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Research priorities for the discovery of a cure for chronic hepatitis B: Report of a workshop

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Abstract

In early 2017, the Hepatitis B Foundation invited 30 experts in the fields of hepatitis B and liver cancer research to identify projects they deemed important to the goal of finding a cure for chronic hepatitis B and D and the diseases with which these viral infections are associated. They were also asked to identify general categories of research and to prioritize sub-project topics within those areas. The experts generally agreed on broadly defined areas of research, but there was usually little difference between the highest and lowest scoring projects; for the most part, all programs described in this document were considered valuable and necessary. An executive summary of this discussion was recently published (Alter et al., Hepatology 2017). The present manuscript reports the areas of research identified by the workshop participants, provides a brief rationale for their selection, and attempts to express differences among the priorities assigned to each area of research, when such distinctions were expressed.

Keywords

Hepatitis B; Hepatitis D; Antiviral therapy; Hepatocellular carcinoma

1. Introduction

In early 2017, the Hepatitis B Foundation invited 30 experts in the fields of hepatitis B and liver cancer research to identify projects they deemed important to the goal of finding a cure for chronic hepatitis B and D and for diseases with which these viral infections are associated. They were asked both to identify general categories of research and to prioritize sub-project topics within those areas. All 30 invited experts participated in a virtual workshop, and 25 of them served as co-authors of this report. An executive summary of the discussion was published in *Hepatology* (Alter et al., 2017). This article provides a detailed account of that workshop.

The experts generally agreed on the broadly defined areas of research needed to discover curative therapies, and there was usually little difference between the highest and lowest scoring projects. For the most part, all programs described in this document, which were proposed by a sub-set of experts, were considered valuable and necessary. This article reports the areas of research identified, with a brief rationale for their selection, and attempts to express differences in assigned priorities, when such distinctions were expressed.

A recent manuscript (Revill et al., 2016) describes general goals for hepatitis B research and is the basis for a new coalition, the “International Coalition for the Elimination of Hepatitis B Virus”, (ICE-HBV) (http://ice-hbv.org/). Here we build upon their report and add details, to create a specific road map for policy makers from government and other funding institutions, and for those planning long-term research infrastructures.

2. Toward curative therapy for chronic hepatitis B

From a clinical perspective, the goal of therapeutics is to reduce a patient's risk of death due to liver disease, ideally to that of a person who has never been infected with HBV, or perhaps more realistically, to that of one with a resolved infection (Block et al., 2016). Resolved infections are usually characterized by robust humoral and cellular immune recognition of virus-infected hepatocytes, with consequent loss of detectable antigenemia and viremia (McMahon et al., 2016; Rehermann and Bertoletti, 2015). This goal is ambitious, but we believe it can be achieved through an aggressive, focused effort. Multiple parallel research approaches will be needed, because different treatment strategies may be required, depending upon the age of patients, length of time of infection, status of immune response, genotype of the infecting virus and other variables (Bertoletti and Kennedy, 2015; McMahon et al., 2014; McMahon, 2016; Zhang et al., 2015).

There is a general consensus that achieving sustained repression of cccDNA within infected cells, by silencing or degradation, is a very high priority (Alter et al., 2017; Revill et al., 2016; Guo & Guo, 2015) How this can be achieved is more uncertain, therefore exploring the possibilities of targeting other viral and host functions that influence the viral life cycle must also be pursued. Whether the restoration of a beneficial immunological recognition of the virus is necessary for sustained viral repression is also unclear, and must also be explored.

The current standard of care is based upon inhibition of the viral polymerase with nucleoside analogues (*i.e.* lamivudine, entecavir, tenofovir, telbivudine). Polymerase inhibitors can reduce the five-to ten-year risk of liver cancer in older patients, who have a lifetime risk of ~25% if untreated (40% in males, lower in females). However, although these inhibitors suppress viral replication and retard disease progression, they do not cure the disease (Gordon et al., 2014; Lok et al., 2016; Papatheodoridis et al 2015), and viremia and disease progression typically rebound once therapy is stopped (Terrault et al., 2016). New therapeutics, used alone or in combination with current approaches, are clearly needed.

Prevention of hepatitis B virus infection by vaccination has been successful but although it has reduced the incidence of new infections in the USA and in other parts of the world (Buckley and Strom, 2016), there are still at least 250 million chronically infected individuals, for whom the vaccine is of no benefit (Gish et al., 2015a, b; Hoofnagle et al., 2007; El-Serag and Davila, 2011). Although vaccination efficacy is very high, coverage is incomplete in many endemic regions, and it can also be ineffective in areas in which maintenance of the cold chain is difficult. Immigration from areas of high to low endemicity (such as the USA), together with the current burden of chronic infection, will also maintain the virus in populations for decades to come, unless better curative therapies are developed soon (Kowdley et al., 2012; Gish et al., 2015a, b). Taken together, the humanitarian, public health and financial burdens of chronic hepatitis B are enormous and provide strong motivation to find a cure.

There has been tremendous progress in basic scientific and clinical research on hepatitis B and D, but the therapeutic benefit of targeting host and viral functions beyond the

polymerase remains largely unexplored. Important, specific questions remain unanswered, including:

- If and how HBV cccDNA can be targeted?
- What other viral targets are possible?
- How does chronic infection cause liver cancer?
- Can, and to what extent can carcinogenesis be blocked even without a cure for chronic infection?
- What would be the effect of a curative therapy given later in life? Will adult therapies also work in children, and *vice versa*?
- To what extent will management and cure of HBV also effectively manage and cure disorders associated with HDV?

These are some of the research priorities outlined in Table 1, raised by the group of experts. Taken together, this survey identifies and organizes those key research areas into subcategories, facilitating further discussion.

3. General research priorities

Table 1 shows an organization of the results of initial discussions of broad areas of research need. Each area stated in Table 1 is then divided into specific subcategories throughout the text. The subcategorizations, although not arbitrary, are governed more by practical reasons to facilitate discussion than by biology. In this regard, there was some thought that HBV and HDV should be considered together, while others strongly felt that, at least for some questions, HDV should be distinct. The category of HBV- and HDV-associated hepatocellular carcinoma (HCC) is also discussed narrowly, in terms of virology, and research prioritization might benefit from a more comprehensive consideration.

Each subcategory, and then each project within a subcategory, was ranked by the authors and contributors as of primary (1) or secondary (2) importance. At least 4 separate responses were used to designate the priority as “1” or “2” (except in the case of HDV, where fewer than 4 scores were received).

However, it is important to note, that all categories were considered to be essential, and the “ranking” values for category and subcategory priority, were very close. We therefore use the descriptors of “enthusiasm”, since there was enthusiasm for all projects.

4. Specific subcategories of research priorities

4.1. Virology and viral therapeutics

Current HBV therapeutics are limited to the interferons and nucleoside analog polymerase inhibitors (NUCs), (Terrault et al., 2016). However, used alone or in combination, neither interferons or NUCs are reliably curative, and they are recommended for use in only a minority of the chronically infected (Terrault et al., 2016).

In addition to the HBV viral polymerase, there are at least four other “obvious” virus specified gene products that could serve as therapeutic targets, and possibly even more than four, depending upon how the activity associated with virus gene product is assayed (Block et al., 2016). The major HBV gene products, in addition to the polymerase, were therefore listed and subdivided into specific research project possibilities.

4.1.1. Areas identified as top priorities

4.1.1.1. cccDNA (Table 2).: As shown in Table 2, repression by elimination or transcriptional repression of cccDNA was considered to be a top priority, with there being nearly unanimous ranking of this category as highest priority. cccDNA is the nuclear form of the viral genome, which persists as a mini chromosome (Seeger and Mason, 2015). It is essential to the viral life cycle, and is the source of all viral gene products, with the exception of subgenomic transcripts that may come from viral DNA integrated in to the host genome (Seeger and Mason, 2015). HBV cccDNA appears to be very stable, persisting as a mini-chromosome, separate from the host chromosome, and continues to specify viral gene products even years after highly effective repression of the viral polymerase, by polymerase inhibitors (Werle-Lapostolle et al., 2004).

cccDNA persistence is the reason that polymerase inhibitor therapy cannot be discontinued and is the source of rebounding virus after cessation of polymerase inhibition and the reactivation of virus in individuals treated with immunosuppressive agents (McMahon et al., 2014; Lok et al., 2016). A better understanding of the biology and mechanisms of regulation of cccDNA metabolism was therefore considered to be a high priority. Several approaches, including drug-induced epigenetic modification of the cccDNA mini-chromosome, leading to transcriptional silencing or destabilizing cccDNA with (CRISPR) designer endonucleases, are just two possibilities that can be envisioned (Guo and Guo, 2015; Kennedy et al., 2015). Taken together, compared with all other virological targets, the greatest enthusiasm was expressed for pursuing cccDNA as a target.

4.1.1.2. HBx (Table 3).: The function of HBx has been debated since recognition that the viral genome specifies its open reading frame, and there is a growing consensus that it is essential for replication of the virus *in vivo* (Seeger and Mason, 2015). There is also compelling evidence that HBx plays a role, perhaps critical, in enabling transcription of HBV cccDNA. HBx could therefore offer a viral protein target that, if inhibited, would repress HBV cccDNA transcription. Understanding HBx and making it a target for new therapeutics generated elevated interest,and great enthusiasm, as illustrated in Table 3.

4.1.1.3. HBs (HBsAg) (Table 4).: HBsAg is an essential viral protein, being needed for the secretion and infectivity of the virion (Seeger and Mason, 2015; Block, et al 2011). In addition, HBsAg, is present in high concentrations in the circulation and has been implicated in immune modulation and maintaining chronicity (Op den Brouw et al., 2009). That said, although the evidence for HBsAg’s role in maintaining immunological tolerance to HBV in chronically infected patients is unproved, reduction in circulating HBsAg is associated with more favorable outcomes and failure to reduce HBsAg is a strong negative predictor of interferon responsiveness (Moucari et al., 2009; Gish et al., 2010; Lee et al., 2011).

Similarly, HBsAg decline in patients treated with NUCs (Wursthorn et al., 2010) is also associated with favorable outcomes, such as durable off drug responses and HBsAg loss.

Thus, despite the fact that HBsAg reduction could be a consequence of these clinical outcomes, rather than their cause, some of the respondents expressed considerable enthusiasm for determining whether HBsAg might have an immunomodulatory role in HBV pathogenesis beyond its function as the viral envelope polypeptide that might justify its targeting for therapeutic intervention. Moreover, since HDV requires HBsAg to complete its life cycle, effective inhibition of HBsAg should also inhibit HDV. Therefore, there was considerable enthusiasm for the study of, and targeting of HBsAg.

4.1.2. Areas assigned secondary priority

4.1.2.1. HBc (HBcAg) (Table 5).: HBcAg is an essential viral protein. In addition to mediating capsid formation, it has been reported to be involved in viral and possibly host gene regulation and immuno-modulation, although the evidence for this is limited. For these reasons, HBcAg has been considered an attractive antiviral target, and there are already multiple HBcAg targeting drugs currently in development (Liang et al., 2015). For these reasons, it received good, but secondary, enthusiasm from reviewers.

4.1.2.2. HBe (HBeAg) (Table 6).: HBe is a proteolytic processing product of a polypeptide that over-laps HBc open reading frame, and is secreted from infected hepatocytes (Miller, 1987; Seeger and Mason, 2015). Its function is unclear, but has been associated with higher viremia in people, regulation of host immune recognition of the virus (Millich and Liang, 2003; McMahon et al., 2014) and even establishment of chronicity, in murine studies (Tian et al., 2016). HBe negative hepatitis is also associated with poorer outcome (McMahon et al., 2014). It rates high, therefore, as a research priority but because of a current lack of functional assay for HBe and uncertainty about its role in virology and pathogenesis, its prioritization as an antiviral target followed other virology research priorities.

4.1.2.3. RNAaseH (Table 7).: RNAaseH is an essential viral gene product, and inhibiting its function should be, in principle, as antiviral as inhibiting the polymerase (Tavis and Lomonosova, 2015). HIV and all retroviruses specify and depend upon RNAaseH enzymes, but small-molecule inhibitors that are selective and safe have been elusive. RNAaseH as a target for HBV was considered to be an important line of research exploration and received enthusiasm.

4.1.2.4. Integrated HBV DNA (Table 8).: HBV genomic DNA can be found integrated into the chromosomes of infected cells, and is regularly detected within the chromosomes of hepatocellular cancers (Brechot et al., 1980; Shafritz et al., 1981; Block et al., 2003). However, integration is not an essential step in the viral productive life cycle, since cccDNA is the necessary and sufficient template for viral transcripts (Seeger and Mason, 2015). However, there is evidence that integrated DNA plays a role in oncogenesis in some (perhaps a minority) of HCC cases (Lin et al., 2016; Yang et al., 2017). Moreover, there are recent reports that HBV integrated DNA (which is usually not capable of transcribing

full viral genomic RNA), does produce significant amounts of HBsAg (Wooddell et al., 2015; 2017), and thus could contribute to pathogenesis, to the extent that HBsAg contributes to pathogenesis (Gish et al., 2015a, b). There is therefore renewed interest in exploring the role of integrated HBV DNA in the pathogenesis of chronic hepatitis and the possibility of targeting it therapeutically.

4.1.2.5. Other virus-specified targets for antiviral intervention (Table 9).: It is possible that there are viral gene products, still not identified, that can serve as antiviral targets. This possibility needs to be explored, for a complete and satisfactory understanding of the biology of HBV, as well as for therapeutic purposes. Also included in this "other" category is the concept of identification of drugs that cause selective elimination of infected cells. There was good enthusiasm for these concepts.

4.2. Immunological and non-immunological host factors

The virus depends upon the host to complete its life cycle, and on host functions, which include constitutive cell processes as well as innate and adaptive immunological defenses. Innate and adaptive immune responses are critical, on one hand, in resolving infection and, on the other, in promotion of viral pathogenesis. For example, most adult infections induce a robust immunological response that results in resolution (Bertoletti and Kennedy, 2015). However, chronic hepatitis B is characterized by an unbeneficial and inadequate immunological response to HBV (Chisari and Ferrari, 1995).

Taken together, a better understanding of the virus-host relationship is central to cure research. Indeed, there is a school of thought that believes restoration of a beneficial immunological response to HBV is essential to achieving even a functional cure, let alone, complete clinical cure. Therefore, analysis of the host factors and immune responses were considered to be extremely high priorities for both clinical and basic sciences.

4.2.1. Areas identified as top priorities

4.2.1.1. Adaptive immune response (Table 10).: T and B cell responses to HBV play a central role in both viral pathogenesis and resolution of infection (Bertoletti and Kennedy, 2015; Chisari and Ferrari, 1995; Rehermann and Bertoletti, 2015). Circulating antibody to HBs that neutralizing infection is considered a hallmark of resolution and protection, but a robust and beneficial T cell response is required for clearance of infected cells. The fact that these systems fail in chronic infection is well established and we are beginning to understand how to restore them. Our current understanding of chronic infection largely depends on observational clinical studies. Lack of biologically relevant animal models prevents better understanding of the immunological mechanism of chronic infection and development of therapeutics to restore a functional immune response that ultimately resolves the virus infection. New animal models will allow for critical experiments to examine the impact of immune restoration on the course of chronic HBV infection. The key questions are highlighted in the list in the Table, and all were considered to be priorities.

4.2.1.2. Innate immune responses (Table 11).: Interferons are effective in a minority of patients, and activators of innate host defenses can have HBV-suppressive affects *in vivo* and

in vitro (Isogawa et al., 2005; Chang et al., 2012). There is therefore a great deal remaining to be learned about the interplay between innate host defenses and the regulation of chronic hepatitis B, and this is considered to be a high priority area for research.

4.2.2. Areas assigned secondary priority

4.2.2.1. Non-immunological host factors (Table 12).: HBV depends upon host functions to complete its life cycle, from binding to hepatocytes, to transcription of its RNA and finally to uncoating and morphogenesis of the virion. Different viral genotypes may even behave differently with respect to natural history, pathogenesis and treatment response (Kim et al., 2011). Knowing these steps and the basis of genotype-dependent pathogenesis and drug sensitivity may provide clues for new strategies of intervention.

4.3. Clinical questions (Table 13)

Separating our clinical and basic science questions is somewhat arbitrary, since there is and should be, overlap. And, although clinical studies permit addressing questions not possible with basic science systems, coordination is important, and this is reflected in some of the research management priorities in this section. Never the less, clinical phase studies do require distinct resources, compared to the basic sciences, and are usually separately managed and are thus listed in the Clinical Questions Table (Table 13).

4.4. Hepatocellular carcinoma and cirrhosis (Table 14)

HCC and cirrhosis are the most important morbid consequences of chronic hepatitis B and D. The incidence of both diseases is growing at an alarming rate in the USA and the world, and is of extremely high public health importance. Of course, although HBV is a major cause of liver cirrhosis and HCC, these diseases are caused by many other etiologies, and these subject areas require broader attention than is given here.

This section was intended to identify the most significant priorities from the perspective of HBV and HDV. In this regard, advances in prevention of HCC and liver cirrhosis by effective management of hepatitis B and D, effective risk screening and cancer surveillance, can go a long way toward preventing disease. However, it is necessary to manage the diseases, once established, and new, more effective treatments are needed. A better understanding of the molecular mechanisms of these diseases and new therapeutics are critical.

4.5. Hepatitis D (Table 15)

HDV requires HBV co-infection of the same hepatocytes to complete its life cycle (Hughes et al., 2011; Alfaiate et al., 2015). Elimination of HBV would therefore be expected to result in elimination of HDV, and there was some discussion of whether to separate HDV and HBV priorities. There are approximately 15–25 million people world-wide infected with HDV, and the super-infection of chronic hepatitis B is usually associated with a far more aggressive disease than hepatitis B alone (Noureddin and Gish, 2014). HDV clearly presents unique virological and clinical challenges, which might be overlooked if not considered on its own. It was therefore finally determined to treat some HDV subject areas separately from HBV; the key areas are presented as lists that parallel those used for HBV.

4.6. Research reagents (Table 16)

The study of hepatitis B and D, and even of hepatocellular carcinoma, have been frustrated by a lack of critical research tools, ranging from experimental models (*in vitro* and *in vivo*) to standardized controls. Some resources, such as standardized tools, can be easily provided and have great impact, with little investment. Others, such as new experimental systems, will require greater investment of effort and funds, but will provide tools that will assist and expedite the success of many of the projects proposed in this document.

It is also important to note, as was considered in the September 2016 conference convened by the September, American Association for the Study of Liver Diseases (ASSLD) and the European Association for the Study of the Liver (EASL), that as new medicines for chronic hepatitis B enter clinical phases of evaluation, new reagents and assays are likely to be needed. That is, the current endpoints to evaluate medication performance are largely dependent upon determination of viral load (viral DNA levels in the blood), liver-derived enzymes detected in the blood (e.g. ALT, AST), and antibodies to HBsAg. These tests may prove inadequate for new approaches being anticipated. There should therefore be an effort to produce new biomarkers, or to redeploy currently available tests, so as to be able to determine:

- **a.** which patients will benefit most from a given therapy;
- **b.** when to begin and stop therapy;
- **c.** the extent to which a new therapy is achieving its intended biochemical effect on its clinical target; and
- **d.** the superiority and/or complementarity of a new therapy, compared to other HBV medications, given alone or in combination.

5. Conclusion: moving forward to the elimination of hepatitis B

It has been more than 50 years since the discovery of the hepatitis B virus and its association with hepatitis and liver cancer (Block et al., 2016)). There is now a growing urgency for development of potentially curative therapies, and there is also optimism that the time is right for significant progress, given the combination of newly available resources and new information about the virus. For example, there are now robust tissue culture systems to infect and grow HBV, permitting study of many steps in its life cycle beyond reverse transcription. Similarly, there is a growing body of clinical information, providing a new understanding of clinical and immunopathological complexities.

Although it is clearly important to explore all viral gene products and replication steps for intervention opportunities, our team of experts concluded that the elimination of HBV cccDNA is the most likely approach to produce either a complete cure or prolonged control of infection after a finite course of therapy. The extent to which this can be durably achieved with drugs, biologicals, genetic manipulations and/or immunomodulation are major questions to be answered. While transcriptional silencing of cccDNA may be easier to achieve than its physical elimination, it would probably require life-long treatment to produce life-long effects, unless therapy were to trigger a durable downstream effect, such as

immune-mediated destruction or non-cytolytic elimination of cccDNA from infected cells. A vigorous, comprehensive research effort, involving multiple, complementary approaches, must therefore be taken.

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Table 1

General research areas identified by workshop participants that should be addressed to achieve improved understanding of the pathogenesis of chronic hepatitis B and D and to discover new breakthrough therapies.

Broad Areas of Research Action	
Virology & Viral Therapeutics	Improve understanding of HBV and HDV virology; emphasize research to define the molecular mechanisms responsible for cccDNA biogenesis, homeostasis and decay; Exploit each step in the life cycle of the virus for therapeutic purposes
Immunology & non-immunological host factors	Improve understanding of adaptive and innate immunology of HBV and HDV acute, chronic, and resolved infections, and reactivation; expand research on viral and host functions influencing the viral life cycle with a goal of exploiting for therapeutic purposes
Clinical Questions	Design human-phase efficacy and toxicity trials to provide information about virology and disease mechanism and to adapt to the likely new mechanisms associated with new therapeutics
Hepatocellular carcinoma and liver cirrhosis	Improve understanding of molecular pathways leading to cirrhosis and HCC, to develop new early detection markers and new therapeutics
Hepatitis D	Improve understanding of hepatitis D molecular biology, effect upon disease progression and immunology to explore new therapeutics
Research Reagents	Develop and standardize new research reagents and systems to study HBV, HDV, and HCC for the purposes of drug discovery and development
Other	Establish new, and expand current, inter-institution and inter-laboratory collaborative networks for basic science discovery and validations

Table 2

Research on covalently closed circular DNA (cccDNA) was identified as a top priority for discovering a cure for chronic hepatitis B. Six specific research areas were identified. In this and the following tables, each area was ranked in importance as either first (1) or second (2).

cccDNA	
Project	**Rank**
Therapeutic benefit of inhibitors of cccDNA, alone and in combination	1
Basic academic research to define the molecular mechanisms responsible for cccDNA biogenesis, homeostasis, and decay, and to determine the half-life of preformed cccDNA in cell culture and animal models	1
Preclinical pharmaceutical research to discover and develop small molecules and biologics that directly target cccDNA life cycle vulnerabilities discovered in the priority above	1
Clinical research designed to test small molecules and biologics to either (a) eliminate cccDNA from the liver, or (b) reduce the number of cccDNA-positive hepatocytes (to a point at which all newly formed virus particles are neutralized by circulating anti-HBs antibodies)	1
Determination of mechanisms of cccDNA regulation	2
Basic academic and pharmaceutical research to determine if genetic, epigenetic, or other strategies that are known to (a) suppress cccDNA transcription, or (b) to prevent its recycling (e.g. capsid inhibitors) can eliminate cccDNA in HBV-infected cells in cell culture and animal models	2

Table 3

Research on HBx function was identified as a top priority for discovering a cure for chronic hepatitis B.

HBx	
Project	**Rank**
Determine and validate the functions of HBx most important to its biological and virological roles	1
Specifically, confirm and expand upon understanding of HBx's HBV regulatory action on cccDNA other viral functions	1
Determine the benefit of HBx as an antiviral target, alone and in combination	1
Determine and validate role(s) of HBx in hepatitis B pathogenesis, including HCC	2
Develop antiviral agents that target HBx	2

Table 4

Research on the function of hepatitis B surface antigen was identified as a top priority for discovering a cure for chronic hepatitis B.

HBs	
Project	**Rank**
Determine and validate role(s) of HBs in hepatitis B chronicity and pathogenesis, including HCC and immunosuppression	1
Develop and validate antiviral agents that target HBs	1
Benefit of HBs as an antiviral target, alone or in combination	2
Determine detailed molecular mechanisms of HBs morphogenesis and role in virion production and secretion	2

Table 5

Research on the hepatitis B core protein was considered to be a secondary priority for discovering a cure for chronic hepatitis B.

HBc	
Project	**Rank**
Benefit as an antiviral target, alone or in combination	1
Development of antiviral agents that target HBc	1
Determine if HBc has regulatory functions for viral and cell genes	2
Determine HBc role(s) in HBV life cycle, pathogenesis, and immunomodulation, beyond its role as a structural component of the virus	2

Table 6

Research on HBe function was considered to be a secondary priority for discovering a cure for chronic hepatitis B.

HBe	
Project	**Rank**
Benefit as an antiviral target, alone or in combination	1
Development of antiviral agents that target HBe	1
Determine if HBe has regulatory functions for viral and cell genes	2
Determine the role(s) of HBe in the HBV life cycle, pathogenesis and immunomodulation beyond its role as a structural component of the virus	2

Table 7

Research on RNase H function was considered to be a secondary priority for discovering a cure for chronic hepatitis B.

RNaseH	
Project	**Rank**
Development of antiviral agents that target RNaseH	1
Benefit as an antiviral target, alone or in combination	2

Table 8

Research on HBV DNA integration into the host cell chromosomes was considered to be a secondary priority for discovering a cure for chronic hepatitis B.

Integrated HBV DNA	
Project	**Rank**
Can integrated HBV DNA cause independent production of HBsAg, making loss of HBsAg and/or measuring levels of HBsAg in some persons not a useful treatment endpoint or functional cure?	1
How can information about integrated HBV DNA be used in the management (treatment and risk assessments) of disease?	1
Better understanding of the role integrated HBV DNA plays in oncogenesis and in contributing gene products that affect chronic liver disease	2
Determination of the mechanism of regulation of integrated DNA expression	2

Table 9

Certain subcategories of research that do not specifically fall into the those listed in Tables 2–9 were considered to be of secondary importance for discovering a cure for chronic hepatitis B.

Other Viral Targets	
Project	**Rank**
Development of strategies leading to the selective elimination of HBV- infected cells. Leading candidates are (a) therapeutic immunization, (b) checkpoint inhibition, and (c) bispecific antibody therapy that can deliver cytolytic or noncytolytic antiviral effector molecules selectively to infected cells	1
Identify viral gene products other than those currently known, that regulate HBV and its pathogenicity	1
How can information about other viral gene products and RNAs that regulate the HBV life cycle and host pathogenicity be used in the management (treatment and risk assessments) of chronic hepatitis B?	2

Table 10

Among studies of immunological and non-immunological host factors, research on adaptive immune responses was considered to be a top priority for discovering a cure for chronic hepatitis B.

Adaptive Immune Response	
Project	**Rank**
How can information about HBV immunovirology be used in the management (treatment and risk assessments) of HBV disease?	1
Role of T cells, T cell exhaustion, function of patient age, length of time of infection, other viral load issues	1
Possibility and benefits of restoration of immunorecognition of HBV	1
Safety of restoring cytolytic immune responses in patients with late-stage disease	1
Role of B cells, as antibody producers and other possible functions	2
Role of immunological checkpoints and other regulators of immunoresponsiveness in maintaining chronicity and disease	2

Table 11

Among studies of immunological and non-immunological host factors, research on innate immune response to HBV infection was considered to be a top priority for discovering a cure for chronic hepatitis B.

Innate Immune Response	
Project	**Rank**
Comprehensive analysis of the hepatic innate and adaptive immune systems, and their roles in chronicity	1
How can understanding the innate and adaptive immune systems help in the therapeutic management of HBV?	1
Role of innate defenses in regulating HBV infection, and in pathogenesis	1
Specific innate host defense factors that are responsible for the role of the innate defense response in influencing HBV acute and chronic infections, and in HCC	2
Nature and mechanism of HBV refractoriness to type I IFN	2

Table 12

Among studies of immunological and non-immunological host factors, research on nonimmunological host factors was considered to be a secondary priority for discovering a cure for chronic hepatitis B.

Non-Immunological Host Factors	
Project	**Rank**
More complete understanding of the HBV life cycle, from receptor binding and entry, to nuclear transport and uncoating, and morphogenesis and secretion of virus and particles, with a focus on steps that are vulnerable to intervention	1
Determination of if and how information about the role of host factors in HBV virology can be used in the management (treatment and risk assessments) of HBV disease	1
Role of viral genotypes in pathogenicity, natural history and responsiveness to therapy	1
Role of host factors in determining outcomes (acute versus chronic, responsiveness to therapies, development of disease	2
Role of genetic factors in determining outcomes of disease natural history and responsiveness to therapy	2

Table 13

Workshop participants considered research on clinical questions to be a top priority for discovering a cure for chronic hepatitis B. The questions in the table were ranked for their relative importance.

Clinical Questions for HBV and HDV	
Project	**Rank**
Determine if people in different clinical stages respond differentially to different experimental therapeutics	1
What triggers seroconversion after years of chronic infection?	1
Explore molecular, cellular, and immunological mechanisms of reactivation in the absence of, and as a function of medical drug interventions	1
What does reactivation via medical interventions, such as immunosuppressive agents, anti-TNF, etc., teach about chronicity?	1
Does “functional” cure and management of HBV result in sustained clinical benefit to those with HDV?	1
Why do only subpopulations treated with IFN or NUCs become functionally cured?	1
Expansion of current, and development of new clinical trial programs and networks in the US and internationally, to evaluate HBV and HDV	1
Establish clinical centers, worldwide, that can carry out trials under FDA standards	2
Possibility that DAAs are sufficient to achieve functional (conditional) cure, and if responsiveness to the drugs varies with clinical stage	2
Are people responsive to treatments in a stage-specific manner?	2
Expansion of current, and development of new clinical networks and clinical programs that integrate basic and clinical studies to better understand HBV human biology and develop best management	2
Create a public database that will allow comprehensive and timely reporting of all drugs, new or old, showing association with HBV reactivation	2

Table 14

Workshop participants considered the study of hepatocellular carcinoma and liver cirrhosis associated with chronic hepatitis B to be a high priority for discovering a cure.

Hepatocellular Carcinoma/Viral Oncology/Liver Cirrhosis	
Project	**Rank**
Development of safe and well-tolerated oral agents for primary prevention of HCC in patients at increased risk (those with chronic HBV and HDV, among others)	1
Development of safe and well-tolerated oral,agents for prevention of HCC recurrence post-surgical resection and liver transplantation in patients with chronic HBV and HDV (among others)	1
Test current promising and already available agents (including statins, and other complementary agents) in large RCTs for primary and secondary prevention of HCC and liver cirrhosis in persons with chronic viral hepatitis	1
Develop more effective screening and surveillance tools for HCC and liver cirrhosis (e.g., biomarker assays, imaging modalities)	1
Better understanding of molecular mechanisms leading to HBV/HDV-associated HCC	1
Development of more effective agents and locoregional therapies for treating established HCC in patients with chronic HBV and HDV (among others)	2
Develop HCC research and treatment clinical networks to share specimens, and clinical, histological, and imaging data.	2
Collect more detailed and more accurate HCC incidence data	2
Determine the role of metabolic liver diseases and obesity in contributing to hepatitis associated HCC	2

Table 15

Workshop participants considered that research needed to achieve a cure for chronic hepatitis D was a secondary priority.

Hepatitis D-Specific Antivirals and Immunology	
Project	**Rank**
Development of next generation anti-HDV agents based on HDV-specific gene products	1
Role of B and T cell exhaustion in chronicity and pathogenesis	1
How can information about HDV immunovirology be used in the management (treatment and risk assessments) of HDV disease	1

Table 16

Workshop participants identified multiple specific reagents and technical capabilities that are needed to support the research described in Tables 2–15.

Reagents and technical capabilities for HBV & HDV research	
Project	**Rank**
Development of animal models of human HBV and HDV disease	1
Standardized virological and immunological assays for critical analytes [e.g., HDV RNA, HBV cccDNA, HBs (quantitative), anti-HDV, Ig, cytokine response] and T and B cells assays as new endpoints for therapeutic drug evaluation	1
Development of cell lines and primary human liver cell systems that support the full infectious cycle, and are authentic liver lines	2