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Original article

THE IMPACT OF STORAGE CONDITIONS ON PLATELETS IN PLATELET-RICH PLASMA (PRP) AND PLATELET CONCENTRATE (PC).

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Abstract.

Introduction:

The ultimate aim of blood transfusion services is to give recipients whole blood and blood components that are secure, efficient, and potent. Control, assurance, and audit of quality are the foundational elements of quality. Product standards are developed and accurately met through the process of quality control.

Method:

In this study, samples of 2.3% PRBCs (50/2155), 9.3% platelet concentrate (42/451), and 3.6% of total whole blood collection (48/1,321) were prepared and tested for haematological parameters of quality control for the period from 1 January 2022 to 31 December 2022. Parameters for 8 samples of platelet concentrate available on day 5, were compared for day 0 and day 5.

Result:

Haematological metrics for whole blood, PRBCs, and platelet concentrate underwent quality examination. 50/2121, or 2.3% of the total whole blood collection, was used as a sample for the analysis of haematological parameters. Volume and haematocrit tests were performed on a total of 55 out of 2335 (2.3%) units. With a range of 344–365 mL, the average capacity was 358 mL. With a range of 35.1-48.0%, the mean haematocrit was 42.2%. Between 271 to 389 mL, the mean capacity was 311 mL. The range of the mean haematocrit was 57.3 to 81.9%. There was a slight decrease in volume, platelet count and WBC count of platelet concentrates on day 5 as compared to day 0.

Conclusion:

For effective and safe blood transfusion services, continuous quality improvement is crucial.

Recommendation:

Attention should be taken to applying consistent criteria for quality assessment, and objective analysis should be used regularly to verify that consistent values are being followed.

Keywords: Platelet output, Haematocrit, Quality Control, Quality Assurance, Submitted: 2023-09-04 Accepted: 2023-09-13

1. INTRODUCTION.

Small nucleated cytoplasmic particles called platelets are crucial for blood clotting and wound healing. [1] The TCA cycle and respiratory chain provide both anaerobic and aerobic metabolism

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for platelet energy. The anaerobic component is linked to the conversion of glucose to lactic acid, whereas the aerobic oxygen-dependent component causes complete substrate breakdown. [2]

Platelet-rich plasma (PRP) is made from blood that has higher platelet concentrations than whole blood. PRP has been utilized to treat musculoskeletal disease and injury in horses for more than ten years [3-5]. Although the platelets in PRP receive most of the attention, they also contain plasma, white blood cells (WBC), and red blood cells (RBC). PRP's physiologic actions are ultimately caused by the proteins found in each of its constituent parts [6-8]. Growth factors with chemotactic, mitogenic, and matrix synthesis activities in tissues include insulin-like growth factor (IGF), platelet-derived growth factor (PDGF), and transforming growth factor (TGF). Additionally present in PRP, inflammatory mediators and catabolic enzymes such as matrix metalloproteinase-9 (MMP-9) are related to the quantity of WBC [9].

A byproduct of whole blood recovered by centrifugation is platelet-rich plasma (PRP). It has a high concentration of platelets, which upon activation release several growth factors including platelet-derived growth factor (PDGF), transforming growth factor (TGF), vascular endothelial growth factor (VEGF), insulin-like growth factor-1 (IGF-1), and fibroblast growth factor basic (FGF-B).[10-12] Recent research has revealed that these growth factors encourage cellular development, proliferation, and differentiation, which helps in tissue regeneration, resulting in a boost in the popularity of PRP and its widespread use in orthopaedic injury [12-14].

Fitness for use or compliance with criteria are examples of quality [15]. The constant and reliable performance of services, processes, and products by the prescribed requirements is referred to as quality assurance [15]. Because blood banking is an essential component of healthcare, it is crucial to analyze whole blood and blood components properly to ensure recipients receive the most benefit [16]. In modern transfusion medicine, blood-component transfusion therapy has evolved into the gold standard of care. The new developments and improvements in blood collection, anticoagulant compositions, component separation equipment, storage, refrigeration, screening of transfusion transmissible diseases, and other methods of donor selection and screening have made it possible to use blood transfusion therapy widely and wisely [17].

Obtaining some intact platelets might be intriguing, but multiple studies showed that PRP's ability to collect considerable numbers of leukocytes was also beneficial, for instance in the treatment of chronic elbow tendinopathy. [18-20] There isn't currently any evidence linking growth factor release to lingering white blood cells in PRP. Growth factors, platelets, and circulating cells are the products' determining components, and several specialists in various fields of sports medicine noted that higher cell concentrations were employed with relatively small amounts of leukocytes to provide superior clinical outcomes.

2. MATERIALS AND METHODS.

This study was carried out from the first of January 2022 to the last day of December 2022 at Nalanda Medical College and Hospital, Patna, Bihar, India.

Data was gathered for the aforementioned period during which, with written consent, blood was drawn from 3476 healthy donors weighing more than 45 kg in sterile single, double, or triple blood bags containing the anticoagulant Citrate Phosphate Dextrose Adenine 1 (CPDA 1).

Out of these 3476 units, 1321 units (350 mL of whole blood from donors weighing 45 to 60 kg) were used as whole blood, and 2155 units (450 mL of blood from healthy donors weighing more than 60 kg) were processed for component preparation in a chilled centrifuge

Units were centrifuged at 3800 (4400 x g) rotations per minute at 4 °C for 9 minutes to separate them into PRBCs and FFP after 2-4 hours of holding time. FFP and platelet concentrate units were centrifuged at 2 spin centrifugations at 1500 rpm for 9 minutes at 22°C followed by 2500 (4400 x g) rpm for 15 minutes at 22 C to separate whole blood into PRBCs [21].

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We prepared samples of 3.6% of the total whole blood collection (48/1321), 2.3% of PRBCs (50/2155), and 9.3% of platelet concentrate (42/451) and tested them for haematological parameters for quality control by the norms outlined in the "Technical Manual of Transfusion Medicine." [25].

Blood components or units with obvious haemolysis or discoloration were excluded from the quality analysis.

Haematocrit and total volume measurements were performed on samples of whole blood and PRBCs. Formulas were used to determine volume:

- *Whole blood: 1 gm=1 05 mL*
- *PRBCs: 1 gm = 1 09 mL*
- *Platelet concentrate: 1 gm=1 035 mL*

Weight was determined by dividing the empty weight by the filled weight of the bag.

Standard calibrated weighing scales were used to measure weight.

The total volume, swirling motions, platelet production, and WBC counts per unit of the platelet concentrate were all measured. The data for the total volume of platelet concentrate, total volume, swirling motion, platelet count and WBC counts per unit was compared on day 0 and day 5 for 8 samples available on the 5th day.

After a brief squeeze along the bag's sides against the light, platelet swirling movement was visually appraised and recorded as "present" or "absent" [16].

Haematocrit, platelet, and WBC counts were performed.

2.1. Statistical analysis.

SPSS software version 12 was used to do statistical analysis in terms of mean, range, and standard deviation.

3. RESULTS.

Blood was taken from healthy, pre-screened donors in the amount of 4456 units, of which 2121 were used as whole blood and 2335 were divided into components (FFP and PRBCs; FFP, PRBCs, and PC).

Table1: Quality control results of whole blood and PRBCs

Parameter	Whole blood				Packed red blood cells			
	Recommended [1,7]	Mean	Range	Concordance	Recommended [1,7]	Mean	Range	Concordance
Volume (in mL)	351±10%	358	344-365	99.9%	271±61	311	271-389	99.9%
Hematocrit	>31%	42.2%	35.1-48.0%	99.9%	>54%	68.4%	57.3%-81.9%	99.9%

50/2121, or 2.3% of the total whole blood collection, was used as a sample for the analysis of haematological parameters. Volume and haematocrit tests were performed on a total of 55 out of 2335 (2.3%) units. With a range of 344–365 mL, the average capacity was 358 mL. With a range of 35.1-48.0%, the mean haematocrit was 42.2%. Between 271 to 389 mL, the mean capacity was 311 mL. The range of the mean haematocrit was 57.3 to 81.9%. [Table-1].

Table 2: Quality control results of platelet concentrate.

Parameter	Quality requirement [1,7]	Mean±SD (mL)	Range	Concordance
RBC Contamination	$<0.1\times10^{11}$/liter	$0.071\pm0.04\times10^{12}$/liter	$0.01-0.13\times10^{12}$/liter	94.1%
WBC contamination	5.4×10^{6} - 4×10^{8} in 449 mL bag.	$2.20\pm1.46\times10^{8}$ /unit	$0.16-5.1\times10^{8}$ /unit	94.5%
Volume	51-69 mL/bag	62.6±4.45	51-69 mL	100%
Platelet count	$>5.4\times10^{10}$/ bag in 76% of bags	$7.3\pm1.42\times10^{10}$/unit	$5.0-11.8\times10^{10}$/ unit	94%
Inspection	Swirling movement of platelets	Present in all units		

A total of 45 out of 450 (9.3%) units underwent volume, platelet count, and contamination tests for RBC and WBC [Table/Fig-4]. With a range of 50–70 mL, the mean volume was 66.6 mL. With a range of 0.16-5.1x10^{8}/unit, the mean WBC contamination was 2.2x 10^{8}/unit. With a range of 0.01–0.13x10^{12}/litre, the mean RBC contamination was 0.071x10^{12} [Table-2]. With a range of $5.0–11.8\times10^{10}$/unit, the mean platelet count was 7.3×10^{10}/unit.

Table 3: Quality control results of platelet concentrate (Day 0 versus Day 5).

Parameter	Mean±SD (mL)(On day 0)	Mean±SD (mL)(On day 5)
WBC contamination	3.2±2.1×10^8 /unit	2.57±1.87×10^8 /unit
Volume	63.1±4.1	61.8±3.81
Platelet count	6.9±1.26×10^{10}/unit	6.16±1.20×10^{10}/unit
Inspection	Clear Swirling movement of platelets	Swirling movement of platelets in some part of the bag in 2 samples

8 samples were assessed on day 5 for volume, swirling motions, platelet count and WBC count. The mean WBC contamination for 8 samples compared on day 0 and day 5 was 3.2 2.1± 10^8/unit and 2.57 ±87 x10^8/unit respectively. Volume of platelet concentrate on day 0 and day 5 was 63.±4.1and 61.8±3.81 respectively. Swirling was observed only in some parts of the bag in 2 out of 8 samples on day 5. The mean platelet count for samples compared on day 0 and day 5 was 6.9±1.26×10^{10}/unit and 6.16±1.20×10^{10} /unit respectively.

4. DISCUSSION.

In addition to maintaining an appropriate supply of blood, blood banks also have the twin obligation of supplying recipients with safe blood and components that are as effective as possible [22]. The ultimate production objective of transfusion medicine is a "zero-risk blood supply" [23].

Internal Quality Control is a crucial component of every laboratory service's quality assurance. It is the pre-defined collection of methods that are used for ongoing performance standards evaluation of normal work [24]. Decreasing the number of negative blood reactions is made possible by maintaining quality control standards in blood banks. It is intended to monitor variances in production processes, and product quality, and verify that manufacturing steps satisfy set requirements for acceptance by periodic quality analysis of blood components [25].

The present study's overall quality control audit was found to have complied with national norms while staying within allowable bounds.

According to accepted standards [26], the recommended volume for whole blood was 350 /450±10 mL with a hematocrit of >30% and the recommended volume for PRBCs was 280±40 mL with a hematocrit of >55%. The mean volume of PRBCs in the current study was 310 mL with a range of 270-390 mL, while the mean volume of whole blood units was 359 mL with a range of 345-375 mL. While the mean hematocrit of PRBCs was 69.5% with a range of 56.3 to 80.9%, the mean hematocrit of whole blood units was 41.7% with a range of 36.2-49%. All of the whole blood and PRBC units examined had volume and hemoglobin concentrations that were well within acceptable ranges, demonstrating the high caliber of our blood bank. In a study conducted by Upadhyay S et al. [27], with results that are similar to those of this study, the mean volume of whole blood units was 410.81 mL with a range of 391-522 mL and the hematocrit of whole blood units was 43.73.2% with a range of 38-52.5% [27]. The hematocrit was 54.2% with a range of 41-69%, and the mean volume of PRBC units was 285.24 mL.

According to accepted standards, the platelet quality control criteria were also within the usual range. Other investigations conducted by Raturi M et al., Singh RP et al., Fijnheer R et al., Hirosue A et al., and others also produced simi- lar findings. [16,28-30]. A study of the various hematological characteristics of platelet concentrate was also conducted by Raveendran R. et al. However, their mean platelet count was higher than this study's, and both studies met the normal standard requirements for volume and WBC contamination [31]. There was a slight decrease in mean values for platelet count and WBC counts on day 5 as compared to day 0. Because individuals with thrombocytopenia require platelet concentrate transfusions to stop or prevent bleeding, platelet transfusions must be of high quality to be effective.

As a result, the metrics measuring the quality of the platelets in our blood bank were in line with the aforementioned research and with generally accepted practices, proving their quality.

5. CONCLUSION.

Our blood bank's platelet concentrate, PRBCs, and whole blood all satisfy established national standards for quality. The quality of platelet concentrate was well maintained on day 5. Safe blood transfusion is a fundamental human right that should be made accessible through effective quality control throughout the whole blood collection, component preparation, and recipient distribution procedures. It is important to create, monitor, and accurately document quality indicators. The importance of consistent updating and adherence to standard operating procedures cannot be overstated. Participation in a program that promotes donor and recipient hemovigilance will also establish ethical transfusion procedures.

6. LIMITATIONS.

The limitations of this study include a small sample population who were included in this study. The findings of this study cannot be generalized for a larger sample population. Furthermore, the lack of a comparison group also poses a limitation for this study's findings.

7. RECOMMENDATION.

Attention should be taken to applying consistent criteria for quality assessment, and objective analysis should be used regularly to verify that consistent values are being followed.

8. ACKNOWLEDGEMENT.

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9. LIST OF ABBREVIATIONS.

PRP- platelet-rich plasma
PC- platelet concentrate
PRBC- Packet red blood cell
TCA- Tricyclic antidepressants
WBC- white blood cells
RBC- red blood cells
IGF- insulin-like growth factor
PDGF- platelet-derived growth factor
TGF- transforming growth factor
VEGF- vascular endothelial growth factor
FGF-B- fibroblast growth factor basic
FFP- Fresh frozen plasma

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11. Conflict of interest.

The authors report no conflicts of interest in this work.

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