RANDOMIZED DOUBLE-BLIND STUDY OF NITAZOXANIDE COMPARED TO PLACEBO IN SUBJECTS WITH HBEAG-NEGATIVE CHRONIC HEPATITIS B VIROLOGICALLY SUPPRESSED FOR AT LEAST TWELVE MONTHS ON TENOFOVIR DISOPROXIL FUMARATE, TENOFOVIR ALAFENAMIDE OR ENTECAVIR

PROTOCOL NO. RM08-2001

Study Sponsor:

The Romark Institute for Medical Research

[redacted]

Current Version:

v4.0 Incorporating Amendments 1, 2, and 3

Replaces:

v1.0 (Original)

v2.0 Amendment 1, Dated 10 April 2019

v3.0 Amendment 2, Dated 13 June 2019

Effective Date: January 2, 2020

This study will be conducted in compliance with this protocol, current International Council for Harmonisation Good Clinical Practice (ICH GCP), United States (US) Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46, 21 CFR 50, 21 CFR Part 312), and applicable regulatory requirements.

INVESTIGATOR'S AGREEMENT

CONFIDENTIALITY

The information in this protocol is provided to you, as an Investigator or consultant, for review by you, your staff, and an applicable institutional review committee. By accepting this document, you agree that information contained herein will be considered confidential and will not be disclosed to others, without written authority from The Romark Institute for Medical Research, except to the extent necessary to obtain: (a) Institutional Review Board approval and (b) informed consent from those persons to whom the investigational medicinal product will be administered.

APPROVAL OF FINAL PROTOCOL

My signature below constitutes agreement with this protocol. I am providing the necessary assurances that this study will be conducted by me and my staff according to all stipulations of the protocol, including all statements regarding confidentiality, and in complete accordance with all applicable regulations including current Good Clinical Practice guidelines and the ethical guidelines set by the World Medical Assembly (Declaration of Helsinki, last amendment in Fortaleza, Brazil October 2013). Furthermore, my signature below indicates that source documents will be available for review by the Sponsor or their designated representative.

Principal Investigator Signature:

Principal Investigator Date

Print name: _____

With the signature below, the Sponsor approves of this protocol.

Sponsor Signature:

The Romark Institute for Medical Research Date

Print name: _____

1. SYNOPSIS

Title: Randomized Double-Blind Study of Nitazoxanide Compared to Placebo in Subjects With HBeAg-Negative Chronic Hepatitis B Virologically Suppressed for at Least Twelve Months on Tenofovir Disoproxil Fumarate, Tenofovir Alafenamide or Entecavir

Study Number: RM08-2001

IND Number: 143,329

Indication: Treatment of hepatitis B e antigen (HBeAg) negative chronic hepatitis B (CHB).

Design: Randomized double-blind placebo-controlled dose-range-finding trial.

Number of Subjects: 48.

Population: Adults (at least 21 years of age) with HBeAg negative CHB virologically suppressed for at least 12 months on tenofovir disoproxil fumarate (TDF), tenofovir alafenamide (TAF) or entecavir (ETV).

Randomization: 1:1:1:1 (12 subjects per group).

Study Dose and Administration: Group 1: Placebo for 12 weeks.

Group 2: 600 mg nitazoxanide (NTZ) once daily for 12 weeks.

Group 3: 600 mg NTZ twice daily for 12 weeks.

Group 4: 900 mg NTZ twice daily for 12 weeks.

All subjects will continue TDF, TAF or ETV therapy throughout the study.

NTZ 300 mg extended release tablets and matching placebo tablets will be administered in double-blind fashion orally with food.

After 12 weeks, subjects will continue treatment with the same treatment regimen if they have achieved any decline in quantitative hepatitis B surface antigen (HBsAg) from Baseline to Week 12. Stopping rules are provided based upon mean change in quantitative (qHBsAg) response.

Objectives: To evaluate safety, effectiveness, and pharmacokinetic-pharmacodynamic (PK/PD) relationships associated with three different NTZ treatment regimens added to TDF, TAF or ETV in treating CHB.

Primary Efficacy Parameter: Mean change in HBsAg from Baseline to Week 12.

Secondary Efficacy Parameters:

i. Sustained HBsAg loss with hepatitis B virus (HBV) deoxyribonucleic acid (DNA) suppression 24 weeks after end of treatment with NTZ and TDF, TAF or ETV.

ii Change in qHBsAg from Baseline to different time points on treatment.

iii. HBsAg loss.

iv. HBsAg seroconversion.

v. HBV DNA suppression.

vi. Change from Baseline in Fibrosis 4 (FIB-4) score.

vii. Change from Baseline in Fibroscan® score.

Safety Parameters: Adverse events, laboratory safety tests

Biological Samples: Blood and urine samples collected at Screening, Baseline (Day 1), Day 3, Weeks 1, 2, 4, 8, 12, 16, 20, 24 and every 12 weeks thereafter during treatment and at off-treatment follow-up Weeks 4, 8, 12, 24 and 48.

Study Center: National University Hospital of Singapore

Screening Period: August 2019 through April 2020

2. STUDY SCHEMA

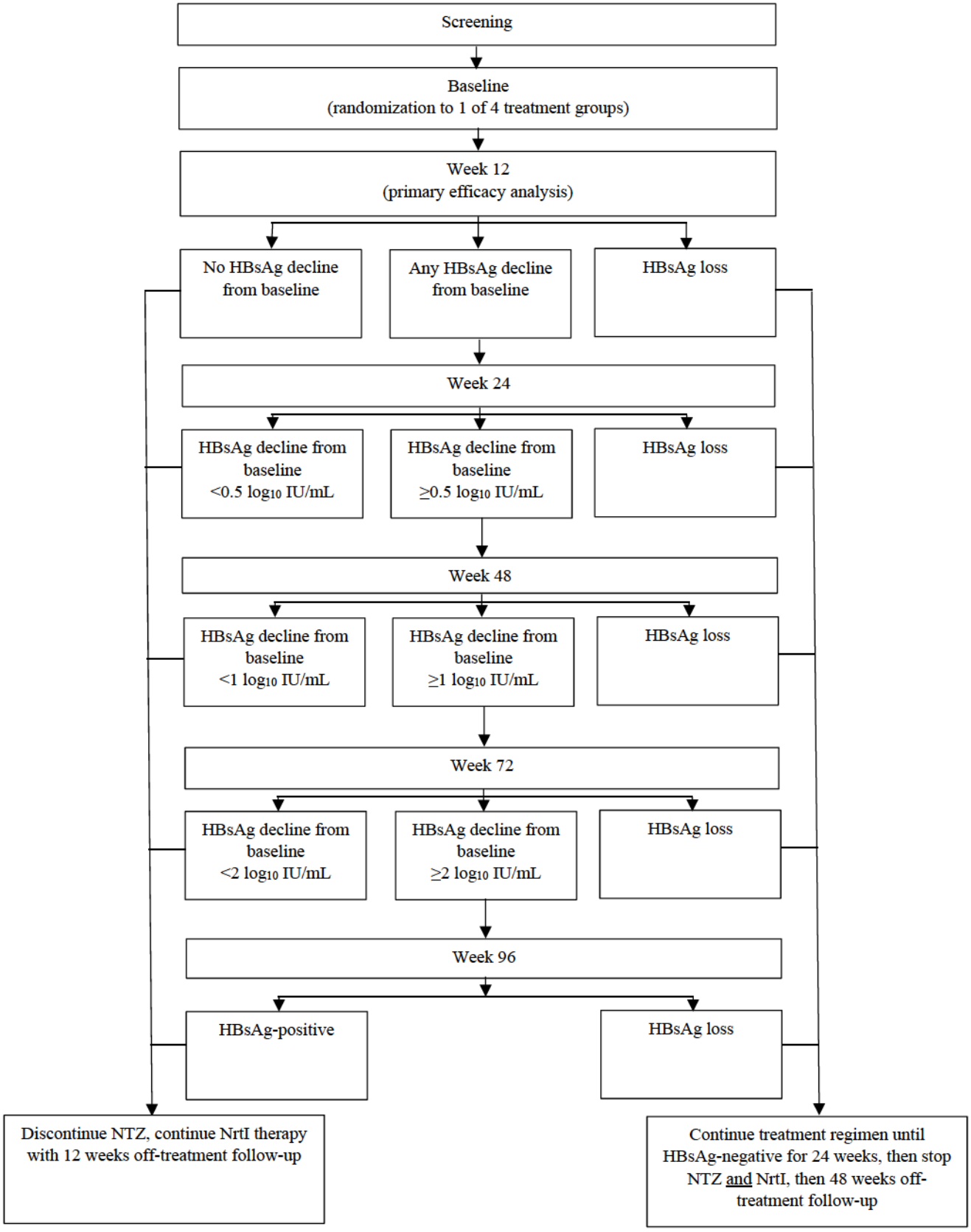

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Table 1: Abbreviations

Abbreviation	Term
AFP	alpha-fetoprotein
ALT	alanine aminotransferase
AST	aspartate aminotransferase
BID	twice daily
BUN	blood urea nitrogen
cccDNA	covalently closed circular viral DNA
CHB	chronic hepatitis B
CPK	creatine phosphokinase
CRF	case report form
CTP	Child-Turcotte-Pugh
DNA	deoxyribonucleic acid
EDC	electronic data collection
ETV	entecavir
GCP	Good Clinical Practice
GGT	gamma glutamyl transferase
HBcAg	HBV core antigen
HBcrAg	HBV core-related antigen
HBeAg	Hepatitis B e antigen
HBsAg	Hepatitis B surface antigen
HBV	Hepatitis B virus
HCC	hepatocellular carcinoma
HCV	Hepatitis C Virus
HDV	Hepatitis D virus
HIV	human immunodeficiency virus
ICF	informed consent form
ICH	International Council for Harmonisation
IgM	Immunoglobulin M
INR	international normalized ratio
IRB	institutional review board
IUD	intrauterine device

Abbreviation	Term
NTZ	nitazoxanide
PT	prothrombin time
QD	once daily
qHBsAg	quantitative HBsAg
TAF	tenofovir alafenamide
TDF	tenofovir disoproxil fumarate

4. INTRODUCTION

4.1. Chronic Hepatitis B

Hepatitis B virus (HBV) is a partially double-stranded deoxyribonucleic acid (DNA) virus belonging to the Hepadnaviridae family of viruses.

Chronic hepatitis B (CHB) infection remains a major public health problem, affecting more than 250 million people worldwide. These patients have a higher risk of developing severe liver diseases such as cirrhosis, liver failure, or hepatocellular carcinoma (World Health Organization, 2018).

Currently available therapies for CHB include nucleoside/nucleotide reverse transcriptase inhibitors (NrtIs) and pegylated interferon. These therapies do not eliminate the covalently closed circular viral DNA (cccDNA), which functions as a nonreplicative minichromosome and persists throughout the lifespan of infected hepatocytes, and therefore, the goals of treatment are sustained suppression of HBV DNA and remission of liver disease. While six NrtIs have been approved by the U.S. FDA for treatment of CHB (tenofovir disoproxil fumarate (TDF), tenofovir alafenamide (TAF), entecavir (ETV), lamivudine, adefovir and telbivudine), TDF, TAF and ETV are the preferred NrtIs for treatment (Lok et al, 2016; Terrault et al, 2018).

Response rates for patients with hepatitis B e antigen (HBeAg) negative CHB enrolled in randomized clinical trials of TDF, TAF or ETV are as follows (Terrault et al, 2018):

HBV DNA suppression (less than 60 IU/mL)	90 to 93%.
alanine aminotransferase (ALT) normalization	76 to 88%.
Loss of hepatitis B surface antigen (HBsAg)	less than 1%.

Long-term administration of NrtIs is standard of care for HBeAg-negative CHB because it reduces liver-related complications by suppressing viral replication. The best and safest stopping rule is HBV DNA suppression with loss of HBsAg. Loss of HBsAg is associated with reduced risk of hepatic decompensation and improved survival (Terrault et al, 2016; Sarin et al, 2016; European Association for the Study of the Liver, 2017). In this context, a treatment for CHB that could induce loss of HBsAg leading to functional cure is needed.

4.2. Nitazoxanide

Nitazoxanide (NTZ) is a thiazolide anti-infective with *in vitro* activity against parasites, anaerobic bacteria, and viruses (Anderson and Curran, 2007).

Alinia® NTZ for Oral Suspension (patients 1 year of age and older) and Alinia NTZ Tablets (patients 12 years and older) are marketed in the United States for the treatment of diarrhea caused by *Giardia lamblia* or *Cryptosporidium parvum*. Alinia for Oral Suspension and Alinia Tablets have not been shown to be superior to placebo for the treatment of diarrhea caused by *Cryptosporidium parvum* in human immunodeficiency virus (HIV)-infected or immunodeficient patients (Romark, 2017).

4.2.1. Antiviral Activity of NTZ in Cell Cultures

Tizoxanide, the active metabolite of NTZ, has been shown to possess antiviral activity in cell culture against a broad range of viruses including influenza A and B viruses, parainfluenza

(Sendai) virus, respiratory syncytial virus (RSV) A-2, canine coronavirus S-378, rotavirus, norovirus, hepatitis C virus genotypes 1a and 1b, Japanese encephalitis virus, dengue fever virus-2, yellow fever virus, hepatitis B virus, and HIV. The wide spectrum of antiviral activity suggests a cell-mediated effect rather than a specific viral target (Rossignol, 2014).

In cell culture, tizoxanide suppresses HBV DNA in a dose-dependent manner (EC50 = 0.15 µM) and acts synergistically with lamivudine, adefovir dipivoxil, TDF and ETV. Unlike currently available therapies, tizoxanide also suppresses HBeAg and HBsAg in a dose-dependent manner at concentrations similar to those required to suppress HBV DNA (Korba et al., 2008; Romark data on file).

Recent studies have shown that NTZ inhibits HBV transcription from cccDNA by interfering with the interaction between HBV regulatory protein x (HBx) and the host protein, damage-specific-DNA-binding protein 1 (DDB1) (Sekiba et al., 2019).

4.2.2. Clinical Experience with NTZ

4.2.2.1. Overall Clinical Experience

NTZ has been marketed for diarrheal disease caused by *Giardia* or *Cryptosporidium* in the United States since 2003. In Latin America, India, Bangladesh and Egypt the drug is marketed as a broad-spectrum antiparasitic agent. It is estimated that more than 300 million patients have been exposed to NTZ worldwide with the usual adult dose being 500 mg twice daily for three days. No significant drug-related AEs have been reported in post-marketing experience.

Over 6,000 subjects have been treated with NTZ in clinical trials, including 1,760 subjects 12 years of age and older who have been exposed to the NTZ 300 mg extended release tablets (as monotherapy, n = 1,272, or in combination with oseltamivir, n = 488). Of the 1,760 subjects, 1,374 were given a dose of 600 mg twice daily for 5 days. The most commonly (at least 2% of treated subjects) reported adverse events (AEs) regardless of causality assessment were: diarrhea (6.4%), chromaturia (4.7%), headache (3.5%), nausea (3.1%), abdominal pain/abdominal pain upper (3%), bronchitis (2.7%), and vomiting (2.3%). In the placebo-controlled trials, the rates of occurrence of AEs did not differ significantly from those of placebo except for chromaturia (4.7% vs 0.2%). Less than 1% of subjects discontinued therapy because of AEs.

Other clinical studies of NTZ immediate-release tablets, oral suspension, or other formulations have been conducted in more than 4,250 subjects. The dose was typically 500 mg to 1500 mg twice daily for adults and adolescents at least 12 years of age or 7.5 mg/kg/day for children under 12 years of age, and the duration of treatment was typically 3 to 14 days.

The product has been studied for longer durations in subjects with acquired immune deficiency syndrome (AIDS) and cryptosporidiosis, and in subjects with chronic hepatitis C. A total of 660 subjects enrolled in these long-term studies received NTZ for more than 1 month (maximum of 1,528 days), of which 411 were treated for greater than 3 months, mostly at a dose range of 500 mg to 1350 mg twice daily. In 315 subjects with chronic hepatitis C, NTZ was administered at doses ranging from 500 mg to 1350 mg twice daily with food for up to 52 weeks. The AEs associated with long-term use of NTZ were primarily related to the gastrointestinal (GI) tract with mild to moderate and transient abdominal pain, diarrhea, headache and nausea being the most common AEs reported. Mild and transient yellow discoloration of the urine and sclera

have also been reported, particularly in subjects receiving doses of at least 1,000 mg BID for extended durations.

4.2.2.2. Clinical Experience in CHB

Rossignol and Brechot described the results of an open label, pilot clinical trial in Egypt in 9 subjects with chronic CHB without cirrhosis. All were HBsAg positive and two were HBeAg positive. On biopsy, inflammatory grading ranged from 2 to 6/18 and fibrosis staging ranged from 1 to 3/6. NTZ 500 mg immediate release tablets were administered orally twice daily with food for up to 48 weeks. NTZ was well tolerated with occasional mild to moderate side effects primarily related to the gastrointestinal tract, including diarrhea and epigastric pain. These adverse events were transient and resolved during therapy; none required discontinuation of treatment. There were no laboratory abnormalities that were deemed to be related to treatment. HBV DNA became undetectable (less than 38 IU/mL) in the serum of 8 of the 9 subjects (89%) after 4 to 20 weeks of NTZ treatment. The two subjects who were HBeAg positive became HBeAg negative after 4 and 16 weeks of treatment, respectively. Three of the 9 subjects (33%) became HBsAg negative; two of them after 8 weeks and one after 48 weeks of treatment. The subject with the later responses (20 weeks to undetectable HBV DNA, 16 weeks to HBeAg negative, and 48 weeks to HBsAg negative) was treated with 500 mg NTZ only once daily (Rossignol and Brechot, 2019).

A brief report of the nine cases described above along with three other case reports was reviewed by Rossignol and Keeffe in 2008. Two of these additional three cases were HBeAg-positive patients in Egypt, and each received NTZ 500 mg immediate release tablets orally twice daily. Both became HBeAg negative and experienced declines in serum HBV DNA with one of the two clearing serum HBV DNA. The third case was a 60 year-old male in the United States with HBeAg positive chronic hepatitis B who had received lamivudine 100 mg daily for two years and then adefovir 10 mg daily for one year without response. NTZ 500 mg twice daily was added to continued adefovir for a total of two years of combination therapy. This patient developed normal ALT, undetectable serum HBV DNA, negative HBeAg and negative HBsAg and had documented seroconversion to antibody to HBsAg (anti-HBs). No safety data were reported for these three case reports (Rossignol and Keeffe, 2008). Kolozi et al, also reported two of these cases in an abstract, but no safety data was provided (Kolozi et al., 2008).

4.3. Rationale for the Study

There is a need for a new treatment of CHB that provides a functional cure with suppression of HBV DNA and loss of HBsAg. NTZ inhibits replication of HBV *in vitro* by means of a novel mechanism. It is unique in that it inhibits HBV transcription from cccDNA and suppresses HBV DNA while also suppressing HBeAg and HBsAg. This study will evaluate the effect of different doses of NTZ on quantitative HBsAg in an effort to identify a dosage regimen for further development with the objective of inducing loss of HBsAg.

5. STUDY OBJECTIVES

This randomized controlled trial is designed to evaluate safety, effectiveness and pharmacokinetic-pharmacodynamic (PK/PD) relationships associated with three different NTZ treatment regimens added to TDF, TAF or ETV in treating CHB.

6. STUDY DESIGN

The study will be a randomized, double-blind, placebo-controlled design. Forty-eight subjects will be selected according to the inclusion and exclusion criteria listed below. Upon enrollment in the study, subjects will be randomized to one of the following treatment groups (12 subjects per group):

- Group 1: PLACEBO

Subjects will take three placebo tablets twice daily.

- Group 2: 600 mg NTZ once daily (QD).

Subjects will take two NTZ 300 mg extended release tablets and one placebo tablet in the morning and three placebo tablets in the evening.

- Group 3: 600 mg NTZ twice daily (BID).

Subjects will take two NTZ 300 mg extended release tablets and one placebo tablet twice daily.

- Group 4: 900 mg NTZ BID.

Subjects will take three NTZ 300 mg extended release tablets twice daily.

NTZ and placebo tablets will be administered orally with food. All subjects will continue TDF, TAF or ETV therapy throughout the study.

After 12 weeks, subjects will continue treatment with the same treatment regimen if they have achieved any decline in quantitative HBsAg (qHBsAg) from Baseline to Week 12. The subjects will discontinue treatment with the investigational medication (NTZ and/or placebo) due to:

- Adverse events
- Withdrawn consent
- Noncompliance
- The subject fails to achieve any decline in qHBsAg from Baseline to Week 12, at least a 0.5 $\log_{10}$ decline from Baseline to Week 24, at least a 1 $\log_{10}$ decline from Baseline to Week 48, at least a 2 $\log_{10}$ decline from Baseline to Week 72 or HBsAg loss at Week 96.
- The subject experiences HBsAg loss with HBV DNA suppression maintained on treatment for 24 weeks (consolidating therapy). TDF, TAF or ETV therapy will also be discontinued.

Subjects will return to the clinic at Day 3, Weeks 1, 2, 4, 8, 12, 16, 20, 24 and every 12 weeks thereafter during the treatment period. Blood samples will be collected at each visit for laboratory and/or serology tests. Blood samples for trough (just prior to dosing) plasma levels of NrtI and tizoxanide will be collected at Baseline and each scheduled visit while on therapy.

Subjects will return to the clinic for off-treatment follow-up at 4, 8 and 12 weeks following the end of treatment with investigational medication (NTZ and/or placebo). In the event that a subject has sustained HBsAg loss with HBV DNA suppression at the Week 12 follow-up visit, that subject will return for additional follow-up visits 24 and 48 weeks after the end of treatment.

The study will be unblinded to the Sponsor (Romark) and Medical Monitor after all subjects have completed 12 weeks of treatment and complete data is collected through that time point for

analysis of safety and the primary efficacy parameter (mean change in qHBsAg at Week 12). The investigators, subjects and clinical monitors will remain blinded throughout the study.

Rationale for study design: *In vitro* data suggest that NTZ may be effective in inducing loss of HBsAg in patients with CHB. Cell culture studies have shown that combinations of NTZ with the NrtIs, lamivudine, adefovir dipovoxil, TDF and ETV are synergistic in inhibiting HBV replication. The NrtIs are poorly effective in producing HBsAg loss or seroconversion, and therefore, patients must remain on chronic therapy in order to maintain suppression of HBV DNA. With this background, this study is designed to evaluate whether the combination of NTZ plus TDF, TAF or ETV is superior to TDF, TAF or ETV alone in reducing quantitative serum HBsAg. TDF, TAF or ETV were selected for use in this study because they are widely used for treating CHB and because of their potency in reducing serum HBV DNA and superior resistance profiles. This study is a Phase 2 study, and therefore, the number of subjects per treatment arm is low. This study is designed to provide data, including dose-range-finding data, to determine whether NTZ reduces qHBsAg and to guide future development of NTZ in treating CHB infection.

Rationale for dose and duration of treatment: The dose of TDF, TAF or ETV used in this study is the standard dose approved by the FDA and other regulatory authorities. The doses of NTZ were selected based upon safety, tolerability and pharmacokinetics data from prior clinical experience with nitazoxanide. Extended release tablets administered twice daily will be used to achieve steady state plasma concentrations of tizoxanide above the concentrations required to inhibit HBsAg in cell cultures. While it is expected that this approach will be optimal, a 600 mg once daily regimen will also be used to evaluate effectiveness. Two different doses (600 mg and 900 mg) will be used in twice daily regimens. These doses are lower than doses that have been used without safety issues in clinical trials involving long-term (at least 48 weeks) treatment of subjects with chronic hepatitis C or AIDS-related cryptosporidiosis. Doses of 1,000 to 1,350 mg NTZ twice daily have been associated with gastrointestinal side effects (e.g., epigastric pain, diarrhea) that make those regimens difficult to administer for long durations, and therefore, a maximum NTZ dose of 900 mg twice daily has been selected.

The duration of treatment to the primary endpoint (12 weeks) is designed to provide a relatively quick determination regarding whether NTZ induces a meaningful reduction of qHBsAg. Subjects will be allowed to continue treatment beyond 12 weeks if they are showing progressive declines in qHBsAg or if they experience HBsAg loss. The stopping rules require discontinuation of the investigational medication in the event that a subject fails to achieve any decline in qHBsAg from Baseline to Week 12, at least a 0.5 log10 decline from Baseline to Week 24, at least a 1 $\log_{10}$ decline from Baseline to Week 48, at least a 2 $\log_{10}$ decline from Baseline to Week 72 or HBsAg loss at Week 96. The generally accepted stopping rule for peginterferon is less than a 1 $\log_{10}$ decline of HBsAg at Week 12. The stopping rules for this study were selected because (1) there is insufficient information regarding the potential qHBsAg response patterns to NTZ and (2) use of NTZ for durations longer than 12 weeks has not been associated with significant safety issues. Subjects achieving HBsAg loss and HBV DNA suppression will continue treatment with the investigational medication and NrtI until they have maintained HBsAg loss and HBV DNA suppression for 24 weeks on treatment (consolidating therapy). The extended duration of treatment for subjects showing response to treatment is designed to allow subjects to continue treatment if they are experiencing benefit with respect to

qHBsAg. It also allows for collection of data regarding the duration of treatment for future clinical development.

7. SUBJECT SELECTION

The criteria for inclusion and exclusion are defined below:

7.1. Inclusion Criteria

1. Age at least 21 years.
2. CHB virus infection (serum HBsAg-positive for at least 6 months or serum HBsAg positive and negative immunoglobulin M (IgM) antibodies to HBV core antigen (IgM anti-HBc).
3. HBeAg negative.
4. Virologically suppressed (HBV DNA less than the lower limit of quantitation) for at least 12 months on TDF, TAF or ETV therapy.
5. Quantitative HBsAg greater than 100 IU/mL.
6. ALT below 1.5 times the upper limit of normal.
7. Able to comply with the study requirements.

7.2. Exclusion Criteria

1. Unable to take oral medications.
2. Females who are pregnant, breast-feeding or not using birth control. A double barrier method, oral birth control pills administered for at least 2 monthly cycles prior to study drug administration, an intrauterine device (IUD), or medroxyprogesterone acetate administered intramuscularly for a minimum of one month prior to study drug administration are acceptable methods of birth control for inclusion into the study. In addition, female subjects should have a Baseline pregnancy test and should agree to continue an acceptable method of birth control for the duration of the study (including follow-up) if sexually active.
3. Any investigational drug therapy within 30 days prior to enrollment.
4. Other causes of liver disease.
5. Co-infection with HIV, hepatitis C virus (HCV) or hepatitis D virus (HDV) based on an enzyme immunoassay (EIA).
6. History of alcoholism or with an alcohol consumption of greater than 40 g per day.
7. Clinically unstable.
8. Any concomitant condition that, in the opinion of the investigator would preclude evaluation of response or make it unlikely that the contemplated course of therapy and follow-up could be completed.

9. History of hypersensitivity or intolerance to NTZ or any of the excipients comprising the NTZ tablets.
10. Hepatocellular carcinoma.
11. Decompensated liver disease including history of ascites, bleeding esophageal varices, portal hypertension or hepatic encephalopathy.
12. FibroScan® score greater than 11 or history of cirrhosis on liver biopsy.
13. Creatinine clearance <65 ml/minute (by the Cockcroft-Gault equation using ideal body weight).
14. History of clinically relevant psychiatric disease, seizures, central nervous system dysfunction, severe pre-existing cardiac, renal, pathologic bone fracture or other risk factors for osteoporosis, hematological disease or medical illness that in the investigator's opinion might interfere with therapy.
15. Malignant disease within 3 years of trial entry.
16. Rheumatological conditions, inflammatory bowel disease or psoriasis requiring or anticipated to require biological/immunosuppressive therapies.
17. Subjects taking or anticipated to need medications considered to be major CYP2C8 substrates (see Section 23.1).

8. RANDOMIZATION

An independent third party will prepare a master randomization list and maintain the masking of the study. Subjects who qualify for the study will be assigned to treatment using centralized randomization procedures stratified by qHBsAg (less than 3, 3 to 4, greater than 4 $\log_{10}$ IU/mL) and NrtI therapy (TDF, TAF or ETV). The treatment numbers will appear on the medication kits containing the masked study medication. The randomization list will be masked to study participants including Sponsor, Investigators, study monitors, subjects and laboratory personnel. Unmasking for any individual subject will occur only if necessitated by emergency and knowledge of the medication being taken will influence the medical management of the subject. Furthermore, if the event warrants submission of a safety report to regulatory authorities, then the subject's treatment assignment will be unmasked for completion of the report. If a subject is lost to follow up or discontinues the study prior to the assessment at week 12, an additional subject may be enrolled in the same treatment group.

9. STUDY PROCEDURES

The schedule of study procedures for each visit are summarized in Section 23.2 and are described below.

9.1. Pre-study Evaluation (Day -31 to 1)

Before screening, subjects will be informed of the nature of the study, and written consent must be obtained prior to participation. After obtaining informed consent, the following procedures will be carried out:

1. Complete medical history
2. Physical examination, including height, body weight and vital statistics
3. Urine pregnancy test for all females of child-bearing potential
4. Review concomitant medications
5. Collection of blood sample for serological tests for qHBV DNA, qHBsAg, qAnti-HBs, HBeAg, anti-HBe, IgM anti-HBc, anti-HIV, anti-HCV and anti-HDV.
6. Collection of a blood sample for laboratory tests including hemoglobin, hematocrit, complete blood count (total and differential), platelet count, prothrombin time (PT), international normalized ratio (INR), random/fasted blood sugar, total cholesterol, high-density lipoprotein, triglycerides, albumin, iron, ferritin, aspartate aminotransferase (AST), ALT, gamma glutamyl transferase (GGT), creatine phosphokinase (CPK), alkaline phosphatase, bilirubin (total/direct), blood urea nitrogen (BUN), creatinine, sodium, potassium, chloride, and alpha-fetoprotein (AFP).
7. Collection of urine sample for routine urinalysis (appearance, glucose, protein and blood).
8. Calculation of creatinine clearance by the Cockcroft-Gault equation using ideal body weight.
9. FibroScan® test.
10. Ultrasonography for hepatocellular carcinoma (HCC) screening.
11. Confirmation that the subject satisfies all inclusion/exclusion criteria.

9.2. Baseline (Day 1)

At Baseline, the following procedures will be carried out:

1. Physical examination, including body weight and vital statistics.
2. Urine pregnancy test for all females of child-bearing potential.
3. Review concomitant medications.
4. Collection of a blood sample for qHBV DNA, qHBsAg, qAnti-HBs, HBeAg, anti-HBe and anti-HIV (anti-HIV is only for subjects at high risk of contracting HIV according to the discretion of the investigator).
5. Collection of a blood sample to be stored for exploratory analyses of HBV RNA, HBV core-related antigen (HBcrAg), qAnti-HBc, quantitative cccDNA, HBs fragments, HBsAg-anti-HBs immune complex, immunological studies and/or host DNA (to assess relationships with virologic response).
6. Collection of a blood sample for laboratory tests including hemoglobin, hematocrit, complete blood count (total and differential), platelet count, PT, INR, albumin, AST, ALT, GGT, CPK, alkaline phosphatase, bilirubin (total/direct), BUN, creatinine, sodium, potassium, chloride, and AFP.

7. Collection of blood samples for trough (just prior to dosing) plasma levels of NrtI and tizoxanide.
8. Collection of a urine sample for routine urinalysis (appearance, glucose, protein and blood).
9. Obtain a 12-lead electrocardiogram (ECG).
10. Instruct the subject regarding:
    a. Administration of study medication.
    b. Use of birth control. Female subjects of childbearing potential, if sexually active, will be instructed to continue an acceptable method of birth control for the duration of the study and for one month post-treatment. Acceptable methods of birth control include a double barrier method, oral birth control pills, an IUD, or medroxyprogesterone acetate administered intramuscularly.
    c. Study visits. Subjects will be provided instructions for returning to the clinic for study visits.
    d. Seeking emergency care or contacting the study physician or nurse. Subjects must be instructed to seek emergency medical care or contact the study physician or nurse if he/she develops any of the following symptoms during the study (treatment period or off-treatment follow-up period).

        - CALL 911 (United States) or LOCAL EMERGENCY SERVICES (outside the United States) OR HOSPITAL OR CONTACT STUDY PHYSICIAN IF YOU HAVE:
            - Trouble breathing including shortness of breath.
            - Severe pain in your chest, shoulder or belly.
        - CALL STUDY NURSE OR PHYSICIAN IF YOU EXPERIENCE:
            - Fever that does not go away or gets higher.
            - Feeling very weak or tired.
            - Unusual (not normal) muscle pain.
            - Trouble breathing.
            - Stomach pain or nausea and vomiting.
            - Feeling cold, especially in your arms and legs.
            - Feeling dizzy or light-headed.
            - Fast or irregular heartbeat.
            - Yellowing of eyes or sclera.
            - Brown or dark urine.
            - Confusion.
            - Tightness in your throat or chest, rash (especially hives) or itchiness.

- Loss of appetite for several days or longer.
- Bowel movements (stools) turn light in color.

11. Randomization (see Section 8).

    At the Baseline visit, subjects will be randomly assigned to one of the following treatment groups:

    Group 1: PLACEBO - Subjects will receive three placebo tablets twice daily with food in addition to continuing TDF, TAF or ETV therapy.

    Group 2: 600 mg NTZ QD - Subjects will receive two NTZ 300 mg extended release tablets and one placebo tablet in the morning and three placebo tablets in the evening with food in addition to continuing TDF, TAF or ETV therapy.

    Group 3: 600 mg NTZ BID - Subjects will receive two NTZ 300 mg extended release tablets and one placebo tablet twice daily with food in addition to continuing TDF, TAF or ETV therapy.

    Group 4: 900 mg NTZ BID - Subjects will receive three NTZ 300 mg extended release tablets twice daily with food in addition to continuing TDF, TAF or ETV therapy.

    Subjects will be given a 4-week supply of medication and instructions on administration of the study medication. The NTZ and placebo tablets (yellow) must be administered with food. TDF, TAF or ETV tablets should be administered daily at the same time each day. The first dose of study medication should be taken in the clinic under the observation of the investigator.

9.3. Evaluations During Treatment

Subjects will return to the clinic for evaluation at Day 3, and Weeks 1, 2, 4, 8, 12, 16, 20, 24 and every 12 weeks thereafter during treatment. Visit windows will be ±1 day through Week 4 and ±3 days thereafter. The following evaluations will be performed:

1. A brief physical examination including body weight and vital statistics
2. A urine pregnancy test for all females of child-bearing potential (except for Day 3)
3. Review concomitant medications
4. Collection of blood sample for qHBV DNA, qHBsAg, qAnti-HBs, HBeAg, anti-HBe and anti-HIV (anti-HIV is only for subjects at high risk of contracting HIV according to the discretion of the investigator)
5. Collection of blood sample to be stored for exploratory analyses of HBV RNA, HBcrAg, qAnti-HBc, quantitative cccDNA, HBs fragments, HBsAg-anti-HBs immune complex, immunological studies and/or host DNA (to assess relationships with virologic response)
6. Collection of blood sample for laboratory safety: hemoglobin, hematocrit, complete blood count, (total and differential), platelet count, PT, INR, AST, ALT, GGT, CPK, alkaline phosphatase, bilirubin total/direct, albumin, BUN, creatinine, sodium, potassium, chloride, and AFP (except for Day 3).

7. Collection of blood samples for trough (just prior to dosing) plasma levels of NrtI and tizoxanide.
8. Collection of a urine sample for routine urinalysis (appearance, glucose, protein and blood) (except for Day 3).
9. Obtain a 12-lead ECG (Week 12 and end of treatment visits only).
10. FibroScan® testing (at end of treatment only).
11. Ultrasonography for HCC screening (every 6 months).
12. Review of adverse events/side effects.
13. Review of compliance, collection of any unused medications from the previous visit, and dispense new supply of study medication.

9.4. Follow-up After End of Treatment with NTZ/Placebo

Subjects will return to the clinic for follow-up evaluation 4, 8 and 12 weeks after the end of treatment with the investigational medication (NTZ/placebo). In the event that a subject has sustained HBsAg loss with HBV DNA suppression at the Week 12 follow-up visit, that subject will also return for additional follow-up visits at 24 and 48 weeks after the end of treatment. The following evaluations will be performed during each follow-up visit:

1. Brief physical examination including body weight and vital statistics.
2. Review concomitant medications.
3. Collection of blood sample for qHBV DNA, qHBsAg, qAnti-HBs, HBeAg, anti-HBe and anti-HIV (anti-HIV is only for subjects at high risk of contracting HIV according to the discretion of the investigator).
4. Collection of blood sample to be stored for exploratory analyses of HBV RNA, HBcrAg, qAnti-HBc, quantitative cccDNA, HBs fragments, HBsAg-anti-HBs immune complex, immunological studies and/or host DNA (to assess relationships with virologic response).
5. Collection of blood sample for laboratory safety: hemoglobin, hematocrit, complete blood count, (total and differential), platelet count, PT, INR, AST, ALT, GGT, CPK, alkaline phosphatase, bilirubin total/direct, albumin, BUN, creatinine, sodium, potassium, chloride, and AFP.
6. Collection of urine sample for routine urinalysis (appearance, glucose, protein and blood).
7. FibroScan® testing (at end of follow-up only).
8. Ultrasonography for HCC screening (every 6 months).
9. Review of adverse events/side effects.

All adverse events must be followed up until they resolve or have stabilized (even beyond the follow-up visit dates provided above).

9.5. Unscheduled Visit

Subjects returning to the clinic for an unscheduled visit due to adverse events will be evaluated at the discretion of the Investigator as is medically warranted. Tests and/or procedures performed at this visit may include, but are not limited to the following:

1. Physical examination, including body weight and vital statistics.
2. Review of compliance with study medication.
3. Review concomitant medications.
4. Collection of blood sample for qHBV DNA, qHBsAg, qAnti-HBs, HBeAg, anti-HBe and anti-HIV (anti-HIV is only for subjects at high risk of contracting HIV according to the discretion of the investigator).
5. Collection of blood sample for laboratory safety: hemoglobin, hematocrit, complete blood count, (total and differential), platelet count, PT, INR, AST, ALT, GGT, CPK, alkaline phosphatase, bilirubin total/direct, albumin, BUN, creatinine, sodium, potassium, chloride, and AFP.
6. Collection of urine sample for routine urinalysis (appearance, glucose, protein and blood).
7. Review of adverse events/side effects.

9.6. Study Medication Discontinuation and Withdrawal Criteria

Treatment with the investigational medication (NTZ/placebo) will be discontinued for individual subjects for the following reasons:

1. Subject withdraws consent.
2. A subject must discontinue study medication for the following medical or administrative reasons [reasons for discontinuing must be recorded in the case report form (CRF)]:
    - Violation of eligibility criteria.
    - Material deviation from the treatment plan specified in the protocol.
    - Progression of the disease which in the opinion of the investigator precludes further participation.
    - Women who become pregnant.
3. An adverse event deemed sufficiently severe to require study discontinuation. These include:
    - Grade 2 or higher rash at least possibly related to study drug.
    - Grade 3 or 4 acute systemic allergic reaction.
    - Grade 2 serum creatinine considered at least possibly related to the study drug by the investigator, or Grade 3 or 4 proteinuria on urinalysis, confirmed by retesting.
    - Grade 2 vomiting that persists despite anti-emetics, or Grade 3 or higher vomiting.
    - Grade 3 or higher diarrhea at least possibly related to study drug.

- Treatment-emergent elevation of ALT greater than 2x baseline, in combination with either bilirubin greater than 2x the upper limit of normal (ULN) or INR greater than 1.5x ULN or serum albumin less than the lower limit of normal (LLN), confirmed by retesting.
- Any Grade 4 AE or confirmed abnormality (except for AST/ALT elevations) considered at least possibly related to the study drug.
- Clinical signs of hepatic decompensation.
- QTcF prolongation (QTcF greater than 500 msec, or increase from baseline greater than 60 msec) at any time point, confirmed by repeat ECG.

4. Subject is:
   - HBsAg positive with no decline in qHBsAg from Baseline at Week 12.
   - HBsAg positive with less than a 0.5 $\log_{10}$ decline in qHBsAg from Baseline at Week 24.
   - HBsAg positive with less than a 1 $\log_{10}$ decline in qHBsAg from Baseline at Week 48.
   - HBsAg positive with less than a 2 $\log_{10}$ decline in qHBsAg from Baseline at Week 72.
   - HBsAg positive at Week 96.
5. HBsAg loss with HBV DNA suppression maintained on treatment for 24 weeks (consolidating therapy).

IMPORTANT: TDF, TAF or ETV therapy will only be discontinued after a subject experiences sustained HBsAg loss with HBV DNA suppression for 24 weeks on treatment (consolidating therapy). Otherwise, subjects will continue TDF, TAF or ETV therapy after discontinuing the investigational medication (NTZ/placebo).

9.7. Rescue Therapy

Additional HBV therapy may be initiated at the discretion of the investigator in the following circumstances:

- Hepatitis B flare defined as abrupt rise in ALT greater than 10x upper limit of normal.
- Evidence of liver decompensation with bilirubin greater than 2x upper limit of normal, prolongation of PT greater than 1 second associated with hepatitis B reactivation, confirmed by repeated test within 4 weeks apart.
- Evidence of clinical liver decompensation with development of ascites, encephalopathy, variceal bleeding.

9.8. Virology Testing and Plan for Monitoring Resistance

A central laboratory will perform all clinical virology testing. All virologic assessments will be performed using an FDA-approved or FDA-cleared assays. Quantitative HBsAg will be determined via a chemiluminescent immunoassay. In the case of virologic failure on treatment (quantifiable HBV DNA after being less than LLOQ), genotypes and phenotypes of Baseline and virologic failure isolates will be determined, and newly emerged drug resistance-associated substitutions/mutations will be identified.

10. DATA MANAGEMENT

10.1. Electronic Data Entry

Data will be transcribed from source documents into an electronic data capture (EDC) system.

The responsible study monitor(s) will verify data, which can be performed remotely or at the clinical study site. The Investigator will ensure that the data recorded are accurate and complete.

Queries emerging during data cleaning will be generated within the EDC system by data management or clinical research associates. The Investigator or his/her designee will answer the queries and update the source data, if needed.

Adverse events and comorbid conditions will be coded by data management using the latest version of the Medical Dictionary for Regulatory Activities (MedDRA); medications will be coded by data management using the World Health Organization (WHO) DRUG dictionary.

After the Last Subject Last Visit (LSLV) has taken place, the database will be cleaned as necessary. As soon as the database is considered clean, it will be locked. The locked database will be used in the final statistical analysis for study reporting. Measures will be undertaken to protect subject data handled during inspections against disclosure to unauthorized third parties. Subject confidentiality will be maintained at all times.

10.2. Protocol Deviations

Protocol deviations will be documented by the Investigator, reported to the institutional review board (IRB) as appropriate, and also reviewed by the assigned clinical research associate (CRA). Deviations will be reported in the clinical trial management system. Each deviation will be classified as major or minor according to the following definitions:

Major protocol deviation: A deviation that has an impact on subject safety, may substantially alter risks to subjects, may have an effect on the integrity of the study data, or may affect the subject's willingness to participate in the study.

Minor protocol deviation: All other protocol deviations.

10.3. Data Quality Assurance

The accuracy and reliability of the study data will be assured by the selection of qualified Investigators and appropriate study centers, review of protocol procedures with the Investigator and associated personnel prior to the study, and by periodic monitoring visits by the Sponsor. Written instructions will be provided for collection, preparation, and shipment of samples.

The monitor will review the source data for accuracy and completeness, and any discrepancies will be resolved with the Investigator or designee, as appropriate.

The Sponsor will be entitled to inspect and audit the facilities used in the clinical and laboratory parts of the study, as well as to make anonymized copies of all the data files pertaining to the study. Similar procedures may also be conducted by agents of any regulatory body, either as part of a national Good Clinical Practice (GCP) compliance program or to review the results of this study in support of a regulatory submission. The Investigator should immediately notify the Sponsor if they have been contacted by a regulatory agency concerning an upcoming inspection.

11. STATISTICAL CONSIDERATIONS

11.1. Sample Size Calculation

The study is designed to evaluate the safety and efficacy of three different NTZ treatment regimens compared to placebo as add-on therapy to TDF, TAF or ETV in reducing quantitative HBsAg (in order to select a dose for future clinical trials).

A student's t-test with a 0.05 two-sided significance level will have 90% power to detect the difference between a mean change of 0.5 log10 for one treatment group and no change for the other treatment group when sigma (common standard deviation) is 0.33 and the sample size in each group is 10. A sample size of 12 per group was selected in order to allow for the possibility of up to 2 non-evaluable subjects per group (due to drop-out, etc.).

11.2. Efficacy Variables

Primary Efficacy Parameter: The mean change in qHBsAg from Baseline to Week 12.

Secondary Efficacy Parameters:

i. Sustained HBsAg loss with suppression of HBV DNA for 24 weeks after the end of treatment.

ii. Change in qHBsAg from Baseline to different time points on treatment.

iii HBsAg loss.

iv. HBsAg seroconversion.

v. HBV DNA suppression.

vi. Change from Baseline in Fibrosis 4 (FIB-4) score.

vii. Change from Baseline in FibroScan® score.

11.3. Response Definitions

HBsAg Loss: HBsAg below the lower limit of detection.

HBsAg seroconversion: Loss of HBsAg and gain of anti-HBs.

HBV DNA suppression: HBV DNA less than lower limit of quantitation; target not detected.

11.4. Statistical Methodology

The statistical methodology is described briefly below and will be described in detail in the Statistical Analysis Plan.

11.4.1. Efficacy Analyses

The study will be unblinded to the Sponsor (Romark) and Medical Monitor for analysis of efficacy and safety data after all subjects have completed 12 weeks of treatment and complete data is collected through that time point. The investigators, subjects and monitors will remain blinded throughout the study.

Efficacy analyses will be based on a population consisting of all subjects randomly assigned to the study ("intent-to-treat" population). In the event that there are significant numbers of subjects who fail to complete the study, violate the protocol, or are noncompliant with the study medication, efficacy analyses will also be conducted for a population excluding these subjects ("per protocol population").

Primary efficacy analyses will be conducted as follows: For each NTZ treatment group, the mean change in qHBsAg from Baseline to Week 12 will be compared to that of the placebo treatment group.

Secondary efficacy analyses will be conducted as follows:

- For each of the three NTZ groups, the proportion of subjects with sustained suppression of HBV DNA and HBsAg loss at 24 weeks after the end of treatment will be compared to the proportion for the placebo treatments group.
- For each NTZ dose group and for each study time point, the change in qHBsAg from Baseline will be compared to the change in qHBsAg for the placebo treatment group.
- For each of the three NTZ groups and for each study time point, the proportion of subjects with HBsAg loss will be compared to the proportion for the placebo treatments group.
- For each of the three NTZ groups and for each study time point, the proportion of subjects with HBsAg seroconversion will be compared to the proportion for the placebo treatments group.
- For each of the three NTZ groups and for each study time point, the proportion of subjects with quantifiable HBV DNA at Baseline who experienced HBV DNA suppression will be compared to the proportion for the placebo treatments group.
- For each NTZ dose group and for each study time point, the mean change in Fibrosis 4 (FIB-4) score from Baseline will be compared to the change for the placebo treatment group.
- For each NTZ dose group, the mean change in FibroScan® score from Baseline to end of treatment and to end of off-treatment follow-up will be compared to the change for the placebo treatment group.

Means will be compared by two-sided student's t tests, $\alpha = 0.05$. Nonparametric data will be analyzed using the Wilcoxon rank sum (Mann-Whitney) test. Proportions will be compared using a two-sided Fisher's Exact test, $\alpha = 0.05$. In the setting of an early Phase 2 study, no adjustments for multiplicity are planned.

11.4.2. Safety Analyses

All randomized subjects who receive the study medication will be assessed for drug safety. Safety analyses will be done descriptively.

12. INVESTIGATIONAL PRODUCTS

12.1. Identity of Investigational Products

NTZ 300 mg extended release tablets manufactured for Romark Laboratories, L.C. by Xcelience Inc., Tampa, Florida. Each tablet is a yellow, round, convex, film-coated, bi-layer tablet for oral administration, each tablet contains 300 mg of NTZ and the inactive ingredients.

The study medications will be provided by the study sponsor.

12.2. Packaging and Labeling

All study medication will be packaged and labeled by the study sponsor and its contractors. NTZ and placebo tablets will packaged in white 30cc HDPE bottles, each containing 30 tablets. The bottles will be stored at room temperature. Do not store above 25ºC and protect from moisture. The box and bottles will bear a label with the following information:

External Box Label

Investigator Name

180 Tablets **RM08-2001** Treatment #

Lot# Nitazoxanide/Placebo 300 mg Tablets

EXP Date Take one tablet from bottle A, B, and C by mouth each morning with food, and take one tablet from bottle D, E, and F by mouth each evening with food.

"For clinical trial use only"

Do not store above 25ºC. Protect from moisture.

Study Sponsor: The Romark Institute for Medical Research

[redacted]

Individual Bottle Label (AM Doses)

30 Tablets **RM08-2001** Treatment #

Lot# Nitazoxanide/Placebo 300 mg Tablets Bottle # (A-C)

Take one tablet by mouth each morning with food.

"For clinical trial use only"

Do not store above 25ºC. Protect from moisture.

Study Sponsor: The Romark Institute for Medical Research

[redacted]

Individual Bottle Label (PM Doses)

30 Tablets	**RM08-2001**	Treatment #
Lot#	Nitazoxanide/Placebo 300 mg Tablets	Bottle # (D-F)
	Take one tablet by mouth each evening with food.	
	"For clinical trial use only"	
	Do not store above 25ºC. Protect from moisture.	
	Study Sponsor: The Romark Institute for Medical Research	
	[redacted]	

Subjects will be provided with an information card containing the name, address, and telephone number of the best contact if they have questions.

12.3. Drug Accountability

Medication supplies will be dispensed at Baseline (Day 1) and every 4 to 12 weeks during the treatment period, depending on the visit schedule. Subjects will be asked to return any unused supplies at each visit.

The investigator is required to maintain adequate records of the disposition of all study drug, including dates, quantity and use by subject. Unused supplies must be returned to the Sponsor or destroyed in accordance with institutional procedures in effect at the study site.

12.4. Subject Compliance and Disallowed Medications

Subject compliance with the protocol will be checked by the Investigator and recorded in the CRF at each visit.

Subjects will be considered non-compliant if they have missed more than 20% of the doses of the study medication or if they take any antiviral drug active against HBV other than the investigational medication, TDF, TAF and ETV.

During the active treatment course of the study, concomitant use of medications considered major CYP2C8 substrates (Section 23.1) are prohibited. Subjects are also prohibited from taking any other investigational medication during the study.

13. ADVERSE EVENTS

The term "adverse event" is defined for purposes of this study as any unwanted physical, psychological or behavioral change experienced by a subject during the course of the study and after taking the first dose of study medication regardless of its severity or relation to the study. Adverse events may include symptoms, signs, unexpected worsening of pre-existing conditions, clinically significant changes in laboratory values, diseases and syndromes, and significant and unexpected failures of pharmacological action of other medications. Symptoms of CHB will not be reported as adverse events.

Adverse events will be recorded on the appropriate EDC forms throughout the study, and the severity of each adverse event will be graded on a four-point scale: mild, moderate, severe, or life-threatening (See Section 23.3). The duration of the adverse event and relationship to the study drug will also be recorded. All adverse events must be followed until their resolution or stabilization even beyond the planned study period.

13.1. Definitions

The following definitions will apply to the reporting of adverse events:

1. Serious Adverse Event: Any adverse experience occurring at any dose that is fatal or life threatening; requires in-patient hospitalization or prolongation of an existing hospitalization; is a persistent significant disability/incapacity; is a congenital anomaly or birth defect; or is an important medical event that may not result in death, be life-threatening, or require hospitalization but, based upon appropriate medical judgment, may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed above.
2. Unexpected Adverse Event: Any adverse experience that is not identified in nature, severity, or frequency in the Investigator's Brochure for NTZ or the respective prescribing information for TDF, TAF or ETV.
3. Severity of adverse events will be assessed by the Investigator using the Toxicity Grading Scale Tables provided in Section 23.3 (derived and adapted from "Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events," Version 2.1, Publish Date: July 2017)
4. Causality (relationship to treatment) will be assessed as follows:

The Investigator will assess causal relationship between Investigational Product and each adverse event and answer "yes" or "no" to the question, "Do you consider that there is a reasonable possibility that the event may have been caused by the Investigational Product?" Under double-blind treatment conditions, it should be assumed that all subjects are taking the test drug.

13.2. Clinical Adverse Events

At the time of each return visit, the subject will be questioned regarding the occurrence and nature of any adverse events. All events must be recorded in the subjects' medical records and in the EDC system. Any subject affected will be examined by the Investigator as deemed necessary to ascertain the course of the event and any residual effects.

All moderate and severe adverse events will be reviewed by the Principal Investigator who will determine using his/her best clinical judgment whether they warrant the subject to be discontinued from the study. The Sponsor will be notified immediately if a subject is discontinued from the study. For all adverse events that require the subject to be discontinued from the study, relevant clinical assessments and laboratory tests will be repeated as clinically indicated until final resolution or stabilization of the event(s).

All subjects will be instructed to contact the Investigator, Investigator's assistants, or clinical personnel should the subject have any serious adverse experiences. Serious adverse events,

including death regardless of the cause, must be reported to the Sponsor immediately (within 24 hours of the initial report).

A serious event requiring immediate notification by telephone is an event that:

- results in death
- is life threatening
- requires inpatient hospitalization or prolongation of an existing hospitalization
- is a persistent or significant disability/incapacity
- is a congenital anomaly/birth defect
- an important medical event that may not result in death, be life-threatening, or require hospitalization but, based upon appropriate medical judgment, may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed above.

An overdose is defined as any intentional or unintentional consumption of the drug by any route that exceeds the highest dose stated in the Investigator's Brochure, approved product labeling (for TDF, TAF or ETV) or in an investigational protocol, whichever dose is larger. Overdoses without an associated adverse event should be recorded, but not reported as an adverse event.

13.3. Reporting Requirement

The Principal Investigator is required to notify The Romark Institute for Medical Research (Sponsor) within 24 hours of learning of any unexpected, fatal, or life-threatening experience and all unusual, alarming, or serious reactions to medication regardless of any opinions as to the cause/effect relationship. All serious adverse events will also be reported to the IRB. Adverse events should be reported to:

The Romark Institute for Medical Research

Medical Affairs

[redacted]

13.4. Medication Modification/Withdrawal Due to Adverse Event

If a severe adverse reaction or laboratory abnormality develops during combination therapy, the dose should be modified or discontinued, at the discretion of the Investigator and in accordance with the approved product labeling for TDF, TAF or ETV, until the adverse reaction abates. If intolerance persists after the dose has been adjusted, the study medication should be discontinued.

13.5. Medication Errors

A medication error is defined as any preventable event related to dosing instructions, product labeling, or packaging that causes or leads to inappropriate medication use or subject harm while the medication is in the control of the investigative site or subject. Medication errors which result in adverse events should be recorded and reported as adverse events. All other medication errors should be reported to the study Sponsor through the Medical Affairs department within 7 days of identification by the site.

14. DISCONTINUATION

The study may be discontinued under the following circumstances:

1. The Sponsor reserves the right to discontinue the study at any time.
2. Adverse event listings will be produced for safety monitoring at least once every two weeks during recruitment. The data will be tested to determine if there are greater than 20% of the subjects who have had at least Grade 3 adverse events considered by the Investigator to be related to the study drug (defined by the Toxicity Grading Scale Tables provided in Section 23.3).

If greater than 20% of the subjects have at least one Grade 3 or Grade 4 adverse event considered to be related to the study drug by the study Medical Monitor, then the study must be stopped and the Institutional Review Boards, local regulatory authorities and FDA's Division of Antiviral Drug Products will be notified. All safety and activity data will be submitted to the FDA in a timely manner.

15. RETENTION OF RECORDS

Essential Documents (EDs) are documents that individually and collectively permit evaluation of the conduct of a trial and quality of the data produced. They demonstrate the compliance of the Investigator, Sponsor, and monitor with the GCP standards and with all applicable regulatory requirements.

In compliance with the International Council for Harmonisation (ICH)/GCP guidelines, the Investigator/institution will maintain all CRFs and all EDC source forms and source documents that support the data collected from each subject, as well as all study documents as specified in ICH/GCP Section 8, Essential Documents for the Conduct of a Clinical Trial, and all study documents as specified by the applicable regulatory requirement(s). A list of these documents is found in Section 23.4: List of Essential Document for the Investigative Site. The Investigator/institution will take measures to prevent accidental or premature destruction of these documents.

Essential documents listed in Section 23.4 must be retained for the duration required by applicable regulatory authorities or until the Sponsor informs the Investigator/institution these documents are no longer needed.

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

The Sponsor is responsible for organizing and maintaining the Trial Master File which is a clear documentation of the course of the study.

16. MONITORING THE STUDY

Monitoring will be conducted by the Sponsor and/or a contract research organization (CRO) according to the Sponsor's standard operating procedures. Site visits will be conducted by the Sponsor at regular intervals to conduct inspections.

Any data transcribed into the EDC system will be 100% source verified.

The Investigator will grant representatives of the Sponsor's clinical operations team and quality team, as well as regulatory agencies and ethical committees access to inspect facilities and records (including subject charts) relevant to this study and agrees to assist the monitors in their activities, if requested.

The data from this study will not be monitored by an independent data monitoring committee.

17. INFORMED CONSENT

Each subject must give written consent according to local requirements after the nature of the study has been fully explained. The informed consent form (ICF) must be signed before performance of any study-related activity. The informed consent form will be approved by both the Sponsor and by the reviewing IRB. They will be in accordance with principles that originated in the Declaration of Helsinki, current ICH and GCP guidelines, applicable regulatory requirements, and Sponsor policy.

Before enrollment in the study, the Investigator or an authorized member of the investigational staff must explain to potential subjects the aims, methods, reasonably anticipated benefits, and potential hazards of the study, and any discomfort participation in the study may entail. Subjects will be informed that their participation is voluntary and that they may withdraw consent to participate at any time. They will be informed that choosing not to participate will not affect the care he/she will receive. Finally, they will be told that the Investigator will maintain a subject identification register for the purposes of long-term follow-up if needed and that their records may be accessed by health authorities and authorized Sponsor staff without violating the confidentiality of the subject, to the extent permitted by the applicable law(s) or regulations. By signing the ICF subjects are authorizing such access, and agree to allow the study physician to re-contact the subject for the purpose of obtaining consent for additional safety evaluations, if needed.

The subject will be given sufficient time to read the ICF and the opportunity to ask questions. After this explanation and before entry into the study, consent should be appropriately recorded by means of the subject's personally dated signature. After having obtained the consent, a copy of the ICF must be given to the subject.

If the subject is unable to read or write, an impartial witness should be present for the entire informed consent process (which includes reading and explaining all written information) and should personally date and sign the ICF after the oral consent of the subject is obtained, if permitted by local law.

The consent forms must be signed prior to the performance of any study related activity.

Signed ICFs must remain in the subject's file(s) and be available for verification by representatives of Romark, the IRB, and FDA/relevant regulatory agencies at any time.

18. ETHICS

The clinical trial will be performed in accordance with the guidelines set by the World Medical Assembly (Declaration of Helsinki, last amendment in Fortaleza, Brazil, October 2013). Prior written approval of the study protocol and of the informed consent form will be obtained from the appropriate local Medical Ethics Review Board.

18.1. Study-specific Design Considerations

Potential subjects will be fully informed of the risks and requirements of the study and during the study subjects will be given any new information that may affect their decision to continue participation. They will be told that their consent to participate in the study is voluntary and may be withdrawn at any time with no reason given and without penalty or loss of benefits to which they would otherwise be entitled. Only subjects who are fully able to understand the risks, benefits, and potential AEs of the study, and provide their consent voluntarily will be enrolled.

18.2. Investigator Responsibilities

The Investigator is responsible for ensuring that the clinical study is performed in accordance with the protocol, current International Council for Harmonisation (ICH) guidelines on GCP, and applicable regulatory and country specific requirements.

Good Clinical Practice is an international ethical and scientific quality standard for designing, conducting, recording, and reporting studies that involve the participation of human subjects. Compliance with this standard provides public assurance that the rights, safety, and wellbeing of study subjects are protected, consistent with the principles that originated in the Declaration of Helsinki, and that the clinical study data are credible.

18.3. Institutional Review Board

Before the start of the study, the Investigator (or Sponsor where required) will provide the IRB with current and complete copies of the following documents:

- Final protocol and, if applicable, amendments.
- Sponsor-approved ICF (and any other written materials to be provided to the subjects).
- Investigator's Brochure (or equivalent information) and addenda.
- Sponsor-approved subject recruiting materials.
- Information on compensation for study-related injuries or payment to subjects for participation in the study, if applicable.
- Investigator's curriculum vitae or equivalent information (unless not required, as documented by IRB).

- Information regarding funding, name of the Sponsor, institutional affiliations, other potential conflicts of interest, and incentives for subjects.
- Any other documents that the IRB requests to fulfill its obligation.

This study will be undertaken only after the IRB has given full approval of the final protocol, amendments (if any), the ICF, applicable recruiting materials, and subject compensation programs, and the Sponsor has received a copy of this approval. This approval letter must be dated and must clearly identify the IRB and the documents being approved.

During the study, the Investigator (or Sponsor where required) will send the following documents and updates to the IRB for their review and approval, where appropriate:

- Protocol amendments.
- Revision(s) to ICF and any other written materials to be provided to subjects.
- If applicable, new or revised subject recruiting materials approved by the Sponsor.
- Revisions to compensation for study-related injuries or payment to subjects for participation in the study, if applicable.
- Investigator's Brochure addenda or new edition(s).
- Summaries of the status of the study at intervals stipulated in guidelines of the IRB (at least annually).
- Reports of AEs that are serious, unlisted, and associated with the investigational drug.
- New information that may adversely affect the safety of the subjects or the conduct of the study.
- Deviations from or changes to the protocol to eliminate immediate hazards to the subjects.
- Report of deaths of subjects under the Investigator's care.
- Notification if a new Investigator is responsible for the study at the site.
- Annual IND Update Report, Short Term Study Specific Safety Summary and Line Listings, where applicable.
- Any other requirements of the IRB.

For all protocol amendments (excluding the ones that are purely administrative, with no consequences for subjects, data or study conduct), the amendment and applicable informed consent form and assent form revisions must be submitted promptly to the IRB for review and approval before implementation of the change(s), except when necessary to eliminate immediate hazard to the study subjects. If a deviation from, or a change to the protocol was implemented to eliminate an immediate hazard to study subjects, then the implemented deviation or change, the reasons for it, and, if appropriate, the protocol amendment should be submitted to the IRB as soon as possible.

The re-approval of the clinical study by the IRB should be documented in writing.

At the end of the study, the Investigator (or Sponsor where required) will notify the IRB about the study completion.

18.4. Privacy of Personal Data

The collection and processing of personal data from subjects enrolled in this study will be limited to those data that are necessary to investigate the safety, quality, and utility of the investigational study drug(s) used in this study.

These data must be collected and processed with adequate precautions to ensure confidentiality and compliance with applicable data privacy protection laws and regulations. Appropriate technical and organizational measures to protect the personal data against unauthorized disclosures or access, accidental or unlawful destruction, or accidental loss or alteration must be put in place. Sponsor personnel whose responsibilities require access to personal data agree to keep the identity of study subjects confidential.

The informed consent obtained from the subject includes explicit consent for the processing of personal data and for the Investigator to allow direct access to his or her original medical records for study-related monitoring, audit, IRB review, and regulatory inspection. This consent also addresses the transfer of the data to other entities and to other countries.

19. DATA CONFIDENTIALITY, DISCLOSURE OF DATA, AND PUBLICATION

Data generated for the study should be stored by the Investigator in a limited-access file area and be accessible only to representatives of the study site, Romark, the IRB, and FDA/relevant regulatory agencies. Medical information resulting from a subject's participation in this study may be given to the subject's personal physician or to the appropriate medical personnel responsible for the subject's welfare.

No information that can be related to a specific individual subject may be released or used in any fashion without the signed written consent of that subject. All reports and communications relating to study subjects will identify subjects only by initials and subject identification number. The Investigator will keep complete subject identification for purposes of long-term follow-up, if needed. This information will be treated with strict adherence to professional standards of confidentiality.

Site personnel will enter data relating to each subject's participation in the study into the EDC system provided by the Sponsor. In the EDC system, identification numbers and initials will be used to identify subjects. Subject names will not be used in the CRFs. Management of data from the EDC system and the production of the clinical study report will be the responsibility of the Sponsor. Access to the database will be restricted to employees who have been trained to use the system. Access to the EDC system and study report will be limited to the IRB, FDA or other regulatory agencies and the Sponsor.

Presentation and/or publication of the results of the study is encouraged provided that The Romark Institute for Medical Research is notified in advance of the author's intent and is given the opportunity to review the manuscript or abstract 45 days prior to its submission for presentation at a scientific meeting or for publication in a scientific journal. The Investigators will have complete autonomy regarding the content and wording of any abstracts, presentations,

and scientific publications arising from this study, including the decision of whether or not to publish.

20. DATA AND REPORT REQUIREMENTS

Data required by The Romark Institute for Medical Research prior to approval and initiation of the study are as follows:

1. Curriculum vitae of the Principal Investigator and all Co-Investigators.
2. Copy of the IRB-approved Informed Consent and subject information forms.
3. Copy of the IRB approval for the conduct of the study.

Data and materials required by The Romark Institute for Medical Research before the study can be considered complete and terminated are as follows:

1. Pre- and post-treatment history, physical examination and subject evaluations.
2. Pre- as well as interim and post-treatment laboratory findings and all special test results.
3. EDC forms properly completed and signed by the Principal Investigator.
4. Drug Inventory Logs indicating drug dispensed and return of the unused supplies to the Sponsor or destruction by study site.
5. Signed Informed Consent/Assent from each subject.

21. CONTACT INFORMATION

Table 2: Contact Information

Medical Monitors		
Name:	[redacted]	
Title:	Medical Monitor	
Tel:	[redacted]	
Fax:	[redacted]	
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Sponsor Medical Affairs		
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Sponsor Project Management		
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Title:	EVP, COO	
Tel:	[redacted]	
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E-mail:	[redacted]	
Contract Research Organization		
Name:	[redacted]	
Title:	Director, [redacted]	
Tel:	[redacted]	
E-mail:	[redacted]	
Investigational Product Supplier		
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22. LIST OF REFERENCES

Anderson VR, Curran MP. Nitazoxanide: A review of its use in the treatment of gastrointestinal infections. Drugs 2007;67:1947-1967.

Backman JT, Filppula AM, Niemi M, Neuvonen PJ. Role of cytochrome P450 2C8 in drug metabolism and interactions. Pharmacol Rev 2016;68(1):168-241.

European Association for the Study of the Liver. EASL 2017 Clinical Practice Guidelines on the management of hepatitis B virus infection. J Hepatol 2017;67:370-398.

Kolozi W, El-Gohary Y, Keeffe E, Rossignol JF. Treatment of chronic hepatitis B (CHB) with nitazoxanide (NTZ) alone or NTZ Plus adefovir (ADV) for two years with loss of hepatitis B e antigen (HBeAg) and hepatitis B surface antigen (HbsAg): report of two cases [Abstract]. Am J Gastroenterol 2008;103 (S1):S150-S151.

Korba BE, Mueller AB, Farrar K, Gaye K, Mukerjee S, Ayers MS, Rossignol JF. Nitazoxanide, tizoxanide and other thiazolides are potent inhibitors of hepatitis B virus and hepatitis C virus replication. Antiviral Res 2008;77:56-63.

Lok A SF, McMahon BJ, Brown RS Jr, Wong JB, Ahmed AT, et al. Antiviral therapy for chronic hepatitis B viral infection in adults: a systematic review and meta-analysis. Hepatology 2016;63:284-306.

Romark, L.C. Alinia (nitazoxanide) prescribing information. April 2017.

Rossignol JF. Nitazoxanide: A first-in-class broad-spectrum antiviral agent. Antiviral Res 2014;110:94-103.

Rossignol JF, Brechot C. A pilot clinical trial of nitazoxanide in the treatment of chronic hepatitis B. Hepatol Comm 2019;6:744-747.

Rossignol JF, Keeffe EB. Thiazolides: a new class of drugs for the treatment of chronic hepatitis B and C. Future Microbiol 2008;3:539-545.

Sarin SK, Kumar M, Lau GK, Abbas Z, Chan HL, Chen CJ et al. Asian-Pacific clinical practice guidelines on the management of hepatitis B: a 2015 update. Hepatol Int 2016;10:1-98.

Sekiba K, Otsuka M, Ohno M, Yamagami M, Kishikawa T, et al. Inhibition of HBV transcription from cccDNA with nitazoxanide by targeting the HBX-DDB1 interaction. Cell Mol Gastroenterol Hepatol 2019;7:297-312.

Terrault NA, Bzowej NH, Chang K-M, Hwang JP, Jonas MM, Murad MH. American Association for the Study of Liver Diseases. AASLD guidelines for treatment of chronic hepatitis B. Hepatology 2016;63:261-283.

Terrault NA, Lok ASF, McMahon BJ, Chang KM, Hwang JP, et al. Update on prevention, diagnosis and treatment of chronic hepatitis B: AASLD 2018 hepatitis B guidance. Hepatology 2018;67:1560-1598.

World Health Organization. Hepatitis B Fact Sheet. July 18, 2018. Accessed at: http://www.who.int/news-room/fact-sheets/detail/hepatitis-b. Accessed date: February 5, 2019.

23. APPENDICES

23.1. Appendix I: Major CYP2C8 Substrates

Table 3: Major CYP2C8 Substrates

Generic Name	Brand Name(s)	Therapeutic Use and/or Drug Class
Amodiaquine	BASOQUIN, CAMOQUIN, FLAVOQUIN	Antimalarial
Cerivastatin (acid, parent)	BAYCOL *	Antihyperlipidemic, HMG-CoA reductase inhibitor
Daprodustat (GSK1278863)	**	Antianemic, prolyl hydroxylase inhibitor
Dasabuvir (ABT-333)	EXVIERA	Antiviral, NSB5 inhibitor
Enzalutamide	XTANDI	Anticancer, antiandrogen
Montelukast	SINGULAIR	Antiasthmatic, LTRA
Pioglitazone	ACTOS	Antidiabetic, PPAR-γ agonist
Repaglinide	NOVONORM, PRANDIN	Antidiabetic, meglitinide analog

Source: Backman et al., 2016.

*Only available outside of US.

**Currently listed as investigational only and not yet granted a trade name by the manufacturer.

23.2. Appendix II: Schedule of Study Visits and Procedures

Table 4: Schedule of Assessments

		Treatment Period											Follow-up Period				
		Day		Week								Every 12 Weeks Thereafter	Week				
	SCREEN	1	3	1	2	4	8	12	16	20	24		4	8	12	24[2]	48[2]
Informed consent	x																
Review inclusion/exclusion criteria	x																
Medical history	x																
Height	x																
Physical exam including weight, vital signs	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Urine pregnancy test[1]	x	x		x	x	x	x	x	x	x	x	x					
Review concomitant medications	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Blood sample for viral serology[3]	x																
Blood sample for viral serology[4]		x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Blood sample for exploratory analyses		x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Blood sample for PK (pre-dose)		x	x	x	x	x	x	x	x	x	x	x					
Blood sample for safety tests	x	x		x	x	x	x	x	x	x	x	x	x	x	x	x	x
Urine collection for urinalysis	x	x		x	x	x	x	x	x	x	x	x	x	x	x	x	x
Fibroscan®	x[5]											x[5]			x[5]		x[5]
Ultrasonography HCC screening	x[6]										x[6]	x[6]	x[6]	x[6]	x[6]	x[6]	x[6]
Electrocardiogram		x						x				x[7]					
Randomization		x															
Adverse events			x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Dispense drugs & accountability		x				x	x	x	x	x	x	x					

[1] For women of childbearing potential only.

[2] Week 24 and 48 follow-ups are only required if subjects have sustained HBsAg loss with HBV DNA suppression at the Week 12 off-treatment follow-up visit.

[3] Tests performed on Screening sample will include qHBV DNA, qHBsAg, anti-HBs, HBeAg, anti-HBe, IgM anti-HBc, anti-HCV, anti-HDV, anti-HIV.

[4] Tests performed will include: qHBV DNA, qHBsAg, qAnti-HBs, HBeAg, anti-HBe, and anti-HIV (anti-HIV is only for subjects at high risk of contracting HIV according to the discretion of the investigator).

[5] FibroScan® will be performed at Screening, the end-of-treatment visit, and at the last off-treatment follow-up visit (either 12 or 48 weeks after end of treatment).

[6] Ultrasonography for HCC will be performed at Screening and every 6 months during the study.

[7] A 12-lead ECG will be obtained at Baseline, Week 12 and at end of treatment only.

23.3. Appendix III: Toxicity Grading for Adult Adverse Events

See "Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events; Version 2.1, Publish Date: July 2017" (Table 5).

General Instructions

Estimating Severity Grade: If the need arises to grade a clinical adverse event that is not identified in this table, use the category "Estimating Severity Grade" located at the top of Table 5. In addition, all deaths related to an AE are to be classified as Grade 5.

Determining Severity of Grade: If the severity of an adverse event could fall under either one of two grades (e.g., the severity of an adverse event could be either Grade 2 or Grade 3), select the higher of the two grades for the adverse event.

Table 5: Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events; Version 2.1, Publish Date: July 2017

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
ESTIMATING SEVERITY GRADE				
Clinical adverse event NOT identified elsewhere in this grading table	Mild symptoms causing no or minimal interference with usual social & functional activities with intervention not indicated	Moderate symptoms causing greater than minimal interference with usual social & functional activities with intervention indicated	Severe symptoms causing inability to perform usual social & functional activities with intervention or hospitalization indicated	Potentially life-threatening symptoms causing inability to perform basic self-care functions with intervention indicated to prevent permanent impairment, persistent disability, or death
CLINICAL- CARDIOVASCULAR				
Arrhythmia (by ECG or physical examination) *Specify type, if applicable*	No symptoms AND No intervention indicated	No symptoms AND Non-urgent intervention indicated	Non-life-threatening symptoms AND Non-urgent intervention indicated	Life-threatening arrhythmia OR Urgent intervention indicated
Blood Pressure Abnormalities ***Hypertension*** (with the lowest reading taken after repeat testing during a visit) ≥ 18 years of age	140 to < 160 mmHg systolic OR 90 to < 100 mmHg diastolic	≥ 160 to < 180 mmHg systolic OR ≥ 100 to < 110 mmHg diastolic	≥ 180 mmHg systolic OR ≥ 110 mmHg diastolic	Life-threatening consequences in a participant not previously diagnosed with hypertension (e.g., malignant hypertension) OR Hospitalization indicated
Hypotension	No symptoms	Symptoms corrected with oral fluid replacement	Symptoms AND IV fluids indicated	Shock requiring use of vasopressors or mechanical assistance to maintain blood pressure

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Cardiac Ischemia/Infarction *Report only one*	NA	NA	New symptoms with ischemia (stable angina) OR New testing consistent with ischemia	Unstable angina OR Acute myocardial infarction
Heart Failure	No symptoms AND Laboratory or cardiac imaging abnormalities	Symptoms with mild to moderate activity or exertion	Symptoms at rest or with minimal activity or exertion (e.g., hypoxemia) OR Intervention indicated (e.g., oxygen)	Life-threatening consequences OR Urgent intervention indicated (e.g., vasoactive medications, ventricular assist device, heart transplant)
Hemorrhage (with significant acute blood loss)	NA	Symptomatic AND No transfusion indicated	Symptoms AND Transfusion of ≤ 2 units packed RBCs indicated	Life-threatening hypotension OR Transfusion of > 2 units packed RBCs indicated
Prolonged PR Interval or AV Block	PR interval 0.21 to <0.25 seconds	PR interval ≥ 0.25 seconds OR Type I 2nd degree AV block	Type II 2nd degree AV block OR Ventricular pause ≥ 3.0 seconds	Complete AV block
Prolonged QTc Interval (Per Bazett's formula)	0.45 to 0.47 seconds	> 0.47 to 0.50 seconds	> 0.50 seconds OR ≥ 0.06 seconds above baseline	Life-threatening consequences (e.g. Torsade de pointes, other associated serious ventricular dysrhythmia)
Thrombosis/Embolism *Report only one*	NA	Symptoms AND No intervention indicated	Symptoms AND Intervention indicated	Life-threatening embolic event (e.g., pulmonary embolism, thrombus)
CLINICAL- DERMATOLOGIC				
Alopecia (scalp only)	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	NA	NA
Bruising	Localized to one area	Localized to more than one area	Generalized	NA
Cellulitis	NA	Non-parenteral treatment indicated (e.g., oral antibiotics, antifungals, antivirals)	IV treatment indicated (e.g., IV antibiotics, antifungals, antivirals)	Life-threatening consequences (e.g., sepsis, tissue necrosis)
Hyperpigmentation	Slight or localized causing no or minimal interference with usual social & functional activities	Marked or generalized causing greater than minimal interference with usual social & functional activities	NA	NA

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Hypopigmentation	Slight or localized causing no or minimal interference with usual social & functional activities	Marked or generalized causing greater than minimal interference with usual social & functional activities	NA	NA
Petechiae	Localized to one area	Localized to more than one area	Generalized	NA
Pruritis (without skin lesions) (See also Injection Site Reactions: Pruritis associated with injection)	Itching causing no or minimal interference with usual social & functional activities	Itching causing greater than minimal interference with usual social & functional activities	Itching causing inability to perform usual social & functional activities	NA
Rash *Specify type, if available*	Localized rash	Diffuse rash OR Target lesions	Diffuse rash AND Vesicles or limited number of bullae or superficial ulcerations of mucous membrane limited to one site	Extensive or generalized bullous lesions OR Ulceration of mucous membrane involving two or more distinct mucosal sites OR Stevens-Johnson syndrome OR Toxic epidermal necrolysis (TEN)
CLINICAL- ENDOCRINE and METABOLIC				
Diabetes Mellitus	Controlled without medication	Controlled with medication OR Modification of current medication regimen	Uncontrolled despite treatment modification OR Hospitalization for immediate glucose control indicated	Life-threatening consequences (e.g., ketoacidosis, hyperosmolar non-ketotic coma, end organ failure)
Gynecomastia	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Obvious on visual inspection AND Causing pain with greater than minimal interference with usual social & functional activities	Disfiguring changes AND Symptoms requiring intervention or causing inability to perform usual social & functional activities	NA
Hyperthyroidism	No symptoms AND Abnormal laboratory value	Symptoms causing greater than minimal interference with usual social & functional activities OR Thyroid suppression therapy indicated	Symptoms causing inability to perform usual social & functional activities OR Uncontrolled despite treatment modification	Life-threatening consequences (e.g., thyroid storm)
Hypothyroidism	No symptoms AND Abnormal laboratory value	Symptomatic causing greater than minimal interference with usual social & functional activities OR Thyroid	Symptoms causing inability to perform usual social & functional activities OR Uncontrolled	Life-threatening consequences (e.g., myxedema coma)

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
		replacement therapy indicated	despite treatment modification	
Lipoatrophy (e.g., fat loss from the face, extremities, buttocks)	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Obvious on visual inspection AND Causing greater than minimal interference with usual social & functional activities	Disfiguring changes	NA
Lipohypertrophy (e.g., abnormal fat accumulation on the back of the neck, breasts, and abdomen)	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Obvious on visual inspection AND Causing greater than minimal interference with usual social & functional activities	Disfiguring changes	NA
CLINICAL- GASTROINTESTINAL				
Anorexia	Loss of appetite without decreased oral intake	Loss of appetite associated with decreased oral intake without significant weight loss	Loss of appetite associated with significant weight loss	Life-threatening consequences OR Aggressive intervention indicated [e.g., tube feeding or total parenteral nutrition (TPN)]
Ascites	No symptoms	Symptoms AND Intervention indicated (e.g., diuretics, therapeutic paracentesis)	Symptoms recur or persist despite intervention	Life-threatening consequences
Bloating or Distension *Report only one*	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	NA
Cholecystitis	NA	Symptoms AND Medical intervention indicated	Radiologic, endoscopic, or operative intervention indicated	Life-threatening consequences (e.g., sepsis, perforation)
Constipation	NA	Persistent constipation requiring regular use of dietary modifications, laxatives, or enemas	Obstipation with manual evacuation indicated	Life-threatening consequences (e.g., obstruction)
Diarrhea	Transient or intermittent episodes of unformed stools OR Increase of ≤ 3 stools over baseline per 24-hour period	Persistent episodes of unformed to watery stools OR Increase of 4 to 6 stools over baseline per 24-hour period	Increase of ≥ 7 stools per 24-hour period OR IV fluid replacement indicated	Life-threatening consequences (e.g., hypotensive shock)
Dysphagia or Odynophagia	Symptoms but able to eat usual diet	Symptoms causing altered dietary intake with no	Symptoms causing severely altered dietary	Life-threatening reduction in oral intake

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Report only one and specify location		medical intervention indicated	intake with medical intervention indicated	
Gastrointestinal Bleeding	Not requiring intervention other than iron supplement	Endoscopic intervention indicated	Transfusion indicated	Life-threatening consequences (e.g., hypotensive shock)
Mucositis or Stomatitis *Report only one and specify location*	Mucosal erythema	Patchy pseudomembranes or ulcerations	Confluent pseudomembranes or ulcerations OR Mucosal bleeding with minor trauma	Life-threatening consequences (e.g., aspiration, choking) OR Tissue necrosis OR Diffuse spontaneous mucosal bleeding
Nausea	Transient (< 24 hours) or intermittent AND No or minimal interference with oral intake	Persistent nausea resulting in decreased oral intake for 24 to 48 hours	Persistent nausea resulting in minimal oral intake for > 48 hours OR Rehydration indicated (e.g., IV fluids)	Life-threatening consequences (e.g., hypotensive shock)
Pancreatitis	NA	Symptoms with hospitalization not indicated	Symptoms with hospitalization indicated	Life-threatening consequences (e.g., circulatory failure, hemorrhage, sepsis)
Perforation (colon or rectum)	NA	NA	Intervention indicated	Life-threatening consequences
Proctitis	Rectal discomfort with no intervention indicated	Symptoms causing greater than minimal interference with usual social & functional activities OR Medical intervention indicated	Symptoms causing inability to perform usual social & functional activities OR Operative intervention indicated	Life-threatening consequences (e.g., perforation)
Rectal Discharge	Visible discharge	Discharge requiring the use of pads	NA	NA
Vomiting	Transient or intermittent AND No or minimal interference with oral intake	Frequent episodes of vomiting with no or mild dehydration	Persistent vomiting resulting in orthostatic hypotension OR Aggressive rehydration indicated (e.g., IV fluids)	Life-threatening consequences (e.g., hypotensive shock)
CLINICAL- MUSCULOSKELETAL				
Arthralgia	Joint pain causing no or minimal interference with usual social & functional activities	Joint pain causing greater than minimal interference with usual social & functional activities	Joint pain causing inability to perform usual social & functional activities	Disabling joint pain causing inability to perform basic self-care functions
Arthritis	Stiffness or joint swelling causing no or minimal interference with usual	Stiffness or joint swelling causing greater than minimal interference with	Stiffness or joint swelling causing inability to	Disabling joint stiffness or swelling causing inability

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
	social & functional activities	usual social & functional activities	perform usual social & functional activities	to perform basic self-care functions
Myalgia (generalized)	Muscle pain causing no or minimal interference with usual social & functional activities	Muscle causing greater than minimal interference with usual social & functional activities	Muscle causing inability to perform usual social & functional activities	Disabling muscle pain causing inability to perform basic self-care functions
Osteonecrosis	NA	No symptoms but with radiographic findings AND No operative intervention indicated	Bone pain with radiographic findings OR Operative intervention indicated	Disabling bone pain with radiographic findings causing inability to perform basic self-care functions
Osteopenia				
Age ≥ 30 years	BMD t-score –2.5 to –1	NA	NA	NA
Age < 30 years	BMD z-score –2 to –1	NA	NA	NA
Osteoporosis				
Age ≥ 30 years	NA	BMD t-score < -2.5	Pathologic fracture (e.g., compression fracture causing loss of vertebral height)	Pathological fracture causing life-threatening consequences
Age < 30 years	NA	BMD z-score < -2		
CLINICAL- NEUROLOGIC				
Acute CNS Ischemia	NA	NA	Transient ischemic attack	Cerebral vascular accident (e.g., stroke with neurological deficit)
Altered Mental Status (For Dementia, see *Cognitive, Behavioral, or Attentional Disturbance* below)	Changes causing no or minimal interference with usual social & functional activities	Mild lethargy or somnolence causing greater than minimal interference with usual social & functional activities	Confusion, memory impairment, lethargy, or somnolence causing inability to perform usual social & functional activities	Delirium OR Obtundation, OR Coma
Ataxia	Symptoms causing no or minimal interference with usual social & functional activities OR No symptoms with ataxia detected on examination	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	Disabling ataxia causing inability to perform basic self-care functions
Cognitive, Behavioral, Attentional Disturbance (includes dementia and attention deficit disorder)	Disability causing no or minimal interference with usual social & functional	Disability causing greater than minimal interference with usual social & functional activities OR	Disability causing inability to perform usual social & functional activities OR	Disability causing inability to perform basic self-care functions OR

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Specify type, if applicable	activities OR Specialized resources not indicated	Specialized resources on part-time basis indicated	Specialized resources on a full-time basis indicated	Institutionalization indicated
Headache	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	Symptoms causing inability to perform basic self-care functions OR Hospitalization indicated OR Headache with significant impairment of alertness or other neurologic function
Neuromuscular Weakness (includes myopathy and neuropathy) *Specify type, if applicable*	Minimal muscle weakness causing no or minimal interference with usual social & functional activities OR No symptoms with decreased strength on examination	Muscle weakness causing greater than minimal interference with usual social & functional activities	Muscle weakness causing inability to perform usual social & functional activities	Disabling muscle weakness causing inability to perform basic self-care functions OR Respiratory muscle weakness impairing ventilation
Neurosensory alteration (includes paresthesia and painful neuropathy) *Specify type, if applicable*	Minimal paresthesia causing no or minimal interference with usual social & functional activities OR No symptoms with sensory alteration on examination	Sensory alteration or paresthesia causing greater than minimal interference with usual social & functional activities	Sensory alteration or paresthesia causing inability to perform usual social & functional activities	Disabling sensory alteration or paresthesia causing inability to perform basic self-care functions
Seizure: New Onset	NA	NA	1 to 3 seizures	Prolonged and repetitive seizures (e.g., status epilepticus) OR Difficult to control (e.g., refractory epilepsy)
Seizure: Pre-existing	NA	Increased frequency from previous level of control without change in seizure character	Change in seizure character from baseline either in duration or quality (e.g., severity or focality)	Prolonged and repetitive seizures (e.g., status epilepticus) OR Difficult to control (e.g., refractory epilepsy)
Syncope	Near syncope without loss of consciousness (e.g., pre-syncope)	Loss of consciousness with no intervention indicated	Loss of consciousness AND Hospitalization or intervention required	NA
CLINICAL- PREGNANCY, PUERPERIUM, AND PERINATAL				
Stillbirth (report using mother's participant ID) *Report only one*	NA	NA	Fetal death occurring at ≥ 20 weeks gestation	NA
Preterm Birth (report using mother's participant ID)	Live birth at 34 to < 37 weeks gestational age	Live birth at 28 to < 34 weeks gestational age	Live birth at 24 to < 28 weeks gestational age	Live birth at < 24 weeks gestational age

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Spontaneous Abortion or Miscarriage (A pregnancy loss occurring at < 20 weeks gestational age, report using mother's participant ID) *Report only one*	Chemical pregnancy	Uncomplicated spontaneous abortion or miscarriage	Complicated spontaneous abortion or miscarriage	NA
CLINICAL- PSYCHIATRIC				
Insomnia	Mild difficulty falling asleep, staying asleep, or waking up early causing no or minimal interference with usual social & functional activities	Moderate difficulty falling asleep, staying asleep, or waking up early causing more than minimal interference with usual social & functional activities	Severe difficulty falling asleep, staying asleep, or waking up early causing inability to perform usual social & functional activities requiring intervention or hospitalization	NA
Psychiatric Disorders (includes anxiety, depression, mania, and psychosis) *Specify disorder*	Symptoms with intervention not indicated OR Behavior causing no or minimal interference with usual social & functional activities	Symptoms with intervention indicated OR Behavior causing greater than minimal interference with usual social & functional activities	Symptoms with hospitalization indicated OR Behavior causing inability to perform usual social & functional activities	Threatens harm to self or others OR Acute psychosis OR Behavior causing inability to perform basic self-care functions
Suicidal Ideation or Attempt *Report only one*	Preoccupied with thoughts of death AND No wish to kill oneself	Preoccupied with thoughts of death AND Wish to kill oneself with no specific plan or intent	Thoughts of killing oneself with partial or complete plans but no attempt to do so OR Hospitalization indicated	Suicide attempted
CLINICAL- RESPIRATORY				
Acute Bronchospasm	Forced expiratory volume in 1 second or peak flow reduced to ≥ 70 to < 80% OR Mild symptoms with intervention not indicated	Forced expiratory volume in 1 second or peak flow 50 to < 70% OR Symptoms with intervention indicated OR Symptoms causing greater than minimal interference with usual social & functional activities	Forced expiratory volume in 1 second or peak flow 25 to < 50% OR Symptoms causing inability to perform usual social & functional activities	Forced expiratory volume in 1 second or peak flow < 25% OR Life-threatening respiratory or hemodynamic compromise OR Intubation
Dyspnea or Respiratory Distress *Report only one*	Dyspnea on exertion with no or minimal interference with usual social & functional activities OR Wheezing OR Minimal increase in respiratory rate for age	Dyspnea on exertion causing greater than minimal interference with usual social & functional activities OR Nasal flaring OR Intercostal retractions	Dyspnea at rest causing inability to perform usual social & functional activities OR Pulse oximetry < 90%	Respiratory failure with ventilator support indicated (e.g., CPAP, BPAP, intubation)

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
		OR Pulse oximetry 90 to < 95%		
CLINICAL- SENSORY				
Hearing Loss ≥ *12 years of age*	NA	Hearing aid or intervention not indicated	Hearing aid or intervention indicated	Profound bilateral hearing loss (> 80 dB at 2 kHz and above) OR Non-serviceable hearing (i.e., >50 dB audiogram and <50% speech discrimination)
Tinnitus	Symptoms causing no or minimal interference with usual social & functional activities with intervention not indicated	Symptoms causing greater than minimal interference with usual social & functional activities with intervention indicated	Symptoms causing inability to perform usual social & functional activities	NA
Uveitis	No symptoms AND Detectable on examination	Anterior uveitis with symptoms OR Medical intervention indicated	Posterior or pan-uveitis OR Operative intervention indicated	Disabling visual loss in affected eye(s)
Vertigo	Vertigo causing no or minimal interference with usual social & functional activities	Vertigo causing greater than minimal interference with usual social & functional activities	Vertigo causing inability to perform usual social & functional activities	Disabling vertigo causing inability to perform basic self-care functions
Visual Changes (assessed from baseline)	Visual changes causing no or minimal interference with usual social & functional activities	Visual changes causing greater than minimal interference with usual social & functional activities	Visual changes causing inability to perform usual social & functional activities	Disabling visual loss in affected eye(s)
CLINICAL- SYSTEMIC				
Acute Allergic Reaction	Localized urticaria (wheals) with no medical intervention indicated	Localized urticaria with intervention indicated OR Mild angioedema with no intervention indicated	Generalized urticaria OR Angioedema with intervention indicated OR Symptoms of mild bronchospasm	Acute anaphylaxis OR Life-threatening bronchospasm OR Laryngeal edema
Chills	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	NA
Cytokine Release Syndrome (disorder characterized by nausea, headache, tachycardia, hypotension,	Mild signs and symptoms AND Therapy (i.e., antibody infusion) interruption not indicated	Therapy (i.e., antibody infusion) interruption indicated AND Responds promptly to symptomatic treatment OR Prophylactic medications indicated for ≤ 24 hours	Prolonged severe signs and symptoms OR Recurrence of symptoms following initial improvement	Life-threatening consequences (e.g., requiring pressor or ventilator support)

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
rash, and/or shortness of breath)				
Fatigue or Malaise *Report only one*	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	Incapacitating symptoms of fatigue or malaise causing inability to perform basic self-care functions
Fever (non-axillary temperatures only)	38.0 to < 38.6°C or 100.4 to < 101.5°F	≥ 38.6 to < 39.3°C or ≥ 101.5 to < 102.7°F	≥ 39.3 to < 40.0°C or ≥ 102.7 to < 104.0°F	≥ 40.0°C or ≥ 104.0°F
Pain (not associated with study agent injections and not specified elsewhere) *Specify location* (Also see Injection Site Reactions: Injection site pain)	Pain causing no or minimal interference with usual social & functional activities	Pain causing greater than minimal interference with usual social & functional activities	Pain causing inability to perform usual social & functional activities	Disabling pain causing inability to perform basic self-care functions OR Hospitalization indicated
Serum Sickness (disorder characterized by fever, arthralgia, myalgia, skin eruptions, lymphadenopathy, marked discomfort, and/or dyspnea)	Mild signs and symptoms	Moderate signs and symptoms AND Intervention indicated (e.g., antihistamines)	Severe signs and symptoms AND Higher level intervention indicated (e.g., steroids or IV fluids)	Life-threatening consequences (e.g., requiring pressor or ventilator support)
Unintentional Weight Loss (excludes postpartum weight loss)	NA	5 to < 9% loss in body weight from baseline	≥ 9 to < 20% loss in body weight from baseline	≥ 20% loss in body weight from baseline OR Aggressive intervention indicated (e.g., tube feeding, total parenteral nutrition)
CLINICAL- URINARY				
Urinary Tract Obstruction	NA	Signs or symptoms of urinary tract obstruction without hydronephrosis or renal dysfunction	Signs or symptoms of urinary tract obstruction with hydronephrosis or renal dysfunction	Obstruction causing life-threatening consequences
CLINICAL- INJECTION SITE REACTIONS				
Injection Site Pain or Tenderness *Report only one*	Pain or tenderness causing no or minimal limitation of use of limb	Pain or tenderness causing greater than minimal limitation of use of limb	Pain or tenderness causing inability to perform usual social & functional activities	Pain or tenderness causing inability to perform basic self-care function OR Hospitalization indicated
Injection Site Erythema or Redness OR Injection	2.5 to < 5 cm in diameter OR 6.25 to < 25 cm^2 surface area AND	≥ 5 to < 10 cm in diameter OR ≥ 25 to < 100 cm^2 surface area OR	≥ 10 cm in diameter OR ≥ 100 cm^2 surface area OR Ulceration OR Secondary	Potentially life-threatening consequences (e.g., abscess, exfoliative

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Site Induration or Swelling *Report only one* *> 15 years of age*	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	infection OR Phlebitis OR Sterile abscess OR Drainage OR Symptoms causing inability to perform usual social & functional activities	dermatitis, necrosis involving dermis or deeper tissue)
Injection Site Pruritus	Itching localized to the injection site that is relieved spontaneously or in < 48 hours of treatment	Itching beyond the injection site that is not generalized OR Itching localized to the injection site requiring ≥ 48 hours treatment	Generalized itching causing inability to perform usual social & functional activities	NA
LABORATORY- CHEMISTRY	***Standard International Units are listed in Italics***			
Acidosis	NA	pH ≥ 7.3 to < LLN	pH < 7.3 without life-threatening consequences	pH < 7.3 with life-threatening consequences
Albumin, Low (g/dL; *g/L*)	3.0 to < LLN *30 to < LLN*	≥ 2.0 to < 3.0 *≥ 20 to < 30*	< 2.0 *< 20*	NA
Alkaline Phosphatase, High	1.25 to < 2.5 x ULN	2.5 to < 5.0 x ULN	5.0 to < 10.0 x ULN	≥ 10.0 x ULN
Alkalosis	NA	pH > ULN to ≤ 7.5	pH > 7.5 without life-threatening consequences	pH > 7.5 with life-threatening consequences
ALT or SGPT, High *Report only one*	1.25 to < 2.5 x ULN	2.5 to < 5.0 x ULN	5.0 to < 10.0 x ULN	≥ 10.0 x ULN
Amylase (Pancreatic) or Amylase (Total), High *Report only one*	1.1 to < 1.5 x ULN	1.5 to < 3.0 x ULN	3.0 to < 5.0 x ULN	≥ 5.0 x ULN
AST or SGOT, High *Report only one*	1.25 to < 2.5 x ULN	2.5 to < 5.0 x ULN	5.0 to < 10.0 x ULN	≥ 10.0 x ULN
Bicarbonate, Low (mEq/L; *mmol/L*)	16.0 to < LLN *16.0 to < LLN*	11.0 to < 16.0 *11.0 to < 16.0*	8.0 to < 11.0 *8.0 to < 11.0*	< 8.0 *< 8.0*
Bilirubin: *Direct Bilirubin, High*	NA	NA	> ULN with other signs and symptoms of hepatotoxicity.	> ULN with life-threatening consequences (e.g., signs and symptoms of liver failure)
Bilirubin: *Total Bilirubin, High*	1.1 to < 1.6 x ULN	1.6 to < 2.6 x ULN	2.6 to < 5.0 x ULN	≥ 5.0 x ULN
Calcium, High (mg/dL; *mmol/L*)	10.6 to < 11.5 *2.65 to < 2.88*	11.5 to < 12.5 *2.88 to < 3.13*	12.5 to < 13.5 *3.13 to < 3.38*	≥ 13.5 *≥ 3.38*
Calcium (Ionized), High (mg/dL; *mmol/L*)	> ULN to < 6.0 *> ULN to < 1.5*	6.0 to < 6.4 *1.5 to < 1.6*	6.4 to < 7.2 *1.6 to < 1.8*	≥ 7.2 *≥ 1.8*
Calcium, Low (mg/dL; *mmol/L*)	7.8 to < 8.4 *1.95 to < 2.10*	7.0 to < 7.8 *1.75 to < 1.95*	6.1 to < 7.0 *1.53 to < 1.75*	< 6.1 *< 1.53*
Calcium (Ionized), Low (mg/dL; *mmol/L*)	< LLN to 4.0 *< LLN to 1.0*	3.6 to < 4.0 *0.9 to < 1.0*	3.2 to < 3.6 *0.8 to < 0.9*	< 3.2 *< 0.8*
Cardiac Troponin I, High	NA	NA	NA	Levels consistent with myocardial infarction or unstable angina as defined by the local laboratory
Creatine Kinase, High	3 to < 6 x ULN	6 to < 10x ULN	10 to < 20 x ULN	≥ 20 x ULN
Creatinine, High **Report only one*	1.1 to 1.3 x ULN	> 1.3 to 1.8 x ULN OR Increase to 1.3 to < 1.5 x participant's baseline	> 1.8 to < 3.5 x ULN OR Increase to 1.5 to < 2.0 x participant's baseline	≥ 3.5 x ULN OR Increase of ≥ 2.0 x participant's baseline

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Creatinine Clearance or eGFR, Low **Report only one* (i.e., Cockcroft-Gault in mL/min or Schwartz, MDRD, CKD-Epi in mL/min/1.73 m^2)	NA	< 90 to 60 ml/min or ml/min/1.73 m2 OR 10 to < 30% decrease from participant's baseline	< 60 to 30 ml/min or ml/min/1.73 m2 OR 30 to < 50% decrease from participant's baseline	< 30 ml/min or ml/min/1.73 m2 OR ≥ 50% decrease from participant's baseline or dialysis needed
Glucose (mg/dL; *mmol/L*) ***Fasting, High***	110 to 125 *6.11 to < 6.95*	> 125 to 250 *6.95 to < 13.89*	> 250 to 500 *13.89 to < 27.75*	≥ 500 *≥ 27.75*
Glucose (mg/dL; *mmol/L*) ***Nonfasting, High***	116 to 160 *6.44 to < 8.89*	> 160 to 250 *8.89 to < 13.89*	> 250 to 500 *13.89 to < 27.75*	≥ 500 *≥ 27.75*
Glucose, Low (mg/dL; *mmol/L*)	55 to 64 *3.05 to <3.55*	40 to < 55 *2.22 to < 3.05*	30 to < 40 *1.67 to < 2.22*	< 30 *< 1.67*
Lactate, High	ULN to < 2.0 x ULN without acidosis	≥ 2.0 x ULN without acidosis	Increased lactate with pH < 7.3 without life-threatening consequences	Increased lactate with pH < 7.3 with life-threatening consequences
Lipase, High	1.1 to < 1.5 x ULN	1.5 to < 3.0 x ULN	3.0 to < 5.0 x ULN	≥ 5.0 x ULN
Lipids Disorders: ***Cholesterol, Fasting,*** / ***High*** (mg dL; *mmol/L*)	200 to < 240 *5.18 to < 6.19*	240 to < 300 *6.19 to < 7.77*	≥ 300 *≥ 7.77*	NA
Lipids Disorders: *LDL, Fasting, High* (mg dL; *mmol/L*)	130 to < 160 *3.37 to < 4.12*	160 to < 190 *4.12 to < 4.90*	≥ 190 *≥ 4.90*	NA
Lipids Disorders: ***Triglycerides, Fasting, High*** (mg dL; *mmol/L*)	150 to 300 *1.71 to 3.42*	>300 to 500 *>3.42 to 5.7*	>500 to < 1,000 *>5.7 to 11.4*	> 1,000 *> 11.4*
Magnesium, Low (mEq/L; *mmol/L*) (To convert a magnesium value from mg/dL to mmol/L, laboratories should multiply by 0.4114)	1.2 to < 1.4 *0.60 to < 0.70*	0.9 to < 1.2 *0.45 to < 0.60*	0.6 to < 0.9 *0.30 to < 0.45*	< 0.6 *< 0.30*
Phosphate, Low (mg/dL; *mmol/L*)	2.0 to < LLN *0.65 to < LLN*	1.4 to < 2.0 *0.45 to < 0.65*	1.0 to < 1.4 *0.32 to < 0.45*	< 1.0 *< 0.32*
Potassium, High (mEq/L; *mmol/L*)	5.6 to < 6.0 *5.6 to < 6.0*	6.0 to < 6.5 *6.0 to < 6.5*	6.5 to < 7.0 *6.5 to < 7.0*	≥ 7.0 *≥ 7.0*
Potassium, Low (mEq/L; *mmol/L*)	3.0 to < 3.4 *3.0 to < 3.4*	2.5 to < 3.0 *2.5 to < 3.0*	2.0 to < 2.5 *2.0 to < 2.5*	< 2.0 *< 2.0*
Sodium, High (mEq/L; *mmol/L*)	146 to < 150 *146 to < 150*	150 to < 154 *150 to < 154*	154 to < 160 *154 to < 160*	≥ 160 *≥ 160*
Sodium, Low (mEq/L; *mmol/L*)	130 to < 135 *130 to < 135*	125 to < 130 *125 to < 130*	121 to < 125 *121 to < 125*	≤ 120 *≤ 120*
Uric Acid, High (mg/dL; *mmol/L*)	7.5 to < 10.0 *0.45 to < 0.59*	10.0 to < 12.0 *0.59 to < 0.71*	12.0 to < 15.0 *0.71 to < 0.89*	≥ 15.0 *≥ 0.89*
LABORATORY- HEMATOLOGY	***Standard International Units are listed in Italics***			
Absolute CD4+ Count, Low (cell/mm3; *cells/L*) *(not HIV infected)*	300 to < 400 *300 to < 400*	200 to < 300 *200 to < 300*	100 to < 200 *100 to < 200*	< 100 *< 100*
Absolute Lymphocyte Count, Low (cell/mm3; *cells/L*) *(not HIV infected)*	600 to < 650 *0.600 x 10^9 to < 0.650 x 10^9*	500 to < 600 *0.500 x 10^9 to < 0.600 x 10^9*	350 to < 500 *0.350 x 10^9 to < 0.500 x 10^9*	< 350 *< 0.350 x 10^9*

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Absolute Neutrophil Count (ANC), Low (cells/mm3; *cells/L*)	800 to 1,000 *0.800 x 10^9 to 1.000 x 10^9*	600 to 799 *0.600 x 10^9 to 0.799 x 10^9*	400 to 599 *0.400 x 10^9 to 0.599 x 10^9*	< 400 *< 0.400 x 10^9*
Fibrinogen, Decreased (mg/dL; *g/L*)	100 to < 200 *1.00 to < 2.00* OR 0.75 to < 1.00 x LLN	75 to < 100 *0.75 to < 1.00* OR ≥ 0.50 to < 0.75 x LLN	50 to < 75 *0.50 to < 0.75* OR 0.25 to < 0.50 x LLN	< 50 *< 0.50* OR < 0.25 x LLN OR Associated with gross bleeding
Hemoglobin, Low (g/dL; *mmol/L*) *(male only)*	10.0 to 10.9 *6.19 to 6.76*	9.0 to < 10.0 *5.57 to < 6.19*	7.0 to < 9.0 *4.34 to < 5.57*	< 7.0 *< 4.34*
Hemoglobin, Low (g/dL; *mmol/L*) *(female only)*	9.5 to 10.4 *5.88 to 6.48*	8.5 to < 9.5 *5.25 to < 5.88*	6.5 to < 8.5 *4.03 to < 5.25*	< 6.5 *< 4.03*
INR, High (not on anticoagulation therapy)	1.1 to < 1.5 x ULN	1.5 to < 2.0 x ULN	2.0 to < 3.0 x ULN	≥ 3.0 x ULN
Methemoglobin (% hemoglobin)	5.0 to < 10.0%	10.0 to < 15.0%	15.0 to < 20.0%	≥ 20.0%
PTT, High (not on anticoagulation therapy)	1.1 to < 1.66 x ULN	1.66 to < 2.33 x ULN	2.33 to < 3.00 x ULN	≥ 3.00 x ULN
Platelets, Decreased (cells/mm^3; *cells/L*)	100,000 to < 125,000 *100.000 x 10^9 to < 125.000 x 10^9*	50,000 to < 100,000 *50.000 x 10^9 to < 100.000 x 10^9*	25,000 to < 50,000 *25.000 x 10^9 to < 50.000 x 10^9*	< 25,000 *< 25.000 x 10^9*
PT, High (not on anticoagulation therapy	1.1 to < 1.25 x ULN	1.25 to < 1.50 x ULN	1.50 to < 3.00 x ULN	≥ 3.00 x ULN
WBC, Decreased (cells/mm3; *cells/L*)	2,000 to 2,499 *2.000 x 10^9 to 2.499 x 10^9*	1,500 to 1,999 *1.500 x 10^9 to 1.999 x 10^9*	1,000 to 1,499 *1.000 x 10^9 to 1.499 x 10^9*	< 1,000 *< 1.000 x 10^9*
LABORATORY- URINALYSIS	***Standard International Units are listed in Italics***			
Glycosuria (random collection tested by dipstick)	Trace to 1+ or ≤ 250 mg	2+ or > 250 to ≤ 500 mg	> 2+ or > 500 mg	NA
Hematuria (not to be reported based on dipstick findings or on blood believed to be of menstrual origin)	6 to < 10 RBCs per high power field	≥ 10 RBCs per high power field	Gross, with or without clots OR With RBC casts OR Intervention indicated	Life-threatening consequences
Proteinuria (random collection tested by dipstick)	1+	2+	3+ or higher	NA

23.4. Appendix IV: List of Essential Documents for the Investigative Site

1. Investigator's Brochure and Updates.
2. Signed protocol (all versions) and amendments, if any, and sample EDC forms.
3. Information given to trial subject:

a. Informed consent form (all versions), any other written information, advertisement for subject recruitment (if used).

4. Financial agreement between the Investigator/institution and the Sponsor for the trial.
5. Signed agreements between involved parties:
    a. Financial Disclosure of Investigator/institution and sponsor.
    b. Confidential Disclosure Agreement of Investigator/institution and Sponsor.
6. Dated, documented approval/favorable opinion of IRB of the following:
    a. Protocol and any amendments.
    b. EDC system (if applicable).
    c. Informed consent form and any revisions.
    d. Any other written information to be provided to other subjects.
    e. Advertisement for subject recruitment (if used).
    f. Subject compensation (if any).
    g. Any other documents given approval.
    h. Continuing review of the trial.
7. Institutional Review Board composition.
8. Regulatory notice of Principal Investigator and sub-Investigators, FDA Form 1572.
9. Curriculum vitae and/or other relevant documents evidencing qualifications of Investigator and sub-Investigators.
10. Normal values/ranges and updates for medical/ laboratory/technical procedures and/or tests included in the protocol.
11. Medical/laboratory technical procedures/tests and updates:
    a. Certification or
    b. Accreditation or
    c. Established quality control and/or external quality assessment or
    d. Other validation (where required).
12. Shipping records for investigational product(s) and trial-related materials.
13. Site initiation monitoring report.
14. Relevant communications other than site visits:
    a. Letters/emails.
    b. Meeting notes.
    c. Notes of telephone calls.
15. Signed informed consent forms.
16. Source documents.
17. Signed, dated, and completed EDC forms to include documentation of EDC form corrections.
18. Notification by originating Investigator to Sponsor of serious adverse events and related reports.

19. Notification by Investigator, where applicable, to IRB of unexpected serious adverse drug reactions and of other safety information.
20. Notification by Sponsor to Investigator of safety information.
21. Interim or annual reports to IRB.
22. Subject screening log.
23. Subject identification code list.
24. Subject enrollment log.
25. Investigational product accountability records (receipt, storage, dispensing, shipment).
26. Signature sheet.
27. Record of retained body fluids/tissue samples (if any).
28. Final report by Investigator/institution to IRB.

23.5. Appendix V: Protocol Revision History

Table 6: Protocol Revision History

Summary of Changes	
Amendment 1	
Purpose:	Administrative update to remove vendor names, clarify study medication packaging and labeling, and remove redundant text.
Change 1:	Title Page, added statement of compliance
Change 2:	Section 5, *Randomization* Deleted: "([redacted])"
Change 3:	Section 6, *Study Procedures* Deleted below text due to redundancy with section 13. "Conduct of the study: All aspects of the study will be carefully monitored by qualified individuals designated by the principal investigator. Monitoring will be conducted according to Good Clinical Practice (GCP) standard operating procedures for compliance with applicable government regulations. The individuals responsible for monitoring the trial will have access to all records necessary to ensure the integrity of the recorded data will periodically review the progress of the study. Study Visits:"
Change 4:	Section 7.1, *Electronic Data Entry* Changed text from: "[redacted] will provide the electronic data collection (EDC) system, [redacted] is a source data collection solution that will allow the site staff to record subject data on electronic protocol-driven source forms by using tablets provided by [redacted] Its purpose is to eliminate the need for data transcription into a traditional electronic case report form, and to allow both real-time access to source data by authorized Sponsor personnel and real-time edit checks.

	[redacted] is the [redacted] web portal. It delivers real-time access to source data, and enables authorized Sponsor personnel to instantly review, clean and analyze study data. The site staff will use [redacted] as they meet with study subjects and perform study procedures. Computerized data cleaning checks will be used in addition to manual review, including listings review, to check for discrepancies and to ensure consistency and completeness of the data. If the [redacted] system is not available for any reason, source data may be captured on paper and transcribed to the [redacted] system at a later time. The responsible study monitor(s) will verify data, which can be performed remotely or at the clinical study site. The Investigator will ensure that the data recorded are accurate and complete. Queries emerging during data cleaning will be generated by data management or clinical research associates in [redacted]. The Investigator or his/her designee will answer the queries and update the source data, if needed. " to "Data will be transcribed from source documents into an electronic data capture (EDC) system. The responsible study monitor(s) will verify data, which can be performed remotely or at the clinical study site. The Investigator will ensure that the data recorded are accurate and complete. Queries emerging during data cleaning will be generated within the EDC system by data management or clinical research associates. The Investigator or his/her designee will answer the queries and update the source data, if needed." Deleted: "All listings, summaries and analyses will be produced using SAS Statistical Software (SAS Institute, Inc., Cary, NC)."
Change 5:	Section 9.2, *Packaging and Labeling* Changed text from: "Monthly supplies of medication will be packaged for each patient in unit-dosed blister cards. The blister cards will be stored in a controlled environment of 2°C to 25°C with excursions up to 40°C allowed. Each blister card will bear a label with the following information: Investigator name Patient number Week XX - XX Number of tablets Instructions for dosing Protocol number Caution: New Drug – Limited by Federal Law to Investigational Use DO NOT USE BEYOND END OF STUDY KEEP OUT OF REACH OF CHILDREN Instructions for storage Address of the Sponsor" to: "NTZ and placebo tablets will packaged in white 30cc HDPE bottles, each containing 30 tablets. The bottles will be stored at room temperature and will bear a label with the following information: Investigator name Patient number Week XX - XX 30 tablets Take 3 tablets orally with food in the morning and in the evening RM08-2001 Caution: New Drug – Limited by Federal Law to Investigational Use

	DO NOT USE BEYOND END OF STUDY KEEP OUT OF REACH OF CHILDREN Store at room temperature Study Sponsor: The Romark Institute for Medical Research [redacted]
Change 6:	Section 12, *Electronic Data Collection (EDC) System* Deleted section because of redundancy with Section 7.1. Subsequent section numbering was updated. Original text was: "12. ELECTRONIC DATA COLLECTION (EDC) SYSTEM An EDC system will be used for this study [redacted] Prior to study initiation, site staff and authorized Romark personnel will be trained to use this system. All electronic source forms are to be filled out completely by the examining site staff and reviewed and signed off on by the Investigator(s). "
Change 7:	Section 20.1, *Schedule of Assessments* Updated for consistency with section 6.0, Study Procedures
Change 8:	Added Appendix 20.4 Protocol Revision History. Subsequent section numbering was updated.
Action:	A revised protocol version 2.0 dated April 10, 2019 was generated
Amendment 2	
Purpose:	Incorporating FDA's recommended updates to the protocol and other minor changes.
Change 1:	Section 12.2: Investigational Product label text is revised. Storage conditions revised to specify upper limit of 25ºC and protection from moisture to comply with requirements for Singapore. Text added to specify that an information card will be provided to each subject with contact information for questions.
Change 2	GLOBAL: Baseline was specified as Day 1 rather than Day 0.
Change 3	Section 6.0, Section 9.2 (new item #7), Section 9.3 (Item #7) and Table 4 (Schedule of Assessments): Revised to add collection of blood samples for trough (just prior to dosing) plasma levels of NrtI and tizoxanide at Baseline and visits throughout the study.
Change 4	Section 9.2, Section 9.3, Section 9.4, Section 9.5 and Table 4: At the discretion of the investigator, subjects at high risk for contracting HIV may have repeat testing during the course of the trial.
Change 5	Section 9.2 (Item #9), Section 9.3 (Item #9) and Table 4 (Schedule of Assessments) and Footnote #7: A 12-lead ECG was added at Baseline, Week 12 and end of treatment.
Change 6	Section 9.1 (Item #6), Section 9.2 (Item #6), Section 9.3 (Item #6), Section 9.4 (Item #5) and Section 9.5 (Item #5): Creatine phosphokinase was added to clinical laboratory testing.
Change 7	Section 9.6, (Item #3): FDA recommended AEs requiring subject discontinuation were added.
Change 8	Section 4.2.2.: Revised to include clinical and safety data from literature publications regarding treatment of CHB with NTZ. Citations were added to Section 22.
Change 9	Section 9.8, revised to state: "…via a chemiluminescent immunoassay." instead of "…determined using the ARCHITECT HBsAg assay by Abbott."
Change 10	Section 21: Added the contact information of the Contract Research Organization monitoring the study.
Change 11	Section 9.3: Revised the study visit windows.
Change 12	Section 9.2, added: "The first dose of study medication should be taken in the clinic under the observation of the investigator."
Change 13	Section 7.2 (Item #17), added: "Subjects taking medications considered major CYP2C8 substrates, see Appendix I".
Change 14	Added Appendix I and Table 3 "Major CYP2C8 Substrates" and updated the appendix and table numbers throughout the protocol.

Change 15	Section 12.4, updated title to read: "Subject Compliance and Disallowed Medications" and added the following: "During the active treatment course of the study, concomitant use of medications considered major CYP2C8 substrates (Appendix I) are prohibited. Subjects are also prohibited from taking any other investigational medication during the study."
Change 16	Section 9.1 (Item #7), Section 9.2 (Item #8), Section 9.3 (Item #8), Section 9.4 (Item #6) and Section 9.5 (Item #6): Urine appearance was added to clinical laboratory testing.
Action:	A revised protocol version 3.0 dated June 13, 2019 was generated.
Amendment 3	
Purpose:	Revise protocol defined stopping rules, and other minor changes.
Change 1	Section 1, section 6, section 9.6 (item #4): Revised stopping rules.
Change 2	Section 1: Added study center and screening period.
Change 3	Section 2: Updated the study schema.
Change 4	Section 4.2.2.2: Changed "…treated with 500 mg nitazoxanide only once daily…" to "…treated with 500 mg NTZ only once daily…". Changed "…reviewed by Rossignol and Keeffe in 2009" to "…reviewed by Rossignol and Keeffe in 2008"
Change 5	Section 8: Added, "If a subject is lost to follow up or discontinues the study prior to the assessment at week 12, an additional subject may be enrolled in the same treatment group."
Change 6	Section 9.1 (Item #5): Changed HBV DNA to qHBV DNA and HBsAG to qHBsAg.
Change 7	Section 9.1 (Item #5), section 9.2 (Item #4): Changed anti-HBs to qAnti-HBs.
Change 8	Section 9.2 (Item #11): Removed "…for 12 weeks…" from the Group 3 and 4 dosing instructions and added "…with food…" to the Group 2 dosing instructions.
Change 9	Section 12.3: Added the following italicized language, "…every 4 *to 12* weeks during the treatment period*, depending on the visit schedule*."
Change 10	Section 21: Removed [redacted] as second Medical Monitor.
Change 11	Section 23.3: Added "(Table 5)" and removed "…on the following pages". Added "Table 5" and removed "…the next page".
Change 12	Amendment 2 Summary of Changes (Item #3): Changed "…Baseline and Week 1" to "…Baseline and visits throughout the study".
Action:	A revised protocol version 4.0 dated January 2, 2020 was generated.