haematologica
Journal of the Ferrata Storti Foundation

Unrelated cord blood transplantation and post-transplant cyclophosphamide

by Patrizia Chiusolo, Andrea Bacigalupo, Sabrina Giammarco, Elisabetta Metafuni, Federica Sora', Maria Assunta Limongiello, Filippo Frioni, Luca Di Marino, Monica Rossi, Claudio Pellegrino, Caterina Giovanna Valentini, Annalisa Ruggeri, Luciana Teofili and Simona Sica

Received: Janauy 14, 2026.
Accepted: May 19, 2026.

Citation: Patrizia Chiusolo, Andrea Bacigalupo, Sabrina Giammarco, Elisabetta Metafuni, Federica Sora', Maria Assunta Limongiello, Filippo Frioni, Luca Di Marino, Monica Rossi, Claudio Pellegrino, Caterina Giovanna Valentini, Annalisa Ruggeri, Luciana Teofili and Simona Sica.
Unrelated cord blood transplantation and post-transplant cyclophosphamide.
Haematologica. 2026 May 28. doi: 10.3324/haematol.2026.300530 [Epub ahead of print]

Unrelated cord blood transplantation and post-transplant cyclophosphamide

Patrizia Chiusolo[1,2], Andrea Bacigalupo[1,2], Sabrina Giammarco[1,2], Elisabetta Metafuni[1,2], Federica Sora'[1,2], Maria Assunta Limongiello[1], Filippo Frioni[2], Luca Di Marino[2], Monica Rossi[1], Claudio Pellegrino[2], Caterina Giovanna Valentini[3], Annalisa Ruggeri[4], Luciana Teofili[2,3], Simona Sica[1,2]

1.Ematologia, Dipartimento di Scienze Laboratoristiche ed Ematologiche, Fondazione Policlinico Universitario Agostino Gemelli IRCCS.
2.Sezione di Ematologia, Università Cattolica del Sacro Cuore, Roma, Italy.
3.Servizio di Emotrasfusione, Dipartimento di Scienze laboratoristiche ed Ematologiche, Fondazione Policlinico Universitario A Gemelli IRCCS.
4. San Raffaele Scientific Institute, Hematology and Bone marrow Transplantation Unit, Milan, Italy.

Corresponding Author
Andrea BAcigalupo
Istituto di Ematologia,
Fondazione Policlinico Universitario A Gemelli IRCCS
Roma, Italy
andrea.bacigalupo@unicatt.it

Disclosures : the Authors declare no conflict of interest

Contributions: Patrizia Chiusolo and Andrea Bacigalupo designed the study , analyzed the data and wrote the manuscript ; Sabrina Giammarco, Elisabetta Metafuni, Federica Sora', Maria Assunta Limongiello, Filippo Frioni, Luca Di Marino, Monica Rossi, Claudio Pellegrino, Caterina Giovanna Valentini, Annalisa Ruggeri, Luciana Teofili, Simona Sica provided clinical for the patients and data collection; Simona Sica and Patrizia Chiusolo supervised the study.

Data sharing : the data is available upon request

To the Editor

The use of unrelated cord blood transplantation (UCBT) has been declining over the years, for patients who lack an HLA matched donor, due to the competition of haploidentical grafts **(1).** However UCBT may have advantages, such as a strong graft versus leukemia (GvL) effect **(2,3).** There are disadvantages, such as delayed engraftment and extensive chronic graft versus host disease (GvHD), may be as high as 30%, after double UCBT **(4).** We have reported a first patient with acute myeloid leukemia (AML) undergoing a successful unrelated cord blood transplant (UCBT) using reduced dose post-transplant cyclophosphamide (PTCY) , cyclosporin (CSA) and mycophenolate (MMF) for graft versus host disease (GvHD) prophylaxis **(5)**. We are now reporting the interim analysis of 14 patients undergoing UCBT in our Center, on the same clinical trial (NCT03802773), approved by our local Ethical Committee (ID 2382).

Patients Eligible for the study were patients who lacked a suitable haploidentical related or a 7/8 unrelated donor. The study enrolled 14 patients, including 12 females and 2 males, with a median age at transplant of 56 years (range: 36-68); the median weight was 58 kg (50-85 kg) **(Tab.1)**. Patients were grafted between 2017 and 2023. Diagnosis, phase of the disase, are detailed in Table 1, together with HLA allele matching and number of CB infused cells x10^7/kg; CB units were prioritized on the following criteria: cell dose, DRB1 match, non ABO major mismatch. The median Hematopoietic Cell Transplant-Co-Morbidity Index (HCT-CI) was 4 (0-7).

Conditioning regimen. The conditioning regimen in 13/14 patients, was a combination of thiotepa 10 mg/kg, fludarabine 150 mg/m^2 and busulfan 3,2mg/kgx3 (TBF3); with high HCT-CI and or advanced age the days of busulfan infusion were reduced to 2 (TBF2) or 1 day (TBF1), thiotepa and fludarabine remaining at the dame dose; one patient in very poor clinical conditions (n.12) received a combination of fludarabine cyclophosphamide and total body irradiation 2 Gy.

GvHD prophylais. All patients received PTCY (30 mg/kg on days +3 and +5), combined with a calcineurin inhibitor (all patients received CsA except for one patient who received tacrolimus, starting on day zero) ; mycophenolate mofetil (MMF) from day+1 to +36 was given to the first 10 patients and was omitted in the last 4 patients, following an amendment to the study (16/07/2020). Subcutaneous GCSF , 250 ug/ day, was given daily from day +5 untill neutrophil engraftment.

Engraftment. All patients received an optimal CD34+ dose -according to EBMT guidelines- with a median of CD34 cells 1.57x 10^5/kg and total nucleated cells 3.05x10^7/kg (range 1.8-5.0). Two patients were not evaluable for engraftment, because of early sepsis (n.3 and n.9) (Stenotrophomonas maltophilia and multidrug-resistant Klebsiella pneumoniae). One patient was not evaluable because of autologous leukemic recovery.

The cumulative incidence of engraftment in the 11 evaluable patients, was 82%: two patients (18%) had primary graft failure (PrGF)- one was rescued with a mismatched unrelated donor HSCT, one died on day +54. PrGF occurred in 2/8 (25%) patients grafted with UCB matched at 4/8 alleles, and

in 0/4 patients grafted with UCB matched at 5/8 alleles; PrGF occurred in 1/7 remission and in 1/5 R/R patients, and both occurred in patients receiving the highest intensity conditioning (TBF3), and a high cell dose (3.1 and 5.0x10^7/kg) **(Tab.1)**. The time to first neutrophil engraftment was 23 days (range 15-33) and the median time to platelet engraftment was 38 days (34-42). Two patients had acute GvHD grade I and 1 patient had grade II GvHD. Minimal chronic GvHD was diagnosed in 1 patient and moderate chronic GvHD in 2 patients. The cumulative incidence of acute GvHD grade II was 7%. The cumulative incidence of moderate chronic GvHD at 3 years is 11%. Cyclosporine was discontinued at a median interval of 141 days (97-390). The median absolute CD4 counts on day +50 was 75/cmm (range 50-116).

Survival . The overall actuarial survival at 3 years is 50% (95%CI 23-76%), with a median overall survival of over 1000 days (range 7-3002): when stratified for remission status this was 75% for remission patients and 17% for R/R patients.

The 3 year CI of transplant related mortality (TRM) is 21%; and for remission or R/R patients it is respectively 25% and 17%; the 3 year CI of relapse is respectively 12% and 66%.

Conclusions. A previous study tested the use of intrabone CB transplants with anti-thymocyte globulin (ATG) prophylaxis **(6):** although engraftment was satisfactory, an update of that study on 127 patients (unpublished), showed a TRM of 36% , of which 55% was due to infections and 22% to GvHD. For this reason we have tested a PTCY based, as opposed to an ATG based GvHD prophylaxis: the overall TRM in the first 14 patients, is 21%, with no case of severe acute or chronic GvHD, no case of GvHD related death, and the actuarial 3 year survival for remission patients , is in excess of 70%. There have been 2 patients with primary graft failure, which is 18% of evaluable patients: one was rescued with a mismatched unrelated graft, the other deceased with aplasia. Intrabone injection of UCBT may be one way to improve engraftment **(6)**, combined with low dose PTCY**.** The cumulative incidence of acute and chronic GvHD was very low, which led to an amendment of the protocol, eliminating MMF. Because of a low incidence of GvHD , cyclosporin could be discontinued early at a median interval of 141 days from transplant. Sepsis remains a common complication of CB transplants, as demonstrated by our study population, in which all 3 transplant related deaths occurred due to sepsis. Only one patient developed EBV DNA-emia, requiring rituximab treatment, in contrast with prior trials including ATG **(7)**. CMV reactivation was seen in the first patients, but not later, due to the use of letermovir prophylaxis. The limitations of this study are currently related to the small number of patients enrolled; nevertheless we believe we have shown a proof of concept, suggesting PTCY as GvHD prophylaxis in UCBT. An amendment of the study has just been approved (ID 2382 , amendment 26101 ;CET 5.february.2026) : the study is now multicenter, it will include pediatric patients, the dose of PTCY will be further reduced to 25 mg/kgx2, and double CB units as well as intrabone infusion, will be included.

REFERENCES

1. Passweg JR, Baldomero H, Bader P, et al. Use of haploidentical stem cell transplantation continues to increase: the 2015 European Society for Blood and Marrow Transplant activity survey report. Bone Marrow Transplant. 2017;52(6):811-817.
2. Milano F, Gooley T, Wood B, et al. Cord-blood transplantation in patients with minimal residual disease. N Engl J Med. 2016;*375*(10):944-953.
3. Horgan C, Mullanfiroze K, Rauthan A, et al. T-cell replete cord transplants give superior outcomes in high-risk and relapsed/refractory pediatric myeloid malignancy. Blood Adv. 2023;7(10):2155-2165.
4. Michel G, Galambrun C, Sirvent A, et al. Single- vs double-unit cord blood transplantation for children and young adults with acute leukemia or myelodysplastic syndrome. Blood. 2016;127(26):3450-3457.
5. Bacigalupo A, Sica S, Laurenti L, Sora' F, et al. Unrelated cord blood transplantation and post-transplant cyclophosphamide. Haematologica. 2019;104(2):e77-e78.
6. Frassoni F, Gualandi F, Podestà M, Piaggio G, et al. Direct intrabone transplant of unrelated cord-blood cells in acute leukaemia: a phase I/II study. Lancet Oncol. 2008;9(9):831-839.
7. Brunstein CG, Weisdorf DJ, DeFor T, et al. Marked increased risk of Epstein-Barr virus-related complications with the addition of antithymocyte globulin to a nonmyeloablative conditioning prior to unrelated umbilical cord blood transplantation. Blood. 2006;108(8):2874-2880.

Tab.1 Clinical characteristics of patients

	G/A	Diagn	Phase	HLA	Cells	Condit	Engr	Plt	GvHD Ac	GvHD Chr	Status	FU	Cause
N.1	F/44	AML	CR2	5/8	4.2	TBF3	15	36	0	0	alive	3002	-
N.2	F/54	ALL	CR1	5/8	4.5	TBF3	25	25	I	0	alive	2740	-
N.3	F/59	sAML	REL	2/8	5.0	TBF1					dead	15	seps
N.4	M/56	AML	CR2	4/8	1.8	TBF3	22	40	0	mod	alive	2568	-
N.5	F/49	sAML	REL	5/8	2.3	TBF3	18	34	0	min	alive	2567	-
N.6	F/58	AML	CR1	4/8	3.1	TBF3	PGF	-	0	0	alive	2446	-
N.7	F/66	sAML	REL	4/8	2.7	TBF1	25	40	0	0	dead	614	rel
N.8	F/63	AML	REL	5/8	2.7	TBF2	27	41	0	0	dead	561	rel
N.9	F/63	sAML	CR2	5/8	2.8	TBF2					dead	7	seps
N.10	M/43	AML	CR1	4/8	5.0	TBF3	PGF				dead	54	seps
N.11	F/46	AML	CR1	4/8	2.8	TBF3	23	42	II	mod	alive	2127	-
N.12	F/53	FCL	REL	4/8	3.0	FC	18	38	0	0	dead	300	rel
N.13	F/68	AML	REL	2/8	4.1	TBF2					dead	55	rel
N.14	F/36	AML	CR2	5/8	3.6	TBF2	33	38	0	0	alive	1364	-

Abbreviations: G/A: gender and age; Diagn: diagnosis; HLA: number of HLA matched alleles; Cells: number of cells infused x10^7/kg; Engr: days of neutrophil engraftment; GvHD: graft versus host disease, acute (ac)(GRADE 0, I, II) and chronic (chr) (min= minimal; mod= moderate); FU= follow in days from transplant ; Cause: cause of death: seps= sepsis; rel= relapseAML: acute myeloid leukemia; sAML: secondary AML; ALL= acute lymphoblastic leukemia; FCL: follicular cell lymphoma; CR: comple remissione (1 or 2); REL: relapse; TBF: 13 patients received a combination of thiotepa, busulfan , fludarabine;TBF1, TBF2, TBF3 indicates days of busulfan; PGF: primary graft failure (see text for details).