PROTOCOL

PROTOCOL TITLE:	Single dose oral bioequivalence study of Primaquine phosphate tablets USP 15 mg (Test) and Primaquine 15 mg Tablet (Reference) in healthy adult human subjects under fasting conditions.
STUDY DESIGN:	An open label, randomized, two-period, two-treatment, two-sequence, crossover, balanced, single dose oral bioequivalence study.
INVESTIGATIONAL PRODUCTS:	• *Test Product (T):* Primaquine phosphate tablets USP 15 mg Manufactured By: IPCA, 125, Kandivli Industrial Estate, Kandivli (West), Mumbai – 400 067, Maharashtra, India. • *Reference Product (R):* Primaquine 15 mg Tablet – Manufactured by: Sanofi, Bridgewater, NJ, USA.

PROTOCOL VERSION NO.: 03

PROTOCOL DATE: October 04, 2022

SPONSOR NAME AND ADDRESS

The Chancellor, Masters & Scholars of the University of Oxford

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Study Funder

UK MRC

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Reference numbers

Trial registration number: ISRCTN54640699

MORU Ref: MAL21010

OxTREC Ref: 40-21

Roles of the sponsor and funder

The study sponsor designed the study and will oversee collection, analysis and interpretation of the data, writing of the report; and the decision to submit the report for publication.

The funder, the UK MRC, had no role in the design of the study and will not have any role in its execution, analyses, and interpretation of data or decision to submit results.

PROTOCOL VERSION CONTROL

Version number	Date
03	October 04, 2022
02	March 15, 2022
01	October 12, 2021

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LIST OF ABBREVIATIONS

AE	**:**	Adverse Event
ALCOA+	**:**	A commonly used acronym for "attributable, legible, contemporaneous, original, accurate, available, enduring, consistent and complete"
ANOVA	**:**	Analysis of Variance
AUC	**:**	Area Under Curve
BMI	**:**	Body Mass Index calculated as weight in kg / (height in meter)^2
BUN	**:**	Blood Urea Nitrogen
CBC		Complete blood count
cGMP	**:**	Current Good Manufacturing Practice
C-PQ	**:**	Carboxyprimaquine
CRF	**:**	Case Report Form
Ct	**:**	Last quantifiable concentration
CYP	**:**	Cytochrome P450
DMP	**:**	Data Management Plan
ECG	**:**	Electrocardiogram
EDC		Electronic Data Capture
GCP	**:**	Good Clinical Practice
GLM	**:**	General Linear Model
HBsAg	**:**	Hepatitis B Surface Antigen
HCT	**:**	Haematocrit
HCV	**:**	Hepatitis C virus
HIV	**:**	Human Immunodeficiency Virus
ICD	**:**	Informed Consent Document
ICH	**:**	The International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human use
ICMR	**:**	Indian Council of Medical Research
IEC	**:**	Independent Ethics Committee
IP	**:**	Investigational Product
K_2EDTA	**:**	Dipotasium Ethylene Diamine Tetra Acetic Acid
Kg	**:**	Kilogram
Ltd	**:**	Limited

LCMS/MS	**:**	Liquid Chromatography coupled to tandem Mass Spectrometry
LLOQ	**:**	Lower Limit of Quantitation
m	**:**	Meter
mL	**:**	millilitre
mg	**:**	milligram
mmHg	**:**	Millimetre of mercury
MSR	**:**	Medical Screening Record
NUMPT	**:**	The number of points of the terminal log-linear phase used to estimate the terminal rate constant
OH-PQm	**:**	Hydroxylated-PQ metabolites
OTC	**:**	Over The Counter
OxTREC	**:**	Oxford Tropical Research Ethics Committee
PI	**:**	Principal Investigator
PK	**:**	Pharmacokinetics
PQ	**:**	Primaquine
R	**:**	Reference product
RBC	**:**	Red Blood Cells
REGEND	**:**	Regression end time point for calculation of Kel
REGSTART	**:**	Regression start time point for calculation of Kel
rpm	**:**	Revolutions per minute
RPR	**:**	Rapid Plasma Reagin
SAE	**:**	Serious Adverse Event
SAS	**:**	Statistical Analysis Software
SGOT	**:**	Serum Glutamic Oxaloacetic Transaminase
SGPT	**:**	Serum Glutamic Pyruvic Transaminase
SOP	**:**	Standard Operating Procedure
T	**:**	Test product
THC	**:**	Tetrahydrocannabinol
USA	**:**	United States of America
WBC	**:**	White Blood Cells
WHO	**:**	World Health Organization
β-HCG	**:**	Beta Human Chorionic Gonadotropin

ºC	**:**	Degree Celsius
AUC_% Extrap _obs	**:**	Percentage of extrapolated AUC

DECLARATION OF INVESTIGATORS

We, the undersigned, have read and understood this protocol and hereby agree to conduct the study in accordance with this protocol and to comply with all requirements regarding the obligations of investigators and all other pertinent requirements of 'WHO', ICH 'Guidance on Good Clinical Practice', the ICMR Ethical guidelines, 'New Drugs and Clinical Trials Rules, 2019', Declaration of Helsinki, Principles of Good Laboratory Practice and all applicable guidelines and regulations.

We agree to comply with all relevant SOPs required for the conduct of this study. We further agree to ensure that all associates assisting in the conduct of study are informed regarding their obligations. We have no competing interests.

Bob Taylor 4-10-22

Signature & date

Dr. Bob Taylor

Principal Investigator

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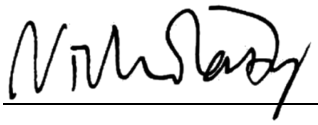
DECLARATION OF SPONSOR

I, on behalf of ***The Masters & Scholars of the University of Oxford*** have read, understood and approve this protocol. I agree to comply with all the obligations of sponsor, all other pertinent requirements of study and all applicable guidelines and regulations.

I also agree to comply with cGMP practices for providing the study drug for human consumption. I declare no competing interests.



7 October 2022

Signature Date

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1.0 FACILITIES

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1.6 INDEPENDENT ETHICS COMMITTEE

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2.0 PROTOCOL SUMMARY

Protocol Title:	Single dose oral bioequivalence study of Primaquine phosphate tablets USP 15 mg (Test) and Primaquine 15 mg Tablet (Reference) in healthy adult human subjects under fasting conditions.
Short Title:	Primaquine 15 mg bioequivalence study
Regulatory Submission:	WHO
Objectives:	• To compare and evaluate the oral bioavailability of Primaquine phosphate tablets USP 15 mg of IPCA with that of Primaquine 15 mg Tablet of Sanofi in healthy, adult, human subjects under fasting conditions. • To monitor the safety and tolerability of the subjects.
Study Design:	An open label, randomized, two-period, two-treatment, two-sequence, crossover, balanced, single dose oral bioequivalence study.
No. of subjects:	50
Housing:	At least 11 hours prior to dosing until at least 24 hours post dose in each period.
Washout period:	At least 10 days.
Study duration:	Considering the minimum washout period, expected study duration of clinical part is 15 days from the day of check-in of first period.
Administration of investigational products:	One tablet of either test product Primaquine phosphate tablets USP 15 mg of IPCA or reference product Primaquine 15 mg Tablet of Sanofi will be administered orally to the subjects as per the randomization schedule in a sitting posture with about 240 mL of water at ambient temperature in each period under the supervision of trained study personnel. This activity will be followed by mouth and hand check of the subjects to assess compliance to dosing. Investigational products (test or reference) must be swallowed whole and must not be chewed, crushed or divided.
Food and fluid restrictions:	Fasting for at least 10 hours prior to dosing until at least 04 hours post-dose in each period. Water will be restricted from at least 01 hour prior to dosing until at least 02 hour post-dose in each period (no fluid, except water given with dosing).
Posture restrictions:	Subject will remain seated upright for initial 04 hours post-dose and only necessary movement will be allowed during this period. They will not be allowed to lie down except as directed by the physician and secondarily due

	to adverse events during this restriction period. Thereafter subjects will be allowed to ambulate freely during the remainder of the study.
Collection of blood samples:	In each period, total 26 venous blood samples (06 mL each) will be collected at pre-dose (0.0 hour) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0, 48.0** and **72.0** hours post dose in labelled K_2EDTA vacutainers through an indwelling cannula placed in the forearm vein/dorsal aspect of hand of the subjects. If required, a sample may alternatively be collected through a fresh venepuncture. The pre-dose (0.0 hour) blood sample will be collected within 120 minutes prior to dosing in each period. All post-dose samples in each period will be collected within +2 minutes of the scheduled time; however, ambulatory samples will be collected within +60 minutes of the scheduled time. The following blood sampling times will be considered for analysis: **For Primaquine:** 0.0 (Pre-dose) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0** and **48.0** hours post dose. **For Carboxyprimaquine:** 0.0 (Pre-dose) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0, 48.0** and **72.0** hours post dose.
Ambulatory visits:	Subjects will report to the clinical facility at **36.0, 48.0** and **72.0** hours post-dose for compliance assessment, well-being assessment and collection of ambulatory blood samples in each period.
Handling of blood samples:	After collection, the blood samples will be placed in an ice bath or other chilling device until centrifugation. Blood samples will be placed in a refrigerated centrifuge within 45 minutes of blood sample collection, and then will be spun at 4000 rpm at 4°C ± 2ºC for 10 minutes. The plasma will then be separated, transferred to labelled polypropylene tubes in duplicates (primary and secondary aliquots with equal volume) and stored in freezer at –20ºC ± 7ºC at the clinical facility until shipment to the analytical facility. The samples will be stored in a freezer at –20ºC ± 7ºC at the analytical facility until analysed. All the temperature excursions will be handled as per in-house SOP. In case the processing error occurred due to any reason (e.g. mixing of sample while segregation, mechanical failure in centrifuge, etc.) during the sample processing, re-spin the sample (s) under the same conditions, if required.

	Note: Transfer of plasma samples into the freezer shall take place as soon as possible so the total elapsed time from blood collection to placement of plasma samples in the freezer does not exceed 90 minutes.
Total blood loss:	The volume of blood required from each subject for the study will be 348 mL.
Safety assessment:	*Vital signs:* • Sitting blood pressure, pulse rate and body temperature will be measured at the time of check-in and prior to check-out in each period. Subjects must have clinically acceptable vital signs prior to check-in of each period. • Sitting blood pressure, pulse rate and body temperature will be measured prior to dosing in each period and during the visit for the last study sample. • Sitting blood pressure and pulse rate will be measured at 2.0 hours (± 40 minutes) post dose in each period. • Sitting blood pressure, pulse rate and body temperature will be measured at 6.0 and 10.0 hours (± 40 minutes) post dose and as needed in each period. *Physical examination (clinical examination)* of the subject will be conducted by a qualified medical designate at the time of check-in and prior to check-out in each period and during the visit for the last study sample. *Well-being assessment:* Subjects will be advised to report any AE that may occur during the study and will be specifically asked for these by trained study personnel in a non-leading manner at the time of physical examination (clinical examinations), during vital signs recording, at about 16.0 and 24.0 hours post dose, during ambulatory visits and as needed in each period. Adverse events will be managed and recorded as appropriate by the investigator or available physician. A complete blood count (CBC) will be measured prior to check in of period II. Only subjects within normal limits or clinically insignificant falls will be dosed in period II. A urine pregnancy test will be performed prior to check-in of each period and serum (β-HCG) pregnancy test will be performed at the time of the end of study laboratory assessment for female volunteers of child bearing potential. Volunteers with a negative test result for pregnancy (prior to check-in of each period) will only be dosed. All study subjects who are dosed, will be assessed for their well-being through a physical examination (clinical examination) which will include vital sign measurement (sitting blood pressure, pulse rate and body

	temperature) and laboratory tests at the end of the study or as applicable (for details refer to section# 13.2; safety assessment during study).
Analytical methodology:	Plasma samples will be assayed by a validated LCMS/MS method developed at Cliantha Research, Ahmedabad, which is specific for the determination of Primaquine and metabolite Carboxyprimaquine. The following blood sampling times will be considered for analysis: **For Primaquine:** 0.0 (Pre-dose) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0** and **48.0** hours post dose. **For Carboxyprimaquine:** 0.0 (Pre-dose) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0, 48.0** and **72.0** hours post dose.
Pharmacokinetic parameters:	Pharmacokinetic parameters Cmax, AUCt, AUC72, AUCi, Tmax, Kel, AUC_%Extrap_obs and tHalf will be calculated using Phoenix® WinNonlin® professional software (Version 8.1.1 or higher; Pharsight Corporation, USA) or SAS® statistical software (Version: 9.4 or higher; SAS Institute Inc, USA).
Statistical analysis:	Statistical analysis will be performed on the pharmacokinetic parameters using SAS® statistical software (Version: 9.4 or higher; SAS Institute Inc., USA).
Assessment of bioequivalence:	The 90% confidence interval of the relative mean (geometric least square mean) of the test to reference product for Ln-transformed Pharmacokinetic parameters C_{max} and AUCt should be within 80.00% to 125.00% to establish bioequivalence. Data of inactive metabolite of Carboxyprimaquine will be provided as supportive data.

SEQUENCE OF EVENTS

Day	Proposed time relative to dosing	Details of events	Applicable to
-28	Within 28 days prior to dosing	Informed consent document presentation for screening & Screening of volunteers	P- I
-1	When volunteers report to facility	Attendance	Each period
	Prior to Check-in	Study specific Informed consent document presentation	P- I
		Urine scans for drugs of abuse and alcohol test	Each period
		Measurement of CBC (Complete blood count)	P-II
		Urine pregnancy test for female volunteers of child bearing potential only	Each period
		Physical examination (Clinical examination), vital signs recording & well-being assessments	
		Compliance check	
		Inclusion Criteria and Exclusion Criteria assessment	P- I
	At least 11 hours prior to dosing	Subjects check-in	Each period
-1, 0 & 1	Check-in to 24 hours post dose	Housing duration	
-1	At least 10 hours prior to dosing	Dinner	
0	Prior to dosing	Pre-dose vital signs recording & well-being assessments	
		Cannulation	
	01 hour pre-dose to 02 hour post-dose	Water restriction	
	0 hour	Dosing	
	At least 04 hours post-dose	Posture Restriction - Seated upright position	
0 to 3	At pre-dose (0.0 hour) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0, 48.0** and **72.0** hours post dose	Collection of 06 mL each venous blood samples in labelled K_2EDTA-vacutainers Compliance assessment and well-being assessments will be performed during ambulatory samples	
0	At 2.0, 6.0 & 10.0 hours (± 40 minutes) post dose	Post-dose vital signs recording & well-being assessments	
	1) After 04 hours post dose 2) Further meals will be provided at appropriate times	1) Lunch 2) Further meals like snacks & dinner	
1	At about 16.0 and 24.0 hours post dose	Well-being assessments	
	Prior to check-out	Physical examination (Clinical examination), vital signs recording & well-being assessments	
	After 24.0 hours post dose	Check-out	
3	At about 72.0 hours post dose	1) Physical examination (Clinical examination), vital signs recording & well-being assessments 2) Collection of blood for end study laboratory assessment (including Serum (β-HCG) pregnancy test only for female subjects having child bearing potential)	P-II (End study)

3.0 BACKGROUND INFORMATION

3.1 DETAILS OF INVESTIGATIONAL PRODUCTS

- *Test Product (T):* Primaquine phosphate tablets USP 15 mg – Manufactured By: IPCA, 125, Kandivli Industrial Estate, Kandivli (West), Mumbai – 400 067, Maharashtra, India.
- *Reference Product (R):* Primaquine 15 mg Tablet – Manufactured by: Sanofi, Bridgewater, NJ, USA. The Sanofi produced primaquine is the WHO-mandated reference product.

3.2 REFERENCE PRODUCT SUMMARY

Indication:

Primaquine phosphate is indicated for the radical cure (prevention of relapse) of vivax Malaria.

Pharmacokinetics:

Peak plasma concentrations occur about 1 to 3 hours after a dose is taken and then rapidly diminish with a reported elimination half-life of 7 hours (3.7 to 9.6 hours).

Taking primaquine after a meal may reduce abdominal pain or cramps associated with ingestion of the drug.

Recommended Dose:

Primaquine phosphate should be administered concurrently in order to eradicate the exoerythrocytic parasites in a dosage of 1 tablet (equivalent to 15 mg base) daily for 14 days.

3.3 RISKS AND BENEFITS

Participation in this study will yield no direct benefits to the subjects.

The risks as mentioned below are minimized considering the fact that only a single dose in each period is to be administered to the subjects with an appropriate washout interval between treatment periods.

The adverse events are nausea, vomiting, epigastric distress, and abdominal cramps, leukopenia, hemolytic anemia in G6PD deficient individuals, and methemoglobinemia in nicotinamide adenine dinucleotide (NADH) methaemoglobin reductase deficient individuals, cardiac arrhythmia and QT interval prolongation, dizziness, rash and pruritus.

Blood taking causes discomfort but this tends to be transient. Rarely, a veinpuncture or finger stick site may become infected. Full aseptic techniques will be used when taking blood to minimise the infection risk. The volume of blood taken is very small and not a health threat.

There is no direct short term benefits to the community. If we are able to eventually make paediatric PQ available, people in malaria endemic countries will benefit.

3.4 SUBJECT SAFETY AND DATA QUALITY ASSURANCE

Risks in this study to subject safety and data quality are assessed as follows:

a) Critical Parameters and procedures:

- Subject safety
  - PI oversight
  - Training and delegation of the staff
  - Inclusion of correct subject population
  - Compliance of Equipment's' used during the study
  - Management of deviations with corrective and preventive actions, as applicable
  - Ensuring adequate safety assessment throughout the study
  - Handling, use and storage/retention of IPs as per protocol
  - Compliance to protocol
  - Subjects are adequately informed about the risk/adverse events and study requirements
  - IEC approval
  - Regulatory approval
  - Vendor management
- Data quality
  - Tools for data collection and analysis designed as per study endpoints and objectives
  - Data collected as per ALCOA+
  - Adequate audit trails irrespective of type of media used
  - Data and record handling to ensure traceability and reconstruction of the study
  - Data access and storage, as per applicable regulatory requirements
  - Use of validated systems to generate critical study data
  - Adequate data back-up system
  - Secured agreement between sponsor and all involved parties

b) Risk identification and assessment:

COVID-19 pandemic has created lot of uncertainty in the current situation and has put subject's safety, protocol compliance and data validity at high risk.

Risk associated with possible COVID 19 Exposure:

Risk Mitigation plan/Risk Evaluation & Mitigation strategy will be made to minimize the risk for COVID 19 exposure and to handle possible situations in COVID 19 outbreak

scenario.

Other identified critical parameters for this study are assessed as low risk with minimal impact on human subject safety, and/or reliability of study results, as this study is governed by the well-defined quality management system operative at Cliantha Research. This includes well trained and delegated staff, SOPs and plans including document control procedures, monitoring, quality control and audits, calibrated instruments, validated systems as well as appropriately designed data collection tools such as CRFs and other forms.

Risks will be continuously tracked and dynamically assessed through principal investigator/responsible stakeholder oversight, monitoring, quality control, auditing and deviation management procedure.

This will be communicated to the affected parties and reported in the clinical study report, as needed.

3.5 PANDEMIC COVID-19 RESPONSE PLAN

Regulatory authorities have recognized that the Coronavirus Disease 2019 (COVID-19) pandemic may impact the conduct of clinical study.

Due to COVID-19 pandemic, challenges may arise for clinical study conduct, for example, quarantines of site personnel/study participants, travel limitations, interruptions to the supply chain for the IP(s), or other considerations if site personnel or study participants become infected with COVID-19. These challenges may lead to difficulties in meeting protocol specified procedures, including administration or use of investigational product, housing duration or adhering to protocol specified visits and laboratory/diagnostic testing.

To accommodate these challenges and mitigate safety risks associated with COVID-19, changes may be required from approved protocol which include (but not limited to) change in study procedures timing, change in subject's housing duration; additional test or parameter may be performed to standard inclusion or exclusion criteria, etc. The changes made to the procedure will prioritize subject's safety and data validity and integrity. For any significant change, a planned protocol deviation will be filled and notified to ethics committee and/or local regulatory (as applicable).

Additional health checks including body temperature or other vital sign monitoring, etc. may be performed during the study, even if not specified in the protocol. Subject who is tested positive to COVID 19 during the study will be withdrawn from the study. This subject and other subjects in close contact will be handled as per government directive effective during study execution period.

All participants will be pre-screened prior to enrolment into the study and evaluated for symptoms of COVID-19 according to in-house SOP and the most recent Ministry of Health, India guidelines available at the time of pre-screening. They will be instructed to adhere to social distancing recommendations according to these guidelines until the time of their scheduled visit and report any change in their status.

As the science and regulations are continuously being adapted to the evolving information around the pandemic, additional measures apart from the ones mentioned here may be

undertaken to ensure subject safety and appropriate study conduct. The Ethics committees, sponsors would be informed for their review and approval as applicable. The updated information would be presented to the subjects where applicable and re-consent for the study sought.

All specific measures taken during the study to manage the COVID-19 related risk will be documented as part of study documentation and reported in the clinical study report.

4.0 STUDY BACKGROUND & RATIONALE

Primaquine (PQ) or PQ phosphate is an 8-aminoquinoline that was developed in the 1950s by the US army. It is used for as antirelapse in *Plasmodium vivax* and *P. ovale* and has been recommended for many years by the WHO for transmission blocking of *P. falciparum* malaria.

Primaquine was originally developed as 0.25–0.5 mg base/kg body weight for 14-day course of treatment for the radical cure of *P. vivax* and *P. ovale* through clearance of hypnozoites following administration of blood-stage schizonticidal drugs such as chloroquine or artemisinin-based combination therapies (ACTs) (Vale, Moreira et al. 2009). Even though a 14-days course is a standard treatment for radical cure of *P. vivax*, a 7-day shorter course has also been evaluated in large RCT studies. In patients with normal G6PD, a 7-day PQ treatment was non-inferior to 14-day primaquine that might improve adherence for radical cure of *P. vivax* malaria (Taylor, Thriemer et al. 2019).

There is now a growing interest in single low dose PQ for blocking the transmission of *Plasmodium falciparum*, following WHO's recommendation in 2012 (World Health Organization 2016). WHO recommends the use of a single 0.25-mg/kg dose of primaquine (PQ) in combination with standard ACT for the treatment of *P. falciparum* malaria in elimination and resistance containment settings. Several trials have shown that single low dose primaquine reduced mosquito infectivity (Goncalves, Pett et al. 2017) (Dicko, Brown et al. 2016).

Despite its long use, the mechanism of action for PQ is largely unknown. The general understanding for the link between PQ efficacy and metabolism through CYP2D6 is supported by both animal and clinical studies. This includes the association of CYP2D6 poor metaboliser phenotype status with poor antirelapse efficacy in *P. vivax* (Bennett, Pybus et al. 2013).

Camarda et al have recently demonstrated that the antimalarial activity of PQ was via a two-step biochemical relay. The activity of hydroxylated-PQ metabolites (OH-PQm), produced by CYP 2D6, are enhanced ca.1000 fold in the presence of cytochrome P450 NADPH:oxidoreductase (CPR) from the liver and bone marrow. Enhancement of OH-PQm efficacy is due to the direct reduction of quinoneimine metabolites by CPR with the concomitant and excessive generation of H_2O_2, leading to parasite killing. (Camarda, Jirawatcharadech et al. 2019).

Primaquine is metabolised by monoamine oxidases (MAOs) to its main non active metabolite, carboxyprimaquine (C-PQ) (Marcsisin, Reichard et al. 2016), which is easily measurable unlike the oxidative metabolites.

Non-compartmental pharmacokinetic analyses assessing interactions between PQ and chloroquine, dihydroartemisinin/piperaquine, and pyronaridine/artesunate demonstrated that higher PQ exposures when administered in combination than when it is administered alone (Pukrittayakamee, Tarning et al. 2014) (Hanboonkunupakarn, Ashley et al. 2014) (Jittamala, Pukrittayakamee et al. 2015).

In a population pharmacokinetic modelling studies characterizing the enantiospecific properties of PQ, exposure to PQ, particularly to the (-)-S-primaquine but not the carboxy metabolites, increased by up to 30% when co-administered with commonly used antimalarial drugs. Such interactions were enantiospecific with a relatively higher effect on (-)-S-primaquine than on (+)-R-primaquine. No drug–drug interaction effects were seen on the pharmacokinetics of either carboxyprimaquine enantiomer (Chairat, Jittamala et al. 2018).

Children are disproportionally affected by malaria of which vivax malaria affects young children less than 15 years of age peaking between 2 and 6 years that carries the highest risk of morbidity and mortality. Children under the age of 5 years accounted 67% of deaths from malaria in African region in 2019 (WHO 2020).

However, there are no paediatric PQ formulations that are friendly to children. The availability of paediatric drugs in the right dosage forms with acceptable taste and odour/flavour is critically important for increasing adherence and allowing PQ regimens that do not need tablet fractions or require crushing tablets.

PQ is now considered essential for eliminating malaria but to ensure child friendly PQ is made available it either has to be WHO prequalified or registered with a stringent drug regulatory authority.

WHO prequalification offers the possibility of a line extension based on demonstrating proportionality with the adult 15 mg tablet. For a generic manufacturer, a bioequivalence study must be performed comparing the generic (test) product with the reference 15 mg of primaquine.

This study is to establish the bioequivalence of a new scored 15 mg generic PQ tablet, produced by IPCA in India. The study will be conducted to international standards, following the WHO guideline /annex 9/ and other applicable requirements for bioequivalence evaluation.

5.0 STUDY OBJECTIVES

- To compare and evaluate the oral bioavailability of Primaquine phosphate tablets USP 15 mg of IPCA with that of Primaquine 15 mg Tablet of Sanofi in healthy, adult, human subjects under fasting conditions.
- To monitor the safety and tolerability of the subjects.

6.0 STUDY DESIGN

An open label, randomized, two-period, two-treatment, two-sequence, crossover, balanced, single dose oral bioequivalence study.

7.0 STUDY POPULATION

A sufficient number of subjects will be enrolled to ensure dosing of **50** healthy adult human subjects.

Based on data provided by sponsor, the sample size estimation for this study is as mentioned below.

Assuming a 5% difference of exposure and maximum concentration between the test and reference primaquine tablets.

Intra-subject C.V (%) ~ 25%

Significance level = 5 %

Power = 95 %

Based on the above estimates, a sample size of 45 subjects should be sufficient to establish bioequivalence with adequate power. Considering dropouts and withdrawals, **50** subjects will be required.

The sample size calculations have been performed in PASS software. The sample size formulae used are from Chow, Shein-Chung, et al. Sample size calculations in clinical research. Chapman and Hall/CRC, 2017 and Julious, Steven A. Sample sizes for clinical trials. Chapman and Hall/CRC, 2009.

8.0 SELECTION OF SUBJECTS

Cliantha has an experienced volunteer recruitment team which has access to a Cliantha database of thousands of volunteers. The recruitment team will ensure sufficient number of screening of volunteers through this database. Subject will go through screening procedures at facility after due consent.

At the facility, on check in day subjects will be given copies of the consent form of the study and study personnel will give a presentation for the same. Subjects who have signed the consent form will be enrolled in the study.

Selection of subjects for the study will be done based on assessment against the inclusion and exclusion criteria below.

Subjects from the pool of healthy volunteers who were screened within 28 days prior to the first dosing day will be considered as potential participants in the study.

Subjects must be enrolled in the study only after providing written informed consent.

8.1 INCLUSION CRITERIA

Volunteers must fulfil all of the following inclusion criteria to be eligible for participation in the study, unless otherwise specified.

1) Age: 18 to 45 years old, both inclusive.
2) Gender: Male and/or non-pregnant, non-lactating female.
   A. Female of childbearing potential must have a negative serum beta human

chorionic gonadotropin (β-HCG) pregnancy test performed within 28 days prior to first dosing day. They must be using an acceptable form of contraception.

B. For female of childbearing potential, acceptable forms of contraception include the following:

i. Non hormonal intrauterine device in place for at least 3 months prior to the start of the study and remaining in place during the study period, **or**

ii. Barrier methods containing or used in conjunction with a spermicidal agent, **or**

iii. Surgical sterilization **or**

iv. Practicing sexual abstinence throughout the course of the study.

C. Female will not be considered of childbearing potential if one of the following is reported and documented on the medical history:

i. Postmenopausal with spontaneous amenorrhea for at least one year, **or**

ii. Bilateral oophorectomy with or without a hysterectomy and an absence of bleeding for at least 6 months, **or**

iii. Total hysterectomy and an absence of bleeding for at least 3 months.

3) BMI: 18.5 to 30.0 kg/m^2, both inclusive; BMI value should be rounded off to one significant digit after decimal point (e.g. 30.04 rounds down to 30.0, while 18.45 rounds up to 18.5).

4) Able to communicate effectively with study personnel.

5) Willing to provide written informed consent to participate in the study.

6) Non-smokers and non-tobacco user (i.e. having no past history of smoking and tobacco consuming for at least one year prior to study).

7) All volunteers must be judged by the principal or sub-investigator or physician as normal and healthy during a pre-study safety assessment performed within 28 days of the first dose of study medication which will include:

a) A physical examination (clinical examination) with no clinically significant finding.

b) Results within normal limits defined site normal range (as mentioned in APPENDIX-II) for the following tests:

Hematology				
Haemoglobin	Total RBC count	Total WBC count	Platelet count	
Differential leukocyte count:				
Neutrophils	Lymphocytes	Eosinophils	Monocytes	Basophils

Blood indices: HCT			
Biochemistry			
BUN	Serum creatinine	Random glucose	SGPT & SGOT
Alkaline phosphatase	Uric acid	Serum bilirubin	
Serum total protein: Total proteins, Albumin			
Serum electrolytes: Serum sodium, serum chloride, serum potassium, serum phosphorous, serum calcium			
Urinalysis			
Colour, quantity, specific gravity, odour, appearance, reaction, albumin, bilirubin, ketone bodies, sugar, urobilinogen and microscopical examination (*performed based on clinical judgment*)			
Immunological Tests			
HIV-I & II	HBsAg	Syphilis (RPR)	Anti HCV
Serum (β-HCG) pregnancy test (for female of child bearing potential)			
Additional Test			
Glucose-6-Phosphate Dehydrogenase (G6PD)			

- Additional tests and/or examinations (apart from mentioned in protocol) may be performed, if necessary, based on principal investigator discretion.
- All results will be assessed against the current laboratory normal ranges at the time of testing and a copy of the normal ranges used will be included in the study documentation.

8.2 EXCLUSION CRITERIA

Volunteers must not be enrolled in the study if they meet any one of the following criteria:

1) History of allergic responses to Primaquine or other related drugs, or any of its formulation ingredients.
2) Have significant diseases or clinically significant abnormal findings during screening [medical history, physical examination (clinical examination), laboratory evaluations, ECG, chest X-ray recording, and, for females, gynaecological history.
3) Any disease or condition like diabetes, psychosis or others, which might compromise the haemopoietic, gastrointestinal, renal, hepatic, cardiovascular, respiratory, central nervous system or any other body system.
4) History or presence of bronchial asthma.
5) Use of any hormone replacement therapy within 3 months prior to the first dose of study medication.
6) A depot injection or implant of any drug within 3 months prior to the first dose of study medication.

7) Use of CYP enzyme inhibitors or inducers within 30 days prior to the first dose of study medication (see http://medicine.iupui.edu/clinpharm/ddis/main-table).
8) History or evidence of drug dependence or of alcoholism or of moderate alcohol use.
9) History of difficulty with donating blood or difficulty in accessibility of veins.
10) A positive hepatitis screen (includes subtypes B & C).
11) A positive test result for HIV antibody and / or syphilis (RPR).
12) Volunteers who have received a known investigational drug within seven elimination half-life of the administered drug prior to the first dose of study medication.
13) Volunteers who have donated blood or loss of blood 50 ml to 100 ml within 30 days or 101 ml to 200 ml within 60 days or >200 ml within 90 days (excluding volume drawn at screening for this study) prior to first dose of study medication, whichever is greater.
14) History of difficulty in swallowing or of any gastrointestinal disease, which could affect drug absorption.
15) Intolerance to venepuncture
16) Any food allergy, intolerance, restriction or special diet that, in the opinion of the principal investigator or sub-investigator, could contraindicate the volunteer's participation in this study.
17) Institutionalized volunteers.
18) Use of any prescribed medications within 14 days prior to the first dose of study medication.
19) Use of any OTC products, vitamin and herbal products, etc., within 7 days prior to the first dose of study medication.
20) Use of grapefruit and grapefruit containing products within 7 days prior to the first dose of study medication.
21) Ingestion of any caffeine or xanthine products (i.e. coffee, tea, chocolate, and caffeine-containing sodas, colas, etc.), recreational drugs, alcohol or other alcohol containing products within 48 hours prior to the first dose of study medication.
22) Ingestion of any unusual diet, for whatever reason (e.g.: low sodium) for three weeks prior to the first dose of study medication.
23) Volunteers with glucose-6-phosphate dehydrogenase (G6PD) deficiency.
24) Volunteers with family or personal history of hemolytic anemia.

9.0 REQUIREMENTS AND RESTRICTIONS FOR THE STUDY

- ***HOUSING REQUIREMENTS*:**

All subjects will be housed in the clinic from at least 11 hours prior to dosing until at least 24 hours post dose in each period. During this period, subjects will not be allowed to leave the clinical facility.

In period I, several extra subjects will be recruited in case there is a short fall of subjects. These subjects will only be dosed to ensure that 50 subjects receive primaquine in this phase. These not dosed will be check-out after dosing of period I.

- ***WASHOUT PERIOD:***

  There will be at least 10 days washout period between each treatment administrations.

- ***STUDY DURATION:***

  Considering the minimum washout period, expected study duration of clinical part is 15 days from the day of check-in of first period.

- ***AMBULATORY VISITS:***

  Subjects will report to the clinical facility at **36.0, 48.0** and **72.0** hours post-dose for compliance assessment, well-being assessment and collection of ambulatory blood samples in each period.

- ***FOOD AND FLUID RESTRICTION AND REQUIREMENTS:***

  The subjects will fast for at least 10 hours prior to dosing until at least 04 hours post-dose in each period. A standard meal will be served at least 04 hours after dosing. Further meals/snacks after 04 hours post dose will be served at appropriate times. Subjects may consume only the food provided while confined in the clinical facility. Meal(s)/snack(s) will be of identical composition and similar quantities will be provided at approximately same times in all the periods.

  Water will be restricted from at least 01 hour prior to dosing until at least 02 hour post-dose in each period (no fluid, except water given with dosing). Free access to water will be allowed outside this interval.

- ***URINE SCAN FOR DRUGS OF ABUSE AND ALCOHOL TEST:***

  Urine scan for drugs of abuse (marijuana-THC, amphetamine-AMP, barbiturates-BAR, cocaine-COC, benzodiazepines-BZO and morphine-MOP) and alcohol test (by blood sample/ urine sample) will be performed on the day of check-in of each period. Subjects must test negative in urine scan for drugs of abuse and alcohol test before check-in of each period.

- ***POSTURE AND ACTIVITY RESTRICTIONS:***

  Subject will remain seated upright for initial 04 hours post-dose and only necessary movement will be allowed during this period. They will not be allowed to lie down except as directed by the physician and secondarily due to adverse events during this restriction period. Thereafter subjects will be allowed to ambulate freely during the remainder of the study.

  Subjects will not engage in any strenuous activity while confined to the clinic and will follow the rules governing their activities as set forth by the clinic.

- ***ADMINISTRATION OF CONCOMITANT MEDICATION:***

  If the subject requires any medication other than specified in the protocol, due to an adverse event or any other reason, decision to administer the concomitant medication will be taken

by the principal investigator or sub-investigator. It will be assessed whether to continue or discontinue the subjects based on the pharmacokinetics and time of administration of the medication.

- ***CONTRACEPTIVE REQUIREMENTS:***

  All study subjects should be advised to use a spermicidal agent along with a barrier method of contraception in addition to their current contraceptive method during the course of the study including the washout period. No hormonal contraceptives or hormonal replacement therapies are permitted in this study.

- ***RESTRICTIONS:***

  Items restricted prior to and during this study are described in the table below. Assessment of compliance to these restrictions will be performed prior to check-in and during ambulatory visit in each period.

Restrictions	Start of Restriction	End of Restriction
Caffeine or xanthine containing products (i.e. coffee, tea, chocolate, and caffeine-containing sodas, colas, etc.)	48 hours prior to dosing in each period	until last blood sample collected in each period
Recreational drugs	48 hours prior to dosing in each period	until last blood sample collected in each period
Alcohol or other alcohol containing products	48 hours prior to dosing in each period	until last blood sample collected in each period
Grapefruit and grapefruit containing products	07 days prior to Period - I dosing	until last study procedure
Unusual diet (e.g.: low sodium)	03 weeks prior to Period - I dosing	until last study procedure
Prescribed medications	14 days prior to Period - I dosing	until last study procedure
OTC products, vitamin and herbal products, etc.	07 days prior to Period - I dosing	until last study procedure

Volunteers who violate any of the above restrictions may be excluded or dropped from the study at the discretion of the investigator. Individual exceptions to the above restrictions may be approved by the sponsor and/or investigator.

10.0 RANDOMIZATION

The order of receiving the investigational product for each subject will be determined according to a randomization schedule. Subjects will be randomized to one of the two sequences: either TR or RT. Equal allocation of the sequence will be ensured. The

randomization schedule will be generated by using SAS® statistical software (Version: 9.4 or higher; SAS Institute Inc., USA).

The randomization schedule will be generated by biostatistician of Cliantha Research.

The randomisation schedule will be kept securely by the study pharmacist or project manager and will be responsible for ensuring subjects are allocated to the correct sequence of primaquine, following an in-house SOP.

The personnel involved in the dispensing of investigational products will be accountable for ensuring compliance to the randomization schedule. The analytical staff concerned will not have access to the randomization schedule during the course of analysis.

11.0 ADMINISTRATION OF INVESTIGATIONAL PRODUCTS

One tablet of either test product Primaquine phosphate tablets USP 15 mg of IPCA or reference product Primaquine 15 mg Tablet of Sanofi will be administered orally to the subjects as per the randomization schedule in a sitting posture with about 240 mL of water at ambient temperature in each period under the supervision of trained study personnel.

This activity will be followed by mouth and hand check of the subjects to assess compliance to dosing.

Investigational products (test or reference) must be swallowed whole and must not be chewed, crushed or divided.

12.0 COLLECTION AND HANDLING OF BLOOD SAMPLES

In each period, total 26 venous blood samples (06 mL each) will be collected at pre-dose (0.0 hour) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0, 48.0** and **72.0** hours post dose in labelled K_2EDTA vacutainers through an indwelling cannula placed in the forearm vein/dorsal aspect of hand of the subjects. If required, a sample may alternatively be collected through a fresh venepuncture.

The pre-dose (0.0 hour) blood sample will be collected within 120 minutes prior to dosing in each period. All post-dose samples in each period will be collected within +2 minutes of the scheduled time; however, ambulatory samples will be collected within +60 minutes of the scheduled time. The actual end-point time of blood collection will be recorded in appropriate data sheet. All deviations outside the range allowed above will be documented as protocol deviations.

Note*: If a mealtime coincides with a blood sample collection time, the collection of blood sample will precede the meal. Occurrence of an AE may necessitate alteration of this sequence.*

The intravenous indwelling cannula will be kept in situ as long as possible during the confinement period. The cannula will be kept patent by injecting 0.5 mL of **normal saline solution** after each sampling. In such cases, blood samples will be collected after discarding the first 0.5 mL of blood containing normal saline from the cannula. Alternatively, blood samples may be withdrawn by a fresh venepuncture using a disposable sterile syringe and a needle at each time of collection.

In addition to the blood taken for pharmacokinetic analysis, blood will be required for screening, study specific tests if any, blood containing normal saline taken prior to each sample collected through the cannula and end study laboratory tests. The volume of blood required from each subject for the study will be as follows:

Description	Blood loss
Blood withdrawn for screening	5.5 mL
Blood loss of pharmacokinetic samples for all periods	312 mL
Discarded blood containing normal saline	23 mL
Blood drawn for CBC on the day of check-in of Period II	02 mL
Blood withdrawn for post study laboratory assessment	5.5 mL
Total blood loss for the study*	**348 mL**

*Note:

Based on investigator discretion, additional blood sample (up to 10 mL) may be collected for additional tests/repeats, sample clots, sample spillage or any other justifiable reason, if deemed necessary.

If alcohol test is performed from blood, additional 2 mL blood sample will be collected in each period in addition to blood loss listed above.

After collection, the blood samples will be placed in an ice bath or other chilling device until centrifugation. Blood samples will be placed in a refrigerated centrifuge within 45 minutes of blood sample collection, and then will be spun at 4000 rpm at 4°C ± 2ºC for 10 minutes. The plasma will then be separated, transferred to labelled polypropylene tubes in duplicates (primary and secondary aliquots with equal volume) and stored in freezer at –20ºC ± 7ºC at the clinical facility until shipment to the analytical facility. The samples will be stored in a freezer at –20ºC ± 7ºC at the analytical facility until analysed. All the temperature excursions will be handled as per in-house SOP.

In case the processing error occurred due to any reason (e.g. mixing of sample while segregation, mechanical failure in centrifuge, etc.) during the sample processing, re-spin the sample (s) under the same conditions, if required.

Note: Transfer of plasma samples into the freezer shall take place as soon as possible so the total elapsed time from blood collection to placement of plasma samples in the freezer does not exceed 90 minutes.

The following blood sampling times will be considered for analysis:

For Primaquine:

0.0 (Pre-dose) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0** and **48.0** hours post dose.

For Carboxyprimaquine:

0.0 (Pre-dose) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0, 48.0** and **72.0** hours post dose.

After completion of last pharmacokinetic blood sample of the study, samples of aliquot# 01 (primary aliquot) will be shipped to analytical facility.

Aliquot# 02 (secondary aliquot) will be forwarded to analytical facility **only after** confirmation of receipt of aliquot# 01 by the analytical facility, if necessary.

Unless the frozen plasma samples are delivered directly from the clinic to the analytical facility, the following procedures will be followed:

All the samples will be shipped frozen at a temperature –20ºC or colder using sufficient quantity of dry ice until receipt at analytical facility. Samples will be shipped by courier to analytical facility as mentioned in the protocol. Temperature loggers will be used during shipment to record the temperature inside the shipment packs.

13.0 ASSESSMENT OF SAFETY

13.1 PRE-STUDY SAFETY ASSESSMENT

The following assessments will be conducted prior to the study

- *Demographic Data:* Date of birth, height, weight, and BMI
- *Vital signs:* Blood pressure, pulse rate, respiration rate, body temperature
- *Medical history and current status:* The volunteer's health status will be determined
- *Medications and therapy:* Determination of any current medication and usage and concomitant therapy in the previous 30 days
- *Physical examination (clinical examination):* A comprehensive physical examination (clinical examination) with ECG and chest X-ray recordings will be conducted. Clinically significant findings will be documented.

  Note: A chest X-ray will be requested only if none available in the past 6 months or if clinically indicated at the time of screening.
- Gynaecological history for the female volunteers.
- *Clinical Laboratory screening:* Blood and urine samples will be tested for standard parameters (as per section# 8.1; Inclusion Criteria).
- *Serum β-HCG test:* For female volunteers of child bearing potential, a serum β-HCG test will be performed during screening.

13.2 SAFETY ASSESSMENT DURING STUDY

A duty physician will be available within the facility throughout the subjects' housing (from check-in to check-out). The following will be done:

Vital signs:

- Sitting blood pressure, pulse rate and body temperature will be measured at the time of check-in and prior to check-out in each period. Subjects must have clinically acceptable vital signs prior to check-in of each period.

- Sitting blood pressure, pulse rate and body temperature will be measured prior to dosing in each period and during the visit for the last study sample.
- Sitting blood pressure and pulse rate will be measured at 2.0 hours (± 40 minutes) post dose in each period
- Sitting blood pressure, pulse rate and body temperature will be measured at 6.0 and 10.0 hours (± 40 minutes) post dose and as needed in each period.

Physical examination (clinical examination) of the subject will be conducted by a qualified medical designate at the time of check-in and prior to check-out in each period and during the visit for the last study sample.

Well-being assessment:

Subjects will be advised to report any AE that may occur during the study and will be specifically asked for these by trained study personnel in a non-leading manner at the time of physical examination (clinical examinations), during vital signs recording, at about 16.0 and 24.0 hours post dose, during ambulatory visits and as needed in each period. Adverse events will be managed and recorded as appropriate by the investigator or available physician.

A CBC will be measured prior to check in of period II. Only subjects within normal limits or clinically insignificant falls will be dosed in period II.

A urine pregnancy test will be performed prior to check-in of each period and serum (β-HCG) pregnancy test will be performed at the time the end of study laboratory assessment for female volunteers of child bearing potential. Volunteers with a negative test result for pregnancy (prior to check-in of each period) will only be dosed.

In the event of detection of any abnormality during safety assessments, the clinical investigator must be consulted for necessary action, which will be recorded.

All study subjects who are dosed, will be assessed for their well-being through a physical examination (clinical examination) which will include vital sign measurement (sitting blood pressure, pulse rate and body temperature) and the following laboratory tests at the end of the study or as applicable.

Hematology				
Haemoglobin	Total RBC count	Total WBC count	Platelet count	
Differential leukocyte count:				
Neutrophils	Lymphocytes	Eosinophils	Monocytes	Basophils
Blood indices: HCT				
Biochemistry				
BUN	Serum creatinine	Random glucose	SGPT & SGOT	
Alkaline phosphatase	Uric acid	Serum bilirubin		
Serum total protein: Total proteins, Albumin				
Serum electrolytes: Serum sodium, serum chloride, serum potassium, serum phosphorous,				

serum calcium
Serum (ß-HCG) pregnancy test (for female of child bearing potential)

13.3 SAFETY ANALYSIS

Any clinically significant results in the end study parameters will be listed in the clinical report. All the clinically significant parameters will be considered as adverse events and followed up until resolution, or until the event is otherwise explained, or until the subject is lost to follow-up. The frequency of follow-up tests to be repeated will be decided by the investigator or physician on a case-by-case basis depending on the severity and/or seriousness of the event.

14.0 ADVERSE EVENT REPORTING

14.1 DEFINITIONS

The term adverse event (AE) is defined as any untoward medical occurrence in a clinical investigation patient or a subject who is administered an investigational medicinal product, which does not necessarily have a causal relationship with the investigational medicinal product treatment.

An adverse event can therefore be any unfavourable or unintended sign (including a clinically significant laboratory test finding), symptom, or disease temporally associated with use of a medicinal (investigational) product, whether or not considered related to the medicinal (investigational) product. This includes all events both expected and unexpected (the nature or severity of which is not consistent with information in the relevant source document (i.e. Investigator's brochure or reference product labelling).

An adverse event may be:

- a new illness
- worsening of a concomitant illness
- an effect of the study investigational product, including the reference product labelling; it could be a clinically significant laboratory value which the investigator or medically qualified designate considers to be clinically important
- a combination of two or more of these factors

Medical events occurring prior to the administration of first dose of investigational product are not considered to be adverse events since no investigational product has been administered yet.

Planned surgical procedures themselves are not adverse events. They are therapeutic measures for conditions that required surgery. The condition for which the surgery is required is an adverse event, if it occurs or is detected during the study. If the condition(s) was (were) known before the start of study treatment, it (they) should have been recorded in the medical history section of the CRF.

Adverse events may be considered by the investigator to be adverse reactions if a causal relationship to the investigational medicinal product is at least a reasonable possibility, i.e.

the relationship cannot be ruled out.

14.2 ADVERSE EVENT MONITORING

Subjects will be monitored throughout the study after the enrolment period for adverse events (AE) until the visit when post-study procedures are conducted. Subjects will be advised to report any AE and will be specifically asked in a non-leading manner about any AE at regular periods as indicated. Necessary treatment of an AE will be performed by the investigator or physician and recorded. The investigator or physician is responsible for assessing seriousness and causality of every AE.

14.3 CLASSIFICATION OF ADVERSE EVENTS WITH RESPECT TO INTENSITY

The intensity of an AE is classified according to the following criteria:

- Mild: Awareness of signs or symptoms that are easily tolerated and do not limit usual activities; the subject may experience slight discomfort.
- Moderate: Discomfort sufficient to cause interference with normal activities.
- Severe: Incapacitating or preventing with inability to perform normal activities or require complex medication or hospitalization.

Note: Severity is not a measurement of seriousness.

Each degree of intensity occurring within the course of observation of an untoward medical occurrence will be documented. However these will be grouped together and evaluated as a single AE and the associated severity reported as the maximum intensity reached. An AE classified as severe may not be necessarily defined as serious.

14.4 CLASSIFICATION OF CAUSALITY OF ADVERSE EVENTS

The investigator must evaluate every AE experienced during the study for its relation to the drug administered. The causal relationship of an AE with the investigational product is classified as follows:

A) Certain

- Event or laboratory test abnormality, with plausible time relationship to drug intake
- Cannot be explained by disease or other drugs
- Response to withdrawal plausible (pharmacologically, pathologically)
- Event definitive pharmacologically or phenomenologically (i.e. an objective and specific medical disorder or a recognized pharmacological phenomenon)
- Rechallenge satisfactory, if necessary

B) Probable/Likely

- Event or laboratory test abnormality, with reasonable time relationship to drug intake
- Unlikely to be attributed to disease or other drugs
- Response to withdrawal clinically reasonable

- Rechallenge not required

C) Possible

- Event or laboratory test abnormality, with reasonable time relationship to drug intake
- Could also be explained by disease or other drugs
- Information on drug withdrawal may be lacking or unclear

D) Unlikely

- Event or laboratory test abnormality, with a time to drug intake that makes a relationship improbable (but not impossible)
- Disease or other drugs provide plausible explanations

E) Conditional/Unclassified

- Event or laboratory test abnormality
- More data for proper assessment needed, or
- Additional data under examination

F) Unassessable/Unclassifiable

- Report suggesting an adverse reaction
- Cannot be judged because information is insufficient or contradictory
- Data cannot be supplemented or verified

Note: The terms Conditional/Unclassified and Unassessable/Unclassifiable would be considered as 'not related' with respect to relationship to study drug.

Subjects will be monitored throughout the study period for adverse events. They will be asked to bring to the notice of the study personnel, doctor or nurse any adverse event that may occur during their stay. Subjects will also be specifically asked about any adverse event throughout their stay in the clinic, at the time of check in and at the time of check out and during ambulatory visit and for subsequent periods. Medically qualified personnel will be available round the clock during the time of housing in the facility. A consultant physician will remain on call during the conduct of the study.

Details of consultant physicians are as follows:

CLIANTHA RESEARCH	
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All adverse events will be managed at/by Cliantha Research and will be followed up until considered resolved, or the condition stabilizes, or the event is otherwise explained, or the subject is lost to follow-up. This may involve additional visits. An adverse event may be managed at the clinical facility or a nearby hospital as required.

For proper assessment and management of adverse events, adequate arrangements and actions which include regular monitoring of subjects by a doctor during the housing period, management and referrals to a hospital or a specialty unit will be made available.

14.5 ASSESSMENT OF SERIOUSNESS

An adverse event is considered to be serious if any of the following criteria are met:

- results in death
- is life threatening (Note: the term 'life threatening' in the definition of serious refers to an event in which the patient was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe)
- results in hospitalization or prolongation of existing hospitalization
- results in persistent or significant disability or incapacity
- is a congenital anomaly / birth defect
- is a medically important event that may jeopardize the subject or may require intervention to prevent one or the other outcomes listed above, according to the clinical judgment of the investigator.

14.6 DOCUMENTATION OF ADVERSE EVENTS

Each adverse event shall be recorded on an 'Adverse Event Record' form irrespective of its association with the ongoing study medication. The IEC and the sponsor shall be informed regarding the same as necessary.

The investigator, and/or trained and qualified staff at Cliantha Research will record all conditions/symptoms/findings both expected (known pharmacological response) and unexpected, occurring from administration of first dose of study medication until the completion of all post-treatment procedures. All study related adverse events will be followed up until resolution, or until the condition stabilizes, or until the event is otherwise explained, or until the subject is lost to follow-up. All abnormal laboratory tests that are considered clinically significant (CS) by the investigator/physician will be reported as adverse events. The clinical investigator is responsible for determining the relationship to study drug and seriousness.

For the purposes of this study, the period of observation of adverse events extends from the start of treatment with the investigational products until the completion of all post-treatment procedures. During this period, all adverse events spontaneously reported by the subject, observed by the investigator / clinical staff or elicited by general questioning of the subjects will be documented in the CRF/adverse event record form and will be listed in the report. Each adverse event shall be evaluated for duration, severity, action taken, date and time of resolution and association with the study treatment.

14.7 REPORTING OF ADVERSE EVENTS

A summary of all adverse events recorded will be sent to the sponsor at the conclusion of study.

The Investigator will report all serious adverse events (including death, irrespective of the reason) to Central licensing authority, sponsor or his representative and Ethics Committee by any available mode of communication within 24 hours of their occurrence. The report of serious adverse event of death, after due analysis shall be forwarded to sponsor, Central licensing authority, Chairperson of Ethics Committee and Head of the Institution where the study has been conducted within 14 calendar days of knowledge of occurrence/reporting. The report of serious adverse events other than death, after due analysis will be forwarded to the sponsor, Central licensing authority, Chairperson of Ethics Committee and Head of the Institution where the study has been conducted within 14 calendar days of knowledge of occurrence/reporting. In case the investigator fails to report serious adverse events within stipulated period, investigator shall furnish the reason for the delay to the satisfaction of Central licensing authority along with report of serious adverse events.

The sponsor or his representative will report the serious adverse event(s) to the Central licensing authority, Chairperson of Ethics Committee and Head of the Institution where the study has been conducted within 14 calendar days of knowledge of occurrence/reporting.

The Ethics Committee will inform to the applicable regulatory authorities as per the required timelines, as applicable.

The written report will include a detailed description of the observed symptoms, laboratory tests and treatment of the event. Subject demographic information, medical history, concomitant medication and date/time of occurrence and report of the event must also be clearly stated. The investigator will judge the possible causal relationship between the event and the investigational medicinal product. All communication of AE information will be in English.

The study may be suspended or terminated depending on the frequency and seriousness of the adverse events encountered during the study.

14.8 REPORTING OF PREGNANCY

All pregnancy cases encountered from the start of treatment with the investigational products (whenever reported or observed) till the collection of the last sample in the study and completion of all post-treatment procedures will be recorded. All pregnancies will be followed up until completion/termination of the pregnancy. The investigator will inform the sponsor and Independent ethics committee of these events within 24 hours of being aware. An immediate report must be followed by a detailed written report (Pregnancy Notification Form) on the event. If the pregnancy continues to term, the outcome (health of the infant) must also be reported.

While pregnancy itself will not be considered an AE or SAE, however any complication during pregnancy or elective termination of a pregnancy for medical reasons will be recorded as an AE or a SAE. A spontaneous abortion will always be considered a SAE and will be reported to the sponsor. Elective termination (i.e., without medical reasons) of an

uncomplicated pregnancy will be considered an elective procedure and not an adverse event.

15.0 ANALYTICAL METHODOLOGY

Plasma samples will be assayed by a validated LCMS/MS method developed at Cliantha Research, Ahmedabad, which is specific for the determination of Primaquine and metabolite Carboxyprimaquine.

Repeat analysis will be performed as per in-house SOP of Cliantha Research.

Incurred sample reanalysis will be performed as per in-house SOP of Cliantha Research.

Note: All the instruments to be used in the study should be pre-qualified with precision and accuracy batch (With quality control samples at higher level, middle level and lower level) before study sample analysis.

The following blood sampling times will be considered for analysis:

For Primaquine:

0.0 (Pre-dose) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0** and **48.0** hours post dose.

For Carboxyprimaquine:

0.0 (Pre-dose) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.333, 2.667, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, **36.0, 48.0** and **72.0** hours post dose.

16.0 SAMPLES TO BE ANALYZED

All available samples will be analysed (if any subject is discontinued and no post dose samples are collected, then pre-dose sample of that period of such subject would not be analysed).

16.1 SAMPLE STORAGE:

PK samples will be stored at Cliantha's facility to allow for possible re-analysis if requested by the WHO. If there is no request for re-analysis, the samples will be destroyed.

17.0 PHARMACOKINETIC AND STATISTICAL ANALYSIS

17.1 PK POPULATION:

PK population is defined as below for evaluation of bioequivalence:

- Subjects who complete all periods of the study, and who are not excluded by pharmacokineticist will be included in the bioequivalence evaluation.
- Data from any subject with missing concentration values in any period (like missed blood samples, lost samples, samples unable to be quantitated) may be used for bioequivalence evaluation if primary pharmacokinetic parameters can be reliably estimated using the remaining data points otherwise that specific pharmacokinetic parameter data will be excluded from the bioequivalence evaluation.

- If the pre-dose concentration appears to be >5% of the Cmax in any subject in any period, then the subject will be excluded from the bioequivalence evaluation.
- Subjects' pharmacokinetics data will be excluded from statistical considerations if in any subject there is lack of any measurable concentrations or there are only very low plasma concentrations for reference product. A subject is considered to have very low plasma concentrations if its AUC for reference product is less than 5% of geometric mean AUC of reference product (which should be calculated without inclusion of data from the outlying subject).
- If subject have missing sample at 72.0 hrs then data (i.e AUC72) of that subject will be excluded for that particular period for Carboxyprimaquine only.
- Kel and related elimination phase parameters will not be estimated for cases that do not exhibit a terminal log-linear phase in the concentration versus time profile.

17.2 PHARMACOKINETIC ANALYSIS

The following pharmacokinetic parameters will be determined from the time and concentration data using a non-compartmental analysis of Phoenix® WinNonlin® professional software (Version 8.1.1 or higher; Pharsight Corporation, USA) or SAS® statistical software (Version: 9.4 or higher; SAS Institute Inc, USA).

All Below Limit of Quantitation (BLQ) concentration values will be set to zero before pharmacokinetic analysis.

The actual time of blood collection for all samples collected will be used for the calculation of pharmacokinetic parameters.

For Primaquine below pharmacokinetic parameters will be calculated

Primary Pharmacokinetic Parameters:		
Cmax	:	Maximum measured plasma concentration over the time span specified.
AUCt	:	The area under the plasma concentration versus time curve will be calculated using the linear trapezoidal rule from the zero time point to the last quantifiable concentration.
Secondary Pharmacokinetic Parameters		
AUCi	:	The area under the plasma concentration versus time curve from zero to infinity will be calculated by adding Ct/Kel to AUCt, where Ct is the last quantifiable concentration and Kel is the elimination rate constant.
Tmax	:	Time of the maximum measured plasma concentration. If the maximum plasma concentration occurs at more than one time point, the first is chosen as Tmax.
Kel	:	The terminal elimination rate constant will be obtained from the slope of the line, fitted by linear least squares regression, through the terminal points of the natural log of the concentration versus time plot for these points

tHalf	:	The half-life will be calculated by the equation tHalf = 0.693/ Kel

T/R ratio will be reported for Cmax, AUCt and AUCi.

AUC_%Extrap_obs, REGSTART, REGEND and NUMPT will be reported.

For Carboxyprimaquine below pharmacokinetic parameters will be calculated.

Primary Pharmacokinetic Parameters:		
Cmax	:	Maximum measured plasma concentration over the time span specified.
AUC72	:	The area under the plasma concentration versus time curve will be calculated using the linear trapezoidal rule from the zero time point to 72 hrs.
Secondary Pharmacokinetic Parameters		
Tmax	:	Time of the maximum measured plasma concentration. If the maximum plasma concentration occurs at more than one time point, the first is chosen as Tmax.

T/R ratio will be reported for Cmax and AUC72.

17.3 STATISTICAL ANALYSIS:

Statistical analysis will be performed on population defined in section 17.1 using SAS® statistical software (Version: 9.4 or higher; SAS Institute Inc, USA).

- *Descriptive statistics:* Arithmetic mean, geometric mean, standard deviation, coefficient of variance, median, maximum and minimum for all applicable pharmacokinetic parameters will be calculated.
- *Analysis of variance:* Ln-transformed data of Cmax, AUC72 and AUCt will be evaluated statistically using the PROC GLM from SAS® for difference due to treatment, period, sequence and subject(sequence) as fixed effects.

  Treatment and period will be tested using Mean Square Error and Sequence will be tested using Subject (sequence) as the error term at 5% level of significance.
- *90% confidence interval estimation:*

  The statistical method for testing bioequivalence will be based on the determination of the 90% confidence interval around the ratio of the Ln-transformed population means (Test/Reference) for the primary PK parameters Cmax, AUC72 and AUCt. This method is equivalent to carrying out two one-sided tests with the null hypothesis of bioinequivalence at the 5% significance level.
- Variability and power:

  Intra-subject variability and power (using Two one-sided test method) will be computed and reported for ln-transformed Pharmacokinetic parameters for Cmax, AUC72 and AUCt.

- Bioequivalence criteria:

  The 90% confidence interval of the relative mean (geometric least square mean) of the test to reference product for Ln-transformed Pharmacokinetic parameters Cmax, and AUCt should be within 80.00% to 125.00% to establish bioequivalence for Primaquine. Data of inactive metabolite of Carboxyprimaquine will be provided as supportive data.

17.4 PRESENTATION OF DATA:

All available plasma concentration data will be presented.

Pharmacokinetic data calculated from all available plasma concentration data will be presented.

Individual and mean plots (Linear and semi logarithmic) will be presented.

18.0 HANDLING OF INVESTIGATIONAL PRODUCTS

The sponsor will supply a sufficient quantity of investigational products for administration to the subjects as well as retention purpose. The investigational products shall be supplied preferably in sealed packages or strips with appropriate label and certificate of analysis.

The shipment of investigational products in appropriate condition will be ensured by the sponsor. If the shipment is accompanied with temperature data logger, the temperature log readings should reach Cliantha Research prior to first dose administration of the investigational product and the records will be compiled with the study data.

It is the responsibility of the sponsor to ensure that all drug supplies provided for the study are manufactured under current Good Manufacturing Practices (cGMP) and are suitable for human use.

The investigational products will be stored securely under conditions as specified by the sponsor on label. The investigational products will be dispensed according to the randomization schedule. The remaining investigational products will be kept in their original containers as retention samples as per applicable regulatory requirements.

Adequate records for receipt, accountability, usage and disposition or return of investigational products will be maintained at Cliantha Research.

19.0 SUBJECT WITHDRAWAL OR DISCONTINUATION CRITERIA

The investigator may withdraw a subject from the study for any of the following reasons.

- The subject suffers from significant intercurrent illness or undergoes surgery during the course of the study or the subject has any significant symptoms or signs during the course of the study
- Any subject found to have entered the study in violation of this protocol. This will include non-compliance to pre-study directions regarding diet, alcohol and drug use, or if the subject is uncooperative during the study
- Any subject who requires the use of an unacceptable concomitant medication.
- If, in the opinion of the investigator, it is not in the subject's best interest to continue.

- Any subject who wishes to withdraw the consent from the study.
- Any other justifiable reason, which should be adequately documented.
- After dosing, if the subject vomits at or before 2 times the median Tmax (2 X 2.0 hours) of Primaquine in any period.

Subject withdrawal during the study shall be handled as per the in-house procedure with adequate documentation. If a subject is withdrawn based on an adverse event, subject will be followed until the event has resolved and all the safety data normally required at the end of the study is obtained as far as possible.

REPLACEMENT OF SUBJECTS

Subjects once dosed will not be replaced.

Any subject withdrawn/dropped out from the study due to any reason prior to dosing of period-I will be replaced with another subject that will be enrolled with a same subject number as that of withdrawn/dropped subject.

20.0 ACCESSIBILITY AND CONFIDENTIALITY OF DATA

Direct/remote access of the raw data will be limited to the concerned study personnel, IEC, sponsor's monitor, auditor, and the regulatory agencies for inspection and audits/monitoring.

The study can be monitored at the discretion of the sponsor by any of its representatives. The data that contains subject's personal information will be maintained as confidential documents as far as possible and will be accessible only to the concerned study personnel and quality assurance auditors during audits. IEC, sponsor/ its representatives and regulatory authorities can be allowed direct/remote access to such data, if they demand during inspection or audits/monitoring of the study, but must agree to respect the confidentiality of the data.

The study will comply with the Data Protection Act 2018, which requires that personal data must not be kept as identifiable data for longer than necessary for the purposes concerned.

Anonymised data may be shared in the future with other researchers, according to MORU data sharing policy.

21.0 QUALITY CONTROL AND QUALITY ASSURANCE

Internal quality assurance (QA) / quality control (QC) measures in built in Cliantha Research's processes from protocol development, study conduct to finalization of the study report. Details of the QA/QC processes employed are outlined in the respective in-house SOP(s) of Cliantha Research. An external audit may also be conducted.

Study activities including the raw data generated during the study conduct and the final report will be liable for quality audits for conformance to this protocol, compliance to all governing in-house SOPs and applicable regulatory requirements.

22.0 ETHICS

22.1 STATEMENT OF COMPLIANCE

The study will commence only after written approval is obtained from the IECs, which will also the Sponsor's ethics committee, OxTREC.

The study will be conducted in accordance with this protocol, relevant in house SOPs, pertinent requirements of the 'WHO', ICMR ethical guidelines and ICH 'Guidance on Good Clinical Practice', 'Principles of Good Laboratory Practice', 'New Drugs and Clinical Trials Rules, 2019', 'Declaration of Helsinki' and all applicable guidelines and regulations.

22.2 INDEPENDENT ETHICS COMMITTEE

This protocol, any modifications, and corresponding informed consent document will be reviewed by the IEC prior to study initiation. The study will not be initiated without approval from the IEC.

All revisions and amendments to the protocol must be approved by the sponsor and the IEC. All revisions and amendments to the informed consent must be approved by the IEC.

22.3 INFORMED CONSENT AND CONSENT DOCUMENT

Volunteers will undergo informed consent procedure for screening prior to screening procedure and will render their consent by reading, signing and dating an informed consent document for screening.

The volunteers will undergo informed consent procedure for the study prior to check-in of period-I.

The study will be explained to all prospective volunteers by the principal investigator or person designated by the principal investigator. The explanation will describe all pertinent aspects of the study stated in the ICD including the drug product being evaluated, the potential hazards involving drug allergies and possible adverse reactions.

Each participant will acknowledge receipt of this information and confirm their freely rendered offer to volunteer in this study by reading, signing and dating an informed consent document.

In case of illiterate volunteers, their thumb impression may be obtained if they are unable to sign. In such a case, an impartial witness must be present during the entire informed consent procedure. After the informed consent procedure and after the volunteer has orally consented for participation in the study, the impartial witness will personally sign and date the consent document. By signing the consent document, the impartial witness attests that the information in the consent document and any other written information was accurately explained to, and apparently understood by, the volunteer and that informed consent was freely given by the volunteer.

A copy of the signed ICD will be provided to study subjects for their reference; however, the original document will be maintained at Cliantha Research as a part of the study documentation.

22.4 VOLUNTEER COMPENSATION

Subjects will not be paid for any loss of wages for the days of their participation; however adequate compensation will be given to subjects in appropriate proportion at the end of each of the periods of the study as per compensation policy of Cliantha Research as approved by the IEC.

This compensation is a total of 15,150 rupees (just under 160 pounds sterling).

In case of drop-out or withdrawal of a subject before completion of the study, compensation will be provided as per the compensation policy of Cliantha Research.

Events	Compensation to be paid to subjects
• At the end of Period – I (approximate 25% of total amount)	Rs. 3800
• At the end of Period – II (approximate 65% of total amount)	Rs. 9900
• At the time of end study assessment (remaining amount of total compensation)	Rs. 1450

Subjects will receive full compensation if they are withdrawn from the study by the investigator on medical grounds to safeguard your safety e.g. a primaquine related AE, or are withdrawn on compassionate grounds.

The extra subjects not dosed during phase I will receive the compensation during check-out phase I (as per compensation policy).

All other subjects will receive compensation for the days that they participated in the study. This will be reduced to 50% for those subjects who do not follow the protocol.

23.0 RECORDING AND HANDLING OF DATA

The screening related data will be directly recorded in MSR and the study related data generated during the conduct of the study will be directly recorded in the respective CRF which will be considered as source data and will kept along with other source data (e.g. X ray reports, ECG, laboratory reports, bioanalytical data, etc.). CRF can also contain data transcribed from other source documents (as appropriate). All CRFs must be completed using black ink or typed. Corrections of data on the CRFs are to be made only by drawing a single line through the incorrect data and writing the correct data next to the incorrect data. Each correction must be initialed and dated by the person making the correction. The investigator(s) will assume responsibility for ensuring the completeness and accuracy of all clinical documents. Electronic CRFs may be used. If computerized systems are used to create, modify, maintain, archive, retrieve or transmit source data, they must comply with the applicable regulatory regulations and/or guidance.

The computer-generated chromatograms will also be treated as raw data. All raw data will be checked for correctness and completeness.

All data generated in connection with this study, together with a copy of this protocol and signed ICDs and the final report will be retained at Cliantha Research / third party archive facility, as per applicable regulatory requirements.

24.0 CLINICAL DATA MANAGEMENT

Data Management will be performed by Cliantha Research using CodeAngelo CDMS tool (a proprietary web-based tool of Cliantha research). Data entry screens will be designed to look like paper forms for data entry and Data Manager will be responsible for development, testing, and maintenance of clinical database and eCRFs. User Acceptance Testing will be performed before making the study live for data entry. Once data entry is completed, activities like data validation, quality review, medical coding, lab data reconciliation, data base lock and datasets export etc. will be performed. Integrated query management system, all queries will be tracked and maintained in the EDC system. Detailed Data Management activities and process will be explained in the study specific Data Management Plan (DMP) and DMP will be approved by the sponsor.

25.0 FINANCE AND INSURANCE

25.1 FINANCE

This study is being entirely funded by UK Medical Research Council. It has had no role in the design of this study and will not have any role during its execution, analyses, interpretation of the data, or decision to submit the results.

25.2 INSURANCE

The study will be covered by an insurance contract with 'The New India Assurance Co. Ltd'.

The Masters & Scholars of the University of Oxford has a specialist insurance policy in place which would operate in the event of any participant suffering harm as a result of their involvement in the research (Newline Underwriting Management Ltd, at Lloyd's of London).

26.0 REPORT

All appropriate data from the study will be included in a final report. The report will contain data regarding the analytical methodology and the chromatography (including at least 20% of the serially selected subject chromatograms), the pharmacokinetic calculations, the statistical analysis of the data, and a clinical report along with raw data. The final report will be in an ICH-E3 format as per the structure and content of clinical study reports guidelines.

The final compiled/integrated (clinical, bioanalytical, pharmacokinetic and statistical) report will be provided by Cliantha Research.

27.0 PUBLICATION POLICY & AUTHORSHIP

The results of the study will be the property of the sponsor and no one can publish study data

without permission of the sponsor. The results will be published and authorship will follow international guidelines. The investigator must receive written permission from the sponsor prior to publication of any study related data.

28.0 TERMINATION OF THE STUDY

If the study is prematurely terminated or suspended for any reason, the investigator/ Cliantha Research will inform the study subjects, sponsor & the IEC. The investigator/Cliantha Research will assure appropriate therapy and follow-up for the study subjects.

(i) The sponsor reserves the right to discontinue the study at any time.

(ii) The principal investigator reserves the right to discontinue the study at any time for safety reasons.

(iii) The IEC may terminate the approval of the study for safety and ethical reasons.

Reasons for the termination/suspension will be provided to the study subjects, sponsor and IEC. Also the reasons for the termination/suspension will be provided to the applicable local regulatory authority in 30 working days or as per the required timelines of applicable regulatory requirements.

29.0 ADHERENCE TO PROTOCOL

Any significant changes in the study procedure or study design will only be effected upon mutual agreement between the sponsor and Cliantha Research and after obtaining a favorable opinion from the ethics committee. All such changes will be documented in the amended version of the protocol and a list of changes made with reference to previous version will be generated and submitted to the IEC along with the amended version of the protocol.

An exception can be made where the principal investigator considers that such a change is necessary to eliminate an immediate hazard to one or more study subjects. In such a case, the change may be implemented prior to approval by the other parties, however, the sponsor and IEC must be informed as soon as possible. Any deviation from the protocol will be recorded and explained. The information on the protocol deviations will be included in the clinical study report.

30.0 REFERENCES & CITED LITERATURE

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31.0 APPENDICES

i. Study Meal Plan

ii. Clinical Laboratory Parameters Range and Pre-defined Site Normal Range

iii. List of changes

APPENDIX-I: STUDY MEAL PLAN

Check-In Night Dinner (Day -1)

Food items	*Servings*	*Ingredients*	*Qty. of Ingredients*	*Energy (Calories)*	*Carbohydrates (grams)*	*Fats (grams)*	*Proteins (grams)*
ROTI	6 NOS. / 120 gm	Wheat flour	120 gm	375.276	77.004	1.836	12.684
		oil	2ml	18.000	0.000	2.000	0.000
RICE	1 Bowl/ 150 gm	Rice	45 gm	157.230	35.208	0.234	3.573
DAL TUVER	1 Bowl/ 150 gm	Tuver	30 gm	96.528	16.569	0.468	6.510
		Oil	1 ml	9.000	0.000	1.000	0.000
		Tomato	20 gm	3.618	0.640	0.050	0.152
		Onion	20 gm	9.280	1.912	0.048	0.300
PAPAD	1 NOS.	Udad Dal	7 gm	22.798	3.570	0.118	1.864
SWEET (MOHANTHAL)	1 NOS./ 40 gm	Besan	30 gm	96.261	14.016	1.593	6.465
		Ghee	10 gm	90.000	0.000	10.000	0.000
		Sugar	20 gm	79.984	19.996	0.000	0.000
PANEER MIX VEG.	1 Bowl/ 200 gm	Potato	50 gm	33.895	7.445	0.115	0.770
		Tomato	20 gm	3.618	0.640	0.050	0.152
		onion	20 gm	9.280	1.912	0.048	0.300
		Capsicum	20 gm	2.972	0.368	0.068	0.222
		Peas	15 gm	11.647	1.782	0.019	1.087
		Paneer	30 gm	77.430	3.723	4.434	5.658
		Oil	5 ml	45.000	0.000	5.000	0.000
Total				**1141.817**	**184.785**	**27.081**	**39.737**
Total Nutrients Calories				**NA**	**739.140**	**243.729**	**158.948**
Total Nutrients Calories in Percentage of total				**NA**	**64.734**	**21.346**	**13.921**

Note:

- 1 gram of Carbohydrates and Protein is equal to 4 calorie and 1 gram of Fat is equal to 9 calorie.
- Salt, Black pepper, Chaat Masala, Onion slices may be served as taste enhancers.
- Curry leaves, condiments & spices can be used as per requirement.
- Nutritive value source: Nutritive value of Indian foods, NIN, ICMR, Hyderabad 2017.

Lunch (Day 0)

Food items	*Servings*	*Ingredients*	*Qty. of Ingredients*	*Energy (Calories)*	*Carbohydrates (grams)*	*Fats (grams)*	*Proteins (grams)*
Jeera Rice	1 Bowl/ 150 gm	Rice	50 gm	174.700	39.120	0.260	3.970
		oil	2 ml	18.000	0.000	2.000	0.000
Kadai Paneer	1 bowl / 200 gm	Paneer	50 gm	129.050	6.205	7.390	9.430
		Capsicum	10 gm	1.486	0.184	0.034	0.111
		Tomato	20 gm	3.618	0.640	0.050	0.152
		Onion	20 gm	9.280	1.912	0.048	0.300
		Carrot	10 gm	3.023	0.555	0.047	0.095
		French Beans	10 gm	2.302	0.268	0.026	0.249
		Oil	5 ml	45.000	0.000	5.000	0.000
Roti	7 nos./ 150 gm	wheat flour	150 gm	469.095	96.255	2.295	15.855
		oil	2 ml	18.000	0.000	2.000	0.000
Dal Makhani	1 bowl / 150 gm.	Black Gram Hole	30 gm	83.418	13.197	0.474	6.591
		Tomato	20 gm	3.618	0.640	0.050	0.152
		Onion	20 gm	9.280	1.912	0.048	0.300
		Oil	5 ml	45.000	0.000	5.000	0.000
Curd	1 bowl / 100 gm.	Curd	100 gm	73.120	4.940	4.480	3.260
Total				**1087.990**	**165.828**	**29.202**	**40.465**
Total Nutrients Calories				**NA**	**663.312**	**262.818**	**161.860**
Total Nutrients Calories in Percentage of total				**NA**	**60.967**	**24.156**	**14.877**

Note:

- 1 gram of Carbohydrates and Protein is equal to 4 calorie and 1 gram of Fat is equal to 9 calorie.
- Salt, Black pepper, Chaat Masala, may be served as taste enhancers.
- Nutritive value source: Nutritive value of Indian foods, NIN, ICMR 2017, Hyderabad.
- Curry leaves, condiments & spices can be used as per requirement.

Snack (Day 0)

Food items	*Servings*	*Ingredients*	*Qty. of Ingredients*	*Energy (Calories)*	*Carbohydrates (grams)*	*Fats (grams)*	*Proteins (grams)*
Kachori khasta	150 gm	Maida (refined flour)	50 gm	172.680	37.135	0.380	5.180
		Moong dal	20 gm	63.606	10.518	0.270	4.776
		Oil	5 ml	45.000	0.000	5.000	0.000
chutney	2 tsp	coriander	30 gm	8.430	0.579	0.210	1.056
		Roasted Chana (dadia)	10 gm	32.087	4.672	0.531	2.155
Total				**321.803**	**52.904**	**6.391**	**13.167**
Total Nutrients Calories				**NA**	**211.616**	**57.519**	**52.668**
Total Nutrients Calories in Percentage of total				**NA**	**65.759**	**17.874**	**16.367**

Note:

- 1 gram of Carbohydrates and Protein is equal to 4 calorie and 1 gram of Fat is equal to 9 calorie.
- Curry leaves, condiments & spices can be used as per requirement.
- Tomato ketchup may be served as taste enhancer.
- Nutritive value source: Nutritive value of Indian foods, NIN, ICMR, Hyderabad 2017

Dinner (Day 0)

Food items	*Servings*	*Ingredients*	*Qty. of Ingredients*	*Energy (Calories)*	*Carbohydrates (grams)*	*Fats (grams)*	*Proteins (grams)*
ROTI	6 nos./ 120 gm	Wheat flour	120 gm	375.276	77.004	1.836	12.684
		Oil	5 ml	45.000	0.000	5.000	0.000
KHICHADI	1 bowl / 150 gm	Rice	30 gm	104.820	23.472	0.156	2.382
		Tuver Dal	10 gm	32.176	5.523	0.156	2.170
SALAD	Half bowl/40 gm	Cucumber	20 gm	3.244	0.564	0.036	0.166
		Onion	20 gm	9.280	1.912	0.048	0.300
KADHI	1 bowl./ 150 gm	Curd	50 gm	36.560	2.470	2.240	1.630
		Besan	10 gm	32.087	4.672	0.531	2.155
		oil	2ml	18.000	0.000	2.000	0.000
POTATO-CABBAGE VEG.	1 bowl / 100 gm.	Cabbage	100 gm	19.520	3.250	0.120	1.360
		Potato	50 gm	33.895	7.445	0.115	0.770
		Tomato	10 gm	1.809	0.320	0.025	0.076
		Oil	5 ml	45.000	0.000	5.000	0.000
Total				**756.667**	**126.632**	**17.263**	**23.693**
Total Nutrients Calories				**NA**	**506.528**	**155.367**	**94.772**
Total Nutrients Calories in Percentage of total				**NA**	**66.942**	**20.533**	**12.525**

Note:

- 1 gram of Carbohydrates and Protein is equal to 4 calorie and 1 gram of Fat is equal to 9 calorie.
- Curry leaves, condiments & spices can be used as per requirement.
- Salt, Black pepper, Chaat Masala may be served as taste enhancers
- Nutritive value source: Nutritive value of Indian foods, NIN, ICMR, Hyderabad 2017.

APPENDIX-II: CLINICAL LABORATORY PARAMETERS RANGE AND PRE-DEFINED SITE NORMAL RANGE

Healthy Volunteers are to be enrolled in the study based on the defined inclusion and exclusion criteria. Evaluating a volunteers' eligibility or safety in a clinical study will be done with reference to past/ current medical history, physical examination (clinical examination), clinical laboratory parameters, X ray, 12 -lead ECG and other relevant tests by a medical doctor and is a clinical decision.

At all times it is important to use scientific judgment and clinical correlation with consideration that subject's safety and wellbeing are of prime importance. Volunteers whose laboratory test values are within pre-defined site normal range will be enrolled in the study.

Blood Chemistry					
Test Name	**Unit**	**Reference Range**	**Clinical Relevant Direction**	**Acceptable (Approx. %)**	**Pre-defined site normal range**
Blood Urea Nitrogen (BUN)	mg/dL	6.00 – 20.00	Increase	-	0 to 23.0
Serum Creatinine	mg/dL	0.60 – 1.20	Increase	-	0 to 1.20
Random Glucose	mg/dL	70.00 – 140.00	Either	10 (Higher side)	70.0 to 154.0
SGPT	U/L	10.85-71.27	Increase	10	0 to 78.40
SGOT	U/L	14.79-48.68	Increase	10	0 to 53.55
Alkaline Phosphatase	U/L	20.00 – 130.00	Increase	10	0 to 143
Bilirubin – Total	mg/dL	0.19 – 1.32	Increase	-	0 to1.32
Total Protein	g/dL	6.70-8.60	Either	10	6.03 to 9.46
Serum Albumin	g/dL	3.50-5.50	Either	10	3.15 to 6.05
Serum Sodium	mmol/L	136.00 – 145.00	Either	3	131.92 to 149.35
Serum Potassium	mmol/L	3.50-5.20	Either	Lower side 5% & Higher side 2%	3.33 to 5.3
Serum Chloride	mmol/L	96.00 – 106.00	Either	5	91.2 to 111.3
Serum Calcium	mg/dL	8.80 – 10.40	Either	5	8.36 to 10.92
Serum Phosphorous	mg/dL	2.70 - 4.50	Either	10	2.43 to 4.95
Uric Acid (for male)	mg/dL	2.50 – 8.00	Increase	5	0 to 8.4
Uric Acid (for female)	mg/dL	1.30 – 6.00	Increase	5	0 to 6.3

Urinalysis			
Test Name	**Unit**	**Reference Range**	**Pre-defined site normal range**
pH (Reaction)	-	4.8 – 7.4	4.3 – 8.5
Specific Gravity	-	1.001-1.035	1.001 to 1.175
Albumin (Protein)	-	Negative	Negative
Sugar (Glucose)	-	Negative	Negative
Bilirubin	-	Negative	Negative
Urobilinogen	-	Negative	Negative
Ketones	-	Negative	Negative
Amorphous Material	-	Absent	Absent
Bacteria	-	Absent	Absent
Trichomonas	-	Absent	Absent
Monila	-	Absent	Absent
Pus Cells	/h.p.f.	0 – 3	0 to 5
Red Blood Cells	/h.p.f.	0 – 3	0 to 3[#]
Epithelial Cells (for male)	/h.p.f.	0-3	0-8
Epithelial Cells (for female)	/h.p.f.	0-5	0-8
Casts	/l.p.f.	Absent	0 to 2
Crystals	-	Absent	Absent

Hematology					
Test Name	**Unit**	**Reference Range**	**Clinical Relevant Direction**	**Acceptable (%)**	**Pre-defined site normal range**
Haemoglobin (for male)	g/dL	12.0-16.5	Decrease	-	12.5 to 17.5
Haemoglobin (for female)	g/dL	11.0-15.0	Decrease	-	11.5 to 16
Total RBC count (for male)	10^12/L	4.10- 6.30	Either	15 (upper side)	4.10 to 7.2
Total RBC count (for female)	10^12/L	3.25- 5.83	Either	15 (upper side)	3.46 – 6.7
Total WBC count	/µL	4432-11172	Either	5	4210-11730
Platelet Count (PLT)	/µL	150000 – 410000	Decrease	10	135000 to 600000
Haematocrit (PCV/ HCT) (for male)	%	40.0 – 50.0	Decrease	7.5	37 to 60
Haematocrit (PCV/ HCT) (for female)	%	36.0 – 46.0	Decrease	Lower side 6 % and upper side 11	34.0 to 51.06
Neutrophils	%	40.4-73.7	Either	20	32.3 to 88.4
Lymphocytes	%	16.0-45.2	Either	30	11.2 - 58.8
Eosinophils	%	0.6-10.4	Increase	25	0 to 13.0
Monocytes	%	4.5-10.3	Increase	30	0 to 13.4
Basophils	%	0.0-0.8	Increase	50	0 to 1.2

Immunological Tests			
Test Name	**Unit**	**Reference Range**	**Pre-defined site normal range**
Anti-HCV	-	Non reactive	Non reactive
HIV I & II	-	Non reactive	Non reactive
HBsAg	-	Non reactive	Non reactive
Syphilis Test (Rapid Plasma Reagin)	-	Non reactive	Non reactive
ß-Human Chorionic Gonadotropin (ß-HCG)	IU/L	Non Pregnant Female< 5.0	0 to < 5.0

Note: *If any test is outsourced then the results will be evaluated considering the reference range of the outsourced laboratory.*

#for Female volunteers, correlate with menstrual history and up to 0-5 cells/h.p.f. may be considered acceptable.

APPENDIX-III: LIST OF CHANGES

The following are the summary of changes made from version# 02 (March 15, 2022) to version# 03 (October 04, 2022):

1. Title page (CONTRACT RESEARCH ORGANIZATION):
Old text: Cliantha Research Limited 1st floor, Silver Arcade, Near Ashwamegh III, Samrajya, Mujmahuda Road, Akota, Vadodara- 390020, Gujarat, India Tel# +91-265-2324374-76 Fax# +91-265-2324378
New text: **Cliantha Research Limited** **Cliantha Corporate,** **TP 86, FP 28/1,** **Off S.P. Ring Road, Sarkhej,** **Ahmedabad-382210,** **Gujarat, India** **Tel#+91-2717-698500**
Rationale for Change: Based on Sponsor recommendation
2. DECLARATION OF INVESTIGATORS
Old text: Dr. Virendra Solanki, MBBS, Site Principal Investigator Cliantha Research Limited 1st floor, Silver Arcade, Near Ashwamegh, III, Samrajya, Mujmahuda Road, Akota, Vadodara- 390020, Gujarat, India Tel# +91-265-2324374-76 Fax# +91-265-2324378 Email: vsolanki@cliantha.com

New text:

Dr. Mayur Soni, MBBS,
Site Principal Investigator
Cliantha Research Limited
Cliantha Corporate,
TP 86, FP 28/1,
Off S.P. Ring Road, Sarkhej,
Ahmedabad-382210,
Gujarat, India
Tel# +91-2717-698500
Email: msoni@cliantha.com

Rationale for Change: Due to change in clinical site

3. DECLARATION OF SPONSOR

Old text:

Professor Nicholas Day
Sponsor representative
Director,
Mahidol Oxford Tropical Medicine Research Unit,
Faculty of Tropical Medicine, Mahidol University,
3/F, 60th Anniversary Chalermprakiat Building,
420/6 Rajvithi Road,
Bangkok, 10400 Thailand
Tel# +662-203-6333
Email: bob@tropmedres.ac

New text:

Professor Nicholas Day
Sponsor representative
Director,
Mahidol Oxford Tropical Medicine Research Unit,
Faculty of Tropical Medicine, Mahidol University,
3/F, 60th Anniversary Chalermprakiat Building,
420/6 Rajvithi Road,
Bangkok, 10400 Thailand
Tel# +662-203-6333
Email: **Nickd@tropmedres.ac**

Rationale for Change: Typographical error

4. Section #1.1 (CLINICAL FACILITY)

Old text:

Cliantha Research Limited 1st floor, Silver Arcade, Near Ashwamegh III, Samrajya, Mujmahuda Road, Akota, Vadodara- 390020, Gujarat, India Tel# +91-265-2324374-76 Fax# +91-265-2324378
New text: **Cliantha Research Limited** **Cliantha Corporate,** **TP 86, FP 28/1,** **Off S.P. Ring Road, Sarkhej,** **Ahmedabad-382210,** **Gujarat, India** **Tel#+91-2717-698500**
Rationale for Change: Based on Sponsor recommendation
5. Section #1.2 (SCREENING FACILITY)
Old text: Cliantha Research Limited 1st floor, Silver Arcade, Near Ashwamegh III, Samrajya, Mujmahuda Road, Akota, Vadodara- 390020, Gujarat, India Tel# +91-265-2324374-76 Fax# +91-265-2324378
New text: **Cliantha Research Limited** **Cliantha Corporate,** **TP 86, FP 28/1,** **Off S.P. Ring Road, Sarkhej,** **Ahmedabad-382210,** **Gujarat, India** **Tel#+91-2717-698500**

Rationale for Change: Sponsor recommendation
6. Section#1.5 (INDEPENDENT ETHICS COMMITTEE)
Old text: Sanjeevani Independent Ethics Committee, GF-28, 29 & 44, Avishkar Complex, Near GEB Colony, Old Padra Road, Vadodara-390015 Gujarat Tel: +91-9925014449 Email: sanjeevani.iec@gmail.com
New text: **IBIOME - IEC** **B - 501, Krishna Complex,** **Near Rajpath Club,** **S. G. Highway,** **Bodakdev, Ahmedabad – 380 054,** **Gujarat – India** **Tel#: +91-97244 05402** **Email: ibiomeiec@gmail.com**
Rationale for Change: Due to change in Clinical Site
7. Section#1.7 (CLINICAL DATA MANAGEMENT FACILITY)
Old text: Cliantha Research Arista Eight Corporate House, Near Satyam House Behind Rajpath Club, Bodakdev, Ahmedabad - 380054, Gujarat, India Tel: +91 79 6621 9500 Fax: +91 79 6621 9549
New text: **Cliantha Research Limited** **Cliantha Corporate,** **TP 86, FP 28/1,** **Off S.P. Ring Road, Sarkhej,**

Ahmedabad-382210, **Gujarat, India** **Tel# +91-2717-698500**
Rationale for Change: Administrative change
8. Section#2.0 (Handling of blood samples) & Section#12.0 (COLLECTION AND HANDLING OF BLOOD SAMPLES)
Below paragraph has been added: **In case the processing error occurred due to any reason (e.g. mixing of sample while segregation, mechanical failure in centrifuge, etc.) during the sample processing, re-spin the sample (s) under the same conditions, if required.**
Rationale for Change: Based on current in-house practice
9. Section#9.0 (REQUIREMENTS AND RESTRICTIONS FOR THE STUDY)
Old text: In phase I, several extra subjects will be recruited in case there is a short fall of subjects. These subjects will only be dosed to ensure that 50 subjects receive primaquine in this phase. These not dosed will remain in the ward until the end of phase I.
New text: In **period I**, several extra subjects will be recruited in case there is a short fall of subjects. These subjects will only be dosed to ensure that 50 subjects receive primaquine in this phase. These not dosed will **be check-out after dosing** of **period I**.
Rationale for Change: Typographical error
10. Section#2.0 (Pharmacokinetic parameters) & Section#17.2 (PHARMACOKINETIC ANALYSIS)
Old text: Pharmacokinetic parameters Cmax, AUCt, AUC72, AUCi, Tmax, Kel, AUC_%Extrap_obs and tHalf will be calculated using Phoenix® WinNonlin® professional software (Version 8.1 or higher).
New text: **Pharmacokinetic parameters Cmax, AUCt, AUC72, AUCi, Tmax, Kel, AUC_%Extrap_obs and tHalf will be calculated using Phoenix® WinNonlin® professional software (Version 8.1.1 or higher; Pharsight Corporation, USA) or SAS® statistical software (Version: 9.4 or higher; SAS Institute Inc, USA).**
Rationale for Change: Based on current in-house practice

11. Section#14.4 (CLASSIFICATION OF CAUSALITY OF ADVERSE EVENTS):

Old text:

CLIANTHA RESEARCH	
Dr. Sejal Mehta D.G.O, D.N.B Vatsalya Maternity & Nursing Home "EARTH"- The Landmark, 1st floor, Opp. Satsang Party Plot, Nr. Yogi Ashish Society, Sun Pharma Road, Atladara, Vadodara, Mobile# +91 – 9909944341	Dr. Shaival Trivedi, M.D. (Medicine) Consultant physician 2, Santoor Park, Behind Mother School, Makaranad Desai Marg, Gotri Road, Vadodara, 390021 Mobile# +91 – 9825197211 Home Tel# +91–265 – 6556337

New text:

CLIANTHA RESEARCH	
Dr Neeru S. Thakkar M.D. (Gynec) **Brinda Women's Hospital** **B/H. Gordhandas Trust Hospital** **Vastrapur Lake,** **Ahmedabad-380 015** **Cell phone#: +91 9426048614** **Office Tel#: +91-79- 26769092**	**Dr. Praveen Garg, M.D. (Medicine)** **Shashwat Hospital & Research Centre** **D2 Block, Shantiniketan Apartment,** **Opposite H.B. Kapadia School,** **Gurukul Road, Memnagar,** **Ahmedabad-380052** **Mobile# +91 – 9825155017** **Office Tel# +91–79 – 2741 7979**

Rationale for Change: Change in clinical site.

12. Section#22.4 (Volunteer compensation):

Old text:

This compensation is a total of 10,080 rupees (just under 100 pounds sterling).

In case of drop-out or withdrawal of a subject before completion of the study, compensation will be provided as per the compensation policy of Cliantha Research.

Events	Compensation to be paid to subjects
• At the end of Period – I (approximate 25% of total amount)	Rs. 2520
• At the end of Period – II (approximate 65% of total amount)	Rs. 6560
• At the time of end study assessment (remaining amount of total compensation)	Rs. 1000

New text:

This compensation is a total of **15,150** rupees (just under **160** pounds sterling).

In case of drop-out or withdrawal of a subject before completion of the study, compensation will be provided as per the compensation policy of Cliantha Research.

Events	Compensation to be paid to subjects
• At the end of Period – I (approximate 25% of total amount)	Rs. **3800**
• At the end of Period – II (approximate 65% of total amount)	Rs. **9900**
• At the time of end study assessment (remaining amount of total compensation)	Rs. **1450**

Rationale for Change: Due to change in clinical site and policy update

13. Administrative Changes

The Table of Contents has been updated.

The following are the summary of changes made from version# 01 (October 12, 2021) to version# 02 (March 15 2022):

1. Title page (Reference number):
Old text: **Trial registration number:** to follow **MORU Ref:** MAL21010 **OxTREC Ref:** 40-21
New text: **Trial registration number: ISRCTN54640699** **MORU Ref:** MAL21010 **OxTREC Ref:** 40-21
Rationale for Change: Due to administrative reason
2. List of Abbreviation:
Below text has been added: CBC: Complete blood count
Rationale for Change: based on sponsor recommendation
3. Section #2.0 (Total Blood Loss)
Old text: The volume of blood required from each subject for the study will be 346 mL.
New text: The volume of blood required from each subject for the study will be **348 mL.**
Rationale for Change: due to change in the blood loss
4. Section#2.0 (Safety Assessment & sequence of event) & 13.2
Below Text has been added: A complete blood count (CBC) will be measured prior to check in of period II. Only subjects within normal limits or clinically insignificant falls will be dosed in period II.
Rationale for Change: based on sponsor recommendation
5. Section#2.0 (Assessment of bioequivalence) & 17.3

Old text: Data of active metabolite of Carboxyprimaquine will be provided as supportive data.
New text: Data of **inactive** metabolite of Carboxyprimaquine will be provided as supportive data.
Rationale for Change: Due to typographical error
6. Section#3.5
Old text: To accommodate these challenges and mitigate safety risks associated with COVID-19, changes may be required from approved protocol which include (but not limited to) conducting **the study in multiple groups**, change in study procedures timing, change in subject's housing duration; additional test or parameter may be performed to standard inclusion or exclusion criteria, etc. The changes made to the procedure will prioritize subject's safety and data validity and integrity. For any significant change, a planned protocol deviation will be filled and notified to ethics committee and/or local regulatory (as applicable).
New text: To accommodate these challenges and mitigate safety risks associated with COVID-19, changes may be required from approved protocol which include (but not limited to) change in study procedures timing, change in subject's housing duration; additional test or parameter may be performed to standard inclusion or exclusion criteria, etc. The changes made to the procedure will prioritize subject's safety and data validity and integrity. For any significant change, a planned protocol deviation will be filled and notified to ethics committee and/or local regulatory (as applicable).
Rationale for Change: based on sponsor recommendation
7. Section#3.5
Below mentioned text has been removed: If study is conducted in multiple groups, then bio-analysis will be initiated only after completion of clinical phase of all groups.
Rationale for Change: based on sponsor recommendation
8. Section#9.0 (housing requirements):
Below paragraph has been added:

In phase I, several extra subjects will be recruited in case there is a short fall of subjects. These subjects will only be dosed to ensure that 50 subjects receive primaquine in this phase. These not dosed will remain in the ward until the end of phase I.

Rationale for Change: based on sponsor recommendation

9. Section#12.0:

Old text:

Description	Blood loss
Blood withdrawn for screening	5.5 mL
Blood loss of pharmacokinetic samples for all periods	312 mL
Discarded blood containing normal saline	23 mL
Blood withdrawn for post study laboratory assessment	5.5 mL
Total blood loss for the study*	**346 mL**

*Note:

Based on investigator discretion, additional blood sample (up to 10 mL) may be collected for additional tests/repeats, sample clots, sample spillage or any other justifiable reason, if deemed necessary.

If alcohol test is performed from blood, additional 2 mL blood sample will be collected in each period in addition to blood loss listed above.

New text:

Description	Blood loss
Blood withdrawn for screening	5.5 mL
Blood loss of pharmacokinetic samples for all periods	312 mL
Discarded blood containing normal saline	23 mL
Blood drawn for CBC on the day of check-in of Period II	**02 mL**
Blood withdrawn for post study laboratory assessment	5.5 mL
Total blood loss for the study*	**348 mL**

*Note:

Based on investigator discretion, additional blood sample (up to 10 mL) may be collected for additional tests/repeats, sample clots, sample spillage or any other justifiable reason, if deemed necessary.

If alcohol test is performed from blood, additional 2 mL blood sample will be collected in each period in addition to blood loss listed above.

Rationale for Change: Due to additional of extra laboratory test at period-II check-in

10. Section#17.2:

Below mentioned criteria has been removed

All missing concentration data will be disregarded from pharmacokinetic analysis.
Rationale for Change: based on sponsor recommendation.
11. Section#17.3 (Analysis of variance):
Below paragraph has been removed: If the study will be conducted in groups then below statistical model will be used: group, sequence, group*sequence, period(group), treatment, and subject(sequence*group) will be used as a fixed effects. Treatment and period(group)will be tested using Mean Square Error and Sequence will be tested using subject(sequence*group) as the error term at 5% level of significance
Rationale for Change: based on sponsor recommendation
12. Section#17.3 (Variability and power):
Old text: Inter-subject variability and power (using Two one-sided test method) will be computed and reported for ln-transformed Pharmacokinetic parameters for Cmax, AUC72 and AUCt.
New text: **Intra**-subject variability and power (using Two one-sided test method) will be computed and reported for ln-transformed Pharmacokinetic parameters for Cmax, AUC72 and AUCt.
Rationale for Change: based on sponsor recommendation
13. Section#22.4 (Volunteer compensation):
Old text: Subjects will not be paid for any loss of wages for the days of their participation; however adequate compensation will be given to subjects in appropriate proportion at the end of each of the periods of the study as per compensation policy of Cliantha Research as approved by the IEC. This compensation is a total of 10,040 rupees (just under 100 pounds sterling). In case of drop-out or withdrawal of a subject before completion of the study, appropriate compensation will be provided as per compensation policy of Cliantha Research as approved by the IEC.
New text: Subjects will not be paid for any loss of wages for the days of their participation; however adequate compensation will be given to subjects in appropriate proportion at the end of each of the periods of the study as per compensation policy of Cliantha Research as approved by the IEC.

This compensation is a total of **10,080** rupees (just under 100 pounds sterling).

In case of drop-out or withdrawal of a subject before completion of the study, compensation will be provided as per the compensation policy of Cliantha Research.

Events	Compensation to be paid to subjects
• **At the end of Period – I (approximate 25% of total amount)**	**Rs. 2520**
• **At the end of Period – II (approximate 65% of total amount)**	**Rs. 6560**
• **At the time of end study assessment (remaining amount of total compensation)**	**Rs. 1000**

Subjects will receive full compensation if they are withdrawn from the study by the investigator on medical grounds to safeguard your safety e.g. a primaquine related AE, or are withdrawn on compassionate grounds.

The extra subjects not dosed during phase I will receive the compensation during check-out phase I (as per compensation policy).

All other subjects will receive compensation for the days that they participated in the study. This will be reduced to 50% for those subjects who do not follow the protocol.

Rationale for Change: Due to change in the blood loss

14. Administrative Changes

The Table of Contents has been updated.