Sharma, and Gabriela Rondon assisted with data collection and reviewed and approved the manuscript. Armeen Mahvash reviewed the radiologic images and reviewed and approved the manuscript. Amin Alousi and Elizabeth J. Shpall assisted with patient accrual and reviewed and approved the manuscript. Dimitrios P. Kontoyiannis and Richard E. Champlin critically reviewed and approved the manuscript. Stefan O. Ciurea designed the study and contributed to patient accrual, data collection, interpretation of the results, and manuscript preparation.

REFERENCES

1. Hirsch HH. BK virus: opportunity makes a pathogen. *Clin Infect Dis*. 2005;41:354-360.
2. Pahari A, Rees L. BK virus-associated renal problems: clinical implications. *Pediatr Nephrol*. 2003;18:743-748.
3. Barber CE, Hewlett TJ, Geldenhuys L, et al. BK virus nephropathy in a heart transplant recipient: case report and review of the literature. *Transpl Infect Dis*. 2006;8:113-121.
4. Basara N, Rasche FM, Schwalenberg T, et al. Hydronephrosis resulting from bilateral ureteral stenosis: a late complication of polyoma BK virus cystitis? *J Transplant*. 2010; [Epub 2010 Sept 27].
5. Bruno B, Zager RA, Boeckh MJ, et al. Adenovirus nephritis in hematopoietic stem cell transplantation. *Transplantation*. 2004;77: 1049-1057.
6. Iwamoto S, Azuma E, Hori H, et al. BK virus-associated fatal renal failure following late-onset hemorrhagic cystitis in an unrelated bone marrow transplantation. *Pediatr Hematol Oncol*. 2002;19:255-261.
7. Limaye AP, Smith KD, Cook L, et al. Polyomavirus nephropathy in native kidneys of non-renal transplant recipients. *Am J Transplant*. 2005;5:614-620.
8. Shapiro S, Robin M, Esperou H, et al. Polyomavirus nephropathy in the native kidneys of an unrelated cord blood transplant recipient followed by a disseminated polyomavirus infection. *Transplantation*. 2006;82:292-293.
9. Stracke S, Helmchen U, von Muller L, et al. Polyoma virus-associated interstitial nephritis in a patient with acute myeloic leukaemia and peripheral blood stem cell transplantation. *Nephrol Dial Transplant*. 2003;18:2431-2433.
10. Atencio IA, Shadan FF, Zhou XJ, et al. Adult mouse kidneys become permissive to acute polyomavirus infection and reactivate persistent infections in response to cellular damage and regeneration. *J Virol*. 1993;67:1424-1432.
11. Cundy KC. Clinical pharmacokinetics of the antiviral nucleotide analogues cidofovir and adefovir. *Clin Pharmacokinet*. 1999;36: 127-143.
12. Dropulic LK, Jones RJ. Polyomavirus BK infection in blood and marrow transplant recipients. *Bone Marrow Transplant*. 2008;41: 11-18.
13. Giralt S, Ballen K, Rizzo D, et al. Reduced-intensity conditioning regimen workshop: defining the dose spectrum. Report of a workshop convened by the Center for International Blood and Marrow Transplant Research. *Biol Blood Marrow Transplant*. 2009;15:367-369.
14. Przepiorka D, Weisdorf D, Martin P, et al. 1994 Consensus Conference on Acute GVHD Grading. *Bone Marrow Transplant*. 1995;15:825-828.
15. 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.
16. Gaston KE, Gabriel DA, Lavelle JP. Rare cause of ureteral obstruction. *Urology*. 2005;66:1110.
17. Mackenzie EF, Poulding JM, Harrison PR, et al. Human polyoma virus (HPV): a significant pathogen in renal transplantation. *Proc Eur Dial Transplant Assoc*. 1978;15:352-360.
18. Coleman DV, Mackenzie EF, Gardner SD, et al. Human polyomavirus (BK) infection and ureteric stenosis in renal allograft recipients. *J Clin Pathol*. 1978;31:338-347.

Intravenous Busulfan-Cyclophosphamide as a Preparative Regimen Before Allogeneic Hematopoietic Stem Cell Transplantation for Adult Patients with Acute Lymphoblastic Leukemia

Wei Tang, Ling Wang, Wei-Li Zhao, Yu-Bao Chen, Zhi-Xiang Shen, Jiong Hu

The use of i.v. busulfan (BU) instead of the oral formulation can improve outcomes in patients undergoing allogeneic hematopoietic stem cell transplantation (allo-HSCT) by reducing toxicity and transplantation-

related mortality (TRM). There are limited reports of i.v. BU used to treat patients with acute lymphoblastic leukemia (ALL). The present study was performed to evaluate the efficacy and toxicity of i.v. BU/cyclophosphamide (CY) conditioning in adult ALL. We retrospectively analyzed 42 consecutive patients who underwent allo-HSCT with BU/CY conditioning between January 2007 and October 2010 with an HLA-matched donor (sibling, n = 18; unrelated, n = 24). Thirty-three patients were in first complete remission (CR1), 2 were in second complete remission (CR2), and 7 were in a more advanced stage. Median patient age was 28 years (range, 17~55 years). The median follow-up was 15 months (range, 1~48 months). Overall, 13 patients died, for a 30-month overall survival of 56.5% $\pm$ 10.6% (65.7% $\pm$ 12.5% for patients in CR1 vs 25.4% $\pm$ 15.5% for those in CR2 or beyond; $P < .001$). Eleven patients experienced relapse between 2 and 26 months after allo-HSCT, with a 30-month relapse rate (RR) of 40% $\pm$ 10.9% (32.0% $\pm$ 12.7% for patients in CR1 vs 71.4% $\pm$ 17.1% for those in CR2 or beyond; $P = .001$). The incidence of grade II-IV acute graft-versus-host disease (GVHD) was 39.2% $\pm$ 8.8%, and that of grade III-IV acute GVHD was 7.4% $\pm$ 4.1%. The incidence of chronic GVHD was 63.9% $\pm$ 11.7%, and that of extensive chronic GVHD was 19.3% $\pm$ 7.9%. Only 2 cases of clinically diagnosed veno-occlusive disease (VOD) were documented (4.7%), and 1 of these patients died of severe VOD. Other BU/CY conditioning–associated toxicities were diffuse alveolar hemorrhage in 1 patient and hemorrhagic cystitis in 8 patients. Four patients died due to TRM, for a 30-month TRM of 9.7% $\pm$ 4.6%. This study demonstrates that i.v. BU/CY can be considered a feasible conditioning regimen for adult ALL, with low incidences of VOD and TRM.

INTRODUCTION

The selection of conditioning regimen for allogeneic hematopoietic stem cell transplantation (allo-HSCT) should be optimized to provide the maximal antileukemic effect with minimal toxicity. Combinations of cyclophosphamide (CY) with total body irradiation (TBI) or busulfan (BU) have been used for more than 20 years as conditioning regimens for various hematologic malignancies [1-3]. TBI has the advantage of eradicating leukemic cells of the central nervous system or testicles as "sanctuary sites" [4]. BU is an alkylating agent commonly used in high-dose chemotherapy regimens, which offered the advantage of easier administration and a lack of the toxicity associated with TBI, including interstitial pneumonitis, cataract, growth retardation, and other endocrine disturbances [4,5]. A BU-based regimen has proven superior for treating chronic myelogenous leukemia (CML), whereas a TBI-based regimen has been shown to be superior for treating acute myelogenous leukemia (AML), in terms of lower leukemia relapse and transplantation-related mortality (TRM) [6-9]. For acute lymphoblastic leukemia (ALL), the available data are controversial, based on the limited number of patients reported [10]. In a recent meta-analysis, BU/CY regimens were associated with higher TRM but a similar relapse rate compared with TBI/CY regimens [11]. BU/CY regimens also are associated with more complications, including liver veno-occlusive disease (VOD) and hemorrhagic cystitis [11-13]. In pediatric patients, the main cause of inferior outcomes with BU/CY regimens is the higher mortality rate [14]. Of note, most of these studies compared TBI/CY and oral BU/CY regimens,

characterized by wide interpatient and intrapatient variability in pharmacokinetics. Currently used i.v. BU formulations have more reliable and consistent pharmacokinetics [5]. In previous studies, the i.v. BU/CY was associated with decreased incidence of VOD, VOD-related mortality, and overall 100-day mortality [15,16]. There are limited published reports on the role of i.v. BU/CY as a conditioning regimen for ALL. Here we report the clinical outcomes in adult patients with ALL undergoing allo-HSCT from HLA-matched sibling or unrelated donors.

PATIENTS AND METHODS

Patients and Eligibility Criteria for Allo-HSCT

This study retrospectively analyzed the outcomes of 42 consecutive adult patients with ALL who underwent allo-HSCT between January 2007 and October 2010 in the Blood and Marrow Transplantation Center of Rui Jin Hospital, Shanghai Jiao Tong University School of Medicine. Eligibility criteria for allo-HSCT were as follows: either an HLA-matched sibling or an unrelated donor available, performance status <2, normal renal and hepatic function (serum creatinine ≤1.5 mg/100 mL, serum bilirubin ≤1.0 mg/100 mL, serum glutamic-pyruvic transaminase ≤3 times the upper normal limit), cardiac left ventricular ejection fraction ≥50%, normal pulmonary function tests (including forced expiratory volume in 1 minute), and negative serology for hepatitis B and human immunodeficiency virus. All patients provided written informed consent to undergo allo-HSCT.

Conditioning Regimen

Patients received i.v. BU (Busulfex) at 1.6 mg/kg every 12 hours for 8 doses over 4 days (from day −7 to day −4 before allo-HSCT). The i.v. BU dose was based on actual body weight and was administered by infusion over 4 hours [17]. The i.v. BU was followed by CY 60 mg/kg/day i.v. over 4 hours for 2 days (days −3 and −2). Allogeneic bone marrow or peripheral blood stem cells were infused on day 0, followed by granulocyte colony-stimulating factor 5 μg/kg i.v. starting on day +3 after allo-HSCT until the absolute neutrophil count exceeded 0.5×10^9/L. Graft-versus-host disease (GVHD) prophylaxis consisted of cyclosporine 1.5 mg/kg twice daily via continuous i.v. infusion, with a switch to the oral formulation if tolerated. The cyclosporine dose was adjusted to a therapeutic level of 200∼250 μg/mL. In addition, methotrexate was given at doses of 15 mg/m^2 on day +1 and 10 mg/m^2 on days +3, +6 and +11, and mycophenolate mofetil 1.0 g was given twice daily from day +1 to day +30. Antithymocyte globulin rabbit (Thymoglobulin; Genzyme, Cambridge, MA) at a total dose of 6 mg/kg was given from day −4 to day −1 to patients undergoing allo-HSCT from an unrelated donor.

Supportive Care

Phenytoin was administered as seizure prophylaxis before and during BU treatment in all patients. For VOD prophylaxis, lipo-prostaglandin E1 (lipo-PGE1) 0.5 μg/kg was given regularly at the start of conditioning until day +21. Mesna, antiemetics, blood components, and other supportive care measures were provided according to institutional guidelines.

Toxicity

The definition of VOD used in the present study was based on the Seattle clinical criteria [18], with a diagnosis of VOD requiring at least 2 of the following within 20 days of transplantation: serum bilirubin >2 mg/dL (34 μM/L), hepatomegaly, and weight gain >5% over baseline. VOD was classified as mild, moderate, or severe. Mild VOD was defined as the absence of adverse effects of liver dysfunction with complete resolution of symptoms and signs. Moderate VOD was defined as adverse effects of liver dysfunction requiring therapy, such as diuretics for fluid retention and analgesics for pain. Severe VOD was defined as the persistence of symptoms after day 100 or death before day 100 with ongoing VOD. Acute GVHD (aGVHD) and chronic GVHD (cGVHD) were diagnosed and graded accordingly [19,20].

Statistical Analysis

The outcomes of allo-HSCT are presented in terms of overall survival (OS), event-free survival (EFS), relapse rate (RR), TRM, and regimen-related toxicities. The probabilities of leukemia relapse and TRM were calculated using cumulative incidence estimates. Survival rates were calculated using Kaplan-Meier estimates. Univariate comparisons were performed using the log-rank test [21]. For analysis of OS, failure was defined as the time of death from any cause. For analysis of TRM, failure was defined as death occurring while the patient was in continuous complete remission (CR). For analysis of relapse, failure was defined as clinical or hematologic recurrence of ALL at any site. For analysis of EFS, treatment was considered to have failed at the time of clinical or hematologic relapse at any site or at the time of death from any cause. Data for patients who were alive and in CR were censored at the time of the last follow-up visit.

Table 1. Patient Characteristics

Sex, n	
Male	28
Female	14
Age, years, median (range)	28 (17∼55)
Donor type, n	
HLA-matched sibling	18
HLA-matched unrelated donor	24
Disease stage, n	
CR1	33 (Ph$^+$ ALL, 7)
CR2	2 (Ph$^+$ ALL, 1)
Relapsed/refractory	7 (Ph$^+$ ALL, 2)
HSC source, n	
Bone marrow	3
Peripheral blood	39
Follow-up, months, median (range)	15 (1∼48)

RESULTS

Patients and Characteristics

A total of 42 patients were included in the analysis. Demographic data for these patients are summarized in Table 1. The median patient age was 28 years (range, 17∼55 years). Thirty-three patients were in first CR (CR1), 2 were in second CR (CR2), and 7 were in a more advanced stage (refractory/relapse). Twenty-four patients underwent allo-HSCT with an HLA-matched unrelated donor, and 18 patients underwent allo-HSCT with an HLA-matched sibling donor. The stem sell source was peripheral blood stem cells in 39 patients and bone marrow in 3 patients. The median follow-up was 15 months (range, 1∼48 months).

Engraftment and Chimerism

For all 42 patients, the median number of mononucleated cells and CD34 cells infused was 5.0×10^8/kg (range, 1.7∼8.9 × 10^8/kg) and 4.9 × 10^6/kg (range, 1.0∼15.6 × 10^6/kg), respectively.
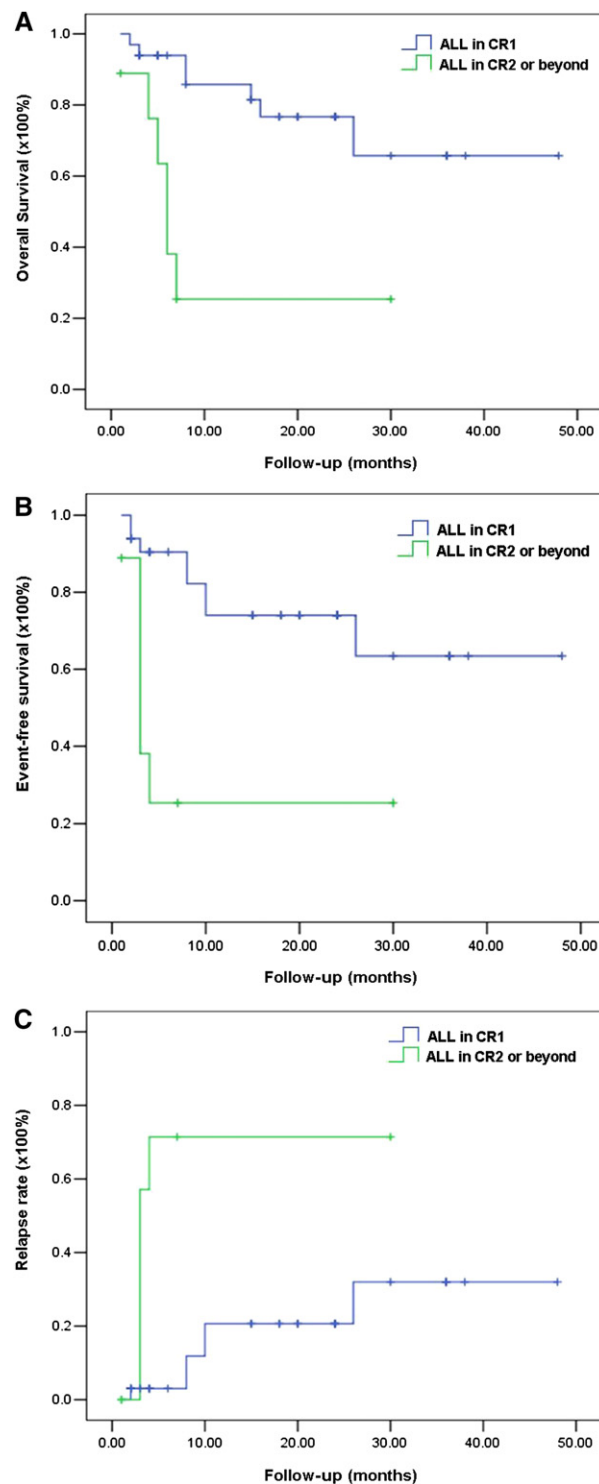
Figure 1. A, OS of patients undergoing allo-HSCT with an i.v. BU/CY regimen. The 30-month OS after allo-HSCT was 65.3% ± 12.5% for patients undergoing transplantation while in CR1 and 25.4% ± 15.5% for those undergoing transplantation in CR2 or a more advanced stage (P <.001). B, EFS of patients undergoing allo-HSCT with an i.v. BU/CY regimen. The 30-month EFS was 63.4% ± 12.3% for patients undergoing transplantation while in CR1 and 25.4% ± 15.5% for those undergoing transplantation in CR2 or a more advanced stage (P = .001). C, RR of patients undergoing allo-HSCT with an i.v. BU/CY regimen. The 30-month RR after allo-HSCT was 32.0% ± 12.7% for patients undergoing transplantation while in CR1 and 71.4% ± 17.1% in those undergoing transplantation in CR2 or a more advanced stage (P = .001).

Neutrophil engraftment (ie, absolute neutrophil count $\geq 0.5 \times 10^9$/L) occurred in all 42 patients, at a median of 13 days (range, 7~23 days), and the median time of platelet recovery (ie, $\geq 20 \times 10^9$/L) was 17 days (range, 9~61 days) in 41 patients. One patient died of VOD on day +29 before the documentation of platelet engraftment. The development of donor-derived hematopoiesis was further documented by short tandem repeat polymerase chain reaction, and all 41 evaluable patients achieved 100% donor type on day +28 to +30 posttransplantation.

OS and EFS

A total of 13 patients died during follow-up, for an overall estimated 30-month OS of 56.5% ± 10.6%. Patients who underwent transplantation while in CR1 had superior outcomes, with a 30-month OS of 65.7% ± 12.5% (median not reached), compared with 25.4% ± 15.5% (median, 6 months; P < .001) (Figure 1A) for patients who underwent transplantation while in CR2 or beyond. The overall 30-month EFS was 55.2% ± 10.4%; EFS was 63.4% ± 12.3% (median not reached) in patients undergoing transplantation while in CR1, compared with 25.4% ± 15.5% (median, 3 months) in patients in CR2 or beyond (P = .001) (Figure 1B).

Relapse after Allo-HSCT

Eleven patients relapsed at 2-26 months after allo-HSCT, with an overall accumulated 30-month RR of 40.0% ± 10.9%. Five of 9 patients who underwent transplantation while in CR2 or beyond relapsed (RR, 71.4% ± 17.1%), compared with 6 of 33 patients in CR1 (RR, 32.0% ± 12.7%) (Figure 1C). Among the patients in CR1, 5 relapse events occurred within the first 12 months posttransplantation (range, 2-10 months); only 1 patient relapsed at 26 months after transplantation from an unrelated donor with persistent chronic GVHD. All 5 patients undergoing all-HSCT while in a more advanced disease stage relapsed within 6 months posttransplantation. Of note, among all the relapse events, only 1 patient developed relapse in the testis, with subsequent bone marrow relapse 3 months after orchiectomy and radiation therapy.

Treatment-Related Toxicity and TRM

Fourteen of 41 evaluable patients developed grade II-IV aGVHD, with an overall incidence of 39.2% ± 8.8% (25.9% ± 11.7% for patients with a sibling donor and 50.2% ± 12.6% for those with an unrelated donor; P = .11). Only 3 patients had grade III-IV aGVHD, with an overall incidence of 7.4% ± 4.1% (5.6% ± 5.4% for patients with a sibling donor and 8.9% ± 6.0% for those with an unrelated donor; P = .67). During follow-up, the overall incidence of cGVHD in 33 evaluable patients was 63.9% ± 11.7% (77.1%

Table 2. Impact of Disease Stage on Outcome of Allo-HSCT

	CR1	≥CR2	P Value
Number of patients	33	9	
30-month OS	65.7% ± 12.5%	25.4% ± 15.5%	<.001
30-month EFS	63.4% ± 12.3%	25.4% ± 15.5%	.001
30-month RR	32.0% ± 12.7%	71.4% ± 17.1%	.001
30-month TRM	6.4% ± 4.4%	22.2% ± 13.9%	.13

± 12.8% for patients with an unrelated donor vs 49.3% ± 18.0% for those with a sibling donor; $P = .066$). The incidence of extensive cGVHD was 19.3% ± 7.9% (11.1% ± 10.5% for patients with a sibling donor vs 28.6% ± 12.1% for those with an unrelated donor; $P = .14$).

For conditioning regimen–related toxicity, we documented 1 case of mild VOD and 1 case of severe VOD, for an overall incidence of 4.7%. The other toxicity included 1 case of diffuse alveolar hemorrhage and 8 cases of hemorrhagic cystitis. Four patients died due to transplantation toxicity; of these, 3 died within 100 days posttransplantation, with an overall 30-month TRM of 9.7% ± 4.6%. The causes of death were VOD (n = 1), grade IV aGVHD (n = 1), diffuse alveolar hemorrhage (n = 1), and pulmonary infection after treatment for grade III aGVHD (n = 1).

Prognostic Factors

We further analyzed the potential prognostic factors associated with clinical outcome. Although based on a limited number of patients, disease stage at transplantation was significantly associated with OS, EFS, and RR, but not with TRM, on univariate analysis. For patients undergoing transplantation while in CR1, either donor type (sibling vs unrelated) or the presence of the Philadelphia chromosome was not associated with OS, EFS, RR, or TRM (Tables 2 and 3).

DISCUSSION

Analysis of results from large-scale clinical trials demonstrates the important role of allo-HSCT with HLA-matched sibling or unrelated donors in the treatment of adult ALL [22,23]. Combinations of CY with TBI or BU were the standard conditioning regimens for decades. For ALL, TBI/CY has remained a preferred regimen because of its benefit in

controlling "sanctuary site" leukemia and lower TRM compared with conventional oral BU/CY regimens [4,10]. A recent meta-analysis including more than 800 patients with ALL showed that oral BU/CY regimens were associated with lower EFS ($P < .001$), similar leukemia relapse ($P = .42$), and higher TRM than TBI/CY regimens [11]. The BU/CY regimen also was associated with higher rates of VOD and hemorrhagic cystitis [11]. It has been shown that BU systemic exposure in terms of area under the plasma concentration-versus-time curve (AUC) is associated with BU toxicity, manifested as gastrointestinal toxicity, hepatotoxicity, mucositis, and aGVHD with increasing AUC [24]. The risk of death was significantly lower for patients with a per-dose AUC of 950~1520 μMol/min compared with patients with either lower or higher AUC values [24]. The complete bioavailability of i.v. BU provides dose assurance by reducing interdose and interpatient variability, which may decrease the toxicity and TRM associated with oral formulations [5].

Kashyap et al. [15] reported a multiple-center study of patients undergoing allo-HSCT in which the incidence of clinically diagnosed VOD was 8% (5/61) in patients receiving i.v. BU and 33% (10/30) in those receiving oral BU. VOD-related mortality was 3.3% (n = 2) in the i.v. BU group and 20% (n = 6) in the oral BU group ($P = .001$). Oral BU was associated with higher 100-day TRM (33% vs 13%; $P = .02$). In a retrospective study of 236 patients with leukemia and myelodysplastic syndrome, aGVHD developed in 30.1% of patients overall, with no difference between the oral BU group (29.8%) and i.v. BU group (30.9%) [16]. Hepatic VOD was significantly more prevalent in the oral BU group (42.0% vs 18.2%; $P = .001$), as was severe VOD (6.1% vs 0). Other regimen-associated toxicities, including grade III-V gastrointestinal bleeding ($P = .004$), diarrhea ($P = .026$), coagulation abnormalities ($P = .007$), and metabolic abnormalities ($P = .042$), were significantly less prevalent in the i.v. BU group. The accumulated NRM was also lower in the i.v. BU group than in the oral BU group (13.8% vs 24.4%; $P = .048$). In a matched-pair analysis comparing outcomes in patients receiving i.v. BU/CY in 4 clinical trials with Center for International Blood and Marrow Transplant Research data for patients receiving oral BU, the overall incidence of VOD or

Table 3. Prognostic Analysis for Transplantation Outcome in Patients in CR1

	n	30-Month OS	30-Month EFS	30-Month RR	30-Month TRM
Donor type		14	19		
Sibling	14	81.8% ± 11.6%	72.7% ± 13.4%	26.7% ± 14.1%	0
Unrelated	19	55.5% ± 18.2%	55.5% ± 18.2%	37.5% ± 19.8%	11.2% ± 7.5%
Philadelphia chromosome status					
Ph⁻	26	59.3% ± 15.6%	59.3% ± 15.6%	35.7% ± 16.5%	8.2% ± 5.6%
Ph⁺	7	85.7% ± 13.2%	71.4% ± 17.1%	28.6% ± 17.1%	0

mortality in the first 28 days was 4.6% (4/83) in the i.v. BU patients and 20.3% (38/149) in the oral BU patients (*P* < .001). The 100-day TRM was 8.7% in the i.v. BU patients and 22.5% in the oral BU patients (*P* = .015). In logistic regression analysis, only oral BU administration was a significant risk factor for VOD [25].

Although the potential benefit of i.v. BU in the conditioning regimen is known, there is little data on direct comparisons of i.v. BU/CY regimens and oral BU/CY or TBI/CY regimens, particularly in patients with ALL. The present study was performed to evaluate the efficacy and toxicity of i.v. BU/CY as a conditioning regimen in adult ALL. We found that i.v. BU/CY conditioning was associated with promising survival (OS, 65.7% ± 12.5%; EFS, 63.4% ± 12.3%), suggesting that i.v. BU/CY may be an acceptable option in patients with ALL in CR1. This was due mostly to the low incidence of clinically diagnosed VOD (<5%) as well as the low VOD-related mortality and overall TRM (9.7%). The low incidence of VOD might be attributed to the inclusion of i.v. BU and prostaglandin E1 (PGE1) prophylaxis in our protocol [26]. Although there is no effective prophylactic treatment for VOD, several clinical trials have tested various prophylactic strategies, including low-dose heparin and PGE1. Some have reported that lipo-PGE1 is an effective prophylaxis for VOD [26,27]. A more recent study demonstrated that prophylactic lipo-PGE1 might not necessarily decrease the incidence of VOD but might reduce the severity of VOD; none of the 40 patients developed severe VOD, leading to a low 100-day TRM [28]. Based on these data, the efficacy of preventing VOD by replacing oral BU with i.v. BU combined with PGE1 prophylaxis can be considered confirmed in a clinical trial. Although we used standard doses of i.v. BU in the preparation regimen, another explanation for the low TRM may be the possible low AUC value of BU. Pharmacokinetic studies of i.v. BU from Korea [29] and Japan [30] found similar pharmacokinetic profiles of i.v. BU as have been reported in Western countries. Of even more interest, the Japanese study found high interpatient and intrapatient consistency of i.v. BU pharmacokinetics, which might possibly preclude the need for monitoring [30]. Unfortunately, there are no data available for i.v. BU in the Chinese population; based on the foregoing findings, studies of i.v. BU in the Chinese population are needed to provide key information on i.v. BU pharmacokinetics, which might help determine an ideal BU dose to ensure antileukemia efficacy and low toxicity, as demonstrated by several groups undergoing allo-HSCT with BU-based conditioning regimens [31-33].

Leukemia relapse remains the leading cause of failure of allo-HSCT [10]. The RR is similar with different conditioning regimens for ALL. In our series, leukemia relapse occurred in 32% of the patients who underwent transplantation while in CR1. Most relapses occurred within the first 12 months after allo-HSCT; only 1 patient relapsed at 26 months posttransplantation. For patients who underwent transplantation in a more advanced disease stage, all relapses occurred very early after transplantation. OS was ~65% for patients undergoing allo-HSCT while in CR1, compared with only 25% for those undergoing allo-HSCT in a more advanced disease stage. This observation suggests that for patients in CR1, i.v. BU/CY can be a feasible and effective conditioning regimen, with leukemia RRs comparable to previous reports, ranging from 22% to 37% in standard or high-risk ALL [22,23,34]. Given the relatively short follow-up in the present study, long-term observation is warranted to confirm the benefit of i.v. BU/CY regimens for patients undergoing allo-HSCT while in CR1. As for salvage therapy for patients with more advanced ALL, allo-HSCT is unlikely to benefit most patients, as has been shown in 2 large studies [35,36]. On the other hand, at least i.v. On the other hand, i.v. BU/CY was not an ideal regimen based on such a high relapse rate with extremely short median OS and EFS. Of note, relapse from an extramedullary sanctuary site has been documented in only 1 patient, thus indicated that the efficacy of controling sanctuary site leukemia by BU/CY might be comparable to TBI-based regimen.

The present study has revealed several important findings. First, an i.v. BU/CY regimen in adult ALL can be feasible with limited toxicity, particularly a low incidence of VOD when combined with prophylactic PGE1. Second, the overall TRM of i.v. BU/CY is low in patients in CR1, leading to improved OS. Third, i.v. BU/CY regimens are not suitable for advanced-stage ALL, because of a high relapse rate. Randomized trials are warranted to confirm the exact role of i.v. BU/CY conditioning regimens in allo-HSCT for adult ALL.

ACKNOWLEDGMENTS

REFERENCES

1. Fefer A, Einstein AB, Thomas ED, et al. Bone marrow transplantation for hematologic neoplasia in 16 patients with identical twins. *N Engl J Med.* 1974;290:1389-1393.
2. Santos GW, Tutschka PJ, Brookmeyer R, et al. Marrow transplantation for acute nonlymphocytic leukemia after treatment with busulfan and cyclophosphamide. *N Engl J Med.* 1983;309:1347-1353.
3. Tutschka PJ, Copelan EA, Klein JP. Bone marrow transplantation for leukemia following a new busulfan and cyclophosphamide regimen. *Blood.* 1987;70:1382-1388.

4. Ferry C, Socie G. Busulfan-cyclophosphamide versus total body irradiation–cyclophosphamide as a preparative regimen before allogeneic hematopoietic stem cell transplantation for acute myeloid leukemia: what have we learned? *Exp Hematol.* 2003;31:1182-1186.
5. Nath CE, Shaw PJ. Busulphan in blood and marrow transplantation: dose, route, frequency and role of therapeutic drug monitoring. *Curr Clin Pharmacol.* 2007;2:75-91.
6. Clift RA, Buckner CD, Thomas ED, et al. Marrow transplantation for chronic myeloid leukemia: a randomized study comparing cyclophosphamide and total body irradiation with busulfan and cyclophosphamide. *Blood.* 1994;84:2036-2043.
7. Devergie A, Blaise D, Attal M, et al. Allogeneic bone marrow transplantation for chronic myeloid leukemia in first chronic phase: a randomized trial of busulfan-cytoxan versus cytoxan-total body irradiation as preparative regimen: a report from the French Society of Bone Marrow Graft (SFGM). *Blood.* 1995;85:2263-2268.
8. Kim I, Park S, Kim BK, et al. Allogeneic bone marrow transplantation for chronic myeloid leukemia: a retrospective study of busulfan-cytoxan versus total body irradiation-cytoxan as preparative regimen in Koreans. *Clin Transplant.* 2001;15:167-172.
9. 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.
10. Avivi I. Stem cell transplantation in adult ALL patients. *Best Pract Res Clin Haematol.* 2003;15:653-674.
11. Xu SX, Tang XH, Xu HQ, et al. Total body irradiation plus cyclophosphamide versus busulphan with cyclophosphamide as conditioning regimen for patients with leukemia undergoing allogeneic stem cell transplantation: a meta-analysis. *Leuk Lymphoma.* 2010;51:50-60.
12. Grochow LB, Jones RJ, Brundrett RB, et al. Pharmacokinetics of busulfan: correlation with veno-occlusive disease in patients undergoing bone marrow transplantation. *Cancer Chemother Pharmacol.* 1989;25:55-61.
13. Hassan M, Oberg G, Ehrsson H, et al. Pharmacokinetic and metabolic studies of high-dose busulphan in adults. *Eur J Clin Pharmacol.* 1989;36:525-530.
14. Davies SM, Ramsay NK, Klein JP, et al. Comparison of preparative regimens in transplants for children with acute lymphoblastic leukemia. *J Clin Oncol.* 2000;18:340-347.
15. Kashyap A, Wingard J, Cagnoni P, et al. Intravenous versus oral busulfan as part of a busulfan/cyclophosphamide preparative regimen for allogeneic hematopoietic stem cell transplantation: decreased incidence of hepatic veno-occlusive disease (HVOD), HVOD-related mortality, and overall 100-day mortality. *Biol Blood Marrow Transplant.* 2002;8:493-500.
16. Kim S, Lee J, Choi S, et al. Allogeneic bone marrow transplantation in adult leukemia patients conditioned with busulfan plus cyclophosphamide: a retrospective comparison of oral versus intravenous busulfan. *Haematologica.* 2005;90:285-286.
17. Fernandez H, Tran H, Albrecht F, et al. Evaluation of safety and pharmacokinetics of administering intravenous busulfan in a twice-daily or daily schedule to patients with advanced hematologic malignant disease undergoing stem cell transplantation. *Biol Blood Marrow Transplant.* 2002;8:486-492.
18. Carreras E, Bertz H, Arcese W, et al. Incidence and outcome of hepatic veno-occlusive disease after blood or marrow transplantation: a prospective cohort study of the European Group for Blood and Marrow Transplantation. *Blood.* 1998;92:3599-3604.
19. Glucksberg H, Storb R, Fefer A, et al. Clinical manifestations of graft-versus-host disease in human recipients of marrow from HLA-matched sibling donors. *Transplantation.* 1974;18:295-304.
20. 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.
21. Lee ET, editor. *Statistical Methods for Survival Data Analysis.* Belmont, CA: Lifetime Learning; 1980 p. 355.
22. Goldstone AH, Richards SM, Lazarus HM, et al. In adults with standard-risk acute lymphoblastic leukemia, the greatest benefit is achieved from a matched sibling allogeneic transplantation in first complete remission, and an autologous transplantation is less effective than conventional consolidation/maintenance chemotherapy in all patients: final results of the International ALL Trial (MRC UKALLXII/ECOG E2993). *Blood.* 2008;111:1827-1833.
23. Nishiwaki S, Inamoto Y, Sakamaki H, et al. Allogeneic stem cell transplantation for adult Philadelphia chromosome–negative acute lymphocytic leukemia: comparable survival rates but different risk factors between related and unrelated transplantation in first complete remission. *Blood.* 2010;116:4368-4375.
24. Andersson BS, Thall PF, Madden T, et al. Busulfan systemic exposure relative to regimen-related toxicity and acute graft-versus-host disease: defining a therapeutic window for intravenous BuCy2 in chronic myelogenous leukemia. *Biol Blood Marrow Transplant.* 2002;8:477-485.
25. Sobocinski KA, Thall PF, Bekele BN, et al. Matched pairs analysis of IV vs PO busulfan as a conditioning agent prior to transplantation. ASH Annual Meeting abstract. *Blood.* 2004;104:349.
26. Gluckman E, Jolivet I, Scrobohaci ML, et al. Use of prostaglandin E1 for prevention of liver veno-occlusive disease in leukaemic patients treated by allogeneic bone marrow transplantation. *Br J Haematol.* 1990;74:277-281.
27. Cho B, Sung IK, Kim DW, et al. A trial use of prophylactic low-dose lipo PGE1 (Eglandin) for the prevention of hepatic veno-occlusive disease after hematopoietic stem cell transplantation in children with hematologic malignancies. *Korean J Pediatr Hematol Oncol.* 2000;7:242-248.
28. Song JS, Seo JJ, Moon HN, et al. Prophylactic low-dose heparin or prostaglandin E1 may prevent severe veno-occlusive disease of the liver after allogeneic hematopoietic stem cell transplantation in Korean children. *J Korean Med Sci.* 2006;21:897-903.
29. Cho YH, Lim HA, Lee MH, et al. Pharmacokinetics and tolerability of intravenous busulfan in hematopoietic stem cell transplantation. *Clin Transplant.* 2007;21:417-422.
30. Kim SW, Mori S, Tanosaki R, et al. Busulfex (iv BU) and CY regimen before SCT: Japanese-targeted phase II pharmacokinetics combined study. *Bone Marrow Transplant.* 2009;43:611-617.
31. Deeg HJ, Storer B, Slattery JT, et al. Conditioning with targeted busulfan and cyclophosphamide for hemopoietic stem cell transplantation from related and unrelated donors in patients with myelodysplastic syndrome. *Blood.* 2002;100:1201-1207.
32. Bornhäuser M, Storer B, Slattery JT, et al. Conditioning with fludarabine and targeted busulfan for transplantation of allogeneic hematopoietic stem cells. *Blood.* 2003;102:820-826.
33. Bartelink IH, Bredius R, Belitser SV, et al. Association between busulfan exposure and outcome in children receiving intravenous busulfan before hematologic stem cell transplantation. *Biol Blood Marrow Transplant.* 2009;15:231-241.
34. Cornelissen JJ, van der Holt B, Verhoef G, et al. Myeloablative allogeneic versus autologous stem cell transplantation in adult patients with acute lymphoblastic leukemia in first remission: a prospective sibling donor versus no-donor comparison. *Blood.* 2009;113:1375-1382.
35. Fielding AK, Richards SM, Chopra R, et al. Outcome of 609 adults after relapse of acute lymphoblastic leukemia (ALL): an MRC UKALL12/ECOG 2993 study. *Blood.* 2007;109:944-950.
36. Reman O, Buzyn A, Lheritier V, et al. Rescue therapy combining intermediate-dose cytarabine with amsacrine and etoposide in relapsed adult acute lymphoblastic leukemia. *Hematol J.* 2004;5:123-129.