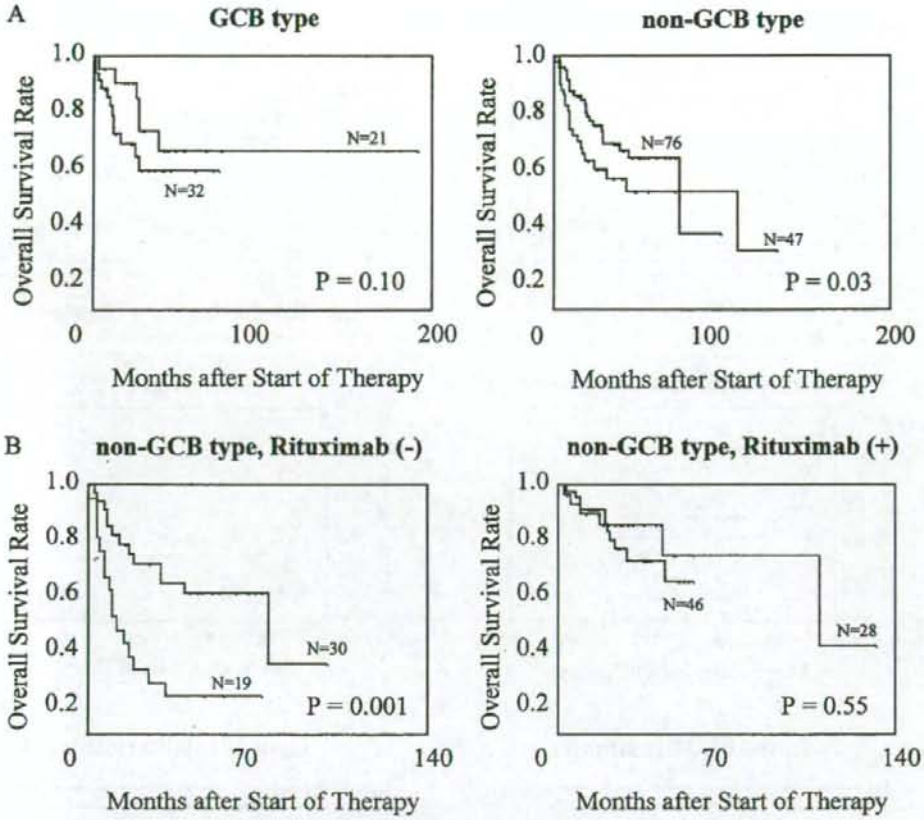

Figure 4. A: Overall survival curves using the Kaplan-Meier method are shown according to anamorsin expression levels in GCB type or non-GCB type. (—: weak or negative anamorsin expression group, ═: strong anamorsin expression group.) In the GCB type, no significant difference of overall survival rate is shown by the log-rank test ($p = 0.10$); however in the non-GCB type, a significant difference is shown ($p = 0.03$). B: Overall survival curves using the Kaplan-Meier method are shown according to anamorsin expression levels within the non-GCB type. (—: weak or negative anamorsin expression group, ═: strong anamorsin expression group.) A significant difference of overall survival rate is shown by the log-rank test in the group receiving chemotherapy only [Rituximab(−)] ($p = 0.001$), while no significant difference is shown in the group receiving combined immunochemotherapy [Rituximab(+)] ($p = 0.55$).

play significant roles in tumor cell growth and survival of DLBCL. Both molecules were also evaluated as prognostic biomarkers of DLBCL: BCL2 indicates an unfavorable prognosis, while BCL6 indicates a more positive prognosis [20,21]. Several new drugs that target each of these molecules have been developed for the treatment of lymphoma including DLBCL [22,23]. Since the growth of anamorsin-null cells is markedly reduced, drugs targeted against anamorsin may be useful for treatment of DLBCL in patients who show abundant anamorsin expression.

The level of anamorsin expression indicated by immunostaining paraffin-embedded sections predicted the outcome for patients with DLBCL. To explore other clinical applications for anamorsin measurement, we will use the quantitative reverse transcriptase polymerase chain reaction (RT-PCR) to try to establish a quantitative measurement of anamorsin mRNA. Measurement of expression levels of mRNA for Wilm's tumor gene (WT1), measured by quantitative RT-PCR, was proven a useful indicator of minimal residual disease (MRD) in leukemia and myelodysplastic syndrome [24]. Anamorsin expression levels as determined by quantitative RT-PCR will be used to evaluate infiltration of tumor cells with strong anamorsin expression into bone marrow or peripheral blood, and monitoring MRD after treatment. Further study will be necessary to elucidate the precise mechanisms of how anamorsin contributes to the abnormal growth of DLBCL.

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Bone Marrow Transplantation (2008), 1–7

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ORIGINAL ARTICLE

Busulfex (i.v. BU) and CY regimen before SCT: Japanese-targeted phase II pharmacokinetics combined study

S-W Kim[1], S-i Mori[1], R Tanosaki[1], T Fukuda[1], M Kami[2], H Sakamaki[3], T Yamashita[3], Y Kodera[4], S Terakura[5], S Taniguchi[6], S Miyakoshi[7], N Usui[8], S Yano[8], Y Kawano[9], Y Nagatoshi[10], M Harada[11], Y Morishima[12], S Okamoto[13], AM Saito[14,15], Y Ohashi[15], R Ueda[16] and Y Takaue[1]

[1]*Hematology and Hematopoietic Stem Cell Transplantation Division, National Cancer Center Hospital, Tokyo, Japan;* [2]*Division of Exploratory Research, Institute of Medical Science, University of Tokyo, Tokyo, Japan;* [3]*Department of Hematology, Komagome Hospital, Tokyo, Japan;* [4]*Department of Hematology, Japanese Red Cross Nagoya First Hospital, Nagoya, Japan;* [5]*Department of Hematology and Oncology, Nagoya University Graduate School of Medicine, Nagoya, Japan;* [6]*Department of Hematology, Toranomon Hospital, Tokyo, Japan;* [7]*Department of Hematology, Tokyo Metropolitan Geriatric Hospital, Tokyo, Japan;* [8]*Division of Clinical Oncology and Hematology, Department of Internal Medicine, Jikei University School of Medicine, Tokyo, Japan;* [9]*Department of Pediatrics, Graduate School of Medical and Dental Sciences, Kagoshima University, Kagoshima, Japan;* [10]*Section of Pediatrics, National Kyushu Cancer Center, Fukuoka, Japan;* [11]*National Omuta Hospital, Omuta, Japan;* [12]*Department of Hematology and Cell Therapy, Aichi Cancer Center Hospital, Nagoya, Japan;* [13]*Division of Hematology, Department of Medicine, Keio University School of Medicine, Tokyo, Japan;* [14]*Department of Hematology and Oncology, Graduate School of Medicine, University of Tokyo, Tokyo, Japan;* [15]*Department of Biostatistics/Epidemiology and Preventive Health Sciences, School of Health Sciences and Nursing, University of Tokyo, Tokyo, Japan and* [16]*Department of Internal Medicine and Molecular Science, Nagoya City University Graduate School of Medical Sciences, Nagoya, Japan*

To evaluate the toxicity and efficacy of an i.v. preparation of BU (12.8 mg/kg), combined with CY (120 mg/kg), a prospective study was performed on 30 Japanese patients (median age, 30 years) with hematologic malignancies undergoing hematopoietic SCT (28 allogeneic transplants from an HLA-matched donor and 2 autologous transplants). There were no significant toxicities, and all but one patient showed evidence of granulocyte engraftment at a median of 14 days for allogeneic and 11 days for autologous transplantation. Grades II–IV acute and chronic GVHD occurred in 9 (9/27, 33%) and 16 patients (16/27, 59%), respectively. Non-relapse mortality at days 100 and 365 was 3 and 17%, respectively. The pharmacokinetics of i.v. BU showed close inter- and intrapatient consistency; the area under the plasma concentration–time curve of the first administration remained at less than 1500 μmol min/l in 27 of the 29 patients (93%), and between 900 and 1350 μmol min/l in 22 patients (73%). As all of the profiles overlap with data from non-Japanese patients, we conclude that racial factors may not seriously influence the bioactivity of i.v. BU.

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Keywords: busulfex; BU; hematologic disease

Correspondence: Dr Y Takaue, Hematology and Hematopoietic Stem Cell Transplantation Division, National Cancer Center Hospital, 5-1-1 Tsukiji, Chuo-Ku Tokyo 104-0045, Japan.
E-mail: ytakaue@ncc.go.jp
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Introduction

In hematopoietic SCT (HSCT), high-dose BU has been widely used, mostly in combination with CY.[1] To overcome the disadvantage of oral BU including gastrointestinal absorption,[2–16] i.v. BU was recently introduced into clinical use.[17–20] The initial experience with i.v. BU showed satisfactory dose assurance with reliable predictability of pharmacokinetics without dose adjustment.[19] Hence, it is very probable that its use reduces the incidence of various risks at transplantation such as hepatic venoocclusive disease (VOD), as shown by Kashyap *et al.*[21]

Nevertheless, drug profiles of i.v. BU preparation have not been fully evaluated in different races, who may have different pharmacokinetics. As part of our pivotal study in Japan, we conducted a phase II study with pharmacokinetic analysis of a combined i.v. BU and CY (BU/CY) regimen administered before allogeneic or autologous HSCT. A population pharmacokinetic analysis suggested that i.v. BU pharmacokinetics show high inter- and intrapatient consistency.[22] This study with the same population further focused on complete pharmacokinetic profiles with additional clinical and safety data.

Patients and methods

Eligibility criteria

Patients with acute leukemia, CML, MDS or malignant lymphoma were eligible for this study. Patients aged 5–55 years with a Lansky Performance Status >70 (over 5 and less than 16 years of age) or an Eastern Cooperative Oncology Group Performance Status ≤2 (16–55 years of

age) who were expected to survive beyond 100 days after HSCT were eligible. The eligibility criteria also included serum creatinine less than twice the upper normal limit, as well as serum total bilirubin less than 1.5 times, and aspartate aminotransferase, alanine aminotransferase and gamma-glutamyltranspeptidase less than three times the upper normal limit. Left ventricular ejection fraction $\geqslant$50% or arterial blood oxygen saturation $\geqslant$94%, and in adult patients a carbon monoxide lung diffusing capacity $\geqslant$60%, were required. Patients with arrhythmia, hypertension or diabetes mellitus that was difficult to control despite medication, severe cardiopulmonary or renal disease, chronic active hepatitis, liver cirrhosis, acute hepatitis, ascites more than 1 l, central nervous system disorders, active infection; positive hepatitis B surface antigen, hepatitis B core antibody, hepatitis C virus antibody or human immunodeficiency virus antigen/antibody; or prior HSCTs were all excluded. Patients were also required to have either BM available from an HLA-matched related or unrelated donor or G-CSF-mobilized PBSCs available from an HLA-matched related donor without T-cell depletion. The study was conducted in conformity with ICH-GCP and the Declaration of Helsinki. The protocol and informed consent forms were approved by each institution's Research Ethics Committee. All patients gave written informed consent prior to their participation in the study.

Conditioning regimen

The i.v. BU (KRN246; Kirin Pharma Co. Ltd., Tokyo, Japan) was given at 0.8 mg/kg through a central venous catheter for 2 h every 6 h at a total of 16 doses for 4 days on days −7 to −4. CY 60 mg/kg was administered through a central venous catheter for 3 h at a total of two doses for 2 days on days −3 and −2. After a rest on day −1, BM or G-CSF-mobilized PBSC without T-cell depletion was infused on day 0. A fixed-dose regimen for BU was calculated based on either the ideal body weight or actual body weight, whichever was less, for adults (18–55 years of age) and the actual body weight for children (over 5 and less than 18 years of age).

Supportive care

For seizure prophylaxis, phenytoin was administered at 5–10 mg/kg/day (upper limit of 300 mg/kg/day) in 2–3 divided doses starting from 2 days before initiation (day −9) to 48 h after completion of BU administration (day −2). G-CSF was administered on day 1 or 5 until engraftment. For patients undergoing allogeneic HSCT, GVHD prophylaxis consisted of CYA (3 mg/kg/day by continuous i.v. infusion from day −1 in related and 3–5 mg/kg/day in unrelated transplantation) and short-term methotrexate, that is, 10 mg/m^2 on day 1 and 7 mg/m^2 on days 3 and 6 in related pairs or 10 mg/m^2 on day 1 and 7 mg/m^2 on days 3, 6 and 11 in unrelated pairs. Mesna was administered at a dose equivalent to 120% of CY on days −3 and −2. Other supportive treatments including antiemetic administration, antibiotic treatment, transfusion support, GVHD treatment and VOD treatment were given according to the standards of each hospital.

Evaluation of clinical data

The efficacy variables were myeloablation, engraftment, relapse, overall survival (OS) and disease-free survival (DFS). The safety variables were non-relapse mortality and adverse events included convulsive seizure, VOD, acute GVHD and other organ toxicities. Engraftment was defined as an absolute neutrophil count of 0.5×10^9/l for three consecutive days. Engraftment failure was defined as the failure to reach an absolute neutrophil count of 0.5×10^9/l by day 28 after transplantation. OS was measured as the time from the day of transplantation until death from any cause, and DFS as the time from the day of transplantation until disease relapse or death from any cause. Relapse, OS and DFS were calculated using the Kaplan–Meier method.[23] non-relapse mortality was defined as any death without progression of the underlying disease. Patients were monitored daily for adverse events, hematology and transplant-related complications. After discharge, patients were followed weekly for adverse events and transplant-related complications, and monitored weekly for hematologic and biochemical data through 100 days after transplantation. The appearance of VOD by day 30 was evaluated based on any two of the major criteria as established by McDonald *et al.*[24] and Jones *et al.*[25] GVHD was graded according to the consensus criteria.[26,27] Kirin Pharma Co. Ltd. provided financial support for the medical costs associated with the conditioning regimen, including i.v. BU for enrolled patients, monitored source data and entered these data in a database. Statistical analysis was performed using SAS software (version 8.02; SAS Institute, Cary, NC, USA).

PK sampling and analysis

The objective of this study was to describe the PK characteristics of i.v. BU, with parameters including BU concentrations for the first and ninth administrations and the accumulation of i.v. BU. Plasma samples were collected from all patients at designated times, in conjunction with the first and ninth doses as follows: immediately before drug infusion and at 15, 30 and 45 min after the start of infusion, at 5 min before the end of infusion and at 15, 30, 60, 120, 180 and 240 min after completion of infusion. In addition, one sample was taken immediately before the 13th infusion and 5 min before its completion. The plasma was assayed using a gas chromatographic-mass spectrometric detection method.[10]

Plasma concentrations for first and ninth dose in individual subjects were analyzed by the non-compartmental method using WinNonlin (version 3.3; Pharsight Corp., Mountain View, CA, USA). The maximum plasma concentration (C_{max}) and the time to reach maximum plasma drug concentration (t_{max}) were observed values. The terminal half-life ($t_{1/2}$) was calculated as $\ln 2/k_{el}$, where k_{el} was the elimination rate constant, determined by log-linear regression of the terminal phase data points. The area under the plasma concentration–time curve from time 0 to infinity (AUC_{inf}) for the first dose was calculated as $AUC_{0-t} + C_t/k_{el}$, where AUC_{0-t} was the AUC from time 0 to the last detectable time, calculated using linear trapezoidal rule, and C_t was the plasma concentration at

the last detectable time. AUC at steady state (AUC_{ss}) for the ninth dose was calculated by the linear trapezoidal rule. Clearance (CL) was calculated as dose/AUC. Volume of distribution (V_z) was calculated as CL/k_{el}. CL and V_z were normalized to actual individual body weight (CL/ABW and V_z/ABW) on the day of dosing. Summary statistics were obtained for C_{max}, t_{max}, $t_{1/2}$, AUC, CL/ABW and V_z/ABW at the first and ninth dose. The AUC at dose 1 (AUC_{inf}) and dose 9 (AUC_{ss}) and the trough concentration ($C_{p,trough}$) and peak concentration ($C_{p,peak}$) at doses 9 and 13 were calculated and compared by preparing each plot.

Results

Patient characteristics

Thirty Japanese patients were registered in this prospective trial between July 2002 and October 2003. The disease characteristics and status at transplantation are given in Table 1. The median age of the patients was 30 years (range, 7–53 years). The median body mass index (BMI) was 22.65 (14.4–29.1), and the mean BMI was 22.32 ± 3.47. There were no patients with moderate or severe obesity (BMI <30). The diseases were AML in 13 patients (43%), ALL or CML in chronic phase in five patients each (17%), non-Hodgkin lymphoma (NHL) in four patients (13%) and MDS in three patients (10%). In total, 11 of the 12 patients with AML were in CR. Four of the five patients with ALL were in CR. Three patients with MDS included refractory anemia, refractory anemia with excess blasts and refractory anemia with excess blasts in transformation. Four patients with NHL included diffuse large B-cell lymphoma in CR ($n=2$), primary refractory peripheral T-cell lymphoma ($n=1$) or suspected extranodal marginal zone B-cell lymphoma of mucosa-associated lymphoid tissue in CR ($n=1$). One patient with AML who was in remission at registration was subsequently withdrawn from protocol treatment due to onset of cardiac myopathy on day −3, and CY was changed to fludarabine. Owing to an additional protocol violation, this patient was excluded from the objective group in the analysis.

Engraftment

Twenty-eight patients (97%) achieved engraftment at a median of 14 days (range, 9–20 days) and 11 days after allogeneic and autologous HSCT, respectively (Table 2). One patient who received unrelated BMT for CML had graft failure. No secondary engraftment failure was observed.

Toxicity and complications

All adverse events were those that are commonly observed in HSCT and no characteristic events related to i.v. BU were observed. None of the patients had to interrupt i.v. BU treatment because of adverse events. The number of observed adverse events was 714 in 27 patients who received allogeneic HSCT and 19 in two patients who received autologous HSCT. The most frequent adverse events in the 27 allogeneic HSCT patients were vomiting and nausea in 20 patients each (74%), anorexia in 19 patients (70%), stomatitis and diarrhea in 18 patients each (67%) and headache in 17 patients (63%; Table 2). Both of the autologous HSCT patients showed stomatitis, vomiting, catheter-related infection, anorexia and dysgeusia. No seizures were observed, and with regard to other neuropsychological profiles, seven patients experienced mild dysgeusia, one moderate systemic burning sensation, one severe tremor, one severe mood change and one severe insomnia in an allogeneic setting. With regard to cardiovascular profiles, one patient experienced mild cardiac failure and the other developed moderate cardiomyopathy due to CY in the allogeneic setting, as described above. This patient had completed i.v. BU administration for 4 days and CY once. When the patient complained of chest discomfort, the heart rate was 101 beats/min, and her electrocardiography showed ST depressions in leads II, III, aVF and V_1–V_6 1 h after the completion of the first dose of CY, which made suspected diagnosis of CY-induced cardiomyopathy. The signs and symptoms subsided shortly, and the second dose of CY on day −2

Table 1 Patient characteristics

Variables	n *(%)*	
	Allogeneic HSCT (n=28)	*Autologous HSCT (n=2)*
Patient age (years) (range, median)	7–53, 30	48–50, 49
5–17	3 (11)	0
18–49	20 (71)	1 (50)
50–55	5 (18)	1 (50)
Gender		
Men	18 (64)	2 (100)
Women	10 (36)	0
Disease		
AML	12 (43)	1 (50)
ALL	5 (18)	0
CML	5 (18)	0
Myelodysplastic syndrome	3 (11)	0
Non-Hodgkin lymphoma	3 (11)	1 (50)
Disease status		
CR, CP, RA	23 (82)	2 (100)
NR, RAEB, RAEB-t	5 (18)	0
Prior chemotherapy	26 (93)	2 (100)
Prior radiotherapy	2 (7)	0
Source of stem cells		
BM	18 (64)	0
Peripheral blood cells	10 (36)	2 (100)
Related or unrelated donor		
Related	19 (68)	NA
Unrelated	9 (32)	NA
Cell dose infused		
Nucleated ($\times 10^8$/kg, median, range)	2.6 (0.7–4.4)	NA
CD34 positive ($\times 10^6$/kg, median, range)	2.7 (2.1–6.3)	2.9 (2.7–3.1)

Abbreviations: CP = chronic phase; HSCT = hematopoietic SCT; NA = not applicable; NR = non-remission; RA = refractory anemia; RAEB = refractory anemia with excess of blasts; RAEB-t = refractory anemia with excess of blasts in transformation.

Table 2 Regimen-related toxicity, engraftment, GVHD and death

Outcome	*Allogeneic HSCT (n=28) (%)*	*Autologous HSCT (n=2) (%)*
Toxicity		
Vomiting	21 (75)	2 (100)
Nausea	21 (75)	1 (50)
Anorexia	19 (68)	2 (100)
Stomatitis	18 (64)	2 (100)
Diarrhea	18 (64)	0 (0)
Headache	18 (64)	0 (0)
Seizure	0 (0)	0 (0)
VOD	1 (4)	0 (0)
	Allogeneic HSCT (n=27) (%)	*Autologous HSCT (n=2) (%)*
Engraftment	26 (96)	2 (100)
Median (days)	14	11
Range (days)	9–20	11
Graft failure	1 (4)	0 (0)
Acute GVHD	13 (48)	—
Grade I	4 (15)	—
Grade II	5 (19)	—
Grade III	2 (7)	—
Grade IV	2 (7)	—
Chronic GVHD	16 (59)	—
Death	8 (30)	0 (0)
Relapse	4 (15)	0 (0)
Non-relapse	4 (15)	0 (0)

Abbreviations: HSCT = hematopoietic SCT; VOD = venoocclusive disease.

was substituted by fludarabine with no subsequent complications.

One patient who received allogeneic HSCT was diagnosed with mild VOD on day 1 based on two diagnostic criteria,[24,25] which resolved on day 3. In another patient, elevated total bilirubin and body weight gain were found on days 60–69, and this was not confirmed to be VOD based on these criteria. Opportunistic infection occurred in 16 of 27 patients (59%), with a median onset of day 113 (range, 7–399). Pulmonary complications occurred in 7 of 27 patients (26%), with a median onset of day 149 (range, 65–335).

GVHD

Acute GVHD occurred in 13 of the 27 patients (48%) who received allogeneic HSCT; four (15%) had grade I, five (19%) grade II and two each (7%) grades III or IV (Table 2). Acute GVHD was documented in 7 of the 19 patients (37%) who received related transplantation (six had grades II–IV), and in six of the eight patients (75%) who received unrelated transplantation (three patients had grades II–IV). Acute GVHD occurred with a median onset of day 45 (range, 7–98). Chronic GVHD occurred in 16 of 27 patients (59%) with a median onset of day 133 (range, 39–239).

Causes of death

Four patients (15%) died of non-relapse causes (Table 2). One patient who received allogeneic HSCT died of multi-organ failure due to aggravated GVHD on day 69. Three patients who received allogeneic HSCT died of chronic GVHD on day 223, hepatic failure due to unknown reasons on day 266 (with extensive chronic GVHD and methicillin-resistant *staphylococcus aureus* (MRSA) pneumonia) and pneumonia due to adenovirus and cytomegalovirus on day 124. Four patients (15%) died of relapse.

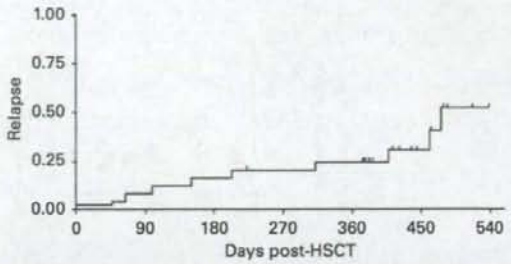

Figure 1 Disease relapse after i.v. BU and CY prior to allogeneic hematopoietic SCT in patients with leukemia and lymphoma.

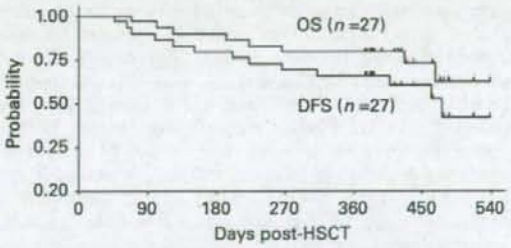

Figure 2 Overall survival and disease-free survival after i.v. BU and CY prior to allogeneic hematopoietic SCT in patients with leukemia, myelodysplastic syndrome and lymphoma.

Relapse and survival

Relapse occurred in 9 of the 23 evaluable allogeneic HSCT patients with leukemia and lymphoma (39%). None of the 23 evaluable patients had central nervous system relapse. The relapse rates at days 100 and 365 were 18% (95% confidence interval (CI), 0–38%) and 26% (95% CI, 8–45%), respectively (Figure 1). The median day of relapse was day 202 (range, 46–476).

OS at days 100 and 365 in allogeneic HSCT was 96% (95% CI, 88–100%) and 78% (95% CI, 62–94%), respectively, with the median follow-up of 413 days (range, 69–537 days) (Figure 2). The median day of death in eight allogeneic HSCT patients was day 208 (range, 69–467). DFS at days 100 and 365 in allogeneic HSCT was 81% (95% CI, 63–99%) and 63% (95% CI, 45–81%), respectively (Figure 2). The two autologous HSCT patients were alive disease-free at day 365.

PK analysis

Intensive PK sampling was assessed at doses 1 and 9 of i.v. BU, and peak and trough levels were obtained at dose 13. Although these analyses were completed in all 30 patients,

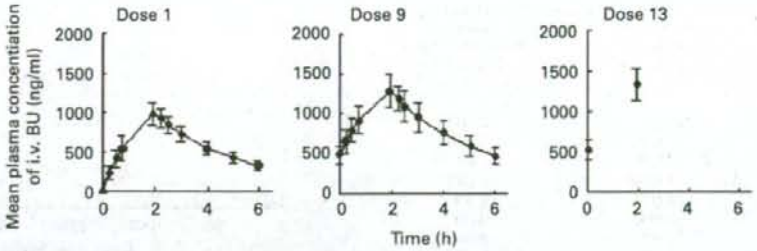

Figure 3 Pharmacokinetic results of i.v. BU at doses 1, 9 and 13 ($n=30$).

data from one patient were excluded from the objective analysis group as noted above. All PK parameters for dose 1 were obtained from 29 patients. For dose 9, all PK parameters except for C_{max} and t_{max} were obtained from 28 patients because the last sample for one patient was collected after initiation of the next dose (Figure 3). The documented plasma concentration of i.v. BU increased over the 2-h period of infusion, with C_{max} observed in the last 5 min, and this was followed by a rapid decrease. The profile of trough and peak levels was essentially the same between doses 9 and 13.

The resulting parameters are listed in Table 3. The mean AUC for doses 1 and 9 was 1171 μmol min/l (coefficient of variation (CV) = 19%) and 1242 μmol min/l (CV = 17%), and the mean C_{max} was 994 ng/ml (CV = 12%) and 1311 ng/ml (CV = 15%), respectively. The mean CL/ABW was 2.66 ml/min/kg (CV = 17%) and 2.46 ml/min/kg (CV = 15%), respectively. V_z/ABW was 0.601/kg (CV = 9%) and 0.601/kg (CV = 11%), respectively. The AUC of the initial dose was below 1500 μmol min/l in 27 patients (90%), and this was within the range of 900–1350 μmol min/l in 21 of the 29 patients (72%).

The AUC for doses 1 and 9 are compared in Figure 4, which supports both intra- and interpatient predictability and consistency. In the patient who developed VOD, the AUC for doses 1 and 9 was 1102 and 1181 μmol min/l, respectively, whereas for the remaining patients without VOD, it was 1173 μmol min/l (CV = 19%) and 1244 μmol min/l (CV = 17%).

Pediatric patients

A 7-year-old girl with AML in first remission received allo-BMT from a matched unrelated donor. Her body weight and BMI were 17.8 kg and 14.4, respectively. Her AUC was 963.9 μmol min/l. Her regimen-related toxicities were grade 3 vomiting and grade 2 acute hemorrhagic gastritis and hypoalbuminemia. She is alive without graft failure or relapse.

A 13-year-old boy with CML in first chronic phase received allo-BMT from a matched unrelated donor. His body weight and BMI were 46.7 kg and 18.8, respectively. His AUC was 932.6 μmol min/l. His regimen-related toxicities were grade 4 anorexia and grade 2 fatigue and vomiting. He did not achieve engraftment by day 28, and he soon received a second allo-BMT from a mismatched

Table 3 Pharmacokinetics of i.v. BU ($n=30$*)

	C_{max} (ng/ml)	$t_{1/2}$ (h)	AUC (μmol min/l)	CL/ABW (ml/min/kg)	V_z/ABW (l/kg)
Dose 1					
Mean	999	2.64	1171	2.67	0.596
Median	997	2.66	1144	2.65	0.596
s.d.	124	0.41	216	0.44	0.054
Maximum	1320	3.52	1698	3.72	0.716
Minimum	796	1.97	811	1.94	0.483
Dose 9					
Mean	1317	2.86	1247	2.46	0.601
Median	1315	2.82	1198	2.36	0.605
s.d.	192	0.37	205	0.36	0.068
Maximum	1720	3.59	1686	3.05	0.786
Minimum	964	2.27	889	1.80	0.466

Abbreviations: ABW = actual body weight; AUC = area under the plasma concentration–time curve; CL = clearance; C_{max} = maximum plasma concentration; s.d. = standard deviation; $t_{1/2}$ = terminal half-life; t_{max} = time to observed maximum plasma concentration from dosing; V_z = volume of distribution.

*For dose 9, all PK parameters except for C_{max} and t_{max} were obtained from 29 patients because the last sample for one patient was collected after initiation of the next dose.

For dose 1, AUC_{inf} is shown; for dose 9, AUC_{ss} for the 6-h dosing interval is presented.

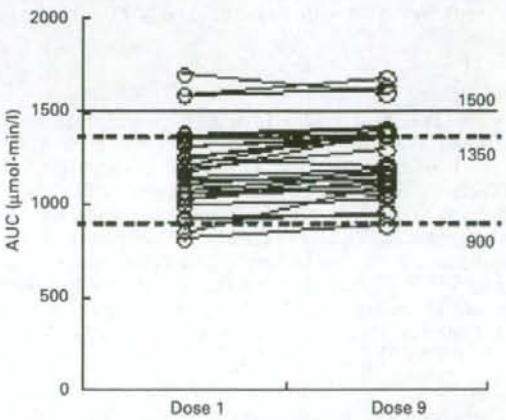

Figure 4 Individual patient area under the plasma concentration–time curve (AUC) values of i.v. BU at doses 1 and 9 ($n=29$).

related donor. He is alive without graft failure or relapse after the second transplant.

A 17-year-old woman with AML in first relapse received allo-BMT from a matched unrelated donor. Her body weight and BMI were 43.2 kg and 17.3, respectively. Her AUC was 902.7 μmol min/l. Her regimen-related toxicities were grade 4 thrombocytopenia, grade 3 febrile neutropenia and grade 2 nausea, vomiting and stomatitis. She died of disease progression on day 193.

Discussion

It has been reported that a high steady-state concentration of BU causes toxicities including VOD,[5–10] whereas a low steady-state concentration leads to graft rejection[10–15] or relapse/progression of the disease.[11] Targeted dose adjustment of BU to maintain the overall systemic exposure within a proper range may reduce these risks.[4–7,14,15] Although it has been reported that there are ethnic differences in PK for a wide range of drugs,[28] this has not been seriously examined with i.v. BU. Therefore, we conducted this drug bioavailability study in a Japanese population. The data obtained were compared with those published mostly overseas. In this study, all observed treatment-related toxicities were as expected, with a low incidence of severe complications. One patient was clinically diagnosed with VOD. This patient showed body weight gain, liver enlargement and right upper abdominal pain, but had no jaundice. As his body weight returned to the baseline within 2 days, this could have been due to overhydration. One patient who developed graft failure had CML and underwent unrelated BMT following interferon therapy, all of which are well-known risks of graft failure.[10,29] The incidence of relapse and the survival rate in this study were similar to those in previous studies.[11,19]

In studies with an oral preparation of BU, it was unclear whether plasma levels of BU correlate with severe regimen-related toxicities.[4,6–8,11] In the pivotal study for US approval of i.v. BU, plasma levels of BU exceeded 1500 μmol min/l in two of the five patients who developed VOD,[19] whereas in our study there was no case of VOD in three patients who had a level over 1500 μmol min/l. This may suggest an ethnic difference in the PK of BU. On the other hand, a population pharmacokinetic analysis of i.v. BU is rare.[30] Our earlier small-scale study revealed high inter- and intrapatient consistency for i.v. BU pharmacokinetics.[22] However, the value of therapeutic drug monitoring remains crucial. Our study demonstrated no essential difference in PK analysis from earlier published Western data,[19] and this supports the notion that racial factors may not seriously influence the bioactivity of i.v. BU.

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Possible Association between Obesity and Posttransplantation Complications Including Infectious Diseases and Acute Graft-versus-Host Disease

Shigeo Fuji,[1] Sung-Won Kim,[1] Ken-ichi Yoshimura,[2] Hideki Akiyama,[3] Shin-ichiro Okamoto,[4] Hiroshi Sao,[5] Junko Takita,[6] Naoki Kobayashi,[7] Shin-ichiro Mori[1] for the Japan Marrow Donor Program

Both obesity and malnutrition are considered risk factors for complications after bone marrow transplantation (BMT). To elucidate the impact of pretransplantation body mass index (BMI) on clinical outcome, we performed a retrospective cohort study with registration data from the Japan Marrow Donor Program (JMDP). Between January 1998 and December 2005, a total of 3935 patients received unrelated BMT through the JMDP; of these, 3827 patients for whom pretransplantation height and weight data were available were included in the study. Patients were stratified according to pretransplantation BMI values (low BMI: BMI < 18 kg/m^2, n = 295; normal BMI: 18 ≤ BMI < 25 kg/m^2, n = 2906; overweight: 25 ≤ BMI <30 kg/m^2, n = 565; obese: 30 kg/m^2 ≤ BMI, n = 61). In a univariate analysis, pretransplantation BMI was associated with a significantly greater risk of grade II-IV acute graft-versus-host disease (GVHD; $P = .03$). Multivariate analysis showed that pretransplantation BMI tended to be associated with an increased risk of grade II-IV acute GVHD ($P = .07$). Obesity was associated with an increased risk of infection compared with normal BMI (odds ratio = 1.9; 95% confidence interval = 1.1 to 3.2; $P = .02$). Our findings demonstrate a correlation between pretransplantation BMI and posttransplantation complications. Although BMI depends strongly on multiple factors, the effect of obesity on clinical outcome should be evaluated in a prospective study.

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KEY WORDS: Obesity, Allogeneic transplantation, Infection, Acute graft-versus-host disease

INTRODUCTION

Both obesity and malnutrition are considered risk factors for complications and increased relapse and nonrelapse mortality in hematopoietic stem cell transplantation (HSCT). An inferior outcome after allogeneic HSCT has been reported in obese adult patients in both allogeneic [1,2] and autologous HSCT [3-5]. Furthermore, our group recently reported that hyperglycemia during the neutropenic period is associated with an increased risk of acute graft-versus-host disease (GVHD) and subsequent nonrelapse mortality [6]. Obesity obviously is associated with an increased risk of hyperglycemia [7], which can lead to an inferior outcome after allogeneic HSCT. Recently, obesity was reported to be associated with low-grade systemic inflammation and was identified as a possible risk factor for autoimmune diseases [8-10]. Alternatively, malnutrition has been reported to be associated with an increased risk of early death after allogeneic HSCT [11,12]. Several reports have noted an association between malnutrition and a high incidence of infectious disease in conventional chemotherapy settings [13-15].

Although we can speculate that these infectious complications may be associated with nonrelapse mortality in HSCT, there is currently no agreement

From the [1]Department of Hematology and Stem Cell Transplantation, National Cancer Center Hospital, Tokyo, Japan; [2]Department of Clinical Trial Management, Translational Research Center, Kyoto University Hospital, Kyoto, Japan; [3]Department of Hematology, Tokyo Metropolitan Cancer and Infectious Diseases Center, Komagome Hospital, Tokyo, Japan; [4]Division of Hematology, Department of Medicine, Keio University School of Medicine, Tokyo, Japan; [5]Department of Hematology, Meitetsu Hospital, Nagoya, Japan; [6]Department of Pediatrics, Graduate School of Medicine, University of Tokyo, Tokyo, Japan; and [7]Department of Hematology, Sapporo Hokuyu Hospital, Sapporo, Japan.

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Correspondence and reprint requests: Shin-ichiro Mori, MD, Department of Hematology and Stem Cell Transplantation, National Cancer Center Hospital, Tokyo, Japan, 5-1-1, Tsukiji, Chuo-Ku, Tokyo 104-0045, Japan. (e-mail: shmori@ncc.go.jp).

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regarding a suitable target range of pretransplantation body mass index (BMI) for clinical management. Previous studies have included various kinds of stem cell sources, and some have included HSCT with T cell depletion. The aim of the present study was to retrospectively evaluate the impact of pretransplantation BMI on the clinical outcome after unrelated bone marrow transplantation (BMT) for hematologic malignancies, using registration data from the Japan Marrow Donor Program (JMDP). The results should provide insight into how to better manage nutritional support for patients undergoing HSCT.

PATIENTS AND METHODS

A total of 3935 patients with various hematologic malignancies underwent BMT through the JMDP between January 1998 and December 2005. Data from 3827 of these patients for whom pretransplantation height and weight data were available were included in the present study. Patient characteristics are summarized in Table 1. The median patient age was 39 years (range, 18 to 72 years), and diagnoses included acute myeloid leukemia (AML; n = 1165), acute lymphoblastic leukemia (ALL; n = 755), myelodysplastic syndrome/myeloproliferative disease (MDS/MPD; n = 597), malignant lymphoma (ML; n = 500) and chronic ML (CML; n = 576), other leukemia (n = 69), multiple myeloma (n = 71), and other (n = 94). Standard risk included acute leukemia in first complete remission (CR1), CML in first chronic phase, MDS in refractory anemia, and lymphoma in CR1. The rest of the patients were categorized as a high-risk group. Bone marrow was the sole stem cell source for transplantation. Total body irradiation (TBI) was used in 2849 patients. GVHD prophylaxis included cyclosporine (CSP)-based (n = 1520) and tacrolimus (TAC)-based regimens (n = 2155), or other combinations (n = 152), with the addition of low-dose antithymocyte globulin (ATG) in 205 patients. Alleles at the HLA-A, -B, and -DRB1 loci were identified by high-resolution DNA typing. The median follow-up period was 565 days. Informed consent was obtained from patients and donors in accordance with the Declaration of Helsinki, and the study design was approved by the JMDP's Institutional Review Board.

The study's primary endpoints were nonrelapse mortality at 100 days and 1 year, overall survival at 1 year, and progression-free survival at 1 year. For nonrelapse mortality, an event was death without disease progression after BMT. For overall survival, an event was death from any cause after BMT. For progression-free survival, an event was disease progression or death after BMT. Secondary endpoints were the incidence of infection (bacterial, viral, fungal, and others); incidence of lung organ toxicity including interstitial pneumonia, adult respiratory distress syndrome, bronchiolitis obliterans, pulmonary hemorrhage, and others, excluding pneumonia with obvious infectious diseases; and incidence of hepatic toxicity, including veno-occlusive disease and drug toxicity. Acute GVHD was classified as grade 0, I, II, III, or IV according to established criteria [16]. The probability of acute GVHD, nonrelapse mortality rate, overall survival, progression-free survival, and relapse rate were estimated using the Kaplan-Meier method. Death without acute GVHD was treated as censoring in the analysis of acute GVHD, and death without progression was treated as censoring in the analysis of relapse. Dichotomous variables between groups were compared using the χ^2 test, and survival times were compared using the log-rank test. An order-restricted version of the log-rank test (a log-rank trend test) was used to test ordered differences between the estimated survival curves. Multivariate analyses were performed using a logistic regression model or a Cox proportional hazards model, as appropriate. The following covariates were included in the univariate analysis: BMI (BMI < 18 kg/m^2, $18 \leq$ BMI < 25 kg/m^2, $25 \leq$ BMI < 30 kg/m^2, and 30 kg/m$^2 \leq$ BMI), sex (donor–recipient pairs), patient age (age < 30 years, $30 \leq$ age < 50 years, age ≥ 50 years), donor age (age < 40 years, age ≥ 40 years), type of disease, risk of leukemia relapse (standard vs high), conditioning (TBI-based vs non–TBI-based), GVHD prophylaxis (CSP-based vs TAC-based), genotypic HLA match versus HLA mismatch, ABO match versus mismatch (major mismatch vs minor mismatch vs major/minor mismatch vs match), cell dose in the graft (dose $< 3.0 \times 10^8$/kg, $3.0 \leq$ dose $< 5.0 \times 10^8$/kg, $\geq 5.0 \times 10^8$/kg), and use of ATG/antilymphocyte globulin (ALG) (ATG/ALG vs no ATG/ALG). All P values were 2-sided. A P value $< .05$ was considered statistically significant.

RESULTS

Patient Characteristics

Table 1 gives the BMI distribution of the study group. Patients were classified into 4 groups based on pretransplantation BMI values according to consensus weight designations from the World Health Organization [17] and the National Heart Lung and Blood Institute Expert Panel [18], as follows: low BMI (BMI < 18 kg/m^2; n = 295), normal BMI ($18 \leq$ BMI < 25 kg/m^2; n = 2906), overweight ($25 \leq$ BMI < 30 kg/m^2; n = 565), and obesity (30 kg/m$^2 \leq$ BMI; n = 61). The prevalence of obesity was quite low compared with that in previous reports from Western countries [1-4]. Significant differences in patient characteristics were observed with regard to age, sex disparity, total nucleated cells (TNCs) per body weight, and primary disease. The low-BMI group

Table 1. Patient Characteristics

	n (%)				
	BMI < 18 kg/m^2	18 ≤ BMI <25 kg/m^2	25 ≤ BMI < 30 kg/m^2	30 kg/m^2 ≤ BMI	
	(n = 295)	(n = 2906)	(n = 565)	(n = 61)	*P* value
Recipient age, years					
< 30	116 (39)	734 (25)	90 (16)	14 (23)	< .0001
30 ≤ age < 50	121 (41)	1473 (51)	322 (57)	36 (59)	
> 50	58 (20)	699 (24)	153 (27)	11 (18)	
Donor age, years					
< 40	217 (74)	2099 (72)	385 (68)	38 (62)	.27
≥ 40	75 (25)	741 (25)	162 (29)	18 (30)	
Sex, donor/recipient					
Match	181 (61)	1833 (63)	374 (66)	39 (64)	< .0001
Male/female	70 (24)	519 (18)	87 (15)	9 (15)	
Female/male	43 (16)	495 (17)	87 (15)	9 (15)	
TNC ($\times 10^{-8}$/kg)					
TNC < 3.0	12 (4)	323 (11)	160 (28)	31 (51)	< .0001
3.0 ≤ TNC< 5.0	60 (20)	1085 (37)	267 (47)	16 (26)	
5.0 ≤ TNC	187 (63)	1191 (41)	78 (14)	1 (2)	
Year of transplantation					
1998	12 (4)	83 (3)	11 (2)	0 (0)	.18
1999	21 (7)	248 (9)	39 (7)	0 (0)	
2000	26 (9)	363 (12)	81 (14)	7 (11)	
2001	43 (15)	398 (14)	74 (13)	7 (11)	
2002	47 (16)	409 (14)	72 (13)	14 (23)	
2003	50 (17)	404 (14)	87 (15)	10 (16)	
2004	40 (14)	463 (16)	88 (16)	12 (20)	
2005	56 (19)	538 (19)	113 (20)	11 (18)	
Diagnosis					
Acute leukemia	186 (63)	1469 (51)	304 (54)	29 (48)	.02
CR1/CR2/>CR2	81/33/65	594/301/541	113/79/107	7/4/18	
Chronic leukemia	30 (10)	449 (15)	84 (15)	13 (21)	
CP1/CP2/AP/BC	16/5/5/3	251/66/65/53	53/8/12/11	5/3/3/2	
MDS/MPD	37 (13)	462 (16)	87 (15)	11 (18)	
RA/RAEB/others	7/12/10	99/155/166	25/33/20	7/3/1	
ML	35 (12)	400 (14)	62 (11)	4 (7)	
CR/>CR	10/19	138/230	24/33	1/3	
MM	5 (2)	56 (2)	10 (2)	0 (0)	
CR/>CR	1/1	10/33	1/7	0/0	
Disease stage*					
Standard	110 (37)	1034 (36)	202 (36)	19 (31)	.67
High	158 (54)	1686 (58)	324 (57)	38 (62)	
Blood type disparity					
Match	146 (49)	1477 (51)	276 (49)	28 (46)	.98
IA	8 (3)	103 (4)	18 (3)	2 (3)	
MA	71 (24)	650 (22)	127 (22)	13 (21)	
MI	65 (22)	586 (20)	121 (21)	14 (23)	
HLA disparity					
HLA allele match	185 (63)	1660 (57)	342 (61)	36 (59)	.01
HLA allele mismatch	70 (24)	857 (29)	149 (26)	18 (30)	
1 allele mismatch	59 (20)	728 (25)	118 (21)	11 (18)	
2 allele mismatch	10 (3)	116 (4)	31 (5)	6 (10)	
3 allele mismatch	1 (0)	13 (0)	0 (0)	1 (2)	
Conditioning regimen					
Conventional	235 (80)	2308 (79)	443 (78)	52 (85)	.25
Reduced-intensity	59 (20)	539 (19)	105 (19)	5 (8)	
TBI for conditioning					
No	80 (27)	654 (23)	146 (26)	12 (20)	.14
Yes	214 (73)	2188 (75)	402 (71)	45 (74)	
ATG for conditioning					
No	268 (91)	2670 (92)	517 (92)	55 (90)	.21
Yes	23 (8)	155 (5)	25 (4)	2 (3)	
GVHD prophylaxis					
CSP-based	137 (46)	1141 (39)	226 (40)	16 (26)	.19
TAC-based	153 (52)	1651 (57)	312 (55)	39 (64)	
Others	3 (1)	48 (2)	7 (1)	2 (3)	
Comorbidity					
Liver dysfunction					
No	239 (81)	2436 (84)	481 (85)	49 (80)	.78
Yes	41 (14)	360 (12)	66 (12)	7 (11)	

(Continued)

Table 1. (*Continued*)

	n (%)				
	BMI < 18 kg/m^2	18 ≤ BMI <25 kg/m^2	25 ≤ BMI < 30 kg/m^2	30 kg/m^2 ≤ BMI	
	(n = 295)	(n = 2906)	(n = 565)	(n = 61)	*P* value
Renal dysfunction					
No	273 (93)	2706 (93)	528 (93)	54 (89)	.90
Yes	7 (2)	90 (3)	19 (3)	2 (3)	
Heart dysfunction					
No	260 (88)	2601 (90)	519 (92)	54 (89)	.32
Yes	20 (7)	195 (7)	28 (5)	2 (3)	
Pulmonary dysfunction					
No	267 (91)	2709 (93)	528 (93)	56 (92)	.27
Yes	13 (4)	87 (3)	19 (3)	0 (0)	

*Disease stage: Standard risk stage included CR1 in acute leukemia, first chronic phase in CML, and CR1 in lymphoma. Others were classified as high-risk stage.

included more young patients, patients receiving high TNCs per body weight, patients with acute leukemia, and male patients with a female donor.

Clinical Outcomes

The incidence of grade II-IV acute GVHD was 42% in the low-BMI group, 45% in the normal-BMI group, 48% in the overweight group, and 58% in the obesity group (Figure 1A). Thus, increased BMI was significantly associated with a higher incidence of grade II-IV acute GVHD (P = .03 by the log-rank trend test). Other factors associated with a higher incidence of grade II-IV acute GVHD were HLA allele disparity, GVHD prophylaxis with CSP (vs with TAC), and donor age ≥ 40 years. Multivariate analysis showed that pretransplantation BMI tended to be associated with an increased risk of grade II-IV acute GVHD (P = .07, log-rank trend test) (Table 2). The incidence of grade III-IV acute GVHD was 17% in the low-BMI group, 17% in the normal-BMI group, 19% in the overweight group, and 25% in the obesity group (Figure 1B). An increase in BMI tended to be associated with a higher incidence of grade III-IV acute GVHD, but this trend was not significant (P = .087, log-rank trend test). Multivariate analysis showed no association between pretransplantation BMI and the incidence of grade III-IV acute GVHD (P = .19, log-rank trend test) (Table 3).

Nonrelapse mortality was 29% in the low-BMI group, 31% in the normal-BMI group, 32% in the overweight group, and 40% in the obesity group at 1 year after BMT (P = .19, log-rank trend test) (Figure 2A). Overall survival was 61% in the low-BMI group, 58% in the normal-BMI group, 59% in the overweight group, and 53% in the obesity group at 1 year after BMT (P = .98, log-rank trend test) (Figure 2B). Progression-free survival was 54%, 52%, 56%, and 47% (P = .72, log-rank trend test), and the relapse rate was 24%, 24%, 18%, 21%, respectively, in the 4 groups at 1 year after BMT (P = .04 by log-rank trend test) (Figure 2C and D). The incidence of systemic infectious diseases, including bacterial, fungal, and viral infections, was 39%, 43%, 46%, and 59%, respectively, in the 4 groups (Figure 3). Obesity was significantly associated with increased incidence of infectious disease compared with normal

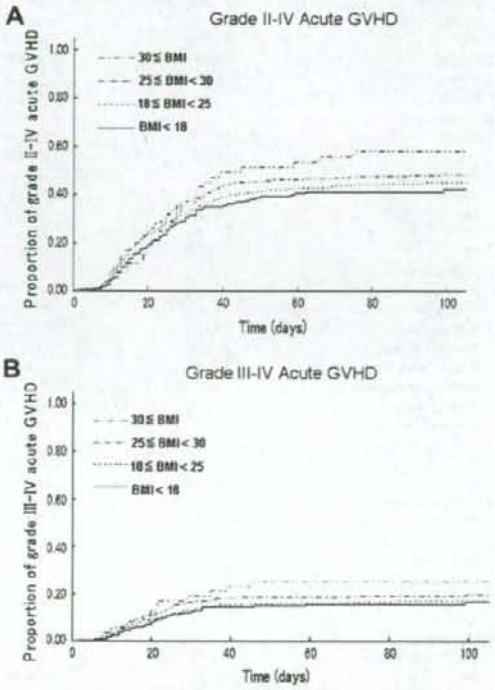

Figure 1. Probability of grade II-IV acute GVHD (A) and grade III-IV acute GVHD (B).

Table 2. Univariate and Multivariate Analyses of Risk Factors for Grade II-IV Acute GVHD

	Univariate Analysis			Multivariate Analysis		
Covariates	HR	95% CI	*P* value	HR	95% CI	*P* value
Recipient BMI						
18 ≤ BMI < 25 kg/m^2	1.00		.026*	1.00		.066*
BMI < 18 kg/m^2	0.91	0.75 to 1.10		1.06	0.85 to 1.31	
25 ≤ BMI < 30 kg/m^2	1.11	0.97 to 1.27		1.19	0.93 to 1.52	
30 ≤ BMI kg/m^2	1.28	0.89 to 1.85		1.29	0.82 to 2.03	
Recipient age, years						
<30	1.00		.85*			
30 ≤ age < 50	1.00	0.88 to 1.13				
≥ 50	0.99	0.86 to 1.14				
Donor age, years						
< 40	1.00		< .0001	1.00		< .0001
≥ 40	1.30	1.17 to 1.45		1.28	1.13 to 1.44	
Sex, donor/recipient						
Match	1.00		.053	1.00		.20
Male/female	1.12	0.98 to 1.27		1.09	0.95 to 1.26	
Female/male	1.15	1.01 to 1.32		1.12	0.97 to 1.30	
TNC (× 10^{-8}/kg)						
TNC <3.0	1.00		.76*			
3.0 ≤ TNC < 5.0	1.03	0.88 to 1.20				
5.0 ≤ TNC	0.99	0.85 to 1.16				
Diagnosis						
Acute	1.00		.28			
Chronic	1.08	0.93 to 1.24				
MDS/MPD	1.01	0.87 to 1.16				
ML	1.17	1.01 to 1.35				
MM	0.92	0.62 to 1.36				
Blood type disparity						
M	1.00		.15	1.00		.49
IA	1.19	0.92 to 1.55		1.14	0.85 to 1.52	
MA	1.03	0.90 to 1.16		1.02	0.89 to 1.17	
MI	1.14	1.00 to 1.29		1.11	0.96 to 1.27	
HLA disparity						
HLA allele match	1.00		< .0001	1.00		< .0001
HLA 1 allele mismatch	1.30	1.16 to 1.47		1.36	1.21 to 1.54	
HLA 2 allele mismatch	1.49	1.18 to 1.88		1.51	1.19 to 1.93	
HLA 3 allele mismatch	2.23	1.16 to 4.30		2.23	1.15 to 4.31	
Conditioning regimen:						
TBI for conditioning						
No	1.00		.67			
Yes	0.98	0.87 to 1.10				
Intensity of conditioning:						
Conventional	1.00		.42			
Reduced-intensity	0.95	0.84 to 1.08				
ATG for conditioning						
No	1.00		.58			
Yes	0.94	0.75 to 1.17				
GVHD prophylaxis						
CSP-based	1.00		.025	1.00		.0003
TAC-based	0.89	0.80 to 0.98		0.80	0.71 to 0.89	
Others	1.21	0.84 to 1.75		0.99	0.66 to 1.49	
Comorbidity						
Liver dysfunction						
No	1.00		.52			
Yes	0.95	0.82 to 1.11				
Renal dysfunction						
No	1.00		.84			
Yes	1.03	0.77 to 1.38				
Heart dysfunction						
No	1.00		.58			
Yes	0.94	0.77 to 1.16				
Lung dysfunction						
No	1.00		.15	1.00		.09
Yes	1.22	0.93 to 1.60		1.29	0.96 to 1.74	

*The log-rank trend test was used for calculating *P* values.

Table 3. Univariate and Multivariate Analyses of Risk Factors for Grade III-IV Acute GVHD

	Univariate Analysis			Multivariate Analysis		
Covariate	HR	95% CI	*P* value	HR	95% CI	*P* value
Recipient BMI						
18 ≤ BMI < 25 kg/m^2	1.00		.086*	1.00		.19*
BMI < 18 kg/m^2	0.99	0.73 to 1.35		1.01	0.72 to 1.43	
25 ≤ BMI < 30 kg/m^2	1.16	0.94 to 1.45		1.16	0.79 to 1.72	
30 ≤ BMI kg/m^2	1.56	0.90 to 2.71		1.42	0.70 to 2.87	
Recipient age, years						
< 30	1.00		.97*			
30 ≤ age <50	0.98	0.81 to 1.19				
≥ 50	1.00	0.79 to 1.25				
Donor age, years						
< 40	1.00		< .0001	1.00		< .0001
≥ 40	1.52	1.28 to 1.79		1.53	1.27 to 1.84	
Sex, donor/recipient						
Match	1.00		.057	1.00		.15
Male/female	1.21	0.99 to 1.49		1.13	0.90 to 1.42	
Female/male	1.23	1.00 to 1.51		1.24	0.99 to 1.56	
TNC (× 10^{-8}/kg)						
TNC < 3.0	1.00		.56*			
3.0 < TNC < 5.0	0.90	0.71 to 1.15				
5.0 < TNC	0.91	0.72 to 1.16				
Diagnosis						
Acute leukemia	1.00		.66			
Chronic leukemia	1.13	0.90 to 1.41				
MDS/MPD	0.99	0.78 to 1.24				
ML	0.98	0.77 to 1.26				
MM	0.71	0.35 to 1.43				
Blood type disparity						
M	1.00		.55			
IA	1.04	0.67 to 1.61				
MA	1.11	0.91 to 1.13				
MI	1.15	0.94 to1.40				
HLA disparity						
HLA allele match	1.00		.0002	1.00		< .0001
HLA 1 allele mismatch	1.36	1.13 to 1.64		1.43	1.18 to 1.74	
HLA 2 allele mismatch	1.57	1.10 to 2.24		1.57	1.07 to 2.30	
HLA 3 allele mismatch	1.49	0.48 to 4.66		1.47	0.47 to 4.60	
Conditioning regimen						
TBI for conditioning						
No	1.00		.49			
Yes	0.94	0.78 to 1.13				
Intensity of conditioning regimen						
Conventional	1.00		.29			
Reduced-intensity	1.11	0.91 to 1.36				
ATG for conditioning						
No	1.00		.26			
Yes	0.80	0.55 to 1.18				
GVHD prophylaxis						
CSP-based	1.00		.029	1.00		.02
TAC-based	0.93	0.79 to 1.10		0.81	0.67 to 0.97	
Others	1.78	1.09 to 2.91		1.27	0.71 to 2.28	
Comorbidity						
Liver dysfunction						
No	1.00		.95			
Yes	0.99	0.78 to 1.27				
Renal dysfunction						
No	1.00		.20			
Yes	1.32	0.87 to 1.99				
Heart dysfunction						
No	1.00		.26			
Yes	0.82	0.57 to 1.16				
Lung dysfunction						
No	1.00		.11	1.00		.20
Yes	1.39	0.93 to 2.07		1.36	0.86 to 2.16	

*The log-rank trend test was used for calculating P values.

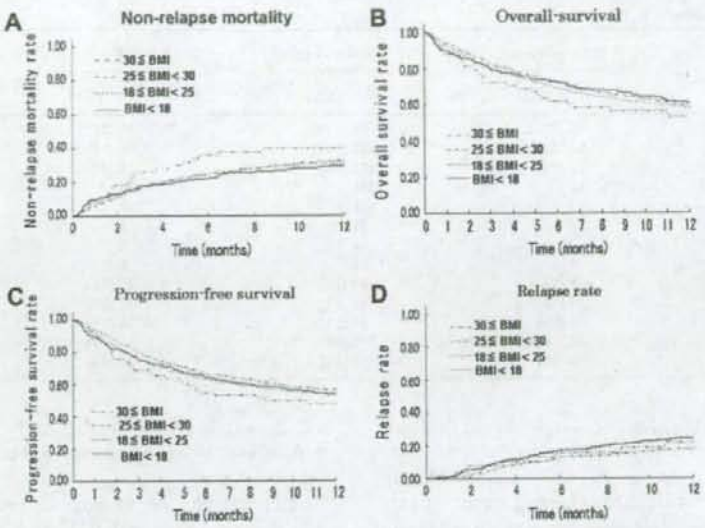

Figure 2. Probability of nonrelapse mortality (A), progression-free survival (B), overall survival (C), and relapse/progression (D).

weight (odds ratio = 1.9; 95% confidence interval [CI] = 1.1 to 3.2; $P = .02$). The incidence of liver dysfunction, including sinusoidal occlusive syndrome, was 19% in the low-BMI group, 20% in the normal-BMI group, 21% in the overweight group, and 25% in the obesity group; the differences were not statistically significant. The incidence of interstitial pneumonia, excluding obvious infectious diseases such as cytomegalovirus or *Pneumocystis jirovecii* pneumonia, was 13% in the low-BMI group, 13% in the normal-BMI group, 12% in the overweight group, and 15% in the obesity group; again, the differences are not statistically significant. The causes of death are given in Table 4. More infections and GVHD-related deaths were seen in the obesity group. If only early mortality is considered, then the nonrelapse mortality within 100 days was 17% in the low-BMI group, 18% in the normal-BMI group, 17% in the overweight group, and 25% in the obesity group. Obesity tended to be associated with greater early nonrelapse mortality, but this difference was not statistically significant ($P = .83$). The incidence of infection-related mortality within 100 days was 5%, 5%, 4% and 8%, respectively, in the 4 groups. Bacterial infection was the main cause of infection-related mortality, with 6 cases (40%) in the low-BMI group, 91 cases (67%) in the normal-BMI group, 17 cases (74%) in the overweight group, and 3 cases (60%) in the obesity group.

To investigate whether pretransplantation BMI had an additional impact on outcome in the patients who developed acute GVHD, we stratified the patients according to the grade of acute GVHD and analyzed the association between pretransplantation BMI and early nonrelapse mortality. We found that pretransplantation BMI had no additional impact on early nonrelapse mortality.

DISCUSSION

Both obesity and malnutrition are considered risk factors for complications, especially infectious diseases. To elucidate the impact of pretransplantation BMI on the clinical outcome, in this study we retrospectively reviewed the data of patients who underwent unrelated BMT, stratified according to recipient BMI, and found results similar to those reported previously [1,2]. The present study has an obvious limitation,

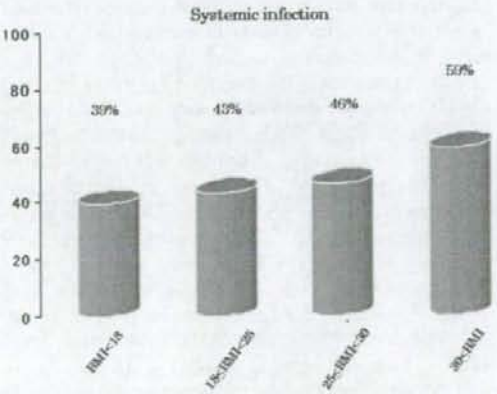

Figure 3. Incidence of systemic infections.

Table 4. Causes of Death

	BMI < 18 kg/m^2	18 ≤ BMI < 25 kg/m^2	25 ≤ BMI < 30 kg/m^2	30 kg/m^2 ≤ BMI
	(n = 295)	(n = 2906)	(n = 565)	(n = 61)
Relapse, n (%)	66 (22%)	609 (21%)	95 (17%)	8 (13%)
Nonrelapse mortality, (%)	97 (33%)	988 (34%)	206 (36%)	24 (39%)
Infection, n (%)	25 (8%)	276 (9%)	56 (10%)	8 (13%)
Bacterial, n	13	151	34	4
Fungal, n	3	37	7	1
Viral, n	4	39	5	1
Mixed, n	2	16	3	1
Others, n	3	33	6	1
Acute GVHD, n (%)	9 (3%)	78 (3%)	26 (5%)	4 (7%)
Chronic GVHD, n (%)	4 (1%)	45 (2%)	13 (2%)	2 (3%)
Graft failure, n (%)	2 (1%)	31 (1%)	6 (1%)	1 (2%)
Organ dysfunction, n (%)	47 (16%)	395 (14%)	83 (15%)	7 (11%)
Others, n (%)	10 (3%)	163 (6%)	22 (4%)	2 (3%)

lacking concise data regarding weight-based dose adjustment of chemotherapy, which is critical for analyzing the incidence of organ dysfunction. Dosing schemes for preparative chemotherapy regimens vary widely among transplantation centers. In addition, centers differ in their use of ideal body weight, actual body weight, and compensatory calculations that yield doses between the actual and ideal weights [19,20]. Another limitation of this study is that low prevalence of obesity in Japan makes the study's statistical power less reliable. For example, patients with morbid obesity (BMI > 35 kg/m^2), considered a significant comorbidity in a hematopoietic cell transplantation-specific comorbidity index, are quite rare in Japan [21]. Similar analyses need to be performed in Western countries to clarify the impact of obesity, especially morbid obesity, after allogeneic HSCT.

Our findings demonstrate that obesity is associated with an increased risk of infectious disease compared with normal weight. Hyperglycemia, caused primarily by insulin resistance in obesity, can lead to increased incidence of infectious disease. As reported by Sheean et al. [22], hyperglycemia after HSCT may be a risk factor for infectious disease. Recently, Derr et al. [23] reported an association between hyperglycemia before a neutropenic period and increased risk of infectious diseases during a neutropenic period after HSCT. In our study, an increased incidence of acute GVHD was associated with an increased risk of infectious disease. On the other hand, low BMI, which suggests the presence of malnutrition, was not associated with an increased risk of infectious diseases or transplantation-related mortality, inconsistent with previous reports [10,11]. This could be because the incidence of acute GVHD was lower and the dose of TNC per body weight was higher in the low-BMI group. Even if we further divide the BMI < 18 kg/m^2 group into 3 subgroups (BMI < 16 kg/m^2, 16 ≤ BMI < 17 kg/m^2, and 17 ≤ BMI < 18 kg/m^2), we find no differences in the incidence of acute GVHD or infectious disease, or in clinical outcomes (data not shown). It is possible that in the Japanese population, BMI < 18 kg/m^2 may not directly reflect a malnutritional status.

Importantly, our findings also suggest an association between increased BMI and a significantly increased incidence of acute GVHD grade II-IV. This observation is based on multiple factors, and no single clear scientific explanation for it exists, but several mechanisms can be hypothesized. First, the dose of the conditioning regimen and GVHD prophylaxis could be improperly adjusted in obese patients, possibly leading to increased tissue damage or poorer GVHD prophylaxis and, ultimately, a higher incidence of acute GVHD. With regard to the conditioning regimen, the relapse rate was lower in the overweight and obese patients compared with the low-BMI and normal-BMI patients, but the incidence of regimen-related toxicity (ie, liver dysfunction and interstitial pneumonitis) did not differ significantly among these groups. With regard to GVHD prophylaxis, there might not have been any significant difference in drug exposure, because dose adjustment of the calcineurin inhibitor usually is done through serial monitoring of drug concentration. Second, the stem cell dose could influence the incidence of acute GVHD. But in this study, the stem cell dose was analyzed independently, and no association was found between stem cell dose and the incidence of acute GVHD. Third, there was an obvious selection bias in each group. For example, it is possible that obese patients may be less likely to find an unrelated donor with an adequate dose of cells for transplantation. While the donor search continued, the number of chemotherapy courses could increase, and the patient's general condition (including disease status and organ function) could become worse. Finally, even though there were no direct data regarding glucose levels in this study, obesity is likely associated with hyperglycemia [7-9], possibly resulting in elevated levels of several cytokines [24-27], inducing a vicious cycle [28-30]. Our group previously reported an association

between hyperglycemia during neutropenia and the development of acute GVHD [6], possibly due to the augmented production of cytokines stimulated by the conditioning regimen. Furthermore, recently it has become clear that adipocytokines, which are secreted mainly from adipocytes, play important roles in the control of immunity [31-33]. In particular, the level of leptin has been found to be proportional to body fat weight and to affect T regulatory cell (Treg) proliferation and function [34,35]. Thus, it could be hypothesized that in obese patients, a higher leptin level suppresses Treg activity, increasing the risk of acute GVHD. These mechanisms are based on the results of animal models, however, and await confirmation in human studies.

The clinical significance of our findings merits careful consideration, because pretransplantation BMI is one of the few factors that can be properly managed and corrected during the unstable, fast-moving pretransplantation period. On the other hand, malnutrition can be corrected by appropriate nutritional support, and obesity can be controlled through an appropriate diet and exercise program during chemotherapy. This study suggests that such a pretransplantation nutritional support program can improve clinical outcomes after allogeneic BMT.

In conclusion, this retrospective analysis of registration data found an association between pretransplantation obesity and increased risk of infectious disease, possibly leading to increased risk of mortality. Although body weight is affected by multiple clinical factors, the effect of obesity on clinical outcome, as suggested here, needs to be confirmed by a prospective study to identify better patient management approaches.

ACKNOWLEDGMENTS

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ORIGINAL ARTICLE

Reduced-intensity unrelated donor bone marrow transplantation for hematologic malignancies

Sung-Won Kim · Keitaro Matsuo · Takahiro Fukuda · Masamichi Hara · Kosei Matsue · Shuichi Taniguchi · Tetsuya Eto · Mitsune Tanimoto · Atsushi Wake · Kazuo Hatanaka · Shinji Nakao · Yoji Ishida · Mine Harada · Atae Utsunomiya · Masahiro Imamura · Yoshinobu Kanda · Kazutaka Sunami · Fumio Kawano · Yoichi Takaue · Takanori Teshima

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Abstract To review a current experience of unrelated bone marrow transplantation (BMT) with reduced-intensity conditioning (RIC) regimens, we conducted a nationwide survey with 77 patients (age, 25–68 years). The backbone RIC regimen was a combination of fludarabine or cladribine, busulfan or melphalan and total body irradiation at 2–4 Gy. Five patients died early, but 71 (92%) achieved initial neutrophil recovery. Thereafter, 36 patients (47%) died of therapy-related complications, 23 (30%) of whom died within day 100. Grades II–IV acute graft-versus-host disease (GVHD) occurred in 34 of the 68 evaluable patients (50%). In a multivariate analysis, a regimen containing antithymocyte globulin (ATG) was significantly associated with a decreased risk of acute GVHD ($P = 0.041$). Thirty-three patients are currently alive with a median follow-up of 439 days (28–2002 days), with an OS of 50% at 1 year. In conclusion, unrelated BMT with RIC regimens can be a curative treatment in a subset of patients.

S.-W. Kim · T. Fukuda · Y. Takaue
Hematology and Hematopoietic Stem Cell Transplantation Division, National Cancer Center Hospital, Tokyo, Japan

K. Matsuo
Division of Epidemiology and Prevention, Aichi Cancer Center Research Institute, Nagoya, Japan

M. Hara
Department of Hematology, Ehime Prefectural Central Hospital, Matsuyama, Japan

K. Matsue
Division of Hematology/Oncology, Kameda Medical Center, Kamogawa, Japan

S. Taniguchi · A. Wake
Department of Hematology, Toranomon Hospital, Tokyo, Japan

T. Eto
Department of Hematology, Hamanomachi Hospital, Fukuoka, Japan

M. Tanimoto
Department of Hematology and Oncology, Okayama University Graduate School of Medicine and Dentistry, Okayama, Japan

K. Hatanaka
Department of Internal Medicine, Rinku General Medical Center, Izumisano, Japan

S. Nakao
Department of Hematology, Kanazawa University Graduate School of Medical Science, Kanazawa, Japan

Y. Ishida
Division of Hematology/Oncology, Iwate Medical University School of Medicine, Morioka, Japan

M. Harada
Department of Medicine and Biosystemic Science, Kyushu University, Fukuoka, Japan

A. Utsunomiya
Department of Internal Medicine, Imamura Bun-in Hospital, Kagoshima, Japan

M. Imamura
Department of Hematology and Oncology, Hokkaido University Graduate School of Medicine, Sapporo, Japan

Y. Kanda
Division of Hematology, Saitama Medical Center, Jichi Medical University, Saitama, Japan