BIOCHEMISTRY





Lecture #23

اللهم استعملنا لنصرة دينك

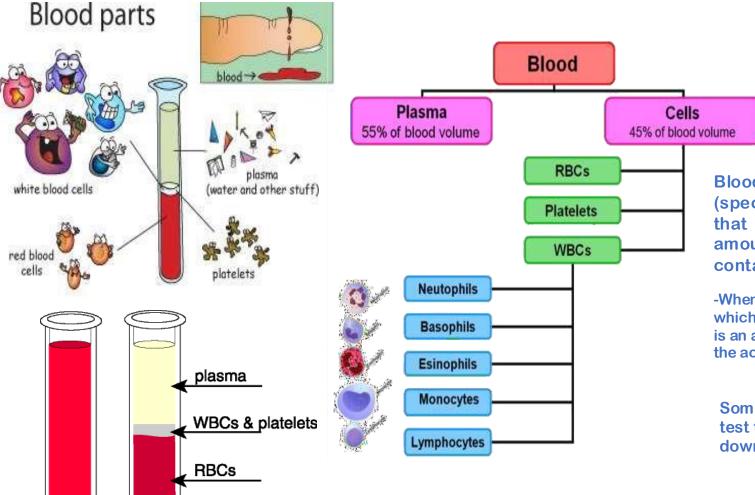
Written by: Leen Mamoon and Hala Swiedan

Plasma Proteins

Edited by: Raghad Hamdan



Blood

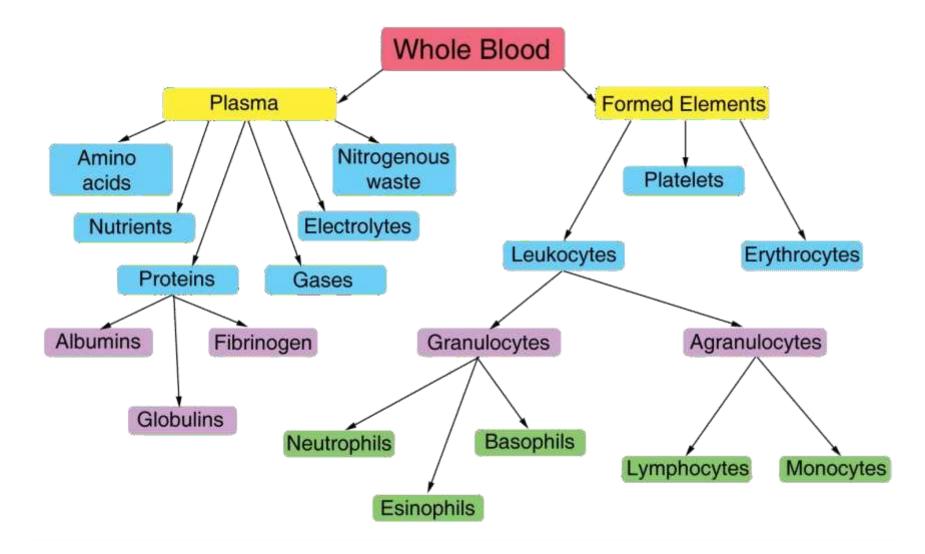


Blood constitutes huge amounts of different cells (specially RBCs), and 55% fluid = plasma, you can note that sometimes when you draw blood (to know the amount of hemoglobin for example), the end of the tube contains gel, this gel is **heparin**, why we use it?

-When a blood sample is drawn, it naturally starts to coagulate, which can interfere with the accuracy of the test results. Heparin is an anticoagulant, meaning it prevents this clotting by inhibiting the action of certain clotting factors in the blood.

Sometimes we use empty tube when we do blood test to make the blood clot; therefore the RBCs go downward and the plasma floats.

Blood: plasma vs. cells



Plasma

- It is the liquid medium in which blood cells are suspended.
- Composition: Water (92%)
 Solids (8%) everything except water
 - Organic:
 - Plasma proteins: Albumin, Globulins & Fibrinogen
 - Non-protein nitrogenous compounds: urea, free amino acids, uric acid, creatinine, creatine & NH3

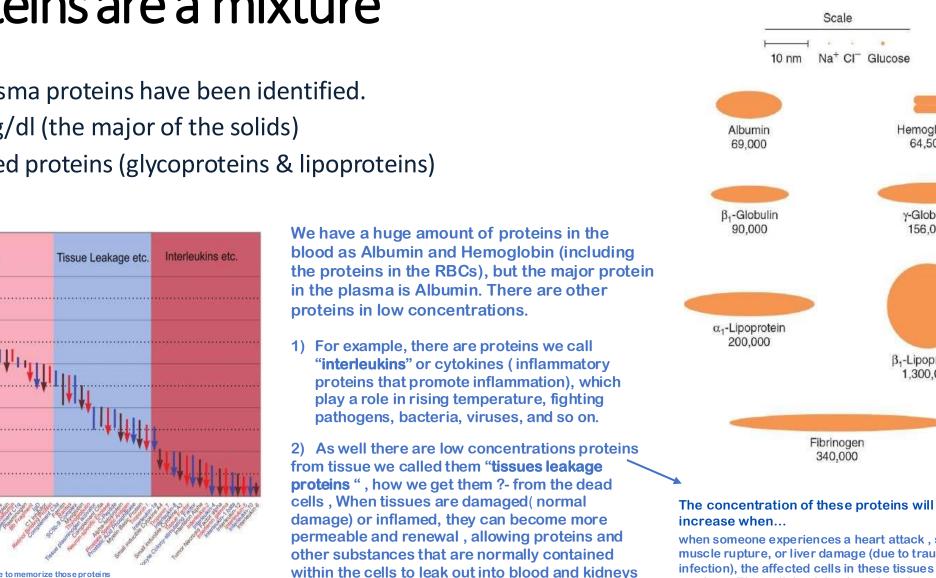
Don't memorize

these things

- Lipids: Cholesterol, TG, phospholipids, free fatty acids
- Carbohydrates: Glucose, fructose, pentoses
- Other substances as: Ketone bodies, bile pigments, vitamins, enzymes & hormones
- Inorganic: Na+,K+,Ca2+,Mg2+,Cl-,HCO3-,HPO42-, SO42-

Plasma proteins are a mixture

There is something called wide dynamic range, which means how much wide is the concentration of the protein.



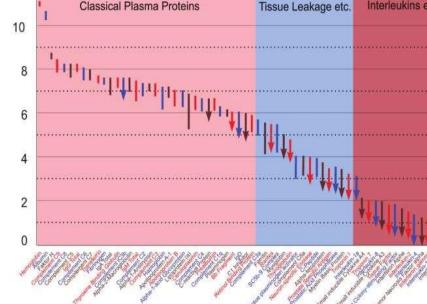
and then we get ride from them out the body.

More than 500 plasma proteins have been identified.

- Normal range 6-8 g/dl (the major of the solids)
- Simple & conjugated proteins (glycoproteins & lipoproteins)



12



You're don't have to memorize those protein

when someone experiences a heart attack, skeletal

muscle rupture, or liver damage (due to trauma or viral infection), the affected cells in these tissues die in large numbers. This cell death leads to the release of intracellular proteins and enzymes into the bloodstream so doctors can note that there is a damage in lever for example

CI

Glucose

Hemoglobin

64,500

y-Globulin

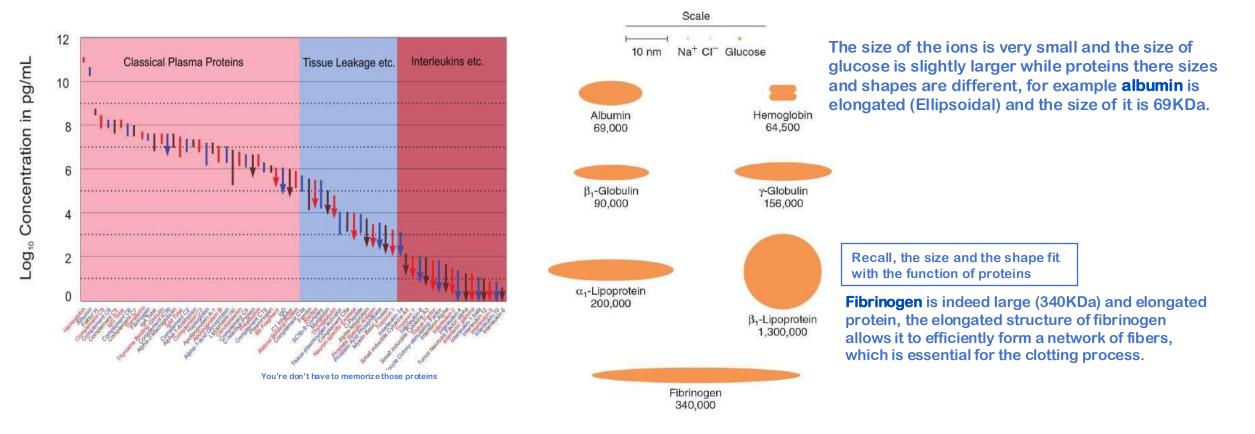
156,000

B₁-Lipoproteir

.300.000

Plasma proteins are a mixture

- More than 500 plasma proteins have been identified.
- Normal range 6-8 g/dl (the major of the solids)
- Simple & conjugated proteins (glycoproteins & lipoproteins)



-Numbers are not required except for those mentioned in the next slides.

TABLE 52-2 Majnr Funcõons of Blood

- 1. Respiration—transport of oxygen from the lungs to the tissues and of CO₂ from the tissues to the lungs
- 2. Nutrition—transport of absorbed food materials
- Excretion—transport of metabolic waste to the kidneys, lungs, skin, and intestines for removal
- 4. Maintenance of the normal acid-base balance in the body
- 5. Regulation of water balance through the effects of blood on the exchange of water between the circulating fluid and the tissue fluid
- 6. Regulation of body temperature by the distribution of body heat
- 7. Defense against infection by the white blood cells and circulating antibodies immunoglobulins, and antibodies.
- 8. Transport of hormones and regulation of metabolism
- 9. Transport of metabolites
- **10.** Coagulation

Don't memorize this slide , just know there are serval functions depending on the type of proteins



The separation of plasma proteins

Phoresis= movement Electro =electrical field Chroma=color

Electrophoresis (most common): serum (defebrinated plasma),

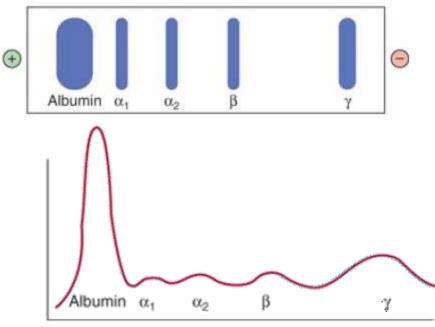
five bands (albumin, $\alpha 1$, $\alpha 2$, $\beta,$ and $\gamma)$

NORMAL VALUES:

Name	Absolute values (g/l)	Relative values (%)
Albumins	35 - 55	50 - 60
al-globulins	2 - 4	4.2 – 7.2 5 %
a2-globulins	5-9	6.8 – 12 9 %
β-globulins	6-11	9.3 – 15 12 %
γ-globulins	7 – 17	13 – 23 18 %

-Electrophoresis is a powerful technique used to separate high concentration proteins based on their size, charge (isoelectric point), or both. When we use this technique we will see five bands or clusters (albumin, $\alpha 1$, $\alpha 2$, β , and γ)

recall that we mentioned before (in mid material) there is something called isoelectric point of amino acid, also proteins have an isoelectric point (negative charges and positive charges equal to each other), because proteins have C terminus and N terminus and R group so we can say that this protein at physiological PH is negativity charged / neutral and so on...

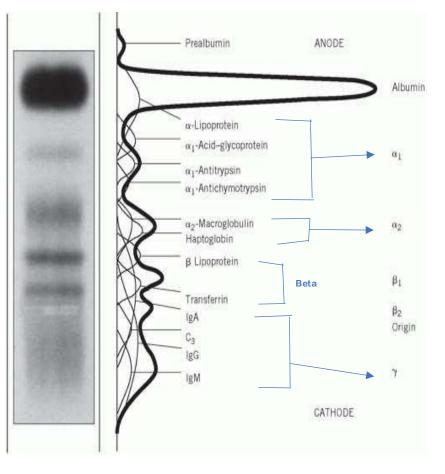


Chromatogram

Electrophoresis of plasma proteins

- Albumin is smaller than globulin, and slightly negatively charged
- Globulins (3 bands):
 - α band:
 - α1 region consists mostly of α1antitrypsin

Hypoalbuminemia is a condition where albumin levels are low but still present.Analbuminemia is a genetic disorder resulting in the complete absence of albumin

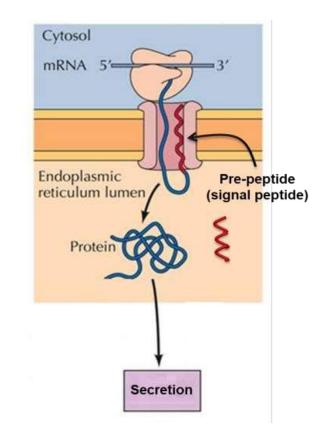


α2 region is mostly haptoglobin, α2-macroglobulin, & ceruloplasmin β band: transferrin, LDL, complement system proteins γ band: the immunoglobulins

Synthesis of plasma proteins

- Mostly liver (major source of proteins in plasma)(albumin, globulins), γ-globulins (plasma cells; lymph nodes, bone marrow, spleen).
- Most plasma proteins are synthesized as preproteins (signal peptide).
- Various posttranslational modifications (proteolysis, glycosylation, phosphorylation, etc.).
- Transit times (30 min to several hours).
- Most plasma proteins are glycoproteins (N- or Olinked).
 - Albumin is the major exception.

The signal peptide guides the newly synthesized protein to the ER or other cellular compartments, Inside the ER, the protein undergoes post-translational modifications before being transported to its final destination, for example by Glycosylation, then those proteins go to the golgi and then get released, (After the protein enters the ER, the signal peptide is typically cleaved off (المحيد في because it did it's function)by a signal peptidase enzyme. Recall, Gamma globulins, or immunoglobulins, are a class of proteins in the blood that function as antibodies, playing a crucial role in the immune system by identifying and neutralizing pathogens like bacteria and viruses, they are from B cells.



Glycosylation is a post-translational modification where carbohydrate molecules (glycans) are added to proteins or lipids.

Proteolysis is the biological process of breaking down proteins into smaller peptides or amino acids by enzymes called proteases (The duration of synthesizing proteases is variable)

Plasma Proteins & genetic variation

- The follow a mendelian or monogenic trait. They exist in population in at least two phenotypes.
 - The ABO blood groups are the best-known
 - examples
- Electrophoresis or isoelectric focusing is used for analysis.

Plasma Proteins Half-Lives

- Albumin & haptoglobin (20 & 5 days, respectively)
- Diseases can affect half-lives
 - In protein-losing gastroenteropathy such as Crohn's disease, albumin may be reduced (1 day).
 Losing protein through the gastrointestinal tract refers to the loss of proteins from

Losing protein through the gastrointestinal tract refers to the loss of proteins from the body via the gut. This can happen in several medical conditions. For instance, inflammation in the colon can lead to diarrhea, which damages the colon tissue and results in the degradation and loss of proteins in huge amounts

Genetic polymorphism

Genetic polymorphism refers to the occurrence of two or more genetically distinct forms, or alleles, of a gene. Sometimes it does have a clinical significance.

-Eye color is a classic example of genetic polymorphism, influenced by multiple genes that contribute to the diversity of eye colors in humans.

> The "half-life" of a protein refers to the time it takes for half of the protein molecules in a cell or system to be degraded or eliminated.



Albumin

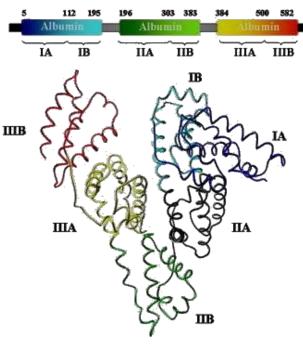
- The Major Protein in Human Plasma, 69 kDa, halflife (20 days)
- The main contributor to the osmotic pressure (75-80%)
- Liver: 12 g/day (25% of total protein synthesis) (liver function test)
- 3/5 total plasma proteins (3.4-4.7g/dL)
- Synthesized as a preproprotein
- Monomeric (made just of one polypeptide)
- Ellipsoidal shape (does not increase viscosity like fibrinogen)

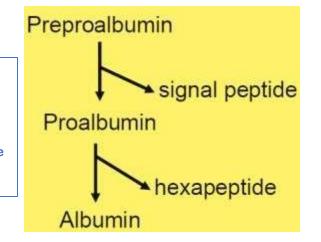


Anionic at pH 7.4 with 20 negative charges

-Osmotic pressure is crucial for maintaining fluid balance between the blood vessels and surrounding tissues, and albumin plays role in that, the concentration of albumin must be high (It helps retain water within the blood vessels by attracting and holding onto water molecules)

Pre= proteins are synthesized and enter the ER **Pro=**cleaving or removing part of proteins (this is process of protein maturation), some proteins are synthesized as proproteins (inactive protein that need processing to do their function and get released)





Albumin is synthesized as preproalbumin , the pre region (signal peptide) is removed—pro region (hexapeptide) is removed —mature protein (albumin)

Albumin's binding capacity

Binds to these molecules nonspecifically

- binds various ligands:
 - Free fatty acids (FFA) Certain
 - **Steroid,** (steroid hormones(Androgens, estrogen, cortisol) are hydrophobic, they need carrier (albumin)
 - hormones Bilirubin
 - Plasma tryptophan
 - Metals: Calcium, copper and heavy metals
 - Drugs: sulfonamides, penicillin G, dicumarol, aspirin (drug-drug

When muscles require energy (doing their metabolism) they signal adipocytes (fat cells) to release fatty acids, since fatty acids are hydrophobic, they cannot dissolve easily in the aqueous environment of the blood, to prevent them from clustering together and to facilitate their transport, albumin binds to these free fatty acids. It transports the bound fatty acids through the bloodstream to various tissues, including skeletal muscle.

> Bilirubin is a product that is produced during the normal breakdown of red blood cells. It is a byproduct of the degradation of heme (hydrophobic and toxic), a component of hemoglobin in red blood cells. So albumin carry the heme to prevent it from clustering and entering the CNS which damages it.

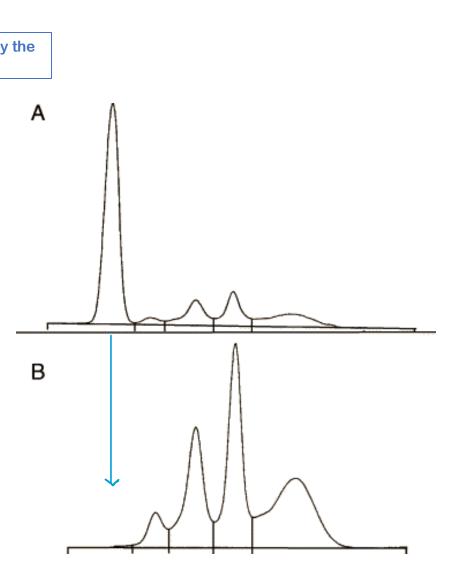
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Analbuminemia

Analbuminemia is a rare genetic disorder characterized by the absence or extremely low levels of albumin in the blood.

- There are human cases of analbuminemia (rare)
- Patients show moderate edema!!!

Albumin is the most abundant protein in the blood plasma, playing crucial roles in maintaining osmotic pressure, so if we don't have albumin the blood will be very fluidic and a lot of water will go into the tissues (due to a large amount of water compared to the protein) but edema doesn't take place , because body compensate by producing more of the other proteins, **"moderate edema"** refers to a noticeable but not severe accumulation of fluid in the tissues, causing swelling. In the context of analbuminemia, moderate edema occurs because of the lack of albumin.



Other clinical disorders

Hypoalbiminemia: edema seen in conditions where albumin level in blood is less than 2 g/dl

Malnutrition (generalized edema)

Gastrointestinal loss of proteins

Analbuminemia is a genetic disorder resulting in the complete absence of albumin, The body may compensate by increasing the production of other plasma proteins, which helps to partially maintain oncotic pressure and thus mitigates the severity of edema "moderate edema ".

Hypoalbuminemia is a condition where albumin levels are low but still present, here cells and tissue can't compensate due to the presence of albumin. Albumin presence results in feedback inhibition (even it was in little amounts). So, severe edema is a result of Hypoalbuminemia.

Hyperalbuminemia: dehydration (relative increase)



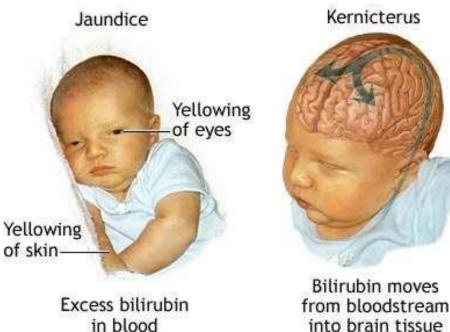
Other clinical disorders

Albumin can bind to molecules nonspecifically, molecules can include certain drugs. Any drug taken, first it gets processed & modified by liver cells ,then it becomes inactive and exits the body. Some exits to the blood, then some of these drugs in blood bind to Albumin, so the drug becomes inactive since it is not free. However, some of this drug remains active if not bound.

- Drug-drug interaction:
 - Bilirubin toxicity (aspirin is a competitive ligand): kernicterus and mental retardation
 - Phenytoin-dicoumarol interaction (epilepsy vs. anti-coagulant, respectively))

In some cases like Bilirubin toxicity, mostly occurs in children when they take aspirin. Aspirin will inhibit bilirubin to bind to Albumin. So bilirubin will be free in huge amounts, thus entering the CNS and damaging it. It will cause children to suffer from mental retardation.

«سَبِيلُ العُلا عَالٍ علىٰ مَن تَعَللا ومَن جَدّ في سَعيٍ لأمرٍ تَمَكنا»



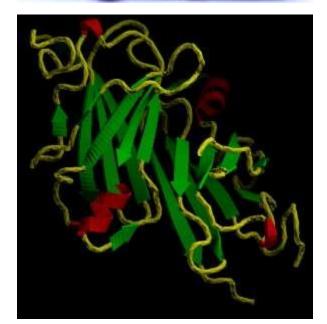
Phenytoin and dicoumarol should not be taken together. Phenytoin treats epilepsy, while dicoumarol is an anti-coagulant. Both bind to Albumin at the same place, so if taken together, they will compete on who binds to Albumin. Thus, the levels of these two drugs will be high in blood and they can cause severe side effects.

Prealbumin (transthyretin)

- It exists as a 62-kDa glycoprotein.
- It has short half-life (≈2 days).
- It is a sensitive indicator of poor protein nutrition.
- Main function: (T2, T4)
 - T4 (Thyroxine) and T3 carrier

Prealbumin will not give albumin, pre here indicates to the location (it's saperation) in chromatogram , pre = coming before albumin.

rion.



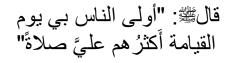
سبحان الله والحمد لله ولا المه الا الله والله اكبر

Acute-phase proteins

- Plasma proteins whose Levels increase (up to 1000 folds), acute inflammation, tissue damage, chronic inflammation & cancer.
- Solution C-reactive protein (CRP), $\alpha 1$ -antitrypsin, haptoglobin, & fibrinogen $\frac{\text{Examples of}}{\text{these proteins}}$
- Interleukin-1 (IL-1) is the main stimulator

stimulates the releasing of Acute phase proteins.

Inflammation (presence of pathogens) leads to: the body increase the concentration of these proteins to protect the body. For example, if someone has a fever this will make the blood test, which shows the acute phase proteins, be of high level so he has systemic inflammation (inflammation in the whole body). This is a result of having viruses/bacteria...



Purpose of acute of phase proteins

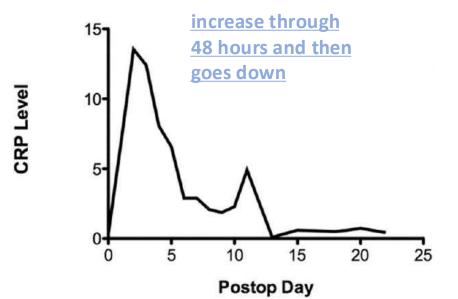
Will be explained in the next slides.

Protein	Function
C-reactive protein	Stimulates the complement pathway
α1-antitrypsin	neutralizes certain proteases released during acute inflammation
Fibrinogen	Coagulation factor
Transferrin	Iron binding (preventing microbe uptake of iron)
Haptoglobin	Hemoglobin binding (iron protection)
Ceruloplasmin	Iron oxidation (iron binding by ferritin)

C-reactive protein (CRP)

CRP binds on the surface of the cell wall of bacteria called (pneumococci).

- It is able to bind to a polysaccharide called fraction C in the cell wall of a bacterial species called pneumococci.
- It is undetectable in healthy individuals.
- It helps in the defense against bacteria and foreign substances.
- It is detectable in many inflammatory diseases (Acute rheumatic fever, bacterial infection, gout, etc.) & Tissue damage
- Its level reaches a peak after 48 hours of incident (monitoring marker).



The body knows that if there is inflammation (caused by this bacteria) the body increases releasing of CRP level.

Globulins

α1-globulins	α2- globulins	β- globulins	γ-globulins
 α1-antitrypsin α1-fetoprotein α1- acid glycoprotein Retinol binding protein 	 Ceruloplasmin Haptoglobin α2-macroglobulin 	 CRP Transferrin Hemopexin β2- microglobulin 	 IgG IgA IgM IgD IgE

Found in alpha1 (the band coming after albumen)

 α 1-antitrypsin

Usually, your body releases these enzymes which degrade the proteins in tissue (causing tissue damage). When cell release proteinase, it releases antiproteinase with them to maintain and control the balance. Example on antiproteinase: antitrypsin.

Also known as (AKA) α1-antiproteinase (52 kDa) It neutralizes the trypsin & trypsin-like enzymes (such as elastase). 90% of α 1- globulin band What is MM? Our cells are diploid (2n, 2 chromosomes you get, a chromosome from your Polymorphic (75) الله عدة أشكال ؛ Like eye color. mother (which indicates to M protein) and one from your father (also M). Alleles Pi^M, Pi^S, Pi^Z, Pi^F (MM is the most common) Deficiency (genetic): Emphysema is found in people with ZZ or SZ. Since Z is not really active in stopping the trypsin MS and MZ usually not affected (proteinase). It is ncreased level of $\alpha 1$ - antitrypsin (acute phase response) Active elastase + α_1 -AT \rightarrow Inactive elastase: α_1 -AT complex \rightarrow No proteolysis of lung \rightarrow No tissue damage

proteinase = enzyme that break protein

Active elastase + \downarrow or no $\alpha_1\text{--}AT \rightarrow$ Active elastase \rightarrow Proteolysis of lung \rightarrow Tissue damage

اذا ما فهمتوا هاد السلايد قدموا سلايدين في شرح اله ^-^

Smoking & α1- antitrypsin deficiency

modification (oxidation) for met that exists in antitrypsin, as a result it will not work efficiently.

Chronic inflammation Oxidation of Met358 devastating in patients methionine

methionine-sulfoxide

ZZ type + smoker = 💽 💽

So smokers have higher ability to have emphysema.

تجميع أفكار للسلايدين الى قبل:)

Acute phase protein \rightarrow trying to protect your body against inflammation.

Example of these proteins : alpha 1 Anti trypsin

Anti: trying to inhibit trypsin (control protein Trypsin : enzyme breaks proteins degradation) بحاول يثبط الإنزيم الى بكسر

للزم يكون في توازنه بن الإنزيمين

There are many types of antitrypsin \rightarrow M, S, Z, F

What is the difference between them ¿ They differ in how good they are in inhibiting trypsin. (يختلفوا (بالكفاءة

 $M \rightarrow most effective.$

 $Z \rightarrow$ less effective =not effective

رادا عندائ جين (عطالت M وكمان . تين أعطاك M MM colie 3 (by selles mm.

اله trypsin ماشي و عم بكسر عن جنب و طرف مافي اشي يوقف (لأنه ال Z ضعيف جدا ما عنده قدره بشط الإنزيم) لذلك ال Trypsin رح يدمر النسيج بالرئة و تصير الرئة نافخة emphysema

معدد بال من الطبيعي بكون عنا الموجود بال Antitry من الأنذ يمات المضاوة مثل المعام في مشكلة عند المدخنين إنه رح يصير أكسدة للحمض الاميني Met الموجود بال .Antitrypsin inflamation المكن بريهو بدوالة ال

طيب في ناس عندهم جينين أعطوا ZZ شو بصير عندهم ؟

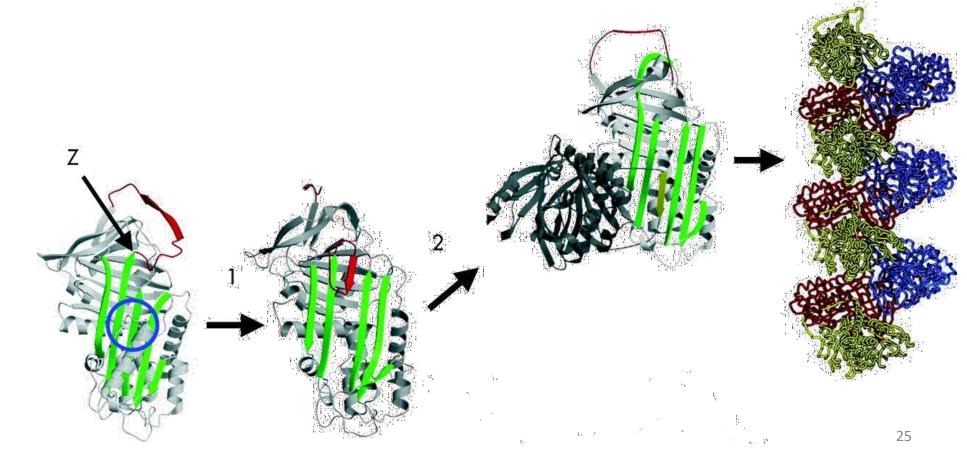
نتيجة الأكسدة رح يبطل الإنزيم شغال و رح يعمل emphysema للمدخنين... it's not a joke الحل: تترك التدخين لأنه

يقول الله عز وجل : (سَنُرِيهِمْ آيَاتِنَا فِي الْأَفَاقِ وَفِي أَنَّهُ الْحَقُ) أَنَّفُسِهِمْ حَتَّى يَتَنَيَّنَ لَهُمْ أَنَّهُ الْحَقُ

Liver disease & $\alpha 1$ - antitrypsin deficiency

Cirrhosis can be caused by ZZ

Liver disease: ZZ phenotype polymerization (loop with β-sheet), aggregates in liver, cirrhosis (10%)



Hemoglobin exist in RBC, when RBC die hemoglobin will be released.

Haptoglobin (Hp)

- It is an acute phase protein.
- α2 glycoprotein (90kDa)
- A tetramer $(2\alpha, 2\beta)$ + Polymorphic

Heme converted to bilirubin and bilirubin is toxic.
 The most important thing in Heme is iron. Millions of RBCs will be damaged which means millions of iron will be lost. We have to compensate this iron, we need it! لازم نرجع نسحبه

Recovery of hemoglobin should occur for two reasons:

Haptoglobin bind to Hemoglobin to maintain it (preventing filtration).

- Two genes, designated Hp1 and Hp23, producing phenotypes:
 - \cong Hp 1-1 $\rightarrow \alpha$ 1, α 1 + 2 β
 - \blacksquare Hp 2-1 \rightarrow α1, α2 + 2β
 - 𝔅 Hp 2-2 → α2, α2 + 2β

Haptoglobin is synthesized by our bodies to maintain hemoglobin. It exists in certain percentage. A lot of RBCs will be degraded Hemolysis means the release large amounts of hemoglobin. So part of haptoglobin percentage will bind to hemoglobin. Then those haptoglobins will be damaged. If we lost large amounts of RBC = larger percentage of haptoglobin will bind and larger amounts of haptoglobin will be lost.

- Binds the free hemoglobin (65 kDa); prevents loss of hemoglobin & its iron into urine
- Hb-Hp complex has shorter half-life (90 min) than that of Hp (5 days)
- Decreased level in hemolytic anemia haptoglobin
 People with hemolytic anemia have a low level of haptoglobin

نقص حدید ، فقر دم Lysis of RBC

https://voutu.be/KCh-7Ghj0jY?si=9omMkr-6F6Y7VTMR

Watch this , may be helpful

Ceruloplasmin

Protein which is binding to copper

- A copper-containing glycoprotein (160 kDa)
 - It contains 6 atoms of copper
- Metallothioneins (regulate tissue level of Cu)
- It regulates copper level: contains 90% of serum Cu.
- A ferroxidase: oxidizes ferrous to ferric Having Enzyme activity
 - Important for transferrin binding
- Albumin (10%) is more important in transport

Ceruloplasmin + albumin are copper carrying proteins. However, albumin is more important in binding and transporting copper.

Amine oxidase

Copper-dependent superoxide dismutase

- Cytochrome oxidase
- Tyrosinase

The problem of copper and iron \rightarrow both of them are toxic.

(make oxidation/ tissue damage>>> should be free for being toxic.

أنتم حملة الراية ، والراية لا تحملها يد ضعيفة...

Pathological conditions related to ceruloplasmin

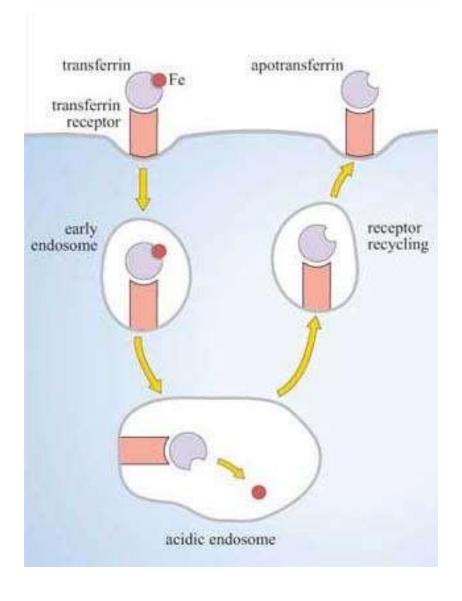
- Ceruloplasmin deficiency can arise from genetic causes or lack of dietary Low copper. amount
 - Hypoceruloplasmenia
 - Ceruloplasmin levels are ~50% of normal
 - No clinical abnormalities
 - Aceruloplasminemia
 - No ferroxidase activity of ceruloplasmin 0
 - If left untreated, accumulation of iron in tissues and organ failure Θ
 - - Wilson's disease Depending on transporting of copper not ceruloplasmin
 - Defective transporter (copper-binding P-type ATPase or ATP7B protein) 0 leading to excess liver copper, increased apoceruloplasmin, and copper toxicosis.

Transferrin

Transfer + ferrin = iron transport

- A β1-globulin that functions as iron transporter
- A glycoprotein synthesized by the liver.

You only have to know that this protein transfers iron.

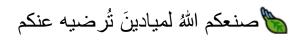


Alpha-2 macroglobulin

- It is a large plasma protein.
- It is responsible for the transport of 10% of zinc and cytokines in blood.
- α2-macroglobulin binds to and inactivates diverse type of proteases.
 - Blood coagulation

Protein degradation

«يا نُخبة الجيل يا مَن أنتم أملٌ طالَ الظّلامُ فنرجو منكم الفجرا»

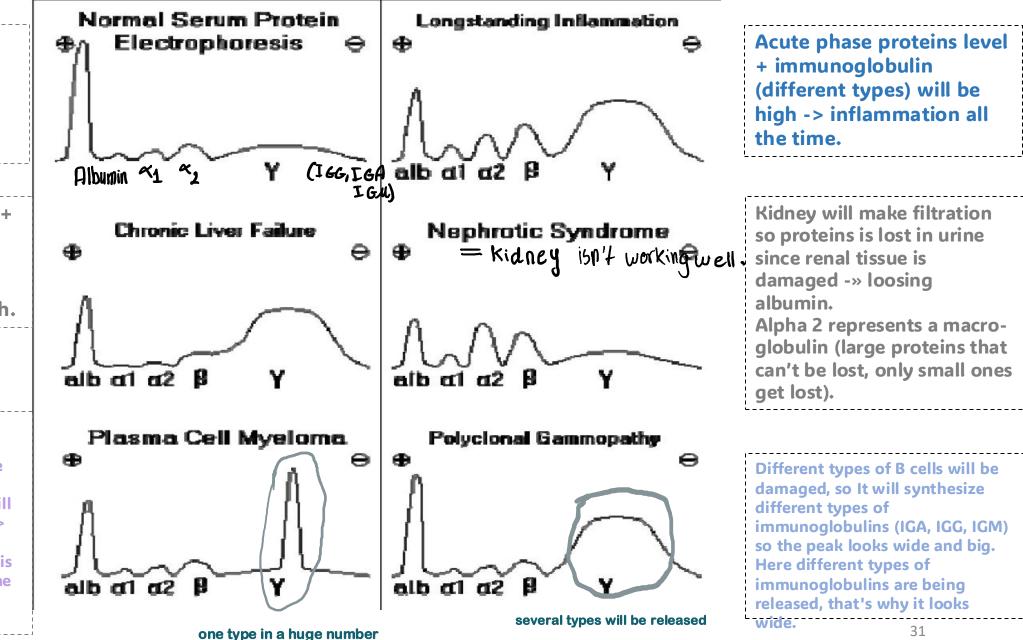


Very important (memorize the chromatogram and the condition).

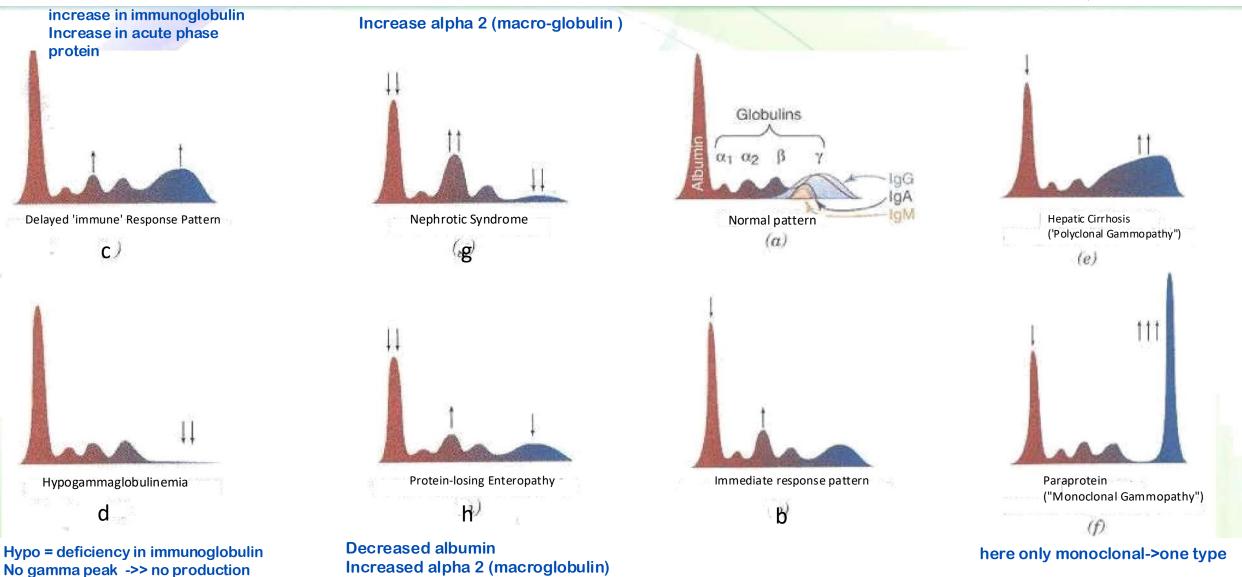
All of them are peaks, except gamma band, because of large quantities and different sizes.

Can't synthesize albumin + alpha one + alpha 2 -> All will be of low level compared with gamma, which relatively looks high.

Maturation of B cells and they make and release one antibody, they become plasma cells. These plasma cells could be cancerous, when it becomes cancerous it will release one type of antibody ->> increase in number of cells and increase in antibody number. This peak look sharp because only one antibody level will increase in blood.



ن وأخيرااا يعطيكم العافية



(alpha one + beta) will relatively decrease

of immunoglobulin



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

Corrections from previous versions:

Versions	Slide # and Place of Error	Before Correction	After Correction
$V1 \rightarrow V2$	Slide 15	Albuminemia	Analbuminemia
	Slide 28		Change the position of blue sentence
	Slide 27		Link is added
V2 → V3			

Additional Resources Used:

رسالة من الفريق العلمى:

- 1. Book pages
- 2. YouTube videos
- 3. Webpages
- 4. Anything else...

تم الاعتداء بكل وحشية على أبسرى فلوطينيين, تم استئصال طحال وكبد أبسير فلوطيني -من ذوي الاحتياجات الخاصة !- وأصيب بالعمى من أثر النعذيب..وتم تسليمه بعد ذلك لأحد مستشفيات عُرْة من قبل قوات الاحظال ليستشه هد بعدها بأربعة أيام إثر الجررائم التي ارتكبها الاحظال بحقه ! الاحظال يقوم بالعمليات الجراحية غالبا بدون تخدير، بهدف تعذيب الأبسير !هذا كله جزء ضئيل مما يتعرض له أهلنا في عُرْة اللهم إنهم قد طغوا في البلاد وأكثروا فيها الفساد.. فصئبً عليهم يا ربنا غضبك صبًا وأهلكهم واجعل كيدهم في نحور هم يا ربنا ومولانا ومعيننا أنت المُعز وأنت المُزل

> ادعوا لكل من شارك في كتابة هذا الموديفايد براحة البال وصفاء الذهن "اللهم هوّن ثم هوّن ثم هوّن ثم أرح نفساً لا يعلم بحالها إلا أنت "