Plerixafor use in autologous hematopoietic stem cell mobilization: Experience from a single center in Southern India

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Abstract:
BACKGROUND: Plerixafor is used for patients at risk of Stem cell mobilization failure based on clinical factors or low peripheral blood CD34 count. It is also added upfront to any mobilization irrespective of risk factor, but the cost-effectiveness of the approach is an issue. Data on plerixafor in different settings of autologous hematopoietic stem cell (HSC) collection from India are scant. We are hereby reporting the experience of failure/success of mobilization rate and few important significant variables (CD34+ dosage, failed collection) between plerixafor and granulocyte colony-stimulating factor alone groups among autologous hematopoietic stem cell transplantation (aHSCT) at our institute.

METHODS: This was a record-based single-center study on patients who underwent aHSCT from January 2013 to June 2019 at a tertiary care hospital. Descriptive statistics were used for baseline characteristics, transplant-related factors, and peritransplant outcomes. All statistical analyses were performed at the 5% significance level.

RESULTS: During the study duration, a total of 96 patients had undergone autologous hematopoietic stem cell collection (aHSCC), all by peripheral blood stem cell harvest, requiring 131 apheretic collections. Of the total 131 collections in 96 patients, plerixafor was used in 63 apheresis collections (48% of total pheresis) in 40 patients. Among the 40 patients who were administered plerixafor to augment the collection, 34 patients had upfront use of plerixafor. We did not observe any significant adverse event related to plerixafor use.

CONCLUSION: A rational utilization of plerixafor can facilitate the process and logistics of aHSCC outcome.

Keywords: Autologous hematopoietic stem cell transplantation, India, plerixafor

Introduction

Treatment of several malignancies and bone marrow (BM) failure syndromes have been revitalized by hematopoietic stem cell transplantation (HSCT) as a therapeutic approach. The administration of hematopoietic growth factors (GFs), specifically granulocyte colony-stimulating factor (G-CSF) alone or G-CSF in combination with chemotherapy, is a standard approach to mobilize HSCs. The minimum threshold for autologous transplantation is currently defined as 2 x 10^6 CD34+ cells/kg body weight. The cell dose required for transplantation is associated with rapid and sustained blood count recovery, which
in turn helps in reduced hospitalization, blood product usage, and infections.\textsuperscript{[1,4]} However, G-CSF-based mobilization regimens have a failure rate of 2%–20% among healthy donors and 10%–50% in autologous patients, respectively.\textsuperscript{[1]}

Poor HSC mobilization is defined in various ways – for example (1) the failure to achieve a minimum level of 5–20 CD34+ cells/\(\mu\)L in peripheral blood after completion of the mobilization regimen, (2) the inability to collect at least 1–2 \(\times\) 10\(^6\) CD34+ cells/kg during a single apheresis procedure, (3) failure to collect a total of 5 \(\times\) 10\(^6\) CD34+ cells/kg with all collections.\textsuperscript{[3]} Several factors predict potential difficulty in HSC mobilization. Some common factors are the advanced age of the patient, BM damage by an underlying malignant disease correlated with poor yield. Other factors such as dose-intensive chemotherapy in multiple cycles by forcing HSC cycling lead to the exhaustion of HSC self-renewal and reconstitution potential and damages BM macrophage effector cells.\textsuperscript{[14]} Thus, the most common cause of mobilization failure in autologous donors is prior exposure to myelotoxic chemotherapy.\textsuperscript{[7]} DNA cross-linking agents such as melphalan, carmustine, and purine analogs such as fludarabine damage stem cells and their marrow niches. Furthermore, lenalidomide is also associated with an increased risk of mobilization failure, especially after receiving four or more cycles.\textsuperscript{[8,9]} Therefore, the use of stem cell-toxic chemotherapies should be avoided, if autologous HSCT (aHSCT) is planned. Previous extensive radiotherapy to BM sites is also a factor for poor mobilization.\textsuperscript{[4]} Persistent low platelet counts before mobilization have also been an independent risk factor for poor mobilization and related to low PB CD34+ harvest.\textsuperscript{[4]}

Plerixafor (AMD3100), a small molecule that inhibits stromal cell-derived factor-1\(\alpha\) (SDF-1\(\alpha\)) binding to the C-X-C chemokine receptor type 4 (CXCR-4) receptor, is approved for patients who show inadequate mobilization of CD34+ peripheral blood stem cells (PBSCs).\textsuperscript{[7]} It acts by reducing the binding and chemotaxis of HSCs to the BM stroma. It is generally used at a dose of 240 \(\mu\)g/kg/day subcutaneously about 12 h before the scheduled apheresis, as it generates peak CD34+ cells level by 6–9 h after administration.\textsuperscript{[13]} However, most patients generally mobilize with traditional approaches, namely steady-state GCSF mobilization or chemoembolization, and considering the cost, plerixafor-based mobilization had been reserved as a salvage strategy for failed collection. More recently, preemptive use of plerixafor is being practiced at many centers for patients at risk of mobilization failure based on clinical factors or a low peripheral blood CD34 count (<10 or 20/\(\mu\)L) on the day before pheresis. In addition, plerixafor can be added upfront to any mobilization irrespective of risk factor, but the cost-effectiveness of this approach is an issue. These settings and strategies for plerixafor use (salvage, preemptive, or upfront) have been discussed in some recent reviews, and different transplant centers may be following one or the other strategies consistently or tailored to a given patient.\textsuperscript{[10]} However, data on the use of plerixafor in different settings of autologous and/or allogeneic transplants from India are very scant.\textsuperscript{[9,11–14]} Therefore, we are hereby reporting the experience of failure/success of mobilization rate and few important significant variables (CD34+ dosage, failed collection) between plerixafor and G-CSF alone groups among aHSCT at our institute.

**Methodology**

Medical records of patients who underwent autologous hematopoietic stem cell collection (aHSCC) and subsequently aHSCT from January 2013 to June 2019 in the department of Medical Oncology and Transfusion Medicine at a tertiary care university hospital in South India were reviewed for enrollment into the study. Baseline patient and disease characteristics, transplant indication, mobilization and harvest details, engraftment time, and other peri-transplant outcomes were collected from medical records and analyzed.

**Baseline characteristics and transplant indication**

Autologous transplants mainly were done for malignant disorders at our center. Common indications include multiple myeloma (MM) as consolidation therapy or for progressive disease, refractory or relapsed lymphomas, and high-risk pediatric solid tumors (neuroblastoma). In addition, characteristics of the underlying disease, including BM involvement and details of treatment with radiotherapy and chemotherapy, including the number of lines of therapy, regimen, and cycles, were collected. Chemotherapeutic agents known to cause a severe decline in stem cell function or loss of stemness are referred to as stem cell toxic drugs, usually implicated ones are melphalan, carmustine, and dacarbazine platinum analogs, fludarabine, lenalidomide.\textsuperscript{[12,14]}

**Peripheral blood stem cell mobilization**

Before 2015, most patients were mobilized with GFs alone (5 \(\mu\)g/kg twice a day for 4–5 days). From October 2015, with the availability of generic plerixafor and our center’s empanelment under the state health insurance scheme for monetary support for transplant procedures, it became economically feasible to use plerixafor. From here on, plerixafor use was more frequent, though, primarily, it was used upfront based on clinical risk factors and at the physician’s discretion. Furthermore, plerixafor was used as salvage after a failed first collection. Because of the nonavailability of in-house...
facility for CD34 until the later part of 2019, preemptive use of plerixafor based on PB CD34 before the day of planned pheresis was not practiced. A periodic appraisal through auditing the data was intended to rationalize the use of plerixafor in the future.

**Peripheral blood stem cell pheresis and stem cell storage**

Most of our procedures were done on COBE spectra apheresis system. First, the stem cell harvest product was analyzed for total leukocyte counts, mononuclear cell count (MNC), and total CD34+ cells at the end of the entire collection. The following day, a second procedure was planned if the CD34 cells collected were <1–2 × 10⁶ CD34+ cells/kg during a single apheresis procedure. For patients with adequate CD34 in the harvest, if the possibility of stem cell infusion was within 72 h from the time of collection (generally for myeloma transplants with high-dose melphalan conditioning), then they were stored in a refrigerator, maintaining the temperature between 2°C and 8°C with no further processing. If not, the stem cell products were volume reduced for plasma removal by refrigerated centrifugation followed by cryopreservation in dimethyl sulfoxide (at a concentration of 10% in final product v/v) within 6–8 h of collection. Subsequently, they were rapidly frozen by dump freezing technique at −80°C and stored till the day of infusion.

**Peri-transplant outcomes**

After stem cell infusion (day 0), patients were monitored for regimen-related toxicities, febrile neutropenia, and other complications that were managed with supportive care as indicated. In addition, details were collected from the medical records regarding the day of neutrophil and platelet engraftment (defined as absolute neutrophil count >0.5 × 10⁹/L in the first of 3 consecutive days and platelet count >20 × 10⁹/L in the first of 3 consecutive days without transfusion support, respectively), duration of hospitalization (defined from day 0 to the day of discharge from bone marrow transplant [BMT] unit), blood product use, and transplant-related mortality (in first 30 days, from any cause).

Engraftment syndrome was defined as the occurrence of noninfectious fever, skin rash, diarrhea, hepatic and renal dysfunction, encephalopathy although transient, and capillary leak features, such as noncardiogenic pulmonary infiltrates, hypoxia, along with weight gain in the absence of no alternative etiologic basis other than engraftment. [13]

Primary engraftment failure was defined as no evidence of engraftment or hematological recovery of autologous cells within the 1st month after transplant, with no evidence of disease relapse. [14]

**Statistical analysis**

The data were tabulated in a Microsoft Excel sheet and analyzed using SPSS for Windows version 20 (SPSS IBM Corp. Ltd. Armonk, NY). Descriptive statistics were used for baseline characteristics, transplant-related factors, and peri-transplant outcomes. Differences in proportions were assessed using the Chi-square test or Fisher's exact test. Differences in means or median were tested using Student’s t-test or Mann–Whitney-U test as appropriate. All statistical analyses were performed at the 5% significance level.

**Results**

During the study duration, a total of 96 patients had undergone autologous HSCC collection (aHSCC), all by PBSC harvest, requiring 131 collections by apheresis [Figure 1]. Ninety-one aHSCCs were performed from January 2013 to June 2019, of which 40 transplants were done between 2013 and 2015, and 51 transplants were done from 2016 to June 2019. As stated earlier, plerixafor became available for use from October 2015 onwards.

**Baseline characteristics**

The total number of patients who underwent aHSCC during the study period was 96. The demographics and baseline characteristics of patients who underwent aHSCC, with/without plerixafor use, are given in Table 1.

**Mobilization and pheresis details (total n = number of pheresis done – “131”)**

Of the total 131 collections in 96 patients, plerixafor was used in 63 apheresis collections (48% of total pheresis) in 40 patients (42% of total patients). Among the 40 patients who were administered plerixafor to augment the collection, 34 patients had upfront use of plerixafor as per the physician’s discretion based on various factors in the patient’s baseline profile. These are summarized in Table 2.

The features of stem cell mobilization, number of phereses, and CD34 count in the harvested product among patients with the usage of plerixafor and only G-CSF mobilization group are shown in Table 3. The median CD34 count in the pheresis done with plerixafor use was 3.95 × 10⁶/kg (0.05–13.4). A median count of CD34 cells of 3.55 × 10⁶/kg (0.15–8.8) in pheresis was done without plerixafor.

Among the 34 patients in the upfront plerixafor usage group, 30 patients underwent the transplant. The reason for abandoning the BMT included inadequate collection, i.e., <1 × 10⁶ CD34+ cells/kg in 4 and, in addition, disease progression in one patient. On the other hand, 55 patients have undergone BMT in the G-CSF only mobilization
group. Thirteen patients had a poor collection; out of them, 12 underwent transplants based on MNC count of the harvest and the physician’s discretion with successful neutrophil and platelet engraftment. Thus, the mobilization failure rate was higher in the G-CSF group (23%) compared to the plerixafor group (10%) and was statistically significant \((P < 0.000)\) as well.

We used plerixafor as a secondary adjunct for six patients after the failed first collection with a CD34+ cell of \(1.4 \pm 0.5 \times 10^6/\text{kg}\). The patient characteristic is given in Table 4. Postplerixafor usage for the second harvest, all the patient’s had an adequate collection, i.e., CD34+ cell count – \(4.8 \pm 2 \times 10^6/\text{kg}\) and all the patients went ahead with the planned transplant.
We did not observe any significant adverse event related to plerixafor use. Myalgia and bone pain were attributed to concurrent GCSF use, although the exact proportion of patients experiencing these side effects and the severity was not available in the retrospective records.

**Peri-transplant outcomes (n = total number of patients = 91)**

Among the 40 patients in the plerixafor usage group, 36 patients and 55 out of 56 patients among the G-CSF had undergone a transplant, respectively. Day 30 transplant outcome for these 91 patients is given in Table 5. There was no significant difference in the time to neutrophil or platelet engraftment, the occurrence of engraftment syndrome, blood product use, or duration of hospital stay between the two groups. More patients had failed engraftment, and day 30 TRM was higher in the GCSF alone group, although statistically not significant. The most common cause of death for the patients with day 30 TRM was infection and sepsis.

**Discussion**

aHSCT provides a curative treatment option for many high risks and refractory/relapsed hematological malignancies. The collection of HPCs for both autologous and allogeneic HSCT has almost completely shifted to PBSC harvest over the past three decades. Our study demonstrates the safety and efficacy of plerixafor in HSC mobilization and adequate PBSC collection when used upfront for patients with clinical risk factors or when used as salvage for patients who fail GSCF mobilization.

Plerixafor reversibly inhibits the binding of SDF-1α to the CXCR4 in the stromal cells of the marrow. This results in the release of CD34+ cells into the circulation.[17,18] At present, it is recommended for use in the mobilization of HPCs (in combination with filgrastim) for collection and transplantation in patients with non-Hodgkin lymphoma and MM.[17] However, conventional mobilization regimens using G-CSF or chemo-mobilization can have a 10%–50% failure rate in patients planned for autologous transplant.[1,6,19,20] In our study, the failure rate with stable GCSF mobilization was 23%, comparable to the literature.

Plerixafor can be used in up-front, preemptive, immediate salvage, and remobilization settings, with protocols for appropriately selected patients. Plerixafor usage in the setting of the failed first collection after conventional mobilization has a success rate of about 90% in immediate salvage.[11,21] In our small subset of 6 patients where plerixafor was used as salvage, all patients could achieve optimal collection to undergo transplant. The most common and cost-effective setting of plerixafor use is preemptive based on the peripheral blood CD34 on day 4 or 5 of GCSF mobilization with about 75%–95% of patients achieving optimal collection with generally a single or two phereses.[10,22-24] As in house CD34 enumeration was unavailable during the study period, preemptive plerixafor use was not done in our study. With upfront plerixafor, irrespective of clinical factors or PB CD34 count, the optimal collection is achieved in 77% of patients though sometimes at a higher total cost.[14,25] In our study, plerixafor was primarily used in the upfront setting based on clinical risk predictors of poor mobilization, or sometimes for a logistic reason to avoid the second pheresis, and about 88% of patients had a successful collection comparable to the other

**Table 2: Reasons for upfront use of plerixafor (n=34)**

| Reasons                                               | n (%) |
|-------------------------------------------------------|-------|
| Stem cell toxic chemotherapy prior + >2               | 31 (91)|
| chemotherapy regimen + radiotherapy                   |       |
| Single regimen chemotherapy + radiotherapy            | 14 (41)|
| Age >60 years                                         | 2 (3) |
| Miscellaneous                                         | 2 (3) |
| *There may be multiple reasons for a given patient.   |       |

**Table 3: Pheresis and harvest details of peripheral blood stem cell collections with and without plerixafor use**

| Features                          | Plerixafor mobilization | G-CSF only mobilization |
|-----------------------------------|-------------------------|-------------------------|
|                                  | 63 (total number of apheresis for 40 patients) | 68 (total number of apheresis for 56 patients) |
| Precollection WBC                 | 38,498±16,523/cmm       | 35,322±13,921/cmm       |
| CD34 + collection (x10^9/kg), mean±SD | 5.265±2.6              | 3.266±2.6               |
| Order of apheresis procedure      |                         |                         |
| First, n (%)                      | 19 (47.5)               | 45 (80)                 |
| Second                            | 19 (38) (47.5%)         | 10 (20) (18%)           |
| Third                             | 2 (6) (5%)              | 1 (3) (2%)              |
| CD34 + collection (x10^9/kg) of first pheresis | 3.95 (0.05-13.4)       | 3.6 (0.15-8.8)         |

**“131” number of pheresis was done in “96” number of patients**

| Median number of pheresis         | 1 (1-3)                 | 1 (1-3)                 |
| Failed collection (with CD34 <1x10^9/kg with one or more pheresis), n (%) | 2 (5)                   | 13 (19)                 |
| Number of patients who underwent transplant (aHSCT), n (%) | 36 (90)                 | 55 (98)                 |

*aHSCT=Autologous hematopoietic stem cell collection, SD=Standard deviation, G-CSF=Granulocyte colony-stimulating factor, WBC=White Blood cells*
studies reporting on upfront plerixafor use.\textsuperscript{[1,12,23,26]} In our study, about 52% of the patients in the plerixafor group (both upfront and salvage) needed a second or a third pheresis. In the GCSF group, 20% required more than one apheresis.

The literature data shows a two to three-fold higher CD34 collection in the plerixafor group compared to the GCSF alone group.\textsuperscript{[1,14,19,23,27,28]} Most of the studies on plerixafor use report that a significantly greater number of patients eventually undergo transplant after plerixafor use compared to stable GCSF mobilization alone (RR = 2.59, 95% confidence interval: 1.40–4.81; \( P < 0.0001 \)), respectively.\textsuperscript{[17,19,29–31]} We did not observe any significant difference in the number of patients finally undergoing transplant in the plerixafor group vs. GCSF only group (90% vs. 98%, respectively). 23% of patients failed to collect the optimal dose of CD34 in the GCSF group. Most of the patients in the GCSF group eventually underwent transplants based on the MNC and the treating physician’s discretion. There is a period bias here as most of the GCSF alone mobilization was during the initial transplant unit setup. As the center’s experience grew with the staff working there, the confidence in the counts and related outcomes could have played a role in findings. In patients failing plerixafor-based mobilization, alternative salvage measures can include marrow harvest, chemotherapeutic agents such as cyclophosphamide or addition of GM-CSF.\textsuperscript{[1,2,6,27]}

Peritransplant outcomes of time to neutrophil and platelet engraftment, the incidence of engraftment syndrome, average blood product use, hospitalization days were similar between the two groups in our study. Our results are comparable to other studies reporting similar time to engraftment in the plerixafor group vis-à-vis G-CSF alone group.\textsuperscript{[6,7,19,20]} Although statistically not significant, engraftment failure (5.4% vs. 2.7%) and day 30 TRM (11% vs. 5.5%) was higher in the GCSF group than the plerixafor group and comparatively higher than that reported for autologous transplant in the literature, perhaps reflecting the learning curve of our transplant unit. Higher CD34 and, if collected more than the optimal dose with the help of plerixafor, can help rescue some cases at a very high risk of engraftment failure.

Although one of the few from India remarking on plerixafor use in mobilization for autologous transplant and its comparison with stable GSCF mobilization for clinical outcomes, our study had limitations of being a

| Patient profile who failed the first collection (\( n=5 \)) |
|----------------------------------------------------------|
| 30/female, Hodgkin’s lymphoma, stem cell toxic chemotherapy prior + >2 chemotherapy regimen + radiotherapy + bone marrow involvement |
| 43/female, Hodgkin’s lymphoma, stem cell toxic chemotherapy prior + >2 chemotherapy regimen + radiotherapy + bone marrow involvement |
| 65/male, multiple myeloma, single regimen chemotherapy |
| 59/female, multiple myeloma, single regimen chemotherapy |
| 37/male, Hodgkin’s lymphoma, stem cell toxic chemotherapy prior + >2 chemotherapy regimen + radiotherapy |
| 45/male, Hodgkin’s lymphoma, stem cell toxic chemotherapy prior + >2 chemotherapy regimen + radiotherapy |

**Table 5: Transplant outcomes**

| Features                                                                 | With plerixafor use | With only G-CSF (\( n=55 \)) |
|-------------------------------------------------------------------------|---------------------|-------------------------------|
|                                                                         | Upfront (\( n=30 \)) | Secondary/salvage (\( n=6 \)) | Overall (\( n=36 \))           |
| Diagnosis                                                               |                     |                               |                               |
| MM                                                                     | 17                  | 2                             | 19                            | 21                            |
| HL                                                                      | 6                   | 4                             | 10                            | 15                            |
| NHL                                                                    | 7                   | -                             | 7                             | 15                            |
| GCT/AML                                                                | -                   | -                             | 2 (AML)                       |
| Neuroblastoma                                                          | -                   | -                             | 2                             |
| Neutrophil engraftment (median days)                                    | 10 (9-14)           | 10.5 (9-17)                   | 10 (9-17)                     | 10 (9-23)                     |
| Platelet engraftment (median days)                                      | 12 (8-20)           | 12.5 (11-13)                  | 12 (8-20)                     | 13 (8-36)                     |
| Engraftment syndrome                                                    | 4                   | 1                             | 5                             | 5                             |
| Failed engraftment                                                     | 2                   | Nil                           | 1                             | 3                             |
| Blood product use                                                       |                     |                               |                               |                               |
| PRBC                                                                   | -                   | -                             | 2 (2-4)                       | 2 (1-11)                      |
| Platelets (SDP/equivalent)                                             | -                   | -                             | 4 (2-6)                       | 4 (1-31)                      |
| Median days of BMT hospitalization                                      | -                   | 21 (12-45)                    | 22 (13-52)                    |
| Day 30 TRM                                                             | -                   | -                             | 2                             | 6                             |
| Cause of day 30 TRM                                                    | -                   | -                             | 5 (sepsis)                    |
| Progressive disease                                                    |                     |                               |                               |                               |

\( ^{a} \text{HSCST}=\text{Autologous hematopoietic stem cell transplantation}, \text{AML}=\text{Acute myeloid leukemia}, \text{GCT}=\text{Germ cell tumor}, \text{MM}=\text{Multiple myeloma, HL}=\text{Hodgkin’s lymphoma, NHL}=\text{Non-HL, G-CSF}=\text{Granulocyte colony-stimulating factor, PRBC}=\text{Packed Red Blood cells, SDP}=\text{Single Donor platelets, BMT}=\text{Bone marrow transplant, TRM}=\text{Transplant related mortality} \)
retrospective study, including missing data in certain areas, small sample size, and period bias.[9,11,12,14,23,32] Our study also does not report preemptive plerixafor use, the most common strategy followed in most transplant centers. Nevertheless, based on our results of upfront plerixafor use, we suggest that clinical risk predictors should also be considered besides PB CD34 in practicing preemptive plerixafor. Kumar et al. from India evaluated the cost-effectiveness of preemptive single-dose plerixafor use in myeloma transplant, overall cost-benefit favored plerixafor use.[9] Although preemptive is the most commonly followed strategy, upfront plerixafor use based primarily on clinical factors and physicians’ discretion is increasingly used by many centers to save time and resources. A prospectively conducted cost-effective analysis for upfront plerixafor use can define its role in this setting more clearly.

Conclusion

Plerixafor use in the mobilization of HSCs is guided by several factors, and a rationale utilization with proper patient selection can facilitate the overall process and logistics of transplant for good clinical outcomes.

Financial support and sponsorship
Nil.

Conflicts of interest
There are no conflicts of interest.

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