Stem cell transplantation in patients with severe congenital neutropenia without evidence of leukemic transformation


Severe congenital neutropenia (CN) (Kostmann syndrome) is a hematologic disorder characterized by a maturation arrest of myelopoiesis at the promyelocyte/myelocyte stage of development. This arrest results in severe neutropenia leading to absolute neutrophil counts (ANC) below 0.2 × 10^9/L associated with severe bacterial infections from early infancy. Data on over 300 patients with CN collected by the Severe Chronic Neutropenia International Registry (SCNIR) beginning in 1994 indicate that more than 90% of these patients respond to recombinant human granulocyte-colony stimulating factor (r-HuG-CSF) treatment with an ANC greater than 1.0 × 10^9/L. For patients who are refractory to r-HuG-CSF treatment and continue to have severe and often life-threatening bacterial infections, hematopoietic stem cell transplantation is the only currently available treatment. We report on a total of 11 patients with CN who underwent HSCT for reasons other than malignant transformation between 1976 and 1998. Of these patients, 8 were nonresponders or showed only partial response to r-HuG-CSF treatment with ongoing infections. Results from these patients suggest that transplantation of stem cells from an HLA-identical sibling is beneficial for patients refractory to r-HuG-CSF. (Blood. 2000;95:1195-1198)

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Introduction

Kostmann described an inherited hematologic disorder with severe neutropenia with an absolute neutrophil count (ANC) below 0.2 × 10^9/L and early onset of severe bacterial infections.1,2 Most children died because of these infections despite antibiotic treatment. Different treatment strategies for congenital neutropenia (CN) included use of steroids and lithium,3,4 but these treatments did not show any long-term effect on neutrophil counts. Bone marrow transplantation (BMT) was the only curative treatment option for patients with HLA-compatible donors.5 Some patients who survived infections and treatment, however, underwent malignant transformation into acute myeloid leukemia (AML).6,7

The availability of recombinant human granulocyte colony-stimulating factor (r-HuG-CSF) in 19878,9 dramatically changed both the prognosis of CN and the quality of life of patients with CN.10,11 Since the establishment of the Severe Chronic Neutropenia International Registry (SCNIR) in 1994, data on 304 patients with CN have been collected. In clinical trials, more than 90% of these patients responded to r-HuG-CSF treatment with an increase in ANC greater than 1.0 × 10^9/L. Importantly, all responding patients required significantly fewer antibiotics and days of hospitalization.12-17 Hematopoietic stem cell transplantation (HSCT) remains the only currently available treatment for those patients refractory to r-HuG-CSF treatment who continue to have severe and life-threatening bacterial infections. HSCT performed after the development of overt myelodysplastic syndromes (MDS)/AML is associated with a high mortality rate (70% in the SCNIR) (data from the SCNIR annual report, 1998). Data from the SCNIR also demonstrate that for all CN patients, approximately 10% will develop leukemia regardless of their treatment or response.13,15,16 For these reasons, there is an increased interest in the outcome of HSCT in this patient population. We report on 11 patients with CN who underwent HSCT for reasons other than malignant transformation between 1976 and 1998.

Patients and methods

Patients

Patient data were collected from 10 European countries, Israel, Australia, Canada, and the United States. All patients or their legal guardians have given written consent for data collection and referral to the SCNIR. Treatment was performed following the generally accepted guidelines for cytokine administration, but each referring physician treated his/her individual patient as medically indicated.

As of August 1, 1998, SCNIR has registered 304 patients with CN. Of these 304 patients, 29 have been reported to have undergone HSCT. Eighteen of these patients underwent HSCT because of malignant transformation. We now report on 11 patients who underwent HSCT for reasons other than malignant transformation. The indication for transplantation for these 11 patients was primarily the lack of or partial response to r-HuG-CSF (8 patients). Nonresponders to r-HuG-CSF were defined as those patients...
showing no or an insufficient response to r-HuG-CSF, even at doses as great as 120 µg/kg/d. Partial responders increased their ANC to 0.5 to 1.0 × 10⁹/L, but still had bacterial infections. The dose of r-HuG-CSF could not be increased in these patients because of the large volume and frequency of injections required.

Of the other patients, the indications for transplantation included neutropenia before the availability of r-HuG-CSF (n = 1), pancytopenia and a G-CSF–receptor mutation (n = 1), and a G-CSF–receptor mutation without further signs of myelodysplasia or cytogenetic aberrations (n = 1). The last patient received HSCT because this mutation is thought to be one step in the pathway to development of leukemia.18-21

### Pretransplant conditioning regimens

The conditioning regimens are summarized in Table 1. One patient received immunosuppressive therapy with cyclophosphamide alone (200 mg/kg). Ten patients received combination chemotherapy that included busulfan (8 to 24.4 mg/kg, median 16 mg/kg, usually divided into 16 doses) and cyclophosphamide (120 to 200 mg/kg). In addition to the combination of busulfan and cyclophosphamide, additional agents used in the conditioning included antithymocyte globulin in 3 patients, thiotepa 6 to 10 mg/kg in 2 patients, and melphalan 140 mg/m² in 1 patient.

### Stem cell source

Most patients received cells from an HLA-identical sibling, either bone marrow (n = 5), peripheral blood progenitor cells (PBPCs) (n = 1), cord blood (n = 1), or bone marrow and cord blood (n = 1). The remaining 3 patients received either single-antigen–mismatched paternal donor bone marrow, single-antigen–mismatched unrelated donor marrow, or antigen-mismatched paternal donor PBPCs.

### Table 1. Case reports and current status of all patients

<table>
<thead>
<tr>
<th>Patient No.</th>
<th>Reason for Transplant</th>
<th>Stem Cell Source</th>
<th>Donor</th>
<th>Conditioning</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Neutropenia</td>
<td>Bone marrow</td>
<td>HLA-identical sibling</td>
<td>CY</td>
<td>Graft rejected; autologous engraftment with recurring neutropenia; subsequent availability of r-HuG-CSF has allowed complete response.</td>
</tr>
<tr>
<td>2</td>
<td>Nonresponder</td>
<td>Bone marrow</td>
<td>HLA-identical sibling</td>
<td>BU/CY</td>
<td>GVHD grade I; no problems at time of report.</td>
</tr>
<tr>
<td>3</td>
<td>Nonresponder</td>
<td>Bone marrow</td>
<td>HLA-identical sibling</td>
<td>BU/CY + ATG</td>
<td>Fungal infection pre-BMT; treated successfully.</td>
</tr>
<tr>
<td>4</td>
<td>Nonresponder</td>
<td>PBPCs</td>
<td>HLA-identical sibling</td>
<td>BU/CY</td>
<td>Alive and well.</td>
</tr>
<tr>
<td>5</td>
<td>Nonresponder</td>
<td>Bone marrow</td>
<td>Single-antigen–mismatched unrelated</td>
<td>BU/CY</td>
<td>Acute GVHD grade IV of intestines, resolved; chronic GVHD skin; failure to thrive syndrome.</td>
</tr>
<tr>
<td>6</td>
<td>Pancytopenia</td>
<td>Bone marrow</td>
<td>Single-antigen–mismatched father</td>
<td>BU/CY</td>
<td>Died on day +111 of grade IV GVHD and septicemia.</td>
</tr>
<tr>
<td>7</td>
<td>Nonresponder</td>
<td>PBPCs</td>
<td>Haploidentical father</td>
<td>BU/CY + ATG + Thiotepa</td>
<td>Died on day +70 of multorgan failure.</td>
</tr>
<tr>
<td>8</td>
<td>Nonresponder</td>
<td>Bone marrow, cord blood</td>
<td>HLA-identical sibling</td>
<td>BU/CY + ATG</td>
<td>Cystectomy due to hemorrhagic cystitis; presently alive and well.</td>
</tr>
<tr>
<td>9</td>
<td>Partial response; recurrent infections</td>
<td>Bone marrow</td>
<td>HLA-identical sibling</td>
<td>BU/CY + ATG</td>
<td>Persistent quadriparesis; neurological symptoms were present before BMT.</td>
</tr>
<tr>
<td>10</td>
<td>G-CSF receptor mutation</td>
<td>Bone marrow</td>
<td>HLA-identical sibling</td>
<td>BU/CY + melphalan</td>
<td>Acute GVHD grade III of skin and intestines; chronic GVHD of skin; fungal pneumonia developed after treatment with steroids; stable hematopoiesis.</td>
</tr>
<tr>
<td>11</td>
<td>Partial response; thoracic granulomatous infection</td>
<td>Cord blood</td>
<td>HLA-identical sibling</td>
<td>BU/CY + Thiotepa</td>
<td>Complete engraftment; no problems reported.</td>
</tr>
</tbody>
</table>

CY indicates cyclophosphamide; BU, busulfan; ATG, antithymocyte globulin; r-HuG-CSF, recombinant human granulocyte colony-stimulating factor; GVHD graft-versus-host disease; BMT, bone marrow transplantation; PBPCs, peripheral blood progenitor cells.

### Results

There were 6 boys and 5 girls. Their age at time of BMT ranged from 0.6 to 15.9 years. At the time of this report, post-BMT follow-up has been 0.9 to 22.5 years (median follow-up, 1.9 years), and 9 of the 11 patients are alive. The indications for transplantation, stem cell source, clinical problems and other information for all 11 patients are summarized in Table 1.

All patients who are alive are currently independent of erythrocyte and platelet transfusions. Only 1 patient has required continued treatment with r-HuG-CSF. This patient had initially rejected the transplant and subsequently had autologous recovery with continuing neutropenia that responds well to r-HuG-CSF.

One patient received a nonmyeloablative conditioning regimen (cyclophosphamide alone) and rejected the graft. The other 10 patients who received myeloablative regimens with busulfan and cyclophosphamide engrafted, regardless of whether the donor was HLA-identical or not.

All 8 patients who received HSCT from a sibling donor are alive. As already noted, 1 of these patients rejected the graft following a nonmyeloablative preparative regimen. Median follow-up for the 7 patients transplanted with a myeloablative regimen is 10 months, with a range of 2 months to 6 years. Complications following sibling donor transplant included hemorrhagic cystitis necessitating cystectomy in 1 patient and severe graft-versus-host disease (GVHD) in another patient. The other 5
patients were cured of their CN without significant complications. Persistence of preexisting mild quadriparesis following HSCT is noted in 1 patient.

Alternative donor transplants were used for 3 patients. In 1 case, a single-antigen–mismatched unrelated donor was the source of stem cells, and in the other 2 cases, a mismatched paternal transplant was performed. All 3 patients engrafted with donor cells and achieved normal neutrophil counts without requiring G-CSF. All 3 patients receiving HLA-mismatched stem cells suffered serious complications following transplant, including grade II-IV GVHD,3 extensive chronic GVHD,1 and fungal infection.1 One patient died from complications related to acute GVHD, and another patient died from multiorgan failure. One patient remains alive, with normal neutrophil counts, but experiences failure to thrive from extensive chronic GVHD.

Bone marrow chimerism was tested in 5 patients. The percent-
ages of donor cells were 30% at 2 years in 1 patient, 84% at 6 months in another patient, and 100% at 6 weeks, 8 weeks, and 1 year in the other 3 patients. Both patients who lacked 100% donor cells nevertheless maintained normal neutrophil counts and were infection free. Two patients had posttransplant marrow aspirates that demonstrated a normal morphological appearance and none of the features associated with CN.

Following engraftment, infections were uncommon; 2 patients developed fungal infections while on steroids for GVHD.

Discussion

We report on 11 patients with CN who underwent transplantation for reasons other than malignant transformation. Data collected from the SCNIR on more than 300 patients diagnosed with CN indicate that nonresponse to r-HuG-CSF treatment is rare, occurring in fewer than 10% of these patients. However, for those patients who do not respond to r-HuG-CSF treatment, alone or in combination with other cytokines such as stem cell factor,22 transplantation is clearly the only currently available treatment. When transplantation is successful, patients who receive it are cured from the defective hematopoiesis and do not need further cytokine treatment.

All CN patients, regardless of their treatment or response, are at risk of developing MDS or leukemia at an incidence of 10% (unpublished data from the SCNIR). Outcome of transplantation after the development of frank leukemia is poor. In our study, only 3 of the 18 patients who underwent transplantation as a treatment for leukemia have survived (unpublished data from the SCNIR). A mutation within a critical region of the intracellular part of the G-CSF receptor seems to play an important role in the pathway toward leukemia.20 A mutation has been detected in each CN patient tested (n = 8) who developed leukemia. However, this mutation has also been detected in a number of CN patients whose condition has not yet transformed into MDS or leukemia.19,20,23

Of the 11 patients reported here, 8 patients were nonresponders or showed only minor response to r-HuG-CSF treatment. Their ANC remained about 0.5 × 10⁹/L, and they continued to have infectious episodes. One patient was transplanted in 1976 when no cytokine treatment was available; 1 patient developed panmye-
penia; and another patient who responded well to r-HuG-CSF treatment developed a mutation of the G-CSF receptor without further signs of myelodysplasia or cytogenetic aberrations. Because this mutation seems to play an important role, as already mentioned, and because an HLA-identical sibling was available, transplantation was discussed with the parents and subsequently performed.

Our data document that the outcome of stem cell transplantation in patients with CN whose condition has not transformed into MDS or leukemia is better than that of transplantation in patients who develop leukemia. One reason for the superior outcome in the group of patients reported here compared with those patients with CN and leukemia who undergo transplantation may be the higher proportion that underwent HSCT from an HLA-identical sibling. In this report, 8 of the 11 patients had a sibling donor compared with only 1 of 18 patient transplanted for leukemia (unpublished data from the SCNIR). Secondly, patients transplanted for treatment of leukemia have already received intensive chemotherapy, which might increase the risk of treatment-related mortality during the transplant. Furthermore, other studies have shown that survival in patients who develop MDS or leukemia arising in the context of other inherited predispositions to leukemia is poor.24-26

For those patients with CN in whom transplant is contemplated, a marrow ablative regimen of busulfan and cyclophosphamide appears effective. The only patient who rejected the graft had received cyclophosphamide alone as preparative treatment before transplantation. In two patients, mixed chimerism developed, but in both cases the children have normal neutrophil counts and remain free from infection. We do not know if children with mixed chimerism will still be at risk of developing leukemia. These children continue to be observed to see if myelodysplasia or leukemia will develop.

It remains difficult to give a recommendation for transplantation for patients with CN who respond well to r-HuG-CSF and show no evidence of impending malignant transformation. Of the 11 patients reported here, 2 (18%) died after transplantation. Both patients were transplanted with stem cells from alternative donors (1 from a single-antigen–mismatched and 1 from an antigen-mismatched father). The 8 patients who underwent transplantation from an HLA-identical sibling survived. However, 1 patient who initially engrafted later developed autologous reconstitution with a subsequent G-CSF–responsive neutropenia. Two other patients had serious posttransplant complications from either GVHD or hemorrhagic cystitis. The risks associated with transplant from an HLA-identical sibling may outweigh the risk of leukemic transformation when r-HuG-CSF is continued in those patients who respond well to therapy. If the risks of HSCT can be decreased, perhaps by employing a low-intensity regimen and inducing tolerance by mixed chimerism, then HSCT from a sibling donor may find a greater role in the treatment of CN, even in patients responsive to r-HuG-CSF.

We conclude that stem cell transplantation from an HLA-
identical sibling is beneficial for CN patients refractory to r-HuG-CSF. For those patients in whom a G-CSF receptor mutation is identified, stem cell transplantation from an HLA-identical sibling is an option. Data on alternative sources of donor stem cells are insufficient to assess outcome in patients with congenital neutropenia. The finding of resolution of neutropenia and freedom from infection in the 2 patients in whom mixed chimerism developed is intriguing and suggests that alternative approaches to HSCT may be useful in the treatment of this disease. In this report, 2 of 3 patients who had alternative-source stem cell transplants died. However, new strategies for transplantation, such as highly purified CD 34+ peripheral stem cells, are promising. In the meantime, in light of the results of studies, we suggest that the use of r-HuG-CSF remain first-line treatment for most patients.
Acknowledgments

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References

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