ORIGINAL RESEARCH

TRANSFUSION

Granulocyte transfusions made with modified fluid gelatin in pediatric and adolescent patients with prolonged neutropenia

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Abstract

Background: Granulocyte transfusions (GT) are used to treat progressive systemic or local infections in prolonged neutropenic patients with antibiotic or antifungal resistance. Granulocytes are most commonly collected from whole blood by apheresis using hydroxyethyl starch (HES) as the red blood cell (RBC) sedimentation agent. This is the first study on the safety and efficacy of transfusing granulocytes collected with modified fluid gelatin (MFG) instead of HES to pediatric patients.

Methods: Clinical data from 46 pediatric and adolescent patients receiving at least one MFG-based granulocyte transfusion and in total 295 granulocyte concentrates from July 2013 to August 2019 at our local university medical center were evaluated retrospectively.

Results: Forty-one patients (89%) survived at least 21 days after their last granulocyte transfusion. These survivors had lower CRP values and higher leukocyte counts after GT than non-survivors (mean delta of -5.34 mg/dl vs. -11.99 mg/dl and $+0.62 \times 10^3/\mu l$ vs. $+0.18 \times 10^3/\mu l$ of all GT, respectively). The neutrophil corrected count increment (CCI) was 68.72 mm²/ml in survivors versus 28.00 mm²/ml in non-survivors. There were no major or severe adverse events.

Conclusion: This study suggests that modified fluid gelatin is a safe and effective alternative to hydroxyethyl starch for the collection of granulocytes for transfusion to prolonged neutropenic patients with progressive systemic or local infections refractory to antibiotic or antifungal therapy.

KEYWORDS
granulocytapheresis, granulocyte transfusion, modified fluid gelatin

Jürgen Föll and Norbert Ahrens contributed equally.

1 | INTRODUCTION

Febrile neutropenia (FN) is a common complication of cancer treatment. Reported mortality rates in children and adolescents with prolonged FN because of sepsis range from 1% to 10% depending on the institution.[1–3] Mortality among patients with severe disseminated infections as a complication of prolonged neutropenia is still high despite improvements in chemotherapy protocols for pediatric hematology, solid tumor malignancies, conditioning regimens for hematopoietic stem cell transplantation, and appropriate broad-spectrum antimicrobials or antimycotic prophylaxis and treatment supported by the administration of granulocyte colony-stimulating factor (G-CSF).[4,5] Patients with drug-resistant pathogens are at particularly high risk, and clinical experience suggests that recovery of bone marrow neutrophil production is key to infection control.[6,7] Therefore, granulocyte transfusion (GT) is used to treat these life-threatening infections in neutropenic individuals.

Granulocytes are collected from the blood of healthy donors by apheresis. In order to obtain sufficient numbers of cells for transfusion, donors should be pretreated. Usually, G-CSF and dexamethasone are applied.[8] A red blood cell (RBC) sedimentation agent is required to separate granulocytes from RBCs. High molecular weight hydroxyethyl starch (HES) is commonly used for this purpose, but its safety has been questioned in recent years. HES is known to affect blood coagulation, renal function, and clinical outcomes.[9] In the human body, HES is distributed to the interstitial tissue that acts as a second compartment for long-time deposition. Pruritus and blood coagulation disorders are frequent side effects of hydroxyethyl starch. Excessive HES exposure can result in the occurrence of foamy macrophages and acquired lysosomal storage disease.[10] Compared to single apheresis, multiple aphereses lead to the accumulation of HES in interstitial tissues because of its two-compartment distribution. From these, HES is slowly released back into the blood. The elimination patterns are similar, but HES persistence is more protracted in the latter case. In one study of elimination of hydroxyethyl starch from donor blood, it was predicted that it would take the serum level 72 weeks to reach baseline after multiple aphereses compared to 38 weeks after a single apheresis procedure.[11] In view of these issues, the administration of HES to healthy blood donors has raised ethical concerns and prompted a search for safer alternatives.

A recent study revealed that modified fluid gelatin (MFG) may be a suitable alternative RBC sedimentation agent for granulocyte production.[12] MFG has a biological half-life of 2.5 h in the body, which is much shorter than that of high-molecular-weight HES; granulocyte yields seem to be lower with MFG, but granulocyte function is at least comparable.[13,14]

The granulocyte dose may be crucial. In an animal model of septicemia, granulocyte transfusions saved all dogs treated with 2×10^8 neutrophils per kilogram, but those transfused with 1×10^8 neutrophils per kilogram died.[15] Recently, the randomized Resolving Infection in Neutropenia with Granulocytes (RING) trial described by Price et al.[16,17] revealed that granulocyte transfusion had no overall effect on the primary outcome in neutropenic patients with infection but failed to provide a definitive answer on the efficacy of high-dose GT therapy.[16,17] However, this study was underpowered for the primary outcome, and a post hoc analysis showed that patients who received an average of 0.6×10^9 granulocytes per kilogram tended to have a better outcome than those who did not.[16] According to reviews from the Cochrane Database of Systematic Reviews, there is a low grade of evidence regarding the efficacy of granulocyte transfusions for preventing infections in neutropenic patients, and the effect of such prophylactic granulocyte transfusions is dose-dependent.[18,19] A further study concluded that there is a certain benefit of prophylactic transfusions if the patients receive sufficient doses of granulocytes (at least 10×10^{10} granulocytes per day).[20]

Most studies on granulocyte transfusions for prolonged FN were conducted in adult populations, and data on their use in pediatric patients is limited. Cairo et al. report significantly improved survival of newborns receiving leukocyte transfusions for sepsis.[21] Buffy coat granulocyte transfusions, on the other hand, did not prove to be effective in neutropenic neonates with presumed sepsis.[22] Evidence in support of granulocyte transfusions for neutropenic pediatric patients with infections was also collected in a recent retrospective single-center single-arm analysis, which reported complete or partial resolution of infection in 92% of cases that were given granulocyte transfusions.[23] Like in adults, clinical response in children was very recently confirmed to be dose-dependent.[24] The feasibility of granulocyte transfusions in children and adolescents with severe infections during neutropenia was also demonstrated in a study of 59 patients, who rarely developed side effects despite high doses of granulocytes.[25]

To the best of our knowledge, all studies in pediatric patients were done using granulocytes obtained from apheresis using HES as the RBC sedimentation agent, and none of those have used granulocytapheresis products obtained using MFG instead of HES for RBC sedimentation. Thus, we present the results of the first study of pediatric and adolescent patients with prolonged FN treated with granulocytes collected from apheresis using MFG instead of HES.

2 | PATIENTS AND METHODS

2.1 | Granulocytapheresis

Unrelated healthy males were recruited to donate granulocytes for pediatric and adolescent patients with prolonged FN. Granulocytes were mobilized with 480 μg G-CSF (Neupogen, Amgen Inc.) and 8 mg dexamethasone (Ratiopharm, Ulm, Germany). RBC sedimentation was achieved by MFG, consisting of 435.8 mL Gelafundin 4% (B. Braun Melsungen AG, Melsungen, Germany) and 64.2 ml sodium citrate (1 mol/L, Seracit, Serag-Wiessner, Naila, Germany), prepared under aseptic conditions. This was used instead of ACD-A for anticoagulation. Apheresis was performed with the Cobe Spectra system (Terumo BCT) in the PMN mode and a separation factor of 250 until September 2015, and with the Spectra Optia (Terumo BCT) in the PMN mode with a packing factor of 3.0 thereafter. Settings were adjusted to a visually estimated hematocrit of 5% to 7.5%.

Granulocyte concentrates were transfused without delay after their pharmaceutical release, which generally took place in the afternoon.

The study was approved by the independent ethics committee of the University of Regensburg (approval # 16-101-0153).

2.2 | Patients

Forty-six pediatric and adolescent patients with prolonged febrile neutropenia who received granulocyte transfusions from July 2013 to August 2019 were included in this retrospective single-center analysis. Patients received prophylactic GT to prevent infection and bridge the time until engraftment or marrow recovery after intensive radiotherapy/chemoradiotherapy, or therapeutic GT to treat severe disseminated or local infection, as appropriate.

2.3 | Blood counts

Samples for laboratory and hematological investigations were generally taken in the morning. A Sysmex XE-5000 automated hematology system (Sysmex, Kobe, Japan) was used to measure cell counts in the final apheresis product as well as in blood samples obtained from donors using EDTA tubes before mobilization, before and after apheresis.

2.4 | Statistics

Patient data were documented and stored in the hospital information system (SAP, Walldorf, Germany), and laboratory data in the laboratory data information system (Swisslab). Data dumps were first imported to a spreadsheet program (Excel 2013, Microsoft Corp.) for plausibilization and then exported to SPSS (version 25, IBM, Armonk, NY) for statistical data analysis and graphics. Laboratory value differences between before and after each GT were calculated and tested for significance using the Mann–Whitney-U test with an error probability of 5%. Body surface area (BSA) was calculated using the DuBois formula. The corrected count increment (CCI) was calculated by multiplying the count increment (cells/ml) by body surface area (mm^2) and dividing the product by the number of granulocytes transfused (10^{10}) resulting in granulocytes mm^2/ml .[25]

3 | RESULTS

3.1 | Indications

A total of 46 pediatric and adolescent patients (median age: 8.55 years, range: 0.21–21.75) were included in the analysis. Patients were divided into four groups according to the underlying diagnoses: hematological diseases, solid tumors, hemoglobinopathies, and hereditary immunodeficiencies (Table 1). The intent of granulocyte transfusion was therapeutic in 35 cases (76%) and prophylactic in 11 cases (24%). Two patients from the prophylactic group developed severe sepsis and were therefore also treated therapeutically with GT.

The indications for granulocyte transfusion were prolonged neutropenia following chemotherapy in 22 patients (48%), hematopoietic stem cell transplantation in 21 patients (46%), and neutropenia because of an underlying disease (aplastic anemia with bone marrow failure, stage IV post-transplant lymphoproliferative disorder, and *PIGO* deficiency, respectively) in three cases (6%).

Before the start of GT therapy, 30 patients (65%) had agranulocytosis (less than $0.1 \times 10^3/\mu l$), a further 12 patients (26%) had severe neutropenia ($0.1–0.5 \times 10^3/\mu l$), and 3 additional patients (7%) had moderate neutropenia ($0.5–1.0 \times 10^3/\mu l$), while the remaining patient (2%) suffered from primary immunodeficiency.

3.2 | Granulocyte transfusion therapy

The total number of granulocyte transfusions administered was 295, with a median of 3.0, a mean of 6.41, and a range of 1–37 granulocyte transfusions per patient. The median interval between transfusions was 3.0 days

TABLE 1 Patient characteristics

Number of patients (*n*)	46
Female	18 (39%)
Male	28 (61%)
Children (<18 years)	41
Adolescents (×18 years)	5
Age[a]	8.55 (0.25–21.75) years
Weight[a]	26 (4–106) kg
Height[a]	127 (58–194) cm
Body surface area	Mean: 1.1 (0.25–2.38) m^2 Median: 0.97 m^2
Neutrophil corrected count increment (CCI, mean)	All patients: 65.93 mm^2/ml Survivors: 68.72 mm^2/ml Non-survivors: 28.00 mm^2/ml
Underlying hematological disease	Number of patients (*n*)
Acute lymphoblastic leukemia	10
Acute myeloid leukemia	13
Myelodysplastic syndrome	2
Severe aplastic anemia	3
Hemophagocytic lymphohistiocytosis	1
Polycythemia vera	1
Adipose-derived mesenchymal stromal/stem cell lymphoma, recurrence	1
Post-transplant lymphoproliferative disorder	3
Underlying solid tumor disease	
Neuroblastoma	2
Stomach cancer	1
Ewing sarcoma	1
Hepatoblastoma	1
Atypical teratoid/rhabdoid tumor (AT/RT)	1
Hemoglobinopathies	
Beta thalassemia major	5
Sickle cell disease (HbSS)	1
Spondylocostal dysostosis	1
Inherent immunodeficiencies	
PIGO deficiency	1

[a]Median with range in parenthesis.

(mean: 4.8, range: 1–138 days including patients with multiple transfusion episodes). This was the most rapid pace at which donors could be recruited.

The median granulocyte content of the products was 2.8×10^{10} (mean: 3.37×10^{10}, range: 0.1–11.6×10^{10}), and 103 products (35%) contained $\geq 4.0 \times 10^{10}$ granulocytes per bag. Thus, the median dose administered was 11.81×10^8 granulocytes per kilogram (mean: 6.5×10^8, range: 0.3–81.8×10^8).

3.3 | Outcomes

Three out of 35 patients (9%) in the therapeutic arm and two out of 11 (18%) in the prophylactic treatment arm died within 21 days after the last GT, yielding an overall 21-day mortality rate of 11%. In the therapeutic subgroup, two deaths occurred on the day of the last transfusion, and one on the day after the last transfusion; the cause of death was cardiovascular failure as a complication of

severe cytomegalovirus pneumonia in association with absolute neutropenia after allogeneic stem cell transplantation in one case, and septic shock with multiple organ-system failure (MOF) in the other two. In these, no bacterial or fungal infection was found by microbiological examinations. The two deaths in the prophylactic treatment group occurred 1 or 2 days, respectively, after the last granulocyte transfusion; the cause of death was multiple organ-system failure (MOF) in each case. The five non-survivors received seven (median; range 3–24) granulocyte transfusions compared to three (median; range 1–37) transfusions in the survivors ($n = 41$) (Figure 1).

Patients were observed for 24 days following the first GT (median; range five for one diseased patient up to 636 days). Of the 41 survivors who lived at least 21 days after their last granulocyte transfusion, seven (17%) succumbed to their underlying disease before the end of the observation period (August 31, 2019). Thus, the overall survival rate, calculated as the number of patients that survived at least 30 days after their last transfusion ($n = 34$) divided by the overall sample size ($n = 46$), was 74% (Figure 2).
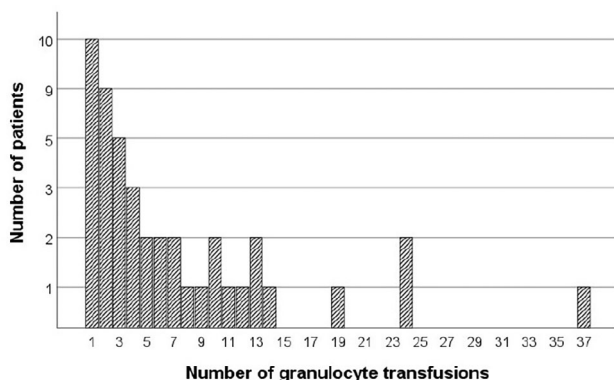

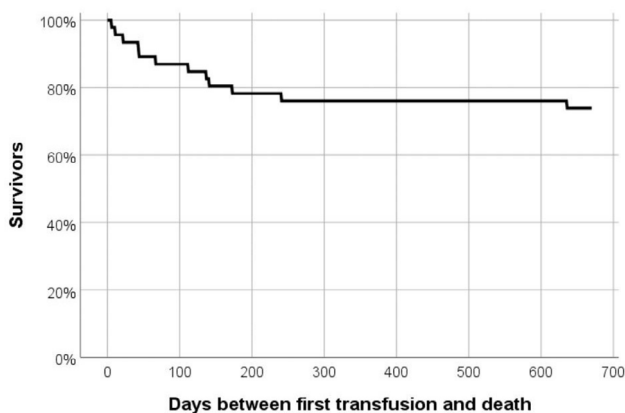**FIGURE 1** Number of granulocyte transfusions per patient

FIGURE 2 Survival following the start of granulocyte transfusion therapy

The secondary outcome variables, body weight, number of granulocyte transfusions received, granulocyte dose, and time between granulocyte transfusions, did not differ significantly between survivors and non-survivors overall. However, the body weight of patients aged 8.55 years or younger, the median age as a cutoff that was found by post-hoc analysis, was significantly lower in survivors (median: 15 kg, mean: 14.5 kg, range: 4–24) than in non-survivors (median of 24.5 kg, mean of 24.5 kg, range: 24–25, $p = .008$), whereas the difference in bodyweight between survivors and non-survivors who were older than 8.55 years was not significant ($p = .268$, data not shown).

3.4 | Adverse events

Adverse events (AEs) occurring up to 2 days after granulocyte transfusion were recorded as part of our hemovigilance system. Fever occurred in six cases (13%) and allergic reactions (exanthema) in two (4%), which were classified as possible adverse reactions to the granulocyte transfusions. Fungal pneumonia occurred due to underlying diseases in two cases and therefore, was not considered to be related to granulocyte transfusion. There were no severe pulmonary adverse events and three patients with mild pulmonary side effects (coughing or transient oxygen supply).

3.5 | Laboratory results

A decreasing trend of CRP values was observed across all patients during GT therapy, which showed a mean of all CRP changes of −9.91 mg/L per GT in 30 days. The cumulative decline was seen in both survivors and non-survivors (Figure 3A). However, the non-survivors had higher initial CRP levels and smaller decreases (mean: −5.34 mg/L per GT) compared to the survivors (−11.99 mg/L per GT). The difference in all CRP values ($n = 1216$) between non-survivors and survivors was highly significant ($p = 5.4 \times 10^{-16}$, Figure 3A).

Overall, the white blood cell (WBC) count increment in response to GT was a median of all GT of $0.53 \times 10^3/\mu l$ (mean: $2.17 \times 10^3/\mu l$, range: $-3.25–26.55 \times 10^3/\mu l$), independent of the patient's weight (Figure 4A). Survivors tended to have higher WBC counts (median: $0.67 \times 10^3/\mu l$, mean: $2.23 \times 10^3/\mu l$, range: $0.0–39.39 \times 10^3/\mu l$) than non-survivors (median: $0.52 \times 10^3/\mu l$, mean: $1.25 \times 10^3/\mu l$, range: $0.01–12.98 \times 10^3/\mu l$, Figure 3B), but the difference was not significant ($p = .12$); count increments were also higher in survivors (median: $0.62 \times 10^3/\mu l$, mean: $2.45 \times 10^3/\mu l$, range: $-1.82–26.55 \times 10^3/\mu l$) than in
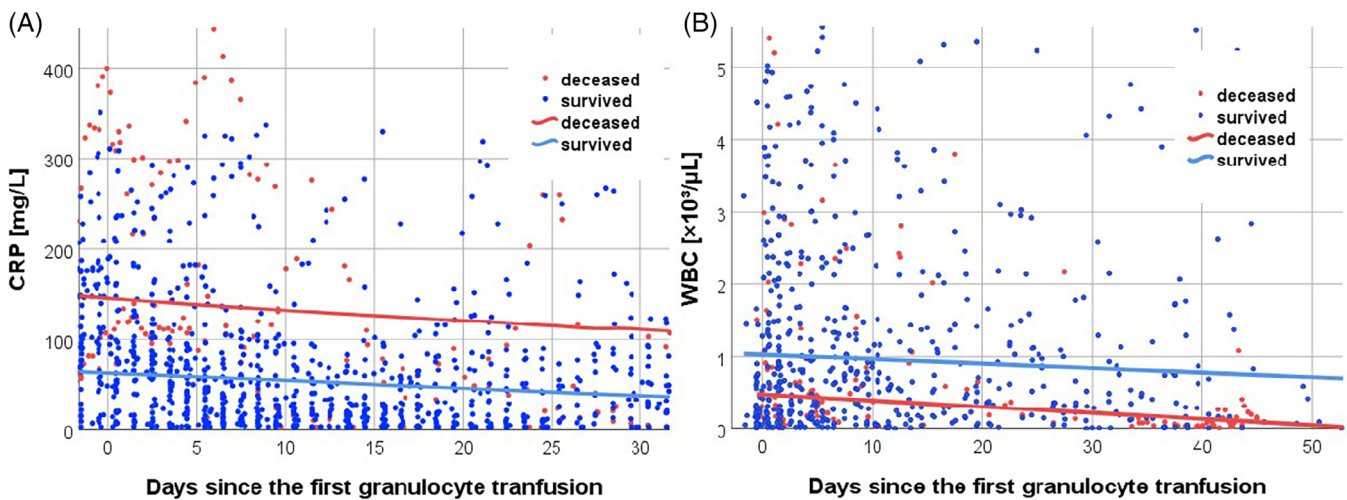
FIGURE 3 C-reactive protein (CRP) levels (A) and white blood cell (WBC) counts (B) in patients who survived at least 21 days after their last granulocyte transfusion (blue) and those who did not (red)

non-survivors (median: $0.18 \times 10^3/\mu l$, mean: $0.64 \times 10^3/\mu l$, range: -3.25–$5.20 \times 10^3/\mu l$).

Overall, the average neutrophil count increment was $2.02 \times 10^3/\mu l$ (median: $0.53 \times 10^3/\mu l$, range: -0.75–$20.69 \times 10^3/\mu l$, and the neutrophil CCI was 65.93 mm^2/ml (Figure 4B,C). The post-transfusion increase in neutrophil count tended to be higher in survivors (mean of all transfusions: $2.19 \times 10^3/\mu l$, median: $0.70 \times 10^3/\mu l$, range: -0.75–$20.69 \times 10^3/\mu l$) than in non-survivors (mean of all transfusions: $0.63 \times 10^3/\mu l$, median: $0.29 \times 10^3/\mu l$, range: -0.56–$5.95 \times 10^3/\mu l$), and the difference was significant ($p = .026$). Neutrophil CCI also differed significantly between survivors (68.72 mm^2/ml) and non-survivors (28.00 mm^2/ml, Table 1).

Furthermore, the size of the white blood cell and neutrophil granulocyte count increment was granulocyte dose-dependent (Figure 5A,B).

4 | DISCUSSION

Here, we retrospectively investigated the efficacy and safety of transfusing granulocytapheresis products manufactured with MFG to pediatric and adolescent patients with prolonged neutropenia for infection treatment or prevention. Prophylactic GT was administered to bridge the time until bone marrow recovery, and therapeutic GT was used to treat febrile neutropenic patients with infections refractory to systemic or local antibiotics. Granulocyte transfusion resulted in a decrease in CRP levels in all patients, but the decrease was greater in survivors than in non-survivors. Thus, CRP appears to be an indicator of clinical response to GT therapy. This confirms previous findings from the Netherlands, where curative

GT was uniformly followed by a CRP decrease (median of 285.5 mg/L before to 90.5 mg/L after 2 GT).[26] As in our study, the CRP decline was more pronounced in survivors than it was in non-surviving patients (in the median from 282.5 to 73.5 mg/L compared to 306 to 188.5 mg/L, respectively). This is consistent with German data from Hannover where CRP dropped for all patients from a median of 171 mg/L before to 140 mg/L after granulocyte transfusions (post) with survivors showing lower post CRP values than non-survivors (119 and 187 mg/L, respectively).[27]

WBC and neutrophil counts increased in all patients after GT. The sequence of sampling on the day following transfusion ensured that temporarily trapped granulocytes inside the lung could redistribute, but it was with the same prone to overlooking successfully transfused granulocytes that were rapidly used up for pathogen elimination.

Impaired increments for both WBC and neutrophils were especially seen in non-survivors. Apart from neutrophil consumption, neutrophil antibodies in the patient could have contributed to reduced increments. The neutrophil CCI in survivors was higher than that in non-survivors. Our data furthermore show that neutrophil and WBC numbers increased almost proportionally to the granulocyte dose per kilogram of body weight. Jendiroba & Freireich pointed to the importance of dosage as key to successful granulocyte transfusion.[28] Likewise, Price et al. found that higher doses were associated with better treatment responses and higher survival rates.[16] The same was recently confirmed for children who, despite their lower body weight, also showed increased effects with higher cell doses.[24]

In this study, the patient's body weight, number of granulocyte transfusions, granulocyte dose, and time
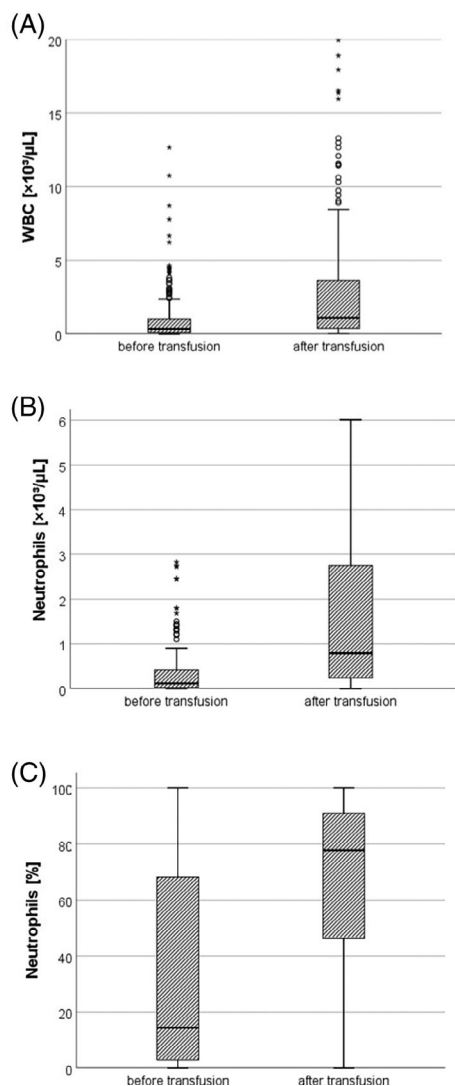
FIGURE 4 White blood cell (WBC) counts (A) as well as absolute (B) and relative (C) neutrophil counts before and after granulocyte transfusion

interval between granulocyte transfusions did not seem to differ significantly between survivors and non-survivors. Young survivors (≤ 8.55 years) had a lower body weight than young non-survivors. Apart from that, the immune system in younger patients is likely to differ, younger patients may have different disease compositions, and the pretreatment could differ that may affect immunization to granulocytes. These factors might explain why younger patients were more likely to survive.

Our data did not clearly capture the time interval between the diagnosis of neutropenia and the first granulocyte transfusion. Time intervals between GC transfusions, however, were recorded, and they did not differ significantly between survivors and non-survivors. Delayed granulocyte transfusion is known to negatively correlate with the survival of neutropenic children with sepsis. Uppuluri et al. found that the timing of granulocyte transfusions plays a key role in determining their clinical efficacy: the chances of survival are better when transfusion is performed early in the septic crisis.[29] Garg et al. also concluded that overall survival was significantly better in patients who received GT within 7 days of neutropenic sepsis than in those who received it later.[30]

Granulocyte transfusions have various common side effects. An estimated 17.5–41% of patients who receive granulocyte products manufactured with hydroxyethyl starch develop fever after the transfusion, and pulmonary complications, including potentially fatal reactions, and alloimmunization with decreasing therapeutic efficiency of later transfusions are common adverse reactions of HES-based granulocyte transfusion.[16,31] Therefore, indication for GT is recognized in patients whose chances of survival are 30% or less without transfusion.[31]

Some medical centers, including ours, have observed lower frequencies of side effects. Seidel et al. reported that
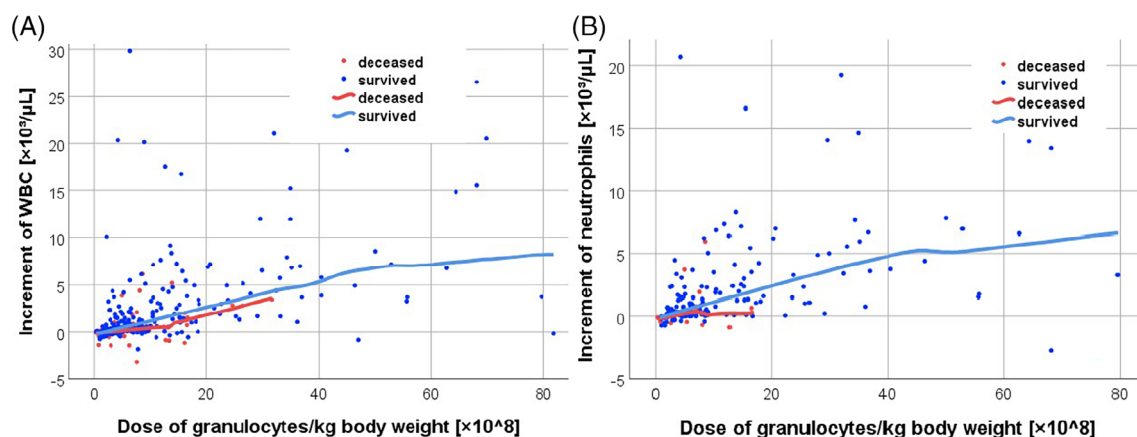

FIGURE 5 White blood cell (WBC, A) and neutrophil increments (B) in patients who survived at least 21 days after their last granulocyte transfusion (blue) and those who did not (red), by dose of granulocytes/kg body weight

less than one in six patients developed mild fever and chills as a side effect of HES-containing granulocyte transfusions in their prospective clinical study,[23] which is close to the rate of 17% in response to MFG-based granulocyte transfusions in the present study. Explanations for these differences could be not only less pre-immunization and better product safety profiles in the present study but also differences in the pretreatment regimens and the inclusion of mild side effects like exanthemata in AE reporting.

Adverse event reporting in granulocyte transfusion is inherently difficult to standardize because of the variety in clinical conditions and partly immunosuppressive treatments. In addition, side effect frequencies are known to depend upon the concept of hemovigilance with active versus passive reporting systems.

Clinical data and experience with MFG in granulocytapheresis and its impact on the side effects of granulocyte transfusion are sparse. Dullinger and Doblinger have shown that HES can be replaced by MFG, but it is important to weigh individual factors to decide whether preventing side effects or collecting larger granulocyte yields is more important in determining whether the generally used agent, HES or the alternative, MFG, should be used.[12,14] This adds to experiences from the advent of granulocyte transfusion,[32,33] and is in line with our observations and 89% survival rate.

Of the five (11%) out of 46 patients who died within 21 days of their last granulocyte transfusion, two succumbed to their underlying disease. Though all of these patients were classified as non-survivors in this study, a causal relationship of the latter two deaths to granulocyte transfusion is unlikely. The other three deaths were due to sepsis or infection, which suggests that the administered doses may not have provided sufficient amounts of functional granulocytes in these cases.

According to Gea-Banacloche, researchers generally recommend a median transfusion dose of at least 4.0×10^{10} granulocytes per unit.[31] Estcourt et al. mentioned dosages of 1.0×10^{10} granulocytes per day to reduce the risk of infection.[18,19] The present study was not sufficiently powered to demonstrate a significant difference in granulocyte dose in survivors and non-survivors. According to critics of the above studies, the positive outcome of the high-dose granulocyte transfusions was not a random occurrence but rather a largely site-specific effect because the dose was not stochastically distributed.[31] Teofili et al. even found that overdosage may have an unfavorable effect: their preliminary data suggest that levels of inflammatory response mediators increase in a dose-related manner after GT, which may explain the detrimental effect of high-dose transfusions.[34] However, their retrospective review of data from 96 patients with hematological malignancies receiving granulocyte transfusions collected from relatives or friends revealed that infection-related mortality was not influenced by the number of GTs or by the total amount of granulocytes transfused per infectious episode: The poor outcome of patients receiving low-dose GTs ($<1.5 \times 10^8$ cells/kg), which were viewed as inadequate, was similar to that of patients in the high-dose group receiving GTs with a presumed-adequate median dose of 3×10^8 cells/kg.[34]

This study has limitations resulting from its retrospective and single-arm study design. Therefore, a randomized, prospective trial comparing HES and MFG-based harvesting methods in GT production for pediatric and adolescent high-risk patients would be desirable to investigate the efficacy and safety of both granulocyte harvesting methods for therapeutic or prophylactic purposes.

CONFLICT OF INTEREST

The authors have no conflicts of interest to disclose.

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REFERENCES

1. Hann I, Viscoli C, Paesmans M, Gaya H, Glauser M. The International Antimicrobial Therapy Cooperative Group (IATCG) of the European Organization for Research and Treatment of Cancer (EORTC). A comparison of outcome from febrile

neutropenic episodes in children compared with adults: results from four EORTC studies. Br J Haematol. 1997;99(3):580–8.

2. Thangthong J, Anugulruengkitt S, Lauhasurayotin S, Chiengthong K, Poparn H, Sosothikul D, et al. Predictive factors of severe adverse events in pediatric oncologic patients with febrile neutropenia. Asian Pac J Cancer Prev. 2020;21(12):3487–92.

3. Kuderer NM, Dale DC, Crawford J, Cosler LE, Lyman GH. Mortality, morbidity, and cost associated with febrile neutropenia in adult cancer patients. Cancer. 2006;106(10):2258–66.

4. Penack O, Becker C, Buchheidt D, Christopeit M, Kiehl M, von Lilienfeld-Toal M, et al. Management of sepsis in neutropenic patients: 2014 updated guidelines from the Infectious Diseases Working Party of the German Society of Hematology and Medical Oncology (AGIHO). Ann Hematol. 2014;93(7):1083–95.

5. Fioredda F, Calvillo M, Bonanomi S, Coliva T, Tucci F, Farruggia P, et al. Congenital and acquired neutropenias consensus guidelines on therapy and follow-up in childhood from the Neutropenia Committee of the Marrow Failure Syndrome Group of the AIEOP (Associazione Italiana Emato-Oncologia Pediatrica). Am J Hematol. 2012;87(2):238–43.

6. Akova M. Epidemiology of antimicrobial resistance in bloodstream infections. Virulence. 2016;7(3):252–66.

7. Dale D, Reynolds H, Pennington J, Elin R, Herzig G. Experimental Pseudomonas pneumonia in leukopenic dogs: comparison of therapy with antibiotics and granulocyte transfusions. Blood. 1976;47(5):869–76.

8. Liang D-C. The role of colony-stimulating factors and granulocyte transfusion in treatment options for neutropenia in children with cancer. Paediatr Drugs. 2003;5(10):673–84.

9. Bellmann R, Feistritzer C, Wiedermann CJ. Effect of molecular weight and substitution on tissue uptake of hydroxyethyl starch: a meta-analysis of clinical studies. Clin Pharmacokinet. 2012;51(4):225–36.

10. Auwerda JJ, Leebeek FWG, Wilson JHP, van Diggelen OP, Lam KH, Sonneveld P. Acquired lysosomal storage caused by frequent plasmapheresis procedures with hydroxyethyl starch. Transfusion. 2006;46(10):1705–11.

11. Maguire LC, Strauss RG, Koepke JA, Bowman RJ, Zelenski KR, Lambert RM, et al. The elimination of hydroxyethyl starch from the blood donors experiencing single of multiple intermittent-flow centrifugation leukapheresis. Transfusion. 1981;21(3):347–53.

12. Dullinger K, Pamler I, Brosig A, Mohrez M, Hähnel V, Offner R, et al. Granulocytapheresis with modified fluid gelatin versus high-molecular-weight hydroxyethyl starch: a matched-pair analysis. Transfusion. 2017;57(2):397–403.

13. Mitra S, Khandelwal P. Are all colloids same? How to select the right colloid. Indian J Anaesth. 2009;53(5):592–607.

14. Doblinger N, Bredthauer A, Mohrez M, Hähnel V, Graf B, Gruber M, et al. Impact of hydroxyethyl starch and modified fluid gelatin on granulocyte phenotype and function. Transfusion. 2019;59(6):2121–30.

15. Appelbaum F, Bowles C, Makuch R, Deisseroth A. Granulocyte transfusion therapy of experimental Pseudomonas septicemia: study of cell dose and collection technique. Blood. 1978;52(2):323–31.

16. Price TH, Boeckh M, Harrison RW, McCullough J, Ness PM, Strauss RG, et al. Efficacy of transfusion with granulocytes from G-CSF/dexamethasone-treated donors in neutropenic patients with infection. Blood. 2015;126:2153–61.

17. Cancelas JA. Granulocyte transfusion: questions remain. Blood. 2015;126(18):2082–3.

18. Estcourt LJ, Stanworth S, Doree C, Blanco P, Hopewell S, Trivella M, et al. Granulocyte transfusions for preventing infections in people with neutropenia or neutrophil dysfunction. The Cochrane Library. 2015;6:CD005341.

19. Estcourt LJ, Stanworth SJ, Hopewell S, Doree C, Trivella M, Massey E. Granulocyte transfusions for treating infections in people with neutropenia or neutrophil dysfunction. Cochrane Database Syst Rev. 2016;4:CD005339.

20. Cairo MS, Rucker R, Bennetts GA, Hicks D, Worcester C, Amlie R, et al. Improved survival of newborns receiving leukocyte transfusions for sepsis. Pediatrics. 1984;74(5):887–92.

21. Baley JE, Stork EK, Warkentin PI, Shurin SB. Buffy coat transfusions in neutropenic neonates with presumed sepsis: a prospective, randomized trial. Pediatrics. 1987;80(5):712–20.

22. Díaz R, Soundar E, Hartman SK, Dreyer Z, Teruya J, Hui S-KR. Granulocyte transfusions for children with infection and neutropenia or granulocyte dysfunction. Pediatr Hematol Oncol. 2014;31(5):425–34.

23. Seidel MG, Minkov M, Witt V, Matthes-Martin S, Pötschger U, Worel N, et al. Granulocyte transfusions in children and Young adults: does the dose matter? J Pediatr Hematol Oncol. 2009;31(3):166–72.

24. Johann PD, Wuchter P, Trojanova L, Sturm D, Lu KH-N, Kulozik AE, et al. Cui bono? Identifying patient groups that may benefit from granulocyte transfusions in pediatric hematology and oncology. J Pediatr Hematol Oncol [Internet]. 2021 [cited 2021 Nov 7];Publish Ahead of Print; https://journals.lww.com/10.1097/MPH.0000000000002349

25. Massey E, Harding K, Kahan BC, Llewelyn C, Wynn R, Moppett J, et al. The granulocytes in neutropenia 1 (GIN 1) study: a safety study of granulocytes collected from whole blood and stored in additive solution and plasma. Transfus Med. 2012;22(4):277–84.

26. Drewniak A, Boelens J-J, Vrielink H, Tool ATJ, Bruin MCA, van den Heuvel-Eibrink M, et al. Granulocyte concentrates: prolonged functional capacity during storage in the presence of phenotypic changes. Haematologica. 2008 Jul;93(7):1058–67.

27. Grigull L, Pulver N, Goudeva L, Sykora K-W, Linderkamp C, Beilken A, et al. G-CSF mobilised granulocyte transfusions in 32 paediatric patients with neutropenic sepsis. Support Care Cancer. 2006;14(9):910–6.

28. Jendiroba DB, Freireich EJ. Granulocyte transfusions: from neutrophil replacement to immunereconstitution. Blood Rev. 2000;14(4):219–27.

29. Uppuluri R, Ramachandrakurup S, Vaidhyanathan L, Kandath S, Subburaj D, Raj R. Changing trends in the use of granulocyte transfusions in neutropenic children with sepsis in India. Ind J Hematol Blood Transfus. 2017;33(2):207–10.

30. Garg A, Gupta A, Mishra A, Singh M, Yadav S, Nityanand S. Role of granulocyte transfusions in combating life-threatening infections in patients with severe neutropenia: experience from a tertiary care Centre in North India. PLoS One. 2018;13(12):e0209832.

31. Gea-Banacloche J. Granulocyte transfusions: a concise review for practitioners. Cytotherapy. 2017;19(11):1256–69.

32. Huestis DW, Loftus TJ, Gilcher R, Lichtiger B, Rock G, Price TH, et al. Modified fluid gelatin. An alternative macromolecular agent for centrifugal leukapheresis. Transfusion. 1985; 25(4):343–8.

33. Rock G, Wise P, Kardish R, Huestis DW. Modified fluid gelatin in leukapheresis accumulation and persistence in the body. Transfusion. 1984;24(1):68–73.

34. Teofili L, Valentini CG, Di Blasi R, Orlando N, Fianchi L, Zini G, et al. Dose-dependent effect of granulocyte transfusions in hematological patients with febrile neutropenia. PLoS One. 2016;11(8):e0159569.