CONCISE COMMUNICATION

Virological and Immunological Effects of Combination Antiretroviral Therapy with Zidovudine, Lamivudine, and Indinavir during Primary Human Immunodeficiency Virus Type 1 Infection

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Forty-seven patients presenting with primary human immunodeficiency virus (HIV) infection were treated with zidovudine 200 mg 3 times a day, lamivudine 150 mg 2 times a day, and indinavir 800 mg 3 times a day for 1 year. From a mean pretreatment viral RNA level of 4.93 $\log_{10}$ copies/mL, the proportions of patients having <500 copies/mL at 24 and 52 weeks were 92.0% and 89.2%, respectively. For the 35 patients with data available at 24 and 52 weeks, the corresponding proportions for the <50 copies/mL analysis were 86.6% and 79.3%, respectively. The change in virus load was −2.19 and −2.41 $\log_{10}$ copies/mL at weeks 8 and 52, respectively. CD4 cell counts increased, from a mean of 546 cells/mm^3, by 142 cells/mm^3 at week 24 and by 210 cells/mm^3 at week 52. Three patients discontinued the study because of drug-related toxicity. Six (12.8%) patients had adverse experiences associated with nephrolithiasis. Combination therapy with zidovudine, lamivudine, and indinavir during primary HIV infection results in a profound and sustained reduction in virus load with concurrent recovery of the CD4 cell population.

Primary human immunodeficiency virus (HIV) infection represents a dynamic phase of HIV-1 infection: an intact immune system is challenged with a rapidly replicating lentivirus that preferentially infects and destroys cells that coordinate the host's ability to respond to the virus [1]. After primary infection, it is thought that most of the immunopathological processes that will eventually lead to AIDS have been established [2]. It is during this primary infection phase that long-lived $CD4^+$ lymphocytes become infected with HIV [3], making viral eradication unlikely, even with years of potent antiretroviral therapy. The immunological and virological outcomes following primary infection are thought to determine the rate of subsequent disease progression [4]. Increased severity of symptoms and signs of primary HIV infection, low CD4 nadir, and high levels of circulating plasma HIV RNA following the seroconversion illness are all associated with rapid disease progression [5]. This would suggest that those patients presenting or identified with an illness associated with primary HIV infection are likely to show rapid progression of the disease, providing a theoretical rationale for initiation of antiretroviral therapy during primary HIV-1 infection. Zidovudine, lamivudine, and indinavir have demonstrated clinical, immunological, and virological benefit in chronic HIV infection [6]. We therefore evaluated this antiretroviral combination in patients diagnosed with primary HIV-1 infection.

Methods

A multinational open-label study was conducted at 6 sites in the United States, Europe, and Australia. The study objective was to evaluate the efficacy of zidovudine, lamivudine, and indinavir, as measured by the rate of suppression of serum HIV RNA to below the level of detection. The secondary objective was to evaluate the

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All participants gave written informed consent. This study conformed to the national requirements for human experimentation in the United States, Australia, Switzerland, Belgium, and Italy.

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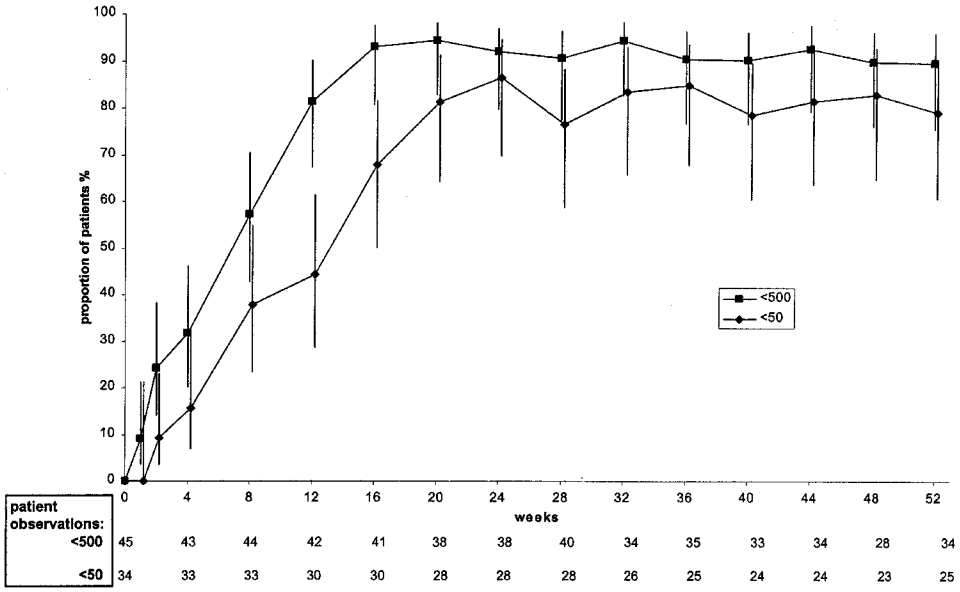

Figure 1. Results of intention-to-treat analysis, showing proportions (%) of patients with viral RNA levels of <500 and <50 copies/mL. *Vertical bars,* 95% confidence intervals.

safety and tolerability of this combination in patients with primary HIV infection. The protocol was approved by an institutional review board for each site.

Study population. For the purposes of this study, primary HIV infection was defined as at least 1 clinical and 1 laboratory feature of primary infection. Clinical criteria included symptoms and signs of acute retroviral syndrome, history of such symptoms within the past 70 days, or exposure within the past 3 months and a negative antibody test within the past 6 months. Laboratory criteria were detectable HIV RNA in either plasma or serum, an indeterminate Western blot test, a negative EIA, or a low-positive EIA with increasing reactivity over time. All patients had to be >18 years of age, with no pulmonary infections within the past 21 days and routine laboratory parameters not >1.5 times the upper limit of normal. All women of childbearing potential were required to have a negative pregnancy test before enrollment.

Study medications were as follows: zidovudine 200 mg 3 times a day, lamivudine 150 mg 2 times a day, and indinavir 800 mg 3 times a day. In instances of zidovudine intolerance, we permitted substitution with stavudine. Patients were assessed weekly for the first 4 weeks, then monthly through the first year.

Laboratory assessments. Samples were collected for centralized batch testing of viral RNA (vRNA) with both the Roche Amplicor assay (detection level 500 RNA copies/mL) and the ultradirect assay (detection level 50 RNA copies/mL) (Amplicor; Roche Diagnostic Systems, Branchburg, NJ). $CD4^+$ and $CD3^+$ lymphocytes were assessed locally with flow cytometric assays.

Adherence assessment. Patients returned all unused medication for counting at each subsequent visit and documented, on diary cards, all doses they ingested.

Statistical analyses. The primary efficacy analysis evaluated the proportions of patients with vRNA levels of <500 copies/mL (Amplicor assay) at each week of the study. Secondary analyses focused on the proportions of patients with vRNA levels of <50 copies/mL (ultradirect assay) and on changes from baseline in vRNA and CD4 cell count. The data were analyzed with intention-to-treat model-based approaches. Specifically, for the analyses of proportions of patients with vRNA levels of <500 and <50 copies/mL, patients who discontinued the study for therapy-related reasons (adverse experience or poor vRNA response) were scored as virologic failures from the time of discontinuation onward, and the required proportions were then determined by use of a longitudinal generalized estimating equations approach [7] that also used available data for patients who discontinued therapy for nontherapy-related reasons. A similar approach was used for the CD4 and vRNA change from baseline analyses. For the latter, to calculate changes from baseline only, a value of 500 was used for vRNA assay results reported as <500 copies/mL, and a value of 250 was used for results reported as negative (i.e., no amplifiable signal).

Because stavudine could be substituted for zidovudine as part of toxicity management, it was not possible to calculate adherence for zidovudine. Adherence was measured for indinavir and lamivudine with the following equation: percentage compliance = 100 × (total doses taken while on study/total dose that could have been taken, assuming that there was no change in dosing). Cumulative frequency distributions for percentage compliance were then graphed separately for each drug.

Results

Forty-seven patients were enrolled in the following cities: New York, 12; Seattle, 10; Sydney, 8; Geneva, 6; Brussels, 4; Basel, 3; Zurich, 2; and Milan, 2. Only 3 women were enrolled. The mean age of participants was 32 years (range, 20–59 years), and the majority of patients ($n = 42$) were white. Forty patients

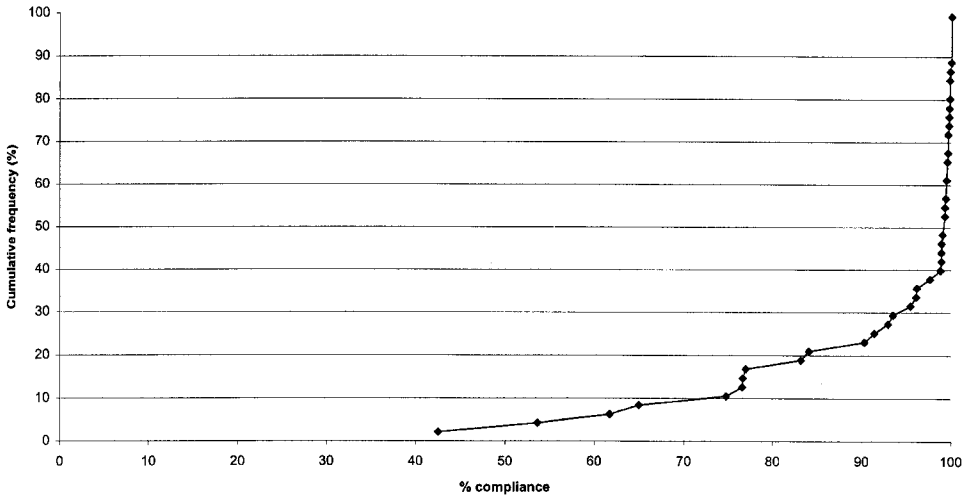

Figure 2. Cumulative frequency distribution of percentage compliance with indinavir treatment

had typical symptoms of primary HIV infection, either at screening (21) or within the past 70 days (19); 4 patients were asymptomatic but had a negative test within the past 6 months. Of these 47 subjects, 37 (79%) completed 52 weeks of treatment with zidovudine (or stavudine), lamivudine, and indinavir; 10 (21%) patients discontinued because of adverse events ($n = 3$), withdrawal from study ($n = 5$), or loss to follow-up ($n = 2$). No patient discontinued therapy because of virological failure.

Virological response. Expressed as change from a mean baseline of 4.93 $\log_{10}$ copies/mL (SD, 0.97 $\log_{10}$ copies/mL; range, 2.79–6.93 $\log_{10}$ copies/mL), there was a mean drop of 1.85 $\log_{10}$ copies/mL at week 4, 2.33 $\log_{10}$ copies/mL at week 24, and 2.41 $\log_{10}$ copies/mL at week 52.

The proportions of patients with a vRNA level of <500 copies/mL, found by use of the model-based approach, are shown in figure 1. In this analysis, the proportions of patients with a vRNA level of <500 copies/mL were 92% at 24 weeks and 89.8% at 52 weeks. Corresponding proportions for the <50 copies/mL analysis (figure 1) were 86.6% at 24 weeks and 79.3% at 52 weeks (because the ultradirect assay was not performed at 1 site with 12 patients, this figure is based on 35 patients). There were no subjects continuing on protocol therapy who had increasing HIV RNA levels.

Immunological response. CD4 counts increased rapidly after initiation of treatment, with a model-based increase of 103 cells/mm^3 by week 2 from a baseline of 535 cells/mm^3 (SD, 175.3 cells/mm^3). By week 32, the estimated increase had almost doubled to 211 cells/mm^3. This increase was essentially maintained through week 52 (210 cells/mm^3 above baseline).

Safety. All 47 patients in the study had at least 1 clinical adverse experience during the trial; 45 of these were judged to be possibly, probably, or definitely related to study drugs. Three (6.4%) patients had clinical serious adverse experiences. Renal colic requiring hospitalization 4 months after therapy initiation in 1 patient (attributed to indinavir) and a constellation of asthenia/fatigue, myalgia, headache, and anemia 4 months after therapy began in a second patient were judged to be definitely drug related; we attributed them to zidovudine. A third patient with a history of depression experienced aggressive behavior 5 months after therapy initiation; this patient attempted suicide with an overdose of study drugs 10 months into the study. This event was not felt to be related to the study therapy. One of 3 patients with serious adverse experiences and an additional 2 subjects withdrew from the trial because of nausea or flank pain. Clinical or laboratory evidence felt to be related to nephrolithiasis was noted in 6 (12.8%) patients. The most common (>20% incidence) adverse experiences considered to be drug related included fatigue or asthenia, abdominal pain, diarrhea, nausea, vomiting, headache, and dry skin. Although it was not prospectively addressed in this study, clinical features of lipodystrophy were noted in 1 subject.

No patient died during this study. No HIV progression to Centers for Disease Control and Prevention category B or C [8] occurred during the study, although the patient who discontinued therapy because of renal colic developed herpetic esophagitis 56 weeks after discontinuing all antiretroviral drugs.

Adherence. Adherence (mean ± SD) to the 3-times-a-day regimen for indinavir during the 52-week study period was 92.2% ± 13.5% and 93.0% ± 15.4% for lamivudine. Figure 2 is a graph of the cumulative frequency distributions for indinavir adherence. Seventy-five percent of patients who received indinavir and lamivudine were at least 99% adherent, as assessed by pill counts of returned medication. A total of 17% of patients had <80% adherence with indinavir, and 13% of patients were <80% adherent with the twice-daily lamivudine. No association was found between compliance and a plasma virus load of <500 copies/mL at week 52 or at time of study drop out. Surprisingly, 6 of 8 subjects with poor compliance

(<80% indinavir compliance) had a virus load of <500 copies/mL at study completion or at time of discontinuation, suggesting that, among these patients, good virological suppression could be achieved despite suboptimal compliance.

Discussion

This study illustrates the rapid and sustained reduction in HIV RNA after initiation of combination antiretroviral therapy during primary HIV-1 infection. More than 90% of subjects achieved vRNA levels of <500 copies/mL within 12 weeks of beginning therapy and maintained virus suppression throughout follow-up. Commensurate with this reduction in circulating virus was a resurgence of $CD4^+$ lymphocytes. There was a rapid increase of ~100 $CD4^+$ cells at week 2, followed by a more gradual recovery throughout the remainder of follow-up.

Although all subjects enrolled in this trial reported some clinically adverse experiences, there were few drug-related serious adverse experiences. Only 3 (6.4%) of 47 subjects discontinued therapy because of drug-related clinical experiences. Fatigue and gastrointestinal symptoms were common but did not appear to decrease adherence to therapy or to impact the virologic response to therapy.

Interestingly, in patients starting their first antiretroviral combination, adherence in this study was high, with 75% of patients reporting at least 99% adherence to indinavir. The absence of virologic failures in this study may be attributed in part to excellent adherence with this regimen. A recent study of zidovudine, lamivudine, and ritonavir in patients with primary HIV infection concluded that adherence in the setting of patients undergoing seroconversion was difficult, with 5 (41.7%) of 12 patients in that study remaining on combination regimens at 12 months [9].

Aggressive combination treatment for primary HIV infection has become a standard recommendation [10], despite the lack of clinical trial data supporting this approach. Nonrandomized open-label studies of combination therapy during primary HIV infection have suggested that patients at this phase of infection respond well to treatment [11]. Unfortunately, increasing awareness of long-term toxicities seen with antiretroviral therapy is a concern [12], particularly if these patients are expected to remain on therapy indefinitely.

The perceived sense of urgency in treating patients with primary HIV infection was originally based on the supposition that such patients may be able to achieve viral eradication if treatment were initiated early enough and continued for long enough [13]. Unfortunately, the discovery of long-lived latently infected lymphocytes has made viral eradication with antiretroviral agents an unachievable goal at present [5]. Others have argued that initiating treatment during primary infection may be the only way to preserve HIV-specific T cell function [1], possibly allowing discontinuation of therapy at some future time. Rapid viral rebound and possible recurrence of symptomatic primary HIV infection has been noted on discontinuation of treatment [14]. Alternatively, the significance of an anecdotal report of a recently infected patient remaining in virological remission after stopping therapy has also been widely debated [15]. Whether early aggressive treatment of primary HIV infection will ultimately alter the course of disease is yet to be determined; further studies of immediate versus deferred therapy are needed to evaluate this potential.

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