Diagnosing and Screening for Hepatitis D Viral Infection

Robert G. Gish, MD[1,2]

1. Robert G. Gish Consultants LLC, La Jolla CA. 2. Medical Director, Hepatitis B Foundation, Doylestown PA, Robert G. Gish, Email: rgish@robertgish.com

Abstract

Hepatitis delta virus (HDV) infection causes the most virulent form of viral hepatitis and occurs in some patients chronically infected with hepatitis B virus (HBV). Antiviral therapies used for HBV and hepatitis C virus (HCV) are not effective against HDV, and interferon is partially effective with 1-20% maintained viral response (MVR) viral clearance, further exacerbating poor outcomes in HDV patients. Despite increased morbidity and mortality, screening for HDV is limited due to perceptions of low prevalence, historically limited access to testing, and poor awareness and lack of medications for treatment. Recent epidemiological studies reveal that HDV prevalence is increasing in areas of historically low prevalence, creating urgency for the implementation of widespread HDV screening and awareness. Emerging therapies have shown promise in treating HDV and further reinforce the need for increased HDV screening in individuals with chronic HBV infection.

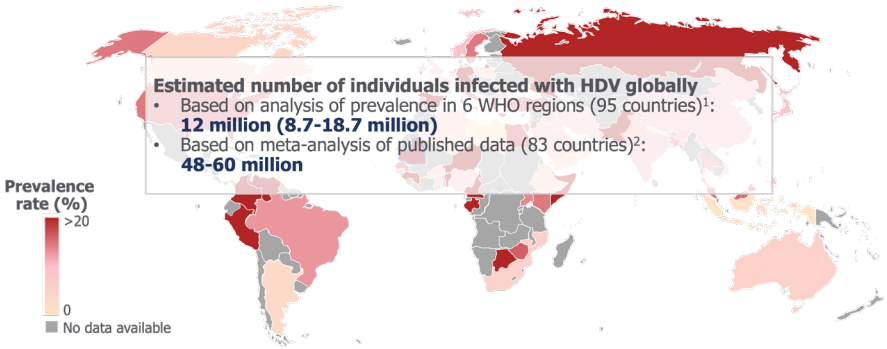
Introduction

HDV is a small satellite of HBV that can cause the most aggressive form of viral hepatitis.[1-3] HDV requires the presence of HBV for viral replication and dissemination in liver cells.[1-3] HDV infection leads to rapid progression of liver disease, increasing the risk of cirrhosis, decompensated cirrhosis, liver transplant, hepatocellular carcinoma (HCC), and mortality compared to HBV monoinfection.[4-6]

Clinical features of HDV infection can be acute or chronic (ie, chronic hepatitis D [CHD]). Acute HDV infections can occur as simultaneous coinfections with HBV or as superinfections in patients with chronic HBV. Acute coinfections follow a course similar to that of HBV acute infections, but with a more rapid progression and worse clinical outcomes. Acute coinfections lead to significantly severe outcomes if acute hepatitis is present and if the patient remains chronically infected. Fortunately, acute HDV coinfections are cleared in 95% of adult patients.[7,8] Superinfections have a more severe disease course with an increased risk of acute liver failure, compared to acute HDV coinfections.[9] Superinfections are often mistaken as HBV flares or acute HBV infections in undiagnosed HBV patients.[4,10]

CHD occurs in 5% of HDV coinfections and over 90% of superinfections.[11,12] It is clinically defined as the detection of HDV antibodies more than 6 months after infection, with detectable serum HDV RNA or detectable HDV antigen (HDAg) in the blood. Although HDAg can be measured, it is not used clinically to diagnose or monitor HDV infection.[13,14] CHD is the most serious type of viral hepatitis.[15] It progresses to cirrhosis in 10%-15% of cases within 2 years and roughly 80% of cases within 15 years.[16]

Despite the burden and increased mortality caused by HDV infection, the true prevalence of HDV infection is unknown. HDV seroprevalence global estimates range from 15 million to 72 million people (Figure 1).[17-19] Certain areas with high HDV prevalence around the world have been identified and are referred to as hot spots.[17] Migration



WHO=World Health Organization.

Figure 1. Approximately 4.5%-13% of HBsAg-positive carriers are coinfected with HDV. Estimated number of HDV-infected individuals varies significantly based on epidemiological analyses done.[19-21]

from these hot spots is thought to increase the spread of HDV into countries with traditionally low prevalence.[22] This spread of HDV could result in increased infection rates globally if not properly identified and monitored.

HDV prevalence in the US is unknown due to incomplete reporting, but several studies have aimed to evaluate prevalence in different populations (Figure 2). Gish and colleagues measured the prevalence of HDV in HBV cases to be 8%, but the study only tested 42% of HBV cases.[23] A study by the Veterans Affairs medical system found a prevalence of 3.4%, but testing methods used were found to be suboptimal.[24] In a study of a US Midwestern population, 12% of HBV cases were tested for HDV and a 3.3% prevalence was obtained.[25] The prevalence of HDV

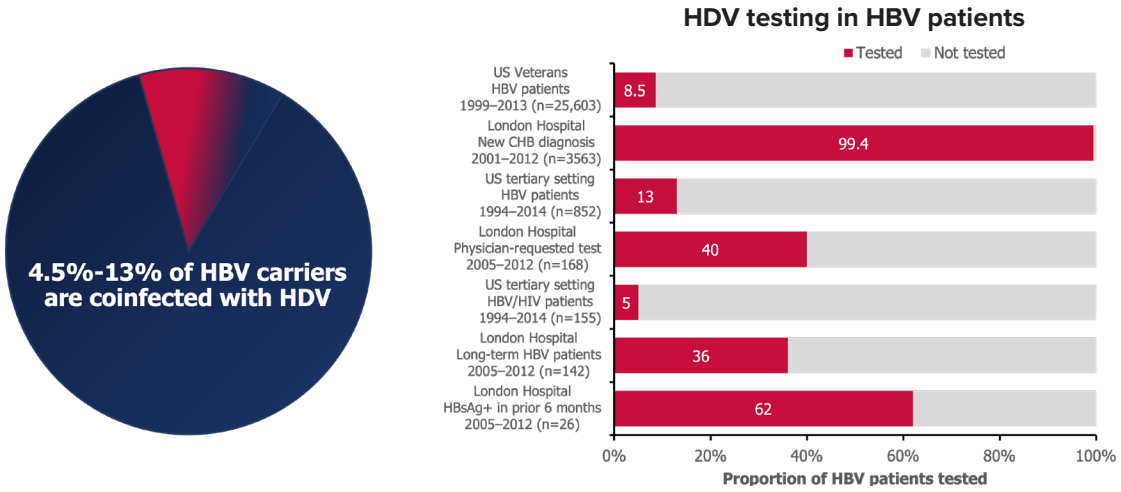


Figure 2. HDV screening is conducted inconsistently in HBV patients despite the known risk of coinfections and superinfections. This lack of consistency leads to a lack of comprehensive prevalence data.[19,21,24,25] CHB=chronic hepatitis B virus; HBsAg=hepatitis B surface antigen; HBV=hepatitis B virus; HDV=hepatitis D virus; HIV=human immunodeficiency virus.

infection among people who inject drugs (PWID) in the US is also increasing. In a study of the PWID population in Baltimore, Maryland, around 50% of HBV-infected PWID were also infected with HDV.[26] A study of HBV-infected PWID conducted in San Francisco found a 36% prevalence of HDV.[27]

The lack of comprehensive prevalence data is attributed to several factors, including limited HDV awareness and interest, lack of samples to validate available tests, limited number of labs that offer HDV testing, and lack of testing and screening guidelines. This lack of awareness and interest has also caused a lag in the development of effective, well-tolerated medications for HDV. Current treatment for HDV is off-label use of PEG-IFN-α, which has low efficacy for HDV, is not well tolerated, and has high rates of relapse. Thus, the limited awareness of HDV ultimately hinders multiple facets of effective management in affected patients. US data on HDV suggests that less than 1% of HBV patients are infected with HDV, but our data could be severely skewed due to the available tests. We still do not trust test sensitivity, and false-negative rates may be as high as 15% for antibody. PCR performance knowledge is even worse due to most labs utilizing primers for *italiense* (genotype 1) , and the test S/S in other genotypes has yet to be fully explored. Issues with tests are improving over time with the standardization of assays, development of new assays, and harmonization across genotypes.[28,29]

HDV Awareness

Perceptions of HDV prevalence have varied widely over time. After implementation of the HBV vaccine (thought to also protect against HDV), a decrease in HDV prevalence was observed.[30] This contributed to the widespread perception that HDV was no longer a threat.[31] This perception of eradication led to a decrease in HDV testing, further contributing to perception of decreased prevalence.

Unfortunately, recent studies have demonstrated that HDV prevalence is increasing around the world. In European countries with traditionally low prevalence, recent studies revealed a trend toward increasing prevalence due to immigration.[32-34] A study conducted in Australia found similar trends.[35] These studies support the need for universal, routine testing of patients with HBV for anti-HDV to avoid underestimating the true prevalence of HDV infection.

Although these studies demonstrate the need for HDV screening, awareness of HDV among healthcare providers and at-risk patients is low.[22] HDV testing was most likely to be ordered by gastroenterologists (49%) and internal medicine physicians (40%).[25] However, high-risk patients are more likely to seek aid from other specialties, such as infectious disease and addiction medicine specialists. Therefore, to increase screening rates in high-risk populations, it is essential to raise awareness among addiction, infectious disease, and other primary care specialties.

HDV Diagnostic Tests

Currently, the main testing method for HDV infection is antibody detection by enzyme-linked immunosorbent assays (ELISA) or radioimmunoassay and confirmation by HDV RNA detection. Positive detection of HDV RNA for more than 6 months indicates chronic infection. One key challenge in HDV diagnostic testing is the comparability, validity, and standardization of HDV diagnostic tests. Validating these tests has been particularly difficult due to the limited availability of HDV-positive samples. The first standardized test for HDV RNA, specific to HDV *italiense*, was created by the World Health Organization in 2013.[36] Although *italiense* is the most common HDV genotype, there are seven others (Figure 3).[17,19,37] To date, only a limited number of labs offer testing for these markers and none of these tests have been FDA cleared. Below is a summary and comparison (Table 1) of the commercially available tests for HDV.

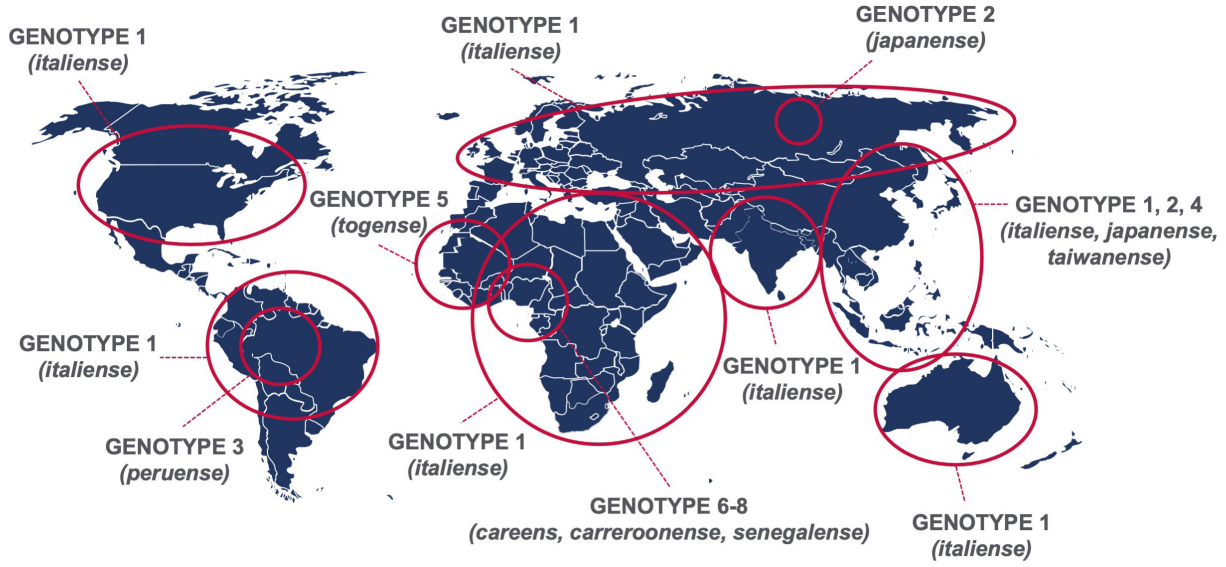


Figure 3. Eight HDV genotypes with varying geographical distribution.[17,19,37] *Italiense* (genotype 1) is the most common HDV genotype.

HDV RNA

HDV RNA is measured using qualitative or quantitative reverse transcription polymerase chain reaction (RT-PCR).[38,39] Utilized as a marker for HDV replication, HDV RNA tests are positive in all patients with chronic infection. HDV RNA tests become negative when the virus is cleared spontaneously or with treatment. This test is used to monitor and predict treatment response.[36,39]

Samples	Markers	
Serum	**Anti-HDV IgG**	**HDV RNA**
Acute HDV Coinfection	+	+
Acute HDV Superinfection	+	+
Chronic HDV	+	+ (10^5-10^7 copies/mL)
Chronic Active HDV-HBV	+	+ (10^5-10^7 copies/mL)
Cirrhosis	+	+ (10^3-10^7 copies/mL)
HDV Recovery	+	-
Liver	**HDV RNA**	
Chronic HDV	+	

Table 1. HDV Markers in the Clinical Form of HDV

Tests Not Utilized Clinically

HDV Antigen

HDV antigen is expressed in liver cell nuclei. To detect HDV antigen, a liver biopsy is required, followed by measurement using immunohistochemistry.[31,40] The emergence of molecular assays has significantly reduced the use of tissue immunohistochemistry.

Anti-HDV Antibody

Anti-HDV IgG is detected in all individuals infected with HDV and can persist long after viral clearance, leading to false-positive results. Anti-HDV IgM is only detectable within 2-3 weeks of onset and disappears after 2 months in patients with acute infection or persists long-term in patients with chronic infection.[17,41] HDV IgM tests have limitations, including not being a reliable marker of HDV viremia and not providing the ability to discern between acute versus chronic infections.[42]

HDV Testing and Screening Guidelines

Currently there is no standard consensus on HDV testing and screening (Figure 4). The American Association for the Study of Liver Diseases (AASLD) suggests that total antibody tests be performed in patients with chronic HBV who are at high risk, such as PWID and immigrants from hot spots.[43] However, there are no peer-reviewed

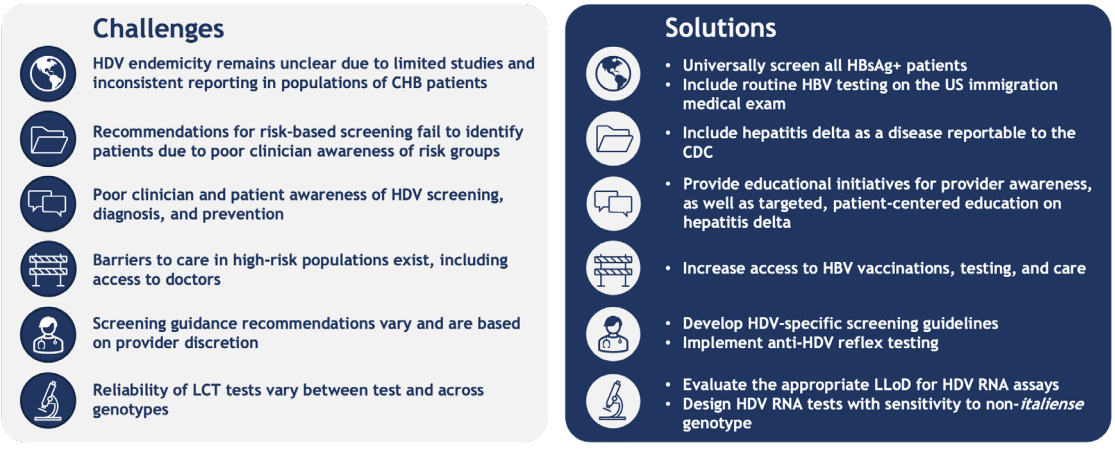


Figure 4. Awareness, educational, and diagnostic challenges exist that hinder HDV screening.[25,43-46] Proposed solutions are presented here to improve HDV screening. CDC=Centers for Disease Control and Prevention; CHD=chronic hepatitis B; HBsAg+=hepatitis B surface antigen positive; HBV=hepatitis B virus; HDV=hepatitis D virus; LDT=lab-developed test; LLoD=lower limit of detection; RNA=ribonucleic acid.

references that suggest risk-based testing captures all HDV-infected individuals or that this testing approach will lead to HDV elimination. In fact, AASLD has moved away from risk-based testing for HCV and has shifted to universal testing recommendations.[47] Due to the lack of evidence for risk-based testing, the European Association for the Study of the Liver (EASL) and the Hepatitis B Foundation recommend testing for HDV in all chronic hepatitis B patients.[48,49] Scaling up screening services was outlined as a key WHO strategy to meet their goal of eliminating viral hepatitis as a public health problem by 2030.[50]

Discussion

Chronic HDV is the most severe form of viral hepatitis, occurring exclusively in patients infected with HBV. HDV is associated with increased risk of cirrhosis, faster progression to hepatic decompensation, and increased mortality, compared to HBV alone.[4] Despite the increased morbidity and mortality, the global prevalence of HDV is unknown due to several factors.[51-53] HDV is thought to be a rare disease in major markets like the US and Europe, but prevalence appears to be increasing in these markets.[23-25,32-34] Increased awareness of HDV is crucial to bolster prevention and early intervention efforts to avoid progressive fibrosis and cirrhosis.[19] Widespread screening for HDV in HBV-infected individuals is crucial to this endeavor.

Historically, uncertainty around accuracy and availability of HDV testing contributed to global underdiagnosis of HDV. In 2013, the first and only standardized HDV RNA test was created by the WHO, bolstering the validity and accuracy of HDV RNA testing.[36] Continued standardization and increased availability of accurate HDV diagnostic tests will allow a better understanding of HDV's prevalence and facilitate early intervention.[48]

Furthermore, increasing HDV screening in HBV-positive patients is critical in improving diagnostic rates and outcomes of patients living with HDV. Currently, screening and testing for HDV is done sparingly. Screening criteria often vary, with some countries applying a risk-based approach and others focusing on the country of origin.[19] It is clear from the limited prevalence data available that screening for HDV should be standardized for all HBV-positive patients, rather than just high-risk HBV-positive patients. Widespread screening will capture the true prevalence of HDV and reduce risk of poor outcomes in patients living with HDV.

Though bulevirtide is conditionally approved in the EU,[54,55] there are currently no effective, FDA-approved treatments for HDV, creating a clear unmet need for patients living with HDV.[48,56] Approval of novel therapies for HDV would likely drive increased awareness and demand for screening and testing in HBV-infected patients. Several products in late-stage trials have demonstrated promising efficacy in clearing HDV RNA. The development of these promising therapies sheds light on the current state of HDV.

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Disclosures

Dr. Robert G. Gish is a consultant for Gilead Sciences, Inc.

Affiliations

1. Robert G. Gish Consultants LLC, La Jolla, California, USA

2. Hepatitis B Foundation, Doylestown, Pennsylvania, USA

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