Published in final edited form as:

J Pediatr Hematol Oncol. 2018 May ; 40(4): e220–e224. doi:10.1097/MPH.0000000000001106.

Allogeneic matched related donor bone marrow transplantation for pediatric patients with severe aplastic anemia using “low dose” cyclophosphamide, ATG plus fludarabine

Chayamon Takpradit, MD^{1,2}, Susan E Prockop, MD^{1,3}, Nancy A Kernan, MD^{1,3}, Andromachi Scaradavou, MD^{1,3}, Kevin Curran, MD^{1,3}, Julianne Ruggiero¹, Nicole Zakak¹, Richard J O'Reilly^{1,3}, and Farid Boulad, MD^{1,3}

¹Department of Pediatrics, Bone Marrow Transplant Service, Memorial Sloan-Kettering Cancer Center, New York, NY ²Department of Pediatrics, Faculty of Medicine, Siriraj Hospital, Mahidol University, Bangkok, Thailand ³Division of Pediatric Hematology/Oncology, New York Presbyterian Hospital Weill Cornell Medical College, New York, NY

Abstract

Background—The combination of cyclophosphamide (CY) and anti-thymocyte globulin (ATG) has been used as a standard conditioning regimen for matched related donor transplantation in patients with severe aplastic anemia.

Procedure—To decrease the regimen related toxicity while maintaining appropriate engraftment and survival rates, fludarabine (FLU) was added to the regimen. Four pediatric patients received matched related donor bone marrow transplantation with CY (50 mg/Kg × 2) [instead of the 50 mg/Kg × 4 standard dosing], equine ATG (30 mg/Kg × 3) with the addition of FLU (30 mg/m² × 4). Graft versus host disease (GVHD) prophylaxis included a calcineurin inhibitor and methotrexate.

Results—No grade 4 acute toxicities occurred during the first 30 days post transplant. All patients engrafted with normalization of peripheral blood counts and transfusion independence. One patient developed grade 1–2 acute GVHD followed by chronic GVHD that resolved. With a median follow up of 41.7 months, all four patients are alive transfusion-free with complete donor chimerism. This combination of low dose CY/ATG + FLU regimen was overall very well tolerated and contributed to a successful outcome including engraftment, chimerism and survival.

Conclusion—This small pilot study shows that this cytoreductive regimen could be considered as the standard of care for transplantation of pediatric patients with aplastic anemia from HLA-matched siblings.

Correspondence: Farid Boulad, MD, Memorial Sloan-Kettering Cancer Center, 1275 York Avenue, New York, NY 10065, Tel: 212 639 6684, Fax: 212 717 3555 bouladf@mskcc.org.

Conflict of Interest:

The authors declare no conflict of interest.

Keywords

Aplastic anemia; hematopoietic stem cell transplantation; low dose cyclophosphamide; fludarabine; toxicity

INTRODUCTION

Acquired severe aplastic anemia (SAA) is a rare, life-threatening disease characterized by marrow failure.[1] Hematopoietic stem cell transplantation (HSCT) is the standard therapeutic approach for patients with severe aplastic anemia when a matched-related donor is available. The overall outcome of matched sibling transplantation in pediatric patients is overall superior to that in adult patients with an overall 5-year survival of 82%, with transplant related mortality primarily due to infections and graft failure.[2] The standard conditioning regimen for matched sibling HSCT of patients with SAA has comprised cyclophosphamide (CY) at a dose of 200 mg/Kg and equine anti-thymocyte globulin (ATG) (CY/ATG). This regimen promoted superior engraftment and survival rate compared with the historical regimen of CY alone.[3, 4] However, the use of high dose cyclophosphamide has been associated with acute toxicity and severe morbidity including severe mucositis, hemorrhagic cystitis, veno-occlusive disease (VOD), invasive fungal infections, as well as transplant related mortality.[5–7]

A number of investigators have attempted to decrease the toxicity of cyclophosphamide in the context of transplantation of both adult and pediatric patients with SAA by adding fludarabine (FLU) to the CY/ATG regimen (CY/ATG/FLU), while decreasing the dose of cyclophosphamide, therefore maintaining the immunosuppressive degree of the cytoreduction.[8–11] This regimen has been used successfully, particularly in alternative donor HSCT often with the addition of low dose total body irradiation (TBI).[12] In this series, we treated four pediatric patients with this approach of low dose CY, ATG plus FLU followed by unmodified grafts from human leukocyte antigen (HLA)-matched related donors. We describe their course and outcome.

PATIENTS AND METHODS

Patients

From January 2013 to January 2014, four patients with idiopathic acquired severe or very severe aplastic anemia received hematopoietic stem cell transplantation from HLA-matched related donors at Memorial Sloan Kettering Cancer Center. Approval of the Institutional Review Board/ Privacy Board was obtained.

Cytoreductive regimen and transplantation

All four patients received: fludarabine 30 mg/m² once daily intravenously (IV) on days -5, -4, -3, and -2; cyclophosphamide, at a total dose of 100 mg/Kg – half the standard dose of 200 mg/Kg - with 50 mg/Kg once daily IV on days -3 and -2; equine anti-thymocyte globulin (ATGAM, Upjohn, Kalamazoo, MI, USA) 30 mg/Kg once daily IV on days -4, -3, and -2. Adequate hydration was given during the conditioning chemotherapy and Mesna was

administered to prevent hemorrhagic cystitis. All patients received an unmodified bone marrow (BM) graft that was infused on day 0.

Graft versus host disease (GvHD) prophylaxis

A combination of calcineurin inhibitor including cyclosporine (patient 2 and 3) or tacrolimus (patient 1 and 4) and standard dose methotrexate (MTX) was administered to all 4 patients. MTX was given at a dose of 15 mg/m² IV on day +1, then 10 mg/m² IV on days +3, +6 and +11. All four patients received leucovorin rescue following MTX.

Supportive care and infectious prophylaxis

Irradiated leucocyte-reduced red blood cell and platelet transfusions were administered to maintain hemoglobin above 7 g/dL and platelet count $> 20 \times 10^9/L$. Filgrastim was administered at 5 mcg/Kg IV twice daily from day +1 until the ANC surpassed $2 \times 10^9/L$ for 3 consecutive days and was tapered thereafter. Intravenous low dose heparin and ursodiol were used for VOD prophylaxis. Antimicrobial prophylaxis included sulfamethoxazole/trimethoprim or pentamidine, acyclovir and micafungin. Broad-spectrum antibiotics were administered for the treatment of febrile neutropenia.

Assessment of engraftment and response

Neutrophil engraftment was defined as the first of 3 consecutive days with an absolute neutrophil count (ANC) $> 0.5 \times 10^9/L$, and platelet recovery was defined as the day the platelet count was $> 20 \times 10^9/L$ without platelet transfusions. Engraftment and chimerism were evaluated by short tandem repeat (STR) polymorphism analysis and fluorescence in situ hybridization (FISH) technique in all 4 sex-mismatched patients. Absolute lymphocyte counts; T-, B- and NK-cell populations were monitored. Acute and chronic GvHD were evaluated according to the 2014 National Institutes of Health Consensus.[13]

Toxicity

Regimen related toxicity (RRT) was graded according to Common Terminology Criteria for Adverse Events version 4.03.[14]

RESULTS

Patient characteristics (Table 1)

There were four patients (2 males and 2 females), with a median age of 12 years (range: 6 – 20 years) at transplantation. Three patients were diagnosed with SAA and one patient with very severe aplastic anemia (vSAA). The median cell count of absolute neutrophil, platelet and absolute reticulocyte at the time of diagnosis were $0.3 \times 10^9/L$, $7 \times 10^9/L$ and $24000/\mu L$, respectively. Patient 2 and patient 4 had been previously treated with filgrastim +/- eltrombopag without response. Three patients received less than 20 transfusions prior to HSCT while patient 4 was heavily transfused with red blood cells and platelets. The median time from diagnosis to transplantation was 3 months (range: 0.7 – 4.9 months). Specifically, patient 4 was diagnosed with vSAA and treated with immune suppression and eltrombopag with no response. In addition, this patient had a prolonged pre-transplant course that

included a severe episode of sepsis with septic shock. Her transplant was postponed again because her sibling donor was found to have acute cytomegalovirus (CMV) infection with viremia and upper respiratory infection. Other patient characteristics are listed in table 1.

Engraftment and GvHD

The median total nucleated, CD34+ and CD3+ cell doses of the grafts were $2.7 \times 10^8/\text{Kg}$ ($2.4 - 4.5 \times 10^8/\text{Kg}$), $4.9 \times 10^6/\text{Kg}$ ($3.4 - 5.3 \times 10^6/\text{Kg}$) and $3.3 \times 10^7/\text{Kg}$ ($3.2 - 5.1 \times 10^7/\text{Kg}$), respectively. Neutrophil engraftment occurred on day +14 (range: 12 – 17), and platelet engraftment to $20 \times 10^9/\text{L}$ and $50 \times 10^9/\text{L}$ on days +21 and +23 respectively (range: 20 – 26). There was no evidence of early or late, primary or secondary graft failure/rejection.

Three of the four patients were evaluated for lineage-specific donor chimerism, which was performed on peripheral blood leucocytes, myeloid, T-, B- and NK-cells. As shown in Figure 1, patients 1, 2 and 3 had evidence of 92–100% donor cells in peripheral blood leukocytes, myeloid, B- and NK-cells throughout their time post transplant. However, T-cells were 71% and 53% donor for patient 1 and 2 early post transplant before increasing to 100% donor for both patients by 17–22 months post transplant. For patient 3, T-cells were 100% donor throughout his time post transplant. Data on patient 4 was limited as this patient continued follow-up elsewhere, but showed evidence of 99–100% donor cells by FISH analysis from early to 2 years post transplant.

Patient 3 who had 100% donor T-cells since early post HSCT, developed grade 1–2 acute GvHD involving his liver and lower gastrointestinal tract starting on day +45 post HSCT, and responded to treatment with corticosteroids. At five months post HSCT, he developed chronic GvHD with lymphopenia and polyserositis manifestation, pleural and pericardial effusion. His symptoms were successfully treated with methylprednisolone.

Toxicity, complications and outcome (Table 2)

No patient developed grade 4 non-hematologic acute toxicity during the first 30 days post HSCT. Only patient 4 developed grade 1 mucositis, while the other patients had no mucositis at all. Nausea and vomiting were reported in all patients but only two patients (1 and 2) required total parenteral nutrition support based on clinical status. After completion of equine ATG, patient 1 and 3 developed a rash and fever without identified causative organism. Both patients were successfully treated with broad-spectrum antibiotics and supportive therapy. Patient 3 and 4 required narcotic medications for relief of bone pain (secondary to filgrastim-associated engraftment) and dysmenorrhea, respectively. Cyclosporine was substituted for tacrolimus on day +35 on patient 4 after the development of grade 3 thrombotic microangiopathy. Patient 1 and 4 had grade 3 metrorrhagia requiring medical treatment and transfusions accordingly. There was no episode of invasive fungal infections. No patients developed hemorrhagic cystitis or VOD.

One patient developed coagulase-negative staphylococcus bacteremia, which was reported at 1 month post HSCT and resolved following treatment with broad-spectrum antibiotics. There was no evidence of infections/reactivation of CMV, Epstein-Barr virus, human herpesvirus 6, adenovirus, BK polyomavirus or Toxoplasma gondii in all 4 patients following transplantation.

With a median follow-up of 41.7 months (range: 34 – 47 months), all four patients are alive, and transfusion free. All patients achieved transfusion independence with normalization of peripheral blood counts within 36 days post transplant. The median number of red cell and platelet transfusions received during hospitalization were 2 (range: 1 – 5) and 7 (range: 4 – 19), respectively. The median duration of hospitalization was 27 days (range: 25 – 30 days).

At five months post HSCT, patient 3 was admitted with tachypnea, tachycardia and hypoxia. Work-up revealed evidence of polyserositis with pleural and pericardial effusion. This was assumed to be a rare manifestation of GvHD. His symptoms resolved rapidly after treatment with systemic steroids. At eight months post HSCT, this patient was readmitted with renal insufficiency and a creatinine rising to a maximum of 2.9 mg/dL. The patient eventually responded to the discontinuation of cyclosporine and treatment with immunosuppressive therapy with steroids and mycophenolate mofetil.

DISCUSSION

The preparative or conditioning regimen is an essential component in hematopoietic stem cell transplantation. The three aims of cytoreduction include immuno-ablation, myeloablation and disease eradication. However, when HSCT is performed in patients with SAA, only immuno-ablation is required for engraftment.

Over the past decades, a number of investigators have tried to change the conditioning regimen for the transplantation of patients with SAA to improve outcome and minimize toxicity. Since the 1970's, the major problem with matched related donor HSCT using CY alone at a dose of 200 mg/Kg was a risk of graft rejection that was as high as 30%. [15,16] Storb *et al* demonstrated that the addition of ATG to high dose CY promoted both excellent engraftment and long term outcome, with a lower rate of GvHD. [3,17] However, despite the satisfactory outcome, there remained significant rates of toxicity and infections including bacterial sepsis and fungal infections related to the standard dose of CY.⁵ Similarly, in unrelated donor HSCT, the incidence of graft rejection remained significant as the CY/ATG regimen was found to be insufficient to sustain hematopoietic reconstitution in patients with SAA receiving grafts from unrelated donors. [18] Therefore, TBI was added to the CY/ATG regimen to intensify the immunosuppression effect. Studies from the USA and Japan, using low dose TBI 2–10 Gy, CY 120–200 mg/Kg plus ATG as a conditioning regimen, showed a low risk of rejection at 11%. Yet, the incidence of acute toxicity related to the TBI/CY/ATG regimen was notably high with an increase in lung and kidney injuries. [19,20]

To minimize the toxicity of conditioning regimens while maintaining the immunosuppressive effect, FLU-based conditioning regimens have been used recently in both matched related and unrelated donor HSCT for patients with idiopathic acquired SAA. Kang *et al* reported the outcome of patients who received transplants from HLA-matched unrelated donors following a CY200/ATG plus FLU regimen. All patients developed both transaminitis and microscopic hematuria and the transplant related mortality was high at 32%. [8,11] Though the dose of CY was de-escalated to 120 mg/Kg in several studies, there was still evidence of VOD and invasive fungal infection in almost 10% of the recipients of matched related grafts. [10,21]

In our report, we treated patients with a lower dose of CY at 100 mg/Kg, plus FLU in the conditioning regimen while keeping the same dose of ATG. All patients engrafted without evidence of hemorrhagic cystitis, invasive fungal infection or VOD. Patients developed some nausea and vomiting and transaminitis, but no grade 4 acute toxicities occurred during the first 30 days post transplant. These results show that using a lower dose of CY at 100 mg/Kg plus FLU in the HSCT conditioning regimen for pediatric patients with SAA had minimal toxicity.

Several investigators tried to decrease the transplant toxicity of cyclophosphamide by decreasing the CY dose and adding FLU to the cytoreductive regimen. However the risk of graft failure remained a concern, in particular in the multiply transfused patients with SAA, or those for whom there was a prolonged time interval between diagnosis and transplantation.[22,23] European Group for Blood and Marrow Transplant-SAA working party reported the high incidence of graft failure at 17–18% with the low dose of CY at 40 mg/Kg, ATG plus FLU regimen in alternative donor HSCT.[9,12] As expected, the graft failure rate decreased significantly in recipients of matched sibling transplant using low dose CY 40–120 mg/Kg, ATG plus FLU, reaching the 3–8% level.[10,21,24] In our report, the use of the cytoreductive regimen with 100 mg/Kg of CY with ATG plus FLU led to sustained engraftment in all four patients without evidence of graft failure/rejection. This report included one patient who had received multiple transfusions prior to HSCT and who proceeded to transplant 4 months post diagnosis.

Other investigators evaluated an alemtuzumab-based conditioning regimen together with low dose CY. In a multicenter retrospective study, Marsh et al treated 21 patients using fludarabine 30 mg/m² for 4 days, cyclophosphamide 300 mg/m² for 4 days, and alemtuzumab median total dose of 60 mg (range:40–100 mg), followed by unmodified grafts from HLA-matched siblings. Overall survival at 2 years was 95% but with a 9.5% incidence of graft failure and full donor chimerism only in 42% of patients.[25]

In summary, in this small pilot study, the combination of low dose cyclophosphamide, equine ATG and the addition of fludarabine allowed durable engraftment, with tolerable regimen related toxicity and comparable overall survival in pediatric patients receiving transplants from HLA-matched siblings. The comparison of low dose CY-FLU/ATG-based regimen to the standard regimen using CY200/ATG will need to be further evaluated in a large number of patients to allow a definitive assessment of the optimal conditioning regimen.

Acknowledgments

This research was funded in part through the NIH/NCI Cancer Center Support Grant P30 CA008748. We acknowledge the expert care provided to patients by staff of the Department of Pediatrics at Memorial Sloan Kettering Cancer Center. We thank Joseph Olechnowicz for editorial assistance.

Abbreviation Key

ATG	anti-thymocyte globulin
FLU	Fludarabine

CY	Cyclophosphamide
GvHD	Graft versus host disease
SAA	Severe aplastic anemia
vSAA	Very severe aplastic anemia
HSCT	Hematopoietic stem cell transplantation
TBI	Total body irradiation
VOD	Veno-occlusive disease
IV	Intravenous
BM	Bone marrow
MTX	Methotrexate
HLA	Human leukocyte antigen
ANC	Absolute neutrophil count
STR	Short tandem repeat
FISH	Fluorescence in situ hybridization
RRT	Regimen related toxicity
FA	Fanconi anemia
CMV	Cytomegalovirus
EBV	Epstein-Barr Virus

References

1. Young NS, Calado RT, Scheinberg P. Current concepts in the pathophysiology and treatment of aplastic anemia. *Blood*. 2006; 108(8):2509–2519. [PubMed: 16778145]
2. Gupta V, Eapen M, Brazauskas R, et al. Impact of age on outcomes after bone marrow transplantation for acquired aplastic anemia using HLA-matched sibling donors. *Haematologica*. 2010; 95(12):2119–2125. [PubMed: 20851870]
3. Storb R, Etzioni R, Anasetti C, et al. Cyclophosphamide combined with antithymocyte globulin in preparation for allogeneic marrow transplants in patients with aplastic anemia. *Blood*. 1994; 84(3): 941–949. [PubMed: 8043876]
4. Ades L, Mary JY, Robin M, et al. Long-term outcome after bone marrow transplantation for severe aplastic anemia. *Blood*. 2004; 103(7):2490–2497. [PubMed: 14656884]
5. Storb R, Blume KG, O'Donnell MR, et al. Cyclophosphamide and antithymocyte globulin to condition patients with aplastic anemia for allogeneic marrow transplantations: the experience in four centers. *Biology of blood and marrow transplantation : journal of the American Society for Blood and Marrow Transplantation*. 2001; 7(1):39–44.
6. Lee JH, Choi SJ, Lee JH, et al. Non-total body irradiation containing preparative regimen in alternative donor bone marrow transplantation for severe aplastic anemia. *Bone marrow transplantation*. 2005; 35(8):755–761. [PubMed: 15735661]

7. Kahl C, Leisenring W, Deeg HJ, et al. Cyclophosphamide and antithymocyte globulin as a conditioning regimen for allogeneic marrow transplantation in patients with aplastic anaemia: a long-term follow-up. *British journal of haematology*. 2005; 130(5):747–751. [PubMed: 16115132]
8. Kang HJ, Shin HY, Choi HS, et al. Fludarabine, cyclophosphamide plus thymoglobulin conditioning regimen for unrelated bone marrow transplantation in severe aplastic anemia. *Bone marrow transplantation*. 2004; 34(11):939–943. [PubMed: 15489866]
9. Bacigalupo A, Locatelli F, Lanino E, et al. Fludarabine, cyclophosphamide and anti-thymocyte globulin for alternative donor transplants in acquired severe aplastic anemia: a report from the EBMT-SAA Working Party. *Bone marrow transplantation*. 2005; 36(11):947–950. [PubMed: 16205733]
10. George B, Mathews V, Viswabandya A, et al. Fludarabine and cyclophosphamide based reduced intensity conditioning (RIC) regimens reduce rejection and improve outcome in Indian patients undergoing allogeneic stem cell transplantation for severe aplastic anemia. *Bone marrow transplantation*. 2007; 40(1):13–18. [PubMed: 17450183]
11. Kang HJ, Shin HY, Park JE, et al. Successful engraftment with fludarabine, cyclophosphamide, and thymoglobulin conditioning regimen in unrelated transplantation for severe aplastic anemia: A phase II prospective multicenter study. *Biology of blood and marrow transplantation : journal of the American Society for Blood and Marrow Transplantation*. 2010; 16(11):1582–1588.
12. Bacigalupo A, Socie G, Lanino E, et al. Fludarabine, cyclophosphamide, antithymocyte globulin, with or without low dose total body irradiation, for alternative donor transplants, in acquired severe aplastic anemia: a retrospective study from the EBMT-SAA Working Party. *Haematologica*. 2010; 95(6):976–982. [PubMed: 20494932]
13. Jagasia MH, Greinix HT, Arora M, et al. National Institutes of Health Consensus Development Project on Criteria for Clinical Trials in Chronic Graft-versus-Host Disease: I. The 2014 Diagnosis and Staging Working Group report. *Biology of blood and marrow transplantation : journal of the American Society for Blood and Marrow Transplantation*. 2015; 21(3):389–401. e381.
14. (U.S.) NCI. 2010. Common Terminology Criteria for Adverse Events (CTCAE) Version 4.03. Feb 23, 2017 <http://evsncinhi.gov/ftp1/CTCAE/CTCAE_403_2010-06-14_QuickReference_5x7pdf>. February 23, 2017
15. Storb R, Longton G, Anasetti C, et al. Changing trends in marrow transplantation for aplastic anemia. *Bone marrow transplantation*. 1992; 10(Suppl 1):45–52.
16. McCann SR, Bacigalupo A, Gluckman E, et al. Graft rejection and second bone marrow transplants for acquired aplastic anaemia: a report from the Aplastic Anaemia Working Party of the European Bone Marrow Transplant Group. *Bone marrow transplantation*. 1994; 13(3):233–237. [PubMed: 8199566]
17. Storb R, Leisenring W, Anasetti C, et al. Long-term follow-up of allogeneic marrow transplants in patients with aplastic anemia conditioned by cyclophosphamide combined with antithymocyte globulin. *Blood*. 1997; 89(10):3890–3891. [PubMed: 9160700]
18. Deeg HJ, Anasetti C, Petersdorf E, et al. Cyclophosphamide plus ATG conditioning is insufficient for sustained hematopoietic reconstitution in patients with severe aplastic anemia transplanted with marrow from HLA-A, B, DRB matched unrelated donors. *Blood*. 1994; 83(11):3417–3418. [PubMed: 8193380]
19. Deeg HJ, Amylon ID, Harris RE, et al. Marrow transplants from unrelated donors for patients with aplastic anemia: minimum effective dose of total body irradiation. *Biology of blood and marrow transplantation : journal of the American Society for Blood and Marrow Transplantation*. 2001; 7(4):208–215.
20. Kojima S, Matsuyama T, Kato S, et al. Outcome of 154 patients with severe aplastic anemia who received transplants from unrelated donors: the Japan Marrow Donor Program. *Blood*. 2002; 100(3):799–803. [PubMed: 12130489]
21. Resnick IB, Aker M, Shapira MY, et al. Allogeneic stem cell transplantation for severe acquired aplastic anaemia using a fludarabine-based preparative regimen. *British journal of haematology*. 2006; 133(6):649–654. [PubMed: 16704442]
22. Champlin RE, Horowitz MM, van Bekkum DW, et al. Graft failure following bone marrow transplantation for severe aplastic anemia: risk factors and treatment results. *Blood*. 1989; 73(2):606–613. [PubMed: 2644980]

23. Scheinberg P, Young NS. How I treat acquired aplastic anemia. *Blood*. 2012; 120(6):1185–1196. [PubMed: 22517900]
24. Maury S, Bacigalupo A, Anderlini P, et al. Improved outcome of patients older than 30 years receiving HLA-identical sibling hematopoietic stem cell transplantation for severe acquired aplastic anemia using fludarabine-based conditioning: a comparison with conventional conditioning regimen. *Haematologica*. 2009; 94(9):1312–1315. [PubMed: 19734425]
25. Marsh JC, Gupta V, Lim Z, et al. Alemtuzumab with fludarabine and cyclophosphamide reduces chronic graft-versus-host disease after allogeneic stem cell transplantation for acquired aplastic anemia. *Blood*. 2011; 118(8):2351–2357. [PubMed: 21518925]

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Figure 1.
Lineage-specific donor chimerism analysis post HSCT.

Table 1

Patient and transplant characteristics

Patient	Age (years)/ Sex	Initial counts				Previous Tx (No. units)	Time to HSCt (months)	Donor	CMV status (donor/recipient)	CD34+ cells/Kg	Engraftment Neut Plt	Last Tx RBC Plt	aGvHD/ cGvHD	Outcome
		ANC ($\times 10^9/L$)	Hb (g/dl)	Retic (/μL)	Plt ($\times 10^9/L$)									
1	13/F	0.4	8.6	39000	9	4	4.9	Brother	Neg/Pos	4.6×10^6	$d + 12$ $d + 20$	$d + 12$ $d + 19$	None	Alive 46 months
2	6/M	0.2	9	24000	6	3	0.7	Sister	Neg/Pos	5.2×10^6	$d + 14$ $d + 20$	$d + 17$ $d + 19$	None	Alive 42 months
3	11/M	0.7	7.2	24000	8	8	1.8	Sister	Neg/Neg	3.4×10^6	$d + 15$ $d + 21$	$d + 36$ $d + 20$	grade I-II/ cGvHD	Alive 41 months
4	20/F	0.1	8.6	14000	5	>20	4.1	Brother	Pos/Neg	5.3×10^6	$d + 17$ $d + 26$	$d + 20$ $d + 25$	None	Alive 34 months

Abbreviation: ANC = absolute neutrophil count; Hb = hemoglobin; Retic = reticulocyte count; Plt = platelet count; Tx = transfusion; Neut = neutrophil; RBC = red blood cell; aGvHD = acute graft versus host disease; cGvHD = chronic graft versus host disease

Table 2

Regimen related toxicity (RRT)

Toxicity	Patient 1	Patient 2	Patient 3	Patient 4
Oral mucositis	0	0	0	1
Gingival bleeding	1	0	0	0
Nausea/ Vomiting	3	3	2	1
Epigastric pain	0	0	2	0
SGOT/SGPT	1	0	3	3
Infusion related reaction	1	0	1	0
Fever	3	0	3	0
Headache	0	1	0	1
Bone pain	0	0	2	0
Dysmenorrhea	0	NA	NA	3
Metrorrhagia	3	NA	NA	3
Thrombotic microangiopathy	0	0	0	3

Abbreviations: CTCAE = common terminology criteria for adverse events; SGOT = serum glutamic-oxaloacetic transaminase; SGPT = serum glutamic-pyruvic transaminase; NA = not applicable,