NOVARTIS

Clinical Development

KAE609/Cipargamin

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An adaptive, randomized, active-controlled, open-label, sequential cohort, multicenter study to evaluate the efficacy, safety, tolerability, and pharmacokinetics of intravenous cipargamin (KAE609) in adult and pediatric patients with severe Plasmodium falciparum malaria (KARISMA – KAE609's Role in Severe Malaria)

Statistical Analysis Plan (SAP)

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List of abbreviations

AE	Adverse event
ACPR	Adequate clinical and parasitological response
ATC	Anatomical Therapeutic Classification
AUC	Area Under the Curve
Bid	bis in diem/twice a day
CDC	Centers for Disease Control and Prevention
CSR	Clinical Study report
CTC	Common Toxicity Criteria
CTCAE	Common Terminology Criteria for Adverse Events
DBL	Data base lock
ETF	Early treatment failure
DMC	Data Monitoring Committee
FAS	Full Analysis Set
eCRF	Electronic Case Report Form
IV	Intravenous
IVR	Interactive Voice Response
IWR	Interactive Web Response
LCF	Late clinical failure
L.O.S.	Level of significance
LPF	Late parasitological failure
MedDRA	Medical Dictionary for Drug Regulatory Affairs
NCI	National Cancer Institute
o.d.	Once Daily
PK	Pharmacokinetics
PPS	Per-Protocol Set
qd	Qua'que di'e / once a day
SAP	Statistical Analysis Plan
SOC	System Organ Class
TFLs	Tables, Figures, Listings
WHO	World Health Organization

1 Introduction

The purpose of this Statistical Analysis Plan (SAP), is to describe the implementation of the statistical analysis, which is planned in the protocol (Version 00). Mock shells for the clinical study report (CSR) will be prepared according to this SAP. Statistical outputs for the CSR will be generated after the final database lock (DBL) according to this SAP and the mock shells. A separate set of mock shells may be prepared and executed for DMCs based on this SAP.

Any data analysis carried out independently by the investigator should be submitted to Novartis before publication or presentation.

All CSR analyses will be carried out using SAS, Version 9.4 or higher.

1.1 Study design

This will be an adaptive, multicenter, randomized, open label, sequential cohort study in patients aged ≥ 12 years old in Cohorts 1-2 and ≥ 6 months to < 12 years old in Cohorts 3-5 with a diagnosis of moderately severe and severe *P. falciparum* malaria.

The study has five age descending cohorts; Cohorts 1 and 2 (≥ 12y), Cohort 3 (6y-< 12y), Cohort 4 (2y-< 6y), and Cohort 5 (6m-< 2y).

Cohorts 1 and 2:

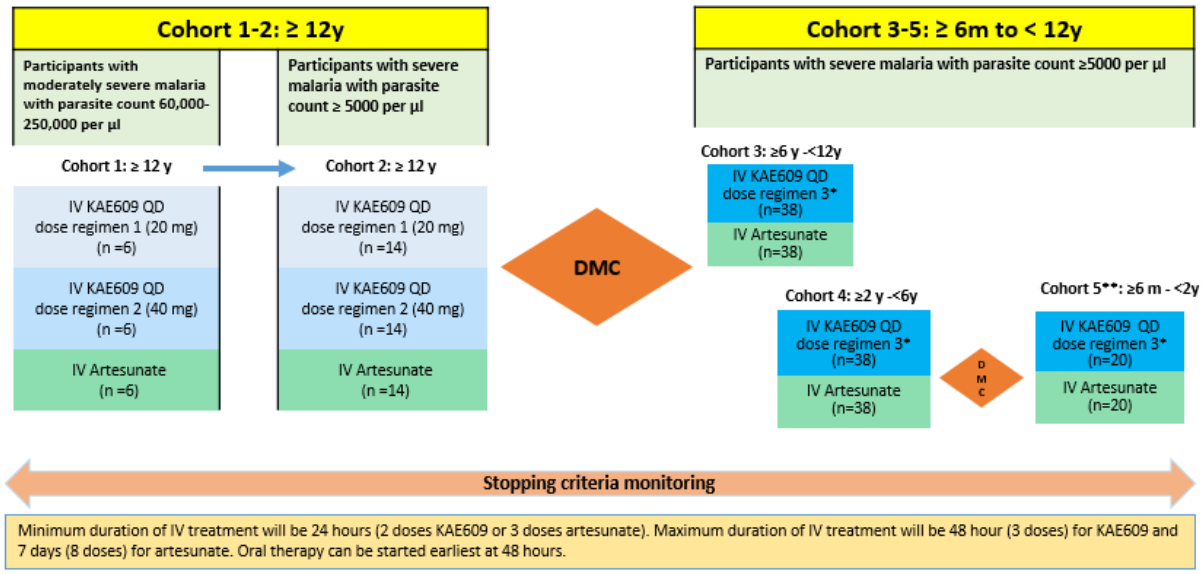
Patients will be randomized to one of the three treatment regimens in a 1:1:1 ratio. Two cipargamin based regimens will have the dose levels of 20 mg and 40 mg, whereas, the control regimen will be IV artesunate, a standard of care. Cohorts 1 and 2 plan to include 18 and 42 patients aged ≥ 12 years with body weight between 35-75 kg, respectively. The results from Cohorts 1 and 2 will be analyzed to determine the safe and efficacious exposure range and the corresponding doses (cipargamin (KAE609) dose regimen 3) for the patients in Cohorts 3 to 5.

In case of cipargamin dose adaptation, additional patients may be recruited with new dose levels in each cohort to ensure 6 and 14 patients per regimen in Cohorts 1 and 2, respectively.

Cohorts 3-5:

Two treatment regimens based on IV artesunate and IV cipargamin will be selected. Patients will be randomized to one of these regimens in 1:1 ratio. Cipargamin dose is not decided a priori at this time. Cohorts 3-5 include patients with age < 12 years and weight > 5kg. Cohort 3 includes 38 patients per regimen with age 6-<12 years; Cohort 4 also includes 38 patients per regimen with age 2-<6 years; whereas, Cohort 5 includes 20 participants per regimen with age 6 months - <2 years. Additional patients may be recruited in each cohort after dose modification to ensure designated number of patients on the same cipargamin dose level. After the completion of Cohorts 1 and 2, the DMC have endorsed the following cipargamin dosing schedule for children in Cohorts 3 and 4: 40 mg for body weight ≥35 kg, 24 mg for body weight 20 to <35 kg, 12 mg for body weight 10 to <20 kg, and 6 mg for body weight 5 to <10 kg.

Figure 1-1 Study Design



Notes:
KAE609 (Cipargamin)
Moderately severe malaria – Prostration and/or repeated vomiting (Barnes et al 2004)
Study to progress from Cohort 1 to Cohort 2 without interruption if none of the KAE609 arms in Cohort 1 meets the stopping criteria.
*Results from Cohort 1 and Cohort 2 will be analyzed to determine the safe and efficacious exposure range and the corresponding doses (KAE dose regimen 3) for participants in Cohorts 3 to 5.
Doses for Cohort 4 will be confirmed after PK analysis from upto 6 KAE609 participants from Cohort 3 and for Cohort 5, it would be post completion of Cohort 4 .
** Cohort 5 recruitment is dependent on availability of adequate toxicology package to dose down to ≥ 6 m
KAE609 doses across the study will be adaptive depending on the safe and efficacious exposure range identified before the study (for Cohorts 1-2) and after interim assessment of Cohort 1 & 2 (for Cohorts 3-5)

Randomization is stratified within each cohort according to country and previous antimalarial therapy use to achieve the balanced treatment allocation for these stratification factors.

Patients will be admitted to the hospital on Day 1. They will remain in hospital under close supervision until they are discharged by the investigator or designee on Day 4. At the discretion of the investigator, patients may stay for additional days if needed. The patients will then be followed up at Days 6, 8, 15, 22, and 29. Visits to assess safety and efficacy will be scheduled during the follow-up period as described in the schedule of assessments tables (Table 8-1 and Table 8-2) in the protocol.

In all cohorts, patients on IV cipargamin regimens will receive once daily injection of IV cipargamin over a minimum of 2 days and a maximum of 3 days; with IV artesunate as rescue medication. Patients in the IV artesunate regimen will be administered a minimum of 3 doses during the first 2 days of treatment. There is no rescue medication for IV artesunate regimen since it is the standard of care therapy recommended by WHO; however, if the patients are not responding in first 2 days, they will be treated for longer with IV artesunate. Patients in all regimens will receive Coartem BID for 3 days following IV therapy.

1.2 Study objectives and endpoints

Table 1-1 Objectives and related endpoints

Objective(s)	Endpoint(s)
Primary objective(s)	**Endpoint(s) for primary objective(s)**
• To assess the efficacy of IV cipargamin	• Proportion of patients with ≥ 90% *P. falciparum* parasite reduction at 12 hours, see Section 2.5.1 for the primary estimand
Secondary objective(s)	**Endpoint(s) for secondary objective(s)**
• To assess clinical outcome	• Proportion of patients with clinical success (see Section 2.7.2.1) over time. Clinical success at 48 hours is considered as the key secondary endpoint, see Section 2.6.1 for the secondary estimand.
• To assess the presence/absence of individual signs of severe malaria	• Proportion of patients with individual signs of severe malaria over time (see Section 2.7.2.2).
• To assess the risk of hemolysis	• Proportion of patients developing hemolysis (early and delayed) after treatment (see Section 2.8.2)
• To assess the risk of long term neurological sequelae	• Proportion of patients with neurological sequelae at Day 29 (see Section 2.7.2.3)
• To evaluate parasite clearance dynamics	• Proportions of patients with ≥ 90% parasite reduction at 24 and 48 hours • PCE slope half-life • Time to *P. falciparum* parasite clearance (PCT) • *P. falciparum* parasite reduction ratios (PRR) at 12, 24 and 48 hours • Proportion of patients with recrudescence and reinfection by Day 29 See Section 2.7.2
• To assess other efficacy endpoints	• Time (days and hours) to switch to oral therapy • Day of discharge from hospital • Time (hours) to recover from prostration See Section 2.7.2
• To evaluate the safety and tolerability of IV cipargamin	• Standard safety/tolerability assessments (incidence of serious adverse events (SAEs), mortality, in-hospital mortality, adverse events (AEs), and routine safety and laboratory assessments) See Section 2.8
• To assess the plasma pharmacokinetics of IV cipargamin	• PK parameters of cipargamin: Cmax, $T_{1/2}$, AUC, CL and Vz • Alpha-1-acid glycoprotein level over time and correlation of AAG concentration with cipargamin PK parameters See Section 2.9

Objective(s)	Endpoint(s)
Exploratory objective(s)	**Endpoint(s) for exploratory objective(s)**
	

2 Statistical methods

2.1 Data analysis general information

Data will be analyzed by Novartis using SAS version 9.4 or higher according to the data analysis presented in Section 12 of the study protocol, which is also available in Appendix 16.1.1 of the CSR. Detailed information is given in the following sections.

Data analyses required for the study Data Monitoring Committee (DMC) will be analyzed by a Novartis internal independent biostatistics team. The outline of the DMC analysis outputs is presented in the DMC charter and will be detailed in a separate mock shell based on this SAP.

Information on visit windowing, imputation rules and the methods of efficacy and safety analyses is given in the sections to follow. This SAP covers the methods for: DMC meetings, planned interim assessment, and the final analysis.

Unless otherwise stated, summary tables/figures/listings will be on all patients included in the population under consideration.

Categorical data will be presented as frequencies and percentages. For continuous data, n, mean, standard deviation, median, 25th and 75th percentiles (for selected parameters), minimum, and maximum will be presented. Additionally, geometric mean will be presented for PK parameters which may be better described using the lognormal distribution.

Unless otherwise stated, p-values (if required) will be provided for two-sided alternative hypotheses and presented up to 4 significant digits after decimal place; confidence intervals will be presented up to 2 significant digits after decimal place.

The dose level of treatment groups in a cohort may be adjusted if a chosen dose is not the optimal dose. Once the dose level has been adjusted, it becomes a different treatment group. If a dose regimen is considered similar among the 5 cohorts, data will be summarized by treatment

group for all 5 cohorts pooled and separately pooled Cohorts 3-5 in addition to summary by cohort and treatment group.

When data from different cohorts are pooled, an IV cipargamin regimen in Cohorts 3 to 5 (aged < 12 years) will be analyzed together with the equivalent IV cipargamin regimen as used in Cohorts 1 and 2 (aged ≥ 12 years). Such an IV cipargamin dose regimen will be denoted and labeled using the same cipargamin treatment group.

Key efficacy and safety data will be summarized separately for adults (aged ≥ 18 years) and children (aged <18 years).

Note: similar doses among 5 cohorts are based on the equivalent adult dose according to the probable weight bands for cohorts 3-5 as defined in protocol. However, they may be reassessed based on PK exposures at the end of study.

All post-text tables will be generated using Microsoft Word cell-based format so that they can be easily cut and pasted to create intext tables for the CSR. For each cohort, some treatment groups may consist sub-optimal dose levels. All treatment groups will be displayed in the post-text tables. However, only those treatment groups containing the dose levels that are deemed to be optimal for each cohort are important for displaying in the intext tables. Therefore, post-text tables will display all cohorts including the pooled cohort (if applicable) and all treatment groups under a cohort side by side as columns.

Randomization is stratified by country and use of any pretreatment of antimalarial drugs before screening period for each cohort. However, due to small sample sizes within a stratum, statistical analysis will not use these strata. For pooled analyses, study cohort (Cohorts 1 and 2 will be pooled together) will be used as stratum.

Parasite evaluations will be done by local microscopists for all the samples. Parasite evaluations will be also done using central laboratory for samples in Cohorts 1 and 2. Local readings will be used for the initial screening and local decisions for management of malaria. The statistical analysis will be performed using local laboratory readings for all cohorts and repeated using central laboratory readings for Cohorts 1 and 2. The identification for out of range P. falciparum parasite counts or other species for the exclusion of patients from the analyses sets will be based on local laboratory readings if they are available and central laboratory readings will be used in case of non-availability of the readings from the local laboratory. Both local laboratory and central laboratory readings will be all listed in the CSR appendix.

2.1.1 General definitions

Study treatment: IV Cipargamin and IV artesunate are randomized treatments. Coartem tablets (20/120 mg and 80/480 mg tablets) are used for oral therapy following the IV therapy for all patients. Coartem is not an investigational treatment and will not be presented in the treatment group name. Treatment group name will be used in statistical analyses and outputs. These treatment groups will be used as the column headers in the CSR outputs.

For example, the initial treatment groups in Cohort 1-2 will be displayed as:

- IV Cipargamin 20mg

- IV Cipargamin 40mg
- IV Artesunate

As per study design, other IV cipargamin dose levels may also be used. Treatment groups for each of the combinations will be used accordingly. For Cohorts 3 to 5, the IV Cipargamin dose level will be the equivalent dose level intended for patients ≥35 kg.

Baseline: The last measurement made prior to administration of the first dose of study treatment. Note: this may include measurements taken on the day of randomization (e.g. lab, ECG, vitals).

Study day: Study day will be calculated with respect to the first dose of study treatment. If a patient did not receive any dose of study treatment then the randomization date will be used as the date of first dose of study treatment. The first day of administration of study treatment (first dose) is defined as Day 1. Day -1 will be the day before Day 1. Day 0 does not exist. For some variables, assessments are collected more than once a day during first 3 days. Study day calculation for such assessments is based on the treatment start date and time.

For the assessments that are performed more than once a day: Study day =Integer (datetime of assessment – datetime of first dose of study treatment)/(60*60*24) + 1.

For assessments that are performed once for a day: Study day =date of assessment – date of first dose of study treatment + 1.

For assessments collected prior to Day 1, study day = date of assessment - date of first dose of study treatment.

Body temperature: Body temperature is measured using different routes (oral/rectal/tympanic/axillary), which will lead to variations in the readings. Therefore, all the temperature readings will be standardized with respect to tympanic equivalent as follows for the reporting purpose:
Body temperature (oral/tympanic/rectal equivalent) = Axillary temperature + 0.5ºC.

Fever Clearance time (FCT) (in patients with temperature of ≥ 37.5°C axillary or ≥ 38.0°C oral/tympanic/rectal) is defined as time from the first IV study medication until the first time the body temperature decreased below and remained below 37.5°C axillary or 38.0°C oral/tympanic/rectal for at least a further 24 hours,

Parasite clearance time (PCT) is defined as time from the first IV study medication until the first total and continued disappearance of asexual parasite forms which remained at least a further 48 hours.

Parasitaemia: P. falciparum asexual parasite counts are based on the local lab assessment for all cohorts and repeated using central lab assessments for Cohorts 1 and 2. Similar use of local lab readings and central lab readings will be applied to PRR, recrudescence, reinfection, and clinical success below.

Parasite detection limit for central lab: 0.1 parasite/uL will be used as detection limit or assay sensitivity limit for the parasite counts using the central lab.

Parasite detection limit for local lab: 10 parasite/uL will be used as detection limit or assay sensitivity limit for the parasite counts using the central lab.

PRR is defined as the ratio of asexual parasite at baseline divided by asexual parasite at post-baseline. If the asexual parasite count at post-baseline is 0, the detection limit will be used to calculate the ratio.

Recrudescence and reinfection

Recrudescence is defined as appearance of asexual parasites after clearance of initial infection with a genotype identical to that of parasites present at baseline. Recrudescence must be confirmed by PCR analysis.

Reinfection or new infection is defined as appearance of asexual parasites after clearance of initial infection with a genotype different from those parasites present at baseline.

Clinical success

Clinical success is a composite endpoint defined based on the following criteria.

A patient is said to have achieved the clinical success at a given timepoint if the patient did not die either during the initial hospitalization nor due to malaria post the initial hospitalization, shows absence of *P. falciparum* asexual parasites, and did not have any of the key signs or symptoms of severe malaria on that given timepoint.

Note: Deaths occurring post initial hospitalization will be evaluated by designated adjudication committee to confirm if they are related to malaria.

Key signs of Severe Malaria

Key signs of Severe Malaria are Altered consciousness, Renal impairment, Acidosis, and Respiratory distress.

Signs of severe malaria

The following signs of severe malaria will be monitored at every visit during the entire study duration.

- **Altered consciousness** – Prostration (inability to sit or drink or breast feed) or GCS < 11 for patients > 5 years / BCS < 3 for patients ≤ 5 years of age
- **Renal Impairment** - Serum creatinine > 3xULN or > 3 mg/dL or need for renal replacement therapy (RRT)
- **Acidosis** - Plasma lactate > 4 mmol/L or Respiratory distress.
  Respiratory distress is defined as rapid, deep and labored breathing (costal indrawing, use of accessory muscles and nasal flaring)- present or absent.
  In database, Plasma lactate is captured under lactic acid.
- **Severe anemia** - Hb < 5 g/dl or Hb < 7g/dl in pediatric and adults respectively or need of blood transfusion
- **Jaundice** - Serum bilirubin > 3 mg/dl
- **Hypoglycemia**- plasma glucose < 40 mg/dL
- **Blantyre coma scale (BCS)** is a number from 0 to 5 based on Appendix 5, Table 5-1 for patients ≤ 5 years. The score is determined by adding the results from three categories: Motor response, verbal response, and eye movement. The minimum score is 0 which indicates poor results while the maximum is 5 indicating good results.

- **Glasgow Coma Scale (GCS)** is a clinical scale used to reliably measure a person's level of consciousness for age > 5 years. The score is determined by adding the results from three categories: Motor response, verbal response, and eye movement. A person's GCS score can range from 3 (completely unresponsive) to 15 (responsive) based on Appendix 5, Table 5-2.

2.2 Analysis sets

Randomized set

All patients who are randomized into the study.

Full analysis set (FAS)

FAS will be comprised of all patients from Randomized set who take at least one dose of study treatment during the treatment period and whose baseline *P. Falciparum* asexual parasitaemia count is greater than 0 using local lab (central lab in case local lab assessment is not available). Following the intent-to-treat principle, patients will be analyzed according to the treatment group assigned to at randomization.

Unless otherwise specified, misrandomized patients will be excluded.

Misrandomized patients, if identified from IRT, include screen-failures, but have been randomized by the investigator before eligibility was finally assessed or mistakenly, and have not been treated. If patients were re-screened and successfully randomized, they will be included according to the treatment assigned in the last randomization.

Safety set (SAF)

Safety set includes all patients who take at least one dose of study drug during the treatment period. Patients will be analyzed according to treatment received. In particular, patient will be analyzed in the same treatment group as randomized if s/he receives at least one dose of the study medication of that group.

Per-protocol set (PPS)

Per-protocol set will be comprised of patients in FAS who did not have any important protocol deviations affecting efficacy.

Important protocol deviations and non-protocol deviations for exclusion from PPS are specified in Table 5-15 and will be identified by the clinical team before the database lock.

PK analysis set

All patients who have at least one valid (i.e. not flagged for exclusion) PK concentration measurement, receive at least one dose of study drug, and do not have any protocol deviations that have an impact on PK data.

2.2.1 Subgroup of interest

The following subgroups will be used for descriptive summary of the selected efficacy and safety analyses in all cohorts pooled. The details will be provided in the corresponding sections.

- Age groups as per Section 2.3
- Baseline weight groups as per Section 2.3
- Sex (male vs female)
- Different level of baseline HRP2 as per Section 2.3
- Geographic region (Africa, Asia)
- Use of previous antimalarial therapy before screening (Yes/No, stratification factor)

In addition, if deemed appropriate, additional subgroup analysis may be performed for data exploration; such as different *P. falciparum* gene resistance markers.

2.3 Patient disposition, demographics and other baseline characteristics

2.3.1 Medical history

Any condition entered on the *Relevant medical history / current medical conditions* CRF will be coded using the MedDRA dictionary. They will be summarized by system organ class (SOC), preferred term (PT) and treatment group for each cohort, for Cohorts 1 and 2 pooled and all cohorts pooled. A summary will also be provided of medical conditions that were ongoing at the time of screening.

Clinically relevant findings that are present prior to signing informed consent must be recorded on the appropriate CRF that captures medical history. Significant findings made after signing informed consent which meet the definition of an Adverse Event must be recorded as an adverse event. The findings occurring after signing ICF and before start of the study treatment will be listed.

Analyses will be based on the Full analysis set.

2.3.2 Patient demographics and other baseline characteristics

Demographic data and baseline disease characteristics will be descriptively presented and tabulated per treatment group, as well as overall, using FAS and PPS. If there are patients in the SAF excluded from FAS, a summary will also be provided for the SAF.

The following demographic variables will be summarized:

- Age and age categories (6 m to < 2 yrs, 2 to <6 yrs, 6 to <12 yrs, 12 to <18 yrs, 18 to <65 yrs, and ≥ 65, <18 vs ≥18 yrs)
- Sex (male, female)
- Race (Caucasian, black, Asian, native American, pacific islander, unknown, other)

- Ethnicity
- Body weight and weight categories (5 to <10, 10 to <15, 15 to <25, 25 to <35, 35 to <55, and ≥55 kg)
- Body height (cm)
- BMI and BMI categories (<16, 16 to 25, >25) (Kg/m^2)
- Head circumference <cm>(for Cohort 5)

The following disease characteristics at baseline will be summarized

- Body temperature and categories equivalent to tympanic method (<38, 38 to <39, ≥39) (°C)
    - *Falciparum* species: (*P. Falciparum* asexual forms, *P. Falciparum* gametocytes, *P. Vivax, P. Ovale, P. malariae, P. Knowlesi*, and Mixed-infection (two or more of *Falciparum* asexual forms, *P. Vivax, P. Ovale, P. malariae, P. Knowlesi)*, n (%)) using local lab for all patients as well as central lab for patients in Cohorts 1 and 2.
- *P. falciparum* density per micro ltr. and its categories (5,000 to <15,000, 15,000 to <50,000, 50,000 to <100,000, 100,000 to <150,000, 150,000 to < 200,000, ≥ 200,000 parasites/μL; <100,000 /μL, ≥100,000 parasites /μL) using local lab for all patients as well as central lab for patients in Cohorts 1 and 2.
- Gametocytes counts /μL
- Results of gene resistance markers of *P. falciparum*
- Baseline HRP2 concentrations ng/ml
- Baseline HRP2 concentration categories ng/ml (≤33th percentile, 34th to 67th percentile, >67th percentile; < median, ≥ median)
- Results of AAG
- HRP2/platelet status: plasma HRP2>1000ng/ml and <200,000 platelets/microliter

  Note- Categories that are with non-zero counts will be displayed.

2.3.3 Patient disposition

The number of patients who were screened and screening failures will be presented. In addition, the reasons for screen failures will be provided. The number and percent of patients who completed or discontinued the IV treatment and study (including the reason for discontinuation) will be presented for each treatment group and overall.

For each protocol deviation, the number and percent of patients for whom the deviation applies will be tabulated. Protocol deviations or non-protocol deviation criteria leading to exclusion from analysis sets will be summarized.

Randomized set will be used.

2.4 Treatments (study treatment, rescue medication, concomitant therapies, compliance)

2.4.1 Study treatment / compliance

Total number of doses and total dose (in mg) for each IV study drug will be summarized by treatment group. Number and percent of patients with different number of doses will be provided. Duration of IV treatment (in days) will be summarized by treatment group. Total number of doses, total dose of Coartem as oral therapy will also be summarized by treatment group; total dose in mg for Coartem is number of tablets multiplied with strength of the tablets in mg.

Number and percent of patients with minimal doses of IV study drug (2 doses for KAE609 and 3 doses for artesunate) will be provided.

Safety set will be used.

2.4.2 Prior, concomitant and post therapies

Prior and concomitant medications will be summarized in separate tables by treatment group.

Concomitant IV rescue and other anti-malarial medications will also be summarized by treatment group and drug.

Note – If a patient is randomized to IV cipargamin regimen but receiving IV artesunate per investigator's discretion, the IV artesunate will be classified as rescue medication. IV artesunate as rescue medications will be captured in DAR CRF whereas, other antimalarials will be recorded in concomitant medications CRFs.

Prior medications are defined as drugs taken and stopped prior to the first dose of study medication. Any medication given at least once between the day of first dose of study medication and the last day of study visit will be a concomitant medication, including those which were started pre-baseline and continued into the treatment period.

Prior and concomitant medications will be coded according to the latest version of WHO Drug Reference List dictionary which employs the ATC. The number and percentage of patients taking prior and concomitant medications will be summarized for each treatment by ATC class and preferred term (PT).

Number and percentages of the patients using concomitant medications in the management of signs and symptoms of severe malaria by treatment group will be summarized. A listing will be provided. These drugs will be identified from the clinical database prior to database lock. Number and percent of patients receiving concomitant non-study drug antimalarial will be provided by treatment group (See Table 5-11).

A listing of the patients who received prohibited prior and concomitant drugs will be provided. It is anticipated that only few patients will receive prohibited prior and concomitant drugs. Separate summary table for prohibited prior and concomitant drugs may also be provided if a higher number of patients are reported to receive such medicines. Prohibited drugs will be identified from the clinical database prior to database lock.

Safety set will be used. In case of differences in the safety set and FAS, the analysis will also be repeated on FAS.

2.5 Analysis of the primary objective

2.5.1 Primary Estimand

An estimand is a precise description of the treatment effect reflecting the clinical question posed by the trial objective. It summarizes at a population-level, what the outcomes would be in the same patients under each of the treatment conditions being compared. Attributes of an estimand include the population, variable (or endpoint) and treatment of interest. It also specifies how intercurrent events are addressed and provides a population-level summary for the variable. The structured framework of estimand detailed in ICH E9 (R1) was newly adopted by the Regulatory Members of the ICH Assembly under Step 4 in Nov-2019.

The clinical question of interest is: What is the difference between IV cipargamin and IV artesunate in early reduction of asexual parasite load in patients with severe *P. falciparum* malaria who are treated with at least one dose of IV treatment?

The justification for targeting this treatment effect is that severe *P. falciparum* malaria is caused by asexual parasite burden, leading to sequestration and microvascular obstruction. Rapid reduction of *P. falciparum* asexual parasites by 12 hours should prevent patients from further deterioration, and so allow eventual recovery from the disease.

The primary estimand is described by the following attributes:

1. Population: Patients with severe *P. falciparum* malaria and who have been treated with at least one dose of IV study medication (either cipargamin or artesunate). Further details about the population are provided in Section 5 in the protocol.
2. Variable: A binary outcome indicating ≥ 90% reduction in *P. falciparum* parasite at 12 hours (H12) after first IV dose as compared to baseline. A patient is considered as achieving primary endpoint if *P. falciparum* asexual parasite reading at 12 hours is either absent or with a positive value that is ≤ 10% of the baseline value.
3. Treatment of interest: IV cipargamin vs IV artesunate. Further details about cipargamin and artesunate are provided in Section 6 in the protocol .
4. Intercurrent events: (1) taking IV rescue antimalarial medication prior to H12 parasite assessment, or (2) mortality prior to H12 parasite assessment. Details on how to handle the intercurrent event are provided in Section 2.5.3.
5. Population level summary measure: Difference in proportion of patients with ≥ 90% reduction in baseline parasite count between IV cipargamin dose regimen and IV artesunate will be evaluated within each cohort separately except Cohort 1, which will be combined with Cohort 2.

2.5.2 Statistical hypothesis, model, and method of analysis

The statistical null hypothesis is that the difference in proportion of patients with ≥ 90% reduction at 12 hours between a cipargamin dose regimen and artesunate is ≤ -0.1 with the alternative hypothesis that the difference is > -0.1.

The statistical hypothesis will be tested at the 2-sided 10% significance level without multiplicity adjustment for multiple comparison due to multiple cipargamin dose regimen. Two-sided 90% confidence intervals (CI) for the difference between each cipargamin dose regimen vs artesunate will be provided using the Wilson uncorrected method: a cipargamin dose regimen is said to be non-inferior to artesunate if the lower limit of CI is > -0.1.

The hypothesis will be tested for each Cohort separately except Cohort 1, which will be combined with Cohort 2.

Data from patients in Cohorts 1 and 2 will be pooled together to allow evaluation of non-inferiority to artesunate since both cohorts include patients aged ≥ 12 years although patients in Cohort 1 are milder in clinical condition compared to patients in Cohort 2. The underlying assumptions are (1) the patients for each treatment group follow a binomial distribution separately in each Cohort except Cohort 1, which will be combined with Cohort 2, (2) patients for each treatment group, follow the same binomial distribution in Cohorts 1 and 2.

FAS will be used.

Wilson uncorrected method:

Let

a = # responders in cipargamin arm

c = # non-responders in cipargamin arm

m = sample size of the cipargamin regimen (a+c)

b = # responder in the artesunate arm

d = # non-responders in the artesunate arm

n = sample size of the artesunate regimen (b+d)

$\theta = \pi_1 - \pi_2$ is the difference between two arms

$\hat{\theta} = {}^{a}/_{m} - {}^{b}/_{n}$ is estimated difference between two arms

Method based on the Wilson score method for the single proportion, without continuity correction:

$L = \hat{\theta} - \delta$, $U = \hat{\theta} + \varepsilon$ where

$$\delta = \sqrt{\{(a/m - l_1)^2 + (u_2 - b/n)^2\}} = z\sqrt{\{l_1(1 - l_1)/m + u_2(1 - u_2)/n\}}$$

$$\varepsilon = \sqrt{\{(u_1 - a/m)^2 + (b/n - l_2)^2\}} = z\sqrt{\{u_1(1 - u_1)/m + l_2(1 - l_2)/n\}}$$

l_1 and u_1 are the roots of $|\pi_1 - a/m| = z\sqrt{\{\pi_1(1 - \pi_1)/m\}}$, and l_2 and u_2 are the roots of $|\pi_2 - b/n| = z\sqrt{\{\pi_2(1 - \pi_2)/n\}}$.

Where z is the standard normal deviate associated with 10% level of significance (l.o.s.) two sided.

2.5.3 Handling of intercurrent events of primary estimand

The primary estimand will account for the different intercurrent events as follows:

- If a patient receives any IV rescue antimalarial medication prior to the *P. falciparum* asexual parasite assessment at 12 hours, the patient will be classified as not achieving ≥ 90% reduction,
- If a patient dies prior to the *P. falciparum* asexual parasite assessment at 12 hours, the patient will be classified as not achieving 90% reduction.

Composite strategy is used to handle intercurrent events as the intercurrent events indicate that the study IV treatment is not working quickly enough to prevent clinical deterioration.

2.5.4 Handling of missing values not related to intercurrent event

All patients should be hospitalized at the time of 12 hours post treatment unless death or discontinuation from the study have occurred. Death is considered as an intercurrent events and is handled in Section 2.5.3. Any missing data not related to intercurrent events will be handled according to Table 2-1.

Since patients are hospitalized at 12 hours, these missing data should be rare. After receiving the IV study medication, a patient's parasite load should decrease over time such that the parasite count is lower at 12 hours than at previous visits (6 hours or earlier). That is, the probability that a patient achieves at least 90% parasite reduction is higher at 12 hours than at 6 hours. Therefore, the imputation of using parasite assessment at the latest visit prior to 12 hours is conservative. This imputation may slightly under estimate the proportion of patients with at least 90% parasite reduction at 12 hours.

Table 2-1 Handling of missing parasite data required for deriving primary endpoint

Event	Outcome
Discontinuation of study prior to parasite assessment at 12 hours, such as withdrawal of consent, COVID-19, etc.	Impute using the latest assessment prior to 12 hours including baseline. Note: the outcome is 'no' if baseline value is used.
Intermediate parasite count missing at 12 hours due to other reason (such as missing sample, technical reason, etc.)	

2.5.5 Sensitivity analyses

- After receiving antimalarial medication, a patient's parasite usually decreases log-linearly after a lag stage (White 2011, Flegg et al 2011). For artemisinin derivatives, parasitaemia starts to decline in a log-linear manner almost as soon as parasiticidal drug concentrations are reached (White 2011). The Tmax is about 0.5 to 15 minutes for IV artesunate (Artesunate PI). The Tmax is about 4 hours for oral cipargamin and should be much shorter for IV cipargamin. Missing data of parasite counts at 12 hours could be well estimated using a log linear interpolation of the 2 parasite counts immediate before and after 12 hours. Since there are 3 planned parasite assessments on or after 2 hours, missing parasite count at 12 hours due to early discontinuation could be estimated using patient specific log linear regression if there are at least 2 parasite counts prior to the discontinuation. A sensitivity analysis will be performed using interpolation for missing data at intermediate time points

and extrapolation using a log linear regression if there are at least 2 parasite counts prior to discontinuation. Regression line will be fitted for each patient separately. In the event that a patient does not have at least 2 parasite counts prior to discontinuation, the patient will be imputed as not achieving ≥ 90% reduction at 12 hours. Parasite counts below the limit of detection will be substituted using the limit of detection. If a patient's parasite count becomes below the limit of detection, only the first parasite count below detection will be used.

- The severity of disease is different between Cohort 1 and Cohort 2, which might impact the ability to achieve ≥ 90% parasite reduction, ie., resulting in a different binomial distribution between Cohorts 1 and 2. Therefore, two-sided 90% confidence intervals (CI) for the difference between each IV cipargamin dose regimen vs IV artesunate will be provided for Cohort 1 and Cohort 2 separately, using handling of missing values not related to intercurrent events specified in Section 2.5.4 and in this section.
- In order to assess the impact of IV rescue medication on parasites reduction, a listing of parasites details over time will be provided for the patients receiving IV rescue.

2.5.6 Supplementary analysis

For each Cohort (except Cohort 1, which will be pooled with Cohort 2), the difference between a cipargamin dose regimen and artesunate in the proportion of patients with ≥ 90% reduction in parasite at 12 hours will be performed using the Fisher exact test.

For each cipargamin dose regimen that is selected for Cohorts 3-5, the difference versus artesunate in proportion of patients with ≥ 90% reduction in parasite at 12 hours will be evaluated by pooling data from all cohorts to show if the IV cipargamin dose regimen is superior to IV artesunate. The treatment difference will be evaluated based on 2-sided 95% CIs using a Mantel-Haenszel estimate of the common risk difference stratified by cohort (Cohorts 1 and 2 will be pooled together and used as a single stratum)(SAS manual version 13.2). P-values will be provided using a Cochran-Mantel-Haenszel test (CMH) stratified by cohort.

Similarly, between treatment difference will be evaluated in children by repeating the above supplementary analysis on pooled Cohorts from 3-5 only.

A 2-sided 95% CIs for the proportion of patients with achieving primary endpoint for each treatment group will be provided based on the Clopper-Pearson exact method on pooled cohort.

These supplementary analyses target a different hypothesis to show superiority of each cipargamin dose regimen over artesunate and will be performed using the FAS and handling of intercurrent events and missing values not related to intercurrent events according to Section 2.5.3 and Section 2.5.4.

2.5.7 Supportive analyses

The analyses specified in Section 2.5.2, Section 2.5.5 and Section 2.5.6, will be repeated using the PPS.

Primary endpoint will be summarized descriptively by the subgroups defined in Section 2.2.1.

Local lab data will be used for all primary endpoint analysis for all cohorts. All the primary endpoint analysis will also be performed using central lab data for Cohorts 1 and 2.

2.6 Analysis of the key secondary objective

2.6.1 Secondary estimand

The secondary clinical question of interest is: What is the difference between IV cipargamin and IV artesunate in clinical success at 48 hours without the need of IV rescue antimalarial medication?

The justification for the secondary estimand is that it will reflect the clinical outcome including recovery from the most severe symptoms of the disease with IV treatment before earliest possible administration of oral antimalarial medication at 48 hours. This objective is being evaluated for possible use in Phase III study.

The secondary estimand is described by the following attributes:

1. Population: patients with severe *P. falciparum* malaria and treated with at least one dose of IV study medication. Further details about the population are provided in Section 5 in the protocol.
2. Variable**:** A binary outcome indicating clinical success- patient is alive and has absence of asexual parasites and no critical signs of severe malaria (See Section 2.1.1 for further details) at 48 hours.
3. Treatment of interest: IV cipargamin vs IV artesunate. Further details about cipargamin and artesunate are provided in Section 6 in the protocol.
4. Intercurrent event**:** receive IV rescue antimalarial medication before assessing the clinical success prior to H48.
5. Summary measure: Proportion of patients with clinical success with 90% CI within a treatment group as well as the difference between the cipargamin and artesunate treatment group.

2.6.2 Statistical hypothesis, model, and method of analysis for secondary estimand

There are no pre-specified hypotheses for secondary endpoints. Model, method of analysis, and handling of missing values/censoring/discontinuations are presented in the following sub-section.

The clinical success at 48 hours will be evaluated using the FAS with data from all cohorts pooled.

Intercurrent events and other non-intercurrent events including partial missing data that may occur and affect the derivation of secondary estimand will be handled in Section 2.6.3.

Two-sided 90% CIs for the difference between a cipargamin dose regimen and artesunate will be provided using a Mantel-Haenszel estimate of the common risk difference stratified by cohort where Cohorts 1 and 2 will be combined. Mantel-Haenszel estimate is a weighted risk difference with weight roughly proportional to strata size. It's an unbiased estimation of risk difference in the population under the following assumptions either (1) the risk difference is same across different strata or (2) the strata sizes in study resemble the strata size in the disease

population at large. Two-sided 90% CIs for the difference for the Mantel-Haenszel estimate of the common risk difference will be calculated using the local lab data only.

Descriptive statistics will be provided by cohort using the local lab data in addition to all cohorts pooled. Descriptive statistics will be provided for Cohorts 1 and 2 using the central lab data.

2.6.3 Handling of intercurrent events and missing data for secondary estimand

Handling of intercurrent events will be done according to Table 2-2; and missing data not related to intercurrent events will be handled according to Table 2-3.

Table 2-2 Handling of intercurrent events and other events that may occur and affect the derivation of secondary estimand

Event	Outcome
Receive IV antimalarial rescue medication prior to 48 hours	Not clinical success

Table 2-3 Handling of missing data that may occur and affect the derivation of secondary estimand

Reason for missing assessment	Method of imputation
Discontinue the study prior to 48 hours, such as withdrawal of consent, COVID-19, etc.	Not clinical success due to early discharge
Missing parasite assessment at 48 hours[$]	• Not clinical success if asexual parasites present in the latest visit prior to 48 hours • Ignored[*] otherwise
Altered consciousness: missing Prostration, missing Glasgow Coma Score or missing Blantyre coma score in relevant age groups[$]	Use the result from the latest visit prior to 48 hours (including baseline) for each missing item
Renal impairment: missing SCr[$]	• Not clinical success if Renal Replacement Therapy is used at 48 hours • Use the latest SCr prior to 48 hours
Acidosis and respiratory distress: missing plasma lactate or missing respiratory distress assessment[$]	Use the result from the latest visit prior to 48 hours (including baseline) for each missing item
[*] determination of clinical success depends on other assessments. [$] Partial data missing, unlikely since patients are in the hospital	

Note: partial data missing should be rare since patients are still hospitalized at 48 hours.

2.6.4 Sensitivity analysis for secondary estimand

For patients who discontinue the study prior to the H48 assessments, such as withdrawal of consent, COVID-19, etc. the worst outcome (not clinical success) is used. This may under estimate the clinical success rate at 48 hours. To assess the impact of worst assessment on the treatment difference, a sensitivity analysis will be performed by imputing missing outcome using the best outcome as "Clinical success".

2.6.5 Supplementary analysis for secondary estimand

Analysis defined for secondary estimand will be repeated by pooling all patients from Cohorts 3-5.

2.6.6 Supportive analysis for secondary estimand

Clinical success at 48 hours will be summarized descriptively by the subgroups defined in Section 2.2.1.

The analyses specified in Section 2.6.2 and Section 2.6.5, will be repeated using the PPS. Additionally, analysis specified in Section 2.6.2 may be performed using the PPS after excluding the patients who took other antimalarial drugs (PD ID= COMD01) before assessing the clinical success at 48 hours, if in case there are enough number of such patients exists.

A 2-sided 90% CIs for the proportion of patients with clinical success for each treatment group will be provided based on the Clopper-Pearson exact method on pooled cohorts.

A 2-sided 90% CIs for treatment difference between cipargamin and artesunate will be provided by cohort using the Wilson uncorrected method.

2.7 Analysis of remaining secondary efficacy objective(s)

Other secondary endpoints will be descriptively presented and tabulated (n, mean, standard deviation, median, minimum, and maximum for continuous variables; n and percent for categorical variables) by treatment group for each cohort separately. Cohort 2 will be also evaluated together with Cohort 1. For stratified analysis, Cohort 1 will be combined with Cohort 2. The cipargamin dose regimen that is dropped after reviewing data from Cohorts 1-2 will be included only in the by-cohort analyses. For those treatment groups that are selected for Cohorts 3-5, descriptive statistics will be provided by treatment group using all pooled Cohorts (1-5) as well as separately pooled Cohorts 3-5. When data from different cohorts are pooled, an IV cipargamin regimen in Cohorts 3 to 5 (aged < 12 years) will be analyzed together with the equivalent IV cipargamin regimen as used in Cohorts 1 and 2 (aged ≥ 12 years). Such an IV cipargamin dose regimen will be denoted and labeled using the same cipargamin treatment group. Analysis will be based on FAS.

2.7.1 Secondary efficacy endpoints

Secondary efficacy endpoints include:

- Clinical success at 24 hours and 72 hours, hospital discharge Day 6, Day 8, Day 15, Day 22, and Day 29. See Section 2.1.1 for definition of clinical success.
- Individual signs of severe malaria over time
  - Altered consciousness - Prostration or GCS < 11 for patients > 5 years old or BCS < 3 for patients ≤ 5 years old
  - Renal Impairment- Serum creatinine > 3 x ULN or > 3 mg/dL or need for renal replacement therapy
  - Acidosis- Plasma lactate > 4 mmol/L Or Respiratory distress – present or absent

  - Severe anemia- Hb < 5 g/dl or Hb < 7g/dl in pediatric and adults respectively with parasite count of > 10,000/µl or need of blood transfusion
  - Jaundice- Serum bilirubin > 3 g/dl with parasite count of > 100,000/µl
  - Hypoglycemia- plasma glucose < 40 mg/dL
- Proportion of patients with convulsion developing or persisting > 6 hour after start of IV study treatment
- Proportion of patients developing any new severe malaria signs (as per WHO severe malaria criteria) after treatment
- Proportion of patients with neurological sequelae (by adjudication) at Day 29
- Time to oral antimalarial therapy (in hours)
- Time to recover from prostration (in hours) in patients who are prostrated at baseline
- Proportions of patients with ≥ 90% *P. falciparum* asexual parasite reduction from baseline at 24 and 48 hours
- *P. falciparum* asexual parasite reduction ratios (PRR) at 12, 24 and 48 hours
- Time to *P. falciparum* asexual parasite clearance (PCT) (in hours)
- Time to fever clearance (FCT) (in hours)
- PCE slope half-life
- Proportion of patients with recrudescence and reinfection by Day 29

Hospital discharge day will be defined as the visit day where healthcare resource utilization has the hospital discharge date.

2.7.2 Statistical hypothesis, model, and method of analysis

There are no pre-specified hypotheses for secondary endpoints. Model, method of analysis, and handling of missing values/censoring/discontinuations are presented in the following sub-sections by topic.

2.7.2.1 Analysis of clinical success at timepoints other than 48 hours

The clinical success at hours 24, 72, at hospital discharge, Days 8, 15, 22, and 29 will be evaluated using the FAS and data from all cohorts pooled.

At each visit, 2-sided 90% CIs for the proportion of patients with clinical success for each treatment group will be provided based on the Clopper-Pearson exact method. In addition, 2-sided 90% CIs for the difference between a cipargamin dose regimen and artesunate will be provided using a Mantel-Haenszel estimate of the common risk difference stratified by cohort where Cohorts 1 and 2 will be combined.

Two-sided 90% CIs for the difference for the Mantel-Haenszel estimate of the common risk difference will be calculated using the local lab data only. Descriptive statistics will be provided for Cohorts 1 and 2 using the central lab data.

Intercurrent events and non-intercurrent events that may occur and affect the derivation of clinical success are handled in Table 2-4.

Table 2-4 Handling of other events that may occur and affect the derivation of clinical success at a visit other than 48 hours

Event	During hospital period (hours 24, 72 and at hospital discharge)[$]	After discharge from hospital (Days 8, 15, 22, and 29)
Receive IV antimalarial rescue medication prior to the visit	Not clinical success	Not clinical success
Death not related to malaria (as per adjudication committee)	Not applicable	Use the worst clinical success outcome between the hospital discharge visit and the last available visit
Discontinue the study prior to the visit due to other reasons, such as withdrawal of consent, lost to follow-up, COVID-19, etc.	Not clinical success due to early discharge	Use the worst clinical success outcome between the hospital discharge visit and the last available visit
Skipping the visit	Use the worst clinical success outcome from the visit before or visit after	Use the worst clinical success outcome from the visit before or visit after
Missing parasite assessments with visit	• Not clinical success if asexual parasites present in the previous visit • Ignored[*] otherwise	• Not clinical success if asexual parasites not cleared at the hospital discharge or parasites reappearance in a previous visit • Ignored[*] otherwise
Altered consciousness: missing Prostration, missing Glasgow Coma Score in adults or missing Blantyre coma score in children	Use the latest visit (including baseline) prior to the visit for each missing item	• Not clinical success if prostration or coma is reported as AEs for the visit • Ignored[*] otherwise
Renal impairment: missing SCr	• Not clinical success if Renal Replacement Therapy is used at the visit • Use the latest SCr prior to the visit	• Not clinical success if Renal Replacement Therapy is used at the visit • Not clinical success if renal failure or renal impairment is reported as AE for the visit • Ignored[*] otherwise
Acidosis and respiratory distress: missing plasma lactate or missing respiratory distress assessment	Use the result from the latest visit prior to the visit for each missing item	• Not clinical success if lactic acidosis or respiratory distress is reported as AE for the visit • Ignored[*] otherwise
[$] Partial data missing is unlikely prior to hospital discharge [*] Patient is considered as clinical success unless the patient does not meet clinical success due to other criterion		

Additional analyses

At each visit, 2-sided 90% CIs for the proportion of patients with clinical success for each treatment group will be provided based on the exact method and 2-sided 90% CIs for treatment difference vs artesunate will be provided using the Wilson uncorrected method by cohort.

For death not related to malaria and discontinuation of study after the hospital discharge, the following 2 sensitivity analyses will be performed:

- Use the last available clinical success outcome prior to death or discontinuation.
- Use multiple imputation methods (see Apendix 5.8.4)

2.7.2.2 Analyses of Individual signs of severe malaria

The following analysis for new severe malaria signs (as per WHO severe malaria criteria) after the IV study treatment will be performed.

The shift table of post-baseline presence relative to baseline presence of Individual signs of severe malaria or not will be provided by visit (24 hours, 48 hours, 72 hours, at hospital discharge, Day 6, Day 8, Day 15, Day 22, and Day 29), cohort, and treatment group. There are 7 individual signs and symptoms of malaria (See Section 2.1.1). Number and percent of patients with count of 0, 1, 2, 3, 4, 5, 6, and 7 different signs and symptoms by timepoint will be provided for each cohort and treatment group.

For BCS and GCS, individual scores for each category (eye opening, verbal response, motor system) will be listed. Refer to Appendix 5, Section 5.1 for more details.

In order to assess the impact of IV rescue medication, a listing of signs and symptoms of severe malaria over time will be provided for the patients receiving IV rescue.

Analysis will be based on FAS using observed data.

2.7.2.3 Analyses of neurological assessments

Analysis will be based on FAS using observed data.

Number and percent of patients will be provided by cohort and treatment group for the following neurological events related to severe malaria:

- Convulsion developing or persisting > 6 hour after the IV study treatment; where "persisting " means that convulsion is present at baseline and reoccurred after IV study treatment.
  - Moderate or severe neurological sequelae for each individual function domain and overall as assessed by investigator (Refer to Appendix 5, Section 5.1).
  - Overall moderate or severe neurological sequelae related to malaria episode at Day 29 confirmed by adjudication.

Following neurological examination results will be listed and summarized descriptively by visit, cohort, and treatment group.

- Type of convulsion
- Presence of ≥ 2 episodes of convulsions excluding febrile seizures during last 24 hours

- Any episode of convulsion lasting for ≥ 15 minutes
- Cranial Nerve Palsy
- Motor systems: Posturing, cerebellar ataxia, strength, Deep Tendon reflex, and tone
- Sense organs that characterize the vision, hearing, and speech

Neurological examinations are described in Appendix 5, Section 5.1.

2.7.2.4 Time to event analyses

Time to event will be calculated from the time of first study medication to the date of first event if a patient experiences the event and be censored at the time of last assessment if a patient does not experience the event. Randomization date will be used in the absence of study medication date.

For the following time to event variables, the first visit with the event will be used to calculate the time if a patient experiences the event. If a patient does not experience the event, the time will be censored at the last visit assessment for the specific type of event (such as last sit assessment, etc.) with the exception of date of discontinuation (or death) for time to oral antimalarial therapy.

- Time to *P. falciparum* asexual parasite clearance (PCT) (in hours)
- Time to fever clearance (FCT) (in hours)
- Time to oral antimalarial therapy (in hours),
- Time to recover prostration (in hours) in patients who are prostrated at baseline

Descriptive statistics (mean, standard error, median, quartiles) will be presented using the Kaplan-Meier method by treatment and cohort. Kaplan-Meier curves will be provided by treatment and cohort. For all cohorts pooled, difference for the cipargamin regimen vs artesunate will be evaluated using a log-rank test stratified by cohort (Cohorts 1 and 2 pooled as one cohort). Two-sided 90% CIs for the hazard ratio will be provided.

PCT will be calculated based on *P. falciparum* asexual parasite counts. Patients without parasite clearance for whatever reason will be censored at the time of last parasite assessment. Patients who received any non-study IV antimalarial medication (including IV rescue medication) before parasite clearance will be censored at the first use of antimalarial medication.

Patients who did not have a fever at pre-dose will not be included in the analysis of FCT. Patients without fever clearance for whatever reason will be censored at the time of last temperature assessment. Patients who received any antimalarial medication (including rescue medication) before fever clearance will be censored at the first use of antimalarial medication.

Patients without switching to oral medication for whatever reason will be censored at the time of last study assessment. Patients who received any IV rescue medication will be censored at the first use of IV rescue medication.

Patients who did not have a prostration at pre-dose will not be included in the analysis of time to recover prostration. Patients who do not recover prostration after the IV treatment for whatever reason will be censored at the last study assessment.

Above analysis will be based on FAS.

Tail of KM plots: Since the tail of KM plots after the day of last planned assessment is not reliable due to small numbers of patients at risk, the time to event will be reset to planned last day, i.e. Day 29, regardless of censoring. This principle will be used for relevant tables using the KM method.

2.7.2.4.1 Time to event analysis for Recrudescence

Recrudescence is defined as appearance of asexual parasites after clearance of initial infection with a genotype identical to that of parasites present at baseline. Recrudescence must be confirmed by PCR analysis.

Incidence rates of recrudescence up to Day 29 will be estimated by the Kaplan-Meier method based on the subset of FAS patients who have clearance of initial infection by Day 7. Descriptive statistics (mean, standard error, median, quartiles) will be presented using the Kaplan-Meier method by treatment and cohort. Kaplan-Meier curves will be provided by treatment and cohort. For all cohorts pooled, difference for the cipargamin regimen vs artesunate will be evaluated using a log-rank test stratified by cohort (Cohorts 1 and 2 pooled as one cohort). Two-sided 90% CIs for the hazard ratio will be provided.

Time to event (recrudescence) will be calculated from the time of first study medication to the date of first event if a patient experience the event and be censored at the time of last parasite assessment if a patient does not experience the event. Reappearance of the parasites but with missing PCR data will be considered as censored at the time of reappearance of parasites.

2.7.2.4.2 Time to event analysis for reinfection

Reinfection is defined as appearance of asexual parasites after clearance of initial infection with a genotype different from those parasites present at baseline. Reinfection must be confirmed by PCR analysis.

Incidence rates of reinfection at Day 29 will be estimated by the Kaplan-Meier method based on the subset of FAS patients who have clearance of initial infection by Day 7. Descriptive statistics (mean, standard error, median, quartiles) will be presented using the Kaplan-Meier method by treatment and cohort. Kaplan-Meier curves will be provided by treatment and cohort. For all cohorts pooled, difference for the cipargamin regimen vs artesunate will be evaluated using a log-rank test stratified by cohort (Cohorts 1 and 2 pooled as one cohort). Two-sided 90% CIs for the hazard ratio will be provided.

Time to event (reinfection) will be calculated from the time of first study medication to the date of first event if a patient experiences the event and be censored at the time of last parasite assessment if a patient does not experience the event. Reappearance of the parasites but with missing PCR data will be considered as censored at the time of reappearance of parasites.

2.7.2.5 Analyses of secondary endpoints related to parasite clearance dynamics

The following analyses are described for the ≥ 90% *P. falciparum* asexual parasite reduction, PRR and PCE slope half-life. The analysis will be based on FAS.

- Proportions of patients with ≥ 90% *P. falciparum* asexual parasite reduction from baseline at 24 and 48 hours

  For these two endpoints, data will be analyzed similar to the analysis of primary endpoint as specified in section 2.5.2. Intercurrent events as well as missing data will also be handled similar to that of primary endpoint as specified in section 2.5.3 and section 2.5.4.

- *P. falciparum* asexual parasite reduction ratios (PRR) at 12, 24 and 48 hours

  For each patient, the asexual PRR at a specific timepoint will be calculated as the ratio of asexual parasite at baseline divided by asexual parasite at that timepoint. If the asexual parasite count at a timepoint is 0/below assay detection limit, the detection limit will be used to calculate the ratio.

  Descriptive statistics including median and geometric mean of asexual PRR at Hour 12, 24 and 48 will be provided by cohort and treatment group using the FAS. For each PRR, 2-sided 90% CIs for geometric mean for each treatment group and geometric mean ratio between each IV cipargamin regime vs IV artesunate by cohort will be provided using an analysis of covariance (ANCOVA) model on the log scale with log baseline value as a covariate and treatment group as a factor by assuming PRRs follow log normal distribution and transformed back using the exponential.

  For all cohorts pooled, the analysis will be performed with cohort as an additional factor in the ANCOVA model. Observed data will be used.

- PCE slope half-life

  Parasite clearance rate constant and slope half-life for parasite clearance will be calculated for each patient using the WWARN (World Wide Antimalarial Resistance Network) Parasite Clearance Estimator as described in Flegg et al. (2011). The following are the key steps:

  For each patient separately:

  Step 1: Perform data cleaning. All further steps are performed on data with outliers, tails, extreme values and trailing zeros removed. For convenience of notation, if there is a zero parasitaemia that directly follows the last positive parasitaemia, it is replaced with the detection limit. This data point is subsequently counted as a non-zero parasite count.

  Step 2: Perform checks to see if the clearance rate constant cannot be estimated:

  (i) Number of non-zero parasite measurements (including a zero replaced with the detection limit) less than three

  (ii) Initial parasitaemia too low: initial parasitaemia < 1,000 parasites per microliter

  (iii) Final recorded parasitaemia too high: final parasitaemia ≥ 1,000 parasites per microliter

  (iv) A zero parasitaemia has been recorded, but the last positive parasitaemia is too high and the zero count is uninformative: last non-zero parasitaemia ≥ 1,000 parasites per microliter

  A zero parasitaemia is defined to be uninformative if the normal linear regression fitted to all the data points (excluding the zero count) gives a confidence interval for the time when the parasitaemia is below the level of detection which includes the time-point when the zero parasitaemia was recorded.

Step 3: Perform additional check to see if there are not enough data to estimate a lag phase *(v)* There are fewer than three measurements in the first 24 hours or a time difference between measurements in the first 24 hours is more than 14 hours

Step 4: Perform model fitting

- If none of the exclusion criteria (i)-(iv) in **Step 2** and (v) in **Step 3** are satisfied, fit polynomial models to the natural log-parasitaemia *versus* time data
- If none of the exclusion criteria (i)-(iv) in **Step 2** are satisfied but exclusion criterion (v) in **Step 3** is satisfied fit tobit linear regression if a zero parasitaemia has been recorded or linear regression otherwise.
- If one or more of the exclusion criteria are satisfied in **Step 2**, the clearance rate constant and duration of lag phase cannot be estimated.

Step 5: Estimate clearance rate constant (K) and duration of lag phase (t_{lag})

The above derivation will be performed using the PCE tool in R developed by WWARN. Descriptive statistics for parasite clearance rate constant and slope half-life for parasite clearance will be provided by cohort and treatment group using the FAS. 2-sided 90% CIs for each treatment group and for treatment ratio between each IV cipargamin regime vs IV artesunate by cohort will be provided using an analysis of covariance (ANCOVA) model on the log scale with log baseline value as a covariate and treatment group as a factor. For all cohorts pooled, the analysis will be performed with cohort as an additional factor in the ANCOVA model.

- Analysis of hospitalization data

Day of discharge from hospital will be summarized descriptively by cohort and treatment group. Number and percent of patients discharged on different days will be summarized. Day of death will be used in case of death during hospitalization. Time to discharge from hospitalization will also be analyzed using time to event analysis as described in Section 2.7.2.4. Patients who do not discharge or die during study will be censored at the last assessment or at the time of death respectively.

2.8 Safety analyses

All safety analyses will be performed using the safety set. All listings will be presented by cohort, treatment group, patient and visit/time if applicable. Summary tables will be presented by cohort (including pooled cohort,) and treatment group. Additionally, if there exists a similar treatment regimen among the cohorts, safety analysis will be performed by pooling the cohorts that have used the similar treatment regimen.

2.8.1 Adverse events (AEs)

The number and percent of patients with treatment emergent adverse events will be presented. The treatment emergent adverse events (events started on or after the first dose of study medication or events present prior to the first dose of study medication but increased in severity based on preferred term on or after the first dose of study medication) will be summarized by primary System Organ Class (SOC) and Preferred Term (PT).

Adverse events will be summarized by presenting, for each treatment group, the number and percentage of patients having at least one AE, having an AE in each primary system organ class and having each individual AE (preferred term). Summaries will also be presented for AEs by severity (Toxicity grade) and for study treatment related AEs. If a patient reported more than one adverse event with the same preferred term, the adverse event with the greatest severity will be presented. If a patient reported more than one adverse event within the same primary system organ class, the patient will be counted only once with the greatest severity at the system organ class level, where applicable.

The latest MedDRA version available before the DBL will be used for reporting the adverse events and the same will be described in a footnote.

The most common adverse events reported (≥ z % in any group for each preferred term in the table by SOC and PT) will be presented in the clinical study report by descending frequency according to its incidence in overall group starting from the most common event. Here threshold value z is set to 5 (%) but will be reviewed and updated as needed.

Separate summaries will be provided for study medication related adverse events, mortality, in hospital mortality, mortality related to malaria by adjudication, serious adverse events, adverse events leading to treatment discontinuation, adverse events leading to dose adjustment, and the safety stopping criteria, i.e., "All cause death or SAE possibly related to study drug". Summaries will also be provided separately for adverse events occurring during the IV study period (prior to oral standard of care, Coartem,) and for adverse events post the IV study period (after taking oral Coartem).

Serious adverse events in screening phase will be flagged in data listing due to rare frequency.

Patients who experienced a grade 3 or grade 4 AE will be summarized.

For the legal requirements of ClinicalTrials.gov and EudraCT, two required tables on treatment emergent adverse events which are not serious adverse events with an incidence greater than 5% and on treatment emergent serious adverse events and SAE suspected to be related to study treatment will be provided by system organ class and preferred term on the safety set. Five percent cut-off is to be reviewed and updated as needed prior to data base lock.

If for a same patient, several consecutive AEs (irrespective of study treatment causality, seriousness and severity) occurred with the same SOC and PT:

- a single occurrence will be counted if there is ≤ 1 day gap between the end date of the preceding AE and the start date of the consecutive AE
- more than one occurrence will be counted if there is > 1 day gap between the end date of the preceding AE and the start date of the consecutive AE
- For occurrence, the presence of at least one SAE / SAE suspected to be related to study treatment / non SAE has to be checked in a block e.g., among AE's in a ≤ 1 day gap block, if at least one SAE is occurring, then one occurrence is calculated for that SAE.

Number of deaths resulting from SAEs suspected to be related to study treatment and number of deaths resulting from SAEs irrespective of causality will be provided. Separate summaries

of deaths during initial hospitalization, post hospitalization, and post hospitalization malaria related deaths confirmed by adjudicated committee may be provided.

The adverse events and SAEs pooled from all Cohorts, if similar treatment regimen exists among the cohorts, will be summarized overall and for the subgroups defined in Section 2.2.1.

Algorithms for date imputations is provided in Appendix 5.

2.8.1.1 Adverse events of special interest / grouping of AEs

The adverse events of special interest are defined in the Case retrieval Sheet (CRS) which is updated for each MEDDRA dictionary. The CRS data are stored in TMS.ECRS SAS dataset corresponding to a subset of eCRS SAS view for which the following filtering criteria are applied

-Drug code = KAE609

-the latest version of MedDRA at the time of final database lock.

-End date is null (this means this is the latest eCRS version).

Potential risks based on the current eCRS are listed in Table 2-5.

The number and percent of patients with these special AEs will be summarized. In addition, listings of related adverse events will be provided.

Table 2-5 List of safety topics of interest

Risk or Adverse Reaction Name	Risk SOC	MedDRA Term	MedDRA Level	MedDRA Qualifier	MedDRA Code
Adrenal gland disorder	Endocrine disorders	Adrenal gland disorder [KAE609] (CMQ)	CMQ		90008629
Anemia	Blood and lymphatic system disorders	Haematopoietic erythropenia (SMQ)	SMQ	Broad	20000029
		Haemolytic disorders (SMQ)	SMQ	Broad	20000019
Antimalarial drug resistance	General disorders and administration site conditions	Development of clinical resistance to KAE609 in P. vivax and P. falciparum [KAE609] (CMQ)	CMQ		90008634
		Lack of efficacy (PSUR) [STANDARD] (NMQ)	NMQ		90000022
Gastrointestinal disorders	Gastrointestinal disorders	Gastrointestinal nonspecific symptoms and therapeutic procedures (SMQ)	SMQ	Narrow	20000140
Hepatotoxicity (GenMed)	Hepatobiliary disorders	Drug related hepatic disorders - comprehensive search (SMQ)	SMQ	Broad	20000006
Hypoglycemia	Metabolism and nutrition disorders	Hypoglycaemia (SMQ)	SMQ	Narrow	20000226
Inflammation	Investigations	Inflammation [KAE609] (CMQ)	CMQ		90012465
Injection site reactions	Injury, poisoning and procedural complications	ADR Injection site reaction [ADR_STD] (NMQ)	NMQ		90003467
Phototoxicity	Skin and subcutaneous tissue disorders	Photosensitivity reactions [STANDARD] (NMQ)	NMQ		90000921
QTc prolongation	Cardiac disorders	Torsade de pointes/QT prolongation (SMQ)	SMQ	Broad	20000001
Semen discoloration	Reproductive system and breast disorders	Fertility disorders-male (based on SMQ) [STANDARD] (NMQ)	NMQ	Broad	90001282

2.8.2 Laboratory data

Laboratory data for hematology and biochemistry parameters will be assessed from a point of care device. In case of non-availability of the point of care device due to any reason, local laboratory evaluation is preferred. However, statistical analysis will be based on the combined data from both types of assessments.

Descriptive statistics will be generated for all clinical laboratory tests performed (actual values and changes from baseline) for three groups of laboratory tests (hematology, clinical chemistry and urinalysis) by laboratory test and treatment group. Change from baseline will only be summarized for patients with both baseline and post baseline values and will be calculated as:

change from baseline = post baseline value – baseline value

The following laboratory parameters will be analyzed for hematology test group: hemoglobin, platelets, white blood cell count, hematocrit, red blood cell (RBC) count, lymphocytes, lymphocytes (%), monocytes, monocytes (%), eosinophils, eosinophils (%), neutrophils, neutrophils(%) , basophils, and basophils (%).

The following laboratory parameters will be analyzed for Biochemistry test group: creatinine, total bilirubin (TBL), direct bilirubin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), glucose, blood urea nitrogen (BUN), sodium, potassium, calcium, albumin, glucose, Bicarbonate, Estimated glomerular filtration rate (eGFR), lactate, LDH, and INR (in selected cases).

Box plots for selected biochemistry and hematology parameters normalized using ULN (upper limit of normal) will be provided by timepoint and treatment group.

For liver enzymes (ALT, AST, etc.), the shift table of CTCAE grades relative to baseline will be provided. For other lab parameters, the shift tables using low/normal high will be provided for each visit and a worst post-baseline value. Summary with frequency and percentage of patients with liver related events as defined in Table 2-6 will be provided by treatment:

Table 2-6 Liver-related events

Parameter	Criterion
ALT	>3xULN; >5xULN; >8xULN;>10xULN; > 20XULN
AST	>3xULN; >5xULN; >8xULN; >10xULN; >20XULN
ALT or AST	>3xULN; >5xULN; >8xULN; >10xULN; >20XULN
ALT or AST and TBL	ALT or AST > 3 × ULN and TBL > 2 x ULN ALT or AST > 5 × ULN and TBL > 2 x ULN ALT or AST > 10 × ULN and TBL > 2 x ULN
TBL	>1.5xULN, >2xULN, >3XULN
ALP	>2xULN, >3xULN
ALP & TBL	ALP >3xULN & TBL >2xULN
(ALT or AST) & TBL & ALP	ALT or AST >3xULN & TBL >2xULN & ALP <2xULN

Peak ALT and AST and peak TBL will be plotted using eDISH (evaluation of Drug Induced Serious Hepatotoxicity) method and Modified eDISH (mDISH) method, see Lin, et. al. (2012).

Listing of patients with the following laboratories will be provided:

- Serum creatinine increase 25 – 49% compared to baseline
- Serum creatinine increase ≥ 50% compared to baseline
- New Plasma lactate > 4 mmol/l
- New dipstick proteinuria ≥ 1+
- New dipstick hematuria ≥ 1+

For urinalysis, frequency tables will be presented. Number and percent of patients in each category will be presented for each visit.

Laboratory measurements, which are recorded as below the assay detection limits, will be imputed as half of the detection limit; and those above the detection limit will be imputed as within the detection limit, for the summary statistics.

Analysis of Hemolysis

Early Hemolytic anemia is defined as 10% or greater decrease in hemoglobin levels and an increase of LDH levels to >390 U/L, or an increase of ≥10% above baseline occurring up to Day 8 of the study.

Delayed hemolytic anemia occurs > 7 days after initiation of parenteral study drug (IV artesunate or IV cipargamin) during the study period. It is defined as 10% or greater decrease in hemoglobin levels and an increase of LDH levels to >390 U/L, or an increase of ≥10% above Day 8 value. If Day 8 value is missing, use the closest value.

The number (and proportion) of patients with early hemolysis and number (and proportion) of patients with delayed hemolysis will be summarized by phase (Early or Delayed) and visit (Early: Days 1 to 3, Days 4 to 8, Days 1 to 8; Delayed: Days 9 to 15, Days ≥ 16, ≥ Day 9), cohort, and treatment group. For delayed hemolysis, visits after Day 8 will be reported.

Besides, number and percent of patients with decrease in hemoglobin ≥ 10% (from baseline for early and from Day 8 for delayed) will be summarized by phase (Early or Delayed) and visit, cohort, and treatment group.

Analysis of Lactate

A shift table presenting a shift in lactate levels from baseline to each post-baseline visit will be provided by visit, cohort, and treatment group. The following categories will be used; ≤ 2 , > 2 - <4 , ≥ 4 mmol/l.

The percentage change from baseline in lactate level will be summarized by visit, cohort, and treatment group.

Time to normalized lactate level (lactate ≤ 2 mmol/l) (in hours) in patients with baseline lactate level above normal will be summarized descriptively using the Kaplan-Meier method by cohort and treatment group. The Kaplan-Meier curve will be also presented. If for a patient the blood lactate does not normalize, the time will be censored at the last blood lactate assessment. Patients with missing baseline lactate values will be excluded.

2.8.3 Other safety data

2.8.3.1 ECG and cardiac imaging data

The following quantitative variables will be summarized for averaged triplicate ECG: heart rate, RR interval, PR interval, QRS interval, QT interval, QTcF (QT interval corrected for heart rate according to Fredericia) and QTcB (QT interval corrected for heart rate according to Bazett) for each timepoint and maximum post baseline value.

Patients with newly notably high QT, HR and fever (≥38 °C, tympanic equivalent) will be summarized.

The analysis results for categorical outliers and T-wave morphology may be summarized in frequency tables with counts and percentages for both number of participants and number of timepoints. For categorical outliers, the number of participants (%) and timepoints (and at the maximum post baseline) will be determined by treatment group.

QTc will be summarized categorically by computing the number and percentage of patients at each time point and at the maximum post baseline value with following:

- QT, QTcF, or QTcB
  - > 450 and ≤ 480 ms
  - > 480 and ≤ 500 ms
  - > 500 ms
  - Increase from Baseline of ≥ 30 ms to < 60ms
  - Increase from Baseline of ≥ 60 ms
- PR
  - Increase from baseline >25% and to a value > 200 ms
  - > 120 ms
- HR for age ≥ 18 years
  - Decrease from baseline >25% and to a value < 50 bpm
  - value of <50 bpm
  - Increase from baseline >25% and to a value > 100 bpm
  - > 100 bpm
- HR for age <18 years
  - > 90^{th} percentile defined in Table 2-7
  - < 10^{th} percentile defined in Table 2-7
- QRS
  - Increase from baseline >25% and to a value > 120 ms
  - > 120 ms

For T-wave morphology, if performed, the analysis will be focused on the treatment emergent changes.

Table 2-7 Heart rate cut-offs (beats/minute) based on centile charts

Age Range#	10th percentile	90th percentile
Birth	107	148
0 – <3m	123	164
3 – <6m	120	159
6 – <9m	114	152
9 – <12m	109	145
12 – <18m	103	140
18 – <24m	98	135
2 – <3y	92	128
3 – <4y	86	123
4 – <6y	81	117
6 – <8y	74	111
8 – <12y	67	103
12 – <15y	62	96
15 – <18y	58	92

Age ranges given in years (y) and months (m). "Birth" refers to the immediate neonatal period.
Reference: Lancet. 2011 March 19; 377(9770): 1011–1018. doi:10.1016/S0140-6736(10)62226-X.

A line plot by patient (Spaghetti plot) will be provided by visit and treatment group.

In addition, shift tables comparing baseline ECG results (normal, abnormal, not available, total) with the post-baseline results will be provided by visit and at the worst post-baseline result (normal, abnormal, not available, total).

The relationship between change from baseline in QTcF/QTcB and cipargamin plasma concentration will be assessed graphically with change from baseline in QTcF/QTcB plotted against cipargamin plasma concentrations.

Cipargamin plasma concentration-QT relationship may also be explored using linear mixed effects model.

A listing of all newly occurring or worsening abnormalities as compared with baseline will be provided, as well as a by-patient listing of all quantitative ECG parameters.

2.8.3.2 Vital signs

The following quantitative variables will be summarized by treatment, cohort, and visit: Body height (cm) in Cohorts 1-4, Weight (kg), head circumference (Cohort 5), Temperature (tympanic equivalent) (^{o}C), Pulse (beats/min), respiratory rate (breaths/min), Systolic blood pressure (mmHg) and Diastolic blood pressure (mmHg).

Analysis in vital sign measurement using descriptive summary statistics for the change from baseline for each post-baseline visit will be performed. Change from baseline will only be summarized for patients with both baseline and post-baseline values and will be calculated as:

Change from baseline = post-baseline value – baseline value

The number and percentage of patients with newly occurring notable vital signs will be presented. Criteria for notable vital sign abnormalities are provided in Table 2-8 and Table 2-9 below.

A listing of all newly occurring or worsening abnormalities as compared with baseline will be provided, as well as a by-patient listing of all vital signs.

All patients level vital signs data will be listed.

Table 2-8 Criteria for notable vital sign abnormalities for age ≥ 18 years

Vital sign (unit)	Criteria
Systolic blood pressure (mmHg)	≥ 180 mmHg/≤ 90 mmHg with increase/decrease from baseline of ≥ 20 mmHg
Diastolic blood pressure (mmHg)	≥ 105 mmHg/≤ 50 mmHg with increase/decrease from baseline of ≥ 15 mmHg
Pulse (beats/min)	≥ 120 bpm/≤ 50 bpm with increase/decrease from baseline of ≥ 15 bpm
Respiratory rate	≥30 bpm ≤ 10 bpm
Temperature (°C)	≥ 37.5 (axillary) or ≥ 38.0 (other routes)
Weight	decrease > 10% from Baseline increase > 10% from Baseline

Table 2-9 Criteria for notably abnormal vital signs for age <18 years

Vital sign (unit)		Criteria	
Systolic blood pressure [mmHg]	High	≥ 95th percentile of the age and height group[1]	
	Low	≤ 5th percentile of the age and height group[1]	
Diastolic blood pressure [mmHg]	High	≥ 95th percentile of the age and height group[1]	
	Low	≤ 5th percentile of the age and height group[1]	
Body temperature [°C]	High	≥ 38°C (*tympanic equivalent*)	
	High	1-<6 months	> 160
		6-<12 months	>150
		12-<18 months	> 140
		18-<24 months	> 135
		2-<3 years	> 128
		3-<4 years	> 123
		4-<6 years	> 117
		6-<8 years	> 111
		8-<12 years	> 103
		12-<15 years	> 96
		≥ 15 years	> 92

Vital sign (unit)		Criteria	
Pulse rate [bpm] [2]	Low	1-<6 months	<120
		6-<12 months	<110
		12-<18 months	< 103
		18-<24 months	< 98
		2-<3 years	< 92
		3-<4 years	< 86
		4-<6 years	< 81
		6-<8 years	< 74
		8-<12 years	< 67
		12-<15 years	< 62
		≥ 15 years	< 58
Respiratory rate [breath per minute] [2,3,4]	High	1-<6 months	>55
		6-<12 months	>50
		12-<18 months	> 46
		18-<24 months	> 40
		2-<3 years	> 34
		3-<4 years	> 29
		4-<6 years	> 27
		6-<8 years	> 24
		8-<12 years	> 22
		12-<15 years	> 21
		≥ 15 years	> 20
	Low	1-<6 months	<33
		6-<12 months	< 30
		12-<18 months	<28
		18-<24 months	< 25
		2-<3 years	< 22
		3-<4 years	< 21
		4-<6 years	< 20
		6-<8 years	< 18
		8-<12 years	< 16
		12-<15 years	< 15
		15-18 years	< 13
Weight	High	increase from baseline of ≥ 2 BMI-for-age percentile categories[5]	
	Low	decrease from baseline of ≥ 2 BMI-for-age percentile categories[5]	

bpm=beats per minute; NHLBI= National Heart, Lung, and Blood Institute;

[1] Fourth Report on Diagnosis, Evaluation and Treatment of High Blood Pressure in Children and Adolescents. Pediatrics 2004; 114; 555 (see section 5.6 for detailed computation)

[2] Fleming S, Thompson M, Stevens R, et al. Normal ranges of heart rate and respiratory rate in children from birth to 18 years of age: a systematic review of observational studies. Lancet 2011; published online March 15. DOI:10.1016/S0140-6736(10)62226-X.

[3] Eldridge L. What is a Normal Respiratory Rate?, Updated May 16, 2014;

[4] Kou .R., Shuei L., Bradypnea, Department of Physiology, School of Medicine, National Yang-Ming University, Taipei, Taiwan,
https://rd.springer.com/referenceworkentry/10.1007/978-3-540-29676-8_246

Vital sign (unit)		Criteria
[5] BMI-for-age percentiles categories (P3, P5, P10, P25, P50, P75, P85, P90, P95, P97) are obtained from the WHO Growth Charts https://www.who.int/childgrowth/standards/bfa_boys_p_exp.txt https://www.who.int/childgrowth/standards/bfa_girls_p_exp.txt https://www.who.int/tools/growth-reference-data-for-5to19-years/indicators/bmi-for-age		

In addition to above analysis, for patients < 18 years of age, number and percentage of patients with increase or decrease from baseline of 1 BMI-for-age percentile category (See Table 2-11) will be provided by visit, cohort and treatment group.

The head circumference data will be analyzed descriptively only since it is not expected to change the values significantly within four weeks of the study.

2.9 Pharmacokinetic endpoints

Only cipargamin pharmacokinetics will be determined in this study.

PK parameters such as AUC_{inf}, AUC_{last}, AUC_{0-t}, C_{max}, C_{2min}, T_{max}, $T_{1/2}$, CL/F, and Vz are determined using non-compartmental method(s) and will be used for the analysis of PK.

Summaries of PK concentrations will be provided by cohort, treatment, and visit. PK concentrations below the limit of quantification (LLOQ) will be treated as zero in summary statistics and for geometric related mean and CV, concentrations below LLOQ will be included as half of the LLOQ value. Descriptive statistics of pharmacokinetic parameters will include arithmetic and geometric means, standard deviation (SD), coefficient of variation, (CV %), median, minimum and maximum, etc. CV% geo-mean will be calculated using this formula; (sqrt (exp (variance for log transformed data)-1))*100.

Two-sided 90% confidence intervals for the mean or geometric mean will be calculated by cohort and treatment group using normal or log-normal approximation as appropriate.

For those treatment groups that are selected for Cohorts 3-5, PK analysis will be provided by treatment group using all pooled Cohorts (1-5) as well as separately pooled Cohorts 3-5. When data from different cohorts are pooled, an IV cipargamin regimen in Cohorts 3 to 5 (aged < 12 years) will be analyzed together with the equivalent IV cipargamin regimen as used in Cohorts 1 and 2 (aged ≥ 12 years).

The pharmacokinetic parameters will also be listed by cohort, treatment and patient.

AAG

α -1-acid glycoprotein (AAG) data will be collected only for patients randomized to cipargamin. Summary statistics including geometric mean will be provided by visit, cohort and pooled cohorts also. The relationship between cipargamin exposure and AAG will be explored graphically using scatter plot including the regression line for cipargamin exposure on AAG. Scatter plot for cipargamin PK parameters and AAG will be provided by cohort and pooled. Listing will be provided by cohort, patient and visit.

2.10 CCI

CCI

2.11 Patient-reported outcomes

NA

2.12 CCI

CCI

2.13 CCI

CCI


CCI


CCI


CCI

2.14 Overview of analysis methods

An overview of statistical analyses and methods applied to efficacy variables and safety variables are given in Table 2-11 and Table 2-12.

Table 2-10 Overview of analysis methods for baseline, disposition and concomitant medications

Variable(s)	Listings	Summary statistics for binary/ categorical data	Summary statistics for continuous data
Medical history	X	X	-
Demographics and baseline characteristics	X	X	X
Patient disposition	X	X	-
Prior medication use	X	X	-
Concomitant medication use	X	X	-
Concomitant IV rescue medication use	X	X	-
Concomitant Non-study drug antimalarial medication use	X	X	-
Prior other prohibited medications use	X	Optional	-
Concomitant other prohibited medications use	X	Optional	-

Table 2-11 Overview of analysis methods for efficacy variables

Variable(s)	Listings	Summary statistics for binary/ categorical data	Summary statistics for continuous data	Summary statistics for time to event variable	Graphs	Method of analysis for between treatment comparison by cohort	Method of analysis for pooled cohorts			Subgroup summary
							Summary	between treatment comparison	By treatment	
≥ 90% reduction in *P. falciparum* parasite at 12 hours	X	X	-	-	-	• Wilson's uncorrected method, • Fisher Exact test,	X	Mantel-Haenszel estimate	-	X
≥ 90% reduction in *P. falciparum* parasite at 24 and 48 hours	X	X	-	-	-	• Wilson's uncorrected method, • Fisher Exact test,	X	Mantel-Haenszel estimate	-	X
Clinical success at 48 hours	X	X	-	-	-	Wilson's uncorrected method,	X	Mantel-Haenszel estimate	Clopper-Pearson	X
Clinical success at other timepoints	X	X	-	-	-	Wilson's uncorrected method,	X	Mantel-Haenszel estimate	Clopper-Pearson	X
Individual signs of severe malaria over time	X	X	-	-	-	-	-	-	-	-
Convulsion after >6 hours post treatment	X	X	-	-	-	-	-	-	-	-
Neurological sequalae by Day 29 and other neurological assessment		X		-	-	-	X	-	-	
PRR at 12, 24, 48 hours	X	-	X	-	-	ANCOVA	X	ANCOVA	-	-

Variable(s)	Listings	Summary statistics for binary/ categorical data	Summary statistics for continuous data	Summary statistics for time to event variable	Graphs	Method of analysis for between treatment comparison by cohort	Method of analysis for pooled cohorts			Subgroup summary
							Summary	between treatment comparison	By treatment	
Time to parasite clearance (PCT)	X	-	-	X	X	-	X	• Stratified Log-rank test • Hazard ratio	-	-
Time to fever clearance (FCT)	X	-	-	X	X	-	X	• Stratified Log-rank test • Hazard ratio	-	-
Time to oral antimalarial therapy	X	-	-	X	X	-	X	• Stratified Log-rank test • Hazard ratio	-	-
Time to hospitalization discharge	X	-	-	X	-	-	X	• Stratified Log-rank test • Hazard ratio	-	-
Time to recover from prostration	X	-	-	X	X	-	X	• Stratified Log-rank test • Hazard ratio	-	-
Time to recrudescence/re-infection	X	-	-	X	X	-	X	• Stratified Log-rank test • Hazard ratio	-	-
Disappearance or Development of gametocytaemia	X	X	-		-	-	X	-	-	-

Variable(s)	Listings	Summary statistics for binary/ categorical data	Summary statistics for continuous data	Summary statistics for time to event variable	Graphs	Method of analysis for between treatment comparison by cohort	Method of analysis for pooled cohorts			Subgroup summary
							Summary	between treatment comparison	By treatment	
PCE slope half-life	X	-	X		-	ANCOVA	X	ANCOVA	-	-
CCI										
Meeting the lack of efficacy criteria (use of rescue medication)	X	X	-		-	-	X	-	-	

Table 2-12 Overview of analysis methods for safety/PK variables

Variable(s)	Listings	Summary statistics for binary/ categorical data	Summary statistics for continuous data	Graphs	Pooled summary
AE	X	X	-	-	X
SAE	X	X	-	-	X
Adverse events of special interest	X	X	-	-	X
Hematology change from baseline	X	-	X	X	X
Biochemistry change from baseline	X	-	X	X	X
Liver abnormalities	X	X	-	-	X
ECG abnormality	X	X	-	-	X
QTc change from baseline	X	-	X	X	X
Vital signs change from baseline	X	-	X	-	X
Notable vital signs abnormality	X	X	-	-	X
Drug concentrations	X	-	X	X	X
PK parameters, AAG	X	-	X	-	X
CCI					
Number of patients meeting the safety stopping criteria	X	X	-	-	X

2.15 Interim analysis

2.15.1 Interim analysis/ DMC review

Key Safety/efficacy/PK data will be analyzed after completion of Cohort 2. The results will be communicated to relevant internal and external people who will contribute to the selection of effective and safe dose for next Cohort or continuation of the same Cohort.

DMC analysis: DMC analyses will be performed at the following timepoints

- After cohort 2 completion
- After cohort 4 completion
- Unplanned

Below is the outline of the data required for the DMC/data review.

Table 2-13 Specification of pre-specified reports for DMC/data review

Output	Analysis set	DMC after Cohort 4 or unplanned safety meetings (CTT blinded)	IA after Cohort 2 (CTT unblinded)
Baseline			
Patient disposition	FAS	Yes	Yes

Output	Analysis set	DMC after Cohort 4 or unplanned safety meetings (CTT blinded)	IA after Cohort 2 (CTT unblinded)
Recruitment by country and center	FAS	Yes	Yes
Demographic characteristics	FAS	Yes	Yes
Background disease characteristics	FAS	Yes	Yes
Safety			
Concomitant medications/significant non-drug therapies	Safety	Yes	Yes
All adverse events, mortality, in-hospital mortality, and Serious adverse events	Safety	Yes	Yes
Number of patients with adverse events of special interest	Safety	Yes	Yes
General, and liver, specific safety laboratories (categorical analysis)	Safety	Yes	Yes
Vital signs (clinically notable)	Safety	Yes	Yes
ECG	Safety	Yes	Yes
Proportion of patients developing delayed hemolysis after treatment	Safety	Yes	Yes
Proportion of patients with neurological sequelae at Day 29	Safety	Yes	Yes
Number of patients meeting the safety stopping criteria	Safety	Yes	Yes
Efficacy			
Number of patients meeting the lack of efficacy criteria (use of rescue medication)	FAS	Yes	Yes
Proportion of patients with individual signs of severe malaria over time	FAS	Yes	Yes
Clinical success	FAS	No*	No
Parasite clearance time (PCT)	FAS	Optional	Yes

Output	Analysis set	DMC after Cohort 4 or unplanned safety meetings (CTT blinded)	IA after Cohort 2 (CTT unblinded)
Proportion of patients with ≥ 90% *P. falciparum* parasite reduction at 12/24/48 hours	FAS	Optional	Yes
P. falciparum parasite reduction ratios (PRR) at 12, 24 and 48 hours	FAS	No*	Yes
KAE609 (Cmax, AUC, concentration)	PK	Optional	Yes
CCI			

* Yes if the DMC analysis after Cohort 4 is upgraded to an IA.

2.15.2 Stopping criteria for the study

Data related to participant's efficacy and safety will be continuously monitored by the sponsor. If participants randomized to any IV cipargamin arms meet either the safety criteria or lack of efficacy criteria as listed in Table 2-14 below, the study may be paused and a DMC review conducted.

Table 2-14 Criteria to temporarily pause the recruitment and initiate DMC review

Cohorts with planned recruitment	Number of participants meeting the safety criteria*	Number of participants meeting the lack of efficacy criteria**
Cohort with 6 participants	2	2
Cohort with 14 participants	2	2
Cohort with 20 participants	3	3
Cohort with 38 participants	6	6
Throughout the study (116 participants within a single IV cipargamin treatment arm)	14	14

*Safety criteria - All cause death or SAE possibly related to cipargamin (as per sponsor)
**Lack of efficacy criteria – Use of IV artesunate as rescue medication (as per protocol defined criteria) in participants randomized to IV cipargamin

Number and percent of patient meeting with safety criteria and lack of efficacy criteria will be presented by cohort and treatment regimen as well in pooled cohorts 1-5.

3 Sample size calculation

3.1 Primary estimand

The primary objective is to show that an IV cipargamin dose regimen is non-inferior to the IV artesunate in proportion of patients with ≥ 90% *P. falciparum* parasite reduction at 12 hours for each cohort. In the artesunate Phase III studies SEAQUAMAT (Dondorp et al 2005) and AQUAMAT (Dondorp et al 2010), proportion of patients with ≥ 90% *P. falciparum* parasite reduction was not reported for both artesunate and quinine. Therefore, a non-inferiority margin in proportion of patients with ≥ 90% parasite reduction for IV cipargamin vs IV artesunate cannot be derived statistically based on historical data. A non-inferiority margin of 10% is set based on clinical judgement. The KM plot for time to > 90% parasite clearance by (Kremsner et al (2016))indicated that the proportion of patients with 90% parasite reduction at 12 hours was about 50%, 45%, and 40% for artesunate 3 dose IM (n=341), 3 dose IV (n=337), and 5 dose IM (n=333), respectively. Therefore, we assume that the proportion of patients with 90% *P. falciparum* parasite reduction is 45% for IV artesunate. CCI [redacted] Based on these assumptions, a sample size of 13 patients per treatment group will provide 90% power to show that a cipargamin treatment group is non-inferior to artesunate for each cohort in the proportion of patients with ≥ 90% *P. falciparum* parasite reduction at 12 hours using a non-inferiority margin of 10% absolute difference at the 1-sided 5% significance level (nQuery 7.0 Table PTE1a). Considering that 5% of patients that may be excluded from the analysis (Kremsner et al (2016)), at least 14 patients will be randomized to each treatment regimen in each cohort except Cohort 1 to achieve 13 evaluable patients per regimen for the primary analysis. The sample size for Cohort 1 will be 6 patients in each arm, which is not based on the statistical justification (no formal testing will be performed in this Cohort 1) but based on clinical judgment to rule out the risk of any potential untoward safety or lack of efficacy issues in this small group of moderately severe malaria patients before enrolling the severe malaria patients into the study.

3.2 Secondary endpoint(s)

Secondary estimand of clinical success at 48 hours:

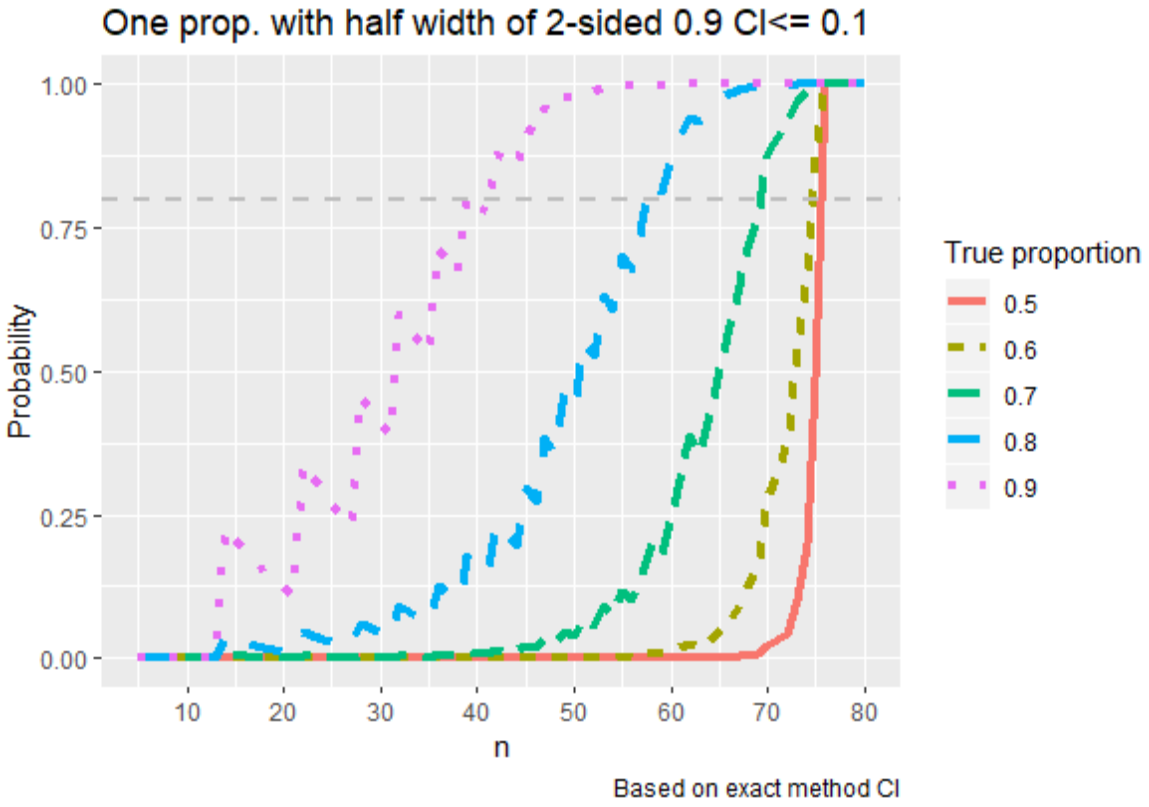
The key secondary estimand of clinical success at 48 hours may be used as a primary efficacy endpoint for the Ph.3 study. The key secondary objective is to estimate clinical success rates for cipargamin and artesunate individually as well as the difference between two treatment groups at 48 hours with good precision. The cipargamin treatment group for this estimation will be the one which is used in all the cohorts across the study; patients from this treatment group will be pooled from all Cohorts. Similarly, for the artesunate group, patients will be pooled from all Cohorts.

The sample size calculation considers the precision of the estimates for the individual treatment groups as well as the difference between the two treatment groups; and the precision is measured in the form of half width of the associated 90% CI as mentioned below.

Precision of the estimates of single proportion

For a binary endpoint, let X be a number of clinical successes with n patients, X follows a binomial distribution $Bin(n, p)$ where p is the true proportion of patients with clinical success for a treatment group at a visit. Proportion (p) is estimated by X/n with a 90% CI based on the exact method; and the half-width of 90% CI is considered as the precision of the estimate. Consider d as the targeted half width of CI and P_l and P_u as lower and upper limits of 90% CI based on X and n (exact method), the probability that the observed half width ($0.5[P_u - P_l]$) is at most the targeted half, ie., $Pr\{0.5(P_u - P_l) \leq d | Bin(n,p))$ will be used to justify the sample size for a single proportion.

Figure 3-1 Plot of probability that half width of 2-sided 90% CIs for a single proportion is at most 0.1 for various true proportions



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For designated half width d, the sample size, n is chosen so that the probability that the half width of 2-sided exact 90% CI for p is at most d is at least 80% at a likely true p.

Figure 3-1 displays the probabilities that the half width of 2-sided exact 90% CIs for a single proportion ≤0.1 for true proportions of 0.5 to 0.9 by 0.1. To maintain the same probability, such as 80% represented in the horizontal dash line, the sample size is higher when the true

proportion is closer to 0.5 and is lower when it is away from the 0.5. For instance, when p =0.5, n=76 is required to estimate the true proportion with at least 80% probability that the half width of 2-sided exact 90% CI is ≤10%. Similarly, when p =0.7, n=70 is required to estimate the true proportion with at least 80% probability that the half width of 2-sided exact 90% CI is ≤10%.

Precision of the estimates of difference between two proportions

For estimating the difference between two proportions, the half width of 2-sided 90% CIs calculated using Wilson's uncorrected method is considered as the precision of the estimation. The probability that the half width of 2-sided 90% CI for difference between 2 proportions is at most a targeted width (d) at likely proportions will be considered for the sample size selection.

Figure 3-2 displays the probability that the half width of Wilson's uncorrected 2-sided 90% CIs for the difference between 2 proportions is most 0.1 for true equal proportions of 0.5, 0.6, 0.7, 0.75, 0.8, and 0.9. To maintain the same probability, such as 80% represented in the horizontal dash line, the sample size is higher when the two equal true proportions are closer to 0.5. For instance, when p =0.5 for both treatment groups, n=133 is required in order to have at least 80% probability that the half width of 2-sided 90% CIs for the difference is at most 0.1. Similarly, when p =0.75 for both treatment groups, n=107 is required in order to have at least 80% probability that the half width of 2-sided 90% CI for the difference is at most 0.1.

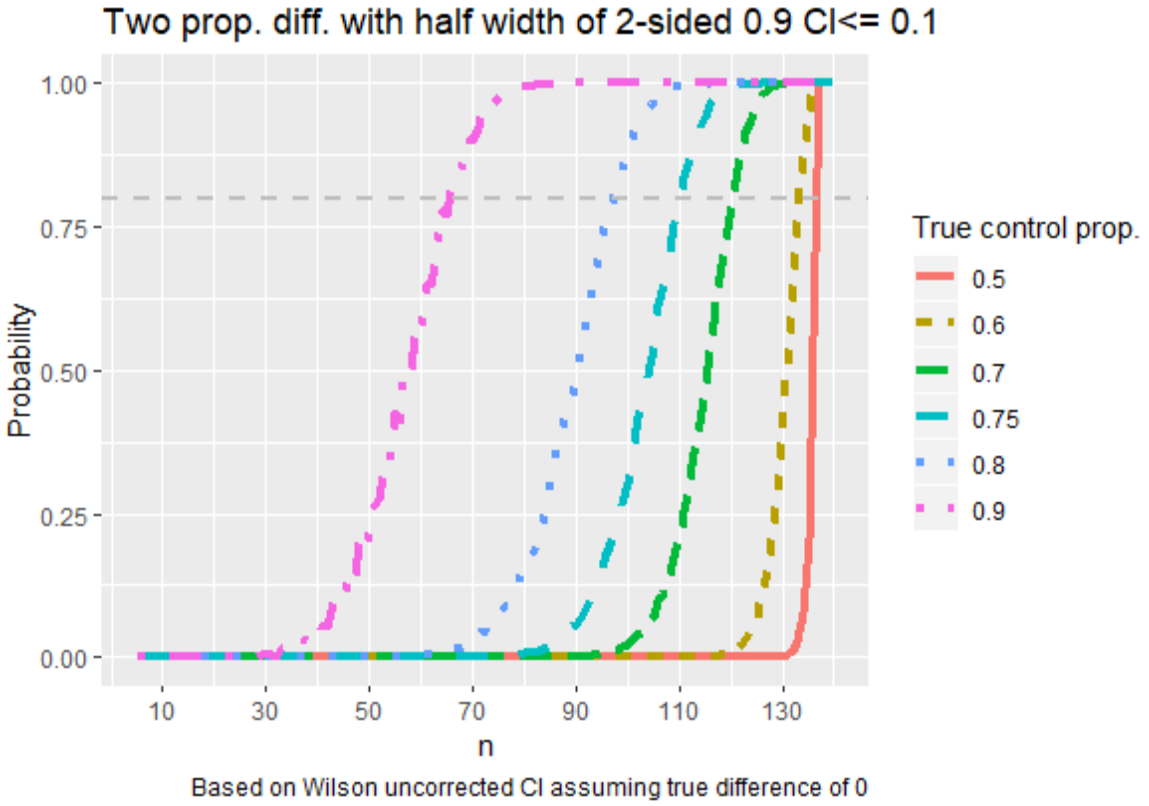
Compared with Figure 3-1, Figure 3-2 shows that the required sample size for the precision in estimating the difference between 2 proportions is higher than the corresponding sample size required for the precision in estimating a single proportion. Therefore, the sample size for the study will be justified based on the half width of 2-sided 90% CIs for the difference between 2 proportions.

The proportion of patients who were alive without new/worsening complication at the end of study for artesunate was 0.679 in SEQUAMAT and 0.791 in AQUAMAT (private communication). Therefore, it is estimated that the proportion of patients meeting the proposed clinical endpoint is about 0.75 in the artesunate arm. To date, cipargamin has not been tested in patients with severe malaria. It is assumed that the proportion in the cipargamin regimen would be same as that of artesunate.

From Figure 3-2, a sample size of n=107 per regimen is required in order to have at least 80% probability that the half width of 2-sided 90% CIs for the difference between 2 proportions is at most 0.1 if the true proportion is 0.75 for both treatment groups. As patients treated with the same treatment from all cohorts will be pooled together for the analysis using the FAS, this sample size is considered for the artesunate and the cipargamin dose regimen that is used in all cohorts.

Taking account that about 8% patients may be excluded from the FAS, patients from all cohorts will be randomized to yield about 116 randomized each for the artesunate and the cipargamin dose regimen that is used in all cohorts so that 107 patients per group are evaluable for this analysis. The enrollment may be increased if the number of patients excluded from the FAS is higher. Considering the required sample size for each cohort for the primary endpoint (see Section 3.1) and the potential patient population available for recruitment, the 116 randomized for a treatment group will split into 6 patients for Cohort 1, 14 patients for Cohort 2, 38 patients for Cohorts 3 and 4, and 20 patients for Cohort 5.

Figure 3-2 Plot of probability that half width of 2-sided 90% CIs for 2 proportion difference is at most 0.1 for various true equal proportions



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Safety events

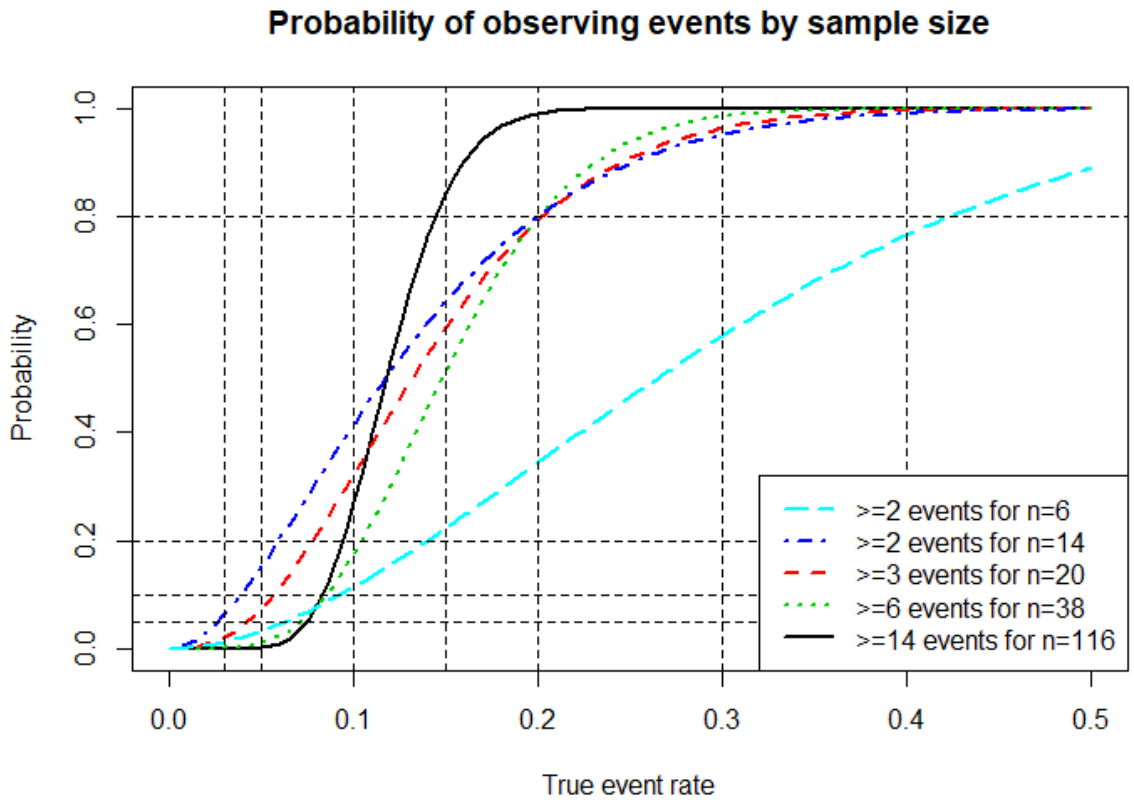
Safety events will be evaluated using the safety set, which is expected to similar to the randomized set. Occurrence of a safety event for a regimen (cipargamin or artesunate) will be evaluated by cohort and overall with all patients pooled from all cohorts. The probability of observing selected minimal number of events is displayed in Figure 3-3 by sample size. From this plot,

- for a treatment group in a cohort with 6 patients, the probability of observing at least 2 events is > 75% if true event rate is ≥ 40% but < 20% if true event rate is ≤ 10%
- for a treatment group in a cohort with 14 patients, the probability of observing at least 2 events is about 80% if the true event rate is ≥ 20% but < 20% if the true event rate is ≤ 5% ;
- for a treatment group in a cohort with 20 patients, the probability of observing at least 3 events is about 80% if the true event rate is ≥ 20% but < 20% if the true event rate is ≤ 7%
- for a treatment group in a cohort with 38 patients, the probability of observing at least 6 events is about 80% if the true event rate is ≥ 20% but < 20% if the true event rate is ≤ 10%

- for a regimen that is selected for Cohorts 3-5 with 116 patients, the probability of observing at least 14 events is > 80% if the true event rate is ≥ 15% but < 20% if the true event rate is ≤ 9%.

The minimal number of events mentioned above may be used a guidance to interpret the occurrence of safety events in a treatment group in a cohort or pooled cohorts.

Figure 3-3 Probability of observing selected minimal number of events by sample size



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4 Change to protocol specified analyses

None

5 Appendix

5.1 Neurological examination

Detailed neurological examination of the patient will be done and relevant medical history collected under following categories to assess the extent of neurological signs and symptoms at baseline and monitor extent of neurological sequelae in the follow up visits. The neurological assessments include Consciousness, Cranial Nerve Palsy , Motor system , Convulsions, and sense organs (vision, hearing, speech).

The determination of coma scores, which are part of neurological assessments, are described in Tables 5-1 and 5-2.

Table 5-1 Blantyre Coma Score (for patients ≤ 5 years)

Eye Opening (E)	Score	Verbal Response (V)	Score	Best Motor Response (M)	Score
Able to follow the moving object	1	Normal cry or appropriate speech (for patients who are old enough to talk) in response to painful stimuli	2	Localize pain stimulus	2
Unable to follow the moving object	0	Moan or an abnormal cry in response to painful stimuli	1	Withdraw limb from pain stimulus	1
-	-	No cry with painful stimuli	0	No response or inappropriate response to pain stimulus	0

Table 5-2 Glasgow Coma Scale (for patients > 5 years)

Eye Opening (E)	Score	Verbal Response (V)	Score	Best Motor Response (M)	Score
Spontaneous	4	Oriented	5	Obeying Commands	6
To speech	3	Confused	4	Localizing	5
To pressure	2	Words	3	Normal flexion	4
None	1	Sounds	2	Abnormal flexion	3
-	-	None	1	Extension	2
-	-	-	-	None	1

Note- Individual deficits that are found during neurological assessments are recorded in the clinical database and will be mapped to the corresponding scores according to Table 5-1 and Table 5-2 by SAS programming.
Detailed methodology for each of the neurological assessments will be provided in a neurological assessment manual and the relevant site staff will be trained prior to the start of the study to maintain uniform standard of neurological examination across the study sites.

5.1.1 Grading of neurological sequelae

Grading for individual domains:

Individual deficits for each neurological functional domain (motor, vision, hearing and speech) will be recorded on the CRF according to Table 5-3. These deficits are pre-classified as moderate severe grades as per Table 5-3. For e.g. if a patient is recorded with deficit of Monoplegia/paresis then grading of this deficit is moderate or if a patient is recorded with deficit of cranial nerve palsies then grading of this deficit is severe. Because the grading of individual deficits are fixed, these will not be captured in the clinical database but will be mapped to the their respective severity grades (moderate/ severe) according to Table 5-3 at the time of statistical analysis.

Table 5-3 Grading of Neurological Sequelae

S.No	Neurological functional domain	Graded as Moderate	Graded as Severe
1	**Motor**		
a)	Monoplegia/paresis	X	
b)	Hemiplegia/paresis		X
c)	Quadriplegia/paresis		X
d)	Continued posturing		X
e)	Cerebellar Ataxia		X
f)	Cranial nerve palsies	X	
2	**Vision**		
a)	Blindness bilateral		x
b)	Blindness unilateral	X	
c)	Bilateral vision impairment	X	
d)	Unilateral vision impairment	X	
3	**Hearing and Speech**		
a)	Bilateral deafness		x
b)	Unilateral deafness	X	
c)	Bilateral hearing impairment	X	
d)	Unilateral hearing impairment	X	
e)	Speech difficulties	X	

S.No	Neurological functional domain	Graded as Moderate	Graded as Severe
f)	Unable to speak		X
4	**Convulsion**		
a)	Any convulsion In patient with no history of convulsion	X	
b)	Status epilepticus		X

Overall grading of neurological sequelae:

Overall grading for neurological sequelae will be derived using individual grading as mentioned above.

Following algorithm will be used to determine the overall grading of the neurological sequelae.

- A patient with at least 1 severe deficit in any functional domain, the overall grading will be severe.
- A patient with at least 1 moderate deficit in 2 or more functional domains, the overall grading will be severe.
- A patient with at least 1 moderate deficit in any one functional domain, the overall grading will be moderate.
- In case the medical history CRF mentioned a pre-existing neurological problem and there is no significant deterioration of symptoms during the malaria episode, the neurological problems will not be considered as being sequelae of the acute disease episode. This evaluation will be performed by the Adjudication committee.

5.2 Imputation rules

5.2.1 Study drug

The study drug administration date should be complete since it's taken in the hospital. In case missing or partial, the visit date will be used as the study drug administration date.

5.2.2 AE date imputation

The following missing dates will not be imputed

- Missing AE start dates
- AE start dates missing the year
- Partial/missing AE end dates

For other type of partial missing start dates, rules specified in Tables 5-5 to Table 5-6 will be used

Table 5-4 AE/Treatment Date Abbreviations

	Day	Month	Year
Partial Adverse Event Start Date	<not used>	AEM	AEY
Treatment Start Date (TRTSTD)	<not used>	TRTM	TRTY

Table 5-7 describes the possible combinations and their associated imputations. The upper text indicates the imputation (NC, A, B, C etc.) and the lower text the relationship of the AE start date to the treatment start date (TRTSTD).

Table 5-5 Imputation algorithm

	AEM MISSING	AEM < TRTM	AEM = TRTM	AEM > TRTM
AEY MISSING	NC Uncertain	NC Uncertain	NC Uncertain	NC Uncertain
AEY < TRTY	(D) Before TRTSTD	(C) Before TRTSTD	(C) Before TRTSTD	(C) Before TRTSTD
AEY = TRTY	(B) Uncertain	(C) Before TRTSTD	(B) Uncertain	(A) After TRTSTD
AEY > TRTY	(E) After TRTSTD	(A) After TRTSTD	(A) After TRTSTD	(A) After TRTSTD

The legend to the above table is shown in Table 5-6.

Table 5-6 Imputation algorithm legends

Relationship	
Before TRTSTD	Indicates AE start date prior to Treatment Start Date
After TRTSTD	Indicates AE start date after Treatment Start Date
Uncertain	Insufficient to determine the relationship of AE start date to Treatment Start Date
Imputation calculation	
NC / Blank	No convention/imputation
(A)	01MONYYYY
(B)	TRTSTD+1
(C)	15MONYYYY
(D)	01JULYYYY
(E)	01JANYYYY

Few examples are shown in Table 5-7.

Table 5-7 Example scenarios

Partial AE start date	Treatment start date	Relationship with TRTSTD	Imputation Calculation	Imputed Date
12mmyyyy	20OCT2001	Uncertain	NC	<blank>
ddmmm2000	20OCT2001	Before	(D)	01JUL2000
ddmmm2002	20OCT2001	After	(E)	01JAN2002
ddmmm2001	20OCT2001	Uncertain	(B)	21OCT2001
ddSEP2001	20OCT2001	Before	(C)	15SEP2001
ddOCT2001	20OCT2001	Uncertain	(B)	21OCT2001
ddNOV2001	20OCT2001	After	(A)	01NOV2001

5.2.3 Concomitant medication date imputation

Missing concomitant medication dates will be imputed similar as to AE dates.

5.2.3.1 Other imputations

NA

5.2.4 Visit windows

When visit windows are used, all visits will be re-aligned, i.e., they will be mapped into one of the visit windows. E.g., if the Day 4 visit of a patient is delayed and occurs on Day 7, say, it will be re-aligned to visit window Day 8. In the case of major deviations from the visit schedule, or due to unscheduled visits, several assessments of a patient may fall in a particular visit window (either scheduled or unscheduled). Statistical approaches to handle multiple assessments in a given visit window are specified below.

- Of note, patients are allowed to have gaps in visits. All data collected will be displayed in listings.
- The following rules are used to determine the window for other visits post baseline:
  - "Lower limit" = "upper limit of prior applicable visit" + 1.
  - "Upper limit" = "target day of current visit" + integer part of ("target day of next applicable visit" – "target day of current visit")/2
  - No upper limit for Day 29 visit to include all assessments

For assessments that are scheduled to be performed only once on a day, Table 5-8 describes the analysis windows mapping to visits (not just scheduled visits) based on study days alone. For the assessments that may be performed on multiple timepoints on a day, Table 5-9 describes the analysis windows mapping to visits based on study day and time. Repeat and/or unscheduled visits (which will be numbered in the database according to new NCDS standards) will be mapped for analysis purposes in the same way. If there are multiple measurements within an analysis window, the conventions defined in Table 5-10 will be used to determine the appropriate measurement to be selected for analysis.

The mapped visits will be used in the by visit analyses. However, the listings will show all collected data regardless of used in the by visit analyses.

Table 5-8 Analysis visit windows based on study days alone

Analysis Visit	Target Day	Analysis visit window for assessment group						
		ECG	Body weight[1]	Body height/Head circumference/ body weight[2]	INR/LDH	CCI	AAG	CCI
Baseline	1	Up to Day 1	Up to Day 1	Up to Day 1	Up to Day 1		Up to Day 1	
Day 2	2	Day 2	Day 2-15	NA	NA		Day 2	
Day 3	3	Day 3	NA	NA	NA		Day 3 and above	
Day 4	4	Day 4 to Day 16	NA	NA	NA		NA	
Day 6	6	NA	NA	NA	NA		NA	
Day 8	8	NA	NA	NA	Day 2 and above		NA	
Day 15	15	NA	NA	NA	NA		NA	
Day 22	22	NA	NA	NA	NA		NA	
Day 29/ End of study	29	Day 17 and above	Day 16 and above	Day 2 and above	NA		NA	

[1] Cohorts 1-3 [2] Cohorts 4-5

Table 5-9 Analysis visit windows based on study day and time

Target visit	Target timepoint	Blood smear	PK (Cohorts 1 to 3)	PK (Cohorts 4-5)	Neurological/severe malaria assessment/ body temperature/blood pressure/pulse	Blood chemistry/hematology Point of Care device	Urinalysis
Baseline	0 hrs	up to 0 hrs	up to 0 hrs	NA	up to 0 hrs	up to 0 hrs	up to 0 hrs
Day 1	2 m	NA	>0 – 6m	>0 – 31 min	NA	NA	NA
	10 m	NA	>6m – 35 m	NA	NA	NA	NA
	30 m	NA	NA	NA	>0 – 3.25 hrs[1]	NA	NA
	1 hrs	>0 - 1.5 hrs	>35 m- 2.5 hrs	>31 min-12.5 hrs	NA	NA	NA
	2 hrs	>1.5 – 3 hrs	NA	NA	NA	NA	NA
	4 hrs	>3 – 5 hrs	>2.5 – 8 hrs	NA	NA	NA	NA
	6 hrs	>5 - 9 hrs	NA	NA	>3.25-9 hrs	> 0 – 9 hrs[2]	NA
	12 hrs	>9 - 15 hrs	>8- 18 hrs	NA	>9 - 15 hrs	>9 – 18 hrs	NA
	18 hrs	>15 - 21 hrs	NA	NA	>15 - 21 hrs	NA	NA
Day 2	24 hrs	>21 – 27 hrs	>18 – 24 hrs 1 min	>12.5 hrs – 36 hrs	>21 – 30 hrs	>18 -30 hrs	>0 – 30 hrs
	24 hrs 2 min	NA	>24 hrs 1 min-24 hrs 31 min	NA	NA	NA	NA
	25 hrs	NA	>24 hrs 31 min– 30.5 hrs	NA	NA	NA	NA
	30 hrs	>27 – 33 hrs	NA	NA	NA	NA	NA
	36 hrs	>33 – 39 hrs	>30.5 –42 hrs	NA	>30 – 42 hrs	>30 – 42 hrs	>30 -42 hrs

Target visit	Target timepoint	Blood smear	PK (Cohorts 1 to 3)	PK (Cohorts 4-5)	Neurological/severe malaria assessment/ body temperature/blood pressure/pulse	Blood chemistry/hematology Point of Care device	Urinalysis
	42 hrs	>39- 45 hrs	NA	NA	NA	NA	NA
	48 hrs	>45 – 60 hrs	m>42 – 48 hrs 1 min	>36 hrs	>42 -60 hrs	>42 -60 hrs	>42 – 60 hrs
	48 hrs 2 min	NA	>48 hrs 1 min – 48 hrs 31 min	NA	NA	NA	NA
	49 hrs	NA	>48 hrs 31 min – 54.5 hrs	NA	NA	NA	NA
Day 3 e	60 hrs	NA	>54.5 – 66 hrs	NA	NA	NA	NA
Day 4	72 hrs	>60 hrs to Day 5	>66 – 96 hrs	NA	>60 hrs to Day 5	>60 hrs to Day 5	>60 hrs to Day 5
Day 6	120 hrs	Day 6 – 7	>96 hrs – 144 hrs	NA	Day 6 – 7	Day 6 – 7	Day 6 -7
Day 8	168 hrs	Day 8 - 11	>144 hrs	NA	Day 8 - 11	Day 8 – 11	Day 8 -11
Day 15	336 hrs	Day 12 - 18	NA	NA	Day 12 - 18	Day 12 – 18	Day 12 - 18
Day 22	504 hrs	Day 19 - 25	NA	NA	Day 19 - 25	Day 19 - 25	Day 19 - 25
Day 29	672 hrs	Day 26 and above	NA	NA	Day 26 and above	Day 26 and above	Day 26 and above

[1] for body temperature and vital signs; [2] for Point of Care device

Table 5-10 Rules for flagging variables

Timing of measurement	Type of data	Rule
Baseline	All data	The last measurement made prior to administration of the first dose of study treatment – note this may include measurements taken on the day of randomization. If a patient did not receive any dose of study treatment then the randomization date will be used.
Post-baseline efficacy	parasite count and temperature	The measurement closest to the target day/time will be used. In the event two measurements are taken equally apart, the first one will be used.
Post-baseline safety	Summary visit information (e.g. lab, ECG, etc.)	The measurement closest to the target day/time will be used. In the event two measurements are taken equally apart, the first one will be used.
Post-baseline safety	Notable abnormalities (e.g. lab, ECG, VS)	The most extreme measurement in the window will be used. Note this means a patient can have a notably high and notably low measurement within a window

5.3 Non-study drug antimalarials

Table 5-11 Table of non-study drug antimalarials with respective ATC codes

Non-study drug antimalarials	ATC code	ATC name
Aminoquinolines	P01BA01	Chloroquine
	P01BA02	Hydroxychloroquine
	P01BA03	Primaquine
	P01BA06	Amodiaquine
Biguanides	P01BB01	Proguanil
	P01BB02	Cycloguanil embonate
	P01BB51	Proguanil, combinations
Methanolquinoline	P01BC01	Quinine
	P01BC02	Mefloquine
Diaminopyridimidines	P01BD01	Pyrimethamine
	P01BD51	Pyrimethamine, combinations
Artemisinin and derivatives, plain	P01BE01	Artemisinin
	P01BE02	Artemether
	P01BE03	Artesunate
	P01BE04	Artemotil
	P01BE05	Artenimol
Artemisinin and derivatives, combinations	P01BF02	Artesunate and mefloquine
	P01BF03	Artesunate and amodiaquine
	P01BF04	Artesunate, sulphamethopyrazine and pyrimethamine
	P01BF05	Artenimol and piperaquine
	P01BF06	Artesunate and pyronaridine
Other Antimalarials	P01BX01	Halofantrine
	P01BX02	Arterolane and Piperaquine

5.4 Medications for management severe malaria

Medications that are used for management of severe malaria will be identified from the clinical database before DBL.

5.5 AEs coding/grading

Adverse events are coded using the Medical dictionary for regulatory activities (MedDRA) terminology. AEs are assessed by investigators according to the most current Common Terminology Criteria for Adverse Events (CTCAE) version 5.0.

5.6 Laboratory parameters derivations

Grade categorization of lab values will be assigned programmatically as per NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 (specify version used in the RAP). The calculation of CTCAE grades will be based on the observed laboratory values only, clinical assessments will not be taken into account. The criteria to assign CTCAE grades are given in Novartis internal criteria for CTCAE grading of laboratory parameters. The latest available version of the document based on the underlying CTCAE version 5.0 at the time of analysis will be used.

A severity grade of 0 will be assigned for all non-missing lab values not graded as 1 or higher. Grade 5 will not be used. For laboratory tests that are graded for both low and high values, summaries will be done separately and labelled by direction, e.g., sodium will be summarized as hyponatremia and hypernatremia.

5.7 Computation of Blood Pressure Percentiles for Arbitrary Sex, Age, and Height

Below algorithm is described in the Appendix B of the following reference: The Fourth Report on Diagnosis, Evaluation and Treatment of High Blood Pressure in Children and Adolescents. Pediatrics 2004; 114; 555.

- To compute the systolic blood pressure (SBP) percentile of a boy whose age is y years and height =h inches with SBP = x mmHg:
    - Convert the height of *h* inches to a height Z-score relative to boys of the same age (as per data tables provided Table 5-14; this is denoted by Z_{ht}. For age < 2 years, age is recorded in the CRF with month as the unit and will be converted into year with 1 decimal, the corresponding height Z score can be calculated using months (See Table 5-13).
    - Compute the expected SBP (μ) for boys of age *y* years and height *h* inches given by

$$\mu = \alpha + \sum_{j=1}^{4} \beta_j (y - 10)^j + \sum_{k=1}^{4} \gamma_k (Z_{ht})^k$$

where, regression coefficients are given in the 3rd column of Table 5-12

- Then convert the boy's observed SBP (x) to a Z-score (Z_{BP}) given by

$$Z_{BP} = \frac{(x - \mu)}{\sigma}$$

where, σ is given in the 3rd column for standard deviation row of Table 5-12

- To convert the bp Z-score to a percentile (P), compute P = ϕ (Z_{BP}) x 100% where ϕ (Z) = area under a standard normal distribution to the left of Z.

- Likewise, to compute percentiles for SBP for girls, diastolic blood pressure (DBP) (K5) for boys, and DBP (K5) for girls, use the regression coefficients from the 4th, 5th, and 6th columns of Table 5-12.

For example, a 12-year-old boy, with height at the 90th percentile for his age-sex group, has a height Z-score = 1.28, and his expected SBP (μ) is

μ = 102.19768 + 1.82416 (2) + 0.12776 (2^2) + 0.00249 (2^3) –0.00135 (2^4) + 2.73157 (1.28) –0.19618 (1.28^2) –0.04659 (1.28^3) + 0.00947(1.28^4) = 109.46 mmHg.

Table 5-12 Regression coefficients from Blood Pressure Regression Models

Regression Coefficients From Blood Pressure Regression Models*

Variable Name	Symbol	Systolic BP		Diastolic BP5	
		Male	Female	Male	Female
Intercept	α	102.19768	102.01027	61.01217	60.50510
Age					
Age–10	β_1	1.82416	1.94397	0.68314	1.01301
(Age–10)^2	β_2	0.12776	0.00598	-0.09835	0.01157
(Age–10)^3	β_3	0.00249	-0.00789	0.01711	0.00424
(Age–10)^4	β_4	-0.00135	-0.00059	0.00045	-0.00137
Normalized height					
Zht	γ^1	2.73157	2.03526	1.46993	1.16641
Zht^2	γ^2	-0.19618	0.02534	-0.07849	0.12795
Zht^3	γ^3	-0.04659	-0.01884	-0.03144	-0.03869
Zht^4	γ^4	0.00947	0.00121	0.00967	-0.00079
Standard deviation	σ	10.7128	10.4855	11.6032	10.9573
ρ†		0.4100	0.3824	0.2436	0.2598
n (persons)		32,161	31,066	24,057	23,443
n (*visits*)		*42,074*	*41,017*	*29,182*	*28,794*

BP, blood pressure; Diastolic BP5, diastolic measurement at Korotkoff 5.

* The coefficients were obtained from mixed-effects linear regression models.

† The value of ρ represents the correlation between BP measurements at different ages for the same child after correcting for age and Zht. This computation was necessary because some studies contributing to the childhood BP database provided BP at more than one age.

Computation of Z-score for height:

As per the recommendation from Centers for Disease Control and Prevention (CDC) use WHO growth charts for 0 to < 2 years of age and CDC growth charts for age ≥2 years. Accordingly, convert the given height into corresponding Z-score as per Table 5-13 and Table 5-14 given for the growth standards using the following formula.

$$Z_{ht} = \frac{(\frac{height}{median})^{L} - 1)}{L * S}$$

Where, median, L, S (i.e. CV) are provided in the Table 5-13 for age up to 2 years and in Table 5-14 for age 2-20 years; height in centimeters.

Table 5-13 Table for WHO standard Length-for-age growth chart for age 0 to to 2 years

		Boy		Girl	
Month	L	Median (cm)	S (CV)	Median (cm)	S (CV)
0	1	49.8842	0.03795	49.1477	0.0379
1	1	54.7244	0.03557	53.6872	0.0364
2	1	58.4249	0.03424	57.0673	0.03568
3	1	61.4292	0.03328	59.8029	0.0352
4	1	63.886	0.03257	62.0899	0.03486
5	1	65.9026	0.03204	64.0301	0.03463
6	1	67.6236	0.03165	65.7311	0.03448
7	1	69.1645	0.03139	67.2873	0.03441
8	1	70.5994	0.03124	68.7498	0.0344
9	1	71.9687	0.03117	70.1435	0.03444
10	1	73.2812	0.03118	71.4818	0.03452
11	1	74.5388	0.03125	72.771	0.03464
12	1	75.7488	0.03137	74.015	0.03479
13	1	76.9186	0.03154	75.2176	0.03496
14	1	78.0497	0.03174	76.3817	0.03514
15	1	79.1458	0.03197	77.5099	0.03534
16	1	80.2113	0.03222	78.6055	0.03555
17	1	81.2487	0.0325	79.671	0.03576
18	1	82.2587	0.03279	80.7079	0.03598
19	1	83.2418	0.0331	81.7182	0.0362
20	1	84.1996	0.03342	82.7036	0.03643
21	1	85.1348	0.03376	83.6654	0.03666
22	1	86.0477	0.0341	84.604	0.03688
23	1	86.941	0.03445	85.5202	0.03711

Reference:

https://www.cdc.gov/growthcharts/who/boys_length_weight.htm

https://www.cdc.gov/growthcharts/who/girls_length_weight.htm

Table 5-14 Table for CDC growth standard height-for-age growth charts for age 2 -20 years

	Boy			Girl		
Age(year)	L	Median (cm)	S (CV)	L	Median (cm)	S (CV)
2	0.941524	86.4522	0.040322	1.072449	84.97556	0.040791
3	-0.39092	95.27359	0.040534	0.541981	94.21336	0.042018
4	0.827637	102.5105	0.041344	0.225706	101.0339	0.04326
5	1.266367	109.1751	0.042593	-0.05773	107.9566	0.044277
6	1.137443	115.6609	0.043673	-0.21907	115.0055	0.044964
7	0.753244	122.0305	0.044403	-0.21021	121.7617	0.045461
8	0.455268	128.1237	0.045127	-0.07928	127.8263	0.045968
9	0.415687	133.7345	0.046217	0.084148	133.1304	0.046884
10	0.505564	138.8234	0.04761	0.284749	138.2112	0.048705
11	0.487939	143.7304	0.048938	0.74429	144.2609	0.050524
12	0.420919	149.3088	0.049948	1.303045	151.4866	0.048599
13	0.81624	156.4099	0.050333	1.242968	157.3437	0.043859
14	1.670433	164.1418	0.048945	0.956572	160.4777	0.041022
15	2.20518	170.1393	0.04589	0.89557	161.898	0.040084
16	2.113023	173.6101	0.043086	0.941146	162.569	0.039821
17	1.724738	175.341	0.041408	0.999506	162.9238	0.039732
18	1.399999	176.185	0.040644	1.047571	163.1308	0.039687
19	1.229163	176.6179	0.040391	1.083315	163.259	0.039657
20	1.167279	176.8492	0.04037	1.108046	163.3383	0.039636

Reference: https://www.cdc.gov/growthcharts/data/zscore/statage.csv

5.8 Statistical models

5.8.1 Primary analysis

The primary analysis of parasite clearance rate ≥ 90% reduction at 12 hours is defined in Section 2. The between treatment difference will be evaluated using a Wilson uncorrected method using PROC FREQ with RISKDIFF (CL= NEWCOMBE) option in the TABLES statement.

5.8.2 Key secondary analysis

The key secondary analysis of clinical success at 48 hours is defined in Section 2.

SAS procedure FREQ with EXACT statement for one-way tables will be used to estimate the proportion of responders (binary outcome = 1 or “Yes”), along with the associated 95% (=100 × (1 – *two-sided alpha level*)) two-sided Clopper- Pearson CI.

Between treatment difference will be evaluated using a Mantel-Haenszel estimate of the treatment difference in responder rates stratified by cohort (Cohorts 1 and 2 pooled) using PROC FREQ with RISKDIFF(COMMON) option in the TABLES statement.
Clinical success at other timepoints will be analyzed similarly.

5.8.3 Other secondary/exploratory analysis

Kaplan-Meier estimates

An estimate of the survival function in each treatment group will be constructed using Kaplan-Meier (product-limit) method as implemented in PROC LIFETEST with METHOD=KM option. The PROC LIFETEST statement will use the option CONFTYPE=LOGLOG.

Median survival for each treatment group will be obtained along with 95% confidence intervals calculated from PROC LIFETEST output using the method of (Brookmeyer, Crowley 1982). Kaplan-Meier estimates of the survivor function with 95% confidence intervals at specific time points will be summarized. The standard error of the Kaplan-Meier estimate will be calculated using Greenwood's formula (Collett 1994).

Difference in survival rate using log-rank test

Between treatment difference will be evaluated using a log-rank test from PROC LIFETEST with STRATA <treatment > with test=logrank option and TEST <treatment > statements. For pooled time to event data from cohort 1 and 2, stratified log-rank test will be performed using STRATA <cohort> statement with GROUP = <treatment> option.

Confidence interval for geometric mean ratio

For PK parameters, such as $AUC_{0\text{-}24h}$, Cmax, the values for individuals will be converted into $\log_e$ scale. Mean and corresponding 2-sided 100*(1-α)% confidence intervals (CI) will be calculated using PROC TTEST by assuming normality of data on log scale. The geometric mean, the lower and upper confidence limits will be calculated by exponentiating the arithmetic means, and lower and upper confidence limit that were derived using log data.

5.8.4 Sensitivity analysis for between treatment difference in clinical success using multiple imputation

The difference in clinical success between the two treatment groups at different visits (24 hours, 48 hours, 72 hours, hospital discharge , Day 6, Day 8, Day 15, Day 22, and Day 29) will be evaluated using Wilson's uncorrected method for individual cohorts and Mental-Hanszel estimate for pooled cohorts as described in Section 2.7 with multiple imputations as sensitivity analysis for missing clinical endpoint not due to death related to malaria (missing due to death not related malaria or missing due to lost to followup) after the hospital discharge and discontinuation of study.

The feasibility of following plan will be tested using blinded data at CSR dryrun and may be revised later if necessary.

Clinical success is a dichotomous composite (comprised of malaria related death, *P. falciparum* asexual parasites count, and major clinical signs of severe malaria) variable which is derived according to Section 2.1.1 and restated below.

A patient is said to have achieved the clinical success at a given timepoint if the patient did not die either during the initial hospitalization nor due to malaria post the initial hospitalization, shows absence of *P. falciparum* asexual parasites (using local lab for all cohorts and repeated for central lab for Cohorts 1 and 2), and did not have any of the key signs or symptoms of severe malaria on that given timepoint. Note: Deaths occurring post initial hospitalization will be evaluated by designated adjudication committee to confirm if they are related to malaria.

In the current study, Blood film for microscopic counts of *P. falciparum* asexual parasite are collected at screening, pre-dose, Hours 1, 2, 4, 6, 12, 18, 24, 30, 36, 42, 48, 72 (hospital discharge), Days 6, 8, 15, 22, 29, and unscheduled visit. Neurological assessments are performed on screening, pre-dose, Hours 6, 12, 18, 24, 36 48, 72 (hospital discharge), Days 6, 8, 15, 22, 29, and unscheduled visit. Altered consciousness will be determined using these neurological assessments as defined in Section 2.1.1. Plasma lactate and serum creatinine assessments are performed on screening, Hours 6, 12, 18, 24, 36 48, 72 (hospital discharge), Days 6, 8, 15, 22, 29, and unscheduled visit. Note: if hospital discharge does not occur at Hour 72 or a scheduled visit, these assessments should be performed at hospital discharge. Renal replacement therapy (RRT) will be captured on concomitant and non-drug therapy CRF page. These assessments are required for determination of renal impairment. Respiratory distress is recorded by investigator in the severe malaria CRF and will also be identified using adverse event pages.

Key signs of Severe Malaria are (a) Altered consciousness, (b) Renal impairment, (c) Acidosis. These signs are defined below.

Altered consciousness – Prostration (inability to sit or drink or breast feed) or GCS < 11 for patients > 5 years / BCS < 3 for patients ≤ 5 years of age. Prostration, GCS/ BCS is recorded by investigator in the relevant CRF. In case that a patient completes the visit but the data are missing, this will be determined to be 'yes' if a patient has an AE of "Altered state of consciousness" with starting day since previous visit to the current visit and 'no' otherwise.

Renal Impairment - Serum creatinine > 3xULN or > 3 mg/dL or need for renal replacement therapy (RRT). Serum creatinine is recorded in the point of care devices (POC) or using local lab in case of non-availability of POC and renal replacement is recorded by investigator in the concomitant CRF. In case that a patient completes the visit but the serum creatinine is missing, this will be determined to be 'yes' if a patient has an "renal replacement" concomitant with starting day since previous visit to the current visit and 'no' otherwise.

Acidosis - Plasma lactate ≥ 4 mmol/L (In database, lactate is captured under lactic acid) or Respiratory distress. Plasma lactate is recorded in the point of care devices (POC) or using local lab in case of non-availability of POC and respiratory distress is recorded by investigator in the severe malaria CRF. In case that a patient completes the visit but the data are missing, this will be determined to be 'yes' if a patient has an AE of "Respiratory distress" with starting day since previous visit to the current visit and 'no' otherwise.

Single imputation rule:

Following single imputations will be performed before applying the multiple imputations:

After a patient has parasite clearance, any intermediate missing parasite between 2 negative parasite assessments is imputed as negative parasite assessment since it is very likely that the intermediate missing should also be negative as prior and post missing data show negative result.

Once a patient died before hospital discharge or is declared as malaria related death at a timepoint, the patient will be considered as not achieving the clinical success for the timepoint onwards.

If a patient has parasite reappearance after the hospital discharge, the patient is considered as not achieving the clinical success for the timepoint onwards.

If a patient receives IV antimalarial rescue medication prior to hospital discharge, the patient is considered as not achieving the clinical success for the timepoint onwards.

Other imputations that are specified in the previous sections will be performed prior to multiple imputation.

Assumptions considered

Since the parasite count data is usually skewed (N Alexander 2012), it is assumed to follow log-normal distribution.

Since the control is standard of care (SoC), data from control group should not be used to impute the missing data for the test group and vice versa to avoid the potential bias. Therefore, imputation technique such as 'Jump to reference' or 'Copy to reference' cannot be used since under these techniques, model uses the data from the reference group while imputing the missing values in the test group and vice versa. Instead, for each treatment group, missing values will be imputed separately using missing at random (MAR) assumption (Carpenter, Kenward 2013). Also, since the patients will be remained hospitalized during the IV treatment period, there is a less chance of missing on-treatment data.

The 0 parasite counts (below assay detection limit) will be first imputed with the detection limit before transforming into the log since log does not exist for 0.

Description of MI procedure

Multiple imputation (MI) is a simulation based approach where missing values are replaced by multiple Bayesian draws from the conditional distribution of missing data given the observed data and covariates, creating multiple completed data sets. These completed data sets can then be analyzed using standard methods and software for balanced data. (Rubin (1987)) presented rules how to combine the multiple sets of estimates to produce overall estimates and confidence intervals that adequately incorporate missing data uncertainty.

FCS method

Fully conditional specification (FCS) method is used to impute the missing data. FCS method uses a separate conditional distribution for each imputed variable. The FCS statement uses a multivariate imputation by chained equations method to impute values for a data set with an arbitrary missing pattern, assuming a joint distribution exists for the data. A FCS method does

not start with an explicitly specified multivariate distribution for all variables, but rather uses a separate conditional distribution for each imputed variable. For each imputation, the process contains two phases: the preliminary filled-in phase followed by the imputation phase. At the filled-in phase, the missing values for all variables are filled in sequentially over the variables taken one at a time. These filled-in values provide starting values for these missing values at the imputation phase. At the imputation phase, the missing values for each variable are imputed sequentially for a number of burn-in iterations before the imputation.

For continuous variables, a regression method; and for dichotomous variables, a discriminant function method is used to impute missing values.

MI model for Clinical success

Since the clinical success is a composite variable, based on individual signs of severe malaria renal impairment, acidosis, prostration, these individual variables collected before Hours 24 will also be included in the multiple imputation model along with the parasite data while imputing clinical success at Hours 24, which is the first timepoint at which the clinical success is being evaluated.

For the imputation of clinical success at 48 hour, individual signs of severe malaria before 24 hours and at 36 hours will be included but not at the 24 hours in the imputation model. Instead, clinical success at 24 hours will be included in the model. Individual signs at Hours 24 is not included because clinical success at 24 hours is based on the individual signs at Hours 24. The individual signs at 48 hours will also not be included because if the clinical success at Hour 48 is missing then the individual signs will also be missing and hence there is no gain in including these variables in the model, which will unnecessarily increase the burden on the imputation model. Similar approach will be used for the imputation of clinical success at other visits.

Among the following variables, different sets of variables will be used for multiple imputation model depending on which timepoint is concerned- Baseline weight, age, country, log-parasites counts for initial infection (from baseline upto Hours 18 for KAE609 and from baseline upto Hours 48 for artesunate), presence of parasitaemia (yes/no) (post 18 hours for KAE609, post 48 hours for artesunate), altered consciousness (yes/no) at each visit, renal impairment (yes/no) at each visit, acidosis (yes/no) at each visit, respiratory distress (yes/no) at each visit, and clinical success at each visit. Dichotomous variable of existence of parasites after 24 hours for KAE609 and 48 hours for artesunate is considered instead of log-count itself because it is more likely that initial parasites will be disappeared by then resulting into 0 counts for patients treated with KAE609 and artesunate respectively.

Since hospital discharge may be at different timepoint for different patients, it's not possible to provide the fixed number of covariates for imputation of clinical success at discharge. Also, it is expected that most of the patients will be discharged by 72 hours, and this timepoint is considered for the multiple imputation.

The missing values at a particular time point will be imputed using the data prior to that time point only along with other demographic variables. E.g. missing value at 48 hours will be imputed using the data available prior to 48 hours but not using the later timepoint. Clinically, it makes sense to use earlier data to predict the future outcome. This will be controlled by

defining a separate FCS statement for each individual variable, for which the missing values are to be imputed. Data at study discontinuation visit or unscheduled visits will be mapped to the planned visit as per the visit-mapping window.

It has been observed in the Phase II study of oral KAE609 formulation for uncomplicated malaria that the most patients have achieved parasite clearance by 24 hours. This implies that post 24 hours results may be negative for all KAE609 patients, which results into a constant value for all the patients. For artesunate, the median parasite clearance time is expected to be >40 hours. Therefore, post 48 hours, results may be negative for all artesunate patients, which results into a constant value for all the patients. In such cases, variables with constant value will be dropped from imputation model, otherwise, SAS procedure, PROC MI will fail with error.

Clinical success at different visits will be imputed using discriminant function by FCS method. The imputations will be done separately for each treatment group. The number of imputations will be set to 100; the seed for the random function will be set to 60912201 for this study.
To generate the multiple imputed data sets, the SAS procedure MI can be used as follows:

The input data set <CLNSUC_dd> should have one record per patient with country, age, baseline weight, gender, log (parasite counts) from baseline up to 18 hours for KAE (and up to 48 hours for artesunate), parasitaemia status from 24 to 72 hours (presence/absence) for each visit for KAE609 (72 hours for artesunate), altered consciousness (yes/no) at each visit, renal impairment (yes/no) at each visit, acidosis (yes/no) at each visit, and clinical success (yes/no) at each visit.

Two outputs will be prepared displaying i) the missing data patterns and the group means as stored in the data set msgpat and ii) the variance information and estimated means with confidence interval and range based on data sets varinfo and param which need to be merged by <treatment group> and <variable>.

The imputed data are saved in data set <impdata>.

For each of the completed dataset, clinical success will be derived according the definition in Section 2.1.1. The percentage of patients with clinical success ($\hat{P}$) at different timepoints such as 24 hours, 48 hours, 72 hours, hospital discharge, Day 6, Day 8, Day 15, Day 22, and Day 29 will be calculated as n/N x100% for each treatment group and for each of the complete datasets; where n=no. of patients with clinical success and N=no. of patients in FAS.

Combining clinical success rates and 90%CI for each treatment group

Results from the imputation model will be used to estimate the combined clinical success rates and associated 90%CIs.

Rubin (1987) presented rules how to combine the multiple sets of estimates to produce overall estimates and confidence intervals that adequately incorporate missing data uncertainty.

Following this, average estimate of clinical success rate (P) from m completed datasets will be calculated as $\overline{P_m} = \frac{\sum \hat{P}_i}{m}$, where $\hat{P}_i$, is the estimate of P from ith completed dataset.

Estimate of the pooled variance of the m completed dataset is $T_m = \left(1 + \frac{1}{m}\right) B_m + \overline{U_m}$, where $B_m = \frac{\Sigma(\hat{P}_i - \overline{P_m})^2}{m-1}$ (between variance) and $\overline{U_m} = \frac{\Sigma \hat{U}_i}{m}$ (within imputation variance), $\hat{U}_1 = \frac{\hat{P}_i(1-\hat{P}_i)}{n}$.
90% CI for the P will be derived based on Wilson uncorrected method using multiple imputed datasets (MI-Wilson) described by Anne, as follows.

Let t be the quantile of the t_v distribution with the confidence level 1-α such that
$P\left(-t \le \frac{P-\overline{P_m}}{\sqrt{T_m}} \le t\right)$ =1-α .....Eq (1),
where $v = (m-1)(1 + \frac{1}{r_m})^2$, and $r_m = \left(1 + \frac{1}{m}\right) B_m / \overline{U_m}$

Substituting the expression for T_m and squaring the terms inside the above probability expression of equation (1) will lead the quadratic equation for P. Solving the quadratic equation for P, two limits (MI-Wilson) will be obtained as follows.

$$\frac{2\overline{P_m} + \frac{t^2}{n} + \frac{t^2 r_m}{n}}{2\left(1 + \frac{t^2}{n} + \frac{t^2 r_m}{n}\right)} \pm sqrt\left[\frac{\left(2\overline{P_m} + \frac{t^2}{n} + \frac{t^2 r_m}{n}\right)^2}{4\left(1 + \frac{t^2}{n} + \frac{t^2 r_m}{n}\right)^2} - \frac{\overline{P_m}^2}{1 + \frac{t^2}{n} + \frac{t^2 r_m}{n}}\right] \quad \text{----------Eq (2)}$$

It may be possible that $B_m = 0$ *or* $B_m = \overline{U_m} = 0$ leading r_m to be undefined. In this case set $r_m = 0$ with ν=∞ (Anne, Jerome 2018). Note: a t-distribution with ∞ degree of freedom is the standard normal distribution.

As the MIANALYZE procedure expects the parameter estimate in the variables ESTIMATE, and the corresponding standard error in the variable STDERR whereas the MI-Wilson procedure works on solving quadratic equation, MIANALYZE procedure can not be used to combine the 90% CIs for multiple imputations, however, it can still be used to obtain the combined point estimate, $\overline{P_m}$. For 90% CIs, equation (2) will be used.

Between treatment difference and 90% CI in combined clinical success rates based on MI

After obtaining the individual estimates of clinical success rates and the 90% CIs for each treatment group, the between treatment difference and the corresponding 90% CI for the difference will be calculated as described in Newcombe 1998.

Let $\overline{P_{m1}}$ and $\overline{P_{m2}}$ be the average estimate of clinical success rates, L_{m1} and L_{m2} be the lower confidence limits, and U_{m1} and U_{m2} be the upper confidence limits based on the multiple imputed datasets for the test group and control group, respectively,

Then the Newcombe confidence limits for the difference in clinical success rates, $D_m = \overline{P_{m1}} - \overline{P_{m2}}$ is

$D_{mL} = D_m - \sqrt{(\overline{P_{m1}} - L_{m1})^2 + (U_{m2} - \overline{P_{m2}})^2}$

$D_{mU} = D_m + \sqrt{(U_{m1} - \overline{P_{m1}})^2 + (\overline{P_{m2}} - L_{m2})^2}$

Example SAS code

```
#Note: The following codes is written to impute clinical
success at hours 24, 48, 72, Days 6, 8, 15, 22 and Day 29 for
KAE609.
#Since hospital discharge is at different timepoint for
different patients, its not considered for the imputation
since its not feasible  to provide the fixed number of
covariates  for imputation of clinical success at discharge.
# In general, these items are missing altogether if a patient
misses a visit. If a patient does not miss a visit, missing
individual item should be minimal.
The following are the convention for variable naming:
```

- Suffix after '_' indicates timing, ‖B= means baseline, H36 means 36 hours posting dosing, D15 means at Day 15, etc.
- Prefix 'par': means dichotomous parasite count (absent or presence)
- Prefix ‖ln=: means log parasite count (continuous)
- Prefix ‖CLSUC=: means clinical success (yes or no)
- Prefix ‖REN=: means renal impairment (yes/no)
- Prefix ‖ACID=: means acidosis (yes/no)
- Prefix ‖ALTCON=: means altered consciousness (yes/no)

```
ODS LISTING CLOSE;
ODS OUTPUT MissPattern=msgpat VarianceInfo=varinfo
ParameterEstimates=param;
/*FOR KAE609*/
proc mi data= <CLINSUC_dd> seed=2201 nimpute=&nimp out=
clinsuc_mi;
   where trt01p=KAE609;  *Imputation within treatment;
class  COUNTRY gender par_h24 par_h36 par_h48 par_h72
CLSUC_h24 CLSUC_h48 CLSUC_h72 CLSUC_D6 CLSUC_D8 CLSUC_D15
CLSUC_D22 CLSUC_D29 REN_B REN_H6 REN_H12 REN_H18 ACID_B
ACID_H6 ACID_H12 ACID_H18
```

```
ALTCON_B ALTCON_H6 ALTCON_H12 ALTCON_H18;*put all categorical
variables upto 24 hours since they are needed to impute
clinical response at 24 hours;
fcs discrim (CLSUC_h24  = COUNTRY age gender ln_s ln_b ln_h1
ln_h2 ln_h4 ln_h6 ln_h12 ln_h18 REN_B REN_H6 REN_H12 REN_H18
ACID_B ACID_H6 ACID_H12 ACID_H18 ALTCON_B ALTCON_H6 ALTCON_H12
ALTCON_H18 )
fcs discrim (CLSUC_48 = COUNTRY age gender ln_s ln_b ln_h1
ln_h2 ln_h4 ln_h6 ln_h12 ln_h18 par_h24 par_h36  REN_B REN_H6
REN_H12 REN_H18 REN_H36 ACID_B ACID_H6 ACID_H12 ACID_H18
ACID_H36 ALTCON_B ALTCON_H6 ALTCON_H12 ALTCON_H18 ALTCON_H36
CLSUC_h24)
fcs discrim ( CLSUC_h72 = COUNTRY age gender ln_s ln_b ln_h1
ln_h2 ln_h4 ln_h6 ln_h12 ln_h18 par_h24  par_h36 par_h48
REN_B REN_H6 REN_H12 REN_H18 REN_H36 ACID_B ACID_H6 ACID_H12
ACID_H18 ACID_H36 ALTCON_B ALTCON_H6 ALTCON_H12 ALTCON_H18
ALTCON_H36 CLSUC_h24 CLSUC_h48)
fcs discrim ( CLSUC_D6 = COUNTRY age gender ln_s ln_b ln_h1
ln_h2 ln_h4 ln_h6 ln_h12 ln_h18 par_h24  par_h36 par_h48
par_h72 REN_B REN_H6 REN_H12 REN_H18 REN_H36 ACID_B ACID_H6
ACID_H12 ACID_H18 ACID_H36 ALTCON_B ALTCON_H6 ALTCON_H12
ALTCON_H18 ALTCON_H36 CLSUC_h24 CLSUC_h48 CLSUC_h72
)
fcs discrim ( CLSUC_D8 = COUNTRY age gender ln_s ln_b ln_h1
ln_h2 ln_h4 ln_h6 ln_h12 ln_h18 par_h24  par_h36 par_h48
par_h72 REN_B REN_H6 REN_H12 REN_H18 REN_H36 ACID_B ACID_H6
ACID_H12 ACID_H18 ACID_H36 ALTCON_B ALTCON_H6 ALTCON_H12
ALTCON_H18 ALTCON_H36 CLSUC_h24 CLSUC_h48 CLSUC_h72 CLSUC_D6
)

fcs discrim ( CLSUC_D15 = COUNTRY age gender ln_s ln_b ln_h1
ln_h2 ln_h4 ln_h6 ln_h12 ln_h18 par_h24  par_h36 par_h48
par_h72 REN_B REN_H6 REN_H12 REN_H18 REN_H36 ACID_B ACID_H6
ACID_H12 ACID_H18 ACID_H36 ALTCON_B ALTCON_H6 ALTCON_H12
```

```
ALTCON_H18 ALTCON_H36 CLSUC_h24 CLSUC_h48 CLSUC_h72 CLSUC_D6
CLSUC_D8
)

fcs discrim ( CLSUC_D22 = COUNTRY age gender ln_s ln_b ln_h1
ln_h2 ln_h4 ln_h6 ln_h12 ln_h18 par_h24  par_h36 par_h48
par_h72 REN_B REN_H6 REN_H12 REN_H18 REN_H36 ACID_B ACID_H6
ACID_H12 ACID_H18 ACID_H36 ALTCON_B ALTCON_H6 ALTCON_H12
ALTCON_H18 ALTCON_H36 CLSUC_h24 CLSUC_h48 CLSUC_h72 CLSUC_D6
CLSUC_D8 CLSUC_D15
)
fcs discrim ( CLSUC_D29 = COUNTRY age gender ln_s ln_b ln_h1
ln_h2 ln_h4 ln_h6 ln_h12 ln_h18 par_h24  par_h36 par_h48
par_h72 REN_B REN_H6 REN_H12 REN_H18 REN_H36 ACID_B ACID_H6
ACID_H12 ACID_H18 ACID_H36 ALTCON_B ALTCON_H6 ALTCON_H12
ALTCON_H18 ALTCON_H36 CLSUC_h24 CLSUC_h48 CLSUC_h72 CLSUC_D6
CLSUC_D8 CLSUC_D15 CLSUC_D22
)
var COUNTRY age weight gender
ln_s ln_b ln_h1 ln_h2 ln_h4 ln_h6 ln_h12 ln_h18 par_h24
par_h36 par_h48 par_h72
CLSUC_h24 CLSUC_h48 CLSUC_h72 CLSUC_D6 CLSUC_D8 CLSUC_D15
CLSUC_D22 CLSUC_D29 REN_B REN_H6 REN_H12 REN_H18 REN_H24
ACID_B ACID_H6 ACID_H12 ACID_H18 ACID_H24 ALTCON_B ALTCON_H6
ALTCON_H12 ALTCON_H18 ALTCON_H24
run;

ODS LISTING;

/*Artusunate*/

/*Similar code will be used for the imputation of clinical
success for artesunate with the change of variables related to
parasite count data such as- a). Delete par_h24, par_h36, and
par_h48 from CLASS and VAR statement. b) Include ln_h24,
ln_h36 and ln_h48 in VAR statement, and c) Replace par_h24,
```

```
par_h36, and par_h48 with ln_h24, ln_h36 and ln_h48 in FCS
statements respectively */
```

5.9 Rule of exclusion criteria of analysis sets

Table 5-15 Protocol deviations and non-PD criteria leading to exclusion from analysis sets

Analysis Set	PD (Description and ID) that causes Patients to be excluded	Non-PD criteria that cause Patients to be excluded
Randomized	• Informed Consent for study participation or parental consent not obtained and patient entered trial (INCL02)	• Not randomized
FAS	NA	• Not in Randomized set • Misrandomized if identified from IRT • Baseline parasitaemia count is 0 or missing using local lab (central lab in case local lab is not available) • No study drug taken
PPS	• Not severe malaria for Cohort 2-5 (INCL01b) and not moderately severe malaria for Cohort 1 (INCL01a_1) • Concomitant antimalarial medications (COMD01) if received before assessing the primary endpoint at 12 hours • Unacceptable prior antimalarial drugs within 12 hours of screening (EXCL01.02)	• Not in FAS; • Baseline Parasite species is other than *Plasmodium* falciparum OR mixed infection using local lab (central lab in case local lab is not available) • Baseline Plasmodium falciparum parasite count <60000/uL or >250000/uL for cohorts 1-2 or <5000/ uL for cohorts 3-5 at using local lab (central lab in case local lab is not available)
SAF	NA	• Not in Randomized set; • No study drug taken
PK	• Patients who received concomitant prohibited medication which may have an impact on PK exposure (COMD04)	• Not in SAF • No evaluable pharmacokinetic parameter data • Did not take at least on dose of IV study drug

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