# *Erythrocyte Sedimentation Rate (ESR)*

## ***Erythrocyte Sedimentation Rate (ESR):***

It is the rate at which red blood cells sediment in a period of one hour. It is a common hematology test, and is a non-specific measure of inflammation. To perform the test, anti-coagulated blood is traditionally placed in an upright tube, known as a Westergren tube, and the rate at which the red blood cells fall is measured and reported in mm at the end of one hour.

There are ٣ stages in erythrocyte sedimentation:

١. Stage ١ : Rouleaux formation (negatively charged red blood cells)
٢. Stage ٢ : Sedimentation or settling stage
٣. Stage ٣ : Packing stage - ١٠ minutes (sedimentation slows and cells start to pack at the bottom of the tube)

In normal conditions, the red blood cells are negatively charged. Therefore, negatively charged red blood cells repel each other and do not stack over each other. Besides, if the viscosity of blood is high, red blood cells would to the base, thus lowering the ESR.

The rate of erythrocyte sedimentation is affected by both inflammatory and non-inflammatory conditions. In inflammatory conditions, fibrinogen, other clotting proteins, and alpha globulin are positively charged, thus increasing the ESR. In non-inflammatory conditions, plasma albumin concentration, size, shape, and number of red blood cells, and the concentration of immunoglobulin can affect the ESR.

The value of ESR does not change whether dialysis is performed or not. Therefore, ESR is not a reliable measure of inflammation in those with kidney injuries as the ESR value is already elevated.

ESR begins to rise at ٢٤ to ٤٨ hours of inflammation, decreases slowly as inflammation resolves, and can take weeks to months to return to normal levels. For ESR values more than ١٠٠ mm/hour

## ***Clinical significance:***

The physician use ESR test to follow the patient repose for treatment. ESR level is increased in the following cases:

١. Inflammation.
٢. Auto immune disease as Rheumatoid arthritis and cancer.
٣. In women during menstruation and pregnancy
٤. Non-inflammatory conditions that can cause raised ESR include anemia, renal failure, obesity and ageing

ESR level is decreased in the following cases:

١. ESR level is decreased in case of sickle shape red blood cells or spherocytosis.
٢. An increased number of red blood cells (polycythemia) causes reduced ESR as blood viscosity increases.
٣. Leukemia,

٤. Congestive heart failure.

## *Normal Values:*

The normal value of ESR increases in progressive of age and female more than male:

| Female | Male | Age |
|---|---|---|
| ٢٠ mm/hr | ١٥ mm/hr | Younger than ٥٠ year |
| ٣٠ mm/hr | ٢٠ mm/hr | Older than ٥٠ year |
| ٤٢ mm/hr | ٣٠ mm/hr | Older than ٨٥ year |

## *Experiment Principle (Westergren Method):*

The rate of red blood cells sedimentation is measured by Westergen tube. Westergen tube is very easy to use and doesn't need any energy source. It is a straight tube with ٣٠ cm height and ٢,٥٥ mm in diameter and is scaled from (٠-٢٠٠) millimeters.

Nowadays, automated analyzers had been introduced into the clinical laboratory; the ESR test has been automatically performed.

## *Procedure:*

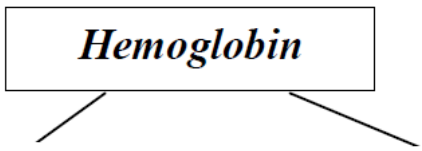
١. Dilute ٢ mL of blood with ٠,٥ mL of ٣,٨٪ sodium citrate or ٠,٥ mL of ٠,٨٥٪ sodium chloride.
٢. Fill westergren tube with the sample till the zero mark (above) and leave it vertically in its special shelf at room temperature and without vibration or direct exposure to light.
٣. After ٦٠ minutes, the distance from the zero mark to the top of the erythrocyte level is read in milliliters. This reading is the value of the ESR.

# *Blood Hemoglobin (Hb)*

Hemoglobin abbreviated Hb, is the iron-containing oxygen-transport metalloprotein in the red blood cells. Hemoglobin in the blood carries oxygen from the lungs or gills to the rest of the body (i.e. the tissues). There it releases the oxygen to permit aerobic respiration to provide energy to power the functions of the organism in metabolism. A healthy individual has ١٢ to ١٦ grams of hemoglobin in every ١٠٠ ml of blood. The iron could be recycled from the old red blood cells.

Each subunit of hemoglobin is a globular protein (globin) with a heme group inside. The globin is a tetramer (which contains four subunit proteins) called hemoglobin A, consisting of two α and two β subunits non-covalently bound, each made of ١٤١ and ١٤٦ amino acid residues, respectively. This is denoted as $\alpha_2\beta_2$. The subunits are structurally similar and about the same size. Each subunit has a molecular weight of about ١٦،٠٠٠ daltons. The four polypeptide chains are bound to each other by salt bridges, hydrogen bonds, and the hydrophobic effect. While heme group consists of an iron (Fe) ion (charged atom) held in a heterocyclic ring, known as a porphyrin. This porphyrin ring consists of four pyrrole molecules cyclically linked together (by methine bridges) with the iron ion bound in the center. The iron ion, which is the site of oxygen binding, coordinates with the four nitrogen atoms in the center of the ring, which all lie in one plane. The iron is bound strongly (covalently) to the globular protein via the N atoms of the imidazole ring of F٨ histidine residue (also known as the proximal histidine) below the porphyrin ring.

When the number of red blood cells or hemoglobin decreases, this condition is known as anemia; it means a decrease in the body's ability to produce healthy red blood cells, or it is damaged very quickly before another cells are replaced. As a result, red blood cells that carry oxygen to tissues are decrease and a state of exhaustion is generated.




### *Heme*

It turns after the iron is separated from it to the yellow pigments that are subtract with yellow, either separation iron used with bone marrow in the formation new hemoglobin molecule and more than the need of the body stored in the liver and spleen

### *Globin*

It is added to the blood proteins and is used in the formation of new hemoglobin molecules

Some symptoms of red blood cells decrease are: paleness of the skin, paleness in the color of the lips, color of the syphilis, weakness of the eyelids soles and the hand grip, accelerating the heartbeat when exerting the least effort, tightness of breath when exerting the slightest effort, pain in the chest.

## ***Normal Values:***

١. Male ١٣-١٧ g / dL
٢. Female ١٢-١٦ g / dL

## ***Common anemia types and their causes include:***

١. ***Iron deficiency anemia:*** The bone marrow needs iron to produce hemoglobin, and the body acquires iron from the food; so anemia may be caused by an iron-deficient diet or by the inability to absorb iron from foods due to intestinal dysfunction.
٢. ***Vitamin deficiency anemia:*** In addition to iron, the body needs folic acid and vitamin B١٢ to produce enough healthy red blood cells. So, if there is lack in these substances in diet then anemia occurs.
٣. ***Anemia caused by chronic diseases:*** Chronic diseases such as AIDS (HIV), cancer, liver fatigue and the last stage of kidney failure may interfere with the production of red blood cells, which leads to a chronic anemia. The kidneys produce a hormone called erythropoietin that stimulates the bone marrow to produce red blood cells so kidney failure is considered as an important cause of anemia.

٤. ***Aplastic anemia:*** Anemia is life threatening and results from a decline in the ability of the bone marrow to produce three types of blood cells, namely red and white blood cells and platelets. A serious infection such as hepatitis, exposure to toxic chemicals or some drugs may lead to aplastic anemia.

٥. ***Hemolytic anemia:*** This type develops as red blood cells are damaged more rapidly than they do by the bone marrow. Some blood diseases may increase red blood cell damage, or autoimmune disease may make the body produce antibodies to red blood cells that damage them early and may lead to hemolytic anemia causing a pale skin and spleen inflation.
٦. ***Sickle cell anemia:*** It is a hereditary and dangerous type of anemia and is caused by a distorted shape of hemoglobin which forces the red blood cells to take an abnormal crescent shape. These abnormal cells die early, causing a chronic lack of red blood cells, and sickle-shaped red blood cells may prevent blood flow through small blood vessels in the body, leading to other symptoms that are often painful.

## ***Procedure:***

There are a number of ways to measure the proportion of hemoglobin, the best and most accurate is that depends on the calculation of the rate of platelet sedimentation in micro centrifuge device.

- Fill a capillary tube containing anticoagulant with blood produced by the prick of finger and then close one end of the capillary with wax dedicated for this purpose.
- This capillary tube is placed against another capillary tube containing distilled water in the micro centrifuge for five minutes.
- Put the capillary tube containing the model (plasma + platelets) on the ruler to calculate platelet sedimentation so that the upper limit of the plasma corresponds to the upper limit of the end of the ruler.

Platelet sedimentation rate is read on the ruler and the following law is applied to calculate hemoglobin.

***Hb (g/L) = platletes- ١/٣***