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Effectiveness of Triple Saline Washed Leucoreduced Red Cells in Prevention of Post Transfusion Reaction Among Beta- Thalassemia Major Patients

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ABSTRACT

Introduction: Beta-thalassemia major, a genetic blood disorder, requires lifelong blood transfusions to manage chronic anemia, but these transfusions pose significant risks, including febrile non-hemolytic transfusion reactions (FNHTRs), alloimmunization, allergic reactions, and iron overload. The triple saline wash method, a form of leukoreduction, efficiently reduces leukocyte counts in packed red blood cells (PRBCs), mitigating these complications. This study evaluates the effectiveness of this technique in minimizing post-transfusion reactions, improving transfusion safety for patients with beta-thalassemia major. **Objective:** To assess the efficacy of triple saline-washed leucoreduced PRBCs in minimizing post-transfusion reactions and correlating leukoreduction levels with clinical outcomes. **Methods:** A cross-sectional study was conducted from January 2023 to October 2024 at a tertiary care center. Fifty-one transfusion-dependent beta-thalassemia major patients were enrolled. Inclusion criteria included regular transfusions and confirmed diagnosis. Leukocyte counts were measured before and after triple saline washing, and post-transfusion reactions were monitored. Statistical analysis compared reaction incidences with leukoreduction levels. **Results:** Among the participants, 68.6% achieved <85% leukoreduction, while 31.4% exceeded this benchmark. Patients with higher leukoreduction rates (>85%) experienced significantly fewer reactions (6.3%) compared to those with lower rates (40.0%, p = 0.014). FNHTRs were the most common adverse reactions (53.3%), followed by allergic reactions (40.0%). Pre-transfusion hemoglobin levels mostly ranged from 8.5–9 g/dL (39.2%), aligning with guidelines. The O+ blood group predominated (37.3%). Overall, 98.82% of transfusions were uneventful. **Conclusion:** The triple saline wash method demonstrated significant efficacy in reducing post-transfusion reactions among beta-thalassemia major patients, especially those achieving >85% leukoreduction. This method offers a cost-effective, practical solution for transfusion safety in resource-limited settings, warranting further research on long-term outcomes.

INTRODUCTION

Beta-thalassemia major is a hereditary blood disorder caused by mutations in the beta-globin gene, leading to defective hemoglobin synthesis. This condition is characterized by chronic anemia, requiring regular blood transfusions for survival. Although blood transfusions are a cornerstone of treatment, they carry risks such as alloimmunization, febrile non-hemolytic transfusion reactions (FNHTRs), allergic reactions, iron overload, and transfusion transmitted infections. These complications not only increase morbidity but also complicate management, necessitating additional interventions like iron chelation therapy and immuno-modulatory treatments [1].

Among these complications, FNHTRs and alloimmunization are significantly associated with the presence of leukocytes in transfused blood products. Leukocytes in stored blood can trigger immune responses, leading to the production of antibodies against donor antigens and the release of inflammatory cytokines. These immune-mediated reactions pose challenges, particularly for patients requiring lifelong transfusions. To mitigate these risks, leukodepletion a process that reduces leukocyte content in blood products has become an integral component of transfusion medicine [2]. Leukodepletion can be achieved through filtration, buffy coat removal, or washing red blood cells (RBCs) with saline solutions. Of these, the triple saline wash method has emerged as a cost-effective and efficient approach, particularly in resource-limited settings.

The triple saline wash method involves washing packed red blood cells (PRBCs) three times with saline to remove leukocytes, plasma proteins, platelets, and other cellular debris. This process not only reduces leukocyte counts but also eliminates inflammatory cytokines and residual plasma proteins, thereby minimizing the risk of transfusion reactions. The effectiveness of this method can be assessed using leukoreduction log values, which reflect the percentage reduction in leukocyte count. For instance, a 1-log reduction corresponds to a 90% reduction, while a 3-log reduction achieves 99.9% leukoreduction. Studies indicate that achieving high levels of leucodepletion significantly reduces FNHTRs, allergic reactions, and other transfusion-related adverse events [3].

Patients with beta-thalassemia major represent a unique population with specific transfusion needs. Regular transfusions are essential to maintain pre-transfusion hemoglobin levels between 9 and 10.5 g/dL, ensuring optimal growth, development, and quality of life. However, repeated exposure to donor antigens increases the risk of alloimmunization, where the immune system develops antibodies against transfused red cell antigens. This condition complicates future transfusions by making it increasingly difficult to find compatible blood units [4]. Moreover, chronic transfusions contribute to iron overload, necessitating lifelong chelation therapy to prevent complications such as liver fibrosis, cardiomyopathy, and endocrine dysfunction. Such challenges emphasize the significance of employing strategies like leukodepletion to limit transfusion-related complications.

The implementation of triple saline-washed PRBCs has shown promising results in reducing post-transfusion reactions. This method is particularly advantageous for beta-thalassemia major patients, as it addresses the dual challenges of FNHTRs and allergic reaction. By removing over 85% of residual leukocytes and cytokines, this technique lowers the immunological burden on the recipient.

Clinical studies have demonstrated a significant decrease in the incidence of adverse transfusion reactions with the use of triple saline-washed red cells, validating its safety and effectiveness [5].

In a study conducted at the Department of Immunohematology and Blood Transfusion of Government Wenlock Hospital, the effectiveness of the triple saline wash method in preventing post- transfusion reactions among beta-thalassemia major patients was evaluated. The study measured leukocyte counts before and after the washing process and documented the incidence and types of post-transfusion reactions. Key findings indicated that patients with higher leukoreduction rates (≥85%) experienced significantly fewer adverse reactions compared to those with lower leukoreduction rates. The results highlight the clinical relevance of this intervention in enhancing transfusion safety [6].

The demographic profile of the study population reflects the chronic nature of beta-thalassemia major, with most patients falling within the 11-20 years age group and requiring frequent transfusions. The majority of adverse reactions observed were FNHTRs, followed by allergic reactions, emphasizing the need for effective leukoreduction techniques. Global literature supports these findings, demonstrating that leukoreduction filters and washing methods can achieve up to a 3-log reduction in leukocyte count, substantially lowering the risk of transfusion-related complications [7].

The use of triple saline-washed leucodepleted red cells is particularly relevant in resource-constrained settings where advanced leukodepletion filters may not be readily available. This method offers a practical and cost-effective solution to improve transfusion safety without compromising efficacy. By reducing the incidence of post-transfusion reactions, it also lessens the burden on healthcare systems, as fewer complications translate to shorter hospital stays and reduced need for additional interventions.

The triple saline wash approach offers an improvement in transfusion strategies for beta-thalassemia major patients [8]. Its ability to effectively reduce leukocyte counts and minimize post-transfusion reactions makes it an invaluable tool in managing this high-risk population. Future research should focus on optimizing this technique and exploring its long-term benefits in improving the quality of life and clinical outcomes for patients with transfusion-dependent conditions.

This study aimed to evaluate the quality of leukoreduction in packed red blood cells (PRBCs) using the triple saline wash method, quantified by log reduction values, and to correlate the findings with post-transfusion clinical outcomes in chronically transfused patients with beta thalassemia. The objectives included assessing the effectiveness of the triple saline wash method in achieving leukoreduction and studying the post-transfusion reactions associated with the use of triple-saline-washed red cells.

MATERIALS AND METHODS

This cross-sectional study was conducted at the Regional Advanced Paediatric Care Centre (RAPCC) and Regional Blood Transfusion Centre, Government Wenlock Hospital, Mangalore, from January 2023 to October 2024. The Ethical approval has been obtained to conduct the study. The study included 51 transfusion-dependent beta-thalassemia major patients who met the inclusion criteria of confirmed diagnosis, age above six months, regular transfusions, and willingness to consent. Patients with beta-thalassemia minor or intermedia, acute illness, or unwillingness to participate or follow up were excluded. The research aimed to evaluate transfusion outcomes among these participants, contributing to the understanding of clinical management in chronically transfused patients.

RESULTS

Patients under the age of ten constituted the largest group (33.3%), while those over the age of twenty accounted for 13.7%, with the majority of patients falling within the intermediate age range (52.9%). Additionally, there was a slightly higher proportion of male patients (54.9%) compared to female patients (45.1%)

Table 1: Weight distribution of patients

		No of patients	Percentage
Weight	<20	14	27.5%
	20 - 30	10	19.6%
	30 - 40	13	25.5%
	>40	14	27.5%

The weight distribution of patients was balanced, with (27.5% each). The 20–30 kg group accounted for 19.6% of the highest proportions in the <20 kg and >40 kg groups patients, while 25.5% were in the 30–40 kg range.

Table 2: Blood group distribution of patients

		No of patients	Percentage
Blood Group	A-	2	3.9%
	A+	8	15.7%
	AB+	5	9.8%
	B-	1	2.0%
	B+	12	23.5%
	O-	4	7.8%
	O+	19	37.3%

The most prevalent blood group among the patients was O+ (37.3%), followed by B+ (23.5%), indicating a significant predominance of these types. Less common blood groups included A+ (19.6%) and AB+ (13.7%), while the rarest were B- (2.0%) and A- (3.9%), highlighting the variability in blood group distribution within the patient population.

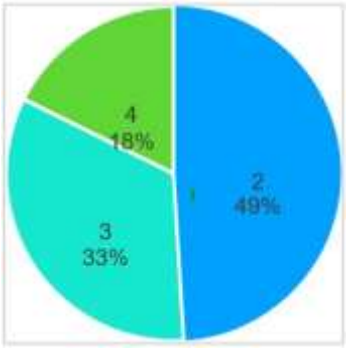

Figure 1: Transfusion interval distribution of patients

Most patients had a transfusion interval of 2 weeks (49.0%), indicating the highest frequency of transfusions, followed by 3-week intervals (33.3%). The least common interval was 4 weeks (17.6%), reflecting a smaller subset of patients requiring less frequent transfusions. This distri bution highlights the varying transfusion needs among the patient population.

Table 3: Types of adverse transfusion reaction

		No of patients	Percentage
Adverse Transfusion Reactions	Allergic Reactions	6	40.0%
	FNHTR	8	53.3%
	Hemolytic Transfusion Reactions	1	6.7%

Febrile non-haemolytic transfusion reactions (FNHTR) were the most common adverse transfusion reactions, accounting for 53.3%, followed by allergic reactions at 40.0%. Haemolytic transfusion reactions were the least frequent, occurring in only 6.7% of cases, highlighting FNHTR as the predominant concern in transfusion-related complications.

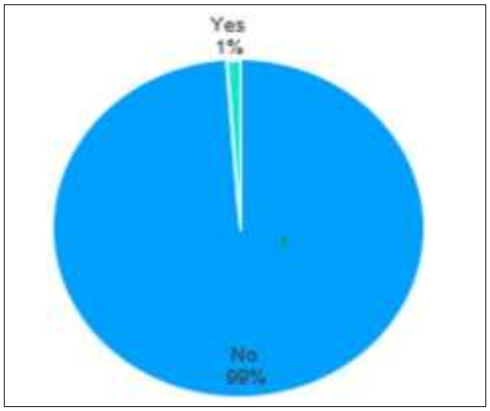

Figure 2: Adverse transfusion reactions in patients

The majority of transfusions (98.82%) were uneventful,reactions, indicating a high level of safety in the transfusion with only a small percentage (1.18%) associated with adverse process.

Table 4: Distribution of patients by average pre-transfusion haemoglobin levels

		No of patients	Percentage
Average Pre-Transfusion HB	<7.5	6	11.8%
	7.5 – 8	5	9.8%
	8 - 8.5	13	25.5%
	8.5 – 9	20	39.2%
	9 - 9.5	4	7.8%
	>9.5	3	5.9%

The majority of patients had an average pre-transfusion hemoglobin level of 8.5–9 g/dL (39.2%), followed by those with levels of 8–8.5 g/dL (25.5%). Smaller proportions were observed in patients with hemoglobin levels >9.5 g/dL (5.9%) and <7.5 g/dL (11.8%), indicating that most patients-maintained levels closer to the target range.

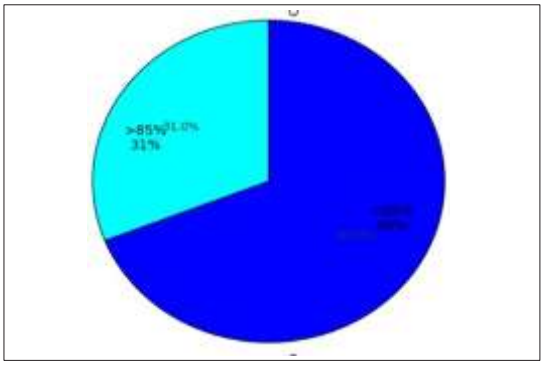

Figure 3: Distribution of patients by average % of LR attained per month

The majority (68.6%) of patients achieved less than 85% of their monthly target LR, while 31.4% exceeded 85%. This indicates that most patients struggled to meet the benchmark, highlighting potential areas for intervention to improve adherence and outcomes.

Table 5: Adverse transfusion reactions and average % of LR attained per month

		average % of LR attained per month				P value
		<85%		>85%		
		Count	Column N %	Count	Column N %	
Total of Adverse Transfusion Reactions During the Study	1	14	40.0%	1	6.3%	0.014
	NIL	21	60.0%	15	93.8%	

Patients achieving <85% average LR per month experienced significantly more adverse transfusion reactions (40.0%) compared to those exceeding 85% (6.3%). The statistically significant difference (p = 0.014) underscores the importance of maintaining higher leucoreduction levels to reduce adverse outcomes.

DISCUSSION

Our study highlights the effectiveness of triple-saline-washed leucoreduced red cells in preventing post-transfusion reactions in beta-thalassemia major patients. By reducing leukocyte count significantly, this method minimized adverse reactions, with 98.82% of patients remaining reaction-free. Conducted at Government Wenlock Hospital, the study correlates leukoreduction efficacy with improved clinical outcomes in chronically transfused beta-thalassemia major patients [9].

Comparative studies validate our findings on leucoreduction efficacy in minimizing transfusion reactions. Kumar et al. (2006) showed a reduction in NHFTR from 4% to 1% in thalassemia patients using pre-storage filters (PALL, USA), with only 1% of 303 patients experiencing NHFTR. Brownlee et al. (2009) emphasized achieving 99.9% leukocyte reduction to prevent reactions, aligning with our low incidence (1.18%). Ahmed et al. (2000) observed a 15.2% NHFTR rate, increasing with transfusion exposure, highlighting the critical importance of minimizing transfusion reactions [10, 11, 12].

The clinical benefits of washing red blood cells (RBCs) before transfusion have been widely studied. Schmidt et al. (2016) demonstrated that washing RBC units reduced the production of human leukocyte antigen (HLA)-specific antibodies in children with chronic kidney disease, supporting our findings. Cardigan et al. (2020) emphasized the theory and practice of washed RBCs, highlighting their effectiveness in reducing transfusion-related complications. However, Busch et al. (2017) reported no significant differences in clinical outcomes between patients receiving washed RBCs and those who did not, suggesting that the benefits of this technique may depend on specific patient populations and conditions [13,14,15].

The 1999 TRICC trial by Hebert et al. demonstrated reduced mortality and multiorgan dysfunction in ICU patients managed with a conservative transfusion strategy compared to a liberal approach. A French hemovi gilance study highlighted significant reductions in bacterial sepsis (3.8% to 1.7%) and NHFTRs (32.9% to 25.8%) after implementing universal leukoreduction. These findings underscore the importance of universal leukoreduction in minimizing transfusion reactions, particularly in thalassemia patients requiring long-term transfusions [16].

Pandey et al. (2020) emphasized flow cytometry as a reproducible, objective method for counting residual white blood cells (rWBCs) in leukodepleted blood components, surpassing the Nageotte counter's accuracy. Our study revealed that most patients were aged 11–20 years (52.9%), followed by under 10 years (33.3%) and over 20 years (13.7%), reflecting early transfusion needs in beta-thalassaemia major. Males (54.9%) outnumbered females (45.1%), aligning with thalassaemia prevalence trends in males [17].

Our study found a significant correlation between leukoreduction (LR) efficacy and adverse transfusion reactions. Patients with LR <85% per month had more reactions (40.0%) than those with LR >85% (6.3%) (p = 0.014), emphasizing the importance of high LR levels, consistent with Taher et al. (2013). Most patients had pre-transfusion hemoglobin levels of 8.5–9 g/dL (39.2%), aligning with recommendations by Taher et al. (2013) and Cappellini et al. (2014) [18, 19].

The blood group distribution and pre-transfusion hemoglobin levels in our study align with existing literature. The predominance of O+ and B+ types mirrors trends reported by Shah et al. (2013). Taher et al. (2013) recommend maintaining pre-transfusion hemoglobin levels of 9–10 g/dL, consistent with our findings. Lal et al. (2020) emphasized strict transfusion protocols to minimize complications, while Beshlawy et al. (2024) highlighted effective leukoreduction in managing transfusion-dependent thalassemia. Chiang jong et al. (2021) and Fletcher et al. (2023) reinforced the clinical importance of leukoreduction [18, 20, 21, 22, 23, 24].

Shanthala Devi et al. (2014) reported three reactions (0.2%) among 161 thalassemic patients: two with

leukoreduced PRBCs (buffy-coat method) and one with non-leucoreducedPRBCs, but none with bedside filter transfusions, highlighting bedside filters as a superior leukodepletion strategy. Sharma et al. (2010) advocated third- and fourth-generation leukofilters for optimal leukoreduction. Taher et al. (2013) emphasized effective blood management and psychological support for transfusion-dependent thalassemia. These findings under score a comprehensive approach integrating strict protocols and holistic patient care [18, 25, 26].

Sharma et al. (2010), Macnamara et al. (2019), and Shapiro (2020) emphasized that leukoreduction reduces leukocyte-associated adverse effects in PRBCs. Bhattac harya et al. (2011) observed 105 adverse events, 41% being FNHTRs, in a study of 56,503 transfusions. Tan et al. (1993) reported an 8.5% transfusion reaction rate among 26 multi-transfusion-dependent thalassemic patients, with signifi cantly fewer reactions in the filtered blood group ($p<0.05$). Our study corroborates these findings, highlighting the cost-effective efficacy of triple saline-washed leucoreduced red cells in resource-limited settings [26, 27, 28, 29, 30].

CONCLUSION

Our study demonstrated that the triple saline wash method significantly reduced leukocyte counts in PRBCs, effectively minimizing post-transfusion reactions in beta-thalassemia major patients. Adverse reactions occurred in only 1.18% of patients, primarily febrile non-haemolytic (53.3%) and allergic reactions (40.0%), underscoring the importance of achieving >85% leukoreduction. Demographics aligned with existing literature on age, gender, weight, blood groups, and transfusion intervals. Most patients had pre-transfusion hemoglobin levels between 8.5–9 g/dL, consistent with optimal guidelines. Our findings advocate for leucodepleted antigen-matched units, and future research on long-term monitoring, patient education, and enhanced transfusion management.

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