Biol Blood Marrow Transplant 22 (2016) 902–909

Biology of Blood and Marrow Transplantation

journal homepage: www.bbmt.org

ASBMT American Society for Blood and Marrow Transplantation

Feasibility and Outcome of Haploidentical Hematopoietic Stem Cell Transplantation with Post-Transplant High-Dose Cyclophosphamide for Children and Adolescents with Hematologic Malignancies: An AIEOP-GITMO Retrospective Multicenter Study

Massimo Berger [1,*], Edoardo Lanino [2], Simone Cesaro [3], Marco Zecca [4], Elena Vassallo [1], Maura Faraci [2], Massimiliano De Bortoli [3], Veronica Barat [1], Arcangelo Prete [5], Franca Fagioli [1]

[1] *Pediatric Onco-Hematology and Stem Cell Transplant Division, City of Health and Science, Regina Margherita Children's Hospital, Turin, Italy*
[2] *Department of Pediatric Hematology-Oncology, IRCCS G. Gaslini, Genoa, Italy*
[3] *Pediatric Hematology-Oncology, Azienda Ospedaliera Universitaria Integrata, Verona, Italy*
[4] *Pediatric Hematology-Oncology, IRCCS Policlinico San Matteo Foundation, Pavia, Italy*
[5] *Pediatric Oncology and Hematology Unit "Lalla Seràgnoli", Department of Pediatrics, University of Bologna Sant'Orsola-Malpighi Hospital, Bologna, Italy*

Article history:
Received 8 October 2015
Accepted 1 February 2016

Key Words:
PTCy-haplo HSCT
Pediatric
Hematological malignancies

A B S T R A C T

Post-transplant high-dose cyclophosphamide (PTCy) is a novel approach to prevent graft-versus-host disease (GVHD) and rejection in patients given haploidentical hematopoietic stem cell transplantation (HSCT). Thirty-three patients with high-risk hematologic malignancies and lacking a match-related or -unrelated donor were treated with PTCy haploidentical HSCT in 5 Italian AIEOP centers. Nineteen patients had a nonmyeloablative preparative regimen (57%), and 14 patients received a full myeloablative conditioning regimen (43%). No patients received serotherapy; GVHD prophylaxis was based on PTCy (50 mg/kg on days +3 and +4) combined with mycophenolate plus tacrolimus or cyclosporine A. Neutrophil and platelet engraftment was achieved on days +17 (range, 14 to 37) and +27 (range, 16 to 71). One patient had autologous reconstitution for anti-HLA antibodies. Acute GVHD grades II to IV and III to IV and chronic GVHD developed in 22% (95% CI, 11 to 42), 3% (95% CI, 0 to 21), and 4% (95% CI, 0 to 27) of cases, respectively. The 1-year overall survival rate was 72% (95% CI, 56 to 88), progression-free survival rate was 61% (95% CI, 43 to 80), cumulative incidence of relapse was 24% (95% CI, 13 to 44), and transplant-related mortality was 9% (95% CI, 3 to 26). The univariate analysis for risk of relapse incidence showed how 3 significant variables, mother as donor ($P = .02$), donor gender as female ($P = .04$), and patient gender as female ($P = .02$), were significantly associated with a lower risk of relapse. Disease progression was the main cause of death. PTCy is a safe procedure also for children and adolescents who have already received several lines of chemotherapy. Among the different diseases, a trend for better 1-year rates of overall survival was obtained for nonacute leukemia patients.

INTRODUCTION

Haploidentical hematopoietic stem cell transplantation (haplo-HSCT) was initially performed with conventional post-transplant immunosuppression that often caused a strong bidirectional alloreactivity that in turn caused high primary graft failure, hyperacute graft-versus-host disease (GVHD), and very poor outcomes [1-3]. In the 1980s and, mostly, in the 1990s the use of ex vivo T cell–depleted grafts were followed by high engraftment probability, very low GVHD occurrence, and high transplant-related mortality (TRM) mainly because of impaired immune reconstitution [4-7].

Starting from basic research, 2 different approaches have gained scientific relevance: manipulated ex vivo T cell–depleted haplo-HSCT [8-12] and in vivo T cell–replete haplo-HSCT followed by post-transplant high-dose cyclophosphamide (PTCy) [13-15]. Initially, PTCy was found to

Financial disclosure: See Acknowledgments on page 908.

* Correspondence and reprint requests: Massimo Berger, MD, PhD, Pediatric Onco-Hematology and Stem Cell Transplant Division, City of Health and Science, Regina Margherita Children's Hospital, Piazza Polonia 94, 10126 Turin, Italy.
E-mail address: massimo.berger@unito.it (M. Berger).

http://dx.doi.org/10.1016/j.bbmt.2016.02.002
1083-8791/

reduce the incidence and severity of GVHD after HSCT in rodent models. When this observation was tested in humans, a randomized clinical trial demonstrated that a lower dose of Cy (7.5 mg/kg i.v. on days 1, 3, 5, 7, and 9 and then weekly) was inferior to cyclosporine A in preventing acute GVHD after HLA-matched sibling HSCT [16]. Subsequent studies showed that tolerance to minor histocompatibility antigens could be induced only when a single dose of ≥150 mg/kg Cy was given between 48 and 72 hours after alloantigen exposure. Moreover, tolerance was not induced if the same dose of Cy was given 24 or 96 hours after transplantation [17-19]. From a biologic point of view, hematopoietic stem cells are relatively quiescent and express high levels of aldehyde dehydrogenase, which likely confer cellular resistance to cyclophosphamide. The Seattle and the John Hopkins groups were the first to use the PTCy protocol early after stem cell infusion to control GVHD by eliminating rapidly dividing, donor-derived T cells generated by the major HLA mismatch graft [13,14,19]. However, most current literature on haplo-HSCT PTCy studies come from adult patients, and there is a very limited experience in the pediatric setting.

The aim of this study was to test PTCy after haplo-HSCT in a multicenter pediatric population, which lacks HLA-compatible related or unrelated donors. These pediatric patients were mostly referred to the Italian Associazione Italiana di Ematologia e Oncologia pediatrica (AIEOP, the Italian Association of Pediatric Haemato-Oncology) centers by low-income countries.

METHODS

From January 2012 to February 2015, 33 patients underwent a haploidentical transplant in 5 AIEOP centers (Turin, Genoa, Verona, Pavia, and Bologna). Patient median age was 12 years (range, 1-21), and 20 were boys (61%) and 13 girls. The details of patient, transplant, and donor characteristics are outlined in Table 1. Five patients had also experienced a failure of previous allogeneic HSCT (15%). The decision to proceed with haplo-HSCT was based on the absence of a compatible related or unrelated donor and/or the urgency of the procedure. Twenty-three patients had haplo-HSCT for acute leukemia (69%), and only 8 had HSCT in first complete remission (CR1) acute leukemia (24%). Five patients had haplo-HSCT for lymphoma, but none of them had HSCT in CR.

All transplants were performed in air-filtered rooms. Antimicrobial prophylaxis was started during conditioning and consisted of acyclovir 500 mg/m^2 3 times a day from day −5, cotrimoxazole 5 mg/kg over 2 consecutive days per week until day −2 and after engraftment, and antifungal prophylaxis with fluconazole unless contraindicated; otherwise, echinocandine was given. Acyclovir was maintained for 3 months after calcineurin inhibitors were stopped, whereas antifungal and cotrimoxazole were stopped when CD4$^+$ lymphocyte counts were above 200/μL.

Twice-weekly cytomegalovirus (CMV) PCR monitoring was started from day +15 until day +100 and then monitored weekly until day +180 or when clinically indicated. Weekly Epstein-Barr virus (EBV) and adenovirus PCR monitoring was started from day +15 to day +100 or when clinically indicated. The aspergillus galactomannan antigen test was performed twice a week.

Bacterial and fungal cultures were performed weekly or when clinically indicated. Piperacillin-tazobactam alone or in combination with an aminoglycoside was given as empirical therapy for febrile neutropenia, unless previous colonization for resistant bacteria was documented, and appropriate antimicrobial therapy was started. First-line pre-emptive CMV reactivation was based on i.v. ganciclovir, whereas foscarnet was preferred for low WBC patient counts. EBV reactivation was treated with rituximab at 375 mg/m^2 for 4 doses at weekly intervals, whereas polyoma virus or adenovirus-related hemorrhagic cystitis were initially treated by supportive care only.

The study was approved by the local Institutional Review Board. All parent or legal guardians signed the appropriate consent forms.

The nonmyeloablative (NMA) protocol consisted of fludarabine 150 mg/m^2 over 5 days together with Cy at 29 mg/kg over 2 days and a single total body irradiation dose at 200 cGy on day −1. For 10 patients the myeloablative (MA) regimen was busulfan-based with thiotepa and fludarabine, 2 patients received total body irradiation (1200 cGy) together with fludarabine, 1 patient received treosulfan combined with thiotepa and fludarabine, and, finally, 1 patient received thiotepa together with fludarabine and etoposide. The choice of MA or NMA conditioning was based on the patient performance status together with the center's policy.

Table 1
Clinical Data of Patients (N = 33) Receiving PTCy HSCT

Characteristic	Value
Gender	
Male	20 (61)
Female	13 (39)
Disease	
ALL	15 (45)
AML	7 (21)
Dendritic cell leukemia	1 (3)
MDS	4 (12)
CML	1 (3)
Lymphoma (HL and NHL)	5 (15)
Disease status	
CR1	8 (24)
CR2	10 (30)
CR3	5 (15)
Other	10 (30)
Donor relation	
Mother	18 (54)
Father	10 (30)
Brother	3 (9)
Sister	2 (6)
Sex mismatch	
Female donor−male recipient	13 (39)
Male donor−female recipient	6 (18)
Male donor−male recipient	7 (21)
Female donor−female recipient	7 (21)
Full haploidentical donor	
Yes	25 (75)
No	8 (24)
NK alloreactivity	
Yes	17 (51)
No	16 (49)
Conditioning regimen	
NMA	19 (57)
MA	14 (42)
GVHD prophylaxis	
Luznik-like	20 (61)
Others	13 (39)
BM stem cell source (n = 30), median (range)	
TNC	4.6 (1.9-11.1)
CD34$^+$	5.3 (1-12.9)
CD3$^+$	.7 (.4-1.2)
PBSC stem cell source (n = 3), median (range)	
TNC	8.4 (8.3-11.1)
CD34$^+$	13.2 (6.8-18.7)
CD3$^+$	2.3 (1.5-2.8)

ALL indicates acute lymphoblastic leukemia; AML, acute myeloid leukemia; MDS, myelodysplastic syndrome; CML, chronic myelogenous leukemia; HL, Hodgkin lymphoma; NHL, non-Hodgkin lymphoma; BM, bone marrow; TNC, total nucleated cell; PBSC, peripheral blood stem cell.
Values are total number of cases with percents in parentheses, unless otherwise noted.

For all patients GVHD prophylaxis was 50 mg/kg Cy on days +3 and +4 and mofetil mycophenolate 15 mg/kg 3 times a day from days +5 to +35. Twenty patients received tacrolimus .01 to .03 mg/kg from days +5 to +180, whereas the others received cyclosporine A (1 to 3 mg/kg) from days +5 to +180. One patient received steroids in place of calcineurin inhibitors until day +30. Granulocyte colony-stimulating factor was started from day +5 until neutrophil counts were above .5 × 10^3/μL for 3 consecutive days. Tacrolimus or cyclosporine A blood level concentrations were monitored 2 to 3 times per week.

HLA Typing and Compatibility

All HLA typings were performed in European Federation of Immunogenetics-accredited laboratories. All patients and donors were typed by high-resolution molecular standard technique at HLA-A, -B, -C, and -DRB1 loci. Twenty-five patients were fully haploidentical with their donors, 8 patients had HSCTs from a non−full haploidentical donor. HLA phenotyping was performed by the same methods and similar protocols. Briefly, typing