Hematology

PHYSIOLOGY

☐ Sheet ☐ Slide ☐ Handout

Number: **5**

Subject: **Anemia, polycythemia and RBCs tests**

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Price:

- Some points were written in an order that differs from the record.
- References:
  - ✓ *Guyton And Hall medical physiology.*
  - ✓ *Robbins Basic Pathology.*
- Topics of the lecture:
  - ✓ Anemia.
  - ✓ Polycythemia.
  - ✓ RBCs test.
  - ✓ Introduction to WBCs.

❖Review:

- Normochromic cells >>> MCHC is normal, between 32-36. If lower than 32>> Hypochromic. If higher than 36>>> Hyperchromic.
- Normocytic cells >>> MCV is normal, between 80-90 μ^3. If lower than 80>>> Microcytic. If higher than 90>>>Macrocytic.
- Macrocytic cells are seen in vitamin B12 deficiency, while microcytic hypochromic cells are seen in iron deficiency and when there is failure in the synthesis of the protoporphyrin ring of the hemoglobin.
- Hemoglobin is composed of two parts: protein 96% and heme 4%.

- **Normal RBC count**:
  - 4 millions/ 100 ml in females
  - 5millions/ 100ml in males.
- An RBC count that is lower than the normal range>>>the case is considered anemia. *However, this is not so accurate; it is explained later.*
- An RBC count that is higher than the normal range>>> the case is considered **erythrocytosis or polycythemia.**

❖Anemia:

- Sometimes is called erythrocytopenia.
- Anemia is a group of disorders that is characterized by a deficiency of circulating RBCs.
- ***To be more accurate;***
  → Anemia is defined as a deficiency of hemoglobin in the blood, this can be the result of either too few RBCs or too little hemoglobin in the cells. While Erythorocytopenia means a decreased number of RBCs. Thus anemia is mainly **low hemoglobin** concentration that MAY or MAY NOT be accompanied with a decrease in RBCs. i.e. Anemia is not always associated with a decreased RBCs count, which means that it is **not the same** as erythrocytopenia. Anemia is more a general term than erythrocytopenia
- There are many classifications of anemia, but here we will focus on the pathological types of anemia, either increased peripheral consumption or decreased blood production.

• Types of anemia:

A. **Increased peripheral consumption** (Dr. Saleem calls it increased blood loss)**,** could be caused by**:**
   - **Acute or chronic hemorrhage.**
   - **Hemolysis:** hemolysis can be caused by:
     - A problem in the cell itself, like a deficiency of some enzymes such as G6PD (Glucose-6-phosphate dehydrogenase) or pyruvate kinase.
     - Extra corpuscular causes of hemolysis, i.e. outside the cells, in the plasma for example: which include blood group incompatibility, venoms, drugs, few penicillin, toxins and infections like malaria.

B. **Decreased blood production** could be caused by:

1. inadequate dietary intake of some essential substances for erythropoiesis such as deficiency in vitamin B12, folic acid, pyridoxine(vitamin B6), ascorbic acid or proteins.
2. Problem in the bone marrow, as the bone marrow is the place where the whole process takes place.

- Note: all vitamins are required for erythropoiesis; however, vitamin B12 and folic acid are the most important. Dr Saleem says that vitamin B12 is important for every process in the body.

Remember that vitamin B6 works as a coenzyme in the first step in the synthesis of the heme portion of hemoglobin, the condensation of glycine with succinyl COA.

• Effects of anemia on the circulation:

- Viscosity of the blood depends largely on the blood concentration of RBCs. In cases of severe anemia the RBCs concentration is decreased >>> the blood viscosity may fall to as low as 1.5 times that of the water's viscosity rather than the normal value of about 3. Therefore resistance to blood flow in peripheral blood vessels decreases >> more than normal amounts of blood flow through the tissues and returns to the heart, increasing cardiac output.
- Moreover, Hypoxia leads to vasodilatation of peripheral tissue blood vessels>> causing further increase in the amount of blood returned to the heart and increasing the cardiac output.

→Thus one of the major effects of anemia is increased cardiac output as well as increased pumping workload on the heart.

- ***Extra note from Guyton:*** when an anemic person starts to exercise, the heart is not capable to pump greater quantities of blood than what it is

already pumping>>> hypoxia of the tissues results and acute heart failure may ensue.

❖Polycythemia or erythrocytosis:

- **Polycythemia** is defined as an increase in the concentration of the RBCs in the circulation in which hemoglobin may reach up to 20mg/100 ml of blood.
- Patients with polycythemia, for unknown reason, are advised to donate blo od routinely.

- Dr. Saleem says that *usually erythrocytosis is used to describe an increase in the RBCs that occurs for a short period of time and then RBCs count is back to normal. On the other hand polycythemia usually describes an increase in the RBCs count that continues for a long period indicating an underlying problem*. **However, polycythemia is the same as erythrocytosis according to *Robbins Basic Pathology*.**

• Classification of polycythemia or erythrocytosis:

1. **Relative polycythemia:**
   Results from dehydration, in which the plasma decreases so the total volume of blood decreases, while the RBCs concentration is still the same >>>as a result the RBCs concentration increases ***RELATIVELY.***

2. **True polycythemia** :

   →It is further divided into:

A. **Primary polycythemia**: results from an increased proliferation of the erythrocyte progenitors (*think of malignancy* in such case).
B. **Secondary polycythemia**: that results from elevated levels of erythropoietin. Erythropoietin may increase in cases such as hypoxia resulting in *physiological polycythemia*.
    - **Note**: Some drugs such as excessive amounts of androgens or thyroxin and cobalt (experimentally used) may lead to polycythemia.

→Note that some cases of polycythemia are associated with increased level of erythropoietin while others are not. *Dr. Saleem focused on this point.*

• Effects of polycythemia on the circulation:

1. Viscosity is greatly increased >>>the blood flow through the peripheral blood vessels is often **very sluggish**>>> the rate of the venous return to the heart is decreased.
   On the other hand the blood volume is greatly increased (from 5 liters up to 9) which tends to increase the venous return to the heart. Thus the cardiac output in polycythemia is not far from normal because these two factors more or less neutralize each other.
   Dr. Saleem says that the viscosity, resistance and venous retain are all increased >>> increasing the load on the heart. So in both cases, anemia and polycythemia, the heart work are increased and this may lead to heart failure.

2. **Decrease in the oxygenated RBCs**: The blood flow in the peripheral tissues is sluggish and the amount of the blood is increased>>> so more blood pass through the capillaries before entering the venous plexus as well as it passes slower than normal >>>so more hemoglobin will be deoxygenated.
   →*Extra note: polycythemia vera is a pathological polycythemia caused by a genetic disorder that results in excess production of RBCs. People with disease usually have ruddy complexion with a bluish tint skin due to an increase in the deoxygenated RBCs.*

❖ Diagnostic tests of the RBC's:

→In hematology many tests are done for experimental or diagnostic purposes. We will discuss two tests; Erythrocyte sedimentation rate and osmotic fragility tests.

➢ Erythrocyte sedimentation rate:

- General test that measures the rate at which the RBCs sediment in a tube after a specific time and it is used to help in the diagnosis of some diseases. However, it is not a **specific test**; that is why it is not performed alone to give a definitive diagnosis.
- **the principle of the test:**

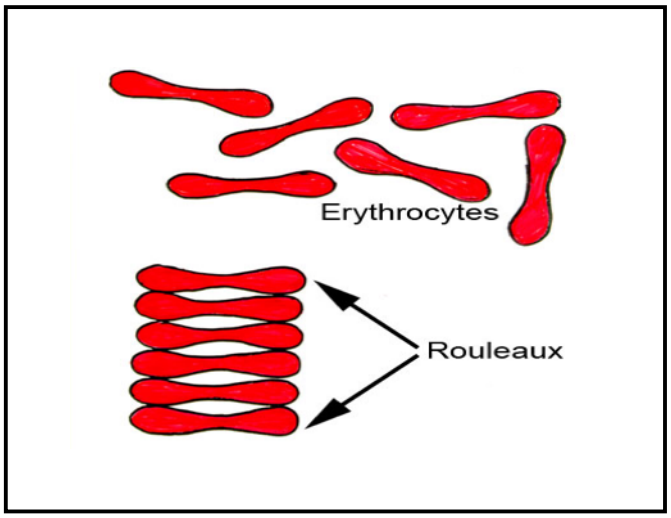
  →We place a fresh anti-coagulated blood sample in an upright tube >>> allow the RBCs to settle in the lab for one hour at room temperature >>> we measure the clear plasma layer>>> the rate at which the RBCs have sediment as mm/hr, will be known.
- There are many methods used to measure the ESR including:
  1. Wintrobe's method.
  2. Westergren's method.
  3. Landau method.
  4. Cultur method.

- Note: Wintrobe's method and Westergren's method are the main methods used. Normal values differ among different methods; in Westrgren's for

example numbers are much higher than in Wintrobe's method, while in cultur method number are less.

- Normal values of the plasma height in Wintrobe's method:
  - In males it's up to 5 ml
  - In females it's up to 15 ml.

→More than these numbers indicates an abnormality.



- RBCs accumulate on each other forming what is called **rouleaux,** ***shown in the figure.***

- **Factors affecting ESR**:

1. **Erythrocyte size:** for example microcytic cells will settle faster than macrocytic cells.
2. **Shape of erythrocyte:** any change in the shape will affect ESR. Abnormally shaped cells, such as in sickle cell anemia or spherocytic anemia, fail to form rouleaux, and the ESR in such cases is decreased.
3. **Count of RBC's**: in severe anemia, where the RBCs count is decreased, ESR is elevated. While in polycythemia, ESR is either normal or low.
4. **Proteins of the plasma**: mainly fibrinogen and globulins as they play a role in viscosity of the blood.(they increase ESR)
5. **Mechanical and technical factors**: for example, the tube must be 100% vertical and if it is slightly tilt (only 3 degrees), this can cause a drop by 30%. ESR also can be affected by temperature.

- **ESR increases in:**

1. **Infections** whether acute or chronic such as hepatitis and tuberculosis.
2. All **connective tissue destructive** diseases such as rheumatoid arthritis.

→In other words, ESR is elevated when there is **inflammation.**

→Thus an elevated ESR indicates a disease without specifying it.

That's why the ESR is done in accompanies with many other tests in order to be able to give a definitive diagnosis for a case. **ESR is a NON- specific test.**

➢ Osmotic fragility test:

- Specific test to some extent.
- It is used to see how fragile the RBCs are, meaning how easily they break.
- ***Remember:*** The normal isotonic solution (physiological) is 0.9% NaCl. Below that is hypotonic solution and higher is hypertonic solution. When RBCs are placed in hypotonic solutions, this will burst (osmotic lysis). In contrast, when they are placed in hypertonic solutions they will shrink.

- The test is performed by adding fresh blood samples, in equal amounts, into solutions with different salt concentrations (From 0.1 to 0.8 of NaCl, i.e. hypotonic solutions) >>> they are left for few hours to allow hemolysis to occur.

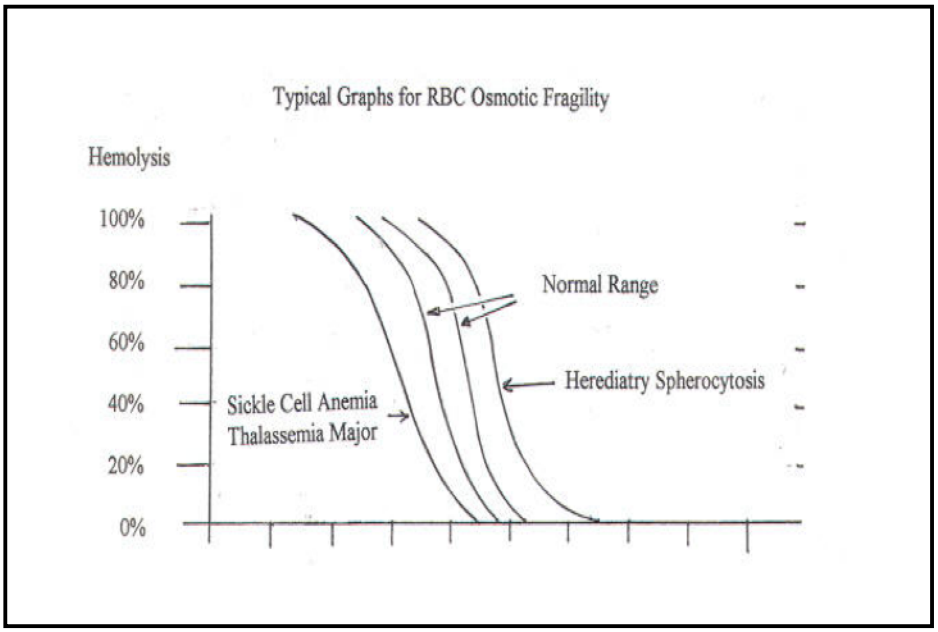
- Normal blood cells can stay intact at lower salt concentrations than fragile blood cells. Normally, blood begins to lyse at concentration of 0.5 or 0.55 and lysis is completed at concentration of 0.3.
- If RBCS hemolysis begins at a lower than 0.5 then the osmotic fragility **test** is increased(the cells have decreased osmatic fragility). On the other hand if the hemolysis begins at a higher concentration than

normal, for example at concentration of 0.6 or 0.65, then the RBCs is said to be fragile and the **test** result is decreased(fragility increased).

- **The results:**

Hereditary spherocytosis: is an autosomal dominant abnormality of erythrocytes. Where they are sphere-shaped (spherocytosis) rather than the normal biconcave disk shaped due to abnormal membrane proteins.

1. The fragility is **decreased in the following cases(i.e**. lysis begins at concentration lower than normal**)**:
   - Iron deficiency anemia
   - Thalassemia
   - Sickle cell anemia.
2. The fragility is **increased in the following cases (i.e.** lysis begins at concentration higher than normal**):**
   - **H**ereditary spherocytosis.
   - **H**emolytic anemia.



- Note that most diseases linked to abnormal fragility test result are associated with abnormalities in the membrane of the RBCs.
- Sometimes the blood samples are left for 24 hours in the solution to get the exact results.

- **Factors affecting osmotic fragility test:**

- Shape of the RBCs mainly.
- Then the volume.
- Surface area.
- The functional state of the RBCs.

❖ Introduction to WBCs:

- **There are 5 types:**
  - Granulocytes: Neutrophils, Eosinophils, Basophils.
  - Lymphocytes.
  - Monocytes.

- **General features:**

  - Nucleated cells.
  - Don't contain hemoglobin.
  - Larger than RBCs.
  - Active cells, moving from blood to the tissues and vice versa.

- Much less in number (normal range is from 5,000 to 10,000). Below this range is called **leukopenia** and above the range is called **leukocytosis.**
- Half life ranges from hours, days, months & up to years.
- Note that there is no difference in the WBCs count between sexes. However the count differs according to other factors such as :
    1. **Time;** the count is at the maximum level in the **afternoon** and the least in the morning. *DR. Saleem said that the maximum level is reached in the evening and this diurnal variation is due to the difference in the activity*.
    2. **After meals, exercise and excitement:** Increases.
    3. **Pregnancy:** Increases.
    4. **Infections:** Increases.

Sorry for any mistakes,
Wish you all best of luck.
Special thanks to Joud Al-Majali and Tala Rawashdeh~