Twenty-four-week Clevudine Therapy Showed Potent and Sustained Antiviral Activity in HBeAg-positive Chronic Hepatitis B

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Clevudine is a pyrimidine analogue with potent and sustained antiviral activity against HBV. The present study evaluated the safety and efficacy of 30 mg clevudine once daily for 24 weeks and assessed the durable antiviral response for 24 weeks after cessation of dosing. A total of 243 hepatitis B e antigen (HBeAg)-positive chronic hepatitis B patients were randomized (3:1) to receive clevudine 30 mg once daily (n = 182) or placebo (n = 61) for 24 weeks. Patients were followed for a further 24 weeks off therapy. Median serum HBV DNA reductions from baseline at week 24 were 5.10 and 0.27 $\log_{10}$ copies/mL in the clevudine and placebo groups, respectively ($P < 0.0001$). Viral suppression in the clevudine group was sustained off therapy, with 3.73 $\log_{10}$ reduction at week 34 and 2.02 $\log_{10}$ reduction at week 48. At week 24, 59.0% of patients in the clevudine group had undetectable serum HBV DNA levels by Amplicor PCR assay (less than 300 copies/mL). The proportion of patients who achieved normalization of alanine aminotransferase (ALT) levels was 68.2% in the clevudine group and 17.5% in the placebo group at week 24 ($P < 0.0001$). ALT normalization in the clevudine group was well maintained during post-treatment follow-up period. The incidence of adverse events (AEs) was similar between the clevudine group and the placebo group. No resistance to clevudine was detected with 24 weeks of administration of drug. *Conclusion:* A 24-week clevudine therapy was well tolerated and showed potent and sustained antiviral effect without evidence of viral resistance during treatment period in HBeAg-positive chronic hepatitis B. (HEPATOLOGY 2007;45:1172-1178.)

Abbreviations: AE, adverse event; cccDNA, covalently closed circular deoxyribonucleic acid; HBeAg, hepatitis B e antigen; ULN, upper limit of normal; WHV, woodchuck hepatitis virus.

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Chronic infection with HBV is common, globally affecting more than 350 million people, and frequently leads to serious consequences such as cirrhosis and hepatocellular carcinoma.[1] The aims of treatment of chronic hepatitis B are to achieve sustained suppression of HBV replication and remission of liver disease.[2] Since currently approved oral antiviral agents for chronic HBV infection do not provide a cure or durable remission in the majority of patients, there is a growing demand for new antiviral agents with improved efficacy. An ideal regimen should be safe with more potent and sustained viral suppression and with minimal chance of developing resistant mutants.

Clevudine [1-(2-deoxy-2-fluoro-β-L-arabinofuranosyl) thymine, L-FMAU] is a nucleoside analogue of the unnatural β-L configuration that has potent activity against HBV and some activity against Epstein-Barr virus in vitro.[3-5] The lack of cytotoxicity reflects the inability of human cellular DNA polymerases α, β, γ, and δ to utilize the 5′-triphosphate of clevudine as a substrate.[6,7] Moreover, clevudine was found to have no effect on mitochondrial structure, DNA content, or function.[6,7] Clevudine is efficiently phosphorylated by 3 intracellular enzymes to clevudine-triphosphate in target cells.[8] A unique advantageous characteristic of clevudine is prolonged sustained suppression of viral replication even after withdrawal of treatment. In woodchucks infected with woodchuck hepatitis virus (WHV), clevudine 10 mg/kg for 4 weeks led to prompt and profound viral suppression with up to 8 $\log_{10}$ reduction of plasma WHV DNA, which was sustained for more than 12 weeks after cessation of dosing.[9] Serum WHV DNA remained undetectable for up to 56 weeks after WHV-infected animals were given oral doses of 10 mg/kg daily clevudine for 12 weeks.[10] The sustained viral suppression has been demonstrated to be associated with significant reduction of covalently closed circular DNA (cccDNA) in hepatocytes.[11,12]

In a phase I/II dose-escalating clinical study[13] and a randomized phase II clinical trial,[14] 4-week and 12-week clevudine therapy produced potent viral suppression during therapy and induced a prolonged antiviral effect after withdrawal of treatment. The present study was conducted to evaluate the safety and efficacy of 30 mg clevudine once a day for 24 weeks and to assess the durable antiviral response for 24 weeks after cessation of dosing.

Patients and Methods

Study Design

This double-blind, randomized, placebo-controlled phase III study was conducted at 33 institutions in South Korea. Patients were randomized, based on a predetermined computer generated list, to receive clevudine 30 mg or placebo (3:1) once daily for 24 weeks with an additional 24-week follow-up period following the cessation of dosing.

The study was conducted in accordance with the principles of the Declaration of Helsinki and was consistent with Good Clinical Practice guidelines. Written informed consent was obtained from all study participants before being tested for eligibility criteria. The study protocol and the informed consent form were approved by the ethics committee at each study site and by the Korean Food and Drug Administration.

Patients were monitored at baseline, days 8, 15, and 29, and every 4 weeks thereafter during the dosing period. After week 24, patients were followed at weeks 28, 34, 40, and 48 without therapy. Patients underwent clinical assessments of tolerability (open-ended interview), physical exam, electrocardiography, and blood draws to measure laboratory parameters and serum HBV DNA levels.

Study Population

Eligible patients were adults, 18 to 60 years of age, with hepatitis B e antigen (HBeAg)-positive chronic hepatitis B, defined as the presence in serum of hepatitis B surface antigen for more than 6 months. Eligible patients also had serum HBV DNA levels more than 6 $\log_{10}$ copies/mL and serum ALT levels between 1.2 and 15 times the upper limit of normal (ULN). Exclusion criteria included coinfection with hepatitis C, hepatitis D, or the human immunodeficiency virus; evidence of cirrhosis or hepatocellular carcinoma; previous exposure to any nucleoside analog that is active against HBV; and use of interferon alpha within 6 months before enrollment. Breastfeeding or pregnant women or women of childbearing age unwilling to use barrier contraceptive methods were also excluded.

The sample size was calculated to detect 50% of statistically significant differences in the proportion of patients with serum HBV DNA below 4700 copies/mL at week 24 between the clevudine treatment group and the placebo group, assuming that placebo and clevudine responses were 10% and 60%, respectively. The sample size was estimated using the Z-test with an alpha level of 0.05 and 95% power for comparing placebo group with clevudine treatment group. The ratio of patient numbers in each group (placebo, clevudine 30 mg) was 1:3. According to this assumption, a sample size of 39 patients in the clevudine group and 13 patients in the placebo group was calculated. However, in order to evaluate the safety and tolerability of clevudine as well as its efficacy, a total of 248 patients, more than 4 times the calculated sample size, were enrolled.

Table 1. Patient Characteristics at Baseline

	Clevudine (n = 182)	Placebo (n = 61)	Total (n = 243)	P
Median age (years)	37.5	34	36	0.0947*
Sex, n (%)				0.059†
Male	149 (81.9)	43 (70.5)	192 (79.0)	
Female	33 (18.1)	18 (29.5)	51 (21.0)	
Median HBV DNA (log_{10} copies/mL)	8.29	8.38	8.32	0.3185*
Median ALT (U/L)	124	128	124	0.7546*
Mean ALT (U/L)	159.7	186.8	166.1	0.2718‡
ALT $< 2 \times$ ULN at baseline n (%)	48 (26.4)	16 (26.2)	64 (26.3)	0.6136§
ALT 2–5 $\times$ ULN at baseline n (%)	94 (51.7)	28 (45.9)	122 (50.2)	
ALT $>5 \times$ ULN at baseline n (%)	40 (22.0)	17 (27.9)	57 (23.5)	

*Wilcoxon 2-sample test. †χ^2-test. ‡t test. §Two-sided Fisher's exact test.

Efficacy End-points

The primary efficacy endpoint was reduction in serum HBV DNA, defined as a median log_{10} decrease from baseline at the end of treatment and during the follow-up period after withdrawal of treatment. Serum HBV DNA levels were measured at a central laboratory using the Digene Hybrid Capture II assay (Digene Corp., Gaithersburg, MD) with a lower limit of detection of 4700 copies/mL. When HBV DNA was undetectable by Digene Hybrid Capture II assay, the COBAS Amplicor PCR assay (Roche Molecular Systems, Branchburg, NJ) with a lower limit of detection of 300 copies/mL was used to measure lower levels of HBV DNA.

Secondary endpoints included the proportion of patients with undetectable HBV DNA, as measured by the COBAS Amplicor PCR assay; HBeAg loss; HBeAg seroconversion (HBeAg loss and appearance of hepatitis B e antibody); and serum ALT normalization.

Safety Analysis

The safety analysis included data from all 243 eligible patients who received at least 1 dose of study medication after randomization. Safety evaluations included analysis of adverse events (AEs), serious AEs, and deaths. Exacerbation of hepatitis B was defined as ALT and/or AST elevations (1) $>20 \times$ ULN or (2) $>10 \times$ ULN and a 10-fold change from the lowest on-study value. Exacerbation of hepatitis B was considered as a serious AE.

Genotypic Analysis

Genotypic analysis of the HBV DNA polymerase domain (amino acids 119 to 247) by dideoxy sequencing were performed at baseline and at the end of the 24-week dosing period. For detection of lamivudine-related YMDD (Tyrosine-Methionine-Aspartate-Aspartate) mutations at rt180 and rt204, RFLP (Restriction Fragment Length Polymorphism) assay were performed at baseline and at the end of the 24-week dosing period as described elsewhere.[15] HBV DNA breakthrough was defined as an increase in the level of HBV DNA of at least 1 log_{10} copies/mL from the lowest point while on treatment.

Statistical Analysis

Results were analyzed on the basis of intention-to-treat. All 243 eligible patients who received at least 1 dose of study medication after randomization were included in the safety analysis. Patients discontinuing the study after receiving the first study drug dose were included in the efficacy analysis until the time of their discontinuation.

The overall treatment comparison was assessed using the Wilcoxon 2-sample test for continuous data, and the chi-squared test or a 2-sided Fisher's exact test for categorical data. Statistical significance was performed with an alpha level of 0.05.

Results

Study Population

A total of 243 eligible patients were enrolled at 33 sites between June 23, 2003 and December 13, 2003 and received at least 1 dose of clevudine 30 mg (n = 182) or placebo (n = 61) in a blinded fashion. The 2 treatment groups were well balanced at the baseline (Table 1).

From 243 patients, 230 patients (173 in the clevudine group and 57 in the placebo group) completed the 24-week treatment period and 221 patients (171 in the clevudine group and 50 in the placebo group) completed the 48-week study period. A total of 11 patients discontinued the study in the clevudine group due to violation of eligibility criteria in 5 patients, withdrawal of consent in 3 patients, poor compliance in 2 patients, and medication error in 1 patient. A total of 11 patients discontinued the study in the placebo group due to withdrawal of consent in 6 patients, AEs in 2 patients, violation of eligibility criteria in 2 patients, and concomitant medication in 1 patient (Fig. 1).

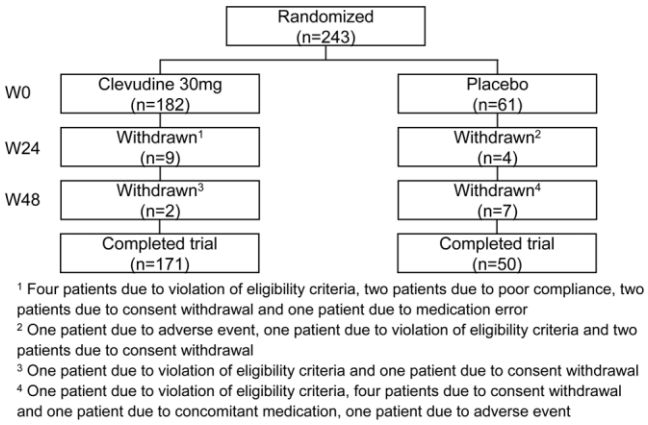

Fig. 1. Flowchart of the present study.

Virologic and Serologic End Points

Clevudine treatment for 24 weeks produced prompt and profound viral suppression. Median serum HBV DNA reductions from baseline at week 24 were 5.10 and 0.27 $\log_{10}$ copies/mL in the clevudine and placebo groups, respectively ($P < 0.0001$). Viral suppression in the clevudine group was sustained even after withdrawal of treatment with 3.73 $\log_{10}$ reduction at week 34 and 2.02 $\log_{10}$ reduction at week 48, compared with 0.51 $\log_{10}$ reduction at week 34 and 0.68 $\log_{10}$ reduction at week 48 in the placebo group ($P < 0.0001$) (Fig. 2).

At week 24, 102 of 173 of patients (59.0%) in the clevudine group but none of 57 patients in the placebo group had undetectable serum HBV DNA levels by Amplicor PCR assay (less than 300 copies/mL) ($P < 0.0001$).

At week 24, HBeAg loss and seroconversion occurred in 11.1% and 7.6%, respectively, of the patients in the clevudine group. These rates were similar to those in the placebo group (12.3% and 8.8%, respectively). At week 48, HBeAg loss and seroconversion occurred in 15.3% and 10.0%, respectively, of the patients in the clevudine group. These rates were also similar to those in the placebo group (12.0% and 12.0%, respectively).

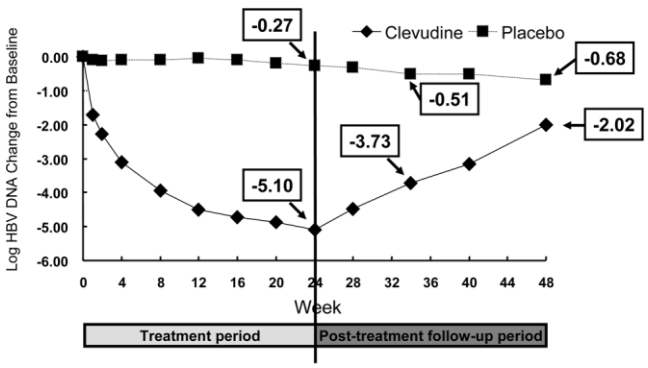

Fig. 2. Median $\log_{10}$ hepatitis B virus (HBV) DNA change from baseline.

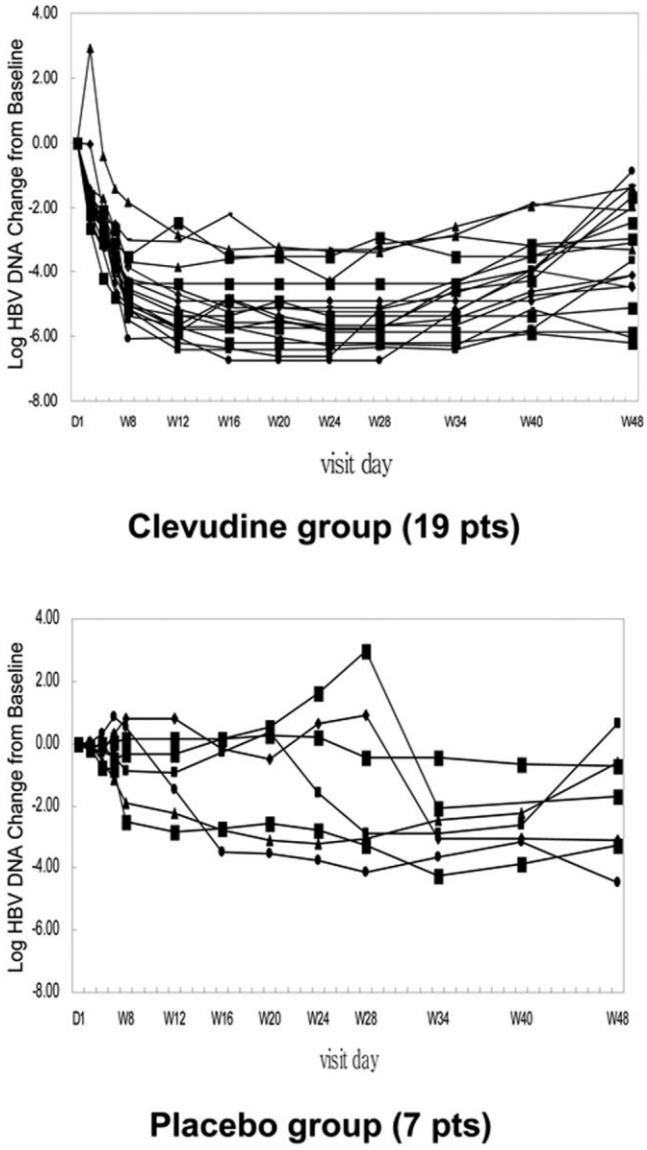

Fig. 3. Median $\log_{10}$ hepatitis B virus (HBV) DNA change from baseline in the patients with HBeAg loss at week 24.

In all the 19 patients in the clevudine group who achieved HBeAg loss during the 24-week treatment period, viral DNA levels were significantly reduced and well maintained after withdrawal of therapy. In contrast, 3 out of 7 patients in the placebo group with HBeAg loss showed no consistent reduction of viral DNA levels (Fig. 3).

Biochemical Endpoints

The proportion of patients who achieved normalization of ALT levels was 68.2% in the clevudine group and 17.5% in the placebo group at week 24 ($P < 0.0001$). In accordance with sustained viral suppression, ALT nor-

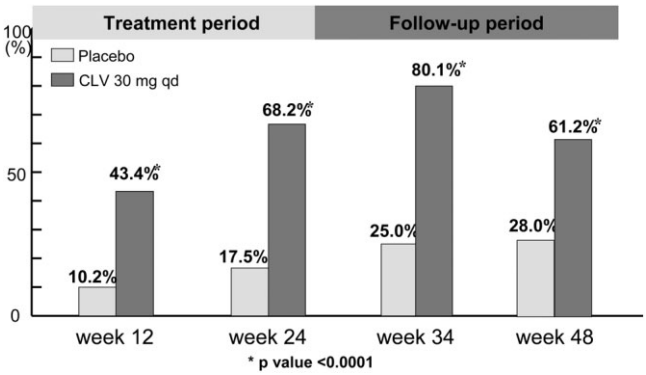

Fig. 4. Changes in the serum ALT normalization rate.

malization in the clevudine group was well maintained during the post-treatment follow-up period. The ALT normalization rates in the clevudine group increased further after withdrawal of therapy, up to 80.1% at week 34, and then decreased to 61.2% at week 48; these proportions were significantly higher than in the placebo group (Fig. 4).

Genotypic Analysis

Comparative analysis of genomic sequence of HBV isolated from the serum of the patients at baseline and at the end of 24 weeks dosing period showed 6 substitutions [rtA181A/T (n = 3), rtA181T (n = 2), and rtV191V/I (n = 1)] in conserved sites of the DNA polymerase domain in 5 patients in the clevudine group. However, these patients had consistently reduced serum HBV DNA levels during treatment and showed no evidence of HBV DNA breakthrough. In the placebo group, 4 nucleotide substitutions [rtL132M (n = 1), rtA181S (n = 1), and rtV191V/I (n = 2)] were observed in 4 patients.

Lamivudine-related YMDD mutations at rt180 and rt204 were not detected by RFLP assay at baseline and at the end of 24 weeks dosing period in both groups.

Safety and Tolerability

The median exposure to study drug was 167 days in the 2 groups, which was close to the intended exposure of 168 days. No discontinuation of treatment due to AEs occurred in the clevudine group. In the placebo group, there were 2 discontinuations of study due to AEs; exacerbation of hepatitis B in 2 patients.

The AEs reported during the treatment period and the post-treatment follow-up period are summarized in Table 2. During the treatment period, the incidence of AEs was similar in the 2 groups. The most frequent AEs, i.e., occurring in ≥5% of patients overall, were upper respiratory symptoms (14.3%), asthenia, dyspepsia, abdominal pain, and headache in the clevudine group. In the placebo group, the most frequent AEs were upper respiratory symptoms (16.4%), ALT increased (14.8%), asthenia, abdominal pain, dyspepsia, and diarrhea. The incidence of serious AEs during treatment was 5.0% in the clevudine group and 19.7% in the placebo group ($P < 0.001$). ALT elevations to a level >5 times the ULN were observed significantly less frequently in the clevudine group (19.2%) than in the placebo group (36.1%). Exacerbation of hepatitis B during treatment were observed in 4 patients in the clevudine group (2.2%) and 9 patients in the placebo group (14.8%) ($P < 0.0001$). In the clevudine group, all the exacerbations of hepatitis B were self-limiting with continued treatment without signs of hepatic decompensation. In the placebo group, 2 patients with exacerbation of hepatitis B discontinued participating in the study due to persistence of exacerbation and signs of hepatic decompensation.

During the post-treatment follow-up period, the incidence of AEs and serious AEs were similar in the 2 groups. However, ALT elevations to a level >5 times the ULN were observed significantly less frequently in the clevudine group (7.1%) than in the placebo group (19.7%).

Discussion

In this large, randomized placebo-controlled trial, clevudine 30 mg daily for 24 weeks showed highly potent antiviral activity in HBeAg-positive chronic hepatitis B patients. Clevudine suppressed HBV DNA by a median of 5.10 $\log_{10}$ copies/mL, and 59% of clevudine treated patients had undetectable levels of HBV DNA levels by PCR assay after 24 weeks of treatment. Although direct head-to-head comparisons are not available, the degree of HBV DNA reduction by clevudine at week 24 (5.10 $\log_{10}$ copies/mL) appears higher than those achieved by treatment with 100 mg lamivudine once daily at week 22 (about 3.2 $\log_{10}$ copies/mL),[16] 10 mg adefovir dipivoxil once daily at week 48 (3.57 $\log_{10}$ copies/mL),[17] 0.5 mg

Table 2. Summary of Cumulative Safety Data

Timing and event	No. of Patients (%) Clevudine (n = 182)	Placebo (n = 61)	*P*
During treatment			
Discontinuation due to adverse event	0	1 (1.6)	0.2510*
Any adverse event	95 (52.2)	36 (59.0)	0.3552†
Serious adverse event	9 (5.0)	12 (19.7)	0.0004†
ALT ≥ 5 × ULN	35 (19.2)	22 (36.1)	0.0072†
Exacerbation of hepatitis B	4 (2.2)	9 (14.8)	<0.0001*
During treatment-free follow-up			
Any adverse event	54 (29.7)	15 (24.6)	0.4463†
Serious adverse event	13 (7.1)	5 (8.2)	0.7807*
ALT ≥ 5 × ULN	13 (7.1)	12 (19.7)	0.0053†

*Two-sided Fisher's exact test. †χ^2-test.

entecavir once daily at week 22 (about 4.8 $\log_{10}$ copies/mL),[16] and 180 μg peginterferon alfa-2a once weekly at week 48 (4.5 $\log_{10}$ copies/mL).[18] However, in interpreting our HBV-DNA data, it must be kept in mind that the present study used 2 assays, including 1 hybridization test and 1 PCR-based assay for the assessment of HBV DNA levels, which makes data interpretation somewhat complicated. Unfortunately, the dynamic range of quantification of the 2 HBV DNA assays that were commercially available at the initiation time of the present clinical trial varied considerably, and neither covered the full range of HBV DNA values that can be observed in untreated and treated patients with chronic HBV.[19] In the ongoing trials, we can now use the more recently developed real-time PCR assays (HBV Beacom assay and COBAS TaqMan HBV), which have a dynamic range of 1.0×10^2 to 1.0×10^9 copies/mL.[20,21]

As shown previously in animal models[9,10] and by short-term phase II clinical trials,[13,14] clevudine has a peculiar characteristic of prolonged viral suppression after withdrawal of treatment. At 24 weeks after discontinuation of treatment, the median HBV DNA was still 2 logs below the baseline level. The mechanisms of delayed viral recrudescence remain to be elucidated. The measured plasma half-life of clevudine (43 to 61 hours) argues against its persistence in the blood.[13] In HepG2 cells, clearance of intracellular phosphorylated clevudine metabolites and clearance of intracellular phosphorylated lamivudine metabolites were similar (Cheng YC, unpublished results). These data negate the possibility that sustained viral suppression of clevudine may be due to retention of clevudine in blood or retention of phosphorylated clevudine metabolites in hepatocytes. The most plausible mechanism of sustained viral suppression may be that the potent suppression of viral replication leads to reduction in cccDNA in hepatocytes as observed in woodchucks.[11,12] With clevudine treatment, WHV cccDNA within hepatocytes declined more slowly than replicating viral DNAs, consistent with a half-life of 33 to 50 days.[11] The loss of cccDNA was comparable to that expected from the estimated death rate of hepatocytes in these woodchucks, suggesting that elimination of infected hepatocytes is one of the major mechanisms for reduction of cccDNA in clevudine treated woodchucks. In addition, clevudine therapy has been proven to break humoral and cell-mediated immune tolerance in chronic WHV infection by reducing viral and antigen load.[22] We previously demonstrated that episodes of acute exacerbations during the early treatment period in the clevudine-treated groups were associated with more prolonged viral suppression.[14] Collectively, these findings strongly suggest that the lack of rebound after clevudine therapy might be due to reduction of intrahepatic cccDNA through the elimination of HBV-infected hepatocytes. The actual measurement of clevudine triphosphate levels and cccDNA levels in hepatocytes from the patients undergoing clevudine therapy still remains to be investigated.

In accordance with potent and sustained viral suppression, clevudine treatment induced a high normalization rate of serum ALT levels at the end of the 24-week treatment period, and the ALT normalization was well maintained during the post-treatment follow-up period.

In this phase III study, clevudine was well tolerated for 24 weeks at the doses administered, with no specific pattern of AEs emerging during the study. Serious AEs including exacerbation of hepatitis B during treatment were significantly less frequent in the clevudine group than in the placebo group.

There was no emergence of drug resistance during the treatment. The rtA181A/T, rtA181T, and rtV191V/I substitutions observed in patients in the clevudine group were not associated with HBV DNA breakthrough. However, the resistance profile of clevudine remains to be determined by longer-term resistance surveillance studies.

The rtV191V/I, rtA181A/T, rtL132M, and rtA181S substitutions we found during our studies have not been described or characterized as resistant variants to lamivudine, adefovir, and entecavir. The rtA181T mutation was found in some patients treated with lamivudine and adefovir, but its relevance to resistance is not clear.[23,24]

Despite the potent HBV DNA suppression, the rates of HBeAg loss and seroconversion in the clevudine group were no better than in the placebo group. This may be attributed to the short period of clevudine therapy. Serologic response of clevudine should be determined by longer-term treatment studies. Interestingly, while HBeAg loss in the clevudine group was consistently associated with significant and persistent reduction of HBV DNA levels, HBeAg loss in the placebo group was not necessarily associated with consistent reduction of viral DNA levels.

In summary, a 24-week clevudine therapy was safe and well tolerated, and produced a highly potent and sustained antiviral effect associated with sustained normalization of ALT levels without emergence of drug resistance. We conclude that clevudine shows promising potential as an effective antiviral therapy for chronic HBV infection. Further studies are underway to investigate the safety and efficacy of clevudine at a larger scale and over a longer period.

References

1. Lee WM. Hepatitis B infection. N Engl J Med 1997;337:1733-1745.

2. Lok A, McMahon BJ. AASLD chronic hepatitis B: update of recommendations. HEPATOLOGY 2004;39:857-861.
3. Chu CK, Ma T, Shanmuganathan K, Wang C, Xiang Y, Pai SB, et al. Use of 2′-fluoro-5-methyl-β-L-arabinofuranosyluracil as a novel antiviral agent for hepatitis B virus and Epstein-Barr virus. Antimicrob Agents Chemother 1995;39:979-981.
4. Yao GQ, Liu SH, Chou E, Kukhanova M, Chu CK, Cheng YC. Inhibition of Epstein-Barr virus replication by a novel L-nucleoside, 2′-fluoro-5-methyl-β-L-arabinofuranosylurail. Biochem Pharmacol 1996;51:941-947.
5. Chin R, Shaw T, Torresi J, Earnest-Silveira L, Trautwein C, Bock T, et al. In vitro susceptibilities of wild-type and drug-resistant hepatitis B virus to (-)-β–D-2,6-diaminopurine dioxolane and 2′-fluoro-5-methyl-β-L-arabinofuranosyluracil. Antimicrob Agents Chemother 2001;45:2495-2501.
6. Pai SB, Liu SH, Zhu YL, Chu CK, Cheng YC. Inhibition of hepatitis B virus by a novel L-nucleoside, 2′-fluoro-5-methyl-β-L-arabinofuranosyluracil. Antimicrob Agents Chemother 1996;40:380-386.
7. Ma T, Pai SB, Zhu YL, Lin JS, Shanmuganathan K, Du J, et al. Structure-activity relationships of 1-(2-deoxy-2-fluoro-β-L-arabinofuranosyluracil) pyrimidine nucleosides as anti-hepatitis B virus agents. J Med Chem 1996; 39:2835-2843.
8. Liu SH, Grove KL, Cheng YC. Unique metabolism of a novel antiviral L-nucleoside analog, 2′-fluoro-5-methyl-β-L-arabinofuranosyluracil: a substrate for both thymidine kinase and deoxycytidine kinase. Antimicrob Agents Chemother 1998;42:833-839.
9. Peek SF, Cote PJ, Jacob JR, Toshkov IA, Hornbuckle WE, Baldwin BH, et al. Antiviral activity of clevudine [L-FMAU, (1-(2-fluoro-5–methyl β-L-arabinofuranosyl)uracil)] against woodchuck hepatitis virus and gene expression in chronically infected woodchucks (*Marmota monax*). HEPATOLOGY 2001;33:254-266.
10. Chu CK, Boudinot FD, Peek SF, Hong JH, Choi Y, Korba BE, et al. Preclinical investigation of L-FMAU as an anti-hepatitis B virus agent. Antivir Ther 1998;3(Suppl 3):113-121.
11. Zhu Y, Yamamoto T, Cullen J, Saputelli J, Aldrich CE, Miller DS, et al. Kinetics of hepadnavirus loss from the liver during inhibition of viral DNA synthesis. J Virol 2001;75:311-322.
12. Summers J, Mason WS. Residual integrated viral DNA after hepadnavirus clearance by nucleoside analog therapy. Proc Natl Acad Sci U S A 2004; 101:638-640.
13. Marcellin P, Mommeja-Marin H, Sacks SL, Lau GKK, Sereni D, Bronowicki JP, et al. A phase II dose escalating trial of clevudine in patients with chronic hepatitis B. HEPATOLOGY 2004;40:140-148.
14. Lee HS, Chung YH, Lee KS, Byun KS, Paik SW, Han JY, et al. A 12-week clevudine therapy showed potent and durable antiviral activity in HBeAg-positive chronic hepatitis B. HEPATOLOGY 2006;43:982-988.
15. Chayama K, Suzuki Y, Kobayashi M, Kobayashi M, Tsubota A, Hashimoto M, et al. Emergence and takeover of YMDD motif mutant hepatitis B virus during long-term lamivudine therapy and re-takeover by wild type after cessation of therapy. HEPATOLOGY 1998;27:1711-1716.
16. Lai CL, Rosmawati M, Lao J, Van Vlierberghe H, Anderson FH, Thomas N, et al. Entecavir is superior to lamivudine in reducing hepatitis B virus DNA in patients with chronic hepatitis B infection. Gastroenterology 2002;123:1831-1838.
17. Marcellin P, Chang TT, Lim SG, Tong MJ, Sievert W, Shiffman ML, et al. Adefovir dipivoxil for the treatment of hepatitis B e antigen-positive chronic hepatitis B. N Engl J Med 2003;348:808-816.
18. Lau GK, Piratvisuth T, Luo KX, Marcellin P, Thongsawat S, Cooksley G, et al. Peginterferon alfa-2a, lamivudine, and the combination for HBeAg-positive chronic hepatitis B. N Engl J Med. 2005;352:2682-2695.
19. Perrillo RP. Current treatment of chronic hepatitis B: benefits and limitations. Semin Liver Dis 2005;25:S20-S28.
20. Sum SS, Wong DK, Yuen MF, Yuan HJ, Yu J, Lai CL, et al. Real-time PCR assay using molecular beacon for quantitation of hepatitis B virus DNA. J Clin Microbiol 2004;42:3438-3440.
21. Lindh M, Hannoun C. Dynamic range and reproducibility of hepatitis B virus (HBV) DNA detection and quantification by Cobas Taqman HBV, a real-time semiautomated assay. J Clin Microbiol 2005;43:4251-4254.
22. Menne S, Ronecker CA, Korba BE, Gerin JL, Tennant BC, Cote PJ. Immunization with surface antigen vaccine alone and after treatment with 1-(2-fluoro-5-methyl-β-L-arabinofuranosyl)-uracil (L-FMAU) breaks humoral and cell-mediated immune tolerance in chronic woodchuck hepatitis virus infection. J Virol 2002;76:5305-5314.
23. Yeh C-T, Chien R-N, Chu C-M, Liaw Y-F. Clearance of the original hepatitis B virus YMDD-motif mutants with emergence of distinct lamivudine-resistant mutants during prolonged lamivudine therapy. HEPATOLOGY 2000;31:1318-1326.
24. Locarnini S, Qi X, Arterburn S, Snow A, Brosgart CL, Currie G, et al. Incidence and predictors of emergence of adefovir resistant HBV during four years of adefovir dipivoxil (ADV) therapy for patients with chronic hepatitis B [Abstract]. J Hepatol 2005;42:36A.