TREATMENT WITH LAMIVUDINE, ZIDOVUDINE, OR BOTH IN HIV-POSITIVE PATIENTS WITH 200 TO 500 CD4+ CELLS PER CUBIC MILLIMETER

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Abstract *Background.* The reverse-transcriptase inhibitor lamivudine has in vitro synergy with zidovudine against the human immunodeficiency virus (HIV). We studied the activity and safety of lamivudine plus zidovudine as compared with either drug alone as treatment for patients with HIV infection, most of whom had not previously received zidovudine.

Methods. Three hundred sixty-six patients with 200 to 500 CD4+ cells per cubic millimeter who had received zidovudine for four weeks or less were randomly assigned to treatment with one of four regimens: 300 mg of lamivudine every 12 hours; 200 mg of zidovudine every 8 hours; 150 mg of lamivudine every 12 hours plus zidovudine; or 300 mg of lamivudine every 12 hours plus zidovudine. The study was double-blind and lasted 24 weeks, with an extension phase for another 28 weeks.

Results. Over the 24-week period, the low-dose and high-dose regimens combining lamivudine and zidovudine were associated with greater increases in the CD4+ cell count ($P=0.002$ and $P=0.015$, respectively) and the percentage of CD4+ cells ($P<0.001$ for both) and with greater decreases in plasma levels of HIV type 1 (HIV-1) RNA ($P<0.001$ for both) than was treatment with zidovudine alone. Combination therapy was also more effective than lamivudine alone in lowering plasma HIV-1 RNA levels and increasing the percentage of CD4+ cells ($P<0.001$ for all comparisons), and these advantages persisted through 52 weeks. Adverse events were no more frequent with combination therapy than with zidovudine alone.

Conclusions. In HIV-infected patients with little or no prior antiretroviral therapy, treatment with a combination of lamivudine and zidovudine is well tolerated over a one-year period and produces more improvement in immunologic and virologic measures than does treatment with either agent alone. (N Engl J Med 1995;333:1662-9.)

ZIDOVUDINE is the recommended initial therapy for human immunodeficiency virus (HIV) infection[1] and is generally tolerated well.[2] However, clinical studies indicate that its beneficial effects are limited in duration,[3] partly because of the development of resistance by HIV.[4,5] Combination therapy with antiretroviral agents may have more sustained antiviral effects, decrease the emergence of drug resistance, and affect a wider range of cellular or tissue reservoirs of HIV infection.[6] Studies evaluating treatment with a combination of two reverse-transcriptase inhibitors have found favorable effects on CD4+ counts and plasma viremia.[7-9] Even in early, asymptomatic disease, there is substantial ongoing replication of HIV type 1 (HIV-1),[10-12] and turnover both of HIV-1 in plasma and of HIV-1–producing cells is generally high, with a half-life on the order of two days.[13,14] There is a need for potent, safe antiretroviral therapy, which may be achieved by using agents in combination.

Lamivudine, or (−)-2′-deoxy-3′-thiacytidine (also known as 3TC), a reverse-transcriptase inhibitor, has in vitro activity against a range of isolates of HIV-1, including virus resistant to zidovudine.[15-17] Lamivudine and zidovudine are synergistic in vitro.[18] A mutation in the HIV-1 polymerase gene at codon 184 that is selected when lamivudine is present confers resistance to that drug.[19-21] Viruses with this mutation remain sensitive to zidovudine.[19,20] The in vitro introduction of the mutation into virions containing mutations that confer resistance to zidovudine resensitizes those viruses to zidovudine.[19] In phase 1 studies lamivudine has had high oral bioavailability and is well tolerated.[22,23] We studied two doses of lamivudine in combination with zidovudine in patients with little or no prior antiretroviral therapy who had 200 to 500 CD4+ cells per cubic millimeter.

METHODS

Study Population

Patients were found to be seropositive for HIV-1 by a standard enzyme-linked immunosorbent assay, with confirmation by the Western blot assay. Eligible patients had received zidovudine for four weeks or less, were 12 or more years of age, had Karnofsky scores of 60 or above, and had 200 to 500 CD4+ cells per cubic millimeter. The criteria for exclusion from the study included results of liver-function tests more than five times the upper limit of the normal range, an absolute neutrophil count below 1000 cells per cubic millimeter, a hemoglobin level below 9.2 g per deciliter for male patients or below 8.8 g per deciliter for female patients, a positive serum pregnancy test for female patients, prior therapy with antiretroviral drugs other than zidovudine, and active opportunistic infections or cancers requiring systemic therapy. Patients who had had an illness considered to define the presence of the acquired immunodeficiency syndrome (AIDS) but who no longer required therapy for the acute illness were allowed to enroll in the study.

Study Design and Treatment Regimens

The study was a double-blind, randomized, multicenter, placebo-controlled trial lasting 24 weeks, with a blinded extension phase for an additional 28 weeks, conducted at 26 sites in North America. Approval was obtained from the investigational review

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*The other members of the North American HIV Working Party are listed in the Appendix.

Table 1. Base-Line Characteristics of the Study Patients and Outcomes after 24 Weeks of Study.*

Characteristic	Zidovudine Only (N = 93)	Lamivudine Only (N = 87)	Combination Therapy		P Value
			low dose (N = 92)	high dose (N = 94)	
Median age — yr	34.0	34.4	33.9	33.9	0.855†
Sex — % of patients					
Male	87	83	92	87	0.292‡
Female	13	17	8	13	
Race or ethnic group — % of patients					0.938‡
White	61	60	64	59	
Black	16	18	16	15	
Hispanic	20	22	18	24	
Other	2	0	1	2	
CD4+ cells					
No./mm^3					
Mean	349±11.4	340±12.8	366±11.8	380±11.5	0.109†
Median	336	332	360	372	
Percent of all lymphocytes					
Mean	20.7±0.8	20.7±0.7	21.0±0.7	21.6±0.6	0.441†
Median	19.3	19.6	21.3	21.8	
Positive ICD p24 antigen assay — % of patients	40	34	33	26	0.157§
HIV RNA					
No. of patients	90	80	88	93	
Mean no. of copies/ml	87,500	79,200	87,100	80,800	
Median no. of copies/ml	45,500	41,400	25,900	33,200	
Mean log copies/ml	4.6±0.1	4.5±0.1	4.4±0.1	4.4±0.1	0.387†
No. of patients with ≥20,000 copies/ml	63	54	49	58	0.251§
Asymptomatic — % of patients	74	73	68	76	0.265‡
Discontinued study drug early — no. of patients (%)	25 (27)	19 (22)	23 (25)	24 (26)	0.887§
Adverse events	11	6	8	4	0.278§
New AIDS-defining illness	1	0	0	0	
Causing withdrawal from study	2	1	1	1	
Other	8	5	7	3	
Lost to follow-up	11	7	10	13	0.677§
Other reasons for discontinuation¶	3	6	5	7	

*Plus–minus values are means ±SE. ICD denotes immune-complex–dissociated. Because of rounding, percentages do not all total 100.

†By the van Elteren test, with control for the study site.

‡By the Cochran–Mantel–Haenszel test, with control for the study site.

§By an exact test.

¶Reasons include noncompliance and the decision of the patient or the investigator not to continue the drug.

board at each institution, and the patients gave written informed consent.

Patients were randomly assigned to receive one of four oral treatments: 200 mg of zidovudine (Retrovir, Glaxo Wellcome, Research Triangle Park, N.C.) every 8 hours plus placebo resembling lamivudine (the zidovudine-only group); 300 mg of lamivudine (Epivir, Glaxo Wellcome) every 12 hours plus placebo resembling zidovudine (the lamivudine-only group); 150 mg of lamivudine every 12 hours plus 200 mg of zidovudine every 8 hours (the low-dose combined-therapy group); or 300 mg of lamivudine every 12 hours plus 200 mg of zidovudine every 8 hours (the high-dose combined-therapy group).

Adverse events were managed in accordance with predetermined guidelines, and the severity of the events was graded according to the criteria of the AIDS Clinical Trials Group.[24]

Evaluation of Patients

Patients were seen during the screening period, at randomization, and at week 2, week 4, and every four weeks thereafter. A full history was taken, and a physical examination performed, at the time of screening and at weeks 24 and 52. At each visit medications were reviewed and vital signs, Karnofsky scores, and results of laboratory tests of safety were obtained. All female patients had pelvic examinations and Pap smears at the start of the study and at weeks 24 and 52 to evaluate the progression of possible cervical disease. All laboratory studies were performed by Roche Biomedical Laboratories (Research Triangle Park, N.C., and Raritan, N.J.).

Assessments were also made when a patient discontinued the use of the study drug and four weeks later. Patients who discontinued the study drug were requested to return for CD4+ cell counts every four weeks through week 24.

Measures of HIV-1 Disease

The measures of HIV-1 disease used in the study included assays of T-lymphocyte subgroups, reverse-transcriptase polymerase-chain-reaction (PCR) assays for HIV-1 RNA in plasma (Roche), the immune-complex–dissociated HIV p24 antigen assay (Coulter, Hialeah, Fla.), and assays for β_2-microglobulin and neopterin. The threshold level of detection for HIV-1 RNA was 200 copies per milliliter. Lower values were recorded as 200 copies per milliliter. The primary outcome measures evaluated were changes from base line in levels of HIV-1 RNA and CD4+ cell counts in the treatment groups.

Statistical Analysis

The patients were randomly assigned to the four treatment groups in equal numbers at each participating center with the use of permuted blocks. A summary measure[25] was used to characterize the profile of HIV disease markers over the first 24 weeks of the study. The time-weighted area under the curve (as calculated by the trapezoidal rule) minus the base-line value was used to compare the groups with respect to the data collected over time during the first 24 weeks of the study. For patients who completed the 52 weeks of the study, the average of the last three evaluations (at weeks 44, 48, and 52) minus the base-line value was used as a summary measure in comparing the groups with respect to the durability of the treatment effect. The summary measures were compared by the van Elteren test,[26] an extension of the Wilcoxon two-sample test that permits stratified analyses, after stratification according to study site. Primary pairwise comparisons between each treatment group and the zidovudine-only group were adjusted with use of the Bonferroni correction for multiple comparisons. All P values were two-sided.

A secondary analysis among the patients who had 20,000 or more copies of HIV-1 RNA per milliliter at base line used the reverse-transcriptase PCR assay to compare the proportions who had a reduction by 2 log from the base-line value in at least one measurement made at week 2, 4, 8, 12, 16, 20, or 24; this analysis used the method of Wei and Johnson.[27] A similar analysis was used to compare the proportions of patients with a reduction of 50 percent from base line on the immune-complex–dissociated p24 antigen assay. Exact tests were used to compare the groups with respect to the incidence of adverse events during as many as 30 days after permanent discontinuation of the study drug. The patients' base-line characteristics were compared by a Cochran–Mantel–Haenszel test after stratification according to study site, or by the van Elteren test after the same stratification. The times to the permanent discontinuation of the study

drug and the times to the first severe or life-threatening laboratory abnormality or clinical adverse event as defined in the protocol were plotted with Kaplan–Meier curves and compared by the log-rank test. All the analyses of efficacy were performed on an intention-to-treat basis.

Four scheduled interim evaluations of the safety data were reviewed by an independent Data and Safety Monitoring Board. The study data were analyzed as planned after the last randomized patient had completed week 24 of the study.

RESULTS

Demographic and Base-Line Variables

A total of 366 patients were randomly assigned to the four treatment groups. The groups were well balanced with respect to demographic variables (Table 1). Thirteen percent of the patients were female, and 39 percent belonged to minority groups. Base-line characteristics of HIV-1 disease were similar among the four groups (Table 1). From 11 to 20 percent of the patients in each group had previously received antiretroviral therapy (zidovudine only), and the median duration of that therapy in the four groups was three weeks or less (P = 0.22).

Table 2. Time-Weighted Changes from Base Line in Immunologic and Virologic End Points during 24 Weeks of Study.*

End Point	Zidovudine Only	Lamivudine Only	Combination Therapy	
			low dose	high dose
Primary				
Absolute change in CD4+ count				
No. of patients†	85	80	85	85
Mean no. of cells/mm^3	16.6±9.4	23.7±7.4	54.9±11.3	44.9±8.6
Median no. of cells/mm^3	11.9	24.0	66.3	41.3
P value (vs. zidovudine only)‡	—	0.52	0.002§	0.015§
P value (vs. lamivudine only)¶	—	—	0.025	0.061
Change in log HIV RNA				
No. of patients†	86	77	80	88
Mean no. of copies/ml	−0.31±0.03	−0.59±0.04	−1.12±0.07	−1.15±0.07
Median no. of copies/ml	−0.31	−0.60	−1.20	−1.10
P value (vs. zidovudine only)‡	—	<0.001§	<0.001§	<0.001§
P value (vs. lamivudine only)¶	—	—	<0.001§	<0.001§
Secondary				
Change in CD4+ as a percentage of all lymphocytes				
No. of patients†	85	81	83	85
Mean % change	1.6±0.3	1.5±0.3	3.6±0.4	4.2±0.4
Median % change	2.0	1.1	3.2	4.2
P value (vs. zidovudine only)‡	—	0.12	<0.001§	<0.001§
P value (vs. lamivudine only)¶	—	—	<0.001§	<0.001§

*Means and medians shown are the changes from base line in the time-weighted area under the curve for the measure indicated during the 24 weeks of the study. Positive numbers denote increases, and negative numbers decreases. Plus–minus values are means ±SE.

†Numbers shown are the numbers of patients for whom values were obtained at randomization and at least twice during the study and who were included in the analysis.

‡By primary pairwise comparison of the median values shown, with use of the van Elteren test with control for study site.

§This P value was significant after the use of the Bonferroni correction for multiple comparisons (adjusted level of significance, 0.05 ÷ 3, or 0.017).

¶By secondary pairwise comparison of the median values shown, with use of the van Elteren test with control for study site.

Among the 366 patients initially randomized, 275 (75 percent) were still receiving the study medication at week 24 (Table 1). Kaplan–Meier plots of the time to the permanent discontinuation of the study drug revealed no significant difference among treatment groups in rates of discontinuation over the 52-week period (P = 0.76; data not shown). Eleven percent of the patients initially randomized were lost to follow-up (Table 1).

There were no significant differences between the patients who completed the 24-week study period and those who did not with respect to age, sex, the absolute CD4+ count, the percentage of CD4+ cells, or the HIV-1 RNA concentration (data not shown). Nor did the median absolute CD4+ count and the log concentration of HIV-1 RNA at base line differ statistically within each treatment group between the patients who completed 24 weeks of the study and those who did not (data not shown).

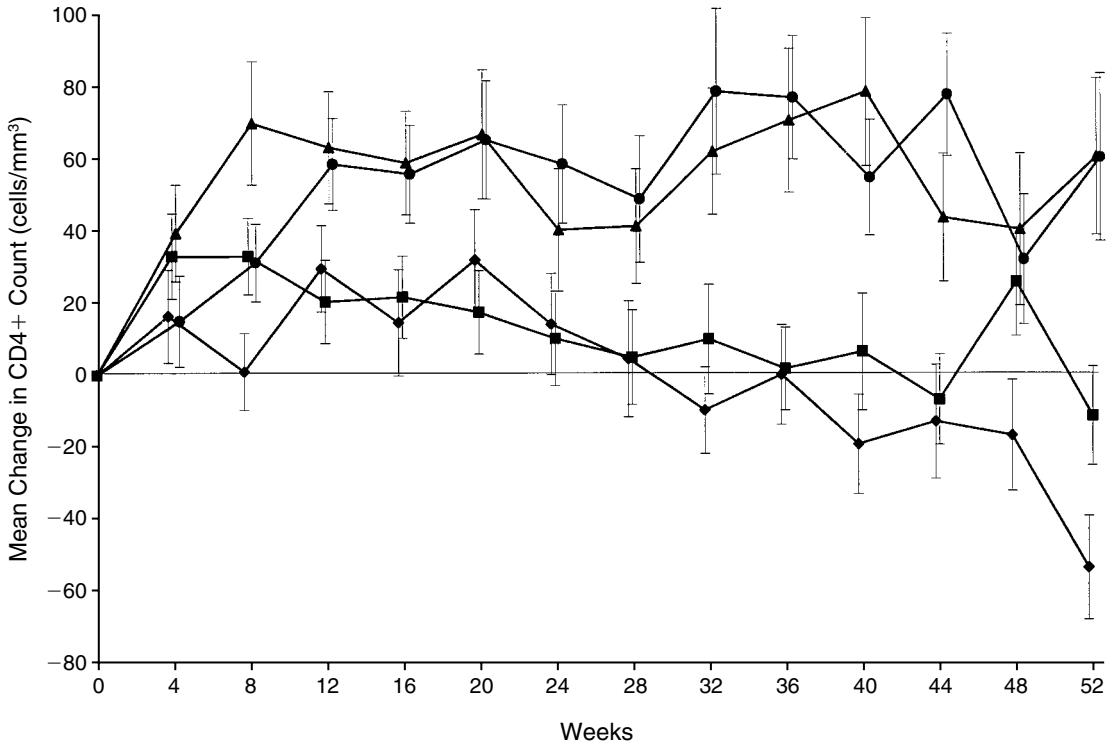
Immunologic and Virologic Effects

Immunologic Activity

The median time-weighted increases from base line in the absolute CD4+ count during the first 24 weeks of the study are shown in Table 2. The differences between the zidovudine-only group and each of the combination-therapy groups were significant. When the change in the absolute CD4+ count in the lamivudine-only group was compared with the change in each combination-therapy group, the differences were not statistically significant. There was no statistically significant difference between the two combination-therapy groups or between the two monotherapy groups.

The greatest mean (±SE) change in the absolute CD4+ count was 32±14 cells per cubic millimeter in the zidovudine-only group, 33±11 cells per cubic millimeter in the lamivudine-only group, 70±17 cells per cubic millimeter in the low-dose combination-therapy group, and 65±16 cells per cubic millimeter in the high-dose combination-therapy group. At 24 weeks, the mean change from base line in the CD4+ count was 14±14 cells per cubic millimeter in the zidovudine-only group, 10±13 cells per cubic millimeter in the lamivudine-only group, 40±17 cells per cubic millimeter in the low-dose combination-therapy group, and 58±16 cells per cubic millimeter in the high-dose combination-therapy group (Fig. 1).

At week 52, data from 45 to 55 percent of the patients in each group were available for analysis, partly because some patients had not completed the extension phase by the time of the analysis. Among the patients who could be studied, the mean increases in the CD4+ count at week 52 were 61±22 and 60±23 cells per cubic millimeter in the low-dose and high-dose combination-therapy groups, respectively. At 52 weeks the zidovudine-only group had a mean decrease of 53±14 cells per cubic millimeter in the CD4+ count, resulting in a difference of approximately 114 cells per cubic millimeter between that group and either combination-therapy



PATIENTS WHO COULD BE EVALUATED

Zidovudine only	(–◆–)	93	87	82	80	73	71	68	69	63	60	60	57	55	46
Lamivudine only	(–■–)	87	81	74	75	68	68	67	65	62	57	56	62	54	48
Low-dose combined therapy	(–▲–)	92	84	75	77	71	71	68	71	68	60	58	58	53	46
High-dose combined therapy	(–●–)	94	87	78	77	76	66	69	61	60	56	58	49	47	42

Figure 1. Mean (±SE) Changes from Base Line in Absolute CD4+ Counts, According to the Week of the Study.
The number of patients shown for each week in each treatment group is the number who could be evaluated at that time. After week 24, the numbers of patients indicate the numbers available for study at each point in the analysis; the numbers do not indicate rates of withdrawal from the study. Some patients had not completed the extended phase of the study by the time of this analysis.

group (Fig. 1). When we assessed the durability of the response, both combination-therapy groups had a more sustained increase in the absolute CD4+ cell count than the zidovudine-only group (Table 3). The changes from base line in the percentage of CD4+ cells (among all lymphocytes) followed a pattern similar to that of the changes in the absolute CD4+ count (Tables 2 and 3).

Antiretroviral Activity

The median 24-week time-weighted change from base line in the plasma concentration of HIV-1 RNA was a decrease of 50 percent in the zidovudine-only group and of 75 percent in the lamivudine-only group, whereas there were decreases of 94 and 92 percent in the low-dose and high-dose combination-therapy groups, respectively (Table 2). The differences between the combination-therapy groups and the monotherapy groups were significant, but the difference between the two combination-therapy groups was not. The effect of lamivudine monotherapy was significantly greater than that of zidovudine monotherapy over the first 24 weeks, primarily because of a greater initial reduction in the viral load. The greatest mean reductions in the plasma concentration of HIV-1 RNA were 0.52±0.04 log in the zidovudine-only group, 1.19±0.07 log in the lamivudine-only group, 1.56±0.10 log in the low-dose combination-therapy group, and 1.55±0.09 log in the high-dose combination-therapy group. At week 24, the mean decrease was 0.25±0.06 log in the zidovudine-only group, 0.42±0.05 log in the lamivudine-only group, 0.90±0.09 log in the low-dose combination-therapy group, and 0.99±0.09 log in the high-dose combination-therapy group (Fig. 2).

There were significantly greater sustained reductions in HIV-1 RNA concentrations in both combination-therapy groups over the 52-week period than in either monotherapy group (Table 3). There was no significant difference in effect between the combination-therapy groups or between the monotherapy groups.

In a subgroup of 224 patients who began the study with 20,000 or more copies of HIV-1 RNA per milliliter, the maximal mean decreases in HIV-1 RNA were 0.55 log (median, 0.60) in the zidovudine-only group and 1.18 log (median, 1.37) in the lamivudine-only group. The maximal mean decreases were 1.87 log (me-

Table 3. Durability of the Response to Treatment over the 52 Weeks of the Extended Study.*

End Point	Zidovudine Only	Lamivudine Only	Combination Therapy	
			low dose	high dose
Primary				
Absolute change in CD4+ count				
No. of patients	52	56	50	43
Mean no. of cells/mm^3	−27.8±12.0	5.3±11.0	52.0±18.4	59.4±15.0
Median no. of cells/mm^3	−26.5	5.8	69.4	66.0
P value (vs. zidovudine only)†	—	0.35	<0.001‡	<0.001‡
P value (vs. lamivudine only)§	—	—	0.073	0.02‡
Change in log HIV RNA				
No. of patients	38	42	33	36
Mean no. of copies/ml	−0.18±0.07	−0.36±0.07	−0.80±0.11	−1.03±0.10
Median no. of copies/ml	−0.20	−0.29	−0.81	−1.00
P value (vs. zidovudine only)†	—	0.31	<0.001‡	<0.001‡
P value (vs. lamivudine only)§	—	—	0.007‡	<0.001‡
Secondary				
Change in CD4+ as a percentage of all lymphocytes				
No. of patients	52	56	50	43
Mean % change	0.3±0.6	1.0±0.4	5.8±0.8	6.2±0.7
Median % change	0.4	0.4	5.0	6.2
P value (vs. zidovudine only)†	—	0.79	<0.001‡	<0.001‡
P value (vs. lamivudine only)§	—	—	0.005‡	0.003‡

*Means and medians shown in this analysis were calculated from the average of the values obtained at the last three visits during the 52-week period for each available patient, minus the base-line value for that patient. Positive numbers denote increases, and negative numbers decreases. Plus–minus values are means ±SE.

†By primary pairwise comparison of the median values shown, with use of the van Elteren test with control for study site.

‡This P value was significant after the use of the Bonferroni correction for multiple comparisons (adjusted level of significance, 0.05 ÷ 3, or 0.017).

§By secondary pairwise comparison of the median values shown, with use of the van Elteren test with control for study site.

dian, 2.11) in the low-dose combination-therapy group and 1.88 log (median, 2.06) in the high-dose combination-therapy group, a reduction of 99 percent. The percentage of patients with at least one HIV-1 RNA measurement during treatment that showed a reduction of 2 log or more was higher in the combined-therapy groups than in either monotherapy group (2 percent of patients receiving zidovudine only, 9 percent of those receiving lamivudine only, 61 percent of those receiving low-dose combination therapy, and 69 percent of those receiving high-dose combination therapy; $P<0.001$ for the comparison of low-dose combination therapy with zidovudine monotherapy).

In general, comparisons of the 24-week time-weighted average levels of neopterin and β_2-microglobulin obtained with zidovudine monotherapy and either dose of combination therapy favored the combination therapy (data not shown). Among the patients whose levels of immune-complex–dissociated p24 antigen at randomization were at least double the lower limit of the assay, the proportion who had at least a 50 percent reduction in p24 antigen was significantly higher in the low-dose combination-therapy group (63 percent) than in the zidovudine-only group (32 percent, $P=0.011$).

Safety

The median follow-up in the treatment groups ranged from 320 to 364 days. The percentages of patients who had serious adverse events attributed to any cause or who were withdrawn from the study because of adverse events are shown in Table 4. Serious adverse events did not occur more often in any one organ system than in the others (data not shown). There were no significant differences among treatment groups in the time to the first adverse event ($P=0.217$ by the log-rank test). However, adverse events occurred more rapidly in the three groups receiving zidovudine than in the lamivudine-only group ($P=0.044$). There were three non–drug-related deaths during the study.

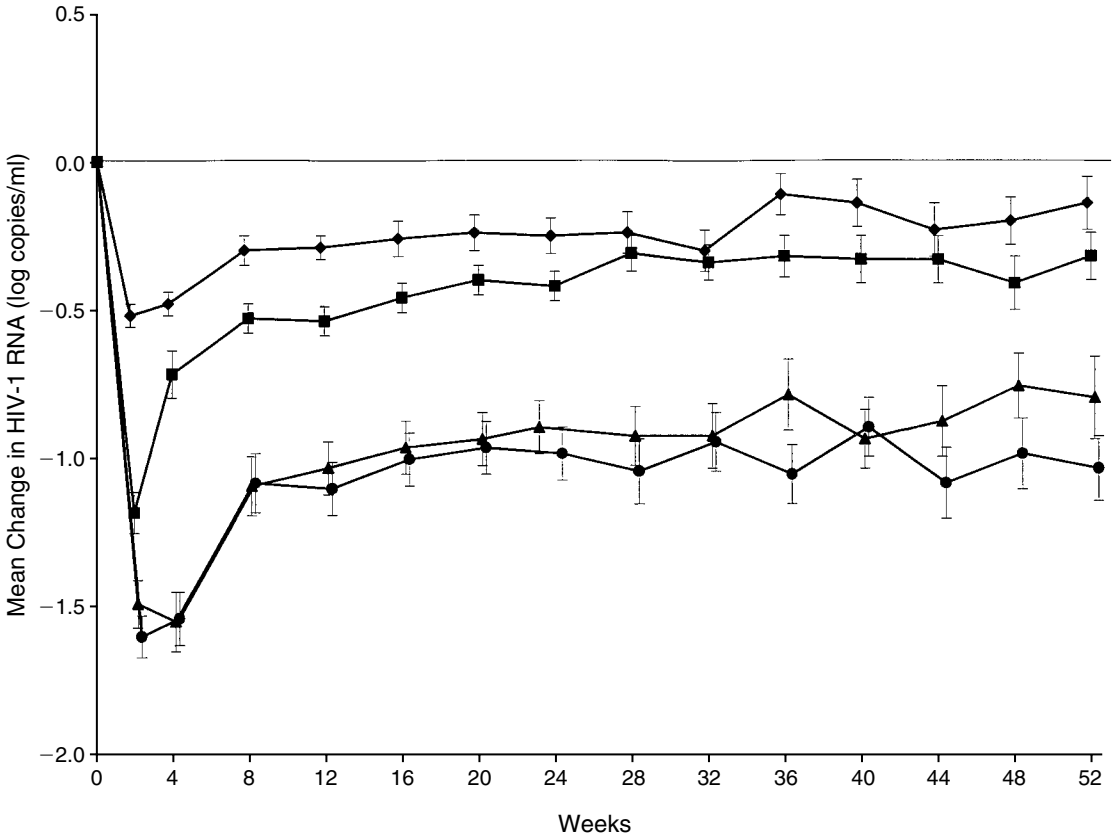
The clinical adverse events attributed to study drugs were most commonly gastrointestinal effects, usually nausea, and were more frequent in the three groups receiving zidovudine ($P=0.06$ for all gastrointestinal events, and $P<0.001$ for nausea). Of 29 patients withdrawn from the study during the first 24 weeks because of clinical adverse events, 14 were withdrawn because of gastrointestinal side effects and 9 were withdrawn because of malaise and fatigue.

Neurologic adverse events, predominantly headache, were the second most common class of events attributed to the study drugs, and they were distributed evenly among treatment groups. Peripheral neuropathy was usually mild; two patients in the zidovudine-only group and one in the lamivudine-only group required an interruption of the dose, with only one permanent withdrawal. There were no withdrawals due to pancreatitis.

Two patients were withdrawn because of anemia, one in the zidovudine-only group and one in the low-dose combination-therapy group. Nineteen patients (5 percent), none of whom received lamivudine monotherapy, had absolute neutrophil counts below 750 cells per cubic millimeter. No other severe laboratory abnormality affected more than 5 percent of the study population. The results of liver-function tests tended to be mildly elevated; 85 percent of all abnormal alanine aminotransferase values measured were within five times the upper limit of normal.

Discussion

The results of this study show the potent antiretroviral activity of lamivudine in combination with zidovudine in HIV-infected patients with limited or no previous use of zidovudine. The combinations of lamivudine and zidovudine resulted in significantly greater and more sustained decreases in plasma levels of HIV-1 RNA over the 24 weeks of the study than did treatment with either drug alone. A persistent decrease in plasma levels of HIV-1 RNA, by 0.8 to 1 log, was observed in the combination-therapy groups even through week 52.



PATIENTS WHO COULD BE EVALUATED

Zidovudine only	(–◆–)	90	83	81	77	73	68	63	62	50	43	39	36	35	35	32
Lamivudine only	(–■–)	80	75	71	71	67	67	61	60	47	45	41	31	39	35	34
Low-dose combined therapy	(–▲–)	88	80	77	71	68	65	63	59	42	38	30	31	31	31	29
High-dose combined therapy	(–●–)	93	87	87	75	75	72	63	66	44	41	40	39	34	34	34

Figure 2. Mean (±SE) Changes from Base Line in the Log Concentration of HIV RNA, According to the Week of the Study.
The number of patients shown for each week in each treatment group is the number who could be evaluated at that time. After week 24, the numbers of patients indicate the numbers available for study at each point in the analysis; the numbers do not indicate rates of withdrawal from the study. Some patients had not completed the extended phase of the study by the time of this analysis.

When the change in HIV-1 RNA levels was analyzed in the 224 patients who had base-line levels 2 log or more above the threshold of detection of the reverse-transcriptase PCR assay, the median peak decrease in levels among patients receiving combination therapy at either dose was 2.1 log, and approximately two thirds of the patients in the combination-therapy groups had at least one value that was 2 log or more below the base-line value during the study. This double-blind, randomized clinical trial used changes in plasma levels of HIV-1 RNA as a primary end point. The plasma level of HIV-1 RNA is a strong predictor of the progression of HIV-1 infection, independently of the CD4+ cell count.[28,29] In addition, retrospective analyses of several prospective clinical trials have shown that a reduction in the plasma level of HIV-1 RNA in response to therapy is an independent predictor of clinical benefit.[30-32]

The decreases in HIV-1 RNA levels were accompanied by substantial increases from the base-line CD4+ cell count over the first 24 weeks of the study in patients who received the combination therapy at either dose, as compared with zidovudine monotherapy. Unlike many studies evaluating other therapies,[3,7-9,33-36] our study showed sustained increases from the base-line CD4+ cell count with both combination treatments and no return toward base-line values in patients followed for 52 weeks. The difference between the combination groups and the zidovudine-only group in the mean increase from base line at 52 weeks was more than 100 cells per cubic millimeter. Although conflicting results have been reported,[2] several earlier studies of antiretroviral therapy have shown clinical benefit when the therapy being evaluated produced changes in CD4+ cell counts[33-36] that were either less substantial or less prolonged than the changes we observed. Given the greater magnitude and duration of the effect of treatment combining lamivudine and zidovudine on the CD4+ cell count and the viral burden, it is possible that this combination will

Table 4. Occurrence of Adverse Events According to Treatment Group.*

Event	Zidovudine Only (N = 93)	Lamivudine Only (N = 87)	Combination Therapy		P Value†
			low dose (N = 92)	high dose (N = 94)	
			no. of patients (%)		
Withdrawal due to adverse event	11 (12)	7 (8)	7 (8)	8 (9)	0.771
Serious adverse event‡	12 (13)	7 (8)	10 (11)	8 (9)	0.678
Death	2 (2)	1 (1)	0	0	—
Severe clinical toxicity§	10 (11)	5 (6)	7 (8)	8 (9)	0.687
Severe laboratory toxicity§	21 (23)	13 (15)	24 (26)	20 (21)	0.32

*The median number of patient-days studied in the analysis was 340.

†By a two-tailed exact test.

‡Includes fatal, life-threatening, disabling, or incapacitating events; events that resulted in hospitalization or prolonged it; or any congenital anomaly, cancer, or overdose, regardless of the relation attributed to the study drug.

§Regardless of the relation attributed to the study drug. Severe denotes grade III or IV in the toxicity grading scale of the AIDS Clinical Trials group.

have a more sustained clinical benefit than those reported previously. However, prospective clinical trials with sufficient power to detect differences in clinical end points are needed to substantiate this hypothesis.

Several factors may explain the observed effects of combined therapy with lamivudine and zidovudine. Both drugs are potent inhibitors of HIV-1 and act synergistically against primary clinical isolates in vitro.[37] Because the phosphorylation of lamivudine and zidovudine differs in resting and activated CD4+ lymphocytes, they may target different populations of infected cells.[38] The sustained antiviral effect of the combination may also be due in part to the reduction in the development of resistance to zidovudine. In a parallel study comparing zidovudine monotherapy with the combination of lamivudine plus zidovudine in previously untreated patients, mutations confirming resistance to zidovudine appeared more slowly in the patients treated with the drug combination.[39,40] In vitro selection of the methionine-to-valine mutation at amino acid 184 of the reverse-transcriptase enzyme restores susceptibility to zidovudine in molecular clones that carry mutations conferring zidovudine resistance.[19,40] Thus, interactions between drug-resistance mutations in the HIV-1 *pol* gene may contribute to the prolonged effect of the combined therapy.

The overall effects on plasma levels of HIV-1 RNA and CD4+ cell counts did not differ significantly between the two monotherapy groups, despite the probability that lamivudine-resistant variants of HIV-1 would emerge rapidly in the patients receiving lamivudine monotherapy.[41] Explanations for the observed effect of this monotherapy are speculative. The mutation conferring resistance to lamivudine does result in an amino acid change at the active site of the reverse-transcriptase enzyme in a motif that is highly conserved across most retroviruses[42] and could result in virions with impaired replication.[43,44]

An important finding of this study was the low frequency of serious adverse events in any treatment group. Lamivudine monotherapy was well tolerated. Mean hemoglobin levels increased in patients receiving this therapy (data not shown), and no episodes of severe neutropenia were observed. Adding lamivudine to zidovudine did not alter the safety profile observed with zidovudine alone. There were no significant differences in adverse events or immunologic or antiviral effects according to the dosage of combination therapy, and therefore the low dose should be used in treatment and in further clinical trials.

In summary, treatment combining lamivudine and zidovudine may be appropriate as the initial therapy for asymptomatic HIV-infected patients who have 200 to 500 CD4+ cells per cubic millimeter. This conclusion is based on the pronounced and durable effect of the combination treatment, as compared with zidovudine monotherapy, on virologic and immunologic markers of HIV-1 infection and on the excellent tolerability of the two drugs in combination. This conclusion is supported by evidence that asymptomatic patients, in general, have a high overall burden of HIV-1[10-12] that is undergoing rapid turnover.[13,14] The early institution of potent combination therapy may be needed to limit viral replication and delay both the development of resistance to antiretroviral drugs and the clinical progression of infection.[45] The data from this study show that combinations of lamivudine and zidovudine have a sustained effect on CD4+ cell counts and viral replication in HIV-1–infected patients.

Appendix

In addition to the study authors, the North American HIV Working Party included B. Akil, G. Beall, N. Bellos, P.S. Berry, C. Brummitt, B. Cameron, C. Cohen, H. Donabedian, K.H. Mayer, G.F. McKinley, G.E. Sepulveda, M. Thompson, C. Tsoukas, S. Walmsley, J. Horton, T. Kerkering, E. Matthew, D. Pearce, D. Peterson, J.C. Pottage, Jr., J. Sampson, W.B. Smith, and B. Yangco. The study personnel at the individual sites were J. Johnson, J. Cowan, C. Kaczka, S.H. Price, R. Zameck, D. Dawson, B. Pobiner, C. Gilbert, J. Scott-Lennox, R. Demasi, P. Jarrett, G. Yuen, J. Esinhart, and G. Pakes.

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