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ORIGINAL ARTICLE

Non-myeloablative allogeneic hematopoietic transplantation for patients with hematologic malignancies: 9-year single-centre experience

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ABSTRACT

Matched related and unrelated allogeneic non-myeloablative hematopoietic transplantation (NMT) is increasingly being used in patients with hematologic malignancies. Conditioning regimens and indications for NMT vary considerably from centre to centre. Our institution uses intravenous fludarabine and cyclophosphamide, plus graft-versus-host disease (GVHD) prophylaxis with tacrolimus and mycophenolate mofetil. We retrospectively analyzed 89 consecutive patients who underwent NMT (65 related, 24 unrelated) at our institution from October 2002 to September 2011. The most frequent indications for NMT were acute myelocytic leukemia (high-risk in first complete or subsequent remission: $n = 20$, 22.5%) and relapsed follicular lymphoma ($n = 18$, 20.2%). The cumulative incidence of acute GVHD (grades 2–4) was 28.1% ($n = 25$), and rates were similar for related ($n = 18$, 28%) and unrelated ($n = 7$, 29%) NMT. At a median follow-up of 22.6 months, the cumulative incidence of chronic GVHD (limited and extensive) was 68% ($n = 61$): 68.5% ($n = 44$) for related and 71% ($n = 17$) for unrelated NMT. The 100-day transplant-related mortality rate was 2.2%: 1.5% for related and 4.2% for unrelated NMT. Of the 89 patients, 30 (33.7%) have relapsed: 41.5% after related and 12.5% after unrelated NMT. Relapse rates were similar in patients with myeloid and lymphoid malignancies (36.4% vs. 33.3%). The 3-year overall and progression-free survival rates were 50.0% and 43.4% respectively, with multivariate analysis showing that neither rate was affected by age, disease group, status at transplantation, or related compared with unrelated NMT. Our findings indicate that, despite its limitations, including the incidence of chronic GVHD, NMT is an important treatment modality for a selected subgroup of patients with hematologic malignancies.

KEY WORDS

Non-myeloablative stem-cell transplantation, conditioning regimens, graft-versus-host disease

1. INTRODUCTION

Non-myeloablative allogeneic hematopoietic transplantation (NMT) is increasingly being used as a treatment strategy in patients with hematologic malignancies. Its scientific premise is that a less-intensive preparative regimen would likely result in less toxicity to organs and would therefore be better tolerated by patients otherwise precluded from standard myeloablative transplantation because of advanced age or comorbidities[1–3].

Despite the more frequent use of NMT, the technique might be associated with a high incidence of chronic graft-versus-host disease (GVHD)—in some settings, higher than that with standard myeloablative transplantation. Although the incidence of GVHD can be lowered with the use of alemtuzumab, that agent has been associated with a higher incidence of cytomegalovirus reactivation. Additionally, NMT patients will often require donor lymphocyte infusion to control their disease.

After transplantation, GVHD is a major determinant of morbidity, quality of life, and survival[4]. However, a graft-versus-tumour (GVT) effect can also play a major role in lowering the risk of cancer relapse. Chronic GVHD has been associated with improvements in relapse-free survival, particularly in more indolent malignancies, including low-grade lymphoma and chronic leukemia[5,6]. Relapse rates were shown to be lower in patients receiving allogeneic than autologous hematopoietic stem-cell transplants, particularly from unrelated donors[7,8], with higher relapse rates in patients receiving syngeneic[9] or T-cell–depleted[10] allografts.

The present study describes our experience of patients who have undergone NMT at our centre over a 9-year period, especially rates of survival and GVHD. In particular, we retrospectively assessed the effects on patient outcomes of donor–recipient relatedness and type of malignancy.

2. METHODS

2.1 Patient Population

This retrospective analysis enrolled consecutive patients who underwent NMT at our institution during a 9-year period between October 1, 2002, and September 30, 2011. Eligibility requirements for NMT included an Eastern Cooperative Oncology Group performance status of 0–2, absence of active infection at the time of transplantation, and normal or near-normal lung, renal, and cardiac function.

2.2 Donors

All donors included in this analysis were matched for human leukocyte antigens at the A, B, and DR loci. Unrelated donors were low-resolution molecular matches at those loci. Donor peripheral blood stem cells were mobilized with filgrastim (10 μg/kg daily, given subcutaneously for 4 days), with stem-cell collection initiated on the 5th day of filgrastim treatment. Apheresis was continued until at least 2.5 million CD34 cells per kilogram recipient weight were collected (1 or 2 procedures).

2.3 Conditioning Regimens and Graft-Versus-Host Donor Prophylaxis

The conditioning regime for recipients consisted of intravenous fludarabine (30 mg/m^2) plus cyclophosphamide (300 mg/m^2) daily for 5 days. The NMT regimen, designed to minimize cytopenia and not requiring stem-cell rescue, has previously been described[11].

Prophylaxis for GVHD consisted of oral mycophenolate mofetil (500 mg twice daily) from day 1 to day 50) and tacrolimus from day 1 to day 5 at doses to ensure trough levels between 5 μg/mL and 15 μg/mL[12]. If GVHD did not occur by day 50, tacrolimus was tapered over the next 2–3 months. Supportive care included prophylactic transfusion of red blood cells if hemoglobin dropped below 80 g/L and of platelets if platelet count dropped below 10×10^3/mm^3

2.4 Statistical Considerations

Descriptive statistics are used to describe the baseline characteristics of patients, disease, and disease status at conditioning. Categorical variables in patients undergoing related and unrelated NMT were compared using two-sided Fisher exact tests, and continuous variables were compared using the Student *t*-test. The number of relapses per person–year was calculated by dividing the number of relapses in a cohort (or overall) by the total number of person–years in the cohort (or overall). Cox regression by the Breslow method was used to analyze specific cohort relapses, and log-rank *p* values were determined. Overall survival (OS) and progression-free survival (PFS) were calculated using the Kaplan–Meier method and compared using the log-rank test. Progression-free survival was defined as the time from stem-cell infusion to relapse or death from any cause. Overall survival was defined as the time from infusion to death from any cause. Cox regression analysis was performed separately for OS and PFS. All variables included in the model were appropriately selected based on clinical judgment. All continuous variables were tested for linearity on the log-hazard scale. The proportional hazards assumption was tested using plots of residuals and including an interaction term with the log time-to-event.

3. RESULTS

3.1 Patient Characteristics

Table I shows the demographic and clinical characteristics of the 89 patients who underwent NMT at our institution during the study period. The median age of the recipients was 59 years (range: 17–69 years) and of the donors, 57 years (range: 35–71 years). Most donors (n = 51, 57.3%) and recipients (n = 61, 68.5%) were men. The most frequent indications for NMT were acute myelocytic leukemia [AML (high-risk in first complete or subsequent remission): n = 20, 22.5%] and relapsed follicular lymphoma (n = 18, 20.2%). Other indications were chronic lymphocytic leukemia (n = 6), diffuse large B-cell lymphoma (n = 7), Hodgkin lymphoma (n = 6), mantle cell lymphoma (n = 6), multiple myeloma (n = 8), plasma cell leukemia (n = 2), myelodysplasia (n = 6), myeloproliferative disorders (n = 3), and chronic myelomonocytic leukemia (n = 4). All patients received immunosuppressive medication as GVHD prophylaxis.

3.2 GvHD

The overall cumulative incidence of acute GVHD (all grades) was 36.0% (n = 32): 35% (n = 23) in patients undergoing related NMT and 37% (n = 9) in patients undergoing unrelated NMT (p = 0.85). The overall cumulative incidence of grades 2–4 acute GVHD was 28.1% (n = 25): 28% (n = 18) for related and 29% (n = 7) for unrelated NMT (p = 1.000, Figure 1).

At a median follow-up of 1.88 years (range: 0.09–8.45 years), the cumulative incidence of chronic GVHD (limited and extensive) was 68.5% (n = 61): 67% (n = 44) for related and 79% (n = 19) for unrelated NMT (p = 0.21). The cumulative incidence of extensive chronic GVHD was 47.2% (n = 42): 45% (n = 29) for

TABLE I Characteristics of the study patients

Characteristic	*Value*
Patients (*n*)	89
Age (years)	
Recipients	
Median	59
Range	17–69
Donors	
Median	57
Range	35–71
Sex (*n* men:women)	
Recipients	61:28
Donors	52:37
Disease type and status at conditioning (*n*)	
Acute myeloid leukemia	20
CR1	8
CR2	11
>CR2	2
Follicular lymphoma	18
CR1	1
CR2	6
>CR2	11
Multiple myeloma	8
CR1	1
CR2	5
>CR2	2
Diffuse large B-cell lymphoma	7
CR1	0
CR2	1
>CR2	6
Chronic lymphocytic leukemia	6
CR1	1
CR2	1
>CR2	4
Hodgkin lymphoma	6
CR2	1
>CR2	4
Relapse	1
Mantle cell lymphoma	6
CR1	1
CR2	1
PR2	4
Plasma-cell leukemia	2
CR2	1
>CR2	1
Myelodysplastic syndrome	6
Myeloproliferative disorder	3
Chronic myelomonocytic leukemia	4
CR1	1
CR2	1
Progression	2
Other leukemia or lymphoma	3
>CR2	3

CR*n* = complete response *n*; PR*n* = partial response *n*.

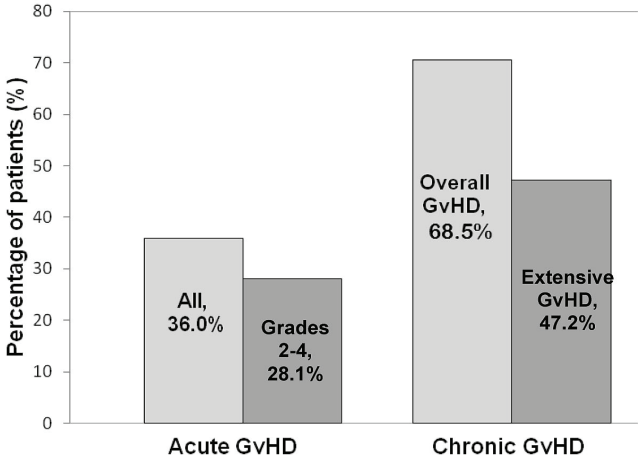

FIGURE 1 Cumulative rates of acute and chronic graft-versus-host disease (GVHD) in patients receiving non-myeloablative transplantation.

related and 54% (n = 13) for unrelated NMT (p = 0.42, Figure 1).

3.3 Transplant-Related Mortality and Relapse

The overall 100-day transplant-related mortality rate in patients undergoing NMT was 2.2%. Of the 89 patients, 30 (33.7%) relapsed with a follow-up time of 1.55 years on average (range: 0.016–8.4 years). Of the 30 relapsed patients, 3 had unrelated transplants, and 27, related transplants (p = 0.06, Figure 2.)

3.4 OS

The 1-year OS rate was 70.3% in recipients of related transplants and 72.0% in recipients of unrelated transplants. The 3-year OS rates were 50.2% and 45.0% respectively (p = 0.8). Patients with myeloid and lymphoid malignancies had similar 1-year (63.7% vs. 74.8%) and 3-year (42.3% vs. 52.9%) OS rates (p = 0.1, Figure 3).

3.5 PFS

The 1-year PFS rate in patients undergoing related and unrelated NMT was 51.8% and 68.6% respectively. The 3-year PFS rate was 41.5% and 42.9% (p = 0.4). Patients with myeloid and lymphoid malignancies had similar 1-year (50.5% vs. 58.1%) and 3-year (44.2% vs. 42.0%) PFS rates (p = 0.4, Figure 4).

3.6 Factors Associated with Outcome

Multivariate Cox regression analysis was performed to estimate hazard ratios and 95% confidence intervals for potential predictors associated with OS, PFS, or transplant-related mortality. Factors analyzed included recipient age, donor type (related vs. unrelated), disease group at conditioning (myeloid vs.

lymphoid), and disease status at conditioning. Of those factors, none were significantly associated with either OS or PFS (Table II).

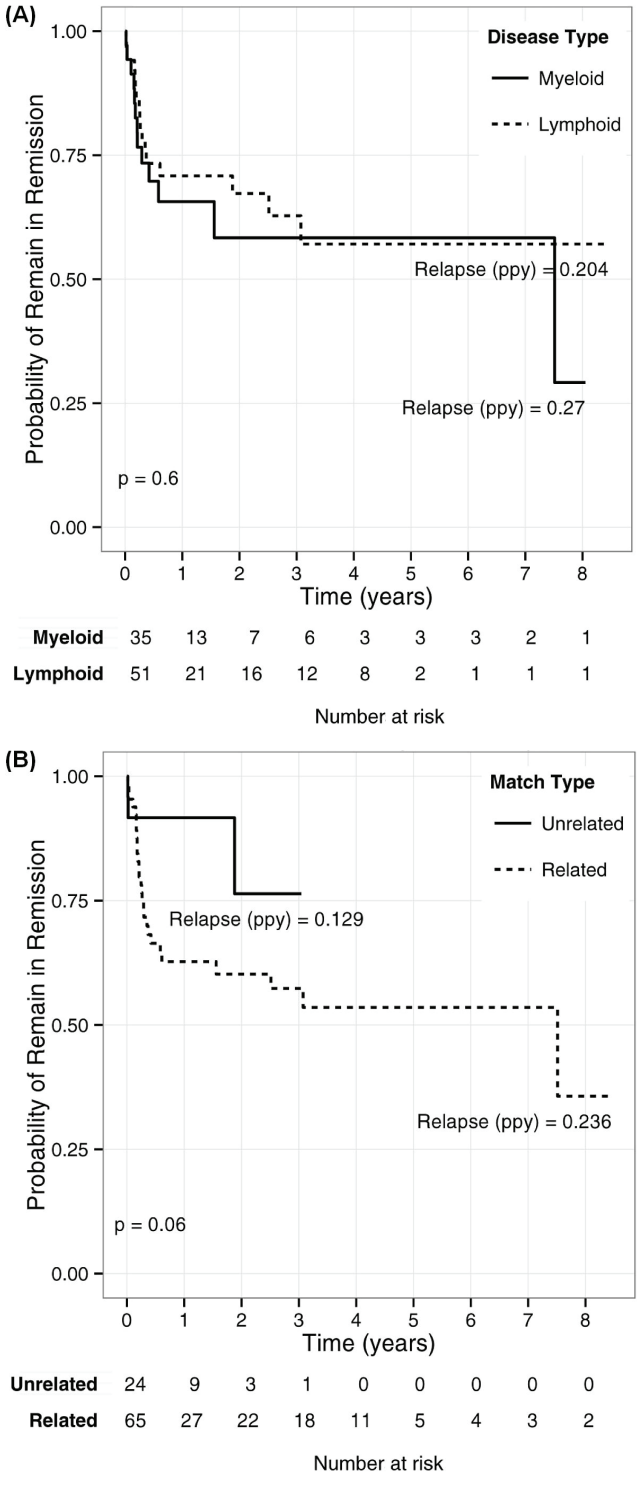

FIGURE 2 Time to relapse for all patients by (A) disease type and (B) donor type (matched related vs. unrelated). Number of relapses per person-year (ppy) is calculated by dividing the number of relapse in a cohort (or overall) by the total number of person–years in the cohort (or overall).

4. DISCUSSION

This retrospective analysis of 89 patients suggests that NMT is an important treatment modality for

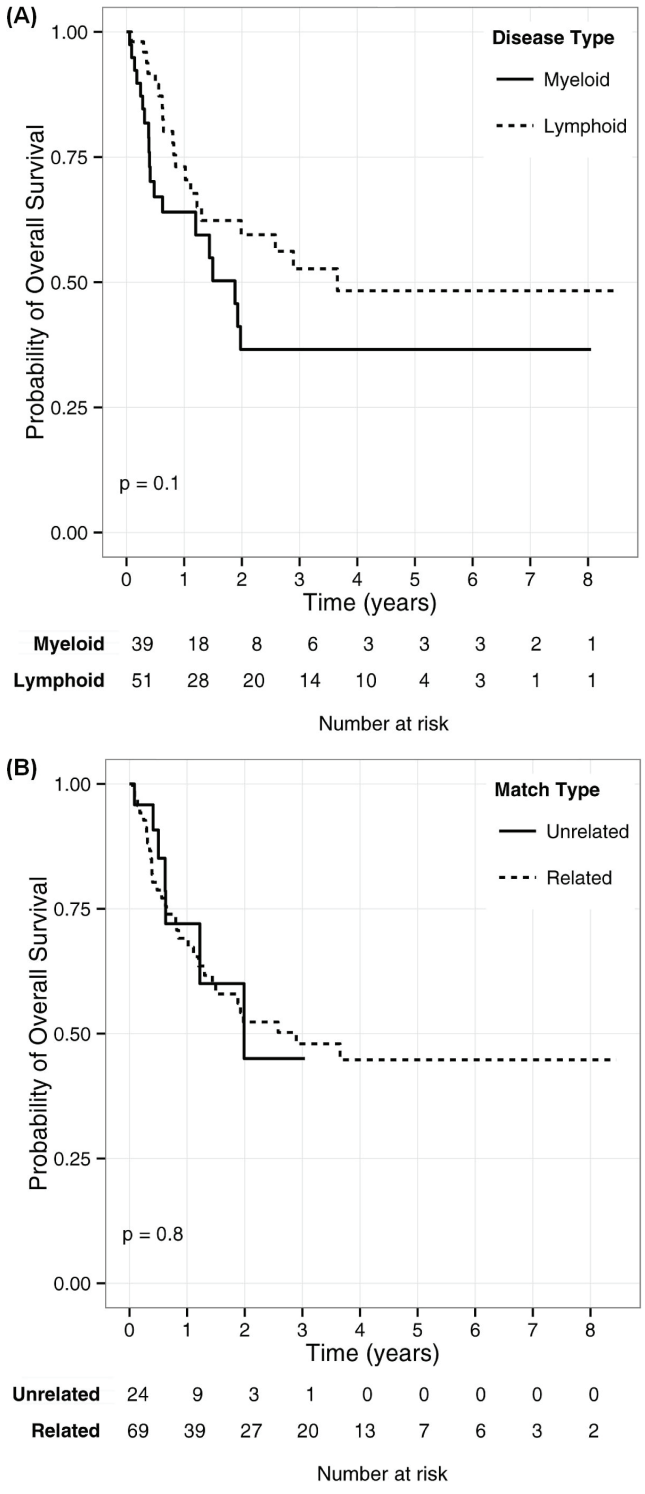

FIGURE 3 Kaplan–Meier curves for overall survival in patients receiving non-myeloablative transplantation, by (A) disease type and (B) donor type (matched related vs. unrelated).

a selected subgroup of patients with hematologic malignancies. Our centre performs allogeneic transplants for the Atlantic provinces of Canada, consisting of New Brunswick, Nova Scotia, Prince Edward Island, and Newfoundland and Labrador, encompassing a total population of about 2.5 million. Currently, 25% of the transplants performed in patients with these malignancies are non-myeloablative. Data about the proportion of patients with AML aged 55–65 years who undergo transplantation are not available. During the study period, NMT was used in all patients 55–65 years of age receiving an allogeneic transplant, and also in patients less than 55 years of age with impaired performance status (although there were few of the latter). The most frequent indications for NMT in our series were AML and indolent relapsed lymphoma. In keeping with other reports, the 100-day mortality rate in our study was less than would be expected for a myeloablative transplant strategy; however, OS and PFS rates were similar to those previously observed[13–19].

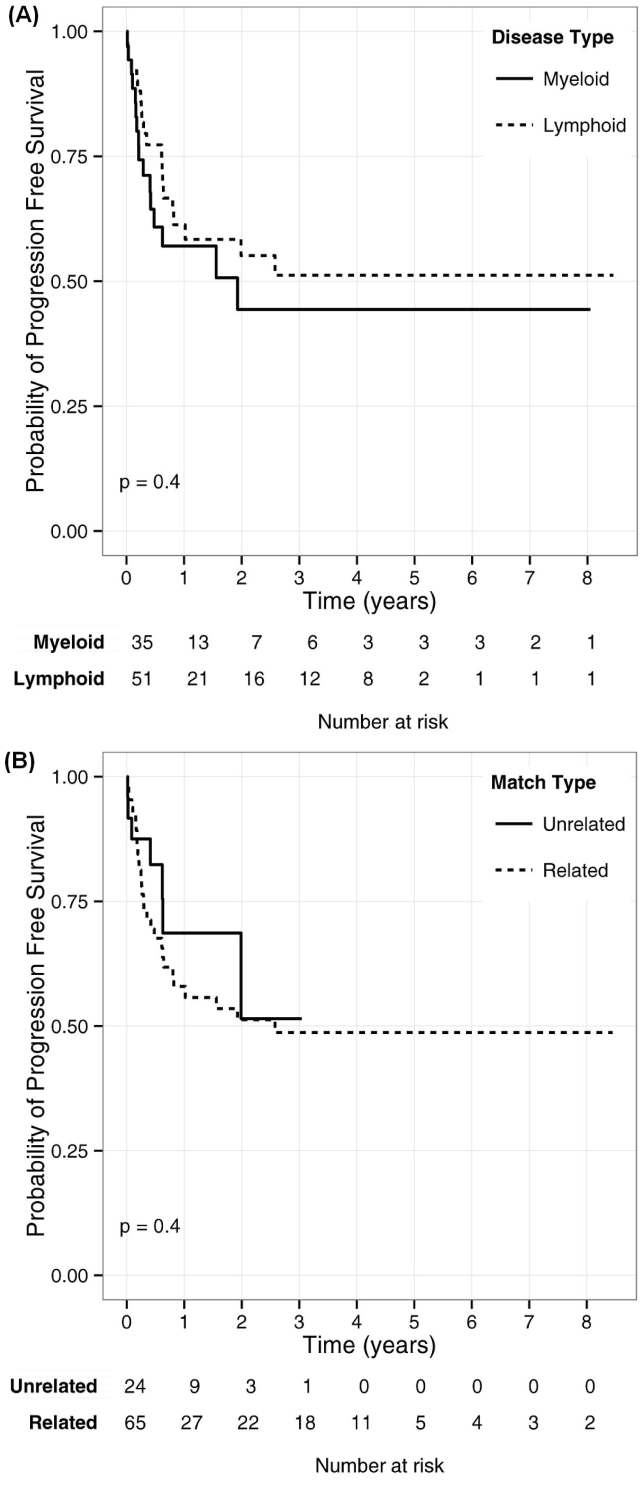

FIGURE 4 Kaplan–Meier curves for progression-free survival in patients receiving non-myeloablative transplantation, by (A) disease type and (B) donor type (matched related vs. unrelated).

We observed high rates of NMT-associated overall and extensive chronic GVHD in our patient cohort. Those findings accord with results in previous studies, which found that the incidence of GVHD was high in patients undergoing NMT and suggested that the incidence might be higher in patients undergoing NMT than in patients undergoing conventional myeloablative allografting[13,14,20–23]. The difference might in part relate to the characteristics of patients who undergo NMT, in that they are generally older and receive mobilized peripheral blood grafts rather than bone marrow grafts (peripheral blood grafts being associated with a higher incidence of GVHD in the myeloablative setting[3,24]).

Our multivariate regression analysis found that recipient age, disease group at conditioning (myeloid vs. lymphoid), and disease status at conditioning were not significantly associated with OS or PFS. In agreement with earlier results[23], no statistically significant difference in survival outcomes was observed in patients who underwent transplantation from related and unrelated donors. Patients receiving transplants from unrelated donors might have experienced longer delays to transplantation, suggesting more stable disease. Although the precise biology of such a response is unclear, it is believed to involve reactions

TABLE II Multivariate Cox regression analysis of factors associated with overall survival and progression-free survival

Factor	*Survival type*			
	Overall		*Progression-free*	
	p *Value*	*HR*	p *Value*	*HR*
Age	0.5092	0.987	0.1262	0.973
Disease group	0.1222	0.484	0.5559	0.772
CR1/PR1	0.4596	0.506	0.9755	0.976
CR2/PR2	0.6052	0.666	0.6203	0.720
Donor type	0.6248	1.257	0.4335	0.708

CR*n* = complete response *n*; PR*n* = partial response *n*.

to polymorphic minor histocompatibility antigens expressed either specifically on hematopoietic cells or more widely on a number of tissue cells. Graft-versus-tumour was first described in 1956, following from the observation that transplanted immunocompetent cells could eliminate leukemic cells in mice, independent of chemotherapy[25]. Since then, studies in humans have demonstrated that donor lymphocyte infusions can induce complete remission in some patients with hematologic malignancies[26,27]. Our study lends further support to the concept and suggests the need for further exploration of the potential benefits of using unrelated donors.

Notably, we observed a statistically nonsignificant trend toward improved OS in patients with lymphoproliferative diseases, suggesting that those patients benefit more than patients with myeloid disease from an immune-mediated GVT effect, although the latter group might have had higher-risk disease[28]. That finding accords with results suggesting that GVT effects might be more beneficial in diseases that progress slowly, but are less effective in rapidly-growing cancers[10,19,29]. Malignancies showing high sensitivity to GVT effects have been found to share a number of characteristics. They are, in general, indolent disorders that are not immediately life-threatening, thereby providing a longer window for a GVT effect to develop. In addition, these malignancies often arise from antigen-presenting cells, suggesting that their responsiveness might be partly related to effective *in vivo* presentation[30]. Insensitive malignancies, including high-grade lymphomas and AML, typically proliferate at a rapid rate, outpacing a developing immune response, and generally lack co-stimulatory molecules to effectively stimulate an immune response.

A major limitation of our study is its retrospective design. In addition, our sample size was relatively small, precluding accurate comparisons—especially between recipients from matched related and unrelated donors. In addition, the study was restricted to patients at a single centre, all of whom were received the same conditioning regimen. Earlier studies have suggested that outcomes might be influenced by the conditioning regimen[14,17,18], suggesting the need for studies to determine the optimal conditioning regimen for NMT.

5. CONCLUSIONS

Our results indicate that, despite its limitations (including a higher risk of relapse[31]), NMT yields encouraging OS and PFS results in selected high-risk patients. Acute and chronic GVHD remain significant concerns, suggesting the need for additional studies to reduce the incidence of chronic GVHD. However, the apparently improved survival rates associated with GVT effects suggest a need to identify strategies that eliminate the risk of GVHD while maintaining the beneficial effects of allogeneic NMT. Such strategies might include alternative conditioning regimens, different graft content, and new immunosuppressive therapies for both prophylaxis and treatment of GVHD.

6. CONFLICT OF INTEREST DISCLOSURES

All authors declare that no financial conflict of interest exists.

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