Blood Groups

Chapter 21

INTRODUCTION

When blood from two individuals is mixed, sometimes clumping (agglutination) of RBCs occurs. This clumping is because of the immunological reactions. But, why clumping occurs in some cases and not in other cases remained a mystery until the discovery of blood groups by the Austrian Scientist **Karl Landsteiner,** in 1901. He was honored with Nobel Prize in 1930 for this discovery.

ABO BLOOD GROUPS

Determination of ABO blood groups depends upon the immunological reaction between antigen and antibody. Landsteiner found two antigens on the surface of RBCs and named them as A antigen and B antigen. These antigens are also called agglutinogens because of their capacity to cause agglutination of RBCs. He noticed the corresponding antibodies or agglutinins in the plasma and named them anti-A or α-antibody and anti-B or β-antibody. However, a particular agglutinogen and the corresponding agglutinin cannot be present together. If present, it causes clumping of the blood. Based on this, Karl Landsteiner classified the blood groups. Later it became the 'Landsteiner Law' for grouping the blood.

LANDSTEINER LAW

Landsteiner law states that:

1. If a particular **agglutinogen** (antigen) is present in the RBCs, corresponding **agglutinin** (antibody) must be absent in the serum.
2. If a particular agglutinogen is absent in the RBCs, the corresponding agglutinin must be present in the serum.

Though the second part of Landsteiner law is a fact, it is not applicable to Rh factor.

■ BLOOD GROUP SYSTEMS

More than 20 genetically determined blood group systems are known today. But, Landsteiner discovered two blood group systems called the ABO system and the Rh system. These two blood group systems are the most important ones that are determined before blood transfusions.

■ ABO SYSTEM

Based on the presence or absence of antigen A and antigen B, blood is divided into four groups:

1. 'A' group
2. 'B' group
3. 'AB' group
4. 'O' group.

Blood having antigen A belongs to 'A' group. This blood has β-antibody in the serum. Blood with antigen B and α-antibody belongs to 'B' group. If both the antigens are present, blood group is called 'AB' group and serum of this group does not contain any antibody. If both antigens are absent, the blood group is called 'O' group and both α and β antibodies are present in the serum. Antigens and antibodies present in different groups of ABO system are given in Table 21.1. Percentage of people among Asian and European population belonging to different blood group is given in Table 21.2.

'A' group has two subgroups namely 'A_1' and 'A_2'. Similarly 'AB' group has two subgroups namely 'A_1B' and 'A_2B'.

■ DETERMINATION OF ABO GROUP

Determination of the ABO group is also called blood grouping, blood typing or blood matching.

Principle of Blood Typing – Agglutination

Blood typing is done on the basis of agglutination. Agglutination means the collection of separate particles like RBCs into clumps or masses. Agglutination occurs if an antigen is mixed with its corresponding antibody which is called **isoagglutinin.** Agglutination occurs when

TABLE 21.1: Antigen and antibody present in ABO blood groups

Group	Antigen in RBC	Antibody in serum
A	A	Anti-B (β)
B	B	Anti-A (α)
AB	A and B	No antibody
O	No antigen	Anti-A and Anti-B

TABLE 21.2: Percentage of people having different blood groups

Population	A	B	AB	O
Indians	23	33	7	37
Asians	25	25	5	45
Europeans	42	9	3	46

A antigen is mixed with anti-A or when B antigen is mixed with anti-B.

Requisites for Blood Typing

To determine the blood group of a person, a suspension of his RBC and testing antisera are required. Suspension of RBC is prepared by mixing blood drops with isotonic saline (0.9%).

Test sera are:

1. Antiserum A, containing anti-A or α-antibody.
2. Antiserum B, containing anti-B or β-antibody.

Procedure

1. One drop of antiserum A is placed on one end of a glass slide (or a tile) and one drop of antiserum B on the other end.
2. One drop of RBC suspension is mixed with each antiserum. The slide is slightly rocked for 2 minutes. The presence or absence of agglutination is observed by naked eyes and if necessary, it is confirmed by using microscope.
3. Presence of agglutination is confirmed by the presence of thick masses (clumping) of RBCs
4. Absence of agglutination is confirmed by clear mixture with dispersed RBCs.

Results

1. *If agglutination occurs with antiserum A:* The anti-serum A contains α-antibody. The agglutination occurs if the RBC contains A antigen. So, the blood group is A (Fig. 21.1).
2. *If agglutination occurs with antiserum B:* The anti-serum B contains β-antibody. The agglutination occurs if the RBC contains B antigen. So, the blood group is B.
3. *If agglutination occurs with both antisera A and B:* The RBC contains both A and B antigens to cause agglutination. And, the blood group is AB.
4. If agglutination does not occur either with antiserum A or antiserum B: The agglutination does not occur because RBC does not contain any antigen. The blood group is O.

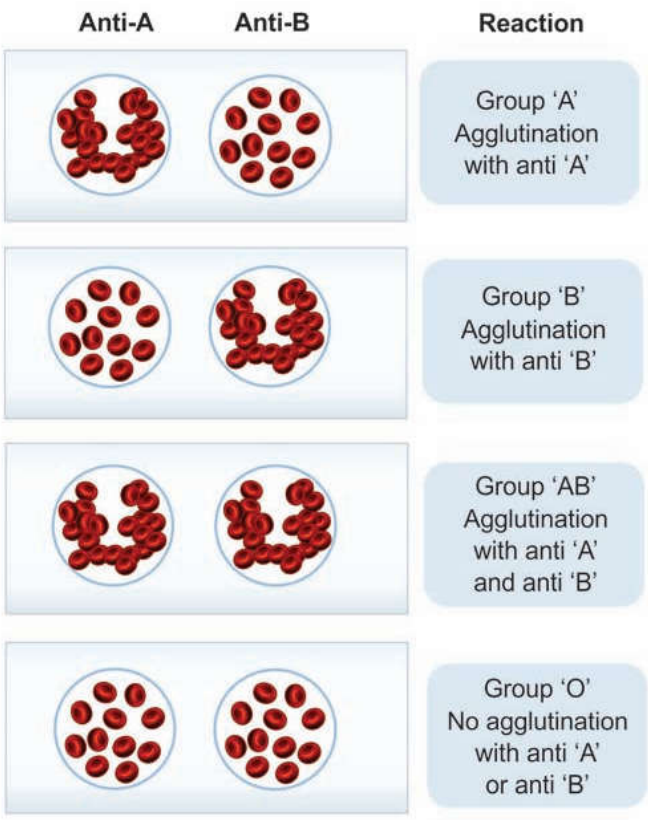

FIGURE 21.1: Determination of blood group

IMPORTANCE OF ABO GROUPS IN BLOOD TRANSFUSION

During blood transfusion, only compatible blood must be used. The one who gives blood is called the **'donor'** and the one who receives the blood is called **'recipient'.**

While transfusing the blood, antigen of the donor and the antibody of the recipient are considered. The antibody of the donor and antigen of the recipient are ignored mostly.

Thus, RBC of 'O' group has no antigen and so agglutination does not occur with any other group of blood. So, 'O' group blood can be given to any blood group persons and the people with this blood group are called **'universal donors'.**

Plasma of AB group blood has no antibody. This does not cause agglutination of RBC from any other group of blood. People with AB group can receive blood from any blood group persons. So, people with this blood group are called **'universal recipients'.**

MATCHING AND CROSS-MATCHING

Blood matching (typing) is a laboratory test done to determine the blood group of a person. When the person needs blood transfusion, another test called cross-matching is done after the blood is typed. It is done to find out whether the person's body will accept the donor's blood or not.

For blood matching, RBC of the individual (recipient) and test sera are used. Cross-matching is done by mixing the serum of the recipient and the RBCs of donor. Cross-matching is always done before blood transfusion. If agglutination of RBCs from a donor occurs during cross-matching, the blood from that person is not used for transfusion.

Matching = Recipient's RBC + Test sera.
Cross-matching = Recipient's serum + Donor's RBC.

INHERITANCE OF ABO AGGLUTINOGENS AND AGGLUTININS

Blood group of a person depends upon the two genes inherited from each parent. Gene A and gene B are dominant by themselves and gene O is recessive. Inheritance of blood group is represented schematically as given in Table 21.3.

Agglutinogens appear during the 6th month of fetal life. Concentration at birth is 1/5 of the adult concentration. It rises to the adult level at puberty. Agglutinogens are present not only in RBCs but also present in many organs like salivary glands, pancreas, kidney, liver, lungs, etc. The A and B agglutinogens are inherited from the parents as Mendelian phenotypes.

Agglutinin α or β is not produced during fetal life. It starts appearing only 2 or 3 months after birth. Agglutinin is produced in response to A or B agglutinogens which enter the body through respiratory system or digestive system along with bacteria.

Agglutinins are the gamma-globulins which are mainly IgG and IgM immunoglobulins.

TRANSFUSION REACTIONS DUE TO ABO INCOMPATIBILITY

Transfusion reactions are the adverse reactions in the body, which occur due to transfusion error that involves transfusion of incompatible **(mismatched)** blood. The reactions may be mild causing only fever and hives (skin disorder characterized by itching) or may be severe leading to renal failure, shock and death.

In mismatched transfusion, the transfusion reactions occur between donor's RBC and recipient's plasma. So, if the donor's plasma contains agglutinins against recipient's RBC, agglutination does not occur because these antibodies are diluted in the recipient's blood.

But, if recipient's plasma contains agglutinins against donor's RBCs, the immune system launches a response

TABLE 21.3: Inheritance of ABO group

Gene from parents	Group of offspring	Genotype
A + A A + O	A	AA or AO
B + B B + O	B	BB or BO
A + B O + O	AB O	AB OO

against the new blood cells. Donor RBCs are agglutinated resulting in transfusion reactions.

Severity of Transfusion Reactions

Severity of transfusion reactions varies from mild (fever and chills) to severe (acute kidney failure, shock and death). Severity depends upon the amount of blood transfused, type of reaction and general health of the patient.

Cause for Transfusion Reactions

Transfusion of incompatible blood produces hemolytic reactions. The recipient's antibodies (IgG or IgM) adhere to the donor RBCs, which are agglutinated and destroyed. Large amount of free hemoglobin is liberated into plasma. This leads to transfusion reactions.

Signs and Symptoms of Transfusion Reactions

Non-hemolytic transfusion reaction

Non-hemolytic transfusion reaction develops within a few minutes to hours after the commencement of blood transfusion. Common symptoms are fever, difficulty in breathing and itching.

Hemolytic transfusion reaction

Hemolytic transfusion reaction may be acute or delayed. The acute hemolytic reaction occurs within few minutes of transfusion. It develops because of rapid hemolysis of donor's RBCs. Symptoms include fever, chills, increased heart rate, low blood pressure, shortness of breath, bronchospasm, nausea, vomiting, red urine, chest pain, back pain and rigor. Some patients may develop pulmonary edema and congestive cardiac failure.

Delayed hemolytic reaction occurs from 1 to 5 days after transfusion. The hemolysis of RBCs results in release of large amount of hemoglobin into the plasma. This leads to the following complications.

1. *Jaundice*

Normally, hemoglobin released from destroyed RBC is degraded and bilirubin is formed from it. When the serum bilirubin level increases above 2 mg/dL, jaundice occurs (Chapter 40).

2. *Cardiac Shock*

Simultaneously, hemoglobin released into the plasma increases the viscosity of blood. This increases the workload on the heart leading to **heart failure.** Moreover, toxic substances released from hemolyzed cells reduce the arterial blood pressure and develop circulatory shock (Fig. 21.2).

3. *Renal Shutdown*

Dysfunction of kidneys is called renal shutdown. The toxic substances from hemolyzed cells cause constriction of blood vessels in kidney. In addition, the toxic substances along with free hemoglobin are filtered through glomerular membrane and enter renal tubules. Because of poor rate of reabsorption from renal tubules, all these substances precipitate and obstruct the renal tubule. This suddenly stops the formation of urine (anuria).

If not treated with artificial kidney, the person dies within 10 to 12 days because of jaundice, circulatory shock and more specifically due to renal shutdown and anuria.

■ Rh FACTOR

Rh factor is an antigen present in RBC. This antigen was discovered by Landsteiner and Wiener. It was first discovered in **Rhesus monkey** and hence the name 'Rh factor'. There are many Rh antigens but only the D antigen is more antigenic in human.

The persons having D antigen are called 'Rh positive' and those without D antigen are called 'Rh negative'. Among Indian population, 85% of people are Rh positive and 15% are Rh negative. Percentage of Rh positive people is more among black people.

Rh group system is different from ABO group system because, the antigen D does not have corresponding natural antibody (anti-D). However, if Rh positive blood is transfused to a Rh negative person anti-D is developed in that person. On the other hand, there is no risk of complications if the Rh positive person receives Rh negative blood.

■ INHERITANCE OF Rh ANTIGEN

Rhesus factor is an inherited dominant factor. It may be homozygous Rhesus positive with DD or heterozygous Rhesus positive with Dd (Fig. 21.3). Rhesus negative occurs only with complete absence of D (i.e. with homozygous dd).

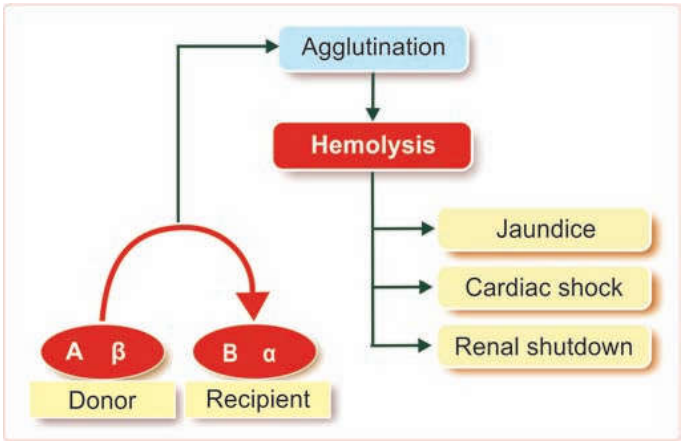

FIGURE 21.2: Complications of mismatched blood transfusion

TRANSFUSION REACTIONS DUE TO Rh INCOMPATIBILITY

When a Rh negative person receives Rh positive blood for the first time, he is not affected much, since the reactions do not occur immediately. But, the Rh antibodies develop within one month. The transfused RBCs, which are still present in the recipient's blood, are agglutinated. These agglutinated cells are lysed by macrophages. So, a delayed transfusion reaction occurs. But, it is usually mild and does not affect the recipient. However, antibodies developed in the recipient remain in the body forever. So, when this person receives Rh positive blood for the second time, the donor RBCs are agglutinated and severe transfusion reactions occur immediately (Fig. 21.4). These reactions are similar to the reactions of ABO incompatibility (see above).

HEMOLYTIC DISEASE OF FETUS AND NEWBORN – ERYTHROBLASTOSIS FETALIS

Hemolytic disease is the disease in fetus and newborn, characterized by abnormal hemolysis of RBCs. It is due to Rh incompatibility, i.e. the difference between the Rh blood group of the mother and baby. Hemolytic disease leads to erythroblastosis fetalis.

Erythroblastosis fetalis is a disorder in fetus, characterized by the presence of erythroblasts in blood. When a mother is Rh negative and fetus is Rh positive (the Rh factor being inherited from the father), usually the first child escapes the complications of Rh incompatibility. This is because the Rh antigen cannot pass from fetal blood into the mother's blood through the **placental barrier.**

However, at the time of parturition (delivery of the child), the Rh antigen from fetal blood may leak into mother's blood because of **placental detachment.** During postpartum period, i.e. within a month after delivery, the mother develops Rh antibody in her blood.

When the mother conceives for the second time and if the fetus happens to be Rh positive again, the Rh antibody from mother's blood crosses placental barrier and enters the fetal blood. Thus, the Rh antigen cannot cross the placental barrier, whereas Rh antibody can cross it.

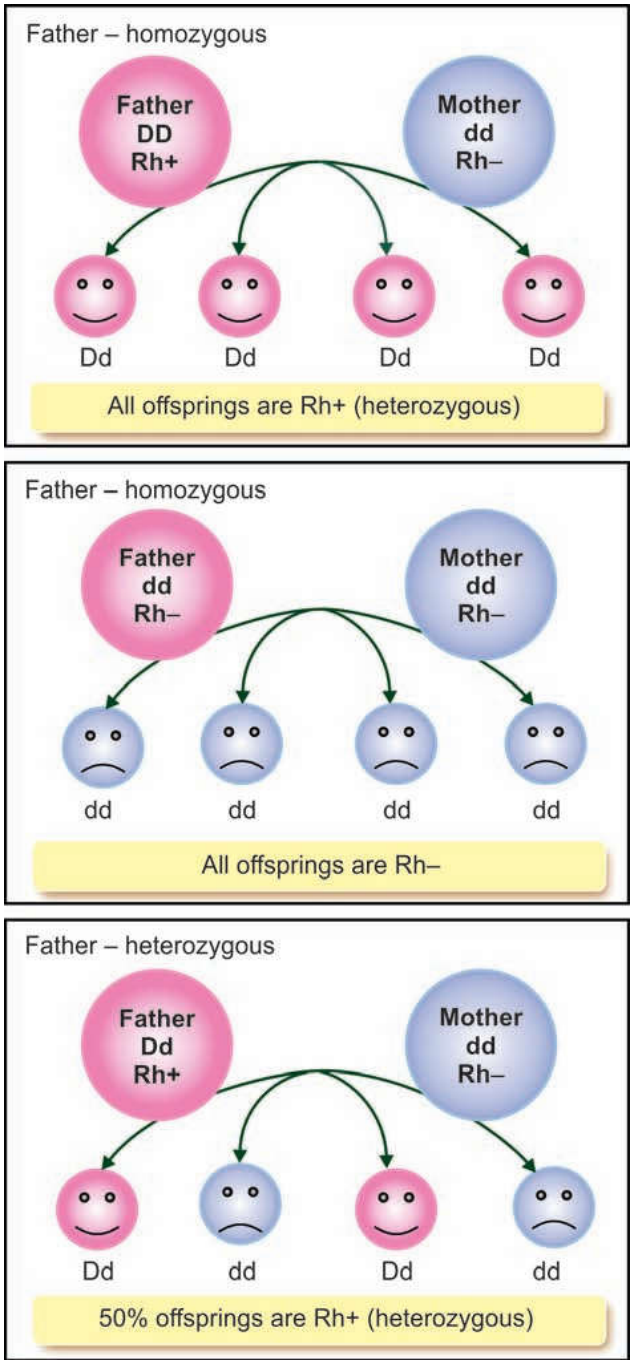

FIGURE 21.3: Inheritance of Rh antigen

Rh antibody which enters the fetus causes agglutination of fetal RBCs resulting in hemolysis.

Severe hemolysis in the fetus causes jaundice. To compensate the hemolysis of more and more number of RBCs, there is rapid production of RBCs, not only from bone marrow, but also from spleen and liver. Now, many large and immature cells in proerythroblastic stage are released into circulation. Because of this, the disease is called erythroblastosis fetalis.

Ultimately due to excessive hemolysis severe complications develop, viz.

1. Severe anemia
2. Hydrops fetalis
3. Kernicterus.

1. *Severe Anemia*

Excessive hemolysis results in anemia and the infant dies when anemia becomes severe.

2. *Hydrops Fetalis*

Hydrops fetails is a serious condition in fetus, characterized by edema. Severe hemolysis results in the development of edema, enlargement of liver and spleen and cardiac failure. When this condition becomes more severe, it may lead to **intrauterine death** of fetus.

3. *Kernicterus*

Kernicterus is the form of **brain damage** in infants caused by severe jaundice. If the baby survives anemia in erythroblastosis fetalis (see above), then kernicterus develops because of high bilirubin content.

The blood-brain barrier is not well developed in infants as in the adults (Chapter 163). So, the bilirubin enters the brain and causes permanent brain damage. Most commonly affected parts of brain are basal ganglia, hippocampus, geniculate bodies, cerebellum and cranial nerve nuclei. The features of this disease are:

i. When brain damage starts, the babies become lethargic and sleepy. They have high-pitched cry, hypotonia and arching of head backwards.
ii. As the disease progresses, they develop hypertonia and opisthotonus (Chapter 155).
iii. Advanced signs of the disease are inability to suckle milk, irritability and crying, bicycling movements, choreoathetosis (Chapter 151), spasticity, (Chapter 34) seizures (Chapter 161), fever and coma.

Prevention or treatment for erythroblastosis fetalis

i. If mother is found to be Rh negative and fetus is Rh positive, anti D (antibody against D antigen) should be administered to the mother at 28th and 34th weeks of gestation, as prophylactic measure. If Rh negative mother delivers Rh positive baby, then anti D should be administered to the mother within 48 hours of delivery. This develops passive immunity and prevents the formation of Rh antibodies in mother's blood. So, the hemolytic disease of newborn does not occur in a subsequent pregnancy.
ii. If the baby is born with erythroblastosis fetalis, the treatment is given by means of exchange transfusion (Chapter 22). Rh negative blood is transfused into the infant, replacing infant's own Rh positive blood. It will now take at least 6 months for the infant's new Rh positive blood to replace the transfused Rh negative blood. By this time, all the molecules of Rh antibody derived from the mother get destroyed.

OTHER BLOOD GROUPS

In addition to ABO blood groups and Rh factor, many more blood group systems were found, such as Lewis blood group and MNS blood groups. However, these systems of blood groups do not have much clinical importance.

LEWIS BLOOD GROUP

Lewis blood group was first found in a subject named Mrs Lewis. The antibody that was found in this lady reacted with the antigens found on RBCs and in body

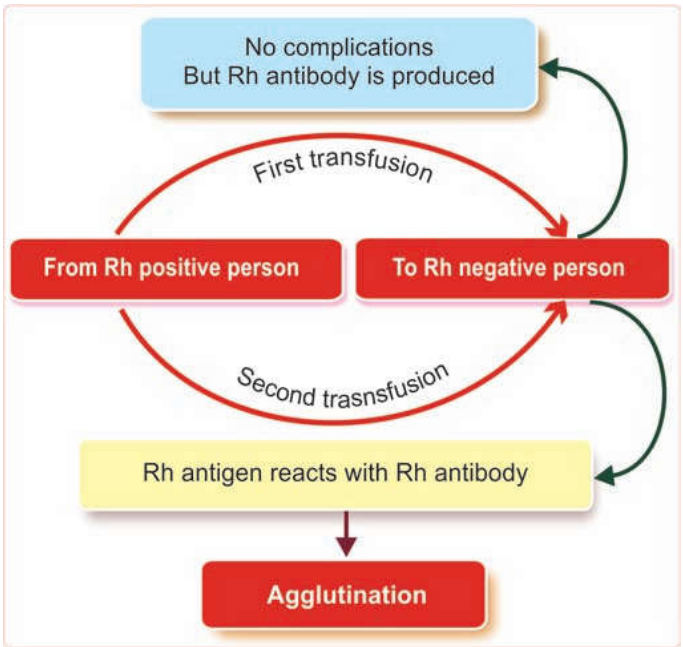

FIGURE 21.4: Rh incompatibility

fluids such as saliva, gastric juice, etc. The antigens, which are named Lewis antigens are formed in the tissues, released in the body secretions and then absorbed by the RBC membrane. Because of secretion along with body secretions, these antigens are also known as secretor antigens. Presence of Lewis antigens in children leads to some complications such as retarded growth. Sometimes, it causes transfusion reactions also.

MNS BLOOD GROUPS

MNS blood groups are determined by their reactions with anti-M, anti-N and anti-S. However, these blood groups rarely cause any trouble like hemolysis following transfusion.

OTHER BLOOD GROUPS

Other blood groups include:

i. Auberger groups
ii. Diego group
iii. Bombay group
iv. Duffy group
v. Lutheran group
vi. P group
vii. Kell group
viii. I group
ix. Kidd group
x. Sulter Xg group.

IMPORTANCE OF KNOWING BLOOD GROUP

Nowadays, knowledge of blood group is very essential medically, socially and judicially. The importance of knowing blood group is:

1. Medically, it is important during blood transfusions and in tissue transplants.
2. Socially, one should know his or her own blood group and become a member of the Blood Donor's Club so that he or she can be approached for blood donation during emergency conditions.
3. It general among the couples, knowledge of blood groups helps to prevent the complications due to Rh incompatibility and save the child from the disorders like erythroblastosis fetalis.
4. Judicially, it is helpful in medico-legal cases to sort out parental disputes.