

☒ Sheet

☐ Slides

Number: 17

Done by: Anas Mourad

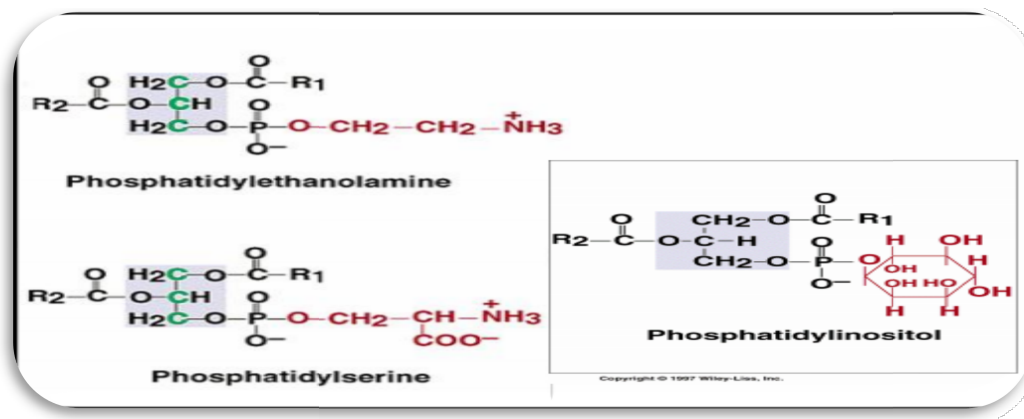
Corrected by: Jawad Masarweh

Subject: Biosynthesis of TAG

Doctor: Faisal Alkhatib



We are going through a brief revision of what we took at the end of the last lecture:

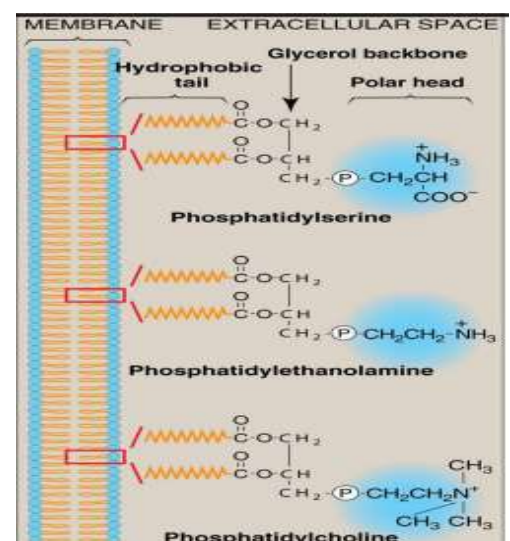


It's important to be able to recognize these structures. The doctor will not ask about them but it's important to know the relationships between these compounds (remember that these reactions don't occur at our body).

- ✓ What is the reaction that converts phosphatidylserine to phosphatidylethanolamine?

Decarboxylation. "it's easy to answer this question because we know the structures of the two compounds".

- ✓ Phosphatidylglycerol = phosphatidylinositol.
- ✓ The hydrocarbon chain forms the non-polar portion of the molecule which is the **interior** of the phospholipid bi-layers.
- ✓ The polar head including the negative charge on the phosphate and the positive charge on the amine, will be the **exterior** (in contact with water).

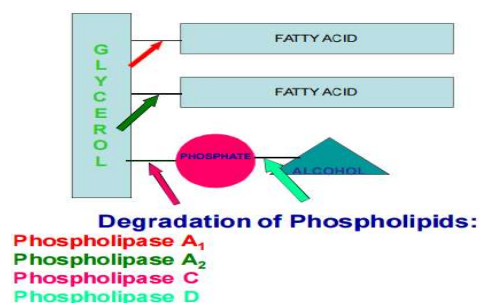


## Degradation of phospholipids:

Phospholipases are the enzymes that catalysis the degradation of phospholipids.

Look at the figure on the right:

We have 2 ester bonds and 2 phosphoester bonds so generally speaking we have 4 ester bonds and they are different in locations so we can predict that there are different enzymes for their degradation.



The Phospholipase **A1** which act on the intact molecule targets "attacks" the ester bond between the first fatty acid and the glycerol.

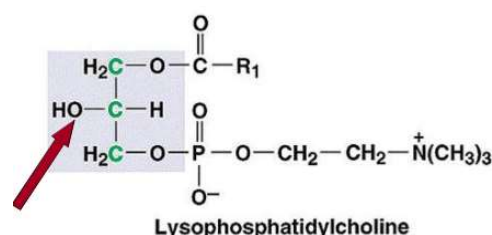
The **Phospholipase A2** which act on the intact molecule also, targets the ester bond between the second fatty acid and the glycerol.

\*Note that A2 doesn't act after A1 they both act on the intact molecule (independent from each other).

The next enzyme is known as **Phospholipase C** which acts on the intact molecule and **attacks** the *phosphoester* bond between the glycerol and the phosphate.

Finally, **Phospholipase D** catalyzes the hydrolysis of the bond between the phosphate and the alcohol.

Note that So, these enzymes acts on the intact molecule only.

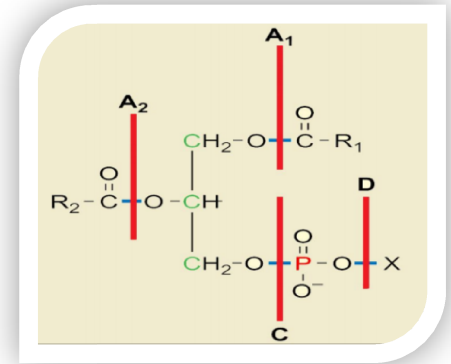


On the right, it's a phospholipid that has already lost the fatty acid at carbon number 2, so this is the product (lysophosphatidylcholine) of **Phospholipase A2**,

**Phospholipase B** acts on the hydroxyl group of the second carbon

Lysophosphatidylcholine have the ability to **lyse** cells "the membranes"; thus It has a strong detergent action, it's a **solubilizing modification** agent, and it's degraded by "**Phospholipase B**"(can be called **Lysphospholipase**).

The figure on the right summarizes the structure of the glycerophospholipids and the lines shows where the phospholipases enzymes do act.



Some notes about Phospholipase:

### 1. **Phospholipase A2:**

- It's presented in snake and bee venoms.

So when a snake bites, it injects the tissue with Phospholipase A2 that acts on the phospholipids of the cell, releasing lysophosphatidylcholine which cause lyses of cell membrane.

- It has a role in releasing arachidonic acid.

Usually the second fatty acid in the phospholipid is an unsaturated fatty acid, sometimes its arachidonic acid "an unsaturated fatty acid", so the **Phospholipase A2** releases arachidonic acid which is a precursor of the prostaglandins and different eicosanoids.

Prostaglandins have a role in inflammation, so the inflammation response is based on releasing arachidonic acid by the action of **Phospholipase A2**, that's why Phospholipase A2 is inhibited by glucocorticoids such as Cortisone or cortisol, thus acting as anti-inflammatory. (they inhibit the release of arachidonic acid → there is no synthesis of Prostaglandins → inflammation is stopped).

### 2. **Phospholipase C:**

If the (x) head group "illustrated in the last figure" was inositol with two phosphates (PIP<sub>2</sub>) the action of Phospholipase C will give us DAG (diacylglycerol) and inositol triphosphate (IP<sub>3</sub>) both of them are used as second messengers.

So, you are required to know:

- 1) Where is the location of the bonds hydrolyzed by each one of these phospholipases.

- 2) The structure of phospholipids in order to tell what are the products of using one of these Phospholipases.

\*please refer to slides for better understanding.

### **Biosynthesis of Triacylglycerol & Phosphoacylglycerol**

We talked about the synthesis of Triacylglycerol and we are going to discuss the synthesis of Phosphoacylglycerols.

Phosphatidic Acid:

It's the Common Intermediate in both of them.

#### **Biosynthesis of glycerophospholipids:**

We have 3 components:

- 1) Alcohol1: DAG (diacylglycerol) which is a glycerol with two fatty acids.
- 2) Phosphate.
- 3) Alcohol2.

We have to join them together to make the phospholipid molecule.

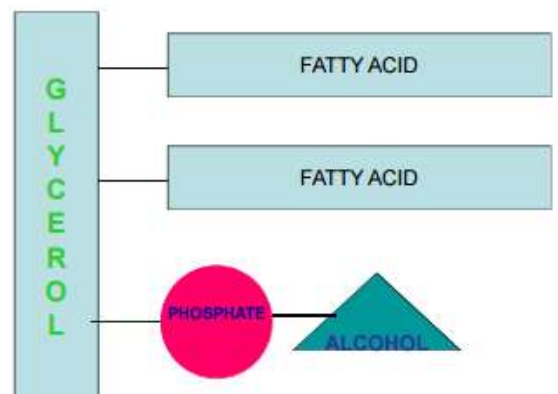
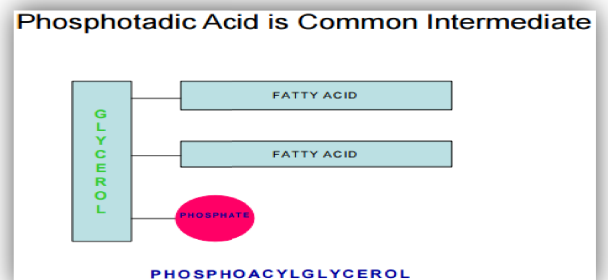
Remember that always when joining two monomers "building blocks" together we have to have one of them in the activated form.

Such as UDP-glucose in the glycogen **synthesis**.

There are two strategies to do that:

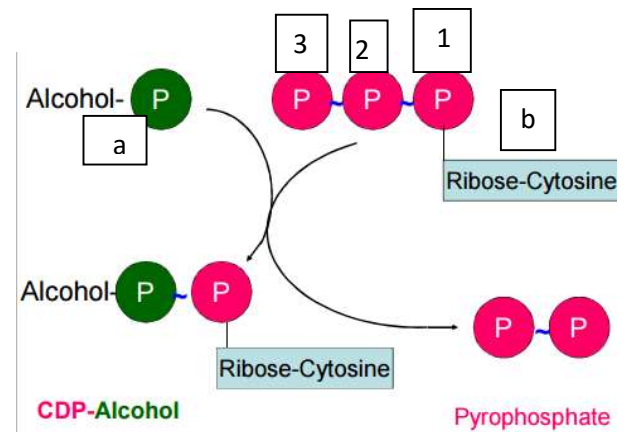
- 1) Transfer ~ (Phosphate-Alcohol1) to Alcohol2.
- 2) Transfer ~ (Phosphate-Alcohol2) to Alcohol1.

\*Note that phosphorylation of alcohol is the activation of it. "(~)" means activated".



- **Formation of Activated Carrier:**
  - a) is the phosphorylated alcohol.
  - b) CTP: cytosine "nitrogenous base" triphosphate, it's the same as ATP but we have cytosine instead of adenine.

So, alcohol phosphate will come and replace the second(#2) and the third(#3) phosphate to produce a pyrophosphate and a CDP-Alcohol.



How many high energy bonds are cleaved by this reaction? Only one.

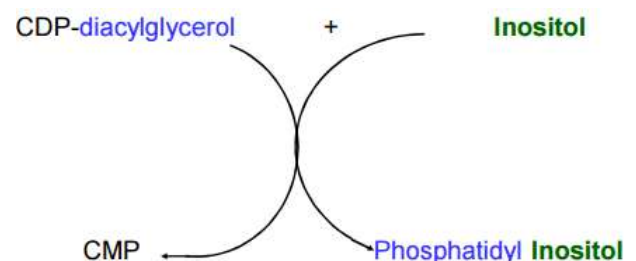
How many high energy bonds are formed by this reaction? Only one too.

CTP will be broken into CMP and PP (pyrophosphate), thus one high energy bond is broken. The CMP will be transferred to the alcohol, thus one high energy bond is formed so, we can predict that ( $\Delta G = 0$ ) → the reaction is reversible we make it irreversible by cleavage of the pyrophosphate giving 2 organic phosphates.

Doctor mentioned that: the formation of the activated carrier is actually producing energy.

For example:

**1-Formation of Phosphatidyl Inositol:**

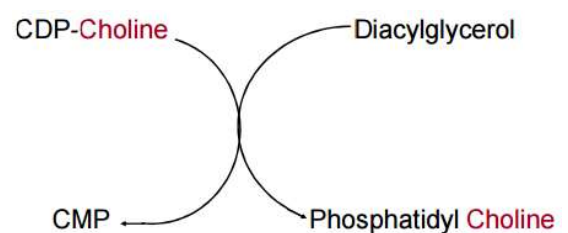


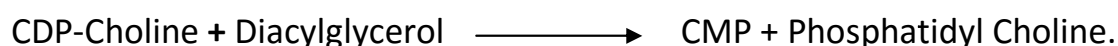
CDP-diacylglycerol + Inositol → Phosphatidyl inositol ( **because we took the diacylglycerol with phosphate** ) + CMP

The given CDP must be converted back to CTP thus require ATP, thus the total energy used will be about 2 ATP.

**2- Formation of Phosphatidyl Choline:**

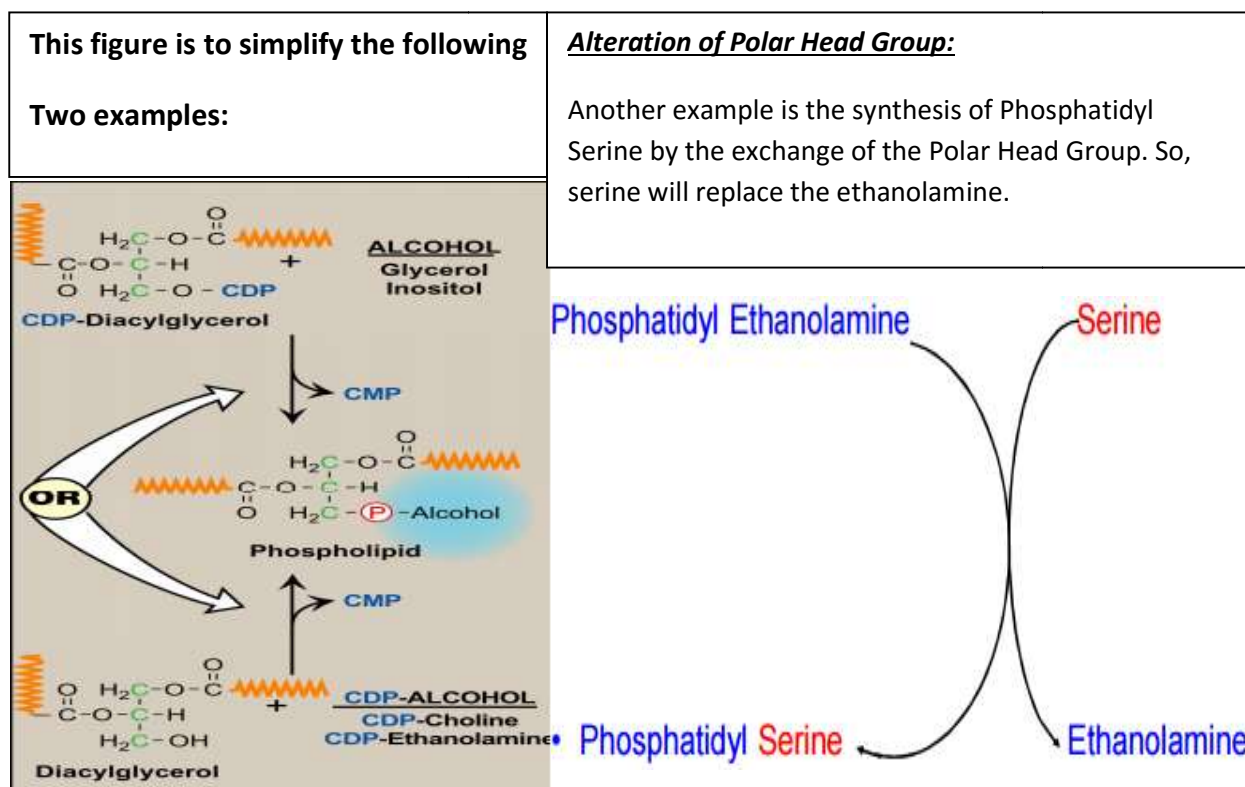
Transfer of Phosphocholine or Ethanolamine to Diacylglycerol to give Phosphatidyl Choline.





**\*Refer to the slides for the rest of examples (very important).**

**We should know that ethanolamine and choline will be transferred as active alcohols while glycerol and inositol will be transferred to an Active Alcohol or they can be synthesized by Alteration of Polar Head group.**

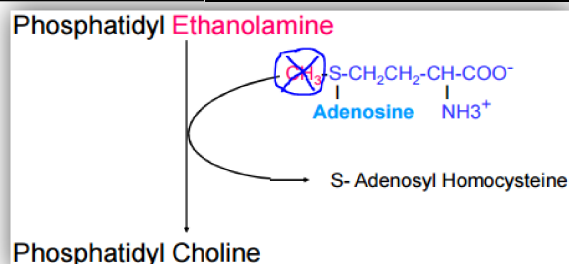


We can convert Phosphatidylserine to Phosphatidylethanolamine by decarboxylation. Or we can synthesis Phosphatidylserine from phosphatidylethanolamine by the exchange of the Polar Head Group. (Serine will replace the ethanolamine).

Note that: all Decarboxylation reactions are irreversible. So, we can't convert Phosphatidyl Ethanolamine to Phosphatidyl Serine by carboxylation.

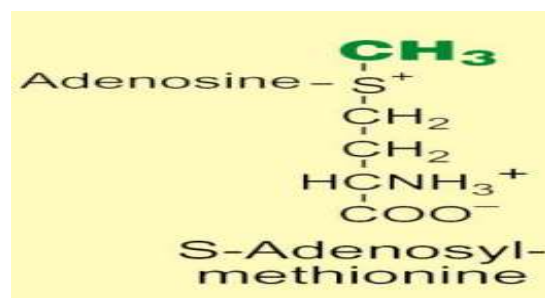


**We can make phosphatidylcholine by methylation of Phosphatidylethanolamine.**



To do that we have to replace the three hydrogens in the Phosphatidyl Ethanolamine by three methyl groups to get Phosphatidyl Choline. Thus we need a methyl donor → S-Adenosyl Methionine (SAM).

The structure of S-Adenosyl Methionine:



SAM acts as methyl donor in many reactions in the body.

It's active donor of methyl group because its sulfur has three bonds making the molecule unstable. So, it's easy to lose the methyl group to phosphatidylethanolamine forming phosphatidylcholine.

Note that the remaining structure is called S-Adenosyl Homocysteine Homocysteine (Homocysteine: similar to cysteine but with 4 carbons).

S-Adenosyl Homocysteine Homocysteine can be either converted back to SAM in a cyclic way or the Homocysteine can be degraded into cysteine ... etc

Note that we need Three SAM, because we are replacing three hydrogens by three methyl groups.

\*Please refer to slides there are many illustration figures that were captured from the book.

❖ Now we are going to mention two roles of phospholipids:

### 1. Surfactant Action of Phospholipids:

Surfactant (detergent): lowers the surface tension of water. "التوتر السطحي"

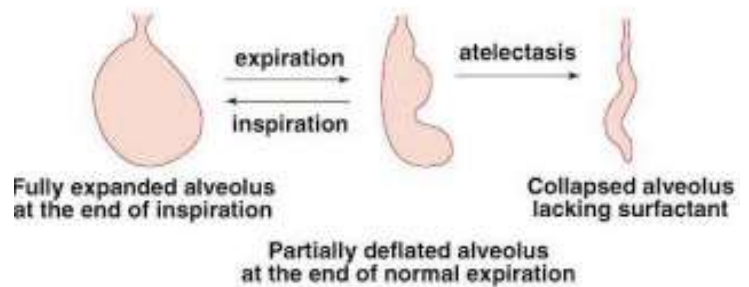


Water will form a spherical shape when a drop is added in a test tube (due to surface tension) while acetone will spread out

If we add surfactant to water it will spread on the plate, it'll no longer has surface tension. This is done by breaking the hydrogen bonds (responsible for the surface tension).

And this is an important function in the lungs.

The structure on the right representing the smallest lung unit which is the alveolus.



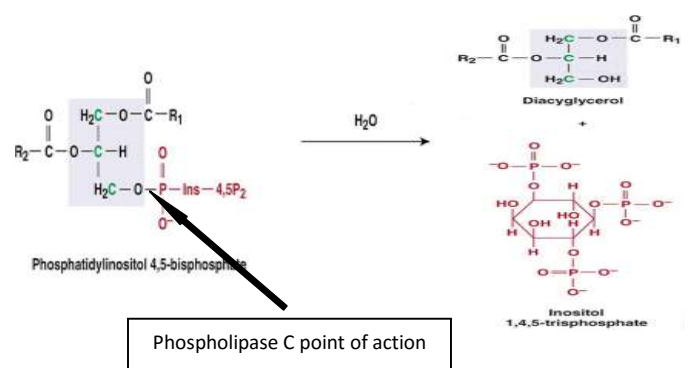
During inspiration it's fully expanded filled with air and then during expiration it's partially deflated and it'll not collapse due to the presence of surfactants. And if the surfactants were absent it will undergo atelectasis (the walls of the alveolus are going to collapse and the breathing process will become much harder)

Main surfactant in the lung is dipalmitoylphosphatidylcholine (DPPC)

Before birth during pregnancy the fetus doesn't have this because he/she is unable to breath (no respiration in the uterus) and during the last two or three weeks of delivery, the synthesis of surfactants. That's why if the baby born before term (in the 8<sup>th</sup> or the 7<sup>th</sup> month), he/she may suffer from respiratory problems due to the absence of surfactants. And if the **physician** expected early delivery, they injects the mother with cortisone or any other drug that will induce the synthesis of surfactants in the baby.

## 2. Synthesis of 2<sup>nd</sup> messengers:

As you can see the figure on the right this is a phosphatidylinositol 4, 5-bisphosphate (PIP<sub>2</sub>), it's also a component of the cell membrane.



It contains phospholipid and inositol with two phosphates "that's why we call it **phosphatidylinositol 4, 5-bisphosphate** ", and by the action of **phospholipaseC**, it will give us inositol tri-phosphate (IP3) and Diacylglycerol (DAG) as shown in the figure.

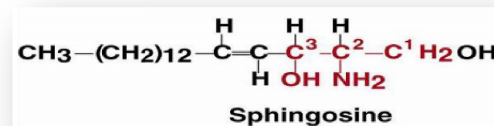
They both " inositol 1,4,5-triphosphate(IP3) and Diacylglycerol(DAG)" act as secondary messengers in some cells in response of binding of some hormones on the receptor.

### Metabolism of Sphingolipids

Sphingolipids can be Sphingophospholipids & Glycosphingolipids. Sphingophospholipids contains phosphate but glycolipids don't contain phosphate. On the other hand, glycolipids contain carbohydrate while Sphingophospholipids don't contain carbohydrate.

The alcohol in both cases is sphingosine (NOT Glycerol) .Note that it's an amine alcohol.

C1 a hydroxyl group, C2 an amine group, again at C3 a hydroxyl group



It contains 18 carbon atoms.

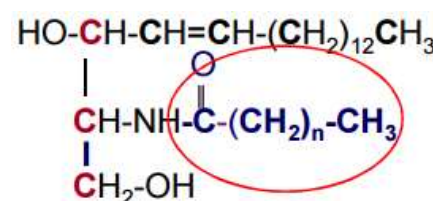
It's not important to know the details (location of double bonds, etc).

If you compare it with glycerol you'll find some similarities.

\*Note that the structure of it looks like monoacylglycerol (MAG).

In addition to that, a fatty acid can join the structure at the location of carbon number two making a bond "amide bond" with the amine group, producing ceramide

Now, the ceramide structure looks like diacylglycerol.



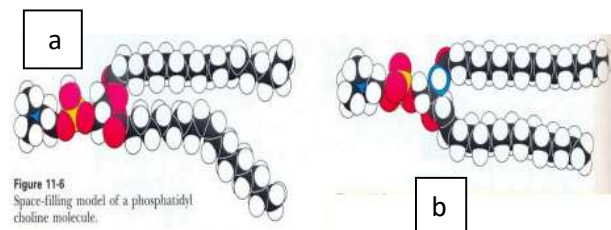
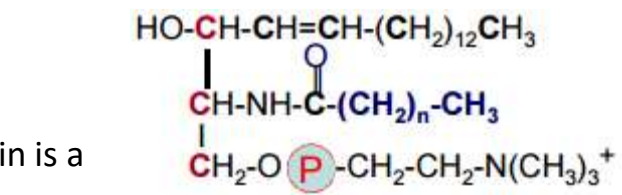
\*Note: -amide: amide bond.

And if we add phosphorylated choline forming sphingomyelin (look like glycerophospholipids e.g. phosphatidylcholine)

Notice that the long hydrocarbon chain is a part of the sphingosine. So, it can't be hydrolyzed by phospholipase for example "phospholipase A2 can't act here as well".

The space filling models are highly similar so study them well in order to know the differences. So, from this similarity we can make a conclusion that the sphingomyelin is a membrane component too.

- a) Is the phosphoglycerol.
- b) Is the sphingomyelin.



**Sphingomyelin** is found in all kinds of cells but especially in neurons in the myelin sheath "Schwann cells".

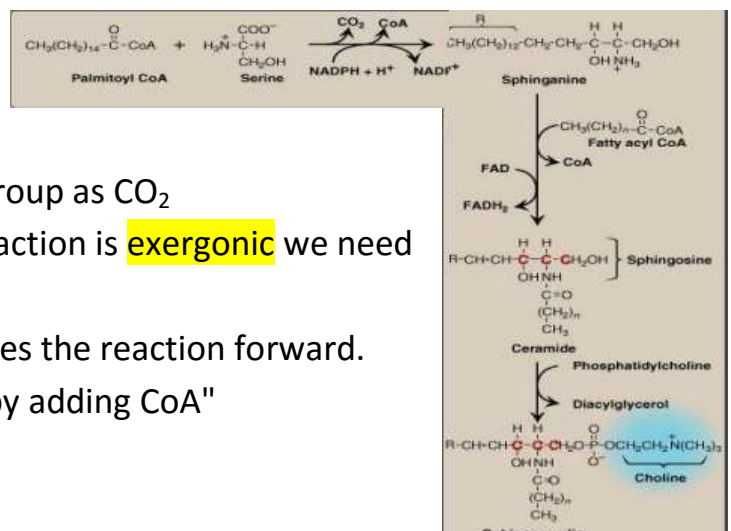
### Synthesis of Sphingomyelin:

It starts with the condensation between Palmitoyl CoA and serine in the presence of pyridoxine phosphate "vitamin B6" that is a cofactor. Forming sphinganine

Note that the source of the nitrogen and hydroxyl group in the sphingosine is Serine.

This doesn't require ATP because:

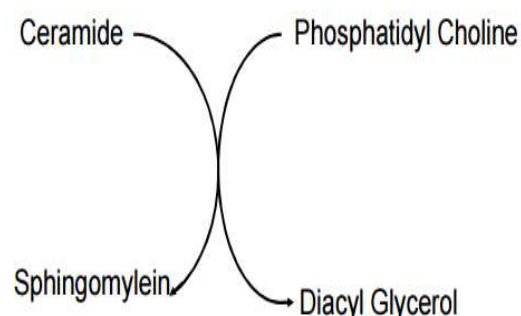
- a) There is a loss of a carboxyl group as  $\text{CO}_2$  "Decarboxylation". So, the reaction is **exergonic** we need no energy.
- b) The cleavage of the CoA pushes the reaction forward. "palmitic acid was activated by adding CoA"



Note that 2C (serine) + 16C (Palmitoyl CoA) = 18 carbon atoms.

The next step is accepting a fatty acid followed by **de-saturation** (removal of 2H+) forming Ceramide.

Note that the ceramide is the parent compound of all sphingolipids so we can add Phosphocholine "the source of it is Phosphatidyl Choline" to produce sphingomyelin and diacylglycerol.



**Glycolipids** are formed by linking one or More Sugars to Ceramide:

So, Ceramide +:

- a) Glucose or Galactose → Cerebroside (found in brain)
- b) Sulfated Galactose → Sulfoglycosphingolipids.
- c) Oligosaccharide → Globoside
- d) Oligosaccharide with NANA → Gangliosides

Sugars are added as ACTIVE sugars and added one by one

Ceramide + phosphocholine = **sphingomyelin**



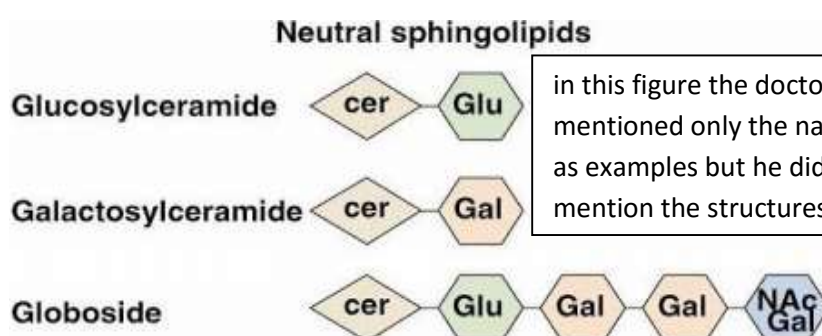
### Neutral sphingolipids:

They are called **Cerebroside**

Cerebro: brain

Osides: glycosidic bond

So, they are sugars connected by glycosidic bonds to Ceramides.



in this figure the doctor mentioned only the names as examples but he didn't mention the structures.

### Acid sphingolipids:

Containing sulfate group OR NANA.



## Gangliosides:

They have oligosaccharides with NANA (**N-Acetylmuramic acid**)

They are commonly named by abbreviation GM or GD or GT

GM: as in ganglioside

MonoNANA (one NANA)

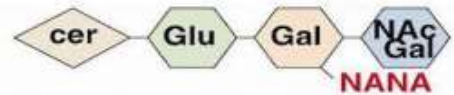
while GD is DiNANA (2 NANA molecules).

### Gangliosides

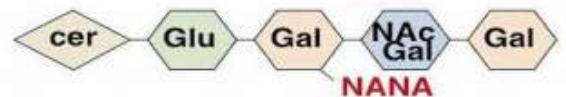
GM3



GM2



GM1



Note that: the longer the carbohydrate chains the smaller the number as in GM1 is made of 4 sugars and so on, the reason behind this, will be explained next lecture.

The End ...

Sorry for any mistake