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The effect of repeated stimulated granulocyte donations on hematopoietic indexes in donors: a 24-year donor center experience

James Szymanski¹, James Troendle², Susan Leitman¹, Hong Hong¹, Yu Ying Yau¹, Cathy Cantilena¹

¹Department of Transfusion Medicine, Clinical Center, National Institutes of Health, Bethesda, Maryland ²Office of Biostatistics Research, National Heart, Lung, and Blood Institute, Bethesda, Maryland

Abstract

BACKGROUND—Short- and long-term effects of mobilization regimens in hematopoietic stem cell and granulocyte donors have been well characterized. In this study, we examined the longitudinal hematopoietic changes related to repeat stimulated granulocyte donation.

STUDY DESIGN AND METHODS—Complete blood counts for consecutive granulocyte donors between October 1994 and May 2017 were compared to unstimulated granulocyte donors. Platelethpheresis donors served as controls. The longitudinal change in precollection white blood cell (WBC) counts for these donor groups were modeled using a linear mixed-effects model. The investigated variables were granulocyte, lymphocyte, and monocyte counts and the granulocyte collection yield. Contrasts were performed to explore the effect of donation number on precollection counts.

RESULTS—For the granulocyte-colony-stimulating factor plus dexamethasone (G-CSF/Dex)-stimulated group, both the granulocyte and the lymphocyte counts decreased $6.51 \times 10^9/L$ (-23.1% , $p < 0.001$) and $0.21 \times 10^9/L$ (-20.4% , $p < 0.001$), respectively, between Donation 1 and Donation 20. This effect was still present at the 3- to 4-year interval ($b = -0.0008313$, $SE = 0.00029$, $p = 0.004$). For the unstimulated donor group between Donation 1 and Donation 20, the lymphocyte count decreased by $0.62 \times 10^9/L$ (-51.5% , $p < 0.001$). This effect was only significant up to Year 2 ($b = -0.0026$, $SE = 0.0010$, $p = 0.013$).

CONCLUSIONS—Past granulocyte donations were found to have a statistically strong negative effect on precollection granulocyte counts and lymphocyte counts and decreased granulocyte yield

Address reprint requests to: James Szymanski, Transfusion Medicine Fellow, Office of the Chief, Department of Transfusion Medicine, National Institutes of Health, Building 10, Room 1C-711, Bethesda, MD 20854; jszymans@montefiore.org.

CONFLICTS OF INTEREST

The authors have disclosed no conflicts of interest.

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SUPPORTING INFORMATION

Additional Supporting Information may be found in the online version of this article.

both in the G-CSF/Dex—stimulated donors and the unstimulated donors. In this statistical model, for both these groups, the effect of past donations on granulocyte and WBC counts were still detectable 2 years later.

In patients with neutropenia, bacterial and fungal infections remain significant sources of morbidity and mortality, despite modern antimicrobial treatment. Neutropenia is a well-established complication of chemotherapy, liquid transplant, and other hematologic conditions. While neutropenic, the risk of infection and mortality increases with the degree and duration of the episode.¹ Granulocyte transfusion has been used for decades as a treatment modality in both patients with neutropenia and those with granulocyte dysfunction who have infections refractory to antibiotics.^{2–4}

The effectiveness of granulocyte transfusions is highly dependent on transfusing an appropriately high dose of granulocytes.² The RING trial, a multicenter randomized controlled study, found that subjects who received an average dose per transfusion of at least 0.6×10^9 granulocytes per kilogram tended to have better outcomes than those receiving a lower dose.⁵ Collection of this dose from healthy donors became possible with the use of granulocyte–colony-stimulating factor (G-CSF) in stimulated apheresis collections.⁶ It was found that using dexamethasone plus G-CSF (G-CSF/Dex), as stimulatory agents, resulted in greater granulocyte yields than either agent alone while causing no increase in donor toxicity.⁷

The short-term adverse effects of G-CSF have been well characterized. In a prospective study of 2408 healthy unrelated peripheral blood stem cell (PBSC) donors who received G-CSF, almost all experienced bone pain, while 70% of donors reported some degree of myalgia, headache, or fatigue, with smaller percentages reporting insomnia, nausea, or fever. Almost all symptoms disappeared after 1 week after donation.^{3,8} Rare side effects of using G-CSF in donors, including splenic rupture, pulmonary hemorrhage, and anaphylactic shock, have also been noted.⁹ In a follow-up study of 6768 National Marrow Donor Program donors who received G-CSF and underwent PBSC donation between 2004 and 2009, there was no evidence of increased donor risk for cancer, stroke, or autoimmune illness.¹⁰

Similarly, the long-term severe adverse effects of repeat stimulated granulocyte donation have been investigated. In a 10-year follow-up study of healthy unrelated granulocyte donors at the National Institutes of Health, 83 subjects who had donated 1120 granulocyte concentrates (mean 13.5 granulocyte donations per donor) were compared with a matched group of platelet (PLT) donors. No difference in the incidence of coronary heart disease, stroke, thrombosis, or malignancy was seen among these two groups.¹¹

It has been previously noted that there are sustained decreases in PLT count with regular plateletpheresis.¹² In serial long-term leukapheresis donors, sustained lymphocyte count decreases with increasing donation number have been reported.¹³ However, the impacts of age, interdonation interval, race, sex, and other concurrent donations were not examined in these studies. In light of the mantra “first do no harm,” the purpose of this study was to adjust and control for these factors and to investigate whether there are longitudinal hematopoietic changes related to repeat stimulated granulocyte donation.

MATERIALS AND METHODS

Study subjects

We retrospectively analyzed the longitudinal changes in the precollection complete blood counts (CBCs) of 493 healthy volunteer allogeneic granulocyte donors who underwent a total of 2543 G-CSF/Dex-stimulated granulocyte donations starting in October 1994 and compared the change in their precollection white blood cell (WBC) counts over time to that of 526 healthy unstimulated granulocyte donors (1391 total donations) and 3743 PLT donors (59,585 total PLT donations; Table 1).

All allogeneic granulocyte donors underwent a thorough history and physical examination with baseline laboratory evaluation and were consented and enrolled in institutional review board-approved protocols before undergoing granulocytapheresis and/or being given G-CSF/Dex. In addition to meeting donor eligibility criteria for allogeneic blood donors, they were excluded for stimulation agent-related risks including uncontrolled hypertension, peptic ulcer disease, and posterior subcapsular cataracts. Donors without a current eye examination or with a history of posterior subcapsular cataracts were not given dexamethasone but were restricted to granulocytapheresis using G-CSF alone. The G-CSF alone-stimulated donors were included in the G-CSF/Dex donor group for purposes of this analysis. Both stimulated and unstimulated granulocyte donors were restricted to a maximum of one granulocytapheresis donation per month.

Unstimulated granulocyte and allogeneic PLT donors met all the eligibility criteria for volunteer whole blood donation. Healthy granulocytapheresis-unstimulated granulocyte donors were exempt from restrictions on foreign travel, use of prescription drugs that would disqualify them from allogeneic donation, and anti-HBc antibody presence. CBCs were performed immediately before every granulocyte and PLT donation. Informed consent was obtained in accordance with the Helsinki Declaration, and our institutional review board approved allogeneic stimulated granulocyte and unstimulated granulocyte apheresis donor protocols.

There was significant donor crossover between groups. Thirty-three percent of the G-CSF/Dex donor group had donated, at least once, using dexamethasone alone. In contrast, the crossover between the G-CSF/Dex group and unstimulated donor group was much lower at 9.8%. All stimulated granulocyte donors had been prior plateletpheresis donors.

Granulocyte stimulation and collection

For the stimulated granulocyte donations before July 2005, donors received 5 µg/kg G-CSF as a single subcutaneous injection 12 to 24 hours before donation and/or 8 mg dexamethasone orally 12 hours before donation. After July 2005, the G-CSF dose was changed to a standard 480 µg subcutaneous injection for all donors. The dexamethasone dose remained unchanged. G-CSF/Dex has been used for granulocyte donor stimulation at our institution since 1994. Stimulated donor granulocytes were collected for use as therapeutic transfusion components, at the request of and consultation with the primary treating physician, for patients with severe neutropenia or disorders of neutrophil function and life-threatening infections unresponsive to antimicrobial agents.

Granulocyte collection procedures were performed using continuous flow centrifugation on an apheresis device (Spectra, Terumo BCT; or CS-3000, Fenwal). Seven to 7.5 liters of whole blood was processed per procedure, at a blood flow rate of 50 to 70 mL/min. A quantity of 500 mL of hydroxyethyl starch solution (Hespan, B. Braun Medical) was used as a red cell sedimenting solution and injected with 30 mL of citrate (Tricitrinol, Cytosol Laboratories) for anticoagulation of the extracorporeal circuit.

Statistical analysis

Data modeling was performed starting with a database of allogeneic granulocyte donors, unstimulated granulocyte donors, and PLT donors, which was compiled retrospectively using a computer spreadsheet (Excel 2013, Microsoft Corporation). For each donation, the number of previous donations for each category (G-CSF+/-Dex, dexamethasone only, PLTs, and unstimulated granulocytes) were tabulated for 0 to 1, 1 to 2, 2 to 3, 3 to 4, and 4+ years ago. The CBC indexes investigated were granulocyte, lymphocyte, and monocyte counts and granulocyte collection yield (granulocyte count in the bag per liter of whole blood processed). To reduce outcome variable skewness, natural log transformations were carried out. For each investigated CBC index, a linear mixed-effect model was built that included as fixed effects donation category, age, race, sex, and the number of previous donations for each category and each time period as previously described. Donors were specified as random effects to control for the intrasubject correlation of repeat donations and crossover between groups. In contrast to multiple linear regression, linear mixed-effects models allow for controlling the variance associated with random factors without data aggregation.¹⁴ The models were built using the computer software (nlme package, R Version 3.3.1) and utilized the computational resources of the high-performance computing Biowulf cluster.¹⁵

To characterize general model fit, pseudo- R^2 was calculated using the R package piecewiseSEM. The conditional pseudo- R^2 describes the proportion of data variance that can be explained by both fixed and random factors.¹⁶ The linear mixed-effects model for precollection granulocyte counts had a conditional pseudo- R^2 of 0.729. The precollection lymphocyte model conditional pseudo- R^2 was 0.742, for precollection monocyte it was 0.528, and for granulocyte yield per liter it was 0.871. Visual inspection of model residual plots did not reveal any obvious deviations from homoscedasticity or normality.

A strong intrasubject correlation of precollection WBC counts for repeated donations was found. The intraclass correlation coefficient is best interpreted, in the case of this model, as the proportion of total variance that is accounted for by the clustering of intrasubject counts. For the granulocyte model the intraclass correlation coefficient was found to be 0.56. For the lymphocyte model it was 0.66 and 0.36 for the granulocyte yield per liter model.

To better illustrate the effect of donation number on WBC count, statistical model contrasts were performed with differing collection numbers spread out among the yearly collection intervals in a balanced manner (Appendix S1, Figure 4, Contrast Model Selection). A 60-year-old donor was set as the baseline to place emphasis on any age-related effects though the mean age of donors was younger. Male sex was used to coincide with our center's majority of granulocyte donors. The five-granulocyte-donations model contrast assumes one stimulated donation for each year of the past 5 years. This assumption was used because, on

average, stimulated granulocyte donors donate $1.3 \times$ /year. The 10-collection-model contrast assumes two stimulated collections for each of the past 5 years. Likewise, the 15-collection model assumes three stimulated collections per year, and the 20-collection model assumes four. The interdonation periods for the contrast model were significantly longer than the Food and Drug Administration established donation deferral periods (Appendix S1 [available as supporting information in the online version of this paper], Fig. 4, contrast model selection).

RESULTS

Precollection granulocyte counts

Significant differences were found in precollection granulocyte counts between the stimulated donor and PLT donor groups and between the individual stimulated donor groups (Fig. 1). The overall G-CSF/Dex-stimulated granulocyte donor group has a mean precollection granulocyte count of $29.2 \times 10^9/\text{L}$, which is $23.5 \times 10^9/\text{L}$ higher ($p < 0.0001$) than the dexamethasone alone-stimulated donors ($5.7 \times 10^9/\text{L}$). The overall dexamethasone-alone donors' predonation granulocyte counts had a marginal mean that was $2.407 \times 10^9/\text{L}$ higher ($p < 0.0001$) than the unstimulated granulocyte donors ($3.3 \times 10^9/\text{L}$). The effect of age was not found to have a significant effect of precollection granulocyte counts. African American race was associated with a decreased precollection granulocyte count in aggregate of $5.24 \times 10^9/\text{L}$ versus Caucasian $6.04 \times 10^9/\text{L}$ ($p < 0.0001$). This held true for stimulated granulocyte, unstimulated granulocyte, and PLT donors. For African Americans undergoing unstimulated granulocyte collections the granulocyte count was $0.293 \times 10^9/\text{L}$ below the all-group mean ($3.26 \times 10^9/\text{L}$, $p < 0.001$), while for G-CSF/Dex collections it was 4.66 below the all-group mean ($29.2 \times 10^9/\text{L}$, $p < 0.001$).

The effect of increased donation number was strongly associated with subsequent decreased precollection granulocyte counts in all donor groups with the exception of the PLT donors ("control" group). The precollection granulocyte count contrast model is shown in Fig. 1. Between granulocyte Donation 1 and Donation 5 the G-CSF/Dex donors group experienced a precollection granulocyte adjusted mean decrease of 4.4% or $1.26 \times 10^9/\text{L}$ ($p < 0.001$). Between Collection 5 and Collection 10 increases, there was a decrement of 7.1% or $1.91 \times 10^9/\text{L}$ ($p < 0.001$).

The unstimulated granulocyte donor group had a decrease precollection granulocyte count of $0.08 \times 10^9/\text{L}$ ($p = 0.011$) between Collection 1 and Collection 5 (-2.7%). Likewise, the dexamethasone-alone donor group decreased $0.11 \times 10^9/\text{L}$ ($p = 0.018$; -2%). The rate of precollection granulocyte count change increased between Collection 5 and Collection 10 for these groups to 5.3 and 6%, respectively. The PLT group precollection granulocyte count had an insignificant increase of 0.6% between Collection 1 and Collection 20 (Appendix S1, Table 3, precollection granulocyte contrast table).

The effect of previous granulocyte donations in the G-CSF/Dex donor group did not completely wane over a 1-month deferral period. Sequential stimulated allogeneic granulocyte donations in the same year had a strong negative effect on precollection granulocyte count ($b = -0.0026$, $SE = 0.00027$, $p < 0.0001$). The significant effect of past G-

CSF/Dex donations carried into the 1- to 2-year interval ($b = -0.0016$, $SE = 0.00027$, $p < 0.0001$), 2- to 3-year interval ($b = -0.0013$, $SE = 0.00029$, $p < 0.0001$), and 3- to 4-year interval ($b = -0.0008313$, $SE = 0.00029$, $p = 0.004$). Prior donations, likewise, had a strong negative effect on current counts for the unstimulated granulocyte donor group, although this effect only was significant up to Year 2 ($b = -0.0026$, $SE = 0.0010$, $p = 0.013$). The same was found for the dexamethasone-alone donor group ($b = -0.0027$, $SE = 0.0007$, $p = 0.0001$). The effect of prior collections greater than 2 years ago for the unstimulated granulocyte, dexamethasone-alone, and PLT donor groups did not reach the level of significance ($p > 0.05$; Appendix S1, Table 7, granulocyte linear mixed-effects model).

Precollection lymphocyte counts

The precollection lymphocyte counts were found to be significantly different between donation groups (Fig. 2). In aggregate, the PLT donors group mean lymphocyte count was $1.63 \times 10^9/L$ (vs. G-CSF/Dex donor group, $1.01 \times 10^9/L$, $p < 0.0001$). The dexamethasone-only donor group mean ($0.73 \times 10^9/L$) was $0.899 \times 10^9/L$ ($p < 0.001$) lower than that for the control PLT group. There was no significant effect due to race after controlling for age, sex, and prior collections, although age had a significant negative effect on lymphocyte count for the G-CSF/Dex donor group ($b = -0.0009$, $p < 0.001$) and the unstimulated granulocyte donor group ($b = -0.00463$, $P < 0.001$).

Between Collection 1 and Collection 10 the G-CSF/Dex group's adjusted mean dropped $0.094 \times 10^9/L$ ($p < 0.001$), a 9.3% decrease. Over this same interval, the unstimulated granulocyte donor group had a larger decrement with an adjusted mean decrease of $0.42 \times 10^9/L$ ($p < 0.001$). Between Collection 1 and Collection 15, the PLT control decreased 1.7% and the dexamethasone-alone donor group decreased 6.9%. The rate of count change increased for the G-CSF/Dex group with increasing collection number. Between Collection 10 and Collection 20, the adjusted mean decreased $0.116 \times 10^9/L$ ($p < 0.001$; Appendix S1, Table 4, precollection lymphocyte contrast table).

Past donations were found to have a strong negative effect on current lymphocyte counts in the unstimulated granulocyte donor groups and, to a lesser degree, in the G-CSF/Dex donor group. This effect, in the unstimulated granulocyte donor group, waned little with time from the 0- to 1-year interval ($b = -0.0303$, $SE = 0.0032$, $p < 0.001$) to the Year 2 to Year 3 interval ($b = -0.021$, $SE = 0.0036$, $p < 0.001$). Between these time points the beta-coefficient remained negative and significant. This effect, in the G-CSF/Dex donor group, largely weakened with time from the 0- to 1-year interval ($b = -0.0144$, $SE = 0.000856$, $p < 0.001$) to the Year 2 to Year 3 interval ($b = -0.0028$, $SE = 0.0009$, $p = 0.0021$). For both groups the beta-coefficient remained significant into Year 4 (Appendix S1, Table 8, lymphocyte linear mixed-effects model).

Precollection monocyte counts

The precollection monocyte counts significantly differed between groups. In aggregate, the G-CSF/Dex donors' group had the largest mean of $0.76 \times 10^9/L$, which was 0.3 greater ($p < 0.001$) than the PLT donors' group. The dexamethasone-alone mean of $0.165 \times 10^9/L$ was $0.267 \times 10^9/L$ below ($p < 0.001$) that of the unstimulated granulocyte donors' group (0.432

$\times 10^9/L$). Increased donation number effected little change in precollection monocyte counts. Between Collection 1 and Collection 5, there was no significant change in the G-CSF/Dex group ($p = 0.29$) and PLT group ($p = 0.20$). The largest absolute monocyte count effect was small and seen in the unstimulated granulocyte donor group, which decreased from a mean of $0.457 \times 10^9/L$ on Collection 1 to $0.389 \times 10^9/L$ by Donation 15 ($P < 0.001$; Appendix S1, Table 5, precollection monocyte contrast table).

Granulocyte yield (granulocytes per liter of whole blood processed)

Increased donation number was associated with decreased granulocyte bag yield per liter processed in the G-CSF/Dex group and the unstimulated granulocyte donor group. The dexamethasone-alone donor group's granulocyte yield per liter did not change significantly between Collection 1 and Collection 15 ($P = 0.83$). Between Collection 1 and Collection 10, the G-CSF/Dex donor group gran bag yield decreased 0.041×10^{10} granulocytes/L (4.3%; $p < 0.001$). The unstimulated granulocyte donor group had a yield decreased (0.042×10^{10} granulocytes/L, $p < 0.001$) over this same collection interval (Appendix S1, Table 6, granulocyte yield contrast table). Age had a significant negative effect on the granulocyte yield per liter for the G-CSF/Dex donor group ($b = -0.001251$, $p < 0.001$) and a paradoxically slight positive effect in the unstimulated granulocyte donor group ($b = 0.0008$, $p = 0.02$). Race did not have a significant effect on granulocyte bag yield per liter (Appendix S1, Table 10, granulocyte yield per liter linear mixed-effects model).

DISCUSSION

This study is the first to describe that in healthy donors, increased number of granulocyte collections is associated with decreased precollection granulocyte counts, lymphocyte counts, and granulocyte yield per liter using careful statistical modeling. Prior studies did not control for the impacts of age, interdonation interval, race, sex, and other concurrent donations. Controlling for these variables, the decrease in WBC counts was still evident in both the stimulated and the unstimulated granulocyte donation groups. No such change was seen in the PLT control group.

The negative effect on predonation granulocyte and lymphocyte counts, from past donations, did not quickly diminish with time in the G-CSF/Dex-stimulated and unstimulated granulocyte donor groups. For both, this effect was found to be most prominent during the first year. The negative beta-coefficient, however, still reached the level of significance at 4 years postdonation in the lymphocyte and granulocyte models. This phenomenon has been previously documented in other studies (Table 2); however, to our knowledge this is the first description of such seen in unstimulated granulocyte donors.

These findings suggest that the granulocytapheresis procedure itself may play a role in the count decrements seen. Prior studies hypothesized that G-CSF was responsible, either through G-CSF-receptor down regulation or depletion of slowreplacing PBSCs.¹⁷ Leukapheresis has been estimated to only remove approximately 10% of PBMCs.¹⁸ However, without G-CSF mobilization, unstimulated granulocyte donors experience minimal PBSC loss, while maintaining marrow reserves.

Hetastarch might play a role in this effect. Studies have shown that it accumulates in marrow and organs of the reticuloendothelial system.¹⁹ In a murine model, hetastarch reduced the inflammatory response by decreasing neutrophil recruitment, hindering PLT-neutrophil interactions, and reducing neutrophil extracellular trap formation.²⁰ Since PBSC donors are not exposed to Hetastarch, the similar hematopoietic count decrements this group experiences are not mediated by such a mechanism (Table 2).

There is no current evidence that these decreases in WBC counts from baseline have clinical consequences for donors. In a 2009 study of 3928 PBSC collections using G-CSF mobilization, no increased incidence of infection was found during the 5-year follow-up period.¹⁷ However, the vast majority of this study's donors underwent one round of mobilization and collection. Thus, these findings may not be generalizable to multiply stimulated and collected granulocyte donors. In our study we found that the effect of increased donation number was strongly associated with subsequent decreased precollection granulocyte counts in all donor groups with the exception of the PLT donors ("control" group). To our knowledge no long-term natural history trials on granulocyte donor infection rates has been undertaken. Although we have shown that there are statistically important negative effects on predonation granulocyte and lymphocyte counts over time, the absolute magnitude of the decrease in these counts may not be clinically meaningful for the donors. To ensure donor safety, additional studies to further characterize this effect should be considered.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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ABBREVIATIONS

CBC(s)	complete blood count(s)
G-CSF/Dex	granulocyte-colony-stimulating factor plus dexamethasone
PBPC(s)	peripheral blood stem cell(s)

REFERENCES

1. Bodey GP. Infection in cancer patients: a continuing association. *Am J Med* 1986;81:11–26.
2. Stanworth SJ, Massey E, Hyde C, et al. Granulocyte transfusions for treating infections in patients with neutropenia or neutrophil dysfunction. *Cochrane Database Syst Rev* 2005;3: CD005339.
3. McPherson RA, Pincus MR. *Henry's clinical diagnosis and management by laboratory methods*. St Louis, Missouri: Elsevier Health Sciences; 2016.
4. Cesaro S, Chinello P, de Silvestro G, et al. Granulocyte transfusions from G-CSF-stimulated donors for the treatment of severe infections in neutropenic pediatric patients with oncohematological diseases. *Support Care Cancer* 2003;11:101–6. [PubMed: 12560938]

5. Price TH, Boeckh M, Harrison RW, et al. Efficacy of transfusion with granulocytes from G-CSF/dexamethasone-treated donors in neutropenic patients with infection. *Blood* 2015;126:2153–61. [PubMed: 26333778]
6. Caspar CB, Seger RA, Burger J, et al. Effective stimulation of donors for granulocyte transfusions with recombinant methionyl granulocyte colony-stimulating factor. *Blood* 1993;81: 2866–71. [PubMed: 7684619]
7. Stroncek DF, Yau YY, Oblitas J, et al. Administration of G-CSF plus dexamethasone produces greater granulocyte concentrate yields while causing no more donor toxicity than G-CSF alone. *Transfusion* 2001;41:1037–44. [PubMed: 11493736]
8. Pulsipher MA, Chitphakdithai P, Miller JP, et al. Adverse events among 2408 unrelated donors of peripheral blood stem cells: results of a prospective trial from the National Marrow Donor Program. *Blood* 2009;113:3604–11. [PubMed: 19190248]
9. Moalic V. Mobilization and collection of peripheral blood stem cells in healthy donors: risks, adverse events and follow-up. *Pathol Biol* 2013;61:70–4. [PubMed: 23199456]
10. Pulsipher MA, Chitphakdithai P, Logan BR, et al. Lower risk for serious adverse events and no increased risk for cancer after PBSC vs BM donation. *Blood* 2014;123:3655–63. [PubMed: 24735965]
11. Quillen K, Yau YY, Leitman SF. The determinants of granulocyte yield in 1198 granulocyte concentrates collected from unrelated volunteer donors mobilized with dexamethasone and granulocyte-colony-stimulating factor: a 13-year experience. *Transfusion* 2009;49:421–6. [PubMed: 19040597]
12. Lazarus EF, Browning J, Norman J, et al. Sustained decreases in platelet count associated with multiple, regular plateletpheresis donations. *Transfusion* 2001;41:756–61. [PubMed: 11399815]
13. Kolf CM, Bolan CD, Wesley RA, et al. Sustained decreases in lymphocyte counts in serial long-term leukapheresis donors. *Transfusion* 2003;43:28A–9A.
14. Verbeke G, Molenberghs G. Linear mixed models for longitudinal data. New York: Springer Science & Business Media; 2009.
15. HPC @ NIH: high-performance computing at the NIH [Internet]. Bethesda: HPC @ NIH; 2017 [Cited 2017 Jun 23]. Available from: <http://hpc.nih.gov>.
16. Nakagawa S, Schielzeth H. A general and simple method for obtaining R² from generalized linear mixed-effects models. *Methods Ecol Evol* 2012;4:133–42.
17. Hölig K, Kramer M, Kroschinsky F, et al. Safety and efficacy of hematopoietic stem cell collection from mobilized peripheral blood in unrelated volunteers: 12 years of single-center experience in 3928 donors. *Blood* 2009;114:3757–63. [PubMed: 19666868]
18. Ratts RB, Weng N-P. Homeostasis of lymphocytes and monocytes in frequent blood donors. *Front Immun* 2012;3:271.
19. Wiedermann CJ, Joannidis M. Accumulation of hydroxyethyl starch in human and animal tissues: a systematic review. *Intensive Care Med* 2014;40:160–70. [PubMed: 24257970]
20. Rossaint J, Berger C, Kraft F, et al. Hydroxyethyl starch 130/0.4 decreases inflammation, neutrophil recruitment, and neutrophil extracellular trap formation. *Br J Anaesth* 2015;114:509–19. [PubMed: 25324349]
21. Brockmann F, Kramer M, Bornhäuser M, et al. Efficacy and Side Effects of Granulocyte Collection in Healthy Donors. *Transfus Med Hemother* 2013;40:258–64. [PubMed: 24179474]
22. Blanchette VS, Dunne J, Steele D, et al. Immune function in blood donors following short-term lymphocytophoresis. *Vox Sang* 1985;49:101–9. [PubMed: 4036082]
23. Martino M, Console G, Dattola A, et al. Short and long-term safety of lenograstim administration in healthy peripheral haematopoietic progenitor cell donors: a single centre experience. *Bone Marrow Transplant* 2009;44:163–8. [PubMed: 19182833]
24. de la Rubia J, de Arriba F, Arbona C, et al. Follow-up of healthy donors receiving granulocyte colony-stimulating factor for peripheral blood progenitor cell mobilization and collection. Results of the Spanish Donor Registry. *Haematologica* 2008;93:735–40. [PubMed: 18387976]
25. Tassi C, Tazzari PL, Bonifazi F, et al. Short- and long-term haematological surveillance of healthy donors of allogeneic peripheral haematopoietic progenitors mobilized with G-CSF: a single institution prospective study. *Bone Marrow Transplant* 2005;36:289–94. [PubMed: 15968277]

Fig. 1.

Predonation granulocyte counts modeled for a Caucasian male donor aged 60 years in donors stimulated with dexamethasone alone, unstimulated granulocyte donors, and PLT donors (left) and donors stimulated with G-CSF/Dex (right).

Fig. 2.

Pre-collection lymphocyte counts ($\times 10^9/L$): pre-collection lymphocyte counts were modeled for a Caucasian male donor aged 60 years by number of donations in allogeneic volunteer granulocyte donors stimulated with G-CSF/Dex, granulocyte donors stimulated with dexamethasone only, PLT donors, and unstimulated granulocyte donors.

Fig. 3.

Granulocyte yield per liter of blood volume processed ($\times 10^{10}$ granulocytes/L) modeled for a Caucasian male donor aged 60 years by number of donations in allogeneic volunteer granulocyte donors stimulated with G-CSF/Dex, granulocyte donors stimulated with dexamethasone only, and unstimulated granulocyte donors.

TABLE 1.

Donor demographics by agent used for stimulation

	G-CSF ± Dex granulocyte donors (n = 229)	Dexamethasone- alone granulocyte donors (n = 264)	Unstimulated granulocyte donors (n = 526)	PLT donors (n = 3743)
Agent	G-CSF ± Dex	Dexamethasone	None	None
Number of donations	1859	684	1391	59,585
Male/female (%)	Male: 151 (66%) Female: 78 (34%)	Male: 176 (67%) Female: 88 (33%)	Male: 373 (71%) Female: 153 (29%)	Male: 1,757 (47%) Female: 1,995 (63%)
Age (years), mean (range)	52.2 (19.7–81.5)	44.5 (19.7–69.9)	43.7 (18.3–78.6)	52.5 (17.1–87.4)
Race	White: 82.1% (n = 188) Black: 6.1% (n = 14) Asian: 2.6% (n = 6) Hispanic: 5.7% (n = 13) Other: 3.4% (n = 8)	White: 73.8% (n = 195) Black: 21.9% (n = 58) Asian: 0.7% (n = 2) Hispanic: 2.6% (n = 7) Other: 0.7% (n = 2)	White: 59.7% (n = 314) Black: 29.8% (n = 157) Asian: 2.7% (n = 14) Hispanic: 4.6% (n = 24) Other: 3.2% (n = 17)	White: 78% (n = 2,924) Black: 8.8% (n = 333) Asian: 4.4% (n = 155) Hispanic: 2.1% (n = 82) Other: 6.6% (n = 249)

TABLE 2.

Studies containing data on donor WBC count change with stimulation

Study	Type	Number	Short term	Long term
Brockmann et al., 2013 ²¹	Granulocytes 5.58 µg/kg G-CSF, 4 mg dexamethasone in 243	378 donors (914 collections)	4 weeks *: lymphocytes 15% decrease; granulocytes decrease 11%	
Blanchette et al., 1985 ²²	Research Lymphapheresis—no stimulation	5 donors (six 8-L procedures each over 12 days)	3 months *: lymphocytes decreased 2% (p > 0.05)	1 year *: lymphocytes increased in four out of five donors (p > 0.05)
Hölig et al., 2009 ¹⁷	PBSCs 7.5 µg/kg G-CSF, 5 to 6 days	3928 donors	4 weeks *: lymphocytes 16.8% decrease (p < 0.01); granulocytes 22% decrease (p < 0.01)	1 year *: lymphocytes 5% decrease (p < 0.01); granulocytes 13% decrease (p < 0.01)
Martino et al., 2009 ²³	PBSCs G-CSF, 10 µg/kg, 5 days	184 donors	Only four donors of 184 had slight leukopenia that resolved within 4 months	2 years *: lymphocytes 1% increase (p < 0.01), granulocytes 11.9% decrease p < 0.01)
de la Rubia et al., 2008 ²⁴	PBSCs	1436 total in registry	4 weeks *: WBC count decreased 16.9%, n = 650 (p < 0.01)	1 year *: WBC count decreased 7%, n = 320 (p = 0.02)
Tassi et al., 2005 ²⁵	PBSCs G-CSF 10 to 12 µg/kg, 5 to 7 days	94 total (80 enrolled)		WBC counts decreased * (1 year, n = 10/83; 2 years, 7/55; 3 years, 5/38); Neutrophils decreased * (1 year, 23/83; 2 years, 12/55; 3 years, 4/38)

* Change from baseline value.