

A combination of dexamethasone, cyclophosphamide, etoposide, and cisplatin is less toxic and more effective than high-dose cyclophosphamide for peripheral stem cell mobilization in multiple myeloma

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Background and Objectives. The purpose of this study was to compare the efficacy and toxicity of two regimens for peripheral blood stem cell (PBSC) mobilization in multiple myeloma (MM) patients.

Design and Methods. From 1995 to 2001, 116 patients were enrolled in two high-dose programs including autologous transplantation, adopting two mobilizing regimens: 61 patients were mobilized with high-dose cyclophosphamide (HD-Cy) at 4 g/m² (group I), and 55 patients with DCEP (dexamethasone, cyclophosphamide, etoposide, and cisplatin) (group II), both followed by granulocyte colony-stimulating factor (G-CSF 5 µg/Kg/day) started 48 hours after chemotherapy.

Results. The median number of CD34⁺ cells harvested was similar in the two groups (5.9 vs 5.82×10⁶ cells/kg). The target of at least 4×10⁶ cells/kg was reached in a higher percentage of patients in the DCEP group (75 vs 59%) ($p=0.05$). The proportion of poor mobilizers (<2×10⁶ CD34⁺ cells/kg) was 21% with HD-Cy and 13% with DCEP ($P=NS$). In group I, 10 patients (16%) required packed red cell transfusions, 5 patients (8%) platelet support, and the majority of patients (87%) had a neutrophil count below 500/µL, whereas none did so in group II ($p=0.0009$, $p=0.01$, $p=0.0009$, respectively). Neutropenia-related fever occurred in 18% of patients in group I versus 0% in group II ($p=0.0005$). WHO grade >II extra-hematologic toxicities (microhematuria, cystitis, infections) were seen in 8 patients (13%) of group I vs 0 in group II ($p=0.007$).

Interpretation and Conclusions. DCEP is a better tolerated and more effective regimen than HD-Cy for peripheral stem cell mobilization in MM patients assigned to high-dose therapy programs.

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Key words: multiple myeloma, stem cell mobilization, DCEP, high-dose cyclophosphamide.

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High-dose chemotherapy followed by autologous rescue with peripheral blood stem cells (PBSC) is the standard therapy for multiple myeloma (MM) patients. Clinical studies on large populations of MM patients have shown that: 1) high-dose therapy followed by PBSC improves event-free and overall survival; 2) higher numbers of PBSC allow a more rapid engraftment with a lower incidence of transplant-related morbidity and mortality;¹⁻³ 3) a variable proportion of patients, so-called *poor mobilizers*, fail mobilization or yield numbers of PBSC inadequate to perform high-dose treatments safely. High-dose cyclophosphamide (HD-Cy) (4 or 7 g/m²) in association with G-CSF is the most used regimen to mobilize PBSC³⁻⁵ even though its toxicity is not negligible, and the anti-tumor activity is scarce.^{5,6} The proportion of *poor mobilizers* after HD-Cy ranges from 10 to 20%.^{5,7,8} To reduce the percentage of *poor mobilizers*, several studies have been designed to identify factors predictive of good yields of PBSC. In a cohort of patients mobilized with HD-Cy we previously defined a scoring system able to predict the risk of inadequate collection of CD34⁺ cells. We showed that a long therapeutic history and considerable tumor involvement of bone marrow are predictive of inadequate collections.⁹ More intensive regimens have been used to increase the yield of CD34⁺ cells¹⁰⁻¹² and, at the same time, to reduce the tumor burden. Many of these regimens are, however, accompanied by an increase of therapy-related toxicity. We have recently demonstrated that the combination of dexamethasone, cyclophosphamide, etoposide and cisplatin (DCEP), in addition to its known anti-tumor activity, is an effective regimen for mobilizing high numbers of peripheral blood progenitor cells, with negligible side-effects.¹³ In this study we analyzed the efficacy and toxicity of DCEP as mobilizing therapy in 55 MM patients and compared their results with those of 61 patients who received cyclophosphamide 4 g/m² as the mobilizing regimen.

Design and Methods

Patients

From 1996 to 2001, 116 MM patients consecutively observed in our Clinic were enrolled in high-dose programs including autologous transplantation. Two mobilizing regimens were used: from 1996 to 1999 patients (n=61) were mobilized with HD-Cy (group I); from January 2000 we adopted the DCEP protocol as the mobilizing regimen in all MM patients for whom high-dose therapy was indicated (n=55; group II). Criteria for inclusion in the high-dose program were: Durie and Salmon stage II, III, or I in progression from a previous monoclonal gammopathy of unknown significance, normal liver and renal function; no severe cardiac or pulmonary disease. The age limit was <60 years for HD-Cy and was increased to 65 years for DCEP.

In both groups the therapeutic history and the following clinical and laboratory characteristics were registered at the start of the mobilizing therapy: age, stage, interval from first treatment to mobilizing therapy, previous exposure to alkylating agents, type of M component, Bence-Jones proteinuria, skeletal involvement, white blood cell (WBC) and platelet counts, hemoglobin level, bone marrow plasmacytosis, and serum creatinine.

PBSC mobilization and collection

In patients of group I, HD-Cy 4 g/m² was administered in two fractions over 24 hours followed by granulocyte colony-stimulating factor (G-CSF 5 µg/kg/day) started 48 hours after chemotherapy until the last day of leukapheresis.

To avoid the risk of hemorrhagic cystitis, hyperhydration (4 L continuous infusion over 24 hours) was commenced 12 hours before starting cyclophosphamide and continued for 12 hours after the second fraction of cyclophosphamide. Mesna (Uromitexan, Asta Medica) was given in four fractions half an hour before and after each infusion of cyclophosphamide.

Group II patients received D-CEP as their mobilizing regimen according to the following schedule: dexamethasone 40 mg for 4 days and 4-day continuous infusion of daily doses of cyclophosphamide 400 mg/m², etoposide 40 mg/m² and cisplatin 10 mg/m²; G-CSF at 5 µg/kg/day was started 48 hours after the end of the infusion. PBSC were collected with a continuous-flow blood cell separator Spectra (COBE BCT), processing a total volume of 2-3 blood mass volumes per leukapheresis.

Flow cytometry

Aliquots of the leukapheresis products were incubated with phycoerythrin-conjugated monoclonal anti-CD34 (HPCA2, Becton Dickinson) for 15 minutes at 20°C, then lysed with a lyse-no-wash standard assay and finally incubated for 10 minutes at 4°C. Cells were processed with a Becton Dickinson FACSsort analyzer. Data acquisition was performed with Cellquest software (Becton Dickinson).

Cryopreservation

Cells, diluted in dimethylsulfoxide (DMSO) solution at a final DMSO concentration of 10%, were frozen to -160°C by means of a KRYO II Series freezer (Planer R203) at a controlled rate and then stored in the liquid phase of nitrogen.

Statistical analysis

Categorical variables were summarized as count and relative frequency, while numerical variables were summarized as median and range. Pearson's χ^2 test and, when possible, Fisher's exact test were used to compare the distribution of categorical variables (e.g. stage and disease status at mobilization, previous alkylating chemotherapy, CD34⁺ count $\geq 2 \times 10^6$ /kg or $\geq 4 \times 10^6$ /kg and all toxicity parameters) between treatment groups. Tests for significant between-group differences in numerical variables (e.g. months from first treatment to mobilization, days from start of chemotherapy to first apheresis, CD34⁺ cell count) were performed using non-parametric one-way analysis of variance techniques, namely the Kruskal-Wallis ANOVA by ranks test. The significance level of *p*-values was set at 0.05. All computations and analyses were performed using STATISTICA for Windows 5.5, StatSoft, Inc. (2000).

Results

Patients

The characteristics of the patients are detailed in Table 1. A significantly higher number of patients with stage III MM was present in the DCEP group than in the HD-Cy one. The higher percentage of patients with disease refractory to previous treatments in the HD-Cy group was not statistically significant. The number of patients previously exposed to alkylating agents and the median months from first treatment to mobilization did not differ in the two groups.

Behavior of leukocytes and CD34⁺ cells

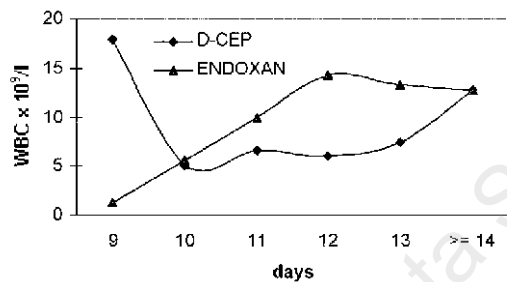
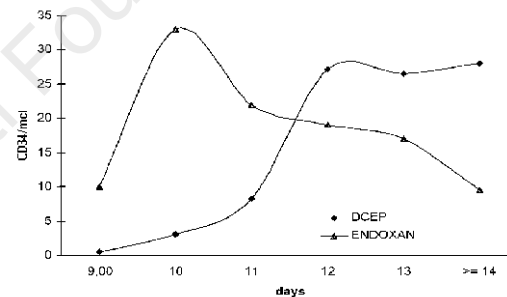
Figures 1 and 2 show the different behaviors of the median values of leukocytes and CD34⁺ cells during the mobilization period in the two groups.

Table 1. Characteristics of the patients at the start of mobilizing therapy.

	HD-Cy (Group I)	DCEP (Group II)
N. of patients	61	55
Male/female	40/21	30/25
Median age (range)	51 (31-61)	54 (30-68)
Myeloma subtype		
IgG	39	38
IgA	17	10
Light chain	4	6
Non-secretory	1	1
Disease status at mobilization		
Responsive	52	9
Refractory	39	16
Months from first treatment to mobilization (range)	4 (0-81)	4 (2-54)
Patients previously treated with alkylating agents	17 (28%)	15 (27%)

Table 2. Characteristics of PBSC collections in the two groups of patients.

	HD-Cy (Group I)	DCEP (Group II)
N. of patients	61	55
N. of apheresis procedures	127	102
N. of procedures per patient	2 (1-6)	2 (1-4)
Patients failing PBSC collection	13 (21%)	7 (13%)
N. of patients yielding $\geq 2 \times 10^6$ /kg	48 (87%)	48 (78%)
CD34 ⁺ cells		
with 1 apheresis	9 (19%)	8 (17%)
with 2 aphereses	27 (56%)	26 (54%)
with >2 aphereses	12 (25%)	14 (29%)
N. of patients yielding $\geq 4 \times 10^6$ /kg CD34 ⁺ cells	36 (59%)	41 (75%)
Days from start of chemotherapy to first apheresis	10 (8-19)	13 (11-15)
N. of CD34 ⁺ cells $\times 10^6$ /kg (range)	5.9 (0.4-19.8)	5.82 (2.1-22.4)

**Figure 1. Comparison of the behavior of leukocyte counts after DCEP and after HD-Cy.****Figure 2. Comparison of the behavior of CD34⁺ cells after DCEP and after HD-Cy.**

In group I, leukocytes, after dropping to less than 1,000/ μ L, rose above this value between day 10 and 11. By contrast, in group II leukocytes, initially increased from baseline values, fell around day 10 (never below 3,000/ μ L), then increased slowly. The behavior of CD34⁺ values also differed in the two groups. The curve representing CD34⁺ values in patients treated with HD-Cy had a narrow peak around day 10. In patients receiving DCEP, the rise of CD34⁺ cells occurred later than in the HD-Cy group (13 days vs 10 days) and their level remained stable for more days.

PBSC mobilization

The characteristics of the leukaphereses are detailed in Table 2. The median number of CD34⁺ cells harvested and the median number of leuka-

phereses per patient were similar in the two groups. In both groups the first collection was performed when the number of peripheral blood CD34⁺ cells reached a count of at least 20/ μ L, with the target of collecting 4×10^6 CD34⁺ cells/kg in each procedure. The percentages of patients yielding more than 2×10^6 CD34⁺ cells/kg were similar in the two groups (78% in group I vs 87% in group II; $p=0.1$), while the number of patients from whom more than 4×10^6 cells/kg were harvested was significantly higher in the DCEP group (75% vs 59%; $p=0.05$). The number of *poor mobilizers* was higher in the HD-Cy group than in the DCEP group (21% vs 13%). The difference did not, however, reach statistical significance. Among *poor mobilizers*, 7 of 13 (54%) in the HD-Cy group and 5 of 7 (71%) in

the DCEP group had been previously exposed to alkylating agents. Similar numbers of patients with a history of exposure to alkylators were successfully mobilized with the two regimens (10/17, or 59% for HD-Cy and 10/15, or 67% for DCEP).

Toxicities of the mobilization regimens

Differently from HD-Cy, DCEP was always well tolerated, and except for the days of continuous infusion of the drugs, patients were managed in an ambulatory care setting. The comparison of the cumulative periods of hospitalization (including the days of drug infusion) between the two groups (DCEP vs HD-Cy) showed that patients mobilized with HD-Cy spent a significantly longer period in bed ($p=0.0000$). Therapy-related transfusion requirements were also significantly different between the two groups. In group I 10 patients (16%) needed packed red cell transfusions and 5 patients (8%) needed platelet transfusion. No patient in group II required transfusion of blood products.

HD-Cy patients showed a significantly higher incidence of hematologic toxicity than did DCEP patients. The majority of patients treated with HD-Cy (53 patients, 87%) had a neutrophil count $<500/\mu\text{L}$ whereas none did so in the DCEP group ($p=0.0009$). As a consequence, 18% of patients treated with HD-Cy experienced fever, but none did in the DCEP group. After HD-Cy an overall number of 58 patients (95%) needed to be hospitalized: 53 because of severe neutropenia (neutrophil count $<500/\mu\text{L}$) and five for other reasons (bone pain, profound fatigue). By contrast, no admissions were registered among the DCEP group. One patient treated with HD-Cy developed disseminated *Herpes zoster* virus infection after chemotherapy which

hampered the CD34⁺ cell collection.

Thrombocytopenia (platelets $<50,000/\mu\text{L}$) was seen in 20 patients (33%) of group I while no group II patients showed platelet numbers below this level after DCEP. Extra-hematologic toxicity, though mostly moderate, was superior in group I (micro-hematuria, cystitis). As expected, nausea was more frequent in the DCEP group but it was always tolerable and well controlled by antiemetic drugs.

Discussion

High-dose therapy supported by autologous PBPC rescue represents the standard front-line therapy for multiple myeloma.^{1-3,14} The mobilizing phase therefore plays a central role in therapeutic programs for MM. HD-Cy is the most used mobilizing therapy, even though other combinations have been reported to be effective.¹⁰⁻¹² We recently showed that the DCEP combination, in addition to its anti-tumor effect, allows collection of adequate numbers of peripheral blood progenitor cells with very mild side-effects.¹³ The aim of this study was to compare the mobilizing efficiency and related toxicities of HD-Cy and DCEP. The analysis was performed on two comparable cohorts of MM patients included in two consecutive programs of high-dose therapy with PBSC rescue. In these programs, the collection of at least 4×10^6 CD34⁺ cells/kg was considered the optimal value for autologous support after high-dose therapy.

In the present study, DCEP showed a higher mobilizing capacity than HD-Cy with less toxicity. In fact, the target of 4×10^6 CD34⁺ cells/kg was reached in a higher percentage of patients in the DCEP group (75% vs 59%) ($p=0.05$). The efficacy of HD-Cy in our study is inferior with respect to that of some reported in the literature,¹⁵ but higher with respect to others.⁵

DCEP was always well tolerated and feasible; no patients experienced severe neutropenia, no febrile complications were registered, and no therapy-related transfusions were required. The period of hospitalization was limited to the days of DCEP infusion. In contrast, all the patients treated with HD-Cy needed hospitalization for severe neutropenia and infectious complications. A significant proportion of patients required transfusion support. Of note, among HD-Cy patients there was a case of disseminated *Herpes zoster* infection which hampered the mobilization. The behavior of the leukocytes, depicted in Figure 1, is consistent with these results. In fact, the leukocyte count after HD-Cy, unlike after DCEP, always dropped under the value of $1 \times 10^9/\text{L}$, which might be related to

Table 3. Toxicity of PBPC mobilization.

	HD-Cy (Group I)	DCEP (Group II)	p
Cumulative period of hospitalization (days)	8 (5-18)	4	0.0000
Pts. needing hospitalization after CHT	58 (95%)	0	0.0009
Pts. with neutrophils $< 500/\mu\text{L}$	53 (87%)	0	0.0009
Pts. with platelets $< 50,000/\mu\text{L}$	20 (33%)	0	0.0009
Pts. with hyperthermia	11 (18%)	0	0.0005
Pts. needing erythrocyte support	10 (16%)	0	0.0011
Pts. needing platelet support	5 (8%)	0	0.0371
Pts. with extrahematologic toxicity	8 (13%)	0	0.0023
Pts. with infections	10 (16%)	0	0.0011
Major	1 (10%)	-	-
Moderate	8 (80%)	-	-
Minor	1 (10%)	-	-

greater toxicity on myelopoiesis.

In conclusion, DCEP is more effective than HD-Cy in mobilizing peripheral blood stem cells in myeloma patients. At the same time it is well tolerated, safe, and overall more feasible than cyclophosphamide. DCEP therefore represents a good alternative as a mobilizing regimen in multiple myeloma patients assigned to high-dose therapy programs.

Contributions and Acknowledgments

AC: primary responsibility for the conception and design of the study and together with LA and SC for the interpretation of the data and writing the manuscript; PZ, SM, AL, CR were responsible for the collection of data and contributed to the analysis of the data; CP performed the statistical analyses; MAM and DT were responsible for the laboratory analysis procedures; EM and ML contributed to the revision of the manuscript. LA, SC: responsible for tables; CP; responsible for the figures.

Disclosures

Conflict of interest: none.

Redundant publications: no substantial overlapping with previous papers.

References

1. Attal M, Harousseau JL, Stoppa AM, Sotto JJ, Fuzibet JG, Rossi JF, et al. Prospective, randomised trial of autologous bone marrow transplantation and chemotherapy in multiple myeloma. *N Engl J Med* 1996; 335:91-7.
2. Barlogie B, Jagannath S, Vesole DH, Naucke S, Cheson B, Mattox S, et al. Superiority of tandem autologous transplantation over standard therapy for previously untreated multiple myeloma. *Blood* 1997; 89:789-93.
3. To LB, Roberts MM, Haylock DN, Dyson PG, Branford AL, Thorp D, et al. Comparison of haematological recovery times and supportive care requirements of autologous recovery phase peripheral blood stem cell transplants, autologous bone marrow transplants and allogeneic bone marrow transplants. *Bone Marrow Transplant* 1992; 9: 277-84.
4. Kotasek D, Shepherd KM, Sage RE, Dale BM, Norman JE, Charles P, et al. Factors affecting blood stem cells collections following high-dose cyclophosphamide mobilization in lymphoma, myeloma and solid tumors. *Bone Marrow Transplant* 1992; 9:11-7.
5. Goldschmidt H, Hegenbart U, Haas R, Hunstein W. Mobilization of peripheral blood progenitor cells with high-dose cyclophosphamide (4 or 7 g/m²) and granulocyte colony-stimulating factor in patients with multiple myeloma. *Bone Marrow Transplant* 1996; 17:691-7.
6. Alegre A, Tomas JF, Martinez-Chamorro C, Gil-Fernandez JJ, Fernandez-Villalta MJ, Arranz R, et al. Comparison of peripheral blood progenitor cell mobilization in patients with multiple myeloma: high-dose cyclophosphamide plus GM-CSF vs G-CSF alone. *Bone Marrow Transplant* 1997; 20:211-7.
7. Demirer T, Bukner CD, Gooley T, Appelbaum FR, Rowley S, Chauncey T, et al. Factors influencing collection of peripheral blood stem cells in patients with multiple myeloma. *Bone Marrow Transplant* 1996; 17:937-41.
8. Desikan KR, Barlogie B, Jagannath S, Vesole DH, Siegel D, Fassas A, et al. Comparable engraftment kinetics following peripheral-blood stem-cell infusion mobilized with granulocyte colony-stimulating factor with or without cyclophosphamide in multiple myeloma. *J Clin Oncol* 1998; 16:1547-53.
9. Corso A, Caberlon S, Pagnucco G, Klersy C, Zappasodi P, Alessandrino EP, et al. Blood stem cell collection in multiple myeloma: definition of a scoring system. *Bone Marrow Transplant* 2000; 26:283-6.
10. Demirer T, Bukner CD, Storer B, Lilleby K, Rowley S, Clift R, et al. The effect of different chemotherapy regimens on peripheral blood stem cell mobilization yield. *Proc ASCO* 1996; 15:1007[abstract].
11. Cook G, Marinaki P, Farrell E, Pearson C, Alcorn MJ, Sharp RA, et al. Peripheral blood progenitor cell mobilisation in patients with multiple myeloma following oral idarubicin and dexamethasone (Z-Dex) induction therapy. *Leukemia* 1997; 11, Suppl 5:35-40.
12. Stewart DA, Guo D, Morris D, Poon MC, Ruether BA, Jones AR, et al. Superior autologous blood stem cell mobilization from dose-intensive cyclophosphamide, etoposide, cisplatin plus G-CSF than from less intensive chemotherapy regimens. *Bone Marrow Transplant* 1999; 23:111-7.
13. Lazzarino M, Corso A, Barbarano L, Alessandrino EP, Cairoli R, Pinotti G, et al. DCEP (dexamethasone, cyclophosphamide, etoposide, and cisplatin) is an effective regimen for peripheral-blood stem cell collection in multiple myeloma. *Bone Marrow Transplant* 2001; 28:835-9.
14. Tribalto M, Amadori S, Cudillo L, Caravita T, Del Poeta G, Meloni G, et al. Autologous peripheral blood stem cell transplantation as first line treatment of multiple myeloma: an Italian Multicenter Study. *Haematologica* 2000; 85:52-8.
15. Facon T, Harousseau JL, Maloisel F, Attal M, Odriozola J, Alegre A, et al. Stem cell factor in combination with filgrastim after chemotherapy improves peripheral blood progenitor cell yield and reduces apheresis requirements in multiple myeloma patients: a randomized, controlled trial. *Blood* 1999; 94:1218-25.

PEER REVIEW OUTCOMES

Manuscript processing

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What is already known on this topic

The mobilization efficacy of intermediate dose cyclophosphamide.

What this study adds

The DCEP regimen represents an alternative regimen for mobilization of multiple myeloma patients.

Potential implications for clinical practice

An higher proportion of myeloma patients are mobilized with a lower toxicity.

Mario Boccadoro, Associate Editor (Turin, Italy)