Durability of Responses to Interferon Alfa-2b in Advanced Hairy Cell Leukemia


Previous studies have demonstrated that significant hematologic improvement occurs in the majority of patients with hairy cell leukemia (HCL) treated with partially purified or recombinant interferon (IFN). Fifty-three patients received IFN alfa-2b for at least 3 months in a dose of 2 x 10^6 U/m^2 subcutaneously thrice weekly. Of the 49 patients evaluable for response (at least 6 months of IFN therapy), there were ten complete responses and 29 partial responses for a total response rate of 80%. The peripheral blood counts and bone marrow continued to improve over the course of a full year of therapy. IFN was well tolerated, with no patients discontinuing therapy because of toxicity. Transient myelosuppression occurred in most patients during the first 10 months of therapy, occasionally precipitating a transient decrease in neutrophil count. Although some patients developed grade 4 neutropenia, none required discontinuation of therapy because of toxicity.

Hairy cell leukemia (HCL) was first described by Bouroncle et al in 1958. Splenectomy has long been considered the initial treatment of choice, but patients with advanced disease postsplenectomy have historically been difficult to treat because of persistent neutropenia following even low-dose chemotherapy.

Quesada et al first demonstrated that a partially purified interferon product was highly effective in advanced HCL, and Ratain et al showed that interferon itself was the active molecule using the recombinant drug. These initial results with recombinant interferon have subsequently been confirmed in a multinational study of 64 patients, and the results of several large series have recently been reviewed.

The objectives of this analysis from a single institution are to extend and update our experience with interferon alfa-2b (IFN) in advanced HCL and to determine the duration of response to IFN by careful assessment of peripheral blood and bone marrow changes following discontinuation of therapy.

MATERIALS AND METHODS

Patient eligibility. Requirements for inclusion in the study were as follows: (1) pathologically confirmed diagnosis of HCL on the basis of a recent (within 1 month) bone core biopsy; (2) significant anemia (hemoglobin, <12 g/dL), thrombocytopenia (platelets, <100,000 cells/µL), neutropenia (neutrophils, <1,000 cells/µL), and/or leukemia (white blood count, >10,000 cells/µL with more than 50% hairy cells); (3) prior splenectomy, unless refused by the patient; (4) normal hepatic and renal function; (5) past the nadir of toxicity of any previous therapy (chemotherapy and/or radiotherapy), preferably no treatment in the previous 4 weeks; and (6) informed consent in accordance with procedures defined by the Department of Health and Human Service.

Study design. IFN (Intron A, Schering Corp, Kenilworth, NJ) was supplied as lyophilized powder (10 x 10^6 U/vial) and reconstituted with sterile water. The initial dose of IFN was 2 x 10^6 U/m^2 self-administered subcutaneously three times weekly. Dose escalation up to 10 x 10^6 U/m^2 was allowed for nonresponders but used in only two patients. Five of the first six patients were initially treated with 10 x 10^6 U/m^2, but it was deescalated when the lower dose was shown to be active. Patients were admitted to the hospital for the first two doses to facilitate teaching of IFN reconstitution and administration and to observe for unexpected toxicity.

Patients were seen monthly at the University of Chicago. Treatment was continued for 12 months in responding patients. Patients completing 12 months of IFN therapy after January 1985 were entered in a randomized study comparing an additional 6 months of IFN therapy to observation. Patients were stratified by response (at the time of randomization), as defined in the next section.

While receiving IFN, a complete blood count (and differential) was obtained weekly until patients showed objective evidence of response, then it was done monthly. A chemistry panel was obtained monthly. Bone marrow biopsy specimens were obtained at least every 3 months. Since the neutrophil alkaline phosphatase score (NAP) has previously been shown to be elevated in HCL, we also determined the NAP every 3 months. After discontinuation of IFN therapy, a complete blood count was obtained monthly. A chemistry panel, NAP, and bone marrow biopsy specimen were obtained every 3 months for the first year of follow-up and then every 6 months.

Response criteria. Patients were considered eligible for response after 6 months of therapy. Although responses occur within 1 to 2 months of starting IFN treatment, prior studies have demonstrated that the absolute neutrophil count (ANC) and hemoglobin level may improve continuously over the first 6 months of therapy. The response criteria used in this analysis are similar to those reported previously.

A complete response (CR) was defined as (a) normalization of peripheral blood counts (hemoglobin, >12 g/dL; platelets, >100,000 cells/µL; and ANC, >1,500 cells/µL) and (2) less than 5% hairy cells in the bone marrow biopsy specimen. (Patients with
focal clumping of hairy cells in the bone marrow were not considered to have a CR.

A partial response (PR) designated normalization of peripheral blood counts but not meeting the pathological criteria for a CR.

A minor response (MR) was defined as normalization of at least one peripheral blood count initially abnormal but not meeting the criteria for a PR. Leukemic patients with only a decrease in WBC to the normal range were also considered to have an MR.

No response (NR) met none of the aforementioned criteria.

Bone marrow studies. All bone marrow biopsy specimens were reviewed initially by a hematopathologist (J.W.V. or M.A.B.). The bone marrow biopsy specimens on all patients not receiving IFN for at least 3 months were rereviewed by two of the authors (R.G.B. and J.W.V.) and analyzed as previously described. This included differential counts of 2,000 to 5,000 cells on the histological sections.

The volume of each marrow subpopulation was quantitated based on the estimated cellularity and the differential as follows: volume of subpopulation = percent cellularity × percentage of subpopulation/10,000. This analysis is similar to the stereological analysis of Flandrin et al1 and our prior use of the hairy cell index (HCI): 

\[
\text{percent cellularity} \times \text{percentage of hairy cells/10,000.}
\]

We have also quantitated the non-hairy cell index (NHCI) at various time points; it was defined as (the percent cellularity/100) − HCI. Occasional bone marrows were severely hypocellular (<10% cellularity), especially after IFN therapy was stopped. Frequently the peripheral blood counts obtained simultaneously suggested that these biopsy specimens were not considered representative of the patient's bone marrow, and thus nine of 56 marrows (19%) obtained after IFN therapy was stopped were excluded from further analysis.

Statistical analysis. Paired parametric (Student's t) and nonparametric (Wilcoxon) tests were used to analyze the changes in peripheral blood counts, NAP, and bone marrow subpopulations during and after IFN therapy. Patients with RBC or platelet transfusion requirements were considered to have a hemoglobin or platelet count of zero for the purpose of this analysis. In general, the Wilcoxon test was used to avoid assuming a normal distribution of data. However, the paired t-test was used to assess the changes in peripheral blood counts after completion of IFN therapy because of the greater power of this test.

RESULTS

Patient characteristics. We have entered 59 patients in our phase II trial from October 12, 1983, to March 15, 1986. Thirty-three of these patients have been reported previously, with follow-up through February 1985. Fifty-three of the 59 patients have completed at least 3 months of therapy (as of March 15, 1986) and are the subject of this paper (Table 1). Four of the remaining six patients have been recently entered in the study. One patient with refractory thrombocytopenia died of an intracerebral hemorrhage during the first week of therapy, and one patient with advanced HCL and severe emphysema (requiring continuous oxygen) died of pneumonia during the first month of therapy.

As noted in Table 1, four patients did not have a prior splenectomy. One patient refused a splenectomy, consistent with the stated eligibility criteria. In the other three nonsplenectomized patients, the decision to use IFN was made on clinical grounds because all three patients had small spleens and a high percentage of hairy cells in the marrow and would not be expected to have a durable response to splenectomy.11

All but one patient had one or more cytopenias or were in the leukemic phase of the disease. Forty patients (76%) were anemic (hemoglobin, <12 g/dL), and 19 patients (36%) were receiving RBC transfusions. Twenty-seven patients (51%) were thrombocytopenic (platelets, <100,000 cells/µL), and 12 patients (23%) required platelet transfusions. Most patients (70%) were neutropenic, and three patients had less than 100 neutrophils/µL prior to IFN therapy. Fourteen patients (26%) were in the leukemic phase of the disease. Five of our patients also had symptomatic bony lesions, including one patient with normal blood counts.

Responses. Of the 53 patients described earlier, 50 patients have completed at least 6 months of therapy. (The other three patients are still receiving IFN). The patient with normal blood counts at study entry, but symptomatic bony lesions, is not evaluable for hematologic response. Major responses were seen in 80% of the 49 evaluable patients, with ten CRs (20%) and 29 PRs (59%). An additional nine patients (19%) had MRs. Only one evaluable patient has not responded to therapy.

Two patients had IFN dose escalations from 2 × 10^6 U/m^2 to 10 × 10^6 U/m^2 after 4 to 5 months of therapy. The IFN dose in the single nonresponder was increased after 4 months of IFN. There was no improvement over the next 2 months, and IFN therapy was discontinued. No further therapy was administered (except for transfusions) over the next 16 months. The other patient in whom the IFN dose was escalated had not responded despite 5 months of IFN, although his ANC had increased from 144 to 620 cells/µL. He showed further improvement over the next month but was not considered a PR until 1 month after stopping IFN treatment (at the end of a full year).

Only one patient had a worsening of his blood counts while receiving IFN. The patient initially responded (MR) to IFN and was randomized to an additional 6 months of IFN at the end of 1 year. After 14 months of therapy, his blood counts deteriorated, and IFN therapy was discontinued.

Changes in peripheral and bone marrow parameters during IFN therapy. As shown in Table 2, the median hemoglobin level improved continuously over the first 12 months of IFN therapy, increasing from 10.6 g/dL to 14.7 g/dL. The platelet count improved dramatically over the first 3 months, but there was only minimal improvement with subsequent therapy. The ANC also improved rapidly, but
Fig 1. Changes in peripheral blood counts after stopping IFN therapy. The data represent the mean values at each time point. (HB, hemoglobin; PLTs, platelets.)

Table 2. Changes in Peripheral Blood and Bone Marrow Parameters During IFN Therapy

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Month (n)</th>
<th>0 (53)</th>
<th>3 (53)</th>
<th>6 (50)</th>
<th>9 (42)</th>
<th>12 (34)</th>
<th>18 (9)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hb (g/dL)</td>
<td></td>
<td>10.6†</td>
<td>12.2 (.001)†</td>
<td>13.7 (.0001)</td>
<td>13.8 (.003)</td>
<td>14.7 (.02)</td>
<td>15.3</td>
</tr>
<tr>
<td>Pts (x 1,000 cells/μL)</td>
<td></td>
<td>95</td>
<td>254 (.001)</td>
<td>240</td>
<td>249 (.06)</td>
<td>277 (.05)</td>
<td>292</td>
</tr>
<tr>
<td>WBC (x 1,000 cells/μL)</td>
<td></td>
<td>3.6</td>
<td>3.8</td>
<td>4.4 (.005)</td>
<td>4.4</td>
<td>5.0 (.003)</td>
<td>6.3 (.05)</td>
</tr>
<tr>
<td>ANC (x 1,000 cells/μL)</td>
<td></td>
<td>0.7</td>
<td>1.8 (.001)</td>
<td>2.0 (.009)</td>
<td>1.9</td>
<td>2.5 (.003)</td>
<td>2.9</td>
</tr>
<tr>
<td>BM cellularity (%)</td>
<td></td>
<td>78</td>
<td>50 (.006)</td>
<td>47</td>
<td>34 (.01)</td>
<td>45 (.02)</td>
<td>48</td>
</tr>
<tr>
<td>BM hairy cells (%)</td>
<td></td>
<td>90</td>
<td>70 (.001)</td>
<td>40 (.009)</td>
<td>24</td>
<td>21 (.006)</td>
<td>10 (.08)</td>
</tr>
<tr>
<td>HCl</td>
<td></td>
<td>0.71</td>
<td>0.30 (.001)</td>
<td>0.15 (.005)</td>
<td>0.07 (.001)</td>
<td>0.09</td>
<td>0.03 (.09)</td>
</tr>
<tr>
<td>NHCl</td>
<td></td>
<td>0.09</td>
<td>0.15 (.001)</td>
<td>0.25</td>
<td>0.18</td>
<td>0.35 (.01)</td>
<td>0.44</td>
</tr>
<tr>
<td>NAPI†</td>
<td></td>
<td>204</td>
<td>100 (.001)</td>
<td>77 (.0001)</td>
<td>31 (.003)</td>
<td>40</td>
<td>40</td>
</tr>
</tbody>
</table>

Abbreviations: Pts, platelets; BM, bone marrow.
*Due to randomization at 12 months to therapy v observation.
†Median.
‡P value v preceding time point by Wilcoxon paired signed-rank test (omitted if P > .1).
§P = .01 v 6 months.
‖P = .06 v 9 months.
¶Normal range, 13 to 100.

there continued to be significant improvement over the first year.

Most of the bone marrow changes also occurred during the first 9 months. The bone marrow cellularity significantly decreased over the first 3 months (Table 2) and decreased further from months 6 to 9, prior to increasing slightly from months 9 to 12. The relative percentage of hairy cells in the bone marrow appeared to decrease continuously over the first year, with a further decrease (P = .08) from months 12 to 18 (nine patients only). The HCl significantly decreased over the first 9 months of IFN, with little subsequent change. The NHCl increased over the first 3 months but changed little over the next 6 months. However, there was significant improvement in the NHCl from months 9 to 12.

Table 2 also illustrates the dramatic changes that occur in the NAP with IFN therapy. The median NAP decreased from 204 to 31 over the first 9 months of IFN, but there was no further change over the next 3 to 9 months.

Toxicity. The nonhematologic toxicity that occurred in out HCL patients was very similar to that described in prior reports.1,11 Constitutional symptoms occurred in all patients and consisted of a "flu-like syndrome," fatigue, and anorexia. Gastrointestinal symptoms were common and included nausea/vomiting (17%) and diarrhea (32%). Neurological toxicity included both peripheral neuropathies (32%) and CNS dysfunction (21%). The latter usually consisted of subtle memory loss or depression, except for one patient with Cancer and Leukemia Group B (CALGB) grade 3 confusion that required a temporary withdrawal of IFN therapy. Other common toxicities included injection site inflammation (45%), dry skin or seborrheic dermatitis (58%), alopecia (17%), and chemical hepatitis (74%). The latter was usually manifested as an elevated SGOT or SGPT level (79%), but the alkaline phosphatase value was occasionally elevated (28%). All toxicity was CALGB grade 1 or 2 (except for the one patient noted before), and no patient discontinued therapy due to toxicity, although one patient with severe fatigue refused randomization.

Transient myelosuppression was a very common event during the initial 1 to 2 months of therapy. Changes in the ANC were of the greatest magnitude (Table 3), but transfusions were occasionally required because of worsening anemia (two patients) and thrombocytopenia (two patients) due to IFN therapy.

Table 3. Transient Myelosuppression Due to IFN Therapy

<table>
<thead>
<tr>
<th>Parameter</th>
<th>n*</th>
<th>Percent Decrease</th>
<th>Time to Nadir (wk)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hemoglobin</td>
<td>33</td>
<td>8†</td>
<td>3</td>
</tr>
<tr>
<td>Platelets</td>
<td>36</td>
<td>25</td>
<td>1</td>
</tr>
<tr>
<td>ANC</td>
<td>47</td>
<td>54</td>
<td>2</td>
</tr>
</tbody>
</table>

*Patients were excluded from this analysis if they were transfusion dependent prior to IFN therapy, had no circulating neutrophils, or had an incomplete collection of blood counts.
†Median.
Follow-up after discontinuation of IFN therapy. Of the 53 patients described in this report, 34 patients have completed at least 12 months of IFN therapy. Twenty-six patients were entered in the randomized study, 13 patients in each arm (12 v 18 months of IFN therapy). Eight patients were not randomized, mainly due to the fact that the randomized study was not opened until February 1985. Twenty-four patients have not been receiving IFN for at least 3 months and were studied further, as described in Materials and Methods. Sixteen patients discontinued IFN therapy after 12 months, seven patients completed 18 months of IFN treatment (6 months after randomization), and one patient did not have a marrow prior to stopping IFN treatment.) Over the first 3 post-IFN months there was a decrease (not statistically significant) in the median cellularity (42.5% v 35%, P > .05) and an increase in the relative proportion of hairy cells (21.5% v 47.5%, P < .001). The decrease in total cellularity appears to be due to a decrease in the myeloid (17.6% v 8.3%, P < .001) and erythroid (18.3% v 11.7%, P < .01) volumes, but there was no significant decrease in the myeloid:erythroid ratio (0.9 v 0.6, P > .05). An example of these morphological changes is presented in Fig 3. There were no statistically significant changes over the subsequent 9 months, although there was an overall increase in the leukemic volume.

A small subset of six patients did have a major increase in HCL over the first 9 months after IFN treatment (Fig 4). Although the follow-up has been short, only one patient has required further therapy after relapsing with a high WBC (23,000 cells/µL) after 6 months of observation. His initial response status was MR, and he has responded to his second course of IFN over the first month of treatment.

DISCUSSION

This paper confirms our preliminary report of the effectiveness of IFN in advanced HCL.3 IFN, when administered at a dose of 2 x 10^6 U/m² three times weekly, leads to normalization of blood counts and improvement in the bone...
marrow in 80% of patients treated for at least 6 months. The therapy is well tolerated, with no patients discontinuing IFN treatment due to toxicity. Comparable results have been reported with the similar drug, interferon alfa-2a. Changes in the peripheral blood counts and bone marrow occur rapidly, with significant improvement over the first 3 months, especially in the platelet count. There continues to be improvement over the first year in both the peripheral blood counts and bone marrow.

One of the primary purposes of this analysis was to define the extent and course of hematologic changes occurring after discontinuing IFN therapy. We demonstrate that responses to IFN are quite durable, with only one of 24 patients followed for a median of 8.5 months who required further therapy. In addition, there were no significant changes in peripheral blood counts over the first post-IFN year, except for a rebound increase in all three cell lines demonstrated at months 1 to 3. There did appear to be significant changes in the bone marrow after stopping IFN therapy that were characterized by a decrease in normal myeloid and erythroid elements. In addition, there was a trend towards an increasing HCI over the first year after stopping IFN treatment, which has not yet reached statistical significance.

Study of the changes in the bone marrow after stopping IFN therapy may provide insight into the mechanisms of interferon-induced myelosuppression. The major hematologic toxicity of interferons is leukopenia, with a decrease of 40% to 60% in the WBC during the first week of treatment but with no further decrease with continued therapy. Ernstoff and Kirkwood have previously analyzed bone marrow in patients with nonhematologic malignancies who received IFN. There was a trend toward an increase in normal precursors but no maturation arrest. Our data demonstrate that there is a marked decrease in normal myeloid and erythroid marrow elements after stopping IFN therapy simultaneous with a rebound increase in the peripheral blood count. This suggests that IFN directly or indirectly inhibits the release of normal marrow elements and that this process is reversed when IFN therapy is discontinued—a hypothesis previously proposed by Ernstoff and Kirkwood.

We also demonstrate in this paper that IFN leads to normalization of another hematologic parameter that is abnormal in patients with HCL, the NAP. A previous report by Golomb et al noted an increased NAP in 15 of 17 patients studied. The median pretreatment NAP in our patients was markedly elevated at 204, which decreased to a nadir of 31 by month 9 but increased after stopping IFN therapy. A high NAP score may reflect a decrease in marrow myeloid elements, since these were the two major changes after discontinuing IFN treatment. A more detailed analysis of the relationship of NAP to peripheral blood counts and bone marrow subpopulations is in progress.

We continue to emphasize that IFN therapy is only palliative because discernible hairy cells remain in the bone marrow of virtually all patients. In addition, the bone marrow remains abnormal, even in complete responders, with increased reticulin and myeloid hypoplasia.

We propose that patients with progressive HCL following splenectomy be treated with IFN for at least 6 months. Responding patients should be treated for 1 year. Information regarding longer periods of treatment will be available after analysis of a multiinstitution study randomizing patients to 12 v 18 months of IFN therapy. Practicing physicians must be aware of the transient myelosuppression that occurs in the first 1 to 2 months of therapy, possibly precipitating the need for RBC and/or platelet transfusions. Following the discontinuation of IFN therapy patients should be monitored with complete blood counts every 1 to 3 months. Although the bone marrow may change significantly after IFN treatment is stopped, it should not necessarily be used as a guide to reinstitution of IFN therapy.

We cannot yet recommend IFN as initial therapy to patients in lieu of splenectomy, although Quesada and co-workers recently report the attainment of a CR in five of seven nonsplenectomized patients. In addition, the eventual role of IFN in the therapy of HCL is uncertain because of the high complete response rates (23% to 65%) reported from preliminary studies of the adenosine deaminase inhibitor pentostatin. If possible, patients with HCL should continue to be entered in clinical trials.

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REFERENCES

5. Ratain MJ, Golomb HM, Vardiman JW, Vokes EE, Jacobs
DURABILITY OF RESPONSE TO IFN IN HCL


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MJ Ratain, HM Golomb, RG Bardawil, JW Vardiman, CA Westbrook, LS Kaminer, BC Lembersky, MA Bitter and K Daly

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