Lecture : 5

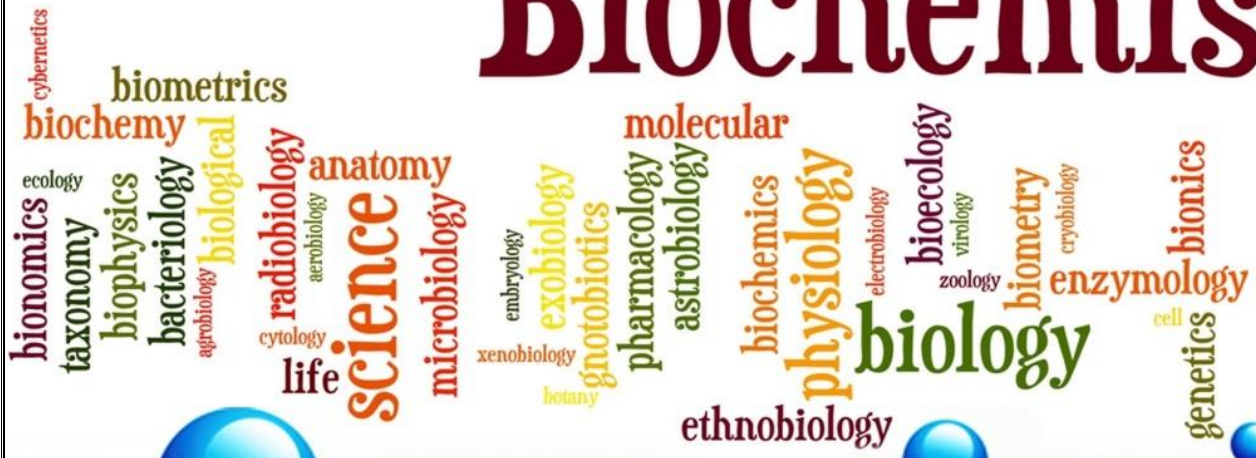
Dr. Name : Nafeth Abu Tarbosh

Done By : Ola Atif

☐ Slide ■ Sheet

Medical Committee
The University of Jordan

Biochemistry

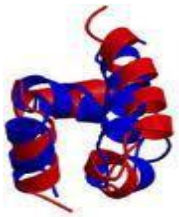


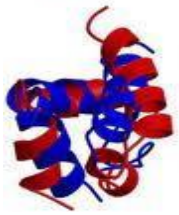
Mousa Subeh

Polypeptide and protein structure

Protein conformation:

Recall that Amino acids unite together by peptide bonds. Any protein exists in many conformations , any little change in the protein's shape gives a new conformation (conformation means shape) , the shape changes by many factors (such as : temperature , environmental factors , regulator for a protein to be active or inactive) .



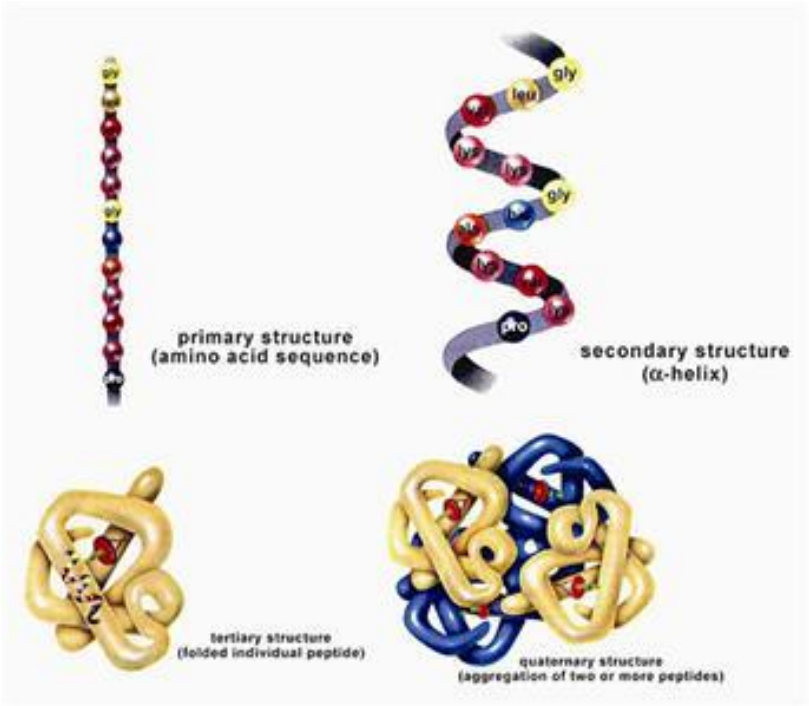


As shown in the picture , these two structures represent the same protein.
However, there are differences in their shapes and these differences make the different conformations.

if we have huge number of inactive conformations we should have at least **one** active conformation called the native conformation.

Native conformation means, the active conformation of the protein which is responsible for its function.

Levels of protein structure :



 # Primary structure (1°) :
The sequence of amino acids , from N to C , when they form a protein , their numbers and arrangement are unique.

Remember :
when 2 amino acids form a peptide bond between them they form Dipeptide
3 amino acids form tripeptide
4 amino acids form tetrapeptide
5 amino acids form pentapeptide

Secondary structure (2°) :

* **Chain of amino acids :** a sequence of amino acids starts with free amino group and ends with free carboxylic group

Some proteins have 1 polypeptide chain (one subunit) like Myoglobin which stores oxygen in the tissues while some others have more than one polypeptide like Hemoglobin

* We determine the number of polypeptide chain in the protein according to the number of free amino and carboxylic groups

For example :
* 1 free amino group and 1 free carboxylic group means 1 polypeptide chain
* 2 free amino groups and 2 free carboxylic groups means 2 polypeptide chain
And so on …

We have a linear chain of amino acids in the ribosome , where the translation of the amino acids occurs . When the mRNA gets on the ribosome , it will translate it to a sequence of amino acids .
the nearby regions start to rotate around each other , and the hydrogen bonding forms between the back bone of the amino acids which makes the secondary structure

examples for secondary structure : α-helix and β-pleated sheet

#**Tertiary structure (3°) :**
3D arrangement of all the atoms (the polypeptide chain) , bonding between the side chains , between the bake bones , and between the side chain with the back bone

Quaternary structure (4°) :

Many polypeptide chains in the protein are present , the most important example for this is : Hemoglobin , it consists of 4 polypeptide chains : 2 α and 2β.

Note: The one polypeptide of Myoglobin resembles each polypeptide of Hemoglobin with slight differences and the certain connections between these repeated polypeptides is what creates the quaternary structure .

** **The primary structure (1°) :**

The number in sequence of amino acids , the liner arrangement and the types of them

-**Their shape of amino acids is zigzag** : because one of the R groups is up and the other one is down to give them enough space , and avoid the steric repulsion between them . What permits this zigzag shape to form is the rotation that occurs in 2 bonds (NOT peptide bond) : but between phi **(Φ)** and psi **(Ψ) bonds** , so the amino acid can go up and then rotates and go down.

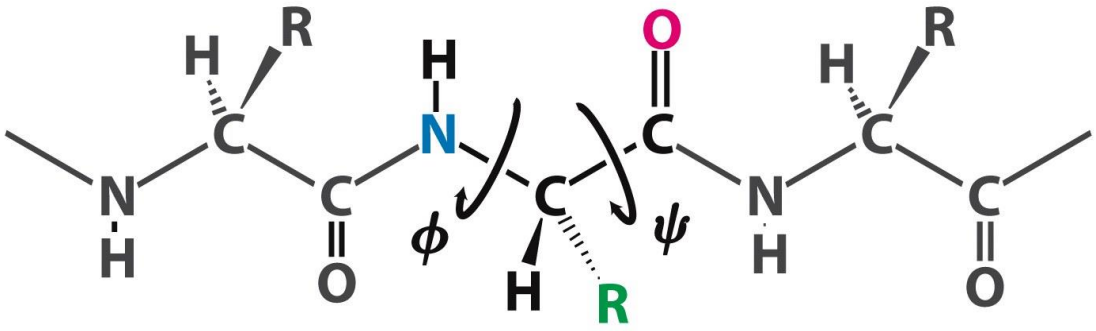


Figure 2.22a
***Biochemistry,* Seventh Edition**

Keep in mind that generally peptide bonds are rigid and no rotation occurs around them

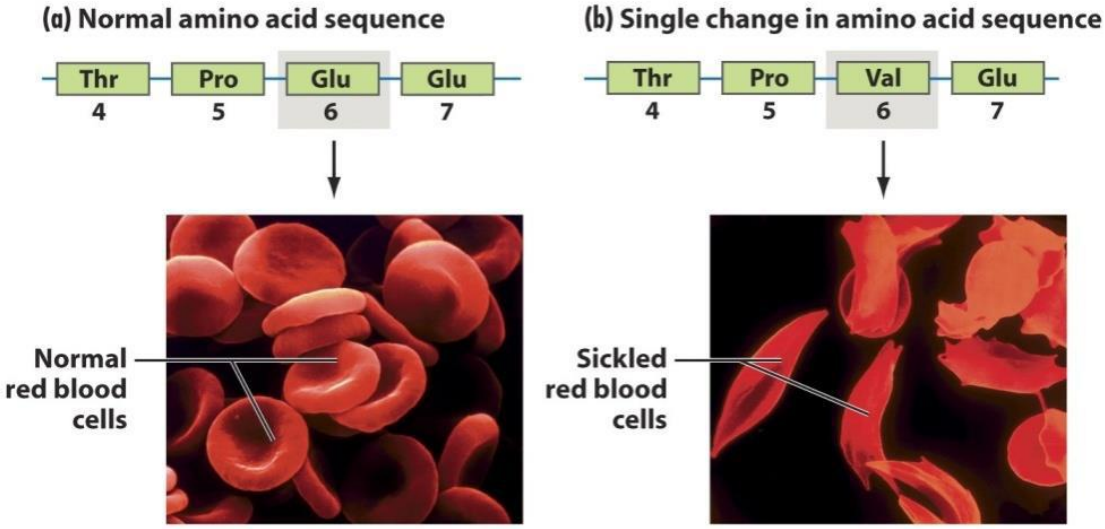
- **The 1° structure determine the final shape of the protein** , because it has the arrangement of the amino acids which determines how the bond will form For example : when the amino acid number 4 is supposed to be methionine the whole protein will be affected slightly or significantly if it's substituted by another amino acid …ext , because every amino acid has a special property so their arrangement determine the bonding to give the final form of the protein

the change of one amino acid causes a change of the protein's shape but some mutations keep the overall shape and changes the function of the protein.

When one amino acid in a sequence of amino acids got a problem (like : mutation) that causes diseases , for example : **sickle cell anemia**

What happened in sickle cell anemia ?

A substitution in amino acid **number 6** .V**aline** instead of **glutamic acid.** Glutamic acid is a negatively charged amino acid , it is changed into valine which is non-polar amino acid . Glutamic acid (number 6) comes on the surface of β-chain in hemoglobin , when it substituted to valine the shape of the hemoglobin will be the same but functionally different.



Why the function will be different ?

On the surface of the hemoglobin (β – chains exactly) there is a small pore where the glutamic acid located inside , because the glutamic acid is negatively charged when the hemoglobin molecules comes in contact with each other a repulsion will happen , so each hemoglobin molecule will stay separately from each other in the RBC , and that makes the RBCs in spherical shape

When we replace the glutamic acid with valine , two hemoglobin molecules will not repulse and come in contact with each other , also a third and fourth molecules will come , as a result that is called **hemoglobin aggregates**

These aggregates will accumulate on the surface of RBC , and that is why the shape becomes sickle

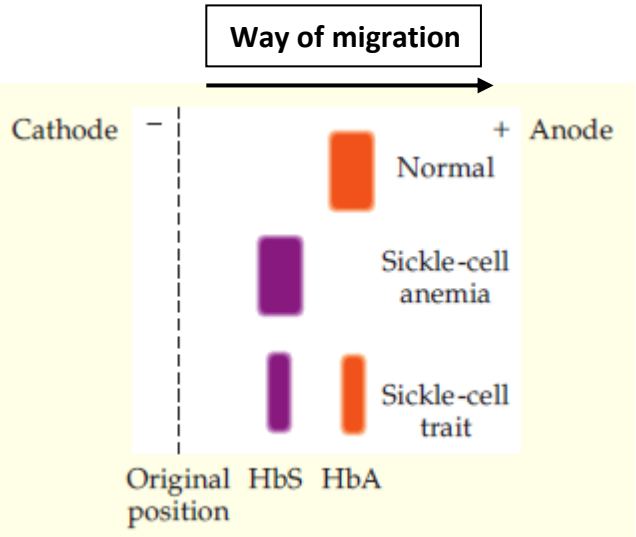
Note:

Sickle cell disease has a strong relationship with the inherited factors that's why it's well found in those individuals whose parents are relatives, like in KSA.

How to know sickle cell animia in the lab ?

By a technique called **Gel Electrophoresis** . Means to expose the proteins to electricity . Bring a substance like a gel (gel has pores NOT totally solid and things can go throw it) , bring this substance , heat it , put a comb in it (like fingers) and let it cool , remove the comb , you will get pores to put proteins in , then put the whole gel in a buffer and expose it to electricity

As shown in the picture :
when the protein gets an electricity , attraction of the opposite charges , proteins of hemoglobin has negative charge so they start to migrate towards the positive electrode (anode)



What is the differences between normal hemoglobin and sickle cell hemoglobin when they get an electricity inside the gel?
The normal hemoglobin has **one more negative charge** than sickle cell hemoglobin **,**
*** Remember** : normal hemoglobin has glutamic acid (negatively charged) but sickle cell hemoglobin has valine (non-polar and not charged)
So the normal hemoglobin will be faster in the gel because the attraction is higher
But if someone has sickle cell anemia we will see the same van of the hemoglobin ; however , it will be closer to the negative electrode (cathode) , because it didn't move at the same speed of the normal hemoglobin
The **heterozygous people** الحاملين للصفة الوراثية

Those people have two bands , the normal and sickle hemoglobin

A student asked the doctor : what will happen if a mutation happened and we get aspartic acid instead of glutamic acid (both are negatively charged) ?
The doctor answered : even they both have negative charge , the protein will change because it is not about the charge only , it also depends on the volume and the bonding of each amino acid . In hemoglobin there is no mutation that replaced the glutamic acid by asparatic acid so we can't know the consequences

****Shape-Determining & stabilizing Interactions in Proteins** :

What bonds determine the shape of protein ?

Hydrophobic interaction and van der val forces
they make any sequence of amino acid in a situation of less energy and more stable state , so it is better for hydrophobic atoms to be close to each other . inside spherical proteins we find inside the protein hydrophobic amino acids and on the surface hydrophilic amino acids polar ones either charged or uncharged

what bonds stabilize the shape of protein ?
disulfide bridges (bonds)

****Secondary structure (2°) :**
#It is hydrogen bonding between the back bone of the amino acids NOT the side chain

#We have a sequence of amino acids (1° structure) which describes the liner sequence of amino acids (number , type of amino acid , their arrangement)

#The 2° Describes how the nearby amino acids rotate around each others structures
It is like building , we start from bases and exoskeleton then we connect the nearby regions by walls to make rooms , after that we connect the far regions by roofs .
The roof which connect the far regions represents the 3° structure and how the far amino acids connects together , the connection between the nearby structures represents the 2° structure by hydrogen bonds

#All the back bone of the amino acids are the same , so the folding of the back bond is the same . so we have few types of folding among the proteins regardless to the amino acids in them .

#If the nearby regions fold around each other most commonly it will form **a spring like structure** which we called **α- helix structure** .

#If we have a strand amino acids and another strand come over it and have a hydrogen bonds between them , they will form **β- pleated sheet**

the hydrogen bonding between the back bone of amino acids form a regular structure which has a certain features

** **α – helix :**

#One turn of α-helix contains hydrogen bonding up and down , and it is **parallel** to the long axis of the α-helix

#One turn (pitch) has 3.6 amino acids exactly ,because the bond happens between oxygen and nitrogen , so not all the amino acid is inside the pitch.

the hydrogen bonding happens between :
amino acid number 1 …with … amino acid number 4
amino acid number 2 …with … amino acid number 5

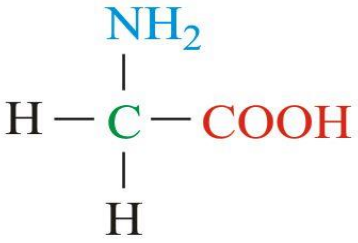
Be Careful : amino acid number 5 …with … amino acid number 2 down and with amino acid number 8 up

#the pitch is **5.4 Å** (Å = 1 x 10 ^ -10)

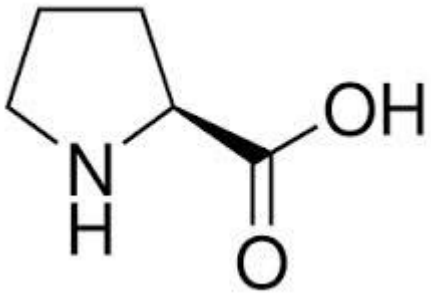
#the turns occurred are **right handed and clockwise**

**** amino acids NOT found in α-helix :**

 # glycine :
when the R group is bigger it gives bulk to stabilize the amino acid to stabilize the shape , but in glycine the R group is hydrogen atom , it is small , has high flexibility and keeps moving . and to put it in the α-helix you have to spend energy , and that is why it is entropically expensive



 # proline :
To do the helical shape it should rotate , but proline's side chain already engaged in the binding of the nitrogen of the back bone (it forms a ring) so there is no rotation around the back bone (psi bond)
Also , it can't make hydrogen bonding so it can't be found in α-helix , because the α-helix basically depends on the hydrogen bonds
So always **proline breaks (ends) the α-helix**



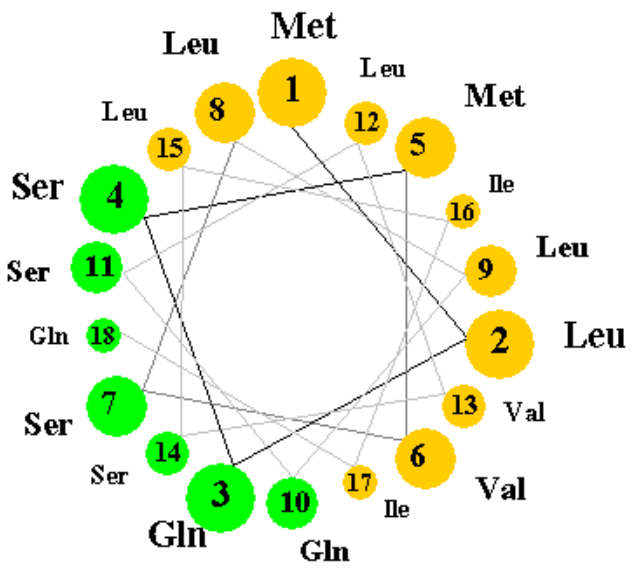
 # pair of charged amino acids with the same charge

They can't be beside each other in the α-helix because they will repulse , and will be entropically expensive

The bonds in the α-helix and any 2° structure is hydrogen bonds
For example : **myoglobin** stores the oxygen inside the tissues is **8 α-helices**
Hydrogen bonding is very weak but because of its big numbers can stabilize the shape of the protein

** **Amphipathic α – helix :**
The channels inside the membrane are proteins , these channels let the hydrophilic materials to pass throw it
The membrane is hydrophobic (consists of : cholesterol and phospholipids) , the internal structure is lipophilic and hydrophobic

The channels are cylinder , the α-helices turn , so the amino acid which directed to the outside is hydrophobic (like the lipids of the membrane) , and the amino acids which directed to the inside are hydrophilic
That's why we called this α-helices **Amphipathic α – helix**



Here are some amazing video hope they help you ☺
** Sickle cell disease
http://www.youtube.com/watch?v=R4-c3hUhhyc

** A word about amino acids and proteins structure
http://www.youtube.com/watch?v=iLHhwDc7Wgw

** proteins structures (very interesting)
http://www.youtube.com/watch?v=Q7dxi4ob2O4

Best wishes