Prospective Evaluation of Alternative Donor from Unrelated Donor and Cord Blood in Adult Acute Leukemia and Myelodysplastic Syndrome

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ABSTRACT

A prospectively registered observational study was conducted to assess the significance of allogeneic hematopoietic stem cell transplantation from highly HLA-matched unrelated donors (UD) and cord blood (CB) on outcomes in adult acute leukemia (AL) and myelodysplastic syndrome (MDS). Between 2007 and 2015, 231 transplant-eligible patients were registered for a phase 2 study of alternative donor transplantation. After registration, a sufficient time period was given to find appropriate UD. Patients received CB transplantation (CBT) if an appropriate UD was unavailable. In total, 119 patients received CBT (106 AL and 13 MDS) and 91 patients received UD transplantation (UDT) (86 AL and 5 MDS). The median age was 39 years in both groups. The primary objective was overall survival (OS); secondary objectives included cumulative incidences of non-relapse mortality (NRM) and relapse, and disease-free survival. Diagnosis, disease status at transplantation, refined-disease risk index, and hematopoietic cell transplant-specific comorbidity index did not differ between UDT and CBT. In multivariate analyses, graft source was not a significant risk factor for all objectives. In adjusted analyses, UDT and CBT showed similar OS, NRM, and relapse in this prospective study. CB can be a comparable alternative stem cell source to UD by achieving a timely transplant.

INTRODUCTION

Hematopoietic stem cell transplantation (SCT) from alternative donors is becoming a choice of standard of care. Since HLA-mismatched umbilical cord blood transplantation (CBT) was introduced as a treatment option for adult hematological disease,[1, 2] transplant outcomes with CBT have dramatically improved.[3] Among HLA well-matched unrelated donor transplantation (UDT) and CBT, HLA-8/8 allele-matched UDT showed superior overall survival (OS) compared with HLA-7/8 allele-matched UDT and CBT, whereas similar OS was observed between HLA-7/8 allele-matched UDT and CBT.[1, 4-7] However, because UDT and CBT are considerably different therapeutic options comprised of distinct graft characteristics, it is always difficult to compare UDT with CBT without any bias. Although previous outcome research showed that HLA well-matched UDT and CBT yielded almost comparable survival rates even for the high-risk leukemia patients,[8] it has been pointed out that various biases are involved in comparing UDT with CBT. One of the potential biases is the difference in the waiting period duration for transplantation.[9, 10] Patients who do not obtain remission or relapse during the donor search period often select immediate transplantation with a readily available graft source. Inevitably, CB recipients have included a higher frequency of non-remission patients in retrospective outcome studies.[1, 7] Nevertheless, a randomized control study comparing UDT with CBT cannot be practical because of ethical considerations. Thus, such a direct comparison has not been performed yet.

To investigate the significance of highly HLA-matched UDT and CBT with as low bias as possible, we designed a prospectively registered observational study. In the present study, adult acute leukemia and myelodysplastic syndrome (MDS) patients with high-risk disease features were prospectively registered, and then UD search was conducted for a certain period. Because of high-risk disease features of patients, we aimed to perform relatively early SCT. After the UD search period, patients received UDT if an appropriate UD was available, whereas most of the other

patients received CBT. We conducted a phase 2 clinical study to confirm the efficacy of CBT within a part of the present study, which is reported elsewhere.[11] Herein, we analyzed the entire cohort and compared CBT with UDT. The primary objective of the present study was to assess survival after CBT in comparison with that after UDT.

PATIENTS AND METHODS

Study Design and Patient Population

Patients aged 16–55 years with acute lymphoblastic leukemia (ALL), acute myeloid leukemia (AML), or myelodysplastic syndrome (MDS) with high-risk features were eligible. After we confirmed the lack of an HLA-matched related donor, provisional registration to the study was conducted. We initially attempted to identify an HLA-8/8 or HLA-7/8 allele-matched UD. At provisional registration, patients with scarce UD candidates were recruited, and the donor search was conducted until no UD candidate was available or an appropriate UD was identified within a sufficient UD search period. To avoid a prolonged UD search, the UD search duration was limited to 180 days. The 180-day criterion was at the latest limit to decide CBT, not to wait for 180 days before proceeding to CBT. It was allowed to proceed to CBT if there were only few opportunities to find appropriate UD. When a suitable UD was not identified within 180 days from starting the UD search, patients were eligible for a single-arm phase 2 clinical study to test effectiveness of CBT in patients with CR status (P2 study).[11] Patients who died before 180 days from starting UD search were categorized as “death before transplantation,” whereas patients who survived more than 180 days from starting UD search but did not undergo any SCT were categorized as “no transplantation.”

For the P2 study, only patients in remission or equivalent status were eligible to ensure appropriate general conditions of recipients.[11] However, there were a significant number of CBT recipients who could not achieve remission or did not fulfill eligibility criteria for the P2 study. All patients who registered to this observational study were included in the present study. To assess patients’ risk of non-relapse mortality (NRM), hematopoietic SCT-specific comorbidity index (HCT-CI) was collected.[12] The refined disease risk index (rDRI) was assessed for patient stratification.[13] The study protocol was approved by the Institutional Review Board of Nagoya University Hospital (No.

2006-8058 for the P2 study and No. 2006-8059 for the observational study) and all participating hospitals, and written informed consent was obtained from each patient in accordance with the Declaration of Helsinki. The P2 study has been registered at UMIN-CTR as UMIN000000600 (https://upload.umin.ac.jp/cgi-bin/ctr_e/ctr_view.cgi?recptno=R000000710).

Graft Selection

Donor choice between HLA-7/8 or -6/8 allele-matched UD and CB was at the discretion of treating physicians and the transplantation center. For the P2 study, CB contained $>2.0 \times 10^7$ frozen total nucleated cell counts (TNCC) per kilogram of recipient, and no more than 2 of 6 HLA serological mismatches between each CB and patient were required. However, a common UCB unit selection strategy was applied to choose a CB unit in the observational study, in which we chose the highest TNCC within 2 loci mismatch among HLA-A, -B, and -DR loci at the antigen level. The overall graft selection strategy for UD is almost identical to National Marrow Donor Program (NMDP) recommendations, whereas that for CB is somewhat different from NMDP recommendations.[6, 14, 15] In NMDP recommendations, HLA class II with DRB1 is tested at allelic level, and the cell dose requirement is higher than our criteria. If it was not possible to find appropriate CB unit as described above, CB more than 2 of 6 HLA-antigen mismatch or less than 2.0×10^7 frozen TNCC could be selected at the discretion of the transplantation center under the observational study arm.

Transplant Procedure

The conditioning regimen was standard dose conditioning in principle. Standard dose conditioning included busulfan/cyclophosphamide; cyclophosphamide/total body irradiation (TBI); melphalan/TBI. Dose intensified conditioning included high-dose cytarabine/cyclophosphamide/TBI; busulfan/cyclophosphamide/TBI. Dose reduced conditioning

included fludarabine/cyclophosphamide with or without TBI; fludarabine/melphalan. Graft-versus-host disease (GVHD) prophylaxis was recommended to use either combination of methotrexate plus cyclosporine or methotrexate plus tacrolimus. Anti-thymocyte globulin was not used in any patients. Other supportive care measures were described elsewhere.[11]

Study Endpoints and Statistical Analysis

The primary endpoint of the present study was overall survival (OS) of CBT and UDT. Secondary endpoints included cumulative incidences of NRM and relapse and disease-free survival (DFS). Neutrophil recovery was defined as achieving an absolute neutrophil count of at least 500/μl for 3 consecutive measurements. Platelet recovery was defined as a count of 20,000/mL without transfusion support. Primary graft failure was defined as failure to achieve neutrophil recovery by day 42. Patients who survived to day 14 and died before neutrophil engraftment were classified as having primary graft failure. Relapse was defined as a recurrence of the underlying hematological disease. NRM was defined as death during continuous remission. DFS was defined as survival in a state of continuous remission. Treatment failure included both relapse and NRM. Diagnosis and clinical grading of acute and chronic graft-versus-host disease (GVHD) were performed according to established criteria.[16, 17] OS and DFS were calculated using the Kaplan-Meier estimate and then compared by the log-rank test.[18] Survival analysis was performed from the date of transplantation to that of event. Probabilities of neutrophil and platelet recovery, NRM, and relapse were calculated using the cumulative incidence estimate to consider competing risks and then compared by Gray's test.[19, 20] Cox proportional hazard analyses were applied to identify significant risk factors for DFS and OS.[21] Competing risk regression models were used for NRM and relapse. By utilizing risk factors, a final multivariate regression model was constructed to assess the difference in graft sources at each endpoint. Multivariate models were built using a forward stepwise selection method

with a threshold *P*-value of less than 0.1. Regardless of the level of significance, the main variable of interest, graft source (CBT vs. UDT), was considered in all steps of model construction. Other variables tested are as follows. Variables applied for analyses of both UDT and CBT were patient age (continuous variable), disease status at transplantation (CR vs. non-CR), rDRI (low vs. intermediate vs. high vs. very high), HCT-CI (0–2 vs. ≥3), intensity of conditioning regimen (intensified vs. others), and diagnosis (AML vs. ALL vs. MDS). The variable applied for analyses of UDT only was no. of HLA-allele mismatches (0, 1, and 2). Variables applied for analyses of CBT only were no. of HLA-serological mismatches (0, 1, 2, and 3), TNCC dose ($\geq 2.73 \times 10^7$/kg vs. $<2.73 \times 10^7$/kg), and CD34+ cell dose ($\geq 1.08 \times 10^5$/kg vs. $<1.08 \times 10^5$/kg). All statistical analyses were performed using Stata software (StataCorp, College Station, TX, USA) and EZR (Saitama Medical Center, Jichi Medical University, Saitama, Japan),[22] a graphical user interface for R (version 3.0.2; The R Foundation for Statistical Computing, Vienna, Austria).

RESULTS

Patient Characteristics

From March 2007 to March 2015, 231 patients underwent provisional registration (Figure 1). Data cutoff occurred on November 30, 2017. After provisional registration, we attempted to find appropriate UD according to the study protocol. In total, 119 patients received CBT and 91 patients received UDT. Six patients withdrew from the study and three died before transplantation. Twelve patients did not receive either UDT or CBT. Of these, five received HLA-mismatched peripheral blood SCT from a family donor and seven chose not to receive allogeneic SCT. Of 119 CBT recipients, 62 patients were eligible and registered to the P2 study reported elsewhere,[11] and the other 57 CB recipients were ineligible for the P2 study. The median patient age at transplant was 39 years in both groups (Table 1). Body weight was significantly heavier in UDT, but the difference was small (median difference, 3.7 kg; $P = 0.037$, Student's t-test). Diagnosis, disease status at transplantation, rDRI, and HCT-CI did not differ between UDT and CBT, except disease status at transplantation of MDS patients. Among MDS patients, two patients in CBT group were non-CR status after chemotherapy, whereas two patients in UDT group were CR status after chemotherapy at the transplantation, respectively (Table 1). The median follow-up for survivors was 65.9 months (range, 30.1–112.7 months) in CBT and 62.0 months (range, 26.7–120.0 months) in UDT.

Characteristics of CBT and UDT

The median time from provisional registration to transplant was 99 days (range, 8–286 days) in CBT and 126 days (range, 77–261 days) in UDT (median difference, 27 days, $P < 0.0001$). The median time from registration to transplantation in CBT and UDT was 105 days vs. 129 days in transplant at CR, whereas the time was 100 days vs. 151 days in transplant at non-CR, respectively. Among AML and ALL patients, the number of patients who received SCT during non-CR status

was 24 patients in CBT and 17 patients in UDT. Of these, 14 patients in CBT and 11 patients in UDT never exhibited CR from initiation of treatment, whereas 10 and 6 patients once experienced CR and thereafter relapsed during the donor search in CBT and UDT, respectively (median, 171 days in CBT and 168 days in UDT, $P = 0.71$). Among those who exhibited relapse after CR, the duration of CR in 8 of 10 CBT and 5 of 6 UDT was longer than 100 days. The numbers of HLA-serological and allele-level mismatches were significantly different between CBT and UDT ($P < 0.001$). Of UDT, 22 were HLA-7/8 allele-matched and three were HLA-6/8 allele-matched (HLA-C and -DRB1 allele mismatch). One-hundred and fourteen of 119 (95.8%) patients in CBT and 78 of 91 (85.7%) patients in UDT received myeloablative conditioning. Dose intensity of conditioning regimen was also significantly different between groups; a dose intensified regimen was more frequently adopted in CBT (Table 1, $P < 0.001$). More than 90% of patients received tacrolimus and short-term methotrexate as GVHD prophylaxis in both CBT (111/119, 93.3%) and UDT (86/91, 94.5%). The median TNCC and CD34+ cell number were 2.19×10^8/kg (range, 0.24–4.70) and 1.83×10^6/kg (range, 0.17–5.80) in bone marrow; 5.64×10^8/kg (range, 3.25–8.79) and 2.07×10^6/kg (range, 1.74–6.52) in peripheral blood stem cell; and 2.70×10^7/kg (range, 1.82–6.06) and 1.02×10^5/kg (range, 0.24–2.82) in CB, respectively.

Engraftment

Successful engraftment was observed in 106 (89.1%) CBT recipients and 86 (94.5%) UDT recipients. Of cases that did not demonstrate engraftment, 10 of 13 (8.4%) cases in CBT and 2 of 5 (2.2%) cases in UDT were attributed to graft failure. The median time to neutrophil engraftment was 21 days (range, 11–43) after CBT, and 15 days (range, 10–27) after UDT. The median time to platelet engraftment was 38 days (range, 18–130) after CBT, and 25.5 days (range, 10–97) after UDT.

Univariate and Multivariate Analyses of Risk Factors Associated with CBT and UDT

Because CBT and UDT reportedly have distinct risk factors, univariate and multivariate analyses were performed to identify risk factors among CBT and UDT, separately (Supplemental Table S1). In CBT, rDRI and diagnosis were significant factors of relapse; rDRI was significant for DFS and OS. In UDT, rDRI and disease status at transplant were significant risks for DFS and OS. Comparing the OS between HLA-8/8 matched UDT and HLA-7/8 or 6/8 matched UDT, there was no significant difference ($P = 0.60$). There was no significant risk factor observed specifically in UDT or CBT.

In multivariate analyses including both UDT and CBT, graft source (CBT vs. UDT) was not a significant risk factor for NRM, relapse, DFS, and OS. In the final multivariate model, no covariate was statistically significant in NRM, whereas rDRI and diagnosis remained significant covariates in relapse, DFS, and OS analyses (Table 2). In adjusted analyses of DFS and OS, CBT and UDT showed similar outcomes: DFS [UDT as reference; CB: hazard ratio (HR), 1.05; 95% confidence interval (95% CI), 0.70–1.59, $P = 0.81$]; OS (CB: HR, 1.17; 95% CI, 0.78–1.77, $P = 0.45$) (Table 2).

Survival Analysis of CBT and UDT

Of 119 CBT recipients, 56 patients died at data cutoff, leading to a median OS of 65.9 months and a 2-year OS rate of 62.2% (95% CI, 52.8–70.2). Of 91 UDT recipients, 38 patients died at data cutoff, leading to a median OS of 61.9 months and a 2-year OS rate of 60.9% (95% CI, 50.0–70.2) (Figure 2, log-rank, $P = 0.59$). In CBT, we observed 56 treatment failures (relapse, n = 37; NRM, n = 19) and 5 cases of second SCT; in UDT, we observed 43 treatment failures (relapse, n = 24; NRM, n = 19) and 2 cases of second SCT. The 2-year DFS was 57.5% (95% CI, 47.8–66.1) in CBT

and 56.1% (95% CI, 45.0–65.8) in UDT (Figure 3, log-rank, $P = 0.92$). When stratified with rDRI, UDT and CBT exhibited almost comparable survival rates (Figure 4). Stratification by rDRI worked excellently in each rDRI risk group except the rDRI-low group, which included a limited number of patients. Disease status at transplantation also efficiently segregated OS both in CBT and UDT and demonstrated comparable survival (Supplemental Figure S1). Comparing CBT with UDT in the subgroup of patients in non-CR at transplantation, we did not observe significant difference between CBT and UDT (log-rank, $P = 0.96$).

NRM, Relapse, and Treatment after Relapse

The cumulative incidences of NRM were 14.7% (95% CI, 8.9–21.7%) at 2 years and 16.0% (95% CI, 9.9–23.5%) at 5 years in CBT, and 20.2% (95% CI, 12.6–29.1%) at 2 years and 21.4% (95% CI, 13.6–30.4%) at 5 years in UDT (Figure 5A), respectively (Gray's test, $P = 0.38$). The cumulative incidences of relapse were 29.7% (95% CI, 21.6–38.2%) at 2 years and 32.4% (95% CI, 24.1–41.1%) at 5 years in CBT, and 24.9% (95% CI, 16.5–34.3%) at 2 years and 27.3% (95% CI, 18.4–36.8%) at 5 years in UDT (Figure 5B), respectively (Gray's test, $P = 0.45$).

Of CBT and UDT, five and one patients with graft failure received second transplantation, and successful engraftment was observed in four and one patients, respectively. Due to relapse after first SCT, 18 CBT recipients and 6 UDT recipients underwent second SCT, and 2 of those CBT recipients and 1 of those UDT recipients maintained CR by second SCT and survived without disease at the time of data collection.

After both CBT and UDT, several patients exhibited late treatment failure, particularly more than 12 months post-CBT (Figures 2 and 3). To elucidate reasons for late treatment failure, we examined deaths after 12 months of transplantation. In CBT, there were 17 treatment failures after 12 months, including 13 relapses. The causes of death were as follows: disease progression, n = 12;

infection, n = 3; and secondary malignancy, n = 2. In UDT, there were 10 treatment failures after 12 months, including 7 relapses. The causes of death were as follows: disease progression, n = 7; GVHD, n = 2; and non-infectious pulmonary disease, n = 1.

DISCUSSION

The results of the present study demonstrated that single-unit CBT and HLA well-matched UDT could convey comparable survival. In terms of NRM and relapse rate, there was no significant difference between CBT and UDT. We observed that there was no significant difference in NRM and relapse comparing CBT with UDT. Although we attempted to discover risk factors specifically associated with CBT or UDT, we did not detect any significant factor. rDRI stratified both outcomes after CBT and UDT efficiently and was a significant risk factor across CBT and UDT.[13] When stratified with rDRI, survival rates after CBT and UDT were almost superimposable in each rDRI score. In multivariate analysis adjusted with rDRI, we could not detect any significant difference between CBT and UDT.

From the results of previous retrospective studies, CBT and UDT can deliver almost comparable survival.[4-7, 23] However, it is always difficult to make a simple comparison between CBT and UDT due to the distinct characteristics of the respective graft sources. Risk factors specific for each graft are different,[24] making it difficult to analyze both CBT and UDT in the same multivariate model. CB is usually readily available within 30 days, whereas obtaining appropriate UD usually takes 1 month or longer.[9, 10] Patients who experienced disease relapse during the UD search period would likely select urgent CBT or haploidentical donor (HID) transplantation. This is likely to increase non-remission patients in the alternative donor group.[1, 7] Therefore, the difference in the length of the UD search period is one of the sources of bias. Delay of UD identification is often correlated with negative outcomes in patients with acute leukemia, such as increased relapse and NRM.[25] To minimize such recognized and unrecognized biases, we prospectively recruited patients, and most of the registered patients received either CBT or UDT.

In the present study, all patients underwent the same provisional registration process, and most of the registered patients further underwent transplantation within a reasonable time period.

Accordingly, we believe that comparability exists between CBT and UDT. Although we still observed a 30 day longer time to transplantation in UDT, we did not find any significant factors associated with outcomes except rDRI during risk factor exploration.[13] Thus, we believe we successfully controlled for other potential biases within the present study. Patient age and HCT-CI are proven important factors[12] that should have influenced the outcome; however, these factors were not significant variables in the present study. Because we recruited only relatively young patients to investigate myeloablative SCT, the age distribution of patients was limited. We considered this to be the main reason we did not observe "patient age" as a significant risk factor.

During the last decade, HID transplantation has emerged as a choice of alternative donor transplantation.[26, 27] Particularly, after the use of post-transplant cyclophosphamide was developed and standardized as a therapeutic option, the number of patients who received HID transplantation increased dramatically. Presently, HID is considered as a relevant alternative donor source.[28] In the era during which the present study was performed, HID transplantation was still under investigation. Thus, we included only five HID transplant recipients in the present study. The influence of HID transplantation was trivial in the current study. Ongoing studies comparing HID transplantation with CBT or UDT could reveal the preference of HID in patients with HLA well-matched UD unavailable.

In the previous studies, NRM after CBT was higher than that after UDT. However, we observed similar incidences of NRM between CBT and UDT. Because we recruited relatively young and fit patients due to the myeloablative SCT setting, we think this is the main reason why we observed considerably low incidences of NRM. The low NRM rate after CBT probably reflected a comparable outcome with UDT. If nonmyeloablative conditioning is used in older patients, the transplant outcome after nonmyeloablative CBT would be significantly lower.[29, 30] Reducing the conditioning intensity is expected not only to reduce NRM but also to increase

disease relapse.[31] Furthermore, an increase in graft failure is also anticipated, particularly after CBT.[32] Thus, the outcome after UDT may be superior to CBT in the nonmyeloablative SCT setting. To overcome the potential problem with engraftment after CBT, several novel methods are under investigation.[33, 34] Increased homing efficiency and expanded stem cell number are now under development. However, these options have not been clinically approved. Simultaneously, we must pay careful attention to relapse. Several recent studies have attempted to enhance conditioning regimen intensity to reduce the incidence of relapse in non-irradiation conditioning.[35, 36] These trials are expected to lower graft failure and relapse rates without increasing NRM.

In conclusion, single-unit CBT for adult hematological malignancies demonstrated a similar survival rate to UDT in this prospective cohort study. Accordingly, choosing CB as an alternative donor source can now be considered in parallel with choosing UD, which may result in achieving a timely transplant with the patient in better remission status.

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Author contributions

S.T. and T.N. designed and conducted the study, interpreted data, and wrote the manuscript; M.S., T.K., Kotaro M., Yukiyasu O., T.G., A.K., K.O., Yasushi O., Noriko F., Nobuharu F., H.Y., M.K., H.I., N.K., T.E., H.A., M.O., S.I., Y.N., M.N., Y.N., S.K., and Y.T. recruited patients and collected clinical data; A.Y., R.S., Y.K., and Y.A. collected and analyzed data; Koichi M. and M.M. supervised the study. All authors read and approved the final manuscript.

Conflict of interest disclosure: Authors declare that there is no relevant competing financial interest.

355 References

356 1. Rocha V, Labopin M, Sanz G, Arcese W, Schwerdtfeger R, Bosi A*, et al.* Transplants of umbilical-
357 cord blood or bone marrow from unrelated donors in adults with acute leukemia. *N Engl J Med*
358 2004 Nov; **351**(22): 2276-2285.
359
360 2. Laughlin MJ, Eapen M, Rubinstein P, Wagner JE, Zhang MJ, Champlin RE*, et al.* Outcomes after
361 transplantation of cord blood or bone marrow from unrelated donors in adults with leukemia. *N*
362 *Engl J Med* 2004 Nov; **351**(22): 2265-2275.
363
364 3. Ballen KK, Gluckman E, Broxmeyer HE. Umbilical cord blood transplantation: the first 25 years
365 and beyond. *Blood* 2013 Jul 25; **122**(4): 491-498.
366
367 4. Atsuta Y, Morishima Y, Suzuki R, Nagamura-Inoue T, Taniguchi S, Takahashi S*, et al.*
368 Comparison of unrelated cord blood transplantation and HLA-mismatched unrelated bone
369 marrow transplantation for adults with leukemia. *Biol Blood Marrow Transplant* 2012 May;
370 **18**(5): 780-787.
371
372 5. Eapen M, Rocha V, Sanz G, Scaradavou A, Zhang MJ, Arcese W*, et al.* Effect of graft source on
373 unrelated donor haemopoietic stem-cell transplantation in adults with acute leukaemia: a
374 retrospective analysis. *Lancet Oncol* 2010 Jul; **11**(7): 653-660.
375
376 6. Spellman SR, Eapen M, Logan BR, Mueller C, Rubinstein P, Setterholm MI*, et al.* A perspective
377 on the selection of unrelated donors and cord blood units for transplantation. *Blood* 2012 Jul 12;
378 **120**(2): 259-265.
379
380 7. Terakura S, Atsuta Y, Tsukada N, Kobayashi T, Tanaka M, Kanda J*, et al.* Comparison of
381 Outcomes of 8/8 and 7/8 Allele-Matched Unrelated Bone Marrow Transplantation and Single-
382 Unit Cord Blood Transplantation in Adults with Acute Leukemia. *Biol Blood Marrow Transplant*
383 2016 Feb; **22**(2): 330-338.
384
385 8. Ustun C, Giannotti F, Zhang MJ, Wang HL, Brunstein C, Labopin M*, et al.* Outcomes of UCB
386 transplantation are comparable in FLT3+ AML: results of CIBMTR, EUROCORD and EBMT
387 collaborative analysis. *Leukemia* 2017 Jun; **31**(6): 1408-1414.
388
389 9. Barker JN, Krepski TP, DeFor TE, Davies SM, Wagner JE, Weisdorf DJ. Searching for unrelated
390 donor hematopoietic stem cells: availability and speed of umbilical cord blood versus bone marrow.
391 *Biol Blood Marrow Transplant* 2002; **8**(5): 257-260.
392
393 10. Kawashima N, Nishiwaki S, Shimizu N, Kamoshita S, Watakabe K, Yokohata E*, et al.* Outcomes
394 of strategic alternative donor selection or suspending donor search based on Japan Marrow Donor
395 Program coordination status. *Int J Hematol* 2018 May; **107**(5): 551-558.
396
397 11. Terakura S, Nishida T, Sawa M, Kato T, Miyao K, Ozawa Y*, et al.* Prospective Phase 2 Study of
398 Umbilical Cord Blood Transplantation in Adult Acute Leukemia and Myelodysplastic Syndrome.
399 *Biol Blood Marrow Transplant* 2019 Sep 20.
400
401 12. Sorror M, Maris M, Storb R, Baron F, Sandmaier B, Maloney D*, et al.* Hematopoietic cell
402 transplantation (HCT)-specific comorbidity index: a new tool for risk assessment before allogeneic
403 HCT. *Blood* 2005 Oct; **106**(8): 2912-2919.
404
405 13. Armand P, Kim HT, Logan BR, Wang Z, Alyea EP, Kalaycio ME*, et al.* Validation and refinement
406 of the Disease Risk Index for allogeneic stem cell transplantation. *Blood* 2014 Jun 5; **123**(23):

3664-3671.

14. Bray RA, Hurley CK, Kamani NR, Woolfrey A, Muller C, Spellman S*, et al.* National marrow donor program HLA matching guidelines for unrelated adult donor hematopoietic cell transplants. *Biol Blood Marrow Transplant* 2008 Sep; **14**(9 Suppl): 45-53.

15. Dehn J, Spellman S, Hurley CK, Shaw BE, Barker JN, Burns LJ*, et al.* Selection of unrelated donors and cord blood units for hematopoietic cell transplantation: guidelines from the NMDP/CIBMTR. *Blood* 2019; **134**(12): 924-934.

16. Przepiorka D, Weisdorf D, Martin P, Klingemann H, Beatty P, Hows J*, et al.* 1994 Consensus Conference on Acute GVHD Grading. *Bone Marrow Transplant* 1995 Jun; **15**(6): 825-828.

17. Lee SJ, Vogelsang G, Flowers MED. Chronic graft-versus-host disease. *Biology of Blood and Marrow Transplantation* 2003; **9**(4): 215-233.

18. Kaplan E, Meier. P. Nonparametric estimation from incomplete observations. *J Am Stat Assoc* 1958; **53:** 457-481.

19. Gooley TA, Leisenring W, Crowley J, Storer BE. Estimation of failure probabilities in the presence of competing risks: new representations of old estimators. *Stat Med* 1999 Mar 30; **18**(6): 695-706.

20. Fine JP, Gray RJ. A Proportional Hazards Model for the Subdistribution of a Competing Risk. *Journal of the American Statistical Association* 1999 1999/06/01; **94**(446): 496-509.

21. Cox DR. Regression Models and Life-Tables. *Journal of the Royal Statistical Society Series B (Methodological)* 1972; **34**(2): 187-220.

22. Kanda Y. Investigation of the freely available easy-to-use software 'EZR' for medical statistics. *Bone Marrow Transplant* 2013 Mar; **48**(3): 452-458.

23. Rocha V, Labopin M, Sanz G, Arcese W, Schwerdtfeger R, Bosi A*, et al.* Transplants of Umbilical-Cord Blood or Bone Marrow from Unrelated Donors in Adults with Acute Leukemia. *New England Journal of Medicine* 2004; **351**(22): 2276-2285.

24. Elmariah H, Pratz KW. Role of Alternative Donor Allogeneic Transplants in the Therapy of Acute Myeloid Leukemia. *J Natl Compr Canc Netw* 2017 Jul; **15**(7): 959-966.

25. Frassoni F, Labopin M, Powles R, Mary JY, Arcese W, Bacigalupo A*, et al.* Effect of centre on outcome of bone-marrow transplantation for acute myeloid leukaemia. Acute Leukaemia Working Party of the European Group for Blood and Marrow Transplantation. *Lancet* 2000 Apr 22; **355**(9213): 1393-1398.

26. Bashey A, Zhang X, Sizemore CA, Manion K, Brown S, Holland HK*, et al.* T-cell-replete HLA-haploidentical hematopoietic transplantation for hematologic malignancies using post-transplantation cyclophosphamide results in outcomes equivalent to those of contemporaneous HLA-matched related and unrelated donor transplantation. *J Clin Oncol* 2013 Apr 1; **31**(10): 1310-1316.

27. Shabbir-Moosajee M, Lombardi L, Ciurea SO. An overview of conditioning regimens for haploidentical stem cell transplantation with post-transplantation cyclophosphamide. *Am J Hematol* 2015 Jun; **90**(6): 541-548.

28. Ruggeri A, Labopin M, Sanz G, Piemontese S, Arcese W, Bacigalupo A*, et al.* Comparison of

outcomes after unrelated cord blood and unmanipulated haploidentical stem cell transplantation in adults with acute leukemia. *Leukemia* 2015 Sep; **29**(9): 1891-1900.

29. Tanaka M, Miyamura K, Terakura S, Imai K, Uchida N, Ago H*, et al.* Comparison of cord blood transplantation with unrelated bone marrow transplantation in patients older than fifty years. *Biol Blood Marrow Transplant* 2015 Mar; **21**(3): 517-525.

30. Rio B, Chevret S, Vigouroux S, Chevallier P, Furst S, Sirvent A*, et al.* Decreased nonrelapse mortality after unrelated cord blood transplantation for acute myeloid leukemia using reduced-intensity conditioning: a prospective phase II multicenter trial. *Biol Blood Marrow Transplant* 2015 Mar; **21**(3): 445-453.

31. Scott BL, Pasquini MC, Logan BR, Wu J, Devine SM, Porter DL*, et al.* Myeloablative Versus Reduced-Intensity Hematopoietic Cell Transplantation for Acute Myeloid Leukemia and Myelodysplastic Syndromes. *J Clin Oncol* 2017 Apr 10; **35**(11): 1154-1161.

32. Terakura S, Kuwatsuka Y, Yamasaki S, Wake A, Kanda J, Inamoto Y*, et al.* GvHD prophylaxis after single-unit reduced intensity conditioning cord blood transplantation in adults with acute leukemia. *Bone Marrow Transplant* 2017 Sep; **52**(9): 1261-1267.

33. Dahlberg A, Milano F. Cord blood transplantation: rewind to fast forward. *Bone Marrow Transplant* 2017 Jun; **52**(6): 799-802.

34. Barker JN, Kurtzberg J, Ballen K, Boo M, Brunstein C, Cutler C*, et al.* Optimal Practices in Unrelated Donor Cord Blood Transplantation for Hematologic Malignancies. *Biol Blood Marrow Transplant* 2017 Jun; **23**(6): 882-896.

35. Sharma P, Pollyea DA, Smith CA, Purev E, Kamdar M, Haverkos B*, et al.* Thiotepa-Based Intensified Reduced-Intensity Conditioning Adult Double-Unit Cord Blood Hematopoietic Stem Cell Transplantation Results in Decreased Relapse Rate and Improved Survival Compared with Transplantation Following Standard Reduced-Intensity Conditioning: A Retrospective Cohort Comparison. *Biol Blood Marrow Transplant* 2018 Aug; **24**(8): 1671-1677.

36. Yamamoto H, Uchida N, Yuasa M, Kageyama K, Ota H, Kaji D*, et al.* A Novel Reduced-Toxicity Myeloablative Conditioning Regimen Using Full-Dose Busulfan, Fludarabine, and Melphalan for Single Cord Blood Transplantation Provides Durable Engraftment and Remission in Nonremission Myeloid Malignancies. *Biol Blood Marrow Transplant* 2016 Oct; **22**(10): 1844-1850.

Figure legends

Figure 1. Treatment allocation according to CONSORT guidelines. AML, acute myeloid leukemia; ALL, acute lymphoblastic leukemia; MDS, myelodysplastic syndrome; JMDP, Japan Marrow Donor Program; UD, unrelated donor; CBT, cord blood transplantation; CR, complete remission; BMT, bone marrow transplantation; PBSCT, peripheral blood stem cell transplantation; SCT, stem cell transplantation.

Figure 2. Overall survival (OS) depicted with Kaplan–Meier estimates of cord blood transplantation (CBT) and unrelated donor transplantation (UDT) recipients. There was no statistically significant difference in OS between CBT and UDT (log-rank test, $P = 0.59$). HR, hazard ratio.

Figure 3. Disease-free survival (DFS) depicted with Kaplan–Meier estimates of cord blood transplantation (CBT) and unrelated donor transplantation (UDT) recipients. DFS was comparable between CBT and UDT (log-rank test, $P = 0.92$). HR, hazard ratio.

Figure 4. Overall survival (OS) stratified by refined disease risk index (rDRI) in (A) cord blood transplantation (CBT) and (B) unrelated donor transplantation (UDT). Int, intermediate.

Figure 5. Cumulative incidence of (A) non-relapse mortality (NRM) and (B) relapse after stem cell transplantation according to graft source. The 2-year cumulative incidences of NRM were 14.7% (95% CI, 8.9–21.7%) in CBT and 20.2% (95% CI, 12.6–29.1%) in UDT (Gray's test, $P = 0.38$). The 2-year cumulative incidences of relapse were 30.6% (95% CI, 22.4–39.2%) in CBT and 24.9% (95% CI, 16.5–34.3%) in UDT (Gray's test, $P = 0.45$).

Table 1. Patient and transplant characteristics

	CBT		UDT		
	No.	%	No.	%	*P*-value
Number	119		91		
Patient age, years, median (range)	39 (16–55)		39 (17–55)		0.80
Body weight, kg, median (range)	55 (35–90)		59 (37–90)		0.037
Sex (male : female)	58:61	(49:51)	50:41	(55:45)	0.37
Diagnosis					0.29
AML	68	57	49	55	
ALL	38	32	37	40	
B-ALL	30	25	32	35	
T-ALL	8	7	5	5	
MDS	13	11	5	5	
AML – disease status at transplant					0.82
CR1	27	23	17	19	
CR2	19	16	16	18	
Non-CR	22	18	16	18	
ALL – disease status at transplant					0.84
B-ALL					
CR1	24	20	25	27	
CR2	5	4	6	7	
Non-CR	1	1	1	1	
T-ALL					
CR1	7	6	5	5	
CR2	0	0	0	0	
Non-CR	1	1	0	0	
MDS – disease status at transplant					0.015
CR after chemotherapy	0	0	2	2	
EB1	4	3	0	0	
EB2	7	6	3	3	
Overt leukemia	2	2	0	0	
Disease risk index					0.89
Low	8	7	6	7	
Int	67	56	56	61	
High	39	33	26	29	
Very high	5	4	3	3	
HCT-CI					0.20
0	91	77	54	60	
1	12	10	14	15	
2	5	4	10	11	
≥3	11	9	13	14	
No. of antigen-level mismatches in HLA-A/B/DR					< 0.001
0	3	3	83	91	
1	29	24	8	9	
2	83	70	0	0	
≥3	4	3	0	0	
No. of allele-level mismatches in HLA-A/B/C/DR					< 0.001
0	3	3	66	73	
1	4	3	22	24	
2	19	16	3	3	
3	30	25			
4	24	20			
5	22	19			
≥6	13	11			
Missing	4	3			
Conditioning regimen					< 0.001
Dose intensified †	83	70	5	6	
Standard dose ‡	31	26	73	80	
Dose reduced §	5	4	13	14	
GVHD prophylaxis					0.98
CyA plus sMTX	5	4	4	4	

Tac plus sMTX	111	93	86	95
Tac plus MMF	1	1	1	1
Missing	2	2	0	0
TNCC, median (range)				
BM （×10^8/kg)			2.19 (0.24–4.70)	
PBSC （×108/kg)			5.64 (3.25–8.79)	
CB （×107/kg)	2.70 (1.82–6.06)			
CD34+ cells, median (range)				
BM （×106/kg)			1.83 (0.17–5.80)	
PBSC （×106/kg)			2.07 (1.74–6.52)	
CB （×105/kg)	1.02 (0.24–2.82)			

AML, acute myeloid leukemia; ALL, acute lymphoblastic leukemia; MDS, myelodysplastic syndrome; EB, excess blasts; HCT-CI, hematopoietic stem cell transplant-specific comobidity index.

† Dose intensified conditioning included high-dose cytrabine/cyclophosphamide/total-body irradiation (TBI); busulfan/cyclophosphamide/TBI.

‡ Standard dose conditioning included busulfan/cyclophosphamide; cyclophosphamide/TBI; melphalan/TBI.

§ Dose reduced conditioning included fludarabine/cyclophosphamide with or without TBI; fludarabine/melphalan.

CyA, cyclosporine A; sMTX, short-term methotrexate; Tac, tacrolimus; MMF, mycophenolate mofetil.

TNCC, total nucleated cell counts; BM, bone marrow; PBSC, peripheral blood stem cell; CB, cord blood.

Table 2. Multivariate analysis including both CBT and UDT

	HR	95% CI	*P*-value
NRM	No significant variable		
CBT (vs. UDT)	0.75	0.40 – 1.42	0.38
Relapse			
CBT (vs. UDT)	1.38	0.79 – 2.34	0.26
rDRI	2.35	1.53 – 3.61	< 0.001
Diagnosis	0.60	0.44 – 0.82	0.001
DFS			
CBT (vs. UDT)	1.05	0.70 – 1.59	0.81
rDRI	2.21	1.62 – 3.01	< 0.001
Diagnosis	0.71	0.56 – 0.91	0.006
OS			
CBT (vs. UDT)	1.17	0.78 – 1.77	0.45
rDRI	2.30	1.69 – 3.13	< 0.001
Diagnosis	0.69	0.54 – 0.88	0.003

HR, hazard ratio; CI, confidence interval; NRM, non-relapse mortality; CBT, cord blood transplantation; UDT, unrelated donor transplantation; rDRI, refined Disease Risk Index; DFS, disease-free survival; OS, overall survival.

Figure 1

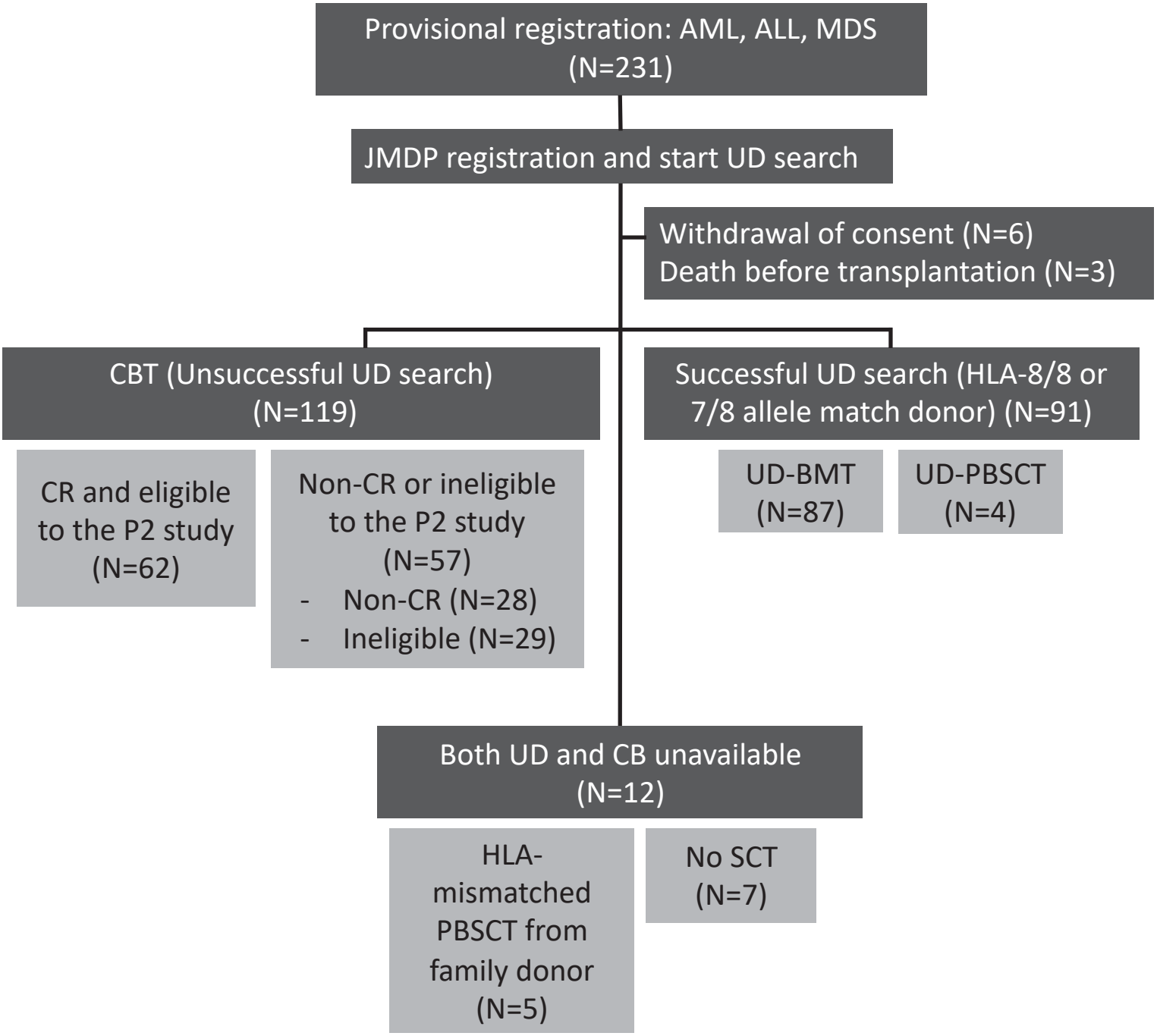
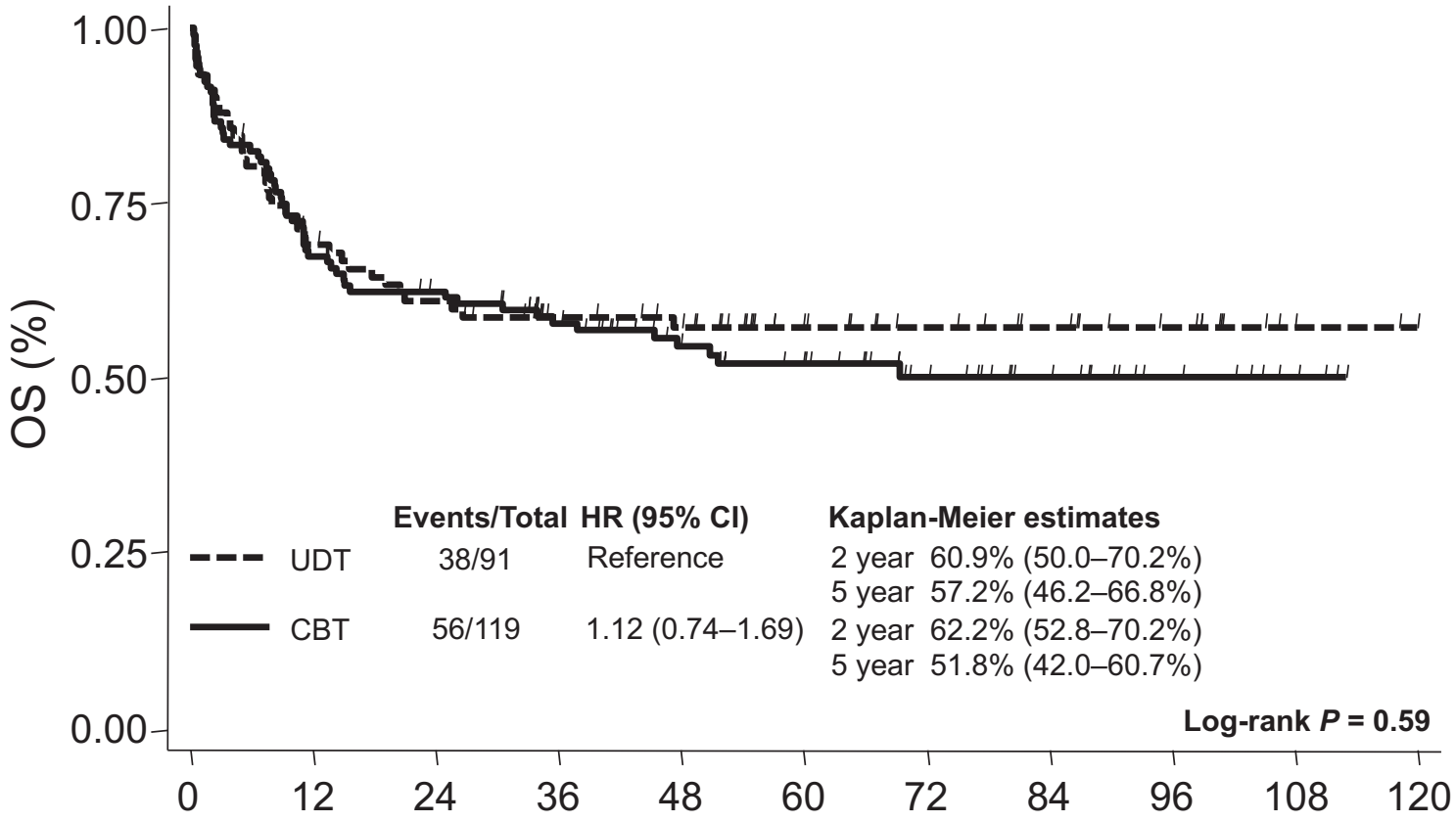

Provisional registration: AML, ALL, MDS (N=231)
JMDP registration and start UD search
Withdrawal of consent (N=6)
Death before transplantation (N=3)
CBT (Unsuccessful UD search) (N=119)
Successful UD search (HLA-8/8 or 7/8 allele match donor) (N=91)
CR and eligible to the P2 study (N=62)
Non-CR or ineligible to the P2 study (N=57)
- Non-CR (N=28)
- Ineligible (N=29)
UD-BMT (N=87)
UD-PBSCT (N=4)
Both UD and CB unavailable (N=12)
HLA-mismatched PBSCT from family donor (N=5)
No SCT (N=7)

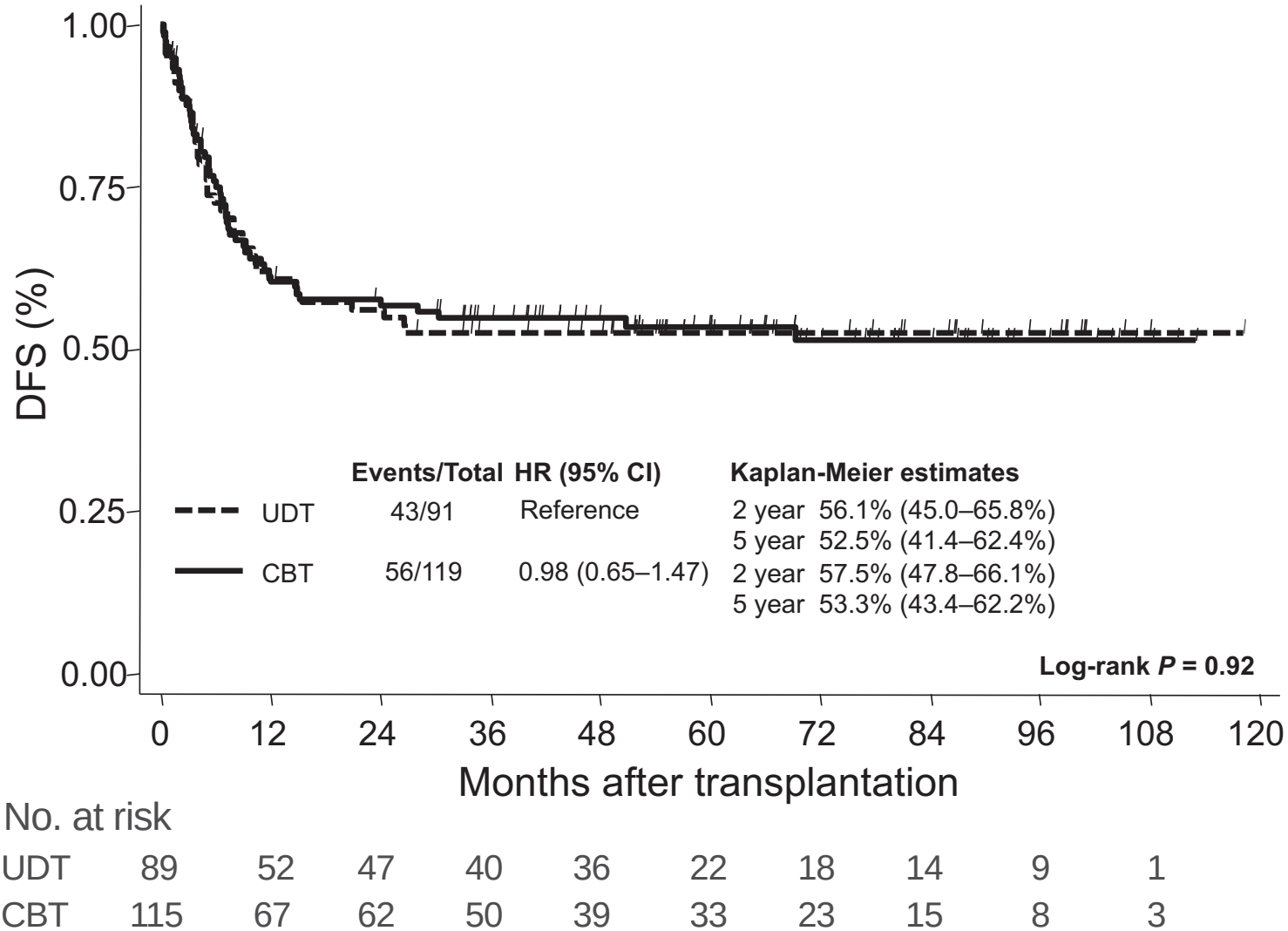
Figure 2



No. at risk

UDT	91	61	53	44	39	25	20	16	11	2
CBT	119	80	72	58	44	36	24	16	9	4

Figure 3



No. at risk

UDT	89	52	47	40	36	22	18	14	9	1
CBT	115	67	62	50	39	33	23	15	8	3

Figure 4

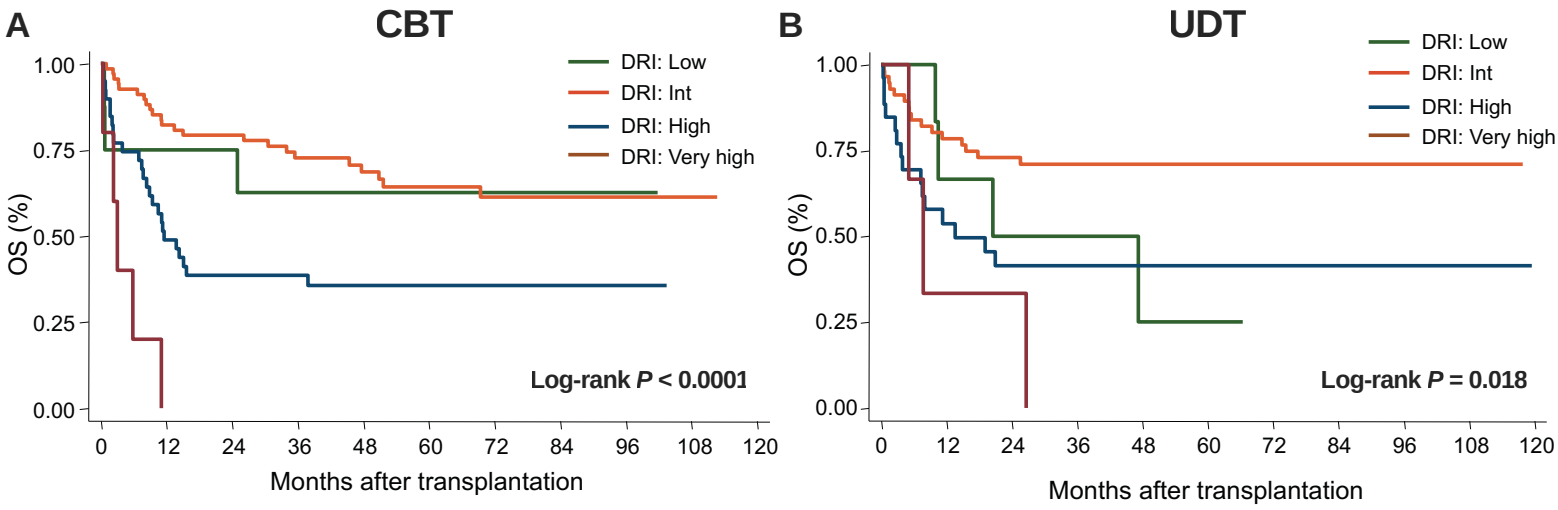
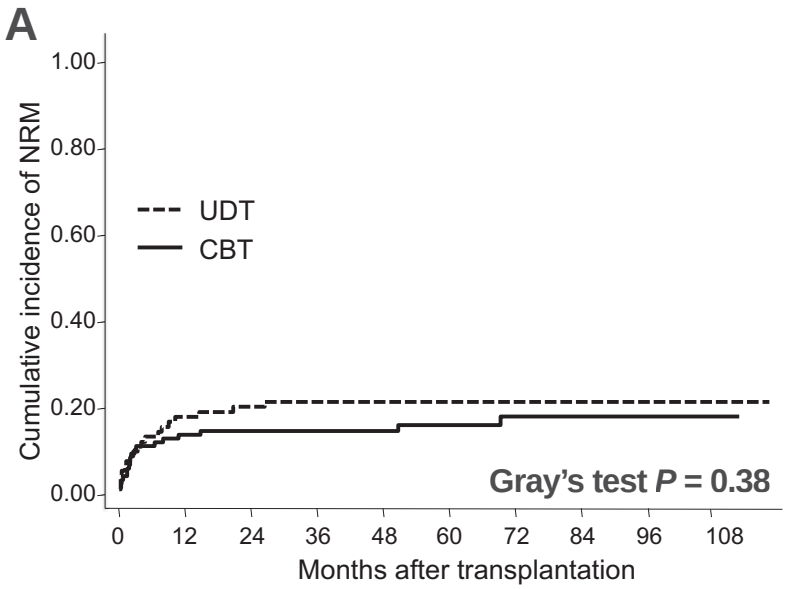
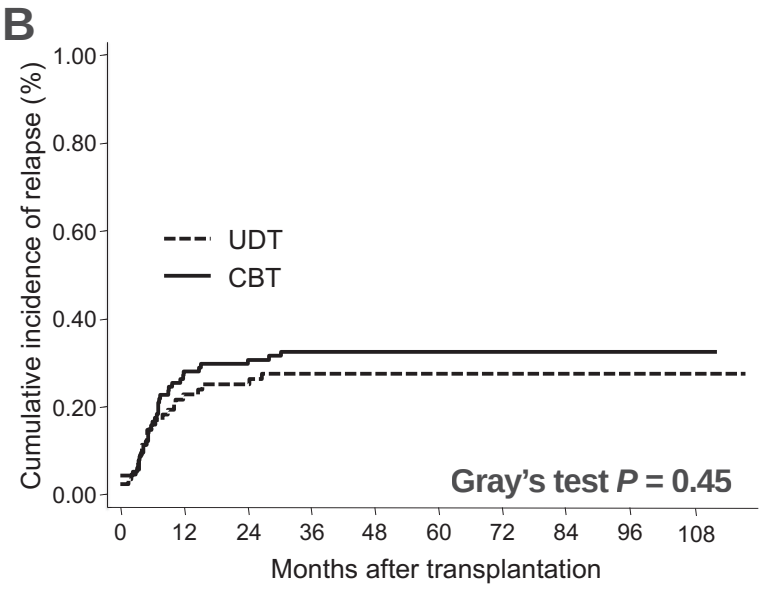

A
CBT
DRI: Low
DRI: Int
DRI: High
DRI: Very high
OS (%)
1.00
0.75
0.50
0.25
0.00
Log-rank P < 0.0001
0
12
24
36
48
60
72
84
96
108
120
Months after transplantation
B
UDT
DRI: Low
DRI: Int
DRI: High
DRI: Very high
OS (%)
1.00
0.75
0.50
0.25
0.00
Log-rank P = 0.018
0
12
24
36
48
60
72
84
96
108
120
Months after transplantation

Figure 5


A
1.00
0.80
0.60
0.40
0.20
0.00
Cumulative incidence of NRM
UDT
CBT
Gray's test P = 0.38
0
12
24
36
48
60
72
84
96
108
Months after transplantation


B
1.00
0.80
0.60
0.40
0.20
0.00
Cumulative incidence of relapse (%)
UDT
CBT
Gray's test P = 0.45
0
12
24
36
48
60
72
84
96
108
Months after transplantation