METABOLISM

بسم الله الرحمن الرحيم

MID – Lecture 8

Metabolism of Glycerophospholipids

﴿ وَإِن تَتَوَلَّوْا يَسْتَبْدِلْ قَوْمًا غَيْرَكُمْ ثُمَّ لَا يَكُونُوا أَمْثَالَكُم ﴾

اللهم استعملنا ولا تستبدلنا

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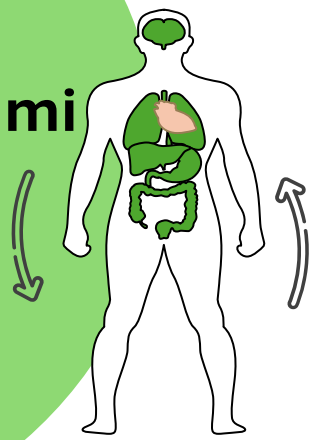
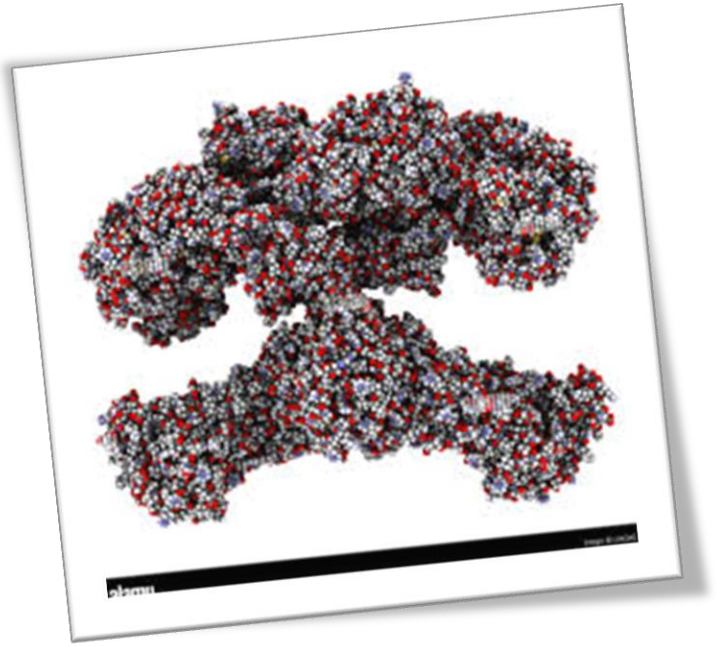


Quiz on the previous lecture



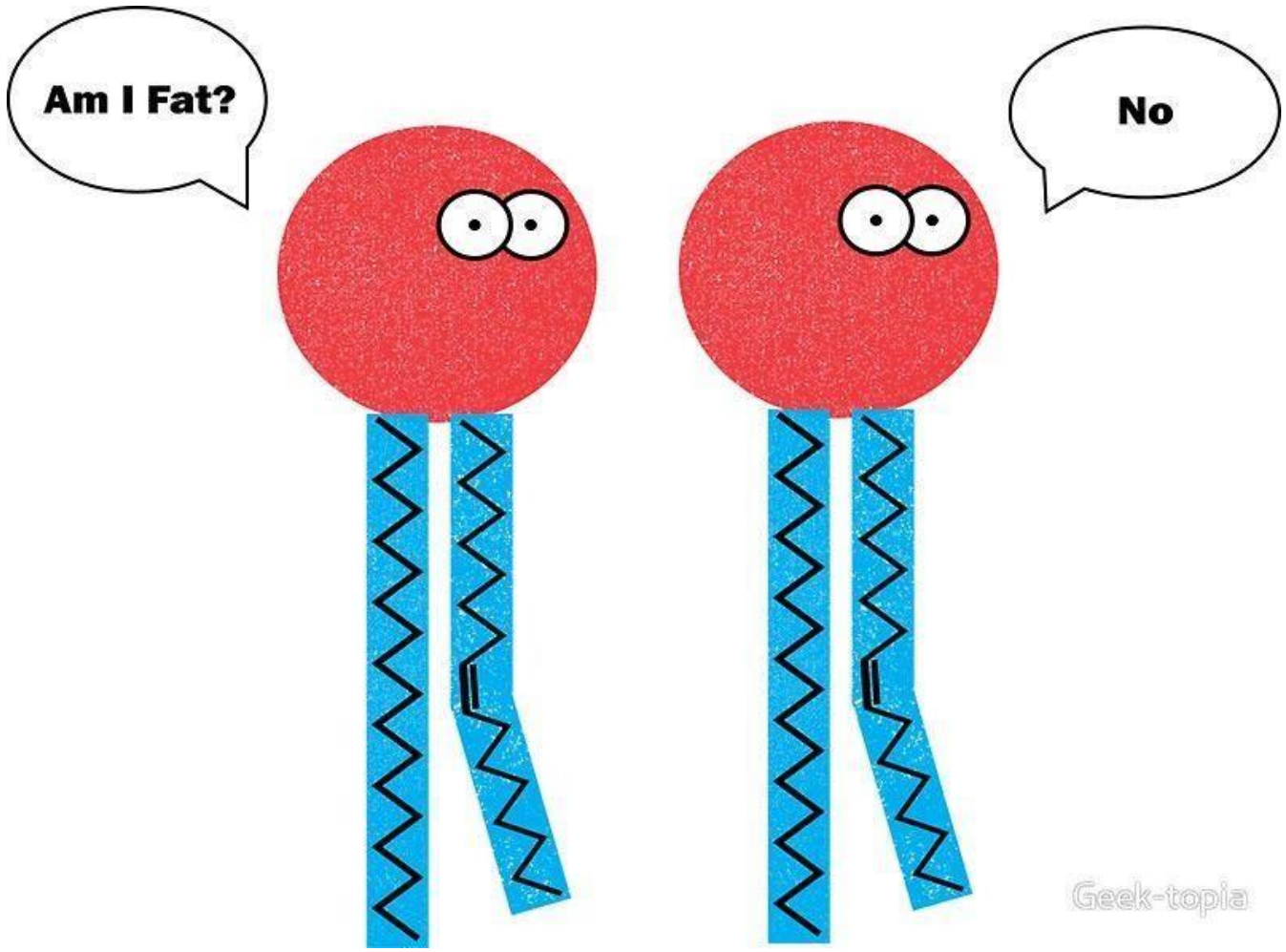
سلامٌ من الله عليكم يا رِفاق،

نعتذر عن عدد "السلايدات" الكبير، حيث كان ذلك لضمان إيضاح المعلومات، وضوح الخط، وجودة الملف.

المحاضرة سهلة بإذن الله، بالتوفيق يا كِرام

Fatty acid synthase

Metabolism of Glycerophospholipids

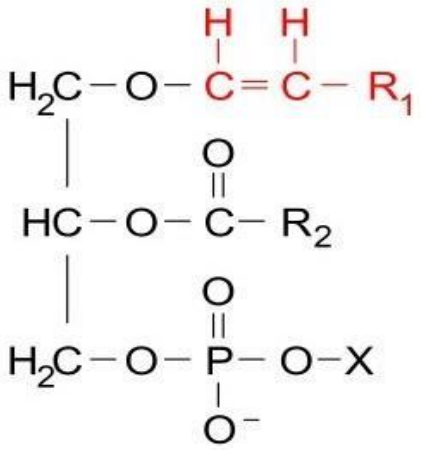


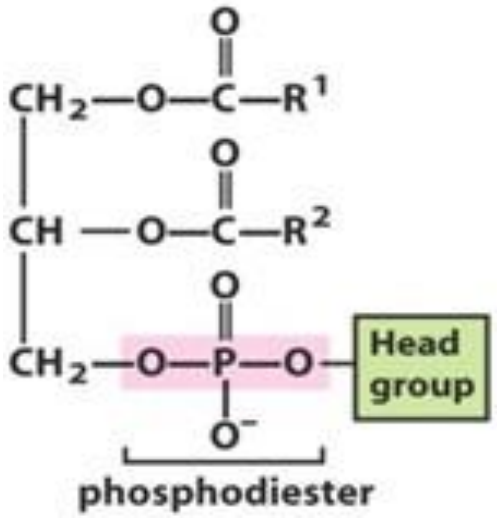
Dr. Diala Abu-Hassan

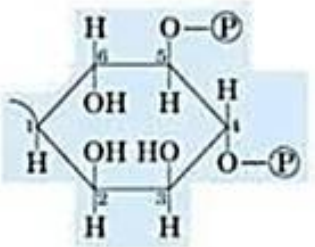
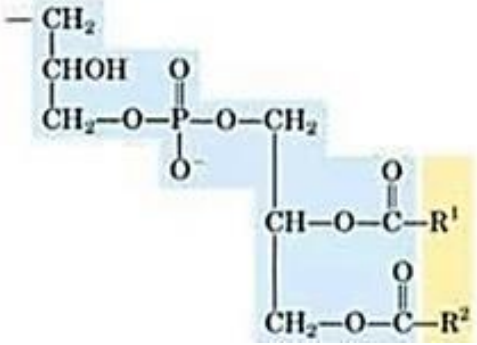
Lippincott's Biochemistry, Ch. 17

Structure and Classification of Glycerophospholipids

- Phosphatidic acids
- Phosphatidylcholine (lecithin)
- Phosphatidylethanolamine
- Phosphatidylserine
- Phosphatidylinositol
- Cardiolipin
- Plasmalogens





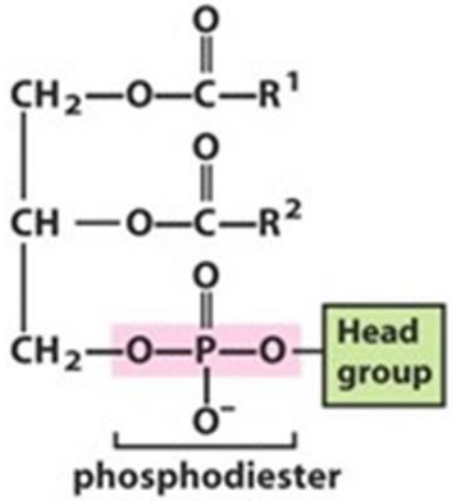
(I)	Phosphatidic acid	—	—H
(III)	Phosphatidylethanolamine	Ethanolamine	$-CH_2-CH_2-\overset{+}{N}H_3$
(II)	Phosphatidylcholine	Choline	$-CH_2-CH_2-\overset{+}{N}(CH_3)_3$
	Phosphatidylserine	Serine	$-CH_2-CH(\overset{+}{N}H_3)-COO^-$
	Phosphatidylglycerol	Glycerol	$-CH_2-CH(OH)-CH_2-OH$
	Phosphatidylinositol 4,5-bisphosphate	*myo*-Inositol 4,5-bisphosphate	
	Cardiolipin	Phosphatidyl-glycerol	

By comparing the structures of choline and ethanolamine, it can be observed that ethanolamine has hydrogens ($-NH_3^+$) in place of the methyl groups ($-N(CH_3)_3^+$) found in choline.

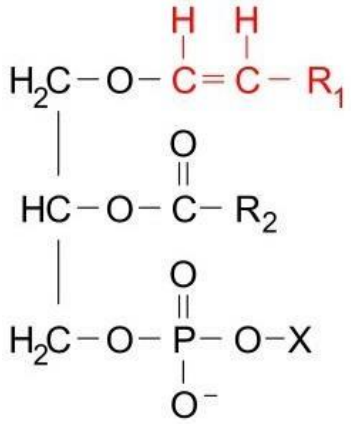
General structure of glycerophospholipids

- Glycerophospholipids are **membrane lipids** composed of:

1. Glycerol molecule (a poly alcohol).
2. **2** fatty acids attached to glycerol molecule on carbons 1&2 via **ester bonds**.
3. A phosphate group on carbon 3.



➢ the phosphate group has a **head group** attached via a **phosphodiester bond**; which can be as simple as a (I)**hydrogen** atom (phosphatidic acid) or a more complex structure as: (II)**choline** (phosphatidylcholine) or (III)**ethanolamine** (phosphatidylethanolamine), etc.



General structure of glycerophospholipids
Complex structures

1. **Cardiolipin:**

- Composed of 2 **phosphatidic acids** connected by a **glycerol** molecule.

➢ So, it has a PG; but it's not a PG itself.

2. **Plasmalogens:**

- Similar to GPs in structure, but **instead** of having a fatty acid on **carbon 1**, there's a **hydrocarbon chain** connected by an **ether** bond (but not ester).

✓ There's also modified GPs, including Phosphatidylinositol-4,5-Bisphosphate (**PIP2**) (and other inositol phosphates).

✓ PIP2 is involved in signaling, underneath the G-protein coupled receptors (**GPCRs**)- see slides 8&9.

- PG = phosphatidylglycerol
- GP = glycerophospholipids

Why do we need to make glycerophospholipids?

Important functions of glycerophospholipids other than their structural function in membranes

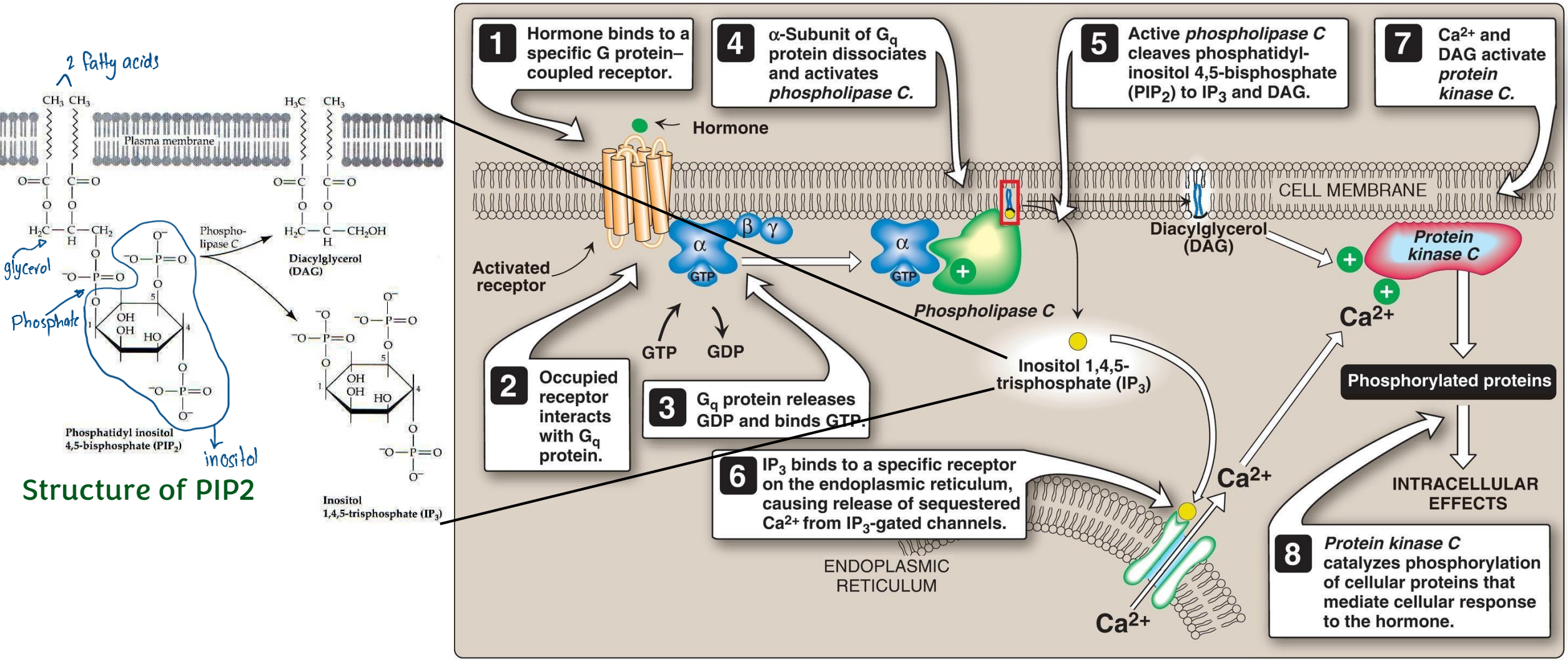
They are more common and abundant than sphingolipids

❖ Other functions of GPs:

1. Signaling by specific types of GP, like PIP2 which is involved in GPCR signaling.

2. Membrane attachment (anchoring proteins to the membrane).

Signaling by PIP2 products



Explanation- Signaling pathways

- Again GPCRs!

1. Activated by different hormones, growth factors, etc.
2. G-protein activation through the alpha subunit by exchanging GDP for GTP.
3. Alpha subunit is now active.
4. Eventually, alpha subunit activates downstream molecules.

➢ for example, the alpha subunit activates adenylyl cyclase which in turn activates another downstream molecules.

➢ But here the focus is on the activation of **phospholipase C**.

Explanation- Signaling pathways
PIP2 signaling

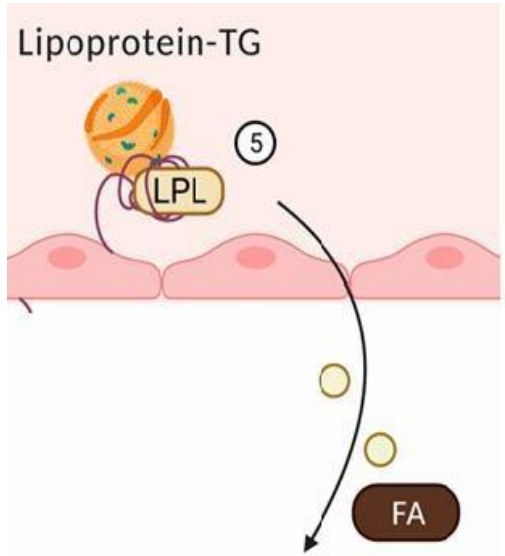
1. Following **alpha-subunit** activation, it **activates Phospholipase C.**
2. Phospholipase C is the enzyme that **degrades** membrane PIP2 into diacylglycerol (DAG, **hydrophobic**) and inositol triphosphate (IP3; negatively charged, thus **hydrophilic**).
    - Phospholipase C cleaves inositol with the phosphate groups (IP3), leaving the fatty acids associated with the glycerol molecule (DAG).
3. DAG remains in the membrane (because of its hydrophobic nature of the FAs), while **IP3** acts as a **second messenger** (leaves the membrane).
4. IP3 binds to the Ca^{2+} channels on the ER membrane (**IP3-gated calcium channels**).
5. Channels **open**, allowing **Ca^{2+}** to exit to the **cytosol**.
6. Ca^{2+} **activates** protein kinase C (**PKC**).
7. PKC **regulate** molecules in the **cytosol**.

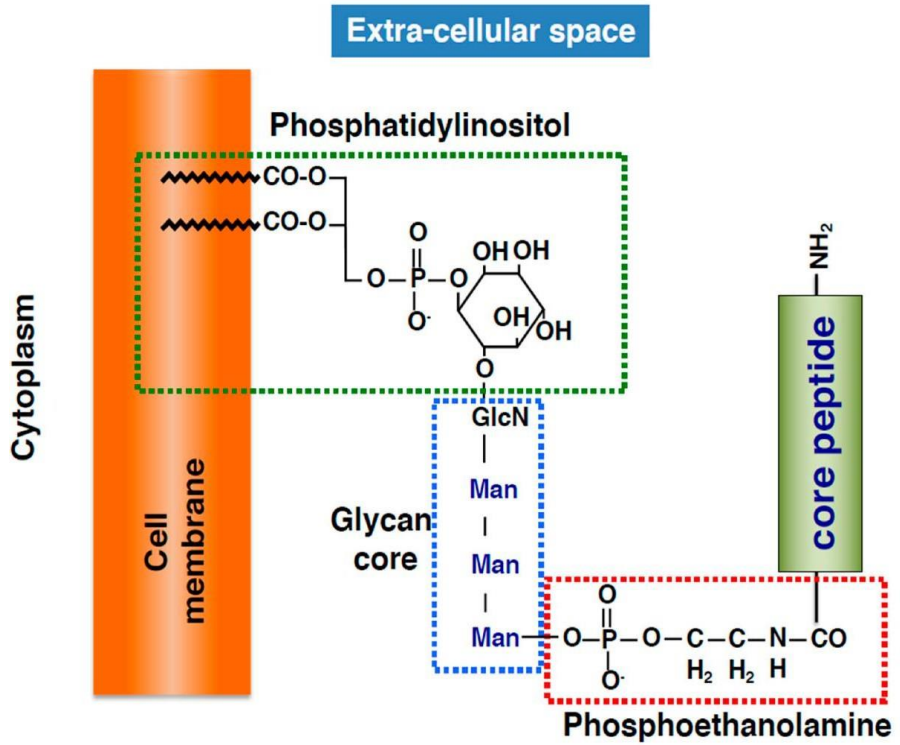
➢ The result, different cellular effects and responses.

GPI for membrane attachment

Glycosyl phosphatidylinositol (GPI) attaches proteins to the plasma membrane, anchoring proteins via phosphatidyl inositol.

- Advantage: lateral mobility
  - Example: lipoprotein lipase





✓ This is phosphatidylinositol (in membrane).
- The inositol is connected to sugars (**glycan core**).
- These sugars are attached to phosphoethanol amine.
- The phosphoethanol amine is attached to the protein (core peptide).

GPI for membrane attachment

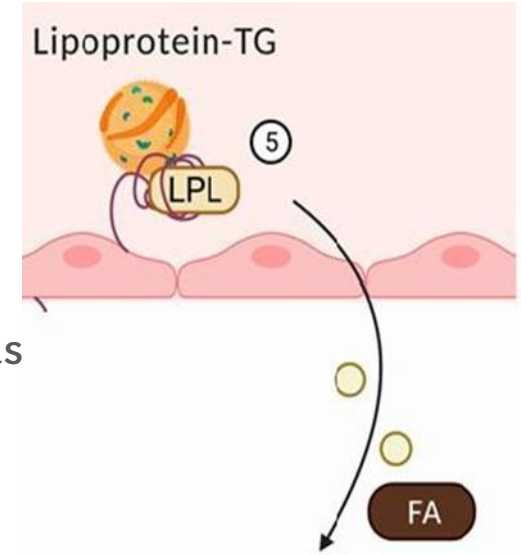
❖ Advantages/ importance:

1. Connection is covalent → **strong attachment.**
2. Provision of **flexibility** and ability to **move.**
   - ❑ If this protein is inserted as an **integral membrane protein**, its movement is restricted to the **plane of the membrane** itself, where it continuously moves **laterally** within the lipid bilayer

✓ These proteins are more mobile within the lipid bilayer because they are attached to the outer leaflet via the GPI anchor, allowing for rapid lateral movement. This can facilitate quick interactions and signaling. On the other hand, transmembrane proteins embedded within the lipid bilayer can be less mobile due to their integral nature and interactions with the cytoskeleton.

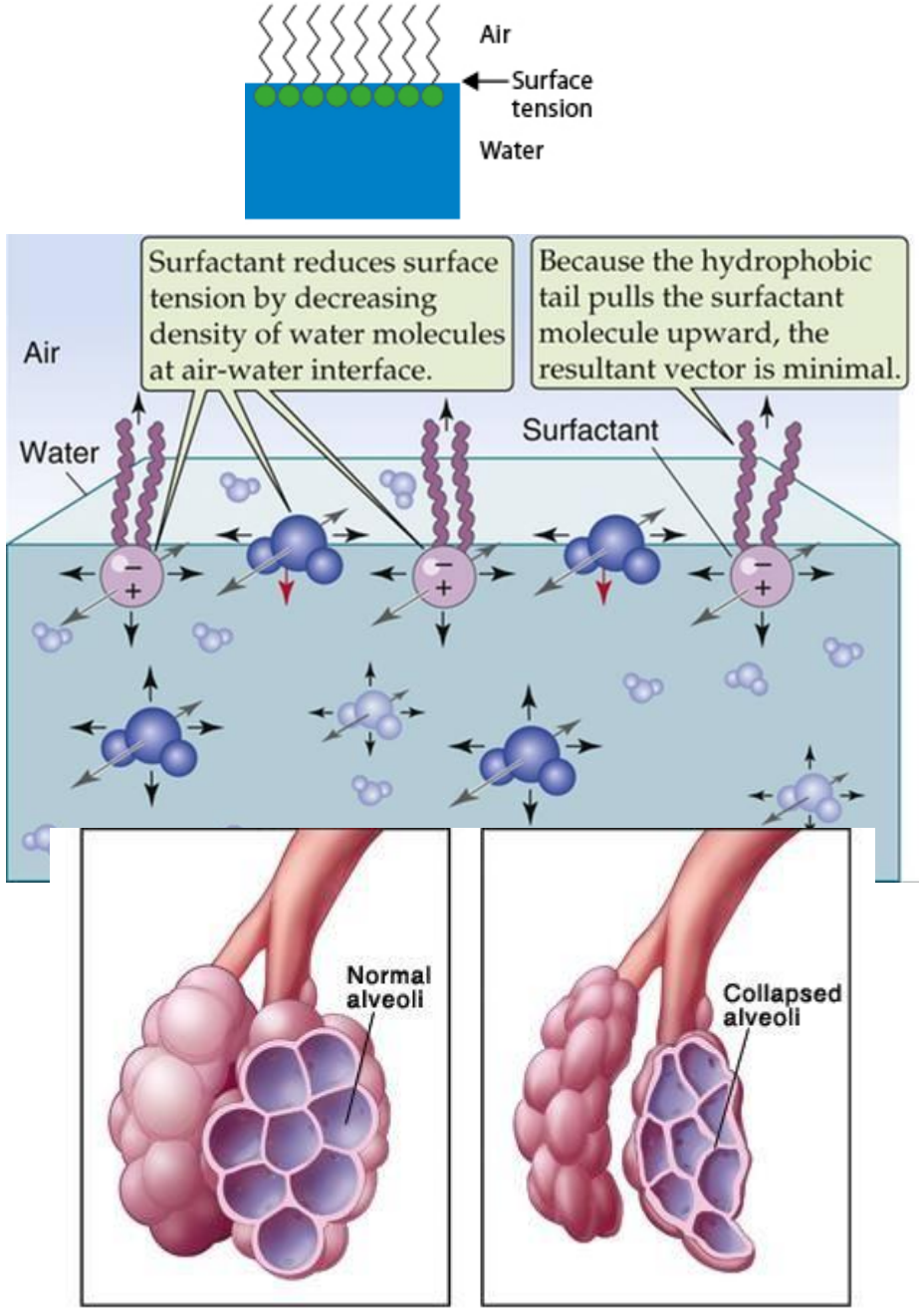
❖ **Lipoprotein lipase (LPL)** is an enzyme that:

1. **Originates** from the membrane of endothelial cells.
2. **Degrades triglycerides** in various types of circulating lipoproteins, converting them into different forms (into free fatty acids and monoacylglycerols which can the be taken up by tissues and used for energy production and storage).
3. Is **anchored** to endothelial cell membranes via **GPI anchors,** providing flexibility for its activity in blood vessels.



Application: Surfactants

- Surfactants are a complex mixture of lipids (90%) and proteins (10%) that make the extracellular fluid layer lining the alveoli and are secreted by type II pneumocytes in the lungs.
- Dipalmitoylphosphatidylcholine (DPPC) is the major lipid in surfactants.
- Surfactants serve to decrease the surface tension of the fluid layer allowing reinflation of alveoli and preventing alveolar collapse (atelectasis).
- Respiratory distress syndrome (RDS) in preterm infants is associated with insufficient surfactant production and/or secretion.
- Prenatal administration of glucocorticoids shortly before delivery to induce expression of specific genes.



Explanation- Surfactants

- The lungs are composed of a functional unit called the **alveolus**, which resembles a small sac with very thin walls.
- Inhalation makes the sacs **expand** while the opposite occurs in exhalation.
- During exhalation and **deflation** of the sacs, **surface tension** builds up inside the alveoli due to interactions between **water molecules.**
- **High surface tension** can cause the alveolar walls to **collapse**, leading to increased resistance during the next inhalation—much like inflating a balloon that's deflated – needs more effort due to surface tension.

➢ As a result, greater effort is needed to breathe; there's resistance.

- to counteract this, **type II pneumocytes** secrete a greasy material called **surfactant**, which reduces surface tension.

- Surface Tension: This is the force exerted by the liquid lining the alveoli that causes the surface to contract. It's due to the cohesive forces between water molecules.
- Role in Exhalation: During exhalation, the alveoli deflate, and the surface tension increases, helping to push air out of the lungs.

Explanation- Surfactants (Cont'd)

- Surfactant allows the alveoli to **slide easily** against each other, making inflation during breathing much easier.
- The key component of surfactant is **dipalmitoyl phosphatidylcholine** (DPPC), which is made of two palmitic acid chains attached to phosphatidylcholine.
- ➢ By reducing surface tension, surfactant ensures **effortless breathing** in healthy individuals.
- Premature babies often lack sufficient surfactant production because their lungs are underdeveloped, causing **respiratory distress syndrome** (RDS), where high resistance to breathing makes inhalation difficult.
- RDS can be treated with Dexamethasone.
- To address this issue, **glucocorticoids** like **dexamethasone** are administered to pregnant women at risk of preterm delivery, it **induces the expression** of genes responsible for producing surfactant, specifically DPPC; therefore, reducing the risk of respiratory distress in premature babies.
- Care is provided to premature babies if they still have insufficient surfactants, so they are placed in incubators to support breathing until lungs maturation.

Extra explanation

❑ Mechanism of Action:

Surfactants are **amphipathic** molecules. The hydrophobic ends of surfactant molecules insert themselves into the water layer lining the alveoli. This disrupts the **hydrogen bonds** between water molecules, which are responsible for the high surface tension.

❑ Benefits in the Alveoli:

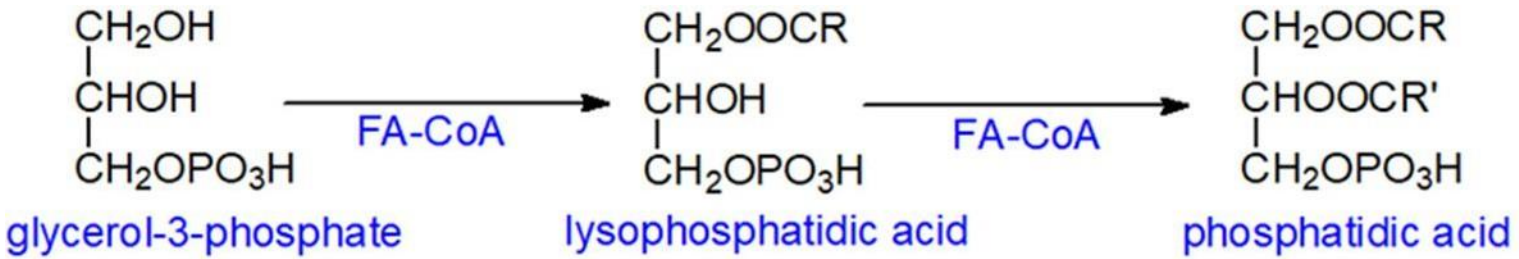
1. **Preventing Collapse:** Lower surface tension reduces the risk of alveolar collapse (atelectasis), especially during exhalation when the alveoli shrink.

2. **Ease of Re-inflation:** Reduced surface tension makes it easier for the alveoli to re-inflate during inhalation.

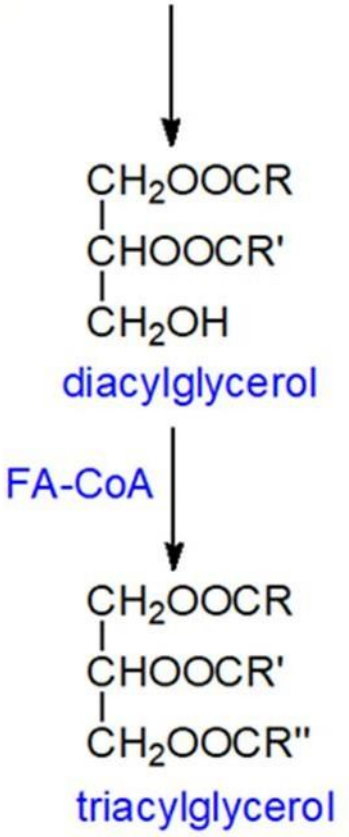
3. **Uniform Expansion:** Smaller alveoli would have higher internal pressure due to higher surface tension, but surfactant helps to equalize this pressure across alveoli of different sizes, which helps in maintaining uniform expansion of alveoli, which improves gas exchange efficiency.

- ✓ In preterm infants, a **deficiency** in surfactant can lead to respiratory distress syndrome (**RDS**). **Surfactant replacement therapy** is often used to treat this condition.
- ✓ Conditions like acute respiratory distress syndrome (**ARDS**) can benefit from treatments that enhance surfactant function.

Synthesis of phosphatidic acid (the parent GP)



Phosphatidic acid is the precursor of glycerophospholipids.

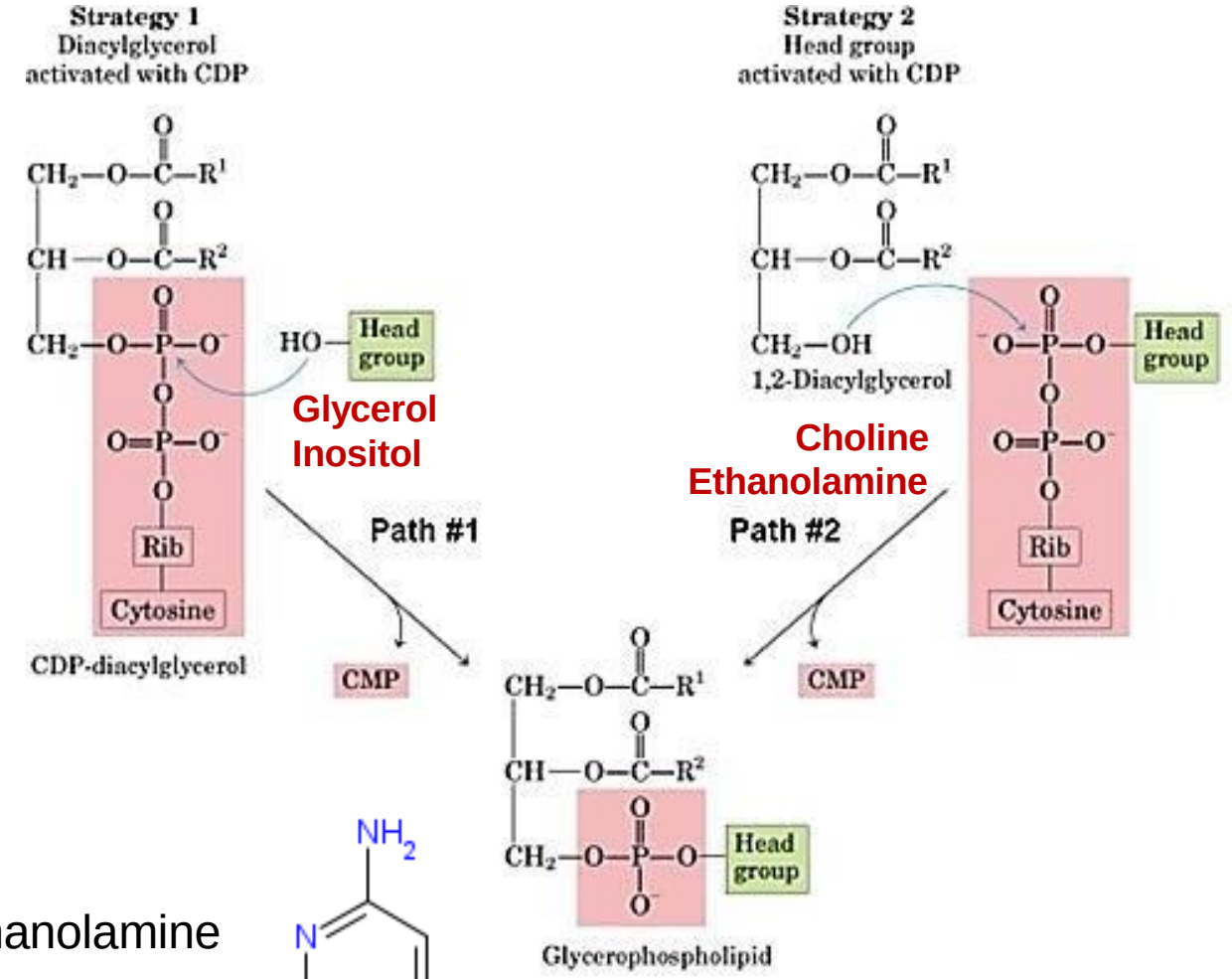


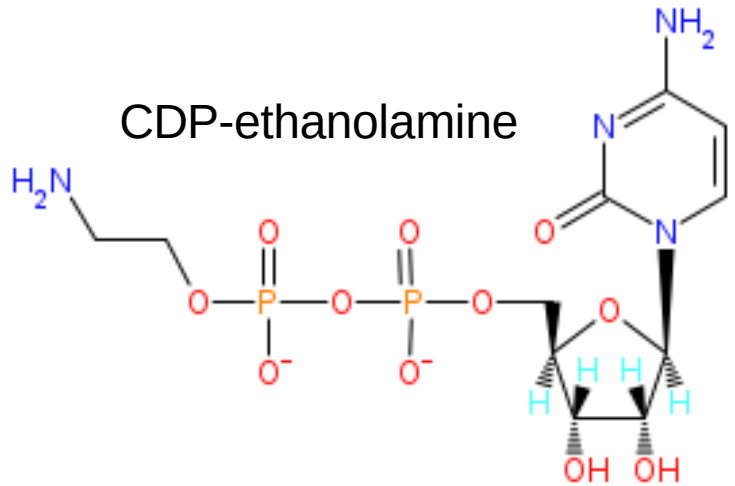
1. **Glycerol-3-phosphate** (G3P), obtained from sugar metabolism.
   - Directly as G3P or by the conversion of dihydroxy acetone phosphate (DHAP).
2. **Fatty acyl transferase enzyme** transfers a FA (activated) to the first carbon, producing **lysophosphatidic acid.**
3. Again, the transfer of the second fatty acid to the second carbon, producing **phosphatidic acid**.

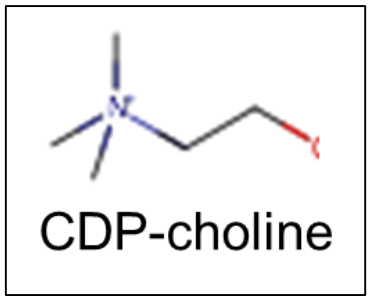
➢ For the storage of TAGs in <u>adipocytes</u>, the phosphate group can be <u>removed</u> to produce **diacylglycerol**, then the third FA is added to it, producing triacylglycerol (TAG). →(synthesis!)

Synthesis

- Location: smooth ER
  - Except for ether lipids
- Activation by CDP is necessary. Either:
  - CDP-DAG (glycerol, inositol)
  - CDP-alcohol (choline, ethanolamine)
- Sources of choline and ethanolamine
  - diet
  - synthesis
  - re-cycling from the turnover of pre-existing phospholipids
- Diet is still essential since demand > supply







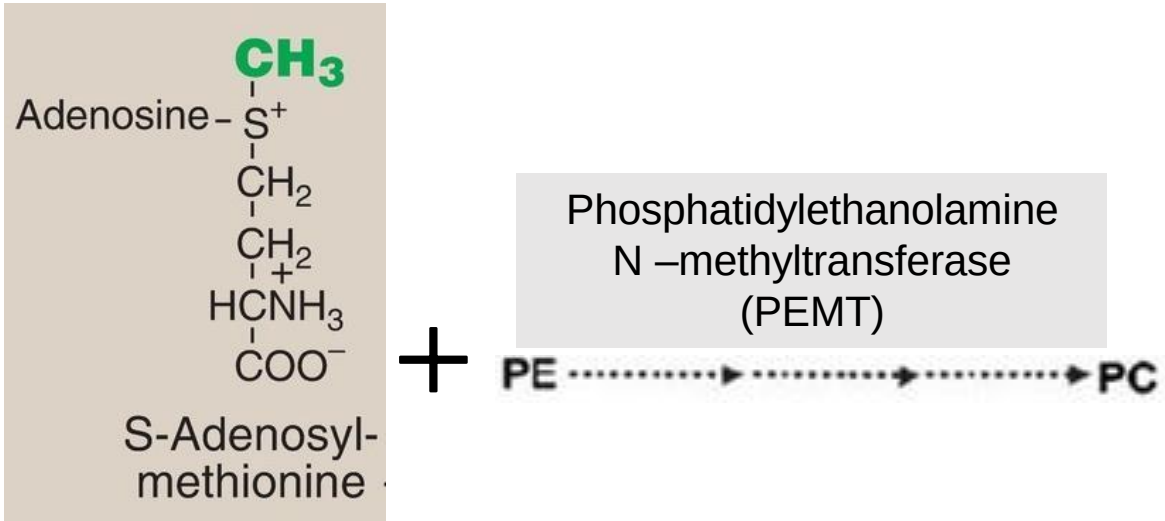
The nucleotide initially enters as CTP and then becomes CDP once added, just like addition of UTP.

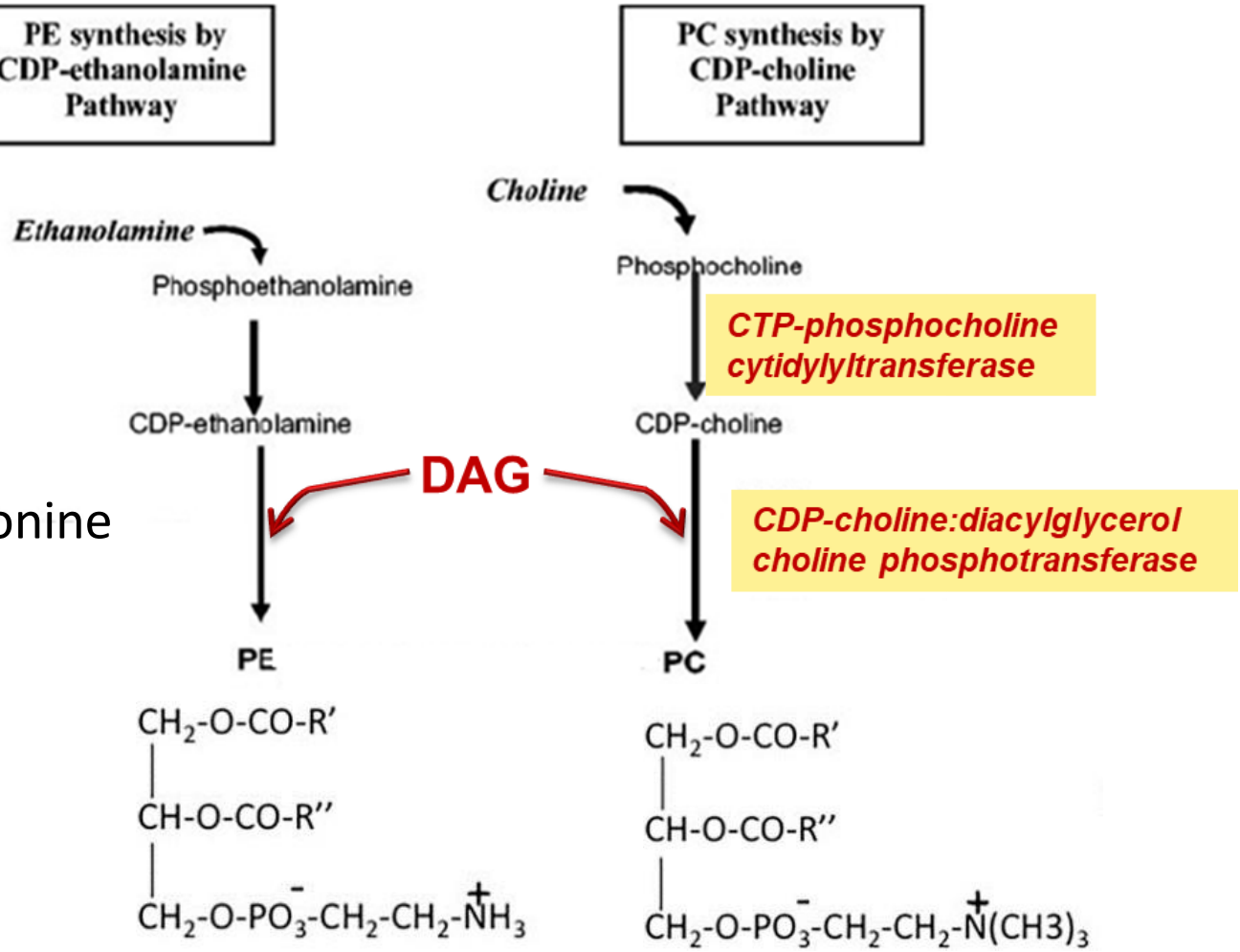
Synthesis of *ph*-choline and *ph*-ethanolamine

- Choline or ethanolamine are phosphorylated by *kinases*, then activated by *transferases* to form, CDP-choline or CDP-ethanolamine.
- Choline phosphate or ethanolamine phosphate is transferred from the nucleotide (releasing CMP) to DAG.

➢ Synthesis of ph-choline from ph-ethanolamine

- Methyl groups are donated by S-adenosylmethionine to convert PE to PC by PE methyltransferase.





Synthesis, generally and specifically[1].

Ph-choline and Ph-ethanolamine.

- The creation of phospholipids like **phosphatidylcholine** and **phosphatidylethanolamine** involves similar steps (as phosphatidic acid) but varies based on the specific head group used (either choline or ethanolamine).

❖ steps:

1. Head groups are added to CTP nucleotides by a **transferase**. (CDP-head group)
2. Diacylglycerol (DAG) is obtained.
3. The head groups are **transferred** to DAG using a **CDP nucleotide**, by the action of a **phosphotransferase**.
4. After the attachment of the head group, CDP nucleotide is **cleaved** (the cut of phosphate, ribose, and cytosine), producing **cytidine mononucleotide** (CMP).
5. Now, depending on the head group; either phosphatidylcholine or phosphatidylethanolamine is produced.

Synthesis, generally and specifically(2).

Others, ex. Ph-inositol and Ph-glycerol.

- For the synthesis of others, the process is similar but the order is different.

❖ Brief steps:

1. **CDP nucleotide**, but instead of carrying the head group, now it carries **DAG** (CDP-DAG).
2. Addition of the **head groups to DAG** by a transferase.
3. Cleavage and **removal** of **CMP**.
4. **Production** of GPs.

➢ Notes about synthesis:

1. The synthesis occurs primarily in the **SER**, but they can come from diet.
2. Phospholipid production doesn't always start from scratch; existing molecules can be **recycled**. For example, **phosphatidylethanolamine** can be converted into **phosphatidylcholine** when needed, which adds flexibility to the body's lipid synthesis process.

phosphatidylcholine is an emulsifier in different food products.

Explanation- Recycling Process.

- Recall structural differences between choline ($-N(CH_3)_3^+$) and ethanolamine ($-NH_3^+$).
- By a methylation reaction, PE can be converted to PC by the action of a **methyltransferase**.
- **Phosphatidylethanolamine N-methyltransferase** (PEMT), adds methyl groups to PE to be converted to PC.
- *From where do we get these methyl groups?*

➢ Addition of a single carbon unit, this carbon unit could be (1)Methyl group ($-CH_3$), (2)formyl group ($-C=O$) or (3)formimino group ($-CHNH$), etc.

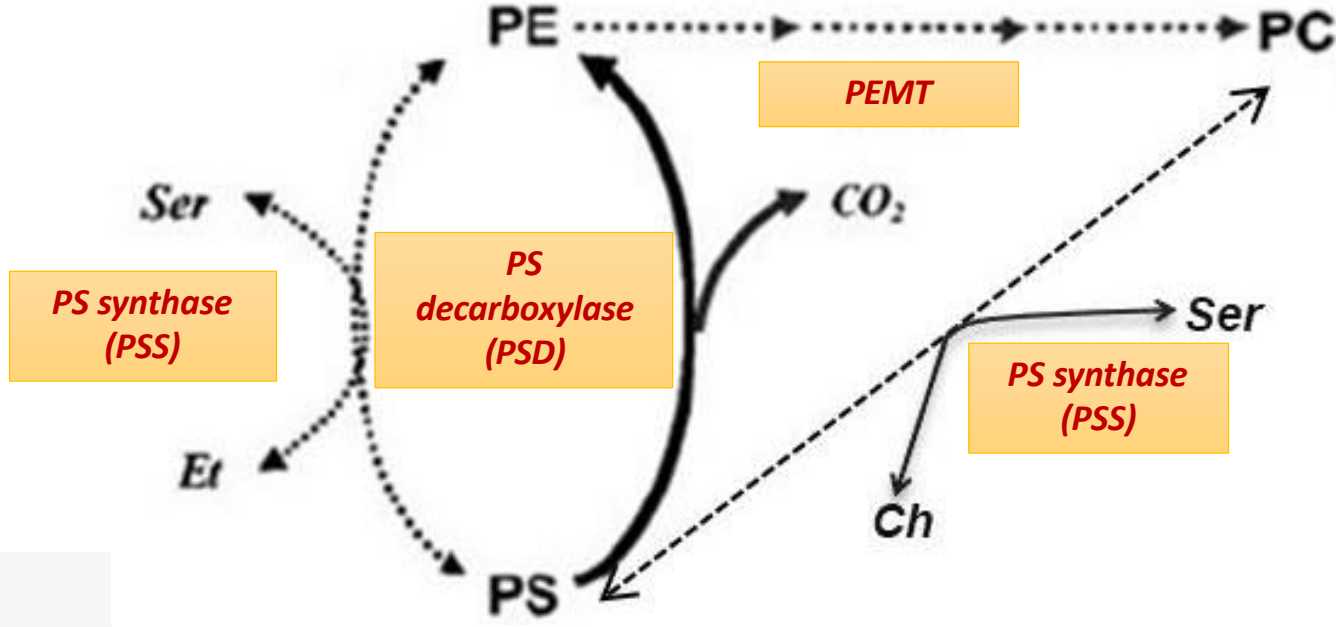
➢ **S-adenosylmethionine** (SAM) that comes from methionine metabolism, transfers **methyl** groups. That is the terminal carbon of methionine can be **cleaved** and used for this transfer of methyl group to a recipient.

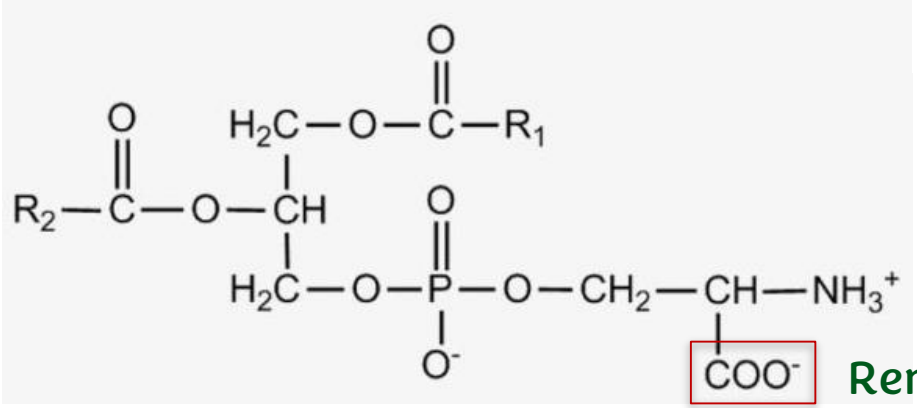
➢ Also, folic acid (**vit. B9**) can transfer methyl groups or other single carbon units → More diversity.

PE = Phosphatidylethanolamine
PC = Phosphatidylcholine

Synthetic pathways for and from ph-serine

- The liver requires another mechanism to produce PC because it uses it to make bile and other plasma lipoproteins.
- PS is decarboxylated to PE by PS decarboxylase (PSD). It can be methylated by PEMT to PC
- PS is exchanged from PE or PC by PS synthases (PSS).





phosphatidylserine

Removal of the carboxyl group from PS, will yield PE.

Metabolism of phosphatidylserine(PS)

❖ exchange between PE and PS:

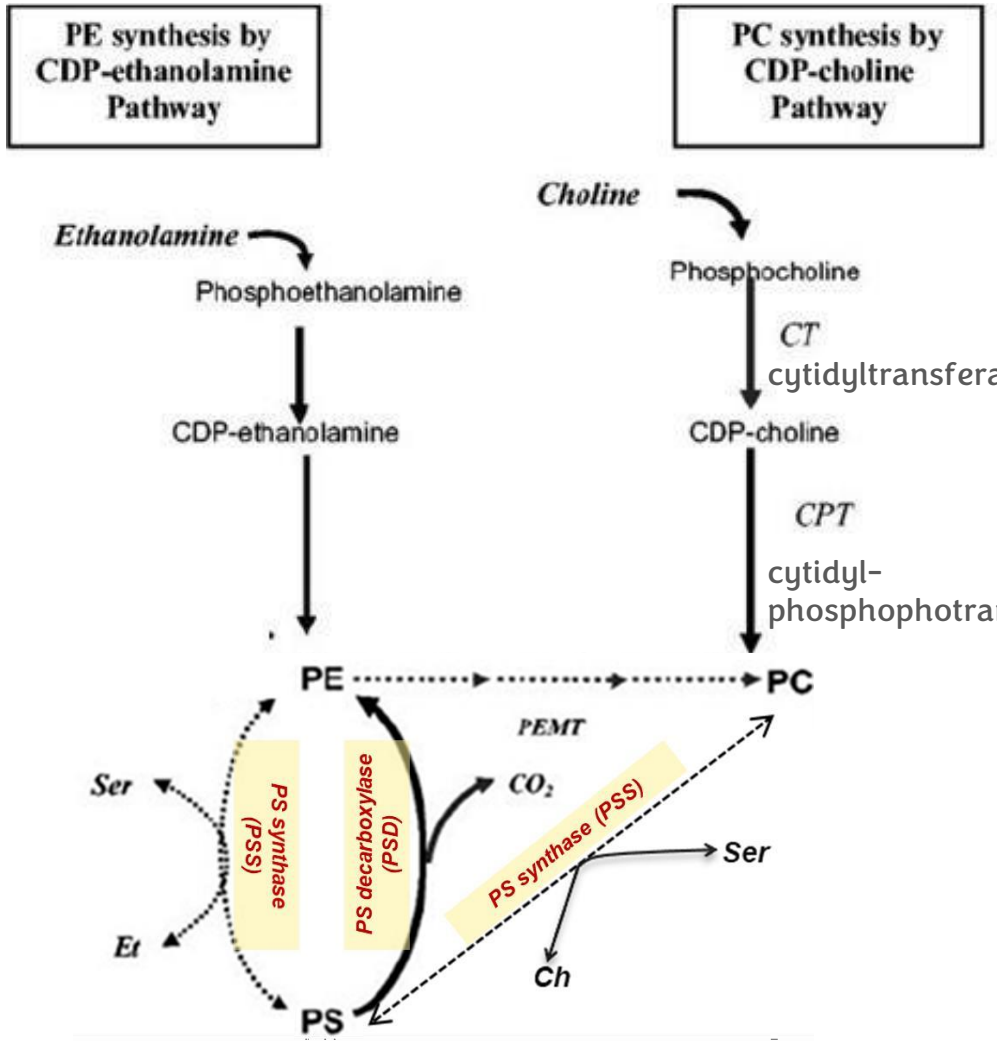
- By a **reversible** exchange reaction, PS can be produced from PE, by replacing ethanolamine with serine A.A using phosphatidylserine **synthase**, or the opposite, to produce PE instead.
- In addition, decarboxylation of PS gives PE.

❖ exchange between PC and PS:

- Phosphatidylserine synthase can also remove the choline and replace it with serine; producing PS from PC in a reversible manner.

Summary of synthesis pf PE, PC, and PS

1. Phosphorylation of the head group (choline or ethanolamine) by kinases.
2. Attach the head group to a CDP molecule (which entered as CTP) by a transferase.
3. CDP-head group (choline or ethanolamine) is transferred to a diacylglycerol using a phosphotransferase to produce PC and PE.



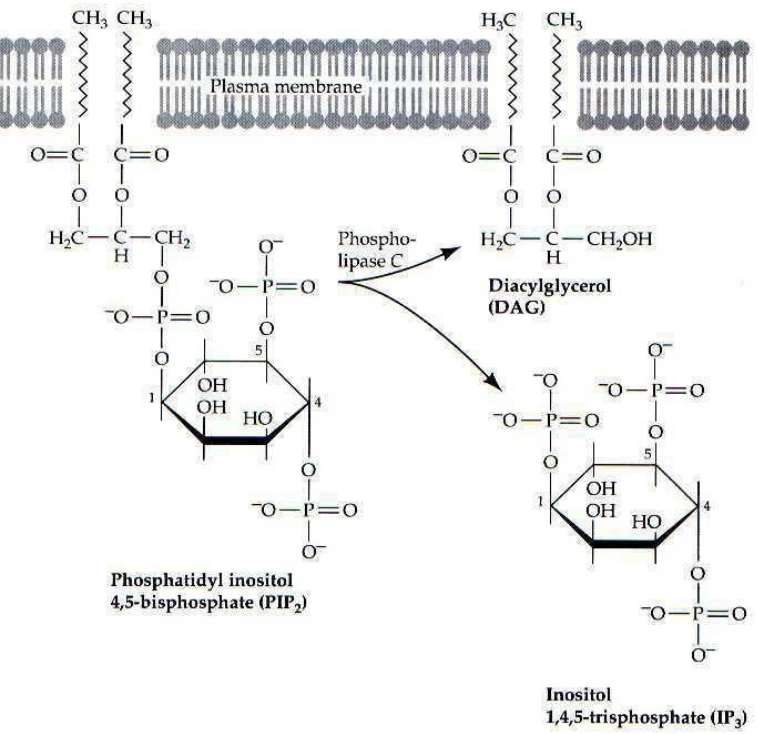
- ❖ Pathways connecting different types:

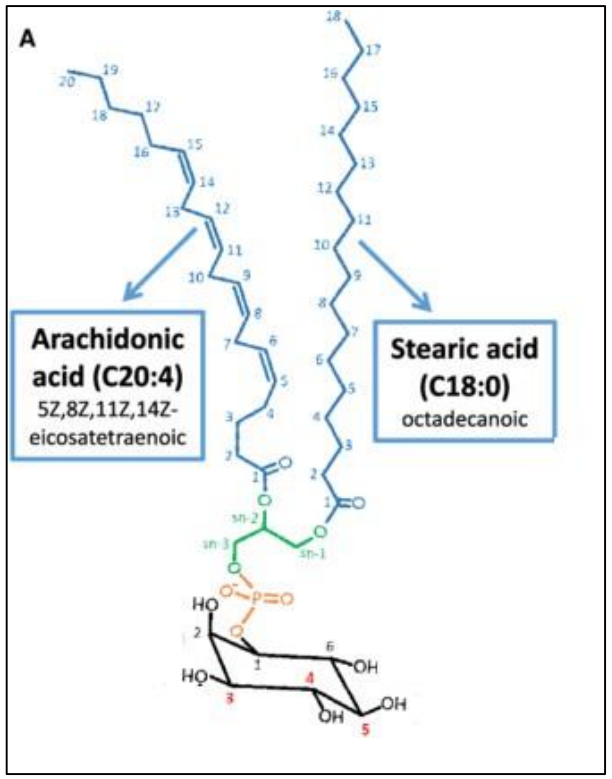
1. Methylation of PE to make PC.
2. Decarboxylation of PS to make PE.
3. Exchange reactions by PS synthase:

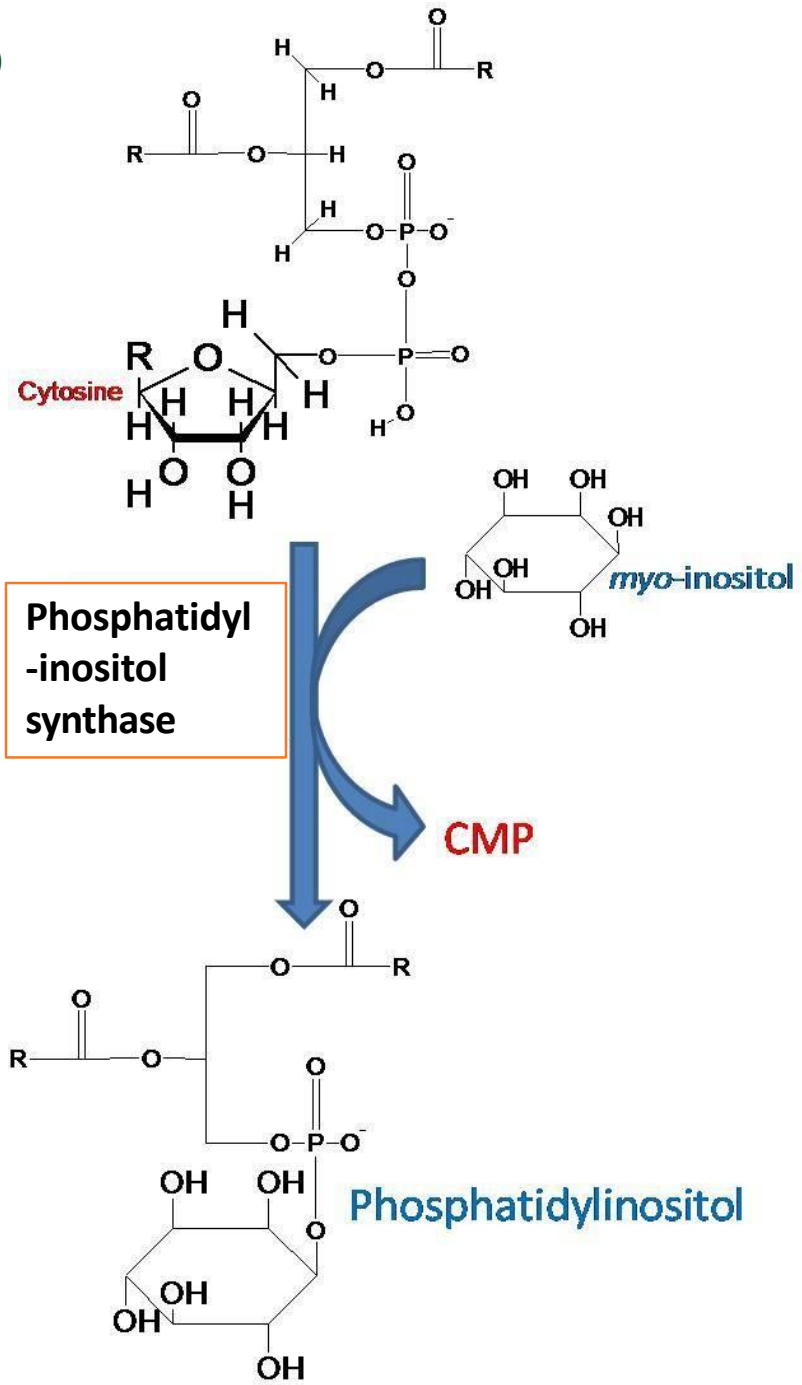
a) Exchange of serine with ethanolamine (reversible)
b) Exchange of serine with choline (reversible)

Synthesis of ph-inositol (1st pathway)

- Inositol is combined with CDP-DAG by PI synthase to produce phosphatidylinositol.
- It is a reservoir of arachidonate.
- It also produces signaling molecules when cleaved by phospholipase C.







Phosphatidylinositol (PI) synthesis

❖ Steps of synthesis: same as before- repeated.

1. DAG is carried by **CDP**, the activated form: CDP-DAG.
2. Inositol is added to the phosphate group on DAG through the action of phosphatidylinositol synthase.
3. CMP is removed, and PI in produced.

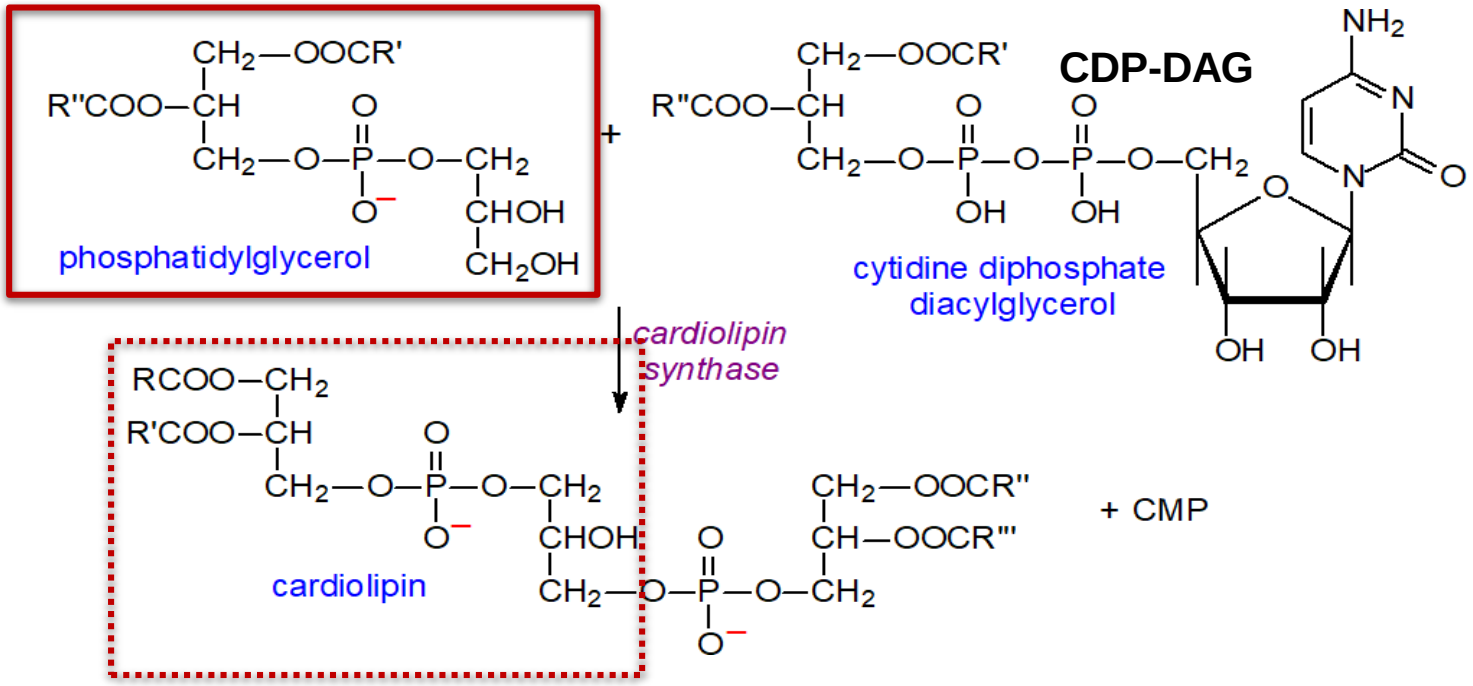
➢ Roles of phosphatidylinositol:

a) Critical for forming GPI anchors, attaching proteins to the plasma membrane.
b) plays a key role in signaling pathways, particularly as a precursor for **PIP2** (Phosphatidylinositol 4,5-bisphosphate).
c) Arachidonic acid reservoir; as its FAs composition is majorly Arachidonic acid and steric acid.

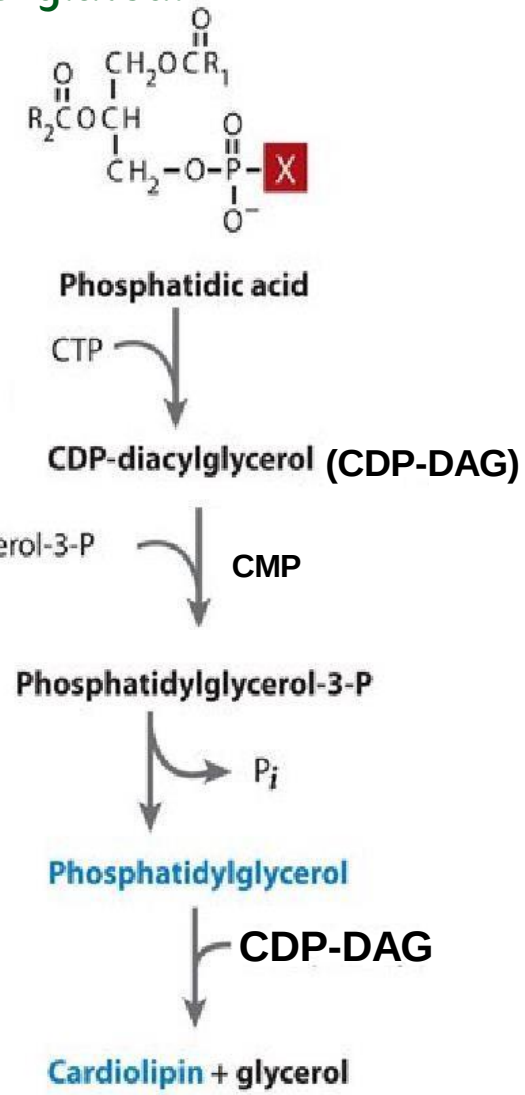
Phosphatidylglycerol and cardiolipin

- Phosphatidylglycerol is synthesized from CDP-DAG and glycerol 3-phosphate.
- Cardiolipin is synthesized by the transfer of DAG from CDP-DAG to a pre-existing molecule of phosphatidylglycerol.

Same as PI in mechanism (as both are alcohols), but the only difference is that here glycerol is phosphorylated and then dephosphorylated.



A phosphate group is required to allow the attachment, which then detaches



Cardiolipin synthesis

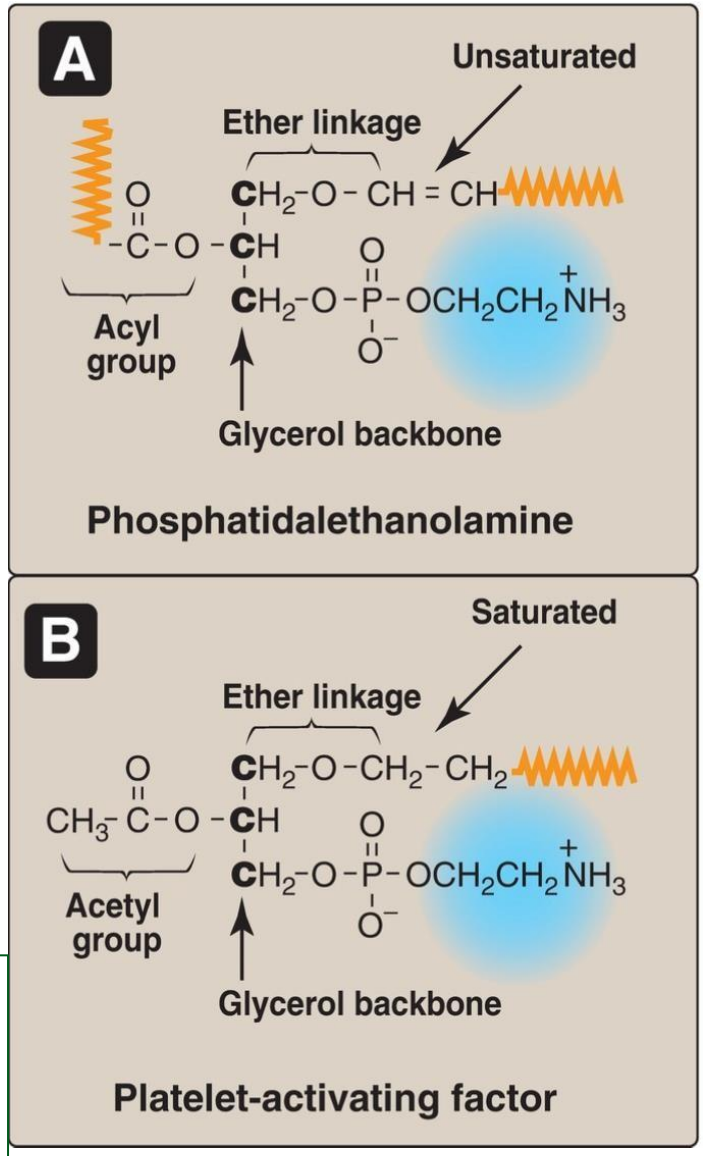
Recall,
Cardiolipin = 2 phosphatidic acids + 1 glycerol.
Phosphatidylglycerol = phosphatidic acid + glycerol.
So, what is left is adding a phosphatidic acid to the phosphatidylglycerol

1. Cardiolipin synthesis begins with phosphatidic acid and phosphatidylglycerol as key precursors.
2. Phosphatidic acid is activated and converted to CDP-diacylglycerol, following a mechanism similar to other CDP-diacylglycerol processes.
3. CDP group is removed and released as CMP.
4. Phosphatidic acid (activated) is then added to phosphatidylglycerol through the action of cardiolipin synthase, linking the molecules together to form cardiolipin (commonly found in the inner mitochondrial membrane).

Ether glycerophospholipids Ex. plasmalogens

- The FA at carbon 1 is replaced by an unsaturated
- alkyl group attached by an ether linkage.

- Plasmalogens: PhosphatidAl**ethanolamine** (abundant in **nerve** tissue, is similar in structure to phosphatidylethanolamine.
- PhosphatidAl**choline** (abundant in **heart** muscle) is another significant ether lipid in mammals.
- Platelet-activating factor (**PAF**) has a **saturated** alkyl group in an ether link to carbon 1 and an **acetyl** residue at carbon 2 of the glycerol backbone.
  - Prothrombotic and inflammatory factor



❖ Differences in PAF:

1. Ethanolamine head group
2. FA on C2 is an Acetyl group (very short)
3. Ether linkage is present but the hydrocarbon chain is saturated (no double bond after the ether linkage)

Ether glycerophospholipids

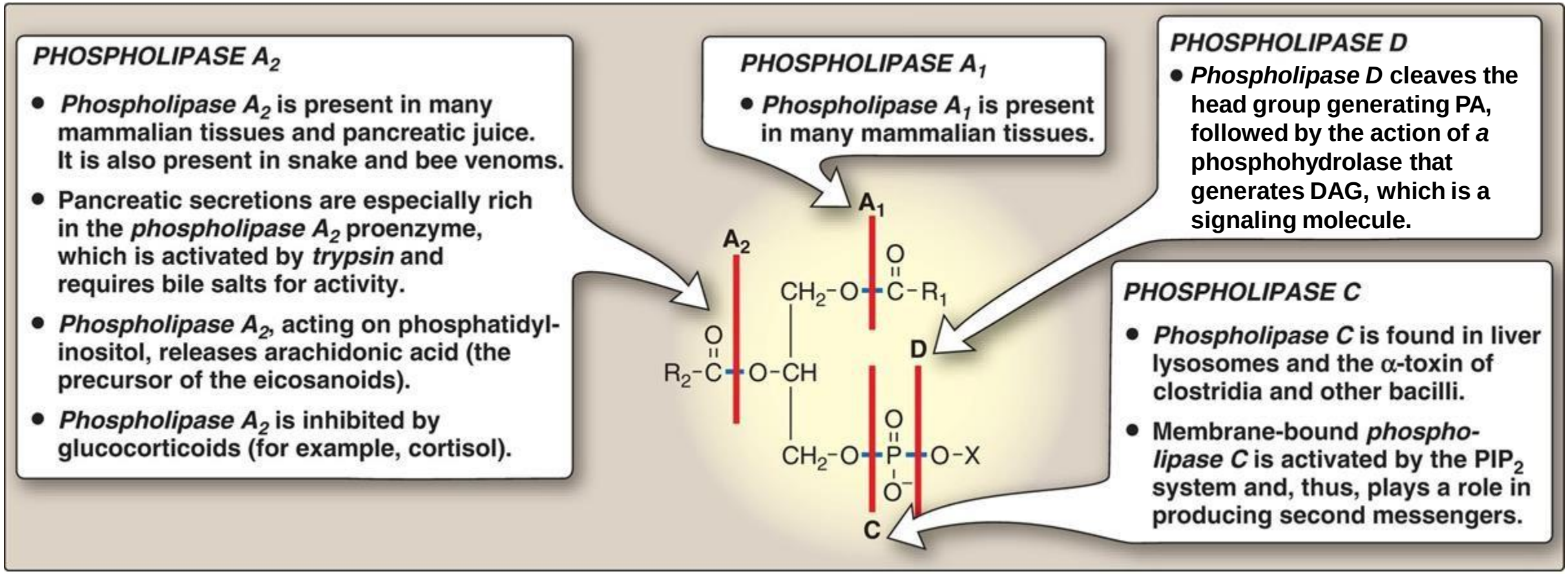
- Plasmalogens are a type of phospholipid with a structure similar to regular phospholipids but with a key difference:
  - They have an **ether linkage** at the first position of the glycerol backbone instead of an ester linkage.

➢ This first tail is a **hydrocarbon chain**, not a fatty acid, and is attached via an oxygen derived from the hydroxyl group of glycerol.

- During synthesis, normal phospholipids such as PC or PE are initially formed, and the fatty acid at the first position is replaced with a hydrocarbon chain, exchange process.
- This replacement requires specific requirements; the hydrocarbon chain must be (1)**unsaturated** with a (2)**single** double bond located between the first and second carbons.
- If the head group is ethanolamine, the resulting plasmalogen is referred to as PhosphatidAlethanolamine (not phosphatidYlethanolamine)

✓ Plasmalogens serve as specialized forms of phospholipids with distinct structural features.

Degradation of Phospholipids



Degradation of Phospholipids

- Phospholipid degradation involves various enzymes, primarily **phospholipases**, which break down phospholipids in different ways depending on the purpose.

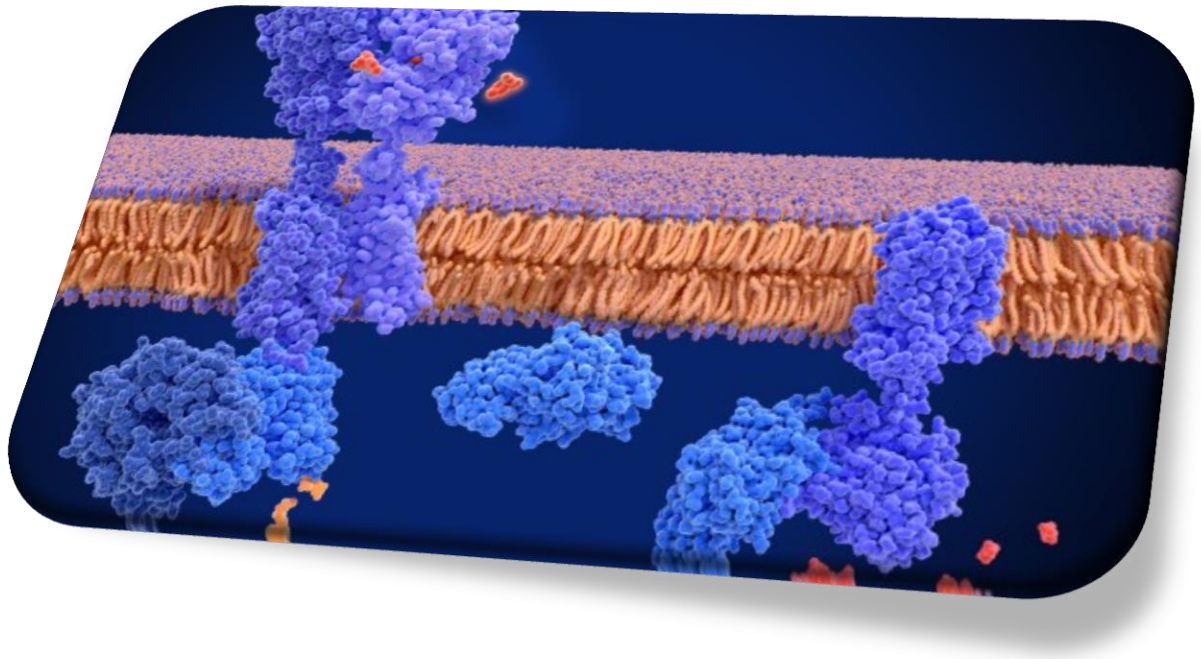
➢ The main types of phospholipases and their functions are:

1. **Phospholipase A**:
   - Phospholipase **A1**: Removes the fatty acid attached to carbon **1** of the glycerol backbone.
   - Phospholipase **A2**: Removes the fatty acid attached to carbon **2**.
   - ➢ This involves the **hydrolysis of the ester** bond between fatty acids and glycerol.
2. **Phospholipase C**: Removes the head group along with the phosphate group, leaving diacylglycerol (DAG).
   - ✓ This enzyme plays a key role in GPCR signaling pathways, as DAG is an important signaling molecule (activates protein kinase C).
3. **Phospholipase D**: Removes only the head group, leaving behind phosphatidic acid, which can serve as a precursor for diacylglycerol.

Summary

- ✓ Glycerophospholipids = glycerol + 2 FAs + phosphate group + head group.
- ✓ Different head groups result in distinct functions of different molecules.
- ✓ GPI anchoring of membrane proteins provides them with flexibility of movement through the membrane.
- ✓ PIP2 is cleaved by phospholipase C downstream in the GPCR pathway, producing DAG and IP3, which eventually activate protein kinase C.
- ✓ Surfactants reduce surface tension in the walls of the alveoli, preventing their collapse and easing our breathing.
- ✓ Phosphatidic acid, the parent GP, is synthesized from G3P by adding two FAs (-phosphate = G3P).
- ✓ PC and PE are produced by the addition of their head groups (carried by CDP) to DAG.
- ✓ PC, PE, and PS can be produced from each other through various reactions (methylation, decarboxylation, and transfer of head groups).
- ✓ PG and PI are produced by the addition of their head groups to DAG that is bound to CDP.
- ✓ Cardiolipin is produced by the addition of phosphatidic acid (carried by CDP) to PG.
- ✓ Ether glycerophospholipids (plasmalogens and platelet-activating factor) are similar to GP but have an ether bond at C1 of glycerol instead of an ester bond.
- ✓ Phospholipids are degraded by phospholipases, which have different types and produce different products.
- ✓ Phospholipase A removes FAs, phospholipase C removes the head group alongside the phosphate leaving DAG, and phospholipase D removes only the head group leaving phosphatidic acid.

Quiz on this lecture



For any feedback, scan the code or click on

Corrections from previous versions:

Versions	Slide # and Place of Error	Before Correction	After Correction
V0 → V1	18	The nucleotide initially enters as CTP and then becomes CMP once added	The nucleotide initially enters as CTP and then becomes CDP once added
	34	GAP was misplaced instead of G3P and DAG multiple times.	Corrected abbreviations.
V1 → V2			

Additional Resources:

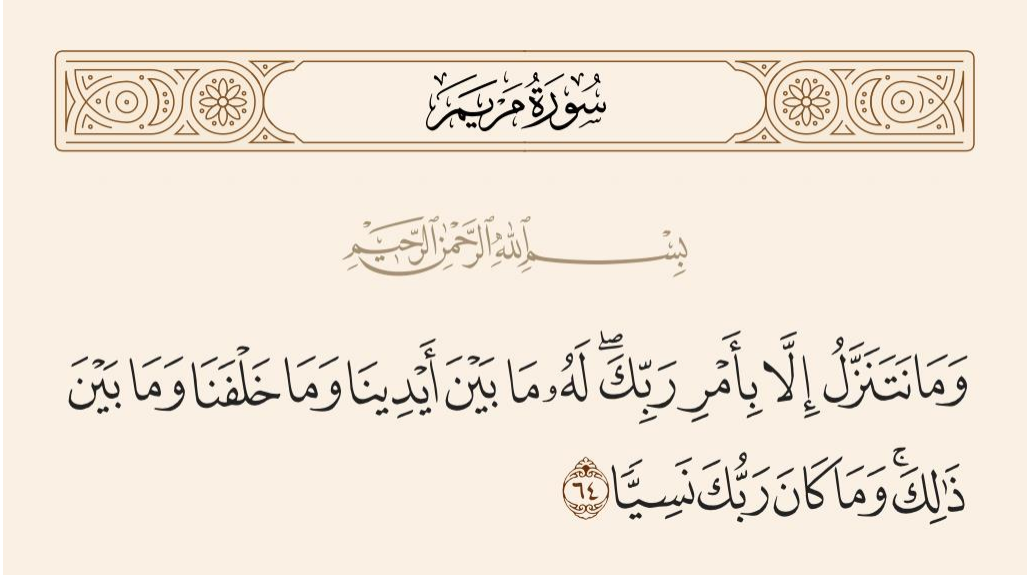
رسالة من الفريق العلمي:

Reference Used:

1. PI and Cardiolipin

Extra References for the Reader to Use:

1. GPs
2. Activation of protein kinase C



https://www.instagram.com/reel/DCuMNDYAdKx/?igsh=cHk2eDBqOHJ0dW96 Reasonable crash out