

Full Length Research Paper

Randomized, prospective, efficacy and pharmacoeconomic trial of short course combination antiviral of lamivudine and adefovir versus entecavir monotherapy in HBeAg-positive chronic hepatitis B

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The aim of this study was to compare the safety, efficacy, and pharmacoeconomics of the diminishing type antiviral combination of lamivudine and adefovir group (LA group) and entecavir monotherapy group (E group) in HBeAg-positive chronic hepatitis B. One hundred (100) patients were randomized equally to LA group, and E group in a multi-center randomized clinical trial. In the LA group, the earliest time for lamivudine discontinuation was 12 weeks and adefovir monotherapy continued until 96 weeks. Group E received entecavir monotherapy for 96 weeks. At 12 weeks, the hepatitis B virus (HBV) DNA suppression and ALT normalization rates in LA group were both comparable to E group. At weeks 24 and 48 of treatment, the difference in virological response (VR) and HBeAg seroconversion between LA group and E group was not significant, while similar results were observed for the biochemical response (BR). At 96 weeks, HBeAg seroconversion of LA group were higher than that of the E group, but the difference was still not statistically significant, while BR and VR in LA and E group were similar. At the same time, no virological breakthrough or drug resistance occurred in either of the two treatment groups by week 96 of the study. Both treatment strategies were well tolerated, with a low incidence of adverse reaction. The costs of all items related to LA group were lower than those related to E group (RMB ¥14,480.13 vs. RMB ¥28,818.47; $t=164.78$, $p<0.001$). This study demonstrates that diminishing type antiviral combination of lamivudine and adefovir is economical, safe, and effective in HBeAg-positive chronic hepatitis B.

Key words: Chronic hepatitis B, HBeAg-positive, lamivudine, adefovir dipivoxil, combination treatment, entecavir, monotherapy, pharmacoeconomic.

INTRODUCTION

Antiviral therapy is a treatment for chronic hepatitis B. Domestically and internationally approved pharmacotherapies include interferon α , pegylated interferon α , lamivudine, adefovir dipivoxil, entecavir, etc. In the clinical setting, these drugs have both advantages

and disadvantages. One of major concerns of lamivudine therapy is increased incidence of drug resistance. Lamivudine is safe and strongly suppresses viral activity, but has high rates of drug resistance. Once the reverse transcriptase 204 mutation is present, cross-resistance to

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telbivudine and entecavir leads to decreased efficacy of those drugs. Adefovir dipivoxil has high rates of seroconversion and low resistance and no cross resistance with other nucleoside analogs, but antiviral activities are weak and onset of therapeutic response is slow. In summary, due to different mutation sites compared to the other three nucleoside analogs, ADV was selected as a basic agent of combination therapy. As the first approved agent for CHB patients, LAM was selected due to the abundant clinical experience and lowest cost. Evidence-based medicine identified that combination therapy could reduce drug-associated resistance to ensure long-term therapy (Degertekin and Lok, 2009), the combination of ADV and LAM results in greater viremia reduction than ADV monotherapy (Wang et al., 2013).

ETV is a deoxyguanosine analog with powerful activity in inhibiting viral replication. It is regarded as a high genetic barrier drug, as more than three sites for drug resistance related mutation are required (European Association for the Study of the Liver, 2012). Available data indicate that ETV was recommended as a first line option for long-term treatment of naive CHB patients instead of LAM resistant patients. In naive CHB patients, the 5-year rate of phenotypic resistance and virus breakthrough-related phenotype resistance were only 1.2 and 0.8%, respectively in patients treated with ETV (Tenney et al., 2009). However, it is very expensive and has been shown to be tumorigenic in laboratory animals while the clinical response is comparable to other antivirals. Thus, LAM and ADV were selected for the *de novo* combination treatment option. It is currently suggested that initial therapies involving either a combination of nucleoside/nucleotide analogs or monotherapy are both good options to prevent the development of resistance, especially, for the patients who need long-term treatment (Ayoub and Keeffe, 2008); however, relevant data from forecast research on efficacy, potential side effects, or an economic evaluation for the two strategies are rare.

It has been difficult to identify a method to judiciously apply combination therapy. Our design of optimizing the schedule for lamivudine/adeфовir combination therapy followed by maintenance adefovir monotherapy for treating HBeAg-positive chronic hepatitis B patients achieved encouraging results and is presented in this report. The purpose of this trial was to compare the safety, efficacy, and pharmacoeconomics of the diminishing type antiviral combination and entecavir monotherapy in HBeAg-positive chronic hepatitis B.

MATERIALS AND METHODS

Case selection

This study is a multi-center, randomized, clinical study whose design was examined and approved by the ethics committee. All patients voluntarily entered the study and paid for medical

treatment on their own. Patients were followed by telephone or home interview, and the data collection was performed by staff blinded to patients' treatment. A total of 100 of HBeAg-positive chronic hepatitis B patients were enrolled. The diagnosis and efficacy assessment criteria conform to the EASL Clinical Practice Guidelines: Management of chronic hepatitis B virus infection.

Inclusion criteria

The inclusion criteria includes the following:

1. Patients are between 30 to 60 years-of-age of both gender;
2. HBV serum markers: positive HBsAg and HBeAg for at least 6 months and serum HBV DNA > 10⁵ copies/ml;
3. Serum alanine aminotransferase (ALT) must have been analyzed within one month prior to the study;
4. The patient must have never received HBV antiviral therapy;
5. Female patients of childbearing age must be on birth control for the duration of the study
6. Serum anti HBeIgM positive.

Exclusion criteria

The exclusion criteria includes the following:

1. Diagnosis of hepatocellular carcinoma;
2. Clinical symptoms of decompensated liver disease;
3. Creatinine clearance < 70 ml/min;
4. ALT > 10-fold over normal limits at the time of screening or has had transient liver decompensation due to acute illness;
5. Hemoglobin < 10 g/dl, neutrophils < 1.5×10⁹/L, and platelets < 80×10⁹/L;
6. Other evidence of active liver disease (hepatitis A, E, C, HDV, HIV, and autoimmune disease);
7. Use of nephrotoxic drugs (example vancomycin, adefovir, cisplatin, etc.) within two months prior to the study;
8. The patient has been determined to have difficulties with compliance. The assessment was based on patients' lifestyle, medicine-taking habit, and their knowledge and attitude towards anti-viral therapies;
9. In addition to chronic hepatitis B, the patient has other serious organic or psychiatric disorders.

Study design

This study was a randomized, controlled, and multi-center clinical trial. Randomization of patient groups was performed with randomization tables, which extended to each clinical center. One hundred (100) patients were randomly assigned at 1:1 ratio into the diminishing type antiviral combination of lamivudine and adefovir group (LA group), entecavir monotherapy group (E group). The first 12 to 24 weeks of the LA group was the intensification phase where combined therapy of lamivudine and adefovir were administered. Patients in the LA group were prescribed LAM 100 mg and ADV 10 mg per day. During the phase of weeks 12-24, if HBV DNA falls below 1.0×10³ copies/ml (undetected level), lamivudine was terminated and adefovir monotherapy is continued. Lamivudine was also terminated at 24 weeks even if HBV DNA did not fall below 1.0×10³ copies/ml, which is also followed by adefovir monotherapy, and then into the maintenance treatment phase. Patients in the E group were prescribed entecavir at 0.5 mg per day. The duration of the clinical trial was 96 weeks.

Observation and follow-up

Follow-up of the two groups were performed at initiation and during

Table 1. Baseline characteristics of patients by group.

Group	Cases	Sex		Age (Years)		ALT(\times ULN)		HBV DNA (log ₁₀ copies/ml)	
		M	F	Mean \pm SD	Median(range)	Mean \pm SD	Median(range)	Mean \pm SD	Median (range)
LA	45	32	13	39.2 \pm 6.3	38.0(30-54)	5.1 \pm 2.1	5.5(2-8.5)	8.2 \pm 0.8	8.5(6.6-9.8)
E	47	33	14	39.0 \pm 6.1	38.0(31-54)	4.6 \pm 2.1	4.0(2-8.5)	8.1 \pm 0.9	8.4(6.5-9.8)

the weekends following weeks 12, 24, 36, 48, 60, 72, 84, 96 during the trial. Follow-up clinical assessments include: history and physical examination, quantitative HBV DNA(PCR), two pairs of semi for hepatitis B(HBV-M), serum biochemistry, alpha-fetoprotein, etc. LAM-, ADV-, and ETV-associated mutations were assessed for patients with virologic breakthrough via direct sequencing. Normal value of ALT was 0-40 IU/L.

Pharmacoeconomics

In the present study, the pharmacoeconomic analysis was of the cost-minimization type. Only the costs of the clinical treatment (those directly related to the health care system: medical care, observation and follow-up, medications) were taken into consideration. Indirect costs related to lost productivity, as well as intangible costs (those related to impaired quality of life), were not calculated.

For patients in the two groups, the costs were calculated separately for each of the following aspects: medical visits; antiviral medications; and medications for the treatment of adverse effects.

Statistical analysis

Quantitative data were presented as the mean \pm standard deviation (SD) (range), categorical data were presented as counts and percentages, and HBV DNA levels were presented as log transformation. Data were analyzed using the SPSS software package version 13.0. The t test was used for quantitative variables, while Pearson Chi-Square was used for categorical variables. All tests of significance were two-tailed, and significance was defined as $P < 0.05$.

RESULTS

Baseline characteristics

Between November 2007 and February 2011, 100 eligible patients participated in this clinical trial. The LA group contained 50 cases, of which three exhibited poor compliance, one traveled abroad for business, and one enrolled in another treatment program at a non-participating center. A total of five cases in the LA group were omitted from analysis and 45 patients completed the trial. The E group contained 50 cases; three patients withdrew due to fear of drug tumorigenicity, resulting in 47 patients completing the trial. The baseline characteristics of the patients were similar and no statistically significant differences were observed (Table 1).

Virological response

Suppression of serum HBV DNA

At baseline, serum HBV DNA levels for the two groups were similar. At 12 weeks, the mean serum levels of HBV DNA for the LA and E groups were lower by 4.663 \pm 0.515 log₁₀ copies/ml and 4.447 \pm 0.611 log₁₀ copies/ml, respectively. There were no statistical difference in HBV DNA reduction between the LA and E groups ($t=1.825$, $P=0.071$). At 24 weeks, the average reduction in HBV DNA levels between group LA (5.132 \pm 0.711 log₁₀ copies/ml) and group E (4.908 \pm 0.736 log₁₀ copies/ml) were comparable ($t=1.48$, $P=0.142$). At 36 weeks, HBV DNA in the LA group continued to show improvement and decreased by 5.197 \pm 0.766 log₁₀ copies/ml, which was still comparable to the E group, whose reduction by 5.078 \pm 0.822 log₁₀ copies/ml ($t=0.718$, $P=0.474$). Both therapy strategies can very effectively inhibit viral replication. The difference was not statistically significant. From 48 weeks, the HBV DNA level of two groups had remained at the undetectable level ($<10^3$ copies/ml). The average reduction in HBV DNA level in the LA group compared to baseline levels was 5.212 \pm 0.785 log₁₀ copies/mL at weeks 48, 60, 72, 84, and 96 of treatment, respectively. In the E group, the average reduction in HBV DNA level was 5.117 \pm 0.850 log₁₀ copies/mL at weeks 48, 60, 72, 84, and 96, respectively. No statistically significant difference existed between the two groups ($t=0.558$, $P=0.578$) (Figure 1).

Rates of undetectable serum HBV DNA

At 12 weeks, groups LA and E had negative HBV DNA test rates of 47% (21/45) and 36% (17/47), the difference between LA and E was not statistically significant ($\chi^2=1.04$, $P>0.05$). At 24 weeks, the negative HBV DNA test rates for two groups were 89% (40/45) and 74% (35/47), and the differences between two groups were also not statistically significant ($\chi^2=3.17$, $P>0.05$). At 36 weeks, the negative HBV DNA test rates in the LA and E group were 93% (42/45) and 89% (42/47), respectively. The difference was not statistically significant ($\chi^2=0.46$, $P>0.05$). From 48 weeks, the negative HBV DNA test rates for two groups were 100% (45/45) and 100% (47/47)

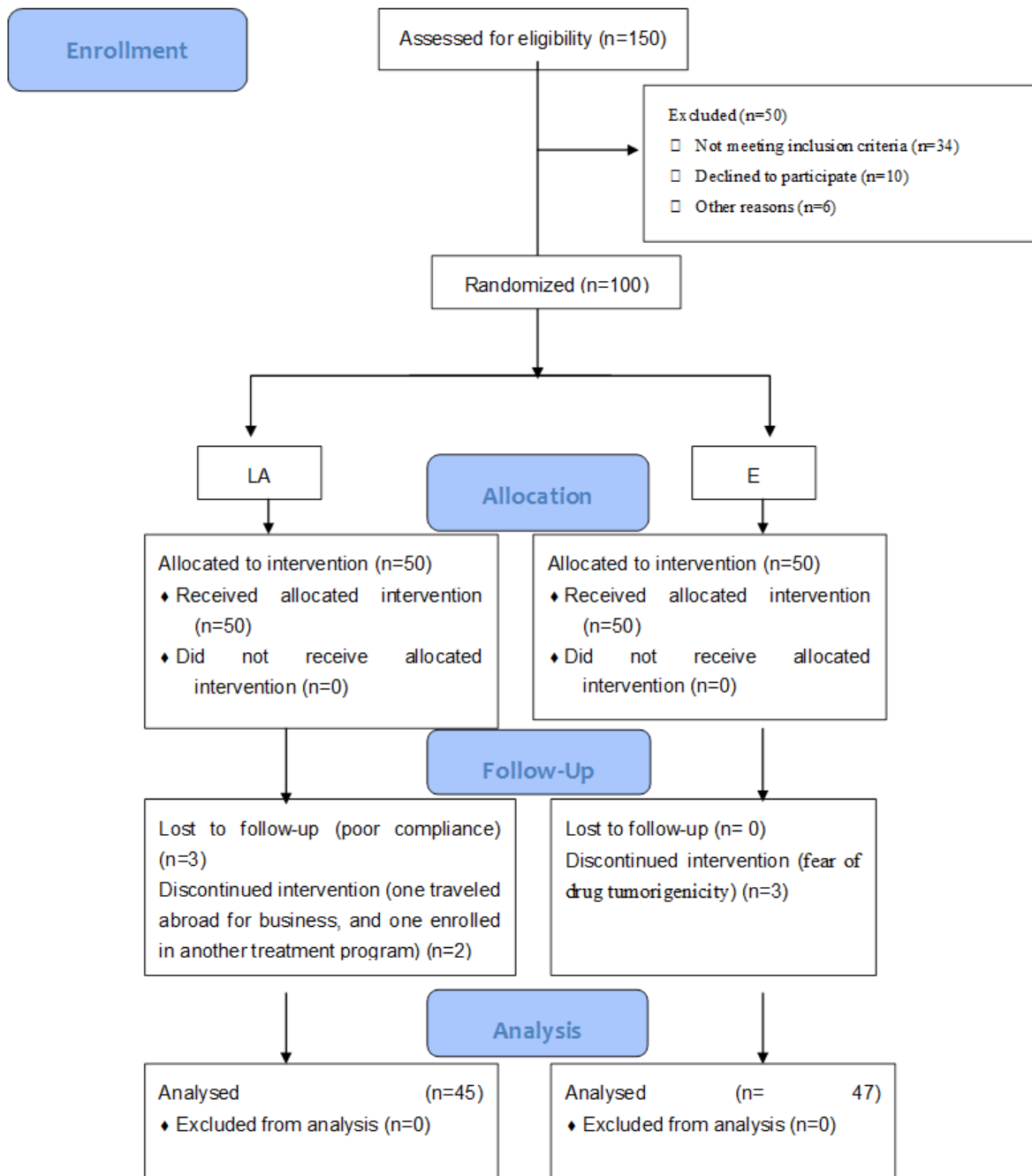


Figure 1. Changes in HBV DNA copy number (log₁₀ copies/ml).

by weeks 48, 60, 72, 84 and 96, and the difference was not statistically significant (P>0.05) (Figure 2).

Serological response (rates of HBeAg seroconversion)

At 24 weeks, there were 8 and 5 patients who had HBeAg/anti-HBe serological conversion in the LA and E

group, respectively; the HBeAg seroconversion rates of the LA and E groups were 18 and 11% , respectively. The difference between two groups was not statistically significant (χ²=0.97, P>0.05). At 48 weeks, the HBeAg seroconversion rates in the LA group continued to show improvement and had achieved 31% (14/45), which was still comparable to the E group, whose seroconversion rates was 26% (12/47) (χ²=0.35, P>0.05). However, after 48 weeks, HBeAg seroconversion of LA group continued

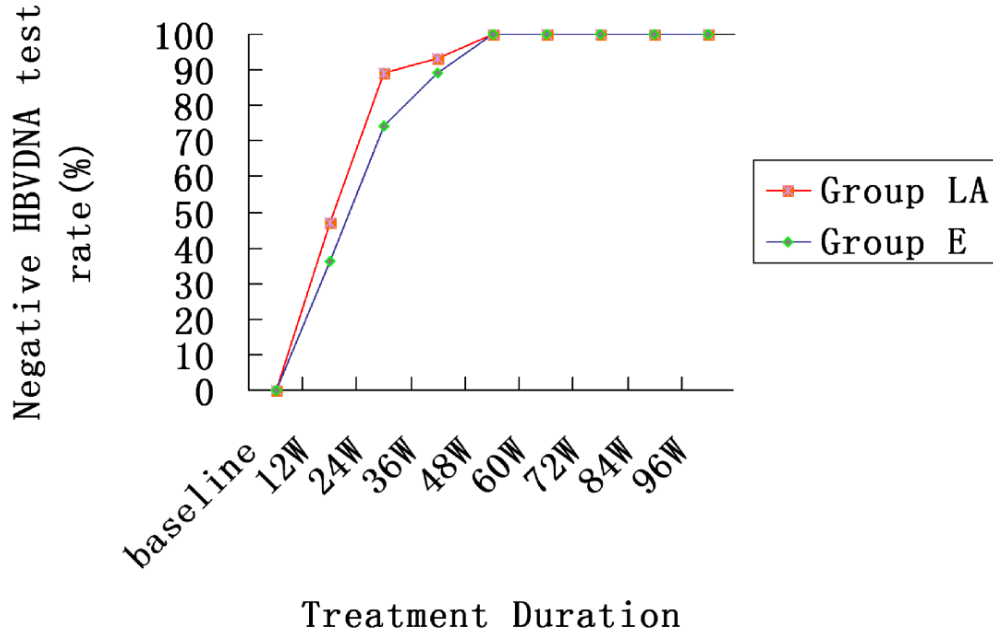


Figure 2. Percentage of negative HBV DNA tests.

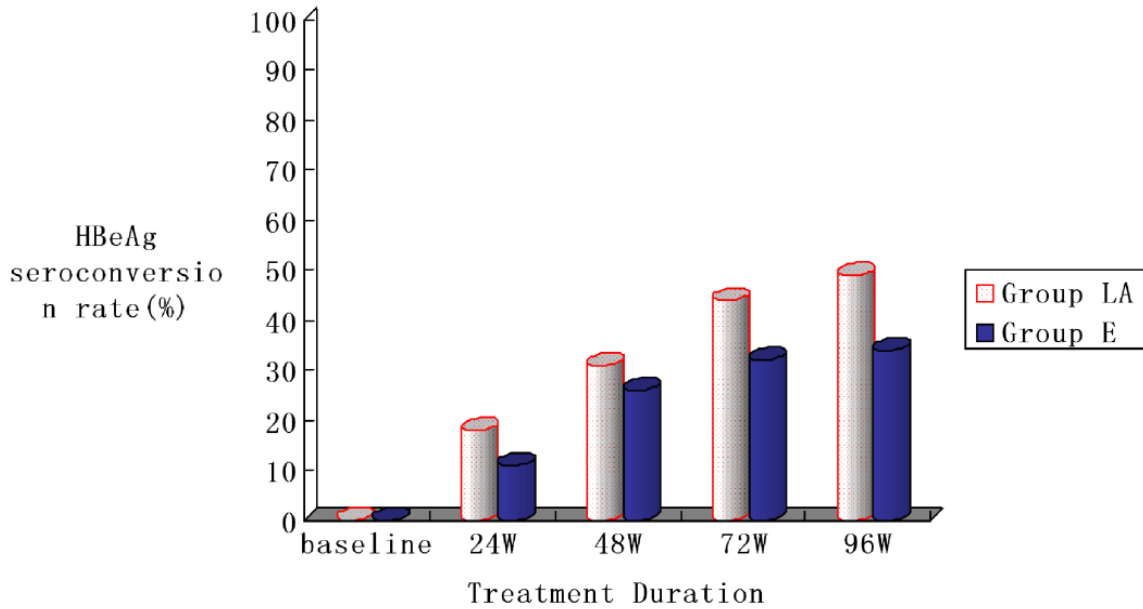


Figure 3. HBeAg seroconversion rate.

to improve. At 72 weeks, the HBeAg seroconversion rates of the LA and E groups were 44% (20/45) and 32% (15/47), respectively; the difference between two groups was not statistically significant ($\chi^2=1.53$, $P>0.05$). At 96 weeks, HBeAg seroconversion of LA group (49%) (22/45) were higher than that of the E group (34%) (16/47), but the difference was still no statistically significant ($\chi^2=2.09$, $P>0.05$) (Figure 3).

Biochemical response (rates of ALT normalization)

For the LA and E groups 12 weeks after trial initiation, serum ALT normalized in 56% (25/45) and 53% (25/47) of patients ($\chi^2=0.05$, $P>0.05$), respectively. At 24 weeks, the percentages of patients with normalized serum ALT in the LA and E groups were 96% (43/45) and 89% (42/47), respectively ($\chi^2 = 1.25$, $P > 0.05$). At 36 weeks, ALT

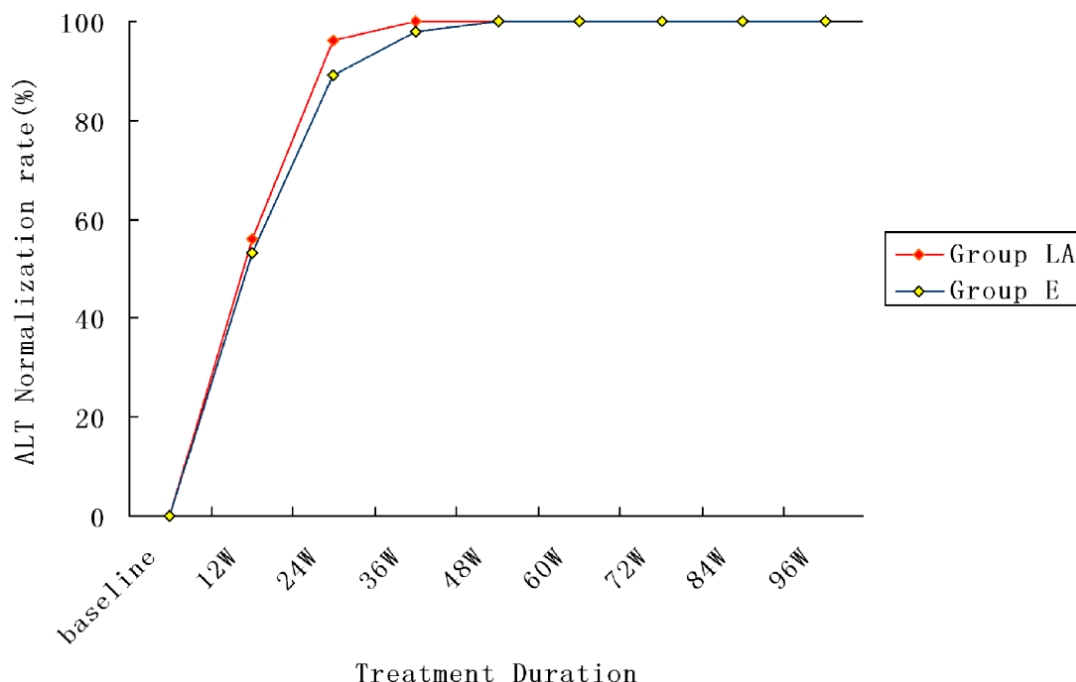


Figure 4. Percentage of patients with normalized serum ALT.

normalization rates in the LA and E groups were 100% (45/45) and 98% (46/47) ($\chi^2=0.97$, $P>0.05$). From 48 weeks, ALT normalization rates in two groups were 100% (45/45) and 100% (47/47) ($P>0.05$). None of these differences were statistically significant (Figure 4).

Virological breakthrough and drug resistance

During the 96-week treatment period, virological breakthrough did not occur in any of the 92 patients included in this study. That is, LAM-, ADV-, or ETV-associated mutations were not detected.

Pharmacoeconomic analysis

In the pharmacoeconomic analysis, we evaluated the direct costs related to treatment of the two groups. It is of note that, as previously mentioned, the costs related to antiviral of patients in two group were included in the final costs. The total costs include antiviral drug costs, outpatient service registration fee, adverse events processing fee, and assistant examination fee containing liver function, renal function, HBV-DNA, AFP, HBV-M and type-B ultrasonic (Figure 5).

In the pharmacoeconomic evaluation, the costs regarding antiviral drug costs were higher in the E group than in the LA group (RMB ¥25,824 vs. RMB ¥11,380; $t=156.4$, $p<0.001$). Costs were higher in the LA group only in the use of renal function. No statistical

difference was found in costs regarding outpatient service registration fee, adverse events processing fee, and assistant examination fee ($p>0.05$).

As seen in Figure 5, the analysis of the total costs per group, revealed greater economy in the LA group than in the E group (RMB ¥14,480.13 vs. RMB ¥28,818.47; $t=164.78$, $p<0.001$).

Safety analysis

Within the 96 weeks of clinical trial, we generally observed that the characteristics and rates of adverse reactions were comparable between the groups. About 18% of the patients developed at least one mild or moderate adverse reactions, which include: abnormal lab results (elevated ALT), fatigue, abdominal discomfort, upper abdominal pain, dizziness, insomnia, etc (Table 2). After receiving heteropathy, these symptoms soon get control.

DISCUSSION

Tremendous progress was made in the treatment of chronic hepatitis B in the past 10 years with the introduction of several novel nucleoside and nucleotide analogues. In clinical use, nucleoside/nucleotide analogues are uncomplicated, efficacious, and safe. Due to high replication rates of HBV, lack of proof reading or editing activity for reverse transcriptase, HBV mutations

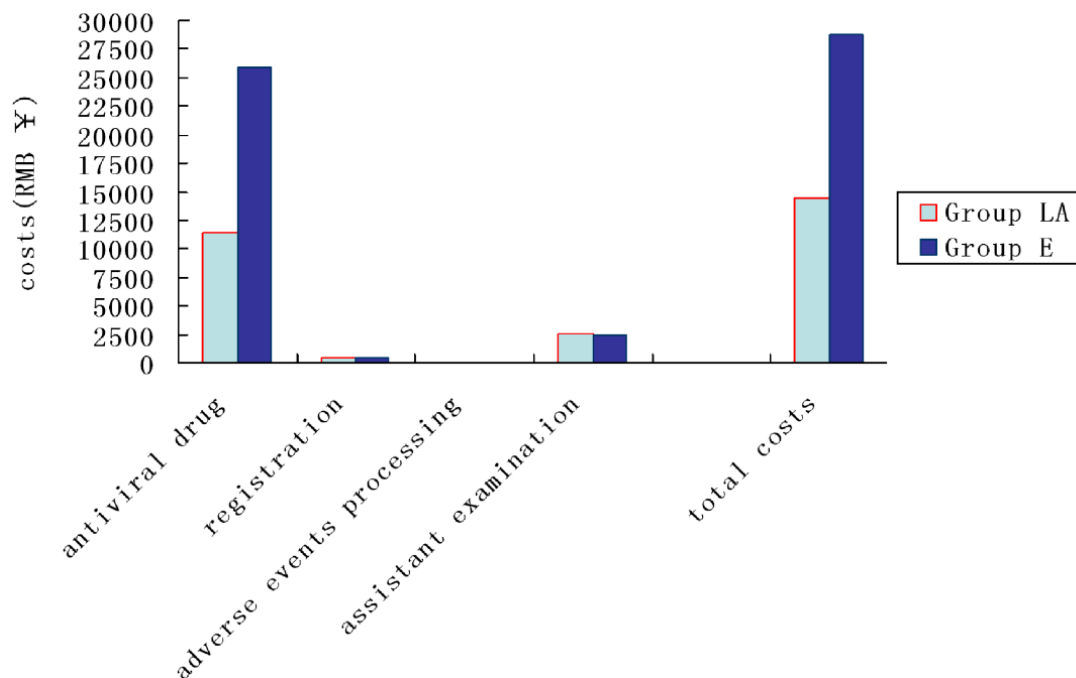


Figure 5. The cost analysis of the parameters investigated in both groups.

Table 2. Adverse events of patients by group.

Group	Elevated ALT	Fatigue	Abdominal discomfort	Dizziness	Insomnia	Epigastric pain
LA	1	4	1	1	1	1
E	3	4	1	0	0	0

can and do develop, resulting in drug resistance during long-term treatment. Drug resistance is one of the most important influencing factors limiting long-term nucleoside treatment for CHB patients (Papatheodoridis and Deutsch, 2008). Current strategy to management of HBV drug resistance includes rescue therapy, which may reduce the efficacy of follow-up pharmacotherapy, increase the risk of drug resistance, and lead to multi-drug resistant HBV that may further limit treatment options (Moriconi et al., 2007). For long-term treatment, it is necessary to consider prevention or delay of drug resistance by developing efficacious therapy with low viral resistance. Drugs with high genetic barrier and/or low resistance such as entecavir are especially important in the prevention of drug resistance. Induction of sustained off-therapy virological and biochemical response at baseline with durable anti-HBe seroconversion in HBeAg-positive patients is a satisfactory end point (European Association for the Study of the Liver, 2012). However, entecavir is expensive, possibly tumorigenic, and no more efficacious than lamivudine in terms of HBeAg seroconversion (Liang, 2012), thereby limiting its

widespread usage.

Another strategy to prevent or delay HBV drug resistance involves combination pharmacotherapy, where two or more antiviral drugs are co-administered starting at treatment initiation. Clinicians throughout the world have tested various approaches to multi-drug therapy. There is clinical evidence showing that lamivudine combined with adefovir dipivoxil can be used to reduce the rate of drug resistance in newly-diagnosed treatment-naïve, chronic hepatitis B patients or in patients with lamivudine-resistance (Seto et al., 2012; Sung et al., 2008).

Ideally, combination HBV therapy should offer compounded or complementary efficacy, low cross-resistance, high resistance barrier, long-term safety, and economic feasibility. Even though lamivudine/adevovir combination therapy satisfies the former three requirements, it would nonetheless increase patients' economic burden and potentially induce multi-drug resistant HBV. Therefore, long-term usage of lamivudine/adevovir combination therapy may not be the preferred strategy for chronic hepatitis B.

Tan et al. (2012) considers YMDD variants as naturally-occurring mutations and lamivudine merely plays a selective role. Since adefovir belongs to a new generation of anti-HBV drugs, little is known about specific adefovir-resistant mutations. During the intensification phase, the combination group was co-administered lamivudine and adefovir, which managed both wild-type HBV as well as those with primary mutations, thereby preventing the selection of drug resistance HBV mutations in patients. The rate of development of HBV drug resistance is positively correlated with baseline viral load (Fung et al., 2009). Lamivudine has strong antiviral effects, where 30% of patients test negative for HBV DNA at three months, with peak of HBV DNA suppression at around 6 months. During the consolidation phase at 3 to 6 months, serum HBV DNA rapidly declines, thereby decreasing the likelihood of drug resistance.

Clinical studies indicate that the onset of HBV viral load suppression by adefovir is slower, requiring 2 to 3 months to reach the anticipated clinical response (Mao et al., 2007). However, in patients receiving lamivudine therapy for chronic hepatitis B, the YMDD mutation can be detected as early as 6 months (Liu et al., 2004). Given these time frames, a negative serum HBV DNA 3 to 6 months after initiation of combination lamivudine/adevovir therapy signals the an appropriate time to discontinue lamivudine therapy in order to reduce the rates of drug resistance and cross-resistance as well as the cost of long-term combination therapy as opposed to monotherapy with adefovir. This specific strategy and administration schedule of lamivudine/adevovir combination therapy takes advantage of the strengths of both individual drugs in order to maximize clinical efficacy and reduce drug resistance. With the YMDD mutation yet to appear and the ability of the rapidly declining HBV DNA levels at 12 weeks in predicting the long-term efficacy of adefovir (Hass et al., 2009), the possibility of future adefovir-resistance is significantly reduced while adefovir efficacy is potentially increased.

In general, pharmacoeconomic analysis consists of two essential elements: costs and outcome, which are, respectively the nominator and the denominator of the equation. The pharmacoeconomic analysis in this study was based on cost minimization, in which we compared the costs of two treatment modalities whose final outcome measure was the resolution of the Complete response rate composed of Rates of undetectable serum HBV DNA (virological response, VR), rates of HBeAg seroconversion and rates of ALT normalization (biochemical response, BR). The analysis showed that the costs of all items related to diminishing type antiviral combination of lamivudine and adefovir were lower than those related to the entecavir monotherapy (RMB ¥14,480.13 vs. RMB ¥28,818.47; $t=164.78, P < 0.001$).

Our results indicated that, during the first 12 weeks of

diminishing type antiviral combination of lamivudine and adefovir (LA), the HBV DNA suppression and ALT normalization rates were both comparable to the entecavir monotherapy (E) group. Both therapy strategies effectively inhibit viral replication and improve liver function. The difference was not statistically significant. At weeks 24 and 48 of treatment, the difference in VR and HBeAg seroconversion between LA group and E group was not significant, while similar results were observed for the BR. However, after 48 weeks, HBeAg seroconversion of LA group continued to improve. At 96 weeks, HBeAg seroconversion of LA group were higher than that of the E group, but the difference was still no statistically significant, while BR and VR in LA and E group were similar. At the same time, no virological breakthrough or drug resistance occurred in either of the two treatment groups by week 96 of the study. Both treatment strategies were well tolerated, with a low incidence of adverse reaction. One caveat of this study is that no matching placebos were employed in this study.

In summary, this study demonstrates that diminishing type antiviral combination of lamivudine and adefovir is economical, safe, and effective in HBeAg-positive chronic hepatitis B. We propose that combination therapy with lamivudine and adefovir for 12 to 24 week followed by long-term adefovir monotherapy be recommended for widespread usage in HBeAg-positive chronic hepatitis B. This strategy was worthy for further clinical application in countries, where ETV is not available or very expensive. A larger study is needed to determine the long-term advantages and disadvantages between the 2 groups.

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