Cyclophosphamide and antithymocyte globulin conditioning may be sufficient for Korean patients with early stage severe aplastic anemia transplanted with marrow from donors other than HLA-identical siblings

We gave a regimen of cyclophosphamide and antithymocyte globulin (CY/ATG) to six patients with early stage severe aplastic anemia (SAA) transplanted with marrow from alternative donors. All patients engrafted and are alive with durable engraftment at a median follow-up of 406 days. The CY/ATG regimen may be sufficient in Korean patients with early stage SAA receiving marrow transplantation from alternative donors.

A cyclophosphamide (CY) and antithymocyte globulin (ATG) conditioning regimen, which has been successful in bone marrow transplantation (BMT) for severe aplastic anemia (SAA) using an HLA-identical sibling donor,[1] was reported to be insufficient for alternative donor transplants in SAA patients of Western countries.[2] The major problem was graft rejection. The rates of graft rejection and graft-versus-host disease (GVHD) after allogeneic BMT including unrelated donor transplants seemed to be lower in reports from Asian countries of the Far East.[3-6] We applied a CY/ATG regimen to six patients with early stage SAA transplanted with marrow from donors other than HLA-identical siblings.

Five patients received allogeneic marrow grafts from unrelated donors, and one from an HLA-phenotypically one major locus mismatched sibling donor. Among five donor-recipient pairs of unrelated transplants, three were HLA-phenotypically matched, one had one major locus mismatch, and one had one minor locus mismatch. The matching of donors and recipients was based on serological typing of HLA A, B, and DR. The conditioning regimen was CY (50 mg/kg/d × 4) plus ATG (30 mg/kg/d × 3) in all patients except one (UPN 120) who had had an anaphylactic reaction to ATG prior to BMT and received fludarabine (30 mg/m²/d × 3) in the place of ATG. All patients received prophylactic therapy with cyclosporine and methotrexate for GVHD. Acute and chronic GVHD were classified according to standard criteria.[7,8] Hematopoietic chimerism was evaluated in all patients, using peripheral blood samples from the donor and the recipient, by polymerase chain reaction (PCR) amplification of short tandem repeats or amelogenin loci.[9]

The pre-transplant characteristics of the patients and transplant outcomes are detailed in Table 1. The median age at BMT was 20 years. One patient (UPN 136) had SAA/paroxysmal nocturnal hemoglobinuria syndrome. Two patients (UPNs 101 and 131) had pre-transplant neutrophil counts below 200/μL. The median interval from diagnosis to BMT was 4.0 months. Two patients (UPNs 120 and 145) had received previous therapy with ATG (± CSA). All had received transfusions. All six patients engrafted and are alive with durable engraftment at a median follow-up of 406 days (range, 328 to 643). All patients achieved an absolute neutrophil count (ANC) over 1,000/μL, unsupported platelet count over 100,000/μL, reticulocyte count over 1%, and RBC transfusion independence. All patients achieved complete donor chimerism documented by hematopoietic chimeric analysis (Table 2). UPN 130 had defective glycosylphosphoinositol (GPI)-anchored proteins (CD55 and CD59) on circulating granulocytes and red blood cells by flow cytometric analysis before BMT. Flow cytometric findings normalized after BMT (day 47, 60 and 231). This patient experienced pancytopenia from post-transplant day 57 and showed persistent mixed chimerism. However, pancytopenia spontaneously recovered on day 87 and complete donor chimerism was achieved 8 months post-transplant. UPN 101 showed transient appearance of cells of recipi-

Table 1. Pre-transplant patient characteristics and transplant outcomes.

UPN	Sex/Age	Dx	Pre-transplant blood cell count			Previous transfusion (unit)		Time to BMT from Dx (mos.)	Previous therapy	Type of donor	HLA locus mismatch	Time to engraftment (post-transplant day)			GVHD		F/U (days)
			cReti	PLT	ANC	RBC	PLT					ANC > 500	PLT > 20	Reti > 1%	Acute	Chronic	
101	M/15	SAA	0.06	5	112	6	180	3.8	None	Unrelated	None	18	27	26	Gr. I	Lim	643+
120	M/23	SAA	1.27	17	434	17	53	55.0	ATG	Unrelated	DR (major)	25	32	32	None	Lim	503+
130	M/28	SAA	0.02	12	72	12	66	1.6	None	Sibling	B (major)	15	21	22	None	Lim	426+
136	F/17	PNH / SAA	0.46	18	975	8	10	4.2	None	Unrelated	None	20	23	40	Gr. III	Ext	386+
141	F/15	SAA	0.51	16	394	6	6	2.6	None	Unrelated	None	21	26	30	None	None	359+
145	M/25	SAA	0.47	17	520	14	298	12.7	ATG/CSA	Unrelated	B (minor)	21	29	29	Gr. III	Ext	328+

UPN= unique patient number; Dx = diagnosis; ms; = months; SAA = severe aplastic anemia; PNH = paroxysmal nocturnal hemoglobinuria; cReti = corrected reticulocyte count (%); PLT = platelet count (× 10³/μL); ANC = absolute neutrophil count (/μL); ATG = antithymocyte globulin; CSA = cyclosporine A; Reti = reticulocyte count (%); GVHD = graft-versus-host disease; Gr = grade; Lim = limited; Ext = extensive; F/U = follow-up duration.

Table 2. Results of assays of hematopoietic chimerism.

UPN	1 mo	2 mo	3 mo	4 mo	5 mo	6 mo	7 mo	8 mo	9 mo	12 mo	15 mo	18 mo	21 mo
101	CC	CC	CC	CC	CC	CC	CC			8%*	6%*	17%*	CC
120	CC	CC	CC	CC					CC	CC	CC		
130	CC	CC	CC			CC			CC	CC			
136	10%*	5%*	13%*	37%*	20%*	8%*	<5%*	CC	CC	CC			
141	10%*	CC	<5%*	CC	CC			CC					
145	CC	CC	CC			CC			CC				

**The proportions of recipient cell population. UPN = unique patient number; mo = months post-transplant ; CC = complete donor chimerism.*

ent origin between 12 and 18 months after BMT, but this patient maintained normal peripheral blood counts during the period of mixed chimerism. Three patients developed acute GVHD. One patient had grade I GVHD and two had grade III GVHD. Chronic GVHD occurred in five patients, but immunosuppressive treatment could be tapered off in four patients.

Our promising results could be considered under two aspects. Firstly, our patients were transplanted in a relatively early stage of the disease. Earlier use of BMT, when one has a matched sibling donor, in order to decrease transfusion-induced sensitization has led to increased survival.[10] A similar advantage may be present in the setting of BMT using alternative donors. Among our six patients, four had an HLA-matched unrelated donor or an HLA one-locus mismatched sibling donor and underwent BMT at 1.6 to 4.2 months after diagnosis of SAA. The other two patients had no matched unrelated donor and they were transplanted with marrow from HLA one-locus mismatched unrelated donors after IST failed. Early transplantation in the course of disease might have contributed to the favorable clinical outcomes in our patients. Secondly, a lesser degree of genetic diversity in histocompatibility antigens may be present among Koreans.[3] Adverse effects of the HLA barrier, such as graft rejection and GVHD, have been suggested to be somewhat different between populations of Western countries and Far Eastern Asian countries.[3-6]

In conclusion, a CY/ATG regimen may be sufficient in Korean patients with early stage SAA transplanted with marrow from alternative donors. Although the number of patients in the present study is small, the results are encouraging. A search for an unrelated donor should be undertaken early in SAA patients who lack an HLA-identical sibling, and stem cell transplantation using alternative donors may be considered at an earlier stage of disease.

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Key words: SAA; allogeneic BMT; alternative donor.

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