Lesson → 1.

Historical Introduction To Transfusion Medicine (Blood Banking).

1) 1628 ÷ British physician William Harvey discoverd the circulation of Blood.

2) 1932 ÷ First Blood bank was established in Leningrad Hospital.

3) 1939–40 ÷ The RH blood group system was discoved by Karl Landsteiner, Alexender Wiener, Philip Levine and R.E Stetson.

4) 1981 ÷ First acquired immune deficiency syndrome (AIDS) case reported.

5) 1984 ÷ HTV identified as cause of AIDS.

6) William Harvey ÷ Discovered Blood circulation.

7) Lamb, Human ÷ Richard lower successfully transfused Blood from Lamb, Human in 1967.

8) Karl Landsteiner discovered three Blood group A, B and O.

9) In 1939/40 Karl Landsteiner discovered RH Blood group.

10) In 1984 HIV was identified.

11) 1795 ÷ Syng Physick performed the first Blood

transfusion man to man.

12) 1932 ∴ First Blood bank established in Leningrad Hospital.

13) 1981 ∴ First AID case reported.

14) 1984 ∴ ~~HIV~~ identified

15) 1628 ∴ William Harvey discovered Blood circulation

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Lesson → 2.

Antigen and AntiBody.

1) **Antigen** — An antigen is any substance which when enters the body induces the immune system of the body to produce antibodies to neutralise.

2) **AntiBody** — Antibodies are 'Y' shaped protein produced by the immune system of the body to identify and neutralise the antigens.

3) **Haptens** — These are low molecular weight molecules. They can bind to immune cell but fail to induce immune response.

⟹ **Difference between Antigen and Haptem**

S.No	Antigen	Haptem
1)	Complete molecule	Incomplete antigen.
2)	Can bind to the MHC	Cannot bind to the MHC complex.
3)	Can directly bind to the Antibodies.	Cannot directly bind to the Antibodies.
4)	Can trigger immune response by itself.	Cannot trigger immune response by itself.

Classification of Antigens

1) **Autoantigens** — These are endogenous antigen that are recognised as nonself by the immune system which should be considered otherwise.

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under normal conditions.

1) **Isoantigens** → Isoantigen are those substa which have antigenic properties and are present in some individuals of given species. All Human being are divided into four group ÷ A, B, AB and O.

3) **Species Specific Antigens** → Tissues of all individuals in a species posses certain species - specific antigens.

4) **Organ Specific Antigens** → Antigen character istics of an organ.

5) **Heterophile Antigen** → Heterophile Antigen are the shared antigen that are present in many species such as Human, animals, plants as well as micro organisms, without species specific

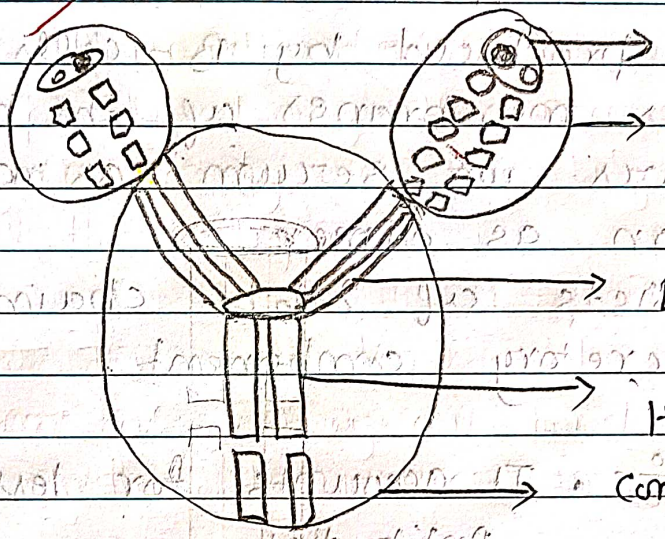
6) **Microbial Antigens** → The invading microbes or pathogens as exogenous Antigen, e.g. viruses, bacteria, fungi, protozoa Helmiths. their secretion and toxins

Properties of Antigens

The Antigen is a foreign substance in induce an immune response.
The Antigen have a molecular mass of 14,000 to

6,00,000 Da.
- They are mainly proteins and/or polysaccharide.
- Antigens are species specific.

Structure of Antibody



Classification of Antibodies

1) IgG. → It is the most abundant type in the Blood and account for 70-75% of Human Antibodies.
- It also possesses the property of opsonisation ie It marks the pathogen to phagocytosis.
- It has a molecular weight approximately 150,000 daltons.
- It molecular formula is H_2L_2 and thus it Has two identical binding sities.

2) IgM → It is the largest of the immunoglobulins
- It is the first antibody to appear in reponse to Antigen.

- It represents 5-10% of the serum Antibodies.
- IgM antibodies are short lived as compared to IgG Antibodies.

3) **IgA** :- It is the second most Abundant class of Antibodies. It account for 10-15% of Human Antibodies.
- It is produced by B-cells located in the mucous membranes of the body.
- It occurs in serum as monomer an in Secretion as dimer.
- It consists of $2H_2L_2$ chains, one joint chain and secretory component.

4) **IgD** :- It accounts for less than 1% of Human Antibodies.
- It exist as monomer.
- It has no Antibody function.
- It molecular weight is about 1,80,000 daltons

5) **IgE** :- Its original role is to protect against parasites.
- The molecular weight is 1,90,000 daltons.
- It does not fix complement.
- It does not cross through placenta.

Antigenic Determinants

7) **Isotypes** :- In isotype Antibodies, the location of Antigenic determinant is the constant region of the heavy or light chain of the Antibody.

2) Allotype :- In allotype Antibodies, the location of Antigenic determinants is the allelic from of Ig genes of the Antibody.

3) Idiotype :- In idiotype Antibodies, the location of Antigenic determinants is on the variable heavy region and variable light region of an Individual Antibody molecules.

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Lesson → 3.

ABO Blood Group System.

1) **Antigen** :- An Antigen is any Substance which when enters the body induces the immune system of the body to product antibodi to neutralise.

2) **Antibody** :- Antibodies are 'Y' shaped proteins produc By the immune system of the body to identify and neutralise the Antigent.

3) **ABO Blood group** :- It defined as the classification of Blood based on the presence and Absence of Antibodies and inherited antigenic substance on the surface of Red Blood cell. The various Blood group A, B, AB and O.

4) **ABO Blood group System** :- The ABO Blood group system is used to denote the presenc of one, both or neither, the presence of antigens on the surface of erythrocytes.

5) **Agglutination** :- It is defined as the clumping of particles. The antigens react with Antibodies. The antigens are called agglutinogens whereas antibodies ar called as Agglutinins. and the reactio

o **Antibodies involved in ABO Blood group System** :-

Antibodies involved in ABO Blood group system are known agglutinins. These Antibodies are present in Blood plasma.

i) Natural Antibodies.
ii) Immune or Acquired Antibodies.

Significance of Blood Groups

The ABO Blood group system is regarded as the most important Blood system in transfusion medicine because of severe haemolytic transfusion reaction and Haemolytic disease to the newborn.

- Blood group A is compatible with Blood group A and AB.
- Blood group AB is compatible with the patient having Blood group AB.
- Blood group B is compatible with Blood group B and AB.
- Blood group O is compatible with the all Blood groups.

⇒ from the above, it clear that :
- Blood group O is universal donor.
- Blood group AB is universal recipient.

Methods to Determine Blood Group

1) Slide Method

⇒ Principle : It is based on the agglutination reactio

When red Blood cell carrying one or both the Antigent are exposed to corresponding Antibodies, they interact with each other to form visible clumping.

⇒ **Requirement** :- i) clean glass slide.
ii) Blood sample.
iii) Alcohol Swabs.
iv) Dropper.
v) Lancet
vi) Anti-A test serum (Blue colour
vii) Anti-B test serum (Yellow)
viii) Marker
ix) Tooth pick.

⇒ **Procedure** :- i) one end of slide is labelled as Anti-A and other end as Anti-B.
2) Allow to stand for 2-3 mint at room temperature
3) The results are read directly from the slide

⇒ **Results** :- • Group A, if agglutination occurs with the Anti-A test serum.
• Group B, if agglutination occurs with the Anti-B test serum.
• Group AB if agglutination occurs with both test serums.
• Group O, if no agglutination in either case.

S.No	Anti-A Reaction	Anti-B reaction	Result.
1.	Agglutination	—	Blood group-A
2	—	Agglutination	Blood group-B
3	Agglutination	Agglutination	Blood group-AB
4	—	—	Blood group O

2) Tube Method :-

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RH Blood Group System.

1) RH Antigens, RH factors :- These are transmembrane protein present on the surface of erythrocytes. RBCs that are RH positive are designated as D ie - RH D failing which RH negative.

2) RH Antibodies :- These are immune antibodies resulting from specific antigenic stimulation such as pregnancy. Thus RH Antibodies are not naturally occurring Antibodies.

3) RH Blood group System :- It is a Human Blood group system and second most important Blood Group system after the ABO Blood group system. RH D, d c, c, E and e. The Blood group d has not so far detected.

4) Direct coomb's test :- It detects Antibodies that are stuck to the surface of the red Blood cells.

5) Indirect coomb's test :- It detect Antibodies that are floating freely in the Blood.

Principle and Procedure of RH Grouping

⇒ Principle :- RH-D typing test is based on the principle of Agglutination. Agglutination

of patient RBC with anti-D serum indicates positive test. Absence of Agglutination is a negative test result for RH-D Antigen.

Methods for the determination

1) Tube Technique Method

⇒ Requirements :- i) Monoclonal Anti-D reagent.
ii) 5% washed red cell suspension
iii) 22% bovine albumin.
iv) Test tube
v) Pasteur pipette.
vi) Centrifuge.

⇒ Procedure :- Take three test tube and label 1st second tubes as test and tube 3rd as control.

2) Place 1 drop of anti-D (D_1) in 1st. tube and 1 drop of anti-D (D_2) in 2nd test tube.

3) Place 1 drop of 22% bovine albumin / control in tube 3rd

4) Add 1 drop of 5% red cell suspension in each tube

5) Mix well, centrifuge at 1000 rpm for 1 minute.

6) Resuspend cell buttom and look for agglutination.

⇒ Result :- i) Agglutination in tube 1 and 2 Shows RH-D positive.

ii) No agglutination in control shows RH-D Negative.

⇒ **Method for Weak RH-D Testing** :- 1) Incubate at 37°c for 30 min.

2) Wash three times with Normal saline.

3) look for agglutination.

⇒ **Result** :- Agglutination :- Du positive.
No Agglutination :- Du Negative.

2) **Indirect Agglutination Test (IAT)** :- It detects Antibodies that are floating freely in the Blood.

⇒ **Requirements** :- i) Test tubes
ii) Test Serum
iii) Coomb's control cell
iv) Monoclonal Anti-D mixture.
v) Antihuman globulin (AHG)
vi) Incubator
vii) centrifuge
viii) microscope.

⇒ **Procedure** :- i) Label three tubes as T (Test), P(+) and N(−) control.
ii) In test tube marked as T, put 2 drops of test serum and 1 drop of coomb control cell.
iii) Wash the cell thrice in each tube with normal Saline.
iv) Add 2 drop of AHG to each test tube.
v) Incubate for 10-15 min. in room temperature.
vi) centrifuge at 1500 rpm for 1 min.
vii) Resuspend cells in Normal saline.

⇒ **Result** :- i) Agglutination in Test and positive control :- RH-D positive.
ii) No Agglutination in Test and Negative control :- RH-D Negative.

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Lesson → 5.

Anticoagulants used in Blood Bank.

1) **Anticoagulants** :- These are the chemical substance which prolong the coagulation of Blood and preserve the Blood in blood Bank.

2) **EDTA (Ethylene diamine tetra acetic Acid)** :- It is most widely used Anticoagulant. It is a powerful calcium chelating agent.

3) **Heparin** :- It is a natural anticoagulant used to prevent clot formation in Blood vessel. It is abundantly available in liver.

4) **ATP (Adenosine triphosphate)** :- It is source of energy for RBC's.

5) **Double oxalate** :- It is a mixture of 3 part of ammonium oxalate and 2 part of potassium oxalate. It is used as Anticoagulant.

Type of Anticoagulant Solution :-

1) Anticoagulant citrate dextrose (ACD) Solution.
2) Citrate phosphate dextrose (CPD) Solution.
3) Citrate phosphate dextrose Adenine-1 (CPDA1) solution.
4) Citrate phosphate dextrose Adenine-1 (CPDA2) solution.

Advantages and Disadvantages of various Anticoagulants :-

1) Double oxalate 2) EDTA 3) Trisodium citrate

4) Heparin:

EDTA (Ethylene diamine tetra acetic Acid).

⇒ Advantage :- i) Preserves cellular morphology of Blood cell
ii) used for platelet count as in inhibits the clumping of platelets.

⇒ Disadvantage :- i) Shrinkage of RBC's and WBC's
ii) Degenrative changes in Blood cells.

Heparin

⇒ Advantage :- i) Resistant to inhibition by activated platelets
ii) Improved pharmacokinetic Profile.

⇒ Disadvantage :- i) It is expensive.
ii) Prevents coagulation for limited time.

ACD-A and ACD-B Solution

ACD-A Solution

Trisodium citrate — 22.0 g
Citric Acid — 8.0 g
Dextrose — 24.6 g
Distilled water — 1000 mL

ACD-B Solution

Trisodium citrate — 13.2g
Citric Acid — 4.8g
Dextrose — 14.8g
Distilled water — 1000mL

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Composition of Anticoagulants

i) **Citrates** :- The sodium citrate bind with the free calcium resulting in the formation of complex. Therefore the process of coagulation of Blood is prevented / Prolonged.

ii) **Citric Acid** :- used with dextrose in anticoagulation preparation.

- Citric Acid regulates pH and thereby protects RBC.
- Used as trisodium citrate and sodium citrate as Anticogulants.

⇒ **Adenine** :- As substrate for ATP Synthesis.

⇒ **Dextrose** :- Provides energy (ATP) to RBC's.

⇒ **Sodium phosphate** :- Acts as buffer.

Citrate Phosphate dextrose (CPD) Solution

Trisodium citrate	:	26.3 g
Citric Acid	:	3.27 g
Dextrose	:	25.5 g
Sodium diphosphate	:	2.28 g
Distilled water	:	1000 mL.

14 mL of CPD Solution is taken for 100 mL of Blood.

Citrate phosphate dextrose (CPD) anticoagulant in Blood transfusion has the following Aduantage

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For Blood preservation over ACD Anticoagulant:

- Isotonicity for RBCs
- More physiological PH.
- 15 % less citrate ion
- Improved red cell oxygen transport.

Lesson-6

Criteria to select Blood DoNoR.

1) **Cancer :-** It is a group of diseases involving ab normal cell growth with the potential to invade or speed to other part of the body

2) **Hypertension :-** A condition in which the force of Blood against the artery wall is too high as compared to the normal.

3) **Diabetes :-** It is a metabolic disease. that causes High blood sugar as compared to the Normal.

4) **Hypothyroidism :-** A condition in which thyroid gland does not produce enough thyroid hormone

5) **Hyperthyroidism :-** A condition in which thyroid gland produce excess amount of thyroid Hormone as compared to the normal.

The purpose to select Blood donor :-

- The health and safety of the donor as well as the recipient must be protected.
- The individual with good Health only be accepted as donor of whole Blood and Blood components.
- Staff should suitably be qualified and trained in the donor selection process.
- Good communication must be established Between the staff and the donor and donor should be ensured.

Medical History

- Have undergone ear/body piercing or tattoo in the last six months.
- Have undergone immunisation in the past one month.
- Have consumed Alchol in the past 48 Hours.
- Have common cold, sore throat or cough at the time of Blood donation.
- Have taken any antibiotics or any other medicine in the 48 Hours.
- Have taken antiplatelet drugs such as aspirin in the past 72 Hours.
- Women who are pregnant or breast feeding are not eligible to donate Blood.

Health Criteria

1) Whole Blood volume . 350 ml — 45 Kg
Collected v/s weight . 450 ml — 55 Kg
of donor . Apheresis — 50 Kg.

2) Pulse rate : 60 — 100 beats per min and regular.

3) Temperature : Normal body temperature for an Adult is 94.6°F (37°c).

4) Haemoglobin : Normal less than 12.5 gm/dL.

5) Blood pressure :
Systolic : 90 — 150 mm of Hg
Diastolic : 50 — 100 mm of Hg.

6) Platelets count : It should not be less than 1.5 Lakh per cubic mm.

Lesson → 7

Blood Collection and Storage.

1) **Blood transfusion** — A Blood transfusion is the transfer of Blood or its product from one person (donor) to another person Intravenously.

2) **Blood collection** — The Blood collection or collection of Blood generally involve the removal of Blood. It is also a common term in Blood sampling.

3) **Venipuncture** — It is a method of collection of Blood by the puncture of vein for any therapeutic purpose.

4) **finger stick (pick method)** — It is a method of collection of Blood by pricking the tip of the finger by sterilised lancet

5) **Arterial Method** — The Blood is collection by the puncture of radial artery at the wrist or the femoral artery in the groin.

Methods of Blood Collection

1) Venipuncture method. (for venous blood)
2) finger prick method. (for capillary Blood)
3) Heal stick method. (for capillary Blood)
4) Arterial method. (for arterial Blood).

1) **Venipuncture Method** :- It is a method of collection of Blood by the Puncture of vein for any therapeutic purpose.

⇒ **Materials / Required** :-
- Syringe
- Needle
- Tourniquet
- vacutainer.
- wipes/swabs.
- Gauze
- Bandages.
- Gloves
- Collection tubes.

⇒ **Procedure** :- 1) Positively identify the patient.
2) Check the requision form for requested test.
3) Position the patient on a chair.
4) Wash your hand.
5) The site is cleansed with antiseptic solution.
6) Ask the patient to make a fist.
7) Insert the needle through the skin into the lumen of the vein.
8) The needle should form a 15-30° angle with the arm Surface.
9) Dispose off contaminated materials in a designated container.

⇒ **Precautions** :- 1) wear gloves and Lab. coat or gown while handling Blood collection.
2) Clean up any Blood spills with disinfectant e.g freshly prepared bleach.
3) Be sure to identify the patient correctly.
4) The site to be punctured must be free from

Hematoma and edema.

5) Allow the antiseptic solution to dry before puncturing
6) Never extract the Blood when the patient is standing.

Advantages and Disadvantage of veni Puncture

⇒ Advantages :- 1) More volume of Blood can be Collected.
2) Multiple collection can be done.
3) Less painful.
4) More options with regards to the site selection.

⇒ Disadvantages :- 1) should be performed by a skilled phlebotomist to avoid complication.
2) Lengthy process.
3) The site to be punctured must be free from Hematoma or edema.

Transportation and storage of Blood

The Blood is transported and stored as per the Blood cold chain system. This system is for transporting and storage of Blood and its components so that they are kept at a correct temperature at all times form collection to Administration to the patient.

The equipment of Blood cold chain includes Blood bank, plasma, platelet agitator cum incubators, Blood transport Boxes.

Characteristics of an ideal Blood donor

The selection of Blood donor must be based on regularly reviewed selection criteria without discrimination of any kind including gender race nationality or region.

Types of Blood Donor

1) Voluntary donor :- They donate Blood of their own.
2) Replacement donor :- They are relatives, friends or near and dears of the patient.
3) Autologus Blood donors :- It refers to the patients who donate their own Blood for themselves.
4) Platelet donors :- They donate Blood components through the process of Blood cell separation.

Screening of Blood donor

The screening criteria of Blood donor is essential to make Blood supply safe.

⇒ To protect donor :- It ensures the safety of the donor to donate the Blood.

⇒ To protect the recipient :- It ensures the protection of recipient with

respect to transfusion — transmitted infection or any other adverse effect.

Method of Transfusion

1) Wash the hands well with soap and water and wear sterile gloves.
2) Make the donor lie down comfortably.
3) Place a bag on a balance.
4) Ask the donar to open and close the fist during collection of Blood.
5) Do not leave the donor unattended during the entire procedure which takes 8–10 min.
6) Provide light refreshment to the donor.

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4/12/22