Case Report

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A case of HBV-induced liver failure in the REEF-2 phase II trial: Implications for finite treatment strategies in HBV 'cure'

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Summary

Nucleoside analogues are the mainstay of treatment for patients with chronic HBV infection but have no direct effect on covalently closed circular DNA. Long-term HBV viral suppression is now routine, but the desirable endpoint of functional cure is rarely achieved. Newer therapies, targeting other aspects of the replicative life cycle of HBV, present opportunities to deliver finite therapy and HBV 'cure'. This is an area of keen focus for the HBV community. We describe a severe case of hepatitis B reactivation, occurring shortly after the withdrawal of a nucleoside analogue within the protocol of a clinical trial (REEF-2). Despite best supportive care and prompt re-introduction of tenofovir, the patient developed subacute liver failure, requiring emergency orthotopic liver transplantation. As we strive to achieve HBV cure, this case highlights the potential risks of finite therapy and highlights the need for improved biomarker-driven strategies and re-evaluation of study protocols.

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Introduction

Highly potent nucleoside analogues (NA), such as tenofovir alafenamide, tenofovir disoproxil, and entecavir, are the mainstay of treatment for patients with HBeAg-negative chronic hepatitis B infection, effectively suppressing viral replication and reducing the risk of long-term complications, such as cirrhosis and hepatocellular carcinoma.[1] However, these agents do not directly target intrahepatic covalently closed circular DNA (cccDNA), and the optimal endpoint of HBsAg loss is rarely achieved.[2] Given the potential side effects of long-term NA therapy, the community has been exploring the safety of NA withdrawal in the context of interventional clinical trials and novel therapies, primarily focusing on selected non-cirrhotic individuals who remain HBsAg positive.[3] Limited data suggest that stopping NA treatment may be followed by enhanced rates of HBsAg seroclearance.[4]

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Case of a 54-year-old man

In this case report, we describe a 54-year-old male with HBeAg-negative chronic HBV infection, who experienced a life-threatening flare of HBV shortly after stopping NA therapy within a clinical trial. Despite the prompt reintroduction of NA therapy, he rapidly developed subacute liver failure and required urgent liver transplantation. Although reports of hepatic decompensation in this context are rare, our case highlights the urgent need for better baseline predictors of severe relapse, more stringent retreatment criteria in clinical trials given the aspiration for HBsAg loss ("functional cure"), and a deeper understanding of the underlying immunobiology. Furthermore, the severity of this case should prompt a re-evaluation of the overall merits of a finite therapy paradigm within clinical trial protocols with new HBV therapies.

This patient has been under regular follow-up at the Institute of Liver Studies, King's College Hospital, for over 10 years. He had demonstrated persistently undetectable HBV DNA levels since commencement of tenofovir monotherapy in December 2009, with optimal concordance and clinic attendance. HBsAg titres ranged from 904 to 3,611 IU/ml, with cross-sectional imaging, normal platelet count, and serial Fibroscan® readings confirming the absence of significant liver fibrosis (liver stiffness measurements ranging from 3.6 to 6.1 kPa).

After discussion and full informed consent, he was enrolled into the REEF-2 study (ClinicalTrials.gov Identifier: NCT04129554). REEF-2 is an ongoing randomised, double-blinded, placebo-controlled, phase IIb study exploring a range of endpoints with JNJ-3989 (small-interfering RNA) + JNJ-6379 (capsid assembly modulator) + NA combination therapy. The patient was randomized to the control arm, and during 48 weeks of continued treatment with tenofovir + placebo, his liver biochemical parameters were all within normal range, including undetectable HBV DNA (<lower limit of quantification) and an HBsAg level of 652 IU/ml at week 48. In accordance with the study protocol, his trial medication (tenofovir + placebo) was stopped at this time point, and monitoring visits were carried out at follow-up weeks 2 and 4, and at monthly intervals thereafter. Retreatment criteria were defined as follows: i) immediate retreatment with NA with signs of decreasing liver function based on laboratory findings (*e.g.* international normalised ratio [INR], direct bilirubin) or clinical assessment (*e.g.* ascites, hepatic encephalopathy); and ii) consideration of retreatment in cases of HBeAg seroreversion, increases in HBV DNA >2,000 IU/ml in combination with alanine aminotransferase

(ALT) >5x the upper limit of normal, or increases in HBV DNA >20,000 IU/ml (confirmed by 2 tests performed ≥4 weeks apart). An Independent Data Monitoring Committee (IDMC) and an Independent Flare Expert Panel (iFLEP) were commissioned for the conduct of the study.

Blood tests performed at week 8 (day 56) of follow-up revealed a high HBV DNA level of 7.07 $\log_{10}$ IU/ml. There was a slight increase in ALT (60 U/L, normal range: <43 U/L), but other liver parameters were within the normal range, including aspartate aminotransferase (AST, 39 U/L), total bilirubin (14 μmol/L), and INR (0.9). He remained well and asymptomatic, and an unscheduled visit was arranged for 2 weeks later. Blood tests at this subsequent visit (day 71) revealed a further rise in ALT (513 U/L) and AST (750 U/L, normal range: <36 U/L) and whilst synthetic liver function remained normal, the patient was asked to come back to the clinic and restarted on tenofovir (day 76). By then, he had developed nausea and general malaise, and serum aminotransferases had risen further (AST: 1,055 U/L, ALT: 1,962 U/L), in parallel with rising HBV DNA (8.98 $\log_{10}$ IU/ml), HBsAg (>124,925 IU/ml), and newly deranged synthetic liver profile (total bilirubin: 32 μmol/L, INR: 1.8). Unfortunately, his condition deteriorated with the onset of jaundice over the next few days, and he was admitted to King's College Hospital (day 83). Cross-sectional imaging performed at the time of admission revealed a normal sized liver with unremarkable morphology, and a non-invasive screen excluded other causes of acute hepatic injury. He received full supportive therapy. Serial blood tests over the next few days revealed a further rise in serum aminotransferases (peak ALT: 3,432 U/L, peak AST: 2,599 U/L), in parallel with worsening hyperbilirubinaemia and coagulopathy. He deteriorated significantly on day 92 of follow-up, developing grade 2 hepatic encephalopathy and asterixis, and was transferred to the liver intensive care unit for further optimisation and commencement of renal haemofiltration.

Despite a dramatic reduction in HBV DNA levels within 21 days of starting tenofovir therapy (fall from 952,000,000 IU/ml to 1,440 IU/ml), serial cross-sectional imaging confirmed a progressive reduction in liver volume, indicating ongoing hepatocyte cytolysis and inadequate hepatic regeneration. Bilirubin and INR levels continued to rise with peak values of 403 μmol/L and 3.8, respectively. At this point, model for end-stage liver disease (MELD) score was 40, consistent with a very poor short-term prognosis and he was listed for super-urgent liver transplantation (day 93 of follow-up). In the UK, qualifying for transplantation in this context is based on a composite of age, INR, serum bilirubin, presence of hepatic encephalopathy, and time from jaundice to encephalopathy of more than 7 days, and the patient met all of these criteria. A suitable liver graft became available early in the morning of day 100 of follow-up, and he underwent standard liver transplantation with a whole, DBD (donation after brainstem death), non-fatty graft later that day. The explanted liver was grossly atrophic and weighed 601 g, whilst histologically there was widespread loss of liver parenchyma and replacement with collapsed stroma, congested blood contents and reactive ductules. Many of the remaining hepatocytes stained positive for HBsAg. The patient received standard post-transplant care, including the administration of hepatitis B immunoglobulin (Hepatect®) and tacrolimus-based immunosuppression, made a good recovery, and was discharged on day 112 of follow-up. The temporal relationship between HBV DNA, HBsAg, ALT, bilirubin, and INR levels is shown in Fig. 1.

Discussion

The clinical picture we describe is 1 of severe HBV reactivation after NA discontinuation in the context of a clinical trial (REEF-2), in which the patient was in the placebo group and did not receive active treatment. At present, NAs remain the mainstay of treatment in patients with HBeAg-negative chronic HBV disease according to European Association for the Study of the Liver (EASL) guidelines,[5] although the lack of significant HBsAg loss has encouraged the clinical and research community to explore the feasibility and safety of finite therapy in non-cirrhotic populations.[3] Several studies have shown that withdrawal of NAs can accelerate loss of HBsAg and achievement of "functional cure,"[2,4] suggesting that low-level viral reactivation after long-term suppression with NA therapy may be beneficial in priming the immune system to eradicate residual cccDNA-containing hepatocytes. Such findings have encouraged clinical practice guidelines from international committees, such as EASL and the Asian Pacific Association for the Study of the Liver (APASL), to recommend that withdrawal of NAs can be considered in

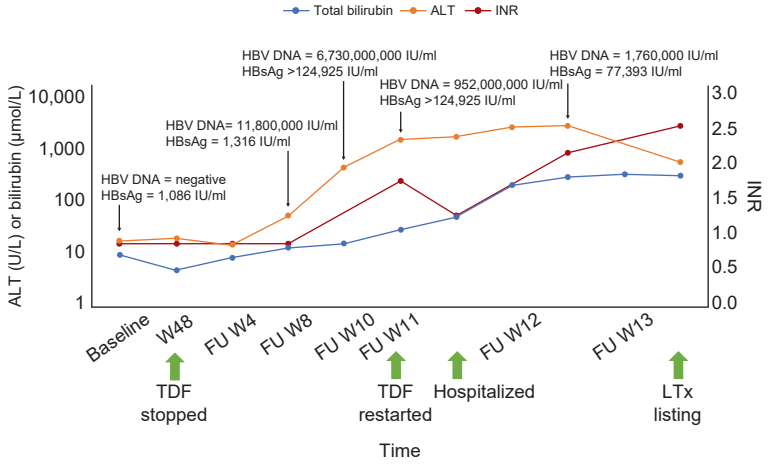

Fig. 1. HBV DNA, HBsAg, total bilirubin, ALT, and INR over time. ALT, alanine aminotransferase; INR, international normalised ratio; FU, follow-up; TDF, tenofovir disoproxil fumarate.

selected non-cirrhotic, HBeAg-negative patients who have achieved an adequate period of virological suppression, provided that close monitoring can be guaranteed.[5]

Several groups have shown that virological relapse is relatively common after withdrawal of NA therapy. In a recent meta-analysis of 37 studies, the cumulative incidence of virological relapse (variably defined in studies as HBV DNA >60 IU/ml, >200 IU/ml, or >2,000 IU/ml) was 44% at 6 months and 63% at 12 months. Despite this, clinical relapse (defined as a combination of virological relapse and an increase in serum aminotransferases above the normal range) was found to occur significantly less frequently (35% at 12 months).[6] Severe adverse outcomes following withdrawal of NA therapy have been reported, although these events are especially rare in the non-cirrhotic population. For example, despite the absence of any apparent risk factors for hepatic decompensation, *Chen et al.* described 5 patients without cirrhosis in their cohort (3 HBeAg positive, 2 HBeAg negative) who developed hepatic decompensation following withdrawal of NA therapy, with events occurring at 40, 40, 144, 169, and 290 weeks after stopping treatment. One fatality was noted despite early reintroduction of tenofovir therapy.[7] Meanwhile, Ma *et al.* described 3 non-cirrhotic patients with hepatic decompensation, including 1 death, with the time from discontinuation of treatment to clinical decompensation ranging from 15 to 91 weeks.[8]

Recent studies performed in South East Asia have suggested that corticosteroids may improve short-term survival in patients with severe liver injury secondary to hepatitis B infection.[9] However, results are conflicting and there is a paucity of randomized controlled studies exploring the combination of corticosteroid and NA therapy compared with standard of care. Mechanistically, any potential benefit of corticosteroids is expected to be limited in those with more advanced liver failure, in whom there is overwhelming hepatocyte necrosis and insufficient cellular regeneration. In our case, the patient had rapidly declining liver reserve, a MELD score of 40, and progressive reduction in liver volumes on serial imaging, and thus it was considered that corticosteroids would not provide any additional benefit.

If a finite therapy approach is to be considered more widely by the community, it is clear that a delicate balance exists between the potential immunological benefits of NA discontinuation, including accelerated rates of HBsAg loss, and the inherent risk of excessive hepatocyte damage and consequent liver failure. To address this balance, retreatment criteria have been suggested based on a combination of synthetic liver function, serum aminotransferases, and HBV DNA level.[10] The initial retreatment criteria used in the REEF-2 study were closely aligned with these field recommendations. In our patient, the HBV DNA titre was very high at week 8 of follow-up (7.07 log), but this alone was not sufficient to trigger retreatment within the initial study protocol as serum aminotransferases were not significantly elevated and synthetic liver function was normal, and it was the first HBV DNA reading >20,000 IU/ml. After safety panel review and internal discussion, the trial sponsor adapted the criteria for off-treatment monitoring and NA retreatment so that any future HBV titre above 5 log should trigger immediate retreatment with NA, regardless of a confirmatory test or other biochemical parameters or ALT values. This change was implemented as an urgent safety measure into the study protocol and other protocols with finite treatment concepts.

In summary, we describe the case of a non-cirrhotic HBeAg-negative patient who rapidly developed subacute liver failure following NA withdrawal and required emergency liver transplantation within 100 days. Such events are rare but highlight the potential risks of withdrawing NA therapy and the need for evolved, standardised re-treatment criteria if these steps are taken. Based on our case, re-treatment with NA should be initiated in response to significant increases in serum HBV DNA, regardless of other liver parameters. Whilst our patient resided in the UK and underwent super-urgent liver transplantation, those living in other countries may not have such readily available access to this service. Moving forward, the community needs to tread carefully in advancing new therapies to achieve HBV cure and should re-assess the role of NAs in clinical trials.

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Conflicts of interest

Kosh Agarwal has received grants from Abbott and MSD; has served as a consultant for Janssen, Assembly, Arbutus, Immunocore, Roche, BMS, BI, Novartis, Shinoigi, and Sobi; has served as a speaker for Gilead and Sobi; and has served on a data safety monitoring board or advisory board for Drug Farm, NUC-B, and Aligos. James Lok and Ivana Carey have no disclosures to report. Yatin Shivkar, Michael Biermer, and Isabelle Lonjon-Domanec are full-time employees of Janssen Research and Development. Thomas Berg has received grants from Abbvie, BMS, Gilead, MSD/Merck, Humedics, Intercept, Merz, Novartis, and Sequana Medical; has served as a consultant for Abbvie, Alexion, Bayer, Gilead, Eisai, Intercept, Ipsen, Janssen, MSD/Merck, Novartis, Roche, Sequana Medical, and Shionogi; has served as a speaker for Abbvie, Alexion, Bayer, Gilead, Eisai, Intercept, Ipsen, Janssen, MedUpdate GmbH, MSD/Merck, Novartis, and Sequana Medica; has received travel support from Gilead, Abbvie, Intercept, and Janssen; has served on a data safety monitoring board or advisory board for Janssen; and has served as Secretary General of EASL.

Please refer to the accompanying ICMJE disclosure forms for further details.

Authors' contributions

All authors were involved in the interpretation of the data, the conception and the drafting of the manuscript and approved the final version of the manuscript.

Data availability statement

The data sharing policy of Janssen Pharmaceutical Companies of Johnson & Johnson is available at https://www.janssen.com/clinical-trials/transparency. As noted on this site, requests for access to the study data can be submitted through Yale Open Data Access (YODA) Project site at http://yoda.yale.edu.

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Supplementary data

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References

[1] Su T-H, Hu T-H, Chen C-Y, Huang Y-H, Chuang W-L, Lin C-C, et al. Four-year entecavir therapy reduces hepatocellular carcinoma, cirrhotic events and mortality in chronic hepatitis B patients. Liver Int 2016;36(12):1755–1764.

[2] Jeng W-J, Chen Y-C, Chien R-N, Sheen I-S, Liaw Y-F. Incidence and predictors of hepatitis B surface antigen seroclearance after cessation of nucleos(t)ide analogue therapy in hepatitis B e antigen-negative chronic hepatitis B. Hepatology 2018;68(2):425–434.

[3] 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.

[4] Berg T, Simon KG, 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.

[5] 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.

[6] Hall SAL, Vogrin S, Wawryk O, Burns GS, Visvanathan K, Sundararajan V, et al. Discontinuation of nucleot(s)ide analogue therapy in HBeAg-negative chronic hepatitis B: a meta-analysis. Gut 2021. gutjnl-2020-323979.

[7] Chen C-H, Hung C-H, Wang J-H, Lu S-N, Hu T-H, Lee C-M. Long-term incidence and predictors of hepatitis B surface antigen loss after discontinuing nucleoside analogues in noncirrhotic chronic hepatitis B patients. Clin Microbiol Infect 2018;24(9):997–1003.

[8] Ma T-L, Hu T-H, Hung C-H, Wang J-H, Lu S-N, Chen C-H. Incidence and predictors of retreatment in chronic hepatitis B patients after discontinuation of entecavir or tenofovir treatment. PLoS One 2019;14(10):e0222221.

[9] Zhao B, Zhang H-Y, Xie G-J, Liu H-M, Chen Q, Li R-F, et al. Evaluation of the efficacy of steroid therapy on acute liver failure. Exp Ther Med 2016;12(5):3121–3129.

[10] van Bömmel F, Berg T. Risks and benefits of discontinuation of nucleos(t)ide analogue treatment: a treatment concept for patients with HBeAg-negative chronic hepatitis B. Hepatol Commun 2021;5(10):1632–1648.