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Total Body Irradiation–Based Myeloablative Haploidentical Stem Cell Transplantation Is a Safe and Effective Alternative to Unrelated Donor Transplantation in Patients Without Matched Sibling Donors

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A B S T R A C T

We enrolled 30 patients on a prospective phase II trial utilizing a total body irradiation (TBI)–based myeloablative preparative regimen (fludarabine 30 mg/m^2/day × 3 days and TBI 150 cGy twice per day on day -4 to -1 [total dose 1200 cGy]) followed by infusion of unmanipulated peripheral blood stem cells from a haploidentical family donor (haplo). Postgrafting immunosuppression consisted of cyclophosphamide 50 mg/kg/day on days 3 and 4, mycophenolate mofetil through day 35, and tacrolimus through day 180. Median patient age was 46.5 years (range, 24 to 60). Transplantation diagnosis included acute myelogenous leukemia (n = 16), acute lymphoblastic leukemia (n = 6), chronic myelogenous leukemia (n = 5), myelodysplastic syndrome (n = 1), and non-Hodgkin's lymphoma (n = 2). Using the Dana Farber/Center for International Blood and Marrow Transplant Research/Disease Risk Index (DRI), patients were classified as low (n = 4), intermediate (n = 12), high (n = 11), and very high (n = 3) risk. All patients engrafted with a median time to neutrophil and platelet recovery of 16 and 25 days, respectively. All evaluable patients achieved sustained complete donor T cell and myeloid chimerism by day +30. Acute graft-versus-host disease (GVHD) grades II to IV and III and IV was seen in 43% and 23%, respectively. The cumulative incidence of chronic GVHD was 56% (severe in 10%). After a median follow-up of 24 months, the estimated 2-year overall survival (OS), disease-free survival (DFS), nonrelapse mortality, and relapse rate were 78%, 73%, 3%, and 24%, respectively. Two-year DFS and relapse rate in patients with low/intermediate risk disease was 100% and 0%, respectively, compared with 39% and 53% for patients with high/very high risk disease. When compared with a contemporaneously treated cohort of patients at our institution receiving myeloablative HLA-matched unrelated donor (MUD) transplantation (acute myelogenous leukemia [n = 17], acute lymphoblastic leukemia [n = 15], chronic myelogenous leukemia [n = 7], myelodysplastic syndrome [n = 7], non-Hodgkin lymphoma [n = 1], chronic lymphoblastic leukemia [n = 1]), outcomes were statistically similar, with 2-yr OS and DFS being 78% and 73%, respectively after haplo transplantation versus 71% and 64%, respectively, after MUD transplantation. In patients with DRI low/intermediate risk disease, 2-yr DFS was superior after haplo compared with MUD transplantations (100% versus 74%, $P = .032$), whereas there was no difference in DFS in patients with high/very high risk disease (39% versus 37% for haplo and MUD respectively, $P = .821$). Grade II to IV acute GVHD was seen less often after haplo compared with MUD transplantation (43% versus 63%, $P = .049$), as was moderate-to-severe chronic GVHD (22% versus 58%, $P = .003$). Myeloablative haplo transplantation using this regimen is a valid option for patients with advanced hematologic malignancies who lack timely access to a conventional donor. Outcomes appear at least equivalent to those seen in contemporaneous patients who underwent transplantation from MUD.

INTRODUCTION

Seventy percent of patients who urgently need a hematopoietic cell transplant (HSCT) do not have an available HLA-matched sibling donor. In such patients, a search

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for an HLA-matched unrelated donor (MUD) can identify an 8/8 HLA-identical donor for approximately 30% to 40% of transplant recipients. The probability of finding an acceptable MUD varies by racial/ethnic groups, ranging from 75% in white Europeans, to 30% to 40% in Mexican and Central/South Americans, to 15% to 20% for African Americans and black Caribbeans [1]. In addition, MUD transplantation is also complicated by the amount of time it takes from search initiation to transplantation, causing some patients to relapse or physically deteriorate while waiting for transplantation. In contrast, a haploidentical family member (haplo) can be identified and rapidly utilized in nearly all cases.

Historically, HSCT from partially HLA-mismatched relative has been complicated by unacceptably high incidences of graft rejection, severe graft-versus-host disease (GVHD), and nonrelapse mortality (NRM) [2,3]. To address the risk of graft rejection and GVHD, extensive T cell depletion has been utilized in association with antithymocyte globulin (ATG) and high peripheral blood stem cell (PBSC) dose [4]; however, NRM from infectious complications remains a challenge. More recently, the investigators at Johns Hopkins University have pioneered a method to selectively deplete alloreactive cells in vivo by administering high doses of cyclophosphamide (Cy) in a narrow window after transplantation [5]. After nonmyeloablative (NMA) conditioning, this approach has resulted in low NRM (4% and 15% at 1 and 2 years, respectively), because of low rates of GVHD and infectious complications. Immune reconstitution was promising with low risk of cytomegalovirus (CMV) or invasive mold infections. Using high-dose, post-transplantation cyclophosphamide (PT/Cy), crossing the HLA barrier in HSCT is now feasible without the need for extensive T cell depletion or serotherapy.

Although NMA haplo-HSCT utilizing PT/Cy results in low rates of GVHD, infection, and NRM, relapse remains the predominant cause of treatment failure, occurring in approximately 45% to 51% of patients [5,6]. To reduce the risk of relapse often associated with the use of NMA preparative regimens, our group and others have demonstrated the feasibility of myeloablative T cell–replete haplo-HSCT utilizing PT/Cy [7-9]. We have previously demonstrated that after a busulfan-based myeloablative haplo-HSCT, outcomes were promising with a 1-year estimate of overall survival (OS), disease-free survival (DFS), relapse, and NRM of 69%, 50%, 40%, and 10%, respectively, for all patients, and 88%, 67%, 33%, and 0% respectively for standard-risk patients (acute leukemia in remission or chronic-phase chronic myelogenous leukemia) [9]. An unexpected finding in that study was a high incidence of hemorrhagic cystitis (HC), and we hypothesized that the replacement of total body irradiation (TBI) for busulfan in the conditioning regimen would decrease the incidence of HC without affecting the overall safety and efficacy of the procedure.

PATIENTS AND METHODS

Eligibility and Enrollment

Thirty patients were accrued to this prospective study. Written informed consent was obtained for all of the patients in accordance with the Declaration of Helsinki. The study was approved by the institutional review board at Northside Hospital. Patients were eligible for inclusion if they were between 18 and 60 years of age, had a high-risk hematologic malignancy, were without a readily available matched related or unrelated donor, and had adequate organ function as defined by bilirubin <2.5 mg/dL, creatinine <2 mg/dL, cardiac ejection fraction ≥45%, forced expiratory volume in 1 second and forced vital capacity ≥60% predicted, Karnofsky performance status ≥70%, and were human immunodeficiency virus negative. Donors were required to be first-degree relatives (parent, child, sibling) of the recipient and partially HLA matched at 5 of 10 to 8 of 10 loci with the recipient. Donors were excluded if they had a positive HLA crossmatch in the host-versus-graft direction or high titer donor-specific antibodies, as determined by the pretransplantation panel reactive antibody testing.

Treatment Plan

Transplantation conditioning consisted of fludarabine 30 mg/m^2/day on days -7 to -5 and TBI 150 cGy twice daily on days -4 to -1 (total dose 1200 cGy). On day 0, patients received an unmanipulated PBSC allograft with a CD34 dose capped at 5×10^6/kg recipient weight (our institutional standard-of-care for all allogeneic transplantation protocols, based on historical data showing a correlation between CD34 dose and chronic GVHD [10,11]). On days +3 and +4, patients received 2 doses of Cy 50 mg/kg/day with mesna. Post-transplantation immunosuppression was initiated on day +5 with intravenous tacrolimus (target level 5 to 15 ng/mL) and oral mycophenolate mofetil (MMF) (15 mg/kg 3 times daily with a maximum daily dose of 3 gm). No immunosuppressive agents were administered until 24 hours after the last dose of PT/Cy, including corticosteroids. MMF and tacrolimus were discontinued without taper at day +35 and +180, respectively, in the absence of GVHD.

Antimicrobial prophylaxis was administered according to institutional practice guidelines. Standard prophylaxis was started on day 0 including a quinolone antibiotic and acyclovir. Antifungal prophylaxis consisted of an echinocandin (caspofungin or micafungin) until day +5, when the patient was started on oral therapy with either voriconazole or posaconazole. Filgrastim 5 μg/kg was given daily starting day +5 and continuing until neutrophil engraftment. Standard pneumocystis prophylaxis was started on day +30 and continued at least 6 months after transplantation and until immunosuppression was discontinued. Quantitative CMV PCR was monitored weekly starting day +1 and pre-emptive therapy was initiated if viral reactivation was detected (≥400 copies/mL). Quinolone antibiotic was continued until day +100 for BK virus prophylaxis.

Study Endpoints

Neutrophil engraftment was defined as the first of 3 days of an absolute neutrophil count of $>.5 \times 10^9$/L after transplantation. *Platelet engraftment* was defined as a platelet count of >20,000/μL without transfusion for the 7 preceding days. Acute GVHD was scored based on the modified Keystone criteria [12]. Grades III and IV acute GVHD was termed *severe* acute GVHD. Chronic GVHD diagnosis and grading were based on the National Institutes of Health consensus criteria [13]. BK virus HC was defined by the presence of urinary symptoms/signs in association with BK viruria. BKV-HC was classified according to the to the NCI Common Terminology Criteria for Adverse Events version 4.0: grade II, frequency with moderate dysuria and/or macroscopic hematuria; grade III, hospitalization indicated for transfusion, i.v. pain medications, and bladder irrigation; and grade IV, catastrophic bleeding; major nonelective intervention indicated. *Severe BK virus HC* was defined as grade ≥ III. Patient outcomes are reported as of January 1, 2015. The major study endpoints were sustained donor engraftment, incidence and severity of GVHD, NRM, DFS, and OS. Patients were considered to have died of NRM if there was no evidence of disease relapse or progression before death.

Chimerism Analysis

We assessed donor-recipient chimerism by the PCR-based amplification of a polymorphic short tandem repeat regions, followed by fragment separation by high resolution capillary electrophoresis (ABI 3130 XL Genetic Analyzer, Life Technologies, Carlsbad, CA) and quantitation using GeneMapper Software (Life Technologies). Peripheral blood samples were collected for chimerism analysis on days 30, 60, and 90 after transplantation. Samples were separated into myeloid and T cell lymphoid fractions by indirect sorting with immunomagnetic beads (StemCell Technologies, Vancouver, British Columbia, Canada). Primary antibodies were specific to CD33 and CD66b for myeloid cell and to CD3 for lymphoid T cell fractionation, respectively. The quality of sort was assessed using multiparametric flow cytometry (FACSCanto cytometer, DIVA analysis software; BD BioSciences, San Jose, CA). Genomic DNA was extracted from immunosorted cells and multiplex PCR was performed using commercial fluorescently labeled primer sets (ProfilerPlus, COfiler, NGM kits; Life Technologies). Four or 5-color fluorescence detection was performed on ABI 3130xl Genetic Analyzer and quantified using GeneMapper Software. For each informative short tandem repeat loci, allelic peak heights were determined, and the percentage of host alleles calculated as (Σ[host alleles peak height])/(Σ[host + donor alleles peak heights]) * 100. The range of the error of chimerism was determined to be nonuniform between different levels of chimerism and did not exceed 3.23%, 6.66%, 8.33%, 8.89%, 8.60%, 5.31%, and 3.07% for 1% to 5%, 6% to 20%, 21% to 40%, 41% to 60%, 61% to 80%, 81% to 95%,

and 95% to 99% host, respectively. Ranges for chimerism error assessment were selected empirically in our laboratory.

Comparison to Contemporaneous Cohort of Myeloablative Matched Unrelated Donor Transplantation Patients

A contemporaneous group of transplant recipients receiving myeloablative conditioning followed by MUD transplantation were utilized as a comparator cohort. Forty-eight consecutive patients receiving a myeloablative T cell–replete MUD transplant were analyzed. All donor-recipient pairs were HLA matched at 8/8 alleles (HLA-A, -B, -C, and -DR).

Statistical Analysis

The patient characteristics were compared between haplo and MUD transplantation groups using the Wilcoxon rank-sum test for the continuous variables and Fisher's exact test for the categorical variables. The OS and DFS were estimated by the Kaplan-Meier method. Comparisons of OS and DFS between different transplantations were evaluated using the log-rank test. The cumulative incidences, accounting for competing risks, were estimated for NRM, relapse, GVHD and BK cystitis. NRM and relapse were competing risks and they were taken together to define DFS. In the competing risks models of GVHD (or BK cystitis), death without GVHD (or death without BK cystitis) was defined as a competing risk. The cumulative incidences of these endpoints were compared between different transplantations using the Gray's test. All *P* values were 2-sided and *P* values less than .05 were considered as significant.

RESULTS

Characteristics of the Study Cohort

A total of 30 patients with a median age of 46.5 years (range, 24 to 60) with high risk hematologic malignancies were enrolled between April 2012 and May 2014. Patient and donor characteristics are listed in Table 1. Transplantation diagnosis included acute myelogenous leukemia (n = 16), acute lymphoblastic leukemia (n = 6), chronic myelogenous leukemia (n = 5), myelodysplastic syndrome (n = 1), and non-Hodgkin lymphoma (n = 2). Using the recently published Dana Farber/Center for International Blood and Marrow Transplant Research disease risk index (DRI) [14], patients were classified as low (n = 4), intermediate (n = 12), high (n = 11), and very high (n = 3) risk. Donor-recipient pairs were matched at a median of 5 (out of 10) HLA loci in the graft-versus-host direction (range, 5 to 7) and a median of 6 (out of 10) HLA loci in the host-versus-graft direction (range, 5 to 8). No patients had donor-specific antibodies before transplantation.

Table 1
Patient and Transplantation Characteristics

Characteristic	Value
Patients (n = 30)	
Age, median (range), yr	
Patient	46 (24-60)
Donor	32 (18-65)
HLA match	
GVH	
5/10	18
6/10	8
7/10	4
8/10	0
HVG	
5/10	13
6/10	12
7/10	3
8/10	2
Donor/recipient gender	
Male/female	8
Female/female	8
Male/male	10
Female/male	4
Donor relationship	
Parent	2
Sibling	12
Child	16
Transplanted cell dose, median (range)	
CD34 ($\times 10^6$/kg)	5.01 (4.01-5.06)
CD3 ($\times 10^7$/kg)	15.56 (3.74-53.8)
Disease	
AML	16
CR1	7
CR2/3	7
PIF	2
ALL	6
CR1	4
CR2	1
PIF	1
CML	5
CP	2
AP/BC	3
MDS	1
NHL	2
Disease risk (DFDRI)	
Very high	3
High	11
Intermediate	12
Low	4
HCT-CI	
0	7
1-2	13
3-5	10

GVH indicates graft-versus-host direction; HVG, host-versus-graft direction; AML, acute myelogenous leukemia; CR, complete remission; PIF, primary induction failure; ALL, acute lymphoblastic leukemia; CML, chronic myelogenous leukemia; CP, chronic phase; AP, accelerated phase; BC, blast crisis; MDS, myelodysplastic syndrome; NHL, non-Hodgkin lymphoma; DFDRI, Dana Farber disease risk index; HCT-CI, hematopoietic cell transplantation comorbidity index.

Engraftment and Chimerism

There were no cases of primary or secondary engraftment failure. Median times to neutrophil and platelet recovery were 16 and 25 days, respectively. Achievement of full donor chimerism was rapid, with all evaluable patients achieving durable complete (>95%) donor T cell and myeloid chimerism by day +30.

GVHD and NRM

The cumulative incidences of grades II to IV and grades III and IV acute GVHD were 43% and 23%, respectively. The cumulative incidences of any, moderate-to-severe, and severe chronic GVHD were 56%, 22%, and 10% respectively (Figure 1). Of the 16 patients who developed chronic GVHD, National Institutes of Health severity grade was mild, moderate, and severe in 10, 3, and 3 patients, respectively. Systemic immunosuppression was required for treatment in all patients with moderate/severe chronic GVHD and in 4 of 10 patients with mild chronic GVHD. When considering the 22 patients who were alive and in remission 1 year after transplantation (of the 28 evaluable patients with sufficient follow-up), only 5 of 22 patients required continued systemic immunosuppression past 1 year after transplantation. NRM at 2 years was 3%, which consisted of 1 death from noninfectious respiratory failure/acute respiratory distress syndrome 8 months after transplantation in a patient with chronic GVHD.

Regimen-related Toxicity and Infectious Complications

As noted in our prior experience with busulfan-based myeloablative haplo-HSCT, post-transplantation fever was common and occurred in the first 5 post-transplantation days in all patients. In 23 of 30 patients, no infectious cause was found for the fever, whereas an infectious source was noted in 7 patients (gram-positive bacteremia [n = 6],

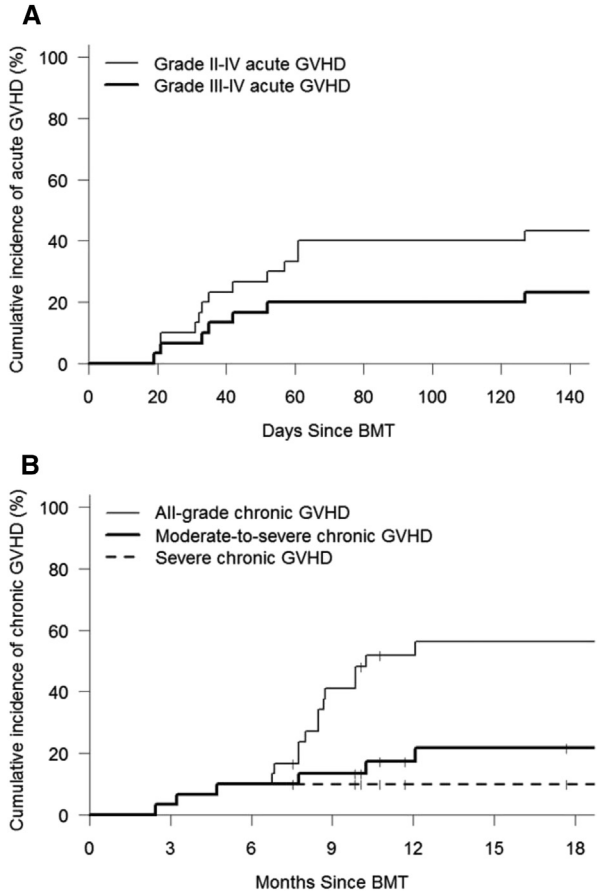

Figure 1. Cumulative incidence of (A) acute GVHD and (B) chronic GVHD.

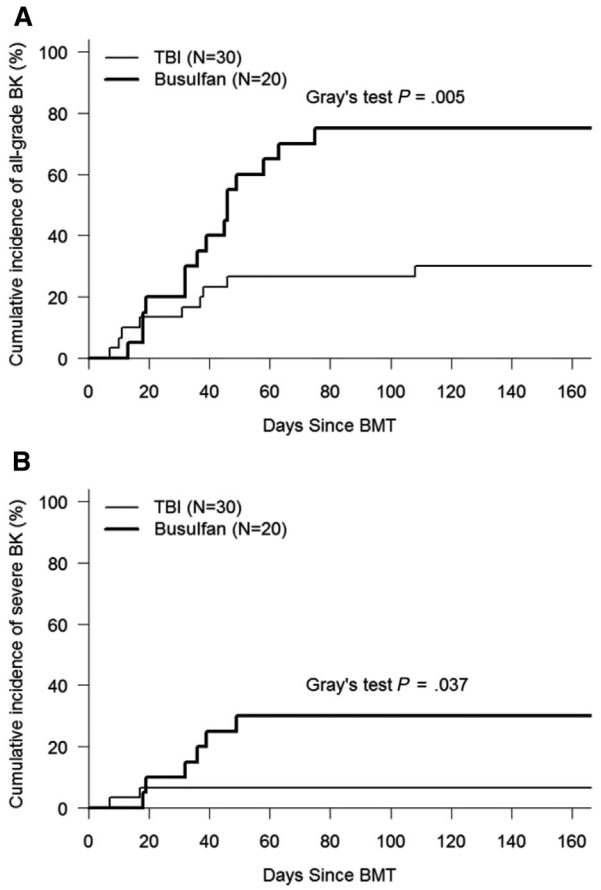

Figure 2. Cumulative incidence of (A) hemorrhagic cystitis of any grade and (B) grade ≥ 3 hemorrhagic cystitis.

metapneumavirus [n = 1]). Fevers resolved in all patients after administration of PT/Cy. CMV reactivation (≥400 copies/mL) occurred in 15 of 26 (58%) of at-risk patients (either donor and/or recipient with CMV-positive serostatus) at a median of day +43 after transplantation (range, 11 to 157) and was treated pre-emptively as per institutional guidelines. CMV disease did not occur. There were no episodes of invasive mold infection or infectious death in the first 100 days after transplantation. There were no cases of Epstein-Barr virus reactivation.

BK virus–associated HC of any grade occurred in 30% of patients and was severe (grade ≥3) in 7%. The median post-transplantation day for developing cystitis was day 31 (range, 7 to 108). Compared with our previous experience with busulfan-based myeloablative haplo-HSCT [9], HC occurred significantly less often after TBI-based myeloablative haplo-HSCT (any grade: 30% versus 75%, $P = .005$; severe HC: 7% versus 30%, $P = .037$) (Figure 2).

Relapse, DFS, and OS

With a median follow-up of 24 months (range, 8 to 44 months), estimated 2-year OS, DFS, and relapse was 78%, 73%, and 24%, respectively for all patients; 100%, 100%, and 0%, respectively, for patients with low/intermediate disease risk; 52%, 39%, and 53% for patients with high/very high disease risk (Figure 3). Twenty-four patients remain alive and 22 of these remain disease-free. Five of 6 deaths on study were attributable to disease relapse/progression.

Comparison of Transplantation Outcomes Between Myeloablative Haplo Versus MUD Transplantation

We compared outcomes of patients on this study receiving myeloablative haplo transplantation with a contemporaneously treated cohort of consecutive patients at our institution receiving myeloablative T cell–replete MUD transplantation. MUD donor-recipient pairs were HLA matched at 10/10 loci in 94% and 9/10 (single HLA-DQ mismatch) in 6%. Conditioning regimens for MUD transplantations included TBI/Cy (n = 4), TBI/etoposide (VP16) (n = 10), busulfan/cyclophosphamide (Bu/Cy) (n = 31), and fludarabine/busulfan for 4 days (Flu/Bu4) (n = 3). Haploidentical and MUD transplantation patients were well matched according to age, diagnosis, disease risk, CMV serostatus and comorbidity index, both when considering the entire MUD cohort or the subset of MUD patients receiving PBSC (Table 2). The groups did differ in the use of PBSC as the stem cell source, which was utilized in all 30 haploidentical transplant recipients compared with 32 of 48 MUD transplant recipients (100% versus 67%, $P < .001$). GVHD prophylaxis was tacrolimus and methotrexate in all MUD patients, and no patients received serotherapy.

When compared with the outcomes for recipients of myeloablative MUD transplantation, outcomes after myeloablative haplo-HSCT were statistically similar with 2-yr OS and DFS of 78% and 73%, respectively, after haplo transplantation versus 71% and 64%, respectively, after MUD transplantation (Figure 4A). In patients with DRI low/intermediate risk disease, 2-yr DFS was superior after

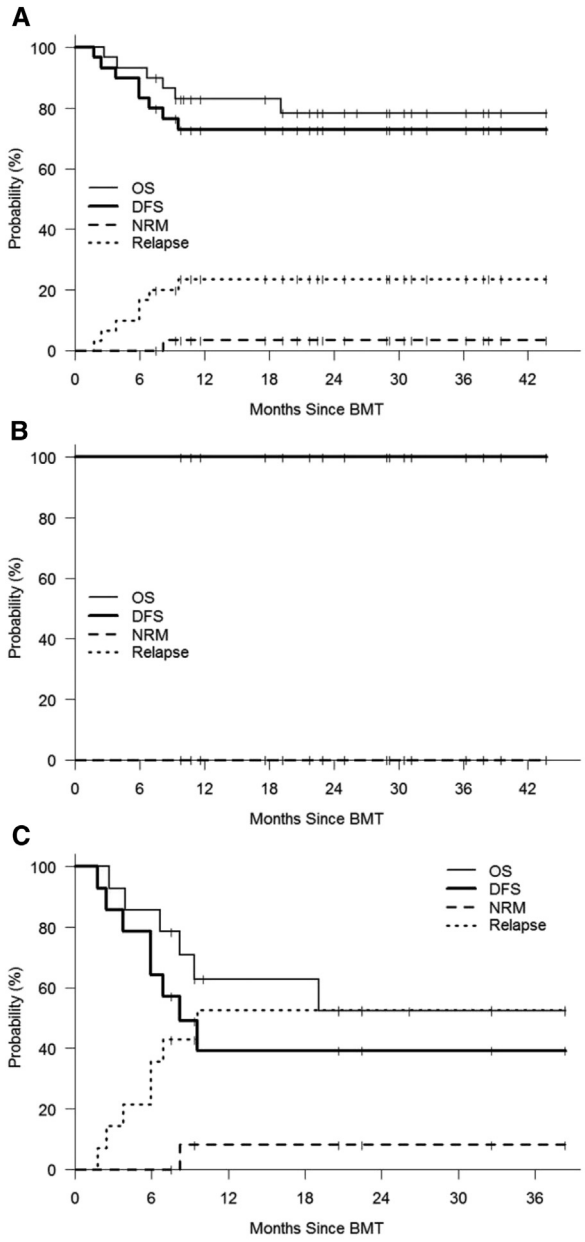

Figure 3. Kaplan-Meier analysis of overall survival, disease-free survival, nonrelapse mortality and relapse for (A) all patients, (B) patients with low/intermediate disease risk, and (C) patients with high/very high disease risk.

haplo compared with MUD transplantations (100% versus 74%, $P = .032$) (Figure 4B), whereas there was no difference in DFS in patients with high/very high risk disease (39% versus 37% for haplo and MUD, respectively, $P = .821$) (Figure 4C). Grade II to IV acute GVHD was seen less often after haplo compared with after MUD transplantation (43% versus 63%, $P = .049$), as was moderate-to-severe chronic GVHD (22% versus 58%, $P = .003$) (Figure 5). When MUD transplantations were restricted to the 32 patients receiving PBSC as the stem cell source, similar trends were seen (Table 3).

DISCUSSION

We report the outcomes of a prospective study of a TBI-based myeloablative, T cell–replete haploidentical PBSC transplantation. All patients engrafted and demonstrated 100% donor T cell and myeloid chimerism by day +30. The 2-year NRM and OS was 3% and 78%, respectively, for all patients, and 0% and 100%, respectively, for patients with low/intermediate disease risk. The cumulative incidence of grade III and IV acute GVHD and moderate-to-severe chronic GVHD was 23% and 22%, which are similar to those expected with myeloablative PBSC transplantations from HLA-matched donors. Remarkably, for a study of myeloablative transplantation from haploidentical donors, serious infections were rare. Notably, there were no serious CMV and invasive mold infections and no infectious mortality. BK virus–associated cystitis, which was a major cause of morbidity in our previously reported experience with busulfan-based myeloablative haploidentical transplantation [9], was significantly less frequent after TBI-based myeloablative conditioning with clinically significant (grade ≥ 3) HC occurring in only 2 (7%) patients.

In the past decade, there has been a growing interest in the use of haplo-HSCT because of the rapid and nearly universal availability of donors, which is a critical issue in patients with advanced hematologic malignancies. Enthusiasm for this approach has been furthered by significant advances in the field and improvements in supportive care, which have increased the safety of providing transplantations for patients across HLA barriers by mitigating the risks of infection, graft failure, and GVHD. A major advance in the success of haplo-HSCT is the use of properly timed PT/Cy, a technique pioneered by investigators at Johns Hopkins University [5,15]. In this strategy, patients receive an unmanipulated bone marrow (BM) allograft after an NMA conditioning regimen consisting of fludarabine, low-dose TBI (200 cGy), and Cy. After PT/Cy, patients receive GVHD prophylaxis consisting of tacrolimus and MMF, which results in acceptably low rates of graft rejection and GVHD, the 2 major historical barriers to successful haplo-HSCT. Furthermore, there was effective clinical immune reconstitution as demonstrated by the low incidence of severe opportunistic infections. However, relapse represented the major cause of treatment failure, occurring in 45% to 51% of transplant recipients [5,6]. One explanation for the high rate of relapse, as in other NMA HSCT trials, is that the transplantation conditioning was not intense enough to achieve sufficient tumor cytoreduction [16-19].

To reduce the risk of relapse in patients with high risk hematologic malignancies, our group has previously reported on the results of myeloablative haploidentical PBSCT, utilizing a busulfan-based conditioning regimen, unmanipulated PBSC allograft, and PT/Cy, tacrolimus, and MMF [9]. In addition to intensification of the preparative regimen dose as a strategy to reduce the risk of recurrence in high-risk malignancies, we also postulated that the use of PBSC, instead of BM, as the stem cell source may provide additional benefit in patients with high-risk malignancies. The use of PBSC, although associated with a higher incidence of chronic GVHD, has also been correlated with a significantly decreased risk of relapse in several large meta-analyses [20-22]. In our previous study, we provided transplantation to 20 patients with high-risk hematologic malignancies, including 11 (55%) who were not in remission at the time of transplantation. All patients had durable engraftment, with low rates of treatment-related mortality (10%). The 1-year OS and DFS were respectable at 69% and 50%, respectively, for all patients; and 88% and 67%, respectively, when considering only patients who underwent transplantation in remission.

Table 2
Comparison of Haplo and MUD Transplantation Recipient Characteristics

Characteristic	Haplo (n = 30)	MUD (n = 48)	MUD-PBSC (n = 32)	*P* Value (MUD versus Haplo)	*P* Value (MUD-PBSC versus Haplo)
Age, median (min, max)	46 (24, 60)	45 (19, 69)	44 (19, 67)	.805	.549
Gender				.639	1.000
Female	16 (53%)	29 (60%)	18 (56%)		
Male	14 (47%)	19 (40%)	14 (44%)		
Diagnosis				.509	.896
ALL	6 (20%)	11 (23%)	7 (22%)		
AML	16 (53%)	21 (43%)	15 (47%)		
CLL	0 (0%)	1 (2%)	1 (3%)		
CML	5 (17%)	7 (15%)	5 (16%)		
MDS	1 (3%)	7 (15%)	3 (9%)		
NHL	2 (7%)	1 (2%)	1 (3%)		
Transplantation status					
CR/CP	22 (73%)	35 (73%)	23 (72%)		
Advanced disease	8 (27%)	13 (27%)	9 (28%)		
Disease risk (DFDRI)				.091	.298
Low/intermediate	16 (53%)	35 (73%)	22 (69%)		
High/very high	14 (47%)	13 (27%)	10 (31%)		
HCT-CI				.400	.156
0	7 (23%)	17 (35%)	15 (47%)		
1/2	13 (43%)	14 (30%)	10 (31%)		
≥3	10 (33%)	17 (35%)	7 (22%)		
CMV serostatus				.135	.141
D+R+	17 (57%)	15 (31%)	10 (31%)		
D−R+	4 (13%)	12 (25%)	9 (28%)		
D+R−	4 (13%)	6 (13%)	3 (10%)		
D−R−	5 (17%)	15 (31%)	10 (31%)		
Stem cell source				<.001	1.00
PBSC	30 (100%)	32 (67%)	32 (100%)		
BM	0 (0%)	16 (33%)	0 (0%)		
Preparative regimen					
TBI/Cy (+Flu - Haplo)	30 (100%)	4 (8%)	3 (9%)	<.001	<.001
TBI/VP16	0 (0%)	10 (21%)	7 (22%)		
Bu/Cy	0 (0%)	31 (65%)	21 (66%)		
Flu/Bu4	0 (0%)	3 (6%)	1 (3%)		
GVHD prophylaxis					
Tac/Mtx	0 (0%)	48 (100%)	32 (100%)	<.001	<.001
PTCy/Tac/MMF	30 (100%)	0 (0%)	0 (0%)		
HLA match (MUD)					
10/10	N/A	45 (94%)	30 (94%)		
9/10 (DQ mm)	N/A	3 (6%)	2 (6%)		
Transplantation period	May 2011-May 2014	Jan 2010-Dec 2013	Jan 2010-Oct 2013		

Data presented are n (%), unless otherwise specified.
D indicates donor; R, recipient; Flu, fludarabine; VP16, etoposide; Bu, busulfan; Tac, tacrolimus; Mtx, methotrexate; N/A, not available; DQ mm, HLA-DQ mismatch.

The rates of grade III and IV acute GVHD and chronic GVHD were 10% and 35%, respectively, which appear lower than our current experience with TBI-based myeloablative haplo-HSCT. A relationship between TBI-based myeloablative conditioning with increased acute [23,24] and chronic [23] GVHD has been reported in the setting of PBSC transplantation, in contrast to the lack of effect in the setting of marrow transplantation [25,26].

Several other groups have published similar experiences with myeloablative haplo-HSCT with PT/Cy. Grosso et al. [7] reported a "2-step" strategy where a defined dose of haploidentical T cells (2×10^8/kg) were infused after myeloablative doses of TBI. Patients then received 60 mg/kg of CY on 2 consecutive days, followed later by infusion of highly purified $CD34^+$ cells from the donor. All patients engrafted and the cumulative incidences of grade III and IV acute GVHD and NRM were 7.4% and 22.5%, respectively, for the 27 patients treated. With a median follow-up of 40 months, OS was 48%. A second study from the same group [27], which includes only patients in remission at the time of transplantation, demonstrated a 2-yr NRM, relapse, and progression-free survival (PFS) of 4%, 19% and 74%, respectively (which compares similarly to those seen in our study—3%, 24%, and 73%, respectively). Raiola et al. [8] reported on 50 patients receiving a myeloablative haplo-HCT, a BM graft, and PT/CY. The regimens used were thiotepa, busulfan, and fludarabine (n = 35) or TBI and fludarabine (n = 15). Forty-five patients (90%) engrafted with an 18-month cumulative incidence of NRM, relapse, and PFS of 18%, 22%, and 51%, respectively. PFS was 67% for patients who underwent transplantation in remission versus 37% for patients who underwent transplantation with active disease.

When comparing our results with TBI-based myeloablative haplo-HSCT with the other 2 published experiences referenced above, it becomes evident that disease risk, as defined by either the DRI or disease status at the time of transplantation, is the primary driver of outcomes. The role of preparative regimen intensity is more difficult to define, although relapse rates in all 3 studies (19% to 24%) appears lower than reported for NMA haplo bone marrow transplantation (BMT) (45% to 51%) [5,6]. Whether PBSC or BM is the preferred stem cell source after myeloablative haplo-HSCT remains unclear; however, BM appears to be associated with a higher rate of graft failure, occurring in approximately 10% of patients in both the series by Raiola et al. [8] as well as the experience of Symons et al., recently

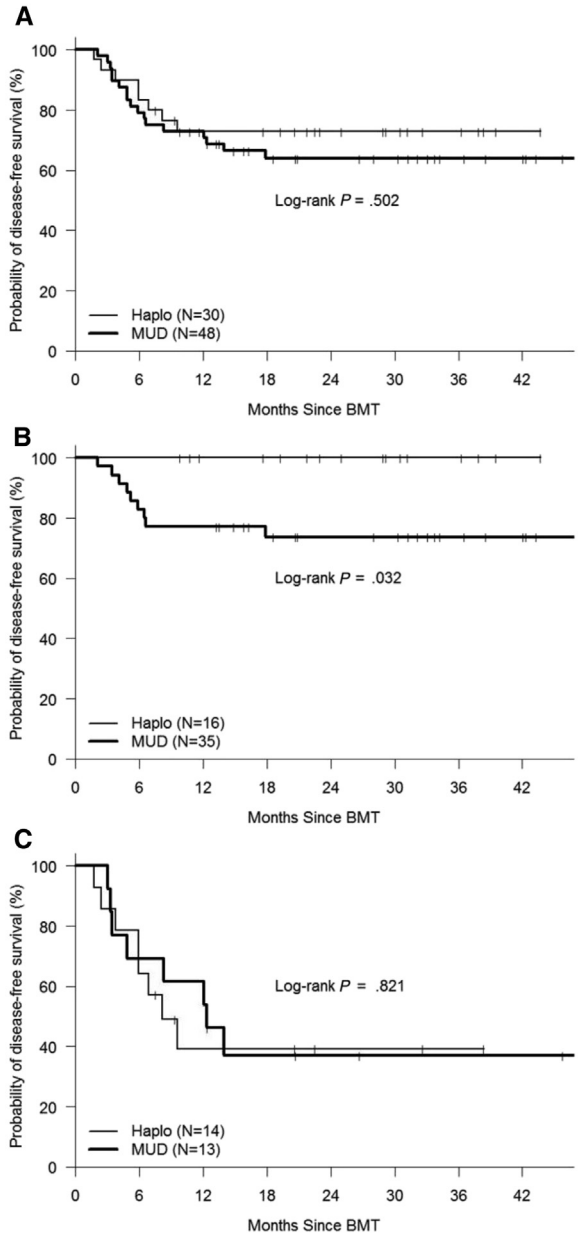

Figure 4. Kaplan-Meier analysis of disease-free survival, comparing recipients of myeloablative haplo-HSCT versus myeloablative MUD transplants for (A) all patients, (B) patients with low/intermediate disease risk, and (C) patients with high/very high disease risk.

reported at the 2015 Blood and Marrow Transplantation Tandem Meetings [28]. Graft failure has not been reported with PBSC based myeloablative haplo-HSCT and PT/Cy.

Our study has much in common with the "2-step" strategy by Grosso et al., utilizing TBI-based myeloablative conditioning, PBSC as the stem cell source, and PT/Cy, tacrolimus, and MMF for GVHD prophylaxis. Both report similar safety profiles with universal engraftment, low rates of infectious complications, low rates of NRM (3% versus 4%), and favorable 2-yr survival (78% versus 77%). The rates of grade III and IV GVHD and total chronic GVHD appear higher in our report (23% versus 4%, and 56% versus 22%), although chronic GVHD rates are naturally difficult to compare between different institutions because of differences in

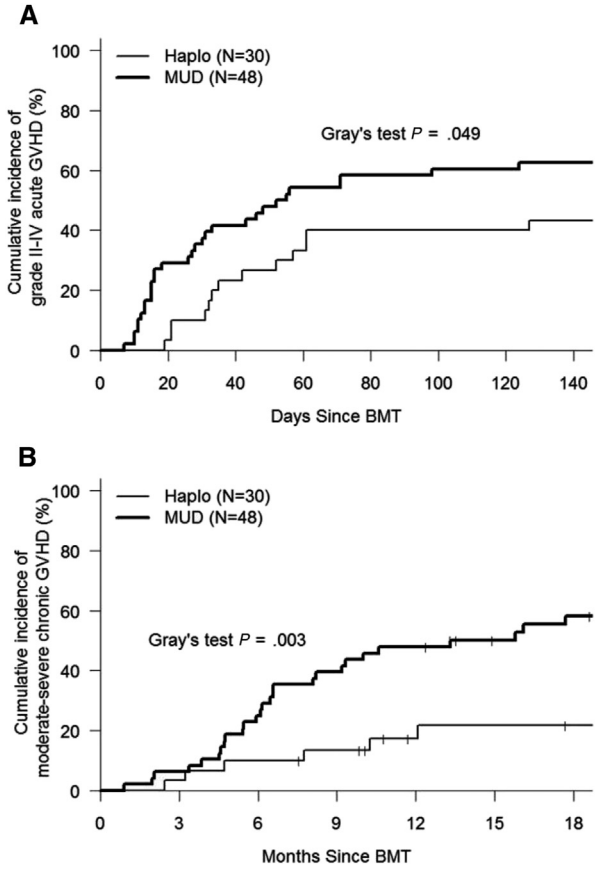

Figure 5. Cumulative incidence of (A) grade II to IV acute GVHD, and (B) moderate-to-severe chronic GVHD, comparing recipients of myeloablative haplo-HSCT versus myeloablative MUD transplantation.

long-term GVHD surveillance practices. The majority of chronic GVHD in our study was mild (10 of 16 cases) by National Institutes of Health criteria (1 or 2 organs with a severity score of 1, indicating no significant impairment of function or activities of daily living). It is noteworthy that severe chronic GVHD (10% versus 8%) was similar between

Table 3
Comparison of Haplo and MUD Transplantation Outcomes

Outcome	Haplo (n = 30)	MUD (n = 48)	MUD-PBSC (n = 32)	*P* Value (MUD versus Haplo)	*P* Value (MUD-PBSC versus Haplo)
Acute GVHD 2-4	43%	63%	63%	.049	.069
Acute GVHD 3-4	23%	23%	31%	.929	.377
Chronic GVHD	56%	69%	69%	.315	.242
Mild	34%	11%	5%		
Moderate	12%	35%	39%		
Severe	10%	23%	25%		
NRM (2 yr)	3%	13%	16%	.196	.130
Low risk	0%	6%	9%	.344	.247
High risk	8%	31%	30%	.162	0.181
Relapse (2 yr)	24%	23%	28%	.917	.725
Low risk	0%	20%	23%	.057	.044
High risk	53%	32%	40%	.224	.454
DFS (2 yr)	73%	64%	56%	.502	.216
Low risk	100%	74%	68%	.032	.017
High risk	39%	37%	30%	.821	.858
OS (2 yr)	78%	71%	62%	.550	.185
Low risk	100%	78%	67%	.062	.019
High risk	52%	54%	50%	.926	.768

the 2 protocols, as were OS and NRM. The requirement for stringent ex vivo T depletion of the hematopoietic cell product differentiates the "2-step" approach from ours and may limit its widespread applicability. Furthermore, given the resistance of hematopoietic stem cells to CY [29], such delayed infusion of selected $CD34^+$ cells may be unnecessary.

An unexpected outcome of our previous busulfan-based myeloablative haplo-HSCT experience [9] was a higher-than-expected rate of BK virus–induced HC. Although there was no mortality associated with this complication, it caused significant morbidity in patients. Several prior retrospective studies have suggested an association of high-dose busulfan with the development of HC [30-33], particularly in the setting of HLA-mismatched transplantations, where HC incidence ranged from 35% to 58% [32,34,35]. TBI, on the other hand, has been associated with significantly less risk of HC in both retrospective and prospective studies [25,36]. In light of these findings, we initiated the current clinical trial utilizing TBI-based myeloablative conditioning, instead of busulfan, for haplo-HSCT. In the current study, the incidence of BK virus–induced HC was significantly lower than that seen in our prior study (30% versus 75%, $P = .005$, respectively, for all HC; 7% versus 30%, $P = .037$ respectively for grade ≥ 3 HC). We believe that the switch from busulfan to TBI in the conditioning regimen was the major reason for this effect, as all other aspects of the regimen (stem cell source, GVHD prophylaxis, antimicrobial prophylaxis) remained the same. Clinically significant morbidity from BK virus occurs only with grade ≥ 3 HC, which typically requires hospital admission for pain control and/or bladder irrigation. This was relatively rare in our study, occurring in 2 cases (6.6% of 30 patients). As a comparison, Grosso et al. reported 1 case (3.6% of 28 patients), using their "2-step" TBI-based myeloablative haplo-HSCT [27].

In this report, we further show that survival outcomes after myeloablative haplo-HSCT are similar to that seen in a well-matched group of contemporaneously treated patients receiving myeloablative MUD transplantation. Despite a trend to higher disease risk among haplo-HSCT recipients (47% versus 27% high/very high disease risk, $P = .091$), there was no statistical difference in the incidence of relapse and survival. Furthermore, in the subset of patients with low/intermediate risk disease, 2-yr DFS was superior after haplo compared with MUD transplantations (100% versus 74%, $P = .032$). The incidence of grade II to IV acute GVHD (43% versus 63%, $P = .049$) and moderate-to-severe chronic GVHD (22% versus 58%, $P = .003$) was significantly lower after haploidentical versus MUD transplantation, leading to a low NMR (3% versus 13%, haplo-HSCT versus MUD; $P =$ not significant). The lower incidence of chronic GVHD occurred despite the greater use of PBSC in the haplo-HSCT group.

When considering the optimal transplan donor type, MUD versus haplo-HSCT, one must also consider the inherent advantages of haplo donors including near universal and rapid availability, as well as lower costs related to donor searching and graft acquisition. Whereas as almost all patients have an available haplo-matched family member, the availability of an 8/8 matched unrelated donor varies according to ethnic/racial background, ranging from 75% for white patients of European descent to less than 20% for black Americans of all ethnic backgrounds; for Hispanics, Asians, Pacific Islanders, and Native Americans, availability ranges from 27% and 52% [1]. Furthermore, given the complexities inherent in registry searching, time from initiation of donor searching to transplantation can be significant, averaging around 3 to 4 months [37-39]. Invariably, a proportion of patients will never proceed to transplantation, either because an appropriately matched donor cannot be found, or because disease recurrence or medical complications in the patient intervene. In 1 such study, about 45% of all patients for whom an unrelated allograft had been searched finally underwent transplantation, due to either lack of donor (20%) or deterioration of the patient's condition before donor identification (35%) [40]. Delays in getting patients to transplantation are adversely associated with patient outcome [39,41,42]. Therefore, getting patients to transplantation as quickly as feasibly possible should be of paramount importance.

Use of unrelated donors has also emerged as a significant cost driver, especially when the costs of stem cell procurement are included [40,43,44]. Total costs, including costs for donor search and graft acquisition, have been reported to be 50% to 60% higher after MUD versus sibling donor transplantations [40,45]. In the study by van Agthoven et al. [40], average 2-year costs, including donor identification expenses per patient undergoing transplantation (in Euros), were €98,334 (sibling bone marrow transplantation), €98,977 (sibling PBSC transplantation), and €151,754 (MUD), representing a 53% increase in costs for unrelated donor versus sibling donor transplantation. Nearly one third of the costs for MUD transplantation was spent on the search for a donor. When considering haplo-HSCT, costs for donor searching and stem cell procurement should be comparable to sibling donor transplantation, except for the extra HLA typing costs of additional family members. Therefore, from a macroeconomic perspective, we speculate that there should be significant cost saving from a strategy that prioritizes haplo-HSCT over MUD transplantation.

In conclusion, our results show that myeloablative haplo-HSCT using this TBI-based regimen results in favorable engraftment (100%), low NRM (3%), and an acceptable relapse rate (24%), similar to that expected after myeloablative MUD transplantation. Two-year DFS is not different from that seen in contemporaneous recipients of MUD transplants, although there appears to be improved DFS in haplo-HSCT patients with standard-risk disease. Haplo-HSCT may provide a benefit over MUD transplantation because of (1) increased donor availability, particularly in non-Caucasian patients; (2) more rapid access to donors, which may avoid unnecessary transplantation delays; and (3) lower donor searching and graft acquisition costs. Myeloablative haplo-HSCT is a valid option for patients with advanced hematologic malignancies who lack timely access to a conventional donor.

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REFERENCES

1. Gragert L, Eapen M, Williams E, et al. HLA match likelihoods for hematopoietic stem-cell grafts in the U.S. registry. *N Engl J Med*. 2014;371:339-348.

2. Beatty PG, Clift RA, Mickelson EM, et al. Marrow transplantation from related donors other than HLA-identical siblings. *N Engl J Med*. 1985; 313:765-771.
3. Szydlo R, Goldman JM, Klein JP, et al. Results of allogeneic bone marrow transplants for leukemia using donors other than HLA-identical siblings. *J Clin Oncol*. 1997;15:1767-1777.
4. Aversa F, Terenzi A, Tabilio A, et al. Full haplotype-mismatched hematopoietic stem-cell transplantation: a phase II study in patients with acute leukemia at high risk of relapse. *J Clin Oncol*. 2005;23:3447-3454.
5. Luznik L, O'Donnell PV, Symons HJ, et al. HLA-haploidentical bone marrow transplantation for hematologic malignancies using nonmyeloablative conditioning and high-dose, posttransplantation cyclophosphamide. *Biol Blood Marrow Transplant*. 2008;14:641-650.
6. Brunstein CG, Fuchs EJ, Carter SL, et al. Alternative donor transplantation after reduced intensity conditioning: results of parallel phase 2 trials using partially HLA-mismatched related bone marrow or unrelated double umbilical cord blood grafts. *Blood*. 2011;118:282-288.
7. Grosso D, Carabasi M, Filicko-O'Hara J, et al. A 2-step approach to myeloablative haploidentical stem cell transplantation: a phase 1/2 trial performed with optimized T-cell dosing. *Blood*. 2011;118: 4732-4739.
8. Raiola AM, Dominietto A, Ghiso A, et al. Unmanipulated haploidentical bone marrow transplantation and posttransplantation cyclophosphamide for hematologic malignancies after myeloablative conditioning. *Biol Blood Marrow Transplant*. 2013;19:117-122.
9. Solomon SR, Sizemore CA, Sanacore M, et al. Haploidentical transplantation using T cell replete peripheral blood stem cells and myeloablative conditioning in patients with high-risk hematologic malignancies who lack conventional donors is well tolerated and produces excellent relapse-free survival: results of a prospective phase II trial. *Biol Blood Marrow Transplant*. 2012;18:1859-1866.
10. Zaucha JM, Gooley T, Bensinger WI, et al. CD34 cell dose in granulocyte colony-stimulating factor-mobilized peripheral blood mononuclear cell grafts affects engraftment kinetics and development of extensive chronic graft-versus-host disease after human leukocyte antigen-identical sibling transplantation. *Blood*. 2001;98:3221-3227.
11. Mohty M, Bilger K, Jourdan E, et al. Higher doses of CD34+ peripheral blood stem cells are associated with increased mortality from chronic graft-versus-host disease after allogeneic HLA-identical sibling transplantation. *Leukemia*. 2003;17:869-875.
12. Przepiorka D, Weisdorf D, Martin P, et al. 1994 Consensus Conference on Acute GVHD Grading. *Bone Marrow Transplant*. 1995;15:825-828.
13. Filipovich AH, Weisdorf D, Pavletic S, et al. National Institutes of Health consensus development project on criteria for clinical trials in chronic graft-versus-host disease: I. Diagnosis and Staging Working Group Report. *Biol Blood Marrow Transplant*. 2005;11:945-956.
14. Armand P, Kim HT, Logan BR, et al. Validation and refinement of the Disease Risk Index for allogeneic stem cell transplantation. *Blood*. 2014; 123:3664-3671.
15. O'Donnell PV, Luznik L, Jones RJ, et al. Nonmyeloablative bone marrow transplantation from partially HLA-mismatched related donors using posttransplantation cyclophosphamide. *Biol Blood Marrow Transplant*. 2002;8:377-386.
16. Aoudjhane M, Labopin M, Gorin NC, et al. Comparative outcome of reduced intensity and myeloablative conditioning regimen in HLA identical sibling allogeneic haematopoietic stem cell transplantation for patients older than 50 years of age with acute myeloblastic leukaemia: a retrospective survey from the Acute Leukemia Working Party (ALWP) of the European group for Blood and Marrow Transplantation (EBMT). *Leukemia*. 2005;19:2304-2312.
17. Mohty M, Labopin M, Volin L, et al. Reduced-intensity versus conventional myeloablative conditioning allogeneic stem cell transplantation for patients with acute lymphoblastic leukemia: a retrospective study from the European Group for Blood and Marrow Transplantation. *Blood*. 2010;116:4439-4443.
18. Ringdén O, Labopin M, Ehninger G, et al. Reduced intensity conditioning compared with myeloablative conditioning using unrelated donor transplants in patients with acute myeloid leukemia. *J Clin Oncol*. 2009;27:4570-4577.
19. Shimoni A, Hardan I, Shem-Tov N, et al. Allogeneic hematopoietic stem-cell transplantation in AML and MDS using myeloablative versus reduced-intensity conditioning: the role of dose intensity. *Leukemia*. 2006;20:322-328.
20. Zhang H, Chen J, Que W. Allogeneic peripheral blood stem cell and bone marrow transplantation for hematologic malignancies: meta-analysis of randomized controlled trials. *Leuk Res*. 2012;36:431-437.
21. Stem Cell Trialists' Collaborative Group. Allogeneic peripheral blood stem-cell compared with bone marrow transplantation in the management of hematologic malignancies: an individual patient data meta-analysis of nine randomized trials. *J Clin Oncol*. 2005;23:5074-5087.
22. Chang YJ, Weng CL, Sun LX, Zhao YT. Allogeneic bone marrow transplantation compared to peripheral blood stem cell transplantation for the treatment of hematologic malignancies: a meta-analysis based on time-to-event data from randomized controlled trials. *Ann Hematol*. 2012;91:427-437.
23. Nagler A, Rocha V, Labopin M, et al. Allogeneic hematopoietic stem-cell transplantation for acute myeloid leukemia in remission: comparison of intravenous busulfan plus cyclophosphamide (Cy) versus total-body irradiation plus Cy as conditioning regimen—a report from the acute leukemia working party of the European group for blood and marrow transplantation. *J Clin Oncol*. 2013;31:3549-3556.
24. Copelan EA, Hamilton BK, Avalos B, et al. Better leukemia-free and overall survival in AML in first remission following cyclophosphamide in combination with busulfan compared with TBI. *Blood*. 2013;122: 3863-3870.
25. Ringden O, Remberger M, Ruutu T, et al. Increased risk of chronic graft-versus-host disease, obstructive bronchiolitis, and alopecia with busulfan versus total body irradiation: long-term results of a randomized trial in allogeneic marrow recipients with leukemia. Nordic Bone Marrow Transplantation Group. *Blood*. 1999;93:2196-2201.
26. Litzow MR, Perez WS, Klein JP, et al. Comparison of outcome following allogeneic bone marrow transplantation with cyclophosphamide-total body irradiation versus busulphan-cyclophosphamide conditioning regimens for acute myelogenous leukaemia in first remission. *Br J Haematol*. 2002;119:1115-1124.
27. Grosso D, Gaballa S, Alpdogan O, et al. A two-step approach to myeloablative haploidentical transplantation: low nonrelapse mortality and high survival confirmed in patients with earlier stage disease. *Biol Blood Marrow Transplant*. 2015;21:646-652.
28. Symons HJ, Chen A, Gamper C, et al. Haploidentical BMT using fully myeloablative conditioning, T cell replete bone marrow grafts, and post-transplant cyclophosphamide (PT/Cy) has limited toxicity and promising efficacy in largest reported experience with high risk hematologic malignancies. *Biol Blood Marrow Transplant*. 2015;21:S29.
29. Kastan M, Schlaffer E, Russo J, et al. Direct demonstration of elevated aldehyde dehydrogenase in human hematopoietic progenitor cells. *Blood*. 1990;75:1947-1950.
30. Yaghobi R, Ramzi M, Dehghani S. The role of different risk factors in clinical presentation of hemorrhagic cystitis in hematopoietic stem cell transplant recipients. *Transplant Proc*. 2009;41:2900-2902.
31. Trotman J, Nivison-Smith I, Dodds A. Haemorrhagic cystitis: incidence and risk factors in a transplant population using hyperhydration. *Bone Marrow Transplant*. 1999;23:797-801.
32. Silva Lde P, Patah PA, Saliba RM, et al. Hemorrhagic cystitis after allogeneic hematopoietic stem cell transplants is the complex result of BK virus infection, preparative regimen intensity and donor type. *Haematologica*. 2010;95:1183-1190.
33. Seber A, Shu XO, Defor T, et al. Risk factors for severe hemorrhagic cystitis following BMT. *Bone Marrow Transplant*. 1999;23:35-40.
34. Ogawa H, Ikegame K, Yoshihara S, et al. Unmanipulated HLA 2-3 antigen-mismatched (haploidentical) stem cell transplantation using nonmyeloablative conditioning. *Biol Blood Marrow Transplant*. 2006; 12:1073-1084.
35. Lu DP, Dong L, Wu T, et al. Conditioning including antithymocyte globulin followed by unmanipulated HLA-mismatched/haploidentical blood and marrow transplantation can achieve comparable outcomes with HLA-identical sibling transplantation. *Blood*. 2006;107:3065-3073.
36. Kroger N, Zabelina T, Kruger W, et al. Comparison of total body irradiation vs busulfan in combination with cyclophosphamide as conditioning for unrelated stem cell transplantation in CML patients. *Bone Marrow Transplant*. 2001;27:349-354.
37. Tiercy JM. Unrelated hematopoietic stem cell donor matching probability and search algorithm. *Bone Marrow Res*. 2012;2012:695018.
38. Rambaldi A, Bacigalupo A, Fanin R, et al. Outcome of patients activating an unrelated donor search: the impact of transplant with reduced intensity conditioning in a large cohort of consecutive high-risk patients. *Leukemia*. 2012;26:1779-1785.
39. Heemskerk MB, van Walraven SM, Cornelissen JJ, et al. How to improve the search for an unrelated haematopoietic stem cell donor. Faster is better than more!. *Bone Marrow Transplant*. 2005;35:645-652.
40. van Agthoven M, Groot MT, Verdonck LF, et al. Cost analysis of HLA-identical sibling and voluntary unrelated allogeneic bone marrow and peripheral blood stem cell transplantation in adults with acute myelocytic leukaemia or acute lymphoblastic leukaemia. *Bone Marrow Transplant*. 2002;30:243-251.
41. Frassoni F, Labopin M, Powles R, et al. Effect of centre on outcome of bone-marrow transplantation for acute myeloid leukaemia. Acute Leukaemia Working Party of the European Group for Blood and Marrow Transplantation. *Lancet*. 2000;355:1393-1398.
42. Craddock C, Labopin M, Pillai S, et al. Factors predicting outcome after unrelated donor stem cell transplantation in primary refractory acute myeloid leukaemia. *Leukemia*. 2011;25:808-813.
43. Saito AM, Cutler C, Zahrieh D, et al. Costs of allogeneic hematopoietic cell transplantation with high-dose regimens. *Biol Blood Marrow Transplant*. 2008;14:197-207.
44. Khera N, Zeliadt SB, Lee SJ. Economics of hematopoietic cell transplantation. *Blood*. 2012;120:1545-1551.
45. Matthes-Martin S, Potschger U, Barr R, et al. Costs and cost-effectiveness of allogeneic stem cell transplantation in children are predictable. *Biol Blood Marrow Transplant*. 2012;18:1533-1539.