Biol Blood Marrow Transplant 22 (2016) 1065–1072

Biology of Blood and Marrow Transplantation

journal homepage: www.bbmt.org

Clinical Research: Alternative Donors

Frequency and Risk Factors Associated with Cord Graft Failure after Transplant with Single-Unit Umbilical Cord Cells Supplemented by Haploidentical Cells with Reduced-Intensity Conditioning

Stephanie B. Tsai [1,2,*], Hongtao Liu [1], Tsiporah Shore [3], Yun Fan [4], Michael Bishop [1], Melissa M. Cushing [5], Usama Gergis [3], Lucy Godley [1], Justin Kline [1], Richard A. Larson [1], Guadalupe Martinez [6], Sebastian Mayer [3], Olatoyosi Odenike [2], Wendy Stock [1], Amittha Wickrema [7], Koen van Besien [3], Andrew S. Artz [1]

[1] *Section of Hematology/Oncology, Department of Medicine, University of Chicago Medical Center, Chicago, Illinois*
[2] *Section of Hematology-Oncology, Department of Medicine, Loyola University Medical Center, Maywood, Illinois*
[3] *Hematopoietic Stem Cell Transplant Program, Weill-Cornell Medical College, New York, New York*
[4] *Department of Hematology, Beijing Hospital, Beijing, China*
[5] *Department of Pathology, Weill-Cornell Medical College, New York, New York*
[6] *Stem Cell Laboratory, Department of Hematology, University of Chicago, Chicago, Illinois*
[7] *Department of Pathology, University of Chicago Medical Center, Chicago, Illinois*

Article history:
Received 17 September 2015
Accepted 11 February 2016

Key Words:
Graft failure
Alternative donor
Allogeneic stem cell transplantation
umbilical cord blood
Haploidentical graft

A B S T R A C T

Delayed engraftment and cord graft failure (CGF) are serious complications after unrelated cord blood (UCB) hematopoietic stem cell transplantation (HSCT), particularly when using low-cell-dose UCB units. The haplo-cord HSCT approach allows the use of a lower dose single UCB unit by co-infusion of a $CD34^+$ selected haploidentical graft, which provides early transient engraftment while awaiting durable UCB engraftment. We describe the frequency, complications, and risk factors of CGF after reduced-intensity conditioning haplo-cord HSCT. Among 107 patients who underwent haplo-cord HSCT, 94 were assessable for CGF, defined as <5% cord blood chimerism at day 60 in the myeloid and CD3 compartments, irrespective of neutrophil and platelet counts. CGF occurred in 14 of 94 assessable patients (15%). Median survival after CGF was 12.7 months with haploidentical or mixed haploidentical–autologous hematopoiesis persisting in the 7 surviving. Median progression-free survival after CGF was 7.7 months and was not statistically different from those without CGF (10.47 months; $P = .18$). In univariate analyses, no UCB factors were associated with CGF, including cell dose, cell viability, recipient major ABO mismatch against the UCB unit, or degree of HLA match. We also found no association of CGF with recipient cytomegalovirus serostatus, haploidentical donor age, or day 30 haploidentical chimerism. However, higher haploidentical total nucleated and $CD34^+$ cell doses and day 30 UCB chimerism < 5% in either the myeloid or CD3 compartments were associated with greater risk of CGF. We conclude that assessing chimerism at day 30 may foretell impending CGF, and avoidance of high haploidentical cell doses may reduce risk of CGF after haplo-cord HSCT. However, long-term survival is possible after CGF because of predominant haploidentical or mixed chimerism and hematopoietic function.

INTRODUCTION

Umbilical cord blood (UCB) is an alternative option to standard graft sources for hematopoietic stem cell transplantation (HSCT) and has been successfully used in both children and adults. Although UCB transplantation appears to achieve similar overall and leukemia-free survival relative to adult donor bone marrow or peripheral blood stem cell transplantation, a major limitation of UCB has been early morbidity and mortality from delayed hematopoietic recovery and a higher incidence of graft failure [1-3]. Delayed engraftment has been attributed to the low-stem-cell and T cell content of UCB units, a higher degree of HLA disparity in the donor–recipient pair, and poor T cell function after

Financial disclosure: See Acknowledgments on page 1071.
* Correspondence and reprint requests: Stephanie B. Tsai, MS, MD, Loyola University Medical Center, Division of Hematology-Oncology, 2160 South First Avenue, Building 112, Maywood, IL 60153.
E-mail address: s13@bu.edu (S.B. Tsai).

http://dx.doi.org/10.1016/j.bbmt.2016.02.010
1083-8791/

UCB transplant, leading to higher rates of infections in the early post-transplant period [2,4-6]. Nonetheless, UCB has much appeal, such as its availability to a wider range of recipients, observed low rates of graft-versus-host-disease in single UCB unit approaches, and potent graft-versus-tumor effect [6-8]. Theories attribute the superior graft-versus-tumor effects to aspects of the fetal immune system and its tolerance of the recipient environment, greater HLA-mismatch than conventional matched donors [9-11], and persistence of maternal microchimerism in the UCB graft [11,12]. Application of reduced-intensity conditioning (RIC) has extended UCB HSCT to older and less-fit patients [13,14]. As a result, UCB has become an increasingly used graft source for patients lacking HLA matched related donors.

Graft failure is one of the most feared complications after allogeneic HSCT, historically associated with high mortality [15]. Variably defined in the literature by failure to achieve neutrophil engraftment and/or donor chimerism, it invariably represents failure of sustained hematopoietic function by the intended graft. Factors associated with increased risk of graft failure after allogeneic HSCT include degree of HLA mismatch [16-18], use of unrelated grafts [4,18], T cell depletion [19], low cell dose [6], use of RIC [18,20] presence of donor-specific antibodies (DSAs) [21,22], and major ABO mismatch [23,24]. Mechanisms of graft failure described include infections in the recipient [25], drug toxicity, and rejection, which is thought to be recipient immune-mediated, involving T lymphocytes [26,27], natural killer cells [28-30], and DSAs [31-33]. Reported rates of graft failure in UCB transplantation range from 10% to 30%, with risk factors of greater HLA mismatch, lower cell dose, and RIC [2,4-6].

At our institutions we have advanced a UCB HSCT approach after RIC that allows the use of a lower dose single UCB unit by co-infusion of a $CD34^+$ selected haploidentical graft, which we refer to as haplo-cord HSCT. Although the ultimate goal is durable UCB hematopoiesis, the haploidentical graft provides early hematopoietic function until transition to UCB engraftment. With this approach we have achieved fast engraftment, comparable rates of relapse, and low rates of graft-versus-host-disease [34]. Given that haplo-cord HSCT is performed with the goal of durable umbilical cord engraftment, we sought to define the frequency of cord graft failure (CGF) and to describe subsequent outcomes.

METHODS

Eligibility and Enrollment

Adults with hematologic malignancies who underwent first allogeneic HSCT with haplo-cord HSCT with RIC on 2 consecutive Institutional Review Board–approved protocols at the University of Chicago Medical Center (UCMC) and the Weil Cornell Medical College (WCMC) between 2006 to 2013 were reviewed (clinical trial.gov NCT00943800 and NCT01810588). Inclusion criteria required no available matched related donor or matched unrelated donor. Other protocol inclusion and exclusion criteria were previously reported and similar in both protocols [34]. All patients provided written informed consent.

Treatment Plan

RIC used fludarabine (30 mg/m^2 daily days −7 to −3), melphalan (140 mg/m^2 on day −2 or 70 mg/m^2 daily days −3 and −2), and rabbit antithymocyte globulin (4.5 to 6.0 mg/kg daily divided over 3 to 4 days) and occasionally (n = 5) total body irradiation of 400 cGy (200 cGy twice a day for 1 day) if there was increased risk of central nervous system relapse. Immunosuppression used tacrolimus from days −1 through 180 and mycophenolate mofetil starting on day −2 at either 15 mg/kg every 8 hours or 1000 mg every 8 hours until day 28.

Graft Sources

Preferred haploidentical grafts were related, nonmaternal, and younger donors. Haploidentical donors grafts were harvested after granulocyte colony-stimulating factor mobilization at 5 μg/kg subcutaneously twice a day or 10 μg/kg daily, and $CD34^+$ cells were selected by the Isolex 300i CD34 depletion device (Isolex 300i Magnetic Cell Selection Systems; Nexell Therapeutics, Irvine CA) before April 2010 or the CliniMACS device (Miltenyi Biotec Inc., San Diego, CA) thereafter, aiming for a CD3 cell dose below 1×10^4/kg in the initial phase of the study. As of early 2012 the algorithm was adjusted to also limit the $CD34^+$ cell dose of the haploidentical donor to approximately 3 to 5×10^6/kg of recipient weight.

Preferred UCB units were HLA matched using standard UCB matching criteria: HLA-A and -B, by antigen matching, and at DRB1 by allele matching with at least 4/6 match. The minimum cell dose varied by protocol and protocol stage from $.5 \times 10^7$ total nucleated cells (TNCs)/kg recipient weight to 2.0×10^7 TNCs/kg. Preference was first given to optimal HLA matching once the minimum cell dose requirements were met.

Presence of DSAs against the haploidentical and UCB grafts was evaluated by solid-phase immunoassay (Luminex Corporation, Austin, TX). When DSAs were present, desensitization was performed as previously reported [35]. Haploidentical cells were infused on day 0, and UCB units were infused either later on the same day or on the following day.

Definitions and Study Endpoints

Disease risk was classified based on the American Society for Blood and Marrow Transplantation Request for Information 2006 risk scoring schema (http://www.asbmt.org). Chimerism at UCMC was performed by PCR for variable number of tandem repeats in unfractionated whole blood to assess myeloid chimerism and in the T cell compartment by CD3 sorting. At WCMC, CD33 chimerism was performed instead of unfractionated whole blood chimerism for myeloid chimerism, along with CD3 sorting. Protocol chimerism time points included baseline and post-transplant on days 14, 30, 60, 100, 180, and 360, or more frequently if clinically indicated. Results shown reflect peripheral blood unless only a marrow sample was available. Neutrophil recovery was defined as the first of 3 consecutive days with an absolute neutrophil count of at least 500/μL, and platelet recovery was defined as the first of 7 consecutive days with platelet count of at least 20,000/μL free of transfusion.

Umbilical CGF was the primary endpoint, defined as umbilical cord chimerism < 5% at day 60 in the myeloid and CD3 compartments irrespective of hematopoietic function. Death before day 60 excluded patients from the primary CGF analysis because cord engraftment may be either lost or gained at time points before day 60. Primary graft failure referred to a lack of neutrophil recovery by day 28. Postprocessing UCB viability was measured at various cord banks using Trypan Blue staining or, more recently, flow cytometric methodologies as per each cord bank protocol. At UCMC, post-thaw UCB viability was measured using Trypan Blue staining. At WCMC, post-thaw UCB viability was measured using Trypan Blue staining until recently when methodology was changed to flow cytometry with 7-actinomycin D staining. Data were censored as of July 31, 2014.

Statistical Analysis

Descriptive tables with patient and disease characteristics were tabulated. Univariate analyses were performed on various factors to identify potential risk factors for CGF. The limited number of events precluded multivariable models. Statistical analysis used to determine association between CGF and predictor variables were Fisher's exact test for dichotomous and logistic regression for continuous predictor variables. *P* values reported reflect 2-sided tests with an alpha of .05 considered significant, without adjustment for multiple testing. Overall survival (OS) and progression-free survival (PFS) estimates were generated by the Kaplan-Meier method. Patient death in remission defined transplant- or treatment-related mortality. STATA version 12 (StataCorp LP, College Station, TX) was used for all analyses.

RESULTS

Characteristics of the Study Cohort

A total of 107 adult patients with hematologic malignancies underwent RIC haplo-cord HSCT between January 2007 and December 2013 at UCMC and WCMC. Table 1 depicts baseline characteristics. Diseases indications were acute myelogenous leukemia (AML; 51%), acute lymphoblastic leukemia (12%), myelodysplastic syndrome (11%), and other (25%). Median patient age was 50 years (range, 18 to 73), and many had active disease at time of transplant (47%). The mean UCB cell dose was 2.1×10^7 TNCs/kg (range, .77 to