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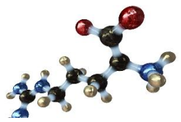


BioChemistry

وَلَقَدْ خَلَقْنَا الْإِنسَانَ وَنَعْلَمُ مَا تُوَسْوِسُ بِهِ نَفْسُهُ ۖ وَنَحْنُ أَقْرَبُ إِلَيْهِ مِنْ حَبْلِ الْوَرِيدِ

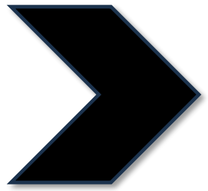
Final | Lecture #2

Protein Structure pt.2

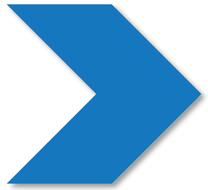
Written by : Ali al-sharaeh
Yaman Khalil

Reviewed by : Ahmad Mohy

Color coding used in the modified:

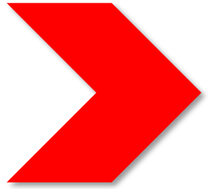


Black: the original slides



Blue: the doctor’s explanation/words

Gray: additional information and explanation



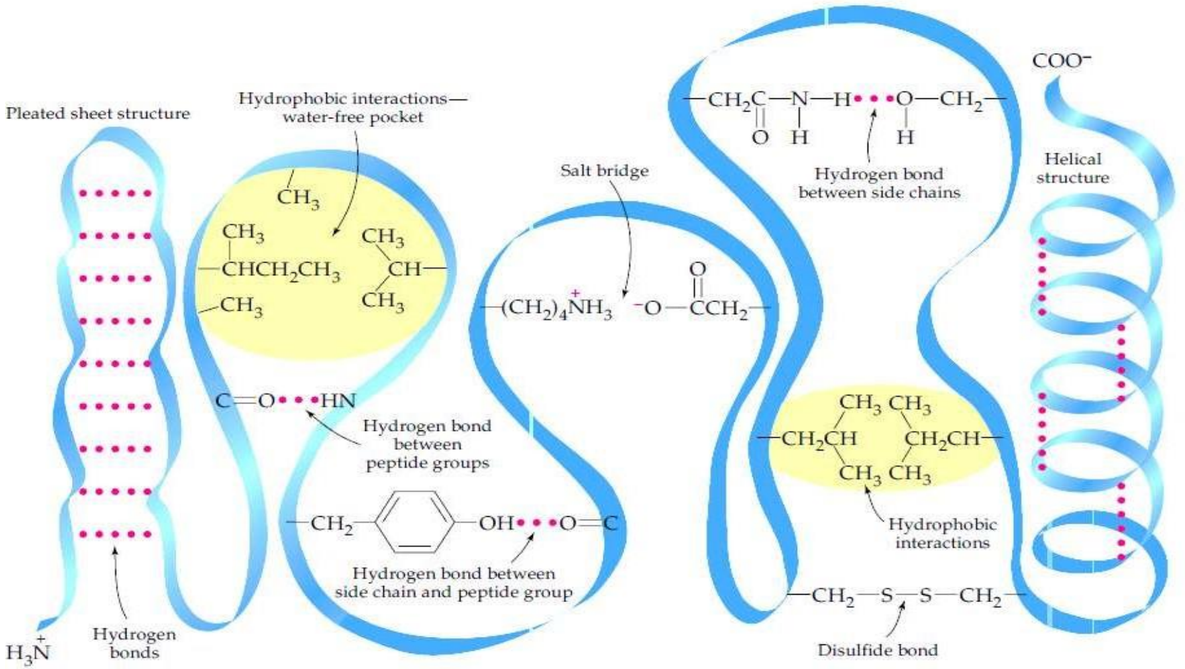
Red: important information

Shape-determining forces

- For secondary structures: hydrogen bonds between components of peptide bonds (**main force**), R groups also play a role in determining the structure=> remember there are some amino acids that can't exist in alpha helix or beta sheet (because of these R groups) (Don't focus on that point just kind of knowledge)

- Tertiary structure : Non covalent interactions between R groups, the peptide bond components do play a role but mainly R group interactions .

- That's why we find lysine near glutamate or aspartate (electrostatic) and hydrophobic amino acids clusters inward and so on . van der walls also MUST be in a close range (not too far neither too close), and although it is weak but a lot of them assist in making the structure

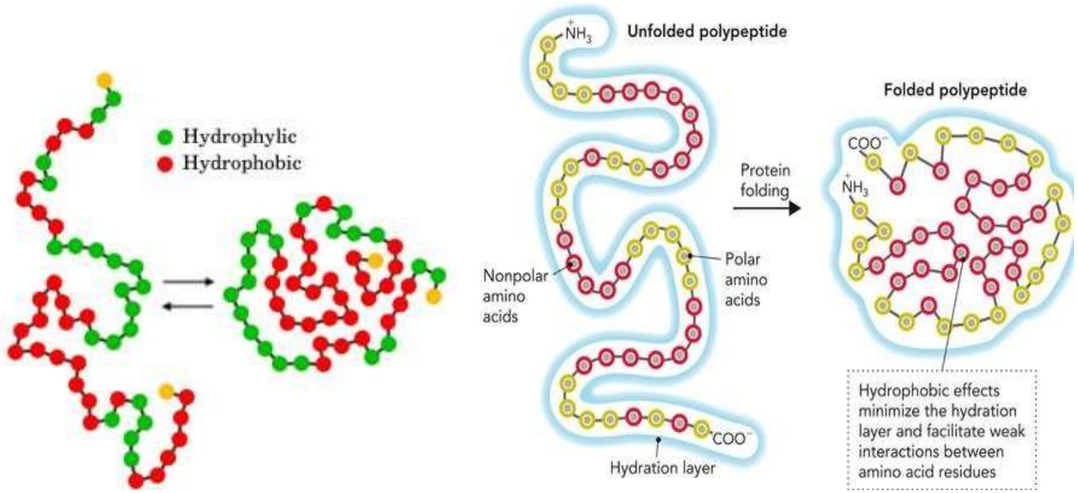


Localization of amino acids

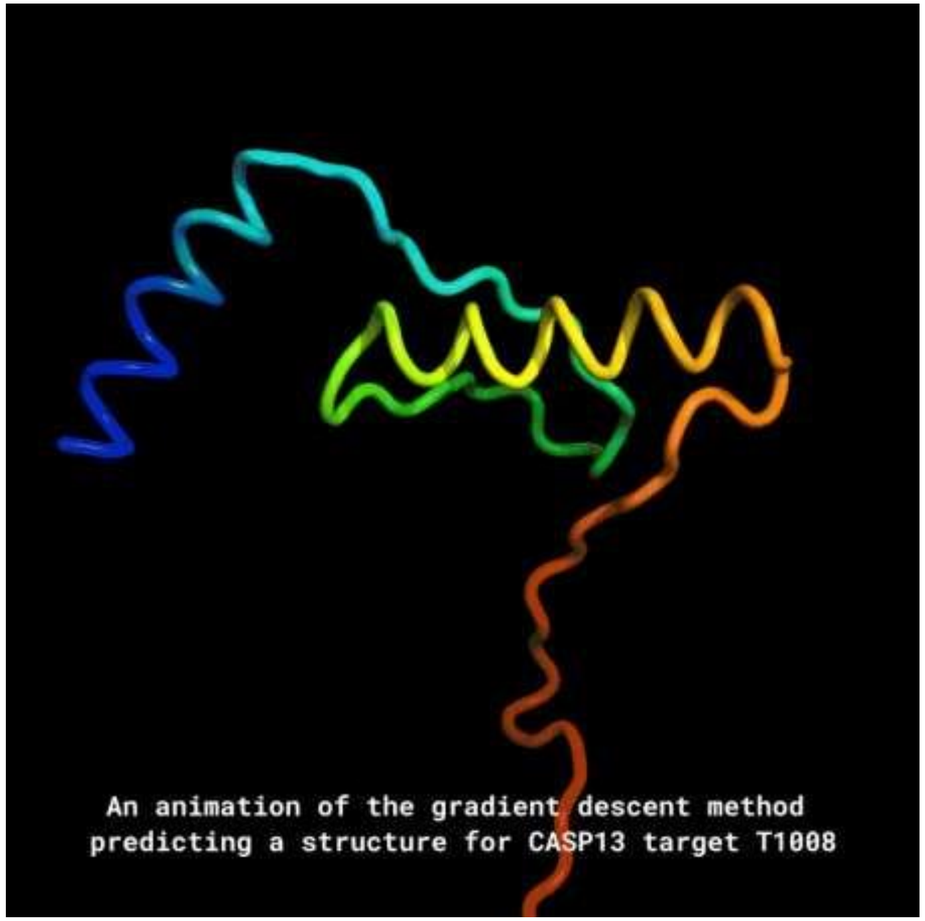
- Polar amino acids can be found in the interior of proteins
- In this case, they form hydrogen bonds to other amino acids or to the polypeptide backbone
- They play important roles in the function of the protein

Can polar amino acids be found in the interior?...YES

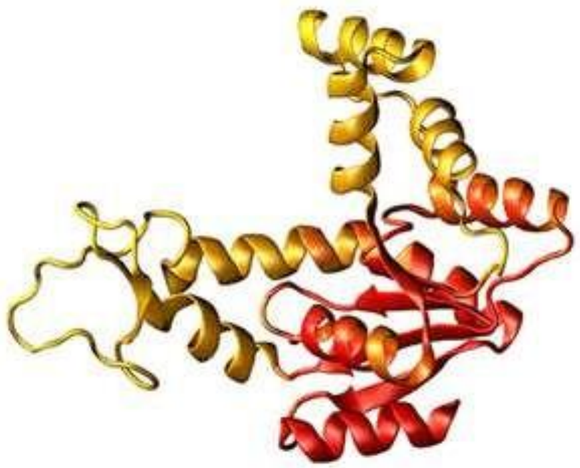
- the most important force in determining the 3d structure are hydrophobic interactions . For example, hydrophobic amino acids aggregate together into a cluster, forming a Hydrophilic surface and a hydrophobic core .
- Non polar amino acids can also be on the surface but mainly they are directed inward

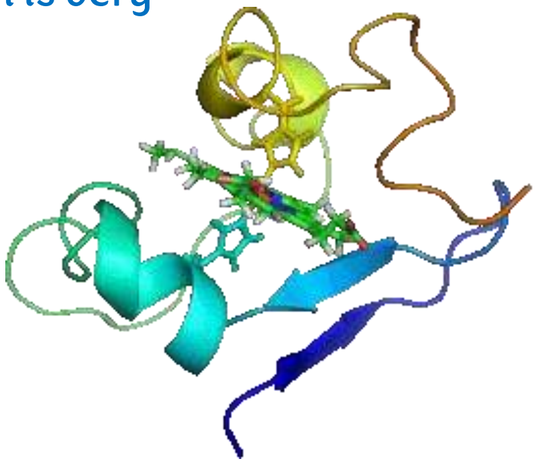


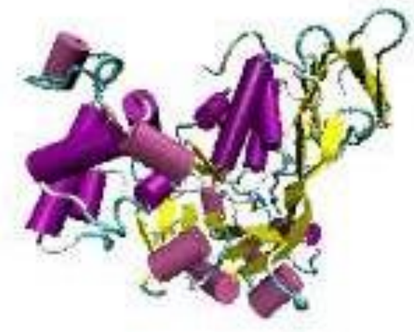
A hypothetical look at protein folding,
folding is a gradual process.(you can see the animation of folding in the lecture)



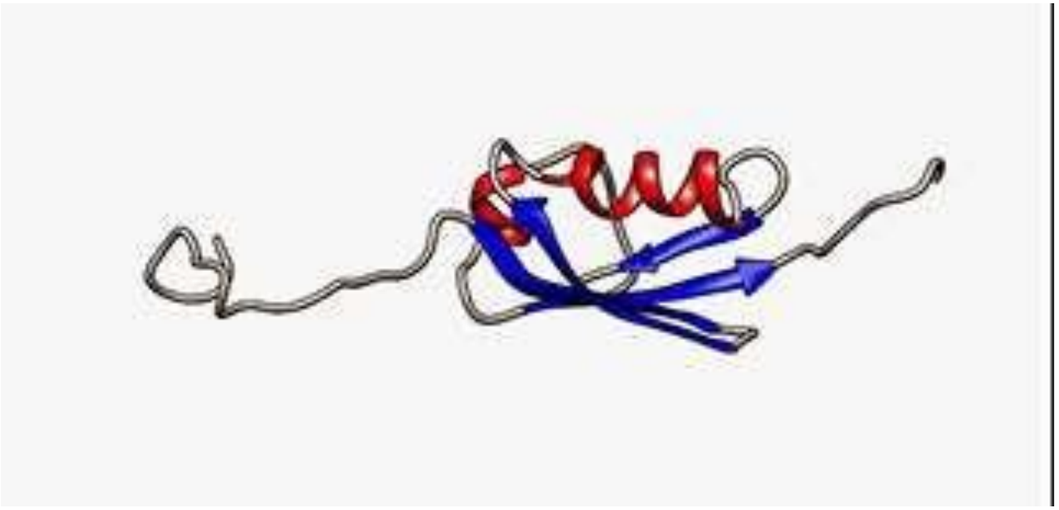
Protein are NOT static, they keep shaking (dynamic), which is very important for the structure.

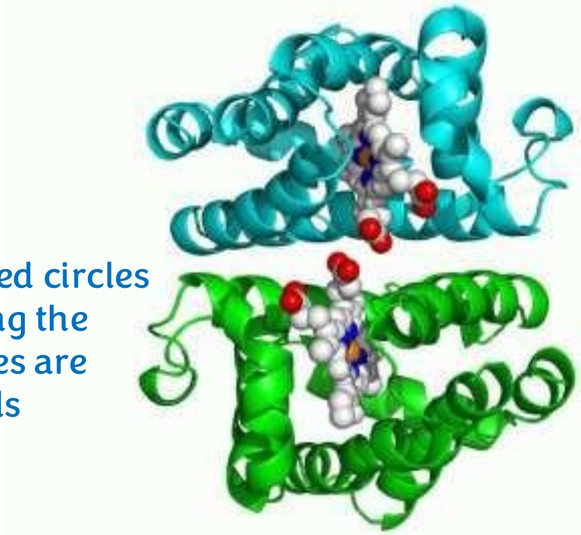






Yellow and teal are alpha helix, while the arrows are beta sheets





The red circles among the helixes are metals

Stabilizing factors

Covalent bonds and strong electrostatic interactions

- There are two forces that do not ***necessarily*** determine the three-dimensional structure of proteins, but stabilize these structures:
  - Disulfide bonds => Covalent bond
  - Metal ions => Electrostatic interactions & (coordinate) covalent bonds

(Further explanation) Overall, these forces don't determine the 3D structure, but in some limited cases they have a role .

Disulfide bonds

- The side chain of cysteine contains a reactive sulfhydryl group, thiol, (—SH), which can oxidize to form a disulfide bond (—S—S—) to a second cysteine.
- The crosslinking of two cysteines (which are both molecules instead of residue) to form a new amino acid, called cystine.

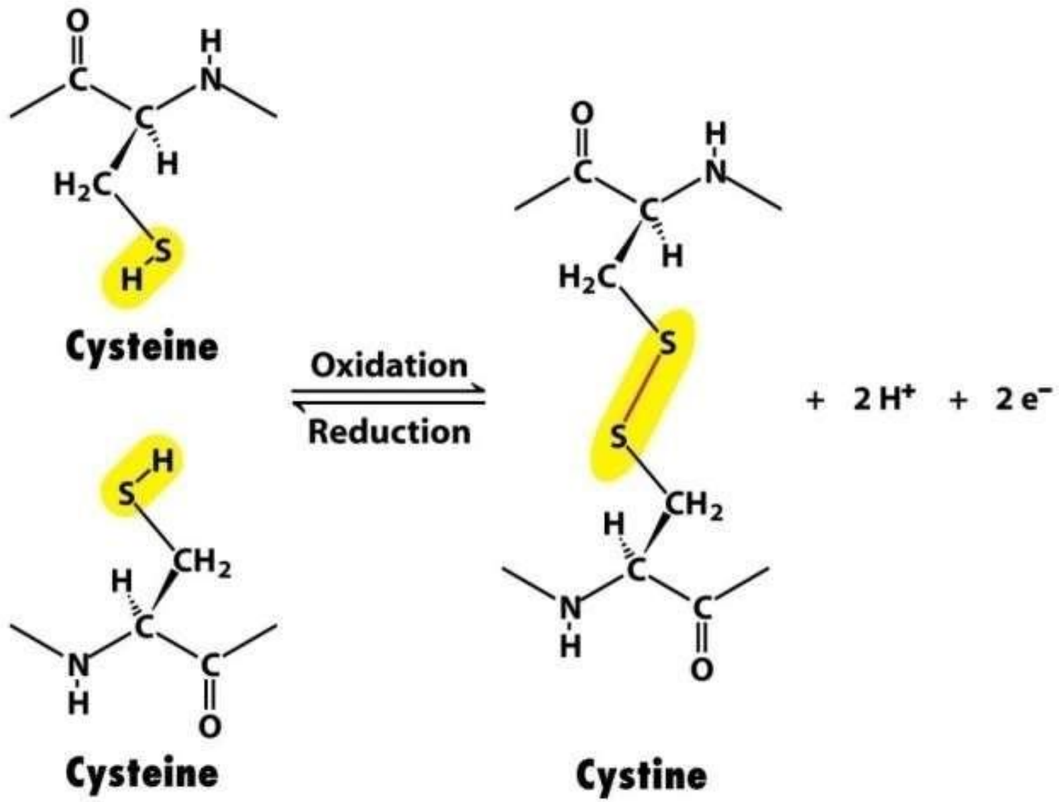
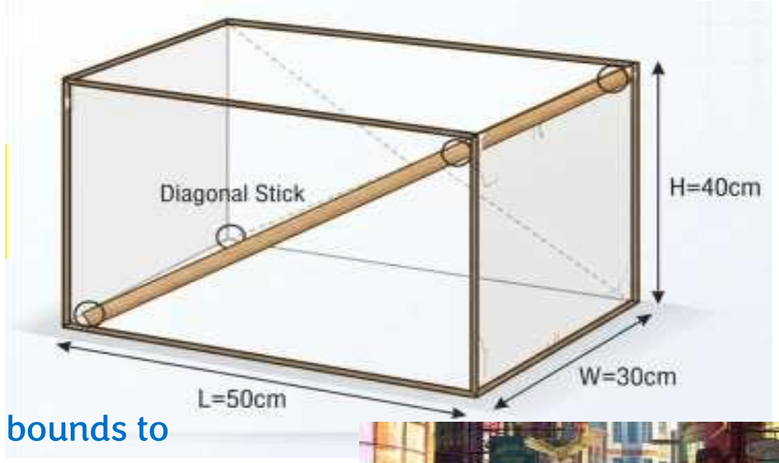


Figure 2-21
Biochemistry, Sixth Edition

metal ions

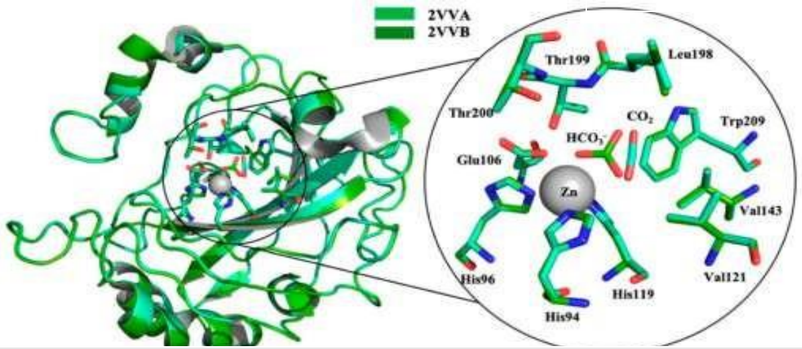
- Several proteins can be complexed to a single metal ion that can stabilize protein structure by forming:



- Covalent interaction (myoglobin) : heme (iron in heme) group which bounds to Histidine (which helps with stabilization)
- Salt bridges (carbonic anhydrase): electrostatic interactions within a protein

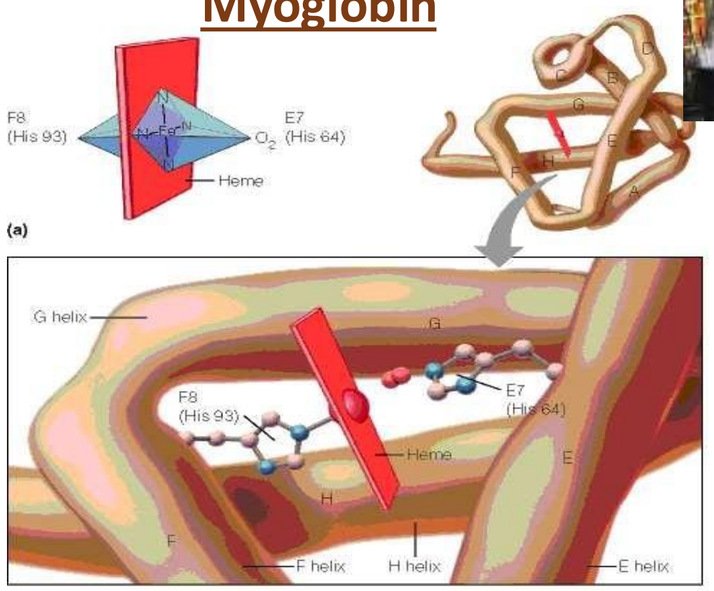
Carbonic anhydrase

Ribbon structure



This enzyme is responsible of the reaction between H2O and CO2 to produce H2CO3 and then it dissoiactes into HCO3- and H+ in the blood. It has a negative Zinc ion that forms salt bridges with Histidine residues.

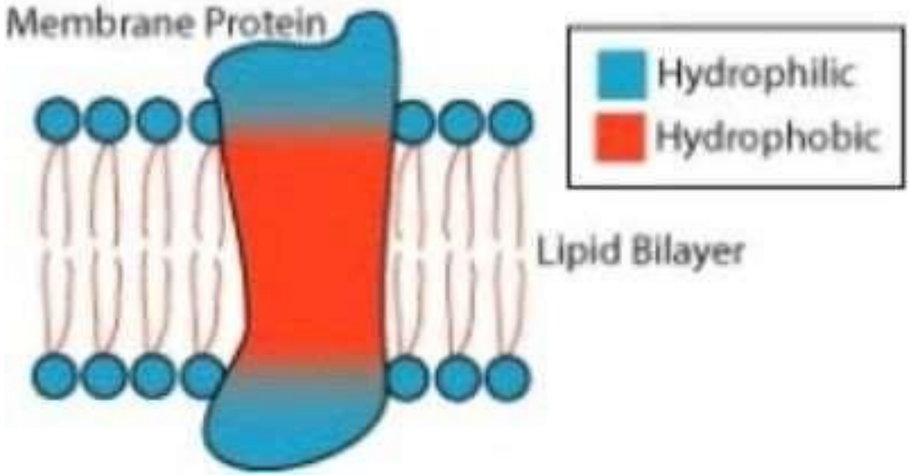
Myoglobin



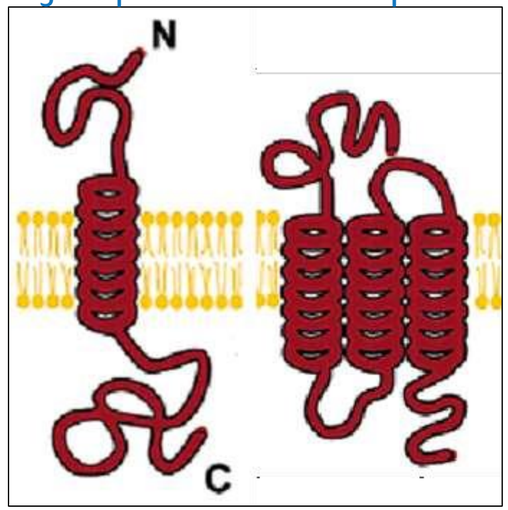
α-helices as transmembrane domains

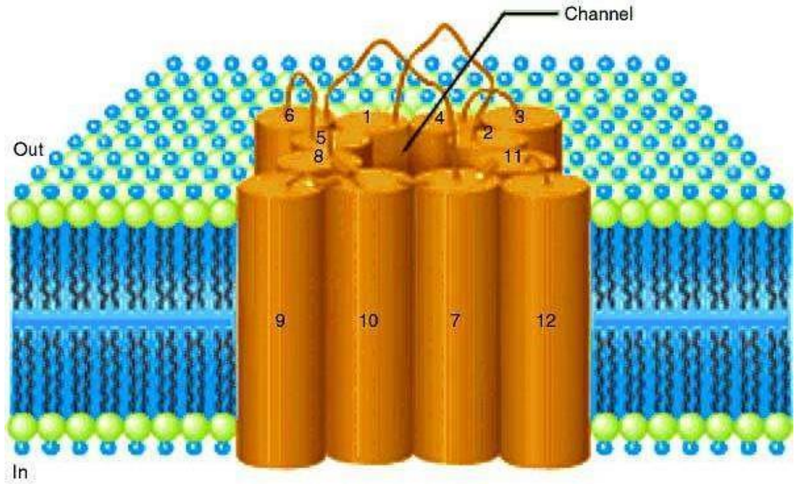
- Membrane-spanning proteins contain a **transmembrane domain that is an α–helix made of hydrophobic amino acids.**
- The α–helices of proteins with multiple transmembrane domains are connected by **loops containing hydrophilic amino acids located outside of the membrane.**
- Membrane ion channel proteins contain **amphipathic** α–helices.

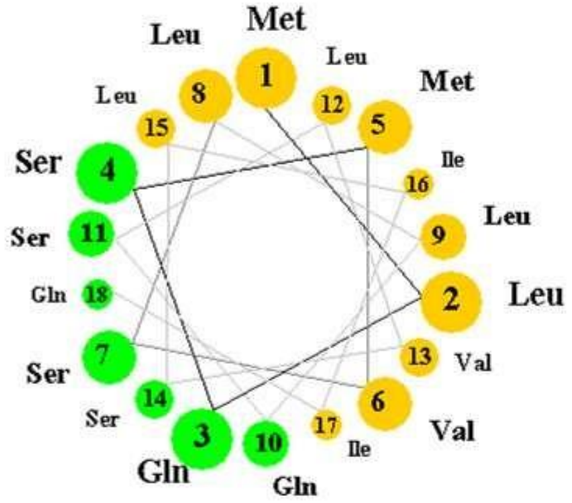
For bacteria and mitochondria, the transmembrane domain is made of beta strands



Single alpha helix multiple helices



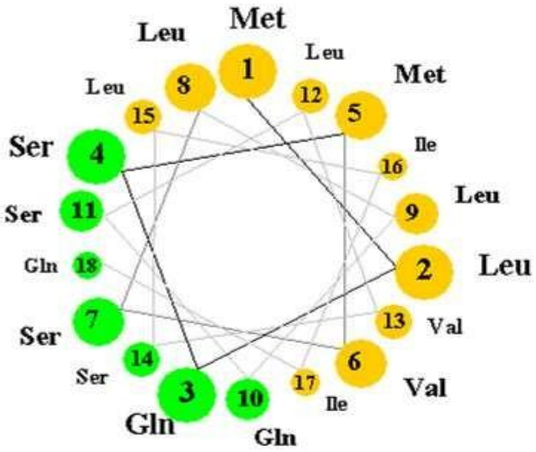




This slide is to understand why it is amphipathic

α-helices as transmembrane domains

- Ions and hydrophilic substances cannot pass through the membrane because it is hydrophobic, so they pass via channels.
- Channels are made by transmembrane protein alpha helices.
- If these helices were purely hydrophobic, Ions and hydrophilic substances would not pass.
- If they were purely hydrophilic, the protein would not be stabilized in the membrane
- Therefore, these helices are amphipathic (hydrophilic at the inner side toward the opening of the channel and hydrophobic at the outer side toward the membrane)



Quaternary structure

Primary, secondary, and tertiary structures are present among all proteins, but quaternary structure exists only in proteins with more than one polypeptide chain.

What is it?

- Proteins have a **quaternary** structure if they are composed of **more than one polypeptide chain.**
  - They are oligomeric proteins (oligo = a few or small or short; mer = part or unit)
- *A quaternary structure Is the spatial arrangement of subunits and the nature of their interactions.*

Polypeptide chains

Usually, we name proteins with large numbers of subunits multimeric proteins like **Pyruvate dehydrogenase complex enzyme**.

- A protein can be a:

They don't have quaternary structure →

  - Monomer: One subunit
  - Dimer: Two subunits
    - The simplest: a homodimer
  - A trimer: Three subunits
  - A tetramer: Four subunits
  - ...etc

Like insulin receptor , consisting of 2 α and 2 β subunits

- Each polypeptide chain is called a subunit.
- Oligomeric or multimeric proteins are made of multiple polypeptides that are
  - identical → homooligomers (homo = same)
  - different → heterooligomers (hetero = different)

If it is a heterooligomer, we designate the subunits by greek letters α,β,γ,δ,ε,... etc.

For example, Gproteins are heterotrimers (they have 3 subunits α,β,and γ)

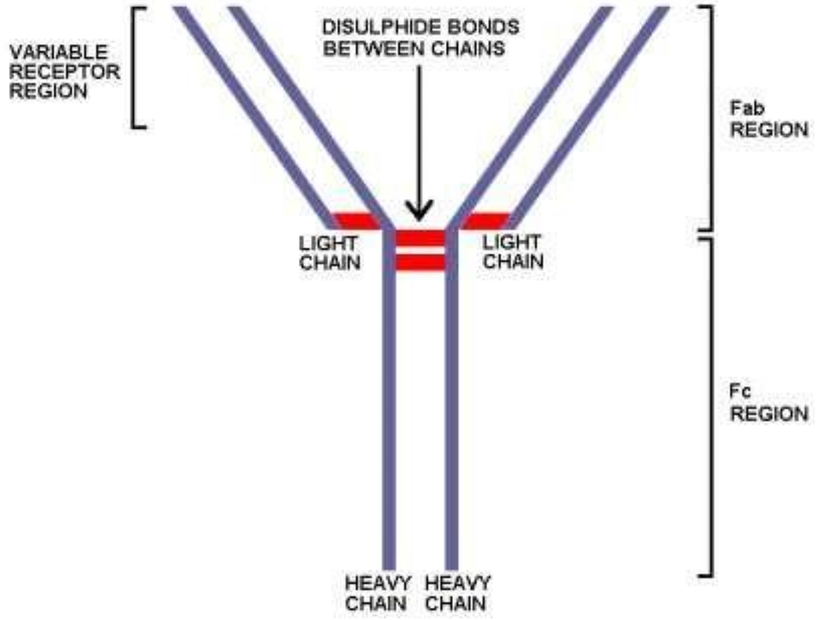
How are the subunits connected?

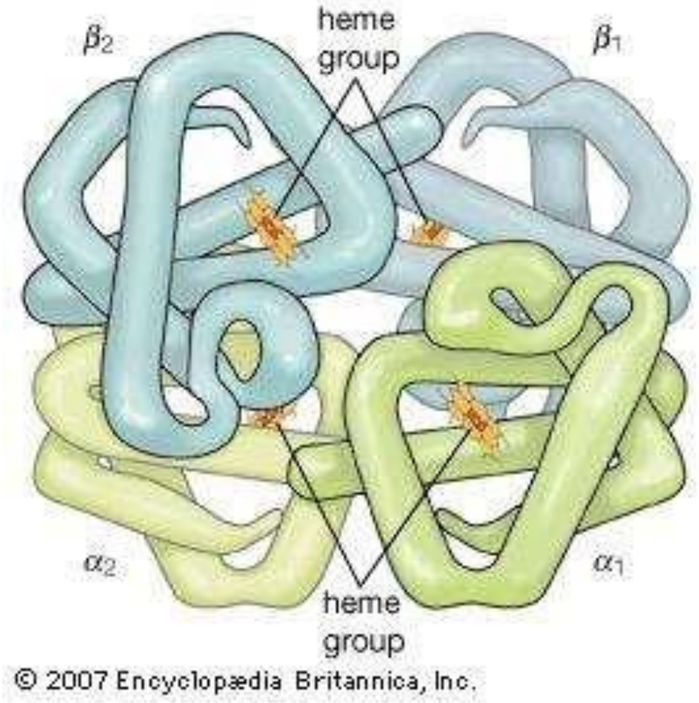
- Sometimes subunits are **disulfide-bonded together**, other times, **noncovalent bonds** stabilize interactions between subunits

Covalently

- Covalently (disulfide bonds): Immunoglobin (antibody)
- Non-covalently: Hemoglobin

We are going to study these proteins in details next lectures





Organized protein structures

Super-secondary structures

- Proteins contain **regions with an ordered organization of secondary structures.**
  - Motifs
  - Modules
  - Domains
  - Folds

There are differences between books about the classification of these structures, so we will not talk about the classification and specific differences between them.

Features of super-secondary structures

- They are distinct 3-dimensional structures.
- They are composed of **multiple secondary structures** that are compact in space.
- They can be small or large.
- In relation to the primary structure of proteins, the super-secondary structures can be recurring and continuous or separated by other structures or come from different polypeptides.

See next slide for further explanation.

- **If they are continuous and located on the same polypeptide,** they can fold independently of the rest of the protein.

Even if it was separated from the protein, it stays folded and functional

- They can be shared among several proteins with similar function and/or structure
- They are linked to a specific function:
  - Enzymatic activity
  - Binding ability (e.g., a DNA-binding domain)

For example, ATP binding proteins have the same binding domain (they might have different structures and functions, but this domain is similar among them)

Also, enzymes that have similar functions (on different substrates) have similar domains

Features of super-secondary structures

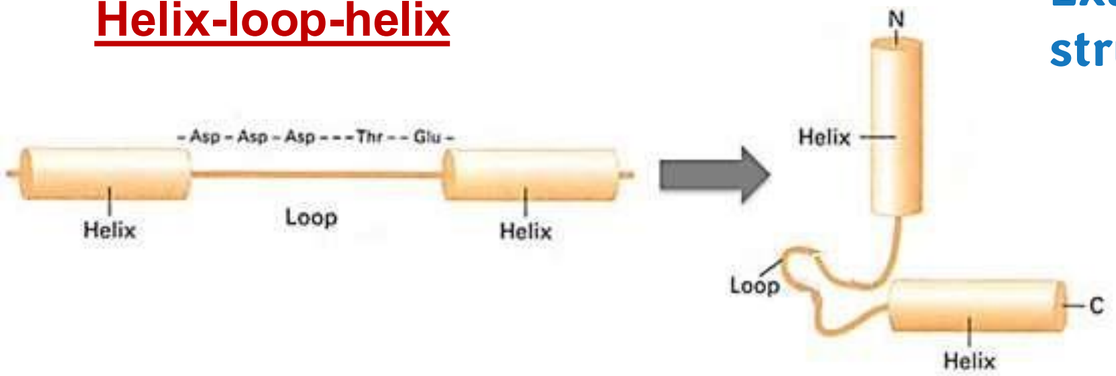
- In relation to the primary structure of proteins, the super-secondary structures can be recurring and continuous or separated by other structures or come from different polypeptides.

For example:

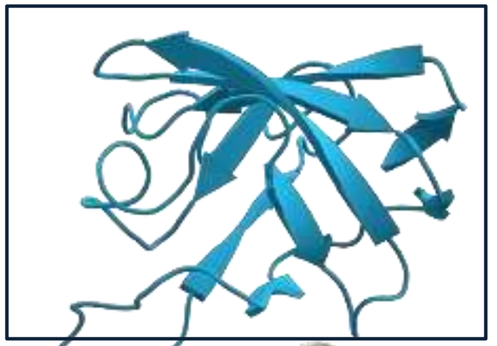
They may contain multiple alpha helices connected via loops or turns (continuous)(consecutive).

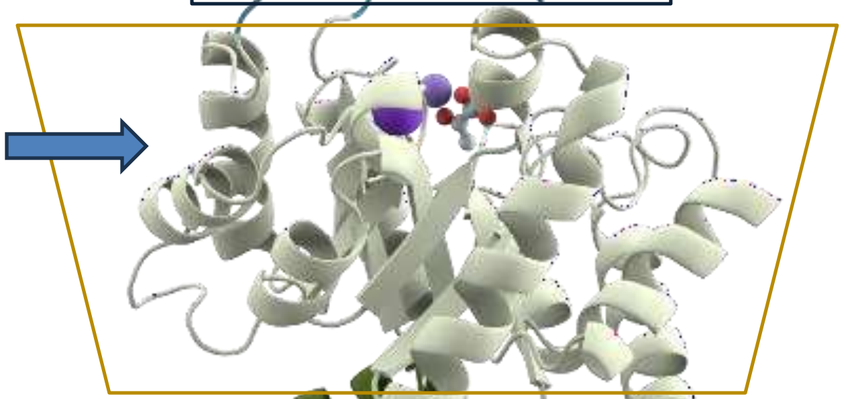
Or secondary structures far from each other when talking about primary structure (polypeptide sequence), but they become near to each other due to protein folding (separated)

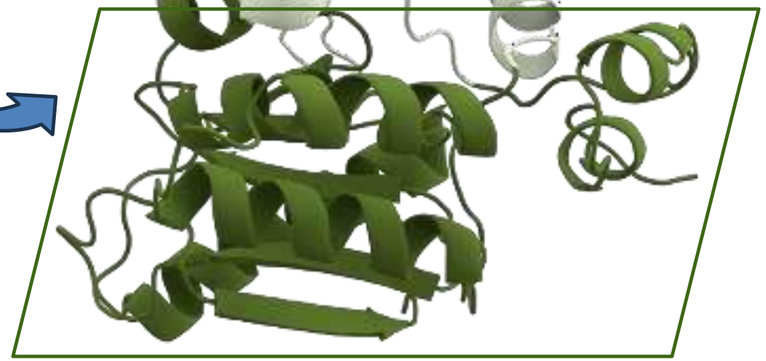
Helix-loop-helix



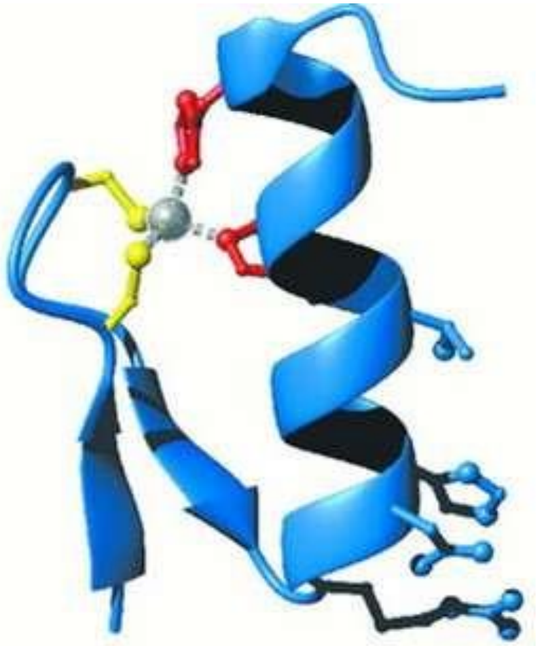
Examples of super-secondary structures





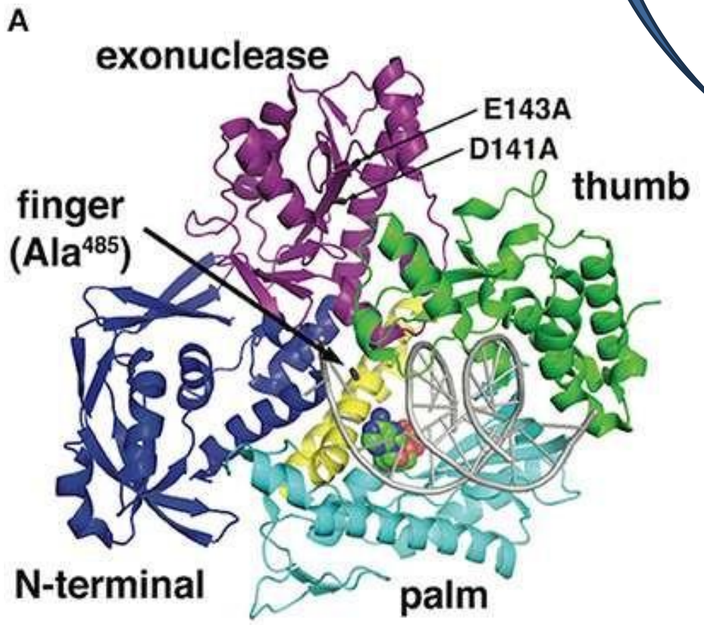


Again, even if it was separated from the protein, it stays folded and functional (independent)



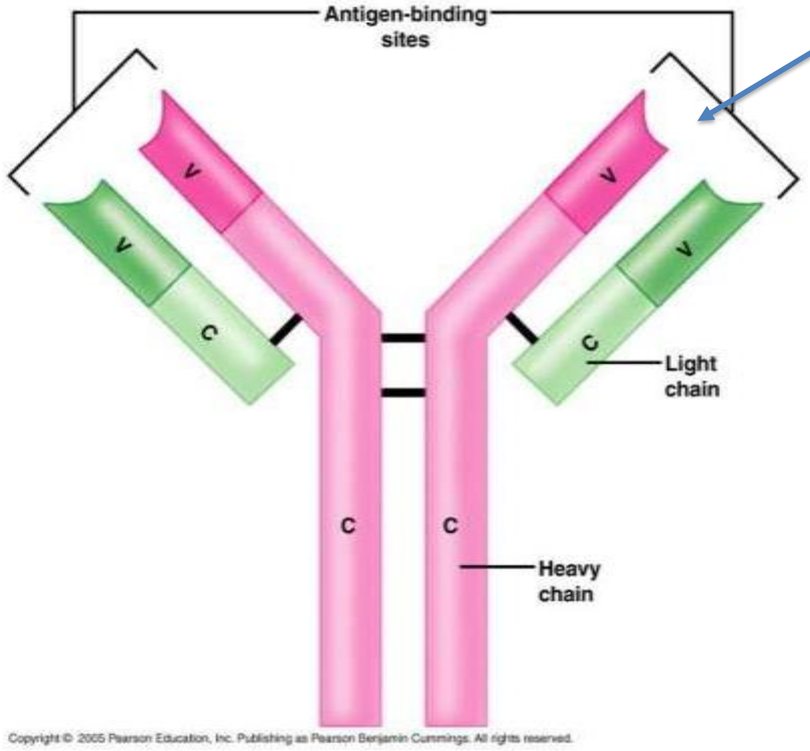
Zinc finger domain: Two beta strands with an alpha helix end folded over to bind a zinc ion. **Important in DNA-binding proteins.**

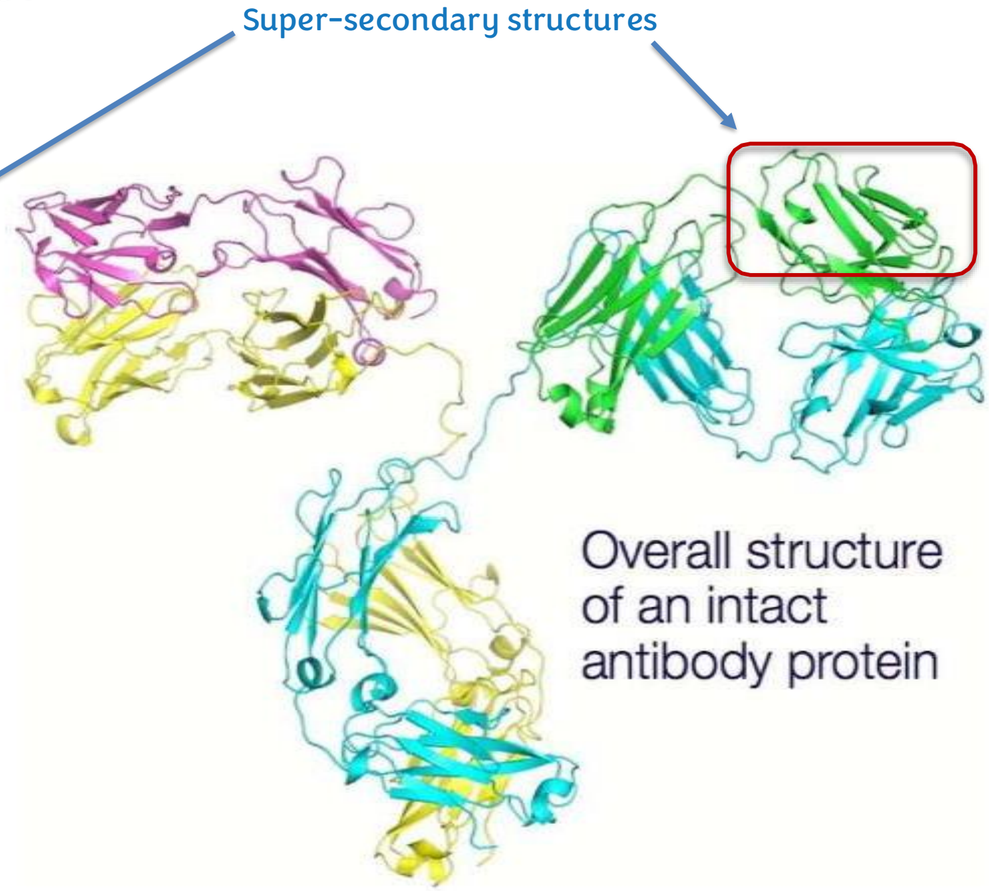
Nucleotide-binding fold

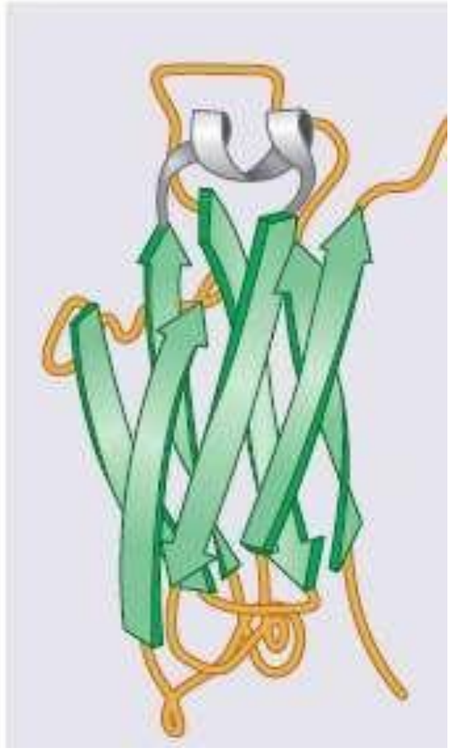


A more complex motif/domain/fold is…

- The immunoglobulin fold that enables interaction with molecules of various structures and sizes.







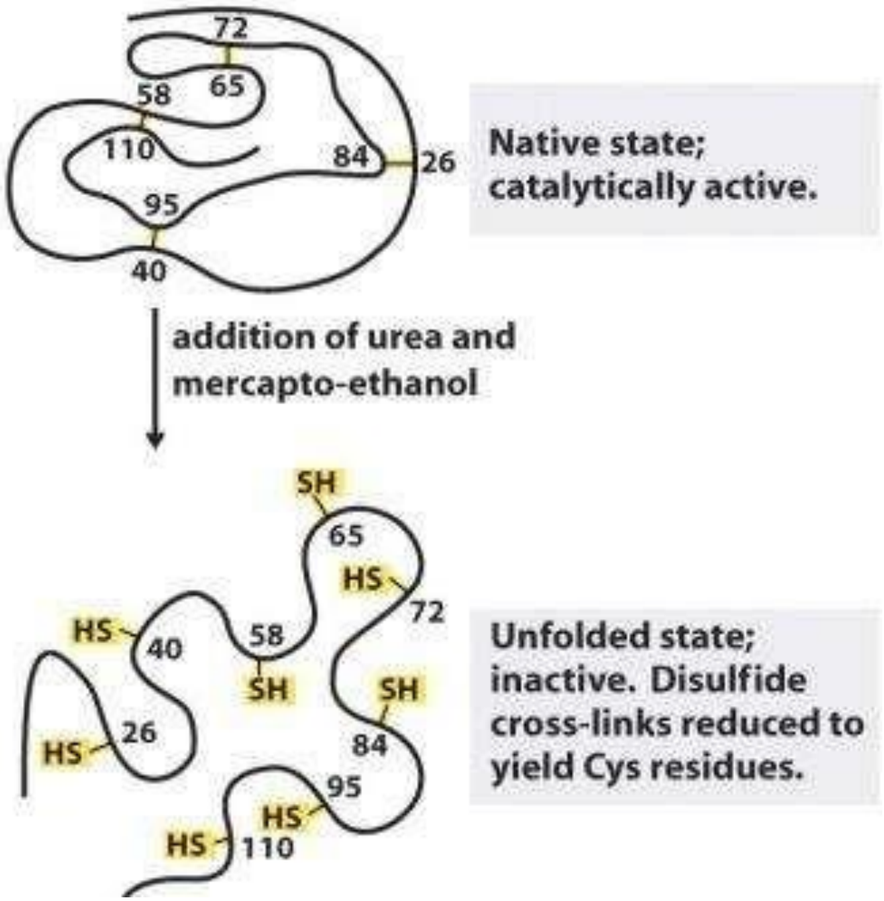
Properties of Proteins:

Denaturation and Renaturation

Denaturation The unfolding of proteins

- Denaturation is the **disruption of the native conformation of a protein via breaking the noncovalent bonds that determine the structure of a protein**
- **Complete** disruption of tertiary structure is achieved by **reduction of the disulfide bonds in a protein** Reduction here = breaking the disulfide bond
- The denatured protein loses its properties such as activity **and become insoluble.**

Due to the exposure of non-polar amino acids because of unfolding, which causes adhesion between protein molecules by hydrophobic interactions, forming a hydrophobic clusters



When we boil an egg, the albumin is denatured due to heat

Denaturing agents

1. Heat: disrupts low-energy van der Waals forces in proteins

Due to the fast movement of electrons when increasing temperature

2. Extremes of pH: **change in the charge of the protein's amino acid side chains** (electrostatic and hydrogen bonds).
3. Detergents: Triton X-100 (nonionic, uncharged) and sodium dodecyl sulfate (SDS, anionic, charged) disrupt the hydrophobic forces.

Or sodium lauryl sulfate

   SDS also disrupt electrostatic interactions.
4. Urea and guanidine hydrochloride disrupt hydrogen bonding and hydrophobic interactions.

5. Reducing agents: β-mercaptoethanol (β-ME) and dithiothreitol (DTT)

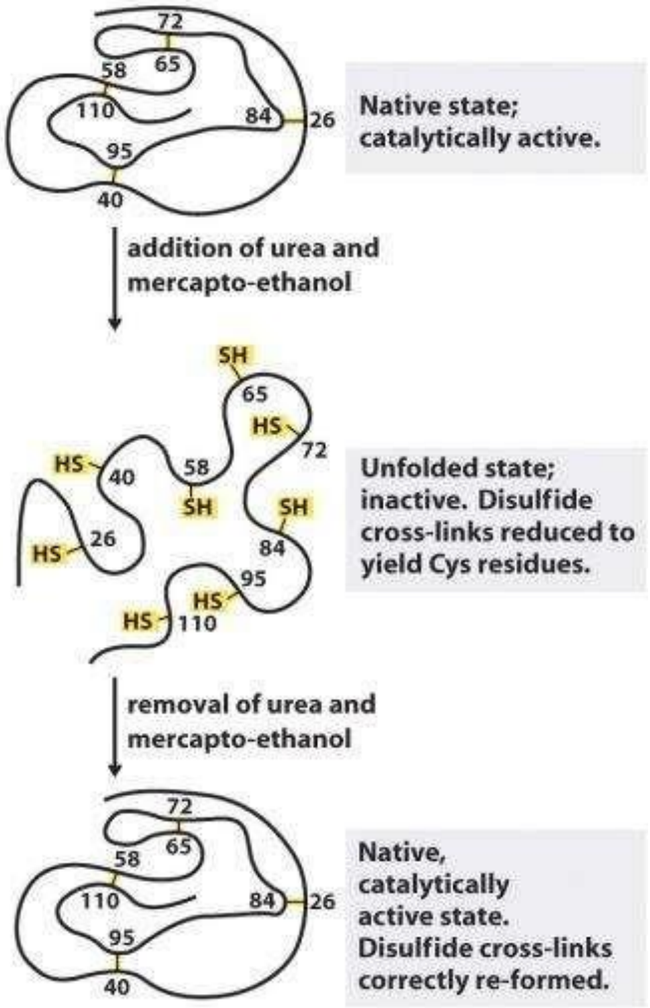
   Both reduce disulfide bonds.

No complete denaturation without the presence of reducing agents

Renaturation

- Renaturation is the process in which the **native conformation of a protein is re-acquired.** It returns spontaneously when removing the denaturing agents (Because its native conformation has the lowest energy)
- Renaturation can occur **quickly and spontaneously**, and disulfide bonds are formed correctly.
- If a protein is unfolded, it can **refold** to its correct structure placing the S-S bonds in the right orientation (adjacent to each other prior to formation), **then the correct S-S bonds are reformed.**
- This is particularly true for **small** proteins.

Usually, when we put a reducing agent, the protein remains functional. Because the disulfide bond acts as a stabilizer in most cases (it doesn't determine the 3D structure it stabilize it)



Factors that determine protein structure

- The **least amount of energy** needed to stabilize the protein. This is determined by:
  - The amino acid sequence (the primary structure), mainly the internal residues.
  - The proper angles between the amino acids
    - The different sets of weak noncovalent bonds that form between the mainly the R groups.
    - Non-protein molecules.

Quiz!

Good luck!

For any feedback, scan the code or click on it.

Corrections from previous versions:

Versions	Slide # and Place of Error	Before Correction	After Correction
V0 → V1			
V1 → V2			

Additional Resources:

Reference Used:

Prof. Mamoun Ahram recorded lecture

Extra References for the Reader to Use:

Marks’ Basic Medical Biochemistry (6th edition)

رسالة من الفريق العلمي: