



A non-myeloablative conditioning regimen in allogeneic stem cell transplantation from related and unrelated donors in elderly patients

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We describe our experience with the use of a single non-myeloablative preparative regimen in stem cell transplantation (NST) in 37 heavily pretreated patients ≥ 55 years. The conditioning regimen consisted of fludarabine, low-dose busulfan, and antithymocyte globulin. Acute graft-versus-host disease (GVHD) grade III-IV and chronic GVHD developed in 15.6% and 44.4% of cases, respectively. With a median follow-up period of 22 (range 3-113) months, the 1-year overall survival and disease-free-survival rates were 55% and 53%, respectively, while the overall non-relapse mortality was 35%. In conclusion, reduced intensity stem cell transplantation is feasible and effective in patients ≥ 55 years. Age *per se*, should no longer be considered as a contra-indication to stem cell transplantation.

Key words: allogeneic bone marrow transplantation, graft-versus-host disease, immunotherapy, elderly, co-morbidity.

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Allogeneic hematopoietic stem cell transplantation (SCT) is a potentially curative therapeutic procedure for patients with a broad spectrum of hematologic malignancies. However, because of the significant toxicity of myeloablative conditioning, SCT has been restricted to young patients without co-morbidities. The incidence of most of the hematologic malignancies for which SCT may be indicated increases with age. For most of these diseases, such as chronic myeloid leukemia, acute myeloid leukemia (AML), and myelodysplastic syndrome (MDS), the median age at diagnosis ranges between 60 and 70 years.¹ Hence, the majority of patients in need of SCT are currently ineligible due to age limitations. The development of reduced-intensity conditioning (RIC) in preparation for SCT revolutionized the field and resulted in extended application of transplant procedures to patients previously considered not unsuitable as candidates because of age or concurrent co-morbidities.² Previously published studies proved the feasibility and efficacy of RIC in patients not eligible for conventional myeloablative conditioning.^{3,4} Recent studies showed that SCT from matched related and unrelated donor is a reasonable option for elderly patients. In these studies, patients were treated with different preparative regimens, and consequently, the efficacy of each of these regimens was difficult to evaluate.⁵⁻⁷ In this article we review our experience using a uniform RIC protocol for recipients above 55 years old.

Design and Methods

Patients' characteristics

This study included patients aged ≥ 55 years old with hematologic malignancies treated with a single RIC regimen at the Hadassah-Hebrew University Hospital, Jerusalem, Israel. The patients' characteristics are listed in Table 1. There were 30 males and 7 females with a median age of 60 years (range 55-69). The vast majority of the patients were heavily pretreated and had advanced risk disease (75%) prior to transplantation (Table 1). Donors were HLA-A,B,C and high resolution DR fully matched siblings (n=25), a 1-locus mismatched sibling (n=1), matched unrelated donors (n=9), and 1-locus mismatched unrelated donors (n=2). The source of stem cells was peripheral blood (n=33) or bone marrow (n=4). Patients were referred for SCT during the period from 1996 to 2005. Each participant signed an approved informed consent form. Eight of these 37 patients were included in our previous report.⁷

Donors

Stem cells were mobilized into the peripheral blood by subcutaneous granulocyte colony-stimulating factor and collected on days 5 and 6. Bone marrow aspiration was done under anesthesia using standard aspiration needles.

Conditioning regimen and graft-versus-host disease (GVHD) prophylaxis

All patients were conditioned with i.v. fludarabine 30 mg/m²/day (days -10 to -5), oral

Table 1. Patients' characteristics.

	Related donor	Unrelated donor
Patient no.	26	11
Sex (female/male)	6/20	1/10
Median age, range	60.3 years, (55-69)	59.7 years, (55-65)
Previous transplant	7/26	1/11
Mismatched donor	1/26	2/11
Female donor to male patient	8/26	3/11
Diagnosis and disease status before transplant		
Acute myeloid leukemia		
First complete remission	1	0
Second complete remission	1	1
Untreated relapse	1	1
Refractory	3	2
Acute lymphoblastic leukemia		
First complete remission	1	0
Chronic myeloid leukemia		
First chronic phase	1	0
Second chronic phase	1	0
Accelerated phase	0	1
Blastic phase	1	0
Myelodysplastic syndrome <i>Advanced</i>	6	2
Myelofibrosis with myeloid metaplasia	1	2
Non-Hodgkin's lymphoma		
DLBCL, CR2	1	0
DLBCL, Post Auto-BMT relapse	3	0
Transformed low grade	2	1
Refractory disease		
Multiple myeloma		
Post Auto-BMT relapse	2	0
Refractory to initial treatment	1	0
Partial remission	0	1

busulfan 4 mg/kg/day (n=13 patients), or i.v. busulfex 3.2 mg/kg/day (n=24 patients) (days -6 to -5) and i.v. ATG Fresenius (5-10 mg/kg/day, n=22) or thymoglobulin (2.5 mg/kg/day, n=15) (days -4 to -1).

GVHD prophylaxis consisted of short-term cyclosporine 3 mg/kg i.v. daily in two divided doses, starting on day -4. The cyclosporine dosage was tapered during the second to third month post transplant, according to chimeric status and evidence of GVHD. T-cell depletion was not done. Acute and chronic GVHD were graded according to the IBMTR indices. Immediately upon the appearance of GVHD, i.v. methylprednisolone (2 mg/kg) and cyclosporine were administered. In order to assess engraftment, degree of chimerism, minimal residual disease and relapse, patients were monitored by cytogenetic analysis and donor and host-specific DNA markers, using amelogenine gene polymerase chain reaction (PCR) bands and variable number of tandem repeats (VNTR-PCR) assay.

Definitions-statistics

Pre-engraftment deaths were excluded from the analysis of acute GVHD and deaths before day 100 were excluded from the analysis of chronic GVHD. Non-relapse mortality was defined as death from any cause not associated with the original disease. Overall survival was defined as the time from the day of the transplant to death from any cause, or last follow-up.

Relapse was defined as the recurrence of the hematologic malignancy, after the initial achievement of complete remission. Disease-free survival was defined as the time from the day of the transplant until relapse or death from any cause. In patients who did not achieve remission of the disease post-transplant, day 0 was considered as the day of relapse for the disease-free survival analysis.

The probabilities of overall survival and disease-free survival were plotted using the Kaplan-Meier method. The statistical significance was estimated by the log-rank test and logistic regression analysis.

Results and Discussion

Engraftment

Engraftment of neutrophils and platelets was achieved in 36 out of 37 (97.3%) patients within a median of 15 days (range 9-36) and 12 days (range 0-63), respectively. None of the patients developed graft failure.

Chimerism

Complete donor chimerism in the peripheral blood was achieved in 30/32 (94%) available patients. The first day of detection of complete chimerism was before day +35 for 17 patients, while 14 patients achieved complete chimerism between day +35 and day +65.

Graft-versus-host disease

Twenty-two out of 32 evaluable patients (68.7%), developed acute GVHD grade I-IV. However, the incidence of grade III-IV acute GVHD was only 15.6% (5/32 patients). Twelve out of 27 (44.4%) evaluable patients developed chronic GVHD (extensive in 6 cases). No significant relation was found between age and acute GVHD ($p=0.34$).

Non-relapse mortality

Eight out of 37 (21.6%) patients died during the first 100 days due to transplant-related complications. Five patients died from multi-organ failure associated with sepsis, one from pneumonia, one from idiopathic interstitial pneumonitis, and one from acute GVHD. During the first year post-transplant four more patients died of transplant-related complications, three from chronic GVHD, and one from sepsis. One patient died from coronary disease while he was in continuous complete remission (CCR), 61 months after his transplant. With a median follow-up of 22 months (range 3 to 113), the overall non-relapse mortality in this group of patients was 35% (13/37 patients).

Transplant outcome

Seventeen patients survived (median follow-up 22 months), 14 of them in CCR. Disease response and relapse are shown in Table 2. The 1-year overall survival, and disease-free survival rates for the whole group were 55% and 53%, respectively.

Table 2. Transplant outcome in relation to disease type.

Type of disease	N.	Follow up (months) Median, range	Remission	Relapse	Alive	Alive	NRM
			post-transplant N.	post-transplant N.	in CCR N.	in CCR N.	
Myeloid	25	28 (3-112)	19	6	13	10	7
Non-Hodgkin's lymphoma	7	16 (3-61)	3	1	1	1	4
Multiple myeloma	4	14 (3-21)	3	0	3	3	1
Acute lymphoblastic leukemia	1	5	1	0	0	0	1

NRM: non-relapse mortality.

Age represents one of the most significant factors that adversely affects the outcome of SCT.^{8,9} The inverse relation between age and transplant outcome is related to many factors. The number and severity of co-morbidities, including organ malfunction, in addition to being overweight and having associated metabolic disorders may make aging individuals prone to increased transplant-related toxicity and mortality. The aim of our study was to evaluate the feasibility and effectiveness of our NST protocol in allogeneic SCT from related and unrelated donors in patients older than 55 years. All evaluable patients achieved rapid complete donor chimerism, without the need for immune system manipulation such as cyclosporine withdrawal or donor lymphocyte infusions. Moreover, despite the fact that we included patients transplanted from unrelated donors (two of whom were mismatched), no patient developed primary or secondary graft rejection, further proving that this preparative regimen is sufficiently immunosuppressive to allow engraftment.

Interestingly, the cumulative incidence of severe acute GVHD was low, and only four patients died from GVHD. Additionally, in this study no significant association was observed between the incidence or severity of acute GVHD and the age of the recipients. The low incidence of severe GVHD in our study can be explained by the inclusion of ATG, and remnants of host veto cells. Despite the high-risk status of our patients, it is of interest to note that the majority of transplant-related deaths occurred during the first 100 days, with only few deaths observed after this period.

Even though most of our patients were transplanted with active refractory disease, 19/32 patients with active disease achieved complete remission, accounting for an overall CR rate of 60%. Ten out of 25 patients with myeloid diseases are in CCR, while only one out of seven patients with non-Hodgkin's lymphoma is in CCR (six of the seven patients with non-Hodgkin's lymphoma included in our study had refractory/relapsed high-grade or transformed low-grade lymphoma). Post-transplant administration of monoclonal antibodies

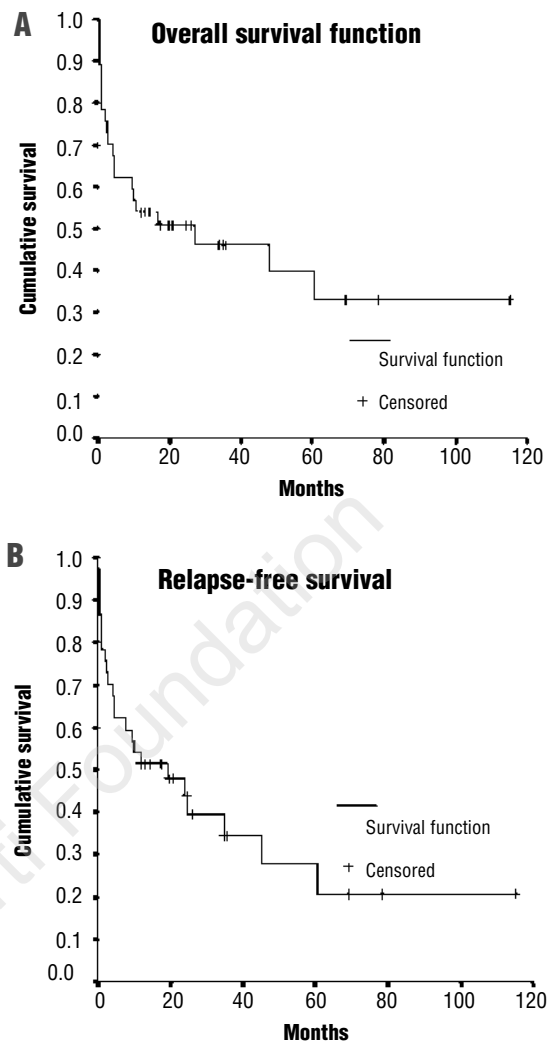


Figure 1. A. Overall survival and B disease-free survival for the whole group.

may improve the outcome of these patients, as previously shown.¹⁰

Many RIC regimens have been proposed during the last years, differing in the intensity of the preparative regimen. Less intense regimens are associated with less toxicity but at the cost of reduced anti-leukemic activity, while the opposite is true for more intensive regimens.^{11,12} In a recent study, Gupta *et al.*,¹³ tested the feasibility and efficacy of matched sibling allogeneic SCT with a non-myeloablative conditioning regimen in 24 patients older than 60 years with AML/MDS. Conditioning consisted of fludarabine plus low-dose total body irradiation. Non-relapse mortality rates were 8% and 25% at 100 days and 2 years, respectively, while 27% of the patients relapsed post-transplant. With a median follow-up of 21 months, 11 out of 24 patients survived free of disease. However, most of the patients included in this study had early stage disease at the time of transplantation (13/15 AML patients were in first complete remission, while four of the nine MDS

patients had refractory anemia). In our study, 18 patients with AML or MDS were included and among them, only one of ten AML patients was in first complete remission, while all eight patients with MDS had refractory anemia with excess blasts. In our study, with a median follow-up of 25 months, eight out of these 18 patients survive in CCR, this being similar to rate in Gupta's report.

Taken together, the results of our study are in accordance with those of recent studies evaluating the feasibility and effectiveness of RIC protocols in the elderly. Published studies showed that SCT is a reasonable option for elderly patients.^{5-7,14}

In conclusion, the results of our study suggest that our RIC regimen is feasible in patients older than 55 years who can then undergo SCT from either matched or minimally-mismatched related or unrelated donors. The effectiveness of our preparative regimen is promising in view of the fact that the vast majority of patients were

treated in an advanced stage of disease with a life expectancy of no more than a few months. Prospective randomized studies and longer observation periods are required to confirm our data and tentative conclusions.

PT, RO, SS, MYS contributed equally to the work and assume primary responsibility for it; RO, SS were responsible for the design of the study, supervision of data collection, data analysis, and writing the manuscript; PT, MYS were responsible for the supervision of data collection and interpretation, data analysis, and writing the manuscript; MB, IBR, SS, AA, SE, BG, IZ, SM, AL were involved in the investigations, interpretation of the data and revision of the manuscript.

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