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ORIGINAL ARTICLE

Effectiveness and Safety of Entecavir or Tenofovir in a Spanish Cohort of Chronic Hepatitis B Patients: Validation of the Page-B Score to Predict Hepatocellular Carcinoma

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Abstract

Background Long-term antiviral therapy has resulted in viral suppression and biochemical response in chronic hepatitis B, although the risk of hepatocellular carcinoma has not been abolished. The Page-B score could be useful to estimate the probability of HCC.

Aims To analyze the effectiveness and safety of entecavir or tenofovir for more than 4 years and the usefulness of Page-B score in the real-world setting.

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Methods Analysis of Caucasian chronic hepatitis B subjects treated with entecavir or tenofovir from the prospective, multicenter database CIBERHEP.
Results A total of 611 patients were enrolled: 187 received entecavir and 424 tenofovir. Most were men, mean age 50 years, 32% cirrhotic and 16.5% HBeAg-positive. Mean follow-up was 55 (entecavir) and 49 (tenofovir) months. >90% achieved HBV DNA <69 IU/mL and biochemical normalization by months 12 and 36, respectively. Cumulative HBeAg loss and anti-HBe seroconversion were achieved by 33.7 and 23.8%. Four patients lost HBsAg; three HBeAg-positive. Renal function remained stable on long-term follow-up. Fourteen (2.29%) developed HCC during follow-up all of them with baseline Page-B ≥10. Nine were diagnosed within the first 5 years of therapy. This contrasts with the 27 estimated by Page-B, a difference that highlights the importance of regular HCC surveillance even in patients with virological suppression.
Conclusions Entecavir and tenofovir achieved high biochemical and virological response. Renal function remained stable with both drugs. A Page-B cut-off ≥10 selected all patients at risk of HCC development.

Keywords Hepatitis B · Hepatocellular carcinoma · Page-B · Tenofovir · Entecavir · Effectiveness · Safety

Introduction

Entecavir (ETV) and tenofovir disoproxil fumarate (TDF) are approved first-line treatments for patients with chronic hepatitis B (CHB) infection. In controlled trials in this population, long-term therapy with either TDF or ETV resulted in viral suppression in most hepatitis B e antigen (HBeAg)-positive and HBeAg-negative patients [1].

Persistent suppression of viral replication leads to favorable clinical outcomes, such as improvements in liver fibrosis—in some cases even reversion of cirrhosis [2]—and a lower risk of hepatic decompensation and death, particularly in patients with cirrhosis [3]. Nonetheless, the likelihood of developing hepatocellular carcinoma (HCC) remains even with successful treatment, although the number of HCC cases decreases after several years of viral suppression.

Several scores for predicting the risk of HCC have been developed, the majority in Asian populations under no therapy. Recently, a new a score was developed for Caucasians receiving antiviral therapy with ETV or TDF, called the Page-B score [4]. This score estimates HCC risk within the first 5 years of treatment based on age, sex and platelet count at the start of therapy. A Page-B cut-off of ≥10 points selected all patients who developed HCC, with a sensitivity and negative predictive value of 100%.

Several studies have investigated the efficacy and safety of ETV or TDF in clinical practice. Most have a relatively short follow-up and are focused on effectiveness [5, 6]. There is little data on the efficacy and side effects of these two nucleos(t)ide analogues (NUCs) in clinical practice in very homogeneous patient cohorts [7, 8], particularly with regard to renal function over long-term follow-up.

The aim of this study was to analyze the effectiveness and safety of TDF or ETV in real-world practice in Caucasian patients followed for more than 4 years. The secondary aim was to validate the Page-B score, a model to predict the risk of developing HCC in this patient population.

Patients and Methods

Study Design and Patients

CIBERHEP is an observational, prospective and monitored national registry of patients with chronic hepatitis B infection managed in routine clinical practice. The registry is governed by the Spanish Association for the Study of the Liver (AEEH) and the Networked Biomedical Research Centre for the Study of the Liver and Digestive Diseases (CIBERehd). Monitoring is a key element of the database, ensuring accuracy of data and minimization of bias. Patients were included between April 2005 and September 2015 in 20 community and university hospitals throughout Spain. All patients gave written consent for the collection of anonymized medical data from their medical records.

CIBERHEP uses an electronic web-based case report form where demographical, clinical and laboratory data, treatment history, and follow-up are recorded. The decision to treat, the choice of treatment, and the follow-up are entirely at the discretion of the treating physician. Laboratory testing is performed at the local clinical laboratories in each center every 6 months. The following routine blood tests are carried out: alanine aminotransferase (ALT), serum hepatitis B virus (HBV) DNA levels (lower limit of detection at each laboratory varied from 10 to 69 IU/mL), HBeAg, HBsAg, and renal function parameters (serum creatinine and estimated glomerular filtration rate (eGFR) determined by the modification of diet in renal disease (MDRD) equation [9]).

The study population comprised adult Caucasians (≥18 years old) with CHB, both HBeAg-positive and HBeAg-negative, and who had received TDF or ETV for a

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minimum of 6 months. Patients with hepatitis delta virus, hepatitis C virus or human immunodeficiency virus coinfection were excluded. As one of the study endpoints was to validate the Page-B score, decompensated patients or those who underwent liver transplantation were also excluded.

The diagnosis of cirrhosis was based on histology, ultrasonography findings (nodular hepatic edge, spleen >12 cm, portal vein >16 mm), a previous history of hepatic decompensations (ascites, hepatic encephalopathy and/or upper gastrointestinal tract bleeding) and/or analytical data (persistent platelet count <140 × 10E9/mL). The flowchart for selecting patients from the CIBERHEP cohort is shown in Fig. 1.

The primary endpoints were virological response (defined as HBV DNA < 69 IU/mL), biochemical response (ALT normalization, defined as ALT < 35 IU/mL for women and <50 IU/mL for men), HBeAg loss, anti-HBe seroconversion, and hepatitis B surface antigen (HBsAg) loss.

HCC surveillance was conducted at each site at the discretion of the investigators following the guidelines of the European (EASL) and Spanish (AEEH) Associations for the Study of the Liver [10, 11]. The diagnosis of HCC was based on histological and/or radiological findings [12]. Follow-up was the time interval between study entry and the last available clinical information, up to February 2016. Analysis time was the time interval between study entry and the HCC diagnosis or end of follow-up in the absence of HCC development. For every HCC case observed in the CIBERHEP cohort, the number of estimated cases based on the baseline Page-B score of patients at risk was calculated.

Statistical Analysis

All statistical analyses were performed using IBM SPSS 20 (SPSS Inc., Chicago, USA).

Standard descriptive statistics of patient' characteristics by antiviral drug and later HCC development were conducted. The nonparametric Kruskal–Wallis test or the two-sided Fisher exact test was used for comparisons. Comparative analyses were performed using Pearson's Chi-square test for qualitative variables and Student's *t* test for analyses of variance for quantitative variables. Nonparametric tests were used in the case of a non-normal distribution, e.g., Wilcoxon test for paired comparisons. Cumulative probabilities of HCC occurrence and HBeAg loss in different subgroups were estimated using the Kaplan–Meier method and compared using the log rank test. Discrimination of the Page-B model in our cohort was assessed using Harrell's *c*-index. Significance was set at $p < 0.05$.

Predicted HCC incidence was calculated using Page-B [4], a Cox proportional hazards model relating HCC risk to

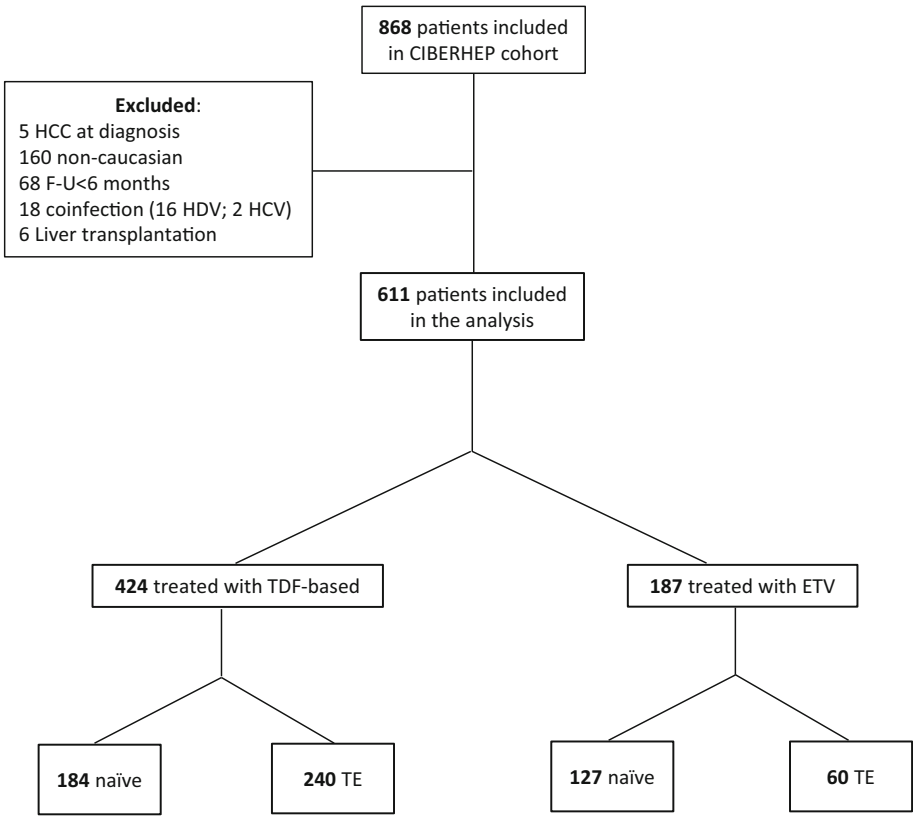

Fig. 1 Flowchart of enrollment and final treatment groups. *ETV* entecavir, *F-U* follow-up, *HCC* hepatocellular carcinoma, *HCV* hepatitis C virus, *HDV* hepatitis delta virus, *PT* previously treated, *TDF* tenofovir disoproxil fumarate

age, sex and platelet count at the beginning of TDF or ETV therapy. The Cox model coefficients were obtained from Papatheodoridis et al. [4]. Detailed survival estimates in yearly increments from 1 to 5 were obtained from the Page-B authors. We used linear interpolation to estimate the baseline disease-free survival for events occurring between full calendar years.

Results

Baseline Characteristics

In total, 809 patients were evaluated and 611 were enrolled. The reasons for exclusion and type of treatment are shown in Fig. 1. ETV at dose of 0.5 or 1 mg per day (patients previously treated with lamivudine) was given to 187 patients, and TDF at dose of 245 mg per day was given alone or in combination regimens to 424 patients: 383 TDF monotherapy, 14 emtricitabine plus TDF, 13 ETV plus TDF, 14 lamivudine plus TDF. Mean age was 50 ± 13 years. Most patients were men, HBeAg-negative, and 300 (49.1%) had been previously treated with first-generation NUCs (Table 1). All patients were Caucasian, the majority of European descent (91% Southern Europe and 7% Eastern Europe). A larger percentage of patients treated with ETV were naïve ($p < 0.001$). Treatment-experienced patients receiving ETV had higher baseline ALT and HBV DNA levels than those given a TDF-containing regimen ($p = 0.01$ and $p = 0.005$, respectively). Among naïve patients, ALT and HBV DNA levels were similar regardless of the analogue given. Overall, 197 patients (32.2%) had liver cirrhosis at the beginning of NUC therapy and 52 (8.5%) showed sonographic signs of portal hypertension.

Mean duration of follow-up was 55 ± 22 months in patients receiving ETV and 49 ± 29 in those given TDF-containing regimens ($p = 0.002$). Overall, the number of patients who reached 12, 24, 36, 48 and 60 months of follow-up was 579, 506, 428, 342, and 224, respectively.

Primary Endpoints

Virological Response

In patients treated with TDF-containing regimens and having available data, HBV DNA was <69 IU/mL in 92% ($n = 224$) by month 12, a value that rose to 98% at month 60 ($n = 56$). Prior treatment did not affect response: a similar percentage of treatment-naïve and treatment-experienced patients reached HBV DNA < 69 IU/mL by month 12 (92% vs. 91.5%). In patients receiving ETV and with available data, HBV DNA was < 69 IU/mL in 85% ($n = 102$) by month 12, increasing to 100% at month 60 ($n = 44$).

Table 1 Baseline characteristics of patients with chronic hepatitis B according to antiviral therapy with second-line analogues

	Total $N = 611$	TDF-containing regimens $N = 424$	ETV $N = 187$	p value
Age, years, mean ± SD	50 ± 13	50 ± 13	50 ± 13	0.98
Men, *n* (%)	444 (72.7%)	305 (71.9%)	139 (74.3%)	0.31
Treatment experienced, *n* (%)	300 (49.0%)	240 (56.6%)	60 (32.1%)	<0.001
Cirrhosis, *n* (%)	197 (32.2%)	133 (31.4%)	64 (34.2%)	0.25
HBeAg-positive, *n* (%)	101 (16.5%)	67 (15.8%)	34 (18.2%)	0.27
ALT, IU/mL, mean ± SD				
Overall, $N = 609$	90 ± 235	69 ± 139	138 ± 367	<0.001
Naïve, $N = 310$	128 ± 315	101 ± 187	166 ± 436	0.13
Treatment-experienced, $N = 299$	51 ± 87	44 ± 78	78 ± 111	0.01
Platelets, 10E9/mL, mean ± SD	191 ± 70	192 ± 70	189 ± 71	0.39
Creatinine, mg/dL, mean ± SD	0.96 ± 0.28	1.11 ± 0.22	1.06 ± 0.20	0.001
HBV DNA, log IU/mL, mean ± SD				
Overall, $N = 556$[a]	4.0 ± 2.4	3.8 ± 2.3	4.9 ± 2.4	<0.001
Naïve, $N = 299$	5.2 ± 2.1	5.1 ± 2.1	5.4 ± 2.3	0.23
Treatment experienced, $N = 257$	3.0 ± 2	2.7 ± 1.9	3.9 ± 2.3	0.005
Undetectable HBV DNA, *n* (%)[a]	159 (26.0%)	130 (30.7%)	29 (15.5%)	<0.001
PAGE-B score, mean ± SD	13 ± 5	13 ± 5	13 ± 6	0.42

ETV entecavir, *MELD* model for end-stage-liver disease, *TDF* tenofovir disoproxil fumarate, *SD* standard deviation

[a] Available in 558 patients: 382 treated with TDF-based therapy and 176 with ETV

At baseline, there was a higher percentage of patients with HBV DNA < 69 IU/mL among HBeAg-negative patients than HBeAg-positive patients (28.8 vs. 18.6%, $p = 0.039$). This difference tended to increase after the beginning of treatment (Month 12: 95.0 vs. 71.2%, $p < 0.001$; Month 24: 95.1 vs. 87.5%, $p = 0.05$; Month 36: 96.6 vs. 87%, $p = 0.03$), and lost significance by month 48 (97.3 vs. 95.7%, $p = 0.68$) and 60 (98.6 vs. 100%, $p = 0.62$). Patient age had an impact on early virological response in both therapies. Baseline HBV DNA levels (4.2 vs. 4.0 log IU/mL, $p = 0.43$) and percentage of patients with baseline HBV DNA < 69 IU/mL (28 vs. 32%, $p = 0.55$) were similar between older and younger patients. However, subjects older than 65 achieved a lower virological response by month 12 with either TDF (80.6 vs. 93.8%, $p = 0.013$) or ETV (64.7 vs. 90.5%, $p = 0.005$) than younger patients. This difference disappeared during follow-up (95 and 80% at month 24 for TDF-containing and ETV regimens, respectively). A summary of the results for the primary endpoints is shown in Table 2.

Biochemical Response

Overall, 84 and 90% of patients achieved normalization of ALT levels at 36 and 60 months after starting therapy, with a similar percentage for ETV (84 and 92%) and TDF (84 and 88%). Abnormal baseline ALT levels were seen in 184 (43.5%) and 126 (67.7%) patients treated with a TDF-containing regimen or ETV, respectively. In these patients, ALT had normalized in 83.9 and 79.6% at month 36 of follow-up and in 84 and 87.5% at month 60 (Table 2).

HBeAg Loss and Seroconversion

In total, 101 (18.6%) patients (67 treated with TDF and 34 with ETV) were HBeAg- positive at the beginning of NUC therapy; 34 (33.7%) and 24 (23.8%) patients, respectively, achieved HBeAg loss and anti-HBe seroconversion during follow-up. HBeAg loss and anti-HBe seroconversion were achieved by 31.3 and 22.4% of patients treated with TDF-containing regimens, and 26.5 and 38.2% of those receiving ETV. The cumulative HBeAg loss and anti-HBe seroconversion rates did not differ between the two regimens (Fig. 2).

HBsAg Loss

Four (0.7%) patients lost HBsAg during follow-up. All were treatment-naïve, three were HBeAg-positive at baseline (two treated with TDF and one with ETV) and younger than 65 years. The remaining patient was HBeAg-negative at baseline and was treated with ETV.

Secondary Outcomes

Safety

Seven (1.7%) patients treated with TDF-containing regimens and 4 (2.1%) treated with ETV required dose adjustment, mainly due to renal impairment. The status of renal function during therapy with ETV and TDF-containing regimens is summarized in Table 3. Overall, mean serum creatinine levels and glomerular filtration estimated

Table 2 Virological and biochemical data, and HBsAg loss during follow-up, according to the nucleos(t)ide analogue regimen received

	Baseline	Month 12	Month 36	Month 60
HBV DNA < 69 UI/mL, *n* (%)				
TDF-containing regimens				
All, *N* = 424	136 (32.1%)	206 (92.0%)	118 (96.0%)	56 (98.2%)
Treatment naïve, *N* = 184	11 (5.9%)	98 (92.5%)	46 (97.9%)	18 (94.7%)
ETV				
All, *N* = 187	30 (16.0%)	87 (85.3%)	65 (100%)	44 (100%)
Treatment naïve, *N* = 127	9 (7.1%)	65 (83.3%)	45 (90.0%)	32 (100%)
Normal ALT levels *n* (%)				
TDF-containing regimens				
All, *N* = 424	255 (60.1%)	222 (86%)	121 (84.0%)	53 (88.3%)
Abnormal ALT at BL, *N* = 169	–	78 (73.6%)	47 (83.9%)	21 (84.0%)
ETV				
All, *N* = 187	80 (42.8%)	97 (85.1%)	70 (84.5%)	47 (92.2%)
Abnormal ALT at BL, *N* = 106	–	59 (80.8%)	39 (79.6%)	28 (87.5%)
Cumulative HBsAg loss, *n* (%)				
TDF-containing regimens, *N* = 424	–	0 (0%)	2 (0.47%)	2 (0.47%)
ETV, *N* = 187	–	0 (0%)	2 (1.07%)	2 (1.07%)

BL baseline, *ETV* entecavir, *TDF* tenofovir disoproxil fumarate, % percentage

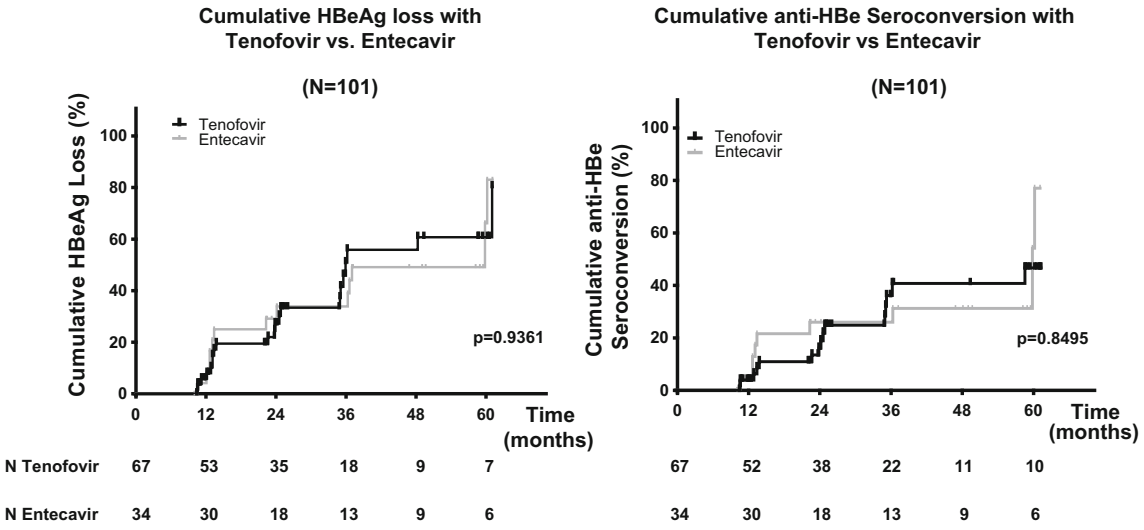

Fig. 2 Cumulative HBeAg loss and anti-HBe seroconversion in patients receiving TDF-containing regimens or ETV

by the MDRD at month 36 of follow-up did not differ from baseline ($p = 0.064$ and $p = 0.13$, respectively) and improved significantly at month 60 ($p < 0.001$ and $p < 0.001$, respectively). These results were maintained on separate analysis of patients receiving each NUC (TDF: $p = 0.05$ and $p = 0.17$ at month 36; $p = 0.019$ and $p = 0.025$ at month 60; ETV: $p = 0.58$ and $p = 0.56$ at month 36; $p = 0.019$ and $p = 0.010$ at month 60). Naïve and previously treated (PT) patients showed similar serum creatinine and estimated glomerular filtration values at month 36 of follow-up (median change from baseline: 0 mg/dL, $p = 0.35$ and 0 mL/min, $p = 0.46$ for naïve; 0 mg/dL, $p = 0.1$ and 0 mL/min, $p = 0.19$ for PT) and both presented slight improvements at month 60 (median change from baseline: −0.05 mg/dL, $p = 0.007$ and 4.2 mL/min, $p = 0.004$ for naïve; 0.05 mg/dL, $p = 0.04$ and 5.8 mL/min, $p = 0.06$ for PT).

Concerning age, renal function was stable in patients older than 65 years at the beginning of ETV or TDF in comparison with month 36 and 60 (median change from baseline: −0.01 mg/dL, $p = 0.92$ and 0 mL/min, $p = 0.73$ at month 36; 0.03 mg/dL, $p = 0.45$ and −2.8 mL/min, $p = 0399$ at month 60). A statistical improvement was observed in younger individuals (median change from baseline: 0 mg/dL, $p = 0.05$ and 0 mL/min, $p = 0.09$ at month 36; −0.05 mg/dL, $p = 0.002$ and 5.8 mL/min, $p < 0.001$ at month 60).

Death and HCC Development

Six (1.4%) patients treated with TDF-containing regimens and 6 (3.2%) with ETV died during therapy, 83.3 and 33.3%, respectively, due to liver-related causes. Nineteen (4.5%) TDF patients and 10 (5.4%) ETV were lost during follow-up.

Fourteen (2.29%) patients developed HCC during the study period: 3 given ETV and 11 given TDF. All were men, mean baseline MELD score of 11, and Page-B of 17. As is shown in Fig. 3, most (71%) had cirrhosis at the start of therapy; only four non-cirrhotic patients developed HCC, a statistical difference ($p = 0.003$). Two patients (14%) were HBeAg-positive, 9 (64.3%) had been previously treated, and 4 (28.6%) had undetectable HBV DNA at the start of ETV or TDF. Mean baseline HBV DNA level in the remainder was similar to that of patients who did not develop HCC (4.0 vs. 4.1 log IU/mL, $p = 0.72$). In the 13 patients with available HBV DNA status at the time of the HCC diagnosis, HBV DNA was undetectable in all cases.

Overall, the incidence of HCC within the first 5 years of therapy was 1.5%, 3.6% in patients with cirrhosis and 0.5% in those without. The annual HCC incidence according to cirrhosis status is shown in Table 4.

Performance of the Page-B Score in the CIBERHEP Cohort

During the first 5 years of treatment with either ETV or TDF, there were nine cases of HCC. According to the Cox model coefficients and the detailed survival estimations from Papatheodoridis et al, the Page-B score adapted to the complete time at risk of each patient estimated that 27.8 HCC cases would occur within the 5 years of complete follow-up in the CIBERHEP cohort. The predictability of Page-B score in out cohort was good (*c*-index: 0.732), though lower than in the original validation dataset (*c*-index: 0.82).

Table 3 Renal function during follow-up according to nucleos(t)ide analogue regimen

	Baseline	Month 12	Month 36	Month 60
eGFR, mL/min, mean (SD)				
Median change from baseline, mL/min (range)				
Overall, $N = 611$	88.9	86.6	87.8	88.5
		0 (−83.2 to 76.2)	0 (−79.4 to 97)	4.9 (−65.2 to 73)
Naïve patients, $N = 311$	90.6	88.5	89.6	90.9
		0 (−83.2 to 76.2)	0 (−79.4 to 64.1)	4.2 (−65.2 to 72.9)
Age ≥65 years, $N = 78$	70.6	66.6	68.5	67.3
		−2.7 (−46.6 to 29.8)	0 (−35.2 to 34.8)	−2.8 (−28.6 to 45)
TDF-containing regimens, $N = 424$	90.8	90.3	88.9	85.1
		0 (−71.9 to 48.6)	0 (−49.3 to 55.4)	4.4 (−40 to 51.7)
Age ≥65 years, $N = 52$	74.2	69.2	71.0	70.2
		0 (−16.1 to 29.8)	2.2 (−20.8 to 34.8)	−2.6 (−9.98 to 45)
ETV, $N = 187$	81.2	79.0	84.8	90.7
		0.2 (−83.2 to 76.2)	0 (−79.4 to 97.0)	5.09 (−65.2 to 72.9)
Age ≥ 65 years, $N = 26$	68.3	66.2	63.1	66.4
		−5.5 (−46.6 to 6.6)	−6.6 (−35.2 to 23.1)	−3.0 (−28.6 to 34.9)
Creatinine, mg/dL, mean (SD)				
Median change from baseline, mL/min (range)				
Overall, $N = 611$	0.91	0.9	0.91	0.93
		0 (−1.7 to 1.2)	0 (−1.8 to 0.5)	−0.05 (−1.96 to 0.6)
Naïve patients, $N = 311$	0.9	0.9	0.9	0.91
		0 (−0.38 to 1.15)	0 (−0.39 to 0.5)	−0.05 (−0.43 to 0.6)
Age ≥65 years, $N = 78$	1	1.09	1.05	0.96
		0.03 (−1.7 to 0.42)	−0.01 (−1.8 to 0.5)	0.03 (−1.96 to 0.31)
TDF-containing regimens, $N = 424$	0.9	0.9	0.91	0.95
		0 (−1.7 to 0.57)	0 (−1.8 to 0.5)	−0.05 (−1.96 to 0.25)
Age ≥65 years, $N = 52$	0.97	1.05	1	0.94
		0 (−1.7 to 0.2)	−0.05 (−1.8 to 0.5)	0.03 (−1.96 to 0.12)
ETV, $N = 187$	0.99	1	1	0.9
		0 (−0.3 to 1.15)	0 (−0.47 to 0.36)	−0.05 (−0.6 to 0.63)
Age ≥65 years, $N = 26$	1	1.01	1.01	1.11
		0.08 (−0.1 to 0.42)	0.04 (−0.18 to 0.3)	0.02 (0−06 to 0.3)

eGFR estimated glomerular filtration rate, *ETV* entecavir, *SD* standard deviation, *TDF* tenofovir disoproxil fumarate

One hundred sixty (26.2%) patients presented a high risk of HCC development (Page-B score >17), 215 (35.2%) medium (Page-B 10–17) and 236 (38.6%) low risk (Page-B <10). The 5-year cumulative probability of HCC in subjects with low, medium and high risk based on Page-B score was 0, 2.8 and 5%.

As originally documented in the derivation cohort from the Page-B score, all Caucasian patients receiving ETV or TDF included in the CIBERHEP cohort who developed HCC had a baseline Page-B score ≥10. The exactitude index and predictive values of this cut-off in our cohort of patients were similar to those described in the Page-B cohort (Table 5).

Discussion

This study confirms the high rate of virological and biochemical response of treatment with ETV or TDF-containing regimens observed in the real-world with a large cohort of patients, more than 600 CHB- 428 (70%) and 224 (37%) followed for 36 and 60 months, respectively. The percentage of patients showing HBV DNA < 69 IU/mL and normalized ALT levels during follow-up was similar to that seen in the ETV and TDF registration studies [1, 13], and previous real-world cohorts [5, 6, 14]. In the present study, HBeAg-positive and HBeAg-negative patients were both included, and virological response was faster in

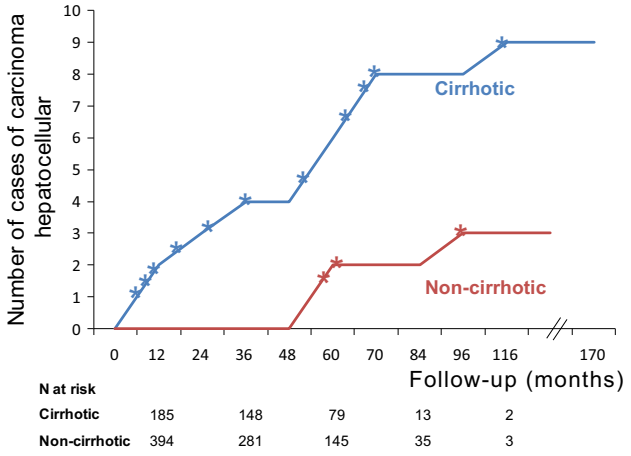

Fig. 3 Observed incidence of hepatocellular carcinoma (HCC) by baseline cirrhosis status in the CIBERHEP Overall, there were 12 cases of HCC, 9 in cirrhotic patients and 3 in non-cirrhotic. *Asterisk* indicates each HCC case and *lines* the cirrhosis status (*blue* for cirrhotic and *red* for non-cirrhotic patients)

Table 4 Annual hepatocellular carcinoma incidence according to cirrhosis status and number of patients at risk

Year	Overall	Cirrhosis	No cirrhosis
1	4/611 (0.7%)	3/197 (1.5%)	1/414 (0.2%)
2	4/506 (0.8%)	3/167 (1.8%)	1/339 (0.3%)
3	5/429 (1.2%)	4/148 (2.7%)	1/281 (0.4%)
4	6/342 (1.8%)	5/116 (4.3%)	1/226 (0.4%)
5	9/224 (4.0%)	7/79 (8.9%)	2/145 (1.4%)

Table 5 Accuracy of Page-B risk score cut-off of ≥10 for predicting the development of hepatocellular carcinoma in the Page-B and CIBERHEP cohorts

	Page-B risk score ≥10	
	Page-B cohort $N = 484$	CIBERHEP cohort $N = 611$
Sensitivity	100%	100%
Specificity	41.2%	25.1%
Positive predictive value	9.8%	3.1%
Negative predictive value	100%	100%

HBeAg-negative, although the difference showed a gradual decrease over time. This trend had been reported in the French cohort [5], but because of our longer follow-up, we found that the observed difference had disappeared at 5 years after the start of therapy. Interestingly, patient age at the beginning of ETV or TDF had an impact on the response to these drugs. Although the number of patients older than 65 years was relatively low ($N = 81$), the percentage achieving HBV DNA < 69 IU/mL either with ETV or TDF was lower than in the rest of patients during the first months of follow-up. However, this percentage was similar by month 24.

Concerning serological response, only 4 patients lost HBsAg during follow-up, a smaller number than in previous European cohorts [5, 6] where 11 of 400 and 14 of 440 patients achieved HBsAg loss. A possible explanation for this finding is the higher percentage of HBeAg-positive patients included in these studies compared to ours. In this regard, only 1 of the 375 (0.3%) HBeAg-negative patients in the TDF registration study who remained on treatment at year 7 achieved HBsAg loss [1].

Long-term follow-up of renal function evaluating serum creatinine levels and the estimated glomerular filtration rate (eGFR) showed no statistical changes in the first 4 years of therapy and a tendency to improvement at year 5. Neither the analogue-based regimens nor previous therapies had an impact on renal function during treatment. In our cohort, only age at the start of therapy seemed to have an influence on renal function, as worsening occurred only in patients older than 65 years. However, it should be noted that our study has an important limitation, since comorbidities were not included in the initial database and therefore were not analyzed in this study. Thus, confounding bias cannot be ruled out since some conditions that can deteriorate renal function such as diabetes and arterial hypertension are more common in older patients. In this line, a recent retrospective study including 3175 CHB patients treated with oral antiviral agents for more than 1 year showed that age, hypertension, and diabetes together with transplantation, underlying chronic kidney disease, and diuretic use were the main risk factors for a renal function decline [15]. Although both ETV and TDF are excreted in urine, only TDF has been associated with nephrotoxicity in post-commercialization studies [16]. However, it should be stressed that most cases of TDF-related nephrotoxicity have been described in HIV-infected patients treated with long-term TDF [17, 18]. No relevant renal events were documented in the registration studies of either TDF or ETV [1, 19–22], whereas in later real-world cohorts, data on a possible impact of TDF on glomerular filtration have been contradictory [23]. Nonetheless, and in line with our findings, Liaw et al. reported no differences in renal function in decompensated patients treated with TDF, TDF plus emtricitabine, or ETV [24]. Moreover, another study including 80 patients treated with a TDF-containing regimen and 80 with ETV, both stratified by age, diabetes, and prior transplantation to avoid bias, ruled out differences in renal function between the two groups [25].

To assess and validate the Page-B score [4], only Caucasian patients were included in this study. Fourteen (2.29%) patients developed HCC during follow-up, and

most of them (71%) had underlying liver cirrhosis. This value is lower than that reported for the Page-B cohort, in which the incidence of HCC was 3.8% in the first 5 years of follow-up, the total follow-up time in the study. Of note, in the CIBERHEP cohort, the estimated number of HCC cases based on the baseline Page-B score and the individual completed time at risk was 27.8. This discrepancy cannot be explained by differences in the baseline characteristics, since follow-up (median 50 months), patient age (52 years), percentage with cirrhosis (20%) and HBeAg-positive status (16%) was similar in the two cohorts. However, in contrast to the Page-B cohort [4], in which all patients underwent regular ultrasound study (every 6 months in cirrhotic and every 12 months in non-cirrhotic patients), HCC surveillance in the CIBERHEP cohort was carried out at the discretion of the investigators at each site. Thus, these differences between the Page-B and CIBERHEP cohorts highlight the importance of periodical HCC surveillance even in patients who have achieved virological suppression. In our study, all patients with available viral load at the time HCC developed had undetectable HBV DNA. A Page-B cut-off $\geq$10 may be useful for selecting patients who will benefit from HCC surveillance. In agreement with the results from the original cohort, all our patients who developed HCC had a baseline Page-B score above this value, making the negative predictive value 100%.

The main limitation of this study is that not all the patients included completed the 5-year follow-up leading to a lower number of HCC cases in our cohort in comparison with the expected by the Page-B model. In addition, baseline comorbidities, which can introduce bias in the analysis regarding the impact of age on renal function during NUC therapy, were not collected or included in the analysis.

In conclusion, in this real-world cohort of CHB patients treated with ETV- or TDF-containing regimens during a mean follow-up of 52 months, high virological and biochemical response rates were found, with a low incidence of drug discontinuations and deaths. Long-term analysis of renal function showed a tendency to improvement over time with both drugs, except in patients older than 65 years at the start of therapy. Application of the Page-B score in this cohort indicated that a cut-off score $\geq$ 10 identifies patients at risk of developing HCC within the first 5 years of ETV or TDF therapy.

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Compliance with ethical standards

Conflict of interest Mar Riveiro-Barciela has received grants from Gilead. David Tabernero has served as speaker for Gilead. Sabela Lens has served as speaker for Gilead, Janssen and Abbvie. Jose L. Calleja has served as speaker and consultant for Gilead, BMS, MSD and Abbvie. Javier García-Samaniego has served as speaker and consultant for Gilead and BMS. Maria Buti has served as speaker for Gilead and BMS. The rest of authors declare that they have no conflict of interest.

References

1. Buti M, Tsai N, Petersen J, et al. Seven-year efficacy and safety of treatment with tenofovir disoproxil fumarate for chronic hepatitis B virus infection. *Dig Dis Sci*. 2015;60:1457–1464.
2. Marcellin P, Gane E, Buti M, et al. Regression of cirrhosis during treatment with tenofovir disoproxil fumarate for chronic hepatitis B: a 5-year open-label follow-up study. *Lancet*. 2013;381:468–475.
3. 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.
4. Papatheodoridis G, Dalekos G, Sypsa V, et al. PAGE-B: a risk score for hepatocellular carcinoma in Caucasians with chronic hepatitis B under a 5-year entecavir or tenofovir therapy. *J Hepatol*. 2016;64(4):800–806.
5. Marcellin P, Zoulim F, Hezode C, et al. Effectiveness and safety of tenofovir disoproxil fumarate in chronic hepatitis B: a 3-year, prospective, real-world study in France. *Dig Dis Sci*. 2016;61(10):3072–3083.
6. Petersen J, Heyne R, Mauss S, et al. Effectiveness and safety of tenofovir disoproxil fumarate in chronic hepatitis B: a 3-year prospective field practice study in Germany. *Dig Dis Sci*. 2016;61(10):3061–3071.
7. Idilman R, Gunsar F, Koruk M, et al. Long-term entecavir or tenofovir disoproxil fumarate therapy in treatment-naive chronic hepatitis B patients in the real-world setting. *J Viral Hepat*. 2015;22:504–510.
8. Tsai MC, Chen CH, Tseng PL, et al. Comparison of renal safety and efficacy of telbivudine, entecavir and tenofovir treatment in chronic hepatitis B patients: real world experience. *Clin Microbiol Infect*. 2016;22:95–e1–7.
9. Levey AS, Coresh J, Greene T, et al. Using standardized serum creatinine values in the modification of diet in renal disease study equation for estimating glomerular filtration rate. *Ann Intern Med*. 2006;145:247–254.
10. European Association For The Study Of The Liver. EASL clinical practice guidelines: management of chronic hepatitis B virus infection. *J Hepatol*. 2012;57:167–185.
11. Buti M, Garcia-Samaniego J, Prieto M, et al. Consensus document of the Spanish Association for the Study of the Liver on the treatment of hepatitis B infection (2012). *Gastroenterol Hepatol*. 2012;35:512–528.
12. European Association For The Study Of The Liver. EASL-EORTC clinical practice guidelines: management of hepatocellular carcinoma. *Eur J Cancer*. 2012;48:599–641.
13. Ono A, Suzuki F, Kawamura Y, et al. Long-term continuous entecavir therapy in nucleos(t)ide-naive chronic hepatitis B patients. *J Hepatol*. 2012;57:508–514.
14. Ahn J, Lee HM, Lim JK, et al. Entecavir safety and effectiveness in a national cohort of treatment-naive chronic hepatitis B patients in the US—the ENUMERATE study. *Aliment Pharmacol Ther*. 2016;43:134–144.
15. Shin JH, Kwon HJ, Jang HR, et al. Risk factors for renal functional decline in chronic hepatitis B patients receiving oral antiviral agents. *Medicine*. 2016;95:e2400.
16. Viread: EPAR- product information. http://www.ema.europa.eu/ema/indexjspcurl=pagesmedicines/human/medicines/000419/

human_med_001144.jsp&mid=WC0b01ac058001d124. Accessed 19 March 2013.
17. Fontana RJ. Side effects of long-term oral antiviral therapy for hepatitis B. *Hepatology*. 2009;49:S185–S195.
18. Gupta SK. Tenofovir-associated Fanconi syndrome: review of the FDA adverse event reporting system. *AIDS Patient Care STDs*. 2008;22:99–103.
19. Marcellin P, Heathcote EJ, Buti M, et al. Tenofovir disoproxil fumarate versus adefovir dipivoxil for chronic hepatitis B. *N Engl J Med*. 2008;359:2442–2455.
20. Martinot-Peignoux M, Carvalho-Filho R, Lapalus M, et al. Hepatitis B surface antigen serum level is associated with fibrosis severity in treatment-naive, e antigen-positive patients. *J Hepatol*. 2013;58:1089–1095.
21. Yuen MF, Seto WK, Fung J, Wong DK, Yuen JC, Lai CL. Three years of continuous entecavir therapy in treatment-naive chronic hepatitis B patients: VIRAL suppression, viral resistance, and clinical safety. *Am J Gastroenterol*. 2011;106:1264–1271.
22. Xu Y, Zhang YG, Wang X, et al. Long-term antiviral efficacy of entecavir and liver histology improvement in Chinese patients with hepatitis B virus-related cirrhosis. *World J Gastroenterol*. 2015;21:7869–7876.
23. Lampertico P, Chan HL, Janssen HL, Strasser SI, Schindler R, Berg T. Review article: long-term safety of nucleoside and nucleotide analogues in HBV-monoinfected patients. *Aliment Pharmacol Ther*. 2016;44:16–34.
24. Liaw YF, Sheen IS, Lee CM, et al. Tenofovir disoproxil fumarate (TDF), emtricitabine/TDF, and entecavir in patients with decompensated chronic hepatitis B liver disease. *Hepatology*. 2011;53:62–72.
25. Gish RG, Clark MD, Kane SD, Shaw RE, Mangahas MF, Baqai S. Similar risk of renal events among patients treated with tenofovir or entecavir for chronic hepatitis B. *Clin Gastroenterol Hepatol*. 2012;10:941–946. **(quiz e968)**.