High Levels of Hepatitis B Surface Antigen Increase Risk of Hepatocellular Carcinoma in Patients With Low HBV Load

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CLINICAL LIVER

This article has an accompanying continuing medical education activity on page e13. Learning Objective: Upon completion of this assessment, successful learners will be able to use HBsAg level to define different HCC risk in HBV carriers with low viral load.

See editorial on page 1057; see Covering the Cover synopsis on page 1048.

BACKGROUND & AIMS: Patients with chronic hepatitis B virus (HBV) infection have a high risk for developing hepatocellular carcinoma (HCC). Patients with lower levels of hepatitis B surface antigen (HBsAg) have higher chances of losing HBsAg than those with high levels. However, little is known about whether higher levels of HBsAg increase risk for HCC. **METHODS:** We followed 2688 Taiwanese HBsAg-positive patients without evidence of cirrhosis for a mean time period of 14.7 years. In addition to the known risk factors of HCC, we investigated the association between levels of HBsAg and development of HCC. **RESULTS:** Of the patients followed, 191 developed HCC, with an average annual incidence rate of 0.5%. Baseline levels of HBsAg and HBV were associated with development of HCC, and risk increased with level. Compared to HBsAg level, by receiver operating characteristic curve analysis, HBV DNA level better predicted the development of HCC during 10-year and 15-year periods (both, $P < .001$). However, when we evaluated hepatitis B e antigen−negative patients with levels of HBV DNA <2000 IU/mL, factors that determined HCC risk included sex, age, and levels of alanine aminotransferase and HBsAg (≥1000 IU/mL), but not level of HBV DNA. Multivariate analysis showed that the adjusted hazard ratio for HCC in patients with levels of HBsAg ≥1000 IU/mL versus <1000 IU/mL was 13.7 (95% confidence interval: 4.8−39.3). **CONCLUSIONS: Among patients infected with HBV genotype B or C, determinants of HCC risk include their sex, age, hepatitis B e antigen status, HBV genotype, and levels of alanine aminotransferase and HBV DNA, but not level of HBsAg. Among hepatitis B e antigen−negative patients with low viral loads, HCC risk is determined by levels of HBsAg and alanine aminotransferase and age, but not HBV DNA.**

Keywords: Chronic Hepatitis B; Liver Disease; Virology.

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Hepatitis B virus (HBV) infection is a global health problem resulting in >1 million deaths per year.[1] Patients with chronic HBV infection are at risk of developing cirrhosis, hepatic decompensation, and hepatocellular carcinoma (HCC), with an estimated lifetime risk of 25%−40% in carriers who acquire the virus early in life.[1–4]

The REVEAL-HBV (Risk Evaluation of Viral Load Elevation and Associated Liver Disease/Cancer-Hepatitis B Virus) study from Taiwan indicated that HBV DNA is the major driver of disease progression in patients with chronic HBV infection.[5–7] In particular, patients with serum HBV DNA levels ≥2000 IU/mL at study entry have an increased risk of developing HCC.[5] In contrast, those with HBV DNA levels <2000 IU/mL are usually designated inactive or low-risk HBV carriers.[3,8,9] However, data from longitudinal studies indicated that these subjects still carry an annual incidence rate of 0.06% for HCC development.[5,10] Therefore, identification of factors predictive of HCC other than viral load in these low-risk patients remains mandatory and deserves additional studies.

Recently, hepatitis B surface antigen (HBsAg) quantification has become increasingly recognized as a marker for evaluating

Abbreviations used in this paper: ALT, alanine aminotransferase; CI, confidence interval; HBsAg, hepatitis B surface antigen; HBV, hepatitis B virus; HBeAg, hepatitis B e antigen; HCC, hepatocellular carcinoma; HCV, hepatitis C virus; HR, hazard ratio; ROC, receiver operating characteristic; SD, standard deviation.

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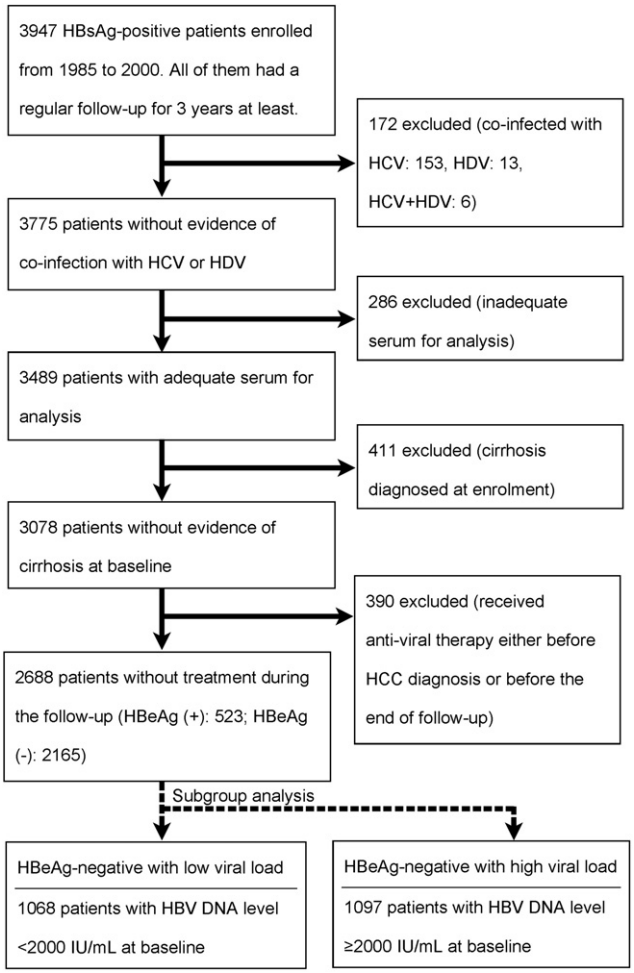

Figure 1. Flow of study participants.

viral replication and possible host immune control over HBV infection.[11–17] A lower HBsAg level is shown to be associated with a higher chance of HBsAg loss and lower risk of hepatitis activity in patients with HBV genotype B or C infection.[11,17,18] In addition, HBsAg level <1000 IU/mL was found to convincingly define inactive carrier state in Italian patients with HBV genotype D infection.[16] Because a lower HBsAg level usually signifies a better prognosis, it is of clinical interest to know whether a higher HBsAg level would be associated with a higher risk of HCC, especially in the special population of lowly viremic patients.

To address this interesting and important issue, we enrolled a large cohort of 2688 treatment-naïve patients who were diagnosed with chronic HBV infection and received long-term follow-up at the National Taiwan University Hospital. The primary aim of our study was to explore whether HBsAg level could complement HBV DNA level as a predictor of HCC development.

Materials and Methods

Patient Cohort

Figure 1 shows the inclusion and exclusion process of patients in the Elucidation of Risk Factors for Disease Control or Advancement in Taiwanese Hepatitis B Carriers (ERADICATE-B) study. Part of this cohort (patients enrolled from 1985 to 1995) had been used to investigate the issue of HBsAg loss.[17] The enrollment time frame was extended to 2000 in this study. In total, 3947 HBsAg-positive patients aged older than 28 years were consecutively enrolled between 1985 and 2000. All of them had been HBsAg-positive for longer than 6 months and received more than 3 years of regular follow-up at the National Taiwan University Hospital. After excluding patients with evidence of hepatitis C virus (HCV) or hepatitis D virus co-infection, and those without adequate serum samples for analysis, 3489 patients remained. We further excluded 411 patients who were diagnosed with cirrhosis at baseline because this is an indication for antiviral therapy in practice guidelines,[19–21] and 390 patients who received antiviral therapy either before HCC diagnosis or before the end of follow-up because of the possible modification of HCC risk by treatment.[22] Finally, a total of 2688 HBV carriers were included into analysis. A subgroup analysis was also performed on hepatitis B e antigen (HBeAg)–positive patients (n = 523) and HBeAg-negative patients (n = 2165), who were divided into high viral load group (1097 with HBV DNA level ≥2000 IU/mL) and low viral load group (1068 with HBV DNA <2000 IU/mL). All enrolled patients gave informed consent as approved by the National Taiwan University Hospital Ethical Committee.

Data Collection

Patients were tested for serological markers (HBsAg, HBeAg, anti-HBe, antibodies against hepatitis C virus [anti-HCV], and antibodies against hepatitis D virus), and had liver function tests and α-fetoprotein levels at baseline. Throughout the follow-up period, if alanine aminotransferase (ALT) levels were within normal limits, liver function tests and α-fetoprotein were assayed every 6 months, and at least every 3 months if ALT levels were elevated. Serum samples collected at each visit were stored at −20°C until analysis. Serum α-fetoprotein and abdominal ultrasonography using a high-resolution and real-time scanner were performed for HCC surveillance every 3 to 6 months from enrollment.

Diagnosis of Cirrhosis and HCC

Cirrhosis was diagnosed by histology or ultrasonographic findings, together with clinical features such as thrombocytopenia, gastroesophageal varices, or ascites.[23] For the diagnosis of cirrhosis made via abdominal ultrasound, the findings had to be consistent on at least 2 occasions 6 months apart.[6] HCC was diagnosed either by histology/cytology or by typical image findings (arterial enhancement and venous wash-out by contrast-enhanced computed tomography or magnetic resonance imaging scanning) in hepatic nodules >1 cm.[24]

Serological Assays

Serum HBsAg, HBeAg, anti-HBe, anti-HCV, and anti–hepatitis D virus were tested by commercial assays (Abbott Laboratories, Abbott Park, IL).

Quantification of HBV DNA and HBsAg Levels

Serum samples at enrollment were tested for both HBV DNA and HBsAg levels. HBV DNA level was quantified using the Abbott RealTime HBV assay, 0.2 mL protocol (Abbott Laboratories) with a low detection limit of 15 IU/mL. HBsAg level was quantified using the Architect HBsAg QT (Abbott Laboratories) according to manufacturer's instructions.[11,15] The detection range of Architect assay is 0.05 to 250 IU/mL. If the HBsAg level