Stem cell mobilization in HIV seropositive patients with lymphoma

Alessandro Re,[1] Chiara Cattaneo,[1] Cristina Skert,[2] Pascual Balsalobre,[3] Mariagrazia Michieli,[4] Mark Bower,[5] Andrés J. M. Ferreri,[6] Marcus Hentrich,[7] José M. Ribera,[8] Bernardino Allione,[9] Philipp Schommers,[10] Silvia Montoto,[11] Camillo Almici,[12] Pierino Ferremi,[12] Mario Mazzucato,[13] Salvatore Gattillo,[14] Salvatore Casari,[15] Michele Spina,[16] José L. Diez-Martin,[3] Umberto Tirelli,[16] and Giuseppe Rossi[1] on the behalf of GECAT (Cooperative European Group on AIDS and Tumors)

[1]Hematology, Spedali Civili, Brescia, Italy; [2]Stem Cell Transplantation Unit, University of Brescia, Italy; [3]Hematology. Hospital General Universitario Gregorio Marañon, Instituto de Investigación Sanitaria Gregorio Marañon, Madrid, Spain; [4]Unit of high dose chemotherapy and cellular therapies, CRO, Aviano, Italy; [5]Oncology, Chelsea and Westminster Hospital, London, UK; [6]Unit of Lymphoid Malignancies, Department of Onco-Hematology, San Raffaele Scientific Institute, Milan, Italy; [7]Hematology, Oncology and Palliative Care, Harlaching Hospital, Munich, Germany; [8]Hematology, Institut Català d'Oncologia-Hospital Germans Trias i Pujol, Badalona, Spain; [9]Hematology, Ospedale Ss. Antonio e Biagio, Alessandria, Italy; [10]First Department of Internal Medicine, University of Cologne, Germany; [11]Haemato-Oncology, Barts Cancer Institute, Queen Mary University of London, UK; [12]Immunohematology and Transfusion Medicine, Spedali Civili, Brescia, Italy; [13]Immunotransfusion Center and Clinical Analysis, CRO, Aviano, Italy; [14]Transfusion and Blood Donation Center, San Raffaele Scientific Institute, Milan, Italy; [15]Infectious Disease, Spedali Civili, Brescia, Italy; and [16]Medical Oncology A, CRO, Aviano, Italy

ABSTRACT

High-dose chemotherapy with autologous peripheral blood stem cell rescue has been reported as feasible and effective in HIV-associated lymphoma. Although a sufficient number of stem cells seems achievable in most patients, there are cases of stem cell harvest failure. The aim of this study was to describe the mobilization policies used in HIV-associated lymphoma, evaluate the failure rate and identify factors influencing mobilization results. We analyzed 155 patients who underwent attempted stem cell mobilization at 10 European centers from 2000-2012. One hundred and twenty patients had non-Hodgkin lymphoma and 35 Hodgkin lymphoma; 31% had complete remission, 57% chemosensitive disease, 10% refractory disease, 2% untested relapse. Patients were mobilized with chemotherapy + G-CSF (86%) or G-CSF alone (14%); 73% of patients collected >2 and 48% >5 x 10^6 $CD34^+$ cells/kg. Low CD4+ count and refractory disease were associated with mobilization failure. Low $CD4^+$ count, low platelet count and mobilization with G-CSF correlated with lower probability to achieve >5 x 10^6 $CD34^+$ cells/kg, whereas cyclophosphamide ≥3 g/m^2 + G-CSF predicted higher collections. Circulating $CD34^+$ cells and CD34/WBC ratio were strongly associated with collection result. HIV infection alone should not preclude an attempt to obtain stem cells in candidates for autologous transplant as the results are comparable to the HIV-negative population.

Introduction

High-dose chemotherapy (HDT) with autologous stem cell transplantation (ASCT) is a potentially curative treatment for several hematologic malignancies, including Hodgkin's lymphoma (HL) and non-Hodgkin's lymphoma (NHL), with peripheral blood as the preferred hematopoietic stem cell (SC) source.[1,2] The lowest SC dose to safely support HDT conditioning regimens in patients with lymphoma is considered to be 2 x 10^6 $CD34^+$ cells/kg[3-7] and although this is achievable in most patients, there are cases of stem cell harvest failure. In the HIV-negative population, failure rates are estimated to be between 5% and 30%, with different mobilization regimens and patient populations, and up to 60% in high-risk patients such as those exposed to fludarabine.[8-10] Indeed, there is much interest in novel agents and strategies to minimize mobilization failure.[9,11] The chance of cure for HIV-infected patients with lymphoma has greatly increased after the advent of combination antiretroviral therapy (cART) in 1996,[12,13] and, more recently, HDT with ASCT in HIV-positive patients with lymphoma has been reported to be as feasible and effective as in HIV-negative counterparts.[14-18] However, although the mechanism is not completely understood, depletion of hematopoietic progenitor cells has been described in HIV-infected subjects, as measured by reduction in long-term colony-initiating cell (LTCIC) numbers and increased rate of hematopoietic SC apoptosis.[19,20] Moreover, reduced $CD34^+$ cell mobilization using G-CSF has been reported in patients with severe immunodeficiency.[21] Several groups reported successful SC mobilization and ASCT in HIV-positive patients receiving cART as either rescue or consolidation of treatment for NHL or HL, usually in small series of selected patients. Effective antiretroviral therapy could help to correct the defective hematopoiesis and finally protect from mobilization failure.[22] In the HIV-negative patients, several parameters have been identified predicting poor SC collections (including older age, type and status of underlying hematologic disease, number and type of prior treatments, prior radiotherapy, marrow involvement and thrombocytopenia at mobilization).[23-26] Proper analyses in an HIV setting are missing. The purpose of the present study was to describe the mobilization policies used in HIV-associated lymphoma, to evaluate the failure rate, and identify factors influencing mobilization results. Moreover, the role of 'ongoing' parame-

 doi:10.3324/haematol.2013.089052
Manuscript received on April 11, 2013. Manuscript accepted on August 20, 2013.
Correspondence: sandrore@aruba.it

Table 1. Characteristics of 155 patients at first mobilization and distribution among successful mobilization and failure.

Patients' characteristic	Total number of pts	Mobilization		P^
		Successful	Failure	
	155 n. (%)	113 n. (%)	42 n. (%)	
Age, years°	42 (24-71)	41.5 (28-65)	42 (27-71)	0.76
Sex				
Male	114 (86)	89 (88)	25 (81)	0.29
Female	18 (14)	12 (12)	6 (19)	
Histopathology (WHO)				
Hodgkin lymphoma	35 (25)	23 (22)	12 (32)	0.27
DLBCL	58 (42)	45 (44)	13 (34)	0.24
Burkitt lymphoma	23 (16)	18 (18)	5 (13)	0.52
Plasmablastic lymphoma	13 (9)	8 (8)	5 (13)	0.33
Anaplastic lymphoma	6 (4)	3 (3	3 (8)	0.20
Others	5 (4)	5 (5)	0 (0)	0.16
Status of lymphoma				
Complete remission	43 (31)	35 (35)	8 (22)	0.17
Chemosensitive disease	78 (57)	60 (59)	18 (50)	0.33
Refractory disease	14 (10)	5 (5)	9 (25)	<0.001
Untested relapse	2 (2)	1 (1)	1 (3)	0.47
Bone marrow involvement				
No	107 (77)	78 (77)	29 (76)	0.91
Previous	28 (20)	22 (22)	6 (16)	0.43
At mobilization	4 (3)	1 (1)	3 (8)	0.03
Prior extended radiotherapy				
No	117 (92)	80 (90)	37 (97)	0.15
Yes	10 (8)	9 (10)	1 (3)	
Number of prior lines of CT				
1	46 (33)	38 (37)	8 (21)	0.07
2	77 (55)	53 (52)	24 (63)	0.24
> 3	17 (12)	11 (11)	6 (16)	0.42
Plt x10^9/L	199 (30-689)	206 (30-473)	85 (63-689)	0.17
PMN x10^9/L	2.7 (0.5-23.1)	2.8 (0.6-23.1)	2.6 (0.5-17.7)	0.59
Time from HIV Dx, months°	67.8 (1.6-376)	69.5 (1.6-376)	64.5 (3.4-289)	0.77
Pts on HAART				
Yes	136 (97)	99 (97)	37 (97)	0.92
No	4 (3)	3 (3)	1 (3)	
CD4+/mcL°	237 (9-1146)	245 (9-1146)	207 (64-918)	0.07
HIV-viremia				
Detectable	35 (28)	28 (30)	7 (21)	0.34
Undetectable	92 (72)	66 (70)	26 (79)	
HIV-viremia, cp/mL°	0 (0-750000)	0 (0-750000)	0 (0-896)	0.76
Type of mobilization				
CT + G-CSF	133 (86)	100 (89)	33 (79)	0.12
G-CSF alone	22 (14)	13 (11)	9 (21)	
G-CSF dosage				
5 mcg/kg	11 (8)	9 (9)	2 (5)	0.46
10 mcg/kg	107 (78)	78 (79)	29 (76)	0.75
> 10 mcg/kg	19 (14)	12 (12)	7 (19)	0.34
Mobilizing CT regimen	3 (3)	7 (22)	<0.001	
CTX > 3g/m^2	34 (26)	27 (27)	7 (22)	0.56
Platinum-based	32 (24)	23 (23)	9 (28)	0.55
Ifosfamide-based	18 (13)	16 (16)	2 (6)	0.16
Etoposide-based	13 (10)	11 (11)	2 (6)	0.43
HD ARA-C	12 (9)	10 (10)	2 (6)	0.52
Others	13 (10)	10 (10)	3 (10)	0.92

**For several parameters the sum does not add to the total due to missing values. ^Association between mobilization failure and characteristics are tested using Mann-Whitney and χ^2-test when appropriate. °Continuous variables are expressed as median value (range).*

ters (circulating pre-apheresis peripheral blood CD34+ cells and the ratio between CD34+ count/WBC count evaluated the same day) in predicting the collection outcome was assessed as potential early markers of failure.

Methods

This is a retrospective multicenter analysis of mobilization (and remobilization) attempts in HIV-positive patients with lymphoma, performed consecutively and registered in the ASCT database of 10 European centers from April 2000 to May 2012. All HIV-positive patients diagnosed with HL or NHL who were potential candidates for ASCT and who had started SC mobilizing procedures were eligible; at least one CD34+ cell measurement on peripheral blood should have been performed on the predicted day of collection.

This study is a collaborative effort within the Cooperative European Group on AIDS and Tumors (GECAT). All patients had given written informed consent to PBSC mobilization and collection either within Ethical Committee approved clinical trials or in the context of standard clinical practice.

Data regarding SC collection attempts were analyzed on clinical records to evaluate mobilization and remobilization success/failure rates and to identify predicting factors. "Mobilization failure" was defined as a collection of <2 x 10^6 CD34+ cells/kg and "optimal mobilization" when >5 x 10^6 CD34+ cells/kg were collected. The variables evaluated at the time of mobilization and considered for correlation with mobilization results (including demographic, lymphoma and HIV infection-related factors and mobilization-related parameters) and the parameters registered during mobilization attempt ("ongoing parameters") as predictors of SC yield are reported in the *Online Supplementary Appendix*. The impact of all these variables was evaluated both for the probability of mobilization failure and for the capability to achieve an optimal SC collection. A mobilization attempt was defined as "first mobilization" when patients had never undergone a mobilization attempt before or "remobilization" when patients had previously experienced a mobilization failure. Stem cell harvest by apheresis was performed according to the policies of the center concerned. Collection procedures started after a peripheral blood CD34+ cell count of <20/mcL was reached.

Statistical analysis

Standard descriptive statistics, such as median, range and proportions were used to summarize the patient sample. The χ^2 test was used to compare differences in percentage, and the Mann-Whitney U test was used to compare continuous values. Logistical regression model was used for univariate and multivariate analysis of predictor variables for minimal (>2 x 10^6 CD34+ cells/kg) and optimal (>5 x 10^6 CD34+ cells/kg) SC collection. Continuous variables were categorized as follows: each variable was first divided into 4 categories at approximately the 25th, 50th, and 75th percentile. If the Odds Ratios (OR) in 2 or more adjacent categories were not substantially different, these categories were grouped together. If no clear pattern was observed, the median was taken as the cut-off point. Variables found to be significant (P<0.1) in univariate analysis were tested in multivariate analysis, which was performed using a stepwise logistical regression model. P<0.05 was considered statistically significant.

Results

Patient population and CD34+ cells yield at first mobilization attempt

We analyzed 155 HIV-positive patients who underwent initial SC mobilization. One hundred and twenty patients had NHL and 35 HL. Thirty-one percent of patients were in complete remission, 57% had chemosensitive disease, 10% refractory disease and 2% untested relapse. The majority of patients were mobilized with CT + G-CSF (86%) and the remainder with G-CSF alone (14%). All patients but 4 were on cART (including zidovudine, which is known for hematopoietic toxicity, in 10) 27 and 2 of them started cART at mobilization. Overall, in 73% of patients (113 of 155) a collection of >2 x 10^6 CD34+ cells/kg was achieved and in 48% (74 of 155) an optimal yield (>5 x 10^6 CD34+ cells/kg) was obtained. The median number of CD34+ cells collected was 6.12 x 10^6/kg (range 2-33 x 10^6/kg), after a median of 2 aphereses (range 1-4). Interestingly, a higher number of CD34+ cells was collected after CT + G-CSF compared to G-CSF alone: 6.55 x 10^6 CD34+ cells/kg (range 2.0-33.0) and 3.85 x 10^6 CD34+

Table 2. Univariate and multivariate statistical analysis of factors influencing mobilization failure (CD34+ < 2x10^6/kg) in 155 first mobilization attempts.

Prognostic factor*	Mobilization failure	
	Univariate *P* OR (95%CI)	Multivariate *P* OR (95%CI)
Lymphoma refractory	0.001 4.8 (1.9-12.1)	0.03 3.7 (1.1-12.4)
Platelet < 160 x 10^9/L	< 0.001 3.1 (1.9-5.0)	NS
CD4+ count < 237/mcL	0.08 2.8 (1.3-5.8)	0.009 2.8 (1.3-6.2)
CTX 1.5 gr/m^2 as mobilizing tx	0.008 4.2 (1.4-12)	NS
Prior lines of therapy (> 2)	0.01 1.28 (1.06-1.55)	NS
Bone marrow disease at mobiliz.	0.03 10.4 (1.2-90.6)	NS

**Only those parameters that achieved statistical significance (P<0.05) are listed.*

Table 3. Univariate and multivariate statistical analysis of factors influencing optimal mobilization (CD34+ >5x10^6/kg) in 155 first mobilization attempts.

Prognostic factor*	Optimal mobilization	
	Univariate *P* OR (95% CI)	Multivariate *P* OR (95% CI)
Lymphoma refractory	0.04 0.34 (0.11-0.98)	NS
Platelet < 160 x 10^9/L	0.001 0.37 (0.2-0.7)	0.004 0.33 (0.1-0.7)
CD4+ count < 237/mcL	0.08 0.55 (0.30-1.05)	0.001 0.52 (0.26-0.8)
Mobilizing strategy (G-CSF *vs.* G-CSF + CT)	0.02 0.31 (0.12-0.80)	0.008 0.21 (0.07-0.7)
CTX > 3 g/m^2 as mobilizing tx	0.01 2.1 (1.20-3.80)	0.006 3.1 (1.4-6.8)
CTX 1.5 g/m^2 as mobilizing tx	0.03 0.20 (0.05-0.90)	NS

**Only those parameters that achieved statistical significance (P<0.05) are listed.*

Table 4. Circulating CD34⁺ cells, WBC and CD34/WBC ratio in 155 patients at first mobilization and their distribution among successful mobilization and failure.

Parameters	Total number of pts	Mobilization		P
	n. 155 median (range)	Successful n. 113 median (range)	Failure n.42 median (range)	
Median WBC x10⁹/L*	6.9 (0.7-97.1)	6.6 (0.7-97.1)	11.2 (0.7-85.3)	0.7
Median CD34⁺ cells/mcL*	21.2 (0-684)	32.6 (0-684)	4.4 (0.07-37)	<0.0
Median CD34/WBC ratio*	2.2 (0-59.1)	3.55 (0-59.1)	0.4 (0.1-3.8)	<0.0001

Measured at predicted day of collection (with circulating WBC of at least 1000/mcL).

cells/kg (range 2.3-31.0), respectively (P=0.01). Mobilization failure occurred in 27% of patients (42 of 155). Patients' characteristics and their distribution among successful (>2 x 10^6 CD34⁺ cells/kg) and failed (<2 x 10^6 CD34⁺ cells/kg) mobilization are reported in Table 1. Interestingly, 6 of 10 (60%) patients receiving zidovudine collected >2 x 10^6 CD34⁺ cells/kg, with optimal collection in 4 (40%).

Univariate and multivariate analysis for mobilization failure and for optimal mobilization

Logistical regression analysis of predictive factors for mobilization failure is reported in Table 2.

In univariate analysis, refractory disease, platelet count less than 160 x 10^9/L, CD4⁺ count less than 237/mcL, cyclophosphamide 1.5 g/m² as mobilizing treatment, two or more lines of prior therapies and bone marrow disease at mobilization were significantly associated with the risk of mobilization failure. In multivariate analysis, only low CD4⁺ cell count and refractory disease remained significant. Indeed, 44% of patients with CD4⁺ count below 100/mcL failed mobilization, as did 64% of patients with refractory disease. Univariate and multivarite analyses for optimal mobilization are reported in Table 3. In univariate analysis, refractory disease, platelet count <160 x 10^9/L, CD4⁺ count <237/mcL, G-CSF alone as mobilizing treatment compared to G-CSF + CT and cyclophosphamide 1.5 g/m² were associated with failure to achieve a collection of >5 x 10^6 CD34⁺ cells/kg, while cyclophosphamide >3 g/m² correlated with optimal collection. In multivariate analysis, only low CD4⁺ cell count, low platelet count and mobilization with G-CSF alone remained significantly associated with lower probability of optimal collection, whereas cyclophosphamide >3 g/m² + G-CSF remained significantly correlated with optimal collection.

Peripheral CD34⁺ cell counts and CD34⁺ cells/WBC counts ratio

Table 4 shows the peripheral blood WBC counts, circulating CD34⁺ cells and their ratio (CD34/WBC ratio) measured at predicted day of collection in the whole series of 155 first mobilization cases and their distribution in the 2 groups of mobilization success and failure. A clear difference was seen in absolute CD34⁺ count and in CD34/WBC ratio between the 2 groups. To verify if preapheresis CD34⁺ count and CD34⁺/WBC ratio could overcome the predictive significance of 'base-line parameters' on collection results, we performed the logistical regression multivariate analysis for collection failure and for optimal collection including the 'ongoing parameters'. Both CD34⁺ absolute count and CD34⁺/WBC ratio were independently associated with the probability of mobilization failure (P<0.001 for both) and CD34/WBC ratio alone (P<0.001) with the probability of optimal collection. However, low CD4⁺ count and refractory disease maintained their independent impact on mobilization failure, and low platelet count maintained its adverse impact in obtaining an optimal collection (*data not shown*). To further examine the correlation between 'ongoing parameters' and the probability of SC collection, we segregated the CD34/WBC ratio into quartiles. The lowest quartile was for ratio <0.7 and the higher for >5.4. The percentage of patients who failed SC harvest correlated with the quartile, with 68%, 24%, 9% and 0% of failure rate, respectively, for lower to higher quartile.

Table 5. Stem cell mobilization and remobilization attempts in 155 HIV positive patients.

Attempt	N pts	N success	N optimal
1	155	113	74
2	25*	9#	2*
3	3	0	
4	2	0	

Including 2 patients in whom > 2 x 10^6 CD34⁺ cells/kg had been obtained at the 1st attempt and had a second one to achieve >5. # including 2 patients in whom > 2 x 10^6 CD34⁺ cells/kg was obtained after pooling 1st + 2nd attempts.

Remobilization

A total of 42 of 155 patients failed initial mobilization and 23 of these 42 patients were remobilized, in 15 cases with CT + G-CSF, in 7 cases with G-CSF alone and in one case with plerixafor + G-CSF. Following remobilization, only 7 of 23 (30%) patients achieved at least 2 x 10^6 CD34⁺ cells/kg. Two more patients achieved >2 x 10^6 CD34⁺ cells/kg after pooling first and second attempts. Few patients proceeded to further attempts (Table 5). Among remobilization strategies, G-CSF alone and cyclophosphamide 1.5 g/m² had the highest failure rates, respectively 86% (6 of 7 patients) and 83% (5 of 6 patients), compared to 44% (4 of 9 patients) for other CT regimens plus G-CSF. The only patient who received plerixafor + G-CSF failed collection. Multivariate analysis of factors influencing failure at remobilization was limited by the small number of cases, but showed a low CD4⁺ count as the only independent significant factor correlated to failure (OR 2.9; P=0.02); however, adding the 'ongoing parameters' to the analysis, only CD34/WBC ratio resulted significantly correlated with collection (OR 0.17; P=0.03) (*data not shown*).

Then, of the whole series of 155 patients who underwent SC mobilization, 113 collected enough CD34⁺ cells

at first mobilization attempt, plus 9 after remobilization and pooling harvests (total 122 of 155, 79%), while 33 of 155 (21%) failed to collect enough cells to perform ASCT even after repeated attempts. Finally, 104 patients actually received HDT with SC rescue. Neutrophil engraftment, defined as an absolute neutrophil count >0.5 x 10^9/L, occurred in all patients at a median of 11 days (range 8-33 days). Platelet engraftment, defined as self-supporting platelet count >20 x 10^9/L, occurred in all evaluable patients (4 patients died early before platelet engraftment), except in 2 who were lost to follow up five and nine months after transplant, at a median of 14 days (range 7-455 days).

Discussion

This is the first study to address SC mobilization and collection in HIV-positive patients with lymphoma. Despite prior concerns that HIV might inhibit hematopoiesis and reduce SC mobilization,[19-21] in a series of 155 consecutive HIV-associated lymphoma, the majority of patients (73%) achieved enough CD34$^+$ cells to proceed to transplant at the first mobilization attempt and almost half (48%) reached an optimal SC collection. Moreover, engraftment kinetics in patients who actually received ASCT appear reassuring and comparable with the general HIV-negative population.[2] The 27% rate of mobilization failure, with no difference between HL and NHL, is still troublesome, but it does not appear much different from the HIV-negative counterpart, where a wide range of failure rate is reported, and might depend on different mobilization strategies and patient populations.[8-10]

The widespread use of cART, which seems capable of correcting the defective hematopoiesis of HIV-infected subjects,[22] might play a pivotal role in this. The mechanism by which antiretroviral therapy favorably impacts on hematopoiesis in HIV-infected subjects is not completely understood, since HIV does not directly infect the CD34$^+$ cells.[28] *In vitro* studies showed that Ritonavir, a protease inhibitor frequently used in antiretroviral combination treatments, markedly decreased apoptosis and increased colony formation if added to CD34$^+$ cells cultures from HIV-infected patients and even from HIV-negative controls. Thus, it has been hypothesized that cART could overcome inhibition of hematopoiesis with a mechanism unrelated to its antiviral activity.[29]

In our series, only two factors were independently associated with mobilization failure: chemo-refractory disease and a low CD4$^+$ count, which can be regarded as the expression of patients' immune status and of responsiveness to antiretroviral therapy. Numerous previous studies have demonstrated the influence of patients' immune status (expressed by the number of CD4$^+$ cells) on clinical aggressiveness of HIV-associated lymphoma, response to treatment and risk of relapse.[30-32] In addition, a recent study from the GICAT (Gruppo Italiano Cooperativo AIDS e Tumori) showed a correlation between CD4$^+$ count and the chance for HIV-positive patients with lymphoma to receive HDT and SC transplantation in the salvage setting.[33] The present study further shows that the CD4$^+$ count correlates even with the possibility of mobilizing and harvesting hematopoietic SC.

Our data also emphasize that different methods of hematopoietic SC mobilization are in use without an established standard. The majority of patients were mobilized after CT + G-CSF (with a wide variety of CT regimens) and only a small proportion received G-CSF alone. Mobilization after CT + G-CSF, a strategy widely used in Europe, has, in this study, allowed higher CD34$^+$ cell yields compared to G-CSF alone, as previously reported in the general HIV-negative population.[34] While the use of G-CSF alone has been advocated to reduce toxicity and costs,[10] adding chemotherapy to G-CSF as mobilizing agent allows further tumor cytoreduction and *in vivo* purging of mobilized tumor cells, besides allowing higher CD34 yield.[9,35] Even if a target of 2 x 10^6/Kg CD34$^+$ cells is considered appropriate to ensure engraftment after myeloablative treatment, in the HIV-negative population, a higher amount of CD34$^+$ cells reinfused has a favorable impact on patient's outcome, mainly in terms of prompt engraftment.[36,37] Such a correlation between CD34$^+$ cell dose and quality of engraftment has been suggested for both neutrophil and platelet delayed engraftment also in a small series of HIV-associated lymphoma.[17] Thus, achieving an optimal CD34 collection, defined as a CD34 cell number >5 x 10^6/kg, might be preferred, particularly in a subset of patients, such as HIV-positive patients with lymphoma, potentially at higher risk of delayed engraftment and infectious complication.

Other than the use of G-CSF alone, a lower likelihood of achieving an optimal SC collection was predicted in our series by a low platelet count at the time of mobilization, a potential indicator of a reduced bone marrow reserve, which is a well known predictor of poor collection also in HIV-negative populations.[23-26] Moreover, the use of cyclophosphamide at the dosage of 1.5 g/m^2 as mobilizing regimen predicted a lower yield of CD34$^+$ cells as already reported in lymphoma series in the general population.[38] On the other hand, high-dose cyclophosphamide (at least 3 g/m^2) correlated with higher CD34$^+$ cell collection compared to the other CT regimens.

Cyclophosphamide monotherapy was the most used regimen in this series, and its mobilization potential is well known, since it was the traditional regimen employed in seminal studies on PBSC collection.[39] Its retrospective comparison with other CT mobilizing programs in our study is hampered by the wide variety of regimens used, which leads to an excessive fragmentation of data.

On the whole, our study shows that, as in the HIV-negative population, also in HIV-positive subjects the identified prognostic factors, available before starting mobilization, were not strong enough to accurately predict the collection result. The impact of pre-apheresis peripheral blood CD34$^+$ cell count and CD34$^+$/WBC ratio was also evaluated as potentially useful ongoing indicators of collection success or failure. Indeed, both CD34$^+$ cell count and CD34$^+$/WBC ratio resulted strongly associated to subsequent SC yield. Given the recent availability of mobilizing agents like plerixafor, whose use 'on demand' might boost 'ongoing' CD34$^+$ cell mobilization, in cases at risk of failure such parameters may be shown to be clinically useful. Accordingly, within the HIV-negative population, standard definition of "predicted poor mobilizers" has been recently proposed to identify patients who may potentially benefit from early intervention with new mobilizing agents.[40] While the results of plerixafor in HIV-negative populations seem encouraging,[9,10,11,41] no experiences have been reported so far on its use as mobilizing

agent in HIV-positive patients, although it was originally introduced as a potentially useful anti-HIV agent.[42] Our results would support a trial on plerixafor in HIV-positive subjects predicted to be at high risk of mobilization failure. The rarity of the use of plerixafor in the present series might lie in the small number of patients who received a second mobilization attempt and on the limited availability of this agent in Europe during the study period. Only 55% of our patients who failed first mobilization went forward for a second attempt, and overall, only 21% reached a sufficient number of $CD34^+$ cells after the second attempt. That means that HIV-positive patients failing a first mobilization are actually very unlikely to proceed to autologous transplant. There could be several reasons for this, including early disease progression, but it is also due to the difficulty of harvesting SC after a first collection failure.

Taken together, our data suggest that HIV should not preclude an attempt to obtain SC in candidates for autologous transplant as the results are comparable to the HIV-negative population. CT + plus G-CSF seems to mobilize better than G-CSF alone and, if using cyclophosphamide, at least 3 g/m^2 seems recommendable. However, we need to optimize current mobilization protocols, and more effective mobilization agents are welcome in HIV-positive patients, as in the general population. Improving antiretroviral therapy is highly advisable even to improve SC collection, and a better understanding of variables associated with mobilization success may further optimize SC collection. The results of this study might help to decide optimal mobilizing strategy in HIV-related lymphoma and could provide the framework for a rational investigation into the use of new mobilizing agents.

Authorship and Disclosures

Information on authorship, contributions, and financial & other disclosures was provided by the authors and is available with the online version of this article at www.haematologica.org.

References

1. Schmitz N, Buske C, Gisselbrecht C. Autologous stem cell transplantation in lymphoma. Semin Hematol. 2007;44(4):234-45.
2. Schmitz N, Linch DC, Dreger P, Goldstone AH, Boogaerts MA, Ferrant A, et al. Randomised trial of filgrastim-mobilised peripheral blood progenitor cell transplantation versus autologous bone-marrow transplantation in lymphoma patients. Lancet. 1996;347(8998):353-7.
3. Siena S, Schiavo R, Pedrazzoli P, Carlo-Stella C. Therapeutic relevance of CD34 cell dose in blood cell transplantationfor cancer therapy. J Clin Oncol. 2000; 18(6):1360-77.
4. Solà C, Maroto P, Salazar R, Mesía R, Mendoza L, Brunet J, et al. Bone Marrow Transplantation: Prognostic Factors of Peripheral Blood Stem Cell Mobilization with Cyclophosphamide and Filgrastim (r-metHuG-CSF): The CD34+ Cell Dose Positively Affects the Time to Hematopoietic Recovery and Supportive Requirements after High-Dose Chemotherapy. Hematology. 1999; 4(3):195-209.
5. Bender JG, To LB, Williams S, Schwartzberg LS. Defining a therapeutic dose of peripheral blood stem cells. J Hematother. 1992;1(4):329-41.
6. Gandhi MK, Jestice K, Scott MA, Bloxham D, Bass G, Marcus RE. The minimum CD34 threshold depends on prior chemotherapy in autologous peripheral blood stem cell recipients. Bone Marrow Transplant. 1999;23(1):9-13.
7. To LB, Haylock DN, Simmons PJ, Juttner CA. The biology and clinical uses of blood stem cells. Blood. 1997;89(7):2233-58.
8. Pusic I, Jiang SY, Landua S, Uy GL, Rettig MP, Cashen AF, et al. Impact of mobilization and remobilization strategies on achieving sufficient stem cell yields for autologous transplantation. Biol Blood Marrow Transplant. 2008;14(9):1045-56.
9. Gertz MA. Current status of stem cell mobilization. Br J Haematol. 2010; 150(6): 647-62.
10. To LB, Levesque JP, Herbert KE. How I treat patients who mobilize hematopoietic stem cells poorly. Blood. 2011;118(17):4530-40.
11. Pusic I, DiPersio JF. The use of growth factors in hematopoietic stem cell transplantation. Curr Pharm Des. 2008;14(20):1950-61.
12. Levine AM. Acquired Immunodeficiency syndrome-related lymphoma: clinical aspects. Semin Oncol. 2000;27(4):442-53.
13. Palella FJ, Delaney KM, Moorman AC, Loveless MO, Fuhrer J, Satten GA, et al. Declining morbidity and mortality among patients with advanced human immunodeficiency virus infection. N Engl J Med. 1998;338(13):853-60
14. Gabarre J, Azar N, Autran B, Katlama C, Leblonde V. High-dose therapy and autologous haematopoietic stem-cell transplantation for HIV-1-associated lymphoma. The Lancet. 2000;355(9209):1071-2
15. Re A, Cattaneo C, Michieli M, Casari S, Spina M, Ruolo M, et al. High-dose therapy and autologous peripheral-blood stem cell transplantation as salvage treatment for HIV associated lymphoma in patients receiving highly active antiretroviral therapy. J Clin Oncol. 2003;21(23):4423-7.
16. Krishnan A, Molina A, Zaia J, Smith D, Vasquez D, Kogut N, et al. Durable remissions with autologous stem cell transplantation for high-risk HIV-associated lymphomas. Blood. 2005;105(2):874-8.
17. Serrano D, Carrion R, Balsalobre P, Miralles P, Berenguer J, Buno I, et al. HIV associated lymphoma successfully treated with peripheral blood stem cell transplantation. Exp Hematol. 2005;33(4):487-94.
18. Diez-Martín JL, Balsalobre P, Re A, Michieli M, Ribera JM, Canals C, et al; European Group for Blood and Marrow Transplantation Lymphoma Working Party. Comparable survival between HIV+ and HIV- non-Hodgkin and Hodgkin lymphoma patients undergoing autologous peripheral blood stem cell transplantation. Blood. 2009;113(23):6011-4.
19. Sloand EM, Young NS, Sato T, Kumar P, Kim S, Weichold FF, et al. Secondary colony formation after long-term bone marrow culture using peripheral blood and bone marrow of HIV-infected patients. AIDS. 1997;11(13):1547-53.
20. Zauli G, Capitani S. HIV-1-related mechanisms of suppression of CD34+ hematopoietic progenitors. Pathobiology. 1996;64(1):53-8.
21. Schooley RT, Mladenovic J, Sevin A, Chiu S, Miles SA, Pomerantz RJ, et al. Reduced mobilization of CD34+ stem cells in advanced human immunodeficiency virus type 1 disease. J Infect Dis. 2000; 181(1):148-57.
22. Baillou C, Simon A, Leclercq V, Azar N, Rosenzwajg M, Herson S, et al. Highly active antiretroviral therapy corrects hematopoiesis in HIV-1 infected patients: interest for peripheral blood stem cell-based gene therapy. AIDS. 2003;17(4):563-74.
23. Micallef IN, Apostolidis J, Rohatiner AZ, Wiggins C, Crawley CR, Foran JM, et al. Factors which predict unsuccessful mobilisation of peripheral blood progenitor cells following G-CSF alone in patients with non-Hodgkin's lymphoma. Hematol J. 2000;1(6):367-73.
24. Hosing C, Saliba RM, Ahlawat S, Körbling M, Kebriaei P, Alousi A, et al. Poor hematopoietic stem cell mobilizers: a single institution study of incidence and risk factors in patients with recurrent or relapsed lymphoma. Am J Hematol. 2009;84(6):335-7.
25. Wuchter P, Ran D, Bruckner T, Schmitt T, Witzens-Harig M, Neben K, et al. Poor mobilization of hematopoietic stem cells-definitions, incidence, risk factors, and impact on outcome of autologous transplantation. Biol Blood Marrow Transplant. 2010;16(4):490-9.
26. Sancho JM, Morgades M, Grifols JR, Juncà J, Guardia R, Vives S, et al. Predictive factors for poor peripheral blood stem cell mobilization and peak CD34(+) cell count to guide pre-emptive or immediate rescue mobilization. Cytotherapy. 2012;14(7):823-9.
27. Walker RE, Parker RI, Kovacs JA, Masur H, Lane HC, Carleton S, et al. Anemia and erythropoiesis in patients with the acquired immunodeficiency syndrome (AIDS) and Kaposi sarcoma treated with zidovudine. Ann Intern Med. 1988;108(3):372-6.
28. Neal TF, Holland HK, Baum CM, Villinger F,

Ansari AA, Saral R, et al. CD34+ progenitor cells from asymptomatic patients are not a major reservoir for human immunodeficiency virus-1. Blood. 1995;86(5):1749-56.
29. Sloand EM, Maciejewski J, Kumar P, Kim S, Chaudhuri A, Young Neal. Protease inhibitors stimulate hematopoiesis and decrease apoptosis and ICE expression in CD34+ cells. Blood. 2000;96(8):2735-9.
30. Mounier N, Spina M, Gabarre J, Raphael M, Rizzardini G, Golfier JB, et al. AIDS-related non-Hodgkin lymphoma: final analysis of 485 patients treated with risk-adapted intensive chemotherapy. Blood. 2006;107 (10):3832-40.
31. Rossi G, Donisi A, Casari S, Re A, Cadeo G, Carosi G. Cancer. The International Prognostic Index can be used as a guide to treatment decisions regarding patients with human immunodeficiency virus-related systemic non-Hodgkin lymphoma. Cancer. 1999;86(11):2391-7.
32. Tedeschi R, Bortolin MT, Bidoli E, Zanussi S, Pratesi C, Vaccher E, et al. Assessment of immunovirological features in HIV related non-Hodgkin lymphoma patients and their impact on outcome. J Clin Virol. 2012; 53(4):297-301.
33. Re A, Michieli M, Casari S, Allione B, Cattaneo C, Rupolo M, et al. High dose therapy and autologous peripheral blood stem cell transplantation as salvage treatment for HIV-associated lymphoma: long term results of the GICAT study with analysis of prognostic factors. Blood. 2009; 114(7):1306-13.
34. Narayanasami U, Kanteti R, Morelli J, Klekar A, Al-Olama A, Keating C, et al. Randomized trial of filgrastim versus chemotherapy and filgrastim mobilization of hematopoietic progenitor cells for rescue in autologous transplantation. Blood. 2001; 98(7):2059-64.
35. Meldgaard KL, Jensen L, Gaarsdal E, Nikolaisen K, Johnsen HE. A comparative study of sequential priming and mobilisation of progenitor cells with rhG-CSF alone and high-dose cyclophosphamide plus rhG-CSF. Bone Marrow Transplant. 2000; 26(7):717-22.
36. Schulman KA, Birch R, Zhen B, Pania N, Weaver CH. Effect of CD34(+) cell dose on resource utilization in patients after high-dose chemotherapy with peripheral-blood stem-cell support. J Clin Oncol. 1999; 17(4):1227-33.
37. Weaver CH, Hazelton B, Birch, R, Palmer P, Allen C, Schwartzberg L, West W. An analysis of engraftment kinetics as a function of the CD34 content of peripheral blood progenitor cell collections in 692 patients after the administration of myeloablative chemotherapy. Blood. 1995; 86(10):3961-9.
38. Ahn JS, Park S, Im SA, Yoon SS, Lee JS, Kim BK, et al. High-dose versus low-dose cyclophosphamide in combination with G-CSF for peripheral blood progenitor cell mobilization. Korean. J Inter Med. 2005; 20(3):224-31.
39. Tarella C, Zallio F, Caracciolo D, Cherasco C, Bondesan P, Gavarotti P, et al. Hematopoietic progenitor cell mobilization and harvest following an intensive chemotherapy debulking in indolent lymphoma patients. Stem Cells. 1999;17(1):55-61.
40. Olivieri A, Marchetti M, Lemoli R, Tarella C, Icone A, Lanza F, et al. Proposed definition of "poor mobilizer" in lymphoma and multiple myeloma: an analytic hierarchy process by ad hoc working group Gruppo Italiano Trapianto di Midollo Osseo. Bone Marrow Transplant. 2012;47(3):342-51.
41. DiPersio JF, Micallef IN, Stiff PJ, Bolwell BJ, Maziarz RT, Jacobsen E, et al. Phase III prospective randomized double-blind placebo-controlled trial of plerixafor plus granulocyte colony-stimulating compared with placebo plus granulocyte-colony-stimulating factor for autologous stem-cell mobilization and transplantation for patients with non-Hodgkin's lymphoma. J Clin Oncol. 2009;27(28):4767-73.
42. Hendrix CW, Collier AC, Lederman MM, Schols D, Pollard RB, Brown S, et al. Safety, pharmacokinetics, and antiviral activity of AMD3100, a selective CXCR4 receptor inhibitor, in HIV-1 infection. J Acquir Immune Defic Syndr. 2004;37(2):1253-62.