



Single-Unit versus Double-Unit Umbilical Cord Blood Transplantation in Children and Young Adults with Residual Leukemic Disease

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COVER LETTER

Marseilles, July the 17th, 2018

Dear Editor,

We are pleased to submit for publication in Biology of Blood and Marrow Transplantation our study “Single vs double-unit cord-blood transplantation in children and young adults with residual leukemic disease.”

In 2016, we report the results of a French prospective and randomized study comparing “single vs double-unit cord blood transplantation for children and young adults with acute leukemia or myelodysplastic syndrome”. Similarly to the prospective study conducted in the US on the same subject, the double-unit strategy proved ineffective at improving the overall outcome when a single cord had an adequate cell dose. However, the effect on post-transplant alloreactivity (i.e. graft vs host and graft vs leukemia effect) was unclear and none of the 2 studies has taken into account in their analysis the pre-transplant minimal residual disease (MRD), a major prognostic factor in the context of non-cord transplantation for acute leukemia.

The purpose of this additional report, based on data from the French prospective study, was to investigate the influence of pre-transplant MRD on relapse and survival after single or double-unit cord transplantation. The key points are that 1/ unrelated cord blood transplantation offers high survival rates even in patients with positive pre-transplant MRD and that 2/ double-cord could have a better graft vs leukemia effect than single-cord transplant in these MRD+ patients.

We think that these new results may be of interest for hematologists and decided to submit this additional study to Biology of Blood and Marrow Transplantation, as a regular manuscript. Please note that we intend to email the signed Author Agreement Form.

Thank you for giving us this opportunity.

Yours sincerely

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UCBT offers high survival rates in children and young adults with acute leukemia.

Even with positive pre-transplant MRD, UCBT leads to high cure rate.

Double UCBT could have a better graft versus leukemia effect in positive MRD patients.

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ABSTRACT

We previously reported in a French prospective randomized study that transplantation of 2 unrelated cord blood (UCB) units instead of 1 does not decrease the risk of transplantation failure but may enhance alloreactivity. We present here the influence of pre-transplant minimal residual disease (MRD) on leukemia relapse and survival after single *versus* double-UCB transplantation. Among 137 children and young adults who were transplanted in the randomized study, 115 had available MRD assessment immediately before their conditioning regimen. MRD was considered positive when $\geq 10^{-4}$, which was the case of 43 out of 115 patients. Overall, the 3-year survival probability was $69.1 \pm 4.4\%$ and it was not significantly influenced by the MRD level: $70.7 \pm 5.4\%$ in MRD- ($<10^{-4}$) patients (n=72), $71.1 \pm 9.4\%$ in MRD+ with $10^{-4} \leq \text{MRD} < 10^{-3}$ (n=26), and $58.8 \pm 11.9\%$ in MRD+ $\geq 10^{-3}$ patients (n=17). In the MRD+ group, we found a significantly lower risk of relapse in the double- *versus* single-unit arm ($10.5 \pm 7.2\%$ vs $41.7 \pm 10.4\%$; $p=0.025$) leading to a higher 3-year survival rate ($82.6 \pm 9.3\%$ vs $53.6 \pm 10.3\%$, $p=0.031$). This difference was only observed in patients who had not received anti-thymocyte globulin (ATG) during their conditioning regimen. In the MRD- group, no difference was found between the single- and the double-unit arms. We conclude that, even in case of positive pre-transplant MRD, UCB transplantation in children and young adults with acute leukemia results in a high cure rate and that a double-unit strategy may enhance graft-vs-leukemia effect and survival in these patients.

INTRODUCTION

Cryopreserved unrelated cord blood (UCB) can be used as a stem cell source for patients with acute leukemia (AL) who need hematopoietic stem cell transplantation (HSCT) and lack an HLA-identical donor.¹ Historically, it was hypothesized that the greater the number of hematopoietic stem cells in UCB units the better the outcome.² In this context, some retrospective analysis suggested the relapse rate was lower after double- compared with single-unit transplantation.³⁻⁵ However, in two prospective randomized studies conducted in the US and in France, the double-unit strategy proved ineffective at improving the overall outcome of an UCB transplant in children and young adults with AL when a single cord had an adequate cell dose.^{6,7} Although graft-versus-host disease (GvHD) was more frequent after double-unit transplantation in both studies, no enhanced graft-versus-leukemia (GvL) effect could be observed in the US study, and in the French study the GvL effect was strictly limited to patients who hadn't received any anti-thymocyte globulin (ATG) in their conditioning regimen. Noteworthy, none of these two studies has analyzed the results of randomization according to pre-transplant minimal residual leukemia disease (MRD).

The presence of MRD before transplantation is a major prognostic factor in the context of non-cord HSCT for acute leukemia.⁸⁻¹⁴ Interestingly, a recently published large retrospective and single center study reported that among patients with positive pre-transplantation MRD, the probability of relapse was lower after UCB than after HLA-matched or HLA mismatched unrelated donor transplantation. In the cord blood group, which included a large majority of patients receiving a double-unit, the overall survival and relapse rate were similar among patients with MRD and those without MRD.¹⁵

We present here the results of the French “single- *versus* double-unit” prospective and randomized study according to the pre-transplant MRD. The main purposes of this additional report were 1/ to examine if the promising results of UCB transplantation, retrospectively reported in a single center study for patients with positive MRD, are reproduced in our prospective and multicenter study, and

2/ to investigate a potentially different effect of the single- *versus* double-unit randomization according to pre-transplant MRD.

METHODS

Inclusion criteria

In the prospective and randomized “single vs double-unit” French study, patients were eligible if they met all of the following criteria: (1) age < 35 years, (2) acute leukemia in complete remission or myelodysplastic syndrome (MDS) with < 20% bone marrow blasts, (3) requiring unrelated HSCT, and (4) absence of unrelated donor considered acceptable on the basis of HLA compatibility and donor availability. In addition, eligible patients had to have at least two unrelated cord blood (UCB) units which were 4-6 HLA identical to the patient and between them, and which contained $> 3 \times 10^7$ total nucleated cells (TNCs) per kilogram of recipient body weight for the first UCB unit, and $> 1.5 \times 10^7$ TNCs/kg for the second. The children and young adults with acute leukemia who were enrolled in this study and had assessment of their MRD immediately before their conditioning regimen were included in the present report. This randomized study was approved by the French National Program for Clinical Research and the French National Cancer Institute and was registered at www.clinicaltrials.gov as #NCT01067300. All patients (or their parents or legal guardians) provided written informed consent.

Method for MRD quantification and stratification

Methods for MRD quantification differed according to primary disease and to the transplant center’s practice. In patients with acute lymphoblastic leukemia (ALL), most pre-transplant MRD measurements were performed on bone marrow aspirates by using RQ-PCR of T cell receptor (TCR) and IgH gene rearrangements, although multi-color multi-parameter flow cytometry assessment was also used in 4 centers. In patients with AML, MRD was evaluated by PCR of leukemia-specific fusion transcripts or flow cytometry if available, or by Wilms Tumor Gene (WT1) expression, using the standardized European LeukemiaNet (ELN) method.¹⁶

The MRD level was considered positive when $\geq 0.01\%$ (1 leukemic cell among 10,000 BM mononucleated cells) and negative when undetectable or $< 0.01\%$, for RQ-PCR of TCR and IgH gene rearrangements, leukemia-specific fusion transcripts and flow cytometry. WT1 expression was considered a positive MRD when > 0.1 WT1 RNA copies for 100 abl in peripheral blood or > 2 for 100 abl in bone marrow.

Transplantation procedures

The detailed transplantation procedures have been previously described.⁷ Briefly, all patients received a myeloablative conditioning regimen with either busulfan, cyclophosphamide and anti-thymocyte globulin (BuCyATG) if they were less than 4-year-old or fludarabine, 12 grays total body irradiation (TBI) and cyclophosphamide (FluTBICy) if older. BuCyATG was mandatory for children under 4-years of age and could also be used for older patients with AML, according to the policy of each transplant center. Donor-recipient HLA matching was assessed at low/intermediate resolution level molecular typing for HLA-A and -B loci and high resolution genotyping for HLA-DRB1 locus. HLA compatibility was expressed as the number of identical loci out of 6. All selected UCB had to fulfill the eligibility criteria detailed above. The GVHD prophylaxis consisted in Cyclosporine A and steroids after BuCyATG, and Cyclosporine A and Mycophenolate Mofetil after FluTBICy.

Study endpoints and statistical methods

The main outcomes investigated here were relapse risk, disease-free survival (DFS) at 3 years and survival (OS). Relapse rates were calculated using the cumulative incidence function with non-relapse mortality as competing risk.¹⁷ Comparisons were made using the Fine and Gray model.¹⁸ The probabilities of DFS and OS were calculated using the Kaplan-Meier estimator and compared with the log-rank test.¹⁹ For this analysis, we used February 2017 as the reference date (ie, the month patients' outcomes were locked). All probabilities were given at 3 years and provided with their 95% confidence interval.

RESULTS

The study cohort

The study flow chart is presented in Figure 1. Among 137 patients who were transplanted in the single- *versus* double-UCB study, 115 had acute leukemia and adequate MRD assessment before their conditioning regimen (79 out of 80 ALL and 36 out of 57 AML). All were included in the analysis presented here and represent the “study cohort”. The remaining 22 patients who lacked adequate pre-transplant MRD data had MDS in 5 cases, AML in 16 and ALL in 1. Seventy four out of 115 patients received FluTBICy without ATG as conditioning regimen (64 ALL and 10 AML) whereas 41 received BuCyATG (26 AML and 15 ALL). Fifty-six patients out of 115 were in the single UCB arm *versus* 59 in the double UCB arm.

Pre-transplant MRD groups

Pre-transplant MRD was estimated molecularly in 72/79 ALL cases, and by flow cytometry in the remaining 7. In AML patients, pre-transplant MRD was performed using leukemia specific fusion transcript in 27 cases, WT1 expression in 5, and flow cytometry in 4. Forty-three patients had positive pre-transplant MRD (MRD+): 4 with MRD load $\geq 10^{-2}$; 13 with MRD load $\geq 10^{-3}$ and $< 10^{-2}$; and 26 with MRD load $\geq 10^{-4}$ and $< 10^{-3}$. The 72 other patients were considered negative (MRD-): 58 had undetectable pre-transplant MRD and 14 had detectable but $< 10^{-4}$ MRD load. As shown in Table 1, the two MRD groups were well balanced according to gender, age at transplant, diagnosis, hematological pre-transplant status, conditioning regimen, transplanted cell dose, graft HLA-compatibility and allocation to the single or the double-unit arm of the randomized study.

Overall results according to pre-transplant MRD quantification

The 3-year survival probability was $69.1 \pm 4.4\%$ for the 115 patients of the whole study cohort (it was $69.7 \pm 4\%$ for all 137 patients transplanted in the single- vs double study). We did not detect any significant influence of pre-transplant MRD status on post-transplant outcome for the whole study cohort (Figure 2). The 3-year overall survival was $70.7 \pm 5.4\%$ in MRD- ($<10^{-4}$) patients, $71.1 \pm 9.4\%$ in MRD+ between $<10^{-3}$ and $\geq 10^{-4}$, and $58.8 \pm 11.9\%$ in MRD+ $\geq 10^{-3}$ patients, $p=0.593$. Although a non-significant trend for a better 3-year overall survival in MRD- than in MRD+ patients was observed in the single-unit arm ($78.1 \pm 7.3\%$ vs $53.6 \pm 10.3\%$; $p=0.071$), this was not at all detectable in the double-unit arm ($64.8 \pm 7.6\%$ vs $82.6 \pm 9.3\%$; $p=0.113$) (Figure 3A and 3B).

Results of the single- vs double-unit randomization in MRD+ patients (Figure 4A and 4B, Table2)

In the MRD+ group, we found a significantly higher 3-year overall survival after double-unit than after single-unit UCB transplant ($82.6 \pm 9.3\%$ vs $53.6 \pm 10.3\%$; $p=0.031$). This statistical difference was also found for the 3-year DFS ($82.6 \pm 9.3\%$ vs $53 \pm 10.5\%$; $p=0.028$) and the 3-year relapse rate ($10.5 \pm 7.2\%$ vs $41.7 \pm 10.4\%$; $p=0.025$).

As detailed in table 2, these differences were particularly marked in the subgroup of pre-transplant MRD+ patients who received FluTBICy as conditioning regimen and cyclosporine-mycophenolate without ATG as GvHD prophylaxis: in this subgroup, the 3-year overall survival reached $91.7 \pm 8\%$ after double-unit vs $57.8 \pm 12.2\%$ after single-unit transplantation, $p=0.027$.

Conversely, no statistical difference between the two UCB arms was found in the BuCyATG/MRD+ subgroup who received cyclosporine A, steroids and pre-transplant ATG for GvHD prophylaxis.

We also analyzed the results of randomization in MRD+ patients according to their diagnosis (supplementary Table): we found significantly higher OS, DFS, and lower relapse rate in the double-unit UCB transplant arm than in the single-unit one for MRD+ patients with ALL whereas there was no difference for MRD+ AML patients.

Results of the single- vs double-unit randomization in MRD- patients (Figure 4C and 4D, Table2)

In the MRD- group, there was no difference between the two UCB arms in terms of risk of relapse, DFS, or survival, for the entire group as well as for both conditioning regimen subgroups.

DISCUSSION

This additional analysis to the French prospective “single vs double-UCBT” trial shows that UCB transplantation offers a good chance of cure for children and young adults with acute leukemia who need HSCT, even in case of positive pre-transplant MRD. The 3-year overall survival, which was $69.1 \pm 4.4\%$ for the whole cohort, remained at $71.1 \pm 9.4\%$ when the MRD level was positive $\geq 10^{-4}$ but $< 10^{-3}$, and at $58.8 \pm 11.9\%$ for patients with an MRD+ exceeding 10^{-3} . These results confirm, in a prospective and multicenter cohort, those reported by Milano et al.¹⁵ Overall, their 4-year survival was 71% after UCB for acute leukemia and myelodysplastic syndrome, without any significant difference between patients with positive and undetectable pre-transplant MRD. However, two other retrospective studies reported less favorable results in patients with ALL who were MRD+ before UCB transplant. In the Eurocord pediatric study²⁰, a positive MRD before single-unit UCB transplant was associated with a significantly higher rate of relapse, a lower DFS and a lower survival (4-year OS was only 41% in MRD+ patients compared to 58% in the MRD- group). Another group also reported a 40% survival in a small retrospective cohort (n=10) of MRD+ children and adults.²¹

Interestingly, our analysis suggests that double UCB may offer a better GvL effect than single UCB in MRD+ patients, resulting in a significantly lower risk of relapse after double-unit transplantation (10.5% vs 41.7%), and leading to a significantly better survival rate (82.6% vs 53.6%). This strong double-UCB transplant-related GvL effect has already been described in several retrospective studies. Verneris et al. found a significantly lower relapse risk after two UCB than after one (19% vs 34%) in 177 patients with ALL or AML.³ Similarly, an EBMT study compared one vs two-unit UCB transplantation in 134 adults with AL.⁴ They found a significantly lower relapse risk and a higher LFS and survival after 2 UCB (21% vs 42% for relapse risk, and 62% vs 42% for 2-year survival). Finally, a

Eurocord analysis reported a lower relapse risk in the double-UCB group than in the single-UCB group (18% vs 26%), among 591 adult patients with AL.⁵

As suggested in the Milano et al. study, alloreactivity after double-UCB could be even higher than after transplant from other stem cell sources.¹⁵ They found a significantly lower risk of relapse after UCB transplantation (the UCB group contained a large majority of double UCB transplant, 89%) than after transplantation from a matched (MUD) or mismatched (MMUD) unrelated donor, when the pre-transplant MRD was positive. This difference was smaller and not statistically significant between the three stem cell source groups (UCB, MUD, and MMUD) in the subgroup of pre-transplant MRD negative patients. Comparing different stem cell sources (matched sibling donor [MSD], MUD, MMUD, or double UCB) in 536 patients with hematological malignancies, Brunstein et al. reported a lower risk of relapse in the double-UCB transplantation group (15%), than in the three other groups (43%, 37% and 35% for the MSD, MUD and MMUD respectively).²² Other retrospective cohorts also showed good and even better results with UCB transplantation compared to other stem cell sources.^{23–25}

In both the US and the French randomized studies that compared single- vs double-UCB, GvHD was more frequent in the double-unit arm^{6,7}, confirming several previous retrospective reports.^{3–5,26} On the other hand, more GvHD has been repeatedly associated with lower risk of relapse after UCBT.^{27,28} Taken together, these data support the hypothesis of a higher alloreactivity with double cord-blood transplants, which may lead to a higher GvL effect, and potentially a lower risk of relapse. In the US randomized study, however, despite a higher incidence of GVHD, no enhanced GvL effect could be observed after double-unit transplantation but it's worth noting that, the reported rates of relapse at 1-year after single- and double-unit UCBT were remarkably low at respectively 12% and 14%. This low relapse risk may be explained by the TBI regimen with a higher total dose in their study than in ours (1320 *versus* 1200 Gray). In any case, the French results presented here suggest that the GvL effect generated by the double transplant could be more easily perceptible in the clinical setting of

positive pre-transplant MRD, which is intrinsically associated with more aggressive diseases than in cases of negative pre-transplant MRD.

Interestingly, the significantly higher GvL effect that we describe after double-UCB was only observed in patients who had received a conditioning regimen with FluTBICy, without pre-transplant ATG. One could argue that ATG may suppress a potentially higher GvL effect of double-unit transplantation. Several studies have underlined, in the setting of UCB transplantation, the deep impact of ATG on immune reconstitution, alloreactivity and clinical outcomes.^{29–34} Zheng et al³³ retrospectively analyzed post-transplant outcome of 207 children with malignant disease who underwent UCB transplant, according to whether or not they had received ATG in their conditioning regimen. They found a higher relapse risk, a higher viral infection rate and a lower DFS in those who had received ATG. Similarly Lindemans et al²⁹ compared three groups (early, late ATG, no ATG) of children who had UCBT for malignant and non-malignant diseases and found a better immune reconstitution, a lower rate of viral infection but a higher rate of severe acute GvHD in the “no ATG” group. Interestingly, Admiral et al³⁰ found worse immune reconstitution, more viral infections, and more relapse in children who had high versus low ATG exposure after UCBT, suggesting that ATG pharmacokinetic could be the way for a better ATG management.^{34–36} The impact of ATG on relapse risk has also been demonstrated in non-cord HSCT.^{31,35,37}

In this study, the strong GvL effect of double-unit transplantation appeared limited to ALL as it wasn't observed in AML. This result must be interpreted with caution because a large majority (86%) of patients who received FluTBICy without ATG had ALL whereas 63% of the BuCyATG group had AML. Other factors, such as HLA disparity and GVH disease, could have played a role. So we cannot conclude that it is a disease-specific effect. Another limitation of the study is that, although based on a prospective and randomized trial, the additional pre-transplant MRD analysis was not initially planned as a study endpoint. Therefore, we did not prospectively define a standard MRD assessment and as a consequence, biological methods to quantify MRD differed according to transplant centers and primary diseases. It should however be noted that MRD assessment was performed in a large

majority of ALL patients by using RQ-PCR of TCR and IgH gene rearrangements according to the ELN recommendations, warranting the standardization of results. On the other hand, the greater heterogeneity in AML may have jeopardized the reliability of our results. Finally, we defined a threshold for MRD positivity at the level $\geq 10^{-4}$ ($\geq 0.01\%$) and this could be subject to controversy. However, this MRD cut-off has been shown to be relevant in several pediatric and young adult cohorts³⁸⁻⁴³ and seems appropriate considering the sensitivity of the different techniques used for MRD detection.^{42,44,45}

We conclude that, even in case of positive pre-transplant MRD, UCB transplantation in children and young adults with acute leukemia leads to a high rate of disease-free survival and that a double-unit strategy may enhance GvL and survival in these patients.

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AUTHORSHIP CONTRIBUTIONS

LB and GM designed research, performed research, analyzed data and wrote the manuscript. CG, JHD designed research, performed research and analyzed data, AL and KB designed research and performed statistical analysis, AS, CR, CP, BB, CJ, IY, EF, CP, AM, DP, VG, DB, FR, CR and MM contributed to research and to writing the manuscript.

CONFLICT OF INTEREST DISCLOSURES

The authors declare no competing financial interests.

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TABLES

Table 1: Patients characteristics according to pre-transplant MRD status

	MRD + (n=43)	MRD - (n=72)	P
Gender			
Male	25 (58.1%)	45 (62.5%)	0.64
Female	18 (41.9%)	27 (37.5%)	
Age			
< 18 years	36 (83.7%)	57 (79.2%)	0.55
≥ 18 years	7 (16.3%)	15 (20.8%)	
Diagnosis			
ALL	30 (69.8%)	49 (68.1%)	0.85
AML	13 (30.2%)	23 (31.9%)	
Hematological status			
CR1	19 (44.2%)	40 (55.6%)	0.24
CR≥2	24 (55.8%)	32 (44.4%)	
Conditionning regimen			
FluTBICy	30 (69.8%)	44 (61.1%)	0.35
BuCYATG	13 (30.2%)	28 (38.9%)	
Number of UCB			
1	24 (55.8%)	32 (44.4%)	0.24
2	19 (44.2%)	40 (55.6%)	
TNC infused (x10⁷/Kg) (mean±SEM)			
1 UCB	4.93 ± 0.66	4.23 ± 0.37	0.36
2 UCB	7.57 ± 0.89	9.36 ± 1.29	0.26

HLA compatibility			
1 UCB :			
4/6	4 (16.7%)	7 (21.9%)	0.74
5-6/6	20 (83.3%)	25 (78.1%)	
2 UCB :			
4/6	6 (31.6%)	14 (35%)	0.80
5-6/6	13 (68.4%)	26 (65%)	

Table 2: Results of randomization according to pre-transplant MRD and conditioning regimen

	Single-unit UCB N=56	Double-unit UCB N=59	P
3-year CI of relapse			
Patients MRD+ (n=43)	41.7 ±10.4%	10.5 ±7.2 %	0.025*
Patients MRD- (n=72)	18.8 ±7%	20 ±6.4%	0.972
FluTBICy MRD+ (n=30)	35.3 ±12.1%	0%	0.019*
FluTBICy MRD- (n=44)	15.8 ±8.6%	12 ±6.7%	0.669
BuCYATG MRD+ (n=13)	57.1 ±21.1%	33.3 ±21.3%	0.291
BuCYATG MRD- (n=28)	23.1 ±12.4%	33.3 ±12.8%	0.570
3-year disease-free survival			
Patients MRD+	53 ±10.5%	82.6 ±9.3%	0.028
Patients MRD-	74 ± 8%	56.8 ± 8%	0.153
FluTBICy MRD+	56.6 ±12.7%	91.7 ±8%	0.025
FluTBICy MRD-	84.2 ±8.4%	59.4 ±9.9%	0.096
BuCYATG MRD+	42.9 ±18.7%	66.7 ±19.2%	0.288
BuCYATG MRD-	59.3 ±14.3%	53.3 ±12.9%	0.721
3-year overall survival			
Patients MRD+	53.6 ±10.3%	82.6 ±9.3%	0.031
Patients MRD-	78.1 ±7.3%	64.8 ±7.6%	0.237
FluTBICy MRD+	57.8 ±12.2%	91.7 ±8%	0.027
FluTBICy MRD-	84.2 ±8.4%	63.5 ±9.7%	0.128
BuCYATG MRD+	42.9 ±18.7%	66.7 ±19.2%	0.367
BuCYATG MRD-	69.2 ±12.8%	66.7 ±12.2%	0.959

FIGURE LEGENDS

Figure 1: Flow chart

Figure 2: Overall results according to pre-transplant MRD status

2A: Three-year disease-free survival

2B: Three-year cumulative Incidence of relapse

2C: Three-year overall survival

Figure 3: Three-year overall survival in MRD + *versus* MRD - patients

3A: After single-unit UCB transplant

3B: After double-unit UCB transplant

Figure 4: Three-year overall survival, relapse, and disease free survival after single-unit or double-unit UCB according to pre-transplant MRD status

4A: Positive pre-transplant MRD patients : overall survival and relapse

4B: Positive pre-transplant MRD patients : disease-free survival

4C: Negative pre-transplant MRD patients : overall survival and relapse

4D: Negative pre-transplant MRD patients : disease-free survival

FIGURES

Figure 1: Flow chart

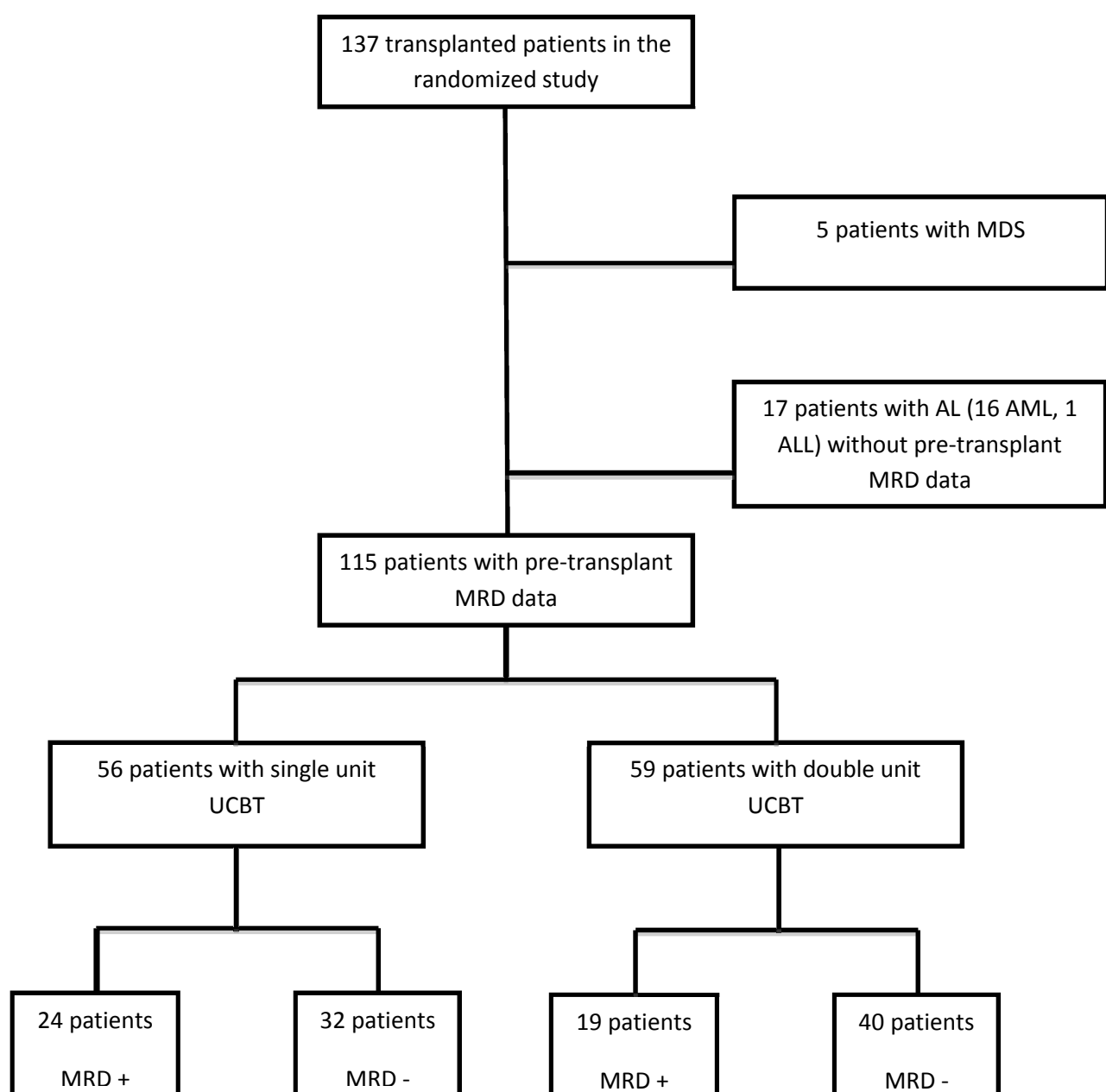
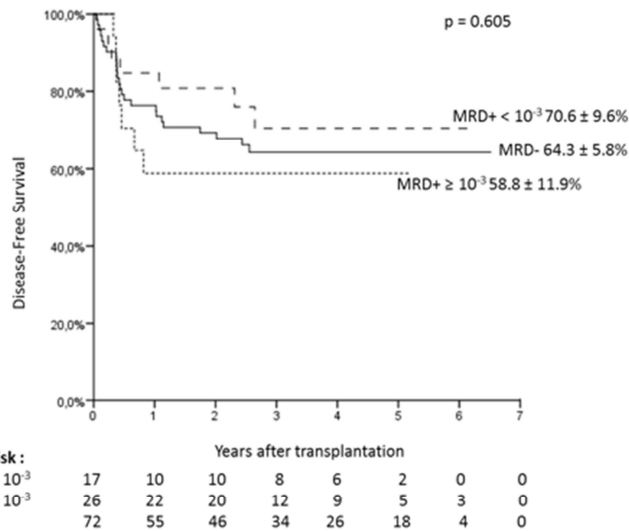
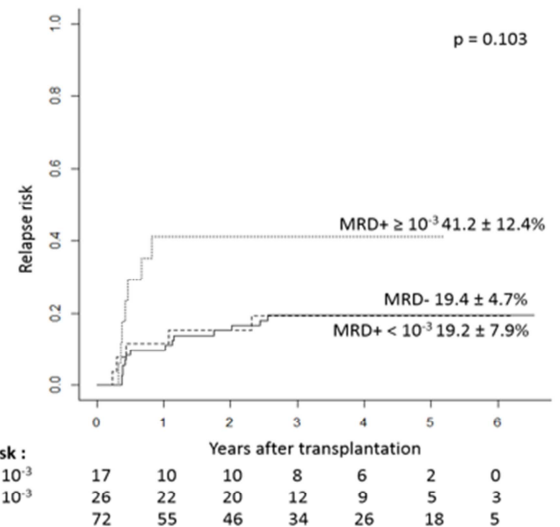


Figure 2: Overall results according to pre-transplant MRD status

2A: Three-year disease-free survival



2B: Three-year cumulative Incidence of relapse



2C: Three-year overall survival

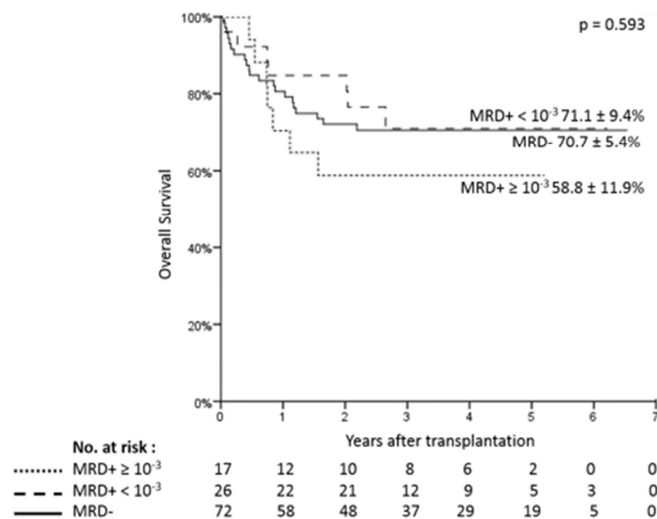


Figure 3: Three-year overall survival in MRD + *versus* MRD - patients

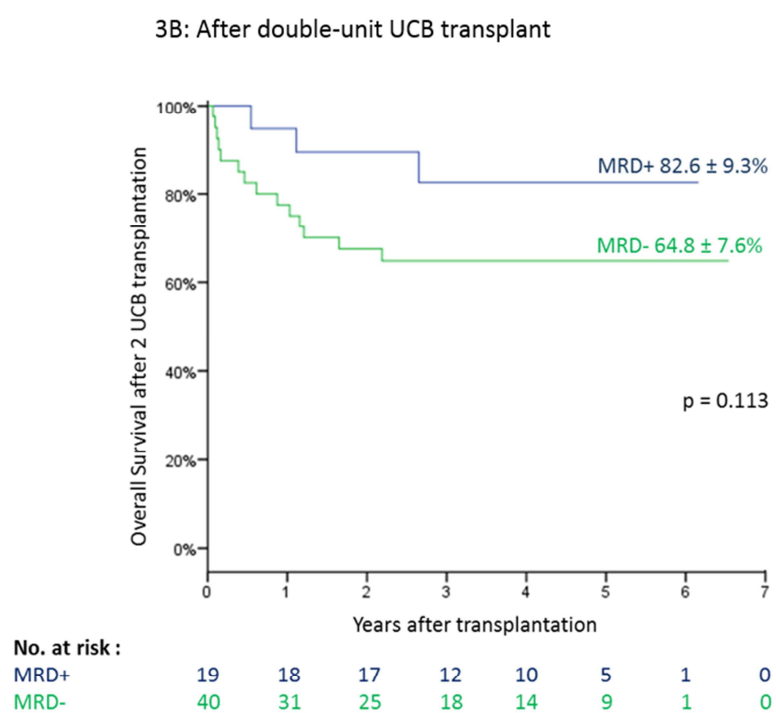
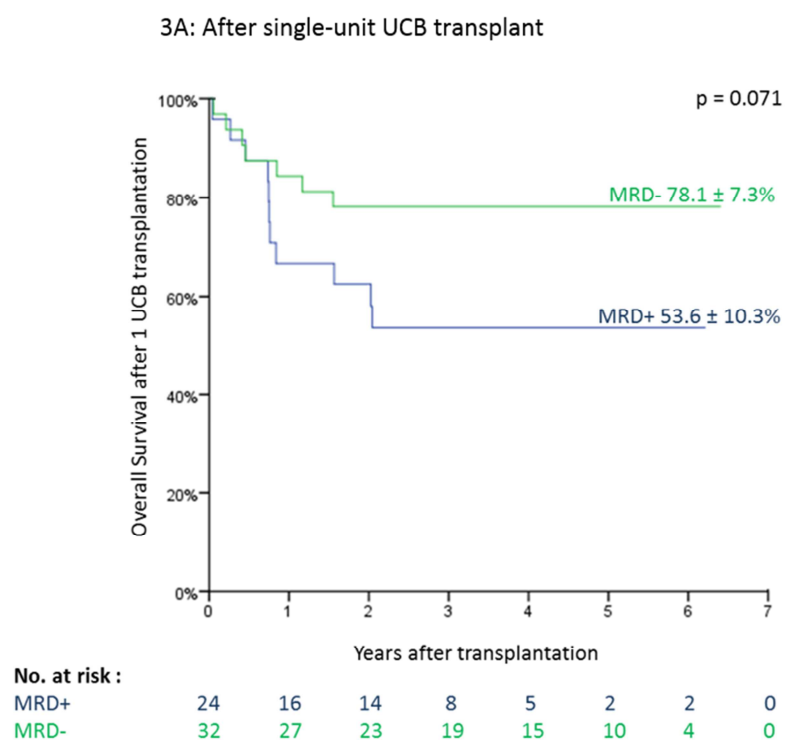


Figure 4: Three-year overall survival, relapse, and disease free survival after single-unit or double-unit UCB according to pre-transplant MRD status

