University of Jordan
Faculty of Medicine

Medical Committee
The University of Jordan

Introduction to

BIOCHEMISTRY

Lecture #: (12)

■ Sheet ☐ Slides ☐ Other

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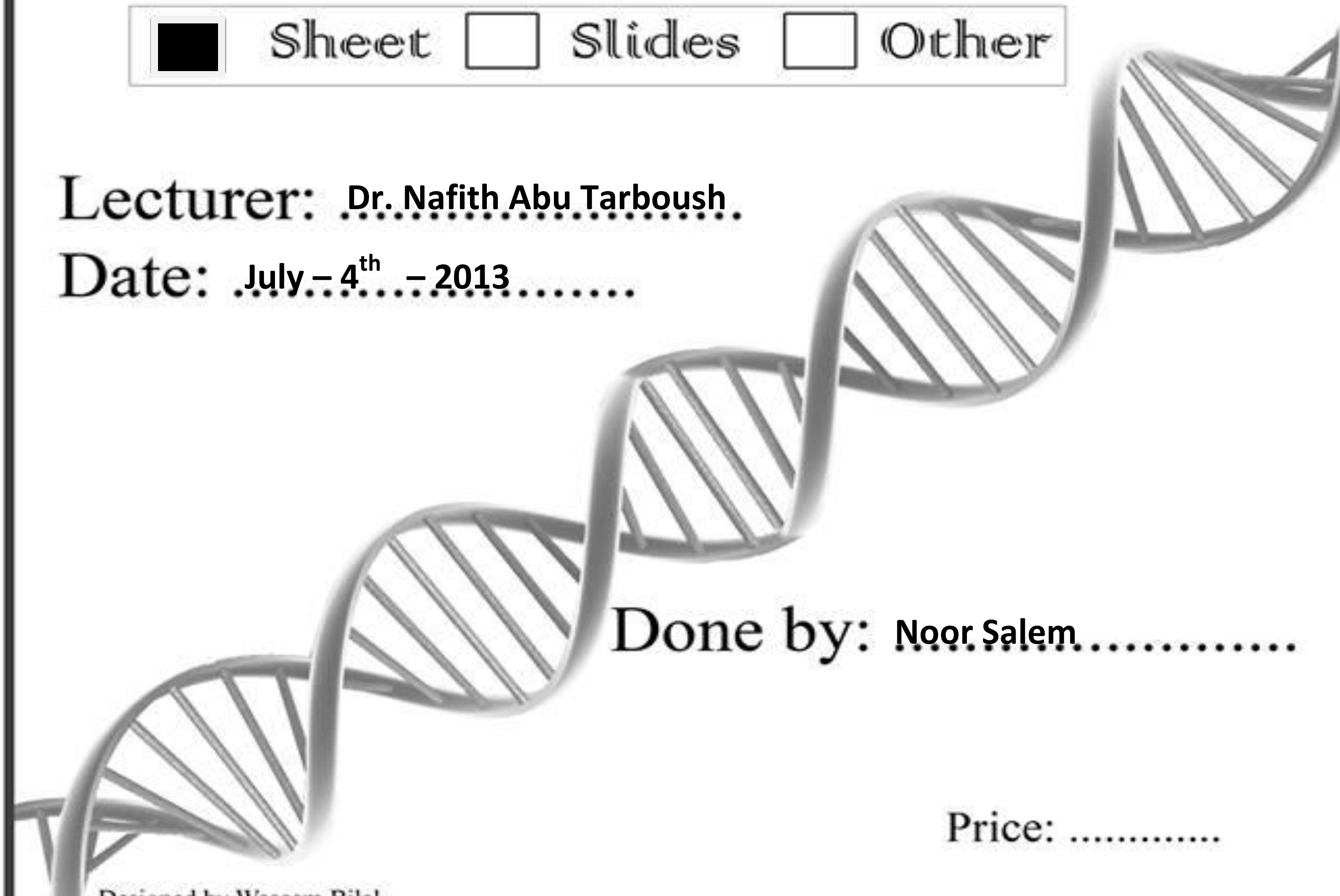

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Review :

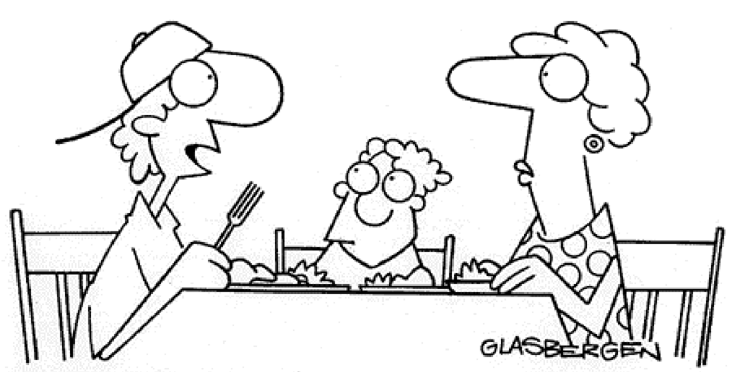
- **Secondary Structure :** The 3D arrangement of **close** amino acids together made through hydrogen bonding between the backbones of these amino acids.
    - α - Helix
    - β - Sheet
- **Tertiary Structure :** 3D arrangement of **all** molecules (hydrogen bond between side chains)

- **Are there other regular secondary structures?**

  Yes, not very complex ones and they are :
    - **β - Turns** (β - Reverse Turns or β - Bends)
        - Connects two helices or two sheets (2 secondary structures).
        - Composed of **4 amino aci**ds connected by a **hydrogen bond** making a **sharp** turn to **reverse** the direction of the chain (To fold structure) (hydrogen bond between the first and the forth amino acid)
    - **Loops** (Ω - Loops)
        - Just like β - Sheets only with **more than 4 amino acids** and a longer structure.

  There are also more complex ones called **Super-secondary Structures**, also known as **Motifs.**
    - Super-secondary structures can be defined as combinations of alpha-helices and beta-structures connected through loops, that form patterns that are present in many different protein structures.
    - **Domains**
        - Motifs with a **certain function** and they perform the exact **same** function **regardless** of their location.
        - Important in **predicting** how proteins work.
        - **Example :** Leucine Zipper
            - DNA-binding protein and that is its <u>**only**</u> function *wherever it is found*.
            - Two α helices connected together in the shape of **scissors** to surround DNA from both sides.
            - There are **hydrophobic interactions** binding them together in the **middle** (2 leucine molecules)
    - There are others that you must know **only by name and know that they are super-secondary structures**, nothing else.
        - βαβ
        - β - Meander
        - αα Unit (aka Helix-Turn-Helix)
        - β - Barrel : Composed of β sheets
        - Greek Key : Looks like a Greek Key.

- **Secondary structures** are sequences of amino acids that have formed α-helices or β-sheets.
    - Some of them **are composed solely of one type** of secondary structure while others are **mixed** and that is related to function.
        - **Fibrous Proteins**
            - Imagine a structure made entirely of α-helices by thinking of springs overlapping to form a compact structure. And of β-sheets by cards stacking over one another.
                - They all have the same shape so they can be combined and water will be **expelled** from all binding locations making them **hydrophobic and tight.**
                - Those form **bundles** and usually have **structural functions** and they don't need to engage in the body's reactions *(Remember they're insoluble because they don't have open side chains to react with water)*.
                - Examples :
                    - **Collagen** *(Controversial choice because it looks like an α-helix but actually isn't so the professor didn't want to use it as an example)*
                    - **Keratin**
                        - Composed of **α-helices**
                        - **Found in :**
                            - Nails
                            - Hair
                            - Outer Layer of Skin
                            - Sheep's Wool
                    - **Fibroin**
                        - Composed of **β-sheets**
                        - **Found in :**
                            - Silk
                            - Spider Webs
        - **Globular Proteins**
            - Look like a globe
            - Usually contain **more than one type** of secondary structure .
            - Adding an α-helix to a β-sheet doesn't form a well-packed structure, spaces remain.



"Everyone in my biology class voted against dissecting a frog. But we almost had enough votes to dissect the teacher!"

    - They are **soluble** due to the spaces that **allow water in** and that helps them move through blood and extracellular matrix.
    - The **hydrophilic** amino acids arrange themselves to be on the **outer surface**.
    - The **hydrophobic** ones coil **inside,** thus giving the protein a globular structure
    - Important for **transport and storage**.
    - **Examples :**
        - **Myoglobin**
            - **One** single polypeptide chain
            - **8 α-helices** but with many loops and turns.
            - **153** amino acids

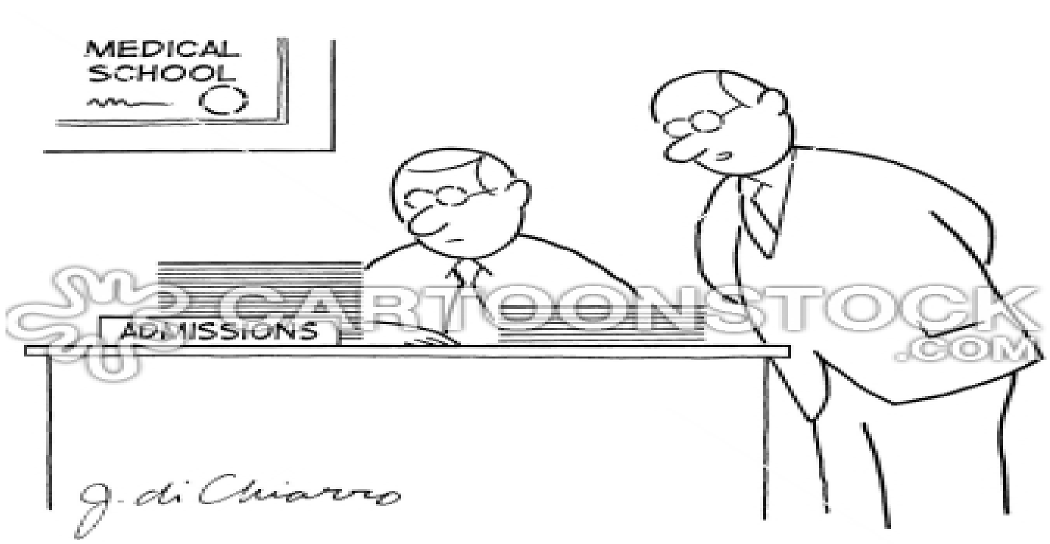
- **Polypeptide Chain :** A sequence of amino acids which has one free amino group and one free carboxylic group (Translated from one gene)
- Some proteins are composed of only one polypeptide chain coiled in a 3D manner, this is what is known as **tertiary structure.**
    - Defined as the *3D arrangement of all atoms in the molecule*.
    - Bonds involved : **Any** bond can be involved in this structure depending on type of amino acid.
        - Covalent (Disulfide Bridges) → From Cysteines
        - Hydrogen Bonds
        - Ionic Interactions
        - Hydrophobic Interactions (Pockets)
        - Van der Waals Forces
        - Metal Coordination
- When you have more than one **already-coiled** polypeptide chain (each one called a supplement) joining each other to form one structure, that is called a **quaternary structure.**
    - Must have **at least 2** polypeptide chains.
    - Does **not** include covalent bonds. **Only** non-covalent.
    - **Examples :**
        - **Hemoglobin**
            - **4** polypeptide chains
            - Each chain was made on its own (the 2 αs were made on their own, same to the two βs)
            - Had a **Heme** added to them which made them "stick" to each other through non-covalent interactions.
            - **Transports oxygen**
            - **First** structure to recognize through X-Ray Crystallography
- <u>**Proteins (Regardless of if they're 3^o or 4^o) can be divided into :**</u>
    - **Simple**
        - A coiled and folded sequence of amino acids with a certain function
    - **Conjugated**
        - **Must** have a non-protein part added in order to function.
        - *Hemoglobin* and *Myoglobin* are examples as they can't function without a **Heme.**
            - Heme is not an amino acid encoded by a genetic code but made by enzymes in the body.

- **<u>How to determine a protein's final shape and structure?</u>**

  There are 2 main techniques and they are usually used together to get a full picture of the protein :

  - **X-Ray Crystallography**
    - Proteins usually exist in solutions or buffers but they have the ability to **form crystals when dehydrated.**
    - A plastic plate with "wells" is covered by a plastic cover slip.
    - One drop of the protein solution is put on the cover slip (The drop must be opposite the well) and it is left over time (With special crystal-related solutions).
    - Water evaporates slowly allowing the protein to **preserve** its bonds' structure and shape.
    - The crystal formed is put under an X-Ray beam and the protein's shape is shown.
      - Each atom has a different shape because of the **different electrons around it.**
    - The X-Ray's results are inserted into a computer program which includes a lengthy procedure with Fourier's equation to tell you what each atom represents in a 3D manner.
    - Most of the photos used in the slides are from this technique.
    - **Pros** : Gives the *3D* structure of a protein and gives a better representation *(More accurate*).
    - **Cons :** Its results are of proteins in a solid state with fixed bonds (Much like freezing) which *doesn't represent real life* since proteins move in solutions and can have conformations.
  - **Nuclear Magnetic Resonance** (NMR)
    - Resembles MRI.
    - A force is projected onto atoms to make them **vibrate** in order to know their shape.
    - **Pros :** Can be performed while the protein is in a solution allowing us to observe the protein's *active* state.
    - **Cons :** Gives a *2D* structure.

  So the best thing is to use both techniques to determine the structure .



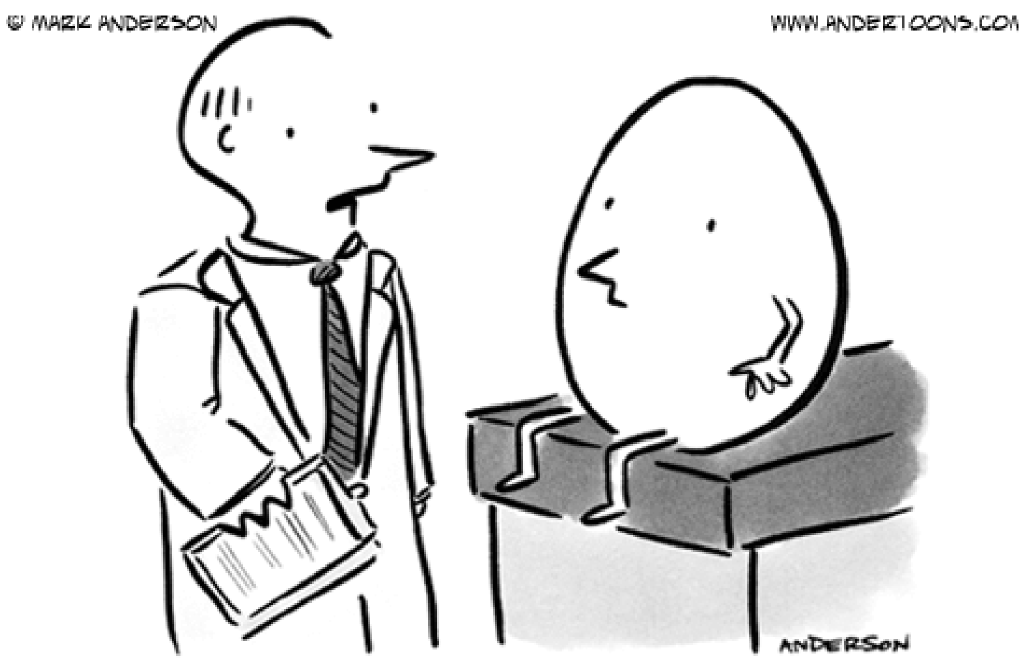
"Just show me the applications with bad handwriting."

- **<u>Proteins can be complex</u>** (Added to non-protein parts)
    - **Glycoproteins**
        - **Carbohydrates** can link to
            - **Nitrogen**
                - Called **N-Linked**
                - N usually comes from *Asparagine*
            - **Oxygen**
                - Called **O-linked**
                - O usually comes from *Serine*, *Threonine* and sometimes *Tyrosine* (Usually it's engaged in other active processes) or *Lysine* if it has been hydroxylated.
    - **Lipoproteins** (With lipids)
        - Chylomicrons (Least Dense)
        - VLDL
        - LDL
        - HDL
    - Complex Lipid-Protein-Carbohydrate
        - **ABO blood grouping** : Glycosphingolipids
            - Protein connected to **sphingosine** with **ceramide** then connected to **4** carbohydrate residues with **Fucose.**
            - If it connects to **N-Acetyl-Galactosamine**, it will be from the **A** type.
            - If it connects to **Galactose**, it will be from the **B** type.
            - If it **doesn't connect anything**, it will be from the **O** type.

be **phosphorylated** in order to activate or deactivate them.

- P is usually added to the hydroxyl groups of *Serine, Threonine* and *Tyrosine*
- **Kinase :** A protein that adds P to any structure (doesn't mean activation)
- **Phosphatase :** The enzyme that removes the P from any structure

- <u>**All proteins share chemical properties**</u>
  - **Hydrolysis**
    - The **reverse** of protein synthesis
      - Protein synthesis (Dehydartion) : Condensation reactions that includes joining two amino acids through removing the H from one and the OH from the other and making an amide (Peptide) bond.
    - Done by **adding water** to break the peptide bonds with the aid of an enzyme
      - Hydroxyl group is added to carboxylic group
      - Hydrogen is added to amino group
    - **Catalysts (**Hydrolytic Enzymes) must be added to **speed up the process**
      - Pepsin
      - Trypsin
      - Chymotrypsin
      - Carboxypeptidase A & B
  - **Denaturation**
    - The loss of the 3D (higher) structure of the protein = Getting back to the primary (**Linear**) structure (Arrangement)
    - Does **not** affect peptide bonds but it affects any bonds involved in shaping the protein.
    - Mostly **irreversible** but some proteins can return to its original shape
    - Caused by :



"Blood pressure's fine, reflexes are normal, and your cholesterol is sky high. You're the picture of health."

- **pH change**
    - **Disrupts salt bridges** (Ionic interactions) by **changing the charge** on the amino acids.
- **Heat**
    - By **increasing the kinetic energy of atoms** which decreases bonds between molecules, making them **susceptible to breaking.**
    - Solidifies the structure because the proteins (Especially globular ones) get their **hydrophobic parts exposed** to the water (After the return to linear arrangement) which **decreases their solubility**, allows more hydrophobic interactions and making them **stack** over one another.
    - Heating over 50 degrees denatures most proteins.
    - Happens during boiling or frying eggs.
- **Mechanical Agitation**
    - Whisking, beating or quickly moving a protein causes many of its bonds to **break**.
    - The more bonds break, the more **hydrophobic areas are exposed** to water, causing the protein to **solidify**.
    - Happens during making whipped cream or meringue.
- **Alcohol and Detergents**
    - Used in hospitals as an **antiseptic** because it **denatures bacteria.**
    - Have lots of OH groups which bind with the protein, **weakening** the already existent bonds.

- **<u>Protein Folding and Prediction</u>**
    - A sequence of amino acids' folding pattern and function can be predicted through **bioinformatics**
        - The science of predicting where amino acids sequences are put on an on-line software to see what other proteins it will match through comparing sequences.
        - **Homology :** Sequence of similar
            - If you have a homology of 25% or 30% then they are mostly of the same group of proteins.
- **<u>How do proteins fold?</u>**
    - Proteins are translated then they leave the ribosome but most **cannot** fold in the extracellular environment on their own, they need other proteins to help them.
    - Those proteins which help other proteins fold are called **Chaperones**.
        - **HSP70** was the *first* protein to be recognized as a chaperon (most common one)
- **<u>Diseases linked to protein folding</u>**
    - **Mad-Cow Disease** (Bovine Spongiform Encephalopathy)
        - Bovine = Cow / Spongiform = Turns the brain into a spongy state / Encephalopathy = Affects the Cephalon in the head.
        - The **human** form of the disease is called **Creutzfeldt–Jakob disease**.
        - A protein named Prion has a sequence of amino acids that gets disrupted at **Methionine 129** by mutating to another amino acid, regardless of what the other amino acid is.

- This mutation affects the folding of the protein by changing it from α**-helices connected by loops** to a **4 β-sheets** which causes the tissue to **stack** in the brain.

The mid-term exam is till here

- **Alzheimer's**
  - Loss of memories
  - A Protein called **Amyloid** is **mis-folded** which gives the body an error message causing it to **break it into AP proteins (breaks down to amino acid sequences each of 40 amino acids)**
  - The AP proteins begin to stack and **accumulate in the nervous system**

- **<u>What is Heme?</u>**
  - **Macro-cyclic** structure which has **4 cyclic nitrogenous** rings (hexocyclic structure), all of them linked to an **iron metal** in the middle.
  - Iron can make **6 bonds**. In the Heme, it is already engaged to 4 Nitrogens and most of the time it has 5 (Connected to an amino acid, usually it's the cyclic structure of Histidine) called (5 coordinate heme) or 6 (called 6 coordinate heme) (Usually kept empty for transport of Cyanide, O, Azides or CO) bonds depending on the function.
    - If the function is **electron transport**, then it **doesn't matter** if the Iron doesn't have all bonds connected , usually 6 coordinate heme
    - If the function is **transporting materials** like O, then one bond **must be empty** for the material to attach , usually 5 coordinate heme
  - Hemes differ in what they are attached to the cyclic structure
  - **Heme B** *(Most common)* exists in Myoglobin and Hemoglobin.
  - **Heme C** exists in enzymes in the internal membrane of the mitochondria (Cytochrome C)
  - **Cytochrome A** has **Heme A3**
  - Histidine on the 5th bond is **very close** to the 6th attachment spot so when a material attaches, it doesn't attach vertically but in an **angle** which decreases the affinity with iron so it can disconnect to move to its destination.
  - Only Fe^{+2} (Reduced state) can connect to other molecules because Fe^{+3} (State after connecting to O) has **very low affinity** to other molecules (Connected to the **Methemoglobin disease** : People with it don't have an enzyme that reduces Hb so it can't connect to oxygen)