TITLE PAGE

Protocol Title: A single centre, open-label, parallel-group, single oral dose study to evaluate the pharmacokinetics, safety and tolerability of Pyrimethamine in healthy Japanese and Caucasian male subjects

Protocol Number: 204678

Short Title: Phase I study of Pyrimethamine in healthy Japanese and Caucasian Subjects

Compound Number: GR99352

Sponsor Name and Legal Registered Address:

GlaxoSmithKline Research & Development Limited
980 Great West Road
Brentford
Middlesex, TW8 9GS
UK

Medical Monitor Name and Contact Information:

PPD
GlaxoSmithKline K.K.
6-15, Sendagaya 4-chome Shibuya-ku,
Tokyo 151-8566
Japan

Regulatory Agency Identifying Number(s): N/A

Approval Date: 19-JULY-2017

SPONSOR SIGNATORY:

PPD

2017. 7. 19.

Kihito Takahashi
Vice President and Senior Managing Director
Development and Medical Affairs Division
GlaxoSmithKline K.K.

Date

The IMMS document approved is as follows:
Unique ID : PPD
System Version : 1.0
Effective Date : 2017.7.19

PPD

TABLE OF CONTENTS

PAGE

1. SYNOPSIS

Protocol Title: A single centre, open-label, parallel-group, single oral dose study to evaluate the pharmacokinetics, safety and tolerability of Pyrimethamine in healthy Japanese and Caucasian male subjects

Short Title: Phase I study of Pyrimethamine in healthy Japanese and Caucasian Subjects

Rationale: Pyrimethamine in combination with a sulphonamide is effective in the treatment of toxoplasmosis. However, in Japan, Pyrimethamine has not been approved by the Japanese regulatory body (Pharmaceutical and Medical Devices Agency [PMDA]/ Ministry of Health, Labour and Welfare [MHLW]), and the use of Pyrimethamine needs to rely on obtaining it via personal import or provided by a grant-in-aid from the Japanese government. The development of Pyrimethamine was requested by the Japanese Ministry of Health, Labor and Welfare (MHLW) because a high medical need exists for the treatment of toxoplasmosis with Pyrimethamine in Japan.

The pharmacokinetics (PK) of Pyrimethamine has been investigated following administration of Sulfadoxine/Pyrimethamine tablet (Fansidar®) in healthy Japanese subjects [Otomo H, et al. 1985]. However, as this study did not provide sufficient information for approval of Pyrimethamine in Japan, the PMDA has requested confirmation of the PK of Pyrimethamine in another PK study in Japanese and Caucasian healthy subjects. The primary adverse reaction of Pyrimethamine is abnormalities of the blood and lymphatic system, such as anaemia, leucopenia and thrombocytopenia. Therefore, for all subjects receiving Pyrimethamine, coadministration of calcium folinate is recommended to reduce the risk of bone marrow depression.

Thus, the primary objective of this study is to investigate the PK following a single oral dose of Pyrimethamine 50 mg in a fasted condition in healthy Japanese and Caucasian male subjects.

Objectives and Endpoints:

Objective	Endpoint
Primary	
• To investigate PK following a single oral dose of Pyrimethamine 50 mg in fasted healthy Japanese male subjects.	• Cmax, AUC(0-t), AUC(0-inf), AUC(0-24), tmax, t1/2, CL/F, Vd/F from the plasma concentrations of Pyrimethamine in healthy Japanese male subjects, as data permit.
Secondary	
• To investigate PK following a single oral dose of Pyrimethamine 50 mg in fasted healthy Caucasian male subjects.	• Cmax, AUC(0-t), AUC(0-inf), AUC(0-24), tmax, t1/2, CL/F, Vd/F from the plasma concentrations of Pyrimethamine in healthy Caucasian male subjects, as data permit.

• To assess safety and tolerability following a single oral dose of Pyrimethamine in fasted healthy Japanese and Caucasian male subjects.	• AE and change from the baseline of clinical laboratory values, vital signs and ECG.

Overall Design:

This study will be a single centre, open-label, parallel-group, single oral dose study to evaluate the PK, safety and tolerability of Pyrimethamine in healthy Japanese and Caucasian male subjects.

Number of Participants:

Sufficient Japanese and Caucasian healthy male subjects will be enrolled such 6 subjects of each population complete dosing and critical assessments.

If participants prematurely discontinue the study, additional replacement participants may be recruited and assigned to the same treatment at the discretion of the Sponsor in consultation with the investigator.

Treatment Groups and Duration:

Subjects will have a screening visit within 30 days prior to the first dose of study treatment. Subjects will be housed in the Clinical Research Unit from Day -1 through Day 8, and return to the unit on Day 15 (±1) and 22 (±1) for the remaining PK samples and safety assessments post dose.

During the treatment period, subjects will receive a single oral dose of Pyrimethamine 50 mg in the fasted state after an overnight fast (at least 10 hours). Oral calcium folinate 15 mg will be coadministered with Pyrimethamine on Day 1, and will continue to be administered once a day until Day 8.

Blood sampling for PK analysis and safety assessments will be performed prior to dosing and over 22 days after dosing. The duration of each subject's participation will be approximately 2 months from screening to the follow-up.

2. SCHEDULE OF ACTIVITIES (SOA)

	Scr	Day -1	Day 1							Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 15 [4]	Follow-up Day 22 [4]
			Pre	0 h	1 h	2 h	4 h	6 h	12 h	24 h	48 h	72 h	96 h	120 h	144 h	168 h	336 h	504 h
Admission to Unit		X																
Informed consent	X																	
Subject demography/ Medical history	X																	
Physical examination	X	X								X	X	X	X	X	X	X		X
12-lead ECG	X		X				X		X	X	X							X
Vital signs	X		X				X		X	X	X	X	X	X	X	X	X	X
Urine drug screen	X	X																
Alcohol breath test	X	X																
CO breath test	X	X																
Serological test	X																	
Haematology, clinical chemistry and urinalysis	X	X								X			X			X	X	X
Study treatment dosing				X														
Calcium folinate dosing				X [1]						X	X	X	X	X	X	X		
PK Sampling			X		X	X	X	X	X	X	X	X		X		X	X	X
SAEs [2]	←==========→																	
AEs [3]				←==========→														
Con Med review	←==========→																	
Discharge																X		
Outpatient Visit	X																X	X

1. Calcium folinate will be coadministered with Pyrimethamine on Day 1.
2. All SAEs will be collected from the signing of the ICF until the follow-up visit at the time points specified.
3. All AEs will be collected from the start of treatment until the follow-up visit at the time points specified.
4. Subjects will return to the unit on Day 15 (±1) and 22 (±1) for the remaining PK samples and safety assessments post dose.

3. INTRODUCTION

3.1. Study Rationale

Pyrimethamine in combination with a sulphonamide is effective in the treatment of toxoplasmosis. However, in Japan, Pyrimethamine has not been approved by the Japanese regulatory body PMDA/MHLW, and the use of Pyrimethamine needs to rely on obtaining it via personal import or provided by a grant-in-aid from the Japanese government. The development of Pyrimethamine was requested by the Japanese MHLW because a high medical need exists for the treatment of toxoplasmosis with Pyrimethamine in Japan.

The PK of Pyrimethamine has been investigated following administration of Sulfadoxine/Pyrimethamine tablet (Fansidar®) in healthy Japanese subjects [Otomo H, et al. 1985]. However, as this study did not provide sufficient information for approval of Pyrimethamine in Japan, the PMDA has requested confirmation of the PK of Pyrimethamine in another PK study in Japanese and Caucasian healthy subjects. The primary adverse reaction of Pyrimethamine is abnormalities of the blood and lymphatic system, such as anaemia, leucopenia and thrombocytopenia. Therefore, for all subjects receiving Pyrimethamine, coadministration of calcium folinate is recommended to reduce the risk of bone marrow depression.

Thus, the primary objective of this study is to investigate the PK following a single oral dose of Pyrimethamine 50 mg in a fasted condition in healthy Japanese and Caucasian male subjects.

3.2. Background

Pyrimethamine is an inhibitor of the enzyme dihydrofolate reductase (DHFR). It blocks the reduction of dihydrofolic acid to tetrahydrofolic acid which is an essential coenzyme in the production of nucleic acids, thereby leading to disruption of protein synthesis and nuclear division. The affinity of Pyrimethamine for protozoal DHFR is much greater than that for the mammalian enzyme. Sulphonamides act synergistically with Pyrimethamine by arresting production of dihydrofolic acid from para-aminobenzoic acid. This results in sequential blockade of the folate pathway of *Toxoplasma Gondii* which, in contrast to man, is unable to utilise preformed folate. Pyrimethamine peak plasma levels in humans generally occur 2 to 4 hours after oral administration of a 100 mg dose of Pyrimethamine and the plasma half-life is approximately 90 hours following a single oral dose of the combination of Sulfadoxine 1000 mg/Pyrimethamine 50 mg (Fansidar®). In healthy Japanese subjects, the peak plasma concentrations of Pyrimethamine occurred at 3.3 hours and the mean elimination half-life was 106.9 hours [Otomo H, et al. 1985].

Pyrimethamine in combination with a sulphonamide is effective in the treatment of toxoplasmosis, including ocular and congenital infections and toxoplasmosis in immune-deficient individuals. Pyrimethamine for the treatment of toxoplasmosis in combination with a sulphonamide was approved in 69 countries including in the US, UK, Germany, Canada and Australia., as of Dec 2014. The trade name is DARAPRIM™.

Summary information of this medicine is described in the PRODUCT INFORMATION DARAPRIM TABLETS [Amended 22Apr2016].

3.3. Benefit/Risk Assessment

More detailed information about the known and expected benefits and risks and reasonably expected adverse events (AEs) of Pyrimethamine may be found in the PRODUCT INFORMATION DARAPRIM TABLETS [Amended 22Apr2016].

3.3.1. Risk Assessment

Potential Risk of Clinical Significance	Summary of Data/Rationale for Risk	Mitigation Strategy
Investigational Product (IP) Pyrimethamine		
Pancytopenia (hematologic effects of folate deficiency)	The GSK safety database contains 45 case reports of pancytopenia. 24 of these are related to Pyrimethamine as the only causative agent. The rest of the cases are confounded by concomitant use of sulphonamides, since it is recommended that Pyrimethamine be given simultaneously with sulphonamides to achieve optimum effectiveness. Some cases are also confounded by antibiotics and antivirals. The majority of pancytopenia cases result from insufficient folate supplementation during therapy. Substantial folate deficiency - apart from pancytopenia - may cause isolated blood dyscrasias, such as: anemia, leucopenia or thrombocytopenia and such events have also been reported. This association has been known for decades and it is supported by many literature reference articles [Pajor A, 1990].	Subjects will be supplemented with a single daily oral dose of calcium folinate for 8 consecutive days (See the SoA for details). Frequent laboratory assessments for hematologic effects (full blood counts) will be carried out during the study period, and subjects monitored for signs and symptoms of depression of haematopoesis (e.g., leucopenia, anaemia, thrombocytopenia, etc.).

Potential Risk of Clinical Significance	Summary of Data/Rationale for Risk	Mitigation Strategy
Dermatitis	GSK safety database contains 8 case reports of dermatitis, consisting of: 4 reports of exfoliative dermatitis, 2 cases of bullous dermatitis and 2 cases of allergic dermatitis. Six of the 8 reports of severe skin reactions were confounded by the use of either sulphonamides (e.g., sulphadiazine) or by other co-suspect medications, so determination of the contribution of each drug in these events is difficult. The remaining 2 cases are inconclusive due to insufficient information. Since it is recommended that Pyrimethamine be given simultaneously with sulphonamides and since long-acting sulphonamides are well-known for causing various skin and subcutaneous tissue disorders, the risk of severe cutaneous reactions should be always considered when combined therapy is used [Taylor WRJ, et al. 2004].	Subjects will not receive sulphonamide in this study as a concomitant medicine (because the safety concern is associated with the recommended combined anti-parasite therapy rather than with Pyrimethamine alone and is most probably directly related to the second component of the therapy, i.e., to a sulphonamide). Subjects will be monitored for signs and symptoms of dermatitis during the study period.

3.3.2. Benefit Assessment

This study is being conducted in healthy subjects with no significant medical history. Subjects will not receive benefit from this study. Knowledge from this study may contribute to the development of Pyrimethamine in Japan and may benefit patients with toxoplasmosis in the future.

3.3.3. Overall Benefit: Risk Conclusion

Based upon the safety assessments and risk mitigation strategy of this protocol and the potential benefits to the Japanese patients with toxoplasmosis, it is concluded that there is a positive risk benefit.

In consideration of the potential benefits and risk reviewed above, GSK concludes the Benefit:Risk for a subject's participation in the GSK study 204678 is favourable and supports the study conduct.

4. OBJECTIVES AND ENDPOINTS

Objective	Endpoint
Primary	
• To investigate PK following a single oral dose of Pyrimethamine 50 mg in fasted healthy Japanese male subjects.	• Cmax, AUC(0-t), AUC(0-inf), AUC(0-24), tmax, t1/2, CL/F, Vd/F from the plasma concentrations of Pyrimethamine in healthy Japanese male subjects, as data permit.
Secondary	
• To investigate PK following a single oral dose of Pyrimethamine 50 mg in fasted healthy Caucasian male subjects.	• Cmax, AUC(0-t), AUC(0-inf), AUC(0-24), tmax, t1/2, CL/F, Vd/F from the plasma concentrations of Pyrimethamine in healthy Caucasian male subjects, as data permit.
• To assess safety and tolerability following a single oral dose of Pyrimethamine in fasted healthy Japanese and Caucasian male subjects.	• AE and change from the baseline of clinical laboratory values, vital signs and ECG.

5. STUDY DESIGN

5.1. Overall Design

This study will be a single centre, open-label, parallel-group, single oral dose study to evaluate the PK, safety and tolerability of Pyrimethamine in healthy Japanese and Caucasian male subjects.

5.2. Number of Participants

Sufficient Japanese and Caucasian healthy male subjects will be enrolled such 6 subjects of each population complete dosing and critical assessments.

If participants prematurely discontinue the study, additional replacement participants may be recruited and assigned to the same treatment at the discretion of the Sponsor in consultation with the investigator.

5.3. Participant and Study Completion

A participant is considered to have completed the study if he has completed all phases of the study including the last visit.

The end of the study is defined as the date of the last visit of the last participant in the study.

5.4. Scientific Rationale for Study Design

The PK of Pyrimethamine has been investigated following administration of Sulfadoxine/Pyrimethamine tablet (Fansidar®) in healthy Japanese subjects [Otomo H, et al. 1985]. However, this study did not include sufficient information for approval of Pyrimethamine in Japan. Confirmation of the PK of Pyrimethamine in a Japanese population, as well as comparison of the data in Japanese relative to Caucasian subjects under the same conditions should be beneficial when confirming the relevance of the existing global experience with Pyrimethamine for patients in Japan.

Daily therapeutic doses of Pyrimethamine have been shown to depress haematopoesis in some 25% to 50% of patients. The likelihood of inducing leucopenia, anaemia or thrombocytopenia is reduced by concurrent administration of calcium folinate. In addition, PMDA recommends using calcium folinate in this study to be conducted in the healthy subjects. Calcium folinate 15 mg will be dosed from Day 1 to Day 8 to prevent bone marrow depression.

The elimination half-life was approximately 107 hours after dosing 50 mg Pyrimethamine (i.e., two Sulfadoxine/Pyrimethamine (500 mg/25 mg) tablets in healthy Japanese subjects and the mean elimination half-life is reported to be approximately 90 hours in non-Japanese subjects. Considering the long half-life of Pyrimethamine, the follow-up period for 22 days together with the parallel study design are considered to be appropriate for evaluation of the PK characteristics of Pyrimethamine in both Japanese and Caucasian subjects, as well as the evaluation of safety in Japanese subjects because this is the first clinical study of this Pyrimethamine formulation in Japanese.

5.5. Dose Justification

Pyrimethamine has been approved in the US, UK and Australia and the recommended doses for adults with toxoplasmosis are as shown in the table below;

Country	Regimen
US	The adult starting dose is 50 to 75 mg of the drug daily, together with 1 to 4 g daily of a sulfonamide of the sulfapyrimidine type, e.g. sulfadoxine. This dosage is ordinarily continued for 1 to 3 weeks, depending on the response of the patient and tolerance to therapy. The dosage may then be reduced to about one half that previously given for each drug and continued for an additional 4 to 5 weeks [DARAPRIM® (pyrimethamine) 25 mg tablets Rx Only PRESCRIBING INFORMATION].
UK	A loading dose of DARAPRIM 100 mg should be given for the first 1 to 2 days, followed by 25 to 50 mg daily. This should be given together with 2 to 4 g of sulphadiazine daily in divided doses [DARAPRIM® Tablets].
Australia	50 mg for initial dose and 25 mg for subsequent daily dose with sulphadiazine or other sulphonamides [DARAPRIM® Tablets].

While a different dose is recommended for malaria, one of the proposed dose regimens for Japan for the treatment of toxoplasmosis in combination with a sulphonamide is as follows; 100 mg on the first day and 25-50 mg for subsequent daily doses. This dosage regimen is used clinically in the UK and was referred to in three randomized studies [Bosch-Driessen LH, et al. 2002; Soheilian M, et al. 2005; Rothova A, et al. 1993].

The previous study in healthy Japanese subjects using two Sulfadoxine/Pyrimethamine (500 mg/25 mg) tablets has confirmed the safety of administration of 50 mg Pyrimethamine in Japanese [Otomo H, et al. 1985; Kusuhara H, et al. 2011]. However, there is no report in which the safety has been confirmed after dosing with 100 mg Pyrimethamine in healthy Japanese subjects.

The 50 mg Pyrimethamine is within the range of that approved in the US, UK and Australia, and taking safety in healthy subjects mentioned above into consideration, 50 mg is considered the appropriate dose to investigate PK in both populations. This study will use the Pyrimethamine tablets (DARAPRIM™, Arrow Pharmaceuticals) approved and marketed in Australia.

6. STUDY POPULATION

Prospective approval of protocol deviations to recruitment and enrollment criteria, also known as protocol waivers or exemptions, is not permitted.

6.1. Inclusion Criteria

Participants are eligible to be included in the study only if all of the following criteria apply:

Age

1. Between 20 and 64 years of age inclusive, at the time of signing the informed consent.

Type of Participant and Disease Characteristics

2. Participants who are overtly healthy as determined by medical evaluation including medical history, physical examination, laboratory tests, and cardiac monitoring.

Weight

3. Body weight ≥ 50 kg and body mass index (BMI) within the range 18.5 – 30.0 kg/m^2 (inclusive).

Sex and Ancestry

4. Japanese or Caucasian male.

- A male participant must agree to use contraception as detailed in Appendix 5 of this protocol during the treatment period and until follow-up.
- Japanese ethnic origin defined as having been born in Japan, having four ethnic Japanese grandparents, holding a Japanese passport or identity papers and being able to speak Japanese. Subjects should also have lived outside Japan for less than 10 years at the time of screening.
- Caucasian subjects as defined as an individual having four grandparents who are all descendants of the original peoples of Europe.

Informed Consent

5. Capable of giving signed informed consent which includes compliance with the requirements and restrictions listed in the ICF and in this protocol.

6.2. Exclusion Criteria

Participants are excluded from the study if any of the following criteria apply:

Medical Conditions

1. Alanine aminotransferase (ALT) > 1.5x upper limit of normal (ULN)
2. and bilirubin > 1.5xULN (isolated bilirubin > 1.5xULN is acceptable if bilirubin is fractionated and direct bilirubin < 35%).
3. QTcF > 450 msec

   NOTES:

- The QTc is the QT interval corrected for heart rate according to Fridericia's formula (QTcF), and/or another method, machine-read or manually over-read.
- The specific formula that will be used to determine eligibility and discontinuation for an individual subject should be determined prior to initiation of the study. In other words, several different formulae cannot be used to calculate the QTc for an individual subject and then the lowest QTc value used to include or discontinue the subject from the trial.
- For purposes of data analysis, QTcF, another QT correction formula, or a composite of available values of QTc will be used as specified in the Reporting and Analysis Plan (RAP).

4. Current or chronic history of liver disease, or known hepatic or biliary abnormalities (with the exception of Gilbert's syndrome or asymptomatic gallstones).
5. History of cardiovascular, respiratory, hepatic, renal, gastrointestinal, endocrine, hematological, or neurological disorders capable of significantly altering the absorption, metabolism, or elimination of drugs; constituting a risk when taking the study treatment; or interfering with the interpretation of data.
6. Abnormal blood pressure as determined by the investigator.
7. Hematological values: outside normal range at screening.
8. Serum creatinine level: outside normal range at screening visit.

Prior/Concomitant Therapy

9. Past or intended use of over-the-counter or prescription medication including herbal medications within 14 days prior to dosing.

Prior/Concurrent Clinical Study Experience

10. Participation in the study would result in loss of blood or blood products in excess of 500 mL within 3 months.
11. Exposure to more than 4 new chemical entities within 12 months prior to the first dosing day.
12. Current enrolment or past participation within the last 30 days before signing of consent in this clinical study involving an investigational study treatment or any other type of medical research.

Diagnostic assessments

13. Presence of Hepatitis B surface antigen (HBsAg) at screening or positive Hepatitis C antibody test result at screening.

    NOTE:

    - Subjects with positive Hepatitis C antibody due to prior resolved disease can be enrolled, only if a confirmatory negative Hepatitis C RNA test is obtained.

14. Positive Hepatitis C RNA test result at screening or within 3 months prior to first dose of study treatment.

NOTE:

- Test is optional and subjects with negative Hepatitis C antibody test are not required to also undergo Hepatitis C RNA testing.

15. Positive pre-study drug/alcohol screen.
16. Positive human immunodeficiency virus (HIV) antibody test.
17. Regular use of known drugs of abuse.

Other Exclusions

18. Regular alcohol consumption within 6 months prior to the study defined as:

    For an average weekly intake of > 14 units for males. One unit is equivalent to 10 g of alcohol: a can of mid-strength (~375 mL) beer, 1 glass (100 mL) of table wine or 1 measure (30 mL) of spirits (including rice wine).
19. History or regular use of tobacco- or nicotine-containing products within 6 months prior to screening.
20. Sensitivity to any of the study treatments, or components thereof, or drug or other allergy that, in the opinion of the investigator or medical monitor, contraindicates participation in the study.

6.3. Lifestyle Restrictions

6.3.1. Meals and Dietary Restrictions

- Refrain from consumption of red wine, Seville oranges, grapefruit or grapefruit juice and/or pummelos, citrus fruits, grapefruit hybrids or fruit juices from 7 days prior to the first dose of study medication until after the last PK sample.
- Subjects should fast for a minimum of 10 hours prior to any blood draws for safety laboratory testing.
- During the in-house period there are meal restrictions (e.g., timing of meals on Day 1 and limitations on number of meals/snacks per day).
- During hospitalization, subjects can consume water, however they should refrain from any drinking from 1 hour prior to dosing until 4 hours post dose. Also, they should refrain from excessive drinking after 4 hours post dose.

6.3.2. Caffeine, Alcohol, and Tobacco

- Participants will abstain from ingesting caffeine- or xanthine-containing products (e.g., coffee, tea, cola drinks, and chocolate) for 24 hours prior to the start of dosing until collection of the final PK sample.
- Participants will abstain from alcohol for 24 hours prior to the start of dosing until collection of the final PK sample.

- Participants who use tobacco products will be instructed that use of nicotine-containing products (including nicotine patches) will not be permitted for 6 months prior to the screening until collection of the final PK sample.

6.3.3. Activity

Participants will abstain from strenuous exercise for 48 hours prior to each blood collection for clinical laboratory tests. Participants will remain in a sitting or semi-supine position for approximately 4 hours after dosing on Day 1 of treatment period. Participants may participate in light recreational activities during studies (e.g., watching television, reading).

6.4. Screen Failures

Screen failures are defined as participants who consent to participate in the clinical study but are not subsequently entered in the study. 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, screen failure details, eligibility criteria, and any serious adverse events (SAEs).

Individuals who do not meet the criteria for participation in this study (screen failure) may not be rescreened.

7. TREATMENTS

Study treatment is defined as any investigational treatment(s), marketed product(s), placebo, or medical device(s) intended to be administered to a study participant according to the study protocol.

Pyrimethamine (DARAPRIM™, Arrow Pharma Pty Ltd., Australia) will be purchased and labelled at Pharmaceutical Packaging Professionals, Australia.

Calcium folinate (Leucovorin®, Pfizer Japan Inc., Japan) will be purchased by GSK Japan, and labelled at Pharmaceutical Packaging Professionals, Australia.

7.1. Treatments Administered

Study Treatment Name:	Pyrimethamine	Calcium folinate
Dosage formulation:	Tablet	Tablet
Unit dose strength(s)/Dosage level(s):	25 mg/unit dose	5 mg/unit dose
Route of Administration:	Oral	Oral
Dosing instructions:	2 tablets will be taken on Day 1 in fasted condition with 240 mL water	3 tablets will be taken on Day 1 together with Pyrimethamine, and once a day until Day 8 (a total of 8 days). Each administration will be with 240 mL water. (See Section 7.7)
Packaging and Labeling:	Study Treatment will be provided in container. Each container will be labelled as required per country requirement.	Study Treatment will be provided in container. Each container will be labelled as required per country requirement.
Manufacturer:	Arrow Pharma Pty Ltd., Australia	Pfizer Japan Inc., Japan

7.2. Dose Modification

This is a single-dose study and dose modification will not be considered.

7.3. Method of Treatment Assignment

This is an open-label and non-randomized study which consists of two groups (Japanese and Caucasian). Subjects in each group will be assigned in accordance with the unique number (randomization schedule) generated by the Biomedical Data Sciences Department at GSK, prior to the start of the study, using validated internal software.

7.4. Blinding

This is an open-label study.

7.5. Preparation/Handling/Storage/Accountability

1. The investigator or designee must confirm appropriate temperature conditions have been maintained during transit for all study treatment received and any discrepancies are reported and resolved before use of the study treatment.

2. Only participants enrolled in the study may receive study treatment and only authorized site staff may supply or administer study treatment. All study treatments must be stored in a secure, environmentally controlled, and monitored (manual or automated) area in accordance with the labeled storage conditions with access limited to the investigator and authorized site staff.
3. The investigator, institution, or the head of the medical institution (where applicable) is responsible for study treatment accountability, reconciliation, and record maintenance (i.e., receipt, reconciliation, and final disposition records).
4. Further guidance and information for the final disposition of unused study treatment are provided in the Study Reference Manual.

- Under normal conditions of handling and administration, study treatment is not expected to pose significant safety risks to site staff. Take adequate precautions to avoid direct eye or skin contact and the generation of aerosols or mists. In the case of unintentional occupational exposure notify the monitor, Medical Monitor and/or GSK study contact.
- A Material Safety Data Sheet (MSDS)/equivalent document describing occupational hazards and recommended handling precautions either will be provided to the investigator, where this is required by local laws, or is available upon request from GSK.

7.6. Treatment Compliance

- When the individual dose for a participant is prepared from a bulk supply, the preparation of the dose will be confirmed by a second member of the study site staff.
- When participants are dosed at the site, they will receive study treatment directly from the investigator or designee, under medical supervision. The date and time of each dose administered in the clinic will be recorded in the source documents. The dose of study treatment and study participant identification will be confirmed at the time of dosing by a member of the study site staff other than the person administering the study treatment. Study site personnel will examine each participant's mouth to ensure that the study treatment was ingested.

7.7. Concomitant Therapy

This study includes, as a part of the study treatment, administration of calcium folinate to mitigate a potential risk of pancytopenia that has developed following administration of Pyrimethamine (Section 3.3.1).

Calcium folinate 15 mg will be coadministered with Pyrimethamine on Day 1, and will continue to be administered once a day at the same time as Day 1 until Day 8 (a total of 8 days). Details for the administration with calcium folinate are also found in SoA and Section 7.1.

Any medication or vaccine (including over-the-counter or prescription medicines, vitamins, and/or herbal supplements) that the participant is receiving at the time of enrollment or receives during the study must be recorded along with:

- reason for use
- dates of administration including start and end dates
- dosage information including dose and frequency

The Medical Monitor should be contacted if there are any questions regarding concomitant or prior therapy.

Participants must abstain from taking prescription or non-prescription drugs (including vitamins and dietary or herbal supplements) within 7 days (or 14 days if the drug is a potential enzyme inducer) or 5 half-lives (whichever is longer) before the start of study treatment until completion of the follow-up visit, unless, in the opinion of the investigator and sponsor, the medication will not interfere with the study.

7.8. Treatment after the End of the Study

Subjects will not receive any additional treatment from GSK after completion of the study because only healthy volunteers are eligible for study participation.

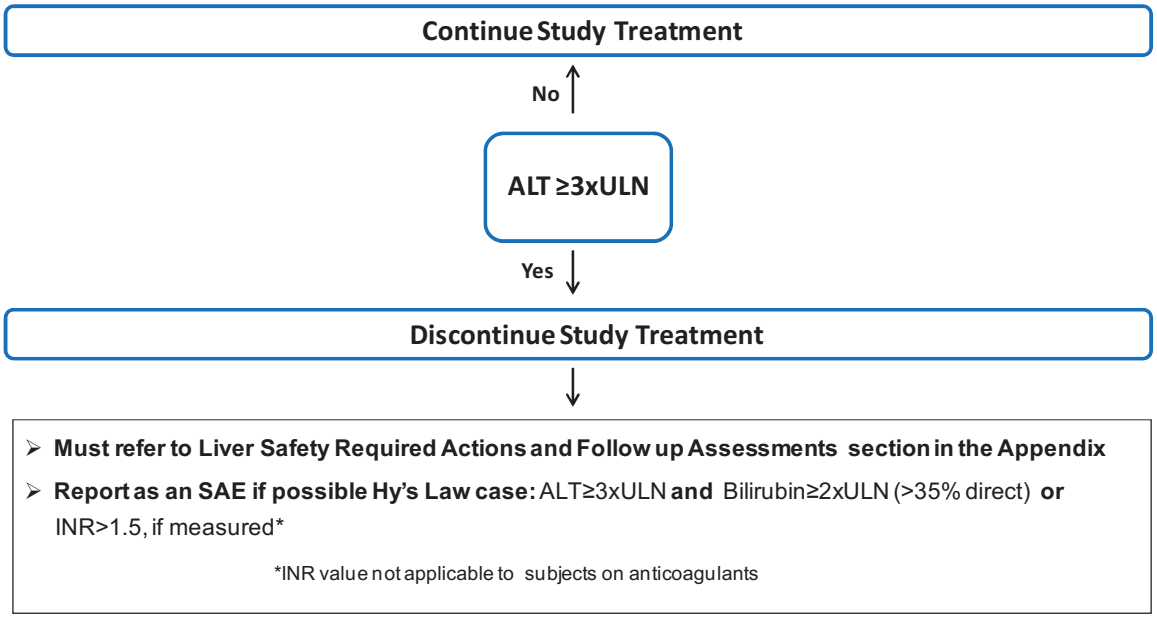
8. DISCONTINUATION CRITERIA

8.1. Discontinuation of Study Treatment

8.1.1. Liver Chemistry Stopping Criteria

Study treatment will be discontinued **for a participant** if liver chemistry stopping criteria are met:

Phase I Liver Chemistry Stopping Criteria – Liver Stopping Event Algorithm



Liver Safety Required Actions and Follow-up Assessments Section can be found in Appendix 6.

8.1.2. QTc Stopping Criteria

A subject that meets the bulleted criterion based on the average of triplicate electrocardiogram (ECG) readings will be withdrawn from study treatment.

- QTcF > 500 msec

8.2. Withdrawal from the Study

- A participant may withdraw from the study at any time at his own request, or may be withdrawn at any time at the discretion of the investigator for safety, behavioral, compliance or administrative reasons.
- If the participant withdraws consent for disclosure of future information, the sponsor may retain and continue to use any data collected before such a withdrawal of consent.
- If a participant withdraws from the study, he may request destruction of any samples taken and not tested, and the investigator must document this in the site study records.
- Refer to the SoA for data to be collected at the time of study discontinuation and follow-up and for any further evaluations that need to be completed.

8.3. Lost to Follow-Up

A participant will be considered lost to follow-up if he repeatedly fails to return for scheduled visits and is unable to be contacted by the study site.

The following actions must be taken if a participant fails to return to the clinic for a required study visit:

- The site must attempt to contact the participant and reschedule the missed visit as soon as possible and counsel the participant on the importance of maintaining the assigned visit schedule and ascertain whether or not the participant wishes to and/or should continue in the study.
- Before a participant is deemed lost to follow-up, the investigator or designee must make every effort to regain contact with the participant (where possible, 3 telephone calls and, if necessary, a certified letter to the participant's last known mailing address or local equivalent methods). These contact attempts should be documented in the participant's medical record.
- Should the participant continue to be unreachable, he will be considered to have withdrawn from the study with a primary reason of lost to follow-up.

9. STUDY ASSESSMENTS AND PROCEDURES

- Study procedures and their timing are summarized in the SoA.
- Protocol waivers or exemptions are not allowed.
- Immediate safety concerns should be discussed with the sponsor immediately upon occurrence or awareness to determine if the participant should continue or discontinue study treatment.
- Adherence to the study design requirements, including those specified in the SoA, is essential and required for study conduct.
- All screening evaluations must be completed and reviewed to confirm that potential participants meet all eligibility criteria. The investigator will maintain a screening log to record details of all participants screened and to confirm eligibility or record reasons for screening failure, as applicable.
- Procedures conducted as part of the participant's routine clinical management (e.g., blood count) and obtained before signing of ICF may be utilized for screening or baseline purposes provided the procedure met the protocol-specified criteria and was performed within the time frame defined in the SoA.
- The maximum amount of blood collected from each participant over the duration of the study, including any extra assessments that may be required, will not exceed 500 mL.
- Repeat or unscheduled samples may be taken for safety reasons or for technical issues with the samples.

9.1. Adverse Events

The definitions of an AE or SAE can be found in Appendix 4.

The investigator and any designees are responsible for detecting, documenting, and reporting events that meet the definition of an AE or SAE and remain responsible for following up AEs that are serious, considered related to the study treatment or the study, or that caused the participant to discontinue the study (see Section 8).

9.1.1. Time Period and Frequency for Collecting AE and SAE Information

- All SAEs will be collected from the signing of the ICF until the follow-up visit at the time points specified in the SoA (Section 2).
- All AEs will be collected from the start of treatment until the follow-up visit at the time points specified in the SoA (Section 2).
- Medical occurrences that begin before the start of study treatment but after obtaining informed consent will be recorded on the Medical History/Current Medical Conditions section of the case report form (CRF) not the AE section.

- All SAEs will be recorded and reported to the sponsor or designee within 24 hours, as indicated in Appendix 4. The investigator will submit any updated SAE data to the sponsor within 24 hours of it being available.
- Investigators are not obligated to actively seek AEs or SAEs in former study participants. However, if the investigator learns of any SAE, including a death, at any time after a participant has been discharged from the study, and he considers the event to be reasonably related to the study treatment or study participation, the investigator must promptly notify the sponsor.
- The method of recording, evaluating, and assessing causality of AEs and SAEs and the procedures for completing and transmitting SAE reports are provided in Appendix 4.

9.1.2. Method of Detecting AEs and SAEs

Care will be taken not to introduce bias when detecting AE and/or SAE. Open-ended and non-leading verbal questioning of the participant is the preferred method to inquire about AE occurrence.

9.1.3. Follow-up of AEs and SAEs

After the initial AE/SAE report, the investigator is required to proactively follow each participant at subsequent visits/contacts. All SAEs will be followed until the event is resolved, stabilized, otherwise explained, or the participant is lost to follow-up (as defined in Section 8.3). Further information on follow-up procedures is given in Appendix 4.

9.1.4. Regulatory Reporting Requirements for SAEs

- Prompt notification by the investigator to the sponsor of a SAE is essential so that legal obligations and ethical responsibilities towards the safety of participants and the safety of a study treatment under clinical investigation are met.
- The sponsor has a legal responsibility to notify both the local regulatory authority and other regulatory agencies about the safety of a study treatment under clinical investigation. The sponsor will comply with country-specific regulatory requirements relating to safety reporting to the regulatory authority, IEC, and investigators.
- Investigator safety reports must be prepared for suspected unexpected serious adverse reactions (SUSAR) according to local regulatory requirements and sponsor policy and forwarded to investigators as necessary.
- An investigator who receives an investigator safety report describing a SAE or other specific safety information (e.g., summary or listing of SAE) from the sponsor will review and then file it along with the Investigator's Brochure (IB) and will notify the IEC, if appropriate according to local requirements.

9.1.5. Pregnancy

- Details of all pregnancies in female partners of male participants will be collected after the start of study treatment and until the follow-up visit.
- If a pregnancy is reported, the investigator should inform GSK within 24 hours of learning of the pregnancy and should follow the procedures outlined in Appendix 5.
- Abnormal pregnancy outcomes (e.g., spontaneous abortion, fetal death, stillbirth, congenital anomalies, ectopic pregnancy) are considered SAE.

9.2. Treatment of Overdose

For this study, any dose of Pyrimethamine greater than 50 mg within a 24-hour time period will be considered an overdose.

GSK does not recommend specific treatment for an overdose.

In the event of an overdose, the investigator should:

1. Contact the Medical Monitor immediately.
2. Closely monitor the participant for AE/SAE and laboratory abnormalities until Pyrimethamine can no longer be detected systemically (at least 22 days).
3. Obtain a plasma sample for PK analysis within 22 (±1) days from the date of the last dose of study treatment if requested by the Medical Monitor (determined on a case-by-case basis).
4. Document the quantity of the excess dose as well as the duration of the overdosing in the CRF.

Decisions regarding dose interruptions or modifications will be made by the investigator in consultation with the Medical Monitor based on the clinical evaluation of the participant.

9.3. Safety Assessments

Planned time points for all safety assessments are provided in the SoA.

9.3.1. Physical Examinations

- A physical examination will include, at a minimum, assessments of the Skin, Cardiovascular, Respiratory, Gastrointestinal and Neurological systems.
- Height and weight will also be measured and recorded only at screening.
- Investigators should pay special attention to clinical signs related to previous serious illnesses.

9.3.2. Vital Signs

- Oral temperature, pulse rate, and blood pressure will be assessed.

- Blood pressure and pulse measurements will be assessed with a completely automated device. Manual techniques will be used only if an automated device is not available.
- Blood pressure and pulse measurements should be preceded by at least 5 minutes of rest for the participant in a quiet setting without distractions (e.g., television, cell phones).
- Single vital signs will be measured in a semi-supine position after 5 minutes rest and will include temperature, systolic and diastolic blood pressure, and pulse rate.

9.3.3. Electrocardiograms

- Single 12-lead ECG will be obtained as outlined in the SoA (see Section 2) using an ECG machine that automatically calculates the heart rate and measures PR, QRS, QT, and QTc intervals. Refer to Section 8.1 for QTc withdrawal criteria and additional QTc readings that may be necessary.

9.3.4. Clinical Safety Laboratory Assessments

- Refer to Appendix 2 for the list of clinical laboratory tests to be performed and to the SoA for the timing and frequency.
- The investigator must review the laboratory report, document this review, and record any clinically relevant changes occurring during the study in the AE section of the CRF. The laboratory reports must be filed with the source documents.
- All laboratory tests with values considered clinically significantly abnormal during participation in the study should be repeated until the values return to normal or baseline or are no longer considered significantly abnormal by the investigator or medical monitor.
- If such values do not return to normal/baseline within a period of time judged reasonable by the investigator, the etiology should be identified and the sponsor notified.
- All protocol-required laboratory assessments, as defined in Appendix 2, must be conducted in accordance with the laboratory manual and the SoA.
- If laboratory values from non-protocol specified laboratory assessments performed at the institution's local laboratory require a change in participant management or are considered clinically significant by the investigator (e.g., SAE or AE or dose modification), then the results must be recorded in the CRF.

9.4. Pharmacokinetics

- Whole blood samples of approximately 2 mL will be collected in EDTA blood collection tube for measurement of plasma concentrations of Pyrimethamine as specified in the SoA. Instructions for the collection and handling of biological samples will be provided by the sponsor. The actual date and time (24-hour clock time) of each sample will be recorded.

10. STATISTICAL CONSIDERATIONS

The objectives of this study are to evaluate safety and tolerability of Pyrimethamine and to estimate Pyrimethamine PK parameters in healthy Japanese and Caucasian male subjects. No formal statistical hypotheses will be tested. Descriptive statistics will be used to assess safety and tolerability objectives by group. An estimation approach will be used to address the PK study objectives, where point estimates and corresponding 95% confidence intervals (CI) will be constructed, unless otherwise stated.

10.1. Sample Size Determination

10.1.1. Sample Size Assumptions

The sample size is based on feasibility where sufficient subjects will be enrolled into the study to support the inclusion of 12 evaluable subjects for analysis and 6 subjects are assigned to each group (Japanese or Caucasian).

Between-subject estimates of variability of the PK endpoints Cmax and AUC(0-inf) of Pyrimethamine are estimated to be 18.1% (coefficient of variance, CV%) and 44.6% (CV%), respectively (based on Japanese PK data reported by Otomo H, et al. 1985). Based on these variability estimates, and a sample size of 6 (active treatment per group), the upper and lower bounds of 95% CI will be approximately 20.7% for Cmax and 56.4% for AUC(0-inf) respectively. For instance, the 95% CI of geometric mean will be 331.3-483.0 if the geometric mean of Cmax (ug/mL) is 400, and the 95% CI of geometric mean will be 25.6-62.5 if the geometric mean of AUC(0-inf) (ug·hr/mL) is 40.

10.1.2. Sample Size Sensitivity

Based on sample size of 6, if the between-subject CVb% observed in the Cmax is 10% (CVb%:19.9%) and 20% (CVb%:21.7%) greater than between-subject CVb% in the reference report, the precision estimate for the upper limit value and lower limit value of a mean of 95% CI will be 23.0% and 25.3%, respectively.

If the between-subject CVb% observed in the AUC is 10% (CVb%:49.1%) and 20% (CVb%:53.5%) greater than between-subject CVb% in the reference report, the precision estimate for the upper limit value and lower limit value of a mean of 95% CI will be 62.8% and 69.3%, respectively.

10.2. Populations for Analyses

For purposes of analysis, the following populations are defined:

Population	Description
Screened	Consisting of all participants screened in the study.
Screening Failure	Participants who sign the ICF in the study but are never subsequently administered. All participants who sign the ICF have

	the screening test in the study.
Safety	All participants who take at least one dose of study treatment.
Pharmacokinetic	This population is defined as all participants administered at least one dose of study treatment and who have PK sample taken and analyzed.

10.3. Statistical Analyses

The study results will be presented in tabular and/or graphical format and summarised descriptively. These analyses will be performed in each group. Complete details of the planned statistical analyses will be provided in the RAP.

10.3.1. Safety Analyses

All safety analyses will be performed on the Safety Population.

Statistical Analysis Methods
AE, clinical laboratory values, vital signs, ECG will be summarized by group.

10.3.2. Pharmacokinetic Analyses

All PK analyses will be described in the PK Population. The details of the PK analysis will be provided in the RAP.

Statistical Analysis Methods
Pyrimethamine concentrations
Plasma concentrations of Pyrimethamine at each assessment point will be listed to prepare figures showing individual concentration-time profiles on both linear and semi-log scales. In addition, the mean and median plasma concentrations at each assessment point will be calculated from concentration data of individual subjects to prepare figures showing mean and median plasma concentration-time profiles on both linear and semi-log scales by group.
Pyrimethamine PK parameters
The PK parameters [i.e., Cmax, AUC(0-t), AUC(0-inf), AUC(0-24), tmax, t1/2, CL/F, Vd/F] will be calculated by non-compartmental analysis from plasma Pyrimethamine concentration-time data using Phoenix WinNonlin (version 6.3 or higher), as data permit. Calculations will be based on the actual sampling times recorded during the study. All derived parameters will be listed and summarized descriptively by each group.

Others
An exploratory analysis will be performed using PK parameter plots in order to assess the results in the Japanese and Caucasian subjects.

10.3.3. Interim Analyses

No interim analyses will be planned.

11. REFERENCES

Otomo H, Tanabe K, Hioki A, Tanaka, H, Odagiri O, Uto M, Kuruma I. Pharmacokinetics of antimalarial Fansidar® in healthy Japanese subjects. *Journal of New Remedies & Clinics* 1985; 34 (3): 415-423.

Pajor A. Pancytopenia in a patient given pyrimethamine and sulphamethoxidiazine during pregnancy. *Archives of Gynecology and Obstetrics* 1990; 247 (4): 215-217.

Taylor WRJ, White NJ. Antimalarial drug toxicity: a review. *Drug Safety* 2004; 27 (1): 25-61.

Bosch-Driessen LH, Verbraak FD, Suttorp-Schulten MSA, van Ruyven RLJ, Klok AM, Hoyng CB, Rothova A. A Prospective, Randomized Trial of Pyrimethamine and Azithromycin Vs Pyrimethamine and Sulfadiazine for the Treatment of Ocular Toxoplasmosis. *American Journal of Ophthalmology* 2002; 134 (1): 34-40.

Soheilian M, Sadoughi MM, Ghajarnia M, Dehghan MH, Yazdani S, Behboudi H, Anisian A, Peyman GA. Prospective Randomized Trial of Trimethoprim/Sulfamethoxazole versus Pyrimethamine and Sulfadiazine in the Treatment of Ocular Toxoplasmosis. *Ophthalmology* 2005; 112 (11): 1876-1882.

Rothova A, Meenken C, Buitenhuis HJ, Brinkman CJ, Baarsma GS, Boen-Tan TN, de Jong PTVM, Klaassen-Broekema N, Schweitzer CMC, Timmerman Z, de Vries J,. Zaal MJW, Kijlstra A. Therapy for Ocular Toxoplasmosis. *American Journal of Ophthalmology* 1993; 115 (4): 517–523.

Kusuhara H, Ito S, Kumagai Y, Jiang M, Shiroshita T, Moriyama Y, Inoue K, Yuasa H, Sugiyama Y. Effects of a MATE Protein Inhibitor, Pyrimethamine, on the Renal Elimination of Metformin at Oral Microdose and at Therapeutic Dose in Healthy Subjects. *Clinical pharmacology & Therapeutics* 2011; 89 (6): 837-844.

PRODUCT INFORMATION DARAPRIM TABLETS [Amended 22Apr2016]

"DARAPRIM® (pyrimethamine) 25 mg tablets Rx Only PRESCRIBING INFORMATION" Revised 03/2017 U.S. Food and Drug Administration. https://www.accessdata.fda.gov/drugsatfda_docs/label/2017/008578s019lbl.pdf [accessed 22/06/2017]

"Daraprim Tablets" Revised 27/05/2016 EMC. http://www.medicines.org.uk/emc/medicine/729/SPC/ [accessed 22/06/2017]

12. APPENDICES

12.1. Appendix 1: Abbreviations and Trademarks

Abbreviations

AE	Adverse event
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
AUC	Area under the concentration-time curve
AUC(0-24)	Area under the concentration-time curve from time 0 to 24
AUC(0-inf)	Area under the concentration-time curve from time 0 to infinity
AUC(0-t)	Area under the concentration-time curve from time 0 to t
BMI	Body mass index
BUN	Blood urea nitrogen
CFR	Code of Federal Regulations
CI	Confidence intervals
CIOMS	Council for International Organizations of Medical Sciences
CL/F	Apparent clearance following oral dosing
Cmax	Maximum observed concentration
CONSORT	Consolidated Standards of Reporting Trials
CPK	Creatine phosphokinase
(e)CRF	(electronic) Case report form
CSR	Clinical Study Report
CV	Coefficient of variation
DHFR	Dihydrofolate reductase
ECG	Electrocardiogram
e.g.	exempli gratia
GCP	Good Clinical Practice
GSK	GlaxoSmithKline
h/hr(s)	Hour(s)
HBsAg	Hepatitis B surface antigen
HIPAA	Health Insurance Portability and Accountability Act
HIV	Human Immunodeficiency Virus
HPLC	High performance liquid chromatography
IB	Investigator's Brochure
ICF	Informed Consent Form
ICH	International Conference on Harmonization
i.e.	id est
IEC	Independent Ethics Committees
IgG	Immunoglobulin G
IgM	Immunoglobulin M
INR	International Normalized Ratio
IP	Investigational Product
IUD	Intrauterine device
IUS	Intrauterine hormone-releasing system

kg	Kilogram
LDH	Lactate dehydrogenase
MCH	Mean Corpuscular Hemoglobin
MCV	Mean Corpuscular Volume
mg	Milligram
MHLW	Ministry of Health, Labor and Welfare
mL	Milliliter
MSDS	Material Safety Data Sheet
msec	Millisecond
pH	Pondus Hydrogenii
PK	Pharmacokinetic(s)
PMDA	Pharmaceutical and Medical Devices Agency
QTc	QT interval corrected
QTcF	QT interval corrected for heart rate by Friderician formula
RAP	Reporting and Analysis Plan
RBC	Red Blood Cells
RNA	Ribonucleic acid
SAE	Serious adverse event
SoA	Schedule of Activities
SUSAR	Suspected unexpected serious adverse reactions
t1/2	Terminal half-life
tmax	Time to maximum observed concentration
ug	Microgram
ULN	Upper limit of normal
Vd/F	Apparent volume of distribution following oral dosing
WBC	White blood cells
WOCBP	Women of Childbearing Potential

Trademark Information

Trademarks of the GlaxoSmithKline group of companies
DARAPRIM

Trademarks not owned by the GlaxoSmithKline group of companies
Fansidar
WinNonlin
Leucovorin

12.2. Appendix 2: Clinical Laboratory Tests

- The tests detailed in Table 1 will be performed by the local laboratory.
- Protocol-specific requirements for inclusion or exclusion of participants are detailed in Section 6 of the protocol.
- Additional tests may be performed at any time during the study as determined necessary by the investigator or required by local regulations.
- The results of each test must be electronic transfer of data, except drug screen, breath tests and serology.

Table 1 Protocol-Required Safety Laboratory Assessments

Laboratory Assessments	Parameters			
Hematology	Platelet Count RBC Count Hemoglobin Hematocrit	RBC Indices: MCV MCH %Reticulocytes	WBC count with Differential: Neutrophils Lymphocytes Monocytes Eosinophils Basophils	
Clinical Chemistry[1]	BUN	Potassium	Aspartate Aminotransferase (AST)	Total and direct bilirubin
	Creatinine	Sodium	Alanine Aminotransferase (ALT)	Total Protein
	Glucose fasting	Calcium	Alkaline phosphatase	
Routine Urinalysis	• Specific gravity • pH, glucose, protein, blood, ketones, bilirubin, urobilinogen, nitrite, leukocyte esterase by dipstick • Microscopic examination (if blood or protein is abnormal)			
Other tests	• Breath test (Alcohol and CO)			
Other Screening Tests	• Urine drug screen (to include at minimum: amphetamines, barbiturates, cocaine, opiates, cannabinoids and benzodiazepines) • Serology (HIV antibody, hepatitis B surface antigen [HBsAg], and hepatitis C virus antibody)			

NOTES :

1. Details of liver chemistry stopping criteria and required actions and follow-up assessments after liver stopping or monitoring event are given in Section 8.1 and Appendix 7 All events of ALT ≥ 3 × upper limit of normal (ULN) and bilirubin ≥ 2 × ULN (> 35% direct bilirubin) or ALT ≥ 3 × ULN and international normalized ratio (INR) > 1.5, if INR measured, which may indicate severe liver injury (possible Hy's Law), must be reported as an SAE (excluding studies of hepatic impairment or cirrhosis).

12.3. Appendix 3: Study Governance Considerations

Regulatory and Ethical Considerations

- This study will be conducted in accordance with the protocol and with:
  - Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences (CIOMS) International Ethical Guidelines
  - Applicable ICH Good Clinical Practice (GCP) Guidelines
  - Applicable laws and regulations
- The protocol, protocol amendments, ICF, Investigator Brochure, and other relevant documents (e.g., advertisements) must be submitted to an IEC by the investigator and reviewed and approved by the IEC before the study is initiated.
- Any amendments to the protocol will require IEC approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study participants.
- The investigator will be responsible for the following:
  - Providing written summaries of the status of the study to the IEC annually or more frequently in accordance with the requirements, policies, and procedures established by the IEC
  - Notifying the IEC of SAE or other significant safety findings as required by IEC procedures
  - Providing oversight of the conduct of the study at the site and adherence to requirements of 21 CFR, ICH guidelines, the IEC, European regulation 536/2014 for clinical studies (if applicable), and all other applicable local regulations

Financial Disclosure

Investigators and sub-investigators will provide the sponsor with sufficient, accurate financial information as requested to allow the sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate regulatory authorities. Investigators are responsible for providing information on financial interests during the course of the study and for 1 year after completion of the study.

Informed Consent Process

- The investigator or his/her representative will explain the nature of the study to the participant or his legally authorized representative and answer all questions regarding the study.
- Participants must be informed that their participation is voluntary. Participants or their legally authorized representative will be required to sign a statement of

informed consent that meets the requirements of 21 CFR 50, local regulations, ICH guidelines, Health Insurance Portability and Accountability Act (HIPAA) requirements, where applicable, and the IEC or study center.

- The medical record must include a statement that written informed consent was obtained before the participant was enrolled in the study and the date the written consent was obtained. The authorized person obtaining the informed consent must also sign the ICF.
- Participants must be re-consented to the most current version of the ICF(s) during their participation in the study.
- A copy of the ICF(s) must be provided to the participant or the participant's legally authorized representative.

The ICF may contain a separate section that addresses the use of remaining mandatory samples for optional exploratory research in accordance with SOP-GSKF-410. The investigator or authorized designee will explain to each participant the objectives of the exploratory research. Participants will be told that they are free to refuse to participate and may withdraw their consent at any time and for any reason during the storage period. A separate signature will be required to document a participant's agreement to allow any remaining specimens to be used for exploratory research. Participants who decline to participate will not provide this separate signature.

Data Protection

- Participants will be assigned a unique identifier by the sponsor. Any participant records or datasets that are transferred to the sponsor will contain the identifier only; participant names or any information which would make the participant identifiable will not be transferred.
- The participant must be informed that his personal study-related data will be used by the sponsor in accordance with local data protection law. The level of disclosure must also be explained to the participant.
- The participant must be informed that his medical records may be examined by Clinical Quality Assurance auditors or other authorized personnel appointed by the sponsor, by appropriate IEC members, and by inspectors from regulatory authorities.

Publication Policy

- The results of this study may be published or presented at scientific meetings. If this is foreseen, the investigator agrees to submit all manuscripts or abstracts to the sponsor before submission. This allows the sponsor to protect proprietary information and to provide comments.
- The sponsor will comply with the requirements for publication of study results. In accordance with standard editorial and ethical practice, the sponsor will generally support publication of multicenter studies only in their entirety and not as individual site data. In this case, a coordinating investigator will be designated by mutual agreement.

- Authorship will be determined by mutual agreement and in line with International Committee of Medical Journal Editors authorship requirements.

Data Quality Assurance

- All participant data relating to the study will be recorded on printed or electronic CRF unless transmitted to the sponsor or designee electronically (e.g., laboratory data). The investigator is responsible for verifying that data entries are accurate and correct by physically or electronically signing the CRF.
- The investigator must maintain accurate documentation (source data) that supports the information entered in the CRF.
- The investigator must permit study-related monitoring, audits, IEC review, and regulatory agency inspections and provide direct access to source data documents.
- The sponsor or designee is responsible for the data management of this study including quality checking of the data.
- Study monitors will perform ongoing source data verification to confirm that data entered into the CRF by authorized site personnel are accurate, complete, and verifiable from source documents; that the safety and rights of participants are being protected; and that the study is being conducted in accordance with the currently approved protocol and any other study agreements, ICH GCP, and all applicable regulatory requirements.
- Records and documents, including signed ICF, pertaining to the conduct of this study must be retained by the investigator for 25 years from the issue of the final Clinical Study Report (CSR)/ equivalent summary unless local regulations or institutional policies require a longer retention period. No records may be destroyed during the retention period without the written approval of the sponsor. No records may be transferred to another location or party without written notification to the sponsor.

Source Documents

- Source documents provide evidence for the existence of the participant and substantiate the integrity of the data collected. Source documents are filed at the investigator’s site.
- Data reported on the CRF or entered in the eCRF that are transcribed from source documents must be consistent with the source documents or the discrepancies must be explained. The investigator may need to request previous medical records or transfer records, depending on the study. Also, current medical records must be available.
- Definition of what constitutes source data can be found in the list of the source documents.

Study and Site Closure

GSK or its designee reserves the right to close the study site or terminate the study at any time for any reason at the sole discretion of GSK. Study sites will be closed upon study completion. A study site is considered closed when all required documents and study supplies have been collected and a study-site closure visit has been performed.

The investigator may initiate study-site closure at any time, provided there is reasonable cause and sufficient notice is given in advance of the intended termination.

Reasons for the early closure of a study site by the sponsor or investigator may include but are not limited to:

- Failure of the investigator to comply with the protocol, the requirements of the IEC or local health authorities, the sponsor's procedures, or GCP guidelines
- Inadequate recruitment of participants by the investigator
- Discontinuation of further study treatment development

12.4. Appendix 4: Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting

Definition of AE

AE Definition
• An AE is any untoward medical occurrence in a clinical study participant, temporally associated with the use of a study treatment, whether or not considered related to the study treatment. • NOTE: An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of a study treatment.

Events <u>Meeting</u> the AE Definition
• Any abnormal laboratory test results (hematology, clinical chemistry, or urinalysis) or other safety assessments (e.g., ECG, radiological scans, vital signs measurements), including those that worsen from baseline, considered clinically significant in the medical and scientific judgment of the investigator (i.e., not related to progression of underlying disease). • Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition. • New conditions detected or diagnosed after study treatment administration even though it may have been present before the start of the study. • Signs, symptoms, or the clinical sequelae of a suspected drug-drug interaction. • Signs, symptoms, or the clinical sequelae of a suspected overdose of either study treatment or a concomitant medication. Overdose per se will not be reported as an AE/SAE unless it is an intentional overdose taken with possible suicidal/self-harming intent. Such overdoses should be reported regardless of sequelae.

Events <u>NOT</u> Meeting the AE Definition
• Any clinically significant abnormal laboratory findings or other abnormal safety assessments which are associated with the underlying disease, unless judged by the investigator to be more severe than expected for the participant's condition. • The disease/disorder being studied or expected progression, signs, or symptoms of the disease/disorder being studied, unless more severe than expected for the participant's condition. • Medical or surgical procedure (e.g., endoscopy, appendectomy): the condition that leads to the procedure is the AE. • Situations in which an untoward medical occurrence did not occur (social and/or

convenience admission to a hospital).

- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

Definition of SAE

If an event is not an AE per definition above, then it cannot be an SAE even if serious conditions are met (e.g., hospitalization for signs/symptoms of the disease under study, death due to progression of disease).

A SAE is defined as any untoward medical occurrence that, at any dose:

a. Results in death

b. Is life-threatening

The term 'life-threatening' in the definition of 'serious' refers to an event in which the participant was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.

c. Requires inpatient hospitalization or prolongation of existing hospitalization

In general, hospitalization signifies that the participant has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AE. If a complication prolongs hospitalization or fulfills any other serious criteria, the event is serious. When in doubt as to whether "hospitalization" occurred or was necessary, the AE should be considered serious.

Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE.

d. Results in persistent disability/incapacity

- The term disability means a substantial disruption of a person's ability to conduct normal life functions.
- This definition is not intended to include experiences of relatively minor medical significance such as uncomplicated headache, nausea, vomiting, diarrhea, influenza, and accidental trauma (e.g., sprained ankle) which may interfere with or prevent everyday life functions but do not constitute a substantial disruption.

e. Is a congenital anomaly/birth defect

f. Other situations:

- Medical or scientific judgment should be exercised in deciding whether SAE reporting is appropriate in other situations such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the participant or may require medical or surgical intervention to prevent one of the other outcomes listed in the above definition. These events should usually

be considered serious.

Examples of such events include invasive or malignant cancers, intensive treatment in an emergency room or at home for allergic bronchospasm, blood dyscrasias or convulsions that do not result in hospitalization, or development of drug dependency or drug abuse.

Recording AE and SAE

AE and SAE Recording

- When an AE/SAE occurs, it is the responsibility of the investigator to review all documentation (e.g., hospital progress notes, laboratory, and diagnostics reports) related to the event.
- The investigator will then record all relevant AE/SAE information in the CRF.
- It is **not** acceptable for the investigator to send photocopies of the participant's medical records to GSK in lieu of completion of the GSK/AE/SAE CRF page.
- There may be instances when copies of medical records for certain cases are requested by GSK. In this case, all participant identifiers, with the exception of the participant number, will be redacted on the copies of the medical records before submission to GSK.
- The investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE.

Assessment of Intensity

The investigator will make an assessment of intensity for each AE and SAE reported during the study and assign it to 1 of the following categories:

- Mild: An event that is easily tolerated by the participant, causing minimal discomfort and not interfering with everyday activities.
- Moderate: An event that causes sufficiently discomfort and interferes with normal everyday activities.
- Severe: An event that prevents normal everyday activities. An AE that is assessed as severe should not be confused with an SAE. Severe is a category utilized for rating the intensity of an event; and both AE and SAE can be assessed as severe.

  An event is defined as 'serious' when it meets at least 1 of the predefined outcomes as described in the definition of an SAE, NOT when it is rated as severe.

Assessment of Causality

- The investigator is obligated to assess the relationship between study treatment and each occurrence of each AE/SAE.

- A "reasonable possibility" of a relationship conveys that there are facts, evidence, and/or arguments to suggest a causal relationship, rather than a relationship cannot be ruled out.
- The investigator will use clinical judgment to determine the relationship.
- Alternative causes, such as underlying disease(s), concomitant therapy, and other risk factors, as well as the temporal relationship of the event to study treatment administration will be considered and investigated.
- The investigator will also consult the IB and/or Product Information, for marketed products, in his/her assessment.
- For each AE/SAE, the investigator **must** document in the medical notes that he/she has reviewed the AE/SAE and has provided an assessment of causality.
- There may be situations in which an SAE has occurred and the investigator has minimal information to include in the initial report to GSK. However, **it is very important that the investigator always make an assessment of causality for every event before the initial transmission of the SAE data to** GSK**.**
- The investigator may change his/her opinion of causality in light of follow-up information and send an SAE follow-up report with the updated causality assessment.
- The causality assessment is one of the criteria used when determining regulatory reporting requirements.

Follow-up of AE and SAE

- The investigator is obligated to perform or arrange for the conduct of supplemental measurements and/or evaluations as medically indicated or as requested by GSK to elucidate the nature and/or causality of the AE or SAE as fully as possible. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals.
- If a participant dies during participation in the study or during a recognized follow-up period, the investigator will provide GSK with a copy of any post-mortem findings including histopathology.
- New or updated information will be recorded in the originally completed CRF.
- The investigator will submit any updated SAE data to GSK within 24 hours of receipt of the information.

Reporting of SAE to GSK

SAE Reporting to GSK via Electronic Data Collection Tool
• The primary mechanism for reporting SAE to GSK will be the electronic data collection tool. • If the electronic system is unavailable for more than 24 hours, then the site will use the paper SAE data collection tool (see next section). • The site will enter the SAE data into the electronic system as soon as it becomes available. • The investigator or medically-qualified sub-investigator must show evidence within the eCRF (e.g., check review box, signature, etc.) of review and verification of the relationship of each SAE to IP/study participation (causality) within 72 hours of SAE entry into the eCRF. • After the study is completed at a given site, the electronic data collection tool will be taken off-line to prevent the entry of new data or changes to existing data. • If a site receives a report of a new SAE from a study participant or receives updated data on a previously reported SAE after the electronic data collection tool has been taken off-line, then the site can report this information on a paper SAE form.

SAE Reporting to GSK via Paper CRF
• Facsimile transmission of the SAE paper CRF is the preferred method to transmit this information to the **medical monitor**. • In rare circumstances and in the absence of facsimile equipment, notification by telephone is acceptable with a copy of the SAE data collection tool sent by overnight mail or courier service. • Initial notification via telephone does not replace the need for the investigator to complete and sign the SAE CRF pages within the designated reporting time frames.

12.5. Appendix 5: Contraceptive Guidance and Collection of Pregnancy Information

Contraception Guidance

Male participants

- Male participants with female partners of child-bearing potential are eligible to participate if they agree to ONE of the following during the protocol-defined time frame in Section 6.1:
  - Are abstinent from penile-vaginal intercourse as their usual and preferred lifestyle (abstinent on a long term and persistent basis) and agree to remain abstinent
  - Agree to use a male condom plus an additional method of contraception with a failure rate of < 1% per year as described in Table 2 when having penile-vaginal intercourse with a woman of childbearing potential
- Men with a pregnant or breastfeeding partner must agree to remain abstinent from penile-vaginal intercourse or use a male condom during each episode of penile penetration during the protocol-defined time frame
- Refrain from donating sperm for duration of study and until follow-up visit

Table 2 Highly Effective Contraceptive Methods

Highly Effective Contraceptive Methods That Are User Dependent [a] *Failure rate of < 1% per year when used consistently and correctly.*
Combined (estrogen- and progestogen-containing) hormonal contraception associated with inhibition of ovulation[b] • oral • intravaginal • transdermal
Progestogen-only hormonal contraception associated with inhibition of ovulation[b] • injectable
Highly Effective Methods That Are User Independent
• Implantable progestogen-only hormonal contraception associated with inhibition of ovulation[b] • Intrauterine device (IUD) • Intrauterine hormone-releasing system (IUS) • bilateral tubal occlusion
Vasectomized partner *(A vasectomized partner is a highly effective contraception method provided that the partner is*

the sole male sexual partner of the WOCBP and the absence of sperm has been confirmed. If not, an additional highly effective method of contraception should be used.)
Sexual abstinence (*Sexual abstinence is considered a highly effective method only if defined as refraining from heterosexual intercourse during the entire period of risk associated with the study drug. The reliability of sexual abstinence needs to be evaluated in relation to the duration of the study and the preferred and usual lifestyle of the participant.)*

NOTES:

a. Typical use failure rates may differ from those when used consistently and correctly. Use should be consistent with local regulations regarding the use of contraceptive methods for participants in clinical studies.
b. Hormonal contraception may be susceptible to interaction with the study drug, which may reduce the efficacy of the contraceptive method. In this case two highly effective methods of contraception should be utilized during the treatment period and for at least, corresponding to time needed to eliminate study treatment plus 30 days for study treatments with genotoxic potential after the last dose of study treatment.

Collection of Pregnancy Information

Male participants with partners who become pregnant

- Investigator will attempt to collect pregnancy information on any male participant's female partner of a male study participant who becomes pregnant while participating in this study. This applies only to participants who receive study treatment.
- After obtaining the necessary signed informed consent from the pregnant female partner directly, the investigator will record pregnancy information on the appropriate form and submit it to GSK within 24 hours of learning of the partner's pregnancy.
- Partner will also be followed to determine the outcome of the pregnancy. Information on the status of the mother and child will be forwarded to GSK.
- Generally, follow-up will be no longer than 6 to 8 weeks following the estimated delivery date. Any termination of the pregnancy will be reported regardless of fetal status (presence or absence of anomalies) or indication for procedure.

12.6. Appendix 6: Liver Safety Required Actions and Follow-up Assessments

Phase I liver chemistry stopping criteria and required follow-up assessments

<table>
<tr><th colspan="3">Liver Chemistry Stopping Criteria</th></tr>
<tr><td>ALT-absolute</td><td colspan="2">ALT ≥ 3xULN
If ALT ≥ 3xULN AND bilirubin[1,2] ≥ 2xULN (> 35% direct bilirubin) or INR > 1.5, Report as an SAE.
See additional Actions and Follow-Up Assessments listed below</td></tr>
<tr><th colspan="3">Required Actions and Follow-up Assessments</th></tr>
<tr><th>Actions</th><th colspan="2">Follow-Up Assessments</th></tr>
<tr><td>• Report the event to GSK within 24 hrs
• Complete the liver event CRF, and complete an SAE data collection tool if the event also meets the criteria for an SAE[2]
• Perform liver event follow-up assessments
• Monitor the subject until liver chemistries resolve, stabilise, or return to within baseline (see MONITORING below)
MONITORING:
If ALT ≥ 3xULN AND bilirubin ≥ 2xULN or INR > 1.5
• Repeat liver chemistries (include ALT, AST, alkaline phosphatase, bilirubin) and perform liver event follow-up assessments within 24 hrs
• Monitor subjects twice weekly until liver chemistries resolve, stabilise or return to within baseline
• A specialist or hepatology consultation is recommended
If ALT ≥ 3xULN AND bilirubin < 2xULN and INR ≤ 1.5:
• Repeat liver chemistries (include ALT, AST, alkaline phosphatase, bilirubin) and perform liver event follow-up assessments within 24-72 hrs</td><td colspan="2">• Viral hepatitis serology[3]
• Obtain INR and recheck with each liver chemistry assessment until the transaminases values show downward trend
• Obtain blood sample for PK analysis, obtained within 24 hrs of the dose
• Serum creatine phosphokinase (CPK) and lactate dehydrogenase (LDH)
• Fractionate bilirubin, if total bilirubin ≥ 2xULN
• Obtain complete blood count with differential to assess eosinophilia
• Record the appearance or worsening of clinical symptoms of liver injury, or hypersensitivity, on the AE report form
• Record use of concomitant medications on the concomitant medications report form including acetaminophen, herbal remedies, other over the counter medications
• Record alcohol use on the liver event alcohol intake case report form
If ALT ≥ 3xULN AND bilirubin ≥ 2xULN or INR > 1.5:
• Anti-nuclear antibody, anti-smooth muscle antibody, Type 1 anti-liver kidney</td></tr>
</table>

Liver Chemistry Stopping Criteria	
• Monitor subjects weekly until liver chemistries resolve, stabilize or return to within baseline	microsomal antibodies, and quantitative total immunoglobulin G (IgG) or gamma globulins • Serum acetaminophen adduct high performance liquid chromatography (HPLC) assay (quantifies potential acetaminophen contribution to liver injury in subjects with definite or likely acetaminophen use in the preceding week [James LP, 2009] • Liver imaging (ultrasound, magnetic resonance, or computerised tomography) and/or liver biopsy to evaluate liver disease; complete Liver Imaging and/or Liver Biopsy CRF forms

1. Serum bilirubin fractionation should be performed if testing is available. If serum bilirubin fractionation is not immediately available, discontinue study treatment for that subject if ALT ≥ 3xULN and bilirubin ≥ 2xULN. Additionally, if serum bilirubin fractionation testing is unavailable, record presence of detectable urinary bilirubin on dipstick, indicating direct bilirubin elevations and suggesting liver injury.
2. All events of ALT ≥ 3xULN and bilirubin ≥ 2xULN (> 35% direct bilirubin) or ALT ≥ 3xULN and INR > 1.5, if INR measured, which may indicate severe liver injury (possible 'Hy's Law'), must be reported as an SAE (excluding studies of hepatic impairment or cirrhosis); INR measurement is not required and the threshold value stated will not apply to subjects receiving anticoagulants.
3. Includes: Hepatitis A IgM antibody; Hepatitis B surface antigen and Hepatitis B Core Antibody (IgM); Hepatitis C RNA; Cytomegalovirus IgM antibody; Epstein-Barr viral capsid antigen IgM antibody (or if unavailable, obtain heterophile antibody or monospot testing); Hepatitis E IgM antibody.

References

James LP, Letzig L, Simpson PM, Capparelli E, Roberts DW, Hinson JA, Davern TJ, Lee WM. Pharmacokinetics of Acetaminophen-Protein Adducts in Adults with Acetaminophen Overdose and Acute Liver Failure. *Drug Metabolism and Disposition* 2009; 37 (8): 1779-1784.

12.7. Appendix 7: Country-specific requirements

Not applicable.

12.8. Appendix 8: Protocol Amendment History

Not applicable.