

## CONTROLLED TRIAL OF INTERLEUKIN-2 INFUSIONS IN PATIENTS INFECTED WITH THE HUMAN IMMUNODEFICIENCY VIRUS

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### ABSTRACT

**Background** Interleukin-2 is a cytokine that regulates the proliferation and differentiation of lymphocytes. In preliminary studies, intermittent infusions of interleukin-2 led to increases in CD4 counts in patients with human immunodeficiency virus (HIV) infection and more than 200 CD4 cells per cubic millimeter. We conducted a controlled study to evaluate the long-term effects of such therapy on both CD4 counts and the viral burden.

**Methods** Sixty HIV-infected patients with base-line CD4 counts above 200 cells per cubic millimeter were randomly assigned to receive either interleukin-2 plus antiretroviral therapy (31 patients, 1 of whom was lost to follow-up) or antiretroviral therapy alone (29 patients). Interleukin-2 was administered every two months for six cycles of five days each, starting at a dosage of 18 million IU per day. Safety and immunologic and virologic measures were monitored monthly until four months after the last treatment cycle.

**Results** In patients treated with interleukin-2, the mean ( $\pm$ SE) CD4 count increased from  $428 \pm 25$  cells per cubic millimeter at base line to  $916 \pm 128$  at month 12, whereas in the control group, the mean CD4 count decreased from  $406 \pm 29$  cells per cubic millimeter to  $349 \pm 41$  ( $P < 0.001$ ). There were no significant differences between the groups in serial measurements of the plasma HIV RNA or p24 antigen concentration during the 12 months of treatment. Constitutional symptoms (fever, malaise, and fatigue) and asymptomatic hyperbilirubinemia were the chief dose-limiting toxic effects of interleukin-2 therapy.

**Conclusions** In patients with HIV infection and base-line CD4 counts above 200 cells per cubic millimeter, intermittent infusions of interleukin-2 produced substantial and sustained increases in CD4 counts with no associated increase in plasma HIV RNA levels. (N Engl J Med 1996;335:1350-6.)

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THE symptoms of infection with the human immunodeficiency virus (HIV) result from a progressive immunodeficiency due to the destruction of CD4 T lymphocytes, which ultimately renders HIV-infected patients susceptible to opportunistic infections and malignant disorders. Immune-system-based approaches to the treatment of HIV infection use pharmacologic augmentation of immunity in an attempt to prevent, delay, or reverse this deterioration.<sup>1-3</sup> This approach is likely to be increasingly useful as the suppression of viral rep-

lication by combinations of new, potent antiviral agents facilitates expansion of the immune system.<sup>4-7</sup>

Interleukin-2 is a cytokine secreted by activated T lymphocytes that regulates the proliferation and differentiation of lymphocytes, including CD4 T cells.<sup>8-11</sup> Although measurement of the viral burden indicates how fast the immune system will decline, the number or percentage of CD4 cells remains the best single indicator of the capacity of the immune system to prevent the development of opportunistic infections.<sup>12-16</sup> In an uncontrolled pilot study, intermittent interleukin-2 therapy resulted in sustained increases in the number of CD4 cells, primarily in patients with base-line CD4 counts greater than 200 cells per cubic millimeter.<sup>17</sup> The immune-system activation induced by interleukin-2 therapy was associated with transient increases in the plasma HIV load in some patients. To validate these preliminary observations, and to determine the long-term effects of intermittent interleukin-2 therapy on the HIV load, we carried out a randomized, controlled trial of intermittent interleukin-2 therapy in HIV-infected patients with CD4 counts greater than 200 cells per cubic millimeter.

### METHODS

#### Study Design

Patients 18 years or older who had HIV type 1 infection and CD4 counts above 200 cells per cubic millimeter at screening were eligible for enrollment if they had never received interleukin-2, had no history of an opportunistic infection defined as one indicating progression to the acquired immunodeficiency syndrome (AIDS), and had received no corticosteroids, cytotoxic chemotherapy, or experimental therapy in the preceding four weeks. The study was approved by the institutional review board of the National Institute of Allergy and Infectious Diseases (NIAID), and all patients provided written informed consent. The study was reviewed twice by a five-member data and safety monitoring board.

Patients were randomly assigned to receive antiretroviral thera-

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The U.S. government has been issued a patent for immunologic enhancement with intermittent interleukin-2 therapy, listing Drs. Kovacs and Lane as inventors.

py plus intermittent intravenous infusions of interleukin-2 or antiretroviral therapy alone. For purposes of randomization, patients were stratified according to the CD4 count (>500 or ≤500 cells per cubic millimeter) and plasma HIV RNA concentration (<10,000 or ≥10,000 HIV RNA copies per milliliter) measured at screening. The study was not placebo controlled, so as to avoid unnecessary hospitalizations for the intravenous infusion of placebo, given that the constitutional side effects of interleukin-2 make blinding impossible.

Patients were evaluated monthly for 14 months after randomization. Patients treated with interleukin-2 were hospitalized at the Warren Grant Magnuson Clinical Center every other month, from month 0 through month 10, for the administration of six cycles of interleukin-2. Interleukin-2 (aldesleukin; Proleukin, Chiron, Emeryville, Calif.) was diluted in 5 percent aqueous dextrose solution containing 0.1 percent albumin and administered by continuous infusion, starting at a dose of 18 million IU per day for five days, with dose reductions of 3 to 6 million IU as needed when there was clinical or laboratory evidence of toxicity.<sup>17</sup> Safety and immunologic and virologic measures were assessed monthly for all patients; in the interleukin-2 group, laboratory values were obtained before each cycle of interleukin-2 was begun. At the end of 14 months, all patients were eligible to receive interleukin-2.

Antiretroviral therapy (with zidovudine, didanosine, zalcitabine, or stavudine, alone or in combination) was provided to all patients, and the agents used could be changed at any time during the course of the study. The specific regimen was determined by the patient and the referring physician, in consultation with the study team.

Measurement of lymphocyte subgroups and identification of surface markers were performed according to the guidelines of the Centers for Disease Control and Prevention.<sup>18</sup> Particle-associated plasma HIV RNA concentrations were determined with use of the branched-chain DNA assay (Chiron).<sup>19,20</sup> Levels of p24 antigen were determined by an immune-complex-dissociated assay (Coulter, Hialeah, Fla.).<sup>21</sup>

**Statistical Analysis**

The primary end point of the study was the effect of intermittent interleukin-2 therapy on the CD4 count over time. Secondary end points included changes in the plasma HIV load, level of p24 antigenemia, percentage of CD4 cells, and number and percentage of CD8 cells. A sample of 60 patients was considered adequate to provide 90 percent power to detect a difference of 35 cells per cubic millimeter in the CD4 count after one year, with a type I error of 0.05, assuming a 20 percent dropout rate. Changes over time for each individual patient were summarized for each variable by the difference between average base-line and post-treatment measurements and by the ordinary least-squares estimate of the slope of the measurements over time. Base-line values (month 0) were the means of the measurements made at two screening visits and the first study visit. Data were analyzed with use of the SAS System for Microsoft Windows, release 6.10 (SAS Institute, Cary, N.C.), according to the group to which the patient was originally randomly assigned (intention-to-treat analysis). Two-sample t-tests (for unequal variances) were used to test for differences between the group means; the results were confirmed with the Wilcoxon rank-sum test. All statistical tests were two-sided, and a P value ≤0.05 was considered to indicate statistical significance.<sup>22</sup>

To ensure that group differences were not attributable to observed imbalances at base line, we also tested the effect of treatment while controlling for base-line covariates (CD4 count, viral load, and history of antiviral therapy) by means of multiple linear regression.

**RESULTS**

Sixty patients were enrolled in the study between April and December 1993. Thirty-one were randomly assigned to the interleukin-2 group, and 29 to the control group. The base-line characteristics of

**TABLE 1. BASE-LINE CHARACTERISTICS ACCORDING TO STUDY GROUP.**

| CHARACTERISTIC*                           | INTERLEUKIN-2 GROUP (N=31) | CONTROL GROUP (N=29) |
|---|----------------------------|----------------------|
| Age (yr)                                  |                            |                      |
| Mean                                      | 38                         | 37                   |
| Range                                     | 23-52                      | 23-49                |
| Sex (no.)                                 |                            |                      |
| Male                                      | 30                         | 29                   |
| Female                                    | 1                          | 0                    |
| Race or ethnic group (no.)                |                            |                      |
| Non-Hispanic white                        | 28                         | 28                   |
| Non-Hispanic black                        | 1                          | 1                    |
| Hispanic                                  | 2                          | 0                    |
| HIV risk group (no.)†                     |                            |                      |
| Homosexual or bisexual                    | 29                         | 28                   |
| Recipient of blood-product transfusion    | 1                          | 0                    |
| Heterosexual                              | 2                          | 1                    |
| Weight (kg)                               |                            |                      |
| Mean                                      | 74                         | 76                   |
| Range                                     | 56-87                      | 58-97                |
| Duration of antiretroviral therapy        |                            |                      |
| 0-6 mo (no.)                              | 3                          | 5                    |
| >6 mo (no.)                               | 28                         | 24                   |
| Mean (mo)                                 | 34                         | 29                   |
| Range (mo)                                | 2-64                       | 2-74                 |
| White-cell count (cells/mm <sup>3</sup> ) |                            |                      |
| Mean                                      | 4649                       | 4830                 |
| Range                                     | 3167-6267                  | 2400-7667            |
| Hemoglobin (g/dl)                         |                            |                      |
| Mean                                      | 13.7                       | 14.1                 |
| Range                                     | 11.3-15.3                  | 11.4-15.9            |
| CD4 count (cells/mm <sup>3</sup> )        |                            |                      |
| Mean                                      | 427                        | 406                  |
| Range                                     | 188-753                    | 206-888              |
| CD4 percentage                            |                            |                      |
| Mean                                      | 26                         | 26                   |
| Range                                     | 11-51                      | 9-52                 |
| CD8 count (cells/mm <sup>3</sup> )        |                            |                      |
| Mean                                      | 949                        | 987                  |
| Range                                     | 184-1755                   | 488-2771             |
| CD8 percentage                            |                            |                      |
| Mean                                      | 54                         | 57                   |
| Range                                     | 27-72                      | 36-81                |
| Plasma HIV load‡                          |                            |                      |
| <10,000 HIV RNA copies/ml (no.)           | 11                         | 9                    |
| ≥10,000 HIV RNA copies/ml (no.)           | 20                         | 20                   |
| Mean (copies/ml)                          | 39,000                     | 41,000               |
| Range (copies/ml)                         | 9000-191,000               | 9000-414,000         |
| p24 Antigen level§                        |                            |                      |
| <15 pg/ml (no.)                           | 6                          | 6                    |
| ≥15 pg/ml (no.)                           | 25                         | 23                   |
| Mean (pg/ml)                              | 47                         | 60                   |
| Range (pg/ml)                             | 14-326                     | 14-548               |

\*For weights and laboratory measures, the mean of three base-line values for each patient was used in calculating the group means.

†The HIV risk groups were self-reported and are not mutually exclusive.

‡For plasma HIV load, the lower limit of sensitivity was 10,000 HIV RNA copies per milliliter; values below that were assigned a value of 9000 HIV RNA copies per milliliter.

§For p24 antigen level, the lower limit of sensitivity was 15 pg per milliliter; values below that were assigned a value of 14 pg per milliliter.

**TABLE 2. MODERATE AND SEVERE CLINICAL SIDE EFFECTS AND LABORATORY ABNORMALITIES IN 30 PATIENTS DURING 157 CYCLES OF INTERLEUKIN-2 TREATMENT.**

| SIDE EFFECT                             | NO. OF CYCLES (%) | NO. OF PATIENTS (%) |
|---|-------------------|---------------------|
| <b>Clinical effects*</b>                |                   |                     |
| Fatigue or malaise                      | 69 (44)           | 27 (90)             |
| Headache                                | 11 (7)            | 6 (20)              |
| Diarrhea                                | 7 (4)             | 6 (20)              |
| Stomatitis                              | 7 (4)             | 6 (20)              |
| Abdominal pain                          | 6 (4)             | 5 (17)              |
| Fever                                   | 5 (3)             | 4 (13)              |
| Sinus congestion                        | 5 (3)             | 3 (10)              |
| Myalgia                                 | 4 (3)             | 4 (13)              |
| Hypotension                             | 4 (3)             | 3 (10)              |
| <b>Laboratory abnormalities†</b>        |                   |                     |
| Bilirubin >2.5 mg/dl                    | 13 (8)            | 10 (33)             |
| Alkaline phosphatase >580 U per liter   | 5 (3)             | 3 (10)              |
| Phosphorus <1.5 mg/dl                   | 5 (3)             | 5 (17)              |
| Calcium <1.75 mmol/liter                | 4 (3)             | 3 (10)              |
| Granulocytes <750 cells/mm <sup>3</sup> | 4 (3)             | 3 (10)              |
| Hemoglobin <8.5 g/dl                    | 2 (1)             | 2 (7)               |
| Platelets <50,000 cells/mm <sup>3</sup> | 1 (1)             | 1 (3)               |
| Creatinine >2.5 mg/dl                   | 1 (1)             | 1 (3)               |
| Alanine aminotransferase >300 U/liter   | 1 (1)             | 1 (3)               |
| Creatine kinase >1930 U/liter           | 1 (1)             | 1 (3)               |

\*The following occurred in less than 2 percent of the cycles: rash, central nervous system abnormalities, nausea or vomiting, phlebitis, arthralgia, fluid retention, and dyspnea.

†To convert bilirubin values to micromoles per liter, multiply by 17.1; to convert creatinine values to micromoles per liter, multiply by 88.4; to convert phosphorus values to millimoles per liter, multiply by 0.3229.

the two groups were similar when assessed by the chi-square or Wilcoxon rank-sum test (Table 1). One patient assigned to interleukin-2 withdrew from the study before receiving interleukin-2 and was lost to follow-up. One patient in the control group received interleukin-2 from his primary care physician during the initial 14 months of the study.

There were no significant differences between the groups in antiviral regimens either at enrollment or during the study. Two thirds of the patients received combination antiretroviral therapy. Sixty percent of the patients in the interleukin-2 group had at least one change in their antiviral regimen during the study, as compared with 69 percent of the control group.

The 30 patients in the interleukin-2 group received 157 cycles of interleukin-2. Ten patients missed a total of 23 cycles for the following reasons: withdrawal from the study because of intolerable side effects (five patients, 16 cycles); noncompliance (one patient, 2 cycles); a CD4 count above 3000 cells per cubic millimeter (two patients, 3 cycles); anemia (one patient, 1 cycle); and cardiomyopathy (one patient, 1 cycle). The mean total dose per cycle decreased from 76 million IU for cycle 1 to 39 million IU for

cycle 6. At the same time, the percentage of patients completing a cycle without modification of the dose or premature discontinuation of the drug increased from 47 percent for cycle 1 to 80 percent for cycle 6.

Moderate and severe clinical side effects and laboratory abnormalities that occurred during the administration of interleukin-2 are summarized in Table 2. Fatigue, malaise, and other constitutional symptoms were the most common clinical toxic effects (recorded for 44 percent of cycles). Asymptomatic elevations of the bilirubin level were the most common laboratory abnormalities (8.3 percent of cycles). All patients had at least one moderate or severe toxic effect, primarily constitutional symptoms (which occurred in 90 percent of patients). Toxic effects decreased in frequency as the dosage was decreased during later cycles.

No differences in routine indicators of the safety of therapy were identified by linear regression, or slope, analysis, except for the total white-cell count, polymorphonuclear-cell count, and lymphocyte count, which showed a significant increase during the controlled study in the interleukin-2 group as compared with the control group (Table 3). When we examined the change in weight over time with linear regression, we found no significant difference between the two groups; the interleukin-2 group had a mean ( $\pm$ SE) loss of  $0.01 \pm 0.06$  kg per month; the control group, a loss of  $0.17 \pm 0.06$  kg per month ( $P = 0.06$ ).

In the interleukin-2 group hypothyroidism requiring thyroid supplementation developed in two patients; psoriatic arthritis developed in one patient with a history of psoriasis, but he completed the study at a reduced dose; one asymptomatic patient was noted to have a cardiomyopathy of undetermined cause that persisted for over one year despite his receiving no further interleukin-2; and one patient was given a diagnosis of Bowen's disease (squamous-cell carcinoma in situ).

Among the 29 patients in the control group, 10 had 32 episodes of moderate or severe clinical side effects or laboratory abnormalities during the study period, including headache (18 episodes), fatigue (4 episodes), weight loss (1 episode), diarrhea (4 episodes), nausea or vomiting (1 episode), fever (1 episode), elevated lipase concentration ( $>562$  U per liter, 2 episodes), and elevation in the alanine aminotransferase concentration ( $>300$  U per liter, 1 episode).

During the 14 months of the study, one patient in the interleukin-2 group died. This patient entered the study with a declining CD4 count (mean baseline CD4 count, 188 cells per cubic millimeter) and received two shortened cycles of interleukin-2; he withdrew from the study because of intolerable side effects with a CD4 count of 102 cells per cubic millimeter. He had a rapidly declining CD4 count and a rising viral load; *Mycobacterium avium* bacteremia subsequently developed (CD4 count, 51 cells per

**TABLE 3.** CHANGES IN IMMUNOLOGIC AND VIROLOGIC MEASURES ACCORDING TO STUDY GROUP.\*

| VARIABLE                                | INTERLEUKIN-2 GROUP (N=30) | CONTROL GROUP (N=29) | P VALUE† |
|---|----------------------------|----------------------|----------|
|   | mean ±SE                   |                      |          |
| <b>Hematologic measures</b>             |                            |                      |          |
| White-cell count (cells/mo)             | 67.8±21.8                  | -37.4±14.7           | <0.001   |
| Polymorphonuclear-cell count (cells/mo) | 21.8±11.7                  | -22.7±10.6           | 0.006    |
| Lymphocyte count (cells/mo)             | 30.0±13.7                  | -16.9±6.4            | 0.004    |
| <b>Immunologic measures</b>             |                            |                      |          |
| CD4 count (cells/mo)                    | 36.7±9.4                   | -4.8±1.6             | <0.001   |
| CD4 percentage (/mo)                    | 0.86±0.20                  | -0.26±0.08           | <0.001   |
| CD8 count (cells/mo)                    | -5.3±6.4                   | -8.8±4.3             | 0.65     |
| CD8 percentage (/mo)                    | -0.64±0.13                 | 0.14±0.08            | <0.001   |
| CD4 cells expressing CD25               |                            |                      |          |
| No. (cells/mo)                          | 29.9±6.1                   | -0.2±0.7             | <0.001   |
| Percentage (/mo)                        | 1.04±0.18                  | -0.01±0.02           | <0.001   |
| CD8 cells expressing HLA-DR             |                            |                      |          |
| No. (cells/mo)                          | -9.1±2.9                   | -4.5±2.5             | 0.23     |
| Percentage (/mo)                        | -0.48±0.10                 | 0.12±0.10            | <0.001   |
| <b>Virologic measures</b>               |                            |                      |          |
| p24 (log pg/mo)                         | 0.004±0.005                | 0.004±0.005          | 0.99     |
| HIV RNA (log HIV RNA copies/mo)         | 0.011±0.005                | 0.019±0.006          | 0.35     |

\*Data are expressed as the mean (±SE) slope of the indicated measure over time.

†P values were not adjusted for multiple comparisons.

cubic millimeter [9 percent]), and he was given a presumptive diagnosis of progressive multifocal leukoencephalopathy before his death, six months after his entry into the study. No other patient in the interleukin-2 group had an opportunistic infection during the 14-month controlled study.

Disseminated bacillary angiomatosis developed in one control patient at month 8 (CD4 count, 216 cells per cubic millimeter [10 percent]) but responded to therapy. A second patient in the control group had focal central nervous system lesions at month 14 (CD4 count, 74 cells per cubic millimeter [9 percent]) that responded to empirical anti-toxoplasma therapy.

**Changes in Immunologic Measures**

There was a significant difference in the mean slopes of the CD4 count over time between the two groups. In the interleukin-2 group, the mean slope was +36.7 cells per month, whereas in the control group it was -4.8 cells per month (P<0.001). Similarly, the percentage of CD4 cells also increased significantly in the interleukin-2 group (Table 3). Both groups had a slight drop in the CD8 cell count (P=0.65); however, the percentage of CD8 cells increased slightly in the control group while decreasing in the interleukin-2 group (Table 3) (P<0.001).

Consistent with the slope analysis was the fact that both the CD4 cell count and the percentage of CD4

cells increased in the interleukin-2 group during the period when interleukin-2 was administered (Fig. 1A and 1B). Fifty-seven percent of the patients treated with interleukin-2 had an increase of more than 50 percent over the base-line CD4 count at the end of approximately one year (mean of the measurements in months 11 and 12), as compared with none of the control group. The mean (±SE) net change from base line in the CD4 count at the end of the study (mean of the measurements at months 13 and 14) was an increase of 412±96 cells per cubic millimeter for the 29 surviving patients in the interleukin-2 group and a decrease of 48±23 cells per cubic millimeter for the 29 patients in the control group (P<0.001).

Among the other immunologic measures that were monitored, the interleukin-2 group had a significant decline in the percentage of CD8 cells that were positive for HLA-DR and a significant increase in the number and percentage of CD4 cells that were positive for CD25 (Table 3).

**Changes in Virologic Measures**

No significant differences were seen between the groups in the mean slope over time for either HIV RNA levels (log-transformed) or p24 antigen levels (also log-transformed) (Fig. 1C and Table 3). Similarly, no significant differences were seen between the groups in the net change in these values from base line to the end of the study (the mean of the values measured at months 13 and 14).

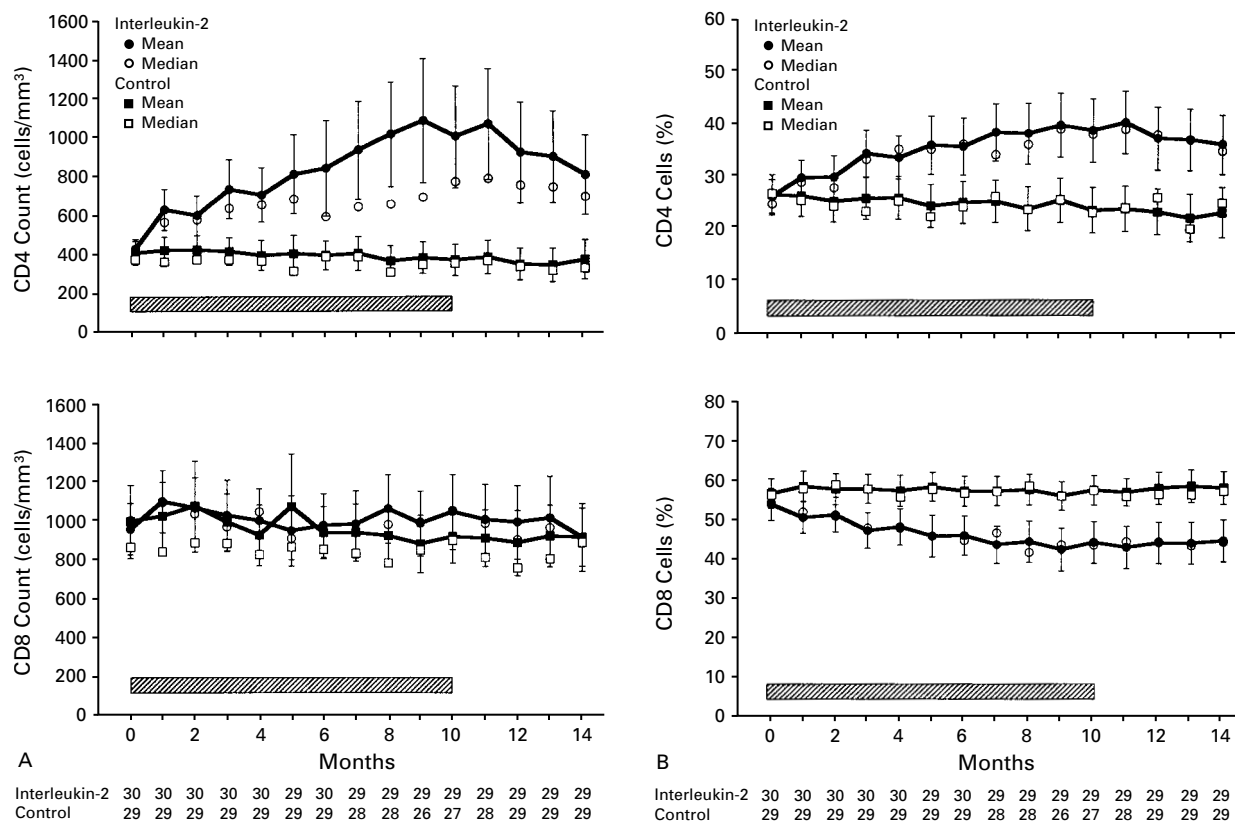
**Correlates of the Response to Interleukin-2**

There was a significant correlation between the CD4 response to interleukin-2 therapy and the base-line CD4 count (P=0.02), but not the base-line viral load. In the control group, there was a significant inverse correlation between the base-line viral load and the slope of the CD4 counts over time (P=0.001).

**Long-Term Follow-up**

After month 14, all patients were eligible to receive intermittent interleukin-2 on an ongoing basis. Twenty-one patients in the interleukin-2 group (19 of whom have continued in the study) and 24 in the control group (12 of whom are still in the study) elected to receive interleukin-2 during this extended study period. Eleven patients, three in the interleukin-2 group and eight in the control group, subsequently enrolled in a study examining the combination of the protease inhibitor indinavir and intermittent interleukin-2.

Four patients died of AIDS-related complications during long-term follow-up. Cryptosporidiosis developed at month 16 in one patient in the interleukin-2 group who had not had an increase in the CD4 count (CD4 count, 202 cells per cubic millimeter [9 percent]); he died six months later, approximately



**Figure 1.** Mean and Median CD4 and CD8 Counts (Panel A) and Percentages (Panel B) and Mean Plasma Viral Load and p24 Antigen Level (Panel C, facing page) in the Interleukin-2 and Control Groups during the 14 Months of the Controlled Study.

The error bars represent  $\pm 2$  SE and approximate the 95 percent confidence intervals. Values at month 0 (base line) are the means of three values measured before the beginning of the study. The shaded bars represent the times during which interleukin-2 was administered (month 0 to month 10). One patient received his sixth cycle of interleukin-2 at month 11. The numbers at the bottom of the panels indicate the numbers of patients for whom data were available.

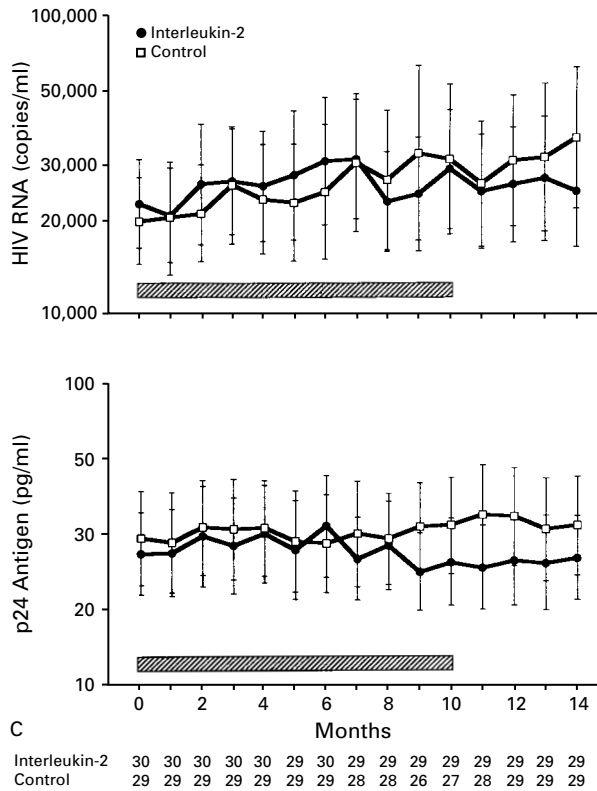
a year after his last cycle of interleukin-2. Biopsy-documented progressive multifocal leukoencephalopathy developed in one control patient, at month 18, during his third cycle of interleukin-2 (CD4 count, 144 cells per cubic millimeter [9 percent]); he died two months later. Another control patient died with a wasting syndrome 21 months after receiving two cycles of interleukin-2 (last CD4 count, 18 cells per cubic millimeter [2 percent]). A third control patient who never received interleukin-2 had *Pneumocystis carinii* pneumonia 31 months after study enrollment (CD4 count, 11 cells per cubic millimeter [2 percent]) and subsequently died. In addition, Hodgkin's disease (not an AIDS-defining disease) developed at month 16 in the patient in the interleukin-2 group who had had Bowen's disease (CD4 count, 792 cells per cubic millimeter [18 percent]); *P. carinii* pneumonia developed in a control patient 18 months after he received his second cycle of interleukin-2 with indinavir (CD4 count, 12 cells per cubic millimeter [3 percent]); and lymphoma developed at

month 20 in the control patient with presumptive toxoplasmosis.

In the interleukin-2 group, the mean CD4 count was maintained at approximately double the baseline value during the extended phase of the study (Fig. 2). Although the mean values were not as high as those seen during the controlled phase of the study, this difference reflected, at least in part, the individualization of dosing regimens during the extended phase, which focused on maintaining CD4 counts above base-line values while decreasing the frequency of administration of interleukin-2. For the control patients, many of whom chose to receive interleukin-2 in the extended phase of the study, there was an increase in CD4 counts after the initiation of interleukin-2 therapy.

### DISCUSSION

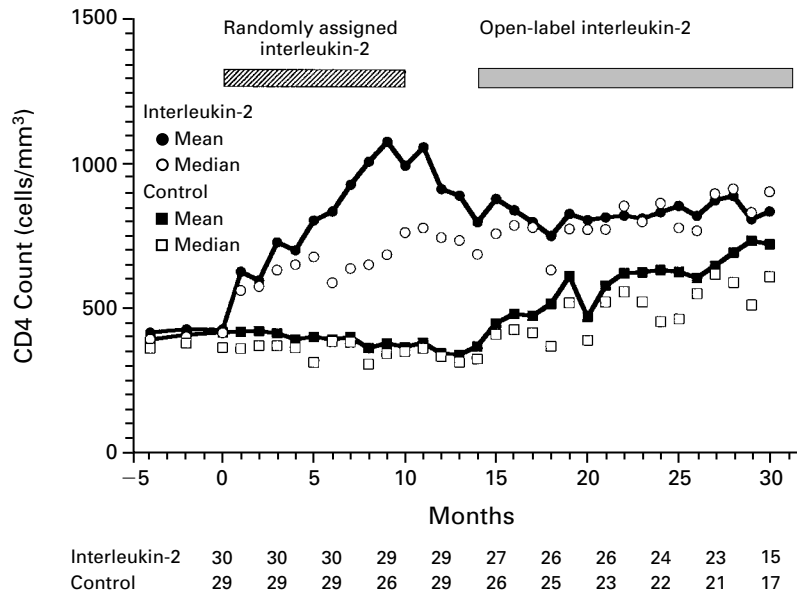
In this study we have shown that intermittent interleukin-2 therapy can lead to substantial and sustained increases in the number and percentage of



CD4 cells in HIV-infected patients with base-line CD4 counts of more than 200 cells per cubic millimeter. One year after the beginning of interleukin-2 therapy, the mean CD4 count in the interleukin-2 group was approximately double the base-line value. This increase has been sustained for more than two years by the continued administration of interleukin-2. In five patients, CD4 counts remained above 1000 cells per cubic millimeter for at least 18 months after interleukin-2 was discontinued. To date, no combination of antiretroviral agents has been shown to be capable of inducing increases in CD4 counts of this magnitude or duration.

The effects of interleukin-2 in promoting T-cell proliferation lead to a peripheral expansion of mature CD4 T cells, which may be maintained by the increase in the expression of CD25 (interleukin-2-receptor  $\alpha$  chain) on these cells. The increase in the number of CD4 cells is unlikely to represent simply the redistribution of cells to the circulation, since there is increased lymphocyte proliferation as well as lymph-node enlargement during interleukin-2 infusions. Moreover, bone marrow biopsies show an increase in lymphoid aggregates, and the increase in some patients in our study was sustained for more than a year without continued interleukin-2 therapy.

The effect of intermittent therapy with interleukin-2 was not limited to CD4 cells. Whereas CD8



**Figure 2.** Mean and Median CD4 Counts in the Interleukin-2 and Control Groups during the Controlled and Extended Phases of the Study.

The values shown are based on data for all patients who were available for follow-up, regardless of their status in the study. The numbers at the bottom of the figure indicate the numbers of patients for whom data were available.

counts remained stable, there was a decrease in the percentage of CD8 cells expressing HLA-DR, a marker of activation ( $P < 0.001$ ). This decrease was only partly related to the decline in the total percentage of CD8 cells<sup>17</sup>; it was paralleled by a decrease in the expression of CD38, another marker of activation (data not shown), and may reflect the ability of interleukin-2 to enhance CD8 effector function.

A crucial observation in this study is that the administration of interleukin-2 did not lead to long-term increases in the plasma viral load. Thus, intermittent interleukin-2 therapy can have a substantial impact on the chief immunologic abnormality associated with HIV infection, the loss of CD4 T cells, without leading to an overall increase in the level of HIV. In the control group, the viral load at study entry was predictive of the net change in the CD4 count, a finding that highlights the importance of the plasma viral load as a predictor of future immune status.<sup>12,13</sup>

The toxicity of interleukin-2 therapy in this study was substantially lower than that reported in our earlier study, in which a dose of 18 million IU per day was given for a longer portion of the study.<sup>17</sup> At the mean dose of approximately 8 million IU per day in the latter part of the present study, few dose reductions were necessary, and substantial immunologic effects were nonetheless seen.

The availability of potent HIV-protease inhibitors has raised the hope that sustained suppression of viral replication can be achieved with combination antiretroviral therapy.<sup>4,7,23</sup> Maximal suppression of HIV by such regimens may lead to improved CD4 responses to interleukin-2 therapy. Preliminary experience with intermittent interleukin-2 plus indinavir, a recently approved protease inhibitor, suggests that responses to interleukin-2 are enhanced when there is profound suppression of viral replication.<sup>24</sup> Complementing antiretroviral therapy with interleukin-2 is an attractive approach. Since interleukin-2 targets the immune system rather than the virus, alterations in the viral genome should not lead to resistance to the effects of interleukin-2 on CD4 cells. Indeed, some patients have continued to respond to interleukin-2 therapy for more than 50 months (unpublished data).

Although interleukin-2 can have a profound and sustained effect on CD4 counts in HIV-infected patients with base-line CD4 counts above 200 cells per cubic millimeter, the long-term clinical benefits of this increase remain to be established.

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