Fat Metabolism (Biosynthesis of Triglycerides)

Fats are a group of simple lipids which are esters of long-chained fatty acids and a trihydric alcohol viz. glycerol. They are derived from plants—primarily seeds, fruits, and nuts—composed of triglycerides that serve as vital energy storage. They are typically liquid (oils) at room temperature, though some are solid. Primarily unsaturated, these healthy fats are used for cooking, industrial applications (cosmetics, lubricants), and in food products.

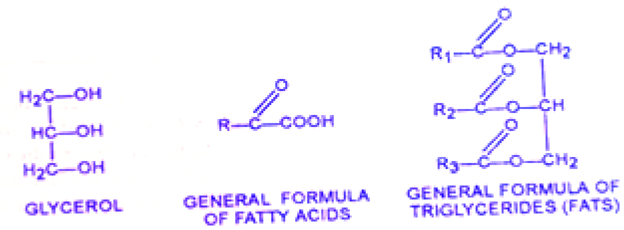


Fig: Structure of glycerol, fatty acid and natural fat

They are also known as Vegetable oils, plant lipids, vegetable fats, and seed oils. Fats are mainly comprised of C16 and C18 fatty acids, including palmitic, stearic, oleic, and linoleic acids. They are mostly liquid at room temperature (oils), but can be solid (e.g., coconut oil, cocoa butter). Plant oils are often rich in unsaturated fats (e.g., linoleic, oleic), making them liquid at room temperature. About 90% of plant fatty acids are comprised of just six types: palmitic, palmitoleic, stearic, oleic, linoleic, and linolenic acid.

Fats in plants, primarily stored as liquid triacylglycerols (oils), are concentrated in seeds and fruits for energy, or as structural lipids in membranes. Seeds can contain up to 70% lipid, while leaves contain 1–7%. Major fatty acids include palmitic, stearic, oleic, linoleic, and linolenic acids.

Key Distribution Locations and Functions:

- **Seeds (Storage):** Primary storage site for triglycerides, acting as energy for germination. Examples include groundnut, soybean, sunflower, and nuts, which often contain up to 65% fat.
- **Fruit Mesocarp (Storage):** Some plants store high amounts of oil in the fleshy fruit layer, such as avocado and oil palm.
- **Leaves (Structural):** Leaves contain lipids, largely within chloroplast membranes (galactolipids) to support photosynthesis, typically accounting for up to 7% of their dry weight.
- **Surface (Protection):** Waxes and cutin cover the plant surface to provide an impervious barrier against water loss and environmental hazards.

- **Organelle Synthesis:** Lipids are synthesized in chloroplasts (fatty acids), the endoplasmic reticulum (phospholipids), and specialized oil bodies.

Fat metabolism in plants

Fat metabolism in plants involves synthesizing fatty acids within plastids (chloroplasts) from acetyl-CoA, differing from animals where this occurs in the cytosol. Key processes include developing storage lipids like triacylglycerols (TAGs) in the endoplasmic reticulum and breaking them down during germination via *β* oxidation for energy. These lipids also form plant membranes, waxy cuticles, and signaling molecules.

Fat metabolism involves two processes –

synthesis and **degradation**

Fat synthesis involves the following three steps –

- Synthesis of fatty acids
- Synthesis of glycerol
- Condensation of fatty acids and glycerol into fats.

Synthesis of fatty acids

Plants synthesise a huge variety of fatty acids although only a few are major and common constituents. Broadly speaking, long-chain fatty acids are synthesised ***de novo*** from small precursors ultimately derived from photosynthate. Two enzyme systems are utilised, **acetyl-CoA carboxylase** and **fatty acid synthase** (**Fig. 1**). The end products of this synthesis are usually the saturated fatty acids **palmitate** and **stearate** with the latter predominating (in most plants by 2-3 times that of palmitate). Once the long-chain acids have been produced they can be subject to **elongation**, **desaturation** and further **modifications** (Fig. 1). Unlike acetyl-CoA carboxylase and fatty acid synthase, which are soluble enzymes, the **elongases** are membrane-bound and sited in the endoplasmic reticulum. Elongases are coded by *FAE* genes while the **desaturases** are coded by *FAD* genes.

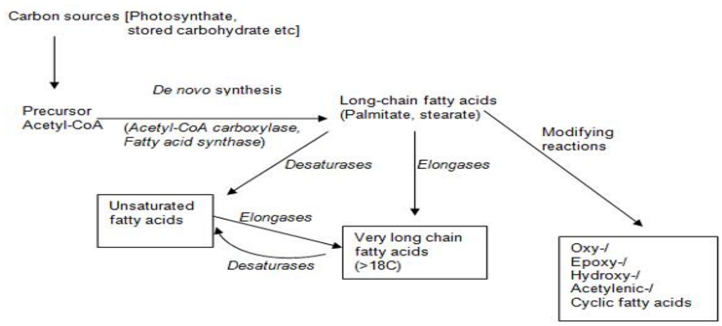


Figure 1. **Fatty acid biosynthesis in plants**

Fatty acid desaturases are usually membrane-bound and utilise complex lipid substrates such as phosphatidylcholine or monogalactosyl diacylglycerol. An exception is the stearoyl-acyl carrier protein (ACP) **Δ9-desaturase** that is present in the chloroplast stroma and converts stearate to oleate. When desaturases produce polyenoic fatty acids, the latter usually have a methylene-interrupted structure, such as linoleic (*cis, cis* Δ9,12-octadecadienoic) acid or α-linolenic (all *cis* Δ9,12,15-octadecatrienoic) acid.

(*a*) **Synthesis of fatty acids.** Nearly all the long chain fatty acids contain an even number of carbon atoms. For example, lauric acid contains 12 C atoms, myristic acid contains 14 C atoms, palmitic acid contains 16 C atoms and stearic acid contains 18 C atoms and so on (Fig. 20.2). This suggests that these are built up by the repeated addition of C units. Further studies demonstrated that acetyl Co~A, produced from carbohydrates during respiration, provides the necessary carbon atoms for fatty acid synthesis. Later, it was shown that CO_2 was involved in the formation of a 3C compound, malonyl Co~A from acetyl Co~A (Klein, 1957).

The reaction is as follows :

$$\underset{\text{Acetyl Co-A}}{CH_3CO-S-CoA} + HCO_3^- + ATP \xrightarrow[\text{Biotin}]{Mn^{++}} \underset{\text{Malonyl Co-A}}{COOH-CH_2-CO-SCoA} + ADP + Pi$$

This reaction requires the involvement of vitamin biotin, manganese ions, ATP and an enzyme *carboxylase*.

Further steps in the synthesis of fatty acids are as follows :

(*i*) The acetyl Co~A gets attached to an acyl carrier protein (ACP).

$$\underset{\text{Acetyl CoA ACP}}{CH_3-CO-S-CoA} + \begin{matrix} HS \\ HS \end{matrix}\!\!>\!\text{Enzyme} \rightarrow \underset{\text{Acetyl-S-ACP}}{CH_3-CO-S}\!\!\begin{matrix} HS \\ \end{matrix}\!\!>\!\text{Enzyme} + HS-CoA$$

(*ii*) The malonyl Co~A reacts with reduced ACP and forms malonyl derivative of ACP (malonyl — S — ACP) —

$$\text{Malonyle}-S-CoA + ACP-SH \longrightarrow \text{Malonyl}-S-ACP + CoA-SH$$

(*iii*) The malonyl S — ACP then condenses with the primer acetyl — S — ACP. The CO_2 is released and the condensation reaction leads to a chain of 4C atom.

$$\text{Malonyl}-S-ACP + \text{Acetyl}-S-ACP \longrightarrow \text{Acetoacetyl}-S-ACP + CO_2 + ACP-SH$$

(*iv*) This 4C unit (Aceto acetyl — S — ACP) is then converted to a saturated fatty acid by three successive reactions as follows :

Reduction

$$\text{Acetoacetyl}-S-ACP + NADPH + H^+ \longrightarrow \alpha\text{-Hydroxybutryl}-S-ACP + NADP^+$$

Dehydration

$$\alpha\text{-Hydroxybutyryl}-S-ACP \longrightarrow \text{Crotonyl}-S-ACP + H_2O$$

Reduction

$$\text{Crotonyl}-S-ACP + NADPH + H^+ \longrightarrow \text{Butyryl}-S-ACP + NADP^+$$

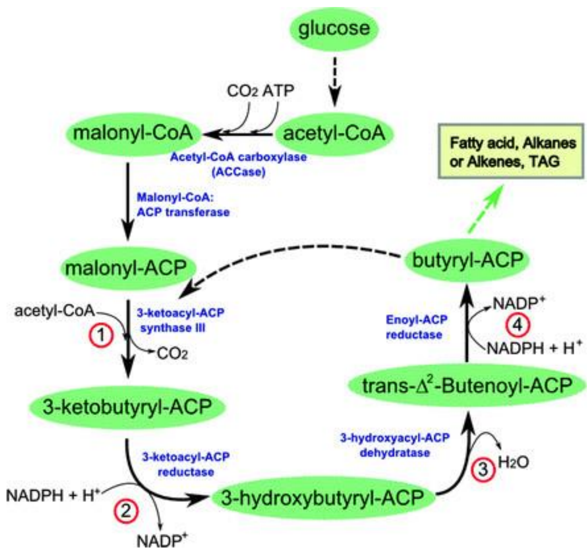
The reaction sequence is repeated to give 6C unit, 8C unit, 10C unit and so on. Finally the most important fatty acids with 16C and 18C units are produced. When the required chain length has been attained, the acyl group is transferred to HS-group of coenzyme-A. Finally the Co~A derivative of fatty acids are formed, which are usedup in fat synthesis.

The above mentioned reactions occur in the cytosol at the endoplasmic reticulum. These reactions are catalysed by a multienzyme complex, called **fatty acid synthetase complex.** The enzyme complex probably contains six enzymes, each catalysing one step.

The overall generalized equation for the synthesis of **palmitate** is summarized as follows :

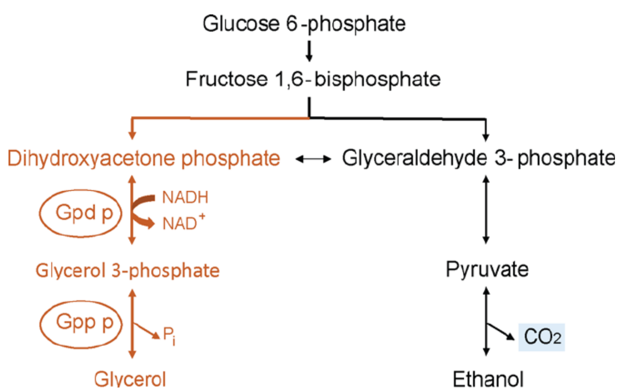
$$8\ \text{Acetyl CoA} + 7\ ATP + 14\ NADPH \longrightarrow \text{Palmitate} + 14\ NADP^+ + 8\ CoA + 6\ H_2O + 7\ ADP + 7\ Pi$$

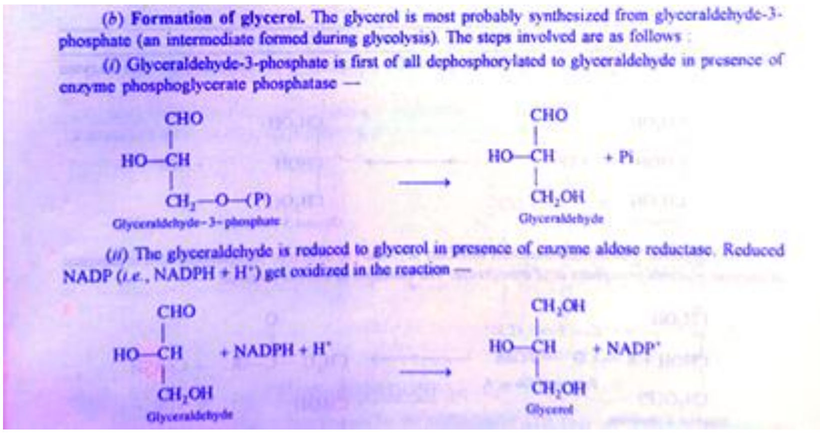
Some plants can produce unusual fatty acids in their seed oils, many of which have useful industrial applications. These include hydroxyl fatty acids, cyclopropane fatty acids, epoxy fatty acids and conjugated unsaturated fatty acids. It is noteworthy that these unusual fatty acids accumulate preferentially in triacylglycerols and are essentially excluded from membrane acyl lipids – presumably because they would impair function.



Fatty acid biosynthesis. Acetyl-CoA is converted into malonyl-CoA by the enzyme acetyl-CoA carboxylase. The malonyl group is transferred to an acyl carrier protein to yield malonyl-ACP. Malonyl-ACP undergoes a condensation reaction with acetyl-CoA to yield acyl-ACP with the concomitant release of CO2 (marked as reaction 1). Acyl-ACP then undergoes a series of reactions involving reduction, dehydration, and another reduction reaction to yield longer-chain ACPs (marked as reactions 2, 3, and 4, respectively). The three reactions are catalyzed by 3-ketoacyl-ACP reductase, 3-hydroxyacyl-ACP dehydratase, and enoyl-ACP reductase, respectively. Sequential repetition of the last four steps (marked as reactions 1 through 4) starting from the condensation (using three different ketoacyl-ACP synthases, 3-ketoacyl-ACP-synthase III (KASIII), KASII (not shown), and KASI (not shown)) with malonyl-ACP yields the 16 or 18-carbon product. The fatty acid is finally released by thioesterases.

Synthesis of Glycerol





(b) **Formation of glycerol.** The glycerol is most probably synthesized from glyceraldehyde-3-phosphate (an intermediate formed during glycolysis). The steps involved are as follows :

(i) Glyceraldehyde-3-phosphate is first of all dephosphorylated to glyceraldehyde in presence of enzyme phosphoglycerate phosphatase —

Glyceraldehyde-3-phosphate → Glyceraldehyde + Pi

(ii) The glyceraldehyde is reduced to glycerol in presence of enzyme aldose reductase. Reduced NADP (i.e., NADPH + H^+) get oxidized in the reaction —

Glyceraldehyde + NADPH + H^+ → Glycerol + $NADP^+$

Biosynthesis of glycerol

- Glucose is oxidized via glycolysis to **dihydroxy acetone phosphate**

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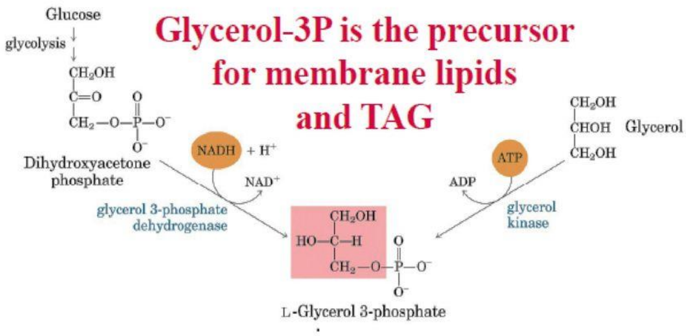
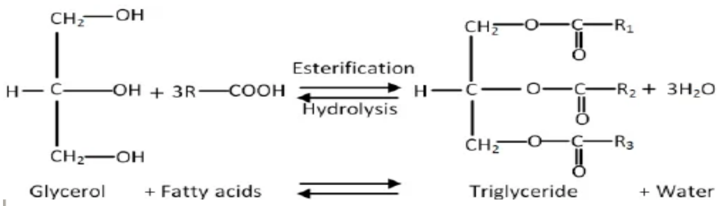
reduced to **glycerol-3 phosphate** by the enzyme glycerol-3 phosphate dehydrogenase.

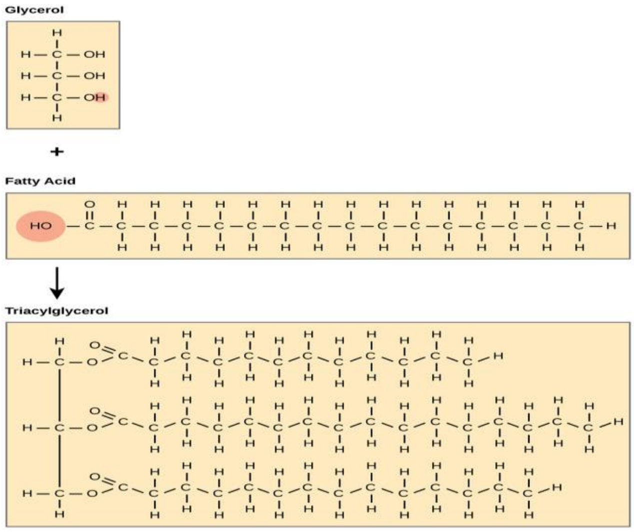


Combining Glycerol and Fatty acid to form Triglycerides

A fat molecule consists of two main components: glycerol and fatty acids. Glycerol is an alcohol with three carbons, five hydrogens, and three hydroxyl (OH) groups. Fatty acids have a long chain of hydrocarbons with a carboxyl group attached and may have 4-36 carbons; however, most of them have 12-18.

In a fat molecule, the fatty acids are attached to each of the three carbons of the glycerol molecule with an ester bond through the oxygen atom. During the ester bond formation, three molecules are released. Since fats consist of three fatty acids and a glycerol, they are also called triacylglycerols or triglycerides.





The overall process of triglyceride (triacylglycerol) biosynthesis consists of **four** biochemical pathways:

- fatty acyl-CoA biosynthesis
- conversion of fatty acyl-CoA to phosphatidic acid
- conversion of phosphatidic acid to diacylglycerol
- conversion of diacylglycerol to triacylglycerol

(*i*) First of all, the fatty acids are converted to their acyl — CoA derivatives in presence of ATP and enzyme *fatty acyl CoA synthetase*. This conversion occurs in the microsomes —

$$\underset{\text{Fatty acid}}{R—COOH} + CoASH + ATP \longrightarrow \underset{\text{Fatty acyl–CoA}}{R—\overset{O}{\overset{\|}{C}}—S—CoA} + AMP + PPi$$

(*ii*) The glycerol is phosphorylated to glycerol-3-phosphate in presence of ATP and enzyme *glycerokinase*:

a) $$\underset{\text{Glycerol}}{CH_2OH—CHOH—CH_2OH} + ATP \longrightarrow \underset{\text{Glycerol-3-phosphate}}{CH_2OH—CHOH—CH_2O(P)} + ADP$$

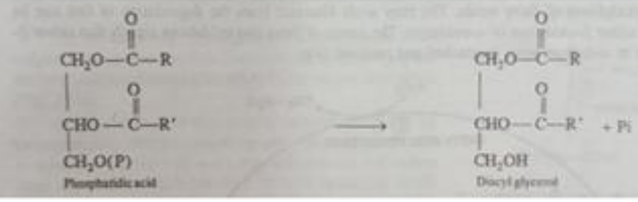
Glycerol-3-phosphate combines with fatty acyl-CoA to form lyso-phosphatidic acid in presence of enzyme *glycerol phosphate acyl transferase*. The molecule of CoA is released —

b) $$\underset{\text{Glycerol-3-phosphate}}{CH_2OH—CHOH—CH_2O(P)} + \underset{\text{Fatty acyl Co}\sim\text{A}}{R—CO—SCoA} \longrightarrow \underset{\text{Lysophosphatidic acid}}{CH_2O—\overset{O}{\overset{\|}{C}}—R \;|\; CHOH \;|\; CH_2O(P)} + CoASH$$

c) Lysophosphatidic acid combines with second molecule of fatty acyl-CoA to form phosphatidic acid in presence of enzyme *I, acyl glycerol phosphate acyl transferase* —

$$\underset{\text{Lysophosphatidic acid}}{CH_2O—\overset{O}{\overset{\|}{C}}—R \;|\; CHOH \;|\; CH_2O(P)} + \underset{\text{Fatty acyl Co}\sim\text{A}}{R'—CO—SCoA} \longrightarrow \underset{\text{Phosphatidic acid}}{CH_2O—\overset{O}{\overset{\|}{C}}—R \;|\; CHO—\overset{O}{\overset{\|}{C}}—R' \;|\; CH_2O(P)} + CoA\,SH$$

(iii) The phosphate residue is cleared from phosphatidic acid to form a diacyl glycerol in presence of enzyme *phosphatidate phosphohydrolase* —



(iv) Finally the third hydroxyl group of diglyceride is esterified with fatty acyl CoA and produces a triacyl glycerol (neutral fat) in presence of enzyme *acyl transferase* —