Phase II will be summarized by treatment group in Phase I, separately. The subjects who missing responder/non-responder classification will be considered as non-responder.

6.2.8 Phase I and Phase II: Change in substantia nigra iron index from baseline to Day 42

The actual value of substantia nigra iron index on baseline and Day 42, and change from baseline to Day 42 will be summarized via descriptive statistics by treatment group (for Phase I only) for each treatment phase.

Substantia nigra iron index will be measured using MRI relaxation rate (R2*) or QSM.

6.2.9 Phase I and Phase II: Change in thalamus and motor cortex iron index from baseline to Day 42

The actual value of thalamus and motor cortex iron index on baseline and Day 42, and change from baseline to Day 42 will be summarized via descriptive statistics by treatment group (for Phase I only) for each treatment phase, separately.

Iron index will be measured using MRI relaxation rate (R2*) or QSM.

6.2.10 Phase III: Change in IRLSS total score from baseline to each visit

The actual value of IRLSS total score on baseline and each scheduled visit (i.e. Weeks 15, 25, 34, 43 and 52 via phone contact), and change from baseline to each scheduled visit will be summarized via descriptive statistics.

6.3 EXPLORATORY ANALYSES

6.3.1 Phases I and II: Vitamin D at each visit

The actual value of serum Vitamin D concentration on baseline, each post-baseline visit, and change from baseline to each post-baseline visit will be summarized via descriptive statistics by treatment group (for Phase I only) for each treatment phase.

A subject data listing of Vitamin D and phosphate for each visit will be provided.

6.3.2 Phases I and II: Change in thalamus and motor cortex iron index from baseline to Day 42 for other brain regions

The actual value of iron index in other brain regions (including caudate, putamen, nucleus accumbens, globus pallidus, subthalamic nuclei (STN), dentate and pulvinar) on baseline and Day 42, and change from baseline to Day 42 will be summarized via descriptive statistics by treatment group (for Phase I only) for each treatment phase, separately.

7. SAFETY ANALYSIS

All safety analyses will be performed on the Safety Population. No formal statistical testing will be performed for safety. Safety assessments include:

- Adverse events
- Laboratory assessments (hematology, iron indices, chemistry, and other)
- Vital signs

7.1 ADVERSE EVENTS

Adverse events (AEs) will be coded by System Organ Class (SOC) and Preferred Term (PT) using the Medical Dictionary for Regulatory Activities (MedDRA®, version 24.0 or higher). The verbatim term will be included in the AE listings.

A treatment-emergent adverse event (TEAE) is defined as an AE that occurs or worsen on or after the first dose of study drug in Phase I. Only TEAEs will be included in summary tables by treatment group (for Phase I only) for each phase. For AEs occurring on the first dosing day, if the start time cannot be ascertained, the event will be counted as treatment emergent. All AEs, treatment emergent or otherwise, will be presented in subject data listings.

The incidence of TEAEs will be summarized as the number (percentage) of subjects with TEAEs within SOC and PT by treatment group (for Phase I only) for each phase. Subjects who report the same PT on multiple occasions will be counted once for the PT: under the highest severity (severe > moderate > mild) when summarized by severity and under the closest relationship (probably related > possibly related > unlikely related > none) to study drug when summarized by relationship. If a subject reports multiple PT for a SOC, the subject will be counted only once for that SOC. Events with unknown severity or relationship will be counted as unknown.

TEAEs will be summarized as below.

- An overview table, including number of subjects with
    - TEAEs
    - serious AEs (SAEs)
    - study drug related TEAEs
    - study drug related SAEs
    - TEAEs by severity
    - TEAE of special interest (AESI)
    - TEAEs leading to study discontinuation
    - TEAEs leading to death
- TEAE by SOC and PT
- TEAE by SOC, PT, and Severity
- Study drug related TEAEs by SOC, PT
- SAEs by SOC and PT
- TEAEs leading to study discontinuation by SOC and PT

All TEAE tables will be sorted by SOC and PT in decreasing frequency of the number and percentage of subjects in the treatment group.

7.1.1 Adverse Events of Special Interest

Adverse events of special interest include hypophosphatemia, hypersensitivity/anaphylactoid reactions, and injection/infusdion site reactions. The search strategy will be identified as follows:

- TEAEs of hypophosphatemia by SOC and PT <Use the following MedDRA PTs to identify these TEAEs >
  - MedDRA PT Blood phosphorus decrease
  - MedDRA PT Blood phosphorus abnormal
  - MedDRA PT Hypophosphataemia
  - MedDRA PT Hypophosphataemic rickets
  - MedDRA PT Rickets familial hypophosphataemic

- TEAEs indicative of hypersensitivity/anaphylactoid reactions by SOC and PT
  - Hypersensitivity/anaphylactoid reaction search is based on the following search criteria: 2 SMQ (Anaphylactic reaction and Angioedema), and 1 PT of Hypersensitivity

- TEAEs of injection/infusion site reactions by SOC and PT <Use the following MedDRA PTs to identify these TEAEs >
  - MedDRA HLT Infusion site reactions
  - MedDRA HLT Injection site reactions
  - MedDRA HLT Administration site reactions NEC
  - MedDRA PT Infusion related reaction

The AEs of special interest will be summarized by SOP and PT.

7.1.2 Deaths, Serious and Other Significant Adverse Events

The listings of serious AEs, AE leading to study discontinuation, and subjects who died during the study will be listed.

7.2 CLINICAL LABORATORY PARAMETERS

Laboratory assessments include hematology, clinical chemistry/ phosphorus, and iron indices:

- Hematology: Hgb, Hct, RBC, WBC, MCV, MCH, MCHC, RDW, platelets, differential count and reticulocyte count.

- Clinical chemistry: sodium, potassium, chloride, BUN, creatinine, albumin, alkaline phosphatase, total bilirubin, gamma-GGT, AST, ALT, LDH, calcium, phosphorus, glucose, bicarbonate and magnesium.

- Iron indices and Phosphorus: serum iron, serum ferritin, and total iron binding capacity (TIBC), percentage serum transferrin saturation (TSAT).

- Other: urine pregnancy (if indicated), other metals (serum copper and zinc), 1,25 dihydroxy vitamin D, 25 hydroxy Vitamin D (25OH-D) and erythropoietin.

All laboratory parameters will be presented in standard units. The actual value and change from baseline will be summarized by visit and treatment group (for Phase I only) using descriptive statistics.

The number and percent of subjects with treatment-emergent potentially clinically significant (PCS) laboratory values after baseline will be summarized by visit and treatment group (for Phase I only) for each phase. The denominator is all subjects with normal baseline and at least one post baseline assessment in the safety population of that phase and the numerator is the number of subjects with PCS (i.e., meets Grade III or Grade IV toxicity criteria from the National Cancer Institute Common Terminology Criteria) at post-baseline.

7.3 VITAL SIGNS AND PHYSICAL EXAMINATION FINDINGS

7.3.1 Vital Signs

Vital signs will be collected for Phase I and Phase II, include sitting body temperature, blood pressure (BP) and heart rate. On study drug dosing days BP and heart rate will be collected immediately pre-dosing, immediately and 30 minutes post dosing, and body temperature will be collected on the dosing dates.

All vital signs will be presented in conventional units as in Table 3. Frequency and percentage of subjects with values considered PCS occurring at post-baseline in dosing days will be summarized by visit and by treatment group (for Phase I only) for each Phase. The denominator is all subjects with a baseline assessment in the full analysis population and the numerator is the number of subjects with PCS at post-baseline. Criteria for PCS are presented below Table 2.

Table 2 Criteria for Potentially Clinically Significant Vital Signs

Vital Sign	Criterion	Definition of PCS at Any Post-baseline Time Point
Systolic blood pressure	Low	Value ≤90 mmHg and decreased ≥20 mmHg [1]
	High	Value ≥180 mmHg and increased ≥20 mmHg [1]
Diastolic blood pressure	Low	Value ≤50 mmHg and decreased ≥15 mmHg [1]
	High	Value ≥105 mmHg and increased ≥15 mmHg [1]
Pulse	Low	Value ≤50 bpm and decreased ≥15 bpm [1]
	High	Value ≥ 120 bpm and increased ≥15 bpm [1]

[1] For immediately and 30 minutes post-dose assessments on Dosing Days, the change (decrease or increase) is based on the pre-dose value obtained on each corresponding dosing day.

7.3.2 Physical Examination

Any significant findings from physical examinations will be documented in medical history or adverse events.

7.4 OTHER ANALYSES

Cambridge-Hopkins Diagnostic Questionnaire (CHDQ), Hopkins-Henning Telephone Diagnostic Interview (HTDI), Hopkins RLS-Sleep Quality Questionnaire (HRSQ), and urine

8. INTERIM ANALYSES AND DATA AND SAFETY MONITORING BOARD (DSMB)

8.1 INTERIM ANALYSES

No formal interim analysis is planned for this study.

8.2 DATA AND SAFETY MONITORING BOARD (DSMB)

No Data Safety Monitoring Board for this study.

9. SUMMARY OF MAJOR CHANGES IN THE PLANNED ANALYSES

Compared with the study protocol 1VIT15042 (Date: 4 September 2019), below items are the major changes made in this SAP:

1. Due to early termination of the study, no statistical analysis will be performed. Hence no primary, and secondary end points will not be tested with statistical model.

2. Physical examinations was removed from safety analysis list and any significant findings will go to medical history or adverse event summaries.

3. The secondary endpoint “Number of Subjects who remained in the study to Week 52” will be summarized in disposition table for n (%) of study completer. It will not be considered as an efficacy endpoint in this SAP.

4. Adverse events of special interest were added in this SAP in Section 7.1.1.