The Virologic, Immunologic, and Clinical Effects of Interleukin 2 With Potent Antiretroviral Therapy in Patients With Moderately Advanced Human Immunodeficiency Virus Infection

A Randomized Controlled Clinical Trial—AIDS Clinical Trials Group 328

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Background: Interleukin 2 (IL-2) administration increases CD4 counts in persons with higher counts. This study investigated persons with moderately advanced human immunodeficiency virus infection receiving highly active antiretroviral therapy (HAART).

Methods: Two hundred four patients with CD4 T-cell counts from 50/µL to 350/µL who were treatment naive or had been treated only with reverse transcriptase inhibitors began a specified protease inhibitor HAART regimen. Virologic responders (≤5000 copies/mL) at 12 weeks were randomized to open-label continuous-infusion IL-2 (IV IL-2), subcutaneous IL-2 (SC IL-2), or HAART alone. Thirty were not randomized and 15 enrolled in a substudy, leaving 159 for analysis. Subjects continued HAART alone for 72 weeks (n=52) or with IV IL-2 (n=53) or SC IL-2 (n=54) for 5 days every 8 weeks. The IV IL-2 subjects could switch to SC IL-2 if their CD4 T-cell count increased by 100/µL or by 25%.

Results: Patients receiving IV or SC IL-2 had greater increases in CD4 cell counts. At week 84, median increases were 459/µL, 312/µL, and 102/µL. Increases of greater than 50% at week 60 (primary end point) were achieved in 39 patients (81%) and 32 (67%) in the IV and SC IL-2 arms, respectively, compared with 13 (29%) in the HAART arm (P<.001 for both). Treatment with IL-2 did not increase plasma human immunodeficiency virus RNA levels. There were fewer new AIDS-defining events in the IV (P=.006) and SC (P=.03) IL-2 groups than in the HAART group (0, 1, and 7, respectively). Drug-related adverse events were more frequent with IL-2 treatment.

Conclusion: Addition of IL-2 to HAART can significantly expand CD4 T-cell counts in moderately advanced human immunodeficiency virus infection, without loss of virologic control.

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Human immunodeficiency virus (HIV) infection is characterized by impaired lymphocyte function and decreasing CD4 T-lymphocyte counts, leading to immunodeficiency and reduced survival. Potent antiretroviral drugs can produce long-term suppression of plasma HIV RNA with increases in CD4 T-cell counts and delayed disease progression.1,2 In more advanced infection, the durability of virologic suppression may be limited, CD4 cell counts frequently may not be restored to reference levels, and immunologic function remains impaired.3-6 Approaches to preserve immune function and immune reconstitution would have value.

Interleukin 2 (IL-2) is a cytokine with potent immunomodulating effects. Interleukin 2 induces proliferation and differentiation of CD4 and CD8+ lymphocytes, protects T lymphocytes from cell death, and enhances the cytotoxic activity.7,9 Interleukin 2 can induce substantial expansion of CD4 cells, associated with a dramatic prolongation of their half-life.10,21 Maintenance therapy in which IL-2 is administered when CD4 cell counts fall to predetermined levels is now commonly used.12 Patients with relatively high baseline CD4 cell counts have shown the best responses, with less favorable responses reported in more immunosuppressed patients receiving less potent antiretroviral therapy.14,21,22 A randomized study in subjects receiving HAART found...
only modest enhancement of CD4 cell counts in advanced (<200/µL) infection with subcutaneous (SC) IL-2 treatment compared with HAART alone (median gains of 65/µL vs 18/µL after 24 weeks [P<.001]).

This AIDS Clinical Trials Group (ACTG) study evaluated the immunologic, virologic, and clinical effect of intravenous (IV) or SC IL-2 administration with a specified HAART regimen in moderately advanced HIV infection.

**STUDY DESIGN**

Adults infected with HIV type 1 (HIV-1) without active AIDS-defining illnesses and with CD4 cell counts of 50/µL to 350/µL were eligible. Prior protease inhibitor therapy or IL-2 therapy, autoimmune disease, or cardiac disease were exclusionary. Subjects could not have prespecified laboratory abnormalities. The study was approved by institutional review boards, and the patients provided informed consent. Patients were enrolled at 26 ACTG sites. During step 1, subjects received 12 weeks of indinavir sulfate and specific combinations of 2 nucleosides (minimum of 1 new), including zidovudine, lamivudine, didanosine, or stavudine. Substitution for intolerance or for viral breakthrough was permitted. Patients with no more than 3000 plasma HIV RNA copies/mL at week 12, suggesting responses to HAART, were randomized 1:1:1.1 to HAART alone for 72 weeks, HAART plus continuous-infusion IL-2 (HAART + IV IL-2) at 9 mIU daily for 5 days every 8 weeks, or HAART + SC IL-2 at 7.5 mIU twice daily for 5 days every 8 weeks. Subjects could receive up to 9 cycles of IL-2 (Figure 1). Recombinant human IL-2 was used (Proleukin; Chiron, Emeryville, Calif). There were up to 2 dose reductions for clinical adverse effects or laboratory toxic effects (6 or 3 mIU daily for IV IL-2 and 4.5 or 2.5 mIU twice daily for SC IL-2). Patients in the IV IL-2 arm could switch to the SC IL-2 arm after 3 or 6 cycles if they achieved greater than a 25% increase and a 100/µL increase in CD4 cell count above the randomization baseline. Postrandomization AIDS-related events were assessed using protocol-specified criteria. All patients underwent chest radiography and physical examination and were seen every 4 weeks until week 20 and every 8 weeks thereafter.

A substudy (ACTG 5046) examined immunization responses in 38 patients. Another substudy (ACTG 928) in 15 patients added a tumor necrosis factor inhibitor, etanercept, to minimize the toxic effects of IL-2 and, while not affecting toxicity, it enhanced IL-6 and C-reactive protein expression.

**LABORATORY METHODS**

Safety laboratory procedures and CD4/CD8+ T-cell counts were performed every 8 weeks at ACTG-certified laboratories. Batched HIV RNA levels were measured at entry, randomization, and weeks 60 and 84 by means of the branched DNA assay (Chiron) in an ACTG-certified laboratory. The RNA levels below 50 copies/mL were analyzed as if they were 50 copies/mL. Skin testing for delayed-type hypersensitivity was assessed at baseline and weeks 36, 60, and 84. Eighty-seven patients participated in the immunology substudy, wherein enumeration of lymphocyte subpopulations, lymphocyte proliferation, and cytokine and soluble activation marker levels were performed at entry, randomization, and weeks 36, 60, and 84.

Lymphocyte subpopulations were measured using ACTG consensus methods that used fluorochrome-labeled monoclonal antibodies (Becton-Dickinson Pharmingen, San Diego, Calif) to the following antigens: CD3, CD4, CD8, CD16, CD19, CD25, CD28, CD38, CD45RA, CD56, CD62L, CD95, and HLA-DR. Naïve cells were defined as CD45RA−CD62L−, memory/effector cells as CD45RA−CD62L+; memory/effector cells as CD45RA−CD62L++; CD4+CD25+, and activated cells as HLA-DR+. Functional subsets were defined according to the expression of CD95+ or CD28+. Lymphocyte proliferation assays used ACTG consensus methods, and cytokine and soluble activation marker levels were measured in cryopreserved specimens.

**STATISTICAL METHODS**

Randomization was stratified by participation in the ACTG 928 and by the nucleoside analogue combinations. Each IL-2 treatment arm was to be compared (separately) with the HAART alone arm using 2-sided tests with significance levels of .025. (No other correction for multiple comparisons was performed.) The primary efficacy end point was the proportion of patients with CD4 T cells (a week 60 count of ≥50% larger than the randomization count). The accrual goal was 120 randomized patients (40 per arm) with week 12 and week 60 CD4 cell counts who were not enrolled in the ACTG 928. This sample size provided at least 80% power to detect a difference of 35% in the true proportions of patients with a CD4 T-cell response, using the Fisher exact test. The week 12 CD4 and CD8+ T-cell counts were those closest to randomization. The week 60 counts were those between weeks 32 and 68 that were closest to week 60.

The protocol specified that the secondary CD4 T-cell analysis should apply random effects models to the slope of log CD4 cell counts between weeks 12 and 60. For other secondary analy-
ses, proportions of patients with an end point were compared using Fisher exact tests. Analyses of variables with many possible values used the Wilcoxon rank sum test.

Step-up logistic regression models using the likelihood ratio test were performed to model the probability of CD4 cell response by week 60 in all randomized patients; treatment variables were always in the model, and candidate exploratory variables included the factors in Table 1 and white blood cell count, lymphocyte count, lymphocyte percentage, and real-time plasma HIV RNA levels (at entry and randomization). Continuous covariates were divided into quartiles for these models. Sets of step-up logistic regression models were performed in the immunology substudy, trying all the variables of entry and randomization percentages and counts for the T-cell subsets measured. All of these measurements were dichotomized at the median observed value.

RESULTS

PATIENTS

Two hundred four patients were enrolled and 174 were randomized to step 2: 15 for the ACTG 928 and 159 for the ACTG 328 (Figure 1). Data for the ACTG 928 patients appear only in the tables regarding toxic effects and deaths (Table 2 and Table 3). Reasons why patients were not randomized to step 2 included plasma HIV RNA levels of greater than 5000 copies/mL (n=10), inability to comply (n=9), or 1 of multiple other reasons (n=11). The baseline characteristics are presented in Table 1. The groups were similar, although the HAART alone group was likely to be older than 49 years and to have higher CD4 and CD8+ T-cell counts.

DURATION OF AND MODIFICATIONS IN THERAPY AND IL-2 DOSING

Sixteen patients changed their HAART drugs. Forty patients discontinued study drug therapy before week 60 (10 receiving HAART alone, 13 receiving HAART + SC IL-2, and 17 receiving HAART + IV IL-2); the reasons for most of these changes were mild or moderate toxic effects, complexity of medications, clinical events, and noncompliance. Fifty-one patients discontinued study drug therapy before week 84. Four patients died before week 60.27 The median number of cycles in the HAART + IV IL-2 arm was 9; in the HAART + SC IL-2 arm, 8. Only 14 patients (26%) in the IV IL-2 arm received 4 or more cycles and only 4 (8%) received 7 or more cycles. By week 84, most IV IL-2 patients had been receiving SC IL-2 for 1 year (Figure 2).

The median dose per cycle of IV IL-2 was 45 mIU (protocol-specified initial dose); the median dose per cycle of SC IL-2 was 75 mIU (protocol-specified initial dose) for only the first 2 cycles (third cycle, median dose, 45 mIU; from the fourth cycle, median dose, 45 mIU). The median dose per cycle of SC IL-2 received after switching was similar to that in the SC IL-2 group. The median total dose during the first 9 cycles was 427 mIU in the IV IL-2 patients and 413 mIU in the SC IL-2 patients.

Table 1. Characteristics of Patients at Time of Randomization*

<table>
<thead>
<tr>
<th>Baseline Characteristics</th>
<th>Step 1 (n = 189)</th>
<th>HAART Alone (n = 52)</th>
<th>HAART + SC IL-2 (n = 54)</th>
<th>HAART + IV IL-2 (n = 53)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, mean (range), y</td>
<td>38 (18-70)</td>
<td>41 (19-70)</td>
<td>38 (25-60)</td>
<td>38 (18-58)</td>
</tr>
<tr>
<td>Age group, y</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;30</td>
<td>23 (12)</td>
<td>7 (13)</td>
<td>5 (9)</td>
<td>6 (11)</td>
</tr>
<tr>
<td>30-39</td>
<td>96 (51)</td>
<td>19 (37)</td>
<td>35 (65)</td>
<td>27 (51)</td>
</tr>
<tr>
<td>40-49</td>
<td>46 (24)</td>
<td>12 (23)</td>
<td>10 (19)</td>
<td>15 (28)</td>
</tr>
<tr>
<td>≥50</td>
<td>24 (13)</td>
<td>14 (27)</td>
<td>4 (7)</td>
<td>5 (9)</td>
</tr>
<tr>
<td>No. male/female</td>
<td>172/17</td>
<td>47/5</td>
<td>47/7</td>
<td>50/3</td>
</tr>
<tr>
<td>Race/ethnicity</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>105 (56)</td>
<td>31 (60)</td>
<td>29 (54)</td>
<td>30 (57)</td>
</tr>
<tr>
<td>Black</td>
<td>51 (27)</td>
<td>11 (21)</td>
<td>12 (22)</td>
<td>18 (34)</td>
</tr>
<tr>
<td>Hispanic/Latino</td>
<td>29 (15)</td>
<td>9 (17)</td>
<td>10 (19)</td>
<td>5 (9)</td>
</tr>
<tr>
<td>Other</td>
<td>4 (3)</td>
<td>1 (2)</td>
<td>3 (6)</td>
<td>0</td>
</tr>
<tr>
<td>Previous IV drug exposure</td>
<td>22 (12)</td>
<td>6 (12)</td>
<td>5 (9)</td>
<td>8 (15)</td>
</tr>
<tr>
<td>Median CD4 cell count/µL</td>
<td>200</td>
<td>305</td>
<td>248</td>
<td>239</td>
</tr>
<tr>
<td>Median % of CD4 cells</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Median CD8 cell count/µL</td>
<td>799</td>
<td>883</td>
<td>825</td>
<td>744</td>
</tr>
<tr>
<td>Median % of CD8 cells</td>
<td>59</td>
<td>55</td>
<td>55</td>
<td>54</td>
</tr>
<tr>
<td>Median log10 branched DNA, cells/µL</td>
<td>4.44</td>
<td>1.70</td>
<td>1.85</td>
<td>1.70</td>
</tr>
<tr>
<td>Nucleoside analogue</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Zidovudine + didanosine</td>
<td>5 (3)†</td>
<td>1 (2)</td>
<td>2 (4)</td>
<td>2 (4)</td>
</tr>
<tr>
<td>Zidovudine + lamivudine</td>
<td>44 (28)†</td>
<td>14 (27)</td>
<td>15 (28)</td>
<td>15 (28)</td>
</tr>
<tr>
<td>Stavudine + didanosine</td>
<td>22 (14)†</td>
<td>8 (15)</td>
<td>7 (13)</td>
<td>7 (13)</td>
</tr>
<tr>
<td>Stavudine + lamivudine</td>
<td>88 (55)†</td>
<td>29 (56)</td>
<td>30 (56)</td>
<td>29 (55)</td>
</tr>
</tbody>
</table>

Abbreviations: HAART, highly active antiretroviral therapy; IL-2, interleukin 2; IV, intravenous; SC, subcutaneous.

*Unless otherwise indicated, data are expressed as number (percentage) of patients. Excludes AIDS Clinical Trials Group 928 patients.
†Indicates at the time of randomization.
Every attempt was made to obtain week 60 CD4 cell counts (primary end point) on all patients, even for subjects who had stopped protocol treatment. Patients who died by week 60 were counted as nonresponders. Of 159 step 2 patients, 141 (89%) were included in this analysis; 16 had stopped protocol therapy more than 8 weeks before their week 60. Shown in Figure 3, each IL-2 arm had significantly more week 60 responders than did the HAART arm: 13 (29%) of 45 in the HAART arm, 39 (81%) of 48 in the HAART/IV IL-2 arm, and 32 (67%) of 48 in the HAART/SC IL-2 arm (P < .001 for the IV and SC arms). The advantage of IL-2 treatment was irrespective of the nucleoside analogue combination. Similarly, both IL-2 arms were superior to HAART alone in the proportions of responders at week 36 (142 patients).

### Table 2. Clinical Events and Deaths*

<table>
<thead>
<tr>
<th>Event</th>
<th>HAART Alone</th>
<th>HAART + SC IL-2</th>
<th>HAART + IV IL-2</th>
</tr>
</thead>
<tbody>
<tr>
<td>AIDS-defining events, total No.</td>
<td>7</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Kaposi sarcoma</td>
<td>2 (0.2, 5) [356, 289]</td>
<td>1 (5) [582]</td>
<td>0</td>
</tr>
<tr>
<td>Burkitt lymphoma†</td>
<td>1 (12) [350]</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Liver and spleen lymphoma†</td>
<td>1 (13) [171]</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Pneumocystis jiroveci pneumonia</td>
<td>1 (1) [144]</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Esophageal candidiasis</td>
<td>1 (3) [108]</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Castleman disease†</td>
<td>1 (7) [100]</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Other HIV-related events, total No.</td>
<td>1</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>Oral hairy leukoplakia</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Oral thrush</td>
<td>0</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Oral candidia</td>
<td>0</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Any death</td>
<td>3‡</td>
<td>2§</td>
<td>2</td>
</tr>
</tbody>
</table>

*Data in parentheses indicate months to the event; in brackets, CD4 cell count per microliter.
†Later died.
‡All due to AIDS-defining events.
§Indicates 1 suicide and 1 automobile crash.
||Indicates 1 liver failure and 1 bowel infarction (in a patient from the AIDS Clinical Trials Group 928).

### Table 3. Lymphocyte Phenotype Panel*

<table>
<thead>
<tr>
<th>Phenotype</th>
<th>HAART Alone</th>
<th>HAART + IV IL-2</th>
<th>HAART + SC IL-2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>At Randomization</td>
<td>Change</td>
<td>At Randomization</td>
</tr>
<tr>
<td>CD4 naive</td>
<td>54</td>
<td>18</td>
<td>84</td>
</tr>
<tr>
<td>CD4/38/DR</td>
<td>22</td>
<td>−2</td>
<td>23</td>
</tr>
<tr>
<td>CD4/95</td>
<td>143</td>
<td>78</td>
<td>169</td>
</tr>
<tr>
<td>CD4/28</td>
<td>176</td>
<td>94</td>
<td>222</td>
</tr>
<tr>
<td>CD4/25</td>
<td>182</td>
<td>66</td>
<td>207</td>
</tr>
<tr>
<td>CD4 memory/effector</td>
<td>154</td>
<td>57</td>
<td>160</td>
</tr>
<tr>
<td>CD8 naive</td>
<td>176</td>
<td>4</td>
<td>199</td>
</tr>
<tr>
<td>CD8/38/DR</td>
<td>238</td>
<td>−50</td>
<td>229</td>
</tr>
<tr>
<td>CD8/95</td>
<td>447</td>
<td>36</td>
<td>387</td>
</tr>
<tr>
<td>CD8/28</td>
<td>238</td>
<td>25</td>
<td>257</td>
</tr>
<tr>
<td>CD8/25/45RO</td>
<td>91</td>
<td>14</td>
<td>104</td>
</tr>
<tr>
<td>CD8 memory</td>
<td>567</td>
<td>−73</td>
<td>518</td>
</tr>
</tbody>
</table>

*Data are given as median counts at randomization and as median changes in counts between weeks 12 and 60.
†P values are for each IL-2 arm vs HAART alone.

### Figure 2

Number of patients on each interleukin 2 (IL-2) treatment arm who received intravenous (IV) or subcutaneous (SC) IL-2 at each of the first 9 cycles of therapy. Patients switched to SC IL-2 treatment for protocol-prespecified increases in CD4 cell counts.
Figure 3. The proportion of patients who experienced CD4 responses by treatment interval and the patterns of CD4 and CD8+ cell counts from week 12 to 84 by treatment arm. A, Percentage of patients with a greater than 50% increase in CD4 cell counts from randomization to 36, 60, and 84 weeks. The *P* values are from a Fisher exact test on the proportion with responses. The week 60 change was the primary end point of the study. B, Mean CD4 and CD8+ cell counts by treatment arm. A, Percentage of patients with a greater than 50% increase in CD4 cell counts from randomization to 36, 60, and 84 weeks. The *P* values are from a Fisher exact test on the proportion with responses. The week 60 change was the primary end point of the study. B, Mean CD4 and CD8+ cell counts by treatment arm.

Based on the random-effects models (a protocol-designated secondary end point), the CD4 T-cell slopes were greater in the IV and SC IL-2 arms than in the HAART alone arm from week 12 to weeks 36, 60, and 84 (*P < .001 at each time point*). Figure 3B shows the average CD4 and CD8+ T-cell counts by arm for weeks 12 to 84. From weeks 12 to 60, there were significantly higher increases in CD4 cell counts in both IL-2 arms compared with the HAART arm (*P = .001 for both comparisons*). No significant differences were observed in CD8+ T-cells counts (data not shown).

**PLASMA HIV RNA**

No significant differences were observed in the percentages of patients with HIV RNA of less than 50 or less than 500 copies/mL (Figure 4). At randomization, there were fewer patients in the HAART + SC IL-2 arm with less than 50 copies/mL than in the HAART alone arm. No significant differences were seen in median HIV RNA levels or the percentage of patients with virologic failure (>0.7-log increase in HIV RNA levels) from randomization to week 60 or to week 84.

**CLINICAL EVENTS AND DEATHS**

Table 2 shows the timing and CD4 levels at the diagnosis of new AIDS-defining clinical events. More AIDS-related events were observed in the HAART alone arm than in either of the IL-2 arms (7 in the HAART alone arm vs 0 in the IV IL-2 + HAART arm [*P = .006*] and 1 in the SC IL-2 + HAART arm [*P = .031*]). The malignancies, which included Kaposi sarcoma, lymphomas, and Castleman disease, occurred in patients with the last CD4 cell counts before diagnosis ranging from 171/µL to 582/µL. There were no significant differences in HIV-related non-AIDS-defining events in the 3 arms (1, 3, and 1) or in the number of deaths from any cause (3, 2, and 2). Inclusion of events for the 15 ACTG 928 participants did not affect the results.

**IMMUNOLOGIC EFFECTS OF IL-2 ADMINISTRATION**

There were no significant differences in skin test reactivity or in lymphocyte proliferation, and the differences in soluble marker levels had large *P* values (.02-.05; data not shown).

### Table 2: Timing and CD4 Levels at Diagnosis of New AIDS-Defining Clinical Events

<table>
<thead>
<tr>
<th>Treatment Arm</th>
<th>VL &lt;50 Copies/mL</th>
<th>VL &lt;500 Copies/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td>HAART SC IL-2</td>
<td>62/385</td>
<td>81/389</td>
</tr>
<tr>
<td>IV IL-2</td>
<td>84/216</td>
<td>88/36</td>
</tr>
<tr>
<td>HAART SC IL-2</td>
<td>62/385</td>
<td>81/389</td>
</tr>
<tr>
<td>IV IL-2</td>
<td>84/216</td>
<td>88/36</td>
</tr>
<tr>
<td>HAART SC IL-2</td>
<td>62/385</td>
<td>81/389</td>
</tr>
<tr>
<td>IV IL-2</td>
<td>84/216</td>
<td>88/36</td>
</tr>
</tbody>
</table>

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Table 3 shows the median week 12 measurements and median changes from weeks 12 to 60 in the T-cell phenotypic panel for each of the 3 treatment arms. For each phenotype, 51 patients (split among the 3 arms) had results from weeks 12 and 60. The HAART + IV IL-2 arm had significantly (P<.05) greater increases than did the HAART alone arm in the numbers of CD4 memory/effector, CD4 naïve, CD4/CD95+, CD4/CD28−, and CD8+/CD28− cell counts. The HAART + SC IL-2 arm had significantly greater increases than did the HAART alone arm in CD4 memory/effector, CD4/CD95+, CD4/CD28−, CD4/CD25+, and CD8+ memory cell counts. These differences persisted through week 84 (data not shown). There were no significant changes in the number or the percentage of CD4 or CD8+ T cells expressing activation markers.

MODELS FOR CD4 T-CELL RESPONSE AT 60 WEEKS

In the final step-up logistic model of week 60 CD4 T-cell response for the overall study, both IL-2 arms were highly significantly different from the HAART alone arm (P<.001 for IV IL-2 + HAART and P = .003 for SC IL-2 + HAART). The only other variables significant for larger probability of week 60 CD4 T-cell response were entry (week 0) plasma HIV RNA level of less than 10 000 copies/mL (P = .01) and randomization CD8+ T-cell percentage of less than 47% (P = .05); both cutoff values represent the first quartile of these variables. No other variables, notably including a pretreatment IL-2 CD4 T-cell count of less than or greater than 200/µL, were significant in the final model.

ADVERSE EVENTS

Table 4 shows the percentage of patients in each arm with grade 3 or grade 4 laboratory or clinical toxic effects. Only categories of adverse events in which at least 5% of the 174 patients had grade 3 or worse adverse events are reported. Overall, both IL-2 arms were associated with significantly more grade 3 or 4 clinical toxic effects usually associated with IL-2 treatment. The HAART + IV IL-2 arm had more gastrointestinal tract toxic effects (especially nausea and vomiting), skin/mucocutaneous toxic effects, and general toxic effects (specifically fever and chills), and SC IL-2 treatment was associated with more fatigue.

Intermittent IV or SC IL-2 treatment combined with antiretroviral therapy has been shown to produce substantial and sustained increases in CD4 cell counts in patients with early-stage or midstage HIV infection, ie, CD4 cell counts of greater than 250/µL.13-21 There are fewer studies in patients with more advanced HIV infection. In this randomized controlled study, the immunologic and virologic effects of beginning HAART and then IL-2 by the intravenous or subcutaneous routes was compared with HAART alone in patients with CD4 cell counts between 50/µL and 350/µL before HAART. In patients with moderately advanced disease who responded virologically to HAART after 12 weeks, significant increases in CD4 cell counts were seen with IL-2 therapy and greater expansion of multiple phenotypic subsets of CD4 T cells was observed. At least a 50% increase in CD4 cells was achieved by week 60 in 81% of patients in the IV IL-2 + HAART arm, 67% of those in the SC IL-2 + HAART arm, and 29% of those in the HAART alone arm. This was achieved without an increase in plasma HIV RNA levels.

At the initiation of this study, it was unclear whether the CD4 cell response to IL-2 treatment would be af-

**Table 4. Patients With the Most Frequent Drug-Related Grade 3 or Worse Adverse Events*†**

<table>
<thead>
<tr>
<th>Adverse Event</th>
<th>HAART (n = 57)</th>
<th>HAART + SC IL-2 (n = 59)</th>
<th>P Value†</th>
<th>HAART + IV IL-2 (n = 58)</th>
<th>P Value†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Any clinical toxic effect</td>
<td>30</td>
<td>53</td>
<td>.02</td>
<td>67</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>Fatigue</td>
<td>7</td>
<td>24</td>
<td>.02</td>
<td>21</td>
<td>.07</td>
</tr>
<tr>
<td>Fever</td>
<td>2</td>
<td>9</td>
<td>.06</td>
<td>10</td>
<td>.03</td>
</tr>
<tr>
<td>Chills/sweats</td>
<td>0</td>
<td>9</td>
<td>.09</td>
<td>19</td>
<td>.02</td>
</tr>
<tr>
<td>Skin rash/mucocutaneous</td>
<td>2</td>
<td>9</td>
<td></td>
<td>14</td>
<td>.03</td>
</tr>
<tr>
<td>Nausea</td>
<td>4</td>
<td>14</td>
<td></td>
<td>19</td>
<td>.02</td>
</tr>
<tr>
<td>Vomiting</td>
<td>2</td>
<td>7</td>
<td></td>
<td>14</td>
<td>.03</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>2</td>
<td>3</td>
<td></td>
<td>10</td>
<td></td>
</tr>
<tr>
<td>Myalgia/discomfort</td>
<td>9</td>
<td>17</td>
<td></td>
<td>16</td>
<td></td>
</tr>
<tr>
<td>Any laboratory toxic effect</td>
<td>33</td>
<td>46</td>
<td></td>
<td>40</td>
<td></td>
</tr>
<tr>
<td>Neutropenia</td>
<td>5</td>
<td>3</td>
<td></td>
<td>9</td>
<td></td>
</tr>
<tr>
<td>Transaminase level elevation</td>
<td>9</td>
<td>9</td>
<td></td>
<td>7</td>
<td></td>
</tr>
<tr>
<td>Total bilirubin level elevation</td>
<td>9</td>
<td>22</td>
<td></td>
<td>12</td>
<td></td>
</tr>
<tr>
<td>Creatinine phosphokinese level elevation</td>
<td>4</td>
<td>3</td>
<td></td>
<td>9</td>
<td></td>
</tr>
<tr>
<td>γ-Glutamyl transferase elevation</td>
<td>4</td>
<td>10</td>
<td></td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>Glucose level elevation</td>
<td>7</td>
<td>5</td>
<td></td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>Triglyceride level elevation</td>
<td>4</td>
<td>9</td>
<td></td>
<td>3</td>
<td></td>
</tr>
</tbody>
</table>

Abbreviations: HAART, highly active antiretroviral therapy; IL-2, interleukin 2; IV, intravenous; SC, subcutaneous.
*Data are expressed as percentage of patients. Data include 15 patients entered on substudy with etanercept treatment.
†P values refer to comparisons of each IL-2 arm with the HAART arm. P = .10 except where indicated.
fected by the administration route, especially in patients with low baseline CD4 cell counts, because the study suggesting equivalent results was conducted in patients with CD4 cell counts greater than 250/µL. The SC route seemed more practical for long-term administration. We sought to address these issues by randomizing patients to receive SC IL-2 or IV IL-2 at the onset of the study but allowing subjects to switch to SC IL-2 later if they reached certain increases in CD4 cell counts; therefore we did not directly compare the routes of IL-2 administration. The slope of CD4 cells was larger in the IV IL-2 arm than in the SC IL-2 arm for 72 weeks, although most patients in the IV arm had been receiving SC IL-2 for a year. Differences in the rate and frequency of adverse and toxic effects were observed, with patients tolerating IL-2 treatment reasonably well. This occurred with similar IL-2 dosing per cycle. A study of quality of life in the 3 groups suggested a short-term detrimental effect during courses of IL-2 treatment and some evidence of a long-term beneficial effect in the SC IL-2 arm.28

The sustained 3- to 4-fold greater increases in CD4 cell counts with IL-2 could have clinical relevance in patients with advanced HIV infection or in those with insufficient immunologic responses to suppressive antiretroviral therapy.29 Participants had increases in CD4 cell counts regardless of whether the counts were above or below 200/µL at the time of randomization.

As noted in patients with earlier-stage disease, the increase in CD4 cell count as a result of IL-2 therapy appears to be associated with a general peripheral expansion of CD4 cell populations that includes naive and memory/effector cells as well as increases in the number of cells expressing CD28, CD25, and CD95.30

Despite increases in the number of CD4 cells and their subsets, we were unable to demonstrate in vitro evidence of increased functionality. In a substudy of this trial,25 responses to immunization were not enhanced among IL-2 recipients. However, a modest enhancement in vaccine responsiveness in IL-2 recipients has been reported.31

In this study, the number of CD4 cells expressing CD4/CD28 increased in both of the IL-2 arms. Expression of this coreceptor for T-cell activation is critical for T-cell responsiveness and it predicted responses to immunizations in HIV infection.30,31 Although exposure to IL-2 induces expression of the α receptor chain (CD25) that is needed for high-affinity IL-2 binding and promotes cellular expansion, high-level expression of CD25 also is seen among CD4 regulatory T cells that attenuate T-cell immunity.32 While IL-2 administration enhances the expansion of CD25+ cells, these cells do not appear to have the same immunosuppressive activities that are characteristic of CD4 regulatory T cells.31

The CD4 cell count is the most reliable predictor of short-term morbidity, with counts below 200/µL associated with a heightened risk of opportunistic infections and death.3,32 While IL-2 administration can increase the CD4 cell count in persons with HIV infection, there has been no convincing evidence that the CD4 T-cell count increases seen after IL-2 administration confer any clinical benefit. Although we were unable to demonstrate laboratory evidence of enhanced immune function, the apparent lower frequency of AIDS-defining events in the IL-2 arms and an earlier metaanalysis33 of several clinical studies suggesting clinical benefits are encouraging but require confirmation. Two large randomized studies powered to discern differences in the occurrence of the clinical end point, the Evaluation of Subcutaneous Proleukin in a Randomized International Trial (ESPRIT)33 and the Phase III Multicenter Randomized Study of the Biological and Clinical Efficacy of Subcutaneous Recombinant, Human Interleukin-2 in HIV-Infected Patients With Low CD4 Counts Under Active Antiretroviral Therapy (SILCAAT),34 are ongoing.

In conclusion, this study provides evidence that IL-2 combined with HAART has the potential to significantly increase CD4 cell counts and partially restore naive and memory CD4 cells and other phenotypic subpopulations of T cells in patients with moderately advanced HIV infection, without increasing HIV replication. The short-term occurrence of AIDS-defining diseases might have been reduced in patients receiving IL-2. Whether IL-2 produces long-term clinical benefit will be answered by large, ongoing clinical trials.

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