Significance of Granulocyte Colony-Stimulating Factor-Combined High-Dose Cytarabine, Cyclophosphamide, and Total Body Irradiation in Allogeneic Hematopoietic Cell Transplantation for Myeloid Malignant Neoplasms

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ABSTRACT

Allogeneic hematopoietic cell transplant (HCT) is a curative procedure for myeloid malignant neoplasms, but relapse after HCT remains critical. A conditioning regimen involving granulocyte colony-stimulating factor-combined high-dose cytarabine, cyclophosphamide, and total body irradiation (G-CSF–combined high-dose cytarabine/cyclophosphamide/total-body irradiation [HDCA/CY/TBI]) was reported to improve outcomes after cord blood transplant (CBT) for myeloid malignant neoplasms, but this regimen was not previously evaluated among patients undergoing bone marrow transplant (BMT) or peripheral blood stem cell transplant (PBSCT).

Methods. We retrospectively analyzed 28 patients who underwent allogeneic HCT including BMT from a related (1 patient) or unrelated donor (9 patients), PBSCT from a related donor (7 patients), or single-unit CBT from an unrelated donor (11 patients) after a G-CSF–combined HDCA/CY/TBI regimen.

Results. All patients achieved neutrophil and platelet engraftment, which were significantly more rapid in the BMT/PBSCT group than in the CBT group. Eighteen patients were alive at a median follow-up of 54.3 months. The 3-year relapse and nonrelapse mortality rates were 28.6% and 7.1%, respectively, which were similar between the BMT/PBSCT and CBT groups. Overall survival and disease-free survival at 5 years after HCT were 62.6% and 64.3%, respectively, which were also similar between the BMT/PBSCT and CBT groups. Only disease status at HCT had a significant impact on overall survival and disease-free survival (86.7% with standard risk vs 38.5% with high risk and 86.7% with standard risk vs 38.5% with high risk, respectively).

Conclusion. A G-CSF–combined HDCA/CY/TBI regimen is a promising conditioning in patients with myeloid malignant neoplasms who undergo not only CBT but also BMT or PBSCT.

ALLOGENEIC hematopoietic cell transplant (HCT) is a curative procedure for myeloid malignant neoplasms. However, post-HCT relapse remains critical. To reduce post-HCT relapse, intensification of conditioning regimens, which provides strong antitumor activity, has been attempted; however, this resulted in increased nonrelapse mortality (NRM) and inferior overall survival (OS) compared with conventional regimens [1–3]. Recently, an intensified

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myeloablative conditioning regimen involving addition of high-dose cytarabine (HDCA) to the conventional combination of cyclophosphamide (CY) and total body irradiation (TBI) was reported to improve outcomes after cord blood transplant (CBT) for myeloid malignant neoplasms compared with the CY/TBI regimen [4]. However, bone marrow transplant (BMT) and peripheral blood stem cell transplant (PBSCT) from related or unrelated donors receiving the HDCA/CY/TBI regimen did not improve outcomes [5]. Thus, the efficacy of the HDCA/CY/TBI regimen differed between donor sources. This is because, in CBT, the antileukemic effect depends on the intensity of the conditioning regimen because of the weakness of the graft-vs-leukemia effect, whereas in BMT or PBSCT, the intensified regimen induces tissue damage and increases NRM because of the frequency of graft-vs-host disease (GVHD) [4,5]. Granulocyte colony-stimulating factor (G-CSF) increases the susceptibility of dormant myelogenous leukemic cells to cytarabine through the induction of cell cycle entry. Several studies have reported that addition of G-CSF combined with HDCA to the CY/TBI regimen resulted in a high incidence of neutrophil engraftment, reduced relapse rate, and better OS after CBT for myeloid malignant neoplasms [6–9]. A G-CSF–combined HDCA/CY/TBI regimen was not previously evaluated among patients undergoing BMT or PBSCT. Therefore, it is unclear whether the efficacy of the G-CSF–combined HDCA/CY/TBI regimen differs between donor sources in the same manner as a HDCA/CY/TBI regimen. Therefore, we report a retrospective study that assessed the efficacy of a G-CSF–combined HDCA/CY/TBI regimen for BMT or PBSCT compared with that of CBT for transplant outcomes in adult patients with myeloid malignant neoplasms.

PATIENTS AND METHODS

This retrospective study was approved by the Institutional Review Board of Yamaguchi University Hospital (no. H28-033). All procedures were performed in accordance with the Declaration of Helsinki. All patients provided written informed consent for the use of their data at the time of original treatment.

Patients

We analyzed adult patients with myeloid malignant neoplasms, including acute myelogenous leukemia, chronic myelogenous leukemia, and myelodysplastic syndrome, who underwent BMT from related or unrelated donors, PBSCT from related donors, or single-unit CBT from unrelated donors as their first allogenic HCT with a G-CSF–combined HDCA/CY/TBI regimen between 2009 and 2015 at Yamaguchi University Hospital. Patients were considered to have standard risk if they were in first complete remission or the first chronic phase of chronic myelogenous leukemia at the time of HCT. Other patients were considered to have high risk.

Conditioning Regimen

All patients received the same myeloablative conditioning. Total body irradiation of 12 Gy was delivered in 6 fractions for 3 days (days −8, −7, and −6). After completion of TBI, 2 or 3 g/m^2 HDCA was administered intravenously for 2 hours every 12 hours for 2 consecutive days (days −5 and −4). The G-CSF was administered by continuous infusion of lenograstim or filgrastim at a daily dose of 5 μg/kg or 300 μg/m^2, respectively, starting 12 hours before the first HDCA dose and continuing until the last HDCA dose. A CY (60 mg/kg) was administered intravenously for 2 hours for 2 consecutive days (days −3 and −2), as described previously [6]. If the patients' condition permitted them to receive the intensified conditioning, they underwent allogenic HCT with this regimen. Antithymocyte globulin was not administered as part of the conditioning regimen, and T-cell depletion of the graft was not performed in any patient.

Graft-vs-Host Disease Prophylaxis

For prophylaxis of GVHD, patients received short-term methotrexate (15 mg/m^2 on day 1 and 10 mg/m^2 on days 3, 6, and 11) and a calcineurin inhibitor. Cyclosporine A at 3 mg/kg/d or tacrolimus at 0.03 mg/kg/d was administered by continuous infusion starting on day −1, and treatment was switched to oral administration when the patient could reliably consume oral medication. In BMT using HLA of an identical sibling or CBT, methotrexate on day 11 was omitted. Moreover, cyclosporine A was used for HCT from a HLA-identical sibling donor, and tacrolimus was used for alternative donor HCT.

Assessment of Engraftment and GVHD

The day of neutrophil engraftment was defined as the first day of 3 consecutive days on which the absolute neutrophil count exceeded 0.5×10^9/L. The day of platelet engraftment was defined as the day on which the absolute platelet count exceeded 20×10^9/L without platelet transfusion. Both acute and chronic GVHD were diagnosed and graded on the basis of published criteria [10,11].

Statistical Analysis

Continuous and dichotomous variables in the 2 groups were compared using the Mann-Whitney test and Fisher exact test, respectively. The incidences of relapse and NRM were calculated using the Gray test, and each event was considered a competing risk [12]. The incidences of neutrophil and platelet engraftment were calculated using the Gray test, and death without engraftment or relapse was considered a competing risk [12]. The cumulative incidence of GVHD was also calculated using the Gray test, and death without GVHD or relapse was considered a competing risk [12]. Disease-free survival (DFS) and OS were estimated using the Kaplan-Meier method and compared between groups using the log-rank test. Factors with P values $< .05$ were considered statistically significant. All analyses were performed with EZR (Saitama Medical Center, Jichi Medical University, Saitama, Japan), which is a modified version of R software that contains a graphical user interface and is designed for statistical functions that are frequently used in biostatistics (R Foundation for Statistical Computing, Vienna, Austria) [13].

RESULTS

Patient Characteristics

This study included 28 patients, and their characteristics are presented in Table 1. Prophylaxis of GVHD was significantly different between the BMT/PBSCT and CBT groups. There were no other significant differences between the groups regarding patient characteristics other than donor source.

Table 1. Patient Characteristics

	Total	BMT/PBSCT	CBT	*P* Value
No. of patients	28	17	11	
Age, median (range), y	42 (16–55)	35 (16–55)	42 (27–53)	.18
Sex				
Male	18	13	5	.13
Female	10	4	6	
HCT-CI				
0	22	14	8	.25
1	2	2	0	
2	4	1	3	
Patient-donor sex				
Match	18	11	7	1
Mismatch	10	6	4	
Donor source				
Rel-BM	1	1	0	<.01
Rel-PB	7	7	0	
UR-BM	9	9	0	
UR-CB	11	0	11	
Diagnosis				
AML	25	16	9	.49
CML	2	1	1	
MDS	1	0	1	
Disease status				
Standard risk	15	9	6	1
High risk	13	8	5	
GVHD prophylaxis				
CyA+MTX	8	8	0	<.01
Tac+MTX	20	9	11	
Follow-up period, median (range), mo	54.3 (25.8–91.0)	52.1 (25.8–86.0)	54.3 (27.7–91.0)	.89

Abbreviations: AML, acute myelogenous leukemia; BMT, bone marrow transplant; CBT, cord blood transplant; CML, chronic myelogenous leukemia; CyA, cyclosporine; GVHD, graft-vs-host disease; HCT-CI, hematopoietic cell transplant comorbidity index; MDS, myelodysplastic syndrome; MTX, methotrexate; PBSCT, peripheral blood stem cell transplant; Rel-BM, related-bone marrow; Rel-PB, related-peripheral blood; Tac, tacrolimus; UR-BM, unrelated-bone marrow; UR-CB, unrelated-cord blood.

Regimen-Related Toxicities

Regimen-related toxicities are summarized in Table 2. The regimens were generally well tolerated. Among the 28 patients, grade 3 toxicities were observed with oral mucositis (n = 19) and diarrhea (n = 10). No other grade 3 and no grade 4 toxicities were observed. Regimen-related toxicities were not significantly different between the BMT/PBSCT and CBT groups. There was no regimen-related death early after HCT.

Table 2. Regimen-Related Toxicities

	BMT/PBSCT					CBT				
Grades*	0	1	2	3	4	0	1	2	3	4
Oral mucositis	0	0	4	13	0	0	0	5	6	0
Diarrhea	0	2	9	6	0	1	1	5	4	0
Liver	14	1	2	0	0	9	2	0	0	0
Cystitis	16	1	0	0	0	11	0	0	0	0

Abbreviations: BMT, bone marrow transplant; CBT, cord blood transplant; PBSCT, peripheral blood stem cell transplant.

*Grades were evaluated according to the National Cancer Institute Common Toxicity Criteria (ver. 4.0).

Engraftment

In all patients, neutrophil engraftment was obtained at a median of 19.5 days (range, 12–24 days), and platelet engraftment was obtained at a median of 30.5 days (range, 12–99 days). Neutrophil and platelet engraftment were significantly more rapid in the BMT/PBSCT group than in the CBT group (median, 16 days [range, 12–22 days] in BMT/PBSCT group vs median, 21 days [range, 19–24 days] in CBT group; $P < .01$; median, 24 days [range, 12–43 days] in BMT/PBSCT group vs median, 39 days [range, 24–99 days] in CBT group; $P < .01$, respectively).

Graft-vs-Host Disease

Acute GVHD developed in 16 of 28 evaluable patients with neutrophil engraftment (grade I in 8 patients, grade II in 7 patients, and grade IV in 1 patient). The cumulative incidence of grade II to IV acute GVHD was 28.6% (95% CI, 13.3–46.0), and there was no significant difference between the BMT/PBSCT and CBT groups (23.5% vs 36.4%; $P = .51$). Among the 25 patients who survived without disease relapse >100 days after HCT, 17 developed chronic GVHD (limited type in 8 patients and extensive type in 9 patients). Extensive-type chronic GVHD had a cumulative incidence of 32.1% (95% CI, 15.8–49.8) and was significantly more frequent in the CBT group than the BMT/PBSCT group (54.5% vs 17.6%; $P = .03$).

Survival

Eighteen patients were alive at a median follow-up of 54.3 months (range, 25.8–91.0 months). Causes of death in the other 10 patients included disease relapse and complications associated with treatment of relapse after HCT (8 patients), grade IV acute GVHD and thrombotic microangiopathy (1 patient), and cytomegalovirus and human herpesvirus 6 encephalitis (1 patient). The cumulative incidence of NRM was 7.1% (95% CI, 1.2–20.7) at 3 years after HCT, which was not significantly different between the BMT/PBSCT and CBT groups (0.0% vs 18.2%; $P = .07$; Fig 1A). In 8 patients, disease relapse occurred at a median of 5.8 months (range, 1.4–12.7 months) after HCT. The 3-year relapse rate was 28.6% (95% CI, 13.3–46.0), which was similar between the BMT/PBSCT and CBT groups (29.4% vs 27.3%; $P = .82$; Fig 1B). The OS and DFS at 5 years after HCT were 62.6% (95% CI, 41.2–78.1) and 64.3% (95% CI, 43.8–78.9), respectively. The OS and DFS at 5 years after HCT were similar between the BMT/PBSCT and CBT groups (68.0% and 54.5%; $P = .40$; and 70.6% and 54.5%; $P = .46$, respectively [Fig 2A]). The following variables were analyzed as other possible factors affecting OS and DFS: patient age (<40 years vs ≥ 40 years), sex, HCT-comorbidity index, patient-donor sex mismatch, disease status at HCT (standard vs high risk), and development of grade II to IV acute GVHD or extensive type chronic GVHD.

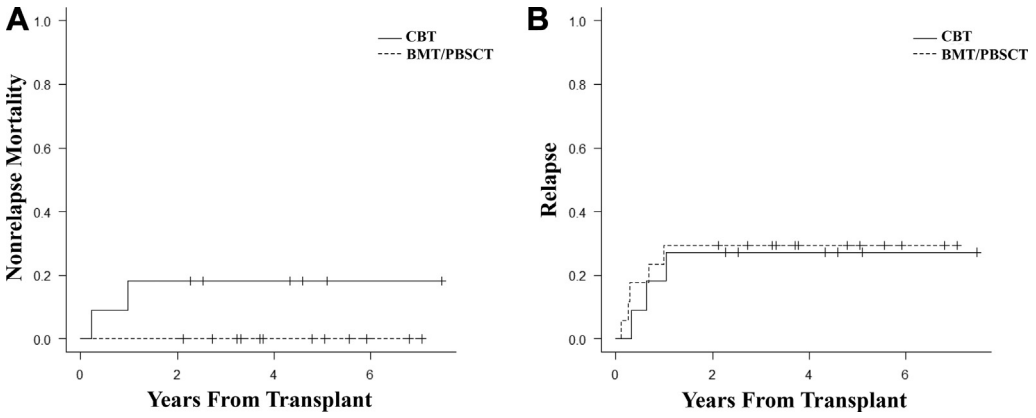

Fig 1. **(A)** Cumulative incidence of nonrelapse mortality according to donor sources. **(B)** Cumulative incidence of relapse according to donor sources. BMT, bone marrow transplant; CBT, cord blood transplant; PBSCT, peripheral blood stem cell transplant.

Consequently, only disease status at HCT had a significant impact on OS and DFS (86.7% with standard risk vs 38.5% with high risk at 5 years after HCT; $P < .01$; and 86.7% with standard risk vs 38.5% with high risk at 5 years after HCT; $P < .01$, respectively [Fig 2B]).

DISCUSSION

We found that BMT or PBSCT following a G-CSF–combined HDCA/CY/TBI regimen can provide favorable survival with a low NRM that is comparable to that of CBT in adult patients with myeloid malignant neoplasms. Moreover, this study demonstrates that this conditioning regimen can yield excellent outcomes, especially in patients with standard risk disease during HCT. There are several reasons why the intensified myeloablative conditioning provides favorable outcomes. First, the combination of G-CSF and HDCA increases the susceptibility of leukemic cells to cytarabine and thus reduces disease relapse [14,15]. Second, the addition of G-CSF–combined HDCA to conditioning promotes neutrophil engraftment [8,9], which can suppress infectious complications and thereby reduce NRM [8,9,14,15]. Third, we evaluated transplant cases after 2009. During this decade, the progression of support therapies, such as management of infection or organ failure, reduced NRM after HCT [16]. Therefore, the incidence of NRM in this study should be lower than that in previous studies, which evaluated intensified conditioning in the earlier period [1–5]. The reported cumulative incidences of NRM in these earlier studies, 30% to 40%, were higher than in this study [1–5]. Fourth, almost all patients in this study had low HCT-comorbidity index scores or were in good physical condition; therefore, low NRM rates were obtained [17].

Our study had several limitations. The results of this study should be compared with those of standard myeloablative conditioning, such as a CY/TBI regimen or non–G-CSF–combined conditioning, such as a HDCA/CY/TBI regimen, to indicate improvement in outcomes after BMT or PBSCT with a G-CSF–combined HDCA/CY/TBI regimen. This is a retrospective study including a limited number of patients; therefore, a prospective randomized study is warranted to confirm our results. Despite these limitations, our study

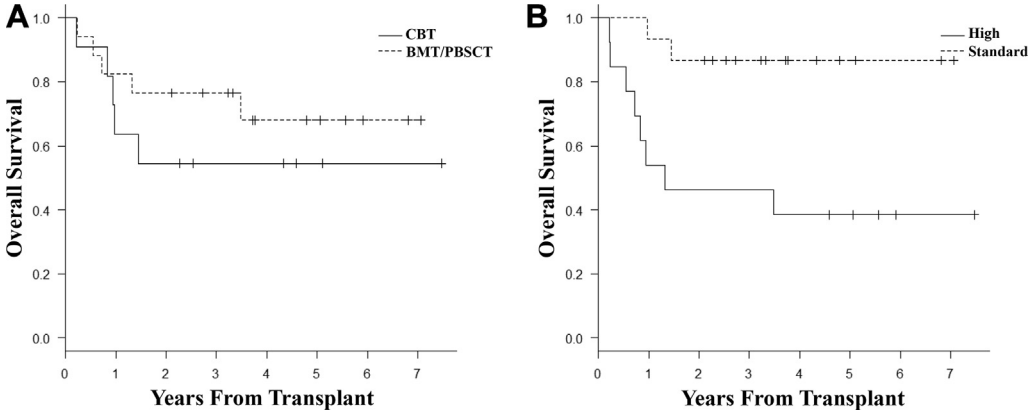

Fig 2. Overall survival according to **(A)** donor sources and **(B)** disease risk at transplant. BMT, bone marrow transplant; CBT, cord blood transplant; PBSCT, peripheral blood stem cell transplant.

suggests that intensification of conditioning can be one of the strategies for improving outcome after HCT in patients with good physical condition, among the recent progression of supportive therapies.

In conclusion, a G-CSF–combined HDCA/CY/TBI regimen is a promising conditioning regimen in adult patients with myeloid malignant neoplasms who undergo not only CBT but also BMT or PBSCT. Larger prospective studies are required to validate these results and to indicate the advantages of intensified conditioning.

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