Low-Dose Deoxycoformycin in the Treatment of Hairy Cell Leukemia

By Eric H. Kraut, Bertha A. Bouroncle, and Michael R. Grever

Ten patients with progressive hairy cell leukemia were treated with 2'deoxycoformycin (dCF) by intravenous bolus (4 mg/m²) given every other week. All ten patients are evaluable for response and nine of the ten patients have achieved a complete remission. In addition to clearing of hairy cells from the bone marrow, eight patients had resolution of their monocytopenia. Seven of the nine patients remain in unmaintained remission with a median duration of 6.2 months. Two patients have had relapse in the bone marrow alone and continue to have normal peripheral blood counts. They are being followed without treatment. Toxicity was minimal at this low dose with one patient having a mild, reversible reduction in creatinine clearance. Four other patients had reversible neutropenia. There were no significant infections associated with treatment. Low-dose deoxycoformycin administered intravenously every other week represents an extremely effective treatment for hairy cell leukemia.

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Hairy cell leukemia was first described at the Ohio State University by Bouroncle, Wiseman, and Doan in 1958, as a new hematologic and pathologic entity. It is a chronic form of leukemia usually manifested by splenomegaly, pancytopenia, and the presence in the blood and bone marrow of the pathognomonic hairy cell, a mononuclear cell with cytoplasmic projections. Present evidence suggests that this is a lymphoproliferative disease that like chronic lymphocytic leukemia originates most frequently in the B lymphocyte line. The clinical course is variable with some patients remaining asymptomatic for months to years, while others require treatment for symptoms related to hypersplenism, recurrent infection, or bone marrow failure.

The standard treatment for these patients has been splenectomy. Although splenectomy may improve the hematologic complications of this disease, it may not prolong survival. Moreover, a significant number of patients do not respond or relapse after splenectomy. Attempts have been made to develop other effective treatment for hairy cell leukemia. Low-dose chlorambucil has offered palliation in some patients but does not completely reverse the pancytopenia or markedly alter the bone marrow infiltration. Intensive chemotherapy has been used successfully on only a few patients, and has been associated with significant morbidity and mortality.

Bone marrow transplantation has been accomplished in one patient with hairy cell leukemia, but the associated risk and age limitations make it unfeasible in most patients. Several studies of alpha interferon in patients with this disease have demonstrated response rates of 43% to 94% with hematologic improvement and clearing of hairy cells from the bone marrow. It is too early, however, to determine the effect of alpha interferon on survival.

In 1980, we began a study of the experimental drug 2'deoxycoformycin (dCF) in patients with lymphoproliferative malignancies. This agent is a potent inhibitor of adenosine deaminase, which is an important enzyme in purine catabolism. During the course of our early investigations, we demonstrated activity of this drug in a patient with refractory hairy cell leukemia. Therefore, we initiated a specific phase II study of dCF in patients with hairy cell leukemia, the results of which are reported here.

MATERIALS AND METHODS

In May 1984, we began the study of low-dose dCF 4 mg/m² administered intravenously every other week in patients with hairy cell leukemia. Informed consent approved by the Ohio State University Human Subjects Review Board was obtained before administration of Deoxycoformycin. 2'Deoxycoformycin was obtained from the Investigational Drug Branch National Cancer Institute (NCI), Bethesda, MD. The eligibility criteria for entrance to the study included the following: (1) A confirmed diagnosis of hairy cell leukemia based on clinical characteristics and demonstration of hairy cells in the peripheral blood and bone marrow examination. (2) Progressive disease as demonstrated by anemia (hemoglobin <12 g/dL), thrombocytopenia platelets <100,000/μL), neutropenia (neutrophils <1,500/μL), and/or leukemia (white blood cell count >10,000/μL with more than 50% hairy cells. (3) Patients with normal hepatic and renal function with a creatinine clearance >50 mL/min.

Six of our ten patients had been splenectomized. However, splenectomy was not a requirement before receiving a trial of dCF. The non-splenectomized patients had either refused this operation or were not considered to be good surgical candidates. Patients received dCF at a dose of 4 mg/m² by intravenous bolus injection every other week. Patients were carefully monitored for a change in clinical status by physical examination and peripheral blood count with a differential prior to each treatment. Treatment was modified in the face of a significant change in renal function or neutropenia according to the following guidelines. A serum creatinine was determined on each patient in the outpatient clinic immediately before each treatment. If the value had increased by more than 20% over the baseline value, the dose was held until a 24-hour urine for creatinine clearance was obtained. The drug was administered only when the creatinine clearance was 50 mL/min or better. If the white blood cell count had decreased below 1,500/μL after drug administration, then the drug was held until recovery or bone marrow evaluation demonstrated that this reduction was secondary to disease.

Patients were seen every other week and response assessment was made after a minimum of two courses of therapy. Criteria for response were defined as follows: (1) Complete remission (CR): The...
absence of hairy cells in the bone marrow aspirate and biopsy with restoration of the peripheral blood counts to the following values: Hemoglobin >12 g/dL, platelets >100,000/μL, and neutrophils >1,500/μL. (2) Partial remission (PR): Absence of hairy cells in the peripheral blood and a 50% or greater reduction in the percentage of hairy cell infiltration in the bone marrow biopsy, and improvement in peripheral counts as noted under CR. (3) Minor Response (MR): Improvement in one or more of the peripheral blood elements as defined above, or greater than 50% reduction in circulating hairy cells.

Patients who had obtained a complete remission were followed off treatment with duration of response measured from the time that the remission was documented.

Adenosine deaminase assay. Determination of the ADA activity of the peripheral blood buffy coat preparation was performed before and one-half hour following the intravenous administration of dCF to document that enzyme inhibition in circulating leukocytes occurred in vivo. The buffy coat preparation was obtained by dextran sedimentation from heparinized blood. The cells were washed twice in Seligmann's balanced salt solution with 1% EDTA, resuspended in 5 mmol/L Tris-HCl (pH 7.4) and 0.25 mol/L sucrose, and counted. After sonication for 30 seconds, the homogenate was centrifuged at 50,000 g and 4°C for 30 minutes. Supernatant ADA was assayed in triplicate.

The enzyme assay was carried out in a total volume of 70 μL containing 5 mmol/L Tris-HCl (pH 7.4) and 4 mmol/L adenosine at 37 °C. Radiolabeled 8-14C-adenosine (ICN Pharmaceuticals, Inc., Irvine, Calif), 54 mCi/mmol, was added to the cold substrate to detect product conversion. After ten minutes, the assay was terminated by adding 20 μL of 4 mol/L formic acid and placing the sample in an ice bath. A 5-μL aliquot was placed on a cellulose acetate thin-layer chromatography plate (Analabs, Inc, North Haven, Conn) and developed in water for 45 minutes. The addition of unlabeled markers (adenosine and inosine) at the origin permitted identification of both substrate and product migration bands for scavenging and subsequent counting in a scintillation counter. The ADA activity was expressed as micromoles of inosine formed per hour per 10^6 leukocytes.

Since there was a variable percentage of circulating hairy cells present in the peripheral blood of each patient, an ADA assay was also performed on a mononuclear cell preparation of the peripheral blood before dCF administration to provide an estimate of the enzyme activity in a preparation enriched for the hairy cells. The mononuclear cell preparation was obtained from heparinized peripheral blood subjected to a Ficoll-hypaque (Pharmacia, Piscataway, NJ) separation. The cells were washed with Seligmann's balanced salt solution with 1% EDTA, resuspended in 5 mmol/L Tris-HCl (pH 7.4) and 0.25 mol/L sucrose, and counted. The mononuclear cells were sonicated on ice for 60 seconds. The homogenate was centrifuged at 50,000 g at 4°C for 30 minutes. Supernatant ADA was assayed as previously described.26

RESULTS

The characteristics of the ten patients are shown in Table 1. The mean age was 53 years (range 37 to 70) and nine of the patients were male. Six of the ten patients had previously been splenectomized and the median time to treatment after splenectomy was 28 months (range 6 to 67 months). Three patients had received chemotherapy in addition to splenectomy, either chlorambucil or 6 mercaptopurine. None of the patients were treated with interferon. The initial peripheral blood counts prior to treatment are demonstrated in Table 2. Five patients were granulocytopenic, seven patients were anemic, and seven patients were thrombocytopenic. All patients had hypercellular bone marrow biopsies with diffuse infiltration with hairy cells.

All ten patients are evaluable for response and toxicity. Nine of the ten patients have obtained a documented complete remission demonstrated by normalization of blood counts (Table 2) as well as clearing of hairy cells from the bone marrow. The average number of courses to complete remission was six (range 4 to 13). Seven of the nine patients that achieved complete remission remain in remission without maintenance therapy from 1 to 10 months. Two patients (Nos. 1 and 3) have had bone marrow relapse only at 12 and 8 months, respectively. Their peripheral blood counts remain normal and they are being followed without treatment until deterioration of peripheral blood counts occur. In contrast to the difficulty obtaining a bone marrow aspirate before a complete remission were able to be aspired. In addition, eight patients who were severely monocytopenic prior to treatment (<100 monocytes/μL) had a demonstrable increase in the peripheral blood monocyte counts (250 to 1,080 monocytes/μL).

One of the patients who entered into complete remission had been the first patient entered on protocol and his course was quite instructive. He had originally reached a partial remission after only two doses of dCF. Further treatment was held when his creatinine clearance dropped from 55 to 43 cc/min, although this later returned to baseline level. He was followed off therapy for 9 months until there was evidence of disease progression. His therapy was restarted at 75% of the initial dose to avoid renal toxicity. He tolerated the dCF reasonably well and obtained a complete remission after 12 doses of dCF.

The only patient who has not achieved complete remission status started out with a high peripheral white blood cell count of 159,000/μL, consisting of predominantly hairy cells and has slowly responded with reduction in the peripheral blood count to 20,000.

Toxicity with this low-dose regimen was mild. Most patients reported mild to moderate nausea lasting for less than 24 hours controlled by antiemetics. Only one patient had a reversible reduction in creatinine clearance. Four patients had therapy temporarily held due to reversible neutropenia. One patient developed an inflammation of

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Abbreviation: 6MP, 6 mercaptopurine.

Table 1. Patient Characteristics
multiple solar dermatitis. This resolved when the drug was held, and the patient had no further problems. No significant infectious complications developed related to dCF treatment. One patient who had been hospitalized several times for treatment of infection secondary to an esophageal fistula and neutropenia developed pneumonia 2 weeks after dCF was initiated. This resolved on antibiotics and appeared to be a complication of his underlying problem and not secondary to the drug. One patient developed a fever within 24 hours after receiving the drug, and it resolved within 48 hours.

Adenosine deaminase activity was measured on buffy-coat preparations and peripheral blood mononuclear cell preparations when cell numbers allowed. Values differed significantly between the buffy-coat and the mononuclear cell preparation in each individual, and there was also a wide range of values among individuals (Table 3). Significant inhibition of ADA activity was achieved in all but one patient during the first course of therapy, but in subsequent courses inhibition was achieved in this patient. There was no correlation between ADA values and response to therapy or time to response.

DISCUSSION

Our results demonstrate that low-dose dCF is a highly effective treatment for patients with hairy cell leukemia. Nine of ten evaluable patients have obtained a complete hematologic remission with minimal toxicity. Although our study involves a limited number of patients, the response rate of 90% is equal to the best results achieved with alpha interferon.15-18 Complete remissions in this disease following treatment with dCF have also been reported by two other groups using this drug with a slightly different dose and schedule.21-24 Complete remissions were associated with clearing of hairy cells from the peripheral blood and bone marrow, clearance of bone marrow fibrosis, and ability to aspirate marrow. We also observed reversal of the severe mononcytopenia characteristic of hairy cell leukemia.

Seven of the nine patients remain in unmaintained remission for up to 10 months. Two patients have had early relapse with recurrence of hairy cells in the bone marrow but no change in their peripheral blood counts. Since they are doing well clinically, they are being followed carefully without treatment until further evidence of disease progression occurs. This decision was based on experience obtained from studies on the natural history of this disease19,28 which suggests that disease progression may be slow and normalization of peripheral blood counts may be sufficient to prolong survival. Furthermore our experience with one patient who initially progressed off therapy and then subsequently responded to re-institution of dCF suggests that patients can be successfully retreated. Whether drug resistance will develop in patients awaits further testing.

In the earlier phase I clinical trials when patients were treated with higher doses of dCF, prohibitive renal, neurologic, and ocular toxicities were noted.20,25 We have recently reported that low doses of dCF (4 mg/m²) administered to a large number of patients with chronic lymphocytic leukemia were well tolerated.19 In both studies complications have been minimal with low-dose dCF. In fact, in our group of patients with hairy cell leukemia who were at high risk of infection due to neutropenia and monocytopenia, there was no apparent increase in infections observed.

This study was originally initiated because dCF had shown activity in heavily treated patients with chronic lymphocytic leukemia and other lymphoproliferative malignancies.19,26 However, the exquisite sensitivity of hairy cell leukemia to the drug was unexpected. It has been suggested that the toxicity of dCF to normal as well as malignant lymphocytes can be explained by depletion of cellular nicotinamide-adenine dinucleotide (NAD) and subsequently adenosine triphosphate.27 Evaluation of nucleotide pools and NAD in vivo following administration of dCF is currently being explored. The exact mechanism of tumor cell cytotoxicity, however, remains unknown.

Since the description of hairy cell leukemia in 1958, the recommended management has been conservative with...
spleenectomy being the treatment of choice in patients with progressive disease. Chemotherapy was infrequently used due to low response rates and significant toxicity. The success of dCF in our patients with progressive hairy cell leukemia including four who had not been splenectomized suggests that it also should be used in the nonsplenectomized patient regardless of spleen size. With the demonstration that two active agents exist for the treatment of hairy cell leukemia, the management of these patients needs reevaluation. Both alpha interferon and dCF offer significant palliation for this disease. Several multigroup studies are now underway to evaluate which of these agents is most appropriate for both the nonsplenectomized and splenectomized patients with progressive disease.

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REFERENCES

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