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Case Report

Unrelated Cord Blood Transplantation Using a Reduced-Intensity Conditioning Regimen without Total Body Irradiation in Two Patients with Multiple Myeloma

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Reduced-intensity unrelated cord blood transplantation was performed on two patients (a 55-year-old woman and a 52-year-old man) with multiple myeloma that had progressed after high-dose chemotherapy (melphalan : 200 mg/m^2) with autologous stem cell transplantation support. The conditioning regimen consisted of fludarabine (180 mg/m^2) and busulfan (8 mg/kg) without total body irradiation. Tacrolimus was administered as a graft-versus-host disease prophylaxis. The engraftments were rapid (day +17 in patient 1 and day +26 in patient 2). Regimen-related toxicity was tolerable and acute graft-versus-host disease (grade I) appeared only in patient 2. Complete donor chimerism continued following the treatment and no disease progression was observed in the succeeding 12 months. These results demonstrate the feasibility and effectiveness of reduced-intensity cord blood transplantation after autologous stem cell transplantation in older patients.
Key words multiple myeloma, reduced-intensity unrelated cord blood transplantation, total body irradiation

INTRODUCTION

Multiple myeloma (MM) is difficult to cure with conventional chemotherapy using melphalan and prednisolone. Although high-dose chemotherapy with autologous stem cell transplantation (ASCT) support is more effective than conventional chemotherapy in terms of the response rate and the five-year overall survival rate, high frequencies of relapse and disease progression were still observed, and the survival curve did not plateau even when the treatment accompanied tandem transplantation[1,2]. Allogeneic stem cell transplantation has been reported to have a graft-versus-myeloma (GVM) effect and at present is the only therapy that has been shown to cure MM. Despite restricting allogeneic stem cell transplantation to younger patients, the transplantation-related mortality (TRM) and overall survival rate, however, have not been favorable[3,4]. Reduced-intensity stem cell transplantation (RIST) consists of a reduced-intensity conditioning regimen, which focuses on immunosuppression rather than on myeloablation. It has the advantage of reducing regimen-related toxicity (RRT) and can be performed on older patients or patients who have disease- and treatment-related complications[5]. Moreover, preferable outcomes resulting from RIST following ASCT have been reported[6-8].

Recently, the feasibility of unrelated cord blood transplantation (UCBT) in adults as well as in children has been shown[9]. Reduction in the severity of graft-versus-host disease (GVHD) and outcomes similar to those observed with allogeneic bone marrow transplantation (BMT) have also been reported[10,11]. Because HLA compatibility and the severity of GVHD are not correlated, donor selection is easier than that for unrelated bone marrow grafts. Additionally, frozen cord blood grafts are more readily available than bone marrow grafts.

The effectiveness of RIST on MM using unrelated cord blood (reduced-intensity unrelated cord blood transplantation : RI-UCBT) has been shown[12-14]. The scale of these RI-UCBT studies, however, was small and the long-term effects of this treatment remain ambiguous. Moreover, the make-up of optimum conditioning regimen that attains minimum RRT, minimum GVHD, and a high rate of engraftment is controversial.

Here, we describe two cases of refractory MM after ASCT in which UCBT using a reduced-intensity conditioning regimen without total body irradiation (TBI) was performed with no observable disease progression in the 12 months succeeding treatment.

PATIENT 1

Patient 1 was a 55-year-old woman whose father died of

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MM. In April 1998, serum monoclonal paraprotein (IgGκ) was detected and she was diagnosed with monoclonal gammopathy of undetermined significance. In August 2003, her serum IgG level was found to be elevated and a diagnosis of MM (stage IIIA) was made. She received three cycles of VAD therapy (vincristine, adriamycin, and dexamethasone) starting one month following the diagnosis. A high dose of melphalan (200 mg/m^2) was administered intravenously followed by autologous peripheral blood stem cell transplantation in December 2003. Though serum paraprotein was not detected by immunofixation, disease progression was observed in the bone in February 2004. Radiographic studies revealed a lytic bone lesion in the diaphysis of the patient's right humerus, to which we delivered radiation therapy. She was admitted to our hospital on April 6, 2004 for RI-UCBT. At the time of admission, hematologic and serum biochemistry were as follows : white blood cell count 3100 cells/μl, hemoglobin 9.6 g/dl, platelet count 1.01 × 10^5 cells/μl, total protein 6.9 g/dl, creatinine 0.70 mg/dl, β_2-microglobulin 3.0 mg/l, IgG 2079 mg/dl, IgA 141 mg/dl, and IgM 49 mg/dl. A bone X-ray showed a punched-out lesion in the diaphysis of the right humerus. Serum monoclonal paraprotein was not detected. Fluorescence *in situ* hybridization showed that the chromosomes of the myeloma cells, including chromosome 13, were normal.

The patient received a dose-reduced conditioning regimen and cord blood was infused on April 16, 2004 (day 0 ; Fig. 1). The infusion contained 2.3 × 10^7 cells/kg body weight and two of the six HLA loci were mismatched. Neutrophil engraftment was achieved on day +17 and the last platelet transfusion was given on day +40. Full donor chimerism of peripheral blood T lymphocytes and bone marrow cells was confirmed on day +28. Apart from myelosuppression, nausea

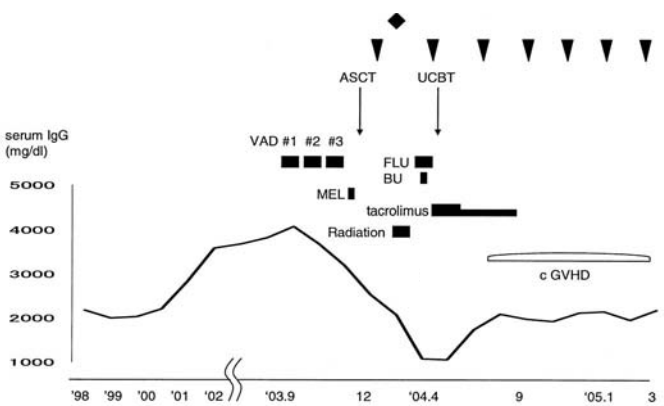

Fig. 1 The clinical course of patient 1.
Abbreviations : VAD, vincristine, adriamycin, and dexamethasone ; MEL, melphalan ; FLU, fludarabine ; BU, busulfan ; cGVHD, chronic GVHD associated with limited-type skin rash and dry mouth ; arrowheads, no serum paraprotein was detected by immunofixation ; diamond, an osteolytic lesion appeared.

(NCI-CTC, grade 1) was the only indication of RRT to occur. On day +11, we observed a fever of 38℃ and facial swelling that lasted a few days. We judged these symptoms to be indicative of pre-engraftment syndrome. Acute GVHD was not observed. Herpetic encephalitis occurred on day +45 and was treated with acyclovir. The patient was discharged on day +89. Administration of tacrolimus continued until day +145. Chronic GVHD included rash (limited type) and dry mouth, which were managed with steroid ointment and artificial saliva. The patient developed a varicella-zoster virus infection in her skin on day +225, and acyclovir was administered. In the 12 months following RI-UCBT, no disease progression was observed.

PATIENT 2

Patient 2 was a 52-year-old man with type II diabetes. The patient was diagnosed with MM (Bence-Jones type, stage IIIA) when a pathologic fracture of the thoracic spine developed. The patient received three cycles of VAD therapy starting in June 2003, while local radiotherapy was performed simultaneously. A high dose of melphalan (200 mg/m^2) was administered intravenously followed by autologous peripheral blood stem cell transplantation in November 2003. He received interferon α (3 × 10^6 units, three times per week) as maintenance therapy. However, because Bence-Jones protein continued to be detected and disease progression was observed throughout the bones, additional radiation therapy was given. We decided to perform RI-UCBT because the physical performance of the patient decreased due to multiple pathologic fractures and an HLA-matched bone marrow graft was unavailable. The patient was admitted to our hospital on April 17, 2004. At the time of admission, hematologic and serum biochemistry were as follows : white blood cell count 3300 cells/μl, hemoglobin 10.2 g/dl, platelet count 6.6 ×10^4 cells/μl, total protein 6.9 g/dl, creatinine 1.02 mg/dl, β_2-microglobulin 3.9 mg/l, IgG 1897 mg/dl, IgA 138 mg/dl, and IgM 104 mg/dl. A bone X-ray showed multiple fractures and lytic lesions. Urine was positive for Bence-Jones protein by immunofixation. Fluorescence *in situ* hybridization showed that the chromosomes of the myeloma cells, including chromosome 13, were normal.

The patient received a reduced-intensity conditioning regimen and cord blood was infused on April 23, 2004 (day 0). The infusion contained 2.8 × 10^7 cells/kg body weight and two of the six HLA loci were mismatched. Neutrophil engraftment was achieved on day +26 and the last platelet transfusion was given on day +53. Full donor chimerism of peripheral blood T lymphocytes and bone marrow cells was confirmed on day +28. Apart from myelosuppression, nausea and fatigue (NCI-CTC, grade 1) were the only signs of RRT to occur. Pre-engraftment fever, which lasted for a few days, was observed on day +11. Acute GVHD appeared in the

patient's skin (stage 1). The patient was discharged on day +49. Administration of tacrolimus was discontinued on day +67. Chronic GVHD was not observed. The patient developed a varicella-zoster virus infection in his skin on day +95 and was successfully treated with acyclovir. Bence-Jones protein was not detected in the urine after day +100. In the 12 months following RI-UCBT, no disease progression was observed.

METHODS

To perform RI-UCBT, fludarabine (30 mg/m^2, from day − 8 to day − 3) and busulfan (4 mg/kg, from day − 6 to day − 5) were administered intravenously and orally, respectively. Cord blood (HLA compatibility of more than four of the six loci and nucleated cells of more than 2.0×10^7 cells per kg of the recipient's body weight) was transfused on day 0. Tacrolimus (0.12 mg/kg/day, twice a day starting on day − 1) was administered orally as a prophylaxis for GVHD. A granulocyte colony-forming unit (filgrastim : 300 μg/day) was used from day +6 until neutrophil engraftment. Ciprofloxacin, itraconazole, and acyclovir were administered orally as prophylaxes for infection. Both patients provided written informed consent.

DISCUSSION

We performed RI-UCBT on two patients with MM that had progressed after ASCT. In both cases, transplantation using a traditional myeloablative conditioning regimen was thought to be inappropriate due to the age and decreased physical performance of each of the patients. Moreover, we used cord blood due to the unavailability of suitable bone marrow grafts. Rapid neutrophil engraftment was achieved. Serum paraprotein was not detected and no disease progression was found in the 12 months following RI-UCBT. RRT as well as acute and chronic GVHD were tolerable and the period of hospitalization was shorter than that for traditional myeloablative transplantation.

At present, allogeneic stem cell transplantation is the only therapy that has been shown to cure MM. Some reports have suggested the existence of a GVM effect in allogeneic transplantation[13]. Since MM tends to occur in the elderly, application of transplantation with a myeloablative conditioning regimen, however, has been restricted to a minority of the affected patients. Moreover, the associated overall survival and progression-free survival rates did not exceed those of ASCT due to a high TRM[3,4]. RIST, which consists of a reduced-intensity conditioning regimen, has as one of its benefits a reduction of TRM and can be performed in older patients or patients who have disease- and treatment-related complications[5-8]. Malony *et al*.[7] reported about 54 cases of MM that were treated with RIST using total body irradiation (2 Gy ± fludarabine) following ASCT. Engraftment was attained in all of the cases and there was one TRM. The estimated progression-free survival rate for all of the patients at two years was 55%. Kroger[8] performed RIST consisting of fludarabine and melphalan ± anti-thymocyte globulin (ATG) on 120 MM patients. In this study, engraftment was rapid, the TRM rate was 18%, the two-year overall survival rate was 59%, and the two-year progression-free survival rate was 39%. Both studies support the effectiveness and feasibility of RIST.

Although the conditioning regimens of many other studies consisted of TBI or ATG in addition to fludarabine and an alkylating agent[16], the regimen in this study consisted of only fludarabine and busulfan. Yanada[17] *et al*. have reported that a conditioning regimen using TBI correlated with a higher risk of TRM and a lower overall survival rate for older patients. Tissue destruction and the high level of inflammatory cytokines caused by TBI possibly predispose patients to acute GVHD[18]. ASCT followed by RIST may reduce the tumor burden and induce immunosuppression ; as a result, engraftment can be promoted and the severity of GVHD can be reduced[6-8]. In our study, the consistently tolerable RRT, rapid engraftment, and mild GVHD despite a lack of TBI or ATG might be due to ASCT being performed before RIST.

The major advantage of using an unrelated cord blood graft is the lower rates of acute and chronic GVHD than those of unrelated BMT (UBMT)[9-12]. Since HLA compatibility and the severity of GVHD are not correlated, HLA-mismatch transplantation can be done without the need for intensive immunosuppression. According to Rocha[11], UBMT and UCBT achieved similar results in terms of relapse after day

Table 1. RI-UCBT following ASCT : a review of the literature

Reference	Diagnosis	Number of cases	Conditioning regimens	GVHD prophylaxis	Acute GVHD	Progression free survival (months)
Yamada *et al*. [13]	MM	2	TBI(2 Gy), fludarabine, melphalan	mycophenolate mofetil, cyclosporine	grade I and II	more than 6 and 16
Ando *et al*. [14]	MM	1	TBI(4 Gy), fludarabine, melphalan	cyclosporine, methylprednisolone	none	4.5
This study	MM	2	fludarabine, busulfan	tacrolimus	none and grade I	more than 12

GVHD : graft-versus-host disease, MM : multiple myeloma, TBI : total body irradiation

100 post-transplant. Patients that received UCBT had a decreased rate of acute GVHD despite the number of HLA mismatches being higher than that of the UBMT group. Other benefits of using cord blood are that cord blood grafts are more readily available than bone marrow grafts and the risk to the donor is small. The high frequency of graft failure and impossibility of donor lymphocyte infusion (DLI) have been previously discussed. Using a sufficient amount of cord blood cells, however, can reduce the risk of graft failure[11]. As for DLI, unlike in chronic myeloid leukemia, its efficacy in MM has not been shown and therefore it is not thought to be a major problem[10].

Although the results of this study demonstrated the feasibility of RI-UCBT in terms of its low RRT, tolerable GVHD, and effectiveness, the sample size was small and the long-term outcome of this treatment was not investigated. A large-scale study is required to further investigate the optimum conditioning regimen, the optimum levels of immunosuppression, the safety of the treatment, and the long-term outcome resulting from RI-UCBT. For the time being, RI-UCBT should be performed in older patients with MM that is refractory or relapses after conventional chemotherapy or ASCT. After establishing the safety and effectiveness of RI-CBT, this treatment can be extended as a first-line therapy in combination with ASCT.

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