Chronic Hepatitis B Virus Infection: Developing Drugs for Treatment

Guidance for Industry

U.S. Department of Health and Human Services

Food and Drug Administration

Center for Drug Evaluation and Research (CDER)

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Clinical/Antimicrobial

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I. INTRODUCTION

The purpose of this guidance is to assist sponsors in the clinical development of drugs and biologics for the treatment of chronic hepatitis B virus (HBV) infection from the initial investigational new drug application (IND) through the new drug application (NDA)/biologics license application (BLA) and postmarketing phases.[2] Sponsors are also encouraged to communicate with the Division of Antivirals through the pre-IND consultation program to obtain advice in the development of drugs with unique considerations based on mechanism of action, novel treatment approaches, or the use of novel biomarkers.[3]

This guidance does not address development of vaccines or blood-derived products, which are regulated by the Center for Biologics Evaluation and Research. This guidance also does not discuss general issues of statistical analysis or clinical trial design. Those topics are addressed in the ICH guidances for industry

[1] This guidance has been prepared by the Division of Antivirals in the Center for Drug Evaluation and Research at the Food and Drug Administration.

[2] For the purposes of this guidance, all references to *drugs* include both human drugs and therapeutic biological products unless otherwise specified.

[3] See the Division of Antivirals Pre-IND Letter of Instruction web page at https://www.fda.gov/drugs/pre-ind-consultation-program/division-anti-viral-dav-pre-ind-letter-instruction.

E9 Statistical Principles for Clinical Trials (September 1998) and *E10 Choice of Control Group and Related Issues in Clinical Trials* (May 2001), respectively.[4]

The contents of this document do not have the force and effect of law and are not meant to bind the public in any way, unless specifically incorporated into a contract. This document is intended only to provide clarity to the public regarding existing requirements under the law. FDA guidance documents, including this guidance, should be viewed only as recommendations, unless specific regulatory or statutory requirements are cited. The use of the word *should* in Agency guidances means that something is suggested or recommended, but not required.

II. BACKGROUND

HBV is an enveloped DNA virus belonging to the *Hepadnavirus* family. The highly stable covalently closed circular viral DNA (cccDNA) functions as a nonreplicative minichromosome and persists throughout the lifespan of infected hepatocytes. The cccDNA is not eliminated by currently approved therapies that include drugs from the nucleoside/nucleotide reverse transcriptase inhibitor (NrtI) class, and pegylated interferon.

Chronic HBV (CHB) infection results in progressive liver disease ranging from asymptomatic to severe disease with complications including cirrhosis, liver failure, and the development of hepatocellular carcinoma (HCC). In untreated adults with CHB, the cumulative 5-year incidence of cirrhosis is 8 to 20 percent; and among those with cirrhosis, the 5-year cumulative risk of hepatic decompensation is 20 percent, and risk of HCC is 2 to 5 percent (Terrault et al. 2016). An effective vaccine and antiviral therapies are approved for the prevention of HBV infection and treatment of CHB, respectively.

Currently available therapies achieve sustained suppression of HBV DNA while on treatment, but rates of HBV surface antigen (HBsAg) loss with or without seroconversion to anti-HBsAg (HBsAb) remain low. Sustained HBV DNA suppression is associated with serum alanine aminotransferase (ALT) normalization and improvement in liver histology including regression of hepatic fibrosis and cirrhosis (Chang et al. 2010; Marcellin et al. 2013; Buti et al. 2015). Effective antiviral therapy for CHB reduces disease-related complications, such as hepatic decompensation and liver failure, and decreases risk of HCC (Lok et al. 2016; Papatheodoridis et al. 2017). Clearance of HBsAg is associated with reduced risk of hepatic decompensation and improved survival (Terrault et al. 2016). HBsAg loss is considered the best predictor of sustained remission off-treatment (Terrault et al. 2016). New finite duration therapies target

[4] We update guidances periodically. For the most recent version of a guidance, check the FDA guidance web page at https://www.fda.gov/regulatory-information/search-fda-guidance-documents.

treatment regimens that can achieve sustained suppression of HBV DNA off-treatment with HBsAg loss (with or without HBsAb seroconversion) with low risk of virologic relapse (as defined by HBV DNA) and minimal risk of liver disease progression after the treatment is stopped (Lok et al. 2017).

III. DEVELOPMENT PROGRAM

A. General Drug Development Considerations

This section discusses nonclinical and early phase clinical development considerations, followed by issues related to the target population for drug development, assessment of activity in early phase trials, and safety considerations.

1. Early Phase Development Considerations

Early clinical evaluation should provide sufficient data to establish safety and evidence of antiviral activity to support the phase 3 trials.

a. Pharmacology/toxicology development considerations

Pharmacology/toxicology development considerations for single investigational drugs intended to treat CHB should follow the approaches outlined in existing guidances for drug development.[5]

When a combination of two or more early stage investigational drugs to treat CHB is being developed, sponsors should discuss with the FDA whether combination toxicology studies are needed (including the design of such studies) to support clinical trials for the intended combination. When combination toxicology studies are conducted, usually no more than two drugs should be tested simultaneously in a particular arm of a toxicology study. Nonclinical combination studies of an investigational drug plus an approved drug or licensed biological product generally are not needed unless data from nonclinical studies of an investigational drug suggest a potential for serious synergistic toxicity with an approved drug or licensed biological product.

In general, sponsors developing drugs intended to treat CHB with proposed treatment durations of 6 months or more should conduct carcinogenicity studies.[6] Sponsors should submit carcinogenicity studies with an initial NDA. However, under limited circumstances and with prior written agreement, the FDA may consider allowing sponsors to submit the completed carcinogenicity studies during the postmarketing period under section 505(o)(3) of the Federal Food, Drug, and Cosmetic Act (FD&C Act)[7], as long as the studies have been initiated before NDA submission.

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[5] See the ICH guidances for industry *M3(R2) Nonclinical Safety Studies for the Conduct of Human Clinical Trials and Marketing Authorization for Pharmaceuticals* (January 2010) and *S6(R1) Preclinical Safety Evaluation of Biotechnology-Derived Pharmaceuticals* (May 2012).

[6] See the ICH guidance for industry *S1A The Need for Long-Term Rodent Carcinogenicity Studies of Pharmaceuticals* (March 1996).

[7] See also the draft guidance for industry *Postmarketing Studies and Clinical Trials—Implementation of Section 505(o)(3) of the Federal Food, Drug, and Cosmetic Act* (October 2019). When final, this guidance will represent the FDA's current thinking on this topic. For the most recent version of a guidance, check the FDA guidance web page at https://www.fda.gov/regulatory-information/search-fda-guidance-documents.

Sponsors developing biological products should follow approaches outlined in ICH S6(R1) and discuss their proposals for a carcinogenicity risk assessment with the FDA during clinical development to facilitate a final assessment needed to support a BLA.

b. Nonclinical virology development considerations

Sponsors should consider recommendations for general antiviral drug development in the draft guidance for industry *Antiviral Product Development—Conducting and Submitting Virology Studies to the Agency* (February 2014).[8] The FDA encourages detailed study reports describing the mechanism of action, antiviral activity in cell culture, cytotoxicity, and mitochondrial toxicity (Marroquin et al. 2007; Arnold et al. 2012), animal models,[9] and resistance studies. Additionally, sponsors are advised to provide the following nonclinical virology data for investigational drugs developed specifically for the treatment of CHB.

Resistance and cross-resistance

HBV is differentiated into 10 genotypes (A through J) with many different subtypes. Drugs targeting viral proteins or sequence targets will likely be impacted by genotype- and subtype-specific variations that occur in the drug target as well as variations that evolve in the presence of the drug that allow the virus to develop resistance. Therefore, characterizing resistance pathways and their relationship to HBV genomic variations helps identify subjects who will benefit from the drug and also helps identify potential cross-resistance with other drugs having a similar mechanism of action.

HBV does not generally grow well enough in cell culture to select for resistant virus in the presence of drug. When possible, we recommend that resistance assessments be performed for all animal studies that assess the antiviral activity of an investigational drug in infected animals receiving the drug that experience viral breakthrough. In addition, a resistance monitoring plan for identifying changes in viral targets associated with treatment failure should be included in the protocols or can be submitted separately for each clinical trial that will treat subjects with CHB.

- Amino acid substitutions or nucleotide mutations associated with the development of resistance (i.e., changes from baseline) to an investigational drug should be determined by sequencing the drug target and validated by introducing resistance-associated substitutions or mutations into the HBV reference genome using site-directed mutagenesis and determining the fold-shift in

[8] When final, this guidance will represent the FDA's current thinking on this topic.

[9] We support the principles of the 3Rs (reduce/refine/replace) for animal use in testing when feasible. FDA encourages sponsors to consult with review divisions when considering a nonanimal testing method believed to be suitable, adequate, validated, and feasible. FDA will consider if the alternative method could be assessed for equivalency to an animal test method.

susceptibility. Results from these studies help identify resistance pathways and support the drug's proposed mechanism of action. Lack of a shift in susceptibility does not exclude a resistance association for a specific substitution or mutation that occurs in two or more independent events.

Cross-resistance should be assessed to determine if resistance against approved HBV drugs with the same target confers resistance to the drug being developed and vice versa. The development of cross-resistance to epitopes of approved HBV vaccines should be assessed.

Considerations for oligonucleotide-based investigational drugs

Knockdown of viral protein expression via oligonucleotide-based therapeutics is an active area for the development of antiviral drugs. These drugs, which have a nucleic acid target, present potential off-target binding at mismatched sequences that could lead to species-specific toxicities not necessarily detected in toxicology studies. Therefore, we recommend that sequence-dependent off-target assessments be conducted for such drugs using appropriate in silico and in vitro methodologies to identify potential off-target mismatches that can be investigated and potentially monitored during clinical development. The Division recognizes that many additional factors, such as temporal and cell type specific expression, pharmacokinetic (PK) properties, and hybridization-dependent efficiencies may be considered in the overall risk assessment of potential off-target effects (refer to Lindow et al. 2012 for a general approach). The following points should be considered when designing sequence-dependent off-target assessments:

- Several different classes of oligonucleotide-based therapeutics (small interfering RNAs and antisense oligonucleotides, etc.) exist. The criteria for defining high-risk off-target mismatches are expected to differ for each class; appropriate criteria should be provided and justified for each drug.

- All elements of the oligonucleotide drug product that are potentially available for mismatch binding should be assessed, including both the sense and antisense strands, overlapping ends, etc.

- There are multiple in silico approaches that may be considered, and we recommend sponsors justify the methods and approaches used. In silico studies can be used to do the following:

  - Identify all potential off-target mismatches, regardless of tissue expression, in the human transcriptome, including the mitochondrial transcriptome; for each of these, describe available information on mouse knockouts and human genetic diseases. Sponsors should specify and justify the criteria for defining a potential off-target mismatch.

  - Determine whether each off-target site in the human transcriptome is conserved in the animal species in which antiviral assessments and pivotal toxicology studies are performed.

  - Determine the variation within the off-target mismatches in the transcriptomes of different ethnic populations in the United States, when feasible, to assess whether certain populations may be more susceptible to off-target effects than others.

- In vitro assessments of possible off-target binding (cell-based analysis, RNA-seq, etc.) should be conducted to confirm or eliminate the importance of each off-target mismatch determined via in silico assessments.

- An overall risk assessment of potential off-target effects should be provided along with the off-target assessment data.

Considerations for drugs developed to modulate innate and adaptive immune responses

Drugs developed to modulate innate and adaptive immune responses to chronic HBV infection are likely to target host factors and induce or repress immune biomarkers before having an impact on HBV replication or clearance of HBV infected hepatocytes. Nonclinical studies should describe the specific mechanism of action of the drug and demonstrate that immune modulation in cell culture and animal models of HBV infection results in suppression of HBV replication, as measured by HBV DNA or HBsAg loss, or that the HBV cccDNA reservoir is reduced by assessing cccDNA levels. In addition, given that these drugs may target host factors, it is important to determine that the target of the drug is conserved, having similar affinity between the animal species being assessed and the human target.

Targeting host factors

For drugs targeting host factors, polymorphisms in the gene encoding the target should be assessed to determine whether the drug will be more effective or less effective in different populations. Sponsors should identify key racial and ethnic groups in the United States who will be part of the proposed

indication at the initiation of drug development. If a nonclinical assay to assess the drug effect is available, multiple samples from each of these groups in the United States should be evaluated to determine whether race or ethnicity may be a factor contributing to efficacy (Forde et al. 2013). Samples should be collected during clinical trials to determine the virus genotype of subjects who respond less favorably to treatment.

c. Clinical pharmacology considerations

In general, dose selection for early efficacy trials should be predicted to provide plasma drug exposures that exceed by severalfold the protein binding-adjusted, cell culture EC_{50} value of the drug for the relevant HBV genotype/subtype. In cases where either total (protein bound plus unbound) or free (unbound) drug concentrations in plasma may not be related to antiviral activity, sponsors may provide justification for dose selection based on drug concentration in other relevant tissues. The dose selection should also consider the safety data from the previous phase 1 trials and animal studies.

Sponsors should refer to the appropriate clinical pharmacology guidances for industry to inform the need and design of drug-drug interaction studies and PK studies in patients with renal or hepatic impairment.[10] We encourage sponsors to conduct these studies, if needed, early in development to inform the management of drug interactions and the inclusion of patients with renal and hepatic impairment in phase 3 trials as appropriate. See section III. B. 6., Dose Selection, for dose selection for phase 2 and 3 trials and section III. C. 2.,

Pharmacokinetic/Pharmacodynamic Considerations, for other PK and pharmacodynamic considerations.

2. Drug Development Population

Therapies should be developed for use in a wide range of patients with CHB, including pediatric populations.

Early phase clinical trials should focus on the adult population without cirrhosis. Initial trials can be conducted in HBV e antigen positive (HBeAg-positive) or HBV e antigen negative (HBeAg-negative)

[10] See the guidance for industry *Pharmacokinetics in Patients With Impaired Hepatic Function: Study Design, Data Analysis, and Impact on Dosing and Labeling* (May 2003). See also the guidances for industry *Clinical Drug Interaction Studies—Cytochrome P450 Enzyme- and Transporter-Mediated Drug Interactions* (January 2020), *In Vitro Drug Interaction Studies—Cytochrome P450 Enzyme- and Transporter-Mediated Drug Interactions* (January 2020), and the draft guidance for industry *Pharmacokinetics in Patients With Impaired Renal Function—Study Design, Data Analysis, and Impact on Dosing* (September 2020). When final, this guidance will represent the FDA's current thinking on these topics.

patients with active disease who are treatment-naïve, or those who were previously treated but currently viremic, off-treatment. Trials can also be conducted in HBeAg-positive or HBeAg-negative patients who are virally suppressed on NrtIs. In addition to endpoints discussed in section III. B., sponsors can evaluate exploratory endpoints in early phase trials to gather data to inform and support the choice of appropriate endpoints in late phase trials, particularly those evaluating treatments of finite durations. Some of these exploratory endpoints may include the following:

- Change in quantitative HBsAg (qHBsAg) concentration at various time points on treatment
- Quantitative HBeAg levels
- Quantitative HBV RNA levels
- Quantitative HBV core-related antigen (HBcrAg) levels
- cccDNA quantification
- Quantitative HBsAg levels from integrated HBV genome fragments
- Quantitative HBsAg-anti-HBs immune complex levels

Also depending on the drug's mechanism of action, liver biopsy findings can be used in certain proof-of-concept studies to further understand the effect of the drug on the cccDNA reservoir and/or to help better understand potential surrogate markers of antiviral activity.

CHB is a global disease, and clinical trials are often conducted in multiple countries. FDA will accept a well-designed and well-conducted foreign clinical study not conducted under an IND as support for an IND or application for marketing approval if the trial was conducted in accordance with good clinical practice and if the FDA is able to validate the data from the trial through an onsite inspection if the Agency deems it necessary.[11] When sponsors rely on foreign data, these should be supported with information about predominant HBV genotypes and subtypes in the region or regions (Schweitzer et al. 2015). Development programs should include a sufficient number of U.S. patients to ensure prevalent genotypes in the U.S. population are represented. FDA strongly encourages sponsors to provide a plan to address inclusion of clinically relevant subpopulations with regard to age, sex, race, and ethnicity in the clinical trials to support an NDA or BLA, no later than the end-of-phase 2 meeting.[12]

[11] See 21 CFR 312.120; 21 CFR 314.106.

[12] See the guidance for industry *Collection of Race and Ethnicity Data in Clinical Trials* (October 2016).

3. Safety Considerations

In general, we recommend that initial marketing applications for drugs intended to treat CHB contain a total safety database of about 1,000 to 1,500 subjects exposed to the proposed dose and duration of treatment. Depending on the drug safety profile and concerns identified during the development process, a larger database or long durations of posttreatment follow-up may be needed.

In addition to routine safety monitoring, the safety monitoring plans should consider the mechanism of action of the drug, as there may be different safety concerns associated with specific drugs both during treatment and after treatment cessation. Some of the notable safety concerns associated with immune modulatory therapies are immune-mediated hepatitis flares and autoimmunity (e.g., immune-related adverse events observed with checkpoint inhibitors). The specific criteria for defining and monitoring for hepatitis flares during treatment and after stopping therapy should be prespecified in the clinical trial protocols. The safety profile observed in early phase trials should guide the monitoring plan for late phase trials.

Severe acute exacerbations of HBV infection may occur after discontinuation of anti-HBV therapy, particularly in the absence of HBsAg loss. Subjects should be monitored closely with both laboratory and clinical follow-up after discontinuation of anti-HBV therapy. The duration of follow-up should consider the mechanism of action and the half-life of the specific investigational drug. In certain circumstances, resumption of anti-HBV therapy may be warranted. The detailed plan and criteria for treatment reinitiation should be prespecified in the clinical trial protocols.

Clinical trial protocols should include predefined algorithms for data collection in the setting of significant hepatic events to ensure that the relevant data are available for further assessment and adjudication of these cases to differentiate between potential etiologies. The outcomes for all serious hepatic events should be systematically evaluated during clinical development. Evaluation by an independent adjudication committee is encouraged.

For a drug approved for use in patients without cirrhosis or with compensated cirrhosis, the database needed to extend use to the decompensated cirrhotic population would depend on the safety profile of the investigational drug and the overall benefit-risk profile for the indicated population. Similarly, obtaining safety data in other subpopulations, such as in patients coinfected with hepatitis D virus (HDV), may be important for certain clinical development programs. We encourage sponsors to discuss with the FDA safety-related considerations, including but not limited to the size of the safety database, before the initiation of phase 3 trials.

B. Phase 3 Efficacy Trial Considerations

Sponsors can submit an NDA/BLA to support marketing approval of a drug in a single patient population (e.g., treatment-naïve patients or patients who are virally suppressed on NrtIs). Such an application should include at least two adequate and well-controlled trials conducted in the proposed population. Alternatively, sponsors can choose to pursue an indication for different populations (e.g., a trial in treatment-naïve subjects and a second trial in subjects who are virally suppressed on NrtIs). In these situations, the NDA/BLA should contain at least one adequate and well-controlled trial in each patient population, with adequate supporting data.

1. Trial Design

Randomized controlled trials are recommended to establish efficacy because of the heterogeneity of the natural course of CHB. Appropriate trial designs depend on whether the therapeutic is intended for chronic suppressive therapy or therapy of finite duration as discussed below.

a. Chronic suppressive therapy

Sponsors developing drugs for chronic suppressive therapy of CHB should consider the following trial design options:

- A randomized controlled noninferiority (NI) (or superiority) trial comparing the investigational drug with an approved active control arm—The primary efficacy endpoint should be undetectable HBV DNA[13] after 48 weeks on-treatment in HBeAg-positive subjects, HBeAg-negative subjects, or both. The active comparator should be an antiviral drug that is recommended for treatment of CHB and reflects current practice at the time of trial initiation. The patient population may be treatment-naïve or previously treated subjects with detectable HBV DNA. Potential concerns related to the development of resistance when evaluating an investigational drug with a low barrier to resistance as a monotherapy should be addressed. For an NI design, determination of the NI margin should be discussed with the FDA.

[13] Defined as less than the lower limit of quantification (LLOQ), specifying if the results are target detected (TD) or target not detected (TND).

- A randomized controlled *add-on* superiority trial comparing an investigational drug plus an approved NrtI with an NrtI alone—However, for this trial design, it is not clear what the most appropriate primary endpoint should be to demonstrate the investigational drug's clinical contribution. Presently, HBV DNA is not recommended as a primary endpoint for an add-on trial because it is unclear what incremental numerical benefits in HBV DNA, over the substantial HBV DNA suppression achieved by an NrtI alone, are predictive of clinical benefit. Other endpoints such as HBsAg clearance or another surrogate found to be predictive of clinical benefit may be considered; however, the trial design and endpoints should be discussed with the FDA in advance of trial initiation.

b. Finite duration therapy

The appropriate trial design depends on the patient population being studied and the treatment regimen being evaluated. See section III. B. 7., Efficacy Endpoints for a description of efficacy endpoints that could be used for evaluation of finite duration therapies in clinical trials.

Sponsors developing drugs for finite duration therapy of CHB should consider the following trial design options:

Virally suppressed on NrtIs

To evaluate the primary efficacy outcome of sustained HBV DNA suppression off-treatment with HBsAg loss in subjects with active disease (HBeAg-positive or HBeAg-negative CHB) who are virally suppressed on NrtIs, sponsors can consider an add-on superiority trial against placebo with a current NrtI treatment regimen as the background therapy. The co-primary efficacy endpoints[14] of HBsAg loss and sustained HBV DNA suppression should be assessed at the 6-month posttreatment time point with additional follow-up to monitor for durability of response (i.e., sustained HBV DNA suppression and HBsAg loss) off-treatment. In general, off-treatment refers to discontinuation of all therapies (i.e., investigational agent and background NrtI regimen). To demonstrate sustained response off-treatment, sponsors should systematically assess the duration of a treatment consolidation period, defined as the duration of continued treatment needed after achieving HBsAg loss, during late phase trials. This may vary based on the mechanism of action and half-life of the specific investigational drug.

[14] See draft guidance for industry *Multiple Endpoints in Clinical Trials* (January 2017). When final, this guidance will represent the FDA's current thinking on this topic.

Sponsors should use the following criteria for stopping NrtI therapy at the end of the investigational treatment period when evaluating a finite duration therapy: (1) applied equally across treatment arms, as applicable; (2) well-defined in the protocol; and (3) stringent, such as HBsAg loss or substantial HBsAg decline or marked reduction in other important biomarkers identified in phase 2 trials. It is expected that few subjects would meet such criteria on the placebo arm. The criteria for stopping NrtI therapy should be based on clinical evidence that also reflects current practice guidelines recommended by the authoritative scientific bodies to ensure that the discontinuation of NrtI therapy does not pose undue safety risk to the trial participants. Sponsors should discuss with the FDA the use of biomarkers as a trigger for treatment interruption in advance of trial initiation.

Alternatively, an outcome of sustained HBV DNA suppression off-treatment without HBsAg clearance can be evaluated after a finite treatment duration using a superiority trial design comparing the investigational drug plus an NrtI with an NrtI alone.

Treatment-naïve

An outcome of sustained HBV DNA suppression off-treatment with HBsAg loss can be evaluated in treatment-naïve subjects by demonstrating superiority to an active control, or, in patients in whom treatment is currently not indicated per treatment guidelines, superiority to placebo. In certain patient populations (e.g., for patients in the immune-tolerant phase with mild necroinflammation or fibrosis) comparison with placebo may be feasible as current treatment guidelines do not recommend treatment for these patients. In some of the trial design scenarios, subjects in the placebo group may be rolled over to an active investigational drug before the completion of the trial based on a prespecified interim analysis. Sponsors should prospectively plan these analyses[15] and should discuss with the FDA before trial enrollment.

Sponsors considering an NI trial design should discuss in advance with the FDA their trial designs and justifications of the proposed NI margin based on historical evidence of treatment effect of the active control. In general, the active comparator in an NI trial should be an FDA-approved drug that is considered the standard of care for the specific indication and population being studied. A detailed protocol and statistical analysis plan (SAP) should be submitted for review.

[15] See the guidance for industry *Adaptive Designs for Clinical Trials of Drugs and Biologics* (November 2019).

2. Trial Population

Patients fulfilling one of the following two criteria for CHB should be enrolled (Centers for Disease Control and Prevention 2012):

(1) Negative immunoglobulin M (IgM) antibodies to HBV core antigen and a positive result on one of the following tests: HBsAg, HBeAg, or nucleic acid test for hepatitis B virus DNA (including qualitative, quantitative, and genotype testing); or

(2) Positive HBsAg result or positive nucleic acid test for HBV DNA (including qualitative, quantitative, and genotype testing) or positive HBeAg on two occasions at least 6 months apart (any combination of these tests performed 6 months apart is acceptable).

Sponsors should consider evaluating drug efficacy in key CHB subpopulations, including but not limited to the following:

- HBeAg-positive and HBeAg-negative subjects
- Subjects with cirrhosis
- Subjects with decompensated liver disease

3. Entry Criteria

The presence or absence of cirrhosis at study entry should be documented based on clinical assessment or previous liver biopsy, when available. As the criteria for diagnosing cirrhosis using noninvasive methods are not yet well defined, particularly in patients who are virally suppressed, the use of a noninvasive modality to define presence or absence of cirrhosis in a trial protocol should be supported by references that summarize performance characteristics and sensitivity and specificity of the modality for identifying patients with cirrhosis.

4. Randomization, Stratification, and Blinding

Sponsors should conduct randomized, double-blind trials whenever feasible to reduce the likelihood of potential biases. In general, trials should be designed to evaluate the effect of investigational therapies in subjects with key disease characteristics. Patient subpopulations could be evaluated in separate trials or as separate strata in a single trial. If multiple patient populations are included in a single trial, sponsors should consider stratifying groups at randomization based on baseline variables (e.g., HBeAg status, HBsAg level, presence or absence of cirrhosis, and HBV DNA level) and ensure that an adequate number of subjects are in each stratum to provide informative data.

5. Specific Populations

a. Patients with HBV/HIV-1 coinfection

The overall treatment goals for patients with HBV/HIV coinfection remain identical to those described for the HBV-monoinfected population. Sponsors should consider including subjects coinfected with HIV in the clinical trials evaluating finite duration HBV therapies. In cases where there is a strong rationale for exclusion of patients with HBV/HIV coinfection, the rationale should be addressed in the trial protocol. The concurrent use of HIV antiretroviral drugs that are also effective against HBV (e.g., HIV nucleoside/nucleotide reverse transcriptase inhibitors) may have implications for treatment cessation when evaluating finite duration HBV therapies and possibly confound interpretation of efficacy outcome as subjects continue their HIV nucleoside/nucleotide reverse transcriptase inhibitor -based antiretroviral regimen. Because of the various interactions between HIV and HBV therapies, we recommend sponsors discuss their plans with and obtain feedback from the FDA.

b. Patients with HBV/HDV coinfection

Infection with HDV only occurs in the setting of concurrent HBV infection (Wranke and Wedemeyer 2016). According to the World Health Organization, an estimated 15 million to 20 million people worldwide are living with HBV/HDV coinfection (World Health Organization 2017). A meta-analysis reported a much higher worldwide HBV/HDV coinfection prevalence of 62 million to 72 million (Chen et al. 2019). Relative to HBV monoinfection, HBV/HDV coinfection leads to more severe liver disease resulting in a greater risk of cirrhosis, HCC, and hepatic decompensation/failure.

The ultimate goal of treatment in patients with HBV/HDV coinfection is clearance or long-term suppression of both viruses. CHB treatment leading to loss of HBsAg may ultimately lead to the clearance of HDV infection (Wranke and Wedemeyer 2016). HDV superinfection frequently leads to spontaneous suppression of HBV (Huang and Lo 2014), and the effect of specific HBV therapies on the

interplay between the two viruses cannot be predicted. The possibility of HBV reactivation needs to be considered when HDV is suppressed after HDV treatment initiation. To mitigate the risk of HBV reactivation and associated hepatitis flares, it is preferred to have patients on NrtI suppressive therapy before they receive HDV treatment. Recommendations for trials in patients with HBV/HDV coinfection are beyond the scope of this guidance and are included in a separate draft guidance.[16] Sponsors should discuss development plans directly with the FDA.

c. Pediatric patients

Pediatric assessments are required under section 505B of the FD&C Act as part of the overall drug development program for a "new active ingredient, new indication, new dosage form, new dosing regimen, or new route of administration,"[17] unless those assessments are waived.[18] Sponsors should discuss their plans for pediatric assessments with the review division and be aware of timing and content requirements for pediatric study plans under section 505B(e) of the FD&C Act. Sponsors are required to submit pediatric study plans no later than 60 days after an end-of-phase 2 meeting or such other time as may be agreed upon by the FDA and the sponsor.[19] Because progressive liver disease is uncommon in young children with HBV infection, it is generally not recommended to include patients younger than 2 years of age in most development programs. Further, treatment generally is not recommended in children younger than 2 years of age, as per current treatment guidelines (Terrault et al. 2016).

In general, pediatric clinical trials can be initiated after phase 2 adult data characterizing the safety profile and preliminary evidence of efficacy are available. Given the natural history of CHB infection, the dynamic relationship between viral replication and the host immune response in pediatric patients may differ from that observed in adults, hence extrapolation of efficacy may not be possible in all scenarios and could be considered on a case-by-case basis depending on the mechanism of action of the investigational drug or drugs and the pediatric age group being evaluated.

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[16] See the draft guidance for industry *Chronic Hepatitis D Virus Infection: Developing Drugs for Treatment* (November 2019). When final, this guidance will represent the FDA's current thinking on this topic.

[17] See section 505B(a)(1)(A) of the FD&C Act; 21 U.S.C. 355c(a)(1)(A).

[18] See section 505B(a)(5) of the FD&C Act.

[19] See section 505B(e)(2)(A)(ii) of the FD&C Act; see also the guidance for industry *Pediatric Study Plans: Content of and Process for Submitting Initial Pediatric Study Plans and Amended Initial Pediatric Study Plans* (July 2020).

In cases in which extrapolation of efficacy from adults to pediatrics is possible, after critical PK parameters of a drug are identified in adults, pediatric development programs can focus on identifying dosing regimen(s) that achieve pediatric exposures similar to adults to demonstrate effectiveness in pediatric populations in which treatment is indicated as per current treatment guidelines. Additional clinical data should be obtained to assess whether antiviral activity is comparable to that observed in adult trials. Early discussion with the FDA on the appropriate endpoints and pediatric trial design is encouraged.

In the absence of a serious safety signal in adults, sponsors should enroll adolescents[20] in phase 3 adult trials or conduct a dedicated adolescent trial concurrently with the phase 3 adult trials. Confirmatory PK, safety, and efficacy data from this age group should be included as part of the data included at the time of filing of the original NDA/BLA.[21]

Typically, the nonadolescent pediatric population (for the purpose of this guidance, 2 to younger than 12 years of age) is divided into several groups or cohorts according to age or weight for enrollment into trials. Weight, rather than age, is the preferred criterion for enrollment because dosing recommendations for most antiviral drugs are weight-based. In addition, within clinical studies, sponsors should enroll the cohorts in parallel rather than in series, unless a drug has a specific safety or drug disposition factor that warrants a different approach. Dose selection for pediatric treatment trials (e.g., phase 2 or 3) should be supported by PK data generated from the initial PK/safety phase of the pediatric study and results of available modeling and simulation. Sponsors should discuss the dose selection with the FDA before initiating the phase 2 or 3 pediatric trials.

The pediatric trials should also obtain data to support safety in pediatric populations. In general, a safety database of about 100 subjects receiving the proposed dose and treatment duration is recommended. The participants should be adequately distributed across the pediatric age group for which studies are required. Long-term follow-up data should be collected from pediatric trials to assess long-term safety as well as durability of treatment response. If clinical trials in adults have demonstrated differences in safety profile or dosing based on fibrosis stage, pediatric subjects should be assessed for presence or absence of cirrhosis using the most appropriate modality for each study location.

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[20] For the purpose of this guidance, *adolescents* are defined as age 12 to younger than 18 years of age.

[21] We note that, for applications to which section 505B applies, all pediatric assessments must be submitted with the application unless those assessments have been deferred (section 505B(a)(1)(A)).

Section 505B of the FD&C Act also mandates that the requisite pediatric assessments be conducted using a formulation of the drug that is appropriate for each pediatric group being studied.[22] Adult formulations generally are considered appropriate for adolescent patients (Momper et al. 2013), but younger patients, who may not be able to swallow pills, may require different formulations. Therefore, pediatric formulation development should begin as early as possible to enable the development of appropriate pediatric formulations of investigational drugs.

6. Dose Selection

The results from proof-of-concept antiviral activity trials can be used to guide the selection of doses to be evaluated in subsequent trials. Sponsors are encouraged to use various quantitative clinical pharmacology approaches that leverage the totality of available information on exposure and pharmacodynamic assessments such as biomarkers that measure target engagement, HBsAg decline, HBV DNA decline, HBV RNA decline, and HBcrAg decline to select doses for phase 3 trials.

7. Efficacy Endpoints

New finite duration therapies could be evaluated in clinical trials using any of the following efficacy endpoints:

- Sustained suppression (6 months or longer) of HBV DNA (less than LLOQ, TD, or TND) off-treatment after a finite duration of therapy

- Sustained suppression (6 months or longer) of HBV DNA (less than LLOQ, TD, or TND) off-treatment with HBsAg loss (less than 0.05 international unit/milliliter (IU/mL)) with or without HBsAb seroconversion after a finite duration of therapy

At present, utility of reduction in HBsAg from baseline (without complete clearance) for assessing response to CHB therapies is unclear because of inconsistent correlations between qHBsAg and clinical response (Hu et al. 2018; Thompson et al. 2010; Chan et al. 2011).

[22] See section 505B(a)(2)A) of the FD&C Act.

A limited number of secondary endpoints (e.g., HBeAg loss, anti-HBe seroconversion in HBeAg positive patients, ALT normalization) should be considered for testing using appropriate statistical methods for multiplicity. Biochemical serum markers such as ALT values vary between laboratories, and lack of normalization of ALT may often be confounded by the presence of other chronic liver diseases such as nonalcoholic fatty liver disease.

Other important endpoints: Assessing progression of liver disease

Except for patients with advanced or decompensated cirrhosis, a statistically rigorous evaluation of endpoints of liver progression can be challenging because these events occur infrequently until late in the course of CHB. However, treatment effects on these endpoints provide useful clinical information, and trials evaluating them could be used to support an expanded indication or patient population and could be summarized in appropriate sections of the label.

Some of the parameters or clinical outcomes that sponsors can consider include the following:

- Change in Model for End Stage Liver Disease scores
- Change in Child-Turcotte-Pugh scores
- Progression to liver failure requiring transplantation or resulting in death
- Occurrence of HCC

Treatment-related regression of fibrosis or cirrhosis, as assessed by liver biopsy or noninvasive methods, may be appropriate for display in the label. However, sponsors should discuss with the Division plans for labeling and performance characteristics of the noninvasive modality when protocols evaluating these endpoints are being designed.

Patient-Reported Outcome Measures

Sponsors who are considering incorporating patient-reported outcome measures[23] as endpoints in clinical trials to measure treatment benefit should discuss patient-reported outcome instruments with the Agency during the clinical development process.

8. Trial Procedures and Timing of Assessments

Biochemical, serological, virological, and histological endpoints can be used to assess the effectiveness of therapy. For drugs with finite treatment durations, the optimal time point to assess the primary efficacy endpoint of sustained virologic response is 6 months or longer after cessation of therapy. Additionally, the most appropriate time point to assess efficacy endpoints depends on the mechanism of action and half-life of the drug. Longer term follow-up may be useful to confirm durability of treatment response and to measure clinical outcomes.

9. Statistical Considerations

In general, a detailed protocol and SAP stating the trial hypotheses, analysis methods, and all other relevant details should be provided to the Division of Antivirals before trial initiation. To improve the precision of treatment effect estimation and inference, sponsors should consider adjusting for prespecified baseline factors that are anticipated to be prognostic of the outcome (e.g., HBeAg status, HBV DNA level, and HBsAg level). If randomization is stratified by baseline covariates, the analysis should account for the stratification factors. A discussion of how the analyses will account for the stratified randomization should be included in the protocol. For details see the draft guidance for industry *Adjusting for Covariates in Randomized Clinical Trials for Drugs and Biological Products* (May 2021).[24] For statistical analysis methods and issues, see the guidances for industry *Providing Clinical Evidence of Effectiveness for Human Drug and Biological Products* (May 1998), *Non-Inferiority Clinical Trials to Establish Effectiveness* (November 2016), and *Adaptive Designs for Clinical Trials of Drugs and Biologics* and the FDA white paper Statistical Considerations on Subgroup Analysis in Clinical Trials (Alosh et al. 2015).

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[23] See the guidance for industry *Patient-Reported Outcome Measures: Use in Medical Product Development to Support Labeling Claims* (December 2009).

[24] When final, this guidance will represent the FDA's current thinking on this topic.

a. Analysis populations

All subjects who are randomized and received at least one dose of assigned therapy during the trial should be included in the primary efficacy analysis for all fully blinded trials. For unblinded/partially blinded trials, all randomized subjects should be considered. Any possibility of randomized subjects who do not receive treatment in either or both arms should be minimized.

b. Efficacy analyses

The primary analysis should compare the proportion of responders across trial treatment arms. This analysis determines whether effectiveness has been demonstrated.

For subgroup analyses, the analysis of the primary efficacy endpoint should be performed within important demographic and baseline characteristics (e.g., geographic region, sex, race, age group, HBV genotype, HBeAg status, screening HBV DNA, baseline weight, body mass index, baseline ALT, baseline fibrosis/cirrhosis, and (if applicable) prior response to previous treatment regimens). The purpose of these analyses is to explore the consistency of the primary efficacy endpoint result across these subgroups.

Treatment-by-region interaction should be investigated and reported to assess consistency of the efficacy results. Treatment-by-HBeAg status interaction should also be investigated if HBeAg-positive and HBeAg-negative subjects are enrolled in the trial.

c. Handling missing data

Sponsors should make every attempt to limit discontinuation of subjects from the trial. When the loss is unavoidable, sponsors should explain the causes of missing data and attempt to determine the final status of a subject who does not complete the protocol. Analyses excluding subjects with missing data or other posttreatment outcomes can be biased because subjects who do not complete the trial may differ substantially in both measured and unmeasured ways compared with subjects who remain in the trial. The primary method of handling missing data in the analysis should be prespecified in the protocol and SAP. Sensitivity analyses should demonstrate that the primary analysis results are robust to the assumptions regarding missing data.

10. Accelerated Approval (Subpart H/I) Considerations

For CHB, HBV DNA suppression with or without HBsAg loss is considered a validated surrogate endpoint that has been demonstrated to predict clinical outcomes, and this endpoint could be used to support a traditional approval. Sponsors should discuss with the FDA plans to use any other surrogate endpoints that are reasonably likely to predict clinical benefit to support accelerated approval.[25] Accelerated

[25] See section 506(c) of the FD&C Act; 21 CFR part 314, subpart H; 21 CFR part 601, subpart E.

approval is subject to the requirement that postmarketing confirmatory trials are conducted to verify and describe the anticipated effect on irreversible morbidity or mortality or other clinical benefit.[26]

11. Benefit-Risk Considerations

A thorough and comprehensive benefit-risk assessment ensures that the benefits outweigh potential risks to the intended population. A benefit-risk assessment takes into consideration the demonstrated therapeutic effect of the new drug and observed safety profile in the context of underlying disease and current treatment options available for the indication.

C. Other Considerations

1. Clinical Virology Considerations

Samples for HBV quantification, genotypic, and phenotypic analysis should be obtained at different time points during treatment and follow-up. Timing of sample collection should be based on initial observations of potency and on-treatment and off-treatment durability. The genotypes and phenotypes of baseline and virologic failure isolates should be determined (virologic failure defined as a confirmed increase of greater than or equal to 1 $\log_{10}$ HBV DNA copies/mL above nadir, quantifiable HBV DNA of greater than or equal to 1 $\log_{10}$ copies/mL above LLOQ after being less than LLOQ, or never achieved HBV DNA levels less than LLOQ for two consecutive visits). Transient increases in HBV DNA after the completion of treatment with a finite regimen may occur before sustained suppression. A strategy for distinguishing treatment failures from subjects experiencing a temporary increase should be agreed upon with the Division during protocol development.

Clinical assessment during the development of activators of the immune system is likely to be challenging, given that the greatest impact of these types of drugs will likely be a reduction in infected cells resulting in depletion of the cccDNA reservoir. Complete depletion of the cccDNA reservoir to below the limit of detection may take a long time and will vary depending on the mechanism of action of the drug. Currently, the only biomarker sufficient to predict a sustained response off-treatment is HBsAg loss, the assessment of which may be complicated by HBsAg expressed from integrated HBV DNA genome fragments. In addition to host immune markers and serum HBV DNA, clinical trial protocols should assess several exploratory HBV endpoints (HBcrAg, HBV RNA, etc.) early in the development program in an attempt to identify potential markers that correlate with the drug's activity and may predict response to therapy.

[26] See 21 CFR 314.510; 21 CFR 601.41.

Genotypes of baseline and on-therapy virologic failure isolates should be compared and newly emerged drug resistance-associated substitutions/mutations should be identified. HBV DNA from subjects with genotypic resistance to the investigational drug should be cloned in an HBV genome background and susceptibility to the investigational drug should be determined.

- There are 10 recognized HBV genotypes (genotypes A through J) as well as subtypes identified for genotypes A through F. The different HBV genotypes/subtypes encode distinct viral proteins and may exhibit differential responses to an investigational drug, which could confound efficacy results in clinical trials if the drug is only effective against some genotypes/subtypes (Congly et al. 2013). Therefore, we recommend determining the genotypes/subtypes of HBV infection present at baseline to determine if the investigational drug exhibits antiviral activity against all HBV genotypes/subtypes. For subjects with suppressed HBV DNA at baseline, historical genotype assessments may be used if available. The assay, with performance characteristics, used to genotype the HBV samples in enrolled subjects should be included with the clinical trial protocol. It may be important to confirm the genotype/subtype by phylogenetic analysis.

- For resistance analyses, any changes, including mixtures, in the amino acid sequence of the target protein or DNA sequence for genome targeting drugs, present in on-treatment or follow-up samples but not in the baseline sample, can be reported as having developed during therapy. In addition, baseline samples should be analyzed to identify HBV genetic polymorphisms that are associated with differential antiviral activity against the investigational drug. Sponsors should consult the FDA early for the most current format for submission of resistance data and if Next Generation Sequencing (NGS) will be used.

- There is a risk of developing resistance against an antiviral drug that targets similar viral proteins in different virus species in patients coinfected with HIV and HBV. Because of this risk, we recommend assessing for the development of resistance and cross-resistance in the viral proteins of both HIV-1 and HBV when appropriate.

- For all virologic assessments in clinical trials, we recommend the use of FDA-approved or FDA-cleared assays, when available, and a central laboratory. Sponsors can collect results from local lab tests, identifying the assays used. If investigational assays are used, performance characteristics of the assays determined from analytical validation studies using geographically and temporally distinct isolates should be provided in addition to detailed descriptions of the methodology.[27] Drugs that require assays to identify the infected population benefiting from

[27] See the IDE (Investigational Device Exemption) web page, available at https://www.fda.gov/MedicalDevices/DeviceRegulationandGuidance/HowtoMarketYourDevice/InvestigationalDeviceExemptionIDE/ucm046164.htm.

treatment (e.g., specific genotypes or resistant populations) may require a companion diagnostic. Additional recommendations can be found in the draft guidance for industry and Food and Drug Administration staff *Principles for Codevelopment of an In Vitro Companion Diagnostic Device with a Therapeutic Product* (July 2016).[28]

- Sponsors are encouraged to submit a resistance monitoring plan early in development. If resistance evaluation in clinical trials involves NGS, we recommend that sponsors discuss details of the NGS approach with the FDA. Submission of NGS data in FASTQ format is strongly encouraged. Additional recommendations can be found in the guidance for industry technical specifications document *Submitting Next Generation Sequencing Data to the Division of Antiviral Products* (July 2019).

- HBV should be genotyped for any instances where HBV DNA is detected in long-term follow-up to distinguish relapse from reinfection.

[28] When final, this guidance will represent the FDA's current thinking on this topic.

2. Pharmacokinetic/Pharmacodynamic Considerations

Trials conducted in HBV-infected subjects should include assessment of PK and the relationship between drug exposure (e.g., minimum or maximum plasma concentration (C_{min} or C_{max}), area under the curve) and virologic success and toxicity in all subjects.

Sponsors can use a combination of intensive and sparse sampling throughout development to characterize the PK of the investigational drug. For example, sponsors should implement an intensive sampling schedule in early phase monotherapy trials. In longer term trials, an intensive sampling schedule might not be feasible. Alternatively, sponsors can combine sparse sampling from these trials with intensive PK data from earlier trials for population PK analysis. Sponsors should obtain multiple sparse PK samples from as many subjects as possible, including at the time of key virologic assessments. It is important to document dosing times and plasma sampling times.

Sponsors can use the following two broad approaches to characterize the relationship between drug exposure and viral kinetics or virologic suppression of the investigational drug, depending on the development stage and purpose of the analysis. Both approaches allow for exploration of relevant covariates.

(1) To aid the design of phase 2b or phase 3 trials, with respect to selection of the dosage regimen, a mechanistic approach relating drug concentrations and viral kinetics should be considered. A mechanistic modeling approach should also account for the development of resistance to the investigational drug and the intended patient population. For combination therapy, the potential of additive or synergistic antiviral effects can be incorporated in the model to assist optimization of the dose combination.

(2) A simplified analysis relating the proportion of subjects with virologic suppression or virologic failure and appropriate exposure variable (e.g., minimum concentration or area under the plasma drug concentration versus time curve) can be used to support evidence of activity and to support dose selection.

Exposure-response safety analyses should consider the mechanistic on-target and off-target effects of the investigational drug and adverse events that are more frequent in the investigational drug arm. The appropriate exposure parameter and modeling approach depends on the investigational drug and toxicity.

3. Labeling Considerations

Severe acute exacerbations of CHB may occur after discontinuation of anti-HBV therapy. Hepatic function should be monitored closely with both clinical and laboratory follow-up for at least several months in subjects who discontinue anti-HBV therapy. In certain circumstances, resumption of anti-HBV therapy may be warranted. These concerns should be adequately conveyed in drug labeling.

Development of HIV-1 resistance against anti-HBV drugs with activity against HIV-1 is a potential risk that should be conveyed in labeling.

GLOSSARY OF ACRONYMS

ALT	alanine aminotransferase
CC_{50}	concentration inhibiting 50 percent cell growth
cccDNA	covalently closed circular DNA
CHB	chronic hepatitis B
$EC_{50/90}$	effective drug concentration inhibiting 50 or 90 percent virus replication
FD&C Act	Federal Food, Drug, and Cosmetic Act
HBeAg	HBV e-antigen
HBsAb	antibody specific to HBsAg
HBsAg	HBV surface antigen
HBV	hepatitis B virus
HBV DNA	hepatitis B virus DNA
HCC	hepatocellular carcinoma
HDV	hepatitis delta virus
HIV	human immunodeficiency virus
IFN	interferon
IgM	immunoglobulin M
IU	international unit
LLOQ	lower limit of quantification
mL	milliliter
NRTI	HIV nucleoside/nucleotide reverse transcriptase inhibitor
NrtI	HBV nucleoside/nucleotide reverse transcriptase inhibitor
NGS	Next Generation Sequencing
NI	noninferiority
PK	pharmacokinetic
qHBsAg	quantitative HBsAg
RNA	ribonucleic acid
rt	reverse transcriptase
SAP	statistical analysis plan
TD	target detected
TND	target not detected

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