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AN EXPERIMENTAL STUDY OF BLOOD COMPONENT PREPARATION AND ITS USES IN CLINICAL SPECIALTIES

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INTRODUCTION

The term transfusion literally means the proper selection and utilization of blood components and the removal of blood or blood components in the treatment of various medical diseases. The transfusions have increasing role in management of patients care. The bold components and blood derivatives has broadened the application of transfusion, the technical advancement, particularly the efficient use of components and fraction, recombinant products, AIDS epidemic have revolutionized concept of transfusion, however, the prudent used of blood product is governed by understanding of indication, limitation, potential benefits, and hazards of the transfusion (Henery 1996; Wintrobe's 1999). The blood transfusion has important role in modern practice of medicine and without it many surgical procedure, cardiac surgery, treatment various hematological disorder are impossible, due to limited resources of blood and increased demand there is now significant need of rational use of blood and its component. (Makroo et al.,1999).

Blood transfusion is an integral and indispensable part of the health care system. Without blood transfusion, effective management of severe trauma, major surgery, obstetrics complication is not possible. Furthermore, there is a key role for transfusion in the management of serious medical and pediatrics conditions such as gastrointestinal bleeding, cancer, leukemia, hemophilia, hemolytic anemia etc. (WHO AITBS 2004) Blood is a specialized living tissue that is composed of a liquid called blood plasma and blood cells suspended within the plasma & that circulates through the heart, arteries, veins, and capillaries carrying nourishment, electrolytes, hormones, vitamins, antibodies, heat, and oxygen to the body's tissues. (Bernice et al., 2002). Blood maybe transfused as whole blood or as one of its components. Because patients seldom require all of the components of whole bold, it makes sense to transfuse only that portion needed by the patient for a specific condition or disease. This treatment, referred to as "blood component therapy", allows several patients to benefit from one unit of donated whole blood. Blood components include red blood cells, plasma, platelets, and cryoprecipitatedantihemophilic factor (AHF) etc. up to four components may be derived from one unit of blood. (Makroo et al.,1999).

AIM AND OBJECTIVE

The study was carried out with the aim of emphasizing on the importance of component therapy in present clinical scenario with the objectives.

1. To determine the total number of blood units dispensed by RedAid Blood Centre, Khanna, Punjab during the period of study.
2. Percentage utilization of various blood components by clinical patients.
3. Average components unit utilization in various Specialties i.e. in surgery & Trauma, Medicine, Obstetrics/ Gynecology and Pediatrics.

MATERIAL AND METHODS

The present study is carried out to estimate the usage of various blood components (RBCs, PC & FFP) in different medical or surgical specialties (surgery & trauma, medicine, obstetrics & gynecology, pediatrics) at RedAid Blood Centre, Khanna, Punjab. 3000 cases of medical or surgical conditions requiring transfusion of components (RBCs, PC & FFP) formed the basis for selection in the study. All the relevant clinical information concerning age, sex, provisional diagnosis, indication for transfusion and the average number of units of various components dispensed per patients and specialties are analyzed**.** Criteria of selection of cases for the study are surgery, Medicine, Obstetrics/Gynecology, and Pediatrics.

Component Preparation

The procedure followed for components separation fusion testing is as per the guidelines as prescribed by (DGHS "Technical Manual", 2nd edition, 2003 & AABB Technical Manual, 15th edition, 2005).

SOP for preparation of Red Cells from the Whole Blood

Material Required

- Refrigerated centrifuge (Roto Silinta 630 RI/Rotixa 50 RS)
- Plasma expresser (Fresenius Hemocare)
- Tube stripper (Fresenius Hemocare)
- Bucket corrector, (Fresenius Insignia)
- Clamps
- Tube sealer (Fresenius Hemoseal)

PROCEDURE

General instructions

1. All the blood bags must be prepared within 6hrs of phlebotomy.
2. All the satellite bags must be accurately identified, numbered, labeled as coming from the original unit.
3. All blood bags are accurately balanced before centrifugation.
4. Contents in diagonally opposite bucket of the centrifuge must be equal in weight.
5. Plastic or weighted rubber discs and large rubber bands are used for balancing.
6. The balanced bucket is carefully placed diagonally opposite in refrigerated centrifuge.

Procedure for separation of red cells

1. The stabilizer switches on from the right side by lifting up the MCB then push the green button.
2. The centrifuge was switched as from the lower right side
3. Open the lid; press the open button on the upper right side.
4. Put the balanced bags in the centrifuge, close the lid and chose the program1 (1750rpm for 11 min at 22℃) for PRP. Chose program 3 (3947 rpm for 5min 30 sec at 4℃) for RBC and FFP only.
5. After centrifugation is over, it gives a beep, open the lid and arrange all the blood bags on Plasma Expresser.
6. Check whether the line separating red cells & plasma layer is clear or not, if it is hazy or contaminated with RBC's, don't separate the plasma and mix the bag properly and re-spin.
7. If the separation layer is clear, then express out all the plasma into the empty satellite bag, while the bag with additive solution is clamped.
8. Irrespective of the spin, almost entire plasma (platelet rich or otherwise) can be extruded out of the red cells.
9. Now clamp the bag which plasma is there. Break the seal of the additive solution and add the additive solution into the red cell bag. Red cells in additive solution (RC) are ready.
10. Detach the bag, which has red cells in additive solution. Paste products labed it.

Separation of Fresh Frozen Plasma (FFP)

Material Required

- Refrigerated centrifuge (Roto Silinta 630 RI/Rotixa 50 RS)
- Plasma expressor (Fresenius Hemocare)
- Tube stripper (Fresenius Hemocare)
- Bucket corrector, (Fresenius Insignia)
- Clamps
- Tube sealer (Fresenius Hemocare)

Procedure

1. Blood component preparation mainly depends on speed and time of centrifugation. The blood bag centrifuge ROTO SILENTA is preprogrammed.
2. The FFP can be obtained either from PRP by giving it greater centrifugal force as compared to the soft spin or directly from whole blood after giving it hard spin.

FFP from PRP

From PRP we can obtain FFP by submitting it to the following program.

Table:- 1 Program for preparation of FFP from PRP.

Application	Accel No.	Decel No.	Speed rpm	RCF	Time Min.	Temp. ℃
PC(Modified)	6	B5	3947	4999	5:30 Sec.	22

FFP directly from whole blood

From whole blood we can obtain FFP by submitting it to the following program.

Table 2: Program for preparation of FFP directly from whole blood.

Application	Accel No.	Decel No.	Speed rpm	RCF	Time Min.	Temp.℃
FFP	6	B5	3947	4999	5:30 sec.	4

1. The entire plasma express into the satellite bag, leaving only red cells in the primary bag.
2. The additive solution is added to the red cells and bag is detached.
3. The plasma is immediately put in freezer and qualifies the definition of Fresh Frozen Plasma (FFP) if it has been separated within six hours of collection.
4. Put product labels and enter in quarantine in software.
5. Once out of blast freezer, the FFP is stored at- 30℃ or lower.

Preparation of Platelet Concentrate
Material Required

- Refrigerated Centrifuge (ROTO Silinta 630RI/ Rotixa 50 RS)
- Plasma expresser (Fresenius Hemocare)
- Tube stripper (Fresenius Hemocare)
- Bucket corrector (Fresenius Insignia)
- Clamps
- Tube sealer (Fresenius Hemoseal)

Procedure

1. Blood component preparation mainly depends on speed and time of centrifugation. The blood bag centrifuge ROTO SILENTA is preprogrammed.
2. To make platelet concentrate (PC), we give soft spin in preprogrammed centrifuge. After centrifugation is over, arrange the entire blood bag on Plasma Expresser.
3. Check whether the line separating red cells & plasma layer is clear or not. If it is hazy or contaminated with RBCs, don't separate the plasma and mix the bag properly and re-spin.
4. If the separation layer is clear, than express out all the plasma (platelet Rich Plasma-PRP) into the empty satellite bag, while the bag with additive solution is clamped.
5. If platelet Rich Plasma (PRP) shows some red cell contamination, do not make platelets from this bag
6. Now clamp the bag in which PRP is there and then add the additive solution into the red cell bag and detach that bag.
7. The remaining component is called Platelet Rich Plasma (PRP). Centrifuge the platelet rich plasma at program 3 to get Platelets and FFP.
8. After this spin, platelets will settle down in the bag. So express out all the plasma into the satellite bag, leaving about 50ml (45-65 ml) plasma with the platelets and detach both bags.
9. Despite the plasma being clear in PRP, if after the second spin, the platelet concentrate shows significant red cell contamination, do not separate platelets from this bag. Express the clear plasma into the satellite bag and make FFP only. The bag with red cell contaminated platelet button should be detached and discarded.
10. The bag having platelets is called Platelet Concentrate and the other component is called Fresh Frozen Plasma if it has been separated within six hours of collection and is snap-frozen.
11. Put product labels accordingly and enter in quarantine un software.
12. Ten keep Platelet Concentrate for 1 hour at room temperature.
13. Put them on platelet agitator at 20-24℃

SOP for blood grouping on donor sample
Material Required

- Donor's EDTA sample.
- Anti-A, Anti-B, Anti-D
- 5% suspensions of reagent A, B and O cells.
- Test tubes, test tube racks, marker pencil, marker pen, glass slide.
- Normal saline, Distilled water.

Procedure

Before performing actual grouping all the samples are arranged in the racks in an ascending order for example as 1, 2, 3 … etc. These samples are scanned using the bar code scanner and DD nos. are entered in the Excel worksheet. Same DD nos. is written in the blood grouping register according to their arrangement in the racks.

A. Forward grouping

1. Forward grouping is performed using the slide technique.
2. Write last four digits of the DD no on the slide.
3. Using the wax pencil draw lines on each slide to make three separate portions.
4. Put 2 drops each of anti-A, anti-B and anti-D on the slide, in that order from top to bottom of the slide
5. Using the Pasteur Pipette, put two drops of serum in each of the test tube labeled as A, B and O cells, kept in front of the donor sample tube.
6. Using the antisera and using applicator stick or edge of another slide.
7. Using the same pipette, donor whole blood is taken and one drop is put each against the three antisera puton the slide.
8. Rock the slide gently for approximately one minute and note and record the results.

B. Reverse grouping

1. Label three tubes as A, B and O cells.
2. Add 2 drops of test sera in each tube.
3. Add 1 drops 5% suspension of washed reagent red cells in each corresponding tube.
4. Mix and centrifuge at 1000 rpm for 1 min.
5. Gently suspend the cell button, look for agglutination or hemolysis and record the results immediately.

C. Table:-3 Interpretation of forward and reverse grouping results

Forward grouping			Reverse grouping			Blood Group
Anti A	Anti B	Anti D	A cells	B cells	O cells	
3-4+	-	3-4+	-	3-4+	-	A +ve
3-4+		-	-	3-4+	-	A -ve
-	3-4+	3-4	3-4+	-	-	B +ve
-	3-4+	-	3-4+	-	-	B -ve
3-4+	3-4+	3-4+	-	-	-	AB +ve
3-4+	3-4+	-	-	-	-	AB -ve
-	-	3-4+	3-4+	3-4+	-	O +ve
-	-	-	3-4+	3-4+	-	O -ve

Blood grouping in case of Rh (D) negative samples

1. 2 drops of added anti-D reagent from a different manufacturer in a labeled tube.
2. Add 1 drop 5% suspension of normal saline washed test red cells.
3. Mix and centrifuge at 1000 rpm for 1 min.
4. Gently re-suspend the cell button and look for agglutination.
5. If agglutination is present label as Rh (D) positive, otherwise perform ICT for weak D antigen.
6. If ICT is negative label donor unit as Rh (D) negative.

SOP for red cell cross-match

Material required

- 1 Requisition form for blood component.
- 2 patient's blood sample.
- 3 Gel cards and LISS diluents.
- 4 Gel card incubator and centrifuge (DiMed-IDMicrotyping system).
- 5 Pipette, test tube rack, test tube, pipette tips. (Multichanel pipette BioHit).
- 6 Sample centrifuge (Hittich EBA-20)

Procedure

Blood grouping on patient's sample, product reservation and cross matching.

1. Perform forward ABO grouping of the patient's EDTA sample by slide method to confirm the blood group of the patient.
2. Centrifuge patient's EDTA sample and / or serum sample at 3000 rpm for 3 min.
3. Enter the Patient's requisition in the software.
4. While entering details always scan the PD No.
5. Reserve the required product displayed in the software on the top of the list of the respective blood group.
6. Take that blood unit out of refrigerator.
7. Reserve this unit for cross-match and save it in the software.
8. Arrange the required no. of tubes on the rack according to the plan mentioned in Annexure 1.
9. Break off two segments from the donor bag (one for repeat blood grouping and cross match & the other for keeping for 7 days). Do forward grouping on the donor unit sample to reconfirm the group of the blood unit to be given & mention it in the requisition form. It should be the same as patient's blood group.
10. Paste one of the eye readable DD number from the blood bag on the back side of the requisition from.
11. Paste another eye readable DD no. on to the Gel card to be used for cross matching. Write PD no of the patient in this.
12. Put the donor unit back into the refrigerator.
13. Take 1000 µl of LISS solution in a test tube.
14. To the tube add 10µl of donor red cells to make around 0.8% suspension. Mix properly.
15. From this tube pipette out 50µl of cell suspension and add to the previously labeled gel card.
16. Add 25µl of the patient's plasma (or serum if only plain sample is available) to the same gel card.
17. Put the gel card in the incubator and press the start button.
18. While incubation is in progress do reverse grouping by tube technique and perform repeat Rh D typing of the patient's red cells by tube technique. Record results.
19. After incubation is over put the gel card in card centrifuge and press start button.
20. After centrifugation is over read & record the results as follows.
21. Compatibility result is to be mentioned on the requisition form also.
22. If compatible enter the result in the software, record the result in the form and reserve the unit for issue.
23. If the attendant is not there in the blood bank or asks to reserve it for the next day, paste a small handwritten sticker on the backside of the cross-matched unit mentioning PD No. and name of the patient & cross-match date.
24. Issue blood component unit.

RESULT

The present study is carried out to ascertain the usage of various blood components (RC, PC, & FFP) in different clinical specialties (Surgery & trauma, medicine, Obstetric Gynecology) at RedAid Blood Centre, Khanna, Punjab.

The Dispensation of Various Blood Components (RBC, PC, FFP)

Table 4: Percentage of different blood components utilized by clinical patients.

Components	Total No. of units	Percentage
RC	4033	75.9%
PC	701	13.2%
FFP	575	10.8%

A total requisition for various types (RBC, PC, FFP) of component requirement were screened and all the relevant clinical information concerning age, sex, specialty, provisional diagnosis, the indication for component therapy and average number of component dispensed per patients and specialty was analyzed. Out of 3000 cases 118 in compete requisition were excluded due to inadequacy of above mentioned criteria for inclusion in the study. A total number of 5309 units of various (RBC, PC, FFP) blood components were utilized during the period of study, in various specialties during of study. Total 4033 units of red cell concentrate, 701 units of platelet concentrates and 575 units of fresh frozen plasma were dispensed. The average blood component units utilized by per patient were 1.769.

Specialty Wise Dispensation of Various Blood Component (RC, PC & FFP)

Table-5: No. of patients and their percentage requiring blood components in different clinical specialty.

Specialty	No. of Patients	Percentage
Surgery & Trauma	600	20%
Medicine	1788	59.6%
Obstetric Gynecology	345	11.5%
Pediatrics	267	8.9%

As evident from Table 5.1 and Pie Chart 5.1 maximum no of units red cells concentrate were dispensed 75.9%, the platelet concentrate 13.2% and fresh frozen plasma constituted utilization was highest.

Dispensation of various blood components (RBC, PC, FFP) among patients of different clinical specialty

Red cell concentration

On analysis of data (refer to Table 5.3 and Pie chart 5.3) The utilization of blood components in different clinical specialty showed that, out of 4033 units of Red cell concentrate 58.1% (2374) was utilized in medicine, 24.6% (986 units) in surgery & trauma, 12.6% (507 units0 in Obstetrics & Gynecology & 4.7% (193 units) were utilized in pediatrics.

Table 6: Dispensation of red cells among patients of different clinical specialties and their percentage.

S. No.	Specialty	Red cells	Percentage
1.	Surgery Trauma	986	24.6%
2.	Medicine	2347	58.1%
3.	Obstetric Gynecology	507	12.5%
4.	Pediatrics	193	4.7%

The incident of Red cells, utilized in medicine was higher (58.1%) followed by surgery & trauma (24.6%), Obstetric Gynecology (12.6%) & low incidence was found in pediatrics group (4.7%).

Platelet concentrate

As evident from data (Refer to Table 5.4 and Pie chart 5.4) the dispensation of platelet concentrate showed that out of total 701 units of platelet concentrate, 77.9% were utilized in medicine, 11.6% in pediatrics, 5.9% in surgery/ trauma & 4.6% were utilized in Obstetrics & Gynecology.

Table 7: Dispensation of platelet concentrate among patients of different clinical specialties and their percentage.

S. No.	Specialty	Platelet conc.	Percentage
1	Surgery Trauma	42	5.9%
2	Medicine	546	77.9%
3	Obstetric Gynecology	32	4.6%
4	Pediatrics	81	11.6%

The incidence of platelet concentrate utilization was higher in medical specialty (77.9%) and found to be lower among obstetrics & gynecology (4.6%).

Fresh Frozen Plasma

On analysis of data (Refer to Table5.5 and Pie chart 5.5) for the FFP dispensation in different clinical specialties, out of 575 units of FFP. 74.4% were utilized in medicine, 13.7% in surgery & trauma, 9.6% in pediatrics & 2.3% were used in Obstetrics & Gynecology.

Table 8: Dispensation of FFP among patients of different clinical specialties and their percentage.

S no.	Specialty	FFP	Percentage
1	Surgery Trauma	79	13.7%
2	Medicine	428	74.4%
3	Obstetrics Gynecology	13	2.3%
4	Pediatrics	55	9.6%

The utilization of FFP users was higher in medicine (74.4%) followed by surgery/ trauma (13.7%), pediatrics (9.6%) & was found to be lower in obstetrics & Gynecology (2.3%).

AGE DISTRIBUTION & MEDIAN AGE OF PATIENTS REQUIRING COMPONENT THERAPY

The data of an age distribution of patient requiring various components showed out of 2882 patients, The age group between 0-20 years, were 19%, 21-40yrs, 37%, 41-60yrs, 24% &>60 yrs,were 20% The cases in age group 21-40 years showed 37%.

Table 9: Age distribution of patients in years component therapy.

Age group	No. of Patients	Percentage
0-20 yrs.	543	19%
21-40 yrs.	1060	37%
41-60 yrs.	714	24%
>60	565	20%

The blood components utilization was found to be higher in the age group 21-60 yrs. (61%), & lower in age group between 0-20yrs. (19%).

Sex Distribution of Patients Requiring Blood Components Therapy

The sex distribution data of patients requiring blood component are following.

Table 10: Percentage of male & female requiring blood components.

Gender	No. of Patients	Percentage
Male	1345	46.66%
Female	1537	53.34%

Out of 2882 patient, the male patients were 1345 & female patients were 1537. The female patients was found (53.34%), while the male patients were found (46.66%) which show increase frequency of blood components required in female. Refer to Table 5.10 & Pie chart 5.9.

DISCUSSION

The blood transfusion has important role in modern practice of medicine and without it many surgical procedure, cardiac surgery, treatment of various hematological disorder are impossible, due to limited resources of blood and increased demand there is now significant need of rational use of blood and its components.

1. To minimizing the inappropriate use and to plan the blood needs for the future, with these aims the present study was carried out at RedAid Blood Centre, Khanna, Punjab. The survey was done by taking 3000 requisitions for various types of component (RBC, PC, and FFP) requirement. The screening of component therapy requirement was based on, all the relevant clinical information concerning age, sex, clinical specialty, provisional diagnosis, indication for component therapy. Out of 3000 cases in-complete were excluded due to inadequacy of above mentioned criteria for the study.

A total of 5901 units of various blood component were utilized during the period of study and was found to be 1.769 units utilization per patients. Among the entire blood component about 75.9% of red cell concentrate were utilized. The increase Red cell concentrate requirement was due to higher number of cases of anemia of varied etiologic origin, Chronic Failure, bleeding from various causes & other surgical or medical conditions resulting in hypovolemia.

The utilization of platelet concentrate was 13.2% & FFP was 10.8% is in correlation with findings of Stanworth et al; 2000, they reported Red cell concentrate utilization at (58.2%) The largest utilization of red cell concentrate was 58.1% in medical specialties followed by 28.6% in surgery and trauma and 12.3% in Obstetrics & Gynecology of Abayomi et al; 1999; they reported the highest utilization in obstetrics & gynecology followed by surgery and pediatrics.

The utilization of red cell concentrate in medical specialties was higher due to large numbers cases belonging to anemia, dialysis, bleeding from GIT, CRF patients, patients of hematemesis which were managed on conservative line of treatment so included in specialties.

Similarly the analysis on data and platelet concentrate utilization showed the maximum utilization of platelet components (77.9%) in medical specialties followed by 11.6% in pediatrics and 5.9% in surgery and trauma and lowest utilization was found in obstetrics & gynecology (4.6%).

The higher rate of utilization in medical specialties may be due to a relatively higher number cases belonging to medical diseases like septicemia, thrombocytopenia, CRF, Aplastic anemia, bone marrow depression due to various factors and Dengue out breaks.

The data on FFP showed the highest utilization was found in medical specialties (74.4%) followed by 13.7% in surgery and trauma and 9.6% in pediatrics and 2.3% in obstetrics & gynecology. The higher utilization of FFP in medical specialties due to high number of cases of liver disease, hypovolemia and patients of DIC.

The age wise utilization rate of blood component when analyzed showed that the peak age of component therapy was found to be higher in the age of 21-60 years and about 61% requirement component was found to be in this age brackets as most of the medical and surgical ailments have increased predisposition during this age bracket. The major clinical or surgical conditions were the anemia, infections, surgical procedures, hemorrhoids etc.

When the data of red cell concentrate were analyzed on the basis of age relation the utilization was found to be higher in the age group of 0-40 years (65.4%) due to

most of the cases were of thalassemia associated with anemia, jaundice, enteric fever etc.

When the data on platelet concentrate was reviewed the highest utilization was found in the age group of 21-60 years (70.3%) followed by patient belonging to age group of 0-20years (16.8%). The higher utilization of platelet concentrates in age group 21-60 years may be due to many kinds of surgery, bleeding piles, liver cirrhosis, CRF etc.

Similarly when the utilization of FFP was reviewed in relation to age its showed the higher rate of utilization found in 0-40 years of age due to large group of cases belonging to anemia, thalassemia major and minor, liver disease, DIC etc.

The sex incidence of blood component utilization was found to be higher in female 53.34% compared to male 46.6% due to most of the female were anemic.

REFERENCES

1. American Association of Blood Banking; Technical Mannual, 1996.
2. Arkin CF: Quality Contro; and Standardization in the Hematology Laboratory. I Brick RL, editor. Hematology: Clinical and Laboratory Practice. St Lousie. Mosby, 1993; P-17-37.
3. Astiz ME and Rackow EC. Assessing perfusion failure during circulatory shock. Critical Care. 1993; 3: 237.
4. AuBuchon JP. Autologous transfusion and directed donations: Current controversies and future directions. Transfusion Medicine Rev. 1989; 2: 290.
5. Baer D.M. and Krause, R.B. (1968): Spurious laboratory values resulting from simulated mailing conditions. American Journal of clinical Pathology, 50: 111.
6. Barbra. J. Bain: Blood Cell (A Practical Guide) Performing a Blood Count. 3rd edition. 2001; Blackwell Science Ltd London, P-23.
7. Bernice Glatzer 2002: Accessed from www.google.com
8. Bode AP: The use of flow cytometery in the study of blood platelets. In: Riley RS, Mahin EJ, Ross W. Clinical applications of flo9w cytometery. New York: IgakuShoin, 1993; P-613-33.
9. Diamond LK. A history of Blood transfusion. In: Wintrobe MM, ed. Blood, pure and eloquent. New York: Mc Graw Hill, 1980.
10. Director General of Health Science manual 2nd edition.
11. Erslev Aj, Caro J and Schuster SJ. Is there an optimal hemoglobin level? Trans Med Rev, 1989; 3: 237.
12. Eschbach JW Correction of the anemia of end-stage renal disease with recombinant human erythropoietin. N Engl J Med, 1987; 316:73.
13. Farr AD. The first human transfusion. Med Hist, 1980; 24: 143.
14. Firkin de Gruchy's Clinical Haematology in Medical Practice 5th edition, 1992.
15. Godkar text book of Medical Laboratory Technology first edition, p-439-441.
16. Goodenough LT A two-institution study of transfusion practice in 78 consecutive adult elective open-heart procedures. Am J ClinPathol, 1989; 91: 468.
17. Garratty G and Petz LD. Acquired immune hemolytic anemias. New York: Churchill-Livingstone, 1980.
18. Greenwalt TJ and Zelenski KR>Transfusion support for haemoglobinopathies. ClinHaematoligy, 1984; 13: 151.
19. Gmur J, Burger J, Schanz U Safety of stringent leukemia. Lancet, 1991; 338:190:91.
20. Harker LA Mechanish of abnormal bleeding in patients undergoing cardiopulmonary bypass: Acquired transient platelet dysfusncion associated with selective alpha-granule release. Blood, 1980; 2: 56: 824.
21. Harvey Simon, MD, Editor-in-Chief, Associate Professor of Medicine, Harvard Medical School; Physcian, Massachusetts Genernal Hospital: Anaemia, 2004.
22. Hennery JB, 1996.
23. Inkster M Preservation of antithrombin III activity in stored whole blood. Transfusion, 1984; 24: 57.
24. International Committee for Standardization in Hematology (1983): Protocol for evaluation of automated blood cell counters: Clinical and Laboratory Hematology, 6: 69.
25. International Council for Standardization in Hematology (1994): Guidelines for the evaluation of blood cell analyzers including those used for differential leukocyte and reticulocyte counting and cell maker applications. Clinical and Laboratory Hematology, 1994; 16: 157.
26. J. Ochie, A Kolhatkar: Medical Laboratory Science Theory and Practice (Production of cells). 2000; 1st edition, 272: 386-87.
27. J.M. England chapter Automated blood cell counters page no, 161.
28. Kumar Robbins- Basdic Pathology 7th Edition, 2003.
29. Makroo RN, 1999: Compendium of Transfusion medicine, 2nd edition.
30. Manual Gude for ADIVA 60 OT automated haematology system.
31. Murphy S, Indications for platelet transfusions in children with acute leukemia. Am J Hematol, 1982; 12: 347.
32. National Institutes of Health Consensus Conference. Fresh frozen plasma: Indicationand risks. JAMA, 1985; 253: 551.
33. National Institutes of Health Consensus Conferences. Perioperative red blood cell transfusion. JAMA, 1988; 260: 2700-2703.
34. Petz LD. Blood Transfusion in acquired hemolytic anemias. In: transfusion medicine. 3rd ed. New York: Churchill Livingstone, 1996; 469-499.

35. Petz LD. Red blood cell Transfusion. Clinoncol, 1983; 2: 505.
36. Ramnik Sood – Medical laboratory Technology; First edition.
37. Red Blood cell indices: implications for practice NBIN, 2004; 4(4): 231-239.
38. Rev. Nutr. V. 15n. 2 compinas maio/ago. 2002.
39. Slichtr SJ. Controversies in Platelet transfusion therapy. Ann Rev Med, 1980; 31: 509.
40. Shirey RS, Boyd JS, Ness PM (1994).
41. Wain SL. The controversy of unmodified versus citrated blood transfusion in the early 20th century. Todd. Sanford and Davidsohn. Clinical diagnosis & Management by laboratory method Seventeenth (17th) Edition 2003.
42. Transfusion, 1984; 24: 404.
43. Wintrobe's Clinical Hematology 10th edition, 1999.
44. Williams and Wilkins, 1996.
45. World Health Organization: Geneva (A.I.T.B.S.2004).
46. Yango, 1994.