


Haematological changes in stored whole blood - An observational study in a tertiary hospital blood bank

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Introduction: Several studies are going on to study the effects of transfusing stored RBCs on recipients. Several haematological changes occur in stored blood which may have some medical effects on the recipients. The biochemical changes may vary from donor to donor as well as from Blood bank to blood bank. Also effects of transfused RBCs will vary from recipient to recipient. Lot of clinical trial should be taken before assessing the safety of stored RBCs. **Objectives:** To study the various haematological changes occurring in a stored whole blood. **Material and methods:** Analysis of hematological changes occurring in stored whole blood in 20 voluntary donars was done in a blood bank of a tertiary hospital. **Results:** Of the various haematological parameters studied only significant changes were seen in total WBC count, percentage of lymphocytes and polymorphs and MPV after storage. There was a significant decrease in Total WBC and polymorphs concentration values. **Conclusion:** Certain haematological changes do occur in stored whole blood over duration of storage. As the demand for blood is high due to various medical conditions; the treating consultant should be aware of the haematological changes seen in stored whole blood.

Keywords: Blood Bank, Stored Blood, Whole Blood

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Introduction

Preservation and long term storage of Red Blood Cells (RBCs) is needed to ensure a readily available, safe blood supply for transfusion medicine. Blood collection and storage systems licensed by the Food and Drug Administration allow red cells to be stored up to 42 days.

During storage, in fact, preserved blood cells undergo progressive structural and functional changes that may reduce red cell function and viability after transfusion [1].

Storage has a negative effect on RBC oxygen delivery [2]. Considerable evidence suggests that transfusion increases the risk of serious complications and death in critically ill patients, especially in patients who are undergoing cardiac surgery [3].

Clinical implications, collectively known as the RBC storage medium lesion, is in part related to bioreactive substances released by leucocytes in the storage medium, such as histamine, lipids, and cytokines, which may exert direct effect on metabolic and physical changes associated with the senescence, such as membrane reticulation, decrease in cell size, increase of cell density, alteration of cytoskeleton, enzymatic desilylation, and phosphatidylserine exposure, RBCs lose potassium 2,3- diphosphoglycerate (2,3-DPG), Adenosine Triphosphate (ATP) stores, lipids and membrane, while becoming more rigid and demonstrating reduced oxygen off-loading [4].

Platelets circulate longer when stored at room temperature and are more activated and able to form clot more effectively when stored at 4°C [5].

White cells lose their phagocytic property within 4-6 hrs of collection and become non-functional after 24 hrs of storage [6].

It is important to remember they do not lose their antigenic property and are capable of sensitizing the recipient to produce non-haemolytic febrile transfusion reactions. Few lymphocytes may remain viable even after 3 weeks of storage.

This work examines changes that occur in some hematological parameters of whole blood stored in blood bank in a resource limited setting with a view to assessing the possible duration of storage for maximal utility in blood transfusion.

Materials and Methods

This study was conducted in Bharati Vidyapeeth Deemed University Medical College and Hospital, Sangli which is a tertiary Hospital. Blood (450 ml) was drawn from twenty healthy volunteer donors into Citrate Phosphate. Dextrose Adenine (CPDA-1) anticoagulant has been added & placed on the quarantine shelf of the blood bank refrigerator.

The donors were 20 in number; they had their ages ranging from 20 to 28 years (mean age 25.2 yrs). The donors were all male and tested negative for: HCV, HbsAg, Syphilis and HIV 1 & 2. The blood was kept for 28 days and samples were evaluated on days 1, 7, 14, 21 and 28. Blood bag of 450 ml which contains CPDA-1 was used. Most blood collection bags (adult) contain 63 ml CPDA anticoagulant which is sufficient to anticoagulated and ensure the viability of blood cells in 450 ml \pm 10% blood for up to 28-35 days when the blood is stored at 2- 8°C [1,2,3].

Blood bags were carefully stored in a quarantine shelf in the blood bank, with temperature ranging from 2-6°C. These haematological parameters were measured using the CellItac E, fully auto 5 part differential Hematology Analyzer by Nihon Kinden. It enumerates 17 parameters with 5-part differentiation of WBC.

Results

Mean values of hematological parameters: At the end of the study period as shown in (Table 1), the mean values of some hematological parameters were as follows: WBC (2.04×10^3 /ul), Hgb (14.04 g/dl), PCV (37.80 %), Lymphocytes (2.722×10^3 /uL), Polymorphs (2.002×10^3 /uL), Monocytes (0.6262×10^3 /uL), Eosinophils (0.29×10^3 /uL), Basophils (0.144×10^3 /uL), MCHC (34.52 g/dl), MCV (84.47 fl), MCH (30.20 pg), Platelet (153.2×10^3 /uL).

Analysis of variance showed that at the end of the study period, significant differences were noted in 5 parameters out of 20 parameters evaluated, as shown in (Table 1) WBC (F= 7.056, P<0.05), Differential lymphocytes (F=2.456, P< P>0.0001), absolute and differential polymorphs (F= 53.40, P<0.0001 and F=82.01, P<0.0001), MPV (F=6.41, P<0.0001). The lymphocyte percentage and MPV is increased while the percentage of polymorphs and Total leucocyte count is decreased during storage.

Table 1: Mean test of significance for haematological parameters for all the days

Parameters	Day 1	Day 7	Day 14	Day 21	Day 28	Total	F value	P value
WBC X 10 ³ / ul	7.59	5.80	4.41	3.21	2.57	2.04	7.056	0.000***
Lym x 10 ³ / ul	3.20	2.95	2.74	2.48	2.24	2.722	2.456	0.053*
Lym %	30	47	61	63	81	56.4	10.71	0.000***
Poly X 10 ³ / ul	5.66	2.21	1.28	0.63	0.23	2.002	53.40	0.000***
Poly %	59	43	30	19	12	32.6	82.01	0.000***
Eosino X10 ³ / ul	0.36	0.31	0.29	0.26	0.23	0.29	1.06	0.116 ns
Eosino %	04	04	03	03	02	3.20	1.080	0.320 ns
Mono X10 ³ / ul	0.73	0.69	0.64	0.56	0.51	0.626	1.12	0.120 ns
Mono %	07	07	06	06	05	6.2	1.09	0.356 ns
BAso X 10 ³ / ul	0.19	0.15	0.14	0.12	0.12	0.144	1.34	0.124 ns
Baso %	01	01	01	00	00	0.60	1.15	0.368 ns
RBC X 10 ⁶ / ul	4.87	5.75	6.61	7.35	8.11	6.538	1.090	0.369 ns
HGB gm/dl	13.6	14.2	14.3	14.1	14.0	14.04	0.182	0.945 ns
HCT %	37.89	36.55	38.18	37.89	38.52	37.80	1.246	0.345 ns
MCV fl	82.15	83.90	84.12	86.01	86.19	84.47	0.589	0.645 ns
MCHC g/dl	34.40	34.44	34.52	34.61	34.66	34.52	0.939	0.486 ns
MCH pg	30.35	30.27	30.23	30.16	30.00	30.20	0.415	0.790 ns
RDW %	12.84	14.00	14.38	14.76	15.02	14.2	6.130	0.001***
PLT x 10 ³ / ul	245	173	138	114	96	153.2	1.635	0.199 ns
MPV fl	7.97	8.47	8.68	8.88	9.17	43.17	6.41	0.000***

Discussion

Certain changes occur in some haematological parameters of stored whole blood. Some of the parameters are increased as compared to their initial values on day of collection while some are decreased. This is comparable with the work done by Cohl et al [5].

When the mean values of WBC on day 1 was compared to day 7, it was observed that there was rapid deterioration in granulocytes WBC. The mechanism of leucocyte depletion during whole blood storage may include loss of cell viability due to ATP depletion. Moreover, leucocytes are also consumed in the formation of micro-aggregates, which are conglomerates of leucocytes, platelets, fibrin, cold-insoluble globulin and cellular debris formed during storage [5, 7-9].

This study revealed a progressive fall in all types of leucocyte however, the pattern of changes observed in the serial differential count would suggest that

And monocytes. The clinical significance of this observation is that stored whole blood would be particularly ineffective as a clinical tool in the management of aplastic anaemia and other leucopenic patients since the most critical entity in these cases is almost always neutropenia.

Furthermore, this data revealed a specific survival advantage of lymphocytes in stored whole blood, which will imply that stored whole blood carries the risk of graft-versus-host disease if viable donor lymphocytes get engrafted in immuno-deficient recipients and premature neonates [9].

This is particularly important within the context of the current HIV pandemic, which is strongly associated with anaemia and frequent transfusions [10].

The platelets also revealed progressive decline in count, during the period of storage. In similarity to leucocytes, the fall in platelet levels may be related to loss of cell viability due to ATP depletion as well as platelet consumption due to microaggregates formation [5].

MPV showed drastic increase from day 14 down to day 28. This agrees with the work done by Cohl et al. [5], Due to depletion of red cell ATP there is gradual but steady fall in haematocrit during storage. A progressive increase in RDW was noticed in this study, after comparison of the mean cell value of day 1 and 14 and this agrees with the study of Cohl et al. [4].

Other hematological parameters remained fairly stable during this study, hence may be considered acceptable for clinical utility [11, 12].

Previous studies suggested that ultimate result due to various haematological and biochemical changes results in ATP depletion. [13, 14]. Therefore adenine is added to the anticoagulant CPDA -1, prolonging the duration of storage. [15, 16, 17, 18].

Conclusion

Certain haematological changes do occur in stored whole blood over a duration of storage. Rapid degeneration of leukocytes could lead to immunodulation related to blood transfusion. The answer to the question of whether there is a difference between fresh and old blood seems to be an unequivocal yes. The storage lesion is a clear process by which RBCs degrade over time.

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