

Peripheral Blood Hematopoietic Stem-cell Mobilization for Autologous Transplantation

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Abstract

Autologous hematopoietic stem-cell transplantation is important in the treatment of several lymphohematopoietic malignancies. Procurement of adequate stem cells, for which CD34 positivity serves as a useful surrogate, is necessary prior to the procedure. Several methods of mobilization are widely used, but methods for individual patients should depend on their specific clinical situation. More than 20% of patients do not achieve adequate mobilization and those who require numerous aphereses to accomplish adequate CD34+ cell yield may experience delayed hematopoietic recovery.

Keywords

Hematopoietic stem-cell mobilization, autologous transplantation

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Autologous hematopoietic stem-cell transplantation is effective in the treatment of many lymphohematopoietic malignancies, including Hodgkin and non-Hodgkin lymphoma and multiple myeloma. The procedure requires the prior collection of an adequate number of hematopoietic stem cells capable of rapidly producing mature hematologic cells. Acquiring such cells can be particularly challenging in patients who have been treated extensively with therapies toxic to stem cells, particularly in individuals of older age.

This article describes current approaches to stem-cell mobilization and focuses on the identification and treatment of those individuals who present with difficulties.

Theory

Animal studies demonstrate that hematopoietic stem cells, and not differentiated progenitors, are responsible for early hematopoietic reconstitution following autologous transplantation.¹ Collection of an adequate number of these cells, responsible for hematopoietic reconstitution, is a prerequisite for safe autotransplantation. Since there is no simple, rapid method for identification of these vital cells, quantification of CD34+ cells in the collected product has been used as a generally reliable surrogate. The number of CD34+ cells infused into the patient predicts the speed of hematologic recovery following transplantation.^{2–4} In general, infusion of $\geq 2 \times 10^6$ CD34+ cells/kg reliably results in rapid hematopoietic recovery following myeloablative preparative therapy. It is important to note, however, that only an exceedingly small proportion of those CD34+ cells contribute

meaningfully to hematopoietic recovery and that CD34+ dose is only a surrogate for these cells.

Principles of Hematopoietic Stem-cell Mobilization

Although unstimulated marrow or blood may serve as a source of stem cells for autologous transplantation, the use of 'mobilized' peripheral blood is a better source of cells and is now uniformly utilized for this purpose. The number of CD34+ cells in the blood is dramatically increased by mobilizing them from the marrow with hematopoietic growth factors.^{5,6} Growth factors, particularly granulocyte-colony-stimulating factor (G-CSF), alone or in combination with certain non-stem-cell-toxic chemotherapeutic agents (most commonly cyclophosphamide or etoposide), are the most frequently used mobilizing agents. The number of CD34+ cells in the bloodstream is increased several-fold by these measures, and CD34+ cells are efficiently collected by apheresis of approximately four total patient blood volumes or of about 20 liters. The number of CD34+ cells/kg in the collection is easily tabulated and the need for additional apheresis is determined. In order to ensure reliably rapid hematopoietic recovery, most centers target a dose of $4\text{--}5 \times 10^6$ CD34+ cells/kg, but doses $\geq 2 \times 10^6$ are generally considered adequate for safe transplantation.⁷ Collection of adequate numbers of CD34+ cells within one or two aphereses avoids occupying valuable machine and personnel time and considerable expense and inconvenience, particularly compared with patients who require more than five procedures. In addition, patients who are poor mobilizers, particularly those who require additional mobilization procedures, have poor survival compared with those who

Table 1: Factors That Predict Poor Stem Cell Mobilization

Older age
Extensive previous chemotherapy (months of treatment, number of cycles)
Use of stem cell toxic chemotherapy (e.g. melphalan, bischloroethylnitrosourea, fludarabine, lenalidomide)
Prior radiation therapy (to marrow)
Female sex
High body mass index
Marrow disease
Low platelet count before mobilization

mobilize adequate numbers of cells.⁸ Furthermore, patients who are supermobilizers, i.e. those who collect $\geq 8 \times 10^6$ CD34+ cells/kg, have improved survival.⁹ Whether these outcomes are directly related to the CD34+ dose or, as is more widely accepted, that good mobilization is a surrogate for healthier and younger patients with various good-risk factors, is uncertain.

Mobilization Methods

The two main methods of mobilization of stem cells utilize hematopoietic growth factors with or without chemotherapy. G-CSF alone has been the most common method at many centers. The precise mechanism by which G-CSF releases CD34+ cells into the bloodstream from their marrow niche is uncertain. G-CSF causes proliferation of neutrophils and release of proteases that degrade proteins anchoring stem cells to the marrow stroma. Protease-independent mechanisms also appear to contribute to the release of stem cells into the circulation.¹⁰ More than 20% of patients treated with G-CSF fail to mobilize CD34+ cells adequately for collection of $\geq 2 \times 10^6$ CD34+ cells within five days. A recent study of patients with non-Hodgkin lymphoma demonstrated that only 20% of patients were able to collect a target dose of $\geq 5 \times 10^6$ CD34+ cells/kg within four apheresis days. Thus, efficient collection of the targeted numbers of CD34+ cells needed for rapid and reliable hematologic recovery occurs in a minority of patients.

Recent evidence indicates the CXCR4 chemokine receptor is expressed on CD34+ cells and mediates adhesion of CD34+ stem cells to the marrow stroma via interactions with the chemokine stromal-derived factor-1 α (SDF-1 α). AMD3100 (plerixafor) is a novel small molecule that reversibly inhibits binding of SDF-1 α to CXCR4 and mobilizes CD34+ cells from the marrow into the circulation. The combination of plerixafor and G-CSF is well tolerated and improves mobilization of CD34+ cells significantly compared with G-CSF alone. A multicenter, randomized, double-blind, placebo-controlled trial in non-Hodgkin lymphoma demonstrated that the addition of plerixafor improved the percentage of patients procuring $\geq 5 \times 10^6$ CD34+ cells within four apheresis days from 20 to 59%.¹¹ A similar study in myeloma improved procurement of 6×10^6 CD34+ cells within two aphereses from 34.4 to 71.6%.¹² The enhanced mobilization resulted in rapid and durable hematologic recovery after transplantation.

It has been well documented that patients who fail to mobilize adequate numbers of hematopoietic stem cells have poor prognoses, generally undergo additional mobilization attempts with high failure rates, have compromised quality of life, and accumulate considerable expenses.^{8,13,14}

The expense of plerixafor as an additional mobilizing agent is balanced by the need for fewer aphereses and the higher proportion of patients obtaining adequate CD34+ cell numbers and therefore not requiring additional mobilization attempts. In addition, risks related to aphereses and patient convenience argue in favor of this more effective mobilization method.

Chemotherapeutic agents have been extensively utilized in combination with G-CSF to improve stem cell mobilization. Mobilization has been integrated into salvage chemotherapy regimens, which effectively mobilize CD34+ cells, e.g. ifosfamide, carboplatin, and etoposide (ICE) for relapsed Hodgkin and non-Hodgkin lymphoma.¹⁵ In a patient in whom autologous transplantation is planned and who is receiving planned courses of ICE or other salvage regimens, the final chemotherapy course followed by G-CSF can be an efficient method of mobilization, serving as needed treatment as well. In this circumstance, mobilization can be accomplished without disrupting planned chemotherapeutic treatment designed to minimize the extent of malignancy prior to transplantation and without adding a separate additional mobilization regimen.

Alternatively, high-dose cyclophosphamide¹⁶⁻¹⁸ or etoposide (in Hodgkin or non-Hodgkin lymphoma³) may, in combination with G-CSF, serve as effective treatment of malignancy as well as a mobilizing regimen. Mobilization is generally more effective than with G-CSF alone, yielding significantly more CD34+ cells compared with G-CSF alone.^{16,18,19} Administration of chemotherapy-based mobilization regimens results in the development of neutropenic fever in 20-30% of patients,³ an important disadvantage of this approach. It is important to note, however, that these drugs are effective treatments with similar response rates to standard salvage regimens, so if they are intelligently integrated into treatment regimens and serve as alternatives to other treatment, they may be an efficient method of mobilization. Etoposide has rates of disease response similar to alternative regimens for Hodgkin and non-Hodgkin lymphoma,³ but its integration into the treatment plan requires foresight.

The inability to precisely predict the day that apheresis should be initiated is another disadvantage of chemotherapy-based mobilization regimens. Most centers utilize a threshold for peripheral blood CD34+ cells to determine whether and when apheresis should be initiated. The pre-apheresis CD34+ cell count in peripheral blood correlates closely with the number of CD34+ cells collected.²⁰⁻²² Different centers employ various thresholds for the minimum CD34+ cell count to begin collection. Enumeration methods vary and other factors including patient size, blood volume processed, and collection efficiency influence the number of CD34+ cells collected. In some circumstances, peripheral blood CD34+ cell quantification may exclude patients who otherwise might collect adequate cells for transplantation. Instead of relying on peripheral blood CD34+ cell counts, apheresis may be initiated when the white blood cell (WBC) count reaches 5,000/ μ l. A recent study from our institution showed that the first day's harvest is highly predictive of patients at high risk for achieving an inadequate yield of CD34+ cells within five apheresis days (Hien Duong, MD, personal communication). Additional treatment, e.g. with AMD3100, might then be initiated only in those patients at risk for inadequate CD34+ cell harvests in order to

improve yield. This and other risk-adapted approaches are under investigation at many centers.

The authors have been particularly interested in patients who are poor mobilizers. Factors that predict poor mobilization are listed in *Table 1* and include older age, extensive prior chemotherapy, and extensive marrow involvement with malignancy.^{13,23-25} Patients who fail to mobilize adequate numbers of CD34+ cells on their initial attempt are at high risk for failing additional attempts at peripheral blood stem-cell mobilization using other methods. Similarly, marrow harvest generally yields low numbers of CD34+ cells in these individuals. The combination of plerixafor and G-CSF was reported to successfully remobilize more than 60% of patients with non-Hodgkin lymphoma who failed to procure adequate numbers of CD34+ cells with G-CSF alone.²⁶ In those at high risk for poor mobilization, the use of plerixafor in combination with G-CSF is a reasonable strategy.

We have recently identified another circumstance where even patients who attain adequate CD34+ cell numbers are at risk for delayed hematopoietic recovery. We studied the influence of the number of aphereses required to obtain adequate ($\geq 2 \times 10^6$) CD34+ cells, based on our suspicion that those patients requiring numerous procedures might not only have fewer CD34+ cells circulating, but also might have lower proportions of CD34+ cells with marrow repopulating potential. It is known that well-trained marathoners mobilize CD34+ cells while running

a marathon, but these mobilized cells are CD34 and CD33+ differentiated progenitors, not CD34+ and CD33- cells in which repopulating potential exists.²⁷ Our patients who required more apheresis days, particularly those requiring five or more procedures, had significantly slower neutrophil recovery, slower platelet recovery, and a higher risk for failure to achieve platelet recovery within eight weeks post-transplantation.²⁸ These data serve as a reminder that CD34+ cell number is only a surrogate for the cells that repopulate the marrow. Additional clinical circumstances may be identified in which its usefulness as a single predictor of hematopoietic recovery is limited. Interestingly, the need for numerous aphereses also identified patients at high risk for the development of therapy-related myelodysplastic syndrome (MDS)/acute myelogenous leukemia (AML), as had been previously reported.²⁹

As the number of autologous transplants performed annually continues to increase,³⁰ it is fitting that the mechanisms of stem-cell mobilization are stimulating targeted approaches. The addition of plerixafor has been an important advance. In proper clinical circumstances, the integration of chemotherapy-based methods of mobilization is efficient and appropriate. In patients in whom additional chemotherapy is not useful, its use for mobilization alone is not warranted. Careful risk stratification may further improve efficiency so that the approach to mobilization is individualized according to patient characteristics. Identification of patients at high risk for delayed hematologic recovery despite adequate CD34+ cell harvests is a fertile area for further work. ■

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