The Role of New Viral Biomarkers in Chronic Hepatitis B: Ready to Use in the Clinical Practice?

Kronik Hepatit B'de Yeni Viral Göstergelerin Rolü: Klinik Uygulamada Kullanıma Hazır mı?

⊙ Suna YAPALI

Acıbadem University Faculty of Medicine, Department of Gastroenterology, Istanbul, Turkey

ABSTRACT

Recent advances have been made to develop and improve the serologic and molecular virologic tools for the diagnosis and optimal management of chronic hepatitis B (CHB) infection. Several biomarkers associated with the natural course of chronic hepatitis B infection virus (HBV) and the efficacy of antiviral treatment have been defined. This article reviews the impact of hepatitis B surface antigen, hepatitis B core-related antigen and HBV RNA on the natural history, treatment response and outcomes of CHB infection.
Keywords: Hepatitis B surface antigen, hepatitis B core-related antigen, hepatitis B virus RNA

ÖZ

Son yıllarda kronik hepatit B (KHB) enfeksiyonunun tanısı ve optimal yönetimi için serolojik ve moleküler virolojik araçlar geliştirilmiştir. Kronik hepatit B virüsünün (HBV) doğal seyri ve antiviral tedavinin etkinliği ile ilişkili birçok viral gösterge tanımlanmıştır. Bu makalede, hepatit B yüzey antijeni, hepatit B kor-ilişkili antijen ve HBV RNA'nın KHB enfeksiyonunun doğal seyri, tedavi cevabı ve sonuçları üzerine etkisi derlenmiştir.
Anahtar Kelimeler: Hepatit B yüzey antijeni, hepatit B kor-ilişkili antijen, hepatit B virus RNA

Introduction

Chronic hepatitis B (CHB) infection affects over 350 million people in the world and it is responsible for more than 500.000 deaths annually (1). Cirrhosis and hepatocellular carcinoma (HCC) are the major complications of CHB infection. With the availability of potent antiviral agents, management of CHB patients improved. However, it is still difficult to predict the risk of liver disease progression and the risk of HCC in CHB infection. Furthermore, the clearance of covalently closed circular DNA (cccDNA) from the host genome and eradication of the virus cannot be achieved with the current treatments. The treatment recommendations of the professional liver societies are mainly based on the clinical status, serum hepatitis B virus (HBV) DNA, alanine aminotransferase (ALT) level, hepatitis B e antigen (HBeAg) status and liver histology (if available). High serum HBV DNA level is an independent predictor of HCC and an important determinant for the decision of treatment (2). With the recent advances in the molecular investigations, the role of new biomarkers in the natural history and during the treatment of CHB has been increasingly recognized. This article reviews the role of new biomarkers during the natural course and treatment of CHB infection, with a special emphasis on hepatitis B surface antigen (HBsAg) and hepatitis B core-related antigen (HBcrAg) levels and HBV-RNA quantification.

Hepatitis B Surface Antigen Quantification

Qualitative HBsAg is an important marker in the detection of HBV infection. The majority of HBsAg is secreted by subviral particles and to a lesser extent by infectious virions (3). HBsAg reflects the content of intrahepatic HBV DNA, cccDNA transcription and host immune response to HBV infection (4,5) The kinetics of

HBsAg production are complex and show significant differences during the natural course of CHB and between HBV-genotypes. There is strong correlation of HBsAg with HBV replication only in the early phases of infection. The association between HBsAg and HBV DNA has been reported to be more prominent in HBV-genotype D, compared to HBV-genotype A. HBsAg levels were found to be related to HBV-reactivation in low viremic HBeAg-negative carriers (6). In a retrospective 8-year follow-up of HBeAg-negative CHB patients, HBsAg levels <100 IU/mL and HBV DNA <2.000 IU/mL were found to predict the inactive phase with a high chance of HBsAg loss. CHB patients with a HBV DNA level of 2.000-5.000 IU/mL who have an annual decline of HBsAg ≥0.5 log IU/mL have also been reported to have a high probability to become inactive carriers. HBsAg level has a good performance in identifying the phases of CHB during the natural course of the infection and can be helpful in determining the follow-up periods (7).

Serum HBsAg level correlates well with the cccDNA and intrahepatic HBV DNA and a is a strong predictor of response to pegylated-interferon (Peg-IFN) in HBeAg-positive patients. A reduction in HBsAg levels shows the induction of immune control. HBsAg level has been found to predict good response to Peg-IFN and lamivudine treatment better than HBV DNA (8). HBsAg level is an important predictor of response to Peg-IFN-α, however there may be genotypic differences (9,10). Quantification of HBsAg in HBeAg-positive patients may help in decision making regarding individualized treatment. In HBeAg-positive CHB patients, an HBsAg level of <1.500 IU/mL at week 12 predicts a high probability HBeAg seroconversion, while an HBsAg level of >20.000 IU/mL for HBV genotype B and C or no decline in HBsAg levels for HBV genotype A and D are associated with a very low rate of HBeAg seroconversion (9,10). The European Association for the Study of the Liver (EASL) recommends considering specific stopping rules according to HBsAg levels at week 12 of Peg-IFN treatment (11). In HBeAg-negative CHB patients, early serum HBsAg decline was found to be associated with sustained virological response in patients receiving Peg-IFN-α treatment (12). The combination of lack of decrease in HBsAg level and <2 log IU/mL decline at the 12th week of Peg-IFN treatment has 100% negative predictive value for treatment response and discontinuation of treatment is recommended by the EASL guideline (10).

Compared to Peg-IFN treatment, HBsAg decline during nucleoside/tide analogs (NA) therapy is much slower. The immune modulation by IFN results in a more rapid HBsAg decline. On the other hand, NAs inhibit only the reverse transcription of the pregenomic RNA but do not target the cccDNA directly and have a less pronounced effect on HBsAg secretory pathways (13,14). Furthermore, integrated HBV DNA in HBeAg-negative patients leads to production of HBsAg.

HBsAg levels in HBeAg-negative patients may be helpful to predict in which patient treatment can be stopped. A study by Hadziyannis et al. (15) showed that discontinuation of antiviral therapy led to sustained off-treatment HBV DNA suppression in 55% of patients and subsequent HBsAg loss in 39% of HBeAg-negative CHB patients who received adefovir treatment for 4-5 years. Lower HBsAg levels at the end of treatment were predictive for later HBsAg loss.

In a systematic review of 22 studies, an HBsAg level of <100 or <200 IU/mL at the end of therapy has been suggested to be a good indicator of sustained response (16). HBsAg levels have promising evidence for predicting in which patients treatment can be stopped, however, more data are needed to confirm the thresholds for discontinuation of treatment.

Hepatitis B Virus Core-related Antigen

HBcrAg is a novel biomarker of CHB infection, which measures serum levels of HBcAg, HBeAg and the empty particle (p22) (17). Quantification of HBcrAg is first developed in Japan by comparing the chemiluminescence signals generated by known concentration of recombinant pro-HBeAg (18). This assay (Fujirebio, Tokyo, Japan) is currently available with a lower limit of detection of 2.0 log U/mL, and a linear range of 3.0 log U/mL-7.0 log U/mL.

Serum HBcrAg level was found to correlate with serum HBV DNA, intrahepatic DNA and cccDNA levels (17). HBcrAg levels may show variation during the natural course of CHB infection. Studies in the European and Asian cohorts showed that HBcrAg levels were higher in HBeAg-positive CHB patients in comparison to HBeAg-negative patients (19,20). In an Asian cohort, it was found that HBcrAg levels were 8.54 log U/mL, and 7.92 log U/mL in the immune tolerant phase and in the immune clearance phase, respectively. Lower HBcrAg levels suggested a better immune control in HBeAg-positive patients. Among HBeAg-negative patients, HBcrAg levels were 2.60 log U/mL, and 4.92 log U/mL in inactive carriers and CHB patients, respectively. HBcrAg levels in HBeAg-negative patients with CHB showed a positive correlation with necroinflammatory activity and fibrosis (19). After HBsAg seroclearance, HBcrAg levels were found to be undetectable in 79% of patients, while 21% had median HBcrAg 2.7 log U/mL (19). HBcrAg levels were found to be associated with development of HCC in both treated and untreated patients (21,22). In a large cohort of 1031 CHB patients with a median 10-year follow-up, HBcrAg >2.9 log U/mL (HR, 5.05; 95% confidence interval, 2.40-10.63) was associated with an increased incidence of HCC (22).

HBcrAg may have an important role in predicting the clinical outcomes in patients on treatment. In their study, Wong et al. (23) reported that HBcrAg was detected in 101 (78%) of 130 samples with undetectable HBV DNA collected from NA-treated patients and there was a positive correlation between HBcrAg and cccDNA. As HBV DNA is undetectable in the majority of patients on treatment, HBcrAg levels may be considered as an indirect marker of cccDNA. The decline in HBcrAg has been found to be slower than in HBV DNA levels during NA treatment (24). The rationale is; NAs inhibit HBV DNA replication by the action on the reverse transcription but without elimination of cccDNA from the infected hepatocyte. In some Asian studies, the decline of HBcrAg was found to correlate with the decline of intrahepatic cccDNA in patients receiving entecavir treatment (23,25,26).

In a study of 46 patients treated with Peg-IFN, a baseline HBcrAg level of >8 log U/mL was reported to have low probability of HBeAg seroconversion (>94.4% negative predictive value) and suppression of HBV DNA at 12 weeks (27). Furthermore, changes in HBcrAg levels may be helpful in predicting clinical outcomes during treatment. In a study of 58 HBeAg-positive CHB patients treated with Peg-IFN, the HBcrAg at week 12 of therapy was

found to be predictive of HBeAg seroconversion at 24 weeks after completion of therapy (28).

The role of HBcrAg for predicting HBsAg loss was examined in a study of 62 HBeAg-negative CHB patients receiving Peg-IFN alone or in combination with tenofovir, the chance of HBsAg loss was higher in patients with baseline HBcrAg <3.7 log U/mL (29).

The majority of HBeAg-negative CHB patients remain on treatment until HBsAg loss is achieved. There are many attempts to determine the predictors of relapse after discontinuation of treatment. The decision for discontinuation of treatment can be determined by viral serological markers, serum HBV DNA, ALT and more recently, HBsAg levels. HBcrAg levels may provide prognostic information for the decision of treatment discontinuation. In studies performed in Japan, a high end-of-treatment HBcrAg level was shown to predict relapse after discontinuation of NA therapy (30,31,32,33). In a study of 113 patients receiving NA treatment (lamivudin: 32, entecavir: 81), end-of-treatment HBcrAg level >3.7 log IU/mL predicted virological relapse within 1 year of NA discontinuation (33). The Japanese Society of Hepatology guidelines have recently recommended the use of an HBsAg level of <1.9 log U/mL and an HBcrAg level of <3.0 log U/mL to identify patients at low risk of relapse (34). However, there is a need to examine the performance of these criteria in different populations.

Recently, HBcrAg positivity has been found to be a significant risk factor for HBV reactivation in HBsAg-negative, anti-HBc-positive patients undergoing high-risk immunosuppressive therapy (especially rituximab-containing chemotherapy), and to have potential to identify patients who would most benefit from prophylactic antiviral treatment (35).

The use of HBcrAg will most probably lead to progress in the management of CHB. However, most of the studies are performed in Japan and Asian countries, and these data need to be confirmed in the Western populations.

Hepatitis B Virus RNA Quantification

During HBV replication, pregenomic RNAs are encapsided into HBV core particles in the cytoplasm, and all pregenomic RNAs are reverse transcribed into plus stranded genomic DNA in the core particle (36). HBV RNA has also been detected in the serum of CHB patients and found to strongly correlate with HBV DNA levels (36). Studies have reported decreased HBV RNA levels in patients receiving NA and the combination of Peg-IFN and NA (37,38). The decrease in HBV RNA levels was predictive for HBeAg seroconversion suggesting a better role of HBV RNA ± HBV DNA for treatment monitoring (37,38).

Conclusion

HBsAg, HBcrAg and HBV RNA quantification provides new perspectives in understanding HBV virology and immunopathogenesis. The implementation of HBsAg quantification has already been integrated into European treatment guidelines. The use of HBcrAg seems to be in the pipeline, however, the results need to be validated in other populations with different ethnic and genotypic characteristics. There is a growing body of evidence for the impact of HBV RNA in patients receiving treatment. In future, the use of new biomarkers may guide the clinicians to predict the natural course, treatment response and the outcome of CHB, adapting individualized treatment approaches.

Ethics

Peer-review: Internally peer-reviewed.

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