Treatment of Diamond-Blackfan Anemia With Recombinant Human Interleukin-3

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This report describes the response of eighteen Diamond-Blackfan anemia (DBA) patients to recombinant human interleukin-3 (rhIL-3). rhIL-3 was administered subcutaneously once daily on an escalating dose schedule (0.5 to 10 μg/kg/d). The rhIL-3 dose was escalated every 21 days until erythroid response was attained, grade III or IV nonhematologic toxicity was observed, or the maximum rhIL-3 dose was reached. Four patients experienced clinically significant erythroid responses. Two of the responders were steroid-dependent and transfusion-independent, while two were steroid-independent and transfusion-dependent. Baseline clinical or laboratory parameters, in particular, in vitro bone marrow erythroid progenitor assays, were not useful in predicting rhIL-3 response. rhIL-3 administered at 5 to 10 μg/kg/d was associated with an increase in total white blood cell count, secondary to increases in neutrophils, eosinophils, and lymphocytes. Patients experienced a dose-dependent elevation in absolute eosinophil counts across the entire dose range. Two of the responding patients remain on maintenance rhIL-3, without diminution of effect at 244 and 370 + days. rhIL-3 was discontinued in the other two responders, because of the development of deep venous thrombosis.

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DIAMOND-BLACKFAN anemia (DBA) is a rare inherited anemia associated with macrocytosis, reticulocytopenia, and in the majority of cases, a normocellular bone marrow (BM) with a selective deficiency of erythroid progenitors.¹,² Phenotypic abnormalities are observed in less than 30% of patients. Inheritance in most cases is sporadic, although both autosomal dominant and autosomal recessive cases have been reported.

The current treatment of choice is corticosteroids. Approximately two-thirds of DBA patients respond, although only 50% of patients achieve long-term remissions, which are usually steroid-dependent.³,⁴ Approximately 20% experience spontaneous remissions, independent of age or management.⁵ Therefore, about one-quarter of patients require either prolonged supportive care with red blood cell (RBC) transfusions or alternative therapies. A limited number of responses have been reported after treatment with androgens, 6-mercaptopurine, cyclophosphamide, high-dose steroids, plasmapheresis, antithymocyte globulin, or cyclosporin A.⁶ Although rarely performed, allogeneic BM transplant can restore normal erythropoiesis in DBA patients.⁷,⁸

The precise pathophysiology of DBA is unknown, and may reflect multiple etiologies, given the heterogeneity observed in congenital anomalies, inheritance patterns, and response to steroid treatment.

Most DBA patients have a decreased number of clonable erythroid burst-forming units (BFU-E) and erythroid colony-forming units (CFU-E) measured in standard in vitro BM culture systems.⁹,¹⁰ Recent evidence suggests that the disease reflects abnormalities in the cascade of events leading to erythroid differentiation of early hematopoietic progenitors.¹¹-¹³ For example, Bagnara et al¹³ showed that in the presence of hematopoietic growth factors, CD34⁺-purified populations from DBA BM proliferate and differentiate into normal numbers of granulocyte/macrophage colonies (CFU-GM) and megakaryocyte colonies (CFU-MK). However, in the presence of erythropoietin (Epo), interleukin-3 (IL-3), interleukin-6 (IL-6), granulocyte-macrophage colony stimulating factor (GM-CSF), or erythroid potentiating activity (EPA), alone or in combination, differentiation of CD34⁺ cells to erythroid colonies (BFU-E) is severely decreased.

Currently, there is no evidence that DBA is caused by a deficiency in hematopoietic growth factors. DBA patients have normal levels of erythropoietin relative to their degree of anemia¹⁴ and their stimulated mononuclear cells produce normal amounts of IL-3 and GM-CSF.¹⁵ Although the hematologic abnormalities in DBA are similar to those seen in Steel (Sl) or W mice, genetic disorders that have been ascribed on one hand to a mutant stem cell factor (SCF) receptor, and on the other to a defective gene encoding the SCF receptor, c-kit,¹⁶,¹⁷ recent studies suggest that neither Sl nor W mice are appropriate models. Studies of c-kit and SCF genes in 21 DBA patients by Southern blotting failed to show any gross structural abnormalities.¹⁷,¹⁸ Abkowitz et al¹⁹ showed that BM mononuclear cells from two DBA patients expressed normal numbers of high-affinity SCF receptors, and that SCF serum levels were normal, as was nucleotide sequencing of c-kit and SCF messenger RNAs.

Despite the fact that DBA patients do not appear deficient in the hematopoietic growth factors that stimulate
erythropoiesis, it is theoretically possible that administration of these hematopoietic growth factors in pharmacologic doses might potentiate erythropoietic development. IL-3 is a hematopoietic growth factor that stimulates early erythroid progenitors.20,21 Halperin et al22 showed that the addition of IL-3 to in vitro cultures of BM from 5 of 6 DBA patients increased the number and size of BFU-E colonies and partially corrected their responsiveness to erythropoietin. In addition, recent phase 1/II clinical trials of recombinant human interleukin-3 (rhIL-3) in patients with bone marrow failure states have shown erythroid responses in a small proportion of patients.23-26 On the basis of these in vitro and in vivo data, we initiated a trial of rhIL-3 in DBA patients. To date, we have treated 18 patients. Four patients experienced clinically significant erythroid responses, again showing the heterogeneity of this disease.

MATERIALS AND METHODS

Patient characteristics and eligibility. Eighteen patients were enrolled on this study, whose clinical histories and baseline hematologic parameters (Table 1) were consistent with the diagnosis of DBA. Patients were eligible for enrollment if they required RBC transfusions or had a baseline hemoglobin <8 g/dL. Patients on steroids and desferoxamine were not excluded, and these drugs were maintained without dose modifications during the initial phase of rhIL-3 treatment. Patients were ineligible for enrollment if they had received another hematopoietic growth factor within 4 weeks or failed to meet standard criteria of adequate hepatic and renal function (serum aspartate aminotransferase [SGOT] and serum alanine aminotransferase [SGPT] <3 × upper limit of normal; creatinine <1.8 mg/dL and blood urea nitrogen [BUN] <30). This study was approved by the Institutional Review Board of Memorial Sloan-Kettering Cancer Center. Informed consent was obtained from all patients or their parents according to the guidelines of the Food and Drug Administration.

Recombinant human IL-3. Recombinant human IL-3 (SDZ ILE 964) was provided by the Cytokine Development Unit of Sandoz Pharmaceuticals Corporation (East Hanover, NJ). rhIL-3 was expressed in Escherichia coli by recombinant DNA technology.27 The 133 amino-acid protein is nonglycosylated and has a molecular weight of 14 to 16 Kd, and a specific activity in the chronic myeloid leukemia (CML)-proliferation assay of 2 to 10 X 10⁶ U/mg of protein. The study drug was supplied in a sterile, lyophilized, water-soluble form.

Study design. This was a single center, nonrandomized, open-label study designed to test the tolerability and biologic effects of rhIL-3 in pediatric patients with inherited cytopenias, of which DBA was a subgroup. Patients were treated consecutively in cohorts of six at the following initial rhIL-3 doses: 0.5, 1.25, 2.5, 5.0, and 10 mg QD. The initial dose was increased to 20 mg QD if tolerated. Patients were treated until adequate responses were noted, or until a maximum of 6 months of treatment was reached. Dosage was individualized on the basis of hematologic response. In addition, patients were treated with corticosteroids, splenectomy, and transfusion therapy, as determined by previous treatments. Clinical and laboratory data were analyzed before treatment and at regular intervals. AZT was also given to those patients with human T-cell leukemia (HTLV)-related lymphoma (CML)-proliferation assay of 2 to 10 X 10⁶ U/mg of protein. The study drug was supplied in a sterile, lyophilized, water-soluble form.
10.0 μg/kg/d, (designated levels 1 through 5). rhIL-3 was administered by single daily subcutaneous injection for 21 consecutive days. After the first 21-day treatment cycle, patients (except patients 1 and 2) underwent a 21-day washout period in which no rhIL-3 was administered. Patients exhibiting no response or a partial response (defined as hemoglobin maintenance of 8 to 10 g/dL) were escalated to the next dose level for another 21 days of treatment. Dose escalation continued without further washout periods until a complete response was achieved (defined as maintenance of hemoglobin >10 g/dL without transfusion support) or patients reached the maximum dose level (10 μg/kg/d). Patients not showing a response were discontinued from the study. Patients achieving a complete response were continued on daily subcutaneous rhIL-3 injections at the dose at which the response was observed. Patients escalating to the maximum dose and achieving only a partial response were also continued on maintenance at this rhIL-3 dose. Patients continued to receive RBC transfusions if the hemoglobin decreased to levels at which they were symptomatic (range 5.0 to 8.0 g/dL). rhIL-3 was discontinued for nonhematologic grade III or IV toxicity related to the study drug. Toxicity gradings were based on modified National Cancer Institute (NCI) toxicity parameters.28 If the RBC response was maintained at least 2 months on maintenance therapy, steroids were slowly tapered on steroid-dependent patients.

Clinical and laboratory assessments. Before treatment, detailed clinical histories were obtained, which included blood counts and transfusion records for the preceding 6 months. In addition, patients underwent complete physical examination, chest x-ray, electrocardiogram, and urinalysis. Laboratory analysis included complete blood count including manual differential and reticulocyte count, routine chemistry profile, coagulation parameters, ferritin, iron studies, hemoglobin electrophoresis, haptoglobin, erythropoietin levels, and whole-blood histamine levels. BM aspiration was performed for morphologic analysis and in vitro quantitation of BM progenitor levels. During the dose-escalating treatment cycles (induction), a complete blood count with manual differential and reticulocyte count was obtained twice per week, and biochemistry profile, whole-blood histamine level, and coagulation profile every 3 weeks. BM aspiration was performed every 3 weeks and analyzed as described at baseline. During maintenance rhIL-3 therapy, complete blood count with manual differential and reticulocyte count was obtained weekly and chemistry profile, coagulation profile, and whole blood histamine levels every 3 weeks. Ferritin, iron studies, Epo levels, and hemoglobin electrophoresis were obtained approximately every 3 months.

During induction and maintenance, patients kept daily logs documenting rhIL-3 dose, injection site, adverse events, concomitant medications, and daily temperatures. Patients or their parents were taught to administer the subcutaneous rhIL-3 injections. Patients were examined weekly by the principle investigator (PI) or the referring physician. Regular telephone contact was maintained with the PI and/or research nurse.

In vitro BM progenitor assays. BM mononuclear cells (BMMNC) were obtained after separation with Lymphoprep (Nycomed Pharma, Oslo, Norway; density: 1.077 ± 0.001 g/mL) and plated at cell concentrations ranging from 0.5 to 2 × 10^5 per mL. Depending on cell recovery, BMMNC were plated in triplicate or duplicate in 1-mL 35-mm Petri dishes in 1.290 methylcellulose with various combinations to stimulate BFU-E growth.

Colonies were counted after incubation for 14 ± 1 days at 37°C in 5% CO2 and 7% O2. BFU-E were defined as either two or more clustered hemoglobinized subclones or a single large colony composed of at least 100 hemoglobinized cells.

RESULTS

Erythroid lineage response. Four of the 18 rhIL-3-treated DBA patients experienced clinically significant erythroid responses. At baseline, two responders (pts 2 and 16) were steroid-dependent, transfusion-independent (Fig 1) and two responders (pts 5 and 13) were steroid-independent, transfusion-dependent (Fig 2).

Patient 2 entered the trial with a hemoglobin of 8 g/dL on 7.5 mg of prednisone daily (Fig 1). She received 21-day cycles of rhIL-3 at doses of 0.5 and 1.25 μg/kg without significant erythroid response. At rhIL-3 doses of 2.5 μg/kg/d, she developed a brisk reticulocytosis followed by a significant increase in hemoglobin. She was continued on daily dose of maintenance rhIL-3 at 2.5 μg/kg and on day 150 of the trial achieved a maximum hemoglobin of 13.5 g/dL. Subsequently, her steroids were tapered over a 3-month period and then discontinued. Although she experienced a slight decline in hemoglobin and absolute reticulocyte count (ARC) during this taper, off steroids her hemoglobin stabilized in the 10.5 to 12.2 g/dL range. She continues on rhIL-3 treatment at 370+ days.

![Fig 1. Effect of rhIL-3 administration on Hgb (m) and ARC (c) in two steroid-dependent transfusion-independent DBA patients. rhIL-3 dosing is depicted by the horizontal arrows and steroid taper by dotted bars.](www.bloodjournal.org)
Patient 13, before rhIL-3 treatment, required RBC transfusions every 4 weeks (Fig 2). On induction cycles of rhIL-3 at 5.0 µg and 10 µg/kg/d, he developed a dose-dependent reticulocytosis. On maintenance rhIL-3 dosing at 10 µg/kg, his transfusion requirement decreased by 50% with an 8-week interval between transfusions. On day 131 of the trial, the patient was hospitalized because of complaints of a progressively tender, swollen right calf and distal thigh. At that time, complete blood counts showed a WBC count of 22.0 × 10⁹/L, Hgb 10.3 g/dL and platelets of 337 × 10⁹/L with an AEC of 9,240 cells/mm³. On detailed questioning, the patient admitted to having fallen on his mid-thigh at a hockey game 10 days earlier. Doppler studies revealed a DVT involving the right popliteal and femoral veins. IL-3 was discontinued and his absolute reticulocyte counts and transfusion requirements returned to their prestudy baseline levels. Anticoagulation resulted in full resolution of the clot without sequelae.

Clinically relevant erythroid responses or significant changes in ARC were not observed in the remaining 14 patients.

**Effects on other erythroid parameters.** Tables 2 and 3 summarize various laboratory parameters measured sequentially in patients 2 and 5 (responders). On rhIL-3 therapy both patients experienced increases in BM cellularity without concomitant change in BM myeloid:erythroid cell ratios or percentage of BM-nucleated erythroid cells. Both patients exhibited decreases in ferritin and total iron levels. In patient 2, during the initial increase in hemoglobin there was a concurrent decrease in Hgb F%, mean corpuscular volume (MCV), and EPO levels. However, with the drop in hemoglobin of 1 to 2 g/dL during the steroid taper, these parameters increased, although remained below initial baseline levels. In patient 5, whom was transfusion-dependent at study entry, initial erythroid parameters reflected transfused RBCs. During rhIL-3 therapy, as transfusion-independence was achieved, the characteristic DBA erythroid phenotype of stress erythropoiesis became evident with elevation of MCV and Hgb F%. Although not shown, the other two responding patients experienced similar patterns of change in these parameters.

**Effects on nonerythroid cell lineages.** To determine the effect of rhIL-3 administration on nonerythroid lineages, the mean total WBC count, mean absolute neutrophil, lym-
phocyte, monocyte, eosinophil, and basophil counts and mean platelet counts were calculated for the entire group of 18 patients at baseline and at the end of every 21-day treatment cycle. The mean WBC count was increased significantly above baseline at rhIL-3 doses of 5.0 and 10.0 μg/kg/d primarily because of modest increases in absolute neutrophil and lymphocyte counts (Fig 3). In addition, patients experienced a dose-dependent elevation in absolute eosinophil count across the entire dose range (Fig 3). The median AEC at rhIL-3 dosings of 5.0 and 10.0 μg/kg/d were 1,320 and 2,992 cells/mm³, respectively. Monocyte, basophil, and platelet counts were not affected over the rhIL-3 dose range examined (data not shown).

**Progenitor cells.** At study entry, clonable BM BFU-E were analyzed in all but one patient (pt 18), using a variety of erythroid colony-stimulating factors alone or in combination (Fig 4). With Epo alone, all patients exhibited very low numbers of BFU-E (range 0 to 28, mean 3) as compared with 15 controls (range 68 to 125, mean 96). The growth of in vitro BFU-E with IL-3 + EPO stimulation was not predictive of in vivo rhIL-3 erythroid response. Two of the responding patients exhibited only minor increases in clonable BM BFU-E and the other two responding patients displayed no in vitro BFU-E colony growth with EPO + IL-3. Conversely, many of the patients with substantial augmentation of BFU-E with IL-3 in vitro did not display erythroid responses to exogenously administered rhIL-3 (including patient 3 who displayed a near normalization of BFU-E colonies). The addition of KL to in vitro BM cultures, either in combination with EPO or EPO + IL-3, resulted in the greatest stimulation of BFU-E growth in controls and most of the DBA patients. However, again only one DBA patient achieved normal BFU-E numbers compared with controls and this parameter was not helpful in predicting erythroid response to exogenously administered rhIL-3.

**Toxicity of escalating doses of rhIL-3.** The majority of patients developed fever and/or chills following the first dose of rhIL-3 and thereafter took antihistamines and acetaminophen before rhIL-3 administration. Therefore, the incidence of fever and chills cannot be accurately assessed at each dose level. Headaches, flushing, bone and/or joint pains, myalgias, irritability, and gastrointestinal complaints were experienced with increasing frequency with rhIL-3 dose escalation (Table 4). These events were mild to moderate (grade 1 or 2) and did not require drug discontinuation or medical intervention. Erythema and pruritus at the injection site were frequent complaints at all dose levels. In general, these local reactions developed within 2 hours of injection and resolved within 24 hours.

Patient 5 experienced mild paresthesias of the digits on rhIL-3 doses of 2.5 to 10.0 μg/kg/d. Patient 7 developed hypotension after injection of 10 μg/kg of rhIL-3 requiring fluid resuscitation and prompting rhIL-3 discontinuation. As noted previously, two patients developed DVT on maintenance rhIL-3 therapy.

No changes were observed in hepatic, renal, or coagulation parameters. Whole-blood histamine levels did not increase during rhIL-3 treatment.

**DISCUSSION**

This report describes the response of the largest group of DBA patients treated with rhIL-3 to date. Four of 18 patients experienced a clinically significant erythroid response and two remain on long-term rhIL-3 therapy at 244+ and 370+ days.

Dunbar et al²⁹ reported treatment of six transfusion-dependent, steroid-unresponsive DBA patients with rhIL-3 (Hoechst-Roussel Pharmaceuticals [Somerville, NJ] in collaboration with Immunex Corp [Seattle, WA]) at doses of 60 to 125 μg/m²/d administered subcutaneously for 4 to 6 weeks. Significant increases in ARC were observed in three patients, two of whom remained transfusion-independent once rhIL-3 therapy had been discontinued. This response rate is consistent with our data, but the long-lasting response after discontinuation of rhIL-3 is inconsistent with the responses observed in patients 5 and 13 of this report. Both of these patients, who discontinued rhIL-3 because of toxicity,
experienced loss of erythroid effect and returned to their baseline transfusion requirements within 8 to 10 weeks of rhIL-3 discontinuation. Olivieri et al.30 also treated seven steroid-unresponsive, transfusion-dependent DBA patients with rhIL-3 (Hoechst-Roussel/Immunex) at doses of 125 to 500 μg/m²/d. Only 1 of the 7 patients experienced an increase in ARC, but this was not sustained on maintenance rhIL-3 therapy. In both these studies, the rhIL-3 administered was a yeast-derived glycosylated product, while the rhIL-3 used in the present study is E coli derived and non-glycosylated. Glycosylation may alter the bioavailability, pharmacokinetics, and receptor binding of the cytokine, which may explain the differences observed in response in these three studies.

Alternatively, discordance in response rates may be due to differences in patient selection. In phase I/II trials of G-CSF and GM-CSF treatment of aplastic anemia, neutrophil responses were observed more frequently in patients with baseline ANC greater than 200 cells/mm³ than in patients with ANC less than 200 cells/mm³.31,32 Erythroid activity (defined crudely by ARC and % BM-nucleated erythroid cells) varies greatly in DBA patients. If degree of erythroid activity influences response to rhIL-3, this may account for the different response rates observed in these three trials. In the Olivieri trial,30 in which there were no long-term clinical erythroid responses, criteria for study entry were an ARC of 0 and absence of BM-nucleated erythroid cells. In the Dunbar trial,29 responders exhibited greater erythroid activity than nonresponders at study entry. In the present study, the four responders all displayed erythroid activity at study entry (ARC ranged from 6 to 21 × 10⁹/L and % BM-nucleated erythroid cells from 6 to 33%) (Table 1). However, a number of patients with similar erythroid activity did not respond. As in the Olivieri trial, the patients (14, 15, 17) with no detectable erythroid activity showed no response. Thus, it appears that while the presence of erythroid activity does not predict clinical response to rhIL-3, lack of measurable erythroid activity has been associated with failure to

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**Table 4. Toxicities Associated With Escalating Doses of rhIL-3**

<table>
<thead>
<tr>
<th>Toxicities</th>
<th>rhIL-3 Dose μg/kg/d</th>
<th>No. of Patients Experiencing Adverse Events (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.5 (5)</td>
<td>1.25 (7)</td>
</tr>
<tr>
<td>Headache</td>
<td>0 (0)</td>
<td>1 (14)</td>
</tr>
<tr>
<td>Flushing</td>
<td>2 (40)</td>
<td>2 (28)</td>
</tr>
<tr>
<td>Bone/joint pain</td>
<td>0 (0)</td>
<td>2 (28)</td>
</tr>
<tr>
<td>Muscle aches/malaise</td>
<td>1 (20)</td>
<td>2 (28)</td>
</tr>
<tr>
<td>Site erythema/pruritis</td>
<td>3 (60)</td>
<td>4 (57)</td>
</tr>
<tr>
<td>Irritability</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Nausea/vomiting</td>
<td>0 (0)</td>
<td>1 (14)</td>
</tr>
<tr>
<td>Edema</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Paresthesias</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Hypotension</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>DVT</td>
<td>0 (0)</td>
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respond and may account for the difference in response rates observed in these three trials.

In a further attempt to identify predictors of response to rhIL-3, we examined other baseline characteristics in our 18 patients including sex, initial response to steroid therapy, current steroid usage, associated congenital abnormalities, and use of chelation therapy and found no differences between responders and nonresponders. Epo levels were assessed at baseline, often 1 to 3 days after RBC transfusion, and therefore unlikely reflect the concurrent hemoglobin. Therefore, Epo level could not be assessed as a predictor of IL-3 response. Surprisingly, in vivo erythroid response to rhIL-3 could not be predicted on the basis of in vitro BM BFU-E growth, in response to Epo, Epo + IL-3, or Epo + IL-3 + KL. These results are similar to those observed by Alter et al in the same patients. Olivieri et al also observed that 0 of the 7 DBA patients with an IL-3-induced increase in BFU-E-derived colonies in vitro showed an in vivo response to rhIL-3. There is no obvious explanation for this discrepancy between the in vitro and in vivo results. Perhaps we are analyzing the wrong progenitor cells (ie, only more differentiated) or are not supplementing adequate quantities of other cytokines or agents that are available in vivo to act in concert with rhIL-3. In the case of in vitro responders that do not respond to exogenously administered rhIL-3, there may be inhibitors of rhIL-3 action in vivo that are not in the in vitro system.

rhIL-3 administration at doses of 5 to 10 µg/kg resulted in an elevation of total WBC counts secondary to increases in neutrophils, lymphocytes, and eosinophils. Because all patients developed eosinophilia in a dose-dependent fashion, eosinophilia is a reliable measure of biologic effect.

rhIL-3 was well tolerated by most patients during short-term induction therapy. However, of the 4 patients placed on maintenance therapy, 2 developed DVT requiring rhIL-3 discontinuation. DVT is uncommon in children and not associated with DBA. Although the DVT experienced by patient 13 could be related to local trauma, patient 5 denied a history of trauma and had no evidence of infection near the site of the thrombosis. In both patients, thrombosis occurred weeks after the development of a stable hemoglobin and therefore was unlikely secondary to a rapidly increasing blood viscosity. The patients had normal platelet counts and coagulation parameters (prothrombin time, partial thromboplastin time, fibrinogen level). Antithrombin III (AT III) and protein C and S levels measured after discontinuation of rhIL-3 were normal, but levels at the time of the event are unknown. AT III and protein C and S levels are currently being evaluated in other patients on long-term rhIL-3 treatment to rule out rhIL-3–induced decreases in these factors.

At the time of the thrombotic events, patients 5 and 13 had elevated AEC of 10,064 and 9,240 cells/mm3, respectively. Chronic hypereosinophilia has been associated with thromboembolic events in a variety of conditions including idiopathic hypereosinophilic syndrome (HES). HES patients experience both small and large vessel thrombi, thought to be secondary to emboli released from intracardiac thrombi or caused by eosinophilic-induced damage to the vascular endothelium. In experimental models, eosinophil secretion products such as major basic protein (MBP) have been shown to damage endothelial cells. In addition, eosinophilic cationic protein (ECP), which is another eosinophilic agent, may act as a hypercoagulant contributing to the possibility of thrombosis.

It is possible the development of DVT in these two patients was related to hypereosinophilia; however, the correlation between DVT and prolonged rhIL-3 induced hypereosinophilia is not entirely clear. Patients 2 and 16, who continued on long-term rhIL-3 therapy, have not developed DVT despite similar AEC elevations for even more prolonged periods than patients 5 and 13. Although both patients were on steroids at initiation of rhIL-3 treatment, steroids did not alter the dose-dependent elevation of eosinophils. Because both patients have been off steroids now for months, it is unlikely steroids have prevented the development of DVT. Nonresponders in this trial also developed eosinophilia, although compared with the patients on maintenance, AEC elevation was less (Fig 3) and of a much shorter duration.

Currently, the short- and long-term consequences of rhIL-3–induced eosinophilia are unknown. The nature of rhIL-3–induced eosinophils are being investigated, as well as alternative rhIL-3 treatment schedules designed to decrease the degree of eosinophilia (ie, intermittent dosing). At the same time, the benefit of rhIL-3 in at least a few DBA patients suggests that the long-term efficacy and frequency of response should be examined on a larger scale.

REFERENCES

Treatment of Diamond-Blackfan anemia with recombinant human interleukin-3 [see comments]

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