Background: A complete virological response is closely related to the long-term outcome of patients with chronic hepatitis B and prevention of emerging HBV mutations. We aimed to evaluate the efficacy of tenofovir disoproxil fumarate (TDF) monotherapy compared to entecavir-adefovir dipivoxil (ETV-ADV) combination therapy in patients with suboptimal responses to long-term lamivudine-adefovir dipivoxil (LAM-ADV) therapy for nucleoside analogue-resistant chronic hepatitis B.

Methods: Patients (n=60) were randomized to TDF monotherapy or ETV-ADV combination therapy for 96 weeks. All patients had the rt204I/V mutation and serum HBV DNA was measured (>60 IU/ml) during LAM-ADV therapy. The primary end point was a complete virological response (HBV DNA <20 IU/ml) at week 96.

Results: The median duration of prior LAM-ADV rescue therapy was 43 (7–108) months. A complete virological response was achieved in 86.6% and 53.3% of patients in the TDF and ETV-ADV groups, respectively, at week 96 (P=0.005). Reduction in serum HBV DNA was significantly greater in the TDF group than in ETV-ADV group (-3.2 ±1.2 versus -2.6 ±1.2; P=0.01). Hepatitis B e antigen loss (22.2% versus 16.6%; P=0.731) and biochemical responses (76.7% versus 73.3%; P=0.766) were not different between the TDF and ETV-ADV groups. No newly emerged mutations were detected. Both therapies demonstrated favourable safety profiles.

Conclusions: TDF therapy achieved a better complete virological response than ETV-ADV therapy in chronic hepatitis B patients with suboptimal response to long-term LAM-ADV rescue therapy. (KCT0000627).

Original article

Tenofovir disoproxil fumarate monotherapy is superior to entecavir-adefovir combination therapy in patients with suboptimal response to lamivudine-adefovir therapy for nucleoside-resistant HBV: a 96-week prospective multicentre trial

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Introduction

HBV infection is characterized by varying degrees of viral replication and hepatic inflammation, which often leads to cirrhosis, hepatic decompensation and hepatocellular carcinoma (HCC) [1,2]. Antiviral therapy can suppress HBV replication and hepatic necro-inflammation, and thus improve the long-term outcomes of patients with chronic hepatitis B [3,4]. A complete virological response is strongly correlated with the prevention of disease progression in patients with or without cirrhosis [5,6]. Lamivudine (LAM) is a potent and safe antiviral agent.
but long-term monotherapy often leads to high rates of viral resistance [7,8]. A combination therapy with ade-fovir dipivoxil (ADV) and LAM markedly reduces the ADV-resistance when compared with ADV monother-
aphy, and has been recommended as a rescue therapy in patients with LAM-resistant chronic hepatitis B [9,10]. A virological response to LAM-ADV rescue therapy is associated with baseline HBV DNA level, gender, presence of the hepatitis B e antigen (HBeAg) and viral genotype. However, a complete virological response was not sufficiently achieved during long-term LAM-ADV rescue therapy in several reports [11–13].

Entecavir (ETV) and tenofovir disoproxil fumarate (TDF) are potent HBV inhibitors with a high barrier to viral resistance and are used as the first-line therapy in treatment-naïve patients with chronic hepatitis B [14–16]. The antiviral efficacy of ETV was compromised in nucleoside analogue (NA)-experienced chronic hepatitis B patients, especially in patients with the LAM-resistant mutation [17], but combination therapy with ADV showed promising outcomes in patients with partial virological responses to long-term LAM-ADV rescue therapy [18]. TDF-based rescue strategies also demonstrated excellent virological responses, and long-term viral suppression was maintained with TDF monotherapy or TDF-NA combination therapy in NA-experienced patients [19,20]. However, in previous studies, TDF monotherapy showed limited efficacy in patients with ADV-resistance, and a cumulative virological response to TDF monotherapy was inferior to combination therapy with LAM in NA-experienced patients [21,22].

Minimal data are available regarding the long-term outcomes of ETV-ADV combination therapy and TDF-based rescue therapies in NA-experienced chronic hepatitis B patients. The aim of the present study was to compare the long-term efficacy of TDF monotherapy and ETV-ADV combination therapy in patients with NA-resistant chronic hepatitis B who showed suboptimal responses to LAM-ADV rescue therapy.

Methods

Study design

The present study was a prospective, randomized, active-control, open-label, multicentre, 96-week clinical trial conducted with patients who were being treated with LAM-ADV rescue therapy but had suboptimal responses. Patients were randomized and stratified by their HBV DNA level at screening (<2,000 IU/ml; 2,000–2,000,000 IU/ml; >2,000,000 IU/ml) using a web-based central application system to receive either TDF (300 mg) once daily or ETV (1 mg) and ADV (10 mg) once daily. Study subjects were evaluated every 12 weeks for adverse events and pill counts (as a compliance measure), and haematology, biochemistry assay, and urinalysis were conducted. Serum HBV DNA levels were monitored every 12 weeks with the COBAS AmpliPrep/COBAS TaqMan HBV Test, v2.0 (Roche Diagnostics, Indianapolis, IN, USA), which has a lower detection limit of 20 IU/ml. Hepatitis B surface antigen (HBsAg) quantification was performed every 12 weeks with an Architect HBsAg assay (Abbott, Abbott Park, IL, USA). HBV-drug-resistant mutations including LAM, ADV, ETV and TDF (rtI180, rt204, rt181, rt236, rtI69, rt184, rt202, rt250 and rt194) were identified using a restriction fragment mass polymorphism (RFMP) assay (Genematrix, Yongin, Korea) [23]. HBeAg and anti-HBe antibodies using microparticle enzyme immunoassays available as commercial kits (Abbott, Wiesbaden, Germany) were detected at baseline, week 48 and week 96. This study was approved by the Institutional Ethics Committee of seven investigational sites in Korea, and written informed consent was obtained from all patients (WHO International Clinical Trials Registry Platform main ID KCT0000627).

Patients

The first patient was randomized on 14 August 2013, and the last visit of subjects happened on 20 May 2016. The inclusion criteria were age 20–75 years, confirmed emergence of the rtM204 mutation with a virological breakthrough (Δ serum HBV DNA >1 log IU/ml) any time before screening, initiation of LAM-ADV combination therapy at least 24 weeks prior to selection, serum HBV-DNA-positive continuously in real-time PCR assay during the LAM-ADV rescue therapy, and serum HBV DNA >60 IU/ml at screening. Patients were excluded if they had ETV-resistant mutations (rtI169, rtI184, rtS202 and rtM250) any time before screening, decompensated cirrhosis (uncontrolled ascites, history of variceal bleeding, history of hepatic encephalopathy, Child-Pugh score ≥8), evidence of renal insufficiency defined as creatinine clearance (Ccr) <60 ml/min (using the Cockcroft–Gault equation), undergone interferon or other immunomodulatory treatment for a HBV infection 6 months before the screening for this study, a medical condition that requires concurrent use of systemic corticosteroid or other immunosuppressive agent (including chemotherapeutic agent), a history of abusing alcohol (more than 40 g/day in men, 20 g/day in women) or illicit drugs, other concomitant chronic viral infections (HCV or HIV), one or more additional known primary or secondary causes of liver disease (other than hepatitis B [for example, autoimmune hepatitis, haemochromatosis, alpha-1 antitrypsin deficiency, Wilson’s Disease, other congenital or metabolic conditions affecting the liver, congestive heart failure or other severe
cardiopulmonary disease], a history of HCC or findings suggestive of possible HCC at screening, severe bone disease (osteomalacia, chronic osteomyelitis, osteogenesis imperfecta, osteochondroses) or they were pregnant or breastfeeding.

Outcome assessment and definition
The primary end point was the proportion of patients with a complete virological response (HBV DNA <20 IU/ml) at week 96. Secondary end points were the changes in mean serum HBV DNA levels, serum HBsAg levels during 96 weeks of treatment, the proportion of patients with normal alanine aminotransferase (ALT), HBeAg loss or seroconversion, resistance mutations to ADV, ETV or TDF, and the development of fatal complications, such as decompensated cirrhosis and HCC. A biochemical response was defined as a decrease in the serum ALT levels to the normal range. Virological breakthrough was defined as an increase in the serum HBV DNA level by 1 log_{10} above the nadir on at least two consecutive occasions. Liver cirrhosis was diagnosed by histology or imaging features along with the presence of thrombocytopenia, gastro-oesophageal varices, ascites or encephalopathy [24].

Statistical analysis
A sample size of 60 patients had an 80% power to indicate the superiority of TDF monotherapy over ETV-ADV combination therapy for the primary end point, with a 5% significance level, assuming response rates of 61% in the TDF group and of 29% in ETV-ADV group [18,19]. Efficacy and safety analyses included all randomized patients who received at least one dose of the study medication. All missing data were treated as representing failure. Data were expressed as the median (range). χ² test or Fisher’s exact test were used for comparisons of variables between groups where appropriate. A P-value less than 0.05 was considered statistically significant. All analyses were performed using SPSS 14.0 software (SPSS Inc., Chicago, IL, USA).

Results
Patient characteristics
68 patients were screened from August 2013 to May 2014. 60 patients were randomized and 28 subjects of each group completed 96 weeks of treatment (Figure 1). The median month of LAM-ADV rescue therapy was 43 (7–108) months. 27 subjects of the TDF group and

**Figure 1. Patient dispositions**

![Diagram of patient dispositions](image-url)
29 of the ETV-ADV group had received LAM before LAM-ADV rescue therapy and 4 subjects had received clevudine ($P=0.332$). Previous alcohol consumption history was not statistically different between the two groups (14 in TDF group and 12 in ETV-ADV group; $P=0.602$) and there were no statistically significant differences between the two groups (Table 1).

Virological response

The proportion of subjects achieving the primary end point (HBV DNA <20 IU/ml at week 96) was 86.6% (26/30) in the TDF group and 53.3% (16/30) in the ETV-ADV group ($P=0.005$). During the study period, virological response rates were 70%, 83% and 80% in the TDF group and 30%, 53% and 53% in the ETV-ADV group at weeks 24, 48 and 72, respectively (Figure 2). The mean reductions in the HBV DNA level at week 48 and 96 were -3.01 ±0.85 and -3.24 ±1.12 log_{10} IU/ml in the TDF group and -2.35 ±1.16 and -2.59 ±1.15 log_{10} IU/ml in the ETV-ADV group. The mean HBV DNA reduction from baseline to week 96 in the ETV-ADV group was significantly smaller than that in the TDF group ($P=0.01$; Figure 3). In univariate analysis, TDF monotherapy ($P=0.007$) was significantly associated with complete virological response at week 96 but age ($P=0.65$), gender ($P=0.393$), cirrhosis ($P=0.236$), LAM-ADV duration ($P=0.829$), ADV resistance mutations ($P=0.643$) and HBeAg positivity ($P=0.583$) were not related with primary end point. Multivariate analysis revealed that only TDF monotherapy (OR 6.54, 95% CI 1.6, 25.4; $P=0.007$) remained as a significant independent predictive factor for cumulative virological response at week 96 (Univariate and multivariate analysis of baseline factors associated with complete virological response at week 96 in Additional file 1). At screening, 8 out of the 29 patients in the TDF group and 4 out of the 30 patients in the ETV-ADV group had ADV-resistant mutations (rt181TV and/or rt236T). The mean change from the baseline HBV DNA level, according to the ADV-resistant mutations at screening, was not statistically different in the ETV-ADV group ($P=0.461$) and in the TDF group ($P=0.415$) during the study period (Mean change in serum hepatitis B virus (HBV) DNA levels from baseline over 96 weeks in Additional file 1). Among these 12 patients with ADV-resistant mutations, a complete virological response was achieved in 10 patients at week 96. No patients experienced virological breakthrough during the study. A higher baseline serum HBV DNA level (>2,000 IU/ml) did not affect virological responses during the study period in either group (data not shown).

Serological and biochemical responses

The median levels of HBsAg at baseline, week 48 and week 96 were 3.72 (range 2.84–4.37), 3.51 (range 2.03–4.33) and 3.65 (range 1.9–4.02) log_{10} IU/ml in the TDF group and 3.59 (range 2.67–4.63), 3.56 (range 2.56–4.51) and 3.49 (range 2.41–4.5) log_{10} IU/ml in the ETV-ADV group, respectively. The mean reductions of the HBsAg level at weeks 48 and 96 were -0.16 ±0.22 and -0.18 ±0.27 log_{10} IU/ml in the TDF group and -0.07 ±0.19 and -0.16 ±0.32 log_{10} IU/ml in the ETV-ADV group, respectively. Reductions in the mean HBsAg from baseline to week 96 were not different between groups ($P=0.669$). The loss of HBeAg was achieved in 5 and 6 subjects in the TDF group (n=27) and in 1 and 4 subjects in the ETV-ADV group (n=24) at 48 and 96 weeks, respectively. The proportions of biochemical response rates were 80% and 76.7% in the

Table 1. Baseline characteristics of subjects

<table>
<thead>
<tr>
<th></th>
<th>Total (n=60)</th>
<th>TDF (n=30)</th>
<th>ETV+ADV (n=30)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years*</td>
<td>46.5 (24–69)</td>
<td>48.5 (33–69)</td>
<td>44 (24–66)</td>
</tr>
<tr>
<td>Male, n (%)</td>
<td>42 (70)</td>
<td>21 (70)</td>
<td>21 (70)</td>
</tr>
<tr>
<td>LAM-ADV duration, months*</td>
<td>43 (7–108)</td>
<td>42 (7–104)</td>
<td>43 (11–108)</td>
</tr>
<tr>
<td>Cirrhosis, n (%)</td>
<td>14 (23.3)</td>
<td>6 (20)</td>
<td>8 (26.7)</td>
</tr>
<tr>
<td>Prothrombin time, seconds*</td>
<td>11.6 (9.7–14.9)</td>
<td>11.4 (10.2–14.9)</td>
<td>11.6 (9.7–13.6)</td>
</tr>
<tr>
<td>Albumin, g/dl*</td>
<td>4.5 (3.7–5.0)</td>
<td>4.5 (3.7–4.9)</td>
<td>4.5 (4.0–5.0)</td>
</tr>
<tr>
<td>Total bilirubin, mg/dl*</td>
<td>0.6 (0.3–1.8)</td>
<td>0.7 (0.3–1.6)</td>
<td>0.6 (0.3–1.8)</td>
</tr>
<tr>
<td>Creatinine, mg/dl*</td>
<td>0.9 (0.6–1.2)</td>
<td>0.8 (0.7–1.1)</td>
<td>0.9 (0.6–1.2)</td>
</tr>
<tr>
<td>Phosphorus, mg/dl*</td>
<td>3.2 (2.1–4.3)</td>
<td>3.2 (2.1–4.3)</td>
<td>3.4 (2.1–4.2)</td>
</tr>
<tr>
<td>Creatinine clearance, ml/min*</td>
<td>96 (60.2–145.8)</td>
<td>97.4 (61.4–145.8)</td>
<td>89.2 (60.2–143.1)</td>
</tr>
<tr>
<td>ALT, IU/l*</td>
<td>27 (10–558)</td>
<td>28 (10–558)</td>
<td>25 (11–108)</td>
</tr>
<tr>
<td>HBeAg-positivity, n (%)</td>
<td>51 (85)</td>
<td>27 (90)</td>
<td>24 (80)</td>
</tr>
<tr>
<td>HBV DNA, log_{10} IU/ml*</td>
<td>3.28 (1.8–6.2)</td>
<td>3.34 (1.8–6.2)</td>
<td>3.24 (1.9–5.3)</td>
</tr>
<tr>
<td>HBsAg, log_{10} IU/ml*</td>
<td>3.7 (2.67–4.63)</td>
<td>3.72 (2.84–4.37)</td>
<td>3.59 (2.67–4.63)</td>
</tr>
</tbody>
</table>

*Median (range). ADV, adefovir dipivoxil; ALT, alanine aminotransferase; ETV, entecavir; HBeAg, hepatitis B e antigen; HBsAg, hepatitis B surface antigen; LAM, lamivudine; TDF, tenofovir disoproxil fumarate.
Figure 2. Proportion of patients with complete virological responses (serum HBV DNA <20 IU/ml) by study visit and treatment group.

Missing equals failure analysis. ADV, adefovir dipivoxil; ETV, entecavir; TDF, tenofovir disoproxil fumarate.

Figure 3. Mean change in serum HBV DNA levels from baseline over 96 weeks by time and study group.

The error bars indicate 95% CIs. ADV, adefovir dipivoxil; ETV, entecavir; TDF, tenofovir disoproxil fumarate.
TDF group and 73.3% and 73.3% in the ETV-ADV group at 48 and 96 weeks, respectively. Biochemical responses \((P=0.766)\) and the loss of HBeAg \((P=0.731)\) were not statistically different between the groups at week 96.

Resistance surveillance

The rt204I/V mutation, with or without the rt180 mutation, were documented in all 60 subjects before LAM-ADV rescue therapy. At the screening period of this study, 47 (80%) patients had the rt204I/V and/or rt180 mutation, 6 (10%) patients had the rt204I/V and rt181T/V mutations, and 4 (7%) patients had the rt181T/V and rt236T mutations \((HBV\ polymerase\ mutations\ at\ screening\ in\ Additional\ file\ 1)\). One subject had wild-type HBV at screening. Mutation profiles were evaluated with the RFMP assay in 14 and 5 subjects who had minimal viraemia at week 48 and week 96, respectively. The baseline mutation profiles persisted in 10 out of 14 patients at week 48 and in all of 5 patients at week 96, respectively (Table 2). No newly emerged mutations were detected in the RFMP assay during the study period, including ETV or TDF-resistant mutations in suboptimal responders. Only 1 out of 8 patients with a baseline ADV-resistant mutation did not show a complete virological response. However, the patient showed complete virological response during TDF monotherapy and had minimal viraemia (serum HBV DNA level, 45.6 IU/ml) only at week 96. The mutation profile could not be detected with the RFMP assay because of low viral load.

Safety

Both TDF monotherapy and ETV-ADV combination therapy were well tolerated and adverse events were similar between groups. The incidence of adverse events was not statistically different between groups, with 76% in the TDF group and 60% in the ETV-ADV group \((P=0.133)\). No patient died during the study period. In the TDF group, one patient received surgical treatment for a knee ligament injury after an automobile accident and a second patient required a red blood cell transfusion due to a relapse of Crohn’s disease, which was stable before screening. In the ETV-ADV group, one patient received several extracorporeal shock wave lithotripsy therapies for a ureteral stone and a second patient required treatment for chest pain after a sports injury. All adverse events and serious adverse events assessed were considered unrelated with the study drugs. Two subjects experienced decreased C\textsubscript{Cr} (<50 ml/min) and four subjects showed serum hypophosphataemia (<2.0 mg/dl) during the study period but all abnormal laboratory findings were recovered by week 96 (Safety profiles through the 96-week study period in Additional file 1). The mean C\textsubscript{Cr} was 99.25 ±24.31 and 91.31 ±24.24 ml/min at baseline and at week 96, respectively, in the TDF group and 93.36 ±24.9 and 89.42 ±22.03 ml/min at baseline and at week 96, respectively, in the ETV-ADV group. No significant difference in the mean C\textsubscript{Cr} was found between the TDF and ETV-ADV group from baseline to week 96 \((P=0.535)\).

Discussion

This is the first study, to our knowledge, to directly compare the long-term efficacy of TDF monotherapy with ETV-ADV combination therapy in patients who had suboptimal responses to long-term LAM-ADV rescue therapy. In this study, the complete virological response rate at week 96 was 87% in the TDF group and

### Table 2. Trend of mutations during the 96-week study period

<table>
<thead>
<tr>
<th>Baseline HBV DNA\textsuperscript{a}</th>
<th>Mutations at screening</th>
<th>Intervention</th>
<th>HBV DNA\textsuperscript{a} at week 48</th>
<th>Mutation at week 48</th>
<th>HBV DNA\textsuperscript{a} at week 96</th>
<th>Mutation at week 96</th>
</tr>
</thead>
<tbody>
<tr>
<td>1,600</td>
<td>M204I</td>
<td>ETV + ADV</td>
<td>156</td>
<td>M204I</td>
<td>&lt;20</td>
<td>ND</td>
</tr>
<tr>
<td>4,120</td>
<td>M204I</td>
<td>ETV + ADV</td>
<td>194</td>
<td>M204I</td>
<td>67</td>
<td>ND</td>
</tr>
<tr>
<td>5,530</td>
<td>L180M, M204I</td>
<td>ETV + ADV</td>
<td>69</td>
<td>L180M, M204I</td>
<td>30</td>
<td>ND</td>
</tr>
<tr>
<td>195,128</td>
<td>L180M, M204I</td>
<td>ETV + ADV</td>
<td>154</td>
<td>M204V</td>
<td>37</td>
<td>ND</td>
</tr>
<tr>
<td>1,570</td>
<td>L180M, M204V</td>
<td>ETV + ADV</td>
<td>440</td>
<td>L180M, M204V</td>
<td>227</td>
<td>L180M, M204V</td>
</tr>
<tr>
<td>3,560</td>
<td>L180M, M204V</td>
<td>ETV + ADV</td>
<td>499</td>
<td>L180M, M204V</td>
<td>197</td>
<td>L180M, M204V</td>
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<tr>
<td>10,400</td>
<td>L180M, M204V</td>
<td>ETV + ADV</td>
<td>2,070</td>
<td>L180M, M204V</td>
<td>525</td>
<td>L180M, M204V</td>
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<tr>
<td>18,400</td>
<td>L180M, M204V</td>
<td>ETV + ADV</td>
<td>8,280</td>
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<td>1,980</td>
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<tr>
<td>531</td>
<td>L180M, M204I/V</td>
<td>ETV + ADV</td>
<td>60</td>
<td>L180M, M204I/V</td>
<td>&lt;20</td>
<td>ND</td>
</tr>
<tr>
<td>12,000</td>
<td>L180M, M204V, A181V</td>
<td>ETV + ADV</td>
<td>86</td>
<td>L180M, M204V</td>
<td>&lt;20</td>
<td>ND</td>
</tr>
<tr>
<td>63,500</td>
<td>L180M, M204V</td>
<td>TDF</td>
<td>280</td>
<td>Wild type</td>
<td>&lt;20</td>
<td>ND</td>
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<tr>
<td>801,000</td>
<td>L180M, M204V</td>
<td>TDF</td>
<td>2,660</td>
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<td>142</td>
<td>L180M, M204V</td>
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<tr>
<td>1,470,000</td>
<td>A181T, N236T</td>
<td>TDF</td>
<td>136</td>
<td>A181T, N236T</td>
<td>LT FU</td>
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<tr>
<td>1,740,000</td>
<td>L180M, A181T, N236T</td>
<td>TDF</td>
<td>566</td>
<td>A181T, N236T</td>
<td>&lt;20</td>
<td>ND</td>
</tr>
</tbody>
</table>

\textsuperscript{a}IU/ml. ADV, adefovir dipivoxil; ETV, entecavir; LT FU, lost to follow-up; ND, not detected; TDF, tenofovir disoproxil fumarate.
53% in the ETV-ADV group. Lim et al. [18] reported that ETV-ADV combination therapy is superior to continuing LAM-ADV therapy in suboptimal responders to LAM-ADV rescue therapy but the complete virological response rate (HBV DNA <60 IU/ml) at week 52 was approximately 29%. The extended ETV-ADV therapy showed a benefit in the present study but nearly half of patients didn’t experienced an undetectable HBV DNA (HBV DNA <20 IU/ml) during study period. In contrast, TDF monotherapy was very effective at achieving complete virological responses in patients with LAM-resistance or in ADV-experienced patients from recent prospective clinical trials [25,26]. The response rate of TDF monotherapy in this study was very similar to that of previous studies for LAM-experienced patients with TDF rescue therapy. Therefore, TDF alone or the addition of TDF is recommended as an essential rescue therapy for NA-resistance in most practice guidelines [14–16].

Switching to another antiviral monotherapy with a high genetic barrier to viral resistance or adding a second antiviral with a complementary resistance profile is a primary strategy for antiviral resistance. ETV, an agent with a high genetic barrier, can control the virological breakthrough of LAM-resistant chronic hepatitis B in the early treatment period but the relatively high emerging rate of ETV-resistance is a concern [27]. Compared to LAM-ADV combination therapy, ETV-ADV combination therapy is a reasonable strategy to counter NA-resistant chronic hepatitis B. By switching to a high genetic barrier drug, from LAM to ETV, ADV maintains a complementary resistance profile. In the present study, the 96-week long-term efficacy of ETV-ADV combination therapy was more promising than previous trials because of relatively longer study period in patients with suboptimal responses to LAM-ADV rescue therapy or lower baseline HBV DNA levels in patients with LAM-resistant chronic hepatitis B [18,28]. Taken together, long-term ETV-ADV combination therapy is an acceptable alternative salvage therapy for NA-experienced chronic hepatitis B patients, especially with lower HBV DNA levels.

In previous reports, the efficacy of TDF monotherapy was attenuated in patients with ADV-resistant mutations in vitro [21,29]. Until recently, Korean National Health Insurance Service had recommended TDF-nucleoside combination therapy for patients with nucleoside analogue-resistant chronic hepatitis B because of preventing emergence of TDF-resistant mutation. However, in recent clinical trials, virological responses to TDF monotherapy were not inferior to combination therapy with NA in patients who experienced both nucleoside and nucleotide analogue therapy, and no TDF-resistant mutations emerged during the study period [25,26]. At the start of this study, 8 of the 30 subjects in the TDF group and 4 of the 30 in the ETV-ADV group had rt181T/V and/or rt236T mutations; however, ADV-resistant mutations were not associated with virological responses in both groups. During the 96-week trial, complete virological response was achieved in 11 out of 12 patients with ADV-resistant mutations, including 2 subjects who dropped out at week 84 and 96. Very low level of HBV DNA was detected only at week 96 in 1 patient, but a complete virological response was achieved from week 12 to week 84. TDF monotherapy rapidly suppressed viral replication during the early period of this study in patients with ADV-resistance. ETV-ADV combination therapy also suppressed ADV-resistance in this study, similar to LAM-ADV combination rescue therapy in patients with LAM-resistant chronic hepatitis B. Although, long-term ETV treatment could be associated with the development of ETV-resistant mutations in the ETV-ADV group, the HBV DNA level gradually declined in patients with partial virological responses to ETV-ADV, even LAM-resistant mutations persisted at week 48 and week 96. Continuing treatment of ADV with ETV might prevent the emergence of ETV-mutations and virological breakthroughs in the ETV-ADV group. However, the baseline HBV DNA level was lower than previous studies and only a small number of patients with the rt236T mutation were enrolled in the present study, therefore these results need to be interpreted appropriately.

The minimal decline of serum HBsAg that was observed during this study is comparable with previous reports detailing treatment-experienced patients undergoing long-term TDF therapy [22]. The serum HBsAg decline was not different between the TDF and ETV-ADV groups. Currently, very potent oral antiviral agents can persistently suppress HBV replication, but HBsAg clearance is unlikely to occur despite long-term virological responses [30–32]. Biochemical responses and HBeAg loss were similar with previous clinical trials [26,28]. Long-term antiviral therapy is needed to achieve a serological response in treatment-experienced patients with chronic hepatitis B.

Treatment with TDF alone and ETV-ADV combination therapy were well tolerated during the study period. The safety profiles were consistent with previously reported clinical trials and there was no significant difference in the occurrence of clinical and laboratory adverse events in both groups [18,26]. There were no discontinuations of study medicines and no development of HCC over the 96-week treatment period. Nephrotoxicity is commonly associated with TDF treatment for HIV-infected patients and ADV is known to be related to renal toxicity [33,34]. A decrease in the $C_{Cr}$ below 50 ml/min was reported in one patient from each group but this was all a temporary decline and recovered to above 50 ml/min at week 96 without any dose adjustment. Significant increases in the serum creatinine level or decreases in the serum
phosphorus level were not different between the TDF and ETV-ADV groups in this study. In recent reports, renal toxicity with antiviral therapy was acceptable and no significant difference among each antiviral agent, such as TDF, ETV and ADV, was reported [35,36].

The long-term outcomes of TDF monotherapy and ETV-ADV combination therapy were evaluated in the present clinical trial; however, the small number of study subjects is a limitation of this study. Secondly, the change in bone mineral density was not evaluated in this long-term study.

In conclusion, our results suggest that TDF monotherapy achieved superior complete virological response over ETV-ADV combination therapy in patients with NA-resistant chronic hepatitis B who showed suboptimal responses to long-term LAM-ADV rescue therapy. Both TDF monotherapy and ETV-ADV combination therapy did not promote the emergence of any HBV drug resistance mutation and demonstrated a favourable profile during the 96-week trial period. TDF monotherapy should be considered as the optimal salvage therapy for suboptimal responders to LAM-ADV rescue therapy for NA-resistant chronic hepatitis B.

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Disclosure statement

The authors have nothing to disclose.

Additional file

Additional file 1: Supplementary information can be found at https://www.intmedpress.com/uploads/documents/4049_Lee_Addfile1.pdf

References


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