ASBMT
American Society for Blood and Marrow Transplantation

Stable Long-Term Donor Engraftment following Reduced-Intensity Hematopoietic Cell Transplantation for Sickle Cell Disease

Lakshmanan Krishnamurti,[1] Sandhya Kharbanda,[2] Melinda A. Biernacki,[3] Wandi Zhang,[3] K. Scott Baker,[4] John E. Wagner,[4] Catherine J. Wu[3]

Reduced-intensity conditioning (RIC) regimens have the potential to decrease toxicities related to hematopoietic stem cell transplantation (HCT) in patients with sickle cell disease (SCD) and thus make HCT a more acceptable therapeutic option for this group of patients. We report the results of 7 patients enrolled on a study to evaluate safety and efficacy of HCT using bone marrow from an HLA matched sibling donor following an RIC regimen for patients with high-risk SCD. The conditioning regimen consisted of busulfan, fludarabine, equine antithymocyte globulin, and total lymphoid irradiation with shielding of the liver, lungs, heart, and gonads on day 1. Graft-versus-host disease (GVHD) prophylaxis consisted of cyclosporine and mycophenolate mofetil. The regimen was well tolerated, and all patients had hematopoietic recovery. Six of 7 patients are stably engrafted off immunosuppression and without sickle cell-related symptoms at 2 to 8.5 years after HCT. Consistent with the complete resolution of SCD related symptoms observed in the 6 engrafted patients, erythropoiesis of complete or predominantly donor origin was detected by red blood cell-specific chimerism assays, despite their having persistent mixed chimerism in the mononuclear and lymphoid compartments. These findings demonstrate the curative potential of allogeneic HCT after an RIC regimen in patients with SCD.

Biol Blood Marrow Transplant 14: 1270-1278 (2008)

KEY WORDS: Marrow and stem cell transplantation, Clinical results in inherited disorders, Conditioning regimen, Allo transplantation, Nonmyeloablative, Red cells, anemia—clinical, Sickle cell anemia

INTRODUCTION

Despite the advances in the health care of children with sickle cell disease (SCD) over the last 30 years, this disease continues to be associated with considerable morbidity and premature mortality, with a 25- to 30-year loss of life expectancy [1,2]. Stroke, organ failure, acute chest syndrome, pulmonary hypertension, and recurrent pain crises cause significant morbidity. Transfusion therapy decreases recurrence of stroke and other complications, but at the expense of increased risk for transfusion reactions, infections, and iron overload [3]. Hydroxyurea increases the total hemoglobin concentration, reduces the vaso-occlusive complications of pain and acute chest syndrome, and attenuates mortality in adults [4]. However, <30% of eligible patients are either prescribed or take this drug; not all adults respond to this treatment, and a small and as yet unquantified risk of latent transformation to leukemia with its long-term use remains as a concern [5].

Currently, allogeneic HCT is the only curative therapy for SCD. The results of HCT after conventional myeloablative therapy in young patients (<16 years of age) with SCD are highly encouraging, with an overall event-free survival (EFS) of approximately 85% and transplant-related mortality of <10% [6-13]. Moreover, stabilization or reversal of organ damage from SCD has been documented after HCT [14]. Despite these encouraging results, the use of this treatment approach has been limited by the infrequent availability of an HLA matched sibling donor [15] and the risks of early and late regimen-related

From the [1]Children's Hospital of Pittsburgh, University of Pittsburgh Medical Center, Pittsburgh, Pennsylvania; [2]University of Alabama, Birmingham, Alabama; [3]Dana Farber Cancer Institute, Boston, Massachusetts; and [4]University of Minnesota, Minneapolis, Minnesota.

Correspondence and reprint requests: Lakshmanan Krishnamurti, M.D., Division of Hematology/Oncology/Bone Marrow Transplantation, Children's Hospital of Pittsburgh, 3705 Fifth Avenue, Pittsburgh, PA 15213 (e-mail: krislx@chp.edu).

Received July 9, 2008; accepted August 27, 2008
1083-8791/08/1411-0001$34.00/0
doi:10.1016/j.bbmt.2008.08.016

Table 1. Clinical Characteristics and Hematologic Recovery in Patients undergoing RIC Allogeneic HCT for Severe SCD

Pt #	Age (Years/Sex)	Recipient β - Globin Genotype and Indication for HCT	Donor β -Globin Genotype	Conditioning Regimen	Donor ABO Compatibility	Infused Marrow Product (Cell Dose/kg Body Weight) TNC ($\times 10^8$)	CD34 ($\times 10^6$)	CD3- ($\times 10^7$)	Duration of ANC<0.5$\times 10^9$/L (Days)	Number of Transfusions post HCT (Day of Last Transfusion) PRBC	Platelets
1	8/F	$\beta^s\beta^s$ Stroke, Allosensitization	$\beta\beta^s$	BU, Flu, ATG, TLI*	matched	3.49	6.25	0.5	8	3 (100)	3 (22)
2	8/M	$\beta^s\beta^s$ Repeated ACS	$\beta\beta$	BU, Flu, ATG, TLI*	matched	5	1.26	0.5	8	3 (20)	3 (28)
3	6/M	$\beta^s\beta^s$ Repeated ACS, Silent stroke	$\beta\beta^s$	BU, Flu, ATG, TLI†	matched	4.51	3.49	0.5	14	3 (35)	3 (11)
4	8/M	$\beta^s\beta^0$ Repeated ACS	$\beta\beta^0$	BU, Flu, ATG, TLI†	major mismatch	4.3	4.72	0.54	13	3 (35)	3 (18)
5	18/F	$\beta^s\beta^s$ Stroke, Thrombosis, Allosensitization	$\beta\beta^s$	BU, Flu, ATG, TLI†	matched	2.61	1.02	0.28	13	3 (39)	3 (16)
6	16/M	$\beta^s\beta^s$ Repeated ACS AVN, Recurrent pain crises	$\beta\beta^s$	BU, Flu, ATG, TLI†	matched	4.25	3.7	0.55	13	1 (35)	4 (15)
7	16/F	$\beta^s\beta^s$ Repeated ACS	$\beta\beta$	BU, Flu, ATG, TLI†	matched	3.20	1.35	0.20	17	6 (35)	14 (25)

HCT indicates hematopoietic stem cell transplantation; ANC, absolute neutrophil count; TNC, total nucleated cells; PRBC, packed red blood cell; ATG, amtithymocyte globulin; BU, busulfan; TLI, total lymphoid irridiation; FLU, fludarabine.
*Busulfan was administered orally.
†Busulfan was administered intravenously.

toxicities related to intensive myeloablative therapy, especially in patients >16 years of age. These risks include organ toxicities, acute and chronic graft-versus-host disease (aGVHD, cGVHD), and late effects, including sterility and secondary malignancies. Therefore, as a means to reduce both acute and long-term toxicities, we evaluated the safety and efficacy of a reduced-intensity conditioning (RIC) approach for SCD.

MATERIALS AND METHODS

Study Subjects, and Patient Samples

Pediatric and adult patients with SCD with severe phenotype and without end-organ failure were considered eligible for this study, using the criteria previously described in a national collaborative trial for myeloablative HCT for SCD [13]. The study protocol was approved by the institutional review boards of the University of Pittsburgh, University of Minnesota and University of Alabama, Birmingham. All subjects or their legal guardians provided signed informed consent.

Conditioning and GVHD Prophylaxis Regimens

Preparative regimen

Patients were conditioned for transplantation with a regimen consisting of busulfan (BU) 0.8 mg/kg/dose intravenously every 6 hours, or 4 mg/kg/day orally in 2 divided doses on days −8 and −7, with targeted dosing, to achieve a BU steady state of 600-900 ng/mL; fludarabine (FLU) 35 mg/m^2 i.v. daily on days −6, −5, −4, −3, and −2; and equine antithymocyte globulin (ATG) 30 mg/kg i.v. daily on days −5, −4, −3, −2, and −1. Total lymphoid irradiation (TLI) was administered on day −2 as a single fraction of 500 cGy, with shielding of the liver, lungs, heart, and gonads. As patients were accrued over a 9-year period, a number of adjustments to the conditioning regimen were implemented over time, reflecting changes in clinical practice [16-18]. As shown in Table 1, the primary change was conversion of busulfan from an oral (patients 1-2) to an intravenous (patients 3-7) formulation.

GVHD prophylaxis

Posttransplantation immunosuppression consisted of cyclosporine and mycophenolate mofetil (MMF). Cyclosponine (CSA) was initiated on day −3 at 2.5 mg/kg i.v. over 2 hours every 12 hours for adults with normal renal function. For children <40 kg, the initial dose was 2.5 mg/kg i.v. over 2 hours every 8 hours. CSA was continued until day 180, and dose adjustments were made on the basis of toxicity and to maintain CSA trough levels of 200-300 mg/L. If no GVHD was observed, the CSA dose was tapered 10% per week beginning on day 181. MMF was initiated on day 0 at 15 mg/kg intravenously or orally twice a day. Subsequently, the MMF dose was modified to 15 mg/kg/dose or 1g thrice daily (whichever was lower)