Annual Review of Medicine

New Therapeutics for Hepatitis B: The Road to Cure

Susanna Naggie[1,2] and Anna S. Lok[3]

[1]Department of Medicine, Division of Infectious Diseases, Duke University School of Medicine, Durham, North Carolina 27701, USA; email: susanna.naggie@duke.edu

[2]Duke Clinical Research Institute, Durham, North Carolina 27701, USA

[3]Department of Internal Medicine—Gastroenterology and Hepatology, University of Michigan School of Medicine, Ann Arbor, Michigan 48109, USA; email: aslok@umich.edu

Annu. Rev. Med. 2021. 72:93–105

First published as a Review in Advance on October 21, 2020

The *Annual Review of Medicine* is online at med.annualreviews.org

https://doi.org/10.1146/annurev-med-080119-103356

Keywords

functional cure, chronic viral hepatitis, HBV surface antigen, curative therapy, targeted antivirals

Abstract

Chronic hepatitis B virus (HBV) infection impacts an estimated 257–291 million people globally. The current approach to treatment for chronic HBV infection is complex, reflecting a risk:benefit approach driven by the lack of an effective curative regimen. This complexity and the lack of a durable treatment response, necessitating indefinite treatment in the majority of cases, have resulted in low uptake of testing and treatment, particularly in regions where comprehensive primary care is lacking and access to affordable testing and treatment is limited. Multiple targeted therapies are now in early human study with the primary goal to achieve persistent HBV DNA and hepatitis B surface antigen suppression after a finite course of treatment, which is referred to as functional cure. This article summarizes the current therapies for HBV infection and discusses the limitations of these therapies, novel approaches to HBV cure, and therapeutic endpoints of clinical trials aimed to cure hepatitis B.

INTRODUCTION

Chronic hepatitis B virus (HBV) infection impacts an estimated 257–291 million people globally (1–3). The extensive burden of infection results in approximately 800,000 deaths annually, driven by complications of cirrhosis and hepatocellular carcinoma (HCC). Globally, the number of deaths due to viral hepatitis is increasing; a rise of >60% by 2040 is forecast (4). Although we have effective vaccines to prevent HBV infection and safe and effective antivirals to treat chronic HBV infection, incident infection, particularly in early childhood, remains high, and access to birth-dose HBV vaccination remains low (1). To impact the rising mortality related to HBV infection globally, testing and treatment are critical; yet, only 9% of persons with the infection were diagnosed in 2015, and <10% of those meeting criteria for therapy were receiving treatment (1).

The current approach to treatment for chronic HBV infection is complex, reflecting a risk:benefit approach driven by the lack of an effective curative regimen. Decision making on initiating therapy requires an assessment of the phases of the disease, determined by level of HBV replication and activity and stage of liver disease (5). This complexity and the lack of a durable treatment response, necessitating indefinite treatment in the majority of cases, have resulted in low uptake of testing and treatment, particularly in regions where comprehensive primary care is lacking and access to affordable testing and treatment is limited.

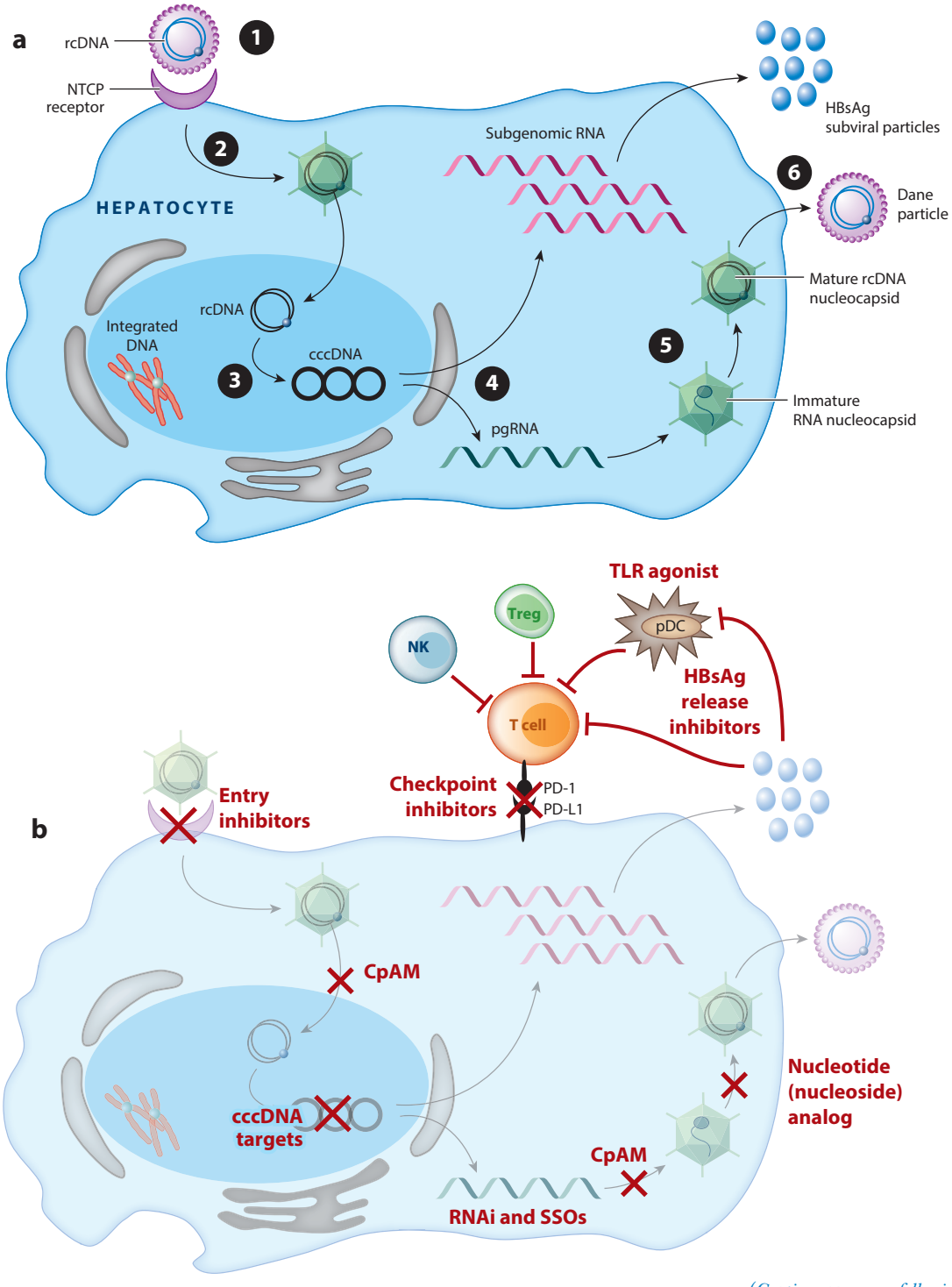
As we have seen with the development of curative, finite therapies for chronic hepatitis C virus (HCV) infection, availability of simple, safe, highly effective curative therapies can change the calculus of medical decision making and cost-effectiveness ratio with regard to widespread testing and treatment of all infected persons. The simplicity of HCV treatment, with similar safety and efficacy across HCV genotypes with and without HIV coinfection, even in patients with end-stage liver or kidney disease, has enabled treatment in care settings and in areas with limited resources (6, 7). The success of therapeutics for HCV infection has renewed hope that cure of HBV infection may also be possible (8). This article summarizes the current therapies for HBV infection and discusses the limitations of these therapies, novel approaches to HBV cure, and therapeutic endpoints of clinical trials aimed to cure hepatitis B.

TARGETS AND MECHANISMS OF HBV THERAPEUTICS

HBV is a partially double-stranded hepatotropic DNA virus that belongs to the Hepadnaviridae family (9). The viral envelope surrounds an icosahedral nucleocapsid, which holds a partially double-stranded, relaxed circular DNA (rcDNA) genome. HBV infects and replicates in hepatocytes (**Figure 1**). The virus binds to the cell surface via the pre-S glycoprotein and interacts with the primary receptor, the hepatic bile acid transporter sodium taurocholate cotransporting polypeptide (NTCP) (10, 11). Following entry, the rcDNA genome is transported into the nucleus and converted into covalently closed circular DNA (cccDNA), which associates with histone and nonhistone proteins to form the viral minichromosome (12). The cccDNA is transcribed into the pregenomic RNA (pgRNA), which serves as the template for reverse transcription to HBV DNA in the hepatocyte cytoplasm and the translational template for core protein and polymerase. The nucleocapsid with partially double-stranded HBV DNA is enveloped and the virion is secreted. However, nucleocapsids can also recycle back into the nucleus and the rcDNA can be converted to cccDNA, so the cccDNA reservoir can be maintained without the need for entry of new HBV.

CURRENT THERAPEUTIC STRATEGIES

There are currently seven antiviral therapies approved by the US Food and Drug Administration for the treatment of chronic HBV infection. Each has one of two therapeutic mechanisms



(*Caption appears on following page*)

Figure 1 (*Figure appears on preceding page*)

HBV life cycle and main targets of direct and indirect viral therapies. (*a*) The HBV life cycle. The virus binds to the cell surface via the pre-S glycoprotein and interacts with the primary receptor, the hepatic bile acid transporter NTCP (10, 11) (❶). Following entry, the nucleocapsid is released into the cytoplasm (❷), and the rcDNA genome is transported into the nucleus and converted into cccDNA (❸), which associates with histone and nonhistone proteins to form the viral minichromosome (12). The cccDNA is transcribed into the pgRNA (❹), which serves as the template for reverse transcription to HBV DNA in the hepatocyte cytoplasm and the translational template for core protein and polymerase (❺). The nucleocapsid with partially double-stranded HBV DNA is enveloped and the virion is secreted (❻). (*b*) The main targets of direct and indirect viral therapies. Direct antiviral therapies include entry inhibitors, HBsAg release inhibitors, nucleotide (nucleoside) analogs, CpAM, interference RNAs and SSOs, and cccDNA targets. Indirect therapies that target the immune system include checkpoint inhibitors and TLR agonists. Abbreviations: cccDNA, covalently closed circular DNA; CpAM, core protein allosteric modulator; HBV, hepatitis B virus; HBsAg, hepatitis B surface antigen; NK, natural killer; NTCP, sodium taurocholate cotransporting polypeptide; pDC, plasmacytoid dendritic cell; pgRNA, pregenomic RNA; rcDNA, relaxed circular DNA; SSO, single-stranded oligonucleotide; TLR, Toll-like receptor; Treg, regulatory T cell.

of action: interferons or nucleoside/nucleotide analogs (NAs). The therapeutic goals of the current antiviral therapies are primarily virologic and biochemical (**Table 1**). A virologic response to therapy is defined by a reduction in HBV DNA level in the blood, with a goal of achieving a level below the limit of assay detection (10–20 IU/mL with current real-time PCR assays). A biochemical response to therapy is defined by a reduction in the level of alanine aminotransferase in the blood to normal. Virologic and biochemical responses are associated with improved clinical outcomes (13–16). Both interferons and NAs have been associated with decreased liver inflammation and fibrosis, as well as reduced rates of cirrhosis and HCC (17, 18).

However, a limitation of our current therapies is the low rate of serologic responses—clearance of hepatitis B early antigen (HBeAg) and hepatitis B surface antigen (HBsAg). A unique feature of HBV is the production of large quantities of subviral particles of HBsAg, outnumbering complete virions by >1000:1. High levels of circulating HBsAg have been suggested to contribute to persistent infection through immune exhaustion (19). Clearance of HBsAg is usually accompanied by the development of HBV surface antibody (anti-HBs) and is associated with a durable suppression of HBV DNA, without the need for ongoing therapy. HBsAg clearance is also associated with improved prognosis, including further decreased risk of HCC compared to HBV DNA suppression without loss of HBsAg (20–22). HBsAg loss is uncommon with our current therapies, particularly NA therapies, resulting in the need for indefinite therapy for the majority of patients. Although interferon therapies have higher rates of HBsAg clearance (~10%), this is true mainly for HBV genotype A, and many patients are unable to tolerate the side effects of interferon. Given the need for frequent clinical and laboratory monitoring, interferon therapy can only be administered in specialized centers.

Table 1 Therapeutic goals and definitions for current and novel HBV therapies

Therapeutic response	Blood				Liver
	ALT	HBV DNA	HBsAg	Anti-HB	cccDNA
Virologic	N/A	Undetectable	Detected	Undetectable	Present
Biochemical	Normal	N/A	Detected	Undetectable	Present
Function cure	Normal	Undetectable	Undetectable	Detected	Present
Complete/sterilizing cure	Normal	Undetectable	Undetectable	Detected	Undetectable

Abbreviations: ALT, alanine aminotransferase; anti-HB, HBV surface antibody; cccDNA, covalently closed circular DNA; HBsAg, hepatitis B surface antigen; HBV, hepatitis B virus.

THERAPEUTIC ENDPOINTS FOR CURE

The concept of cure for a virus that has host genome integration and/or a latency phase is complex. To date, we do not have cures of integrated viruses (e.g., herpesviruses), and in the case of chronic HBV infection, the definition of cure is controversial, reflecting a chasm between ideal and achievable goals. Current HBV therapies suppress HBV replication but do not eliminate the virus. Residual virus in the liver, both replication competent in the form of cccDNA and replication incompetent in the form of integrated HBV DNA, contributes to persistent albeit lower risk of liver-related complications, particularly HCC. Ideally, a cure for chronic HBV would translate to finite therapies that result in loss of HBsAg and eradication of HBV, but these goals may not be realistic. Thus, several definitions of cure have been previously proposed (**Table 1**).

Recently, the European Association for the Study of the Liver (EASL) and the American Association for the Study of Liver Diseases (AASLD) jointly organized an HBV Treatment Endpoint Conference to develop a consensus on the nomenclature for "achievable" cure and to agree on HBV treatment endpoints to guide the design of clinical trials (23).

Functional cure, deemed an achievable goal, is defined as durable HBsAg loss [based on assays with lower limit of detection (LLOD) ~0.05 IU/mL], with or without anti-HBs seroconversion, and undetectable serum HBV DNA after completing a course of treatment (23) (**Table 1**). Functional cure should be differentiated from what has been defined as complete or sterilizing cure, which includes not only durable HBsAg loss and undetectable serum HBV DNA, but also elimination of cccDNA, which serves as a source of viral latency and reactivation in the appropriate setting (e.g., with rituximab treatment). In the setting of functional cure, integrated HBV DNA is still present, which can produce HBsAg, but is not sufficient for productive replication, and cccDNA may still be present but is not transcriptionally active (24). While functional cure may be viewed as less ambitious, it has been shown to be associated with improved clinical outcomes and, from the patients' perspective, removes the stigma of HBV infection.

The development of surrogate biomarkers for functional cure is critical for clinical trial design of novel HBV therapeutics. With current therapies, the primary virologic biomarker is HBV DNA, with the goal of viral suppression below the assay limit of detection. However, for the majority of patients, withdrawal of therapy results in rebound of viremia. Thus, novel biomarkers that reliably represent the intrahepatic reservoir of transcriptionally active cccDNA and risk of relapse after treatment withdrawal are needed.

HBsAg as a Surrogate for HBV Functional Cure

Generally, HBsAg loss is durable and accompanied by undetectable HBV DNA, and viral relapse is rare when treatment is stopped. Ideally, the goal is to achieve not only HBsAg loss but also anti-HBs seroconversion, a sign of immunity, although numerous studies have shown that anti-HBs development is not essential for durable HBsAg loss. This is true for spontaneous as well as treatment-induced HBsAg loss (25).

Circulating HBsAg can be measured qualitatively and quantitatively. Quantitative HBsAg decline precedes HBsAg loss and has been used to predict loss of HBsAg in interferon-based trials and to predict virologic relapse following NA withdrawal (26). The decline in HBsAg is attributed to a decrease in transcriptionally active cccDNA. However, this may not be the case with HBV therapies that have different mechanisms of action [e.g., small interfering RNAs (siRNAs) targeting production of HBsAg]. While a marked decline (e.g., 2–3 log) in HBsAg level or a decline in HBsAg to low levels (e.g., 10 or 100 IU/mL) is associated with a higher chance of HBsAg loss, specific thresholds of quantitative HBsAg that reliably predict HBsAg loss have not

been determined and might vary depending on the mechanism of action of the HBV therapeutic (26). Thus, decline in quantitative HBsAg in phase II trials may not predict HBsAg loss in phase III trials. Furthermore, current assays do not differentiate between HBsAg derived from integrated HBV DNA and HBsAg derived from cccDNA. Thus, it has been argued that cccDNA may be eliminated or rendered transcriptionally inactive, yet low levels of HBsAg derived from integrated HBV DNA may still be detected. In an early study of siRNA targeting cccDNA transcription, HBsAg from integrated HBV DNA was detected in eAg-negative participants, highlighting the need for dual transcription targets.

Measures of cccDNA and HBV Transcription

Persistence of cccDNA is the Achilles heel of HBV cure. Direct measures of cccDNA require liver tissue, as well as stringent specificity to differentiate cccDNA from rcDNA. Several serum markers have been proposed to be surrogates of cccDNA, including quantitative HBsAg, though integrated HBV DNA can also be the source of HBsAg, HBV RNA, and hepatitis B core-related antigen (HBcrAg).

Serum HBV RNA

Serum HBV RNA exists predominantly as intact, pregenomic, and subgenomic species and may serve as a reliable biomarker of intrahepatic cccDNA concentration and transcriptional activity (27, 28). During replication, the cccDNA is transcribed into several RNA species, including pgRNA. PgRNA is packaged in nucleocapsids and serves as the template for reverse transcription to HBV DNA. In patients not receiving antiviral therapy, serum HBV RNA levels are much lower than serum HBV DNA levels. During NA therapy, reverse transcription of pgRNA to HBV DNA is inhibited, but cccDNA transcription to pgRNA continues, and HBV RNA may be detected in serum of patients with undetectable HBV DNA. The presence of serum HBV RNA, particularly pgRNA, is a useful biomarker of cccDNA activity, and in patients with longstanding HBV DNA suppression on NA therapy, elevated pgRNA has been shown to predict viral relapse when NA is stopped (29). There is currently no approved assay for HBV RNA; furthermore, most HBV RNA assays are not specific for pgRNA and may include spliced RNA and mRNA.

Serum HBcrAg

HBcrAg is a biomarker for hepatitis B core antigen. The HBcrAg assay measures denatured HBeAg, HBcAg, and the precore protein p22cr (30). Serum HBcrAg levels have been correlated with serum HBV DNA, intrahepatic pgRNA, and cccDNA levels and transcriptional activity (31, 32). There is no approved assay for HBcrAg. Current research assays lack sensitivity, and the results are less informative in HBeAg-positive patients, as the assay also measures HBeAg.

NOVEL APPROACHES TO HBV CURE

Targeted Antivirals

There are multiple novel antivirals targeting different steps in the HBV life cycle currently in development, reminiscent of the early days of HCV direct-acting antiviral discovery (**Table 2**). Several of these novel agents are now in phase II trials. Here, we focus only on those that have been studied in humans.

Table 2 Summary of novel HBV therapeutics in human studies

Drug name	Mechanism	Delivery	Trial phase
Entry inhibitor			
Myrcludex B	Competition with NTCP	Subcutaneous injection	II
Targeting HBx			
Nitazoxanide	Inhibits HBV transcription from cccDNA	Oral	II
CRV-431	Cyclophilin inhibitor	Oral	I
Inhibition of gene expression/gene silencing			
Antisense oligonucleotide			
GSK3228836	RNA degradation	Subcutaneous injection	II
RNA interference (RNAi)			
JNJ-3989 (formerly ARO-HBV-1001)	RNA degradation	Subcutaneous injection	II
AB-729	RNAi	Subcutaneous injection	I
ALN-HBV (VIR-2218)	RNA degradation	Subcutaneous injection	I
Core protein allosteric modulators (CpAMs)			
ABI-H0731	Core protein binding	Oral	II
RO7049389	Core protein binding	Oral	II
JNJ-56136379	Assembly modulator	Oral	II
AB-506	Core protein binding	Oral	I
ABI-H2158	Core protein binding	Oral	I
GLS4JHS	Core protein binding	Oral	II
HBsAg release inhibitors			
Nucleic acid polymers (REP 2139 and 2165)	HBsAg binding	Intravenous infusion	II
Targeting cell intrinsic and innate immune responses			
RO7020531	TLR7 agonist	Oral	I
Vesatolimod, GS-9620	TLR7 agonist	Oral	II
Selgantolimod, GS-9688	TLR8 agonist	Oral	I
AIC649	TLR9 agonist	Oral	I
Targeting adaptive immune responses			
Checkpoint inhibitors			
Nivolumab	Anti-PD1	Intravenous infusion	I
Cemiplimab, REGN2810	Anti-PD1	Intravenous infusion	I/II
Therapeutic vaccines			
TG1050/T101	Non-replicative adenovirus serotype 5 encoding three HBV proteins	Subcutaneous injection	I
ChAdOx1 HBV	Adjuvanted ChAd and MVA vectored	Intramuscular injection	I
HepTcell	HBV peptide therapeutic vaccine with TLR9 adjuvant IC31	Intramuscular injection	I
JNJ-64300535	Electroporation of DNA vaccine	Electroporation-mediated intramuscular injection	I
INO-1800	DNA plasmids encoding HBsAg and HBcAg plus INO-9112 (DNA plasmid encoding human interleukin 12)	Electroporation-mediated intramuscular injection	I

HBV entry inhibitors. Identification of the entry receptor for HBV has allowed the development of entry inhibitors. As a class, these compounds prevent both HBV and hepatitis D virus (HDV) entry and stop infection of naïve hepatocytes and thus onward infection. Several of these agents block NTCP receptor fusion, which can also block bile salt transport and thus increase serum bile acids. Myrcludex B (bulevirtide) and cyclosporine A both bind NTCP and have exhibited antiviral activity.

Myrcludex B is a first-in-class entry inhibitor in phase IIa study for chronic HBV infection. It has been investigated as monotherapy and in combination with pegylated interferon (peg-IFN) as well as tenofovir disoproxil fumarate (TDF). To date, most studies of myrcludex B have focused on chronic HDV infection, an orphan disease for which there is no approved treatment. With monotherapy, there was only a decrease in HDV RNA, but when combined with peg-IFN, after 48 weeks, 26% of HBV/HDV coinfected patients had HBsAg loss (33).

Nucleocapsid assembly modulators. HBV core protein is essential for HBV genome packaging (pgRNA) and reverse transcription. Depending on the chemical structure of the core protein allosteric modulators (CpAMs), two types of dysfunctional capsids may be formed: unstable aberrant capsids or empty capsids. Theoretically, the dysfunctional capsids would reduce the production of infectious virions and decrease the replenishment of cccDNA in the hepatocyte nucleus. There are currently 10 CpAMs in phase I or II study. Monotherapy with CpAMs reduces serum HBV DNA and HBV RNA levels, but there was no change in HBsAg levels after up to 12 weeks of therapy. It is expected that CpAMs will be used in combination with other novel HBV agents, particularly those targeting HBsAg, to achieve functional cure.

In the phase Ib study, JNJ-6379 was dosed once daily orally for 28 days in patients with chronic HBV infection and without cirrhosis. Rapid HBV DNA suppression was noted, but there was no impact on HBeAg or HBsAg levels (34). However, when studied as triple therapy in combination with siRNA JNJ-3989 and NA, JNJ-6379 showed a >1.5-log decrease in HBsAg levels after 12 weeks of treatment (35). In a phase Ib/IIa study, the CpAM ABI-H0731 was dosed as monotherapy once daily orally for 28 days in patients with chronic HBV infection and without cirrhosis. Rapid HBV DNA and RNA suppression was noted, but there was no impact on HBeAg or HBsAg levels (35, 36). ABI-H0732, when used in combination with NA therapy, results in more rapid decline in HBV DNA and pgRNA levels, compared to NA alone (37).

Post-transcriptional control inhibitors (RNAi or oligonucleotides). RNA interference (RNAi) can directly target HBV transcripts and induce their degradation, resulting in gene silencing. Single-stranded oligonucleotides (SSOs) are small nucleic acids complementary to the target transcript that similarly induce degradation after binding. Several studies of RNAi and SSO are in progress.

In a phase II study, ARC-520 (RNAi) was investigated in combination with entecavir in HBeAg– and HBeAg+ chronic HBV infection and resulted in a rapid and potent decrease in serum HBV DNA (38). However, the decrease in HBsAg levels was only noted in the HBeAg+ patients. Follow-up studies in chimpanzees revealed that the HBsAg being produced in the HBeAg– patients was predominantly derived from integrated virus, which was not targeted by the RNAi (38). This highlights the importance of having a combination of RNAi targeting different regions of the HBV genome. A modified RNAi, ARO-HBV (JNJ-3989), contains two RNAi that are both conjugated to N-acetyl galactosamine to facilitate uptake by the liver. A phase Ib study of subcutaneous delivery of ARO-HBV showed rapid decline in HBV DNA and a robust decline in HBsAg in both HBeAg– and HBeAg+ patients (39). As described above, ARO-HBV has been evaluated as triple therapy in combination with CpAM and NA.

HBsAg release inhibitors. Secretion inhibitors function under the same principle as the RNAi and oligonucleotides, which is to block the release of subviral HBsAg particles that are thought to have immunoinhibitory properties and block innate immune response. REP 2139 is a nucleic acid polymer that blocks the release of the subviral particles (40). Preclinical studies suggested that the selective inhibition of subviral particle release does not increase intracellular HBsAg but is associated with clearance of HBsAg and HBV DNA from the blood and reduced cccDNA from the liver. A phase II, open-label, randomized study assessed the safety and efficacy of addition of either REP 2139–Mg or REP 2165–Mg to a backbone of TDF and peg-IFN (41). In Part A, 40 participants with HBeAg– chronic HBV infection and HBsAg concentrations >1,000 IU/mL were enrolled. All participants received 24 weeks of TDF before the introduction of peg-IFN. Participants were then randomized to either the control arm ($n = 20$) with peg-IFN + TDF or the experimental arm with the addition of either REP 2139–Mg ($n = 10$) or REP 2165–Mg ($n = 10$) to the backbone of peg-IFN + TDF. At week 48, 10 participants in the experimental arm experienced HBsAg loss, and 11 had anti-HBs seroconversion compared to none in the control arm. However, HBsAg seroconversion persisted in only 50% during post-treatment follow-up.

cccDNA targets. The development of small molecules that can inhibit cccDNA formation or destabilize the currently established cccDNA is being explored, but none have been tested in clinical trials yet.

Immunomodulatory Targets Including Therapeutic Vaccines

Impaired immune response in patients with chronic HBV infection creates opportunities for immunomodulatory mechanistic targets that may serve as key components of a combined cure strategy. T cell immune exhaustion is well described in HBV persistence (31). The abundance of subviral HBsAg is likely part of a viral immune evasive strategy that results in T cell exhaustion. In addition, deficient activation of the innate immune response and the development of a tolerogenic intrahepatic environment also play a role in HBV persistence (19). Thus, treatment strategies to restore HBV-specific immune responses with immunomodulatory therapies may be critical to cure strategies that achieve durable viral clearance.

Interferons. Interferons are antiviral proteins that play an important role in the innate immune response to viral infections. Interferons bind to receptors on the cell surface and stimulate IFN-stimulated response elements in the nucleus, which have multiple downstream effects including virus recognition and clearance (42). Historically, peg-IFN has been used for the treatment of chronic HBV and HCV infection, although in both cases, rates of achieving prolonged viral suppression or viral clearance are low (43, 44). Peg-IFN has been shown to inhibit pgRNA encapsidation, enhance cccDNA degradation, and exert epigenetic modification of cccDNA transcription (45). Multiple investigational HBV therapies are being combined with peg-IFN to create combination therapies targeting the virus and the host immune response; these include myrcludex B and nucleic acid polymers. However, the side effect profile of peg-IFN is a challenge.

Toll-like receptor agonists. Toll-like receptors (TLRs) are a critical part of the initial sensing of viral infection in humans, initiating intracellular pathways that induce antiviral mediators including interferons, cellular immune mediators, and antiviral cytokines (46). Early studies have reported that the activation of TLR-mediated pathways results in suppression of HBV replication in vitro and in vivo. An oral agonist of TLR-7 was studied in woodchucks and chimpanzees, and induced a decline in serum HBV DNA and HBsAg levels as well as hepatic HBV DNA

(47, 48). However, when studied in humans, the TLR-7 agonist induced HBV-specific immune responses but no changes were noted in serologic markers of HBV, including HBsAg, HBV DNA, HBeAg, and HBV RNA (49, 50). A novel oral agonist for TLR-8 (GS-9688, selgantolimod) is currently in phase II study. In vitro, selgantolimod induces cellular immune mediators, IFN-γ, and antiviral cytokines including TNF-α (51). Similar to that of TLR-7, a phase Ib study of GS-9688 did not see significant changes for HBsAg, HBV DNA, HBcrAg, or HBV RNA (49). There are currently TLR-7, -8, and -9 agonists in phase I and II trials.

Checkpoint inhibitors. Checkpoint inhibitors against PD-1 and PD-L1 have been shown to be effective in several malignancies by reversing T cell exhaustion and restoring antitumor immunity (52). Given the role of T cell exhaustion in chronicity of HBV infection and the recalcitrance of the latent reservoir in infected hepatocytes, checkpoint inhibition may have a role in latency reversal. Blockade of the PD-1 axis has been associated with restoration of HBV-specific T cell function in vitro (53). Clinical trials of anti-PD-1 are in progress.

Therapeutic vaccines. Therapeutic vaccines are developed to stimulate the host immune response to restore HBV-specific immune control with suppression of HBV replication and ultimately HBsAg loss. To date, early human study results have been disappointing (54). It is likely that a therapeutic vaccine will need to be used in combination with other antiviral or immunomodulatory therapies to achieve functional cure of HBV.

CONCLUSION

Chronic HBV infection causes significant morbidity and mortality globally. Nucleoside analogue therapies have had a dramatic impact on the rates of liver disease and liver-related complications, including death from HBV infection. Yet, due to the need in many cases for prolonged therapy, if not lifelong therapy, uptake of and access to NA therapy globally are low. The rapid discovery of direct-acting antivirals for chronic HCV infection and the dramatic impact of these antivirals on the health of people with HCV infection have renewed interest in pursuing curative therapies for HBV infection. The differences in the viral life cycles, particularly the lack of a nuclear phase for HCV infection, suggest that cure for HBV may be more complicated and more difficult to achieve than for HCV. In addition, current NA therapies are safe and well tolerated (unlike the peg-INF that was used for HCV infection prior to direct-acting antivirals); thus, as we proceed with clinical trials of novel HBV agents, care must be taken to ensure the safety of our research participants. The safety profiles of any new therapies must be comparable to that of NAs. Due to the likely role that immune restoration will play in therapeutic strategies, there is a risk of hepatic flares. Hepatic flares in the setting of active HBV infection can result in fulminant liver failure and death. Several programs have had to close or halt due to safety concerns and/or severe adverse events. There are currently multiple novel agents with unique mechanisms of action in early phases of human study, and many more are clamoring to follow. We must remain vigilant and proceed cautiously so that we can maintain the appropriate balance of risk and benefit.

DISCLOSURE STATEMENT

S.N. receives research grants from Gilead and AbbVie. She serves as an event adjudication committee member for FHI360 and PRA/BMS and a consultant for BioMarin and Vir Biotechnology. A.S.L. receives research grants from Assembly, Bristol-Myers Squibb, Gilead, and TARGET. She

serves on the advisory board or as a consultant for Gilead, GlaxoSmithKline, Huahui, Spring Bank, and TARGET.

LITERATURE CITED

1. WHO (World Health Organ.). 2017. *Global hepatitis report*. Rep., WHO, Geneva, Switz. **https://www.who.int/hepatitis/publications/global-hepatitis-report2017/en/**
2. Polaris Obs. Collab. 2018. Global prevalence, treatment, and prevention of hepatitis B virus infection in 2016: a modelling study. *Lancet Gastroenterol. Hepatol.* 3(6):383–403
3. Schweitzer A, Horn J, Mikolajczyk RT, et al. 2015. Estimations of worldwide prevalence of chronic hepatitis B virus infection: a systematic review of data published between 1965 and 2013. *Lancet* 386:1546–55
4. Foreman KJ, Marquez N, Dolgert A, et al. 2018. Forecasting life expectancy, years of life lost, and all-cause and cause-specific mortality for 250 causes of death: reference and alternative scenarios for 2016–40 for 195 countries and territories. *Lancet* 392:2052–90
5. Terrault NA, Lok ASF, McMahon BJ, et al. 2018. Update on prevention, diagnosis, and treatment of chronic hepatitis B: AASLD 2018 hepatitis B guidance. *Hepatology* 67:1560–99
6. Dore GJ, Feld JJ, Thompson A, et al. 2020. Simplified monitoring for hepatitis C virus treatment with glecaprevir plus pibrentasvir, a randomised non-inferiority trial. *J. Hepatol.* 72:431–40
7. Gupta N, Mbituyumuremyi A, Kabahizi J, et al. 2019. Treatment of chronic hepatitis C virus infection in Rwanda with ledipasvir-sofosbuvir (SHARED): a single-arm trial. *Lancet Gastroenterol. Hepatol.* 4:119–26
8. Naggie S, Muir AJ. 2017. Oral combination therapies for hepatitis C virus infection: successes, challenges, and unmet needs. *Annu. Rev. Med.* 68:345–58
9. Seitz S, Urban S, Antoni C, Böttcher B. 2007. Cryo-electron microscopy of hepatitis B virions reveals variability in envelope capsid interactions. *EMBO J.* 26:4160–67
10. Sureau C, Salisse J. 2013. A conformational heparan sulfate binding site essential to infectivity overlaps with the conserved hepatitis B virus A-determinant. *Hepatology* 57:985–94
11. Yan H, Zhong G, Xu G, et al. 2012. Sodium taurocholate cotransporting polypeptide is a functional receptor for human hepatitis B and D virus. *eLife* 1:e00049
12. Schreiner S, Nassal M. 2017. A role for the host DNA damage response in hepatitis B virus cccDNA formation—and beyond? *Viruses* 9:125
13. Marcellin P, Gane E, Buti M, et al. 2013. Regression of cirrhosis during treatment with tenofovir disoproxil fumarate for chronic hepatitis B: a 5-year open-label follow-up study. *Lancet* 381:468–75
14. Ahn SH, Park YN, Park JY, et al. 2005. Long-term clinical and histological outcomes in patients with spontaneous hepatitis B surface antigen seroclearance. *J. Hepatol.* 42:188–94
15. Liu J, Yang HI, Lee MH, et al. 2014. Spontaneous seroclearance of hepatitis B seromarkers and subsequent risk of hepatocellular carcinoma. *Gut* 63:1648–57
16. Simonetti J, Bulkow L, McMahon BJ, et al. 2010. Clearance of hepatitis B surface antigen and risk of hepatocellular carcinoma in a cohort chronically infected with hepatitis B virus. *Hepatology* 51:1531–37
17. Lai CL, Yuen MF. 2013. Prevention of hepatitis B virus-related hepatocellular carcinoma with antiviral therapy. *Hepatology* 57:399–408
18. Liaw YF, Sung JJ, Chow WC, et al. 2004. Lamivudine for patients with chronic hepatitis B and advanced liver disease. *N. Engl. J. Med.* 351:1521–31
19. Yuen MF, Chen DS, Dusheiko GM, et al. 2018. Hepatitis B virus infection. *Nat. Rev. Dis. Primers* 4:18035
20. Kim GA, Lim YS, An J, et al. 2014. HBsAg seroclearance after nucleoside analogue therapy in patients with chronic hepatitis B: clinical outcomes and durability. *Gut* 63:1325–32
21. Yuen MF, Wong DK, Fung J, et al. 2008. HBsAg seroclearance in chronic hepatitis B in Asian patients: replicative level and risk of hepatocellular carcinoma. *Gastroenterology* 135:1192–99
22. Yip TC, Wong GL, Chan HL, et al. 2019. HBsAg seroclearance further reduces hepatocellular carcinoma risk after complete viral suppression with nucleos(t)ide analogues. *J. Hepatol.* 70:361–70
23. Cornberg M, Lok AS, Terrault NA, Zoulim F. 2020. Guidance for design and endpoints of clinical trials in chronic hepatitis B—report from the 2019 EASL-AASLD HBV Treatment Endpoints Conference. *J. Hepatol.* 72:539–57

24. Chan HL, Thompson A, Martinot-Peignoux M, et al. 2011. Hepatitis B surface antigen quantification: why and how to use it in 2011—a core group report. *J. Hepatol.* 55:1121–31
25. Yip TC, Lok AS. 2020. How do we determine whether a functional cure for HBV infection has been achieved? *Clin. Gastroenterol. Hepatol.* 18:548–50
26. Cornberg M, Wong VW, Locarnini S, et al. 2017. The role of quantitative hepatitis B surface antigen revisited. *J. Hepatol.* 66:398–411
27. Liu Y, Jiang M, Xue J, et al. 2019. Serum HBV RNA quantification: useful for monitoring natural history of chronic hepatitis B infection. *BMC Gastroenterol.* 19:53
28. Giersch K, Allweiss L, Volz T, et al. 2017. Serum HBV pgRNA as a clinical marker for cccDNA activity. *J. Hepatol.* 66:460–62
29. Wang J, Shen T, Huang X, et al. 2016. Serum hepatitis B virus RNA is encapsidated pregenome RNA that may be associated with persistence of viral infection and rebound. *J. Hepatol.* 65:700–10
30. Seto WK, Wong DK, Fung J, et al. 2014. Linearized hepatitis B surface antigen and hepatitis B core-related antigen in the natural history of chronic hepatitis B. *Clin. Microbiol. Infect.* 20:1173–80
31. Testoni B, Lebossé F, Scholtes C, et al. 2019. Serum hepatitis B core-related antigen (HBcrAg) correlates with covalently closed circular DNA transcriptional activity in chronic hepatitis B patients. *J. Hepatol.* 70:615–25
32. Suzuki F, Miyakoshi H, Kobayashi M, Kumada H. 2009. Correlation between serum hepatitis B virus core-related antigen and intrahepatic covalently closed circular DNA in chronic hepatitis B patients. *J. Med. Virol.* 81:27–33
33. Bogomolov P, Alexandrov A, Voronkova N, et al. 2016. Treatment of chronic hepatitis D with the entry inhibitor myrcludex B: First results of a phase Ib/IIa study. *J. Hepatol.* 65:490–98
34. Zoulim F, Lenz O, Vandenbossche JJ, et al. 2020. JNJ-56136379, an HBV capsid assembly modulator, is well-tolerated and has antiviral activity in a phase 1 study of patients with chronic infection. *Gastroenterology* 159(2):521–33
35. Yuen M. 2019. *First clinical experience with RNA interference [RNAi]-based triple combination therapy in chronic hepatitis B (CHB): JNJ-73763989, JNJ-56136379 and a nucleos(t)ide analogue (NA)*. Paper presented at the American Association for the Study of Liver Diseases Annual Meeting, Nov. 8–12, Boston, MA
36. Ma X, Lalezari J, Nguyen T, et al. 2019. *Interim safety and efficacy results of the ABI-H0731 phase 2a program exploring the combination of ABI-H0731 with Nuc therapy in treatment-naive and treatment-suppressed chronic hepatitis B patients*. Paper presented at the International Liver Congress, Apr. 10–14, Vienna, Austria
37. Sulkowski M. 2019. *Continued therapy with ABI-H0731+Nrtl results in sequential reduction/loss of HBV DNA, HBV RNA, HBeAg, HBcrAg and HBsAg in HBeAg-positive patients*. Paper presented at the American Association for the Study of Liver Diseases Annual Meeting, Nov. 8–12, Boston, MA
38. Wooddell CI, Yuen MF, Chan HL, et al. 2017. RNAi-based treatment of chronically infected patients and chimpanzees reveals that integrated hepatitis B virus DNA is a source of HBsAg. *Sci. Transl. Med.* 9:eaan0241
39. Gane E, Locarnini SA, Lim TH, et al. 2018. *First results with RNA interference (RNAi) in chronic hepatitis B (CHB) using ARO-HBV*. Paper presented at the American Association for the Study of Liver Diseases Annual Meeting, Nov. 9–13, San Francisco, CA
40. Bazinet M, Pântea V, Cebotarescu V, et al. 2017. Safety and efficacy of REP 2139 and pegylated interferon alfa-2a for treatment-naive patients with chronic hepatitis B virus and hepatitis D virus co-infection (REP 301 and REP 301-LTF): a non-randomised, open-label, phase 2 trial. *Lancet Gastroenterol. Hepatol.* 2:877–89
41. Bazinet M, Pântea V, Placinta G, et al. 2020. Safety and efficacy of 48 weeks REP 2139 or REP 2165, tenofovir disoproxil, and pegylated interferon alfa-2a in patients with chronic HBV infection naïve to nucleos(t)ide therapy. *Gastroenterology* 158:2180–94
42. Finter NB, Chapman S, Dowd P, et al. 1991. The use of interferon-α in virus infections. *Drugs* 42:749–65
43. McHutchison JG, Lawitz EJ, Shiffman ML, et al. 2009. Peginterferon alfa-2b or alfa-2a with ribavirin for treatment of hepatitis C infection. *N. Engl. J. Med.* 361:580–93
44. Xie Q-L, Zhu Y, Wu L-H, et al. 2015. The efficacy and safety of entecavir and interferon combination therapy for chronic hepatitis B virus infection: a meta-analysis. *PLOS ONE* 10:e0132219

45. Isorce N, Lucifora J, Zoulim F, Durantel D. 2015. Immune-modulators to combat hepatitis B virus infection: from IFN-α to novel investigational immunotherapeutic strategies. *Antivir. Res.* 122:69–81
46. Ma Z, Cao Q, Xiong Y, et al. 2018. Interaction between hepatitis B virus and Toll-like receptors: current status and potential therapeutic use for chronic hepatitis B. *Vaccines* 6:6
47. Lanford RE, Guerra B, Chavez D, et al. 2013. GS-9620, an oral agonist of Toll-like receptor-7, induces prolonged suppression of hepatitis B virus in chronically infected chimpanzees. *Gastroenterology* 144:1508–17, 1517.e1–10
48. Menne S, Tumas DB, Liu KH, et al. 2015. Sustained efficacy and seroconversion with the Toll-like receptor 7 agonist GS-9620 in the Woodchuck model of chronic hepatitis B. *J. Hepatol.* 62:1237–45
49. Gane EJ, Lim YS, Gordon SC, et al. 2015. The oral toll-like receptor-7 agonist GS-9620 in patients with chronic hepatitis B virus infection. *J. Hepatol.* 63:320–28
50. Janssen HLA, Brunetto MR, Kim YJ, et al. 2018. Safety, efficacy and pharmacodynamics of vesatolimod (GS-9620) in virally suppressed patients with chronic hepatitis B. *J. Hepatol.* 68:431–40
51. Mackman RL, Mish M, Chin G, et al. 2020. Discovery of GS-9688 (selgantolimod), as a potent and selective oral Toll-like receptor 8 agonist for the treatment of chronic hepatitis B. *J. Med. Chem.* 63(18):10188–203
52. Pardoll DM. 2012. The blockade of immune checkpoints in cancer immunotherapy. *Nat. Rev. Cancer* 12:252–64
53. Bengsch B, Martin B, Thimme R. 2014. Restoration of HBV-specific CD8+ T cell function by PD-1 blockade in inactive carrier patients is linked to T cell differentiation. *J. Hepatol.* 61:1212–19
54. Lim SG, Agcaoili J, De Souza NNA, Chan E. 2019. Therapeutic vaccination for chronic hepatitis B: a systematic review and meta-analysis. *J. Viral Hepat.* 26:803–17

Annual Review of Medicine

Volume 72, 2021

Contents

Indexes

Errata

An online log of corrections to *Annual Review of Medicine* articles may be found at http://www.annualreviews.org/errata/med