Hematopoietic Cell Transplantation Using Reduced-Intensity Conditioning Is Successful in Children with Hematologic Cytopenias of Genetic Origin

Alok Kothari [1], Alexander Ngwube [1], Robert Hayashi [1], Lisa Murray [1], Jeffrey Davis [2], Paul Haut [3], Brett J. Loechelt [4], Shalini Shenoy [1,*]

[1] Department of Pediatrics, Washington University, St. Louis, Missouri
[2] Department of Pediatrics, Children's and Women's Health Centre of BC, Vancouver, BC, Canada
[3] Department of Pediatrics, Indiana University School of Medicine, Indianapolis, Indiana
[4] Department of Pediatrics, Children's National Medical Center, Washington, D.C.

Article history:
Received 7 February 2015
Accepted 21 March 2015

Key Words:
Congenital hematologic cytopenias
Reduced-intensity conditioning
Hematopoietic cell transplantation

A B S T R A C T

Genetically derived hematologic cytopenias are a rare heterogeneous group of disorders. Allogeneic hematopoietic cell transplantation (HCT) is curative but offset by organ toxicities from the preparative regimen, graft rejection, graft-versus-host disease (GVHD), or mortality. Because of these possibilities, consideration of HCT can be delayed, especially in the unrelated donor setting. We report a prospective multicenter trial of reduced-intensity conditioning (RIC) with alemtuzumab, fludarabine, and melphalan and HCT in 11 children with marrow failure of genetic origin (excluding Fanconi anemia) using the best available donor source (82% from unrelated donors). The median age at transplantation was 23 months (range, 2 months to 14 years). The median times to neutrophil ($>500 \times 10^6$/L) and platelet ($>50 \times 10^9$/L) engraftment were 13 (range, 12 to 24) and 30 (range, 7 to 55) days, respectively. The day +100 probability of grade II to IV acute GVHD and the 1-year probability of limited and extensive GVHD were 9% and 27%, respectively. The probability of 5-year overall and event-free survival was 82%; 9 patients were alive with normal blood counts at last follow-up and all were successfully off systemic immunosuppression. In patients with genetically derived severe hematologic cytopenias, allogeneic HCT with this RIC regimen was successful in achieving a cure. This experience supports consideration of HCT early in such patients even in the absence of suitable related donors.

INTRODUCTION

Hematologic cytopenias due to bone marrow dysfunction (BMD) of genetic etiology include a heterogeneous group of disorders where genetic mutations result in abnormal or arrested hematopoiesis affecting 1 or more cell lines [1-3]. Conservative management of a lifelong defect in hematopoiesis is challenging and includes chronic transfusion therapy with inherent risks of infection, iron overload, anemia, bleeding, alloimmunization, susceptibility to life-threatening infections, and in many BMD, a predisposition to malignant transformation, usually acute myeloid leukemia [4-8].

Allogeneic hematopoietic cell transplantation (HCT) can cure the hematopoietic defects associated with BMD. The heterogeneity of BMD and associated organ involvement in some disorders present unique obstacles to successful HCT. In addition, the challenges of allogeneic transplantation, including identification of a suitable donor and optimization of preparative therapy to reduce regimen-related toxicity while ensuring successful engraftment, are important to overcome. Low rates of mortality, graft-versus-host disease (GVHD), and untoward late effects from HCT will promote consideration of transplantation earlier for BMD [9]. Safe and effective transplantation methods are a key to promoting long-term survival and maintaining quality of life after HCT at a young age before the increasing risks associated with conservative management of the marrow failure. Many BMD are associated with defective DNA repair or maintenance pathways and possess increased sensitivity to DNA-damaging agents, including chemotherapy and radiation. These sensitivities have resulted in increased HCT-related morbidity and mortality in BMD patients [2,9]. Further, the presence of genetic predisposition to malignant transformation requires development of transplantation approaches that minimize additional risks of late malignancies, which makes reduced-intensity conditioning (RIC) regimen an appealing choice.

Trials of HCT after RIC have recently targeted several groups of recipients for reasons such as age and pre-existing organ toxicity. Although RIC regimens are perhaps better tolerated, barriers to using RIC regimens and proceeding with HCT include risk of GVHD from unrelated donor grafts and potentially increased rates of graft rejection, especially in the immune competent and those exposed to multiple blood products [10].

This study describes the outcomes of a prospective, pediatric, multicenter, phase II HCT trial for children with BMD who had undergone RIC to achieve significant host immunosuppression. The primary objective was to determine donor engraftment, overall survival (OS), and event-free survival (EFS) after conditioning with alemtuzumab, fludarabine, and melphalan and HCT from the best available donor in patients with BMD and severe hematologic cytopenia. Fanconi anemia patients were excluded because of the use of the alkylating agent melphalan in the regimen and the unknown toxicity imparted by this drug on affected patients. Secondary objectives were to evaluate for

Financial disclosure: See Acknowledgments on page 1325.

* Correspondence and reprint requests: Shalini Shenoy, MD, Box 8116, Washington University School of Medicine, 660 S. Euclid Ave., St. Louis, MO 63110.
E-mail address: shenoy@wustl.edu (S. Shenoy).

1083-8791/
http://dx.doi.org/10.1016/j.bbmt.2015.03.019

immune reconstitution, infection, and transplantation-related toxicities.

METHODS

Patient and Donor Selection

BMD patients were enrolled at 4 transplantation centers. The protocol was approved by the institutional review board at each of the participating institutions (NCT00920972). Consent was obtained from legal guardians of all patients before enrollment and assents were obtained when appropriate. Eligibility included patients younger than 21 years of age with a diagnosis of a BMD other than Fanconi anemia. Patients with human immunodeficiency virus seropositivity, Lansky performance score ≤ 50, or uncontrolled active bacterial, viral, or fungal infections were ineligible.

Donors were selected based upon the best allele match for HLA-A, HLA-B, HLA-C, and DRB1 by high-resolution typing. In the absence of matched related donor (MRD) marrow, 7 to 8/8 allele/antigen matched unrelated donor (URD) marrow was considered eligible. Umbilical cord blood (UCB) products were required to be matched at a minimum of 4 of 6 HLA loci (low resolution at A and B; high resolution at DRB1) and have a pre-cryopreservation total nucleated cell number of $\geq 4.0 \times 10^7$ per kilogram recipient weight. Marrow products were infused fresh and unmanipulated except as required for ABO incompatibility between donor and recipient.

Conditioning Regimen and GVHD Prophylaxis

All patients received alemtuzumab intravenously (i.v.) daily for 3 days (10 mg, 15 mg, and 20 mg if >10 kg and 10 mg on each day if $\leq$ 10 kg) between day −21 and −19 after a 3 mg test dose per manufacturer recommendations on day −22. The total dose of alemtuzumab was 48 mg in children >10 kg and 33 mg in children $\leq$ 10 kg. Patients were discharged after alemtuzumab administration and readmitted on day −8. Fludarabine (30 mg/m^2 or 1 mg/kg if $\leq$ 10 kg) was administered i.v. daily between days −8 and −4. Melphalan (140 mg/m^2 or 4.7 mg/kg if $\leq$ 10 kg) was administered i.v. on day −3 [11]. GVHD prophylaxis consisted of cyclosporine or tacrolimus and short-course methotrexate (7.5 mg/m^2 on days 1, 3, and 6). Cyclosporine or tacrolimus was started on day −3 with regular monitoring to maintain therapeutic levels, continued to day +100, and tapered gradually to stop by day +180 in the absence of GVHD. In addition, prednisone was administered to all unrelated bone marrow (URD BM) recipients at 1 mg/kg/day from day +7 to +28 and subsequently tapered in the absence of GVHD. UCB recipients received a calcineurin inhibitor and mycophenolate mofetil (MMF) instead. MMF (1 g every 8 hours for children $\geq$ 50 kg or 15 mg/kg every 8 hours for children < 50 kg) was commenced on day -3 and continued through day +45 or for 7 days after engraftment, whichever came later. MMF levels were not monitored.

Supportive Care

All patients received antibiotic prophylaxis with oral ciprofloxacin, itraconazole (until day 100 for BM and day 180 for UCB), acyclovir (until 1 year if herpes simplex virus or varicella zoster virus positive) and trimethoprim-sulfamethoxazole (until 1 year) after engraftment. Patients were monitored weekly for cytomegalovirus (CMV) and Epstein-Barr virus (EBV) DNA until day +100. If CMV was detected, pre-emptive ganciclovir or foscarnet (if before engraftment) therapy was administered until CMV testing was negative × 2. Patients received transfusion of red blood cells if their hemoglobin levels fell below 70 g/L. Platelet transfusions were given for levels below 20×10^9/L. Granulocyte colony–stimulating factor at a dose of 5 μg/kg/day was commenced on day +7 and was continued until the absolute neutrophil count was greater than $.5 \times 10^9$/L for 3 consecutive days.

Endpoints/Statistical Evaluation

Primary endpoints were engraftment, OS, and EFS. *Neutrophil engraftment* was defined as the first of 3 consecutive days with an absolute neutrophil count greater than 500/μL, and *platelet recovery* was defined as the first of 7 consecutive days of a platelet count greater than or equal to 50,000/μL without a transfusion. Donor engraftment was determined by demonstrating chimerism by short tandem repeat analysis in BM and/or peripheral blood. The percentage of donor chimerism was assessed at 1, 3, 6, 9, and 12 months after HCT in the first year, every 6 months in the second year, and yearly thereafter. Formal assessments of immunologic recovery were made on days +100 and +180 and 12 months after HCT. Lymphocyte subpopulations were measured using flow cytometry to calculate absolute lymphocyte count, CD3, CD4, CD8, CD19, and CD16 + 56 cell numbers. Immunoglobulin levels (IgG, IgA, and IgM) were measured at the same intervals.

Statistical analyses were performed in Prism 5.03 (GraphPad, La Jolla, CA). The cutoff date for analysis was April 30, 2014. Continuous variables were summarized as medians and range and categorical variables as percentages. Cumulative incidence of neutrophil and platelet engraftment at 21 days, acute GVHD (aGVHD) at day +100 and chronic GVHD at 1 year were all estimated. OS, EFS, and treatment-related mortality were estimated using Kaplan-Meier estimators and comparisons between groups (MRD and URD) were carried out. *P* values less than .05 were considered significant.

RESULTS

Patient Characteristics

Eleven patients between 2 months and 14 years (median, 23 months) with BMD were enrolled in this study of RIC HCT. Patients, diagnoses, indication for transplantation, transplantation characteristics, and outcomes are summarized in Table 1. Nine were recipients of URD HCT. BM was the source of hematopoietic stem cells in all but 1 patient who received 4 of 6 matched UCB. The median total nucleated and CD34$^+$ cells per kg were 4.1×10^8 (range, $.98 \times 10^8$ to 7.92×10^8) and 4.36×10^6 (range, $.6 \times 10^6$ to 8.82×10^6), respectively.

Table 1
Patient Characteristics and Outcomes

Patient No.	Diagnosis (Indication for Transplantation)	Age at HCT, mo/gender	Follow- up, yr	Donor Source	HLA Match	TNC, per kg ($\times 10^8$)	CD34, per kg ($\times 10^6$)	aGVHD/ cGVHD	Outcome/On or Off IS at Last FU
1	CDA Type 1 (Red cell transfusion dependent)	23/M	10	URD UCB	4/6	.98	.6	0/0	A & W/off
2	DBA (transfusion dependent since 7 weeks of life - unresponsive to steroids)	12/F	8	URD BM	8/8	4.92	2.41	0/0	A & W/off
3	SDS (pancytopenia)	168/F	45 days	URD BM	8/8	3.8	4.2	Gr 4/NE	Died/on
4	Congenital BMD (red cell and platelet transfusion dependent since birth) [31]	2/M	6	MSD BM	8/8	5.8	7.8	0/0	A & W/off
5	CAMT (platelet transfusion dependent)	12/M	5	URD BM	8/8	3.95	4.82	0/0	A & W/off
6	X-linked thrombocytopenia (platelet transfusion dependent from 1 mo. of life)	12/M	7	URD BM	8/8	5.14	8.82	Gr 3/0	A & W/off
7	DBA (transfusion dependent since infancy -unresponsive to steroids)	72/M	6	MSD BM	8/8	3.64	5.2	0/0	A & W/off
8	CAMT (platelet transfusion dependent)	36/M	3.5	URD BM	8/8	3.5	3.31	0/0	A & W/off
9	SCN (severe neutropenia)	156/M	1.5	URD BM	8/8	4.1	3.68	0/0	A & W/off
10	X-linked thrombocytopenia (platelet transfusion dependent)	14/M	1.2	URD BM	8/8	7.92	5.92	0/0	A & W/off
11	DBA (transient response to steroids; recurrence with myelofibrosis at 2.5 years)	34/M	1	URD BM	8/8	7.41	4.36	Gr 4/NE	Dead/NE

TNC indicates total nucleated cells; cGVHD, chronic graft versus host disease; FU, follow-up; IS, immunosuppression; CDA, congenital dyserythropoeitic anemia; M, male; A & W, alive and well; SDS, Shwachman Diamond syndrome; F, female; Gr, grade; NE, not evaluable; CAMT, congenital amegakaryocytic thrombocytopenia.

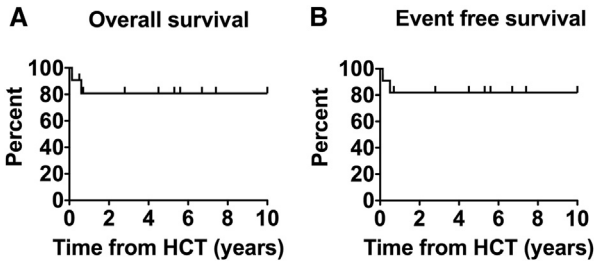

Figure 1. Probability of OS and EFS. (A) Five-year overall survival: 82% (95% CI, 52.3% to 94.9%) and (B) 5-year event-free survival: 82% (95% CI, 52.3% to 94.9%) after hematopoietic cell transplantation for inherited bone marrow failure syndromes. Events were defined as graft rejection, persistent hematologic cytopenias, or death. Dots depict a censored event.

Primary Outcomes

At a median follow-up of 62 months (range, 12 to 186 months), 9 of the 11 recipients had stably engrafted donor cells and had normal blood counts. The probability of EFS and OS at 5 years was 82% (Figure 1A,B). The cumulative incidence of neutrophil recovery was 91% on day 21 and the median time to neutrophil engraftment was 13 days (range, 12 to 24 days). The cumulative incidence of platelet recovery ($>50 \times 10^9$/L) was 73% on day 30 and the median time to recovery was 19 days (range, 7 to 55 days). The recipient who succumbed to aGVHD did not achieve platelet recovery. Each patient had >95% donor chimerism by short tandem repeat analysis in either BM cells or T lymphoid (CD3) and myeloid (CD15) compartments of peripheral blood each time, except for patient 11 (Table 1), who had graft rejection on day 180.

Acute and Chronic GVHD

At 100 days after HCT, the cumulative incidence of grade II to IV aGVHD was 9%.

Two patients with aGVHD died after additional complications as described below. At 180 days after HCT, the cumulative incidence of GVHD was 27%. In addition to the 2 deaths, 1 patient developed grade III skin and gut acVHD that resolved with intensification of immunosuppression. All surviving patients were weaned off immunosuppression by 2 years after HCT (Table 1).

Immune Reconstitution

Absolute numbers of $CD3^+$, $CD4^+$, $CD8^+$ T cells, and B cells showed recovery after day +180 and were in the normal range by 1 year (Figure 2A). Serum concentrations of immunoglobulins (IgA, IgM and IgG) remained at the lower limits of normal during the first year (Figure 2B). The concentration of IgG fell during the first year and was the lowest of all immunoglobulin levels (Figure 2B), presumably because of alemtuzumab-induced donor B cell depletion despite early administration of the agent.

Infections

Infectious complications were common in the first 6 months and gradually decreased in frequency in concert with immunologic recovery. During the first 100 days, 6 patients had bacteremia with organisms including coagulase-negative Staphylococcus, methicillin-resistant Staphylococcus *aureus*, *S. maltophilia,* and *S. pneumoniae.* One patient developed a urinary tract infection (UTI) and another had Clostridium *difficile* detected in stool. Viral infections in 4 patients included CMV and EBV replication, adenovirus, rhinovirus, varicella, and human herpes virus 6. CMV reactivation occurred in 1 of 3 patients who were at high risk for reactivation (donor seropositive/seronegative, recipient seropositive). EBV replication was asymptomatic and not treated; no patient progressed to post-transplantation lymphoproliferative disease. Between 100 and 180 days, 4 patients had bacteremia with coagulase-negative Staphylococcus, *E. faecalis,* and *E. gergoviae*, 1 had a UTI with *K. pneumoniae*, and 2 viral infections (CMV reactivation and parainfluenza) were noted. Between 180 and 365 days, 2 patients had bacteremia and another had an *E. coli*–derived UTI. No fungal infections were observed in any patients.

Mortality

One patient with Diamond Blackfan anemia (DBA with subsequent myelofibrosis) had graft rejection on day 180 (after initial engraftment). Restrictive pulmonary disease (presumed bronchiolitis obliterans either due to the underlying disease or GVHD) was noted during the 6 month follow-up period and treated symptomatically. A second (myeloablative) transplantation was performed on day +346, but he died 10 days later of progressive respiratory failure. Another recipient with Shwachman Diamond syndrome died

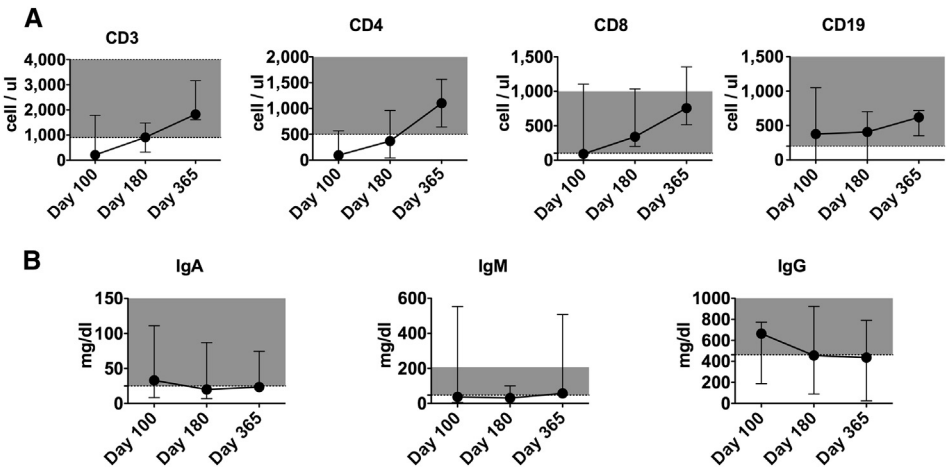

Figure 2. Serial immune reconstitution studies. Serial immune reconstitution showing gradual recovery with time in lymphocyte subset numbers. (A) Absolute numbers of lymphocyte subsets (median and range) CD3, CD4, CD8, and CD19 on days 100, 180 and 365 after transplantation. (B) Immunoglobulin concentrations (median and range) on days 100, 180, and 365 after transplantation. Shaded areas represent the normal values.

of complications of aGVHD and mutiorgan failure on day +45 (Table 1).

DISCUSSION

Although allogeneic HCT reverses the cytopenias of BMD, a key obstacle to contemplating the same early in patients with symptomatic BMD is the risk of mortality, treatment-related toxicities, and GVHD. These side effects are more likely in the absence of suitable matched family donors. This, as well as disease-related factors, such as organ involvement, exposure to transfusions, iron overload, underlying infections, and bleeding risks, compound the risks of transplantation toxicity and graft rejection [9]. Hence, HCT reports on large numbers of patients with BMD are scarce. Patients with many forms of BMD, including DBA, dyskeratosis congenita, severe congenital neutropenia, and congenital amegakaryocytic thrombocytopenia, have an increased risk of eventual clonal malignant transformation in the hematopoietic cells [5-8,12]. Avoiding the use of high-dose chemotherapy or radiation to achieve successful allogeneic donor cell engraftment is beneficial in this situation. The hypothesis for this study was based on substitution of myeloablation with recipient immunoablation in the preparative regimen to achieve donor cell engraftment irrespective of donor source (MRD or URD).

Previous studies have reported on HCT after myeloablative regimens with moderate success but with room for improvement. A registry report on outcomes of 61 patients transplanted for DBA was associated with a mortality rate of 18%. The OS was higher for MRD compared to that for URD (78% versus 45% at 1 year, and 76% versus 39% at 3 years, respectively). The majority of transplants (67%) in this report were from MRD [10]. Zeidler et al. reported on HCT in 11 patients with severe congenital neutropenia (SCN) before malignant transformation. Eight received MRD grafts, and 2 of the 3 alternate donor transplant recipients did not survive. OS after myeloablative regimens was 80% at a median follow-up of 10 months [13]. The French SCN registry reported HCT outcomes on 5 SCN patients before malignant transformation. All received myeloablative regimens with 80% engraftment and 40% mortality due to infection at 1-year after transplantation [14]. Bizzetto et al., on behalf of European Society for Blood and Marrow Transplantation, reported 95% OS at 3 years after related UCBT but only 61% OS after URD UCBT for hereditary BM failure syndromes other than Fanconi anemia [15].

An alemtuzumab-based immunosuppressive RIC regimen was successful in achieving donor cell engraftment in children with transfusion-dependent BMD enrolled on this trial. Survival and cure rates were high (82%) and comparable or better than previous reports. Although the number of patients reported is small, the majority underwent URD, which can be deferred in BMD patients for reasons of toxicity. This regimen provides an opportunity to lower the intensity and deliver HCT to this group of patients who otherwise have a significant risk of morbidity and mortality [12]. The follow-up period is relatively short and long-term tracking for the incidence of late effects including malignancy are in progress. Our experience with this group of BMD mirrors the successful experience previously reported in 7 patients who underwent transplantation for marrow failure with Shwachman Diamond syndrome using a similar regimen, none of which are included in this group [16]. Of note, only 1 patient in this current series received an UCB graft and had a good outcome. This experience is inadequate to determine whether this regimen is adequate to achieve donor engraftment with UCB transplantations and should be used with caution given the high rate of graft rejection described with UCB in other nonmalignant disorders [17].

The incidence of grade II to IV aGVHD at day +100 was low (9%) despite the predominance of URD transplantations. This compares favorably with previous reports that have cited a day +100 incidence of grade II to IV aGVHD of 24% [15]. This could perhaps be attributed to the use of T-depleting monoclonal antibody during conditioning and the young recipient age. The timing of alemtuzumab administration, however, was intentionally distal to the time of transplantation to achieve better recipient immunosuppression (to facilitate engraftment) at the expense of significant T cell depletion of the graft (to offset GVHD). The overall incidence of GVHD in this report is consistent with other reports of HCT using alemtuzumab [18,19]. Alemtuzumab, an antibody against CD52, not only effectively depletes T and B cells but also eliminates a subset of dendritic cells involved in antigen presentation [20]. GVHD incidence from previous reports on BMD transplantations range from 26% to 53% [10,15]. Though 27% in this report developed GVHD in the first year, it was encouraging that all surviving recipients were off immunosuppression without GVHD recurrence by 2 years after HCT. GVHD prophylaxis was weaned between 3 and 6 months in the absence of GVHD.

Withdrawal of immunosuppression and cellular immune reconstitution predictably paralleled each another (Figure 2A). Infections are to be expected with immunosuppressive T cell–depleting regimens and were encountered early in this group [21,22]. Patients were strictly ineligible to proceed to transplantation in the presence of invasive infections. No infection-related mortality was encountered, though multiple infections were documented; more in the first 6 months than the latter part of the first year. However, we believe that the timing of the alemtuzumab avoided delayed immune reconstitution and late infectious complications. CMV reactivation (33% in our series; no CMV disease) is anticipated in at-risk patients; rates as high as 50% to 85% are previously described [23,24]. Careful surveillance for viral reactivation with pre-emptive targeted therapy is strongly recommended until immune reconstitution and subsidence of lymphopenia. In contrast to standard practice, pre-emptive therapy for CMV reactivation was instituted at the detection of CMV replication, however trivial. Immune reconstitution studies were helpful in determining patterns of lymphocyte depletion and recovery. This was tracked carefully due to delayed immune reconstitution and risk for late infections with the peritransplantation use of alemtuzumab [21,23,25-27]. Tracking lymphocyte subpopulations was helpful in predicting infection risks and highlighted the need for very close monitoring before 6 months after HCT (Figure 2). The pace of immune reconstitution was robust after 6 months and matched that noted after HCT without T cell depletion, despite the use of a lymphodepleting antibody [28-30]. Immunoglobulin levels, however, were the last to recover.

In conclusion, this multicenter prospective study, utilizing an alemtuzumab-based RIC was effective in achieving favorable outcomes after HCT in children with BMD. This novel approach provided adequate host immune suppression to support engraftment with acceptable rates of GVHD and treatment-related mortality. Although the study included a small group of patients, the favorable results for this rare group of disorders support a consideration of transplantation early in this young population before the

development of serious marrow-related sequelae in those severely afflicted.

ACKNOWLEDGMENTS

This work was supported by the St. Louis Children's Hospital Foundation.

Financial disclosure: The authors have no financial disclosures to report.

Conflict of interest statement: There are no conflicts of interest to report.

REFERENCES

1. Sieff CA, Nisbet-Brown E, Nathan DG. Congenital bone marrow failure syndromes. *Br J Haematol*. 2000;111:30-42.
2. Alter BP. Bone marrow failure syndromes in children. *Pediatr Clin N Am*. 2002;49:973-988.
3. Myers KC, Davies SM. Hematopoietic stem cell transplantation for bone marrow failure syndromes in children. *Biol Blood Marrow Transplant*. 2009;15:279-292.
4. Donadieu J, Leblanc T, Bader Meunier B, et al. Analysis of risk factors for myelodysplasias, leukemias and death from infection among patients with congenital neutropenia. Experience of the French Severe Chronic Neutropenia Study Group. *Haematologica*. 2005;90:45-53.
5. Rosenberg PS, Alter BP, Link DC, et al. Neutrophil elastase mutations and risk of leukaemia in severe congenital neutropenia. *Br J Haematol*. 2008;140:210-213.
6. Alter B, Giri N, Savage S, Rosenberg P. Cancer in dyskeratosis congenita. *Blood*. 2009;113:6549-6557.
7. Alter B, Giri N, Savage S, et al. Malignancies and survival patterns in the National Cancer Institute inherited bone marrow failure syndromes cohort study. *Br J Haematol*. 2010;150:179-188.
8. Vlachos A, Rosenberg P, Atsidaftos E, et al. Incidence of neoplasia in Diamond Blackfan anemia: a report from the Diamond Blackfan Anemia Registry. *Blood*. 2012;119:3815-3819.
9. MacMillan ML, Wagner JE. Hematopoietic cell transplantation for congenital bone marrow failure. *Curr Opin Oncol*. 2005;17:106-113.
10. Roy V, Perez WS, Eapen M, et al. Bone marrow transplantation for diamond-blackfan anemia. *Biol Blood Marrow Transplant*. 2005;11:600-608.
11. Shenoy S, Grossman WJ, DiPersio J, et al. A novel reduced-intensity stem cell transplant regimen for nonmalignant disorders. *Bone Marrow Transplant*. 2005;35:345-352.
12. Morimoto K, Moore TB, Schiller G, Sakamoto KM. Transplantation for congenital bone marrow failure syndromes. *Bone Marrow Res*. 2011; 2011:849387.
13. Zeidler C, Welte K, Barak Y, et al. Stem cell transplantation in patients with severe congenital neutropenia without evidence of leukemic transformation. *Blood*. 2000;95:1195-1198.
14. Ferry C, Ouachee M, Leblanc T, et al. Hematopoietic stem cell transplantation in severe congenital neutropenia: experience of the French SCN register. *Bone Marrow Transplant*. 2005;35:45-50.
15. Bizzetto R, Bonfim C, Rocha V, et al. Outcomes after related and unrelated umbilical cord blood transplantation for hereditary bone marrow failure syndromes other than Fanconi anemia. *Haematologica*. 2011;96:134-141.
16. Bhatla D, Davies SM, Shenoy S, et al. Reduced-intensity conditioning is effective and safe for transplantation of patients with Shwachman-Diamond syndrome. *Bone Marrow Transplant*. 2008;42:159-165.
17. Kamani NR, Walters MC, Carter S, et al. Unrelated donor cord blood transplantation for children with severe sickle cell disease: results of one cohort from the phase II study from the Blood and Marrow Transplant Clinical Trials Network (BMT CTN). *Biol Blood Marrow Transplant*. 2012;18:1265-1272.
18. Styczynski J, Tallamy B, Waxman I, et al. A pilot study of reduced toxicity conditioning with BU, fludarabine and alemtuzumab before the allogeneic hematopoietic SCT in children and adolescents. *Bone Marrow Transplant*. 2011;46:790-799.
19. Law J, Cowan MJ, Dvorak CC, et al. Busulfan, fludarabine, and alemtuzumab as a reduced toxicity regimen for children with malignant and nonmalignant diseases improves engraftment and graft-versus-host disease without delaying immune reconstitution. *Biol Blood Marrow Transplant*. 2012;18:1656-1663.
20. Ratzinger G, Reagan JL, Heller G, et al. Differential CD52 expression by distinct myeloid dendritic cell subsets: implications for alemtuzumab activity at the level of antigen presentation in allogeneic graft-host interactions in transplantation. *Blood*. 2003;101: 1422-1429.
21. Chakrabarti S, Collingham KE, Marshall T, et al. Respiratory virus infections in adult T cell-depleted transplant recipients: the role of cellular immunity. *Transplantation*. 2001;72:1460-1463.
22. Novitzky N, Rouskova A. Infectious complications following T-cell depleted hematopoietic stem-cell transplantation. *Cytotherapy*. 2001; 3:165-173.
23. Chakrabarti S, Mackinnon S, Chopra R, et al. High incidence of cytomegalovirus infection after nonmyeloablative stem cell transplantation: potential role of Campath-1H in delaying immune reconstitution. *Blood*. 2002;99:4357-4363.
24. Vassiliou GS, Webb DK, Pamphilon D, et al. Improved outcome of alternative donor bone marrow transplantation in children with severe aplastic anaemia using a conditioning regimen containing low-dose total body irradiation, cyclophosphamide and Campath. *Br J Haematol*. 2001;114:701-705.
25. Avivi I, Chakrabarti S, Milligan DW, et al. Incidence and outcome of adenovirus disease in transplant recipients after reduced-intensity conditioning with alemtuzumab. *Biol Blood Marrow Transplant*. 2004; 10:186-194.
26. D'Sa S, Peggs K, Pizzey A, et al. T- and B-cell immune reconstitution and clinical outcome in patients with multiple myeloma receiving T-cell-depleted, reduced-intensity allogeneic stem cell transplantation with an alemtuzumab-containing conditioning regimen followed by escalated donor lymphocyte infusions. *Br J Haematol*. 2003;123: 309-322.
27. van Besien K, Dew A, Lin S, et al. Patterns and kinetics of T-cell chimerism after allo transplant with alemtuzumab-based conditioning: mixed chimerism protects from GVHD, but does not portend disease recurrence. *Leuk Lymphoma*. 2009;50:1809-1817.
28. Atkinson K. Reconstruction of the haemopoietic and immune systems after marrow transplantation. *Bone Marrow Transplant*. 1990;5: 209-226.
29. Fujimaki K, Maruta A, Yoshida M, et al. Immune reconstitution assessed during five years after allogeneic bone marrow transplantation. *Bone Marrow Transplant*. 2001;27:1275-1281.
30. Fallen PR, McGreavey L, Madrigal JA, et al. Factors affecting reconstitution of the T cell compartment in allogeneic haematopoietic cell transplant recipients. *Bone Marrow Transplant*. 2003;32: 1001-1014.
31. Castillo-Caro P, Dhanraj S, Haut P, et al. Proximal radio-ulnar synostosis with bone marrow failure syndrome in an infant without a HOXA11 mutation. *J Pediatr Hematol Oncol*. 2010;32:479-485.