Research Article

Risk of relapse after treatment discontinuation

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Risk factors for relapse after discontinuation of tenofovir or entecavir in hepatitis B e antigen-negative patients

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Abstract

Background and Aim: This study aimed to define the relapse frequency and risk determinants in chronic hepatitis B (CHB) patients who discontinued nucleoside analog (NA) treatment, were HBeAg-negative, and had achieved both a virological and biochemical response.

Materials and Methods: This retrospective cohort study reviewed patients with HBeAg-negative CHB who received antiviral therapy for at least 65 months between January 1, 2013, and December 31, 2020. These patients discontinued treatment after demonstrating a biochemical and virological response and were evaluated at 6, 12, and 24 months post-treatment discontinuation.

Results: Sixty-seven patients with CHB who received NA therapy for at least 65 months, discontinued treatment, and had undetectable HBV DNA and normal ALT values were evaluated. After cessation of NA therapy, a relapse was observed in 38 patients (56.7%). The relapse rate was 71.0% in patients treated with tenofovir disoproxil fumarate (TDF) as the last NA type and 37.9% in patients treated with entecavir (ETV) (p=0.017). The cutoff value for the best estimate of age for predicting relapse was 42 years. The relapse rate was 69.2% in patients aged ≥42 years and 39.2% in patients aged <42 years (p=0.007). The relapse rate was 51.3% in patients with a pre-treatment fibrosis score of 2, 56.0% in those with a fibrosis score of 3, and 100% in those with a fibrosis score of 4 (p=0.089).

Conclusion: Among HBeAg-negative CHB patients who achieved a virological and biochemical response to long-term antiviral therapy, those aged 42 years and older, those with high fibrosis scores before treatment, and those who used TDF before treatment cessation should be closely monitored for relapse, especially in the first 12 months after stopping NA treatment.

Keywords: Hepatitis B; discontinued treatment; relapse.

Introduction

The World Health Organization estimated that as of 2019, 296 million people worldwide were living with chronic hepatitis B (CHB) infection, and only 6.6 million of these patients were on treatment.[1] The main purpose of CHB infection treatment is to prevent potential negative consequences of the disease, such as cirrhosis, hepatic decompensation, hepatocellular carcinoma (HCC), and death.[2] In current guidelines, first-line therapy for CHB includes nucleoside analogues (NA) tenofovir disoproxil fumarate (TDF), tenofovir alafenamide (TAF), and entecavir (ETV), noted for their high barrier to resistance and appropriate safety profiles, as well as leading to long-term undetectable HBV DNA levels in compliant patients.[2,3] Covalently closed circular (CCC) DNA, defined as the stable form of HBV-DNA, is not eradicated by these treatment regimens. Therefore, long-term NA therapy, which suppresses HBV and provides regression of histological activity and fibrosis, is currently the best therapeutic strategy to prevent adverse outcomes such as HCC and cirrhosis.[2,4,5] However, this disease, requiring long-term treatment, raises concerns regarding drug compliance, side effects, and costs.[5] For this reason, the concept of NA discontinuation emerges as a strategy to consider when treatment according to the recommended guidelines does not provide further benefit.[6]

The ideal endpoint in the treatment of CHB is the confirmation of sustained hepatitis B surface antigen (HBsAg) seroclearance in non-cirrhotic patients, with or without anti-HBs seroconversion.[2,3,7] Although this is associated with favorable clinical outcomes, it is observed in long-term follow-up, and the annual seroclearance rate in patients treated with NA is only 0.33%.[7] Therefore, other endpoints were defined in the efficacy evaluation, such as HBV suppression (virological remission), normalization of alanine aminotransferase (ALT) level (biochemical remission), and hepatitis B e antigen (HBeAg) seroconversion. Achieving these endpoints has also been shown to result in histological improvement and/or regression of liver fibrosis.

The European Association for the Study of the Liver (EASL) 2017 guideline proposed that, apart from HBsAg seroclearance, NA can be discontinued under certain conditions if close follow-up can be guaranteed. It is suggested that NA can be discontinued in non-cirrhotic HBeAg-positive CHB patients who achieve HBeAg seroconversion and undetectable HBV DNA levels and complete at least 12 months of consolidation therapy.[2] In non-cirrhotic HBeAg-negative CHB patients, it is stated that NA therapy can be discontinued after at least 2 years of treatment and after ≥3 years of virological suppression.[2,6] The American Association for the Study of Liver Diseases (AASLD) 2018 guideline, on the other hand, states that

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despite a lack of sufficient and conclusive evidence, treatment discontinuation may be considered only in non-cirrhotic patients who have demonstrated loss of HBsAg. It is also recommended that individuals who discontinue NA therapy be followed closely every 3 months for at least 1 year for virological relapses, ALT flares, and clinical decompensation.[3]

In recent years, studies have emerged to evaluate the potential benefits, safety, and effectiveness of discontinuing long-term NA therapy.[6] Several studies have shown that discontinuation of HBeAg-negative CHB therapy may promote a decrease in HBsAg and even loss of HBsAg in some patients, possibly by inducing immune responses.[8–10] Although there is an ongoing debate on this issue, in real-life practice, discontinuation of long-term NA therapy should be considered if better or similar results to its continuation are anticipated.[6] Therefore, it is crucial to identify the predictors of relapse or sustained remission in patients who discontinue long-term NA therapy to determine which patients should not cease treatment. Accordingly, our study aims to define the relapse frequency and risk determinants in CHB patients who discontinued NA therapy, were HBeAg-negative, and had achieved a virological and biochemical response.

Materials and Methods

This retrospective cohort study was conducted in a tertiary center in Southern Anatolia, where the prevalence of HBV is higher than in the rest of the country. After reviewing HBeAg-negative CHB patients who received antiviral therapy for at least 65 months between January 1, 2013, and December 31, 2020, discontinued treatment, and had a biochemical and virological response after discontinuation, a total of 67 patients with available data were identified. The biochemical response was defined as ALT within normal limits (<40 U/L), and the virological response was defined as undetectable HBV DNA.[11]

Age, gender, place of residence, date of the first diagnosis, presence of concomitant Anti HCV, Anti Delta, steatohepatitis, liver histology (fibrosis and HAI), baseline HBV DNA, ALT level, date of initiation of treatment, antiviral treatment given, treatment change, if any, total treatment time, and negative viral load durations of the patients included in the study were recorded. Demographic and clinical data were obtained from patients' electronic medical records.

Patients who discontinued antiviral therapy were followed monthly for the first six months and then every three months. Patients with clinical relapse were re-treated according to the severity of the relapse. ALT, HBV DNA, total bilirubin, and INR levels at 6, 12, and 24 months after discontinuation of treatment, development of any relapse or flare, and any re-initiation of therapy were recorded. In our study, relapse was defined as an increase in HBV DNA above 2000 IU/mL; ALT elevation was defined as higher than 1.2 times normal; flare was defined as HBV-DNA >2000 IU/mL and ALT >ULN x 10.

All collected blood samples were tested on the same regularly calibrated analyzer. Biochemical tests were performed at the hospital's central laboratories using routine automated techniques. Serum hepatitis markers, including HBsAg, HBsAg antibody, HBeAg, HBeAg antibody, anti-hepatitis D virus, and anti-hepatitis C virus, were tested using the Enzyme Immunoassay kit (Abbott Diagnostics, North Chicago, IL). Serum HBV DNA was assayed using the Roche Cobas Amplicor TaqMan HBV Monitor assay (detection limit, 20 IU/mL; Roche Diagnostics, Pleasanton, CA). Patients whose file records could not be reached were excluded from the study.

Statistical Analysis

All tests were analyzed using the statistical package SPSS, version 21.0 (SPSS Inc, an IBM Company). Continuous variables were presented as median (interquartile range [IQR]). Categorical variables were summarized as frequency and percentage. The Chi-square or Fisher's exact test was used to test the relationship between two categorical variables. Non-parametric comparisons between two groups were made using the Mann-Whitney test. The Receiver-operating characteristic (ROC) curve was used to evaluate the predictive power of age for relapse, and the Youden Index (J = Sensitivity + Specificity – 1) was used to determine the cut-off point. Cumulative relapse rate estimates were obtained using the Kaplan-Meier method. The Log-Rank method was used to compare the Kaplan-Meier curves. Cox proportional hazard regression models were used to estimate the risk of relapse for some variables. To identify independent prognostic factors, a multivariate Cox regression model was created with significant associations in univariate analyses and age, pre-treatment fibrosis score, and last NA type before treatment discontinuation. Factors with significance or trends in univariate analyses ($p<0.10$) were used in this model. The hazard ratio (HR) and their 95% confidence intervals (CI) are presented together with the corresponding p-values. A p-value of <0.05 was considered statistically significant.

Results

Patients' Main Characteristics

After stopping the treatment, 67 CHB patients who had received NA treatment for at least 65 months were negative for serum HBV DNA, serum HBeAg, Anti-HCV, and Delta antibody, and had normal ALT values. Their median age was 44 (range: 29 – 69), and 36 (53.7%) were male. The main characteristics of the patients are presented in Table 1. Initial NA therapy was changed in 7 (10.4%) patients. After initial treatments, two (3.0%) patients were switched from ETV, ADF, and 3TC to TDF, and one (1.5%) switched from TDF to ETV. The last type of NA used before cessation was ETV in 43.3% and TDF in 56.7% of patients. The median duration of treatment with NA was 78 months, the duration of virological remission during treatment was 69 months, and the minimum duration of virological remission was 61 months.

Virological and Biochemical Relapse

The frequencies of relapses at 6 and 12 months according to the HBV DNA cutoff points of 2,000–20,000 IU/mL and >20,000 IU/mL and the upper limit of the ALT value are presented in Figure 1. After cessation of NA, relapse was observed in 38 (56.7%) patients. After discontinuation of NA, relapse developed at 6 months in 22 (32.8%) patients and at 12 months in 16 (23.9%) patients. No virological and biochemical relapses were observed in the 24-month follow-up. Virological relapse occurred mostly at the defined cutoff point of HBV DNA 2000-20000 IU/mL. At both 6 and 12 months after treatment cessation, ALT was 1–2 times higher than normal in those with HBV DNA of 2,000-20,000 IU/mL and 10 times above normal in those with HBV DNA >20,000 IU/mL. In 3 patients who developed flare with ALT >10x ULN and HBV DNA >20,000 IU/mL, total bilirubin was between 2–10 mg/dL and INR was high. Re-treatment was started in all patients who developed relapse. Virological and biochemical remission was achieved at 12 months in all patients who relapsed at 6 months, and at 24 months in all patients who relapsed at 12 months.

Table 1. Main characteristics of CHB patients who discontinued effective and long-term NA therapy

Initiation of nucleos(t)ide analogue therapy	
Age (years), median (IQR)	44 (38–51)
Gender (male), n (%)	36 (53.7)
ALT (U/L), n (%)	
Normal	15 (22.4)
>1–2 x ULN	19 (28.4)
>2 x ULN	33 (49.3)
HBV-DNA (IU/ml), n (%)	
>2 million	10 (14.9)
2 thousand – 20 thousand	16 (23.9)
20 thousand – 2 million	41 (61.2)
Presence of steatohepatitis, n (%)	14 (20.9)
Fibrosis score, median (IQR)	2 (2–3)
Histological activity index, median (IQR)	7 (7–8)
Initial NA treatment, n (%)	
Adefovir	2 (3.0)
Lamivudine	2 (3.0)
Tenofovir disoproxil fumarate	30 (44.8)
Entecavir	33 (49.3)
Change in initial therapy, n (%)	7 (10.4)
Last NA before treatment discontinuation, n (%)	
Tenofovir disoproxil fumarate	38 (56.7)
Entecavir	29 (43.3)
NA treatment duration (months), median (IQR)	78 (73–81)
Virological remission duration during NA treatment (months), median (IQR)	69 (64–73)

ALT: Alanine aminotransferase; NA: Nucleos(t)ide analogue; IQR: Interquartile range.

Relapse Indicators

In univariate Cox regression analyses, a higher fibrosis score before NA treatment (p=0.043) and use of TDF as the last NA treatment before discontinuation (p=0.017) were found to be significantly associated with the risk of relapse. There was a trend towards a significant relationship with older age (p=0.066). A multivariate Cox regression model was established with age, pre-treatment fibrosis score, and last NA type before discontinuation of treatment. In this model, there was no independent risk factor, but there was a significant trend between the probability of relapse and use of TDF as the last NA treatment before discontinuation (p=0.070) (Table 2).

At 24 months after discontinuing treatment, relapse was observed in 71.0% of patients with TDF as the last NA type and in 37.9% of patients with ETV (p=0.012) (Fig. 2a). In the ROC analysis performed to evaluate the predictive power of age for relapse, the best predictive power was 0.611 (95% CI; 0.529–0.792; p=0.025), and the cutoff value was 42 years, with 71.0% sensitivity and 62.0% specificity. The relapse rate at the end of 24 months was 69.2% in patients aged ≥42 years and 39.2% in patients aged <42 years (p=0.007) (Fig. 2b). At the end of 24 months, the relapse rate was 51.3% in patients with a pre-treatment fibrosis score of 2, 56.0% in those with a fibrosis score of 3, and 100% in those with a fibrosis score of 4 (p=0.089) (Fig. 2c).

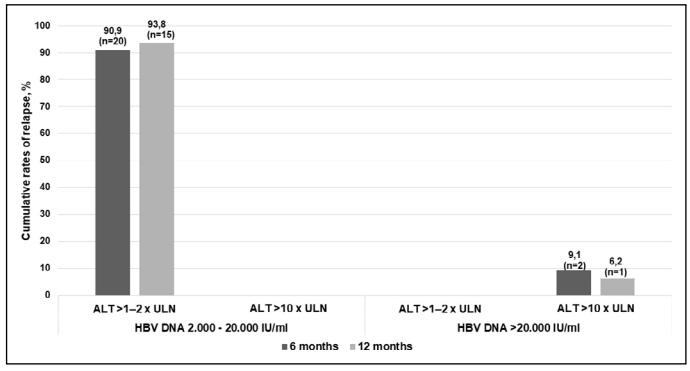

Figure 1. Frequencies of virological relapses according to HBV DNA cut-off points at 6 and 12 months after discontinuation of NA treatments.

Discussion

In the management of CHB, no definite duration of NA treatment has yet been determined for patients with long-term virological suppression due to concern for HBV relapse.[12,13] Studies evaluating the frequency of virological relapses after NA discontinuation in CHB patients who achieved a virological response with long-term treatment have reported variable cutoff values for relapse. In the literature, studies that accept the lowest value at which HBV-DNA is detected as the limit to define virological relapse have shown more than 80% virological relapse.[12,14] In one of the two previous systematic reviews, the virological remission rate at 12 months after discontinuation of therapy in HBeAg-negative patients was 43.7%, while the virological relapse rate was <70% and the clinical relapse rate was 50% in the other one.[15,16] Similar to our study, a prospective observational study in which virological relapse was defined as HBV DNA >2000 IU/mL in HBeAg-negative patients reported virological relapse in approximately half of the patients within 24 months after discontinuation of therapy, despite long-term viral suppression.[17] The majority of relapses occur within the first year after discontinuation of treatment.[16,18] This is consistent with our findings that all relapses occurred within the first 12 months during our 24-month follow-up study.

In the literature, one determinant of relapse after discontinuation of treatment in CHB patients with long-term virological suppression is the last type of NA used.[13] In our study, the median NA treatment duration was 78 months, and the frequency of virological and biochemical relapses 24 months after cessation of treatment was lower in those who received ETV as the last treatment compared to those who received TDF. In two previous studies by Jeng WJ et al.,[18,19] the frequency of clinical relapse within 1 year after discontinuation of ETV treatment was 45.3%, while it was higher (52%) in TDF, as in our study. It has been shown that the timing of clinical and virological relapses after TDF cessation in HBeAg-negative patients is much earlier than after ETV cessation.[13,20] In our study, although no statistically significant independent risk factor was found in the multivariate analysis, receiving TDF treatment increased the risk of recurrence by 1.97 times, which tended to be statistically significant. The mechanism underlying the different relapse frequencies between ETV and TDF discontinuation has not been clearly explained. The different relapse behavior of NAs has been attributed to the different immunomodulatory abilities of these therapies.[13,20,21]

Several studies in HBeAg-negative CHB patients after discontinuation of NAs have found lower relapse rates in younger patients, suggesting age as the sole predictive factor for relapse.[9,21] In our study, although

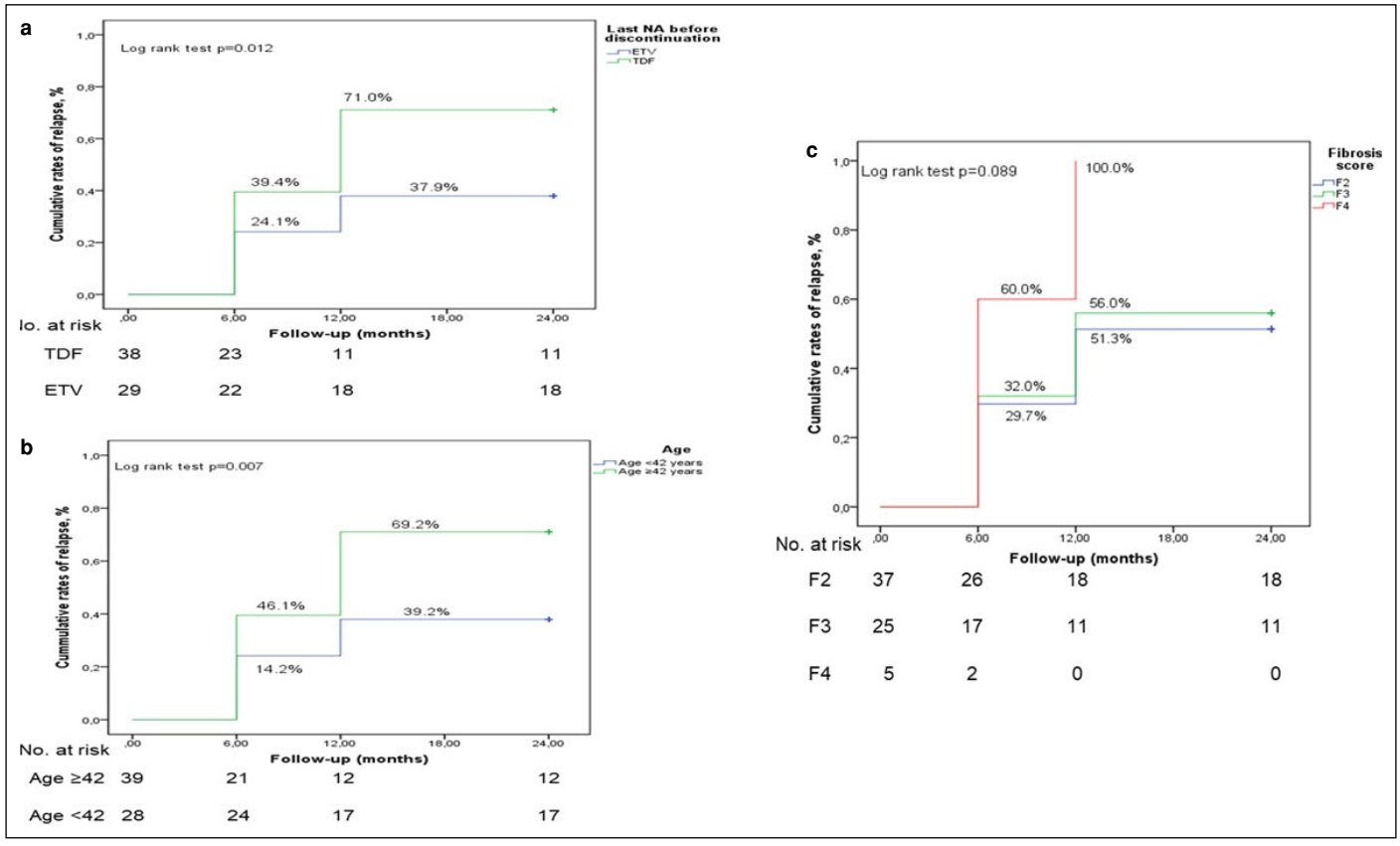

Figure 2. Relapse rates after discontinuation of the nucleos(t)ide analogue in chronic hepatitis B patients; **(a)** last NA treatment type, **(b)** age and **(c)** pre-treatment fibrosis score.

Table 2. Regression analysis of cumulative relapse with cox proportional hazards

	Cumulative relapse (n=36)			
	Univariate analysis HR (95% CI)	***P**	**Multivariate analysis HR (95% CI)**	**P***
Initiation of nucleos(t)ide analogue therapy				
Age (Per year increase)	1.032 (0.998–1.067)	0.066	1.027 (0.992–1.064)	0.128
Gender, male vs female	1.314 (0.685–2.521)	0.412		
Steatohepatitis, yes vs no	1.641 (0.794–3.392)	0.181		
Fibrosis score (Per score increase)	1.737 (1.019–2.961	0.043	1.498 (0.869–2.582)	0.146
Histological activity index (Per index increase)	0.850 (0.682–1.058)	0.145		
ALT >1-2 X ULN vs Normal ALT	0.806 (0.349–1.864)	0.615		
ALT ≥ 2 X ULN vs Normal ALT	0.672 (0.309–1.461)	0.316		
HBV DNA 20 thousand-2 million vs 2 thousand–20 thousand	1.091 (0.489–2.435)	0.831		
HBV DNA >2 million vs 2 thousand – 20 thousand	1.055 (0.361–3.083)	0.922		
Last type of NA before treatment discontinuation, TDF vs ETV	2.358 (1.163–4.779)	0.017	1.973 (0.947–4.111)	0.070
NA treatment duration (months)	0.965 (0.922–1.009)	0.121		
Virological remission duration during NA treatment (months)	1.000 (0.952–1.051)	0.992		

ALT: Alanine aminotransferase; NA: Nucleos(t)ide analogue; TDF: Tenofovir disoproxil fumarate; ETV: Entecavir; CI: Confidence interval; *: P<0.05.

age was not an independent indicator for relapse, it was found that older age had a significant relationship trend. In a study conducted in the Asia-Pacific, where CHB infection develops mostly through perinatal transmission, more than three-quarters of patients aged 25 years and older developed relapse after NA discontinuation, and in HBeAg-negative CHB patients, 25 years of age was accepted as a cutoff value for

sustained response.[22] A longer period of HBV infection in the elderly was thought to affect persistence by complicating viral clearance. In addition, it was noted that younger patients may have a stronger immune system, which may have an impact on the eventual eradication of the virus.[9,21] In our study, this cutoff value was determined as 42 years, and the rate of relapse was found to be higher at the end of 24 months in those aged ≥42 years. The higher age limit determined in our study compared to previous studies may be related to intrafamilial transmission as well as vertical transmission of CHB infection in the region where our study was conducted.[23]

One of the main reasons for the renewed interest in discontinuing antiviral therapy is the potential for HBsAg loss. An observational study found a very high HBsAg seroclearance rate in HBeAg-negative patients during a 5-year follow-up after ceasing adefovir therapy, though this could not be demonstrated in subsequent studies.[9] However, it remains controversial whether the rate of HBsAg loss is higher after discontinuation of therapy than with continuous therapy.[6] After a mean follow-up of 3 years, a randomized trial reported a significantly greater loss of HBsAg and reduction in quantitative HBsAg in patients who discontinued treatment compared to those who continued treatment.[10] On the other hand, another study showed that the rate of HBsAg loss was approximately 1% in those who stopped or continued treatment, and there was no difference between the groups. However, it is noteworthy that the median post-treatment follow-up period (72 months) in this study was shorter.[24] Follow-up time may explain the different results in these studies. In our study, HBsAg loss was not observed in any patient during the 24-month follow-up period. Although evaluation with longer follow-up times is needed, this finding may also be related to the limited benefit of discontinuing NA therapy.

In patients with cirrhosis, treatment discontinuation is not recommended under any circumstances before HBsAg loss due to the risk of liver decompensation.[2,3] Besides, in a study conducted among HBeAg-negative patients with compensated cirrhosis who were treated with ETV for at least 12 months and achieved HBV DNA suppression, there was no significant difference between those who continued and discontinued treatment in terms of liver-related complications or death after a median 59-month follow-up.[25] Based on these studies originating from the Asia-Pacific, the APASL (The Asian Pacific Association for the Study of the Liver) guideline stated that possible discontinuation of NA therapy before HBsAg seroclearance may be considered in patients with compensated cirrhosis, provided a careful non-treatment monitoring plan.[11] Another study with an 18-month follow-up period reported that among HBeAg-negative CHB patients who received treatment for 4 years or more, the rate of not re-initiating treatment was higher in patients with pre-treatment fibrosis score ≤ 3 compared to those with a fibrosis score of 4 (cirrhosis). In addition, an independent relationship was demonstrated between the severity of fibrosis score and the need for re-treatment.[26] In our study, all patients with a baseline fibrosis score of 4 developed relapses at 12 months, and treatment was re-initiated. None of these 5 patients died or developed liver decompensation.

All of the patients included in our study were HBeAg-negative and had positive Anti-HBe titres. The absence of HBeAg-positive patients in the study group is a limitation of our study. In addition, HBsAg levels and other promising predictors such as anti-HBc, HBV RNA, and HBcrAg were not evaluated in this study. Finally, cessation of NA treatment was assumed to be a trigger for re-induction of the T cell response against hepatocytes, and no immunological evaluation was performed to define this interaction.

Conclusion

In conclusion, among HBeAg-negative CHB patients who achieved a virological and biochemical response with long-term antiviral therapy, those aged 42 years and older, those with high fibrosis scores before treatment, and those who used TDF before treatment cessation should be closely followed for relapse, especially in the first 12 months after stopping NA treatment.

Ethics Committee Approval: The Dicle University Clinical Research Ethics Committee granted approval for this study (date: 21.01.2022, number: 77).

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References

1. World Health Organization. Hepatitis B [Available from: https://www.who.int/news-room/fact-sheets/detail/hepatitis-b.] Accessed on March 02, 2022.
2. European Association for The Study of The Liver. EASL 2017 Clinical Practice Guidelines on the management of hepatitis B virus infection. J Hepatol. 2017;67(2):370-398.
3. Terrault NA, Lok AS, McMahon BJ, Chang KM, Hwang JP, Jonas MM, et al. Update on prevention, diagnosis, and treatment of chronic hepatitis B: AASLD 2018 hepatitis B guidance. Hepatology 2018;67(4):1560-1599.
4. Marcellin P, Gane E, Buti M, Afdhal N, Sievert W, Jacobson IM, 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(9865):468-475. [CrossRef]
5. Aoki FY. 46-Antivirals against Herpes Viruses. John E. Bennett RD, Martin J. Blaser, editor. Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases. Philadelphia: Elsevier Health Sciences.; 2020. p. 577-595.
6. Tout I, Lampertico P, Berg T, Asselah T. Perspectives on stopping nucleos (t) ide analogues therapy in patients with chronic hepatitis B. Antiviral Res 2021;185:104992. [CrossRef]
7. Kim G-A, Lim Y-S, An J, Lee D, Shim JH, Kim KM, et al. HBsAg seroclearance after nucleoside analogue therapy in patients with chronic hepatitis B: clinical outcomes and durability. Gut 2014;63(8):1325-1332. [CrossRef]
8. Höner zu Siederdissen C, Hui AJ, Sukeepaisarnjaroen W, Tangkijvanich P, Su WW, Nieto GEG, et al. Contrasting timing of virological relapse after discontinuation of tenofovir or entecavir in hepatitis b e antigen–negative patients. J Infect Dis 2018;218(9):1480-1484. [CrossRef]
9. Hadziyannis SJ, Sevastianos V, Rapti I, Vassilopoulos D, Hadziyannis E. Sustained responses and loss of HBsAg in HBeAg-negative patients with chronic hepatitis B who stop long-term treatment with adefovir. Gastroenterology 2012;143(3):629-636. e1. [CrossRef]
10. Berg T, Simon K-G, Mauss S, Schott E, Heyne R, Klass DM, et al. Long-term response after stopping tenofovir disoproxil fumarate in non-cirrhotic HBeAg-negative patients–FINITE study. J Hepatol 2017;67(5):918-924. [CrossRef]
11. Sarin S, Kumar M, Lau G, Abbas Z, Chan H, Chen C, et al. Asian-Pacific clinical practice guidelines on the management of hepatitis B: a 2015 update. Hepatol Int 2016;10(1):1-98. [CrossRef]
12. Sohn HR, Min BY, Song JC, Seong MH, Lee SS, Jang ES, et al. Off-treat-

ment virologic relapse and outcomes of re-treatment in chronic hepatitis B patients who achieved complete viral suppression with oral nucleos (t) ide analogs. BMC Infect Dis 2014;14(1):1-8. [CrossRef]

13. Kuo MT, Hu TH, Hung CH, Wang JH, Lu SN, Tsai KL, et al. Hepatitis B virus relapse rates in chronic hepatitis B patients who discontinue either entecavir or tenofovir. Aliment Pharmacol Ther 2019;49(2):218-228. [CrossRef]
14. Shouval D, Lai C-L, Chang T-T, Cheinquer H, Martin P, Carosi G, et al. Relapse of hepatitis B in HBeAg-negative chronic hepatitis B patients who discontinued successful entecavir treatment: the case for continuous antiviral therapy. J Hepatol 2009;50(2):289-295. [CrossRef]
15. Chang ML, Liaw YF, Hadziyannis S. Systematic review: cessation of long-term nucleos (t) ide analogue therapy in patients with hepatitis B e antigen-negative chronic hepatitis B. Aliment Pharmacol Ther 2015;42(3):243-257.
16. Papatheodoridis G, Vlachogiannakos I, Cholongitas E, Wursthorn K, Thomadakis C, Touloumi G, et al. Discontinuation of oral antivirals in chronic hepatitis B: a systematic review. Hepatology 2016;63(5):1481-1492.
17. Cao J, Chi H, Yu T, Li Z, Hansen BE, Zhang X, et al. Off-treatment hepatitis B virus (HBV) DNA levels and the prediction of relapse after discontinuation of nucleos (t) ide analogue therapy in patients with chronic hepatitis B: a prospective stop study. J. Infect. Dis. 2017;215(4):581-589. [CrossRef]
18. Jeng W-J, Chen Y-C, Sheen I-S, Lin C-L, Hu T-H, Chien R-N, et al. Clinical relapse after cessation of tenofovir therapy in hepatitis B e antigen–negative patients. Clin Gastroenterol Hepatol 2016;14(12):1813-1820. e1. [CrossRef]
19. Jeng WJ, Sheen IS, Chen YC, Hsu CW, Chien RN, Chu CM, et al. Off-therapy durability of response to entecavir therapy in hepatitis B e antigen-negative chronic hepatitis B patients. Hepatology 2013;58(6):1888-1896. [CrossRef]
20. Su T-H, Yang H-C, Tseng T-C, Liou J-M, Liu C-H, Chen C-L, et al. Distinct relapse rates and risk predictors after discontinuing tenofovir and entecavir therapy. J Infect Dis 2018;217(8):1193-1201. [CrossRef]
21. Jiang Y, Li W, Yu L, Liu J, Xin G, Yan H, et al. Enhancing the antihepatitis B virus immune response by adefovir dipivoxil and entecavir therapies. Cell Mol Immunol 2011;8(1):75-82. [CrossRef]
22. Ha M, Zhang G, Diao S, Lin M, Sun L, She H, et al. A prospective clinical study in hepatitis B e antigen-negative chronic hepatitis B patients with stringent cessation criteria for adefovir. Arch Virol 2012;157(2):285-290. [CrossRef]
23. Bilge B. Prevalence and interfamily transmission pathways of hepatitis B virus. Akademik Gastroenteroloji Dergisi 2017;16(1):12-17. [Turkish]
24. Liem KS, Fung S, Wong DK, Yim C, Noureldin S, Chen J, et al. Limited sustained response after stopping nucleos (t) ide analogues in patients with chronic hepatitis B: results from a randomised controlled trial (Toronto STOP study). Gut 2019;68(12):2206-2213. [CrossRef]
25. Chen Y, Peng C-Y, Jeng W, Chien R, Liaw Y. Clinical outcomes after interruption of entecavir therapy in HB eAg-negative chronic hepatitis B patients with compensated cirrhosis. Aliment Pharmacol Ther 2015;42(10):1182-1191. [CrossRef]
26. Papatheodoridis GV, Rigopoulou EI, Papatheodoridi M, Zachou K, Xourafas V, Gatselis N, et al. DARING-B: discontinuation of effective entecavir or tenofovir disoproxil fumarate long-term therapy before HBsAg loss in non-cirrhotic HBeAg-negative chronic hepatitis B. Antivir Ther 2018;23(8):677-685. [CrossRef].