Official Title: **A RANDOMIZED, SPONSOR-OPEN, INVESTIGATOR-BLINDED, SUBJECT-BLINDED, PLACEBO-CONTROLLED, SINGLE-ASCENDING DOSE (SAD) AND MULTIPLE-ASCENDING DOSE (MAD) STUDY TO INVESTIGATE THE SAFETY, TOLERABILITY, AND PHARMACOKINETICS OF RO7049389 IN HEALTHY CHINESE SUBJECTS**

NCT Number: NCT03570658

Document Date: Protocol Version 4: 20-Nov-2018

PROTOCOL

TITLE:	**A RANDOMIZED, SPONSOR-OPEN, INVESTIGATOR-BLINDED, SUBJECT-BLINDED, PLACEBO-CONTROLLED, SINGLE-ASCENDING DOSE (SAD) AND MULTIPLE-ASCENDING DOSE (MAD) STUDY TO INVESTIGATE THE SAFETY, TOLERABILITY, AND PHARMACOKINETICS OF RO7049389 IN HEALTHY CHINESE SUBJECTS**
PROTOCOL NUMBER:	YP39406
VERSION:	4
EUDRACT NUMBER:	N/A
IND NUMBER:	N/A
TEST PRODUCT:	RO7049389
SPONSOR:	F. Hoffmann-La Roche Ltd
DATE FINAL:	Version 1: 27 January 2017
DATE AMENDED:	Version 2: 16 December 2017 Version 3: 23 February 2018 Version 4: See electronic date stamp below

FINAL PROTOCOL APPROVAL

Approver's Name	Title	Date and Time (UTC)
[redacted]	Company Signatory	20-Nov-2018 10:47:28

CONFIDENTIAL

The information contained in this document, especially any unpublished data, is the property of F. Hoffmann-La Roche Ltd (or under its control) and therefore is provided to you in confidence as an investigator, potential investigator, or consultant, for review by you, your staff, and an applicable Ethics Committee or Institutional Review Board. It is understood that this information will not be disclosed to others without written authorization from Roche except to the extent necessary to obtain informed consent from persons to whom the drug may be administered.

PROTOCOL ACCEPTANCE FORM

TITLE:	**A RANDOMIZED, SPONSOR-OPEN, INVESTIGATOR-BLINDED, SUBJECT-BLINDED, PLACEBO-CONTROLLED, SINGLE-ASCENDING DOSE (SAD) AND MULTIPLE-ASCENDING DOSE (MAD) STUDY TO INVESTIGATE THE SAFETY, TOLERABILITY, AND PHARMACOKINETICS OF RO7049389 IN HEALTHY CHINESE SUBJECTS**
PROTOCOL NUMBER:	YP39406
VERSION NUMBER:	4
EUDRACT NUMBER:	N/A
IND NUMBER:	N/A
TEST PRODUCT:	RO7049389
SPONSOR:	F. Hoffmann-La Roche Ltd

I agree to conduct the study in accordance with the current protocol.

Principal Investigator's Name (print)

Principal Investigator's Signature

Date

Please keep the signed original form in your study files, and return a copy to your local study monitor.

PROTOCOL AMENDMENT, VERSION 4
RATIONALE

Protocol YP39406 has been amended to incorporate the following changes based on the emerging data from this study (YP39406) and the first-in-human (FIH) study (YP39364):

- [redacted] The modification to the study design will support characterization of safety, tolerability and PK profiles of RO7049389 in healthy Chinese subjects.
- [redacted] The highest dose level to be included in this study will now be based on the observed exposure in the FIH study (YP39364) which has been demonstrated to be safe and well tolerated.

Additional minor changes have been made to correct typographical errors and to improve clarity and consistency. Substantial new information appears in italics. This amendment represents cumulative changes to the original protocol.

PROTOCOL AMENDMENT, VERSION 4:
SUMMARY OF CHANGES

PROTOCOL SYNOPSIS

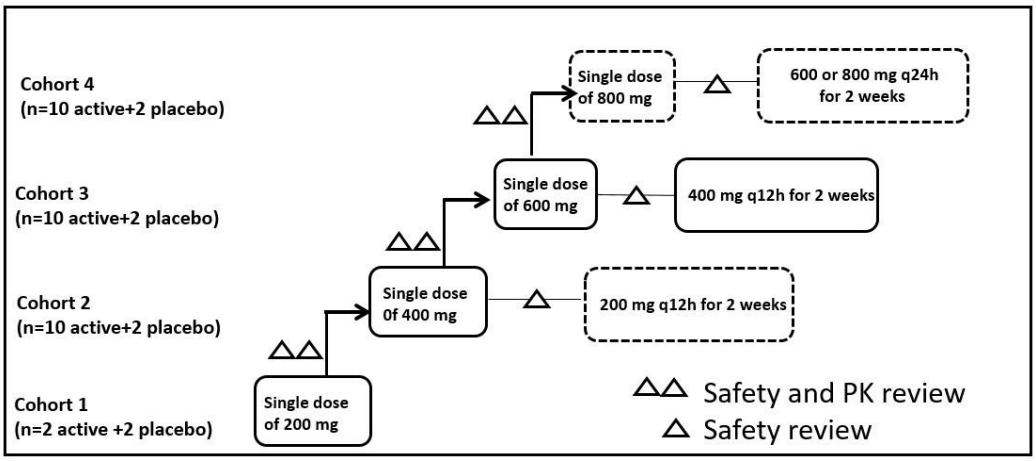
The protocol synopsis has been updated to reflect the changes to the protocol, where applicable.

3.1.1 Overview of Study Design

This study will leverage the data which will have been generated in the FIH study, allowing targeted evaluation of fewer doses. ~~In addition, this study proposes to evaluate SAD and MAD of the same total daily dose level in a single subject cohort after approximately 1 week washout. The total daily dose in the MAD part will be equal or lower than the dose used in the SAD part for the same cohort.~~

[…]

Figure 1Overall Study Design and Dose-escalation – new Figure



[redacted]

Dotted lines indicate optional cohorts. See Table 1 for additional information.

Escalation to the next single dose cohort will be based primarily on the safety and available PK data through 24 hours post-single dose in the previous cohorts. The MAD phase may start after a wash-out period of approximately 1-week, providing that the safety information through 24 hours post-dose in the SAD phase at the same *cohort*~~dose level~~, and available safety information ~~from MAD cohorts~~ in the entry-into-human study (YP39364), have been reviewed.

[….]

[….]

The highest dose level *and exposure* to be included in this study will be within the dose *and exposure* ~~range~~ *that* ~~which~~ has been demonstrated to be safe and well tolerated in FIH Study YP39364.

3.1.1.3 Dietary Requirements

[….]

In the MAD phase *(except cohort 4, which will be in the fasted state)*, a standard breakfast or dinner will be served to healthy volunteers 30 minutes prior to dosing, which should be consumed within 30 minutes.

[….]

3.1.2.1 Dose Escalation Stopping Rules

[….]

- It is predicted that further dose-escalation will not result in a further increase in plasma exposure.
- The proposed dose *or exposure* exceeds those previously evaluated in the study YP39364.

[….]

3.2.1 Rationale for Dosage Selection

[….]

Subsequent dose-levels may be adjusted based on the PK, tolerability and safety results from FIH Study YP39364 and available data from current Study YP39406. The highest dose-level *and exposure* to be included in this study will be within the dose range *and exposure* which has been demonstrated to be safe and well tolerated in FIH Study YP39364.

4.3.2.1 RO7049389 and Placebo

MAD Phase

[….]

Except for cohort 4, which will be in the fasted state, ~~a~~A standard breakfast or dinner will be served to healthy volunteers 30 minutes prior to dosing, which should be consumed within 30 minutes. RO7049389 or placebo will then be administered.

[….]

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF APPENDICES

PROTOCOL SYNOPSIS

TITLE:	**A RANDOMIZED, SPONSOR-OPEN, INVESTIGATOR-BLINDED, SUBJECT-BLINDED, PLACEBO-CONTROLLED, SINGLE-ASCENDING DOSE (SAD) AND MULTIPLE-ASCENDING DOSE (MAD) STUDY TO INVESTIGATE THE SAFETY, TOLERABILITY, AND PHARMACOKINETICS OF RO7049389 IN HEALTHY CHINESE SUBJECTS**
PROTOCOL NUMBER:	**YP39406**
VERSION:	4
EUDRACT NUMBER:	N/A
IND NUMBER:	N/A
TEST PRODUCT:	RO7049389
PHASE:	I
INDICATION:	N/A
SPONSOR:	F. Hoffmann-La Roche Ltd

OBJECTIVES

The **primary objective** of this study is as follows:

- To assess the safety and tolerability of RO7049389 compared to placebo after single- and multiple-ascending oral doses in healthy Chinese subjects.

The **secondary objective** of this study is as follows:

- To assess the pharmacokinetics (PK) of RO7049389 after single-ascending and multiple-ascending oral doses in healthy Chinese subjects.
- To investigate metabolites (M5, M6, and M11) of RO7049389 in selected plasma samples.

STUDY DESIGN

Description of Study

Healthy subjects will be enrolled into one of 3 planned cohorts and one optional cohort. Cohort 1 will include 4 healthy subjects, with 2 randomly assigned to RO7049389 (200mg) and 2 assigned to placebo. From Cohort 2 to Cohort 4, each of the cohorts will include 12 healthy subjects with 10 subjects randomly assigned to RO7049389 and 2 subjects randomly assigned to placebo.

In Cohort 1, subjects will receive one single dose of the assigned study medication. In subsequent cohorts, subjects will receive one single dose of the assigned study medication, followed by a wash out period of approximately 1-week before entering into a MAD phase of the study. Subjects in the MAD cohorts will be from the SAD cohorts after the wash out period, or from new enrollments if there are discontinuations between the SAD phase and MAD phase for reasons other than safety.

Escalation to the next single dose cohort will be based primarily on the safety and available PK data through 24 hours post single dose in the previous cohorts. The MAD phase may start after a wash out period of approximately 1-week, providing that the safety information through 24 hours post dose in the SAD phase at the same dose level, and available safety information from MAD cohorts in the entry-into-human study (YP39364), have been reviewed.

NUMBER OF STUDY SUBJECTS

Due to the nature of this study, the actual number of subjects will be determined during the study. This study may require up to 76 subjects, including:

- About 40 healthy subjects to be enrolled in Cohort 1 of 4 subjects and Cohort 2 to Cohort 4 of about 12 subjects each.
- Up to 36 healthy subjects to be enrolled if new subjects are needed for the MAD phase, in case of non-safety related discontinuations between two periods (i.e., replacement subjects),
- For each part, up to two back-up subjects can undergo the preparatory activities in order to replace participants who withdraw from the study for non-safety reasons.

Subjects who drop out of the study for non-safety reasons may be replaced to ensure sufficient data to characterize the safety and PK profile, and to make a dose-escalation decision. Subjects who withdraw from the study due to poor tolerability, or, due to study drug-related adverse events, will not be replaced.

TARGET POPULATION

The study will be conducted in Chinese healthy, male and female subjects of non-childbearing potential, 18 to 60 years of age.

INCLUSION/EXCLUSION CRITERIA

Inclusion Criteria

Study subjects must meet the following criteria for study entry:

1. Chinese healthy male and female subjects, 18 to 60 years of age, inclusive. Healthy status is defined by absence of evidence of any active or chronic disease following a detailed medical and surgical history, a complete physical examination including vital signs, 12-lead ECG, hematology, blood chemistry, serology and urinalysis.
2. A Body Mass Index (BMI) of between 19 to 27 kg/m^2 inclusive, and a body weight of at least 45 kg.
3. Women should be of non-childbearing potential. Female subjects must be either surgically sterile (by means of hysterectomy and/or bilateral oophorectomy) or post-menopausal for at least one year (defined as amenorrhea $\geq$ 12 consecutive months without another cause, and confirmed by follicle stimulating hormone level $>$ 35 mIU/mL).
4. For men: agreement to remain abstinent (refrain from heterosexual intercourse) or use contraceptive measures, and agreement to refrain from donating sperm, as defined below:
    a. With female partners of childbearing potential, men must remain abstinent or use two methods of contraception with their partner, one of which must be a barrier method (i.e., condom), for the duration of the study and for at least 28 days after the last dose of study drug to avoid exposing the embryo. Men must refrain from donating sperm during this same period.
    b. The reliability of sexual abstinence should be evaluated in relation to the duration of the clinical trial and the preferred and usual lifestyle of the patient. Periodic abstinence (e.g., calendar, ovulation, symptothermal, or post-ovulation methods) and withdrawal are not acceptable methods of contraception.
    c. Other acceptable forms of contraception for this study include vasectomy, bilateral tubal occlusion, intrauterine device (IUD) or proper use of hormonal contraceptives.
5. Informed of, willing and able to comply with all of the protocol requirements and the investigational nature of the study, and have signed an informed consent form (ICF) in accordance with institutional and regulatory requirements.

Exclusion Criteria

Study subjects who meet any of the following criteria will be excluded from study entry:

1. Pregnant (positive pregnancy test) or lactating women, and male subjects with partners who are pregnant or lactating.
2. History or symptoms of any clinically significant gastrointestinal, renal, hepatic, broncho-pulmonary, neurological, psychiatric, cardio-vascular, endocrinological, hematological or allergic disease, metabolic disorder, cancer or cirrhosis.
3. Personal history of congenital long QT syndrome or family history of sudden death.
4. History of Gilbert's syndrome.
5. History of having received or currently receiving any systemic anti-neoplastic (including radiation) or immune-modulatory treatment (including systemic oral or inhaled corticosteroids) $\leq$ 6 months prior to the first dose of study drug or the expectation that such treatment will be needed at any time during the study.
6. Subjects who have had significant acute infection, e.g., influenza, local infection, acute

gastrointestinal symptoms or any other clinically significant illness within two weeks of dose administration.

7. Any confirmed significant allergic reactions (urticaria or anaphylaxis) against any drug, or multiple drug allergies (non-active hay fever is acceptable).
8. Any clinically significant concomitant diseases or condition that could interfere with, or treatment of which might interfere with, the conduct of the study, or that would, in the opinion of the Investigator, pose an unacceptable risk to the subject in this study.
9. Confirmed (based on the average of three separate resting blood pressure measurements, properly measured with well-maintained equipment, after at least 10 minutes rest) systolic BP greater than 140 or less than 90 mmHg, and diastolic BP greater than 90 or less than 50 mmHg at screening.
10. Clinically relevant ECG abnormalities on screening ECG, e.g.,
    a. QTcF $>$ 450 msec or $<$ 300 msec
    b. Notable resting bradycardia (HR $<$ 45 bpm), or HR $>$ 90 bpm.
    c. ECGs with documented machine errors in the interval duration assessments
    d. Evidence of atrial fibrillation, atrial flutter, complete bundle branch block, Wolf-Parkinson-White Syndrome, or cardiac pacemaker.
11. ECG with QRS and/or T-wave judged to be unfavorable for a consistently accurate QT measurement (e.g., neuromuscular artifact that cannot be readily eliminated, arrhythmias, indistinct QTS onset, low amplitude T-wave, merged T- and U-waves, prominent U-waves).
12. Creatinine clearance (CrCl) $\leq$ 70 mL/min (using the Cockcroft-Gault formula).
13. Positive test at screening of any of the following: hepatitis A (HAV IgM Ab), hepatitis B (HBsAg), hepatitis C (HCV RNA or HCV Ab) or human immunodeficiency virus 1 and 2 (HIV Ab).
14. Any other clinically significant abnormalities in laboratory test results at screening. In the case of uncertain or questionable results, tests performed during screening may be repeated before randomization to confirm eligibility.
15. Participation in an investigational drug or device study within 90 days prior to screening or more than 4 times per year.
16. Donation or loss of blood over 500 mL within 3 months prior to screening.
17. Positive test for drugs of abuse (including recreational drugs) and/or positive alcohol test at screening or on Day -1.
18. Any suspicion or history of drug and/or alcohol abuse within the last year.
19. History (within 3 months of screening) of alcohol consumption exceeding two standard drinks per day on average (1 standard drink = 10 grams of alcohol). Alcohol consumption will be prohibited at least 48 hours before screening, 48 hours before and 48 hours after each dose, and 48 hours before each scheduled visit.
20. Use of $>$ 5 cigarettes or equivalent nicotine-containing product per day.
21. Taking any prescribed or over-the-counter medications (including vitamins or herbal remedies) within 2 weeks of first dosing or within 5 times the elimination half-life of the medication prior to first dosing (whichever is longer). Occasional acetaminophen/paracetamol is allowed. Exceptions may be made on a case-by-case basis following discussion and agreement between the Investigator and the Sponsor.
22. Subjects under judicial supervision, guardianship or curatorship.
23. Medical or social conditions that would potentially interfere with the subject's ability to comply with the study visit schedule or the study assessments

LENGTH OF STUDY

The total duration of the SAD part of the study will be approximately 8 weeks (from screening through study completion) for each randomized subject as follows:

- Screening: Up to 4 weeks
- In clinic period: Days −1 to 3
- Ambulatory visits: Days 4, 5, 8
- Safety Follow-up Call: Day 29

The total duration of the MAD part of the study will be approximately 10 weeks* (from screening through study completion) for each randomized subject as follows:

- Screening: Up to 4 weeks
- In Clinic period: Day -1 to Day 16
- Safety Follow-up Visit: Day 21
- Safety Follow-up Call: Day 42

*For subjects who previously participated in the SAD part of the study, their total duration in the study will be approximately 12 weeks: up to 4 weeks screening for SAD, approximately 1 week treatment on SAD, approximately 1 week washout, and 6 weeks for MAD (no additional screening visit).

END OF STUDY

The end of the study is defined as the date when the last study subject last observation (LSLO) occurs. LSLO is expected to occur 28 days after the last dose is administered.

OUTCOME MEASURES

SAFETY OUTCOME MEASURES

The safety outcome measures for this study are as follows:

- Incidence and severity of adverse events.
- Incidence of laboratory abnormalities, based on hematology, clinical chemistry, coagulation, and urinalysis test results.
- ECGs.
- Vital signs including blood pressure (BP), pulse rate and body temperature.

PHARMACOKINETIC OUTCOME MEASURES

The pharmacokinetic outcome measures for this study are as follows:

PK parameters (SAD)

- C_{max}: Maximum observed plasma concentration
- T_{max}: Time to maximum observed plasma concentration
- $AUC_{0\text{-}last}$: Area under the plasma concentration versus time curve up to the last measurable concentration
- $AUC_{0\text{-}inf}$: Area under the plasma concentration versus time curve extrapolated to infinity
- $T_{1/2}$: Apparent terminal phase half-life
- CL/F: Apparent clearance after oral administration

PK parameters (MAD)

- C_{max}: Maximum observed plasma concentration after single dose and at steady-state
- T_{max}: Time to maximum observed plasma concentration after single-dose and at steady-state

- C_{trough}: Trough plasma concentration
- $AUC_{0-\tau}$: Area under the plasma concentration-time curve for a dosing interval after single dose and at steady-state
- $T_{1/2}$: Apparent terminal phase half-life after single-dose and at steady-state
- Accumulation index of RO7049389

PK parameters (SAD and MAD):

Plasma samples collected in the study will be qualitatively and/or quantitatively analyzed for the metabolites (M5, M6, and M11) of RO7049389 in selected samples.

INVESTIGATIONAL MEDICINAL PRODUCT(S)

The investigational medicinal product in this study are:

RO7049389 and placebo, which will be supplied and packaged by the Sponsor. [redacted] A matching-placebo, in terms of size, shape and color, has also been developed for clinical usage.

PROCEDURES

Schedules of Assessments (SoA) are provided in Appendices 1-4.

STATISTICAL METHODS

SAFETY ANALYSES

All study subjects who have received at least one dose of the study medication (RO7049389 or matching placebo), whether prematurely withdrawn from the study or not, and with at least one safety assessment, will be included in the safety analysis. The safety data, including AEs, reasons for withdrawal from study, laboratory data, ECG, concomitant medications, vital signs, and physical examination results will be listed and summarized descriptively. Laboratory test values will be presented by individual listings with flagging of values outside the normal ranges.

PHARMACOKINETIC ANALYSES

Study subjects will be excluded from the pharmacokinetic analysis population if they significantly violate the inclusion or exclusion criteria, deviate significantly from the protocol or if data are unavailable or incomplete which may influence the pharmacokinetic analysis. Excluded cases will be documented together with the reason for exclusion. All decisions on exclusions from the analysis will be made prior to database closure. Non-compartmental analysis will be employed for estimation of PK parameters. All PK parameters will be presented by individual listings and summary statistics for each cohort including means, geometric means, medians, ranges, standard deviations and coefficients of variation. Plasma samples collected in the study will be qualitatively and/or quantitatively analyzed for the presence of metabolites of RO7049389. A population PK model development and analysis may be performed. The Population PK results will be reported in a separated document.

SAMPLE SIZE JUSTIFICATION

The sample size was determined by practical considerations and not based on statistical power calculations. Approximately four subjects will be enrolled in SAD cohort 1 (2 active treatment and 2 placebo). Approximately twelve subjects will be enrolled in each of the other SAD and MAD cohorts. They will be randomized to either active treatment (10 subjects per dose level) or placebo (2 subjects per dose level).

With ten subjects on active drug per dose group or cohort, there is a 94% chance to observe at least one AE that has an incidence rate of 25% in the population.

INTERIM ANALYSIS

No interim analysis is planned for this study.

LIST OF PROHIBITED MEDICATIONS

All medications (prescription and OTC) taken within 4 weeks of study screening will be recorded on the appropriate eCRF.

As a general rule, no concomitant medication will be permitted within 14 days prior to the first dosing or within 5 half-lives of the medication prior to the first dosing (whichever is longer), until the follow-up visit, with the exception of the cases listed in the protocol. Exceptions may be made on a case-by-case basis following discussion and agreement between the Investigator and the Sponsor after the rationale for exception is discussed and clearly documented.

LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation	Definition
AE	Adverse Events
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
AUC	Area under the curve
BID	Twice daily
BP	Blood pressure
cccDNA	Covalently closed circular DNA
CHB	Chronic Hepatitis B
CL	Clearance
CL/F	Apparent clearance
CRO	Contract research organization
CSR	Clinical study report
CTCAE	Common Terminology Criteria for Adverse Events
DDI	Drug-drug interaction
DNA	Deoxyribonucleic acid
EC	Ethics Committee
ECG	Electrocardiograms
eCRF	Electronic case report form
EDC	Electronic data capture
ESF	Eligibility screening form
FDA	Food and Drug Administration
FIH	First in human
FSH	Follicle-stimulating hormone
HBeAG	Hepatitis B envelope antigen
HBsAG	Hepatitis B surface antigen
HBV	Hepatitis B virus
HCV	Hepatitis C virus
HDL	High density lipoproteins
HIV	Human immunodeficiency virus
HR	Heart rate
IB	Investigator's Brochure
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IFN	Interferon
IMP	Investigational medicinal product

IND	Investigational New Drug (application)
INR	International normalized ratio
IRB	Institutional Review Board
IUD	Intrauterine device
IV	Intravenous
LDL	Low density lipoproteins
LSLO	Last subject, last observation
LSLV	Last subject, last visit
MAD	Multiple ascending doses
NOAEL	No-observed-adverse-effect level
NUC	Nucleos(t)ide analogue
PD	Pharmacodynamic
PK	Pharmacokinetic
PT	Prothrombin time
Q12h	Twice daily, every 12 hours
Q24h	Once daily, every 24 hours
QD	Once daily
QRS	QRS complex
QT	QT interval
QTc	QT corrected for heart rate
QTcF	QT corrected for heart rate using the Fridericia correction factor
RBC	Red blood cell
RNA	Ribonucleic acid
RR	RR interval
SAD	Single ascending dose
SAE	Serious adverse event
SD	Single dose
SoA	Schedule of assessments
ULN	Upper limit of normal
V	Volume
WBC	White blood cell

1. BACKGROUND AND RATIONALE

RO7049389, an inhibitor of hepatitis B virus (HBV) capsid assembly, is being developed for the treatment of patients with chronic HBV infection.

1.1 BACKGROUND ON DISEASE

Hepatitis B virus (HBV) infection is a major cause of both acute hepatitis and chronic liver diseases, including cirrhosis and hepatocellular carcinoma. Approximately two billion people worldwide have serological evidence of past or present HBV infection. An estimated 686,000 people are estimated to die each year due to the acute or chronic consequences of hepatitis B (WHO 2016). In China, about 7% of the population (approximately 90 million) is chronically infected with HBV (Yu et al 2014; Liang et al 2009).

Chronic HBV infection can be characterized by the persistence of high levels of HBV antigens (including capsid protein, core protein, hepatitis B envelope antigen [HBeAg], and hepatitis B surface antigen [HBsAg]). Disease progression is a dynamic process with several stages, during which chronic hepatitis B may be presented either as HBeAg-positive or HBeAg-negative hepatitis.

Persistence of HBV infection is a consequence of the presence of covalently closed circular DNA (cccDNA) in the nuclei of infected hepatocytes. Despite the advancement in the understanding of HBV disease biology, eradication of cccDNA (defined as ‘disease cure’) may not be achievable for the foreseeable future. Nevertheless, the sustained clearance of HBsAg has been shown to be protective against disease progression and development of HBV complications including cirrhosis, liver failure and hepatocellular carcinoma (HCC). Accordingly, sustained HBsAg loss with or without seroconversion has been defined as ‘functional cure’, and is specified by current treatment guidelines as the ideal treatment endpoint. Achieving functional cure allows treatment cessation (EASL 2012) with low likelihood of virological relapse. Other clinically meaningful endpoints, irrespective of HBeAg status, are HBV DNA suppression and alanine aminotransferase (ALT) normalization, which indicate the virological and biochemical responses to therapies, respectively. For HBeAg-positive patients, HBeAg seroconversion is indicative of a better prognosis, including lower rates of cirrhosis and slower disease progression. For both HBeAg-positive and –negative chronic hepatitis B (CHB), sustained suppression of HBV replication is associated with biochemical remission, histological improvement and delayed disease progression. However, virological relapse is a major limitation of the currently approved therapies as they rarely result in functional cure (HBsAg loss).

Currently, there are two classes of drugs available for the treatment of CHB: subcutaneously-administered interferon (IFN) preparations, and orally-administered nucleos(t)ide analogues (NUCs). Although both types of treatment can induce the loss of HBV envelope antigen with development of anti-HBeAg antibody (serological response),

the suppression of HBV DNA to an undetectable level by sensitive polymerase chain reaction (PCR) methods (virological response), and are able to normalize liver transaminases levels (biochemical response), neither treatment achieves a high rate of functional cure. HBsAg loss only occurs in approximately 3% of patients after one year of treatment, and < 15% after one to five years' follow-up (EASL 2012). In addition, IFN-based therapies are associated with many side-effects, while NUCs frequently require prolonged or possibly life-long therapy, and some are associated with a high risk of viral resistance.

1.2 BACKGROUND ON RO7049389

RO7049389 is an inhibitor of HBV capsid assembly and belongs to the well-studied class of heteroaryldihydropyrimidine (HAP) compounds. This class of compounds induces formation of abnormal HBV core protein aggregates, which are subsequently depleted. Depleting functional core protein results in interruption of viral assembly and inhibition of HBV replication.

The HBV core protein is involved in multiple steps of the viral life cycle such as encapsidation of pre-genomic ribonucleic acid (pgRNA), and subsequent initiation of reverse transcription. It is also an important component of the cccDNA mini chromosome. Furthermore, it has been suggested that the HBV core protein may play a role in suppressing host innate immune responses (Twu et al 1998; Fernandez et al 2003; Gruffaz et al 2013). Depletion of functional core protein may therefore facilitate host immune restoration. RO7049389 can therefore potentially provide anti-HBV benefits by both direct inhibition of viral replication and augmentation of host immune responses against the virus.

See the RO7049389 Investigator's Brochure for more details on non-clinical and clinical studies.

1.2.1 Previous Non-Clinical Studies

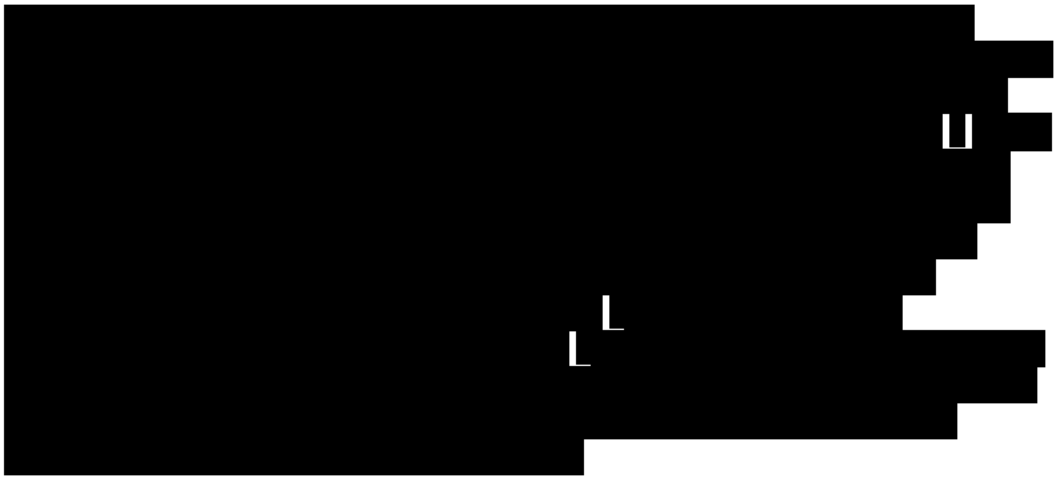
RO7049389 has shown potent antiviral activity through the induction of HBV core protein misassembly and subsequent degradation, and has a high degree of selectivity against HBV. RO7049389 demonstrated activity against most prevalent HBV genotypes (A, B, C, D) and against a panel of nucleos(t)ide analogue-resistant HBV variants tested in vitro.

[redacted]

RO7049389—F. Hoffmann-La Roche Ltd
24/Protocol YP39406, Version 4

RO7049389—F. Hoffmann-La Roche Ltd
25/Protocol YP39406, Version 4

[redacted]



RO7049389 had no effect on neurobehavioral and respiratory function in rats [redacted]

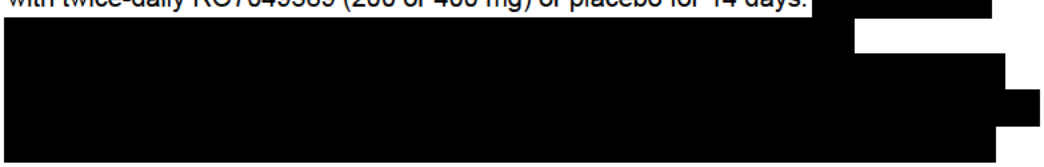
An in vitro study indicated that RO7049389 had no phototoxic potential.

See the RO7049389 Investigator's Brochure for further details on non-clinical studies.

1.2.2 Previous Clinical Studies

The first-in-human (FIH) study with RO7049389 is ongoing. YP39364, a randomized, Sponsor-open, Investigator-blinded, subject-blinded, placebo-controlled study to investigate the safety, tolerability, pharmacokinetics and pharmacodynamics of RO7049389 in: (1) single- (with or without food) and multiple (with microdosing midazolam) ascending doses in healthy volunteers; (2) patients chronically infected with hepatitis B virus.

As of the data cutoff of 2 June 2017, preliminary PK analysis was performed using data from 59 healthy volunteers treated across five SAD cohorts (n=33) and three multiple ascending–dose (MAD) cohorts (n=26). Subjects in the SAD cohorts received a single dose of RO7049389 (150–2000 mg) or placebo; those in the MAD cohorts were treated with twice-daily RO7049389 (200 or 400 mg) or placebo for 14 days. [redacted]



[redacted]

Preliminary blinded safety review was performed in Study YP39364 for 59 healthy volunteers. Thirty-three subjects in five SAD cohorts (150, 450, 1000, 2000, and 1000 mg repeated cohort) received a single dose of RO7049389 or placebo, and 26 subjects in three MAD cohorts (200 mg fasted, 200 mg fed, 400 mg fed) received RO7049389 or placebo BID for 14 days. This preliminary safety review showed that both the single doses and BID dosing were well tolerated in all subjects. [redacted]

See the RO7049389 Investigator's Brochure for further details on clinical studies.

1.3 STUDY RATIONALE AND BENEFIT–RISK ASSESSMENT

1.3.1 Study Rationale

Study YP39406 is designed to assess the safety, tolerability and pharmacokinetics (PK) of single-ascending (SAD) and multiple-ascending doses (MAD) of RO7049389 in healthy Chinese subjects. The study leverages data generated in the FIH study YP39364, allowing targeted evaluation of fewer doses. The data generated from this study will help to evaluate potential ethnic differences between Chinese and non-Chinese subjects with regards to safety, tolerability and pharmacokinetic characteristics

that will provide guidance on future clinical studies and optimize the benefit-risk in Chinese HBV patients.

1.3.2 Benefit/ Risk Assessment

As this is a Phase I study in healthy subjects, no therapeutic benefit is anticipated for these subjects. However, this study will be essential in the development of new treatments for chronic HBV infection in populations of Asian descent, and for selecting the most appropriate dose for a proof-of--concept study in such patients.

The evaluation of the potential risks of treatment and the specific tests, observations, and precautions required for clinical studies with RO7049389 are based on information from non-clinical toxicology and safety pharmacology studies. Furthermore, this study leverages data generated in the ongoing FIH study YP39364. Safety and tolerability will be carefully assessed, and study subjects will be closely monitored. Section 6 of the Investigator's Brochure (Guidance to the Investigator) summarizes key risk management activities to consider when dosing this novel compound.

Based on (i) the calculation of a safe starting-dose for the first-in-human study as recommended in the Food and Drug Administration (FDA) guidance (FDA 2005), and on (ii) exposure multiple calculations, the selected starting-dose of 200 mg is considered to be safe (please refer to Section 3.1.2 for details). The target exposure range of RO7049389 proposed within this study has been shown to be safe and well-tolerated in pre-clinical studies, and under a controlled setting, should provide a better understanding of clinical safety and tolerability to guide future clinical studies. All dose-levels to be included in this study will be within the dose-range which will have been considered to be safe and well-tolerated in the FIH study YP39364. The risks for an individual study subject due to ascending doses of RO7049389 or study-related procedures are considered to be minimal. The starting-dose level of 200 mg may be adjusted based on the available data from FIH study YP39364.

2. OBJECTIVES

2.1 PRIMARY OBJECTIVE

The primary objective of this study is as follows:

- To assess the safety and tolerability of RO7049389 compared to placebo after single- and multiple-ascending oral doses in healthy Chinese subjects.

2.2 SECONDARY OBJECTIVE

The secondary objective of this study is:

- To assess the pharmacokinetics (PK) of RO7049389 after single-ascending and multiple-ascending oral doses in healthy Chinese subjects.
- To investigate metabolites (M5, M6, and M11) of RO7049389 in selected plasma samples.

3. STUDY DESIGN

3.1 DESCRIPTION OF STUDY

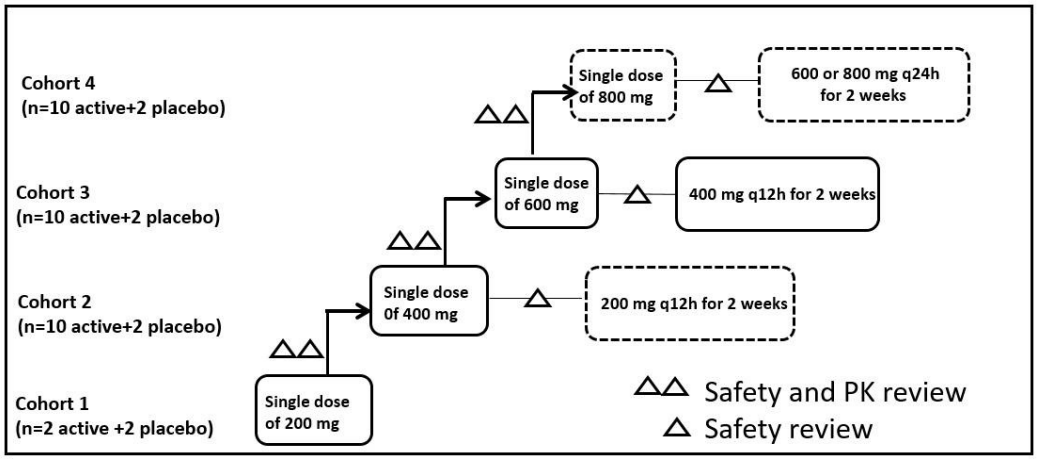
3.1.1 Overview of Study Design

This study will leverage the data which will have been generated in the FIH study, allowing targeted evaluation of fewer doses.

Healthy subjects will be enrolled into one of 3 planned cohorts and one optional cohort. Cohort 1 will include 4 healthy subjects, with 2 randomly assigned to RO7049389 and 2 assigned to placebo. From Cohort 2 to Cohort 4, each of the cohorts will include 12 healthy subjects with 10 subjects randomly assigned to RO7049389 and 2 subjects randomly assigned to placebo.

In Cohort 1, subjects will receive one single dose of the assigned study medication. In subsequent cohorts, subjects will receive one single dose of the assigned study medication, followed by a wash-out period of approximately 1-week before entering into a MAD phase of the study. Subjects in the MAD cohorts will be from the SAD cohorts after the wash-out period (see Figure 1), or from new enrollments if there are discontinuations between the SAD phase and MAD phase for reasons other than safety.

Figure 1 Overall Study Design and Dose-escalation



Dotted lines indicate optional cohorts. See Table 1 for additional information.

Escalation to the next single dose cohort will be based primarily on the safety and available PK data through 24 hours post-single dose in the previous cohorts. The MAD phase may start after a wash-out period of approximately 1-week, providing that the safety information through 24 hours post-dose in the SAD phase at the same *cohort*, and available safety information in the entry-into-human study (YP39364), have been reviewed.

The projected dose levels, which may be adjusted based on the emerging safety, tolerability, and PK data from this study and study YP39364 (entry into human), are shown in Table 1.

Table 1 Initial Dose of RO7049389 or Placebo

	Cohort 1	Cohort 2	Cohort 3	Cohort 4[a]
SAD	200 mg	400 mg	600 mg	800 mg
MAD	N/A	200 mg q12h[b,c]	400 mg q12h	600 or 800 mg q24h

a. Cohort 4 is optional and will be conducted in case that the total daily dose of the estimated efficacious dose from YP39364 is between 400 mg to 600 mg q12h or higher.
b. The dose of 200 mg q12h in MAD cohort 2 will be optional and will be conducted in case the total daily dose of the estimated efficacious dose from YP39364 is lower than 400 mg q12h.

The rationale for dose selection is described in Section 3.2.1.

The highest dose level and exposure to be included in this study will be within the dose and exposure that has been demonstrated to be safe and well tolerated in FIH Study YP39364.

3.1.1.1 SAD Phase

All subjects will be admitted to a clinical research unit for 3 nights from Day -1 through Day 2, and discharged on Day 3. On Day 1, subjects will be administered a single dose of either RO7049389 or placebo. Subjects who will not enter the MAD phase (Cohort 1) will return for a follow-up visit on Day 8 (±1), and will receive a follow-up call on Day 29.

3.1.1.2 MAD Phase

In Cohorts 2, 3 and 4, after a post-SAD phase wash-out period of at least one week, all subjects will be admitted to a clinical research unit for 16 nights from Day -1 to Day 16 (discharged on Day 16). Subjects will be administered multiple oral doses of either RO7049389 or placebo twice daily (q12h) or once daily (q24h) from Day 1 to Day 14 (the currently planned regimen is q12h, but will be evaluated based on emerging data from Study YP39364). The allocation of RO7049389 or placebo will be the same as that which was administered in the SAD phase. All subjects will return for a follow-up visit on Day 21 (±1), and will receive a follow-up call on Day 42.

3.1.1.3 Dietary Requirements

In the SAD phase, all healthy volunteers will receive either RO7049389 or placebo in the fasted state (overnight at least 8 hours). Subjects will not eat for at least 4 hours after the dose is administered. Lunch will be served approximately 4-5 hours post-morning-dose. In the MAD phase (except cohort 4, which will be in the fasted state), a standard breakfast or dinner will be served to healthy volunteers 30 minutes prior to dosing, which should be consumed within 30 minutes. RO7049389 or placebo will then be administered. Lunch will be served approximately 4-5 hours

post-morning-dose. An optional evening snack may be allowed provided that the fasting period is fulfilled before the next morning assessments.

Water will be allowed ad libitum until one hour prior to dosing and after one hour post dosing.

3.1.2 Dose-Escalation Decision Criteria

A Safety Review of the data will be conducted by the Principal Investigator, the Medical Monitor, and the Sponsor Clinical Team prior to each RO7049389 dose-escalation.

Escalation to the next dose cohort will be based primarily on the safety and tolerability information (including adverse events [AEs], electrocardiograms [ECGs], vital signs, clinical laboratory test results) up to 48 hours post-dose (SAD), and secondarily on all available PK data up to 24 hours post-dose (SAD) in a minimum of 8 subjects (receiving active study drug or placebo) within the previous cohort. Doses will be escalated by three-fold increments but for higher doses the increments will be smaller than three-fold. The decision to escalate the dose will be made by mutual agreement between the Sponsor and the Investigator.

The MAD phase (Cohorts 2, 3, and 4) can begin after a washout period of at least one week, and once safety and tolerability information through 24 hours post-dose in the SAD phase (see Figure 1), and available safety and tolerability information from MAD cohorts in the entry-into-human study (YP39364) have been reviewed. Additionally, both the Sponsor and the Investigator must agree that the safety data can support continuing with the multiple-dose phase.

3.1.2.1 Dose-Escalation Stopping Rules

Any serious adverse event considered related to study drug will lead to an immediate halt of study drug dosing, dose escalation and enrollment of new subjects until a thorough investigation has been conducted by the Sponsor's Clinical Team and the Principal Investigator. The sponsor or designee will notify all investigators and IRBs/EC, if required (see Section 5.3), regarding the outcome of such investigation. The study will only continue if agreed by the IRBs/EC.

An internal safety review of the unblinded data by Sponsor representatives from Translational Medicine, Drug Safety, Clinical Pharmacology and Biostatistics will be performed prior to each RO7049389 dose-escalation.

Planned dose-escalation will not be implemented if one of the following circumstances occurs in subjects treated with RO7049389, unless it is obvious that the occurrence is not related to the administration of study medication:

- If at any dose level, more than two subjects on active drug experience:
  - Severe and clinically significant (as defined by the Investigator) RO7049389-related AEs of the same character.

  - Clinically significant RO7049389-related laboratory abnormalities of the same character (e.g., ALT or AST $>3\times$ ULN, and total bilirubin $>2\times$ ULN).
  - Clinically significant RO7049389-related changes in vital signs or ECGs of the same character (e.g., QTcF $>$ 500 msec, or > 60 msec longer than the pre-dose baseline, within the first 48 hours post-dose).
- It is predicted that further dose-escalation will not result in a further increase in plasma exposure.

[redacted]

- The proposed dose *or exposure* exceeds those previously evaluated in the study YP39364.
- Other findings (regardless of the incidence rates) that at the joint discretion of the Sponsor and the Investigator indicate dose-escalation should be halted.

If dose-escalation is stopped, lower doses within the tolerated dose range could be investigated, or a dose repeated in the subsequent cohorts by mutual agreement between the Sponsor and Investigator, in order to increase the amount of data within this tolerated dose range.

For an individual subject, dosing should not continue if there are:

- Clinically significant RO7049389-related changes in safety parameters that are considered not acceptable by the Investigator and/or the Sponsor; or
- Poor gastrointestinal (GI) tolerability that is considered to affect the subject's wellbeing and/or the PK evaluation.

Due to the exploratory nature of this clinical study, its conduct can be discontinued at any time at the discretion of the Sponsor. This will not constitute a premature termination of the study.

3.1.3 End of Study

The end of the study is defined as the date when the last study subject last observation (LSLO) occurs. LSLO is expected to occur 28 days after the last dose is administered.

3.2 RATIONALE FOR STUDY DESIGN

The rationale for the design of study YP39406 is given in Section 1.3.1.

3.2.1 Rationale for Dosage Selection

The first SAD cohort will receive 200 mg (~1/4 of the total projected clinically efficacious dose). This starting-dose is considered to have been safe and well tolerated in the FIH Study YP39364.

[redacted]

[redacted]

Furthermore, the starting-dose level of 200 mg is considered to be safe based on FDA guidance. [redacted] Using the lowest HED of 40 mg/kg [redacted] and a safety factor of 10, the maximum recommended starting-dose (MRSD) for a 60 kg individual is 240 mg. The starting-dose of 200 mg proposed in this study is approximately 1.2-fold lower than the MRSD.

Subsequent dose-levels may be adjusted based on the PK, tolerability and safety results from FIH Study YP39364 and available data from current Study YP39406. The highest dose-level *and exposure* to be included in this study will be within the dose range *and exposure* which has been demonstrated to be safe and well tolerated in FIH Study YP39364.

3.2.2 Rationale for Study Population

The study will be conducted in Chinese healthy, male and female subjects of non-childbearing potential, 18 to 60 years of age. The absence of confounding diseases and co-medications in healthy subjects allows for a clearer and more consistent assessment of drug disposition and safety profile.

The data generated from this study will help to evaluate potential ethnic differences between Chinese and non-Chinese subjects with regards to safety, tolerability and pharmacokinetic characteristics that will provide guidance on future clinical studies and optimize the benefit-risk in Chinese HBV patients.

3.3 OUTCOME MEASURES

3.3.1 Safety Outcome Measures

The safety outcome measures for this study are as follows:

- Incidence and severity of adverse events.
- Incidence of laboratory abnormalities, based on hematology, clinical chemistry, coagulation, and urinalysis test results.
- ECGs.
- Vital signs including blood pressure (BP), pulse rate and body temperature.

3.3.2 Pharmacokinetic Outcome Measures

Plasma concentrations will be measured by specific validated methods. Pharmacokinetic (PK) parameters will be estimated using standard non-compartmental methods for RO7049389.

The pharmacokinetic outcome measures for this study are as follows:

PK parameters (SAD):

- C_{max}: Maximum observed plasma concentration
- T_{max}: Time to maximum observed plasma concentration
- $AUC_{0\text{-}last}$: Area under the plasma concentration versus time curve up to the last measurable concentration
- $AUC_{0\text{-}inf}$: Area under the plasma concentration versus time curve extrapolated to infinity
- $T_{1/2}$: Apparent terminal phase half-life
- CL/F: Apparent clearance after oral administration

PK parameters (MAD):

- C_{max}: Maximum observed plasma concentration after single-dose and at steady-state
- T_{max}: Time to maximum observed plasma concentration after single-dose and at steady-state
- C_{trough}: Trough plasma concentration
- $AUC_{0\text{-}\tau}$: Area under the plasma concentration-time curve for a dosing interval after single-dose and at steady-state
- $T_{1/2}$: Apparent terminal phase half-life after single-dose and at steady-state
- Accumulation index of RO7049389

PK parameters (SAD and MAD):

- Plasma samples collected in the study will be qualitatively and/or quantitatively analyzed for the metabolites (M5, M6, and M11) of RO7049389 in selected samples.

4. MATERIALS AND METHODS

4.1 STUDY POPULATION

Due to the nature of this study, the actual number of subjects will be determined during the study. This study may require up to 76 subjects, including:

- About 40 healthy subjects to be enrolled in Cohort 1 of 4 subjects and Cohort 2 to Cohort 4 of about 12 subjects each.

- Up to 36 healthy subjects to be enrolled if new subjects are needed for the MAD phase, in case of non-safety related discontinuations between two periods (i.e., replacement subjects),
- For each part, up to two back-up subjects can undergo the preparatory activities in order to replace participants who withdraw from the study for non-safety reasons.

Subjects who drop out of the study for non-safety reasons may be replaced to ensure sufficient data to characterize the safety and PK profile, and to make a dose-escalation decision. Subjects who withdraw from the study due to poor tolerability, or, due to study drug-related adverse events, will not be replaced.

4.1.1 Recruitment Procedures

Study subjects will be identified for potential recruitment using pre-screening enrollment logs, IEC/IRB approved newspaper/radio advertisements and mailing lists prior to consenting to take place on this study.

4.1.2 Inclusion Criteria

Study subjects must meet the following criteria for study entry:

1. Chinese healthy male and female subjects, 18 to 60 years of age, inclusive. Healthy status is defined by absence of evidence of any active or chronic disease following a detailed medical and surgical history, a complete physical examination including vital signs, 12-lead ECG, hematology, blood chemistry, serology and urinalysis.
2. A Body Mass Index (BMI) of between 19 to 27 kg/m^2 inclusive, and a body weight of at least 45 kg.
3. Women should be of non-childbearing potential. Female subjects must be either surgically sterile (by means of hysterectomy and/or bilateral oophorectomy) or post-menopausal for at least one year (defined as amenorrhea $\geq$ 12 consecutive months without another cause, and confirmed by follicle stimulating hormone level $>$ 35 mIU/mL).
4. For men: agreement to remain abstinent (refrain from heterosexual intercourse) or use contraceptive measures, and agreement to refrain from donating sperm, as defined below:

a) With female partners of childbearing potential, men must remain abstinent or use two methods of contraception with their partner, one of which must be a barrier method (i.e., condom), for the duration of the study and for at least 28 days after the last dose of study drug to avoid exposing the embryo. Men must refrain from donating sperm during this same period.

b) The reliability of sexual abstinence should be evaluated in relation to the duration of the clinical trial and the preferred and usual lifestyle of the patient. Periodic abstinence (e.g., calendar, ovulation, symptothermal, or post-ovulation methods) and withdrawal are not acceptable methods of contraception.

c) Other acceptable forms of contraception for this study include vasectomy, bilateral tubal occlusion, intrauterine device (IUD) or proper use of hormonal contraceptives.

5. Informed of, willing and able to comply with all of the protocol requirements and the investigational nature of the study, and have signed an informed consent form (ICF) in accordance with institutional and regulatory requirements.

4.1.3 Exclusion Criteria

Study subjects who meet any of the following criteria will be excluded from study entry:

1. Pregnant (positive pregnancy test) or lactating women, and male subjects with partners who are pregnant or lactating.
2. History or symptoms of any clinically significant gastrointestinal, renal, hepatic, broncho-pulmonary, neurological, psychiatric, cardio-vascular, endocrinological, hematological or allergic disease, metabolic disorder, cancer or cirrhosis.
3. Personal history of congenital long QT syndrome or family history of sudden death.
4. History of Gilbert's syndrome.
5. History of having received or currently receiving any systemic anti-neoplastic (including radiation) or immune-modulatory treatment (including systemic oral or inhaled corticosteroids) $\leq$ 6 months prior to the first dose of study drug or the expectation that such treatment will be needed at any time during the study.
6. Subjects who have had significant acute infection, e.g., influenza, local infection, acute gastrointestinal symptoms or any other clinically significant illness within two weeks of dose administration.
7. Any confirmed significant allergic reactions (urticaria or anaphylaxis) against any drug, or multiple drug allergies (non-active hay fever is acceptable).
8. Any clinically significant concomitant diseases or condition that could interfere with, or treatment of which might interfere with, the conduct of the study, or that would, in the opinion of the Investigator, pose an unacceptable risk to the subject in this study.
9. Confirmed (based on the average of three separate resting blood pressure measurements, properly measured with well-maintained equipment, after at least 10 minutes rest) systolic BP greater than 140 or less than 90 mmHg, and diastolic BP greater than 90 or less than 50 mmHg at screening.
10. Clinically relevant ECG abnormalities on screening ECG, e.g.,

a) QTcF $>$ 450 msec or $<$ 300 msec

b) Notable resting bradycardia (HR $<$ 45 bpm), or HR $>$ 90 bpm.

c) ECGs with documented machine errors in the interval duration assessments

d) Evidence of atrial fibrillation, atrial flutter, complete bundle branch block, Wolf-Parkinson-White Syndrome, or cardiac pacemaker.

11. ECG with QRS and/or T-wave judged to be unfavorable for a consistently accurate QT measurement (e.g., neuromuscular artifact that cannot be readily eliminated, arrhythmias, indistinct QTS onset, low amplitude T-wave, merged T- and U-waves, prominent U-waves).
12. Creatinine clearance (CrCl) $\leq$ 70 mL/min (using the Cockcroft-Gault formula).

13. Positive test at screening of any of the following: hepatitis A (HAV IgM Ab), hepatitis B (HBsAg), hepatitis C (HCV RNA or HCV Ab) or human immunodeficiency virus 1 and 2 (HIV Ab).
14. Any other clinically significant abnormalities in laboratory test results at screening. In the case of uncertain or questionable results, tests performed during screening may be repeated before randomization to confirm eligibility.
15. Participation in an investigational drug or device study within 90 days prior to screening or more than 4 times per year.
16. Donation or loss of blood over 500 mL within 3 months prior to screening.
17. Positive test for drugs of abuse (including recreational drugs) and/or positive alcohol test at screening or on Day -1.
18. Any suspicion or history of drug and/or alcohol abuse within the last year.
19. History (within 3 months of screening) of alcohol consumption exceeding two standard drinks per day on average (1 standard drink = 10 grams of alcohol). Alcohol consumption will be prohibited at least 48 hours before screening, 48 hours before and 48 hours after each dose, and 48 hours before each scheduled visit.
20. Use of > 5 cigarettes or equivalent nicotine-containing product per day.
21. Taking any prescribed or over-the-counter medications (including vitamins or herbal remedies) within 2 weeks of first dosing or within 5 times the elimination half-life of the medication prior to first dosing (whichever is longer). Occasional acetaminophen/paracetamol is allowed. Exceptions may be made on a case-by-case basis following discussion and agreement between the Investigator and the Sponsor.
22. Subjects under judicial supervision, guardianship or curatorship.
23. Medical or social conditions that would potentially interfere with the subject's ability to comply with the study visit schedule or the study assessments.

4.2 METHOD OF TREATMENT ASSIGNMENT AND BLINDING

4.2.1 Method of Blinding

Randomization numbers will be generated by the Sponsor or its designee.

The study is Investigator-blinded and Subject-blinded. In order to minimize bias in reporting, collecting, and interpreting safety data. Study subjects, Investigator(s), and all individuals in direct contact with study subjects at the investigative site will remain blinded until the completion of the study, except the pharmacist handling the study drug distribution.

The study is Sponsor-open. At the Sponsor side, the Clinical Pharmacologists, the Study Pharmacometrician, the Study Clinical Pharmacology Scientist, the Study Biostatistician, the Statistical Programmer/Data Acquisition Specialist/Clinical Data Programmer, and the individual responsible for PK sample bioanalysis – members of the Sponsor's project and study teams who do not have direct contact with the subjects – will be unblinded to

treatment assignment. Other members of the Sponsor staff may be unblinded at the Clinical Pharmacologist's discretion, if this is considered necessary to optimize data analysis and dose-escalation decision making.

Before each of the dose-escalation decisions, the individual safety, tolerability, and available PK data will be reviewed in a blinded fashion by the Investigator and the Sponsor's clinical team during the Safety Review Meeting. The Investigator may also be unblinded in mutual agreement with the Clinical Pharmacologist if treatment assignment knowledge is considered critical to allow study progression, and/or is deemed important to define future risk management activities. Unblinding can occur as soon as the Investigator(s) and the responsible Roche scientists have reviewed the data, even if the data have not been entered onto the Case Report Forms (CRFs). If required, unblinded data (individual as well as at group level) may also be presented to the Drug Safety Committee or other experts of the Sponsor.

As per Health Authority reporting requirements, the Sponsor will break the treatment code for all unexpected serious adverse events (SAE; see Section 5.2.1) that are considered by the Investigator to be related to study drug.

All such occurrences should be documented in the study file. Treatment codes should not be broken except in emergency situations and, if possible, the responsible scientific leader should be contacted before the code is opened. At the final monitoring visit, the unused treatment codes will be counted and checked and a statement to the effect that all are intact (or not as the case may be) will be made by the monitor; this statement will be included or referred to in the final study report. All treatment codes will be returned to Roche.

Whenever disclosure of the identity of the test medication is necessary, adequate procedures will be in place to ensure integrity of the data. Any unblinding will be documented in the study report with date, reason for identifying the drug and the name(s) and role(s) in the study of the person(s) unblinded.

4.3 STUDY TREATMENT

4.3.1 Formulation, Packaging, and Handling

4.3.1.1 RO7049389 and Placebo

RO7049389 and matching placebo (IMP) to be used in the study will be provided by Roche.

[redacted]

[redacted] A matching-placebo, in terms of size, shape and color, has also been developed for clinical usage. [redacted]

Film-coated tablets of RO7049389 should be stored under the recommended storage conditions “Store at 2-8°C, protect from light and moisture”. For information on the shelf-life of RO7049389 film-coated tablets and the respective placebos, see the packaging.

Study drug packaging will be overseen by the Roche clinical trial supplies department and bear a label with the identification required by local law, the protocol number, drug identification and dosage. The packaging and labeling of the study drug will be in accordance with Roche standard and local regulations.

Upon arrival of investigational products at the site, site personnel should check them for damage and verify proper identity, quantity, integrity of seals and temperature conditions, and report any deviations or product complaints to the Monitor upon discovery.

For further details, see the Investigator’s Brochure.

4.3.2 Dosage, Administration and Compliance

4.3.2.1 RO7049389 and Placebo

The health care professionals at the study site administering the study drug to the subject must check to be sure that the study drug is swallowed.

The qualified pharmacist at the pharmacy will keep drug accountability and record the study drug batch or lot number received by each subject during the study.

Accountability and subject compliance will be assessed by maintaining adequate study drug dispensing records. The Investigator is responsible for ensuring that dosing is administered in compliance with the protocol. Delegation of this task must be clearly documented and approved by the Investigator.

Active or placebo doses will be administered orally to the study subjects with approximately 240 mL of still water at room temperature. An additional amount of water up to 100 mL can be given to assist dose administration only if needed. The dose in mg, the date and time of dosing, and the exact volume of water should be recorded on the eCRF (the exact volume of water consumed during dose administration will be recorded during the in-clinic period). [redacted]

The qualified individual responsible for dispensing the study drug will prepare the correct dose according to the randomization schedule. This individual will write the date dispensed and study subject's number and initials on the study drug vial label and on the Drug Accountability Record. This individual will also record the study drug batch or lot number received by each study subject during the study.

Guidelines for dosage modification and treatment interruption or discontinuation are provided in Section 3.1.2.

SAD Phase

RO7049389 or matching-placebo will be administered orally with 240 mL of water to the healthy subjects by investigational staff in the morning of Day 1 after an overnight fast of at least 8 hours.

Healthy volunteers will not eat for at least 4 hours after the dose is administered. Water will be allowed until one hour prior to dosing, and from one hour post-dosing. Approximately 4-5 hours after dosing, healthy subjects will be served lunch.

MAD Phase

RO7049389 or matching-placebo will be administered orally with 240 mL of water to the healthy subjects by investigational staff from Day 1 through to Day 13 q12h, and on Day 14 once in the morning.

Except for cohort 4, which will be in the fasted state, a standard breakfast or dinner will be served to healthy volunteers 30 minutes prior to dosing, which should be consumed within 30 minutes. RO7049389 or placebo will then be administered.

Lunch will be served approximately 4-5 hours post-morning-dose. The second daily dose will be administered in the evening, approximately 12 hours after the morning dose.

Timing of meals relative to dosing may be changed according to emerging data.

Guidelines for dietary stipulations are provided in Section 4.4.3 and Section 4.4.4.

4.3.3 Investigational Medicinal Product Accountability

All investigational medicinal products (IMPs) required for completion of this study (RO7049389 and matching-placebo) will be provided by the Sponsor. The investigational site will acknowledge receipt of IMPs, to confirm the shipment condition and content. Any damaged shipments will be replaced.

The Investigator is responsible for the control of drugs under investigation. Adequate records of the receipt (e.g., Drug Receipt Record) and disposition (e.g., Drug Dispensing

Log) of the study drug must be maintained. The Drug Dispensing Log must be kept current and should contain the following information:

- The identification of the study subject to whom the study drug was dispensed (for example, study subject initials and date of birth).
- The date(s), quantity of the study drug dispensed to the study subject.
- The identification of the person who dispensed the drug.
- All records and drug supplies must be available for inspection by the Clinical Trial Monitor at every monitoring visit.

IMPs will either be disposed of at the study site according to the study site's institutional standard operating procedure or returned to the Sponsor with the appropriate documentation. The site's method of IMP destruction must be agreed upon by the Sponsor. Local or institutional regulations may require immediate destruction of used investigational medicinal product for safety reasons. In these cases, it may be acceptable for investigational study site staff to destroy dispensed investigational product before a monitoring inspection provided that source document verification is performed on the remaining inventory and reconciled against the documentation of quantity shipped, dispensed, returned, destroyed and provided that adequate storage and integrity of drug has been confirmed.

The site must obtain written authorization from the Sponsor before any IMP is destroyed, and IMP destruction must be documented on the appropriate form.

Written documentation of destruction must contain the following:

- Identity of investigational product(s) destroyed
- Quantity of investigational product(s) destroyed
- Date of destruction
- Method of destruction
- Name and signature of responsible person [or company] who destroyed investigational product[s]

Accurate records of all IMPs received at, dispensed from, returned to, and disposed of by the study site should be recorded on the Drug Inventory Log.

4.4 CONCOMITANT THERAPY AND FOOD

4.4.1 Permitted Therapy

Concomitant therapy includes any medication, e.g., prescription drugs, OTC drugs, approved dietary and herbal supplements, nutritional supplements and any non-medication interventions (e.g., individual psychotherapy, cognitive behavioral therapy, smoking cessation therapy, and rehabilitative therapy) used by a subject from 4 weeks prior to the first dosing through the study completion. All concomitant

medications should be reported to the Investigator and recorded on the Concomitant Medications eCRF.

As a general rule, no other concomitant medication will be permitted within 14 days prior to the first dosing or within 5 half-lives of the medication prior to the first dosing (whichever is longer), until the follow-up visit, unless the rationale for use is discussed between the Investigator and Sponsor and is clearly documented.

The following medications are exceptions:

- Medications used to treat AEs may only be prescribed after consultation with the Sponsor (with the exception of acetaminophen/paracetamol), unless there is a medical need to ensure the well-being of the subject that should not be delayed. All therapy and/or medication administered to manage adverse events should be recorded on the Adverse Event eCRF.
- Hormone replacement therapy (HRT): continue using if initiated at least 2 months prior to study start.
- Occasional use of acetaminophen (up to 2g per day, not to exceed 3g/5 days) or ibuprofen (up to 400 mg per day) will be permitted. During the period of confinement to the clinical research unit, subjects will be restricted from the use of acetaminophen/paracetamol and other non-prescription medications beginning 4 hours prior to dosing through 4 hours after dosing unless deemed necessary to treat an Adverse Event (AE) by the Investigator.

4.4.2 Prohibited Therapy

All medications (prescription and OTC) taken within 4 weeks of study screening will be recorded on the appropriate eCRF.

As a general rule, no concomitant medication will be permitted within 14 days prior to the first dosing or within 5 half-lives of the medication prior to the first dosing (whichever is longer), until the follow-up visit, with the exception of the cases listed in Section 4.4.1. Exceptions may be made on a case-by-case basis following discussion and agreement between the Investigator and the Sponsor after the rationale for exception is discussed and clearly documented.

4.4.3 Prohibited Food

Use of the following is prohibited:

- Alcohol must not be consumed from 48 hours before screening, 48 hours before admission until completion of follow-up visit.
- [redacted]

4.4.4 Dietary and Special Requirements

The following requirements will also be applied:

- Laboratory safety assessments should be conducted after study subjects have been fasted for a minimum of 8 hours (4h for screening and follow-up).
- The excessive consumption of fluids (greater than 3 liters per day) should be avoided.
- On the days of a study visit caffeine (i.e., beverage including green tea, chocolate or supplements) should not be consumed from 48 hours prior to study drug administration until discharge from the clinic.
- No strenuous exercise is permitted during the study from 96 hours before admission until completion of the follow-up visit.
- The use of tobacco is not permitted during the study.

Meals will be similar in composition and time of administration across all cohorts during the in-house period.

4.5 STUDY ASSESSMENTS

4.5.1 Description of Study Assessments

All examinations listed below will be performed according to the schedule of assessments outlined in Appendix 1, Appendix 2, Appendix 3, and Appendix 4.

All time-points when several assessments coincide, the following sequence should be followed with the PK blood sample to be taken at the nominal time-point:

- Urine collection
- ECG recordings
- Vital signs
- PK, safety blood sampling
- Study drug administration

4.5.1.1 Medical History and Demographic Data

Medical history includes clinically significant diseases, reproductive status, smoking history, use of alcohol and drugs of abuse, and all medications (e.g., prescription drugs, OTC drugs, herbal or homeopathic remedies, nutritional supplements) used by the study subject within 30 days prior to the screening visit.

Demographic data will include age, sex, and self‑reported race/ethnicity.

4.5.1.2 Physical Examinations

A complete physical examination should include an evaluation of the head, eyes, ears, nose, throat, neck and lymph nodes, and the cardiovascular, dermatological, musculoskeletal, respiratory, gastrointestinal, genitourinary, and neurological systems. A

genitourinary examination may be performed in case of evocative symptoms at the Investigator discretion.

Any abnormality identified at baseline should be recorded on the General Medical History and Baseline Conditions eCRF.

At subsequent visits (or as clinically indicated), limited, symptom-directed physical examinations should be performed. Changes from baseline abnormalities should be recorded in study subject's notes. New or worsened clinically significant abnormalities should be recorded as adverse events on the Adverse Event eCRF.

Body weight will be recorded at screening and during study. Height will only be recorded at screening. BMI will be calculated at screening.

4.5.1.3 Vital Signs

Blood pressure (BP), pulse rate, and body temperature (tympanic or oral) will be recorded at the time-points specified in Overall Schedule of Assessments (Appendix 1 and Appendix 3), and Specific Schedule of Assessments (Appendix 2 and Appendix 4).

Blood pressure and pulse rate should be obtained in a quiet room at a comfortable temperature, with the study subject's arm unconstrained by clothing or other material. All measurements will be obtained from the same arm and, with the same cuff size, using a well-calibrated automatic instrument with a digital readout, throughout the study. After the study subject has been in a supine position for at least 5 minutes, blood pressure and pulse rate will be obtained.

Blood pressure and pulse rate will be performed in triplicate (can be as short as 20 sec to 1-min interval between measurements) after the subject has rested in supine position for at least 5 minutes. The mean of three consecutive replicates will be used as the value for the defined time-point.

4.5.1.4 Electrocardiograms

12-Lead ECG

ECGs will be collected after the study subject has been in a supine position for at least 10 minutes. At the specified time-points, 12-lead ECGs will be obtained in triplicate, i.e., three consecutive interpretable 12-lead ECGs within a 2-5-minute interval, and recorded in CRF. Triplicate recordings should be taken for any unscheduled ECG.

All ECG recordings must be performed using a standard digital high-quality, high-fidelity ECG machine equipped with computer-based interval measurements. Automated ECG intervals (PR [PQ], QRS, QT, QTcF [to be derived in eCRF]) and HR will be captured, or calculated, and the changes in T-wave and U-wave morphology will be documented. T-wave information will be captured as normal or abnormal, U-wave information will be captured in two categories: absent/normal or abnormal.

For safety monitoring purposes, the Investigator must review, sign and date all ECG tracings either by wet signature or electronically. Paper or electronic ECG tracings must be appropriately kept by the study center and must fulfill all applicable archiving requirements. The ECG intervals and interpretation will be recorded on the eCRF or may be sent electronically. If considered appropriate by Roche, ECGs may be analyzed retrospectively at a central laboratory.

The following are requirements for ECG assessments:

1. Digital ECG recordings, storage and analysis.
2. Three useful recordings must be collected without artefacts, per time-point.
3. Body position should also be consistently maintained for each ECG performed. In particular, changes in HR should be avoided. The absence of any environmental distractions (TV, radio, conversation) during the pre-ECG rest and the ECG recording in the clinic must be emphasized.
4. Avoid ECG recordings within 2 hours after meals if possible (it is accepted that this is not possible after the breakfast and lunch for some relevant cohorts).
5. Strictly match timing and conditions of ECG recording to baseline. Conditions to be standardized include food intake, activity level, stressors, and room temperature.
6. If possible, the same machine, brand and model, should be used for the same study subject throughout the study.
7. ECGs should be 12-lead, recorded at 25 mm/sec for at least 10 sec.
8. In some cases, it may be appropriate to repeat abnormal ECGs to rule out improper lead placement as contributing to the ECG abnormality.
9. ECG machines should have periodic calibration and service records (minimum once a year).
10. If any QT/QTc values > 500 msec or increases from pre-dose on Day 1 QTc > 60 msec (as provided by the machine), the site should repeat the ECG within the next 5 minutes and notify the Sponsor. If confirmed, ECG recordings should be repeated at least hourly until two successive ECGs show QTc values below the threshold value that triggered the repeated measurement.

4.5.1.5 Laboratory Assessments

Normal ranges for the study laboratory parameters must be supplied to the Sponsor before the study starts. Laboratory safety tests shall be collected at time-points specified in the Schedule of Assessments (Appendix 1 and Appendix 3).

Additional blood or urine samples may be taken at the discretion of the Investigator if the results of any test fall outside the reference ranges, or clinical symptoms necessitate additional testing to monitor study subject safety. Where the clinical significance of abnormal lab results is considered uncertain, screening lab tests may be repeated before randomization to confirm eligibility. If there is an alternative explanation for a

positive urine or blood test for drugs of abuse, e.g., previous occasional intake of a medication or food containing for example codeine, benzodiazepines or opiates, the test could be repeated to confirm washout.

In the event of unexplained abnormal clinically significant laboratory test values, the tests should be repeated immediately and followed up until they have returned to the normal range and/or an adequate explanation of the abnormality is found. Results of clinical laboratory testing will be recorded on the eCRF or be received as electronically produced laboratory reports submitted directly from the local or central laboratory.

The samples listed below will be collected at the time-points indicated in the SoA:

- Hematology: hemoglobin, hematocrit, total white blood cell (WBC) count, differential WBC count (basophils, eosinophils, lymphocytes, monocytes, and neutrophils), platelet count, erythrocytes count (RBC), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC) and reticulocyte counts.
- Coagulation: prothrombin time, international normalized ratio (INR), activated thromboplastin time (aPTT).
- Blood Chemistry: alanine aminotransferase (ALT), aspartate aminotransferase (AST), total and indirect bilirubin, alkaline phosphatase (ALP), gamma-glutamyl transferase (at screening only), creatinine phosphokinase (at screening only), total protein, albumin, urea, creatinine, CrCl (at screening only, using the Cockcroft-Gault formula (Appendix 6), uric acid, total cholesterol, LDL, HDL, triglycerides, fasting glucose, sodium, chloride, potassium, calcium, phosphorus.
- Urinalysis: a midstream, clean-catch urine specimen will be collected for dipstick analysis of protein, blood, glucose, leucocytes and pH. If the dipstick result is 2+ or greater for blood, protein or leukocytes, urine will be sent to the laboratory for microscopy. If there is an explanation for the positive dipstick result, e.g., menses, it should be recorded, and there is no need to perform microscopy. Urine color may be evaluated from urinalysis if considered necessary.
- Drugs of abuse will be measured in urine: cannabinoids, amphetamines, methamphetamines, opiates, methadone, cocaine, benzodiazepines, and barbiturates.
- Alcohol: alcohol breath test.
- Viral Markers:
  - Hepatitis A (HAV IgM Ab), hepatitis B (HBsAg), hepatitis C (HCV RNA or HCV Ab), human immunodeficiency virus 1 and 2 (HIV Ab).
- Pregnancy Test: plasma or serum beta-human chorionic gonadotropin (β-HCG) at screening, urine on all other occasions (females only).
- Hormones: follicle-stimulating hormone (females only to confirm post-menopausal status).

4.5.1.6 Pharmacokinetic Assessments

Blood samples will be collected to evaluate the PK as specified in the SoA (Appendix 1, Appendix 2, Appendix 3, and Appendix 4). When the PK assessment is scheduled for the same nominal time as another scheduled assessment, the PK blood samples should be taken as close as possible to the scheduled time.

Blood samples will be collected from all cohorts. The actual date and time of each blood sample collection will be recorded in CRF.

A decision to stop PK sampling earlier or to collect more samples than currently proposed scheduled times will be based on the PK profile of the study drug. Timing of PK sampling may change based on emerging PK results after agreement with the Sponsor and the Investigator.

A PK sample will also be collected following occurrence of a dose-limiting event or serious adverse event.

PK parameters will be estimated using standard non-compartmental methods for RO7049389.

PK samples will be destroyed no later than 2 years after the date of final closure of the clinical database, unless regulatory authorities require specimens to be maintained for a longer time period.

Plasma samples will be analyzed for metabolite characterization of RO7049389 (M5, M6, and M11) with the use of quantitative and/or qualitative methods.

For sampling procedures, storage conditions, and shipment instructions, see the separate laboratory manual.

4.5.2 Timing of Study Assessments

4.5.2.1 Screening and Pretreatment Assessments

Written informed consent for participation in the study must be obtained before performing any study-specific screening tests or evaluations. Informed Consent Forms for enrolled study subject and for study subjects who are not subsequently enrolled will be maintained at the study site.

All screening and pre-treatment assessments must be completed and reviewed to confirm that Study Subjects meet all eligibility criteria. The Investigator will maintain a screening log to record details of all study subjects screened and to confirm eligibility or record reasons for screening failure.

An Eligibility Screening Form (ESF) documenting the Investigator's assessment of each screened study subject with regard to the protocol's inclusion and exclusion criteria is to be completed by the Investigator and kept at the investigational site.

Screening assessments will be performed within 28 days prior to randomization, unless otherwise specified. If a subject fails an inclusion/exclusion criterion due to a transient and non-clinically significant condition at screening, the Investigator may repeat the relevant screening assessment(s) within the 28-day screening period. If the subject fails a second time they will be classed as a screen failure and cannot be re-screened.

For subjects who participate in both SAD and MAD parts of the study, a brief and limited re-screening will be conducted prior to initiation of the MAD part (see Appendix 3).

Re-screening is allowed for subjects who were screened in the study and met study inclusion/exclusion criteria but failed to be randomized within 28 days after the start of screening period because the recruitment into a cohort was suspended, or a stratum cap in a cohort was reached. In order to re-screen such a subject, all inclusion and exclusion criteria should be re-evaluated and all applicable screening assessments repeated if done more than 28 days before randomization.

4.5.2.2 Assessments during Treatment

Under no circumstances will study subjects who enroll in this study and have completed treatment as specified, be permitted to be allocated a new randomization number and re-enroll in the study.

All assessments must be performed as per SoA (see Appendix 1, Appendix 2, Appendix 3, and Appendix 4).

4.5.2.3 Assessments at Study Completion/Early Termination Visit

Study subjects who complete the study or discontinue from the study early will be asked to return to the clinic 7 days (±1 day) after the last dose of study drug for a follow-up visit and to complete assessments as specified in the SOA.

In case of early termination of a subject in SAD or MAD, a blood sample for PK assessment should be collected at the time of discontinuation.

4.5.2.4 Follow-Up Assessments

After the study completion/early termination visit, adverse events should be followed as outlined in Section 5.5.

4.5.2.5 Assessments at Unscheduled Visits

Please see Appendix 1 and Appendix 3 for assessments that are required to be performed in case of an unscheduled visit. All unscheduled assessments should be reported in eCRF.

Unscheduled local laboratory tests may be ordered per Investigator's discretion and may be used for the individual management of the study subject.

4.6 STUDY SUBJECT, STUDY, AND SITE DISCONTINUATION

4.6.1 Study Subject Discontinuation

The Investigator has the right to discontinue a study subject from RO7049389 or withdraw a study subject from the study at any time. In addition, study subjects have the right to voluntarily discontinue study drug or withdraw from the study at any time for any reason. Reasons for discontinuation of study drug or withdrawal from the study may include, but are not limited to, the following*:*

- Study subject withdrawal of consent at any time.
- Any medical condition that the investigator or Sponsor determines may jeopardize the study subject's safety if he or she continues in the study.
- Investigator or Sponsor determines it is in the best interest of the study subject.
- Study subject non-compliance.

An early termination blood sample for RO7049389 PK assessment will be collected at the time of discontinuation.

4.6.1.1 Discontinuation from Study Drug

Individual study subjects should discontinue RO7049389/placebo if they experience any of the following:

- Clinically significant RO7049389-related changes in safety parameters that are considered not acceptable by the Investigator and/or the Sponsor.
- Poor gastrointestinal (GI) tolerability that is considered to affect the study subject's well-being and/or the PK evaluation.

All study subjects have the right to withdraw from the study at any time for any reason.

The investigators have the right to withdraw study subject from the study in the event of intercurrent illness, adverse events, for administrative or other reasons. The Sponsor should be informed of study subjects' discontinuations from the study or from the study drug.

Study Subjects who withdraw from the study prematurely will be asked to return to the clinic for a follow-up visit in 7 days after the last dose of study drug. The primary reason for premature study drug discontinuation should be documented on the appropriate eCRF.

4.6.1.2 Withdrawal from Study

Every effort should be made to obtain information on study subjects who withdraw from the study. The primary reason for withdrawal from the study should be documented on the appropriate eCRF.

Study subjects will not be followed for any reason after consent has been withdrawn.

When a subject voluntarily withdraws from the study, or is withdrawn by the Investigator, samples collected until the date of withdrawal will be analyzed, unless subject specifically requests for these to be discarded or local laws require their immediate destruction.

Study subjects who withdraw from the study for safety reasons will not be replaced. Study subjects who withdraw from the study for other reasons will be replaced.

4.6.2 Study and Site Discontinuation

The Sponsor has the right to terminate this study at any time. Reasons for terminating the study may include, but are not limited to, the following:

- The incidence or severity of adverse events in this or other studies indicates a potential health hazard to study subjects.
- Study subject enrollment is unsatisfactory.
- Previously unknown data become available which raise significant concerns about the potential risk to participants from continuation of the study

The Sponsor will notify the Investigator and Health Authorities if the study is placed on hold, or if the Sponsor decides to discontinue the study or development program.

The Sponsor has the right to replace a site at any time. Reasons for replacing a site may include, but are not limited to, the following:

- Excessively slow recruitment
- Poor protocol adherence
- Inaccurate or incomplete data recording
- Non-compliance with the International Conference on Harmonization (ICH) guideline for Good Clinical Practice.

5. ASSESSMENT OF SAFETY

5.1 SAFETY PARAMETERS AND DEFINITIONS

Safety assessments will consist of monitoring and recording adverse events, including serious adverse events and non-serious adverse events of special interest; measurement of protocol‑specified safety laboratory assessments; measurement of protocol-specified vital signs, ECGs; and other protocol-specified tests that are deemed critical to the safety evaluation of the study.

Certain types of events require immediate reporting to the Sponsor, as outlined in Section 5.3.

5.1.1 Adverse Events

According to the ICH guideline for Good Clinical Practice, an adverse event is any untoward medical occurrence in a clinical investigation subject administered a pharmaceutical product, regardless of causal attribution. An adverse event can therefore be any of the following:

- Any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.
- Any new disease or exacerbation of an existing disease (a worsening in the character, frequency, or severity of a known condition.
- Recurrence of an intermittent medical condition (e.g., headache) not present at baseline.
- Any deterioration in a laboratory value or other clinical test (e.g., ECG, X‑ray) that is associated with symptoms or leads to a change in study treatment or concomitant treatment or discontinuation from study drug.
- Adverse events that are related to a protocol-mandated intervention, including those that occur prior to assignment of study treatment (e.g., screening invasive procedures such as biopsies).

5.1.2 Serious Adverse Events (Immediately Reportable to the Sponsor)

A serious adverse event is any adverse event that meets any of the following criteria:

- Fatal (i.e., the adverse event actually causes or leads to death).
- Life-threatening (i.e., the adverse event, in the view of the Investigator, places the subject at immediate risk of death).

This does not include any adverse event that had it occurred in a more severe form or was allowed to continue might have caused death.

- Requires or prolongs inpatient hospitalization (see Section 5.2.5.9).
- Results in persistent or significant disability/incapacity (i.e., the adverse event results in substantial disruption of the study subject’s ability to conduct normal life functions).
- Congenital anomaly/birth defect in a neonate/infant born to a mother exposed to study drug.
- Significant medical event in the investigator's judgment (e.g., may jeopardize the study subject or may require medical/surgical intervention to prevent one of the outcomes listed above).

The terms “severe” and “serious” are not synonymous. Severity refers to the intensity of an adverse event (rated as mild, moderate, or severe, or according to a pre-defined grading criteria (e.g., National Cancer Institute Common Terminology Criteria for

Adverse Events [NCI CTCAE] criteria; see Section 5.2.3); the event itself may be of relatively minor medical significance (such as severe headache without any further findings).

Severity and seriousness need to be independently assessed for each adverse event recorded on the eCRF.

Serious adverse events are required to be reported by the Investigator to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5.3.2, for reporting instructions) and to the local health authority and IRB/EC, if required by local requirements.

5.1.3 Non-Serious Adverse Events of Special Interest (Immediately Reportable to the Sponsor)

Non-serious adverse events of special interest are required to be reported by the Investigator to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5.3.2 for reporting instructions). Adverse events of special interest for this study include the following:

- Cases of an elevated ALT or AST in combination with either an elevated bilirubin or clinical jaundice, as defined in Section 5.2.5.6.
- Suspected transmission of an infectious agent by the study drug, as defined below:

  Any organism, virus, or infectious particle (e.g., prion protein transmitting transmissible spongiform encephalopathy), pathogenic or non-pathogenic, is considered an infectious agent. A transmission of an infectious agent may be suspected from clinical symptoms or laboratory findings that indicate an infection in a Subject exposed to a medicinal product. This term applies only when a contamination of the study drug is suspected.

5.2 METHODS AND TIMING FOR CAPTURING AND ASSESSING SAFETY PARAMETERS

The Investigator is responsible for ensuring that all adverse events (see Section 5.1.1 for definition) are recorded on the Adverse Event eCRF and reported to the Sponsor in accordance with instructions provided in this section and in Sections 5.3, 5.4, and 5.5.

For each adverse event recorded on the Adverse Event eCRF, the Investigator will make an assessment of seriousness (see Section 5.1.2 for seriousness criteria), severity (see Section 5.2.3), and causality (see Section 5.2.4).

5.2.1 Adverse Event Reporting Period

Investigators will seek information on adverse events at each study subject contact. All adverse events, whether reported by the study subject or noted by study personnel, will be recorded in the study subject's medical record. Adverse events will then be reported on the Adverse Event eCRF as follows:

After informed consent has been obtained **but prior to initiation of study drug**, only serious adverse events caused by a protocol-mandated intervention should be reported (e.g., serious adverse events related to invasive procedures such as biopsies). Any other adverse event should not be reported.

After initiation of study drug, all adverse events, regardless of relationship to study drug, will be reported until **28 days** after the last dose of study drug.

After a period of 28 days from the last dose of study drug, investigators should report any deaths, serious adverse events, or other adverse events of concern that are believed to be related to prior treatment with study drug (see Section 5.5).

5.2.2 Eliciting Adverse Event Information

A consistent methodology of non-directive questioning should be adopted for eliciting adverse event information at all study subject evaluation time‑points. Examples of non-directive questions include the following:

"How have you felt since your last clinic visit?"

"Have you had any new or changed health problems since you were last here?"

5.2.3 Assessment of Severity of Adverse Events

Table 2 provides guidance for assessing adverse event severity.

Table 2 Adverse Event Severity Grading Scale

Severity	Description
Mild	Discomfort noticed, but no disruption of normal daily activity
Moderate	Discomfort sufficient to reduce or affect normal daily activity
Severe	Incapacitating with inability to work or to perform normal daily activity

Note: Regardless of severity, some events may also meet seriousness criteria. Refer to definition of a serious adverse event (see Section 5.1.2).

5.2.4 Assessment of Causality of Adverse Events

Investigators should use their knowledge of the study subject, the circumstances surrounding the event, and an evaluation of any potential alternative causes to determine whether or not an adverse event is considered to be related to the study drug, indicating "yes" or "no" accordingly. The following guidance should be taken into consideration:

- Temporal relationship of event onset to the initiation of study drug.
- Course of the event, considering especially the effects of dose reduction, discontinuation of study drug, or reintroduction of study drug.
- Known association of the event with the study drug or with similar treatments.
- Known association of the event with the disease under study.
- Presence of risk factors in the study subject or use of concomitant medications known to increase the occurrence of the event.
- Presence of non-treatment-related factors that are known to be associated with the occurrence of the event

5.2.5 Procedures for Recording Adverse Events

Investigators should use correct medical terminology/concepts when recording adverse events on the Adverse Event eCRF. Avoid colloquialisms and abbreviations.

Only one adverse event term should be recorded in the event field on the Adverse Event eCRF.

5.2.5.1 Diagnosis versus Signs and Symptoms

A diagnosis (if known) should be recorded on the Adverse Event eCRF rather than individual signs and symptoms (e.g., record only liver failure or hepatitis rather than jaundice, asterixis, and elevated transaminases). However, if a constellation of signs and/or symptoms cannot be medically characterized as a single diagnosis or syndrome at the time of reporting, each individual event should be recorded on the Adverse Event eCRF. If a diagnosis is subsequently established, all previously reported adverse events based on signs and symptoms should be nullified and replaced by one adverse event report based on the single diagnosis, with a starting date that corresponds to the starting date of the first symptom of the eventual diagnosis.

5.2.5.2 Adverse Events Occurring Secondary to Other Events

In general, adverse events occurring secondary to other events (e.g., cascade events or clinical sequelae) should be identified by their primary cause, with the exception of severe or serious secondary events. However, medically significant adverse events occurring secondary to an initiating event that are separated in time should be recorded as independent events on the Adverse Event eCRF. For example:

- If vomiting results in mild dehydration with no additional treatment in a healthy adult, only vomiting should be reported on the eCRF.
- If vomiting results in severe dehydration, both events should be reported separately on the eCRF.
- If a severe gastrointestinal hemorrhage leads to renal failure, both events should be reported separately on the eCRF.
- If dizziness leads to a fall and subsequent fracture, all three events should be reported separately on the eCRF.

All adverse events should be recorded separately on the Adverse Event eCRF if it is unclear as to whether the events are associated.

5.2.5.3 Persistent or Recurrent Adverse Events

A persistent adverse event is one that extends continuously, without resolution, between study subject evaluation time-points. Such events should only be recorded once on the Adverse Event eCRF. The initial severity of the event should be recorded, and the severity should be updated to reflect the most extreme severity any time the event worsens. If the event becomes serious, the Adverse Event eCRF should be updated to reflect this.

A recurrent adverse event is one that resolves between Subject evaluation time-points and subsequently recurs. Each recurrence of an adverse event should be recorded separately on the Adverse Event eCRF.

5.2.5.4 Abnormal Laboratory Values

Not every laboratory abnormality qualifies as an adverse event. A laboratory test result should be reported as an adverse event if it meets any of the following criteria:

- Accompanied by clinical symptoms.
- Results in a change in study treatment (e.g., dosage modification, treatment interruption, or treatment discontinuation).
- Results in a medical intervention (e.g., potassium supplementation for hypokalemia) or a change in concomitant therapy.
- Clinically significant in the Investigator’s judgment.

It is the Investigator's responsibility to review all laboratory findings. Medical and scientific judgment should be exercised in deciding whether an isolated laboratory abnormality should be classified as an adverse event.

If a clinically significant laboratory abnormality is a sign of a disease or syndrome (e.g., alkaline phosphatase and bilirubin 5 times the ULN associated with cholecystitis), only the diagnosis (i.e., cholecystitis) should be recorded on the Adverse Event eCRF.

If a clinically significant laboratory abnormality is not a sign of a disease or syndrome, the abnormality itself should be recorded on the Adverse Event eCRF, along with a descriptor indicating if the test result is above or below the normal range (e.g., "elevated potassium", as opposed to "abnormal potassium"). If the laboratory abnormality can be characterized by a precise clinical term per standard definitions, the clinical term should be recorded as the adverse event. For example, an elevated serum potassium level of 7.0 mEq/L should be recorded as "hyperkalemia".

Observations of the same clinically significant laboratory abnormality from visit to visit should not be repeatedly recorded on the Adverse Event eCRF, unless the etiology changes. The initial severity of the event should be recorded, and the severity or seriousness should be updated any time the event worsens.

5.2.5.5 Abnormal Vital Sign Values

Not every vital sign abnormality qualifies as an adverse event. A vital sign result should be reported as an adverse event if it meets any of the following criteria:

- Accompanied by clinical symptoms.
- Results in a change in study treatment (e.g., dosage modification, treatment interruption, or treatment discontinuation).
- Results in a medical intervention or a change in concomitant therapy.
- Clinically significant in the Investigator's judgment.

It is the Investigator's responsibility to review all vital sign findings. Medical and scientific judgment should be exercised in deciding whether an isolated vital sign abnormality should be classified as an adverse event.

If a clinically significant vital sign abnormality is a sign of a disease or syndrome (e.g., high blood pressure), only the diagnosis (i.e., hypertension) should be recorded on the Adverse Event eCRF.

Observations of the same clinically significant vital sign abnormality from visit to visit should not be repeatedly recorded on the Adverse Event eCRF, unless the etiology changes. The initial severity of the event should be recorded, and the severity or seriousness should be updated any time the event worsens.

5.2.5.6 Abnormal Liver Function Tests

The finding of an elevated ALT or AST ($\geq 3 \times$ ULN) in combination with either an elevated total bilirubin ($\geq 2 \times$ ULN) or clinical jaundice in the absence of cholestasis or other causes of hyperbilirubinemia is considered to be an indicator of severe liver injury. Therefore, investigators must report as an adverse event the occurrence of either of the following:

- Treatment-emergent ALT or AST $> 3 \times$ ULN in combination with total bilirubin $> 2 \times$ ULN.
- Treatment-emergent ALT or AST $> 3 \times$ ULN in combination with clinical jaundice.

The most appropriate diagnosis or (if a diagnosis cannot be established) the abnormal laboratory values should be recorded on the Adverse Event eCRF (see Section 5.2.5.1) and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event), either as a serious adverse event or a non-serious adverse event of special interest (see Section 5.3.2) and to the local health authority and IRB/EC, if required by local requirements.

5.2.5.7 Deaths

All deaths that occur during the protocol-specified adverse event reporting period (see Section 5.2.1), regardless of relationship to study drug, must be recorded on the Adverse Event eCRF and immediately reported to the Sponsor (see Section 5.4.2).

Death should be considered an outcome and not a distinct event. The event or condition that caused or contributed to the fatal outcome should be recorded as the single medical concept on the Adverse Event eCRF. Generally, only one such event should be reported. The term “sudden death” should only be used for the occurrence of an abrupt and unexpected death due to presumed cardiac causes in a subject with or without preexisting heart disease, within 1 hour of the onset of acute symptoms or, in the case of an unwitnessed death, within 24 hours after the subject was last seen alive and stable. If the cause of death is unknown and cannot be ascertained at the time of reporting, “unexplained death” should be recorded on the Adverse Event eCRF. If the cause of death later becomes available (e.g., after autopsy), “unexplained death” should be replaced by the established cause of death.

5.2.5.8 Preexisting Medical Conditions

A preexisting medical condition is one that is present at the screening visit for this study. Such conditions should be recorded on the General Medical History and Baseline Conditions eCRF.

A preexisting medical condition should be recorded as an adverse event only if the frequency, severity, or character of the condition worsens during the study. When recording such events on the Adverse Event eCRF, it is important to convey the concept that the preexisting condition has changed by including applicable descriptors (e.g., “more frequent headaches”).

5.2.5.9 Hospitalization or Prolonged Hospitalization

Any adverse event that results in hospitalization or prolonged hospitalization should be documented and reported as a serious adverse event (per the definition of serious adverse event in Section 5.2.5.9), except as outlined below.

The following hospitalization scenarios are not considered to be serious adverse events:

- Hospitalization for respite care.
- Planned hospitalization required by the protocol (e.g., for study drug administration and intensive PK sampling).
- Hospitalization for a preexisting condition, provided that all of the following criteria are met:

  The hospitalization was planned prior to the study or was scheduled during the study when elective surgery became necessary because of the expected normal progression of the disease.

  The study subject has not suffered an adverse event.

The following hospitalization scenarios are not considered to be serious adverse events, but should be reported as adverse events instead:

- Hospitalization for an adverse event that would ordinarily have been treated in an outpatient setting had an outpatient clinic been available

5.2.5.10 Overdoses

Study drug overdose is the accidental or intentional use of the drug in an amount higher than the dose being studied. An overdose or incorrect administration of study drug is not an adverse event unless it results in untoward medical effects.

Any study drug overdose or incorrect administration of study drug should be noted on the Study Drug Administration eCRF.

All adverse events associated with an overdose or incorrect administration of study drug should be recorded on the Adverse Event eCRF. If the associated adverse event fulfills serious criteria, the event should be reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5.3.2) and to the local health authority and IRB/EC, if required by local requirements.

5.3 IMMEDIATE REPORTING REQUIREMENTS FROM INVESTIGATOR TO SPONSOR

Certain events require immediate reporting to allow the Sponsor to take appropriate measures to address potential new risks in a clinical trial. The Investigator must report such events to the Sponsor immediately; under no circumstances should reporting take place more than 24 hours after the investigator learns of the event. The following is a list

of events that the Investigator must report to the Sponsor within 24 hours after learning of the event, regardless of relationship to study drug:

- Serious adverse events
- Non-serious adverse events of special interest
- Pregnancies (see Section 5.3.3)

The Investigator must report new significant follow-up information for these events to the Sponsor immediately (i.e., no more than 24 hours after becoming aware of the information). New significant information includes the following:

- New signs or symptoms or a change in the diagnosis
- Significant new diagnostic test results
- Change in causality based on new information
- Change in the event's outcome, including recovery
- Additional narrative information on the clinical course of the event

Investigators must also comply with local requirements for reporting serious adverse events to the local health authority and IRB/EC.

5.3.1 Emergency Medical Contacts

To ensure the safety of Study Subjects, access to the Medical monitors is available 24 hours a day 7 days a week. Medical monitors contact details are listed in the "Protocol Administrative and Contact Information & List of Investigators".

5.3.2 Reporting Requirements for Serious Adverse Events and Non-Serious Adverse Events of Special Interest

Events that Occur prior to Study Treatment Initiation

After informed consent has been obtained but prior to initiation of study treatment, only serious adverse events caused by a protocol-mandated intervention should be reported. The Serious Adverse Event/Adverse Event of Special Interest Reporting Form provided to Investigators should be completed and submitted to the Serious Adverse Event Responsible immediately (i.e., no more than 24 hours after learning of the event).

Events that Occur after Study Treatment Initiation

For reports of serious adverse events and non-serious adverse events of special interest (see Section 5.1.2 and 5.1.3) that occur after initiation of study treatment until the end of the adverse event reporting period (defined in Section 5.5), investigators should record all case details that can be gathered immediately (i.e., within 24 hours after learning of the event) on the appropriate Adverse Event of Special Interest/ Serious Adverse Event eCRF form and submit the report via the electronic data capture (EDC) system. A report will be generated and sent to the Sponsor's Safety Risk Management department.

In the event that the EDC system is unavailable, the Serious Adverse Event/Adverse Event of Special Interest Reporting Form provided to investigators should be completed and submitted to the Serious Adverse Event Responsible immediately (i.e., no more than 24 hours after learning of the event) and to the local health authority and IRB/EC, if required by local requirements.

Once the EDC system is available, all information will need to be entered and submitted via the EDC system.

5.3.3 Reporting Requirements for Pregnancies

5.3.3.1 Pregnancies in Female Subjects

Female subjects of childbearing potential will not be allowed to participate in this study. However, if a female subject becomes pregnant during the study or within 28 days after the last dose of study drug, a Clinical Trial Pregnancy Reporting Form should be completed by the investigator and submitted to the sponsor within 24 hours after learning of the pregnancy. Pregnancy should not be recorded on the Adverse Event eCRF. The Investigator should discontinue study drug and counsel the subject, discussing the risks of the pregnancy and the possible effects on the fetus. Monitoring of the subject should continue until conclusion of the pregnancy.

5.3.3.2 Pregnancies in Female Partners of Male Study Subjects

Male Subjects will be instructed through the Informed Consent Form to immediately inform the Investigator if their partner becomes pregnant during the study or within 28 days after the last dose of study drug. A Clinical Trial Pregnancy Reporting Form should be completed by the Investigator and submitted to the Sponsor within 24 hours after learning of the pregnancy. Attempts should be made to collect and report details of the course and outcome of any pregnancy in the partner of a male study subject exposed to study drug. The pregnant partner will need to sign an Authorization for Use and Disclosure of Pregnancy Health Information to allow for follow-up on her pregnancy. Once the authorization has been signed, the Investigator will update the Clinical Trial Pregnancy Reporting Form with additional information on the course and outcome of the pregnancy. An Investigator who is contacted by the male study subject or his pregnant partner may provide information on the risks of the pregnancy and the possible effects

on the fetus, to support an informed decision in cooperation with the treating physician and/or obstetrician.

5.3.3.3 Abortions

Any spontaneous abortion should be classified as a serious adverse event (as the Sponsor considers spontaneous abortions to be medically significant events), recorded on the Adverse Event eCRF, and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5.3.2) and to the local health authority and IRB/EC, if required by local requirements.

Any induced abortion due to maternal toxicity and/or embryo-fetal toxicity should also be classified as serious adverse event, recorded on the Adverse Event eCRF, and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5.3.2) and to the local health authority and IRB/EC, if required by local requirements.

Elective abortion not associated with toxicity (e.g., induced abortion for personal reasons) does not require expedited reporting but should be reported as outcome of pregnancy on the Clinical Trial Pregnancy Reporting Form.

5.3.3.4 Congenital Anomalies/Birth Defects

Any congenital anomaly/birth defect in a child born to a female subject or female partner of a male subject exposed to study drug should be classified as a serious adverse event, recorded on the Adverse Event eCRF, and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5.3.2) and to the local health authority and IRB/EC, if required by local requirements.

5.4 FOLLOW-UP OF SUBJECTS AFTER ADVERSE EVENTS

5.4.1 Investigator Follow-Up

The Investigator should follow each adverse event until the event has resolved to baseline grade or better, the event is assessed as stable by the Investigator, the subject is lost to follow-up, or the subject withdraws consent. Every effort should be made to follow all serious adverse events considered to be related to study drug or trial-related procedures until a final outcome can be reported.

During the study period, resolution of adverse events (with dates) should be documented on the Adverse Event eCRF and in the subject's medical record to facilitate source data verification. If, after follow-up, return to baseline status or stabilization cannot be established, an explanation should be recorded on the Adverse Event eCRF.

All pregnancies reported during the study should be followed until pregnancy outcome and reported according to the instructions provided in Section 5.3.3.

5.4.2 Sponsor Follow-Up

For serious adverse events, non-serious adverse events of special interest, and pregnancies, the Sponsor or a designee may follow up by telephone, fax, electronic mail, and/or a monitoring visit to obtain additional case details and outcome information (e.g., from hospital discharge summaries, consultant reports, autopsy reports) in order to perform an independent medical assessment of the reported case.

5.5 POST-STUDY ADVERSE EVENTS

The Investigator is not required to actively monitor subjects for adverse events after the end of the adverse event reporting period (defined as 28 days after the last dose of study drug).

If the Investigator becomes aware of any other serious adverse event occurring after the end of the adverse event reporting period, if the event is believed to be related to prior study drug treatment the event should be reported directly to the Sponsor or its designee, either by faxing or by scanning and emailing the Serious Adverse Event Reporting Form using the fax number or email address provided to investigators.

5.6 EXPEDITED REPORTING TO HEALTH AUTHORITIES, INVESTIGATORS, INSTITUTIONAL REVIEW BOARDS, AND ETHICS COMMITTEES

The Sponsor will promptly evaluate all serious adverse events and non-serious adverse events of special interest against cumulative product experience to identify and expeditiously communicate possible new safety findings to investigators, IRBs, ECs, and applicable health authorities based on applicable legislation.

To determine reporting requirements for single adverse event cases, the Sponsor will assess the expectedness of these events using the following reference document:

- RO7049389 Investigator's Brochure

The Sponsor will compare the severity of each event and the cumulative event frequency reported for the study with the severity and frequency reported in the applicable reference document.

Reporting requirements will also be based on the investigator's assessment of causality and seriousness, with allowance for upgrading by the Sponsor as needed.

6. STATISTICAL CONSIDERATIONS AND ANALYSIS PLAN

The primary objective of this study is to characterize the safety profile associated with a single dose and with multiple doses of RO7049389 in healthy volunteers. Statistical analyses will be descriptive in nature. Subjects will be grouped according to the actual treatment they received. All subjects who receive any amount of study medication (RO7049389 or matching placebo) and with at least one safety assessment will be

included in the safety analysis. All subjects who receive placebo will be combined into a single pooled placebo control group.

6.1 DETERMINATION OF SAMPLE SIZE

The sample size was determined by practical considerations and not based on statistical power calculations. Approximately four subjects will be enrolled in SAD cohort 1 (2 active treatment and 2 placebo). Approximately twelve subjects will be enrolled in each of the other SAD and MAD cohorts. They will be randomized to either active treatment (10 subjects per dose level) or placebo (2 subjects per dose level).

With ten subjects on active drug per dose group or cohort, there is a 94% chance to observe at least one AE that has an incidence rate of 25% in the population.

6.2 SUMMARIES OF CONDUCT OF STUDY

The number of study subjects who enroll, discontinue, or complete the study will be summarized by treatment (RO7049389 vs. matching placebo) for each dose cohort in the study. Reasons for premature study withdrawal will be listed and summarized by treatment and dose cohort. Enrollment and protocol deviations will be listed and evaluated for their potential impact on interpretation of study results. Study drug administration (RO7049389 or matching placebo) will be summarized with number of doses and percentage of assigned dose subjects received by treatment and dose cohort.

6.3 ANALYSIS POPULATIONS

6.3.1 Safety Analysis Population

All study subjects who have received at least one dose of the study medication (RO7049389 or matching placebo), whether prematurely withdrawn from the study or not, and with at least one safety assessment, will be included in the safety analysis.

6.3.2 Pharmacokinetic Analysis Population

Study subjects will be excluded from the pharmacokinetic analysis population if they significantly violate the inclusion or exclusion criteria, deviate significantly from the protocol or if data are unavailable or incomplete which may influence the pharmacokinetic analysis. Excluded cases will be documented together with the reason for exclusion. All decisions on exclusions from the analysis will be made prior to database closure.

6.4 SUMMARIES OF TREATMENT GROUP COMPARABILITY

Study subjects randomized to receive placebo will be pooled together to form a pooled placebo group. Demographic and baseline characteristics (age, sex, etc.) will be summarized by treatment (each dose level of RO7049389 vs. pooled placebo group) for the safety population using descriptive statistics. For continuous variables, mean, standard deviation, median, and ranges will be presented. For categorical data, the

proportion of subjects in each category will be summarized. Summaries will be presented in overall and by cohorts.

6.5 SAFETY ANALYSES

All safety analyses will be based on the safety analysis population. Subjects will be grouped based on the treatment they actually receive. Safety will be assessed through physical exams, adverse events, changes in laboratory results, changes in ECG measurements, and changes in vital sign measurements.

6.5.1 Adverse Events

The original terms recorded on the eCRF by the Investigator for adverse events will be standardized by the Sponsor.

Adverse events will be summarized by MedDRA coded term. In addition, adverse events will be summarized by their toxicity level (according to the maximum reported severity level) and severity (severe vs. non-severe). All serious adverse events, including deaths, severe adverse events (Grade $\geq$ 3), and adverse events of special interest will be listed and summarized separately. Adverse events leading to study drug discontinuation or interruption will be summarized as well. Subjects will be tabulated by treatment (RO7049389 vs. pooled placebo group) and dose level.

6.5.2 Clinical Laboratory Test Results

All clinical laboratory data will be stored on the database in the units in which they were reported. Study subject listings and summary statistics at each assessment time will be presented using the International System of Units (SI units; Système International d'Unités). Laboratory data not reported in SI units will be converted to SI units before processing.

Laboratory test values will be presented by individual listings with flagging of values outside the normal ranges.

The proportion of study subjects with laboratory abnormalities will be summarized by treatment (RO7049389 vs. pooled placebo group) and dose level. Additionally, the absolute value and change from baseline in all laboratory tests will be summarized by treatment and visit using descriptive statistics.

6.5.2.1 Standard Reference Ranges and Transformation of Data

Roche standard reference ranges, rather than the reference ranges of the Investigator, will be used for all parameters. For most parameters, the measured laboratory test result will be assessed directly using the Roche standard reference range. Certain laboratory parameters will be transformed to Roche's standard reference ranges.

A transformation will be performed on certain laboratory tests that lack sufficiently common procedures and have a wide range of investigator ranges, e.g., enzyme tests

that include AST, ALT, and alkaline phosphatase and total bilirubin. Since the standard reference ranges for these parameters have a lower limit of zero, only the upper limits of the ranges will be used in transforming the data.

6.5.2.2 Definition of Laboratory Abnormalities

For all laboratory parameters included, there exists a Roche predefined standard reference range. Laboratory values falling outside this standard reference range will be labeled “H” for high or “L” for low in study subject listings of laboratory data.

In addition to the standard reference range, a marked reference range has been predefined by Roche for each laboratory parameter. The marked reference range is broader than the standard reference range. Values falling outside the marked reference range that also represent a defined change from baseline will be considered marked laboratory abnormalities (i.e., potentially clinically relevant). If a baseline value is not available for a study subject, the midpoint of the standard reference range will be used as the study subject’s baseline value for the purposes of determining marked laboratory abnormalities. Marked laboratory abnormalities will be labeled in the study subject listings as “HH” for very high or “LL” for very low.

6.5.3 Vital Signs

Vital signs data will be presented by individual listings with flagging of values outside the normal ranges and flagging of marked abnormalities. In addition, tabular summaries will be used, as appropriate.

6.5.4 ECG Data Analysis

ECG data will be presented by individual listings with flagging of values outside the normal ranges and flagging of marked abnormalities. In addition, tabular summaries will be used, as appropriate.

Summary descriptive statistics for the actual values and changes from baseline will be tabulated by nominal time for HR, QRS duration, PR and QTcF (see Appendix 5). For multiple measurements taken at a nominal time-point, the average of these measurements will be used as the value at that nominal time-point in all summaries. In addition, QTcF will be categorized at each time-point as $\leq$ 450 msec, $>$ 450-480 msec, $>$ 480-500 msec and $>$ 500 msec and summarized. Similarly, a summary will be provided of the QTcF changes from baseline at each time-point categorized as $<$ 30 msec, 30-60 msec, and $>$ 60 msec. Changes of the overall ECG interpretation, T-wave and U-wave morphology will be summarized.

6.5.5 Concomitant Medications

The original terms recorded on the study subjects’ eCRF by the Investigator for concomitant medications will be standardized by the Sponsor by assigning preferred terms.

Concomitant medications will be presented in summary tables and listings.

6.6 PHARMACOKINETIC ANALYSES

Non-compartmental analysis will be employed for estimation of PK parameters. Individual plasma concentrations at each sampling time-point for RO7049389 of each cohort will be presented by listings and appropriate descriptive summary statistics, including means, medians, geometric means, ranges, standard deviations and coefficients of variation. Individual and mean plasma concentration of each cohort versus time data will be plotted on semi-logarithmic scales.

All PK parameters will be presented by individual listings and summary statistics for each cohort including means, geometric means, medians, ranges, standard deviations and coefficients of variation.

The relationship of selected PK parameters from Day 1 with dose will be evaluated graphically to establish dose-linearity. Dose-proportionality may be assessed by estimating the slope of the linear regression of logarithmically transformed variables C_{max}, $AUC_{0\text{-}last}$ and $AUC_{0\text{-}inf}$ versus the logarithmically transformed dose. A 95% confidence interval for the slope which includes the value 1 would be consistent with dose-proportionality.

Plasma samples collected in the study will be qualitatively and/or quantitatively analyzed for the presence of metabolites of RO7049389.

A population PK model development and analysis may be performed. The Population PK results will be reported in a separated document.

6.7 INTERIM ANALYSIS

No interim analysis is planned for this study.

7. DATA COLLECTION AND MANAGEMENT

7.1 DATA QUALITY ASSURANCE

The Sponsor will be responsible for data management of this study, including quality checking of the data. Sites will be responsible for data entry into the Electronic Data Capture (EDC) system.

A comprehensive validation check program will verify the data. Discrepancies will be generated automatically in the system at the point of entry or added manually for resolution by the Investigator.

The Sponsor will produce a Data Handling Manual and a Data Management Plan that describes the quality checking to be performed on the data. Laboratory electronic data

will be sent directly to the Sponsor, using the Sponsor's standard procedures to handle and process the electronic transfer of these data.

System backups for data stored by the Sponsor and records retention for the study data will be consistent with the Sponsor's standard procedures.

7.2 ELECTRONIC CASE REPORT FORMS

Data for this study will be captured via an on line EDC system. The data collected in the source documents is entered onto the study eCRF. An audit trail will maintain a record of initial entries and changes made; reasons for change; time and date of entry; and user name of person authorizing entry or change. For each study subject enrolled, an eCRF must be completed and electronically signed by the Principal Investigator or authorized delegate from the study staff. If a study subject withdraws from the study, the reason must be noted on the eCRF. If a study subject is withdrawn from the study because of a treatment-limiting adverse event, thorough efforts should be made to clearly document the outcome.

The Investigator should ensure the accuracy, completeness and timeliness of the data reported to the Sponsor/CRO in the eCRFs and in all required reports.

eCRFs will be submitted electronically to the Sponsor/CRO and should be handled in accordance with instructions from the Sponsor/CRO.

At the end of the study, the Investigator will receive subject data for his or her site in a readable format on a compact disc that must be kept with the study records. Acknowledgement of receipt of the compact disc is required.

7.3 SOURCE DATA DOCUMENTATION

Study monitors will perform ongoing source data verification to confirm that critical protocol data (i.e., source data) entered into the eCRFs by authorized site personnel are accurate, complete, and verifiable from source documents.

Source documents (paper or electronic) are those in which subject data are recorded and documented for the first time. They include, but are not limited to, hospital records, clinical and office charts, laboratory notes, memoranda, subject -reported outcomes, evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies of transcriptions that are certified after verification as being accurate and complete, microfiche, photographic negatives, microfilm or magnetic media, X-rays, subject files, and records kept at pharmacies, laboratories, and medico-technical departments involved in a clinical trial.

Before study initiation, data to be entered directly into the eCRFs (i.e., no prior written or electronic record of the data) and considered source data must be defined in the Trial Monitoring Plan.

Source documents that are required to verify the validity and completeness of data entered into the eCRFs must not be obliterated or destroyed and must be retained per the policy for retention of records described in Section 7.5.

To facilitate source data verification, the investigators and institutions must provide the Sponsor direct access to applicable source documents and reports for trial-related monitoring, Sponsor audits, and IRB/EC review. The investigational site must also allow inspection by applicable Health Authorities.

7.4 USE OF COMPUTERIZED SYSTEMS

When clinical observations are entered directly into an investigational site's computerized medical record system (i.e., in lieu of original hardcopy records), the electronic record can serve as the source document if the system has been validated in accordance with Health Authority requirements pertaining to computerized systems used in clinical research. An acceptable computerized data collection system allows preservation of the original entry of data. If original data are modified, the system should maintain a viewable audit trail that shows the original data as well as the reason for the change, name of the person making the change, and date of the change.

7.5 RETENTION OF RECORDS

Records and documents pertaining to the conduct of this study and the distribution of IMP, including eCRFs, Informed Consent Forms, laboratory test results, and medication inventory records, must be retained by the Principal Investigator for at least 15 years after completion or discontinuation of the study, or for the length of time required by relevant national or local Health Authorities, whichever is longer. After that period of time, the documents may be destroyed, subject to local regulations. No records may be disposed of without the written approval of the Sponsor. Written notification should be provided to the Sponsor prior to transferring any records to another party or moving them to another location.

8. ETHICAL CONSIDERATIONS

8.1 COMPLIANCE WITH LAWS AND REGULATIONS

This study will be conducted in full conformance with the ICH E6 guideline for Good Clinical Practice and the principles of the Declaration of Helsinki, or the laws and regulations of the country in which the research is conducted, whichever affords the greater protection to the individual. The study will comply with the requirements of the ICH E2A guideline (Clinical Safety Data Management: Definitions and Standards for Expedited Reporting). Studies conducted in the United States or under a U.S. Investigational New Drug (IND) application will comply with U.S. FDA regulations and applicable local, state, and federal laws. Studies conducted in the EU/EEA will comply with the EU Clinical Trial Directive (2001/20/EC).

8.2 INFORMED CONSENT

The Sponsor's sample Informed Consent Form will be provided to the site. If applicable, it will be provided in a certified translation of the local language. The Sponsor or its designee must review and approve any proposed deviations from the Sponsor's sample Informed Consent Forms or any alternate consent forms proposed by the site (collectively, the "Consent Forms") before IRB/EC submission. The final IRB/EC-approved Consent Forms must be provided to the Sponsor for Health Authority submission purposes according to local requirements.

The Consent Forms must be signed and dated by the subject before his or her participation in the study. The case history or clinical records for each subject shall document the informed consent process and that written informed consent was obtained prior to participation in the study.

The Consent Forms should be revised whenever there are changes to study procedures or when new information becomes available that may affect the willingness of the subject to participate. The final revised IRB/EC-approved Consent Forms must be provided to the Sponsor for Health Authority submission purposes.

Subjects must be re-consented to the most current version of the Consent Forms (or to a significant new information/findings addendum in accordance with applicable laws and IRB/EC policy) during their participation in the study. For any updated or revised Consent Forms, the case history or clinical records for each subject shall document the informed consent process and that written informed consent was obtained using the updated/revised Consent Forms for continued participation in the study.

A copy of each signed Consent Form must be provided to the subject. All signed and dated Consent Forms must remain in each subject's study file or in the site file and must be available for verification by study monitors at any time.

8.3 INSTITUTIONAL REVIEW BOARD OR ETHICS COMMITTEE

This protocol, the Informed Consent Forms, any information to be given to the subject, and relevant supporting information must be submitted to the IRB/EC by the Principal Investigator and reviewed and approved by the IRB/EC before the study is initiated. In addition, any subject recruitment materials must be approved by the IRB/EC.

The Principal Investigator is responsible for providing written summaries of the status of the study to the IRB/EC annually or more frequently in accordance with the requirements, policies, and procedures established by the IRB/EC. Investigators are also responsible for promptly informing the IRB/EC of any protocol amendments (see Section 9.5).

In addition to the requirements for reporting all adverse events to the Sponsor, investigators must comply with requirements for reporting serious adverse events to the local health authority and IRB/EC. Investigators may receive written IND safety reports

or other safety-related communications from the Sponsor. Investigators are responsible for ensuring that such reports are reviewed and processed in accordance with Health Authority requirements and the policies and procedures established by their IRB/EC, and archived in the site's study file.

8.4 CONFIDENTIALITY

The Sponsor maintains confidentiality standards by coding each subject enrolled in the study through assignment of a unique subject identification number. This means that subject names are not included in data sets that are transmitted to any Sponsor location.

Subject medical information obtained by this study is confidential and may only be disclosed to third parties as permitted by the Informed Consent Form (or separate authorization for use and disclosure of personal health information) signed by the subject, unless permitted or required by law.

Medical information may be given to a subject's personal physician or other appropriate medical personnel responsible for the subject's welfare, for treatment purposes.

Data generated by this study must be available for inspection upon request by representatives of the U.S. FDA and other national and local Health Authorities, Sponsor monitors, representatives, and collaborators, and the IRB/EC for each study site, as appropriate.

8.5 FINANCIAL DISCLOSURE

Investigators will provide the Sponsor with sufficient, accurate financial information in accordance with local regulations to allow the Sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate Health Authorities. Investigators are responsible for providing information on financial interests during the course of the study and for one year after completion of the study (i.e., LSLV).

9. STUDY DOCUMENTATION, MONITORING, AND ADMINISTRATION

9.1 STUDY DOCUMENTATION

The Investigator must maintain adequate and accurate records to enable the conduct of the study to be fully documented, including but not limited to the protocol, protocol amendments, Informed Consent Forms, and documentation of IRB/EC and governmental approval. In addition, at the end of the study, the Investigator will receive the Subject data, which includes an audit trail containing a complete record of all changes to data.

9.2 SITE INSPECTIONS

Site visits will be conducted by the Sponsor or an authorized representative for inspection of study data, subjects' medical records, and eCRFs. The Investigator will

permit national and local Health Authorities, Sponsor monitors, representatives, and collaborators, and the IRBs/ECs to inspect facilities and records relevant to this study.

9.3 ADMINISTRATIVE STRUCTURE

The Sponsor of the trial is F. Hoffmann-La Roche Ltd. The Sponsor is responsible for the study management, data management, statistical analysis and medical writing for the clinical study report.

9.4 PUBLICATION OF DATA AND PROTECTION OF TRADE SECRETS

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 prior to submission. This allows the Sponsor to protect proprietary information and to provide comments based on information from other studies that may not yet be available to the Investigator.

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 trials only in their entirety and not as individual center data. In this case, a coordinating Investigator will be designated by mutual agreement.

Any formal publication of the study in which contribution of Sponsor personnel exceeded that of conventional monitoring will be considered as a joint publication by the Investigator and the appropriate Sponsor personnel.

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

Any inventions and resulting patents, improvements, and/or know-how originating from the use of data from this study will become and remain the exclusive and unburdened property of the Sponsor, except where agreed otherwise.

9.5 PROTOCOL AMENDMENTS

Any substantial protocol amendments will be prepared by the Sponsor. Substantial protocol amendments will be submitted to the IRB/EC and to regulatory authorities in accordance with local regulatory requirements.

Approval must be obtained from the IRB/EC and regulatory authorities (as locally required) before implementation of any changes, except for changes necessary to eliminate an immediate hazard to subjects or any non-substantial changes, as defined by regulatory requirements.

10. REFERENCES

Björkhem-Bergman L, Bäckström T, Nylén H, et al. Comparison of endogenous 4β-hydroxycholesterol with midazolam as markers for CYP3A4 induction by rifampicin. Drug Metab Dispos. 2013;41(8):1488-1493.

European Association for the Study of the Liver (EASL) Clinical Practice Guidelines: Management of chronic hepatitis B virus infection. J Hepatol. 2012;57:167–185.

FDA Guidance for Industry - Estimating the Maximum Safe Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers (2005). Retrieved from: http://www.fda.gov/downloads/Drugs/Guidances/UCM078932.pdf

Fernández M, Quiroga JA, Carreño V. Hepatitis B virus downregulates the human interferon-inducible MxA promoter through direct interaction of precore/core proteins. J Gen Virol. 2003;84(Pt 8):2073-2082.

Gruffaz M, Testoni B, Luangsay S, et al. Hepatitis B core protein is a key and very early negative regulator of Interferon response. J Hepatol. 2013;58:S155-S156.

Guideline on the Investigation of Drug Interactions (EMA) http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2012/07/WC500129606.pdf

Guideline on the Investigation of Drug Interactions (EMA) Retrieved from:http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2012/07/WC500129606.pdf

Halama B, Hohmann N, Burhenne J. A nanogram dose of the CYP3A probe substrate midazolam to evaluate drug interactions. Clin Pharmacol Ther. 2013;93(6):564-571.

ICH guideline M3 (R2) http://www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Multidisciplinary/M3_R2/Step4/M3_R2__Guideline.pdf

Investigator's Brochure, RO7049389. Second Version, October 2017.

Liang X, Bi S, Yang W et al. Epidemiological serosurvey of hepatitis B in China--declining HBV prevalence due to hepatitis B vaccination. Vaccine. 2009;27(47):6550-6557.Midazolam. In: DRUGDEX System (Micromedex 2.0). Greenwood Village, CO.

Neuenschwander B, Branson M, Gsponer T. Critical aspects of the Bayesian approach to phase I cancer trials. Stat Med. 2008;27(13):2420-2439.

Truven Health Analytics; c1974-2013.Retrieved from: http://www.micromedexsolutions.com/micromedex2/librarian# Accessed August 11,2014.

Twu JS, Lee CH, Lin PM, et al. Hepatitis B virus suppresses expression of human beta-interferon. Proc Natl Acad Sci USA. 1988;85(1):252-256.

WHO Fact Sheet N°204, updated July 2016
http://www.who.int/mediacentre/factsheets/fs204/en/

Yang J, Liao M, Shou M. Cytochrome p450 turnover: regulation of synthesis and degradation, methods for determining rates, and implications for the prediction of drug interactions. Curr Drug Metab. 2008;9(5):384-394.

Yeung W, Whitehead J, Reigner B, et al. Bayesian adaptive dose-escalation procedures for binary and continuous responses utilizing a gain function. Pharm Statist. 2015;14:479-487.

Yu R, Fan R, Hou J. Chronic hepatitis B virus infection: epidemiology, prevention, and treatment in China. Front Med. 2014;8(2):135-144.

Appendix 5
Correction Formulas for QTc Intervals

Fridericia's correction for QTc Measurement – QTcF

$$\text{QTcF (msec)} = \frac{QT(msec)}{\sqrt[3]{RR(msec)/1000}}$$

Example: QTcF of a subject with a QT of 386 msec and a RR of 848 msec

QT (msec) = 386

RR (msec) = 848

$$QTcF = \frac{386}{\sqrt[3]{848/1000}} = 408\ \text{msec}$$

Appendix 6
Cockcroft Gault Equation for Calculation CrCl

The Cockcroft-Gault equation will be used to calculate creatinine clearance (CrCl) (Conventional units = mL/min or SI units = mL/sec). Baseline body weight (ABW) will be used for calculation of CrCl.

Conventional Units:

$$\textit{Males (mL/min)} = \frac{\textit{(140 – Age) * ABW (kg)}}{\textit{72 * Serum Creatinine (mg/dL)}}$$

Females (mL/min) = Male value x 0.85

Conversion Factor for Creatinine Clearance:

- SI Units (mL/sec) = Conventional units (mL/min) $\times$ 0.0167
- Conventional Units (mL/min) = SI Units (mL/sec) / 0.0167

Conversion Factor for Serum Creatinine:

- Conventional units (mg/dL) = SI units (µmol/L) / 88.4
- SI Units (µmol/L) = Conventional Units $\times$ 88.4