A comparison of chemo-free strategy with G-CSF plus plerixafor on demand versus intermediate-dose cyclophosphamide and G-CSF as PBSC mobilization in newly diagnosed multiple myeloma patients: An Italian explorative cost Analysis

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ARTICLE INFO

Keywords:
chemo-free stem cell mobilization
multiple myeloma
plerixafor

ABSTRACT

Background: Upfront single or tandem ASCT still represents an integral part of treatment for patients with multiple myeloma. The combination of intermediate dose (ID) - cyclophosphamide plus G-CSF, has been considered the standard method as mobilization regimen. No prospective randomized clinical trials have compared efficacy and costs using ID - cyclophosphamide against a chemo-free mobilization strategy with G-CSF and plerixafor on demand.

Methods: A prospective single arm of 20 patients enrolled in three Italian Centers mobilized with G-CSF plus plerixafor on demand was compared with a retrospective historical control arm of 30 patients mobilized with ID - cyclophosphamide (4 g/sqm) and G-CSF. Costs of the prospective arm was compared with the ones of the retrospective control arm with the aim to collect ≥4 × 10⁶/kg CD34 + . The exploratory cost analysis was performed using microcosting specific inputs of G-CSF plus plerixafor on demand versus ID - cyclophosphamide + G-CSF considering pre-apheresis, peri-apheresis and post-apheresis session.

Results: Mobilization with ID - cyclophosphamide and G-CSF resulted in a significantly higher CD34+ peak mean on day 1 yield (119 CD34+ μL vs 67.3; p = 0.06) and in total average CD34+ yield (mean collection 10.6 × 10⁶/kg vs 5.8 × 10⁶/kg; p = 0.004) compared to patients mobilized with G-CSF and plerixafor. There was no significant differences (p = 0.36) in the two groups of patients collecting ≥ 4 million CD34 +/kg with ID - cyclophosphamide and G-CSF (93.3 %) vs G-CSF and plerixafor (90.9 %). None of the patients undergoing G-CSF and plerixafor mobilization had febrile neutropenia compared with 7 patients who received ID - cyclophosphamide and G-CSF (0% vs 23 %, p = 0.03) who had a median of 5 days hospitalization (range 4-6). All patients proceeded to ASCT with a mean of 3.6 CD34+/kg infused for G-CSF and plerixafor arm and 4.4 CD34+/kg for the ID - cyclophosphamide + GCSF group (p = 0.37) with a median time to ANC and PLT engraftment not different in the two groups. Total costs of a mobilizing strategy using a combination of G-CSF and plerixafor on demand was 12.690 euros compared to 16.088 euros with ID - cyclophosphamide and G-CSF (p = 0.37); in particular, mobilization cost components were significantly lower for G-CSF and plerixafor vs G-CSF and ID - cyclophosphamide for hospital stay (3080 euros vs 9653 euros; p < 0.001) whereas for mobilizing agent, there was a significative difference with 5470 euros for G-CSF and plerixafor use due to the cost of plerixafor compared with 1140 euros for ID - cyclophosphamide and G-CSF treatment (P = 0.001).

Conclusions: Our data demonstrate that in patients with multiple myeloma eligible for ASCT, a chemo-free
mobilization with G-CSF and plerixafor on demand was associated with efficacy in PBSC collection and optimal safety profile with similar average costs when compared to a chemo-mobilization with ID - cyclophosphamide. A prospective randomized multicenter study could address which is the most cost-effective strategy for this setting of patients.

Clinical Trial Registry: EudraCT 2013 – 004690-27.

1. Introduction

In spite of the introduction of novel multidrug induction agents that have dramatically changed the prognosis and overall survival [1–3], high-dose melphalan followed by single or tandem autologous stem cell transplantation (ASCT) still represents an integral part of treatment for patients with multiple myeloma in first remission [4].

Successful peripheral blood stem cell (PBSC) harvest remains a key factor for ASCT, with a minimum number of $2.0 \times 10^6$ CD34+/kg required for a single procedure. The International Myeloma Working Group has suggested a minimum target of $4 \times 10^6$ CD34+/kg and, if feasible, an average of $8–10 \times 10^6$ CD34+ should be collected, allowing most myeloma patients to undergo two autografts during the course of their disease [5].

The aim of the PBSC mobilization strategy is always to collect an adequate amount of CD34+ cells for transplantation, preferably in the first mobilization attempt and ideally with a minimum number of apheresis sessions to reduce the minimal hospitalization and side effects. In addition to the successful mobilization of PBSC, the costs should be considered and need to include not only the cost of the drugs but also the costs of the entire procedure [6–8].

The combination of intermediate dose - cyclophosphamide (3–4 g/sqm) plus granulocyte-colony stimulating factor (G-CSF), has been considered the standard mobilization regimen for multiple myeloma. It has high efficacy collection yield and low mobilization failure rates [9]. The use of cyclophosphamide used even at intermediate dosage might have high efficacy collection yield and low mobilization failure rates [9].

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The combination of intermediate dose - cyclophosphamide (3–4 g/sqm) plus granulocyte-colony stimulating factor (G-CSF), has been considered the standard mobilization regimen for multiple myeloma. It has high efficacy collection yield and low mobilization failure rates [9].

Steady-state mobilization with the use of G-CSF alone allows self-administration as outpatients with a good predictability of the CD34 peak in the peripheral blood. Adequate PBSC collections might be achieved with minimum side effects and hospitalization [9].

Plerixafor is a selective reversible inhibitor of the CXCR4 chemokine receptor. It blocks the binding of its ligand SDF-1, thus increasing the mobilization of CD34 in the peripheral blood. The use of plerixafor “on demand” is approved in poor mobilizer patients according the European Group for Blood and Marrow Transplantation position statement [11] and allows predictable time to peak CD34+ cells (around 11 h after administration), reducing mobilization failures as compared with G-CSF alone [12,13].

No prospective randomized clinical trials have compared efficacy and costs using a chemo-free mobilization strategy with G-CSF and plerixafor on demand against the use of intermediate dose of cyclophosphamide.

In this paper we report the results of a multicenter study in multiple myeloma patients (scheduled for a tandem ASCT) who prospectively received G-CSF plus plerixafor on demand as upfront mobilization regimen with the aim to compare the cost and the efficacy of this strategy with an historical group of patients who had received cyclophosphamide plus G-CSF as mobilization regimen.

2. Patients and methods

2.1. Patient population

A total of 50 adult patients affected by multiple myeloma who were candidates to receive single or tandem ASCT after a first-line induction and mobilization therapy were included in the study. A prospective single arm of 20 patients enrolled in three Italian Centers (IEO - Milano, INT Pascale - Napoli and CTMO – Reggio Calabria) mobilized with re-combinant human non glycosylated G-CSF (G-CSF) plus plerixafor on demand was compared with a retrospective historical control arm of 30 patients mobilized with intermediate-dose cyclophosphamide (4 g/sqm) and G-CSF at the IEO.

Costs of the prospective arm was compared with the ones of the retrospective control arm with the aim to collect $\geq 4 \times 10^6$/kg CD34+ . Characteristics of patients in two arms were comparable in terms of age, gender, isotype, status at diagnosis and disease status at mobilization, median lines of previous therapies and transplant strategy.

This study has been evaluated and approved by the Ethical Committee of the IEO and Centro Cardiologico Monzino of Milan.

2.2. PBSC mobilization and collection

Cyclophosphamide (4 g/sqm) and G-CSF (5 mg/kg/day started on day +2) was used as PBSC mobilization in the historical control series of 30 patients consecutively mobilized at IEO from November 2008 to November 2015. Cyclophosphamide was administered on an inpatient basis as an intravenous infusion by a central venous catheter (CVC) on day 1 followed by G-CSF 5 μg/kg/day from day 3 to ANC recovery. All patients received oral antimicrobial prophylaxis with levofloxacin, acyclovir and fluconazole. CVC was removed at the discharge; if adequate vascular accesses were not present, another CVC was placed before apheresis collection.

Peripheral blood CD34+ cell count was measured daily when patient’s white blood cell count recovered to $> 1500$ cells/μL or from day +12 onward. If CD34+ cells $> 20/μL$, the collection of PBSC was started.

G-CSF 10 μg/kg/day (non-pegylated G-CSF only) and plerixafor 240 mg/kg/day was used prospectively in 20 patients form May 2015 to August 2017.

Patients received subcutaneously G-CSF 10 μg/kg in the morning for 4 days as outpatients; on day 4, only if peripheral CD34+ count was $\leq 20 μL$, patients received the injection of plerixafor (0.24 mg/kg) as a subcutaneous injection as on demand strategy, by hospitalization regimen. Plerixafor were administered on an inpatient basis.

On day 5–7 patients had their morning dose of G-CSF and measurement of circulating CD34 cells was performed. If CD34+ cells were $> 20/μL$, the collection of PBSC was started. The same procedure was repeated until the target was reached.

Treatment with plerixafor on each evening prior to apheresis and G-CSF on each day of apheresis was continued daily until the target number of CD34+ stem cells as determined by the investigator was collected. Patients received up to a maximum of 3 apheresis.

All collections were performed using the MNC program of the COBE Spectra apheresis system (Caridian) by processing a maximum of two blood volumes.

Evaluation of CD34+ cells was performed using CD34PE/CD45FAC combination with Trucount Tubes (Becton Dickinson). CD34 stem cell enumeration was evaluated on samples of each individual leukapheresis product. Flow cytometric analysis was performed according to ISHAGE gating strategies.

If two or more leukapheresis were needed to achieve the required CD34+ cell number, the assay was repeated on each leukapheresis product prior to infusing into the recipient.
The final products were cryopreserved in 10 %DMSO using a controlled rate freezer and stored in liquid nitrogen. Each bag was divided in order to have a maximum of $350 \times 10^3$ ml WBC cells per cryopreserved bag.

2.3. Transplant Procedure and Supportive Care

All patients received a conditioning regimen with melphalan 200 mg/sqm on day -2 followed by infusion of autologous PBSC on day 0. All patients received G-CSF 5 μg/kg/ from day +5, fungal (fluconazole), herpes (acyclovir) and bacterial prophylaxis (levofloxacin) per institutional guidelines. The time of neutrophil engraftment was considered the first of 3 successive days with ANC > 0.5 cells/L. The time of platelets engraftment was considered the first of 3 successive days with platelets count > $20 \times 10^9$ cells/L in the absence of platelets transfusion for preceding 7 days.

2.4. Cost determination

The exploratory cost analysis was performed using microcosting specific inputs of G-CSF + plerixafor on demand (prospective arm) versus cyclophosphamide + G-CSF (retrospective control) as mobilization regimen. Cost were calculated per patients in both groups by a microcosting evaluation considering:

- **Pre-apheresis session**, that included hospitalization days for cyclophosphamide/plerixafor administration, CVC positioning and cost of mobilizing agents, febrile neutropenia and other Adverse Events (AEs) management cost: drugs / hospitalization, management of AEs, antbiotic treatment until first apheresis procedure.
- **Peri-apheresis session** that included apheresis procedure and CD34+ counts, blood counts, biochemical parameters (creatinin, Na+, K+, Ca++, AB0 group) AEs (number of procedures required, procedures to be performed on the week-end) for 1st apheresis procedure up to the day of last apheresis.
- **Post-apheresis session** that included storage cost, AEs (blood transfusions), The postapheresis period will be up to 15 days from apheresis from end of the day of last apheresis.

Costs were calculated according as presented in Table 1 and estimated from the reimbursement rates of IEO Inpatient fees and adjusted to reflect 2012 Euro values.

2.5. Telomerase evaluation

To evaluate the telomerase expression of PBSC mobilized with G-CSF and plerixafor on demand versus the retrospective historical control, we used a RQ-PCR assay based on TaqMan fluorescence methodology to quantify the full range of hTERT mRNA copy numbers. By plotting the Ct value of an unknown sample on the standard curve, the amount of target sequences in the sample could be calculated. We used different concentrations of ABL1 and hTERT standard templates including $10^2 \times 10^5 \times 10^6$ copies/reaction to perform quantitative PCR and calculate the standard curves, respectively. To normalize the hTERT mRNA expression for sample-to-sample differences in RNA input, RNA quality, and reverse transcriptase efficiency, we amplified the housekeeping gene ABL1. According to each standard curve, we got the copy numbers of ABL1 and hTERT, respectively. The ratio between copy numbers of hTERT and ABL1 represented the normalized hTERT (NhTERT) for each sample and could be compared with that of other samples. NhTERT = (hTERT mRNA copies sample/ABL1 mRNA copies sample) × 100.

2.6. Statistical Analysis

This was an exploratory study without formal sample size calculation, the overall goal was the comparison of costs of PBSC in multiple myeloma patients treated either with cyclophosphamide 4 g/sqm + G-CSF 5μg/Kg or G-CSF 10μg/Kg + plerixafor 0.24 mg/Kg on demand. Patients characteristics and treatments at the time of mobilization as well as mobilization costs, were described by counts and percent (categorical variables) or mean, median Standard Deviation (SD) and interquartile range (IQR) (continuous variables) and compared by Fisher’s exact test or t-test and two-sample Wilcoxon test as appropriate.

All tests were two-tailed and considered significant at the 5 % level. All analyses were done using SAS 9.4 (N.C., Cary).

3. Results

3.1. Patient characteristics

Table 2 shows the baseline characteristics of the 50 eligible patients included in this study. The two treatment groups did not differ significantly at the time of mobilization (baseline) except for the distribution of prior therapies (radiation, thalidomide based and bortezomib based).

There were 27 men and 23 women, median age was 56 (IQR: 51–63), the number of median lines of previous therapy was 1. Ten patients, all in the control arm, had also received radiotherapy.

Since G-CSF and plerixafor mobilization was used in a more recent time period (from May 2015), more patients in this group received bortezomib containing induction regimens before mobilization (100 % vs 33 %, p < .001); no patients in both arms received lenalidomide therapy before mobilization strategy.

3.2. PBSC mobilization and collection results

Mobilization with cyclophosphamide and G-CSF resulted in a nearly significantly higher CD34+ peak mean on day 1 yield (119 CD34+ μL vs 67.3; p = 0.06) compared to patients mobilized with G-CSF and
plerixafor (Table 3). The total average CD34+ yield was consequently significantly higher in patients mobilized with cyclophosphamide and G-CSF (mean collection 10.6 × 10^6/kg vs 5.8 × 10^6/kg; p = 0.004). As for the efficacy of collecting the minimum target required to perform a tandem transplant, there was no significant differences (p = 0.36) in the two groups of patients collecting ≥ 4 million CD34+/Kg with cyclophosphamide and G-CSF (93.3 %) vs G-CSF and plerixafor (90.0 %)

Eight (20 %) of patients in G-CSF and plerixafor on demand group was able to collect ≥ 4 million CD34+/Kg by a single apheresis session compared to 20 (66.7 %) of patients in the group of cyclophosphamide and G-GSF (p = 0.09)

There was a significant difference in patients collecting the optimal target (8 million CD34+/Kg for a tandem transplant) undergoing cyclophosphamide and G-CSF mobilization (60 % vs 15 %, p = 0.003) compared to G-CSF and plerixafor.

Seven patients (35 %) in the prospective arm was able to collect the minimum target required without the use of plerixafor on demand, having a median peak CD34+ of 27.8 (range 20–72.7). As for plerixafor administration on demand, 10 patients received a single administration and 3 patients two administration during mobilization phase to reach the cellular target.

The median number of apheresis session was 1 in cyclophosphamide and G-CSF arm and 2 in patients mobilized with the combination of G-CSF and plerixafor on demand (p = 0.05).

There were two mobilization failures in both groups (collecting a median of 3.6 × 10^6/kg CD34+ (range 2.96–3.80 × 10^6/kg).

As for the number of bags required to cryopreserve the PBSC collected, a median of two bags were frozen in the cyclophosphamide and G-CSF group vs three in the G-CSF and plerixafor arm (p < 0.001).

3.3. Toxicity, supportive care, transplant results

Grade 3−4 neutropenia occurred in all patients who received cyclophosphamide and G-CSF; 3% of them needed for red blood cell (RBC) and 7% platelet (PLT) transfusions.

None of the patients undergoing G-CSF and plerixafor mobilization had febrile neutropenia compared with 7 patients who received cyclophosphamide and G-CSF (0% vs 23 %, p = 0.03) who had a median of 5 days hospitalization (range 4–6). This implied no days of intravenous antibiotics in the prospective arm versus 5 days (range 4–5) for the historical control.

As for central venous access, 16 patients required CVC positioning for apheresis: 8 in the G-CSF and plerixafor arm and 8 in the cyclophosphamide + GCSF arm.

There were no mobilization – related severe adverse events in either group.

All patients proceeded to autologous transplant with a mean of 3.6 CD34+/kg infused for G-CSF and plerixafor arm and 4.4 CD34+/kg for the cyclophosphamide + G-CSF group (p = 0.37). Median time to ANC and PLT engraftment was not different in the two groups with a median of 11 days (range 10−13) and 12 days (range 11−16) for ANC and PLT engraftment in G-CSF and plerixafor compared to 12 days (range 11−14) and 12 days (range 10−17) for ANC and PLT engraftment in cyclophosphamide and G-CSF arm. No patient in both arm presented a long term engraftment failure.

3.4. Mobilization and apheresis costs

The distribution of costs observed in the study are summarized in Table 4 and Figs. 1–2.

Total costs of a mobilizing strategy using a combination of G-CSF and plerixafor on demand was 12.690 euros compared to 16.088 euros when cyclophosphamide and G-CSF was used (p = 0.07). Total costs distribution means were not biased by any outlier (Fig. 2).

Mobilization cost components were significantly lower for G-CSF and plerixafor vs G-CSF and cyclophosphamide for hospital stay (3080 euros vs 9653 euros, p < 0.001).

Patients undergoing cyclophosphamide and G-CSF mobilization strategy required CVC insertion either for chemotherapy administration or apheresis procedures, this reflecting in a higher cost comparing patients receiving G-CSF and plerixafor on demand (2720 euros vs 540 euros; P < 0.001).

As for mobilizing agent the cost analysis also revealed a significative difference with 5470 euros for G-CSF and plerixafor use due to the cost of plerixafor compared with 1140 euros for cyclophosphamide and G-CSF treatment (P = 0.001).

In terms of apheresis procedure costs we observed a cost of 2260

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**Table 2**

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>All patients</th>
<th>G-CSF + Plerixafor</th>
<th>G-CSF + Cyclophosphamide</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, median</td>
<td>56 (51–63)</td>
<td>61 (53–65)</td>
<td>55 (51–61)</td>
<td>0.11</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>27 (54.0)</td>
<td>8 (40.0)</td>
<td>19 (63.3)</td>
<td>0.15</td>
</tr>
<tr>
<td>Female</td>
<td>23 (46.0)</td>
<td>12 (60.0)</td>
<td>11 (36.7)</td>
<td></td>
</tr>
<tr>
<td>Intensity, N</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IgG</td>
<td>47 (94.0)</td>
<td>19 (95.0)</td>
<td>28 (93.3)</td>
<td>1.00</td>
</tr>
<tr>
<td>IgA</td>
<td>3 (6.0)</td>
<td>1 (5.0)</td>
<td>2 (6.7)</td>
<td>1.00</td>
</tr>
<tr>
<td>IgM</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Status at diagnosis</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>All</td>
<td>10 (20.0)</td>
<td>6 (30.0)</td>
<td>4 (13.3)</td>
<td>0.17</td>
</tr>
<tr>
<td>IIIA</td>
<td>35 (70.0)</td>
<td>12 (60.0)</td>
<td>23 (76.7)</td>
<td>0.23</td>
</tr>
<tr>
<td>CR</td>
<td>19 (38.0)</td>
<td>9 (45.0)</td>
<td>10 (33.3)</td>
<td>0.55</td>
</tr>
<tr>
<td>VGFR</td>
<td>5 (10.0)</td>
<td>3 (15.0)</td>
<td>2 (6.7)</td>
<td>0.38</td>
</tr>
<tr>
<td>PR</td>
<td>26 (52.0)</td>
<td>8 (40.0)</td>
<td>18 (60.0)</td>
<td>0.25</td>
</tr>
<tr>
<td>SD</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Prior therapies</td>
<td>10 (20.0)</td>
<td>0</td>
<td>10 (33.3)</td>
<td>0.003</td>
</tr>
<tr>
<td>Thalidomide based</td>
<td>20 (40.0)</td>
<td>0</td>
<td>20 (66.7)</td>
<td>0.01</td>
</tr>
<tr>
<td>Bortezomib based</td>
<td>30 (60.0)</td>
<td>20 (100)</td>
<td>10 (33.3)</td>
<td>0.001</td>
</tr>
</tbody>
</table>

---

**Table 3**

<table>
<thead>
<tr>
<th>Mobilization and Apheresis Results</th>
<th>All patients</th>
<th>G-CSF + Plerixafor</th>
<th>G-CSF + Cyclophosphamide</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>CD34 + Collected Total x 10^6/kg, Mean (SD)</td>
<td>8.7 (6.4)</td>
<td>5.8 (1.6)</td>
<td>10.6 (7.6)</td>
<td>0.004</td>
</tr>
<tr>
<td>CD34 + pre LAF, Mean (SD)</td>
<td>98.9 (79.9)</td>
<td>67.3 (45.5)</td>
<td>119 (90.7)</td>
<td>0.06</td>
</tr>
<tr>
<td>No. of pts collecting ≥ 4 × 10^6, N (%)</td>
<td>46 (92.0)</td>
<td>18 (90.0)</td>
<td>28 (93.3)</td>
<td>0.36</td>
</tr>
<tr>
<td>Single apheresis session with ≥ 4 × 10^6, N (%)</td>
<td>28 (56.0)</td>
<td>8 (20.0)</td>
<td>20 (66.7)</td>
<td>0.09</td>
</tr>
<tr>
<td>No. of pts collecting ≥ 8 × 10^6, N (%)</td>
<td>21 (42.0)</td>
<td>3 (15.0)</td>
<td>18 (60.0)</td>
<td>0.003</td>
</tr>
<tr>
<td>No. Apheresis, Median (min,max)</td>
<td>1 (1,3)</td>
<td>2 (1,3)</td>
<td>1 (1,3)</td>
<td>0.05</td>
</tr>
<tr>
<td>No. frozen PBSC bags, Median (min,max)</td>
<td>2 (1,8)</td>
<td>3 (1,8)</td>
<td>2 (2,3)</td>
<td>&lt; 0.001</td>
</tr>
</tbody>
</table>
eurosintheG-CSFandplerixaforprospectivearmcomparedto1748for
the retrospective one (p<0.001).

3.5. Telomerase evaluation
Our results (Table 5) showed that the expression of human telo-
merase are not significantly different in patient mobilized with G-CSF
and plerixafor versus cyclophosphamide and G-CSF (p = 0.18 two-
sample Wilcoxon test), and also not significantly different to the ex-
pression found in healthy subjects (0.27% vs 0.45%, p = 0.44; 0.63 %
vs 0.45 %, p = 0.33 respectively, one sample signed rank test).

Of note, only in two patients mobilized with G-CSF + cyclopho-
phamide we observe an increase of transcription values.

4. Discussion
In this study we have prospectively evaluated PBSC mobilization
following a chemo-free strategy (G-CSF plus plerixafor on demand)
against a chemo-mobilization with intermediate dose cyclopho-
phamide with the aim to compare the cost and the efficacy of these
different strategies for patient affected by multiple myeloma candidate
to ASCT as consolidation therapy.

Ideally, the best method of PBSC mobilization should obtain the
sufficient number of CD34+ cells necessary to ensure rapid and sus-
tained engraftment in the lowest possible number of apheresis sessions,
thus minimizing possible side effects for the patient. Evaluation of the
cost related to the stem cell mobilization strategy should also be con-
sidered as inclusive in the decision; several retrospective studies re-
cently addressed the question [6–8].

We chose to test a chemo-free approach in three different Italian
Centers and to compare the results with an historical group of patients
who received the “gold standard” treatment with an intermediate dose
of cyclophosphamide for several reasons: the contribution of che-
motherapy mobilization in disease control in multiple myeloma after
combination drugs recently used in induction treatment is not a matter
since several studies demonstrated similar outcomes irrespective of the
method of mobilization [14,15]. Also, a steady state mobilization al-
 lows for a predictable timing of cell collection and minimizes the re-
sources needed for the hospital and the discomfort for the patients.
Moreover, stem cell mobilization with G-CSF alone has become a more
feasible option because of the introduction of plerixafor, used on de-
mand in poor mobilizers patient [13].

First, our data suggest that the total costs of a mobilizing strategy
using a combination of G-CSF and plerixafor on demand compared to
cyclophosphamide is not statistically different (p = 0.07).
Second, mobilization with a chemo-free approach allows to collect
the minimum PBSC target required to perform a transplant with no
significant differences (p = 0.36) when cyclophosphamide and G-CSF
combination strategy was used.
Third, the telomerase expression did not demonstrate any difference
between the two different strategy used.

Our “microcosting” analysis highlighted a pre-apheresis mobiliza-
tion cost components significantly lower for G-CSF and plerixafor on
versus versus cyclophosphamide and G-CSF (3080 euros vs 9653
euros, p < 0.001). In fact, despite a significative difference related to
the drugs costs with 5470 euros for G-CSF and plerixafor use compared
with 1140 euros for cyclophosphamide and G-CSF treatment (P =
0.001), the hospital stay cost was significantly higher for patients mo-
bilized with chemotherapy. This difference was mainly related to the
fact that none of the patients undergoing G-CSF and plerixafor mobi-
lization had febrile neutropenia compared with 7 patients who received
cyclophosphamide and G-CSF (p = 0.03) with 5 days median hospi-
talization (range 4-6) to manage this complication. This data is in line
with what reported in other studies [7,8] and determines an increase in

### Table 4

<table>
<thead>
<tr>
<th>Cost components, Mean (min,max)</th>
<th>Plerixafor/ Plerixafor + G-CSF N = 20</th>
<th>EDX N = 30</th>
<th>Δ</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hospital stay</td>
<td>3080 (8,004,800)</td>
<td>9653 (160,024,600)</td>
<td>-6573</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>CVC</td>
<td>540 (01,200)</td>
<td>2720 (24,003,600)</td>
<td>-2180</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Mobilizing agents</td>
<td>5470 (56,012,640)</td>
<td>1140 (7,401,860)</td>
<td>4330</td>
<td>0.001</td>
</tr>
<tr>
<td>Apheresis procedure</td>
<td>2260 (12,903,910)</td>
<td>1748 (12,903,790)</td>
<td>512</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Storage</td>
<td>1340 (4,003,200)</td>
<td>827 (8,001,200)</td>
<td>513</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Total Costs</td>
<td>12,690 (585,023,900)</td>
<td>16,088 (891,033,050)</td>
<td>-3398</td>
<td>0.07</td>
</tr>
</tbody>
</table>

### Table 5

<table>
<thead>
<tr>
<th>Human Telomerase Expression, %</th>
<th>G-CSF + Plerixafor N = 6</th>
<th>G-CSF + Cyclophosphamide N = 7</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean (SD)</td>
<td>0.34 (0.29)</td>
<td>2.05 (3.20)</td>
<td>0.18 #</td>
</tr>
<tr>
<td>Median</td>
<td>0.27</td>
<td>0.63</td>
<td></td>
</tr>
</tbody>
</table>

1% = 100*(hTERT copies/hABL1 copies); Healthy donors mean = 0.45.
# Wilcoxon test.
the costs of hospitalization for patients treated with chemomobilization. Moreover, patients undergoing cyclophosphamide and G-CSF mobilization strategy required CVC insertion either for chemotherapy administration (100 %) or apheresis procedures (27 %) while only a percentage (40 %) of patients receiving G-CSF and plerixafor on demand required CVC insertion for apheresis courses only.

Regarding the apheresis cost analysis, we observed a cost of 2260 euros in the G-CSF and plerixafor prospective arm compared to 1748 euros for the retrospective one (p < 0.001) related to the median number of apheresis session in the two groups of patients: one in cyclophosphamide and G-CSF arm versus two in patients mobilized with the combination of G-CSF and plerixafor on demand (p = 0.05) reflecting the higher activity of cyclophosphamide in PBSC yield.

As for post-apheresis section, a median of two bags were frozen in the cyclophosphamide and G-CSF group versus three in the G-CSF and plerixafor arm with a different storage cost of 827 euros for chemomobilization and 1340 euros for a chemo-free strategy (p < 0.001). This difference is related to the fact that G-CSF and plerixafor combination induces a greater leukocytosis with 75% of the collections in our prospective arm divided into rates (2/4 bags) with an average of the cellular concentration of 330 × 10^6/kg [3] WBC / bag.

Taken together, our results demonstrated that the total cost of a chemo-free mobilization with G-CSF and plerixafor on demand results was comparable to a chemo-mobilization with intermediate – dose cyclophosphamide and G-CSF.

In terms of PBSC mobilization and harvest, cyclophosphamide and G-CSF resulted in a significantly higher CD34+ peak mean on day 1 yield (p = 0.06) and in total average of CD34+ collected compared to patients mobilized with G-CSF and plerixafor (p = 0.004) as reported in previous studies [9,10].

As for the efficacy of collecting the minimum target required to perform a tandem transplant, there was no significant differences (p = 0.36) in the two groups of patients collecting ≥ 4 million CD34+/Kg with cyclophosphamide and G-CSF (93.3 %) versus G-CSF and plerixafor (90.0 %); of note, seven patients (35 %) in the chemo-free arm was able to collect the minimum target required without the use of plerixafor on demand.

Patients included in this study underwent standard volume apheresis: none of patients processed more than 2 blood volume per apheresis section to avoid thrombocytopenia and electrolyte abnormalities. This choice, aimed to improve safety and patient tolerance of the procedure, should be considered in analyzing the mobilization and harvest results in chemo-free results since several studies with large-volume apheresis reported improved PBSC collection yields [16].

There was a significant difference in patients collecting the optimal target (8 × 10^6/kg CD34+) undergoing cyclophosphamide and G-CSF mobilization (60 % vs 15 %, p = 0.003) compared to G-CSF and plerixafor on demand. All patients proceeded to autologous transplant; despite the difference in mean of PBSC infused for G-CSF and plerixafor arm (3.6 × 10^6/kg CD34+) versus cyclophosphamide + G-CSF arm (4.4 × 10^6/kg CD34+), median time to neutrophil and platelet engraftment was not different in the two groups.

This observation seems to confirm our choice to identify 4 × 10^6/kg CD34+ as target to be collected in exploring costs of the two different mobilization strategy used in the study.

In terms of biological observations, our results showed that the expression of human telomerase in patients mobilized with G-CSF and plerixafor compared to cyclophosphamide and G-CSF are not significantly different, probably related to the presence of highly proliferating cells in leukapheresis products. It has been described that the CD34 stem cells do not present high levels of telomerasers [17].

In conclusion, our data demonstrate that in patients with multiple myeloma eligible for ABMT, a chemo-free mobilization is associated with efficacy in PBSC collection and optimal safety profile with similar average costs when compared to a chemo-mobilization with intermediate dose of cyclophosphamide. A prospective randomized multicenter study could address which is the most cost-effective strategy for this setting of patients.

Credit author statement

D. Laszlo for creation of the published work, leadership responsibility for the research activity

D. Laszlo, GP. Maracceci, M. Martino, B. Luccchetti, A. Magarò, A. Caima, C.Rabascio for preparation, creation of the published work (critical review and final revision)

D. Radice for application of statistical techniques to analyze study data performing cost analysis

S. Menna, MT. Lionetti for management activities to produce metadata, and maintain research data

F. Bertolini for final revision of the manuscript

Acknowledgements

This research was supported by Sanofi for the additional costs of the trial and in part by AIRC and Italian Ministry of Health.

References


lymphoma patients failing previous mobilization attempts: EU compassionate use data. Bone Marrow Transplant 2010;46:52. https://doi.org/10.1038/bmt.2010.54.


