Journal of Blood Medicine

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ORIGINAL RESEARCH

Prediction of Viable CD34 Count in Harvested Product by Peripheral Blood Hematopoietic Progenitor Count of Automated Hematology Analyzer Undergoing Hematopoietic Stem Cell Transplantation

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Introduction: The CD34+ hematopoietic cell count was used to define cell harvest goals. Successful peripheral blood stem cell transplantation depends on infusion of an appropriate number of HPCs to achieve rapid and durable hematologic recovery.

Purpose: In this study, we evaluated the use of the Hematopoietic Progenitor Cell count program on the Sysmex XN-3000 hematology analyzer as an effective parameter for enumerating CD34+ cells.

Patients and Methods: Whole blood samples from 144 subjects who are either healthy donors or patients scheduled to undergo peripheral blood stem cell collection were collected and hemopoietic stem cells were quantified using CD34 cell enumeration by flow cytometry and XN-HPC by hematology analyzer.

Results: The correlation between the two methods was high (r = 0.766; 95% CI: 0.702–0.818). Passing–Bablok showed an intercept at 3.45 (2.54 to 4.74) with a slope of 0.78 (95% CI 0.69 to 0.89). Residual analysis of this model indicated no significant deviation from linearity (p = 0.360). The receiver operating characteristic curve demonstrated an area under curve to be 0.88 (0.82 to 0.92), with a positive predictive value of 80.3%. The correlation between CD34+ and XN-HPC showed a strong relationship and good agreement with minimal bias.

Conclusion: The XN-HPC showed good analytical performance. With the increasing requirements for stem cell transplantation, a technically simple and rapid alternative for stem cell enumeration that is sustainable is highly useful.

Keywords: XN-HPC, CD34+, progenitor, leukemia, allogeneic, autologous

Introduction

Hematopoietic stem cell transplantation (HSCT) uses hematopoietic progenitor cells or stem cells (HSC) to replace diseased bone marrow with healthy progenitor cells to repopulate and propagate the bone marrow.[1,2] HSCT is a well-established treatment for high-risk malignant and non-malignant hematologic diseases, immunologic or metabolic disorders, and solid tumours.[3–6] For haematological malignancies, the source of stem cells has shifted from bone marrow collection, which is an invasive procedure, to a much less invasive method in the form of peripheral blood stem cell (PBSC) collection.[7] PBSC transplantation can be summarized into several distinct phases, but the most crucial phase that would allow for successful transplantation is the first phase, which is the mobilization or movement of stem cells from

Received: 6 March 2024
Accepted: 12 June 2024
Published: 22 August 2024

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the marrow into the peripheral blood induced by timely administration of specific human hematopoietic granulocyte-colony stimulating factors.[8] HSCs used for HSCT in eligible patients can be collected from healthy donors for allogeneic HSCT or, in some instances, the progenitors can be collected from the patients themselves for autologous HSCT. Both procedures require careful consideration by the HSCT transplant team.[7]

The mobilization of stem cells requires close monitoring of stem cell counts that have migrated into the circulation. To ensure the success of the mobilization phase, several authors have recommended that peripheral blood samples be collected daily for CD34+ cell enumeration by flow cytometry analysis until the CD34+ cell count is between 8 and 20 CD34+ cells/μL.[9,10] Donors or patients who do not reach this threshold are considered to have failed PBSC collection and would be counselled for a bone marrow harvest to be performed under general anesthesia. The CD34+ cell dose or the absolute number of CD34+ cells is the best available predictor of graft quality.[11,12]

For a peripheral blood stem cell (PBSC) transplant to be successful, the right amount of HSCs must be infused in order to promote a fast and long-lasting haematologic recovery. The International Society of Hematotherapy and Graft Engineering (ISHAGE) has developed a procedure that relies on flow cytometry to count CD34+ cells for the assessment of circulating HSCs and for determining when to begin apheresis. CD34+ cell count is used as a marker of bone marrow response to stem cell mobilisation accurately that is safe and harmless to the donor.

CD34+ cell count is used to define cell harvest goals and guide growth factor administration and the number of leukapheresis sessions necessary. However, this method is quite costly, as multiple flow cytometry runs must be performed while monitoring the mobilization phase. Our centre uses a threshold of ≥10 cells/μL when deciding whether to proceed with PBSC harvesting for allogeneic and autologous transplantation. Since the first XN model was equipped with the XN-HPC program, many research groups have studied this parameter to ensure its suitability for stem cell enumeration. However, the suggested XN-HPC cutoff values were variable and should be determined for each centre. Sysmex XN analyser uses fluorescence flow cytometry to analyse physiological and chemical properties of cells. It provides the information about cell size, cell structure and cell interior. HPC is counted in the WPC channel of XN analyser. With its unique combination of reagents, WPC channel detects abnormal membrane composition and nuclear content. The lipid membrane composition of immature cells is different from that of mature cells or abnormal blasts. Stem cells' membranes are relatively resistant to permeabilisation by the WPC reagent. As a result, stem cells are medium in size (medium FSC), have a low granularity (low SSC) and relatively low fluorescence intensity (low-medium SFL). In the Sysmex XN analyser, HPCs can be measured in a simple and reliable method within a few minutes without the need for manual gating, pre-treatment or sample washing as compared to immune flow cytometry measurement. However, this requires further validation and comparison with the larger community of XN-HPC users, as this would allow optimization of the timing of apheresis. Therefore, the main aim of this study was to compare HSC enumeration by XN-HPC and CD34+ enumeration by flow cytometry and to determine the correlation and accuracy of its identification.

Materials and Methods

Study Design

This prospective observational study was performed at the National Hematology Referral Centre in Hospital Ampang, Malaysia. Informed consent was obtained before we recruited all healthy Malaysian donors and patients who qualified for autologous hematopoietic stem cell transplantation, regardless of age or gender. Non-Malaysian citizens, and people who were unable or unwilling to give written consent were all excluded from this study.

We recruited patients with Acute Leukaemia, Multiple Myeloma, Hodgkin Lymphoma and non-Hodgkin lymphoma for our allogeneic and autologous stem cell transplantation cases. Peripheral blood stem cells were mobilised following our institution protocol using granulocyte-colony stimulating factor (G-CSF) alone or in combination with chemotherapy. The chemotherapy treatment was followed by administration of G-CSF at a dose of 5 μg/kg/day for patients, and healthy donors were only administered G-CSF at a dose of 10 μg/kg/day. The CD34+ assessment was performed starting from 3 to 4 days after the first administration of G-CSF in healthy donors and after 10–15 days after chemotherapy. Apheresis was started when the quantity of CD34+ cells in PB was at least 20×10^6/L, or in some special circumstances, between 10 and 20×10^6/L, depending on the patient's characteristics.

Peripheral blood for stem cell enumeration was collected preharvest as scheduled by the transplant team and analyzed using the CD34+ cell count and XN-HPC program. This study was reviewed and approved by the Medical Research and Ethics Committee of the Ministry of Health, Malaysia (NMRR-20-2585-57209).

Stem Cell Enumeration by CD34+ Cell Count

Peripheral blood was used for CD34+ HSC enumeration using the BD Stem Cell Enumeration kit on a FACS Calibur flow cytometry analyzer (BD Biosciences, CA, USA) according to the International Society of Hematotherapy and Graft Engineering (ISHAGE) protocol.[13] All samples were collected in K2EDTA tubes, stored at room temperature, and processed within 2 hours of collection. The flow cytometry method for stem cell enumeration uses special beads coated with monoclonal antibodies against CD34 and CD45 and a viability dye. Stem cells were identified using low-side scatter (SSC) CD45+ and CD34+ gating strategies. During sample acquisition, direct volumetric control was used to establish the stem cell concentration.

Stem Cell Enumeration by XN-HPC Programme

The same peripheral blood sample was used for HSC enumeration using the XN-HPC program on a Sysmex XN-3000 hematology analyzer (Sysmex Corporation, Kobe, Japan). HSC identification uses a semiconductor laser beam at a wavelength of 633 nm, which is emitted to the blood cells passing through the flow of cells upon acquisition into this channel. This channel, also known as the white blood cell progenitor and pathogenic cells (WPC), is a fluorescence channel that provides information about the cells in the form of forward-scattered and side-scattered light that is presented as a plot that gates for the specific pattern for stem cell identification and enumeration. Forward scattered light hits the cells directly and emits information about the diameter of the cell that translates as the size of the cells, while side-scattered light hits the cell at an angle and emits information about the cytoplasmic properties or complexity, which is usually low for stem cells. Three consecutive runs were performed for each sample to ensure within run precision.

Pre-Study Evaluation of Sysmex XN-HPC Performance

Repeatability of XN-HPC stem cell enumeration was evaluated by measuring five samples comprising three normal subjects and two XN-CHECK control materials, which were tested on the Sysmex XN-HPC ten times consecutively. The average coefficient of variation (CV%) was 19.7% (range 3.7–27.2%). Instrument precision limit was 30%. For stability testing, test samples stored at room temperature and 4 °C were determined by XN-HPC measurements after 0, 2, 4, 6, and 8 hours. Stability test results were assessed using two-way analysis of variance. The samples were stable for at least 8 hours.

Statistical Analysis

Continuous variables are described as means with standard deviations for normally distributed data or medians with a range for skewed data. Categorical variables were described as frequencies and percentages. The Wilcoxon signed-rank test was used to compare the median values of CD34+ and XN-HPC cell counts. The relationship between CD34+ and XN-HPC cell counts was further analyzed using a linear regression model. Passing and Bablok method was used to estimate the slope and intercept with 95% confidence interval (CI). Using Spearman correlation, the correlation coefficient, or r and its 95% confidence interval were determined. To determine the degree of agreement between the two measurements, the Bland–Altman analysis was used. The difference between the two measurements is shown against the average of the two measurements along with the bounds of agreement using the Bland and Altman plots. Ninety-five percent of the mean difference data should fall within the lower and upper limits of the agreement range, which signifies the measurement accuracy. Receiver operating characteristic (ROC) curve analysis was used to evaluate the diagnostic accuracy of XN-HPC. The optimal cutoff point was determined using the Youden index. Based on earlier research,[14,15] sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were computed at the appropriate XN-HPC count that best predicted the CD34+ count at $10.0x10^6$/L and $20.0x10^6$/L.[16]

Results

Study Population

Patients and donors were referred to the Stem Cell Transplant Unit of the Department of Hematology, Hospital Ampang, Malaysia, for allogeneic or autologous peripheral blood stem cell collection and transplantation. A total of 201 mobilised peripheral blood samples were randomly chosen for preharvest processing out of 39 healthy donors and 105 patients who received PBSC transplantation. Thirty were healthy donors and nine were matched unrelated donors. The majority were adults (n = 133) with a few subjects being less than 18 years old (n = 11). The characteristics of the patients and donors and the number of peripheral blood samples in each group are listed in Table 1. The overall median HSC concentrations were not significantly different across subjects. The median CD34+ cell count was 17.33 $x10^6$/L (interquartile range: 0.67–115.67 $x10^6$/L), whereas the median XN-HPC cell count was 13.64 $x10^6$/L (interquartile range: 0.06–152.31 $x10^6$/L).

Correlation and Agreement Studies

The correlation of CD34+ and XN-HPC cell counts in 201 preharvest peripheral blood sample collections is shown in Table 2 and Figure 1. A good agreement was observed between the two methods for all 201 samples. All preharvest samples showed strong correlation (r = 0.77, 95% CI: 0.70–0.82). The regression line had an intercept of 3.45 (2.54 to 4.74) and a slope of 0.78 (0.69 to 0.89). The regression equation was defined as XN-HPC = 3.45 + 0.78 (CD34+) (Figure 1A). The residual analysis of this model indicated no significant deviation from linearity (P = 0.360). The agreement between the two methods was evaluated using Bland–Altman plot (Figure 1B) that showed good agreement with minimal bias between CD34+ and XN-HPC up to a certain range. About 94% of the data points lie within the ±2 SD of the mean of difference. Good agreement and no systematic difference were shown between the CD34+ cell count and XN-HPC at a range of 0 to 15. Thirteen or 6.4% were identified as outliers, and the XN-HPC count differed from CD34+ cell counts by greater than 1 SD above or below the limit of agreement line. For allogeneic preharvest samples, peripheral blood samples from healthy allogeneic donors (N = 42), the correlation strength was moderate (r = 0.69, slope 0.61, 95% CI: 0.40–0.85) (Table 2). A strong correlation was also found in 20 samples collected from multiple myeloma (MM) patients (r = 0.78, slope 1.24, 95% CI: 0.83–1.77), despite a modest underestimation of XN-HPC noted in the Passing–Bablok regression analysis (95% CI of slope: 1.24–0.23) and Bland–Altman plot (95% CI of mean bias: −2.94 to 17.42) (Figure 1).

Receiver Operating Characteristics (ROC) Analysis and Diagnostic Accuracy

Table 3 shows the overall results of sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) corresponding to XN-HPC count, which best predicted CD34+ cell count at ≥10×106/L and ≥20×106/L 14,15. The value of area under the curve (AUC) with 95% confidence interval are reported. The overall sensitivity and specificity for XN-HPC count of ≥$10×10^6$/L were 90.7% and 74.2%, respectively, compared to CD34+ cell count of ≥$20×10^6$/L (sensitivity: 89.3%, specificity: 73.5%). Based on the CD34+ cell count by flow cytometry, 93 of the samples were less than $10×10^6$/L. The diagnostic accuracy

Table 1 Characteristics of Patients and Donors, Number of Mobilised Peripheral Blood Samples and Values Distribution of Haemopoietic Stem Cells Using CD34 Positive and XN-HPC Cell Counts

Characteristics	n	Age, Median (Range)	Samples, n	CD34+×10^6/L Median (Range)	XN-HPC×10^6/L Median (Range)	p-value*
All	144	37 (11-71)	201	17.33 (0.67–115.67)	13.64 (0.06–152.31)	0.387
Female	62	40 (26-71)				
Male	82	35 (11-67)				
Allogeneic donors	39	30 (11-67)	42	31.17 (6.33–101.67)	39.07 (5.72–152.31)	< 0.001
Autologous patients	105	40 (12-71)				
Multiple myeloma (MM)	20	57.5 (16-71)	25	25.33 (3.33–115.67)	25.02 (2.51–94.83)	0.083
Lymphoma	79	31 (12-62)	128	12.00 (0.67–97.33)	7.89 (0.06–138.88)	0.004
Leukemia	6	44.5 (22-69)	6	7.17 (1.67–44.67)	14.59 (0.73–87.50)	0.917

Note: *Wilcoxon signed-rank test: Comparison between the median values of HPC and CD34 + counts.

Table 2 Comparison Between XN-HPC and CD34+ Cell Counts

Group	r value[a]	Passing–Bablok Regression*		Bland–Altman Difference Plot (×10⁶/L)	
		Slope (95% CI)	Intercept[b] (95% CI)	Mean Bias** (95% CI)	95% Limits of Agreement (Mean Bias ±1.96 SD)
All	0.766	0.78 (0.69 to 0.89)	3.45 (2.54 to 4.74)	−1.56 (−4.81 to 1.69)	−47.36 to 44.23
Allogeneic donors	0.690	0.61 (0.40 to 0.85)	3.98 (−5.41 to 11.29)	−16.31 (−24.97 to −7.64)	−70.79 to 38.18
Autologous patients					
MM	0.781	1.24 (0.83 to 1.77)	0.23 (−5.35 to 6.50)	7.24 (−2.94 to 17.42)	−41.11 to 55.59
Lymphoma	0.742	0.90 (0.77 to 1.06)	2.83 (2.17 to 4.25)	2.03 (−1.17 to 5.24)	−33.89 to 37.96
Leukemia	0.086	1.12 (−0.23 to 2.58)	2.18 (−24.12 to 16.89)	−11.73 (−50.64 to 27.19)	−84.40 to 60.95

Notes: [a]r value: Spearman correlation. [b]Intercept: 95% Confidence Interval (CI): ×10⁶/L. *Passing–Bablok regression; XN-HPC = Intercept + Slope (CD34+). **Mean bias = XN-HPC - CD34+.

of XN-HPC was assessed using the AUC in the ROC analysis (Figure 2). ROC curves were generated to investigate the ability of preharvest HPC concentrations to predict sufficient CD34+ cell harvest. ROC analysis showed excellent test performance for the preharvest XN-HPC concentration (AUC: 0.86 ≥20/μL CD34+). We also generated ROC curves for preharvest HPC concentrations to predict a sufficient concentration of CD34+ cells for PBSC harvest, which was defined as ≥10×10⁶/L CD34+, as the expected threshold. The test performance was also very good (AUC: 0.88 ranging 0.82–0.92). The Youden index method was used to determine the optimal cutoff point. We identified the cut-off values of the XN-HPC count capable of maximizing its efficiency to be used as a "rule-in" and "rule-out" test for starting apheresis.

Discussion

In the current study, we attempted to establish the correlation between XN-HPC and CD34+ hematopoietic stem cell enumeration after mobilization and whether XN-HPC cut-off values could predict the optimal numbers of CD34+ HSC before PBSC harvest was initiated. We recruited subjects that included both allogeneic donors and candidates for autologous transplantation, as in previous studies.[14,15,17–19] Factors that could influence the yield of stem cells included method of collection, timing of the analysis and storage. To minimize and work around the possible biases, the phlebotomist and operators of both the CD34+ HSC enumeration by flow cytometry and the XN-HPC method were the same person throughout the study. The timing of venipuncture was also standardized in the morning session and samples were analyzed immediately upon collection. Overall, the correlation and ROC analyses showed a strong correlation between the two methods and were comparable to the values previously reported.[17–19] Positive bias was very small at 1.5% and 95% limits of agreement of −47 to

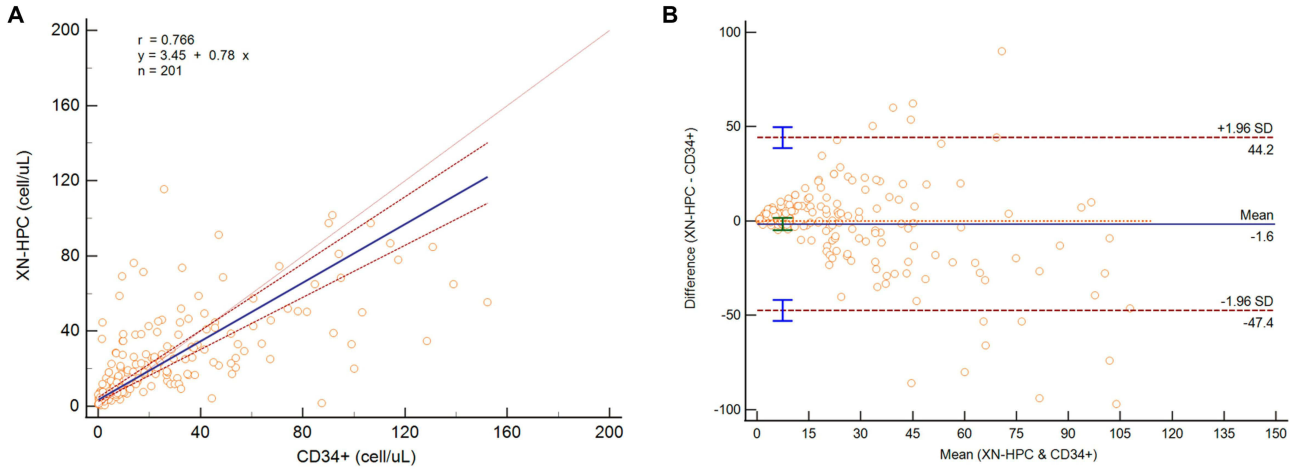

Figure 1 Correlation between XN-HPC and CD34+ cell count analysis using (**A**) Passing and Bablok regression analysis and (**B**) Bland–Altman analysis.

Table 3 Results of Receiver Operating Characteristics Curve Analysis

Total Samples, n	Cut-off CD34+[a]	Number of Positives[b]	AUC (95% CI)	Cut-off XN-HPC	Sensitivity	Specificity	PPV	NPV
All, 201	**≥10.0**	108	0.88 (0.82 to 0.92)	>11.6[c]	90.7	74.2	80.3	87.3
Donor, 42	**≥10.0**	39	0.94 (0.83 to 1.00)	>17.0[c]	82.1	100	100	30.0
MM, 25	**≥10.0**	16	1.00 (NA to NA)	>13.6[c]	100	100	100	100
Lymphoma, 128	**≥10.0**	49	0.85 (0.78 to 0.91)	>11.6[c]	87.8	72.2	66.2	90.5
Leukemia, 6	**≥10.0**	4	0.75 (0.33 to 1.00)	>6.67[c]	75.0	100	100	66.7
All, 201	**≥20.0**	84	0.86 (0.81 to 0.91)	>16.0[d]	89.3	73.5	70.8	90.5
Donor, 42	**≥20.0**	37	0.92 (0.83 to 1.00)	>18.0[d]	81.1	100	100	41.7
MM, 25	**≥20.0**	14	0.97 (0.92 to 1.00)	>13.6[d]	100	81.8	87.5	100
Lymphoma, 128	**≥20.0**	31	0.83 (0.76 to 0.91)	>16.0[d]	90.3	73.2	51.9	95.9
Leukemia, 6	**≥20.0**	2	0.50 (0.00 to 1.00)	>19.3[d]	50.0	100	100	80.0

Notes: [a] Cut-off value of CD34+ cell count ($\times 10^6$/L). [b] Number of CD34+ positive samples. [c] Cut-off value of XN-HPC count ($\times 10^6$/L) that optimally predict PB CD34+ ≥10.0×10^6/L. [d] Cut-off value of XN-HPC count ($\times 10^6$/L) that optimally predict PB CD34+ ≥20.0×10^6/L.

44%. However, the significant difference in HSC enumeration values was notable in the allogeneic donor and lymphoma patient groups (Table 1). For allogeneic donors, Passing–Bablok and Bland–Altman analyses showed that the correlation was good with a negative mean bias of −16.31% indicating a tendency of HSC underestimation using the XN-HPC and 95% limits of agreement of −70.79 to 38.18%. For the lymphoma group, the analyses showed a strong correlation with a positive mean bias of 2.03%, indicating the opposite and 95% limit of agreement of −33.89 to −37.96%.

In the transplant setting, both these potential scenarios would influence the decision on whether to proceed with PBSC harvest or whether further mobilization is required. Therefore, we tested the XN-HPC enumeration values by applying our centre's threshold value of ≥10.0×10^6/L CD34+ HSC, indicating successful mobilization (Table 3). Overall, 53.7% (108 of 201) mobilized peripheral blood samples had a CD34+ cell count ≥10.0×10^6/L by flow cytometry and were identified at a slightly higher XN-HPC cut-off value of 11.6 x10^6/L. ROC curve analysis of XN-HPC count yielded very good accuracy. The sensitivity of this approach was excellent at 90.7%, with high specificity of 74.2%. This indicated that XN-HPC was able to detect and measure CD34+ HSC correctly (Table 3). This was also reflected in the very high positive predictive and negative predictive

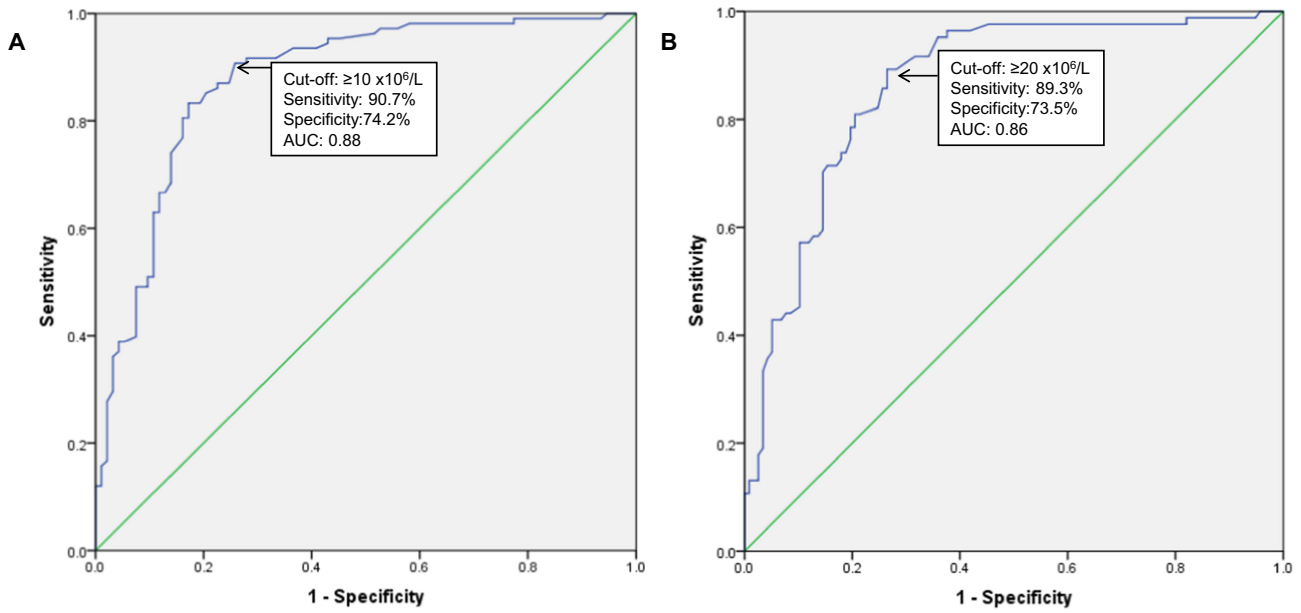

Figure 2 ROC curve for all patients and donors to determine the optimal cut-off for which the HPC concentration can reliably predict CD34+ cell concentration (**A**) ≥10 x10^6/L AUC: 0.88 and (**B**) ≥20 x10^6/L AUC: 0.86.

values of 80.3% and 87.3%, respectively (Figure 2). A few studies have noted significant differences in HSC enumeration in samples from multiple myeloma patients.[15,17] Our findings indicated that although the sample size was small for this group of samples, there was no significant difference between the two methods while the sensitivity and sensitivity were very high. Some authors have indicated caution when using a single cut-off value, as this may cause too many patients to be eligible for harvest before they are adequately mobilized. The laboratory may consider a positive cutoff with high specificity before the initiation of the PBSC harvest. We analysed different XN-HPC cutoff values for each of the different sources of samples from patients who were allogeneic donors or candidates for autologous transplantation. Different cutoff values were derived that maintained high sensitivity and specificity. There was not much difference in the cutoff values, except for the donor category. The highest XN-HPC cut-off was with allogeneic donors at 17.0×10^6/L that could optimally predict CD34+ HSC of $\geq 10.0 \times 10^6$/L but with very good sensitivity and specificity. Autologous samples from leukemia patients showed the lowest cut-off value, but the number of samples for this category was too small.

Overall, our results show a strong correlation with high accuracy. Individual cutoffs should be established for every laboratory that is invested in using the marker for CD34+ enumeration. The use of XN-HPC concentration as a surrogate for CD34+ cell concentration derived from the regression equation may be integrated into the current preharvest workflow with a timely evaluation by CD34+ cell enumeration by flow cytometry. As a surrogate, serial XN-HPC values should be obtained at predetermined time points to increase its specificity. Each transplant centre should determine which patients would be suitable for XN-HPC monitoring and which patients would benefit from CD34+ cell count by flow cytometry. The inherent variability seen across different sample categories, although showing good positive predictive values, may be related to the small number of samples analyzed for each category. The role of previous chemotherapy in autologous samples and its effect on the quality of HSC remain largely unknown. Involvement in the interlaboratory analysis of the same sample is warranted to ensure the robustness of the test platform over time. Most studies agree that the XN-HPC program is fast, simple to use, does not require experienced operators, and is affordable, especially for monitoring purposes.[14,18,19] However, poor agreement between the two methods especially in the clinical decision range and when stem cells were detected at very low concentrations has been reported.[15] It is also crucial that other sources of stem cells can be measured, including marrow, cord blood, and cryopreserved products, especially when matched unrelated donors are not available.

Rapid and durable hematological recovery is dependent on successful peripheral blood stem cell transplantation, which relies on the infusion of an appropriate number of hematopoietic stem cells.[1,20] Therefore, adequately mobilized hematopoietic stem cells in the peripheral blood are crucial before PBSC harvesting is performed. As the main transplant centre in Malaysia, the increasing number of patients brought in for stem cell transplantation to consolidate remission status post-chemotherapy requires a sustainable approach to monitoring stem cell concentration before PBSC harvest. Therefore, incorporating this method for CD34+ monitoring would improve the efficiency of transplant services in general and easing financial constraints while maintaining the availability of the service. Moving forward, a testing algorithm that incorporates XN-HPC in the monitoring of HSC mobilized as well as establishing triggers for CD34+ cell enumeration by flow cytometry will be proposed to evaluate the effectiveness of certain cutoff values for XN-HPC for allogeneic and autologous transplantation.

Conclusion

Our study confirms a strong correlation between XN-HPC count and $CD34^+$ cell count and could be a useful surrogate test to assess optimal timing for PBSC collection. It can be used as an alternative method for CD34+ cell count. However, a workflow that would incorporate the XN-HPC count in daily peripheral blood cell count monitoring during mobilization that is used for screening and confirmation with CD34+ cell count at least one day prior to or on the day of the intended peripheral blood stem cell harvest would greatly increase the sustainability of bone marrow transplant unit services. Further prospective studies are recommended to evaluate the effectiveness of a working algorithm that incorporates XN-HPC cell counts in both healthy donors and autologous patients.

Ethics Approval and Consent to Participate

This study was approved by the Medical Research and Ethics Committee of the Ministry of Health Malaysia (NMRR-20-2585-57209). Study participants provided informed consent to participate. Research was conducted in line with the Belmont Report and the Declaration of Helsinki.

Acknowledgments

Special thanks to Nur Azrin Binti Abu Bakar and Norazrina Masani for data collection and Tg Mohd Ikhwan Tg Abu Bakar Sidik for helping with the data presentation. The authors thank all volunteers who consented to participate in the study, as well as the clinical and laboratory staff of the Department of Hematology, Hospital Ampang, for their various contributions to the study.

Disclosure

The authors declare that they have no known competing financial interests or personal relationships that could have influenced the work reported in this study. None of the authors were reimbursed for conducting this study.

References

1. Copelan EA. Hematopoietic stem-cell transplantation. *N Engl J Med*. 2006;354(17):1813–1826. doi:10.1056/NEJMra052638
2. Hatzimichael E, Tuthill M. Hematopoietic stem cell transplantation. *Stem Cells Cloning*. 2010;3:105. doi:10.2147/SCCAA.S6815
3. Buckley RH, Schiff SE, Schiff RI, et al. Hematopoietic stem-cell transplantation for the treatment of severe combined immunodeficiency. *N Engl J Med*. 1999;340(7):508–516. doi:10.1056/NEJM199902183400703
4. Hsieh MM, Kang EM, Fitzhugh CD, et al. Allogeneic hematopoietic stem-cell transplantation for sickle cell disease. *N Engl J Med*. 2009;361(24):2309–2317. doi:10.1056/NEJMoa0904971
5. Gratwohl A, Baldomero H, Aljurf M, et al. Hematopoietic stem cell transplantation: a global perspective. *JAMA*. 2010;303(16):1617–1624. doi:10.1001/jama.2010.491
6. Henig I, Zuckerman T. Hematopoietic stem cell transplantation—50 years of evolution and future perspectives. *Rambam Maimonides Med J*. 2014;5(4):e0028. doi:10.5041/RMMJ.10162
7. Körbling M, Freireich EJ. Twenty-five years of peripheral blood stem cell transplantation. *Blood J Am Soc Hematol*. 2011;117(24):6411–6416.
8. Takeyama K, Ohto H. PBSC mobilization. *Transfus Apheresis Sci*. 2004;31(3):233–243. doi:10.1016/j.transci.2004.09.007
9. Tricot G, Jagannath S, Vesole D, et al. Peripheral Blood Stem Cell Transplants for Multiple Myeloma: identification of Favorable Variables for Rapid Engraftment in 225 Patients. *Blood*. 1995;85(2):588–596. doi:10.1182/blood.V85.2.588.588
10. Weaver CH, Hazelton B, Birch R, et al. An Analysis of Engraftment Kinetics as a Function of the CD34 Content of Peripheral Blood Progenitor Cell Collections in 692 Patients After the Administration of Myeloablative Chemotherapy. *Blood*. 1995;86(10):3961–3969. doi:10.1182/blood.V86.10.3961.bloodjournal86103961
11. Powles R, Mehta J, Kulkarni S, et al. Allogeneic blood and bone-marrow stem-cell transplantation in haematological malignant diseases: a randomised trial. *Lancet*. 2000;355(9211):1231–1237. doi:10.1016/S0140-6736(00)02090-0
12. Vose JM, Sharp G, Chan WC, et al. Autologous transplantation for aggressive non-Hodgkin's lymphoma: results of a randomized trial evaluating graft source and minimal residual disease. *J Clin Oncol*. 2002;20(9):2344–2352. doi:10.1200/JCO.2002.09.138
13. Sutherland DR, Anderson L, Keeney M, Nayar R, Chin-Yee I. The ISHAGE guidelines for CD34+ cell determination by flow cytometry. *J Chemother*. 1996;5(3):213–226. doi:10.1089/scd.1.1996.5.213
14. Dima F, Barison E, Midolo M, Benedetti F, Lippi G. Assessment of haematopoietic progenitor cell counting with the Sysmex® XN-1000 to guide timing of apheresis of peripheral blood stem cells. *Blood Transfusion*. 2020;18(1):67. doi:10.2450/2019.0086-19
15. Söderström A, Møller BK, Sørensen BS. Evaluation of the Sysmex XN automated hematopoietic progenitor cell enumeration for timing of peripheral blood stem cell harvest. *Transfus Apheresis Sci*. 2020;59(2):102683. doi:10.1016/j.transci.2019.102683
16. Cottler-Fox MH, Lapidot T, Petit I, et al. Stem Cell Mobilization. *Hematology*. 2003;2003(1):419–437. doi:10.1182/asheducation-2003.1.419
17. Tanosaki R, Kumazawa T, Yoshida S, et al. A Novel Enumeration Method for Hematopoietic Progenitor Cells in Collecting Peripheral Blood Stem Cell for Transplantation. *Blood*. 2011;118(21):1922. doi:10.1182/blood.V118.21.1922.1922
18. Peerschke EI, Moung C, Pessin MS, Maslak P. Evaluation of new automated hematopoietic progenitor cell analysis in the clinical management of peripheral blood stem cell collections. *Transfusion*. 2015;55(8):2001–2009. doi:10.1111/trf.13078
19. Grommé M, Russcher H, Braakman E, et al. Multicentre study to evaluate a new enumeration method for hematopoietic stem cell collection management. *Transfusion*. 2017;57(8):1949–1955. doi:10.1111/trf.14183
20. Appelbaum FR. Hematopoietic-cell transplantation at 50. *N Engl J Med*. 2007;357(15):1472–1475. doi:10.1056/NEJMp078166

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