Autologous Stem Cell Transplantation Is an Effective Salvage Therapy for Primary Refractory Multiple Myeloma

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A B S T R A C T

High-dose therapy and autologous stem cell transplantation (ASCT) have proven efficacy in patients with multiple myeloma responding well to induction therapy. For those who fail to achieve a stable partial response (PR), the effect of ASCT is unclear. We report on 126 patients identified from a national database, who underwent ASCT having achieved <PR after induction with modern induction regimens. The overall response rate was 86% (24% complete response). Patients with progressive disease at the time of transplantation had poorer outcomes than those with minimally responsive or stable disease, but clinical benefit was seen in all groups. Day 100 and 1-year nonrelapse mortalities were 2% and 4%, respectively. The 5-year relapse rate and progression-free survival were 84% and 14% (median, 18 months), respectively. The 5-year overall survival was 42% (median, 51 months). Our findings support the use of ASCT in myeloma patients responding suboptimally to modern induction therapies. Patients should not be excluded on the basis of refractoriness to induction, as ASCT is effective in this group conventionally considered to have a poor outcome. Comprehensive multivariate analysis identified no disparate subgroups, meaning ASCT is a reasonable strategy for all fit primary refractory patients.

INTRODUCTION

Since the initial demonstration of high-dose melphalan for multiple myeloma (MM), in excess of 500 reports have been published on its use, with nearly 15,000 patients undergoing autologous stem cell transplantation (ASCT) in Europe. Randomized controlled studies show improved response rates, progression-free survival (PFS), and overall survival (OS) compared with conventional chemotherapy [1]. ASCT has, therefore, become the established front-line therapy for those biologically fit enough for its physiological challenges. The depth of response to induction therapy is correlated with outcome after ASCT; attainment of at least a very good partial response (PR) is associated with superior PFS [2]. However, even with novel agent–containing induction regimens, up to 25% of newly diagnosed patients have poorly responsive disease (<PR), and this proportion rises with sequential relapses.

Studies in other refractory B cell malignancies (eg, non-Hodgkin lymphoma) have yielded disappointing results with ASCT [3]. However, this may not be the case for MM—the few published reports are conflicting and largely predate novel agents. In the early 2000s, Singhal et al. and Kumar et al. reported cohorts with primary refractory MM (PRM) (43 and 50 patients, respectively) who received ASCT after conventional induction therapy [4,5]. They found no difference in long-term outcomes compared with patients undergoing ASCT with chemo-sensitive disease. In contrast, in a post hoc analysis of the IFM 2005-01 trial, Moreau et al. reported failure to achieve ≥very good PR after bortezomib-based induction resulted in inferior PFS after ASCT [2]. Nonetheless, even if outcomes of ASCT for <PR are inferior to those of ASCT for ≥PR, the modality might nevertheless offer clinical benefit.

We sought to delineate the clinical course of patients who underwent ASCT despite failing to achieve a PR after induction with modern therapies. Patients were identified as having achieved minimal response (MR), stable disease (SD), or progressive disease (PD) at the time of ASCT. In this report, we examine the impact of ASCT and discuss the clinical utility of ASCT for aggressive and poorly responding disease.

DESIGN AND METHODS

Patient Selection, Definitions, and Procedures

This retrospective study was approved and registered by the British Society of Blood and Marrow Transplantation Clinical Trials Committee. Eligible patients were identified from the British Society of Blood and Marrow Transplantation Data Registry. Consent was obtained at the time of transplantation, in line with European Bone Marrow Transplant Registry directives with European Bone Marrow Transplant response criteria [6] were used throughout, as the majority of patients underwent transplantation in or before 2006, when the more recent International Myeloma Working Group criteria were published. Patients were eligible if, at the time of ASCT, they had never achieved PR (ie, best response was MR, SD, or PD) and had undergone stem cell collection sufficient to undertake ASCT, by either peripheral apheresis or bone marrow harvesting. Bone marrow aspirate and trephine biopsy was performed at 100 days after transplantation, unless declined.

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Statistical Analysis

Metrics collected for all patients were as follows: age at diagnosis, age at ASCT, gender, Karnofsky status at ASCT, β_2-microglobulin at diagnosis, albumin at diagnosis, serum creatinine at diagnosis, serum creatinine at ASCT, number of lines of prior therapy, disease status at time of ASCT, time from diagnosis to ASCT, time from first therapy to ASCT, and ASCT conditioning regimen. The Kaplan-Meier product-limit estimator was used for median and range of the follow-up time and univariate (UVA) probabilities. Nonrelapse mortality (NRM), relapse, PFS, and survival after ASCT were evaluated in multivariate analyses (MVA) using competing risk analysis to identify patient-, disease-, and transplantation-related variables prognostic of outcomes (relapse and NRM were used as competing risks for each another). The assumption of proportional hazards for each factor in the Cox model was tested using time-dependent covariates. For nonproportional hazards, the post-transplantation time course was broken into 2 periods, using the maximized partial likelihood method to find the most appropriate breakpoint. Interactions between covariates were tested before stepwise modeling. The final MVA model was built using a forward stepwise model. All *P* values were 2-sided.

RESULTS

Patients

One hundred twenty-six eligible patients were identified and underwent transplantation between 2000 and 2008 at 18 centers in the United Kingdom. Patient and transplantation characteristics are shown in Table 1. All patients in this study had primary refractory disease, having failed to achieve at least a PR in response to any and all prior therapies; 67 (53%) patients received ASCT "up front" after 1 line of induction therapy resulting in <PR (including 11 patients with PD during induction); 59 (47%) received ASCT after more than 1 cycle of therapy, again having never achieved a PR or better in response to any therapy. Induction regimens were not standardized but were in keeping with current United Kingdom practice during that time period, and, therefore, some incorporated thalidomide but not lenalidomide or bortezomib. No patients had received prior ASCT. Cytogenetic data were available for too few patients to allow subgroup analysis. Median time to engraftment (defined as peripheral blood neutrophils $> .5 \times 10^9$/L) was 14 days (range, 9 to 117) and platelet engraftment ($>50 \times 10^9$/L unsupported) was 19 days (range, 10 to 132). Median follow-up is 61 months (range, 1 to 112).

NRM

Three of 126 evaluable patients died of treatment-related causes within 100 days. NRM at 100 days, 1 year, and 5 years were 2%, 4%, and 10%, respectively. UVA and MVA are shown in Table 2 (variables listed as collected in the Methods section, and those not included in the table did not reach significance).

Response to ASCT and Relapse Rate

At day 100, the complete response (CR) rate was 21% (95% confidence interval [CI], 13% to 29%) and the PR rate was 74% (95% CI, 65% to 82%) (Table 1). Response rate was not correlated with any demographic or treatment factors. Patients with MR or SD at the time of transplantation demonstrated a CR rate of 24% and PR rate of 70%, compared with those with PD at the time of transplantation, who had 16% and 79%, respectively ($P = .608$). Disease response at day 100 (CR versus PR versus PD) was strongly predictive of OS, PFS, and relapse rate ($P = .02$, $P = .003$, $P = .003$, respectively). Given that administration of high-dose melphalan is the rationale for ASCT, UVA and MVA for response rate, OS, PFS, and relapse rate by melphalan dose were untaken and did not reach significance.

Table 1
Patient Characteristics and Response to ASCT at Day 100

Characteristics	Value
No. of patients, n	126
Sex, n (%)	
Male	77 (61)
Female	49 (39)
Age at diagnosis, median (range), yr	54 (25-69)
Age at transplantation, median (range), yr	56 (33-72)
Time from first treatment to transplantation, median (range), mo	7 (3-73)
>12 months (%)	16
ISS score, n (%)	
I	22 (50)
II	15 (34)
III	7 (16)
Unknown	82
Karnofsky status at transplantation, n (%)	
100	12 (15)
90	39 (48)
80	25 (31)
70	4 (5)
60	1 (1)
Unknown	45
Serum creatinine at diagnosis, median (range), μmoL/L	86 (43-577)
Unknown	69
Serum albumin at diagnosis, median (range), g/L	38 (21-49)
Unknown	80
Serum β2-microglobulin at diagnosis, median (range), mg/L	3.2 (1.1-76)
Unknown	82
Prior lines of therapy, median (range)	1 (1-4)
Unknown (n)	12
Prior exposure to	
Vincristine	79
Idarubicin	21
Cyclophosphamide	49
Melphalan	10
Adriamycin	79
Etoposide	9
Thalidomide	17
Disease status at transplantation, n (%)	
MR	48 (38)
SD	31 (25)
PD	47 (37)
High-dose therapy regimen, n (%)	
Melphalan 200 mg/m^2	62 (52)
Melphalan 140 mg/m^2	16 (13)
Melphalan 100 mg/m^2	8 (6)
Melphalan other dose	34 (27)
Unknown	6
Stem cell source, n (%)	
Peripheral blood	123 (98)
Bone marrow	2 (2)
Combination of both	1 (1)
Response to transplantation at day 100, n (%)	
CR	24 (21)
PR	84 (74)
MR	1 (1)
SD	3 (3)
PD	2 (2)
Death (disease)	0
Death (ASCT-related)	3 (3)
Unknown	6 (6)

ISS indicates International Scoring System.

At the time of analysis, 65 patients had died at a median of 25 months after ASCT (95% CI, 19 to 35): 54 were due to disease progression and 11 unrelated causes. The relapse rates at 1 year and 5 years were 33% and 84%, respectively. PD at the time of transplantation conferred an increase in rate of relapse (47% at 1 year, compared with 18% and 20% for SD and MR, respectively, $P = .022$).

Table 2
Univariate and Multivariate Analysis for OS, Response Rate, PFS, NRM, and Relapse Rate

Outcome	Variable	*P* Value
OS	Disease status at transplantation	.012
PFS	Albumin at diagnosis (continuous)	.042
	Karnofsky status at transplantation	.005
	Disease status at transplantation	.003
	Disease response at day 100*	.003
	Albumin at diagnosis (continuous) (MVA)	.033
	Karnofsky status at transplantation (MVA)	.040
NRM	Albumin at diagnosis (continuous)	.021
	Albumin at diagnosis (continuous, MVA)	.021
Relapse rate	Karnofsky status at transplantation	.002
	Creatinine at diagnosis (continuous)	.037
	Disease status at transplantation	.009
	Disease response at day 100*	.003
	Karnofsky status at transplantation (MVA)	.007
	Disease response at day 100 (MVA)*	.002

All *P* values are for UVA, unless specified.
* Patients surviving beyond day 100 only.

PFS and OS

The PFS at 5 years was 15% (95% CI, 9% to 23%), with a median PFS of 18 months (range, 15 to 21) (Figure 1). OS at 5 years was 42% (95% CI, 32% to 52%), with a median OS of 51 months (range, 44 to 62) (Figure 1).

DISCUSSION

Induction therapy followed by consolidation with high-dose therapy and ASCT is currently standard practice for fit patients with responsive disease [1]. However, in those responding suboptimally to modern induction regimens, the role of ASCT is unclear and little evidence exists as to which patients might benefit.

Our data clearly show that for patients whose disease response before ASCT was <PR, ASCT is an effective means of inducing a deeper remission, with overall response rates of 86%. This is in contrast to previous reports suggesting patients with disease poorly responsive to conventional regimens do not gain clinically useful responses from ASCT [7]. This is extremely important, since depth of response in MM is now well recognized to predict PFS and OS—indeed, in our cohort, depth of response at day 100 correlated with the relapse rate. Patients with PRM had a shorter median PFS than published reports of those who underwent transplantation in ≥PR, but this nevertheless translated into a good 5-year OS in an otherwise difficult-to-manage group of patients [8,9]. Of course, this OS is partially reflective of advances in salvage therapy employed at relapse, just as in ASCT for responsive disease; the combination of ASCT and these agents should be considered additive rather than mutually exclusive. It is also worthy of note that although 52% of the patients in our cohort received melphalan 200 mg/m^2, the remainder were treated with lower doses, which would be expected to reduce the efficacy of the treatment. Our results may, therefore, actually underestimate the efficacy of ASCT with "full-dose" melphalan for PRM. Reassuringly, within our cohort the 1- and 5-year NRM were 4% and 10%—similar to ASCT for responsive disease [1,10].

OS, PFS, and relapse rate were correlated with disease status at transplantation, with patients achieving MR or SD faring better than those with PD, although clinically useful responses were still seen in those patients with PD. These findings are in keeping with a recent demonstrating poor outcomes for patients with PD (PFS and OS of 7 and 13 months, respectively) [11].

Naturally, there are some limitations inherent to a retrospective analysis. First, induction regimens were not standardized but were in keeping with United Kingdom practice during the time frame of data capture. Some induction regimens included thalidomide, and when prior exposure to thalidomide was examined in UVA and MVA, no significant effect on outcomes was seen. However, outside of clinical trials, bortezomib and lenalidomide became available to UK National Health Service patients in 2007 and 2009, respectively, and consequently none of the included patients had received these agents. Although the inclusion of these novel agents has undoubtedly improved response rates, a proportion of patients nevertheless do not achieve ≥PR [12,13], and our data are relevant to this group. In addition, thalidomide-based induction regimens continue to be widely employed, and this is likely to remain the case in

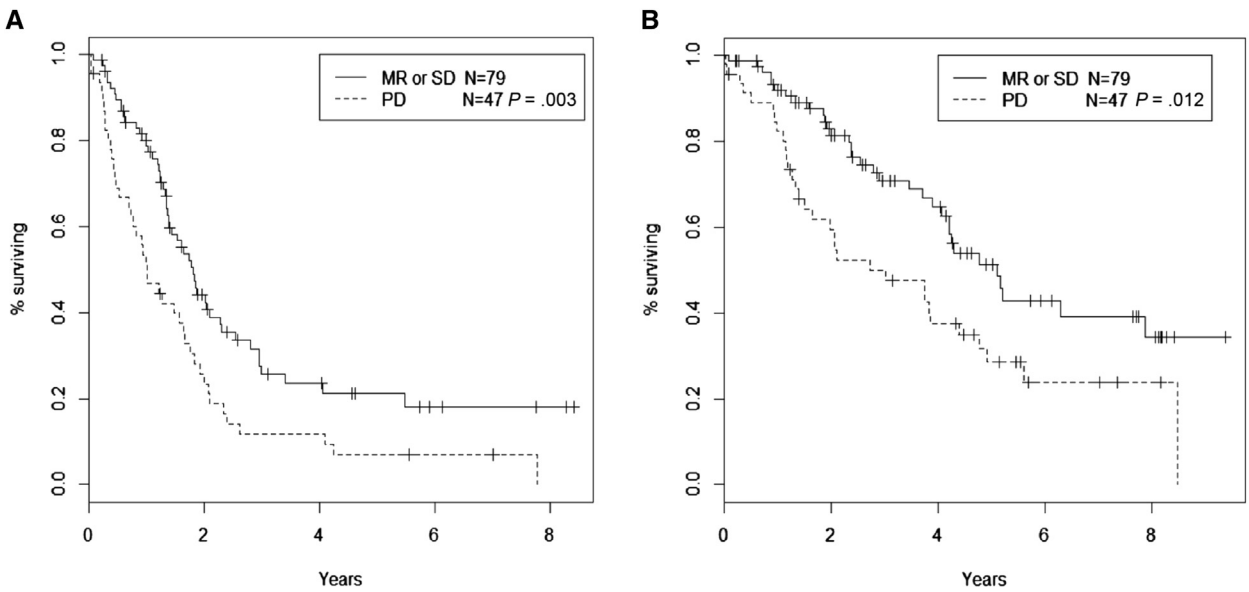

Figure 1. Kaplan-Meier estimates. (A) Shows PFS by disease status at transplantation and (B) shows OS by disease status at transplantation.

health care economies where access to newer agents will be limited outside clinical trials.

Secondly, information on maintenance therapy and subsequent salvage regimens was unavailable; cytogenetic stratification data were also not available for the majority of our patients. A major obstacle to evaluating ASCT for PRM is that the prognosis with nontransplantation therapies is also unknown. Alexanian et al. reported 27 patients receiving ASCT and 60 receiving nontransplantation therapies: ASCT improved median survival by 27 months ($P < .01$) [14]. In that era, neither induction nor salvage regimens incorporated novel agents, the numbers were small, and the control arm received suboptimal therapy for socioeconomic reasons; nevertheless, a clear benefit was shown. Early phase studies evaluating novel and experimental therapies in relapsed refractory MM are myriad, and although cautious interpretation is warranted with such heterogeneous populations, it is clear that the outcomes for this group of patients remain generally poor despite advances in therapy. For example, consider 4 recent early phase studies that have evaluated carfilzomib-lenalidomide-dexamethasone [15], pomalidomide-bortezomib-dexamethasone [16], elotuzumab-bortezomib [17], and vorinostat-bortezomib [18] in relapsed refractory populations after a median of 2 lines of prior therapy, and found PFS of 11.8 months, 7.4 months (duration of response rather than PFS), 9.7 months, and 7.6 months, respectively. Clearly such studies do not represent a comparator group for our cohort but do illustrate that even the next generation of novel agents does not mitigate the adverse prognosis of disease refractory to current induction regimes.

A prospective comparison of ASCT to modern non-ASCT therapy in patients with PRM would be extremely informative, particularly in light of the often poor durability of responses to novel agents at relapse [19]. Nonetheless, it seems fair to conclude that even with a new array of treatment modalities becoming available, the outlook for patients with PRM receiving nontransplantation therapy is poor, and in this context ASCT represents a highly attractive option for establishing disease control. In addition, use of ASCT does not preclude subsequent treatment with experimental agents, which might well be incorporated into maintenance regimens or employed at later relapse.

Our findings support the use of ASCT in myeloma patients responding suboptimally to modern induction therapies, including thalidomide-containing regimens. Patients should not be excluded on the basis of refractoriness to induction, as ASCT is effective in this group conventionally considered to have a poor outcome. Comprehensive MVA identified no disparate subgroups, meaning ASCT is a reasonable strategy for all fit primary refractory patients.

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REFERENCES

1. Child JA, Morgan GJ, Davies FE, et al. High-dose chemotherapy with hematopoietic stem-cell rescue for multiple myeloma. *N Engl J Med*. 2003;348:1875-1883.
2. Moreau P, Attal M, Pégourié B, et al. Achievement of VGPR to induction therapy is an important prognostic factor for longer PFS in the IFM 2005-01 trial. *Blood*. 2011;117:3041-3044.
3. Vose JM, Anderson JR, Kessinger A, et al. High-dose chemotherapy and autologous hematopoietic stem-cell transplantation for aggressive non-Hodgkin's lymphoma. *J Clin Oncol*. 1993;11:1846-1851.
4. Kumar S, Lacy MQ, Dispenzieri A, et al. High-dose therapy and autologous stem cell transplantation for multiple myeloma poorly responsive to initial therapy. *Bone Marrow Transplant*. 2004;34:161-167.
5. Singhal S, Powles R, Sirohi B, et al. Response to induction chemotherapy is not essential to obtain survival benefit from high-dose melphalan and autotransplantation in myeloma. *Bone Marrow Transplant*. 2002;30:673-679.
6. Bladé J, Samson D, Reece D, et al. Criteria for evaluating disease response and progression in patients with multiple myeloma treated by high-dose therapy and haemopoietic stem cell transplantation. Myeloma Subcommittee of the EBMT. European Group for Blood and Marrow Transplant. *Br J Haematol*. 1998;102:1115-1123.
7. Alexanian R, Weber D, Giralt S, et al. Impact of complete remission with intensive therapy in patients with responsive multiple myeloma. *Bone Marrow Transplant*. 2001;27:1037-1043.
8. Bladé J, Rosiñol L, Sureda A, et al. High-dose therapy intensification compared with continued standard chemotherapy in multiple myeloma patients responding to the initial chemotherapy: long-term results from a prospective randomized trial from the Spanish cooperative group PETHEMA. *Blood*. 2005;106:3755-3759.
9. Gertz MA, Kumar S, Lacy MQ, et al. Stem cell transplantation in multiple myeloma: impact of response failure with thalidomide or lenalidomide induction. *Blood*. 2010;115:2348.
10. Attal M, Harousseau JL, Stoppa AM, et al. A prospective, randomized trial of autologous bone marrow transplantation and chemotherapy in multiple myeloma. Intergroupe Français du Myélome. *N Engl J Med*. 1996;335:91-97.
11. Rosinol L, Garcia-Sanz R, Jose'Lahuerta J, et al. Benefit from autologous stem cell transplantation in primary refractory myeloma? Different outcomes in progressive versus stable disease. *Haematologica*. 2012; 97:616-621.
12. Harousseau J-L, Attal M, Avet-Loiseau H, et al. Bortezomib plus dexamethasone is superior to vincristine plus doxorubicin plus dexamethasone as induction treatment prior to autologous stem-cell transplantation in newly diagnosed multiple myeloma: results of the IFM 2005-01 phase III trial. *J Clin Oncol*. 2010;28:4621-4629.
13. Rajkumar SV, Jacobus S, Callander NS, et al. Lenalidomide plus high-dose dexamethasone versus lenalidomide plus low-dose dexamethasone as initial therapy for newly diagnosed multiple myeloma: an open-label randomised controlled trial. *Lancet Oncol*. 2010;11:29-37.
14. Alexanian R, Dimopoulos MA, Hester J, et al. Early myeloablative therapy for multiple myeloma. *Blood*. 1994;84:4278-4282.
15. Wang M, Martin T, Bensinger W, et al. Phase 2 dose-expansion study (PX-171-006) of carfilzomib, lenalidomide, and low-dose dexamethasone in relapsed or progressive multiple myeloma. *Blood*. 2013;122: 3122-3128.
16. Richardson PG, Hofmeister C, Raje NS, et al. MM-005: phase 1 trial of pomalidomide (POM), bortezomib (BORT), and low-dose dexamethasone (LoDEX [PVD]) in lenalidomide (LEN)-refractory and

proteasome inhibitor (PI)-exposed myeloma. *J Clin Oncol*. 2014;32. abstract 8589.
17. Jakubowiak AJ, Benson DM, Bensinger W, et al. Phase I trial of anti-CS1 monoclonal antibody elotuzumab in combination with bortezomib in the treatment of relapsed/refractory multiple myeloma. *J Clin Oncol*. 2012;30:1960-1965.
18. Badros A, Burger AM, Philip S, et al. Phase I study of vorinostat in combination with bortezomib for relapsed and refractory multiple myeloma. *Clin Cancer Res*. 2009;15:5250-5257.
19. Laubach JP, Mahindra A, Mitsiades CS, et al. The use of novel agents in the treatment of relapsed and refractory multiple myeloma. *Leukemia*. 2009;23:2222-2232.

Evaluation of Initial Telomere Length and Changes after Transplantation in Adult Double-Unit Cord Blood Transplant Recipients

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Cord blood (CB) leukocytes have inherent telomere length (TL) variation, and CB hematopoietic stem cells (HSC) can maintain high telomerase levels preventing telomere attrition in vitro. We evaluated TL changes in 13 adult double-unit CB transplant (CBT) recipients. In the 26 units, we observed a marked variation in CB TL at thaw (median, 9.99 kilobases [kb]; range, 6.85 to 13.5). All 13 patients engrafted. Of 11 engrafting with 1 unit, there was no correlation between unit dominance and TL (mean dominant unit TL, 8.84 kb ± 1.76; mean nonengrafting unit TL, 10.3 kb ± 1.81; $P = .77$). Serial measurements of TL up to 1 year after CBT demonstrated an overall mean 3.04 kb ± .16 TL decrease with only 1 patient exhibiting telomere maintenance. In summary, initial TL does not predict CB unit dominance. Moreover, our analysis suggests neonatal hematopoiesis makes a transition to an HSC characterized by changes in average TL and potentially low telomerase asymmetric cell division in adult CBT recipients. Further investigation of alterations in telomere length and its clinical implications after transplantation of this observation are indicated.

INTRODUCTION

Cord blood (CB) is an alternative source of allogeneic hematopoietic stem cells (HSC) for the transplantation of patients lacking suitable HLA-matched adult donors. Although CB transplantation (CBT) has the advantage of reduced stringency of required HLA match, it is limited by the low progenitor cell dose, resulting in impaired engraftment and restricting the application of CBT in larger children and adults. One strategy to extend transplantation access in adult patients is to combine 2 units in a double-unit graft [1]. Intriguingly, a single unit mediates sustained donor hematopoiesis in the majority of patients. However, the mechanisms of unit dominance have not been fully elucidated. Furthermore, although healthy long-term survivors of CBT are documented, the long-term effects of transplanting a limited number of HSC from a CB unit into adult recipients have not been fully established.

It is known that CB progenitors have a significantly higher replicative potential than adult HSC [2], although there is an inherent biological variation at birth. We have previously shown that CB demonstrates maintenance of telomere length (TL) in vitro due to persisting levels of telomerase activity for 4 to 5 months [3]. The effect of replicative stress in vivo on TL, however, is unknown. We, therefore, investigated the extent of TL variability in clinical CB units, the influence of TL upon unit dominance after CBT, and the effect of the in vivo microenvironment on the degree of neonatal TL changes during the first post-transplantation year in adult double-unit CBT recipients.

STUDY DESIGN

Thirteen patients with high-risk hematologic malignancies underwent transplantation with 4 to 6 of 6 HLA-A, -B antigen, -DRB1 allele-matched unrelated donor double-unit CB grafts after myeloablative or nonmyeloablative conditioning, as previously described [4-6]. All patients provided informed consent for transplantation in accordance with the Declaration of Helsinki and signed institutional review board–approved consent for the laboratory analysis of each CB unit and serial peripheral blood samples. Each CB unit was analyzed on the same day as clinical transplantation. Mononuclear cells (MNCs) were isolated from each unit and from peripheral blood at days 28, 100, 180, and 1 year after CBT by density gradient separation with Ficoll-Hypaque. DNA was then isolated from MNC pellets and quantified using a BioTek Synergy H1 Hybrid Multi-Mode Microplate Reader (BioTek, Winooski, VT). Southern Blot was performed using the Roche TeloTAGGG Telomere Length Assay (Roche Diagnostics GmBH, Mannheim, Germany) per the manufacturer's instructions. Clinical engraftment was evaluated after transplantation

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