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Factors affecting lymphocyte collection efficiency for the manufacture of chimeric antigen receptor T cells in adults with B cell malignancies

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Abstract

BACKGROUND: The clinical and procedural parameters that affect the optimal collection of lymphocytes for the production of chimeric antigen receptor (CAR) T cells remain undefined but are increasingly important, as commercial products are now available. We evaluated determinants of low lymphocyte collection efficiency (CE) and the rate of successful CAR T cell manufacture in middle aged and older adults with advanced B-cell malignancies.

STUDY DESIGNS AND METHODS: Mononuclear cell collections using COBE Spectra and Spectra Optia platforms from patients participating in a CD19-directed CAR T cell therapy trial were reviewed. Patient and disease-specific factors, peripheral blood counts, apheresis parameters and product cell counts were analyzed to determine effects on lymphocyte CE.

RESULTS: Ninety-two apheresis events from patients with acute lymphocytic leukemia (ALL) (n=28), chronic lymphocytic leukemia (CLL) (n=18) and non-Hodgkin lymphoma (NHL) (n=46) were available for analysis. Forty-one collections (45%) had a lymphocyte CE of <40%. On multivariable analysis, age (every 10-year increase, OR=1.51, p=0.034), disease type (CLL vs. ALL, OR=0.24, p=0.052; NHL vs. ALL, OR=0.20, p=0.009) and pre-collection platelets (every $10 \times 10^3/\mu\text{L}$ increase, OR=1.07, p=0.005) were appreciably associated with a lymphocyte CE of <40%. No major apheresis complications occurred.

CONCLUSIONS: Lymphocyte collection at our center was well-tolerated and 100% successful in manufacturing CD19-directed CAR T cells from adult patients with B-cell malignancies despite low CE in some patients. A diagnosis of ALL, advancing age and higher pre-apheresis platelet counts were observed to be associated with low CE.

Keywords

Cellular therapy

INTRODUCTION

Immunotherapy using T cells genetically modified to express chimeric antigen receptor (CAR) is a rapidly emerging approach to treat various malignancies. Currently, there are two CAR T cell products approved by the U.S. Food and Drug Administration (FDA).

Axicabtagene ciloleucel (Yescarta) received FDA approval in October 2017 for the treatment of relapsed or refractory (R/R) large B-cell lymphoma.^{1,2} Tisagenlecleucel (Kymriah) was initially approved in August 2017 for patients up to 25 years of age with R/R precursor B-cell ALL and later approved in May 2018 for adult patients with R/R large B cell lymphoma.³⁻⁶ It is anticipated that the demand for lymphocyte products will increase as CAR T cell therapy becomes available at a broader scale, commercially or in the context of clinical trials.

CAR T cell manufacturing typically begins with the collection of nonstimulated peripheral blood lymphocytes through apheresis of mononuclear cells (MNCs), followed by selection and stimulation of T cells, and transduction of the CAR gene using viral vectors and expansion of the final product *in vitro*. While there has been extensive experience with hematopoietic progenitor cell collection for both autologous and allogeneic transplantation, the procedural and clinical factors affecting lymphocyte collection and ways to optimize the yield of T cells for the manufacturing CAR T cells are not fully known. This is particularly relevant given the wide range of patient ages and different diseases currently under study.

The optimal parameters for hematopoietic progenitor cell collection may not be applicable to MNC collections because non-mobilized patients often have low white blood cell counts making isolation of target lymphocytes challenging.⁷ MNC collections for CAR T cells more closely mimic collections for donor lymphocyte infusion (DLI) wherein mature lymphocytes are the target cells of apheresis.⁸ However, in comparison with healthy donors, patients referred for CAR T cell therapy are heavily pretreated and frequently have lymphopenia and/or high numbers of circulating malignant cells.

As CAR T cells become more widely utilized, understanding the parameters that affect lymphocyte collection will aid in obtaining sufficient cells for CAR T cell production. The objective of this study is to identify factors that influence the optimal collection of lymphocytes from predominantly middle age and older adults with advanced B cell malignancies for the manufacture of CAR T cells as measured by lymphocyte collection efficiency.

MATERIALS AND METHODS

Subjects

We reviewed consecutive MNC collections from patients with advanced B cell malignancies enrolled in a CD19 CAR T cell trial (#NCT01865617) at Fred Hutchinson Cancer Research

Center (FHCRC)/Seattle Cancer Care Alliance (SCCA). This trial was a single-center phase I/II study of autologous T cells engineered to express a CD19 CAR for adults with relapsed and/or treatment-refractory B cell malignancies. Inclusion for this retrospective study required consecutive collections with available product lymphocyte count data to calculate lymphocyte CE. The clinical trial was conducted in accordance with the U.S. Food and Drug Administration and the International Conference on Harmonization Guidelines for Good Clinical Practice, the Declaration of Helsinki and the Institutional Review Board (IRB). This retrospective chart review was approved by the IRB of FHCRC. This CD19-directed CAR T cell trial did not exclude patients based on their pre-collection peripheral blood absolute lymphocyte count, CD3 T cell count at screening, circulating tumor cells, or recipients of prior autologous or allogeneic hematopoietic cell transplant (HCT). Patients did not require a test for T cell expansion.

Apheresis

Prior to collection, patients were assessed by apheresis nurses for adequacy of their peripheral veins. If assessment of a patient's veins demonstrated a low likelihood of successful collection through peripheral venous access, then either a tunneled or temporary central venous catheter was placed before the procedure. Non-stimulated MNC collections were performed using the COBE Spectra and Spectra Optia (Terumo BCT, Lakewood, CO, USA). With collections on Spectra Optia, our apheresis unit used the mononuclear cell (MNC) collection protocol for a brief period of time before changing to the continuous mononuclear cell (CMNC) collection protocol. Among the 21 collections on the Spectra Optia platform, only one was conducted using the MNC protocol. The anticoagulant acid citrate dextrose A (ACDA) and heparin were used. Table 1 summarizes the anticoagulant concentration used, inlet flow rate and collect flow rate for each of the protocols used. Approximately 12 liters (median 11.6 L, range 11.1 to 17.4 L) of whole blood was processed. Two collections processed 17 L. Laboratory parameters required for collection before January 2018 included a hematocrit of 30% and a platelet concentration of 50,000/ μ L; those performed after January 2018 required a hematocrit of 28% and a platelet concentration of 20,000/ μ L. All patients only required a single day of collection. Before and after apheresis, a complete blood count (CBC) with automated differential was performed on peripheral blood using a hematology analyzer (XE-5000, Sysmex America Inc., Lincolnshire, IL, USA). Manual differential count was performed whenever blasts or circulating tumor cells were detected. After apheresis, a CBC was performed on the products using the Sysmex analyzer (XS-1000i, Sysmex America Inc., Lincolnshire, IL, USA) to determine the total nucleated cell counts. However, not all products had differential counts; hence, the lymphocyte content of the apheresis product was available in only 92 collections.

The lymphocyte collection efficiency (CE2) was calculated using the equation:

$$\text{Lymphocyte CE (\%)} = [(\text{product lymphocyte count} \times \text{product volume}) / (\text{process blood volume} \times \text{average peripheral blood lymphocyte count})] \times 100.$$

The average peripheral blood lymphocyte count was the pre-collection lymphocyte count + post-collection blood lymphocyte count $\div$ 2.

Manufacture of CD19-directed CAR T Cells at FHCRC

The MNC apheresis product underwent selection using immunomagnetic selection of CD4⁺ and CD8⁺ T cells. Patients had CAR T cells generated from either 1) bulk CD4-selected T cells and bulk-selected CD8 T cells, or 2) bulk CD4-selected T cells and selected CD8 central memory T cells. Preclinical data has demonstrated that CAR T cell products derived from defined T cell subsets have enhanced potency compared with products from unselected T cells that vary in phenotypic composition.⁹ Therefore, our center has utilized a 1:1 ratio of CD4:CD8 populations as this might provide reproducible clinical potency and facilitate more reliable correlations between cell dose and efficacy or toxicity.¹⁰ The selected CD4⁺ and CD8⁺ T cells were then separately lentivirally transduced to express the CD19 CAR and a truncated human epidermal growth factor receptor (EGFRt) that enables identification of transduced cells. The CAR T cells were then expanded and enriched. Equal numbers of the two expanded CD4⁺ and CD8⁺ cells were then combined for infusion to patients after they have received lymphodepleting chemotherapy.¹⁰

Statistical Analysis—Descriptive summary statistics included mean (standard deviation) and median (range) for continuous variables and number (percentage) for categorical variables. The threshold of <40% was determined a-priori to be the clinically meaningful outcome of lymphocyte CE based on our institutional criteria for minimum acceptable operator and instrument performance and a conservative estimate based on the published literature.^{11–15}

Potential predictors included the following: patient-specific factors such as age (every 10-year increase), gender (female vs. male), and weight (every 1 kg increase); disease-specific factors such as disease type (ALL vs. CLL vs. NHL), prior therapy (every 1 additional line of therapy), prior autologous hematopoietic cell transplant (HCT) and prior allogeneic HCT; pre-collection laboratory-specific factors such as, granulocyte (every $0.1 \times 10^3/\mu\text{L}$ increase), lymphocyte (every $0.1 \times 10^3/\mu\text{L}$ increase), monocyte (every $0.1 \times 10^3/\mu\text{L}$ increase), hematocrit (every 1% increase) and platelet count (every $10 \times 10^3/\mu\text{L}$ increase); and collection-specific factors such as, type of venous access (central vs. peripheral), instrument (Spectra Optia vs. COBE Spectra), product total nucleated cell count (every $1 \times 10^9/\mu\text{L}$ increase) and product hematocrit (every 1% increase). Product total nucleated cell count and hematocrit were included as surrogates of collection quality and cellular interface management during the procedure.

We first performed univariate logistic regression models to examine potential predictors of lymphocyte CE <40%. We then used the LASSO penalized regularization regression to minimize the misclassification errors via cross-validation in order to build a parsimonious multivariable prediction model.^{16,17} Robust standard error was used for all variance estimations. C statistic (area under the curve for the receiver operator curve) was used to assess the classification accuracy (discrimination). Hosmer-Lemeshow test was used to assess for goodness of fit testing of the model (calibration). All statistical analyses were performed using Stata 14.2.

RESULTS

Patient and collection characteristics

From June 4, 2013 to March 6, 2018, 197 MNC collections for protocol #NCT01865617 were performed at the SCCA/FHCRC from 189 patients. In this review, we only included 92 collections from 90 patients with available product lymphocyte counts that would allow for the calculation of lymphocyte CE. The distribution of diseases included 50% NHL (n=46), 30% ALL (n=28), and 20% CLL (n=18). Median patient age was 55 (range 20 to 70 years). Patient, disease and treatment characteristics are summarized in Table 2.

Pre-collection peripheral blood counts and apheresis procedural variables are summarized in Table 3. Twenty-seven (29%) apheresis events were from patients with an absolute lymphocyte count of $<0.5 \times 10^3/\mu\text{L}$ (range, $0.16\text{--}0.49 \times 10^3/\mu\text{L}$). Four (4%) of 92 collections had circulating blasts in the peripheral blood detected by CBC with manual differential in the range of 6%–80.0% of the total WBC ($0.19\text{--}20.73 \times 10^3/\mu\text{L}$). The majority (77%) of collections were performed using Spectra Optia via peripheral venous access (65%) (Table 4). No major adverse events occurred during apheresis, including bleeding, symptomatic hypocalcemia, hypotension or allergic reactions. Platelet clumping occurred within the centrifuge of two collections, resulting in early cessation of apheresis.

Collection yields, manufacturing, and CAR T cell infusion

The median MNC apheresis product volume was 226 ml (range 146 to 374 ml). Product counts are summarized in Table 5. Figures 1A and 1B demonstrate a positive correlation between pre-collection peripheral blood lymphocyte and monocyte counts with the product lymphocyte yield.

Only 89 of the 92 (97%) collections proceeded to CAR T cell manufacturing because three patients discontinued the study after collection. All 89 collections yielded adequate T cells for manufacturing of CAR T cells. Eighty-two of 89 MNC apheresis products (92%) that yielded manufactured CAR T cell products were infused. Among patients not infused, one decided to proceed with comfort measures only for their disease, two were subsequently found to be ineligible due to complications from prior therapy, and four did not have evidence of disease on re-evaluation.

Risk factors for low lymphocyte collection efficiency

The average lymphocyte CE of all collections was 43% with a median of 42% (range, 0.3–252%). Forty-one collections (45%) had a lymphocyte CE of $<40\%$. Table 5 compares the characteristics of collections with lymphocyte CE $<40\%$ vs. those $\geq 40\%$. On univariate analysis (Table 5), there was a trend towards reduced lymphocyte CE with higher pre-collection peripheral blood lymphocyte and monocyte counts, but these were not statistically significant upon multivariate analysis (Table 6). There was no clear correlation between pre-collection lymphocyte and MNC peripheral blood counts with lymphocyte CE (Fig. 1C and Fig. 1D). The fitted line of Spectra Optia collections plots above the COBE Spectra line, illustrating increased lymphocyte CE₂ with the Spectra Optia (Fig. 1C and Fig. 1D).

However, the type of instrument used (Spectra Optia vs. COBE Spectra) did not predict for lymphocyte CE in the univariate or multivariate analysis.

Predictors of low CE selected to form the multivariable model included age, disease, pre-collection platelet count, lymphocyte count, and monocyte count. The resulting model incorporating these predictors had a c statistic (area under the curve for the receiver operating curve) of 0.75 for discrimination. The calibration plot showed good fit with a Hosmer-Lemeshow p-value of 0.34. For inference testing, only three predictors – a diagnosis of ALL, advancing age and higher pre-apheresis platelet count – were appreciably associated with lymphocyte CE of <40% in the multivariable analysis (Table 6). A number of other variables such as gender, weight, pre-collection counts (hematocrit, lymphocytes, MNC and granulocytes), prior therapy, type of instrument used for collection, product counts (hematocrit and total nucleated cell count) were not associated with a low lymphocyte CE.

DISCUSSION

In our study of 92 adult patients with advanced B cell malignancy undergoing apheresis for CAR T cells, we derived various patient-specific independent predictors – age, disease, and pre-collection platelet for low CE (<40%). The resulting multivariable model had moderate discriminative power (c statistic of 0.75) and good calibration. If validated externally, these predictors can help apheresis physicians identify patients who may not be able to meet the collection goal for protocols requiring higher product lymphocyte yields. Furthermore, we discovered that the lymphocyte CE was not appreciably affected by the type of instruments used or the extreme ranges of the pre-collection lymphocytes.

Despite a relatively low (<40%) lymphocyte CE in 45% of collections, CAR T cell manufacture was successful in 100% of patients with intention to proceed with CAR T cell infusion, including those who were heavily pretreated and had low absolute lymphocyte counts at the time of apheresis. The apheresis procedure was safe and well tolerated without any major adverse events, such as bleeding, hypotension, allergic reactions or symptomatic hypocalcemia. Two prior studies of lymphapheresis for CAR T cell production reported a complication rate of 9.8–15%, the majority of which are minor and manageable, such as paresthesia, pain, nausea, vomiting and headache.^{8,18}

Allen and coworkers reported their experience at the National Institutes of Health (NIH) with apheresis for CAR T manufacturing in pediatric and young adult patients ages 4 to 30 years with ALL and solid tumors.⁵ Of the 71 collections, two (3%) yielded fewer than the minimum of 0.6×10^9 CD3⁺ cells requested for manufacturing. An additional 14 collections (20%) achieved the minimum, but not the target of 2×10^9 CD3⁺ cells. The COBE Spectra instrument was used for these collections. The average CE2 values for CD3⁺ lymphocytes were reported as 59% and 68% for those products that were below and above the target yield of 2×10^9 CD3⁺ cells, respectively.⁵ A higher proportion of circulating blasts and NK cells, and lower lymphocyte counts were associated with lower CD3⁺ yields. Only 63 collections (88%) were successful in generating sufficient quantities of CAR T cells to meet protocol dose criteria (0.3 to 3.0×10^6 transduced CD3⁺ cells). Lower CD3⁺ yields were significantly

associated with manufacturing failures, which occurred in five of 16 patients (31%) with fewer than the target number of CD3⁺ cells, and three of 55 patients (5%) who met the target.⁸

Ceppi and colleagues recently published their experience with CAR T cell collection and processing at the Seattle Children's Hospital and Research Institute among 99 heavily pretreated pediatric and young adult patients (ages 1.3–25.7 years) with ALL, NHL, and neuroblastoma.¹⁰ This study demonstrated a 100% success rate in achieving the total MNC collection target of 1×10^9 cells/kilogram. This study also exclusively used the COBE Spectra instrument for collections. The average lymphocyte CE2 values for their collections was 83.4% and there was no significant difference between products collected from children with ALL (86.7%) compared to products from children with neuroblastoma (68.8%). Only 96 (94%) of these products yielded adequate CAR T cells. Among the six insufficient CAR T products, one was canceled due to patient death and two had inadequate CD8 cell growth attributed to recent therapy with metaiodobenzylguanine (MIBG) for neuroblastoma. One product was unacceptable due to infection and two had poor expansion; all three patients subsequently underwent repeat collection and then were able to generate acceptable CAR T products.¹⁸

Compared to the studies of Allen et al⁸ and Ceppi et al¹⁸, our study had a much lower average lymphocyte CE of 43%. Notably, the patients in both other studies were much younger (median ages 14.9⁸ and 11.6¹⁸ years, respectively) compared to the much older patients in our study (median age 55 years). However, our observed lymphocyte CE closely approximates that of the reported lymphocyte CE2 of $37.2 \pm 18\%$ in twelve MNC collections performed on healthy non-mobilized adult blood donors (median age 29 years; range 21 to 50 years).¹⁹ Our analysis revealed that advancing age independently predicted for a lymphocyte CE of <40%. This may have accounted for the lower lymphocyte CE in our study compared to studies that included pediatric patients. Furthermore, our study did not demonstrate any difference in lymphocyte CE between the type of instrument used (Spectra Optia vs. COBE Spectra). This is in contrast to a retrospective study of 26 collections from pediatric patients 1.7 to 18 years-old that showed a higher mean MNC CE2 from Spectra Optia vs. COBE Spectra collections.¹⁵

In addition to age, we identified a higher precollection platelet count and a diagnosis of ALL as risk factors for a lymphocyte CE of <40%. We hypothesize that a higher platelet count may have resulted in occult platelet clumping within the centrifuge that may have occurred below the trigger sensitivity of the apheresis machine. Furthermore, the presence of large blasts associated with ALL, and high platelet counts may have caused narrowing of the MNC interface where the mature lymphocytes lie. Further studies are needed to elucidate the mechanisms that result in a low lymphocyte CE among patients with these risk factors to optimize their lymphocyte yield, which may include modification of the default apheresis settings.

Our study has its limitations. First, our analysis was limited 92 out of 197 apheresis products because product lymphocyte counts were not routinely checked in all cases (not required by protocol). While this significantly reduced the number of study participants, the exclusion of

patients was by random chance without a systematic selection bias. Second, this study was limited to COBE Spectra and Spectra Optia instruments. Third, despite moderate discriminative power and good fit, our derived model would require external validation to ensure generalizability.

In conclusion, apheresis at our center was successful in obtaining a satisfactory product to generate CAR T cells despite frequently low lymphocyte CE, based in large part to our unique manufacturing process. Advancing age, diagnosis of ALL and higher platelet counts prior to apheresis predict for low lymphocyte CE. Further studies are needed to validate these predictors, and to inform interventions that would improve the lymphocyte CE among at-risk patients.

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Figure 1.
(A & B) Precollection lymphocyte and mononuclear cell (MNC) counts versus product lymphocyte counts demonstrating a positive correlation on both the COBE Spectra and Spectra Optia platforms. (C & D) Precollection lymphocyte and MNC counts versus calculated lymphocyte efficiency (CE2) without clear correlation.

Table 1.

Anticoagulant concentration, inlet and collect flow rates used

	COBE Spectra[*]	Spectra Optia MNC[*]	Spectra Optia CMNC[*]
Anticoagulant: Heparin(units) /ACDA (ml)	10 units/1 ml	5 units/1 ml	5 units/1 ml
Inlet/AC Ratio	15:1 – 35:1	Ramping recommended start 8:1; ramp no greater than 20:1	8:1 – 30:1
Inlet Flow Rate (ml/min) maximum flow	100	125	100
Collect Flow Rate (ml/min)	1.5	Not applicable	1 to 2.4
Chamber Flush	N/A	Default 16 ml	
Chamber Chase	N/A	Recommended 4 ml	Not applicable

MNC = mononuclear cell collection; CMNC = continuous mononuclear cell collection

^{*} In this study, COBE Spectra, Spectra Optia MNC and Spectra Optia CMNC were used in 71, 1 and 20 of the collections, respectively.

Per Terumo CMNC Procedure Guidelines for Optimizing the Collect Pump Flow Rate

Table 2.

Patient, disease and treatment characteristics

	Diagnosis			
	All collections <i>N</i> = 92	ALL <i>n</i> = 28	CLL <i>n</i> = 18	NHL <i>n</i> = 46
Age, years				
Mean	52	42	61	55
Median	55	39	62	57.5
Range	20–73	20–73	42–73	28–70
Gender, n (%)				
Male	58 (63)	15 (54)	12 (67)	31 (67)
Female	34 (37)	13 (46)	6 (33)	15 (33)
Weight, kg				
Mean	80	76.9	86.5	79.9
Median	79.1	76	82.8	80.2
Range	47.8–148	47.8–128	64.1–119.1	50.2–148
Prior lines of therapy, no.				
Mean	4	3	4	4
Median	4 1–8	3	4	4
Range		1–7	2–8	1–8
Prior autologous HCT, n (%)	19 (21)	0	1 (6)	18 (39)
Prior allogeneic HCT, n (%)	20 (22)	7 (25)	6 (33)	7 (15)

HCT = hematopoietic cell transplant

Table 3.

Precollection laboratory and apheresis procedural parameters

	Diagnosis			
	All collections <i>N</i> = 92	ALL <i>n</i> = 28	CLL <i>n</i> = 18	NHL <i>n</i> = 46
Precollection blood counts, median (range)				
Hematocrit, %	33 (24–46)	31.5 (25–41)	36 (24–46)	33 (26–46)
WBC, $\times 10^3/\mu\text{L}$	4 (0.45–35.6)	3.3 (0.79–20.5)	4.2 (0.5–26)	4.6 (1.7–35.6)
Lymphocytes, $\times 10^3/\mu\text{L}$	0.8 (0.16–23.1)	0.7 (0.16–2.2)	1.1 (0.4–23.1)	0.8 (0.2–12.5)
Neutrophils, $\times 10^3/\mu\text{L}$	2.5 (0.01–14.8)	2 (0.06–8.7)	1.5 (0.01–6)	3 (1–14.9)
Monocytes, $\times 10^3/\mu\text{L}$	0.4 (0–3.1)	0.2 (0.01–1.1)	0.4 (0–1.2)	0.5 (0–3.1)
Platelets, $\times 10^3/\mu\text{L}$	128 (13–668)	113 (24–458)	114 (13–435)	141 (20–668)
<hr/>				
Blasts, %	0 (6–80)	0 (0–20)	0	0 (6–80)
Venous access, n (%)	60 (65)	16 (57)	14 (78)	30 (65)
Peripheral veins	32 (35)	12 (43)	4 (22)	16 (35)
Central				
<hr/>				
Instrument, n (%)				
COBE Spectra	71 (77)	24 (86)	13 (72)	34 (74)
Spectra Optia	21 (23)	4 (14)	5 (28)	12 (26)

WBC = white blood cell count

Table 4.

Apheresis product counts

	Diagnosis			
	All collection <i>N</i> = 92	ALL <i>n</i> = 28	CLL <i>n</i> = 18	NHL <i>n</i> = 46
Absolute TNC count, $\times 10^9/\mu\text{L}$				
Mean	18	8	24	21.8
Median	10.5	7.3	17	11.3
Range	0.12–199	0.12–19.5	0.7–133	1.25–199
Absolute lymphocyte count, $\times 10^3/\mu\text{L}$				
Mean	33.7	18.5	50.7	36.2
Median	21.1	15.7	37.7	21.2
Range	0.29–287.1	0.29–62.6	0.7–184.8	2.6–287.1
Hematocrit, %				
Mean	1.7	1.7	1.8	1.6
Median	1.6	1.7	1.6	1.6
Range	0.1–3.7	0.6–3.5	0.7–3.6	0.1–3.7
Absolute platelet count, $\times 10^3/\mu\text{L}$				
Mean	1223	875	1476	1326
Median	1078	645	1427	1221
Range	9–5851	141–2065	143–5032	9–5851

TNC = total nucleated cell

Table 5.

Univariate analysis for predictors of lymphocyte collection efficiency less than 40%

Parameter	Less than 40% <i>n</i> = 41	40% or greater <i>n</i> = 51	Odds ratio	P-value
Age, years				
Median (range)	58 (24–73)	53 (20–73)	1.21	0.234
Gender, n (%)				
Male	26 (63)	32 (63)	1	.
Female	15 (37)	19 (37)	1.09	0.837
Weight, kg				
Median (range)	79.2 (50.2–128)	79 (47.8–148)	1.01	0.537
Disease type, n (%)				
ALL	16 (39)	12 (23)	1	.
CLL	8 (20)	10 (20)	0.56	0.348
NHL	17 (41)	29 (57)	0.43	0.083
Prior therapy				
Lines of therapy, median (range)	3 (1–8)	4 (1–8)	0.89	0.322
Prior autologous HCT, n (%)	7 (17)	12 (23)	0.83	0.729
Prior allogeneic HCT, n (%)	9 (22)	11 (22)	0.97	0.948
Pre-collection CBC, median (range)				
Hematocrit, %	32 (25–46)	34 (24–46)	0.98	0.622
Neutrophils, $\times 10^3/\mu\text{L}$	2.5 (0.6–14.8)	2.6 (0.01–12.3)	1.01	0.264
Lymphocytes, $\times 10^3/\mu\text{L}$	0.9 (0.2–23.1)	0.8 (0.2–4.1)	1.02	0.076
Monocytes, $\times 10^3/\mu\text{L}$	0.6 (0–3.1)	0.4 (0–1)	1.12	0.043
Platelets, $\times 10^3/\mu\text{L}$	167 (28–668)	116 (13–262)	1.06	0.007
Venous access, n (%)				
Peripheral veins	26 (63)	34 (33)	1	.
Central	15 (37)	17 (67)	1.08	0.864
Instrument used, n (%)				
COBE Spectra	32 (78)	39 (76)	1	.
Spectra Optia	9 (22)	12 (24)	0.86	0.771
Product counts, median (range)				
TNC, $\times 10^9/\mu\text{L}$	8.3 (0.1–199)	13.4 (3–125.9)	1.00	0.861
Hematocrit, %	1.6 (0.1–3.6)	1.5 (0.7–3.7)	0.92	0.786

ALL = acute lymphocytic leukemia; CLL = chronic lymphocytic leukemia; NHL = non-Hodgkin lymphoma; HCT = hematopoietic cell transplant; TNC = total nucleated count

Table 6.

Multivariable analysis for predictors of lymphocyte collection efficiency less than 40%

	Odds ratio	P-value
Patient factors		
Age (every 10-year increase)	1.51	0.044
Disease factors		
Disease type (CLL vs. ALL)	0.16	0.022
Disease type (NHL vs. ALL)	0.17	0.006
Laboratory factors		
Pre-collection platelet count (every $10 \times 10^3/\mu\text{L}$ increase)	1.07	0.022
Pre-collection lymphocyte count (every $0.1 \times 10^3/\mu\text{L}$ increase)	1.02	0.107
Pre-collection monocyte count (every $0.1 \times 10^3/\mu\text{L}$ increase)	1.06	0.388

ALL = acute lymphocytic leukemia; CLL = chronic lymphocytic leukemia; NHL = non-Hodgkin lymphoma