PAREXEL International

Akebia Therapeutics, Inc.

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PHASE 2, RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED, DOSE-FINDING STUDY TO ASSESS THE EFFICACY, SAFETY, PHARMACOKINETICS, AND PHARMACODYNAMICS OF VADADUSTAT IN JAPANESE SUBJECTS WITH ANEMIA SECONDARY TO NON-DIALYSIS DEPENDENT CHRONIC KIDNEY DISEASE (NDD-CKD)

Statistical Analysis Plan

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LIST OF ABBREVIATIONS

AE	adverse event
ALT	alanine aminotransferase
AST	aspartate aminotransferase
BMI	body mass index
CBC	complete blood count
CI	confidence interval
ECG	Electrocardiogram
eCRF	electronic Case Report Form
eGFR	estimated glomerular filtration rate
EOT	end-of-treatment
ESA	erythropoiesis-stimulating agent
Hb	Hemoglobin
ITT	intent-to-treat
IV	Intravenous
LOCF	last observation carried forward
LS mean	least-squares mean
MedDRA	Medical Dictionary for Regulatory Activities
MITT	modified intent-to-treat
NDD-CKD	non-dialysis dependent chronic kidney disease
PD	pharmacodynamic(s)
PK	pharmacokinetic(s)
PP	per protocol
PT	preferred term
Q1	1st quartile
Q3	3rd quartile
RBC	red blood cell
SAE	serious adverse event
SAP	statistical analysis plan
SMQ	standardised MedDRA queries
SOC	system organ class
TEAE	treatment-emergent adverse event
TIBC	total iron binding capacity
TSAT	transferrin saturation
VEGF	vascular endothelial growth factor

1 INTRODUCTION

This study is a Phase 2 clinical study to assess the dose-response relationship between oral vadadustat once daily dosing for 6 weeks and the change in hemoglobin (Hb) in Japanese subjects with anemia secondary to non-dialysis dependent chronic kidney disease (NDD-CKD); in order to define the starting dose for use in Phase 3 clinical studies in Japan.

This Statistical Analysis Plan (SAP) defines the preliminary and final statistical analyses for the clinical study report for this study. This should be read in conjunction with the study protocol that provides all necessary background information and rationale for the current study and its design.

The pharmacokinetic data will be separately obtained from vendor to keep blindness of the study.

This SAP is based upon the following study documents:

- Study Protocol, Version 4 (December 15, 2016)
- electronic Case Report Form (eCRF), Version 3.0 (October 6, 2016)
- Protocol Deviation Specification Form, Version 1.0 (September 29, 2016)

2 STUDY OBJECTIVES

2.1 Primary Objective and Endpoint

The primary objective of this study is to assess the dose-response relationship between oral vadadustat once daily dosing for 6 weeks and the change in Hb in Japanese subjects with anemia secondary to NDD-CKD; this is to define the starting dose for use in Phase 3 clinical studies in Japan.
The primary endpoint that will be used to assess this objective is the mean change in Hb levels from pre-treatment to the end of the primary efficacy period (Week 6). Pre-treatment Hb is defined as the average of 2 Hb values obtained prior to treatment based on the qualifying screening Hb value and the Hb value at the baseline visit.

2.2 Secondary Objectives and Endpoints

The secondary objectives of this study are:

- To assess the safety, tolerability, pharmacokinetics (PK), and pharmacodynamics (PD) of oral vadadustat once daily dosing in Japanese subjects with anemia secondary to NDD-CKD during the 6-week, primary efficacy period
- To evaluate the effect of dose adjustments and demonstrate the maintenance effect on Hb during a 10-week dose-adjustment and maintenance period
- To assess the time to reach the target Hb range from baseline

The efficacy endpoints that will be used to assess these objectives include the following:

- Time to reach target Hb level of 10.0-12.0 g/dL from baseline visit (Day 1)
- Mean Hb levels at the end of the primary efficacy period (Week 6) and at the end of the dose adjustment and maintenance period (Week 16)
- Proportion of subjects who achieve target Hb 10.0-12.0 g/dL at the end of the dose adjustment and maintenance period (Week 16)
- Mean change in Hb between pre-treatment and the end of the dose adjustment and maintenance period (Week 16)
- Mean change in hematocrit, red blood cell (RBC) count, and reticulocyte count from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Mean change in iron indices (ie, iron, total iron-binding capacity [TIBC], transferrin saturation [TSAT], and ferritin) and hepcidin from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Proportion of subjects requiring rescue with RBC transfusion from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Proportion of subjects requiring rescue with ESAs from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Number of dose adjustments from baseline to the end of the dose adjustment and maintenance period (Week 16)
- Maintenance of iron sufficiency (defined as ferritin ≥50 ng/mL and TSAT ≥20%) from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Plasma concentration profile of vadadustat and its metabolites using pre-dose sample from Week 4

The safety endpoints that will be used to assess these objectives include the following:

- Safety assessments, including adverse events (AEs), vital signs, electrocardiograms (ECGs), and other laboratory assay results (eg, chemistry, components of the complete blood count [CBC] other than the ones noted above, and vascular endothelial growth factor [VEGF])

3 INVESTIGATIONAL PLAN

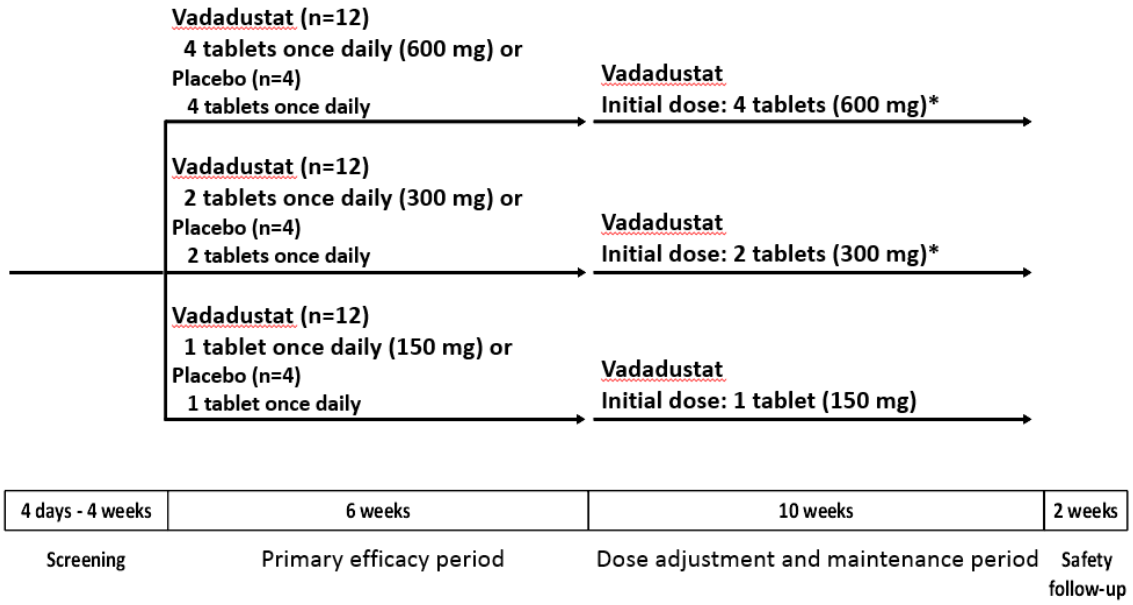
3.1 Overall Study Design and Plan

This is a Phase 2, randomized, double-blind, placebo-controlled, dose-finding study to assess the efficacy, safety, tolerability, PK, and PD of orally administered vadadustat in Japanese subjects with anemia secondary to NDD-CKD.

The study has a planned enrollment of 48 subjects to be enrolled at approximately 25 sites in Japan. There will be 16 subjects in each of the 3 tablet-count groups.

An overview of the study design is presented in Figure 1.

Figure 1: Overview of Study Design



* For subjects who develop an excess Hb response during the primary efficacy period, the number of tablets of study drug will be decreased (see Section 8.2.4). For these subjects, the number of tablets of vadadustat initiated at the Week 6 visit will be lower than indicated.

The study will include the following periods:

- Eligibility screening period (up to 4 weeks)
- Primary efficacy period (6 weeks; Weeks 1 to 6)
- Dose adjustment and maintenance period (10 weeks; Weeks 7 to 16)
- Follow-up period (2 weeks; Weeks 17 and 18)

Subjects will participate in a screening period (4 days to 4 weeks) to determine study eligibility, and eligible subjects will be randomized following the screening period.

Using a central randomization system, subjects will be randomized 1:1:1 to receive 1, 2, or 4 tablets at their baseline visit (Day 1). Within each tablet-count group, subjects will be randomized 3:1 to receive vadadustat (150, 300, or 600 mg vadadustat) or placebo. See the protocol section 8.2.2 for information regarding the randomization scheme.

Study drug treatment will be administered during a 6-week primary efficacy period. See the protocol section 8.2.4 for information on study drug administration. The primary efficacy period includes fixed-dose treatment to establish a dose-response relationship. However, if Hb level increase rapidly or if the Hb level exceeds 13.0 g/dL, the study drug dose can be decreased or interrupted (see the protocol section 8.2.4).

After completing the primary efficacy period, subjects will continue to a 10-week dose adjustment and maintenance period (see the protocol section 8.2.5). Subjects receiving placebo will be switched to vadadustat, and study drug dose will be adjusted to achieve a target Hb of 10.0-12.0 g/dL based on dose adjustment guidelines (see the protocol section 8.2.5).

Vadadustat treatment will stop after the dose adjustment and maintenance period has been completed (Week 16) and subjects will continue in a 2-week follow-up period (Week 17-18).
The clinical and safety assessments will be performed as described in the protocol section 9.3 and as listed in the protocol Appendix A.

3.2 Efficacy and Safety Variables

3.2.1 Efficacy and PD Variables

The following variable is the primary efficacy variable:

- Change from pre-treatment in Hb at Week 6.

The following efficacy endpoints will also be analyzed:

- Actual values and change (absolute and percent) from pre-treatment in Hb, hematocrit, RBC count, and reticulocyte count (both absolute and percent)
- Actual values and change (absolute and percent) from pre-treatment in iron, TIBC, TSAT, ferritin, and hepcidin

3.2.2 Safety Variables

The following variables are the safety endpoints:

- AEs
- Vital signs
- ECGs
- Laboratory parameters

3.2.3 PK Variables

The following variables are the PK variable:

- Pre-dose plasma concentrations of vadadustat at the Week 4 visit
- Pre-dose plasma concentrations of vadadustat's metabolites at the Week 4 visit

4 STATISTICAL METHODS

4.1 Data Quality Assurance

All tables, figures and data listings to be included in the report will be independently checked for consistency, integrity and in accordance with standard PAREXEL procedures.

4.2 General Presentation Considerations

'Pre-treatment' of Hb is defined as the average of 2 Hb values obtained prior to treatment based on the qualifying screening Hb value and the Hb value at the Baseline Visit. 'Pre-

treatment' for other variables is defined as the last available pre-treatment assessment. In general, this is the value at the Baseline Visit. 'End of Treatment (EOT)' is defined as the assessment on the EOT form labeled as 'Week 16 Visit', even if other values closer to Day 113 are available. 'Treatment Day' will be calculated relative to start of dosing at Baseline Visit (i.e. Treatment Day = Assessment Date - Baseline Visit Date + 1).

Continuous data will be summarized in terms of the mean, standard deviation (SD), median, minimum, maximum and number of observations, unless otherwise stated. The minimum and maximum will be reported to the same number of decimal places as the raw data recorded in the database. The mean and median will be reported to one more decimal place than the raw data recorded in the database. The SD will be reported to two more decimal places than the raw data recorded in the database. In general, the maximum number of decimal places reported shall be four for any summary statistic.

Categorical data will be summarized in terms of the number of subjects providing data at the relevant time point (n), frequency counts and percentages. Any planned collapsing of categories will be detailed in the SAP text and the data displays.

Percentages will be presented to one decimal place. Percentages will not be presented for zero counts. Percentages will be calculated using n as the denominator. If sample sizes are small, the data displays will show the percentages, but any textual report will describe frequencies only.

Changes from baseline in categorical data will be summarized using shift tables where appropriate.

P-values greater than or equal to 0.001, in general, will be presented to three decimal places. P-values less than 0.001 will be presented as "<0.001".

Confidence intervals will be presented to one more decimal place than the raw data.

All report outputs will be produced using SAS® version 9.3 or a later version in a secure and validated environment. All report outputs will be provided to the Sponsor in a single Microsoft Word 2010 [Version 14.0] document.

4.3 Study Subjects

4.3.1 Disposition of Subjects

A clear accounting of the disposition of all subjects who enter the study will be provided, from screening to study completion.

The summary of the following subject disposition will be presented.

- A summary of the number of subjects randomized, the number and percentage of subjects treated (with at least one dose of study medication) and the number and percentage of subjects completed through Week 6 Visit and discontinued by Week 6 Visit by treatment group and overall (Analysis population: All Subjects

Randomized). For final analysis, a summary of the number and percentage of subjects completed Week 18 Visit and discontinued from the study by treatment group and overall will be provided (Analysis population: All Subjects Randomized). Withdrawals from the study will also be summarized by major reason.

By-subject listings of eligibility details, randomization details, visit dates and withdrawal/study completion details (including reason for discontinuation and duration of treatment prior to discontinuation) will be provided.

4.3.2 Protocol Deviations

Major protocol deviations are defined as those deviations from the protocol likely to have an impact on the perceived efficacy and/or safety of study treatments. The impact of major protocol deviations on the efficacy and/or safety results will be investigated by assessing the robustness of the study results and conclusions to the choice of analysis population (see Section 4.4), both including and excluding data potentially affected by major protocol deviations.

Protocol deviations and any action to be taken regarding the exclusion of subjects or affected data from specific analyses will be determined in Protocol Deviation Specification document.

The protocol deviation will be summarized as following:

- A summary of the number and percentage of subjects with a protocol deviation by treatment group and overall and by type of deviation (Analysis population: All Subjects Randomized)

A by-subject listing of protocol deviations will be provided.

4.4 Analysis Populations

Upon database release, protocol deviation and analysis population outputs will be produced and will be sent to Sponsor for review. An analysis population classification meeting will be arranged to discuss the outputs and to decide which subjects and/or subject data will be excluded from certain analyses. Decisions made regarding the exclusion of subjects and/or subject data from analyses will be made prior to unblinding and will be documented and approved by Sponsor.

The following summary of analysis population will be provided:

- A summary of the number and percentage of subjects allocated to each analysis population by treatment group and overall (Analysis population: All Subjects Randomized)

A by-subject listing of analysis population details will be provided. This listing will be presented by treatment group and include: site, subject identifier, inclusion/exclusion flag for each population and reason for exclusion from each population. All subjects randomized will appear on this listing.

4.4.1 Analysis Population for the Safety Analyses

The safety summaries and analyses will be based on the **safety population**. The safety population is defined as all enrolled subjects who receive at least 1 dose of study medication. In the event that a subject is allocated the incorrect study treatment as per the study randomization list, subjects will be summarized and analyzed 'as treated' i.e. by allocated treatment group.

4.4.2 Analysis Populations for the Efficacy Analyses

The efficacy summaries and analyses will be based on the **modified intent-to-treat (MITT) population**. The MITT population is defined as all randomized subjects who have received at least 1 dose of study medication and have a pre-treatment Hb average defined as the average of the qualifying screening value and the baseline value, and at least one post-baseline Hb measurement. In the event that a subject is allocated the incorrect study treatment as per the study randomization list, subjects will be summarized and analyzed 'as randomized' i.e. by randomized treatment group.

For the efficacy variables, sensitivity analyses will be performed on the **per-protocol (PP) population** to assess the robustness of the study conclusions to the choice of analysis population. The PP population is defined as the subjects in the MITT population who have completed the study and have efficacy data through Week 6, have a study medication compliance of ≥80%, and do not have any major protocol deviations expected to significantly affect the primary efficacy endpoint. Subjects with major protocol deviations expected to significantly affect the primary efficacy endpoint will be identified and documented prior to data unblinding.

4.4.3 Analysis Population for the PK Analysis

The PK summaries and analyses will be based on the **pharmacokinetics (PK) population**. The PK population is defined as all subjects in the safety population who have a pre-dose PK sample at Week 4. Subjects will be summarized and analyzed by actual treatment group.

4.5 Demographic and Other Baseline Characteristics

The demographic and other baseline characteristics will be summarized for the MITT and safety populations. The following summaries will be provided:

- A summary of demographic variables (e.g. age, gender, weight, height and body mass index [BMI]) by treatment group and all subjects

- A summary of other relevant variables (e.g. estimated Glomerular Filtration Rate [eGFR] at baseline, CKD stage, etiology of CKD and smoking status) by treatment group and all subjects
- A summary of relevant medical history by treatment group and all subjects.
- A summary of prior and concomitant medications within 30 days prior to the first study drug dose up to and including Week 6 by treatment group and overall.
- For the final analysis, a summary of concomitant medications after Week 6 will be provided.

Age will be calculated as the number of complete years between a subject's birth date and the date of informed consent.

Definitions of eGFR and CKD stage are described in section 4.8.4.7.

CKD stage will be determined by eGFR at baseline:

CKD stage	eGFR (mL/min/1.73m^2)
1	90+
2	60 - 89
3a	45 - 59
3b	30 - 44
4	15 - 29
5	< 15

By-subject listings of demographic data and other baseline characteristics (as summarized above) will be provided

4.6 Treatment Compliance

The treatment compliance will be summarized for the MITT population. The following summary will be provided.

- A summary of the treatment compliance measures (i.e. ≥80% compliance and <80%) by treatment group and study visit.

A by-subject listing of treatment compliance data will be provided.

4.7 Efficacy Evaluation

4.7.1 Analysis and Data Conventions

This study is designed to test for dose-response relationship. An analysis of covariance (ANCOVA) model will be used to compare change from pre-treatment in Hb between the three vadadustat dosing groups and the placebo group. The model will include treatment assignment (3 dosed groups and one placebo group) and pre-treatment Hb value as a covariate. A step-down procedure will be used to control the overall type I error rate for the multiple comparisons. Testing of the highest dose compared with placebo will be

conducted first. If and only if this comparison is significant, then testing will proceed to comparison of the next lower dose and placebo, and so on. Therefore, no multiplicity adjustment will be needed for this analysis.

A linear regression dose-response modeling will be performed as a secondary analysis.

$$Y = \beta_0 + \beta_{dose}X$$

where Y is change in Hb and X is vadadustat dose. β_xs are parameters of the model. β_0 is intercept and β_{dose} is slope, respectively.
The null hypothesis for the dose response will be that the regression coefficient of slope parameter is zero. The alternative hypothesis will be that the regression coefficient is not zero. Symbolically, this is expressed as follows:

$$H_0: \beta_{dose} = 0$$
$$H_1: \beta_{dose} \neq 0$$

This hypothesis will be tested using a 0.05 two-sided significance level.

4.7.1.1 Multi-center Studies
Not Applicable. Analysis of center effect is not planned.

4.7.1.2 Adjustments for Covariates
The primary efficacy analysis will be adjusted for the pre-treatment measure of primary efficacy variable.

An 'unadjusted' sensitivity analysis will be performed to assess the robustness of the study conclusion.

4.7.1.3 Handling of Missing Data
For the MITT population analyses for the primary efficacy variable, the Last Observation Carried Forward (LOCF) approach will be used to impute missing data.

For the secondary endpoint analyses, data will be used as observed and no imputation method will be used.

4.7.1.4 Multiple Comparisons/Multiplicity
Analysis of one primary variable [Change in Hb] and one time point of primary interest [Week 6 visit] has been defined for this study. Though 3 pairwise comparison between placebo and dosed group is planned, step-down procedure will be used to control the overall type I error rate. The secondary variables defined are intended to provide supportive evidence relating to the primary objective and no labeling claims are intended. Though one preliminary analysis is planned before the final analysis, primary analysis will not be changed. Hence no adjustments for multiplicity are required.

4.7.1.5 Preliminary (6-Week) Analysis

After all subject performed assessment of the Week 6 Visit, preliminary analysis will be performed. As the study conduct and final analysis will not be modified by this analysis, no alpha adjustment is proposed. It is expected that this 6-week data
will be identical for the preliminary analysis and the final analysis of the complete dataset. Any changes between the preliminary analysis and the final analysis will be documented. The preliminary analysis will be performed by the independent unblinded team. All data through the cutoff date of the last subject randomized who completes week 6 will be analyzed. Blinding of blinded team and investigators will be maintained to prevent potential bias.

4.7.1.6 Examination of Subgroups

Not Applicable. No subgroup analysis is planned.

4.7.2 Primary Efficacy Variable – Mean Change in Hb from Pre-treatment to the End of the Primary Efficacy Period (Week 6)

The primary variable for the assessment of efficacy is the change from pre-treatment to Week 6 in Hb (g/dL). Change in Hb is defined as the Hb measured at the Week 6 visit minus the mean pre-treatment Hb. Pre-treatment Hb is defined as the average of 2 Hb values obtained prior to treatment based on the qualifying screening Hb value and the Hb value at the baseline visit.

For the primary analysis, the LOCF approach will be used to impute missing data. Complete-case analysis will be performed for sensitivity at the scheduled Week 6 visit.

4.7.2.1 Summary of actual value and change in Hb at week 6

The primary analysis population for efficacy will be MITT population. The following summary will be provided:

- A summary of actual value of Hb by treatment group at baseline and week 6. A summary of the change (absolute and percent) from baseline in Hb by treatment group at week 6. 1st and 3rd quartiles (Q1, Q3) as well as the mean, SD, median, minimum, maximum and number of observations will be shown. P values for overall comparison and treated groups vs. placebo on change from baseline will also be presented.
- A waterfall plot for absolute Hb change from baseline for each treatment group at week 6.

The effect of treatment in terms of the change from pre-treatment to Week 6 in Hb will be analyzed using a one-way analysis of covariance (ANCOVA). The statistical model will assess the main effect of treatment, adjusted for pre-treatment average. Least-squares (LS) means for each treatment and their 95% confidence intervals (CIs) will be presented. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done. Unadjusted one-way analysis of variance (ANOVA) will be performed for sensitivity.

The simple linear regression between the change from pre-treatment to Week 6 in Hb and dose will be calculated. The regression coefficients and their standard errors, p-values of test of regression parameters and their 95%CIs will be presented. The test of slope parameter will be used to evaluate dose response relationship.

As sensitivity analyses, efficacy endpoints will also be analyzed using the PP population.

A by-subject listing of actual values and change (absolute and percent) from baseline in Hb will be provided.

4.7.2.2 Summary of actual value and change in Hb from baseline to week 6 and beyond

The analysis population for efficacy will be MITT and PP population. The following summary will be provided:

- A summary of actual value of Hb by treatment group and study visits. 1st and 3rd quartiles (Q1, Q3) as well as the mean, SD, median, minimum, maximum and number of observations will be shown. Summary of pre-treatment average will also be presented.
- A summary of the change (absolute and percent) from baseline in Hb by treatment group and study visits.
- A plot showing the mean Hb value over time within each treatment group. 95% CI of these means will be shown as error bar and sample size in each group at each time point will be shown.
- Plots of individual profile of Hb values over time within each treatment group.
- Box plots of absolute Hb changes from baseline for each treatment group at each post-treatment study visit

A by-subject listing of actual values and change (absolute and percent) from baseline in Hb will be provided.

4.7.3 Secondary Efficacy Variables

Time to reach target Hb level

Time to reach target Hb level of 10.0-12.0 g/dL from baseline is defined for subjects with Hb <10.0 g/dL at the baseline visit as following:

First post-treatment assessment date with value of Hb ≥10.0 and ≤12.0 g/dL – the baseline visit date +1. If target Hb level is not achieved, last Hb assessment date will be used as censored date.

A summary of time to reach target Hb level will be provided for subjects with Hb <10.0 g/dL at the baseline visit by treatment group.

Kaplan-Meier failure (cumulative incidence) plot will be presented for the time to reach target Hb level by treatment group.

A by-subject listing will be provided.

Hb within target level

A summary of the number and percentage of subjects within target Hb 10-12 g/dL at the end of the dose adjustment and maintenance period (Week 16) will be provided by treatment group.

The number and percentage of subjects above / within / below target range who are maintained within the target range at their previous visit after Week 6 will be provided by treatment group.

The effect of treatment in terms of the proportion of subjects within target Hb level at Week 6 will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

A by-subject listing will be provided.

Increase in Hb from pre-treatment average

A summary of the number and percentage of subjects who achieved a ≥1 g/dL increase in Hb from pre-treatment average will be provided by treatment group.

The effect of treatment in terms of the proportion of subjects who achieved a ≥1 g/dL increase in Hb from pre-treatment average will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

Hematocrit, RBC count, and reticulocyte count

Hematocrit, RBC count, and reticulocyte count (both absolute and percent) will be analyzed as secondary efficacy variables.

The following summaries will be provided:

- A summary of hematocrit, RBC count, and reticulocyte count (both absolute and percent) by treatment group and time point. 1st and 3rd quartiles (Q1, Q3) as well as the mean, SD, median, minimum, maximum and number of observations will be presented.
- A summary of the change (absolute and percent) from baseline in hematocrit, RBC count, and reticulocyte count (both absolute and percent) by treatment group and time point
- Box plots of absolute changes from baseline in hematocrit, RBC count, and reticulocyte count (both absolute and percent) for each treatment group at each post-treatment time point

The effect of treatment in terms of the change from baseline to Week 6 in hematocrit and RBC count will be analyzed using ANCOVA. The statistical model will assess the main effect of treatment, adjusted for baseline measurement. LS-means for each treatment and their 95% CIs will be presented. Three post-hoc pairwise comparison, i.e., Placebo vs

150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done. Unadjusted ANOVA will be performed for sensitivity.

A by-subject listing of hematocrit, RBC count, and reticulocyte count (both absolute and percent) will be provided by treatment group.

Iron indices and hepcidin

Iron indices (ie, iron, TIBC, TSAT, and ferritin) and hepcidin will be analyzed as secondary efficacy variables.

The following summaries will be provided:

- A summary of iron indices and hepcidin by treatment group and time point
- A summary of the change (absolute and percent) from baseline in iron indices and hepcidin by treatment group and time point
- Box plots of absolute changes from baseline in iron indices and hepcidin for each treatment group at each post-treatment time point

A by-subject listing of iron indices and hepcidin will be provided by treatment group.

Rescue therapy

Initiation of rescue therapy will be analyzed.

The following summaries will be provided:

- A summary of the number and percentage of subjects requiring rescue with RBC transfusion from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- A summary of the number and percentage of subjects requiring rescue with ESAs from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)

The effect of treatment in terms of the proportion of subjects requiring rescue with RBC transfusion / ESAs will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

By-subject listings of rescue therapies will be provided by treatment group.

Dose adjustment

A summary of the number of dose adjustments from baseline to the end of the dose adjustment and maintenance period (Week 16) will be provided.

A by-subject listing of dose adjustment will be provided by treatment group.

Maintenance of iron sufficiency
A summary of maintenance of iron sufficiency (defined as ferritin ≥50 ng/mL and TSAT ≥20%) from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16) will be provided.

The effect of treatment in terms of the proportion of subjects who maintain iron sufficiency will be analyzed using Fisher's exact test for week 6 only. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

A by-subject listing of iron sufficiency will be provided by treatment group.

4.7.4 Exploratory Efficacy Variable

Time in Therapeutic Range (TTR) for Hb from week 6 to week 20
The half-time interpolation method will be used to calculate TTR for Hb. The total time of follow-up with Hb in target range of 10.0 to 12.0 g/dL is divided by the total time. Half the time between two tests is allocated to the first Hb value, and half to the second Hb value.

A summary of TTR for Hb will be provided by treatment group.

A by-subject listing of TTR for Hb will be provided by treatment group

4.8 Safety Evaluation

All safety summaries and analyses will be based upon the safety population as defined in Section 4.4.

4.8.1 Extent of Exposure

The treatment duration will be summarized for the safety population. The following summary will be provided.

- A summary of the treatment duration (e.g. End Date of Administration - Start date of Administration +1) by treatment group and study visits.

A by-subject listing of start, end and duration of treatment will be provided.

4.8.2 Adverse Events

AEs will be coded using the Medical Dictionary for Regulatory Activities (MedDRA) Version 19.0.

Treatment-emergent adverse events (TEAEs) will be tabulated and are defined as those AEs that either start or worsen in severity on or after the date/time of first dose of study

treatment and on or before the final protocol required visit. In general, it is Week 18 Visit and 14 days after the date of last dose of study treatment.

Where dates are missing or partially missing, AEs will be assumed to be treatment-emergent, unless there is clear evidence (through comparison of partial dates) to suggest that the AE started prior to the first dose of study treatment or more than 14 days after the last dose of study treatment.

The AE will be summarized using MedDRA system organ class (SOC) and preferred term (PT). All AE summaries will provide the number of subjects reporting at least one AE and the total number of events reported. The following summaries will be provided:

- A summary of the number and percentage of subjects reporting at least one TEAE, drug-related TEAE, TEAE leading to withdrawal, serious adverse event (SAE), drug-related SAE, SAE leading to withdrawal and death by treatment group.
- A summary of the number and percentage of subjects reporting a TEAE by treatment group, SOC, and PT
- A summary of the number and percentage of subjects reporting a drug-related TEAE by treatment group, SOC, and PT
- A summary of the number and percentage of subjects reporting a treatment-emergent SAE by treatment group, SOC and PT
- A summary of the number and percentage of subjects reporting a drug-related treatment-emergent SAE by treatment group, SOC, and PT
- A summary of the number and percentage of subjects reporting a TEAE leading to withdrawal by treatment group, SOC, and PT
- A summary of the most common TEAEs by treatment group and PT (reported by >10% of subjects in any treatment group).
- A summary of the number and percentage of subjects reporting a TEAE by treatment group, severity, SOC and PT
- A summary of the number and percentage of subjects reporting a TEAE by treatment group, causality, SOC and PT

AE summaries will be ordered alphabetically for SOC, and PT within SOC.

For each subject and each AE, the worst severity recorded will be attributed and used in the by-severity summaries. Similarly, the worst causality (most related to treatment) will be attributed and used in the by-causality summaries. If severity or causality is missing, data will be treated as observed

A by-subject listing of all AEs (including non-treatment-emergent events) will be provided. This listing will be presented by treatment group and will include: center, subject identifier, age, sex, race, AE (SOC, PT, and verbatim term), date of onset, date of resolution, duration, severity, seriousness, action taken, outcome and causality.

The following AEs are defined as AEs of special interest. Summaries of the number and percentage of subjects reporting the following AEs will be provided.

- PT: "hyperkalemia" (MedDRA 10020646) or "blood potassium increased" (MedDRA 10005725)
- PT: "hyperuricaemia" (MedDRA 10020903) or "blood uric acid level increased" (MedDRA 10022891)
- PT: "Hypertension" (MedDRA 10020772) or "Blood pressure increased" (MedDRA 10005750) or "Blood pressure systolic increased" (MedDRA 10005760) or "Blood pressure diastolic increased" (MedDRA 10005739) or "Blood pressure ambulatory increased" (MedDRA 10005732) or "Blood pressure inadequately controlled" (MedDRA 10051128) or "Accelerated hypertension" (MedDRA 10000358) or "Essential hypertension" (MedDRA 10015488) or "Diastolic hypertension" (MedDRA 10012758) or "Systolic hypertension" (MedDRA 10042957) or "Malignant hypertension" (MedDRA 10025600)
- Standardised MedDRA Query (SMQ): "Cholestasis and jaundice of hepatic origin" (SMQ 20000009) or "Drug related hepatic disorders – severe events only" (SMQ 20000007) or "Liver related investigations, signs and symptoms" (SMQ 20000008)
- Standardised MedDRA Query (SMQ): "Ischaemic heart disease" (SMQ 20000043)
- Standardised MedDRA Query (SMQ): "Central nervous system haemorrhages and cerebrovascular conditions" (SMQ 20000061)
- Standardised MedDRA Query (SMQ): "Embolic and thrombotic events" (SMQ 20000081)
- Standardised MedDRA Query (SMQ): "Cardiac failure" (SMQ 20000004)
- Standardised MedDRA Query (SMQ): "Retinal disorders" (SMQ 20000158, narrow)

4.8.3 Deaths, Serious Adverse Events, and Other Significant Adverse Events

Other significant AEs are those AEs reported as leading to withdrawal from study. The summaries of SAEs and AEs leading to withdrawal from study are described in section 4.8.2.

A summary of the number and percentage of deaths during the study will be provided by treatment group

A by-subject listing of all deaths that occurred during the study will be provided.

4.8.4 Clinical Laboratory Evaluation

4.8.4.1 Serum and urine pregnancy tests

Serum pregnancy test for females of childbearing potential will be performed at the screening visit and will be used for subject's inclusion / exclusion. Urine pregnancy test will be performed at the baseline visit and used for subject's initiation of study drug. Additional pregnancy tests may be conducted during the study to establish the absence of pregnancy based on the investigator's clinical judgment or as required by local regulations.

A summary of subjects with any positive pregnancy test results will be provided by treatment group.

A by-subject listing of serum and urine pregnancy tests will be provided.

4.8.4.2 Coagulation tests

Coagulation tests will be performed at the baseline visit. The following variables will be analyzed.

- prothrombin time
- partial thromboplastin time
- international normalized ratio

A summary of baseline coagulation test results will be provided by treatment group.

A by-subject listing of coagulation tests will be provided.

4.8.4.3 Folate and vitamin B12

Folate and vitamin B12 tests will be performed at the screening visit and will be used for subject's inclusion / exclusion.

A summary of folate and vitamin B12 will be provided by treatment group.

A by-subject listing of folate and vitamin B12 tests will be provided.

4.8.4.4 Urine albumin-to-creatinine ratio (uACR)

Urine albumin-to-creatinine ratio (uACR) will be evaluated at baseline visit.

A summary of uACR will be provided by treatment group.

A by-subject listing of uACR will be provided.

4.8.4.5 Complete Blood Count (CBC)

The following components of the CBC are described in Efficacy / PD analysis section.

- Hb
- Hematocrit
- RBC count
- Automated Reticulocyte Count (both absolute and percent).

The following components of the CBC will be analyzed:

- Mean Corpuscular Volume
- Mean Corpuscular Hb
- Mean Corpuscular Hb Concentration
- Red Cell Distribution Width
- White Blood Cell count with differential (neutrophils, bands, lymphocytes, monocytes, eosinophils, and basophils)
- Platelets

The following summaries will be provided:

- A summary of each component of the CBC by treatment group and time point
- A summary of the change from baseline in each component of the CBC by treatment group and time point
- A summary of the number and percentage of subjects experiencing abnormal values by component of CBC, treatment group and time point.

A by-subject listing of CBC will be provided by treatment group.

4.8.4.6 Hemoglobin

The following Hb analyses will be performed for safety analysis section:

- A summary of the number and percentage of subjects with any Hb > 12.0 g/dL by treatment group
- A summary of the number and percentage of subjects with any Hb > 13.0 g/dL by treatment group
- A summary of the number and percentage of subjects with any Hb > 14.0 g/dL by treatment group
- A summary of the number and percentage of subjects with Hb increase > 1.0 g/dL in any 2-week interval by treatment group

4.8.4.7 Chemistry and estimated glomerular filtration rate (eGFR)

The following laboratory parameters will be analyzed:

- Sodium
- Potassium
- Bicarbonate
- Chloride
- Calcium
- Phosphorus
- Glucose
- Creatinine
- Blood Urea Nitrogen
- Creatine Phosphokinase
- Uric Acid
- Albumin
- Total Protein
- Total Bilirubin
- Alkaline Phosphatase
- Alanine Aminotransferase (ALT)
- Aspartate Aminotransferase (AST)
- Lactate Dehydrogenase
- Total cholesterol
- Estimated Glomerular Filtration Rate (eGFR) (calculated from serum creatinine)

The following summaries will be provided:

- A summary of Chemistry and eGFR by treatment group and time point
- A summary of the change (absolute and percent) from baseline in Chemistry and eGFR by treatment group and time point

- A summary of the number and percentage of subjects experiencing abnormal values by laboratory parameter, treatment group and time point

The following treatment-emergent laboratory test abnormalities of interest will be summarized

- Subjects experiencing at least 1 potassium > upper limit of normal
- Subjects experiencing at least 1 uric acid > upper limit of normal
- Subjects experiencing at least 1 ALT or AST > upper limit of normal
- Subjects experiencing at least 1 ALT or AST > 2x upper limit of normal
- Subjects experiencing at least 1 ALT or AST > 3x upper limit of normal

The eGFR will be calculated from serum creatinine by following formula:

$$\text{eGFR}(\text{mL/min/1.73m}^2) = 194 \times (\text{Scr in mg/dL})^{-1.094} \times (\text{Age})^{-0.287} \times (0.739 \text{ if female})$$

A by-subject listing of Chemistry and eGFR will be provided by treatment group.

4.8.4.8 C-reactive protein and Vascular Endothelial Growth Factor (VEGF)

The following summaries will be provided:

- A summary of C-reactive protein (CRP) and VEGF by treatment group and time point
- A summary of the change (absolute and percent) from baseline in CRP and VEGF by treatment group and time point

The effect of treatment in terms of the change from baseline to Week 6 in CRP and VEGF will be analyzed using ANCOVA. The statistical model will assess the main effect of treatment, adjusted for baseline measurement. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done. Unadjusted ANOVA will be performed for sensitivity.

A by-subject listing of CRP and VEGF will be provided by treatment group.

4.8.5 Vital Signs, Physical Findings and Other Observations Related to Safety

4.8.5.1 Vital Signs

The following variables will be analyzed:

- Blood Pressure (Systolic / Diastolic)
- Heart Rate
- Respiratory Rate
- Body Temperature

The following summaries will be provided:

- A summary of each vital sign parameter by treatment group and time point.
- A summary of the change (absolute and percent) from baseline in each vital sign parameter by treatment group and time point.

Any findings of treatment-emergent vital sign abnormalities will be handled as adverse event and reported in AE tables / listings.

A by-subject listing of vital sign parameter with reference ranges will be provided.

4.8.5.2 12-lead ECG

The following variables will be analyzed at the Baseline Visit:

- Heart rate
- PR interval
- QT interval
- QRS interval
- QTc (corrected)
- Overall result

A summary of each ECG parameter will be provided by treatment group.

A by-subject listing of 12-lead ECG results will be provided.

4.8.5.3 Physical Examination and Weight Assessment

Physical Examination and Weight Assessment will be performed at the Screening Visit and used for subject's inclusion / exclusion. Weight and height will be reported in demographic tables and listings.

4.8.6 Safety Monitoring (Independent Data Monitoring Committee [IDMC], Data Monitoring Committee [DMC], Data and Safety Monitoring Board [DSMB])

Not applicable. No change of study conduct based on preliminary analysis result is planned.

4.9 Other Analyses

4.9.1 PK Analysis

The following summary will be provided:

- The summary of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.
- Box plots of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.
- Dose-normalized box plots of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.

By-subject listings of pre-dose plasma concentrations of vadadustat and its metabolites will be provided by treatment group.

4.10 Determination of Sample Size

The target enrollment will be approximately 48 subjects for the study with 12 subjects enrolled in each of the 4 treatment groups.

The study is powered based on a comparison of the highest dose vadadustat group (600 mg daily) and the placebo group. Based on the results from Study AKB-6548-CI-0005 that was conducted in the United States and evaluated vadadustat in patients with anemia secondary to NDD-CKD, it is assumed that the expected mean Hb changes from baseline to Week 6 will be approximately 0 mg/dL for the placebo group and 1.35 g/dL for the highest dose vadadustat group (600 mg daily), with a common standard deviation of 0.68 g/dL across treatment groups. With these assumptions, the study with n=12 subjects per group will have >95% power to detect the difference between treatment group and control group, at 2-sided alpha of 0.05.

4.11 Changes in the Conduct of the Study or Planned Analysis

Not Applicable.

5 REFERENCES

Not Applicable.

PAREXEL International

Akebia Therapeutics, Inc.

AKB-6548-CI-0021

PHASE 2, RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED, DOSE-FINDING STUDY TO ASSESS THE EFFICACY, SAFETY, PHARMACOKINETICS, AND PHARMACODYNAMICS OF VADADUSTAT IN JAPANESE SUBJECTS WITH ANEMIA SECONDARY TO NON-DIALYSIS DEPENDENT CHRONIC KIDNEY DISEASE (NDD-CKD)

Statistical Analysis Plan

PAREXEL Project Number: 230644

Status; Date: Version 1; 08 March 2017 (Original SAP)
Version 2; 15 May 2017

SPONSOR SIGNATURE PAGE

Approved by: [redacted] [redacted]
Date

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Akebia Therapeutics, Inc.

PAREXEL SIGNATURE PAGE

This document has been approved and signed electronically on the final page by the following:

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	[redacted] Project Role: Primary Statistical Programmer
	[redacted] Project Role: MWS Reviewer

TABLE OF CONTENTS

LIST OF ABBREVIATIONS

AE	adverse event
ALT	alanine aminotransferase
AST	aspartate aminotransferase
BMI	body mass index
CBC	complete blood count
CI	confidence interval
ECG	Electrocardiogram
eCRF	electronic Case Report Form
eGFR	estimated glomerular filtration rate
EOT	end-of-treatment
ESA	erythropoiesis-stimulating agent
Hb	Hemoglobin
ITT	intent-to-treat
IV	Intravenous
LOCF	last observation carried forward
LS mean	least-squares mean
MedDRA	Medical Dictionary for Regulatory Activities
MITT	modified intent-to-treat
NDD-CKD	non-dialysis dependent chronic kidney disease
PD	pharmacodynamic(s)
PK	pharmacokinetic(s)
PP	per protocol
PT	preferred term
Q1	1st quartile
Q3	3rd quartile
RBC	red blood cell
SAE	serious adverse event
SAP	statistical analysis plan
SMQ	standardised MedDRA queries
SOC	system organ class
TEAE	treatment-emergent adverse event
TIBC	total iron binding capacity
TSAT	transferrin saturation
VEGF	vascular endothelial growth factor

1 INTRODUCTION

This study is a Phase 2 clinical study to assess the dose-response relationship between oral vadadustat once daily dosing for 6 weeks and the change in hemoglobin (Hb) in Japanese subjects with anemia secondary to non-dialysis dependent chronic kidney disease (NDD-CKD); in order to define the starting dose for use in Phase 3 clinical studies in Japan.

This Statistical Analysis Plan (SAP) defines the preliminary and final statistical analyses for the clinical study report for this study. This should be read in conjunction with the study protocol that provides all necessary background information and rationale for the current study and its design.

The pharmacokinetic data will be separately obtained from vendor to keep blindness of the study.

This SAP is based upon the following study documents:

- Study Protocol, Version 4 (December 15, 2016)
- electronic Case Report Form (eCRF), Version 3.0 (October 6, 2016)
- Protocol Deviation Specification Form, Version 1.0 (September 29, 2016)

2 STUDY OBJECTIVES

2.1 Primary Objective and Endpoint

The primary objective of this study is to assess the dose-response relationship between oral vadadustat once daily dosing for 6 weeks and the change in Hb in Japanese subjects with anemia secondary to NDD-CKD; this is to define the starting dose for use in Phase 3 clinical studies in Japan.
The primary endpoint that will be used to assess this objective is the mean change in Hb levels from pre-treatment to the end of the primary efficacy period (Week 6). Pre-treatment Hb is defined as the average of 2 Hb values obtained prior to treatment based on the qualifying screening Hb value and the Hb value at the baseline visit.

2.2 Secondary Objectives and Endpoints

The secondary objectives of this study are:

- To assess the safety, tolerability, pharmacokinetics (PK), and pharmacodynamics (PD) of oral vadadustat once daily dosing in Japanese subjects with anemia secondary to NDD-CKD during the 6-week, primary efficacy period
- To evaluate the effect of dose adjustments and demonstrate the maintenance effect on Hb during a 10-week dose-adjustment and maintenance period
- To assess the time to reach the target Hb range from baseline

The efficacy endpoints that will be used to assess these objectives include the following:

- Time to reach target Hb level of 10.0-12.0 g/dL from baseline visit (Day 1)
- Mean Hb levels at the end of the primary efficacy period (Week 6) and at the end of the dose adjustment and maintenance period (Week 16)
- Proportion of subjects who achieve target Hb 10.0-12.0 g/dL at the end of the dose adjustment and maintenance period (Week 16)
- Mean change in Hb between pre-treatment and the end of the dose adjustment and maintenance period (Week 16)
- Mean change in hematocrit, red blood cell (RBC) count, and reticulocyte count from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Mean change in iron indices (ie, iron, total iron-binding capacity [TIBC], transferrin saturation [TSAT], and ferritin) and hepcidin from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Proportion of subjects requiring rescue with RBC transfusion from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Proportion of subjects requiring rescue with ESAs from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Number of dose adjustments from baseline to the end of the dose adjustment and maintenance period (Week 16)
- Maintenance of iron sufficiency (defined as ferritin ≥50 ng/mL and TSAT ≥20%) from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Plasma concentration profile of vadadustat and its metabolites using pre-dose sample from Week 4

The safety endpoints that will be used to assess these objectives include the following:

- Safety assessments, including adverse events (AEs), vital signs, electrocardiograms (ECGs), and other laboratory assay results (eg, chemistry, components of the complete blood count [CBC] other than the ones noted above, and vascular endothelial growth factor [VEGF])

3 INVESTIGATIONAL PLAN

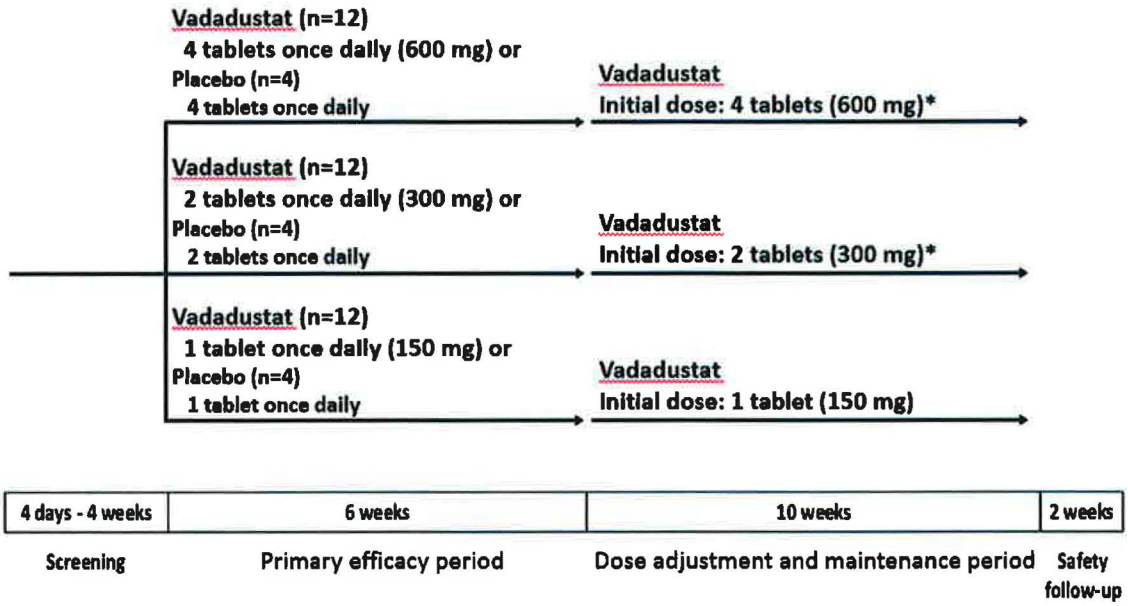
3.1 Overall Study Design and Plan

This is a Phase 2, randomized, double-blind, placebo-controlled, dose-finding study to assess the efficacy, safety, tolerability, PK, and PD of orally administered vadadustat in Japanese subjects with anemia secondary to NDD-CKD.

The study has a planned enrollment of 48 subjects to be enrolled at approximately 25 sites in Japan. There will be 16 subjects in each of the 3 tablet-count groups.

An overview of the study design is presented in Figure 1.

Figure 1: Overview of Study Design



* For subjects who develop an excess Hb response during the primary efficacy period, the number of tablets of study drug will be decreased (see Section 8.2.4). For these subjects, the number of tablets of vadadustat initiated at the Week 6 visit will be lower than indicated.

The study will include the following periods:

- Eligibility screening period (up to 4 weeks)
- Primary efficacy period (6 weeks; Weeks 1 to 6)
- Dose adjustment and maintenance period (10 weeks; Weeks 7 to 16)
- Follow-up period (2 weeks; Weeks 17 and 18)

Subjects will participate in a screening period (4 days to 4 weeks) to determine study eligibility, and eligible subjects will be randomized following the screening period.

Using a central randomization system, subjects will be randomized 1:1:1 to receive 1, 2, or 4 tablets at their baseline visit (Day 1). Within each tablet-count group, subjects will be randomized 3:1 to receive vadadustat (150, 300, or 600 mg vadadustat) or placebo. See the protocol section 8.2.2 for information regarding the randomization scheme.

Study drug treatment will be administered during a 6-week primary efficacy period. See the protocol section 8.2.4 for information on study drug administration. The primary efficacy period includes fixed-dose treatment to establish a dose-response relationship. However, if Hb level increase rapidly or if the Hb level exceeds 13.0 g/dL, the study drug dose can be decreased or interrupted (see the protocol section 8.2.4).

After completing the primary efficacy period, subjects will continue to a 10-week dose adjustment and maintenance period (see the protocol section 8.2.5). Subjects receiving placebo will be switched to vadadustat, and study drug dose will be adjusted to achieve a target Hb of 10.0-12.0 g/dL based on dose adjustment guidelines (see the protocol section 8.2.5).

Vadadustat treatment will stop after the dose adjustment and maintenance period has been completed (Week 16) and subjects will continue in a 2-week follow-up period (Week 17-18).
The clinical and safety assessments will be performed as described in the protocol section 9.3 and as listed in the protocol Appendix A.

3.2 Efficacy and Safety Variables

3.2.1 Efficacy and PD Variables

The following variable is the primary efficacy variable:

- Change from pre-treatment in Hb at Week 6.

The following efficacy endpoints will also be analyzed:

- Actual values and change (absolute and percent) from pre-treatment in Hb, hematocrit, RBC count, and reticulocyte count (both absolute and percent)
- Actual values and change (absolute and percent) from pre-treatment in iron, TIBC, TSAT, ferritin, and hepcidin

3.2.2 Safety Variables

The following variables are the safety endpoints:

- AEs
- Vital signs
- ECGs
- Laboratory parameters

3.2.3 PK Variables

The following variables are the PK variable:

- Pre-dose plasma concentrations of vadadustat at the Week 4 visit
- Pre-dose plasma concentrations of vadadustat's metabolites at the Week 4 visit

4 STATISTICAL METHODS

4.1 Data Quality Assurance

All tables, figures and data listings to be included in the report will be independently checked for consistency, integrity and in accordance with standard PAREXEL procedures.

4.2 General Presentation Considerations

'Pre-treatment' of Hb is defined as the average of 2 Hb values obtained prior to treatment based on the qualifying screening Hb value and the Hb value at the Baseline Visit. 'Pre-

treatment' for other variables is defined as the last available pre-treatment assessment. In general, this is the value at the Baseline Visit. 'End of Treatment (EOT)' is defined as the assessment on the EOT form labeled as 'Week 16 Visit', even if other values closer to Day 113 are available. 'Treatment Day' will be calculated relative to start of dosing at Baseline Visit (i.e. Treatment Day = Assessment Date - Baseline Visit Date + 1).

Continuous data will be summarized in terms of the mean, standard deviation (SD), median, minimum, maximum and number of observations, unless otherwise stated. The minimum and maximum will be reported to the same number of decimal places as the raw data recorded in the database. The mean and median will be reported to one more decimal place than the raw data recorded in the database. The SD will be reported to two more decimal places than the raw data recorded in the database. In general, the maximum number of decimal places reported shall be four for any summary statistic.

Categorical data will be summarized in terms of the number of subjects providing data at the relevant time point (n), frequency counts and percentages. Any planned collapsing of categories will be detailed in the SAP text and the data displays.

Percentages will be presented to one decimal place. Percentages will not be presented for zero counts. Percentages will be calculated using n as the denominator. If sample sizes are small, the data displays will show the percentages, but any textual report will describe frequencies only.

Changes from baseline in categorical data will be summarized using shift tables where appropriate.

P-values greater than or equal to 0.001, in general, will be presented to three decimal places. P-values less than 0.001 will be presented as "<0.001".

Confidence intervals will be presented to one more decimal place than the raw data.

All report outputs will be produced using SAS® version 9.3 or a later version in a secure and validated environment. All report outputs will be provided to the Sponsor in a single Microsoft Word 2010 [Version 14.0] document.

4.3 Study Subjects

4.3.1 Disposition of Subjects

A clear accounting of the disposition of all subjects who enter the study will be provided, from screening to study completion.

The summary of the following subject disposition will be presented.

- A summary of the number of subjects randomized, the number and percentage of subjects treated (with at least one dose of study medication) and the number and percentage of subjects completed through Week 6 Visit and discontinued by Week 6 Visit by treatment group and overall (Analysis population: All Subjects

Randomized). For final analysis, a summary of the number and percentage of subjects completed Week 18 Visit and discontinued from the study by treatment group and overall will be provided (Analysis population: All Subjects Randomized). Withdrawals from the study will also be summarized by major reason.

By-subject listings of eligibility details, randomization details, visit dates and withdrawal/study completion details (including reason for discontinuation and duration of treatment prior to discontinuation) will be provided.

4.3.2 Protocol Deviations

Major protocol deviations are defined as those deviations from the protocol likely to have an impact on the perceived efficacy and/or safety of study treatments. The impact of major protocol deviations on the efficacy and/or safety results will be evaluated prior to unblinding to determine which data and/or subjects, if any, to exclude from the analysis populations (see Section 4.4),

Protocol deviations and any action to be taken regarding the exclusion of subjects or affected data from specific analyses will be determined in Protocol Deviation Specification document 20170207 version 2.0.

The protocol deviation will be summarized as following:

- A summary of the number and percentage of subjects with a protocol deviation by treatment group and overall and by type of deviation and by period (Analysis population: All Subjects Randomized)

A by-subject listing of protocol deviations will be provided.

4.4 Analysis Populations

Upon database release, protocol deviation and analysis population outputs will be produced and will be sent to Sponsor for review. An analysis population classification meeting will be arranged to discuss the outputs and to decide which subjects and/or subject data will be excluded from certain analyses. Decisions made regarding the exclusion of subjects and/or subject data from analyses will be made prior to unblinding and will be documented and approved by Sponsor.

The following summary of analysis population will be provided:

- A summary of the number and percentage of subjects allocated to each analysis population by treatment group and overall (Analysis population: All Subjects Randomized)

A by-subject listing of analysis population details will be provided. This listing will be presented by treatment group and include: site, subject identifier, inclusion/exclusion flag

for each population and reason for exclusion from each population. All subjects randomized will appear on this listing.

4.4.1 Analysis Population for the Safety Analyses

The safety summaries and analyses will be based on the **safety population**. The safety population is defined as all enrolled subjects who receive at least 1 dose of study medication. In the event that a subject is allocated the incorrect study treatment as per the study randomization list, subjects will be summarized and analyzed 'as treated' i.e. by allocated treatment group.

4.4.2 Analysis Populations for the Efficacy Analyses

The efficacy summaries and analyses will be based on the **modified intent-to-treat (MITT) population**. The MITT population is defined as all randomized subjects who have received at least 1 dose of study medication and have a pre-treatment Hb average defined as the average of the qualifying screening value and the baseline value, and at least one post-baseline Hb measurement. In the event that a subject is allocated the incorrect study treatment as per the study randomization list, subjects will be summarized and analyzed 'as randomized' i.e. by randomized treatment group.

For the efficacy variables, sensitivity analyses will be performed on the **per-protocol (PP) population** to assess the robustness of the study conclusions to the choice of analysis population. The PP population is defined as the subjects in the MITT population who have completed the study and have efficacy data through Week 6, have a study medication compliance of $\geq$80%, and do not have any major protocol deviations expected to significantly affect the primary efficacy endpoint. Subjects with major protocol deviations expected to significantly affect the primary efficacy endpoint will be identified and documented prior to data unblinding.

4.4.3 Analysis Population for the PK Analysis

The PK summaries and analyses will be based on the **pharmacokinetics (PK) population**. The PK population is defined as all subjects in the safety population who have a pre-dose PK sample at Week 4. Subjects will be summarized and analyzed by actual treatment group.

4.5 Demographic and Other Baseline Characteristics

The demographic and other baseline characteristics will be summarized for the MITT and safety populations. The following summaries will be provided:

- A summary of demographic variables (e.g. age, gender, weight, height and body mass index [BMI]) by treatment group and all subjects

- A summary of other relevant variables (e.g. estimated Glomerular Filtration Rate [eGFR] at baseline, CKD stage, etiology of CKD and smoking status) by treatment group and all subjects
- A summary of relevant medical history by treatment group and all subjects.
- A summary of prior and concomitant medications within 30 days prior to the first study drug dose up to and including Week 6 by treatment group and overall.
- For the final analysis, a summary of concomitant medications after Week 6 will be provided.

Age will be calculated as the number of complete years between a subject's birth date and the date of informed consent.

Definitions of eGFR and CKD stage are described in section 4.8.4.8.

CKD stage will be determined by eGFR at baseline:

CKD stage	eGFR (mL/min/1.73m^2)
1	90+
2	60 - 89
3a	45 - 59
3b	30 - 44
4	15 - 29
5	< 15

By-subject listings of demographic data and other baseline characteristics (as summarized above) will be provided

4.6 Treatment Compliance

The treatment compliance will be summarized for the MITT and safety population. The following summary will be provided.

- A summary of the treatment compliance measures (i.e. ≥80% compliance and <80%) by treatment group and study visit.

A by-subject listing of treatment compliance data will be provided.

4.7 Efficacy Evaluation

4.7.1 Analysis and Data Conventions

This study is designed to test for dose-response relationship. An analysis of covariance (ANCOVA) model will be used to compare absolute change from pre-treatment in Hb between the three vadadustat dosing groups and the placebo group. The model will include treatment assignment (3 dosed groups and one placebo group) and pre-treatment Hb value as a covariate. A step-down procedure will be used to control the overall type I error rate for the multiple comparisons. Testing of the highest dose compared with

placebo will be conducted first. If and only if this comparison is significant, then testing will proceed to comparison of the next lower dose and placebo, and so on. Therefore, no multiplicity adjustment will be needed for this analysis.

A linear regression dose-response modeling will be performed as a secondary analysis.

$$Y = \beta_0 + \beta_{dose} X$$

where Y is change in Hb and X is vadadustat dose. β_xs are parameters of the model. β_0 is intercept and β_{dose} is slope, respectively.
The null hypothesis for the dose response will be that the regression coefficient of slope parameter is zero. The alternative hypothesis will be that the regression coefficient is not zero. Symbolically, this is expressed as follows:

$$H_0: \beta_{dose} = 0$$
$$H_1: \beta_{dose} \neq 0$$

This hypothesis will be tested using a 0.05 two-sided significance level.

4.7.1.1 Multi-center Studies
Not Applicable. Analysis of center effect is not planned.

4.7.1.2 Adjustments for Covariates
The primary efficacy analysis will be adjusted for the pre-treatment measure of primary efficacy variable.

An 'unadjusted' sensitivity analysis will be performed to assess the robustness of the study conclusion.

4.7.1.3 Handling of Missing Data
For the MITT population analyses for the primary efficacy variable, the Last Observation Carried Forward (LOCF) approach will be used to impute missing data.

- If a Baseline value is missing, the last value prior to baseline will be carried forward to the Baseline value but not to further values during the treatment period.
- For post-Baseline assessments, hemoglobin measurements at end of treatment and follow-up visits are not carried forward. Other than that, the most recent post-baseline assessments to the primary evaluation period (week 6) will be carried forward . Baseline and pre-Baseline values will not be carried forward to the post-Baseline visits.

For the secondary endpoint analyses, data will be used as observed and no imputation method will be used.

4.7.1.4 Multiple Comparisons/Multiplicity

Analysis of one primary variable [Change in Hb] and one time point of primary interest [Week 6 visit] has been defined for this study. Though 3 pairwise comparison between placebo and dosed group is planned, step-down procedure will be used to control the overall type I error rate. The secondary variables defined are intended to provide supportive evidence relating to the primary objective and no labeling claims are intended. Though one preliminary analysis is planned before the final analysis, primary analysis will not be changed. Hence no adjustments for multiplicity are required.

4.7.1.5 Preliminary (6-Week) Analysis

After all subject performed assessment of the Week 6 Visit, preliminary analysis will be performed. As the study conduct and final analysis will not be modified by this analysis, no alpha adjustment is proposed. It is expected that this 6-week data
will be identical for the preliminary analysis and the final analysis of the complete dataset. Any changes between the preliminary analysis and the final analysis will be documented. The preliminary analysis will be performed by the independent unblinded team. All data through the cutoff date of the last subject randomized who completes week 6 will be analyzed. Blinding of blinded team and investigators will be maintained to prevent potential bias.

4.7.1.6 Examination of Subgroups

Not Applicable. No subgroup analysis is planned.

4.7.2 Primary Efficacy Variable – Mean Change in Hb from Pre-treatment to the End of the Primary Efficacy Period (Week 6)

The primary variable for the assessment of efficacy is the change from pre-treatment to Week 6 in Hb (g/dL). Change in Hb is defined as the Hb measured at the Week 6 visit minus the mean pre-treatment Hb. Pre-treatment Hb is defined as the average of 2 Hb values obtained prior to treatment based on the qualifying screening Hb value and the Hb value at the baseline visit.

For the primary analysis, the LOCF approach will be used to impute missing data. Complete-case analysis will be performed for sensitivity at the scheduled Week 6 visit.

4.7.2.1 Summary of actual value and change in Hb at week 6

The primary analysis population for efficacy will be MITT and PP population. The following summary will be provided:

- A summary of actual value of Hb by treatment group at baseline and week 6. A summary of the change (absolute and percent) from baseline in Hb by treatment group at week 6. 1st and 3rd quartiles (Q1, Q3) as well as the mean, SD, median, minimum, maximum and number of observations will be shown. P values for overall comparison and treated groups vs. placebo on change from baseline will also be presented.

- A waterfall plot for absolute Hb change from baseline for each treatment group at week 6.

The effect of treatment in terms of the change from pre-treatment to Week 6 in Hb will be analyzed using a one-way analysis of covariance (ANCOVA). The statistical model will assess the main effect of treatment, adjusted for pre-treatment average. Least-squares (LS) means for each treatment and their 95% confidence intervals (CIs) will be presented. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done. Unadjusted one-way analysis of variance (ANOVA) will be performed for sensitivity.

The simple linear regression between the change from pre-treatment to Week 6 in Hb and dose will be calculated. The regression coefficients and their standard errors, p-values of test of regression parameters and their 95%CIs will be presented. The test of slope parameter will be used to evaluate dose response relationship.

As sensitivity analyses, efficacy endpoints will also be analyzed using the PP population.

A by-subject listing of actual values and change (absolute and percent) from baseline in Hb will be provided.

4.7.2.2 Summary of actual value and change in Hb from baseline to week 6 and beyond

The analysis population for efficacy will be MITT and PP population. The following summary will be provided:

- A summary of actual value of Hb by treatment group and study visits. 1st and 3rd quartiles (Q1, Q3) as well as the mean, SD, median, minimum, maximum and number of observations will be shown. Summary of pre-treatment average will also be presented.
- A summary of the change (absolute and percent) from baseline in Hb by treatment group and study visits.
- A plot showing the mean Hb value over time within each treatment group. 95% CI of these means will be shown as error bar and sample size in each group at each time point will be shown.
- Plots of individual profile of Hb values over time within each treatment group.
- Box plots of absolute Hb changes from baseline for each treatment group at each post-treatment study visit to week 6
- Box plots of absolute Hb changes from baseline for each treatment group at each post-treatment study visit to week 16

A by-subject listing of actual values and change (absolute and percent) from baseline in Hb will be provided.

4.7.3 Secondary Efficacy Variables

The analysis population for efficacy will be MITT and PP population.

Time to reach target Hb Range

Time to reach target Hb level of 10.0-12.0 g/dL from baseline is defined for subjects with Hb <10.0 g/dL at the baseline visit as following:
First post-treatment assessment date with value of Hb ≥10.0 and ≤12.0 g/dL – the baseline visit date +1. If target Hb level is not achieved, last Hb assessment date will be used as censored date.

A summary of time to reach target Hb level will be provided for subjects with Hb <10.0 g/dL at the baseline visit by treatment group.

Kaplan-Meier failure (cumulative incidence) plot will be presented for the time to reach target Hb level by treatment group.

A by-subject listing will be provided.

Hb within target Range

A summary of the number and percentage of subjects within target Hb 10-12 g/dL at the end of the dose adjustment and maintenance period (Week 16) will be provided by treatment group.

The number and percentage of subjects above / within / below target range who are maintained within the target range at their previous visit after Week 6 will be provided by treatment group.

The effect of treatment in terms of the proportion of subjects within target Hb level at Week 6 will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

A by-subject listing will be provided.

Increase in Hb from pre-treatment average

A summary of the number and percentage of subjects who achieved a ≥1 g/dL increase in Hb from pre-treatment average will be provided by treatment group.

The effect of treatment in terms of the proportion of subjects who achieved a ≥1 g/dL increase in Hb from pre-treatment average will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

Hematocrit, RBC count, and reticulocyte count

Hematocrit, RBC count, and reticulocyte count (both absolute and percent) will be analyzed as secondary efficacy variables.

The following summaries will be provided:

- A summary of hematocrit, RBC count, and reticulocyte count (both absolute and percent) by treatment group and time point. 1st and 3rd quartiles (Q1, Q3) as well as

the mean, SD, median, minimum, maximum and number of observations will be presented.
- A summary of the change (absolute and percent) from baseline in hematocrit, RBC count, and reticulocyte count (both absolute and percent) by treatment group and time point
- Box plots of absolute changes from baseline in hematocrit, RBC count, and reticulocyte count (both absolute and percent) for each treatment group at each post-treatment time point

The effect of treatment in terms of the change from baseline to Week 6 in hematocrit and RBC count and reticulocyte count (both absolute and percent) will be analyzed using ANCOVA. The statistical model will assess the main effect of treatment, adjusted for baseline measurement. LS-means for each treatment and their 95% CIs will be presented. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done. Unadjusted ANOVA will be performed for sensitivity.

A by-subject listing of hematocrit, RBC count, and reticulocyte count (both absolute and percent) will be provided by treatment group.

Iron indices and hepcidin

Iron indices (ie, iron, TIBC, TSAT, and ferritin) and hepcidin will be analyzed as secondary efficacy variables.

The following summaries will be provided:
- A summary of iron indices and hepcidin by treatment group and time point
- A summary of the change (absolute and percent) from baseline in iron indices and hepcidin by treatment group and time point
- Box plots of absolute changes from baseline in iron indices and hepcidin for each treatment group at each post-treatment time point

A by-subject listing of iron indices and hepcidin will be provided by treatment group.

Rescue therapy

Initiation of rescue therapy will be analyzed.

The following summaries will be provided:
- A summary of the number and percentage of subjects requiring rescue with RBC transfusion from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- A summary of the number and percentage of subjects requiring rescue with ESAs from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)

The effect of treatment in terms of the proportion of subjects requiring rescue with RBC transfusion / ESAs will be analyzed using Fisher's exact test. Three post-hoc pairwise

comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

By-subject listings of rescue therapies will be provided by treatment group.

Average Daily Dose
A summary of Average Daily Dose by treatment group and visit will be presented with 1st and 3rd quartiles (Q1, Q3) as well as the mean, SD, median, minimum, maximum and number of observations.

Dose adjustment
A overview summary of the number of dose adjustments from baseline to the end of the dose adjustment and maintenance period (Week 16) will be provided.
A summary of the number of dose adjustments by visit from baseline to the end of the dose adjustment and maintenance period (Week 16) will be provided.

A by-subject listing of dose adjustment will be provided by treatment group.

Maintenance of iron sufficiency
A summary of maintenance of iron sufficiency (defined as ferritin ≥50 ng/mL and TSAT ≥20%) from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16) will be provided.

The effect of treatment in terms of the proportion of subjects who maintain iron sufficiency will be analyzed using Fisher's exact test for week 6 only. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

A by-subject listing of iron sufficiency will be provided by treatment group.

4.7.4 Exploratory Efficacy Variable

Time in Therapeutic Range (TTR) for Hb from week 6 to week 20
The half-time interpolation method will be used to calculate TTR for Hb. The total time of follow-up with Hb in target range of 10.0 to 12.0 g/dL is divided by the total time. Half the time between two tests is allocated to the first Hb value, and half to the second Hb value.

A summary of TTR for Hb will be provided by treatment group.

A by-subject listing of TTR for Hb will be provided by treatment group

4.8 Safety Evaluation

All safety summaries and analyses will be based upon the safety population as defined in Section 4.4.

4.8.1 Extent of Exposure

The extent of exposure will be summarized for the safety population. The following summary will be provided.

- A summary of the treatment duration (e.g. End Date of Administration - Start date of Administration +1) by treatment group and study visits.

A by-subject listing of start, end and duration of treatment will be provided.

4.8.2 Adverse Events

AEs will be coded using the Medical Dictionary for Regulatory Activities (MedDRA) Version 19.0.

Treatment-emergent adverse events (TEAEs) will be tabulated and are defined as those AEs that either start or worsen in severity on or after the date/time of first dose of study treatment and on or before the final protocol required visit. In general, it is Week 18 Visit and 14 days after the date of last dose of study treatment. The TEAEs are defined as those adverse events that started after first injection up to 14 days post-last dose.

Where dates are missing or partially missing, AEs will be assumed to be treatment-emergent, unless there is clear evidence (through comparison of partial dates) to suggest that the AE started prior to the first dose of study treatment or more than 14 days after the last dose of study treatment.

Partial AE and concomitant medication start dates will be imputed as follows:

1. If the year (and month and day) is unknown, the date will be imputed as the date of first intake of study treatment if this is possible (i.e., the event end date is not prior to treatment start date).

2. If the month and day is unknown then:

    a. If the year matches the first dose date, then impute the month and day of the first dose date.

    b. If the year is earlier than the first dose date or the first dose date is not available (subject is not dosed) and matches the year of informed consent date, then impute the month and day of the informed consent date;

    c. Otherwise, assign January 1.

3. If the day is unknown then:

a. If the month and year match the first dose date, then impute the day of the first dose date.

b. If the month and year are earlier than the first dose date or the first dose date is not available (subject is not dosed) and matches the month and year of the informed consent date, then impute the day of the informed consent date;

c.Otherwise, assign '1'.

Partial AE and concomitant medication stop dates will be imputed as follows:

1. If the year is unknown, the date will not be imputed and will be assigned a missing value.

2. If the month is unknown, then assign December.

3. If the day is unknown, then assign the last day of the month.

For all other settings (eg. year missing, but day and month available etc.), imputation strategies will be reviewed and decided in the ongoing data cleaning and sponsor meetings.

If the start and/or stop dates of medications other than study treatments are missing or partially missing, the dates will be compared as far as possible with the date of first study treatment administration.

If not specified, observed data approach will be performed.

The AE will be summarized using MedDRA system organ class (SOC) and preferred term (PT). All AE summaries will provide the number of subjects reporting at least one AE and the total number of events reported. The following summaries will be provided:

- A summary of the number and percentage of subjects reporting at least one TEAE, drug-related TEAE, TEAE leading to withdrawal, serious adverse event (SAE), drug-related SAE, SAE leading to withdrawal and death by treatment group.
- A summary of the number and percentage of subjects reporting a TEAE by treatment group, SOC, and PT
- A summary of the number and percentage of subjects reporting a drug-related TEAE by treatment group, SOC, and PT
- A summary of the number and percentage of subjects reporting a treatment-emergent SAE by treatment group, SOC and PT
- A summary of the number and percentage of subjects reporting a drug-related treatment-emergent SAE by treatment group, SOC, and PT
- A summary of the number and percentage of subjects reporting a drug-related TEAE leading to withdrawal by treatment group, SOC, and PT
- A summary of the most common TEAEs by treatment group and PT (reported by >10% of subjects in any treatment group).
- A summary of the number and percentage of subjects reporting a TEAE by treatment group, severity, SOC and PT
- A summary of the number and percentage of subjects reporting a TEAE by treatment group, relatationship, SOC and PT

AE summaries will be ordered alphabetically for SOC, and PT within SOC.

For each subject and each AE, the worst severity recorded will be attributed and used in the by-severity summaries. Similarly, the worst causality (most related to treatment) will be attributed and used in the by-causality summaries. If severity or causality is missing, data will be treated as observed

A by-subject listing of all AEs (including non-treatment-emergent events) will be provided. This listing will be presented by treatment group and will include: center, subject identifier, age, sex, race, AE (SOC, PT, and verbatim term), date of onset, date of resolution, duration, severity, seriousness, action taken, outcome and causality.

The following AEs are defined as AEs of special interest. Summaries of the number and percentage of subjects reporting the following AEs will be provided.

- PT: "hyperkalemia" (MedDRA 10020646) or "blood potassium increased" (MedDRA 10005725)
- PT: "hyperuricaemia" (MedDRA 10020903) or "blood uric acid level increased" (MedDRA 10022891)
- PT: "Hypertension" (MedDRA 10020772) or "Blood pressure increased" (MedDRA 10005750) or "Blood pressure systolic increased" (MedDRA 10005760) or "Blood pressure diastolic increased" (MedDRA 10005739) or "Blood pressure ambulatory increased" (MedDRA 10005732) or "Blood pressure inadequately controlled" (MedDRA 10051128) or "Accelerated hypertension" (MedDRA 10000358) or "Essential hypertension" (MedDRA 10015488) or "Diastolic hypertension" (MedDRA 10012758) or "Systolic hypertension" (MedDRA 10042957) or "Malignant hypertension" (MedDRA 10025600)
- Standardised MedDRA Query (SMQ): "Cholestasis and jaundice of hepatic origin" (SMQ 20000009) or "Drug related hepatic disorders – severe events only" (SMQ 20000007) or "Liver related investigations, signs and symptoms" (SMQ 20000008)
- Standardised MedDRA Query (SMQ): "Ischaemic heart disease" (SMQ 20000043)
- Standardised MedDRA Query (SMQ): "Central nervous system haemorrhages and cerebrovascular conditions" (SMQ 20000061)
- Standardised MedDRA Query (SMQ): "Embolic and thrombotic events" (SMQ 20000081)
- Standardised MedDRA Query (SMQ): "Cardiac failure" (SMQ 20000004)
- Standardised MedDRA Query (SMQ): "Retinal disorders" (SMQ 20000158, narrow)

Serious adverse events that occur during the screening period and are assessed by the investigator as related to study procedures will be tabulated.

4.8.3 Deaths, Serious Adverse Events, and Other Significant Adverse Events

Other significant AEs are those AEs reported as leading to withdrawal from study. The summaries of SAEs and AEs leading to withdrawal from study are described in section 4.8.2.

A summary of the number and percentage of deaths during the study will be provided by treatment group

A summary of the number and percentage of subjects Adverse Events Leading to Death by treatment group, severity, SOC and PT

A by-subject listing of all deaths that occurred during the study will be provided.

4.8.4 Clinical Laboratory Evaluation

4.8.4.1 Liver function abnormality

A summary of Liver function abnormality during the study will be provided by treatment group.

Any subject with at least one of the following liver function abnormalities is summarized:

- ALT ≥2x and <3x ULN
- AST ≥2x and <3x ULN
- Bilirubin ≥2x and <3x ULN
- ALT ≥3x ULN
- AST ≥3x ULN
- Bilirubin ≥3x ULN

4.8.4.2 Serum and urine pregnancy tests

Serum pregnancy test for females of childbearing potential will be performed at the screening visit and will be used for subject's inclusion / exclusion. Urine pregnancy test will be performed at the baseline visit and used for subject's initiation of study drug. Additional pregnancy tests may be conducted during the study to establish the absence of pregnancy based on the investigator's clinical judgment or as required by local regulations.

A by-subject listing of serum and urine pregnancy tests will be provided.

4.8.4.3 Coagulation tests

Coagulation tests will be performed at the baseline visit. The following variables will be analyzed.

- prothrombin time
- partial thromboplastin time
- international normalized ratio

A summary of baseline coagulation test results will be provided by treatment group.

A by-subject listing of coagulation tests will be provided.

4.8.4.4 Folate and vitamin B12

Folate and vitamin B12 tests will be performed at the screening visit and will be used for subject's inclusion / exclusion.

A by-subject listing of folate and vitamin B12 tests will be provided.

4.8.4.5 Urine albumin-to-creatinine ratio (uACR)
Urine albumin-to-creatinine ratio (uACR) will be evaluated at baseline visit.

A summary of uACR will be provided by treatment group.

A by-subject listing of uACR will be provided.

4.8.4.6 Complete Blood Count (CBC)
The following components of the CBC are described in Efficacy / PD analysis section.

- Hb
- Hematocrit
- RBC count
- Automated Reticulocyte Count (both absolute and percent).

The following components of the CBC will be analyzed:

- Mean Corpuscular Volume
- Mean Corpuscular Hb
- Mean Corpuscular Hb Concentration
- Red Cell Distribution Width
- White Blood Cell count with differential (neutrophils, bands, lymphocytes, monocytes, eosinophils, and basophils)
- Platelets

The following summaries will be provided:

- A summary of each component of the CBC by treatment group and time point
- A summary of the change from baseline in each component of the CBC by treatment group and time point
- A summary of the number and percentage of subjects experiencing abnormal values by component of CBC, treatment group and time point.

A by-subject listing of CBC will be provided by treatment group.

4.8.4.7 Hemoglobin
The following Hb analyses will be performed for safety analysis section:

- A summary of the number and percentage of subjects with any Hb > 12.0 g/dL by treatment group
- A summary of the number and percentage of subjects with any Hb > 13.0 g/dL by treatment group
- A summary of the number and percentage of subjects with any Hb > 14.0 g/dL by treatment group
- A summary of the number and percentage of subjects with Hb increase > 1.0 g/dL in any 2-week interval by treatment group

4.8.4.8 Chemistry and estimated glomerular filtration rate (eGFR)

The following laboratory parameters will be analyzed:

- Sodium
- Potassium
- Bicarbonate
- Chloride
- Calcium
- Phosphorus
- Glucose
- Creatinine
- Blood Urea Nitrogen
- Creatine Phosphokinase
- Uric Acid
- Albumin
- Total Protein
- Total Bilirubin
- Alkaline Phosphatase
- Alanine Aminotransferase (ALT)
- Aspartate Aminotransferase (AST)
- Lactate Dehydrogenase
- Total cholesterol
- Estimated Glomerular Filtration Rate (eGFR) (calculated from serum creatinine)

The following summaries will be provided:

- A summary of Chemistry and eGFR by treatment group and time point
- A summary of the change (absolute and percent) from baseline in Chemistry and eGFR by treatment group and time point
- A summary of the number and percentage of subjects experiencing abnormal values by laboratory parameter, treatment group and time point

The following treatment-emergent laboratory test abnormalities of interest will be summarized

- Subjects experiencing at least 1 potassium > upper limit of normal
- Subjects experiencing at least 1 uric acid > upper limit of normal
- Subjects experiencing at least 1 ALT or AST > upper limit of normal
- Subjects experiencing at least 1 ALT or AST > 2x upper limit of normal
- Subjects experiencing at least 1 ALT or AST > 3x upper limit of normal

The eGFR will be calculated from serum creatinine by following formula:

$$\text{eGFR}(\text{mL/min/1.73m}^2) = 194 \times (\text{Scr in mg/dL})^{-1.094} \times (\text{Age})^{-0.287} \times (0.739 \text{ if female})$$

A by-subject listing of Chemistry and eGFR will be provided by treatment group.

4.8.4.9 C-reactive protein and Vascular Endothelial Growth Factor (VEGF)

The following summaries will be provided:

- A summary of C-reactive protein (CRP) and VEGF by treatment group and time point
- A summary of the change (absolute and percent) from baseline in CRP and VEGF by treatment group and time point

The effect of treatment in terms of the change from baseline to Week 6 in CRP and VEGF will be analyzed using ANCOVA. The statistical model will assess the main effect of treatment, adjusted for baseline measurement. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done. Unadjusted ANOVA will be performed for sensitivity.

A by-subject listing of CRP and VEGF will be provided by treatment group.

4.8.5 Vital Signs, Physical Findings and Other Observations Related to Safety

4.8.5.1 Vital Signs

The following variables will be analyzed:

- Blood Pressure (Systolic / Diastolic)
- Heart Rate
- Respiratory Rate
- Body Temperature

The following summaries will be provided:

- A summary of each vital sign parameter by treatment group and time point.
- A summary of the change (absolute and percent) from baseline in each vital sign parameter by treatment group and time point.

Any findings of treatment-emergent vital sign abnormalities will be handled as adverse event and reported in AE tables / listings.

A by-subject listing of vital sign parameter with reference ranges will be provided.

4.8.5.2 12-lead ECG

The following variables will be analyzed at the Baseline Visit:

- Heart rate
- PR interval
- QT interval
- QRS interval
- QTc (corrected)
- Overall result

A summary of each ECG parameter will be provided by treatment group.

A by-subject listing of 12-lead ECG results will be provided.

4.8.5.3 Physical Examination and Weight Assessment

Physical Examination and Weight Assessment will be performed at the Screening Visit and used for subject's inclusion / exclusion. Weight and height will be reported in demographic tables and listings.

4.8.6 Safety Monitoring (Independent Data Monitoring Committee [IDMC], Data Monitoring Committee [DMC], Data and Safety Monitoring Board [DSMB])

Not applicable. No change of study conduct based on preliminary analysis result is planned.

4.9 Other Analyses

4.9.1 PK Analysis

The following summary will be provided:

- The summary of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.
- Box plots of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.
- Dose-normalized box plots of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.

By-subject listings of pre-dose plasma concentrations of vadadustat and its metabolites will be provided by treatment group.

4.10 Determination of Sample Size

The target enrollment will be approximately 48 subjects for the study with 12 subjects enrolled in each of the 4 treatment groups.

The study is powered based on a comparison of the highest dose vadadustat group (600 mg daily) and the placebo group. Based on the results from Study AKB-6548-CI-0005 that was conducted in the United States and evaluated vadadustat in patients with anemia secondary to NDD-CKD, it is assumed that the expected mean Hb changes from baseline to Week 6 will be approximately 0 mg/dL for the placebo group and 1.35 g/dL for the highest dose vadadustat group (600 mg daily), with a common standard deviation of 0.68 g/dL across treatment groups. With these assumptions, the study with n=12 subjects per group will have >95% power to detect the difference between treatment group and control group, at 2-sided alpha of 0.05.

4.11 Changes in the Conduct of the Study or Planned Analysis

Not Applicable.

5 REFERENCES

Not Applicable.

PAREXEL International

Akebia Therapeutics, Inc.

AKB-6548-CI-0021

PHASE 2, RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED, DOSE-FINDING STUDY TO ASSESS THE EFFICACY, SAFETY, PHARMACOKINETICS, AND PHARMACODYNAMICS OF VADADUSTAT IN JAPANESE SUBJECTS WITH ANEMIA SECONDARY TO NON-DIALYSIS DEPENDENT CHRONIC KIDNEY DISEASE (NDD-CKD)

Statistical Analysis Plan

PAREXEL Project Number: 230644

Status; Date: Version 1; 08 March 2017 (Original SAP)
Version 2; 15 May 2017
Version 3; 24 Jul 2017

SPONSOR SIGNATURE PAGE

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LIST OF ABBREVIATIONS

AE	adverse event
ALT	alanine aminotransferase
AST	aspartate aminotransferase
BMI	body mass index
CBC	complete blood count
CI	confidence interval
ECG	Electrocardiogram
eCRF	electronic Case Report Form
eGFR	estimated glomerular filtration rate
EOT	end-of-treatment
ESA	erythropoiesis-stimulating agent
Hb	Hemoglobin
ITT	intent-to-treat
IV	Intravenous
LOCF	last observation carried forward
LS mean	least-squares mean
MedDRA	Medical Dictionary for Regulatory Activities
MITT	modified intent-to-treat
NDD-CKD	non-dialysis dependent chronic kidney disease
PD	pharmacodynamic(s)
PK	pharmacokinetic(s)
PP	per protocol
PT	preferred term
Q1	1st quartile
Q3	3rd quartile
RBC	red blood cell
SAE	serious adverse event
SAP	statistical analysis plan
SMQ	standardised MedDRA queries
SOC	system organ class
TEAE	treatment-emergent adverse event
TIBC	total iron binding capacity
TSAT	transferrin saturation
VEGF	vascular endothelial growth factor

1 INTRODUCTION

This study is a Phase 2 clinical study to assess the dose-response relationship between oral vadadustat once daily dosing for 6 weeks and the change in hemoglobin (Hb) in Japanese subjects with anemia secondary to non-dialysis dependent chronic kidney disease (NDD-CKD); in order to define the starting dose for use in Phase 3 clinical studies in Japan.

This Statistical Analysis Plan (SAP) defines the preliminary and final statistical analyses for the clinical study report for this study. This should be read in conjunction with the study protocol that provides all necessary background information and rationale for the current study and its design.

The pharmacokinetic data will be separately obtained from vendor to keep blindness of the study.

This SAP is based upon the following study documents:

- Study Protocol, Version 4 (December 15, 2016)
- electronic Case Report Form (eCRF), Version 3.0 (October 6, 2016)
- Protocol Deviation Specification Form, Version 1.0 (September 29, 2016)

2 STUDY OBJECTIVES

2.1 Primary Objective and Endpoint

The primary objective of this study is to assess the dose-response relationship between oral vadadustat once daily dosing for 6 weeks and the change in Hb in Japanese subjects with anemia secondary to NDD-CKD; this is to define the starting dose for use in Phase 3 clinical studies in Japan.
The primary endpoint that will be used to assess this objective is the mean change in Hb levels from pre-treatment to the end of the primary efficacy period (Week 6). Pre-treatment Hb is defined as the average of 2 Hb values obtained prior to treatment based on the qualifying screening Hb value and the Hb value at the baseline visit.

2.2 Secondary Objectives and Endpoints

The secondary objectives of this study are:

- To assess the safety, tolerability, pharmacokinetics (PK), and pharmacodynamics (PD) of oral vadadustat once daily dosing in Japanese subjects with anemia secondary to NDD-CKD during the 6-week, primary efficacy period
- To evaluate the effect of dose adjustments and demonstrate the maintenance effect on Hb during a 10-week dose-adjustment and maintenance period
- To assess the time to reach the target Hb range from baseline

The efficacy endpoints that will be used to assess these objectives include the following:

- Time to reach target Hb level of 10.0-12.0 g/dL from baseline visit (Day 1)
- Mean Hb levels at the end of the primary efficacy period (Week 6) and at the end of the dose adjustment and maintenance period (Week 16)
- Proportion of subjects who achieve target Hb 10.0-12.0 g/dL at the end of the dose adjustment and maintenance period (Week 16)
- Mean change in Hb between pre-treatment and the end of the dose adjustment and maintenance period (Week 16)
- Mean change in hematocrit, red blood cell (RBC) count, and reticulocyte count from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Mean change in iron indices (ie, iron, total iron-binding capacity [TIBC], transferrin saturation [TSAT], and ferritin) and hepcidin from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Proportion of subjects requiring rescue with RBC transfusion from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Proportion of subjects requiring rescue with ESAs from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Number of dose adjustments from baseline to the end of the dose adjustment and maintenance period (Week 16)
- Maintenance of iron sufficiency (defined as ferritin ≥50 ng/mL and TSAT ≥20%) from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- Plasma concentration profile of vadadustat and its metabolites using pre-dose sample from Week 4

The safety endpoints that will be used to assess these objectives include the following:

- Safety assessments, including adverse events (AEs), vital signs, electrocardiograms (ECGs), and other laboratory assay results (eg, chemistry, components of the complete blood count [CBC] other than the ones noted above, and vascular endothelial growth factor [VEGF])

3 INVESTIGATIONAL PLAN

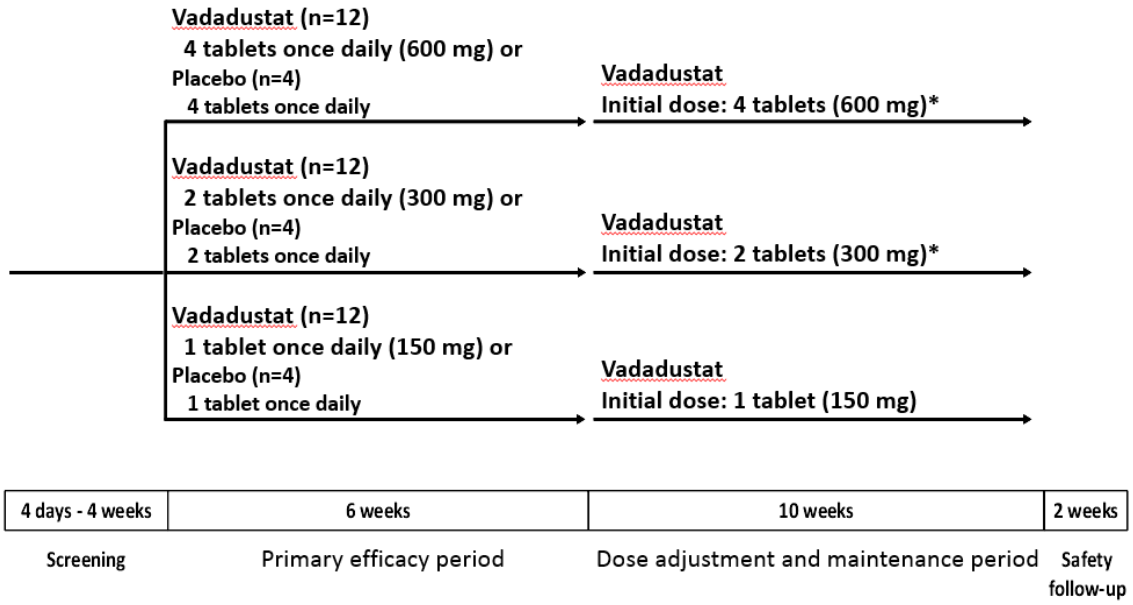
3.1 Overall Study Design and Plan

This is a Phase 2, randomized, double-blind, placebo-controlled, dose-finding study to assess the efficacy, safety, tolerability, PK, and PD of orally administered vadadustat in Japanese subjects with anemia secondary to NDD-CKD.

The study has a planned enrollment of 48 subjects to be enrolled at approximately 25 sites in Japan. There will be 16 subjects in each of the 3 tablet-count groups.

An overview of the study design is presented in Figure 1.

Figure 1: Overview of Study Design



* For subjects who develop an excess Hb response during the primary efficacy period, the number of tablets of study drug will be decreased (see Section 8.2.4). For these subjects, the number of tablets of vadadustat initiated at the Week 6 visit will be lower than indicated.

The study will include the following periods:

- Eligibility screening period (up to 4 weeks)
- Primary efficacy period (6 weeks; Weeks 1 to 6)
- Dose adjustment and maintenance period (10 weeks; Weeks 7 to 16)
- Follow-up period (2 weeks; Weeks 17 and 18)

Subjects will participate in a screening period (4 days to 4 weeks) to determine study eligibility, and eligible subjects will be randomized following the screening period.

Using a central randomization system, subjects will be randomized 1:1:1 to receive 1, 2, or 4 tablets at their baseline visit (Day 1). Within each tablet-count group, subjects will be randomized 3:1 to receive vadadustat (150, 300, or 600 mg vadadustat) or placebo. See the protocol section 8.2.2 for information regarding the randomization scheme.

Study drug treatment will be administered during a 6-week primary efficacy period. See the protocol section 8.2.4 for information on study drug administration. The primary efficacy period includes fixed-dose treatment to establish a dose-response relationship. However, if Hb level increase rapidly or if the Hb level exceeds 13.0 g/dL, the study drug dose can be decreased or interrupted (see the protocol section 8.2.4).

After completing the primary efficacy period, subjects will continue to a 10-week dose adjustment and maintenance period (see the protocol section 8.2.5). Subjects receiving placebo will be switched to vadadustat, and study drug dose will be adjusted to achieve a target Hb of 10.0-12.0 g/dL based on dose adjustment guidelines (see the protocol section 8.2.5).

Vadadustat treatment will stop after the dose adjustment and maintenance period has been completed (Week 16) and subjects will continue in a 2-week follow-up period (Week 17-18).
The clinical and safety assessments will be performed as described in the protocol section 9.3 and as listed in the protocol Appendix A.

3.2 Efficacy and Safety Variables

3.2.1 Efficacy and PD Variables

The following variable is the primary efficacy variable:

- Change from pre-treatment in Hb at Week 6.

The following efficacy endpoints will also be analyzed:

- Actual values and change (absolute and percent) from pre-treatment in Hb, hematocrit, RBC count, and reticulocyte count (both absolute and percent)
- Actual values and change (absolute and percent) from pre-treatment in iron, TIBC, TSAT, ferritin, and hepcidin

3.2.2 Safety Variables

The following variables are the safety endpoints:

- AEs
- Vital signs
- ECGs
- Laboratory parameters

3.2.3 PK Variables

The following variables are the PK variable:

- Pre-dose plasma concentrations of vadadustat at the Week 4 visit
- Pre-dose plasma concentrations of vadadustat's metabolites at the Week 4 visit

4 STATISTICAL METHODS

4.1 Data Quality Assurance

All tables, figures and data listings to be included in the report will be independently checked for consistency, integrity and in accordance with standard PAREXEL procedures.

4.2 General Presentation Considerations

'Pre-treatment' of Hb is defined as the average of 2 Hb values obtained prior to treatment based on the qualifying screening Hb value and the Hb value at the Baseline Visit. 'Pre-

treatment' for other variables is defined as the last available pre-treatment assessment. In general, this is the value at the Baseline Visit. 'End of Treatment (EOT)' is defined as the assessment on the EOT form labeled as 'Week 16 Visit', even if other values closer to Day 113 are available. 'Treatment Day' will be calculated relative to start of dosing at Baseline Visit (i.e. Treatment Day = Assessment Date - Baseline Visit Date + 1).

Continuous data will be summarized in terms of the mean, standard deviation (SD), median, minimum, maximum and number of observations, unless otherwise stated. The minimum and maximum will be reported to the same number of decimal places as the raw data recorded in the database. The mean and median will be reported to one more decimal place than the raw data recorded in the database. The SD will be reported to two more decimal places than the raw data recorded in the database. In general, the maximum number of decimal places reported shall be four for any summary statistic.

Categorical data will be summarized in terms of the number of subjects providing data at the relevant time point (n), frequency counts and percentages. Any planned collapsing of categories will be detailed in the SAP text and the data displays.

Percentages will be presented to one decimal place. Percentages will not be presented for zero counts. Percentages will be calculated using n as the denominator. If sample sizes are small, the data displays will show the percentages, but any textual report will describe frequencies only.

Changes from baseline in categorical data will be summarized using shift tables where appropriate.

P-values greater than or equal to 0.001, in general, will be presented to three decimal places. P-values less than 0.001 will be presented as "<0.001".

Confidence intervals will be presented to one more decimal place than the raw data.

All report outputs will be produced using SAS® version 9.3 or a later version in a secure and validated environment. All report outputs will be provided to the Sponsor in a single Microsoft Word 2010 [Version 14.0] document.

4.3 Study Subjects

4.3.1 Disposition of Subjects

A clear accounting of the disposition of all subjects who enter the study will be provided, from screening to study completion.

The summary of the following subject disposition will be presented.

- A summary of the number of subjects randomized, the number and percentage of subjects treated (with at least one dose of study medication) and the number and percentage of subjects completed through Week 6 Visit and discontinued by Week 6 Visit by treatment group and overall (Analysis population: All Subjects

Randomized). For final analysis, a summary of the number and percentage of subjects completed Week 18 Visit and discontinued from the study by treatment group and overall will be provided (Analysis population: All Subjects Randomized). Withdrawals from the study will also be summarized by major reason.

By-subject listings of eligibility details, randomization details, visit dates and withdrawal/study completion details (including reason for discontinuation and duration of treatment prior to discontinuation) will be provided.

4.3.2 Protocol Deviations

Major protocol deviations are defined as those deviations from the protocol likely to have an impact on the perceived efficacy and/or safety of study treatments. The impact of major protocol deviations on the efficacy and/or safety results will be evaluated prior to unblinding to determine which data and/or subjects, if any, to exclude from the analysis populations (see Section 4.4),

Protocol deviations and any action to be taken regarding the exclusion of subjects or affected data from specific analyses will be determined in Protocol Deviation Specification document 20170207 version 2.0.

The protocol deviation will be summarized as following:

- A summary of the number and percentage of subjects with a protocol deviation by treatment group and overall and by type of deviation and by period (Analysis population: All Subjects Randomized)

A by-subject listing of protocol deviations will be provided.

4.4 Analysis Populations

Upon database release, protocol deviation and analysis population outputs will be produced and will be sent to Sponsor for review. An analysis population classification meeting will be arranged to discuss the outputs and to decide which subjects and/or subject data will be excluded from certain analyses. Decisions made regarding the exclusion of subjects and/or subject data from analyses will be made prior to unblinding and will be documented and approved by Sponsor.

The following summary of analysis population will be provided:

- A summary of the number and percentage of subjects allocated to each analysis population by treatment group and overall (Analysis population: All Subjects Randomized)

A by-subject listing of analysis population details will be provided. This listing will be presented by treatment group and include: site, subject identifier, inclusion/exclusion flag

for each population and reason for exclusion from each population. All subjects randomized will appear on this listing.

4.4.1 Analysis Population for the Safety Analyses

The safety summaries and analyses will be based on the **safety population**. The safety population is defined as all enrolled subjects who receive at least 1 dose of study medication. In the event that a subject is allocated the incorrect study treatment as per the study randomization list, subjects will be summarized and analyzed 'as treated' i.e. by allocated treatment group.

4.4.2 Analysis Populations for the Efficacy Analyses

The efficacy summaries and analyses will be based on the **modified intent-to-treat (MITT) population**. The MITT population is defined as all randomized subjects who have received at least 1 dose of study medication and have a pre-treatment Hb average defined as the average of the qualifying screening value and the baseline value, and at least one post-baseline Hb measurement. In the event that a subject is allocated the incorrect study treatment as per the study randomization list, subjects will be summarized and analyzed 'as randomized' i.e. by randomized treatment group.

For the efficacy variables, sensitivity analyses will be performed on the **per-protocol (PP) population** to assess the robustness of the study conclusions to the choice of analysis population. The PP population is defined as the subjects in the MITT population who have completed the study and have efficacy data through Week 6, have a study medication compliance of ≥80%, and do not have any major protocol deviations expected to significantly affect the primary efficacy endpoint. Subjects with major protocol deviations expected to significantly affect the primary efficacy endpoint will be identified and documented prior to data unblinding.

4.4.3 Analysis Population for the PK Analysis

The PK summaries and analyses will be based on the **pharmacokinetics (PK) population**. The PK population is defined as all subjects in the safety population who have a pre-dose PK sample at Week 4. Subjects will be summarized and analyzed by actual treatment group.

4.5 Demographic and Other Baseline Characteristics

The demographic and other baseline characteristics will be summarized for the MITT and safety populations. The following summaries will be provided:

- A summary of demographic variables (e.g. age, gender, weight, height and body mass index [BMI]) by treatment group and all subjects

- A summary of other relevant variables (e.g. estimated Glomerular Filtration Rate [eGFR] at baseline, CKD stage, etiology of CKD and smoking status) by treatment group and all subjects
- A summary of relevant medical history by treatment group and all subjects.
- A summary of prior and concomitant medications within 30 days prior to the first study drug dose up to and including Week 6 by treatment group and overall.
- For the final analysis, a summary of concomitant medications after Week 6 will be provided.

Age will be calculated as the number of complete years between a subject's birth date and the date of informed consent.

Definitions of eGFR and CKD stage are described in section 4.8.4.7.

CKD stage will be determined by eGFR at baseline:

CKD stage	eGFR (mL/min/1.73m^2)
1	90+
2	60 - 89
3a	45 - 59
3b	30 - 44
4	15 - 29
5	< 15

By-subject listings of demographic data and other baseline characteristics (as summarized above) will be provided

4.6 Treatment Compliance

The treatment compliance will be summarized for the MITT and safety population. The following summary will be provided.

- A summary of the treatment compliance measures (i.e. ≥80% compliance and <80%) by treatment group and study visit.

A by-subject listing of treatment compliance data will be provided.

4.7 Efficacy Evaluation

4.7.1 Analysis and Data Conventions

This study is designed to test for dose-response relationship. An analysis of covariance (ANCOVA) model will be used to compare absolute change from pre-treatment in Hb between the three vadadustat dosing groups and the placebo group. The model will include treatment assignment (3 dosed groups and one placebo group) and pre-treatment Hb value as a covariate. A step-down procedure will be used to control the overall type I error rate for the multiple comparisons. Testing of the highest dose compared with

placebo will be conducted first. If and only if this comparison is significant, then testing will proceed to comparison of the next lower dose and placebo, and so on. Therefore, no multiplicity adjustment will be needed for this analysis.

A linear regression dose-response modeling will be performed as a secondary analysis.

$$Y = \beta_0 + \beta_{dose} X$$

where Y is change in Hb and X is vadadustat dose. β_xs are parameters of the model. β_0 is intercept and β_{dose} is slope, respectively.
The null hypothesis for the dose response will be that the regression coefficient of slope parameter is zero. The alternative hypothesis will be that the regression coefficient is not zero. Symbolically, this is expressed as follows:

$$H_0: \beta_{dose} = 0$$
$$H_1: \beta_{dose} \neq 0$$

This hypothesis will be tested using a 0.05 two-sided significance level.

4.7.1.1 Multi-center Studies
Not Applicable. Analysis of center effect is not planned.

4.7.1.2 Adjustments for Covariates
The primary efficacy analysis will be adjusted for the pre-treatment measure of primary efficacy variable.

An 'unadjusted' sensitivity analysis will be performed to assess the robustness of the study conclusion.

4.7.1.3 Handling of Missing Data
For the MITT population analyses for the primary efficacy variable, the Last Observation Carried Forward (LOCF) approach will be used to impute missing data.
Subjects who underwent dose reduction before week 6 will not
be analyzed differently from subjects who did not undergo dose reduction. This means we will use the actual Hb at week 6 for all subjects except the missing data, which will be carried from the last Hb before week 6 and after baseline.

- If a Baseline value is missing, the last value prior to baseline will be carried forward to the Baseline value but not to further values during the treatment period.
- For post-Baseline assessments, hemoglobin measurements at end of treatment and follow-up visits are not carried forward. Other than that, the most recent post-baseline assessments to the primary evaluation period (week 6) will be carried forward . Baseline and pre-Baseline values will not be carried forward to the post-Baseline visits.

For the secondary endpoint analyses, data will be used as observed and no imputation method will be used.

4.7.1.4 Multiple Comparisons/Multiplicity

Analysis of one primary variable [Change in Hb] and one time point of primary interest [Week 6 visit] has been defined for this study. Though 3 pairwise comparison between placebo and dosed group is planned, step-down procedure will be used to control the overall type I error rate. The secondary variables defined are intended to provide supportive evidence relating to the primary objective and no labeling claims are intended. Though one preliminary analysis is planned before the final analysis, primary analysis will not be changed. Hence no adjustments for multiplicity are required.

4.7.1.5 Preliminary (6-Week) Analysis

After all subject performed assessment of the Week 6 Visit, preliminary analysis will be performed. As the study conduct and final analysis will not be modified by this analysis, no alpha adjustment is proposed. It is expected that this 6-week data
will be identical for the preliminary analysis and the final analysis of the complete dataset. Any changes between the preliminary analysis and the final analysis will be documented. The preliminary analysis will be performed by the independent unblinded team. All data through the cutoff date of the last subject randomized who completes week 6 will be analyzed. Blinding of blinded team and investigators will be maintained to prevent potential bias.

4.7.1.6 Examination of Subgroups

Not Applicable. No subgroup analysis is planned.

4.7.2 Primary Efficacy Variable – Mean Change in Hb from Pre-treatment to the End of the Primary Efficacy Period (Week 6)

The primary variable for the assessment of efficacy is the change from pre-treatment to Week 6 in Hb (g/dL). Change in Hb is defined as the Hb measured at the Week 6 visit minus the mean pre-treatment Hb. Pre-treatment Hb is defined as the average of 2 Hb values obtained prior to treatment based on the qualifying screening Hb value and the Hb value at the baseline visit.

For the primary analysis, the LOCF approach will be used to impute missing data. Complete-case analysis will be performed for sensitivity at the scheduled Week 6 visit.

4.7.2.1 Summary of actual value and change in Hb at week 6

The primary analysis population for efficacy will be MITT and PP population. The following summary will be provided:

- A summary of actual value of Hb by treatment group at baseline and week 6. A summary of the change (absolute and percent) from baseline in Hb by treatment group at week 6. 1st and 3rd quartiles (Q1, Q3) as well as the mean, SD, median, minimum, maximum and number of observations will be shown. P values for overall

comparison and treated groups vs. placebo on change from baseline will also be presented.
- A waterfall plot for absolute Hb change from baseline for each treatment group at week 6.

The effect of treatment in terms of the change from pre-treatment to Week 6 in Hb will be analyzed using a one-way analysis of covariance (ANCOVA). The statistical model will assess the main effect of treatment, adjusted for pre-treatment average. Least-squares (LS) means for each treatment and their 95% confidence intervals (CIs) will be presented. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done. Unadjusted one-way analysis of variance (ANOVA) will be performed for sensitivity.

The simple linear regression between the change from pre-treatment to Week 6 in Hb and dose will be calculated. The regression coefficients and their standard errors, p-values of test of regression parameters and their 95%CIs will be presented. The test of slope parameter will be used to evaluate dose response relationship.

As sensitivity analyses, efficacy endpoints will also be analyzed using the PP population.

A by-subject listing of actual values and change (absolute and percent) from baseline in Hb will be provided.

4.7.2.2 Summary of actual value and change in Hb from baseline to week 6 and beyond

The analysis population for efficacy will be MITT and PP population. The following summary will be provided:
- A summary of actual value of Hb by treatment group and study visits. 1st and 3rd quartiles (Q1, Q3) as well as the mean, SD, median, minimum, maximum and number of observations will be shown. Summary of pre-treatment average will also be presented.
- A summary of the change (absolute and percent) from baseline in Hb by treatment group and study visits.
- A plot showing the mean Hb value over time within each treatment group. 95% CI of these means will be shown as error bar and sample size in each group at each time point will be shown.
- Plots of individual profile of Hb values over time within each treatment group.
- Box plots of absolute Hb changes from baseline for each treatment group at each post-treatment study visit to week 6
- Box plots of absolute Hb changes from baseline for each treatment group at each post-treatment study visit to week 16

A by-subject listing of actual values and change (absolute and percent) from baseline in Hb will be provided.

4.7.3 Secondary Efficacy Variables

The analysis population for efficacy will be MITT and PP population.

Time to reach target Hb Range

Time to reach target Hb level of 10.0-12.0 g/dL from baseline is defined for subjects with Hb <10.0 g/dL at the baseline visit as following:

First post-treatment assessment date with value of Hb ≥10.0 and ≤12.0 g/dL – the baseline visit date +1. If target Hb level is not achieved, last Hb assessment date will be used as censored date.

A summary of time to reach target Hb level will be provided for subjects with Hb <10.0 g/dL at the baseline visit by treatment group.

A by-subject listing will be provided.

Hb within target Range

A summary of the number and percentage of subjects within target Hb 10-12 g/dL at the end of the dose adjustment and maintenance period (Week 16) will be provided by treatment group. In addition, the cumulative proportion of subjects within target range will be displayed graphically from baseline visit to follow up visit by treatment group.

The number and percentage of subjects above / within / below target range who are maintained within the target range at their previous visit after Week 6 will be provided by treatment group.

The effect of treatment in terms of the proportion of subjects within target Hb level at Week 6 will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

Increase in Hb from pre-treatment average

A summary of the number and percentage of subjects who achieved a ≥1 g/dL increase in Hb from pre-treatment average will be provided by treatment group.

The effect of treatment in terms of the proportion of subjects who achieved a ≥1 g/dL increase in Hb from pre-treatment average will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

Hematocrit, RBC count, and reticulocyte count

Hematocrit, RBC count, and reticulocyte count (both absolute and percent) will be analyzed as secondary efficacy variables.

The following summaries will be provided:

- A summary of hematocrit, RBC count, and reticulocyte count (both absolute and percent) by treatment group and time point. 1st and 3rd quartiles (Q1, Q3) as well as the mean, SD, median, minimum, maximum and number of observations will be presented.
- A summary of the change (absolute and percent) from baseline in hematocrit, RBC count, and reticulocyte count (both absolute and percent) by treatment group and time point
- Box plots of absolute changes from baseline in hematocrit, RBC count, and reticulocyte count (both absolute and percent) for each treatment group at each post-treatment time point

The effect of treatment in terms of the change from baseline to Week 6 in hematocrit and RBC count and reticulocyte count (both absolute and percent) will be analyzed using ANCOVA. The statistical model will assess the main effect of treatment, adjusted for baseline measurement. LS-means for each treatment and their 95% CIs will be presented. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done. Unadjusted ANOVA will be performed for sensitivity.

A by-subject listing of hematocrit, RBC count, and reticulocyte count (both absolute and percent) will be provided by treatment group.

Iron indices and hepcidin
Iron indices (ie, iron, TIBC, TSAT, and ferritin) and hepcidin will be analyzed as secondary efficacy variables.

The following summaries will be provided:
- A summary of iron indices and hepcidin by treatment group and time point
- A summary of the change (absolute and percent) from baseline in iron indices and hepcidin by treatment group and time point
- Box plots of absolute changes from baseline in iron indices and hepcidin for each treatment group at each post-treatment time point

A by-subject listing of iron indices and hepcidin will be provided by treatment group.

Rescue therapy & ESA administration
Initiation of rescue therapy will be analyzed.

The following Rescue therapy summaries will be provided:
- A summary of the number and percentage of subjects requiring rescue with RBC transfusion from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- A summary of the number and percentage of subjects requiring rescue with ESAs from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16). An ESA rescue is defined to fulfill the following 4 criteria simultaneously:

1. ESA administration, and
2. ESA administered because the subject experienced a clinically significant worsening of their anemia or symptoms of anemia, and
3. ESA administered because the subject's Hb level is <9.0 g/dL, and
4. Subjects' reason for early study withdrawal is because of "Worsening of anemia requiring ESA rescue or blood transfusion".

The following ESA administration summaries will be provided:

- A summary of the number and percentage of subjects requiring administration with ESAs from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)

The effect of treatment in terms of the proportion of subjects requiring with RBC transfusion / ESAs administration will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

By-subject listings of rescue therapies and ESA administration will be provided by treatment group.

Average Daily Dose
A summary of Average Daily Dose by treatment group and visit will be presented with 1st and 3rd quartiles (Q1, Q3) as well as the mean, SD, median, minimum, maximum and number of observations.

Dose adjustment
A overview summary of the number of dose adjustments from baseline to the end of the dose adjustment and maintenance period (Week 16) will be provided.
A summary of the number of dose adjustments by visit from baseline to the end of the dose adjustment and maintenance period (Week 16) will be provided.

A by-subject listing of dose adjustment with HB change will be provided by treatment group. The three flags of HB change will be presented as follows:

- HB>13 g/dL at any time of study
- HB >12 g/dL to 13 g/dL at dose adjustment and maintenance period only
- HB increase >1 g/dL over 2 weeks at any time of study.

Maintenance of iron sufficiency
A summary of maintenance of iron sufficiency (defined as ferritin ≥50 ng/mL and TSAT ≥20%) from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16) will be provided.

The effect of treatment in terms of the proportion of subjects who maintain iron sufficiency will be analyzed using Fisher's exact test for week 6 only. Three post-hoc

pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

A by-subject listing of iron sufficiency will be provided by treatment group.

4.7.4 Exploratory Efficacy Variable

Time in Therapeutic Range (TTR) for Hb from week 6 to week 20

The half-time interpolation method will be used to calculate TTR for Hb. The total time of follow-up with Hb in target range of 10.0 to 12.0 g/dL is divided by the total time. Half the time between two tests is allocated to the first Hb value, and half to the second Hb value.

A summary of TTR for Hb will be provided by treatment group.

A by-subject listing of TTR for Hb will be provided by treatment group

4.8 Safety Evaluation

All safety summaries and analyses will be based upon the safety population as defined in Section 4.4.

4.8.1 Extent of Exposure

The extent of exposure will be summarized for the safety population. The following summary will be provided.

- A summary of the treatment duration (e.g. End Date of Administration - Start date of Administration +1) by treatment group and study visits.

A by-subject listing of start, end and duration of treatment will be provided.

4.8.2 Adverse Events

AEs will be coded using the Medical Dictionary for Regulatory Activities (MedDRA) Version 19.0.

Treatment-emergent adverse events (TEAEs) will be tabulated and are defined as those AEs that either start or worsen in severity on or after the date/time of first dose of study treatment and on or before the final protocol required visit. In general, it is Week 18 Visit and 14 days after the date of last dose of study treatment. The TEAEs are defined as those adverse events that started after first injection up to 14 days post-last dose.

Where dates are missing or partially missing, AEs will be assumed to be treatment-emergent, unless there is clear evidence (through comparison of partial dates) to suggest

that the AE started prior to the first dose of study treatment or more than 14 days after the last dose of study treatment.

Partial AE and concomitant medication start dates will be imputed as follows:

1. If the year (and month and day) is unknown, the date will be imputed as the date of first intake of study treatment if this is possible (i.e., the event end date is not prior to treatment start date).

2. If the month and day is unknown then:

    a. If the year matches the first dose date, then impute the month and day of the first dose date.

    b. If the year is earlier than the first dose date or the first dose date is not available (subject is not dosed) and matches the year of informed consent date, then impute the month and day of the informed consent date;

    c. Otherwise, assign January 1.

3. If the day is unknown then:

    a. If the month and year match the first dose date, then impute the day of the first dose date.

    b. If the month and year are earlier than the first dose date or the first dose date is not available (subject is not dosed) and matches the month and year of the informed consent date, then impute the day of the informed consent date;

    c.Otherwise, assign '1'.

Partial AE stop dates will be imputed as follows:

1. If the year is unknown, the date will not be imputed and will be assigned a missing value.

2. If the month is unknown, then assign December.

3. If the day is unknown, then assign the last day of the month.

For all other settings (eg. year missing, but day and month available etc.), imputation strategies will be reviewed and decided in the ongoing data cleaning and sponsor meetings.

If the start and/or stop dates of medications other than study treatments are missing or partially missing, the dates will be compared as far as possible with the date of first study treatment administration.

If not specified, observed data approach will be performed.

The AE will be summarized using MedDRA system organ class (SOC) and preferred term (PT). All AE summaries will provide the number of subjects reporting at least one AE and the total number of events reported. The following summaries will be provided:

- A summary of the number and percentage of subjects reporting at least one TEAE, drug-related TEAE, TEAE leading to withdrawal, serious adverse event (SAE), drug-related SAE, SAE leading to withdrawal and death by treatment group.
- A summary of the number and percentage of subjects reporting a TEAE by treatment group, SOC, and PT
- A summary of the number and percentage of subjects reporting a drug-related TEAE by treatment group, SOC, and PT
- A summary of the number and percentage of subjects reporting a treatment-emergent SAE by treatment group, SOC and PT
- A summary of the number and percentage of subjects reporting a drug-related treatment-emergent SAE by treatment group, SOC, and PT
- A summary of the number and percentage of subjects reporting a drug-related TEAE leading to withdrawal by treatment group, SOC, and PT
- A summary of the number and percentage of subjects reporting a TEAE leading to withdrawal by treatment group, SOC, and PT
- A summary of the most common TEAEs by treatment group and PT (reported by >10% of subjects in any treatment group).
- A summary of the number and percentage of subjects reporting a TEAE by treatment group, severity, SOC and PT
- A summary of the number and percentage of subjects reporting a TEAE by treatment group, relatationship, SOC and PT

AE summaries will be ordered alphabetically for SOC, and PT within SOC.

For each subject and each AE, the worst severity recorded will be attributed and used in the by-severity summaries. Similarly, the worst causality (most related to treatment) will be attributed and used in the by-causality summaries. If severity or causality is missing, data will be treated as observed

A by-subject listing of all AEs (including non-treatment-emergent events) will be provided. This listing will be presented by treatment group and will include: center, subject identifier, age, sex, race, AE (SOC, PT, and verbatim term), date of onset, date of resolution, duration, severity, seriousness, action taken, outcome and causality.

The following AEs are defined as AEs of special interest. Summaries of the number and percentage of subjects reporting the following AEs will be provided.

- PT: “hyperkalemia” (MedDRA 10020646) or “blood potassium increased” (MedDRA 10005725)
- PT: “hyperuricaemia” (MedDRA 10020903) or “blood uric acid level increased” (MedDRA 10022891)
- PT: “Hypertension” (MedDRA 10020772) or “Blood pressure increased” (MedDRA 10005750) or “Blood pressure systolic increased” (MedDRA 10005760) or “Blood pressure diastolic increased” (MedDRA 10005739) or “Blood pressure ambulatory increased” (MedDRA 10005732) or “Blood pressure inadequately controlled” (MedDRA 10051128) or “Accelerated hypertension” (MedDRA 10000358) or “Essential hypertension” (MedDRA 10015488) or “Diastolic hypertension”

(MedDRA 10012758) or "Systolic hypertension" (MedDRA 10042957) or "Malignant hypertension" (MedDRA 10025600)
- Standardised MedDRA Query (SMQ): "Cholestasis and jaundice of hepatic origin" (SMQ 20000009) or "Drug related hepatic disorders – severe events only" (SMQ 20000007) or "Liver related investigations, signs and symptoms" (SMQ 20000008)
- Standardised MedDRA Query (SMQ): "Ischaemic heart disease" (SMQ 20000043)
- Standardised MedDRA Query (SMQ): "Central nervous system haemorrhages and cerebrovascular conditions" (SMQ 20000061)
- Standardised MedDRA Query (SMQ): "Embolic and thrombotic events" (SMQ 20000081)
- Standardised MedDRA Query (SMQ): "Cardiac failure" (SMQ 20000004)
- Standardised MedDRA Query (SMQ): "Retinal disorders" (SMQ 20000158, narrow)

Serious adverse events that occur during the screening period and are assessed by the investigator as related to study procedures will be tabulated.

4.8.3 Deaths, Serious Adverse Events, and Other Significant Adverse Events

Other significant AEs are those AEs reported as leading to withdrawal from study. The summaries of SAEs and AEs leading to withdrawal from study are described in section 4.8.2.

A summary of the number and percentage of deaths during the study will be provided by treatment group

A summary of the number and percentage of subjects Adverse Events Leading to Death by treatment group, severity, SOC and PT

A by-subject listing of all deaths that occurred during the study will be provided.

4.8.4 Clinical Laboratory Evaluation

4.8.4.1 Liver function abnormality

A summary of Liver function abnormality by visit will be provided by treatment group.
Any subject with at least one of the following liver function abnormalities is summarized:

- ALT ≥2x and <3x ULN
- AST ≥2x and <3x ULN
- Bilirubin ≥2x and <3x ULN
- ALT ≥3x ULN
- AST ≥3x ULN
- Bilirubin ≥3x ULN

4.8.4.2 Serum and urine pregnancy tests

Serum pregnancy test for females of childbearing potential will be performed at the screening visit and will be used for subject's inclusion / exclusion. Urine pregnancy test will be performed at the baseline visit and used for subject's initiation of study drug.

Additional pregnancy tests may be conducted during the study to establish the absence of pregnancy based on the investigator's clinical judgment or as required by local regulations.

A by-subject listing of serum and urine pregnancy tests will be provided.

4.8.4.3 Coagulation tests

Coagulation tests will be performed at the baseline visit. The following variables will be analyzed.

- prothrombin time
- partial thromboplastin time
- international normalized ratio

A summary of baseline coagulation test results will be provided by treatment group.

A by-subject listing of coagulation tests will be provided.

4.8.4.4 Folate and vitamin B12

Folate and vitamin B12 tests will be performed at the screening visit and will be used for subject's inclusion / exclusion.

A by-subject listing of folate and vitamin B12 tests will be provided.

4.8.4.5 Urine albumin-to-creatinine ratio (uACR)

Urine albumin-to-creatinine ratio (uACR) will be evaluated at baseline visit.

A by-subject listing of uACR will be provided.

4.8.4.6 Complete Blood Count (CBC)

The following components of the CBC are described in Efficacy / PD analysis section.

- Hb
- Hematocrit
- RBC count
- Automated Reticulocyte Count (both absolute and percent).

The following components of the CBC will be analyzed:

- Mean Corpuscular Volume
- Mean Corpuscular Hb
- Mean Corpuscular Hb Concentration
- Red Cell Distribution Width
- White Blood Cell count with differential (neutrophils, bands, lymphocytes, monocytes, eosinophils, and basophils)
- Platelets

The following summaries will be provided:

- A summary of each component of the CBC by treatment group and time point
- A summary of the change from baseline in each component of the CBC by treatment group and time point
- A summary of the number and percentage of subjects experiencing abnormal values by component of CBC, treatment group and time point.

A by-subject listing of CBC will be provided by treatment group.

4.8.4.7 Chemistry and estimated glomerular filtration rate (eGFR)

The following laboratory parameters will be analyzed:

- Sodium
- Potassium
- Bicarbonate
- Chloride
- Calcium
- Phosphorus
- Glucose
- Creatinine
- Blood Urea Nitrogen
- Creatine Phosphokinase
- Uric Acid
- Albumin
- Total Protein
- Total Bilirubin
- Alkaline Phosphatase
- Alanine Aminotransferase (ALT)
- Aspartate Aminotransferase (AST)
- Lactate Dehydrogenase
- Total cholesterol
- Estimated Glomerular Filtration Rate (eGFR) (calculated from serum creatinine)

The following summaries will be provided:

- A summary of Chemistry and eGFR by treatment group and time point
- A summary of the change (absolute and percent) from baseline in Chemistry and eGFR by treatment group and time point
- A summary of the number and percentage of subjects experiencing abnormal values by laboratory parameter, treatment group and time point

The eGFR will be calculated from serum creatinine by following formula:

$$\text{eGFR}(\text{mL/min}/1.73\text{m}^2) = 194 \times (\text{Scr in mg/dL})^{-1.094} \times (\text{Age})^{-0.287} \times (0.739 \text{ if female})$$

A by-subject listing of Chemistry and eGFR will be provided by treatment group.

4.8.4.8 C-reactive protein and Vascular Endothelial Growth Factor (VEGF)
The following summaries will be provided:

- A summary of C-reactive protein (CRP) and VEGF by treatment group and time point
- A summary of the change (absolute and percent) from baseline in CRP and VEGF by treatment group and time point

The effect of treatment in terms of the change from baseline to Week 6 in CRP and VEGF will be analyzed using ANCOVA. The statistical model will assess the main effect of treatment, adjusted for baseline measurement. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done. Unadjusted ANOVA will be performed for sensitivity.

A by-subject listing of CRP and VEGF will be provided by treatment group.

4.8.5 Vital Signs, Physical Findings and Other Observations Related to Safety

4.8.5.1 Vital Signs
The following variables will be analyzed:

- Blood Pressure (Systolic / Diastolic)
- Heart Rate
- Respiratory Rate
- Body Temperature

The following summaries will be provided:

- A summary of each vital sign parameter by treatment group and time point.
- A summary of the change (absolute and percent) from baseline in each vital sign parameter by treatment group and time point.

Any findings of treatment-emergent vital sign abnormalities will be handled as adverse event and reported in AE tables / listings.

A plot showing the mean blood pressure over time within each treatment group. 95% CI of these means will be shown as error bar and sample size in each group at each time point will be shown.

A by-subject listing of vital sign parameter with reference ranges will be provided.

4.8.5.2 12-lead ECG
The following variables will be analyzed at the Baseline Visit:

- Heart rate
- PR interval
- QT interval
- QRS interval

- QTc (corrected)
- Overall result

A summary of each ECG parameter will be provided by treatment group.

A by-subject listing of 12-lead ECG results will be provided.

4.8.5.3 Physical Examination and Weight Assessment

Physical Examination and Weight Assessment will be performed at the Screening Visit and used for subject's inclusion / exclusion. Weight and height will be reported in demographic tables and listings.

4.8.6 Safety Monitoring (Independent Data Monitoring Committee [IDMC], Data Monitoring Committee [DMC], Data and Safety Monitoring Board [DSMB])

Not applicable. No change of study conduct based on preliminary analysis result is planned.

4.9 Other Analyses

4.9.1 PK Analysis

The following summary will be provided:

- The summary of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.
- Box plots of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.
- Dose-normalized box plots of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.

Concentrations below the limit of quantification (BLQ) will be replaced by the 0 for the calculation of descriptive statistics. So concentration contains BLQ values are treated as 0 for all summary statistics except geometric mean and its related parameters.

By-subject listings of pre-dose plasma concentrations of vadadustat and its metabolites will be provided by treatment group.

4.9.2 Treatment-Emergent Outliers analysis

The purpose of this analysis is to identify the treatment Emergent outliers by each prescribed dose at each period. The definition of prescribed doses is the subjects with latest dose amount at planned visit when dose change. e.g. if at Week 2 with dose 600 mg and dose reduced to 450 mg at week 4, then visit of prescribed dose for VADA 450 was assigned to Week 4.
However, if subject had dose reduction on 150 mg or had interruption of study medication, the prescribed dose will be considered as 0 mg.

Only treatment-emergent outliers will be counted but outliers at the baseline and screening visits will be excluded.

The following components will be analyzed by primary efficacy period and dose adjustment and maintenance period for all subjects and for subjects with non-elevated values at baseline:

- Non-elevated at baseline definition for each component
  - Systolic Blood Pressure <140 mmHg
  - Diastolic Blood Pressure<90 mmHg
  - Heart Rate <100 bpm
  - Potassium<4 mEq/L

The following component with following criteria will be applied.

- Systolic Blood Pressure
  - Any SBP >=160 to <180 mmHg
  - Any SBP >=180 mmHg
  - Any SBP increase from baseline >=20 to <40 mmHg
  - Any SBP increase from baseline >=40 mmHg
- Diastolic Blood Pressure
  - Any DBP >=100 to <110 mmHg
  - Any DBP >=110 mmHg
  - Any DBP increase from baseline >=10 to <20 mmHg
  - Any DBP increase from baseline >=20 mmHg
- Heart Rate
  - Any pulse >=100 to <120 bpm
  - Any pulse >=120 bpm
  - Any pulse increase from baseline >=20 to <30 bpm
  - Any pulse increase from baseline >=30 bpm
- Potassium
  - Any Potassium >5.5 to 6.0 mEq/L
  - Any Potassium >6.0 to 6.5 mEq/L
  - Any Potassium >6.5 mEq/L
  - Any Potassium increase from baseline >0.5 to 1.0
  - Any Potassium increase from baseline >1.0 to 1.5 mEq/L
  - Any Potassium increase from baseline > 1.5 mEq/L

4.10 Determination of Sample Size

The target enrollment will be approximately 48 subjects for the study with 12 subjects enrolled in each of the 4 treatment groups.
The study is powered based on a comparison of the highest dose vadadustat group (600 mg daily) and the placebo group. Based on the results from Study AKB-6548-CI-0005 that was conducted in the United States and evaluated vadadustat in patients with anemia secondary to NDD-CKD, it is assumed that the expected mean Hb changes from

baseline to Week 6 will be approximately 0 mg/dL for the placebo group and 1.35 g/dL for the highest dose vadadustat group (600 mg daily), with a common standard deviation of 0.68 g/dL across treatment groups. With these assumptions, the study with n=12 subjects per group will have >95% power to detect the difference between treatment group and control group, at 2-sided alpha of 0.05.

4.11 Changes in the Conduct of the Study or Planned Analysis

Not Applicable.

5 REFERENCES

Not Applicable.

5.1 Statistical Analysis Plan Amendment 3

5.1.1 Modifications and changes

5.1.1.1 **Rationale for the amendment**

The purpose of this amendment is to reflect the change for the efficacy endpoints and also illustrate more details from analysis perspective.

- The main changes includes:
- Efficacy related:
    - Additional by-subject listing of dose adjustment with HB change with different HB cutoff point at dose adjustment and maintenance period will be provided
- Safety related:
    - Additional description for treatment emergent outlier analysis.
    - Separate ESA administration summaries and ESA rescue summaries will be provided:
- Other:
    - Keep consistent with mock of shell and remove duplicates description in the SAP.

5.1.1.2 Global Changes

None

5.1.1.3 **Specific changes**

Change#1

Section 4.7.1.3 Handling of Missing Data, page 15,

For the MITT population analyses for the primary efficacy variable, the Last Observation Carried Forward (LOCF) approach will be used to impute missing data.

Has been changed to:

For the MITT population analyses for the primary efficacy variable, the Last Observation Carried Forward (LOCF) approach will be used to impute missing data.
Subjects who underwent dose reduction before week 6 will not
be analyzed differently from subjects who did not undergo dose reduction. This means we will use the actual Hb at week 6 for all subjects except the missing data, which will be carried from the last Hb before week 6 and after baseline.

Change#2

Section 4.7.3 Secondary Efficacy Variables, page 18,

A summary of time to reach target Hb level will be provided for subjects with Hb <10.0 g/dL at the baseline visit by treatment group.

Kaplan-Meier failure (cumulative incidence) plot will be presented for the time to reach target Hb level by treatment group.

Has been changed to:

A summary of time to reach target Hb level will be provided for subjects with Hb <10.0 g/dL at the baseline visit by treatment group.

Change#3

Section 4.7.3 Secondary Efficacy Variables, page 18,

Hb within target Range

A summary of the number and percentage of subjects within target Hb 10-12 g/dL at the end of the dose adjustment and maintenance period (Week 16) will be provided by treatment group.

The number and percentage of subjects above / within / below target range who are maintained within the target range at their previous visit after Week 6 will be provided by treatment group.

The effect of treatment in terms of the proportion of subjects within target Hb level at Week 6 will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.
A by subject listing will be provided.

Has been changed to:

Hb within target Range

A summary of the number and percentage of subjects within target Hb 10-12 g/dL at the end of the dose adjustment and maintenance period (Week 16) will be provided by

treatment group. In addition, the cumulative proportion of subjects within target range will be displayed graphically from baseline visit to follow up visit by treatment group.

The number and percentage of subjects above / within / below target range who are maintained within the target range at their previous visit after Week 6 will be provided by treatment group.

The effect of treatment in terms of the proportion of subjects within target Hb level at Week 6 will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

Change#4

Section 4.7.3 Secondary Efficacy Variables, page 18,

Rescue therapy
Initiation of rescue therapy will be analyzed.

The following summaries will be provided:

- A summary of the number and percentage of subjects requiring rescue with RBC transfusion from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- A summary of the number and percentage of subjects requiring rescue with ESAs from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)

The effect of treatment in terms of the proportion of subjects requiring rescue with RBC transfusion / ESAs will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

By-subject listings of rescue therapies will be provided by treatment group.

Has been changed to:

Rescue therapy & ESA administration
Initiation of rescue therapy will be analyzed.

The following Rescue therapy summaries will be provided:

- A summary of the number and percentage of subjects requiring rescue with RBC transfusion from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)
- A summary of the number and percentage of subjects requiring rescue with ESAs from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16). An ESA rescue is defined to fulfill the following 4 criteria simultaneously:

1. ESA administration, and
2. ESA administered because the subject experienced a clinically significant worsening of their anemia or symptoms of anemia, and
3. ESA administered because the subject's Hb level is <9.0 g/dL, and
4. Subjects' reason for early study withdrawal is because of "Worsening of anemia requiring ESA rescue or blood transfusion".

The following ESA administration summaries will be provided:

- A summary of the number and percentage of subjects requiring administration with ESAs from baseline to the end of the primary efficacy period (Week 6) and the end of the dose adjustment and maintenance period (Week 16)

The effect of treatment in terms of the proportion of subjects requiring rescue with RBC transfusion / ESAs will be analyzed using Fisher's exact test. Three post-hoc pairwise comparison, i.e., Placebo vs 150 mg, placebo vs 300 mg, and placebo vs 600 mg, will be done.

By-subject listings of rescue therapies and ESA administration will be provided by treatment group.

Change#5

Section 4.7.3 Secondary Efficacy Variables, page 20,

A by-subject listing of dose adjustment will be provided by treatment group.

Has been changed to:

A by-subject listing of dose adjustment with HB change will be provided by treatment group. The three flags of HB change will be presented as follows:

- HB>13 g/dL at any time of study
- HB >12 g/dL to 13 g/dL at dose adjustment and maintenance period only
- HB increase >1 g/dL over 2 weeks at any time of study.

Change#6

Section 4.8.2 **Adverse Events**, page 21,

Partial AE stop dates and concomitant medication will be imputed as follows:

Has been changed to:

Partial AE stop dates will be imputed as follows:

Change#7

Section 4.8.4.1 Liver function abnormality, page 24,

A summary of Liver function abnormality during the study will be provided by treatment group

Has been changed to:

A summary of Liver function abnormality by visit will be provided by treatment group.

Change#8

Section 4.8.4.5 Urine albumin-to-creatinine ratio (uACR), page 28,

Urine albumin-to-creatinine ratio (uACR) will be evaluated at baseline visit.
A summary of uACR will be provided by treatment group.
A by-subject listing of uACR will be provided.

Has been changed to:

Urine albumin-to-creatinine ratio (uACR) will be evaluated at baseline visit.
A by-subject listing of uACR will be provided.

Change#9

Section 4.8.4.7 Hemoglobin, page 25,

4.8.4.7 Hemoglobin
The following Hb analyses will be performed for safety analysis section:
• A summary of the number and percentage of subjects with any Hb > 12.0 g/dL by treatment group
• A summary of the number and percentage of subjects with any Hb > 13.0 g/dL by treatment group
• A summary of the number and percentage of subjects with any Hb > 14.0 g/dL by treatment group
• A summary of the number and percentage of subjects with Hb increase > 1.0 g/dL in any 2-week interval by treatment group

Has been changed to:

This section has been removed.

Change#10

Section 4.8.4.8 Chemistry and estimated glomerular filtration rate (eGFR), page 26,

The following laboratory parameters will be analyzed:
- Sodium
- Potassium
- Bicarbonate
- Chloride
- Calcium
- Phosphorus

- Glucose
- Creatinine
- Blood Urea Nitrogen
- Creatine Phosphokinase
- Uric Acid
- Albumin
- Total Protein
- Total Bilirubin
- Alkaline Phosphatase
- Alanine Aminotransferase (ALT)
- Aspartate Aminotransferase (AST)
- Lactate Dehydrogenase
- Total cholesterol
- Estimated Glomerular Filtration Rate (eGFR) (calculated from serum creatinine)

The following summaries will be provided:

- A summary of Chemistry and eGFR by treatment group and time point
- A summary of the change (absolute and percent) from baseline in Chemistry and eGFR by treatment group and time point
- A summary of the number and percentage of subjects experiencing abnormal values by laboratory parameter, treatment group and time point

The following treatment-emergent laboratory test abnormalities of interest will be summarized

- Subjects experiencing at least 1 potassium > upper limit of normal
- Subjects experiencing at least 1 uric acid > upper limit of normal
- Subjects experiencing at least 1 ALT or AST > upper limit of normal
- Subjects experiencing at least 1 ALT or AST > 2x upper limit of normal
- Subjects experiencing at least 1 ALT or AST > 3x upper limit of normal

Has been changed to:

The following laboratory parameters will be analyzed:

- Sodium
- Potassium
- Bicarbonate
- Chloride
- Calcium
- Phosphorus
- Glucose
- Creatinine
- Blood Urea Nitrogen
- Creatine Phosphokinase
- Uric Acid
- Albumin
- Total Protein
- Total Bilirubin
- Alkaline Phosphatase
- Alanine Aminotransferase (ALT)

- Aspartate Aminotransferase (AST)
- Lactate Dehydrogenase
- Total cholesterol
- Estimated Glomerular Filtration Rate (eGFR) (calculated from serum creatinine)

The following summaries will be provided:
- A summary of Chemistry and eGFR by treatment group and time point
- A summary of the change (absolute and percent) from baseline in Chemistry and eGFR by treatment group and time point
- A summary of the number and percentage of subjects experiencing abnormal values by laboratory parameter, treatment group and time point

Change#11

Section 4.8.5.1 Vital Signs, page 27,

The following variables will be analyzed:
- Blood Pressure (Systolic / Diastolic)
- Heart Rate
- Respiratory Rate
- Body Temperature

The following summaries will be provided:
- A summary of each vital sign parameter by treatment group and time point.
- A summary of the change (absolute and percent) from baseline in each vital sign parameter by treatment group and time point.

Any findings of treatment-emergent vital sign abnormalities will be handled as adverse event and reported in AE tables / listings.

Has been changed to:

The following variables will be analyzed:
- Blood Pressure (Systolic / Diastolic)
- Heart Rate
- Respiratory Rate
- Body Temperature

The following summaries will be provided:
- A summary of each vital sign parameter by treatment group and time point.
- A summary of the change (absolute and percent) from baseline in each vital sign parameter by treatment group and time point.

Any findings of treatment-emergent vital sign abnormalities will be handled as adverse event and reported in AE tables / listings.

A plot showing the mean blood pressure over time within each treatment group. 95% CI of these means will be shown as error bar and sample size in each group at each time point will be shown

Change#12

Section 4.9.1 PK Analysis, page 27,

The following summary will be provided:

- The summary of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.
- Box plots of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.

Dose-normalized box plots of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.

Has been changed to:

The following summary will be provided:

- The summary of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.
- Box plots of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.

Dose-normalized box plots of pre-dose plasma concentrations of vadadustat and its metabolites at Week 4 Visit by treatment group.

Concentrations below the limit of quantification (BLQ) will be replaced by the 0 for the calculation of descriptive statistics. So concentration contains BLQ values are treated as 0 for all summary statistics except geometric mean and its related parameters.

Change#13

Section 4.9.2 Treatment-Emergent Outliers analysis ,

This is newly added section in SAP amendment v3.0.

Has been changed to:

The purpose of this analysis is to identify the treatment Emergent outliers by each prescribed dose at each period. The definition of prescribed doses is the subjects with latest dose amount at planned visit when dose change. e.g. if at Week 2 with dose 600 mg and dose reduced to 450 mg at week 4, then visit of prescribed dose for VADA 450 was assigned to Week 4.
However, if subject had dose reduction on 150 mg or had interruption of study medication, the prescribed dose will be considered as 0 mg.

Only treatment-emergent outliers will be counted but outliers at the baseline and screening visits will be excluded.

The following components will be analyzed by primary efficacy period and dose adjustment and maintenance period for all subjects and normotensive at baseline:

- Normotensive at baseline definition for each interested component
  - Systolic Blood Pressure <140 mmHg
  - Diastolic Blood Pressure<90 mmHg
  - Heart Rate <100 bpm
  - Potassium<4 mEq/L

The following component with following criteria will be applied.

- Systolic Blood Pressure
  - Any SBP >=160 to <180 mmHg
  - Any SBP >=180 mmHg
  - Any SBP increase from baseline >=20 to <40 mmHg
  - Any SBP increase from baseline >=40 mmHg
- Diastolic Blood Pressure
  - Any DBP >=100 to <110 mmHg
  - Any DBP >=110 mmHg
  - Any DBP increase from baseline >=10 to <20 mmHg
  - Any DBP increase from baseline >=20 mmHg
- Heart Rate
  - Any pulse >=100 to <120 bpm
  - Any pulse >=120 bpm
  - Any pulse increase from baseline >=20 to <30 bpm
  - Any pulse increase from baseline >=30 bpm
- Potassium
  - Any Potassium >5.5 to 6.0 mEq/L
  - Any Potassium >6.0 to 6.5 mEq/L
  - Any Potassium >6.5 mEq/L
  - Any Potassium increase from baseline >0.5 to 1.0
  - Any Potassium increase from baseline >1.0 to 1.5 mEq/L
  - Any Potassium increase from baseline > 1.5 mEq/L

Normotensive at baseline of Systolic Blood Pressure with are defined as and following criteria will be applied