STANDARD OPERATING PROCEDURES (SOPs)

FOR BLOOD BANK (06)

Book 1

Department of Health & Family Welfare, GNCTD

SOP for General Administration and Equipment Management, Ist Edition: August; 2016
Quality Assurance Cell
Delhi State Health Mission
Department of Health and Family Welfare
Government of NCT of Delhi

Compilation facilitated by : State QA Cell (Nodal Officer: Dr. Monika Rana , Consultant : Ramesh Pandey , Communitization Officer : Arvind Mishra , Statistical Officer : Shahadat Hussain), ARC (Maneesh and Irshad).
Designed and Formatted by: Graphic Designer: Mansi Rana

PREFACE

With the aim of providing Quality Health care in its Hospitals and Blood Bank, Delhi Government has proposed to prepare Standard Operating Procedures for different techniques to be followed in Blood Banks and Blood Storage units.

Delhi Government has entrusted the responsibility of drafting SOPs of Blood Banks and BSUs to the committee chaired by Dr. Bharat Singh along with the members.

The technical experts in the field of Blood Transfusion including Dr. Mausumi Swami, D.D.U. Hospital, Dr. Meenakshi Sidhar Dr. BSA Hospital and Dr. Alok Singh DHAS have contributed in drafting these procedures. Dr. Shivani Paik DDDMASC hospital has helped as co-opted member. The efforts and hard work of the experts are praiseworthy.

While drafting these SOPs the Transfusion Medicine Technical manual of DGHS GOI, Drug and Cosmetic Act, NACO and NABH standards for Blood Banks were kept in mind.

These SOPs will be of great help to all Blood Banks and BSUs in improving quality and achieving uniform standards in Blood Bank. These SOPs will also facilitate the Blood Banks in accreditation.

Dr. Bharat Singh
Chairperson, Drafting Committee

This document has been prepared by the Expert Committee comprising of:

Sr. No.	Name	Designation	
1.	Dr. Bharat Singh, M.D	Consultant Pathologist (SAG) & Head, Regional Blood Transfusion Centre, GTB Hospital & Hony. Director, State Blood Transfusion Council, Govt. Of Delhi	Chairperson
2.	Dr. Mausumi Swami	Head, Department of Blood Bank & Regional Blood Transfusion Centre, DDU Hospital New Delhi-110064	Member
3.	Dr. Meenakshi Sidhar	HOD, Blood Bank Dr. BSA Hospital, Rohini New Delhi-110085	Member
4.	Dr. Alok Singh	HOD, Blood Bank DHAS Hospital Delhi-110032	Member
5.	Dr. Shivani Paik	Incharge BSU, DDMASC Delhi-110046	Member

The SOPs have been prepared by a Committee of Experts and are being circulated for customization and adoption by all hospitals. These are by no means exhaustive or prescriptive. An effort has been made to document all dimensions / working aspects of common processes / procedures being implemented in provision of healthcare in different departments. This document pertains to the Blood Banks / Blood Storage Units . The individual hospital departments may customize / adapt / adopt the SOPs relevant to their settings and resources. The customized final SOPs prepared by the respective Departments must be approved by the Medical Director / Medical Superintendent and issued by the Head of the concerned department. The stakeholders must be trained and familiarized with the SOPs and the existing relevant technical guidelines / STGs / Manuals mentioned in the SOPs must also be made available to the stakeholders.

1. DETAILS OF THE DOCUMENT (FIRST PAGE)

Name of the Blood Bank
Name of the Hospital , Delhi-1100
License no -

Address:

Document Name :	
Document No. :	
No. of Pages :	
Date Created :	
Prepared By :	**Designation :** **Name :** **Signature :**
Approved By :	**Designation :** **Name :** **Signature :**
Responsibility of Updating :	**Designation :** **Name :** **Signature :**

2. AMENDMENT SHEET (SECOND PAGE OF DOCUMENT)

AMENDMENT SHEET

S.No.	page no.	Details of the amendment	Reasons	Signature of the preparatory authority	Signature of the approval authority

3. CONTROL OF THE DOCUMENT (THIRD PAGE OF THE DOCUMENT)

The holder of the copy of this manual is responsible for maintaining it in good and safe condition and in a readily identifiable and retrievable.

The holder of the copy of this manual shall maintain it in current status by inserting latest amendments as and when the amended versions are received.

The Manual is reviewed once a year and is updated as relevant to the Hospital policies and procedures.

The Authority over control of this manual is as follow:

Prepared By	**Approved By**	**Issued By**
Name: Designation : HOD /Dept. In charge Signature:	Medical Superintendent Name: Signature:	Quality – Nodal Officer Name: Signature:

The procedure Manual with Original Signature of the above on the Title page is considered as **"Master Copy"** , and the photocopies of the master copy for the distribution are considered as **"Controlled Copy'**.

Distribution List of the Manual

Sr. No.	Officials	Signature of Officials receiving copy

INDEX

1. SOP FOR DONOR COMPLEX

Number DC/001	Effective Date	Pages 2	Author	Authorized
Version	Review period 2 years	No. of copies: 5	Approved By	Revision Date
Location Donor Complex			Subject: Donor Registration	
Function To obtain complete demographic Information about prospective blood donor			Distribution • HOD • Quality Manager • Technical Manager • Concerned station • Master Copy	

Sl.No.	Subject	SOP NO.
1.	Donor registration	DC/001
2.	Donor screening	DC/002
3.	Hemoglobin estimation using Hemoglobin analyzer	DC/003
4.	Selection & preparation of Blood bags selection of Blood Bag.	DC/004
5.	Labeling of blood bag.	DC/005
6.	Phlebotomy- preparation of donor for phlebotomy	DC/006
7.	Bleeding of Donor	DC/007
8.	Donor safety-Post Donation care	DC/008
9.	Adverse donor reaction and their management	DC/009
10.	Single needle platelet aphaeresis or bio cell separator	DC/010
11.	Blood Collection Monitor- steps to use	DC/-011

1.0 PURPOSE

To obtain complete demographic information about prospective blood donor

2.0 SCOPE

The processes of identifying a donor is extremely important because it serves to provide both identification of the source of the blood product& recall information for future donation

3.0 RESPONSIBILITY

Attending the donors to get the necessary demographic information on donor selection & registration form. Doctor must be able to provide accurate answer to donor's general queries.

Responsible Person

Registration desk /Staff Nurse/Doctor

4.0 ACTIVITY

4.1 Material

4.1.1 Donor's Selection, Registration/ Consent Form
4.1.2 Pens
4.1.3 General information in the form of pamphlets about blood donation

4.2 Method

4.2.1 Whenever a prospective donor comes to the blood bank, he /she should be greeted with a pleasant smile and asked to sit down comfortably.

4.2.2 He/she is offered a glass of water and asked to relax.

4.2.3 He is given the donor selection and registration form and asked to fill,
For donor registration form please refer the annexure no –1

4.2.4 If he /she have any difficulty in filling up the form then donor should be helped in the same and form should be completely filled.

4.2.5 After filling up the form and duly signed by the donor, he/she is sent to the Medical Officer/ Doctor on Duty in the donor screening room.

4.2.6

5.0 PRECAUTION

Certain information i.e. full name & date of birth must be verified at all points along the donor process to ensure & guard against any possible mix –up of donor records.

6.0 RECORD

The donor selection & registration form on to which complete information is recorded will become a permanent record, which must be retained in compliance with regulation.

7.0 REFERENCES

- 7.0.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
- Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
- Standards for blood bank and blood Transfusion Services NACO 2007

8.0 ANNEXURES

Annexure no 1 Donor Selection & Registration Form

Annexure no 2 Donor Consent Form for Apheresis

Number DC/ 002	Effective date	Pages 3	Author	Authorized by
Version	Review Period 2 years	No. of Copies :5	Approved By	Revised Date
Location Donor Complex			Subject Donor Screening	
Function: Assessing suitability of donor for Blood donation			Distribution • HOD • Quality Manager • Technical Manager • Designated area • Master Copy	

1.0 PURPOSE

The purpose of donor screening is two-fold. First, the screening process is necessary to assess whether the individual is healthy enough to donate with no detrimental effect to the donor . The patient or recipient must be protected from potential adverse effects from the transfusion of a donated product.

2.0 SCOPE

The donor selection process is one of the most important steps in maintaining the safety of the blood donor and quality of blood product.

3.0 RESPONSIBILTIES

3.1 Responsibilities of Junior Resident/SR/MO
3.2 To obtain complete medical history from donor
3.3 Alleviate the apprehension and medical queries regarding blood donation
3.4 Provide relevant information to the donor
3.5 Medical examination

4.0 ACTIVITIES

The donor screening process has three major components /activities.

4.1 Donor Registration
4.2 Medical history
4.3 Physical examination

Upon successful completion of these three interrelated but separate parts, the donor is declared fit for donation and selected for phlebotomy.

The donor screening process has one of three outcomes for the prospective donor.

- **Acceptance, 2) Temporary deferral, 3) Permanent deferral**

Accepted donor continues on to the donation process.

Temporary deferred donors are advised on how long they must wait before trying to donate again. They are also advised on what they should do to increase their chances of acceptance

after the waiting period is over. For example, if donor's hemoglobin is too low to allow the donor to safely donate, he or she is advised on how to increase the hemoglobin level through dietary change/medication.

Permanent deferral is given to those people who cannot be accepted as a blood donor under any circumstances.
The history and physical examination must take place on the actual day of donation /Donor Registration.

The donor screening process begins with obtaining complete and accurate demographic information about the donor at the time of initial donor registration. The information must fully identify the donor and enter the donor to existing donor record. Current information must be obtained and recorded for each donation. Data collected for each prospective donor should include the following required information.

Date and time of donation
Name: First, Last (and middle initial if available)
Address: residence /or business.
Telephone number
Email if any:
Gender :
Age and /or date of birth:
Occupation:
Unique characteristics of the donor: Donors with known Rh negative blood group/ Bombay phenotype –record of which should be maintained separately.

MATERIALS REQUIRED:

- Donor Questionnaire
- Donor card

5. PROCEDURE:

CRITERIA FOR SELECTION OF BLOOD DONORS

A. Accept only voluntary/replacement non-remunerated blood donors if following criteria are fulfilled:
The interval between blood donations should be not less than three months. The donor shall be in good health, mentally alert and physically fit and shall not be a jail inmate or a person having multiple sex partners or a drug-addict. The donors shall fulfill the following requirements, namely: -

(a) the donor shall be in the age group of 18 to 65 years

(b) the donor shall not be less than 45 kilograms

(c) temperature and pulse of the donor should be normal

(d) the systolic and diastolic blood pressures should be within normal limits (Systolic-100-160mmHg & Diastolic 60-90 mmHg) , controlled hypertensive on single drug may be considered

(e) haemoglobin should not be less than 12.5 g/dL

(f) the donor should be free from acute respiratory diseases

(g) the donor should be free from any skin diseases at the site of phlebotomy

(h) the donor should be free from any disease transmissible by blood transfusion, in so far as can be determined by history and examination indicated above

(i) the arms and forearms of the donor shall be free from skin punctures or scars indicative of professional blood donors or addiction of self injected narcotics

(j) donor should not be under the influence of alcohol

B. Defer the donor for the period mentioned as indicated in the following table:

CONDITIONS	**PERIOD OF DEFERMENT**
(1)	(2)
(a) Abortion /Delivery	6 months
(b) History of blood transfusion	6 months
(c) Surgery	12 months
(d) Typhoid	12 months after recovery
(e) History of Malaria duly treated	3 months (endemic) 3 years (non endemic area)
(f) Tattoo	6 months
(h) Breast feeding	12 months after delivery
(i) Immunization (Tetanus, Plague, Cholera, Typhoid, Rubella, Gamma-globulin)	15days

(j) Rabies vaccination	12 months
(k) Hepatitis in family or close contact	12 months
(l) Immunoglobulin	12 months
(m) On Antibiotic/Aspirin(for platelet)	3 days after stoppage
(n) Menstruation	Till periods finish
(o) Drugs-isotretinoin for acne,finasteride for prostate hyperplasia	One month after last dose
(p) Drugs- cortisone	7 days after stoppage

C. Defer the donor permanently if suffering from any of the following diseases:

a. Cancer
b. Heart disease
c. Abnormal bleeding tendencies
d. Unexplained weight loss
e. Diabetes - controlled on Insulin
f. Hepatitis B/ C infection
g. Chronic renal disease/ failure
h. Signs and symptoms, suggestive of AIDS
i. Liver disease
j. Tuberculosis
k. Polycythemia Vera
l. Asthmatics on steroids
m. Epilepsy
n. Leprosy
o. Schizophrenia
p. Endocrine disorders
q. It is important to ask donors if they have been engaged in any risk behaviour. Allow sufficient time for discussion in the private cubicle. Try and identify result-seeking

donors and refer them to VCTC (voluntary counselling and testing center). Reassure the donor that strict confidentiality is maintained.

D. Private Interview:

A detailed sexual history should be taken. Positive history should be recorded on confidential notebook- to be maintained by the counsellor.

6. Physical Examination

The physical examination begins by observing the prospective donor's generable appearance.
The donor should be in good health, mentally alert and physically fit.
The interviewer decides whether the donor is obviously in need of sleep or not.
Hemoglobin, blood pressure, temperature, donor weight, height and local skin examination on phlebotomy site is checked by designated person.

Hemoglobin: -Hemoglobin level for blood donation must be 12.5g/dl (125 g /l) or greater and hematocrit must be 38 per cent (0.38).

Pulse: The pulse of donor should be between 60 and 100 beats per minute and regular. If the donor is an athlete a lower pulse may be accepted. Pulse should be taken for at least 30 second.

Blood Pressure: -Systolic and diastolic blood pressure should be within normal limits with or without medication (systolic – 100 to180 mm of Hg& Diastolic –50 to100 mm of Hg)

Temperature:

- Oral temperature must not exceed 37.5^{o} c (99.5^{o} F).
- Donor weight: To avoid acute hypovolemic reaction from donation the potential donor must weight a minimum of 45 Kg.
- The arm and forearm of donor should be free from skin disease at the site of phlebotomy.
- The arm and forearm of the donor should be free from skin punctures or scars indicative of professional donor or addiction of self injected narcotics.
- Examination of Respiratory system, Cardiovascular system and per abdomen should be carried out if necessary.

Informed Consent

If nothing abnormal has been found in the prospective donor's history and medical examination the final step just prior to donation is the informed consent process. The donor record includes the signed statement of donor countersigned by Consultant that acknowledges the informed consent process.

Annexure no 2: Format of informed consent

RECORDS

Information obtained from donor on registration form and results of their physical examination entered in donor record register

Deferred donor and reason for their deferral should be recorded in the register

Record of donor reaction and its management

REFERENCE

- Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
- Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
- Standards for blood bank and blood Transfusion Services NACO 2007

Number DC/00	Effective Date	Page 2	Author	Authorized
Version -	Review period 2years	No. of copies:5	Approved By	Revision Date
Location **Donor Complex**			**Subject** **Hemoglobin estimation**	
Function: Method of estimation of Donor's Haemoglobin by portable Haemoglobin analyser			**Distribution** • HOD • Quality Manager • Technical Manager • Designated area • Master Copy	

0.0 PURPOSE

Estimation of donor's hemoglobin level for screening

2.0 SCOPE

Quantitative measurement of Hemoglobin level of prospective blood donor.

3.0 RESPONSIBILITY

Designated Technical staff.

4.0 ACTIVITY

4.1 PRINCIPLE

Based upon photometric method using dry chemistry & taking the measurement of absorbance at dual wavelength (570&880 nm.) Chemical reaction takes place in the cuvette and photometer automatically displays the result in less than 60 seconds.

4.2 Material

4.2.1 Spirit swab
4.2.2 Lancet
4.2.3 Dry cotton
4.2.4 Hb analyzer
4.2.5 Hb cuvettes
4.2.6 Tissue paper

4.3 Procedure

4.3.1 Switch on the machine.
4.3.2 Wait till the machine display is ready
4.3.3 Clean the fingertips with sterile spirit swab and wait to dry.

4.3.4 Puncture the finger firmly near the tip with a sterile disposable lancet. There should be free flow of blood.
4.3.5 Wipe the first drop of blood from fingertip.
4.3.6 Touch the microcuvette to the fingertip. It will be charged by capillary action.
4.3.7 Wipe the excess amount of blood from the side of the cuvette. Wipe it gently in such a way that blood inside the cuvette should not be disturbed.
4.3.8 Run the microcuvette in the analyzer chamber and note the display value of hemoglobin of the donor.

4.4 Precaution

If there is error display after switching on the analyzer do not proceed for the test. Refer to the service manual or call service engineer.

4.5 RECORD

4.5.1 Enter the hemoglobin values in donor selection and registration from.
4.5.2 Enter the hemoglobin values in donor record register.

5. REFERENCE:

Manufacturer guidelines (Hb analyzer)

Number DC/00	Effective Date	Page 1	Author	Authorized
Version - 04	Review period 2years	No. of copies:5	Approved By	Revision Date
Location **Bleeding room**			**Subject** **Selection & Preparation of blood bag**	
Function **Selection of blood bag**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated area • Master Copy	

1.0 PURPOSE

According to the component to be prepared from the blood unit & the weight of the donor the blood bags are selected for blood collection.

2.0 SCOPE

Optimize the use of single unit of blood & to minimize any detrimental post donation effects to the donor.

3.0 RESPONSIBILITY

3.1 Medical Officer In Charge of donor complex is responsible for selection of type of blood bag

3.2 Designated technician or nursing staff is responsible for preparation of blood bag.

4.0 ACTIVITY

4.1 Material

4.1.1 Different type of Blood Bags

4.1.2 Identification labels

4.1.3 Marker pen

4.1.4 Procedure

4.1.4.1 Check the bag visually. In case of discoloration, or growth in anticoagulant do not use it.

4.1.4.2 Product levels are dry and show no evidence of defacement or leaking anticoagulant.

4.1.4.3 Check the expiry date of the bag

4.1.4.4 Fix identical donor identification number labels to ensure accountability in all bags, satellite bags, pilot tubes and donor records and any special identification tags e. g. voluntary/ replacement/autologous etc.

5.0 RECORD

Enter the following details on donor registration/consent form and register.

5.1 Type of bag
5.2 Manufacturer's Name
5.3 Batch No.
5.4 Expiry Date of the bag
5.5 Segment No.

6.0 REFERENCES

6.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
6.2 Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
6.3 Standards for blood bank and blood Transfusion Services NACO 2007

Number DC/00	Effective Date	Page 1	Author	Authorized
Version - 0	Review period 2years	No. of copies:5	Approved By	Revision Date
Location **Bleeding room**			**Subject** **Labeling of Blood Bag**	
Function **Preparation of blood bag and pilot tubes**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated area • Master Copy	

1.0 PURPOSE

To label the bags and pilot tubes after verification of donor details in order to accurately relate the blood product to the donor.

2.0 SCOPE

The unit number label is the unique identifier for the donor & all the blood components separated from the unit collected from the donor.

3.0 RESPONSIBILITY

It is the responsibility of the phlebotomist/ nursing staff collecting the blood unit to ensure proper labeling & recording of the donor details even if donor area attendant affixes the label.

4.0 ACTIVITY

4.1 Material

4.1.1 Numerics coded labels

4.2 Procedure

4.2.1. Give each donor a unique number & once his blood is collected, identify by that number only.

4.2.2. Don't write donor's name on his /her blood bag or sample tube. This maintains the donor's confidentiality.

4.2.3. Affix per printed number label on the primary bag and on all the satellite bags in case of multiple bags and pilot tubes

4.2.4. Verify the donor identity by tallying with the name on the registration form. Affix the unit number label, now in the registration form.

4.2.5. Cross check the number on the bag, pilot tubes and registration form to ensure identity. Record the entry in the donor register using the same number.

4.2.6. Transcribe this number on all records hence forth for storage, testing and distribution.

4.2.7. While issuing the unit use the same number on issue record.

5.0 RECORD

Make sure that the labels are pasted and number is written on all records & there are no transcription errors as this number will trace any blood product from donor to recipient & vice – versa in case of requirement.

While issuing the unit use the same number on issue records.

6.0 REFERENCES

6.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.

6.2 Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)

6.3 Standards for blood bank and blood Transfusion Services NACO 2007

Number DC/000	Effective Date	Page 2	Author	Authorized
Version	Review period 2 years	No. of copies:5	Approved By	Revision Date
Location Bleeding room			Subject Phlebotomy	
Function Preparation of donor for phlebotomy			Distribution • HOD • Quality Manager • Technical Manager • Designated area • Master Copy	

1.0 PURPOSE

To minimize the risk of bacterial contamination of collected blood unit as well as prevention of donor from infection through the veni-puncture site.

2.0 SCOPE

Donor and blood product safety.

3.0 RESPONSIBILITIES

3.1 Prepare phlebotomy site according to SOP.

3.2 Responsible person : Nursing staff/ Junior Resident/ Medical Officer/ Senior Resident

4.0 ACTIVITIES

4.1 Material

0.5% aqueous solution of iodophore compound (Betadene/ iodine)
Spirit / Alcohol swab & 2x2 inches sterile gauze/cotton pieces.

4.2 Procedure

4.1.1 Ask the donor to wash his both arm if required.
4.1.2 Ask the donor to lie on the donor couch in comfortable position.
4.2.3 Apply blood pressure cuff with tubing directed upward as high on upper arm as possible to prevent interference with skin preparation (at least 2 inches from anticubital area) and inflate to 60 to 80 mm Hg.
4.2.4 Position donor arm in a naturally extended (not hyper extended)position on armrest at a 20- 30 degree angle from the body

4.2.5 Place the gripper(ball) in donor hand and request the donor to squeeze firmly

4.2.6 Palpate the anticubital area for veins with the help of middle three fingers to differentiate anatomical structures. Palpate along the course of vein in the upward direction and little skin over the vein may be pulled with the thumb of the opposite hand to determine how well supported or rolling the vein may be.

4.2.7 Select the skin entry site ¼ to ½ inch below or to the side of the intended point of entry for bevel up technique or directly over the vein for bevel down technique.

4.2.8 Secure the donor arm position by reminding the donor not to move at the time of puncture.

4.2.9 Release the cuff, clean the proposed site of venipuncture in the following manner.

4.2.10 Scrub area at least 4 cm (1.5 inches) in all direction starting at the intended site of the venipuncture and moving outward concentrically for a minimum of 30 seconds with methylated spirit /alcohol swab and let it dry & then clean with 0.5 % aqueous solution of iodophore compound (Betadine) in same manner.

4.2.11 Remove the iodine by spirit swab in same manner & let it dry.

4.2.12 Dispose off used swabs into a waste bin meant for bio hazardous materials

5.0 Precaution

4.1 After the skin has been prepared it must not be touched again. Do not re palpate at the intended site.

4.2Discard the used swab. If it is physically soiled / contaminated, take a new swab & repeat skin preparation procedure as detailed earlier.

4.3 The antiseptic solution must be allowed to dry before the phlebotomy is performed.

4.4 Always wear the protective gloves.

6.0 REFERENCES

6.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.

6.2 Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)

6.3 Standards for blood bank and blood Transfusion Services NACO 2007

Number DC/0	Effective Date	Page 4	Author	Authorized
Version - 04	Review period 2years	No. of copies:5	Approved By	Revision Date
Location **Bleeding room**			**Subject** **Bleeding of donor**	
Function **Phlebotomy and Blood collection**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated area • Master Copy	

1.0 PURPOSE

To collect one unit of whole blood preferably within 4 to 10 minutes.

2.0 SCOPE

Blood unit collected in less than 10 minutes of duration prevent clot formation and provides better platelet yield thus resulting in maximum component utilization and therapeutic benefit to the patients.

3.1 **RESPONSIBILITIES**

3.1.1 Prior to phlebotomy, phlebotomist must complete and verify the following on both the blood bag &record.

3.1.2 Identical coded & numeric donor identification number to ensure rationality on all copies of donor record, primary and satellite bags, pilot tubes or inline pouch any special identification tags, e. g. autologous etc.

3.1.3 Product bag labels are dry and show no evidence of defacement or leaking anticoagulant.

3.1.4 Take possession of donor record (registration form), blood bag and pilot tubes. Keep all in close proximity to donor to prevent confusion between donors.

3.1.5 Ask donor to state name. Compare with name recorded on donor record because phlebotomist is the last person to verify donor identification and acceptability.

3.1.6 Check the values of blood collection monitor & set the weighing scale at zero prior to phlebotomy.If required reset the value according to the desired blood volume to be drawn (eg- 350 or 450ml) .

3.1.7 If an ordinary weighing scale is being used calculate the final weight of the bag according to the formula:

(weight of empty bag in gms **+** volume collected in ml X 1.053)

Responsible person
Nursing staff/doctor posted in phlebotomy room.

4.0 ACTIVITIES

4.1 Material

4.1.1 Sterile blood bag containing anticoagulant CPDA-1 or Additive Solution with integrally attached needle.
4.1.2 Blood collection monitor.
4.1.3 Sterile gauze and clean instruments (scissors& artery forceps)
4.1.4 Pilot tubes for sample collection
4.1.5 Tube stripper
4.1.6 Dielectric sealer.

4.2 Method

4.2.1 Veni-puncture
4.2.2 Clamp the tubing before starting the procedure.
4.2.3 Make the vein prominent by re inflating blood pressure cuff to 40-60 mmHg. Higher pressure may occlude the radial pulse and increase the possibility of spurting or clot formation.
4.2.4 Advice donor to alternatively squeeze and relax hand on hand gripper with the final squeeze held firmly around the hand gripper.
4.2.5 Twist off needle cover and inspect needle for defects. This should be done out of donor view if possible. Discard needle cover.
4.2.6 In case of failure to clamp the tube segment, anticoagulant may cause burning sensation at venipucture site and should be flicked from the needles. Hold needle upright out of donor's view and tap the needle hub once with opposite hand.
4.2.7 Pull skin tight below the venipuncture site with the thumb or fingers of the free hand. This helps prevent sudden movement of the arm and anchors the vein and minimizes discomfort.
4.2.8 With bevel up hold needle at 30 to 45 degree angle and pierce skin with a smooth quick thrust at the previously selected point of entry. For the phlebotomist convenience skin puncture may also be made with the bevel down.
4.2.9 When bevel is completely under the skin lower the angle of needle to 10 degrees or less and then with a steady push advance needle to penetrate vein wall. One of the flowing sensation may be felt or observed.
4.2.10 Sudden cessation of tissue resistance indicates vein entry.
4.2.11 Abrupt rupture of tissue if the vein is overly distended.
4.2.12 Sliding or deflection from the target if the needles slip over or under the vein.
4.2.13 Back flow of blood into tubing adjacent to needle if using bag with an in line pouch indicated vein entry.

4.2.14 Thread needle approximately ½ inch inside lumen of vessel to maintain secure position which will lessen chances of clot formation.

4.2.15 Release clamp and observe blood flow through tubing and the time it entered the bag.

4.2.16 Instruct donor to relax grip and then to rhythmically squeeze hand gripper every 5-10 seconds, relaxing between squeeze.

4.2.17 Secure the needle to donor's arm with a 2-3 inch strip of leucoplast across the hub or on the tubing near the hub for optimal positioning. To prevent displacement of needle, additional short tape should be placed across the tubing lower on that arm.

4.2.18 Reduce pressure of blood pressure cuff to 30-40 mmHg to facilitate blood flow and promote donor comfort.

4.2.19 Monitor filling of blood bag

4.2.20 Mix blood with anticoagulant. In the absence of automatic shaker, take special care to thoroughly agitate blood with anticoagulant in the primary bag by gently lifting and tilting the bag every 100 ml or at least once every minute.

4.2.21 Ask donor to squeeze his fingers in the absence of resilient hand gripper to cause contraction of fore arm muscle and resultant increase venous flow. This should be done slowly and repeatedly every 5-10 seconds.

4.2.22 Precaution should be taken to avoid puncturing artery by feeling pulsating sensation of artery.

4.2.23 Time duration to be noted from the time blood enters the bag unit till flow is discontinued.

4.2.24 Sign donor record .

4.2.25 Give special care to slow collection, i. e. unit not half full within 3-4 minutes.

4.2.26 Check tubing patency and pathway for obstructions.

4.2.27 Assess donor for pain (venospasm) and /or reaction symptoms or change in arm position.

4.2.28 Make careful bevel adjustments. Bevel may be moved away from the vein wall or valve. Slight withdrawal or turning the needle or advancing of the needle may also prove beneficial; however avoid contaminating the needle.

4.2.29 Observe donor squeezing technique. Tense muscles in the shoulder or tight squeezing without a period to fill the vein between squeeze can be counterproductive.

4.2.30 Check tubing to determine if above technique were successful.

4.2.31 Interact with donor to prevent reaction.

4.2.32 Allow collection to continue uninterrupted until desired volume has been collected with the following exceptions.

4.2.33 Hematoma formation during and after venipuncture.

4.2.34 Complaints of discomfort .

4.2.35 Donor reaction, excessive collection time .

4.2.36 A second venipuncture may be performed if :

- 50 ml or less blood has already been collected
- The donor allows a second venipuncture.
- An acceptable vein is available on the opposite arm.

4.2.37 Discontinue blood collection .
4.2.38 Watch for a signal or a filled unit by automatic blood collection monitor.
4.2.39 Put the clamp on the tube segment and remove blood pressure cuff and needle anchoring tapes. Put a dry cotton ball and hold lightly over venipuncture site. Remove needle from vein smoothly and quickly with cotton ball held lightly over site. To prevent vessel injury do not apply pressure to the site until the needle is removed.
4.2.40 Extend donor arm vertically from shoulder.
4.2.41 Immediately following needle removal retain 2cotton ball to provide pressure to the venipuncture site.
4.2.42 Instruct donor to use the opposite hand to apply firm pressure over and slightly distal to the venipuncture site to obstruct venous flow.
4.2.43 Encourage donor to maintain a relaxed position rather than tensing the muscle. This precaution will minimize bleeding into the area.
4.2.44 Post donation strip the tubing clean so that all blood from tubing enters the bag and can be well mixed with anticoagulated blood.
4.2.45 Seal the tubing using dielectric sealer.
4.2.46 Discard the needle into sharp container
4.2.47 Measures to prevent clot formation

- Proper mixing
- Limit collection time for use of whole blood to 15 minutes. If collection time is more than 15 minutes and unit is still not completed discontinue the procedure and label the unit as under collected. Allowable limit for transfusion purpose needs to be within± 10% of the optimum volume.

4.3 STORAGE

4.3.1. Place unit and pilot tubes in proper storage area.
4.3.2. Blood unit collected for platelet preparation will be kept in utility tray at room temperature in component lab maintaining the room temperature between 22° to 24°C.
4.3.3. Blood unit collected in single blood bags will be kept immediately at 2-6 °C in blood bag refrigerators.
4.3.4. **Pilot tube sample will be stored at 2-8 degree Celsius in refrigerator.**
4.3.5. Precaution to take while storing blood in blood bank refrigerator
4.3.6. Open the refrigerator door only when required.
4.3.7. The blood unit should be kept in an upright position in blood bag holding cassettes on the shelf to allow the cold air to move around freely inside the refrigerator. They should never be packed tightly.
4.3.8. Tested and untested blood should be kept in separate blood bank refrigerators.
4.3.9. Blood should be arranged group wise for easy access.
4.3.10. Do not keep food or drink in the blood bank refrigerators.
4.3.11. Defrost the blood bank refrigerators periodically to maintain uniform cooling.
4.3.12. Once the testing report is obtained shift the non reactive quarantine blood to the tested blood refrigerator.

4.3.13. Blood should be issued to the patient according to serial number (FIFO) and / or clinical indication of transfusion.

4.3.14.

5.0 RECORDS

5.1 Complete record and charting.
5.2 Signature of phlebotomist .
5.3 Successful or unsuccessful procedure.
5.3.1. Second venipuncture if required.

5.3.2 Donor complaints sign & symptoms of donor reaction and treatment given.
5.3.3 Length of donation
5.3.4 Documentation of reason for unsuccessful venipuncture.

7.0 REFERENCES

7.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
7.2 Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
7.3 Standards for blood bank and blood Transfusion Services NACO 2007

Number DC/0	Effective Date	Page 2	Author	Authorized
Version -	Review period 2years	No. Of copies:5	Approved By	Revision Date
Location **Donor Complex**			**Subject** **Donor safety**	
Function **Post Donation care**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated area • Master Copy	

1.0 PURPOSE

To ensure donor safety

2.0 SCOPE

The donor needs to be observed after blood collection in order to attend to any adverse reactions in the immediate post donation period.

3.0 RESPONSIBILITY

Nursing staff &doctor posted in Phlebotomy attends to the donor.

4.0 ACTIVITY

4.1 Material

4.1.1 Sterile swab

4.1.2 Band – aid

4.1.3 Leaflet for post donation instruction.

4.1.4 Donor card

4.2 Procedure

4.2.1 Inspect venipuncture site while arm is in elevated position, lift cotton swab. Affix Band aid, if no oozing of blood. If donor is allergic to tape use roller gauze.

4.2.2 Provide extra rest time for first time donor.

4.2.3 Never leave the donor to wear his shoes or tie the laces immediately after the blood donation to prevent donor reaction due to lowering of head.

4.2.4 Help donor to get down from the couch to prevent arm strain and hematoma. Direct the donor to refreshment room for rest and light refreshment.

4.1.3 He is asked to report if any discomfort or dizziness immediately to the attending staff nurse and should be attended by the doctor.

4.1.5 If he feels any discomfort he is advised accordingly. A record of donor reaction is noted in the Donor reaction register. An emergency kit is kept ready at all time.

4.1.6 If donor is comfortable after having refreshments he / she is allowed to leave after sufficient time.

4.1.7 Donor should be thanked for valuable contribution and handed over donor card and encouraged for future donations.

4.1.8 Post donation instruction leaflet should be given before leaving the refreshment room.

5.0 RECORD

5.0.1 Record any adverse reaction in the donor reaction register.

5.0.2 Note the leaving time of the donor in the donor feedback register

6.0 REFERENCE

6.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.

6.2 Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)

6.3 Standards for blood bank and blood Transfusion Services NACO 2007

Number DC/012	Effective Date	Page 2	Author	Authorized
Version - 04	Review period 2years	No. of copies:5	Approved By	Revision Date
Location **Donor Complex**			**Subject** **Blood Collection**	
Function **Adverse donor reaction and their management**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated area • Master Copy	

1.0 PURPOSE

Any adverse reaction in the immediate post – donation period requires to be referred to the Blood Bank Officer.

2.0 SCOPE

Management of donor reaction to prevent any adverse effect & to advice about future blood donations.

Although the donation process is usually safe and uncomplicated, occasional donor may experience adverse reaction from the process.

3.1 RESPONSISIBILITY

3.1.1 To give proper attention and care to the donor

3.1.2 Assess the donor reaction and provide proper management

Responsible person

The doctor in charge of donor complex

3.2 MATERIALS REQUIRED:

Emergency tray- as per Annexure-

4.0 ADVERSE REACTIONS

4.1 FAINTING OR VASOVAGAL REACTION

Symptoms:

4.1.1 Mild : Sweating , Weakness, Dizziness, Pallor. The skin of donor feels cold to touch due to fall in blood pressure.

4.1.2 Moderate to Severe: Loss of consciousness, convulsion and involuntary passage of urine / faeces.

4.2 Management:

4.2.1 Whenever there is a first sign of donor reaction, immediately deflate the blood pressure cuff and withdraw needle.

4.2.2 Raise the legs above the level of the donor's head by placing him on his back. Ensure sufficient airway.

4.2.3 Loosen tight clothing.

4.2.4 The donor should respond by coughing which will elevate the blood pressure.

4.2.5 Give inhalation of aromatic spirit of ammonia, if required

4.2.6 Apply cold compresses to donor head.

4.2.7 Check periodically – BP, pulse and Respiration

4.2.8 Administer 1V normal saline or dextrose saline infusion if hypotension is prolonged.

4.3 NAUSEA AND VOMITING

4.3.1 **Management:**

4.3.2 Make the donor comfortable and relaxed.

4.3.3 Ask him to breathe slowly and deeply.

4.3.4 Apply cold compresses to the donor forehead and back of neck.

4.3.5 Turn the donor head to side to avoid aspiration of vomitus.

4.3.6 If he vomits, provide towel or tissue paper.

4.3.7 Give water to clean / rinse his / her mouth.

4.3.8 If vomiting still persists give antiemetic medication

4.4 TWITCHING OR MUSCULAR SPASM

Anxious donor with Hyperventilation may suffer from this type of reaction.

4.4.1 **Management:**

Ask the donor to breathe into paper bag. Do not give oxygen

4.5 CONVULSIONS: (OCCURS VERY RARELY)

Management:

4.5.1 Tilt the donor head to the side. Place tongue depressor between the teeth to avoid him from biting the tongue.

4.5.2 Prevent the donor from injuring himself/ herself

4.5.3 Ensure sufficient airway.

4.5.4 Call Emergency to shift the patient, if required

4.6 HAEMATOMA

4.6.1 **Management:**

4.6.2 Release the tourniquet / BP cuff pressure immediately.

4.6.3 Apply pressure on venipuncture site & withdraw the needle from the vein.

4.6.4 Place 3-4 sterile gauge pieces or cotton swabs over the haematoma.

4.6.5 Raise arm above the heart level.

4.6.6 Apply pressure with the tip of the fingers for 7-10 minutes.

4.6.7 Apply ice to reaction site

4.6.8 Advice the donor for topical application of Thrombophob ointment, if bruising occurs.

4.7 SERIOUS CARDIAC PROBLEM (extremely rare)

4.7.1 **Management:**

4.7.2 Cardiopulmonary resuscitation if the donor is in cardiac arrest.
4.7.3 Continue the CPR until medical aid arrives
4.7.4 Call Emergency to shift the patient.

5 **RECORD**

Record all the events and management in the donor reaction register

6 **REFERENCES**

6.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
6.2 Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
6.3 Standards for blood bank and blood Transfusion Services NACO 2007

Number DC/0	Effective Date	Page 2	Author	Authorized
Version –	Review period 2years	No. of copies:5	Approved By	Revision Date
Location **Apheresis Room**			**Subject** **Single needle Platelet Apheresis on Cell Separator**	
Function **To collect Single Donor Platelet from a healthy donor**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated area • Master Copy	

1.0 PURPOSE

To prepare single donor platelet concentrate in the blood center with the help of Cell separator for use in patients with thrombocytopenia.

2.0 SCOPE

The single unit of platelet apheresis of high dose minimizes the exposure to multiple donors & maximizes the benefits of the particular blood component to the patient.

3.0 RESPONSIBILITY

3.1 The procedure is performed by trained technical / medical personnel under the supervision of Doctor who is responsible for the smooth conduction of the procedure.

3.2 Make sure that all pre requisites have been completed.

4.0 ACTIVITIES

4.1 Principle

4.1.1 The apheresis procedure is based on the principle of dual step centrifugation based on the specific gravity of blood component.

4.1.2 In spite of single arm procedure the separation is in continuous flow.

4.1.3 The procedure takes place in the form of cycles, the inlet cycle and the return cycle.

4.1.4 During the Inlet cycle whole blood is drawn from the donor and separated in the centrifuge compartment. 40% of the drawn blood goes into the reservoir bag.

4.1.5 During the Return cycle red cell and plasma are returned back to the donor and the blood in the reservoir bag enters the centrifuge compartment for the separation, thus maintaining continuous flow separation.

4.1.6 Platelets are retained inside the collection container of the machine and the remaining blood is returned back to the donor.

4.2 Prerequisites:

4.2.1 Selection of donor

4.2.2 Properly screened healthy donor is selected for donation. Donor selection follows the standard blood donation criteria and in addition should qualify the following criteria:

6.2.5 Pre procedure platelet count should be above 150, 000 / µl

6.2.6 Interval between donations should be at least 48 hours and donors should not undergo platelet- pheresis more than twice in a week .

6.2.7 If the donor donates a unit of whole blood or if it becomes impossible to return the donor red cell during the previous platelet apheresis at least three months should be elapsed before a subsequent plateletpheresis procedure unless the extra corporeal volume is < 100 ml.

6.2.8 Donors who have taken aspirin medication 72 hour prior to donation should not be accepted as a Plateletpheresis donor.

6.2.9 He should not be fasting prior to the procedure

6.2.10 A Prominent and easily accessible anticubital vein on one of the arm is selected for the apheresis.

6.2.11 For routine platelet apheresis the donor is tested for the Transfusion transmitted diseases .

6.2.12 Other donor parameters such as Hematocrit, Height and weight are required to be entered into the machine.

6.2.13 Informed written consent is taken from the donor

4.4 Material

Single needle closed system cell separator kit compatible with the aphaeresis system.
Material for phlebotomy Please refer to the SOP for phlebotomy
EDTA Vaccutainer for product sampling
Tube sealer
Emergency medicine tray.

4.4 Procedure as per product manual

5.0 RECORD

5.1 Apheresis consent form, qualification criteria, TTI screening report to be maintained along with aphaeresis detail in register.

6.0 REFERENCES

6.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.

6.2 Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)

6.3 Standards for blood bank and blood Transfusion Services NACO 2007

Number DC/	Effective Date	Pages 1	Author	Authorized
Version -	Review period 2 years	No. of copies:5	Approved By	Revision Date
Location Donor Complex			Subject Blood Collection Monitor	
Function **To describe the steps to use the Blood Collection Monitor**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated area • Master Copy	

1.0 PURPOSE

To describe the steps to use the Blood Collection Monitor

2.0 SCOPE

It is important to know to know the proper steps to use the Blood Collection Monitor to ensure optimal collection and mixing of blood with the anticoagulant in the blood bag.

3.0 RESPONSIBILITY

Responsible Person

Doctor /Staff Nurse/ phlebotomist

4.0 ACTIVITY

4.1 Material

Blood Collection Monitor and Blood Bag

4.2 Method

4.2.1 First switch on the BCM by pressing on/off switch on the side of the machine

4.2.2 Place the blood bags together in BCM plate properly and route in the clamp

4.2.3 Set the collected blood volume (350/450ml) on BCM by pressing up/ down arrow button on key pad.

4.1.4 Then after prick press the start key button when blood just starts to come in the blood bag.

4.1.5 If donor flow rate is below 20ml/min and above 120ml/min then BCM gives an alarm.

4.1.6 You can mute the alarm by pressing the start button.

4.1.7 Check the blood prick site and adjust the needle if required/ possible.

4.1.8 After optimal amount of blood is collected the BCM automatically clamps the tube as well as gives the complete process alarm.

4.1.9 The clamps will release automatically when the filled bags are lifted from the BCM plate for 2 seconds.

5.0 PRECAUTION

5.1 Ensure that the bags are placed properly in the BCM plate

5.2 The tubing of the blood bag should not be stretched.

6.0 REFERENCES

Manufacturer's instruction

Standard Operating Procedures : Blood & Components Preparation & Storage.

Number CL/0	Effective Date	Page 2	Author	Authorized
Version -	Review period 2 years	No. of copies:5	Approved By	Revision Date

Location **Blood component preparation Lab**	**Subject** **Blood component preparation and storage.**
Function **Calibration of Refrigerated Centrifuge.**	**Distribution** • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy

Sl. No.	Subject	Sop. No.
1.	Blood component Preparation and storage- Calibration of Refrigerated Centrifuge	CL/001
2.	Blood component preparation and storage- pre centrifugation preparations	CL /002
3.	Preparation of Blood components (PRP) using Triple bag with additive solution.	CL/003
4.	Preparation of Blood components using Automated Component Extractor	CL/004
5.	Preparation of Blood components using Triple bag with additive solution	CL/005
6.	Labeling of Blood Components	CL/006
7.	Blood components Preservation & Storage	CL/007
8.	Preparation of washed Packed Red Cells.	CL/008

1.0 PURPOSE

To deliver highest product yield in the shortest time, at the lowest possible spin, so as to cause the least amount of trauma to each product, while at the same time maintaining the optimal temperature for component viability.

2.0 SCOPE

Successful preparation of blood components requires adequate but not excessive centrifugation. The equipment must perform in a consistent and dependable manner.

3.0 RESPONSIBILITY

3.1 After successful calibration, programming should be locked.

3.2 If further change in the pre- existing program or new additional programming is required, the designated technologist must inform the technical supervisor to unlock the centrifuge for additional / resetting of program.

3.3 Cross check the temperature of the rotors at least once in a month.

Responsible Person

Technical Supervisor and Technologist

4.0 ACTIVITY

4.1 Material

4.1.1 Refrigerated Centrifuge
4.1.2 Blood bag Buckets
4.1.3 Blood unit 450ml /350ml
4.1.4 Dry Rubber material for balance
4.1.5 Double pan weighing balance
4.1.6 Plasma Extractor- automated/ manual
4.1.7 Equipment & Reagents for measuring the quality standards

4.2 Method

Differential centrifugation speeds commonly used are of two types:

4.2.1 Note:- **Final adjusted time will include acceleration but not deceleration time and varies according to manufacturer guidelines.**

4.2.2 Optimal speed of centrifuge can be achieved by calculating relative centrifugal force (in g) using following formula:

Relative centrifugal force (RCF) in g = 28.38 R (rpm / 1000)^2

R= radius of centrifuge rotor in inches

4.2.3 Set the preliminary values in refrigerated centrifuge according to blood bags to be centrifuged & set the program.

4.2.4 Centrifuge the bags at their designated program

4.2.5 Prepare the desired component. If the quality control values are optimal as per the standard guidelines, finalize the program & lock it. If the quality control values are not satisfactory, repeat the exercise by slight modification in acceleration, time & RPM till the desirable results are obtained.

4.2.6 The same method is being used to set different programmes for blood component preparation using different blood bags. Based on these calculations, six programmes are calibrated and standardized for the preparation of various blood components.

Program No.	Acceleration (Minutes)	Deceleration (Minutes)	Speed (RPM)	Time (minutes)	Temperature °C
1	9	4	3800	09	04
2	4	4	1800	12	22
3	9	4	3200	12	22
4	9	5	3200	12	04
5	9	5	3900	08	22
6	7	4	1080	06	22

4.3 Application of programs

Application	Type of Bag	Program No.
Packed red blood cells (PRBC) & Fresh frozen Plasma(FFP)	Double	1
Packed red blood cells (PRBC) & Platelet rich Plasma(PRP)	Triple (By 1st spin)	2
Leucoreduced red cells(LPRC) & Fresh frozen Plasma(FFP)	T/B & I/B (1st spin)	5
Platelet concentrate(PC) & Fresh frozen Plasma(FFP)	Triple (By 2nd spin)	3
Packed red blood cells (PRBC) & Platelet rich plasma(PRP)	Double	2
Leucoreduced platelets(LPC)	T/B & I/B (By 2nd spin)	6
Cryopoor Plasma(Factor VIII deficient & plasma) Cryoprecipitate(Cryo)	Thawed FFP	1

5.0 RECORD

The entire final set of programmes is stored in the memory of refrigerated centrifuge software.

Date of calibration and its validity Period (One year).

6.0 REFERENCE

- Technical Manual AABB 13th edition.
- Training Module for Blood Bank Officers 2006-2007
- Transfusion Medicine Technical manual DGHS 2003, 2nd edition

Number CL/002	Effective Date	Page 2	Author	Authorized
Version - 0	Review period 2 years	No. of copies:5	Approved By	Revision Date
Location **Blood component preparation Room**			**Subject** **Blood component preparation and storage.**	
Function **Pre- Centrifugation Preparation.**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy	

1.0 PURPOSE

To monitor weight of blood bags and for balancing centrifuge buckets with blood bags.

2.0 SCOPE

Properly balanced centrifuge processes the blood with smooth conduction of centrifugation & is able to separate the different layers of cellular & acellular blood components to achieve the optimal result.

3.0 RESPONSIBILITY

3.1 Blood after collection is kept at room temperature (22 ± 2° C) if it is to be processed for blood components, especially for preparation of platelet concentrate.

3.2 Get the blood shifted to the Component Lab from Phlebotomy room immediately and process it within 6 – 8 hrs of collection.

3.3 Physical checking of the total number of units collected in different blood bags.

3.4 Any discrepancy will be entered in register and brought to the notice of the Senior Technologist.

3.5 Check the donor ID on primary and its satellite bags to ensure they are having the same number.

3.6 Decision for components preparation from under / over collected units will be taken after discussing with senior Technologist or Blood Bank Officer.

3.7 Enter the blood components to be prepared in component register according to donor ID number based on selected blood bag for component preparation.

Responsible Person Technical supervisor/ Senior Technologist posted for the blood component preparation

4.0 ACTIVITY

4.1 Material

4.1.1 Collected whole blood units
4.1.2 Double pan Balance
4.1.3 Centrifuge Buckets
4.1.4 Balancing dry rubber material
4.1.5 Refrigerated Centrifuge

4.2 Procedure

4.2.1 Cross check the donor ID in primary and satellite bags.
4.2.2 Keep blood bags into the centrifuge buckets in vertical position keeping the primary bag away from central partition & Satellite bags near the center of the bucket.
4.2.3 Fold upper 1/3 with the ports & integrally connected tubing of satellite bags.
4.2.4 Balance in pairs using dry rubber balancing material.
4.2.5 Switch on the power supply and centrifuge switch after ensuring that lid of centrifuge is closed.
4.2.6 Wait for digital display to stabilize. Select the proper programme to enter the values and let the desired temperature be attained.
4.2.7 Once the temperature is achieved, open the lid and carefully place the weighed and balanced bags. Be sure that primary bag is kept outwards.
4.2.8 The centrifuge is carefully lidded and shut.
4.2.9 Recheck the values.

5.0 RECORD

Before packing the buckets & loading the blood component, make entry in the register.

6.0 REFERENCE

Technical Manual AABB 13th edition.
Transfusion Medicine , Technical Manual, DGHS, 2nd Edition, 2003

Number CL /0	Effective Date	Page 2	Author	Authorized
Version - 0	Review period 2years	No. of copies:5	Approved By	Revision Date
Location Blood component preparation Room			Subject Blood component preparation and storage.	
Function Preparation of Blood components (PRP) using Triple bag with additive solution			Distribution • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy	

1.0 PURPOSE

To deliver highest product yield in the shortest time, at the lowest possible spin, so as to cause the least amount of trauma to each component, while at the same time maintaining the optimal temperature for component viability.

2.0 SCOPE

Successful preparation of blood components requires adequate but not excessive centrifugation. The equipment must perform in a consistent and dependable manner.

3.0 RESPONSIBILITIES

3.1 After successful calibration, programming should be locked.

3.2 If further change in the pre- existing program or new additional programming is required, the designated technologist must inform the technical supervisor to unlock the centrifuge for additional / resetting of program.

3.3 Cross check the temperature of the rotors at least once in a month.

4.0 ACTIVITY

4.1 Material

4.1.1 Triple Blood Bags with additive solution (350ml / 450 ml)
4.1.2 Deep Freezer (- 80^{o}C)
4.1.3 Collected whole blood units
4.1.4 Double pan Balance
4.1.5 Centrifuge Buckets
4.1.6 Balancing dry rubber material
4.1.7 Refrigerated Centrifuge
4.1.8 Plasma expressor

4.2 Procedure

4.2.1 Centrifuge blood bags at program No.2

4.2.2 After centrifugation gently take out the bags, keep them straight and place the primary bag in upright position on a plasma expressor and the spring allowing the plate of the expressor to be in contact with the bag.

4.2.3 Break the seal / closure of the primary bag and allow platelet rich plasma [PRP] to extract in the empty satellite bag. Plasma does not go into the SAGM bag because manufacturer provides it with a locked mechanism.

4.2.4 Remove maximum volume (200-300ml.) of PRP into empty satellite bag.

4.2.5 Apply the metal or plastic clip to the tubing of PRP bag.

4.2.6 Transfer the SAGM to the primary bag containing PRBC after breaking the seal and hanging the SAGM bag at a height using component / IV stand.

4.2.7 Seal the tubing in between primary and two satellite bags at three places, cut it as packed red cells and store at their required temperature.

4.2.8 Give second centrifuge to the rest of two satellite bags after proper balancing at program No. 3. This time platelets are the heaviest product in PRP bag and they settle at the bottom of bag.

4.2.9 After second centrifuge, place the satellite bag [containing platelets poor in plasma and sediment platelet] into plasma expressor. Express plasma into the plasma bag.

4.2.10 Take out the bag from plasma expressor, remove the clamp and allow supernatant to flow back in to the plasma bag leaving behind 50 to 70 ml of plasma with sediment platelets in the bag.

4.2.11 Seal the tubing of both the bags at three places and cut from the middle one and store the plasma at their required temperature.

4.2.12 Leave the platelets concentrate at the table top for 30 minutes to one hour at room temperature and then shift to the platelet agitator.

5.0 RECORD

Enter record in blood component register.

6.0 REFERENCE

- Technical Manual AABB 13th edition.
- Transfusion Medicine , Technical Manual, DGHS, 2nd Edition, 2003

Number CL / 004	Effective Date	Page 2	Author	Authorized
Version - 04	Review period 2 years	No. of copies: 5	Approved By	Revision Date
Location **Blood component preparation Room**			**Subject** **Blood component preparation and storage.**	
Function **Preparation of blood components using Automated component extractor** (ACE)			**Distribution** • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy	

1.0 Purpose

Preparation of buffy coat removed blood components.

2.0 Scope

Transfusion of buffy coat removed blood component minimizes the possibilities of post transfusion febrile non – hemolytic reaction.

3.0 Responsibility

3.1 Blood after collection is kept at room temperature (22 ± 2° C) if it is to be processed for blood components, especially for preparation of platelet concentrate.

3.2 Get the blood shifted to the Component Lab from Phlebotomy room immediately and process it within 6 – 8 hrs of collection.

3.3 Physical checking of the total number of units collected in different blood bags.

3.4 Any discrepancy will be entered in register and brought to the notice of the Senior Technologist.

3.5 Check the donor ID on primary and its satellite bags to ensure they are having the same number.

3.6 Decision for components preparation from under / over collected units will be taken after discussing with senior Technologist or Blood Bank Officer.

3.7 Enter the blood components to be prepared in component register according to donor ID number based on selected blood bag for component preparation

Responsible Person

Technical supervisor/ Senior Technologist posted for the blood component preparation.

4.0 ACTIVITY

4.1 Material

Quadruple Blood Bags (Top & Bottom or Integral filter bags) (450ml) with additive solution

4.1.1 Automated component extractor.
4.1.2 Metallic plates/dies with hooks.
4.1.3 Deep Freezer (- 80^{o}C)
4.1.4 Collected whole blood units
4.1.5 Double pan Balance
4.1.6 Centrifuge Buckets
4.1.7 Balancing dry rubber material
4.1.8 Refrigerated Centrifuge

4.2 Procedure

4.2.1 Switch on the power of centrifuge and set the temperature of centrifuge to the desired temperature at least 15 minutes before starting the centrifuge, so that desired temperature is attained depending upon the components to be prepared.

4.2.2 Balance the blood bags carefully before placing in the centrifuge cups using double pan balance. Weight to be adjusted using rubber material.

4.2.3 Centrifuge the bags at program no.5

4.2.4 Once the centrifuge has stopped take out the cups / buckets containing blood bags carefully and place on the working table.

4.2.5 Switch on Automated component extractor(ACE) which is pre-programmed .

4.2.6 Wait till the ACE Screen display requisite Protocol, load unit & - Start to process.

4.2.7 Place the bags on ACE machine. Hang the primary bag on hanger making sure that the label of the bag faces to the fixed wall of the ACE.

4.2.8 Place the bags upper port tubing through the top clamp before its Y junction and the bottom tube of SAGM bag through the bottom left clamp. Clamp any one of the top transfer bags by plastic clip.

4.2.9 Press START - Screen display break cannulas & accordingly break the top cannula primary bag first and later the cannula of SAGM bags.

4.2.10 The Plasma starts flowing from top into the satellite bag & RBC from bottom into the SAGM bag.

4.2.11 Once the process is almost complete machine makes a click sound. Immediately place a plastic clamp on the top tubing adjacent to top clamp.

4.2.12 The display show Heat sealing – the bottom tubing in the clamp gets heat – sealed.

4.2.13 Once the procedure is complete the display shows “remove the bag”.

4.2.14 Remove the bags from the machine.(For top and bottom bags with filter, after removing the bags from the ACE put the RBC bag in a hanger for filtering leukocytes.) Separate the SAGM Red cell bag. Seal the plasma bag on the di-– electric sealer & separate the same.

4.2.15 The Plasma bags separated is labeled as FFP. The date of expiry is written on the bag (1 Year from date of collection) and keep it into the Deep Freezer till cleared for Infectious Markers.

4.2.16 Place the buffy coat bags attached with satellite bag, which were hung for 1 hour to 1 hr 30 mins and place it in the bucket

4.2.17 Balance the buckets using rubber material

4.2.18 Use metallic Plate /disc with hooks to keep the buffy coat bag in an upright position in the cups/ buckets.

4.2.19 Place the buckets in the centrifuge.

4.2.20 Centrifuge at program No. 6

4.2.21 After centrifugation take out the buckets carefully from centrifuge & place on the working table.

4.2.22 After the protocol is selected, hang the bag on the hanger.

4.2.23 Install the top tubing on the upper optic assembly & close the cover completely.

4.2.24 Follow the instruction on the display panel.

4.2.25 Press Start Remove Unit.

4.2.26 Put the expiry date on platelet concentrate, taking day of collection as zero days.

4.2.27 After automatic heat sealing, remove the bag.

4.2.28 Once the components have been prepared the magnetic plates should be removed from the ACE & kept at designated place.

4.2.29 At the end of day-to-day work, switch off the machine

4.3 Precautions

4.3.1 Transfer the air before sealing

4.3.2 Always check the magnetic plate before placing it on the ACE

5.0 RECORD

Enter record in blood component register & blood bank software

6.0REFERENCE

- **User manual of ACE**
- Technical Manual AABB 13th edition.
- Transfusion Medicine , Technical Manual, DGHS, 2nd Edition, 2003

Number CL /00	Effective Date	Page 2	Author	Authorized
Version - 04	Review period 2 years	No. of copies:5	Approved By	Revised Date

Location Blood component preparation Room	Subject Blood component preparation and storage.
Function Preparation of Cryoprecipitate from FFP	Distribution • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy

1.0 PURPOSE

To deliver highest product yield in the shortest time, at the lowest possible spin, so as to cause the least amount of trauma to each product, while at the same time maintaining the optimal temperature for component viability.

2.0 SCOPE

Successful preparation of blood components requires adequate but not excessive centrifugation. The equipment must perform in a consistent and dependable manner.

3.0 RESPONSIBILITIES

3.1 After successful calibration, programming should be locked.

3.2 If further change in the pre- existing program or new additional programming is required, the designated technologist must inform the technical supervisor to unlock the centrifuge for additional / resetting of program.

3.3 Cross check the temperature of the rotors at least once in a month.

Responsible Person

Technical Supervisor and Technologist

4.0 ACTIVITY

4.1 Material

4.1.1 Cryo bath.

4.1.2 Refrigerated Centrifuge Deep Frezer -40 to – 80^{o}C

4.1.3 Double pan Balance

4.1.4 Centrifuge Buckets
4.1.5 Balancing dry rubber material
4.1.6 FFP

4.2 Procedure

4.2.1 The FFP prepared frozen at – 80^{o}C for a minimum of 24 hour period is preserved in deep freezer.

4.2.2 Take out the frozen unit from the deep freezer & leave it at 4^{o}Cfor 24 hrs. Once the tubing becomes soft put the FFP bag in Cryo bath at 4^{o}C till it becomes slushy.

4.2.3 Give second centrifuge after proper balancing at program No.1 as per SOP –for Calibration of refrigerated centrifuge

4.2.4 After centrifuge place the satellite bag containing Cryo poor plasma and sediment Cryo precipitate into plasma expressor.

4.2.5 Take out the clip and allow supernatant to extract into the bag leaving 20 to 25 ml of plasma with sediment cryo precipitate.

4.2.6 Take out the bag from plasma expressor, seal the tubing of both the bags at three places and cut from the middle one and store the Cryo poor plasma & Cryoprecipitate at their required temperature(-40 to - 80^{o}C).

5.0 RECORD

Enter record in blood component register

6.0 REFERENCE

- Technical Manual AABB 13th edition.
- Transfusion Medicine , Technical Manual, DGHS, 2nd Edition, 2003

Number CL -006	Effective Date	Page 2	Author	Authorized
Version – 04	Review period 2 years	No. of copies:5	Approved By	Revision Date
Location Blood component preparation Room			Subject Labeling of blood Components	
Function To ensure safe blood supply			Distribution • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy	

1.0 PURPOSE

1.1 To label the blood component bags in order to accurately relate the blood component to the donor.

1.2 To easily trace the component.

2.0 SCOPE

The blood after it is collected remains in quarantine and is released for transfusion only after all tests (grouping & infectious markers) are completed. Before these blood components are taken on inventory for use, the units are affixed with printed labels meeting Drug regulatory norms. The label is required for identification & retrieval of blood units for use, disposal and follow up in case of adverse reactions. The sero-reactive units are discarded observing the Bio-medical waste rules.

3.0 RESPONSIBILITY

It is the responsibility of the designated Technologist from the component laboratory to label the blood & component units.

4.0 ACTIVITY

4.1 Material

4.1.1 Pre-printed adhesive component labels for all component printed as per regulatory norms.

4.1.2 The printed labels are colour coded for all component as per Blood Group A- yellow, group B- pink , group O- blue and group AB- white . Negative labels also have the same color code.

4.2 Method

4.2.1 After collection and processing, the component units remain in quarantine storage areas.

4.2.2 Once all reports of blood group and TTI testing are ready, place the bags on a table in chronological order.

4.2.3 Segregate those which are found reactive for any TTI or found unsuitable for use .

4.2.4 Date of collection and date of expiry is very important. The expiry date depends on the type of components. In case of triple and quadruple bag with additive solution the expiry date is 42 days and for double and single bags, it is 35 days. In case of triple or quadruple bags if for some reason the component could not be separated then the date of expiry will be 28 days as the primary bags contains CPD as anti-coagulant. The day of collection is considered the day zero for calculating the expiry dates.

4.2.5 After the bags are labeled ask a second Technologist to double check the number, group & result of infectious markers on the bags tallying them with the records.

4.2.6 Enter all labeled blood bags group wise in the stock book, which is also maintained group wise. In the stock book keep a footnote for use.

4.2.7 Label FFP &Cryo deficient plasma and platelet concentrates in the same manner. Cryoprecipitate labels do not indicate blood groups.

4.2.8 All plasma components have an expiry date of one year. The expiry date of the platelet concentrate is 5 days taking date of production as 0 day.

5.0 RECORD

Enter all labeled bags numbers in the inventory (stock register) of units for use

5 REFERENCE

- Technical Manual AABB 13th edition.
- Transfusion Medicine , Technical Manual, DGHS, 2nd Edition, 2003
- NABH Blood Bank standards 2nd Edition, June 2013

Annexure 1

LABELLING REQUIREMENT

COMPONENT	**The label specimen receptacles and containers should conation at least the following information**.
GENERAL LABELLING REQUIREMENT	
	Specify
	• *Nature of whole blood or component (or intended component) • *Volume of component • *Unique numeric or alphanumeric donation identification • Rh Group, specifying "Rh D positive "if D positive or " Rh D Negative if D negative. • *Date of collection / preparation and expiry date • Temperature of storage • Name of anticoagulant (not required for frozen, deglycerolized, rejuvenated, or washed red cell. • Approximate volume of blood collected from the donor. • That the donor or component must not be used for transfusion if abnormal hemolysis or any other deterioration is detected. • That blood or component must be administered through a 170 – 200 um filter.
SUPPLEMENTARY SPECIFIC LABELLING REQUIREMENT	
	Specify
Fresh frozen Plasma	Whether component is from whole blood or aphaeresis donation Volume and composition of anticoagulant used Whether quarantined or screened Storage Temperature Instruction for use after thawing.
Platelets Aphaeresis	• Volume of content & storage temperature • Whether or not leukocyte depleted.
Plateles recovered	• Donation no. (if platelets are pooled labeling system must allow identified of original donation.) • Whether or not leukocyte depleted; • Storage Temperature
Red cells	• Name and volume of component; • Composition anticoagulant preservative solution.
Red cell buffy coat Removed	Composition of anticoagulant solution Composition and volume of additive solution
Red cells in Additive sol.	• Composition and volume of additive solution.
Red cells leukocyte Depleted	• Composition of anticoagulant solution
Red cells Washed	• Time of preparation of expiry • Composition and volume of anticoagulant solution
Whole Blood	• Volume of preparation

	• Composition and volume of anticoagulant solution.
Red Cells Aphaeresis	• Dose of red cells or the total Hb content • No. of unit equivalents. Option for split no. 2 unit collection is administration to more than one recipient. • Whether or not leukocyte depleted; • Composition of anticoagulant solution • Composition of volume of any additive solution

Number CL /00	Effective Date	Page 2	Author	Authorized
Version - 04	Review period 2 years	No. of copies:5	Approved By	Revised Date
Location **Blood component preparation Room**			**Subject** **Blood component preparation and storage.**	
Function **Blood Component Preservation & storage.**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy	

1.0 PURPOSE

Once the blood is collected from the donor, there begins a series of changes in vitro that alter physiological properties. The anticoagulant / preservative solution is vital in maintaining the viability, preservation & storage as it:

1.1 Ensures that blood & products maintains their in vivo properties
1.2 Inhibits the growth of microorganism in the product
1.3 Prevents clotting of the product.

2.0 SCOPE

The expiration date for red cell is determined by the number of red cell surviving in transfused patient 24 hour after transfusion.

1.0 RESPONSIBILITY

This is the responsibility of TTI Technologist to physically check the labeled blood component & remove all the components prepared from units found positive for the infectious markers from quarantine area.

All the non-reactive units to be taken into stock must be shifted to designated storage area/equipment (Blood Bank refrigerators/ walk-in-cooler, deep freezers, platelet agitator & incubator).

2.0 ACTIVITY

2.1 **Material**

2.1.1 Blood bank refrigerators
2.1.2 –80^{o}C freezer
2.1.3 Platelet Agitator & incubator
2.1.4 Utility tray / Transportation boxes
2.1.5 Transportation trolley
2.1.6 Plasma Storage bags/ canisters.

2.2 **Method**

2.2.1 All untested unit should be kept in the quarantine area

2.2.2 After testing is over, release the fully tested unit. Transfer those deemed suitable for clinical use from quarantine area to the stock area after labelling.

2.2.3 Label those found unsuitable for use with a biohazard label and keep for disposal.

2.2.4 The shelves of Blood Bank refrigerators should be pre – labeled for group specific components. Store whole blood & red cell concentrates in color – coded blood holding plastic casettes & keep them group wise & according to the expiry dates (FIFO)

2.2.5 The shelves of platelet agitator should be pre – labeled for group specific platelets.

2.2.6 Keep Single Donor Platelet (through Plateletpheresis) in a Platelet agitator with incubator at $22^{o}C \pm 2^{o}C$ in a separate shelf.

2.2.7 Take due care to maintain sterility of all components by keeping all storage areas clean.

2.2.8 Monitor to ensure the storage conditions to be appropriate for each product.

2.2.9 Monitor the temperature of all storage areas with continuous graphic recorder.

2.2.10 Change the charts every week and archive them. Check the alarm system every month.

5.0 RECORD

Record all blood / components released for use in Designated stock Register & unsuitable units to be discarded in the disposal Register.

6.0 REFERENCE

- Technical Manual AABB 13th edition.
- Transfusion Medicine , Technical Manual, DGHS, 2nd Edition, 2003
- NABH Blood Bank standards 2nd Edition, June 2013

Annexure 2

Blood Component Storage

Blood component	Storage temperature	Length of storage	Transportation temperature	Transportation time
Whole blood (for transfusion as whole blood)	2-6^{o}C±1^{o}C	≤35 days with adenine supplemented anticoagulant (CPDA)	2-10^{o}C	≤ 24 hour
Whole blood (for component preparation)	1-6^{o}C 22± 2^{o}C (if to be used for the preparation of platelets	Up to 8 hour before use Up to 24 hour before use		≤ 24 hour
Red cells	2-6^{o}C±1^{o}C	≤35 days in adenine supplemented anticoagulant (CPDA)	2-10^{o}C	≤24 hour
Red cell in additive solution (SAGM)	2-6^{o}C	≤ 42 days in adenine supplemented anticoagulant (CPDA)	2-10^{o}C	≤24 hour
Red cell in additive solution (SAGM) buffy coat (leucoreduced)	2-6^{o}C	42 days	2-10^{o}C	
Plasma ,fresh frozen	-18 to-80^{o}C	1year	Similar to storage temperature	
Liquid plasma	2-6^{o}C	40days	Similar to storage temperature	
Plasma, thawed	Thawed between 30-37^{o}C	Transfused as soon as possible	Similar to storage temperature	
Plasma , cryoprecipitate depleted	-18 to-25^{o}C	4 years	Similar to storage temperature	
Buffy coat pooled	Not suitable for storage If unavoidable : 22± 2^{o}C	Administered as soon as possible after collection with maximum storage of 24 hour	Similar to storage temperature(with continuous flat bed gentle agitation	
Platelets (single unit concentrates , buff coat	22± 2^{o}C	5 days (close system) with continuous flat bed gently agitation < 6 hour (after open	Similar to storage temperature(with continuous flat bed gentle	

removed , apheresis)		system)	agitation	
Platelet ,pooled	22± 2°C	5 days (Closed system) 24 hrs. (open system)	Similar to storage temperature(with continuous flat bed gentle agitation	
Cryoprecipitate	--18 to-25°C	1 year	Similar to storage temperature	

3.IMMUNOHEMATOLOGY

INDEX

Number RCS/00	Effective Date	Page 2	Author	Authorized By
Version - 0	Review period 2years	No. of copies:5	Approved By	Revised Date

Location	**Subject**
Red cell Serology Lab.	**Preparation of Red Cell Suspension and storage**
Function **Preparation of RBC suspension of appropriate concentration for a given test**	**Distribution** • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy

1.0 PURPOSE

To obtain accurate results in red cell serology e.g. – ABO Blood grouping, antibody screening and compatibility testing.

2.0 SCOPE

When red cells are washed thrice with normal saline & remained suspended within their required concentration, it maintains the sensitivity of the test which may otherwise be diminished if cell suspension is of high concentration.

3.0 RESPONSIBILITY

3.1 Always prepare the suspension from anti- coagulated blood.

3.2 Do not take cell from clotted blood by breaking the clot. If anti coagulated sample is inadequate or hemolysed, request the treating doctor to send fresh EDTA sample.

3.3 Do not cut short the washing steps.

Responsible Person

Designated Technician posted in Red cell Serology Lab.

4.0 ACTIVITY

4.1 Principle

The concentration of erythrocytes in a saline suspension is important for the accuracy of testing in the blood bank. All technicians who work in the Blood Bank should follow the methods for preparing accurate suspension.

4.2 Material

4.2.1 The specimen should consist of 2-3ml of EDTA blood.

4.2.3 12x75 mm test tubes

4.2.3 Glass Pasteur pipettes

4.2.4 Normal saline (0.9%)

4.2.5 Rubber stopper or plastic cap for closing the test tubes

4.2.6 Test tube stand

4.2.7 Centrifuge machine with timer.

4.2.8 Preparation of an approximate 2%-5% red blood cell suspension

4.2.9 Place 1or 2 ml of anti coagulated blood in a 12x75 mm test tube

4.2.10 Fill 3/4th of the test tube with normal saline and centrifuge at 1500- 2000 rpm for 1-2 minutes.

4.2.11 Aspirate or Decant the supernatant saline

4.2.12 Repeat washing (steps 2 and 3) until the supernatant saline is clear (usually 3 times).

4.2.13 Pipette 10ml of normal saline (0.9%) into a clean test tube.

4.2.14 Add 0.2 ml/0.5ml (for 2% and 5% respectively) of the washed packed red cell button to the 10ml of saline.

4.2.15 Cover the tube until use and store the red cell suspension at $4^{o}C$.

4.2.16 Before use, invert the tube several times until the cells are in suspension.

4.3 LIMITATIONS

Hemolysis of red blood cell from improper washing may result in false result. A cell suspension that is heavy or of light concentration may produce false positive or false negative result.

4.4 PRECAUTIONS

The supernatant fluid should be clear after the final wash. The red cell suspension should not be used after 24 hrs. of preparation .

5.0 RECORD

Record the result of group specificity and sensitivity of the prepared cells with anti sera in use

6.0 REFERENCE

- Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
- Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
- Standards for blood bank and blood Transfusion Services NACO 2007

Number RCS/002	Effective Date 01-01-2016	Page 2	Author	Authorized
Version -	Review period 2years	No. of copies:5	Approved By	Revised Date

Location **Red cell Serology Lab.**	**Subject** **Preparation of Red Cell Suspension and storage**
Function **To Prepare RBC suspension of pooled cells of appropriate concentration for red cell serology**	**Distribution** • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy

1.0 PURPOSE

This procedure is used for preparation of RBC suspension of pooled cells of appropriate concentration

2.0 SCOPE

This procedure applies to all testing that requires red cell suspension preparation.

3.0 RESPONSIBILITY

Every morning shift duty technician must prepare A, B, O, red cell suspension for the day's use.

4.0 ACTIVITY

4.1 Principle

The concentration of erythrocytes in a saline suspension is important for the accuracy of testing in the blood bank. All technicians who work in the Blood Bank should follow the methods for preparing accurate suspension

4.2 Material

4.2.1 The specimen should consist of 2-3ml of EDTA blood.
4.2.2 12x75 mm test tubes
4.2.3 Glass Pasteur pipettes/ micro-pipette
4.2.4 Normal saline (0.9%)

4.2.5 Rubber stopper or plastic cap for closing the test tubes
4.2.6 Test tube stand
4.2.7 Lab Centrifuge machine with timer

4.3 Procedure

4.3.1 Take 3 clean test tubes & label them as 'A; 'B' and 'O'.
4.3.2 Take equal volume of packed cells, from at least 3- 5 samples of each blood group into the respective tubes e.g. 100 µL packed cell from each of the 5 samples of known group 'A' into tube labeled 'A' and similarly for group B and O
4.3.3 Suspend the cells in Normal saline by filling 3/4th of the test tube with normal saline.
4.3.4 Centrifuge the tubes at 1500-2000 rpm for 1-2 minutes or till the RBCs settle down to form a button.
4.3.5 Decant the supernatant saline completely.
4.3.6 Repeat from steps 3, 4& 5 at least 3 times more or till the supernatant saline is clear.
4.3.7 Label 3 fresh tubes as 'A', 'B' & 'O' respectively.
4.3.8 For preparation of 5% suspension: 1 ml of Normal saline + 50µL of washed packed cells is used
4.3.9 The final red cell suspension is stored at 4° C

3.0 RECORD

3.1 Enter the unit numbers from which pooled cells are prepared in the register
3.2 Record the results of testing with the Antisera used in register
3.3 Enter the manufacturer's name and batch number of the Antisera used

4.0 REFERENCE

4.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
4.2 Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
4.3 Standards for blood bank and blood Transfusion Services NACO 2007

Number RCS/00	Effective Date	Page 2	Author	Authorized
Version -	Review period 2years	No. of copies 5	Approved By	Revised Date
Location **Red cell Serology Lab**			**Subject** **ABO Grouping**	
Function **Reverse ABO Grouping by Tube Technique**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy	

1.0 PURPOSE

Detection of natural occurring A& B antibody in serum

2.0 SCOPE

To determine the correct ABO group of individual and ensure the reliability of the result, this procedure describes the method of detection of A & B antibodies in the serum. . It provides guidance for the use of standard red cell suspension in order to confirm the final group after comparing the results with forward grouping.

3.0 RESPONSIBILITY

Designated technician posted in red cell serology lab

4.0 ACTIVITY

4.1 Material

4.1.1 12 x 75 mm test tube
4.1.2 Pooled Red cell of know blood group (A,B & O)
4.1.3 Normal saline (0.9%)
4.1.4 Pasture Pipette/ Micro pipette
4.1.5 Test tube stand
4.1.6 Lab Centrifuge machine with timer

4.2 Method

4.2.1 Label three test tubes –A, B and O

4.2.2 To all the three test tubes, add two drops of serum to be tested using a Pasteur pipette.

4.2.3 To the tube labeled A, add one drop of thoroughly mixed A 5% red cell suspension To the tube labeled B, add one drop of thoroughly mixed 5% B red cell suspension To the tube labeled O, add one drop of thoroughly mixed 5% O red cell suspension Mix and centrifuge all three test tubes at 1000 rpm for 1 minute

4.2.4 Re-suspend all the cells by gentle agitation and examine for agglutination both visually& microscopically if required.

4.3 Reporting results

Agglutination indicates that an antibody specific for either the A or B antigen is present in the serum or plasma being tested. The blood grouping of the individual based on the presence or absence of agglutination is presented in the table.

Reactions of donor unit/patients Serum & Reagent red cells

A1 cell	B Cell	O cell	Antibody	Blood group
+	**+**	**-**	Anti A & Anti B	O
-	**+**	**-**	Anti B	A
+	**-**	**-**	Anti A	B
-	**-**	**-**	No antibody	AB
+	**+**	**+**	Anti- H	Oh (Bombay blood group)

(+)Agglutination (-) No Agglutination

4.4 Note

Agglutination in test tube O cell requires additional laboratory work up. Do not finalize the blood group until problem is solved.

4.5 Procedure Notes

4.5.1 A hemolysed specimen is unsuitable for this test.

4.5.2 If the expected result of both forward and reverse typing are not compatible, either a variation in the donor unit/ patient's sample or a technical error may exist.

4.5.3 Discrepancies in serum (reverse) grouping can be due to additional / unexpected antibody or missing antibodies.

4.5.4 If the results of forward & reverse grouping do not tally, do not enter the final grouping before the discrepancy is resolved.

4.5.5 When result of cells and serum tests for ABO do not match: inform the Blood Bank Officer. The discrepancy must be investigated.

4.5.6 If the blood is from donor unit, the unit must not be released for transfusion until the discrepancy is resolved.

4.5.7 In case of urgency, when the blood is from a potential recipient, it may be necessary to administer group O packed cells of the appropriate Rh group, till the discrepancy is resolved.

5.0 RECORD

5.1 Enter the result of reverse grouping in the Blood grouping register. Enter the result of patient's grouping in the blood requisition form and patient grouping register.

6.0 REFERENCE

6.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.

6.2 Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005

6.3 Standards for blood bank and blood Transfusion Services NACO 2007

Number RCS/00	Effective Date	Page 3	Author	Authorized
Version -	Review period 2years	No. of copies:5	Approved By	Revised Date

Location **Red cell Serology Lab**	**Subject** **ABO Grouping**
Function **Forward ABO grouping by tube technique**	**Distribution** • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy

1.0 PURPOSE

Identification of A&B antigens on red cells

2.0 SCOPE

To determine the correct ABO group of an individual and ensure the reliability of the result, this procedure describes the method of detection of ABO antigens on the red cell. It provides guidance for the use of blood grouping reagents (Antisera) in order to confirm the final group after matching the results with reverse grouping.

3.0 RESPONSIBILITY

3.1 Check antiserum against freshly prepared known pooled cells and enter the result / grade at specified column in forward grouping register.
3.2 If the grading is two grades lower than the result of quality check, change Antisera.
3.3 The blood sample should be tested as soon as possible. If there is delay in testing, the blood sample should be stored at 2º to 6º C.
3.4 Store Antisera at 2º to 6ºC after performing the tests
3.5 The Blood Bank Officer should verify all discrepant or doubtful results

Responsible person

Technician posted in red cell serology lab

4.0 ACTIVITY
4.1 Principle

The ABO blood group (A, B, AB and O) represent the antigens expressed on the red cells of each group. When an antibody and its corresponding antigen are combined in vitro, agglutination of the red cells occur. A natural antibody and its corresponding antigen are not normally present in the same blood specimen.

4.2 Material

4.2.1 Commercial blood grouping anti sera (From two different sources or batches) : Anti – A, Anti – B and Anti AB
4.2.2 12x75 mm test tubes
4.2.3 Normal saline (0.9 % NaCl)
4.2.4 Micro-pipettes or Pasteur pipette
4.2.5 Microscope
4.2.6 Lab centrifuge, Timer

4.3 Quality Control

Reagent Antisera should be tested daily with red cells of known antigen (see Daily Reagent Quality Assurance)

4.4 Procedure

Check the patients' name and identification numbers on the blood specimen and requisition from.

4.4.1 Prepare a 2- 5 % suspension of the patient's red cell in normal saline (see preparation of Red cell suspension).

4.4.2 Label four 12x75 mm test tubes A, B, AB and C (Auto-control)
4.4.3 To the tube labeled A, add 1 drop of antiserum A
4.4.4 To the tube labeled B, add 1 drop anti B antiserum.
4.4.5 To the tube labeled AB, add 1 drop of anti AB antiserum.
4.4.6 To the tube labeled C, add 1 drop of Patient's Serum.
4.4.7 Using a pipette, add 1 drop of the patient's red cell suspension to each of the test tubes.
4.4.8 Mix well and centrifuge the test tubes for 1 min. at1000 rpm.
4.4.9 Re-suspend the cell with gentle agitation and examine microscopically for agglutination.

4.5 Reporting of results

4.5.1 Agglutination of erythrocytes with a specific antiserum is interpreted as a positive (+) test result and indicates the erythrocytes have the corresponding antigen. A negative (O) test indicates that the corresponding antigen is not present. For various agglutination patterns and their respective interpretation refer table.

Control	Anti -A	Anti - B	Anti - AB	Result Blood Group
-	+	-	+	**A**
-	-	+	+	**B**
-	+	+	+	**AB**
-	-	-	-	**O**

(+) Agglutination (-) No. Agglutination

Grading of Antibody Reaction(Agglutination & Haemolysis)

Grade	Description
Negative O	No Aggregates
Mixed Field (MF)	Few isolated aggregates. Mostly free floating cell supernatant appears (Pinkish)
Weak +	Tiny aggregates that are barely visible macroscopically, many free erythrocytes, turbid and reddish Supernatant.
1+	Many small aggregates just visible macroscopically with free erythrocytes, turbid and reddish supernatant.
2+	Medium sized aggregates, some free erythrocytes clear supernatant.
3+	Several large aggregates, some free erythrocytes, clear supernatant.
4+	All erythrocytes are combined into one solid aggregate clear superman.

Haemolysis is considered as positive reaction.

4.5.2 Precautions

4.5.2.1 Each manufacturer provides, with each package of antiserum, detailed instruction for the use of Anti A and Anti B. These directions vary in details; therefore it is important to follow the direction for the specific antiserum in use.

4.5.2.2 Do not rely on the color of dyes to identify reagent Antisera. All tubes must be properly labeled.

4.5.2.3 Do not perform tests at the temperature higher than the room temperature (20- 24^{o}C)

4.5.2.4 Confirm observation of agglutination with a microscope, if required

4.5.2.5 Record result immediately after observation, on a work sheet/ register

4.5.2.6 Remember that contaminated blood specimens, reagents or supplies may interfere with the test result

4.0.7 Limitations

Antisera prepared from human sources are capable of detecting A1 and A2 group; however weak subgroups of A may only be detected with anti- AB

5.0 RECORD

5.1 Enter the result of donor grouping in the donor forward grouping register

5.2 Enter the result of patient's grouping in the blood requisition form, patient grouping register.

6.0 REFERENCES

6.1 Transfusion Medicine, Technical Manual, DGHS, 2nd edition.

6.2 Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)

6.3 Standards for blood bank and blood Transfusion Services NACO 2007

Name of Blood Bank
Name of Hospital Delhi-1100
License no

Number RCS/00	Effective Date	Page 2	Author	Authorized
Version -	Review period 2years	No. of copies:5	Approved By	Revised Date

Location Red cell Serology Lab	Subject Preparation of sensitized Red Cells
Function Preparation of Sensitized red cell for the validation of Coomb's Test	Distribution • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy

1.0 PURPOSE

To check the validity of Coomb's test

2.0 SCOPE

Sensitized cell (also known as check cell) check the presence and efficacy of Coomb's sera used.

3.0 RESPONSIBILITY

Responsible Person

Technologist

4.0 ACTIVITY

4.1 Principle

Sensitized Red Cell is a cell, which has been coated by the specific Antibody but no visible agglutination (Macroscopic or microscopic) is observed.

4.2 Material Required

4.2.1 12x75 test tubes 4.2.1
4.2.2 AHG Serum
4.2.3 Anti D Serum lgG
4.2.4 Rh (D) Positive Cell
4.2.5 Normal Saline (0.9%)
4.2.6 Pasteur pipette/ micro pipette
4.2.7 Test tube stand

4.2.8 Lab Centrifuge with timer
4.2.9 Microscope

4.3 Procedure

4.3.1 Dilute the Anti D (lgG) Serum with Normal Saline.

4.3.2 Do the dilution of anti sera using doubling dilution technique as follows:
Take 11 test tubes and label then 2,4,8,16,32,64,128,256,512,1024, serially and leave the last one blank.

4.3.3 Add three drop of normal saline (0.9%) in all test tube
4.3.4 Add three drop of anti D lgG in first test tube and mix it well with the help of Pasteur pipette.
4.3.5 Transfer three drop from first tube to the next one and up to the tenth tubes and remove one volume from this tube into the last one. Do not discard save it for the use if further dilution is needed.
4.3.6 Add one drop of 5% suspension of pooled O cell into each tubes.
4.3.7 Incubate at 37^{o}C for 30 minute.
4.3.8 Centrifuge at 1000 rpm for one minute and see agglutination
4.3.9 A last positive reaction is the titer of antibody which is selected.
4.3.10 Prepare the anti D solution of selected dilution.
4.3.11 Add 8 drops (0.5ml) of 5% washed cell suspension of O Rh (D)
4.3.12 To the Positive cells add 16 drop (1ml) of selected dilution of Anti –D Serum.
4.3.13 Mix and incubate 37^{o}C for 30 min.
4.3.14 Look for agglutination; wash the cell three times with a large volume of the saline. Decant the suspension of sensitized cell in saline.
4.3.15 Add 1-drop AHG serum to 1 drop of the 5% washed sensitized cells.
4.3.16 Mix& spin immediately at 1000 rpm for one min.
4.3.17 Cell should show +2 agglutination (if there is no agglutination the whole procedure is repeated by taking less diluted anti – D serum.)

Note:- These cell are used as positive control for testing Coomb's Serum/ Coomb's Test.

5.0 RECORD

5.1 Enter the donor unit number from which check cells are prepared in the X – Match register.
5.2 Record the result &date of testing with the Coomb's sera used in X – Matching register.
5.3 Enter the manufacturer's name and batch number of the Coomb'sSera.

6.0 REFERENCE

- Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
- Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
- Standards for blood bank and blood Transfusion Services NACO 2007

Name of Blood Bank
Name of Hospital Delhi-1100
License no -

Number RCS/00	**Effective Date**	**Page** 3	**Author**	**Authorized**
Version -	**Review period** 2years	**No. of copies:5**	**Approved By**	**Revision Date**
Location Red cell Serology Lab		**Subject** Direct Coomb's Test		
Function Performing Direct Coomb's Test by Tube Technique.		**Distribution** • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy		

1.0 PURPOSE

To identify coating of red cell with auto antibodies in particular lgG & C3d

2.0 SCOPE

The direct antiglobulin test is generally used to determine if red cells have been coated, in vivo with immunoglobulin, complement or both, in particular lgG & C3d. Washed red cells from a patient or donor are tested directly with AHG reagent. Test is used in investigating autoimmune hemolytic anemia; drug induced Hemolysis, hemolytic disease of newborn & allows identification of immune reaction to recently transfused red cells.

3.0 RESPONSIBILITY

In addition to the daily reagent quality assurance check, each test must have a saline control run concurrently to rule out auto agglutination. Coomb's control / check cells must be added to each test that does not exhibit agglutination.

Responsible Person

Technologist /final reporting by Blood Bank Officer

4.0 ACTIVITY

4.1 Principle

The direct antiglobulin test is based on the principle that antiglobulin antibodies induce in vitro agglutination of red cells coated with immunological bound antibodies. After red cells are washed to remove free plasma protein from the test mixture, they are tested directly with reagents containing anti lgG and anti C3d (broad spectrum /polyspecific. Monospecific Antisera, for example anti lgG, anti C3d, that are specific for immunoglobulin/ complements, may also be used).

4.3 Specimen

4.3.1 No special preparation of the patient is required before specimen collection. The patient must be positively identified when the specimen is collected. The time of collection and the Doctor's initials should be written.

4.3.2 Specimen should consist of 3-5 ml of clotted blood and 2ml of EDTA blood drawn by aseptic technique.

4.3.3 Newborn or infants sample may be collected as red cell in normal saline or from a cord blood sample.

4.3.4 If a delay in testing occurs, the specimen must be stored at 2-6° C. Antibodies dependent for their detection upon the binding of complement may not be detected if aged serum or plasma from an anti-coagulated sample is used. Hemolysis of the specimen is undesirable.

4.4 Material

4.4.1 Antiglobulin serum (Polyspecific)

4.4.2 IgG sensitized erythrocytes (Coomb's control cells / check cells)

4.4.3 12x75 mm test tubes

4.4.4 Pasteur Pipettes

4.4.5 Normal saline (0.9%)

4.4.6 Test tubes stand

4.4.7 Lab Centrifuge with timer

4.4.8 Microscope.

4.5 Quality control

In addition to the daily reagent assurance check, each test must have a saline control run concurrently to rule out auto agglutination. Coombs control / check cell must be added to each test that does not exhibit agglutination.

Agglutination of the Coombs control cells verifies the reactivity of the antiglobulin antiserum

4.6 Procedure

4.6.1 Prepare a 5% saline suspension of the patient's red cells.

4.6.2 Label one 12x75 mm test tube with the letters DAT. Label second 12 x75 test tubes as DAT control.

4.6.3 Using a disposable pipette, add 2 drops of the of the red cell suspension to each tubes.

4.6.4 Add 2 drops antiglobulin serum to DAT tube and 2 drops of normal saline to the control tube.

4.6.5 Centrifuge at 1000rpm for 1 minute.

4.6.6 Gently re suspend the cell and examine macroscopically and microscopically for agglutination. Grade the reaction and record results.

4.6.7 Verify the AHG activity in the DAT tube that demonstrates no agglutination.

4.6.8 Add one drop of Coombs control cells to the DAT tubes.

4.6.9 Centrifuge at 1000rpm for 1 minute.

4.6.10 Gently re suspend the red cell and examine macroscopically and microscopically for agglutination. If is not present, the test is invalid and must be repeated.

4.6.11 The control cells should not show agglutination. If agglutination is present in the control cells ,it indicates auto-agglutination

4.7 Reporting results

A negative test is demonstrated by the absence of agglutination in the DAT test tubes and a positive test is manifested by the presence of agglutination.

5.0 RECORD

Record the results of the test in patient's register.

6.0 REFERENCE

- Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
- Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
- Standards for blood bank and blood Transfusion Services NACO 2007

Name of Blood Bank
Name of the Hospital Delhi-1100
License no -

Number RCS/00	Effective Date	Page 2	Author	Authorized
Version - 04	Review period 2years	No. of copies:5	Approved By	Revised Date

Location Red cell Serology Lab	Subject Direct Coomb's Test
Function Performing Direct Coomb's Test by Column Agglutination Technique	Distribution • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy

1.0 PURPOSE

To identify the coating of red cells with antibodies, in particular lgG &C3d.

2.0 SCOPE

The direct antiglobulin test is generally used to determine if red cell have been coated, in vivo, with immunoglobulin, complements, or both. The DAT is used to demonstrate in vivo coating of red cell with globulins, in particular lgG & C3d. Red cells suspension from a patient or donor are tested directly with AHG reagent. Test is used in investigating autoimmune hemolytic, drug induced hemolysis, hemolytic disease of newborn & allo immune reaction to recently transfused red cells.

3.0. Responsible Person

Technologist / final reporting by Blood Bank Officer

4.0 ACTIVITIY

4.1 Principle

The direct antiglobulin test is based on the principle that antiglobulin antibodies induce in vitro agglutination of erythrocytes with immunological bound antibodies. After erythrocytes are washed to remove free plasma protein from the test mixture, they are tested directly with reagents containing anti IgG and anti C3d. (broad spectrum /polyspecific). Monospecific Antisera, for example anti lgG, anti C3d, anti C4 that are specific for immunoglobulin,complements, may also be used. The DAT procedure is clinically important in the diagnosis of conditions such as hemolytic anemia or hemolytic transfusion reactions. .

4.2 Material

4.2.1 Coombs Column agglutination Cards with six microtubes containing poly specific AHG (Anti- IgG+C3d)

4.2.2 LISS (Low ionic strength solution) for red cell suspension

4.2.3 Card centrifuge and incubator.

4.2.4 Micropipettes (10, 25, 50, & 1000 µL)

4.3 Preparation of cell suspension (0.8-1% red cell suspension):

4.3.1 Add 10 µL of packed Red Cells (washed once in Saline) in 1mlof LISS solution in a clean test tube and mix uniformly

4.4 Specimen

4.4.1 The specimen should consist of 3ml of EDTA blood.

4.4.2 Newborn or infants samples may be collected as red cells in normal saline or from a cord blood sample.

4.4.3 If a delay in testing occurs, the specimen must be stored at 2-6ºC. Antibodies dependent for their detection upon the binding of complement may not be detected if aged serum or plasma is used. Hemolysis of the specimen is undesirable.

4.5 Procedure

4.5.1 Identify the appropriate microtube of a card with the patient's name/number.

4.5.2 Remove the aluminum foil from as many micro tubes as needed.

4.5.3 Add 50µL of the patient's RBC suspension (0.8-1.0%).

4.5.4 Centrifuge the card in the Card centrifuge for 10 minutes.

4.5.5 Read the results.

4.6 Reporting results

The settling of all RBCs in the bottom of column indicates a negative DCT. If there is visible trapping of RBCs in between the column, that indicates positive DCT.

5.0 RECORD

Record the results of the test in the register

6.0 REFERENCE

- Manufacturer's Guidelines.
- AABB Technical Manual

- Principles & Practice of Transfusion Medicine, 1st Edition 2014

Name of the Blood Bank

Name of Hospital Delhi-1100

License no -

Number **RCS/008**	**Effective Date**	**Page** **3**	**Author**	**Authorized**
Version - 04	**Review period** **2years**	**No. Of copies:5**	**Approved By**	**Revision Date**
Location **Red cell Serology Lab**			**Subject** **Coomb's Testing**	
Function **Performing Indirect Coomb's Test by Tube Technique.**			**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Designated Lab.** • **Master Copy**	

1.0 PURPOSE

To detect the presence of auto or alloantibody in serum.

2.0 SCOPE

The two step procedure (ICT) is used to demonstrate in vitro reactions between red cells & antibodies that sensitize but do not agglutinate red cells that express the corresponding antigen. This is useful for detection and identification of antibodies, blood grouping & compatibility testing.

3.0 RESPONSIBILITY

3.1 Compare information on specimen to request form; first and last name, full hospital number, and date of collection. If this information is not identical, obtain a new specimen.

3.2 The test reagents should be monitored daily or at the time of use, and according to the Quality Control Procedure:

3.3 An auto control, a mixture of patient's red cells and serum, should be tested simultaneously with each antibody-screening test.

3.4 All negative antiglobulin reaction must be tested with Coombs control / check cells. A positive test result at this point will confirm that active antiglobulin was added to the test system and present when the original antiglobulin test was interpreted as negative. If a positive result is not obtained with the control cells, the test is invalid and must be repeated.

Responsible Person

Technologist, final reporting by Blood Bank Officer

4.0 ACTIVITY

4.1 Principle

4.1.1 The indirect antiglobulin test is done to determine the presence of sensitization of red cells with Ig G and / or complement in vitro. Serum from patients (surgical, obstetrical) or donor is tested with group O reagent blood cells from individual donors that represent a variety of the most common blood group antigens

4.1.2 Hemolysis or agglutination at any stage of the test demonstrates the presence of an antibody (a positive test) with specificity of a corresponding antigen on the reagent red blood cells. The absence of hemolysis and /or agglutination indicates that the serum being tested does not contain detectable antibodies directed at antigens present on the reagent red cells being used.

4.1.3 A positive reaction indicates that an antibody is present in the serum being tested. .If the antibody is a specific blood group antibody directed toward an antigen determinant that is absent from the patient's own erythrocytes, it may have been produced as a result of previous transfusions or pregnancies. Infrequently, an antibody may be present that reacts with all red cells tested, including the patient's own cells. This pattern of reactivity is typical of acquired hemolytic anemia.

4.2 Specimen

No special preparation of the patient is required prior to specimen collection. The patient must be positively identified when the specimen is collected. The specimen shall be labeled at the bedside and shall include the patient's first and last name, the date the specimen is collected and the patient's hospital identification number. The time of collection and the Doctor's initials should be written on the requisition form.

4.2.1 The required specimen is 3-5ml of clotted blood. The presence of hemolysis in the specimen makes the specimen unsuitable for testing.

4.2.2 Antibodies that depend on the binding of complement for their detection may not be detected if aged serum or plasma from an anti coagulated sample are used for an antibody detection testing. Sample for an antibody screening may be used up to 72hour after collection. The specimen must be stored at 2-6^{o}C and kept for 7 days

4.3 Material

4.3.1 12X75 mm test tubes

4.3.2 Micro-Pipettes

4.3.3 Normal saline (0.9 %)

4.3.4 Antiglobulin reagent Antisera

4.3.5 Coombs control or check cells (lgG- sensitized cells)

4.3.6 Reagent red blood cells (5% suspension of pooled O cells).
4.3.7 Lab Centrifuge & 37ºC water bath
4.3.8 Test tube stand
4.3.9 Microscope (optional)

4.4 Quality Control of Reagents

4.4.1 The test reagents should be monitored daily or the time of use, and according to the quality Control Procedure.
4.4.2 An auto control, a mixture of patient's red cells and serum, may be tested simultaneously with each test.
4.4.3 All negative anti-globulin reactions must be tested with Coombs control / check cells. A positive test result at this point will confirm that active anti-globulin was added to the test system and was present when the original anti-globulin test was negative. If a positive result is not obtained with the control cells, the test is invalid and must be repeated.

4.5 Procedure

4.5.1 Compare information on specimen to request form; first and last name, hospital number, and date. If this information is not identical, obtain a new specimen.
4.5.2 Prepare a 5% suspension of group O positive red cells
4.5.3 Centrifuge the clotted specimen for 5 minutes at 2500 rpm.
4.5.4 Determine by checking records if the patient has previously been tested.
4.5.5 Label two 12x75 mm test tubes.(sample and auto)
4.5.6 Using a pipette, add two drops of the patient's serum to each tube.
4.5.7 Add 1 drop of red blood cells (5% suspension) to the tubes
4.5.8 Add 1 drop of the patient red cells suspension to the tube labeled Auto-control.
4.5.9 Mix all tubes and centrifuge for 1 minute X 1000 RPM.
4.5.10 Gently re suspend the cells in the tubes and examine macroscopically for agglutination or hemolysis. If agglutination seen, it indicates presence of saline/complete antibodies.
4.5.11 If no agglutination is seen further steps are done.
4.5.12 Wash each tube three to four times with large volumes of 0.9% normal saline. Decant completely after each wash.
4.5.13 Add two drops of antiglobulin (AHG) reagent to each tube.
4.5.14 Mix each tube and centrifuge for 1 minute at 1000rpm.
4.5.15 Gently re suspend the cells and examine it macroscopically with the aid of magnification (Microscopic examination is optional).
4.5.16 To each tube that exhibits no agglutination add 1 drop of Coombs check cells. Mix well and centrifuge for 1 minute at 1000 rpm. Re- suspend the cells and examine macroscopically for agglutination. Record this result as control cells (CC).

4.6 Reporting results

4.6.1 Agglutination or hemolysis indicates a positive reaction. The absence of hemolysis or agglutination constitutes a negative test and indicates the absence of detectable antibodies to specific antigens present on the reagent red cells.
4.6.2 If the auto control demonstrates a positive reaction, the serum contains an autoantibody. The presence of an autoantibody can conceal an underlying alloantibody in the serum.
4.6.3 If the patients have been recently transfused, a mixed field reaction in the auto control suggests that an unexpected antibody directed at an antigen present on surviving donor cells is present.

4.6.4 A negative test is demonstrated by the absence of agglutination in the ICT test tube.

4.7 Limitation

The detection of antibodies in serum can be compromised if the ratio of serum to cells in the test , or the test or the length of incubation is incorrect. Failure to detect all antibodies in a serum can result from:

- Low – titred antibodies that is too weak to be detected by the methods and / or media being used.
- An antibody that may be exhibiting a dosage effect
- The lack of an antigen on the screening cells to an antibody in the serum.
- In rare, cases, the presence of an antibody directed at one of the antigen in the red cell suspend medium may cause false positive reaction.
- False- negative reactions can result from as little as 1/10,000of a drop of the original serum remaining after cell washing. This produces some neutralization of antiglobulin reagent.

5.0 RECORD

Record the results of test in the register.

6.0 REFERENCE

- Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
- Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
- Standards for blood bank and blood Transfusion Services NACO 2007

Name of Blood Bank

Name of Hospital Delhi-1100

License no -

Number RCS/00	Effective Date	Page 2	Author	Authorized
Version - 0	Review period 2years	No. of copies:5	Approved By	Revised Date

Location Red cell Serology Lab	Subject Coomb's Testing
Function Performing of Indirect Coomb's Test by Column Agglutination technique	Distribution • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy

1.0 PURPOSE

To detect the presence of antibodies in serum.

2.0 SCOPE

The procedure (ICT) is used to demonstrate in vitro reactions between red cells & antibodies that sensitize but do not agglutinate cells that express the corresponding antigen. This is useful for detection and identical of antibodies, blood grouping & compatibility testing.

3.0 RESPONSIBILITY

3.1 Compare information on specimen to request form; first and last name, full hospital number, and date if this information is not identical, obtain a new specimen.

3.2 Allow reagent to reach room temperature before use

4.0 ACTIVITY

4.1 Material

4.1.1 CAT Cards with six microtubes containing poly specific AHG (Anti- IgG+C3d)

4.1.2 LISS for red cell suspension

4.1.3 Card centrifuge and incubator.

4.1.4 Micropipettes (10, 25, 50, & 1000 µL)

4.2 Specimen

4.2.1 No special preparation of the patient's is required prior to specimen collection. The patient's must be positively identified when the specimen is collected. The specimen shall be labeled at the bedside and shall include the patient's first and last name, the

date the specimen is collected and the patient's hospital identification number. The time of collection and the Doctor's initials should be written on the requisition form.

4.2.2 Blood should be drawn by an aseptic technique and the specimen tested as soon as possible. Preferably, blood samples should be drawn into citrate, EDTA, Heparin or CPDA.Samples should be well centrifuged. When serum samples are used, the serum should be well cleared by centrifugation for 10 minutes at 1500g, before use. Hemolysis in the specimen makes the specimen unsuitable for testing.

4.2.3 Antibodies that depend on the binding of complement for their detection may not be detected if aged serum or plasma from an anticoagulated sample are used for an antibody detection testing.

4.2.4 Samples that are not for immediate testing should be stored at 2-6ºC after separation , for a maximum of 48 hrs

4.3 Procedure

4.3.1 Identify the appropriate micro-tube of a card with the patient's name/number. Remove the aluminum foil from as many micro-tubes as needed.

4.3.2 Pipette 50µl of the ready to use O cell suspension into the microtube.

4.3.3 Add 25 µL of test serum/plasma.

4.3.4 Incubate for 15 min at 37^{o}C in the CAT Incubator.

4.3.5 Centrifuge the card in the CAT centrifuge for 10 minutes.

4.3.6 Read the results.

4.4 Reporting results

The settling of all RBCs in the bottom of column indicates a negative ICT. If there is any visible trapping of RBCs in the column, it indicates a positive ICT.

5.0 RECORD

Record the results of test in register .

6.0 REFERENCE

- Manufacturer's Guidelines.
- AABB Technical Manual
- Principles & Practice of Transfusion Medicine, 1st Edition 2014

Name of Blood Bank

Name of Hospital Delhi-1100

License no -

Number **RCS/0**	**Effective Date**	**Page** **2**	**Author**	**Authorized**
Version -	**Review period** **2years**	**No. of copies:5**	**Approved By**	**Revision Date**
Location **Red cell Serology Lab**			**Subject** **Blood grouping and Rh typing**	
Function **Performing Blood Grouping by Column Agglutination Technique**			**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Designated Lab.** • **Master Copy**	

1.0 PURPOSE

Determination of the ABO/Rh blood groups combined with reverse grouping

2.0 SCOPE

To determine the correct ABO group of an donor/recipient and ensure the reliability of the result. This procedure describes the method of detection of ABO antigens on the red cell & corresponding naturally occurring antibodies in serum, as well as RhD determination.

2.0 RESPONSIBILITY

3.1 The technologist posted in Red cell serology lab is responsible for maintaining the equipments and switching it off after performing day's work.

3.2 For optimal results, freshly drawn blood samples must be used, preferably using EDTA anticoagulant. Where samples are not for immediate testing, store at 2-6° C for a maximum of 48 hours

3.3 The Blood Bank Officer should validate the results ,verify all discrepant or doubtful results.

4.0ACTIVITY

4.1 MATERIALS REQUIRED

- CAT Card " ABO/D + reverse typing cards" containing monoclonal anti-A1, anti-B and anti-D within the column. The Micro-tube ctrl is the negative control. Two micro-tubes with neutral media serve for reverse grouping with A1 & B cells.
    - LISS solution
    - Test cell reagents- A1 , B & O in a 0.8% ±0.1% suspension.
    - Auto-Dispenser
    - Micro-Pipette
    - Yellow Tips
    - Tubes for suspensions
    - CAT Centrifuge

4.2 Preparation of blood sample

Prepare a 5% red cell suspension by dispensing 0.5 ml of LISS in a clean test tube and adding 50 microlitre of Whole Blood or 25microlitre of packed cells to it. Mix well.

4.3 Procedure

1) Identify the Card with unique patient/donor no. /details.
2) Remove the aluminum foil from as many micro-tubes as required by holding the card in upright position.
3) Pipette 50 µl reagent A1 cells to microtube 5(A1).
4) Pipette 50µl reagent B cells to microtube 6(B).
5) Pipette 50 µl of patient's serum to both microtubes 5&6..
6) Pipette 10 µl of patient's 5% red cell suspension to microtubes 1-4 (A,B,D,ctrl)
7) Incubate at room temp for 10 min
8) Centrifuge the Cards for 10 mins in the CAT Centrifuge.
9) Read and record the results.

4.4 Interpretation

Positive: agglutinated cells forming a red cell line on the surface of medium or agglutinates dispersed in medium

Negative: compact button of cells on the bottom of the microtube.

4.5 Reactions for blood groups ABO

Anti-A	Anti-B	Blood group
+++to++++	negative	A
negative	+++to++++	B
+++to++++	+++to++++	AB
negative	negative	O

4.6 Reactions for reverse grouping

A1	B	Blood group
+to++++	negative	B
negative	+to++++	A
+to++++	+to++++	O
negative	negative	AB

4.7 Reactions for Rh D

5 RECORD

+++to++++	+-to ++	negative
RhD positive	RhD weak positive	RhD negative

Record in the register.

6.0 REFERENCE

- Manufacturer's Guidelines.
- AABB Technical Manual
- Principles & Practice of Transfusion Medicine, 1st Edition 2014

Name of Blood Bank
Name of Hospital Delhi-1100
License no -

Number RCS/00	Effective Date	Page 2	Author	Authorized
Version - 00	Review period 2years	No. of copies:5	Approved By	Revised Date
Location Red cell Serology Lab			Subject Antibody screening and identification	
Function Antibody screening and identification by Column agglutination technique			Distribution • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy	

0.0 PURPOSE
To detect the presence of unexpected antibodies in serum.

1.0 SCOPE
The procedure is used to demonstrate in vitro reactions between red cells & antibodies that sensitize but do not agglutinate cells that express the corresponding antigen. This is useful for detection and identification of unexpected antibodies in serum/plasma.

2.0 RESPONSIBILITY
3.1 Compare information on specimen with the request form; first and last name, hospital CR number, and date if this information is not identical, obtain a new specimen.

3.2 Allow reagent to reach room temperature before use
3.3 Responsible person: technologist in red cell serology lab.

3.0 ACTIVITY

3.1 Material

- Coombs CAT Cards with six microtubes containing poly specific AHG (Anti- IgG+C3d)
- LISS for red cell suspension
- Test cells reagent –I-II-III screening cell panel
- Test cell reagent 11 cell panel for identification.
- CAT centrifuge and incubator.
- Micropipettes (10, 25, 50, & 1000 µL)
- Red cell suspension(0.8-1%)-add 1ml of LISS to 10 µl of packed cells and mix gently in a clean test tube.

3.2 Specimen

3.2.1 No special preparation of the patient's is required prior to specimen collection. The patient's must be positively identified when the specimen is collected. The specimen shall be labeled at the bedside and shall include the patient's first and last name, the date the specimen is collected and the patient's hospital identification number. The time of collection and the Doctor's initials should be written on the requisition form.

3.2.2 Blood should be drawn by an aseptic technique and the specimen tested as soon as possible. Preferably, blood samples should be drawn in EDTA tube.

3.2.3 Samples should be well centrifuged. When serum samples are used, the serum should be well cleared by centrifugation for 10 minutes at 1500rpm, before use. Hemolysis in the specimen makes the specimen unsuitable for testing.

3.2.4 Antibodies that depend on the binding of complement for their detection may not be detected if aged serum or plasma from an anticoagulated sample is used for an antibody detection testing.

3.2.5 Samples that are not for immediate testing should be stored at 2-6ºC after separation, for a maximum of 48 hrs.

3.3 Procedure

3.4 ANTIBODY SCREENING

3.4.1 Identify the appropriate microtube of an CAT card with the patient's name/number. Remove the aluminum foil from as many microtubes as needed.

3.4.2 Pipette 50µl each of the ready to use I-II-III screening cell into the appropriate microtube.

3.4.3 Add 25 µL of test serum/plasma to all the microtubes.

3.4.4 Incubate for 15 min at 37^{o}C in the CAT Incubator.

3.4.5 Centrifuge the card in the CAT centrifuge for 10 minutes.

3.4.6 Read the results on charts provided with the panel.

3.5 ANTIBODY IDENTIFICATION

3.5.1 Identify the appropriate microtube of the card with the patient's name/number. Remove the aluminum foil from as many microtubes as needed.

3.5.2 Pipette 50µl each of the ready to use Panel test cells into the appropriate microtube.

3.5.3 Add 25 µL of test serum/plasma to all the microtubes.

3.5.4 Incubate for 15 min at 37^{o}C in the CAT Incubator.

3.5.5 Centrifuge the card in the CAT centrifuge for 10 minutes.

3.5.6 Read the results on charts provided with the panel

3.5.7 Note the reaction on the antigram.

3.6 Reporting results

Interpretation is done according to the reaction on the antigram charts provided by the manufacturer.

5.0 RECORD

Record the results of test in antibody screening and identification register

6.0 REFERENCE

- Manufacturer's Guidelines.
- AABB Technical Manual
- Principles & Practice of Transfusion Medicine, 1st Edition 2014

Name of Blood Bank
Name of Hospital Delhi-1100
License no -

Number **RCS/0**	**Effective Date**	**Page** **2**	**Author**	**Authorized**
Version - 0	**Review period** **2years**	**No. of copies:5**	**Approved By**	**Revision Date**
Location **Red cell Serology Lab**			**Subject** **Direct Antiglobulin Test (DAT)**	
Function **Performing DAT using Card Screening** **(IgG, IgA, IgM, C3c, C3d, Ctl)**			**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Designated Lab.** • **Master Copy**	

1.0 PURPOSE

To identify the coating of red cells with antibodies

2.0 SCOPE

The direct antiglobulin test is generally used to determine if red cell have been coated, in vivo, with immunoglobulin, complements, or both. Washed red cells from a patient or donor are tested directly with AHG reagent. Test is used in investigating autoimmune hemolytic, drug induced hemolysis, hemolytic disease of newborn & allo immune reaction to recently transfused red cells.

3.0. Responsible Person

Technologist, final reporting by Blood Bank Officer

4.0 ACTIVITIY

4.1 Principle

The direct antiglobulin test is based on the principle that antiglobulin antibodies induce in vitro agglutination of erythrocytes with immunological bound antibodies. After erythrocytes are washed to remove free plasma protein from the test mixture, they are tested directly with reagents containing anti **–IgG, IgA, IgM, C3c, C3d, Ctl**. (broad spectrum /polyspecific). Monospecific Antisera, for example anti lgG, anti –C3d, anti –C4 that are specific for immunoglobulin, complements, may also be used. The DAT procedure is clinically important in the diagnosis of conditions such as hemolytic anemia or hemolytic transfusion reactions.

4.2 Material

- CAT Card with six microtubes containing poly specific AHG (Anti-**IgG, IgA, IgM, C3c, C3d, Ctl**) and negative control.
- LISS for red cell suspension
- CAT centrifuge and incubator.
- Micropipettes (10, 25, 50, & 1000 µL)

4.3 Preparation of cell suspension(0.8-1% RBC):

Add 10 µL of packed RBC Cells (washed once in saline) in 1ml of LISS solution in a clean test tube and mix uniformly

4.4 Specimen

4.4.1 No special preparation of the patient is required before specimen collection. The patient must be positively identified when the specimen is collected.

4.4.2 The specimen must be labeled at the bedside and must include the patient's first last name, the date the specimen is collected and the patient's hospital identification number. The time of collection and the doctor's initials should be written on the required form.

4.4.3 Blood should be drawn by an aseptic technique and the specimen tested as soon as possible. The specimen should consist of 3-5 ml of EDTA, citrate, CPDA, or heparin blood. Samples should be well centrifuged.

4.4.4 If a delay in testing occurs, the specimen must be refrigerated at 2-8ºC after separation, for a maximum of 48 hrs, thereafter at - 20ºC. Antibodies dependent for their detection upon the binding of complement may not be detected if aged serum or plasma from an anti-coagulated sample is used. Hemolysis of the specimen is undesirable.

4.5 Procedure

4.5.1 Identify the appropriate microtube of CAT card with the patient's name/number.

4.5.2 Remove the aluminum foil from as many microtubes as needed.

4.5.3 Add 50µL of the patient's RBC suspension (0.8-1.0%).

4.5.4 Centrifuge the CAT card in the CAT centrifuge for 10 minutes.

4.5.5 Read the results.

4.5.6 Reporting results

The settling of all RBCs in the bottom of column indicates a negative DCT. If there is visible trapping of RBCs in between the column that indicates positive DCT.

5.0 RECORD

Record the results of the test in patient's grouping register

6.0 REFERENCE

- Manufacturer's Guidelines.
- AABB Technical Manual
- Principles & Practice of Transfusion Medicine, 1st Edition 2014

Name of Blood Bank
Name of Hospital Delhi-1100
License no - 0

Number RCS/013	Effective Date	Page 2	Author	Authorized
Version - 0	Review period 2years	No. of copies:5	Approved By	Revised Date

Location Red cell Serology Lab	Subject Rh Phenotyping
Function Performing Rh phenotype using CAT card, "Rh-subgroups+K"	Distribution • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy

1.0 PURPOSE

To identify the different phenotypes of Rh system and presence of Kell antigen

2.0 SCOPE

This test is done for identification of unexpected antibodies, for obtaining compatible blood for a patient with an anybody, and in an D negative donors to prevent immunization due to other Rh antigens in D negative patients

3.0. RESPONSIBLE PERSON

Technologist, final reporting by Blood Bank Officer

4.0 ACTIVITIY

4.1 **Material**

4.1 CAT Card" Rh-subgroups +K" containing monoclonal antibodies anti-C, anti- c, anti-E, The microtube control is the negative control.

4.2 LISS for red cell suspension
4.3 CAT centrifuge and incubator.
4.4 Micropipettes (10, 25, 50, & 1000 µL)

4.2 **Preparation of cell suspension (5% RBC):**

Add 25 µL of packed RBC Cells (washed once in Saline) in 0.5mlof ID-Diluent 2 LISS solution in a clean test tube and mix uniformly

4.3 **Specimen**

No special preparation of the patient is required before specimen collection. The patient must be positively identified when the specimen is collected.

4.3.1 The specimen must be labeled at the bedside and must include the patient's first last name, the date the specimen is collected and the patient's hospital identification number. The time of collection and the doctor's initials should be written on the required form.

4.3.2 Blood should be drawn by an aseptic technique and the specimen tested as soon as possible. The specimen should consist of 3-5 ml of EDTA, citrate, CPDA, or heparin blood. Samples should be well centrifuged

4.3.3 If a delay in testing occurs, the specimen must be refrigerated at 2-8ºC after separation, for a maximum of 48 hrs, thereafter at - 20ºC. Antibodies dependent for their detection upon the binding of complement may not be detected if aged serum or plasma from an anti-coagulated sample is used. Hemolysis of the specimen is undesirable.

4.4 **Procedure**

4.4.1 Identify the appropriate microtube of an ID card with the patient's name/number.

4.4.2 Remove the aluminum foil from as many microtubes as needed.

4.4.3 Add 10-12.5µL of the patient's RBC suspension to all the microtubes of the ID-card

4.4.4 Centrifuge the ID card in the ID centrifuge for 10 minutes.

4.4.5 Read the results.

4.5 **Reporting results**

The settling of all RBCs in the bottom of column indicates a negative test. If there is visible trapping of RBCs in between the column that indicates a positive test.

5.0 RECORD

Record the results of the test

6.0 REFERENCE

- Manufacturer's Guidelines.
- AABB Technical Manual
- Principles & Practice of Transfusion Medicine, 1st Edition 2014

Name of Blood Bank

Name of Hospital Delhi-1100

License no - 0

Number **RCS/0**	**Effective Date**	**Page** **2**	**Author**	**Authorized**
Version - 0	**Review period** **2years**	**No. of copies:5**	**Approved By**	**Revision Date**

Location **Red cell Serology Lab**	**Subject** **Blood Group Determination**
Function **Blood grouping by Microplate method**	**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Designated Lab.** • **Master Copy**

1.0 PURPOSE

Determination of the ABO/Rh blood groups

2.0 SCOPE

To determine the correct ABO group of an individual and ensure the reliability of the result.

3.0-RESPONSIBILITY

3.1 The technologist posted in Immuno-hematology lab

3.2 The Blood Bank Officer should validate and verify all discrepant or doubtful results.

4.0 ACTIVITY

4.1 Materials required

4.1.1 U-shaped microplate

4.1.2 Normal saline

4.1.3 Pipette(50µL)

4.1.4 Yellow universal Tips

4.1.5 Plate Reader

4.1.6 Plate-Shaker

4.1.7 Plate-Centrifuge (190 RCF)

4.1.8 Anti-sera A,B,AB

4.1.9 Reagent red cells A, B, O

4.2 Preparation of blood sample

Preparation of 3-5% red cell Suspension(as per SOP -)

For optimal results, freshly drawn blood samples must be used, preferably using citrate, EDTA or CPDA anticoagulants. Samples drawn in plain tubes may also be used. Where samples are not for immediate testing, store at 2-6° C for a maximum of 48 hours

4.3 Procedure

4.3.1 Identify 6 wells of the microplate for each patient Put 1 drop of patient's red cell suspension into 3 wells of the microplate

4.3.2 Put 1 drop of patient's serum into the remaining 3 wells of the microplate

4.3.3 Add 1 drop of Antisera each(anti A ,anti B, anti AB) to the red cell suspension and 1 drop of reagent red cells(A,B,O) each into the serum

4.3.4 Manually tap the plate or shake the plate by shaker

4.3.5 Shake for 10-30 secs at about 500 rpm

4.3.6 Centrifuge the plate in Plate-Centrifuge for 1 min at 190 RCF

4.3.7 Re-suspend the cells with high speed shaker setting (>1000rpm)

4.3.8 Lower the speed of the shaker to half(500 rpm)

4.3.9 Read and record the results

4.4 Interpretation

Positive: agglutination, Negative: no agglutination

4.5 Reactions for blood groups ABO

Anti-A	Anti-B	Blood group
+++to++++	**negative**	**A**
negative	**+++to++++**	**B**
+++to++++	**+++to++++**	**AB**
negative	**negative**	**O**

Reactions for reverse grouping

A1	B	Blood group
+to++++	negative	B
negative	+to++++	A
+to++++	+to++++	O
negative	negative	AB

5.0 RECORD

Record the results in the ABO Blood Grouping Register

6.0 REFERENCE

- AABB Technical Manual
- Principles & Practice of Transfusion Medicine, 1st Edition 2014

Precautions to be taken while using CAT

- Centrifuge unused cards for 1 cycle in the ID centrifuge before use
- Avoid touching the tip of the pipette with the serum/test cells in the microtube to prevent any carryover of sample
- Whenever the test cells are required, preferably use only DiaMed cells
- Always pipette cells before serum/plasma
- Cells should always be pipette into the well at an angle of 45º
- Serum/plasma should always be added straight from the top
- When serum/plasma samples are used, the serum/plasma should be well cleared by centrifugation for 10 minutes at 1500g, before use (to avoid fibrin residues interfering with the reaction pattern)
- Storage temperature for cards is 18ºC-25ºC (at room temperature) in dark
- Cards should be placed in card racks in upright position only
- All reagents taken from 2-8ºC (fridge) should be used after 15-20 minutes (so that temperature of the reagents reach RT)
- Results can be stored for 2 days at RT and one week at 2-8ºC, provided card is properly sealed with Para films.

STANDARD OPERATING PROCEDURES
CROSS-MATCH LABORATORY
INDEX

Name of Blood Bank
Name of Hospital Delhi-1100
License no -

Number CML/00	Effective Date	Page 4	Author	Authorized
Version - 0	Review period 2years	No. of copies:5	Approved By	Revision Date

Location Cross match- Lab	Subject COMPATIBILITY TESTING (CROSSMATCHING)
Function: Coomb's X –Match by conventional tube technique.	Distribution • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy

1.0 PURPOSE

To check the presence of clinically significant antibody in the patient's serum which can react / or have a capability of destruction of donor's cells.

2.0 SCOPE

This procedure is applied for compatibility testing of all patients requiring transfusion .It has ability to pick up the ABO grouping discrepancy between patient and donor as well as any incompatibility due to incomplete antibodies in patient's serum which can react with donor's red cells.

3.0 RESPONSIBILITY

echnologist posted in Compatibility lab

4.0 ACTIVITIY

4.1 Principle

Pre-transfusion compatibility testing combines a potential recipient's blood specimen with an intended donor. The major cross match consists of combining a sample of the recipient's

serum with a sample of red cells from the intended donor. If an antibody is present in the potential recipient that has specificity for an antigen on the donor red cells, agglutination or hemolysis should be exhibited.

4.2 Specimen

4.2.1 No special preparation of the patient is required before specimen collection.
4.2.2 The patient must be positively identified when the specimen is collected.
4.2.3 The patient's specimen must be labeled at the bedside and the label must include the patient's first and last name, the date when the specimen is collected and the patient's hospital identification number.
4.2.4 The time of collection and the Doctor's initials should be written on the requisition form.
4.2.5 Blood should be drawn by an aseptic technique and the specimen should be collected in a plain and EDTA tube.
4.2.6 Hemolysis renders the specimen unsuitable for testing.
4.2.7 If a delay in testing is necessary, the blood should be refrigerated at 2-4ºC.
4.2.8 In the massively transfused (10 units or more) patients, the general rule of thumb is that a new specimen must be drawn after transfusion of 3 or more units.
4.2.9 Do not accept the requisition form if any one of the above is incomplete.

4.3 Material

4.3.1 AHG reagent (broad – spectrum or IgG)
4.3.2 LISS
4.3.3 12x75 mm disposable test tubes
4.3.4 Pipettes, micro or Pasteur
4.3.5 Scissors
4.3.6 Test tubes rack.
4.3.7 37^{o}C water bath
4.3.8 Microscope,
4.3.9 Timer
4.3.10 Centrifuge with timer

4.4 QUALITY CONTROL

Reagent red cells should be tested daily with known Antisera. All negative AHG reaction must be tested with IgG sensitized red cells and should produce a positive reaction/ agglutination. If the AHG control cells do not agglutinate, the compatibility test is invalid.

4.4 Procedure

4.5.1 Preliminary check as per 4.0.4 above
4.5.2 Compare the information on the patient's specimen with the I nformation on the Blood requisition form: Name, identification number, and date: if this information is not identical, obtain a new specimen.
4.5.3 Centrifuge the clotted specimen for 1-2 minutes at 1500-2000 rpm.
4.5.4 Check past blood bank records to determine if the patient has ever been tested for antibodies or previously transfused

4.5.5 Perform an ABO grouping (forward and reverse), and Rh typing, on the donor and on the recipient's specimen. Record all test result in the blood bank records as well as on the cross match requisition form.

4.5.6 Check date on previous records, if the ABO and Rh are not same; obtain a new specimen for repeat testing.

4.5.7 Take the whole blood/ PRBC to be cross-matched from the blood bank refrigerator in which screened blood units "safe for transfusion "are stored. Check all identification on the units, for example: - Donor No. , check characteristics of the unit, for example – expiration date, and the physical characteristics of the unit, viz. –presence of hemolysis/ discoloration / clots.

4.5.8 Detach a segment from the unit of blood. Cut the ends of the segment and drain it into a test tube

4.5.9 Wash the donor specimen 3 times with normal saline. Decant the final wash completely and prepare a 3 -5% suspension of the red cells.

4.5.10 Enter the donor number and expiration date on the patient requisition form. ABO grouping and Rh typing of the donor unit should be performed.

4.6 **Compatibility procedure**

The compatibility test can be divided into three phases: room temperature, (immediate spin), at 37^{o} C, and AHG phase. Only the immediate spin phase is performed if a patient demonstrates a negative antibody screen in the AHG phase

using pooled O screening cells. The following is an example of a typical cross match configuration including all phases.

4.7 **Room temperature (immediate spin) phase**

4.7.1 Label two 12x75 mm test tubes. One tube should have the patient's last name and donor number; it should be labeled AUTO (control)

4.7.2 To tube no. 1 add 2 drops of patients serum & 1 drop of donor's washed cells

4.7.3 To the AUTO tube add 2 drops of patient's serum & 1 drop of patient's washed cells.

4.7.4 Mix and centrifuge for 1 minute at 1000rpm.

4.7.5 Gently re suspend the cell button and read microscopically. If agglutination is observed, do not proceed with the next phase. Agglutination at this phase is considered to indicate an incompatible cross match. Additional testing may not strengthen the degree of agglutination.

4.7.6 Record result. If no agglutination, proceed with the next phase.

4.7.8 37^{o}C PHASE, Reaction enhancing reagent such as LISS/ enzyme/ albumin is added at this phase.

4.7.9 Incubate the 2 tubes from the immediate spin phase for 30-45 minutes at 37^{o}C and for 15 minutes if LISS is used.

4.7.10 Centrifuge for 1minute @ 1000rpm.

4.7.11 Gently re suspend the cell button and examine microscopically.

4.7.12 Record results.

4.8 ANTIGLOBULIN PHASE (AHG)

4.8.1 Wash the 2 test tubes from the 37°C phase three times with normal saline.

4.8.2 After the last wash decant all saline and add 2 drops of AHG reagent.

4.8.3 Mix and centrifuge for 1 minute at 1000 rpm.

4.8.4 Gently re suspend the cell button and examine microscopically.

4.8.5 Record results.

4.8.6 If either or both of the tubes demonstrate no agglutination, add one drop of IgG sensitized red cells (Coombs control or check cell) to the tube.

4.8.7 Centrifuge the tubes for 1 minute at 1000 rpm and observe for agglutination. Agglutination must occur. The test is invalid if the control cell does not agglutinate, and test must be repeated.

4.8.9 Record results.

4.9 REPORTING RESULTS

A compatible cross match is indicated by the absence of agglutination and / or hemolysis at any stage of the cross match. The absence of agglutination indicates that the patient has no demonstrable antibodies with specificity for any antigens on the donor red cells.

4.10 PROCEDURE NOTES

4.10.1 If incompatibility is demonstrated by agglutination or hemolysis at any stage of the cross match, the donor unit should not be used for transfusion. Exception might include “least incompatible”units in patients with auto antibodies.

4.10.2 If the patient has a positive antibody screen or hemolysis at any phase with a donor unit, the antibody specificity should be determined. Units that are negative for the antigen to the patient’s antibody can be cross – matched.

4.10.3 The cross match will detect the ABO incompatibility.

4.10.4 The cross match will also detect most unexpected antibodies directed against common antigens. If patient exhibit strong rouleaux formation, a saline washed red cells technique may be used.

4.11 SALINE WASHED RED CELLS TECHNIQUE

4.11.1 Re centrifuge the serum – cell mixture when Rouleaux formation is suspected.

4.11.2 Remove the serum

4.11.3 Replace the serum with an equal volume of saline (2 drops) and mix.

4.11.4 Centrifuge the saline –cell mixture and examine for agglutination.

4.11.5 Rouleaux will disappear. True agglutination will remain.

4.12 LIMITATIONS

4.12.1 Low – titre antibodies: The patient antibodies may not be detectable in the pre transfusion compatibility test. These antibodies may be demonstrable with enhancement methods such as enzyme test.

4.12.2 A cross match will not prevent allo-immunization of the patient , guarantee normal survival of transfused red cells and detect all unexpected antibodies in a patient's serum.

5.0 RECORD

5.1 Enter results in cross – match register & compatibility report form.

5.2 Technologist who performed the test and who has checked the results should put his/her initials in all records.

6.0REFERENCE

- Technical Manual AABB 13th Edition.
- Transfusion Medicine Technical Manual, DGHS, 2nd Edition , 2003

Name of the Blood Bank

Name of the Hospital , Delhi-1100

License no

Number **CML/00**	**Effective Date**	**Page** **2**	**Author**	**Authorized**
Version – 0	**Review period** **2years**	**No. of copies:5**	**Approved By**	**Revision Date**
Location **Cross match Lab**			**Subject** **Compatibility testing (crosshatching)**	
Function **Coomb's X – Match by Column Agglutination Technique (CAT)**			**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Designated Lab.** • **Master Copy**	

1.0 PURPOSE

To check the presence of clinically significant antibody in the patient's serum which can react / or have a capability of destruction of donor's cells.

2.0 SCOPE

This procedure is applied for compatibility testing of all patients requiring transfusion .It has ability to pick up the ABO grouping discrepancy between patient and donor as well as any incompatibility due to incomplete antibodies in patient's serum which can react with donor's red cells.

3.0 RESPONSIBILITY

Technologist posted in Compatibility lab

4.0 ACTIVITIY

4.1 Principle

Pre-transfusion compatibility testing combines a potential recipient's blood specimen with an intended donor. The major cross match consists of combining a sample of the recipient's serum with a sample of red cells from the intended donor. If an antibody is present in the potential recipient that has specificity for an antigen on the donor red cells, agglutination or hemolysis should be exhibited.

4.2 Specimen

4.2.1 No special preparation of the patient is required before specimen collection.

4.2.2 The patient must be positively identified when the specimen is collected.

4.2.3 The patient's specimen must be labeled at the bedside and the label must include the patient's first and last name, the date when the specimen is collected and the patient's hospital identification number.

4.2.4 The time of collection and the Doctor's initials should be written on the requisition form.

4.2.5 Blood should be drawn by an aseptic technique and the specimen should be collected in a plain and EDTA tube.

4.2.6 Hemolysis renders the specimen unsuitable for testing.

4.2.7 If a delay in testing is necessary, the blood should be refrigerated at 2-4ºC.

4.2.8 In the massively transfused (10 units or more) patients, the general rule of thumb is that a new specimen must be drawn after transfusion of 3 or more units.

4.2.9 Do not accept the requisition form if any one of the above is incomplete.

4.3 Material

4.3.1 Coombs CAT Cards with six microtubes containing poly specific AHG (Anti-IgG+C3d)

4.3.2 LISS for red cell suspension

4.3.3 CAT centrifuge and incubator.
4.3.4 Micropipettes (10, 25, 50, & 1000 µL

4.4 Preparation of cell suspension (0.8-1% RBC):

4.4.1 Add 10 µL of packed RBC Cells (washed once in Saline) in 1ml of LISS in a clean test tube and mix uniformly.

4.5 Procedure: **MAJOR CROSS MATCH**

4.5.1 Identify the appropriate microtubes of the CAT Card with the recipient's and donor's name/number.
4.5.2 Take off the aluminum foil from as many microtubes as needed.
4.5.3 Add 50 µL of donor's cell suspension (0.8-1.0%) to the appropriate microtubes.
4.5.4 In case an auto-control is to be included, add 50µl of the patient's own red cell suspension to the appropriate microtube.
4.5.5 Add 25µL of the recipient's plasma/serum to all the microtubes.
4.5.6 Incubate at 37ºC for 15 min in the CAT incubator
4.5.7 Centrifuge the card(s) for 10 minutes in the CAT centrifuge
4.5.8 Read the results.

Note: For saline/room temperature cross-match, the same procedure should be carried out using NaCl /Enzyme cards, which, should be incubated at room temperature after addition of cells & plasma/serum

4.6 Procedure: MINOR CROSSMATCH

4.6.1 Identify the appropriate microtubes of the CAT Card with the recipient's and donor's name/number.
4.6.2 Take off the aluminum foil from as many microtubes as needed.
4.6.3 Add 50 µL of patient's cell suspension (0.8-1.0%) to the appropriate microtubes.
4.6.4 Add 25µL of the donor's plasma/serum to all the microtubes.
4.6.5 Incubate at 37ºC for 15 min in the CAT incubator
4.6.6 Centrifuge the card(s) for 10 minutes in the CAT centrifuge
4.6.7 Read the results.

Note: For saline/room temperature cross-match, the same procedure should be carried out using NaCl/Enzyme cards, which, should be incubated at room temperature after addition of cells & plasma/serum

4.7 REPORTING RESULTS

A compatible cross match is indicated by the settling of all RBC s in the bottom of column, which indicates that the patient has no demonstrable antibodies with specificity for any antigen on the donor's red cell. If there is any visibly seen trapping of RBCs in between the column that indicates the incompatibility between patients & donor & that blood must not be transfused to the patient.

5.0 RECORD

Enter result in cross – match register & compatibility report form

Technologist who performed the test and who has checked the result should put his/her initials in the records

6.0 REFERENCE

- Technical Manual AABB 13th Edition.
- Transfusion Medicine Technical Manual, DGHS, 2nd Edition , 2003
- Manufacturer's insert

Name of Blood Bank

Name of Hospital Delhi-1100

License no -

Number CML/00	**Effective Date**	**Page** 2	**Author**	**Authorized**
Version - 0	**Review period** 2years	**No. of copies: 5**	**Approved By**	**Revision Date**
Location Issue Counter		**Subject** Issue of Blood / Blood Components		
Function Receiving patient's sample with requisition form & issuing of blood products		**Distribution** • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy		

1.0 PURPOSE

The blood and components are used as per need of the patients. These are issued as per the requisition of a clinician after ensuring the compatibility testing in Blood Bank.

2.0 SCOPE

To issue proper component as per need of the patient, minimize wastage of time as well as Blood products

3.0 RESPONSIBILTY

It is the responsibility of the technologist on duty in compatibility lab to issue the blood for which requisition is received on the directions of Blood Bank Officer.

4.0 ACTIVITY

4.1 Material

4.1.1 Requisition form

4.1.2 Compatibility report with reaction form

4.1.3 Issue/ release form

4.1.4 Master register

4.1.4 Issue register
4.1.5 Stock register

4.2 Procedure

4.2.1 Receive the requisition form after verifying the patient's name & CR Number, ward, name of Hospital on form as well as on blood sample. Also verify the name and designation of the doctor signing on the requisition form.
4.2.2 Enter the particulars of the patient on the register and mention the date and time of receiving the sample.
4.2.3 Assign the proper unit numbers to the patient from stock register. In order to avoid outdating, implement FIFO (first in first out) policy.
4.2.4 Ensure that the assigned units have been tested for TTI and found suitable for use.
4.2.5 Carry out compatibility testing.
4.2.6 If unit is not to be issued, immediately keep back into the BBR of cross match lab.
4.2.7 When the issue/ release slip comes from ward take out the correct unit from blood bank refrigerator and keep it in thermal box for transport. Also hand over the compatibility report & reaction form.
4.2.8 Make entries in the issue register.
4.2.9 Instruct the individual to take the unit straight to OT/ ward for transfusion and hand over the unit to nursing staff/ transfusionist .

5.0 RECORD

5.1 Make following entries in issue register:
5.2 Name of patient
5.3 Hospital registration number
5.4 Blood group of patient
5.5 Date and time of issue
5.6 Unit no. Issued
5.7 Blood group of unit
5.8 Component of blood
5.9 Compatibility report
5.10 Signature of technician who issues
5.11 Make an entry on master register

6.0 REFERENCE

- Technical Manual AABB 13th Edition.
- Transfusion Medicine Technical Manual, DGHS, 2nd Edition , 2003

Standard Operating Procedures
Transfusion Transmitted Infections
Index

Name of Blood Bank
Name of Hospital Delhi- 1100
License no -

Number TTI/00	**Effective Date**	**Page** 3	**Author**	**Authorized**
Version - 0	**Review period** 2years	**No. of copies:5**	**Approved By**	**Revised Date**
Location TTI Testing laboratory			**Subject** HBsAg Testing	
Function Sample tested for Hepatitis B Surface antigen by ELISA Gen 4			**Distribution** • HOD • Quality Manager • Technical Manager • Designated lab • Master Copy	

1.0 PURPOSE

Detection of Hepatitis B surface antigen in donor's blood for the prevention of transfusion related transmission of Hepatitis B.

2.0 SCOPE

HbsAg is a mandatory test for blood unit screening before it is transfused. This is carried out on all donor unit samples.

3.0 RESPONSIBILITY

Centrifuge the clotted donor's sample and transfer the serum into fresh pre labeled numeric coded test tubes. If there is delay in testing, store the sample at 2-8ºc.

Reagent should be stored properly during testing. Store the reagent according to manufacturer's guidelines and should be brought to room temperature prior to start of the procedure.

Check all packaging before using the kits, if the packaging is damaged, the Technologist must check that the component of kit are intact before using them.

Responsible Person

It is the responsibility of Technologist of TTI lab to ensure correct sample is received from donor complex and carry out the tests.

3.0 ACTIVITY

4.1 Materials required

4.1.1 Serum sample of the blood donors.

4.1.2 Monoclonal Anti HBsAg coated microplate.

4.1.3 Conjugate & conjugate diluent

4.1.4 HBsAg positive control

4.1.5 HbsAg negative control
4.1.6 Substrate Buffer
4.1.7 Chromogen Pink Coloured solution
4.1.8 Stopping solution
4.1.9 Concentrated Washing solution.
4.1.10 D/W
4.1.11 Sodium hypochlorite &sodium bicarbonate
4.1.12 Micro-Pipettes to measure & dispense 50,100,1000 microL
4.1.13 Graduated cylinders of 100ml,1000ml capacity
4.1.14 Container for bio-hazardous waste
4.1.15 Water-bath/microplate incubator
4.1.16 Microplate washer
4.1.17 Microplate reader equipped with 450,490 &620-700nm filters
4.1.18 Absorbent paper
4.1.19 Filter paper sheet
4.1.20 Work-sheet etc

4.2 Principle

4.2.1 The method is a one step enzyme immunoassay technique of the sandwich type using monoclonal antibodies & polyclonal antibodies for the detection of the various subtypes of surface antigen of the hepatitis B virus (HBsAg) recognized by WHO, in human serum and plasma. The solid phase is coated with monoclonal antibodies and the conjugates are monoclonal mouse antibodies and polyclonal goat antibodies against the HBsAg, bound to the peroxidase.

4.2.2 Control sera and samples are distributed into the wells of the microplate
4.2.3 Red colored conjugate is then added
4.2.4 After incubation at 37ºC during one hour and half the unbound conjugate is removed by washing
4.2.5 Distribution of coloured substrate solution
4.2.6 After 30 minutes incubation in presence of the substrate in dark & at room temperature (18-30ºC),the presence of the complexed conjugate is shown by change of colour
4.2.7 Distribution of stopping solution
4.2.8 Reading of the optical densities at 450/620-700nm and interpretation of the results.

4.3 Procedure

4.3.1 Bring all the reagents and test specimen to room temperature before use.
4.3.2 Prepare the diluted washing solution by adding 50ml of wash buffer to 950ml of D/W
4.3.3 Prepare the conjugate working solution by adding conjugate diluents vial into the lyophilized conjugate 10 minutes before starting the test

4.3.4 Take out from the protective packing the support frame and the necessary number of strips. Put the unused strips back in their packing and reclose it

4.3.5 Add 100 micro liter of sample (serum) directly to each well. In each run, will be four negative controls and one positive control.

4.3.6 Add 50µL of conjugate into all the wells. Cover the plate with seal for 90 minutes at 37 ºC.

4.3.7 Wash the plate with micro plate washing procedure, 6 times with at least 0.35 ml wash buffer per well.

4.3.8 Add 100µL substrate. Cover the plate with black cover and incubate for 30 minutes in dark at 20-30ºC

4.3.9 Add 100µL of stopping buffer to each well.

4.3.10 Carefully wipe the plate bottom. Wait for 4mins after addition of stopping solution before reading and within 30 minutes of stopping the reaction.

4.3.11 Read absorbency at 450/620-700nm using a plate reader

ERBA LISA PICO HBsAg

- Add 25 µl of sample diluents each well.
- Add 75 µl of sample diluents to A_1.
- Add 75 µl 3NC and 1PC controls (B_1, C_1, D_1 and E_1).
- Add 75 µl sample F_1 onwards.
- Add 50 µl of conjugate to each well.
- Incubate for 60 minutes at 37°C.
- Wash the plate 5 times, blot, and dry (washing solution – 1:20 dilution).
- Add 50 µl of color reagent to each wells, cover the plate with black cover for 15 minutes in dark at room temperature.
- Add 100 µl of stop solution, read the absorbance at 450 nm with reference filter 620 nm.

4.4 Validation for BIORAD

4.4.1 Calculation for cut – off value determination.

4.4.2 Positive control absorbency of individual positive control should be equal or greater than 1.0

4.4.3 Negative control. Absorbency of any of individual negative control should be equal or less than 0.080

4.4.4 Next: - Average value of negative control. Cut –off value formula = mean of NC +0.050.

4.5 Validations for ERBALISA

BC< 0.05

NC<0.02

PC>1.00

CUT OFF :-NCX + 0.05

4.6 Interpretation

4.6.1 Non – reactive if the absorbency of the test sample is less than the cut –off value, and then sample is considered as non-reactive.

4.6.2 If the Absorbency of the test serum is equal or greater them the cut off value, then it is considered as initially reactive and the donor unit is discarded. This sample should be repeated. If the absorbency of sample is again equal to or more than cut off value then it is considered as repeated reactive and the results are noted.

5.0 Record – same

Paste the printout in the HBs Ag Register and also record the following details:

5.0.1 The date on which the test is run

5.0.2 Lot No. and expiry date of the kit

5.0.3 Initials of the Technologist who performed the test.

5.0.4 The test should be validated by the Blood Bank Officer

5.0.5 Reactive unit are marked in red and segregated for disposal as per BMW rules.

5.0.6 The number of samples to be repeated

5.0.7 Transfer the result to Infectious marker register, Screening Register, Master Register.

6.0 REFERENCE

- Kit Package Insert
- Transfusion Medicine Technical Manual, 2nd Edition, 2003

NOTE: In case Blood Bank is using kit of other manufacturer the SOP procedure , validation and Interpretation is to be modified as per the literature in the kit insert

Name of Blood Bank
Name of Hospital, Delhi-1100
License no -

Number **TTI/00**	**Effective Date**	**Page** **3**	**Author**	**Authorized**
Version - 04	**Review period** **2years**	**No. of copies:5**	**Approved By**	**Revised Date**
Location **TTI Testing Laboratory**			**Subject** **HCV Testing**	
Function **Sample tested for HCV antigen& antibodies by** **ELISA Gen 4**			**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Designated lab** • **Master Copy**	

1.0 PURPOSE

Detection of Hepatitis C antigen & antibody in donor's blood for the prevention of transfusion related transmission of Hepatitis C.

2.0 SCOPE

HCV is a mandatory test for blood unit screening before it is transfused. This is carried out on donor unit's samples.

3.0 RESPONSIBILITY

Centrifuge the clotted donor's sample and transfer the serum into fresh pre labeled numeric coded test tubes. If there is delay in testing, store the sample at 2-8ºc.

Reagent should be stored properly during testing. Store the reagent according to manufacturer's guidelines and should be brought to room temperature prior to start of the procedure.

Check all packaging before using the kits, if the packaging is damaged, the Technologist must check that the component of kit are intact before using them.

Responsible Person

It is the responsibility of Technologist of TTI lab to ensure correct sample is received from donor complex and carry out the tests.

4.0 ACTIVITY

4.1 Materials required

4.1.1 Serum sample

4.1.2 Monolisa HCV Ag-Ab Ultra kit containing the following

4.1.3 Microplate, concentrated washing solution, negative control, positive control, antigen positive control, antigen diluent, conjugate 1, conjugate 2, peroxidase substrate buffer, chromogen& stop solution.

4.1.4 D/W

4.1.5 Sodium hypochlorite and sodium bicarbonate

4.1.6 Disposable gloves, protective glasses, disposable tubes

4.1.7 Micro-Pipettes capable of delivering 50µL,80µL,100µL,200µL&1ml

4.1.8 Graduated cylinders of 10,200&1000ml capacity

4.2 Principle

4.2.1 The method is a qualitative enzyme immunoassay for the detection of HCV infection, based on the detection of capsid antigen and antibodies associated with an infection by Hepatitis C virus in patient serum /plasma. The test includes the following steps

4.2.2 In the first stage, the HCV antibody present in the test sample binds with the HCV recombinant Ag coated on the wells and if Hepatitis C capsid antigen is present this antigen will be bound by the monoclonal antibodies coated on the solid phase in the microplate and by the biotinylated monoclonal antibodies against the capsid hepatitis C antigen(conjugate 1)

4.2.3 After incubation at 37ºC during 90 minutes and a washing step, the conjugate 2 containing peroxidase-labeled anti-human IgG antibodies and streptavidine-peroxidase are added to each well of microplate. If Human IgG is present, having reacted with solid phase, the Anti-human IgG conjugate binds to the Human antibodies. The conjugate Streptavidine/peroxidase binds to the biotin of conjugate 1 if HCV capsid antigen if present.

4.2.4 After incubation for 30 mins at 37ºC,the unbound enzymatic conjugate is removed by washing step and the antigen-antibody complex is revealed by addition of substrate

4.2.5 30 minutes after the reaction has been stopped, the absorbance values are read using a spectrophotometer at 450/620-700nm.

4.3 Procedure

4.3.1 Prepare diluted washing solution R2 by adding 950ml D/w in 50 ml of wash buffer (for 1L)

4.3.2 Prepare working antigen positive control as per kit instructions

4.3.3 Remove the microplate frame and strips from the protective bag

4.3.4 Put 100µL conjugate 1 (R6) into each well.

4.3.5 Put 50µL each of negative control serum(R3) in well A1, antibodies positive control serum in wells B1, C1, D1 and 50 µL of working antigen positive control solution in E1

4.3.6 Put 50µL of first test sample into the well F1 & and further on for rest of the samples.

4.3.7 Homogenize the mixture with atleast 3 aspirations or with the microplate shaker for 5 sces.

4.3.8 Cover the wells & Incubate at 37± 1 ºC for 90 minutes.

4.3.9 After aspirating the contents of the well into a liquid waste container, add a minimum of 370µl of washing solution into each well, aspirate again..The residual volume must be <10 µl

4.3.10 Repeat the washing step as mentioned above for 4 more times

4.3.11 Dispense quickly 100µl of conjugate 2 solution into each well. (Shake gently before use)

4.3.12 Cover the plate with adhesive film and incubate at 37± 1 ºC for 30 minutes.

4.3.13 Remove the adhesive film, empty all the wells by aspiration and wash minimum 5 times.

4.3.14 Prepare the enzymatic development solution and dispense 80µL into each well. Allow the reaction to develop in the dark at room temperature (20-28ºC) for half an hour.(Do not use adhesive film during this incubation).

4.3.15 Add 100µL stop solution and homogenize.

4.3.16 Carefully wipe the plate bottom and read the OD at 450/620-700nm at least 4 minutes after stopping solution addition and within 30 minutes of stopping the reaction.

4.3.17 The result shall be checked and printed.

4.4 Validation

4.4.1 For the negative control, measured absorbance value must be < 60% 0f cut off (O.D < cut off x 0.6)

4.4.2 For the antibodies positive control, the mean of the measured value must be greater than, or equal to, 0.800 and less than or equal to 2.700. If one of the antibodies positive control individual values differ by >30% from the mean value, disregard it and calculate with remaining two positive control values.

4.4.3 The absorbance of the working antigen positive control solution should be >0.500

4.4.4 The validation of the test results is to be done by the Blood Bank Officer.

4.5 Interpretation

4.5.1 the results are automatically calculated by the Cut –off value as “mean of antibodies positive control/4

4.5.2 samples with OD <cut off value-negative

4.5.3 samples with OD≥cut off value-initially reactive-donor unit discarded and retested in duplicate for the O.Ds of the test sample if falls in grey zone.

4.5.4 After retesting, the sample is considered positive if the OD is≥ cut off value , negative if less than the cut off value

4.5.5 Grey zone kept as 10%.value more than cut off –samples in grey zone are considered reactive and discarded and sample repeated. If again reactive considered as repeat reactive

4.5.6 The borderline and reactive sample shall be discarded as per the recommended protocol.

5.0 RECORD

Pasted the printout in the HCV Register and also record the following details:

5.1 The date on which the test is run

5.2 Lot No. and expiry date of the kit.
5.3 Initials of the technologist who performed the test.
5.4 Initials of the supervisor who verifies the result.
5.5 Reactive units are marked in the red.
5.6 The number of the samples to be repeated
5.7 Transfer the result to Infectious marker register, Screening Register, Master Register.

6.0 REFERENCE

- Kit Package Insert.
- Transfusion Medicine Technical Manual, 2nd Edition, 2003

NOTE: In case Blood Bank is using kit of other manufacturer the SOP procedure , validation and Interpretation is to be modified as per the literature in the kit insert

Name of Blood Bank
Name of Hospital Delhi- 1100
License no -

Number TTI/0	**Effective date**	**Page 3**	**Author**	**Authorized**
Version - 04	**Review period 2years**	**No. Of copies:5**	**Approved By**	**Revised Date**
Location TTI Testing Laboratory			**Subject Anti HIV Testing**	
Function Sample tested for HIV1&2 antibodies and p24 antigen of HIV-1 by 4th Generation Kit for HIV			**Distribution** • HOD • Quality Manager • Technical Manager • Designated lab • Master Copy	

1.0 PURPOSE

Detection of HIV antigen & antibody in donor's blood for the prevention of transfusion related transmission of HIV I & II.

2.0 SCOPE

Testing for HIV is a mandatory test for blood unit screening before it is transfused. This is carried out on all donor unit's samples.

3.0 RESPONSIBILITY

Centrifuge the clotted donor's sample and transfer the serum into fresh pre labeled numeric coded test tubes. If there is delay in testing, store the sample at 2-8ºc.

Reagent should be stored properly during testing. Store the reagent according to manufacturer's guidelines and should be brought to room temperature prior to start of the procedure.

Check all packaging before using the kits, if the packaging is damaged, the Technologist must check that the component of kit are intact before using them.

Responsible Person

It is the responsibility of Technologist of TTI lab to ensure correct sample is received from donor complex and carry out the tests.

4.0 ACTIVITY

4.1 Material required

4.1.1 Serum samples
4.1.2 HIV Antigen and Antibody coated Microplate
4.1.3 Concentrated washing solution
4.1.4 Negative control (NC)
4.1.5 HIV Ab positive control
4.1.6 HIV Ag positive control
4.1.7 Conjugate
4.1.8 Conjugate diluents
4.1.9 Substrate
4.1.10 Peroxidase substrate buffer
4.1.11 Strip sealers
4.1.12 Stopping solution
4.1.13 Black plate cover
4.1.14 D/W
4.1.15 Sodium hypochlorite &sodium bicarbonate
4.1.16 Pipettes to measure & dispense 50, 100, 1000µl
4.1.17 Graduated cylinders of 100ml, 1000ml capacity
4.1.18 Container for biohazardous waste
4.1.19 Water-bath/microplate incubator
4.1.20 Microplate washer
4.1.21 Microplate reader equipped with 450,490 & 620-700nm filters
4.1.22 Absorbent paper

4.2 Principle

4.2.1 The method is a fourth generation in vitro enzyme Immunoassay, for the qualitative detection of antibodies for HIV-1, HIV-1 group O, HIV- 2 and p24 Antigen of HIV-1 in Human serum and plasma. It is a solid phase enzyme linked immunosorbent assay based on the principle of the double antigen/antibody sandwich technique for the

detection of the IgM and IgG antibodies against HIV-1 and/or HIV-2 and p24 antigen in human serum/plasma.

4.2.2 This Immunometric assay involves a two – stage reaction.

In the first stage, the HIV l & 2 antibodies if present in the test sample binds with HIV recombinant antigen coated on the wells. Antigen p24, if present binds to the monoclonal anti-p24 antibody on the microwell surface.

The unbound serum sample is removed by washing the wells.

In the second stage, the bounded anti- HIV l & 2 antibodies and bounded p24 antigen are detected by the peroxidase conjugate..

The unbound conjugate is removed by washing the wells.

The presence of the bound conjugate is shown by a blue colour upon additional incubation with TMB substrate.

The enzyme-substrate reaction is stopped by adding stop solution and absorbances are read using a photometer.

4.3 Procedure (BENESPHERA HIV ADVANCE ELISA -AVANTOR)

4.3.1 Take the required number of strips and fix them to frame.

4.3.2 Pipette 100µL of each of negative control, antibody positive control, antigen positive control & samples into the wells.

4.3.3. Incubate the plate at 37ºC for 60 minutes after sealing the plate with sealer

4.3.4 Before the last 5 minutes of incubation, make a 1:51 dilution of conjugate with conjugate diluents (10 ml of diluents in 200µL of conjugate)

4.3.5 Aspirate the contents from all the wells and wash each one 5 times with diluted washing solution (350µL/well/time)

4.3.6 Invert the plate &tap it on the absorbent paper .pipette 100µL of diluted conjugate into each well

4.3.7 Incubate at 37ºC for 30 minutes after sealing the plate

4.3.8 Before the last 5 minutes of incubation make a 1:101 dilution of substrate with substrate buffer (10ml of buffer in 100µl of substrate)

4.3.9 Aspirate the contents from all the wells and wash each one 5 times with diluted washing solution (350µL/well/time)

4.3.10 invert the plate &tap it on the absorbent paper .pipette 100µL of diluted substrate into each well and incubate at room temperature(18-30ºC) for 30 minutes .Avoid exposure to light

4.3.11 pipette 100µL of stop solution into each well and homogenize by tapping the plate

4.3.12 read the absorbance at 450 nm/620 nm within 30 minutes of adding stop solution.

ERBA SURE HIV (GEN4)

- Add 100 µl of controls and specimen to respective wells (4NC, 1Ab PC, 1 Ag PC, (S_1, S_2…).
- Incubate at 37°C for 1 hour.
- Wash 5 times blot, dry.
- Add 100 µl of conjugate to all wells.
- Incubate for 30 minutes at 37°C.
- Wash 5 times blot, dry.
- Add 100 µl of Substrate to each wells.
- Incubate in dark at room temperature for 30 minutes.
- Add 100 µl of stop solution, read the absorbance at 450 nm with reference filter 620 nm.
- Preparation of **conjugate** – 1:51 dilution
- Preparation of **substrate** – 1:101 dilutions
- Preparation of **washing solution** – 1:20 dilution

4.4 Validation

4.4.1 Absorbance of the entire antibody positive control and antigen positive control should be greater than /equal to 1.000

4.4.2 Absorbance of negative controls should be less than /equal to 0.100

4.4.3 Negative control mean should be greater than equal to 0.000and less than or equal to 0.100

4.4.4 Cut off limit calculated as “mean of NC + 0.2”. Grey zone taken as 10%.

4.5 Interpretation

4.5.1 The result is automatically calculated by the cut off value. Values more than cut off are reactive and donor unit discarded. And test repeated, if again reactive considered as repeat reactive. The borderline and reactive sample shall be discarded as per the recommended protocol.

5.0 RECORD

Paste the printout in the HIV Register and also record the following details:

5.1 The date on which the test is run

5.2 Lot No. and expiry date of the kit

5.3 Initials of the technologist who performed the test

5.4 Initials of the Supervisor who verifies the result

5.5 Reactive units are marked in red.

5.6 The number of the units to be repeated.

5.7 Transfer the result to Infection marker register, Screening Register , Master Register & Blood Bank software.

6.0 REFERENCE

- Kit Package Insert
- Transfusion Medicine Technical Manual, 2nd Edition, 2003

NOTE: In case Blood Bank is using kit of other manufacturer the SOP procedure , validation and Interpretation is to be modified as per the literature in the kit insert

Name of Blood Bank
Name of Hospital Delhi
License no

Number **TTI/004**	**Effective Date**	**Pages** **2**	**Author**	**Authorized**
Version - 0	**Review period** **2years**	**No. of copies:5**	**Approved By**	**Revised Date**
Location **TTI Testing Laboratory**			**Subject** **Malaria Parasite Testing**	
Function **Detection of Malaria Parasite antigen by Sure Test Malaria LDH.**			**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Designated lab** • **Master Copy**	

1.0 PURPOSE

Detection of Malaria lactate dehydrogenase in donor's blood sample for the prevention of transfusion related transmission of Malaria.

2.0 SCOPE

The sample from donors is tested for transfusion – transmitted disease and testing for malaria is mandatory.

3.0 RESPONSIBILITY

3.1 Reagent should be stored properly according to manufacturer guidelines and should be brought to room temperature prior to starting the procedure.

3.2 Check all packaging before using the kits, if the packaging is damaged the Technologist must check that component of kit is okay before using them.

3.3 Before using the reagent check the expiry date of reagents.

Responsible Person

Technologist posted in TTI testing laboratory is responsible for performing the test.

4.0 ACTIVITY

4.1 Principle

The test detects the presence of **Plasmodium lactate dehydrogenase**, an enzyme produced in the Plasmodium species. The Sure Test Rapid Malaria antigen test contains a membrane strip, which is pre-coated with a monoclonal antibody as a single line across a test strip. The monoclonal antibody is specific to lactate dehydrogenase of the

Plasmodium species. The conjugate pad is dispensed with the monoclonal antibody, which is pan specific to the lactate dehydrogenase of plasmodium species.

4.2 Material

4.2.1 Fresh anti-coagulated blood collected from the donor.

4.2.2 Sure Test malarial antigen test kit containing: Test device, package insert, instruction card & Assay buffer

4.2.3 Sample pipette

4.2.4 Lancet, Alcohol swab (optional)

4.3 Procedure

4.3.1 Add 5 µL of whole blood into the sample well

4.3.2 Add two drops (60µL) of assay buffer in the buffer well.

4.3.3 Read the results in 20 minutes.

4.4 Interpretation of Results

4.4.1 Positive reaction: the pLDH present in the sample reacts with the pan anti- pLDH conjugate and moves through the test strip where the pLDH is captured by the specific pLDH antibodies, causing the appearance of the colored band.

4.4.2 Negative reaction: only the control band will be the visible.

4.5 Validation of the test

4.5.1 The test can be considered valid when the control band is clearly visible, presenting a dense line.

4.5.2 The test is invalid when the control band does not appear.

Note: In this condition repeat the test.

4.6 Precaution

The regents of different lots must not be mixed and used.

5.0 RECORD

5.1 he date on which the test is run.

5.2 Lot No. and expiry date of the Kit & its components.

5.3 Initials of the Technologist who performed the test.

5.4 The test result is to be validated by Blood Bank Officer.

5.5 Positive units are marked in red and the donor unit is discarded as per BMW rules

5.6 Transfer the result to Infectious marker register, Screening Register, Master Register.

6.0 REFERENCE

- Kit Package Insert.
- Transfusion Medicine Technical Manual, 2nd Edition, 2003

NOTE: In case Blood Bank is using kit of other manufacturer the SOP procedure , validation and Interpretation is to be modified as per the literature in the kit insert

Name of Blood Bank

Name of Hospital Delhi

License no -

Number **TTI/00**	**Effective**	**Pages** **2**	**Author**	**Authorized**
Version - 0	**Review period** **2years**	**No. of copies:5**	**Approved By**	**Revised Date**
Location **TTI Testing Laboratory**			**Subject** **Transfusion transmitted infection**	
Function **Detection of antibodies to TreponemaPallidum.**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated lab • Master Copy	

1.0 PURPOSE

Detection of Anti Treponema Pallidum Antibody in donor's blood samples for the prevention of transfusion related transmission of syphilis.

2.0 SCOPE

The samples from donors are tested for transfusion – transmitted disease and testing for Syphilis is mandatory.

3.0 RESPONSIBILITY

3.1.1 Reagent should be stored properly according to manufacturer guidelines and should be brought to room temperature prior to starting the procedure.

3.1.2 Check all packaging before using the kits, if the packaging is damaged the Technologist must check that component of kit is okay before using them.

3.1.3 Before using the reagent check the expiry date of reagents.

Responsible Person

Technologist posted in TTI testing laboratory is responsible for performing the test.

4.0 ACTIVITY

4.0 Materials required

4.1.1 Specimen –human serum/plasma

4.1.2 TPHA test kit containing test cells, control cells, diluents, positive control serum, nonreactive control serum

4.1.3 Pipettes

4.1.4 96-well micro-titre plates.

4.2 Principle

4.2.1 TPHA reagent is used to detect human serum antibody to T Pallidum by means of indirect haem agglutination (IHA) method. Preserved avian erythrocytes are reacted with antigenic components of pathogenic T. Pallidum (Nichol's strain). These test cells agglutinate in the presence of specific antibodies to T. Pallidum in the patient serum/plasma and show characteristic patterns in micro titration plates. Test results are obtained in 45- 60 minutes and the cell agglutination patterns are both easily read and long lasting. TPHA test result has specificity similar to that of the TPI test and sensitivity comparable to that of the FTA – ABS test.

4.3 Qualitative test

4.3.1 Each sample requires 3 wells of micro titration plate A to C. Put 190 µl of diluent to well A.

4.3.2 Add 10 micro liter of serum to well A (1:20 specimen dilution). Mix thoroughly.

4.3.3 Transfer 25 µl of diluted serum from well A to well B&C.

4.3.4 Ensure that the test and control cells are thoroughly re-suspended. Add 75 µl of control cells to well B and 75 µl of test cells to well C. This will give a final specimen dilution of 1:80.

4.3.5 Shake the plate gently to mix the contents thoroughly.

4.3.6 Incubate for 45- 60 minutes at room temperature (15-30 ºC).

4.3.7 Read result. Results are stable for 24 hours.

4.4 Interpretation

Result	**Test cells**	**Control cells**
Strong Positive	Full cell pattern covering bottom of the well.	No agglutination – tight button
Weak positive	Cell pattern covers approx 1/3 of well button	No agglutination – tight button
Indeterminate	Cell pattern show distinctly Open center	No agglutination – tight button
Negative	Cells settled to a compact button typically with a small clear center	No agglutination – tight button
Non specific	Positive reaction	Positive reaction

Non-specific absorption

- Add 10µl of test specimen to a well then add 190 µl of control cells. Mix well and let stand for 30 minutes.
- Centrifuge for 3 minutes at 1500 rmp
- Take 25µL of the supernatant in two wells
- Add 75 micro l of control cell in one welland 75 micro l of test cell in well 2

- Mix well and incubate at room temperature for 45-60 minutes
- Read and interpret as above
- If the test is repeatedly non-specific the sample should be tested by another method

Limitation

- Strong positive reaction may show folding at the edge of the cell matrix.
- When the test well is positive, the control well should be observed. The control well should settle to a compact button.
- Agglutination in the control cells as well as the test cells indicate the presence of anti-cell antibody and the test should be reported as invalid and repeated.
- A doubtful reaction with test cells should be reported as indeterminate. Sample is retested

TPHA test is highly specific, but false positive result have been known to occur in patients suffering from leprosy, infection mononucleosis and connective tissue disorders.

5.0 RECORD

The details of test should be entered in Infectious marker register.

6.0 REFERENCES

- Kit insert
- Transfusion Medicine Technical Manual, 2nd Edition, 2003

NOTE: In case Blood Bank is using kit of other manufacturer the SOP procedure , validation and Interpretation is to be modified as per the literature in the kit insert

Name of Blood Bank
Name of Hospital Delhi
License no -

Number **TTI/00**	**Effective**	**Pages** **2**	**Author**	**Authorized**
Version - 0	**Review period** **2years**	**No. of copies:5**	**Approved By**	**Revised Date**
Location **TTI Testing Laboratory**			**Subject** **VDRL TESTING**	
Function **Sample Tested for RPR antigen by Flocculation Method.**			**Distribution** • HOD • Quality Manager • Technical Manager • Designated lab • Master Copy	

1.0 PURPOSE

Detection of Anti Treponema Pallidum Antibody in donor's blood samples for the prevention of transfusion related transmission of syphilis.

2.0 SCOPE

The samples from donors are tested for transfusion – transmitted disease and testing for Syphilis is mandatory.

3.0 RESPONSIBILITY

3.1 Reagent should be stored properly according to manufacturer guidelines and should be brought to room temperature prior to starting the procedure.

3.2 Check all packaging before using the kits, if the packaging is damaged the Technologist must check that component of kit is okay before using them.

3.3 Before using the reagent check the expiry date of reagents.

Responsible Person

Technologist posted in TTI testing laboratory is responsible for performing the test.

4.0 ACTIVITY

4.1 MATERIALS REQUIRED

- Reagent kit
- Micropipettes and disposable pipette tips
- Timer
- VDRL shaker
- Incubator 37^0C
- Glassware
- Distilled water.

4.3 PRINCIPLE

During the testing procedure, the specimen, serum or plasma is mixed with the carbogen reagent and allowed to react for 8 minutes. If anti-Lipoidal antibodies are present in the specimen they will react with the carbogen reagent forming visible black floccules. If anti-Lipoidal antibodies are not present in the specimen there will be no flocculation.

4.4 PROCEDURE

1. Carry out the test as per manufacturer's instructions given in the package insert.
2. Remove reagents from the fridge 30 minutes prior to testing. Mix the reagents gently by inverting the vials without foaming.
3. Bring reagents and samples to room temperature before testing.
4. Arrange all donor unit test tube samples, serially in ascending order in a test tube rack. Add required number of internal kit controls and external lab controls.
5. Discard all disposable tips into hypochlorite solution.
6. Place the cards in front of the test tube rack.

RPR Card tests for Syphilis

- Bring reagent and sample to room temperature before sampling.

↓

- Pipette 50ul of test specimen, positive control and negative control on two separate reaction circle of the disposable slide.

↓

- Add 1 drop of well-mixed carbogen reagent next to the test specimen, negative and positive control by the reagent dropper.

↓

- Mix the specimen properly by mixing stick.

↓

- Keep the slide on the VDRL rotator for 8 minutes at 180 rpm.

↓

- Observe the flocculation.
- Shake the plate gently to mix the contents thoroughly.
- ncubate for 45- 60 minutes at room temperature (15-30 ºC).
- Read result. Results are stable for 24 hours.

4.5 Interpretation

- Large and medium black floccules against white background are reactive.
- Small black floccules against white background is weakly reactive.
- No flocculation is negative.

- After test is conformed discard the card in hypochlorite solution. After completing the test and cross checked by the medical officer

5 RECORD

The details of test should be entered in Infectious marker register.

6 REFERENCES

- Kit insert
- Transfusion Medicine Technical Manual, 2nd Edition, 2003

Note: In case Blood Bank is using kit of other manufacturer the SOP procedure, validation and Interpretation is to be modified as per the literature in the kit insert.

Standards Operating Procedures
Bio-Medical Waste Management
Index

Name of Blood Bank
Name of Hospital Delhi
License no -

Number **BMWM/00**	**Effective Date**	**Page** **3**	**Author**	**Authorized**
Version – 04	**Review period** **2 years**	**No. Of copies:5**	**Approved By**	**Revision Date**
Location **Working areas**		**Subject** **Bio-Medical Waste Management**		
Function **General Information**		**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Technical In charge concerned lab.** • **Master File.**		

1.0 PURPOSE

Blood Bank must manage Infectious/ non infectious waste in order to minimize potential personal exposure and to assure environmentally sound disposal of biomedical waste.

2.0 SCOPE

2.1 All biomedical waste generators at each point within the department comply with this policy.

2.2 Minimize the amount of waste generated

2.3 Assist in the identification, classification and segregation of biomedical waste.

3.0 RESPONSIBILITY

It is the responsibility of the designated staff to dispose off waste according to SOP with help of lab attendant. Technical supervisor/ Nursing Sister is to cross check the method of disposal & documentation.

4.0 ACTIVITY

4.1 Categories of regulated waste in Blood Bank

4.1.1 Liquid or semi liquid human blood, blood component and products made from human blood > 20 ml in volume.

4.1.2 Contaminated items that would release blood or body fluid in a liquid state when compressed, such as soaked cotton swab, gauze pieces and surgical sponge.

4.1.3 Sharps including needles, syringes with attached needles, sample tubes and micro-cuvettes.

4.2 Nature of waste Produced

Blood Bank generates both Non – hazardous and Hazardous type of waste – the hazardous being potentially infectious.

4.2.1 Needles, Syringes, Lancets, blood bag needle.
4.2.2 Blood sample in vacutainers and tubes, yellow tips.
4.2.3 Blood bags: Expired blood & blood products or found reactive in mandatory tests (HBsAg, HIV- I & II, HCV, VDRL and Malaria).
4.2.4 Miscellaneous: Cotton swabs, filter or tissue Paper, disposable gloves, empty carton etc.
4.2.5 The Ministry of Environment and Forests have classified the Biomedical Wastes (as mentioned previously), which is notified in the Biomedical Waste handling and management rules.
4.2.6 Non-Hazardous and Hazardous waste is managed as per Hospital Protocol for segregation and final disposal.

4.3 Operational guidelines for waste management in blood bank.

4.3.1 Always dispose of sharps on your own.
4.3.2 Never pass used sharps directly from one person to another.
4.3.4 During exposure prone procedure, the risk of injury should be minimized by ensuring that the operator has the best possible visibility, e.g. by positioning the donor, adjusting well light source and controlling bleeding.
4.3.5 Never recap, bend or break disposable needles.
4.3.6 Directly after use, place needles and syringes in a rigid container until ready for disposal.
4.3.7 Locate sharps disposal containers close to the point of use.
4.3.8 Do not dispose used sharps in any other waste container.
4.3.9 Prevent overflow by sending sharps disposal containers for autoclaving when three – quarters full.

HANDLING SYRINGES AND NEEDLES

DO'S	DON'TS
• Pass syringes & needles in a tray preferably cut with needles cutter.	• Never pass syringe &needles directly to the next person.
• Put needles and syringes in 1% hypo chlorite solution if needle cutter is not available.	• Do not bend or break used needle with hand.

• Remove cap of needle near the site of use.	• Never test the fineness of the needle's tip before use with bare or gloved hand.

• Pick up open needle from tray / drum with forceps.	• Never pick up open needles by hands.
	• Suck air in and out of syringe & needle after removing from packing and before taking blood sample.
	• Never recap used needles

5 RECORD

Enter the time, nature of material, quantity, reason for disposal into waste management / disposal register with initials of designated technician.

6 REFERENCE

- Transfusion medicine Technical manuals, DGHS, 2nd edition.
- Biomedical Waster (Management and handling) rules 1998, Ministry of environment & Forest Notification

Name of Blood Bank
Name of Hospital Delhi
License no -

Number BMWM/00	Effective date	Page 2	Author	Authorized by
Version – 0	Review period 2years	No. of copies:5	Approved By	Revision Date

Location Working areas	Subject Bio-Medical Waste Management
Function Management of Blood Spill	Distribution • HOD • Quality Manager • Technical Manager • Technical In charge concerned lab. Master File.

1.0 PURPOSE

Disinfect the blood spillage area.

2.0 SCOPE

To avoid the spread of infection & to prevent the personnel from exposure to disease which have a potential of transmitting through blood.

3.0 RESPONSIBILITY

In case of spilling of the blood or blood product by a person, he/she should take the responsibility to ensure management of that blood spillage area. If spilled material is unscreened and / positive for TTI extra precaution should be taken. Lab attendant/ housekeeping staff are to clean the surface under the guidance of that person.

4.0 ACTIVITY

4.1 Material

4.1.1 4% sodium hypo chlorite solution
4.1.2 Paper towel / blotting paper
4.1.3 Gloves
4.1.4 Dustbin

4.2.1 Procedure

4.2.1 Spills on the floor, of infected or potentially infected material should be covered with paper towel / blotting paper / newspaper.

4.2.2 Pour adequate amount of 4% Sodium – hypochlorite solution and cover the area of spillage with paper towel/blotting paper/ newspaper on the area.

4.2.3 Keep it covered for at least 20 – 30 minutes and should be removed with gloved hands and discarded in infectious waste, as per guidelines.

5. RECORD

5.1 Enter the time, nature of material spilled, into waste management/ disposal register with initials of designated staff/technician.

5.2 Enter the screening status of spilled blood product.

6.0 REFERENCES

- Transfusion medicine Technical manuals, DGHS, 2nd edition.
- Biomedical Waster (Management and handling) rules 1998, Ministry of environment & Forest Notification

Name of Blood Bank
Name of Hospital Delhi
License no -

Number BMWM/003	Effective Date	Page 2	Author	Authorized By
Version – 04	Review period 2years	No. of copies:5	Approved By	Revision Date

Location Sterilization Room	Subject Biomedical Waste Management
Function Autoclaving of blood bags and other infected materials	Distribution • HOD • Quality Manager • Technical Manager • Technical In charge concerned lab. • Master File.

1.0 PURPOSE

Autoclave the blood bags (TTI reactive & others) and other infected material for safe disposal

2.0 SCOPE

Infectious substances, including regulated medical waste are one class of hazardous materials regulated under the Hazardous Medical Regulation.

3.0 RESPONSIBILITY

It is the responsibility of the Lab attendant to autoclave the blood units and other infectious material under the supervision of Technical supervisor /Nursing staff.

4.0 ACTIVITY

4.1 Blood Bags to be discarded

4.1.1 Expired blood

4.1.3 Under / over collection

4.1.4 Blood found reactive after screening for mandatory testing (HBsAg, HCV, HIV I &II VDRL and MP)

4.1.5 Blood unsuitable for transfusion due to any other reason. (Hemolysed, used for quality control purpose, clotted, leaked etc.)

4.1.6 Blood bag received back for the follow – up of adverse transfusion reaction.

4.1.6.1 Unused blood retuned from wards.

4.1.7 Other potentially infected materials

4.2 Material

4.2.1 Autoclave

4.2.2 Blood bags to be discarded

4.3 Method

4.3.1 Record the particulars of the bag in the register e.g. Donor's number, Date of collection, Expiry, Screening Report, reason for disposal.

4.3.2 Keep the bag in vertical position (standing) in polybags

4.3.3 Follow universal precautions while handling the Blood Bags.

4.3.4 Autoclave the container with bags at 121ºC &15 Lbs for 20 minutes.

4.3.5 After autoclaving, handover the container with bag as such to the authorized person and take his signature as per hospital policies for for disposal as per the BMW guidelines

4.3.6 Maintain the entries in the register.

Note: All sharps should be disposed in sharp containers, autoclaved and send to authorized personnel from Biomedical Waste Department for central disposal .

5.0 RECORD

5.1 Enter the date, time, Status of blood bags into waste management / disposal register with initials of designated Technician/nursing staff countersigned by Technical Supervisor/ authorized official.

5.2 Enter the screening status of blood product.

5.3 Take the signature of person to whom autoclaved material is handed over in hand over registers.

6.0 REFERENCE

- Transfusion medicine Technical manuals, DGHS, 2nd edition.
- Biomedical Waster (Management and handling) rules 1998, Ministry of environment & Forest Notification

Standards Operating Procedures
Adverse Incident Reporting
Index

Name of Blood bank

Name of Hospital Delhi-1100

License no -

Number **AIR/00**	**Effective Date**	**Page** **2**	**Author**	**Authorized**
Version - 04	**Review period** **2years**	**No. of copies:5**	**Approved By**	**Revision Date**
Location **Quality Control Lab**		**Subject** **Non conforming standards**		
Function **Non conformity of quality in Blood Components**		**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Technical In charge concerned lab.** • **Master File.**		

1.0 SCOPE & APPLICATION:

All the blood and blood components which are not meeting the quality standards as specified in the Quality manual should be recorded and decision for use will be taken by the HOD

2.0 RESPONSIBILITY:

(i) It is the responsibility of all the technical staff posted in component lab and Technical supervisor who will submit the report to the Quality Assurance Manager.

(ii) The Quality Assurance Manager is responsible to review and report to the HOD for further investigation and decision.

3.0 DEFINITIONS:

Non conforming standards:

- Any product which is beyond the accepted standards as specified in the Quality mannual

4.0 CORRECTIVE ACTION:

- Is required for error and incident reports and is usually connected to a process improvement activity. It is an immediate remedial action taken to correct the effect of a defined event.

5.0 Preventive Action:

- Follow up action taken to prevent a defined event from re-occurring.

6.0 Incident:

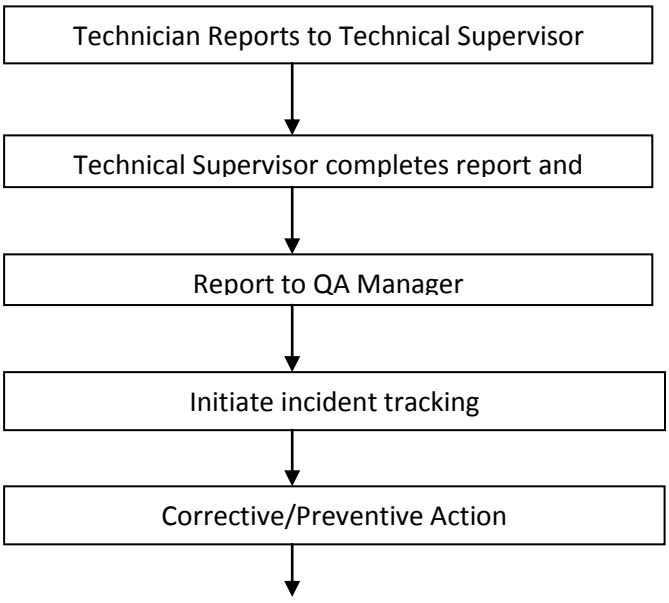
- An Event that results from a deviation from a system, process or procedure that may affect the

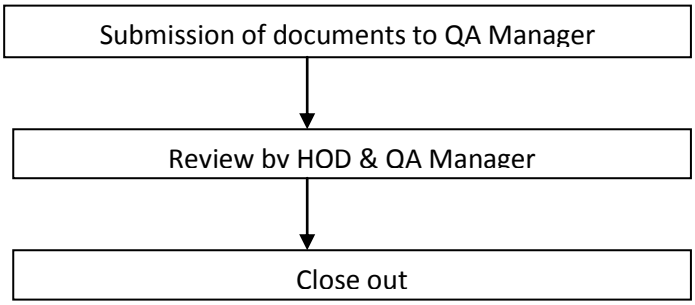
(i) Safety, purity, potency or effectiveness of the product.
(ii) Health or safety of a donor, product recipient, member of staff/public.
(iii) Traceability of records.
This event may have been identified either prior to or after distribution of a product or service.

7.0 PROCEDURE:

(i) Document all incidents on the standard form (Incident Report Form/ register).
(ii) Forward the incident summary report to the Technical supervisor for evaluation and completion.
(iii) Initiate incident tracking..
(iv) Develop corrective/preventive Action in consultation with Technical Supervisor, QA Manager and the HOD.
(v) Forward original documents to the QA Manager within 3 working days of the event.
(vi) The QA Manager reviews the report for completeness and appropriateness of corrective action.
(vii) The status of an event remains active until effective action is taken and closed out. Record the details, date of action and close out and get the report forms signed by the HOD.
(viii) Notify the HOD immediately in case of critical incidents such as those that could result in loss of life, product recall, failure to operate or adverse publicity
(ix) Provide monthly summary reports to the HOD

8.0 Flow Chart for Incident Reporting Process:





9.0 DOCUMENTATION:

Record all incidents on a incident report form/ register. File all record forms.

10.0 REFERENCES

- Transfusion medicine Technical manual 2nd edition, 2004.
- NABH Blood Bank standards 2nd edition, June 2013

Name of Blood Bank

Name of Hospital Delhi-1100

License no -

Number	**Effective Date**	**Page**	**Author**	**Authorized**
AIR/00		**6**		
Version - 04	**Review period**	**No. of copies**	**Approved By**	**Revision Date**
	2years	**5**		

Location	**Subject**
Quality Control Lab	**Adverse incident Reporting**
Function **Mechanism for correction and Prevention of errors and incidents**	**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Technical In charge concerned lab.** • **Master File.**

1.0 SCOPE & APPLICATION:

The procedure covers all incidents that would affect the quality of blood products & services. The procedure applies to all incidents, adverse reactions, equipment used in collection, testing & storage of blood products. The incident reporting process should be clearly defined so that information is traced and acted upon.

2.0 RESPONSIBILITY:

(iii) It is the responsibility of all the technical staff to report any incident/accident to the Technical supervisor who will submit the report to the Quality Assurance Manager.

(iv) The Quality Assurance Manager is responsible to review and report to the HOD for further investigation and implementation of remedial measures if any.

3.0 DEFINITIONS:

Incident Reporting:

- Is a process improvement tool that is used to identify problems, analyze the cause, develop solutions, execute the solution and track the effectiveness.

4.0 CORRECTIVE ACTION:

- Is required for error and accident reports and is usually connected to a process improvement activity. It is an immediate remedial action taken to correct the effect of a defined event.

5.0 Preventive Action:

- Follow up action taken to prevent a defined event from re-occurring.

6.0 Incident:

- An Event that results from a deviation from a system, process or procedure that may affect the
  (iv) Safety, purity, potency or effectiveness of the product.
  (v) Health or safety of a donor, product recipient, member of staff/public.

(vi) Trace ability of records. This event may have been identified either prior to or after distribution of a product or service.

7.0 PROCEDURE:

(x) Document all incidents on the standard form (Incident Report form/ Register).

(xi) Forward the incident summary report to the Technical supervisor for evaluation and completion.

(xii) Initiate incident tracking.

(xiii) Develop corrective/preventive Action in consultation with Technical Supervisor, QA Manager and the HOD.

(xiv) Forward original documents to the QA Manager within 3 working days of the event.

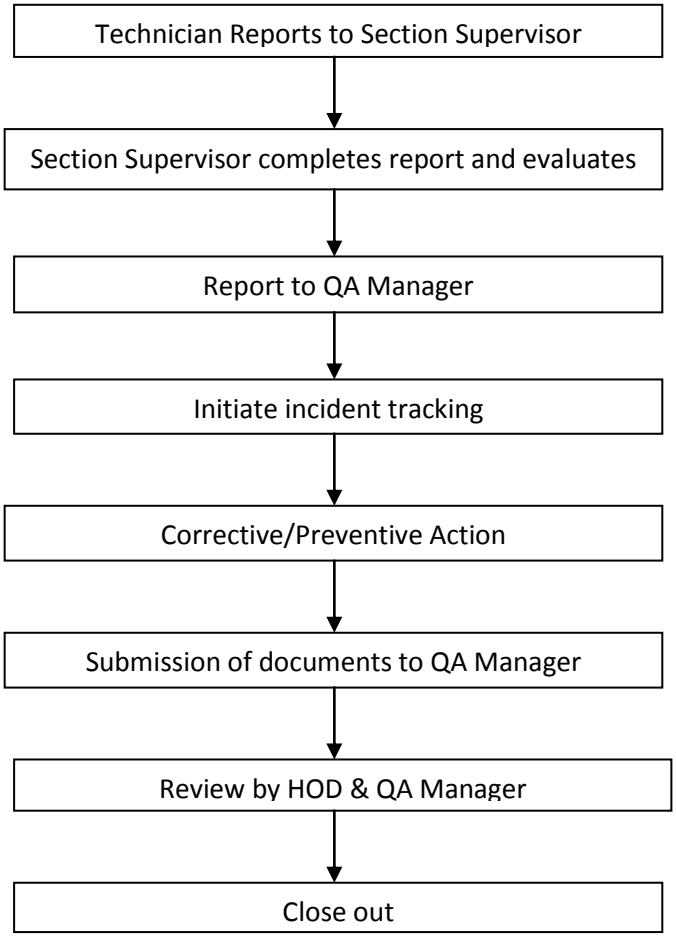
(xv) The QA Manager reviews the report for completeness and appropriateness of corrective action.

(xvi) The status of an event remains active until effective action is taken and closed out. Record the details, date of action and close out and get the reports form signed by the HOD.

(xvii) Notify the HOD immediately in case of critical incidents such as those that could result in loss of life, product recall, failure to operate or adverse publicity

(xviii) Provide monthly summary reports to the HOD

5 Flow Chart for Incident Reporting Process:



5.0 DOCUMENTATION:

Record all incidents on a incident report form. File all record forms.

6.0 REFERENCES

- Transfusion medicine Technical manual 2nd edition, 2004.
- NABH Blood Bank standards 2nd edition, June 2013

STANDARD OPERATING PROCEDURES
QUALITY CONTROL IN BLOOD BANKING

Index

Sl. No.	QUALITY ASSURANCE	SOP No.
1.	1.To approve & random quality check of Antisera	QC/001
2.	2.Monitoring Blood Component Quality	QC/002
3.	3.Equipment Maintenance	QC/003
4.	4.Preparation of Levy –Jennings Chart/QC chart	QC/004
5.	5. Mechanism for correction and Prevention of error and incidents	QC/005
6.	6. Non conformity of quality in Blood Components	QC/006

Name of Blood Bank
Name of Hospital Delhi-
License no –

Number QC/0	Effective Date	Page 4	Author	Authorized
Version - 04	Review period 2 years	No. of copies:5	Approved By	Revision Date
Location Quality Control Lab			Subject Quality Control	
Function Quality check of Blood Group Antisera			Distribution • HOD • Quality Manager • Technical Manager • Designated Lab. • Master Copy	

1.0 PURPOSE

Reagent quality control programme is to ensure that the reagent functions as expected.

2.0 SCOPE

The potency & specificity of reagent may be compromised in certain circumstances, such as contamination of open vials, erroneous labeling or unfavorable environmental conditions during shipment and / or storage. It is therefore a good policy to check the specifications of all reagents at the user laboratory, including those that have been officially licensed. In all cases, use of reagents should confirm with the methods specified in the manufacturer's package insert.

3.0 RESPONSIBILITY

It is the responsibility of the designated staff to store all the reagents as per manufacturer's instructions. It should be reviewed regularly by the Technical Supervisor & any change in the procedure be recorded in the Antisera QC format.

4.0 ACTIVITY

4.0.1 **Appearance**

No turbidity, precipitates, particles or gel formation by visual inspection

4.0.2 **Specificity**

Positive reaction with red cells having corresponding antigen(s); and no reaction with negative control

4.0.3 **Avidity**

It is a measure of the speed with which the antiserum agglutinates with the corresponding red cells.

4.1 Material Required

ANTI -A

- 10% of A cell suspension
- Glass test tube 12 x 75 mm
- Anti serum- A
- Lab centrifuge
- Microscope

- Timer/ stop watch

Procedure

- Put 1-2 drops of (10%) A cells on a glass test tube
- Add 1 drop of Anti serum A and mix
- Simultaneously start the stop watch
- Stop the watch as soon as agglutination becomes visible

Avidity:10-12 seconds
Appearance should be clear
Material Required

ANTI- B

- 10 % B cell suspension
- Glass tube 12 x 75 mm
- Anti serum – B
- Lab centrifuge
- Microscope
- Timer/ stop watch

Procedure

- Put 1-2 drops of 10% B cells on a glass test tube
- Add 1 drop Anti – B and mix
- Simultaneously start a stop watch
- Stop the watch as soon as agglutination becomes visible
- **Avidity:**10-12 seconds
- **Appearance** should be clear

4.0.3.3 **Material Required**

ANTI- D

- 10% Rh positive cell suspension
- Glass tube 12 x 75 mm
- Anti D serum
- Lab centrifuge
- Microscope
- Timer/ stop watch

Procedure

- Put 1-2 drops of (10%) Rh positive cells on a glass test tube
- Add 1 drop Anti – D and mix
- Simultaneously start a stop watch
- Stop the watch as soon as agglutination becomes visible

Avidity: For IgM anti-D:<10 seconds ,for IgG antiD:10-20 seconds
Appearance should be clear

Anti Serum	Test Cells	Limit
Anti –A	**A**	**10-12 Seconds**
Anti – B	**B**	**10-12 seconds**
Anti – D	**A/B/O Rh Positive Cells**	**IgM<10 Seconds** **IgG 10-20 Seconds**

4.0.4 Reactivity- **NO** immune hemolysis, rouleaux formation or reaction with cells other then specified for.

4.0.5 Titre (Potency)

It is reciprocal of the highest dilution showing weak agglutination, denotes the strength of the Reagent.

4.0.5.1 **Material Required**

- Anti Sera
- Pooled Test Cells of A, B (3%in N/S), O positive (10% in N/S)
- Test Tubes
- Micropipette capable of dispensing 500µL
- Centrifuge
- Normal saline

4.0.5.2. Procedure for Anti-A & Anti-B

4.0.5.2.0 Label a row of test tubes, according to Antiserum dilution (1:2 upto 1:512 or more)

4.0.5.2.1 Put 500µL saline into all tubes

4.0.5.2.2 Add 500µL Anti-serum to tube 1 (dilution 1:2)

4.0.5.2.3 Mix the contents of tube 1 with a micropipette, remove 500µL from the mixture to tube 2

4.0.5.2.4 Continue the same technique, through all the tubes and remove 500µL from the dilution tube of 1:512/ 1:1024 and discard.

4.0.5.2.5 Label another row of test tubes according to Anti-serum dilution (1:2 through 1:512/1:1024)and transfer 2 drops of diluted Anti-serum in the respective tubes from the above diluted Anti-serum

4.0.5.2.6 Add 1 drop of either 3% saline suspension of red cells (for A&B cells)

4.0.5.2.7 Centrifuge at 1000rpm for 1 minute

4.0.5.2.8 Gently re-suspend the red cells and look for agglutination macroscopically and if required under microscope.

4.0.5.3 Result

The agglutination titre is recorded as the reciprocal of the highest dilution showing weak agglutination.

Anti Serum	Test Cells	Acceptance criterion
Anti – A	**3% pooled A cells**	**3+ agglutination at 256 dilution or more,**
Anti –B	**3%pooled B cells**	**3+ agglutination at 256 dilution or more**

4.0.5.3.0 Procedure for Anti-D(IgG&IgM)

4.0.5.3.1 Label 1 row of test tubes, according to Antiserum dilution (1:2 through 1:512)

4.0.5.3.2 Put 500µL saline into all tubes

4.0.5.3.3 Add 500µL Antiserum to tube 1 (dilution 1:2)

4.0.5.3.4 Mix the contents of tube 1 with a micropipette, remove 500µL from the mixture to tube 2

4.0.5.3.5 Continue the same technique, through all the tubes and remove 500µL from the dilution tube of 1:512 and discard.

4.0.5.3.6 Label another 2 rows of test tubes according to antiserum dilution (1:2 through 1:512)and transfer 2 drops of diluted antiserum in the respective tubes from the above diluted antiserum

4.0.5.3.7 Add 1 drop of either 10% saline suspension of red cells into both the rows

4.0.5.3.8 Incubate 1st row at 37ºC for 15 minutes

4.0.5.3.9 Incubate 2nd row at room temperature for 5 minutes and centrifuge at 1000 rpm for 1 minute and note the result

4.0.5.3.10 Centrifuge all the tubes of the first row at 1000rpm for 1 minute

4.0.5.3.11 Gently re-suspend the red cells and look for agglutination macroscopically and note the result.

4.0.5.4 Result

The agglutination titer is recorded as the reciprocal of the highest dilution showing weak agglutination.

Anti Serum	Test Cells	Acceptance criterion
Anti – D, IgG	**10% pooled O+ve cells**	**3+ agglutination at 128 dilution or more,**
Anti –D ,IgM	**10% pooled O+ve cells**	**3+ agglutination at 128 dilution or more**

4.0.6 Corrective Action

4.0.6.1 When a reagent produces result outside the limits set by the manufacturer, corrective action is necessary. The following procedure is suggested for confirming a suspected reagent deficiency.

4.0.6.2 Confirm that established methods are being followed, which do not conflict with the manufacturer‘s instruction. Personnel should be aware of the specific function of each reagent and of its limitations

4.0.6.3 Repeat the testing.

4.0.6.4 Test other vials of the same lot as well as different lots of the reagent. Test should also be done if the same type of reagent is available by another manufacturer and compare the result.

5.0 RECORD

5.0.1 Enter the QC result n the Antisera QC format

5.0.2 Maintain a stock register for all reagents

5.0.3 On receipt, make entries of number of vials received, name of manufacturer, batch number and expiry date in this register.

5.0.4 Issue the reagent for use, only after a QC check is performed.

5.0.5 Enter the date, time, and number of vials issued for use in the stock register.

5.0.6 Order all reagents, as soon as the critical level is reached.

6.0 REFERENCE

- Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
- Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
- Standards for blood bank and blood Transfusion Services NACO 2007
-

Name of Blood Bank
Name of Hospital Delhi
License no -

Number QC/0	**Effective date**	**Page 2**	**Author**	**Authorized**
Version - 04	**Review period 2years**	**No. of copies:5**	**Approved By**	**Revised Date**
Location Quality control Lab			**Subject Quality Control**	
Function Monitoring blood component quality.			**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Designated Lab.** • **Master Copy**	

1.0 PURPOSE

The quality of components is evaluated to verify that each unit contain specified amount of components and attains the specified level of activity.

2.0 SCOPE

It is not possible to test each individual blood component unit but atleast 1% (or 4 samples in a month) random sample of the total units processed must always be evaluated. If result complies with designated values, it can be assumed that similarly processed units are of equal quality and are suitable for transfusion.

3.0 RESPONSIBILITY

It is the responsibility of the quality manager to perform quality control of various blood components. The choice and frequency of the test will depend on the components and their intended function, as well as on the quantity of each type processed.

4.0 ACTIVITY

4.1 Component Evaluation

4.1.1 The selected in vitro measurements & subsequent calculations may provide useful information on the survival & function of blood & blood components, though evaluation effectiveness by in vivo testing is often desirable.

4.1.2 The test result should be within acceptable limit of quality standards as described in the Annexure No.

4.2 Corrective Action:

4.2.1 When test result are unacceptable, corrective action must be taken and recorded

4.2.2 A check should be made whether established component preparation procedures/GMP, including phlebotomy procedures, have been followed.

4.2.3 Proper functioning of the component preparation centrifuge & the acceptability of its own quality control result must be ascertained.

4.2.4 Storage temperature should be checked

4.2.5 The quality control test should then be repeated on the same test sample & on another sample from the same batch since an anomalous result may stem either from an error in testing or from the selection of a non representative sample.

5 **RECORD**

5.1 Enter the details of quality control testing of specified blood component on the respective worksheet. Refer to the annexure for QC worksheet component nd their quality standards.

5.2 A review of result and description of any corrective action should also be recorded on the separate sheet.

6 **REFERENCE**

- Transfusion Medicine, Technical Manual, DGHS, 2nd edition.
- Drug and cosmetic Rules 1945, Schedule F , Part XII B (as amended upto 30th June, 2005)
- Standards for blood bank and blood Transfusion Services NACO 2007

Name of Blood Bank
Name of Hospital Delhi
License no -

Number QC/003	**Effective Date**	**Page 3**	**Author**	**Authorized**
Version - 04	**Review period 2years**	**No. of copies:5**	**Approved By**	**Revised Date**
Location Records & Store Room			**Subject Equipments Maintenance**	
Function Maintenance, Calibration and Validation of Equipments.			**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Designated Lab.** • **Master Copy**	

1.0 PURPOSE

To ensure optimum performance from all the blood bank equipments

2.0 SCOPE

This procedure applies to all the instruments and equipments in use within the Blood Bank.

3.0 RESPONSIBILITY

It is the responsibility of the Technical supervisor to ensure:

- Relevance of equipment requirement
- Prepare/ modify specification and validation reports for new equipment
- Write SOP for all equipments, which define the entire maintenance requirement (e.g. routine/preventive) regardless of whether carried out by internal or external agent.
- Prepare the maintenance schedules for all equipments. The schedule is to include.
  - Préventive maintenance
  - Routine maintenance
  - Extra maintenance
  - Calibration

4.0 ACTIVITY

4.1 Procedures

4.2 Maintenance overview:

4.2.1 Identify& ensure each item of equipment in the unit that requires maintenance.

4.2.2 Ensure all items have an Equipment Identification Number.

4.2.3 Include clear outline of the relevant procedures, routine maintenance and preventive maintenance and cleaning of equipment. Write operator instruction for each item of equipment.

4.2.4 Include those responsible and names of service personnel and maintain a documented log of servicing for all items.

4.2.5 Identify the relevant procedures for equipment maintenance; determine the frequency of calibration and cleaning procedures clearly identifying the times e.g. Daily, monthly etc.

4.2.6 Prepare a complete equipment and instrumentation list consisting of the following headings:

4.2.7 Equipment name / description , ID Number, Serial Number, Model Number

4.3 Calibration: Frequency, referenced documents, performed by, traceability documents

4.4 Preventive maintenance: Frequency, reference documents, performed by.

4.5 Routine maintenance: Frequency, reference documents, performed by.

NOTE: Maintained a list of all equipments and instruments used in all sections / departments in the QC lab to ensure all equipments within the department are documented.

4.6 Maintenance Schedules

Draw up suitable schedules by maintenance type and frequency or by equipment type. Define forward dates for the completion of maintenance and record the date of actual performance in the schedule.

4.7 Service contracts

4.7.1 Contracts need to be in place for all equipment items maintained by an external agent.

4.7.1 Each service contract shall define exactly what is carried out / the frequency and by whom it is completed.

4.7.2 At the completion of the service a maintenance report is to be supplied signed by the contractor and the officer in charge. The report shall detail the work carried out by contractor in the equipment log book.

4.8 Repair & breakdown

4.8.1 Operating instructions for each item of equipment shall identify the steps required to be taken in the event of a fault or breakdown, and shall identify who is responsible for organizing service or replacement.

4.8.2 A log book of errors and corrective actions is to be maintained for all equipment items. In the event of equipment breakdown, it shall be identified as being “OUT OF SERVICE”.

4.9 Maintenance overdue

4.9.1 The Quality Control Laboratory shall determine the suitability for ongoing use of any equipment that has passed its due date for routine maintenance (where this routine maintenance does not involve calibration). The laboratory must document their reasons for continuing to use an item of equipment that is overdue for maintenance. Where appropriate this should include explanation (and supportive evidence where available) that product quality has not been compromised by this delay in maintenance.

4.9.2 Where possible documentary evidence from the manufacturer supporting this decision should be provided. Steps should be taken at the next instance of routine maintenance to evaluate whether any discernible damage has been caused to the equipment by the delay in maintenance.

5.0 RECORD

5.1 Maintain individual files of all equipments including service reports.
5.2 Enter the details of all routine as well as trouble – shooting service calls by the manufacturer's engineer in the equipment maintenance register.

5.3 Maintain a file of all manufacturer's instructions where required, display them close to the equipment.
5.4 Record the name address and telephone no. of the service engineer to be contacted in case of need.

6 REFERENCE

NABH Blood Bank Standards 2nd Edition, June 2014

Name of Blood Bank
Name of Hospital Delhi
License no -

Number **QC/00**	**Effective Date**	**Page** **2**	**Author**	**Authorized**
Version - 0	**Review period** **2 years**	**No. Of copies:5**	**Approved By**	**Revised Date**
Location **Quality Control Lab**			**Subject** **Quality Control**	
Function **To prepare Levy-Jennings (LJ) chart**			**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Designated Lab.** • **Master Copy**	

1.0 PURPOSE

It is a measure which must be undertaken for each assay ***to ensure*** that the test is working accurately as per the limits of the test so as to produce valid and acceptable results

2.0 SCOPE

An LJ Chart helps detection of the following:

- Systematic variation
- Random variation
- Lot to lot variation
- Day to day variation

It indicates

1. That the test is valid
2. That all test conditions for that run have been met

3. That all test results for that run are reliable

3.0 RESPONSIBILITY

It is the responsibility of the designated Quality manager to prepare LJ Charts

4.0 ACTIVITY

4.1 Procedure

- Include at least 30 runs on the same graph
- Mean and ± 2SD is calculated& plotted on the graph.
- Values are plotted on the Y axis in the chart and consecutive dates of runs are plotted on X axis.
- Change of operator and batch of assay should be recorded

4.1.1 **Mean** is calculated as $\sum X / n$, where $\sum X$= Sum total of values and n = Number of reactive sample taken for calculation of LJ chart

4.1.2 **Standard deviation** = each of the individual values are compared with the mean (x) to find out the deviations from the mean.

4.1.3 The deviation will be expressed as 'd'. The deviations are then squared.

These squared deviations are added and expressed as $\sum d^2$ or $\sum (x_n - x)^2$ the result is then divided by the number of readings.

4.1.4 The square root of the above value is taken to find out standard deviation.

4.1.5 SD = $\sqrt{\sum (x_n - x)^2/n}$

4.1.6 **Coefficient of Variation** -it is expressed as a percentage and the following formula is used.

CV (%) = SDx 100/Mean (X)

CV less than 10% is considered as an indication of little variation

5.0 Interpretation

5.0.1Systematic Variation

Trend-Results change gradually in either direction indicating slowly changing parameters-deteriorating reagents, equipment failing

Shift-Results fall sharply on one side of the mean(±2SD) indicating a major change has occurred-Switching to new lot of kits, New reagents, changes in incubation temperature or new technical hand

5.0.2Random Variation

- Observance of one result significantly different from other results without any pattern may be due to:
  - Transcription errors
  - Sample mix-up
  - Poor pipette precision
  - Poor mixing of samples
  - Reader not calibrated
  - Washing inconsistent

6.0 CORRECTIVE ACTION

If any variation is noted in the chart, Quality manager is to look into the matter, root cause analysis(RCA) to be done and corrective action should be taken. The Blood Bank In charge should be informed.

7.0 RECORD

Print-out of the charts along with the interpretation and corrective measures, if any, should be recorded in the designated register/ file.

8.0 REFERENCE

NABH Blood Bank Standards 2nd Edition, June 2014

Name of Blood Bank

Name of Hospital Delhi-1100

License no

Number QC/00	**Effective Date**	**Pages :** 6	**Author**	**Authorized**
Version - 0	**Review period 2years**	**No. of copies:5**	**Approved By**	**Revision Date**

Location Quality Control Lab	**Subject** Adverse incident Reporting
Function Mechanism for correction and Prevention of error and incidents	**Distribution** • HOD • Quality Manager • Technical Manager • Technical In charge concerned lab. • Master File.

1.0 SCOPE & APPLICATION:

The procedure covers all incidents that would affect the quality of blood products & services. The procedure applies to all incidents, adverse reactions, equipment used in collection, testing & storage of blood products. The incident reporting process should be clearly defined so that information is tracked and acted on and feedback provided.

2.0 RESPONSIBILITY:

(v) It is the responsibility of all the technical staff to report any incident/accident to the section supervisor who will submit the report to the Quality Assurance Supervisor/Manager.

(vi) The Quality Assurance Supervisor/Manager is responsible to review the completed report and report to the Director for further investigation and implementation of remedial measures if any.

3.0 DEFINITIONS:

Incident Reporting:

- Is a process improvement tool that is used to identify problems, analyse the cause, develop solutions, execute the solution and track the effectiveness.

4.0 CORRECTIVE ACTION:

- Is required for error and accident reports and is usually connected to a process improvement activity. It is an immediate remedial action taken to correct the effect of a defined event.

5.0 PREVENTIVE ACTION:

- Follow up action taken to prevent a defined event from re-occurring.

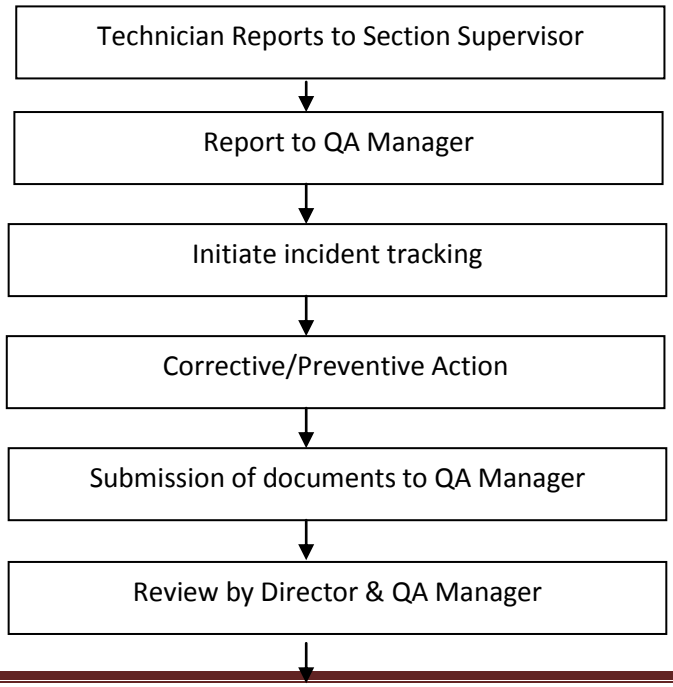
6.0 INCIDENT:

- An Event that results from a deviation from a system, process or procedure that may affect the
  (vii) Safety, purity, potency or effectiveness of the product.
  (viii) Health or safety of a donor, product recipient, member of staff/public.
  (ix) Trace ability of records.
  This event may have been identified either prior to or after distribution of a product or service.

7.0 PROCEDURE:

(xix) Document all incidents on the standard form (Incident Report Form).
(xx) Forward the incident summary report to the section supervisor for evaluation and completion.
(xxi) Initiate incident tracking..
(xxii) Develop corrective/preventive Action in consultation with Section Supervisor, QA Manager and the HOD.
(xxiii) Forward original documents to the QA Manager within 3 working days of the event.
(xxiv) The QA Manager reviews the report for completeness and appropriateness of corrective action.
(xxv) The status of an event remains active until effective action is taken and closed out. Record the details, date of action and close out and get the reports form signed by the HOD.
(xxvi) Notify the HOD immediately in case of critical incidents such as those that could result in loss of life, product recall, failure to operate or adverse publicity
(xxvii) Provide monthly summary reports to the HOD

Flow Chart for Incident Reporting Process



Close out

5.0 DOCUMENTATION:

Record all incidents on a incident report form. File all record forms.

6.0 END OF DOCUMENT

7.0 REFERENCE

NABH Blood Bank Standards 2nd Edition, June 2014

Name of Blood Bank

Name of Hospital Delhi-1100

License no -

Number QC/0	**Effective Date**	**Page** **2**	**Author**	**Authorized**
Version - 04	**Review period 2years**	**No. of copies:5**	**Approved By**	**Revision Date**
Location **Quality Control Lab**		**Subject** **Non conforming standards of Blood products**		
Function **Non conformity of quality in Blood Components**		**Distribution** • **HOD** • **Quality Manager** • **Technical Manager** • **Technical In charge concerned lab.** • **Master File.**		

1.0 SCOPE & APPLICATION:

All the blood and blood components which are not meeting the quality standards as specified in the Quality manual should be recorded and decision for use will be taken by the HOD

2.0 RESPONSIBILITY:

(vii) It is the responsibility of all the technical staff posted for component separation and section supervisor who will submit the report to the Quality Assurance Supervisor/Manager.

(viii) The Quality Assurance Supervisor/Manager is responsible to review the completed report and report to the HOD for further investigation/ decision taking.

3.0 DEFINITIONS:

Non confirming standards:

- Any product which is beyond the accepted standards as specified in the Quality manual

4.0 CORRECTIVE ACTION:

- Is required for error and accident reports and is usually connected to a process improvement activity. It is an immediate remedial action taken to correct the effect of a defined event.

5.0 Preventive Action:

- Follow up action taken to prevent a defined event from re-occurring.

6.0 Incident:

- An Event that results from a deviation from a system, process or procedure that may affect the

(x) Safety, purity, potency or effectiveness of the product.

(xi) Health or safety of a donor, product recipient, member of staff/public.

(xii) Trace ability of records.

This event may have been identified either prior to or after distribution of a product or service.

7.0 PROCEDURE:

(xxviii) Document all incidents on the standard form (Incident Report Form).

(xxix) Forward the incident summary report to the section supervisor for evaluation and completion.

(xxx) Initiate incident tracking

(xxxi) Develop corrective/preventive action in consultation with Section Supervisor, QA Manager and the HOD.

(xxxii) Forward original documents to the QA Manager within 3 working days of the event.

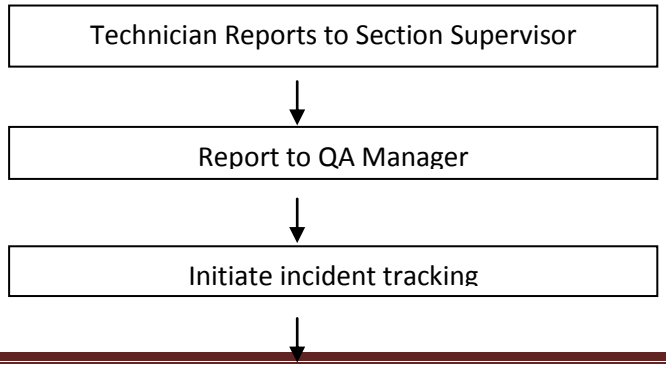
(xxxiii) The QA Manager reviews the report for completeness and appropriateness of corrective action.

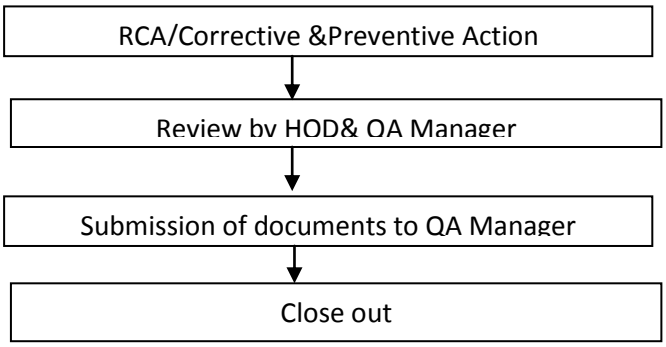
(xxxiv) The status of an event remains active until effective action is taken and closed out. Record the details, date of action and close out and get the report form signed by the HOD.

(xxxv) Notify the HOD immediately in case of critical incidents such as those that could result in loss of life, product recall, failure to operate or adverse publicity

(xxxvi) Provide monthly summary reports to the HOD

Flow Chart for Incident Reporting Process





7.0 DOCUMENTATION:

Record all incidents on a incident report form/ register. File all record forms.

8.0 REFERENCE

NABH Blood Bank Standards 2nd Edition, June 2014

POLICY FOR BLOOD STORAGE UNIT

Scope:

To provide safe blood and blood components to the patients admitted in first referral unit

Facilities available:

- Whole Human blood
- Fresh Frozen plasma
- Packed RBC
- Platelet concentrate

Limitations:

- No bleeding facility. Every donor has to go to the Mother Blood Transfusion Centre for donation and collect the donor card.

Beneficiaries:

In house admitted patients who need transfusion.

Responsibility:

The availability and compatibility testing is the responsibility of Blood Storage Unit In charge

The responsibility of motivating donors for Blood donation and transfusion of blood and its components **is the responsibility of the respective department Head and doctor on duty**.

Procedure:

Request for blood / blood components are sent to Mother Blood Transfusion Centre,…… Hospital, New Delhi, as per requirement of the hospital. Every blood storage centre can keep available five units of A,B,O (positive), two units of AB(positive) and one unit of A,B,O(negative) whole blood . This can be modified according to the actual requirements. The whole Blood / blood components are procured from the mother centre along with issuing letter

which matches the blood bag number written on blood units and special attention given by the BSU Technician to check blood units for hemolysis, leakage, turbidity and change in colour if any. Ensure certificate for the issued blood units have been tested for Malaria, HBsAg, HCV, STS and HIV and found non-reactive.

The blood units are brought to Blood Storage Centre in transport carrier under recommended cold chain Thereafter, these blood units are kept in refrigerators according to respective blood / blood component shelf.

1. Request

a. To ensure patients safety, blood / blood components should not be prescribed unless there is a real indication.

b. Request should be made by a consultant/ Senior Residents.

c. Blood transfusion request form should be filled completely by a doctor

d. Consent for transfusion should be taken from patient / guardian after explaining the transfusion requirement and adverse effects of Blood transfusion by the doctor on duty.

e. In case of unaccompanied or unconscious patients who are incapable of giving consent ,it can be given by minimum two treating doctors stating that transfusion is absolutely necessary to save the life of the patient.

f. Blood sample should be taken for ABO & Rh grouping and cross matching and labelled at bedside in both EDTA and serum vials.

f. The entire request for blood / blood component should be sent to the Blood Storage Centre.

g. The blood/components will be released only on exchange basis i.e. one donor per request (and not per adult volume).

h. Blood components will be released without donor only in emergency life threatening situations with proper justification by the dealing Consultant/ SR with signature and stamp.

2. Grouping

a. All patients should be grouped in case:

i. Any intermediate or major surgery is planned

ii. Any invasive procedures are planned where a risk exists and blood transfusion is a possibility

iii. Transfusion of blood / blood components is planned

iv. The patient is diagnosed with a medical disease with likelihood of blood /blood components transfusion requirement.

v. For FFP and Platelets only grouping is recommended.

3. Blood Reservation

a. Blood should be reserved before all elective surgeries the procedure will be as under: The requisition for blood and the blood sample will be sent to the blood storage center immediately on admission / when the need for transfusion is established.

ii. Availability of the donor (for replacement) must be provided by the requesting doctor.

iii. The blood storage center will ascertain availability of stocks and inform the same to the consultant / ward nurse.

iv. Cross matching and issue of blood will be done on receiving firm demand from the ward /

OR

v. In case of rare blood groups ('AB' Rh positive and all Rh negative blood groups), the treating consultant will discuss with the medical officer in charge of blood storage centre for availability

before the patients admission in order that suitable arrangements can be made in advance.

vi. In case of non-availability of required blood group, blood storage facility will try to arrange the same from mother blood transfusion Centre.

4. Issuing Blood

a. Blood and blood components will be issued from the BSU only after compatibility testing which normally would take 45-60 minutes for red cells. However for all planned surgeries and requirements the requests for compatibility should be sent well in advance and during routine working hrs only. Issuing will be done on receiving request from the department.

Blood compatibility slip with reaction Performa will be issued by the Blood storage Centre along with all blood products. This has to be kept in patients file and responsibility of the document will be that of the In charge nurse of the floor / ward.

c. After completion of the transfusion, duly filled transfusion reaction/ feedback Performa has to be dispatched to the Blood Storage Centre within 24 hrs of transfusion.

d. The date of collection and expiry of all units of blood / FFP etc. will be mentioned on all units & these will be issued as per inventory maintenance, observing 'first in-first out" (FIFO) policy in order to optimize blood usage.

e. **BSU should follow all the technical procedure as per Blood Bank SOP .**

5. Procedure before Transfusion

a. Blood / Blood components should be checked by the doctor and the following details should be verified: blood bag with patients' blood group & name for correct identification of recipient and counterchecked.

b. Check BP / Pulse / Temperature and record in the case file before transfusion.

6. Procedure during Transfusion

a. Monitoring of vitals can be done by nurses
b. Visual observation is often the best way of accessing the patient during transfusion.
c. Record base line observations at the start of each unit and of each transfusion. Temperature / Pulse should be measured 15 minutes after the start of each unit and hourly thereafter. Monitor rate of flow to ensure transfusion progress. Under no circumstances should any drug be administered through the same IV line.
d. Management of Blood transfusion reactions:

Step 1 - Stop transfusion

Step 2 - Keep IV line open with 0.9 % NaCl

Step 3 - Notify attending physician and blood bank, If transfusion is terminated

Step 4 - Send freshly collected post – transfusion sample of blood in EDTA and Plain vials (preferably from opposite arm) and sample of urine to BSU.

Step 5 - Send the residual blood component unit along with administration set to BSU.

e. If no transfusion reactions then after complete transfusion, fill the feedback form and send it to the blood storage center.

f. Whole blood / packed cells can be transfused over 3 – 4 hours.

g. FFP: Can be transfused over 30 minutes

h. Blood should be transfused immediately after receiving. Blood must not be warmed by insertion in hot water, microwave or on a radiator.

i. Blood transfusion set must be changed every 2 units or at least every 12 hourly**.**

Policy of returning unused blood to Mother Blood Transfusion centre:

Screened whole blood and components are procured from mother blood bank, New Delhi once a week.

Whole blood/components are returned to same mother blood bank atleast 07 days before expiry.

8. Policy for discard of expired / contaminated / hemolysed blood begs/ partially used blood bags:

All contaminated hemloysed, expired blood and partially used blood bags are sent for autoclaving first and then dispose off as per BMW guidelines.

9. Policy of Quality Control:

Quality control of Blood and blood product is maintained by the mother blood transfusion centre.

10. Laws governing blood transfusion services:

Blood Bank rules Schedule –F Part XII B of Drug and Cosmetic Rules 1945

11. REFERENCE:

NACO Guidelines for setting up Blood Storage Centre, 2007

Annexure No. 1

POST DONATION INSTRUCTIONS

- Leave band-aid in place for at least 4 hours and not to let it get wet.
- Avoid lifting/ heavy exercise for at least 24 hours
- Increase fluid intake for 48 hours
- Avoid driving for next two hours
- Do not smoke or chew tobacco for4 hours after donation
- Inform a blood bank staff member if you have any unexpected reaction-lightheadedness, nervousness, flushed look or slight perspiration above the lip.
- If there is bleeding from the site where the needle was placed, raise your arm and apply pressure on the bandage until it stops.
- Record your experience on feedback form / register.
- If you have any questions on blood donation, ask blood bank staff before leaving. For any persistent problem ,contact blood bank .

Thank you for your donation. We hope to see you again soon.

Annexure No.2

CHECK LIST OF EMERGENCY DRUGS

Authorized Person: Medical Officer
Responsible Person: Nursing Sister
To check periodically every three months for availability and expiry of medicines

S.N.	INJECTION	S.N.	FIRST AID
1	Dopamine	1	T.T. (Tetanus Toxoid)
2	Hydrocortisone	2	Spirit ammonia aromatic
3	Perinorm	3	Betadine solution
4	Lasix	4	Bandage
5	Deriphyllin	5	Glucose powder
6	Mephentin	6	Band aid
7	Dexamethasone	7	Antiseptic solution
8	Atropine		**MISCELLANEOUS**
9	Avil	1	Hot water bottle
10	Sodium bicarbonate	2	Ice Cap
11	Calcium carbonate	3	Tongue depressor
12	Distilled water	4	Paper bag
13	Adrenaline	5	Towel
14	Noradrenaline	6	Tissue Paper
15	5% glucose	7	Oxygen cylinder with mask
16	Normal saline	8	Syringes
		9	1.V.infusion set
		10	1.V. canula / or scalp vein
		11	1.V.stand

Prepared By……. **Checked by……..**

Date……. **Sign of HOD…….**

Annexure No.3

Quality Control of Antisera

Date of testing: _____

Antisera: _____ Date of Expiry _____

Purpose of testing**: Approval/Random** Lot No. _

Appearance : -……..

Specificity:-……..

Reactivity :-……

Volume :-

Titer :- tested with ……..cells.

Antiserum	1	2	3	4	5	6	7	8	9	10	Result
Ratio	1:2	1:4	1:8	1:16	1:32	1:64	1:128	1:256	1:512	1:1024	
Aggu. Grade.											

Titer :- tested with ……cells.

Antiserum	1	2	3	4	5	6	7	8	9	10	Result
Ratio	1:2	1:4	1:8	1:16	1:32	1:64	1:128	1:256	1:512	1:1024	
Aggu. Grade.											

Avidity:-

☐ Approved ☐ Not approved

Remark:-_____

Signature of Quality Manager

Annexure No.4

Quality Control of Antisera and reagent

Reagent and Solutions	**Frequency of testing**
Anti- human serum	Each day of use
Blood grouping – serums	Each day of use
Lectin	Each day of use
Antibody screening and reverse Grouping cells	Each day of use
Hepatitis test reagents	Each run
Syphilis serology reagents	Each run
Enzymes	Each day of use
HIV I & ll reagents	Each run
Normal saline (LISS and PBS)	Each day of use
Bovine Albumin	Each day of use

Annexure No.5

QUALITY REQUIREMENT OF BLOOD COMPONENT

COMPONENT	PROPERTIES	**PARAMETER TO BE CHECKED**	**QUALITY REQUIREMENT**
Whole Blood		Volume. Sample – 1% of all units with a minimum of 4 units per month Anticoagulant used	350 ml or 450 ml (depending on donors excluding anticoagulant
		Haemoglobin Hematocrit Sterility(after 14 days culture) TTI	> 9.7% w/v ≥45 g/ unit 30-40% No growth Non-reactive
Red cells	Contains all red cells from donated units after centrifugation. No procedures taken to remove leucocyte or platelets.		
		Volume . Sample – 1 % of all units	280 ± 40 ml (when processed from 450 ml) 150 – 180 ml (when processed from 350 ml)
		Haematocrit. Sample –– 1 % of all units	60 – 70%
		Haemoglobin Sample – – 1 % of all unitS	≥45 g / unit
Red cell in additive solution	Contains all red cells from donated units after centrifugation suspended in SAGM.		
	or platelets.		
		Volume Sampling – 1 % of all units	350 ± 20 ml (when processed from 450 ml)300±20 ml (when processed from 350 ml)

		Hematocrit Sampling – 1 % of all units	55-66%
		Haemoglobin sampling – 4 units per month	≥45 g/unit
	If steps taken to remove leukocytes/platelets	WBC count	$<5*10^6$ per unit
Platelet, recovered from single unit PRP	Amount of platelet in adult "standard dose " equivalent to that obtained from 4- 6 units of whole blood		
		ABO group HLA testing when required	
		Volume	50- 70 ml
		Platelet count pH. Swirling RBC contaminants. Residual leukocyte count Sampling – 1% of all units : ≥10 unit per month (75 % of unit should fall within specified values)	$\geq 3.5 \times 10^{10}$/per unit 6.0 – 7.3 at the end of expiry present at days 5 nil $<5 \times 10^7$ / single unit equivalent
Platelets from buffy coat			
		All criteria are same as platelet PRP except leucocytes count. Leucocytes count.	8.3×10^8 leucocytes in 100% of unit tested.
plateletpheresis		All criteria are same as platelet PRP except volume,platelet count	
		Platelet count	$>3.7 \times 10^{11}$

			Leucocytes count.	<5x10^6
			Note: if platelet are filtered then leukocyte count should be	<8x10^5

COMPONENT	PROPERTIES	PARMETER TO BE CHECKED	QUALITY REQUIREMENT
Fresh Frozen plasma	Albumin and immunoglobulins at least 70 % of original, factor V111c, and naturally occurring inhibitors.		
		Volume.	150 – 200 ml if processed from 350 ml 200 – 250 ml if processed from 450 ml
		Appearance	Clear
		Red cells	<6x10^9 / L
		Leucocytes	< 0.1 x 10^6 / L
		Platelets Sample – 1% of all units with minimum of 4 unit / month	<5.0 x 10^6 / L
		Stable coagulation factors	200 I.U.
		Factor V111c	0.7 unit / ml (minimum 70 % of original value)
		Fibrinogen sampling – every 2 month pool of	200 – 400mg
		groups during last month of storage	
Plasma, cryoprecipitate depleted	Content of albumin, immunoglobulin and coagulation factors comparable to FFP. Reduced level of		

	factor V, V111, X111 and fibronection.		
Liqiud Plasma	Content of albumin, immunoglobulin and stable coagulation factors only.		
Cryoprecipitate	Contain a major portion of factor V111 , vWF , fibrinogen, factor X111 and fibronectin present in freshly drawn and separated plasma		
		Volume	10 – 25 ml
		Factor V111c Sample – 1% of all units Every two month pool of 6 unit of mixed blood group during first month of storage.	> 80 I. U. / unit in 75 % of units tested.
		Fibrinogen sampling – 1% of all units: with a minimum of 4 unit per month	>150 mg/ unit
		vWF vWFsampling – 1% of all units: with a minimum of 4 unit per month	40 – 70 % of original 55 mg

Annexure No.6 **Date:**

Quality Control Program of Blood Components: - Cryoprecipitate

S.No.	Donor I/DNo.	Date of Collection	Date of Preparation	Volume (ml)	PT (Sec.)	APTT (sec)	F VIII (IU/ml)	Fibrinogen (g/l)	vWF	Remark

Standards

Volume	10-25 ml
Appearance	Clear
Factor VIIIc	>80 I.U. / Unit in 75% of Unit Tested
Fibrinogen	>150 mg / unit
vWF	40 – 70 % of original 55mg

Signature of I/C Blood Bank **Signature of Q/M.**

Annexure No. 7

Equipments that must be observed, standardized and calibrated with at least the following frequencies

S.No	Equipments	Performance	Frequency	Frequency of Calibration
1.	Temperature Recorder	Compare against thermometer	Daily	As often as Necessary
2.	Refrigerated Centrifuge	Observe speed and temperature	Each day of use	As often as necessary
3.	Haematocrit	-	-	Standardize before initial use, after repair or adjustment and Annually
4.	General lab Centrifuge	-	-	Tachometer every 6 month
5.	Automated Blood Typing	observe controls for correct result	each day of use	--
6.	Hemoglobin Meter	standardized against cyanmethamoglobin	each day of use	
7.	Urinometer	Standardize against distilled water	-ditto -	-
8.	Blood container Weighing device	Standardize against container of known Weight	-ditto-	as often as necessary
9.	Water Bath	observe temperature	-ditto-	-ditto-
10	Rh view box (Wherever necessary)	-ditto-	-ditto-	-ditto-
11	Autoclave	-ditto-	Each day of use	-ditto-
12	Serological Rotators	observe control for correct result	Each day of use	speed as often as necessary
13	Laboratory Thermometers	-	-	before initial use
14	Electronic Thermometer	-	Monthly	-
15	Blood Agitators	observe weight of the first container of blood Filled for correct result	Each day use	standardize with container of known mass or volume before initial use, and after repair or adjustment

दिल्ली सरकार

आप की सरकार

Department of Health & Family Welfare, GNCTD