About this Course:

In this course, we will study structure-function relationship, plasma proteins and energy metabolism with dr Nafith, carbs metabolism (from carbs to energy) with dr Nayef, lipid metabolism (from fatty acids to energy) with dr Faisal, and then protein and nucleic acid metabolism with dr Diala, and finally obesity, vitamins, nutrition and other common topics in metabolic processes with dr. Nafith

Here you can find Dr. Nafith’s slides: [http://eacademic.ju.edu.jo/n.abutarboush/Material/Forms/AllItems.aspx](http://eacademic.ju.edu.jo/n.abutarboush/Material/Forms/AllItems.aspx)

A brief introduction:

Generally, Food conversion will produce acetyl CoA at the end. All macromolecules would become a common molecule, this common molecule is called **Acetyl CoA**.

Carbs, Lipids, Proteins and Nucleic Acids will convert to acetyl CoA. Let’s take Carbs as an example, Carbohydrates can be broken down to glucose, glucose to pyruvate and then pyruvate will turn to acetyl CoA.

Acetyl CoA will go to Krebs cycle, this is called energy metabolism, and energy metabolism is common to all macromolecules.

Krebs cycle has two other names
- a- Citric acid cycle
- b- Tricarboxylic Acid cycle

Blood:

Let’s start:
- In the first two lectures, we are supposed to answer the following questions:
  o What is plasma, and how can we extract it?
  o What are the different components of plasma?
  o Plasma proteins’ (general functions, basis of classification, associated processes and molecules)
  o Plasma proteins’ (structure, synthesis, function & diseases associated)
- **Blood components:**
  - Blood components are plasma and cells:
    - **Plasma** forms 55% of the blood volume
    - **Cellular components** (RBC, WBC, and platelets) form 45% of blood volume
      - WBC (white blood cells), also called leukocytes:
        - Granulocytes: Neutrophils, Eosinophils, and Basophils
        - Agranulocytes: Lymphocytes and Monocytes
      - RBC (red blood cells), also called erythrocytes
      - Platelets

The previous underlined examples were not mentioned by Dr. Nafith, but they are found in the slides.

- When the blood is separated by its components in a test-tube, the components from the bottom of the tube to the top of it, would be:
  - RBCs - *bottom*
  - WBCs and Platelets
  - Plasma (the liquid, non-cellular part) – *the upper part.*

- **Extraction**

  But how could we extract the plasma from the blood?

  - By Centrifugation
  - More importantly, by Purification (ESR: Erythrocyte Sedimentation Rate)

  By two techniques, we can extract plasma proteins:

  - Salting out:
    - Proteins are water soluble, when adding salt (ammonium sulfate for example), protein solubility will decrease until it precipitates, the more soluble the protein is, the higher the concentration we need to extract the protein.
    - By this technique, we can extract: fibrinogen, albumin, and globulins

  - Electrophoresis (gel electrophoresis):
    - In this technique, we cannot use plasma directly, because of fibrinogen, when fibrinogen is exposed to air, it turns directly into fibrin. This is why cutting your finger,
leads to clot formation (fibrinogen is a clotting factor). Instead of plasma, we can use serum which is a defibrinated plasma. (to make it easier, serum = plasma – fibrinogen)

When we bring a blood sample, and leave it approximately for an hour, the cellular components will start to precipitate, but it is important to say that this process may take either more or less than an hour in some pathological cases (diseases) or physiological process.

The ESR was previously more common, when it is fast, it can indicate certain diseases and physiological processes (such as pregnancy), but when it takes longer time, it can be an indicator to other diseases and physiological processes, but now we can use direct, specific tests for certain diseases or physiological processes.

❖ More details about electrophoresis

- when putting plasma proteins in the gel, they would move from negative electrode to positive electrode, albumin is the fastest (this is why it has the highest negative charges), then a group of globulin proteins (alpha, beta, gamma) comes next.
  
  Note: Albumin is smaller than globulin, and slightly negatively charged

- resulting bands:
  - Albumin (one large band)
  - Globulins (3 main bands):

  α band:
  - this band separates into two bands, Globulin Alpha1 and Globulin Alpha2, they were both called alpha because they are close to each other in molecular weight. It is important to know that Alpha1 is not a single protein, it is a group of proteins & Alpha2 is a group of proteins, too.
    1. α1 region consists mostly of α1-antitrypsin
    2. α2 region is mostly haptoglobin, α2-macroglobulin, & ceruloplasmin

  β band: Beta proteins are also a group of proteins, and if they were given enough time, they might separate into Beta1 and Beta2. (transferrin, LDL, complement system proteins)

  γ band: the immuno-globulins
  Gamma Globulins- are antibodies, immunoglobulins (IgG, IgD, IgE, IgM and IgA)

- By this technique (electrophoresis), we usually get five bands which are (albumin, globulin α1 , globulin α2, globulin β, and globulin γ) -from the least molecular weight to the most-
Plasma

- **Its definition** (liquid part of the blood where cells are suspended, it is composed of 92% water and 8% solids -plasma proteins in particular-)
- **What can we find in plasma?** We can find **everything** (except cellular components), such as:
  - Gases, wastes (such as nitrogenous wastes), Amino acids, Electrolytes, ions , proteins (albumin, globulin and fibrinogen), enzymes, hormones, Nutrients, Ketone bodies, Carbs as monosaccharides, fatty acids, nucleotides and many other stuff, it is simply a reflection of the person
- We can put “**Everything**” into two main groups:
  - Organic
    - Plasma proteins: Albumin, Globulins & Fibrinogen
    - Non-protein nitrogenous compounds: urea, free amino acids, uric acid, creatinine, creatine & NH₃
    - Lipids: Cholesterol, TG, phospholipids, free fatty acids
    - Carbohydrates: Glucose, fructose, pentose
    - Other substances such as: Ketone bodies, bile pigments, vitamins, enzymes & hormones
  - Inorganic: Na⁺, K⁺, Ca²⁺, Mg²⁺, Cl⁻, HCO₃⁻, HPO₄²⁻, SO₄²⁻

**Plasma Proteins**

- We are gonna study, the following six major groups in plasma:
  - Albumin & pre-albumin
  - α₁-antitrypsin
  - Haptoglobin (Hp)
  - α₁-fetoprotein (AFP)
  - Ceruloplasmin
  - C-Reactive Protein

- But 500 plasma proteins have been identified, some are not detectable in healthy people, but could rise and become detectable in certain diseases

- The normal range of plasma proteins 6-8 grams/ deciliters (Remember : a deciliter = 100 milliliter / a deciliter= 10⁻¹ liter)

- **General properties**
  - All plasma proteins can be synthesized in **liver except** gamma globulin, gamma globulins are synthesized in B cells /plasma cells; lymph nodes, bone marrow, spleen)
  - Most plasma proteins are synthesized as preproteins (signal peptide), meaning plasma proteins in liver are inactive, because they won’t work there, what happens is that a single peptide gets out of liver cells and then to blood where it becomes active, this is a property that all plasma proteins synthesized in liver share,

*preproteins need to undergo posttranslational modifications to become active*
adding a molecule in a process called glycosylation or phosphorylation, other modifications are proteolysis or breaking the protein to become fully functional (such as albumin)

- Some plasma proteins need short time to move from liver cells to blood, others need more time and this is called transit time.

- Simple & conjugated proteins (glycoproteins & lipoproteins)

- Most plasma proteins attached to carbs, in other words, most plasma proteins are Glycoproteins (N- or O-linked). Albumin is the major exception, and this has a clinical significance, because if albumin was bound to a carb molecule, this would increase blood’s viscosity/ thickness (which makes it harder to move), because albumin is already negatively charged and carbohydrates add to its negativity and it is the more abundant plasma protein.

- Plasma proteins have different half lives, it could be determined through isotope labeling studies ($^{131}$I) by injecting the person with a protein of interst, which is radio-labelled (fluorescence for example) to be easily traced, and then a sample is taken to see how much is left

- Albumin has a half life of 20 days
- Haptoglobin has half life of 5 days (if not bound)
- This half life of proteins might be affected by many factors,

  I. Diseases can affect half-lives (ex. Crohn’s disease), albumin may be reduced (1 day)
  II. Protein-losing gastroenteropathy, if someone had a problem with intestines, and the blood which moves near intestines leak some plasma to intestines, the concentration of a protein would decrease dramatically

- Many molecules show electrophoretic and isoelectric focusing mobility.
- Many plasma proteins exhibit polymorphism. Polymorphism is a Mendelian (monogenic) trait that exists in the population in at least two phenotypes—neither of which is rare. (The ABO blood groups are the best-known examples) Plasma proteins showing polymorphism are α1-antitrypsin, haptoglobin, transferring, ceruloplasmin, and immunoglobulin

- **Question:** Why Hemoglobin is not a Plasma Protein?
  **Answer:** Because it is inside RBC, it is from the cellular components, it is not considered as plasma protein. (Hemoglobin molecular weight is 65 kilo Dalton)

- **Plasma Proteins and Polymorphism**

  - Mutations can occur to any gene in any protein, these mutations can either be harmful (causing a disease) or harmless (color of eyes or hair ...) , these mutations can lead to polymorphisms, but **How**? If more than 1% of population got this genetic mutation, it would be called polymorph, the
protein would be said to have two forms (e.g. 2% and 98%)Polymorphism, when a specific site on a chromosome has multiple alleles in the population, it is said to be polymorphic (many forms)

❖ Structure-function relationship

- Albumin Molecular Weight = 69 kilo Dalton (it is ellipsoidal in shape)
- Fibrinogen (soluble, elongated) when you got an injury, fibrinogen turns to fibrin, and fibrin is insoluble, and this what leads to blood clot
- Structure-Function relationship significance: (shape has a great effect on function) If albumin’s structure was elongated like fibrinogen, the human will not live.

❖ Applications:

- Densitometer: a device that measures the density of each band and presents it in peaks

- albumin has the highest concentration -highest peak- of all plasma proteins in any blood sample 50-60% of plasma proteins, then its normal concentration would be approximately (3.5-5.5), don’t memorize the relative values of the globulins but notice that their concentrations are close to each other, refer to the slides to see the values.

  Clinically, You cannot diagnose someone with albumin deficiency unless their albumin concentration was less than 2, this condition is called hypoalbuminemia.

- Hematocrit (packed cell volume PCV) is the cells volume over the whole blood volume, if a person has a hematocrit of 40, this means that 40 percent of the blood volume is cells and the remainder is plasma, let’s have a simple exercise, if the hematocrit is 0.4 (40%) and the total blood volume is 5 liters, calculate the plasma volume

**Answer:**

\[
\text{hematocrit} = \frac{\text{cells volume}}{\text{total blood volume}} \\
0.4 = \frac{\text{cells volume}}{5} \\
\text{Cells volume} = 2 \\
\text{Total blood volume} = \text{cells volume} + \text{plasma volume} \\
\text{Then plasma volume equals 3 liters}
\]

Extra Note: when you go to some references you might see this test called packed red blood cell volume, because of the fact that RBCs form the largest portion of the cellular component.
In adult males, the measured hematocrit is about 47% and in adult females is about 42%

Do you remember when we said that plasma forms 55% of the blood volume and cells form 45% of blood volume, but actually it is not the same in males and females according to the hematocrits mentioned above!

- **A** is normal

- **B** (chronic/acute) infection, inflammation or cancer cases
  - **Explanation**: from the first glance, you might think that it is an indication of **kidney failure** because albumin has low concentration compared to others, but if it was a kidney failure, all proteins should be at low concentration.
  - (Relatively, this case is one out of two options, either the albumin is lower in concentration than gamma globulin or albumin is equal to gamma in concentration. In other words, the globulins are either slightly higher or unchanged, **Einstein** can help in Biochemistry, too)

  Albumin is not an acute phase protein – we will talk about this piece of information in more details in the following lecture.
• **Liver problems, such as hepatitis**
  
  - **Explanation:** All plasma proteins synthesized in liver except immunoglobulins (gamma globulins), so it is a liver problem, because all proteins are relatively low in concentration except *gamma globulins, they are synthesized in beta cells*.

• **Kidney failure (nephrotic syndrome)**
  
  - All proteins concentrations are low

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**Clinical Note:** normally, there should be no proteins in urine, (in some cases, slight amount could be neglected though)

“*If You Want to Go Fast, Go Alone. If You Want to Go Far, Go Together*” - A message to Doctor 2016