

Posttransplant cyclophosphamide as GVHD prophylaxis in patients receiving mismatched unrelated HCT: the PHYLOS trial

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Key Points

- PPTCy-based GVHD prophylaxis leads to a low rate of aGVHD, a low incidence of NRM and acceptable relapse rate after HSCT from MMUD

Posttransplant high-dose cyclophosphamide (PTCy) is effective in overcoming the negative impact of HLA disparity in the haploidentical setting. In light of these results, we investigated the efficacy of PTCy, in improving clinical outcomes of hematopoietic stem cell transplantation (HSCT) from a mismatched unrelated donor (MMUD) in patients with acute myeloid malignancies by reducing the incidence and severity of acute graft-versus-host disease (aGVHD). A prospective, single-arm, phase 2 study (PHYLOS) was conducted by the Gruppo Italiano Trapianto di Midollo Osseo. The ethical committees of the participating centers approved the study (EURODRAC 2017-003530-85). A total of 77 consecutive patients (acute myeloid leukemia: 64; myelodysplastic syndrome: 13) were enrolled at 26 Italian transplant centers (January 2020-November 2022). Median age of the patients was 53 (range, 19-65) years. The 100-day cumulative incidence of grades 2 to 4 aGVHD was 18.2% (95% CI,

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Deidentified individual participant data that underlie the reported results will be made available for a period of 1 year after the publication date. Deidentified patient data are available on request from the corresponding author, Anna Maria Raiola (annamaria.raiola@hsanmartino.it). If available, data will be provided electronically in Excel

worksheets, within a reasonable timeframe. The study protocol is included as a data supplement available with the online version of this article.

The full-text version of this article contains a data supplement.

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10.6-27.6) and of grades 3 to 4 was 6.5% (95% CI, 3.1-15.1). Seventy-one patients (92%) had full-donor chimerism with complete neutrophil engraftment by day +30. One-year cumulative incidence of chronic GVHD was 13.4% (95% CI, 6.9-22.1). One-year cumulative incidence of nonrelapse mortality was 9.1% (95% CI, 4.0-16.9), and the relapse rate was 23.8% (95% CI, 14.9-33.9). One-year overall survival and graft relapse-free survival were 78.6% (95% CI, 67.4-86.3) and 55.3% (95% CI, 43.4-65.7), respectively. Our study in a homogeneous patient cohort suggests that PTCy leads to a low rate of aGVHD and improves clinical outcomes of HSCT from MMUD. This trial was registered at www.clinicaltrials.gov as #NCT03270748.

Introduction

Allogeneic hematopoietic cell transplantation (HCT) is a curative treatment option for several malignant and nonmalignant hematologic diseases, and HLA disparity remains one of the most important factors affecting long-term survival. Indeed, outcomes are inferior with HLA-mismatched unrelated donors (MMUDs).¹⁻³

Single HLA mismatches at HLA-A, -B, -C, or -DRB1 locus (7/8 HLA matched) are associated with an increased risk of severe acute graft-versus-host disease (aGVHD), resulting in increased nonrelapse mortality (NRM) and decreased overall survival (OS) when compared with matched related or unrelated donor HCT.⁴⁻¹⁰ Strategies to facilitate the use of MMUDs are relevant for patients from ethnic minorities or in the presence of familial disease. The probability of finding a matched unrelated donor can vary between 16% and 75%, depending primarily on the patient's ethnic background.¹¹ To overcome HLA diversity and intensify immunosuppression, in vivo T-cell depletion (thymoglobulin/antilymphocyte or alemtuzumab) is used as GVHD prophylaxis for MMUD HCT, but a shared schedule of treatment in homogenous group of patients is lacking and results in terms of acute and chronic GVHD (cGVHD) are heterogeneous.¹²⁻¹⁶

Posttransplant high-dose cyclophosphamide (PTCy) is effective in overcoming the negative impact of HLA disparity with familial haploidentical donors.¹⁷⁻¹⁹ Indeed, when combined with a calcineurin inhibitor and mycophenolate, PTCy led to acceptable rates of engraftment, GVHD, and survival.²⁰⁻²² Several studies have been published on the use of PTCy in matched sibling and unrelated donor as GVHD prophylaxis, in combination or as a single agent.²³⁻²⁷

Results from 2 nonrandomized phase 2 studies suggest that PTCy alone may not be sufficient to prevent GVHD after a transplant with peripheral blood stem cells (PBSCs) from an HLA identical donor (grades 2-4 GVHD 45%) supporting the concept that combination with another immunosuppressive drug may optimize the effect, at least in the PBSC HCT setting.^{28,29}

In contrast, limited data are available in the context of transplant from MMUD with a few retrospective³⁰⁻³³ or prospective studies³⁴⁻³⁶ that included heterogeneous populations. Two European Bone Marrow Transplantation (EBMT) retrospective studies compared PTCy and anti-thymocyte globulin (ATG) in this setting, and both reported better outcomes with PTCy.^{37,38} Therefore, we investigated the efficacy and safety of PTCy, combined with calcineurin inhibitor and mycophenolate mofetil, in the context of a

homogeneous group of 1 antigen/allele mismatched (7/8) unrelated HCT in patients with myeloid malignancies after a myeloablative conditioning (MAC) regimen.

Patients and methods

Study design and eligibility

PHYLOS (NCT03270748 EURODRAC 2017-003530-85) is a prospective, phase 2, multicenter, single-arm, open-label study promoted by Gruppo Italiano per il Trapianto di Midollo Osseo, cellule staminali e terapia cellulare (GITMO).

Eligibility criteria for a first allogeneic HCT were an age of 18 to 65 years, a diagnosis of acute myeloid leukemia (AML) or myelodysplastic syndrome (MDS) in complete remission (CR) at transplant, an Eastern Cooperative Oncology Group performance status ≤ 2 , and an adequate cardiac, liver, pulmonary, and renal function. Patients were excluded if they had a suitable HLA-identical sibling or MUD available. Donor searches were carried out by the Italian Bone Marrow Donor Registry. High-resolution HLA typing was performed for all donor-recipient pairs matching for HLA-A, -B, -C, -DRB1, and -DQB1. Any single antigen or allele mismatch at HLA-A, -B, -C, or -DRB1 was defined as 7/8. The presence of any other mismatches (HLA DQB1 and DPB1) has been reported.

Treatment plan

As conditioning regimen, all patients received busulfan 0.8 mg/kg 4 times per day in 2-hour infusions for 4 consecutive days (16 doses from day -6 to day -3) and fludarabine 40 mg/m² per day for 4 consecutive days (from day -6 to day -3) for a total dose of 160 mg/m². GVHD prophylaxis consisted of cyclophosphamide 50 mg/kg per day on days +3 and +4, cyclosporine or tacrolimus (continuous IV infusion and then oral) starting from day +5, with the dose adjusted to maintain a therapeutic level, to be continued for at least 100 days, and mycophenolate mofetil (15 mg/kg every 12 hours) starting from day +5 to day +35 (supplemental Figure 1). Patients received either granulocyte colony-stimulating factor-mobilized peripheral blood cells or bone marrow (BM) on day 0. ABO incompatibility was managed as per institutional procedures.

Supportive care

Granulocyte growth factor was given from day +5 to neutrophil engraftment. Mesna was administered to reduce the risk of hemorrhagic cystitis. Antimicrobial prophylaxis was administered according to institutional practice guidelines.

End point definitions

The primary end point was cumulative incidence of aGVHD, grades 2 to 4, at 100 days after MMUD transplants with PTCy in patients with AML (CR1 or CR2) or MDS. aGVHD was scored according to the Glucksberg criteria. Secondary efficacy end points were as follows: 1-year OS, 1-year cumulative incidence of cGVHD and graft failure at 100 days and at 1 year, 1-year GVHD-free and relapse-free survival (GRFS), time to hematologic reconstitution (neutrophil and platelet recovery) at 30 days from transplant, time and causes of NRM, cumulative incidence and time of disease relapse, incidence of infectious complications, and time to immune reconstitution. OS was defined as the time from transplant to the date of death regardless of the cause. The duration of follow-up was at least 1 year for all the enrolled patients alive at data cut-off. cGVHD was defined and scored according to the National Institutes of Health consensus criteria. Time to neutrophil engraftment was defined as the first of 3 consecutive days with an absolute neutrophil count $>0.5 \times 10^9/L$ after transplantation. Time to platelet engraftment was defined as the first of 3 consecutive days with a platelet count $>20 \times 10^9/L$ without transfusion. Graft failure was defined as lack of donor-derived engraftment ($<95\%$ donor chimerism) by day 30. NRM was defined as death without evidence of disease. Relapse was defined by either morphological evidence of AML or MDS consistent with pretransplant features. Pretransplant minimal residual disease (MRD) was defined as any evidence of a defined abnormality using conventional cytogenetics, fluorescence in situ hybridization techniques, cytofluorimetric analysis, or molecular probes. GRFS was defined as occurrence of grades 3 to 4 aGVHD or cGVHD requiring systemic immunosuppressive treatment, disease relapse, or death from any cause during the first 12 months after hematopoietic stem cell transplantation (HSCT). Safety assessments included reports of adverse events graded according to the Common Terminology Criteria Adverse Events (version 4.03; 14 June 2010).

Statistical analysis

This was a single-arm phase 2 trial based on Fleming single-stage design. The MMUD transplant reference rate of aGVHD (grades 2-4) was assumed to be 50%.^{13,14} We hypothesized to observe an absolute 15% aGVHD reduction with this protocol, from an expected 50% to 35%. A total of 78 patients were required to test this hypothesis with a 1-sided significance level of 0.05 and a power of 85%. According to the sample size, 31 was the maximum number of patients who may develop aGVHD to reject the null hypothesis.

A safety interim analysis was planned after the first 30 enrolled patients on the incidence of grades 2 to 4 GVHD. An independent panel of experts not involved in the study has monitored the study safety.

Patient, disease, and transplantation characteristics were summarized using descriptive statistics. Categorical variables are reported as absolute value and percentage, whereas quantitative variables are described as median value and range.

Cumulative incidence of relapse, NRM, aGVHD, and cGVHD was calculated using the Fine and Gray method, considering the respective competitive risks (death in remission for relapse, disease

recurrence for NRM, and death for GVHD). One patient was censored at the data of second transplant.

Ethical consideration

The study was conducted according to Good Clinical Practice and the Declaration of Helsinki and was approved by the ethical committee of all participating centers. All patients provided written informed consent.

Results

Patient characteristics

Between 15 January 2020 and 10 November 2022, a total of 81 patients from 26 GITMO centers were screened. Four patients were excluded because they did not meet the inclusion criteria; 77 patients were available for the final analysis: 64 AML and 13 MDS. The baseline demographic disease and transplantation-related characteristics are reported in [Table 1](#). Median age was 53 (range, 19-65) years, and 45 patients were males (54%). Of 64 patients with AML, 13 (20.3%) were classified as high risk according to ELN 2022 (European Leukemia NET), 15 (23.4%) had not achieved CR after first induction, and 28 (43.7%) had a positive MRD at transplant (specific molecular transcript or immunophenotype). The source of hematopoietic stem cells was peripheral blood for 72 patients (93.5%) and BM for 5 patients (6.5%). The interim analysis on the incidence of grades 2 to 4 aGVHD after the first 30 patients did not have a higher-than-expected risk of developing GVHD and/or a higher incidence of transplant-related mortality.

Primary end point: aGVHD

At day 100, cumulative incidence of grades 2 to 4 aGVHD was 18.2% (95% CI, 10.6-27.6; 90% CI, 1.6-25.9) (14 patients) ([Figure 1A](#); [Table 2](#)). Furthermore, 5 patients, 8 patients, and 1 patient had 1, 2, and 3 organs involved (5 only gut; 8 skin and gut; and 1 gut, skin, and liver), respectively. The gut was always an involved organ (14/14). Within 100 days from transplant, 5 patients (6.5%, 95% CI, 3.1; 15.1%) developed grades 3 to 4 aGVHD ([Figure 1B](#)) and 17 patients grade 1; 6 of them received a therapeutic dose of steroid.

Engraftment

There were 5 patients who failed to achieve engraftment and died while having cytopenia, and 1 patient had autologous recovery and underwent a second transplant from a different donor. Furthermore, 71 patients (92%) had full-donor chimerism by day +30 as tested on unmanipulated BM and selected peripheral blood CD3⁺ cells, including 4 of 5 patients transplanted from BM. The median time (days) to neutrophil recovery was +17 (range, 10-35), and the median time to platelet recovery was +22 (range, 11-137). Cumulative incidence of engraftment was 88.3% for neutrophils on day +30 and 83.2% for platelets on day +60 ([Figure 2](#)).

cGVHD

The 1-year cumulative incidence of cGVHD and moderate/severe cGVHD was 13.4% (95% CI, 6.9-22.1) and 9.4% (95% CI, 4.1-17.2) (10 and 7 patients), respectively ([Figure 1C](#)).

Table 1. Patients, disease, and transplant characteristics

Patients	N = 77
Age, median (range), y	53 (19-65)
Patient sex, n (%)	
Male/female	45 (58.4%)/32 (41.6%)
ECOG, n (%)	
0	64 (83.1%)
>0	11 (14.3%)
Unknown	2 (2.6%)
HCT-CI, n (%)	
0-2	64 (83.1%)
≥3	13 (16.9%)
Disease	
AML, n (%)	64 (83.1%)
WHO classification, n (%)	
AML with recurrent genetic abnormalities	16 (25.0%)
AML with myelodysplasia-related changes	15 (23.4%)
Therapy-related myeloid neoplasm	4 (6.3%)
AML, NOS	28 (43.8%)
Myeloid sarcoma	1 (1.6%)
Cytogenetic risk (ELN 2022), n (%)	
Favorable	14 (21.9%)
Intermediate	37 (57.8%)
Adverse	13 (20.3%)
Molecular markers	
<i>FLT 3</i>	15
<i>NPM1</i>	19
<i>IDH1/IDH2</i>	8
<i>ASXL1</i>	3
<i>AML1/ETO RUNX1::RUNX1T1</i>	2
<i>BCR::ABL1</i>	2
No CR postinduction therapy	15 (23.4%)
≥2 CR	11 (17%)
MRD positive at transplant, n (%)	
Molecular markers	14/64 (21%)
<i>NPM</i>	11 (17%)
<i>AML1/ETO</i>	2 (3%)
<i>bcr/abl</i>	1 (1%)
IF	14/64 (21%)
MDS, n (%)	13 (16.9%)
WHO classification, n (%)	
MDS with single lineage dysplasia	2 (15.4%)
MDS with multilineage dysplasia	2 (15.4%)
MDS with excess blasts	6 (46.1%)
MDS with isolated del(5q)	1 (7.7%)
MDS, unclassifiable	2 (15.4%)
HSCT up-front	5
Transplant and donors	
Diagnosis: transplant, median (range), d	202 (115-1482)

Table 1 (continued)

Patients	N = 77
Year of the transplant, n (%)	
2020	21 (27.3%)
2021	32 (41.6%)
2022	24 (31.2%)
Conditioning Bu4Flu, n (%)	77 (100%)
Donor age, median (range), y	29 (18-62)
Donor sex, n (%)	
Male/female	51 (66.2%)/26 (33.8%)
Donor/recipient sex mismatch, n (%)	
Male/male	31 (40.2%)
Male/female	14 (18.2%)
Female/male	20 (26.0%)
Female/female	12 (15.6%)
CMV serostatus, n (%)	
+/+	34 (44.1%)
-/+	19 (24.7%)
+/-	9 (11.7%)
-/-	6 (7.8%)
Missing	9 (11.7%)
Stem cell source, n (%)	
PB	72 (93.5%)
BM	5 (6.5%)
Cell dose	
PB median (range), CD34 ⁺ , ×10 ⁶ /kg	5.9 (2.2-12.2)
BM median (range), CD34 ⁺ , ×10 ⁶ /kg	2.3 (1.4-5.3)
Donor HLA mismatches 7/8, n (%)	77 (100%)
HLA class I mismatch (A, B, C)	72 (93.5%)
HLA class II mismatch (DRB1)	3 (4%)
Missing	2 (2.5%)

CMV, cytomegalovirus; ECOG, Eastern Cooperative Oncology Group; HCT-CI, hematopoietic cell transplantation-comorbidity index; IF, immunophenotyping; NOS, not otherwise specified; PB, peripheral blood; WHO, World Health Organization.

Relapse incidence

The 1-year cumulative incidence of relapse rate was 23.8% (18 patients) (95% CI, 14.9-33.9) (Figure 1D). Relapse was the cause of death in 9 patients (12%). Considering only the AML cohort, 15 of 64 patients relapsed at 1 year.

NRM

The 1-year cumulative incidence of NRM was 9.1% (7 patients) (95% CI, 4.0-16.9) (Figure 1E). The causes of death were sepsis (n = 3), pneumonia (n = 2), and multi-organ failure (n = 2).

OS and GRFS

The 1-year GRFS and OS were 55.3% (95% CI, 43.4-65.7) and 78.6% (95% CI, 67.4-86.3), respectively (Figure 1F-G).

The overall outcomes are outlined in Table 2.

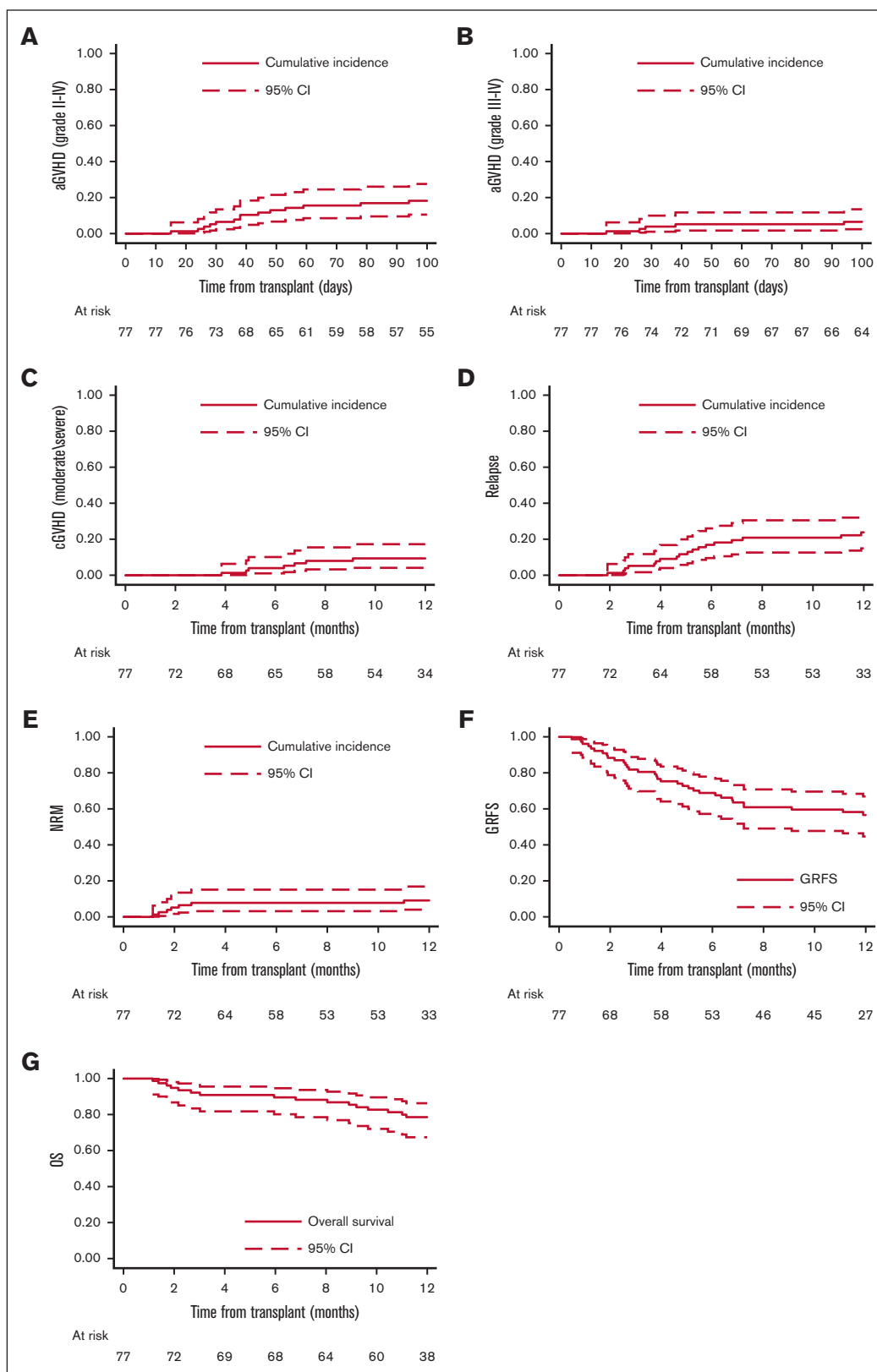


Figure 1. Overall outcomes. (A) Cumulative incidence of aGVHD grades 2 to 4 at 100 days. (B) Cumulative incidence of aGVHD grades 3 to 4 at 100 days. (C) Cumulative incidence of cGVHD moderate/severe at 12 months. (D) Cumulative incidence of relapse at 12 months. (E) Cumulative incidence of NRM at 12 months. (F) GRFS at 12 months. (G) OS at 12 months.

Table 2. End points

	No. of events	Estimate	95% CI, %	
Cumulative incidence of aGVHD at 100 days				
Any grade	28	39.0%	28.1	49.6
Grades 2-4	14	18.2%	10.6	27.6
Grades 3-4	5	6.5%	3.1	15.1
Cumulative incidence of cGVHD at 12 months				
Any grade	10	13.4%	6.9	22.1
Moderate/severe	7	9.4%	4.1	17.2
Cumulative incidence of relapse at 12 months	18	23.8%	14.9	33.9
Cumulative incidence of NRM at 12 months	7	9.1%	4.0	16.9
GRFS at 12 months	33	55.3%	43.4	65.7
OS at 12 months	16	78.6%	67.4	86.3

Toxicity

The most common serious adverse events were infections and infestations (24 events in 21 patients), gastrointestinal disorders (26 events in 18 patients), and blood and lymphatic system disorders (19 events in 14 patients) (Table 3). The most common toxicity was oral mucositis (26 patients). The most frequent infectious complications were of bacterial origin with 18 episodes of bloodstream infections, followed by viral reactivations (only 4 cytomegalovirus reactivations). Cardiovascular events occurred in 3 patients.

Discussion

This multicenter, single-arm phase 2 trial enrolling patients with myeloid malignancies demonstrated that a PTCy-based GVHD prophylaxis in HCT from a MMUD leads to a low rate of aGVHD, with a low incidence of NRM, and acceptable relapse rate. GVHD still represents a major limitation of HSCT in general, especially when there are 1 or more HLA mismatches between donor and recipient.^{8,10} Posttransplant administration of cyclophosphamide has proven to be strategic to prevent GVHD, not only in the HLA-haploidentical HCT setting but also in HLA-matched related or unrelated HCT.^{17-25,38} Nevertheless, only few systematic studies have confirmed the feasibility and efficacy of PTCy as GVHD prophylaxis in MMUD. Indeed, in an initial prospective study, in the context of non-MAC with BM stem cell source, a PTCy-based GVHD prophylaxis strategy prevented both incidence of graft failure and grades 3 to 4 aGVHD, suggesting a prominent role of PTCy in overcoming HLA differences also in an unrelated setting.³⁹ These data were confirmed in a subsequent study in the MMUD HCT using PBSC as the stem cell source, in which PTCy was found to lower GVHD and NRM at levels comparable to those observed in transplant from matched unrelated donors.⁴⁰ However, both studies enrolled patients with any hematologic malignancies, exploiting mostly non-MAC/reduced intensity conditioning regimens.

The PHYLOS trial aimed to investigate the impact of PTCy in AML and MDS in first or subsequent remission, receiving the same MAC, followed by a peripheral blood (PB) transplant in most patients. We adopted this regimen because in a previous GITMO study on patients with AML in CR at transplant,⁴¹ this conditioning schedule, although still myeloablative, resulted in a lower

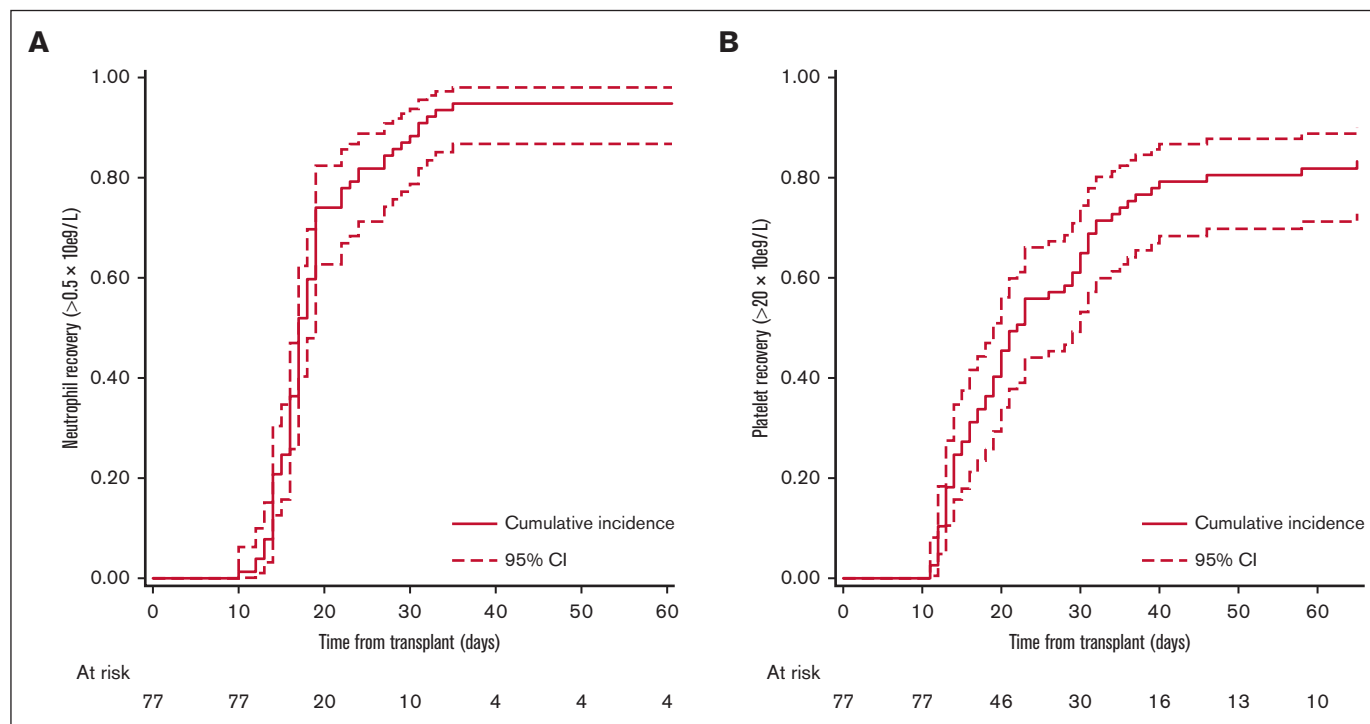


Figure 2. Count recovery. (A) Cumulative incidence of neutrophil recovery. (B) Cumulative incidence of platelet recovery.

Table 3. Toxicity grades 3 to 5 of adverse events until day 360 after HCT

Toxicity	Grades 3-5	
	n	%
Infections and infestations	21	27.27
Gastrointestinal disorders	18	23.38
Blood and lymphatic system disorders	14	18.18
General disorders	2	2.60
Respiratory, thoracic, and mediastinal disorders	8	10.39
Investigations	6	7.79
Vascular disorders	4	5.19
Metabolism and nutrition disorders	2	2.60
Eye disorders	1	1.30
Cardiac disorders	3	3.90
Musculoskeletal and connective tissue disorders	1	1.30
Renal and urinary disorders	1	1.30
Hepatobiliary disorders	1	1.30
Nervous system disorders	1	1.30
Psychiatric disorders	1	1.30

Adverse events graded according to the Common Terminology Criteria Adverse Events (version 4.03: 14 June 2010).

transplant-related mortality compared with busulfan and cyclophosphamide, even in older patients. Consistently, in our experience, the 1-year cumulative incidence of NRM was 9.2% with infections as the dominant cause of death.

Results from our trial are comparable with those reported in a NMDP prospective study, which reported 1-year rates of NRM, relapse, and OS of 8%, 30%, and 70%, respectively, in patients receiving a MAC.⁴² However, in the NMDP study, BM was mandatory as stem cell source, and approximately one-third of patients were matched 4 to 6 of 8, whereas in our study there was no restriction in terms of stem cell source and most patients (72/77) were transplanted from PB (probably due to the ease of collection and an expected faster engraftment). Data on transplant outcomes according to the stem cell source (PB or BM) are lacking in this setting, and our study provides the first and more robust results with PTCy given in the context of a PB-based MMUD HSCT.

Relapse remains an issue, as in our study cumulative incidence was 24% at 1 year, with near one-third of patients harboring high-risk features (biological high-risk, first-line refractory, and/or MRD positive at transplant), comparable to that of previous trials, either prospective or retrospective. Recently, a meta-analysis that included several hundred patients undergoing MMUD HSCT with PTCy reported a pooled relapse rate of 44%.⁴³ Leukemia relapse after transplantation is a clinical need for patients with AML, especially for those with high-risk features. For this reason, in the future, innovative postengraftment treatments, such as cellular therapies or targeted drugs, should be integrated within the allogeneic HSCT (allo-HSCT) backbone, taking advantage of a possibly better GVHD prophylaxis which allows for a smoother post-HSCT clinical course.

Considering current experiences with MMUD, one might wonder whether a direct head-to-head, possibly randomized comparison

between ATG and PTCy as GVHD prophylaxis strategy is possible and eventually needed to set PTCy as a new standard of care for MMUD HSCT.

Some responses come from 2 large retrospective studies that indirectly compared the 2 GVHD prophylaxis strategies. In 2019, an EBMT retrospective study analyzed the results of allo-HSCT from MMUDs in a homogenous population of patients with AML, comparing the outcomes of PTCy vs ATG.³⁷ This study assigned to PTCy a lower rate of grades 3 to 4 aGVHD and similar rates of grades 2 to 4 aGVHD and any-grade cGVHD as compared with ATG. Furthermore, the authors observed superior survival outcomes in terms of leukemia-free survival and GRFS with the use of PTCy. Penack and EBMT³⁸ recently published an analysis on 2123 peripheral blood allo-HSCT, from MMUD (9/10 antigen matched) transplanted between January 2018 and June 2021. In their experience, PTCy was associated with an advantage in NRM, 18% vs 24.9% ($P = .028$), and in OS, 65.7% vs 55.7% (<0.001), in comparison to rabbit anti-thymocyte globulin. Progression-free survival was also better with PTCy at 59.1% vs 48.8% for rabbit anti-thymocyte globulin ($P = .001$). Interestingly, the incidence of cGVHD and aGVHD was not significantly different between the 2 groups. These results were confirmed by smaller monocentric experiences,^{31,44} but patients analyzed in these studies were heterogeneous for conditioning, stem cell source, and phase of disease. Whether these data and the results from prospective, non-randomized trials (including PHYLOS) are sufficient to consider PTCy as the standard for GVHD prophylaxis with MMUD is not clear.

Our trial has some limitations, such as the absence of a control group, a relatively short follow-up (1 year), and the possible impact of different HLA loci mismatches.

In conclusion, the PHYLOS reveals that PTCy-based MMUD transplants are feasible, safe, and effective, with results superimposable to those of haplo-related HSCT. Available data are not mature enough to conclude whether PTCy should be the preferred GVHD prophylaxis in this setting, but they eventually pave the way for future investigations exploiting the PTCy platform even outside the classical setting of haploidentical HSCT. In the future, the role of abatacept will also be a subject for discussion, due to the very promising results achieved in prospective studies after approval by the US Food and Drug Administration as GVHD prophylaxis in unrelated transplants.⁴⁵ This is even more important nowadays, in consideration of the emerging data driving donor selection. Indeed, the choice between a mismatched volunteer donor and a haplo-related donor could be no more hierarchical but inclusive of other factors, such as donor's age, sex mismatch, presence of anti-HLA antibodies, and/or ABO incompatibility,⁴⁶ together with the time required to find a suitable donor and the related costs. Further prospective studies are necessary to better investigate the role of PTCy in different HSCT settings and to dissect the possible impact of specific HLA loci (or even epitopes) or other biological features on GVHD, disease relapse, and finally long-term outcome of HSCT from other than matched related donors.

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Authorship

Contribution: A.M. Raiola designed the study and wrote the manuscript; B.B., A.M. Risitano, E.A., and F.B. designed the study and reviewed the manuscript; G.C. and A.C. performed statistical analysis; G.C., A.C., and E.D. extracted and analyzed row data updating reference list and sent queries to the participating centers; B.B., A.M. Risitano, F.M., I.M.C., F.O., G.S., F.P., M.L.B., V.P., A.M., P.C., S.S., B.L., C.d.G., A.M.C., D.S., E.M., A.L., L.G., P.B., E.T., N.M., C.B., F.Z., M.L., A.G., A.O., E.P., N.S., and M.M. screened potential eligible patients, enrolled them, and provided follow-up data; and all authors approved the final version of the manuscript.

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