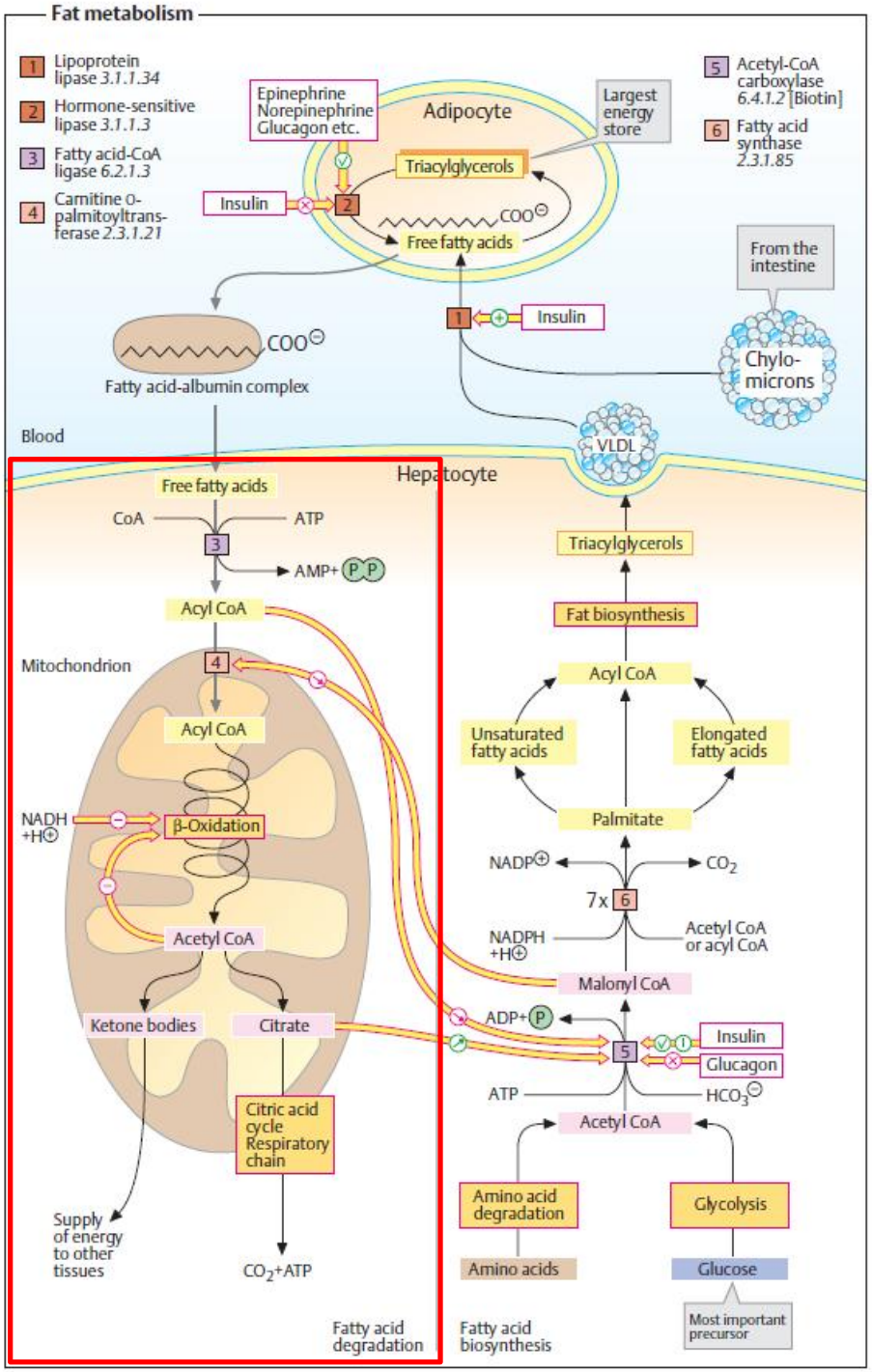
FATTY ACID OXIDATION



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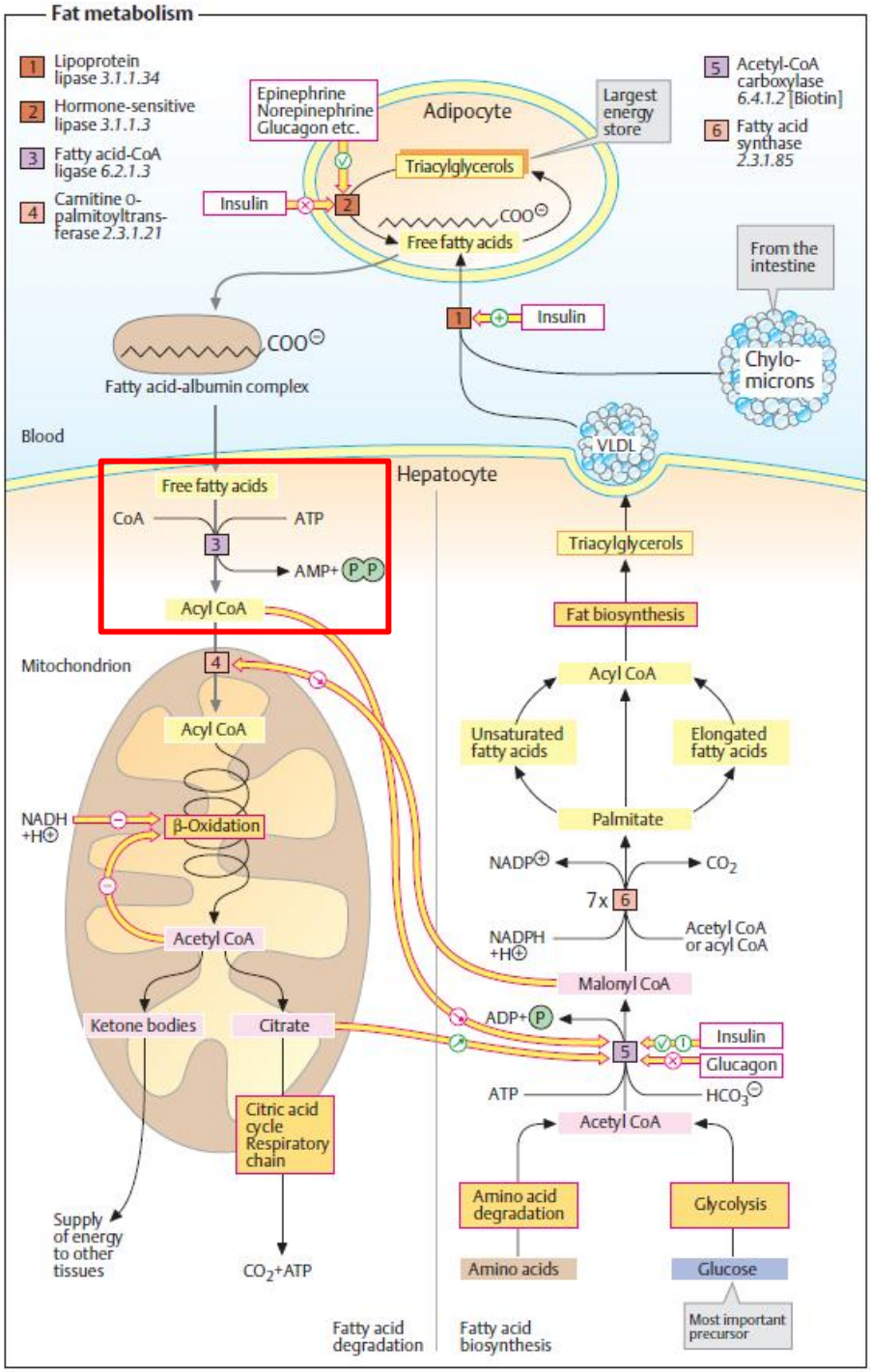
Activation of fatty acid to acylCoA

- This step is the premise to all reactions of fatty acids:

  Catabolism: β-oxidation

  Anabolism: biosynthesis of lipids



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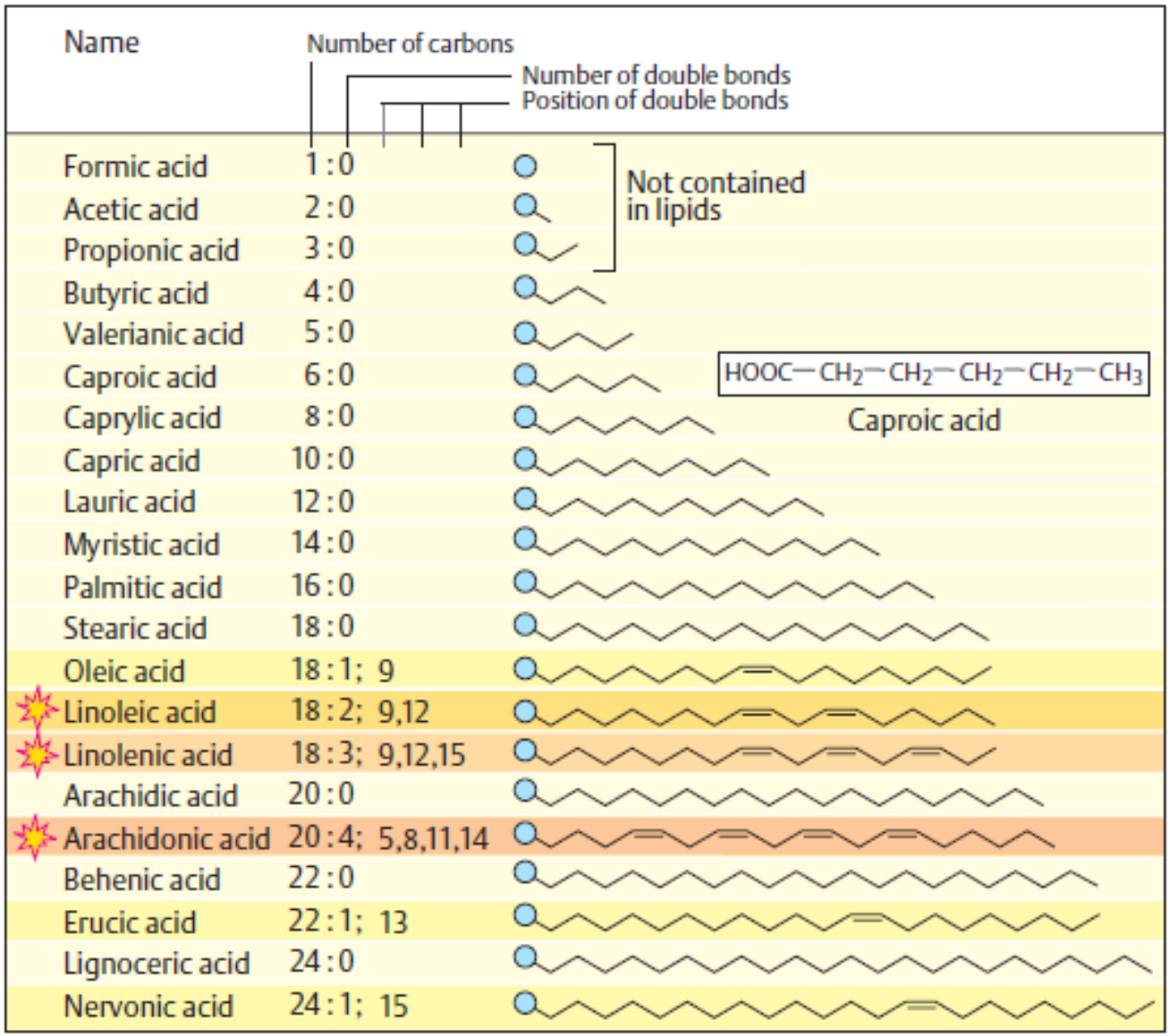
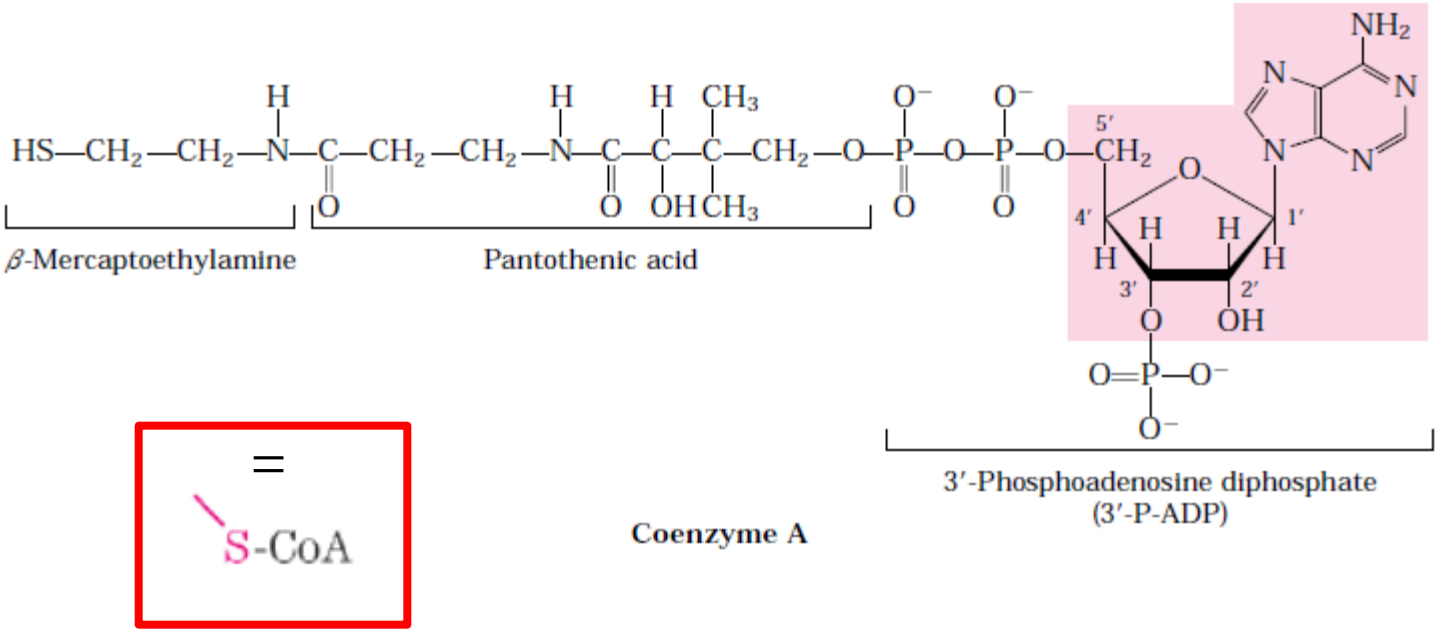
Carboxylic acids

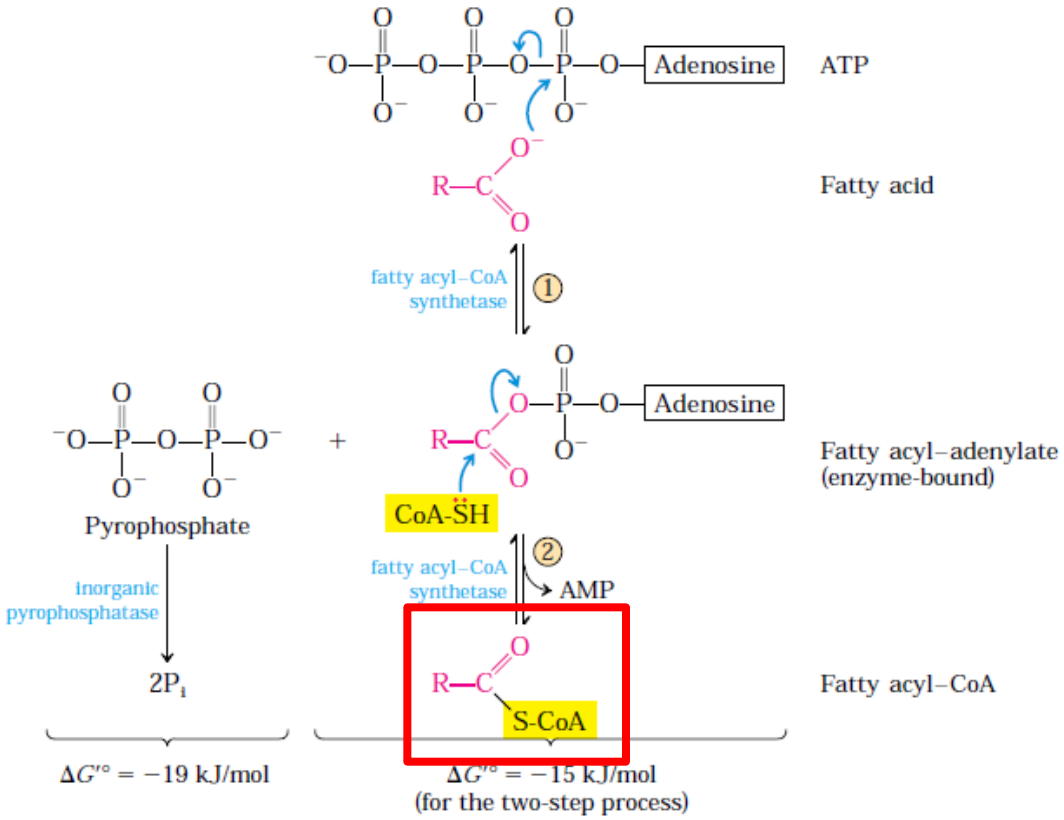




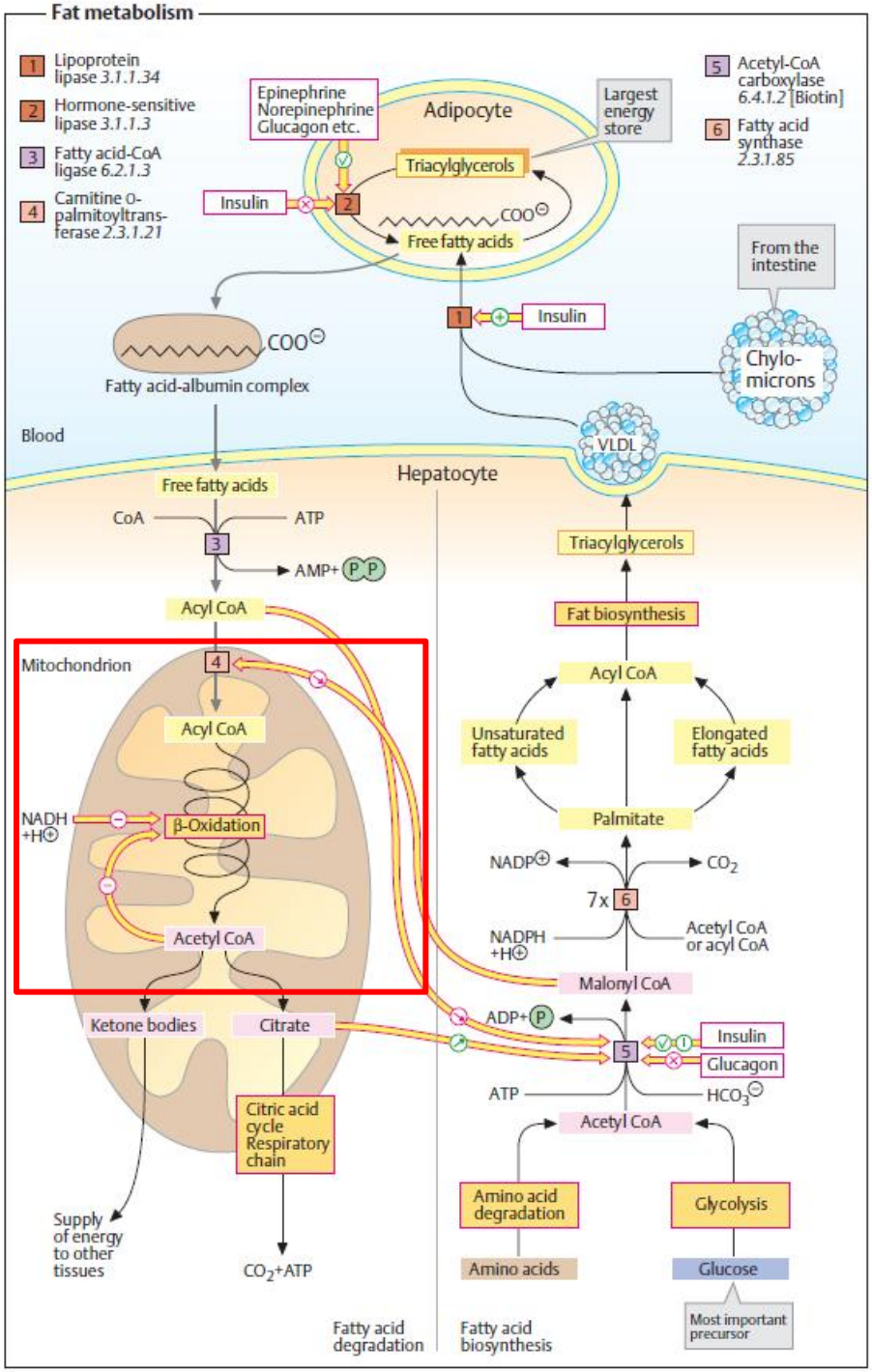
Coenzyme A

Coenzyme A (CoA) functions in acyl group transfer reactions; the acyl group (such as the acetyl or acetoacetyl group) is attached to the CoA through a thioester linkage to the -mercaptoethylamine moiety. NAD^+ functions in hydride transfers, and FAD, the active form of vitamin B_2 (riboflavin), in electron transfers.

The **conversion of a fatty acid to a fatty acyl–CoA** is catalyzed by fatty acyl–CoA synthetase and inorganic pyrophosphatase. Fatty acid activation by formation of the fatty acyl–CoA derivative occurs in two steps. In step 1 , the carboxylate ion displaces the outer two (and) phosphates of ATP to form a fatty acyl–adenylate, the mixed anhydride of a carboxylic acid and a phosphoric acid. The other product is PPi, an excellent leaving group that is immediately hydrolyzed to two Pi, pulling the reaction in the forward direction. In step 2 , the thiol group of coenzyme A carries out nucleophilic attack on the enzyme-bound mixed anhydride, displacing AMP and forming the thioester fatty acyl–CoA. The overall reaction is highly exergonic.



Fatty acyl–CoA esters formed at the cytosolic side of the outer mitochondrial membrane can be transported into the mitochondrion and oxidized to produce ATP, or they can be used in the cytosol to synthesize lipids.



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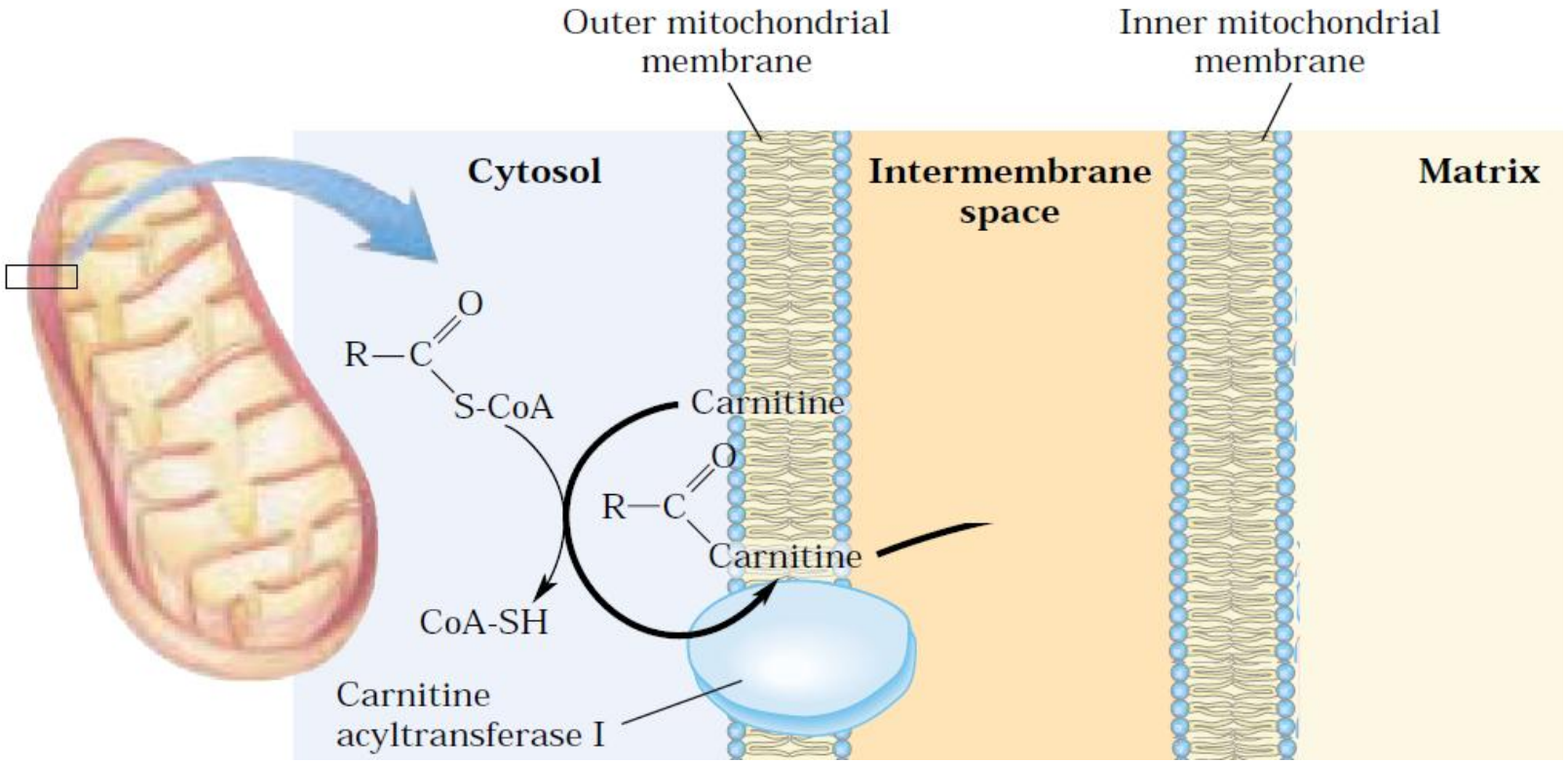
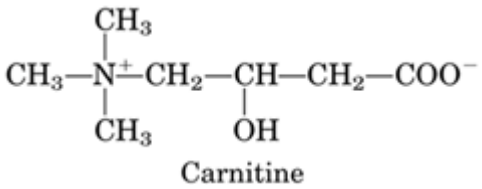
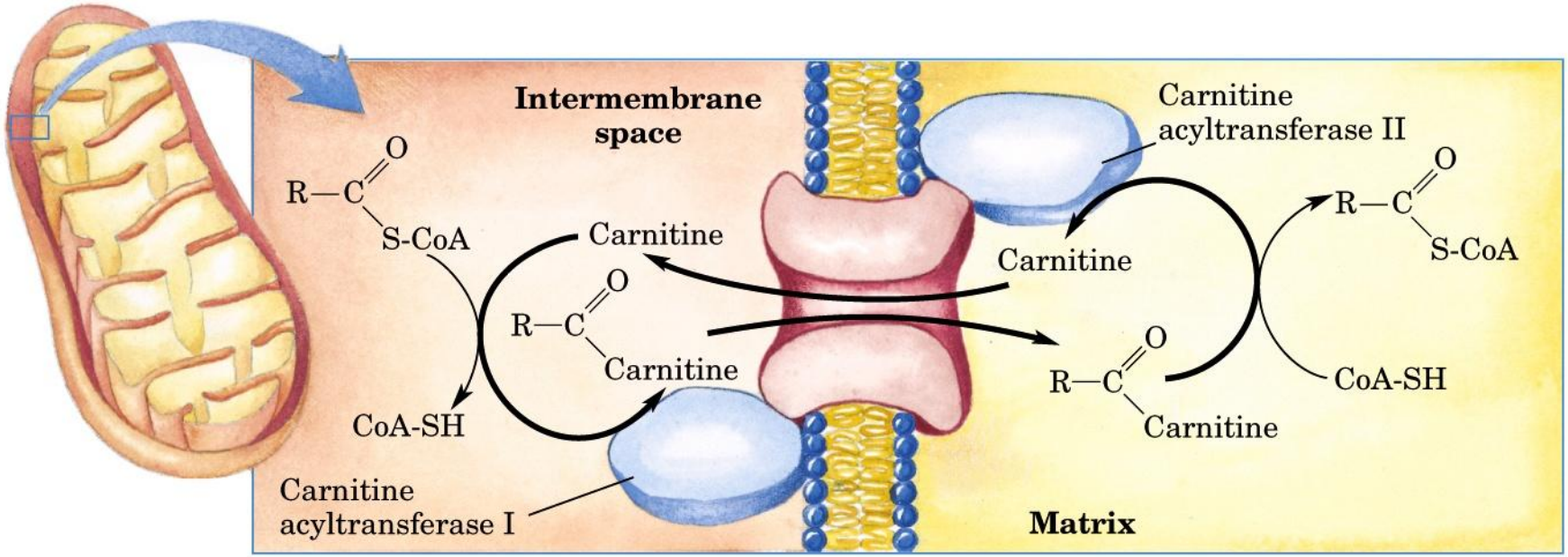


Fatty acids destined for mitochondrial oxidation are transiently attached to the hydroxyl group of **carnitine** to form fatty acyl–carnitine. This transesterification is catalyzed by **carnitine acyltransferase I** (*Mr* 88,000), in the outer membrane. The carnitine ester is formed on the cytosolic face of the outer membrane, then moves across the outer membrane to the intermembrane space.

L-Carnitine ([-],β-hydroxy-γ-*N*-trimethylaminobutyric acid) is a highly polar, water-soluble quaternary amine that exists as a zwitterion under physiological conditions.



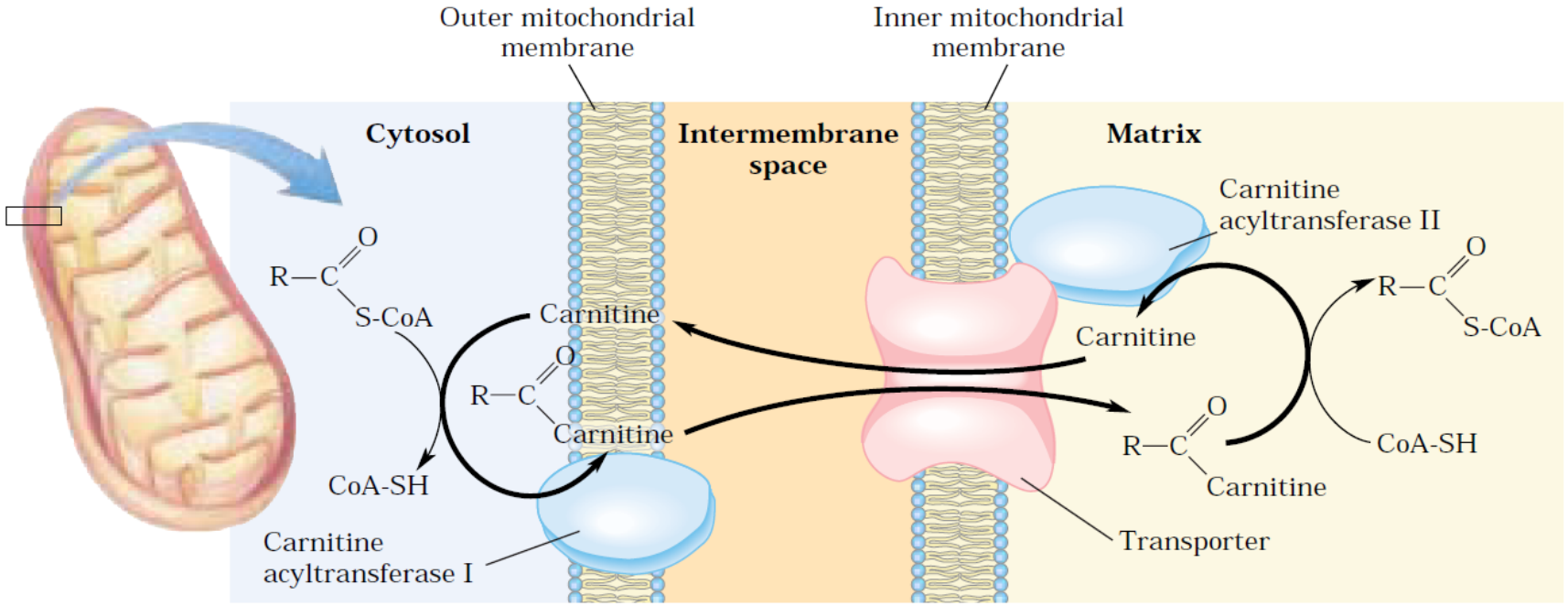
The carnitine shuttle



Fatty acid entry into mitochondria via the acyl-carnitine/carnitine transporter. After fatty acyl–carnitine is formed at the outer membrane or in the intermembrane space, it moves into the matrix by facilitated diffusion through the transporter in the inner membrane.

In the matrix, the acyl group is transferred to mitochondrial coenzyme A, freeing carnitine to return to the intermembrane space through the same transporter. Acyltransferase I is inhibited by malonyl-CoA, the first intermediate in fatty acid synthesis. This inhibition prevents the simultaneous synthesis and degradation of fatty acids.

The carnitine shuttle



The three-step process for transferring fatty acids into the mitochondrion — (*a*) esterification to CoA, (*b*) transesterification to carnitine followed by transport, and (*c*) transesterification back to CoA — links two **separate pools of coenzyme A and of fatty acyl–CoA, one in the cytosol, the other in mitochondria**.

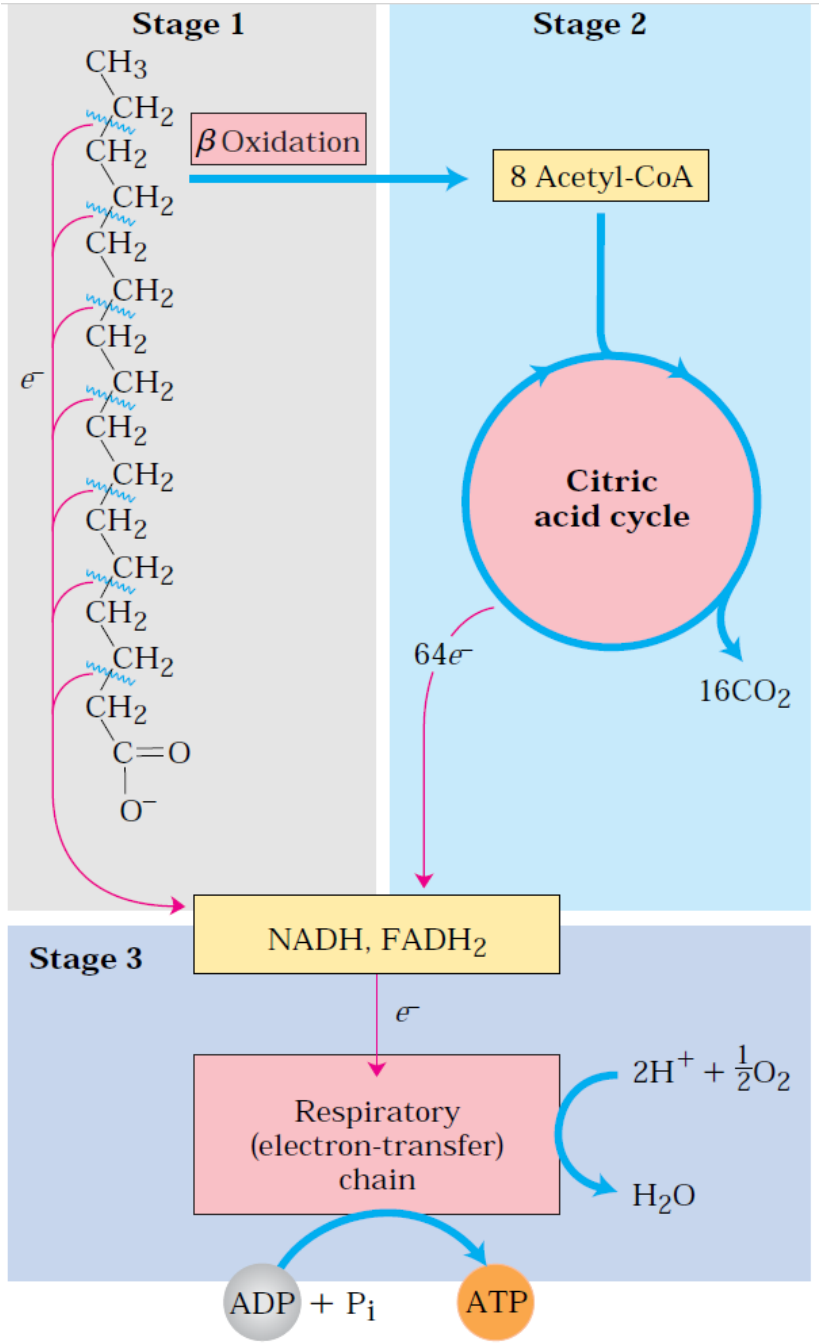
These pools have different functions. Coenzyme A in the mitochondrial matrix is largely used in oxidative degradation of pyruvate, fatty acids, and some amino acids, whereas cytosolic coenzyme A is used in the biosynthesis of fatty acids. Fatty acyl–CoA in the cytosolic pool can be used for membrane lipid synthesis or can be moved into the mitochondrial matrix for oxidation and ATP production. Conversion to the carnitine ester commits the fatty acyl moiety to the oxidative fate.

Stages of fatty acid oxidation.

Stage 1: A long-chain fatty acid is oxidized to yield acetyl residues in the form of acetyl-CoA. This process is called **β-oxidation**.

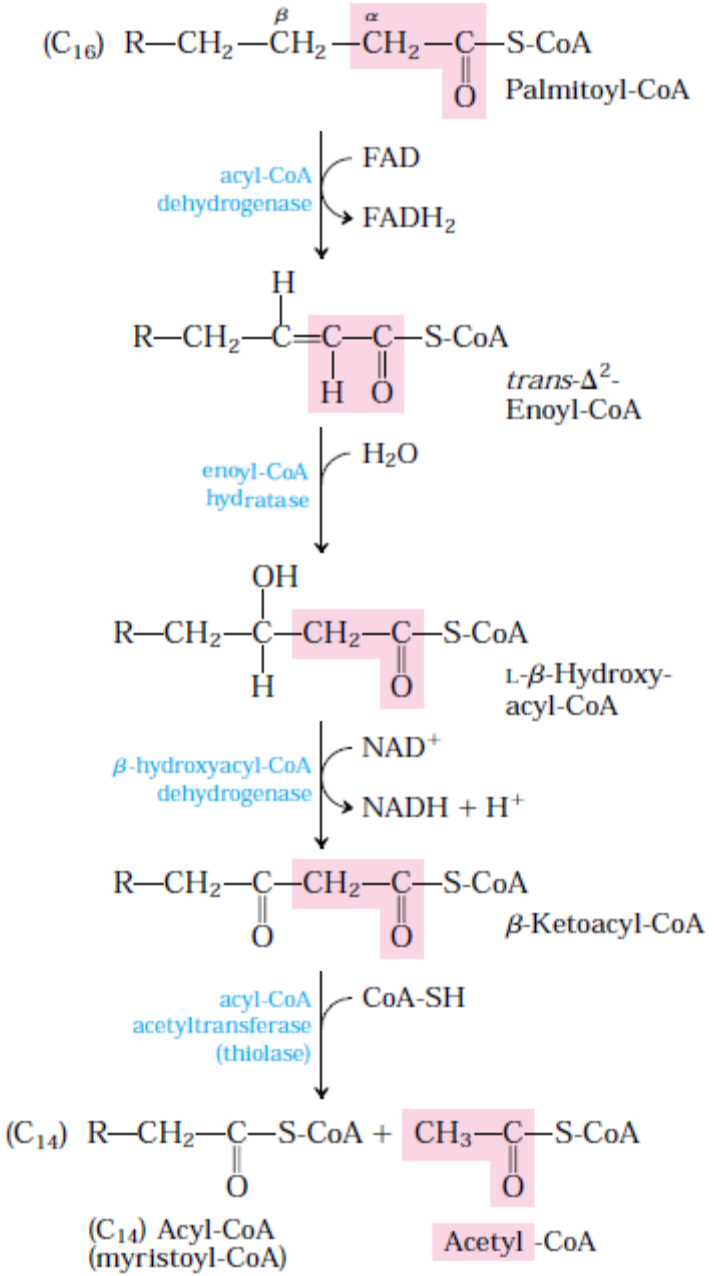
Stage 2: The acetyl groups are oxidized to CO_2 via the citric acid cycle.

Stage 3: Electrons derived from the oxidations of stages 1 and 2 pass to O_2 via the mitochondrial respiratory chain, providing the energy for ATP synthesis by oxidative phosphorylation.



The β–oxidation of saturated fatty acids has four basic steps

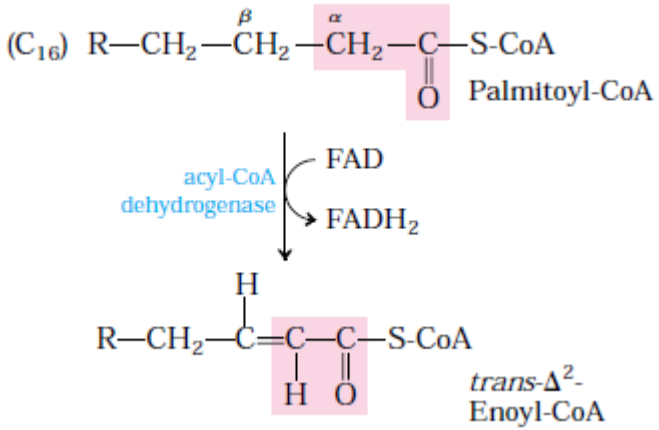
Four enzyme-catalyzed reactions make up the first stage of fatty acid b-β-oxidation.



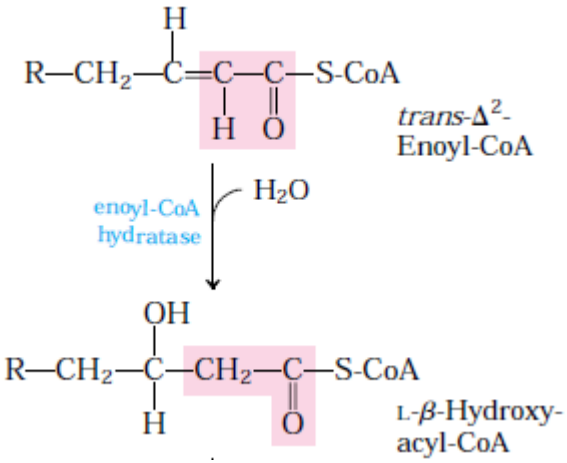
First, **dehydrogenation** of fatty acyl–CoA produces a double bond between the and carbon atoms (C-2 and C-3), yielding a ***trans*-2-enoyl-CoA** (the symbol 2 designates the position of the double bond). Note that the new double bond has the trans configuration, whereas the double bonds in naturally occurring unsaturated fatty acids are normally in the cis configuration.

This first step is catalyzed by three isozymes of **acyl-CoA dehydrogenase,** each specific for a range of fatty-acyl chain lengths: very-long-chain acyl-CoA dehydrogenase (VLCAD), acting on fatty acids of 12 to 18 carbons; medium-chain (MCAD), acting on fatty acids of 4 to 14 carbons; and short-chain (SCAD), acting on fatty acids of 4 to 8 carbons. All three isozymes are flavoproteins with FAD as a prosthetic group.

The electrons removed from the fatty acyl–CoA are transferred to FAD, and the reduced form of the dehydrogenase immediately donates its electrons to an electron carrier of the mitochondrial respiratory chain, the electron-transferring flavoprotein (ETF).



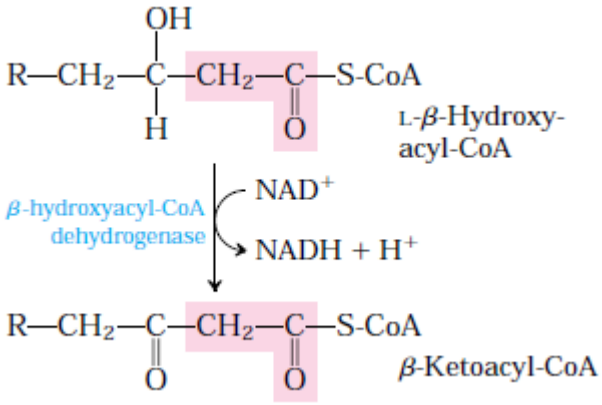
In the second step of the β-oxidation cycle **water is added** to the double bond of the *trans*-2-enoyl-CoA to form the L stereoisomer of **β-hydroxyacyl-CoA (3-hydroxyacyl-CoA).**



In the third step, **L-β-hydroxyacyl-CoA is dehydrogenated** to form **β-ketoacyl-CoA,** by the action of **β-hydroxyacyl-CoA dehydrogenase;** NAD is the electron acceptor.

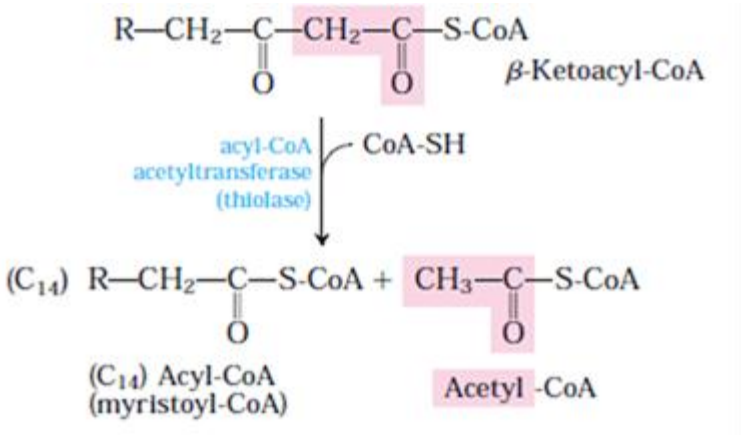
This enzyme is absolutely specific for the L stereoisomer of hydroxyacyl-CoA.

The NADH formed in the reaction donates its electrons to NADH dehydrogenase, an electron carrier of the respiratory chain, and ATP is formed from ADP as the electrons pass to O_2.



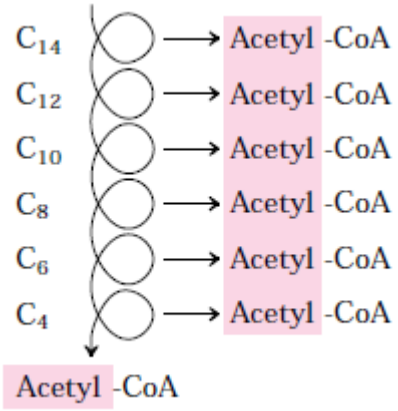
The fourth and last step of the β-oxidation cycle is catalyzed by **acyl-CoA acetyltransferase,** more commonly called **thiolase,** which promotes reaction of -ketoacyl-CoA with a molecule of free coenzyme A to split off the carboxyl-terminal two-carbon fragment of the original fatty acid as acetyl-CoA.

The other product is the coenzyme A thioester of the fatty acid, now shortened by two carbon atoms.

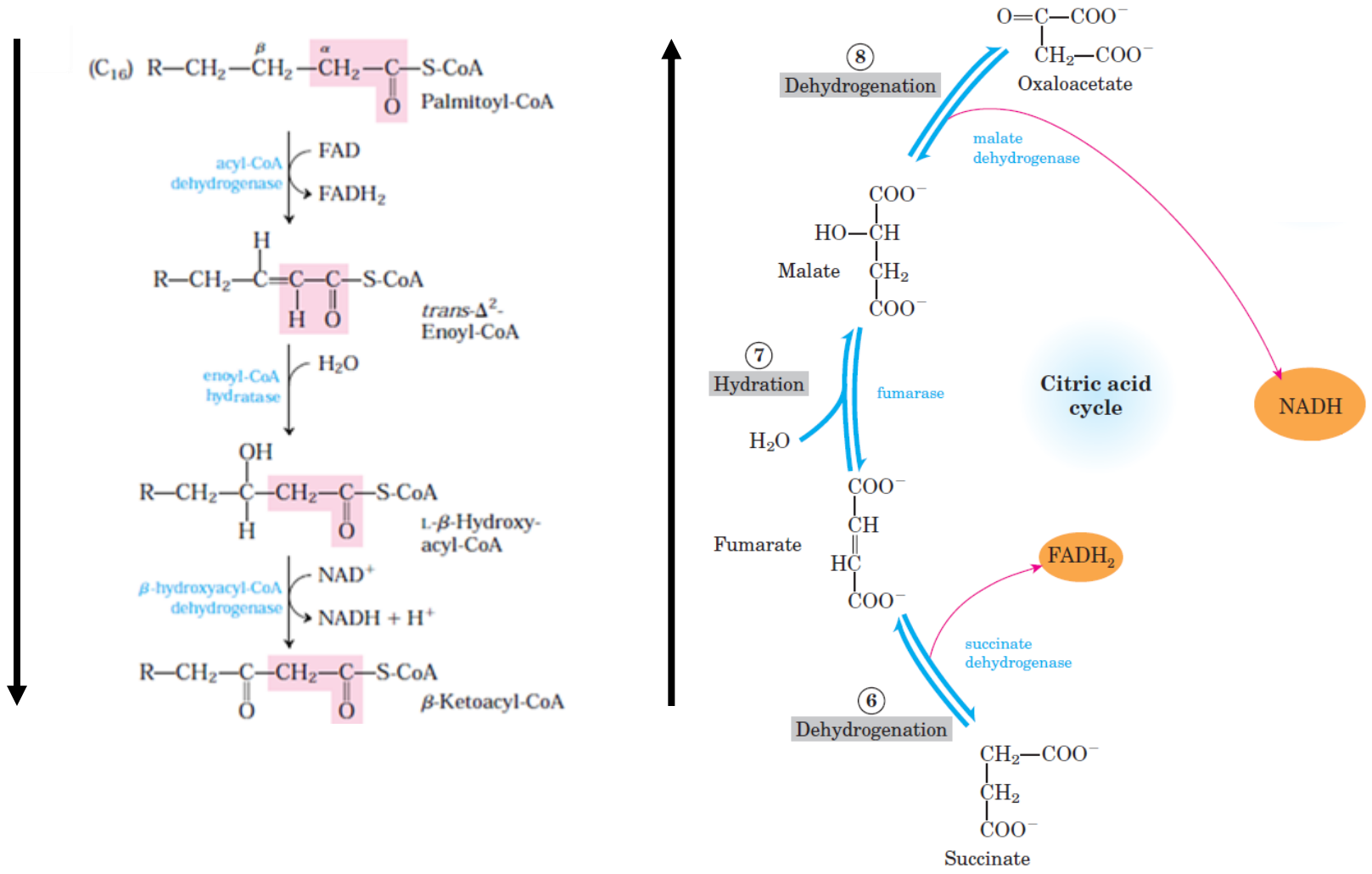


In each pass through the four-step sequence, one acetyl residue (shaded in pink) is removed in the form of acetyl-CoA from the carboxyl end of the fatty acyl chain—in this example palmitate (C16), which enters as palmitoyl-CoA.

Six more passes through the pathway yield seven more molecules of acetyl-CoA, the seventh arising from the last two carbon atoms of the 16-carbon chain. Eight molecules of acetyl-CoA are formed in all.



Three steps of β-oxidation remind the steps occurring in the production of oxaloacetate from succinate in the citric acid cycle



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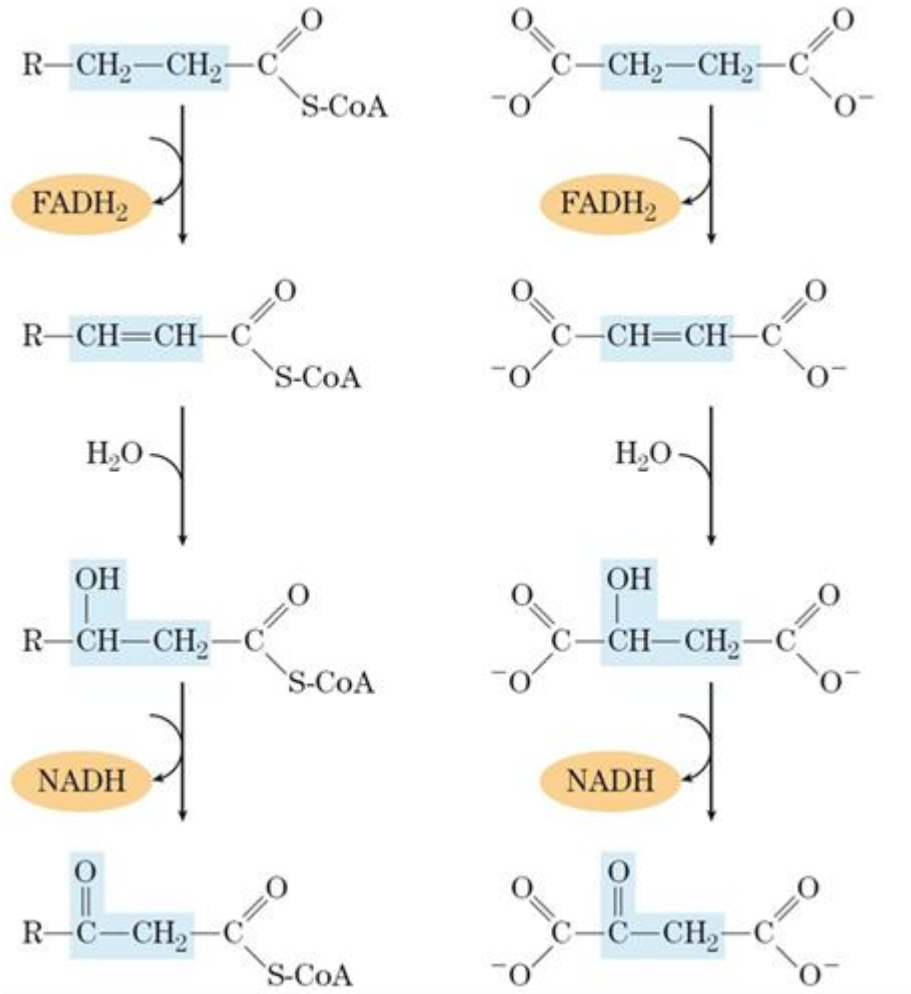

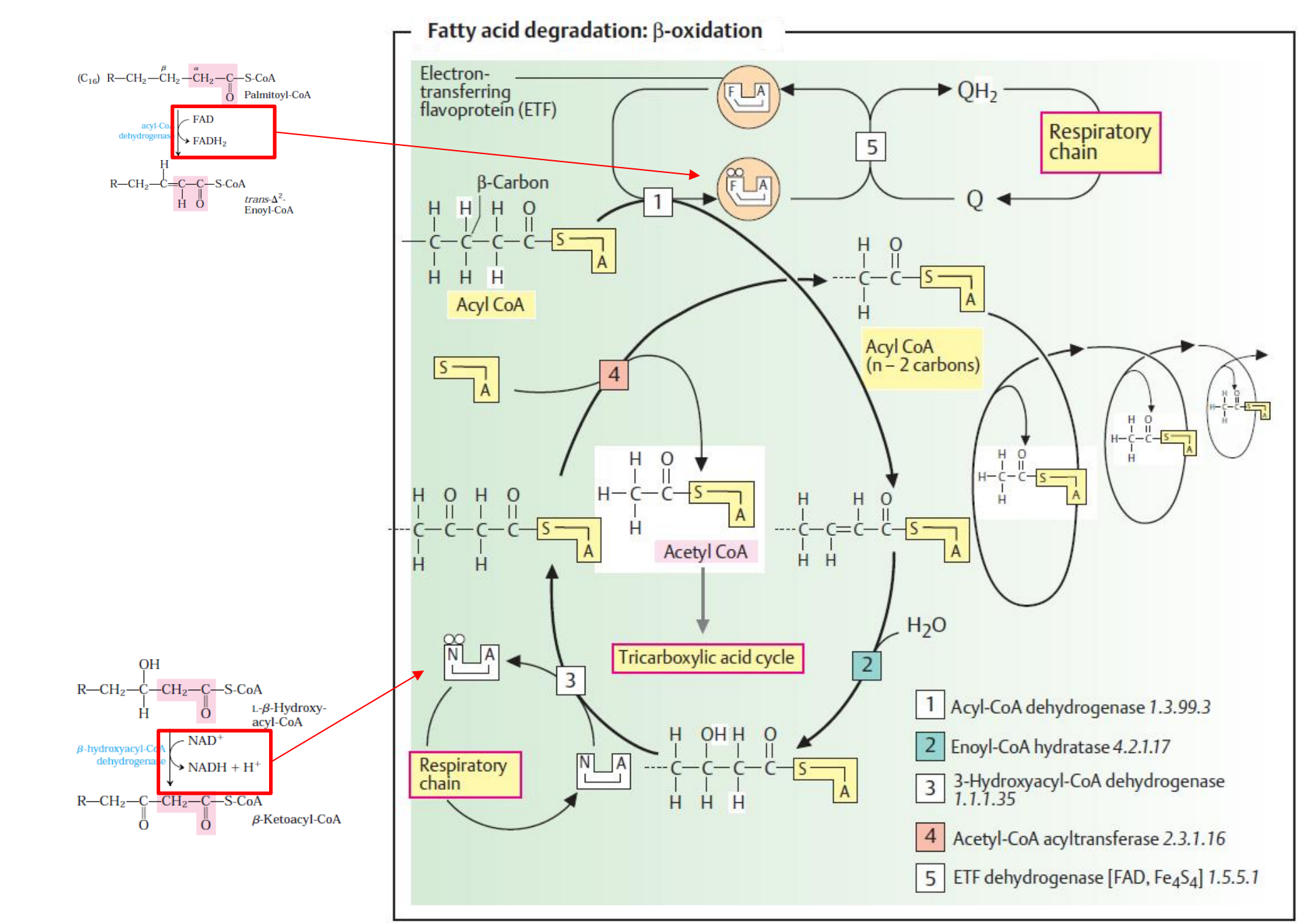

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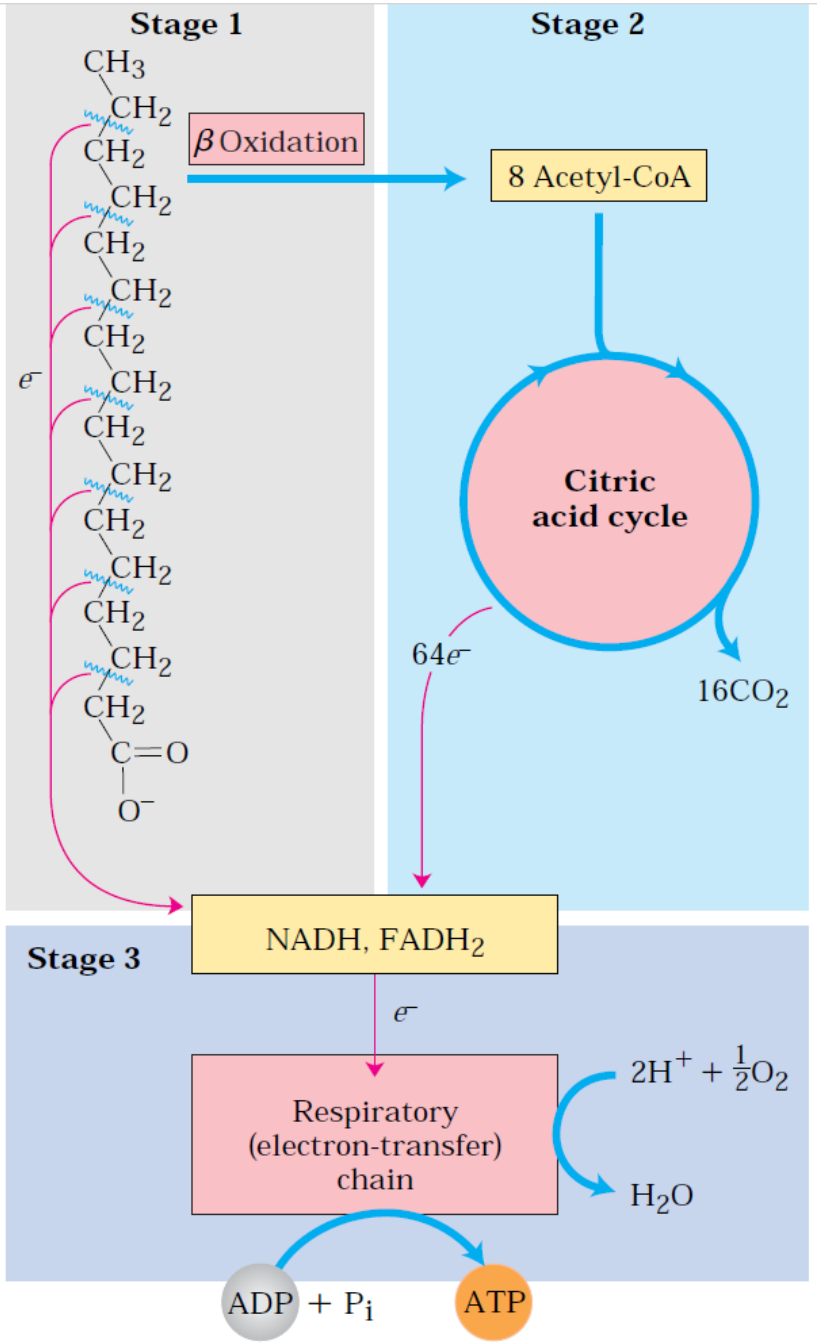


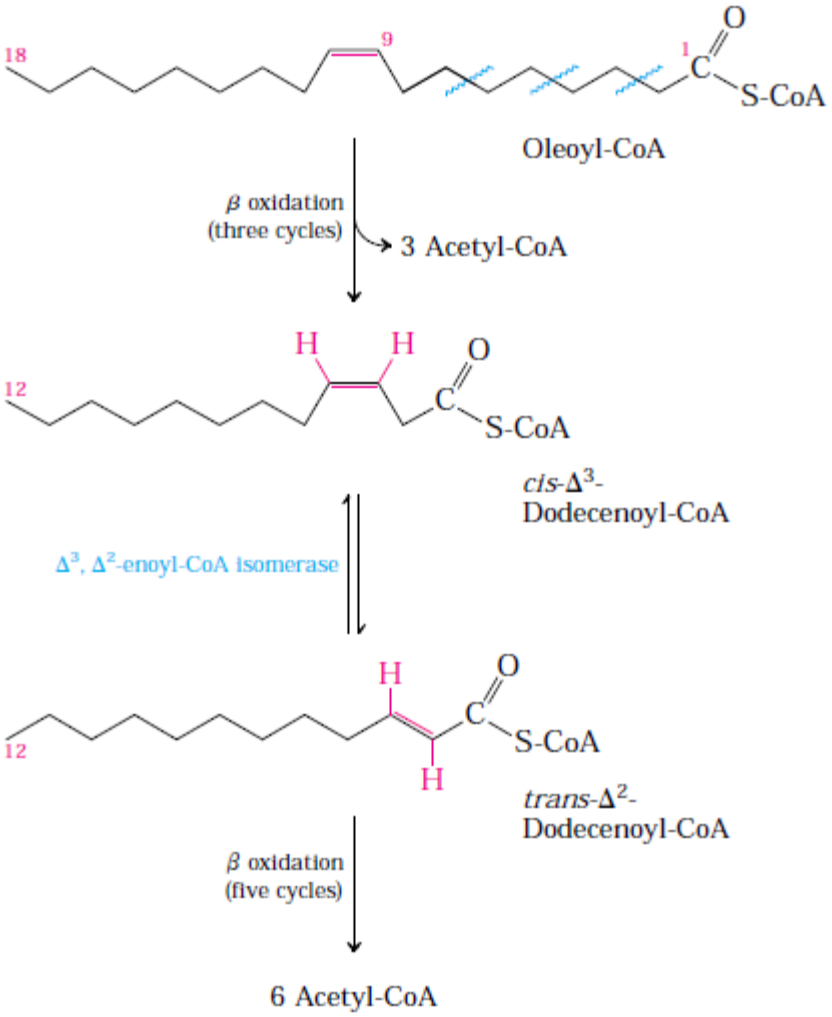
The complete oxidation of palmitoyl-CoA to carbon dioxide and water yields

Palmitoyl-CoA 23 O_2 108 P_i 108 ADP
108 (**106**) **ATP** 16 CO_2 23 H_2O

Oxidation of unsaturated fatty acids requires additional reactions

Here, as an example of a **monounsaturated fatty acid**, oleic acid [oleoyl-CoA (Δ9)].

Oxidation requires an additional enzyme, enoyl-CoA isomerase, to reposition the double bond, converting the cis isomer to a trans isomer, a normal intermediate in oxidation.

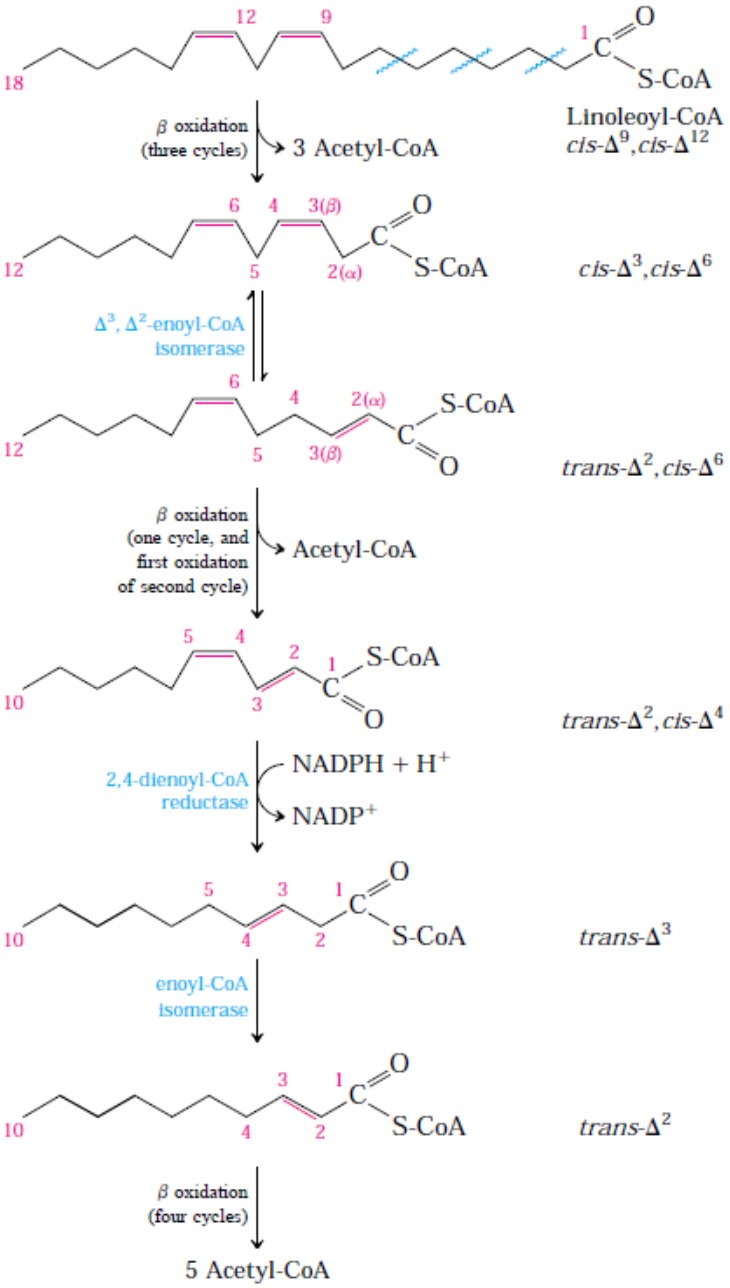


Oxidation of unsaturated fatty acids requires additional reactions

Here an example of **polyunsaturated fatty acid,** linoleic acid as linoleoyl-CoA ($\Delta_{9,12}$).

Oxidation requires a second auxiliary enzyme in addition to enoyl-CoA isomerase: NADPH-dependent 2,4-dienoyl-CoA reductase.

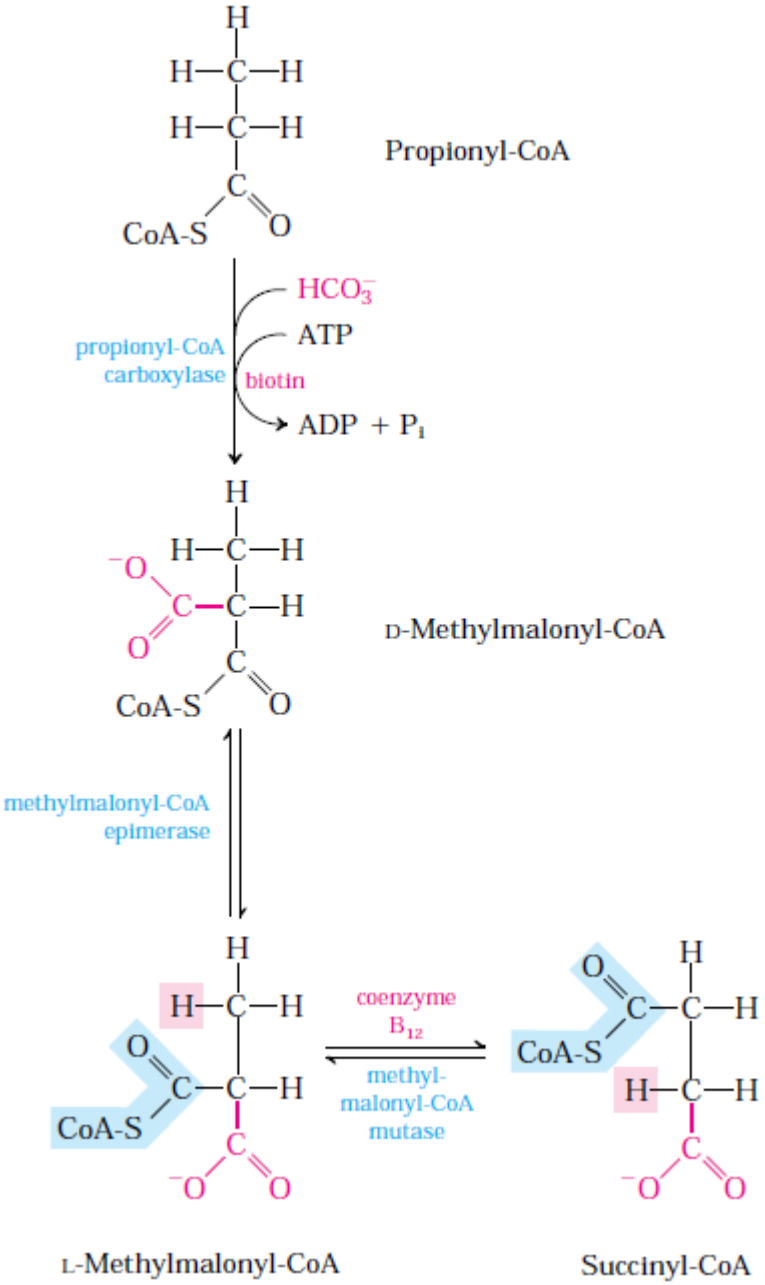
The combined action of these two enzymes converts a *trans*-Δ_2,*cis*-Δ_4-dienoyl-CoA intermediate to the *trans*-Δ_2-enoyl-CoA substrate necessary for oxidation.

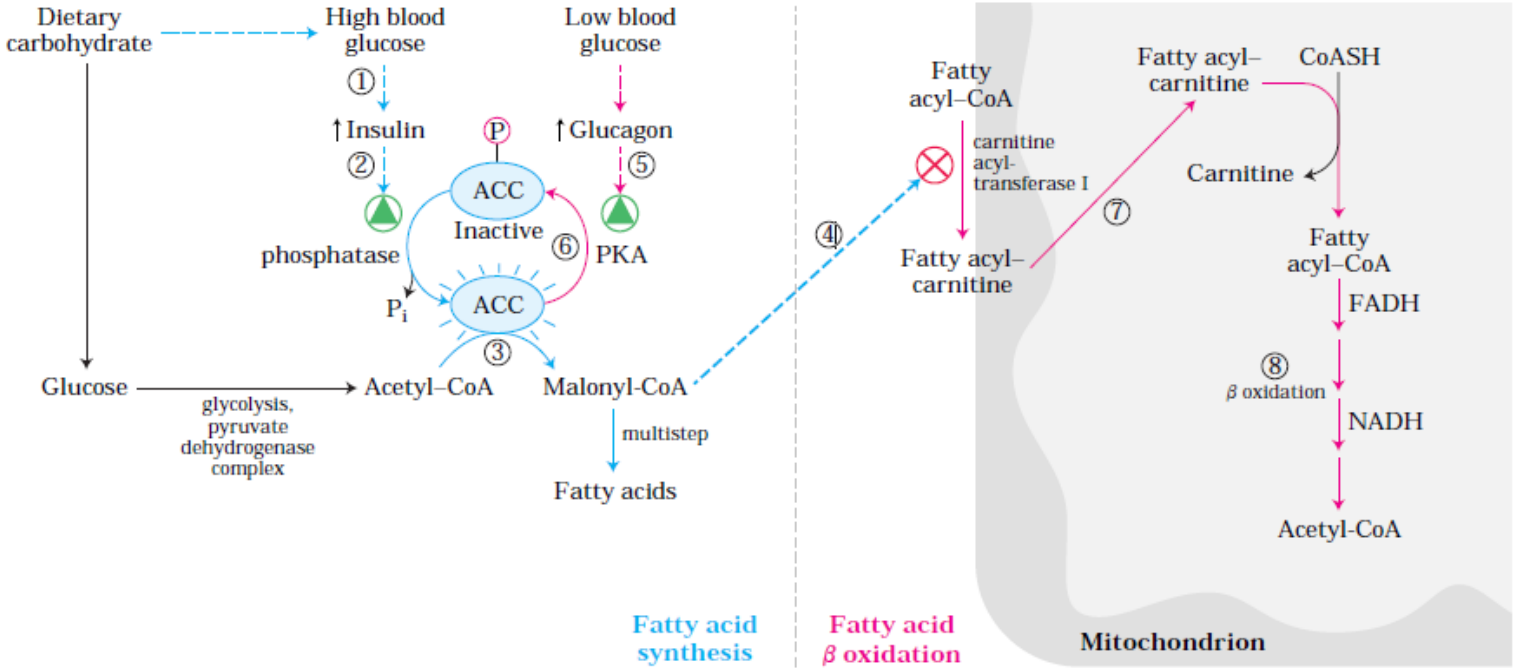


Oxidation of propionyl-CoA produced by oxidation of odd-number fatty acids.

The sequence involves the carboxylation of propionyl-CoA to D-methylmalonyl-CoA and conversion of the latter to succinyl-CoA - which can enter the citric acid cycle.

This conversion requires epimerization of D- to L-methylmalonyl-CoA, followed by a reaction in which substituents on adjacent carbon atoms exchange positions.



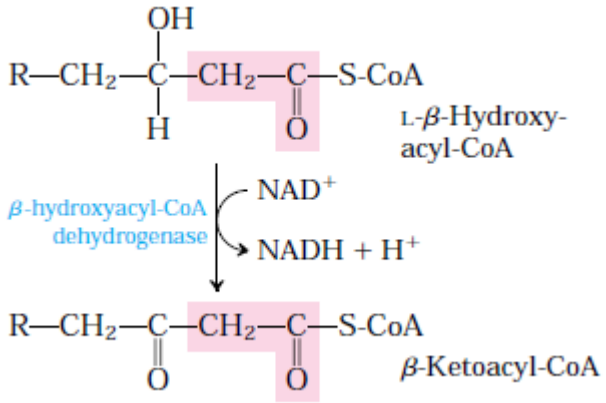


Malonyl-CoA, the first intermediate in the cytosolic biosynthesis of long-chain fatty acids from acetyl-CoA increases in concentration whenever an animal is well supplied with carbohydrate; excess glucose that cannot be oxidized or stored as glycogen is converted in the cytosol into fatty acids for storage as triacylglycerol.

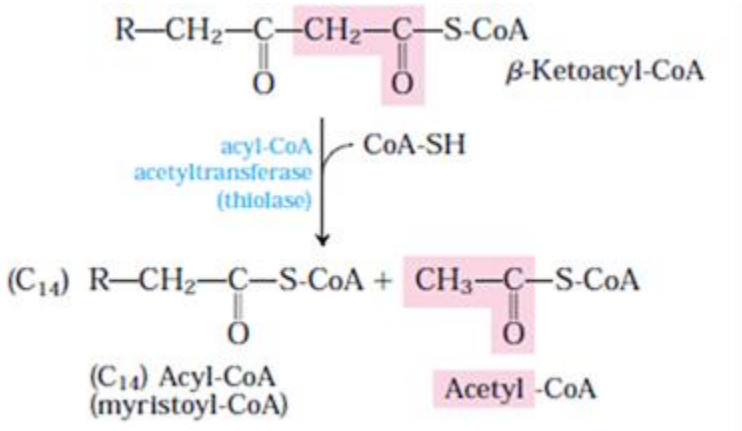
The **inhibition of carnitine acyltransferase I by malonyl-CoA ensures that the oxidation of fatty acids is inhibited** whenever the liver is amply supplied with glucose as fuel and is actively making triacylglycerols from excess glucose.

Two of the enzymes of β-oxidation are also regulated by metabolites that signal energy sufficiency:

- **when the [NADH]/[NAD] ratio is high, β-hydroxyacyl-CoA dehydrogenase is inhibited**.

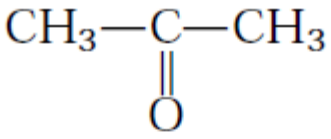


- **high concentrations of acetyl-CoA inhibit thiolase**.

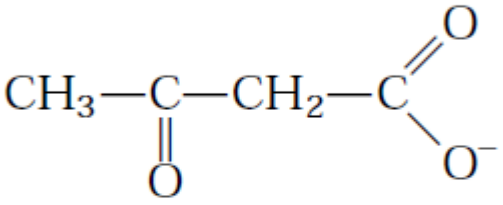


Ketone *bodies*

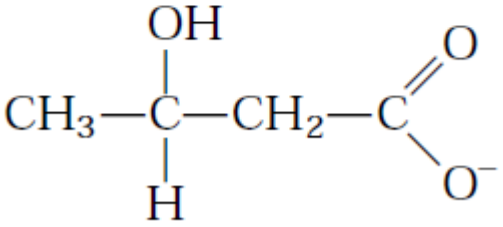
In humans and most other mammals, acetyl-CoA formed in the liver during oxidation of fatty acids can either enter the citric acid cycle or undergo conversion to the "ketone *bodies*," **acetone**, **acetoacetate**, and **D-β-hydroxybutyrate**, for export to other tissues.



Acetone



Acetoacetate

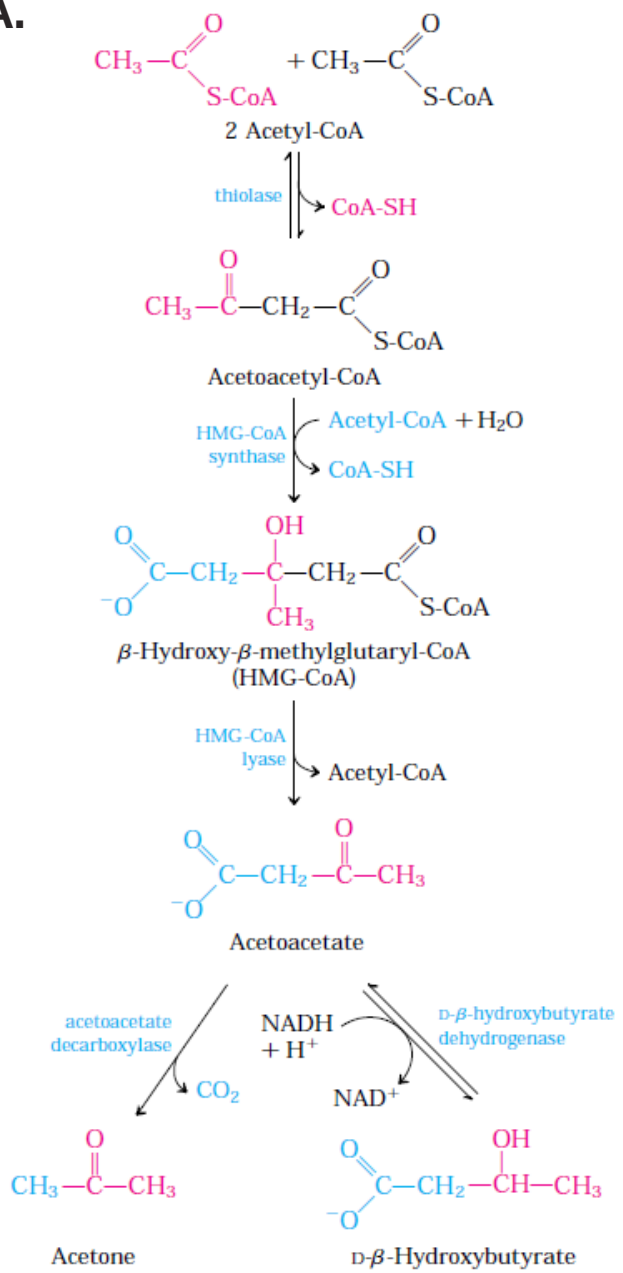


D-β-Hydroxybutyrate

Formation of ketone bodies from acetyl-CoA.

Healthy, well-nourished individuals produce ketone bodies at a relatively low rate. When acetyl-CoA accumulates (e.g. in starvation or untreated diabetes), thiolase catalyzes the condensation of two acetyl-CoA molecules to acetoacetyl-CoA, the parent compound of the three ketone bodies. The reactions of ketone body formation occur in the matrix of liver mitochondria.

The six-carbon compound β-hydroxy-β-methylglutaryl-CoA (HMG-CoA) is also an intermediate of sterol biosynthesis, but the enzyme that forms HMG-CoA in that pathway is cytosolic. HMG-CoA lyase is present only in the mitochondrial matrix.

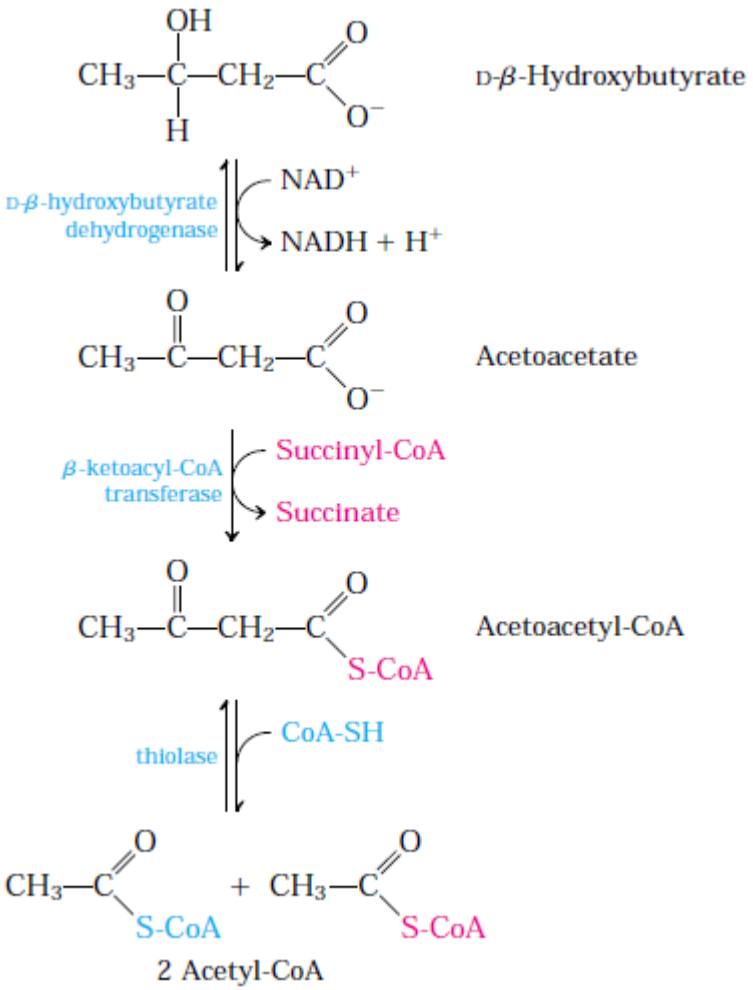


D-β-Hydroxybutyrate as a fuel.

D-β-Hydroxybutyrate synthesized in the liver passes into the blood and thus to other tissues
where it is converted in three steps to acetyl-CoA.

It is first oxidized to acetoacetate, which is activated with coenzyme A donated from succinyl-CoA, then split by thiolase.

The acetyl-CoA thus formed is used for energy production.



Ketone body formation and export from the liver.

Conditions that promote gluconeogenesis (untreated diabetes, severely reduced food intake) slow the citric acid cycle (by drawing off oxaloacetate) and enhance the conversion of acetyl-CoA to acetoacetate.

The released coenzyme A allows continued oxidation of fatty acids.