Preleukapheresis peripheral blood CD34+ cells predict progenitor cell collection yield and the necessary number of procedures to undergo

Sir,

We evaluated the peripheral blood (PB) CD34+ cell content as a predictive parameter of the leukapheresis CD34+ cell yield. Regression analysis showed that a preleukapheresis CD34+ cell concentration of ≥ 40/μL predicted a yield of ≥2×10⁶ CD34+ cells/kg by a single leukapheresis (r = 0.83, p = 0.0001). In addition, CD34+ cell concentrations in preleukaphereis PB ≤ 30/μL and ≤ 15/μL were associated with the need for at least two (p = 0.0028) or at least three (p = 0.02) procedures respectively in order to obtain ≥ 2×10⁶ CD34+ cells/kg.

We studied CD34+ cell concentration in preleukaphereses PB samples and CD34+ cell yield in a number of aphereses to investigate whether these parameters are related. The aim of our work was: a) to establish a statistical relationship between both parameters which would allow us to calculate the threshold concentration of immediate preleukapheresis PB CD34+ cells necessary to obtain ≥ 2×10⁶/kg CD34+ cells in a single apheresis; b) to determine the number of procedures necessary to obtain ≥2×10⁶ CD34+ cells/kg.

CD34+ cells were analyzed in PB samples in patients mobilized either with rhG-CSF or following chemotherapy plus rhG-CSF. Underlying diseases were: breast carcinoma (n= 56), Hodgkin's disease (n=5), non-Hodgkin's lymphoma (n= 12), multiple myeloma (n= 13), acute leukemia (n=4) and CML (n= 1).

Ten liter leukaphereses were performed until more than 2×10⁶ CD34+ cells/kg had been collected. A total of 218 aphereses were evaluable for CD34+ counts. Evaluated paired data (PB-apheresis) corresponding to the first, second or subsequent apheresis procedures involved 87, 80 and 51 samples respectively.

Processing of samples was performed as reported elsewhere[1] with FITC-conjugated CD34 (anti-HPCA-2; Becton Dickinson, Mountain View, CA, USA). Fifty thousand mononuclear cells were analyzed in each sample.

The median concentration of CD34+ cells in preleukapheresis PB samples was 11.96/μL (range: 0.9-1035). The median CD34+ cell count per leukapheresis was 0.61×10⁶/kg (range 0.03-22.51). The results obtained for these parameters are summarized in Table 1.

Preleukapheresis PB CD34+ cell counts showed a strong correlation with harvested CD34+ cell counts per kilogram (r = 0.83, p=0.0001). Linear regression analysis based on 218 paired samples (Figure 1) showed that a preleukapheresis CD34+ cell concentration ≥40/μL predicted that ≥ 2×10⁶ CD34+ cells/kg could be collected by a single leukapheresis. The same analysis showed that target yields of ≥1.5, ≥1 and ≥0.75×10⁶ CD34+ cells/kg could be predicted with preleukaphereis PB CD34+ cells/μL of ≥30, ≥16 and ≥11, respectively.

Table 1. Correlation analysis between circulating CD34+ cells and PBPC collection yields. Preleukapheresis CD34+ cell counts correlated with CD34+ cells both when considering first or second procedures independently and when evaluating all procedures. Results are given as median and range values.

	PB CD34+ (Cell/mL)	*Apheresis CD34+ (Cell x10⁶/kg)*	*Correlation*
All aphereses	11.96 (0.9-1035)	0.61 (0.03-22.51)	r=0.83 p=0.0001
Apheresis 1	9.35 (1.42-1035)	0.71 (0.03-22.51)	r=0.91 p=0.0001
Apheresis 2	13.93 (0.9-162.13)	0.76 (0.03-12.68)	r=0.84 p=0.0001

We applied Student's t test to compare PB CD34+ cell counts in patients who had undergone one, two or more and three or more procedures. In this analysis, we found that mean PB CD34+ cell concentrations ≤ 30/μL and ≤ 15/μL were associated with the need to perform at least two (p= 0.0028) or at least three (p=0.02) apheresis procedures, respectively, to obtain ≥2×10⁶ CD34+ cells/kg.

In PBSCA, the estimation of CD34+ cell yield prior to initiating apheresis procedures,[2-8] has both clinical and economic implications. In the present study, patients with a variety of underlying diseases, premobilization treatments and mobilization schedules

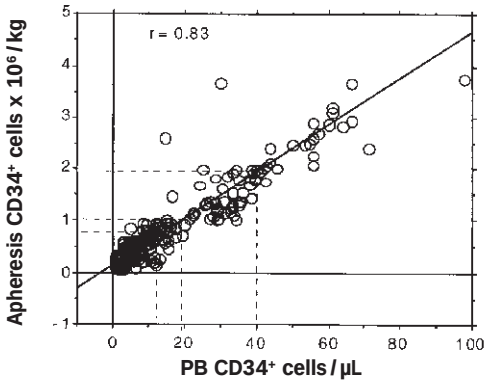

Figure 1. Linear regression analysis of CD34+ cells/μL and yield of CD34+ cells/kg. A number of CD34+cells ≥ 40/μL in the peripheral blood is highly predictive for the collection of ≥ 2×10⁶ CD34+/kg in a standard apheresis procedure of 10 liters.

were evaluated. Regardless of the previous variables, a preleukapheresis PB $CD34^+$ cell concentration $\geq 40/\mu L$ was significantly related to the collection of at least 2×10^6 $CD34^+$ cells/kg in a single apheresis, as previously reported.[9,10] In addition to the above data, we found that to obtain a target number of 2×10^6 $CD34^+$ cells/kg, PB $CD34^+$ cell concentrations $\leq 30/\mu L$ are associated with the need for at least two leukapheresis procedures and PB concentrations $\leq 15/\mu L$ are associated with the need for at least three procedures. In conclusion, our study shows that preleukapheresis PB $CD34^+$ cell concentration can be used to guide PBPC harvest by predicting both the total $CD34^+$ cell yield and the number of aphereses needed to be undergone.

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Phenotypic changes in neutrophils after rhG-CSF administration in non-Hodgkin's lymphoma patients undergoing PBSC transplantation or conventional chemotherapy

Sir,

rhG-CSF induces several phenotypic changes in neutrophils. Increased HLA-DR expression and decreased CD10 expression have recently been described in neutrophils from some patients after rhG-CSF therapy. We evaluated these parameters in 12 non-Hodgkin's lymphoma patients undergoing either PBSC transplantation after high-dose chemotherapy or conventional chemotherapy. The appearance of an HLA-DR-positive neutrophil subpopulation, along with a marked decrease in CD10 expression, was confirmed. However, despite this immature phenotype, rhG-CSF-induced neutrophils displayed enhanced phagocytosis and chemiluminescence.

Recombinant human granulocyte colony-stimulating factor (rhG-CSF) induces several changes in neutrophils.[1,2] Recently, Zarco *et al*.[3] described new phenotypic findings in rhG-CSF-induced neutrophils in six ALL patients undergoing chemotherapy. The appearance of an HLA-DR-positive neutrophil subpopulation, along with a decrease in the percentage of $CD10^+$ neutrophils, appeared of particular interest.

We reviewed the clinical files of patients recently treated with rhG-CSF (Filgrastim) for whom analysis of HLA-DR and CD10 expression on circulating neutrophils before and after rhG-CSF administration was available. Twelve patients (4 females, 8 males), with intermediate and high grade non-Hodgkin's lymphoma (NHL) were evaluated. Six patients had been treated with autologous peripheral blood stem cells (PBSC) transplantation after high-dose chemotherapy,[4] and neutrophils had been studied before the conditioning regimen and after engraftment (i.e. neutrophils > 0.5×10^9/L, and platelets > 20×10^9/L), stimulated by rhG-CSF (5 mg/kg/day). The other 6 patients had been studied before the first course of chemotherapy (Promice-Cytabom)[5] and after a five-day course of rhG-CSF (5 µg/kg/day), administered to