Official Title: A Phase I, Sponsor-Open, Investigator-Blinded, Subject-Blinded, Multi-Center, Placebo-Controlled Study to Evaluate Safety, Tolerability, Pharmacokinetics and Pharmacodynamics of Oral Administration of RO7020531: (1). Single and Multiple Ascending Doses in Healthy Male and Female Subjects; (2). 6-week Treatment of Patients With Chronic Hepatitis B Virus Infection

NCT Number: NCT02956850

Document Date: Protocol Version 7: 24-June-2020

PROTOCOL

TITLE:	**A PHASE I, SPONSOR-OPEN, INVESTIGATOR-BLINDED, SUBJECT-BLINDED, MULTI-CENTER, PLACEBO-CONTROLLED STUDY TO EVALUATE SAFETY, TOLERABILITY, PHARMACOKINETICS AND PHARMACODYNAMICS OF ORAL ADMINISTRATION OF RO7020531: (1). SINGLE AND MULTIPLE ASCENDING DOSES IN HEALTHY MALE AND FEMALE SUBJECTS; (2). 6-WEEK TREATMENT OF PATIENTS WITH CHRONIC HEPATITIS B VIRUS INFECTION**
PROTOCOL NUMBER:	NP39305
VERSION:	7
EUDRACT NUMBER:	2016-003723-38
TEST PRODUCT:	RO7020531
SPONSOR:	F. Hoffmann-La Roche Ltd
DATE FINAL:	Version 1.0: 22 September 2016
DATES AMENDED:	Version 2.0: 07 April 2017 Version 3.0: 24 July 2017 Version 4.0: 22 September 2018 Version 5.0: 22 March 2019 Version 6.0: 20 November 2019 Version 7.0: see electronic time stamp below

FINAL PROTOCOL APPROVAL

Date and Time (UTC)	Title	Approver's Name
24-Jun-2020 12:47:48	Company Signatory	[redacted]

CONFIDENTIAL

PROTOCOL ACCEPTANCE FORM

TITLE:	**A PHASE I, SPONSOR-OPEN, INVESTIGATOR-BLINDED, SUBJECT-BLINDED, MULTI-CENTER, PLACEBO-CONTROLLED STUDY TO EVALUATE SAFETY, TOLERABILITY, PHARMACOKINETICS AND PHARMACODYNAMICS OF ORAL ADMINISTRATION OF RO7020531: (1). SINGLE AND MULTIPLE ASCENDING DOSES IN HEALTHY MALE AND FEMALE SUBJECTS; (2). 6-WEEK TREATMENT OF PATIENTS WITH CHRONIC HEPATITIS B VIRUS INFECTION**
PROTOCOL NUMBER:	NP39305
VERSION:	7
EUDRACT NUMBER:	2016-003723-38
TEST PRODUCT:	RO7020531
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 7: RATIONALE

Protocol NP39305 Version 7 has been amended as summarized below:

- The mode of action of RO7020531 (toll-like receptor (TLR)7 agonist) includes interferon α (IFN-α) induction and a broad spectrum of immune modulation. This amendment introduces a RO7020531 dose reduction to 100 mg QOD and a RO7020531 dosing regimen of 100 mg QW for patients who are treatment naïve (Part 2 Cohort 4) to provide flexibility and choice to Investigators and patients and to help manage better the patients' tolerability issues with flu-like symptoms resulting from the RO7020531 mode of action (Sections 3.1.2.2, 4.4.2.1, 4.4.2.3, and 5.2.1; and Appendix 1 Schedule of Assessments: CHB Patients Who Are Not on Antiviral Treatment (Part 2 Cohort 4). Of note, these dose modifications are not due to any new safety signal.
- To add an additional scenario that is not considered to be a serious adverse event (SAE) (Section 5.3.5.10).
- To clarify the prophylactic and symptomatic use of acetaminophen (paracetamol) for flu-like symptoms (Sections 4.5.1.2 and 4.7.1.1).
- To allow greater flexibility in patient recruitment by allowing re-screening of patients who failed screening once (Section 4.6.2.1).
- To update previous clinical experience in RO7020531 Studies NP39305 and YP39553 (Section 1.2.2) and other TLR 7 agonist (GS-9620) studies (Section 1.1).

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

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PROTOCOL SYNOPSIS

TITLE:	**A PHASE I, SPONSOR-OPEN, INVESTIGATOR-BLINDED, SUBJECT-BLINDED, MULTI-CENTER, PLACEBO-CONTROLLED STUDY TO EVALUATE SAFETY, TOLERABILITY, PHARMACOKINETICS AND PHARMACODYNAMICS OF ORAL ADMINISTRATION OF RO7020531: (1). SINGLE AND MULTIPLE ASCENDING DOSES IN HEALTHY MALE AND FEMALE SUBJECTS; (2). 6-WEEK TREATMENT OF PATIENTS WITH CHRONIC HEPATITIS B VIRUS INFECTION**
PROTOCOL NUMBER:	NP39305
VERSION:	7
EUDRACT NUMBER:	2016-003723-38
TEST PRODUCT:	RO7020531
PHASE:	I
INDICATION:	Chronic Hepatitis B Virus Infection
SPONSOR:	F. Hoffmann-La Roche Ltd

OBJECTIVES

Primary Objectives

Part 1: Single Ascending Dose and Multiple Ascending Dose in Healthy Volunteers

The primary objective is:

- To assess the safety and tolerability of single ascending doses (SAD) and multiple ascending doses (MAD) of RO7020531 administered orally to healthy volunteers (HVs).

Part 2: Chronic Hepatitis B Patients

The primary objective is:

- To assess the safety and tolerability of 6 weeks of treatment with RO7020531 administered orally to chronic hepatitis B (CHB) patients.

Secondary Objectives

Part 1: SAD and MAD in Healthy Volunteers

The secondary objectives are:

- To investigate the plasma pharmacokinetics (PK) of RO7020531, the main active metabolite RO7011785, and additional metabolites, including RO7018822 and RO7033805, following single and multiple ascending oral doses of RO7020531.
- To investigate the urine PK of RO7020531, the main active metabolite RO7011785, and additional metabolites, including RO7018822 and RO7033805, in pooled urine samples of healthy subjects after single ascending oral doses of RO7020531.
- To investigate the effect of RO7020531 on pharmacodynamic (PD) parameters following single ascending doses and multiple ascending doses administered every other day (QOD).

- To evaluate the effect of RO7020531 dosing on electrocardiogram (ECG) parameters after single and multiple ascending oral doses using exposure-response analysis.

Part 2: Chronic Hepatitis B Patients

The secondary objectives are:

- To investigate the plasma PK of RO7020531, the main active metabolite RO7011785, and additional metabolites, including RO7018822 and RO7033805, in CHB patients.
- To investigate the PD markers of TLR7 activation, including cytokines and interferon-stimulated genes (ISGs), following administration of RO7020531 to patients with CHB.

Exploratory Objectives

The exploratory objective for Part 2 (CHB patients) is:

- To investigate the antiviral effect of 6 weeks of treatment with RO7020531 in CHB patients.

STUDY DESIGN

Description of Study

Part 1 will be a randomized, Sponsor-open, Investigator-blinded, subject-blinded, placebo-controlled, SAD and MAD study to evaluate the safety, tolerability, PK, and PD of RO7020531 and metabolites following oral administration to HVs. PK, PD, safety and tolerability data collected in the SAD portion of this study will be used to determine doses at which to initiate the MAD portion of the study.

Part 2 will commence when the data from SAD and MAD in HVs support the progression into the 6-week study in CHB patients after Internal Monitoring Committee (IMC) review. This may occur before all MAD HV cohorts have been completed but not before data from at least the first MAD cohort have been reviewed. It will be a multiple-center, randomized, Sponsor-open, Investigator-blinded, patient-blinded, placebo-controlled study to investigate the safety, tolerability, PK and PD of treatment with RO7020531 for 6 weeks in CHB patients.

NUMBER OF STUDY SUBJECTS

Part 1: SAD and MAD in Healthy Volunteers

Approximately eight HV cohorts may be evaluated in the SAD portion of this study (approximately 80 subjects in total), and approximately three cohorts are anticipated for the MAD (approximately 30 subjects in total). In each cohort, eight subjects will be treated with RO7020531 and two with placebo. A minimum of two females per cohort should be randomized, with at least one female receiving active drug. Depending on the data collected in each cohort, additional cohort(s) may be added in the SAD and MAD portions to collect the necessary information for selecting appropriate doses in patients.

Part 2: Chronic Hepatitis B Patients

Approximately 20-30 virologically suppressed CHB patients may be randomized into at least two cohorts in Part 2 of the study (Cohort 3 will be optional) and up to 15 treatment-naïve CHB patients may be randomized in the optional Cohort 4. Within each dose level, up to 15 patients (8-12 active and 2-3 placebo) will be randomized. A minimum of two females per cohort should be randomized in Part 2, with at least one female per cohort receiving active drug.

TARGET POPULATION

Part 1: SAD and MAD in Healthy Volunteers

The study population consists of healthy male and female subjects, aged 18 to 65 years, inclusive.

Part 2: Chronic Hepatitis B Patients

The study populations in Cohorts 1, 2 and 3 consist of male and female virologically-suppressed CHB patients aged 18 to 65 years, inclusive, who have been on tenofovir, entecavir, adefovir, or telbivudine, either as single agents or in combination, for at least 6 months.

The study population in Cohort 4 consists of HBV treatment-naïve or not on treatment for the past 6 months CHB patients aged 18 to 65 years, inclusive.

INCLUSION/EXCLUSION CRITERIA

Inclusion Criteria

Part 1: SAD and MAD in Healthy Volunteers

HVs must meet the following criteria for study entry:

1. Healthy male and female subjects, 18 to 65 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, and urinalysis.
2. Informed of, and 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.
3. For women of childbearing potential: agreement to remain abstinent (refrain from heterosexual intercourse) or use two approved contraceptive methods, of which one must be a barrier method and the other should be an established non-barrier form of contraception with a failure rate of < 1% per year, during the treatment period and for at least one month after the last dose of study drug.
    a. A woman is considered to be of childbearing potential if she is post menarcheal, has not reached a post menopausal state (≥ 12 continuous months of amenorrhea with no identified cause other than menopause), and has not undergone surgical sterilization (removal of ovaries and/or uterus).
    b. Examples of contraceptive methods with a failure rate of < 1% per year include bilateral tubal occlusion, male sterilization, established proper use of hormonal contraceptives that inhibit ovulation, hormone-releasing IUDs, and copper IUDs.
    c. 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.
4. Male subjects must be willing to use two methods of contraception with their partners, one of which must be a barrier method (i.e. condom), for the duration of the study and for one month after the last dose of study medication. Other acceptable forms of contraception for this study include vasectomy, bilateral tubal occlusion, intrauterine device (IUD) or proper use of hormonal contraceptives. Periodic abstinence is not considered an adequate form of contraception. Men must refrain from donating sperm during this same period.
5. Negative pregnancy test on Day -1 for female subjects.
6. A body mass index (BMI) between 18 to 32 kg/m^2, inclusive.
7. Non-smokers, or use of < 10 cigarettes (or equivalent nicotine-containing product) per day.
8. Negative anti-nuclear antibody (ANA) test; or positive with dilutions not greater than 1:40 and with no associated history or symptoms of potential connective tissue disease or other immune-mediated diseases.

Part 2: Chronic Hepatitis B Patients

Patients must meet the following criteria for study entry:

1. Adult male and female patients, 18 to 65 years of age, inclusive.
2. Informed of, and 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.
3. A BMI between 21 to 32 kg/m^2, inclusive. Males must be above 55 kg and females above 45 kg body weight.
4. Chronic hepatitis B infection (positive test for hepatitis B surface antigen (HBsAg) for

more than 6 months prior to randomization).

5. For Cohort 1, 2, 3 and 4: HBsAg detectable at screening.
6. For Cohort 1, 2, and 3: On treatment with tenofovir, entecavir, adefovir, or telbivudine, either as single agents or in combination, for at least 6 months.
   For Cohort 4: HBV treatment naïve or not on any anti-HBV treatment for the past 6 months.
7. For Cohort 1, 2 and 3: HBV DNA < 90 IU/mL for at least 6 months prior to randomization; HBV DNA < 90 IU/mL at screening by Roche Cobas assay.
   For Cohort 4: HBV DNA at screening $\geq 2 \times 10^4$ IU/mL for HBeAg positive and $\geq 2 \times 10^3$ IU/mL for HBeAg negative patients.
8. For Cohort 1, 2 and 3: Alanine amino transferase (ALT) $\leq 1.5 \times$ upper limit of normal (ULN) during the 6 months prior to randomization confirmed by two measurements separated by at least 14 days (one of the ALT measurements can be done at screening); ALT at screening $\leq 1.5 \times$ ULN.
   For Cohort 4: ALT and aspartate aminotransferase (AST) at screening and Day -1 visit: $\leq 5 \times$ ULN.
9. Screening laboratory values (including hematology, chemistry, urinalysis) obtained up to 28 days prior to first study treatment within acceptable range or judged to be not clinically significant by the Principal Investigator (PI) and Medical Monitor.
10. Gamma glutamyl transpeptidase (GGT), alkaline phosphatase (ALP), albumin, total and direct bilirubin within normal range or judged to be not clinically significant by the Investigator and Medical Monitor at screening.
11. Negative ANA test, or positive with dilutions not greater than 1:40 and with no associated history or symptoms of potential connective tissue disease or other immune-mediated diseases.
12. Liver biopsy, Fibroscan® or equivalent elastography test obtained within 6 months prior to randomization demonstrating liver disease consistent with chronic HBV infection with absence of cirrhosis and absence of extensive bridging fibrosis (cirrhosis or extensive bridging fibrosis are defined as ≥Metavir 3, recommended cutoff for fibroscan 8.5 kPa).
13. For women of childbearing potential: agreement to remain abstinent (refrain from heterosexual intercourse) or use two approved contraceptive methods, of which one must be a barrier method and the other should be an established non-barrier form of contraception with a failure rate of < 1% per year, during the treatment period and for at least one month after the last dose of study drug.
    a. A woman is considered to be of childbearing potential if she is post-menarcheal, has not reached a post-menopausal state (≥ 12 continuous months of amenorrhea with no identified cause other than menopause), and has not undergone surgical sterilization (removal of ovaries and/or uterus).
    b. Examples of contraceptive methods with a failure rate of < 1% per year include bilateral tubal occlusion, male sterilization, established proper use of hormonal contraceptives that inhibit ovulation, hormone-releasing IUDs, and copper IUDs.
    c. 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.
14. 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 or pregnant female partners, men must remain abstinent or be willing to use two methods of contraception with their partners, one of which must be a condom and the other should be an established form of contraception, during the treatment period and for at least one month after the last dose of study drug to avoid exposing the embryo. Other acceptable forms of contraception include vasectomy, bilateral tubal occlusion, IUD or proper use of hormonal contraceptives (e.g. contraceptive pills). 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 and withdrawal are not acceptable methods of contraception.

15. Negative pregnancy test on Day -1 for female patients.

Exclusion Criteria

Part 1: SAD and MAD in Healthy Volunteers

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

1. Pregnant (positive pregnancy test) or lactating women, and male partners of women who are pregnant or lactating.
2. History of immunologically mediated disease (e.g., inflammatory bowel disease, idiopathic thrombocytopenic purpura, lupus erythematosus, autoimmune hemolytic anemia, scleroderma, severe psoriasis, rheumatoid arthritis, multiple sclerosis, or any other autoimmune disease).
3. History or symptoms of any clinically significant disease including (but not limited to), neurological, cardiovascular, endocrine, respiratory, hepatic, ocular, or renal disorder (as per investigator's judgment).
4. Personal or family history of congenital long QT syndrome or sudden cardiac death.
5. Evidence of an active or suspected cancer or a history of malignancy, where in the Investigator's opinion, there is a risk of recurrence.
6. History of having received or currently receiving any systemic anti-neoplastic (including radiation) or immune-modulatory treatment (including systemic oral or inhaled corticosteroids, IFN or PEG-IFN) within the 8 weeks prior to the first dose of study drug or the expectation that such treatment will be needed at any time during the study. Eye drop-containing and infrequent inhaled corticosteroids are permissible up to 4 weeks prior to the first dose of study drug.
7. History of clinically significant thyroid disease; also, subjects with clinically significant elevated thyroid-stimulating hormone (TSH) concentrations at screening.
8. Any confirmed clinically significant allergic reactions (anaphylaxis) against any drug, or multiple drug allergies (non-active hay fever is acceptable).
9. History of clinically significant psychiatric disease, especially major depression (significant psychiatric disease is defined as treatment with an antidepressant medication or a major tranquilizer at therapeutic doses for major depression or psychosis, respectively, or any history of the following: a suicide attempt, hospitalization for psychiatric disease, or a period of disability due to a psychiatric disease).
10. Clinically significant acute infection, e.g., influenza, local infection or any other clinically significant illness within two weeks of randomization.
11. History of clinically significant gastrointestinal (GI) disease including inflammatory bowel disease, peptic ulcer disease, GI hemorrhage.
12. Confirmed systolic blood pressure (BP) greater than 140 or less than 90 mmHg, and diastolic BP greater than 90 or less than 50 mmHg at screening (based on the average of 3 separate resting BP measurements, properly measured with well-maintained equipment, after at least 5 minutes rest).
13. Clinically relevant ECG abnormalities on screening ECG: e.g.,
    a. QTc interval (QTcF > 450 msec or < 300 msec)
    b. Notable resting bradycardia (HR < 45 bpm), or HR > 90 bpm
    c. Difference between highest and lowest of any screening QTc > 30 msec
    d. ECGs with documented machine errors in the interval duration assessments
    e. 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 QRS onset, low amplitude T-wave, merged T- and U-waves, prominent U-waves)

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

14. Any of the following laboratory parameters prior to dosing:
    a. White blood cells (WBC) < 3000 cells/mm^3
    b. Neutrophil count < 1500 cells/mm^3
    c. Platelet count < 140,000 cells/mm^3
    d. Activated partial thromboplastin time (aPTT) > 40 seconds, international normalized ratio (INR) > 1.2
    e. Hb < 12 g/dL in females or 13 g/dL in males
15. Abnormal renal function including serum creatinine > ULN or calculated CrCl < 70 mL/min (using the Cockcroft Gault formula).
16. ALT or AST values at screening above ULN and judged clinically significant by the Investigator.
17. Positive results for anti-mitochondrial antibody (AMA), anti-smooth muscle antibody (ASMA) or thyroid peroxidase antibody.
18. Positive Hepatitis A virus antibody (HAV Ab IgM), HBsAg, Hepatitis C antibody (HCV Ab) or positive for human immunodeficiency virus (HIV) at screening.
19. 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 or judged to be clinically irrelevant for healthy subjects.
20. History of alcohol abuse (consumption of more than 2 standard drinks per day on average; 1 standard drink = 10 grams of alcohol) and/or drug abuse within one year of randomization. Alcohol consumption will be prohibited at least 48 hours before screening, 48 hours before admission until discharge from the clinic, and 48 hours before each scheduled visit.
21. Positive test for drugs of abuse or positive alcohol test at screening or Day − 1.
22. Any clinically significant concomitant disease or condition that could interfere with, or for which the treatment of 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.
23. Use of any medication (prescription or over-the-counter [OTC], including health supplements, vitamins or herbal remedies) within the 2 weeks prior to the first dosing or within 5 half-lives of the medication prior to first dosing (whichever is longer). Exceptions may be made on a case-by-case basis following discussion and agreement between the Investigator and the Sponsor.
24. Participation in an investigational drug or device study within 90 days prior to randomization.
25. Donation or loss of blood over 500 mL, or administration of any blood product, within 90 days prior to starting study medication.
26. Subjects under judicial supervision, guardianship or curatorship.
27. Any medical or social condition which may interfere with the subject's ability to comply with the study visit schedule or the study assessments.

Part 2: Chronic Hepatitis B Patients

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

1. Pregnant (positive pregnancy test) or lactating women.
2. History of liver cirrhosis.
3. History or other evidence of bleeding from esophageal varices.
4. Decompensated liver disease (e.g., Child-Pugh Class B or C clinical classification or clinical evidence such as ascites or varices).

5. History or other evidence of a medical condition associated with chronic liver disease other than HBV infection (e.g., hemochromatosis, autoimmune hepatitis, alcoholic liver disease, toxin exposure, thalassemia, nonalcoholic steato-hepatitis, etc.). A clinical diagnosis of fatty liver is allowed provided that non alcoholic steatohepatitis (NASH) has been excluded by liver biopsy.
6. Documented history or other evidence of metabolic liver disease within one year of randomization.
7. Positive test for Hepatitis A virus (IgM anti-HAV), Hepatitis C virus (HCV), Hepatitis D virus, Hepatitis E virus (HEV), or human immunodeficiency virus (HIV).
8. Expected to need systemic antiviral therapy other than that provided by the study at any time during their participation in the study, with the exception of oral therapy for Herpes simplex virus type I (HSV I) or HSV II.
9. History of or suspicion of hepatocellular carcinoma or alpha fetoprotein $\geq$ 13 ng/mL at screening.
10. History of immunologically mediated disease (e.g., inflammatory bowel disease, idiopathic thrombocytopenic purpura, lupus erythematosus, autoimmune hemolytic anemia, scleroderma, severe psoriasis, rheumatoid arthritis, multiple sclerosis, or any other autoimmune disease).
11. History of clinically significant cardiovascular (postural hypotension), endocrine, renal, ocular, pulmonary or neurological disease (as per Investigator's judgment).
12. History of clinically significant GI disease including inflammatory bowel disease, peptic ulcer disease, GI hemorrhage, or history of pancreatitis.
13. History of clinically significant psychiatric disease, especially major depression (significant psychiatric disease is defined as treatment with an antidepressant medication or a major tranquilizer at therapeutic doses for major depression or psychosis, respectively, or any history of the following: a suicide attempt, hospitalization for psychiatric disease, or a period of disability due to a psychiatric disease).
14. Active or suspected cancer or a history of malignancy, where in the Investigator's opinion, there is a risk of recurrence
15. History of having received or currently receiving any systemic anti-neoplastic (including radiation) or immune-modulatory treatment (including systemic oral or inhaled corticosteroids, IFN or PEG-IFN) within the 8 weeks prior to the first dose of study drug or the expectation that such treatment will be needed at any time during the study. Eye drop-containing and infrequent inhaled corticosteroids are permissible up to 4 weeks prior to the first dose of study drug.
    Cohort 4: Concurrent HBV treatments.
16. History of organ transplantation.
17. Clinically significant thyroid disease; also, patients with clinically significant elevated TSH concentrations at screening.
18. Any confirmed clinically significant allergic reactions (anaphylaxis) against any drug, or multiple drug allergies (non-active hay fever is acceptable).
19. Clinically significant acute infection (e.g., influenza, local infection) or any other clinically significant illness within 2 weeks of randomization.
20. Clinically relevant ECG abnormalities on screening ECG.
21. Any of the following laboratory parameters at screening:
    a. WBC $<$ 3,000 cells/mm^3
    b. Neutrophil count < 1500 cells/mm^3
    c. Platelet count < 140,000 cells/mm^3
    d. aPTT $>$ 40 seconds, INR $>$ 1.2
    e. Hb $<$ 12 g/dL in females or 13 g/dL in males
22. Abnormal renal function including serum creatinine $>$ ULN or calculated CrCl $<$ 60 mL/min (using the Cockcroft Gault formula).

23. Positive results for AMA, ASMA or thyroid peroxidase antibody.
24. Participation in an investigational drug or device study within 30 days prior to randomization.
25. Donation or loss of blood over 500 mL, or administration of any blood product, within 90 days prior to starting study medication.
26. History of alcohol abuse (consumption of more than 2 standard drinks per day on average; 1 standard drink = 10 grams of alcohol) and/or drug abuse within one year of randomization.
27. Positive test for drugs of abuse or positive alcohol test at screening or Day -1. For positive cannabinoids test, the eligibility is at the Investigator's discretion.
28. Patients under judicial supervision, guardianship or curatorship.
29. Any medical or social condition which may interfere with the patient's ability to comply with the study visit schedule or the study assessments.

LENGTH OF STUDY

Part 1: SAD and MAD in Healthy Volunteers

SAD: The total duration of the study will be up to 9 weeks (from screening through study completion) for each randomized subject as follows:

- Screening: Up to 28 days;
- Dosing period: 1 day;
- Follow up: 28 days after dosing.

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

- Screening: Up to 28 days;
- Dosing period: 14 days;
- Follow up: 28 days after last dosing.

Part 2: Chronic Hepatitis B Patients

The total duration of the study will be up to 16 weeks (from screening through study completion) for each randomized patient as follows:

- Screening: Up to 28 days;
- Dosing period: 6 weeks;
- Follow up: 6 weeks after last dosing.

END OF STUDY

The end of the study is defined as the date when the last subject last observation (LSLO) occurs. LSLO is expected to occur 12 weeks after the last patient in Part 2 *(i.e., last patient in Cohort 4)* is randomized.

OUTCOME MEASURES

SAFETY OUTCOME MEASURES

The safety outcome measures for HVs (Part 1) and patients (Part 2) are as follows:

- Incidence and severity of adverse events (AE).
- Incidence of laboratory abnormalities based on hematology, clinical chemistry (including liver function tests), coagulation and urinalysis test results.
- Incidence of vital signs (blood pressure, pulse rate, respiratory rate and body temperature) or ECG (PR [PQ], QRS, QT, QTcF) abnormalities.

A detailed medical history and physical examination will be performed at the time-points indicated in the SoA. Height will only be recorded at screening.

AEs and concomitant medications will be monitored throughout the entire study (screening through follow-up) as defined by International Conference on Harmonization (ICH) guidelines.

Monitoring for liver flares will be conducted for the duration of the CHB patient study (Part 2).

PHARMACOKINETIC OUTCOME MEASURES

The PK evaluations for HVs (Part 1) and patients (Part 2) are as follows:

- Summary descriptive statistics of plasma PK parameters for RO7020531, the main active metabolite RO7011785, and additional metabolites, including RO7018822 and RO7033805, will be computed. These parameters include C_{max}, T_{max}, AUC_{inf}, AUC_{last} and $t_{1/2}$ and will be presented by dose cohorts including mean, standard deviation (SD), coefficient of variation (CV), medians and ranges.
- The total amount of RO7020531, the main active metabolite RO7011785 and additional metabolites, including RO7018822 and RO7033805, in urine over a 24 hour period will be computed and provided in tables and listings.
- In Cohorts 1, 2 and 3 of Part 2 of the study, sparse sampling for tenofovir (including tenofovir alafenamide, if approved for HBV and applicable), entecavir, adefovir and telbivudine will be made. As appropriate, tables and listings of these concentrations will be provided.

PHARMACODYNAMIC OUTCOME MEASURES

Part 1: SAD and MAD in Healthy Volunteers

- Blood samples will be collected to evaluate a number of immune PD outcome measures including, but not limited to, the protein and metabolite markers (neopterin, IFN-α, IP-10, TNF-α, IL-6, IL-10, IL-12p40), and markers of transcriptional responses (ISG15, OAS-1, MX1 and TLR7).

Part 2: Chronic Hepatitis B Patients

- Blood samples will be collected to evaluate a number of immune PD outcome measures including, but not limited to, the protein and metabolite markers (neopterin, IFN-α, IP-10, TNF-α, IL-6, IL-10, IL-12p40) and markers of transcriptional responses (ISG15, OAS-1, MX1 and TLR7).
- Additional immune PD assessments (proteins and mRNA transcripts) may be added to those listed above as needed.

EXPLORATORY OUTCOME MEASURES

Part 2 only: Chronic Hepatitis B Patients

HBV Antiviral Measures

The antiviral outcome measures for this study are the following:

- Quantitative HBV DNA
- HBsAg (qualitative)
- HBsAg (quantitative)
- Hepatitis B $early$ antigen (HBeAg) (qualitative)
- Anti-HBe and anti-HBs antibody status
- Additional exploratory virology laboratory tests, such as HBsAg/anti-HBsAg complex levels, HBeAg levels (semi-quantitative assessment based on signals in the HBeAg assay), HBV core and core related antigens, anti-HBc antibody and HBV RNA in serum may be assessed at the time-points specified in the SoA as an evaluation of potentially exploratory markers of therapeutic response, in conjunction with the viral parameters listed above.

Outcomes of antiviral response will include quantitative HBeAg decline, loss of HBeAg, development of anti-HBe, HBeAg seroconversion (loss of HBeAg and presence of anti-HBe), quantitative HBsAg decline, loss of HBsAg, development of anti-HBs, HBsAg seroconversion (loss of HBsAg and presence of anti-HBs), and maintenance of HBV DNA levels less than 90 IU/mL for Cohorts 1,2 and 3 (at the end of the treatment period and at the end of the follow-up period). For Cohort 4, the outcome of antiviral response will additionally include quantitative HBV DNA level (actual and change from baseline).

Monitoring of viral resistance will be performed in any patient who experiences virological

breakthrough.

Viral genotype may be assessed in CHB patients (Part 2). Viral genotype is known as a predictive factor to response to PEG-IFN treatment (Sonneveld et al 2012).

Other Exploratory Measures

Other exploratory outcome measures for Part 1 and 2 of this study which may be investigated include, but are not limited to, the following:

- Clinical genotyping samples will be collected in all HVs/patients. Genotyping may be conducted as appropriate to explore the impact of genetic polymorphism on drug metabolism, transport, PD response, efficacy, or the safety profile of RO7020531.
- Immunophenotyping might be performed by flow cytometry to assess changes in count and activation status of selected immune cells from Part 1 MAD and Part 2 and its relevance for its treatment response.
- Total transcriptome analysis may be performed from whole blood RNA samples to identify biomarkers potentially predictive of antiviral and/or pharmacodynamic responses.

RESEARCH BIOSAMPLE REPOSITORY (RBR) SAMPLE COLLECTION (optional)

The following RBR samples will be collected for identification of dynamic (non-inherited) biomarkers from HVs/patients who sign RBR informed consent:

- Leftover plasma samples
- Leftover serum samples
- Leftover blood samples

The following samples will be collected for identification of genetic (inherited) biomarkers:

- Whole blood for DNA extraction to assess for biomarkers, including but not limited to single nucleotide polymorphisms (SNPs)

INVESTIGATIONAL MEDICINAL PRODUCT

Test Product

The study drug RO7020531 will be provided in hard gelatin capsules for oral administration containing 1 mg, 10 mg, or 100 mg of RO7020531 drug substance. The study drug must be stored according to the details on the product label: "Store at 2°C – 8°C, protect from light and moisture".

Placebo

The placebo will be provided in hard gelatin capsules identical in size and appearance to the corresponding active capsules, containing microcrystalline cellulose of compendial grade but no active substance.

Part 1: SAD and MAD in Healthy Volunteers

In the SAD cohorts, RO7020531 or matching placebo will be administered orally to the subjects in a fasted state. The SAD portion of the study will include an adaptive number of cohorts (approximately eight). The anticipated dose-escalation sequence for SAD is 3 mg, 10 mg, 20 mg, 40 mg, 60 mg, 100 mg, 140 mg, and 170 mg.

In the MAD cohorts, RO7020531 or matching placebo will be administered orally to the subjects, in a fasted state, QOD from Day 1 through to Day 13. In total, seven doses will be given (on Day 1, Day 3, Day 5, Day 7, Day 9, Day 11, and Day 13). The MAD portion of the study will include an adaptive number of cohorts (approximately three). Dose levels for the MAD portion of the study will be defined during the study conduct based on emerging safety and tolerability data from the SAD portion of the study.

Part 2: Chronic Hepatitis B Patients

The starting dose level in patients will be determined upon completion of Part 1. RO7020531 or matching placebo will be administered orally to patients, in a fasted state, QOD for 6 weeks (*unless in Cohort 4 in case of QW dosing as dose modification*). In total, *up to* 21 doses will be given). In Cohorts 1, 2 and 3, RO7020531 or matching placebo will be administered together with the patient's nucleos(t)ide analogue (NUC) treatment (tenofovir, entecavir, adefovir, or

telbivudine either as single agents or in combination). In Cohort 4, RO7020531 or matching placebo will be administered alone.

NON-INVESTIGATIONAL MEDICINAL PRODUCTS

Nucleos(t)ide Analogues

Tenofovir, entecavir, adefovir and telbivudine are marketed drugs and will be used in the study for an authorized indication per local label. They are not considered as Investigational Medicinal Products (IMP).

During the study treatment and follow-up periods, patients in Cohorts 1, 2 and 3 of Part 2 of the study will continue the use of the NUCs as prescribed by each patient's physician and following the general warnings and precautions in the local labels.

PROCEDURES

Informed consent will be obtained prior to any study-specific procedures. Following eligibility check at screening, patients will be randomized into the study. The assessments and examinations will be conducted as described in the Schedule of Assessments (SoA).

STATISTICAL METHODS

SAFETY ANALYSES

All safety analyses will be based on the safety analysis population.

All subjects who receive at least one dose of the study drug will be included in the safety analysis. The safety data, including AE, reasons for study drug discontinuation and withdrawal from study, laboratory data, ECG, concomitant medications, vital signs, and physical examination results, will be listed and summarized descriptively.

As appropriate, listings, summary tables and graphs (subject plot and/or mean plots) will be provided for safety and tolerability assessments.

AEs will be listed and summarized by body system and preferred term using the Medical Dictionary for Drug Regulatory Affairs (MedDRA).

For laboratory data and vital signs: values will be presented by individual listings with flagging for values outside normal ranges and for abnormalities.

PHARMACOKINETIC ANALYSES

Non-compartmental analysis using WinNonlin software will be used to calculate PK parameters where appropriate. Summary descriptive statistics of plasma PK parameters including C_{max}, T_{max}, AUC_{inf}, AUC_{last} and $t_{1/2}$ for RO7020531 and RO7011785 and additional metabolites including RO7018822 and RO7033805, will be presented by treatment arm including mean, SD, CV, medians and ranges. Where appropriate, data may be pooled and analyzed, for example, all single dose data may be pooled. Listings, summary tables and graphs (individual plots and/or mean plots) by treatment group will be provided. Descriptive statistics of urine PK parameters for RO7020531 and RO7011875 and additional metabolites including RO7018822 and RO703380 will be presented, where available. PK and PD data from this study may be used to develop a population PK/PD model.

HVs/patients will be excluded from the PK analysis if data are unavailable which may influence the analysis. Where appropriate, listings and summary tables of tenofovir (including tenofovir alafenamide, if approved for HBV and applicable), entecavir, adefovir, and telbivudine concentrations will be provided based on the sparse sampling throughout the study.

PHARMACODYNAMIC ANALYSES

Summary descriptive statistics will be presented for the induction of cytokines, chemokines, and neopterin and of interferon-response genes expression separately by treatment arm. Exploratory analysis will be performed to assess the interferon-induced response under different dosing conditions. Graphical and statistical techniques including linear, nonlinear, and logistic regression will be used to explore potential relationships between dosing regimen, PK and PD.

EXPLORATORY ANALYSES

All antiviral endpoints are considered exploratory for this study (Part 2 only). Summary descriptive statistics will be used to summarize the antiviral outcome measures of qualitative HBsAg, quantitative HBsAg actual and change from baseline, and HBeAg, anti-HBe and anti-HBs status at each of the time-points by treatment group. For Cohort 4, the outcome of antiviral response will additionally include quantitative HBV DNA level (actual and change from baseline). Status of HBsAg and HBeAg seroconversion, if any, and maintenance of HBV DNA levels less than 90 IU/mL (Cohorts 1, 2 and 3) will also be summarized. The findings from the resistance analyses will be listed for each patient selected for analysis.

Exploratory analysis may also be performed to identify markers and/or marker panels correlating with and/or potentially predictive of an in vivo PD response, the occurrence of certain AEs (to be defined) and/or changes in viral parameters.

A detailed exploratory analysis plan will be developed when initial study results (AE, viral/antiviral parameters in Part 2) have been reviewed.

SAMPLE SIZE JUSTIFICATION

Part 1: SAD and MAD in Healthy Volunteers

The current planned study design and sample size complies with standard safety review rules applied in single and multiple ascending dose studies.

Part 2: Chronic Hepatitis B Patients

Sample size is determined based on clinical judgment and practical considerations. With eight patients treated with RO7020531, there is a 90% chance to observe at least one adverse event if the underlying event incidence rate is 25% in the patient population.

Interim Analyses

There will be no Interim Analysis for this study.

LIST OF PROHIBITED MEDICATIONS

Part 1: SAD and MAD in Healthy Volunteers

Prohibited medications include:

- Any prescribed or OTC medications, including health supplements, vitamins or herbal remedies within 2 weeks prior to the first dosing or within 5 half-lives of the medication prior to first dosing (whichever is longer), until the follow up visit (Day 8 for SAD and Day 20 for MAD).
- Any systemic anti-neoplastic (including radiation) or immune-modulatory treatment (including systemic oral or inhaled corticosteroids, interferon (IFN) or PEG-IFN) at any time within the 8 weeks prior to the first dose of study drug and until the follow up visit (Day 8 for SAD and Day 20 for MAD). Eye drop-containing and infrequent inhaled corticosteroids are permissible up to 4 weeks prior to the first dose of study drug.

Part 2: Chronic Hepatitis B Patients

Prohibited medications include:

- Any systemic anti-neoplastic (including radiation) or immune-modulatory treatment (including systemic oral or inhaled corticosteroids, IFN or PEG-IFN) at any time within the 8 weeks prior to the first dose of study drug and until the end of follow-up period. Eye drop-containing and infrequent inhaled corticosteroids are permissible up to 4 weeks prior to the first dose of study drug.
- Any systemic antiviral therapy other than nucleos(t)ide analogues (Cohorts 1, 2 and 3 only), at any time from 30 days before screening until the end of follow-up period with the exception of oral therapy for HSV I or HSV II. Investigational drugs or herbal and other remedies which are being taken by the patient for possible or perceived effects against HBV are prohibited.

Special Concomitant Medications in Chronic Hepatitis B Patients (Part 2)

The following medications are not prohibited in Part 2 of the study. However, caution is required in case of co-administration with the study drug, and the Sponsor should be informed before these drugs are administered.

- Drugs that reduce renal function or compete for active tubular secretion may increase serum concentrations of either study drug or the co-administered drug.
- Drugs metabolized by aldehyde oxidase including famciclovir and/or inhibiting this enzyme including tamoxifen, raloxifen, cimetidine, promethazine, clozapine and chlorpromazine, which could decrease the formation of RO7011785.

LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation	Definition
AAV	Adeno-associated virus
AE	Adverse events
ALP	Alkaline phosphatase
ALT	Alanine aminotransferase
AMA	Anti-mitochondrial antibody
ANA	Anti-nuclear antibody
anti-HBc	Antibody to HBcAg
anti-HBe	Antibody to HBeAg
anti-HBs	Antibody to HBsAg
aPTT	Activated partial thromboplastin time
ASMA	Anti-smooth muscle antibody
AST	Aspartate aminotransferase
AUC	Area under the concentration-time curve
$AUC_{0-\infty}$	Area under the plasma concentration versus time curve extrapolated to infinity
AUC_{inf}	Area under the plasma concentration versus time curve extrapolated to infinity
AUC_{last}	Area under the plasma concentration versus time curve up to the last measurable concentration
BMI	Body mass index
BP	Blood pressure
cccDNA	Covalently closed circular DNA
CD	Cluster of differentiation
CHB	Chronic hepatitis B
CL	Clearance
C_{max}	Maximum observed plasma concentration
CRO	Contract research organization
CRM	Continual Reassessment Method
CV	Coefficient of variation
DAIDS	Division of AIDS
DDI	Drug-drug interaction
DLE	Dose-limiting event
DNA	Deoxyribonucleic acid
ECG	Electrocardiogram
eCRF	Electronic case report form
EDC	Electronic data capture

Abbreviation	Definition
EEA	European economic area
EFD	Embryo fetal development
ESF	Eligibility Screening Form
EU	European Union
FSH	Follicle-stimulating hormone
GGT	Gamma glutamyl transpeptidase
GI	Gastrointestinal
GLP	Good Laboratory Practice
HAV	Hepatitis A virus
Hb	Hemoglobin
HbA1c	Glycated hemoglobin
HBcAg	Hepatitis B core antigen
HBeAg	Hepatitis B *early* antigen
HBsAg	Hepatitis B surface antigen
HBV	Hepatitis B virus
HCG	Human chorionic gonadotropin
HCV	Hepatitis C virus
HEV	Hepatitis E virus
HIV	Human immunodeficiency virus
HR	Heart rate
HSV	Herpes simplex virus
HV	Healthy volunteer
IB	Investigator's Brochure
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IFN	Interferon
IgD	Immunoglobulin D
IgM	Immunoglobulin M
IL	Interleukin
IMC	Internal Monitoring Committee
IMP	Investigational medicinal product
IND	Investigational New Drug
INR	International normalized ratio
IP-10	Interferon gamma-inducible protein 10
IRB	Institutional Review Board
ISG	Interferon-stimulated gene
IUD	Intrauterine device

Abbreviation	Definition
IxRS	Interactive (voice/web) response system
LOAEL	Lowest observed adverse effect level
MAD	Multiple ascending dose
MCH	Mean corpuscular hemoglobin
MCHC	Mean corpuscular hemoglobin concentration
MCV	Mean corpuscular volume
mDC	Myeloid dendritic cell
mRNA	Messenger RNA
MX1	Myxovirus resistance 1 gene
NASH	Non-alcoholic steatohepatitis
NK	Natural killer cell
NOAEL	No-observed-adverse-effect level
NUC	Nucleoside/nucleotide analogue
OAS	Oligoadenylate synthetase
OATP	Organic anion transporting polypeptide
PBMC	Peripheral blood mononuclear cells
PD	Pharmacodynamic
pDC	Plasmacytoid dendritic cell
PEG-IFN	Pegylated interferon
P-gp	P-glycoprotein
pH	Measure of acidity or alkalinity
PK	Pharmacokinetic
PQ	PQ interval
PR	PR interval
PT	Prothrombin time
QOD	Every other day
QRS	QRS complex
QT	QT interval
QTc	Corrected QT interval
QTcF	Fridericia's correction of QT interval
QW	Once a week
RBC	Red blood cell
RBR	Research Biosample Repository
RNA	Ribonucleic acid
RR	RR interval
SAD	Single Ascending Dose
SAE	Serious Adverse Event

Abbreviation	Definition
SD	Standard Deviation
SI	Système International d'Unités
SNP	Single nucleotide polymorphism
SoA	Schedule of Assessments
$t_{1/2}$	Half-life
TBNK	T-cells, B-cells and NK-cells
TLR	Toll-like receptor
T_{max}	Time to maximum observed plasma concentration
TNF	Tumor necrosis factor
TSH	Thyroid-stimulating hormone
ULN	Upper limit of normal
V_{ss}	Volume of distribution at steady-state
WBC	White blood cell
WGS	Whole genome sequencing
WHO	World Health Organization

1. BACKGROUND AND RATIONALE

1.1 BACKGROUND ON DISEASE

Chronic hepatitis B (CHB) and its sequelae are major global healthcare problems. Despite the implementation of effective vaccination in many countries, hepatitis B is one of the most common infectious diseases in the world. It is estimated that more than 2 billion people or one third of the world's population have been infected with the hepatitis B virus (HBV) at some time in their lives and an estimated 240 million are now chronically infected (WHO 2002, WHO 2016). Nearly 25% of all chronic HBV carriers develop serious liver diseases such as chronic hepatitis, cirrhosis, and primary hepatocellular carcinoma. More than 686 000 people die every year due to the consequences of hepatitis B (WHO 2016).

The endemicity of HBV varies substantially by region, with East Asia and sub-Saharan Africa having prevalence rates of CHB above 8% (Ott et al 2012). In these highly endemic areas, the most common means of transmission is by perinatal infection, and up to 90% of the population has serological evidence of prior infection (Alter et al 2003). Although prevalence levels in developed countries are relatively low, immigration from highly endemic regions has had a significant influence on the local need for therapy, and even countries with low endemicity are currently experiencing the burden of CHB (Wasley et al 2010).

HBV belongs to the Hepadnaviridae family. It is a partly double-stranded DNA virus with approximately 3200 base pairs. The transcriptional template of HBV is the covalently closed circular DNA (cccDNA), which resides inside the hepatocyte nucleus as a minichromosome (Locarnini et al 2010). Several HBV subtypes have been identified. Most CHB patients are infected with the wild-type strain of HBV, which produces large amounts of the hepatitis B *early* antigen (HBeAg) resulting in the HBeAg-positive form of CHB. However, in a significant proportion of patients, variant forms of the virus predominate later in the course of the disease, which have diminished ability to produce HBeAg. Another serological marker, hepatitis B surface antigen (HBsAg), is a hallmark of the infection and remains persistently positive in CHB patients. There is a correlation between the presence of HBsAg and patients' outcome with HBsAg level being predictive of fibrosis severity, development of hepatocellular carcinoma and survival rates (Fattovich et al 1998, Tseng et al 2012, Martinot-Peignoux et al 2013).

HBV is not cytopathic: both liver damage and viral control are immunomediated (Trepo et al 2014). The clinical outcome of infection is dependent on the complex interplay between HBV replication and both the innate and adaptive immune responses. The dominant cause of the long-term viral persistence and pathogenesis of HBV liver disease is the development of an inefficient antiviral response to the viral antigens (Bertoletti et al 2012).

Currently available treatments for CHB include interferon (IFN), pegylated-interferon (PEG-IFN), and nucleos(t)ide analogues (NUC): lamivudine, adefovir, entecavir, tenofovir and telbivudine (Papatheodoridis et al 2012; Sarin et al 2016; Terrault et al 2016). Although these therapies achieve long-term effects in lowering HBV DNA levels, chronic HBV infection cannot be completely eradicated with currently approved therapeutics due to the persistence of cccDNA in the nucleus of infected hepatocytes (Lucifora et al 2014). With these treatments, rates of HBsAg clearance and seroconversion, which are associated with reduced or reversed cirrhosis and prevention of HCC development, are low (<15% HBsAg seroconversion after 1 to 5 years follow-up) (Chang et al 2010,Marcellin et al 2013). In addition, the notable deficiencies of current HBV treatments include indefinite duration of NUCs and risk of viral resistance with some NUC treatments, while PEG-IFN therapy is poorly tolerated and a significant portion of patients do not have a virological response (Papatheodoridis et al 2008).

Due to the therapeutic limitations of the currently available agents for the management of HBV infection, there is a need for new treatments of CHB that can provide clinical cure (HBsAg loss) and sustained suppression of HBV replication (Wang and Chen 2014).

Toll-like receptors (TLRs) are a family of pathogen-recognition receptors that activate the innate immune response. Stimulation of TLRs leads to the release of multiple cytokines, including type I and type II IFNs, to the induction of pathways and enzymes that destroy intracellular pathogens, and to the maturation of professional antigen-presenting cells, resulting in the activation of the adaptive immune response (Iwasaki and Medzhitov 2004). To date, 11 functional TLRs have been identified in humans. Most TLRs are located in the plasma membrane, except TLR3, TLR7, TLR8 and TLR9, which are intracellularly expressed, particularly in endosomes. TLR7 receptors are able to recognize viral components and induce IFN production and downstream responses (Lester and Li 2014).

A number of small molecule agonists for TLR7 have been identified (Horscroft et al 2012). The stimulation of TLR7 mediates an endogenous type I IFN response, which is critical in the development of a broad, effective and protective immunity against hepatitis viruses (Horscroft et al 2012, Funk et al 2014). Compared to PEG-IFN therapy, treatment with a TLR7 agonist induces broader immuno-modulatory effects that are likely to lead to more effective control and functional cure of chronic HBV infection (Strader et al 2004, Isogawa et al 2005). TLR7 agonists induce the production of multiple isotypes of IFN from plasmacytoid dendritic cells (pDCs) which have been shown in vitro to possess additive or synergistic antiviral effects compared to exogenous PEG-IFN. *A TLR7 agonist, GS-9620, has been evaluated in Phase 2 studies with a once a week (QW) dosing regimen (in combination with NUC treatment) in both NUC-suppressed patients and in patients not currently on treatment. While QW dosing of GS-9620 demonstrated broad immune modulatory effects, it did not affect the levels of HBsAg (Janssen et al 2018 and Agarwal et al 2018). Therefore, a finite treatment of more intensified TLR7 dosing regimen (e.g. every other day [QOD]) in combination with*

other mode of action anti-HBV drugs is more likely to achieve functional cure (permanent loss of HBsAg) of CHB.

1.2 BACKGROUND ON RO7020531

RO7020531, an oral double prodrug of the TLR7-specific agonist, RO7011785, is being developed for the treatment of CHB patients. A prodrug approach was chosen for oral delivery of the TLR7 agonist RO7011785 in order to improve bioavailability and limit TLR7 activation in the gastrointestinal (GI) tract, which may be associated with GI intolerability. Non-clinical studies with RO7020531 suggest that it is rapidly converted to the active metabolite RO7011785. Data from in vivo studies with RO7020531 and in vitro studies with RO7011785 support immune activation as the mechanism of action.

See the RO7020531 Investigator Brochure (IB) for details on non-clinical studies.

1.2.1 Previous Non-Clinical Studies

1.2.1.1 Non-Clinical Pharmacology

RO7011785 showed in vitro potency to activate TLR7 and triggered downstream TLR7-mediated nuclear factor-kappa B (NF-κB) signaling in an engineered HEK293 reporter cell line expressing human TLR7. RO7011785 was shown to be a selective hTLR7 agonist with less potency to activate hTLR8 in vitro. Ex vivo stimulation of human peripheral blood mononuclear cells (PBMCs) from healthy donors by RO7011785 resulted in dose-dependent induction of various cytokines and chemokines, including IFN-α, tumor necrosis factor-α (TNF-α), interleukin-6 (IL-6), and IFN gamma-inducible protein 10 (IP-10).

1.2.1.2 HBV Animal Model

The anti-HBV activity of RO7020531 was evaluated in an HBV mouse model. In this model, C57BL/6 mice were injected with recombinant adeno-associated virus (AAV) harboring 1.3 copies of the HBV genome, and demonstrated high levels of HBV viral makers including HBV DNA, HBsAg, and HBeAg. Oral administration of RO7020531, QW or QOD for 6 weeks, proved to have in vivo anti-HBV activity in this mouse model, i.e., clear reduction in HBV DNA and HBsAg (by more than 1 log after the 42-day treatment period). With the same administration frequency, higher doses resulted in faster and greater reduction in HBV DNA and HBsAg. With the same dose of 100 mg/kg, QOD administration resulted in greater viral reduction on Day 42 than QW administration. In addition, RO7020531 induced interferon-stimulated gene (ISG) expression in peripheral whole blood in this mouse model.

1.2.1.3 Non-Clinical Pharmacokinetics and Metabolism

RO7020531 showed high oral bioavailability and rapid conversion to the active moiety, RO7011785. RO7011785 exhibited a relatively short half-life ($t_{1/2}$) of 1.51 hours in rats and 2.12 hours in monkeys, moderate systemic clearance (CL) of ~50% liver blood flow in rats, and CL of ~30% liver blood flow in monkeys. RO7011785 had a moderate volume of distribution (V_{ss}) in the range of body water or slightly higher for both rats and

monkeys. Differences in metabolite exposure were noted between species, although RO7011785 was the predominant metabolite in both rats and monkeys, confirming good conversion to active drug.

Conversion of RO7020531 to the active form RO7011785 was observed in animal and human hepatocytes, with both the ester hydrolysis and aldehyde oxidase-mediated pathways active. The relative strength of the oxidase versus esterase pathways varied between species and influenced the concentrations of the different intermediate prodrugs: RO7033805 was the predominant intermediate for cynomolgus monkey, whereas RO7018822 was the predominant intermediate for mouse, rat, and human.

RO7020531 was found to be a good substrate of multidrug resistance protein 1 (MDR1; P-glycoprotein [P-gp]), with a high apparent membrane permeability. Therefore, in vivo drug-drug interactions (DDI) due to co-administration with MDR1 inhibitors cannot be excluded. However, interactions of strong MDR1 inhibitors with the absorption of RO7020531 are considered unlikely due to the high permeability, high solubility, and high oral bioavailability of RO7020531, which makes it unlikely that MDR1 plays a major role in RO7020531 transintestinal absorption in vivo.

RO7011785 can be directly glucuronidated by UDP-glucuronosyltransferase 1A1 (UGT1A1); thus, drug-drug interactions with UGT1A1 inhibitors cannot be excluded. Drug-drug interaction potential was low, as determined in vitro with cytochrome P450 (CYP) enzymes and with transporters (organic anion transporting polypeptide 1B1 [OATP1B1], OATP1B3, organic anion transporter 1 [OAT1], OAT3, and breast cancer resistance protein [BCRP]). Therefore, the interactions with CYPs and substrates for hepatic uptake transporters and kidney transporters (for example NUCs) in vivo are unlikely.

1.2.1.4 Toxicology and Safety Pharmacology

Repeat-dose toxicity studies with treatment duration of up to 13 weeks have been performed with RO7020531 in rats and monkeys. The effects observed were considered to be consistent with exaggerated pharmacology, i.e., TLR7 agonism leading to the intended immune/cytokine stimulation and subsequent tolerability or proinflammatory findings. QOD and QW dosing regimens were investigated in Good Laboratory Practice (GLP) toxicology studies; QW dosing was better tolerated. The no-observed-adverse-effect-level (NOAEL) for the 13-week rat toxicology study was 3 mg/kg QOD (area under the concentration–time curve for males and females [AUC M/F] = 186/503 h • ng/mL) and 30 mg/kg QW (AUC M/F = 2950/4660 h • ng/mL). The NOAEL for the 13-week monkey toxicology study was 3 mg/kg QOD (AUC M/F = 858/706 h • ng/mL) and 10 mg/kg QW (AUC M/F = 3340/2960 h • ng/mL).

Neither the double prodrug RO7020531, nor its metabolites RO7011785 and RO7018822, significantly affected potassium channels in the in vitro human ether-à-go-go-related gene (hERG) assay. Dosing at ≥3 mg/kg in monkeys caused reversible heart rate increase (with decreased RR interval and shortened QT and/or heart-rate corrected QT [QTc] interval), non-adverse increase in PR interval and increase in body temperature. There were no RO7020531-related effects on respiratory or central nervous system (CNS) function in rats up to the highest dose tested (300 mg/kg).

No genotoxic potential was observed for RO7020531 in the Ames test, nor in the in vitro and in vivo micronucleus test. No evidence of teratogenicity or embryo-fetal toxicity was observed in definitive (GLP) embryo-fetal toxicity studies in rats (up to 150 mg/kg/day, with $AUC_{0\text{-}24h}$ of 13500 ng•h/mL) or rabbits (up to 100 mg/kg/day, with $AUC_{0\text{-}24h}$ of 46700 ng•h/mL), other than increased incidences of non-adverse fetal skeletal variations. In a definitive (GLP) fertility study in male and female rats, no adverse effects of RO7011785 on mating performance, fertility, or early embryonic development were noted up to the highest tested dose of 300 mg/kg QOD, despite slightly reduced body weight gain in both males and females.

See the RO7020531 IB for details on the properties of RO7020531 and RO7011785.

1.2.2 Previous Clinical Experience

Study NP39305 is the first-in-human study of RO7020531.

As of *01 May 2020*, 110 healthy volunteers (HVs) *in Part 1,* and 30 CHB patients *from Cohorts 1 to 3 and 9 CHB patients from Cohort 4 from Part 2 have* been dosed in *Study NP39305*. The data from subjects *in Part 1 and from patients in Cohort 1 in Part 2* were reviewed by IMC and shared with investigators. The treatment with RO7020531/placebo at all dose levels (Part 1: 3 mg, 10 mg, 20 mg, 40 mg, 60 mg, 100 mg, 140 mg, 170 mg; Part 2: 150 mg) was considered safe and had acceptable tolerability. *An event of flu-like syndrome of moderate intensity was reported from a treatment naïve CHB patient from Part 2 Cohort 4 on Day 3. It was assessed as serious as it required or prolonged hospitalization and as related to the study drug by the investigator, and it resolved on Day 5.* Adverse events (AEs) potentially related to pharmacodynamic (PD) effects expected with TLR7 agonist treatment were reported at dose levels >100 mg, including three HVs and seven CHB patients. They showed reversible signs and symptoms of immune activation after repeated dosing (e.g. flu-like symptoms) that resolved without sequelae. Apart from transient cytopenia observed in one of the patients, which improved during the dosing period, there have been no clinically significant changes in any other safety parameters, including laboratory assessments, vital signs and ECGs.

RO7020531 showed a predictable PK profile. Similar to pharmacokinetics in single (SAD) and multiple (MAD) ascending dose cohorts from HVs, the active TLR7 agonist,

RO7011785, appeared rapidly in plasma following 150 and 170 mg dosing of CHB patients on Day 1 with a T_{max} ranging from 1.0 to 2.0 hr and was eliminated relatively quickly from plasma with a mean terminal half-*life* of ~4 hours.

Individual C_{max} and $AUC_{0\text{-}inf}$ of RO7011785 following 150 and 170 mg of RO7020531 in CHB patients appears to be within the range of prediction from SAD/MAD healthy volunteer data. However, the mean RO7011785 AUC0-inf of 2814 ng*hr/mL on Day 1 is slightly higher (~15%) than the expected value, with greater inter-subject variability, and numerically comparable to mean exposure associated with the monkey lowest observed adverse effect level (LOAEL) (2780 ng*hr/mL).

After Study NP39305 had been started, Study YP39553 *was initiated. YP39553 was* a Phase 1 *bridging* study to evaluate the PK, PD, and safety/tolerability of RO7020531/placebo in Chinese HV in 4 SAD cohorts (40, 100, 140, and 170 mg) and three MAD cohorts (100 mg, 150 mg and 150 mg). *This study has been completed. A total* of 70 Chinese HV *were* dosed in study YP39553. The treatment with RO7020531/placebo was safe with acceptable tolerability when administered orally as a single dose of up to 170 mg and as multiple doses up to 100 mg QOD. A total of nine subjects (8 subjects from the MAD 150 mg cohorts and one subject from the SAD 170 mg cohort) experienced pyrexia/flu-like symptoms after the first, second, or the third dose of the study drug. Of these nine subjects with flu-like symptoms, *six* had AEs of moderate intensity and *three* had AEs of mild intensity. In all these subjects, there were no clinically significant changes in vital signs (except pyrexia) and ECGs. Unscheduled laboratory tests performed at the time of pyrexia (5.5 to 12 hours post dose) revealed that seven subjects from MAD 150 mg cohorts had Grade 1 to 4 lymphopenia and the Principal Investigator decided to discontinue further drug administration in these seven subjects. Lymphocyte counts returned to normal levels within 24 to 48 hours in all subjects. One subject from the 150 mg MAD dose cohort decided to withdraw from the study after the fifth dose.

Following single oral doses of RO7020531, the PK of the active metabolite RO7011785 is linear and mean AUC0-inf increased in a dose proportional manner from 40 mg to 170 mg. No PK accumulation was observed following dosing QOD dosing regimen over the 2-week period. Both single and multiple doses of RO7020531 resulted in dose-dependent increases in TLR7 response marker at the dose of 100 mg or above, and the flu-like symptoms are associated with higher PD response.

1.3 STUDY RATIONALE AND BENEFIT–RISK ASSESSMENT

1.3.1 Study Rationale

The current protocol, NP39305, is the first clinical study of RO7020531, designed to assess the safety, tolerability, PK and PD of oral SADs and multiple ascending doses (MAD) administered to healthy male and female volunteers (Part 1); and safety, tolerability, PK and PD effects of treatment with RO7020531 for 6 weeks in CHB patients

(Part 2). Based on the results of this entry-into-human (EIH) study, further development of RO7020531 will include combination studies with other agents to treat CHB.

All SAD and MAD cohorts will be dosed in the fasted state. Dose-escalation will proceed following evaluation of the safety and tolerability of the available data from all previous cohorts.

In the SAD portion of the study, safety and tolerability of ascending doses will be the primary endpoints for dose-escalation. In addition, PK and PD measured by the appearance of select biomarkers indicating activation of a TLR pathway will be evaluated. These biomarkers will be evaluated for each cohort and will be used to decide whether to initiate the MAD portion of Part 1. When PD biomarkers indicating TLR activity are detected within a particular SAD cohort and the dose in that cohort is considered to have adequate safety and tolerability, then that dose may be evaluated in the first MAD cohort, but only if the next higher single dose is also considered safe and well-tolerated. Evidence for TLR-related PD activity will be provided by measuring post-dose changes of selected biomarkers that are known to be activated following stimulation of TLR7 and TLR8. These biomarkers include the protein and metabolite markers IFN-α, IL-12p40, IL-10, IP-10, TNF-α, IL-6, and neopterin and markers of transcriptional responses (ISG15, OAS-1, MX1 and TLR7). The single dose above the dose found to elicit TLR activation will be evaluated in the SAD prior to moving to the MAD. Dose-escalation in the SAD will not proceed if stopping rules are met or if the mean RO7011785 exposure (AUC) reaches the exposure found to be associated with adverse effects in the monkey GLP toxicology studies.

The MAD portion is designed to dose subjects with RO7020531 using a QOD dose regimen that will proceed for two weeks. Previous clinical and non-clinical data with another TLR7 agonist, RO6864018, have shown that the QOD dose schedule provides additional priming of TLR7 response biomarkers and is thought to be a more effective dose regimen mechanistically for HBV treatment. This approach to the HV MAD will therefore allow characterization of safety, PK and PD following QOD dosing. The dose will not be escalated in the HV MAD if stopping rules have been met or if the mean RO7011785 exposure (AUC) reaches the exposure found to be associated with adverse effects in the monkey GLP toxicology studies (~2.8 $\mu g \cdot h/mL$).

Part 2 will involve HBeAg-positive and HBeAg-negative virologically suppressed CHB patients, as well as HBeAg-positive patients with CHB who are currently not on antiviral treatment. In Cohorts 1, 2 and 3 of Part 2 of the study RO7020531 will be administered to study patients together with the NUCs they have been taking (tenofovir, entecavir, adefovir, or telbivudine, either as single agents or in combination), which are globally marketed drugs (for the rationale of enrollment of NUC-suppressed patients; see Section 3.2.2.2). Based on preclinical evidence, no drug–drug interaction (DDI) is anticipated with the concomitant NUC treatment. In Cohort 4 of Part 2 of the study RO7020531 will be administered to study patients who are treatment-naïve or not on

treatment for the past 6 months. The primary objective of Part 2 will be the assessment of safety and tolerability of multiple doses of RO7020531 administered for 6 weeks, followed by 6 weeks of follow-up. Based on non-clinical and clinical experience with another TLR7 agonist, RO6864018, safety issues should be apparent within 6 weeks of dosing. This study will allow better understanding of the safety and PD in CHB patients before going on to longer dosing in subsequent studies. The goal of the treatment with RO7020531 in CHB patients is to induce immune cell activation and local cyto/chemokines while limiting AEs related to systemic IFN-α.

1.3.2 Benefit-Risk Assessment

No prior clinical experience with RO7020531 exists. The evaluation of potential risks of treatment and specific tests, observations, and precautions required for clinical studies with RO7020531 are based on information from non-clinical toxicology and safety pharmacology studies, and on information from clinical studies with other TLR7 agonists. Safety and tolerability will be carefully assessed, and HVs/patients will be closely monitored. The RO7020531 IB (Guidance to the Investigator) summarizes potential risks and key risk management activities to consider when dosing this compound.

[redacted] In two Phase I studies with RO7020531, 19 subjects (dosed with 140 mg or higher) developed flu-like symptoms with initial onsets usually after the first, second, or the third dose. *In addition, in Study NP39305 Part 2 Cohort 4, one patient experienced an SAE of flu-like syndrome (see Section 1.2.2).* The flu-like symptoms were mild in the majority of these subjects, [redacted]

As for all early clinical studies in HVs, there is no direct benefit to the subjects participating in Part 1 of the study. However, this study will be essential in the development of a new treatment for chronic HBV infection, and for selecting the most appropriate dose for a study in HBV-infected patients. It is possible that no long-term benefit to CHB patients may occur in Part 2 of this study; however, data generated in these patients will enable further development of RO7020531 as potentially part of a clinical cure for CHB.

2. OBJECTIVES

2.1 PRIMARY OBJECTIVES

2.1.1 Part 1: SAD and MAD in Healthy Volunteers

The primary objective is:

- To assess the safety and tolerability of single and multiple ascending doses of RO7020531 administered orally to HVs.

2.1.2 Part 2: Chronic Hepatitis B Patients

The primary objective is:

- To assess the safety and tolerability of 6 weeks of treatment with RO7020531 administered orally to CHB patients.

2.2 SECONDARY OBJECTIVES

2.2.1 Part 1: SAD and MAD in Healthy Volunteers

The secondary objectives are:

- To investigate the plasma PK of RO7020531, the main active metabolite RO7011785, and additional metabolites, including RO7018822 and RO7033805, following single and multiple ascending oral doses of RO7020531.
- To investigate the urine PK of RO7020531, the main active metabolite RO7011785, and additional metabolites, including RO7018822 and RO7033805, in pooled urine samples of healthy subjects after single ascending oral doses of RO7020531.
- To investigate the effect of RO7020531 on PD parameters following single ascending doses and multiple ascending doses administered QOD.
- To evaluate the effect of RO7020531 dosing on electrocardiogram (ECG) parameters after single and multiple ascending oral doses using exposure-response analysis.

2.2.2 Part 2: Chronic Hepatitis B Patients

The secondary objectives are:

- To investigate the plasma PK of RO7020531, the main active metabolite RO7011785, and additional metabolites, including RO7018822 and RO7033805, in CHB patients.
- To investigate the PD markers of TLR7 activation, including cytokines and ISGs, following administration of RO7020531 to patients with CHB.

2.3 EXPLORATORY OBJECTIVES

The exploratory objective for Part 2 (CHB patients) is:

- To investigate the antiviral effect of 6 weeks of treatment with RO7020531 in CHB patients.

3. STUDY DESIGN

3.1 DESCRIPTION OF STUDY

Part 1 will be a randomized, Sponsor-open, Investigator-blinded, subject-blinded, placebo-controlled, SAD and MAD study to evaluate the safety, tolerability, PK, and PD of RO7020531 and metabolites following oral administration to HVs. PK, PD, safety and tolerability data collected in the SAD portion of this study will be used to determine doses at which to initiate the MAD portion of the study.

Part 2 will commence when the data from SAD and MAD in HVs support the progression into the 6-week study in CHB patients after Internal Monitoring Committee (IMC) review as detailed in Section 3.1.2.2. This may occur before all MAD HV cohorts have been completed but not before data from at least the first MAD cohort have been reviewed. Part 2 will be a multiple-center, randomized, Sponsor-open, Investigator-blinded, patient-blinded, placebo-controlled study to investigate the safety, tolerability, PK and PD of treatment with RO7020531 for 6 weeks in patients with CHB infection.

3.1.1 Overview of Study Design

3.1.1.1 Part 1: SAD in Healthy Volunteers

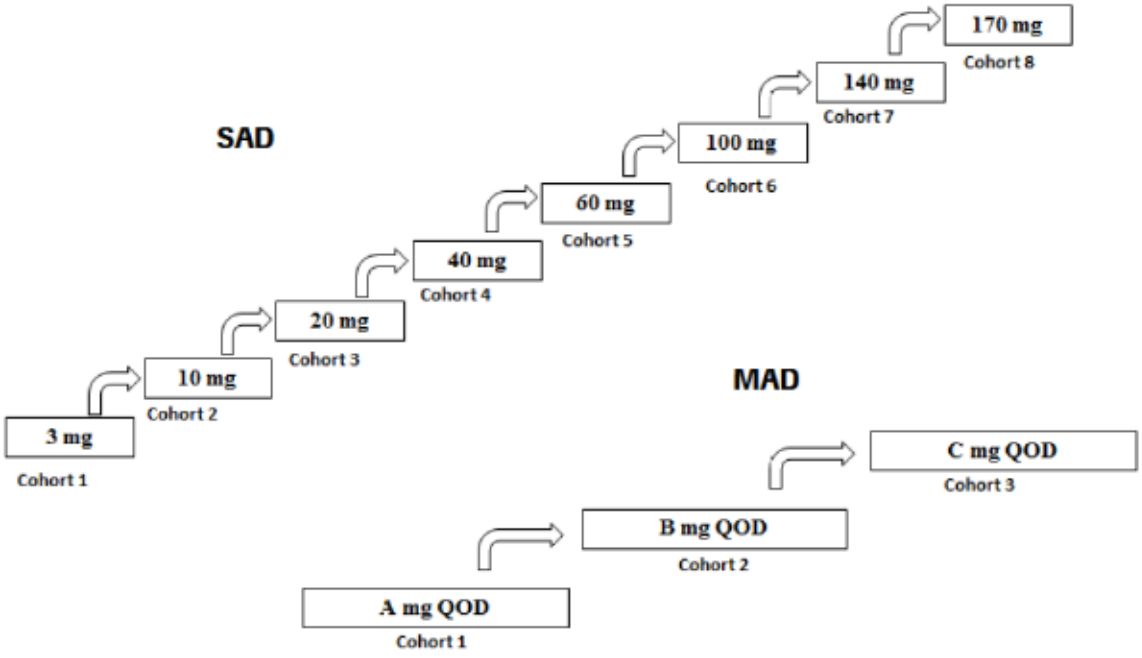
HVs will be screened up to 28 days before randomization into the SAD portion of this study. For dose-escalation, volunteers will be sequentially randomized into one of approximately eight dose cohorts (Figure 1). The anticipated ascending dose scheme is 3 mg, 10 mg, 20 mg, 40 mg, 60 mg, 100 mg, 140 mg, and 170 mg. Additional cohorts may be added, and the doses chosen for evaluation in the SAD may be modified based on data from the previous cohorts. Initial dose-escalation will allow RO7011785 exposures in humans up to those documented at the NOAEL in the most sensitive species from the 13-week GLP toxicology studies (i.e., rats dosed 3 mg/kg QOD with an AUC of 0.4 μg • h/mL). If adequate safety and tolerability are documented in humans, further dose-progression may be explored but not until a consensus agreement is reached between the Sponsor and the Investigator, and endorsed by the Independent Ethics Committee (IEC). Exposures higher than those associated with adverse effects in the monkey GLP toxicology studies (mean RO7011785 AUC ~2.8 μg • h/mL) are not to be explored in this study.

Due to anticipated variability in PD, cohort size will be fixed at eight subjects randomly assigned to RO7020531 and two subjects randomly assigned to placebo. Each cohort will include a minimum of two females, with at least one female receiving active drug. Safety and tolerability at each dose level will determine eligibility for dose-escalation. Measurement of PK and select PD markers will be made with each cohort and, upon availability, will be utilized along with safety data to determine an initial dose to initiate MAD in HVs. The SAD in HVs may continue until the maximum mean RO7011785 AUC of ~2.8 μg • h/mL is reached based on the monkey GLP toxicology assessment or until dose-limiting toxicities are documented (see Section 3.1.2.1 for dose escalation criteria and stopping rules).

The MAD portion of this study in HVs may be initiated before the SAD portion is completed. Therefore, both the SAD and MAD portions of this study in HVs may run in parallel. All single and multiple doses in HVs will be administered in the fasted state. As an additional safety precaution in this EIH study, the subjects participating in each single dose cohort will be dosed according to a sentinel dosing design. Initially, two subjects will be dosed: one subject with RO7020531 and one subject with placebo. If the safety and tolerability results from the first 24 hours following dosing for the initial subjects are acceptable to the Investigator, the other subjects of each cohort may be dosed within shorter intervals.

In the SAD cohorts, volunteers will be asked to remain in the unit from Day -2 until Day 3 with dosing occurring on Day 1. After being discharged on Day 3, subjects will return for an outpatient clinic visit on Day 5 and for a follow-up outpatient visit on Day 8. All subjects will have a follow-up phone call 28 days after the study drug administration. Subjects will be asked to report any AEs that occur during this period.

Figure 1 Study Design for Part 1: SAD and MAD in Healthy Volunteers*



*The figure shows anticipated doses for SAD cohorts 7 and 8.

3.1.1.2 Part 1: MAD in Healthy Volunteers

The trigger to start the first cohort in the MAD includes documentation of adequate safety and tolerability in the SAD. In addition, evidence in at least two subjects within a particular dose cohort exhibiting responses above placebo-defined baseline for select PD biomarkers should be demonstrated. These biomarkers include the protein and metabolite markers IFN-α, IL-12p40, IL-10, IP-10, TNF-α, IL-6, and neopterin and markers of transcriptional responses (ISG15, OAS-1, MX1 and TLR7). Once criteria have been reached to determine an initial dose for MAD, and there is evidence that the next higher dose is also well-tolerated as a single dose, then the MAD part will be initiated at the defined dose level. HVs will be screened up to 28 days before randomization into the MAD portion of this study. Only doses covered by the dose range evaluated in the SAD part will be used in the MAD part. The MAD study in HVs will consist of approximately 3 dose levels (Figure 1). The cohort size will be eight subjects randomly assigned to RO7020531 and two subjects randomly assigned to placebo. Each cohort will include a minimum of two female subjects, with at least one female on active drug.

Subjects randomized in the MAD study will be asked to remain in the unit from Day -2 until Day 14 with QOD dosing initiated on Day 1. In total, seven QOD doses will be administered with the last dose given on Day 13. Subjects will be discharged on Day 14.

After completing 7 QOD doses, each subject in the MAD cohorts will return for an outpatient follow-up visit 7 days after the last dose. All subjects will have a follow-up phone call 28 days after their last dose. Subjects will be asked to report any AEs that occur during this period.

Upon safety review (see Section 3.1.2), dose-escalation will proceed in the MAD part of this study until a maximal tolerated dose (MTD) or a mean RO7011785 AUC of ~2.8 μg • h/mL is reached, or until it is determined that a suitable dose evaluation has been made by the Sponsor. The maximum dose investigated in MAD will not exceed doses evaluated in the SAD part.

Dose-escalation decisions in the MAD may utilize a Continual Reassessment Method (CRM) and may be modified accordingly. Information gained in the MAD part in HVs will be used to guide dose-selection for the study in CHB patients.

Based on the PK, PD and safety evaluation of the previous dose cohorts, additional cohorts of HVs may be enrolled at higher or lower dose levels, or at a dose level already tested.

3.1.1.3 Part 2: Chronic Hepatitis B Patients

Part 2 will commence when the data from SAD and MAD in HVs support the progression into the 6-week study in CHB patients after IMC review as detailed in Section 3.1.2.2 This may occur before all MAD HV cohorts have been completed but not before data from at least the first MAD cohort have been reviewed. The decision to start the patient part and the recommendation for the starting dose level will be shared with investigators and the Ethics Committees/Institutional Review Boards before first dosing of a patient. Part 2 will consist of up to approximately four dose levels (Figure 2). Dose cohorts will randomize CHB patients who will be screened up to 28 days before randomization into the study. Patients in dose cohorts will receive study drug or placebo on a QOD schedule. Treatment duration in all cohorts will be 6 weeks with 6 weeks of follow-up. All doses will be given to patients in fasted state (at least 2 hours after a meal or 2 hours before the next meal).

Cohorts 1, 2, and 3 will include eight patients receiving RO7020531 and two patients receiving placebo. Each cohort in Part 2 will include a minimum of two female patients, with at least one female patient receiving RO7020531. The dose level for the starting dose and subsequent doses in patients will be defined by the IMC based on a comprehensive review of available data in HV (all safety and PK, and select PD data), and will be lower than or equal to the dose that has been studied and has acceptable tolerability in the HV MAD part of the study.

In Cohorts 1, 2, and 3 all patients will continue taking commercially available NUC treatment (tenofovir, entecavir, adefovir, or telbivudine, either as single agents or in combination) orally once a day as per local label during study participation, including screening, the treatment period and follow-up.

The recruitment will start with 10 patients (8 receiving RO7020531 and 2 receiving placebo) randomized to Cohort 1. The first dose of oral RO7020531 or matching placebo will be given to each patient in the clinic on Day 1.

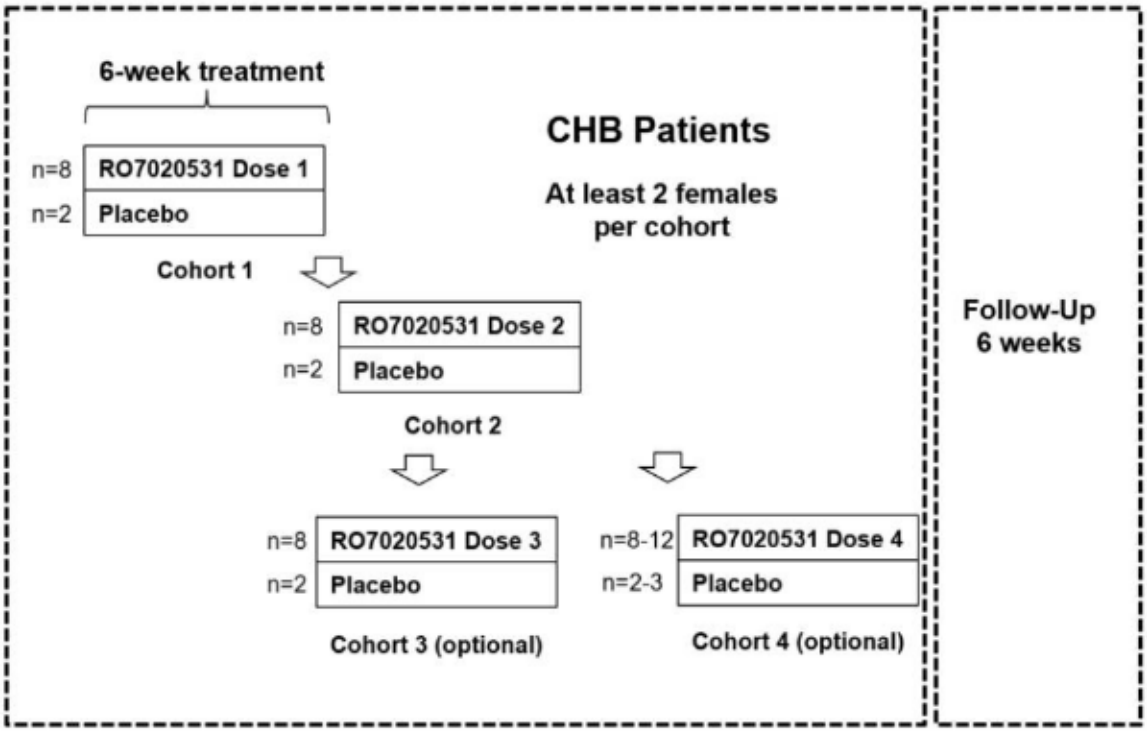
The recruitment will be suspended after the first 10 patients are randomized. When all 10 patients complete 6 weeks of treatment or discontinue study treatment prematurely, the assessment of the safety and tolerability of RO7020531 at the first dose level will be performed by the IMC. Based on the IMC safety review, progression of the study will be defined, including dose level for the next cohort (see Section 3.1.2.2). If the safety and tolerability are acceptable and a decision to open the next cohort is taken by the IMC and acknowledged by the active Investigators, the randomization will resume, and 10 patients will be randomized into Cohort 2.

The review of safety and tolerability in Cohort 2 will be performed in the same fashion by the IMC and will enable opening of optional Cohort 3.

The recommendation for the subsequent cohort dose level will be shared with the IEC/Institutional Review Boards (IRBs). Exploration of doses beyond the maximum proposed dose level will not occur until approval is received from the IEC/IRBs.

Both Cohorts 3 and 4 are optional and the study team may decide to conduct either one of these cohorts, or both. Cohort 4 will enroll CHB patients who are treatment naïve. The dose level for Cohort 4 has been pre-defined at the level of 150 mg RO7020531/placebo for 6 weeks on QOD schedule.

Figure 2 Study Design for Part 2: Chronic Hepatitis B Patients



Follow-Up Period

After 6 weeks of study treatment, a follow-up period of 6 weeks will occur for patients in all treatment arms after receiving the last dose of RO7020531 or placebo.

Optional In-Clinic Stay

To facilitate safety monitoring and the PK/PD assessment, patients will have an option to stay in the clinical unit for one night on Day 1 and/or Day 41. Patients will also have an option to not stay overnight in the clinical unit after the sampling on Day 1 or Day 41 and come to the site in the morning on Day 2 and/or Day 42 as per the SoA (Appendix 1) so that all required PK/PD samples are collected consistently and on time.

Additionally, there will be an optional overnight stay in the clinical unit on Day 1 at the Investigator's discretion for patients in Cohort 4 who develop flu-like symptoms/pyrexia during the first 12 hours post dose and do not respond to symptomatic treatment.

3.1.2 Dose-Escalation Decision Criteria and Stopping Rules

A Safety Review Meeting will be conducted by the Principal Investigator, the Medical Monitor, the Clinical Pharmacologist, and the Sponsor Clinical Team prior to each dose-escalation in the SAD and MAD portions in HVs (Part 1).

IMC meeting(s) will occur to undertake a comprehensive review of available data (all safety and PK, and select PD data) from the SAD and completed MAD HV cohort(s) to decide whether the data support progression into the 6-week study in CHB patients. The IMC will make recommendations for a starting dose, and subsequent doses for the Part 2 of the study. IMC dosing recommendations in Part 2 will be shared with and acknowledged by the active investigators, and shared with IEC/IRBs (see Section 3.1.3 for more details on IMC).

3.1.2.1 Part 1: SAD and MAD in Healthy Volunteers

For all SAD and MAD cohorts, evaluable safety and tolerability data from a minimum of 8 subjects will be required in order to make the decision to escalate to the next dose. The maximum dose explored in HV will not exceed the exposure documented with adverse effects in the GLP toxicology studies.

The decision to escalate to the next dose level in the SAD cohorts will be based primarily on the safety and tolerability information through Day 4 (including AE, ECGs, vital signs, clinical laboratory test results) and, secondarily, on available PK through 24 hours post-dose and PD data at the previous dosage level. In addition, all available safety and PK data from the previous dose level(s) will be reviewed. The anticipated dose-escalation sequence for the SAD is 3 mg, 10 mg, 20 mg, 40 mg, 60 mg, 100 mg, 140 mg and 170 mg. If further cohorts are implemented, dose-escalations or dose modifications will be based on the documented safety and PK but not higher than double from the previous cohort.

The decision to escalate to the next dose level in the MAD cohorts will be based primarily on the safety and tolerability information (including AE, ECGs, vital signs, clinical laboratory test results) through Day 14 and, secondarily, on available PK data. In addition, all available safety and PK data from the previous dose level(s), including SAD cohorts, will be reviewed.

Depending on the evaluation of the safety and/or PK data, the same dose may be repeated; a lower dose may be administered; or intermediate dose-escalation steps other than those anticipated above may be used at the discretion of the Study Investigator and the Sponsor.

The recommendation for the subsequent cohort dose level will be shared with the IEC/IRB. Exploration of doses beyond the maximum proposed dose level will not occur until approval is received from the IEC/IRB.

Planned dose-escalation will be stopped if one of the following circumstances occurs in HVs treated with RO7020531, unless it is apparent that the occurrence is not related to the administration of study medication:

- Within a cohort, three or more HVs on active drug experience:
  - Severe and clinically significant (as defined by the Investigator) RO7020531-related AE of the same character, or
  - Clinically significant RO7020531-related laboratory abnormalities of the same character, or
  - Clinically significant RO7020531-related changes in vital signs or ECGs of the same character (e.g., confirmation of mean QTc 500 msec or 60 msec longer than the pre-dose baseline).
- Within two consecutive dose cohorts, four occurrences of any of the above conditions in HVs receiving active drug.
- Other findings (regardless of the incidence rates) that, at the joint discretion of the Sponsor and the Investigator, indicate dose-escalation should be halted.
- Dose-escalation will not proceed if it is predicted that higher doses of RO7020531 will not result in a further increase in RO7011785 plasma exposure; or if the mean systemic exposure (AUC) reaches the RO7011785 plasma levels found to be associated with adverse effects in the 13-week monkey GLP toxicology studies (i.e., mean AUC ~2.8 μg • h/mL).

If dose-escalation is stopped, lower doses could be investigated by mutual agreement between the Sponsor and the Investigator.

For stopping rules in an individual HV in the MAD study, see Section 4.7.1.1.

3.1.2.2 Part 2: Chronic Hepatitis B Patients

The dose level for the starting dose and subsequent doses in patients will be defined based on a comprehensive review of available safety, PK and select PD data from the SAD and completed MAD cohort(s) in HVs by the IMC (see Section 3.1.3). The starting dose in patients will be lower than or equal to the dose that has been studied and has acceptable tolerability in the MAD part of the study, with evidence of PD effects in HVs (dose not higher than the MTD in HVs).

After the completion of dosing in Cohort 1 of Part 2, the decision on dose level for Cohort 2 will be taken by the IMC upon the review of data once all 10 patients have completed 6 weeks of treatment or have discontinued treatment prematurely (see Section 3.1.3). The IMC decision to open the next cohort will be acknowledged by the active investigators. Dose level selection after Cohort 1 may be guided by predictive models for safety as a function of dose. The dose levels for a subsequent cohort will not be higher than double compared to the previous cohort. The maximum dose in CHB patients will not exceed exposure documented with adverse effects in the monkey GLP toxicology studies.

The decision to open the optional Cohort 3 will be taken in the same fashion by the IMC upon the review of safety and tolerability in Cohort 2.

The dose for optional Cohort 4 has been predefined at the level of 150 mg QOD for 6 weeks. *For dose modifications in this cohort, see Section 4.4.2.3 and Section 5.2.1.*

The recommendation for the subsequent cohort dose level will be shared with the IEC/IRB. Exploration of doses beyond the maximum proposed dose level will not occur until approval is received from the IEC/IRB.

For stopping rules in patients, see Section 4.7.1.1.

3.1.3 Internal Monitoring Committee

For the Safety Review Meeting conducted for dose escalations in Part 1 please see Section 3.1.2.

An IMC will operate in this study and will perform periodic safety data review to ensure that continuation of the study does not pose a health hazard to patients. The IMC will also review any previously unknown or new data from other studies and assess potential additional safety risks of the NP39305 study.

The IMC will perform the following safety data reviews:

- Comprehensive review(s) of available safety and PK, and select PD data from the SAD and completed MAD HV cohort(s) to decide whether the data support progression into the 6-week study in CHB patients and to make recommendations for a starting dose and subsequent doses for the Part 2 of the study;
- Safety of QOD dosing in Cohort 1 will be reviewed once all 10 patients in the Cohort 1 complete 6 weeks of treatment or discontinue treatment prematurely;
- Safety of QOD dosing in Cohort 2 will be reviewed once all 10 patients in the Cohort 2 complete 6 weeks of treatment or discontinue treatment prematurely.

Based on IMC review and acknowledgment by active investigators, progression of the study will be defined, including dose level for the next cohort. IMC recommendations will be shared with IEC/IRBs.

The IMC may perform other periodic or individual patient reviews. Additional review meetings may occur as determined by the IMC. The need for an IMC safety review may be revised as determined by the IMC (e.g. if a dose level is repeated).

Membership of the IMC consists of representatives from Clinical Science (Translational Medicine), Clinical Safety, Biostatistics, Clinical Pharmacology and Statistical Programming and Analysis.

3.1.4 Length of Study and End of Study

Part 1: SAD and MAD in Healthy Volunteers

SAD: The total duration of the study will be up to 9 weeks (from screening through study completion) for each randomized subject as follows:

- Screening: Up to 28 days;
- Dosing period: 1 day;
- Follow up: 28 days after dosing.

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

- Screening: Up to 28 days;
- Dosing period: 14 days;
- Follow up: 28 days after last dosing.

Part 2: Chronic Hepatitis B Patients

The total duration of the study will be up to 16 weeks (from screening through study completion) for each randomized patient as follows:

- Screening: Up to 28 days;
- Dosing period: 6 weeks;
- Follow up: 6 weeks after last dosing.

End of Study

The end of the study is defined as the date when the last subject last observation (LSLO) occurs. LSLO is expected to occur 12 weeks after the last patient in Part 2 *(i.e., last patient in Cohort 4)* is randomized.

3.2 RATIONALE FOR STUDY DESIGN

3.2.1 Rationale for Dosage and Schedule Selection

A starting dose of 3 mg was selected for the SAD portion of this study based on both maximum recommended starting dose (MRSD) (FDA guidance) and exposure multiple calculations. In the 13-week GLP toxicology studies, rats were the most sensitive species with a NOAEL of 3 mg/kg QOD, which converts to a human equivalent dose (HED) of 0.5 mg/kg. Using a safety factor of 10, the MRSD for a 60 kg individual is 3 mg. Using predictions of RO7020531 plasma exposure from physiologically-based PK modeling, under conservative low clearance assumptions, it is predicted that a single dose of 3 mg will lead to a RO7011785 C_{max} of 32 ng/mL and $AUC_{0\text{-}inf}$ of 102 ng • h/mL. This exposure is approximately 10 times lower than the C_{max} in animals at the NOAEL, and approximately 30 times lower than the projected RO7011785 exposure in humans (C_{max} of 980 ng/mL) at therapeutic concentrations.

RO7020531 doses will be escalated following review of the safety, PK and select PD data of all the previous cohorts, and the doses will be designed to elicit systemic exposures of RO7011785 below the concentrations found to be associated with adverse effects in animal studies. Initial dose-escalation will reach up to the exposures at NOAEL in the most sensitive species but could go higher if adequate safety and tolerability are documented in humans; in this case, higher doses will not be explored until a consensus agreement is reached between the Sponsor and the Investigator, and endorsed by the IEC/IRB. Exposures higher than those associated with adverse effects in the 13-week monkey GLP toxicology studies (mean AUC ~2.8 μg • h/mL) are not to be explored in this study.

Dose levels to be explored in CHB patients (Part 2 of the study) will be defined based on emerging data in HVs. The aim of the HBV treatment with TRL7 agonists is an induction of immune cell activation and local cyto- and chemokines with limited AEs related to systemic IFN. It is expected that this can be achieved by investigating several dose levels of RO7020531 in Part 2.

The phenomena of response priming (increase in PD effects with the second dose) and tachyphylaxis (reduced response with chronic and frequent doses) may be expected with RO7020531 based on clinical data observed with another TLR7 agonist, RO6864018. The QOD schedule has been selected for the multiple dosing part of this study in HVs and CHB patients based on information acquired from non-clinical and clinical studies with RO6864018, and aimed to optimize PD responses while minimizing safety risks, such as poor tolerability with every day dosing. It is expected that tachyphylaxis to systemic IFN release might be a desirable effect with the QOD schedule, can potentially limit AE, and improve tolerability.

3.2.2 Rationale for Study Population

3.2.2.1 Part 1: SAD and MAD in Healthy Volunteers

Healthy subjects are chosen for the SAD and MAD portions (Part 1) of this study. Prior clinical PK/PD data are available from both HVs and hepatitis C virus (HCV) patients who received single or multiple oral daily doses of another double prodrug, RO6864018, which is rapidly converted to the TLR7 agonist RO6871765. This in house data and other literature data indicate that PK/PD relationships determined in HVs, HBV and HCV patients were comparable. These data suggest that HVs represent an effective population to evaluate safety, tolerability, PK and PD of TLR agonists that will be predictive of the dose-effect relationships anticipated in CHB patients.

3.2.2.2 Part 2: Chronic Hepatitis B Patients

Cohorts 1, 2 and 3

Patients entering the study will be on tenofovir, entecavir, adefovir, or telbivudine, either as single agents or in combination, for at least 6 months. RO7020531 is not a direct antiviral; therefore, inclusion in the study of virologically suppressed patients permits evaluation of immuno-mediated effects under the nucleotide/nucleoside viremic control. Entecavir and tenofovir are potent HBV inhibitors with a high barrier to resistance. The other NUCs (telbivudine and adefovir) engender higher rates of resistance with long-term monotherapy but are still potent HBV inhibitors and are being used in some instances (Papatheodoridis et al 2012; Sarin et al 2016; Terrault et al 2016). Treatment with NUCs should provide a good basis for the immunomodulatory treatment. Reduction of viral load leads to the partial restoration of immune responses in patients treated with NUCs and improves the function and regulation of host immune cells, including pDCs and NK-cells activity (Tjwa et al 2011; Akbar et al 2011). It was shown that minimizing the HBV viral load by antivirals increases the responsiveness of HBV specific T-cells which are hypo-responsive in cases of persistent HBV infection, and that HBV DNA suppression is an essential requirement for the functional reconstitution of anti-HBV T-cell responses (both CD4 and CD8) (Boni et al 1998; Boni et al 2001; Boni et al 2003).

In addition, virologically suppressed patients should be less likely to develop alanine aminotransferase (ALT) flares resulting from an inflammatory hepatitis than patients with significant viral loads (Agarwal et al 2018). Treatment of virologically suppressed patients is expected to be associated with lower AE rates and fewer treatment discontinuations compared to treatment-naïve patients.

Cohort 4

Cohort 1 of Part 2 of the study has shown good safety and acceptable tolerability in nucleoside/nucleotide analogue (NUC)-suppressed CHB patients dosed with 150 mg RO7020531/ placebo. Based on these data the study will expand the CHB patient population and enroll an exploratory cohort of up to 15 patients (Cohort 4) who are naïve to HBV treatment. This change will allow the evaluation of TLR7 agonist treatment on virological parameters in treatment-naïve patients dosed for 6 weeks.

Studies with TLR7 agonists have shown no clinically significant effect of TLR7 agonist treatment on the level of hepatitis B surface antigen (HBsAg) in NUC-suppressed CHB patients dosed for 12 weeks (Jannsen et al 2018). Thus, no HBsAg change might be expected with 6 weeks of treatment in NP39305 study in Cohorts 1, 2 and 3. However, TLR7 agonists have a potential to decrease HBV DNA due to immune mediated effects (Lau et al 2005). In virally suppressed patients, it is impossible to evaluate effects on HBV DNA, since these patients have undetectable viral load. There have been no studies reported in treatment-naïve CHB patients evaluating monotherapy with a TLR7 agonist. An exploration of antiviral effects in treatment naïve patients will bring value in scientific understanding of TLR7 agonist effects, before evaluation in longer Phase 2 trials or before combination with other antiviral agents.

Health status in study subjects (both Part 1 and Part 2) will be confirmed during screening period. Because of the immunomodulatory mechanism of action of RO7020531, all subjects with a history of immunologically mediated disease will be excluded. All HV/patients will be tested for autoimmune markers to exclude participants with potential connective tissue disease or other immune-mediated diseases.

3.2.2.3 Rationale for Recruitment of Females

This study will randomize at least two females per cohort. Sex differences in TLR7-mediated response have been reported, including higher immune activation and IFN-α production by pDCs in females (Berghofer et al 2006). Females appear to be more sensitive to TLR7 activation than males and exhibit greater PD responses in peripheral blood such as fold change from baseline for several genes and cytokines/chemokines investigated in previous clinical studies of another TLR7 agonist, RO6864018. It was also shown that female HVs appeared to have higher exposure of the active TLR7 agonist, RO6871765. These facts warrant further investigation in this study to evaluate sex differences in PK and PD response, which requires enrollment of females in the study cohorts.

3.2.3 Rationale for Pharmacokinetic Assessments

3.2.3.1 Part 1: SAD and MAD in Healthy Volunteers

As this is a first in human study, extensive serial plasma PK sampling will be taken from the HVs in Part 1 for the SAD and the MAD. In general, these PK data will be used to describe the concentration-time profile for RO7020531 and its metabolites as well as key PK characteristics including elimination half-life, dose/exposure and PK/PD relationships for safety and specific TLR7-dependent biomarkers.

For the SAD in HVs, subjects will be asked to remain in the unit for up to 3 days post-dose for safety monitoring and to collect plasma samples for PK. With the proposed collection of up to 48 hours post-dose, it is assumed that the full concentration-time profile for RO7020531 and its metabolites will be determined. Pooled urine samples will be collected to determine the fraction of RO7020531 and its metabolites that are eliminated in the urine. Previous work with the TLR7 agonist RO6864018 has shown extensive elimination of the active TLR7 agonist RO6871765 in the urine. As both RO7020531 and RO6864018 are from the same chemical class, it is expected that extensive renal elimination of the active TLR7 agonist RO7011785 will occur. It is anticipated that the 24-hour urine collection will allow characterization of this elimination pathway.

For the MAD in HVs, a QOD dosing schedule will be used and extensive PK sampling will be made over a 24-hour period on Day 1 following the first dose and on Day 13 following multiple doses to determine if PK parameters change with multiple doses. With subsequent doses throughout the dosing period, sparse PK sampling will also be used to characterize trough values and C_{max} values to determine that exposure of RO7011785 is maintained during this dose regimen. No urine samples will be collected in the MAD part of the HV study.

3.2.3.2 Part 2: Chronic Hepatitis B Patients

For the MAD in patients, a QOD dosing schedule will be used and a modified PK sampling schedule will be implemented to be less demanding on the patients. In this case, serial PK sampling will be collected on Day 1, Week 1 and on a dosing day within the last week of treatment to determine if PK parameters change with multiple doses. As with the HVs, with subsequent doses throughout the dosing period, sparse PK sampling will also be used to characterize potential trough values and C_{max} values to determine that exposure of RO7011785 is maintained during this dose regimen. This multiple-point PK plasma sampling in patients within the first and last weeks of dosing should enable calculation of C_{max} and AUC for comparison with HVs and analysis of key PK characteristics including elimination half-life, dose/exposure and PK/PD relationships.

In Cohorts 1, 2 and 3 of Part 2, tenofovir (including tenofovir alafenamide, if approved for HBV and applicable), entecavir, adefovir and telbivudine will be monitored by sparse PK sampling throughout the treatment duration to determine the level of steady-state concentration. No DDI is expected between RO and NUCs; these data will be used to determine compliance with NUCs.

The timing of the serial PK collections and the sparse PK sampling is aligned with the collection of plasma PD markers in both HVs and patients. The procedures for the collection, handling and shipping of the PK laboratory samples are specified in a separate laboratory manual.

3.2.4 Rationale for Pharmacodynamic Assessments

TLR7 is expressed on human pDC and B-cells, and its activation induces both humoral and cellular changes (Iwasaki and Medzhitov 2004, Lester and Li 2014). These changes include the production of cytokines and chemokines such as IFN-α, IL-6, TNF-α, IL-10, IL-12p40, IP-10 and changes in the expression of ISGs, e.g., ISG15, OAS-1 and myxovirus resistance 1 gene (MX1) and of the TLR7 gene itself (Fidock et al 2011), as well as changes in markers of immune stimulation such as neopterin. Changes in immune cell absolute numbers and percentages in peripheral blood [e.g. myeloid dendritic cells (mDCs), plasmacytoid dendritic cells (pDCs) and lymphocytes] have also been described following the clinical treatment with another TLR7 agonist, RO6864018 (ANA773), in HCV patients with virological response (Boonstra et al 2012).

To capture the broad immunomodulatory effects of TLR7 agonism, a multipronged approach will be taken. Cytokines, chemokines, peripheral blood RNA transcript expression and peripheral blood immune cell phenotype will be measured. Time-points for the analyses will be taken as outlined in the SoA (Appendix 1). These markers also showed a dose-dependent PD profile with other TLR7 agonists when assessed in patients and HVs. The PD activity in the SAD part of the study will be used to help choose the starting dose in the MAD part of the study. Similarly, the PD activity seen in SAD and MAD will be used together with other factors such as safety considerations to choose the starting dose in CHB patients.

In previous HV clinical studies, IFN-α, neopterin and IP-10 were activated with another TLR7-selective agonist, RO6864018 and no activation of the TLR8 markers, IL-6 and TNF-α was seen. Thus, in the current study, any activation of IL-6 and TNF-α may be indicative of activation of TLR8.

3.2.5 Rationale for Exploratory Assessments

HBV viral markers will be assessed in Part 2 as listed in Section 3.3.3.

Marker panels, e.g., RNA transcription profiles may also be evaluated as potential predictors of TLR7 PD responses.

Viral genotype may be assessed in CHB patients (Part 2). Viral genotype is known as a predictive factor to response to PEG-IFN treatment (Sonneveld et al 2012).

As TLR7 is expressed on B-cells it is assumed that activation of B-cells will occur if exposed to a TLR7 agonist. In order to assess the magnitude of B-cell activation, a panel of activation markers including, but not limited to, CD19, CD86, CD80, CD40, CD27, IgD, may be assessed in selected study centers (Part 1 MAD and Part 2 only).

To evaluate any effect of RO7020531 on T- and NK-cell function, a panel of T- or NK-cell activation/costimulation/coinhibition markers may be evaluated in CHB patients.

Clinical genotyping may also be undertaken as appropriate to explore the impact of genetic polymorphism on drug metabolism, transport, PD response, efficacy, or the safety profile of RO7020531.

3.3 OUTCOME MEASURES

3.3.1 Safety Outcome Measures

The safety outcome measures for HVs (Part 1) and patients (Part 2) are as follows:

- Incidence and severity of AEs.
- Incidence of laboratory abnormalities based on hematology, clinical chemistry (including liver function tests), coagulation and urinalysis test results.
- Incidence of vital signs (blood pressure, pulse rate, respiratory rate and body temperature) or ECG (PR [PQ], QRS, QT, QTcF) abnormalities.

A detailed medical history and physical examination will be performed at the time-points indicated in the SoA. Height will only be recorded at screening.

AEs and concomitant medications will be monitored throughout the entire study (screening through follow-up) as defined by International Conference on Harmonization (ICH) guidelines.

Monitoring for liver flares will be conducted for the duration of Part 2 in CHB patients (see Section 5.2.1).

3.3.2 Pharmacokinetic and Pharmacodynamic Outcome Measures

3.3.2.1 Pharmacokinetic Outcome Measures

The PK evaluations for HVs (Part 1) and patients (Part 2) are as follows:

- Summary descriptive statistics of plasma PK parameters for RO7020531, the main active metabolite RO7011785, and additional metabolites including RO7018822 and RO7033805 will be computed. These parameters include C_{max}, T_{max}, AUC_{inf}, AUC_{last} and $t_{1/2}$ and will be presented by dose cohorts including mean, standard deviation (SD), coefficient of variation (CV), medians and ranges.
- The total amount of RO7020531, the main active metabolite RO7011785 and additional metabolites, including RO7018822 and RO7033805, in urine over a 24 hour period will be computed and provided in tables and listings.
- In Cohorts 1,2 and 3 of Part 2 of the study, sparse sampling for tenofovir (including tenofovir alafenamide, if approved for HBV and applicable), entecavir, adefovir and telbivudine will be made. As appropriate, tables and listings of these concentrations will be provided.

3.3.2.2 Pharmacodynamic Outcome Measures

Part 1: SAD and MAD in Healthy Volunteers

- Blood samples will be collected to evaluate a number of immune PD outcome measures including, but not limited to, the protein and metabolite markers (neopterin, IFN-α, IP-10, TNF-α, IL-6, IL-10, IL-12p40) and markers of transcriptional responses (ISG15, OAS-1, MX1 and TLR7).

Part 2: Chronic Hepatitis B Patients

- Blood samples will be collected to evaluate a number of immune PD outcome measures including, but not limited to, the protein and metabolite markers (neopterin, IFN-α, IP-10, TNF-α, IL-6, IL-10, IL-12p40) and markers of transcriptional responses (ISG15, OAS-1, MX1 and TLR7).
- Additional immune PD assessments (proteins and RNA transcripts) may be added to those listed above as needed.

3.3.3 Exploratory Outcome Measures

Part 2 only: Chronic Hepatitis B Patients

HBV Antiviral Measures

The antiviral outcome measures for this study are the following:

- Quantitative HBV DNA
- HBsAg (qualitative)
- HBsAg (quantitative)
- HBeAg (qualitative)
- Anti-HBe and anti-HBs antibody status
- Additional exploratory virology laboratory tests, such as HBsAg/anti-HBsAg complex levels, HBeAg levels (semi-quantitative assessment based on signals in the HBeAg assay), HBV core and core related antigens, anti-HBc antibody and HBV RNA in serum may be assessed at the time-points specified in the SoA as an evaluation of potentially exploratory markers of therapeutic response, in conjunction with the viral parameters listed above.

For HBV-specific assessments, please see Section 4.6.1.8.

Outcomes of antiviral response will include quantitative HBeAg decline, loss of HBeAg, development of anti-HBe, HBeAg seroconversion (loss of HBeAg and presence of anti-HBe), quantitative HBsAg decline, loss of HBsAg, development of anti-HBs, HBsAg seroconversion (loss of HBsAg and presence of anti-HBs), and maintenance of HBV DNA levels less than 90 IU/mL for Cohorts 1, 2 and 3 (at the end of the treatment period and at the end of the follow-up period). For Cohort 4, the outcome of antiviral response will additionally include quantitative HBV DNA level (actual and change from baseline).

Monitoring of viral resistance will be performed in any patient who experiences virological breakthrough. Please see Section 4.6.1.8.

Viral genotype may be assessed in CHB patients (Part 2). Viral genotype is known as a predictive factor to response to PEG-IFN treatment (Sonneveld et al 2012).

Other Exploratory Measures

Exploratory outcome measures for Part 1 and 2 of this study which may be investigated include, but are not limited to, the following:

- Clinical genotyping samples will be collected in all HVs/patients and genotyping may be conducted as appropriate to explore the impact of genetic polymorphism on drug metabolism, transport, PD response, efficacy, or the safety profile of RO7020531.
- Immunophenotyping might be performed by flow cytometry to assess changes in count and activation status of selected immune cells and its relevance for treatment response in Part 1 MAD and Part 2.
- Total transcriptome analysis may be performed from whole blood RNA samples to identify biomarkers potentially predictive of antiviral and/or pharmacodynamic responses.

For exploratory assessments please see Section 4.6.1.8.

4. MATERIALS AND METHODS

4.1 CENTER

Part 1 SAD and MAD in HVs is a single-center study. An additional site(s) may be included for back-up purposes and may be activated if needed.

Part 2 in CHB patients is a multi-center study. Administrative and Contact Information, and List of Investigators are provided separately.

4.2 STUDY POPULATION

Part 1: SAD and MAD in Healthy Volunteers

Approximately eight HV cohorts may be evaluated in the SAD portion of this study (approximately 80 subjects in total), and approximately three cohorts are anticipated for the MAD (approximately 30 subjects in total). In each cohort, eight subjects will be treated with RO7020531 and two with placebo. A minimum of two females per cohort should be randomized, with at least one female receiving active drug. Depending on the data collected in each cohort, additional cohort(s) may be added in the SAD and MAD portions to collect the necessary information for selecting appropriate doses in patients.

Part 2: Chronic Hepatitis B Patients

Approximately 20-30 virologically suppressed CHB patients may be randomized into at least two cohorts in Part 2 of the study (Cohort 3 will be optional) and up to 15 treatment-naïve CHB patients may be randomized into the optional Cohort 4. Within each dose level, up to 15 patients (8-12 active and 2-3 placebo) will be randomized. A minimum of two females per cohort should be randomized in Part 2, with at least one female per cohort receiving active drug.

4.2.1 Recruitment Procedures

For Part 1, HVs 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.

For Part 2, study sites will search their database of CHB patients for potentially suitable patients. Patients will be identified for potential recruitment using pre-screening enrollment logs. In case the study sites will use newspaper/Social Network Service/poster advertisements, mailing lists or other distributable documents to recruit patients, the Sponsor will ensure that these documents have been reviewed and approved by the IEC/IRB prior to use.

4.2.2 Inclusion Criteria

4.2.2.1 Part 1: SAD and MAD in Healthy Volunteers

HVs must meet the following criteria for study entry:

1. Healthy male and female subjects, 18 to 65 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, and urinalysis.
2. Informed of, and 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.

3. For women of childbearing potential: agreement to remain abstinent (refrain from heterosexual intercourse) or use two approved contraceptive methods, of which one must be a barrier method and the other should be an established non-barrier form of contraception with a failure rate of < 1% per year, during the treatment period and for at least one month after the last dose of study drug.
    a. A woman is considered to be of childbearing potential if she is post menarcheal, has not reached a post menopausal state (≥ 12 continuous months of amenorrhea with no identified cause other than menopause), and has not undergone surgical sterilization (removal of ovaries and/or uterus).
    b. Examples of contraceptive methods with a failure rate of < 1% per year include bilateral tubal occlusion, male sterilization, established proper use of hormonal contraceptives that inhibit ovulation, hormone-releasing IUDs, and copper IUDs.
    c. 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.
4. Male subjects must be willing to use two methods of contraception with their partners, one of which must be a barrier method (i.e. condom), for the duration of the study and for one month after the last dose of study medication. Other acceptable forms of contraception for this study include vasectomy, bilateral tubal occlusion, intrauterine device (IUD) or proper use of hormonal contraceptives. Periodic abstinence is not considered an adequate form of contraception. Men must refrain from donating sperm during this same period.
5. Negative pregnancy test on Day -1 for female subjects.
6. A body mass index (BMI) between 18 to 32 kg/m^2, inclusive.
7. Non-smokers, or use of < 10 cigarettes (or equivalent nicotine-containing product) per day.
8. Negative anti-nuclear antibody (ANA) test; or positive with dilutions not greater than 1:40 and with no associated history or symptoms of potential connective tissue disease or other immune-mediated diseases.

4.2.2.2 Part 2: Chronic Hepatitis B Patients

Patients must meet the following criteria for study entry:

1. Adult male and female patients, 18 to 65 years of age, inclusive.
2. Informed of, and 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.
3. A BMI between 21 to 32 kg/m^2, inclusive. Male patients must be above 55 kg and female patients above 45 kg body weight.
4. CHB infection (positive test for HBsAg for more than 6 months prior to randomization).

5. For Cohort 1, 2, 3 and 4: HBsAg detectable at screening.
6. For Cohort 1, 2 and 3: On treatment with tenofovir, entecavir, adefovir, or telbivudine, either as single agents or in combination, for at least 6 months.

   For Cohort 4: HBV treatment naïve or not on any anti-HBV treatment for the past 6 months.
7. For Cohort 1, 2 and 3: HBV DNA < 90 IU/mL for at least 6 months prior to randomization; HBV DNA < 90 IU/mL at screening by Roche Cobas assay.

   For Cohort 4: HBV DNA at screening $\geq 2 \times 10^4$ IU/mL for HBeAg positive and ≥ 2 x 103 IU/mL for HBeAg negative patients.
8. For Cohort 1, 2 and 3: Alanine amino transferase (ALT) $\leq 1.5 \times$ upper limit of normal (ULN) during the 6 months prior to randomization confirmed by two measurements separated by at least 14 days (one of the ALT measurements can be done at screening); ALT at screening $\leq 1.5 \times$ ULN.

   For Cohort 4: ALT and aspartate aminotransferase (AST) at screening and Day -1 visit: $\leq 5 \times$ ULN.
9. Screening laboratory values (including hematology, chemistry, urinalysis) obtained up to 28 days prior to first study treatment within acceptable range or judged to be not clinically significant by the Principal Investigator (PI) and Medical Monitor.
10. Gamma glutamyl transpeptidase (GGT), alkaline phosphatase (ALP), albumin, total and direct bilirubin within normal range or judged to be not clinically significant by the Investigator and Medical Monitor at screening.
11. Negative ANA test, or positive with dilutions not greater than 1:40 and with no associated history or symptoms of potential connective tissue disease or other immune-mediated diseases.
12. Liver biopsy, Fibroscan® or equivalent elastography test obtained within 6 months prior to randomization demonstrating liver disease consistent with chronic HBV infection with absence of cirrhosis and absence of extensive bridging fibrosis (cirrhosis or extensive bridging fibrosis are defined as ≥ Metavir 3, recommended cutoff for fibroscan 8.5 kPa).
13. For women of childbearing potential: agreement to remain abstinent (refrain from heterosexual intercourse) or use two approved contraceptive methods, of which one must be a barrier method and the other should be an established non-barrier form of contraception with a failure rate of < 1% per year, during the treatment period and for at least one month after the last dose of study drug.
    a. A woman is considered to be of childbearing potential if she is post-menarcheal, has not reached a post-menopausal state (≥ 12 continuous months of amenorrhea with no identified cause other than menopause), and has not undergone surgical sterilization (removal of ovaries and/or uterus).
    b. Examples of contraceptive methods with a failure rate of < 1% per year include bilateral tubal occlusion, male sterilization, established proper use of hormonal contraceptives that inhibit ovulation, hormone-releasing IUDs, and copper IUDs.

c. 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.

14. 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 or pregnant female partners, men must remain abstinent or be willing to use two methods of contraception with their partners, one of which must be a condom and the other should be an established form of contraception, during the treatment period and for at least one month after the last dose of study drug to avoid exposing the embryo. Other acceptable forms of contraception include vasectomy, bilateral tubal occlusion, IUD or proper use of hormonal contraceptives (e.g. contraceptive pills). 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 and withdrawal are not acceptable methods of contraception.

15. Negative pregnancy test on Day -1 for female patients.

4.2.3 Exclusion Criteria

4.2.3.1 Part 1: SAD and MAD in Healthy Volunteers

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

1. Pregnant (positive pregnancy test) or lactating women, and male partners of women who are pregnant or lactating.
2. History of immunologically mediated disease (e.g., inflammatory bowel disease, idiopathic thrombocytopenic purpura, lupus erythematosus, autoimmune hemolytic anemia, scleroderma, severe psoriasis, rheumatoid arthritis, multiple sclerosis, or any other autoimmune disease).
3. History or symptoms of any clinically significant disease including (but not limited to), neurological, cardiovascular, endocrine, respiratory, hepatic, ocular, or renal disorder (as per investigator's judgment).
4. Personal or family history of congenital long QT syndrome or sudden cardiac death.
5. Evidence of an active or suspected cancer or a history of malignancy, where in the Investigator's opinion, there is a risk of recurrence.
6. History of having received or currently receiving any systemic anti-neoplastic (including radiation) or immune-modulatory treatment (including systemic oral or inhaled corticosteroids, IFN or PEG-IFN) within the 8 weeks prior to the first dose of study drug or the expectation that such treatment will be needed at any time during the study. Eye drop-containing and infrequent inhaled corticosteroids are permissible up to 4 weeks prior to the first dose of study drug.

7. History of clinically significant thyroid disease; also, subjects with clinically significant elevated thyroid-stimulating hormone (TSH) concentrations at screening.
8. Any confirmed clinically significant allergic reactions (anaphylaxis) against any drug, or multiple drug allergies (non-active hay fever is acceptable).
9. History of clinically significant psychiatric disease, especially major depression (significant psychiatric disease is defined as treatment with an antidepressant medication or a major tranquilizer at therapeutic doses for major depression or psychosis, respectively, or any history of the following: a suicide attempt, hospitalization for psychiatric disease, or a period of disability due to a psychiatric disease).
10. Clinically significant acute infection, e.g., influenza, local infection or any other clinically significant illness within two weeks of randomization.
11. History of clinically significant GI disease including inflammatory bowel disease, peptic ulcer disease, GI hemorrhage.
12. Confirmed systolic blood pressure (BP) greater than 140 or less than 90 mmHg, and diastolic BP greater than 90 or less than 50 mmHg at screening (based on the average of 3 separate resting BP measurements, properly measured with well-maintained equipment, after at least 5 minutes rest).
13. Clinically relevant ECG abnormalities on screening ECG: e.g.,
    a. QTc interval (QTcF > 450 msec or < 300 msec)
    b. Notable resting bradycardia (HR < 45 bpm), or HR > 90 bpm
    c. Difference between highest and lowest of any screening QTc > 30 msec
    d. ECGs with documented machine errors in the interval duration assessments
    e. 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 QRS onset, low amplitude T-wave, merged T- and U-waves, prominent U-waves)
    f. Evidence of atrial fibrillation, atrial flutter, complete bundle branch block, Wolf-Parkinson-White Syndrome, or cardiac pacemaker.
14. Any of the following laboratory parameters prior to dosing:
    a. White blood cells (WBC) < 3000 cells/mm^3
    b. Neutrophil count < 1500 cells/mm^3
    c. Platelet count < 140,000 cells/mm^3
    d. Activated partial thromboplastin time (aPTT) > 40 seconds, international normalized ratio (INR) > 1.2
    e. Hb < 12 g/dL in females or 13 g/dL in males
15. Abnormal renal function including serum creatinine > ULN or calculated CrCl < 70 mL/min (using the Cockcroft Gault formula).

16. ALT or AST values at screening above ULN and judged clinically significant by the Investigator.
17. Positive results for anti-mitochondrial antibody (AMA), anti-smooth muscle antibody (ASMA) or thyroid peroxidase antibody.
18. Positive Hepatitis A virus antibody (HAV Ab IgM), Hepatitis B surface antigen (HBsAg), Hepatitis C antibody (HCV Ab), or positive for human immunodeficiency virus (HIV) at screening.
19. 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 or judged to be clinically irrelevant for healthy subjects.
20. History of alcohol abuse (consumption of more than 2 standard drinks per day on average; 1 standard drink = 10 grams of alcohol) and/or drug abuse within one year of randomization. Alcohol consumption will be prohibited at least 48 hours before screening, 48 hours before admission until discharge from the clinic, and 48 hours before each scheduled visit.
21. Positive test for drugs of abuse or positive alcohol test at screening or Day – 1.
22. Any clinically significant concomitant disease or condition that could interfere with, or for which the treatment of 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.
23. Use of any medication (prescription or over-the-counter [OTC], including health supplements, vitamins or herbal remedies) within the 2 weeks prior to the first dosing or within 5 half-lives of the medication prior to first dosing (whichever is longer). Exceptions may be made on a case-by-case basis following discussion and agreement between the Investigator and the Sponsor.
24. Participation in an investigational drug or device study within 90 days prior to randomization.
25. Donation or loss of blood over 500 mL, or administration of any blood product, within 90 days prior to starting study medication.
26. Subjects under judicial supervision, guardianship or curatorship.
27. Any medical or social condition which may interfere with the subject's ability to comply with the study visit schedule or the study assessments.

4.2.3.2 Part 2: Chronic Hepatitis B Patients

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

1. Pregnant (positive pregnancy test) or lactating women.
2. History of liver cirrhosis.
3. History or other evidence of bleeding from esophageal varices.

4. Decompensated liver disease (e.g., Child-Pugh Class B or C clinical classification or clinical evidence such as ascites or varices).
5. History or other evidence of a medical condition associated with chronic liver disease other than HBV infection (e.g., hemochromatosis, autoimmune hepatitis, alcoholic liver disease, toxin exposure, thalassemia, nonalcoholic steato-hepatitis, etc.). A clinical diagnosis of fatty liver is allowed provided that non-alcoholic steatohepatitis (NASH) has been excluded by liver biopsy.
6. Documented history or other evidence of metabolic liver disease within one year of randomization.
7. Positive test for Hepatitis A virus (IgM anti-HAV), Hepatitis C virus (HCV), Hepatitis D virus, Hepatitis E virus (HEV), or human immunodeficiency virus (HIV).
8. Expected to need systemic antiviral therapy other than that provided by the study at any time during their participation in the study, with the exception of oral therapy for Herpes simplex virus type I (HSV I) or HSV II.
9. History of or suspicion of hepatocellular carcinoma or alpha fetoprotein $\geq$ 13 ng/mL at screening.
10. History of immunologically mediated disease (e.g., inflammatory bowel disease, idiopathic thrombocytopenic purpura, lupus erythematosus, autoimmune hemolytic anemia, scleroderma, severe psoriasis, rheumatoid arthritis, multiple sclerosis, or any other autoimmune disease).
11. History of clinically significant cardiovascular (including postural hypotension), endocrine, renal, ocular, pulmonary or neurological disease (as per Investigator's judgment).
12. History of clinically significant GI disease including inflammatory bowel disease, peptic ulcer disease, GI hemorrhage, or history of pancreatitis.
13. History of clinically significant psychiatric disease, especially major depression (significant psychiatric disease is defined as treatment with an antidepressant medication or a major tranquilizer at therapeutic doses for major depression or psychosis, respectively, or any history of the following: a suicide attempt, hospitalization for psychiatric disease, or a period of disability due to a psychiatric disease).
14. Active or suspected cancer or a history of malignancy, where in the Investigator's opinion, there is a risk of recurrence.
15. History of having received or currently receiving any systemic anti-neoplastic (including radiation) or immune-modulatory treatment (including systemic oral or inhaled corticosteroids, IFN or PEG-IFN) within the 8 weeks prior to the first dose of study drug or the expectation that such treatment will be needed at any time during the study. Eye drop-containing and infrequent inhaled corticosteroids are permissible up to 4 weeks prior to the first dose of study drug.

    Cohort 4: Concurrent HBV treatments.
16. History of organ transplantation.

17. Clinically significant thyroid disease; also, patients with clinically significant elevated TSH concentrations at screening.
18. Any confirmed clinically significant allergic reactions (anaphylaxis) against any drug, or multiple drug allergies (non-active hay fever is acceptable).
19. Clinically significant acute infection (e.g., influenza, local infection) or any other clinically significant illness within 2 weeks of randomization.
20. Clinically relevant ECG abnormalities on screening ECG.
21. Any of the following laboratory parameters at screening:
    a. WBC < 3,000 cells/mm^3
    b. Neutrophil count < 1500 cells/mm^3
    c. Platelet count < 140,000 cells/mm^3
    d. aPTT > 40 seconds, INR > 1.2
    e. Hb < 12 g/dL in females or 13 g/dL in males
22. Abnormal renal function including serum creatinine > ULN or calculated CrCl < 60 mL/min (using the Cockcroft Gault formula).
23. Positive results for AMA, ASMA or thyroid peroxidase antibody.
24. Participation in an investigational drug or device study within 30 days prior to randomization.
25. Donation or loss of blood over 500 mL, or administration of any blood product, within 90 days prior to starting study medication.
26. History of alcohol abuse (consumption of more than 2 standard drinks per day on average; 1 standard drink = 10 grams of alcohol) and/or drug abuse within one year of randomization.
27. Positive test for drugs of abuse or positive alcohol test at screening or Day -1. For positive cannabinoids test, the eligibility is at the Investigator's discretion.
28. Patients under judicial supervision, guardianship or curatorship.
29. Any medical or social condition which may interfere with the patient's ability to comply with the study visit schedule or the study assessments.

4.3 METHOD OF TREATMENT ASSIGNMENT AND BLINDING

This study is observer-blinded. This means that the subjects (HV and patients), the Investigator(s), and all individuals in direct contact with the HVs/patients at the investigative sites will be blinded. For Part 1 only, the pharmacy staff handling the study drug distribution will be unblinded.

This study is Sponsor open; members of the Sponsor's study team will be unblinded. This may exclude CRO staff in direct contact with the study sites.

4.3.1 Part 1: SAD and MAD in Healthy Volunteers

In each Cohort of Part 1, 10 study subjects will be randomized 4:1 to RO7020531 or to placebo. Each cohort of HVs in Part 1 will randomize a minimum of two females, with at least one female receiving active drug.

The list of randomized treatment assignments will be generated by Roche or its designee. The randomized treatment assignment will be allocated from the list sequentially to subjects in the order in which they are randomized. For each dose cohort (in SAD only), the randomization will be designed such that of the first 2 subjects, 1 will receive active drug, and the other will receive placebo. The remaining 8 subjects in the cohort will be randomized such that 7 receive active drug and 1 receives placebo. The treatment allocation will be managed by the unblinded pharmacist and will be based on the randomization code list.

To allow informed recommendations or decisions regarding the dose decisions in this study, an integrated assessment of the available data on PK, select PD, safety and tolerability will be made prior to each dose decision.

In exceptional cases, (e.g. if deemed important for dose decisions or for the more thorough evaluation of safety-related concerns that may impact dosing of future subjects on this or other currently conducted or shortly to start studies involving administration of RO7020531) and in the interest of the subjects' safety, the investigator may be unblinded after approval by the Clinical Pharmacologist.

The investigator will receive a set of sealed treatment codes. These may have the form of sealed envelopes or scratch codes. If the identity of the test medication needs to be known in order to manage the subject's condition, the treatment code for that subject may be broken.

As per health authority reporting requirements, the sponsor will break the treatment code for all unexpected SAEs (see Section 5.1) that are considered by the investigator to be related to study drug. All treatment code breakages should be documented in the study file. Treatment codes should not be broken except in emergency situations and, if possible, the responsible clinical pharmacologist should be contacted before the code is opened. At the final monitoring visit, the unused code labels 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 code labels will be destroyed by the investigator site after verification by the study monitor, and once permission is granted by 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 at the investigating site, will be documented in the study report with date, reason for identifying the drug and the name of all the person(s) who had to be unblinded.

4.3.2 Part 2: Chronic Hepatitis B Patients

In each Cohort of Part 2, patients will be randomized 4:1 to RO7020531 or to placebo. Each cohort of patients in Part 2 will randomize a minimum of two females, with at least one female receiving active drug. The patient randomization numbers will be generated by the interactive voice/web response system (IxRS) according to specifications provided by the Sponsor to the external randomization vendor. Patients and all study site personnel will be blinded to treatment assignment throughout the Part 2 of the study until all patients have completed their treatment period or discontinued prematurely. Members of the Sponsor's study team will be unblinded. This may exclude CRO staff in direct contact with the study sites.

If unblinding at the site is necessary for patient management (in the case of an SAE), the Investigator will be able to break the treatment code by contacting the IxRS. Treatment codes should not be broken except in emergency situations. If the Investigator wishes to know the identity of the study drug for any other reason, he or she should contact the Medical Monitor directly before the code is broken, if possible. The Investigator should document and provide an explanation for any premature unblinding (e.g., accidental unblinding, unblinding due to a SAE).

As per Health Authority reporting requirements, the Sponsor will break the treatment code for all unexpected SAEs (see Section 5.1) that are considered by the Investigator to be related to study drug.

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 at the investigating site end, will be documented in the study report with date, reason for identifying the drug and the name of all the person(s) who had to be unblinded.

4.4 STUDY TREATMENT

4.4.1 Formulation, Packaging, and Handling

4.4.1.1 RO7020531 and Placebo

RO7020531 and placebo will be supplied by Roche.

Investigational Medicinal Product (IMP): Hard gelatin capsule for oral administration containing 1 mg, 10 mg, or 100 mg of RO7020531 drug substance.

Placebo: Hard gelatin capsule identical in size and appearance to the corresponding active capsules, containing microcrystalline cellulose of compendial grade but no active substance.

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 medication will be in accordance with Roche standard and local regulations.

The study drug must be stored according to the details on the product label: "Store at 2°C–8°C, protect from light and moisture".

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, refer to the RO7020531 IB.

4.4.1.2 Nucleos(t)ide Analogues

Tenofovir, entecavir, adefovir and telbivudine are marketed drugs and will be used in the Cohorts 1, 2 and 3 of Part 2 of the study for an authorized indication per local label. They are not considered as IMPs.

During the study treatment and follow-up periods, patients in Cohorts 1, 2 and 3 will continue the use of the NUCs as prescribed by each patient's physician and following the general warnings and precautions in the local labels.

4.4.2 Dosage, Administration and Compliance

4.4.2.1 RO7020531 and Placebo

Both RO7020531 and placebo will be administered orally in capsules.

Part 1: SAD and MAD in Healthy Volunteers

In the SAD cohorts, RO7020531 or matching placebo will be administered orally to the subjects by investigational staff on the morning of Day 1 after an overnight fast of at least 10 hours. Subjects in SAD cohorts will not eat for 4 hours after the dose is administered. Water will be allowed ad libitum until one hour prior to dosing and after one hour post-dosing. Approximately 4 hours after dosing, subjects will be administered lunch. The SAD portion of the study will include an adaptive number of cohorts (approximately eight). Each dose cohort will include 10 subjects (8 active and 2 placebo). The anticipated dose-escalation sequence for SAD is 3 mg, 10 mg, 20 mg, 40 mg, 60 mg, 100 mg, 140 mg, and 170 mg.

The starting dose of 3 mg RO7020531 will be administered as three 1 mg capsules. All other doses will be administered as a combination of 1 mg, 10 mg or 100 mg capsules with the requisite number of capsules being administered per specific dose cohort. Should an intermediate dose be required due to a change in the anticipated dose-escalation, the dose will be composed of the appropriate combination of 1 mg, 10 mg and 100 mg capsules.

In the MAD cohorts, RO7020531 or matching placebo will be administered orally to the subjects by investigational staff QOD from Day 1 through to Day 13. In total, seven

doses will be given with study drug administration on Day 1, Day 3, Day 5, Day 7, Day 9, Day 11, and Day 13. Each dose will be given in a fasted state (after an overnight fast of at least 10 hours). Water will be allowed ad libitum until one hour prior to dosing and after one hour post dosing. Subjects will not eat for 4 hours after each dose is administered. The MAD portion of the study will include an adaptive number of cohorts (approximately three). Each dose cohort will include 10 subjects (8 active and 2 placebo). Dose levels for MAD portion of the study will be defined during the study conduct based on emerging safety and tolerability data from the SAD portion of the study.

Doses in HVs will be given orally with 240 mL of water. Additional amounts of water up to 100 mL could be given to assist dose administration only if needed.

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 subject 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 subject during the study.

Part 2: Chronic Hepatitis B Patients

In Part 2, RO7020531 or matching placebo will be administered orally to patients every other day from Day 1 through to Day 41 *(unless in Cohort 4 in case of QW dosing as dose modification). In total, up to* 21 doses will be given. The dose level for the Cohort 1 in patients will be determined upon completion of Part 1 (see Section 3.1.2.2). The number of capsules per intake will reflect the dose level for a cohort and will be defined prior to dosing of the first patient in a cohort. For every study drug administration, the dose will be composed of the appropriate combination of 1 mg, 10 mg and 100 mg capsules.

Cohorts 1, 2, and 3 will include 10 patients (8 active and 2 placebo). Every dose will be given in the fasted state (patients will not eat 2 hours before and 2 hours after dosing). *Cohort 4 will include up to 15 patients (8 to 12 active and 2 to 3 placebo).*

In Cohorts 1, 2 and 3, RO7020531 or matching placebo will be administered together with patient's NUC treatment (tenofovir, entecavir, adefovir, or telbivudine either as single agents or in combination). NUC(s) will be administered per local label. On the day of study visits, patients will take study treatment together with NUC(s) in clinic.

In Cohort 4, RO7020531 or matching placebo will be administered alone.

The qualified individual responsible for dispensing the study drug will dispense to the patient the correct bottles of RO7020531 and/or matching placebo according to the IxRS assignment. This individual will write the date dispensed and patient number on the study drug bottle label and on the Drug Accountability Record. This individual will also record the study drug batch or lot number received by each patient during the study.

Guidelines for treatment discontinuation are provided in Section 4.7.

Accountability and patient compliance will be assessed by maintaining adequate study drug dispensing records. Information about patient compliance should be captured in an electronic Case Report Form (eCRF). The data should include information about any missing doses or other dosing errors throughout treatment period.

Patients should bring all bottles of the study drug to the site at every study visit when study drug is taken at the clinic (i.e., Days 3, 7, 21, 41 [and additionally on Days *5,* 15*,* and 35 for Cohort 4 only]). Patients will return all bottles to the site after the last dose of the study drug. Patients will be asked at every visit during treatment period about their intake of the study drug at home.

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.

4.4.2.2 Nucleos(t)ide Analogues

In Cohorts 1, 2 and 3 of Part 2 of the study, enrolled patients will be on tenofovir, entecavir, adefovir, or telbivudine, either as single agents or in combination, for at least 6 months before entering the study. All patients will continue their ongoing treatment with NUCs during the study treatment period and during the study follow-up period as prescribed by each patient's physician.

NUCs will be administered as non-IMP orally once daily per local label (See prescribing information for: tenofovir, entecavir, adefovir, telbivudine). On the day of RO7020531 intake, NUC(s) will be taken together with study treatment in the fasted state (patients will not eat 2 hours before and 2 hours after dosing). On the day of study visits, patients will take their NUC treatment in clinic.

No drug accountability will be performed with NUCs. However, NUC administration during the study, including the follow-up period, should be captured in site documentation and eCRF. This information should include information about any missing doses during treatment and follow-up period. Patients will be asked at every visit about their intake of NUC(s) at home.

4.4.2.3 Dose Modification

RO7020531 Dose Modification for Individual Healthy Volunteers or Patients

Dose modification of RO7020531 will not be performed in individual HVs (Part 1 MAD) or patients (Part 2*) in Cohorts 1, 2 and 3. For patients who are treatment naïve (Part 2 Cohort 4), dose modification of RO7020531 is allowed. Further details are provided in Section* 5.2.1*).*

Nucleos(t)ide Analogues Dose Modification (Part 2 Cohorts 1, 2 and 3)

Dose modification of tenofovir, entecavir, adefovir, or telbivudine is not expected during this study.

4.4.3 Investigational Medicinal Product Accountability

RO7020531 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 HV/patient to whom the study drug was dispensed (for example subjects initials and date of birth).
- The date(s), quantity of the study drug dispensed to the HV/patient.
- The date(s) and quantity of the study drug returned by the HV/patient.

All records and drug supplies must be available for inspection by the Clinical Trial Monitor at every monitoring visit.

IMP 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. Written authorization must be obtained from the sponsor at study start up before destruction.

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 destroyed
- Quantity of investigational product destroyed
- Date of destruction

- Method of destruction
- Name and signature of responsible person [or company] who destroyed investigational product

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

4.4.3.1 Medication Adherence and Reminder System

The information provided in this document is the property of AiCure ("Platform Provider"), and should not be disclosed to others without written authorization from the Platform Provider, except to the extent necessary to ensure adequate conduct of the Study.

This Study might employ a medication adherence monitoring platform ("Platform") for patients in Cohort 4 of Part 2 of the study. The Platform uses artificial intelligence on smartphones to confirm medication ingestion. In addition, built-in reminders and a communication system allow real-time intervention in case of drug interruptions.

Use of this Platform will in no way supersede or replace the physician and/or prescribed medication protocol of the patients. Because the Platform does not change the medication protocol of the patients, but rather encourages adherence to the predefined protocol, use of this Platform presents minimal risk to the patients. Use of the Platform might be required for subjects in Cohort 4 of Part 2 the study.

The monitoring Platform requires that patients take each dose of the medication while using a smartphone. The Platform will be provided to patients preloaded on a smartphone, or patients will download the Platform onto their own mobile device during the first visit.

When at home, Study patients will receive a medication reminder at a time within a predefined window. This notification reminds patients to take their medication dose while using the Platform. Patients will follow a series of prescribed steps in front of the front-facing webcam to visually confirm their ingestion of the medication. The application on the smartphone will make an automated determination of whether the patient has properly taken their medication at the prescribed time. There is no need for a healthcare provider to review the administration, nor would a healthcare provider need to be available at the time the patient takes their medication. The amount of guidance that the device provides to the patient is automatically reduced as the patient becomes more proficient at using the application. If at any time the patient is unable to use the full visual confirmation, the patient will be able to self-report taking the study drug using the AiCure system.

After the device confirms proper medication ingestion, any video recordings will be encrypted and transmitted to a secure centralized location for further analysis, including testing for duplicate enrollment. The captured data and video are reviewable through a roles and rules restricted system ensuring privacy of the information in accordance with

U.S. and applicable European data privacy laws, including General Data Protection Regulation (GDPR) (EU) 2016/679.

Phone numbers of the patients may also be collected and stored in an encrypted manner. Storing the phone numbers will allow for direct communication with patients, including automated messaging from the Platform device and contact by healthcare providers or other monitoring personnel. At no time is the phone number visible to healthcare providers or monitoring personnel. Individuals outside the clinical sites will not be provided with patient names, nor will they be given access to patient medical records.

The Platform may provide significant benefits to Study patients as well as to the other stakeholders in the trial. Patients will benefit from rapid and tailored intervention in case of non-adherence (drug interruptions) without having to visit the clinic for unscheduled visits. Healthcare providers will have access to real-time and continuous adherence data without having to rely on self-reported data or frequent study visits by patients. Patients who regularly fail to take their medication will be contacted by healthcare providers or other Study monitoring personnel for retraining.

4.4.3.1.1 Patient Risk

The Platform provides no more than minimal risk to patients. This protocol only introduces a smartphone-based monitoring application that prompts the user to take their medication, verifies ingestion, and stores encrypted data securely for analysis. Use of the Platform does not affect titration, dosage, route of administration or treatment duration.

All Study data, including any identifiable patient information, will be obtained and encrypted by the application. Patients will be coded according to the protocol and their identity will not be stored with the Study data obtained. After the patient has taken the medication and confirmation of proper ingestion has been completed, the encrypted data will be automatically forwarded to a secure server. The server is compliant with the Health Insurance Portability and Accountability Act (HIPAA) and European data privacy laws, including General Data Protection Regulation (GDPR) (EU) 2016/679, which protects the privacy and security of healthcare and other personal information. The data will be securely stored and only accessible to healthcare providers and other authorized personnel on a need-to-know basis through two-way authentication.

The data may also be retained in a secure manner beyond the term of the trial and utilized to improve the operation of the Platform, categorize adherence activity by disease state or other useful categories, and/or for regulatory filings by the Platform Provider to support future applications for the Platform Provider's product. Individuals who are not associated with the care and treatment of patients will not have access to patient identity or any medical records.

4.4.3.1.2 Patient Confidentiality

The Platform Provider will protect patients' personal information to the full extent required by law. However, information from this Study, including any de-identified video recording(s) of patient performance of various actions, may be submitted to the Study site, and potentially to the U.S. Food and Drug Administration (FDA). Both information obtained by the application, and information in the patient Informed Consent, may be examined by the Study site or the Study site's representatives, and may also be reviewed by the FDA and other regulatory agencies, Institutional Review Board(s) and or Ethics Committee(s). All of these parties are bound to safeguard the rights, safety and well-being of all clinical trial patients, and to maintain all information in confidence, with special consideration given to trials that may include vulnerable patients.

The results of this research project may be presented at meetings or in publications; however, specific patients will not be identified by name in these presentations and/or publications. Information from this Study may also be retained in an encrypted manner by the Platform Provider for the purpose of improving the Platform, to allow for future analysis of various facial and other parameters, the reporting of high level statistical analysis of the Platform, to improve the internal workings of the system running on the smartphone device, or for regulatory filings by the Platform Provider to support future applications for the Provider's product.

4.5 CONCOMITANT THERAPY

4.5.1 Permitted Therapy

4.5.1.1 Part 1: SAD and MAD in Healthy Volunteers

As a general rule, no concomitant medication (including health supplements, vitamins or herbal remedies) will be permitted, 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 AEs should be recorded on the AE eCRF.
- Hormone replacement therapy (HRT), continue using if initiated two months prior to study start.
- Acetaminophen/paracetamol is allowed up to a maximum dose of 2 g per day. 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 AE by the Investigator.

4.5.1.2 Part 2: Chronic Hepatitis B Patients

Concomitant therapy includes any medication, e.g., prescription drugs, OTC drugs, approved dietary and herbal supplements, nutritional supplements used by a patient from 4 weeks prior to screening until the follow-up visit. All concomitant medications should be reported to the Investigator and recorded on the Concomitant Medications eCRF. Information about all HBV treatments received before randomization and during the study till the end of follow-up period should be recorded on the appropriate eCRF.

Acetaminophen (paracetamol) may be used for prophylactic and symptomatic management of flu-like symptoms at the Investigator's discretion. The total daily dose of acetaminophen (paracetamol) should not exceed 2 g per day.

4.5.2 Prohibited Therapy

4.5.2.1 Part 1: SAD and MAD in Healthy Volunteers

All medications (prescription drugs, OTC drugs, approved dietary and herbal supplements, nutritional supplements) taken from 4 weeks prior to study screening until the follow up call will be recorded on the appropriate eCRF.

Prohibited medications include:

- Any prescribed or OTC medications (except for the cases given in Section 4.5.1), including health supplements, vitamins or herbal remedies within 2 weeks prior to the first dosing or within 5 half-lives of the medication prior to first dosing (whichever is longer), until the follow up visit (Day 8 for SAD and Day 20 for MAD).
- Any systemic anti-neoplastic (including radiation) or immune-modulatory treatment (including systemic oral or inhaled corticosteroids, IFN or PEG-IFN) at any time within the 8 weeks prior to the first dose of study drug and until the follow up visit (Day 8 for SAD and Day 20 for MAD). Eye drop-containing and infrequent inhaled corticosteroids are permissible up to 4 weeks prior to the first dose of study drug.

4.5.2.2 Part 2: Chronic Hepatitis B Patients

Prohibited medications include:

- Any systemic anti-neoplastic (including radiation) or immune-modulatory treatment (including systemic oral or inhaled corticosteroids IFN or PEG-IFN) at any time within the 8 weeks prior to the first dose of study drug and until the end of follow-up period. Eye drop-containing and infrequent inhaled corticosteroids are permissible up to 4 weeks prior to the first dose of study drug.
- Any systemic antiviral therapy other than nucleos(t)ide analogues (Cohorts 1, 2 and 3 only) at any time from 30 days before screening until the end of follow-up period, with the exception of oral therapy for HSV I or HSV II. Investigational drugs or herbal and other remedies which are being taken by the patient for possible or perceived effects against HBV are prohibited.

4.5.3 Special Concomitant Medications in Chronic Hepatitis B Patients (Part 2)

The following medications are not prohibited in Part 2 of the study. However, caution is required in case of co-administration with the study drug, and the Sponsor should be informed before these drugs are administered.

- Drugs that reduce renal function or compete for active tubular secretion may increase serum concentrations of either study drug or the co-administered drug.
- Drugs metabolized by aldehyde oxidase including famciclovir and/or inhibiting this enzyme including tamoxifen, raloxifen, cimetidine, promethazine, clozapine and chlorpromazine, which could decrease the formation of RO7011785.

4.5.4 Dietary and Special Requirements

There are data suggesting that concentrated green tea (including bottled green tea beverages) may inhibit aldehyde oxidase (Tayama et al 2011). Therefore, HVs and patients should minimize the amount of bottled green tea beverages and other green tea preparations they drink from Day -1 until the follow-up visit (for SAD and MAD cohorts) or until the end of treatment period (for Part 2).

4.5.4.1 Part 1: SAD and MAD in Healthy Volunteers

In SAD and MAD cohorts, subjects must fast overnight (at least 10 hours) before the dosing and must not eat for 4 hours after the dose is administered. Approximately 4 hours after dosing, subjects will be administered lunch. Meals will be similar in composition and time of administration across all SAD/MAD cohorts.

Water will be allowed ad libitum until one hour prior to dosing and after one hour post-dosing. However, the excessive consumption of fluids (greater than 3 liters per day) should be avoided until completion of the follow-up visit.

Laboratory safety assessments should be conducted after subjects have been fasted for a minimum of 8 hours.

Alcohol must not be consumed from 48 hours before screening, 48 hours before admission until discharge from the clinic, and 48 hours before each scheduled visit. Please refer to Appendix 7 for the calculation of alcohol standard drinks as relates to eligibility criteria.

Caffeine (i.e., beverage, chocolate or supplements) must not be consumed from 48 hours prior to study drug administration until discharge from the clinic.

Strenuous exercise is not permitted during the study from 96 hours before admission until completion of the follow-up visit.

The use of tobacco is not permitted from Day -2 till Day 3 for SAD and from Day -2 till Day 14 for MAD portion of the study.

4.5.4.2 Part 2: Chronic Hepatitis B Patients

No specific food is prohibited in this study.

Dosing with RO7020531 or placebo will be given in fasted state (at least 2 hours after a meal or 2 hours before the next meal), in Cohorts 1, 2 and 3 together with the patient's NUC treatment.

Alcohol consumption is to be strongly discouraged. During the study, patients should consume no more than an average of 20 g of alcohol daily. Please refer to Appendix 7 for the calculation of alcohol standard drinks as relates to eligibility criteria. Patients will be queried on a regular basis about their alcohol consumption and appropriate comments concerning this intake will be recorded on the eCRF.

4.6 STUDY ASSESSMENTS

4.6.1 Description of Study Assessments

All examinations listed below will be performed according to the Schedule of Assessments (SoA) outlined in Appendix 1.

At time-points when several pre-dose assessments coincide, the following sequence should be followed:

- Urine collection (urinalysis, urine PK pre-dose sample).
- ECG recordings.
- Vital signs.
- Pre-dose blood sampling (pre-dose PK, pre-dose PD, HBV assessments, exploratory samples, safety blood tests).
- RO7020531/placebo and NUC intake.

For the post-dose assessments, the following sequence should be followed with the PK blood sample (or PD blood sample if there is no PK sampling) to be taken at the nominal time-point:

- ECG recordings.
- Vital signs.
- Post-dose blood sampling (post-dose PK, post-dose PD).

4.6.1.1 Medical History and Demographic Data

Medical history includes clinically significant diseases, surgeries, reproductive status, smoking history, use of alcohol and drugs of abuse.

In addition, for Part 2, medical history includes detailed HBV history: date of HBV diagnosis, mode of HBV transmission, HBV genotype (if documented), all previous HBV treatments, occurrence of NUC analogue resistance (if any), previous evaluations for cirrhosis, dates/outcomes of liver biopsies (if any).

Previous treatments include all medications (e.g., prescription drugs, over-the-counter drugs, herbal or homeopathic remedies, vitamins, nutritional supplements) used by the HV/patient within 4 weeks prior to the screening visit.

Demographic data will include age, sex, and self-reported race, ethnicity and origin (origin will be collected for Asian subjects only).

4.6.1.2 Physical Examinations

A complete physical examination should be performed at time-points indicated in the SoA (Appendix 1) and includes an evaluation of the head, eyes, ears, nose, throat, neck and lymph nodes, and the cardiovascular, dermatological, musculo-skeletal, respiratory, GI and neurological systems. A genitourinary examination may be performed in case of evocative symptoms at the Investigator's discretion. Any abnormality identified at screening or at baseline should be recorded on the Medical History eCRF.

For patients in Part 2, during the treatment period, a basic and/or symptoms-directed physical examination (at least lungs, cardiovascular, abdomen, extremities, lymph nodes) should be performed according to the SoA (Appendix 1). No recording of weight is required during these basic/symptoms-directed physical exams.

Targeted or symptoms/AE-directed physical examinations should be performed if needed at all other visits at the discretion of the Investigator.

Clinically significant changes from a baseline abnormality showing its improvement or resolution should be recorded on the Additional Observation eCRF. New or worsened clinically significant abnormalities should be recorded as AE on the AE eCRF (Section 5.3.5).

Height will only be recorded at screening. Weight will be recorded at time-points for complete Physical Examination as specified in the SoA (Appendix 1). BMI will be calculated at screening in accordance with the formula provided in Appendix 2.

4.6.1.3 Vital Signs

Blood pressure (BP), pulse rate, respiratory rate and body temperature will be recorded at the time-points specified in the SoA (Appendix 1).

Blood pressure, respiratory rate and pulse rate should be obtained in a quiet room at a comfortable temperature, with the HV/patient's arm unconstrained by clothing or other material. Blood pressure, respiratory rate and pulse rate will be obtained after the HV/patient has been in a supine or sitting position for at least 5 minutes. All blood pressure and pulse rate measurements will be obtained from the same arm (where possible), in the same position and, with the same cuff size, using an automatic instrument with a digital readout, throughout the study. Body temperature measurement can be either oral or tympanic; the method should be maintained throughout the study.

Blood pressure measurement will be performed in triplicate (can be as short as 20 second to 1 minute interval between measurements). The mean of three consecutive replicates will be used as the value for the defined time-point.

4.6.1.4 Electrocardiograms

Triplicate 12-lead ECG recordings (i.e., three useful ECGs without artifacts) will be obtained within approximately 2-5 minutes at each specified time-point. The average of the three readings will be used to determine ECG intervals (PR [PQ], QRS, QT, QTcF). The intervals 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.

Whenever possible, the same brand/model of a standard digital high-quality, high-fidelity electrocardiograph machine equipped with computer-based interval measurements should be used for each HV/patient. The conditions should be as close as possible to pre-dose time-points; this includes but is not limited to food intake, activity level, stressors and room temperature.

To minimize variability, it is important that HV/patients be in a resting position for $\geq$ 10 minutes prior to each ECG evaluation. Body position should be consistently maintained for each ECG evaluation to prevent changes in heart rate. Environmental distractions (e.g., television, radio, conversation) should be avoided during the pre-ECG resting period and during ECG recording. ECGs should be performed prior to any scheduled vital sign measurements and blood draws. In some cases, it may be appropriate to repeat abnormal ECGs to rule out improper lead placement as contributing to the ECG abnormality.

For safety monitoring purposes, the investigator must review, sign and date all ECG tracings. Paper copies will be kept at the study centers with the HV's/patient's clinical file as part of the permanent record. 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.

If any ECG recording documents QT/QTc values > 500 msec or increases from Day -1 QTc > 60 msec (as provided by the machine), the site should repeat the ECG triplicate 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.

4.6.1.5 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 SoA (Appendix 1). Results of clinical laboratory testing will be recorded on the eCRF or will be received as electronically produced laboratory reports submitted directly from the local or central laboratory.

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 HV's/patients' safety. In the event of unexplained abnormal clinically significant laboratory test values, the tests should be repeated immediately and followed up until the values have returned to the normal range and/or an adequate explanation of the abnormality is found.

In Part 2 Cohorts 1, 2 and 3, only a central laboratory will be used; however, unscheduled local laboratory tests may be ordered per Investigator discretion. In this case, a duplicate sample should simultaneously be sent to the central laboratory for analysis.

In Part 2 Cohort 4 at the Day -1 (baseline) visit, a duplicate sample will be taken for liver function tests (ALT) and sent to central laboratory and local laboratory. The local laboratory sample is required to confirm eligibility for liver function parameters before dosing a patient on Day 1. The local laboratory result will not be used for statistical analysis. In the rare event that a patient is deemed to be eligible for the study based on the results of the local test, but the central laboratory test results (conducted after the patient has been dosed) differ, the patient should continue on the study.

At screening, where the clinical significance of abnormal laboratory results is considered uncertain, lab tests may be repeated before randomization to confirm eligibility. If a HV/patient 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 there is an alternative explanation for a positive urine test for drugs of abuse, e.g., previous occasional intake of a medication or food containing for example codeine, benzodiazepines or opiates, the test should be repeated to confirm washout. Please see Section 4.6.2.1 for details on re-screening in Part 2.

For more details on unscheduled laboratory tests see Section 4.6.2.4.

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

For safety outcome measures see Section 3.3.1.

Based on the ongoing data review, any sample type not considered to be critical for safety may be stopped at any time if the data from the samples collected does not produce useful information.

Screening and Safety Samples

Samples for the following blood and urine laboratory tests will be sent to one or several local laboratories (Part 1 only) or central laboratories or to the Sponsor for analysis. Urine dipstick tests will be done locally by the sites for Part 1 and Part 2. Instruction manuals and supply kits will be provided for all laboratory assessments.

For Part 1 and Part 2

- Hematology: Hemoglobin (Hb), hematocrit, total WBC count, differential WBC count (including basophils, eosinophils, lymphocytes, monocytes and neutrophils), platelet count, red blood cells count (RBC), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC) and reticulocyte (%) counts.
- Coagulation: PT, INR, and aPTT.
- Blood Chemistry: ALT, AST, total and indirect bilirubin, ALP, GGT (only at screening), total protein, albumin, urea, creatinine, uric acid, total cholesterol, triglycerides, glucose, sodium, chloride, potassium, calcium, phosphorus, bicarbonate, cystatin C, HbA1c (only at screening).
- 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.

- Pregnancy Test: Serum or plasma beta-human chorionic gonadotropin (β-HCG) at screening, urine on all other occasions (females only).
- Substance use: Drugs of abuse urine test and alcohol test (only at screening and Day -1). A breath test may be used to test for alcohol.
- Viral Serology:
    - Part 1 (only at screening): Hepatitis A (IgM anti-HAV), Hepatitis B (HBsAg), HCV, HIV.
    - Part 2: Viral screening tests (HAV, HCV, Hepatitis D, HEV, HIV). For HBV assessments and schedule see Section 4.6.1.8 (HBV-Specific Viral Assessments) and SoA (Appendix 1).
- Hormones (only at screening): Follicle-stimulating hormone (females only to confirm post-menopausal status).
- Thyroid Function Tests (only at screening): TSH, free T3 and free T4.
- Autoimmune Panel (only at screening): ANA, AMA, ASMA and thyroid peroxidase antibody.

For Part 2 only: Chronic Hepatitis B Patients

- Alpha fetoprotein (only at screening).

4.6.1.6 Pharmacokinetic Assessments

Part 1: SAD in Healthy Volunteers

Blood and urine samples for determination of plasma concentrations of RO7020531, the main active metabolite RO7011785, and additional metabolites including RO7018822 and RO7033805, will be collected at time-points specified in the SoA (Appendix 1).

The volume and pH of urine voided at each interval will be recorded, and aliquots will be collected from the pooled samples at each time interval for analysis.

Part 1 MAD in Healthy Volunteers

In the MAD portion of Part 1, blood samples for determination of plasma concentrations of RO7020531, the main active metabolite RO7011785, and additional metabolites, including RO7018822 and RO7033805, will be collected at the time-points specified in the SoA (Appendix 1).

Plasma and urine concentrations will be measured by a specific and validated method. The PK parameters will be read directly from the plasma concentration versus time profiles, or calculated using standard non-compartmental methods. Total drug concentrations of RO7020531, the main active metabolite RO7011785, and additional metabolites, including RO7018822 and RO7033805, will be calculated over a 24 hour interval.

Details on sampling procedures, sample storage, and shipment are given in the Laboratory Manual.

Part 2: Chronic Hepatitis B Patients

Plasma PK samples for RO7020531, the main active metabolite RO7011785, and additional metabolites, including RO7018822 and RO7033805, as well as the NUC analogues (Cohorts 1, 2 and 3) will be collected as detailed in the SoA (Appendix 1).

4.6.1.7 Pharmacodynamic and Biomarker Assessments

Samples for the following PD/biomarker tests will be sent to several specialized central laboratories for analysis:

- Blood samples collected into serum separator tubes for the analysis of protein biomarkers in peripheral blood.
- Blood samples collected into Paxgene blood RNA tubes for RNA analysis.

Instruction manuals and supply kits will be provided for the collection of all PD samples.

Time-points of PD blood sample collection are provided in the SoA (Appendix 1). Blood samples will be collected to evaluate a number of PD parameters including but not limited to the protein markers of humoral response: neopterin, IFN-α, IP-10, TNF-α, IL-6, IL-10, IL-12p40, and cellular response: transcriptional analysis (whole blood sample for RNA).

Additional PD assessments (proteins and mRNA transcripts) may be added to the list above as needed. Please see PD outcome measures in Section 3.3.2.2.

These samples will be stored for up to 5 years for the protocol assessments defined above, unless otherwise indicated. Samples will be destroyed no later than 5 years after the date of final closure of the clinical database, unless regulatory authorities require specimens to be maintained for a longer time period.

For HVs/patients who consent to Research Biosample Repository (RBR), leftover PD samples will be transferred to RBR. See Section 4.6.1.9.

4.6.1.8 Exploratory Assessments

HBV-Specific Viral Assessments (Part 2)

Blood sample collection times are provided in the SoA (Appendix 1). Please see exploratory outcome measures in Section 3.3.3. Samples for the following laboratory tests will be sent to one or several central laboratories or to the Sponsor for analysis. Instruction manuals and supply kits will be provided for all central laboratory assessments. Validated methods will be used to assess the different markers.

- Quantitative HBV DNA.
- Qualitative HBsAg.
- Quantitative HBsAg level.
- Qualitative HBeAg.
- Semi-quantitative determination of HBeAg level may be performed.
- Anti-HBe and anti-HBs.
- HBsAg/anti-HBsAg complex levels may be assessed to explore the relationship with changes in other viral parameters.
- HBV core and core-related antigens, anti-HBc antibody and HBV RNA in serum may be assessed.
- Viral genotype may be assessed (e.g. by using an immunoassay).
- Viral resistance monitoring: Monitoring of resistance will be performed in any patient who experiences virological breakthrough.

Viral Resistance Monitoring

Blood samples for viral resistance monitoring will be taken at baseline and throughout the study (Part 2), as detailed in the SoA (Appendix 1), to perform sequence analyses in patients who experience virological breakthrough. If, during the treatment period or follow-up period of the study, a patient experiences an increase in serum HBV DNA level of more than one log10 (10-fold) over nadir (virologic breakthrough), an unscheduled visit should be performed within one week of receiving HBV DNA results. At the unscheduled visit, a confirmatory HBV DNA sample and a sample for viral resistance monitoring will be taken (see Section 4.6.2.4) and sent to one or several central laboratories or to the Sponsor for analysis. Additional unscheduled visits and assessments will be carried out as determined necessary by the treating physician. The Medical Monitor for the study should be informed as needed. For patients with confirmed virological breakthrough, administration of study drug will be discontinued (see Section 4.7.1.1) and close monitoring including monitoring for the persistence of resistance mutation(s) will be carried out during the follow-up period. The frequency of the monitoring will be at the discretion of the Investigator and treating physician. For NUC discontinuation, see Section 4.7.1.2.

Sequencing of the HBV genome isolated from the blood of patients with confirmed virological breakthrough will be performed in order to identify potential HBV drug resistance and mutations. Viral phenotypic drug susceptibility testing may also be performed.

Other Exploratory Assessments (Part 1 and Part 2)

Samples for the following exploratory tests will be sent to several specialized central laboratories for analysis.

- Clinical genotyping samples will be collected in all HVs/patients and genotyping may be conducted as appropriate to explore the impact of genetic polymorphism on drug metabolism, transport, PD response, efficacy, or the safety profile of RO7020531.
- Immunophenotyping might be performed by flow cytometry to assess changes in count and activation status of selected immune cells from Part 1 MAD and Part 2 and its relevance for its treatment response.
  Blood samples collected into sodium Cytochex blood collection tubes (for Part 2) for immunophenotyping by flow cytometry. Blood samples at selected sites (for Part 1 MAD and Part 2) will be collected for analysis of B cell activation makers
- Total transcriptome analysis may be performed from whole blood RNA samples to identify biomarkers potentially predictive of antiviral and/or pharmacodynamic responses.

Instruction manuals and supply kits will be provided for the collection of all exploratory samples. Blood sample collection times are provided in the SoA (Appendix 1). Please see exploratory outcome measures in Section 3.3.3.

These samples will be destroyed no later than 5 years after the date of final clinical study report.

For HVs/patients who consent to RBR, leftover samples will be transferred to RBR (see Section 4.6.1.9).

Data arising from biosamples including samples for analyses of inherited DNA will be subject to the confidentiality standards described in Section 8.4.

4.6.1.9 Samples for Research Biosample Repository (optional)

Overview of the Research Biosample Repository

The Roche Biosample Repository (RBR) is a centrally administered group of facilities for the long-term storage of human biologic specimens, including body fluids, solid tissues, and derivatives thereof (e.g., DNA, RNA, proteins, peptides). The collection and analysis of RBR specimens will facilitate the rational design of new pharmaceutical agents and the development of diagnostic tests, which may allow for individualized drug therapy for patients in the future.

Specimens will be collected from HVs/patients who give specific consent to participate in this optional Research Biosample Repository. Collected specimens will be used to achieve the following objectives:

- To study the association of biomarkers with efficacy, AE, or disease progression.
- To increase knowledge and understanding of disease biology.
- To study drug response, including drug effects and the processes of drug absorption and disposition.
- To develop biomarker or diagnostic assays and establish the performance characteristics of these assays.

Approval by the Institutional Review Board or Ethics Committee

Sampling for the RBR is contingent upon the review and approval of the exploratory research and the RBR portion of the Informed Consent Form by each site's IRB or IEC and, if applicable, an appropriate regulatory body. If a site has not been granted approval for RBR sampling, this section of the protocol will not be applicable at that site.

Sample Collection

The following samples will be collected for identification of dynamic (non-inherited) biomarkers:

- Leftover plasma samples
- Leftover serum samples
- Leftover blood samples

The following samples will be collected for identification of genetic (inherited) biomarkers:

- Whole blood for DNA extraction to assess for biomarkers, including but not limited to single nucleotide polymorphisms (SNPs).

The sample collected for DNA extraction may be used for whole genome sequencing (WGS) and other genetic analysis and may be sent to one or more laboratories for analysis.

Genomics is increasingly informing researchers understanding of disease pathobiology. WGS provides a comprehensive characterization of the genome and, along with clinical data collected in this study, may increase the opportunity for developing new therapeutic approaches. Data will be analyzed in the context of this study but will also be explored in aggregate with data from other studies. The availability of a larger dataset will assist in identification of important pathways, guiding the development of new targeted agents.

For all samples, date of consent and specimen collection should be recorded on the associated Research Biosample Repository page of the eCRF. For sampling procedures, storage conditions, and shipment instructions, see the separate laboratory manual.

RBR specimens will be stored and used until no longer needed or until they are exhausted. The Research Biosample Repository storage period will be in accordance with the IEC/IRB-approved Informed Consent Form and applicable laws (e.g., Health Authority requirements).

The repository specimens will be subject to the confidentiality standards (as described under Confidentiality and in Section 8.4).

Confidentiality

Data generated from RBR specimens must be available for inspection upon request by representatives of national and local Health Authorities, and Study monitors, representatives, and collaborators, as appropriate.

HV/patient medical information associated with Research Biosample Repository specimens 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 HV/patient, unless permitted or required by law.

Data derived from Research Biosample Repository specimen analysis on individual HVs/patients will generally not be provided to study investigators unless a request for research use is granted. The aggregate results of any conducted research conducted using RBR specimens will be available in accordance with the effective Roche policy on study data publication.

Genetic research data and associated clinical data may be shared with researchers who are not participating in the study or submitted to government or other health research databases for broad sharing with other researchers. HVs/patients will not be identified by name or any other personally identifying information. Given the complexity and exploratory nature of these analyses, genetic data and analyses will not be shared with investigators or HVs/patients unless required by law.

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

Consent to Participate in the Research Biosample Repository

The Informed Consent Form will contain a separate section that addresses participation in the RBR. The Investigator or authorized designee will explain to each HV/patient the objectives, methods, and potential hazards of participation in the RBR. HVs/patients will be told that they are free to refuse to participate and may withdraw their specimens at any time and for any reason during the storage period. A separate, specific signature will be required to document a HV's/patient's agreement to provide optional RBR specimens. HVs/patients who decline to participate will not provide a separate signature.

The Investigator should document whether or not the HV/patient has given consent to participate by completing the RBR Sample Informed Consent eCRF.

In the event of death or loss of competence of a HV/patient who is participating in the Research, the participant's specimens and data will continue to be used as part of the RBR.

Withdrawal from the Research Biosample Repository

HVs/patients who give consent to provide specimens for the RBR have the right to withdraw their specimens at any time for any reason. If a HV/patient wishes to withdraw consent to the testing of his or her specimens, the Investigator must inform the Monitor in writing of the HV's/patient's wishes using the RBR Withdrawal Form and, if the trial is ongoing, must enter the date of withdrawal on the Research Biosample Repository Withdrawal of Informed Consent eCRF. The HV/patient will be provided with instructions on how to withdraw consent after the trial is closed. A HV's/patient's withdrawal from Study NP39305 does not, by itself, constitute withdrawal of specimens from the RBR. Likewise, a HV's/patient's withdrawal from the RBR does not constitute withdrawal from Study NP39305. Data already generated before time of withdrawal of consent to Research Biosample Repository will still be used.

Monitoring and Oversight

Specimens collected for the Research Biosample Repository will be tracked in a manner consistent with Good Clinical Practice by a quality-controlled, auditable, and appropriately validated laboratory information management system, to ensure compliance with data confidentiality as well as adherence to authorized use of specimens as specified in this protocol and in the Informed Consent Form. Study monitors and auditors will have direct access to appropriate parts of records relating to HV/patient participation in Research Biosample Repository for the purposes of verifying the data provided to Roche. The site will permit monitoring, audits, IEC/IRB review, and Health Authority inspections by providing direct access to source data and documents related to the samples.

4.6.2 Timing of Study Assessments

4.6.2.1 Screening and Pre-treatment 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 screened HVs/patients including HVs/patients who are not subsequently randomized will be maintained at the study site.

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

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

Part 1: SAD and MAD in Healthy Volunteers

A screening examination should be performed from Day -28 to Day -3 for SAD and MAD cohorts. Subjects must fulfill all entry criteria to be accepted into the study. Assessments as detailed in the SoA (Appendix 1) will be conducted.

Part 2: Chronic Hepatitis B Patients

A screening examination should be performed from Day -28 to Day -2 for Part 2. Patients must fulfill all entry criteria to be accepted into the study. Assessments as detailed in the SoA (Appendix 1) will be conducted. All screening assessments must be completed and reviewed to confirm that patients meet all eligibility criteria by Day -1. Only eligible patients should start assessments on Day -1 per SoA (Appendix 1).

Documented liver biopsy or Fibroscan® (or equivalent elastography) test results demonstrating chronic HBV infection with absence of cirrhosis and extensive bridging fibrosis (absence of cirrhosis and extensive bridging fibrosis is defined as Metavir < 3, recommended cutoff for fibroscan 8.5 kPa) must be available within the 6 months prior to randomization. If no results are available, a liver biopsy or Fibroscan® (or equivalent) will be performed during screening.

If a patient in Part 2 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 patient fails a second time, he/she will be classified as a screen failure and cannot be re-screened.

Patients who failed screening may be re-screened only once.

Re-screening is allowed for patients in Part 2 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 of an administrative reason. *The* Sponsor's agreement should be received for re-screening in this case. In order to re-screen such a patient, all inclusion and exclusion criteria should be re-evaluated and all applicable screening assessments repeated if done more than 28 days before randomization. There is no need to repeat alpha-fetoprotein test if done for this study in central laboratory within 6 months before re-screening.

4.6.2.2 Assessments during Treatment

Part 1: SAD and MAD in Healthy Volunteers

All assessments must be performed as per SoA (Appendix 1). There is no window for study visits during MAD treatment (from Day -2 till Day 14).

Part 2: Patients with Hepatitis B Virus

All assessments must be performed as per SoA (Appendix 1). Assessments scheduled on the day of study treatment administration should be performed prior to administration of study treatment, unless otherwise noted in the detailed SoA (Appendix 1).

Safety and PK in the study will be monitored on an ongoing basis with individual data review for every patient.

There is no window for study visits during the 1st week of treatment period (Day -1, 1, 2, 3, 4, 7, 8). Visits Day 21/22 can be carried out on Day 19/20 or Day 23/24. The end of treatment visit can be conducted on Day 39/40 or 41/42. In Cohort 4, there is no window for the Day 5 visit and visits introduced for viral load assessment (Days 15, 28, 35) have a window of ± 2 days.

If a patient misses a visit, the Investigator should contact the Sponsor immediately to discuss the visits and assessments to be performed for the patient to ensure that safety is monitored and that appropriate laboratory samples (including safety, PK and PD) are taken based on each patient's schedule.

4.6.2.3 Follow-Up Assessments

Part 1: SAD and MAD in Healthy Volunteers

All study subjects in SAD and MAD cohorts who complete the study or discontinue from the study early will be asked to return to the clinic approximately 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.

After the follow-up visit, AEs should be followed as outlined in Sections 5.5 and 5.6.

All subjects will have a follow-up phone call approximately 28 days (±3 days) after their last dose administration. This call is for safety purposes to monitor any AE which may have occurred since the follow-up visit. The follow-up call will be reported as the study completion visit in eCRF.

Part 2: Chronic Hepatitis B Patients

A follow-up period of 6 weeks will occur for all treatment arms. All patients who complete the 6-week treatment period or discontinue from the study treatment early will be asked to return for follow-up visits at Week 9 and Week 12 and complete assessments as specified in the SoA (Appendix 1). The visit window for follow-up visits is ±5 days.

After the last follow-up visit (Week 12), AEs should be followed as outlined in Section 5.1 and 5.5.

The last follow-up visit (Week 12) will be reported in eCRF as patient's study completion.

4.6.2.4 Assessments at Early Termination and Unscheduled Visits

Please see Appendix 1 for assessments that are required to be performed in case of an unscheduled visit.

If an unscheduled visit is required for safety reasons, necessary assessments will be undertaken at the discretion of the Investigator. 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 HV/patient. A duplicate sample should simultaneously be sent to the central laboratory for analysis.

In case of early termination of a subject in MAD portion of Part 1, a blood sample for PK assessment may be collected at the time of discontinuation. In case of premature study drug discontinuation in Part 2 (Section 4.7.1.1), a patient should be called for an unscheduled visit and assessments should be performed as listed in the SoA (Appendix 1) under "Study Drug Discontinuation", including physical examination, vital signs, ECG, safety labs, HBV parameters and PD assessments.

For Part 2 only: Chronic Hepatitis B Patients

In case of virological breakthrough, an unscheduled visit should be performed within one week of receiving HBV DNA results showing an increase in viral load of greater than one log10 above nadir. At the unscheduled visit, a confirmatory HBV DNA sample and a sample for viral resistance monitoring will be taken (see Appendix 1 and Section 4.6.1.8) and sent to one or several central laboratories or to the Sponsor for analysis. Additional unscheduled visits and assessments will be carried out as determined necessary by the treating physician. The Medical Monitor for the study should be informed as needed.

4.7 SUBJECT, STUDY, AND SITE DISCONTINUATION

4.7.1 Subject Discontinuation

The Investigator has the right to discontinue a HV/patient from RO7020531 or withdraw a HV/patient from the study at any time. In addition, HVs/patients 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:

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

HVs/patients who discontinue study treatment due to poor tolerability to study drug, or other safety related reasons, will not be replaced. HVs/patients who discontinue study treatment early for non-safety reasons may be replaced to ensure sufficient data to make dose-escalation and drug development decisions.

For Part 1, the decision to replace a subject will be made by a mutual agreement between the Sponsor and Investigator. Further information regarding the replacement policy is provided in Section 6.2.

For Part 2, the decision to replace a patient will be taken by the Sponsor.

4.7.1.1 Discontinuation from RO7020531

HVs/patients have the right to withdraw from the study at any time for any reason.

The Investigators have the right to withdraw HVs/patients from the study in the event of intercurrent illness, AE, pregnancy in female HVs/patients, for administrative or other reasons. The Sponsor should be informed of HVs'/patients' discontinuations from the study or from the study drug.

Part 1: MAD in Healthy Volunteers

For an individual subject in the MAD cohorts, dose continuation will not occur if the subject experiences any of the following:

- Clinically significant RO7020531-related changes in safety parameters that are considered not acceptable by the Investigator and/or the Sponsor; or
- Poor tolerability, which is considered to affect the HV's well-being and/or the PK evaluation.

Subjects who discontinue the MAD study prematurely will be asked to return to the clinic for a follow-up visit within 7 days after the last dose of study drug. The primary reason for premature study drug discontinuation should be documented on the appropriate eCRF. HVs who discontinue study drug prematurely for safety reasons will not be replaced.

For dose escalation criteria and stopping rules in SAD/MAD cohorts in Part 1 see Section 3.1.2.1.

Part 2: Chronic Hepatitis B Patients

Individual patients must discontinue RO7020531/placebo if they experience any of the following:

- Safety and tolerability issues, e.g., flu-like symptoms not tolerable and not manageable with symptomatic/*prophylactic* treatment *with acetaminophen (paracetamol) and/or dose reduction of RO7020531 (Cohort 4 only).*
- Grade 4 ALT (i.e., ≥ 10 × ULN) as defined by the Division of Acquired Immunodeficiency Syndrome (AIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events ("DAIDS AE grading table") (Appendix 6), confirmed within 48-72 hours.
- Grade 3 ALT (i.e., > 5 × ULN) combined with total bilirubin > 2 × ULN (of which > 35% is direct bilirubin), or Grade 3 ALT and INR > 1.5.
- Grade 4 hematology (Hb < 7.0 g/dL, absolute neutrophil count < 400 /mm^3, absolute lymphocyte count < 350/mm^3, or platelets < 25,000/mm^3).
- Any other confirmed (within 48-72h) Grade 3/4 laboratory abnormality deemed clinically significant (based on Investigator's assessment).
- Development of liver decompensation (ascites, varices, Child-Pugh Class B or C clinical classification).
- Confirmed virological breakthrough after ascertaining that patients are compliant with treatment administration.
- For Cohorts 1, 2 and 3: NUC analogue discontinuation due to safety issues or resistance development.

If *4* or more out of *15* patients (*12* receiving RO7020531 and *3* receiving placebo) are discontinued for drug-related safety reasons, enrollment will be halted and safety analysis will be performed. The results of the safety analysis will be provided to IEC/IRB and/or Health Authority, if applicable, before resuming enrollment.

Patients who discontinue study drug before the end of 6-week treatment period:

- Will not resume dosing with RO7020531/placebo. Dosing with NUC analogue in Cohorts 1, 2 and 3 will continue per each patient's physician discretion and local label.

All prematurely discontinued patients will be asked to come back for an unscheduled visit (see Section 4.6.2.4) to have assessments performed as listed in the SoA (Appendix 1) under "Study Drug Discontinuation / Early Termination".

All patients who discontinue the study treatment prematurely should complete the 6-week follow-up period (starting from the date of the last dose taken) as per SoA (Appendix 1). The primary reason for premature study drug discontinuation should be documented on the appropriate eCRF.

4.7.1.2 Discontinuation from Nucleos(t)ide Analogue (only for Part 2 Cohorts 1, 2 and 3)

In case of the development of NUC-resistance and/or NUC-related safety issues, discontinuation of NUC should be performed by each patient's physician in consultation with Investigator and per NUC local label.

Severe acute exacerbations of hepatitis B have been reported in patients who have discontinued anti-HBV therapy. Patients who discontinue NUC treatment should switch to another anti-HBV treatment at the discretion of each patient's physician.

If a patient discontinues NUC(s), the study drug (RO7020531/placebo) will also be withdrawn. Follow-up visits and assessments should be performed as listed in the SoA (Appendix 1).

If a patient does not switch to another anti-HBV treatment after a NUC discontinuation, hepatic function should be monitored closely with both clinical and laboratory follow-up for at least several months.

4.7.1.3 Withdrawal from Study

For withdrawal from the treatment phase in Part 2 see Section 4.7.1.1.

Every effort should be made to obtain information on HVs/patients who withdraw from the study, including withdrawal from the follow-up phase. The primary reason for withdrawal from the study treatment and/or full study (including follow up) should be documented on the appropriate eCRF.

HVs/patients will not be followed for any reason if consent has been withdrawn.

When a HV/patient voluntarily withdraws from the study, or is withdrawn by the Investigator, samples collected until the date of withdrawal will be analyzed, unless HV/patient specifically requests for these to be discarded or local laws require their immediate destruction. A HV's/patient's withdrawal from the study does not, by itself, constitute withdrawal of specimens donated to the Research Biosample Repository.

4.7.2 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 AEs in this or other studies indicates a potential health hazard to HVs/patients.
- HV/patient 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 ICH guideline for Good Clinical Practice

5. ASSESSMENT OF SAFETY

5.1 SAFETY PARAMETERS AND DEFINITIONS

Safety assessments will consist of monitoring and recording AE, including SAEs and non-serious AEs 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 5.1.2 and 5.1.3.

5.1.1 Adverse Events

According to the ICH guideline for Good Clinical Practice, an AE is any untoward medical occurrence in a clinical investigation HV/patient administered a pharmaceutical product, regardless of causal attribution. An AE 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), except as described in Section 5.3.5.9.
- 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.
- AEs 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 SAE is any AE that meets any of the following criteria:

- Fatal (i.e., the AE actually causes or leads to death).
- Life-threatening (i.e., the AE, in the view of the Investigator, places the HV/patient at immediate risk of death).

This does not include any AE 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 5.3.5.10).
- Results in persistent or significant disability/incapacity (i.e., the AE results in substantial disruption of the HV's/patient'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 HV/patient 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 AE (rated as mild, moderate, or severe, or according to a pre-defined grading criteria (see Section 5.3.3); 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 AE recorded on the eCRF.

SAEs 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 5.3.8 for reporting instructions).

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

Non-serious AEs 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 5.3.8 for reporting instructions). AEs 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 5.3.5.6.
- 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 HV/patient exposed to a medicinal product. This term applies only when a contamination of the study drug is suspected.

5.2 SAFETY PLAN

5.2.1 Management of Specific Adverse Events

- *Close monitoring of* flu-like symptoms: TLR7 agonists can be associated with dose-dependent flu-like symptoms; a percentage of HVs/patients may develop fever, chills, fatigue, myalgia, malaise, and headache within several hours after the dosing with RO7020531/*placebo*. Symptomatic treatment with acetaminophen (paracetamol) is recommended in case these events occur. There is an optional overnight stay in the clinical unit on Day 1 at the discretion of the Investigator for monitoring patients in Cohort 4 who develop flu-like symptoms/pyrexia during the first 12 hours post-dosing and do not respond to symptomatic treatment with acetaminophen (paracetamol). Additionally, the safety monitoring after dosing on Day 3 and Day 5 can be extended from 8 to 12 hours, at the discretion of the Investigator, in patients from Cohort 4 who develop flu-like symptoms/pyrexia and do not respond to symptomatic treatment. If at any time patients develop flu-like symptoms with pyrexia Grade 2 or above per DAIDS grading scale (38.6 °C or 101.5 °F), it is recommended that they call the site so that the Investigator can advise whether the patient needs to attend an unscheduled visit or receive symptomatic treatment with acetaminophen (paracetamol) up to 2 g/day.
- [redacted]
  - [redacted]

- For abnormal liver function tests in CHB patients (Part 2), see Section 5.3.5.6. For the drug discontinuation in patients with abnormal liver function tests, see Section 4.7.1.1.

[1] *Based either on central laboratory (as per the SoA in Appendix 1) and/or local laboratory if performed for patient management and/or decision making. In such a scenario, if local laboratory tests are performed, an additional sample will be sent to the central laboratory, but decisions will be based on local laboratory results.*

Liver Flare Monitoring: Liver flares, identified by an abrupt elevation of serum ALT, are acute hepatitis exacerbations due to changes in the immune response against HBV and its downstream mechanisms. These flares will be monitored for the duration of the study in Part 2. ALT elevations of 2 to 3 times the baseline value that are self-limited are likely to reflect a treatment response rather than drug toxicity. Progressive increases, increases inconsistent with the time course of a "flare" or ALT elevations associated with increases in bilirubin and alkaline phosphatase should be treated as AE. The Investigator should aim to exclude development of decompensated liver disease. Patients who develop signs of decompensated liver disease (e.g., ascites, varices, Child-Pugh Class B or C clinical classification (Appendix 3) should discontinue study treatment (Section 4.7.1.1). Patients who develop flares should be monitored more closely with additional unscheduled visits and laboratory assessments.

No dose modification of RO7020531/placebo for *liver-related safety* is expected *in Part 2 Cohort 4. No abnormal liver laboratory parameters were observed in Cohorts 1 to 3, including in those subjects with flu-like symptoms.* At the discretion of the Investigator, study treatment can be discontinued. For the treatment stopping rules in individual HVs/patients and dose escalation stopping rules, please see Sections 3.1.2 and 4.7.1.1.

For the management of NUC analogue-related AE, please refer to the corresponding label.

5.3 METHODS AND TIMING FOR CAPTURING AND ASSESSING SAFETY PARAMETERS

The Investigator is responsible for ensuring that all AEs (see Section 5.1.1 for definition) are recorded on the AE eCRF and reported to the Sponsor in accordance with instructions provided in this section and in Section 5.1 and 5.5.

For each AE recorded on the AE eCRF, the Investigator will make an assessment of seriousness (Section 5.1.2 for seriousness criteria), severity (Section 5.3.3), and causality (Section 5.3.4).

5.3.1 Adverse Event Reporting Period

Investigators will seek information on AEs at each HV/patient contact. All AE, whether reported by the HV/patient or noted by study personnel, will be recorded in the HV's/patient's medical record. AEs will then be reported on the AE eCRF as follows:

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

After initiation of study drug, all AE, regardless of relationship to study drug, will be reported until the end of follow-up, which is 28 days after the last dose for HVs and 6 weeks after the last dose for patients.

After the end of follow-up period (28 days after last dose for HVs and 6 weeks after last dose for patients), investigators should report any deaths, SAE, or other AE of concern that are believed to be related to prior treatment with study drug (see Section 5.5).

5.3.2 Eliciting Adverse Event Information

A consistent methodology of non-directive questioning should be adopted for eliciting AE information at all HV/patient 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?"

5.3.3 Assessment of Severity of Adverse Events

Table 1 provides guidance for assessing AE severity.

Table 1 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).

5.3.4 Assessment of Causality of Adverse Events

Investigators should use their knowledge of the HV/patient, the circumstances surrounding the event, and an evaluation of any potential alternative causes to determine whether or not an AE 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 or discontinuation 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 HV/patient 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

For patients, causality will be assessed individually for RO7020531 and for NUC analogue(s), if applicable.

5.3.5 Procedures for Recording Adverse Events

Investigators should use correct medical terminology/concepts when recording AEs on the AE eCRF. Avoid colloquialisms and abbreviations.

Only one AE term should be recorded in the event field on the AE eCRF.

For Part 2, Cohort 4, there may be a flu-like symptom specific AE eCRF page, within which each of the symptoms experienced can be captured as well as two allocated eCRF pages for capturing hypotension and hypoxia AEs in case these occur and whether they required medical intervention and management.

5.3.5.1 Diagnosis versus Signs and Symptoms

A diagnosis (if known) should be recorded on the AE 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 AE eCRF. If a diagnosis is subsequently established, all previously reported AEs based on signs and symptoms should be nullified and replaced by one AE report based on the single diagnosis, with a starting date that corresponds to the starting date of the first symptom of the eventual diagnosis.

5.3.5.2 Adverse Events Occurring Secondary to Other Events

In general, AEs 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 AEs occurring secondary to an initiating event that are separated in time should be recorded as independent events on the AE 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 GI 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 AEs should be recorded separately on the AE eCRF if it is unclear as to whether the events are associated.

5.3.5.3 Persistent or Recurrent Adverse Events

A persistent AE is one that extends continuously, without resolution, between HV/patient evaluation time-points. Such events should only be recorded once on the AE 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 AE eCRF should be updated to reflect this.

A recurrent AE is one that resolves between HV/patient evaluation time-points and subsequently recurs. Each recurrence of an AE should be recorded separately on the AE eCRF.

5.3.5.4 Abnormal Laboratory Values

Not every laboratory abnormality qualifies as an AE. A laboratory test result should be reported as an AE 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 AE.

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

If a clinically significant laboratory abnormality is not a sign of a disease or syndrome, the abnormality itself should be recorded on the AE 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 AE. 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 AE 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.

5.3.5.5 Abnormal Vital Sign Values

Not every vital sign abnormality qualifies as an AE. A vital sign result should be reported as an AE 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 AE.

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 AE eCRF.

Observations of the same clinically significant vital sign abnormality from visit to visit should not be repeatedly recorded on the AE 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.

5.3.5.6 Abnormal Liver Function Tests

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

For Healthy Volunteers and CHB patients in Cohorts 1, 2 and 3:

- Treatment-emergent ALT or AST $>3\times$ULN in combination with total bilirubin $>2\times$ULN (of which 35% is direct bilirubin)
- Treatment-emergent ALT or AST $>3\times$ULN in combination with clinical jaundice

For CHB patients in Cohort 4:

- Treatment-emergent ALT or AST $>5\times$ULN and $2\times$ baseline in combination with total bilirubin $>2\times$ULN (of which 35% is direct bilirubin)
- Treatment-emergent ALT or AST $>5\times$ULN in combination with clinical jaundice
- Hepatitis flare, defined as ALT $>10\times$ULN and $>2\times$ baseline level

The most appropriate diagnosis or (if a diagnosis cannot be established) the abnormal laboratory values should be recorded on the AE eCRF (see Section 5.3.5.1) and

reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event), either as a SAE or a non-serious AE of special interest (see Section 5.3.8).

For the drug discontinuation in patients with abnormal liver function tests, see Section 4.7.1.1.

For Part 2 (CHB patients), documented liver biopsy or Fibroscan® (or equivalent elastography) test results demonstrating chronic HBV infection with absence of cirrhosis and absence of extensive bridging fibrosis (absence of cirrhosis and absence of extensive bridging fibrosis is defined as Metavir <3, recommended cutoff for fibroscan 8.5 kPa) must be available within 6 months prior to randomization. If no results are available, patients must agree to have either a liver biopsy or Fibroscan® (or equivalent) performed during screening.

For liver flares monitoring see Section 5.2.1.

5.3.5.7 Deaths

All deaths that occur during the protocol-specified AE reporting period (Section 5.3.1), regardless of relationship to study drug, must be recorded on the AE eCRF and immediately reported to the Sponsor (Section 5.4.2). This includes death attributed to progression of chronic HBV infection.

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 AE 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 patient with or without preexisting heart disease, within one hour of the onset of acute symptoms or, in the case of an unwitnessed death, within 24 hours after the patient 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 AE eCRF. If the cause of death later becomes available (e.g., after autopsy), "unexplained death" should be replaced by the established cause of death.

5.3.5.8 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 Medical History eCRF.

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

5.3.5.9 Lack of Efficacy or Worsening of Chronic Hepatitis B Virus Infection

As this is the first study of RO7020531 in CHB patients (Part 2 only) some of whom are virologically suppressed (Cohorts 1, 2 & 3) and some of whom are treatment naïve (Cohort 4), the effects on viral parameters are being explored as part of the exploratory endpoints in this short study. No functional HBV cure outcome is expected in this study. Lack of RO7020531 efficacy in terms of changes in HBeAg and HBsAg does not qualify for AE in this study.

Medical occurrences or symptoms of deterioration that are anticipated as part of chronic HBV infection should be recorded as AEs if judged by the Investigator to have unexpectedly worsened in severity or frequency or changed in nature at any time during the study. When recording an unanticipated worsening of chronic HBV infection on the AE eCRF, it is important to convey the concept that the condition has changed by including applicable descriptors.

In the event of development of liver decompensation (ascites, varices, Child-Pugh Class B or C clinical classification etc.), this should be reported as an AE and the patient should be discontinued from the study Section 4.7.1.1.

For virological breakthrough, see Section 4.6.1.8 and for liver flares monitoring, see Section 5.2.1. Each of these should be reported as AE.

5.3.5.10 Hospitalization or Prolonged Hospitalization

Any AE that results in hospitalization or prolonged hospitalization should be documented and reported as a SAE (per the definition of SAE in Section 5.1.2), except as outlined below.

The following hospitalization scenarios are not considered to be *an* SAE:

- Hospitalization for respite care
- Planned in-clinic stay for HVs in SAD and MAD portions of Part 1.
- Planned in-clinic stay of patients in Part 2 (optional) for intensive PK sampling or because of flu-like symptoms/pyrexia (Part 2, Cohort 4) *as per Section 5.2.1).*

- Hospitalization for a pre-existing 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 HV/patient has not suffered an AE.
- *If for administrative reasons, e.g. the patient lives far way and is kept in the clinic/hospital for patient and/or site convenience (Part 2, Cohort 4).*

5.3.5.11 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 AE 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 AEs associated with an overdose or incorrect administration of study drug should be recorded on the AE eCRF. If the associated AE 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 5.3.8).

5.3.6 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:

- SAE
- Non-serious AE of special interest
- Pregnancies

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 SAEs to the local health authority and IEC/IRB.

5.3.7 Emergency Medical Contacts

To ensure the safety of HVs/patients, 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".

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

5.3.8.1 Events that Occur prior to Study Drug Initiation

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. The Serious Adverse Event/Adverse Event of Special Interest Reporting Form provided to investigators should be completed and submitted to the SAE Responsible immediately (i.e., no more than 24 hours after learning of the event).

5.3.8.2 Events that Occur after Study Drug Initiation

For reports of serious adverse events and non-serious adverse events of special interest (see Sections 5.1.2 and 5.1.3) that occur after initiation of study drug, investigators should record all case details that can be gathered on the Serious Adverse Reporting Form and forward this form to the SAE Responsible within 24 hours.

In the case of electronic reporting, investigators should record all case details that can be gathered immediately (i.e., within 24 hours after learning of the event) on the appropriate Serious Adverse Event / Adverse Event of Special Interest 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 SAE Responsible immediately (i.e., no more than 24 hours after learning of the event).

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

5.3.9 Reporting Requirements for Pregnancies

5.3.9.1 Pregnancies in Female Healthy Volunteers/Patients

A female subject/patient of childbearing potential will be instructed to immediately inform the Investigator if she becomes pregnant until the end of the follow-up period of 28 days in Part 1 or 6 weeks in Part 2.

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 in Part 1 or Part 2. Pregnancy should not be recorded on the AE eCRF. The Investigator should discontinue study drug and counsel the HV/patient, discussing the risks of the pregnancy and the possible effects on the fetus. Monitoring of the patient should continue until conclusion of the pregnancy.

5.3.9.2 Pregnancies in Female Partners of Male Subjects

Male HVs/patients will be instructed through the Informed Consent Form to immediately inform the Investigator if their partner becomes pregnant during the study until the end of follow up period (28 days after the last dose of study drug for Part 1 and 6 weeks after the last dose for Part 2). 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 HV/patient 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 HV/patient 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.

5.3.9.3 Abortions

Any spontaneous abortion should be classified as an SAE (as the Sponsor considers spontaneous abortions to be medically significant events), recorded on the AE eCRF, and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5.3.8).

Any induced abortion due to maternal toxicity and/or embryo-fetal toxicity should also be classified as an SAE, recorded on the AE eCRF, and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5.3.8).

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.

5.3.9.4 Congenital Anomalies/Birth Defects

Any congenital anomaly/birth defect in a child born to a female HV/patient or female partner of a male HV/patient exposed to study drug should be classified as an SAE, recorded on the AE eCRF, and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5.3.8).

5.4 FOLLOW-UP OF SUBJECTS AFTER ADVERSE EVENTS

5.4.1 Investigator Follow-Up

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

During the study period, resolution of AEs (with dates) should be documented on the AE eCRF and in the HV's/patient'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 AE eCRF.

All pregnancies reported during the study should be followed until the pregnancy outcome, and reported according to the instructions provided in Section 5.3.9.

5.4.2 Sponsor Follow-Up

For SAEs, non-serious AEs 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 HV/patients for AEs after the end of the AE reporting period (defined as end of follow-up period: 28 days after last study drug dose for HVs and 6 weeks after last study drug dose for patients).

If the Investigator becomes aware of any other SAE occurring after the end of the AE reporting period, and 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 SAE 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 SAEs and non-serious AEs of special interest against cumulative product experience to identify and expeditiously communicate possible new safety findings to investigators, IRBs, IECs, and applicable health authorities based on applicable legislation.

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

- RO7020531 IB

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 analysis of complete data for the study will be performed when all HVs/patients have either completed the follow-up period or have discontinued early from the study, all data from the study are in the database and have been cleaned and verified, and the database is locked.

This is an exploratory study for which no formal hypothesis testing will be done and thus no adjustment for multiplicity of testing will be performed.

6.1 DETERMINATION OF SAMPLE SIZE

Part 1: SAD and MAD in Healthy Volunteers

The current planned study design and sample size complies with standard safety review rules applied in single and multiple ascending dose studies.

Part 2: Chronic Hepatitis B Patients

Sample size is determined based on clinical judgment and practical considerations. With eight patients treated with RO7020531, there is a 90% chance to observe at least one AE if the underlying event incidence rate is 25% in the patient population.

6.2 REPLACEMENT POLICY (ENSURING ADEQUATE NUMBERS OF EVALUABLE SUBJECTS)

HVs/patients prematurely discontinued from the study for non-safety reasons may be replaced to ensure adequate numbers of evaluable HVs/patients. HV/patient replacement numbers will be provided by Roche or its designee. For more details see Section 4.7.1.

The decision to replace a withdrawn HV/patient will be made at the discretion of the Sponsor and the investigator for Part 1, and at the discretion of the Sponsor for Part 2.

6.3 SUMMARIES OF CONDUCT OF STUDY

The number of HVs/patients who are randomized, discontinue, or complete the study treatment and/or the entire study (including follow up period) will be summarized by treatment. Reasons for premature study withdrawal (treatment phase and follow up phase) will be listed and summarized by treatment. Enrollment and protocol deviations will be listed and evaluated for their potential impact on interpretation of study results.

6.4 ANALYSIS POPULATIONS

6.4.1 Safety Analysis Population

All HVs/patients who have received at least one dose of the study medication, whether prematurely withdrawn from the study or not, will be included in the safety analysis. HVs/patients will be analyzed according to the treatment actually received.

6.4.2 Pharmacokinetic Analysis Population

A per protocol analysis including all HVs/patients randomized and adherent to the protocol will be performed. HVs/patients 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 PK 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.

6.4.3 Pharmacodynamic Analysis Population

The analyses of PD data will include all HVs/patients who were randomized, received at least one dose of study medication (RO7020531 or placebo), and have PD data available. HVs/patients will be analyzed according to the treatment group to which they were randomized.

6.5 SUMMARIES OF TREATMENT GROUP COMPARABILITY

Descriptive statistics will be generated for demographic and baseline disease characteristics including sex, race, ethnicity, origin (for Asian HVs/patients only), age, weight, height, body mass index, HBV DNA and HBsAg levels, and HBV history including but not limited to duration of HBV disease, previous HBV treatments, length of time on NUCs. Data for study drug administration and concomitant medication will be listed. The number of HVs/patients who were randomized, discontinued treatment period, completed treatment period, discontinued study and completed the study (including follow-up period) will be summarized.

6.6 PRIMARY AND SECONDARY STUDY VARIABLES

The primary study variables are those measuring safety and tolerability. The PK and PD measures of the active metabolite RO7011785, following single and multiple doses of RO7020531, are secondary variables.

6.7 SAFETY ANALYSES

All safety analyses will be based on the safety analysis population.

6.7.1 Adverse Events

The original terms recorded on the eCRF by the Investigator for AEs will be standardized by the Sponsor by assigning preferred terms from the Medical Dictionary for Drug Regulatory Affairs (MedDRA). AEs will be summarized by mapped term and appropriate thesaurus level.

AEs will be described by individual listings and frequency tables broken down by body system.

6.7.2 Clinical Laboratory Test Results

All clinical laboratory data will be stored on the database in the units in which they were reported. HV's/patient's 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.

Safety analyses to determine if treatment groups are safe to proceed will use all available data reported from local and/or central laboratory results. Unscheduled local laboratory tests may be ordered per investigator discretion and may be used for the individual management of the HV/patient. A duplicate sample should simultaneously be sent to the central laboratory for analysis (Part 2 only).

6.7.2.1 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.

6.7.2.2 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 HV/patient 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 HV/patient, the midpoint of the standard reference range will be used as the HV/patient's baseline value for the purposes of determining marked laboratory abnormalities. Marked laboratory abnormalities will be labeled in the HV/patient listings as "HH" for very high or "LL" for very low.

6.7.3 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.

6.7.4 ECG Data Analysis

ECG data will be presented by individual listings with flagging of values outside the normal ranges and flagging of marked and clinically significant 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. 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 ≤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.

6.7.5 Concomitant Medications

The original terms recorded on the HVs'/patients' 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.

6.8 PHARMACOKINETIC ANALYSES

Non-compartmental analysis using WinNonlin software will be used to calculate PK parameters where appropriate. Summary descriptive statistics of plasma PK parameters including C_{max}, T_{max}, AUC_{inf}, AUC_{last} and $t_{1/2}$ for RO7020531 and RO7011785 and additional metabolites including RO7018822 and RO7033805, will be presented by treatment arm including mean, SD, CV, medians and ranges. Where appropriate, data may be pooled and analyzed for example, all single dose data may be pooled. Listings, summary tables and graphs (individual plots and/or mean plots) by treatment group will be provided. Descriptive statistics of urine PK parameters for RO7020531 and RO7011875 and additional metabolites including RO7018822 and RO703380 will be presented, where available. PK and PD data from this study may be used to develop a population PK/PD model.

HVs/patients will be excluded from the PK analysis if data are unavailable which may influence the analysis (see Section 6.4.2). Where appropriate, listings and summary tables of tenofovir (including tenofovir alafenamide, if approved for HBV and applicable), entecavir, adefovir and telbivudine concentrations will be provided based on the sparse sampling throughout the study.

6.9 PHARMACODYNAMIC ANALYSES

Summary descriptive statistics will be presented for the induction of cytokines, chemokines, and neopterin and of interferon-response genes separately by treatment arm. Exploratory analysis will be performed to assess the interferon-induced response under different dosing conditions. Graphical and statistical techniques including linear, nonlinear, and logistic regression will be used to explore potential relationships between dosing regimen, PK and PD.

6.10 EXPLORATORY ANALYSES

All antiviral endpoints are considered exploratory for this study (Part 2 only). Summary descriptive statistics will be used to summarize the antiviral outcome measures of qualitative HBsAg, quantitative HBsAg actual and change from baseline, and HBeAg, anti-HBe and anti-HBs status at each of the time-points by treatment group. For Cohort 4, the outcome of antiviral response will additionally include quantitative HBV DNA level (actual and change from baseline). Status of HBsAg and HBeAg seroconversion, if any, and maintenance of HBV DNA levels less than 90 IU/mL (Cohorts 1, 2 and 3) will also be summarized. The findings from the resistance analyses will be listed for each patient selected for analysis.

Exploratory analysis may also be performed to identify markers and/or marker panels correlating with and/or potentially predictive of an in vivo PD response, the occurrence of certain AEs (to be defined) and/or changes in viral parameters.

A detailed exploratory analysis plan will be developed when initial study results (AE, viral/antiviral parameters in Part 2) have been reviewed.

6.10.1 Analyses of Antiviral Measures (Part 2 Only)

Summary descriptive statistics will be used to summarize the antiviral outcome measures of qualitative HBsAg, quantitative HBsAg actual and change from baseline, and HBeAg, anti-HBe and anti-HBs status at each of the time-points by treatment group. Status of HBsAg and HBeAg seroconversion, if any, and maintenance of HBV DNA levels less than 90 IU/mL (Cohorts 1, 2 and 3) will also be summarized.

The findings from the resistance analyses will be listed for each patient selected for analysis.

6.10.2 Analysis to Support Dose-Escalation Decisions and Dose-Selection

Bayesian adaptive methods such as continual re-assessment method (CRM) may be applied once a dose level in the SAD part, which is considered to be safe and also exhibits a response for at least one member of the screening panel of biomarkers (e.g., IL-10, IP-10) indicating activation of the TLR pathway, is identified to initiate dosing of MAD cohorts. However, model-based CRM dose-escalation approaches will only be considered as supporting information for the dose-escalation and/or dose selection. Clinical judgment will always take precedence in making the final decisions.

There are two main endpoints of this study: safety and characterization of TLR PD biomarker activity. Two types of modelling approaches for the next best dose recommendation may be explored in Part 1 MAD for HVs and Part 2 for CHB patients. Operating characteristics of both models will be compared to determine the appropriateness.

a) Based only on dose-limiting event (DLE) responses:

N-CRM with control for the probability of over-dosing, based on occurrence of a DLE may be applied to inform decision about the dose of RO7020531 to be given to the next cohort during dose-escalation. A DLE will be defined as any treatment-related adverse reaction as described in Section 3.1.2 (e.g., AE, laboratory abnormality, etc.) that would prevent another drug administration at the same dose level in a given HV/patient.

Details on the model/algorithm are given below. The relationship between the dose and the probability of observing a DLE will be described by the following two-parameter logistic regression model:

$$\text{logit}(p) = \log\left(\frac{p}{1-p}\right) = \alpha + \beta \log\left(\frac{dose}{ref}\right)$$

where p is the probability of observing a DLE at a given dose, α is the log of the odds for p at the reference dose (ref, here equals to X mg, starting dose of MAD) and β is the change in the log odds for an e1-fold increase in dose (detailed discussion about this model parameterization can be found in Neuenschwander et al 2008).

The model will be estimated using a Bayesian framework; priors for α and β will be specified using the data from the already completed cohorts of the SAD part of the study. Given the data, Markov Chain Monte Carlo (MCMC) sampling will be used for obtaining the posterior distribution of the model parameters.

After each cohort of HVs/patients completes dosing, the model will be updated with the DLE–observed occurrence and a new MTD will be defined as the dose such that:

- The probability of being within the target safety interval (of 20% to 35% DLE rate) is maximized and
- The probability of being within the excessive toxicity interval (above 35% DLE rate) is below 30%.

b) Dose-escalation based on dual endpoint (modelling both DLE and PD biomarker response)

This model may be flexibly applied based on the availability of the data for the panel of biomarkers constituting the result of activation the TLR pathway. The aim of the dual endpoint design is to estimate an optimal dose level which represents an optimal trade-off between safety and biomarker response.

For joint modelling of DLE and PD biomarker response (assuming correlation), the Bayesian approach developed by (Bekele and Shen 2005) and as implemented in the R package crmPack (Bove et al 2016) will be considered for the escalation process. A DLE for this model may be limited to any treatment mechanism of action (MOA)-related adverse reaction (e.g., severe flu-like symptoms, not tolerable and not manageable with

symptomatic treatment). Alternatively, another Bayesian dose-escalation approach modelling the two endpoints separately (assuming no correlation) may also be explored. In this, the biomarker response would be modelled as a linear log-log function of dose and a gain function applied to select the next best dose (Yeung et al 2015).

6.11 INTERIM ANALYSES

There will be no Interim Analysis 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 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. Central and, if applicable, local laboratory data and/or other 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 HV/patient randomized, an eCRF must be completed and electronically signed by the Principal Investigator or authorized delegate from the study staff. If a HV/patient discontinues the study treatment early or withdraws from the study completely, the reason must be noted on the eCRF. If a HV/patient is withdrawn from the study because of a treatment-limiting AE, 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/contract research organization (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 HV/patient 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 HV/patient 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, HV/patient-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, HV/patient 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 IEC/IRB 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, HV/patient 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 each 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 IEC/IRB submission. The final IEC/IRB-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 HV/patient or the HV's/patient's legally authorized representative before his or her participation in the study. The case history or clinical records for each HV/patient 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 HV/patient to participate. The final revised IEC/IRB-approved Consent Forms must be provided to the Sponsor for Health Authority submission purposes.

HVs/patients 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 IEC/IRB policy) during their participation in the study. For any updated or revised Consent Forms, the case history or clinical records for each HV/patient 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 HV/patient or the HV's/patient's legally authorized representative. All signed and dated Consent Forms must remain in each HV's/patient'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 HV/patients, and relevant supporting information must be submitted to the IEC/IRB by the Principal Investigator and reviewed and approved by the IEC/IRB before the study is initiated. In addition, any HV/patient recruitment materials must be approved by the IEC/IRB.

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

In addition to the requirements for reporting all AEs to the Sponsor, investigators must comply with requirements for reporting SAEs to the local Health Authority and IEC/IRB. 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 IEC/IRB, and archived in the site's study file.

8.4 CONFIDENTIALITY

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

HV/patient 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 HV/patient, unless permitted or required by law.

Medical information may be given to a HV/patient's personal physician or other appropriate medical personnel responsible for the HV's/patient'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 IEC/IRB 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 (last HV/patient last visit).

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 IEC/IRB and governmental approval. In addition, at the end of the study, the Investigator will receive the HV/patient data, which includes an audit trail containing a complete record of all changes to data.

Roche shall also submit an Annual Safety Report once a year to the IEC/IRB and Health Authorities, if applicable, according to local regulatory requirements and timelines of each country participating in the study.

9.2 SITE INSPECTIONS

Site visits will be conducted by the Sponsor or an authorized representative for inspection of study data, HVs'/patients' medical records, and eCRFs. The Investigator will permit national and local Health Authorities, Sponsor monitors, representatives, and collaborators, and the IEC/IRBs 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.

The Sponsor is also responsible for managing CROs, IxRS and central laboratories used in the study.

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 IEC/IRB and to regulatory authorities in accordance with local regulatory requirements.

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

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Appendix 1 Schedule of Assessments: SAD

Visit	Screening							Follow Up Visit	Follow Up Call
Day	D-28 to D-3	Day -2	Day -1	Day 1	Day 2	Day 3	Day 5	Day 8[a]	Day 29[b]
Informed Consent	x								
Eligibility	x		x						
Demography	x								
Medical History	x								
Physical Examination[c]	x		x					x	
Vital Signs[d]	x		6	7	2	x		x	
ECG-12 lead[e]	x		6	7	x	x		x	
PK Plasma Sample				12	2	x			
Urine PK Sample				5	x				
PD for Protein Biomarkers			x	4	x	x[f]	x[f]	x	
Whole Blood for RNA			x	4	x				
Hematology, Blood Chemistry, Coagulation, Urinalysis	x		x		x			x	
Pregnancy Test[g]	x		x					x	
Administration of Study Medication[h]				x					
Randomisation				x					
Autoimmune panel, Thyroid function	x								
Viral Serology (HAV, HBV, HCV, HIV)	x								
Follicle Stimulating Hormone[i]	x								
Substance Use[j]	x		x						
In-Clinic Stay[k]		x	x	x	x	x			
Ambulatory Visit	x						x	x	
Whole Blood for RBR DNA[l]				x					
Clinical Genotyping[m]				x					
Adverse Events	x[n]	x[n]	x[n]	x	x	x	x	x	x
Previous and Concomitant Treatments	x	x	x	x	x	x	x	x	x

Appendix 1 Schedule of Assessments: SAD (cont.):

a) A follow up visit to be completed 7 days (±1) after the last dose of study medication.

b) A follow up call to be completed 28 days (±3) after the last dose of study medication.

c) Full physical exam, including recording of weight, is required at screening, Day –1 and Follow-Up visit. At all other visits (or as clinically indicated), limited, symptom-directed physical examinations should be performed at the discretion of the Investigator. Height will only be recorded at screening. BMI will be calculated at screening.

d) Vital signs include blood pressure, pulse rate, respiratory rate and body temperature. Blood pressure, respiratory rate and pulse rate will be obtained after the patient has been in a supine or sitting position for at least 5 minutes. Blood pressure measurement will be performed in triplicate (can be as short as 20 second to 1 minute interval between measurements). Pulse rate and body temperature measurement will be performed as single assessments.

e) 12-lead ECGs will be obtained in triplicate (three consecutive interpretable 12-lead ECGs within a 2–5 minute interval) after the subject has been in a supine position for at least 10 minutes. Automated ECG intervals (PR (PQ), QRS, QT, QTcF) and HR will be captured or calculated, and the changes in T-wave and U-wave morphology will be documented.

f) PD samples at 48 and 96 hours for neopterin only (no other PD markers).

g) Serum or plasma beta-human chorionic gonadotropin (β-HCG) at screening, urine on all other occasions (females only).

h) Study drug will be administered in fasted state (after an overnight fast of at least 10 hours).

i) Follicle stimulating hormone (females only to confirm post-menopausal status, performed at screening only).

j) At screening and Day –1 only (drugs of abuse and alcohol test).

k) Subjects will be admitted to the unit on Day -2 to start time-matching ECG assessments in the morning of Day –1. The discharge from the unit will be on Day 3 after 48 h assessments.

l) Blood for RBR DNA sample will be collected on Day 1 from all patients who signed RBR ICF. If, however, the RBR DNA blood sample is not collected during the scheduled visit, it may be collected at any time during the conduct of the clinical study.

m) If the clinical genotyping blood sample is not collected at Day 1, it may be collected at any time during the conduct of the clinical study.

n) Prior to dosing, only serious adverse events caused by a protocol-mandated intervention should be reported.

Appendix 1 Schedule of Assessments: SAD Detailed

Visit	Day	Scheduled Time (h)	Vital Signs[a]	ECG-12 lead[b]	Hematology, Blood Chemistry, Coagulation, Urinalysis	Plasma PK Sample[c]	Urine PK Sample[d]	PD for Protein Biomarkers[e]	Whole Blood for RNA[e]	Whole Blood for RBR DNA[f]	Clinical Genotyping[g]	Administration of Study Medication[h]
	Day -1	0			X			X	X			
		0.5	X	X								
		1	X	X								
		2	X	X								
		4	X	X								
		6	X	X								
		12	X	X								
	Day 1	Predose	X	X		X	X	X	X	X	X	
		0					0-4					X
		0.25				X						
		0.5	X	X		X						
		1	X	X		X						
		1.5				X						
		2	X	X		X		X	X			
		3				X						
		4	X	X		X						
		6	X	X		X	4-8	X	X			
		8				X						
		12	X	X		X	8-12	X	X			
		18				X	12-24					
	Day 2	24	X	X	X	X		X	X			
		36	X			X						
	Day 3	48	X	X		X		X				
	Day 5	96						X				
Follow Up Visit	Day 8[i]		X	X	X			X	X			

Appendix 1 Schedule of Assessments: SAD Detailed

a) Vital signs include blood pressure, pulse rate, respiratory rate and body temperature. Blood pressure, respiratory rate and pulse rate will be obtained after the patient has been in a supine or sitting position for at least 5 minutes. Blood pressure measurement will be performed in triplicate (can be as short as 20 second to 1 minute interval between measurements). Pulse rate and body temperature measurement will be performed as single assessments.

b) 12-lead ECGs will be obtained in triplicate (three consecutive interpretable 12-lead ECGs within a 2–5 minute interval) after the subject has been in a supine position for at least 10 minutes. Automated ECG intervals (PR (PQ), QRS, QT, QTcF) and HR will be captured or calculated, and the changes in T-wave and U-wave morphology will be documented.

c) Plasma PK samples will be collected at pre-dose and 0.25, 0.5, 1, 1.5, 2, 3, 4, 6, 8, 12, 18, 24, 36, and 48 hours post-dose.

d) A single void urine sample will be collected pre-dose on Day 1. Pooled urine PK samples will be collected 0 to 4 hours, 4 to 8 hours, 8 to 12 hours and 12 to 24 hours post-dose.

e) PD samples will be collected at pre-dose, 2, 6, 12, 24, 48, and 96 hours post-dose. Samples at 48 and 96 hours will be collected for neopterin only (no other PD markers).

f) Blood for RBR DNA sample will be collected on Day 1 from all patients who signed RBR ICF. If, however, the RBR DNA blood sample is not collected during the scheduled visit, it may be collected at any time during the conduct of the clinical study.

g) If the clinical genotyping blood sample is not collected at Day 1, it may be collected at any time during the conduct of the clinical study.

h) Study drug will be administered in fasted state (after an overnight fast of at least 10 hours).

i) A follow up visit to be completed 7 days (±1) after the last dose of study medication.

Appendix 1 Schedule of Assessments: MAD

Visit	Screening	Treatment Period																Follow Up Visit	Follow Up Call
Day	D-28 to D-3	Day -2	Day -1	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10	Day 11	Day 12	Day 13	Day 14	Day 20[a]	Day 41[b]
Informed Consent	X																		
Eligibility	X		X																
Demography, Medical History	X																		
Physical Examination[c]	X		X							X							X	X	
Vital Signs[d]	X		5	6	X	4	X	2	X	2	X	2	X	2	X	6	X	X	
ECG-12 lead[e]	X		5	6		3				X						6		X	
PK Plasma Sample				12	X	3	X	3	X	3	X	3	X	3	X	12	X		
PD for Protein Biomarkers			X	4	X	3	X	3	X	3	X					3	X	X	
Whole Blood for RNA			X	4	X	3	X	3	X	3	X					3	X	X	
B-Cell Activation Panel[f]			X			X											X	X	
Hematology, Blood Chemistry, Coagulation, Urinalysis	X		X				X				X			X		X	X	X	
Pregnancy Test[g]	X		X															X	
Administration of Study Medication[h]				X		X		X		X		X		X		X			
In-Clinic Stay[i]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X		
Ambulatory Visit	X																	X	
Randomisation				X															
Autoimmune panel, Thyroid function	X																		
Viral Serology (HAV, HBV, HCV, HIV)	X																		
Follicle Stimulating Hormone[j]	X																		
Substance Use[k]	X		X																
Clinical Genotyping[l]				X															
Whole Blood for RBR DNA[m]				X															
Adverse Events, Previous and Concomitant Treatments	X[n]	X[n]	X[n]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Appendix 1 Schedule of Assessments: MAD

a) A follow up visit to be completed 7 days (±1) after the last dose of study medication.

b) A follow up call to be completed 28 days (±3) after the last dose of study medication.

c) Full physical exam, including recording of weight, is required at screening, Day –1 and Follow-Up visit. At all other visits, including Day 7 and Day 14 (or as clinically indicated), limited, symptom-directed physical examinations should be performed at the discretion of the Investigator. Height will only be recorded at screening. BMI will be calculated at screening.

d) Vital signs include blood pressure, pulse rate, respiratory rate and body temperature. Blood pressure, respiratory rate and pulse rate will be obtained after the patient has been in a supine or sitting position for at least 5 minutes. Blood pressure measurement will be performed in triplicate (can be as short as 20 second to 1 minute interval between measurements). Pulse rate and body temperature measurement will be performed as single assessments.

e) 12-lead ECGs will be obtained in triplicate (three consecutive interpretable 12-lead ECGs within a 2–5 minute interval) after the subject has been in a supine position for at least 10 minutes. Automated ECG intervals (PR (PQ), QRS, QT, QTcF) and HR will be captured or calculated, and the changes in T-wave and U-wave morphology will be documented.

f) B-cell activation panel may be assessed at selected sites only.

g) Serum or plasma beta-human chorionic gonadotropin (β-HCG) at screening, urine on all other occasions (females only).

h) Study drug will be administered in fasted state (after an overnight fast of at least 10 hours).

i) Subjects will be admitted to the unit on Day –2 to start time-matching ECG assessments in the morning of Day –1. Subjects will stay in the unit till Day 14, with the first dose given on Day 1 and last dose given on Day 13.

j) Follicle stimulating hormone (females only to confirm post-menopausal status, performed at screening only).

k) At screening and Day –1 only (drugs of abuse urine test and alcohol test).

l) If the clinical genotyping blood sample is not collected at Day 1, it may be collected at any time during the conduct of the clinical study.

m) Blood for RBR DNA sample will be collected on Day 1 from all patients who signed RBR ICF. If, however, the RBR DNA blood sample is not collected during the scheduled visit, it may be collected at any time during the conduct of the clinical study.

o) Prior to the first dose of study drug, only serious adverse events caused by a protocol-mandated intervention should be reported.

Appendix 1 Schedule of Assessments: MAD Detailed – 1

Visit	Day	Scheduled Time (h)	Vital Signs[a]	ECG-12 lead[b]	Hematology, Blood Chemistry, Coagulation, Urinalysis	Plasma PK Sample[c]	PD for Protein Biomarkers[d]	Whole Blood for RNA[d]	B-Cell Activation Panel[e]	Whole Blood for RBR DNA[f]	Clinical Genotyping[g]	Administration of Study Medication[h]
Treatment Period	Day -1	0			X		X	X	X			
		0.5	X	X								
		1	X	X								
		2	X	X								
		4	X	X								
		12	X	X								
	Day 1	Predose	X	X		X	X	X		X	X	
		0										X
		0.25				X						
		0.5	X	X		X						
		1	X	X		X						
		1.5				X						
		2	X	X		X	X	X				
		3				X						
		4	X	X		X						
		6				X	X	X				
		8				X						
		12	X	X		X	X	X				
		18				X						
	Day 2	24	X			X	X	X				
	Day 3	Predose	X	X		X	X	X	X			
		0										X
		2	X	X		X	X	X				
		4	X	X								
		6	X			X	X	X				
	Day 4	24	X		X	X	X	X				
	Day 5	Predose	X			X	X	X				
		0										X
		2				X	X	X				
		6	X			X	X	X				
	Day 6	24	X			X	X	X				

Appendix 1 Schedule of Assessments: MAD Detailed – 2

Visit	Day	Scheduled Time (h)	Vital Signs[a]	ECG-12 lead[b]	Hematology, Blood Chemistry, Coagulation, Urinalysis	Plasma PK Sample[c]	PD for Protein Biomarkers[d]	Whole Blood for RNA[d]	B-Cell Activation Panel[e]	Whole Blood for RBR DNA[f]	Clinical Genotyping[g]	Administration of Study Medication[h]
Treatment Period	Day 7	Predose	x	x		x	x	x				
		0										x
		2				x	x	x				
		6	x			x	x	x				
	Day 8	24	x		x	x	x	x				
	Day 9	Predose	x			x						
		0										x
		2				x						
		6	x			x						
	Day 10	24	x			x						
	Day 11	Predose	x		x	x						
		0										x
		2				x						
		6	x			x						
	Day 12	24	x			x						
	Day 13	Predose	x	x	x	x	x	x				
		0										x
		0.25				x						
		0.5	x	x		x						
		1	x	x		x						
		1.5				x						
		2	x	x		x	x	x				
		3				x						
		4	x	x		x						
		6				x	x	x				
		8				x						
		12	x	x		x						
		18				x						
	Day 14	24	x		x	x	x	x	x			
Follow Up Visit	Day 20[i]		x	x	x		x	x	x			

Appendix 1 Schedule of Assessments: MAD Detailed

a) Vital signs include blood pressure, pulse rate, respiratory rate and body temperature. Blood pressure, respiratory rate and pulse rate will be obtained after the patient has been in a supine or sitting position for at least 5 minutes. Blood pressure measurement will be performed in triplicate (can be as short as 20 second to 1 minute interval between measurements). Pulse rate and body temperature measurement will be performed as single assessments.

b) 12-lead ECGs will be obtained in triplicate (three consecutive interpretable 12-lead ECGs within a 2–5 minute interval) after the subject has been in a supine position for at least 10 minutes. Automated ECG intervals (PR (PQ), QRS, QT, QTcF) and HR will be captured or calculated, and the changes in T-wave and U-wave morphology will be documented.

c) Plasma PK samples for RO7020531 will be collected: pre-dose and 0.25, 0.5, 1, 1.5, 2, 3, 4, 6, 8, 12, 18 and 24 post dose from Day 1 to Day 2 and from Day 13 to Day 14. On Days 3, 5, 7, 9, and 11 samples for PK will be taken at pre-dose and 2, 6 and 24 hours post dose.

d) PD samples will be collected: pre-dose and 2, 6, 12 and 24 post dose from Day 1 to Day 2. On Days 3, 5, 7 and 13 samples for PD will be taken at pre-dose and 2, 6 and 24 hours post dose.

e) B-cell activation panel may be assessed at selected sites only.

f) Blood for RBR DNA sample will be collected on Day 1 from all patients who signed RBR ICF. If, however, the RBR DNA blood sample is not collected during the scheduled visit, it may be collected at any time during the conduct of the clinical study.

g) If the clinical genotyping blood sample is not collected at Day 1, it may be collected at any time during the conduct of the clinical study.

h) Study drug will be administered in fasted state (after an overnight fast of at least 10 hours).

i) A follow up visit to be completed 7 days (±1) after the last dose of study medication.

Appendix 1 Schedule of Assessments: CHB Patients on NUCs (Part 2 Cohorts 1, 2 & 3) – 1

Visit	Screening	Treatment Period											Study Drug Discontinuation/Early Termination	Follow Up Visit	Follow Up Visit
Day	D-28 to D-2	Day -1	Day 1	Day 2	Day 3	Day 4	Day 7	Day 8	Day 21[a]	Day 22[a]	Day 41[b]	Day 42[b]		Week 9[c]	Week 12[c]
Informed Consent	x														
Eligibility[d]	x	x													
Demography	x														
Medical History[e]	x														
Physical Examination[f]	x	x							x			x	x		x
Vital Signs[g]	x	x	3	x	3	x	3	x	x	x	x	x	x	x	x
ECG-12 lead[h]	x	x	x	x				x				x	x		x
Hematology, Blood Chemistry, Coagulation[i]	x	x		x				x	x		x		x	x	x
Urinalysis[i]	x	x		x		x		x	x		x		x	x	x
Pregnancy Test[j]	x	x										x	x		x
Administration of Study Medication[k]			x		x		x		x		x				
Randomisation			x												
RO PK Sample			7	x[l]	2		2		2		7	x[l]			
NUC PK Sample			2						2		2				
PD for Protein Biomarkers		x	2	x[l]	2	x[l]	2	x[l]	2	x[l]	2	x[l]	x	x	x
Whole Blood for RNA		x	2	x[l]	2	x[l]	2	x[l]	2	x[l]	2	x[l]	x	x	x
Immunophenotyping		x							x			x	x	x	x
B-cell activation panel (at selected sites)[m]		x							x			x	x	x	x
HBV DNA, HBV DNA, viral resistence monitoring	x	x										x	x		x
HBsAg, HBeAg, HBcAg, HBsAg/anti-HBsAg complex	x	x										x	x		x

Appendix 1 Schedule of Assessments: CHB Patients on NUCs (Part 2 Cohorts 1, 2 & 3) – 2

Visit	Screening	Treatment Period											Study Drug Discontinuation/Early Termination	Follow Up Visit	Follow Up Visit
Day	D-28 to D-2	Day -1	Day 1	Day 2	Day 3	Day 4	Day 7	Day 8	Day 21[a]	Day 22[a]	Day 41[b]	Day 42[b]		Week 9[c]	Week 12[c]
anti-HBs, anti-HBe, anti-HBc	x	x										x	x		x
HBV Viral Genotyping		x													
Autoimmune panel, Thyroid function, Alpha-fetoprotein[n]	x														
Viral Serology (HAV, HCV, HDV, HIV)	x														
Follicle Stimulating Hormone[o]	x														
Substance Use[p]	x	x													
Liver Biopsy/Fibroscan[q]	x														
Whole Blood for RBR DNA[r]			x												
Clinical Genotyping[s]			x												
Adverse Events, Previous and Concomitant Treatments	x[t]	x[t]	x	x	x	x	x	x	x	x	x	x	x	x	x

Appendix 1 Schedule of Assessments: CHB Patients on NUCs (Part 2 Cohorts 1, 2 & 3)

a) Visits Day 21/22 can be also conducted on Day 19/20 or Day 23/24.

b) Visits Day 41/42 can be also conducted on Day 39/40.

c) Visits Week 9 and Week 12 have window ±5 days.

d) Eligibility criteria must be confirmed by Day –1.

e) Medical history should include detailed HBV history.

f) Full physical exam, including recording of weight, is required at screening, Day –1, Day 42 and Week 12. Basic and/or symptoms-directed physical examination (at least lungs, cardiovascular, abdomen, extremities, lymph nodes) should be performed on Day 21 (no recording of weight required). At all other visits (or as clinically indicated), limited, symptom-directed physical examinations should be performed at the discretion of the Investigator. Height will only be recorded at screening. BMI will be calculated at screening.

g) Vital signs include blood pressure, pulse rate, respiratory rate and body temperature. Blood pressure, respiratory rate and pulse rate will be obtained after the patient has been in a supine or sitting position for at least 5 minutes. Blood pressure measurement will be performed in triplicate (can be as short as 20 second to 1 minute interval between measurements).

h) 12-lead ECGs will be obtained in triplicate (three consecutive interpretable 12-lead ECGs within a 2–5 minute interval) after the subject has been in a supine position for at least 10 minutes. Automated ECG intervals (PR (PQ), QRS, QT, QTcF) and HR will be captured or calculated, and the changes in T-wave and U-wave morphology will be documented.

i) During treatment period, samples for clinical laboratory tests (hematology, chemistry, coagulation, urinalysis) should be collected in the morning pre-dose (before taking NUC treatment or/and study drug). At all other time-points, these samples should be taken at the most appropriate time.

j) Serum or plasma beta-human chorionic gonadotropin (β-HCG) at screening, urine on all other occasions (females only).

k) Study drug will be administered together with patient's nucleos(t)ide analogue treatment in fasted state.

l) Time-points for PK and/or PD assessments 24 hours after study drug dosing on previous day.

m) B-cell activation panel may be assessed at selected sites only.

n) Alpha-fetoprotein not required if subject was re-screened within a 6 month period.

o) Follicle stimulating hormone (females only to confirm post-menopausal status, performed at screening only).

Appendix 1 Schedule of Assessments: CHB Patients on NUCs (Part 2 Cohorts 1, 2 & 3)

p) At screening and Day –1 only (drugs of abuse urine test and alcohol test).

q) Screening biopsy, Fibroscan® or equivalent unless already documented within 6 months of randomisation.

r) Blood for RBR DNA sample will be collected on Day 1 from all patients who signed RBR ICF. If, however, the RBR DNA blood sample is not collected during the scheduled visit, it may be collected at any time during the conduct of the clinical study.

s) If the clinical genotyping blood sample is not collected at Day 1, it may be collected at any time during the conduct of the clinical study.

n) Prior to the first dose of study drug, only serious adverse events caused by a protocol-mandated intervention should be reported.

Appendix 1 Schedule of Assessments: CHB Patients on NUCs (Part 2 Cohorts 1, 2 & 3) Detailed

Visit	Day	Scheduled Time (h)	Vital Signs[a]	ECG-12 lead[b]	Hematology, Blood Chemistry, Coagulation	Urinalysis	RO PK Sample[c]	NUC PK Sample[d]	PD for Protein Biomarkers	Whole Blood for RNA[e]	Immunophenotyping	B-cell activation panel[f]	HBV DNA, HBV TNA, viral resistence monitoring	HBsAg, HBeAg, HBcAg, HBsAg/anti-HBsAg complex	anti-HBs, anti-HBe, anti-HBc	HBV Viral Genotyping	Whole Blood for RBR DNA[g]	Clinical Genotyping[h]
Screening	Day -28 - Day -2	***	x	x	x	x							x	x	x			
Treatment Period	Day -1	***	x	x	x	x			x	x	x	x	x	x	x	x		
	Day 1	Predose	x				x	x									x	x
		0.25					x											
		1					x											
		2	x				x	x										
		4					x											
		6	x	x			x		x	x								
		8					x		x	x								
	Day 2	24	x	x	x	x	x		x	x								
	Day 3	Predose	x				x		x	x								
		2-4	x				x											
		4-6	x						x	x								
	Day 4	24	x			x			x	x								
	Day 7	Predose	x				x		x	x								
		2-4	x				x											
		4-6	x						x	x								
	Day 8	24	x	x	x	x			x	x								
	Day 21[i]	Predose	x		x	x	x	x	x	x	x	x						
		2-4					x	x										
		4-6							x	x								
	Day 22[i]	24	x						x	x								
	Day 41[j]	Predose	x		x	x	x	x	x	x								
		0.25					x											
		1					x											
		2					x	x										
		4					x											
		6					x		x	x								
		8					x											
	Day 42[j]	24	x	x			x		x	x	x	x	x	x	x			
Follow Up Visit	Week 9[k]		x		x	x			x	x	x	x						
Follow Up Visit	Week 12[k]		x	x	x	x			x	x	x	x	x	x	x			

Appendix 1 Schedule of Assessments: CHB Patients on NUCs (Part 2 Cohorts 1, 2 & 3) Detailed

a) Vital signs include blood pressure, pulse rate, respiratory rate and body temperature (oral or tympanic). Blood pressure, respiratory rate and pulse rate will be obtained after the patient has been in a supine or sitting position for at least 5 minutes.

b) 12-lead ECGs will be obtained in triplicate (three consecutive interpretable 12-lead ECGs within a 2–5 minute interval) after the subject has been in a supine position for at least 10 minutes. Automated ECG intervals (PR (PQ), QRS, QT, QTcF) and HR will be captured or calculated, and the changes in T-wave and U-wave morphology will be documented.

c) Plasma PK samples will be collected pre-dose and 0.25, 1, 2, 4, 6, 8 and 24 hours post-dose on Day 1 of Week 1 and on Day 41. PK samples will be collected pre-dose and 2–4 hours post-dose on Days 3, 7, and 21.

d) Plasma PK samples for NUCs will be collected pre-dose and 2–4 hours post-dose on Day 1, Day 21 and Day 41.

e) During treatment period, blood samples for protein biomarkers and RNA will be collected 6, 8 and 24 hours post-dose on Day 1; pre-dose, 4–6 and 24 hours post-dose on Day 3, Day 7 and Day 21; and pre-dose, 6 and 24 hours post-dose on Day 41.

f) B-cell activation panel may be tested at selected sites only.

g) Blood for RBR DNA sample will be collected on Day 1 from all patients who signed RBR ICF. If, however, the RBR DNA blood sample is not collected during the scheduled visit, it may be collected at any time during the conduct of the clinical study.

h) If the clinical genotyping blood sample is not collected at Day 1, it may be collected at any time during the conduct of the clinical study.

i) Visits Day 21/22 can be also conducted on Day 19/20 or Day 23/24.

j) Visits Day 41/42 can be also conducted on Day 39/40.

k) Visits Week 9 and Week 12 have window ±5 days.

Appendix 1 Schedule of Assessments: CHB Patients Who Are Not on Antiviral Treatment (Part 2 Cohort 4) – 1

Visit	Screening	Treatment Period														Follow Up Visit	Follow Up Visit
Day	D-28 to D-2	Day -1	Day 1	Day 2	Day 3	Day 5	Day 7	Day 8	Day 15[a]	Day 21[b]	Day 22[a]	Day 28[a]	Day 35[a]	Day 41[c]	Day 42[c]	Study Drug Discontinuation / Early Termination	Week 9[d]	Week 12[d]
Informed Consent	X																	
Eligibility[e]	X	X																
Demography	X																	
Medical History[f]	X																	
Physical Examination[g]	X	X								X					X	X		X
Vital Signs[h]	X	X	6	X	4 or 5[t]	4 or 5[t]	3	X	X	X	X	X	X	X	X	X	X	X
ECG-12 lead[i]	X	X	X	X	X	X		X							X	X		X
Hematology, Blood Chemistry, Coagulation[j]	X	X		X		X		X		X				X		X	X	X
Urinalysis[j]	X	X		X		X		X		X				X		X	X	X
Pregnancy Test[k]	X	X													X	X		X
Administration of Study Medication[l]			X		X	X	X		X	X			X	X				
Randomisation			X															
RO PK Sample			7	x[m]	2	2	2			2				7	x[m]			
PD for Protein Biomarkers		X	2	x[m]	2	2	2	x[m]		2	x[m]			2	x[m]	X	X	X
Whole Blood for RNA		X	2	x[m]	2	2	2	x[m]		2	x[m]			2	x[m]	X	X	X
Immunophenotyping		X								X					X	X		X

Appendix 1 Schedule of Assessments:
CHB Patients Who Are Not on Antiviral Treatment (Part 2 Cohort 4) – 2

Visit	Screening	Treatment Period															Follow Up Visit	Follow Up Visit
Day	D-28 to D-2	Day -1	Day 1	Day 2	Day 3	Day 5	Day 7	Day 8	Day 15[a]	Day 21[b]	Day 22[a]	Day 28[a]	Day 35[a]	Day 41[c]	Day 42[c]	Study Drug Discontinuation / Early Termination	Week 9[a]	Week 12[d]
HBV DNA, HBV TNA, viral resistence monitoring[e]	X	X						X	X	X		X	X		X	X		X
HBsAg, HBeAg, HBcAg, HBsAg/anti-HBsAg complex	X	X								X					X	X		X
anti-HBs, anti-HBe, anti-HBc	X	X													X	X		X
HBV Viral Genotyping		X																
Autoimmune panel, Thyroid function, Alpha-fetoprotein[n]	X																	
Viral Serology (HAV, HCV, HDV, HEV, HIV)	X																	
Follicle Stimulating Hormone[o]	X																	
Substance Use[p]	X	X																
Liver Biopsy/Fibroscan[q]	X																	
Whole Blood for RBR DNA[r]			X															
Clinical Genotyping[s]			X															
Adverse Events, Previous and Concomitant Treatments	X[u]	X[u]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Appendix 1 Schedule of Assessments: CHB Patients Who Are Not on Antiviral Treatment (Part 2 Cohort 4)

a) Visits Day 15, 28 and 35 have a window of ±2 days

b) Visits Day 21/22 can be also conducted on Day 19/20 or Day 23/24.

c) Visits Day 41/42 can be also conducted on Day 39/40.

d) Visits Week 9 and Week 12 have a window of ± 5 days.

e) Eligibility criteria must be confirmed by Day –1.

f) Medical history should include detailed HBV history.

g) Full physical exam, including recording of weight, is required at screening, Day –1, Day 42 and Week 12. Basic and/or symptoms-directed physical examination (at least lungs, cardiovascular, abdomen, extremities, lymph nodes) should be performed on Day 21 (no recording of weight required). At all other visits (or as clinically indicated), limited, symptom-directed physical examinations should be performed at the discretion of the Investigator. Height will only be recorded at screening. BMI will be calculated at screening.

h) Vital signs include blood pressure, pulse rate, respiratory rate and body temperature. Blood pressure, respiratory rate and pulse rate will be obtained after the patient has been in a supine or sitting position for at least 5 minutes. Blood pressure measurement will be performed in triplicate (can be as short as 20 second to 1 minute interval between measurements).

i) 12-lead ECGs will be obtained in triplicate (three consecutive interpretable 12-lead ECGs within a 2–5 minute interval) after the subject has been in a supine position for at least 10 minutes. Automated ECG intervals (PR (PQ), QRS, QT, QTcF) and HR will be captured or calculated, and the changes in T-wave and U-wave morphology will be documented.

j) During treatment period, samples for clinical laboratory tests (hematology, chemistry, coagulation, urinalysis) should be collected in the morning pre-dose (before taking study drug). At all other time-points, these samples should be taken at the most appropriate time.

k) Serum or plasma beta-human chorionic gonadotropin (β-HCG) at screening, urine on all other occasions (females only).

l) Study drug will be administered in fasted state. *In case of 100 mg QW dosing, dose administration will be synchronized with the scheduled visit, as there are timed assessments, e.g., if the last QOD dose was interrupted on Day 8 or Day 9, and allowing for the 5 days stipulation, QW dosing would start on Day 15. However, if e.g. the last QOD dose was interrupted on Day 11 and allowing for the 5 days stipulation, QW dosing would start on Day 21, NOT on Day 15.*

Appendix 1 Schedule of Assessments: CHB Patients Who Are Not on Antiviral Treatment (Part 2 Cohort 4)

m) Time-points for PK and/or PD assessments 24 hours after study drug dosing on previous day.

n) Alpha-fetoprotein not required if subject was re-screened within a 6 month period.

o) Follicle stimulating hormone (females only to confirm post-menopausal status, performed at screening only).

p) At screening and Day –1 only (drugs of abuse urine test and alcohol test).

q) Screening biopsy, Fibroscan® or equivalent unless already documented within 6 months of randomisation.

r) Blood for RBR DNA sample will be collected on Day 1 from all patients who signed RBR ICF. If, however, the RBR DNA blood sample is not collected during the scheduled visit, it may be collected at any time during the conduct of the clinical study.

s) If the clinical genotyping blood sample is not collected at Day 1, it may be collected at any time during the conduct of the clinical study.

t) Depending on whether the safety monitoring is for 8 hours or extended to 12 hours, at the Investigator's discretion, in patients who develop flu-like symptoms/pyrexia and do not respond to symptomatic treatment

u) Prior to the first dose of study drug, only serious adverse events caused by a protocol-mandated intervention should be reported.

v) *Pre-dose sample*

Appendix 1 Schedule of Assessments:
CHB Patients Who Are Not on Antiviral Treatment (Part 2 Cohort 4) Detailed

Visit	Day	Scheduled Time (h)	Vital Signs[a]	ECG-12 lead[b]	Hematology, Blood Chemistry, Coagulation	Urinalysis	RO PK Sample[c]	PD for Protein Biomarkers	Whole Blood for RNA[d]	Immuno-phenotyping	HBV DNA, HBV TNA, viral resistence monitoring[m]	HBsAg, HBeAg, HBcAg, HBsAg/ anti-HBsAg complex	anti-HBs, anti-HBe, anti-HBc	HBV Viral Genotyping	Whole Blood for RBR DNA[e]	Clinical Genotyping[f]
Screening	Day -28 - Day -2	***	x	x	x	x					x	x	x			
Treatment Period	Day -1	***	x	x	x[g]	x		x	x	x	x	x	x	x		
	Day 1	Predose	x				x								x	x
		0.25					x									
		1					x									
		2	x				x									
		4	x				x									
		6	x	x			x	x	x							
		8	x				x	x	x							
		12	x													
	Day 2	24	x	x	x	x	x	x	x							
	Day 3	Predose	x				x	x	x							
		2-4	x				x									
		4-6	x					x	x							
		6-8	x	x												
		8-12[h]	x													
	Day 5	Predose	x		x	x	x	x	x							
		2-4	x				x									
		4-6	x					x	x							
		6-8	x	x												
		8-12[h]	x													

Appendix 1 Schedule of Assessments:
CHB Patients Who Are Not on Antiviral Treatment (Part 2 Cohort 4) Detailed

Visit	Day	Scheduled Time (h)	Vital Signs[a]	ECG-12 lead[b]	Hematology, Blood Chemistry, Coagulation	Urinalysis	RO PK Sample[c]	PD for Protein Biomarkers	Whole Blood for RNA[d]	Immuno-phenotyping	HBV DNA, HBV TNA, viral resistence monitoring[m]	HBsAg, HBeAg, HBcAg, HBsAg/ anti-HBsAg complex	anti-HBs, anti-HBe, anti-HBc	HBV Viral Genotyping	Whole Blood for RBR DNA[e]	Clinical Genotyping[f]
Treatment Period	Day 7	Predose	X				X	X	X							
		2-4	X				X									
		4-6	X					X	X							
	Day 8	24	X	X	X	X		X	X		X					
	Day 15[i]		X								X					
	Day 21[j]	Predose	X		X	X	X	X	X	X	X	X				
		2-4					X									
		4-6						X	X							
	Day 22[j]	24	X					X	X							
	Day 28[i]		X								X					
	Day 35[i]		X								X					
	Day 41[k]	Predose	X		X	X	X	X	X							
		0.25					X									
		1					X									
		2					X									
		4					X									
		6					X	X	X							
		8					X									
	Day 42[k]	24	X	X			X	X	X	X	X	X	X			
Follow Up Visit	Week 9[l]		X		X	X		X	X							
Follow Up Visit	Week 12[l]		X	X	X	X		X	X	X	X	X	X			

Appendix 1 Schedule of Assessments: CHB Patients Who Are Not on Antiviral Treatment (Part 2 Cohort 4) Detailed

a) Vital signs include blood pressure, pulse rate, respiratory rate and body temperature (oral or tympanic). Blood pressure, respiratory rate and pulse rate will be obtained after the patient has been in a supine *or sitting* position for at least 5 minutes.

b) 12-lead ECGs will be obtained in triplicate (three consecutive interpretable 12-lead ECGs within a 2–5 minute interval) after the subject has been in a supine position for at least 10 minutes. Automated ECG intervals (PR (PQ), QRS, QT, QTcF) and HR will be captured or calculated, and the changes in T-wave and U-wave morphology will be documented.

c) Plasma PK samples for RO will be collected pre dose and 0.25, 1, 2, 4, 6, 8 and 24 hours post-dose on Day 1 of Week 1 and on Day 41. PK samples will be collected pre-dose and 2-4 hours post-dose on Days 3*, 5*, 7, and 21.

d) During treatment period, blood samples for protein biomarkers and RNA will be collected 6, 8 and 24 hours post-dose on Day 1; pre-dose *and* 4–6 *hours post-dose on Day 3 and Day 5; pre-dose, 4–6 hours* and 24 hours post-dose on Day 7 and Day 21; and pre-dose, 6 and 24 hours post-dose on Day 41.

e) Blood for RBR DNA sample will be collected on Day 1 from all patients who signed RBR ICF. If, however, the RBR DNA blood sample is not collected during the scheduled visit, it may be collected at any time during the conduct of the clinical study.

f) If the clinical genotyping blood sample is not collected at Day 1, it may be collected at any time during the conduct of the clinical study.

g) A duplicate sample will be taken for liver function tests (ALT) and sent to the central laboratory and local laboratory. The local laboratory sample is required to confirm eligibility for liver function parameters before dosing a patient on Day 1. If the patient is deemed to be eligible for the study based on the results of the local test, but the central laboratory test results differ, the patient should continue on the study.

h) Optionally, the safety monitoring after dosing on Day 3 and Day 5 can be extended to 12 hours in patients who develop flu-like symptoms/pyrexia and do not respond to symptomatic treatment at the Investigator's discretion

i) Visits Day 15, 28 and 35 have a window of ±2 days.

j) Visits Day 21/22 can be also conducted on Day 19/20 or Day 23/24.

k) Visits Day 41/42 can be also conducted on Day 39/40.

l) Visits Week 9 and Week 12 have a window of ±5 days.

m) *Pre-dose sample.*

Appendix 2 Formula for Calculation of BMI

Formula for calculation of BMI

$$BMI = \frac{\text{Weight (kg)}}{\text{Height (m)}^2}$$

Unit Conversion: 1 kg = 2.2 lbs

1 inch = 2.54 cm

Example: BMI of a subject being 1.70 m tall and weighing 80 kg:

$$\frac{80 \text{ kg}}{(1.70 \text{ m})^2} = 27.7 \text{ kg/m}^2$$

The subject's standing height will be measured in bare feet standing with his/her heels and back in contact with the vertical bar of a wall mounted measuring device. The head is held so the subject looks straight forward. A level will be placed on the subject's head to ensure that the subject is looking straight forward. The point at which the lower surface of the level intersects with the vertical measuring device will be the standing height.

Appendix 3 Child-Pugh Classification of Severity of Liver Disease

Clinical and Biochemical Measurements	Points Scored for Increasing Abnormality		
	1	2	3
Encephalopathy (grade)*	None	1 and 2	3 and 4
Ascites	Absent	Slight	Moderate
Bilirubin (mg. per 100 mL)	1 – 2	2 - 3	> 3
Albumin (g. per 100 mL)	3.5	2.8 - 3.5	< 2.8
Prothrombin time (sec. prolonged)	1 – 4	4 - 6	> 6

* According to grading of Trey, Burns and Saunders (1966)

1, 2 or 3 points are scored for increasing abnormality of each of the 5 parameters measured.

Grade A: 5 or 6

Grade B: 7 to 9

Grade C: 10 to 15

Appendix 4 Correction Formulas for QTc Intervals

Fridericia's correction for QTc Measurement - QTcF

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

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

QT (msec) = 386

RR (msec) = 848

$$\frac{QT\ (msec)}{\sqrt[3]{RR\ (msec)/1000}} = 408\ msec$$

Appendix 5 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:

$$\text{Males (ml/min)} = \frac{(140 - \text{Age}) * \text{ABW (kg)}}{72 * \text{Serum Creatinine (mg/dL)}}$$

Females (ml/min) = Male value x 0.85

Conversion Factor for Creatinine Clearance:

SI Units (mL/sec) = Conventional units (mL/min) × 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 × 88.4

Appendix 6 Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events

Electronic version is located on the DAIDS Regulatory Support Center website:

https://rsc.niaid.nih.gov/sites/default/files/daidsgradingcorrectedv21.pdf

Appendix 7 Alcohol Volume Calculation

This appendix is to facilitate the assessment of exclusion criterion for Part 1 (HVs) in Section 4.2.3.1 and Part 2 (patients) in Section 4.2.3.2. It shows a calculation for the study entry assessment on whether a subject drinks more than two standard drinks per day.

A standard drink is any drink containing 10 grams of alcohol. One standard drink always contains the same amount of alcohol regardless of container size or alcohol type, that is beer, wine, or spirit.

The formula for calculating standard drinks:

Number of standard drinks = Volume (liter) x Vol% of alcoholic beverage x 0.789

For example, one bottle of 375 mL of full strength beer 5% alcohol by volume equals to 1.5 standard drinks:

$0.375 \times 5 \times 0.789 = 1.5$ standard drinks