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Double umbilical cord blood transplantation for hematological malignancies: A long-term analysis from the SFGM-TC registry

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Allogeneic hematopoietic stem cell (HSC) transplantation is a curative treatment for many hematologic malignancies for which umbilical cord blood (UCB) represents an alternative source of HSCs. To overcome the low cellularity of one UCB unit, double UCB transplantation (dUCBT) has been developed in adults. We have analyzed the outcome of 136 patients who underwent dUCBT reported to the SFGM-TC registry between 2005 and 2007. Forty-six patients received myeloablative regimens, and 90 patients received reduced-intensity conditioning regimens. There were 84 cases of leukemia, 17 cases of non-Hodgkin lymphoma, 11 cases of myeloma, and 24 other hematologic malignancies. At transplantation, 40 (29%) patients were in complete remission. At day 60 after transplantation, the cumulative incidence of neutrophil recovery was 91%. We observed one UCB unit domination in 88% of cases. The cumulative incidence of day 100 acute graft-versus-host disease, chronic graft-versus-host disease, transplant-related mortality, and relapse at 2 years were 36%, 23%, 27%, and 28% respectively. After a median follow-up of 49.5 months, the 3-year probabilities of overall and progression-free survival were 41% and 35%, respectively, with a significant overall survival advantage when male cord engrafted male recipients. We obtained a long-term plateau among patients in complete remission, which makes dUCBT a promising treatment strategy for these patients. © **2013 Published by Elsevier Inc. on behalf of ISEH - Society for Hematology and Stem Cells.**

Umbilical cord blood (UCB) contains primitive hematopoietic progenitors capable of engraftment and spontaneous in vivo expansion to allow hematopoietic recovery in the allogeneic hematopoietic stem cell transplantation (allo-HSCT) setting [1–5]. According to these properties, UCB transplantation (UCBT) offers an alternative strategy for patients without human leukocyte antigen (HLA) identical sibling or unrelated donor, who represent approximately one third of patients in need of a potentially curative allo-HSCT [6]. UCB cells have many interesting properties,

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such as prompt availability [7], minimal infectious contamination risk, and immunologic immaturity of cells, allowing HLA incompatibilities and a decrease in the probability and severity of acute and chronic graft-versus-host disease (GVHD) [8,9]. UCBT has been developed firstly in children in related and then unrelated settings [10,11]. However, UCB contains fewer progenitors than bone marrow and peripheral stem cells, delaying hematopoietic recovery and leading to a high transplant-related mortality (TRM) [12] in case of graft failure as observed in adults. This main limit for UCBT led us to propose double UCBT (dUCBT) for adult patients when cellularity in a single unit is not sufficient. The first encouraging results led to place dUCB as an alternative stem cell source when no unrelated donor is available [13–15]. After dUCBT, many studies have demonstrated that the sustained hematopoiesis is derived almost exclusively from one of the two UCB units [14–16]. Most important defined criteria in the UCBT settings are HLA matching and total nucleated cells (TNC) number [17] in addition to $CD34^+$ cell content, which is still controversial [18,19]. We decided to perform a multicenter study of the Société Française de Greffe de Moelle et de Thérapie Cellulaire (SFGM-TC) registry to evaluate the different transplant outcomes after dUCBT for hematologic malignancies and to study the different parameters involved in the dominance of one UCB unit after transplantation.

Methods

Patients

This retrospective analysis was conducted on adult patients who underwent dUCBT for hematologic malignancies reported to the SFGM-TC registry between January 2005 and December 2007 after either myeloablative conditioning (MAC) or reduced-intensity conditioning (RIC). One hundred and thirty-six patients from 23 centers were included in the analysis. There were 88 men and 48 women, with a median age of 41 years (range, 18–66 years). There were 76 patients with acute leukemias (42 acute myeloid leukemia, 27 acute lymphoid leukemia, 5 secondary acute leukemia, and 2 biphenotypic acute leukemia), 8 patients with chronic leukemias (5 lymphoid and 3 myeloid), 10 patients with myelodysplasia, 24 patients with Hodgkin and non-Hodgkin lymphomas, 13 patients with multiple myelomas, and 5 patients with other myeloproliferative diseases. Forty-four patients received a previous autologous transplantation. The median interval between diagnosis and transplantation was 20.5 months (range, 3–385 months). At the time of allogeneic transplantation, 40 patients were in first complete remission (CR1), 40 patients were in second complete remission or greater, 21 patients were in partial remission (PR), and 23 patients were in less than PR. The last follow-up analysis was performed in May 2012. Patient characteristics are described in Table 1.

Transplantation procedures

UCB unit properties. Information concerning HLA typing was obtained from the database of the French network of cord blood banks on behalf of the "Agence de la Biomédecine" and Eurocord. HLA typing was evaluated across three loci: HLA-A, HLA-B, and HLA-DRB1; it was based on antigenic level for HLA-A and HLA-B, and on allelic level for HLA-DRB1. We divided the population into three groups according to HLA matching: (1) at least 4/6 HLA matching on HLA-A, -B, and -DRB1 without any mismatch on DRB1 alleles between the recipient and each UCB unit and the two units together (n = 77); (2) at least 4/6 HLA matching on HLA-A and HLA-B between the recipient and each unit, but without complete matching on DRB1 or with less than 4/6 HLA matching between the two units (n = 51); and (3) eight patients with more than two mismatches on the three loci. For ABO matching, a comparison between the patient and each UCB unit has been performed: there was a complete compatibility between the patient and the 2 units in 37 patients, one incompatibility in 49 cases and 2 incompatibilities in 50 cases. There were 42 sex-matched transplantations (between the 2 UCB units and the recipient), 64 with 1 mismatch (45 with male recipient and 19 with female recipient) and 30 with 2 mismatches (18 in male recipient and 12 in female recipient). For cytomegalovirus (CMV), 77 (57%) recipients were negative. The median number of harvested and infused TNCs per kilogram of the patient's weight was 4.6×10^7 (range, 1.6×10^7 to 12.2×10^7) and 3.1×10^7 (range, 0.6×10^7 to 12.1×10^7), respectively. The median number of harvested and infused $CD34^+$ cells was 1.8×10^5 (range, 0.3×10^5 to 10.9 x 10^5) and 1.4×10^5 (range, 0.2×10^5 to 9.3×10^5), respectively. The different UCB unit characteristics are shown in Table 2.

Conditioning regimen and GVHD prophylaxis. Conditioning regimens were classified as proposed by a recent Center for International Blood and Marrow Research workshop [20]. Forty-six patients received MAC, and 90 patients received RIC; 83% of the RIC was performed according to the Minnesota program [21]. The different conditioning regimens are described in Table 3.

The GVHD prophylaxis consisted of cyclosporine A plus mycophenolate mofetil in 77% of cases and other combinations in the other cases. In addition, antithymocyte globulins (ATG) were administered in 25% of cases (Table 3).

Chimerism analysis. Chimerism data were collected from chimerism and HLA French laboratories. Chimerism analysis was performed on marrow and blood samples (total white blood cells or $CD3^+$ cells) using polymerase chain reaction based on informative polymorphic short tandem repeat with an accuracy of ±5%. An evaluation has been done at 1, 2, and 3 months after transplantation; after three months, its frequency varied according to the centers' practices. Sixteen laboratories used quantitative polymerase chain reaction with accuracy less than 0.5%. A cord blood unit was defined as dominant when it represented more than 95% of the total documented cells after transplantation. Mixed chimerism (double chimera) was defined by the presence of cells from the two UCB units for at least 2 months after transplantation in accordance with recent data [22,23].

Statistical analysis

Neutrophil recovery was defined by an absolute neutrophil count of at least 500 cells/mm^3 for three consecutive days; platelet recovery was defined by a count of at least 50.000 cells/mm^3 for seven consecutive days without any transfusion support. GVHD was reported and graded according to published criteria [24]. Chronic GVHD was diagnosed according to standard criteria

Table 1. Patient characteristics

	All patients (n = 136)	MAC (n=46)	RIC (n=90)
Median age at diagnosis years (range)	38.5 (9–63)	33.5 (9–62)	42 (9–63)
Median age at transplantation years (range)	41.1 (18.3–66.6)	34.5 (18.3–63.0)	44.7 (18.6–66.6)
Gender: Male/Female	88 (65%)/48 (35%)	30 (65%)/16 (35%)	58 (65%)/32 (35%)
Median weight at transplantation kg (range)	69.5 (42–120)	69 (45–93)	70 (42–120)
Diagnosis			
AML	42 (31%)	15 (33%)	27 (30%)
ALL	27 (20%)	17 (37%)	10 (11%)
NHL	17 (12%)	4 (9%)	13 (14%)
MM	13 (9%)	1 (2%)	12 (14%)
MDS	10 (8%)	4 (9%)	6 (7%)
MPS (≠ CML)	5 (4%)	2 (4%)	3 (3%)
HG d	7 (5%)	0 (0%)	7 (8%)
CLL	5 (4%)	0 (0%)	5 (6%)
CML	3 (2%)	1 (2%)	2 (2%)
Biphenotypic AL	2 (1%)	1 (2%)	1 (1%)
Secondary AL	5 (4%)	1 (2%)	4 (4%)
Treatment lines before transplant			
1 line	37 (27%)	15 (33%)	22 (24%)
2 lines	53 (39%)	25 (54%)	28 (31%)
≥ 3 lines	30 (22%)	5 (11%)	25 (28%)
No previous treatment	14 (10%)	0 (0%)	14 (16%)
NA	2 (2%)	1 (2%)	1 (1%)
Previous autologous SCT	44 (32%)	4 (9%)	40 (44%)
Disease status at transplant			
CR1	40 (29.5%)	18 (39%)	22 (24%)
CR ≥ 2	40 (29.5%)	15 (33%)	25 (28%)
PR	21 (15%)	3 (7%)	18 (20%)
<PR	23 (17%)	8 (17%)	15 (17%)
Stable/chronic	9 (7%)	2 (4%)	7 (8%)
Unknown	3 (2%)	0 (0%)	3 (3%)

ALL = acute lymphoid leukemia; AL = acute leukemia; AML = acute myeloid leukemia; CLL = chronic lymphoid leukemia; CML = chronic myeloid leukemia; CR = complete remission; HG d = Hodgkin disease; MAC = myeloablative conditioning; NHL = non-Hodgkin lymphoma; MM = multiple myeloma; MDS = myelodysplastic syndrome; MPS = myeloproliferative syndrome; NA = not available; PR = partial remission; RIC = reduced intensity conditioning; SCT = stem cell transplantation.

for patients who survived at least 90 days after transplantation [25]. TRM was defined as death from any cause other than relapse occurring after transplantation. Relapse was defined on the basis of morphologic evidence of hematopoietic disease in bone marrow or other sites. Overall survival (OS) was defined as the time from transplantation to any cause of death, and progression-free survival (PFS) was defined as survival from transplantation to disease progression or death.

Categorical variables related to patients, disease, and transplantation procedure were analyzed using the chi-square test and continuous variables with a Mann–Whitney *U* test. Cumulative incidence curves in competing-risks settings were used to estimate incidence over time for neutrophil and platelet recovery, acute and chronic GVHD, TRM, and relapse. The models by Fine and Gray [26] were used to estimate pretransplant variables, and the competing-risks regression for the multivariate analysis. Overall survival and PFS were estimated using the Kaplan-Meier method with log-rank test for univariate analysis [27]. Cox proportional-hazards regression models were used to assess the influence of pretransplant variables on OS and PFS [28]. This multivariate analysis on OS and PFS was made after stratification on diagnosis studying the following variables: pretransplant disease status, age of the recipient, sex, HLA and ABO matching, CMV status of the recipient, number of infused cells, with or without ATG, and conditioning regimen. In addition, we have built a multivariate model on dominant UCB unit prediction accounting for matching variables and infused cell counts. Statistical analysis was performed with R statistical software (version 2.12).

Results

Engraftment

After transplantation, 127 (93%) patients engrafted with no significant difference between those with RIC or MAC. The cumulative incidences of neutrophil recovery at days 60 and 100 after transplantation were 91% (95% confidence interval [CI], 88.5–93.5) and 92% (95% CI, 89.6–94.6), respectively (Fig. 1A), with a median time of 22 days (range, 0–108 days) after RIC and 31 days (range, 17–64 days) after MAC. Sixty-one of 97 recipients received hematopoietic growth factor support, with no significant faster neutrophil recovery ($p = 0.92$).

The cumulative incidences of platelet recovery at days 100 and 180 after transplantation were 54% (95% CI, 50–58)

Table 2. Umbilical cord blood properties and matching variables according to conditioning

	All patients (n = 136)	MAC (n = 46)	RIC (n = 90)
Stem cell count-median (range)			
At harvesting			
$TNC^{-} \times 10^{7}$/kg bodyweight	4.6 (1.6–12.2)	4.6 (2.5–9.1)	4.5 (1.6–12.2)
$CD34^{-} \times 10^{5}$/kg bodyweight	1.8 (0.3–10.9)	2.2 (0.8–10.9)	1.6 (0.3–7.8)
Infused			
$TNC^{-} \times 10^{7}$/kg bodyweight	3.1 (0.6–12.1)	3.2 (1.2–12.1)	3 (0.6–10.9)
$CD34^{-} \times 10^{5}$/kg bodyweight	1.4 (0.2–9.3)	1.6 (0.5–9.3)	1.1 (0.2–4.8)
HLA matching			
Group 1	77 (56.5%)	28 (61%)	49 (54%)
Group 2	51 (37.5%)	16 (35%)	35 (39%)
Group 3	8 (6%)	2 (4%)	6 (7%)
Sex matching			
(MM) → M recipient	25 (18.5%)	9 (19%)	16 (18%)
(FF) → F recipient	17 (12.5%)	6 (13%)	11 (12%)
(MF) → M recipient	45 (33%)	16 (35%)	29 (32%)
(MF) → F recipient	19 (14%)	5 (11%)	14 (16%)
(FF) → M recipient	18 (13%)	5 (11%)	13 (14%)
(MM) → F recipient	12 (9%)	5 (11%)	7 (8%)
ABO compatibility according to recipient			
Complete compatibility	37 (27%)	14 (30%)	23 (25%)
1 incompatibility	49 (36%)	16 (35%)	33 (37%)
Major	28 (21%)	7 (15%)	21 (23.5%)
Minor	21 (15%)	9 (20%)	12 (13.5%)
2 incompatibilities	50 (37%)	16 (35%)	34 (38%)
2 major	21 (15%)	4 (9%)	17 (19%)
2 minor	17 (13%)	5 (11%)	12 (13.5%)
1 major and 1 minor	12 (9%)	7 (15%)	5 (5.5%)
CMV serologic status			
Seronegative recipients	77 (57%)	31 (67%)	46 (51%)

CD34 = $CD34^{+}$ cells; HLA = human leukocyte antigen; MAC = myeloablative conditioning; RIC = reduced-intensity conditioning; TNC = total nucleated cells.

and 60% (95% CI, 56–64), respectively (Fig. 1B), with a median time of 54 days (range, 0–449 days) after RIC and 56 days (range, 19–133 days) after MAC.

During the follow-up, we observed an autologous reconstitution in seven patients (six after RIC and one after MAC), and two patients (both after RIC) developed a secondary graft failure (at 2 and 6 months after transplantation).

Chimerism results and dominant UCB unit identification

Chimerism analysis was evaluated in 97 patients; it was not performed in 6 patients, and 15 patients were not evaluable for chimerism, among which were 6 early deaths. The median follow-up for chimerism analysis was 9 months (range, 1–44 months). We found a predominance of cells coming from one UCB unit, thus a dominant UCB in 85 (88%) patients. A persistent mixed chimerism after at least 2 months after transplant was observed in 12 (12%) patients.

In case of a dominant UCB unit, we did not find any difference in $CD34^{+}$ and TNC counts between the two units ($p = 0.18$ and $p = 0.11$, respectively). Concerning hematopoietic recovery, its kinetics in the dominant UCB was influenced by kind of conditioning; it was faster after MAC than after RIC ($p < 0.0001$). Next, we analyzed factors potentially associated with the dominant UCB. We performed a multivariate analysis including HLA, sex, and ABO matching and cellularity (TNC and CD34 positive cells infused). There was a trend in the association of sex matching with the predominant unit ($p = 0.094$); the unit that is sex matched with the recipient has a trend of higher probability to become dominant after transplantation.

Acute and chronic GVHD

At day 100 after transplantation, we observed 28 grade I, 27 grade II, 23 grade III, and 6 grade IV acute GVHD cases. The cumulative incidence of grade II or greater acute GVHD was 36% (range, 32–40%), with 20% (range, 17–24) being grades III and IV. GVHD was resolved in 65% of cases.

Among 104 evaluable patients, we observed 12 limited and 14 extensive chronic GVHD cases, and the cumulative incidences of chronic GVHD at 1 and 2 years were 19.5% (range, 15.3–23.9%) and 23% (18.4–27.5%), respectively.

Overall and progression-free survival

After a median follow up of 49.5 months (range, 3–72 months) for surviving patients (last follow-up in May

Table 3. Hematopoietic stem cell transplantation procedures

Procedures	No. of patients (%)
Conditioning regimen	
Myeloablative conditioning	46
Cyclophosphamide, TBI	35 (76%)
Cyclophosphamide, busulfan	8 (17%)
Others	3 (7%)
Reduced intensity conditioning	90
Cyclophosphamide, fludarabine, TBI (Minnesota)	75 (83%)
Melphalan, fludarabine, busulfan	5 (6%)
Cyclophosphamide, fludarabine, busulfan	2 (2%)
Others	8 (9%)
GVHD prophylaxis	
Cyclosporine A, MMF	105 (77%)
Cyclosporine A, steroids	17 (12.5%)
Cyclosporine A, methotrexate	5 (4%)
Cyclosporine A, methotrexate, MMF	3 (2%)
Cyclosporine A alone	3 (2%)
MMF ± steroids	1 (1%)
No prophylaxis	2 (1.5%)
Antithymocyte globulin	34 (25%)

ATG = antithymocyte globulin; MMF = mycophenolate mofetil; TBI = total body irradiation.

2012), the probabilities of 3-year OS and PFS were 41.3% (95% CI, 33.6–50.6) and 34.6% (95% CI, 27.6–44.2), respectively, and the median OS was 17.5 months (95% CI, 9.6–52.6; Fig. 2A and D). We observed better results for patients in CR1 with a 3-year OS of 59.4% (95% CI, 46.1–76.4) versus 33.3% (95% CI, 25.0–44.6) for patients not in CR1 at transplant ($p = 0.0085$; Fig. 2B).

Relapse incidence

Relapse occurred in 45 patients after a median time of 5.5 months (range, 1–50) after transplantation. The 1- and 2-year cumulative incidences of relapse were 21.3% (CI, 17.8–24.8) and 28% (CI, 24.0–31.8), respectively. Three years after transplantation, we observed only three late relapses (one patient with Hodgkin lymphoma in the third line of treatment with grade I acute GVHD of the skin and no chronic GVHD, one patient with multiple myeloma in VGPR at transplantation after three lines of treatment without any acute or chronic GVHD, and one patient with acute lymphoid leukemia in CR1 at transplant without acute or chronic GVHD).

Treatment-related mortality and causes of death

The 3-month and 2-year cumulative incidences of TRM were 12.5% (CI, 9.7–15.3) and 27% (CI, 23.4–31.0), respectively.

At the last follow-up, 54 (40%) patients were alive and 81 (60%) patients had died. One patient was lost to follow-up 4 months after transplantation. Thirty-six patients died from relapse, 44 patients died from HSCT-related causes (23 from infection, 12 from GVHD, 1 from lymphoproliferative disorder, 8 from toxicity, and 1 from secondary malignancy [acute myeloid leukemia]). Considering engraftment, 7 of 9 patients with primary graft failure died (3 from disease progression and 4 from HSCT-related causes), the 2 remaining patients are still alive. The 2 patients with secondary graft failure died from progression. Among the 7 patients with autologous reconstitution, 5 died (2 disease progressions, and 3 HSCT related causes) and 2 are alive. Finally, regarding chimerism, 10 of 12 patients with mixed chimerism died (4 relapses and 6 HSCT-related causes), and the 2 remaining patients were still alive 36 and 64 months after transplant.

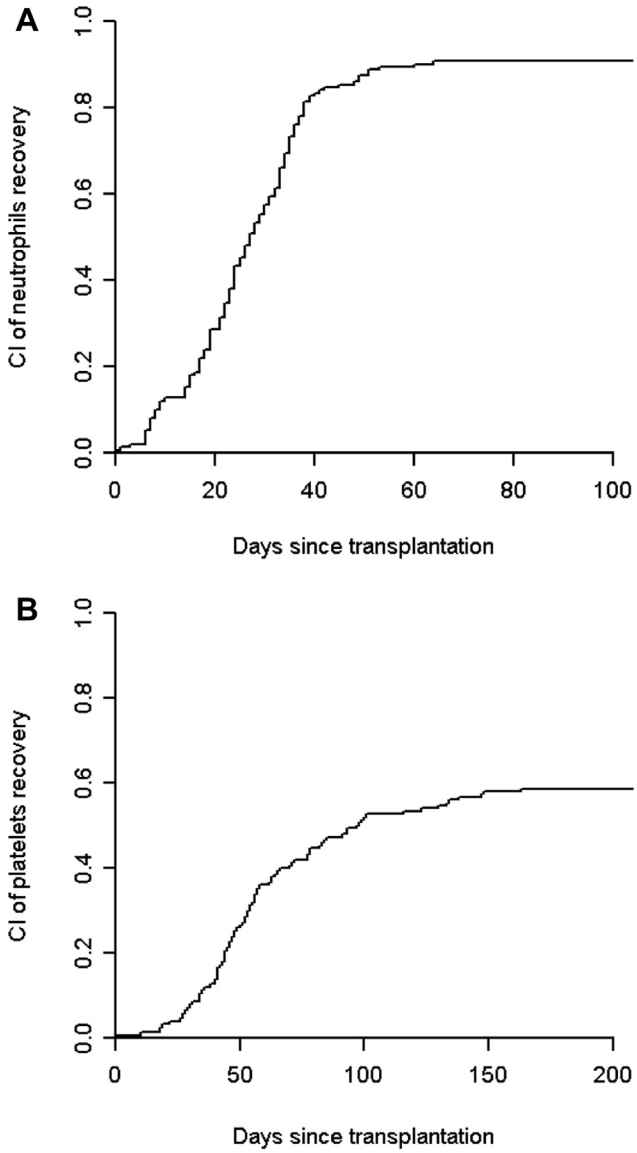

Figure 1. Hematopoietic recovery: cumulative incidence of neutrophil recovery **(A)** and platelet recovery **(B)**.

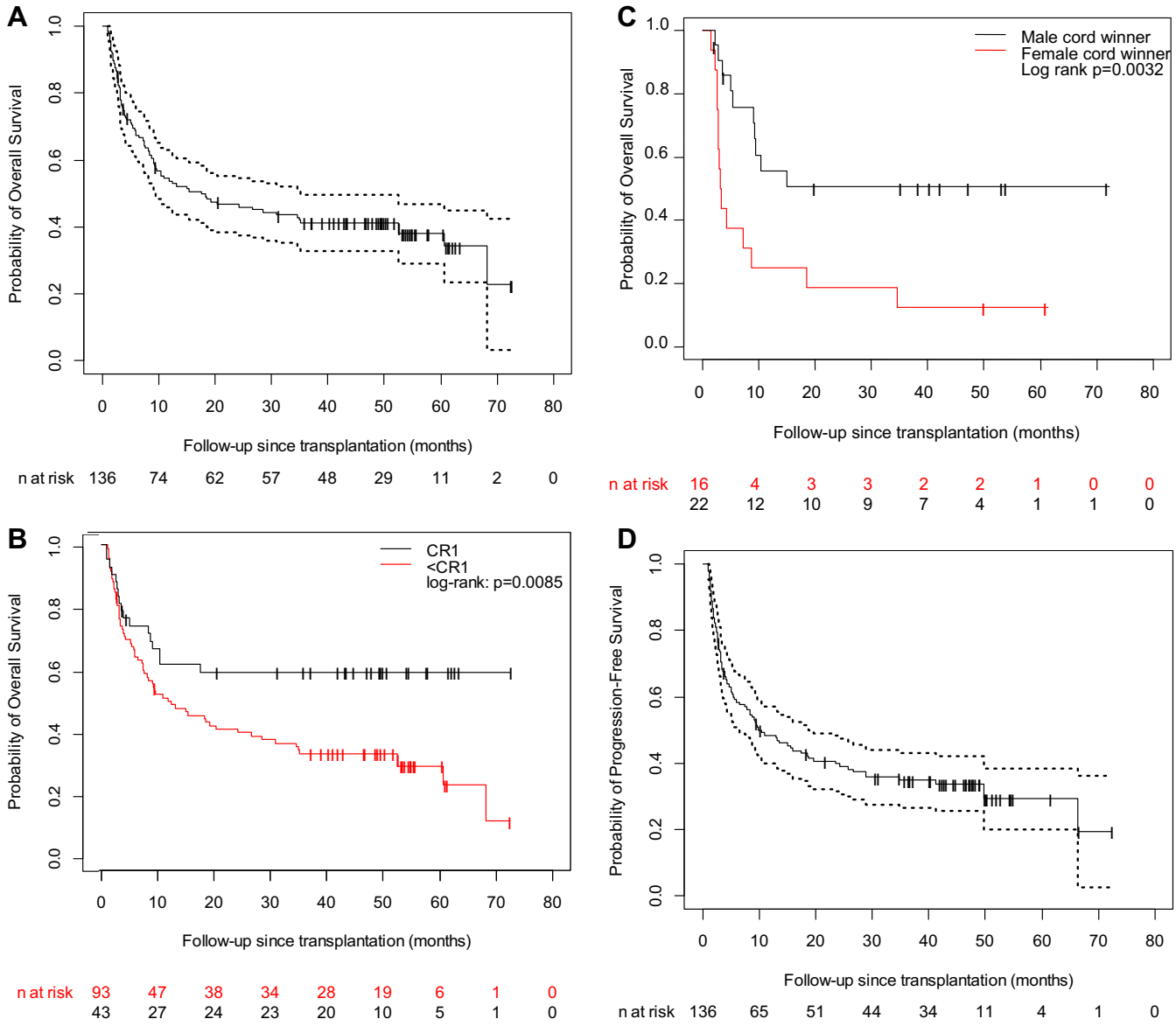

Figure 2. Probability of overall survival—**(A)** of the global population, **(B)** according to status at time of transplantation, **(C)** according to the sex of the dominant UCB unit among male recipients—and progression-free survival **(D)**. Dashed lines represent 95% confidence interval. (Color version of figure is available online.)

Statistical results

Results of multivariate analysis concerning engraftment, acute and chronic GVHD, relapse, TRM, OS, and PFS are shown in Table 4. For sex matching, we found a lower probability of OS in male recipients transplanted with both male and a female UCB units in comparison with male recipients transplanted with two male UCB units (p = 0.043). In this subgroup, there was a trend for a lower probability of PFS (p = 0.07) and for a higher incidence of relapse (p = 0.059) with a lower incidence of chronic GVHD (p = 0.09). In addition, we showed a significant OS advantage when male cord engrafted among male recipients (p = 0.0032; Fig. 2C). This result was not significant among female patients because the small sample size and data available (n = 19).

Discussion

The first results published for dUCBT settings have demonstrated the feasibility and safety of this kind of allogeneic HSCT for patients without any donor or when one UCB unit had insufficient cellularity [13,21,29]. Other studies including both single and double UCB showed a better engraftment with less severe GVHD, a low TRM, and less relapse after dUCBT [14,15,30–32], with an advantage in terms of PFS and OS. More recently, Rocha et al. [33] compared 230 dUCBT with 377 single UCBT from the Eurocord Registry and confirmed the previous results with lower relapse and improved leukemia-free survival (LFS) rates after dUCBT compared with single UCBT for patients transplanted in CR1. Brunstein et al. [34] showed similar leukemia-free survival after dUCBT when they were

Table 4. Significant factors after multivariate analyses for hematopoietic recovery, GVHD, TRM, relapse, OS, and PFS in the global population

Outcome	Variable	HR (95% CI)	*p* value
Neutrophil recovery	Disease status (vs. CR1)		
	PR	0.34 (0.15–0.75)	0.008
	<PR	0.61 (0.37–1.02)	0.057
	ABO matching (vs. matched)		
	1 major incompatibility	0.57 (0.35–0.93)	0.024
Platelet recovery	Disease status (vs CR1)		
	PR	0.28 (0.08–0.92)	0.036
	<PR	0.52 (0.29–0.95)	0.032
Acute GVHD	Disease status (vs. CR1)		
	<PR	0.37 (0.16–0.87)	0.02
	Sex matching (vs. matched)		
	MF→ F	0.34 (0.12–0.95)	0.04
Chronic GVHD	Sex matching (vs. MM → M)		
	FF→ M	0.14 (0.05–0.38)	0.049
	MF→ F	0.11 (0.04–0.26)	0.013
	MM → F	0.21 (0.10–0.44)	0.032
Transplant-related mortality	CMV-positive recipients	3.45 (2.08–5.71)	0.014
	ABO matching (vs. matched)		
	1 minor incompatibility	0.10 (0.06–0.18)	<0.001
	1 minor and 1 major incompatibility	0.09 (0.04–0.20)	0.004
	Serotherapy (vs. none)	4.36 (2.59–7.35)	0.005
Relapse	Disease status (vs CR1)		
	PR	21.27 (9.35–48.42)	<0.001
	Other than CR1	4.87 (2.71–8374)	0.007
	ABO matching (vs. matched)		
	1 major incompatibility	5.28 (2.99–9.30)	0.003
	Conditioning regimen: MAC	0.31 (0.17–0.54)	0.035
Overall survival	Disease status (vs. CR1)		
	PR	4.26 (1.31–13.88)	0.016
	CMV-positive recipients	2.44 (1.36–4.38)	0.003
	Sex matching (vs. MM → M)		
	MF→ M	2.26 (1.03–4.98)	0.043
	ABO matching (vs. matched)		
	1 major incompatibility	3.29 (1.44–7.56)	0.005
	1 major and 1 minor incompatibility	0.23 (0.07–0.70)	0.01
	Serotherapy (vs. none)	2.72 (1.47–5.01)	0.001
Progression-free survival	Disease status (vs. CR1)		
	PR	4.54 (1.57–13.12)	0.005
	CMV-positive recipients	2.32 (1.35–3.99)	0.002
	ABO matching (vs. matched)		
	1 major incompatibility	2.49 (1.15–5.41)	0.020
	1 major and 1 minor incompatibility	0.35 (0.13–0.97)	0.044
	Serotherapy (vs. none)	1.98 (1.14–3.44)	0.015

CI = confidence interval; CMV = cytomegalovirus; CR = complete remission; F = female; GVHD = graft versus host disease; HLA, human leukocyte antigen; HR = hazard ratio; M = male; MAC = myeloablative conditioning; PR = partial remission.

compared with allogeneic HSCT from HLA-matched related or unrelated donors. Similarly, Ponce et al. [35] showed earlier TRM after dUCBT when compared to transplantations from related and unrelated donors but with a lower long-term TRM and less relapse. Recently, a comparison of dUCBT with HSCT from HLA-haploidentical related donors in a phase 2 trial showed encouraging results in favor of dUCB units as an alternative source of hematopoietic stem cells [36].

Our study includes a large number of patients with the largest follow-up (median, 49.5 months; range, 2.7–72) for surviving patients after dUCBT compared with previous studies. A supplementary advantage in our study was the complete documentation of many important pretransplantation and posttransplantation parameters, mainly the cellularity, HLA data, and chimerism monitoring. Concerning hematopoietic recovery, our results are similar to those published in previous studies after MAC [29,30,32] or RIC [14,15]. Nevertheless, we did not show any effect on engraftment of HLA matching and of TNC and $CD34^+$ cell numbers, which can be related to the high median dose of infused cells observed in the dUCB according to recent recommendations [16,19] and well-HLA matched conditions in the majority of the cases. Moreover in our

study, 32.5% of patients had a sufficient number of cells provided by only one UCB unit, for which a single UCBT could have been feasible. In this setting, a recent article on dUCBT showed a significant influence of TNC and $CD3^+$ cell doses on engraftment, but no influence of HLA matching [18]. In addition, we found a significant effect of disease status and ABO compatibility on neutrophil recovery.

The incidence of acute GVHD is in the range of results already described [15,37]. A higher and earlier incidence of acute GVHD without any effect on TRM has been published by MacMillan et al. [31] and Brunstein et al. [38]. We did not find any effect of pretransplantation factors such as RIC and ATG on acute GVHD incidence, as reported by MacMillan et al. [31]. Our data regarding chronic GVHD are in accordance with previous studies [15,29–32,38].

Regarding other transplant outcomes, disease status at the time of transplantation remains a major factor affecting overall, progression-free survival, and relapse as described previously [30,32]. Although an important number of patients included in our study were heavily pretreated and 20% of patients were not in complete remission at the time of transplantation, we found encouraging overall results especially patients transplanted in CR1 with a stable plateau of long-term survival. TRM was acceptable compared to available results [15,30–32], and the main cause of death was infection. In terms of relapse, as opposed to Verneris et al. [30], we found an effect of conditioning regimen on incidence, which is lower after MAC [30]. According to CMV status of the recipient, we found a significant negative effect on TRM, OS, and PFS in seropositive recipients. Regarding HLA matching, we did not show any difference between groups 1 and 2 except for relapse, with a trend in favor of the second group. These preliminary findings need to be considered regarding the probably underestimated number of HLA-A and HLA-B mismatches at the generic level. HLA class I and II allele level determination as well as KIR typing could improve the UCB choice and thus transplantation outcome [39,40].

The documentation of chimerism allowed us to show the dominance of one UCB in the majority of cases as demonstrated previously [14,15,29,32,41]. Similar to other reports, there were only a few patients with a stable mixed donor (double UCB) chimerism [42–44]. The donor chimerism status was determined faster after MAC than after RIC, as reported previously [15]. The presence of a dominant UCB raised a certain number of questions about factors implicated in its determination [45]. Different hypotheses were previously proposed, such as the $CD3^+$ cell number [18,29,46], the order of UCB injection [15], or the $CD34^+$ cell viability [18,47]. The effect of HLA matching on the dominant UCB unit determination remains controversial [18,46]. More recently, in a biologic approach, some authors showed the implication of an immunologic $CD8^+$ T cell–mediated reaction from the dominant unit against the other [23], and the hypothesis of a graft-versus-graft reaction is supported by in vitro data [48] with a potential role of $CD34^-$ cells [49]. In our study, a statistical model accounting for all transplantation variables raises the hypothesis of a potential role of sex matching in the determination of the dominant UCB unit. Moreover, among patients transplanted with one female and one male unit, we showed a significant OS advantage when male cord engrafted among male recipients. This factor has never been considered in previous analyses and needs to be confirmed in larger series to understand the interfering factors independently of cell number and HLA matching.

In conclusion, our findings show encouraging long-term results after dUCBT, although only 30% of patients were in early disease phase at the time of transplantation; disease status before transplantation remains the major prognostic factor for transplantation outcome. We obtained a long-term plateau among patients stratified on disease status (CR1), which makes dUCBT a promising treatment strategy for these patients. The role of the dominant UCB on transplant outcome is still not well defined. We showed a possible role of sex matching on the presence of dominant UCB with a significant advantage in terms of OS when male cord engrafted among male recipients.

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Conflict of interest disclosure

No financial interest/relationships with financial interest relating to the topic of this article have been declared.

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