Pure red cell aplasia and Evans syndrome following hematopoietic stem cell transplantation from blood type-matched unrelated donor

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Pure red cell aplasia (PRCA) and Evans syndrome following allogeneic hematopoietic stem cell transplantation (HSCT) from a blood type-matched donor are very rare. A 29-year-old Japanese woman with hematologic remission of Philadelphia chromosome-positive acute lymphoblastic leukemia underwent bone marrow transplantation from a blood type-matched, HLA 6/6-matched, unrelated donor in May 2008. Her clinical course after transplantation was favorable, and allowed for a gradual reduction in tacrolimus dosage. However, 12 months after transplantation, she developed PRCA related to allogeneic HSCT. The patient's PRCA was alleviated by stopping the dosage reduction for the immunosuppressive therapy and by continued administration of low-dose tacrolimus. Three months after the PRCA diagnosis, the patient developed Evans syndrome, so prednisolone (PSL) treatment was initiated. PSL treatment effectively treated the Evans syndrome, as was evident by the absence of cythemolysis two months later, and no relapse has occurred even after the PSL dose was reduced and then terminated. The fact that PRCA onset in this case occurred after blood type-matched allogeneic HSCT, during the late grafting phase, and in combination with Evans syndrome, suggests that the pathogenic mechanism may differ from that of previously reported cases of PRCA following blood type-incompatible allogeneic HSCT. (*Journal of Hematopoietic Cell Transplantation* Vol. 1 No. 1; 24–28, 2012)

Background

Pure red cell aplasia (PRCA) and autoimmune hemolytic anemia (AIHA) are rare complications that follow allogeneic hematopoietic stem cell transplantation (HSCT).^{1–3} PRCA has occurred after blood type-mismatched allogeneic HSCT, in which case the remaining isohemagglutinin in the recipient plasma is thought to suppress erythropoiesis.⁴ With regard to AIHA, continuous suppression of cellular immunity after allogeneic HSCT is thought to affect humoral immunity, yielding the production of autoantibodies which contribute to hemolysis.^{2,5,6} Here we report a case of PRCA followed by Evans syndrome that developed after unrelated donor allogeneic HSCT from a blood type-matched, human leukocyte antigen (HLA) 6/6-matched donor.

Case Report

The patient was a 29-year-old Japanese woman without remarkable medical or family histories. She developed Philadelphia chromosome (Ph)-positive acute lymphoblastic leukemia in December 2007. She achieved hematologic remission for which mRNA levels of minor BCR-ABL fusion protein were 61 copies/ μ g total RNA after one course of hyper-CVAD therapy. Two courses of hyper-CVAD were combined with imatinib, but molecular remission was not achieved (10 copies of BCR-ABL mRNA/ μ g total RNA). Using a conditioning regimen of total body irradiation (TBI 12 Gy) with etoposide (VP-16) (15 mg/kg/2 days) and cyclophosphamide (CY) (60 mg/kg/2 days), and immunosuppression with tacrolimus and short-term methotrexate (MTX), bone marrow transplantation from a

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blood type-matched (B, Rh +), HLA 6/6-matched, unrelated donor was performed in May 2008. Engraftment was successful with neutrophil recovery on day 17 after the transplant and engraftment of all three cell lineages on day 28. Bone marrow examination on day 29 showed chimerism with donor cells of at least 90% and negative for minor BCR-ABL mRNA. Acute graft-versus-host disease (GVHD) symptoms appeared on the skin (stage 1–2) on days 9–21 and 42–53, but they were successfully treated with tacrolimus alone. Her subsequent clinical course was favorable with a gradual reduction of tacrolimus dosage during follow-up.

In March 2009, the patient experienced cold-like symptoms of headaches and shortness of breath. Blood examination on May 27, 2009 revealed severe anemia with reticulocytopenia (RBC, $194 \times 10^4/\mu\text{l}$, Hb 5.9 g/dl, Ht 18.1%, reticulocyte count less than 0.1%) (Table 1). Bone marrow examination revealed normal cellularity and normal megakaryocyte count on clot section. In differentiation, marked erythroid hypoplasia with erythroid lineage of 2.5% (myeloid/erythroid [M/E] ratio, 36.5) was found.

Morphologically, no dysplasia was seen. Chromosomal analysis revealed normal karyotype. Late-onset rejection or relapse was ruled out as the donor chimerism was more than 90% and minor BCR-ABL mRNA was negative, and thus the patient was diagnosed with PRCA. Parvovirus B19 infection was ruled out because anti-parvovirus B19 antibody levels were not elevated and PCR analysis was negative (Table 1). Drug-induced PRCA was also ruled out because no medications were added prior to PRCA onset. No chronic GVHD symptoms were identified. Combined, this led us to believe that PRCA was related to allogeneic HSCT, and suggested that AIHA was not found (Table 1). Low-dose tacrolimus (1 mg/day) was continued without dose reduction and followed up with red blood cell transfusions. Erythrocyte recovery (Hb, 11.8g/dl; reticulocyte count, $2.39 \times 10^4/\mu\text{l}$) was spontaneously observed on July 22, 2009 (Figure 1).

On August 11, 2009, petechial subcutaneous hemorrhages began to appear on the lower extremities, but we did not identify infection symptoms or GVHD. Blood tests revealed an RBC count of $285 \times 10^4/\mu\text{l}$, Hb 8 g/dl, Ht of 24%,

Table 1. Laboratory findings at pure red cell aplasia onset

WBC	5200/ul	Na	140mEq/l	Mg	2.3mg/dl
Stab	0.5%	K	5.2mEq/l	Cu	160ug/dl
Seg	43.5%	Cl	104mEq/l	Zn	82ug/dl
Eos	10.5%	UA	6.4mg/dl	Fe	151ug/dl
Baso	0.5%	BUN	15.6mg/dl	UIBC	148ug/dl
Mono	2%	Cr	0.79mg/dl	Ferritin	670ng/ml
Ly	42.5%	TP	7.5g/dl	Folic acid	19.1ng/ml
RBC	194/ul	Alb	3.7g/dl	Vit B12	987pg/ml
Hb	5.9g/dl	Alb	58.9%		
Ht	18.1%	$\alpha 1$	3%	CMV-C10/C11	0/0
Plt	143/ul	$\alpha 2$	10.4%		
Ret	<1%	β	7.9%		
		γ	19.8%	Parvo B19	
GOT	15IU/l	BS	109mg/dl	IgG	2.06
GPT	19IU/l	CRP	0.15mg/dl	IgM	0.33
LDH	201IU/l			PCR	negative
ALP	363IU/l	IgG	1523mg/dl		
r-GTP	48IU/l	IgA	408mg/dl	Anti-Ce	positive
T-Bil	0.2mg/dl	IgM	142mg/dl	Anti-Ce Ab	

Vit B12, Vitamin B12; CMV-C10/C11, CMV-Antigenemia C10/C11; Anti-Ce Ab, Anti-Ce Antibody

Figure 1. Clinical course of pure red cell aplasia complicated by Evans syndrome. Hb, hemoglobin; LDH, lactate dehydrogenase; Plt, platelet count; PSL, prednisolone; Ret, reticulocyte count. We diagnosed PRCA after allogeneic HSCT on May 27, 2009, and AIHA after allogeneic HSCT on August 11, 2009.

reticulocyte count of $4.68 \times 10^4/\mu\text{l}$, platelet count of $2.9 \times 10^4/\mu\text{l}$ and lactate dehydrogenase (LDH) level of 203 IU/L. Bone marrow examination revealed erythroblastic and megakaryocytic hyperplasia. Direct and indirect Coombs tests were positive (anti-IgG +, anti-C3b/C3d +, pan-reactive eluate +), but cold agglutinine titration was normal. Antinuclear antibodies were positive (320-fold with homogeneous pattern), but anti-dsDNA IgG were negative. Platelet-associated immunoglobulin G (PAIgG) increased to 2940 ng/ 10^7 cells. Given these results, Evans syndrome was diagnosed, and prednisolone (PSL) therapy (0.5 mg/kg/day) was started. After PSL therapy, Evans syndrome disease activity decreased gradually (Figure 1). PSL was terminated in February 2011, and no relapse of PRCA or Evans syndrome has been observed to date.

Discussion

We report our experiences with a 29-year-old Japanese female patient who presented with PRCA and Evans syndrome following HSCT from a blood type-matched, HLA6/6-matched unrelated donor. Most reported cases of PRCA following allogeneic HSCT are attributed to blood type-mismatched donors (major mismatch in particular), and

cases of PRCA after transplantation from a blood type-matched donor are very rare.^{1,4,6} On the other hand, AIHA after allogeneic HSCT was reported to occur in 4.5% of adult cases and as risk factors, unrelated donors, HLA-mismatches, and chronic GVHD were pointed out.^{2,3} Few cases of Evans syndrome after HSCT have been reported. One patient received a HSCT from a T-cell-depleted unrelated donor,⁷ and another patient was treated with a conditioning regimen of antithymocyte globulin (ATG), which could have over-suppressed T cells, leading to abnormalities in humoral immunity.⁸ Some reports have noted a combination of PRCA with AIHA or Evans syndrome in patients with malignant lymphoma⁹ and after allogeneic HSCT.⁶ To our knowledge, no one has reported a case of PRCA and Evans syndrome following HSCT. However, a combination of PRCA with AIHA or Evans syndrome has been observed in 7 of 357 cases (2.0%) of pancreas transplantation using immunosuppression with alemtuzumab, mycophenolate mofetil, and daclizumab.¹⁰ One factor that may be involved in AIHA and Evans syndrome as well as PRCA is an immune abnormality brought about by excessive immunosuppression, but the causative mechanisms of this remain largely unknown.

The case study did not use drugs such as ATG for the

patient's conditioning regimen, yet the clinical course after transplantation was favorable, yielding only mild GVHD under mild immunosuppression with tacrolimus and MTX. Pathogenesis of PRCA and Evans syndrome after HSCT in the present case was unclear. Given that cold-like symptoms were noted two months before PRCA onset, it is still plausible that a viral infection somehow caused PRCA. In the clinical course of the present case, we observed no clear cythemolysis findings at the time of PRCA onset, and the reactivity to erythrocyte transfusions was good, and at the same time, the platelet count began to decrease gradually (Figure 1). Although the Coombs' test was not performed at the time of PRCA onset, it is possible that the Evans syndrome occurred simultaneously with PRCA. Considering the alleviation of PRCA after that, cythemolysis might be attributed to the Evans syndrome.

One effective treatment for PRCA that develops after blood type-incompatible allogeneic HSCT is to reduce or terminate the immunosuppressant administration, and reduce the recipient-derived plasma cells that produce isohemagglutinin via the graft-versus-plasma cell effect.^{11,12} However, if this treatment does not yield improvement, administration of ATG or steroids, i. e., treatments that strengthen immunosuppression, can also be effective.^{4,13,14} Some reports have noted that performing donor lymphocyte infusion and causing the graft-versus-plasma cell effect can conversely be effective in such cases.^{4,15} In addition, some have reported the efficacy of plasma exchange or rituximab, and thus there is no single treatment strategy.⁴ In the present case, PRCA went into spontaneous remission when the tacrolimus dosage reduction was terminated and administration was maintained at a low dosage. Because Evans syndrome subsequently developed and PSL administration was initiated, however, it is not clear whether low dose tacrolimus was effective.

The fact that PRCA onset in this case occurred after blood type-matched allogeneic HSCT, during the late grafting phase, and in combination with Evans syndrome, suggests that the pathogenic mechanism may differ from that of previously reported cases of PRCA following blood type-incompatible allogeneic HSCT.

Consent

Written informed consent was obtained from the patient in order to publish this Case report.

Competing interests

The author(s) declare that they have no competing interest.

Author contributions

HY was responsible for manuscript preparation and was involved in patient care. KN, KM, KN, KI, KT and KD were responsible for patient care and manuscript review. All authors read and approved the final manuscript.

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