Impact of HBeAg on Hepatocellular Carcinoma Risk During Oral Antiviral Treatment in Patients With Chronic Hepatitis B

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Abbreviations used in this paper: ALT, alanine aminotransferase; CHB, chronic hepatitis B; CT, computed tomography; FiB-4, Fibrosis-4; HBeAg, hepatitis B envelope antigen; HBsAg, hepatitis B surface antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio; IPTW, inverse probability of treatment weighting; IQR, interquartile range; LC, liver cirrhosis; NA, nucleos(t)ide analog; PSM, propensity score matching.

1542-3565/$36.00
https://doi.org/10.1016/j.cgh.2021.09.001

BACKGROUND & AIMS: **Antiviral treatment from hepatitis B envelope antigen (HBeAg)-positive status may attenuate the integration of hepatitis B virus DNA into the host genome causing hepatocellular carcinoma (HCC). We investigated the impact of HBeAg status at the onset of antiviral treatment on the risk of HCC.**

METHODS: **The incidence of HCC was evaluated in Korean patients with chronic hepatitis B who started entecavir or tenofovir in either HBeAg-positive or HBeAg-negative phase. The results in the Korean cohort were validated in a Caucasian PAGE-B cohort.**

RESULTS: **A total of 9143 Korean patients (mean age, 49.2 years) were included: 49.1% were HBeAg-positive and 49.2% had cirrhosis. During follow-up (median, 5.1 years), 916 patients (10.0%) developed HCC. Baseline HBeAg positivity was not associated with the risk of HCC in the entire cohort or cirrhotic subcohort. However, in the non-cirrhotic subcohort, HBeAg positivity was independently associated with a lower risk of HCC in multivariable (adjusted hazard ratio [aHR], 0.41; 95% confidence interval [CI], 0.26–0.66), propensity score-matching (aHR, 0.46; 95% CI, 0.28–0.76), and inverse probability weighting analyses (aHR, 0.44; 95% CI, 0.28–0.70). In the Caucasian cohort (n = 719; mean age, 51.8 years; HBeAg-positive, 20.3%; cirrhosis, 34.8%), HBeAg-positivity was not associated with the risk of HCC either in the entire cohort or cirrhotic subcohort. In the non-cirrhotic subcohort, none of the HBeAg-positive group developed HCC, although the difference failed to reach statistical significance (aHR, 0.21; 95% CI, 0.00–1.67).**

CONCLUSIONS: **This multinational cohort study implies that HBeAg positivity at the onset of antiviral treatment seems to be an independent factor associated with a lower risk of HCC in patients with chronic hepatitis B without cirrhosis, but not in those with cirrhosis.**

Keywords: Cumulative Incidence; DNA; Hepatitis B Virus; Liver Cancer; Neoplasm.

Hepatocellular carcinoma (HCC) is a highly lethal malignancy, and it is the second leading cause of cancer-related mortality worldwide.[1] Chronic hepatitis B (CHB) is a major cause of HCC, especially in East Asia.[2] Effective antiviral treatment with nucleos(t)ide analogues (NAs) is reported to reduce the relative risk of HCC by 45% to 63%.[3,4] However, NA treatment is not able to prevent HCC occurrence completely, so HCC risk prediction is important for optimizing cost-effective surveillance for patients with CHB.

Recently, several studies provided important evidence that antiviral treatment from the early phase of CHB (ie, hepatitis B envelope antigen [HBeAg]-positive status) might attenuate the HCC risk. Mason et al reported that hepatitis B virus (HBV) DNA integration into the host genome and clonal hepatocyte expansion, which is a key mechanism of hepatocarcinogenesis,[5–7] starts in the HBeAg-positive chronic HBV infection phase (previously termed 'immune-tolerant' phase).[8] In addition, a recent study of the effect of RNA interference with ARC-520 on hepatitis B surface antigen (HBsAg) in chimpanzees chronically infected with HBV revealed that the integration of HBV DNA into the host genome occurs during early phase of HBeAg-positive status.[9] Molecular integration events occurring during the HBeAg-positive phase and the subsequent HBeAg seroclearance-associated hepatocyte turnover may predispose to the development of HCC. Thus, it can be postulated that NA treatment starting in the HBeAg-positive phase potentially before HBV DNA-host genome integration might lower the incidence of HCC, possibly by reducing HBV DNA integration and clonal hepatocyte expansion as well as by decreasing liver inflammation.

Previous studies reported that HBeAg status at the onset of NA treatment was not an independent predictor of HCC development.[10–13] However, those studies included relatively small numbers of patients (less than 1700 patients per study) with low proportions of HBeAg-positivity (16%–36%), so it might have been difficult to adequately adjust for all potential confounders. Because the HBeAg-positive phase occurs early in the natural course of chronic HBV infection, HBeAg-positive patients compared with HBeAg-negative patients usually have characteristics associated with lower HCC risk, such as younger age and milder stage of fibrosis.[14] On the other hand, HBeAg-positive patients have higher HBV DNA levels, which can affect the HCC risk as well.[15,16] Thus, the clinical impact of HBeAg-positivity on the probability of HCC development should be investigated after adjusting for those critical confounding factors.

Therefore, we conducted a large, nationwide, multicenter cohort study in Korea to investigate the impact of HBeAg status at the onset of antiviral treatment on the risk of HCC in patients with CHB after rigorously adjusting and balancing for confounding factors including age, hepatic fibrosis, and serum HBV DNA levels. In addition, we compared our results with those obtained in a Caucasian cohort treated with NAs.

Methods

Patients

Among a Korean nationwide multicenter cohort of consecutive patients with CHB, patients older than 19 years who started NAs of high genetic barrier (ie, entecavir or tenofovir disoproxil fumarate) as initial antiviral treatment for more than 6 months at 16 university-affiliated hospitals between January 2007 and December 2018 (Supplementary Table 1) were eligible for this study. A total of 9143 patients with CHB who underwent NA treatment according to the American Association for the Study of Liver Diseases guidelines (Supplementary Methods)[17] were selected for the study (Supplementary Figure 1).

Another independent dataset was included from the extended follow-up study of the Caucasian PAGE-B cohort,[18] fulfilling the same inclusion/exclusion criteria. Among 1951 Caucasian patients with CHB who had received NA therapy for more than 1 year, 719 fulfilled the criteria of our study and were included as a validation cohort.

Outcomes and Assessment

The primary outcome was the development of HCC. The date of starting NA treatment was defined as the index date, and the follow-up duration of our study was ended at the date of HCC diagnosis. Patients who were lost to follow-up without HCC development were censored at the date of last visits. Fibrosis-4 (FIB-4) score was calculated to assess the grade of hepatic fibrosis at the index date (Supplementary Methods).[19]

All patients underwent regular surveillance for HCC with liver ultrasonography and serum alpha-fetoprotein measurement at the index date and every 3 to 6 months thereafter, regardless of the presence or absence of liver cirrhosis (LC). The surveillance was repeated until the date of HCC detection, death, or last follow-up (Supplementary Methods).

Statistical Analyses

Baseline characteristics were described appropriately (Supplementary Methods). The cumulative incidence rates of HCC according to HBeAg status were derived using the Kaplan-Meier method, and the log-rank test was used for the comparison. The Cox proportional hazards analysis was performed to identify predictors of HCC occurrence. Significant variables in the univariable analyses were included in the multivariable models. Schoenfeld residual tests identified that all multivariable models did not violate the proportional hazards assumption. To minimize the potential bias according to the different baseline characteristics between HBeAg-positive and HBeAg-negative patients, we performed propensity score matching (PSM) and inverse probability of treatment weighting (IPTW) analyses (Supplementary Methods). All statistical tests conducted were 2-sided. The P value was considered statistically significant when it was $< .05$. R version 3.6.3 (http://www.r-project.org/) was used for all statistical analyses.

What You Need to Know

Background

The clinical impact of hepatitis B virus envelope antigen (HBeAg) status at the onset of nucleos(t)ide analogue treatment on the risk of hepatocellular carcinoma (HCC) is unclear in patients with chronic hepatitis B (CHB).

Findings

The current study showed that HBeAg-positivity was significantly and independently associated with a lower risk of HCC development in patients with CHB without liver cirrhosis (LC), but not in patients with LC.

Implications for patient care

Our findings support the need for regular monitoring of patients in the HBeAg-positive CHB virus infection phase (previously termed 'immune tolerant') for timely onset of nucleos(t)ide analogue treatment before the development of LC to reduce HCC risk.

Results

Baseline Characteristics

Among the 9143 patients of the entire Korean cohort (mean age, 49.2 years; male, 60.3%), 4651 (50.9%) were HBeAg-negative (HBeAg-negative group) and 4492 (49.1%) were HBeAg-positive (HBeAg-positive group) at baseline (Table 1). As expected, the HBeAg-positive group had younger age, lower prevalence of LC, higher serum HBV DNA levels, and milder severity of hepatic fibrosis according to FIB-4 scores (all $P < .001$). Because LC is the strongest predictor of HCC,[20] we divided the patients according to presence (49.2%) or absence (50.8%) of LC in order to avoid the influence of LC on the HCC risk (Supplementary Table 2). In both the non-LC and LC subcohorts, the HBeAg-positive group showed younger age, higher HBV DNA levels, and lower FIB-4 scores (all $P < .001$) (Supplementary Table 3).

Among the 719 patients of the Caucasian cohort (mean age, 51.8 years; male, 70.0%), 573 (79.7%) were HBeAg-negative and 146 (20.3%) were HBeAg-positive at baseline. Approximately one-third of the patients (34.8%) had LC. The HBeAg-positive group had younger

Table 1. Baseline Characteristics of the Korean Cohort

Variables	Entire cohort (n = 9143)	HBeAg status		P value
		HBeAg-positive (n = 4492)	HBeAg-negative (n = 4651)	
Type of NAs				< .001
Entecavir	4895 (53.5)	2296 (51.1)	2599 (55.9)	
Tenofovir	4248 (46.5)	2196 (48.9)	2052 (44.4)	
Male sex	5510 (60.3)	2680 (59.7)	2830 (60.8)	.26
Age, *y*	49.2 ± 11.4	47.1 ± 11.9	51.2 ± 10.5	< .001
Liver cirrhosis	4499 (49.2)	1847 (41.1)	2652 (57.0)	< .001
Platelet, $\times 1000/mm^3$	153 (113-197)	160 (117-207)	147 (109-187.5)	< .001
Albumin, *g/dL*	4.1 (3.8-4.4)	4.1 (3.7-4.3)	4.1 (3.8-4.4)	< .001
Total bilirubin, *mg/dL*	0.9 (0.7-1.3)	0.9 (0.7-1.3)	0.9 (0.7-1.3)	.04
ALT, *U/L*	96 (53-184)	102 (60-200)	91 (49-171)	< .001
HBV DNA, log_{10} *IU/mL*	6.4 (5.3-7.7)	7.2 (6.0-8.2)	5.9 (4.8-6.8)	< .001
FIB-4 score	2.7 (1.7-4.5)	2.5 (1.5-4.5)	2.8 (1.8-4.5)	< .001

Note: Values are expressed as frequency (%), mean ± standard deviation, or median (interquartile range).
ALT, Alanine aminotransferase; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; NAs, nucleos(t)ide analogues.

age and higher serum HBV DNA levels than the HBeAg-negative group, which was consistent with the findings of the Korean cohort (all $P < .001$) (Supplementary Table 4).

HBeAg Status and HCC Risk in the Entire Korean Cohort

During a median follow-up of 5.1 years (interquartile range [IQR], 3.1–6.8 years), 916 patients (10.0%) developed HCC. The cumulative incidence rates of HCC at years 2, 5, and 8 were 2.7%, 8.1%, and 13.4%, respectively, in the HBeAg-positive group, and 3.9%, 11.0%, and 16.1%, respectively, in the HBeAg-negative group (log-rank $P < .001$) (Figure 1A). HBeAg-positivity was associated with a lower risk of HCC in the univariable analysis, but this association disappeared in the multivariable analysis (adjusted HR [aHR], 1.01; 95% confidence interval [CI], 0.87–1.16; $P = .94$) (Table 2). In both PSM and IPTW analyses (Supplementary Results), HBeAg-positivity was also not associated with HCC risk (PSM: aHR, 1.10; 95% CI, 0.94–1.29 [Figure 2A and Supplementary Table 5]; IPTW: aHR, 1.03; 95% CI, 0.89–1.19 [Figure 3A and Supplementary Table 6]).

LC, the single most important risk factor for HCC,[20] had a statistically significant interaction with HBeAg status on the HCC risk (P for interaction $< .001$). Therefore, we stratified our patients according to the presence of LC and separately analyzed the impact of HBeAg status on HCC risk in both the non-LC and LC subcohorts.

HBeAg Status and HCC Risk in the Non-LC Subcohort of the Korean Cohort

During 5.1 years (IQR, 3.3–6.9 years) of median follow-up for the 4644 Korean patients of the non-LC subcohort, the 2-, 5-, and 8-year cumulative incidence HCC rates were 0.2%, 0.7%, and 2.1%, respectively, in the HBeAg-positive group, and 1.0%, 3.5%, and 5.4%, respectively, in the HBeAg-negative group (log-rank $P < .001$) (Figure 1B). The HBeAg-positive group was associated with a lower risk of HCC in the univariable analysis of the non-LC subcohort (HR, 0.33; 95% CI, 0.22–0.50; $P < .001$) (Table 3). Furthermore, the HBeAg-positive group was independently associated with a significantly lower risk of HCC (aHR, 0.41; 95% CI, 0.29–0.66; $P < .001$) after adjusting for significant confounding variables (Table 3). Even after considering death as a competing risk for HCC development, the HBeAg-positive group was independently associated with a significantly lower risk of HCC (adjusted subhazard ratio, 0.43; 95% CI, 0.27–0.68; $P < .001$) (Supplementary Table 10).

We additionally evaluated whether this result was reproducible after PSM and IPTW adjustments (Supplementary Results). On PSM analysis, the HBeAg-positive group was negatively associated with HCC development both in univariable (HR, 0.49; 95% CI, 0.30–0.81; stratified log-rank $P = .01$) (Figure 2B) and multivariable analyses (aHR, 0.46; 95% CI, 0.28–0.76; $P = .003$) (Supplementary Table 7). In the IPTW analysis, the HBeAg-positive group was also associated with a lower risk of HCC both in univariable (HR, 0.47; 95% CI, 0.30–0.73; weighted log-rank $P < .001$) (Figure 3B) and

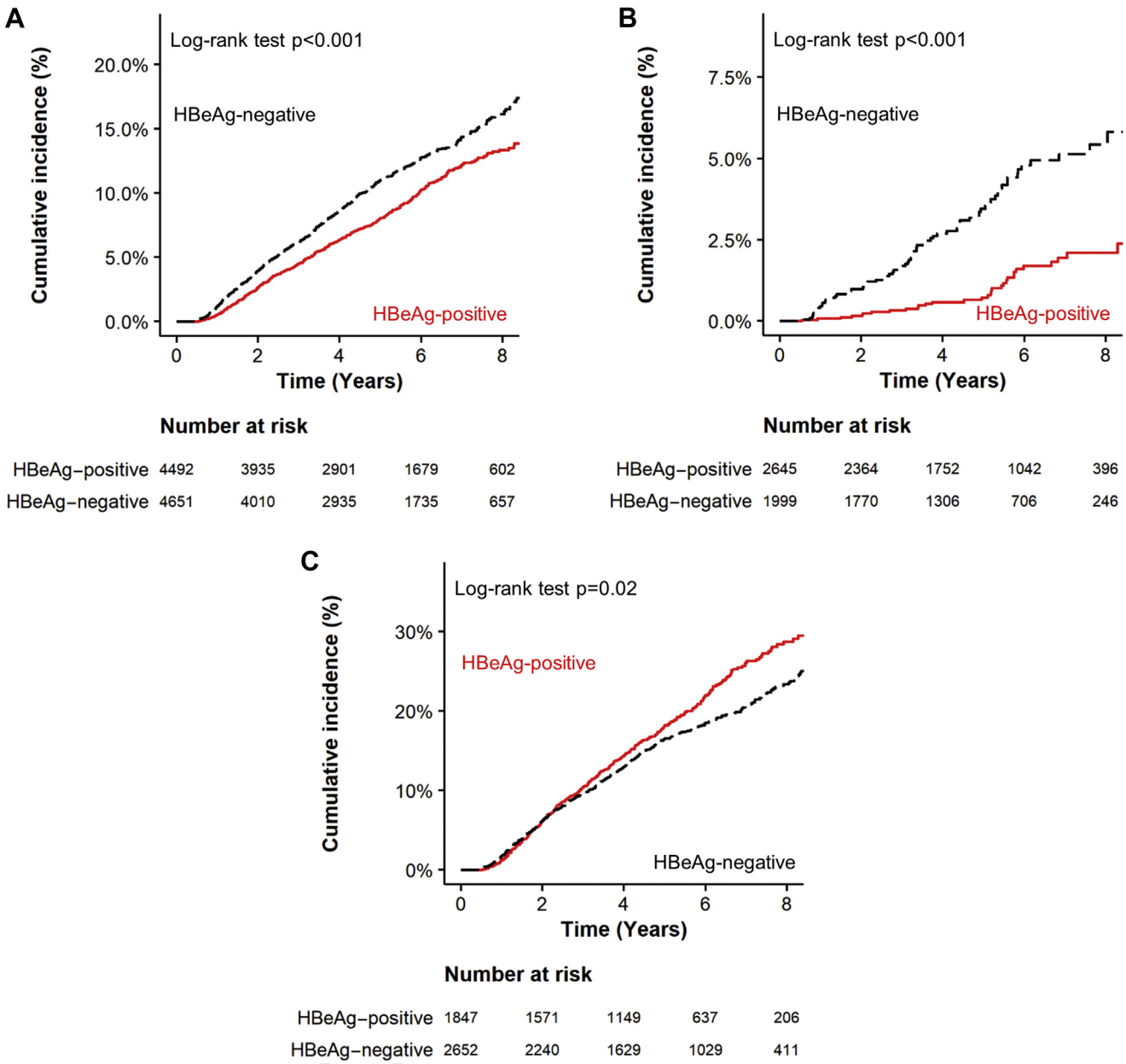

Figure 1. Cumulative incidence of hepatocellular carcinoma according to HBeAg status in the Korean cohort. Kaplan-Meier curves of the entire cohort (*A*), the non-LC subcohort (*B*), and the LC subcohort (*C*). The log-rank test was used for the comparison between the HBeAg-positive and HBeAg-negative groups.

multivariable analyses (aHR, 0.44; 95% CI, 0.28–0.70; $P < .001$) (Supplementary Table 8).

To minimize the impact of HBV DNA level as a confounder on the association between HBeAg status and HCC risk, we performed additional PSM analyses (Supplementary Results). The HBeAg-positive group was also independently associated with a significantly lower risk of HCC (aHR, 0.56; 95% CI, 0.33–0.94; $P = .03$) in the matched population (Supplementary Table 9). All subgroup analyses in the non-LC subcohort, including those stratified according to the HBV DNA levels, consistently showed that the HBeAg-positive group had a lower risk of HCC (Supplementary Figure 3).

HBeAg Status and HCC Risk in the LC Subcohort of the Korean Cohort

During 5.0 years (IQR, 2.9–6.8 years) of median follow-up for the 4499 patients of the LC subcohort, the 2-, 5-, and 8-year cumulative incidence HCC rates were 6.2%, 18.2%, and 28.7%, respectively, in the HBeAg-positive group, and 6.2%, 16.5%, and 23.4%, respectively, in the HBeAg-negative group (log-rank $P = .02$) (Figure 1C). Although the HBeAg-positive group was associated with a higher risk of HCC in the univariable analysis (HR, 1.17; 95% CI, 1.02–1.35; $P = .02$), it was not an independent predictor of HCC in the multivariable

Table 2. The Risk of HCC Development in the Korean Cohort

Variables	Univariable analysis		Multivariable analysis	
	HR (95% CI)	*P* value	HR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg-positive	0.76 (0.67-0.87)	< .001	1.01 (0.87-1.16)	.94
Sex				
Females	1 [Reference]		1 [Reference]	
Males	1.83 (1.58-2.12)	< .001	2.41 (2.07-2.80)	< .001
Age, *y*	1.06 (1.06-1.07)	< .001	1.04 (1.03-1.05)	< .001
Liver cirrhosis	8.17 (6.69-9.99)	< .001	3.86 (3.09-4.83)	< .001
Platelet,[a] $\times 1000/mm^3$	0.988 (0.986-0.989)	< .001		
Albumin, *g/dL*	0.47 (0.43-0.52)	< .001	0.65 (0.58-0.72)	< .001
Total bilirubin, *mg/dL*	1.01 (1.00-1.03)	.14		
ALT, *U/L*	0.994 (0.993-0.995)	< .001	0.998 (0.997-0.999)	< .001
HBV DNA, log_{10} *IU/mL*	0.79 (0.76-0.82)	< .001	0.95 (0.91-0.99)	.02
FIB-4 score[a]	1.04 (1.04-1.05)	< .001	1.02 (1.01-1.03)	< .001

Note: The HR and *P* value were estimated using Cox proportional hazards regression.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, Fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (Spearman's rho, −0.74; $P < .001$), only FIB-4 score was included in the multivariable analysis.

analysis (aHR, 1.09; 95% CI, 0.95–1.26; $P = .22$) (Supplementary Table 11). In both PSM and IPTW analyses (Supplementary Results), HBeAg-positivity was also not associated with HCC risk (PSM: aHR, 1.09; 95% CI, 0.93–1.28 [Figure 2C and Supplementary Table 12]; IPTW: aHR, 1.13; 95% CI, 0.98–1.32 [Figure 3C and Supplementary Table 13]).

HBeAg Status and HCC Risk in the Caucasian Cohort

During a median 7.6 years (IQR, 4.4–9.5 years) of follow-up for the 719 patients of the Caucasian PAGE-B cohort,[18] 40 patients (5.6%) developed HCC. The risk of HCC of the HBeAg-positive group was not significantly different from that of the HBeAg-negative group (aHR, 0.74; 95% CI, 0.25–2.15) (Supplementary Figure 4A) in the entire population. Similar findings were observed both in PSM (stratified log-rank $P = .32$) (Supplementary Figure 5A) and IPTW analyses (weighted log-rank $P = .79$) (Supplementary Figure 6A).

Within the non-LC patients of the Caucasian cohort, the risk of HCC of the HBeAg-positive group was also not statistically significantly different from that of the HBeAg-negative group (aHR, 0.21; 95% CI, 0.00–1.67; $P = .17$; log-rank $P = .1$) (Supplementary Figure 4B). However, interestingly, none of the patients in the HBeAg-positive group developed HCC during the study period. After PSM analysis, the risk of HCC of the HBeAg-positive group was significantly lower than that of the HBeAg-negative group (stratified log-rank $P = .046$) (Supplementary Figure 5B). However, the significant difference was not reproduced after IPTW matching (weighted log-rank $P = .26$) (Supplementary Figure 6B).

Within the patients with LC in the Caucasian cohort, the risk of HCC of the HBeAg-positive group was not significantly different from that of the HBeAg-negative group (aHR, 0.87; 95% CI, 0.30–2.53; $P = .79$) (Supplementary Figure 4C). The result within the patients with LC was also maintained both in PSM (stratified log-rank $P > .99$) (Supplementary Figure 5C) and IPTW analyses (weighted log-rank $P = .77$) (Supplementary Figure 6C).

Discussion

The main finding of our study is that HBeAg status was not associated with the risk of HCC in patients within the entire CHB cohort or the LC subcohort. For patients in the non-LC subcohort, however, the HBeAg-positive group showed a significantly lower risk of HCC compared with the HBeAg-negative group after adjusting for confounding factors. The expected risk reduction in the HBeAg-positive group was approximately 59% compared with the HBeAg-negative group. These results were reproducible in both the PSM and IPTW models. HBeAg positivity at the onset of antiviral treatment showed a minimal risk of HCC in Caucasian patients without LC as well.

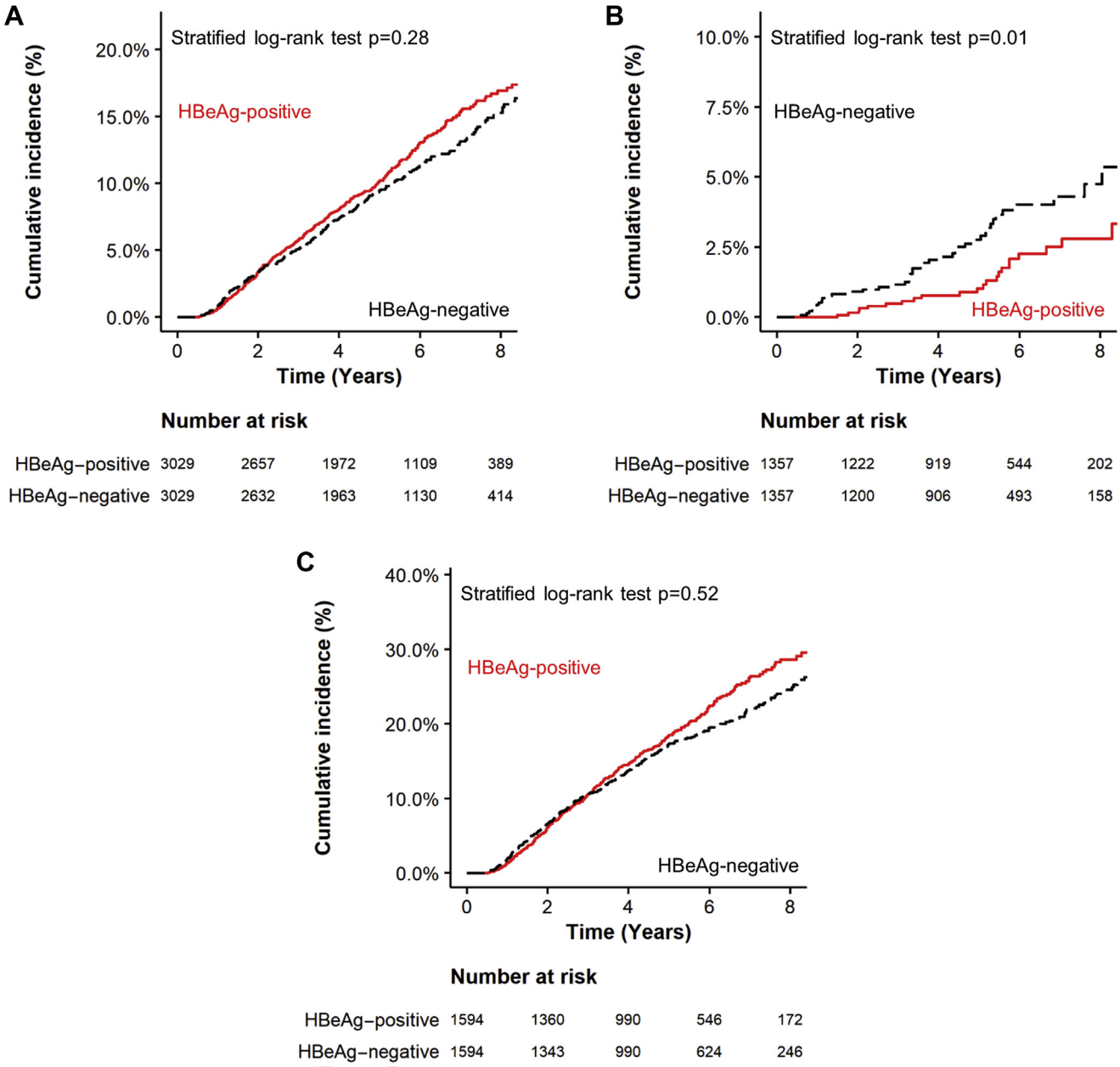

Figure 2. Cumulative incidence of hepatocellular carcinoma according to HBeAg status in the Korean cohort balanced by propensity score matching. Kaplan-Meier curves of the entire cohort (*A*), the non-LC subcohort (*B*), and the LC subcohort (*C*). The stratified log-rank test was used for the comparison between the HBeAg-positive and HBeAg-negative groups.

HBeAg-positivity has not been associated with the HCC risk in several previous studies including patients with CHB treated with oral antivirals.[10–13] Significant differences in baseline characteristics between HBeAg-positive and HBeAg-negative patients with CHB (eg, age, fibrosis stage, and serum HBV DNA level) may affect the results. Thus, all potential confounding factors need to be adjusted in order to properly assess the clinical impact of HBeAg positivity on HCC risk. Among the confounding factors, presence of LC undoubtedly represents the strongest predictor of HCC.[21] To neutralize the strong effect of LC, our patients were evaluated not only as an entire cohort but also as 2 different subcohorts divided according to the presence of LC. Our study included more than 9000 Korean patients with CHB, most of whom had genotype C virus infections characterized by a lengthy HBeAg-positive phase.[22] Thus, great numbers of HBeAg-positive (n = 4492) and HBeAg-negative (n = 4651) patients were included, allowing rigorous adjustments for potential confounding factors.

HBeAg positivity was not associated with the risk of HCC in the entire cohort or in the LC subcohort, which is in agreement with previous reports.[10–13] However, HBeAg positivity was independently associated with a lower HCC risk in the non-LC subcohort even after stratification by age, sex, serum HBV DNA level, and FIB-4 score. Our findings imply that HBeAg status may be a critical predictor of HCC risk in patients without LC, but

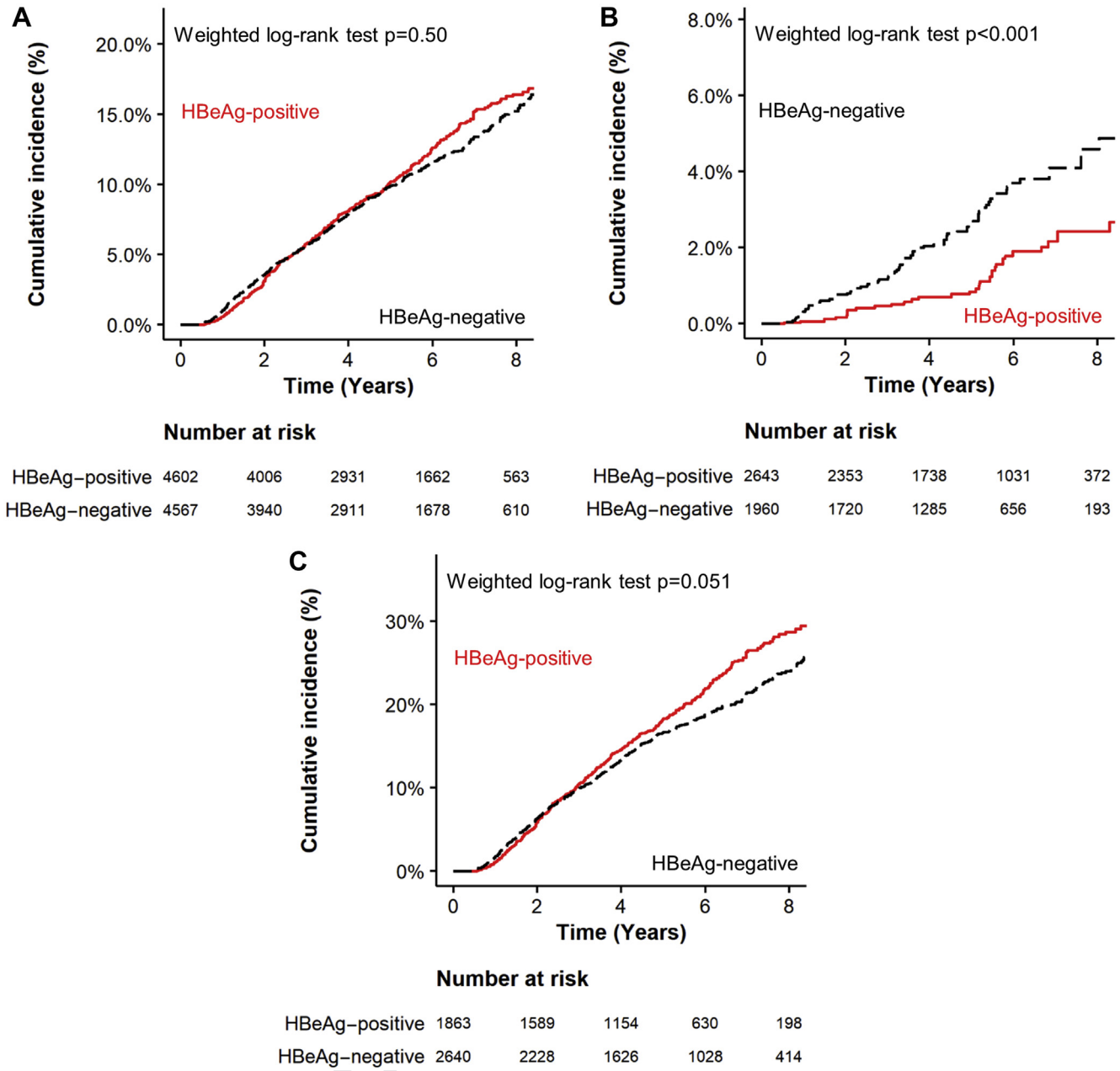

Figure 3. Cumulative incidence of hepatocellular carcinoma according to HBeAg status in the Korean cohort balanced by inverse probability of treatment weighting. Kaplan-Meier curves of the entire cohort (*A*), the non-LC subcohort (*B*), and the LC subcohort (*C*). The weighted log-rank test was used for the comparison between the HBeAg-positive and HBeAg-negative groups.

not in patients who have already developed LC (Supplementary Discussion).

Recent studies indicated that the integration of HBV DNA into the host genome is initiated during the HBeAg-positive phase.[8,9] Patients may have recurrent episodes of acute exacerbations before achieving spontaneous HBeAg seroclearance or seroconversion. During the spontaneous HBeAg seroclearance-associated liver turnover, hepatocytes with integrated HBV DNA can undergo selective clonal expansion, allowing for natural selection. As a result, HBeAg-negative patients have been found to have a >10-fold increase in the size of hepatocyte clones (>10,000 cells) compared with HBeAg-positive patients.[8,23] Antiviral treatment starting in the HBeAg-positive hepatitis phase can minimize the intensity and duration of active immune-mediated hepatic inflammation by directly reducing HBV DNA and thereby attenuating hepatocyte turnover and the selective pressure for clonal expansion of hepatocytes. Moreover, because NA treatment inhibits HBV DNA polymerase, which also synthesizes double-stranded linear DNA that can integrate into host genome in 1 of 10^5–10^6 infected cells,[24,25] early NA treatment might reduce the risk of host genome integration of HBV DNA. Antiviral treatment starting in the HBeAg-positive CHB phase may also reduce the emergence of precore/core mutants, which

Table 3. The Risk of HCC Development in the Non-LC Subcohort of the Korean Cohort

Variables	Univariable analysis		Multivariable analysis	
	HR (95% CI)	*P* value	HR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg-positive	0.33 (0.22-0.50)	< .001	0.41 (0.26-0.66)	< .001
Sex				
Females	1 [Reference]		1 [Reference]	
Males	2.08 (1.34-3.24)	.001	3.35 (2.12-5.30)	< .001
Age, *y*	1.07 (1.05-1.09)	< .001	1.07 (1.05-1.09)	< .001
Platelet,[a] $\times 1000/mm^3$	0.99 (0.98-0.99)	< .001		
Albumin, *g/dL*	0.60 (0.41-0.87)	.007	0.65 (0.44-0.96)	.03
Total bilirubin, *mg/dL*	0.99 (0.87-1.12)	.84		
ALT, *U/L*	0.998 (0.996-0.999)	.002	0.998 (0.996-0.999)	< .001
HBV DNA, log_{10} *IU/mL*	0.74 (0.65-0.85)	< .001	0.89 (0.76-1.04)	.16
FIB-4 score[a]	1.05 (1.03-1.07)	< .001	1.05 (1.02-1.08)	< .001

Note: The HR and *P* value were estimated using Cox proportional hazards regression.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, Fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio; LC, liver cirrhosis.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (Spearman's rho, −0.62; $P < .001$), only FIB-4 score was included in the multivariable analysis.

mainly occurs during spontaneous HBeAg seroclearance process and is reportedly associated with HCC risk as well.[26,27] These findings collectively provide the theoretical basis that antiviral treatment starting in the HBeAg-positive phase can not only reduce chronic necroinflammation and fibrosis progression, but also have a positive effect on pathways of direct carcinogenesis, thereby further lowering the risk of HCC. However, it is still unclear whether NA treatment can attenuate the rate of integration of HBV DNA into the host genome.[28] Therefore, serial assessments of HBV DNA integration during NA treatment are warranted to address this issue in future studies.

Whether to start NA treatment in HBeAg-positive patients who do not fulfill the current therapeutic indications (patients in the HBeAg-positive chronic HBV infection phase)[17] cannot be answered by our data. However, such HBeAg-positive patients should remain under regular follow-up, and treatment should be recommended as early as possible upon any sign of liver disease progression in order to prevent lengthy phases of CHB and/or progression to HBeAg-negative CHB and certainly development of LC.

Our study has several limitations. First, it was a retrospective cohort study. To minimize this limitation, we included a large number of patients – the largest cohort among studies of HCC prediction models[10–13,29–31] to date – and analyzed after rigorous adjustments. Second, the histologic severity of hepatic fibrosis was not evaluated. Instead, we used the FIB-4 score, which can classify the severity of hepatic fibrosis in patients with CHB with moderate sensitivity and accuracy.[32] To adjust for the severity of hepatic fibrosis, the FIB-4 score was included in the multivariable analysis, and subgroup analysis was performed based on the FIB-4 score as well.

In conclusion, our data suggests that baseline HBeAg positivity is independently associated with lower HCC risk in NA-treated patients with CHB without LC, even after adjustments for many confounders including age, severity of hepatic fibrosis, and HBV DNA levels. In contrast, HBeAg status upon NA treatment initiation does not seem to have any effect on the HCC risk if LC has developed. Such results may support the need for regular monitoring of patients in the HBeAg-positive chronic HBV infection phase (previously termed 'immune-tolerant') for timely onset of antiviral treatment before lengthy phases of reactivation and certainly before the development of LC in order to reduce the HCC risk in this setting.

Supplementary Material

Note: To access the supplementary material accompanying this article, visit the online version of Clinical Gastroenterology and Hepatology at www.cghjournal.org, and at http://doi.org/10.1016/j.cgh.2021.09.001.

References

1. Foreman KJ, Marquez N, Dolgert A, et al. Forecasting life expectancy, years of life lost, and all-cause and cause-specific

mortality for 250 causes of death: reference and alternative scenarios for 2016-40 for 195 countries and territories. Lancet 2018;392:2052–2090.
2. de Martel C, Maucort-Boulch D, Plummer M, et al. World-wide relative contribution of hepatitis B and C viruses in hepatocellular carcinoma. Hepatology 2015;62:1190–1200.
3. Wu CY, Lin JT, Ho HJ, et al. Association of nucleos(t)ide analogue therapy with reduced risk of hepatocellular carcinoma in patients with chronic hepatitis B: a nationwide cohort study. Gastroenterology 2014;147:143–151.e5.
4. Wong GL, Chan HL, Mak CW, et al. Entecavir treatment reduces hepatic events and deaths in chronic hepatitis B patients with liver cirrhosis. Hepatology 2013;58:1537–1547.
5. Marongiu F, Doratiotto S, Montisci S, et al. Liver repopulation and carcinogenesis: two sides of the same coin? Am J Pathol 2008;172:857–864.
6. Chemin I, Zoulim F. Hepatitis B virus induced hepatocellular carcinoma. Cancer Lett 2009;286:52–59.
7. Mason WS, Liu C, Aldrich CE, et al. Clonal expansion of normal-appearing human hepatocytes during chronic hepatitis B virus infection. J Virol 2010;84:8308–8315.
8. Mason WS, Gill US, Litwin S, et al. HBV DNA integration and clonal hepatocyte expansion in chronic hepatitis B patients considered immune tolerant. Gastroenterology 2016;151:986–998.e4.
9. Wooddell CI, Yuen MF, Chan HL, et al. RNAi-based treatment of chronically infected patients and chimpanzees reveals that integrated hepatitis B virus DNA is a source of HBsAg. Sci Transl Med 2017;9:eaan0241.
10. Kim JH, Kim YD, Lee M, et al. Modified PAGE-B score predicts the risk of hepatocellular carcinoma in Asians with chronic hepatitis B on antiviral therapy. J Hepatol 2018;69:1066–1073.
11. Papatheodoridis G, Dalekos G, Sypsa V, et al. PAGE-B predicts the risk of developing hepatocellular carcinoma in Caucasians with chronic hepatitis B on 5-year antiviral therapy. J Hepatol 2016;64:800–806.
12. Wong GL, Chan HL, Wong CK, et al. Liver stiffness-based optimization of hepatocellular carcinoma risk score in patients with chronic hepatitis B. J Hepatol 2014;60:339–345.
13. Kim DY, Song KJ, Kim SU, et al. Transient elastography-based risk estimation of hepatitis B virus-related occurrence of hepatocellular carcinoma: development and validation of a predictive model. OncoTargets Ther 2013;6:1463–1469.
14. Fujiwara N, Friedman SL, Goossens N, et al. Risk factors and prevention of hepatocellular carcinoma in the era of precision medicine. J Hepatol 2018;68:526–549.
15. Kim GA, Han S, Choi GH, et al. Moderate levels of serum hepatitis B virus DNA are associated with the highest risk of hepatocellular carcinoma in chronic hepatitis B patients. Alimentary pharmacology & therapeutics 2020;51:1169–1179.
16. Chen CJ, Yang HI, Su J, et al. Risk of hepatocellular carcinoma across a biological gradient of serum hepatitis B virus DNA level. JAMA 2006;295:65–73.
17. Terrault NA, Lok ASF, McMahon BJ, et al. Update on prevention, diagnosis, and treatment of chronic hepatitis B: AASLD 2018 hepatitis B guidance. Hepatology 2018;67:1560–1599.
18. Papatheodoridis GV, Sypsa V, Dalekos GN, et al. Hepatocellular carcinoma prediction beyond year 5 of oral therapy in a large cohort of Caucasian patients with chronic hepatitis B. J Hepatol 2020;72:1088–1096.
19. Sterling RK, Lissen E, Clumeck N, et al. Development of a simple noninvasive index to predict significant fibrosis in patients with HIV/HCV coinfection. Hepatology 2006;43:1317–1325.
20. Wong VW, Janssen HL. Can we use HCC risk scores to individualize surveillance in chronic hepatitis B infection? J Hepatol 2015;63:722–732.
21. Thiele M, Gluud LL, Fialla AD, et al. Large variations in risk of hepatocellular carcinoma and mortality in treatment naïve hepatitis B patients: systematic review with meta-analyses. PLoS One 2014;9:e107177.
22. Kim H, Jee YM, Song BC, et al. Molecular epidemiology of hepatitis B virus (HBV) genotypes and serotypes in patients with chronic HBV infection in Korea. Intervirology 2007;50:52–57.
23. Tu T, Mason WS, Clouston AD, et al. Clonal expansion of hepatocytes with a selective advantage occurs during all stages of chronic hepatitis B virus infection. J Viral Hepatitis 2015;22:737–753.
24. Summers J, Jilbert AR, Yang W, et al. Hepatocyte turnover during resolution of a transient hepadnaviral infection. Proceedings of the National Academy of Sciences of the United States of America 2003;100:11652–11659.
25. Yang W, Summers J. Integration of hepadnavirus DNA in infected liver: evidence for a linear precursor. J Virol 1999;73:9710–9717.
26. Liao Y, Hu X, Chen J, et al. Precore mutation of hepatitis B virus may contribute to hepatocellular carcinoma risk: evidence from an updated meta-analysis. PLoS One 2012;7:e38394.
27. Chotiyaputta W, Lok AS. Hepatitis B virus variants. Nat Rev Gastroenterol Hepatol 2009;6:453–462.
28. Tu T, Budzinska MA, Vondran FWR, et al. Hepatitis B virus DNA integration occurs early in the viral life cycle in an in vitro infection model via sodium taurocholate cotransporting polypeptide-dependent uptake of enveloped virus particles. J Virol 2018;92.
29. Lee HW, Yoo EJ, Kim BK, et al. Prediction of development of liver-related events by transient elastography in hepatitis B patients with complete virological response on antiviral therapy. Am J Gastroenterol 2014;109:1241–1249.
30. Yang HI, Yuen MF, Chan HL, et al. Risk estimation for hepatocellular carcinoma in chronic hepatitis B (REACH-B): development and validation of a predictive score. Lancet Oncol 2011;12:568–574.
31. Yuen MF, Tanaka Y, Fong DY, et al. Independent risk factors and predictive score for the development of hepatocellular carcinoma in chronic hepatitis B. J Hepatol 2009;50:80–88.
32. Xiao G, Yang J, Yan L. Comparison of diagnostic accuracy of aspartate aminotransferase to platelet ratio index and fibrosis-4 index for detecting liver fibrosis in adult patients with chronic hepatitis B virus infection: a systemic review and meta-analysis. Hepatology 2015;61:292–302.

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Acknowledgment Q6
The authors thank Prof Young-Suk Lim and Prof Jonggi Choi (University of Ulsan College of Medicine, Seoul, Republic of Korea) for sharing their data. The authors thank Prof Young Youn Cho (Chung-Ang University Hospital, Seoul, Republic of Korea) for providing intellectual comments.

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Conflicts of interest

These authors disclose the following: Seung Up Kim reports receiving grants from Yuhan Pharmaceuticals, and lecture fees from Bristol-Myers Squibb, Gilead Science, and Yuhan Pharmaceuticals; Yoon Jun Kim reports receiving research grants from Bristol-Myers Squibb, Roche, JW Creagene, Bukwang Pharmaceuticals, Handok Pharmaceuticals, Hanmi Pharmaceuticals, Yuhan Pharmaceuticals, and Pharmaking, and lecture fees from Bayer HealthCare Pharmaceuticals, Gilead Science, MSD Korea, Yuhan Pharmaceuticals, Samil Pharmaceuticals, CJ Pharmaceuticals, Bukwang Pharmaceuticals, and Handok Pharmaceuticals; Jung-Hwan Yoon reports receiving research grant from Bayer HealthCare Pharmaceuticals, Bukwang Pharmaceuticals, and Daewoong Pharmaceuticals; George V. Papatheodoridis has served as advisor/lecturer for Abbvie, Dicerna, Gilead, GlaxoSmithKline, Ipsen, Janssen, Merck Sharp & Dohme, Roche, and Spring Bank and has received research grants from Abbvie, Gilead; Jeong-Hoon Lee reports receiving lecture fees from GreenCross Cell, Daewoong Pharmaceuticals, and Gilead Korea. Q3

Funding Q4

The trial was supported by grants from Liver Research Foundation of Korea as part of Bio Future Strategies Research Project, Seoul National University Hospital Research Fund (grant number 03-2016-0380), and from the National Research Foundation of Korea (NRF) grants funded by the Korea government (MSIP) (grant number NRF-2019R1A2C2010311). Q7

Supplementary Methods

Patients

Patients who developed hepatocellular carcinoma (HCC) within 6 months from the start of nucleos(t)ide analog (NA) treatment were diagnosed with malignancy other than HCC, underwent organ transplantation, or were co-infected with hepatitis C virus or human immunodeficiency virus before or during the follow-up period were excluded. Liver cirrhosis (LC) was diagnosed by radiologic and clinical criteria as follows: (1) platelet count of <100,000/mL and a blunted, nodular liver edge accompanied by splenomegaly (>12 cm) and/or (2) the presence of esophageal or gastric varices, ascites, or hepatic encephalopathy.

Patients without LC had serum alanine aminotransferase (ALT) levels of ≥2 times the upper limit of normal (ULN) and serum hepatitis B virus (HBV) DNA levels of >20,000 or >2000 IU/mL for hepatitis B envelope antigen (HBeAg)-positive or HBeAg-negative cases at baseline. The ULN of ALT was defined as 35 U/L in males and 25 U/L in females. Patients with LC showed detectable levels of serum HBV DNA regardless of serum ALT levels at baseline. This study was approved by the institutional review board of each center (Supplementary Table 1).

Outcomes and Assessment

Fibrosis-4 score = Age (years) × aspartate aminotransferase (U/L) / [platelet count (×1000/mm^3) × ALT1/2 (U/L)].

Patients with inadequate liver ultrasonography were surveilled for HCC by alternative methods such as dynamic computed tomography (CT) and/or magnetic resonance imaging (MRI). HCC was diagnosed according to the American Association for the Study of Liver Diseases guidelines: (1) hepatic nodules ≥1 cm shows typical findings of HCC such as hypervascularity in the arterial phase and washout in the portal or delayed phase in dynamic CT or MRI; (2) if the HCC is not confirmed on either CT or MRI, the other study shows typical findings of HCC or biopsy confirms HCC.

Statistical Analyses

Continuous variables were described as mean ± standard deviation or median with interquartile range (IQR). Categorical variables were described as frequency and percentage. The baseline characteristics were compared by performing the *t*-test or Mann-Whitney *U* test for continuous variables and the χ^2 test or Fisher exact test for categorical variables.

Propensity score matching (PSM) was calculated by fitting a logistic regression model that included the following variables in both the Korean and Caucasian cohorts: age, sex, platelet count, serum levels of albumin, total bilirubin, ALT, and HBV DNA. A 1:1 ratio PSM was performed using the nearest neighbor method. In the inverse probability of treatment weighting (IPTW) analysis, we performed weight truncation at 0.5% and 99.5% to avoid the influence of extreme weights, and we used stabilized weights. The Cox proportional hazards regression model with robust sandwich variance estimator and Kaplan-Meier method were derived in the populations balanced by PSM and IPTW. The stratified log-rank test and the weighted log-test were performed in the PSM-based population and IPTW-based population, respectively.

Supplementary Results

HBeAg Status and HCC Risk in the Entire Korean Cohort

Both PSM and IPTW analyses balanced the differences in baseline characteristics between the HBeAg-positive and HBeAg-negative groups with standardized mean differences (SMDs) of <0.1 for all variables (Supplementary Tables 14 and 15).

HBeAg Status and HCC Risk in the Non-LC Subcohort of the Korean Cohort

After PSM and IPTW analyses for adjusting baseline characteristics between the HBeAg-positive and HBeAg-negative groups of the non-LC subcohort, the baseline characteristics were well-balanced (SMDs of <0.1 for all variables) (Supplementary Tables 16 and 17).

We sub-classified patients according to their HBV DNA levels and performed the PSM analyses separately for each subclassification. The HBV DNA levels of HBeAg-positive and HBeAg-negative groups were exactly matched in the matched population (Supplementary Figure 2). Other baseline characteristics were also well balanced between the HBeAg-positive and HBeAg-negative groups (Supplementary Table 18).

HBeAg Status and HCC Risk in the LC Subcohort of the Korean Cohort

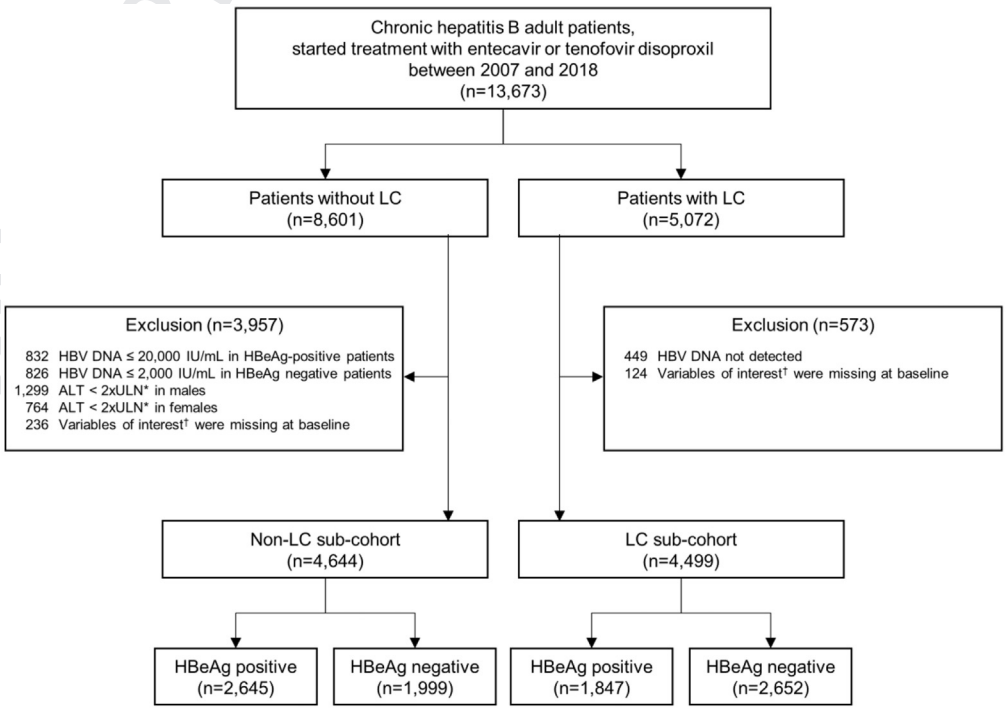
Both PSM and IPTW analyses were able to balance the baseline characteristics of the HBeAg-positive and HBeAg-negative groups in the LC subcohort (Supplementary Tables 19 and 20).

Supplementary Discussion

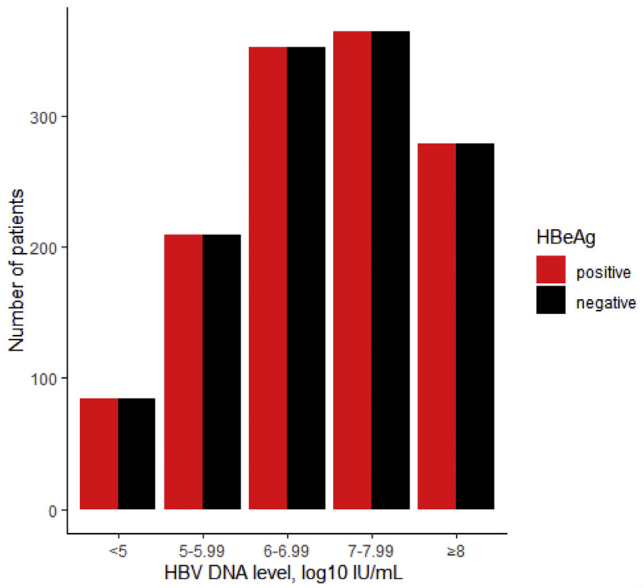
In the LC subcohort, the phase of chronic HBV infection upon NA treatment initiation did not affect the risk of HCC. NA treatment in patients with LC is considered to reduce the HCC risk mainly by blocking indirect

hepatocarcinogenesis mechanisms such as down-regulation of hepatic inflammation and reversal of hepatic fibrosis, which is theoretically unrelated to HBeAg status. In addition, patients who have progressed to LC have already harbored HBV DNA integration, and accumulated additional oncogenic events including inflammation, fibrosis, and long-term expression of viral proteins. Therefore, the HBeAg status at the onset of NA treatment cannot affect the HCC risk in patients with established LC who have many activated oncogenic mechanisms unrelated to the HBeAg phase of chronic HBV infection

Interestingly, NA treatment starting in the HBeAg-positive than HBeAg-negative CHB phase reduced the HCC risk only in our non-LC subcohorts. The effect of HBeAg status was obvious after any type of analysis and adjustment in our large Korean non-LC subcohort (n = 4644) as well as in the smaller Caucasian non-LC subcohort (n = 469) using PSM analysis. It should be noted that none of the Caucasian patients without LC who started NA treatment in the HBeAg-positive CHB phase developed HCC. In patients without LC, direct hepatocarcinogenesis, which is considered to be mainly induced by the integration of HBV DNA, is the predominant mechanism of HCC development, and thus blocking HBV DNA integration may be crucial for reducing the risk of HCC. Therefore, our findings in the non-LC subcohort may support the hypothesis that NA treatment starting in the HBeAg-positive CHB phase could block HBV DNA integration.

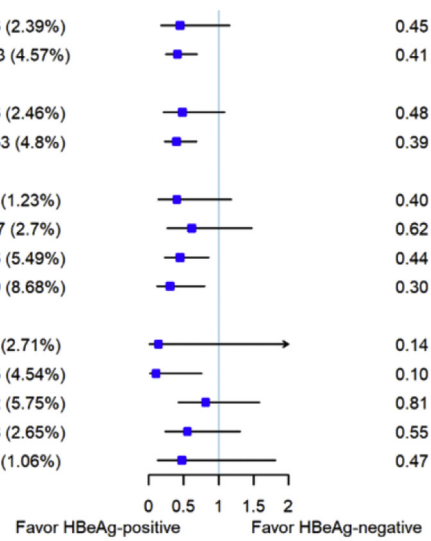


Supplementary Figure 1. CONSORT diagram of the Korean cohort. *Upper limits of normal: 35 U/L for males and 25 U/L for females. †Platelet count, albumin, and total bilirubin. ALT, Alanine aminotransferase; CONSORT, Consolidated Standards of Reporting Trials; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; LC, liver cirrhosis; ULN, upper limit of normal.

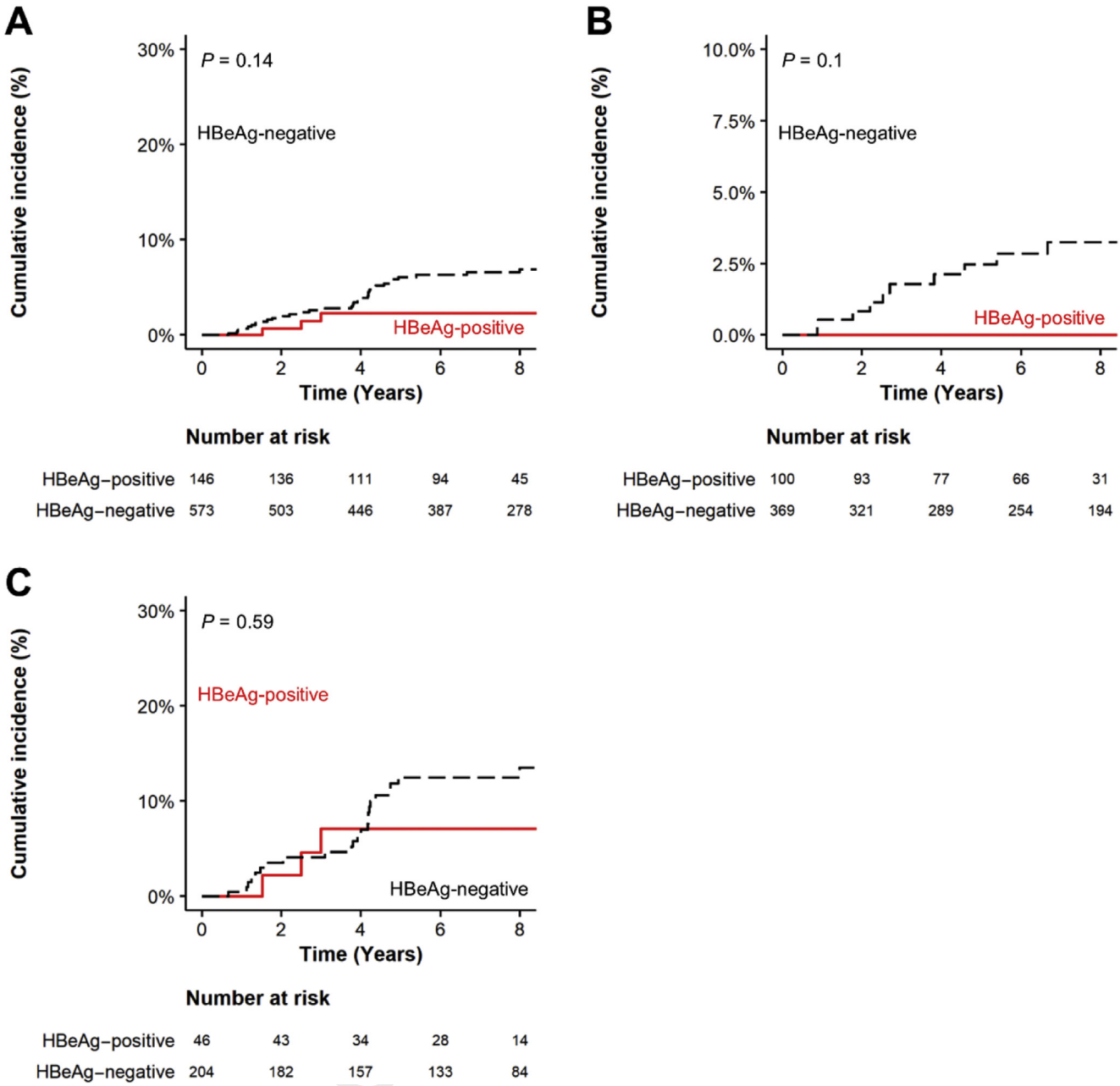


Supplementary Figure 2. Number of the matched patients in each subclassification according to HBV DNA level in the Korean cohort. HBeAg, Hepatitis B envelope antigen; HBV, hepatitis B virus.

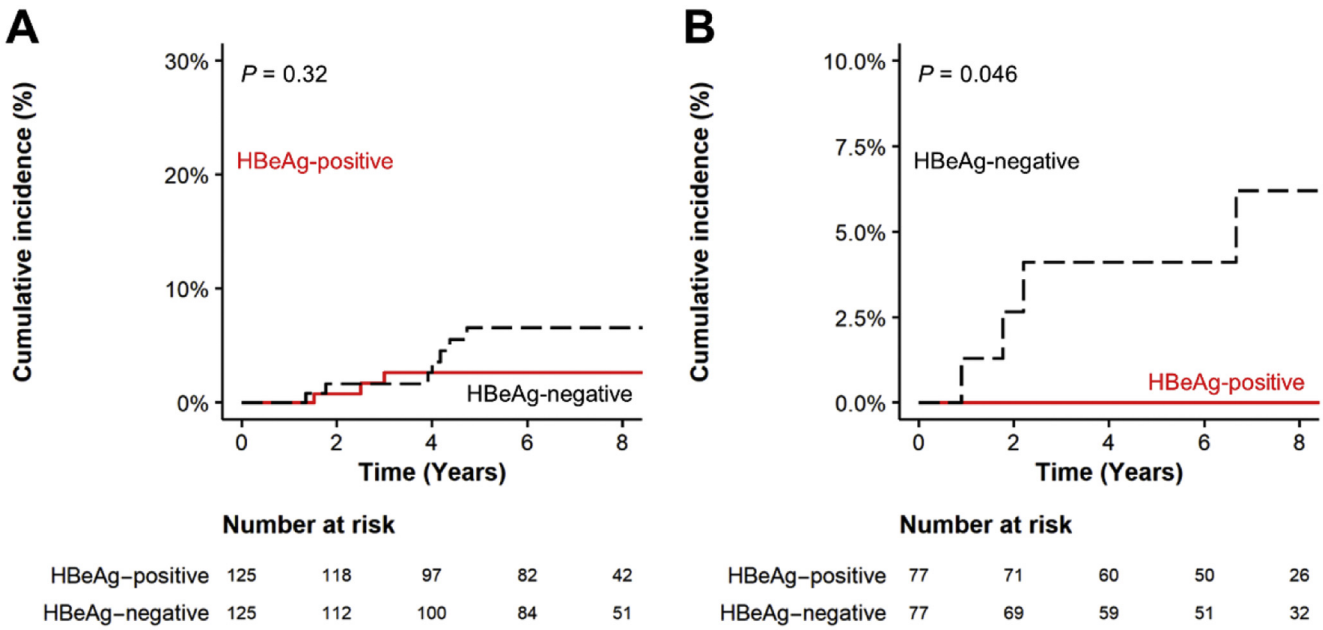
Subgroup	Number of patients HBeAg-positive	HBeAg-negative	Adjusted HR (95% CI)*	P value
Sex				
Females	7/1091 (0.64%)	19/796 (2.39%)	0.45 (0.17-1.15)	0.10
Males	27/1554 (1.74%)	55/1203 (4.57%)	0.41 (0.24-0.69)	<0.001
FIB-4 score†				
Under median, <1.94	11/1386 (0.79%)	23/936 (2.46%)	0.48 (0.21-1.09)	0.08
Over median, ≥1.94	23/1259 (1.83%)	51/1063 (4.8%)	0.39 (0.23-0.68)	<0.001
Age				
<40	6/1172 (0.51%)	7/567 (1.23%)	0.40 (0.13-1.18)	0.10
40-49	10/742 (1.35%)	18/667 (2.7%)	0.62 (0.26-1.48)	0.28
50-59	13/548 (2.37%)	30/546 (5.49%)	0.44 (0.23-0.86)	0.02
≥60	5/183 (2.73%)	19/219 (8.68%)	0.30 (0.11-0.80)	0.02
HBV DNA level, log10 IU/mL				
<5	1/95 (1.05%)	9/332 (2.71%)	0.14 (0.01-3.26)	0.22
5-5.99	1/222 (0.45%)	22/485 (4.54%)	0.10 (0.01-0.76)	0.03
6-6.99	13/411 (3.16%)	30/522 (5.75%)	0.81 (0.42-1.59)	0.55
7-7.99	11/663 (1.66%)	10/378 (2.65%)	0.55 (0.23-1.31)	0.18
≥8	8/1254 (0.64%)	3/282 (1.06%)	0.47 (0.12-1.82)	0.27

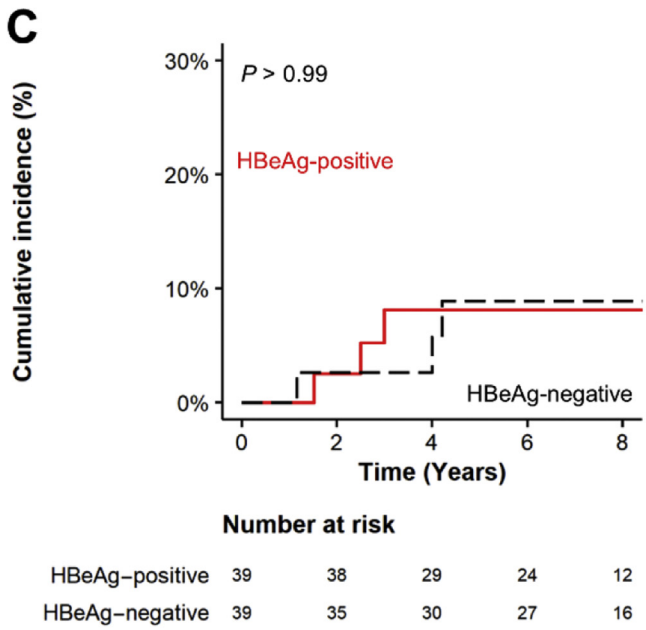


Supplementary Figure 3. Forest plots of adjusted HRs for HCC development by subgroups of sex, FIB-4 score, age, and HBV DNA levels in the non-LC subcohort of the Korean cohort. *Adjusted HRs and *P* values were estimated by multivariable Cox proportional hazards regression model adjusted for significant confounding variables, such as sex, age, hepatitis B virus DNA, and FIB-4 score. †FIB-4 score was stratified according to the median value. CI, Confidence interval; HBeAg, hepatitis B envelope antigen HCC, hepatocellular carcinoma; HR, hazard ratio; FIB-4, Fibrosis-4; LC, liver cirrhosis.



Supplementary Figure 4. Cumulative incidence of HCC according to HBeAg status in the Caucasian cohort. Kaplan-Meier curves of the entire cohort (*A*), the non-LC subcohort (*B*), and the LC subcohort (*C*). The log-rank test was used for the comparison between the HBeAg-positive and HBeAg-negative groups. HBeAg, Hepatitis B envelope antigen; HCC, hepatocellular carcinoma; LC, liver cirrhosis.





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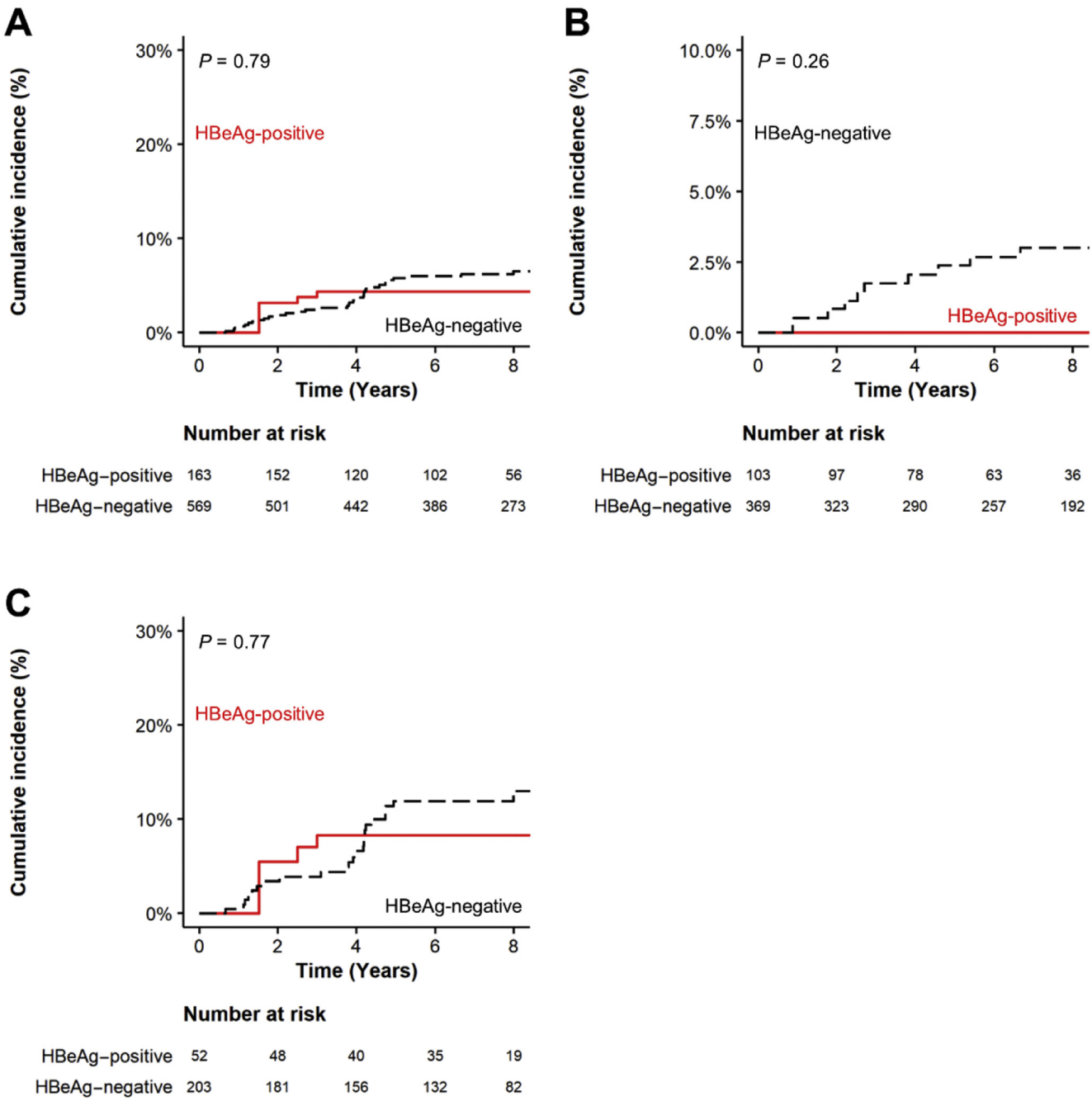
Supplementary Figure 5. Cumulative incidence of HCC according to HBeAg status in the Caucasian cohort balanced by PSM. Kaplan-Meier curves of the entire cohort (*A*), the non-LC subcohort (*B*), and the LC subcohort (*C*). The stratified log-rank test was used for the comparison between the HBeAg-positive and HBeAg-negative groups. HBeAg, Hepatitis B envelope antigen; HCC, hepatocellular carcinoma; LC, liver cirrhosis; PSM, propensity score matching.



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Supplementary Figure 6. Cumulative incidence of HCC according to HBeAg status in the Caucasian cohort balanced by inverse probability of treatment weighting. Kaplan-Meier curves of the entire cohort (*A*), the non-LC subcohort (*B*), and the LC subcohort (*C*). The weighted log-rank test was used for the comparison between the HBeAg-positive and HBeAg-negative groups. HBeAg, Hepatitis B envelope antigen; HCC, hepatocellular carcinoma; LC, liver cirrhosis.

Supplementary Table 1. Participating Centers

Hospital name	Location	IRB number	Year of starting NAs
Seoul National University Hospital	Seoul, Korea	2002-025-109	From 2007 to 2018
Severance Hospital	Seoul, Korea	4-2020-0487	From 2007 to 2018
Asan Medical Center	Seoul, Korea	2019-0507	From 2012 to 2017
Kyungpook National University Hospital	Daegu, Korea	2016-10-011	From 2011 to 2014
Samsung Medical Center	Seoul, Korea	2019-12-069	From 2012 to 2015
Ewha Womans University Mokdong Hospital	Seoul, Korea	2016-07-052	From 2007 to 2018
Korea University Anam Hospital	Seoul, Korea	2016AN0201	From 2007 to 2018
Hallym University Sacred Heart Hospital	Anyang, Korea	HALLYM 2018-10-002-001	From 2011 to 2015
Hanyang University Hospital	Seoul, Korea	HYUH 2016-09-028	From 2011 to 2015
Sanggye Paik Hospital	Seoul, Korea	SGPAIK 2016-10-011-001	From 2011 to 2015
Hanyang University Guri Hospital	Guri, Korea	GURI 2016-10-009-001	From 2011 to 2015
Eulji General Hospital	Seoul, Korea	EMCS 2016-10-009	From 2011 to 2015
Kyung Hee University Hospital	Seoul, Korea	KHUH 2018-02-040	From 2011 to 2015
Soonchunhyang University Hospital	Seoul, Korea	SCHUH 2016-10-022-001	From 2011 to 2015
Kangbuk Samsung Hospital	Seoul, Korea	KBSMC 2016-10-026	From 2012 to 2015
Hallym University Kangdong Sacred Heart Hospital	Seoul, Korea	KANGDONG 2016-10-018	From 2011 to 2015

IRB, Institutional review board; NAs, nucleos(t)ide analogues.

Supplementary Table 2. Baseline Characteristics of the Korean Cohort According to the Presence of Liver Cirrhosis

		Status of LC		
	Entire cohort	Non-LC subcohort	LC subcohort	*P* value
Variables	(N = 9143)	(n = 4644)	(n = 4499)	
Type of NAs				< .001
Entecavir	4895 (53.5)	2273 (48.9)	2622 (58.3)	
Tenofovir	4248 (46.5)	2371 (51.1)	1877 (41.7)	
Male sex	5510 (60.3)	2757 (59.4)	2753 (61.2)	.08
Age, *y*	49.2 ± 11.4	44.4 ± 11.4	54.1 ± 9.0	< .001
HBeAg positivity	4492 (49.1)	2645 (57.0)	1847 (41.1)	< .001
Platelet, $\times 1000/mm^3$	153 (113–197)	189 (153–227)	120 (89–152)	< .001
Albumin, *g/dL*	4.1 (3.8–4.4)	4.2 (3.9–4.4)	4.0 (3.6–4.3)	< .001
Total bilirubin, *mg/dL*	0.9 (0.7–1.3)	0.9 (0.7–1.2)	1.0 (0.7–1.4)	< .001
ALT, *U/L*	96 (53–184)	143 (96–272)	53 (35.5–94)	< .001
HBV DNA, log_{10} *IU/mL*	6.4 (5.3–7.7)	7.3 (6.0–8.2)	5.9 (4.6–6.7)	< .001
FIB-4 score	2.7 (1.7–4.5)	1.9 (1.2–3.2)	3.5 (2.4–5.8)	< .001

Note: Values are expressed as frequency (%), mean ± standard deviation, or median (interquartile range).
ALT, Alanine aminotransferase; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; LC, liver cirrhosis; NA, nucleos(t)ide analog.

Supplementary Table 3. Baseline Characteristics of the Korean Cohort According to the Presence of LC and HBeAg Status

	Non-LC subcohort (n = 4644)			LC subcohort (n = 4499)		
	HBeAg-positive	HBeAg-negative	*P* value	HBeAg-positive	HBeAg-negative	*P* value
Variables	(n = 2645)	(n = 1999)		(n = 1847)	(n = 2652)	
Male sex	1554 (58.8)	1203 (60.2)	.34	1126 (61.0)	1627 (61.3)	.82
Age, *y*	42.7 ± 11.5	46.6 ± 10.9	< .001	53.4 ± 9.3	54.7 ± 8.7	< .001
Platelet, $\times 1000/mm^3$	193 (156–230)	185 (149–220.5)	< .001	116 (87–150)	123 (90–153)	.002
Albumin, *g/dL*	4.1 (3.9–4.4)	4.2 (3.9–4.5)	< .001	3.9 (3.4–4.2)	4.1 (3.7–4.4)	< .001
Total bilirubin, *mg/dL*	0.9 (0.6–1.2)	0.9 (0.7–1.2)	.19	1.0 (0.7–1.5)	1.0 (0.7–1.3)	.06
ALT, *U/L*	145 (97–278)	141 (95–264.5)	.18	52 (36–88.5)	54 (35–100)	.54
HBV DNA, log_{10} *IU/mL*	7.9 (6.9–8.2)	6.3 (5.4–7.4)	< .001	6.3 (5.3–7.2)	5.5 (4.2–6.3)	< .001
FIB-4 score	1.8 (1.1–3.1)	2.0 (1.3–3.4)	< .001	3.8 (2.5–6.3)	3.4 (2.3–5.4)	< .001

Note: Values are expressed as frequency (%), mean ± standard deviation, or median (interquartile range).
ALT, Alanine aminotransferase; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; LC, liver cirrhosis.

Supplementary Table 4. Baseline Characteristics of the Caucasian Cohort

		Status of HBeAg		
	Entire cohort	HBeAg-positive	HBeAg-negative	*P* value
Variables	(N = 719)	(n = 146)	(n = 573)	
Male sex	503 (70.0)	110 (75.3)	393 (68.6)	.14
Age, *y*	51.8 ± 13.5	46.6 ± 15.0	53.2 ± 12.8	< .001
LC	250 (34.8)	46 (31.5)	204 (35.6)	.41
Platelet, $\times 1000/mm^3$	181 (142–225)	181 (150–225)	181 (141–223)	.77
Albumin, *g/dL*	4.3 (4.0–4.5)	4.2 (4.0–4.5)	4.3 (4.0–4.6)	.39
Total bilirubin, *mg/dL*	0.8 (0.6–1.0)	0.7 (0.5–1.0)	0.8 (0.6–1.0)	.11
ALT, *U/L*	96 (62–164.5)	98 (70.2–206)	96 (61–154)	.10
HBV DNA, log_{10} *IU/mL*	6.3 (4.9–7.4)	7.7 (6.1–8.8)	6.1 (4.7–7.0)	< .001

Note: Values are expressed as frequency (%), mean ± standard deviation, or median (interquartile range).
ALT, alanine aminotransferase; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; LC, liver cirrhosis.

Supplementary Table 5. The Risk of HCC Development in the Korean Cohort Balanced by Propensity Score Matching

	Univariable analysis		Multivariable analysis	
Variables	HR (95% CI)	*P* value	HR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg-positive	1.11 (0.95–1.29)	.19	1.10 (0.94–1.29)	.21
Sex				
Females	1 [Reference]		1 [Reference]	
Males	1.68 (1.41–1.99)	< .001	2.24 (1.88–2.67)	< .001
Age, *y*	1.06 (1.05–1.07)	< .001	1.04 (1.03–1.05)	< .001
LC	6.98 (5.53–8.81)	< .001	3.54 (2.72–4.62)	< .001
Platelet,[a] $\times 1000/mm^3$	0.988 (0.987–0.990)	< .001		
Albumin, *g/dL*	0.45 (0.40–0.51)	< .001	0.64 (0.56–0.73)	< .001
Total bilirubin, *mg/dL*	1.01 (0.98–1.04)	.52		
ALT, *U/L*	0.99 (0.99–1.00)	< .001	0.998 (0.997–0.999)	< .001
HBV DNA, log_{10} *IU/mL*	0.81 (0.77–0.85)	< .001	0.95 (0.90–1.01)	.09
FIB-4 score[a]	1.04 (1.03–1.05)	< .001	1.02 (1.01–1.03)	.003

Note: The HR and *P* value were estimated using Cox proportional hazards regression with robust sandwich variance estimator analysis.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio; LC, liver cirrhosis.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (Spearman's rho, −0.73; $P < .001$), only FIB-4 score was included in the multivariable analysis.

Supplementary Table 6. The Risk of HCC Development in the Korean Cohort Balanced by Inverse Probability of Treatment Weighting

	Univariable analysis		Multivariable analysis	
Variables	HR (95% CI)	*P* value	HR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg-positive	1.04 (0.90–1.21)	.56	1.03 (0.89–1.19)	.69
Sex				
Females	1 [Reference]		1 [Reference]	
Males	1.64 (1.39–1.94)	< .001	2.18 (1.84–2.58)	< .001
Age, *y*	1.06 (1.06–1.07)	< .001	1.04 (1.04–1.05)	< .001
LC	8.55 (6.91–10.56)	< .001	4.12 (3.24–5.25)	< .001
Platelet,[a] $\times 1000/mm^3$	0.988 (0.987–0.990)	< .001		
Albumin, *g/dL*	0.52 (0.46–0.59)	< .001	0.68 (0.60–0.76)	< .001
Total bilirubin, *mg/dL*	1.01 (0.99–1.03)	.40		
ALT, *U/L*	0.99 (0.99–1.00)	< .001	0.998 (0.997–0.999)	< .001
HBV DNA, log_{10} *IU/mL*	0.80 (0.77–0.84)	< .001	0.95 (0.90–1.00)	.07
FIB-4 score[a]	1.04 (1.04–1.05)	< .001	1.02 (1.01–1.03)	< .001

Note: The HR and *P* value were estimated using Cox proportional hazards regression with robust sandwich variance estimator analysis.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio; LC, liver cirrhosis.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (weighted Spearman's rho, −0.73; $P < .001$), only FIB-4 score was included in the multivariable analysis.

Supplementary Table 7. The Risk of HCC Development in the Non-LC Subcohort of the Korean Cohort Balanced by Propensity Score Matching

	Univariable analysis		Multivariable analysis	
Variables	HR (95% CI)	*P* value	HR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg-positive	0.49 (0.30–0.81)	.005	0.50 (0.30–0.83)	.008
Sex				
Females	1 [Reference]		1 [Reference]	
Males	2.42 (1.33–4.39)	.004	3.80 (2.13–6.78)	< .001
Age, *y*	1.08 (1.06–1.10)	< .001	1.08 (1.06–1.11)	< .001
Platelet,[a] $\times 1000/mm^3$	0.99 (0.98–0.99)	< .001		
Albumin, *g/dL*	0.49 (0.34–0.70)	< .001	0.66 (0.42–1.05)	.08
Total bilirubin, *mg/dL*	0.97 (0.87–1.08)	.60		
ALT, *U/L*	0.998 (0.995–1.001)	.16		
HBV DNA, log_{10} *IU/mL*	0.83 (0.70–0.98)	.03	0.88 (0.74–1.06)	.17
FIB-4 score[a]	1.09 (1.05–1.13)	< .001	1.02 (0.94–1.09)	.68

Note: The HR and *P* value were estimated using Cox proportional hazards regression with robust sandwich variance estimator analysis.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio; LC, liver cirrhosis.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (Spearman's rho, −0.61; $P < .001$), only FIB-4 score was included in the multivariable analysis.

Supplementary Table 8. The Risk of HCC Development in the Non-LC Subcohort of the Korean Cohort Balanced by Inverse Probability of Treatment Weighting

	Univariable analysis		Multivariable analysis	
Variables	HR (95% CI)	*P* value	HR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg-positive	0.47 (0.30–0.73)	< .001	0.44 (0.28–0.70)	< .001
Sex				
Females	1 [Reference]		1 [Reference]	
Males	1.81 (1.09–3.02)	.02	3.07 (1.80–5.23)	< .001
Age, *y*	1.07 (1.05–1.09)	< .001	1.07 (1.05–1.09)	< .001
Platelet,[a] $\times 1000/mm^3$	0.99 (0.98–0.99)	< .001		
Albumin, *g/dL*	0.53 (0.39–0.73)	< .001	0.66 (0.48–0.91)	.01
Total bilirubin, *mg/dL*	1.01 (0.99–1.04)	.21		
ALT, *U/L*	0.999 (0.997–1.001)	.30	0.999 (0.997–1.001)	.16
HBV DNA, log_{10} *IU/mL*	0.80 (0.71–0.91)	< .001	0.87 (0.76–1.00)	.046
FIB-4 score[a]	1.06 (1.03–1.08)	< .001	1.05 (1.03–1.08)	< .001

Note: The HR and *P* value were estimated using Cox proportional hazards regression with robust sandwich variance estimator analysis.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio; LC, liver cirrhosis.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (weighted Spearman's rho, −0.62; $P < .001$), only FIB-4 score was included in the multivariable analysis.

Supplementary Table 9. The Risk of HCC Development in the Non-LC Subcohort of the Korean Cohort Balanced by Propensity Score Matching in Each Subclassification According to HBV DNA Level

	Univariable analysis		Multivariable analysis	
Variables	HR (95% CI)	*P* value	HR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg-positive	0.58 (0.35–0.96)	.04	0.59 (0.35–0.98)	.04
Sex				
Females	1 [Reference]			
Males	1.74 (1.00–3.02)	.05		
Age, *y*	1.08 (1.05–1.10)	< .001	1.07 (1.05–1.09)	< .001
Platelet,[a] $\times 1000/mm^3$	0.98 (0.98–0.99)	< .001		
Albumin, *g/dL*	0.45 (0.32–0.64)	< .001	0.65 (0.43–0.99)	.046
Total bilirubin, *mg/dL*	1.00 (0.92–1.09)	.99		
ALT, *U/L*	0.998 (0.996–1.001)	.21		
FIB-4 score[a]	1.05 (1.02–1.07)	< .001	1.02 (0.99–1.05)	.19
HBV DNA, log_{10} *IU/mL*				
<5	1 [Reference]			
5–5.99	1.40 (0.39–5.04)	.60		
6–6.99	1.98 (0.60–6.54)	.26		
7–7.99	1.26 (0.37–4.29)	.72		
≥8	0.60 (0.15–2.38)	.46		

Note: The HR and *P* value were estimated using Cox proportional hazards regression with robust sandwich variance estimator analysis.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio; LC, liver cirrhosis.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (Spearman's rho, −0.62; $P < .001$), only FIB-4 score was included in the multivariable analysis.

Supplementary Table 10. The Risk of HCC Development in the Non-LC Subcohort of the Korean Cohort Accounting for Death as a Competing Risk

	Univariable analysis		Multivariable analysis	
Variables	SHR (95% CI)	*P* value	SHR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg-positive	0.32 (0.21–0.48)	< .001	0.43 (0.27–0.68)	< .001
Sex				
Females	1 [Reference]		1 [Reference]	
Males	2.17 (1.36–3.45)	.001	3.05 (1.90–4.88)	< .001
Age, *y*	1.07 (1.05–1.09)	< .001	1.07 (1.05–1.09)	< .001
Platelet,[a] $\times 1000/mm^3$	0.99 (0.98–0.99)	< .001		
Albumin, *g/dL*	0.67 (0.44–1.02)	.06		
Total bilirubin, *mg/dL*	0.97 (0.88–1.07)	.55		
ALT, *U/L*	0.998 (0.995–1.00)	.06		
HBV DNA, log_{10} *IU/mL*	0.74 (0.66–0.82)	< .001	0.90 (0.78–1.04)	.14
FIB-4 score[a]	1.05 (1.02–1.07)	< .001	1.04 (1.01–1.07)	.004

Note: The SHR and *P* value were estimated using Fine-Gray subdistribution hazards regression.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; LC, liver cirrhosis; SHR, subhazard ratio.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (Spearman's rho, −0.62; $P < .001$), only FIB-4 score was included in the multivariable analysis.

Supplementary Table 11. The Risk of HCC Development in the LC Subcohort of the Korean Cohort

Variables	Univariable analysis		Multivariable analysis	
	HR (95% CI)	*P* value	HR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg–positive	1.17 (1.02–1.35)	.02	1.09 (0.95–1.26)	.22
Sex				
Females	1 [Reference]		1 [Reference]	
Males	1.82 (1.56–2.13)	< .001	2.29 (1.95–2.69)	< .001
Age, *y*	1.03 (1.03–1.04)	< .001	1.04 (1.03–1.04)	< .001
Platelet,[a] *×1000/mm^3*	1.00 (0.99–1.00)	< .001		
Albumin, *g/dL*	0.61 (0.55–0.68)	< .001	0.66 (0.59–0.74)	< .001
Total bilirubin, *mg/dL*	1.01 (0.97–1.05)	.72		
ALT, *U/L*	0.998 (0.997–0.999)	< .001	0.998 (0.997–0.999)	< .001
HBV DNA, *log$_{10}$ IU/mL*	0.98 (0.94–1.03)	.45		
FIB-4 score[a]	1.03 (1.02–1.04)	< .001	1.02 (1.01–1.03)	.004

The HR and *P* value were estimated using Cox proportional hazards regression.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio; LC, liver cirrhosis.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (Spearman's rho, −0.72; $P < .001$), only FIB-4 score was included in the multivariable analysis.

Supplementary Table 12. The Risk of HCC Development in the LC Subcohort of the Korean Cohort Balanced by Propensity Score Matching

Variables	Univariable analysis		Multivariable analysis	
	HR (95% CI)	*P* value	HR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg-positive	1.12 (0.96–1.31)	.16	1.09 (0.93–1.28)	.26
Sex				
Females	1 [Reference]		1 [Reference]	
Males	1.77 (1.48–2.11)	< .001	2.17 (1.81–2.61)	< .001
Age, *y*	1.03 (1.02–1.04)	< .001	1.03 (1.02–1.04)	< .001
Platelet,[a] *×1000/mm^3*	1.00 (0.99–1.00)	< .001		
Albumin, *g/dL*	0.63 (0.55–0.71)	< .001	0.67 (0.59–0.76)	< .001
Total bilirubin, *mg/dL*	1.00 (0.96–1.03)	.90		
ALT, *U/L*	0.998 (0.997–0.999)	< .001	0.998 (0.997–0.999)	< .001
HBV DNA, *log$_{10}$ IU/mL*	0.95 (0.91–1.01)	.08		
FIB-4 score[a]	1.02 (1.01–1.03)	< .001	1.01 (1.00–1.03)	.14

The HR and *P* value were estimated using Cox proportional hazards regression with robust sandwich variance estimator analysis.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio; LC, liver cirrhosis.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (Spearman's rho, −0.73; $P < .001$), only FIB-4 score was included in the multivariable analysis.

Supplementary Table 13. The Risk of HCC Development in the LC Subcohort of the Korean Cohort Balanced by Inverse Probability of Treatment Weighting

	Univariable analysis		Multivariable analysis	
Variables	HR (95% CI)	*P* value	HR (95% CI)	*P* value
HBeAg				
HBeAg-negative	1 [Reference]		1 [Reference]	
HBeAg-positive	1.15 (0.99–1.33)	.07	1.13 (0.98–1.32)	.10
Sex				
Females	1 [Reference]		1 [Reference]	
Males	1.74 (1.47–2.04)	< .001	2.17 (1.84–2.57)	< .001
Age, *y*	1.04 (1.03–1.04)	< .001	1.04 (1.03–1.05)	< .001
Platelet,[a] *×1000/mm³*	1.00 (0.99–1.00)	< .001		
Albumin, *g/dL*	0.62 (0.56–0.70)	< .001	0.66 (0.59–0.75)	< .001
Total bilirubin, *mg/dL*	1.00 (0.97–1.04)	.86		
ALT, *U/L*	0.998 (0.997–0.999)	< .001	0.998 (0.997–0.999)	< .001
HBV DNA, log_{10} *IU/mL*	0.98 (0.93–1.03)	.40		
FIB-4 score[a]	1.02 (1.02–1.03)	< .001	1.02 (1.01–1.03)	.004

The HR and *P* value were estimated using Cox proportional hazards regression with robust sandwich variance estimator analysis.
ALT, Alanine aminotransferase; CI, confidence interval; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; HR, hazard ratio; LC, liver cirrhosis.
[a]In consideration of the significant correlation between platelet count and FIB-4 score (weighted Spearman's rho, −0.73; $P < .001$), only FIB-4 score was included in the multivariable analysis.

Supplementary Table 14. Baseline Characteristics of the Korean Cohort Balanced by Propensity Score Matching

		Status of HBeAg			
Variables	Entire cohort (N = 6058)	HBeAg-positive (n = 3029)	HBeAg-negative (n = 3029)	*P* value	Standardized difference
Male sex	3679 (60.7)	1855 (61.2)	1824 (60.2)	.43	0.021
Age, *y*	49.5 ± 11.0	49.5 ± 11.3	49.6 ± 10.6	.72	0.009
LC	3089 (51.0)	1528 (50.4)	1561 (51.5)	.41	0.022
Platelet, *×1000/mm³*	150 (110–194)	150 (110–197)	150 (111–192)	.85	0.003
Albumin, *g/dL*	4.1 (3.8–4.4)	4.1 (3.7–4.3)	4.1 (3.8–4.4)	.14	0.029
Total bilirubin, *mg/dL*	0.9 (0.7–1.3)	0.9 (0.7–1.3)	0.9 (0.7–1.3)	.47	0.006
ALT, *U/L*	95 (53–182)	92 (50–177)	100 (57–188)	.001	0.012
HBV DNA, log_{10} *IU/mL*	6.5 (5.6–7.4)	6.5 (5.6–7.6)	6.4 (5.6–7.3)	.003	0.063
FIB-4 score	2.8 (1.7–4.6)	2.8 (1.7–4.7)	2.8 (1.8–4.6)	1.00	0.007

Note: Values are expressed as frequency (%), mean ± standard deviation, or median (interquartile range).
ALT, alanine aminotransferase; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; LC, liver cirrhosis.

Supplementary Table 15. Baseline Characteristics of the Korean Cohort Balanced by Inverse Probability of Treatment Weighting

Variables	Entire cohort (N = 9143)	Status of HBeAg		*P* value	Standardized difference
		HBeAg-positive (n = 4492)	HBeAg-negative (n = 4651)		
Male sex	60.7	60.6	60.8	.87	0.004
Age, *y*	49.3 ± 11.3	49.3 ± 11.5	49.3 ± 11.0	.76	0.007
LC	51.7	52.2	51.3	.41	0.020
Platelet, $\times 1000/mm^3$	150 (110–194)	150 (109–196)	150 (111–192.6)	.74	0.007
Albumin, *g/dL*	4.1 (3.8–4.4)	4.1 (3.7–4.4)	4.1 (3.8–4.4)	.68	0.007
Total bilirubin, *mg/dL*	0.9 (0.7–1.3)	0.9 (0.7–1.3)	0.9 (0.7–1.3)	.06	0.001
ALT, *U/L*	93 (51–179)	88 (48–168.6)	99 (55–187)	< .001	0.006
HBV DNA, log_{10} *IU/mL*	6.3 (5.2–7.6)	6.4 (5.0–7.8)	6.3 (5.4–7.4)	.51	0.040
FIB-4 score	2.8 (1.7–4.6)	2.8 (1.7–4.6)	2.8 (1.7–4.6)	.81	0.002

Note: Values are expressed as %, mean ± standard deviation, or median (interquartile range).
ALT, Alanine aminotransferase; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; LC, liver cirrhosis.

Supplementary Table 16. Baseline Characteristics of the Non-LC Subcohort of the Korean Cohort Balanced by Propensity Score Matching

Variables	Non-LC subcohort (n = 2714)	Status of HBeAg		*P* value	Standardized difference
		HBeAg-positive (n = 1357)	HBeAg-negative (n = 1357)		
Male sex	1,624 (59.8)	822 (60.6)	802 (59.1)	.46	0.030
Age, *y*	44.8 ± 11.5	44.4 ± 11.9	45.3 ± 11.1	.06	0.072
Platelet, $\times 1000/mm^3$	184.5 (148–221.8)	184 (148–223)	185 (149–220)	1.00	0.014
Albumin, *g/dL*	4.2 (3.9–4.4)	4.2 (3.9–4.4)	4.2 (3.9–4.4)	.33	0.033
Total bilirubin, *mg/dL*	0.9 (0.7–1.2)	0.9 (0.7–1.2)	0.9 (0.7–1.2)	.76	0.028
ALT, *U/L*	147 (96–282.8)	144 (95–283)	149 (97–282)	.62	0.016
HBV DNA, log_{10} *IU/mL*	7.0 (6.1–7.9)	7.0 (6.1–8.0)	6.9 (6.1–7.8)	.06	0.064
FIB-4 score	2.0 (1.3–3.4)	2.0 (1.3–3.3)	2.1 (1.3–3.5)	.43	0.026

Note: Values are expressed as frequency (%), mean ± standard deviation, or median (interquartile range).
ALT, Alanine aminotransferase; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; LC, liver cirrhosis.

Supplementary Table 17. Baseline Characteristics of the Non-LC Subcohort of the Korean Cohort Balanced by Inverse Probability of Treatment Weighting

Variables	Non-LC subcohort (n = 4644)	Status of HBeAg: HBeAg-positive (n = 2645)	Status of HBeAg: HBeAg-negative (n = 1999)	*P* value	Standardized difference
Male sex	59.9	59.8	59.9	.96	0.002
Age, *y*	44.4 ± 11.5	44.4 ± 11.8	44.4 ± 11.2	.99	<0.001
Platelet, $\times 1000/mm^3$	188 (152–226)	190 (152–226)	186 (151–225)	.21	0.017
Albumin, *g/dL*	4.2 (3.9–4.4)	4.2 (3.9–4.4)	4.2 (3.9–4.4)	.42	<0.001
Total bilirubin, *mg/dL*	0.9 (0.7–1.2)	0.9 (0.6–1.2)	0.9 (0.7–1.2)	.49	0.010
ALT, *U/L*	144 (96–280)	140 (95–274.3)	149 (97–286)	.16	<0.001
HBV DNA, log_{10} *IU/mL*	7.2 (6.0–8.2)	7.3 (5.9–8.2)	7.1 (6.1–8.1)	.09	0.058
FIB-4 score	2.0 (1.2–3.3)	2.0 (1.2–3.3)	2.0 (1.2–3.4)	.30	0.002

Note: Values are expressed as %, mean ± standard deviation, or median (interquartile range).
ALT, Alanine aminotransferase; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; LC, liver cirrhosis.

Supplementary Table 18. Baseline Characteristics of the Non-LC Subcohort of the Korean Cohort Balanced by Propensity Score Matching in Each Subclassification According to HBV DNA Level

Variables	Non-cirrhotic subcohort (n = 2574)	Status of HBeAg: HBeAg-positive (n = 1287)	Status of HBeAg: HBeAg-negative (n = 1287)	*P* value	Standardized difference
Male sex	1,549 (60.2)	784 (60.9)	765 (59.4)	.47	0.030
Age, *y*	44.5 ± 11.4	44.3 ± 11.6	44.8 ± 11.1	.24	0.046
Platelet, $\times 1000/mm^3$	184 (147–222)	184 (148–224)	184 (147–220)	.39	0.024
Albumin, *g/dL*	4.2 (3.9–4.4)	4.2 (3.9–4.4)	4.2 (3.9–4.4)	.12	0.059
Total bilirubin, *mg/dL*	0.9 (0.7–1.2)	0.9 (0.6–1.2)	0.9 (0.7–1.2)	.40	0.014
ALT, *U/L*	149 (98–290)	144 (96–278)	153 (100–302)	.06	0.002
HBV DNA, log_{10} *IU/mL*	7.0 (6.1–7.9)	7.0 (6.1–7.9)	7.0 (6.1–7.9)	.94	0.002
FIB-4 score	2.0 (1.2–3.5)	2.0 (1.2–3.5)	2.1 (1.3–3.5)	.14	0.013

NOTE. Values are expressed as frequency (%), mean ± standard deviation, or median (interquartile range).
ALT, Alanine aminotransferase; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; LC, liver cirrhosis.

Supplementary Table 19. Baseline Characteristics of the LC Subcohort of the Korean Cohort Balanced by Propensity Score Matching

Variables	LC subcohort (n = 3188)	Status of HBeAg: HBeAg-positive (n = 1594)	Status of HBeAg: HBeAg-negative (n = 1594)	*P* value	Standardized difference
Male sex	1,958 (61.4)	980 (61.5)	978 (61.4)	.97	0.003
Age, *y*	53.6 ± 8.9	53.7 ± 9.2	53.5 ± 8.6	.57	0.020
Platelet, $\times 1000/mm^3$	119 (87–150.2)	118.5 (88–151)	119 (85–149)	.49	0.028
Albumin, *g/dL*	4.0 (3.5–4.3)	4.0 (3.5–4.3)	4.0 (3.5–4.3)	.38	0.019
Total bilirubin, *mg/dL*	1.0 (0.7–1.4)	1.0 (0.7–1.4)	1.0 (0.8–1.4)	.35	0.012
ALT, *U/L*	54 (36–94)	52 (36–85.8)	57 (37–104.8)	.002	0.010
HBV DNA, log_{10} *IU/mL*	6.0 (5.1–6.8)	6.0 (5.1–6.9)	6.0 (5.1–6.7)	.09	0.053
FIB-4 score	3.6 (2.4–6.0)	3.7 (2.4–6.1)	3.6 (2.4–5.9)	.82	0.009

Note: Values are expressed as frequency (%), mean ± standard deviation, or median (interquartile range).
ALT, Alanine aminotransferase; FIB-4, fibrosis-4; HBeAg, hepatitis B envelope antigen; HBV, hepatitis B virus; LC, liver cirrhosis.

Supplementary Table 20. Baseline Characteristics of the LC Subcohort of the Korean Cohort Balanced by Inverse Probability of Treatment Weighting

Variables	LC subcohort (n = 4499)	Status of HBeAg: HBeAg-positive (n = 1847)	Status of HBeAg: HBeAg-negative (n = 2652)	*P* value	Standardized difference
Male sex	61.4	61.4	61.5	.99	<0.001
Age, years	53.9 ± 8.9	53.8 ± 9.2	54.0 ± 8.8	.50	0.022
Platelet, $\times 1000/mm^3$	120 (88–151)	119 (89–151)	120 (87–152)	.94	0.002
Albumin, *g/dL*	4.0 (3.6–4.3)	4.0 (3.6–4.3)	4.0 (3.6–4.3)	.80	0.018
Total bilirubin, *mg/dL*	1.0 (0.7–1.4)	1.0 (0.7–1.4)	1.0 (0.7–1.4)	.12	0.010
ALT, *U/L*	53 (35–94)	51 (35–84.3)	55 (36–102)	.001	0.005
HBV DNA, log_{10} *IU/mL*	5.8 (4.6–6.7)	5.8 (4.4–6.8)	5.8 (4.7–6.6)	1.00	0.028
FIB-4 score	3.5 (2.4–5.8)	3.6 (2.4–5.8)	3.5 (2.4–5.8)	.79	0.002

Note: Values are expressed as %, mean ± standard deviation, or median (interquartile range).
LC, liver cirrhosis; HBeAg, hepatitis B envelope antigen; ALT, alanine aminotransferase; HBV, hepatitis B virus; FIB-4, fibrosis-4.