

Study of Storage-Related Changes in Whole Blood and Packed Red Blood Cells in the Blood Bank of a Tertiary Care Centre of Western Uttar Pradesh

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Abstract

Introduction: Blood transfusion services are integral to modern medicine. With the increasing shift toward component therapy, understanding the storage-related changes in blood products is essential. Whole blood and packed red blood cells (PRBCs) are commonly stored and used, but undergo significant biochemical and haematological changes over time, known as “storage lesions.” This study was conducted to evaluate such changes during a 35-day storage period for both PRBCs and Whole blood.

Methodology: This observational study included 100 blood units – 50 whole blood and 50 PRBC units – randomly collected from voluntary donors at a tertiary care blood center. Samples were assessed on Days 0, 7, 14, 21, 28, and 35 for haematological (Hb, RBC count, RBC indices) and biochemical (Na⁺, K⁺, LDH) parameters using standard analyzers. analysis was done by SPSS software® version 20.0. Statistical analysis was performed using repeated measures analysis of variance (RM ANOVA), appropriate for assessing intra-unit changes across multiple time points (Days 0, 7, 14, 21, 28, and 35). RM ANOVA accounts for the correlation between repeated measurements on the same blood unit and provides robust evidence for time-dependent changes in both haematological and biochemical parameters. Significance was set at $p \leq 0.05$.

Results: Significant haematological and biochemical changes were observed with increasing storage duration. In PRBCs, haemoglobin decreased from 17.25 ± 1.67 g/dL on Day 0 to 14.56 ± 2.2 g/dL on Day 35 ($p < 0.05$), while in whole blood it increased slightly. Potassium levels increased progressively, while sodium levels declined. LDH levels rose markedly, especially in PRBCs, indicating hemolysis. All changes were statistically significant ($p < 0.05$).

Conclusion: Storage leads to predictable and significant deterioration in blood quality. Monitoring haematological and biochemical parameters is crucial to ensure transfusion efficacy and patient safety, particularly beyond 14 days of storage.

Keywords: Storage lesions, PRBC, Whole Blood, Haematological, Biochemical, Potassium, LDH, Blood Storage.

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Introduction

Blood is a vital biological fluid that performs essential functions such as oxygen delivery, nutrient transport, waste removal, and immune defense in the human body. It comprises plasma and cellular elements, including red blood cells (RBCs), white blood cells (WBCs), and platelets, each playing a distinct physiological role. RBCs primarily function to transport oxygen and carbon dioxide due to the presence of haemoglobin, WBCs are critical for immune defense, and platelets are essential for hemostasis through clot formation [1].

The demand for safe and effective blood transfusions is a critical component of healthcare delivery. Advances in transfusion medicine have led to a shift from whole blood transfusions to component therapy, allowing targeted treatment using packed red blood cells (PRBCs), fresh frozen plasma, and platelet concentrates [2]. This practice enhances the efficiency of transfusion and optimizes the use of donated blood based on patient-specific clinical needs [3].

Despite improved preservation techniques, storage of blood—especially PRBCs and whole blood—is associated with various morphological and biochemical changes collectively termed as “storage lesions.” These include decreased ATP levels, hemolysis, increased potassium, reduced 2,3-diphosphoglycerate (2,3-DPG), and elevated lactate dehydrogenase (LDH) levels, which may impair post-transfusion efficacy and safety. RBCs stored in CPDA-1 solution have a shelf life of 35 days, while the use of additive solutions like SAG-M can extend PRBC storage to 42 days [4].

Storage lesions not only reduce the deformability and oxygen-carrying capacity of RBCs but can also increase the risk of transfusion-related complications such as hyperkalemia, especially in vulnerable populations like neonates or patients requiring massive transfusions [5]. Moreover, biochemical deterioration, such as increased LDH and potassium and reduced pH, may influence transfusion outcomes and contribute to prolonged hospital stays [6].

Several studies, including those by Oyet et al. and Chaurasiya et al., have documented significant haematological and biochemical shifts in stored blood,

reinforcing the need for careful evaluation of storage effects to guide clinical practice [7,8]. Whole blood, though less commonly used in modern transfusion protocols, is still vital in emergency settings and trauma care, especially in resource-limited areas where component separation may not be feasible [9].

In this context, the present study was undertaken to assess the storage-related haematological and biochemical changes in whole blood and PRBCs over a 35-day period at a tertiary care blood center. The findings aim to contribute to the growing body of evidence on storage lesions, with implications for optimizing transfusion practices and ensuring patient safety.

Material and Methods

This hospital-based study was conducted in the Department of Pathology at Muzaffarnagar Medical College, Muzaffarnagar, Uttar Pradesh, over a period of 18 months, which included 12 months of data collection and 6 months of data analysis as an observational analysis of storage-related changes over time, with measurements taken longitudinally from the same blood unit at Day 0, 7, 14, 21, 28, and 35. Therefore, the within-unit trends in haemoglobin and other parameters primarily reflect storage-associated changes, rather than inter-donor variability. The haemoglobin values presented at each time point were derived from repeated measures on the same unit, not cross-sectional comparisons between donors.

That said, we acknowledge that initial baseline variability in haemoglobin among donors could influence the absolute starting values and may affect the magnitude of observed changes. To minimize this confounding we used a random sampling method and included only healthy, voluntary donors who met standard blood bank eligibility criteria, helping to narrow the physiological range of donor haemoglobin values, the statistical analysis focused on time-dependent changes within each product type (PRBCs and whole blood) rather than direct between-group comparisons of absolute values and all measurements were carried out under identical storage conditions and laboratory protocols to reduce technical variability.

A total of 100 blood units were randomly selected using a simple random sampling technique, including 50 units of whole blood and 50 units of packed red blood cells (PRBCs).

All blood was collected in citrate phosphate dextrose adenine (CPDA-1) anticoagulant bags and stored at 2–6°C. Donor demographic details such as age, gender, and blood group were recorded at the time of donation. The units were analyzed with measurements taken longitudinally from the same blood unit at six time intervals: Day 0 (baseline), and subsequently on Days 7, 14, 21, 28, and 35 of storage.

Haematological parameters including haemoglobin (Hb), red blood cell (RBC) count, packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), and mean corpuscular haemoglobin concentration (MCHC) were measured using an automated hematology analyzer in the central clinical laboratory. Biochemical parameters—sodium (Na^+), potassium (K^+), and lactate dehydrogenase (LDH)—were estimated using an automated biochemistry analyzer.

Samples were excluded if the blood was clotted or if the blood bags tested positive for transfusion-transmitted infections. All measurements were carried out under standardized laboratory conditions to minimize pre-analytical variability. Data entry was made in MS Office Excel software in codes and analysis was done by SPSS software® version 20.0. Statistical analysis was performed using repeated measures analysis of variance (RM ANOVA), appropriate for assessing intra-unit changes across multiple time points (Days 0, 7, 14, 21, 28, and 35). RM ANOVA accounts for the correlation between repeated measurements on the same blood unit and provides robust evidence for time-dependent changes in both haematological and biochemical parameters. Significance was set at $p \leq 0.05$. Ethical clearance for the study was obtained in accordance with institutional guidelines and protocols.

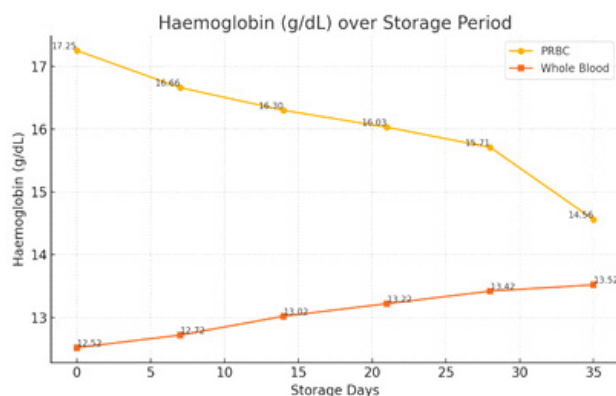
Results

This study analyzed a total of 100 blood units, comprising 50 units each of whole blood and packed red blood cells (PRBC), stored at 2–6°C in CPDA-1 anticoagulant solution. Haematological and biochemical parameters were measured on Days 0, 7, 14, 21, 28, and 35.

Haemoglobin (Hb)

In PRBC: Hb concentration declined significantly across storage ($F = 15.091$, $p = 6.41 \times 10^{-13}$). Mean Hb dropped from 17.25 ± 1.67 g/dL at baseline to 14.56 ± 2.20 g/dL on Day 35, a mean decrease of 2.69 g/dL.

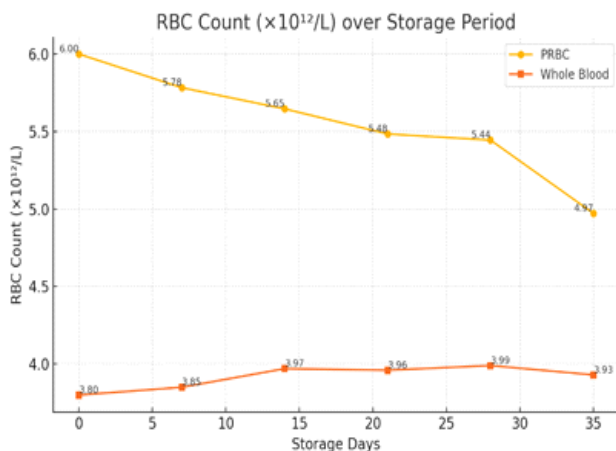
This decline was statistically significant from Day 7 onward. In whole blood, Hb showed no significant change over time ($F = 0.560$, $p = 0.731$), with a slight increase of 1.0 g/dL observed from 12.02 ± 1.96 to 13.02 ± 2.18 g/dL, likely reflecting variability.



RBC Count

RBC count in PRBC units declined significantly ($F = 16.387$, $p = 6.04 \times 10^{-14}$), falling from $5.999 \pm 0.75 \times 10^6/\mu\text{L}$ to $4.972 \pm 0.86 \times 10^6/\mu\text{L}$ by Day 35. This reduction of approximately $1.03 \times 10^6/\mu\text{L}$ confirms red cell loss during storage.

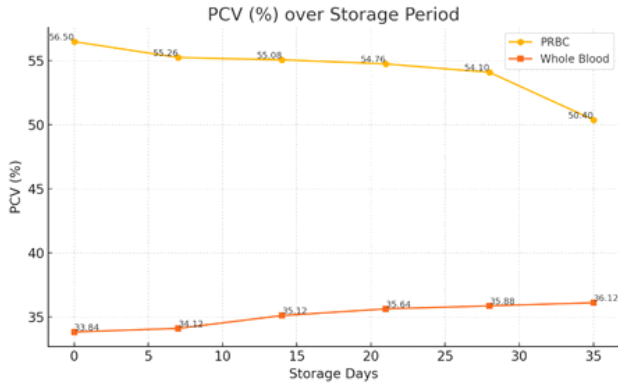
Whole blood showed a mild rise in RBC count over time ($F = 2.462$, $p = 0.034$), increasing from 3.80 ± 0.74 to $3.93 \pm 0.78 \times 10^6/\mu\text{L}$, though clinical relevance is limited.



Packed Cell Volume (PCV / Hematocrit)

HCT showed a significant decrease in PRBCs ($F = 5.852, p = 3.97 \times 10^{-5}$), from $56.5 \pm 5.66\%$ to $50.4 \pm 8.48\%$, a reduction of 6.1%. This likely reflects lysis-related plasma expansion.

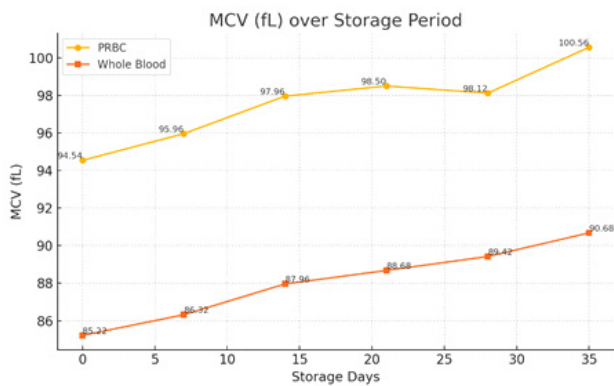
In contrast, whole-blood HCT increased from $33.84 \pm 6.67\%$ to $36.12 \pm 7.42\%$ ($F = 5.654, p = 5.99 \times 10^{-5}$), consistent with hemoconcentration or storage-related shifts.



MCV (Mean Corpuscular Volume)

MCV increased significantly in PRBC ($F = 12.938, p = 3.54 \times 10^{-11}$), rising from 94.5 ± 9.08 fL to 100.6 ± 10.65 fL. This cell swelling reflects osmotic changes during storage.

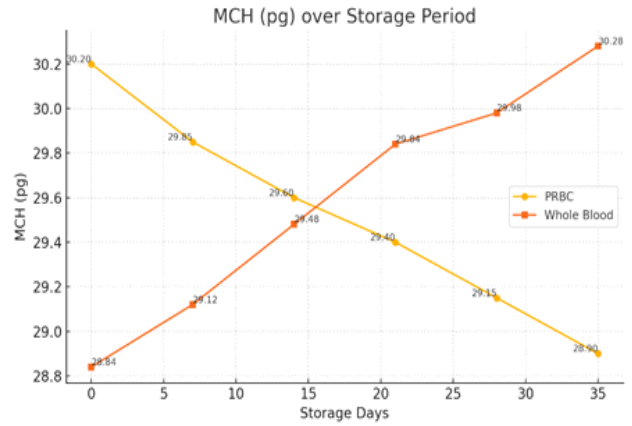
A similar trend was seen in whole blood with a sharp MCV rise ($F = 68.142, p = 4.06 \times 10^{-44}$) from 85.22 ± 9.13 to 90.68 ± 11.78 fL, showing membrane instability.



MCH (Mean Corpuscular Haemoglobin)

MCH declined slightly in PRBCs ($F = 2.289, p = 0.0466$), with means shifting from 30.2 ± 1.72 to 28.9 ± 1.40 pg—just breaching significance.

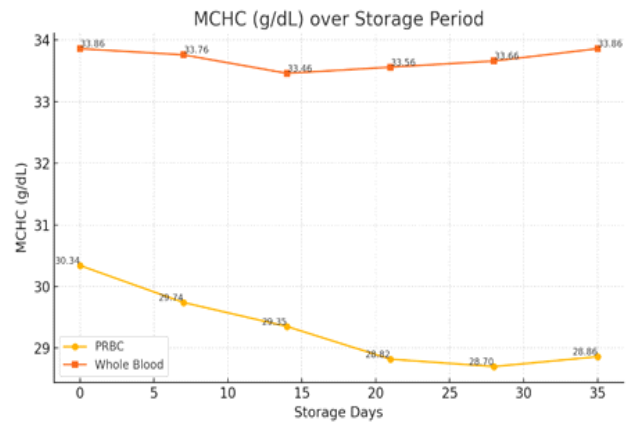
Whole blood displayed a marked MCH rise ($F = 61.992, p = 2.62 \times 10^{-41}$); values peaked beyond Day 21, consistent with increased hemoglobin per cell volume.



MCHC (Mean Corpuscular Haemoglobin Concentration)

MCHC significantly decreased in PRBC units ($F = 7.354, p = 1.92 \times 10^{-6}$), suggesting intracellular Hb dilution.

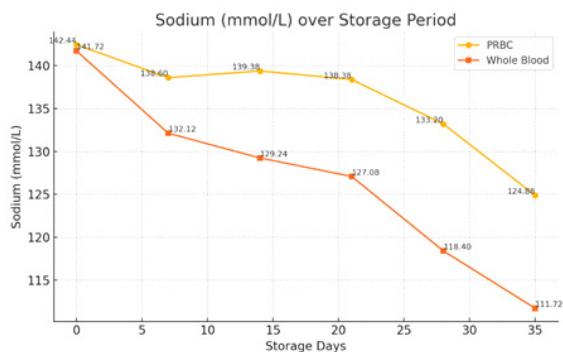
Whole-blood MCHC fluctuated mildly ($F = 3.624, p = 0.0035$), ending slightly lower than baseline.



Sodium (Na⁺)

Na⁺ fell by 17.6 ± 8.69 mmol/L in PRBCs ($F = 18.086, p = 2.90 \times 10^{-15}$), from 142.4 to 124.9 mmol/L by Day 35, suggesting progressive electrolyte leakage.

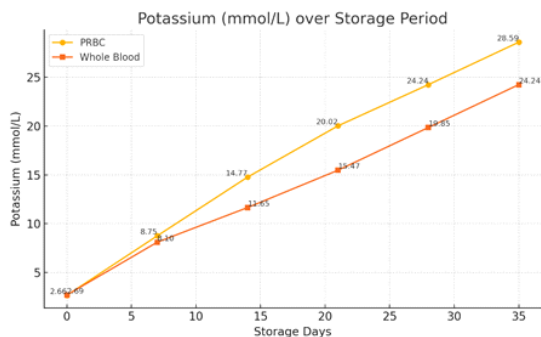
Whole blood units showed a steeper Na⁺ drop of 30.0 ± 2.05 mmol/L ($F = 90.664, p = 2.76 \times 10^{-53}$), reflecting greater ionic imbalance.



Potassium (K⁺)

K⁺ rose dramatically in PRBCs from 2.66 ± 1.34 to 28.59 ± 2.78 mmol/L (F = 13.748, p = 7.73 × 10⁻¹²), a 25.93 mmol/L increase.

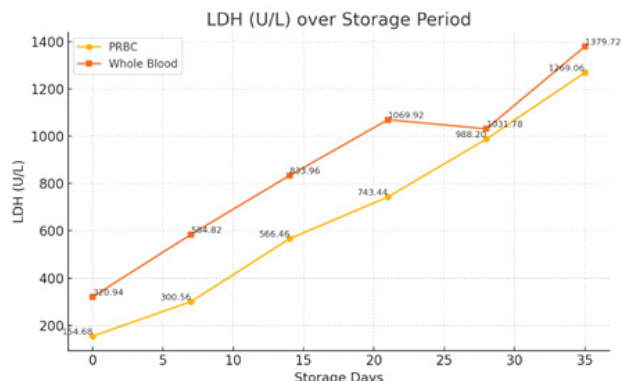
In whole blood, K⁺ rose from 2.69 ± 0.78 to 24.24 ± 2.94 mmol/L (F = 852.909, p = 1.43 × 10⁻¹⁵⁰), confirming accelerated red cell lysis.



Lactate Dehydrogenase (LDH)

LDH levels surged in PRBCs from 154.7 ± 49.8 to 1269.1 ± 392.5 U/L (F = 201.978, p = 9.30 × 10⁻⁸⁵), reflecting haemolysis.

Whole blood showed similar rise from 320.9 ± 116.1 to 1379.7 ± 365.8 U/L (F = 115.064, p = 1.10 × 10⁻⁶¹), confirming stored cell breakdown.



Repeated-Measures ANOVA

Parameter	Group	df ₁	df ₂	F	p-value	Interpretation (p < 0.05)
Hb	PRBC	5	245	15.091	6.41 × 10 ⁻¹³	Significant fall with storage
	Whole blood	5	245	0.560	0.731	No significant trend
RBC count	PRBC	5	245	16.387	6.04 × 10 ⁻¹⁴	Significant fall
	Whole blood	5	245	2.462	0.034	Mild but sig. fall
PCV / HCT	PRBC	5	245	5.852	3.97 × 10 ⁻⁵	Progressive rise
	Whole blood	5	245	5.654	5.99 × 10 ⁻⁵	Progressive rise
MCV	PRBC	5	245	12.938	3.54 × 10 ⁻¹¹	Cells swell over time
	Whole blood	5	245	68.142	4.06 × 10 ⁻⁴⁴	Very large rise
MCH	PRBC	5	245	2.289	0.0466	Marginal rise
	Whole blood	5	245	61.992	2.62 × 10 ⁻⁴¹	Marked rise
MCHC	PRBC	5	245	7.354	1.92 × 10 ⁻⁶	Falls significantly
	Whole blood	5	245	3.624	0.0035	Small but sig. wobble
Na ⁺	PRBC	5	245	18.086	2.90 × 10 ⁻¹⁵	Steady decline
	Whole blood	5	245	90.664	2.76 × 10 ⁻⁵³	Steep decline
K ⁺	PRBC	5	245	13.748	7.73 × 10 ⁻¹²	Sharp rise
	Whole blood	5	245	852.909	1.43 × 10 ⁻¹⁵⁰	Very sharp rise
LDH	PRBC	5	245	201.978	9.30 × 10 ⁻⁸⁵	Massive rise (haemolysis)
	Whole blood	5	245	115.064	1.10 × 10 ⁻⁶¹	Massive rise

df_1 (numerator) = number of time-points - 1
(6 - 1 = 5);

df_2 (denominator) = (n - 1) × df_1 → PRBC: n = 50
→ (50 - 1) × 5 = 245

Whole blood: n = 50 → (50 - 1) × 5 = 245

F compares between-day variance to within-subject variance; the smaller the p-value, the stronger the evidence that the parameter truly changes during storage.

Discussion

Blood storage plays a pivotal role in modern transfusion medicine, especially in settings where blood demand often exceeds immediate availability. However, storage-related changes, termed “storage lesions,” can significantly affect the quality and safety of blood products over time. The present study evaluated both haematological and biochemical parameters in whole blood and packed red blood cells (PRBCs) stored for up to 35 days in CPDA-1 solution at 2–6°C, a common practice in blood banks across India.

Haematological Parameters

Haemoglobin and RBC Count

In the current study, PRBC haemoglobin levels declined significantly from 17.25 g/dL on Day 0 to 14.56 g/dL on Day 35 ($p < 0.05$), indicating degradation of haemoglobin over time. In contrast, whole blood haemoglobin showed a slight increase from 12.52 to 13.52 g/dL. This could be attributed to plasma water evaporation or redistribution of RBCs during storage, which falsely elevates Hb concentration in whole blood units^[8]. These findings are consistent with Chaurasiya et al., who observed similar trends of haemoglobin reduction in PRBCs due to oxidative damage and hemolysis during prolonged storage^[8].

RBC counts in PRBCs decreased progressively (5.9988 → 4.9718 million/ μ L), consistent with the findings of Marabi et al., who reported a decline due to membrane fragility and hemolysis^[10]. In contrast, whole blood RBC counts remained relatively stable (3.80 → 3.93 million/ μ L), suggesting that leukocytes and platelets may help preserve RBC integrity

initially, though their utility declines over time and align with the findings with Gupta et al^[11].

Hematocrit (PCV), MCV, MCH, and MCHC

The hematocrit in PRBCs declined significantly from 56.5% to 50.4%, indicating RBC sedimentation and membrane loss, a well-documented effect during storage^[5]. Interestingly, whole blood hematocrit increased from 33.84% to 36.12%, possibly due to plasma reduction and hemoconcentration—a pattern previously reported by Chhabra et al.^[12].

MCV in PRBCs increased from 94.54 to 100.56 fL, indicating cellular swelling, possibly due to impaired Na^+/K^+ ATPase function and increased intracellular sodium. Similar findings were reported by Oyet et al.^[7], who also observed increased MCV due to RBC fragility during storage. Whole blood MCV followed the same upward trend (85.22 → 90.68 fL), though less pronounced.

MCH and MCHC values in PRBCs slightly decreased over the storage period. MCH dropped from 29.04 to 28.76 pg, and MCHC from 30.34% to 28.86%. These changes reflect reduced haemoglobin content per cell and intracellular dehydration^[13]. Notably, whole blood MCHC remained unchanged at 33.86%, indicating better maintenance of haemoglobin saturation, likely due to the presence of plasma buffering capacity.

Biochemical Parameters

Sodium and Potassium

A significant decline in sodium levels was observed in both PRBCs (142.44 → 124.88 mmol/L) and whole blood (141.72 → 111.72 mmol/L). This drop is attributed to the influx of sodium into RBCs due to membrane pump failure, an effect commonly seen during prolonged hypothermic storage^[14].

Conversely, potassium levels increased markedly during the same period. In PRBCs, potassium rose from 2.66 to 28.59 mmol/L; in whole blood, from 2.69 to 24.24 mmol/L. This increase is a hallmark of storage lesions and is due to the efflux of intracellular potassium from erythrocytes into the extracellular medium as membrane integrity deteriorates^[15]. Mukherjee et al. and Parshuram et al. confirmed similar findings, warning that hyperkalemia could

pose a risk in neonatal and massive transfusion scenarios [6,16].

LDH Levels

LDH serves as an indirect marker of RBC lysis and cellular injury. In this study, LDH levels rose sharply in both PRBCs (154.68 → 1,269.06 U/L) and whole blood (320.94 → 1,379.72 U/L). This pattern clearly reflects ongoing hemolysis and membrane breakdown, supporting previous observations by Oyet et al. [7] and Chaurasiya et al. [8].

Higher LDH values in whole blood than in PRBCs may be explained by the presence of leukocytes and platelets, which also release LDH upon lysis. However, both product types clearly show that LDH increases linearly with time, reinforcing its role as a reliable indicator of blood deterioration [17].

Clinical Implications

The cumulative impact of these storage-related changes has significant clinical implications. Increased potassium and LDH levels suggest caution in using older units for vulnerable populations such as neonates, patients with renal impairment, or those requiring large-volume transfusions. Decreased haemoglobin, hematocrit, and RBC integrity may reduce the efficacy of oxygen delivery, especially in critically ill patients [6,16].

Furthermore, routine monitoring of potassium and LDH during storage could guide quality assurance protocols in blood banks. Adoption of leukoreduction, additive solutions (e.g., SAGM), or newer storage methods may help mitigate these changes [5].

Comparison with Other Studies

The results of the present study align well with findings from other Indian and international studies. Oyet et al. and Chaurasiya et al. reported similar patterns of electrolyte imbalance and hemolytic markers in PRBC and whole blood stored at 2–6°C [7,8]. However, slight variations in absolute values may result from differences in sample processing, anticoagulant type, storage bags, and analysis techniques.

Additionally, while most international guidelines permit storage of PRBCs for up to 42 days, emerging

evidence, including our data, suggests that qualitative deterioration becomes clinically relevant after Day 14–21, particularly for biochemical parameters like potassium and LDH [19,20].

Conclusion

The present study systematically evaluated the haematological and biochemical changes occurring in stored whole blood and packed red blood cells (PRBCs) over a 35-day period at a tertiary care blood center. The findings highlight that storage-induced alterations—commonly referred to as storage lesions—are both predictable and significant.

Haematologically, PRBCs demonstrated a steady decline in haemoglobin concentration, RBC count, and hematocrit, while MCV increased due to cellular swelling. MCH and MCHC showed modest declines, reflecting intracellular haemoglobin loss and membrane fragility. In contrast, whole blood samples exhibited a slight rise in haemoglobin and hematocrit, likely due to plasma loss, while MCHC remained relatively unchanged.

Biochemically, both PRBC and whole blood units showed a progressive and significant increase in extracellular potassium and LDH levels—indicative of red cell lysis. Concurrently, sodium levels declined, reflecting ionic imbalance due to membrane pump failure. These changes became more pronounced beyond 14 days of storage, raising potential clinical concerns, particularly in vulnerable patient groups such as neonates, those with renal impairment, or individuals requiring massive transfusions.

The study underscores the need for stringent monitoring of storage parameters and supports the use of fresher blood units when clinically feasible. Adoption of improved storage technologies and leukoreduction practices may further enhance transfusion safety and efficacy.

Conflict of interest: Nil

Source of funding: Nil

Ethical Clearance was taken IEC of Muzaffarnagar Medical College, Muzaffarnagar, Uttar Pradesh dated 20/03/2023 with approval number MM/IEC/2023/243.

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