# COMPARISON OF CHANGES DUE TO STORAGE IN RED CELL CONCENTRATE FROM AA, AS AND AC BLOOD DONORS IN UNIVERSITY OF ILORIN TEACHING HOSPITAL, NIGERIA.

**Olatunbosun LO,[1,2,6]* Muhibi MA,[2,3] Oke OT,[2] Shittu OA[1,4], Ademosun AA,[6] Olalere, FD,[1,6] Lawal, SA[4] Kareem MA[6], Azeez RT[6], Oyeniyi GM[7], Uthman RD,[6] Idris AO, [6] AbdulRaheem AA.[1]**

[1]Department of Haematology and Blood Transfusion, University of Ilorin Teaching Hospital, Ilorin, Nigeria.
[2]Department of Medical Laboratory Science, Faculty of Basic Health Sciences, Achievers University, Owo, Ondo State, Nigeria.
[3]Department of Medical Laboratory Science, Faculty of Applied Health Sciences, Edo University, Uzairue, Edo State.
[4]Department of Hematology, University of Ilorin, Ilorin, Nigeria.
[5]Department of Hematology and Blood transfusion, Obafemi Awolowo University Teaching Hospital Complex, Ile-Ife.
[6]Department of Medical Laboratory Science, Faculty of Health Sciences, Al-Hikmah University, Ilorin, Nigeria.
[7]Department of Medical Laboratory Science, Faculty of Basic Clinical Sciences, University of Ilorin, Nigeria.
Corresponding Author*: Email: deluy008@gmail.com., Phone: +2348030467510

## ABSTRACT

**Background:** Red blood cell concentrate (RCC) is an essential therapeutic intervention for patients with anaemia, anemic heart failure, or hematological disorders.

**Aim:** This study evaluated red cell indices, RBC enzymes, and membrane integrity in common haemoglobin variants among prospective blood donors before and after RCC preparation.

**Methods:** A cross-sectional descriptive study was conducted among 72 prospective blood donors at the University of Ilorin Teaching Hospital. Participants were profiled using structured questionnaires. Whole blood (pre-donation) and RCC (post-harvest) samples were analyzed for mean cell volume (MCV), mean cell hemoglobin (MCH), mean cell hemoglobin concentration (MCHC), Osmotic Fragility Test (OFT), Lactic Acid Dehydrogenase (LDH), Glucose-6-Phosphate Dehydrogenase (G6PD), and Phosphatidylserine (PS) across haemoglobin genotypes (AA, AS, AC) using hematology analyzers and ELISA-based assays. Data were analyzed descriptively, with significance set at $p < 0.05$.

**Results**: RCC showed increased concentrations of G6PD, LDH, PS, and red cell functional parameters relative to baseline. Between post-harvest and pre-transfusion intervals, G6PD, LDH, and PS rose significantly, while OFT decreased significantly. Over 72-hour storage, G6PD, LDH, and PS progressively increased, though not significantly. Among hemoglobin variants, AA genotype exhibited superior red cell function compared to AC and AS. G6PD was highest in AA ($0.53 \pm 0.29$), while LDH was lower than in AS and AC. PS concentration peaked in AC ($p = 0.028$), and osmotic fragility was highest in AS, though not significant.

**Conclusion:** The study reveals early onset of red cell lesions during RCC processing, worsening after 72 hours of storage, marked by increased enzyme activity and reduced membrane integrity. AA genotype donors demonstrated superior RCC quality, suggesting their blood offers optimal transfusion safety and stability.

**Keywords**: Red Cell, Membrane, Enzymes, Genotype, pH, Concentrates

## INTRODUCTION

Red blood cell (RBC) concentrates transfusion is a life-saving treatment for patients suffering severe blood loss or anemia due to trauma injury, surgery, hemorrhage, hematological disease or malignancy [1]. In recent years, the need for stricter control over the quality of blood and its products has been emphasized. One such quality indicator for stored red cell units is the extent of hemolysis. Detecting excess hemolysis due to component processing and storage has important

implications for the transfused patient. Hemolysis, a critical indicator of red cell concentrates quality, can lead to adverse transfusion reactions, including hemoglobinemia, jaundice, and even life-threatening complications [2]. Current quality control measures may not adequately detect suboptimal red cell concentrates, potentially compromising patient outcomes. The most common red blood cell enzyme disorders are characterized by hemolysis but with wide clinical variability. Glucose-6-phosphate dehydrogenase deficiency and lactate dehydrogenase are the most common red cell enzymes disorder assessed in prospective blood donors [3]. Glucose-6-phosphate dehydrogenase deficiency is a genetic disorder that affects red blood cells. The most common medical problem associated with glucose-6-phosphate dehydrogenase deficiency is hemolytic anemia [4], which occurs when red blood cells are destroyed faster than the body can replace them. This type of anemia leads to paleness, yellowing of the skin, the eyes (jaundice), dark urine, fatigue, shortness of breath, and a rapid heart rate [5]. Lactic acid dehydrogenase (LDH) is abundant in RBCs and its level in plasma can function as another marker of red cell haemolysis in stored blood [6]. It has been proposed that red cells with exposed phosphatidylserine (PS) are more likely to be ingested by macrophages and that the exposure of PS signals the removal of aged red cells from the circulation [7]. Externalized phosphatidylserine (PS) is a sensitive marker for study to show ineffective packed cell transfusion and reasons to be included. As suggested by the currently available data on RBC aging in vivo and in vitro, on the response of RBC to various stress treatments in vitro, and on RBC in pathological conditions. The number of PS-exposing RBC increases with storage in the blood bank [8,9,10]. The finding has not been fully addressed to take into account the donor variability (genetics and lifestyle). Phosphatidylserine (PS) normally localizes to the inner leaflet of cell membranes but becomes exposed in abnormal or apoptotic cells, signaling macrophages to ingest them. Along similar lines, it seemed possible that the removal of red cells from circulation because of normal aging or in hemolytic anemias might be triggered by PS exposure. Likewise, the osmotic fragility test (OFT) is used to measure erythrocyte resistance to hemolysis while being exposed to varying levels of dilution of a saline solution [11]. When erythrocytes are exposed to a hypotonic environment, water enters the cell and causes swelling and eventual lysis [12,13]. The susceptibility of osmotic lysis of erythrocytes is a function of surface area to volume ratio. The exposure of phosphatidylserine (PS) and osmotic fragility are essential characteristics that influence the quality and viability of red cell concentrates (RCCs) during storage [8]. However, assessment of these factors is restricted in most blood banks. Furthermore, existing assessment methods for these factors are restricted, resulting in variable outcomes and possible complications related to transfusions. It is important to thoroughly evaluate these parameters in RCCs with a focus and emphasis on the safety and effectiveness of red cell concentrates transfusions. To best gain knowledge on donor variability and processes involved in preparing packed cells.

## MATERIALS AND METHODS:

This study is descriptive experimental research conducted in University of Ilorin Teaching Hospital (UITH). A total of 72 subjects were recruited as prospective blood donors for RCC concentrate and were assessed pre-donation, post-harvest and pre-transfusion for the quality of RCC concentrate. Ethical clearance was obtained from the Health Research Ethics Committee, Ministry of Health, Ilorin and Ethical committee, University of Ilorin Teaching Hospital, Ilorin. A 4mL whole blood was collected into EDTA for red cell functional parameters, genotype assay, G6PD, LDH, OFT and PS assays. A 1.5mL of RCC collected from blood bag into cryovial bottle at interval of 72hrs for the same aforementioned parameters.

## PREPARATION OF RED CELL CONCENTRATES

Whole blood was collected from a fit prospective blood donor into a sterile plasticized polyvinyl chloride (PVC) plastic double blood bag.  The CPDA-1 anticoagulated whole

blood was centrifuged at a speed of 2,700 g (soft spin) using Biobase BKC-BB6 JINAN Biobase Biotech Blood Bag Cold centrifuge for 10 minutes to separate the blood components into distinct layers, red blood cells (RBCs) at the bottom, White Blood Cells & platelet-rich plasma (PRP) at the top. The plasma of about 100 mL was carefully removed from the primary collection bag into satellite bag using plasma extractor leaving the RBCs and buffy coat behind of about 300 mL. The prepared red cell concentrates were stored at 6°C in a blood bank refrigerator.

## HAEMOGLOBIN ELECTROPHORESIS

Whole blood mixed with hemolysate solution was applied on the membrane cellulose acetate paper gently.  The electrophoresis tank containing buffer solution at alkaline pH was covered appropriately and power source was switched on to run at 450V for 15minutes. Migration of molecules depends on their molecular weight and electrical charge. Hb A has the highest amount of negative charges and thus the fastest migration. Hb S has a low molecular weight and lower negative charge and thus second fastest migration, while Hb C has a very low molecular weight and thus slow migration.

## ESTIMATION OF GLUCOSE-6-PHOPHATE DEHYDROGENASE (G6PD) AND ESTIMATION OF PHOSPHATIDYLSERINE (ELISA, Elabscience, 2024)

A volume of 100ul of standard and sample was added to each well and was incubated for 90 minutes at 37°C. Liquid from each well was decanted and immediately 100ul of biotinylated Ag/Ab detection was added and incubated for 1hour at 37°C. Solution from each well was decanted and 350ul of wash buffer was added and washed 3 times. A volume of 100ul of HRP conjugate was added to each well and incubated for 30min at 37°C. It was aspirated and washed 5 times. A volume of 90ul of substrate reagent was added and incubated for 15minutes at 37°C. A volume of 50ul of stop solution was added to each well and optical density of each well was determined at once with a micro plate reader set at 450nm.

## ESTIMATION OF LACTATE DEHYDROGENASE (LDH) (Colorimetric)

The reaction mixture was prepared by mixture of 950 µL tris buffer plus 50 µl of lactate substrate, and 50 µl $NAD^+$. 50 µl of serum was added to the reaction mixture, mixed well and incubated at 37°c for five minutes. Absorbance was measured 340nm every minute for five minutes. LDH catalyzes the conversion of lactate dehydrogenase to pyruvate, reducing NAD+ to NADH. The rate of NADH production is directly proportional to LDH activity.

## OSMOTIC FRAGILITY TEST

Twelve test tubes were arranged containing varying concentrations of 9.0, 7.5, 6.5, 6.0, 5.5, 5.0, 4.0, 3.5, 3.0, 2.0, 1.0 NaCl stock solution (8.5%) leaving the 12th tube as the blank (5.0ml of water). Fifty microliter of well-mixed blood was added to each tube and mixed immediately by inverting the tubes several times and foam was avoided. The suspensions were left for thirty minutes at room temperature. They were remixed and then centrifuged for five minutes at 1200 rpm.

## RESULTS

A total of 71 adult males and female prospective blood donors for the red cell concentrate were recruited with their clinicodemographic data recorded. Red cell functional indices, haemoglobin genotype, G6PD, LDH, and OFT were evaluated at baseline, post-red cell concentrate preparation and during storage presented in tables as shown below:

**Table 1:** Socio-demographic characteristics of prospective donors for blood products

| Parameters | Mean | Frequency | Percent (%) | Parameters | Frequency | Percent (%) |
|---|---|---|---|---|---|---|
| **AGE** | 34.75 | | | **EDUCATION** | | |
| <=30 | | 24 | 33.8 | Secondary | 25 | 35.2 |
| 31-40 | | 29 | 40.8 | Tertiary | 46 | 64.8 |
| > 40 | | 18 | 25.4 | **RELIGION** | | |
| **SEX** | | | | Christianity | 68 | 95.8 |
| Male | | 68 | 95.8 | Islam | 3 | 14 |
| Female | | 3 | 4.2 | **TRIBE** | | |
| **MARITAL** | | | | Yoruba | 65 | 91.5 |
| Single | | 29 | 40.8 | Others | 6 | 8.5 |
| Married | | 42 | 59.2 | **DAILY STRESS** | | |
| **OCCUPATION** | | | | No Stress | 21 | 29.6 |
| Civil Servant | | 14 | 19.7 | Rarely | 44 | 62.0 |
| Trader | | 11 | 15.5 | Often | 6 | 8.5 |
| Student | | 9 | 12.7 | **CIGARETTE** | | |
| Unemployed | | 37 | 52.2 | Yes | 3 | 4.2 |
| **SLEEP** | | | | No | 68 | 95.8 |
| >6 hours daily | | 52 | 73.2 | **ALCOHOLISM** | | |
| < 5 hours daily | | 14 | 19.7 | Yes | 10 | 14.1 |
| < 4 hours daily | | 5 | 7.0 | No | 61 | 85.9 |

A total of 71 adult males (95.8%) and female (4.2%) prospective blood donors for blood products were recruited with mean age of 34.8 where 40.8% were between the age range of 31-40 and 33.8% were ages below 30 years.    High percentage of the participants were married (59.2%), unemployed (52.2%), acquired Tertiary education (64.8%), Christianity by faith (95.8%) and Yoruba by tribe (91.5%). Principally, 62.0% rarely experienced stress in their daily activities, they are non-cigarette smokers (**95.8%**), non-alcohol drinkers (**85.9**) and usually with sleep pattern of more than 6 hours per day (73.2) on routine basis.

**Table 2**: Clinical Demographic Variables of Prospective Blood Donors for Blood Products

| Parameters | Frequency | Percent (%) | Parameters | Frequency | Percent (%) |
|---|---|---|---|---|---|
| **CATEGORIES** | | | **REASONS FOR DONATION** | | |
| First time donors | 21 | 29.6 | Relative | 42 | 59.2 |
| Second time donor | 13 | 18.3 | Voluntary non-remunerated | 29 | 40.8 |
| Third Time donor | 3 | 4.2 | **ANALGESIC** | | |
| Regular Donors | 34 | 47.9 | Regular | 3 | 4.2 |
| **FREQUENCY** | | | Occasional | 68 | 95.4 |
| 1-2 Years | 58 | 81.7 | **HEMATINIC** | | |
| 3-4 Years | 10 | 14.1 | Regular | 13 | 81.3 |
| More than 4 Years | 3 | 4.2 | Occasional | 3 | 18.8 |

**Table 2** above shows clinico-demographic characteristic of the donation pattern where 47.9% were regular donors for blood products, 29.6% were first timer, 18.3% were second timer and 4.2% were third timer prospective donors. Majority of the donors donate at maximum of twice a year (81.7%), relative donors (59.2), occasionally use analgesic (95.4%) and regularly use hematinics (81.3%).

**Table 3**: Mean difference in red cell functional indices at baseline, post-harvest (red cell concentrate) and pre-transfusion of the concentrate.

| Variables | Mean ± SD | | | | |
|---|---|---|---|---|---|
| | **Pre-donation** | **Post-Harvest** | **Pre-transfusion** | **F-test** | **p-value** |
| PCV (%) | $41.28 \pm 2.06$ | $42.63 \pm 1.87$ | $35.23 \pm 5.12$ | 97.449 | 0.001 |
| HB   (g/dl) | $14.16 \pm 0.81$ | $15.21 \pm 0.41$ | $12.77 \pm 1.84$ | 75.489 | 0.001 |
| RBC X $10^{12}$/L | $4.39 \pm 0.32$ | $4.37 \pm 0.15$ | $3.93 \pm 0.68$ | 23.828 | 0.001 |
| MCV   fl | $92.47 \pm 4.29$ | $100.35 \pm 2.32$ | $92.34 \pm 6.20$ | 59.861 | 0.001 |
| MCH   pg | $32.29 \pm 1.85$ | $37.91 \pm 1.33$ | $32.65 \pm 2.53$ | 181.438 | 0.001 |
| MCHC   g/dl | $34.31 \pm 1.09$ | $37.83 \pm 1.01$ | $35.38 \pm 1.79$ | 127.860 | 0.001 |

**Table 3**: There's an increase in red cell functional indices from the pre-donation to the post-harvest. However, the value declined at pre-transfusion relative to the baseline with a mean difference that was statistically significant (p value = 0.001).

**Table 4**: Pattern of red cell enzymes concentration at baseline, post-harvest (red cell concentrate) and pre-transfusion of the concentrate

| Variables | Mean ± SD |
|---|---|
| | |

|  | Pre donation | Post Harvest | Pretransfusion | F-test | p-value |
|---|---|---|---|---|---|
| LDH **(ng/mL)** | 129.93 ± 22.45 | 277.48 ± 60.90 | 341.59± 48.97 | 379.556 | 0.001 |
| G6PD (ng/mL) | 0.51 ± 0.28 | 0.35 ± 0.21 | 0.22 ± 0.12 | 32.002 | 0.001 |
| PS (ng/mL) | 0.09 ± 0.03 | 0.09 ± 0.03 | 0.16 ± 0.03 | 135.474 | 0.001 |
| OFT **(%)** | 3.45 ± 1.17 | 4.27 ± 1.50 | 5.37 ± 1.25 | 38.202 | 0.001 |

**Table 4**: There was significant increase in LDH, PS and OFT concentration across the 3 stages of evaluation relative to the baseline. Conversely, G6PD shows a consistently low or decreased concentration.

**Table 5**: Mean difference in parameter based on haemoglobin genotype

| Genotype | Mean ± SD | | | | |
|---|---|---|---|---|---|
|  | **A A** | **A C** | **A S** | **F-test** | **p-value** |
| PCV (%) | 41.40 ± 1.09 | 41.20 ± 0.00 | 41.31± 2.22 | 0.045 | 0.956 |
| HB (g/dl) | 14.22 ± 0.87 | 13.86 ± 0.00 | 14.10 ± 0.00 | 0.821 | 0.444 |
| RBC X $10^{12}$/L | 4.46 ± 0.00 | 4.39 ± 0.34 | 4.36 ± 0.21 | 0.082 | 0.922 |
| MCV fl | 94.32 ± 4.31 | 92.40 ± 0.00 | 94.38 ± 4.77 | 0.193 | 0.825 |
| MCH pg | 32.39 ± 1.92 | 31.70 ± 0.00 | 31.82 ± 1.58 | 0.508 | 0.604 |
| MCHC g/dl | 34.41 ± 1.14 | 34.30 ± 0.00 | 33.72 ± 0.59 | 1.742 | 0.183 |

**Table 5** above shows red cell functional parameters with mean standard deviation almost the same across the various hemoglobin genotype (AA, AC, AS). However, hemoglobin value for AS (13.86-+ 0.34) was lower when compared with AA (14.22± 0.87) and AC (14.10 ± 0.00) although the mean difference was not statistically significant. Interestingly, hemoglobin genotype AS has the highest MCV value (94.38 -+ 4.77) while hemoglobin AA has the highest MCH and MCHC value of 32.39 ± 1.92 and 34.41 ± 1.14 respectively.

**Table 6**: Pattern of red cell enzyme polymorphism in hemoglobin variants

| Genotype | | | **Mean ± SD** | | |
|---|---|---|---|---|---|
| | AS | AC | AA | F-test | p-value |
| LDH **(ng/mL)** | 129.20 ± 23.77 | 136.00 ± 14.67 | 121.00 ± 0.00 | 0.548 | 0.581 |
| G6PD **(ng/mL)** | 0.40 ± 0.20 | 0.17 ± 0.00 | 0.53 ± 0.29 | 2.461 | 0.093 |
| PS   **(ng/mL)** | 0.09 ± 0.03 | 0.11 ± 0.00 | 0.07 ± 0.02 | 3.765 | **0.028*** |
| OFT   **(%)** | 4.25 ± 1.41 | 4.00 ± 0.00 | 4.40 ± 2.14 | 0.071 | 0.932 |

**Table 6** shows highest LDH concentration in AC individual (136.00 ± 14.67) followed by AS individual (129.20 ± 23.77), and lowest value of LDH was found in AA individual (121.00 ± 0.00) with a mean difference that was not statistically significant. G6PD value was highest in AA individual at mean value of 0.53 ± 0.29 followed by AS individual (0.40± 0.20) and the lowest in AC individual (0.17± 0.00). The PS was found to be the highest in hemoglobin AC genotype (0.11 ± 0.00) with lowest value in hemoglobin AA (0.07 ± 0.02) and intermediate value in AS (0.09 ± 0.03) the mean value across the hemoglobin genotype was statistically significant with p value (p = 0.028). Furthermore, hemoglobin AA (4.40 ± 2.14) had an OFT cut off mean value higher than AC (4.00 ± 0.00) and AS (4.25 ± 1.41) individual, interestingly the mean difference is not statistically significant.

**Table 7**: Pattern of red cell enzyme LDH, G6PD, PS and OFT at 72 hours interval of 4°C storage

| **Variables** | **Mean ± SD** | | | | | |
|---|---|---|---|---|---|---|
| | **1** | **2** | **3** | **4** | **F-test** | **p-value** |
| LDH **(ng/mL)** | 120.00 ± 8.76 | 120.00 ± 8.76 | 125.00 ± 32.48 | 133.09 ± 18.33 | 1.507 | 0.229 |
| G6PD **(ng/mL)** | 0.50 ± 0.23 | 0.50 ± 0.29 | 0.53 ± 0.30 | 0.59 ± 0.35 | 0.301 | 0.741 |
| PS **(ng/mL)** | 0.08 ± 0.03 | 0.08 ± 0.03 | 0.09 ± 0.03 | 0.12 ± 0.02 | 3.443 | **0.038*** |
| OFT **(%)** | 4.00 ± 0.00 | 4.00 ± 0.00 | 4.27 ± 0.81 | 4.29 ± 1.78 | 0.102 | 0.903 |

**Table 7** shows pattern of changes in G6PD and LDH concentration longitudinally from 72 hours interval assessment to 14 days at 4°C storage. There was a progressive increase that was statistically not significant at first two rounds of 72 hours. Phosphatidylserine concentration longitudinally from seventy-two hours of interval assessment to fourteen days at 4°C storage assessment increases that was statistically significant (p = 0.038), whereas OFT increase was not statistically.

**DISCUSSION**

Transfusion of RBCs is indicated to restore the oxygen-carrying capacity in patients with severe anemia or major blood loss [14, 15]. Factors influencing blood quality have been explored in several studies among which is donor-related characteristics especially in heterogeneous groups blood donors

with a wide range of biological differences. In this study, out of a total of 72 participants as red cell concentrate blood donors, majority were adult males (95.8%) with mean age of 34.8, married (59.2%), unemployed (52.2%) and had a Tertiary education (64.8%). Interestingly in a Muslim dominated area, majority of the participants were Christian by faith (95.8%) and Yoruba by tribe (91.5%). Similarly, the quality of life the participants live reflected in the number of hours they sleep per day and good social life of non- alcoholic and non-cigarette and no addiction to analgesic. In this study, the functional red cell parameters evaluated were within the reference range in pre-donation samples which depict high quality donors were recruited. It was also found that, the level of these parameters was slightly increased in post-harvest samples, which may be possibly due to anticoagulant to blood ratio and the post -donation treatment as red cell concentrates [16]. Meanwhile these parameters were decreased significantly during storage period before issued for transfusion. This observation shows that, the red cell functional parameters tend to decrease based on the storage time and condition. Previous study by Jadison *et al.,* 2021 have reported similar observation while Leung and colleagues [17] study differ slightly in their outcome. This difference may be probably due to the longevity period of the storage of the red cell concentrates or the study participants genetic variability [18,19]. In this study, the level of lactate dehydrogenase (LDH) and phosphatidylserine (PS) investigated were found to increase significantly. This implies that, there was a change in the level of LDH and PS during processing and storage due to possible red cell increased metabolism and enhanced glycolytic activity during processing and storage. This observation was consistent with the report of previous studies and could be ascribed to susceptibility of red blood cells to stress during processing and storage which could lead to increase hemolysis and vesicle concentration in red blood cell concentrates Adsatong *et al.,* [20, 21]. Additionally, the concentration of Glucose-6-dehydrogenase enzyme (G6PD) and OFT were found to decrease significantly in RCC and during storage before transfusion relative to the baseline value. This indicates that, the enzyme activity and membrane integrity are lost probably due to processing stress and storage condition. The result of this current study is in line with the report of Klein and colleagues [22, 23]. Paradoxically, the report of this current study was not in agreement with the findings of Walsh and colleagues [24] which found a normal level of osmotic fragility of red cell in pre-donated and stored red cell concentrates sample. This variation maybe due to difference in clinical condition of the participants, genetic factor, and processing method of the sample.

In this study, the donor's variability based on Hb genotype was investigated and found that the Packed Cell Volume was found to be same in value across the three major hemoglobin genotypes (AA, AS, and AC). Also, the hemoglobin concentration and red cell indices were found similar in both AA and AS individuals but slightly lower in AC individuals while the total red cell count was found similar in AC and AS individuals but slightly higher in AA individuals with no statistical significance. Additionally, red cell enzyme activities vary across the haemoglobin genotype variants. The levels of LDH and G6PD were noted to increase among the AC and AS individuals respectively with mean difference that was not statistically significant. While phosphatidylserine and OFT were lower in AA individuals but highest in AC individuals which was statistically significant. The study of the effect of storage at $4^{\circ}C$ for 2 weeks with a repeated evaluation of the enzymes and membrane integrity at interval of 72 hours showed an increased value in PS and OFT progressively throughout the study period. This finding is in line with the previous studies reported by[8, 25] . This reflect that the longer the period of storage the higher the value of the enzyme with

clinically significant high value at fourth round, relating to possible hemolytic characteristics, creating the risk of hemolysis during transfusion of the red cell concentrates.

**CONCLUSION**:

The red cell indices, enzymes and membrane integrity evaluated favor AS haemoglobin genotype in terms of transfusion outcome compared to AA individuals while AC individuals had lowest value. Hence, high quality RCC favor AA genotype blood donors and generally short duration of storage for optimal transfusion outcome.

**REFERENCES**

1.     Pistorius AMA, Luten M, Bosman GJCGM, DeGrip WJ. A single assay for multiple storage-sensitive red blood cell characteristics by means of infrared spectroscopy. Transfusion (Paris) 2010;50(2):366–375.

2.     Tang H, Hung-yao H, Pei-ru W, et al. Inability to maintain GSH pool in G6PD-deficient red cells causes futile AMPK activation and irreversible metabolic disturbance. Antioxidants & redox signalin 2021;22(9):744–759.

3.     Luzzatto L, Ally M, Notaro R. Glucose-6-phosphate dehydrogenase deficiency. Blood 2020;136(11):1225–1240.

4.     Orlina AR, Aaron M. Josephson, Barbara T. McDonald. The poststorage viability of glucose-6-phosphate dehydrogenase-deficient erythrocytes. Journal of Laboratory and Clinical Medicine 2019;7(5):930–936.

5.     Tripathi P, Kumar R, Agarwal S. Spectrum and hematological profile of hereditary anemia in North Indians: SGPGI experience. Intractable Rare Dis Res 2018;7(4):258–263.

6.     Arif SH, Yadav N, Rehman S, Mehdi G. Study of Hemolysis During Storage of Blood in the Blood Bank of a Tertiary Health Care Centre. Indian J Hematol Blood Transfus 2017;33(4):598–602.

7.     Boas FE, Forman L, Beutler E. Phosphatidylserine exposure and red cell viability in red cell aging and in hemolytic anemia. Proceedings of the National Academy of Sciences 1998;95(6):3077–3081.

8.     Dinkla S, Peppelman M, Raadt J Van Der, et al. Phosphatidylserine exposure on stored red blood cells as a parameter for donor-dependent variation in product quality. Blood Transfus 2014;12(2):204–9.

9.   Verhoeven AJ, Hilarius PM, Dekkers DWC, Lagerberg JWM, Korte D De. Prolonged storage of red blood cells affects aminophospholipid translocase activity. Vox Sang 2006;91(3):244–251.

10.  Bosman GJCGM, Cluitmans JCA, Groenen YAM, Werre JM, Willekens FLA, Novotný VMJ. Susceptibility to hyperosmotic stress–induced phosphatidylserine exposure increases during red blood cell storage. Transfusion (Paris) 2011;51(5):1072–1078.

11.  Parpart AK, Philip B. Lorenz, Ethel R. Parpar, John R. Gregg, Aurin M. Chase. The osmotic resistance (fragility) of human red cells. he Journal of clinical investigation 1947;26(4):636–640.

12.  Luzzatto L, Ally M, Notaro R. Glucose-6-phosphate dehydrogenase deficiency. Blood 2020;136(11):1225–1240.

13.  Bosman GJCGM, Cluitmans JCA, Groenen YAM, Werre JM, Willekens FLA, Novotný VMJ. Susceptibility to hyperosmotic stress–induced phosphatidylserine exposure increases during red blood cell storage. Transfusion (Paris) 2011;51(5):1072–1078.

14.  Pistorius AMA, Luten M, Bosman GJCGM, DeGrip WJ. A single assay for multiple storage-sensitive red blood cell characteristics by means of infrared spectroscopy. Transfusion (Paris) 2010;50(2):366–375.

15.  Tzounakas VL, Kriebardis AG, Georgatzakou HT, et al. Data on how several physiological parameters of stored red blood cells are similar in glucose 6-phosphate dehydrogenase deficient and sufficient donors. Data Brief 2016;8:618–627.

16.  Chen Z, Herzog RW, Kaufman RJ. Cellular stress and coagulation factor production: when more is not necessarily better. Journal of Thrombosis and Haemostasis [homepage on the Internet] 2023 [cited 2025 Apr 20];21(12):3329–3341. Available from: https://www.jthjournal.org/action/showFullText?pii=S1538783623007705

17.  Leung P, Cannon EP, Petrikovics I, Hawkins A, Way JL. In vivo studies on rhodanese encapsulation in mouse carrier erythrocytes. Toxicol Appl Pharmacol 1991;110(2):268–274.

18.  Verhoeven AJ, Hilarius PM, Dekkers DWC, Lagerberg JWM, Korte D De. Prolonged storage of red blood cells affects aminophospholipid translocase activity. Vox Sang 2006;91(3):244–251.

19.  Tang H, Hung-yao H, Pei-ru W, et al. Inability to maintain GSH pool in G6PD-deficient red cells causes futile AMPK activation and irreversible metabolic disturbance. Antioxidants & redox signalin 2021;22(9):744–759.

20.  Tzounakas VL, Kriebardis AG, Georgatzakou HT, et al. Data on how several physiological parameters of stored red blood cells are similar in glucose 6-phosphate dehydrogenase deficient and sufficient donors. Data Brief 2016;8:618–627.

21.    Mercer RM, Corcoran JP, Porcel JM, Rahman NM, Psallidas I. Interpreting pleural fluid results. Clinical Medicine 2019;19(3):213–217.

22.    Nkhoma ET, Poole C, Vannappagari V, Hall SA, Beutler E. The global prevalence of glucose-6-phosphate dehydrogenase deficiency: A systematic review and meta-analysis. Blood Cells Mol Dis 2009;42(3):267–278.

23.    Klein HG, David J. Anstee. Mollison's blood transfusion in clinical medicine. Blackwell publishing 2013;11:1–27.

24.    Walsh TS, McArdle F, McLellan SA, et al. Does the storage time of transfused red blood cells influence regional or global indexes of tissue oxygenation in anemic critically ill patients?*. Crit Care Med 2004;32(2):364–371.

25.    Dinkla S, Peppelman M, Raadt J Van Der, et al. Phosphatidylserine exposure on stored red blood cells as a parameter for donor-dependent variation in product quality. Blood Transfus 2014;12(2):204–9.