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Allogeneic Peripheral Blood Stem Cell Transplantation for Aplastic Anaemia: a Single Centre Experience

T. Farzana, T. S. Shamsi, M.Irfan, S. H. Ansari, M. I. Baig, N. Shakoor
Bismillah Taqee Blood Diseases Centre, Karachi.

Introduction

Aplastic anaemia is a disorder characterized by peripheral blood pancytopenia associated with hypocellular bone marrow trephine biopsy in the absence of any dysmorphic precursors or marrow infiltration. In Pakistan, idiopathic aplastic anaemia is the most common type of aplastic anaemia affecting mainly male population.1 It is associated with immune mediated destruction of haematopoietic stem cells.2 Allogeneic stem cell transplantation offers a 60-80% chance of cure in young patients who have a HLA identical sibling donor.3,4 However, patients without a donor or who are older; immunosuppression remains the only option. Graft rejection, and graft versus host disease have been major complications related to allogeneic transplant in the past. Lately, improved immune suppression protocols have resulted in a decline in graft failures with better outcome.5 Autologous peripheral blood stem cell transplantation has replaced bone marrow transplantation during last 15 years.6 Published data suggests that in allogeneic PBSC, there is rapid engraftment, lesser blood component usage, early hospital discharge and lower incidence of acute GvHD.7-9 Randomised studies comparing PBSC vs. BMT in allogeneic setting clearly show faster haemopoietic and immune recovery and reduced relapse rate in PBSC groups.10,11 However, these studies did not show significant difference in acute and chronic graft versus host disease in two groups. In Pakistan, until recently immunosuppression was the only modality of treatment available for these patients. During the last 3 years peripheral blood stem cell transplantation programme was started in our centre. Here we describe our experience in 20 cases of aplastic anaemia.

Patients and Methods

Twenty-two patients with aplastic anaemia received allogeneic peripheral blood stem cell transplantation at Bismillah Taqee Blood Diseases Centre, Karachi. Patients' characteristics are shown in the Table. Conditioning regimen consisted of cyclophosphamide 50 mg / kg /day for 4 days given to every patient except one patient with pure red cell aplasia who received busulphan 3.5 mg/kg/day for 4 days followed by cyclophosphamide 40 mg/kg/day for 4 days. Two Patients who previously failed immunosuppressive treatment and were heavily transfused received ALG 15 mg/kg/day for 4 days as well. Donors were primed with 10 mcg/kg of G-CSF for 4 days. Peripheral blood stem cells were harvested on 5th day using Haemonetics MCS+ cell separator. Ante-cubital vein was used for stem cell aphaeresis. None of the donors required a central line. Median aphaeresis time with this machine was 250 minutes (180 - 300 minutes) depending upon the volume and rate of blood flow from the donor. Mononuclear cell yield was sufficient with single aphaeresis. Mononuclear cell dose of >4 x108/kg body weight of recipient was given to patients. For GvHD prophylaxis, oral cyclosporin 5 mg/kg/day was started on day -5 so that therapeutic levels are achieved prior to transplant. The dose was adjusted depending on whole blood cyclosporin level. Oral methotrexate 10 mg/kg was given on day +1, +3, + 6, +11 and then weekly for up to 6 months while cyclosporin was continued for 12 months. MMF was added to patients who received ATG. All patients received injection G-CSF 5 mcg/kg/day subcutaneously starting on day +4. All patients were kept in isolation rooms with facilities of HePa air filtration unit and reverse barrier nursing. Leucodepleted blood and platelets were

given as required. Neutrophil engraftment was defined as the first of consecutive 3 days of absolute neutrophil count >0.5x109/l while for platelets, it is defined as the first of 7 consecutive days with an untransfused platelet count >20x109/l. GvHD was diagnosed clinically and graded as described previously.12 After discharge from the hospital, patients were followed up in the clinic twice a week during first 60 days post transplant, once a week till first 100 days and then fortnightly to monthly thereafter. In case of an untoward event they were instructed to report immediately. Patients and parents were instructed to keep a record of temperature, cough, skin rash, dysuria, diarrhoea, dyspnoea, mouth ulcers, dryness of eyes and mouth, excessive hair growth, tremors, fits and swollen gums.

Results

All patients engrafted neutrophils and platelets on a median time of day 9 (7-11) and day 12 (10-17) respectively. In 21 evaluable patients (one death on day+9), platelet count >50x109/l was reached on day 18 (14-35) post transplant. The median number of blood and platelet transfusions was two (range 1-4) and four (range 2-8) respectively. Patients developed neutropenic fever (temp > 100.50 F) received antibiotics as per institution's policy. Acute GvHD requiring treatment was seen in 4 patients, while 17 had no GvHD or only grade I. Median time to discharge from hospital was day +29. Fourteen patients were followed up from 130 - 788 days. Hirsutism was seen in 10 patients, tremors were reported in 4 patients, hypertension was noted in 5 patients, gut and skin chronic GvHD was seen in 4 patients, all 4 were male, 3 of them had female donors. One patient developed shingles and late graft failure was seen in 1 patient. The cause of graft failure presumed to be pregnancy in 10th month post transplant. Overall 4 patients died post transplant in a follow up period of 788 days. The causes of death were intra-cranial
haemorrhage on day+7, herpes encephalitis on day +180, graft failure and mucour mycosis on day +353 and TB meningitis on day +544. Allogenic peripheral blood stem cell transplantation resulted in 80% event free survival in our hands. Only one death was seen beyond one-year post transplant in a patient who was off immunosuppressive drugs.

Discussion

Allogeneic BMT has been curative for aplastic anaemia, b-thalassaemia major and haematological malignancies.3,4 Survival of patients with aplastic anemia treated with transplantation of bone marrow has improved significantly over the past several decades. Allogenic bone marrow transplantation (BMT) for patients with HLA-identical siblings is now the first-line therapy, and long-term survival of approximately 90% can be expected with cyclophosphamide/antithymocyte globulin conditioning and post-grafting methotrexate / cyclosporin immunosuppression.5,13
Recent studies have shown that PBSC results in rapid haematological and immunological recovery without excessive aGvHD compared to BMT.8,14 This results in a reduced transfusion requirement before immunological recovery, early discharge and lower transplantation costs. Data from the literature suggest that PBSC appears safe for paediatric donors, yields sufficient progenitor stem cells and results in prompt engraftment.11 The median neutrophil engraftment was day +9 and all patients achieved it by day +12. Mobilization of PBSC and collection by aphaeresis has practical advantages for the donor over conventional marrow harvest i.e. no general anaesthesia or pain at the sites of bone punctures. G-CSF appears safe and well tolerated in children. There are some adverse effects like myalgia, bone pain and fever.11 Various dose schedules are in use for stem cell mobilization, even lower dose of 5 mcg/kg seemed to have yielded sufficient stem cells. Post aphaeresis, mild thrombocytopenia was observed in all donors.15,16
Many early studies reported around 50% long-term survival in aplastic anaemia post BMT. Recent

studies report survival rates of 60 - 90%. Improved survival in recent years may reflect better patient selection, earlier transplantation, changes in transplantation regimens, and/or supportive care or some combination of these factors.3,5 Although we lost four patients, treatment related mortality i.e. within first 100 days occurred in only one patient. Our initial results of 81% survival in severe aplastic anaemia patients after allografts are encouraging in the developing world setting.
To conclude, allogeneic PBSC transplantation is life saving in severe aplastic anaemia. This procedure produced comparable results in a developing country setting. Since we are the first centre to embark on allogeneic stem cell transplantation, our learning curve is expected to improve with time and better patient selection.

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