Hematology

- Histology
- Biochemistry
- Pathology
- Pharmacology
- **Physiology**
- Microbiology

- Handout
- Slide
- Sheet: **3**

- Dr. name :
  Dr Salim Khresha
- lecture number :
  3
- Done BY :
  Dania Wreikat Al-Adwan

By: Amer ABU Shmais

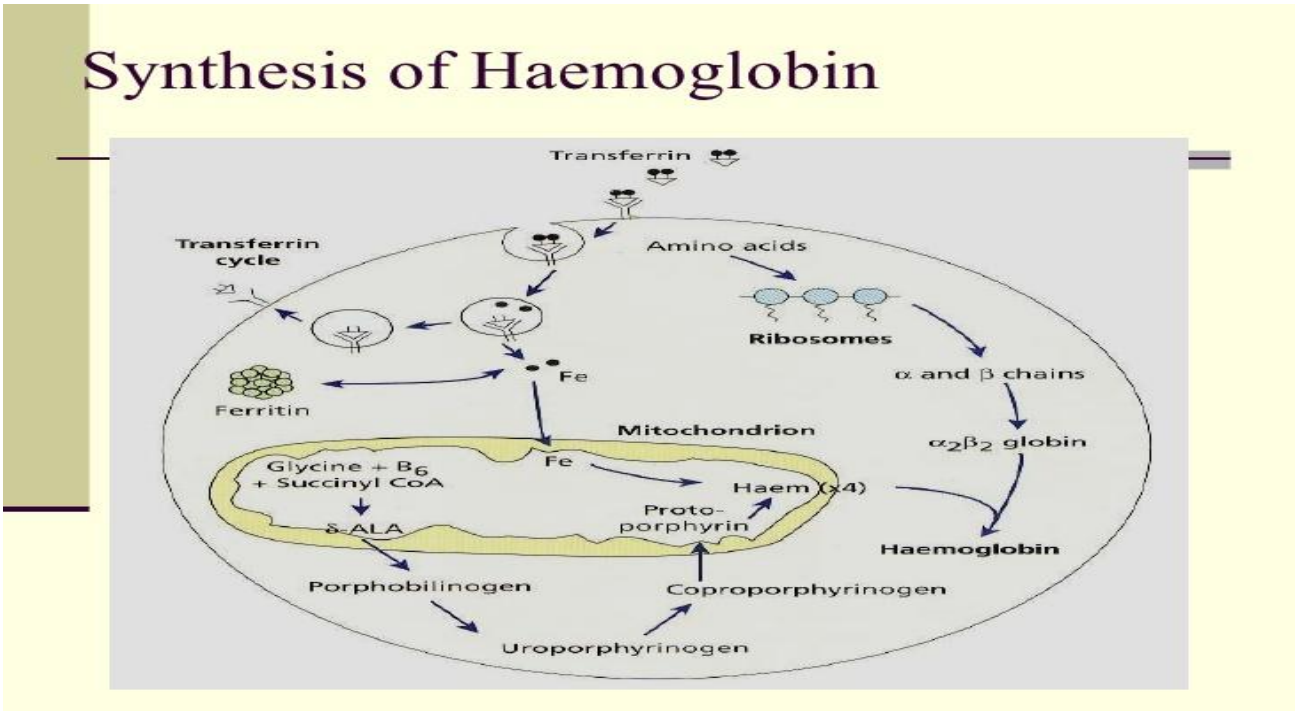
بِسْمِ ٱللَّهِ ٱلرَّحْمَٰنِ ٱلرَّحِيمِ

HEMATOLOGY & BLOOD SYSTEM

Recall:

▪ Hemoglobin synthesis:

Hemoglobin synthesis occurs in all developing RBCs, heme part synthesis occurs in the mitochondria and globin part on the ribosomes. Heme part begins by the combination of glycine and succinyl Co-A under the effect of an enzyme called aminolevulinic acid synthase (ALAS) producing delta aminolevulinic acid (ALA) –an intermediate in the synthesis. So this enzyme is essential for this reaction. For this reaction also we need vitamin B6, which is stimulated by erythropoietin and inhibited by the heme. Then finally, protoporphyrin is formed and binds with ferrous forming heme which then binds to the globin part (the 4 subunits) forming hemoglobin.



- Then,we talked about 6 Types of hemoglobins :
  - 3 of them found only in the fetus ⟹ (Protland ,Gower 1, Gower2)
  - And the other 3 types found in the newborn & in the adult ⟹

  (Adult hemoglobin (A) , Hemoglobin (A2) , Fetal hemoglobin (F))
- There are difference between them in the molecular structure & percentages
- The doctor focus in the percentages of hemoglobin's types :

	Newborn	Adult
Adult hemoglobin (A)	**20 %**	**97%**
Hemoglobin (A2)	**0.5%**	**2.5 %**
Fetal hemoglobin (F)	**80%**	**<1 %**

- **You also remember there are other 2 globins :**
  - **Myoglobin ⟹ in the red muscle or in muscle .**
  - **Neuroglobin ⟹ in nervous system .**

- The fetal hemoglobin is gradually replaced by adult hemoglobin, 6 months after birth, it is totally replaced by adult hemoglobin.

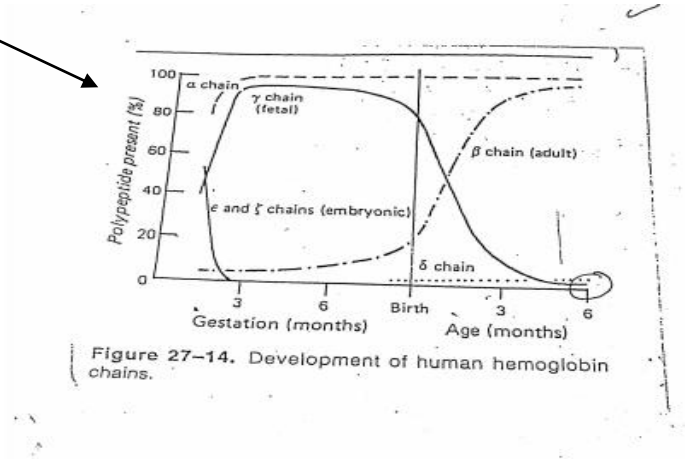
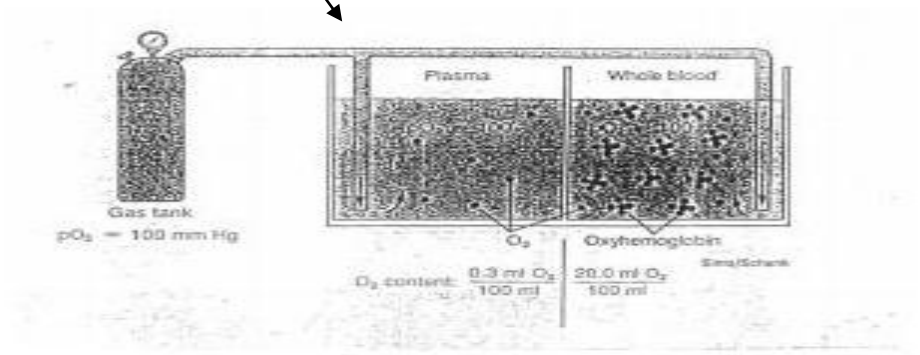


Figure 27–14. Development of human hemoglobin chains.

- 1g of hemoglobin carries 1.34 ml oxygen. We have 20 ml oxygen/100 ml of the whole blood, and 0.3 ml oxygen/100 ml plasma. So the capacity of hemoglobin in the blood to carry oxygen is higher than the capacity of plasma (mostly water) which almost doesn't carry any oxygen.



- **Factors that regulate erthropoiesis :**

1. Oxygen supply
2. Vitamins especially B12 , folic acid
3. Iron
4. Proteins (Note :hemoglobin is a protein that has 96% proteins and 4% Heme

5. Trace element (copper ,cobalt)
   (cobalt binds to vitamin B12)
6. Health bone marrow
7. Liver: is a multifunctional organ (Storage ,protein synthesis ,hormone synthesis), and it produces 10% of erythropoietin.
8. Hormones (erythropoietin ,androgens,thyroid hormone ,growth hormone and corticosteroid hormones)

❖ Let's begin :

This figure called **Hemoglobin-oxygen dissociation curve**

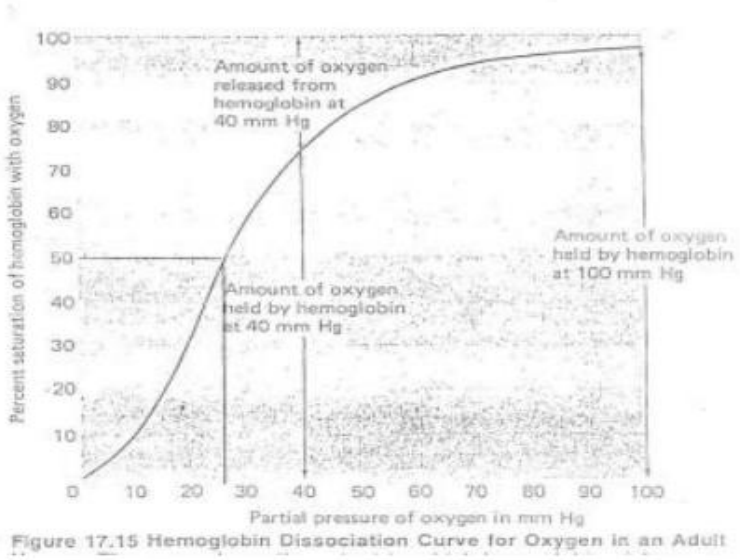


Figure 17.15 Hemoglobin Dissociation Curve for Oxygen in an Adult

In the Column:% of hemoglobin saturation with oxygen ,and in the row: partial pressure of oxygen in mmHg.

From this figure we understand three points:

1- Normally, when the partial pressure of oxygen in the lungs is about 100%; not all hemoglobin becomes saturated with oxygen but only 97%. More or less than that is pathological.
2- At tissue level, when partial pressure of oxygen is 40 mmHg; just 25% of oxygen is released, and the remaining 75% is still held by the hemoglobin.
3- We can determine the partial pressure of oxygen when 50% of hemoglobin is saturated, we call it P50. In the figure we see that the P50=26 mmHg.
   - ✓ **This curve is always the same for any person, despite the difference in Hb concentration.**

- Student Question: Why there's no 100% of hemoglobin saturated with oxygen ?
  Normally not occur ,this condition indicates there's something abnormal such as in the cardiac failure ,where the blood flow moves slowly (almost all blood becomes saturated with oxygen) ,But in polycythemia (not even 70% will be saturated).

- **This figure shows the relation between oxygen content and PO2**

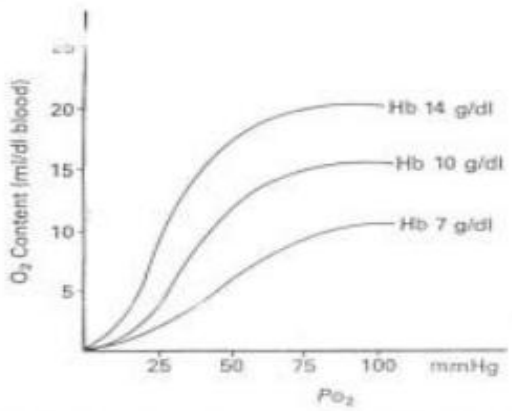
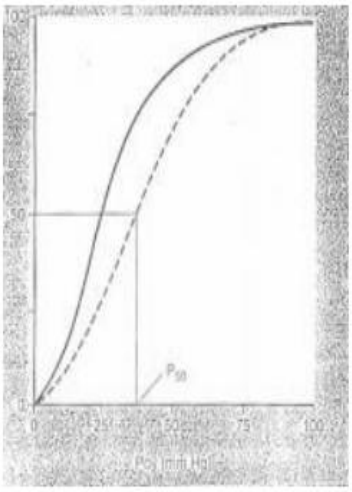


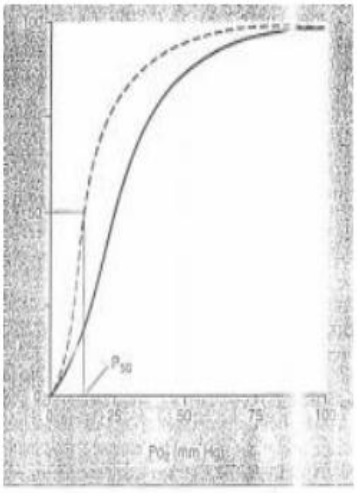
Fig. 6.12 Effect of anaemia on oxygen content of the blood at different Po_2 values.

In this relation we get different curves depending on hemoglobin concentration. So here we have different curves whether hemoglobin concentration is 7, 10 or 14 g/dl.

- Hemoglobin has 4 hemes ,when the blood passes through the body ,the 4 subunits do not bind to the oxygen directly at the same time ,its binding **ONE BY ONE** .This also applied on the other hemoglobin in the same RBC .
- Binding of the first heme with the first oxygen molecule facilitates the binding of the second one and so on till the 4^{th} subunit.

- We said that the hemoglobin dissociation curve in an adult human is always the same and does not change, but sometimes in the same individual in some conditions this curve changes:

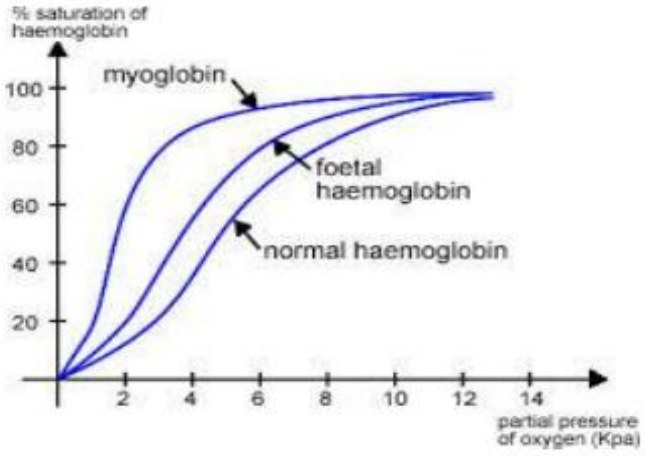




The curve shifts to the right	**The curve shifts to the left**
*P50 is increased than normal	*P50 is decreased than normal
*Affinity of hemoglobin for O2 is decreased, which means there is demand for oxygen.	*Affinity of hemoglobin for O2 is increased, which means either a pathological condition or that there isn't much need for oxygen.
*Factors that shift the curve to the right: ⇧ **Partial pressure for CO2** ⇧ **2,3-DPG** ⇧ **Temperature** ⇩ **PH**	*Factors that shift the curve to the left: ⇩ **Partial pressure for CO2** ⇩ **2,3-DPG** ⇩ **Temperature** ⇧ **PH**

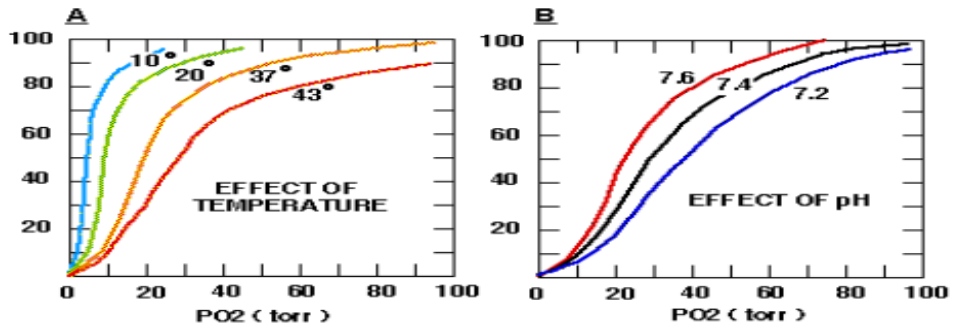
- **Remember :**
  - ✓ **Heme part bind to oxygen**
  - ✓ **Globin part binds other elements such as CO2 ,hydrogen , 2-3DPG**

- **Here in this curve, we see the differences in dissociation of oxygen from adult hemoglobin and fetal hemoglobin.**
- In fetal hemoglobin the curve is shifted to the left, P50 is low and the affinity is high, so it doesn't release oxygen easily . And we see myoglobin and hemoglobin, the myoglobin curve is too much shifted



to the left, which means that myoglobin in the muscles does not release oxygen easily unless the partial pressure for oxygen is very low and there is so much need especially in exercise.

❖ *** Here we see the effect of pH and temperature.**



* When the temperature is high the curve shifts to the right ,and when it is low it shifts to the left.

* When the PH is high the curve shifts to the left, and when it is low it shifts to the right.

➢ **Here we apply The effects of Mammalian Size on oxygen dissociation from hemoglobin :**

The elephant is too much to the left, because it is lazy. The cat and mouse are on the right, they move a lot. The human is in between.

- Too much to the left: Elephant
- Too much to the right: mouse

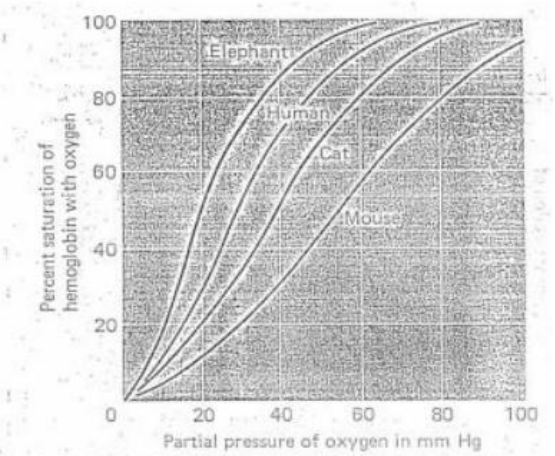


Figure 17.18 Effects of Mammalian Size on Oxygen Dissociation from Hemoglobin.

Blood parameters

We knew 3 parameters :

1) **Hemoglobin concentration of the blood [Hb] : 15g/dl { Regardless of their sex}**
2) **Red blood Cell Count(RCC): 5 millions cell/mL { Regardless of their sex}**
3) **Hematocrit (HCT) : 45% { Regardless of their sex}**

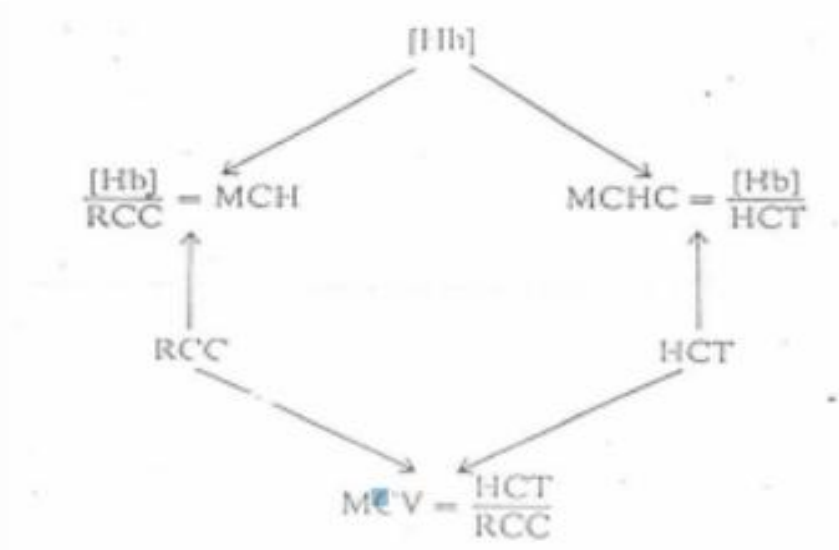
Table 27–5. Characteristics of human red cells.[1]

		Maie	Female
Hematocrit (Hct)(%)		47	42
Red blood cells (RBC) ($10^6/\mu L$)		5.4	4.8
Hemoglobin (Hb) (g/dL)		16	14
Mean corpuscular volume (MCV) (fL)	$= \frac{Hct \times 10}{RBC\ (10^6/\mu L)}$	87	87
Mean corpuscular hemoglobin (MCH) (pg)	$= \frac{Hb \times 10}{RBC\ (10^6/\mu L)}$	29	29
Mean corpuscular hemoglobin concentration (MCHC) (g/dL)	$= \frac{Hb \times 100}{Hct}$	34	34
Mean cell diameter (MCD) (μm)	= Mean diameter of 500 cells in smear	7.5	7.5

[1]Cells with MCVs > 95 fL are called macrocytes; cells with MCVs < 80 fL are called microcytes; cells with MCHs < 25 g/dL are called hypochromic.

Absolutely ,You should study the the numbers for both female and male specifically for the first 4 parameter.

- **<u>We use these three parameters to calculate other parameters as the following diagram shows:</u>**



1) ***Mean Cell Hemoglobin (MCH)***
2) ***Mean Cell Hemoglobin Concentration (MCHC)***
3) ***Mean Cell Volume (MCV)***

1_ *Mean Cell Hemoglobin (MCH)* : Indicates the **amount of hemoglobin in 1 RBC** , and should always correlate with the MCV and MCHC .

- MCH = (Hemoglobin * 10) / RBCs count in millions
- Normal value= 28-32 Picograms/cell, It is almost fixed within this range
- If it was below 28 pg/cell then the cell is hypochromic. If it was above 32 pg/cell,the cell is hyperchromic
- The MCH actually does not give us the right knowledge, it indicates the amount of hemoglobin in one RBC in Picograms, it does not indicate if it is normal or not. You have to relate this MCH to MCV (Mean Cell Volume) and MCHC (Mean Cell Hemoglobin Concentration) .In MCHC,if the hemoglobin occupies 32-36% of MCV then the RBC is normal.
- So MCH alone is not enough, it should be correlated with MCV and MCHC.
- This is like when you want to know the right measures of a person, you should relate the weight to the height and not only depend on one of these measurements; an 80 kgs person seems fat, but knowing its height is 1.85 meters then his weight seems normal).
- For example; in pernicious anemia if we just take MCH it is hyperchromic but when we relate it to MCV it is normochromic

2- Mean Cell Hemoglobin Concentration (MCHC):

Indicates **amount of Hb in RBC** ,whether the red blood cells are normochromic, hypochromic, or hyperchromic.

- MCHC= (Hemoglobin*100)/hematocrit
- Normal value= 32-36%
- An MCHC below 32% indicates hypochromia, an MCHC above 36% indicates hyperchromia, and RBCs with normal MCHC are termed normochromic

3- Mean Cell Volume (MCV): Indicates whether the red blood cells appear normocytic, microcytic (in anemia or thalassemia), or macrocytic. So it **indicates the volume of the cells**.

- MCV= (Hematocrit * 10)/RBCs count in millions
- Normal value= 80-100 fL (femtoliters, or 10^-15 L).
- If the MCV is less than 80 fL the RBCs are microcytic. If the MCV is greater than 100 fL the RBCs are macrocytic. If the MCV is within the normal range the RBCs are normocytic

* So all these parameters are used to diagnose different types of anemia.

***Summary:

1) MCV: gives us an idea about the **size of RBCs** (normocytic or macrocytic or microcytic).

2) MCH: gives us an idea about **the amount of Hb in 1 RBC**.

3) MCHC: gives us an idea about **the amount of Hb in RBCs** (normal – normochromic – or abnormal –hypo or hyper chromic –).

The normal **RANGE** for each parameter :

- ✓ MCV : (80-90) fL
- ✓ MCH : (28-32) pg
- ✓ MCHC : (32-36) g/dl .

MCV is normal if its value within the range or a little bit above or below normal. **This is only for MCV**. MCHC has to be exactly in the range to be normal.

➢ Describe these cells ?

***** Ex .1***

- MCV = 91 normocytic
- MCH = 31
- MCHC = 34 normochromic

****Ex.2***

- MCV = 67 microcytic
- MCH = 22
- MCHC = 33 normochromic

****Ex .3***

- MCV = 67 microcytic
- MCH = 20
- MCHC = 30 hypochromic

***Ex .4**

- MCV = 113 macrocytic
- MCH = 38
- MCHC = 33 normochromic

***Ex .5**

- MCV = 92 normocytic
- MCH = 26
- MCHC = 29 hypoochromic

In general:

- ✓ Normally, we have 4*10^6 RBCs in female and 5*10^6 RBCs in male.
- ✓ Low count of RBCs: Anemia or Erythrocytopenia .(In male ,below 5millions RBCs ,but in female below 4millions RBCs).
- ✓ High count of RBCs: erthrocytosis or polycythemia.

Anemia / Erythrocytopenia

- ***Anemia*:** is a group of disorders characterizes by quantitative or qualitative deficiency of the circulating RBCs.(deficiency in number or contents of RBCs)
  Notice that: sometimes anemic pt has normal count of RBCs but with low Hb content
- ***General causes of anemia :***
  1) Deficiency in the production of RBCs.
  2) Excessive loss.
  3) Excessive destruction
- ***Classification of anemia according to :***

1. Morphology

Types	***Proplem***

Normochromic Normocytic anemia	**Size and content normal, low count of RBCs**
Hypochromic Normocytic anemia	**The size is normal but the content of Hb is low**
Hypochromic Microcytic anemia	**The cells are small with low content of Hb**
Macrocytic	**Large cells with low count e.g. pernicious anemia**
Microspherocytic	**Abnormality in the shape (most probably there is hemolysis)**

2) Etiological :

i. Increased blood loss :

a. Hemorrhage(acute ,chronic)

b. Hemolysis (either in circulation or in the medium ,something in the plasma)

ii. Decreased blood production :

a. Nutritional.

b. Bone marrow failure

❖ ***The Effect of anemia on the body***

When we talked about plasma proteins we said fibrinogen & globins affect the viscosity .On the other hand ,in the blood the main factor that affect viscosity is **RBCs** then **plasma proteins** .So***, viscosity decreases*** in anemic patient because of ***decrease amount of RBCs*** ⟶ the ***resistance to blood flow in the peripheral blood vessels decreases*** ⟶ ***blood flows easily*** and another factor that makes the problem worse, anemia causes hypoxia ,so there's vasodilatation .As the result ,because of viscosity and vasodilatation ⟶ ***increase the amount of blood returning to the heart*** and consequently increase heart rate and beats ⟶ so ***increase the workload on the heart***.

- So anemic patients have to avoid heavy exercises because they might have cardiac failure (because almost all of the oxygen is being utilized).

Erythrocytosis / polycythemia

An increase in the concentration of RBCs in the circulation. Usually, when we say **erythrocytosis** we mean **physiological condition** while in **Polycythemia** we mean an abnormal condition, may be referring to **blood cancer.**

**from Wikipedia :

The 2 terms are not synonymous because polycythemia refers to any increase in red blood cells, whereas erythrocytosis only refers to a documented increase of red cell mass

➢ *Classification :*

1. **relative erythrocytosis** : In dehydration
2. **True erythrocytosis** :

   *With high erythropoietin:

   a. Physiological condition (hypoxia such as being at high altitude)

   b. drugs : cobalt

   c.androgens

   d.thyroxine

   *With low or normal erythropoietin: this is a cancerous case such as Polycythemia Vera.

▪ *The effects of erythrocytosis/polycythemia on the body:*

A. Viscosity increases ⟶ increasing the resistance. The blood flow through the peripheral blood vessels is often **very sluggish**. Increasing blood viscosity ⟶ decreases the rate of venous return to the heart(the heart pumps hardly).BUT, because of the blood volume in polcythemia /erthrocytosis is greatly increased at the same time ,so that it tends to increase venous

return. At the end, **the workload on the heart is increased by increasing the blood volume** (sometimes its reach 7 to 8 liters). (While in anemia, the increase in the workload on the heart is due to viscosity and the increased heart rate)

B. In ploycethemia , not the whole blood becomes oxygenated , so some RBCs will not be oxygenated , that result in some patient will have blue skin ,**Its called Cyanosis.**

Tests on RBCs

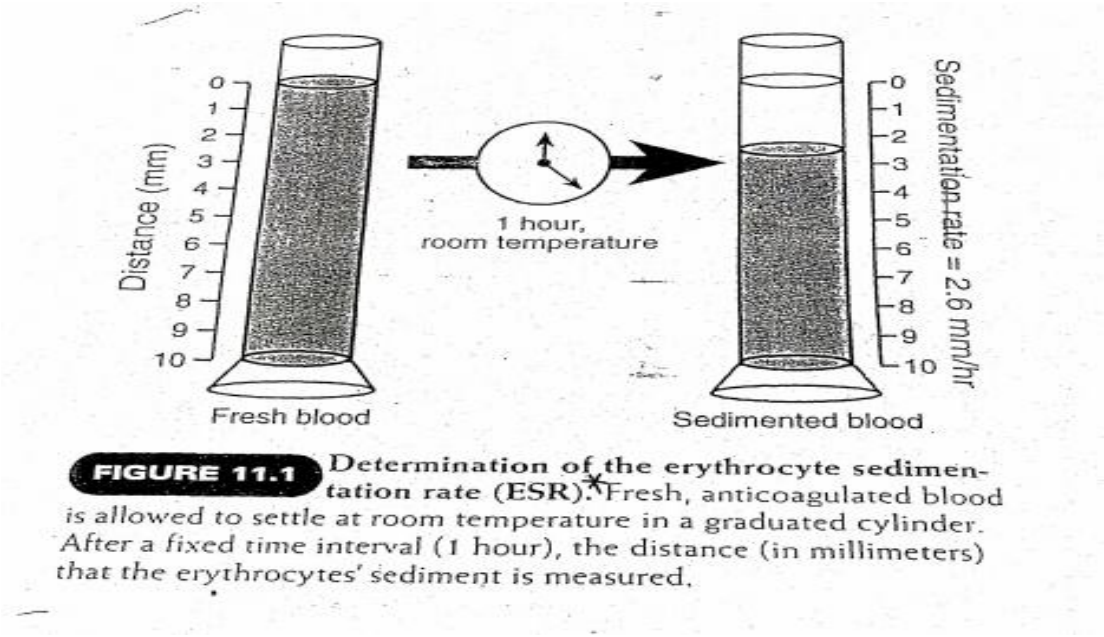


FIGURE 11.1 **Determination of the erythrocyte sedimentation rate (ESR).** Fresh, anticoagulated blood is allowed to settle at room temperature in a graduated cylinder. After a fixed time interval (1 hour), the distance (in millimeters) that the erythrocytes' sediment is measured.

***Indicate the presence of organ disease or blood disease.**

- **Erythrocyte sedimentation rate (ESR):** very important & common test.
  Fresh anti-coagulated blood is placed in a graduated cylinder, kept in lab conditions (normal temperature & humidity) for 1 hour, afterwards the free plasma on the top is measured (the height) which indicates the ESR. In which RBCs precipitate without any effect (only) the time and some other factors.

- There are more than one type of tube ,so other test perform another tubes , in this case the clear data will be differ .
- Normally, free plasma distance usually reaches from (0-5 OR 1-5) mm in males and it reaches up to 15 mm in females if we're using the Wintrobe method.

- There are 4 methods to measure ESR **(Wintrobe, Landau, Cultur & Westergen – Sedimentation Rate Tubes**) vary according to:
  - A. Range of graduation.
  - B. Height of blood column.
  - C. Length of tube.
  - D. Internal diameter of tube.
  - E. The amount of blood.
- Each method has its own normal range for men or women
- **ESR or Rouleaux formation of RBCs** :They sediment above each other (similar to a cylindrical packet of coins).
- Factors affect the ESR :
  1) Erythrocytes
  2) plasma composition
  3) mechanical & technical factors (errors)

1_Erthrocytes

- ✓ Size: the larger the cells the faster the sedimentation rate,so in the microcytic cell ⟶ ESR higher than normal
- ✓ Shape : any change in the shape ,it will affect the ESR so the cell don't settle down easily .Therefore, in sickle cell anemia & in spherocytic cells ⟶ ESR is low (they don't settle &sediment easily)
- ✓ Cell count:
  - **in Anemia** the ESR is relatively **high**
  - **in polycythemia** the ESR is **normal or low**

❖ **The doctor asked :Why ESR is high in anemia?**

Normally RBCs have a negative charge on their surface, so there is repulsion between cells (cells remain suspended) while in anemia the repulsion decreases leading to easily sedimentation (because of the less count).

2_Plasma proteins

Is the most important factor determining the ESR, rouleaux of RBCs are affected mainly by the levels of plasma protein especially the levels of fibrinogen and globulins increase ESR.(when the concentration of fibrinogen and globulins increase⟶ the ESR increases).

3- Mechanical & technical factors (errors)

- ✓ Temperature: the ESR increases with large change in temperature
- ✓ Humidity.
- ✓ Tubes: should stay 100% vertical in the racks. A tilt of 3° can cause errors up to 30%.

- ESR is a non-specific test (not a diagnostic test); it indicates the presence of a disease (x-disease) but doesn't specify it. So we need further investigation.

- ESR increases in:
  - All infections (acute or chronic)
  - Connective tissue destructive diseases.

- ESR is **high** in: Rheumatoid arthritis, Tuberculosis (very high it can reach up to 50 mm) & in Acute hepatitis.
- So we conclude that ***the age*** plays a role in ESR test , **ESR is low in young people and high in elderly** (both males and females), That's because most old people suffer from connective tissue destructive diseases.

❖ **Why the absorption of vitamin B12 happens in the ileum and the absorption of the folic acid happens in the jejunum ?**

- Vitamin B12 absorption in the ileum because there are many receptors for B12 in the ileum and there's less bacteria there .
- Absorption of folic acid in jejunum because there's carboxyl peptidase in the jejunum .

(لا تجعلوا الله أهون الناظرين إليكم)

THE END

**** Your colleague :**

Dania Al-Wreikat