An open label phase 1b study to characterise the pharmacokinetic/pharmacodynamic relationship and safety of MMV367 in healthy adult participants experimentally infected with blood stage *Plasmodium falciparum*

Protocol Number: MMV_MMV367_22_01

Principal Investigator: Associate Professor Bridget Barber

Sponsor: Medicines for Malaria Venture

Local Sponsor: Southern Star Research Pty Ltd

Funded by: Medicines for Malaria Venture and GSK plc

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14 April 2023

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STATEMENT OF COMPLIANCE

Investigator declaration

I have read the protocol and agree that it contains all necessary details for carrying out the study as described. I will conduct this protocol as outlined herein and will make a reasonable effort to complete the study within the time designated.

I agree to personally conduct or supervise the described study.

The study will be conducted in accordance with the following:

- World Medical Association Declaration of Helsinki – Ethical Principles for Medical Research Involving Human Participants (Fortaleza, Brazil 2013)
- NHMRC National Statement on Ethical Conduct in Human Research (2007, updated 2018)
- Integrated addendum to ICH E6(R1): Guideline for Good Clinical Practice ICH E6(R2) (November 2016) – with introductory comments of the Australian Therapeutic Goods Administration
- Current ethics approved Clinical Trial Protocol.

I agree to inform all participants that the study agents are being used for investigational purposes and I will ensure that the requirements related to obtaining informed consent are in accordance with International Council of Harmonisation (ICH) Guidelines for Good Clinical Practice (GCP) section 4.8 and local requirements.

I agree to report adverse events that occur in the course of the study to the Sponsor in accordance with ICH Guidelines for GCP section 4.11 and local requirements.

I have read and understand the information in the Investigator's Brochure(s), including the potential risks and side effects of the study drug.

I agree to promptly report to the Human Research Ethics Committee (HREC) all changes in the research activity and all unanticipated problems involving risk to participants. I will not make any changes to the conduct of the study without HREC and Sponsor approval, except when necessary to eliminate apparent immediate harm to participants.

I agree to maintain adequate and accurate records and make those records available in accordance with ICH Guidelines for GCP section 4.11 and local requirements.

I agree to ensure that all associates, colleagues, and employees assisting in the conduct of the study are informed about their obligations in meeting the above commitments.

I understand that the study may be terminated or enrolment suspended at any time by the Sponsor, with or without cause, or by me if it becomes necessary to protect the best interest of the participants.

_____ Date: _____

Principal Investigator

Signatories

The undersigned parties agree, that the protocol was written in accordance with the World Medical Association Declaration of Helsinki – Ethical Principles for Medical Research Involving Human Participants (Fortaleza, Brazil 2013), the NHMRC National Statement on Ethical Conduct in Human Research (2007, updated 2018), and the Integrated Addendum to ICH E6 (R1): Guideline for Good Clinical Practice E6 (R2) (November 2016) – with introductory comments of the Australian Therapeutic Goods Administration.

This clinical trial protocol has been reviewed and approved by the Sponsor.

Name	Signature	Date
Medical Director: **Stephan Chalon, MD** Medicines for Malaria Venture		
Clinical Scientist: **Etienne Guirou, PhD** Medicines for Malaria Venture		

1 PROTOCOL SUMMARY

1.1 SYNOPSIS

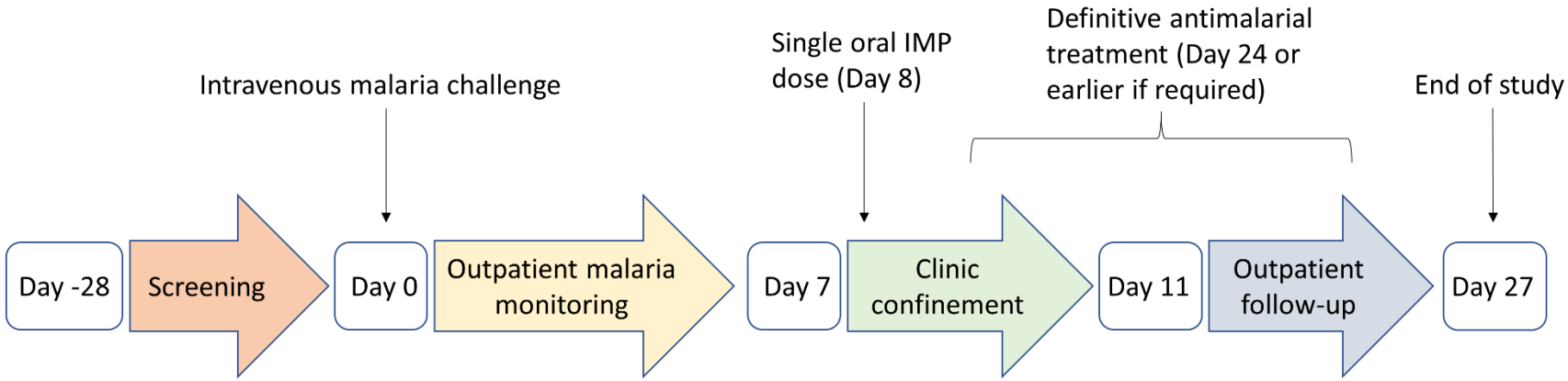
Title:	An open label phase 1b study to characterise the pharmacokinetic/pharmacodynamic relationship and safety of MMV367 in healthy adult participants experimentally infected with blood stage *Plasmodium falciparum.*
Study Description:	This is an open-label, adaptive study that will utilise the *P. falciparum* induced blood stage malaria (IBSM) model to characterise the pharmacokinetic/pharmacodynamic (PK/PD) profile and safety of MMV367 (the IMP). Up to 18 participants will be enrolled in cohorts of up to 6 participants each. The study will proceed as follows for all participants: • Screening period of up to 28 days to recruit healthy adult participants. • Day 0: Intravenous inoculation with approximately 2,800 viable *P. falciparum*-infected erythrocytes. • Days 1-3: Daily follow up via phone call or text message. • Days 4-7: Daily site visits for clinical evaluation and blood sampling to monitor malaria parasitaemia via quantitative polymerase chain reaction (qPCR). • Day 7 PM: Start of confinement within the clinical trial unit. • Day 8: Administration of a single oral dose of the IMP (MMV367). Different doses of MMV367 will be administered across and within cohorts in order to effectively characterise the PK/PD relationship (see Section 4.3 for dose selection rationale). • Days 8-11: Regular clinical evaluation and blood sampling while confined to monitor malaria parasitaemia and measure MMV367 plasma concentration. • Day 11 AM: End of confinement within clinical trial unit. • Days 12-23: Outpatient follow-up for clinical evaluation and blood sampling. • Day 24: Initiation of compulsory definitive antimalarial treatment with Riamet® (artemether/lumefantrine) and/or other registered antimalarials if required (see section 6.1). Treatment will be initiated earlier than Day 24 in the event of: ○ Insufficient parasite clearance following IMP dosing (parasitaemia not reduced ≥10-fold by Day 10 compared with peak parasitaemia on Day 8). ○ Parasite regrowth following IMP dosing (initial parasite clearance is followed by asexual parasite regrowth above 5000 parasites/mL).

	○ Participant discontinuation/withdrawal (see Section 7), ○ Investigator's discretion in the interest of participant safety. • Day 27: End of study visit for final clinical evaluation and to ensure complete clearance of malaria parasitaemia (at least one negative qPCR result required).
Objectives:	<u>Primary Objective</u> To characterise the PK/PD relationship of MMV367 in healthy participants experimentally infected with blood stage *P. falciparum.*
	<u>Secondary Objectives</u> • To evaluate the safety and tolerability of single oral doses of MMV367 in healthy participants experimentally infected with blood stage *P. falciparum*. • To evaluate the safety and tolerability associated with blood-stage malaria infection in healthy malaria naïve participants. • To characterise the pharmacokinetics of MMV367 following single oral dose administration in healthy participants experimentally infected with blood stage *P. falciparum*. • To characterise the parasite clearance kinetics following single doses of MMV367 in healthy participants experimentally infected with blood stage *P. falciparum*.
	<u>Exploratory objectives</u> • To investigate potential resistance of *P. falciparum* to MMV367 in participants experiencing parasite regrowth after MMV367 administration. • To characterise the effect of MMV367 on *P. falciparum* parasite viability.
Endpoints:	<u>Primary Endpoint</u> The PK/PD relationship between MMV367 blood concentrations and blood stage asexual parasitaemia. <u>Secondary Endpoints</u> • The incidence, severity and relationship to MMV367 of adverse events as determined by self-reported symptoms, clinical laboratory analysis, vital signs, physical examinations and ECG assessments. • The incidence, severity and relationship to the malaria challenge agent of adverse events as determined by self-reported symptoms, clinical laboratory analysis, vital signs, physical examinations and ECG assessments. Additionally, the severity of the induced malaria infection in each participant graded by the malaria clinical score.

	• The pharmacokinetics of MMV367 calculated using non-compartmental methods (C_{max}, t_{max}, AUC_{last}, AUC_{inf}, CL/F, Vz/F, λ, $t_{1/2}$). • The parasite clearance kinetics following dosing with MMV367 (parasite reduction ratio over a 48-hour period [PRR_{48}] and corresponding parasite clearance half-life [$PCt_{1/2}$]). Exploratory endpoints • Incidence and level of resistance to MMV367 in parasites determined using *in vitro* methods. • Effect of MMV367 on parasite viability examined by *ex vivo* cultures.
Study Population:	Malaria-naïve healthy adults, aged between 18-55 years old, who meet all the inclusion criteria and none of the exclusion criteria. Participants will be recruited from the South East Queensland region in Australia. Up to 18 participants are planned to be enrolled in cohorts of up to 6 participants.
Phase:	Phase 1b
Description of Clinical Units/Facilities Enrolling Participants:	The trial is planned to be performed at the University of Sunshine Coast in two Clinical Trial Units in Queensland, Australia. Moreton Bay Health Hub Morayfield 19-31 Dickson Road Morayfield QLD 4506 South Bank Building A2, SW1 Complex 52 Merivale Street South Brisbane QLD 4101
Description of Study Interventions:	Investigational Medicinal Product MMV367 dispersible granules 250 mg/g (25% w/w) was manufactured by Piramal Pharma Solutions (India) and will be weighed and resuspended in sterile water at the clinical trial unit pharmacy according to the Pharmacy Manual. Each participant will be administered the appropriate dose orally after a ≥ 8 hour fast at the clinical trial unit under direct staff observation. Malaria Challenge Agent The *P. falciparum* 3D7 master cell bank (MCB) was produced from blood collected from a donor with clinical manifestation of malaria. Each challenge dose will be prepared aseptically at Q-Gen Cell Therapeutics from an aliquot of the *P. falciparum* 3D7 MCB. Each participant will be inoculated intravenously with a dose of approximately 2,800 viable *P. falciparum* 3D7-infected erythrocytes in 2 mL of saline for injection. Definitive antimalarial Medications • Riamet® (each tablet containing 20 mg artemether and 120 mg lumefantrine) is marketed by Novartis Pharmaceuticals Australia Pty Ltd and will be the first-line definitive antimalarial medication in this study. Riamet® tablets will be administered as six oral doses of four tablets each (total course of 24 tablets equivalent to 480 mg

	artemether and 2.88 g lumefantrine). The second dose of four tablets will be administered 8±1 h after the first dose and the remaining doses will be administered twice daily (morning and evening). Each dose will be administered with food or drinks rich in fat. • Primacin® (each tablet containing 13.2 mg primaquine phosphate equivalent to 7.5 mg of primaquine) is marketed by Boucher & Muir Pty Ltd and is the recommended treatment to clear gametocytes. If required, participants will be administered a single dose of 6 tablets (45 mg primaquine) orally. • Malarone® (each tablet containing 250 mg atovaquone and 100 mg proguanil hydrochloride) is marketed by GlaxoSmithKline Australia Pty Ltd. If required, Malarone® will be administered as three oral doses of 4 tablets (one dose daily for 3 days; total course of 12 tablets). • Intravenous artesunate is the recommended parenteral treatment for severe malaria in Australia. If required, the recommended dose regimen will be followed (2.4 mg/kg at approximately 0, 12, 24, 48 hours and then daily for up to 7 days or until able to take oral drugs). ***Note:*** *Primacin, Malarone, and intravenous artesunate will only be administered if required (see section 6.1).*
Study Duration:	Approximately 12 months from enrolment of the first participant until completion of the data analyses.
Participant Duration:	Approximately 56 days for each participant which includes a screening period (up to 28 days), a period of observation following malaria challenge (8 days) and a follow-up after administration of the IMP (20 days).

1.2 SCHEMA


Intravenous malaria challenge
Single oral IMP dose (Day 8)
Definitive antimalarial treatment (Day 24 or earlier if required)
End of study
Day -28
Screening
Day 0
Outpatient malaria monitoring
Day 7
Clinic confinement
Day 11
Outpatient follow-up
Day 27

1.3 SCHEDULE OF ACTIVITIES (SOA)

The full study conduct schedule is presented in Appendix 12.1 and summarised in the tables below.

Screening and malaria challenge period

Study Day	**-28 to -1**	**-3 to -1**	**0**		**1**	**2**	**3**	**4**	**5**	**6**	**7 AM**
	Screening	**Eligibility confirmation**	**Pre-inoculation**	**Post-inoculation**	**Phone contact**			**Outpatient visits**			
Informed consent	X										
Beck Depression Inventory	X										
Cardiovascular risk	X										
Demography	X										
Medical history	X		X								
Prior medications	X		X								
Eligibility	X	X	X								
Urine drug screen	X		X								
Alcohol breath test	X		X								
Viral screen blood sample	X										
RBC alloantibody blood sample	X										
Coagulation profile blood sample	X										
COVID-19 rapid test			X								
COVID-19 PCR test		X								X	
G6PD test blood sample	X										
Serum pregnancy test blood sample	X[c]	X[d]									
Urine pregnancy test (WOCBP)			X								
FSH test (post-menopausal women)	X										
Urinalysis sample	X	X									
Adverse events				X	X	X	X	X	X	X	X
Concomitant medications				X	X	X	X	X	X	X	X
Safety serum retention			X								
Full physical exam	X[a]										
Abbreviated physical exam			X								
Symptom directed physical exam								X	X	X	X
Malaria clinical score			X					X	X	X	X

Study Day	**-28 to -1**	**-3 to -1**	**0**		**1**	**2**	**3**	**4**	**5**	**6**	**7 AM**
Vital signs	X[e]		X	X				X	X	X	X
Electrocardiograph (triplicate)	X		X								
Haematology blood sample	X	X									X
Biochemistry blood sample	X[b]	X									X
Diary card				X	X	X	X	X	X	X	X
Phone contact					X	X	X				
Malaria challenge agent administration				X							
Malaria PCR blood sample			X					X	X	X	X
Future malaria research blood sample (if participant consented)			X								

FSH: Follicle-stimulating hormone; G6PD: Glucose-6-phosphate dehydrogenase; PCR: Polymerase chain reaction; RBC: Red blood cell; WOCBP: Women of childbearing potential.

[a]Body height to be recorded at screening only.

[b]Lipids included at Screening for cardiovascular risk factor.

[c]Serum pregnancy test to be performed for all women at Screening.

[d]Serum pregnancy test to be performed for WOCBP at the eligibility confirmation visit.

[e]At screening, vital signs will be measured after the volunteer has rested in the supine position for 5 minutes. SBP and DBP will be measured again after the volunteer has transitioned to a standing position for 3 minutes to assess postural hypotension (see Section 5.2, exclusion criterion 10). At all other time-points vital signs will be measured after the participant has rested in the seated position for at least 5 min.

IMP administration and clinic confinement period

Study Day	**7 PM**	**8**										**9**			**10**			**11**
Hours post-IMP dose		**Pre-dose**	**0**	**1**	**2**	**3**	**4**	**6**	**8**	**12**	**16**	**24**	**30**	**36**	**48**	**54**	**60**	**72**
Urine drug screen	X																	
Alcohol breath test	X																	
COVID-19 rapid test	X																	
Urine pregnancy test (WOCBP)	X																	
Adverse events		X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Concomitant medications		X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Abbreviated physical exam		X																
Symptom-directed physical exam				X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Malaria clinical score		X						X		X		X	X	X	X	X	X	X
Vital signs		X						X		X		X	X	X	X	X	X	X
Electrocardiograph		X										X			X			X
Haematology blood sample		X										X						X
Biochemistry blood sample		X										X						X
Malaria PCR blood sample		X			X		X		X	X	X	X	X	X	X	X	X	X
Randomisation		X																
IMP administration			X															
IMP PK blood sample		X		X	X	X	X	X	X	X	X	X		X	X		X	X
Parasite *ex vivo* viability blood sample		X							X		X	X		X	X			X
Future malaria research blood sample (if participant consented)_		X																

IMP: Investigational medicinal product; PK: Pharmacokinetics; PCR: Polymerase chain reaction; WOCBP: Women of childbearing potential.

Outpatient follow-up period

Study Day	**12**	**13**	**15**	**17[d]**	**20[d]**	**22**	**24[d]**	**27 EOS[e]**
Hours (days) post-IMP dose	**96±4 (4)**	**120±12 (5)**	**168±24 (7)**	**216±24 (9)**	**288±24 (12)**	**336±24 (14)**	**384±24 (16)**	**456±24 (19)**
Viral screen blood sample								X
RBC alloantibody blood sample								X
Serum pregnancy test blood sample (WOCBP)								X
Urinalysis sample								X
Adverse events	X	X	X	X	X	X	X	X
Concomitant medications	X	X	X	X	X	X	X	X
Safety serum retention blood sample								X
Full physical exam								X
Symptom directed physical exam	X	X	X	X	X	X	X	
Vital signs	X	X	X	X	X	X	X	X
Malaria clinical score	X	X	X	X	X	X	X	X
Electrocardiograph								X
Haematology blood sample			X			X		X
Biochemistry blood sample			X			X		X
Diary card	X	X	X	X	X	X	X	X
Malaria PCR blood sample	X	X	X	X	X	X	X	X
IMP PK blood sample	X	X	X	X	X			
Definitive antimalarial treatment							X[b]	
Parasite drug resistance blood sample	X[a]							
Parasite *ex vivo* growth blood sample	X	X[c]						
Future malaria research blood sample (if participant consented)	X		X					X

EOS: End of study; IMP: Investigational medicinal product; PK: Pharmacokinetics; PCR: Polymerase chain reaction; RBC: Red blood cell; WOCBP: Women of childbearing potential.

[a]One blood sample will be collected for parasite drug resistance assessment only in the event of parasite regrowth. This sample will be collected just prior to initiation of definitive antimalarial treatment.

[b]Definitive antimalarial treatment to be initiated on Day 24, or earlier in accordance with the criteria specified in Section 4.1.

[c]One blood sample will be collected for parasite *ex vivo* growth only in the event of parasite regrowth. This sample will be collected just prior to initiation of definitive antimalarial treatment.

[d]Visit not required if participant has previously been administered definitive antimalarial treatment and returned at least one negative malaria PCR result.

[e]Participants who withdraw, or are withdrawn or discontinued from the trial, will be asked to attend an early termination visit with assessments equivalent to the Day 27 EOS visit.

2 INTRODUCTION

2.1 BACKGROUND AND STUDY RATIONALE

Malaria is a mosquito-borne parasitic disease prevalent in tropical and subtropical regions around the world. It is caused by infection with the *Plasmodium* parasite of which five main species are known to infect humans: *P. falciparum*, *P. vivax*, *P. ovale*, *P. malariae* and *P. knowlesi*. Of these species, *P. falciparum* is responsible for the majority of the malaria burden, particularly in sub-Saharan Africa. The malaria parasite has a complex life cycle in humans consisting of both a liver stage and a blood stage. Clinical symptoms of malaria infection are due to the asexual blood stage of the parasite's life cycle.

Malaria is responsible for significant global morbidity and mortality, with an estimated 247 million new cases and 619,000 deaths worldwide in 2021 [1]. The emergence of antimalarial drug resistance has been a major setback for malaria control and progress towards the goal of elimination. Resistance has emerged to all antimalarials in widespread use, including artemisinin-based combination therapies, the current first line treatment for uncomplicated *P. falciparum* malaria. Thus, the development of novel antimalarial therapies is required to control malaria in regions with high prevalence of artemisinin-resistance.

MMV367 is a first in class, fast acting, orally bioavailable blood stage inhibitor of *P. falciparum*. MMV367 belongs to the pyrrolidinamide class of antimalarials, identified using a high throughput whole cell phenotypic screen. Mode of action studies have identified changes in genes encoding acyl coenzyme A synthetase 10 and 11 (ACS10 and ACS11) to modulate the potency of pyrrolidinamides. MMV367 is being developed primarily for the curative treatment of uncomplicated malaria due to *P. falciparum* in adults and children, as a single dose regimen in a potential fixed-dose combination with another active and safe non-artemisinin antimalarial drug.

The following properties of MMV367 characterised in preclinical studies (see MMV367 Investigator's Brochure) justified the progression to clinical development:

- Potent *in vitro* activity against asexual *P. falciparum* blood forms in a panel of drug sensitive and drug resistant strains and clinical isolates.
- Low *in vitro* frequency of spontaneous resistance.
- Fast antimalarial mode of action with killing rates comparable to artemisinins.
- Oral *in vivo* efficacy against *P. falciparum* with potency comparable or superior to marketed antimalarials.
- Novel antimalarial mechanism of action.
- Suitable physicochemical properties.
- Projected low therapeutic dose in humans.
- Low oral toxicity in two preclinical species (i.e., preclinical safety profile suitable for progression to first-in-human study).

A first in human study to investigate the safety, tolerability, and pharmacokinetics of MMV367 in 47 healthy males and women of childbearing potential has recently been completed in the UK. Preliminary (blinded) safety/tolerability and pharmacokinetic data from this randomised, double-blind, placebo-controlled, single-ascending dose (SAD) study, pilot food effect study, and multiple-dose study are available. MMV367 doses evaluated in this phase 1 study (up to 1500 mg single dose in the SAD, 440 mg single dose in the pilot food effect study, and 400 mg daily for 3 days in the multiple dose study) were not associated with severe or serious adverse events. Thus, based on the results of this completed first-in-human study, the progression of MMV367 as a potential new treatment for acute uncomplicated malaria is supported.

Volunteer infection studies (VIS) using the induced blood stage malaria (IBSM) model have previously successfully characterised the PK/PD relationship of several experimental antimalarial compounds as well as a number of antimalarial drugs currently in use [2-5,11]. The IBSM model involves intravenous inoculation of healthy adult participants with parasite-infected erythrocytes and administration of the test antimalarial at a defined time after inoculation. Frequent blood sampling is performed to measure parasitaemia and drug concentration kinetics over the course of the study, thus providing data for PK/PD modelling analyses. Such VIS have been shown to accurately predict the activity of investigational antimalarials in studies in endemic populations [6, 7].

The current study aims to determine the PK/PD relationship of single doses of MMV367 following administration in healthy adult participants using the *P. falciparum* IBSM model along with further evaluation of the safety and tolerability of MMV367 in the context of this human challenge model. Data obtained in this study will support the use of MMV367 in new antimalarial combination therapies against acute uncomplicated malaria by informing partner drug selection and dosing considerations for future trials in malaria endemic populations.

2.2 RISK/BENEFIT ASSESSMENT

2.2.1 KNOWN POTENTIAL RISKS

Risks associated with the Investigational Medicinal Product (MMV367)

Risks associated with the IMP are informed by extensive preclinical studies and the first in human study recently completed. Full details on preclinical studies are included in the IB and a summary of the preliminary first in human study data (blinded) is included as an addendum to the IB.

Review of preclinical safety data did not identify any important risks associated with MMV367. Based on preclinical observations at toxicological doses in rats and dogs, potential risks associated with MMV367 administration were all considered monitorable in humans prior to initiation of the first-in-human study (gastrointestinal, blood pressure, heart rate, and respiratory rate).

The exposure at the no observed adverse effect level (NOAEL) of the most sensitive preclinical species was identified from the 7-day GLP toxicology study in dogs and considered as the highest acceptable human exposure. The corresponding exposure of 1250 µg.h/mL is the cumulative AUC (0-168 h) of the parent drug MMV367. The average cumulative AUC (0-168 h) was calculated as follows: (AUC on Day 1 + AUC on Day 7) / 2*7. The highest exposure achieved in the first in human study was an AUC_{inf} of 299 µg.h/mL in one volunteer who received a 1500 mg dose, which is 4-fold below the exposure at NOAEL in the most sensitive preclinical species.

Preliminary data from the first in human study indicate that MMV367 is well tolerated in humans. A summary of all treatment-emergent adverse events (TEAEs) reported in the study is included in Table 1 below. No clinically concerning changes in ECGs (QTc interval in particular) or clinical laboratory parameters were identified.

Table 1 Summary of preliminary blinded adverse event data from the first in human study of MMV367

AE verbatim[#]	**100 mg [N=8]**	**300 mg [N=8]**	**750 mg [N=7]**	**1500 mg [N=8]**	**440 mg (fed/fasted) [N=8]**	**400 mg (3 days) [N=8]**
	Number of events (number of participants)					
Paresthesia	1 (1)					
Orthostatic hypotension	1 (1)					1 (1)
Upper respiratory tract infection symptoms		1 (1)				
Period pain		1 (1)				
Dizziness		1 (1)				1 (1)
Presyncope		1 (1)				2 (2)
Lightheadedness			1 (1)			1 (1)
Headache			1 (1)		3 (2)	2 (2)
Conjunctivitis			1 (1)			
Abdominal pain			**1 (1)***			
Epigastric pain			**1 (1)***			
Loose stools			1 (1)			
Increased flatulence				1 (1)		
Dental extraction				1 (1)		
Broken tooth				1 (1)		
Dental pain				1 (1)		
Nasopharyngitis				1 (1)		
Cold/flu symptoms					1 (1)	2 (2)
Rhinitis						1 (1)
Low back pain						2 (2)
Tingly teeth						1 (1)
Cough						1 (1)
TOTAL TEAEs	**2**	**4**	**6**	**5**	**4**	**14**

[#]All adverse events were mild or moderate in severity.
*Adverse events considered related to the IMP.

Risks associated with the *P. falciparum* IBSM model

Risks pertaining to the IBSM model include the development of blood borne infections, reaction to the blood sample, and clinical manifestations of malaria (including haematological adverse events such as neutropenia or leukopenia, liver function abnormalities, and occurrence of cardiac AEs). These are each discussed in detail below.

- **Risk management of blood borne infections**

The *P. falciparum* 3D7 challenge agent will be used to induce blood stage malaria in this trial. Although the challenge agent contains a very small amount of blood, risk of a transfusion-transmissible infection in this trial is extremely low. Firstly, donors were screened and tested negative for the presence of active blood borne infections. Secondly, the Australian Red Cross Lifeblood removed white blood cells from the donor blood to lower the risk of a transfusion-transmissible infection. Thirdly, the volume of blood associated with the malaria challenge inoculum is many thousands of times smaller than in a transfused unit (i.e., a relatively lower risk of infection). As part of the safety monitoring, all participants will be screened for blood borne infections before and after the trial. To date, no blood borne infections have been reported in any of the 453 participants who have received this *P. falciparum* 3D7 challenge agent.

- **Risk management of reaction to the blood sample**

The risk of developing red blood cell (RBC) alloantibodies and/or experiencing an acute haemolytic reaction in this trial is considered extremely low because the donor blood used to produce the challenge agent was blood group O (RhD) negative. People with this blood group are generally considered "universal donors". However, it is possible that participants could suffer a transfusion reaction after they receive the challenge agent or could develop alloantibodies to the donor RBCs that may make blood transfusion more difficult in the future. To date, of the 453 participants who have been administered this *P. falciparum* 3D7 challenge agent, one participant has developed an antibody response to a minor Rh antigen (anti-E antibody). However, there was no laboratory evidence to indicate that the specific Rh phenotype of the donor RBCs in the challenge agent stimulated production of this anti-E alloantibody.

Alloimmunisation has been observed in an IBSM study testing a genetically modified *P. falciparum* 3D7 strain. This study investigated the safety, infectivity and immunogenicity of a strain in which the gene encoding the knob associated histidine rich protein had been deleted in order to explore its potential use as a live-attenuated malaria vaccine. In this study, two participants developed alloimmunisation (anti-C and anti-P1 antibodies in one participant and anti-c antibodies in the other participant) after administration of the highest dose of parasites (approximately 1000-fold higher dose of parasites than routinely administered in IBSM studies using the wild-type *P. falciparum* 3D7 strain). However, the total dose of erythrocytes (including non-parasitized) administered was similar to that routinely administered in IBSM studies (approximately 1×10^8 erythrocytes). Although anti-P1 antibodies are not considered clinically important for transfusion reactions, anti-C and anti-c antibodies may result in a delayed transfusion reaction characterized by slow drop in haemoglobin over two weeks post-transfusion. Consultation with transfusion medicine specialists at the completion of the study indicated there was no immediate risk if these participants were to require emergency administration of unmatched Group O Rh

(D) negative blood, and in the setting of routine blood transfusion a full cross-match would obviate such a reaction. Additionally, since both antibodies were of low titre, there was a possibility that they may diminish over time. None of the other 6 participants enrolled in this study were found to have developed alloimmunisation; this includes 4 participants administered a lower dose of parasites and 2 participants administered the same dose of parasites as the 2 participants who developed alloimmunisation.

Participants will be monitored for signs and symptoms in the period immediately after administration of the challenge agent to further assess the risk of the challenge agent causing a transfusion reaction. All participants will be tested for RBC alloantibodies at screening and at the end of the trial as part of safety monitoring.

Women of childbearing potential (WOCBP) have a small additional risk if they develop RBC alloantibodies, as this could cause problems during pregnancy. WOCBP who have participated in several IBSM trials with *P. falciparum* isolate 3D7 have had no known issues to date. Including WOCBP in the trial enhances the generalisability of the trial results.

- **Risk management of clinical manifestations of malaria**

The number of viable blood stage parasites that will be used to infect the participants in this trial (~2,800) is substantially lower than the starting parasitaemia induced from the bite of a single malaria-infected mosquito (~30,000 parasites are released into the blood when they break out of a single infected liver cell). In this trial, parasite growth and malaria symptoms will be closely monitored in participants following administration of the challenge agent. The timing of IMP dosing (Day 8) has been selected based on extensive experience conducting IBSM trials using the *P. falciparum* 3D7 challenge agent. Clinical symptoms of malaria are likely to be absent or predominantly mild in severity up to Day 8, while parasitaemia will have reached a sufficient magnitude (>5000 parasites/mL) to allow assessment of the antimalarial activity of the IMP.

- **Risk management of liver function abnormalities**

Transient, asymptomatic liver function test (LFT) abnormalities, including rare cases of alanine aminotransferase (ALT) and/or aspartate aminotransferase (AST) elevations >10 fold the upper limit of normal (×ULN), have been reported in several participants in IBSM studies [5,12]. However, changes in bilirubin have only been reported in one participant with undiagnosed pre-existing liver disease [12]. The LFT abnormalities did not require treatment, and resolved by the end of the studies. A few cases of the LFT elevations were considered serious AEs (SAEs) by two Pharma sponsors due to internal processes for SAE notifications. An independent review involving drug-induced liver injury experts found these liver function abnormalities were most likely a direct consequence of the malaria infection rather than a direct drug-induced liver injury caused by the investigational antimalarial drug [13]. As a precaution, participants with LFT results exceeding the acceptable values at screening (Appendix 12.2) will be ineligible to participate in this study. All enrolled participants will undergo regular safety monitoring to assess for asymptomatic liver function abnormalities. Participants will be required to minimise intake of possibly hepatotoxic substances, such as alcohol and paracetamol, during the trial. Drugs of abuse are not permitted under any circumstance.

- **Risk management of cardiac adverse events**

There have been four cardiac SAEs reported in healthy participants in the Netherlands participating in malaria challenge studies using sporozoites (i.e., direct feeds by infected mosquitoes rather than IBSM infection). These four cardiac SAEs are described in the *P. falciparum* 3D7 challenge agent IB; two of the events have also been published as case reports [8, 9]. No cardiac SAEs have been attributed to the challenge agents in IBSM studies. However, in one IBSM trial, two participants (one infected with the *P. falciparum* 3D7 challenge agent and one infected with another malaria challenge agent [*P. falciparum* K13]) developed ventricular extra-systoles that were classified as moderate AEs possibly related to malaria. As a precaution, participants at significant risk of cardiovascular disease are excluded from participating in IBSM studies, and regular safety monitoring, including physical examination and ECG assessments, will be performed.

Risks associated with definitive antimalarial medications

Risks related to use of artemether-lumefantrine (Riamet®), primaquine phosphate (Primacin®) and atovaquone/proguanil (Malarone®) are detailed in the prescribing information provided by manufacturers. Participants who have any known contraindication to any of the definitive antimalarial medications according to the applicable labelling at screening will be excluded from participating in the study.

Primacin® may cause severe haemolytic anaemia in participants with glucose-6-phosphate dehydrogenase (G6PD) deficiency. Participants will be tested for G6PD deficiency at screening to ensure the safety of Primacin®. Participants with G6PD deficiency will not be eligible for the study.

Overall risk management

The overall risks to the participants in this study will be managed by frequent safety monitoring, including close monitoring during the period of confinement at the clinical trial unit following single oral dose administration of MMV367. Safety monitoring will include clinical laboratory safety tests (biochemistry, haematology, and urinalysis), physical examination, vital signs, ECG analysis and adverse event monitoring. Throughout the study, the safety, parasitaemia and PK data will be assessed after the completion of each cohort by the Safety and Data Review Team (SDRT) which will include the Principal Investigator, the Sponsor Medical Director, the partner organisation (GSK plc) Medical Director and the Medical Monitor. Specific details will be provided in the SDRT Charter available at study start.

2.2.2 KNOWN POTENTIAL BENEFITS

There are no expected clinical benefits for the healthy participants who will participate in this trial.

2.2.3 ASSESSMENT OF POTENTIAL RISKS AND BENEFITS

On the basis of the safety provisions and risk minimisation activities outlined in Section 2.2.1, the overall risk to the participants in this trial is considered to be minimal and acceptable, and the potential of future improved treatment for malaria is considered to outweigh these potential risks.

3 OBJECTIVES AND ENDPOINTS

OBJECTIVES	ENDPOINTS
Primary	
To characterise the PK/PD relationship of MMV367 in healthy participants experimentally infected with blood stage *P. falciparum.*	The PK/PD relationship between MMV367 blood concentrations and blood stage asexual parasitaemia.
Secondary	
To evaluate the safety and tolerability of single oral doses of MMV367 in healthy participants experimentally infected with blood stage *P. falciparum*.	The incidence, severity and relationship to MMV367 of adverse events as determined by self-reported symptoms, clinical laboratory analysis, vital signs, physical examinations and ECG assessments.
To evaluate the safety and tolerability associated with blood-stage malaria infection in healthy malaria naïve participants.	The incidence, severity and relationship to the malaria challenge agent of adverse events as determined by self-reported symptoms, clinical laboratory analysis, vital signs, physical examinations and ECG assessments. Additionally, the severity of the induced malaria infection in each participant graded by the malaria clinical score.
To characterise the pharmacokinetics of MMV367 following single oral dose administration in healthy participants experimentally infected with blood stage *P. falciparum*.	The pharmacokinetics of MMV367 calculated using non-compartmental methods (C_{max}, t_{max}, AUC_{last}, AUC_{inf}, CL/F, Vz/F, λ, $t_{1/2}$).
To characterise the parasite clearance kinetics following single doses of MMV367 in healthy participants experimentally infected with blood-stage *P. falciparum*.	The parasite clearance kinetics following dosing with MMV367 (parasite reduction ratio over a 48-hour period [PRR_{48}] and corresponding parasite clearance half-life [$PCt_{1/2}$]).
Exploratory	
To investigate potential resistance of *P. falciparum* to MMV367 in participants experiencing parasite regrowth after MMV367 administration.	Incidence and level of resistance to MMV367 in parasites determined using *in vitro* methods.
To characterise the effect of MMV367 on *P. falciparum* parasite viability.	Effect of MMV367 on parasite viability examined by *ex vivo* cultures.

4 STUDY DESIGN

4.1 OVERALL DESIGN

This is an open-label, adaptive study that will utilise the *P. falciparum* induced blood stage malaria (IBSM) model to characterise the pharmacokinetic/pharmacodynamic (PK/PD) profile and safety of MMV367 (the IMP). Up to 18 participants will be enrolled in cohorts of up to 6 participants each. The study will proceed as follows for all participants:

- Screening period of up to 28 days to recruit healthy adult volunteers.
- Day 0: Intravenous inoculation with approximately 2,800 viable *P. falciparum*-infected erythrocytes.
- Days 1-3: Daily follow up via phone call or text message.
- Days 4-7: Daily site visits for clinical evaluation and blood sampling to monitor malaria parasitaemia via quantitative polymerase chain reaction (qPCR).
- Day 7 PM: Start of confinement within the clinical trial unit.
- Day 8: Administration of a single oral dose of the IMP (MMV367). Different doses of MMV367 will be administered across and within cohorts in order to effectively characterise the PK/PD relationship (see Section 4.3 for dose selection rationale).
- Days 8-11: Regular clinical evaluation and blood sampling to monitor malaria parasitaemia and measure MMV367 plasma concentration.
- Day 11 AM: End of confinement within clinical trial unit.
- Days 12-23: Outpatient follow-up for clinical evaluation and blood sampling.
- Day 24: Initiation of compulsory definitive antimalarial treatment with Riamet® (artemether/lumefantrine) and/or other registered antimalarials if required (see section 6.1). Treatment will be initiated earlier than Day 24 in the event of:
  - Insufficient parasite clearance following IMP dosing (parasitaemia not reduced ≥10-fold by Day 10 compared with peak parasitaemia on Day 8).
  - Parasite regrowth following IMP dosing (initial parasite clearance is followed by asexual parasite regrowth above 5000 parasites/mL).
  - Participant discontinuation/withdrawal (see Section 7),
  - Investigator's discretion in the interest of participant safety.
- Day 27: End of study visit for final clinical evaluation and to ensure complete clearance of malaria parasitaemia (at least one negative malaria PCR result required).

4.2 SCIENTIFIC RATIONALE FOR STUDY DESIGN

The study design of this trial using the IBSM model has been used previously to safely and effectively determine the PK/PD relationship of several antimalarial agents [2-5,11]. The *P. falciparum* 3D7 challenge agent to be used to initiate blood-stage malaria infection in this study has been well characterised, with consistent and reproducible parasite growth occurring following intravenous inoculation of healthy participants in previous studies. Experience from these studies indicates that parasitaemia will be above

5,000 parasites/mL on Day 8 following inoculation, but clinical symptoms of malaria will either be absent or only mild to moderate in severity.

The duration of confinement within the clinic following dosing with MMV367 on Day 8 (72 hours) is considered appropriate to ensure close monitoring of participants for safety purposes and to enable frequent blood sampling for parasitaemia and drug concentration measurements. Blood sampling time points have been selected with consideration of human PK data from the FIH study of MMV367, as well as preclinical data on the antimalarial activity of MMV367. These time points may be adjusted during the study, based on the emerging data in order to optimally determine the exposure/response relationship of MMV367.

The follow-up period from MMV367 dosing on Day 8 to initiation of compulsory definitive antimalarial treatment on Day 24 (16 days) is equal to approximately 22 times the elimination half-life of MMV367 (approximately 17 hours). This duration was selected to allow sufficient time for parasite regrowth, which is required to define the PK/PD relationship of MMV367. Further, this duration will ensure that participants have a negligible blood concentration of the IMP at the completion of the study on Day 27.

Compulsory treatment with the registered antimalarial Riamet® (artemether/lumefantrine) will occur for all participants to ensure all parasites are cleared at the completion of the study. *P. falciparum* 3D7 parasites are known to be sensitive to artemether/lumefantrine, and Riamet® has been effective in clearing parasitaemia in previous IBSM studies using this challenge agent. A single dose of Primacin® will be used to ensure clearance of gametocytes (the life cycle stage responsible for transmission to mosquitoes) at the Investigator's discretion. An alternate registered antimalarial treatment Malarone® (atovaquone/proguanil) will be used in the event of contraindication to Riamet®. Malarone has also previously been effective in clearing *P. falciparum* 3D7 parasitaemia [10]. If in the rare event participants are unable to complete oral antimalarial treatment, they will be admitted to hospital to receive intravenous artesunate, the recommended parenteral treatment of malaria in Australia.

4.3 JUSTIFICATION FOR DOSE

MMV367 will be administered as a single oral dose on Day 8, with different doses to be tested across and within cohorts to effectively characterise the PK/PD relationship. Participants will be randomised to a dose group on the day of dosing.

Rationale for highest dose

The highest dose of MMV367 administered in this study will be no more than 1500 mg. Testing of this dose may be required to determine the maximum parasite killing rate of MMV367. This dose was previously shown to be well tolerated by healthy volunteers and is equal to the maximum single dose of MMV367 administered in the FIH SAD study. There were no IMP-related AEs or SAEs recorded in the 1500 mg dose cohort (n=8, randomised 6 IMP: 2 placebo), and the incidence of all AEs was no higher than in lower dose cohorts. Additionally, PK data indicated that the mean group exposure following dosing with 1500 mg MMV367 did not exceed the exposure cap set for this FIH study. The exposure cap was 376

µg.h/mL for AUC_{inf} and 23.8 µg/mL for C_{max}. Dosing with 1500 mg MMV367 in the FIH study was associated with a mean group exposure of 183 µg.h/mL for AUC_{inf} and 5.43 µg/mL for C_{max}. Thus, a safety margin exists in the event of higher exposure in the healthy participants with induced blood-stage malaria.

Dose selection for Cohort 1

The doses to be tested in Cohort 1 will be as follows:

MMV367 dose	20 mg	90 mg	1500 mg
Number of participants	3	2	1

Preclinical data indicates that MMV367 exhibits potent and fast acting activity against asexual blood stage *P. falciparum*. Preliminary modelling using human PK data generated in the FIH study combined with preclinical PD data (humanised mouse model of malaria) has allowed estimation of efficacious doses in humans with acute uncomplicated *P. falciparum* malaria.

Parasite regrowth following initial parasite clearance is required to effectively characterise PK/PD parameters such as the half maximum effective concentration (EC_{50}) of MMV367. A single dose of 20 mg is expected to result in initial parasite clearance in the IBSM model (log_{10} parasite reduction ratio over a 48 h period [PRR_{48}] median 2.9, 95% CI: 0.26 - 5.8), but subsequent parasite regrowth is expected to occur. It is predicted that parasitaemia will return to the pre-dose baseline with a median of 7 days post-dosing (95% CI: 2.5 - 14). Additionally, modelling indicates that there is a 50% chance for a typical individual dosed with 20 mg to have a full parasitaemia profile above the lower limit of quantification (LLOQ) of the qPCR assay. Thus, a significant amount of data is expected to be generated at this dose level and three participants will be dosed with 20 mg MMV367 in Cohort 1.

Testing of 90 mg MMV367 is considered important because it is predicted to represent an “intermediate” dose level. That is, modelling indicates that 90 mg will result in faster parasite clearance (log_{10} PRR_{48} median 3.6, 95% CI: 1.8 – 6.2) compared with 20 mg for a typical individual, while retaining a good chance of parasite regrowth occurring and thus contributing to refining the MMV367 EC_{50} estimate. Two participants will receive 90 mg MMV367 in Cohort 1.

Testing of 1500 mg MMV367 is required to characterise the PK/PD parameters across the full dose range, in particular the maximum killing rate (E_{max}). The chance of parasite regrowth occurring at this dose level is considered very low and thus data from this dose level will have a long duration of observable maximum killing rate ensuring a reliable estimation of the maximum clearance rate (E_{max}), however it is not anticipated to contribute to refining the estimate of the EC_{50}. Therefore, one participant dosed with 1500 mg is considered sufficient in the first cohort.

Dose selection for Cohort 2

The SDRT will meet following the completion of Cohort 1 to review all safety, PK, and PD data. Doses to be tested in Cohort 2 will be informed by the results obtained in Cohort 1, and will be selected to refine the PK/PD parameter estimates. It may be necessary to test a dose lower than 20 mg in Cohort 2 if PK/PD parameters such as the EC_{50} were not estimated with sufficient certainty and precision in Cohort 1. It may also be necessary to obtain additional data at doses previously tested in Cohort 1. The maximum dose in Cohort 2 will not exceed 1500 mg. Specific doses to be tested in Cohort 2 will be decided by the SDRT based on PK/PD simulations.

Dose selection for Cohort 3 (if required)

The SDRT will meet following the completion of Cohort 2 to review all safety, PK, and PD data. If the SDRT deems sufficient data has been obtained to achieve the primary endpoint (i.e., the PK/PD relationship of MMV367 has been appropriately characterised in the human challenge model), a third cohort of participants will not be enrolled. If the SDRT deems further data is necessary, doses for Cohort 3 will be selected as described above for Cohort 2. The maximum dose in Cohort 3 will not exceed 1500 mg.

4.4 END OF STUDY DEFINITION

A participant is considered to have completed the trial if they have completed all phases of the trial including the last visit or the last scheduled procedure shown in the Schedule of Activities (Section 1.3). The end of the trial globally is defined as the time at which all participants have completed the trial and all study analyses have been completed and reported.

5 STUDY POPULATION

5.1 INCLUSION CRITERIA

Volunteers must fulfil all of the following criteria to be eligible for inclusion in this trial:

Demographics and general considerations

1. Healthy adults aged 18 to 55 years inclusive who will be contactable and available for the duration of the trial and up to two weeks following the EOS visit.
2. Total body weight greater than or equal to 50 kg, and a body mass index (BMI) within the range of 18 to 32 kg/m^2 (inclusive). BMI is an estimate of body weight adjusted for height. It is calculated by dividing the weight in kilograms by the square of the height in metres.
3. Completion of the written informed consent process prior to undertaking any trial-related procedure.
4. Must be willing and able to communicate and participate in the whole trial.
5. Agreement to adhere to Lifestyle Considerations (Section 5.3) throughout the trial duration.

6. Must be able to provide contact details of a support person (responsible adult) who is aware of the participant's participation in the study and is available to provide assistance if required (for example with contacting the participant in the event that study staff are unable to, or with transporting the participant to and from the study site if required).

Vital signs and ECG parameters

7. Vital signs at screening (measured after 5 min in the supine position):
    - Systolic blood pressure (SBP): 90–140 mmHg,
    - Diastolic blood pressure (DBP): 40–90 mmHg,
    - Heart rate (HR): 40–100 bpm.

Note: Symptomatic postural hypotension will be assessed by measuring SBP and DPB in the standing position (see exclusion criterion 10).

8. At Screening and pre-inoculation with the malaria challenge agent (Day 0), normal standard mean of triplicate 12-lead electrocardiogram (ECG) parameters after 5 minutes resting in supine position:
    - QTcF: ≤450 msec (males) or ≤470 msec (females),
    - QRS: 50–120 msec,
    - PR interval: ≤ 210 msec,
    - Normal ECG tracing unless the Principal Investigator or delegate considers an ECG tracing abnormality to be not clinically relevant.

Contraception

9. Women of childbearing potential (WOCBP) who anticipate being sexually active with a male during the trial must agree to use a highly effective method of birth control (see below) combined with a barrier contraceptive from the screening visit until 34 days after the last dose of MMV367 (covering a full menstrual cycle of 30 days starting after 5 half-lives of last dose of MMV367) and have a negative urine pregnancy test result prior to inoculation with the malaria challenge agent on Day 0.

    - Highly effective birth control methods include: combined (oestrogen and progestogen containing) oral/intravaginal/transdermal/implantable hormonal contraception associated with inhibition of ovulation, progestogen-only oral/injectable/implantable hormonal contraception associated with inhibition of ovulation, intrauterine device, intrauterine hormone-releasing system, bilateral tubal occlusion, vasectomised partner, or sexual abstinence or same sex relationship.

    - Female participants who are abstinent (from penile-vaginal intercourse) must agree to start a double method if they start a sexual relationship with a male during the study. Female participants must not be planning in vitro fertilisation within the required contraception period.

    Women of non-childbearing potential (WONCBP) are defined as:

- Natural (spontaneous) post-menopausal defined as being amenorrhoeic for at least 12 months without an alternative medical cause with a screening follicle stimulating hormone level (FSH) >25 IU/L (or at the local laboratory levels for post-menopause).
- Premenopausal with irreversible surgical sterilization by hysterectomy and/or bilateral oophorectomy or salpingectomy at least 6 months before screening (as determined by participant medical history).

10. Males who have, or may have, female sexual partners of childbearing potential during the course of the study must agree to use a double method of contraception including condom plus diaphragm, or intrauterine device, or stable oral/transdermal/injectable/implantable hormonal contraceptive by the female partner, from the time of informed consent through to 94 days after MMV367 administration. This has been calculated based on 90 days (one cycle of spermatogenesis) plus 5 half-lives of the IMP (4 days). Abstinent males must agree to start a double method if they begin a sexual relationship with a female during the study and up to 94 days after the last dose of MMV367. Males that are surgically sterile, or who have undergone sterilisation and have had testing to confirm the success of the sterilisation, may also be included and will not be required to use above described methods of contraception.

5.2 EXCLUSION CRITERIA

Volunteers will be excluded from participating in the study if any of the following criteria apply:

Medical history

1. Known hypersensitivity to artesunate or other artemisinin derivatives, lumefantrine, proguanil/atovaquone, primaquine, or 4-aminoquinolines.
2. Any history of anaphylaxis or other severe allergic reactions, or other food or drug allergy that the Investigator considers may impact on participant safety.
3. History of convulsion (including drug or vaccine-induced episodes). A medical history of febrile convulsion during childhood (< 5 years) is not an exclusion criterion.
4. Presence of current or suspected uncontrolled chronic diseases that may impact participant safety or interpretation of clinical trial results, such as (but not limited to) cardiac or autoimmune disease, diabetes, progressive neurological disease, severe malnutrition, hepatic or renal disease, epilepsy, or asthma.
5. History of malignancy of any organ system (other than localised basal or squamous cell carcinoma of the skin or *in situ* cervical cancer), treated or untreated, within five years of screening, regardless of whether there is no evidence of local recurrence or metastases.
6. Individuals with history of schizophrenia, bipolar disorder psychoses, attempted or planned suicide, or any other severe (disabling) chronic psychiatric diagnosis including generalised anxiety disorder.
7. History of an episode of depression lasting more than 6 months that required pharmacological therapy and/or psychotherapy within the last 2 years.

8. A score of 20 or more on the Beck Depression Inventory-II (BDI-II) and/or a response of 1, 2 or 3 for item 9 of this inventory (related to suicidal ideation).

    - The BDI-II will be used as a validated tool for the assessment of depression at screening. Participants that meet criterion 8 will be referred to a general practitioner or medical specialist as appropriate. Participants with a BDI-II score of 17 to 19 may be enrolled at the discretion of the Investigator if they do not have a history of the psychiatric conditions mentioned in criterion 6 and their mental state is not considered to pose additional risk to the health of the participant during the trial or to the execution of the trial and interpretation of the data gathered.

9. History of splenectomy.
10. Symptomatic postural hypotension at screening (confirmed on two consecutive readings), irrespective of the decrease in blood pressure, or asymptomatic postural hypotension defined as a decrease of SBP of ≥20 mmHg after 3 min standing and/or a decrease of DBP of ≥10 mmHg after 3 min standing. This 3 min standing period will commence after the volunteer has rested for 5 min in the supine position.
11. Cardiac/QT risk:
    - Family history of sudden death or of congenital prolongation of the QTc interval or known congenital prolongation of the QTc interval or any clinical condition known to prolong the QTc interval.
    - History of symptomatic cardiac arrhythmias or of clinically relevant bradycardia.
12. Evidence of increased cardiovascular disease risk (defined as >10%, 5-year risk for those greater than 35 years of age, as determined by the Australian Absolute Cardiovascular Disease Risk Calculator [http://www.cvdcheck.org.au/]). Risk factors include sex, age, systolic blood pressure (mm/Hg), smoking status, total and HDL cholesterol (mmol/L), and reported diabetes status.
13. Presence of clinically significant infectious disease or fever (e.g., sublingual temperature ≥38°C) within the five days prior to inoculation.

Prior medications and treatments

14. Any COVID-19 vaccine within 14 days of malaria inoculation, any other vaccination within 28 days of IMP dosing, and any vaccination planned during the study.
15. Use of prescription drugs (excluding contraceptives), investigational medical products, or non-prescription drugs or herbal supplements, that in the opinion of the investigator may potentially interfere with study interventions, within 14 days or five half-lives (whichever is longer) prior to inoculation. Requirements for concomitant medication use (from inoculation until the end of study) are specified in Section 6.5.
16. Individual who has ever received a blood transfusion.

Malaria exposure

17. Any history of malaria or participation in a previous malaria challenge trial or malaria vaccine trial.

18. Must not have had malaria exposure that is considered by the Principal Investigator or their delegate to be significant. This includes but is not limited to: history of having travelled to or lived (>2 weeks) in a malaria-endemic region during the past 12 months or planned travel to a malaria-endemic region during the course of the trial; history of having lived for >1 year in a malaria-endemic region in the past 10 years; history of having ever lived in a malaria-endemic region for more than 10 years inclusive. For endemic regions see https://malariaatlas.org/explorer/#/, Bali is not considered a malaria-endemic region.

Alcohol use and smoking

19. History or presence of alcohol abuse (regular alcohol consumption in males >21 units per week and females >14 units per week (1 unit = ½ pint beer, or a 25 mL shot of 40% spirit, 1.5 to 2 units = 125 mL glass of wine, depending on type), or drug habituation, or any prior intravenous usage of an illicit substance.
20. Any individual who currently smokes cigarettes on a daily basis (including e-cigarettes, vaping, and other nicotine use).

Blood donation

21. Blood product donation to any blood bank during the 8 weeks (whole blood) or 4 weeks (plasma and platelets) prior to admission in the clinical unit on Day 8.
22. Individual unwilling to defer blood donations for at least twelve months after the EOS visit.

Laboratory results

23. Haematology, biochemistry or urinalysis results at screening or at the eligibility visit (Day -1 to Day -3) that are outside of the standard clinically acceptable laboratory ranges (Appendix 12.2) or are considered clinically significant by the Principal Investigator.
24. Positive result for: hepatitis B surface antigen (HBs Ag), anti-hepatitis B core antibodies (anti-HBc Ab), anti-hepatitis C virus (anti-HCV) antibodies, anti-human immunodeficiency virus 1 and 2 antibodies (anti-HIV1 and anti-HIV2 Ab), COVID-19 by PCR at Screening or RAT on Day 0, red blood cell alloantibodies.
25. Positive urine drug test. Any drug listed in the urine drug screen unless there is an explanation acceptable to the Investigator (e.g., the participant has stated in advance that they consumed a prescription or over-the-counter product that contained the detected drug) and the participant has a negative urine drug screen on retest by the pathology laboratory.
26. G6PD deficiency (result below the lower limit of the laboratory reference range for quantitative G6PD test).
27. Positive alcohol breath test.
28. Positive serum pregnancy test at screening or eligibility visit, positive urine pregnancy test on Day 0.

Other

29. Individual who, in the judgement of the Investigator, is likely to be non-compliant during the trial
30. Individual who is an Investigator, research assistant, pharmacist, trial coordinator, or other staff thereof, directly involved in conducting the trial.

31. Individual without good peripheral venous access.
32. Individual who is breastfeeding or lactating.

5.3 LIFESTYLE CONSIDERATIONS

While participating in this trial, participants are asked to:

- Refrain from alcohol consumption of more than 20 g/2 units/2 standard drinks per day.
- Refrain from tobacco use (includes smoking, e-cigarettes, vaping, and other nicotine use) throughout the study.
- Abstain from any alcohol for the duration of clinical trial unit confinement.
- Abstain from any illicit drug habituation.
- Refrain from eating food containing poppy seeds for 48 hours prior to screening, malaria inoculation, and IMP administration.
- Refrain from excessive consumption of beverages or food containing xanthine bases including Red Bull, chocolate, coffee etc. (more than 400 mg caffeine per day, equivalent to more than 4 cups of coffee per day).
- Refrain from consumption of quinine containing foods/beverages such as tonic water and lemon bitter.
- Abstain from strenuous exercise and/or altering their regular exercise program.

5.4 SCREEN FAILURES

A screen failure occurs when a volunteer consents to participate in the clinical study but is not subsequently enrolled (inoculated with the malaria challenge agent). Volunteers who do not fulfil all the inclusion criteria, and/or fulfil any of the exclusion criteria, should not be enrolled. However, repeat assessments may be performed to confirm and follow-up on out-of-range clinical laboratory test, vital sign, and/or ECG values that determine a volunteer's eligibility.
If a volunteer does not meet all eligibility criteria, and is hence a screen failure, but at some point in the future is expected to meet the eligibility criteria, the volunteer may be re-screened for future cohorts. Volunteers who are re-screened will be re-consented and assigned a new volunteer number and then restart a new screening phase.

A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants, to meet the Consolidated Standards of Reporting Trials (CONSORT) publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, and screen failure details with respect to the eligibility criteria. Screen failure information will not be entered into the eCRF. Volunteers who fail screening due to an underlying medical condition previously unknown to them will be reimbursed for their time, and provided with the appropriate referrals for guidance and counselling for their condition.

5.5 STRATEGIES FOR RECRUITMENT AND RETENTION

Up to 18 healthy, adult, malaria- naïve participants will be enrolled in this trial. It is anticipated that up to 9 reserve participants will be required on Day 0 (3 per cohort) to ensure an adequate number of participants are enrolled and administered the malaria challenge agent.

Participants will be recruited by general or trial specific advertising via print, radio, social media, poster or other media, as approved by the HREC(s).

Participants who withdraw or are withdrawn from the trial will be compensated on a fractional basis for their involvement unless they are withdrawn as a consequence of their misconduct.

Reserve participants who do not participate in the trial will be compensated for the inconvenience associated with their attendance for screening and attendance on Day 0.
The monetary value of the compensation is documented in the Participant Information Sheet.

6 STUDY INTERVENTION

6.1 STUDY INTERVENTIONS ADMINISTRATION

6.1.1 STUDY INTERVENTION DESCRIPTION

IMP
MMV367 belongs to the pyrrolidinamide class of antimalarial compounds discovered using a high-throughput whole cell phenotypic screen. Preclinical studies indicated that MMV367 exhibits potent antimalarial activity against blood-stage *P. falciparum*, with a novel mode of action involving acyl coenzyme A synthetase 10 and 11.

Challenge agent
A *P. falciparum* 3D7 master cell bank (MCB) was produced from a participant with type O Rh (D) negative blood who was infected with the parasite by mosquito bite. Blood was collected and aliquoted into cryovials and stored in liquid nitrogen under controlled conditions. A cryovial will be retrieved from storage, thawed, and used to aseptically prepare the intravenous inoculum at Q-Gen Cell Therapeutics (QIMR Berghofer).

Definitive antimalarial medications
Riamet® (Novartis Pharmaceuticals Australia Pty Ltd) is an oral fixed dose combination therapy containing artemether and lumefantrine. Riamet® is registered in Australia for the treatment of acute uncomplicated malaria.

Malarone® (GlaxoSmithKline Australia Pty Ltd) is an oral fixed dose combination therapy containing atovaquone and proguanil hydrochloride. Malarone® is registered in Australia for the treatment of acute uncomplicated malaria.

Primacin® (Boucher & Muir Pty Ltd) contains the active ingredient primaquine phosphate. Primaquine phosphate is an 8-aminoquinoline effective against the sexual forms (gametocytes) of *Plasmodium* species which are responsible for transmission to mosquitoes. Primacin® is registered for use in Australia.

Artesunate (powder and diluent for reconstitution, imported by Link Pharmaceuticals) is an artemisinin derivative; the intravenous formulation is typically used to treat severe malaria. Although parenteral artesunate does not currently have marketing approval in Australia, the Therapeutic Goods Administration (TGA) allows importation of artesunate for Category A use in patients with severe malaria under the Special Access Scheme (SAS).

6.1.2 DOSING AND ADMINISTRATION

IMP

A single oral dose of MMV367 will be administered on Day 8 after an overnight fast of ≥ 8 hours. The IMP will be administered as an oral suspension in sterile water. Specific dosing instructions will be documented in the pharmacy manual. Participants will be required to remain fasted for 4 hours after dosing. Different doses of MMV367 will be tested across and within cohorts.

The doses to be tested in Cohort 1 will be as follows:

MMV367 dose	20 mg	90 mg	1500 mg
Number of participants	3	2	1

Specific doses to be tested in subsequent cohorts will be decided by the SDRT based on emerging data during the study (safety, PK, and PD data from preceding cohorts). The maximum dose to be tested in the study will be no more than 1500 mg.

Challenge Agent

The malaria challenge agent, containing an estimated 2,800 viable *P. falciparum* 3D7 parasite-infected erythrocytes in a volume of 2 mL, will be administrated intravenously on Day 0. The actual number of parasites inoculated will take into account the loss of viability resulting from cryopreservation, storage and thawing. Participants will undergo intravenous cannulation. The inoculum will be injected, and the cannula flushed with clinical grade saline. The cannula will then be removed. An extra syringe will be prepared to quantify the parasite count of the challenge agent by PCR.

Definitive antimalarial medications

Compulsory definitive antimalarial treatment for all participants will be initiated on Day 24, if not initiated earlier in accordance with the criteria described in Section 4.1.

Riamet® tablets will be administered as six oral doses of four tablets (total course of 24 tablets equivalent to 480 mg artemether and 2.88 g lumefantrine). The second dose of four tablets will be administered 8±1 h after the first dose. The remaining doses will be administered twice daily (morning and evening).

Malarone® will be utilised as a backup medication to Riamet® in the event of allergy or contraindication to Riamet®. A treatment course consists of four tablets administered daily for three days (total course of 12 tablets equivalent to 3 g atovaquone and 1.2 g proguanil hydrochloride).

Participants may be treated with Primacin® at the Investigator's discretion to ensure clearance of gametocytes. If needed, participants will be administered a single oral dose of six Primacin® tablets (total dose 45 mg primaquine) with food.

Intravenous artesunate may be administered in the event a participant vomits or cannot tolerate oral drugs. In this case, the participant will be admitted to hospital for treatment. Artesunate will be administered as 2.4 mg/kg IV bolus on admission, with repeat dosing at 12 hours and 24 hours, then once daily until oral therapy is possible.

6.2 PREPARATION/HANDLING/STORAGE/ACCOUNTABILITY

6.2.1 ACQUISITION AND ACCOUNTABILITY

Acquisition

IMP
MMV367 was manufactured by Piramal Pharma Solutions (India). The Sponsor will provide the IMP to the clinical trial unit.

Challenge agent
Aliquots of the *P. falciparum* 3D7 MCB are stored in liquid nitrogen under controlled conditions at QIMR Berghofer. On the day of inoculation, cryovials will be retrieved from storage and used to aseptically prepare the challenge agent at Q-Gen Cell Therapeutics (QIMR Berghofer). The challenge will then be transported to the clinical trial unit under controlled conditions.

Definitive antimalarial medications
Riamet® (distributed by Novartis Pharmaceuticals Pty Ltd), Primacin® (distributed by Boucher & Muir Pty Limited), and Malarone® (distributed by GlaxoSmithKline Australia Pty Ltd) will be acquired by the clinical trial unit. Artesunate for intravenous administration is available within the Central Pharmacy of Queensland Health.

Accountability

The clinical trial unit pharmacist or delegate is responsible for maintaining accurate study intervention accountability records throughout the study. Dispensing, accountability and documentation will be in accordance with the clinical trial unit standard procedures.

All products will be inventoried upon receipt by the clinical trial unit pharmacist. The condition of the products at the time of receipt by the pharmacist will be documented, as will the time restrictions of use for the syringes containing the challenge agent. The clinical trial unit pharmacist or designee will be responsible for maintaining the accurate *Plasmodium falciparum* 3D7 Challenge Agent Accountability Log as per clinical trial unit standard operating procedures (SOPs). The lot numbers and expiry dates of all study interventions will be documented. The clinical trial unit pharmacist or delegate will ensure that the received products are the specified formulation.

The storage, handling and the disposal of the products will be in accordance with approved procedures. All dosages prescribed and dispensed to the participants and all dose changes during the study must be recorded in the eCRFs and accountability logs. All drug supplies are to be used only in accordance with this protocol, and not for any other purpose. All used medications will be fully documented. All used product containers and packaging (and unused IMP and challenge agent) must be destroyed at the clinical trial unit once drug accountability is final and has been checked by the Sponsor or its delegate, and written permission for destruction has been obtained from the Sponsor.

Study products and study accountability logs will be available to the Sponsor or their representative as part of the study monitoring procedures. Upon completion of the study, copies of all study drug management records will be provided to the Sponsor. Original records will be maintained at the clinical trial unit with the rest of the study records.

6.2.2 FORMULATION, APPEARANCE, PACKAGING, AND LABELLING

The contents of the dispensing labels for the IMP, the malaria challenge agent, and the definitive antimalarial medications will be in accordance with all applicable regulatory requirements.

IMP

MMV367 is formulated as dispersible granules (250 mg/g [25% w/w]). The drug substance was blended with excipients (mannitol, microcrystalline cellulose, povidone, sodium lauryl sulfate, xanthan gum and fumed silica) to improve its oral bioavailability. Aliquots of the powder will be reconstituted in sterile water for oral administration of different doses.

Challenge agent

The malaria challenge agent will contain parasitised and unparasitised erythrocytes, resuspended in 0.9% Sodium Chloride Intravenous Infusion, in a total volume of 2 mL in syringes. The syringes will be double contained following preparation and labelled in accordance with GCP guidelines. Labelling is in accordance with PIC/S Annex 13.

Definitive antimalarial medications

Riamet® tablets (each containing 20 mg artemether/120 mg lumefantrine) are yellow, round, flat tablets marked with N/C and a score line on one side and CG on the other side. Each carton contains 24 tablets.

Malarone® tablets are round, pink and film-coated, and are engraved with "GX CM3". Malarone® tablets are supplied in blister packs of 12 or 24 tablets.

Primacin® tablets (each containing 13.2 mg primaquine phosphate equivalent to 7.5 mg primaquine) are round, flat, orange uncoated tablets available in bottles of 28 or 56 tablets.

Artesunate powder for reconstitution is supplied in vials containing 60 mg artesunate. Each vial is reconstituted with an ampoule (1 mL) of sodium bicarbonate 5%. Sodium chloride 0.9 % (5 mL) is then added to reconstituted artesunate vial to create a 10 mg/mL solution (total volume 10 mL).

6.2.3 PRODUCT STORAGE AND STABILITY

All study interventions will be held in appropriate locked storage conditions at the clinical trial unit until required.

IMP

MMV367 dispersible granules are to be stored between 15°C and 25°C and protected from moisture. The reconstituted IMP for oral administration should be prepared within 8 hours of each administration.

Challenge agent

The *P. falciparum* 3D7 challenge agent is prepared at Q-Gen on inoculation day (Day 0). The time between preparation of the challenge agent and administration to the participant will be a maximum of 4 hours. The syringes containing the challenge agent will be stored in a temperature monitored validated transport container at 2-15°C during transportation from Q-Gen to the clinical trial unit and will be immediately transferred to a temperature monitored onsite refrigerator (2-15°C). The clinical trial unit pharmacist or delegate will document receipt conditions and time restrictions of use. The challenge agent will then be dispensed to participants as per written prescription. Transporter logs should be provided to the Sponsor.

Definitive antimalarial medications

Riamet® tablets are to be stored below 30°C and protected from moisture. Primacin® tablets are to be stored below 25°C. Malarone® tablets are to be stored below 30°C. The reconstituted artesunate for intravenous administration should be prepared freshly for each administration and should not be stored.

6.2.4 PREPARATION

IMP

The IMP will be prepared at the clinical trial unit by reconstituting the dispersible granules in sterile water for oral administration. Detailed preparation instructions will be provided in the pharmacy manual.

Challenge agent

The challenge agent will be prepared aseptically at Q-Gen from frozen cryovials of the *P. falciparum* 3D7 MCB by nominated QIMR Berghofer staff members. The erythrocytes will be thawed, washed, resuspended in saline, diluted in a final volume of 2 mL of clinical grade saline, and dispensed into syringes. Any remaining unused erythrocytes will be discarded as per approved SOPs.

Definitive antimalarial medications

Riamet®, Primacin® and Malarone® are available as tablets and no preparation is required. Artesunate for intravenous administration will be prepared according to manufacturer's instructions within the hospital setting immediately prior to administration.

6.3 MEASURES TO MINIMIZE BIAS: RANDOMIZATION AND BLINDING

All participants will receive an identification number (screening number) as soon as they have signed the informed consent form. Participants who meet the eligibility criteria and are enrolled in the study will be assigned an enrolment number on Day 0 immediately prior to administration of the malaria challenge agent. Participants will be randomised within each cohort to a dose group on Day 8, immediately prior to administration of the IMP. A randomisation schedule will be prepared in advance and take in account potential participant withdrawals prior to scheduled dosing with IMP. The details of the randomisation procedure will be described in the randomisation plan. This is an open label study and therefore no blinding will be performed.

6.4 STUDY INTERVENTION COMPLIANCE

The Investigator will administer the malaria challenge agent intravenously at the clinical trial unit on Day 0. The IMP (single oral dose of MMV367) will be administered in a fasted state at the clinical trial unit on Day 8 under direct observation by staff. The first dose of definitive antimalarial medication (Riamet®, or Malarone® if required) will be administered at the clinical trial unit under direct observation by staff (except in the event a participant tests positive to COVID-19; See Section 8.2.6.11 for details of COVID-19 management plan). The subsequent doses may be taken at home. Participants will receive a phone call or text message from the clinical trial unit staff to ensure compliance.

If Primacin® is required to clear gametocytaemia, it will be administered at the clinical trial unit under direct observation by staff. If a participant requires intravenous artesunate, the participant will be admitted to hospital for treatment.

6.5 CONCOMITANT THERAPY

Concomitant medications, treatments and procedures are those occurring from the time of administration of the malaria challenge agent (Day 0), until EOS. Those occurring prior to Day 0 are

classified as prior medications, treatments and procedures. Medications taken within 28 days before Day 0 will be recorded as prior medication.

On Day 0, participants will be questioned in relation to relevant aspects of compliance with the study protocol, including drug intake since their screening visit. Details of all drugs taken (prescription and over-the-counter, systemic and topical administration) will be recorded at this time and appropriate action taken.

From Day 0 until the EOS visit, no medications will be permitted to be taken without the approval of the Investigator (with the exception of contraceptives and ibuprofen/paracetamol). Ibuprofen (preferred) may be administered at doses of up to 1.6 g/24 hours, or paracetamol up to 4 g/24 hours with investigator approval, for treatment of headache or other pain if required. Ibuprofen is the preferred treatment for headache or pain. To minimise the risk of liver enzyme elevation, use of paracetamol should be kept to a minimum if possible. However, since paracetamol may be required by some participants its use is not excluded. All other medications will be assessed by the investigator on a case by case basis with regard to the participant's wellbeing and potential for the medication to interfere with study interventions.

Any medication taken during the study for treatment of a medical condition or AE is to be recorded in the concomitant medication pages in the eCRF. The exact dose, route and timing of each dose should be recorded.

6.5.1 RESCUE MEDICINE

All participants will be administered definitive antimalarial treatment in this study. Details of these medications are included in Sections 6.1 and 6.2.

7 STUDY INTERVENTION DISCONTINUATION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

7.1 DISCONTINUATION OF STUDY INTERVENTION

Once inoculated with the challenge agent on Day 0, it is expected that some study participants may experience mild to moderate malaria signs and symptoms prior to scheduled dosing with the IMP on Day 8. The following criteria should be considered as guidance for the decision not to proceed with administration of the IMP on Day 8 and to administer definitive antimalarial treatment instead:

1. A participant experiences an SAE or severe AE (≥ CTCAE Grade 3).
2. A participant returns a positive alcohol or urine drug screen.
3. A participant tests positive for SARS-CoV-2.
4. Investigator decision based on other safety signals (clinical, ECG, vitals and laboratory tests) not included in the above criteria if there is a perceived risk to participants.

In the event that definitive antimalarial treatment is initiated instead of dosing with the IMP, participants will be administered their first dose of antimalarial medication at the clinic (except in the event a participant tests positive to COVID-19; See Section 8.2.6.11 for details of COVID-19 management plan) but will then be able to return home. The participant will be contacted daily via phone to check that each dose of antimalarial treatment is being taken, and to ascertain any AEs. Following antimalarial treatment, blood will be taken for malaria PCR until at least one negative result is obtained. The participant will then be withdrawn from the study (see Section 7.2).

7.2 PARTICIPANT DISCONTINUATION/WITHDRAWAL FROM THE STUDY

Participants are free to withdraw from participation in the trial at any time upon request. Participants who decide to withdraw will be reminded of the importance of completing their full course of definitive antimalarial treatment. The Principal Investigator will demonstrate due diligence in following up with the participant to ensure antimalarial therapy has been completed successfully.

The Investigator may discontinue or withdraw a participant from the trial for the following reasons:

- Pregnancy.
- Significant trial intervention non-compliance.
- If any clinical AE, laboratory abnormality, or other medical condition or situation occurs (or is revealed after previously being unknown) such that continued participation in the trial would not be in the best interest of the participant.
- The participant is not dosed with the IMP (see Section 7.1).
- Investigator discretion.

The Principal Investigator will ensure all withdrawn participants successfully complete the full course of definitive antimalarial treatment.

Participants who decide to withdraw, or are withdrawn, will be asked to complete an early termination visit (procedures for this visit will be the same as for the EOS visit). Any ongoing AEs present at the time of withdrawal/discontinuation will be followed-up by the Principal Investigator as specified in Section 8.3.5.

The Sponsor will be informed immediately of any withdrawals. The reason for participant discontinuation or withdrawal from the trial will be recorded in the eCRF.

Participants who withdraw, or are withdrawn or discontinued from the trial, may be replaced after mutual agreement between the Sponsor and the Principal Investigator. The decision regarding the replacement of participants will be documented.

7.3 LOST TO FOLLOW-UP

If a participant does not attend a scheduled visit, the Investigator will apply due diligence by documenting all steps taken to contact the participant (e.g., dates of phone calls, registered letter, home visit, etc.) in the source documents.

It will be explained to the participants during the consenting process and throughout the trial that they must be readily contactable. The period from inoculation to definitive antimalarial treatment is the critical period for participant safety. A participant will be deemed "missing" if they fail to reply to communication to their personal mobile phone and nominated contact's number after 24 h. If after 36 h the participant fails to respond then the Investigator will organise a home visit. Subsequently, if the participant is still absent, the Investigator will request assistance from the local police to locate the missing participant. Once the participant is found, definitive antimalarial treatment will be administered.

A participant will be deemed lost to follow-up in the unlikely event that the participant is unable to be contacted or located despite all of the above measures. This is not anticipated to occur prior to administration of definitive antimalarial treatment, as in this situation, all efforts to locate the participant, including with assistance from local police, would continue until the participant is located and treated.

8 STUDY ASSESSMENTS AND PROCEDURES

The timing of each of the assessments described below is summarised in the SOA table (section 1.3) and the order of assessments at each visit is outlined in detail in the study conduct schedule (Appendix 12.1).

8.1 PHARMACOKINETIC AND PHARMACODYNAMIC ASSESSMENTS

8.1.1 PHARMACOKINETICS

Blood samples will be collected to quantify MMV367 plasma concentrations for PK analysis. Population PK modelling will be performed as part of the PK/PD analysis of MMV367 (primary endpoint). PK parameters of MMV367 will also be calculated using non-compartmental methods (secondary endpoint).

Samples will be analysed for MMV367 plasma concentrations using a validated analytical method in compliance with the SOPs of the laboratory performing the analyses. Assays will be performed in a Sponsor approved laboratory.

Plasma for MMV367 concentrations will be collected at the time points specified in the Schedule of Activities (Section 1.3). Allowed time windows for sample collection are as follows. The actual time of blood sampling will be recorded instead of the nominal time point.

Time point	Allowed time window
Inpatient observation pre-IMP (Day 8)	- 60 minutes
Inpatient observation ≤16 hours post-IMP (Day 8)	± 15 minutes
Inpatient observation ≥24 hours post-IMP until discharge (Days 9-11)	± 60 minutes
Outpatient monitoring (Day 12)	± 4 hours
Outpatient monitoring (Day 13)	± 12 hours
Outpatient monitoring Days 15-27)	± 24 hours

8.1.2 PHARMACODYNAMICS

Blood samples will be collected to monitor malaria parasitaemia using polymerase chain reaction (malaria PCR). Total malaria parasitaemia will be quantified throughout the study by quantitative PCR targeting the gene encoding *P. falciparum* 18S rRNA (malaria 18S qPCR; reported as parasites/500uL packed RBCs). After discharge from the clinical trial unit, parasite lifecycle stages will also be determined by reverse transcriptase qPCR (qRT-PCR). The qRT-PCR assays will target the female gametocyte-specific transcript *pfs25*, the male gametocyte-specific transcript *pfMGET*, and the ring-stage transcript *pfSBP-1*.

Malaria PCR data will be used for the PK/PD analysis of MMV367 (primary endpoint) and to determine the kinetics of parasite clearance following MMV367 dosing (secondary endpoint).

Malaria PCR will also be used to guide the timing of definitive antimalarial treatment initiation. The requirement for Primacin® administration to clear gametocytes will be informed by the parasite lifecycle stage data.

Samples for malaria PCR will be analysed using validated analytical methods in compliance with the SOPs of the laboratory performing the analyses. Assays will be performed in a Sponsor approved laboratory.

Blood samples for malaria PCR will be collected at the time points specified in the SOA (Section 1.3). Allowed time windows for sample collection are as follows. The actual time of blood sampling will be recorded instead of the nominal time point.

Time point	Allowed time window
Pre-malaria inoculation (Day 0)	- 4 hours
Outpatient monitoring pre-IMP (Days 4-6)	± 12 hours
Outpatient monitoring pre-IMP (Day 7 AM)	± 4 hours
Inpatient observation pre-IMP (Day 8)	- 60 minutes
Inpatient observation ≤24 hours post-IMP (Days 8-9)	± 30 minutes
Inpatient observation >24 hours post-IMP until discharge (Days 9-11)	± 60 minutes
Outpatient monitoring (Day 12)	± 4 hours
Outpatient monitoring (Day 13)	± 12 hours
Outpatient monitoring Days 15-27)	± 24 hours

Additional samples may be collected at the discretion of the Investigator based on previous malaria PCR results and clinical symptoms. Following definitive antimalarial treatment, no further blood collection for malaria PCR is required for a participant once a minimum of one negative qPCR result is recorded.

8.2 SAFETY, ELIGIBILITY AND OTHER ASSESSMENTS

All safety assessments may be conducted at unscheduled visits or time points if required for the participant's safety at the discretion of the Investigator.

The following time windows will be allowed for vital signs recording, ECGs and blood sampling for clinical laboratory evaluations:

Time point	Allowed time window
Pre-malaria inoculation (Day 0)	- 4 hours
Outpatient monitoring pre-IMP (Days 4-6)	± 12 hours
Outpatient monitoring pre-IMP (Day 7 AM)	± 4 hours
Inpatient observation pre-IMP (Day 7 PM)	± 4 hours
Inpatient observation pre-IMP (Day 8)	- 2 hours
Inpatient observation ≤24 hours post-IMP (Days 8-9)	± 30 minutes
Inpatient observation >24 hours post-IMP until discharge (Days 9-11)	± 60 minutes
Outpatient monitoring (Day 12)	± 4 hours
Outpatient monitoring (Day 13)	± 12 hours
Outpatient monitoring from (Days 15-27)	± 24 hours

8.2.1 BECK DEPRESSION INVENTORY

Originally described by Beck et al (1961), the Beck Depression Inventory is a validated objective tool for the assessment of depression. Updated in 1996, the Beck Depression Inventory II (BDI-II) is a 21-item, self-report rating inventory that measures characteristic attitudes and symptoms of depression, and participants will be required to complete the BDI-II at Screening for eligibility. The BDI-II takes approximately 10 minutes to complete, although participants require a fifth – sixth grade reading level to adequately understand the questions. A score of >20 at screening and/or a response of 1, 2 or 3 for item 9 indicating current suicidal ideation is exclusionary. A participant with a BDI-II score of 17 to 19 may be enrolled at the discretion the Investigator if they do not have a history of the psychiatric conditions mentioned in exclusion criterion 10-12 and their mental state is not considered to pose additional risk to the health of the participant or to the execution of the trial and interpretation of the data gathered.

8.2.2 PHYSICAL EXAMINATION

A **full physical examination** includes the following:

Weight
Height **(*Screening only*)**
Review of systems excluding genitourinary examination and including the following:
Head, neck (including thyroid), ears, eyes, nose and throat
Heart/circulation
Chest
Lungs
Abdomen
Skin
Neurological exam

A Full physical examination will be performed at Screening and at EOS.

An **abbreviated physical examination** will include heart/circulation, chest, lungs, skin, and abdomen. An abbreviated physical examination will be performed prior to inoculation with the malaria challenge agent and prior to administration of the IMP on Day 8.

When the **symptom-directed physical examination** is performed, body systems will be reviewed only if clinically indicated.

8.2.3 VITAL SIGNS

At screening, vital signs will be measured after the volunteer has rested in the supine position for 5 minutes. SBP and DBP will be measured again after the volunteer has transitioned to a standing position for 3 minutes to assess postural hypotension (see Section 5.2, exclusion criterion 10).

At all other time-points vital signs will be measured after the participant has rested in the seated position for at least 5 min.

The normal ranges for vital signs after administration of the malaria challenge agent are:

Parameter	Range
Systolic blood pressure	90-140 mmHg
Diastolic blood pressure	50-90 mmHg
Heart rate	50-100 bpm
Temperature	35.0-37.5°C
Respiratory rate	10-25 breaths/min

8.2.4 MALARIA CLINICAL SCORE

The malaria clinical score gives an indication of the severity of the induced malaria infection in each participant. Fourteen signs/symptoms frequently associated with malaria are graded using a 4-point scale (absent 0; mild: 1; moderate: 2; severe: 3) and summed to generate a total malaria clinical score (maximum score possible is 42). Severity will be graded in accordance with the CTCAE Version 5.0 Published: November 27, 2017. Mild (1) equates to CTCAE grade 1, Moderate (2) equates to CTCAE grade 2 and Severe (3) equates to CTCAE grade 3. The criteria are shown in the table below.

Symptom or sign	Clinical Score/CTCAE grade			
	Absent (0)	Mild (1)	Moderate (2)	Severe (3)
		CTCAE 1	CTCAE 2	CTCAE 3
Headache		Mild pain	Moderate pain; limiting instrumental activities of daily living (ADL)	Severe pain; limiting self-care ADL
Myalgia		Mild pain	Moderate pain; limiting instrumental ADL	Severe pain; limiting self-care ADL
Arthralgia		Mild pain	Moderate pain; limiting instrumental ADL	Severe pain; limiting self-care ADL

Symptom or sign	Clinical Score/CTCAE grade			
Fatigue		Fatigue relieved by rest	Fatigue not relieved by rest; limiting instrumental ADL	Fatigue not relieved by rest; limiting self-care ADL
Malaise		Uneasiness or lack of well-being	Uneasiness or lack of well-being; limiting instrumental ADL	Uneasiness or lack of well-being limiting self-care ADL
Chills		Mild sensation of cold; shivering; chattering of teeth	Moderate tremor of the entire body; narcotics indicated	Severe or prolonged, not responsive to narcotics
Sweating/hot spells		Mild sweating/hot spells not affecting ADL	Moderate sweating/hot spells; narcotics indicated	Severe or prolonged, not responsive to narcotics
Reduced appetite		Loss of appetite without alteration in eating habits	Oral intake altered without significant weight loss or malnutrition; oral nutritional supplements indicated	Associated with significant weight loss or malnutrition (e.g. inadequate oral caloric and/or fluid intake); tube feeding or TPN indicated
Nausea		Loss of appetite without alteration in eating habits	Oral intake decreased without significant weight loss, dehydration or malnutrition	Inadequate oral caloric or fluid intake; tube feeding, TPN, or hospitalisation indicated
Vomiting		Intervention not indicated	Outpatient IV hydration; medical intervention indicated	Tube feeding, TPN, or hospitalisation indicated
Abdominal discomfort		Mild pain	Moderate pain; limiting instrumental ADL	Severe pain; limiting self-care ADL
Fever		38.0-38.9°C	≥39.0-39.9°C	≥40.0°C
Tachycardia		HR ≥100 Asymptomatic, intervention not indicated	HR ≥100 Symptomatic; non-urgent medical intervention indicated	HR ≥100; Urgent medical intervention indicated
Hypotension		SBP ≤80 Asymptomatic, intervention not indicated	SBP ≤80 Symptomatic; non-urgent medical intervention indicated	SBP ≤80; Urgent medical intervention indicated
Total Score				
Maximum 3 x 14 = 42				

8.2.5 ELECTROCARDIOGRAPHS

A 12-lead ECG will be recorded after the participant has rested supine for at least 5 min. ECG tracings will be retained and labelled as per standard procedures at the clinical trial unit and will be recorded in the eCRF. Any clinically significant findings will be documented as AEs. The Investigator will sign and date each ECG as evidence of their review. ECGs will be recorded in triplicate at the screening and prior to inoculation with the malaria challenge agent on Day 0.

The normal ECG ranges after administration of the malaria challenge agent are:

Parameter	Range
PR interval	≤210 msec
QRS	50–120 msec
QTcF	Males: ≤450 msec Females: ≤470 msec

8.2.6 DIARY CARDS

Participants will be provided with diary cards after malaria inoculation on Day 0 and instructed to record symptoms, details of self-administered take home definitive antimalarial medication and concomitant medications used during the study. Participants will also be provided with thermometers to record their temperature in the event of symptoms of fever. The diary cards will be reviewed at each outpatient visit and any information will be entered in the eCRF.

8.2.7 LABORATORY TESTS

The required pathology tube types, sample volumes, and laboratory processing procedures will be described in the Laboratory Manual. Samples will be collected in accordance with clinical trial unit SOPs.

The Investigator will document the clinical significance of all results falling outside of the normal reference ranges. Changes from baseline (malaria inoculation and/or IMP dosing) will also be considered. Clinically significant abnormal results will be documented as AEs and followed-up as specified in Section 8.3.5.

Additional reflex testing may be conducted by the local laboratory (as per their SOPs) if safety laboratory values for a participant fall outside of the normal range/parameters. Unscheduled testing may be performed at Investigator's discretion.
The parameters that will be measured are listed below.

8.2.7.1 HAEMATOLOGY

White cell count (WCC)
Neutrophils (NEUT)
Lymphocytes (LYM)
Monocytes (MON)
Eosinophils (EOS)

Basophils (BAS)
Mean cell volume (MCV)
Red cell count (RCC)
Haemoglobin (HGB)
Haematocrit (HCT)
Platelet count (PLAT)
Red cell distribution width (RDW)
Reticulocyte count (RETI)
Blood Group and Rh(D) tests (***Screening only***)

8.2.7.2 BIOCHEMISTRY

Sodium (NA)
Potassium (K)
Chloride (CL)
Bicarbonate (BICARB)
Calcium (CA)
Corrected calcium (CCA)
Magnesium (MAG)
Glucose (GLUC)
Urea
Creatinine (CREAT)
Estimated glomerular filtration rate (eGFR) – CKD Epi calculation
Albumin (ALB)
Globulin
Total protein
Total bilirubin (BILI)
Direct (conjugated) bilirubin (BILDIR)
Alkaline phosphatase (ALP)
Alanine aminotransferase (ALT)
Aspartate aminotransferase (AST)
Gamma-glutamyl transferase (GGT)
Lactate dehydrogenase (LDH)
Phosphate (PHOS)
C-Reactive Protein (CRP) ***(not required at Screening)***
Cholesterol (Chol) (***Screening only***)
Triglycerides (Trig) (***Screening only***)
High density lipoprotein (HDL) (***Screening only***)
Low density lipoprotein (LDL) (***Screening only***)

8.2.7.3 VIRAL SCREEN

HIV 1/2 (anti-HIV1 and anti-HIV2 Ab)
Hepatitis B (HBsAg, anti-HBc [IgG + IgM if IgG is positive])
Hepatitis C (anti-HCV)

8.2.7.4 COAGULATION PROFILE

Prothrombin Time (PT)
Activated Partial Thromboplastin Time (APTT)
International Normalised Ratio (INR)

8.2.7.5 RED BLOOD CELL ALLOANTIBODIES

Screening for red blood cell alloantibodies will be performed at screening and at the EOS visit to investigate potential alloimmunization associated with administration of the malaria challenge agent.

8.2.7.6 GLUCOSE 6-PHOSPHATE DEHYDROGENASE (G6PD)

Quantitative G6PD testing performed at screening only for participant eligibility.

8.2.7.7 PREGNANCY TESTING

At screening, a serum β-hCG pregnancy test will be conducted for all female participants, and FSH levels will be tested in post-menopausal females (at least one year post-menopausal). At the eligibility confirmation visit, an additional serum β-hCG pregnancy test will be performed for WOCBP (WONCBP are defined in Section 5.1).

On Day 0 (pre-inoculation) and Day 7 PM, urine β-hCG pregnancy tests will be conducted for WOCBP. An additional serum β-hCG pregnancy test will also be conducted for WOCBP at the EOS visit.

If a participant is found to have a positive urine pregnancy test at any stage during the study, a confirmatory serum β-hCG will be performed. If confirmed positive, the participant will be administered definitive antimalarial treatment and will be discontinued from the study. Pregnancy-specific reporting procedures and follow-up are outlined in Section 8.3.9.

8.2.7.8 URINALYSIS

Urine will be tested by dipstick at the clinical trial unit. If there are any abnormalities (more than trace amounts for protein, blood, and leukocytes), the urine will be sent for formal laboratory urinalysis per the clinical trial unit standard procedure for microscopy, culture and sensitivity.

Glucose (GLUC)
Bilirubin (BILI)
Ketone (KETONES)
Specific gravity (SPGRAV)
Blood
pH
Protein (PROT)
Urobilinogen (UROBIL)
Nitrite
Leukocytes (WBC)
Microscopy, culture and sensitivity (***formal laboratory analysis if required***)

8.2.7.9 URINE DRUG SCREEN AND ALCOHOL BREATH TEST

The urine drug screen and alcohol breath test will be performed at clinical trial unit. If the result of a urine drug screen or alcohol breath test is positive, the participant will not be enrolled in the study (if testing positive on Day 0) or administered the IMP (if testing positive on Day 7 PM) unless there is an explanation acceptable to the Investigator (e.g., the participant has stated in advance that they consumed a prescription or over-the-counter product that contained the detected drug) and the participant has a negative urine drug screen on retest. The urine drug screen may be repeated if the participant denies usage of any of these agents and the test result is believed to be a false positive.

Urine drug screen:	
Amphetamines	Opiates
Methamphetamines	Phencyclidine
Barbiturates	Tetrahydrocannabinol (cannabis)
Benzodiazepines	Tricyclic antidepressants
Cocaine	
Methadone	
Alcohol breath test	

8.2.7.10 SAFETY SERUM RETENTION SAMPLE

Safety serum retention samples will be collected on Day 0 (prior to malaria challenge) and at EOS. Participants consent to the mandatory storage of these samples and their use for retrospective safety assessments if required. Samples will be stored indefinitely at QIMR Berghofer.

8.2.7.11 COVID-19 TESTING

A COVID-19 PCR test will be performed at the eligibility visit (Day -3 to Day -1) and a rapid antigen test (RAT) will be performed immediately prior to scheduled malaria inoculation (Day 0). Any participant testing positive will be ineligible for enrolment in the study.

An additional PCR test will be performed after malaria inoculation on Day 6, and an additional RAT will be performed upon admission to the clinical trial unit on Day 7 PM. Any participant testing positive will be administered definitive antimalarial medication and will be withdrawn from the study without being administered the IMP. If the participant tests positive while in clinic (i.e. the RAT on Day 7 PM), the first dose of definitive antimalarial medication will be administered at the clinic while remaining doses will be taken by the participant at home. If the result of the PCR test on Day 6 comes back positive, definitive antimalarial treatment will be delivered to the participants' home address, and a phone call will be made to ensure that the first dose is taken.

Participants will be contacted daily via phone to ensure treatment compliance for the remaining doses and to ascertain any adverse events. Where possible, an early termination visit (Section 12.1) will be conducted a minimum of 7 days following the positive COVID-19 test when the Investigator deems the risk of transmission is low.

Additional COVID-19 testing may be performed after IMP dosing at the discretion of the Investigator. If a participant tests positive during the confinement period (Day 7 PM to Day 11), definitive antimalarial treatment will be initiated and the participant will be sent home and followed-up via phone to ensure all doses are taken and ascertain any adverse events. Daily RATs will be performed for all remaining participants.

If a participant tests positive to COVID-19 during the outpatient follow-up period (Day 12 to Day 27), definitive antimalarial treatment will be initiated (either in clinic or delivered to the participants' home address as specified above).

Where possible, participants testing positive to COVID-19 after IMP dosing will resume study visits a minimum of 7 days after the date of the positive test.

Clinical management of participants with COVID-19 will follow Queensland Health Guidelines. If a participant's condition was to deteriorate while isolating at home, the participant will be assessed over the phone by the Investigator and may be advised to present to a hospital Emergency Department if required, or clinical site staff may call 000 if appropriate. In the event that a participant is admitted to hospital, the Principal Investigator will continue to liaise with treating clinicians.

8.2.7.12 PARASITE *EX VIVO* VIABILITY

Determining parasite growth in *ex vivo* cultures is an exploratory objective of this study. *Ex vivo* growth is a measure of parasite viability. While 18S qPCR quantifies all parasites (including those non-viable or dead in circulation), only viable parasites are able to replicate in *ex vivo* culture. Modelling of parasite growth observed *ex vivo* enables the fraction of viable parasites present in blood samples to be determined. The results of parasite *ex vivo* growth analyses will be supportive to malaria 18S qPCR data in determining the kinetics of parasite clearance following MMV367 dosing (secondary endpoint). Short term *ex vivo* cultures will be established from blood samples collected pre-MMV367 administration (Day 8), then at several time-points post-MMV367 administration.

8.2.7.13 PARASITE DRUG RESISTANCE

Blood samples will be collected from participants experiencing parasite regrowth to investigate if parasites have acquired resistance to MMV367 (exploratory objective). Parasites will be genotyped and *in vitro* drug sensitivity testing will be performed.

8.3 ADVERSE EVENTS AND SERIOUS ADVERSE EVENTS

8.3.1 DEFINITION OF ADVERSE EVENTS (AE)

An AE is any untoward medical occurrence, i.e., unfavourable and unintended sign (including an abnormal laboratory finding), symptom or disease that occurs in a participant during the course of the trial. An AE does not necessarily have a causal relationship with trial treatments or procedures.

AEs include, but are not limited to:

- A new symptom, sign or medical condition.
- A disease or medical condition detected or diagnosed during the course of the trial even though it may have been present prior to the start of the trial.
- An exacerbation of a pre-existing medical condition/disease.
- An increase in frequency or intensity of a pre-existing episodic disease or medical condition.
- Continuous persistent disease or symptoms present at trial start that worsen following the start of the trial.
- An abnormal assessment (e.g., change on physical examination, ECG finding) if it represents a clinically significant finding that was not present at trial start or worsened during the course of the trial.
- An abnormal laboratory test result if it represents a clinically significant finding (e.g., CTCAE grade 2 or above), symptomatic or not, which was not present at trial start or worsened during the course of the trial or led to dose reduction, interruption or permanent discontinuation of trial treatment.

Borderline abnormal laboratory findings and other objective assessments should NOT be routinely captured and reported as AEs. However, abnormal laboratory findings or other objective measurements that meet the following criteria should be captured and reported in the AE section of the eCRF:

- The result meets the criteria for reporting as an SAE.
- The test result is associated with accompanying symptoms, and/or
- It requires additional diagnostic testing or medical/surgical intervention, and/or
- It leads to a change in trial dosing, or discontinuation from the trial, significant additional concomitant drug treatment, or other therapy, and/or
- It is considered by the Investigator to be clinically significant or represent a clinically significant change from baseline.

Merely repeating an abnormal test, in the absence of any of the above conditions, does not constitute an AE. Any abnormal test result that is determined to be an error does not require reporting as an AE. Surgical procedures themselves are not AEs; they are therapeutic measures for conditions that may, or may not, be AEs. Malaria in itself is not considered an AE.

8.3.2 DEFINITION OF SERIOUS ADVERSE EVENTS (SAE)

An SAE is defined as an event that fulfils at least one of the following criteria:

- Results in death.
- Is life-threatening
  - The term "life-threatening" in the definition of "serious" refers to an event in which the participant was at immediate risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it was more severe.
- Requires inpatient hospitalisation or prolongs existing hospitalisation, unless this is for:
  - Elective or pre-planned treatment for a pre-existing condition that is unrelated to the trial and has not worsened since the start of the trial.
  - Cosmetic surgery, or for social reasons, or respite care in the absence of any deterioration in the participant's general condition.
- Results in persistent or significant disability/incapacity.
- Is a congenital abnormality/birth defect.
- Is considered medically important
  - Medical and scientific judgement should be exercised in deciding whether other AEs are to be considered serious, such as important medical events that may not be immediately life-threatening but may jeopardise the participant or may require intervention to prevent one of the other outcomes listed in the definition above. Examples of such events are: intensive treatment in an emergency room or at home for allergic bronchospasm; blood dyscrasias; convulsions that do not result in hospitalisation; development of drug dependency or drug abuse.

A **Suspected Unexpected Serious Adverse Reaction (SUSAR)** is any SAE where a causal relationship with a trial intervention (malaria challenge agent, MMV367, definitive antimalarial medication) is at least a reasonable possibility, and the event is not listed in the reference safety information (IB or Australian Prescribing Information for the definitive antimalarial medication).

8.3.3 ADVERSE EVENTS OF SPECIAL INTEREST (AESI)

An AE of special interest (AESI), whether serious or non-serious, is one of scientific and medical concern specific to the Sponsor's product or program, for which ongoing monitoring and rapid communication by the Investigator to the Sponsor and medical monitor could be appropriate. Such an event might require further investigation in order to characterise and understand it. Depending on the nature of the event, rapid communication by the trial Sponsor to other parties (e.g., regulators) might also be warranted (CIOMS VI, ICH E2F, 2010). Any abnormalities listed below should be reported as an AESI.

Hepatic:

- ALT or AST > 5× ULN
- ALP > 2 × ULN (in the absence of known bone pathology)
- Total bilirubin > 2× ULN (in the absence of known Gilbert syndrome)
- Any AST or ALT value > 2× ULN and total bilirubin > 1.5× ULN
- Any ALT or AST value > 2 × ULN and INR > 1.5
- Potential Hy's Law cases (defined as ALT or AST > 3 × ULN and total bilirubin > 2× ULN [mainly conjugated fraction] without notable increase in ALP to > 2× ULN)
- Any clinical event of jaundice (or equivalent term)
- ALT or AST > 2× ULN accompanied by (general) malaise, fatigue, abdominal pain, nausea, or vomiting, or rash with eosinophilia (eosinophil percent or count above the ULN).

Cardiac:

- QTcF at any time >480 msec
- Bundle branch block (except right bundle branch block that was present prior to MMV367 administration)
- Any arrhythmia, except:
  - sinus bradycardia that is clinically asymptomatic and not associated with any other relevant ECG abnormalities
  - sinus tachycardia that is clinically asymptomatic, is associated with a body temperature >38.0°C, and not associated with any other relevant ECG abnormalities
  - respiratory sinus arrhythmia
  - wandering atrial pacemaker, or
  - isolated single premature atrial/ventricular complex (i.e., no bigeminy, trigeminy, couplets, triplets or salvos) that does not occur more than once in a particular ECG tracing.

Haematological:

- Haemoglobin drop >20.0 g/L from baseline (at eligibility visit)
- Absolute neutrophil count (ANC) <0.5 × 10^9/L.
- Platelet count <75 × 10^9/L.

Dermatological: *†

Clinical signs of possible cutaneous adverse reactions such as:

- Dermatitis
- Rash including
  - erythematous
  - macular
  - papular
  - maculopapular
  - pruritic
  - pustular
  - vesicular

*If one of these cutaneous reactions is observed, pictures of the lesions should be obtained when feasible.
†Dermatological AEs need not be reported as AESIs if clearly unrelated to the malaria challenge agent or IMP (e.g. rash from cannula dressing or ECG dots).

All AESIs, including those that do not meet the definition of an SAE, must be reported as per section 8.3.7.

8.3.4 CLASSIFICATION OF AN ADVERSE EVENT

8.3.4.1 SEVERITY OF EVENT

In addition to determining whether an AE fulfils the criteria for an SAE or not, the severity of AEs experienced by participants will be recorded in accordance with the Common Terminology Criteria for Adverse Events (CTCAE) v5.0, published 27 November 2017.

The severity of AEs will be graded as follows should there be no CTCAE grading available for a specific AE:

- Grade 1: Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
- Grade 2: Moderate; minimal, local or non-invasive intervention indicated; limiting age-appropriate instrumental activities of daily living.
- Grade 3: Severe or medically significant but not immediately life-threatening; hospitalisation or prolongation of hospitalisation indicated; disabling; limiting self-care activities of daily living.
- Grade 4: Life-threatening consequences; urgent intervention indicated.
- Grade 5: Death related to AE.

A mild, moderate, or severe AE may or may not be serious. These terms are used to describe the intensity of a specific event. Medical judgement should be used on a case-by-case basis.

Seriousness, rather than severity assessment, determines the regulatory reporting obligations.

8.3.4.2 RELATIONSHIP TO STUDY INTERVENTION

The Principal Investigator or delegate must assess the relationship of each event to the malaria challenge agent, IMP (MMV367), the definitive antimalarial medications, and other trial procedures (separately) and decide whether, in their medical judgement, there is a reasonable possibility that the event may have

been caused by any of the trial interventions. Where possible, a distinction should be made between events considered related to the malaria challenge agent, MMV367, and the definitive antimalarial medications.

If there is no valid reason for suggesting a relationship, then the AE should be classified as “not related”. Alternatively, if there is any valid reason for suspecting a cause-and-effect relationship between the study intervention and the occurrence of the AE (even if undetermined or untested), then the AE should be considered as “related” to whichever product is relevant. This should be documented in the participant’s clinical file (source data) and eCRF.

The following may guide this assessment:

Related/suspected:

The temporal relationship between the event and the study intervention is compelling and/or follows a known or suspected response pattern to that product, and the event cannot be explained by the participant’s medical condition, other therapies or accident.

Not related/not suspected:

The event can be readily explained by other factors such as the participant’s underlying medical condition, concomitant therapy or accident and no plausible temporal or biologic relationship exists between the study intervention and the event.

In addition to the assessments of relationship to the trial interventions, the Investigator should comment on the AE record in the eCRF whether an AE is related to the trial participation of the participant (trial procedures etc.).

Of note, a sign or symptom associated with malaria infection that is of expected intensity, frequency and duration for the individual participant in the context of this trial is considered to be a malaria challenge agent-related event.

8.3.5 TIME PERIOD AND FREQUENCY FOR EVENT ASSESSMENT AND FOLLOW-UP

All AEs must be documented and followed up during the study by the Investigator until the event is resolved.
Events that are unresolved at the time of the participant’s last follow-up visit should continue to be followed up by the Investigator until no further medically relevant information in relation to the event can be expected and the Investigator considers it justifiable to terminate the follow-up. The participant will be referred to a general practitioner or medical specialist as appropriate. The Sponsor retains the right to request additional information for any participant with ongoing AE(s)/SAE(s) at the end of the trial, if judged necessary.

All AEs should be treated appropriately. The Investigator will decide upon the appropriate action to be taken in response to an AE, which may include one or more of the following:

- no action taken (i.e., further observation only), or
- administration of a concomitant medication, or
- hospitalisation or prolongation of current hospitalisation (event to be reported as an SAE), or
- other.

8.3.6 ADVERSE EVENT REPORTING

It is the Investigator's responsibility to document and report all AEs occurring in the clinical trial whether spontaneously reported by the participant, observed by the Investigator (either directly or by laboratory or other assessments), or elicited by general questioning. The period of observation for collection of AEs extends from the time of inoculation with the malaria challenge agent to EOS. These AEs must be recorded on specific AE pages of the eCRF. Events reported prior to this will be recorded as medical history, unless the symptoms worsen during the trial.

The following information should be recorded for all AEs:

- Description
- Dates and times of onset and resolution
- Seriousness (SAE or not)
- AESI (yes/no)
- Severity
  - If severity of an AE changes, only one AE will be reported, with the highest severity recorded in the eCRF and listings and tables. In the clinical file (source data), the description of the AE will report the various severities observed over time.
  - If the AE resolves and then reoccurs, then two AEs will be reported.
- Action taken in response to the AE (including treatment given).
- Outcome.
- Relationship to the trial treatments or procedures (causality assessment), including the IMP, malaria challenge agent, definitive antimalarial medications, or any other treatment or procedure conducted during the trial.

8.3.7 SAE AND AESI REPORTING

The Investigator will take immediate appropriate action in response to SAEs and AESIs to ensure participant safety and in an attempt to identify the cause of the event. The Investigator must notify the SAE or AESI to the pharmacovigilance provider (Prime Vigilance) by email within 24 hours of becoming aware of the event.

All reports must be signed by the Principal Investigator or delegate and notified to Prime Vigilance preferably by email or fax to:

Email: MMV@primevigilance.com

Back-up fax number: +44 800 471 5694

Prime Vigilance Contact:

Head Office: +44 1483 307920

Any copies of participant's medical records provided for SAE or AESI reporting must have all participant identifiers redacted before submission.

The report forms will always be completed as thoroughly as possible with all available details of the event and signed by the Principal Investigator or delegate. If the Principal Investigator or delegate does not have all information regarding an event, they will not wait to receive additional information before reporting the event. A follow-up SAE or AESI report should be completed within 14 days, or if there is no new information the report form should be updated when additional information is received.

The Principal Investigator or delegate will always provide an assessment of causality at the time of the initial report.

Email transmission of the report forms is the preferred method to transmit this information to Prime Vigilance. In rare circumstances notification by telephone is acceptable, with a copy of the report form sent by overnight mail.

The Principal Investigator or delegate, or responsible person according to local requirements, will comply with the applicable local regulatory requirements related to the reporting of SAEs to regulatory authorities and the HREC. Any SAE that meets the criteria of a SUSAR will be reported to the TGA by the Sponsor in accordance with the Sponsor's reporting procedures.

8.3.8 REPORTING EVENTS TO PARTICIPANTS

Not applicable.

8.3.9 REPORTING OF PREGNANCY

Pregnancy itself is not defined as an AE/SAE. Any complication or termination of pregnancy for medical reasons are to be reported as an AE/SAE. Spontaneous abortion, still birth or congenital anomaly must be reported as an SAE. Any WOCBP enrolled in the study who becomes pregnant during the study and the following 34 days after MMV367 administration should be followed through delivery or termination of the pregnancy. This has been calculated based on 30 days (one female menstrual cycle) plus 5 half-lives of the IMP. Five half-lives have been calculated as 4 days.

The Investigator will collect pregnancy information and report to Prime Vigilance within 24 hours of becoming aware of a participant's pregnancy using the pregnancy form and following the same process as described in section 8.3.5 for SAEs. Pregnancy follow-up should be recorded on the same form including pregnancy outcomes.

Once pregnancy is confirmed, pregnant female participants will be immediately withdrawn from the study and will be managed as per section 7.2. Where possible, the Investigator will also attempt to collect and report information regarding pregnancy outcomes of female partners of any male participants who were administered MMV367 in this study and the following 94 days after MMV367 administration. This has

been calculated based on 90 days (one cycle of spermatogenesis) plus 5 half-lives of the IMP. Five half-lives have been calculated as 4 days. Appropriate signed informed consent will be required directly from the pregnant female partner to obtain and report this information. Any participant's female partner who becomes pregnant during the study should be followed through delivery or termination of the pregnancy.

9 STATISTICAL CONSIDERATIONS

The following sections describe the statistical analysis as it is foreseen when the study is being planned. A detailed Statistical Analysis Plan (SAP) and Modelling Analysis Plan (MAP) will be finalised and approved prior to study conduct. The SAP and MAP will provide details of all analyses to be performed as well as the format of listings and tables to be provided for completion of the Clinical Study Report (CSR). Any deviations from the SAP will be described and justified in the final CSR.

9.1 STATISTICAL HYPOTHESES

The primary objective of this study is to characterise the PK/PD relationship of MMV367 in healthy participants experimentally infected with blood stage *P. falciparum*. There is no pre-specified statistical hypothesis for testing this objective.

9.2 SAMPLE SIZE DETERMINATION

A maximum sample size of up to 18 participants is expected to be sufficient to achieve the primary endpoint of the study, which is to determine the PK/PD parameters of MMV367. This sample size is not based on formal statistical calculations but rather is based on previous experience of characterising the PK/PD relationship of antimalarials using the IBSM model [3, 11]. The specific number of participants required will be dependent on the emerging results during the study; it is possible that less than 18 participants will be required. The SDRT will review the data after the completion of each cohort and decide when sufficient data has been obtained to complete the primary endpoint. No more than 18 participants will be administered IMP in this study.

9.3 POPULATIONS FOR ANALYSES

At minimum, two populations will be used for data analyses, the Full Analysis Set (FAS) and the IMP Set. The FAS will consist of all enrolled participants (i.e. those inoculated with the malaria challenge agent on Day 0). The IMP set will consist of all enrolled participants who receive the IMP on Day 8.

The FAS will be used to assess all participant disposition, baseline, demographic and protocol deviation data. Safety data may be assessed using both the FAS and IMP sets, as specified in the SAP.

Additional populations for analysis of PK, PD and PK/PD parameters will be defined in the SAP.

The number of participants in each analysis set will be summarised, with a corresponding listing.

9.4 STATISTICAL ANALYSES

9.4.1 GENERAL APPROACH

Continuous variables will be summarised with the number of observations (non-missing values), mean, standard deviation (SD), median, quartiles, minimum and maximum. The minimum and maximum values will be presented to the same number of decimal places as recorded in the raw data. The mean, median and SD will be presented to one more decimal place than the raw data. Categorical variables will be summarised with the number of observations and the numbers and percent from each category. Percentages will be rounded to one decimal place, with the denominator being the number of participants in the relevant population with non-missing data. No missing data will be imputed. If required, confidence intervals (CIs) will be two sided and will use 95% confidence levels. Any analyses requiring significance testing will use a two-sided test at the 5% significance level.

The inoculation baseline will be defined as the last available assessment prior to administration of the malaria challenge agent. The IMP baseline will be defined as the last available assessment prior to the administration of the IMP. Unscheduled visits will be excluded from summary tables. Assessments conducted at Early Termination will be excluded from summary tables.

9.4.2 ANALYSIS OF THE PRIMARY ENDPOINT

The primary endpoint is the PK/PD relationship of MMV367. The basic PK/PD relationship will link the parasite clearance rate to the MMV367 blood concentration with a sigmoidal E_{max} model characterized by the E_{max}, EC_{50} and Hill coefficient parameters. If needed, alternative models will be applied, for instance to account for a lag in the drug response, which will contain additional parameters. The parameters will be estimated using a non-linear mixed-effect modelling of MMV367 concentration-time and parasitaemia-time profiles. First, a population PK model will be constructed with the measured concentrations; it will allow the individual PK parameters obtained and the full PK profile for each individual to be calculated. Second, the PD parameters will be estimated with the measured parasitaemia and the predicted individual PK profiles.

As a general concept the changes in living parasites in blood P are modelled as the effect of a net exponential growth rate GR and clearance rate $Kill$ due to MMV367 with an initial parasitaemia P_{base} at time t_0 (time of first observation). The equations are expressed in the log-scale such as:

$$\begin{aligned} \frac{dPL}{dt} &= GR - Kill \\ PL(t_0) &= PL_{base} \end{aligned}$$

where $PL = \ln(P)$ is the log-transformed parasite counts.

The effect of concentrations on parasite killing/clearance will be described as:

$$Kill = E_{max} \cdot \frac{C_c^{Hill}}{C_c^{Hill} + EC_{50}^{Hill}}$$

where C_c is the concentration in the central compartment, E_{max} is the maximum effect of the drug, EC_{50} is the concentration that results in 50% of the maximum effect and $Hill$ is the Hill coefficient.

Alternative models may be tested if the exploration of the data indicate a different behaviour, e.g. if a lag time is observed before the parasites are cleared, an effect compartment model will be tested.

Other key parameters such as the minimum inhibitory concentration (MIC), the minimal parasiticidal concentration (MPC_{90}), and the parasite reduction rate in 48 h ($log_{10}PRR_{48}$) will be derived from the PD model. The MIC is defined as the concentration at which parasite clearance by the drug equals the parasite growth, i.e., the MMV367 concentration at which the minimum parasite concentration is observed. It is calculated with the following equation in case of the E_{max} sigmoid model.

$$MIC = EC_{50}\left(\frac{GR}{E_{max} + GR}\right)^{1/Hill}$$

The MPC_{90} is defined as the concentration at which the clearance effect is at 90% of the maximum. It is calculated as follows:

$$MPC_{90} = EC_{50} \cdot 9^{1/Hill}$$

The PRR_{48} is defined as the parasite clearance achieved within 48 hours, usually given as the reduction of values on log_{10} transformed scale. The maximum capacity of parasite clearance in 48 hours is determined as follows assuming that concentrations are maintained well above the MPC_{90} for this time span.

$$PRR_{48} = 48h\frac{E_{max} - GR}{\ln(10)}$$

Full details of the modelling analysis will be described in the MAP.

PK and PD parameters will be tabulated as population estimates and relative standard error (RSE). The inter-individual variability (IIV) of PK parameters will be estimated. If the RSE of the PK parameters IIV is larger than that estimated from the FIH study, the FIH study will be used to help inform the IIV of the PK parameters. The IIV of PD parameters will be fixed to plausible values.

9.4.3 ANALYSIS OF THE SECONDARY ENDPOINTS

This study has four secondary endpoints.

- The incidence, severity and relationship to MMV367 of adverse events as determined by self-reported symptoms, clinical laboratory analysis, vital signs, physical examinations and ECG assessments.

- The incidence, severity and relationship to the malaria challenge agent of adverse events as determined by self-reported symptoms, clinical laboratory analysis, vital signs, physical examinations and ECG assessments. Additionally, the severity of the induced malaria infection in each participant graded by the malaria clinical score.
- The pharmacokinetics of MMV367 calculated using non-compartmental methods (C_{max}, t_{max}, AUC_{last}, AUC_{inf}, CL/F, Vz/F, λ, $t_{1/2}$).
- The parasite clearance kinetics following dosing with MMV367 (PRR_{48} and $PCt_{1/2}$).

Safety parameters:
The safety and tolerability of MMV367 and the malaria challenge agent will be assessed by clinical review of the following parameters:

- AEs (including SAEs and AESIs)
- Vital signs
- 12-lead ECGs
- Clinical laboratory analysis
- Physical examination

In addition, the malaria clinical score will be used to assess the severity of the induced malaria infection (see Section 8.2.4).

The analysis of this endpoint is discussed in Section 9.4.4 below.

Parasite clearance kinetics:
The parasite clearance kinetics following dosing with of MMV367 will be determined by calculating the parasite reduction ratio over a 48 hour period (PRR_{48}) and corresponding parasite clearance half-life ($PCt_{1/2}$).

The PRR_{48} will be estimated using the slope of the optimal fit of the log-linear relationship of the parasitaemia decay. The optimal fit will be derived using summarised replicate parasitaemia data, which have been cleaned by dealing with potential outliers, values below the limit of detection and non-detectable values. The optimal fit of the log-linear parasitaemia-by-time relationship is determined by using left and right censoring to systematically remove the potential lag phase and tail phase of the parasitaemia decay. The decay rate, estimated by the slope coefficient from the log-linear decay regression, will be calculated for each participant. The overall dose-specific PRR_{48} will be estimated with its 95% CI by calculating the weighted average slope estimate and corresponding standard error (SE) using an inverse-variance method. Only participants who have optimal regression models with appropriate fit contribute toward the dose-specific PRR_{48}. Details will be presented in the SAP.

The parasite clearance half-life ($PCt_{½}$) will be derived from the optimal decay rate. $PCt_{1/2}$ is a transformation of PRR_{48} into a per hour clearance rate. Details regarding the calculation of this parameter will be presented in the SAP.

PK parameters:

The following PK parameters will be determined using non-compartmental methods from plasma MMV367 concentration-time profiles: AUC_{last}, AUC_{inf}, C_{max}, t_{max}, $t_{1/2}$, CL/F, Vd/F and λ. When applicable, all PK variables (normally and log-normally distributed) will be summarized using arithmetic and geometric means, minimum, median, maximum, standard deviation (SD) and coefficient of variation (CV %) of arithmetic and geometric means. Summary tables showing the median and range for t_{max} and geometric mean and CV% for all other PK variables will be provided by dose.

Plasma concentration values below the lower limit of quantification will be set to zero. All mentioned parameters will be expressed as mean ± SD, unless otherwise stated. Details regarding the calculation of this parameter will be presented in the SAP.

9.4.4 SAFETY ANALYSES

All AE data will be listed for each participant, including severity, relationship to IMP (MMV367) and malaria challenge agent, relationship to other protocol-specific treatments, outcome and actions taken. In addition, listings of AEs leading to discontinuation, AESIs, SAEs and deaths, will be provided as applicable. Adverse events (AE) will be coded according to MedDRA and grouped by system organ class (SOC) and preferred term (PT).

Clinical laboratory parameters, physical examination results, vital signs, and 12-lead ECG results will be listed for all participants and visits. Summary tables for each scheduled visit, including observed values, change from each baseline and clinical significance of each abnormal value, will be presented.

Malaria clinical score data (total score and each sign/symptom individually) will be listed for all participants and visits.

9.4.5 BASELINE DESCRIPTIVE STATISTICS

Demographic data will be summarised by descriptive statistics and listed for all enrolled participants. This will include total number of observations, mean, standard deviation, and range for continuous variables for medians and number and percentages for categorical variables.

Participant disposition will be summarised and presented in a flow diagram. Trial completion, trial withdrawals, exclusions, and violations will be summarised and the reasons for withdrawal, exclusions, and violations will be listed.

Medical history, current medical conditions, prior and concomitant medications, results of laboratory screening tests, drug and alcohol screening tests, and any other relevant baseline information will be listed by participant and treatment group. Medical history will be coded using MedDRA and summarised by SOC and PT.

9.4.6 PLANNED INTERIM ANALYSES

This study has no formal interim analyses other than review of safety, tolerability, parasitaemia and PK data by the SDRT between cohorts.

9.4.7 SUB-GROUP ANALYSES

Not applicable.

9.4.8 TABULATION OF INDIVIDUAL PARTICIPANT DATA

All individual participant data will be listed by measure and time-point.

9.4.9 EXPLORATORY ANALYSES

No formal statistical analysis is planned for exploratory endpoints.

10 SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

10.1 REGULATORY, ETHICAL, AND STUDY OVERSIGHT CONSIDERATIONS

10.1.1 ETHICAL STANDARD

The trial will be conducted in accordance with the protocol approved by the HREC(s), the principles of the Declaration of Helsinki (Recommendations guiding Medical Doctors in Biomedical Research Involving Human Participants, Fortaleza, Brazil 2013), the NHMRC National Statement on Ethical Conduct in Human Research (2007, updated 2018) and the Integrated Addendum to ICH E6 (R1): Guideline for Good Clinical Practice E6 (R2) (November 2016) — with introductory comments of the Australian TGA.

The Principal Investigator will minimise any discomfort experienced by participants during the trial. The only invasive procedures will be the IV inoculation of the malaria challenge agent and the blood collection by cannulation/venepuncture. The total volume of blood drawn from each participant will not exceed 470 mL in any 30-day period during trial participation.

10.1.2 ETHICAL REVIEW

The protocol, Participant Information Sheets, and Informed Consent Forms will be reviewed by the approving HREC(s) and no trial activities will be initiated prior to approval. All amendments and addenda to the protocol and consent forms will similarly be submitted to the approving HREC(s) for approval prior to their implementation.

Changes to the final trial protocol can only be made with the prior consent of the Principal Investigator, the Sponsor and the approving HREC(s). All such changes must be attached to (or incorporated into) the

final protocol and communicated to all relevant members of the clinical trial unit staff and, if appropriate, to trial participants. All deviations from this trial protocol will be included in the trial master file and included in the CSR. The types of amendments are discussed below.

Administrative or minor changes

Administrative or minor changes include but are not limited to changes in trial staff or contact details (e.g., Sponsor instead of contract research organisation monitors). Amendments for administrative or minor changes may be suitable for executive review (expedited) by the approving HREC(s).

Substantial amendment

Significant changes require a substantial amendment. Significant changes include but are not limited to: new data affecting the safety of participants, change of the objectives/endpoints of the trial, eligibility criteria, dose regimen, trial assessments or procedures, treatment or trial duration with or without the need to modify the Participant Information Sheet and Informed Consent Form. Substantial amendments are to be approved by the HREC(s). The implementation of a substantial amendment can only occur after formal approval from the Sponsor, HREC(s), and Principal Investigator.

Urgent amendment

An urgent amendment might become necessary to preserve the safety of the participants included in the trial. The requirements for approval should in no way prevent any immediate action being taken by the Principal Investigator or the Sponsor in the best interests of the participants. Therefore, if deemed necessary, the Investigator can implement an immediate change to the protocol for safety reasons with notification to the medical monitor as soon as practicably possible. This means that, exceptionally, the implementation of urgent amendments will occur before submission to, and approval by, the HREC(s). In such cases, the Principal Investigator must notify the Sponsor within 24 hours. A related substantial amendment will be written and submitted to HREC(s) as soon as practicable but no later than 7 working days, together with a description of the steps that have already been taken in regard to implementation of this amendment.

HREC approval of future research

In the event that the Principal Investigator or the Sponsor want to perform testing on the samples that is not described in the protocol, additional approval will be sought from the approving HREC(s).

10.1.3 INFORMED CONSENT PROCESS

10.1.3.1 CONSENT/ASSENT AND OTHER INFORMATIONAL DOCUMENTS PROVIDED TO PARTICIPANTS

At the screening visit, each potential participant will be given the trial Participant Information Sheet and Informed Consent Form that describes in detail the trial interventions, trial procedures, and risks.

10.1.3.2 CONSENT PROCEDURES AND DOCUMENTATION

During the initial screening visit/recruitment, potential participants will read the Participant Information Sheet. The Investigator will explain the trial via the Participant Information Sheet and the candidate participants will be encouraged to ask questions. Potential participants will be informed that participation is voluntary and that they may withdraw from the trial at any time, without prejudice. Potential participants will have the opportunity to discuss the trial with their family or think about it prior to agreeing to participate.

Individuals willing to be considered for inclusion in the trial will sign and date the Informed Consent Form in the presence of the Investigator. Participants will be given a copy of their signed Informed Consent Form. The conduct of the informed consent process will be documented in the source document (including the date) and the form will be signed before the participant undergoes any trial-specific procedures; only once the participant has consented to the trial may trial-specific screening activities commence.

10.1.4 STUDY DISCONTINUATION AND CLOSURE

The Sponsor, Principal Investigator, approving HREC(s), and regulatory authorities independently reserve the right to discontinue the trial at any time for safety or other reasons. Circumstances that may warrant termination or suspension include, but are not limited to:

- determination of unexpected, significant, or unacceptable risk to participants
- insufficient compliance with protocol requirements
- data that are not sufficiently complete and/or evaluable
- determination that the primary endpoint has been met.

This will be done in consultation with the Sponsor where practical. In the event of premature trial termination or suspension, the above-mentioned parties will be notified in writing by the terminator/suspender stating the reasons for early termination or suspension (with the exception of the Sponsor's responsibility for notifying the regulatory authorities). After such a decision, the Sponsor and the Investigator will ensure that adequate consideration is given to the protection of the participants' interest and safety. The Investigator must review all participants as soon as practical and complete all required records.

10.1.5 CONFIDENTIALITY AND PRIVACY

Participants will be informed that their data will be held on file by the clinical trial unit and that these data may be viewed by staff of the clinical trial unit (including, where necessary, staff of the clinical trial unit other than the named Investigators), trial monitors, and regulatory authorities.

In the event of a notifiable disease being discovered during the trial, the appropriate Public Health authorities will be notified in accordance with the Queensland Public Health Regulation 2018.

Upon request, the Investigator(s)/institution(s) will permit direct access to source data and documents for trial-related monitoring, audits, HREC review, and regulatory inspection(s) by the Sponsor (or their appropriately qualified delegates) and regulatory authorities.

Participants will also be informed that a report of the trial will be submitted to the Sponsor, co-development partner organisation (GSK plc), regulatory authorities, and for publication, but that they will only be identified in such reports by their trial identification number, their sex and age. The Investigators will undertake to hold all personal information in confidence.

Participants will be informed that samples collected for the purposes described in the protocol will be sent to the Sponsor's nominated national or international laboratory for assessment.

10.1.6 FUTURE USE OF STORED SPECIMENS AND DATA

As part of the trial, safety serum retention samples will be stored indefinitely at QIMR Berghofer for retrospective safety assessments that may later be indicated. Participants consent to this storage and the use of the sample for safety assessments when they sign the Informed Consent Form for the trial.

Under a separate protocol and consenting process, blood samples may be collected for storage and use in future malaria research. The separate study will be under the local sponsorship of QIMR Berghofer and will involve a protocol and consent document approved by the QIMR Berghofer HREC. All samples will be stored at QIMR Berghofer in accordance with the laboratory SOPs. The Investigator will ensure that confidentiality will be maintained continuously in all future research that involves use of these samples. No genetic testing will be performed on the stored samples without obtaining consent from the participants. The stored samples will not be sold or used directly for production of any commercial product. There are no benefits to participants in the collection, storage, and subsequent research use of their samples. Reports about future research done with participant samples will NOT be kept in participant health records, but a participant's samples may be kept with the trial records or in other secure areas.

10.1.7 KEY ROLES AND STUDY GOVERNANCE

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Institutional Ethics Committee	QIMR Berghofer Medical Research Institute Human Research Ethics Committee (QIMR Berghofer HREC; EC00278) Locked Bag 2000, Royal Brisbane and Women's Hospital, Brisbane QLD 4029, Australia Tel: +61 (0)7 3362 0117
Biostatistician (parasitaemia)	Stacey Llewellyn QIMR Berghofer Medical Research Institute 300C Herston Rd Herston QLD 4006, Australia Tel: +61 (0)7 3845 3579 Email: Stacey.Llewellyn@qimrberghofer.edu.au
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Clinical Study Unit	USC Clinical Trials Unit <u>Moreton Bay</u> Health Hub Morayfield 19-31 Dickson Road Morayfield QLD 4506 Australia Tel: +61 (0)7 54563965 Email: ctchhm@usc.edu.au <u>South Bank</u> Building A2, SW1 Complex 52 Merivale Street South Brisbane QLD 4101 Tel: +61 (0)7 5409 8630 Email: ctcsouthbank@usc.edu.au

Laboratories	Clinical laboratory: Sullivan Nicolaides Pathology Central Laboratory (SNP) 24 Hurworth Street Bowen Hills, QLD 4006, Australia Tel: +61 (0)7 3377 8782 Malaria PCR: Jake Tickner Queensland Paediatric Infectious Diseases Laboratory (Q-PID), SASVRC, Level 8, Centre for Children's Health Research 62 Graham Street South Brisbane, QLD 4101, Australia Tel: +61 (0)7 3069 7462 Email: j.tickner@uq.edu.au IMP quantification: Dr. Christoph Siethoff CEO Swiss BioQuant AG, Kägenstrasse 18 4153 Reinach, Switzerland Tel: +41 (0) 617 1698 12 Email:Christoph.Siethoff@swissbioquant.com, mail@swissbioquant.com
Serious AE and AESI reporting	Prime Vigilance Tel: +44 (0) 800 471 5694 Email: mmv@primevigilance.com

10.1.8 SAFETY OVERSIGHT

The SDRT will be responsible for decisions related to the safety of participants and the continuation of the trial. The SDRT will meet to assess any event/s that trigger the discontinuation rules or as needed to provide a recommendation and findings to the approving HREC(s) and the Principal Investigator.

The SDRT will meet at the end of each cohort to review safety data (AEs, AESIs, SAEs, and laboratory data), parasitaemia data, and the available PK and PK/PD modelling data. Based on this review, the SDRT will decide on whether to progress to the next cohort and will decide on the doses of MMV367 to be administered.

The SDRT may also decide to modify the frequency and timing of blood sample collection for malaria PCR and/or MMV367 plasma concentration measurements as long as blood volume is not greater than 470 mL in any 30-day period.

The role and composition of the SDRT is outlined in the study specific SDRT Charter.

10.1.9 CLINICAL MONITORING

It will be the Sponsor's responsibility to ensure that the trial is monitored in accordance with the requirements of GCP. The conduct of the trial will be reviewed internally by the clinical trial unit in accordance with their SOPs and work instructions, and GCP guidelines. The trial will be monitored

according to the SOPs of the Sponsor and all serious breaches, suspected breaches and protocol deviations will be reported to the Sponsor. Serious breaches that impact participant safety and/or data integrity will also be reported to the approving HREC.

During the trial, trial monitor(s) (on behalf of the Sponsor) will visit the clinical trial unit regularly (or, as specified in the Site Management and Monitoring Plan, access remotely the clinical unit files) to check the completeness of participant records, accuracy of eCRF entries, adherence to the protocol and to GCP, the progress of enrolment, and to ensure that trial interventions are being stored, dispensed, and accounted for according to specifications. Key trial personnel will be required to be available to assist the monitor during these visits (or provide the Monitor with an access to the required documents, if the visit is performed remotely).

The Investigator will be required to give the monitor access to all relevant source documents to confirm their consistency with the eCRF entries. At a minimum, the Sponsor will require full verification for the presence of informed consent, adherence to the inclusion/exclusion criteria, and documentation of SAEs, AESI and AEs, and the recording of data that was used for all primary and safety variables. Additional checks of the consistency of the source data with the eCRFs will be performed according to the trial-specific monitoring plan.

10.1.10 QUALITY ASSURANCE AND QUALITY CONTROL

The clinical trial unit will perform internal quality management of trial conduct, data and biological specimen collection, documentation and completion.

Quality control procedures will be implemented beginning with the data entry system and data quality control checks that will be run on the database that will be generated. Any missing data or data anomalies will be communicated to the site for clarification and resolution.

The clinical trial unit will provide direct access to source data and documents, and reports for the purpose of monitoring and auditing by the Sponsor, and inspection by regulatory authorities.

Data management, including the development and management of a secure database, will be performed in accordance with regulatory requirements. The designated data management vendor will review the data entered into the eCRFs by clinical trial unit staff for completeness and accuracy. A formal querying process will be followed whereby the data management team will request the site personnel to clarify any apparent erroneous entries or inconsistencies and will request additional information from the site as required.

Medical history/current medical conditions and AEs will be coded using the Medical Dictionary for Regulatory Activities (MedDRA) terminology (version 20.1 or higher). Prior and concomitant medications will be coded using the WHO Drug Dictionary Enhanced (WHO Global; March 2014 or later).

After all data have been captured and reviewed, all queries have been resolved with the site, and any protocol deviations that were identified during the data management processes have been confirmed by the site, the database will be declared to be complete and accurate. The database will be locked and made

available for data analysis. Any changes to the database after that time may only be made, in consultation with the Sponsor and in accordance with documented database unlock and relock procedures.

10.1.11 DATA HANDLING AND RECORD KEEPING

10.1.11.1 DATA COLLECTION AND MANAGEMENT RESPONSIBILITIES

Participants will be assigned a unique identifier when participating in the study, and any participant datasets or records that are transferred to the Sponsor or Clinical Research Organization (CRO) will only contain this identifier. Any other identifiable information about the participant will not be transferred. The Principal Investigator will ensure procedures are in place to appropriately protect the confidentiality of the participant records and data, including adequate safe guards for digital/computer access. The participants will be informed that their personal study-related data will be used by the Sponsor and that their medical records may be examined by auditors and regulatory agencies.

Each participant will have a clinical file (source data) and eCRF (for protocol specific data) into which relevant data will be recorded. All recording of source data and documents will be done in RealTime, the clinical trial unit's eSource system. Pathology reports, ECGs and any other paper source will be uploaded to RealTime.

Guidelines for eCRF completion including correcting data and responding to data queries will be provided by the Sponsor or CRO. A log of names, signatures, and initials of all staff authorised to enter data into a participant's clinical file and eCRF will be kept. Corrections to source document (whether paper or electronic) must have an audit trail (i.e., must not obscure or delete the original entry, and the date/time of correction and identity of the person making the correction must be clearly indicated).

10.1.11.2 STUDY RECORDS RETENTION

In compliance with the ICH/GCP guidelines, the Investigator/Institution will maintain all eSource and all paper source documents that support the data collected from each participant, as well as all study documents as specified in ICH/GCP Section 8, Essential Documents for the Conduct of a Clinical Trial, and all study documents as specified by the applicable regulatory requirement(s). The Investigator/Institution will take measures to prevent accidental or premature destruction of these documents. Essential documents must be retained until at least 2 years after the last approval of a marketing application in an ICH region and until there are no pending or contemplated marketing applications in an ICH region or until at least 2 years have elapsed since the formal discontinuation of clinical development of the IMP. These documents will be retained for a longer period if required according to the applicable regulatory requirements or per agreement with the Sponsor. It is the responsibility of the Sponsor to inform the Investigator/Institution as to when these documents no longer need to be retained. If the responsible Investigator retires, relocates, or for any other reasons withdraws from his responsibility of keeping the study records, custody must be transferred to a person who will accept the responsibility. The Sponsor must be notified in writing of the name and address of the new custodian. Under no circumstance shall the Investigator relocate or dispose of any study documents without having obtained written approval

from the Sponsor. If it becomes necessary for the Sponsor or the appropriate regulatory authority to review any documentation related to the study, the Investigator must permit access to such reports.

10.1.12 PROTOCOL DEVIATIONS

Protocol deviation: any departure, change, and/or addition from the trial design or procedures defined in the protocol that has received approval by the competent authorities and favourable opinion from the approving HREC.

Suspected serious breach: a report that is judged by the reporter as a possible serious breach, but has yet to be formally confirmed as a serious breach by the Sponsor.

Serious breach: a breach of GCP or the protocol that is likely to affect to a significant degree: a) the safety or rights of a trial participant, and/or b) the reliability and robustness of the data generated in the clinical trial.

Reporting requirements:

- All protocol deviations, suspected serious breaches, and serious breaches will be documented in the trial master file and included in the CSR.
- All protocol deviations, suspected serious breaches, and serious breaches will be viewed by the Sponsor and Principal Investigator.
- All protocol deviations, suspected serious breaches and serious breaches will be reported and assessed at each SDRT meeting.
- All serious breaches will be reported by the clinical trial unit to the Sponsor and the approving HREC(s) as early as possible, but within 7 days.
- Protocol deviation logs will be submitted by the clinical trial unit to the Sponsor and approving HREC via inclusion with the annual report.

10.1.13 PUBLICATION AND DATA SHARING POLICY

The data management, statistical, and medical writing team appointed by the Sponsor will collaborate to provide a detailed CSR upon conclusion of the trial. The CSR will include appendices of all tables and listings generated during the analyses of data. The tables, figures and listings will be provided by the Sponsor. The Sponsor will undertake to ensure that all safety observations made during the conduct of the trial are documented in the CSR.

Please refer to the Master Services Agreement for information on publication.

The Principal Investigator shall have the right to publish the results of the research, subject to the sponsor's prior written consent, which shall not be unreasonably withheld or delayed. Following the receipt of such consent, the Principal Investigator shall submit a copy of the proposed publication to the sponsor who shall have 30 days in which to request amendments thereto which, to the extent that such proposed amendments are reasonable, the Principal Investigator shall be obliged to incorporate prior to such publication.

The sponsor undertakes that, prior to publication of any information, article, paper, report or other material concerning the research, it will submit a copy of such publication to the Principal Investigator who shall have 30 days in which to request amendments thereto which, to the extent that such proposed amendment are reasonable, the sponsor shall be obliged to incorporate prior to such publication.

Publication and reporting of results and outcomes of this trial will be accurate and honest, and undertaken with integrity and transparency. Publication of results will be volunteered to fair peer-review. Authorship will be given to all persons providing significant input into the conception, design, and execution or reporting of the research. No person who is an author, consistent with this definition, will be excluded as an author without his/her permission in writing. Authorship will be discussed between researchers prior to trial commencement (or as soon as possible thereafter) and reviewed whenever there are changes in participation. Acknowledgement will be given to collaborating institutions and hospitals and other individuals and organisations providing finance or facilities.

Neither the complete nor partial results of the study achieved under this protocol, nor any of the information provided by the Sponsor for the purposes of performing the study, will be published or passed on to any third party without the consent of the study Sponsor. Any Investigator involved with this study is obligated to provide the Sponsor with complete study results and all data derived from the study.

The Sponsor will ensure that the key design elements of this protocol are posted in a publicly accessible database such as Australian New Zealand Clinical Trials Registry or Clinicaltrials.gov. In addition, upon trial completion and finalisation of the CSR, the results of this trial will be submitted for publication in an open access journal and/or posted in a publicly accessible database of clinical trial results.

10.1.14 CONFLICT OF INTEREST POLICY

Potential conflicts of interest will be identified, assessed, managed and documented, with any action taken and reasons for taking action recorded during the ongoing risk management process.

10.2 LIABILITY/INDEMNITY/INSURANCE

The Local Sponsor will ensure sufficient insurance is available to enable it to indemnify and hold the Investigators and relevant staff as well as any hospital, institution, ethics committee or the like, harmless from any claims for damages for unexpected injuries, including death, that may be caused by the participant's participation in the trial but only to the extent that the claim is not caused by the fault or negligence of the participant(s) or Investigators. The Sponsor adheres to the guidelines of Medicines Australia for injury resulting from participation in a company sponsored trial, including the provision of 'No-fault clinical trial insurance'.

10.3 ABBREVIATIONS

λ	Elimination Rate Constant
AE	Adverse Event
AESI	Adverse Event of Special Interest
ALP	Alkaline Phosphatase
ALT	Alanine Aminotransferase
AST	Aspartate Aminotransferase
anti-HBc Ab	Anti-Hepatitis B Core Antibodies
anti-HCV Ab	Anti-Hepatitis C Virus Antibodies
anti-HIV1 Ab	Anti-Human Immunodeficiency Virus 1Antibodies
anti-HIV2 Ab	Anti-Human Immunodeficiency Virus 2 Antibodies
AUC	Area Under the Curve
AUC_{inf}	AUC to Infinite Time
AUC_{last}	AUC to Last Quantifiable Concentration
BDI-II	Beck Depression Inventory II
B-hCG	Beta Human Chorionic Gonadotropin
BMI	Body Mass Index
CI	Confidence Interval
C_{max}	Maximum Plasma Concentration
CL/F	Apparent Oral Clearance
CRO	Clinical Research Organisation
CSR	Clinical Study Report
CTCAE	Common Terminology Criteria for AEs
DBP	Diastolic Blood Pressure
eCRF	Electronic Case Report Form
ECG	Electrocardiographs
EC_{50}	Half Maximal Effective Concentration
EOS	End of Study
E_{max}	Maximum parasite killing rate
FBC	Full Blood Count
FSH	Follicle-Stimulating Hormone
G6PD	Glucose-6-Phosphate Dehydrogenase
GCP	Good Clinical Practice
HBs Ag	Hepatitis B Surface Antigen
HIV	Human Immunodeficiency Virus
HR	Heart Rate
HREC	Human Research Ethics Committee
IB	Investigator's Brochure
IBSM	Induced Blood Stage Malaria
ICH	International Conference on Harmonization
IMP	Investigational Medicinal Product
LFT	Liver Function Test
MAP	Modelling Analysis Plan
MCB	Master Cell Bank
MedDRA	Medical Dictionary for Regulatory Activities

MIC	Minimum Inhibitory Concentration
MMV	Medicines for Malaria Venture
MPC_{90}	Minimal Parasiticidal Concentration that achieves 90% of maximum effect
NHMRC	National Health and Medical Research Council
NOAEL	No Observed Adverse Effect Level
NTF	Note To File
PD	Pharmacodynamics
PICF	Patient Information Consent Form
PK	Pharmacokinetics
PK/PD	Pharmacokinetics/Pharmacodynamics
PRR	Parasite Reduction Ratio
PRR_{48}	Parasite Reduction Ratio over a 48 hour period
PT	Preferred Term
$Pt_{½}$	Parasite Clearance Half-Life
QIMR Berghofer	QIMR Berghofer Medical Research Institute
qPCR	Quantitative Polymerase Chain Reaction
qRT-PCR	Quantitative Reverse-Transcriptase Polymerase Chain Reaction
RAT	Rapid Antigen Test
RBCs	Red Blood Cells
RDW	Red Blood Cell Distribution Width
Rh	Rhesus Antibody
SAD	Single ascending dose
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SBP	Systolic Blood Pressure
SD	Standard Deviation
SDRT	Safety and Data Review Team
SE	Standard Error
SOA	Schedule of Activities
SOC	System Organ Class
SOP(s)	Standard Operating Procedure(s)
SUSAR	Suspected Unexpected Serious Adverse Reaction
$t_{½}$	Terminal Half-Life
TEAEs	Treatment-Emergent Adverse Events
TGA	Therapeutic Goods Administration
t_{max}	Time taken to reach C_{max}
ULN	Upper Limit of Normal
Vd/F	Apparent Volume Distribution
VIS	Volunteer Infection Study
WBC	White Blood Cell
WOCBP	Women of Childbearing Potential
WONCBP	Women of Non-Childbearing Potential
WHO	World Health Organization

10.4 PROTOCOL AMENDMENT HISTORY

Version	Date	Description of Change	Brief Rationale

11 REFERENCES

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3. McCarthy JS, Lotharius J, Ruckle T, Chalon S, Phillips MA, Elliott S, et al. Safety, tolerability, pharmacokinetics, and activity of the novel long-acting antimalarial DSM265: a two-part first-in-human phase 1a/1b randomised study. Lancet Infect Dis. 2017;17:626-35.

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10. Watts RE, Odedra A, Marquart L, Webb L, Abd-Rahman AN, Cascales L, et al. Safety and parasite clearance of artemisinin-resistant *Plasmodium falciparum* infection: A pilot and a randomised volunteer infection study in Australia. PLoS Med. 2020;17:e1003203.

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12. McCarthy JS, Abd-Rahman AN, Collins KA, Marquart L, Griffin P, Kummel A, et al. Defining the antimalarial activity of cipargamin in healthy volunteers experimentally infected with blood-stage *Plasmodium falciparum*. Antimicrob Agents Chemother. 2021;65: e01423-20.

13. Chughlay MF, Akakpo S, Odedra A, Csermak-Renner K, Djeriou E, Winnips C, et al. Liver enzyme elevations in *Plasmodium falciparum* volunteer infection studies: Findings and recommendations. Am J Trop Med Hyg. 2020;103:378-393.

12 APPENDICES

12.1 FULL STUDY CONDUCT SCHEDULE

12.1.1 SCREENING VISIT (DAY -28 TO DAY -1)

A screening visit will be scheduled after an initial phone interview conducted by clinical trial unit staff has occurred to review background information. For the screening visit, volunteers will be asked to come to the clinical trial unit after an overnight fast of ≥8 hours.

Volunteers will be advised of the possible requirement to repeat some screening tests during the Day -3 to Day -1 eligibility confirmation visit (unless screening occurs within eligibility Day -3 to -1 window) and/or on inoculation day to determine their continuing eligibility.

The screening will be conducted within 28 days prior to the inoculation day and will include all activities listed below:

- A screening number will be assigned to each participant (as per clinical trial unit standard format).
- Elicit a complete medical history and use of medications.
- Elicit a social history including recreational drug, alcohol and nicotine use.
- Elicit demographic data.
- Perform alcohol breath test.
- Perform urine drug screen.
- Perform full physical examination (record body weight and height).
- Ask volunteer to complete the Beck Depression Inventory.
- Assess the cardiovascular disease risk (as per the Australian Absolute Cardiovascular Disease Risk Calculator).
- Record vital signs (after 5 min in supine and then after 3 min in standing for blood pressure).
- Obtain a 12-lead ECG in triplicate.
- Collect urine for urinalysis.
- Collect blood samples for haematology and biochemistry (including lipids for cardiovascular risk factor), red cell antibodies, quantitative G6PD, viral screen, coagulation profile, serum β-hCG pregnancy test (for all females), and FSH test (for post-menopausal females).
- Verify participant meets inclusion/exclusion criteria.

12.1.2 ELIGIBILITY CONFIRMATION VISIT (DAY -3 TO DAY -1)

The participants (including reserve participants) will report to the clinical trial unit between Day -3 to Day -1 for the following baseline assessments, unless screening laboratory assessments were conducted within this period, in which case repeat sampling will not be required.

<u>This visit will include:</u>

- Collect blood samples for haematology and biochemistry.
- Perform serum β-hCG pregnancy test for WOCBP.

- Collect urine for urinalysis.
- Collect sample for COVID-19 PCR test.

The timing of these assessments is to ensure that results are available for review by the Investigator prior to inoculation on Day 0.

12.1.3 MALARIA CHALLENGE AGENT ADMINISTRATION (DAY 0)

Participants, including up to three reserve participants per inoculation day, will report to the clinical trial unit on the morning of the day of inoculation. The Investigator will review the participants' screening and eligibility confirmation visit results prior to their inoculation.

On arrival to the clinical trial unit, participants will be required to undertake further procedures to determine whether they remain eligible. A reserve participant may be asked to replace a participant who does not meet eligibility. The reserve participants will be compensated for the study visit even if not inoculated, as described in the Participant Information Sheet and Consent Form.

<u>The procedures that will be undertaken prior to inoculation include:</u>

- Verify that all applicable eligibility criteria have been met.
- Elicit information regarding any new medical conditions, illnesses and medication use since screening visit.
- Perform COVID-19 rapid test.
- Perform alcohol breath test.
- Perform urine drug screen.
- Perform urine β-hCG pregnancy test for WOCBP.
- Perform abbreviated physical examination.
- Record vital signs.
- Obtain a 12-lead ECG in triplicate.
- Obtain malaria clinical score.
- Cannulate participants with an indwelling intravenous cannula (20 or 22 gauge) for the malaria challenge agent, and record which arm is utilised.
- Collect blood samples for:
    - malaria PCR (baseline sample),
    - safety serum retention sample,
    - Future malaria research sample (if participant has consented).

<u>Administration of the malaria challenge agent:</u>

- Administer the malaria challenge agent of ~2,800 viable *P. falciparum* 3D7 infected human erythrocytes intravenously.
- Observe for a minimum of 60 minutes after inoculation to evaluate for immediate adverse reactions.
- Educate participants on signs and symptoms of malaria.

    a. Signs of malaria include fever (oral temperature above 38°C), chills/shivering/rigors, tachycardia, hypotension.

b. Symptoms of malaria include headache, myalgia, arthralgia, fatigue/lethargy, malaise, sweating/hot spells, loss of appetite, nausea, vomiting and abdominal discomfort.

- Provide participants with diary cards and thermometers to record any temperature readings during the study in the event of symptoms of fever. Participants will also record symptoms, details of self-administered take home definitive antimalarial medication and concomitant medications on the diary cards during the study.
- Record AEs and concomitant medications.
- Record vital signs prior to leaving the clinical trial unit (approximately 60 minutes after inoculation).

12.1.4 OUTPATIENT MONITORING PRE-IMP ADMINISTRATION (DAY 1 TO DAY 7AM)

Monitoring via phone (Days 1 to 3):
During this period, the participants are expected to be asymptomatic. A daily phone call or text message will be made to the participant by clinical trial unit staff to monitor participant well-being and to solicit any AEs and concomitant medications. Participants will be reminded to fill in their diary cards.

Daily visits to the clinical trial unit for monitoring (Days 4 to 7):
Follow-up from Day 4 to Day 7AM will be undertaken through daily morning visits to the clinical trial unit.

The following procedures will occur during these visits:

- Perform symptom-directed physical examination if clinically indicated.
- Record vital signs.
- Obtain malaria clinical score.
- Collect blood sample for malaria PCR.
- Collect sample for COVID-19 PCR test (Day 6AM).
- Collect blood samples for haematology and biochemistry (Day 7AM).
- Record AEs and use of concomitant medications.
- Check participant diary cards.

12.1.5 INPATIENT OBSERVATION AND IMP ADMINISTRATION (DAY 7PM TO DAY 11)

Participants will be admitted to the clinical trial unit on Day 7PM, in preparation for single-dose administration of MMV367 on Day 8 when parasitaemia for the majority of participants is expected to be above 5,000 parasites/mL. Participants will be provided with a light snack the evening prior to dosing and then fast from all food and drink (except water) for a minimum of 8 h on the day prior to dosing until approximately 4 h post-dose, at which time lunch will be provided.

The following procedures will occur on admission to the clinical trial unit Day 7PM:

- Alcohol breath test.
- Urine drug screen.
- Urine β-hCG pregnancy test for WOCBP.
- Perform COVID-19 rapid test.

The following procedures will occur pre MMV367 administration on Day 8:

- Perform abbreviated physical examination.
- Obtain and review a 12-lead ECG.
- Obtain malaria clinical score.
- Record vital signs.
- Record AEs and use of concomitant medications.
- Review haematology and biochemistry results from Day 7 am collection.
- Randomisation.
- Cannulate participants with an indwelling intravenous (18 or 20 gauge) cannula.
- Collect blood samples for
    - haematology and biochemistry,
    - malaria PCR,
    - parasite *ex vivo* viability,
    - IMP PK (baseline sample),
    - future malaria research sample (if participant consented).

Administer the relevant MMV367 dose under direct observation.

The following procedures will occur after MMV367 administration:

- Follow up participants as in-patients until Day 11 to ensure tolerance of MMV367 and adequate clinical response.
- Perform symptom-directed physical examination if clinically indicated.
- Record vital signs three times a day (at time points specified in Section 1.3) whilst confined.
- Obtain malaria clinical score three times a day (at time points specified in Section 1.3) whilst confined.
- Obtain 12-lead ECG once daily (at time points specified in Section 1.3) while confined.
- Collect blood for haematology and biochemistry at 24 hours post-MMV367 administration.
- Collect blood for malaria PCR following MMV367 administration at 2, 4, 8, 12, 16, 24, 30, 36, 48, 54 and 60 hours (see section 8.1 and 8.2 for time allowed time windows).
- Collect blood samples for IMP PK at 1, 2, 3, 4, 6, 8, 12, 16, 24, 36, 48 and 60 hours post-MMV367 administration (see section 8.1 and 8.2 for time allowed time windows).
- Collect blood for parasite *ex vivo* viability at 8, 16, 24, 36 and 48 hours post-MMV367 administration.
- Record AEs and use of concomitant medications.

The following procedures will occur prior to discharge from the clinical trial unit (Day 11AM):

- Perform symptom-directed physical examination if clinically indicated.
- Obtain a 12-lead ECG.
- Obtain malaria clinical score.
- Record vital signs.
- Collect blood samples at 72 hours post-MMV367 administration for:
    - haematology and biochemistry,
    - malaria PCR,

- IMP PK,
- parasite *ex vivo* viability.

- Record AEs and use of concomitant medications.

12.1.6 OUTPATIENT MONITORING POST-IMP ADMINISTRATION (DAYS 12 TO 27)

Follow up visits will be undertaken on an out-patient basis for clinical evaluation and blood sampling on the scheduled days described below. Follow up visits may occur more frequently if clinically indicated. Follow up visits on Days 17, 20 and 24 are not required if participant has previously been administered definitive antimalarial treatment and returned at least one negative malaria PCR result.

<u>The following procedures will take place on Day 12±4hrs:</u>

- Obtain malaria clinical score.
- Record vital signs.
- Record AEs and use of concomitant medications.
- Check participant diary cards.
- Perform symptom-directed physical examination if clinically indicated.
- Collect blood samples for:
  - malaria PCR,
  - IMP PK,
  - parasite *ex vivo* viability,
  - future malaria research sample (if participant consented).

<u>The following procedures will take place on Day 13±12hrs:</u>

- Obtain malaria clinical score.
- Record vital signs.
- Record AEs and use of concomitant medications.
- Check participant diary cards.
- Perform symptom-directed physical examination if clinically indicated.
- Collect blood samples for:
  - malaria PCR,
  - IMP PK.

<u>The following procedures will take place on Day 15±24hrs:</u>

- Obtain malaria clinical score.
- Record vital signs.
- Record AEs and use of concomitant medications.
- Check participant diary cards.
- Perform symptom-directed physical examination if clinically indicated.
- Collect blood samples for:
  - haematology and biochemistry,
  - malaria PCR,

- IMP PK,
- future malaria research sample (if participant consented).

The following procedures will take place on Day 17±24hrs:

- Obtain malaria clinical score.
- Record vital signs.
- Record AEs and use of concomitant medications.
- Check participant diary cards.
- Perform symptom-directed physical examination if clinically indicated.
- Collect blood samples for:
  - malaria PCR,
  - IMP PK.

The following procedures will take place on Day 20±24hrs:

- Obtain malaria clinical score.
- Record vital signs.
- Record AEs and use of concomitant medications.
- Check participant diary cards.
- Perform symptom-directed physical examination if clinically indicated.
- Collect blood samples for:
  - malaria PCR,
  - IMP PK.

The following procedures will take place on Day 22±24hrs:

- Obtain malaria clinical score.
- Record vital signs.
- Record AEs and use of concomitant medications.
- Check participant diary cards.
- Perform symptom-directed physical examination if clinically indicated.
- Collect blood samples for:
  - haematology and biochemistry,
  - malaria PCR.

12.1.7 DEFINITIVE ANTIMALARIAL TREATMENT

All participants will receive compulsory definitive antimalarial treatment with Riamet® and/or other registered antimalarials if required on Day 24±24hrs, or earlier in the following cases:

- Insufficient parasite clearance following IMP dosing (parasitaemia not reduced ≥10-fold by Day 10 compared with peak parasitaemia on Day 8).
- Parasite regrowth following IMP dosing (initial parasite clearance is followed by asexual parasite regrowth above 5000 parasites/mL).

- Participant discontinuation/withdrawal (see Section 7).
- Investigator's discretion in the interest of participant safety.

The following procedures will be performed prior to antimalarial rescue treatment:

- Obtain malaria clinical score.
- Record vital signs.
- Record AEs and use of concomitant medications.
- Check participant diary cards.
- Perform symptom-directed physical examination if clinically indicated.
- Collect blood samples for:
  - malaria PCR,
  - parasite *ex vivo* viability (only in the event of parasite regrowth),
  - parasite drug resistance (only in the event of parasite regrowth).

12.1.8 END OF STUDY VISIT (DAY 27±24HRS) OR EARLY TERMINATION

The following procedures will be performed at the EOS visit:

- Perform a full physical examination (record body weight).
- Obtain a 12-lead ECG.
- Obtain malaria clinical score.
- Record vital signs.
- Record AEs and use of concomitant medications.
- Collect urine for urinalysis.
- Collect blood samples for:
  - haematology and biochemistry,
  - serum β-hCG pregnancy test for WOCBP,
  - RBC alloantibodies,
  - viral screen,
  - malaria PCR (not required if one negative result previously returned post-definitive antimalarial treatment),
  - safety serum retention sample,
  - future malaria research sample (if participant consented).
- Collect diary card.

If a participant is unwell with detectable parasitaemia or at the Investigator's discretion, the participant may be asked to return to the clinical trial unit after the EOS visit for safety follow up.

12.2 STANDARD INCLUSION RANGES FOR CLINICAL LABORATORY PARAMETERS

Test	Unit	Low	High
Biochemistry			
NA	mmol/L	130	150
K	mmol/L	3.0	5.5
CCA	mmol/L	0.9 x LLN	1.1 x ULN
Urea	mmol/L	N/A	1.75 x ULN
CREAT	umol/L	N/A	1.0 x ULN
eGFR	mL/min/1.7 3m^2	60	N/A
GLUC	mmol/L	N/A	1.0 x ULN
BILI	umol/L	N/A	1.25 x ULN
ALP	U/L	N/A	1.5 x ULN
AST	U/L	N/A	1.1 x ULN
ALT	U/L	N/A	1.1 x ULN
GGT	U/L	N/A	1.5 x ULN
HDL	mmol/L	0.9x LLN	N/A
LDL	mmol/L	N/A	2.0 x ULN
Haematology			
HGB	g/L	0.9x LLN	1.1.x ULN
PLAT	x10^9/L	0.9x LLN	1.1 x ULN
WCC	x10^9/L	0.9x LLN	1.1 x ULN
NEUT	x10^9/L	1.0 x LLN	1.1 x ULN
LYM	x10^9/L	1.0 x LLN	1.1 x ULN
Urinalysis			
PROT	N/A	N/A	Trace
KETONES	N/A	N/A	Trace
GLUC	N/A	N/A	Trace
Red Blood Cells	×10^6/L	N/A	20 (females)* 10 (males) *A result >20 is acceptable for females currently menstruating.
White Blood Cells	×10^6/L	N/A	10
Casts	N/A	N/A	Occasional