BLOOD
AND
BLOOD PRODUCTS

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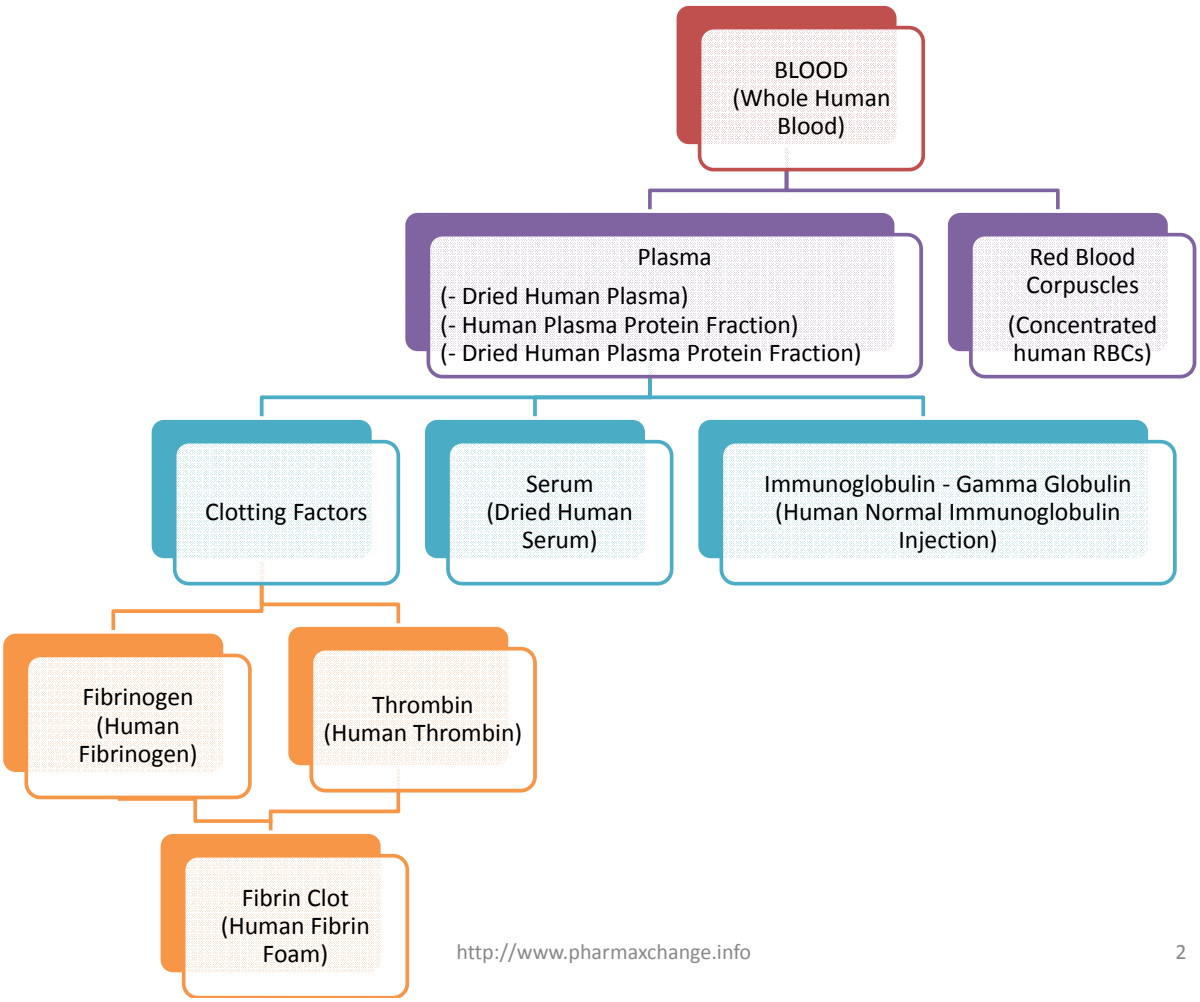


WHOLE HUMAN BLOOD

- Definition : It is the human blood mixed with a suitable anticoagulant

That is :

Human Blood + Anticoagulant = Whole Human Blood

Conditions for Being a Donor

Any person in good health is accepted as a donor provided that he or she:

1) Is not suffering from any disease that can be transmitted by transfusion. This includes syphilis, malaria, and serum jaundice.
2) Is not anemic. The haemoglobin content of the blood should not be less than
   - 12.5% for females
   - 13.3% for males

   (checked by allowing a drop of blood to fall into a copper sulphate solution of specific gravity 1.053 for females, and 1.055 for males. If the drop sinks, the sample is satisfactory)
3) Has been taking medication which might prove toxic or have allergic reactions in a patient e.g. antibiotics

Collection of the blood

- Blood is collected aseptically from the median cubital vein, in the front elbow.
- This blood is put into a sterile container containing an anticoagulant solution and the bottle is gently shaken to ensure that blood and anticoagulant are well mixed, thus preventing the formation of small fibrin clots.
- A maximum of 420ml of blood is taken in one attendance.
- Immediately afterwards the container is sealed and cooled to 4-6 degrees centigrade for storage.

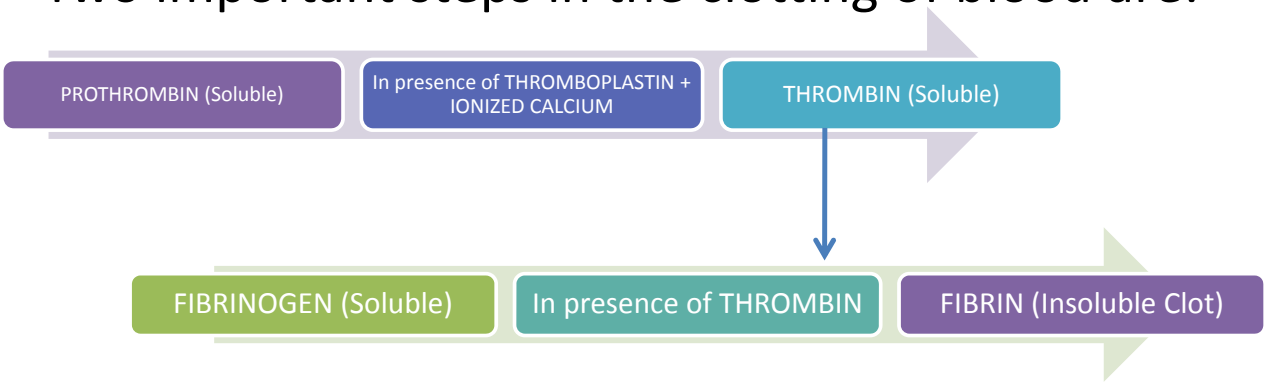
Equipment Used for the Collection

Equipment used for taking the blood is made from plastics, and is disposable

The container earlier consisted of bottles, but Plastic bags have started being used and are the containers of the future.

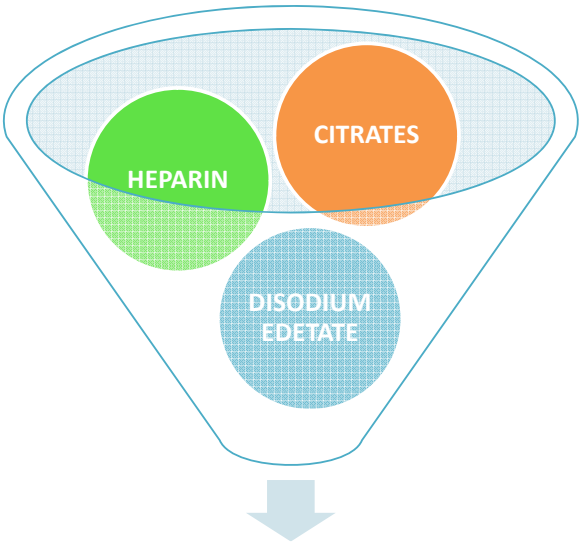
Blood Clotting

Two important steps in the clotting of blood are:



In response to injury, the tissues and blood platelets free substances that activate the clot promoting enzyme THROMBOPLASTIN.
Thromboplastin, with the assistance of ionized calcium and other factors, converts PROTHROMBIN to active clotting enzyme THROMBIN.
Thrombin then acts on FIBRINOGEN, converting it into insoluble FIBRIN, the matrix of the clot.

ANTICOAGULANTS



ANTICOAGULANTS USED

CITRATES

- The solution most often used as a blood anticoagulant is known as Acid-citrate-dextrose (ACD), composed of:
  - Sodium Citrate (2.0 to 2.5 g)
  - Dextrose (3.0 g)
  - Water for Injection (q.s. to 120 ml)
- The citrate prevents clotting by binding the calcium ions as unionized calcium citrate, thus preventing a vital step of clotting.

Why Acid Citrate and not Normal Citrate??

- Earlier the normal, trisodium citrate was used but it has a very high alkaline pH in solution which causes considerable caramelisation of the dextrose (darkening) during sterilization and the two solutions have to be autoclaved separately.
- The Acid Citrate produces a pH of about 5 and causes little or no caramelisation.
- In addition, it is less likely to induce flaking of the glass of the container.
- The higher concentration (2.5g / 120ml) is often preferred because it more effectively reduces the formation of small clots.

Why add Dextrose?

- The dextrose delays haemolysis of the erythrocytes in vitro and prolongs their life after transfusion.
- Its function is hypothesized to be connected with the synthesis of compounds, such as ATP, that are important in making energy available to living cells

HEPARIN

- Naturally occurring anticoagulant.
- Made by the mast cells of the connective tissue surrounding blood vessels.
- It inhibits clotting in the circulatory system.
- Occasionally, it is used in blood for transfusion when large volumes must be given to one patient and the corresponding amounts of citrate would be harmful, e.g. in cardiac surgery.

- It quickly loses activity in blood in vitro and normal quantities are effective for about a day.
- ACD on the other hand, prolongs the storage life to three weeks.
- Heparin is expensive and may continue its action even after transfusion, thus needing administration of neutralizing substances such as protamine sulphate.

DISODIUM EDETATE

- This is also a chelating agent like ACD.
- It has a strong affinity for divalent metals, and thus will bind to calcium firmly.
- It is sometimes preferred when preservation of blood platelets is essential, although the stability of these seems to depend much more on preventing contact with the glass surface: if plastic bags or silicone-treated glass is used, ACD is almost as effective as Disodium Edetate.
- The survival of red blood cells in dextrose-edetate solutions is as good as in ACD.

TESTING OF WHOLE BLOOD

- At the time that blood is collected, two small additional amounts are collected:
  - One, which is often obtained by draining the collecting tube, is put into a small 5 ml bottle and is firmly attached to the main container. This is for testing compatibility with the blood of the recipient. This separate specimen avoids the dangerous procedure of attempting to remove a sample from the main bottle without causing bacterial contamination.
    If a plastic bag is used, it is possible to leave the blood-filled collecting tube attached to the bag and to seal it at several points with a special tool; then a section can be separated for testing without contamination
  - The second, somewhat larger sample is used as soon as possible for :
    - Serological test to confirm the absence of syphilis and other diseases
    - To determine the ABO blood group of the cells and plasma and the Rh grouping of the cells.

BLOOD GROUPS

- Fundamental Aim: is to prevent antigen-antibody reaction.
- Red cells carry an antigen that reacts with the corresponding antibody in the plasma of individuals of certain other groups. If the cells are transfused into an individual with the equivalent antibody in his plasma, they are rapidly destroyed, with serious consequences.
- Although some 9 blood groups are known, only the ABO and Rh are of major importance as causes of haemolytic transfusion reactions

1) ABO System

- The first sign of the haemolytic antigen-antibody reaction is agglutination and, therefore, red-cell antigens and plasma antibodies are called agglutinogens or agglutinins respectively.
- The agglutinated cells haemolyse, freeing haemoglobin and other constituents and causing jaundice and kidney damage: if the latter is extreme, the patient may even die.
- Fortunately most transfusion reactions are mild.

Compatibility Chart

Group	Red Cell Antigen (Agglutinogen)	Plasma Antibody (Agglutinin)	Can Donate To:	Can Receive From:
A	A	Anti-B	A or AB	A or O
B	B	Anti-A	B or AB	B or O
AB	A & B	None	AB only	All groups. (Universal Recipient)
O	O	Anti-A and Anti-B	All groups. (Universal Donor	O only

NOTE - Generally, the recipient's cells are not seriously harmed by the agglutinins in the donor's plasma, because the latter quickly becomes diluted in the recipient's blood (thus a donor of blood group O can donate to all even though it has agglutinins Anti-A and Anti-B)

2) Rh System

- Rh factor, so named cause it was found in the rhesus monkey.
- The red cells of some individuals carry an antigen that is known as the Rh factor.
- If Rh+ blood is transfused into an Rh- recipient, production of antibodies to the Rh+ blood may be stimulated.
- If this occurs, subsequent transfusion of Rh+ blood will cause a haemolytic reaction.

- Haemolytic disease in a new born:
  - If a foetus is Rh+ from it's father, and the mother is Rh- and has the Rh+ antibody in her blood (either from previous transfusion of Rh+ blood or as a result of stimulation by antigens of the foetus), the mother's antibodies may cross the placenta and destroy the foetal erythrocytes.
  - This haemolytic reaction may kill the foetus or cause the infant to be severely anaemic.

STORAGE

- Blood collected must be kept at a temperature between 4 to 6 degrees centigrade, at all times except during short periods of transport and examination, which must not exceed 30 mins.
- Even at this low temperatures, deleterious changes do take place.
  - The leucocytes disintegrate in a few hours
  - The platelets disintegrate in a few days
  - The red cells show a fall in ATP and other organic phosphates, a reduction in oxygen-carrying capacity and, due partly to loss of lipid from their membranes, increased fragility.
- Storage at room temperature even for a day, seriously reduces post-transfusion survival of the erythrocytes

- The fitness of blood for transfusion is based on its appearance. On standing, the cells sediment, leaving a layer of yellow supernatant plasma.
- If the blood has been taken shortly after a heavy fatty meal, the plasma may be turbid and show a white layer of fat on it's surface. On top of the red cells there may be a complete or partial greyish layer of leucocytes.
- The most important feature, however, is the line of demarcation between cells and plasma, which must be sharp: if it is obscured by a diffuse red coloration, indicating haemolysis, the blood is unfit for use.
- Complete haemolysis, especially if it occurs rapidly, is usually a sign of bacterial infection, but its absence is not confirmation of sterility since certain psychrophilic bacteria, predominately pseudomonads and members of the coli-aerogenes group, can grow in blood at refrigerator temperatures without causing haemolysis.
- Many of the organisms isolated from contaminated blood have been capable of using citrate as their sole source of carbon and, as would be expected, this has led to clot formation, as citrate which is the anticoagulant gets assimilated by the bacteria.

USES

- Haemorrhage, shock, burns and uncontrollable diarrhoea and vomiting, can all cause significant losses of blood.
  - Haemorrhage and other diseases may result in deficiency or absence of vital blood constituents such as red cells, platelets, or clotting factors.
  - The transfusion of whole blood can be of great value in all these circumstances but often, because of the risk of transfusion reactions, it is not used where the need is solely to make up blood volume but is restricted to haemorrhage and certain diseases where there is deficiency of the vital oxygen-carrying erythrocytes.
- Normally whole blood is not administered unless the ABO and Rh groups of the donor and recipient are known and a sample of the donor's blood has been tested for compatibility with that of the recipient.
- In an emergency, group O, Rh negative blood may be given while taking necessary precautions.

CONCENTRATED HUMAN RED BLOOD CORPUSCLES

Definition: This is the solution of human RBC's which have been concentrated using centrifugation

Preparation

- It is prepared by removing most of the citrated plasma from whole blood that is not more than a fortnight old and has been allowed to stand or has been centrifuged to deposit the cells

- More than 40% of the supernatant fluid after the settling, is siphoned off using sterile tubes, taking strict aseptic precautions throughout.
- Since there is a risk of bacterial contamination the product must be used within 12 hours.

- The cells are matched with the recipient's plasma and may then be mixed with matched cells from other bottles.
- The haemoglobin content must not be less than 15.5%

Uses

- This product is used when administration of whole blood might overtax the circulation, i.e., in treatment of diseases, such as chronic anaemia (where blood volume has not been reduced), rather than haemorrhage (which would require a replenishment of blood volume as well and thus would require whole blood)
- Another application is in exchange transfusion in infants: a toxic amount of citrate might be given if whole blood was used.

DRIED HUMAN PLASMA

- It is the portion of the blood which has been separated from the cell content, and is dried, and can be used after reconstitution with water.

DRIED HUMAN PLASMA

Disadvantages of Whole Blood

- It has poor keeping properties necessitating use within three weeks
- It requires refrigeration
- It must be compatible with the blood of the recipient

Advantages of Dried Plasma

- Properly stored it keeps well for at least five years
- If protected from light it can be stored at room temperature provided this is below 20 degrees centigrade
- It can be given to patients of any blood group.

THUS, UNDER SUITABLE CIRCUMSTANCES, DRIED PLASMA CAN BE USED AS A SUBSTITUTE FOR WHOLE BLOOD.

Problems to be Overcome During Preparation

- Two major problems have to be overcome
  - Transmission of Viral Jaundice
  - Neutralization of Plasma Agglutinins

1] Transmission of Viral Jaundice

- There are two types:
  - Infective hepatitis (incubation time = 5 weeks, mortality rate = 0.3%)
  - Homologous serum jaundice (incubation time = 20 weeks, mortality rate = 12%)
- Most infections following transfusion are mild
- Control is partly effected by refusing to accept donors with a history of jaundice, but not all cases are recognized and since at present there is no reliable test by which carriers can be detected, an occasional infected bottle is inevitable.
- Attempts have been made to kill the causative viruses by treatment with UV light, but the method is technically difficult.

- Note – if the preparation of a blood product involves pooling material from a larger number of donors, infection in one or two bottles will be distributed throughout the pool and appear in each of the units made from it.
- Nowadays, the pools used for making dried plasma and serum are limited to not more than ten donations, and the incidence of jaundice is only slightly greater than when whole blood is transfused.
- However, in the past, when pools of 300 or more bottles were made, the incidence was 7 to 12%.

2] Neutralization of Plasma Agglutinins

- Agglutinins in the donor's plasma usually do not damage the recipient's red cells.
- Occasionally, however, the plasma agglutinins are very powerful and can cause serious haemolysis of the cells of the recipient.
- This means that incompatibility problems are not entirely eliminated by using products such as plasma and serum, that contain no cells.
- The problem has been overcome since the discovery that red cell agglutinogens also occur as water soluble forms in plasma, saliva and other body fluids.
- Consequently, by mixing plasma from different groups in suitable proportions the powerful agglutinins can be cross-neutralized by soluble agglutinogens, producing a preparation that is safe to transfuse to all groups.
- The most satisfactory ratio for mixing is
  9 of A: 9 of O: 2 of B or AB.