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Modern diagnostic approaches for hepatitis B virus infection

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Abstract: Hepatitis B virus (HBV) infection remains a major global public health problem due to its wide geographic distribution, high rate of chronicity, and strong association with progressive liver disease, including cirrhosis and hepatocellular carcinoma. Despite the availability of effective vaccination programs, HBV continues to pose a significant diagnostic and clinical challenge, particularly because many infected individuals remain asymptomatic for long periods. Accurate diagnosis relies on the combined interpretation of serological, molecular, and biochemical markers. This review highlights modern diagnostic approaches for HBV infection, focusing on the clinical significance of serological markers, the role of molecular techniques in disease monitoring, and methods used to assess liver injury severity.

Keywords: *Hepatitis B virus, HBV diagnosis, Serological markers, Molecular diagnostics, HBV DNA, Liver fibrosis, FibroScan, Chronic hepatitis B*

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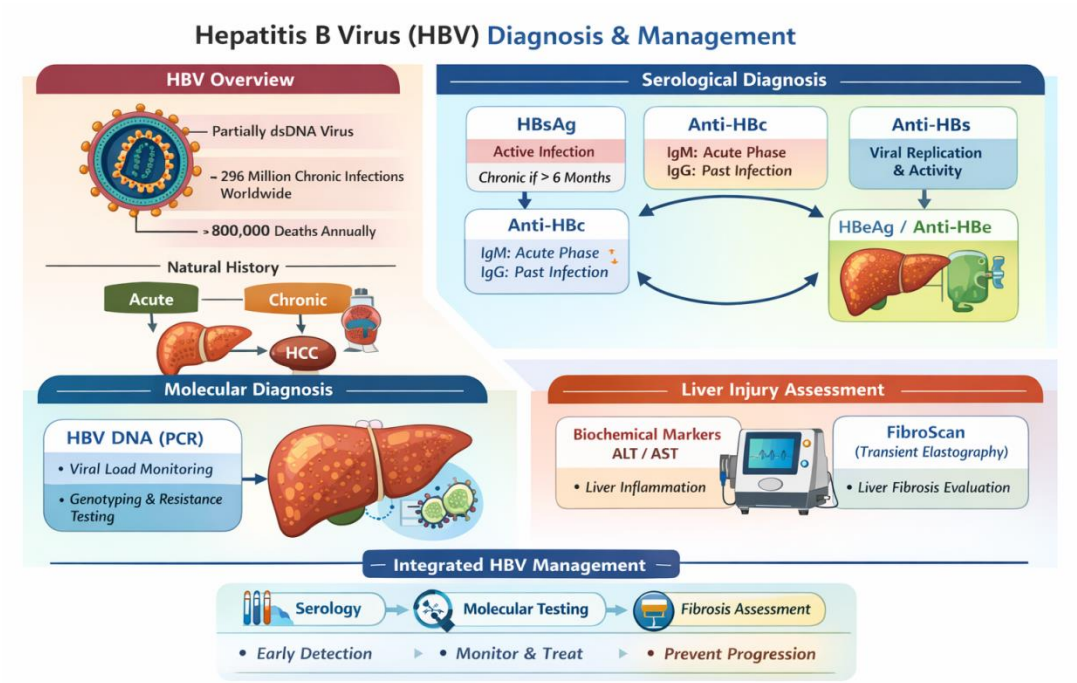
Introduction

Hepatitis B virus (HBV) is a double-stranded, partial-stranded DNA virus that is a leading cause of chronic liver disease worldwide. It has a dynamic disease course that can begin with acute infection and progress to cirrhosis or hepatocellular carcinoma. Modern diagnosis of HBV infection relies on an integrated approach combining serological markers, molecular diagnostics, and assessment of liver damage. Serological markers such as HBsAg, Anti-HBc, Anti-HBs, and HBeAg/Anti-HBe are used to determine the infection status, stage, and viral activity. Measurement of HBV DNA using PCR is the gold standard for estimating viral load, guiding treatment decisions, and monitoring response to therapy. This is complemented by assessing the severity of liver damage using biochemical markers (ALT, AST) and non-invasive techniques such as transient elastography (FibroScan) to estimate the degree of fibrosis. This comprehensive approach enables accurate diagnosis, effective follow-up, and a reduction in long-term disease progression and complications as shown in (Figure 1) [1]

Hepatitis B virus (HBV) is a partially double-stranded DNA virus belonging to the Hepadnaviridae family and is a major cause of acute and chronic liver disease worldwide. According to the World Health Organization, approximately 296 million people were living with chronic HBV infection in 2022, with more than 800,000 deaths annually attributed to HBV-related complications such as liver cirrhosis and hepatocellular carcinoma (HCC) [2]. These figures highlight the substantial global burden of the disease and the ongoing need for effective diagnostic and monitoring strategies.

HBV infection is characterized by a complex and dynamic natural history that depends on the interaction between viral replication and the host immune response. Infection acquired during adulthood is often self-limiting, whereas perinatal or early childhood infection carries a high risk of progression to chronic hepatitis [3]. The chronic form of HBV infection may remain clinically silent for years, during which progressive liver damage can occur without obvious symptoms. As a result, many patients are diagnosed only at advanced stages of disease, when therapeutic options are limited and prognosis is poor.

Accurate laboratory diagnosis plays a central role in the identification of infected individuals, determination of disease phase, assessment of infectivity, and selection of appropriate treatment strategies. Modern diagnostic approaches are based on the integration of serological markers that reflect viral antigens and host immune responses, molecular assays that quantify viral replication, and clinical tools that assess the extent of liver injury. This integrated diagnostic framework is essential for effective patient management, prevention of disease transmission, and reduction of HBV-related morbidity and mortality [4].



Serological markers in the diagnosis of HBV infection

Serological testing represents the cornerstone of HBV diagnosis and is routinely used as the first-line approach in clinical and epidemiological settings. The detection of specific viral antigens and antibodies allows clinicians to distinguish between acute and chronic infection, identify individuals with resolved infection or vaccine-induced immunity, and evaluate viral activity [5]. Correct interpretation of serological profiles requires understanding the temporal appearance and persistence of each marker during the course of infection.

Hepatitis B virus surface antigen (HBsAg)

Hepatitis B surface antigen (HBsAg) is the earliest detectable serological marker following HBV infection and reflects the presence of circulating viral particles. Detection of HBsAg indicates active HBV infection, regardless of clinical symptoms. Persistence of HBsAg for more than six months is a defining criterion for chronic HBV infection [6]. In recent years, quantitative measurement of HBsAg has gained clinical relevance, as declining HBsAg levels may reflect reduced transcriptional activity of covalently closed circular DNA (cccDNA) and improved response to antiviral therapy [7].

Anti-HBc antibodies

Antibodies directed against the hepatitis B core antigen (Anti-HBc) are markers of natural exposure to HBV and are absent in individuals with vaccine-induced immunity. IgM Anti-HBc appears during the acute phase of infection and is a key diagnostic marker, particularly during the window period when HBsAg has disappeared but Anti-HBs is not yet detectable [6]. IgG Anti-HBc persists for life and indicates past exposure to HBV, either in resolved infection or chronic disease. The presence of isolated Anti-HBc may suggest occult HBV infection, especially in immunocompromised patients, underscoring its clinical importance [8].

Antibodies to surface antigen (Anti-HBs)

Anti-HBs antibodies indicate protective immunity against HBV infection. Their appearance following clearance of HBsAg signifies recovery from natural infection, while their presence in the absence of Anti-HBc confirms immunity acquired through vaccination [9]. Measurement of Anti-HBs titers is clinically useful for evaluating vaccine response and determining the need for booster doses in high-risk populations such as healthcare workers and immunosuppressed individuals.

HBeAg and Anti-HBe system

Hepatitis B e antigen (HBeAg) and its corresponding antibody (Anti-HBe) serve as important markers of viral replication and infectivity. The presence of HBeAg is generally associated with high levels of HBV DNA and increased risk of transmission. Seroconversion to Anti-HBe usually reflects partial immune control of viral replication and is associated with improved clinical outcomes [10]. However, the emergence of precore and basal core promoter mutations may result in HBeAg-negative chronic hepatitis, emphasizing the need for molecular testing to accurately assess viral activity.

Molecular diagnosis and its role in disease management

Quantification of HBV DNA using polymerase chain reaction (PCR) techniques is the gold standard for assessing viral replication. HBV DNA levels are essential for confirming active infection, determining disease phase, guiding initiation of antiviral therapy, and monitoring treatment response [11]. Molecular assays also enable HBV genotyping, which may influence disease progression and response to therapy in certain patient populations.

Assessing the severity of liver injury

Assessment of liver injury is a critical component of HBV management. Biochemical markers such as alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are widely used indicators of hepatic inflammation, although they may not accurately reflect the degree of fibrosis [3]. Non-invasive

methods have increasingly replaced liver biopsy in routine practice. Transient elastography (FibroScan) is a validated technique for measuring liver stiffness and estimating fibrosis stage, offering a safe and reliable alternative for disease staging and longitudinal follow-up [12].

Interpretation of common serological patterns

Clinical status	HBsAg	Anti-HBc	Anti-HBs
Person not exposed to infection	Negative	Negative	Negative
Immunity acquired through vaccination	Negative	Negative	Positive
Recovery from previous infection	Negative	Positive	Positive
Acute infection	Positive	IgM Positive	Negative
Chronic infection	Positive	IgG Positive	Negative

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