Blood Chemistry

Constituents of Blood

The chemistry composition of blood is complex . It is an aqueous solution of ions and organic molecules which also have suspended particles . Suspended particles in blood are red blood cells (erythrocytes) , white blood cells (leukocytes) and platelets . Blood without suspended particles is called blood plasma . Blood serum is obtained when fibrinogen , a particular protein is removed from plasma . The major blood constituents are shown in Table 17.1.

a.When blood is drawn and allowed to clot , a clear liquid (serum) licudes from the clotted . Plasma on the other hand separates the cells only when blood is prevented from clotting
b. The blood clot is formed by a protein (fibrinogen) which is present in the soluble form in the plasma and which is transformed to an insoluble network of fibrous material (fibrin , the substance of the blood clot) by the clotting mechanism
c. The change of fibrinogen into fibrin is caused by thrombin , which in fluid blood looks as prothrombin . The conversion of prothrombin depends on the action of thromboplastin and calcium .

Anticoagulants

Clot formation may be prevented by a number of substances as well by vitamin K deficiency . Dicemerol , related to coumarin which comes from clover , inhibits prothrombin synthesis in the liver . It may be used clinically when there is danger of thrombosis by reducing clotting tendency .

Heparin Heparin

A sulphated polysaccharide which inhibits the formation of thrombin from prothrombin , is the most satisfactory

Table 17.1 : Blood Constituents

Compounds	Normal Values	Possible Pathology
Haemoglobin	14-16 g/100 ml whole blood	High in polycythaemia. Low in anaemias.
Non–protein nitrogen	25-25 mgm/100 ml whole blood	High in nephritis and Addison's disease
Uric acid	3-5 mgm/100 ml whole blood	High in gout
Total plasma proteins	6-5-8.2g/100 ml plasma	Low in nephrotic syndrome and malnutrition
Cholesterol	150-300 mgm/100 ml plasma or serum	High in nephrotic Syndrome and hypothyroidism. Low in pernicious anaemia and liver disease
Glucose	80-110 mgm/100 ml whole blood	High in diabetes. Low in Addison's disease
CO_2 combining power	53-80 ml. CO_2/100 ml plasma	Low in acidosis, uncontrolled-led diabetes and nephritis.
Inorganic phosphates	3-4 mgm/100 ml plasma or serum	High in renal rickets and nephritis. Low in infantile rickets
Chloride	570-620 mgm (as NaCl)per 100 ml plasma or serum	High in nephritis. Low in fever or pneumonia

Anticoagulant , since it produced no change in the composition of the blood . However , oxalate and citrate have most widely used as they are cheaper . Use of more of these salts may bring appreciable changes in the distribution of water between the cells and plasma .

Potassium Oxalate

It has been most commonly used since it is more soluble . It acts by precipitating calcium ions as calcium oxalate .

Sodium citrate

Citrate dose not precipitate calcium but converts it to non-ionized form . Citrated plasma is not as satisfactory as serum for calcium estimation . Ethylenediamine tetra acetic acid (EDTA) and its salts act by chelating calcium ions .

Sodium fluoride

It also acts as an anticoagulant but large amounts are required . For blood glucose estimation a mixture of sodium fluoride and potassium oxalate is used as fluoride acts as a preservative by inhibiting glycolytic enzymes .

Functions of Blood

1. Carrying the products of digestion from the small intestine to various organs and tissues
2. Accepting oxygen in the lungs and releasing it in the tissues.
3. Carrying CO_2 from the tissues for elimination by the lungs
4. Removal of waste products from tissues for excretion by the kidneys.
5. Maintenance of water balance and temperature control .
6. Syntheses of antibodies for the protection against bacterial infection.
7. Distribution of hormones , vitamins and enzymes to their sites of action .

Oxygen Transport

During respiration , gases CO_2 and O_2 are interchanged between the body and the environment . This process can occur due to the haemoglobin present in red blood cell (RBC) . The function of the RBC is to carry the inhaled oxygen from

lungs to tissues , where it is utilized for their growth , development and sustenance . Haemoglobin is a globular protein . It consists of four polypeptide chains arranged in a tetrahedral configuration . Haemoglobin contains a non – protein constituent called heme . Heme is an iron porphyrin and is responsible for the red colour of blood . Each of the polypeptide chains of haemoglobin is associated with one heme unit . Haemoglobin combines with oxygen in the lungs (where oxygen is present in higher concentration).Such a combination results in the formation of oxyhaemoglobin . Myoglobin stores oxygen in the muscle tissues . In contrast to haemoglobin it consists of only a single polypeptides chain and is associated with one heme unit .

Formation of CO_2 and its Distribution in Blood

CO_2 is formed in large amounts in the body as the end product of normal metabolism . In tissues about 200 ml of CO_2 is formed perminute at rest and 4 litres at maximal exercise . CO_2 enters the blood stream and from there reaches the lungs .The chief acid present in the blood is CO_2 .CO_2 dissolves in water to from H_2CO3 which on dissociation yields H^+ and HCO_3^- .

$$H_2O + CO_2 \rightleftharpoons H_2CO_3 \rightleftharpoons H^+ + HCO^-_3$$

The only alkali present as such in blood is bicarbonateHCO_3 . It dissociates in water as follows :

$$Na\ HCO_3 + H_2O \longrightarrow H_2CO_3 + NaOH$$

Plasma Proteins

Plasma proteins constitute almost 70 per cent of the plasma and are usually divided into 3 groups : albumin , globulins and fibrinogen . Approximately 55 per cent of the plasma protein is albumin , 38.5 per cent globulins and 6.5 per cent is fibrinogen . Albumin , like other plasma proteins , cannot pass through the walls of the blood vessels (because they are colloids and colloids cannot pass through membranes) . Since albumin is principal plasma protein and the smallest plasma protein both in size and weight (it consists of a single chain of

610 amino acids) , it accounts for most of the colloid osmotic pressure of the blood . This colloid osmotic pressure is caused by the small amounts of plasma that pass through the capillary membranes and tend to accumulate as the venous end of the capillaries . If the plasma protein (primarily albumin) are present in decreased (as during a low protein diet or in nephritis) the osmotic pressure of the plasma decreases . This decreases osmotic pressure of the blood cause a greater net pressure outward at the arterial end of the capillary and a lower net inward venous pressure at the venous end of the capillary . When this occurs , water (fluid) accumulates in the tissues . Such a condition is known as an Oedema .

Kwashiorkor

A severs protein deficiency disease , is characterized by edema of the abdomen and extremities . In children a swollen belly is characteristic . Kwashiorkor is caused by a drop in plasma protein particularly albumin . Under these conditions , water moves from the blood stream into the tissues causing swelling . Oedema can also occur because of heart disease , whereby there is an increase in venous hydrostatic pressure . Many terminal illnesses cause oedema . This becomes a serious problem and tapping and draining may be necessary . Concentrated albumin infusions (25 g in 100 ml diluent) are helpful in the treatment of shock , to increase the blood volume , and to remove fluid from the tissues . The amount of albumin present in the blood is lowered in liver disease because albumin is formed in the liver . Another function of albumin in the blood is to act as a carrier for fatty acids , trace elements and many drugs .

Globulins

The globulins present in the plasma can be separated into different groups by a process known as electrophoresis whereby charged protein particles migrate at varying rates to electrodes of opposite charge , with albumin migrating the

fastest . The distribution of the plasma proteins during electrophoresis. The globulins from complexes (loose combination) with such substances as carbohydrates (mucoprotein and glycoprotein) , lipids (lipoprotein) and metal ions (transferring for Fe and ceruloplasmin for Cu) . These complexes can be transported to all parts of the body . The Y- globulins (immunoglobulins) include the antibodies with which the body fights infectious diseases . Y – globulin has been found to contain as many as 20 different antibodies for immunity against such diseases as measles , infections hepatitis , poliomyelitis , mumps and influenza . Some people lack the ability to make Y – globulin . These people are quite susceptible to infections because they have no antibodies to counteract such diseases . The lack of Y- globulin is called agmmaglobinemia and can be combated by the administration of Y- globulin .

Blood Buffers

The normal pH range of the blood is 7.35 to 7.45 . When the pH falls below this range , the condition is called acidosis . Alkalosis occurs when the pH rises above its normal value . Acidosis is more common than alkalosis because many of the metabolic products produced during digestion are acidic .

Maintenance of Acid – Base Balance (pH)

The mechanisms operating in the body to accomplish the maintenance of acid – base balance are :

1. Buffer systems of the blood
2. Respiratory mechanisms
3. Renal mechanisms

a. Excretion of excess acid or base

b. Formation and excretion of ammonia

Blood Buffers

The blood maintains its pH between 7.35 and 7.45 because of buffers . These buffers are present both in the plasma and in the RBCs . Those in the plasma are primarily sodium buffers ; those in blood cells are mainly potassium buffers . Buffers are

substances , usually a mixture of a weak acid and a salt of a weak acid that resist change in pH. The blood buffers consist of:

a. Bicarbonate buffers

b. Phosphate buffers

c. Protein buffers (including hemoglobin and oxyhemoglobin).

Bicarbonate Buffers

The bicarbonate buffers system in the red blood cells consists of carbonic acid (H_2CO_3) and potassium bicarbonate ($KHCO_3$). The bicarbonate buffer system in the blood plasma consists of carbonic acid and N_2HCO_3. If a strong acid (such as HCl) is added to a sample of blood, it will react with the salt part of the buffer and undergo the following reactions.

$HCl + KHCO_3 \longrightarrow H_2CO_3 + KCl$ (in blood cells)

$HCl + NaHCO_3 \longrightarrow H_2CO_3 + NaCl$ (in blood plasma)

The carbonic acid (H_2CO_3) produced is part of the original buffer.

Note that the strong acid HCl, has been replaced by a very salts and will not affect the pH of the system.

If a strong base like KOH or NaOH is added to a sample blood, the following reactions will occur with the bicarbonate buffer system:

$KOH + H_2CO_3 \longrightarrow KHCO_3 + H_2O$(in blood cells)

$NaOH + H_2CO_3 \longrightarrow NaHCO_3 + H_2O$ (in blood plasma)

The salts $KHCO_3$ and $NaHCO_3$ are part of the original buffer system and the water produced is neutral, so the pH again is not affected. In both cases (reaction with a strong acid or a strong base) more of the buffer is produced plus a neutral compound. The bicarbonate buffers control the pH of blood and the phosphate buffers have an important role inside the cell and the urine.

Phosphate Buffers

The phosphate buffers consist of mixytures of K_2HPO_4 and KH_2PO_4 (also $NaHPO_4$ and NaH_2PO_4) which function similarly the bicarbonate buffers in neutralizing excess acid and base.

$$HCl + K_2HPO_4 \longrightarrow KH_2PO_4 + KCl$$

$$KOH + KH_2PO_4 \longrightarrow K_2HPO_4 + H_2O \qquad \text{More buffer}$$

Haemoglobin Buffers

The haemoglobin buffers account for more than half of the buffering action in the blood. These are haemoglobin buffers and oxyhaemo-globin buffers.

$$\left.\begin{matrix} HHb \\ KHb \end{matrix}\right\} \text{Haemoglobin buffer}$$

$$\left.\begin{matrix} HHbO_2 \\ KHbO_2 \end{matrix}\right\} \text{Oxyhaemoglobin buffer}$$

These buffers, as well as other proteins that act as buffers in the blood stream, pick up excess acid or base to help keep the pH of the blood at 7.35 to 7.45.

Respiratory Mechanism

The process of respiration, i.e. the intake of oxygen and removal of CO_2 requires the transport of these substances by blood. Since CO_2 reacts to form HCO^-_3 and oxygen forms oxyhaemoglobin which is more acidic than haemoglobin, the respiratory process is involved in the delicate acid-base balance of the body. Maintenance of a constant pH of 7.35 to 7.45 is required for health. The respiratory centre in the pH of the blood and immediately causes an increase in the rate and depth of breathing until excess CO_2 (and hence excess H^+) is removed. Changes brought about by respiration are rapid.

Renal Mechanisms

This is by far the most effective mechanism, but it is slow, requiring hours to show result. The kidneys excrete more HCO_3^- and HPO_4^-, when the blood pH is too high and more H^+ (which it gains in exchange for Na^+) and H_2PO_4 when pH is too low. Also the kidney can increase its production of NH_3 (ammonia) which will trap H^+ to form NH_4, thus lowering the

acidity of the blood. Since the cell membrane is not permeable to the charged NH_4, it is "trapped" and excreted in the urine as ammonium salts. Ammonia is produced in the epithelial cells largely by the deamidation of glutamine taken up from the arterial blood.

Glutamine + $H_2O \longrightarrow$ Glutamic acid + NH_3

Urine is slightly acidic because of the phosphates and sulphates, formed principally from catabolism of the food, and extracted mainly as acid ions, $H_2PO_4^-$ and HSO_4^-. Also organic acids formed in metabolism are extracted by the kidney if they do not undergo further metabolism.

Measurement of Acid-Base Balance

The definite way of assessing the state of acid-base balance of the body is to determine the pH of the blood. But clinically this is not always possible. As an alternative method, determination of the CO_2 content of the plasma is considered suitable for clinical purpose. Acidosis and Alkalosis – Acid Base Imbalance – Disturbance in H^+ Homeostasis . Changes in the CO_2 level in blood and corresponding change in HCO_3^- AND pH may be caused by respiratory acidoses , respiratory alkalosis, metabolic acidoses and metabolic alkalosis . If the pH is abnormally high or low , the alkalosis or acidosis is said to be uncompensated . Disturbances in the acid – base balance are known as acidosis and alkalosis and these occur mostly due to abnormalities in respiratory system or due to disturbances in metabolism . Accordingly acidosis and alkalosis are classified as :

1. a. Respiratory acidosis
   b. Respiratory alkalosis
2. a. Metabolic acidosis
   b. Metabolic alkalosis

Respiratory acidosis

It is caused by depression of the respiratory centres by drugs , e.g. barbiturate poisoning , narcotics ingestion , in pulmonary disorders (pneumonia) due to mechanical obstruction of air

passage and during breathing of air with high CO_2 content . There will be a decrease in blood HCO_3^- content and increased levels of CO_2 (H_2CO_3) . The condition is compensated by the action of the kidneys .

Respiratory alkalosis

It is relatively uncommon . It is caused by hypoventilation as in high fever , hysteria , high altitudes and salicylate poisoning or due to dry hot weather . It can also occur during anaesthetic procedures with manual control of breathing causing expulsion of CO_2 and in certain diseases of central nervous system affecting the respiratory system .

Metabolic acidosis

In this condition , there is a deficit of plasma bicarbonate , without much change in the carbonic acid content . Metabolic acidosis can occur in the following conditions :

a. Uncontrolled diabetes m complicated with ketosis
b. Vomiting with loss of fluid not containing acid
c. Poisoning by an acid salt
d. Starvation , high fever
e. Violent exercise
f. Lactic acidosis due to hemorrhage
g. Ingestion of acidifying salts like acetyl salicylic acid , phosphoric acid . HCl , NH_4Cl
h. Renal insufficiency : Retention of acids normally produced , e.g. Terminal stages of nephritis , destructive renal lesions , such as polycystic kidneys , pyelonephritis , renal TB. The CO_2 /bicarbonate buffer system in blood reflects changes in all buffer systems during disturbances . When pH falls , the ratio HCO_3^- / CO_3 + H_2CO_3 also decreases . Excess of CO_2 is blown off through lungs that bring back the original ratio and pH. Thus , acidosis is compensated by a respiratory response .

Metabolic alkalosis

In metabolic alkalosis , the bicarbonate content of the plasma is increased without undue changes in the carbonic acid content.Metabolic alkalosis occurs in the following conditions:

a. Ingestion of large doses of alkalies in the treatment of pepticulcers .

b. Excessive vomiting with loss of large amount of gastric juice as in cases of intestinal obstruction .

c. Removal of large amounts of gastric secretion as in gastric suction. There is also loss of chloride . Compensation is attempted by a depression of respiration so that more CO_2 is retained .