

An update on Hepatitis B virus

Soumendra Nath Maity^{1-3*}, S. S. Kumar⁴, R. Vijayaraghavan¹, Rathnagiri Polavarapu^{2,5,6}

¹Department of Research, Saveetha Institute of Medical and Technical Sciences, Chennai, Tamil Nadu, India, ²Department of Clinical Research Laboratory, Genomix Molecular Diagnostics Pvt. Ltd., Hyderabad, Telangana, India, ³Department of Microbiology, Malla Reddy Institute of Medical Sciences, Hyderabad, Telangana, India, ⁴Department of Physiology, Vishnu Dental College, Bhimavaram, West Godavari, Andhra Pradesh, India, ⁵MNR Foundation for Research and Innovation, Sangareddy, Telangana, India, ⁶Genomix CARL Pvt. Ltd., Pulivendula, Andhra Pradesh, India

ABSTRACT

Hepatitis B virus (HBV) can cause life-threatening liver infection such as liver cirrhosis and hepatocellular carcinoma. In 1963, Blumberg reported 1st time about the new viral antigen from a blood sample of an Australian aborigine and named as "Australian antigen," and later this introduced as hepatitis B surface antigen protein of HBV. Approximately 257 million individuals are infected with HBV infection worldwide, and millions of death occurred annually. The virus can spread through sexual contact and parenteral and perinatal routes. HBV causes liver-associated diseases; it can be acute or chronic and symptomatic or asymptomatic disease. The current review provides a detailed account of HBV, its pathogenesis, diagnosis, and treatment.

Key words: Diagnosis, Hepatitis B virus, infections, treatment

INTRODUCTION

Hepatitis B virus (HBV) can cause life-threatening liver infection such as liver cirrhosis and hepatocellular carcinoma. In 1963, Blumberg reported 1st time about the new viral antigen from a blood sample of an Australian aborigine and named as "Australian antigen," and later this introduced as hepatitis B surface antigen (HBsAg) protein of HBV.^[1] HBV is the major public health problem worldwide. According to the WHO report, 257 million individuals are living with HBV infection. Although effective HBV vaccine available but due to lack of mass vaccination in under developed countries, hepatitis B infection increases rapidly with estimated 30 million new cases each year and 1 million die annually due to HBV disease.^[2] HBV infection can transmit through blood and body fluids and most infective than HIV-related disease.^[3] The present review article was undertaken to explore the pathogenesis, diagnosis, and treatment modes of hepatitis B.

CLASSIFICATION

HBV belongs to the Hepadnaviridae family, consisting of two genera: Orthohepadnavirus can be found in mammals and Avihepadnavirus found in birds. Due to high genetic variability in HBV, they are divided into 8 different genotypes A-H.

Another three clades of HBV isolates are named as HBV chimpanzees, HBV orangutans, and HBV gibbons.^[4]

STRUCTURE AND GENOMIC ORGANIZATION

HBV is a 42-nm enveloped, spherical, double-stranded DNA virus. It is also called as Dane particle. Other two particles are spherical

(20 nm) and filamentous (22 nm) particles composed of HBsAg and host-derived lipids without viral nucleic acids; thus, these are noninfectious.^[5] The complete HBV virion lipid envelopes consist of HBsAg which encoded inner nucleocapsid composed of hepatitis B core antigen (HBcAg) with attached viral DNA and polymerase enzyme. The viral genome is 3.2 kb pairs; partially double-stranded circular DNA and polymerase enzyme are attached with the 5' end of the minus strand.^[1] There are several open reading frames in the viral genome which encodes various proteins of HBV. The S region encodes HBsAg proteins, which further divided into three regions, pre-S1, pre-S2, and S. The core or C gene divided into two distinct regions of core and pre-core. Depending on the initiation of translation from the core and pre-core regions, C open reading frame encodes HBcAg or HBeAg. The P region consists of large polymerase protein gene which functionally divided into three forms, such as the reverse transcriptase domain for catalyzing genome synthesis, ribonuclease H domain for facilitating replication, and the terminal protein region for acting on the initiation of minus-strand synthesis. The HBV X region encodes HBxAg which is necessary for the prognosis of viral infection and plays a major role in oncogenic properties of HBV. Two direct repeats DR1 and DR2 in the 5' end of the plus strand are responsible for the strand-specific DNA synthesis. En1 and En2 are the two enhancer proteins for the expression of the liver-specific viral gene.^[6]

MODE OF TRANSMISSION

Approximately 257 million individuals are infected with HBV infection worldwide and millions of death occurred annually.^[7] The virus can spread through sexual contact and parenteral and perinatal routes. Transmission through needle stick injuries is the most common in health-care workers. HBV transmission can

Address for correspondence:

Soumendra Nath Maity, Department of Research, Saveetha Institute of Medical and Technical Sciences, Chennai, Tamil Nadu, India.
E-mail: nath.soumendra@gmail.com

Received: 10-04-2018

Revised: 25-05-2018

Accepted: 20-06-2018

occur due to unsafe blood transfusion to other individual. Babies are at high risk of HBV infection through their infected mother. Other way of HBV transmission is needle sharing between drug abusers, tattooing, etc.^[7,8]

PATHOGENESIS AND CLINICAL MANIFESTATIONS

HBV causes liver-associated diseases; it can be acute or chronic and symptomatic or asymptomatic disease. Blood is the major source of HBV infection. After entry into the host, virus starts replicating in the liver within 3 days' period. This is the asymptomatic phase, where virus can multiply inside the hepatocytes with minimal cytopathic effect. Ground-glass lesion is the main cytopathological appearance in HBV infection. The pathogenesis of HBV depends on the immune reaction of the host. HBxAg of HBV proved to be more pathogenic in nature due to its apoptosis-inducing properties.^[9] HBxAg mostly reported from hepatocellular carcinoma cases but less frequent in chronic Hepatitis B infection. Thus, HBx Ag is one of the oncogenic factors in HBV infection.^[10] Pathogenicity and virulence properties are varies in different HBV genotypes. HBV genotypes A, D, and E are most commonly isolated from Africa, Europe, and USA. Perinatal transmission of HBV is less frequent in these countries due to a shorter period of high level viremia in mothers.^[11] HBV genotypes B and C are predominant in Asia. Perinatal transmission is higher to longer period of high level viremia and more progressive hepatitis with poor response to the antiviral treatment.^[12] Especially, all HBV-infected persons develop anti-HBc antibodies, which indicate significant ongoing HBV replication. Thus, anti-HBc antibody is one of the major serological markers of inflammatory liver infection. The presence of anti-HBs denotes low level of HBs Ag, but both anti-HBs and HBsAg can persist. Extrahepatic manifestations can occur due to HBs and HBe immune complexes, but it is not a major factor in viral hepatitis.^[13] The mechanism of immune escape of HBV is not clear, even in individual who resolves HBV infection, HBV can replicate even months without inducing an immune response.^[14] HBV persistence is due to the immune evasion properties of the virus. HBV infection has various clinical stages such as acute hepatitis, chronic hepatitis, extrahepatic infection, and occult or latent infection. Approximately two-thirds of patients with acute HBV infection develop mild asymptomatic illness. Some of the infected individual can show symptoms such as nausea, jaundice, and acute liver failure. The incubation period of acute HBV infection may vary from 2 to 6 months. The incubation period is mainly depend on the level of viral exposure.^[15] In this phase, patient may complain about fever, nausea, fatigue, etc. During acute hepatitis, serum ALT level increases with the high level of HBsAg and HBV DNA. Approximately 1% of patients may suffer from acute liver failure with jaundice. Acute liver failure patients need proper clinical management and may require liver transplantation. Some of the patients of acute HBV infection can persist to the disease and progress to chronic hepatitis. In severe chronic hepatitis or liver cirrhosis patients, 50% of them survive up to 5 years of time period.^[16,17] Many of the chronic hepatitis patients are asymptomatic or have mild symptoms. In chronic hepatitis condition, patients may suffer from ascites, encephalopathy, and gastrointestinal bleeding. Around 1–10% of HBV-infected patients may suffer from acute necrotizing vasculitis, glomerulonephritis, serum-sickness, etc.^[18] Some of

the patients may suffer from atypical or occult HBV infection. During occult HBV infection stage, patients may seronegative for all HBV markers but positive for viral DNA.^[19-21] Latent or occult HBV infection may be due to a mutation in HBV genome. However, there is no proper evidence of a relationship between mutations related to occult HBV with liver disease.^[22]

LABORATORY DIAGNOSIS OF HBV

HBV causes a wide variety of liver disease ranging from acute hepatitis, chronic hepatitis to liver cirrhosis, and hepatocellular carcinoma. The diagnosis of HBV infection is mainly based on clinical and serological examination. Other than this, biochemical and histopathological findings are also playing a major role in HBV diagnosis. There are several antigens of HBV, and their respective antibodies can be detected in serum after HBV infection.

DIAGNOSIS OF ACUTE HBV INFECTION

HBsAg-positive serology test indicates acute HBV infection. Repeat the test after 6 months detects the resolve infection or chronic. If the test is negative for HBsAg, then HBV infection can rule out.^[23] Detection of anti-HBc IgM is not more useful in the routine diagnosis of acute HBV infection. Anti-HBcIgM level can rise during chronic hepatitis as well as in acute HBV infection.^[24]

DIAGNOSIS OF CHRONIC HBV INFECTION

Chronic HBV infection can be determined by the presence of HBsAg >6 months in patient's serum. Positive anti-HBs test determines that the patient is immune to HBV and negative test determines no immunity. Detection of HBe Ag along with HBs Ag determines active viral infection with high viral load.^[24]

SEROLOGICAL DIAGNOSIS OF HBV

Several serological methods are available to differentiate the acute and chronic HBV infection. Enzyme immune assay proved to be highly sensitive and specific method to diagnose HBV infection. Enzyme-linked immunosorbent assay is based on the principle of the antigen and antibody reaction. The HBV antigen-based ELISA consists of known specific HBV antibody coated on microtiter well. Patient serum contains the specific antibody to pre-coated antigen and makes antigen-antibody complex which can detect by adding enzyme-conjugated secondary antibody and color substrate. HBV antibody-based ELISA wells coated with known specific antigen to HBV, if patient serum contains specific antibody to HBV reaction takes place. Antigen-antibody complex can be detected by adding enzyme-conjugated secondary antibody and substrate solution. Advancement of chemiluminescence enzyme immunoassay increases the sensitivity and specificity of HBs Ag detection assay. This method is more sensitive and specific method than other enzyme immune assay. Rapid immunochromatography method for the detection of HBs Ag in patient serum proved to be useful as a point of care diagnostic assay. Several studies were performed to check the sensitivity and specificity of commercially available kits. Although enzyme immune assay and rapid test assays are efficient for the diagnosis of HBV infection, due to the high mutation level in HBV genome, serological markers may undetectable in patient serum.^[1,13,24]

MOLECULAR DIAGNOSIS OF HBV

Thus, molecular diagnostic assays are highly encouraged to detect HBV DNA level in seronegative patients. In the past few decades, molecular diagnostic techniques are improved rapidly such as polymerase chain reaction (PCR), real-time PCR, transcription mediated amplification, loop-mediated isothermal amplification, rolling circle amplification, and ligase chain reaction.^[25] PCR method is the gold standard for the nucleic acid amplification. Very low level of HBV DNA can detect using HBV genome-specific primers, which can also differentiate various genotypes in HBV. Most of the PCR-based method can detect HBV DNA level ranging from 50 to 200 IU/ml.^[26,27] However, real-time detection-PCR (RTD-PCR) has higher sensitivity, which detects up to 5–10 IU/ml DNA copies. RTD-based PCR assay can detect the quantity of synthesized DNA in each cycle throughout the amplification process.^[28,29] Ligase chain reaction is useful to amplify shorter DNA targets, which uses DNA ligase enzyme and DNA polymerase enzyme to run the amplification.^[30-32] Recently, loop-mediated isothermal amplification assays gain more importance in molecular diagnostics for its application in point of care diagnostics. This method is highly sensitive method to detect HBV DNA using three pairs of specially designed specific primers. The amplification of nucleic acid is going on under constant temperature (58°C–65°C) in a heat block apparatus, which can yield a large amount of HBV DNA within 30–60 min of time period. The amplification product can visualize in agarose gel electrophoresis as well as under UV light for fluorescence production. The LAMP assay combined with biosensors for one-site detection of HBV DNA showing most efficient molecular tool for future diagnostic of HBV.^[33-36]

PROPHYLAXIS AND TREATMENT

HBV infection can be prevented by obtaining health safety measures and providing proper health education among population about the transmission and pathogenic nature of HBV disease. Other way to prevent this infection is immunization. Both active and passive immunizations are available commercially. In active immunization, recombinant proteins S and Pre-S1 and Pre-S2 gene of HBV are administered intramuscularly on deltoid muscle 0, 1, and 6 months' interval. In passive immunization, hyperimmune hepatitis B immune globulin is administered intramuscularly soon after exposure to infection. It can give protection against illness and carrier state but cannot prevent infection. Combination of active and passive immunization is required for babies born from the infected mother. There is no specific antiviral treatment available for acute infection, but in chronic HBV infection, interferon alpha with lamivudine or famciclovir treatment may give the better outcome.^[9,37-39]

CONCLUSION

The current review provides a detailed account of HBV, its pathogenesis, diagnosis, and treatment. This will help the readers to understand and help researchers to develop new methods of diagnosis and treatment.

REFERENCES

- Gerlich WH. Medical virology of hepatitis B: How it began and where we are now. *Gerlich Virol J* 2013;10:239.
- Blumberg BS. Hepatitis B virus, the vaccine, and the control of primary cancer of the liver. *Proc Natl Acad Sci* 1997;94:7121-5.
- GAR-Global Alert and. Hepatitis B. Available from: <http://www.who.int/csr/disease/hepatitis/whocdscsrlyo20022/en/index3.html>. Response. [Last accessed on 2018 Mar 22].
- Chevaliez S, Pawlotsky JM. Hepatitis C virus serologic and virologic tests and clinical diagnosis of HCV related liver disease. *Int J Med Sci* 2006;3:35-40.
- Gavilanes F, Gonzalez-Ros JM, Peterson DL. Structure of hepatitis B surface antigen. Characterization of the lipid components and their association with the viral proteins. *J Biol Chem* 1982;257:7770-7.
- Crowther RA, Kiselev NA, Bottcher B, Berriman JA, Borisova GP, Ose V, *et al.* Three dimensional structure of hepatitis B virus core particles determined by electron cryomicroscopy. *Cell* 1994;77:943-50.
- World Health Organization. Hepatitis B. 2017. Available from: <http://www.who.int/mediacentre/factsheets/fs204/en>. [Last accessed on 2018 Mar 20].
- Anna SF, Lok J, editors. *Sherlock's Diseases of the Liver and Biliary System*. 12th ed. USA: A Jhon Wiley & Sons Ltd.; 2011.
- Schuster R, Gerlich WH, Schaefer S. Induction of apoptosis by the transactivating domains of the hepatitis B virus X gene leads to suppression of oncogenic transformation of primary rat embryo fibroblasts. *Oncogene* 2000;19:1173-80.
- Nakamoto Y, Guidotti LG, Kuhlen CV, Fowler P, Chisari FV. Immune pathogenesis of hepatocellular carcinoma. *J Exp Med* 1998;188:341-50.
- Dusheiko GM, Bowyer SM, Sjogren MH, Ritchie MJ, Santos AP, Kew MC. Replication of hepatitis B virus in adult carriers in an endemic area. *J Infect Dis* 1985;152:566-71.
- Kao JH, Chen DS. Genotypes and clinical phenotypes of hepatitis B virus in patients with chronic hepatitis B virus infection. *J Clin Microbiol* 2002;40:1207-9.
- Gerlich WH, Kann M. *Hepatitis B. Topley and Wilson's Microbiology and Microbial Infections*. 10th ed. USA: John Wiley & Sons Ltd.; 2009.
- Wieland S, Thimme R, Purcell RH, Chisari FV. Genomic analysis of the host response to hepatitis B virus infection. *PNAS* 2004;101:6669-74.
- McMahon BJ, Alward WL, Hall DB, Heyward WL, Bender TR, Francis DP, *et al.* Acute hepatitis B virus infection: relation of age to the clinical expression of disease and subsequent development of the carrier state. *J Infect Dis* 1985;151:599-603.
- Liaw YF, Tai DI, Chu CM, Chen TJ. The development of cirrhosis in patients with chronic type B hepatitis: A prospective study. *Hepatology* 1988;8:493-6.
- Weissberg JI, Andres LL, Smith CI, Weick S, Nichols JE. Survival in chronic hepatitis B. An analysis of 379 patients. *Ann Intern Med* 1984;101:613-6.
- Dienstag JL. Immunopathogenesis of the extrahepatic manifestations of hepatitis B virus infection. *Springer Semin Immunopathol* 1981;3:461-72.
- Liang TJ, Blum HE, Wands JR. Characterization and biological properties of a hepatitis B isolated from a patient without HBV serological markers. *Hepatology* 1990;12:204-12.
- Paterlini P, Gerken G, Nakajima E, Terre S. Polymerase chain reaction to detect hepatitis B virus DNA and RNA sequences in primary liver cancers from patients negative for hepatitis B surface antigen. *N Engl J Med* 1990;323:80-5.
- Rehermann B, Ferrari C, Pasquinelli C, Chisari FV. The hepatitis virus persists for decades after patients recovery from acute viral hepatitis despite active maintenance of a cytotoxic T-lymphocyte response. *Nat Med* 1996;2:1104-8.
- Blum HE, Von Weizsacker F. The HBsAg negative, anti-HBc/anti-HBs positive patient. *Internist* 1991;32:262-6.
- Dufour DR, Lott JA, Nolte FS. Diagnosis and monitoring of

- hepatic injury. Recommendation for use of laboratory test in screening, diagnosis, monitoring. *Clin Chem* 2000;46:2050-68.
24. Mel K, Gail M, Martin P. The laboratory diagnosis of hepatitis B virus. *Can J Infect Dis Med Microbiol* 2005;16:65-72.
 25. Datta S, Chatterjee S, Veer V, Chakravarty R. Molecular biology of hepatitis B virus for clinicians. *J Clin Exp Hepatol* 2012;2:353-65.
 26. Pawlotsky JM. Molecular diagnosis of viral hepatitis. *Gastroenterology* 2002;122:1554-68.
 27. Weiss J, Wu H, Farrenkopf B, Schultz T, Song G, Shah S, *et al.* Real time TaqMan PCR detection and quantitation of HBV genotypes A-G with the use of an internal quantitation standard. *J Clin Virol* 2004;30:86-93.
 28. Kubista M, Andrade JM, Bengtsson M, Forootan A, Jonak J, Lind K, *et al.* The real-time polymerase chain reaction. *Mol Aspects Med* 2006;27:95-125.
 29. Guirgis BS, Abbas RO, Azzazy HM. Hepatitis B virus genotyping: Current methods and clinical implications. *Int J Infect Dis* 2010;14:e941-53.
 30. Landegren U. Ligation-based DNA diagnostics. *Bioessays* 1993;15:761-5.
 31. Landegren U, Kaiser R, Sanders J, Hood L. A ligase-mediated gene detection technique. *Science* 1998;241:1077-80.
 32. Lisby G. Application of nucleic acid amplification in clinical microbiology. *Mol Biotechnol* 1999;12:75-99.
 33. Craw P, Balachandran W. Isothermal nucleic acid amplification technologies for point-of-care diagnostics: A critical review. *Lab Chip* 2012;12:2469-86.
 34. Gill P, Ghaemi A. Nucleic acid isothermal amplification technologies: A review. *Nucleosides Nucleotides Nucleic Acids* 2008;27:224-43.
 35. Parida M, Sannarangaiah S, Dash PK, Rao PV, Morita K. Loop mediated isothermal amplification (LAMP): A new generation of innovative gene amplification technique; Perspectives in clinical diagnosis of infectious diseases. *Rev Med Virol* 2008;18:407-21.
 36. Notomi T, Okayama H, Masubuchi H, Yonekawa T, Watanabe K, Amino N, *et al.* Loop-mediated isothermal amplification of DNA. *Nucleic Acids Res* 2000;28:E63.
 37. Kanungo R. Ananthanarayan and Paniker's Textbook of Microbiology. 10th ed. India: Universities Press Pvt. Ltd.; 2013.
 38. Freed EO, Martin MA, editors. *Fields Virology*. 5th ed. USA: Lippincott Williams & Wilkins; 2007.
 39. Lampertico P. EASL 2017 clinical practice guidelines on the management of hepatitis B virus infection. *J Hepatol* 2017;67:370-98.

How to cite this Article: Maity SN, Kumar SS, Vijayaraghavan R, Polavarapu R. An update on Hepatitis B virus. *Int. Res. Med. Health Sci.*, 2018; 1(1):8-11.

Source of Support: Nil, **Conflict of Interest:** None declared.