Bone Marrow Transplantation for Children With Myelodysplastic Syndromes

By Eva C. Guinan, Nancy J. Tarbell, Ramana Tantravahi, and Howard J. Weinstein

Therapeutic options for children with de novo or secondary myelodysplastic syndromes (MDSs) are limited. We report the outcome of eight pediatric patients (median age 12 years, range 3 to 19 years) who underwent allogeneic bone marrow transplantation between 1984 and 1987. Two of the eight children had developed secondary myelodysplasia after alkylating agent-based combination chemotherapy. Five patients had clonal chromosomal abnormalities, including four patients with monosomy 7. Seven of eight patients engrafted. Two of these seven subsequently died of complications of acute or chronic graft-versus-host disease (GVHD), and a third patient died at 21 months of pulmonary fibrosis. None of the patients have had recurrence of disease. The four surviving patients remain in complete remission at a median follow-up of 19 months (range 10 to 44 months).

M Y E L O D Y S P L A S T I C syndromes (MDS) represent a spectrum of clonal hematologic disorders which have in common peripheral blood cytopenias in the setting of dysplastic and generally hypercellular bone marrows, although some patients may have hypopcellular or even frankly aplastic marrows. Most patients with these disorders are older (median age 62 to 65 years), although MDS has been estimated as accounting for ~3% of childhood hematologic malignancies and 17% of pediatric patients developing acute nonlymphocytic leukemia (ANLL). In both pediatric and adult survivors of malignancy who have been exposed to alkylating-agent chemotherapy with or without irradiation, MDS is being diagnosed with increased frequency.

Both de novo and secondary MDS are marked by a high rate of evolution into overt ANLL. However, the complete remission (CR) rate after intensive chemotherapy in pediatric patients with MDS has been substantially lower than that in children with de novo ANLL, and the durations of remission have been short. Less aggressive regimens using agents such as low-dose cytosine arabinoside or 13-cis-retinoic acid have been similarly unsuccessful in most patients. Exploration of the role of biologic response modifiers such as granulocyte-macrophage colony-stimulating factor (GM-CSF) and α-interferon (INF-α) in the therapy of MDS has recently begun.

Allogeneic bone marrow transplantation (BMT) is effective in the therapy of some malignant and nonmalignant hematologic disorders. Two previously reported series of patients undergoing BMT for myelodysplasia support its efficacy in this patient population. We describe the outcome of BMT in pediatric patients with myelodysplasia, both de novo and secondary.

MATERIALS AND METHODS

Eight consecutive patients with myelodysplasia and an HLA-identical, mixed leukocyte culture (MLC)-nonreactive donor were treated with allogeneic BMT at the Dana Farber Cancer Institute and The Children’s Hospital between October 1984 and August 1987. The diagnosis was established by bone marrow examination demonstrating myelodysplasia with or without a clonal chromosomal abnormality.

Table 1 shows the characteristics of this group of patients. The ratio of males to females was 6:2. The ages at time of transplantation ranged from 3 to 19 years (median 12 years), and duration of disease prior to transplant ranged from 2 to 10 months (median 3.5 months). Two of the patients (UPN 244 and 172) had received prior chemotherapy for metastatic neuroblastoma and retinoblastoma, respectively, and the latter patient had also received radiation therapy. One (UPN 172) developed MDS 31 months after cessation of primary therapy; the other patient (UPN 244) developed MDS after only six months. All eight patients were previously transfused at the time of marrow transplantation. None of the patients had received other therapy directed at their myelodysplasia.

Peripheral blood and bone marrow samples were reviewed and classified according to the categories defined by the French-American-British (FAB) cooperative group (Table 1). Marrow samples were prepared for cytogenetic analysis by rinsing samples twice with unsupplemented medium at 37°C, after which 3 to 5 × 10^6 cells were inoculated into culture bottles containing RPMI 1640 with 16.0% fetal bovine serum (FBS), L-glutamine, and antibiotics. Cultures were incubated for ~24 hours at 37°C in an incubator with 5% CO₂ atmosphere and humidity. Mitotic cells were blocked with colcemid at a final concentration of 0.01 μg/mL. Standard air-dried slides were stained with an aqueous solution of quinacrine mustard (50 μg/mL) and mounted in pH 5.6 buffer. The preparations were examined under a fluorescence microscope equipped with a camera. Suitable metaphases were photographed. Analysis was done on photographic prints.

Histocompatibility between donor and recipient was determined by HLA-typing and MLC techniques. All donors were HLA-identical, MLC-nonreactive siblings. There were five sex-matched recipient-donor pairs and three sex-mismatched pairs. The conditioning regimen in all patients consisted of high-dose cyclophosphamide (1,800 mg/m²/day bolus infusion over one hour, days −4 and −3 or −3 and −2) and concurrent fractionated total body irradiation (TBI). Owing to participation in other ongoing studies, three patients also received cytosine arabinoside (500 mg/M²/day continuous infusion for seven days) before initiation of cyclophosphamide and TBI. TBI was given on a dedicated facility with a 4-MeV linear accelerator. All patients were treated anteroposterior/poste...
pulmonary function tests.

slowly over 6 months); the eighth patient received a T-cell-depleted
starting day -1.5 mg/kg IV or 6 mg/kg orally (PO) every 12 hours
cyclosporine either methotrexate [10 mg/M² intravenously (IV) days I, 3, 6, and
12] or procarbazine 5.100 mg/M² qd and antilymphocyte serum 0.2 mL/kg orally (PO) every 12 hours
starting day –1 and continued through day 50 and then tapered
slowly over 6 months); the eighth patient received a T-cell-depleted
marrow (purged with anti-Leu1 and complement) without addi-
tional postgrafting immunosuppression (Table 2). The transplants
were performed in laminar airflow rooms; patients were fed sterile or
low-bacteria diets and were treated with oral nonabsorbable antibi-
otics and skin cleansing. The marrow transplant protocols and
consent forms were approved by the Human Subjects Review Board
of the Dana Farber Cancer Institute and The Children’s Hospital.

Prophylaxis for acute graft-v-host disease (GVHD) was assigned
according to protocols in effect at the time that patients came to
transplantation. Seven patients received GVHD prophylaxis with
either methotrexate [10 mg/M² intravenously (IV) days 1, 3, 6, and
11] or methotrexate plus cyclosporine (methotrexate as above with
cyclosporine 1.5 mg/kg IV or 6 mg/kg orally (PO) every 12 hours
starting day –1 and continued through day 50 and then tapered
slowly over 6 months); the eighth patient received a T-cell-depleted
marrow (purged with anti-Leu1 and complement) without addi-
tional postgrafting immunosuppression (Table 2). The transplants
were performed in laminar airflow rooms; patients were fed sterile or
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otics and skin cleansing. The marrow transplant protocols and
consent forms were approved by the Human Subjects Review Board
of the Dana Farber Cancer Institute and The Children’s Hospital.

RESULTS

The outcome of the eight patients is summarized in Table 2.

Engraftment. The seven patients receiving non-T-cell-depleted
marrow all engrafted promptly. Return of the absolute neutrophil count to 500/μL occurred 14 to 28 days
after transplantation (median 23 days). The one patient to
receive a T-cell-depleted graft had an initial increase in
neutrophil count to 500/μL which then decreased by day
21. She remained pancytopenic with an aplastic marrow, and
no dividing cells were available on two evaluations to provide
evidence of donor engraftment. She was retransplanted on
day 40 after preparation with procarbazine 12.5 mg/kg
every other day for three doses and both cyclophosphamide
1,200 mg/M² qd and antilymphocyte serum 0.2 mL/kg
every day for three days. The bone marrow was not T-cell
depleted. The patient subsequently developed a clinical syn-

<table>
<thead>
<tr>
<th>UPN</th>
<th>Age (yr)</th>
<th>Sex</th>
<th>Previous Therapy</th>
<th>Disease Duration (mo)</th>
<th>Marrow Fibrosis</th>
<th>Karyotype</th>
<th>FAB Classification</th>
</tr>
</thead>
<tbody>
<tr>
<td>172</td>
<td>6</td>
<td>M</td>
<td>HN₂, ADR, DTIC, CTX</td>
<td>4</td>
<td>None</td>
<td>45, XY, -7/46, XY, -7</td>
<td>RAEBIT</td>
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<tr>
<td>188</td>
<td>12</td>
<td>F</td>
<td>None</td>
<td>10</td>
<td>Increased reticulin</td>
<td>46, XX/45, XX, -7</td>
<td>RAEB</td>
</tr>
<tr>
<td>200</td>
<td>13</td>
<td>M</td>
<td>None</td>
<td>2</td>
<td>None</td>
<td>45, XY</td>
<td>RAEBIT</td>
</tr>
<tr>
<td>226</td>
<td>19</td>
<td>F</td>
<td>None</td>
<td>10</td>
<td>Myelofibrosis</td>
<td>45, XX, -7</td>
<td>RAEBIT</td>
</tr>
<tr>
<td>244</td>
<td>3</td>
<td>M</td>
<td>VCR, L-PAM, DTIC, CTX</td>
<td>9</td>
<td>None</td>
<td>46, XY/45, XY, -7</td>
<td>RA</td>
</tr>
<tr>
<td>252</td>
<td>11</td>
<td>M</td>
<td>None</td>
<td>2</td>
<td>None</td>
<td>46, XY/45, XY, -7</td>
<td>RAEBIT</td>
</tr>
<tr>
<td>261</td>
<td>15</td>
<td>M</td>
<td>None</td>
<td>3</td>
<td>Increased reticulin</td>
<td>46, XY/45, XY, -7</td>
<td>RAEBIT</td>
</tr>
<tr>
<td>294</td>
<td>18</td>
<td>M</td>
<td>None</td>
<td>2</td>
<td>Increased reticulin</td>
<td>46, XY/45, XY, -7</td>
<td>RAEB</td>
</tr>
</tbody>
</table>

Abbreviations: UPN, unique patient number; VCR, vincristine, L-PAM, melphalan; DTIC, dacarbazine; CTX, cyclophosphamide; HN₂, nitrogen mustard; ADR, adriamycin (doxorubicin); C-DDP, cis-platinum; RA, refractory anemia, RAEB, refractory anemia with excess blasts; RAEBIT, refractory anemia with excess blasts in transition.

Table 2. Patient Conditioning and Outcome

<table>
<thead>
<tr>
<th>UPN</th>
<th>Chemotherapy</th>
<th>TBI*</th>
<th>GVHD Prophylaxis</th>
<th>AGVH</th>
<th>CGVH</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>172</td>
<td>CTX 1,800 mg/M² × 2</td>
<td>1,200 cGy</td>
<td>MTX</td>
<td>GR I: skin</td>
<td>None</td>
<td>Alive and well at 44 mo</td>
</tr>
<tr>
<td>188</td>
<td>CTX 1,800 mg/M² × 2; ARA-C 500 mg/M² × 7</td>
<td>1,200 cGy</td>
<td>MTX</td>
<td>GR I: skin, liver</td>
<td>Severe: skin, liver, GI</td>
<td>Died at 19 mo with severe CGVHD</td>
</tr>
<tr>
<td>200</td>
<td>CTX 1,800 mg/M² × 2; ARA-C 500 mg/M² × 7</td>
<td>1,296 cGy</td>
<td>MTX</td>
<td>GR IV: skin, liver, GI</td>
<td>NA</td>
<td>Died at day 63 with severe AGVHD</td>
</tr>
<tr>
<td>226</td>
<td>CTX 1,800 mg/M² × 2; ARA-C 500 mg/M² × 7</td>
<td>1,200 cGy</td>
<td>T-Cell depletion of marrow</td>
<td>NA</td>
<td>NA</td>
<td>Died at day 58 aplastic with venoocclusive liver disease and infection</td>
</tr>
<tr>
<td>244</td>
<td>CTX 1,800 mg/M² × 2</td>
<td>1,400 cGy</td>
<td>MTX</td>
<td>GR I: skin</td>
<td>None</td>
<td>Died at 21 months of progressive lung disease</td>
</tr>
<tr>
<td>252</td>
<td>CTX 1,800 mg/M² × 2</td>
<td>1,400 cGy</td>
<td>MTX</td>
<td>GR I: skin</td>
<td>None</td>
<td>Alive and well at 20 mo</td>
</tr>
<tr>
<td>261</td>
<td>CTX 1,800 mg/M² × 2</td>
<td>1,400 cGy</td>
<td>MTX, cyclophosphamide</td>
<td>GR I: skin</td>
<td>None</td>
<td>Alive and well at 17 mo</td>
</tr>
<tr>
<td>294</td>
<td>CTX 1,800 mg/M² × 2</td>
<td>1,400 cGy</td>
<td>MTX, cyclophosphamide</td>
<td>GR II: skin, liver</td>
<td>Moderate: skin, GI</td>
<td>Alive with abnormal PFTS, clinical lung disease, resolving CGVHD at 10 mo</td>
</tr>
</tbody>
</table>

Abbreviations: CTX, cyclophosphamide; ARA-C, cytosine arabinoside; MTX, methotrexate; AGVHD, acute GVHD; CGVHD, chronic GVHD; PFTS, pulmonary function tests.

*1,400 cGy = 175 cGy x eight doses; 1,296 cGy = 216 x six doses; 1,200 cGy = 200 x six doses.
drome consistent with severe venocclusive disease of the liver; she also had *Candida tropicalis* fungemia and died on day 38 (after the first transplant).

Donor engraftment was documented in five of the seven engrafting patients by differences in chromosome composition in cases of cross-sex transplants or informative quinacrine polymorphisms or blood type differences in same-sex transplants. The two remaining patients had no detectable markers by which to document donor engraftment as restriction fragment length polymorphisms (RFLPs) and/or RBC antigenic or enzyme differences were not being routinely studied.

**GVHD.** Patients were evaluated for GVHD by a senior attending physician, and their condition was graded according to previously published clinical and pathologic criteria.23 All seven patients who engrafted developed acute GVHD. In four patients, acute GVHD was confined to the skin with an overall grade of I and did not require treatment. Two patients also developed mild liver function abnormalities (grade II GVHD). One of these (UPN 188) resolved her GVHD spontaneously; the other (UPN 294) resolved his symptoms after a short course of prednisone. Another patient (UPN 200) developed fulminant grade IV GVHD and died at day 63 secondary to complications of this syndrome despite therapy with steroids, antilymphocyte serum, and cyclosporine. Two of the six patients at risk (ie, alive at >100 days) developed chronic GVHD. Sicca syndrome, oral lichen planuslike lesions and weight loss developed in one patient (UPN 294) who subsequently had complete resolution of these symptoms on azathioprine (Imuran, 1 mg/kg/day) and prednisone (1 mg/kg/every other day). Ten months after BMT, this patient has significant obstructive airway disease (forced vital capacity 39% predicted, total lung capacity 126% predicted) in the setting of his otherwise well-controlled chronic GVHD. A second patient (UPN 188) developed scleroderma skin, contractures, and an autoimmunelike clinical picture characterized by a protein-losing nephropathy, pleural and pericardial effusions, and ascites. She eventually developed progressive symptoms despite treatment with azathioprine, corticosteroids, and cyclosporine and died 19 months posttransplant.

**Disease-free survival.** At the time of evaluation (June 20, 1988), four patients were alive and all were hematologically normal with at least one bone marrow aspirate post transpl ant that demonstrated correction of their underlying dysplasia. Cytogenetic studies in patients who had had a clonal abnormality demonstrated absence of that clone after BMT. Two other patients (UPN 188 and 244), who died at 19 and 21 months, respectively, were also disease-free at the time of their death.

**Toxicity.** All patients tolerated their conditioning regimens without undue acute toxicity. Patients who received cytosine arabinoside by constant infusion had more skin erythema and breakdown as well as more diarrhea and liver function abnormalities. Two patients (UPN 226 and 200) died within the first 100 days posttransplant (from complications of failure to engraft and acute GVHD, respectively). A third patient (UPN 188) died 19 months posttransplant secondary to complications of chronic GVHD. A fourth patient (UPN 244) developed dyspnea, rales, and hypoxemia 14 months after BMT. He had no evidence of chronic GVHD. Lung biopsy obtained at that time demonstrated severe pulmonary fibrosis of unclear etiology. His pathology was not believed to be consistent with bronchiolitis obliterans. A prolonged course of corticosteroids (at 1 to 2 mg/kg/day) appeared to arrest but not reverse the course of his lung disease. Despite continued steroid therapy, he died 21 months posttransplant of progressive respiratory failure. Of the remaining four patients, three (UPN 261, 252, and 172) are complication-free (at 17, 20, and 44 months posttransplant). The remaining patient (UPN 294) developed idiopathic interstitial pneumonitis at day 100 and has significant obstructive lung disease 10 months posttransplant in the setting of treated chronic GVHD (described above).

**DISCUSSION**

Several investigators have reported their experience in treating pediatric patients with myelodysplasia.3,8,12 The likelihood of such patients achieving CR with aggressive combination chemotherapy regimens is low.24,9,12 and the number of patients sustaining CR for >2 years is even more limited. Recent reviews of less aggressive regimens (generally in adult patients) suggest that sustained CR rates with low-dose cytosine arabinoside or *cis*-retinoic acid are also low.13-15 and the safety and efficacy of the biological response modifiers in treatment of MDS16-19 remain to be established. Although the data regarding the utility of such approaches in a pediatric population is limited, the available literature5,9,12 as well as our own results (H.W. Weinstein, unpublished observations) suggest that these therapies are of minimal benefit in younger patients. In contrast, the disease-free survival of four of the eight patients we report extends the previously reported positive experience with allogeneic BMT, which has yielded disease-free survivals of 40% and 43% in two other published series.4,21

Reported adverse prognostic features for patients with MDS undergoing BMT have included narrow fibrosis, increased marrow blasts, and secondary MDS.5,21 In our series of eight patients, the one patient with myelofibrosis did fail to engraft and subsequently died. However, this patient also received a T-cell-depleted graft, also a risk factor associated with increased graft rejection or failure of engraftment.24-26 The bone marrow biopsies of three other patients showed increased reticulin (Table 1), although the patients were not judged to have myelofibrosis; they all engrafted. The number of patients in any FAB classification was too small to allow us to assess the importance of histology as a prognostic feature, although none of our patients has had disease recurrence. The two patients (UPN 172 and 244) with secondary myelodysplasia both engrafted promptly. One patient remains disease-free at 44 months posttransplant, and the other died at 21 months of pulmonary fibrosis but without evidence of relapse.

In summary, eight children with de novo or secondary myelodysplasia underwent HLA-matched, MLC-compatible BMT. Four of eight patients are disease-free survivors with a median follow-up of 19 months. We were unable to
identify prognostic factors in this limited series. Based on this experience, we recommend BMT as initial therapy for pediatric patients with either de novo or secondary myelodysplasia. In the absence of ex vivo T-cell depletion, our experience suggests that cyclophosphamide and TBI (1,200 to 1,400 cGy) are sufficient to ensure adequate ablation and immunosuppression as well as long-term disease-free survival. Preliminary data suggest that BMT using partially mismatched or unrelated matched marrow may prove a feasible therapeutic alternative for patients with this diagnosis who lack a matched donor.27

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REFERENCES

Bone marrow transplantation for children with myelodysplastic syndromes

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