Table 1. **Patients', disease, and transplant characteristics of pediatric and adult recipients of single-unit cord blood.**

Characteristics	Children (age<16) N	(%)	Adult (age>16) N	(%)
N. of transplants	498		1880	
Patient age at transplant				
Median (range)	5 (0-15)		49 (16-82)	
0-9 years	378	(76)		
10-19 years	120	(24)	88	(5)
20-29 years			236	(13)
30-39 years			317	(17)
40-49 years			351	(19)
50-59 years			492	(26)
≥60 years or older			396	(21)
Patient sex				
Male	275	(55)	1039	(55)
Female	223	(45)	841	(45)
Sex matching				
Matched	207	(42)	696	(37)
Male to female	114	(23)	391	(21)
Female to male	125	(25)	485	(26)
Unknown	52	(10)	308	(16)
Diagnosis				
AML	170	(34)	1115	(59)
ALL	290	(58)	418	(22)
CML	7	(1)	106	(6)
MDS	31	(6)	241	(13)
Disease status				
Standard	247	(50)	673	(36)
Advanced	236	(47)	1127	(60)
Unknown	15	(3)	80	(4)
ABO matching				
Matched	182	(37)	602	(32)
Minor mismatch	127	(26)	522	(28)
Major mismatch	113	(23)	451	(24)
Bidirectional	75	(15)	301	(16)
Unknown	1	(<1)	4	(<1)
HLA mismatched number				
Matched (6/6)	82	(16)	71	(4)
One locus mismatched (5/6)	222	(45)	309	(16)
Two loci mismatched (4/6)	158	(32)	1025	(55)
Three loci mismatched (3/6)	36	(7)	475	(25)
N. of cryopreserved nucleated cells (x10⁷/kg)				
Median	5.30		2.52	
Range	0.81-38.7		0.71-9.98	
N. of cryopreserved CD34-positive cells (x10⁵/kg)				
Median	1.68		0.83	
Range	0.072-65.66		0.07-14.02	
Preparative regimen*				
MAST				
CY+TBI	216	(43)	891	(47)
Other TBI regimen	93	(19)	162	(9)
BU+CY	86	(17)	65	(3)
Other non-TBI regimen	41	(8)	47	(3)
RIST				
FL+BU+other	6	(1)	172	(9)
FL+CY+other	12	(2)	119	(6)
FL+Mel+other	21	(4)	357	(19)
Other RIST	23	(5)	67	(4)
T-cell depletion *in vivo***				
ATG or ALG use	9	(2)	26	(1)
GVHD prophylaxis***				
Cyclosporine A + sMTX	157	(32)	748	(40)
Cyclosporine A + MMF/steroid	37	(7)	99	(5)
Cyclosporine A alone	31	(6)	142	(8)
Tacrolimus + sMTX	216	(43)	434	(23)
Tacrolimus + MMF/steroid	24	(5)	132	(7)
Tacrolimus alone	20	(4)	304	(16)
Others	13	(3)	21	(1)

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**CY: cyclophosphamide; CA: citarabine; BU: busulfan; TBI: total body irradiation; FL: fludarabine; Mel: melphalan, **ATG: antithymocyte globulin; ALG: antilymphocyte globulin; ***sMTX: short-term methotrexate; MMF: mycophenolate mofetil.*

Acute and chronic graft-versus-host disease

The risk of grade 2 to 4 acute GVHD was significantly higher in HLA-mismatched UCB pediatric recipients (RR=2.13, *P*=0.004 for 5/6; RR=2.65, *P*<0.001 for 4/6; RR=2.39, *P*=0.0015 for 3/6; P for trend 0.001) (Table 4). The risk of chronic GVHD and extensive-type chronic GVHD was also significantly higher in 4/6 UCB recipients (RR=2.99, *P*=0.005 for chronic GVHD, and RR=7.62, *P*=0.047 for extensive-type chronic GVHD), and the risks increased according to the number of mismatches (P for trend, 0.002 for chronic GVHD, 0.005 for extensive-type chronic GVHD). In adults, in contrast to the results in children, there were no differences in the risks of grade 2 to 4 acute GVHD in 5/6 and 4/6 UCB recipients (for grade 2 to 4 acute GVHD, RR=1.03, *P*=0.916 for 5/6, RR=1.27, *P*=0.276 for 4/6). The risk of grade 2 to 4 acute GVHD was higher for 3/6 (RR=1.72, *P*=0.017). In adult recipients, the risk of chronic GVHD was increased in recipients of 4/6 UCB (RR=1.90, *P*=0.04), however, there were no differences in the risk of extensive-type chronic GVHD (RR=1.15, *P*=0.758 for 5/6; RR=1.62, *P*=0.253 for 4/6; RR=1.28, *P*=0.574 for 3/6) (Table 4).

Effect of total nucleated cell dose on outcome

An increase in the cryopreserved total nucleated cell dose increased the incidence of neutrophil recovery in both children and adults, as well as the incidence of platelet recovery in children (Table 3). The cumulative incidences of neutrophil recovery were 94% for >10 x 10^7/kg, 88% for 5.0-9.9 x 10^7/kg, 82% for 2.5-4.9 x 10^7/kg, and 86% for <2.5 x 10^7/kg in children (*P*<0.001) (Figure 2A). The cell dose was significantly correlated with the recipient's age at transplant in children (the median ages were one year for >10 x 10^7/kg, 3 years for 5.0-9.9 x 10^7/kg, 8 years for 2.5-4.9 x 10^7/kg, and 12 years for <2.5 x 10^7/kg). The cumulative incidences of neutrophil recovery were 76% for >2.5 x 10^7/kg and 74% for <2.5 x 10^7/kg in adults (*P*=0.007) (Figure 2B). The cumulative incidences of TRM at three years post-transplant were 13% for >10 x 10^7/kg, 14% for 5.0-9.9 x 10^7/kg, 14% for 2.5-4.9 x 10^7/kg, and 14% for <2.5x10^7/kg in children (*P*=0.98) and 29% for >2.5x10^7/kg and 28% for <2.5 x 10^7/kg in adults (*P*=0.77) (*Online Supplementary Figure S2*). The probabilities of overall survival at three years post-transplant were 68% for >10x10^7/kg, 53% for 5.0-9.9 x 10^7/kg, 57% for 2.5-4.9 x 10^7/kg, and 55% for <2.5x10^7/kg in children (*P*=0.30) and 36% for >2.5 x 107/kg and 41% for <2.5x10^7/kg in adults (*P*=0.13). A lower total nucleated cell dose was neither associated with increased mortality in children or adults in multivariate analyses (Table 2). Thus, there was no combined effect of HLA disparity and total nucleated cell dose on mortality neither in children nor in adults (cumulative

incidence of TRM at three years post-transplant, 8% for 6/6, 11% for 5/6 and >5 x 10^7/kg, 11% for 5/6 and 2.5-4.9 x 10^7/kg, 0% for 5/6 and <2.5 x 10^7/kg, 23% for 4/6 and >5 x 10^7/kg, 24% for 4/6 and 2.5-4.9 x 10^7/kg, 25% for 4/6 and <2.5 x 10^7/kg in children, and 23% for 6/6, 29% for 5/6 and >2.5 x 107/kg, 30% for 5/6 and <2.5 x 10^7/kg, 27% for 4/6 and >2.5 x 10^7/kg, 27% for 4/6 and <2.5 x 10^7/kg in adults (*Online Supplementary Figure S3*).

Association of outcomes with the type of HLA mismatches for 4/6 adult recipients

The large number of adult recipients of 4/6 CB enabled us to analyze association of outcomes with the type of HLA mismatches in this population. The number of recipients were 7 for HLA-A double mismatch, 170 for HLA-A and HLA-B mismatch, 190 for HLA-A and HLA-DRB1 mismatch, 36 for HLA-B double mismatch, 581 for HLA-B and HLA-DRB1 mismatch, and 41 for HLA-DRB1 double mismatch. With adjusted analyses, adjusted with same variables in the final model of all adult recipients, there was no significant effect of HLA mismatch types on overall mortality with HLA-A and HLA-B mismatch as the reference (*Online Supplementary Table S3*). The risk of relapse was significantly decreased in HLA-A and HLA-DRB1

Table 2. **Multivariate analyses of overall survival, relapse, and transplant-related mortality.**

Outcome	N.	Overall mortality RR	95%CI	P	Relapse RR	95%CI	P	Transplant related mortality RR	95%CI	P
Children 15 years or younger										
HLA disparity										
Matched (6/6)	82	1.00			1.00			1.00		
5/6	222	1.07	(0.68-1.69)	0.765	1.06	(0.68-1.65)	0.794	1.29	(0.52-3.23)	0.58
4/6	158	1.61	(1.02-2.56)	0.042	0.77	(0.48-1.24)	0.282	3.55	(1.47-8.58)	0.005
3/6	36	1.25	(0.65-2.42)	0.498	0.91	(0.45-1.86)	0.802	1.56	(0.43-5.63)	0.497
Total nucleated cell dose										
≥10.0x10^7/kg	85	1.00			1.00			1.00		
5.0-9.9x10^7/kg	169	1.14	(0.72-1.79)	0.579	1.10	(0.69-1.75)	0.684	0.82	(0.40-1.68)	0.592
2.5-4.9x10^7/kg	190	0.92	(0.58-1.45)	0.707	0.90	(0.56-1.44)	0.651	0.90	(0.45-1.80)	0.77
<2.5x10^7/kg	43	0.88	(0.47-1.67)	0.701	0.98	(0.53-1.83)	0.961	0.67	(0.24-1.88)	0.443
Adults 16 years or older										
HLA disparity										
Matched (6/6)	71	1.00			1.00			1.00		
5/6	309	0.99	(0.71-1.38)	0.944	0.70	(0.47-1.04)	0.075	1.41	(0.83-2.41)	0.205
4/6	1025	0.88	(0.65-1.21)	0.436	0.67	(0.47-0.97)	0.034	1.24	(0.75-2.04)	0.408
3/6	475	0.95	(0.69-1.31)	0.751	0.70	(0.48-1.03)	0.07	1.29	(0.77-2.16)	0.339
Total nucleated cell dose										
≥3.0x10^7/kg	439	1.00			1.00			1.00		
2.5-2.9x10^7/kg	492	0.99	(0.83-1.17)	0.876	0.86	(0.70-1.06)	0.167	1.10	(0.86-1.42)	0.445
2.0-2.4x10^7/kg	705	0.86	(0.72-1.01)	0.06	0.79	(0.65-0.97)	0.021	1.05	(0.83-1.33)	0.694
<2.0x10^7/kg	183	0.93	(0.73-1.18)	0.562	0.79	(0.59-1.07)	0.126	1.00	(0.70-1.45)	0.983

For overall mortality, other predictive variables were advanced disease status at transplant in children, and age at transplant over 50 years, male sex, advanced disease status at transplant, chronic myeloid leukemia (associated with a lower risk of mortality), and reduced-intensity conditioning in adults. For relapse, other predictive variables were advanced disease status at transplant, and acute lymphoblastic leukemia or myelodysplastic syndrome (associated with a lower risk of relapse) in children, and advanced disease status at transplant and myelodysplastic syndrome (associated with a lower risk of relapse) in adults. For transplant-related mortality, there was no other predictive variable in children. Other predictive variables for adults were age at transplant over 50 years and female to male donor-recipient sex mismatch.

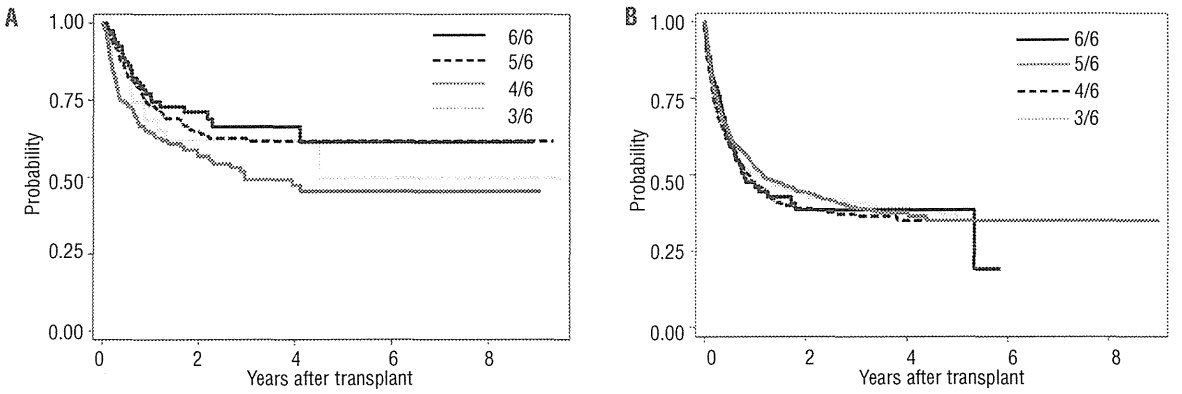

Figure 1. **Unadjusted probabilities of overall survival in HLA disparity groups for pediatric (A) and adult (B) recipients with leukemia.** (A) In children, the unadjusted probabilities of survival at three years post-transplant were 66% for recipients of HLA matched (6/6), 62% for one-locus-mismatched (5/6), 45% for two-loci-mismatched (4/6), and 62% for three-loci-mismatched (3/6) single-unit unrelated cord blood (*P*=0.032). (B) In adults, these probabilities were 38% 37%, 39%, and 40% respectively (*P*=0.567) (B).

mismatch, HLA-B and HLA-DRB1 mismatch, and HLA-DRB1 double mismatch recipients (RR=0.70, *P*=0.045; RR=0.76, *P*=0.047; and RR=0.46, *P*=0.03, respectively). The risk of transplant-related mortality was significantly increased in HLA-DRB1 double mismatch recipients (RR=2.06, *P*=0.025). There was no significant effect of HLA mismatch types for risks of grade 2 to 4 and grade 3 to 4 acute GVHD (*Online Supplementary Table S3*).

Discussion

Our main objective was to assess the effect of HLA disparity on survival after single-unit UCBT in children and adults, and to obtain data that could be useful for the selection of an appropriate cord blood unit for patients with leukemia. Our study is the first to assess the effect of UCB HLA-matching on the transplant outcome in a large

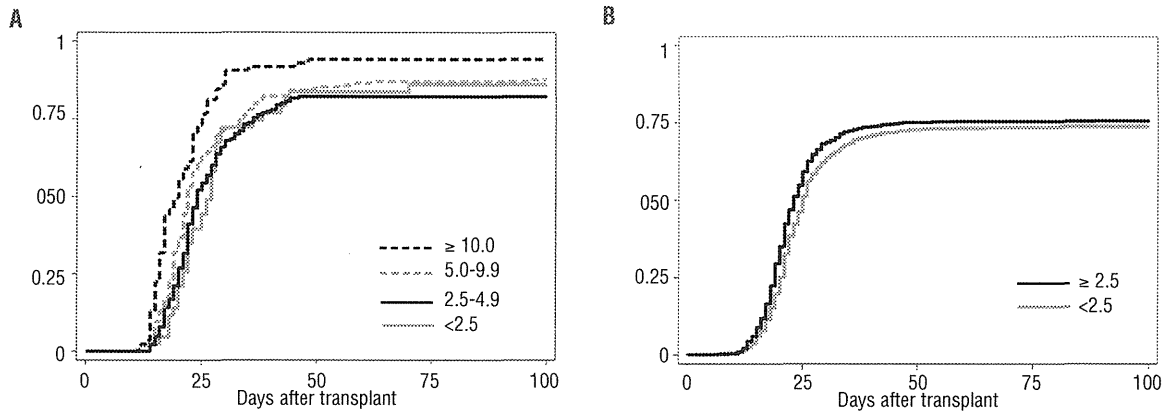

Figure 2. **Unadjusted cumulative incidences of neutrophil recovery in total nucleated cell dose groups for pediatric (A) and adult (B) recipients with leukemia. (A) In children, the unadjusted cumulative incidences of neutrophil recovery were 94% for >10x10^7/kg, 88% for 5.0-9.9 x10^7/kg, 82% for 2.5-4.9x10^7/kg, and 86% for <2.5x10^7/kg (*P*<0.001). (B) In adults, these incidences were 76% for >2.5x10^7/kg and 74% for <2.5 x10^7/kg (*P*=0.007).**

Table 3. **Multivariate analyses of neutrophil and platelet recovery.**

Outcome	Children ≤16 years or younger N	RR	95%CI	*P* value		Adults ≥16 years or older N	RR	95%CI	*P*
Neutrophil recovery									
HLA disparity									
Matched (6/6)	82	1.00				71	1.00		
5/6	222	1.03	(0.77-1.39)	0.823		309	0.89	(0.66-1.19)	0.436
4/6	158	0.96	(0.71-1.30)	0.799		1025	0.92	(0.70-1.22)	0.576
3/6	36	0.67	(0.44-1.03)	0.068		475	0.84	(0.64-1.12)	0.243
Total nucleated cell dose									
≥>10.0x10^7/kg	85	1.00			≥3.0x10^7/kg	439	1.00		
5.0-9.9x10^7/kg	169	0.66	(0.49-0.89)	0.007	2.5-2.9x10^7/kg	492	0.84	(0.72-0.97)	0.021
2.5-4.9x10^7/kg	190	0.50	(0.37-0.67)	<0.001	2.0-2.4x10^7/kg	705	0.79	(0.68-0.90)	0.001
<2.5x10^7/kg	43	0.54	(0.38-0.77)	0.001	<2.0x10^7/kg	183	0.78	(0.64-0.94)	0.009
Platelet recovery									
HLA disparity									
Matched (6/6)	82	1.00				71	1.00		
5/6	222	0.89	(0.66-1.20)	0.438		309	1.05	(0.73-1.52)	0.775
4/6	158	0.75	(0.54-1.05)	0.09		1025	1.05	(0.74-1.48)	0.791
3/6	36	0.71	(0.44-1.15)	0.164		475	0.99	(0.69-1.41)	0.951
Total nucleated cell dose									
≥10.0x10^7/kg	85	1.00			≥3.0x10^7/kg	439	1.00		
5.0-9.9x10^7/kg	169	0.93	(0.68-1.29)	0.681	2.5-2.9x10^7/kg	492	0.84	(0.70-1.01)	0.058
2.5-4.9x10^7/kg	190	0.70	(0.51-0.97)	0.03	2.0-2.4x10^7/kg	705	0.86	(0.73-1.02)	0.078
<2.5x10^7/kg	43	0.70	(0.45-1.07)	0.101	<2.0x10^7/kg	183	0.72	(0.57-0.91)	0.007

For neutrophil recovery, other predictive variables were acute lymphoblastic leukemia in children (with a higher neutrophil recovery), and advanced disease status at transplant in adults. For platelet recovery, other predictive variables were advanced disease status at transplant in children, and age at transplant over 50 years, male sex, and advanced disease status at transplant in adults.

Table 4. **Multivariate analyses of grade 2 to 4/grade 3 to 4 acute graft-*versus*-host disease, and chronic/extensive-type chronic graft-*versus*-host disease.**

Outcome	Grade 2 to 4 acute GVHD N	RR	95%CI	P	Grade 3 to 4 acute GVHD RR	95%CI	P	Chronic GVHD N	RR	95%CI	P	Extensive-type chronic GVHD RR	95%CI	P
Children 15 years or younger														
HLA disparity														
Matched (6/6)	72	1.00			1.00			67	1.00			1.00		
5/6	196	2.13	(1.28-3.58)	0.004	1.75	(0.73-4.24)	0.212	186	1.79	(0.85-3.75)	0.123	4.15	(0.54-31.81)	0.17
4/6	136	2.65	(1.55-4.52)	<0.001	2.25	(0.94-5.41)	0.07	114	2.99	(1.42-6.30)	0.004	7.62	(1.03-56.63)	0.047
3/6	28	2.39	(1.18-4.84)	0.015	2.60	(0.82-8.26)	0.105	23	2.61	(0.96-7.11)	0.061	7.49	(0.81-69.63)	0.077
Adults 16 years or older														
HLA disparity														
Matched (6/6)	56	1.00			1.00			49	1.00			1.00		
5/6	227	1.03	(0.64-1.65)	0.916	0.95	(0.38-2.37)	0.919	193	1.58	(0.83-3.02)	0.161	1.15	(0.47-2.80)	0.758
4/6	765	1.27	(0.82-1.97)	0.276	1.27	(0.55-2.94)	0.573	650	1.90	(1.03-3.51)	0.04	1.62	(0.71-3.72)	0.253
3/6	341	1.72	(1.10-2.70)	0.017	1.13	(0.47-2.68)	0.788	288	1.81	(0.96-3.38)	0.065	1.28	(0.54-3.02)	0.574

For grade 2 to 4 acute GVHD, other predictive variables were total nucleated cell dose (>10x10^7/kg as the reference, RR=1.94 P=0.009 for 5.0-9.9x107/kg, RR=1.73 P=0.028 for 2.5-4.9x107/kg, and R=1.68 P=0.094 for <2.5x10^7/kg) in children, and cyclosporine-based GVHD prophylaxis (vs. tacrolimus-based) in adults. For grade 3 to 4 acute GVHD, male sex and advanced disease status in children, and male sex and male to female donor-recipient sex mismatch and reduced-intensity conditioning in adults. For chronic GVHD, no other predictive variables in children, and other predictive variable for adults was ABO major mismatch, and male to female sex mismatch and advanced risk disease status for decreased risk. For extensive-type chronic GVHD, no other predictive variables in children, and other predictive variable for adults was ABO major mismatch.

number of adult recipients. Our findings in children were similar to those in previous reports.[9,17,18,31,32] An increase in the number of HLA mismatches resulted in an increased risk of acute and chronic GVHD, which led to an increased risk of overall and transplant-related mortality. In contrast to the results in children, the probability of overall or relapse-free survival did not decrease with the number of mismatched antigens in adults. An increase in the number of HLA mismatches in UCB increased the incidence of cGVHD in 4/6 CB recipients; however, there was no increase in the risk of grade 2 to 4 or severe acute GVHD, or extensive-type chronic GVHD. These differences may have contributed to the decreased incidence of relapse without affecting TRM after HLA-mismatched UCBT in adults.

A major potential contributor to the different findings in children and adults is the difference in the nucleated cell dose. There was a dramatic difference in the nucleated cell dose between children and adults. TNC dose in adults is highly concentrated in a very small, low-dose area that is quite different from the doses used in children in our study and from the doses in previous reports, mainly in pediatric recipients.[9,18,32] A positive effect on the transplant outcome with a decreased incidence of acute GVHD and lower mortality with HLA matching might only be seen in the setting of pediatric recipients who receive cord blood with a larger cell dose compared to adults. A report from Eurocord of 171 adult recipients of single-unit CBT did not see a decrease in the probability of overall or relapse-free survival with the number of mismatched antigens.[33] A more recent collaborative study by the Center for International Blood and Marrow Transplant Research, the New York Blood Center National Cord Blood Program, and the Eurocord-Netcord registry with 514 adult recipients did not observe an increase in mortality after HLA-mismatched UCBT.[34]

Another potential cause of different findings in children and adults is differences in diagnosis. Adult recipients had a significantly greater proportion of patients with myeloid malignancy. The incidence of a graft-*versus*-leukemia effect is reportedly higher in myeloid malignancy.[35-37] The decreased risk of relapse with a significant graft-*versus*-leukemia effect in HLA-mismatched UCB recipients was also more prominent in adult recipients with acute myeloid leukemia in our study. Furthermore, there were differences in disease risk between children and adults. Only 36% of adults were in a standard-risk disease status at transplant, while this value was 50% in children. Although we had adjusted for the disease status at transplant, we cannot rule out the possibility that these differences influenced the results.

An increase in the total nucleated cell dose increased the neutrophil recovery rate in both children and adults, consistent with other reports.[18,31-33] A lower total nucleated cell dose was not associated with increased transplant-related or overall mortality in our cohort, thus, we did not see a combined effect of HLA disparity and total nucleated cell dose. This differs from the findings of a recent report from New York Cord Blood Bank.[18] In our cohort, a lower cell dose was associated with a slower recovery; however, the differences in the overall incidences of neutrophil recovery between cell dose groups were small, especially in the adult cohort. This may explain our finding that a lower total nucleated cell dose was not associated with increased mortality. Another probable reason for the different findings is that for our analyses we separated children and adults. A small percentage of older adults who received lower cell dose CB included in the subjects of previous studies may have affected increased mortality with lower cell doses. Lastly, TNC dose in adults is highly concentrated in a very small, low-dose area (nearly 70% lie in the range of 2.0-3.0 x 10^7/kg) which is a unique finding for adult recipients of single-unit cord blood in Japan. Therefore, differences in cell doses between the TNC dose groups is quite small, which is suspected to be one of the reasons for these findings. The results of our study support the current recommended cut-off TNC dose for cord blood search in Japan, which is 2.0 x 10^7/kg.

Although information is still limited because of the limited number of 6/6 and 5/6 CB adult recipients, the large number of adult recipients of 4/6 CB enabled us to analyze the association of outcomes with the type of HLA mismatches in this population. There was no effect of HLA mismatch type on overall mortality; therefore, there is no

preference recommendation for HLA mismatch types from our study. The increase in the number of HLA-DRB1 mismatch was associated with decreased mortality; however, it is important to note that HLA-DRB1 double mismatch was associated with increased transplant-related mortality.

This study included a large number of HLA-A, HLA-B, low-resolution and HLA-DRB1 high-resolution typed CB recipients, but there are limitations. UCB selection is mainly influenced by the availability of an acceptable cell dose, but is also influenced by many unmeasured factors that can affect the outcome. Although we adjusted for known risk factors and disparities between groups, we cannot rule out the influence of a potential selection bias. Another limitation involves the results for 3/6. Since, in current practice in Japan, HLA-DR typing for UCB unit selection is performed at low resolution, with a preference of up to two HLAantigen-mismatched UCB units, most (97%) of the HLA-A, HLA-B, low-resolution and HLA-DRB1 high-resolution 3/6 UCB in the present study were selected as one- or two-antigen-mismatched for theHLA-A, HLA-B, and HLA-DR low-resolution level. If we consider the effect of the current practice for UCB unit selection regarding 3/6 UCB, our conclusions should only apply to HLA-A, HLA-B, and HLA-DRB1 or HLA-A, HLA-B, and HLA-DR zero- to two-mismatched UCBT. Furthermore, we may have underestimated the impact of HLA-matching, since we did not have enough data to include low- or high-resolution information on HLA-C matching, which was recently reported to affect mortality.[38]

In conclusion, we found that the effects of HLA disparity on transplant outcome differed between children and adults. In children, an increased number of mismatched HLA loci correlated with an increased risk of mortality. These findings support the selection of a UCB unit with HLA 6/6 followed by 5/6, consistent with the recommendations from the US and Europe. In adults, there was no increase in mortality with an increase in the number of mismatched HLA loci. In this case, a UCB unit with up to 4/6 can be selected if transplant is urgently needed.

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Authorship and Disclosures

Information on authorship, contributions, and financial & other disclosures was provided by the authors and is available with the online version of this article at www.haematologica.org.

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ORIGINAL ARTICLE

Changes in incidence and causes of non-relapse mortality after allogeneic hematopoietic cell transplantation in patients with acute leukemia/myelodysplastic syndrome: an analysis of the Japan Transplant Outcome Registry

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The outcomes for allogeneic hematopoietic cell transplantation (allo-HCT) are heavily influenced by non-relapse mortality (NRM). We retrospectively assessed the changes in the incidence and causes of NRM after allo-HCT over the past 12 years. NRM, relapse rate and OS were analyzed using the Japan transplant outcome database of 6501 adult patients with acute leukemia or myelodysplastic syndrome who received their first allo-HCT in remission from 1997 through 2008. In multivariate analysis in patients aged 16–49 years, the adjusted hazard ratios (HRs) for NRM for 2001–2004 and 2005–2008 were 0.78 (95% confidence interval, 0.65–0.93) and 0.64 (0.54–0.78), respectively, compared with 1997–2000. The HR for overall mortality in 2005–2008 was 0.81 (0.70–0.93) compared with 1997–2000. In patients aged 50–70 years, the HRs for NRM and overall mortality in 2005–2008 were 0.56 (0.46–0.68) and 0.66 (0.47–0.93), respectively, compared with those in 2001–2004. We found that causes of death that contributed to the changes in NRM varied among subgroups. In conclusion, our study indicated that the incidence of NRM after allo-HCT has significantly decreased over the past 12 years, which has led to an improvement of OS, and also showed reductions in NRM in subgroups consisting of older patients and those who received unrelated cord blood transplantation.

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Keywords: leukemia; allogeneic hematopoietic cell transplantation; non-relapse mortality; GVHD; cord blood transplantation

INTRODUCTION

Allogeneic hematopoietic cell transplantation (allo-HCT) has been recognized as a potent strategy for curing hematological malignancies. However, there have always been concerns about the risk of non-relapse mortality (NRM). As the risk of relapse is known to be significantly reduced after allo-HCT, the outcome of and indications for allo-HCT are heavily influenced by the risk for NRM.

Over the past few decades, many changes have been made to improve the outcome after allo-HCT, including improvements in the conditioning regimen, donor selection, and prophylaxis and treatment for organ complications, GVHD and infectious diseases, which have led to a reduction in NRM.[1–4]

Although an improvement in NRM has been reported in relatively younger patients who have received allo-HCT from a BM or peripheral blood (PB) donor, NRM has not been fully examined in other settings, such as in elderly patients, or in cord blood (CB) transplantation.

To evaluate the effects of these advances, we retrospectively assessed the changes in the incidence and causes of NRM over the past 12 years, using a nationwide registry database of more than 6000 patients who received various types of allo-HCT.

PATIENTS AND METHODS

Data source

The clinical data were extracted from a nationwide transplant outcome registry database provided by the Japan Society for Hematopoietic Cell Transplantation (JSHCT), the Japan Marrow Donor Program (JMDP) and the Japan Cord Blood Bank Network (JCBBN). The JSHCT collect clinical data through the Transplant Registry Unified Management Program, as described previously.[5] This study was approved by the data management committees of JSHCT, JMDP and JCBBN, and by the Institutional Review Board at the National Cancer Center Hospital.

Patients and definitions

We evaluated the data on patients aged between 16 and 70 years who had AML, acute lymphocytic leukemia (ALL) or myelodysplastic syndrome (MDS), and who received their first allo-HCT between 1997 and 2008. We compared the incidence of NRM after allo-HCT in three consecutive 4-year

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periods (1997–2000, 2001–2004 and 2005–2008) for younger patients (16–49 years), and in the latter two periods for older patients (50–70 years). NRM was defined as death without recurrent disease after allo-HCT. Analyses were performed for patients with acute leukemia/MDS in remission or low-risk MDS (refractory anemia with or without ringed sideroblast: RA/RARS). Analyses were performed on the basis of patients' age (16–49 years and 50–70 years) and donor source (HLA-6/6-serum-matched or 1-Ag-mismatched related, unrelated BM and unrelated CB). In the era considered by this study, only BM from unrelated volunteer donors was used in Japan. In 2003, JMDP nationally recommended DNA typing of HLA-A and B, as well as HLA-DRB1. Since 2005, JMDP required all the candidates of unrelated allo-HCT to examine high-resolution typing of HLA-A, B and DRB1, and also recommended high-resolution typing of the C-locus. Conditioning regimens were classified as indicated by Giralt *et al.*[6] The incidences of mortality associated with GVHD, infection and organ failure were analyzed. In patients who had multiple causes among GVHD, infection and organ failure, information regarding the main cause of death was prioritized.

Statistical analysis

Data were retrospectively reviewed and analyzed as of June 2011. Among the three time periods, patient characteristics were compared using the χ^2 test. The primary end point of the study was NRM after allo-HCT. Probabilities of NRM and relapse were estimated with the use of cumulative incidence curves, with relapse viewed as a competing risk of NRM, and with NRM viewed as a competing risk of relapse. The Pepe and Mori test was used to evaluate the differences between groups. For the 151 patients (2%) who were known to have relapsed but whose date of relapse was unavailable, mid-point imputation was performed by substituting the midpoint from HCT to date of last contact as the date of relapse. The probability of OS was estimated using the Kaplan-Meier product limit method, and 95% confidence intervals (CIs) were calculated using the Greenwood formula. To compare the OS between groups, the log-rank test was used. Incidences of NRM, relapse and OS were estimated as probabilities at 3 years from allo-HCT. Multivariate analyses for NRM and relapse were performed using competing risk regression by the method of Fine and Gray, and for OS using a Cox proportional hazard regression model. The multivariate analyses were performed separately among patients aged 16–49 years and patients aged 50–70 years, where the year of allo-HCT (1997–2000 vs 2001–2004 or 2005–2008 among younger patients, 2001–2004 vs 2005–2008 among older patients or those who received unrelated CB transplantation (UCBT)), disease type (AML vs ALL or MDS), patient age (16–29 years vs 30–39 or 40–49 among younger patients, 50–59 vs 60–70 among older patients), patient gender (male vs female), donor source (HLA-6/6-Ag-matched sibling vs other family donors, HLA-6/6-Ag-matched unrelated BM, mismatched unrelated BM or unrelated CB) and conditioning regimens (myeloablative vs reduced-intensity) were considered as covariates. Multivariate analyses were also performed separately for those receiving related allo-HCT, where HLA-6/6-Ag-matched sibling vs other family donors were considered as covariates for the donor source, those receiving unrelated BM transplantation (UBMT), where HLA-6/6-Ag-matched unrelated BM vs mismatched BM were considered as covariates, and those who received UCBT, where the covariates above were examined other than the donor source. We considered two-sided *P*-values of <0.05 to be statistically significant. Statistical analyses were performed with SAS version 9.1.3 (SAS, Cary, NC, USA) and the SPSS software version 11.0.1 (SPSS, Chicago, IL, USA).

RESULTS

Patients

A total of 6501 patients registered from 266 institutions across the country[5] were analyzed, with a median age of 40 years and a median follow-up of 39 months. Characteristics of the patients and

Table 1. Patients' characteristics according to the time period of transplant

Characteristics	*1997–2000,* N *(%)*	*2001–2004,* N *(%)*	*2005–2008,* N *(%)*	P
Total number of patients	1354	2292	2855	
Age at transplant (years)				<0.001
16–34	740 (55)	892 (39)	862 (30)	
35–49	491 (36)	783 (34)	939 (33)	
50–59	116 (9)	489 (21)	743 (26)	
60–70	7 (1)	128 (6)	311 (11)	
Donor source				<0.001
Related BM	511 (38)	367 (16)	504 (18)	
Related peripheral blood	158 (12)	546 (24)	456 (16)	
Unrelated BM	588 (43)	998 (44)	1312 (46)	
Unrelated cord blood	14 (1)	321 (14)	534 (19)	
Others	83 (6)	60 (3)	49 (2)	
Disease type				0.991
AML	699 (52)	1226 (53)	1516 (53)	
ALL	505 (37)	744 (32)	949 (33)	
MDS	150 (11)	322 (14)	390 (14)	
Disease status				0.001
CR1	811 (60)	1288 (56)	1802 (63)	
CR2	311 (23)	552 (24)	654 (63)	
CR3 or beyond	76 (6)	96 (4)	77 (3)	
MDS RA/RARS	83 (6)	202 (9)	267 (9)	
Other remission state/no detailed data	73 (5)	154 (7)	55 (2)	
Conditioning				<0.001
Myeloablative	1131 (84)	1585 (69)	1788 (63)	
Reduced-intensity	21 (2)	394 (17)	689 (24)	
Not categorized	202 (15)	313 (14)	378 (13)	
GVHD prophylaxis				<0.001
CYA-based	1041 (77)	1367 (60)	1354 (47)	
Tacrolimus-based	270 (20)	825 (36)	1373 (48)	
No data available	43 (3)	100 (4)	128 (4)	

Abbreviations: MDS = myelodysplastic syndrome; RA/RARS = refractory anemia with or without ringed sideroblast.

transplantation procedures according to the time period are shown in Table 1. The overall proportions of AML, ALL and MDS were 53%, 34% and 13%, respectively. A total of 1354, 2292 and 2855 allo-HCTs were performed in 1997–2000, 2001–2004 and 2005–2008, respectively. The number and proportion of patients aged 50–70 years (1997–2000, $n=123$, 9%; 2001–2004, $n=617$, 27%; 2005–2008, $n=1054$, 37%), allo-HCT from an unrelated CB donor ($n=14$, 1%; $n=321$, 14%; $n=534$, 19%), and the use of a reduced-intensity conditioning regimen ($n=21$, 2%; $n=394$, 17%; $n=689$, 24%) increased over the three periods. Most of the myeloablative conditioning regimens (96%) consisted of high-dose CY with TBI or BU. Tacrolimus-based GVHD prophylaxis increased, especially in allo-HCT from an unrelated BM and CB donor (BM: $n=218$, 37%; $n=579$, 58%; $n=945$, 72%; CB: $n=3$, 21%; $n=99$, 31%; $n=229$, 43%).

Outcomes of allo-HCT over the three periods

The incidence of NRM of the entire 6501 patients was 23% at 3 years after allo-HCT (Figure 1a). Overall, 265 patients died of acute or chronic GVHD (median OS: 143 days, range: 18–3360), 497 died of infection (median OS: 116 days, range: 0–3184) and 500 died of organ failure (median OS: 145 days, range: 0–4013).

Older patients had a significantly higher incidence of NRM than younger patients (31% vs 20%, $P<0.001$, Figure 1b). The donor source significantly affected the incidence of NRM, and unrelated CB had the highest risk of NRM (related, 17%; unrelated BM, 25%; unrelated CB, 31%, $P<0.001$, Figure 1c). In a comparison of the outcome after allo-HCT among the three time periods in the overall 6501 patients (Figure 2), there were no linear improvements in NRM and OS over the three periods (NRM: 23%, 25% and 21%; OS: 61%, 57% and 60% at 3 years after allo-HCT). By the multivariate analysis that adjusted for disease type, patient age, patient gender, donor source and conditioning regimens, in younger patients (Table 2), the hazard ratios (HRs) for NRM in 2001–2004 and 2005–2008 compared with 1997–2000 were 0.78 (95% CI 0.65–0.93, $P=0.005$) and 0.64 (95% CI 0.54–0.78, $P<0.001$), respectively. The HR for overall mortality in 2005–2008 was significantly lower than that in 1997–2000 (HR 0.81, 95% CI 0.70–0.93, $P=0.004$). The HRs for relapse did not differ significantly among the periods. In older patients, the HRs for NRM and overall mortality in 2005–2008 compared with 2001–2004 were 0.56 (95% CI 0.46–0.68, $P<0.001$) and 0.66 (95% CI 0.47–0.93, $P=0.017$), respectively. However, the HR for relapse in 2005–2008 significantly increased (HR 1.53, 95% CI 1.20–1.97, $P=0.001$).

Allo-HCT from an HLA-matched or 1-Ag-mismatched related donor

In younger patients who received allo-HCT from a related donor (Figure 3a), the incidence of NRM remained rather low throughout the 12 years. Although NRM and OS slightly improved in 2005–2008, the differences were not statistically significant.

In older patients who received allo-HCT from a related donor, NRM was significantly reduced in 2005–2008 compared with 2001–2004 (Figure 3b, HR 0.62, 95% CI 0.44–0.88, $P=0.007$, Table 2). The incidences of death associated with organ failure and GVHD were significantly reduced in 2005–2008 (organ failure, 11 and 6%, $P=0.007$; GVHD, 6 and 3%, $P=0.015$, Figure 4a). In contrast, a significant increase in relapse was observed in 2005–2008 compared with 2001–2004 (21 and 36%, $P<0.001$, data not shown), and the same result was also shown by a multivariate analysis (HR 1.97, 95% CI 1.38–2.81, $P<0.001$, Table 2). This result remained the same when the analyses were restricted to HCT using reduced-intensity regimens or myeloablative regimens. Consequently, the improvement in OS in 2005–2008 was not statistically significant (Figure 3b and Table 2).

Allo-HCT from an unrelated BM donor

A significant reduction in NRM was seen over the three periods among younger patients who received allo-HCT from an unrelated BM donor (Figure 3c), with the HRs of 0.69 (95% CI 0.55–0.88, $P=0.003$) and 0.61 (95% CI 0.47–0.78, $P<0.001$) in 2001–2004 and 2005–2008, respectively (Table 2). The incidences of death associated with GVHD and organ failure were significantly reduced over the three periods (GVHD, 7, 4 and 4%, $P=0.011$; organ failure, 12, 10 and 8%, $P=0.002$, Figure 4b). OS significantly improved in 2005–2008 (Figure 3c and Table 2).

In older patients who received allo-HCT from an unrelated BM donor, NRM and OS significantly improved in 2005–2008 compared with 2001–2004 (Figure 3d). The HR for NRM in 2005–2008 was 0.58 (95% CI 0.41–0.82, $P=0.002$). The incidences of death associated with infection and organ failure were reduced in 2005–2008 (infection, 14 and 10%, $P=0.054$; organ failure,

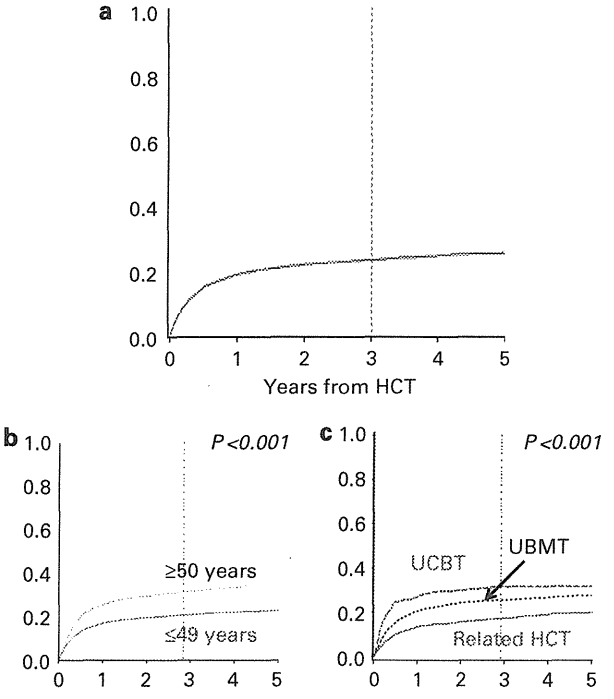

Figure 1. NRM over the past 12 years among 6501 patients who received allo-HCT in remission is shown in (**a**). NRM according to age (**b**) and donor source (**c**) are also shown.

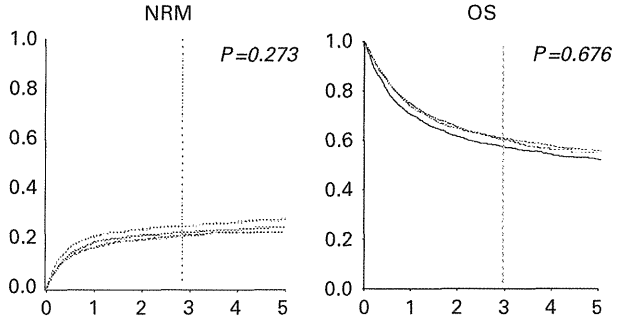

Figure 2. NRM and OS at 4-year periods (1997–2000, solid line; 2001–2004, dotted line; 2005–2008, dashed line) in the overall patients.

Table 2. Multivariate analyses for NRM, relapse and overall mortality after allo-HCT among the three periods

	All patients N = *6501*			*Related HCT* N = *2542*			*UBMT* N = *2898*			*UCBT* N = *869*		
	HR	*95% CI*	P *value*	*HR*	*95% CI*	P *value*	*HR*	*95% CI*	P *value*	*HR*	*95% CI*	P *value*
		N = 4707			*N* = 1846			*N* = 2202			*N* = 518	
Patient age at transplant, 16–49 years												
NRM												
1997–2000	1.00			1.00			1.00					
2001–2004	0.78	(0.65–0.93)	0.005	1.00	(0.75–1.33)	0.980	0.69	(0.55–0.88)	0.003	1.00		
2005–2008	0.64	(0.54–0.78)	<0.001	0.62	(0.44–0.88)	0.007	0.61	(0.47–0.78)	<0.001	1.04	(0.72–1.51)	0.830
Relapse												
1997–2000	1.00			1.00			1.00					
2001–2004	1.16	(0.98–1.37)	0.094	0.95	(0.74–1.21)	0.650	1.39	(1.39–1.06)	0.019	1.00		
2005–2008	1.12	(0.94–1.34)	0.220	1.20	(0.94–1.52)	0.150	1.20	(0.89–1.61)	0.240	0.66	(0.43–1.00)	0.049
Overall mortality												
1997–2000	1.00			1.00			1.00					
2001–2004	0.94	(0.82–1.06)	0.310	1.00	(0.82–1.22)	0.990	0.88	(0.73–1.06)	0.188	1.00		
2005–2008	0.81	(0.70–0.93)	0.004	0.89	(0.71–1.11)	0.285	0.77	(0.62–0.94)	0.010	0.84	(0.57–1.23)	0.373
		N = 1794			*N* = 696			*N* = 696			*N* = 351	
Patient age at transplant, 50–70 years												
NRM												
2001–2004	1.00			1.00			1.00			1.00		
2005–2008	0.56	(0.46–0.68)	<0.001	0.49	(0.33–0.71)	<0.001	0.58	(0.41–0.82)	0.002	0.57	(0.40–0.83)	0.003
Relapse												
2001–2004	1.00			1.00			1.00			1.00		
2005–2008	1.53	(1.20–1.97)	0.001	1.97	(1.38–2.81)	<0.001	1.46	(0.93–2.28)	0.100	0.96	(0.59–1.58)	0.880
Overall mortality												
2001–2004	1.00			1.00			1.00			1.00		
2005–2008	0.66	(0.47–0.93)	0.017	0.87	(0.67–1.15)	0.334	0.82	(0.61–1.09)	0.169	0.67	(0.49–0.91)	0.010

Abbreviations: CI = confidence interval; HCT = hematopoietic cell transplantation; HR = hazard ratio; NRM = non-relapse mortality; UBMT = unrelated BM transplantation; UCBT = unrelated cord blood transplantation. Year of allo-HCT (1997–2000 vs 2001–2004 or 2005–2008 among younger patients, 2001–2004 vs 2005–2008 among older patients or those who received UCBT), disease type (AML vs ALL or MDS), patient age (16–29 years vs 30–39 or 40–49 among younger patients, 50–59 vs 60–70 among older patients), patient gender (male vs female), donor source (HLA-matched sibling vs other family donors, HLA-matched unrelated BM, mismatched unrelated BM or unrelated CB), and conditioning regimens (myeloablative vs reduced-intensity) were considered as covariates. In the analysis for related HCT, donor source (HLA-matched sibling vs other family donors), year of allo-HCT, disease type, patient age, patient gender and conditioning regimens were considered as covariates. In the analysis for UBMT, donor source (HLA-matched BM vs mismatched unrelated BM), year of allo-HCT, disease type, patient age, patient gender and conditioning regimens were considered as covariates. In the analysis for UCBT, year of allo-HCT, disease type, patient age, patient gender and conditioning regimens were considered.

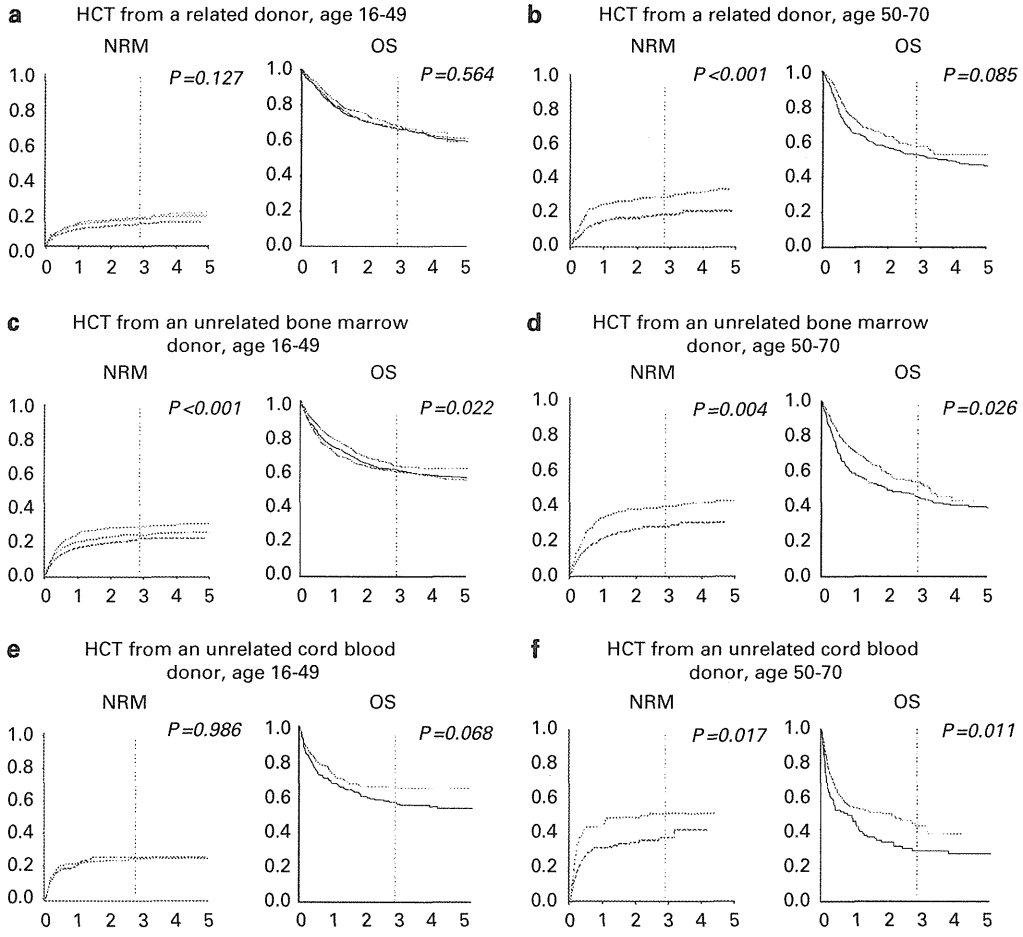

Figure 3. (**a**) NRM and OS at 3 years from HCT among younger patients (16–49 years) who received allo-HCT from a related donor were 15%, 16% and 12% ($P=0.127$), and 67%, 66% and 68% ($P=0.564$), respectively in the period of 1997–2000 ($n=587$, solid line), 2001–2004 ($n=620$, dotted line) and 2005-2008 ($n=639$, dashed line). (**b**) NRM and OS among older patients (50–70 years) who received related donor transplantation were 28% and 17% ($P<0.001$) and 52% and 57% ($P=0.085$), respectively in the period of 2001–2004 ($n=293$, dotted line) and 2005–2008 ($n=321$, dashed line). (**c**) NRM and OS among younger patients who received allo-HCT from an unrelated BM donor were 28%, 24% and 22% ($P<0.001$), and 60%, 60% and 63% ($P=0.022$), respectively in the period of 1997–2000 ($n=560$, solid line), 2001–2004 ($n=803$, dotted line) and 2005-2008 ($n=839$, dashed line). (**d**) NRM and OS among older patients who received allo-HCT from an unrelated BM donor were 39% and 27% ($P=0.004$) and 45% and 54% ($P=0.026$), respectively in the period of 2001–2004 ($n=195$, dotted line) and 2005–2008 ($n=473$, dashed line). (**e**) Non-relapse mortality and OS among younger patients who received allogeneic hematopoietic cell transplantation from an unrelated cord blood donor were 25% and 25% ($P=0.986$), and 55% and 65% ($P=0.068$), respectively in the period 2001–2004 ($n=214$, dotted line) and 2005–2008 ($n=292$, dashed line). (**f**) Non-relapse mortality and OS among older patients who received allogeneic hematopoietic cell transplantation from an unrelated cord blood donor were 51% and 37% ($P=0.017$), and 29% and 44% ($P=0.011$), respectively in the period of 2001–2004 ($n=107$, dotted line) and 2005–2008 ($n=242$, dashed line).

14 and 8%, $P=0.049$, Figure 4c). We found a significant reduction in mortality rates associated with bacterial and fungal infection.

Allo-HCT from an unrelated CB donor

In younger patients who received allo-HCT from an unrelated CB donor, there was no significant difference in the incidence of NRM between the two periods (Figure 3e). In this group, there was a marked reduction in the relapse rate (25 and 18%, $P=0.018$, data not shown; HR 0.66, 95% CI 0.43–1.00, $P=0.049$, Table 2). OS was better in 2005–2008; however, the difference was not statistically significant.

Significant improvements in NRM and OS were observed in 2005–2008 among older patients who received UCBT (Figure 3f). The HRs for NRM and overall mortality in 2005–2008 were 0.57 (95% CI 0.40–0.83, $P=0.003$) and 0.67 (95% CI 0.49–0.91, $P=0.010$), respectively. Reductions in the incidences of death associated with GVHD and infection seemed to contribute to the improvements in NRM (GVHD, 7 and 3%, $P=0.163$; infection, 23 and 13%, $P=0.136$). The mortality rate due to bacterial infection was significantly reduced.

Incidence of and mortality after severe acute GVHD

In subgroups that showed a significant reduction in the incidence of NRM, younger patients who received UBMT, older patients who received related HCT and older patients who received UCBT showed significant reductions in the incidence of GVHD-related mortality. In younger patients who received UBMT, the incidence of severe acute GVHD was significantly reduced over the three

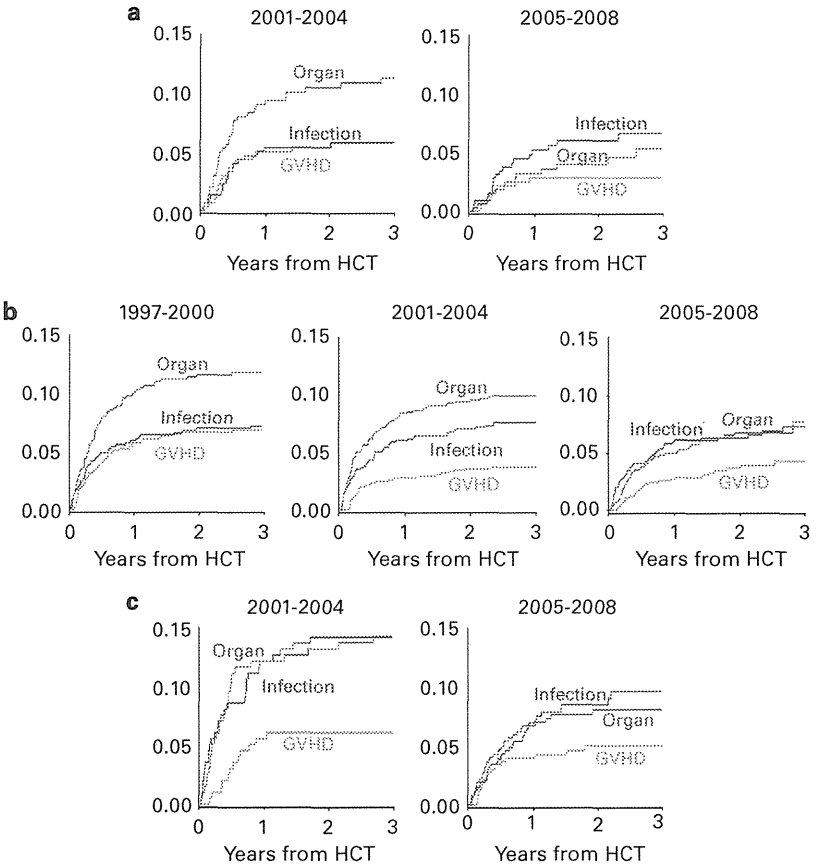

Figure 4. Change in the causes of NRM among different time periods is shown. Cumulative incidences of death due to GVHD, infection and organ failure are separately presented in each time period. (**a**) In older patients who received allo-HCT from a related donor, the incidences of death associated with organ failure and GVHD were significantly reduced in 2005–2008 (organ failure, 11 and 6%, $P=0.007$; GVHD, 6 and 3%, $P=0.015$). (**b**) In younger patients who received allo-HCT from an unrelated BM donor, the incidences of death associated with GVHD and organ failure were significantly reduced (GVHD, 7, 4 and 4%, $P=0.011$; organ failure, 12, 10 and 8%, $P=0.002$). (**c**) In older patients who received allo-HCT from an unrelated BM donor, the incidences of death associated with infection and organ failure were reduced in 2005–2008 (infection, 14 and 10%, $P=0.054$; organ failure, 14 and 8%, $P=0.049$).

periods (16, 15 and 12% at 100 days after allo-HCT, $P=0.021$). In older patients who received related HCT, the incidence of severe acute GVHD was reduced in 2005–2008 relative to 2001–2004, but this difference was not statistically significant (14 and 10%, $P=0.099$). In older patients who received UCBT, there was no remarkable reduction in the incidence of severe acute GVHD in the later period (18 and 16%, $P=0.542$). However, the mortality rate was significantly reduced among older patients who suffered severe acute GVHD after UCBT (92 and 67% at 3 years after allo-HCT, $P=0.022$).

DISCUSSION

In this study that used a large database of 6501 patients, we found that the incidence of NRM after allo-HCT for adult patients has significantly decreased over the past 12 years, which has led to an improvement of OS. As prior studies have primarily focused on the changes in NRM among younger patients who received allo-HCT with myeloablative conditioning,[2,4] this is the first study to show the changes in NRM in subgroups comprising older patients and UCBT.

We found that demographic, disease and transplantation characteristics have been changing, as previous studies reported.[1,2,4] The marked increase in the number of older patients, allo-HCT with reduced-intensity conditioning and UCBT might reflect an increase in allo-HCT for 'more vulnerable' patients. Gooley *et al.*[1] reported that the hematopoietic cell transplantation-specific comorbidity index (HCT-CI)[7] scores were higher in HCT recipients in more recent time periods. Unfortunately, we were not able to evaluate HCT-CI in the current study because of a lack of information.

Among patients who received related HCT, remarkable improvement in NRM was observed in older patients. Another distinguishing finding was an increase in relapse in overall older patients, especially among those who received related HCT in remission. There was no recent shift in the use of allo-HCT in a later remission state, and we obtained a similar result when the analyses were restricted to HCT using reduced-intensity regimens or myeloablative regimens. In addition, the proportional use of anti-thymocyte globulin has remained unchanged over the periods. Less use of PB donors and more aggressive selection of older patients as indicated for allo-HCT may have affected the result. Despite this increase in relapse, older patients who received HCT in remission showed, by multivariate analyses, a significant reduction in mortality with a remarkable reduction in HRs for NRM irrespective of donor sources.

In analyses based on the donor source, UBMT showed remarkable improvements in NRM and OS throughout the age subgroups. Along with high-resolution donor–recipient HLA

matching,[8,9] the lesser proportion of donor/patient pairs with allele mismatches may have reduced the incidence of GVHD-related mortality, and contributed to the improvement in outcomes after UBMT.

Among patients who received UCBT, we found a decreased risk of relapse in younger patients with no change in NRM. On the other hand, older patients had a decreased risk of NRM with no change in relapse. These outcomes may be explained by the changes in clinical practice in 2001–2004, 'learning phase' of UCBT, and that after 2005, including the indication of UCBT and the prophylaxis and treatment for GVHD/infection.

A recent reduction in the incidence of GVHD-related mortality was observed in younger patients receiving UBMT and older patients receiving related allo-HCT or UCBT. With the changes in prophylaxis and treatment against GVHD including high-resolution donor–recipient HLA matching,[8,9] the incidence of grade 3 to 4 severe acute GVHD has decreased in younger patients receiving UBMT and older patients receiving related HCT, which may have led to the reduction in GVHD-related mortality in these subgroups. Interestingly, in older patients receiving UCBT, there was no reduction in the incidence of severe acute GVHD; however, the mortality rate among those who developed severe acute GVHD was reduced. The prompt initiation of treatment after a more thorough examination to diagnose GVHD,[10] supportive care and nutritional management may have improved the prognosis of those who had severe GVHD. Alternatively, the unique HLA epidemiological genetics of Japanese patients may have affected the results.[11,12]

A recent reduction in the incidence of infection-related mortality was observed in older patients receiving UBMT or UCBT. New antifungal drugs, including mold-active azoles, micafungin or liposomal amphotericin B, are now more likely to be administered as empiric or preemptive strategies for patients who have a positive galactomannan Ag test or pulmonary nodules.[1,13,14] As GVHD and infection have been reported to be associated with each other's development and exacerbation,[13,15–18] an improved control of severe GVHD may have led to the reduction of the risk of infection-related mortality.[13,14]

We included all of the organ toxicities that were documented after allo-HCT as the cause of organ failure-related mortality, including conditioning regimen-related toxicity,[19,20] lung injury[15] and late effects on any organs.[21] We observed a reduction in the incidence of organ failure-related mortality in older patients receiving related HCT and those who received UBMT. In the future, more detailed analyses are warranted based on each specific organ toxicity.

As this analysis is based on a retrospectively collected multicenter database, our results may be susceptible to the disadvantages of any retrospective study, such as the heterogeneity in the treatment strategies chosen at the discretion of the physicians. Because of the nature of the multicenter registry, detailed data were not available regarding the incidences of infection and specific organ failure, and prophylactic treatment toward infection. Although we acknowledge this limitation, the results obtained from this large database that contains clinical data on over 6000 patients should provide valuable information. In addition, for the first time, we found reductions in NRM in subgroups consisting of older patients and those who received UCBT. We also showed the causes of death that contributed to the reduction of NRM in each donor/age subgroup. By further evaluating the risks of NRM and relapse in each demographic subgroup, we would be able to more clearly define the indications for allo-HCT, and tailor the strategy for individual patients.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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Author contributions: SK designed the study, prepared the data file, performed the analysis, interpreted the data and wrote the manuscript; KY contributed to the study design, data file preparation, data analysis and interpretation of the data; TY was primarily responsible for the study design, data analysis and interpretation of the data; YA reviewed and cleaned the data, interpreted the data and helped to write the manuscript; TNI reviewed, cleaned and interpreted the data, HA, ST, KM, ST, TE, HO and MK obtained and interpreted the data; JT, KK, KK, RS, YM and HS reviewed, cleaned and interpreted the data; TF designed the study, interpreted the data and helped to write the manuscript.

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Comparison of Unrelated Cord Blood Transplantation and HLA-Mismatched Unrelated Bone Marrow Transplantation for Adults with Leukemia

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Recent advances in unrelated cord blood transplantation (UCBT) and high-resolution typing of human leukocyte antigen (HLA) from an unrelated donor have increased choices in alternative donor/stem cell source selection. We assessed HLA-mismatched locus-specific comparison of the outcomes of 351 single-unit UCB and 1,028 unrelated bone marrow (UBM) adult recipients 16 years old or older at the time of transplantation who received first stem cell transplantation with myeloablative conditioning for acute leukemia or myelodysplastic syndromes. With adjusted analyses, HLA 0 to 2 mismatched UCBT showed similar overall mortality (relative risk [RR] = 0.85, 95% confidence interval [CI], 0.68-1.06; $P = .149$) compared with that of single-HLA-DRB1-mismatched UBMT. UCBT showed inferior neutrophil recovery (RR = 0.50, 95% CI, 0.42-0.60; $P < .001$), lower risk of acute graft-versus-host disease (RR = 0.55, 95% CI, 0.42-0.72; $P < .001$), and lower risk of transplantation-related mortality (RR = 0.68, 95% CI, 0.50-0.92; $P = .011$) compared with single-HLA-DRB1-mismatched UBMT. No significant difference was observed for risk of relapse (RR = 1.28, 95% CI, 0.93-1.76; $P = .125$). HLA 0 to 2 antigen-mismatched UCBT is a reasonable second alternative donor/stem cell source with a survival outcome similar to that of single-HLA-DRB1-mismatched or other 7 of 8 UBMT.

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KEY WORDS: Unrelated cord blood transplantation, HLA-mismatched unrelated bone marrow transplantation

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INTRODUCTION

Allogeneic hematopoietic stem cell transplantation (HSCT) is a widely used, curative treatment for hematologic malignancies. When available, a human leukocyte antigen (HLA)-identical sibling is the donor of choice. However, only about 30% of candidates eligible for allogeneic HSCT will have such a donor. In addition, older patients with older siblings have more difficulty finding such a donor capable of stem cell donation. High-resolution donor-recipient HLA matching has contributed to the success of unrelated donor marrow transplantation, and the current first recommended alternative donor after an HLA-matched sibling for HSCT is an HLA-A, -B, -C, and -DRB1 8 of 8-allele-matched unrelated donor [1-4]. However, there are still a significant number of patients for which finding an HLA 8 of 8-matched unrelated donor is difficult and for whom a second alternative donor/stem cell source should be found.

The effect of HLA mismatches after bone marrow transplantation from unrelated donors (UBMT) has been well studied, and single mismatched UBM donors are usually selected as a second alternative donor/stem cell source [1-4]. Lee al. [3] showed that a single mismatch, antigen-level, or high-resolution, at HLA-A, -B, -C, or -DRB1 loci was associated with higher mortality and decreased survival. However, the reduction in survival may be acceptable in comparison with the survival rates for currently available alternative treatments. Analyses from the Japan Marrow Donor Program (JMDP) showed better survival in HLA class II mismatched recipients; thus, single-DRB1-mismatched UBM donor is currently a second alternative in Japan [1,2,5].

Recent advances in unrelated cord blood transplantation (UCBT) have provided patients with increased choices for a second alternative donor/stem cell source [6]. Clinical comparison studies of cord blood transplantation and HLA-A, -B, and -DRB1 6 of 6 allele-matched bone marrow transplantation for leukemia from unrelated donors in adult recipients showed comparable results [7-9]. More recently, promising outcomes of UCBT were shown compared with HLA-A, -B, -C, and -DRB1 8 of 8 allele-matched UBMT, the current first alternative donor/stem cell source [10-12].

The aim of this study was to determine the utility of UCBT as a second-alternative donor source in adult patients with acute leukemia or myelodysplastic syndromes. It is common today to perform high-resolution typing of HLA for donor selection of unrelated donors; thus, we performed mismatched-allele-specific analyses for comparison of HLA-mismatched UBMT and UCBT in terms of overall survival (OS) and other HSCT outcomes, setting single-DRB1-mismatched UBMT, the current second alternative, as the reference.

PATIENTS AND METHODS

Collection of Data and Data Source

The recipients' clinical data were provided by the Japan Cord Blood Bank Network (JCBBN) and the JMDP [13]. Peripheral blood stem cell donation from unrelated donors was not permitted in Japan during the study period. All 11 cord blood banks in Japan are affiliated with JCBBN. Both JCBBN and JMDP collect recipients' clinical information at 100 days posttransplantation. Patients' information on survival, disease status, and long-term complications including chronic graft-versus-host (cGVHD) disease and second malignancies is renewed annually using follow-up forms. This study was approved by the institutional review board of Nagoya University Graduate School of Medicine.

Patients

The subjects were adult patients of at least 16 years of age with acute myeloid leukemia, acute lymphoblastic leukemia, and myelodysplastic syndromes, who were recipients of first UBMT or UCBT with myeloablative conditioning. All patients in the UCBT cohort received a single-unit CB. Transplantation years were between 1996 and 2005 for UBMT and between 2000 and 2005 for UCBT to avoid the first 3 years of a pioneering period (1993-1995 for UBMT and 1997-1999 for UCBT). There were no statistically significant differences between UBMT in 1996-1999 and UBMT in 2000-2005 in probabilities of OS (41% versus 44%, at 3 years posttransplantation; $P = .86$) and in relapse-free survival (RFS) (40% versus 40%, at 3 years posttransplantation; $P = .93$).

Among 2,253 UBMT recipients with complete HLA high-resolution data, the following recipients with HLA -A, -B, -C, and -DRB1 8 of 8 allele match (n = 1,079) and more than three mismatches (5 of 8 allele match [n = 117], 4 of 8 allele match [n = 24], 3 of 8 allele match [n = 4], 2 of 8 allele match [n = 1]) were excluded. There were no statistically significant differences in risk of mortality or treatment failure (RFS) associated with single high-resolution (allele) versus single low-resolution (antigen) mismatches (data not shown), so in the analyses, allele and antigen mismatches were considered equivalent. HLA matching of cord blood was performed using low-resolution molecular typing methods for HLA-A and -B, and high-resolution molecular typing for HLA-DRB1. Of 557 recipients of CB with complete HLA data, 105 recipients with three mismatches and nine recipients with four mismatches were excluded. A total of 1,028 UBMT recipients (248 HLA class II locus mismatched, 424 HLA class I locus mismatched, and 356 HLA 2 loci mismatched) and 351 UCBT recipients (20 HLA-A, -B, low-resolution and -DRB1 matched, 87

locus mismatched, and 244 2 loci mismatched) were the subjects for analyses. Both host-versus-graft and graft-versus-host directions were accounted for in terms of HLA mismatch.

HLA Typing

Alleles at the HLA-A, -B, -C, and -DRB1 with unrelated bone marrow donor-recipient pairs and for HLA-DRB1 for unrelated cord blood donor-recipient pairs were identified by the methods described previously [1,5,14]. Serologic or antigen-level typing was performed with a standard two-stage complement-dependent test of microcytotoxicity or low-resolution DNA-based typing usually by collapsing the four-digit typing result back to its first two digits in part.

Definitions

The primary outcome of the analyses was OS, defined as time from transplantation to death from any cause. A number of secondary endpoints were also analyzed. Neutrophil recovery was defined by an absolute neutrophil count of at least 500 cells per cubic millimeter for three consecutive points; platelet recovery was defined by a count of at least 50,000 platelets per cubic millimeter without transfusion support. Diagnosis and clinical grading of acute GVHD (aGVHD) were performed according to the established criteria [15,16]. Relapse was defined as a recurrence of underlying hematologic malignant diseases. Transplantation-related death was defined as death during a continuous remission. RFS was defined as survival in a state of continuous remission.

Statistical Analysis

Descriptive statistical analysis was performed to assess patient baseline characteristics, diagnosis, disease status at conditioning, donor-patient ABO mismatches, preparative regimen, and GVHD prophylaxis. Medians and ranges are provided for continuous variables and percentages for categoric variables. Cumulative incidence curves were used in a competing-risks setting to calculate the probability of aGVHD and cGHVD, relapse, and transplantation-related mortality (TRM) [17]. Gray's test was used for group comparison of cumulative incidences [18]. Adjusted comparison of the groups on OS and RFS was performed with the use of the Cox proportional-hazards regression model [19]. For other outcomes with competing risks, Fine and Gray's proportional-hazards model for subdistribution of a competing risk was used [20]. For neutrophil and platelet recovery, death before neutrophil or platelet recovery was the competing event; for GVHD, death without GVHD and relapse were the competing events; for relapse, death without relapse was the competing event; and, for TRM, relapse was the competing event [21]. Adjusted probabilities of OS and RFS were estimated using the Cox proportional-hazards regression model, with consideration of other significant clinical variables in the final multivariate models. The variables considered were the patient's age at transplantation, patient's sex, donor-patient sex mismatch, donor-patient ABO mismatch, diagnosis, disease status at conditioning, the conditioning regimen, and the type of prophylaxis against GVHD. Factors differing in distribution between CB and BM recipients and factors known to influence outcomes were included in the final models. Variables with more than two categories were dichotomized for the final multivariate model. Variables were dichotomized as follows: patient age >40 or <40 years at transplantation, recipient's sex, sex-mismatched donor-patient pair versus sex-matched pair, donor-recipient ABO major mismatch versus others for ABO matching, advanced versus standard (first and second complete remission of acute myeloid leukemia, first complete remission of acute lymphoblastic leukemia, or refractory anemia or refractory anemia with ring sidoblasts of myelodysplastic syndromes) risk of the disease, cyclophosphamide, and total-body irradiation (TBI) or busulfan and cyclophosphamide or others for conditioning regimen, and cyclosporine-based versus tacrolimus-based prophylaxis against GVHD. No significant interactions were identified between each variable and HLA disparity/stem cell source groups. All P values were two-sided.

RESULTS

Patient Characteristics

Table 1 shows characteristics of patients, their disease, and transplantation regimens. Proportions of females, sex-mismatched donor-recipient pairs, and ABO mismatched donor recipient pairs were larger in cord blood recipients ($P < .001$, $P < .001$, and $P < .001$, respectively). UCB recipients were older than recipients of UBM (median age, 37 years versus 34 years; $P < .001$). A preparative regimen with TBI and cyclophosphamide was used in the majority of patients in all groups, and cytosine arabinoside was supplemented for CB recipients in addition to TBI and cyclophosphamide in about half the recipients with cyclophosphamide and TBI. For GVHD prophylaxis, tacrolimus and short-term methotrexate was used preferentially in BM recipients (61% of DRB1-one-mismatched BM recipients), while cyclosporine A and short-term methotrexate was used preferentially in CB recipients (61%). The median follow-up period for survivors was 2.1 years (range, 0.1-6.2) for CB recipients and 5.5 (range, 0.3-11.6) years for BM recipients.

Table 1. Patient, Disease, and Transplantation Characteristics According to Stem Cell Source and Number of Mismatched Loci

	Bone Marrow Transplant			
	Class II One Locus Mismatch	Class I One Locus Mismatch	Two Loci Mismatch	Cord Blood Transplantation
	N (%)	N (%)	N (%)	N (%)
Number of transplantations	248	424	356	351
Patient age at transplantation				
Median (range)	36 (16-60)	34 (16-67)	34 (16-59)	37 (16-58)
Patient sex				
Male	151 (61)	241 (57)	210 (59)	162 (46)
Female	97 (39)	183 (43)	146 (41)	189 (54)
Sex matching				
Matched	145 (58)	268 (63)	217 (61)	170 (48)
Male to female	52 (21)	82 (19)	73 (21)	97 (28)
Female to male	50 (20)	71 (17)	64 (18)	84 (24)
Unknown	1 (<1)	3 (1)	2 (1)	0 (0)
Diagnosis				
AML	135 (54)	204 (48)	172 (48)	193 (55)
ALL	78 (31)	149 (35)	135 (38)	113 (32)
MDS	35 (14)	71 (17)	49 (14)	45 (13)
Disease status				
Standard	124 (50)	214 (50)	168 (47)	147 (42)
Advanced	114 (46)	195 (46)	169 (47)	174 (50)
Unknown	10 (4)	15 (4)	19 (5)	30 (9)
ABO matching				
Matched	119 (48)	184 (43)	153 (43)	114 (32)
Minor mismatch	53 (21)	108 (25)	85 (24)	99 (28)
Major mismatch	67 (27)	116 (27)	97 (27)	73 (21)
Bidirectional	8 (3)	12 (3)	14 (4)	64 (18)
Unknown	1 (<1)	4 (1)	7 (2)	1 (<1)
HLA-mismatched number and direction				
Matched				20 (6)
One locus mismatched				87 (25)
HVG direction	16 (6)	38 (9)		8 (9)
GVH direction	17 (7)	30 (7)		8 (9)
Both directions	215 (87)	356 (84)		71 (82)
Two loci mismatched				244 (70)
Two HVG direction			4 (1)	2 (1)
One HVG direction and one GVH direction			6 (2)	4 (2)
Two GVH direction			4 (1)	3 (1)
One both directions and one HVG direction			42 (12)	40 (16)
One both directions and one GVH direction			29 (8)	28 (11)
Two both directions			271 (76)	167 (68)
No. of nucleated cells infused ($\times 10^7$/kg)				
Median	25.0	24.5	23	2.46
Range	2.40-59.8	2.10-97.5	1.5-66.0	1.41-6.01
Preparative regimen				
CY + TBI	94 (38)	168 (40)	151 (42)	109 (31)
CY + CA + TBI	46 (19)	78 (18)	74 (21)	124 (35)
CY + BU + TBI	20 (8)	39 (9)	27 (8)	15 (4)
Other TBI regimen	45 (18)	70 (17)	61 (17)	80 (23)
BU + CY	34 (14)	54 (13)	30 (8)	21 (6)
Other non-TBI regimen	9 (4)	15 (4)	13 (4)	2 (1)
GVHD prophylaxisis				
Cycrosporine A + sMTX	87 (35)	221 (52)	150 (42)	213 (61)
Cyclosporine A ± other	1 (<1)	5 (1)	5 (1)	24 (7)
Tacrolimus + sMTX	152 (61)	191 (45)	193 (54)	76 (22)
Tacrolimus ± other	8 (3)	5 (1)	6 (2)	35 (10)
Others	0 (0)	2 (<1)	2 (<1)	3 (1)

ALL indicates acute lymphoblastic leukemia; AML, acute myelogenous leukemia; BU, oral busulfan; CA, citarabine; CY, cyclophosphamide; GVH, graft-versus-host; HVG, host-versus-graft; MDS, myelodysplastic syndomes; sMTX, short-term methotrexate.

Outcome

OS and RFS

OS and RFS for CB recipients were similar when compared with that of single-HLA-DRB1-mismatched BM recipients (relative risk [RR] = 0.85, 95% confidence interval [CI], 0.68-1.06; P = .149 for OS and RR = 0.97, 95% CI, 0.92-1.35; P = .747) (Table 2).

The adjusted probabilities of survival at 3 years posttransplantation of CB recipients (47%) were not different from those of single HLA-DRB1 mismatched BM recipients (41%; P = .19) or single HLA class I-mismatched BM recipients (47%; P = .96), but superior to those of 6 of 8 BM recipients (38%; P = .014) (Figure 1A). Figure 1B shows adjusted RFS curves (42% for CB recipients, 36% for single HLA-DRB1-mismatched BM, 44% for single HLA class I-mismatched BM, and 36% for 6 of 8 BM recipients, at 3 years posttransplant) (P values of comparison between CB and single HLA-DRB1-mismatched BM, CB, and single HLA

Table 2. Multivariate Analyses of Overall Survival, Relapse-Free Survival, Relapse, and Transplant-Related Mortality

	Degree of HLA Mismatch	N	Overall Survival			Relapse-Free Survival			Relapse			Transplant-Related Mortality		
			RR	(95% CI)	P value	RR	(95% CI)	P value	RR	(95% CI)	P value	RR	(95% CI)	P value
Bone marrow transplant	Single DRB1 (7/8)	248	1.00			1.00			1.00			1.00		
	Single A or B (7/8)	137	0.84	(0.64-1.11)	.216	0.82	(0.63-1.08)	.158	0.65	(0.41-1.01)	.056	1.07	(0.77-1.49)	.698
	Single C (7/8)	287	0.89	(0.72-1.12)	.324	0.86	(0.69-1.07)	.170	0.60	(0.41-0.87)	.007	1.13	(0.86-1.48)	.391
	C + DRB1 (6/8)	144	0.97	(0.74-1.27)	.831	0.95	(0.73-1.24)	.726	0.76	(0.49-1.17)	.208	1.10	(0.78-1.55)	.600
	A/B + C (6/8)	122	1.22	(0.94-1.59)	.143	1.15	(0.88-1.49)	.300	0.70	(0.44-1.10)	.12	1.42	(1.03-1.96)	.032
	Other two loci (6/8)	90	1.25	(0.92-1.68)	.146	1.13	(0.84-1.53)	.409	0.60	(0.35-1.02)	.061	1.48	(1.03-2.13)	.035
Cord blood transplant		351	0.85	(0.68-1.06)	.149	0.97	(0.92-1.35)	.747	1.28	(0.93-1.76)	.125	0.68	(0.50-0.92)	.011

RR indicates relative risk; CI, confidence interval.
Adjusted by patient age at transplantation >40 versus ≤40, patient sex, donor-patient sex mismatch versus matched, ABO major mismatch versus others, advanced versus standard disease status at transplantation, cyclophosphamide and total-body irradiation or busulfan and cyclophosphamide for conditioning versus other conditioning regimen, and cyclosporine-based versus tacrolimus-based prophylaxis against graft-versus-host disease.

class I-mismatched BM, and CB and 6 of 8 BM recipients were 0.80, 0.12, and 0.43, respectively).

Relapse and TRM

There was no significant increase of relapse rates among CB recipients when compared with DRB1 single-mismatched BM recipients (RR = 1.28, 95% CI, 0.93-1.76; $P = .125$). The risk of TRM was lower in CB recipients compared with that of single HLA-DRB1-mismatched BM recipients (RR = 0.68, 95% CI, 0.50-0.92; $P = .011$) (Table 2). The risk of TRM was also lower in CB recipients when compared with 6 of 8 BM recipients (RR = 0.52, 95% CI, 0.39-0.68; $P < .001$).

Hematologic recovery

Neutrophil and platelet recovery was inferior in CB recipients, as shown in Table 3 (RR = 0.50, 95% CI, 0.42-0.60; $P < .001$ for neutrophil recovery, RR = 0.52, 95% CI, 0.42-0.63; $P < .001$ for platelet recovery).

Acute GVHD and chronic GVHD

The risk of grade 2 to 4 or severe (grades 3-4) aGVHD was lower in CB recipients than that of single HLA-DRB1-mismatched BM recipients (RR = 0.55, 95% CI, 0.42-0.72; $P < .001$ for grade 2 to 4 aGVHD and RR = 0.43, 95% CI, 0.27-0.58; $P < .001$ for severe aGVHD) (Table 4). Unadjusted cumulative incidence of severe aGVHD was 9% for CB, 19% for single HLA-DRB1-mismatched BM, 18% for single HLA

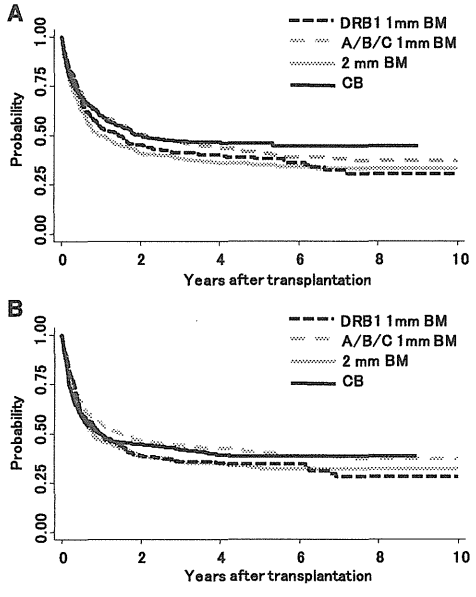

Figure 1. Adjusted probabilities of OS (A) and RFS (B). The adjusted 3-year probabilities of OS for unrelated cord blood recipients, single-HLA-DRB1-mismatched unrelated bone marrow (UBM) recipients, single-HLA-class-I-mismatched UBM, and 6 of 8 UBM recipients were 47%, 41%, 47%, and 38%, respectively (A). The adjusted 3-year probabilities of RFS were 42%, 36%, 44%, and 36%, respectively (B).

Table 3. Multivariate Analyses of Neutrophil and Platelet Recovery

			Neutrophil Recovery			Platelet Recovery		
	Degree of HLA Mismatch	N	RR	(95% CI)	*P* value	RR	(95% CI)	*P* value
Bone marrow transplantation	Single DRB1 (7/8)	248	1.00			1.00		
	Single A or B (7/8)	137	1.31	(1.04-1.65)	.021	1.31	(1.01-1.70)	.039
	Single C (7/8)	287	1.19	(0.98-1.43)	.069	0.98	(0.79-1.21)	.840
	C + DRB1 (6/8)	144	0.96	(0.77-1.20)	.735	0.79	(0.62-1.02)	.065
	A/B + C (6/8)	122	1.14	(0.89-1.45)	.307	0.84	(0.63-1.13)	.255
	Other two loci (6/8)	90	0.89	(0.68-1.14)	.346	0.80	(0.58-1.10)	.174
Cord blood transplantation		351	0.50	(0.42-0.60)	<.001	0.52	(0.42-0.63)	<.001

RR indicates relative risk; CI, confidence interval.

Adjusted by patient age at transplantation >40 versus <40, patient sex, donor-patient sex mismatch versus matched, ABO major mismatch versus others, advanced versus standard disease status at transplant, cyclophosphamide, and total-body irradiation or busulfan and cyclophosphamide for conditioning versus other conditioning regimen, and cyclosporine-based versus tacrolimus-based prophylaxis against graft-versus-host disease.

class I-mismatched BM, and 22% for 6 of 8 BM at 100 days posttransplantation ($P < .001$ between CB and single HLA-DRB1-mismatched BM) (Figure 2A).

Among recipients who survived at least 100 days posttransplantaton, the risk of developing cGVHD and extensive-type cGVHD was not significantly increased in all HLA disparity groups of CB recipients when compared with that of HLA-DRB1-allele/antigen-mismatched BM recipients (RR = 1.36, 95% CI, 0.99-1.88; $P = .057$ for cGVHD, and RR = 0.86, 95% CI, 0.55-1.34; $P = .500$ for extensive-type cGVHD). The unadjusted cumulative incidence of extensive-type cGVHD was 17% for CB recipients, 20% for single HLA-DRB1-mismatched BM, 25% for single HLA class I-mismatched BM, and 30% for 6 of 8 BM recipients at year posttransplantation ($P = .34$ between CB and single HLA-DRB1-mismatched BM) (Figure 2B).

DISCUSSION

Our main objective was to compare OS after transplantation of UCBT and single-HLA-mismatched UBMT and to provide useful data for selection of an appropriate donor and graft source in second stem cell source/donor selection for adults with hematologic malignancy. To the best of our knowledge, this is the first study to involve mismatched allele/antigen-specific analyses including CB for the process of donor selection. Our results suggest that 0 to 2 HLA-mismatched UCB is a reasonable second alternative of choice for adult patients with leukemia, with similar survival to that of single DRB1-mismatched or other 7 of 8 UBM recipients, the current first choice for second alternative donor/stem cells.

Neutrophil and platelet recovery was slower in CB recipients than BM recipients, consistent with the results of previous reports [7-10,12]. This is the major limitation of the use of UCB, and several strategies have been studied to reduce the neutropenic period, such as screening for patients' pretransplantation anti-HLA antibodies and their specificity, transplantation of 2 UCB units if a single UCB unit with an adequate cell dose is not available, or direct infusion of UCB into bone marrow [22-26].

Despite higher HLA disparity at the antigen level (69% 2 antigen mismatch, 25% antigen mismatch, and 6% matched), UCB recipients showed lower incidence of severe aGVHD than single DRB1-mismatched UBM recipients, consistent with other reports that compared UCB with single-mismatched UBM (7 of 8) [8,11,12]. In our study, tacrolimus and short-term methotrexate were used preferentially in BM recipients, whereas cyclosporine A was used in 68% of CB recipients. Prior studies have shown reduced severe aGVHD with tacrolimus, and this difference may have underscored the improved aGVHD control of UCB over mismatched BM in unadjusted analyses [27,28]. It is likely that decreased risk of grade 2 to 4 aGVHD in UCB recipients contributed to decreased risk of TRM among UCB recipients.

Increasing the number of HLA mismatches from 7 of 8 to 6 of 8 was associated with an approximately 10% reduction in survival in UBM recipients, which was quite similar to the results from the National Marrow Donor Program [3]. Because we eliminated data from the first 3 pioneering years of unrelated BMT, most of the bone marrow recipients and donors were allele-typed for at least HLA-A, -B, and -DRB1 before transplantation. Survival outcomes of single class I mismatch were not significantly different from those of single class II mismatch in the current analyses. We believe that allele typing of HLA-A, -B, and -DRB1 before transplantation led to better selection of the donor compared with that in the first several years of UBMT. This study includes a large number of fully typed BM and CB recipients, but there are limitations. The choice of stem cell source is influenced by many unmeasured factors that can affect outcome. It is also influenced by the availability of acceptable HLA disparity for unrelated donors and mainly cell dose for cord blood units. Although we have adjusted for known risk factors and disparities between groups, we cannot rule out the influence of potential selection bias, which can only be excluded in a randomized controlled trial. Transplantation years