

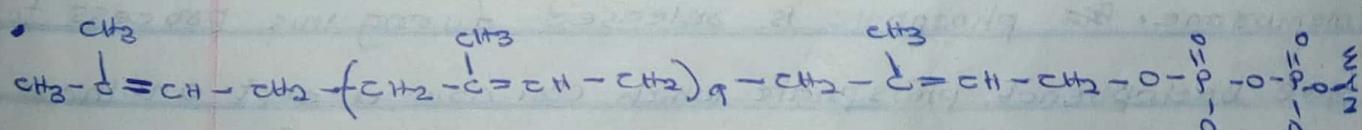
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BCH 403.

Mr. Somadev.

PEPTIDOGLYCAN SYNTHESIS.

Peptidoglycan is a complex component of bacteria cell wall - most bacteria cell walls contain a large complex peptidoglycan molecule consisting of long polysaccharide chains made of alternating N-acetyl muramic acid [NAM] and N-acetyl glucosamine [NAG] residues. Peptide chains are attached to the NAM groups. The polysaccharide chain are connected through their pentapeptides or by interbridges. These synthesis reactions occur in two places i.e. both inside & outside of cell membrane. Peptidoglycan synthesis is a multi-step process that has been best studied in a gram positive bacterium called *Staphylococcus aureus*. Two carriers participate, uridine diphosphate [UDP] and bactoprenol. Bactoprenol is a 3S-carbon alcohol that attaches to NAM by a pyrophosphate group (ppi) and moves peptidoglycan components through the hydrophobic membrane.



The synthesis of peptidoglycan occurs in 8 steps

- ① UDP-derivatives of NAM and NAG are synthesized in the cytoplasm.
- ② Amino acids are sequentially added to UDP-NAM to form the peptidoglycan chain [the two terminal D-alanine are

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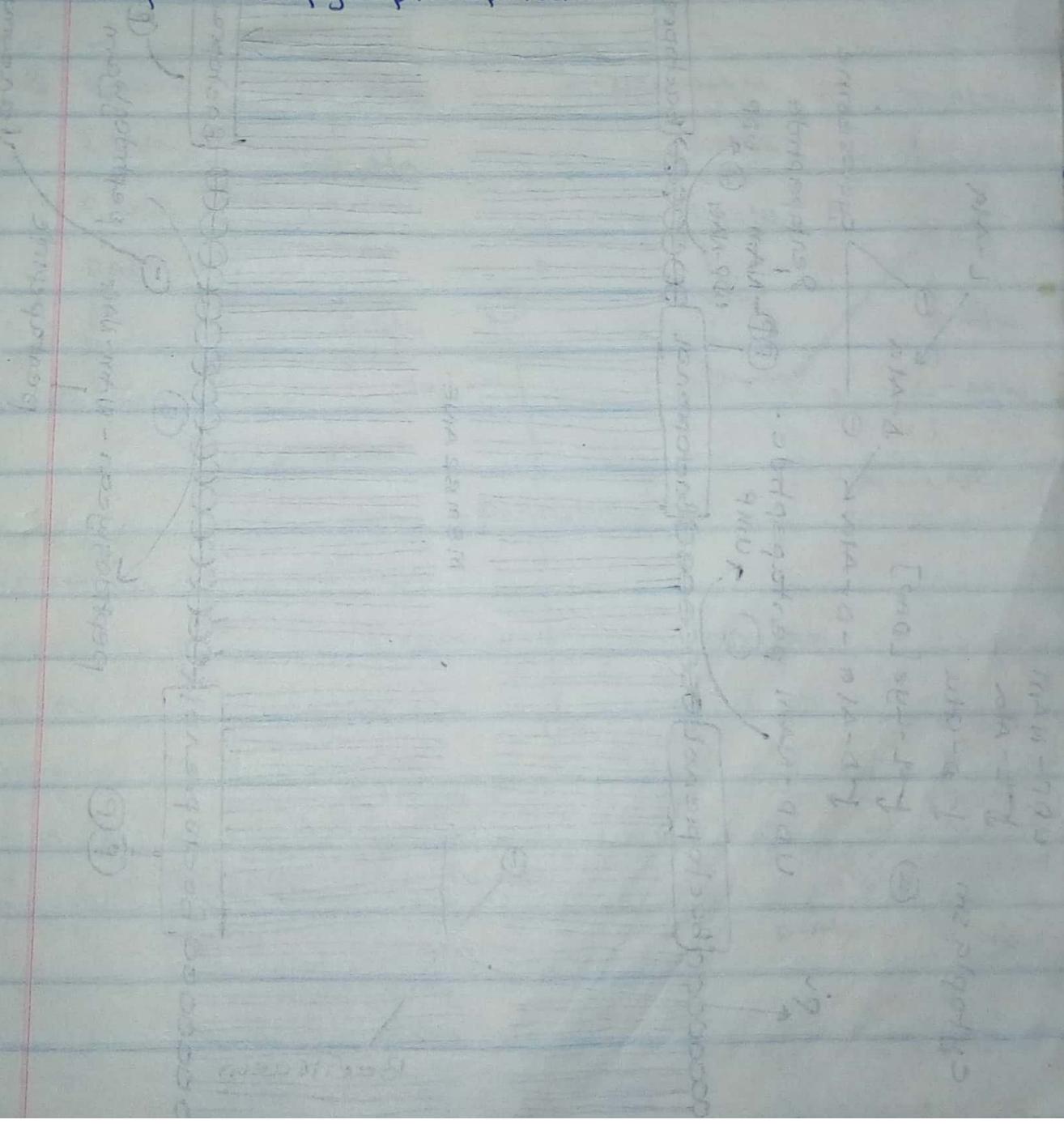
added as a dipeptide). ATP energy is used to make the peptide bonds, but transfer RNA and ribosomes are not involved.

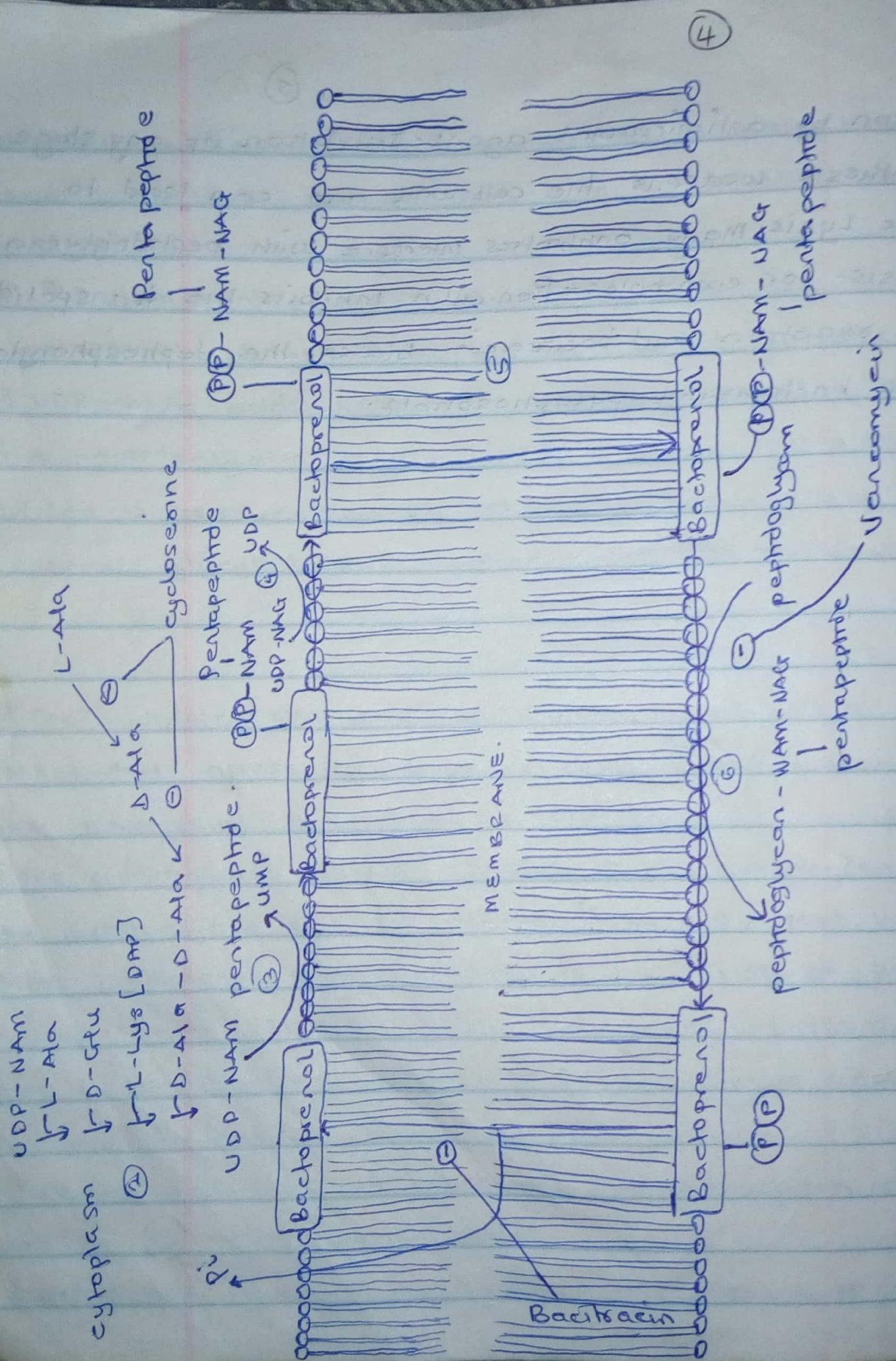
- ③ The NAM-penta-peptide is transferred from UDP to a bactoprenol phosphate at the membrane surface.
- ④ UDP-NAG adds NAG to the NAM-penta-peptide to form the peptidoglycan repeat unit. If a pentaglycine inter-bridge is required, the glycine's are added using special glycyl t-RNA molecules, not ~~ribosomes~~.
- ⑤ The completed NAM-NAG peptidoglycan repeat unit is transported across the membrane to its ^{outer} ~~inner~~ surface by the bactoprenol pyrophosphate carrier.
- ⑥ The peptidoglycan unit is attached to the growing end of the peptidoglycan chain to lengthen it by one repeat unit.
- ⑦ The bactoprenol carrier returns to the inside of the membrane. ^A Phosphate is released during this process to give bactoprenol phosphate which can now accept another NAM peptide.
- ⑧ Finally, peptide cross-link between the peptidoglycan chains are formed by transpeptidation.

Peptidoglycan synthesis is particularly vulnerable to

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disruption by antimicrobial agent. Inhibition of any stage of synthesis weakens the cell wall and can lead to osmotic lysis. Many antibiotics interfere with peptidoglycan synthesis. For example:- Penicillin inhibits the transpeptidation reaction and bacitracin blocks the dephosphorylation of bactoprenol pyrophosphate.

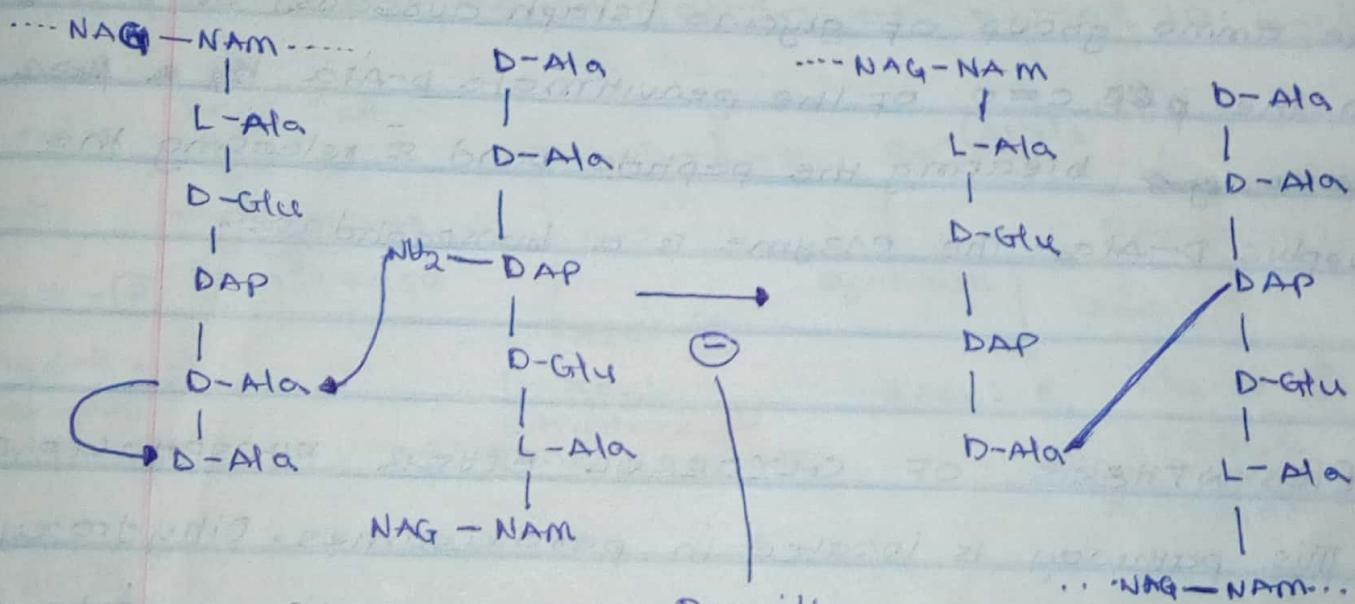




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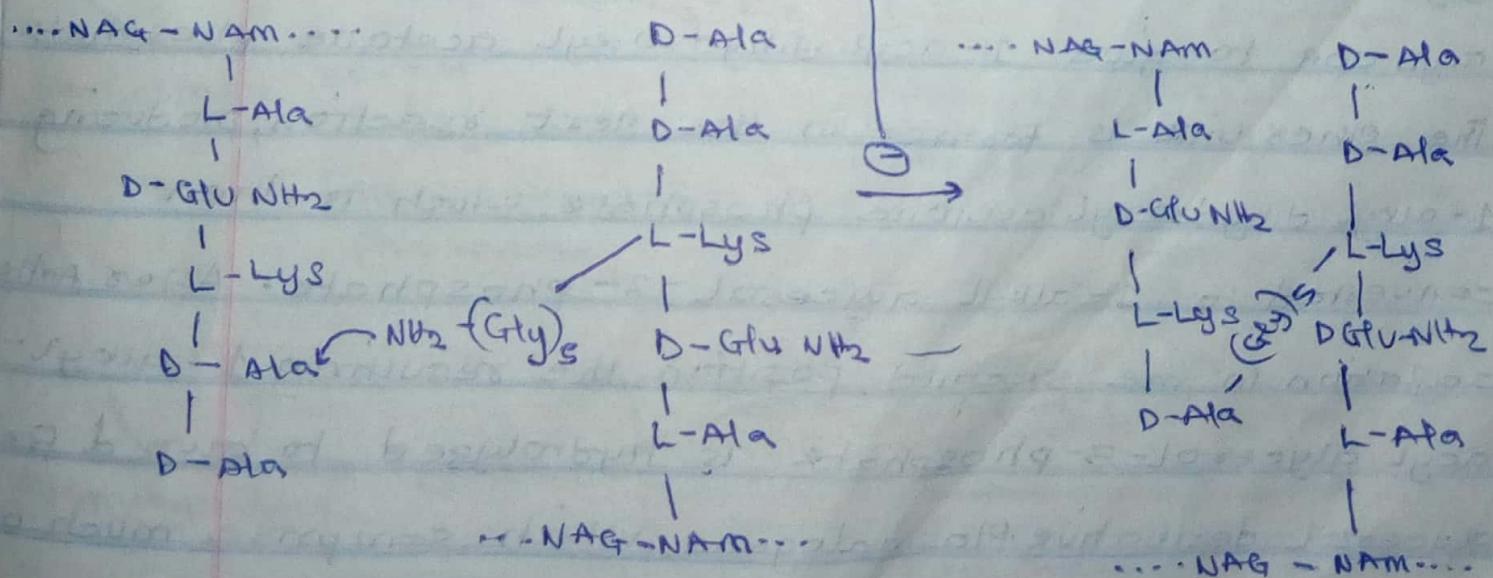
The pentapeptide contains L-Lysine in *Staphylococcus aureus* peptidoglycan, and diaminopimelic acid [DAP] in *E-coli*. Inhibition by bacitracin, cycloserine and Vancomycin has been shown in the diagram.

E-coli transpeptidation. [Gram Negative].



[Gram positive]

S. aureus transpeptidation.



PAF, plasmalogen \Rightarrow e.g. of glycerol ether PL.

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The scheme above is the transpeptidation reactions in the formation of the peptidoglycans of E. coli and Staph aureus.

TRANSPEPTIDATION is the process of cross linkage of peptide chains to produce the insoluble, strong mesh of peptidoglycan. It involves enzymes-catalyzed attack by a free amino group of glycine [Staph aureus] or DAP [E. coli] on the ~~glu~~ C=O of the penultimate D-Ala ~~ring on R side~~ breaking the peptide bond & releasing the surplus D-Ala. The enzyme is a transpeptidase.

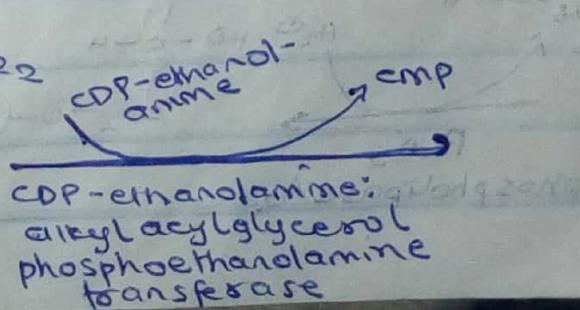
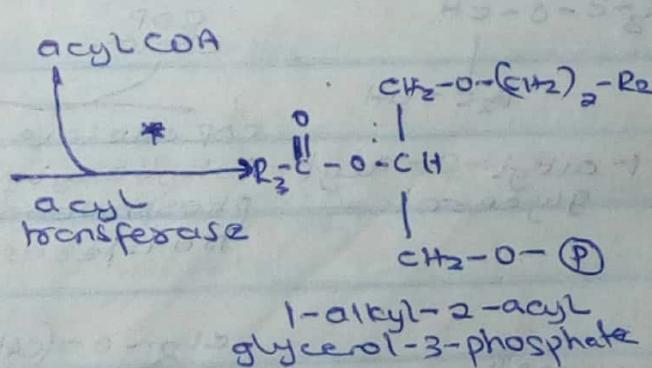
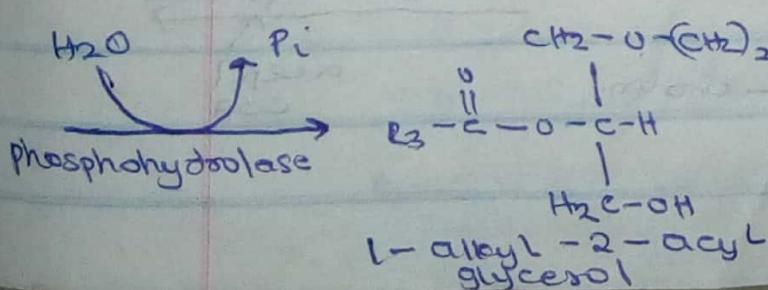
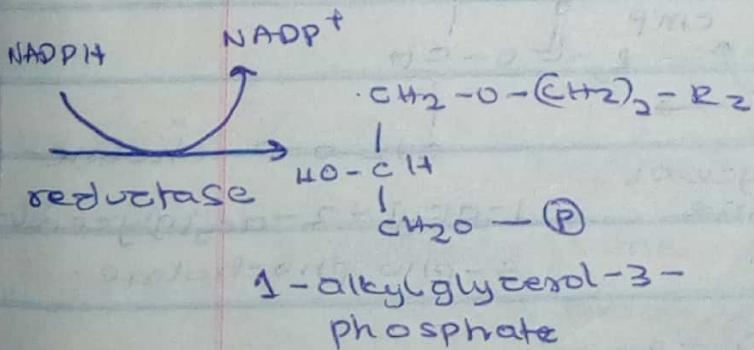
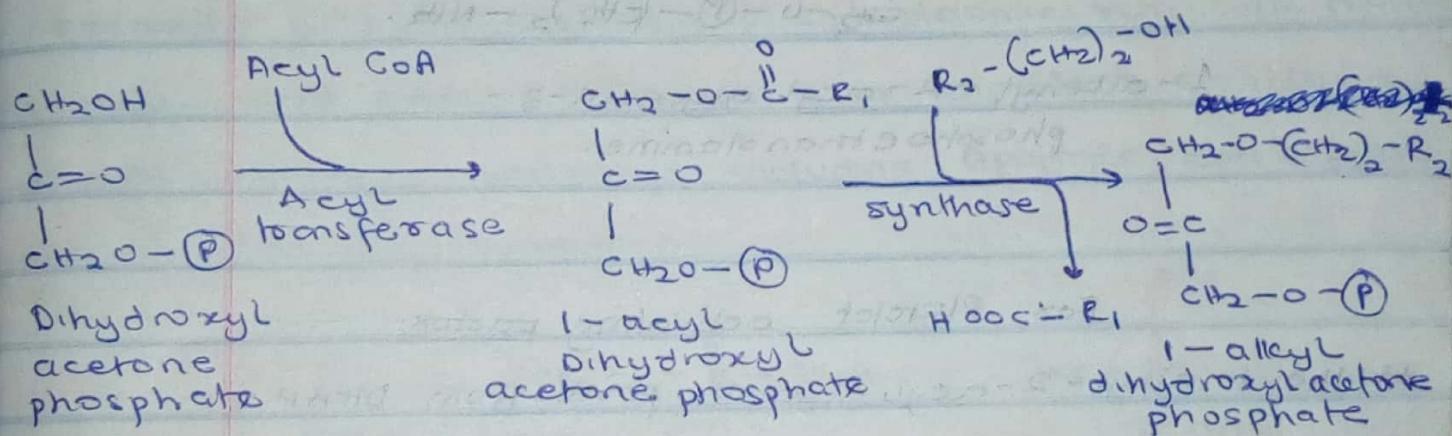
BIOSYNTHESIS OF GLYCEROL ETHER PHOSPHOLIPIDS.

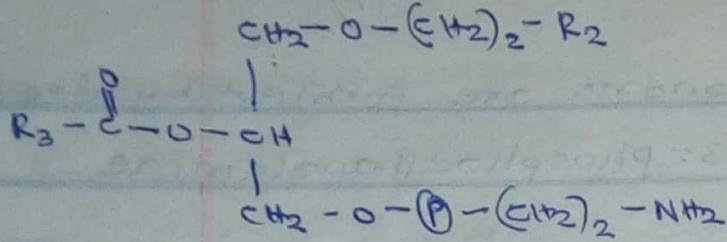
This pathway is located in peroxisomes. Dihydroxyacetone phosphate is the precursor of the moiety of glycerol ether phospholipid. This compound combines with acyl CoA to give 1-acyl dihydroxyacetone phosphate. The ether link is formed in the next reaction, producing 1-acyl dihydroxyacetone phosphate, which is then converted to 1-acyl glycerol-3-phosphate. After further acylation in the second position, the resulting 1-acyl-2-acyl glycerol-3-phosphate is hydrolyzed to give a free glycerol derivative Plasmalogens, which comprise much of

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The phospholipid in mitochondria are formed by desaturation of the analogous 3-phosphoethanolamine derivatives.

Platelet activating factor [PAF] is synthesized from the corresponding 3-phosphocholine derivative. It is formed by many blood cells and other tissues and aggregates platelets at concentration as low as 10^{-11} mol/L



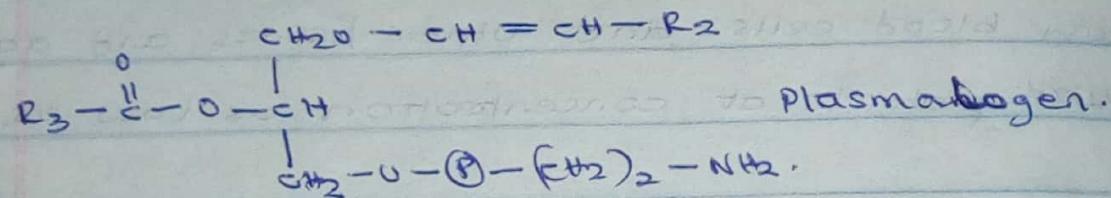


⑧

requirements for
the enzyme

[NADPH, O₂, Cytb5] desaturase

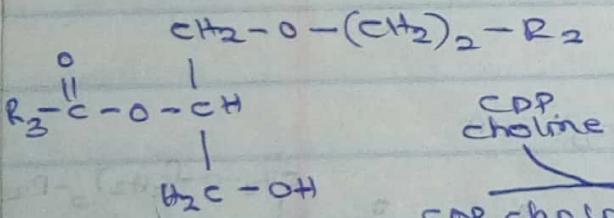
1-alkyl-2-acyl glycerol - 3-
phosphoethanolamine



1-alkenyl-2-acyl glycerol-3-
phosphoethanolamine.

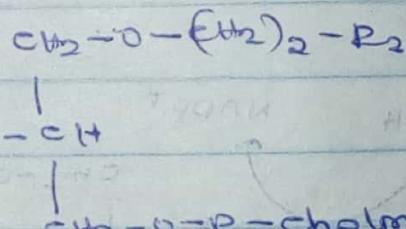
For Platelet activating factor.

At the 1-alkyl-2-acyl glycerol point from DHAP

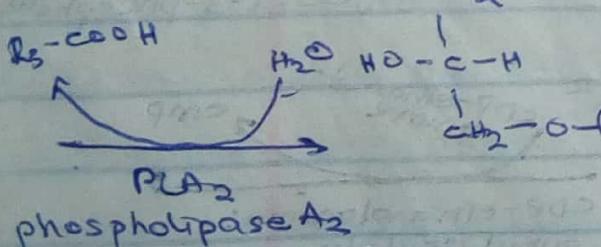


1-alkyl-2-acyl
glycerol

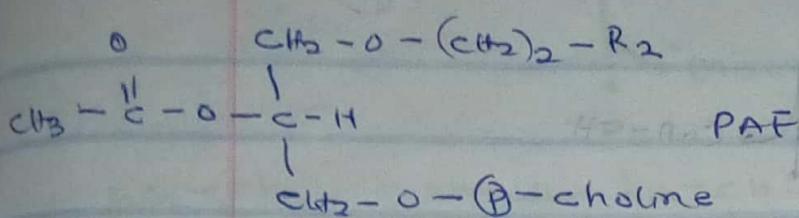
CDP choline
CDP choline:
acylglycerol
phosphocholine
transferase



1-alkyl-2-acylglycerol-
3-phosphocholine.



acetyl transferase
acetyl CoA

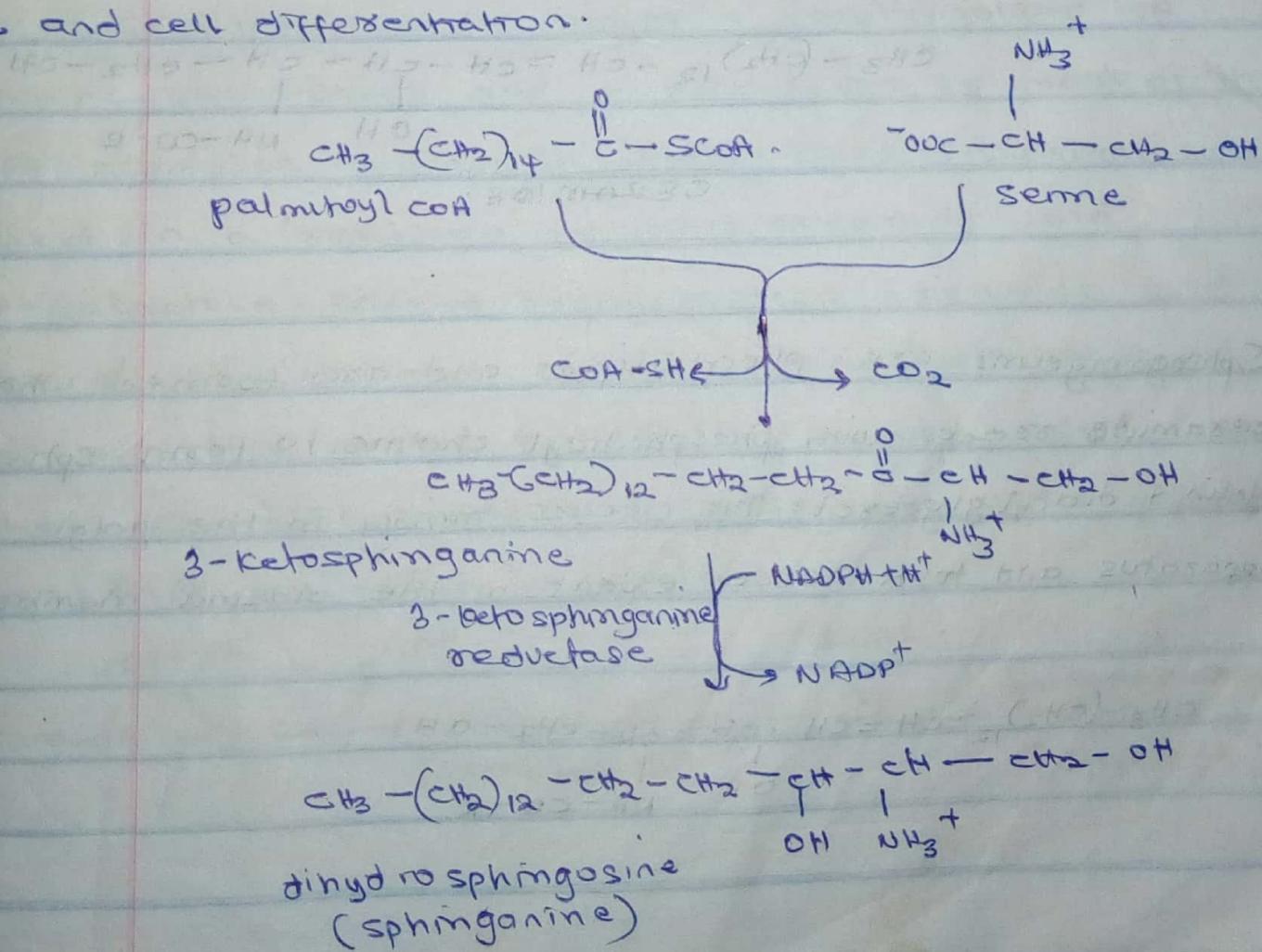


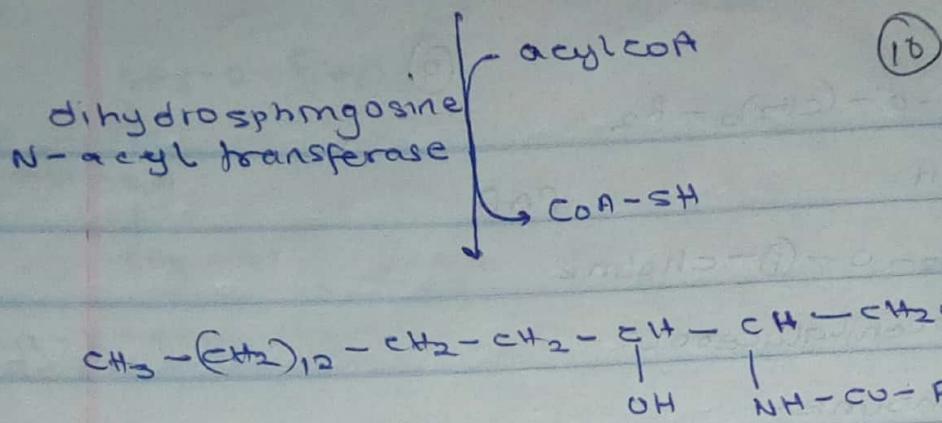
1-alkyl-2-acetylglycerol

3-phosphocholine

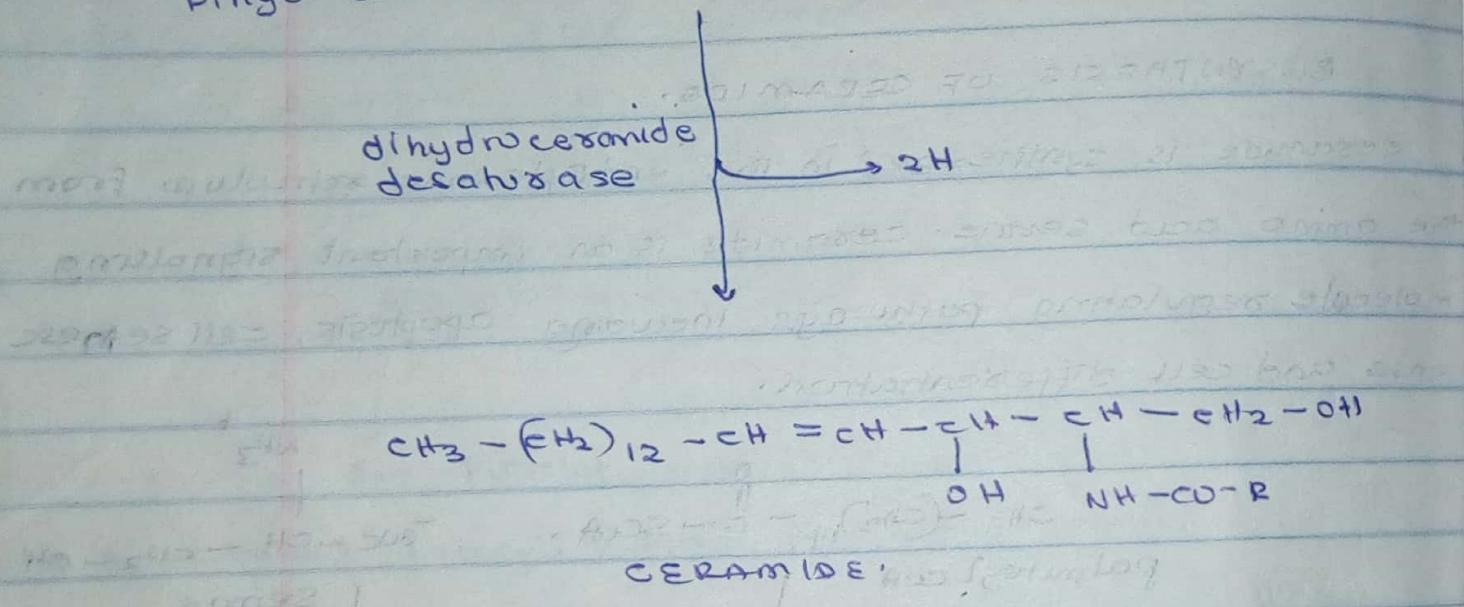
BIOSYNTHESIS OF CERAMIDE.

Ceramide is synthesized in the endoplasmic reticulum from the amino acid serine. Ceramide is an important signalling molecule regulating pathways including apoptosis, cell senescence and cell differentiation.

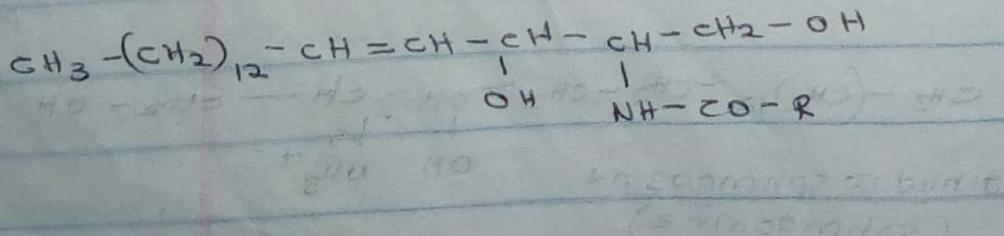


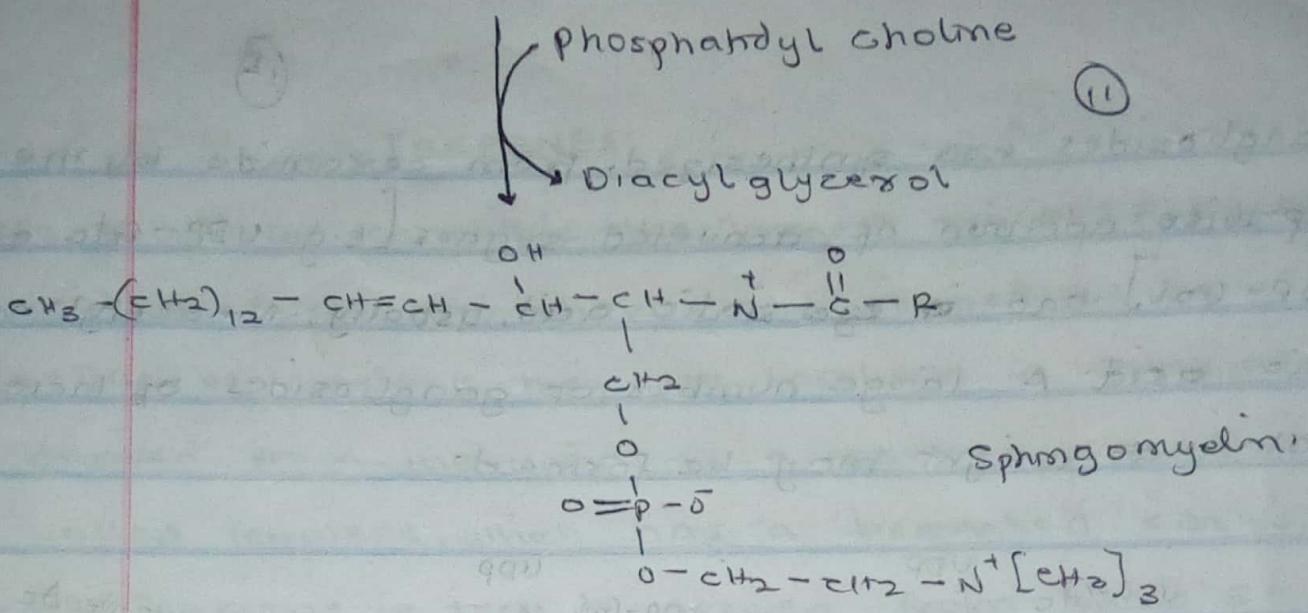


Dihydroceramide.

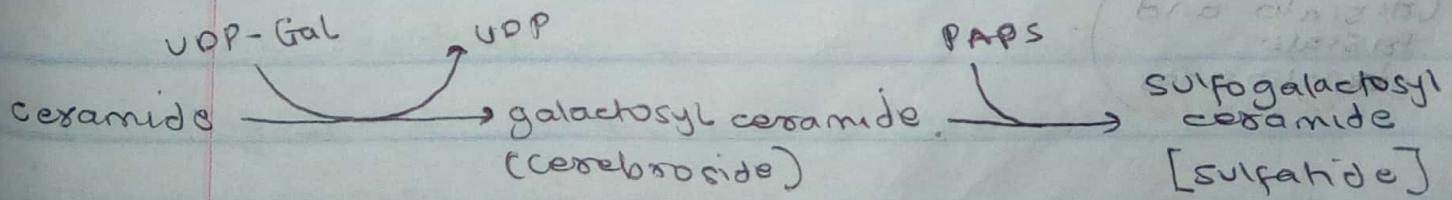


Sphingomyelins are phospholipids and are formed when ceramide reacts with phosphatidyl choline to form sphingomyelin + diacylglycerol. This occurs mainly in the golgi apparatus and to a lesser extent in the plasma membrane.



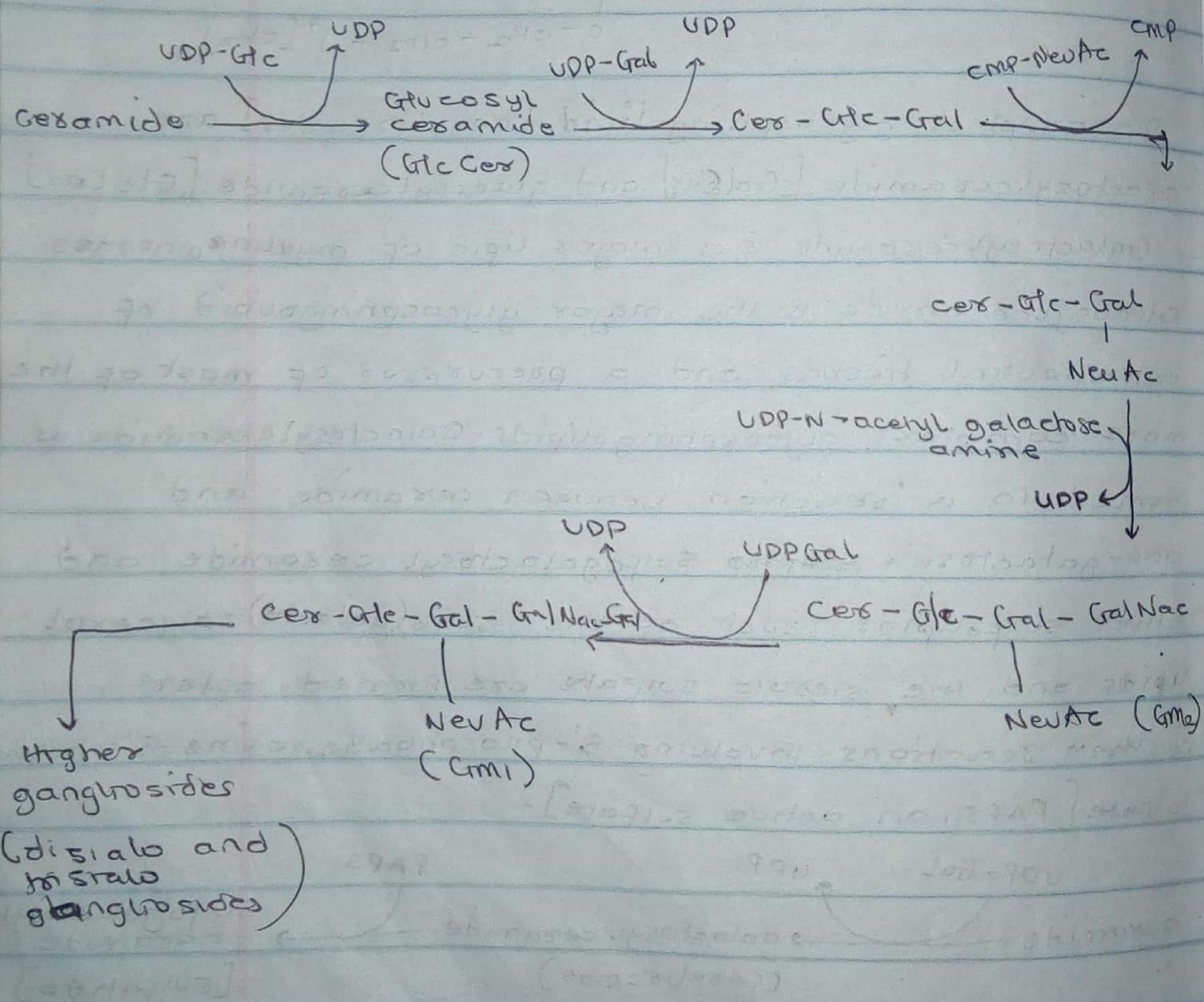


The simplest glycosphingolipid [cerebrosides] are galactosylceramide [GalCer] and glucosylceramide [GlcCer]. Galactosylceramide is a major lipid of myelin, whereas glucosylceramide is the major glycosphingolipid of extraneuronal tissues and a precursor of most of the more complex glycosphingolipids. Galactosylceramide is formed in a reaction between ceramide and UDP-galactose. ~~Sulfato~~ Sulfogalactosyl ceramide and other sulfolipids such as the sulfo(galacto)-glycerol lipids and the steroid sulfate are formed after further reactions involving 3'-phosphoadenosine-5'-phosphate [PAPS, an active sulfate].



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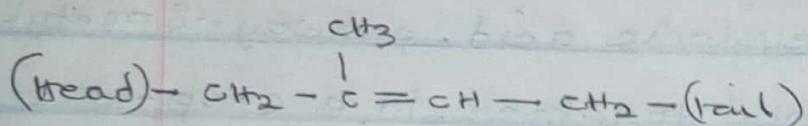
Gangliosides are synthesized from ceramide by the step-wise addition of activated sugars [e.g UDP-Glc and UDP-Gal] and ~~is~~ ^{is} sialic acid, usually, N-acetyl neurameric acid. A large number of gangliosides of increasing molecular weight may be formed.



TERPENES.

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These substances constitute the largest group of secondary plant products and show some properties of lipids - one of which is they are insoluble in water and are derived from the union of a common 5-carbon unit called Isoprene, which has a branched carbon skeleton. Isoprene in turn is derived from basic 5-carbon unit called Isopentane. Except for Isoprene itself, the terpenoids or terpenoids are dimers, trimers, tetramers or polymers in which Isoprene units are usually joined in a head to tail manner.



Classification of Terpenes

Terpenes are classified into many categories based on the number of carbon atoms & Isoprene residues present in their structure.

- ① Monoterpene :- They consist of 10 carbon atoms or 2 Isoprene residues
- ② Sesquiterpenes :- These contain 15 carbon atoms or 3 Isoprene residues
- ③ Diterpenes ; 20 carbon atoms or 4 Isoprene residues
- Sesterterpene : 25 carbon atoms or 5 Isoprene residue

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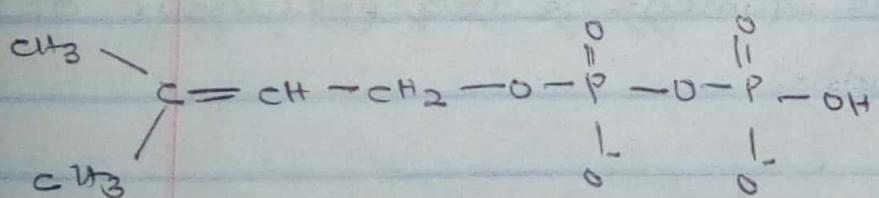
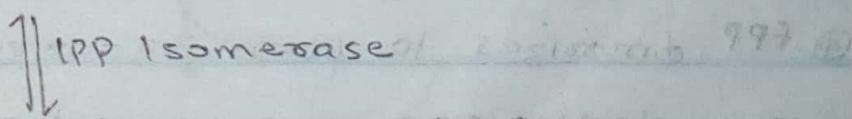
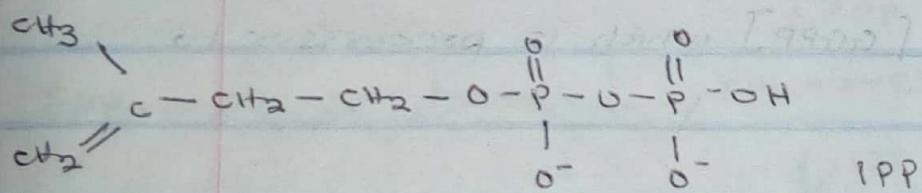
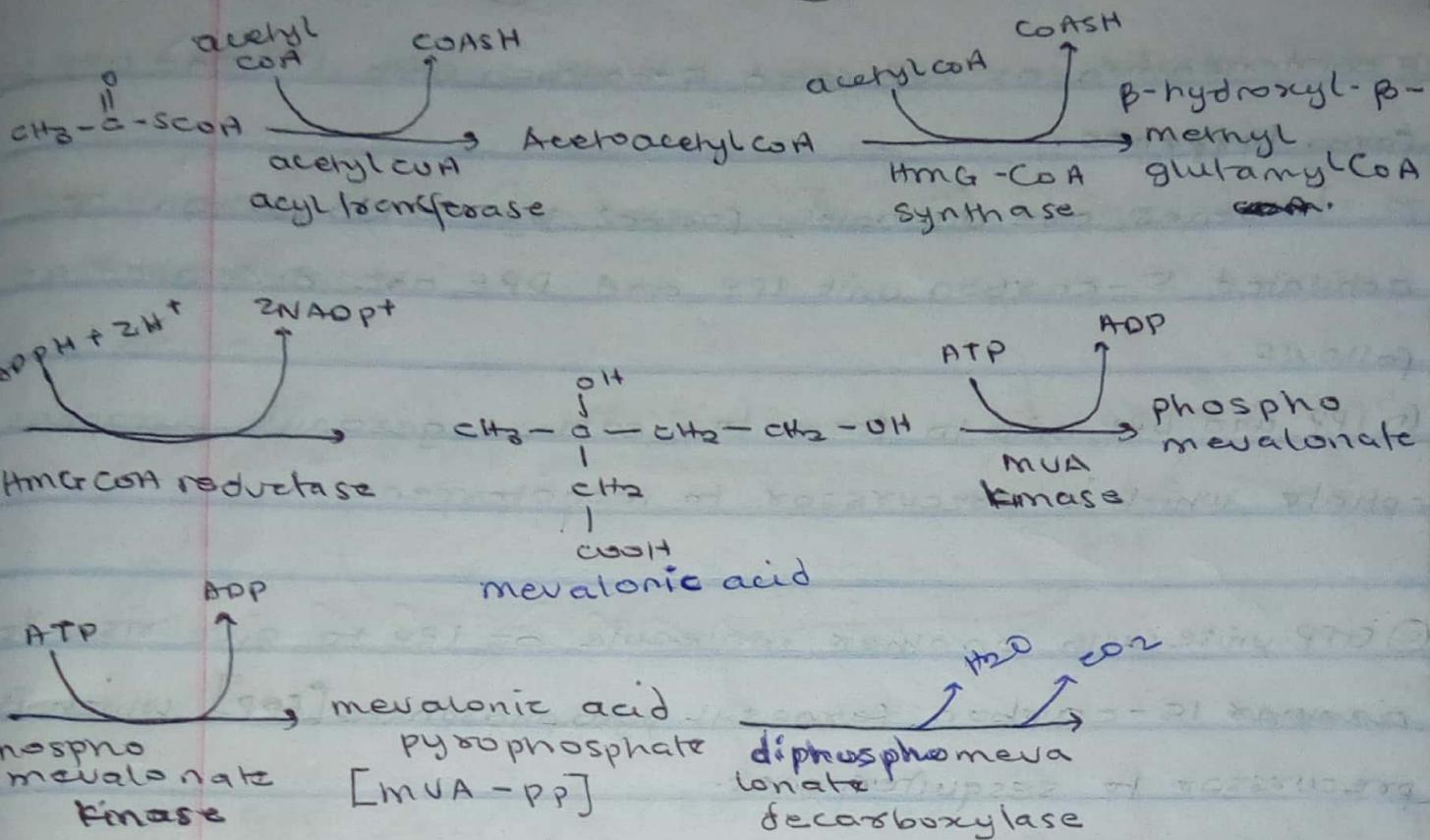
- ④ Triterpenes: These consist of 30 carbon atoms or 6-isoprene units.
- ⑤ Tetraterpenes: consist of 40 carbon atoms or 8-isoprene residues.
- ⑥ Polyterpene: consist of large number of isoprene residues [i.e. more than 8].

Biosynthesis of Terpenes:

This may be studied in 2 parts in the cytosol.

- ① Synthesis of activated 5 carbon units:
Isoprene units is almost entirely synthesized from acetyl CoA through mevalonic acid pathway. 3 acetyl CoA molecules are joined together in a step-wise manner to form a 6-carbon intermediate mevalonic acid. Mevalonic acid is then phosphorylated using 2 ATP molecules to form mevalonic acid pyrophosphate [MVA-PP]. Decarboxylation and dehydration of MVA-PP results in the formation of activated 5 carbon unit called isopentenyl pyrophosphate [IPP]. The latter can be isomerized to another activated 5-carbon unit called di-methylallyl pyrophosphate [DPP]. Both of these activated 5-carbon units are building blocks of terpenes in plants.

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(B) condensation of activated 5-carbon units IPP and DPP to form terpenes.

Terpenes are ultimately formed by condensation of activated 5-carbon unit IPP and DPP are described as follows

(i) IPP and DPP unite to form ten carbon geranyl pyrophosphate which is precursor to monoterpenes.

(ii) GPP unite with another molecule of IPP to give rise to ~~anomous~~ 15-carbon farnesyl pyrophosphate [FPP] which is precursor to sesquiterpenes.

(iii) FPP unite with IPP to form 20-carbon compound geranyl geranyl pyrophosphate [GGPP] which is precursor to diterpenes.

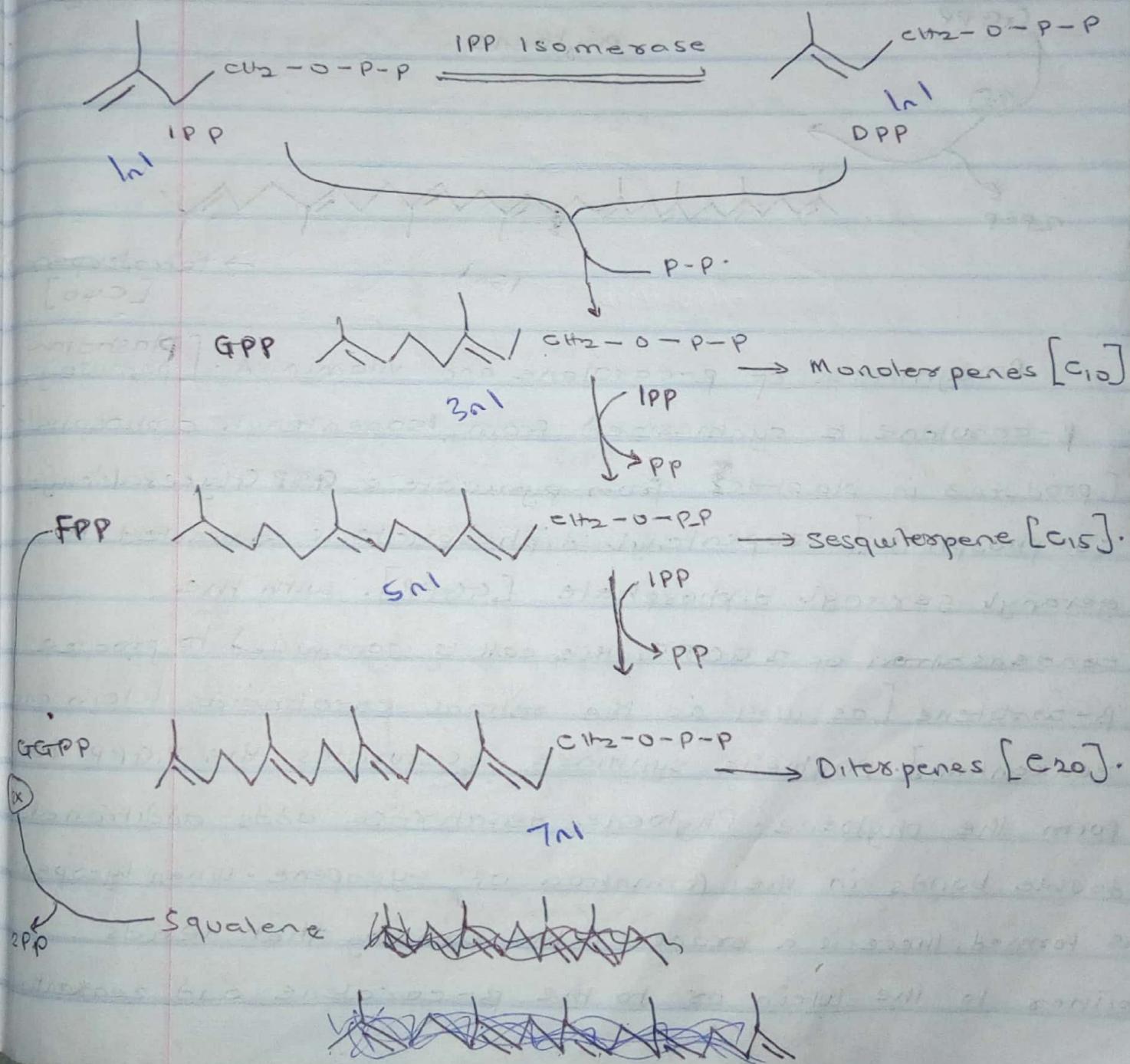
(iv) FPP dimerizes to form 30-carbon compound which after elimination of 2-pyrophosphate group, gives rise to squalene. The squalene is the precursor of triterpenes and steroids.

(v) GGPP can dimerize to form 40-carbon compound which after elimination of 2-pyrophosphate group gives rise

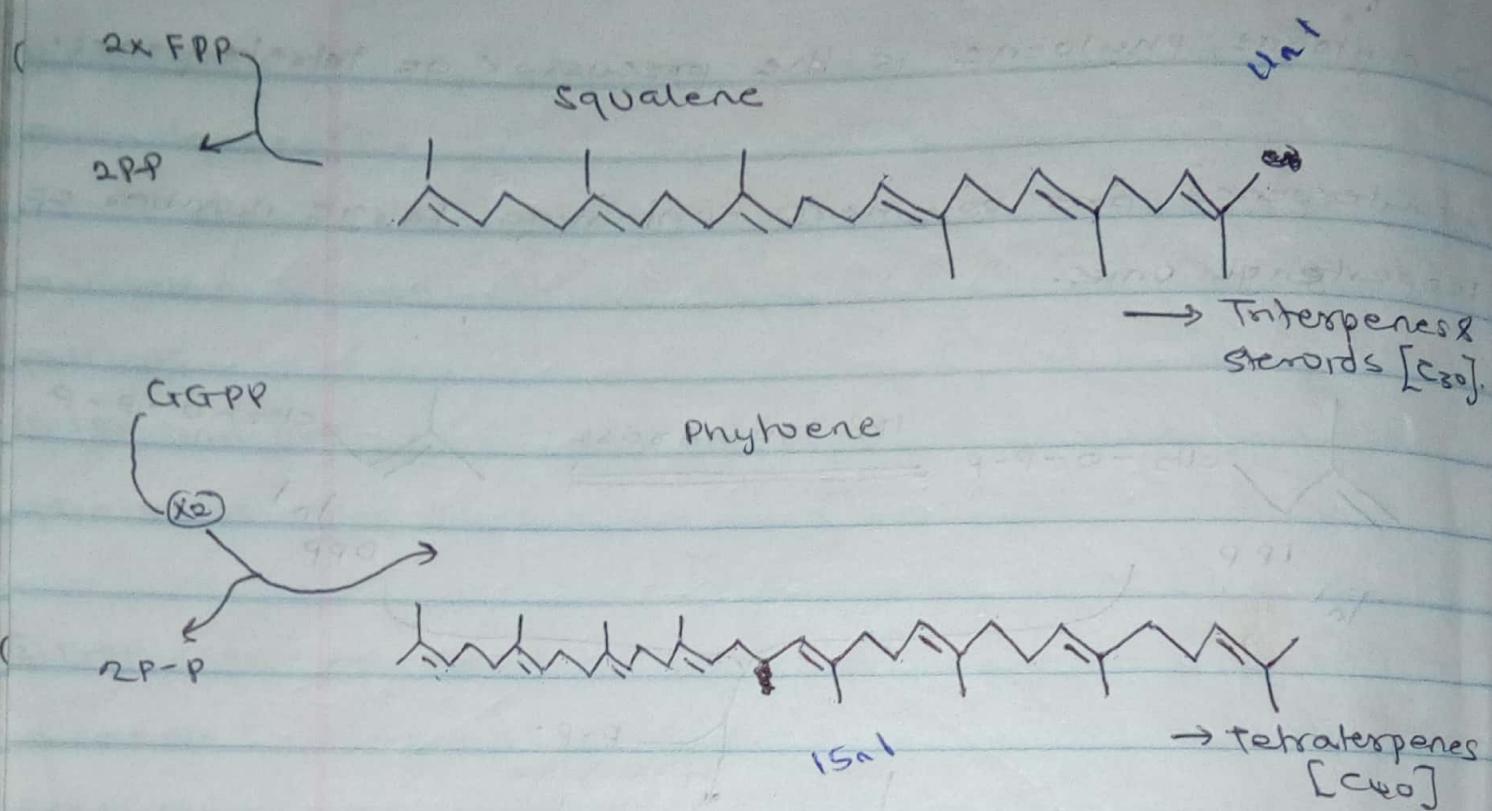
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to phytene, phytene is the precursor of tetraterpenes.

④ Polyterpenes are polymers containing large number of isopentenyl units.



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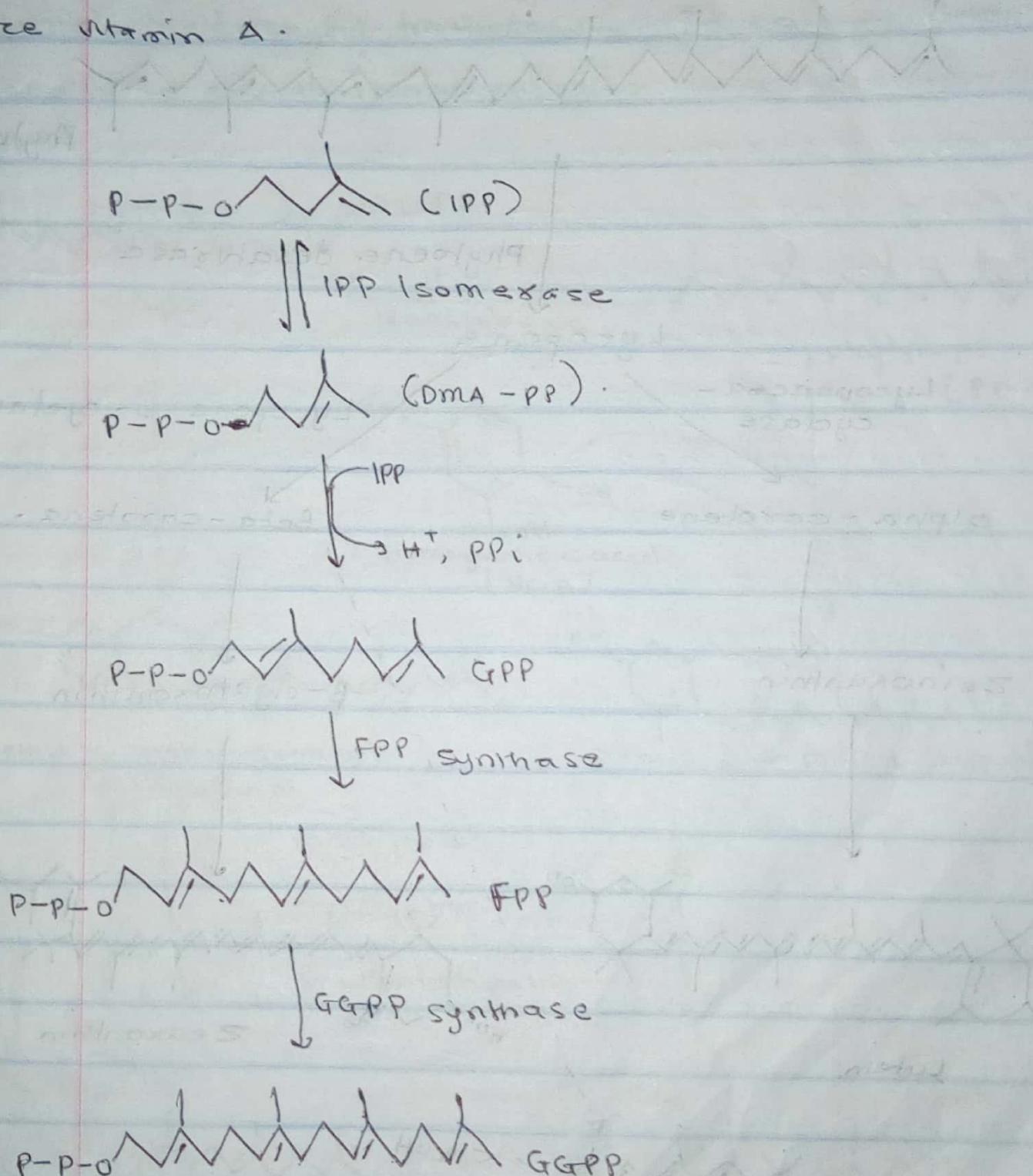


Biosynthesis of β -carotene and vitamin A. [Plastidial pathway]

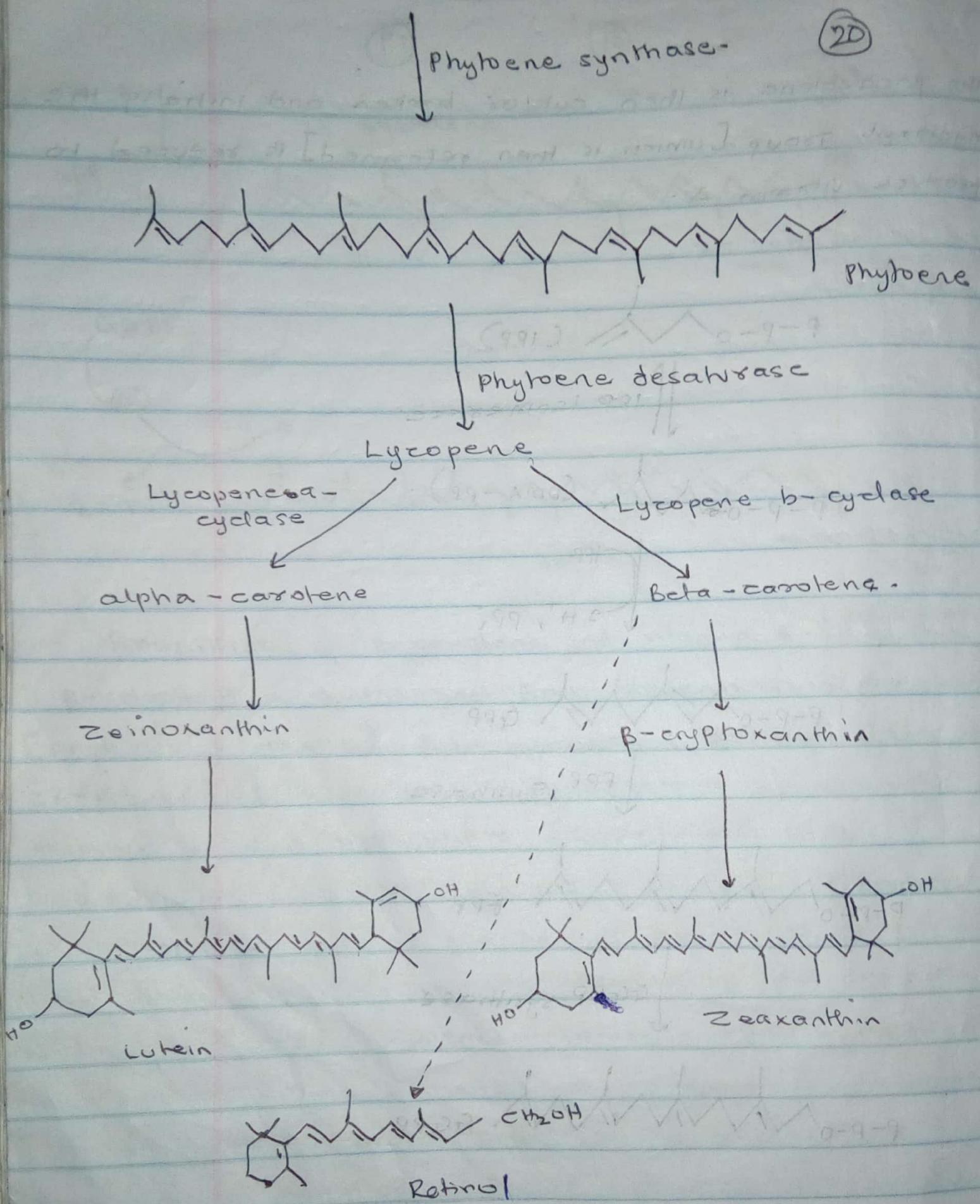
β -carotene is synthesized from isopentenyl diphosphate [produced in plastids] from pyruvate & G3P [glyceraldehyde-3-phosphate]. Isopentenyl diphosphate is converted to geranyl geranyl diphosphate [GGPP]. With the condensation of 2 GGPP, the cell is committed to produce β -carotene [as well as the retinal carotenoids lutein and zeaxanthin]. Phytoene synthase desaturates the GGPP to form the phytoene. Phytoene desaturase adds additional double bonds in the formation of lycopene. When lycopene is formed, there is a branch in the pathway that leads either to the lutein or to the β -carotene and zeaxanthin.

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the β -carotene is then cut or broken and initially the hydroxyl group [which is then reformed] is reduced to produce Vitamin A.



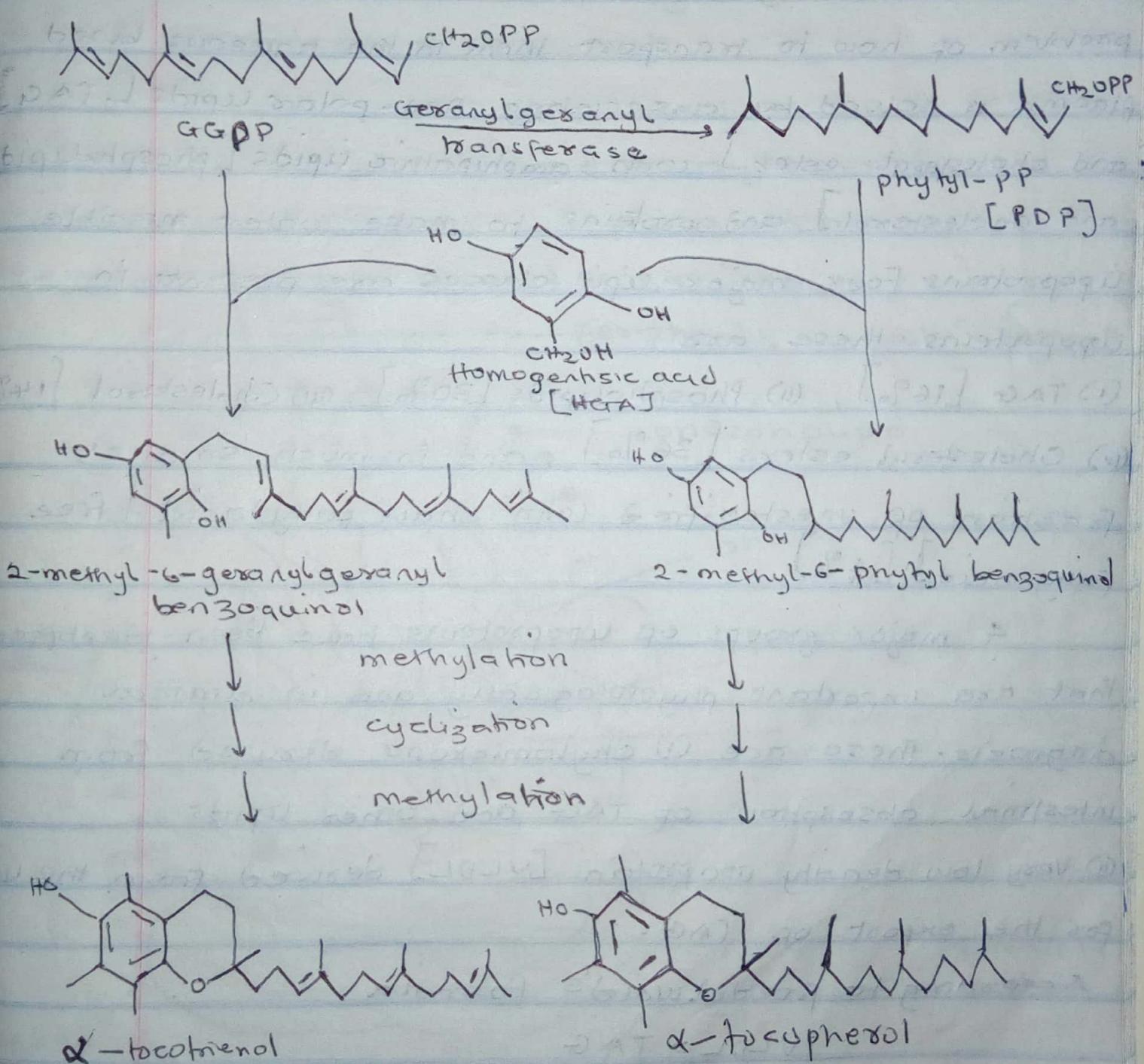
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Biosynthesis of Vitamin E

Homogentisic acid geranyl geranyl transferase [HGGT] and homogentisic acid phytol transferase [HPT] catalyze the first committed step in tocotrienol and tocopherol biosynthesis



IDL - Intermediary density lipoprotein

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* The protein part of a lipid is called apo protein

Lipoproteins.

They are lipid transporters, fats absorbed from the diet and lipid synthesized by the adipose tissue or liver must be transported between the various tissues and organs for utilization and storage. Since lipids are insoluble in water, the problem of how to transport them in the aqueous blood plasma is solved by associating non-polar lipids [TAGs and cholestryl ester] with amphipathic lipids [phospholipids and cholesterol] and proteins to make water miscible lipoproteins. Four major lipid classes are present in lipoproteins these are

- (i) TAG [16%], (ii) Phospholipids [30%], (iii) Cholesterol [14%]
- (iv) Cholestryl esters [36%], and in much smaller fraction of unesterified long chain fatty acid [free fatty acids] [4%]

4 major groups of lipoproteins have been identified that are important physiologically and in clinical diagnosis. These are (i) chylomicrons derived from intestinal absorption of TAG and other lipids

(ii) very low density lipoprotein [VLDL] derived from the liver for the export of TAG.

According to Fredrickson's formula

$$VLDL = \frac{TAG}{5}$$

chylomicron carries endogenous TAG from intestine to all other tissues.
VLDL carries TAG

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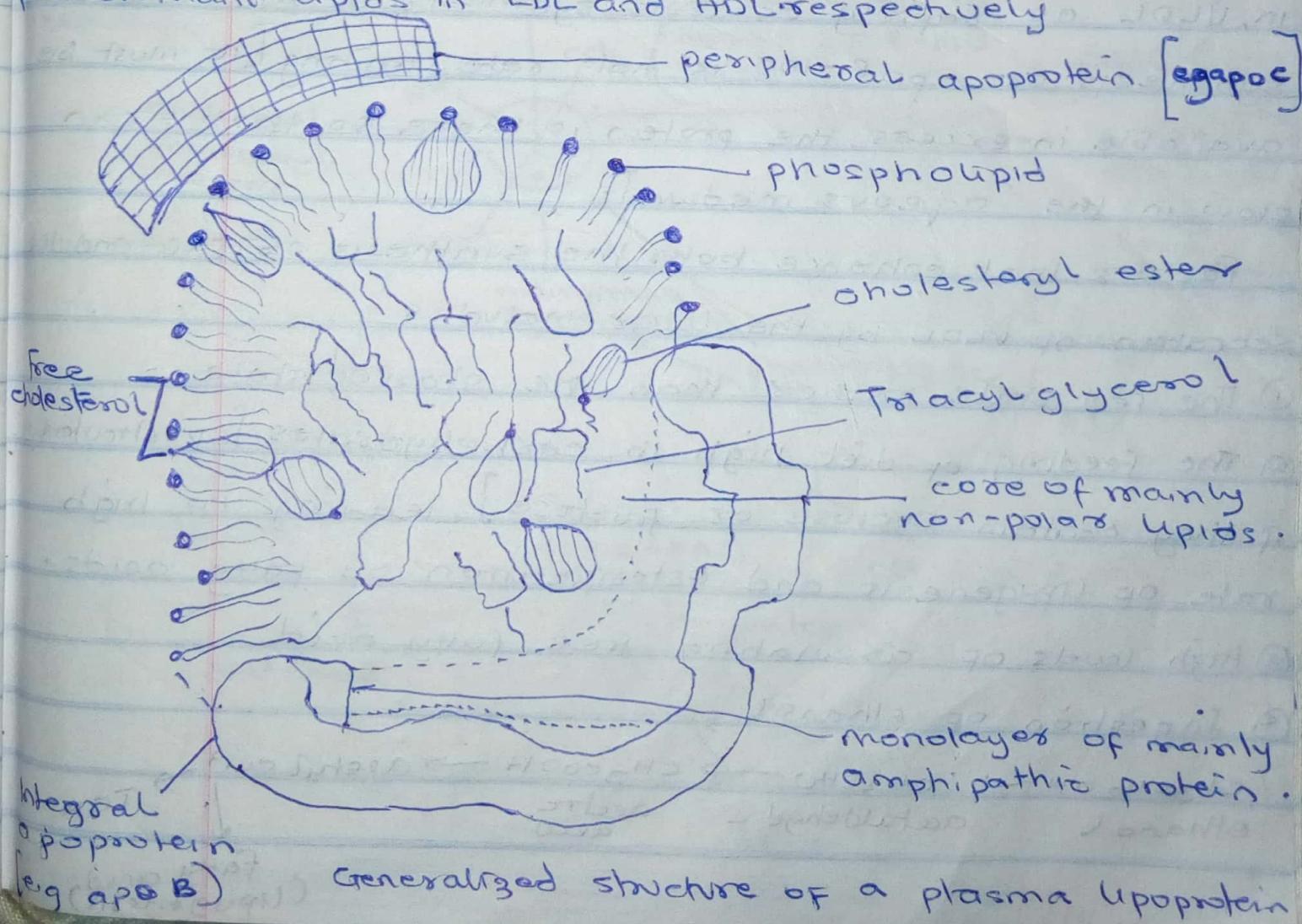
functions of LDL, VLDL, HDL, chylomicrons.

(iii) Low density lipoprotein [LDL] represents a final stage in the catabolism of VLDL

$$\text{LDL} = \frac{\text{Total cholesterol}}{5} - \frac{\text{TAG}}{5} - \text{HDL cholesterol}$$

(iv) HDL: Involved in VLDL and chylomicron metabolism and also in cholesterol transport

TAG is the predominant lipid in chylomicrons and VLDL whereas cholesterol and phospholipids are the predominant lipids in LDL and HDL respectively.



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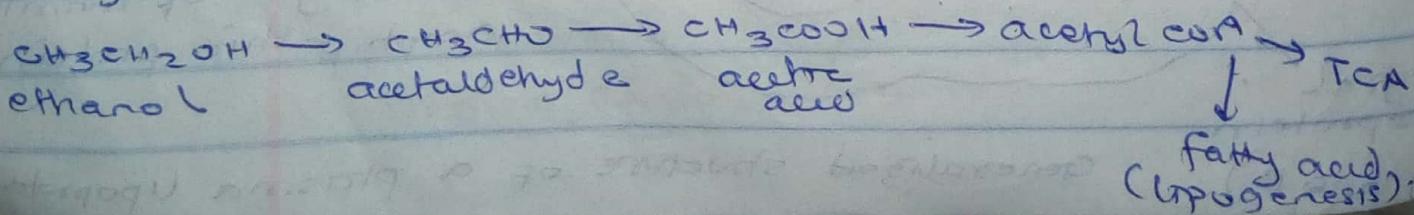
The diagram is the generalized structure of a plasma lipoprotein. A lipoprotein has similar structure of a plasma membrane. small amount of cholestryl ester and TAG are to be found in the surface layers and a little free cholesterol in the core.

Synthesis of VLDL in the liver.

As TAG does not normally accumulate in the liver, it must be inferred that it is transported from the liver in VLDL as rapidly as it is synthesized and that the synthesis of apoB-100 is not rate limiting [it must be available in excess; the protein is there so that it can flow in the aqueous medium].

Factors that enhance both the synthesis of TAG and the secretion of VLDL by the liver includes

- ① The fed state rather than the starved state
- ② The feeding of diet high in carbohydrates [particularly if they contain sucrose or fructose], leading to high rate of lipogenesis and esterification of fatty acids.
- ③ High levels of circulating free fatty acid
- ④ Ingestion of ethanol

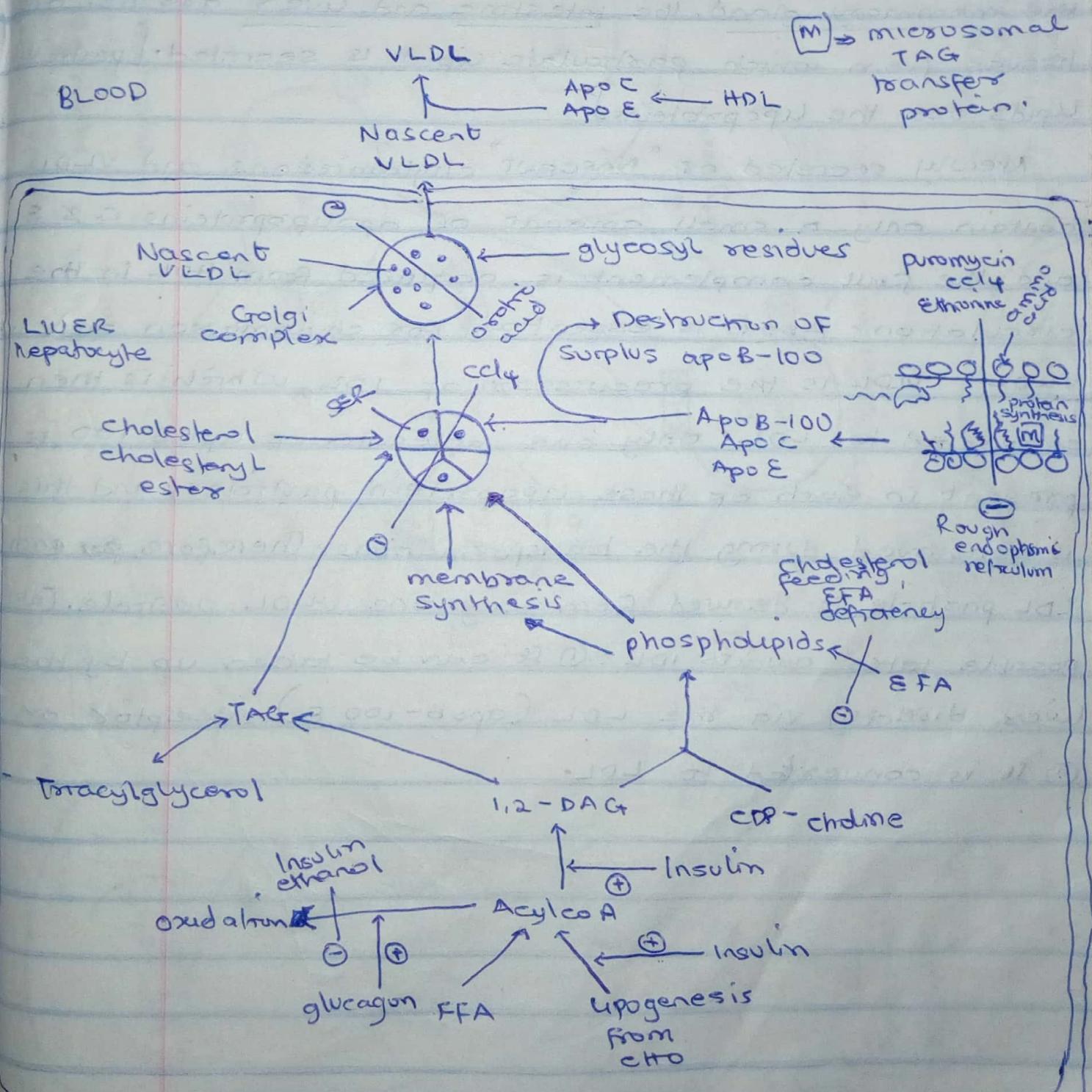


EFA → Essential fatty acid

FFA → free fatty acid

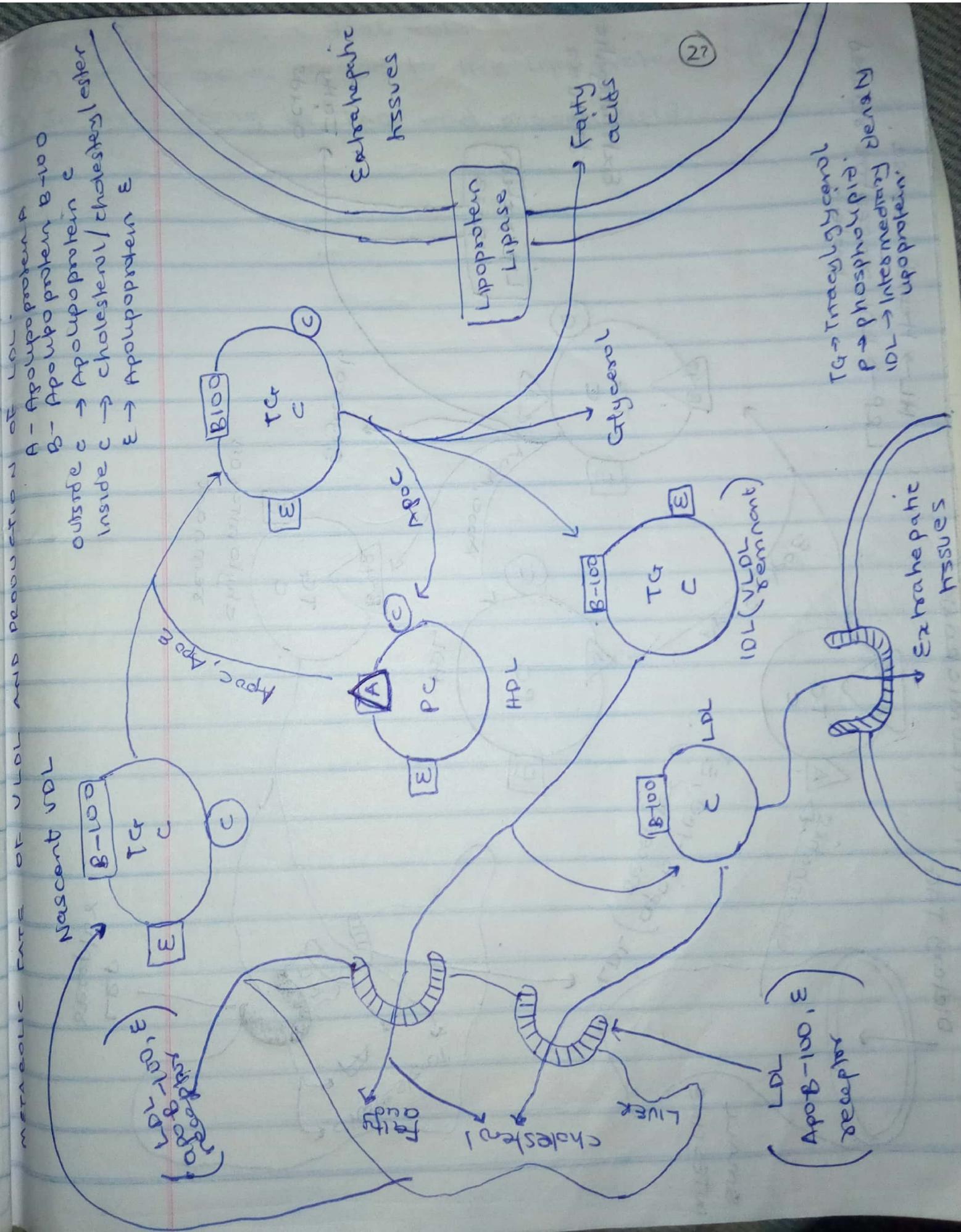
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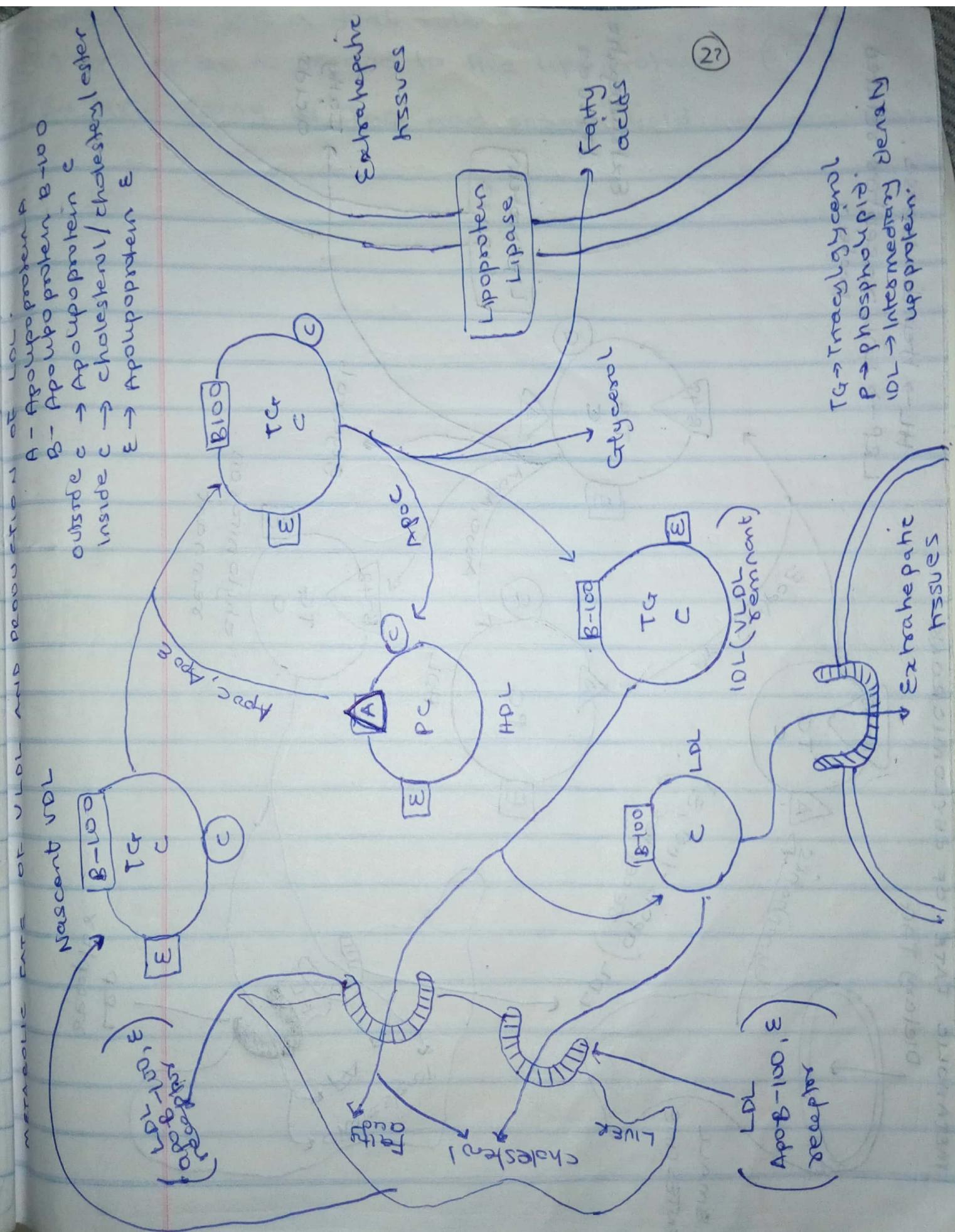
The presence of high concentrations of insulin and low concentrations of glucagon which enhance fatty acid synthesis and esterification and inhibits their oxidation.



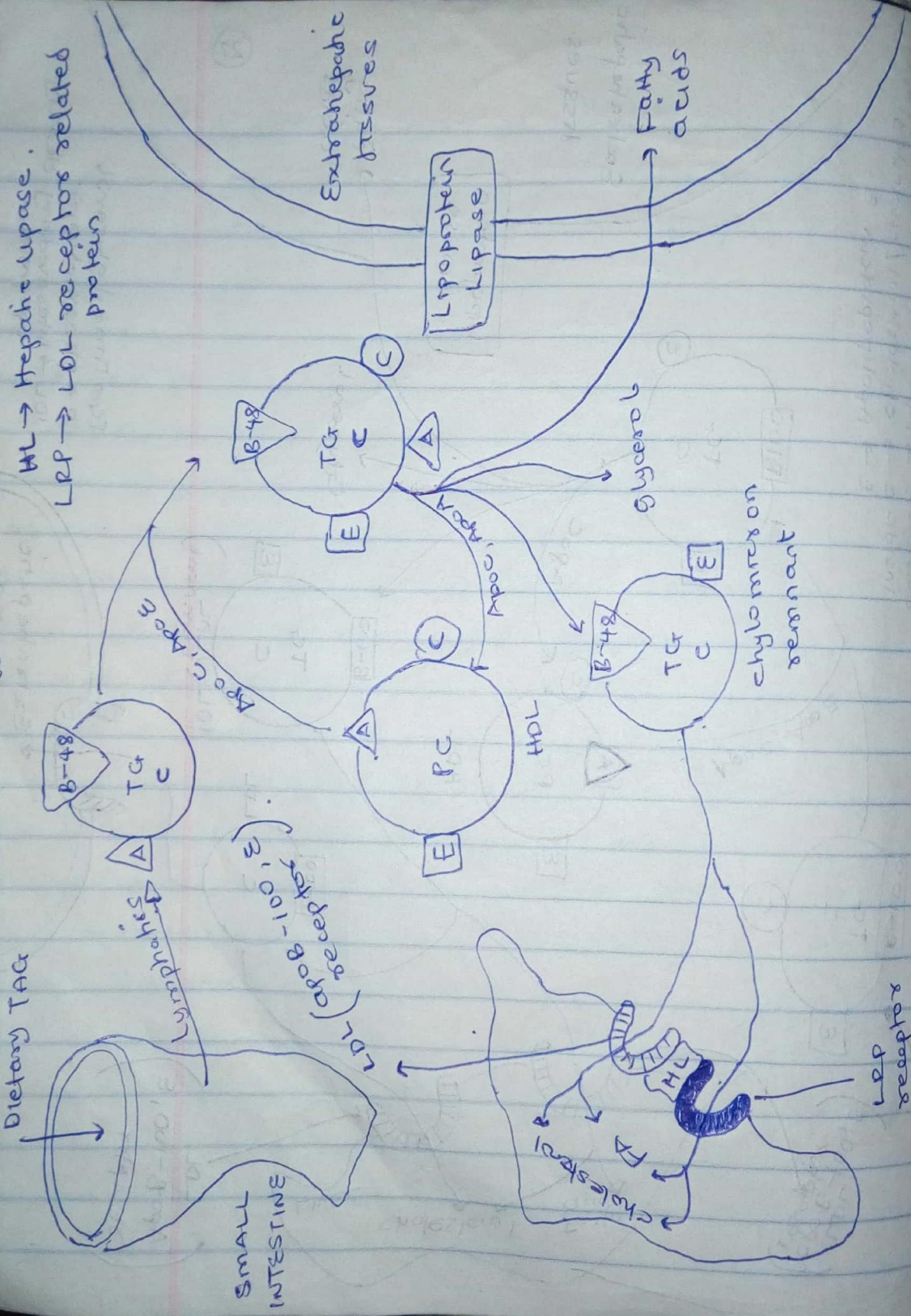
There are striking similarities in the mechanisms of formation of chylomicrons by intestinal cells and of VLDL by hepatic parenchymal cells, perhaps because apart from the mammary gland, the intestine and liver are the only tissues from which particulate lipid is secreted. [particulate lipids are the lipoproteins].

Newly secreted or "NASCENT" chylomicrons and VLDL contain only a small amount of apolipoproteins C & E, and the full complement is acquired from HDL in the circulation. ApoB is essential for chylomicron and VLDL. VLDL is the precursor of IDL, which is then converted to LDL. Only one molecule of apoB 100 is present in each of these lipoprotein particles, and this is conserved during the transformations. Therefore, ~~for~~ each LDL particle is derived from only one VLDL particle. Two possible fates awaits IDL: ① It can be taken up by the liver directly via the LDL (apoB-100, E) receptor or ② It is converted to LDL.





METABOLIC FATE OF CHOLEMICRONES



Hepatic lipase has a dual role.

- ① In-acting as a ligand to the lipoprotein
- ② In hydrolysing its TAG and phospholipid

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