A Phase II Open-Label Study of Ledipasvir/Sofosbuvir for 12 Weeks in Subjects with Hepatitis B Virus Infection (APOSTLE)

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STATEMENT OF COMPLIANCE

The trial will be carried out in accordance with International Conference on Harmonisation Good Clinical Practice (ICH GCP) and the following:

- United States (US) Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46, 21 CFR Part 50, 21 CFR Part 56, 21 CFR Part 312, and/or 21 CFR Part 812)

The protocol, informed consent form, recruitment materials, and all participant materials will be submitted to the Institutional Review Board (IRB) for review and approval. Approval of both the protocol and the consent form must be obtained before any participant is enrolled. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented to the study. In addition, all changes to the consent form will be IRB-approved; a determination will be made regarding whether a new consent needs to be obtained from participants who provided consent, using a previously approved consent form.

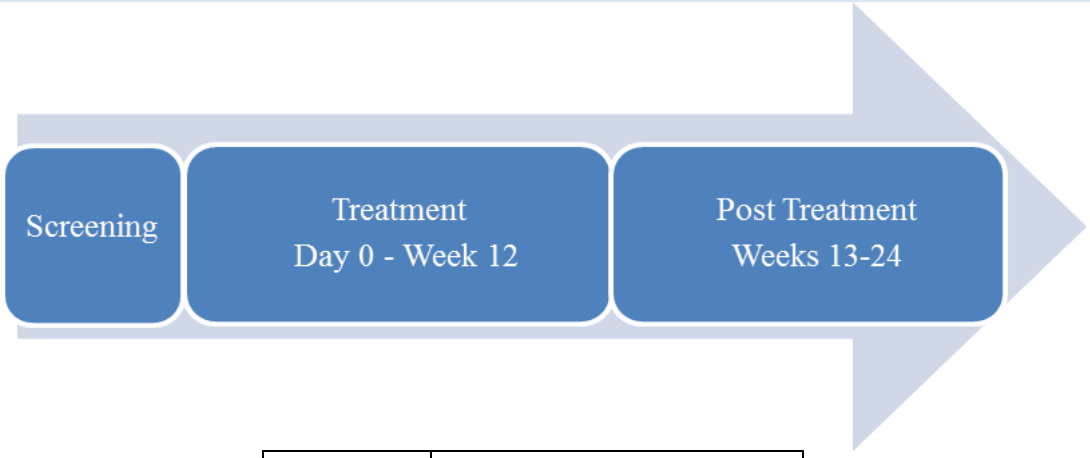
1 PROTOCOL SUMMARY

1.1 SYNOPSIS

Title:	A Phase II Open-Label Study of Ledipasvir/Sofosbuvir for 12 Weeks in Subjects with Hepatitis B Virus Infection (APOSTLE)
Study Description:	Chronic hepatitis B affects approximately 350 million people worldwide. Chronic hepatitis B (CHB) infection leads to progressive liver disease, liver failure and liver cancer. Treatment for CHB is aimed to suppress hepatitis B viral loads (HBV DNA levels) using nucleoside analog treatment, such as tenofovir disoproxil fumarate (TDF) and recently tenofovir alafenamide fumarate (TAF). Suppressive therapy is associated with regression of liver fibrosis and reduced liver related outcomes, however, has to be continued lifelong. Conversely, for chronic hepatitis C, with advent of direct acting antivirals (DAAs) such as ledipasvir/sofosbuvir (LDV/SOF) or Harvoni® functional cure is now easily attainable. This study is aimed at evaluating the antiviral activity of LDV and SOF either in fixed dose combination (FDC) or as monotherapy in subjects infected with HBV, as well as its safety and tolerability. Additionally, the relationship between LDV/SOF pharmacodynamics and HBV parameters will be explored.
Objectives:	**Primary Objectives:** To evaluate the change of serum hepatitis B surface antigen (HBsAg $\log_{10}$ IU/mL) as an indicator of antiviral activity of ledipasvir and/or sofosbuvir in subjects with chronic hepatitis B from baseline to end of 12 weeks of treatment. To evaluate the safety and tolerability of the treatment regimen as assessed by review of the accumulated safety data. **Secondary Objective**: To evaluate the kinetics of circulating HBV DNA in subjects who are infected with HBV from baseline to end of 12 weeks of treatment. **Exploratory Objectives**: To explore the relationship between pharmacodynamics biomarkers and viral parameters.

Endpoints:	**Primary**: HBsAg quantitation at end of 12 weeks treatment. Reports of Grade 3 or 4 clinical or laboratory adverse events. **Secondary**: HBV DNA at end of 12 weeks treatment.
Study Population:	Adults affected with chronic viral hepatitis B (HBV), who are either in low replicative state not requiring treatment or are virally suppressed on HBV medications.
Phase:	2
Description of Sites:	Conducted by the University of Maryland, Institute of Human Virology (IHV) at the IHV Clinical Research Unit (CRU), with Collaborating Sites at the Chinese Culture and Community Service Center, Inc. (CCACC) and Unity Health Care Inc., Parkside.
Description of Study Intervention:	This study will involve 4 Dosing Groups: **Group A**: 10 chronic HBV subjects with chronic HBV in low replicate state, not on antivirals and not requiring therapy, will receive LDV/SOF once daily for 12 weeks. **Group B:** 5 virally suppressed chronic HBV subjects on treatment will receive LDV/SOF once daily for 12 weeks. **Groups C & D**: 10 subjects with chronic HBV in low replicative state, not on antivirals and not requiring therapy, will be randomized in a 1:1 ratio to receive **either** SOF 400 mg (**Group C**) **OR** LDV 90 mg once daily (**Group D**) for 12 weeks.
Study Duration:	Twenty month enrollment, eight month study participation, four months data analysis.
Participant Duration:	Subject participation may be up to eight months in duration.

1.2 SCHEMA



SCR	*Screening*
D0	*Day 0*
W1	*Week 1*
W2	*Week 2*
W4	*Week 4*
W8	*Week 8*
W12/EOT	*Week 12 / End of Treatment*
Week 13	*Post Treatment Week 1*
Week 16	*Post Treatment Week 4*
Week 24	*Post Treatment Week 12*

1.3 SCHEDULES OF ACTIVITIES (SOA)

GROUPS A & B: Subjects receiving 12 weeks of Ledipasvir/Sofosbuvir

Study Visit	SCR -60 to -28	D0	W1 (+/-3 days)	W2 (+/-3 days)	W4 (+/-3 days)	W8 (+/-3 days)	W12 / EOT (+/-7 days)	Week 13 (+/-7 days)	Week 16 (+/-7 days)	Week 24 (+/-7 days)
Informed Consent	X	X								
Demographics	X									
Medical History	X	X								
Concomitant Medication Review	X	X……								X
Dispense Study Medication		X			X	X				
Complete Physical Examination	X	X					X			
Directed Physical Examination			X	X	X	X		X		X
Height & Weight	X									
Vital Signs	X	X	X	X	X	X	X	X	X	X
Adverse Event monitoring		X……								X
Electrocardiogram	X									
Hematology[a]	X*	X	X	X	X	X	X		X	
Chemistry[b]	X*	X	X	X	X	X	X	X	X	X
Lipase and creatine kinase[#]		X	X	X	X	X	X	X	X	X
Coagulation[c]	X*	X			X		X			X
HgbA1c	X***									
Serologies[d]	X*									
HBV Genotyping (if not already on file, or if risk behavior identified)		X								
HBsAg, AntiHBs	X**	X	X	X	X	X	X	X	X	X
Quantitative HBV DNA	X*	X	X	X	X	X	X	X	X	X
FibroSure	X***									
Urinalysis	X									
Urine Drug screen	X									
Urine Pregnancy Test[e]	X	X		X	X	X	X		X	
Quantitative HBsAg	X	X	X	X	X	X	X	X	X	X
Quantitative HBcrAg & HBV RNA	X	X	X	X	X	X	X	X	X	X
Research Storage Labs[f]		X	X	X	X	X	X	X	X	X
Pax DNA		X								
Pax RNA		X		X		X				X

* May use outside labs within the last 30 days (or any historical hepatitis Delta result)
** May use outside labs within the last 90 days
*** May use outside labs within the last 12 months
[a]Complete blood count with differential
[b]Complete metabolic panel: ALT, AST, albumin, alkaline phosphatase, total bilirubin, creatinine, calcium, carbon dioxide, chloride, eGFR calculation, globulin, glucose, potassium, protein, sodium
[c]Prothrombin time and International Normalised Ratio (INR)
[d]Anti-HCV, Total antibody hepatitis delta, Human immunodeficiency virus screening, HBeAg, antiHBe
[e]For women of childbearing potential
[f]Serum storage, plasma storage, and PBMC storage
[#] For Group B

GROUPS C and D: Subjects receiving 12 weeks of monotherapy of either Sofosbuvir or Ledipasvir

Study Visit	SCR (60 day window)	D0	W1# (+/-3 days)	W2# (+/-3 days)	W4 (+/-3 days)	W8# (+/-3 days)	W12 / EOT (+/-7 days)	Week 13# (+/-7 days)	Week 16 (+/-7 days)	Week 24 (+/-7 days)
Informed Consent	X	X								
Demographics	X									
Medical History	X	X								
Concomitant Medication Review	X	X……X								
Randomization		X								
Dispense Study Medication		X			X	X#				
Complete Physical Examination	X	X					X			
Directed Physical Examination			X#	X#	X	X#		X#		X
Height & Weight	X									
Vital Signs	X	X	X#	X#	X	X	X	X#	X	X
Adverse Event monitoring	X	X	X#	X#	X	X	X	X#	X	X
Electrocardiogram	X									
FibroScan (Transient Elastography)		X					X			
Hematology[a]	X*	X	X	X	X	X	X		X	
Chemistry[b]	X*	X	X	X	X	X	X	X	X	X
Lipase and creatine kinase		X	X	X	X	X	X	X	X	X
Coagulation[c]	X*	X			X		X			X
HgbA1c	X***									
Serologies[d]	X*									
HBV Genotyping (if not already on file, or if risk behavior identified)		X								
HBsAg, AntiHBs	X**	X	X	X	X	X	X	X	X	X
Quantitative HBV DNA	X**	X	X	X	X	X	X	X	X	X
FibroSure	X***									
Urinalysis	X									
Urine Drug screen	X									
Urine Pregnancy Test[e]	X	X		X	X	X	X		X	
Quantitative HBsAg	X	X	X	X	X	X	X	X	X	X
Quantitative HBcrAg & HBV RNA	X	X	X	X	X	X	X	X	X	X
Research Storage Labs[f]		X	X	X	X	X	X	X	X	X
Pax DNA		X								
Pax RNA		X		X		X				X

* May use outside labs within the last 30 days (or any historical hepatitis Delta result) or 90 days for those previously completing Group A

** May use outside labs within the last 90 days

*** May use outside labs within the last 12 months

Those previously treated on Group A (tolerated LDV/SOF x 12 weeks) will have lab visits only (with dispensing and medication adherence week 8)

[a]Complete blood count with differential

[b]Complete metabolic panel: ALT, AST, albumin, alkaline phosphatase, total bilirubin, creatinine, calcium, carbon dioxide, chloride, eGFR calculation, globulin, glucose, potassium, protein, sodium

[c]Prothrombin time and International Normalised Ratio (INR)

[d]Anti-HCV, Total antibody hepatitis delta, Human immunodeficiency virus screening, HBeAg, antiHBe

[e]For women of childbearing potential

[f]Serum storage, plasma storage, and PBMC storage

2 INTRODUCTION

2.1 STUDY RATIONALE

Chronic HBV (CHB) infection is endemic in several countries, with very high prevalence in certain areas of Asia and Africa. There are over half a million immigrants from Asia and Africa in the Baltimore-metropolitan DC area [1] this, combined with the fact that Baltimore and Washington DC are among the highest for estimated prevalence of IDUs in 96 large U.S. metropolitan areas [2] means that there is a unmet need for hepatitis B (HBV) care and studies to advance HBV treatment research in this part of the country.

Hepatitis B surface antigen (HBsAg) loss is the current accepted endpoint for anti-HBV therapy and has been associated with improvements in liver histology, including the reversal of cirrhosis, a decreased risk of hepatocellular carcinoma (HCC), and prolonged survival. Recent treatment guidelines have acknowledged the importance of HBsAg clearance in CHB [3, 4]. An emerging theme is that HBsAg clearance is associated with definitive remission of the activity of CHB and an improved long term outcome [4]. Recent data show that the risk of HCC is lower if HBsAg clearance occurs before 50 years of age [5]. Loss of HBsAg is thus a primary goal of CHB therapy and is synonymous to a functional HBV cure.

Nucleos(t)ide analogues (NA) are the current standard of care for CHB, providing durable on-treatment suppression of viral replication and resulting in long-term clinical benefits with a reduced risk of liver complications. Although NAs are highly effective in suppressing HBV viral replication, the incidence of HBsAg loss during NA therapy is cumulative but low (ranging from 0-3% in the first year of NA therapy) [6]. As a result, continuous long-term use is recommended for most patients. Moreover, NA therapy rarely results in HBsAg seroconversion [7]. Therefore, new therapies that that enhance rates of HBsAg loss and subsequent seroconversion after a finite treatment course are needed.

2.2 BACKGROUND

Hepatitis B virus (HBV) chronically infects over 350 million people worldwide and over one million Americans. HBV is transmitted sexually, perinatally and percutaneously [6, 8-9]. The risk of developing chronic HBV after exposure is inversely related to difference in age at infection, ranges from 90% in infants infected via vertical transmission to less than 5% in immunocompetent adults [10]. Therefore, in areas where vertical transmission predominates, the prevalence of chronic disease is significantly greater than in areas in which HBV is transmitted among adults, primarily through sex and intravenous drug use. In the United States, since the advent of universal infant vaccination for HBV in 1991, there have been great reductions in acute HBV and chronic HBV, the reductions however have been offset by substantial migration of persons already chronically infected with HBV in the past 40 years, especially from the endemic region such as Eastern Asia and Sub-Saharan Africa [11]. According to National Health and Nutrition Examination Survey (NHANES), during 2007-2012, among the estimated 199,000 Non-Hispanic blacks with CHB, prevalence of CHB infection was 2.5% among foreign-born non-Hispanic blacks; Correspondingly, during 2011-2012, among the estimated 427,000 non-Hispanic Asians with CHB infection, 93.1% were foreign born [12].

Hepatitis B in Baltimore and Washington DC

Baltimore has been cited as a leading city in terms of injection drug use, ranking second in the nation for intravenous drug use (IDU) per capita in 2004 [13]. A study in early 2000 looking at injection drug users all with chronic HCV in Baltimore, occult HBV was found in 45% of individuals [14]. A more recent study looking at patterns of drug use and infectious disease in Baltimore drug users found that 45% of recent users of cocaine or heroin were infected with hepatitis C [15]. In 2014, another study found that of the 782 participants tested for HCV, only 19 % reported having received an HCV diagnosis in the past, while 48% tested positive for HCV. Of those diagnosed, only 6% reported having received any form of treatment [16]. This draws attention to the ripple effects of under diagnosed HBV among the IDU population in Baltimore.

In 2008, the National Institutes of Health and the DC Department of Health collaborated to establish the DC Partnership for HIV/AIDS Progress (DC PFAP), a partnership for community-based clinical care and research whose aim is to reduce the incidence and prevalence of HIV/AIDS in the District of Columbia. Led by Director Dr. Henry Masur, a plan was developed to create a research program to build a sustainable model for urban areas working to reduce the HIV/AIDS crisis. A needs assessment within the area's medical community demonstrated a significant deficit in hepatitis C virus (HCV) care and treatment. This led to the development of a Hepatitis branch within DC PFAP, rooted in direct subspecialty medical services and clinical research addressing the limitations of standard of care therapy.

The Hepatitis program is led by Scientific Director Dr. Shyamasundaran Kottilil, and is based out of three campuses: (1) clinical partners within Washington DC, (2) National Institutes of Health, Bethesda, MD and (3) University of Maryland Institute of Human Virology, Baltimore, MD. The overarching goals of the programs are to:

- Improved access to subspecialty care for underinsured patients with HIV
- Develop access to HIV and hepatitis related research for residents of DC
- Build credibility and trust in the community for the research process
- Expand integrated care for hepatitis B and C in the community HIV clinics
- Initiate NIH and IHV-UMSOM research protocols at these DC clinics
- Provide national leadership in the development and delivery of effective, safe, convenient therapies, including HBV and HCV.

The clinical aspects of the DC Partnership for HIV/AIDS (DC PFAP) Progress program in Washington, DC are set within three partner centers: Unity Healthcare/Walker Jones and Unity Healthcare/Parkside and Chinese Culture and Community Services. At these locations, DC PFAP and IHV-SOM providers are embedded within established clinics to augment and enhance subspecialty medical care. Patients are managed by state of the art standard of care as well as offered opportunities to participate in clinical research as appropriate. To date, over 6000 HIV infected patients and over 2000 HCV patients have been linked to care and over 200 patients cured of hepatitis C through treatment with novel directly acting therapy within these subspecialties.

Hepatitis B treatment

Currently, there are two classes of drugs approved for the treatment of HBV: nucleos(t)ide reverse transcriptase inhibitors (NRTIs) and interferon-α (IFN-α). First line antiviral therapy includes entecavir (nucleoside analogue), tenofovir (nucleotide analogue), and pegylated IFN-α (PEG-IFN-α). PEG-IFN is indicated for a course of 48-52 weeks. Despite it having weaker antiviral activity than NRTIs, PEG-IFN-α has been associated with a higher rate of HBeAg and HBsAg loss, but rates are still extremely low (HBsAg

loss 2-7%, and 11% at 6 months and 3 years post treatment, respectively) and is associated with several side effects [6].

Relationship between HBsAg levels, cccDNA and Hepatitis B Functional Cure

The goals of antiviral treatment are to decrease the morbidity and mortality related to chronic HBV infection. Antiviral therapy can reverse hepatic fibrosis, and even cirrhosis, which result in reduction of cirrhosis complications and the risk of hepatocellular carcinoma (HCC) [17, 18]. Treatment success (cure) is categorized as (1) immunological cure: defined by HBsAg loss and sustained HBV DNA suppression; and (2) virologic cure defined by eradication of virus, including the covalently closed circular DNA (cccDNA) form. Virologic cure is currently not a feasible or attainable goal at this time. With the currently available therapy, only a minority of patients achieve immunologic cure. However, the vast majority will require chronic (often lifelong) suppressive therapy with, most frequently tenofovir or entecavir.

A major obstacle in attaining HBV cure is the presence of covalently closed circular DNA (cccDNA) in the hepatocyte nucleus. This non-integrated form of HBV DNA in the infected liver cells, serves as the primary template for transcription of all viral RNAs and plays a vital role in the HBV life cycle as well as drives the production of HBsAg and HBeAg, as well as progeny virions [19]. The half-life of cccDNA is long, perhaps measured in years, and this explains why HBV reactivates either spontaneously or following immune suppression [20]. Therefore, efficient eradication of cccDNA from the infected hepatocytes is now considered the most important task in achieving HBV cure [21]. Quantitation of intrahepatic HBV cccDNA could be accomplished but not without potential complications from liver biopsy. Fortunately, it has been shown that serum HBsAg titer correlates well with intrahepatic HBV cccDNA in patients with chronic hepatitis B naïve to antiviral therapy [22].

Though current HBV antiviral therapy is efficient in inhibiting viral replication by blocking reverse transcription of pgRNA to HBV DNA, it only has a marginal effect on cccDNA production, stability, or transcription [20]. Thus, continued transcription of cccDNA may explain the relatively low decline in serum HBsAg levels during NA therapy. Additionally, it is estimated that it will take an estimated median 36-52.2 years before HBsAg clearance is attained with NA therapy [23-24]. Nevertheless, a robust HBsAg decline during NA treatment is associated with immune responses and appear to lead to subsequent HBsAg loss [25]. In those patients with chronic hepatitis B treated with more than 10 years of lamivudine therapy, an on-treatment reduction of HBsAg by >0.166 $\log_{10}$ IU/mL per year predicted subsequent seroclearance with a negative predictive value of 97.8% [26]. Likewise, a decline of HBsAg >1 log after 1 year of therapy with antiviral telbivudine in HBeAg positive CHB patients was associated with HBsAg loss and enhancement in antiviral T cell reactivity, suggesting immune restoration against HBV infection [27].

Study Hypothesis: Ledipasvir-Sofosbuvir and HBsAg Decline

When patients co-infected with both hepatitis B and hepatitis C have were treated for their hepatitis C (HCV) with twelve (12) weeks of fixed dose combination ledipasvir-sofosbuvir (Harvoni®) at 90 mg and 400 mg daily, respectively, there was an observed decline in the mean quantitative HBsAg level on treatment with mean reduction of 0.14, 0.25, and 0.47 $\log_{10}$ IU/mL, at weeks 1, 4, and 12, respectively [28]. Overall, 37 of 111 (33%) subjects studied in this cohort had an HBsAg decline of greater than or equal to 0.5 $\log_{10}$ IU/mL at 12 weeks on ledipasvir-sofosbuvir [28]. In addition, a recently published study by Dr. Loggi and colleagues, found a similar case of HBV-HCV co-infected patient that was treated with anti-HCV direct-acting antivirals and observed a HBV DNA increase of more than 2 log but with simultaneous HBsAg decline, and subsequent HBsAg loss [29]. Whether these unexpected observations

are a result of direct antiviral properties of ledipasvir-sofosbuvir, or due to a complex interaction between the immune response and interactions between the HBV and HCV during DAA therapy, remains a mystery that this and future trials will hopefully enlighten. This study hypothesizes that a similar reduction in HBsAg levels would be seen in mono-infected hepatitis B infected subjects, either in low replicative state or virally suppressed, over the course of 12 week treatment with ledipasvir-sofosbuvir. In addition, this pilot study will explore if the antiviral effects are due to either sofosbuvir, ledipasvir or both.

2.3 RISK/BENEFIT ASSESSMENT

2.3.1 KNOWN POTENTIAL RISKS

The risks related to phlebotomy include pain, bruising, fainting and very rarely, infection. These risks will be minimized by having trained staff perform the procedures.

The background risk of major birth defects and miscarriage for the indicated population is unknown. In the U.S. general population, the estimated background risk of major birth defects and miscarriage in clinically recognized pregnancies is 2–4% and 15–20%, respectively [30].

There is potential for loss of confidentiality, which will be minimized by providing privacy during study visits. In order to protect all participant confidentiality, all specimens and research records will be coded by number. All paper records will be kept locked with limited access. Electronic data will be password protected and will not include participant names or contact details.

Risk of using LDV/SOF (Harvoni®), SOF (Sovaldi®), or LDV (GS-5885 or ledipasvir)
The most common side effects in all people who take LDV and SOF either in fixed dose combination or as monotherapy are fatigue, headache, nausea, diarrhea and insomnia. In some people with advanced liver disease such as cirrhosis, other common side effects may include: asthenia, myalgia, irritability, dizziness, cough and dyspnea [30]. Discontinuation due to side effects is rare about 1%.

This study also intends to evaluate the safety of using ledipasvir and/or sofosbuvir in subjects with hepatitis B alone, i.e. Not co-infected with hepatitis C. It is well known that successful treatment of hepatitis C, including therapies with ledipasvir/sofosbuvir, in a patient co-infected with hepatitis B are at risk of hepatitis B reactivation, mostly presenting as elevation of liver enzymes but hepatic failures as well as death have been reported. In fact between November 2013 to October 2016, there have been 29 cases reported to the FDA, of which there were two deaths, one requiring liver transplantation, and six hospitalizations [31]. This remains to be a small proportion of people treated for hepatitis C. Of note, the two deaths were on treatment with a different direct acting HCV drugs daclatasvir-asunaprevir and were not on ledipasvir/sofosbuvir [31]. It is known that in people co-infected with both HBV and HCV, their hepatitis C virus usually predominates leading suppression of HBV viral replication [32-33]. Therefore, it is believed that with HCV cure, there is a loss of inhibition against HBV resulting in higher chance of HBV reactivation. We believe that in a different setting such as in subjects only mono-infected with HBV, this risk of HBV reactivation is negligible at best. Nevertheless, subjects in this study will be closely watched and followed with frequent monitoring of liver profile during and after ledipasvir and/or sofosbuvir therapy.

In patients with heart conditions that require them to take amiodarone, administration of LDV/SOF has the potential to cause serious symptomatic bradycardia, as well as fatal cardiac arrests, and cases have required pacemaker interventions. Thus, potential volunteers for any study group who have taken amiodarone in the last 90 days or are required to take amiodarone will be excluded from this study.

The risk profile of LDV/SOF FDC will be also utilized for those in the monotherapy arms as it more closely represents the risks of either medication than risk profiles stated from use of either ledipasvir or sofosbuvir with other hepatitis C treatments such as interferon or ribavirin. Neither ledipasvir nor sofosbuvir have been studied alone as treatment in a viral hepatitis population, while LDV/SOF FDC has been safely used in the hepatitis C population with and without other concomitant medications.

Risk of Blood Draws
Collecting blood samples from a vein may cause pain, bruising, lightheadedness, fainting, bleeding and rarely infection at the site of the needle stick.

Risk of Electrocardiogram (ECG)
After an ECG, subjects may experience mild skin irritation, slight redness or itching where the recording patches were placed. They may also need to have chest hair shaved for the procedure.

Risk of FibroScan
FibroScans are non-invasive procedures. Subjects may experience a sense of vibration from the probe tip and a temporary red mark on the skin following the procedure.

Risk of Allergic Reaction
Allergic reaction is always possible. Serious allergic reactions that may occur can sometimes be life-threatening. Allergic reactions may include pruritus, rashes, difficulty of breathing and wheezing, sudden drop in blood pressure, swelling around the mouth, throat, or eyes, palpitations, and sweating.

Risk of Loss of Confidentiality
There is always a potential for the loss of confidentiality. This risk will be minimized by keeping all study data stored and secured. Electronic data will be password protected. Private information will only be given out as listed in the HIPAA form.

Unexpected Risks and Discomforts
There may be risks in this study that are not yet known or happen rarely when subjects take these study drugs. Subjects will be told of any new information that could impact their safety during study participation.

Pregnancy and Breast-Feeding Risks
Because the effects of LDV and/or SOF on an unborn baby or a nursing infant are not known, care must be taken to avoid pregnancy in female subjects or in female partners of male subjects during this study and following completion of study treatment (30 days after completion for women, 14 days after completion for men). Subjects will not be enrolled if pregnant or while breast-feeding.

2.3.2 KNOWN POTENTIAL BENEFITS

There is no direct benefit with participation. In this study, we will be evaluating whether or not LDV and SOF either in combination or as monotherapy will lead to decline in HBsAg levels in patients. If this study finds that LDV/SOF has an effect, future studies may lead to evaluating benefit of LDV and/or SOF for HBV patients.

2.3.3 ASSESSMENT OF POTENTIAL RISKS AND BENEFITS

As there are limited treatment options available for chronic hepatitis B, to investigate agents such as LDV and SOF, which are already proven to be safe with limited side effects profile will provide more benefits than risks in the long term. The risks to participants will be minimized by frequent scheduled and unscheduled visits as needed to monitor and ensure safety.

The value of the information gained will outweigh the risks of participation in the study because any additional treatment options and potential cure for HBV will further eliminate financial and emotional burdens to the affected population globally.

3 OBJECTIVES AND ENDPOINTS

OBJECTIVES	ENDPOINTS	JUSTIFICATION FOR ENDPOINTS
Primary		
To evaluate the change of serum hepatitis B surface antigen (HBsAg $\log_{10}$ IU/mL) level as an indicator of antiviral activity of ledipasvir and sofosbuvir either FDC or as monotherapy in subjects with chronic hepatitis B from baseline to end of 12 weeks treatment.	The primary endpoint is HBsAg ($\log_{10}$ IU/mL) at the end of 12 weeks treatment.	HBV surface antigen (HBsAg) is a marker of HBV infection and an essential part of the HBV lifecycle. Based on data from a phase 2 study evaluating 12 weeks of LDV/SOF in subjects coinfected with HCV and HBV, a decline from baseline in HBsAg was observed. This study will evaluate whether similar changes in HBsAg are observed in HBV monoinfected subjects as the primary efficacy endpoint.
To evaluate the safety and tolerability of the treatment regimens as assessed by review of the accumulated safety data.	The primary safety and tolerability endpoint is any AE(s) leading to permanent discontinuation of study drug(s).	One of the study aims is to evaluate the safety and tolerability of LDV and SOF either FDC or as monotherapy in HBV monoinfected subjects, thus any AE leading to discontinuation should be an endpoint. Clinically significant laboratory abnormalities and vital sign changes will be captured as an AE.

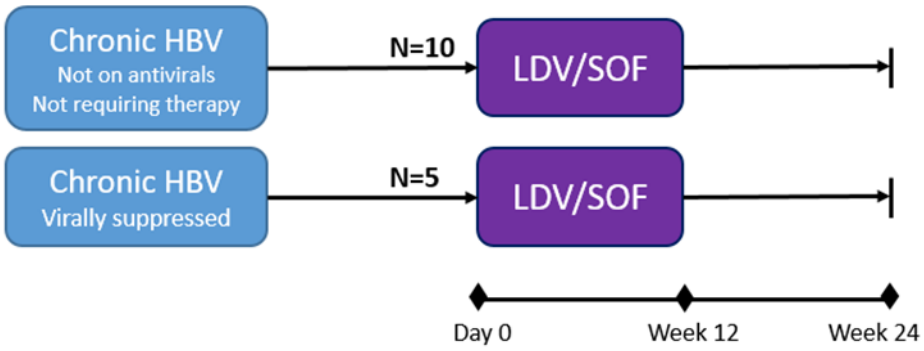
Secondary		
To evaluate the kinetics of circulating HBV DNA in subjects infected with chronic HBV.	The secondary endpoint is HBV DNA (log_{10}IU/mL) at the end of 12 weeks treatment.	HBV DNA in the blood is the approved clinical marker to monitor for HBV replication. Changes in HBV DNA would provide important information on the antiviral activity of LDV and/or SOF treatment.
Exploratory		
To explore the relationship between pharmacodynamic changes and viral parameters.	The exploratory endpoints may include immunologic changes, such as, change from baseline in cytokine profile, and changes in viral parameters including HBV RNA and HBV core related antigen during and after treatment with LDV and/or SOF.	Immunologic changes may occur with a decline in HBsAg in HBV infected subjects. If HBsAg decline is observed during treatment, we will explore immunologic changes in relation to decline in HBsAg levels. Changes in viral parameters including serum HBV RNA and Hepatitis B core related antigen (HBcrAg) will be explored to further evaluate the antiviral activity of LDV and/or SOF. Both serum HBV RNA and HBcrAg are experimental markers of HBV replication and reflect the transcriptional activity of intrahepatic cccDNA. Changes in one or both of these viral parameters may provide important information on the antiviral activity of LDV and/or SOF treatment.

4 STUDY DESIGN

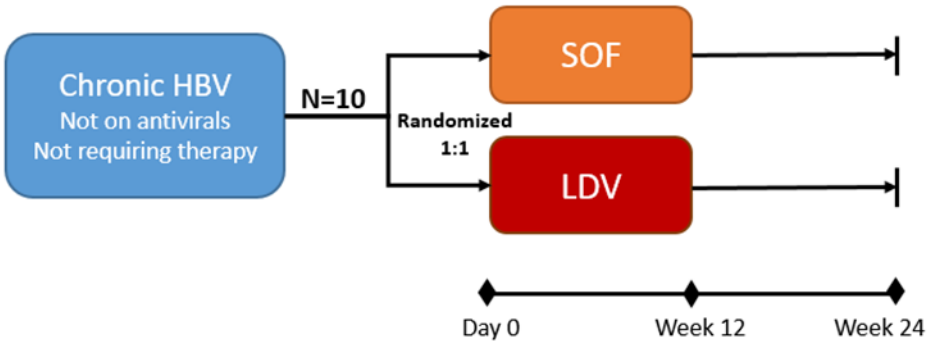
4.1 OVERALL DESIGN

This is a phase 2, open-label, interventional study for evaluating antiviral activity by measuring change of HBsAg quantification, safety and tolerability of LDV and/or SOF either FDC or as monotherapy for treatment of chronic HBV infection. It is anticipated that approximately 40 HBV mono-infected subjects will be screened to enroll 25 subjects, which will include referrals from DC Partnership for HIV/AIDS Progress (DCPFAP) program. Study subjects will be screened and enrolled at the Institute for Human Virology Clinical Research Unit (IHV CRU), Chinese Culture and Community Service Center, Inc. (CCACC) or Unity Health Care, Inc. Parkside by research study staff. Eligible participants will receive 12 weeks of LDV and/or SOF either as FDC or monotherapy, and continue for additional 12 weeks of follow up. Serial measurement of viral, and safety labs will be performed.

Groups A & B – Subjects enrolled will receive twelve weeks of combination therapy ledipasvir/sofosbuvir 400mg/90mg once daily. **Group A** will enroll up to 10 chronic HBV subjects who are in a low replicative state and therefore, not requiring treatment. **Group B** will enroll 5 chronic HBV subjects virally suppressed on anti-HBV medications. All subjects will be followed until 12 weeks post-treatment. Subjects will have study visits on Day 0 (Baseline), Week 1, Week 2, Week 4, Week 8, Week 12, and post-treatment Weeks 13, 16 and 24.



Groups C & D – Ten subjects will be randomized in a 1:1 ratio to receive twelve weeks daily dose of either sofosbuvir (400 mg once daily) or ledipasvir (90 mg daily). All subjects will be followed until 12 weeks post-treatment. Subjects will have study visits on Day 0 (Baseline), Week 1, Week 2, Week 4, Week 8, Week 12, and post-treatment Weeks 13, 16 and 24.



It is anticipated that LDV and/or SOF will result in decline of mean quantitative HBsAg >0.4 $\log_{10}$/mL from baseline to end of treatment in HBV mono-infected patients similar to that observed in HBV/HCV co-infected patients. With a small cohort, no interim analysis is planned.

4.2 SCIENTIFIC RATIONALE FOR STUDY DESIGN

Based on retrospective data from a phase 2 study evaluating 12 weeks of LDV/SOF in subjects coinfected with HCV and HBV, a decline from baseline in HBsAg was observed. This study is designed to evaluate whether similar changes in HBsAg are observed in HBV monoinfected subjects, either in low replicative state or virally suppressed, with LDV and/or SOF treatment for duration of 12 weeks and further translate the antiviral activity of LDV and/or SOF.

4.3 JUSTIFICATION FOR DOSE

Dosing for this clinical trial will utilize the FDA approved dosing and length of treatment for chronic hepatitis C treatment as the most safety data is available for this choice. Additionally, as declines in quantitative hepatitis B surface antigen levels were seen when LDF/SOF was taken by those with HCV and HBV, this study aims to replicate the treatment to compare quantitative changes in a monoinfected population. LDV/SOF will be taken orally as a once-daily dose regardless of food. As it is a fixed-dose combination, no dose adjustments will be made.

Study medications used for the monotherapy arms will use corresponding dose as those used in combination LDV/SOF therapy (i.e. 400mg SOF or 90 mg LDV). Likewise, there will be no dose adjustments made.

4.4 END OF STUDY DEFINITION

A participant is considered to have completed the study if he or she has completed all phases of the study including the last visit or the last scheduled procedure shown in the Schedule of Activities (SoA), Section 1.3.

The end of the study is defined as completion of the last visit or procedure by all subjects as shown in the SoA.

5 STUDY POPULATION

5.1 INCLUSION CRITERIA

In order to be eligible to participate in this study, an individual must meet all of the following criteria:

Participants in Groups A, C & D (Chronic HBV, low replicative state not requiring treatment):

1. Provision of signed and dated informed consent form
2. Stated willingness to comply with all study procedures and availability for the duration of the study
3. Male or female, aged 18 or older at screening
4. Diagnosed with chronic hepatitis B infection defined as one of the following:
    a. HBsAg or HBV DNA positivity for at least 6 months
    b. Medical records indicating a chronic HBV infection
5. HBeAg negative at screening
6. HBV DNA > lower level of quantitation (LLOQ)
7. Quantitative HBsAg at least 10 IU/mL at screening
8. Ability to take oral medication and be willing to adhere to the twelve week study drug regimen
9. For females of reproductive potential: usual practice of complete abstinence from sexual intercourse with a member of the opposite sex OR use of at least one form of highly effective contraception for at least 1 month prior to enrollment and agreement to use such a method during study participation and for an additional 30 days after the end of study drug administration
10. For males of reproductive potential: usual practice of complete abstinence from sexual intercourse with a member of the opposite sex OR use of at least one form of highly effective contraception for at least 1 month prior to enrollment and agreement to use such a method during study participation and for an additional 14 days after the end of study drug administration
11. Ability to communicate effectively with the study investigator and key staff
12. Medical management provided by a primary care provider
13. Ability to store medications at a room temperature of less than 86 degrees Fahrenheit
14. Not on antiviral therapy or requiring treatment for HBV during screening

Participants in Group B (Chronic HBV, virally suppressed):

1. Provision of signed and dated informed consent form
2. Stated willingness to comply with all study procedures and availability for the duration of the study
3. Male or female, aged 18 or older at screening
4. Diagnosed with chronic hepatitis B infection defined as one of the following:
    a. HBsAg or HBV DNA positivity for at least 6 months
    b. Medical records indicating a chronic HBV infection
5. Receiving oral anti-HBV medications (either tenofovir alafenamide, tenofovir disoproxil fumarate, entecavir, or a combination of no more than 2 of these agents) for at least three months prior to enrollment
6. HBV DNA < lower level of quantitation (LLOQ) at screening and for at least three months prior
7. Quantitative HBsAg at least 10 IU/mL at screening
8. Ability to take oral medication and be willing to adhere to the twelve week study drug regimen
9. For females of reproductive potential: usual practice of complete abstinence from sexual intercourse with a member of the opposite sex OR use of at least one form of highly effective contraception for at least 1 month prior to enrollment and agreement to use such a method during study participation and for an additional 30 days after the end of study drug administration
10. For males of reproductive potential: usual practice of complete abstinence from sexual intercourse with a member of the opposite sex OR use of at least one form of highly effective contraception for at least 1 month prior to enrollment and agreement to use such a method during study participation and for an additional 14 days after the end of study drug administration
11. Ability to communicate effectively with the study investigator and key staff
12. Medical management provided by a primary care provider
13. Ability to store medications at a room temperature of less than 86 degrees Fahrenheit

5.2 EXCLUSION CRITERIA

An individual who meets any of the following criteria will be excluded from participation in this study:

1. Coinfection with hepatitis C, hepatitis D or human immunodeficiency virus (HIV)
2. Pregnancy or lactation
3. Known allergic reactions to sofosbuvir or ledipasvir
4. Treatment with another investigational drug or other intervention within three months
5. Evidence of cirrhosis or hepatic decompensation such as:
    - Platelets less than 100,000 /mm^3
    - Albumin less than 3.5 g/dL
    - INR greater than 1.7 or Prothrombin time of 1.5 times the upper limit of normal (ULN)
    - Total bilirubin of 1.5 times the upper limit of normal
    - FibroTest (or FibroSure®) of 0.75 or greater
6. Abnormal hematological and biochemical parameters at screening including:
    - White blood cell count less than 2500 cells/uL
    - Absolute neutrophil count (ANC) less than 1,000 cells/mm3 (less than 750 mm3 for African or African-American subjects)
    - Hemoglobin less than 12 g/dL for males, less than 11 g/dL for females

- AST or ALT of two times the upper limit of normal
- Estimated GFR less than 50 mL/min
- Glycosylated hemoglobin (HbA1c) greater than 8.5%

7. Current or prior history of any of the following:
    - Immunodeficiency disorders or autoimmune disease (e.g. Systemic lupus erythematosus, rheumatoid arthritis, inflammatory bowel diseases, sarcoidosis, psoriasis of greater than mild severity)
    - Severe pulmonary disorders, significant cardiac diseases
    - Gastrointestinal disorder with post-operative condition that could interfere with the absorption of the study drugs
    - Significant psychiatric illness that in the judgment of the Investigator, is a contraindication to protocol participation or impairs a volunteer's ability to give informed consent
    - Any malignancy diagnosed within 5 years (not including recent localized treatment of squamous or non-invasive basal cell skin cancer; cervical carcinoma in situ appropriately treated prior to screening)
    - Solid organ transplantation
    - Poor venous access
8. Screening ECG with clinically significant findings
9. Evidence of HCC (e.g., α fetoprotein > 50ng/mL or radiologic evidence)
10. Clinically significant illicit drug or alcohol abuse within 12 months of screening. Subjects on methadone maintenance treatment or prescribed opioid may be included.
11. Use of amiodarone within 90 days of enrollment; or carbamazepine, phenytoin, phenobarbital, oxcarbazepine, rifabutin, rifampin, rifapentine, St. John's wort, rosuvastatin, or interferon within 30 days of enrollment or expected use of these prohibited drugs during study participation. Use of or expected need of proton-pump inhibitors more than 20 mg omeprazole equivalent or H2 receptor antagonist more than 40 mg famotidine BID equivalent within 7 days of enrollment.

5.3 LIFESTYLE CONSIDERATIONS

Not Applicable

5.4 SCREEN FAILURES

Screen failures are defined as participants who consent to participate in the clinical trial but are not subsequently eligible or entered into the study. A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants, to meet the Consolidated Standards of Reporting Trials (CONSORT) publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details, eligibility criteria, and any serious adverse event (SAE).

Individuals who do not meet the criteria for participation in this trial (screen failure) because of a lab value may have a single retest of screening labs if there is reason to believe the retest value will be within accepted parameters, if the initial value was either due to a sample processing error or due to an extenuating circumstance such as intercurrent infection. Rescreened participants should be assigned the same participant number as for the initial screening.

5.5 STRATEGIES FOR RECRUITMENT AND RETENTION

We anticipate screening 40 subjects from the local area outpatient clinic population and by advertisements in order for approximately 25 to complete study treatment. Separate screening and enrollment consents will be utilized for this trial. It is expected that at least 50% will be from minority populations with a near equal mix of genders based on the population affected by chronic hepatitis B in the local area. Subjects may be approached at outpatient visits by their clinicians, and referred to the study team if interested in the study or may come from ongoing study populations at the sites.

UMB Study Team Roles at Collaborating Sites: UMB Associate Investigators have been actively involved in patient care at the collaborating sites (Unity Parkside and Chinese Culture and Community Services). These sites will sign a UMB Reliance Agreement prior to initiating any study activities. The site AI/Collaborating Investigator if applicable may see subjects at non-study visits and will communicate study relevant findings to UMB/IHV study team. The UMB Associate Investigators will oversee all research operations at the sites.

If Participants Are Seen at Collaborating Sites: The UMB study staff will see participants for all study visits at the sites. All phlebotomy will be done by a phlebotomist working in the clinic or clinic staff. A specimen bag and specimen tubes will be prepared by UMB staff and will be labeled with the participant's study number. All laboratory studies will be received and reviewed by UMB study staff. Clinic staff will not participate in study-related visits unsupervised by UMB study staff. The primary responsibility of the site AI and the clinic is to provide timely and effective communication regarding any potential adverse events if a subject/patient is seen for care outside of the normal study visit. Any visits required of subjects that do not occur when study staff are present will need to be scheduled at another participating site with research team staffing. Any medical problems not related to the study will be addressed by the participant's primary care or hepatitis providers. This will be communicated to the participants when consented for the protocol.

Pregnant women, those lacking consent capacity including mentally ill, prisoners, cognitively impaired subjects and children will not be eligible for enrollment. However, in this academic research setting employees or students may participate if they desire, but will not be consented by an academic superior or supervisor and will be notified in the consent process that employment status or academic standing at will not be affected by participation or nonparticipation in this study. Any subject imprisoned during the study will be removed, and may be replaced at the Investigator's discretion.

Subjects will receive remuneration of $120 cash or gift cards at each scheduled study visit, and parking vouchers will be provided to subjects. Total compensation for each participant over the length of the study is $1200.00. For subjects residing more than 20 miles from the research clinic, Uber gift cards may be used to reimburse transportation to and from study visits, or they can receive additional mileage compensation in the amount of 50 cents per mile.

6 STUDY INTERVENTION

6.1 STUDY INTERVENTION(S) ADMINISTRATION

6.1.1 STUDY INTERVENTION DESCRIPTION

Ledipasvir/Sofosbuvir FDC:
Ledipasvir/Sofosbuvir (HARVONI®) is a fixed-dose combination tablet for daily oral administration. It was approved by the Food and Drug Administration (FDA) for the treatment of chronic hepatitis C in 2014. Ledipasvir (LDV) is an HCV NS5A inhibitor and sofosbuvir (SOF) is a nucleotide analog inhibitor of HCV NS5B polymerase. The pharmacokinetic properties of LDV, SOF, and the predominant circulating metabolite GS-331007 have been evaluated in healthy adult subjects and in subjects with chronic hepatitis C. Both were absorbed quickly with the peak median plasma concentration between 0.8 to 4.5 hours post-dose. Please refer to the product insert for additional information [30].

This fixed dose combination drug is not indicated for the treatment of chronic hepatitis B as is being studied in this clinical trial.

Sofosbuvir:
Sofosbuvir (SOF) is a nucleotide analog inhibitor of HCV NS5B polymerase that was approved by the Food and Drug Administration (FDA) for the treatment of chronic hepatitis C in 2013. It's FDA approved trade name is SOVALDI®. The pharmacokinetic properties of SOF and its predominant circulating metabolite GS-331007 have been evaluated in health adult subjects and in subjects with chronic hepatitis C. SOF is absorbed quickly with the peak median plasma concentration between 0.8 to 4.5 hours post-dose. Please refer to the SOVALDI® package product insert for additional information [34].

SOF monotherapy is currently not indicated for the treatment of chronic hepatitis B as is being studied in this clinical trial.

Ledipasvir (also known as GS-5885):
Ledipasvir (LDV) is an HCV NS5A inhibitor which is approved by the FDA in combination with sofosbuvir for the treatment of chronic hepatitis C. The pharmacokinetic properties of LDV have been evaluated in healthy adult subjects and in subjects with chronic hepatitis C. LDV is absorbed quickly with the peak median plasma concentration between 0.8 to 4.5 hours post-dose. Please refer to the Investigator's Brochure for additional information [350].

LDV monotherapy is currently not indicated for the treatment of chronic hepatitis B as is being studied in this clinical trial.

6.1.2 DOSING AND ADMINISTRATION

Ledipasvir/Sofosbuvir FDC:
Each tablet contains 90 mg LDV and 400 mg SOF for daily oral administration with or without food

Sofosbuvir:
Each tablet contains 400 mg SOF for daily administration with or without food.

Ledipasvir (also known as GS-5885):
Each tablet contains 90 mg LDV for daily administration with or without food.

If a dose of FDC, SOF or LDV is missed, but discovered the same day the dose may be taken, however two doses must not be taken on the same day. Subjects will be instructed to call for replacement study product if stored over 86 degrees Fahrenheit.

6.2 PREPARATION/HANDLING/STORAGE/ACCOUNTABILITY

6.2.1 ACQUISITION AND ACCOUNTABILITY

Gilead Sciences, Inc. will provide the study drug for this clinical trial. It will be delivered to the investigational pharmacy at the Institute of Human Virology and dispensed by the Investigational Pharmacist to eligible subjects in four-week supplies at Day 0, Week 4, and Week 8 visits. Unused pills and empty bottles will be returned by the subjects to the pharmacy and may be destroyed onsite per standard operating procedure (SOP) after Sponsor approval.

6.2.2 FORMULATION, APPEARANCE, PACKAGING, AND LABELING

All bottle containers will be labeled to include the statement, “Caution: New Drug – Limited for Federal (or United States) law to investigational use only”.

Sample label:

APOSTLE Dr. Chua
Subject #
Harvoni 90mg/400mg #28
Take 1 tablet daily.

Disp: Exp:
RETURN ALL USED/UNUSED DRUGS TO CLINIC
Institute of Human Virology
INVESTIGATIONAL DRUG SERVICE
UNIVERSITY OF MARYLAND MEDICAL SYSTEM
420-706-0275
Caution: New Drug – Limited by Federal (United States) law to investigational use only.

Ledipasvir/Sofosbuvir FDC:
HARVONI tablets are orange, diamond-shaped, film-coated, debossed with “GSI” on one side and “7985” on the other side of the tablet. Each bottle contains 28 tablets (NDC 61958-1801-1), a silica gel desiccant, polyester coil, and is closed with a child-resistant closure [30].

Sofosbuvir:
Sofosbuvir (SOVALDI®) tablets are as a yellow-colored, capsule-shaped, film-coated tablet debossed with “GSI” on one side and “7977” on the other side. Each tablet contains 400 mg sofosbuvir [34]. Each bottle contains 28 tablets, a silica gel desiccant and polyester coil, and is closed with a child-resistant closure [34].

Ledipasvir (also known as GS-5885):
Ledipasvir (GS-5885) tablets, 90 mg, are white, capsule-shaped, plain-faced, film-coated tablets. The tablets are approximately 15 mm in length and 7 mm in width. Thirty (30) tablets are packaged in 75-mL white, high density polyethylene bottles with polyester coil packing material. Each bottle is capped with a white continuous thread, child-resistant polypropylene screw cap with an induction-sealed, aluminum-faced liner [35].

6.2.3 PRODUCT STORAGE AND STABILITY

Ledipasvir / Sofosbuvir FDC, Sofosbuvir or Ledipasvir:
Bottles should be stored at room temperature below 30 °C (86 °F), dispensed in the original containers with intact seals [30, 34, 35].

6.2.4 PREPARATION

Bottles will be re-labeled by the Investigational Pharmacist as prescribed to each study subject after consent is given and subjects are found to be eligible.

6.3 MEASURES TO MINIMIZE BIAS: RANDOMIZATION AND BLINDING

Volunteers for Groups C and D will be randomized using block randomization in a 1:1 ratio of SOF:LDV. They will be block randomized in blocks of two. This will be done by the research pharmacist utilizing a random number generator called Stattrek. There will be a total of five blocks generated of two volunteers in each randomized block.

6.4 STUDY INTERVENTION COMPLIANCE

Study drug compliance will be assessed by subject recall at each visit. Drug records will be maintained by the IND pharmacist, and subjects will be instructed to return any unused pills or empty bottles for destruction at the site.

6.5 CONCOMITANT THERAPY

For this protocol, a prescription medication is defined as a medication that can be prescribed only by a properly authorized/licensed clinician. Medications to be reported in the source documents are concomitant prescription medications, over-the-counter medications and supplements. Concomitant medications taken from the time of screening through end of study treatment will be recorded at each study visit. All subjects will be asked about prohibited agents and those to be used with caution taken within the 90 days preceding screening during the screening visit and available medical records will be reviewed.

Treatment with the following drugs is either prohibited or to be used with caution due to the potential for drug-drug interactions and risk of adverse events.

Disallowed Concomitant Medications [30]

Drug Class	Prohibited Agents	Clinical Comment
Antiarrhythmics	amiodarone	Coadministration of amiodarone with HARVONI may result in serious symptomatic bradycardia. The mechanism of this effect is unknown.
Anticonvulsants	carbamazepine phenytoin phenobarbital oxcarbazepine	Coadministration is expected to decrease the concentration of ledipasvir and sofosbuvir, leading to reduced therapeutic effect of HARVONI.
Antimycobacterials	rifabutin rifampin rifapentine	Coadministration is expected to decrease the concentration of ledipasvir and sofosbuvir, leading to reduced therapeutic effect of HARVONI.
Herbal Supplements	St. John's wort	Coadministration of HARVONI with St. John's wort, a P-gp inducer, is not recommended due to decreased concentration of ledipasvir and sofosbuvir.
HMG-CoA Reductase Inhibitors	rosuvastatin	Coadministration may significantly increase the concentration of rosuvastatin which is associated with increased risk of myopathy, including rhabdomyolysis.

Medications to be Used with Caution [30]

Drug Class	Used with Caution	
Antiarrhythmics	digoxin	Coadministration may increase the concentration of digoxin
Antacids	Tums, Maalox, etc.	It is recommended to separate antacid and HARVONI administration by 4 hours to avoid decreased concentration of ledipasvir.
H2-receptor antagonists	Nizatidine Famotidine Cimetidine ranitidine	H2-receptor antagonists may be administered simultaneously with or 12 hours apart from HARVONI at a dose that does not exceed doses comparable to famotidine 40 mg twice daily to avoid decreased concentration of ledipasvir.
Proton-pump inhibitors	Dexlansoprazole Esomeprazole	Proton-pump inhibitor doses comparable to omeprazole

Proton-pump inhibitors (cont).	Lansoprazole Omeprazole Pantoprazole Rabeprazole	20 mg or lower can be administered simultaneously with HARVONI under fasted conditions to avoid decreased concentration of lediipasvir.

6.5.1 RESCUE MEDICINE

Any subject from Groups A, C or D who meets criteria for HBV treatment during the study will be referred to their clinical provider for assessment or to study NCT02995252 which offers FDA approved treatment for up to two years for subjects willing to participate in every three-month research lab collection.

7 STUDY INTERVENTION DISCONTINUATION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

7.1 DISCONTINUATION OF STUDY INTERVENTION

Subjects may withdraw consent at any time for any reason. Subjects may have study drug stopped by the investigator should any untoward effect occur but continue to participate in post-treatment visits.

A subject must be discontinued from study drug but may continue with post-treatment visits if:

- The subject has a Grade 3 or 4 adverse event or tolerability issue related to study drug
- The subject has a GFR of <30 mL/min (confirmed by repeat measurement) or a two log increase in HBV DNA while on study drug (confirmed by repeat measurement)
- Significant study intervention non-compliance

Discontinuation from treatment is permanent. Once a subject has been discontinued, they will not be allowed to restart study drug. Subjects who require discontinuation will continue to follow the general study schedule of assessments unless unwilling to do so.

Discontinuation from LDV/SOF does not mean discontinuation from the study, and remaining study procedures should be completed as indicated by the study protocol. If a clinically significant finding is identified (including, but not limited to changes from baseline) after enrollment, the investigator or qualified designee will determine if any change in participant management is needed. Any new clinically relevant finding will be reported as an adverse event (AE).

7.2 PARTICIPANT DISCONTINUATION/WITHDRAWAL FROM THE STUDY

Participants are free to withdraw from participation in the study at any time upon request.
An investigator may discontinue or withdraw a participant from the study for the following reasons:

- The subject withdraws consent.
- The subject has a medical condition or personal circumstance which, in the opinion of the investigator and/or Sponsor, places the subject at unnecessary risk through continued participation in the trial or does not allow the subject to adhere to the requirements of the protocol.
- The subject fails to comply with the dosing, evaluations, or other requirements of the study.
- An investigator feels it is in the best interest of the subject to discontinue
- If a patient becomes pregnant during the course of the study, the patient will be discontinued from the study once safety is evaluated

When a subject/patient discontinues/withdraws prior to study completion, all applicable activities scheduled for the final study visit should be performed at the time of discontinuation. Any adverse events which are present at the time of discontinuation/withdrawal should be followed as per protocol. The subject will then be referred back to their primary care physician for determination of appropriate management of the patient's HBV infection.

The reason for participant discontinuation or withdrawal from the study will be recorded in the RedCap database. Subjects who sign the informed consent form but do not receive the study intervention may be replaced. Subjects who sign the informed consent form, and receive the study intervention, and subsequently withdraw, or are withdrawn or discontinued from the study, may be replaced at the discretion of the investigator.

A participant will be considered lost to follow-up if they fail to return for two scheduled visits and are unable to be contacted by the study site staff.

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

- The site will attempt to contact the participant and reschedule the missed visit within the study visit window if possible, and counsel the participant on the importance of maintaining the assigned visit schedule and ascertain if the participant wishes to and/or should continue in the study.
- Before a participant is deemed lost to follow-up, the investigator or designee will make every effort to regain contact with the participant (where possible, 3 telephone calls and, if necessary, a certified letter to the participant's last known mailing address). These contact attempts should be documented in the participant's medical record or study file.
- Should the participant continue to be unreachable after two missed study visits, they will be considered to have withdrawn from the study with a primary reason of lost to follow-up.

8 STUDY ASSESSMENTS AND PROCEDURES

8.1 EFFICACY ASSESSMENTS

Description of procedures / evaluations:

- **Biological specimen collection and laboratory evaluations**. Specimens will be obtained at the IHV CRU, CCACC Clinic or Unity Parkside Clinic. Clinical laboratories will be sent out to LabCorp for processing, and results will be reviewed by a study practitioner. Research labs will be delivered to the research processing lab within the IHV and research tests will be batched and performed from previously collected and frozen samples. Serum storage samples (6ml) and plasma storage samples (6 ml) will be collected at each visit from Day 0 onward. Quantitative HBsAg/HBcrAg (8 ml) will be collected at screening and every visit thereafter. Optional PBMC storage will be collected (20-50 ml) each visit from Day 0 onward. Optional Pax DNA samples (8.5ml) will only be collected if not already stored for the subject from previous study participation. Optional Pax RNA samples (2.5ml) will be collected at timepoints noted in the SoA.

8.2 SAFETY AND OTHER ASSESSMENTS

Once subjects have signed the screening consent, assessment of eligibility may begin. Screening may be completed from 60 to 28 days prior to Day 0 in order to collect two samples for HBsAg and HBcrAg at least 28 days apart prior to dosing for Groups A & B. Groups C & D do not require the timing for the second quantitative samples, and have a 60 day screening window. If found to be eligible at screening, medical history, physical exam and concomitant medications will be reviewed on Day 0 prior to study consenting by a licensed provider on the study team. If ineligible, subject will not move on to sign study consent at that time.

After study consent is obtained, the remaining Day 0 events listed in the SoA will be completed. Subjects will have laboratory assessments obtained prior to first dose of study medication to be given during the visit. Subjects will be provided with labeled study medication for home dosing after Day 0.

- **Physical examination** is to include assessment of head, neck and thyroid; eyes, ears, nose, throat, mouth and tongue; respiratory; cardiovascular; lymph nodes and abdomen; skin, hair and nails; musculoskeletal; and neurological systems. Targeted exams may be limited to the systems of concern. Physical exams will be completed by a licensed practitioner.
- **Outside clinical labs** may be utilized for eligibility for CBC, chemistry panel, coagulation factors, Hepatitis C and D and HIV serologies from the last 30 days, or last 90 days for those re-enrolling who have previously completed Group A. Outside clinical HBsAg and anti-HBs and quantitative HBV DNA results may be utilized from the last 90 days, and FibroSure and HgbA1C tests may be utilized from up to 12 months prior to screening for eligibility following local HIPAA requirements.
- Clinical lab results may be shared with the subject upon request, however research results will not be provided to subject medical providers.
- Study drug adherence will be assessed by subject recall and pill counts at all scheduled visits.

- Assessment of adverse events will be completed as per the SoA.

8.3 ADVERSE EVENTS AND SERIOUS ADVERSE EVENTS

8.3.1 DEFINITION OF ADVERSE EVENTS (AE)

Adverse event means any untoward medical occurrence associated with the use of an intervention in humans, whether or not considered intervention-related (21 CFR 312.32 (a)).

8.3.2 DEFINITION OF SERIOUS ADVERSE EVENTS (SAE)

An adverse event (AE) or suspected adverse reaction is considered "serious" if, in the view of either the investigator or sponsor, it results in any of the following outcomes: death, a life-threatening adverse event, inpatient hospitalization or prolongation of existing hospitalization, a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions, or a congenital anomaly/birth defect. Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the participant and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

8.3.3 CLASSIFICATION OF AN ADVERSE EVENT

8.3.3.1 SEVERITY OF EVENT

The investigator will grade the severity of each AE according to the “NIH National Cancer Institute Common Terminology Criteria for Adverse Events ” (Version 4.03, June 2010; Published: May 08, 2009), which can be found at: *https://evs.nci.nih.gov/ftp1/CTCAE/CTCAE_4.03_2010-06-14_QuickReference_5x7.pdf*

Adverse Events not found in the Toxicity Table will be assessed for severity and classified into one of the categories below:

- **Grade 1** (Mild): Event requires minimal or no treatment and do not interfere with the participant’s daily activities.
- **Grade 2** (Moderate): Event results in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning.
- **Grade 3** (Severe): Event interrupts a subject’s usual daily activity or functioning and may require systemic drug therapy or other treatment. Severe events are usually incapacitating.
- **Grade 4** (Potentially Life threatening): Events causing inability to perform basic self-care functions OR Medical or operative intervention indicated to prevent permanent impairment, persistent disability, or death.
- **Grade 5** (Death)

8.3.3.2 RELATIONSHIP TO STUDY INTERVENTION

All adverse events (AEs) must have their relationship to study intervention assessed by the clinician who examines and evaluates the participant based on temporal relationship and clinical judgment. The degree of certainty about causality will be graded using the categories below. In a clinical trial, the study product must always be suspect.

- **Definitely Related** – There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out. The clinical event, including an abnormal laboratory test result, occurs in a plausible time relationship to study intervention administration and cannot be explained by concurrent disease or other drugs or chemicals. The response to withdrawal of the study intervention (dechallenge) should be clinically plausible.
- **Probably Related** – There is evidence to suggest a causal relationship, and the influence of other factors is unlikely. The clinical event, including an abnormal laboratory test result, occurs within a reasonable time after administration of the study intervention, is unlikely to be attributed to concurrent disease or other drugs or chemicals, and follows a clinically reasonable response on withdrawal.
- **Potentially Related** – There is some evidence to suggest a causal relationship (e.g., the event occurred within a reasonable time after administration of the trial medication). However, other factors may have contributed to the event (e.g., the participant's clinical condition, other concomitant events). Although an AE may rate only as "possibly related" soon after discovery, it can be flagged as requiring more information and later be upgraded to "probably related" or "definitely related", as appropriate.
- **Unlikely to be related** – A clinical event, including an abnormal laboratory test result, whose temporal relationship to study intervention administration makes a causal relationship improbable (e.g., the event did not occur within a reasonable time after administration of the study intervention) and in which other drugs or chemicals or underlying disease provides plausible explanations (e.g., the participant's clinical condition, other concomitant treatments).
- **Not Related** – The AE is completely independent of study intervention administration, and/or evidence exists that the event is definitely related to another etiology. There must be an alternative, definitive etiology documented by the clinician.

8.3.3.3 EXPECTEDNESS

The Principal Investigator will be responsible for determining whether an adverse event (AE) is expected or unexpected. An AE will be considered unexpected if the nature, severity, or frequency of the event is not consistent with the risk information previously described for the study intervention.

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

All adverse events (AEs) occurring from the time of ingesting one dose of the study drugs through the end of study will be documented, recorded, and reported. Prior to the first dose, only events related to study procedures will be captured as AEs. If a diagnosis is clinically evident, the diagnosis rather than the individual signs and symptoms or lab abnormalities will be recorded as the AE.

All AEs will be captured and tracked on an AE form in the subject's research file as well as entered into the REDCap reporting database. Information to be collected includes event description, date of onset, clinician's assessment of severity, relationship to study product (assessed only by those with the training and authority to make a diagnosis), and date of resolution/stabilization of the event. All AEs occurring while on study must be documented appropriately regardless of relationship. All AEs will be followed to adequate resolution as long as the subjects are willing to participate.

Any medical condition that is present at the time that the participant is screened will be considered as baseline and not reported as an AE. An elective surgery/procedure scheduled to occur during a study will not be considered an AE if the surgery/procedure is being performed for a pre-existing condition and the surgery/procedure has been pre-planned prior to study entry. However, if the study participant's condition deteriorates at any time during the study, it will be recorded as an AE.

Changes in the severity of an AE will be documented to allow an assessment of the duration of the event at each level of severity to be performed. AEs characterized as intermittent require documentation of onset and duration of each episode.

The Principal Investigator and/or study team will record all reportable events with start dates occurring any time after informed consent is obtained the last day of study participation. At each study visit, the study team member will inquire about the occurrence of AE/SAEs since the last visit.

8.3.5 ADVERSE EVENT REPORTING

As per UMB policies, all Reportable New Information will be reported to the UMB IRB within allowed timeframes.

Line listings, frequency tables, and other summary AE data will be submitted to the IND Sponsor (if required) when needed for periodic safety assessments, review of IND annual reports, review of IND safety reports, and preparation of final study reports.

8.3.6 SERIOUS ADVERSE EVENT REPORTING

The study clinician will immediately report to the sponsor any serious adverse event, whether or not considered study intervention related, including those listed in the protocol or investigator brochure and must include an assessment of whether there is a reasonable possibility that the study intervention caused the event. Study endpoints that are serious adverse events (e.g., all-cause mortality) must be reported in accordance with the protocol unless there is evidence suggesting a causal relationship between the study intervention and the event (e.g., death from anaphylaxis). In that case, the investigator must immediately report the event to the sponsor.

All serious adverse events (SAEs) will be followed until satisfactory resolution or until the site investigator deems the event to be chronic or the participant is stable. Other supporting documentation of the event may be requested by the study sponsor and should be provided as soon as possible.

The study sponsor will be responsible for notifying the Food and Drug Administration (FDA) of any unexpected fatal or life-threatening suspected adverse reaction as soon as possible, but in no case later than 7 calendar days after the sponsor's initial receipt of the information if IND is found to be necessary.

In addition, the sponsor must notify FDA and all participating investigators in an Investigational New Drug (IND) safety report of potential serious risks, from clinical trials or any other source, as soon as possible, but in no case later than 15 calendar days after the sponsor determines that the information qualifies for reporting as applicable with IND.

Any SAE will be reported to the IRB per institutional guidelines. SAEs (regardless of relationship) must also be reported to Gilead Drug Safety & Public Health (DSPH) Office within 15 calendar days of first becoming aware of the event.

8.3.7 REPORTING EVENTS TO PARTICIPANTS

Any adverse event or serious adverse event that affects the risk/benefit ratio to subjects will prompt a revision to the informed consent document. All subjects will be notified of the additional risk and re-consented at their next visit, or earlier if the situation requires.

8.3.8 EVENTS OF SPECIAL INTEREST

Not Applicable

8.3.9 REPORTING OF PREGNANCY

Pregnancy itself is not an AE. However, complications of pregnancies are AEs and may be SAEs. Pertinent obstetrical information of all pregnancies will be reported to the Gilead DSHP via fax or email within 3 business days from site awareness of the pregnancy. Study drug must be stopped immediately. The participant will be advised to notify her obstetrician of study agent exposure.

Pregnancy outcome data (e.g., delivery outcome, spontaneous, or elective termination of the pregnancy, presence of absence of birth defects, congenital abnormalities, or other complications) will be reported to the Sponsor and DSPH within 3 business days of the site's awareness on a protocol-specified form.

8.4 UNANTICIPATED PROBLEMS

8.4.1 DEFINITION OF UNANTICIPATED PROBLEMS (UP)

The Office for Human Research Protections (OHRP) considers unanticipated problems involving risks to participants or others to include, in general, any incident, experience, or outcome that meets **all** of the following criteria:

- Unexpected in terms of nature, severity, or frequency given (a) the research procedures that are described in the protocol-related documents, such as the Institutional Review Board (IRB)-approved research protocol and informed consent document; and (b) the characteristics of the participant population being studied;
- Related or possibly related to participation in the research ("possibly related" means there is a reasonable possibility that the incident, experience, or outcome may have been caused by the procedures involved in the research); and

- Suggests that the research places participants or others at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or recognized.

8.4.2 UNANTICIPATED PROBLEM REPORTING

The investigator will report unanticipated problems (UPs) to the reviewing Institutional Review Board (IRB) and to the Sponsor. The UP report will include the following information:

- Protocol identifying information: protocol title and number, PI's name, and the IRB project number;
- A detailed description of the event, incident, experience, or outcome;
- An explanation of the basis for determining that the event, incident, experience, or outcome represents an UP;
- A description of any changes to the protocol or other corrective actions that have been taken or are proposed in response to the UP.

To satisfy the requirement for prompt reporting, UPs will be reported using the following timeline:

- UPs that are serious adverse events (SAEs) will be reported to the IRB and to the study sponsor within 7 days of the investigator becoming aware of the event.
- Any other UP will be reported to the IRB and to the study sponsor at the time of continuing review.

8.4.3 REPORTING UNANTICIPATED PROBLEMS TO PARTICIPANTS

Any UP that affects the risk/benefit ratio to subjects will prompt a revision to the informed consent document. All subjects will be notified of the additional risk and re-consented at their next visit, or earlier if the situation requires.

9 STATISTICAL CONSIDERATIONS

9.1 STATISTICAL HYPOTHESES

The research study hypothesis is that in the HBV-monoinfected patients, LDV and/or SOF treatment has antiviral activity against HBV. The corresponding endpoint to assess efficacy of treatment is the HBsAg level. We expect that after 12 weeks of treatment the decline in HBsAg from baseline to end of treatment will be at least 0.4 $\log_{10}$/mL.

The secondary endpoint to assess treatment's antiviral activity against HBV is the level of HBV DNA. The expected decline in HBV DNA for HBV-monoinfected patients after 12 weeks of LDV and/or SOF treatment between baseline to end of treatment will be at least 2 $\log_{10}$ IU/mL.

9.2 SAMPLE SIZE DETERMINATION

Group A:
A total of 10 eligible patients with CHB, not currently on oral antiviral therapy will be accrued. Sample size and power calculations were based on a paired t-test approach assuming at least 0.4 log_{10}/mL decline in in HBsAg between baseline (pre-LDV/SOF) and post-treatment.

A sample size of 10 will have an adequate 80% power to detect a difference in mean levels of HBsAg of at least 0.4, assuming a standard deviation of differences of 0.4 (based on preliminary data), and using a paired t-test with a 0.05 two-sided significance level.

Groups B, C & D:
A total of 15 subjects with chronic HBV (5 in each Group) is consistent with the number of subjects in proof-of-concept pilot studies evaluating antiviral effects of novel therapies for chronic HBV infection. No formal power and sample size calculations were done.

9.3 POPULATIONS FOR ANALYSES

The intent-to-treat analysis population will include all subjects who received at least one dose of study drug. All subjects on the study will be followed through end of study or censored at the date they were discontinued from study.

9.4 STATISTICAL ANALYSES

9.4.1 GENERAL APPROACH

Subject's on-study demographic and clinical characteristics will be estimated and reported using the following summary statistics, means and standard deviations, medians and ranges, proportions and confidence intervals. Baseline parameters will include sex, self-identified race/ethnicity, age, body mass index, presence or absence of cirrhosis, HBsAg, HBV DNA level, HBV genotype and for group B, active HBV treatment.

9.4.2 ANALYSIS OF THE PRIMARY EFFICACY ENDPOINT(S)

The mean (standard deviation) of HBsAg will also be reported at baseline and at post-treatment. The primary efficacy analysis will determine individual difference in HBsAg from baseline (Day 0) to post-treatment using a paired *t*-test. HBsAg (log_{10}/mL) will be expressed means ± standard deviation (SD).

9.4.3 ANALYSIS OF THE SECONDARY ENDPOINT(S)

Safety will be evaluated by assessment of clinical laboratory test, physical examinations, and vital signs measurements at various time points during the study, as well as the documentation of AEs. A treatment-emergent AE will be defined as any new or worsening adverse event that beings on or after the date of first dose of study drug up to the date of last dose of study drug plus 30 days.

The toxicity data will be monitored closely throughout the study for all 25 enrolled subjects. AEs will be summarized based on the date of onset of the event for each subject. Addressing the safety of LDV and SOF, a maximum width 95% confidence interval for any grade 3 or higher toxicity will be about 40%. For 25 patients in the study, if the true unknown probability of a rare toxicity is 10%, the probability of observing 1 or more toxicities is 93%, for 5% it is 74%, and, if the true toxicity rate is 3%, then the probability of observing one or more rare toxicities is 53%.

9.4.4 SAFETY ANALYSES

See 9.4.3

9.4.5 BASELINE DESCRIPTIVE STATISTICS

Not Applicable

9.4.6 PLANNED INTERIM ANALYSES

There is no planned interim analysis.

9.4.7 SUB-GROUP ANALYSES

Not Applicable

9.4.8 TABULATION OF INDIVIDUAL PARTICIPANT DATA

Subject specific data will be tabulated by time point for HBV DNA and quantitative HBsAg.

9.4.9 EXPLORATORY ANALYSES

The exploratory endpoints may include immunologic changes, such as, change from baseline in cytokine profile, relationship between pharmacodynamics biomarkers and changes in viral parameters including HBV RNA and HBV core related antigen during and after treatment with LDV and/or SOF. The exploratory analysis will determine individual difference in the exploratory endpoints from baseline (Day 0) to post-treatment using a paired t-test. All inferential test will be indicated statistical significance with a P-value of less than 0.05 (two-sided).

10 SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

10.1 REGULATORY, ETHICAL, AND STUDY OVERSIGHT CONSIDERATIONS

10.1.1 INFORMED CONSENT PROCESS

10.1.1.1 CONSENT/ASSENT AND OTHER INFORMATIONAL DOCUMENTS PROVIDED TO PARTICIPANTS

Consent forms describing in detail the study intervention, study procedures, and risks are given to the participant and written documentation of informed consent is required prior to starting intervention/administering study intervention. The following consent materials are submitted with this protocol: screening consent, study consent, HIPAA consent.

10.1.1.2 CONSENT PROCEDURES AND DOCUMENTATION

Informed consent is a process that is initiated prior to the individual agreeing to participate in the study and continues throughout the individual's study participation. Consent forms will be Institutional Review Board (IRB)-approved and the participant will be asked to read and review the document. The investigator will explain the research study to the participant and answer any questions that may arise. A verbal explanation will be provided in terms suited to the participant's comprehension of the purposes, procedures, and potential risks of the study and of their rights as research participants. Participants will have the opportunity to carefully review the written consent form and ask questions prior to signing.

This study will utilize both screening and study consents. The participants should have the opportunity to discuss the study consent with their family or surrogates or think about it prior to agreeing to participate, and may do the same with screening. The participant will sign the screening informed consent document prior to any procedures being done specifically for the screening, and the study consent prior to administration of the study drug. Participants must be informed that participation is voluntary and that they may withdraw from the study at any time, without prejudice. Copies of the informed consent documents will be given to the participants for their records. The informed consent process will be conducted and documented in the source document (including the date), and the screening form signed, before the participant undergoes any study-specific procedures. The rights and welfare of the participants will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

Additionally, employees or students will be made aware that employment status or academic standing will not be affected by your participation or non-participation in this study. Students or employees will not be consented by anyone in a supervisory role.

Non-English-Speaking Participants
If a non-English speaking participant is unexpectedly eligible for enrollment, the participant will be provided with the UMB Short Written Consent Form for Non-English Speaking Research Participants in the participant's native language and a verbal explanation of the purpose, procedures and risks of the

study. The IRB-approved English consent form will serve as basis for the verbal explanation of the study. The investigator will obtain an interpreter, either in person or by phone translation service, unless the investigator is fluent in the prospective participant's language. Preferably, the interpreter will be someone who is independent of the participant (i.e., not a family member).

The IRB-approved English consent form will be signed by the investigator obtaining consent and a witness to the oral presentation. The UMB Short Written Consent Form will be signed by the participant and a witness who observed the presentation of information. The interpreter may sign the consent document as the witness and, in this case, will note, "Interpreter" under the signature line. A copy of both signed forms will be provided to the participant to take home. The investigator obtaining consent will document the consent process in the participant's medical record. Further, all instances of use of the UMB Short Written Consent Form will be reported to the IRB at the time of annual review.

Illiterate Subjects
As the majority of the patient populations from which the study participants are drawn are literate, written consent will typically be provided for both screening and treatment consents. However, oral consent will be obtained for illiterate participants as consistent with UMB IRB Policy without separate IRB approval for each specific use. At Continuing Reviews, the UMB IRB will be informed of the number of illiterate participants who provided consent verbally. A non-study team witness will be present for the oral consenting process.

10.1.2 STUDY DISCONTINUATION AND CLOSURE

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided by the suspending or terminating party to study participants, investigator, funding agency, the Investigational New Drug (IND) sponsor and regulatory authorities. If the study is prematurely terminated or suspended, the Principal Investigator (PI) will promptly inform study participants, the Institutional Review Board (IRB), and sponsor and will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule.

Circumstances that may warrant termination or suspension include, but are not limited to:

- Determination of unexpected, significant, or unacceptable risk to participants
- Demonstration of efficacy that would warrant stopping
- Insufficient compliance to protocol requirements
- Data that are not sufficiently complete and/or evaluable
- Determination that the primary endpoint has been met
- Determination of futility

Study may resume once concerns about safety, protocol compliance, and data quality are addressed, and satisfy the sponsor, SMC, IRB and/or Food and Drug Administration (FDA) as appropriate.

10.1.3 CONFIDENTIALITY AND PRIVACY

Participant confidentiality and privacy is strictly held in trust by the participating investigators, their staff, and the sponsor. This confidentiality is extended to cover testing of biological samples and genetic

tests in addition to the clinical information relating to participants. Therefore, the study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the sponsor.

All research activities will be conducted in as private a setting as possible.

The study monitor, other authorized representatives of the sponsor, representatives of the Institutional Review Board (IRB), regulatory agencies or pharmaceutical company supplying study product may inspect all documents and records required to be maintained by the investigator, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at each clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the reviewing IRB, Institutional policies, or sponsor requirements.

Study participant research data, which is for purposes of statistical analysis and scientific reporting, will be entered into the REDCap database for future analysis. This will not include the participant's contact or identifying information. Rather, individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by the IHV research staff will be secured and password protected. At the end of the study, all study databases will be de-identified and archived at the IHV.

10.1.4 FUTURE USE OF STORED SPECIMENS AND DATA

Data collected for this study will be analyzed and stored at University of Maryland, Institute of Human Virology. With the participant's approval within the consent and as approved by local Institutional Review Board (IRB), de-identified biological samples may be shared with Gilead Sciences, Inc. with the goal of sharing of data. These samples could be used to research immune response to hepatitis B, its complications and other conditions for which individuals with hepatitis B are at increased risk, and to improve treatment. Gilead Sciences, Inc. will also be provided with a code-link that will allow linking the biological specimens with the phenotypic data from each participant, maintaining the blinding of the identity of the participant.

During the conduct of the study, an individual participant can choose to withdraw consent to have optional biological specimens for future research stored, in which case they will be destroyed.

When the study is completed, access to study data and/or samples will be provided by Dr. Kottilil.

10.1.5 KEY ROLES AND STUDY GOVERNANCE

Principal Investigator
Joel Chua, MD; Assistant Professor
University of Maryland, Institute of Human Virology
725 W. Lombard Street
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10.1.6 SAFETY OVERSIGHT

Safety oversight will be under the direction of a Safety Monitoring Committee (SMC) composed of individuals with the appropriate expertise, including experience with the treatment and/or care of those with hepatitis B & C. At least two members of the SMC will be independent from the study conduct and free of conflict of interest, or measures should be in place to minimize perceived conflict of interest. The SMC will meet at least semiannually to assess safety and efficacy data. The SMC will provide its input to the study sponsor and PI.

10.1.7 CLINICAL MONITORING

Clinical site monitoring is conducted to ensure that the rights and well-being of trial participants are protected, that the reported trial data are accurate, complete, and verifiable, and that the conduct of the trial is in compliance with the currently approved protocol/amendment(s), with International Conference on Harmonisation Good Clinical Practice (ICH GCP), and with applicable regulatory requirement(s).

- Monitoring will begin as the first two subjects screen for the clinical trial. This will entail a clinical research nurse providing 100% verification of screening consent and data verification of the first two subjects. An independent SMC member will be available for consultation as needed for any medical concerns.
- Once subjects begin enrollment, consent and eligibility verification will continue at 100%, with 15% targeted subject data review.

10.1.8 QUALITY ASSURANCE AND QUALITY CONTROL

The IHV CRU will perform internal quality management of study conduct, data and biological specimen collection, documentation and completion. An individualized quality management plan will be developed.

Quality control (QC) procedures will be implemented beginning with the data entry system and data QC checks that will be run on the database will be generated. Any missing data or data anomalies will be addressed.

10.1.9 DATA HANDLING AND RECORD KEEPING

10.1.9.1 DATA COLLECTION AND MANAGEMENT RESPONSIBILITIES

All research data and results will be recorded using data collection forms. Source documents will support the data collected and will include, but not limited to; clinical findings and observations, laboratory and test data, hospital medical records, physician or office charts, physician or nursing notes, recorded data from automated instruments, x-rays, etc. Research data will be entered into a secure electronic database. REDCap tracks all data corrections made by authorized users, providing audit trails for monitoring or query.

10.1.9.2 STUDY RECORDS RETENTION

The investigator is responsible for retaining all essential documents listed in the ICH Good Clinical Practice (GCP) Guideline. All essential documentation for all study subjects is to be maintained by the investigators in a secure storage facility for a minimum of 5 years. The FDA requires study records to be retained for up to 2 years after marketing approval or disapproval (21 CFR 312.62), or until at least 2 years have elapsed since the formal discontinuation of clinical development of the investigational agent for a specific indication. These records are also to be maintained in compliance with IRB, state, and federal medical records retention requirements, whichever is longest. All stored records are to be kept confidential to the extent required by federal, state, and local law.

10.1.10 PROTOCOL DEVIATIONS

A protocol deviation is any noncompliance with the clinical trial protocol or International Conference on Harmonisation Good Clinical Practice (ICH GCP). The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly.

These practices are consistent with ICH GCP:

- 4.5 Compliance with Protocol, sections 4.5.1, 4.5.2, and 4.5.3
- 5.1 Quality Assurance and Quality Control, section 5.1.1
- 5.20 Noncompliance, sections 5.20.1, and 5.20.2.

It is the responsibility of the site investigator to use continuous vigilance to identify and report deviations that constitute Reportable New Information within 5 working days of identification of the protocol deviation as per local IRB requirements. The site investigator is responsible for knowing and adhering to the reviewing IRB requirements

10.1.11 PUBLICATION AND DATA SHARING POLICY

This trial will be registered at ClinicalTrials.gov, and results information from this trial will be submitted to ClinicalTrials.gov. In addition, every attempt will be made to publish results in peer-reviewed journals.

10.1.12 CONFLICT OF INTEREST POLICY

The independence of this study from any actual or perceived influence, such as by the pharmaceutical industry, is critical. Therefore, any actual conflict of interest of persons who have a role in the design, conduct, analysis, publication, or any aspect of this trial will be disclosed and managed. Furthermore, persons who have a perceived conflict of interest will be required to have such conflicts managed in a way that is appropriate to their participation in the design and conduct of this trial.

10.2 ADDITIONAL CONSIDERATIONS

Not Applicable

10.3 ABBREVIATIONS

Abbreviation	Term
AE	Adverse event
AI	Associate Investigator
AIDS	Acquired immune deficiency syndrome
ALT	Alanine transaminase
AST	Aspartate transaminase
CBC	Complete blood count
cccDNA	Covalently closed circular DNA
CFR	Code of Federal Regulations
CHB	Chronic Hepatitis B
CMP	Complete metabolic panel
CRF	Case report form
CRU	Clinical research unit
DC PFAP	DC Partnership for HIV/AIDS Progress
DNA	Deoxyribonucleic acid
DSPH	Gilead Drug Safety & Public Health
ECG	Electrocardiogram
ELISA	Enzyme-linked immunosorbent assay
ELISPOT	Enzyme-Linked ImmunoSpot
FDA	Food and Drug Administration
FDC	Fixed Dose Combination
GCP	Good Clinical Practices
HBV	Hepatitis B virus
HBcrAg	Hepatitis B core related antigen
HBeAg	Hepatitis B envelope antigen

HBsAg	Hepatitis B surface antigen
HgbA1c	Hemoglobin A1c
HCC	Hepatocellular carcinoma
hCG	Human chorionic gonadotropin
HCV	Hepatitis C virus
HIPAA	Health Insurance Portability and Accountability Act
HIV	Human immunodeficiency virus
ICF	Informed consent form
ICH	International Conference on Harmonization
IDU	Intravenous drug use
IFN-α	Interferon-α
IHV	Institute of Human Virology
IND	Investigational New Drug
IRB	Institutional Review Board
LDV	Ledipasvir
LLOQ	Lower Limit of quantification
MTA	Material transfer agreement
NA	Nucleoside or nucleotide analogue
NCT	National Clinical Trial
NHANES	National Health and Nutrition Examination Survey
NIH	National Institutes of Health
NRTIs	Nucleos(t)ide reverse transcriptase inhibitors
OAV	Oral antivirals
OHRP	Office for Human Research Protections
PBMC	Peripheral blood mononuclear cells
PCR	Polymerase chain reaction
PEG-IFN-α	Pegylated interferon-α
PI	Principal Investigator
QA	Quality assurance
QC	Quality control
RNA	Ribonucleic acid
SAE	Serious adverse event
SAR	Suspected adverse reaction
SC	Study Coordinator
SMC	Safety monitoring committee
SoA	Schedule of Activities
SOF	Sofosbuvir
SOP	Standard operating procedure
TAF	Tenofovir alafenamide fumarate
TDF	Tenofovir disoproxil fumarate

UA	Urinalysis
ULN	Upper limit of normal
UMB	University of Maryland Baltimore
UMSOM	University of Maryland School of Medicine
UP	Unanticipated problem
UPnonAE	Unanticipated problem that is not an adverse event
WBC	White blood cell

10.4 PROTOCOL AMENDMENT HISTORY

Version	Affected Sections	Description of Change	Brief Rationale
1.1 (10 Oct 2017)	-	Updated study background, rationale, sample storage & references	Respond to IRB requests prior to approval
2.0 (05 Jan 2018)	Title Page	Updated IND number and removed KlinEra as Sponsor	To be consistent with FDA filing and IND decisions
3.0 (04 Apr 2018)	SoA	Added qualifier removing need to repeat Hepatitis B genotype if on file	Unless new risk behavior identified, genotype will remain constant.
4.0 (04 Apr 2018)	SoA	Added qualifier removing need to repeat Hepatitis B genotype if on file.	Unless new risk behavior identified, genotype will remain constant.
5.0 (01 Nov 2018)	All	Addition of 3 study treatment groups, increase total N. Addition of two sites, CCACC and Unity Parkside.	To explore effects of each drug individually and combined effect on quantitative HBsAg in virally suppressed HBV patients.
6.0 (18 Mar 2019)	SoA, Eligibility Criteria, Title Page	Addition of lipase & creatine kinase labs. Clarification of inclusion OAVs and exclusion of interferon. Add IND#.	Changes made per FDA recommendations.

11 REFERENCES

1. Migration Policy Institute [Internet}. Washington DC. [cited 2017 July 1]. Available from: *http://www.migrationpolicy.org/programs/data-hub/us-immigration-trends?gclid=CITxl8uuvM0CFcUmhgodz4wEDQ*.
2. Brady, J.E., et al., Estimating the prevalence of injection drug users in the U.S. and in large U.S. metropolitan areas from 1992 to 2002. J Urban Health, 2008. 85(3): p. 323-51.
3. Lok, A.S. and B.J. McMahon, Chronic hepatitis B: update 2009. Hepatology, 2009. 50(3): p. 661-2
4. European Association For The Study Of The Liver., EASL Clinical Practice Guidelines: management of chronic hepatitis B. J Hepatol, 2009. 50(2): p. 227-42.
5. Yuen, M.F., et al., HBsAg Seroclearance in chronic hepatitis B in Asian patients: replicative level and risk of hepatocellular carcinoma. Gastroenterology, 2008. 135(4): p. 1192-9.
6. Terrault, N.A., et al., AASLD guidelines for treatment of chronic hepatitis B. Hepatology, 2016. 63(1): p. 261-83.
7. Kwon, H. and A.S. Lok, Hepatitis B therapy. Nat Rev Gastroenterol Hepatol, 2011. 8(5): p. 275-84.
8. Ott, J.J., et al., Global epidemiology of hepatitis B virus infection: new estimates of age-specific HBsAg seroprevalence and endemicity. Vaccine, 2012. 30(12): p. 2212-9.
9. Lavanchy, D., Hepatitis B virus epidemiology, disease burden, treatment, and current and emerging prevention and control measures. J Viral Hepat, 2004. 11(2): p. 97-107.
10. Dienstag, J.L., Hepatitis B virus infection. N Engl J Med, 2008. 359(14): p. 1486-500.
11. Mitchell, T., et al., The increasing burden of imported chronic hepatitis B--United States, 1974-2008. PLoS One, 2011. 6(12): p. e27717.
12. Roberts, H., et al., Prevalence of chronic hepatitis B virus (HBV) infection in U.S. households: National Health and Nutrition Examination Survey (NHANES), 1988-2012. Hepatology, 2016. 63(2): p. 388-97.
13. Friedman, S.R., et al., Estimating numbers of injecting drug users in metropolitan areas for structural analyses of community vulnerability and for assessing relative degrees of service provision for injecting drug users. J Urban Health, 2004. 81(3): p. 377-400.
14. Torbenson, M., et al., High prevalence of occult hepatitis B in Baltimore injection drug users. Hepatology, 2004. 39(1): p. 51-7.
15. Harrell, P.T., et al., Latent classes of heroin and cocaine users predict unique HIV/HCV risk factors. Drug Alcohol Depend, 2012. 122(3): p. 220-7.
16. Whitehead, N.E., et al., Awareness of biologically confirmed HCV among a community residing sample of drug users in Baltimore City. J Community Health, 2014. 39(3): p. 487-93.
17. Marcellin, P., et al., Regression of cirrhosis during treatment with tenofovir disoproxil fumarate for chronic hepatitis B: a 5-year open-label follow-up study. Lancet, 2013. 381(9865): p. 468-75.
18. Liaw, Y.F., et al., Lamivudine for patients with chronic hepatitis B and advanced liver disease. N Engl J Med, 2004. 351(15): p. 1521-31.
19. Nassal, M., HBV cccDNA: viral persistence reservoir and key obstacle for a cure of chronic hepatitis B. Gut, 2015. 64(12): p. 1972-84.
20. Lok AS, Zoulim F, Dusheiko G, et al. Hepatitis B cure: from discovery to regulatory approval. J Hepatol. 2017; 67: p. 847-61.
21. Lucifora, J. and U. Protzer, Attacking hepatitis B virus cccDNA--The holy grail to hepatitis B cure. J Hepatol, 2016. 64(1 Suppl): p. S41-8.

22. Wang, M., et al., Serum hepatitis B surface antigen is correlated with intrahepatic total HBV DNA and cccDNA in treatment-naive patients with chronic hepatitis B but not in patients with HBV related hepatocellular carcinoma. J Med Virol, 2013. 85(2): p. 219-27.
23. Chevaliez S, Hezode C, Bahrami S, et al. Long-term hepatitis B surface antigen (HBsAg) kinetics during nucleoside/nucleotide analogue therapy: finite treatment duration unlikely. J Hepatol. 2013; 58: p. 676-83.
24. Zoutendijk R, Hansen BE, van Vuuren AJ, et al. Serum HBsAg decline during long-term potent nucleos(t)ide analogue therapy for chronic hepatitis B and prediction of HBsAg loss. J Infect Dis 2011; 204: p 415-8.
25. Höner Zu Siederdissen C, Maasoumy B, Cornberg M. What is new on HBsAg and other diagnostic markers in HBV infection? Best Pract Res Clin Gastroenterol. 2017; 31(3): p. 281-9.
26. Seto WK, Wong DK, Fung J. Reduction of hepatitis B surface antigen levels and hepatitis B surface antigen seroclearance in chronic hepatitis B patients receiving 10 years of nucleoside analogue therapy. Hepatology 2013; 58: p. 923-31.
27. Wursthorn K, Jung M, Riva A, et al. Kinetics of hepatitis B surface antigen decline during 3 years of telbuvidine treatment in hepatitis B e antigen-positive patients. Hepatology. 2010; 52: p. 1611-20.
28. Liu CJ, Chuang WL, Sheen IS, et al. Decline in HBsAg levels observed during treatment with ledipasvir/sofosbuvir (LDF/SOF) in patients with chronic hepatitis B virus (HBV) and hepatitis C virus (HCV) infection. Unpublished internal data, Gilead Sciences, Inc.
29. Loggi E, Gitto S, Galli S, et al. Hepatitis B virus reactivation among hepatitis C patients treated with direct-acting antiviral therapies in routine clinical practice. J Clin Virol. 2017; 93: p. 66-70.
30. Harvoni® Package Insert [Internet]. Gilead Sciences, Inc. 4/2017. [cited 2017 July 1]. Available from: *http://www.gilead.com/~/media/Files/pdfs/medicines/liver-disease/harvoni/harvoni_pi.pdf*.
31. Bersoff-Matcha SJ, Cao K, Jason M, et al. Hepatitis B virus reactivation associated with direct-acting antiviral therapy for chronic hepatitis C virus: a review of cases reported to the U.S. Food and Drug Administration Adverse Event Reporting System. Ann Intern Med. 2017; 166(11): 792-8.
32. Bini EJ, Perumalswami PV. Hepatitis B virus infection among American patients with chronic hepatitis C virus infection: Prevalence, racial/ethnic differences, and viral interactions. Hepatology. 2010; 51(3): 759-66.
33. Caccamo G, Saffioti F, Raimondo G. Hepatitis B virus and hepatitis C virus dual infection. World J Gastroenterol. 2014; 20(40): 14559-67.
34. Sovaldi® Package Insert [Internet]. Gilead Sciences, Inc. Revised version 11/2017.
35. Ledipasvir (GS-5885) Investigator's Brochure, 4th edition. Gilead Sciences, Inc. July 3, 2013.