Metabolism of fatty acids. Ketone bodies

Lipids II

Ketone bodies

Under certain metabolic conditions associated with a high rate of fatty acid oxidation, liver produces considerable quantities of compounds like acetoacetate and β-OH butyric acid, which pass by diffusion into the blood.

Acetoacetate undergoes spontaneous decarboxylation to produce acetone. *These three substances are collectively known as "ketone bodies".*

Ketone bodies

Liver appears to be the only organ which produces ketone bodies and add to the blood.

Extrahepatic tissues can pick

up ketone bodies from the circulating blood and utilise them as respiratory substrates.

Acetone is a waste product which can be excreted via the lungs or with urine.

Synthesis of ketone bodies

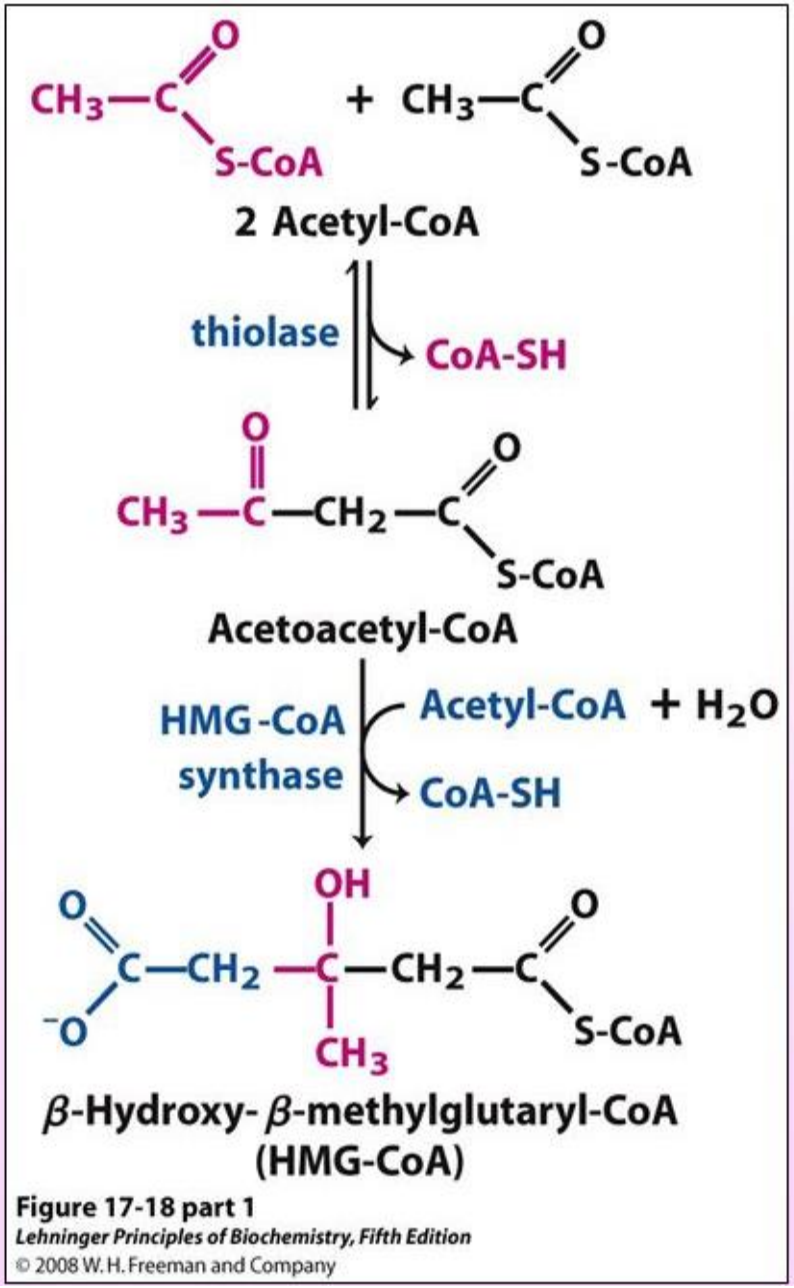

Figure 17-18 part 1
Lehninger Principles of Biochemistry, Fifth Edition

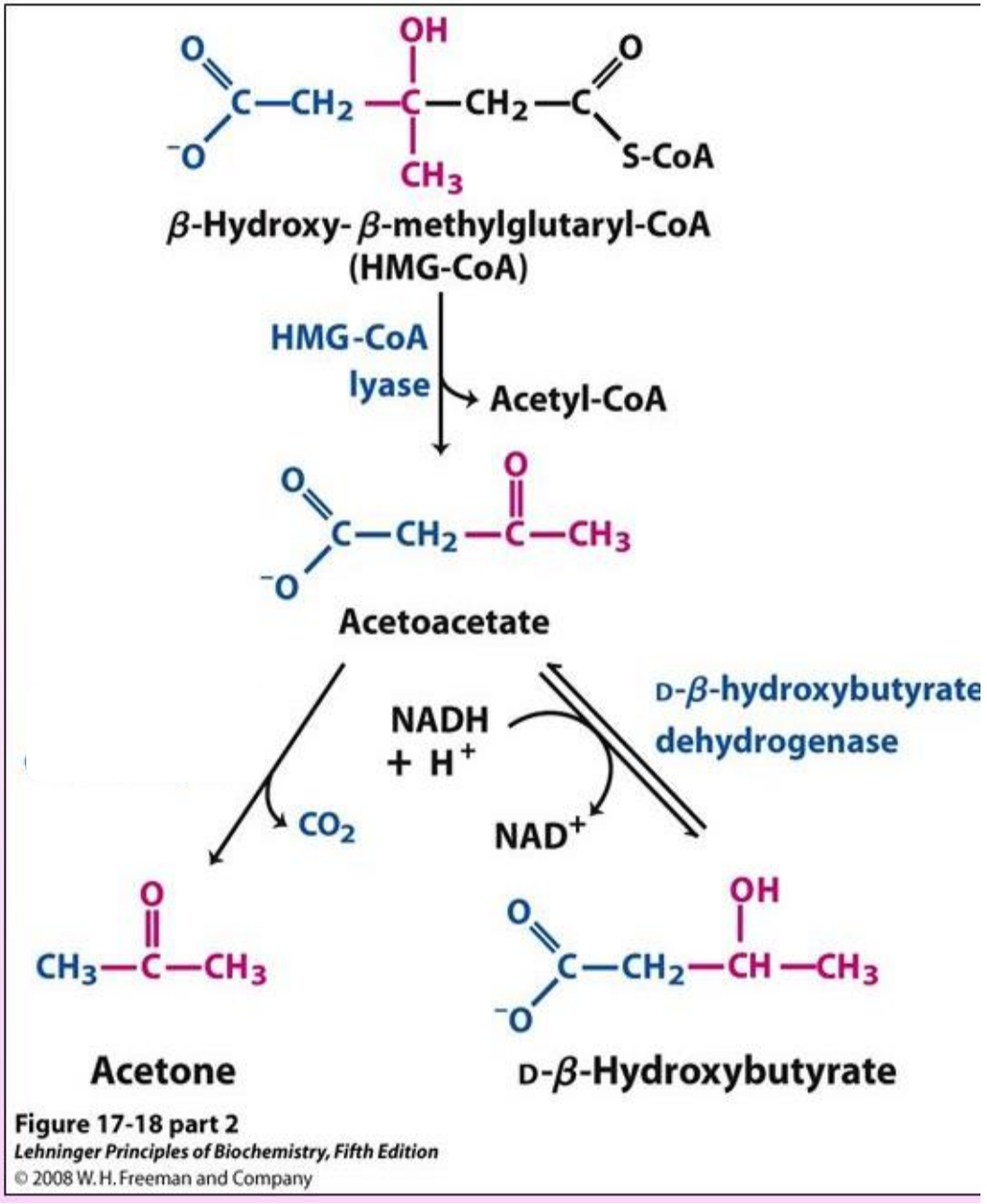

Figure 17-18 part 2
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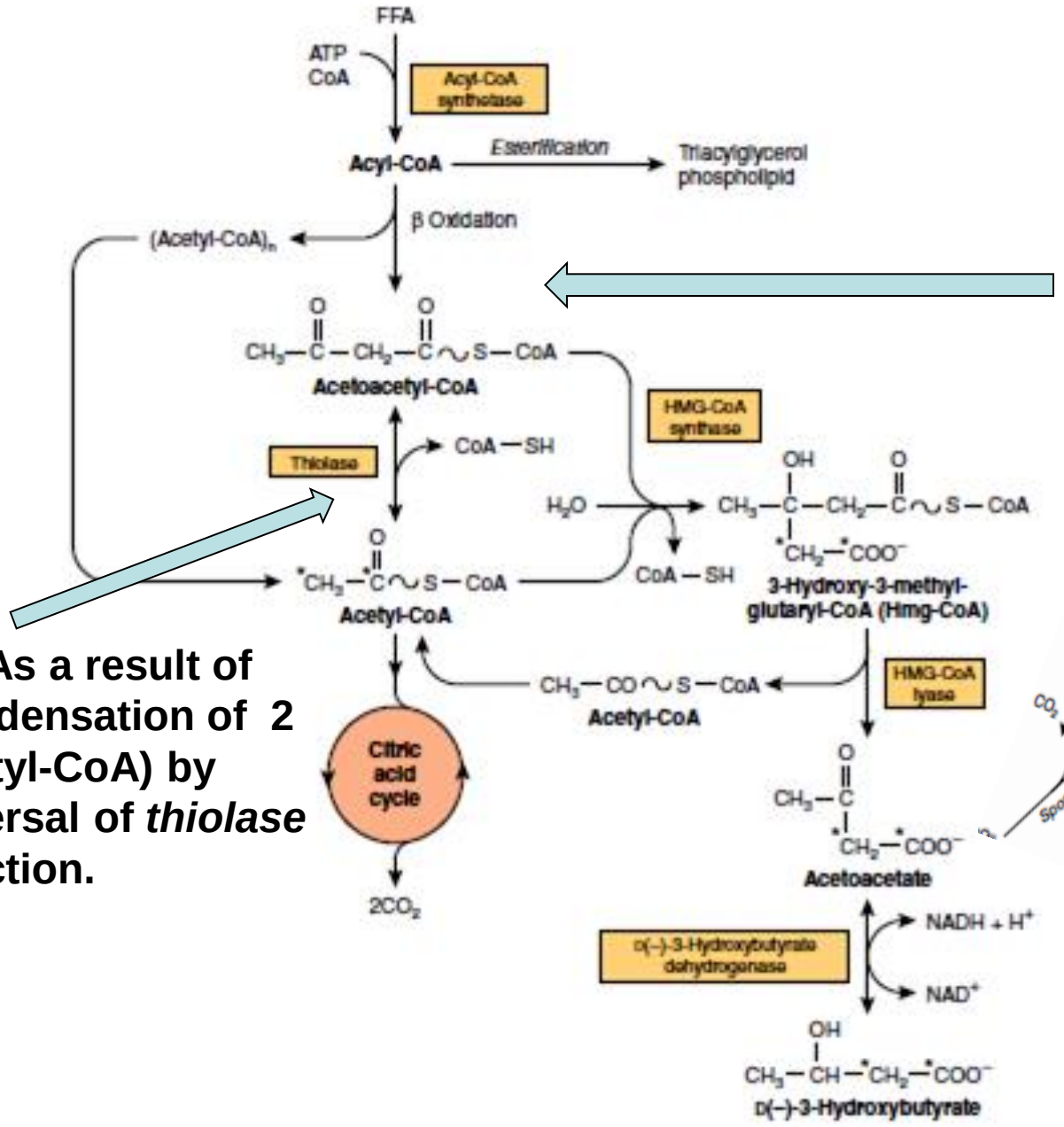
Reactions of synthesis of ketone bodies



Aceto-acetyl-CoA is the starting material for ketogenesis.

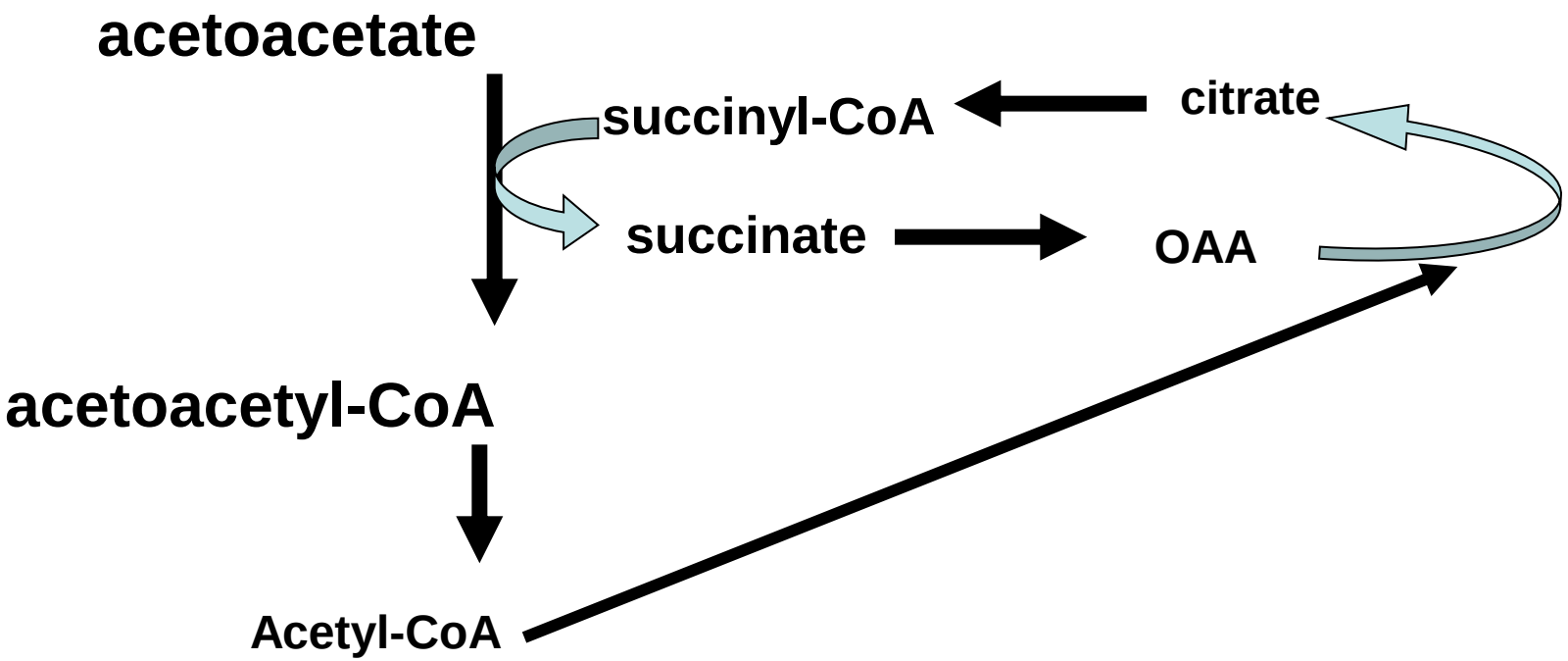
This can arise in two ways:

(a) directly from the terminal four carbons of a fatty acid during β-oxidation

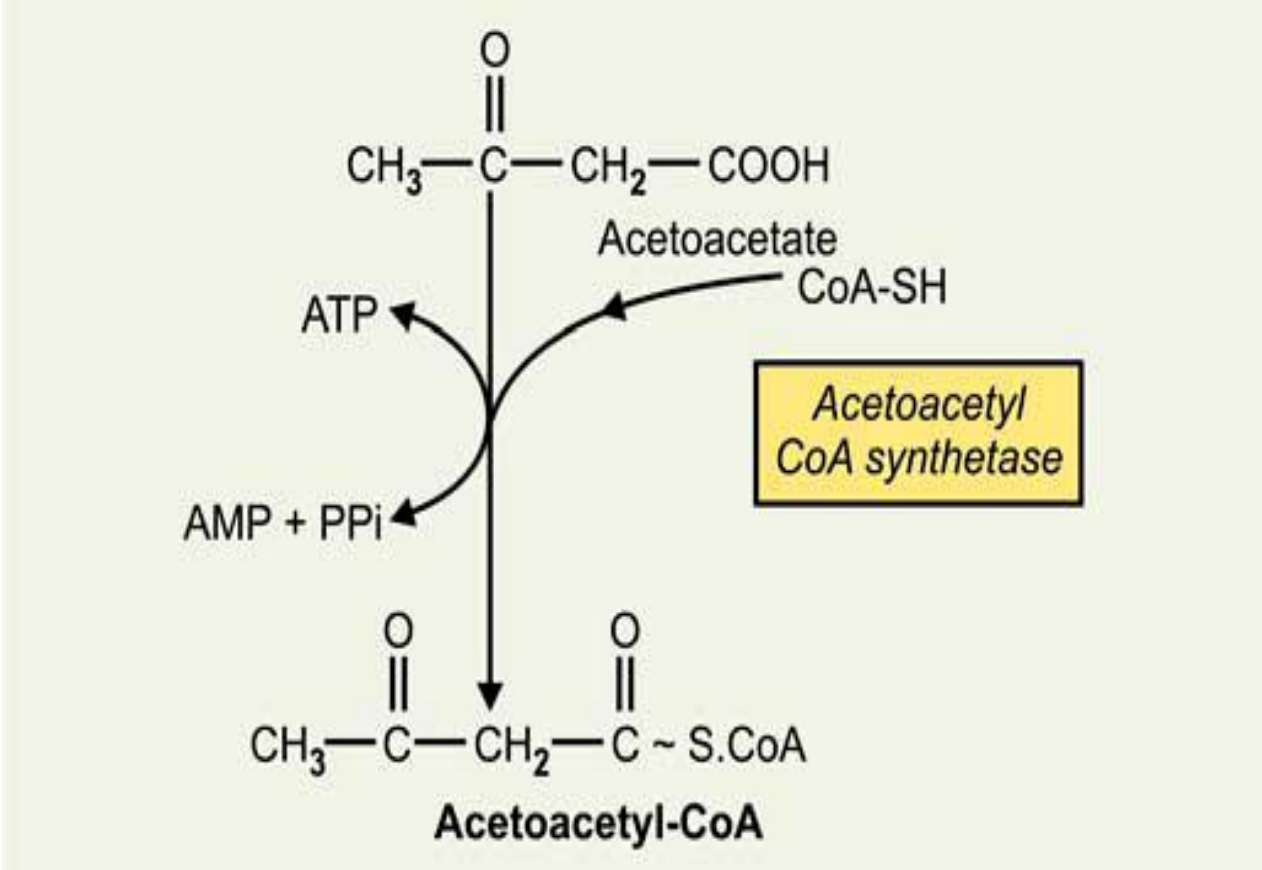
(b) As a result of condensation of 2 acetyl-CoA) by reversal of *thiolase* reaction.

In extrahepatic tissues acetoacetate is activated to acetoacetyl-CoA by

succinyl-CoA-acetoacetate CoA transferase



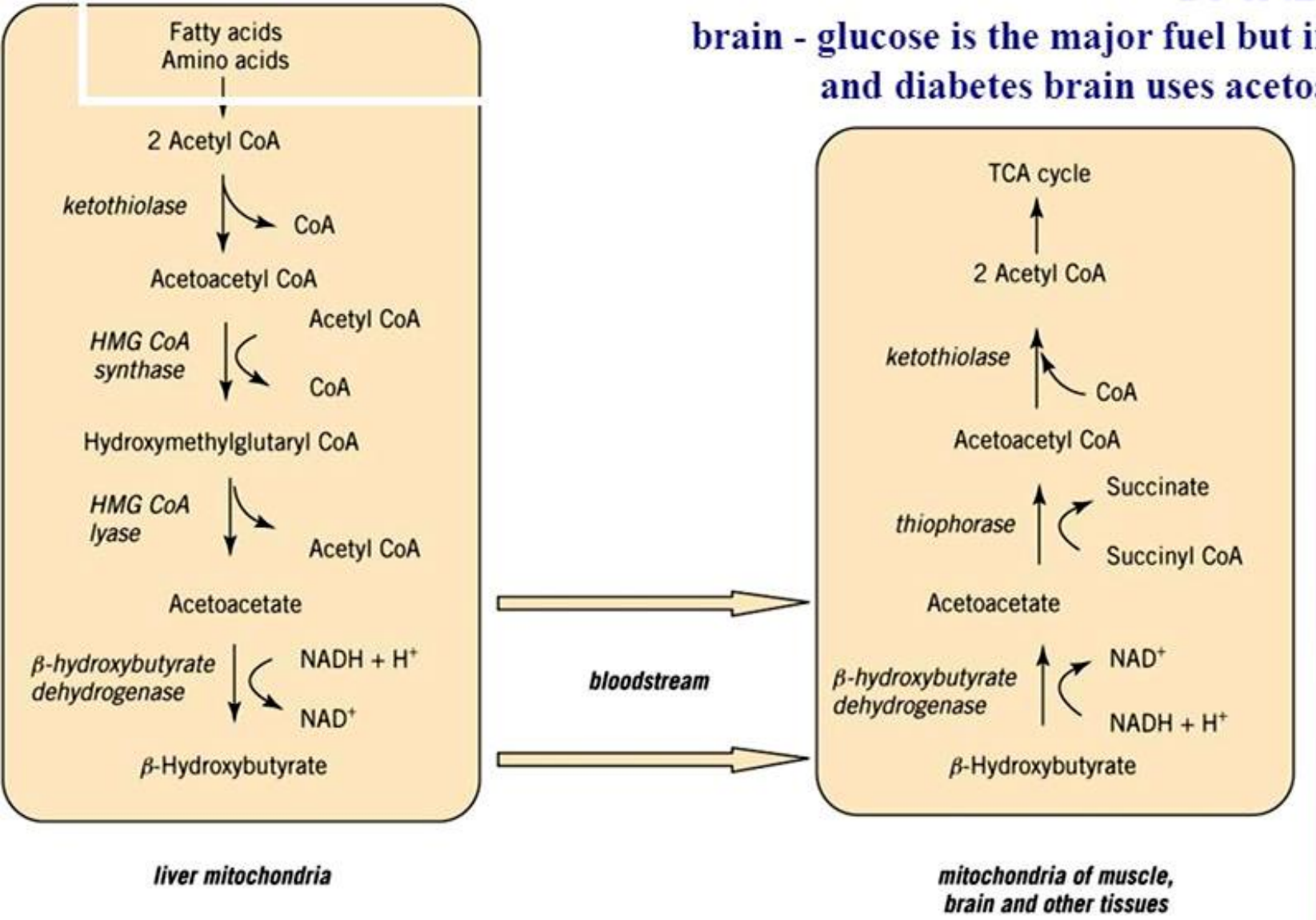
Second mechanism

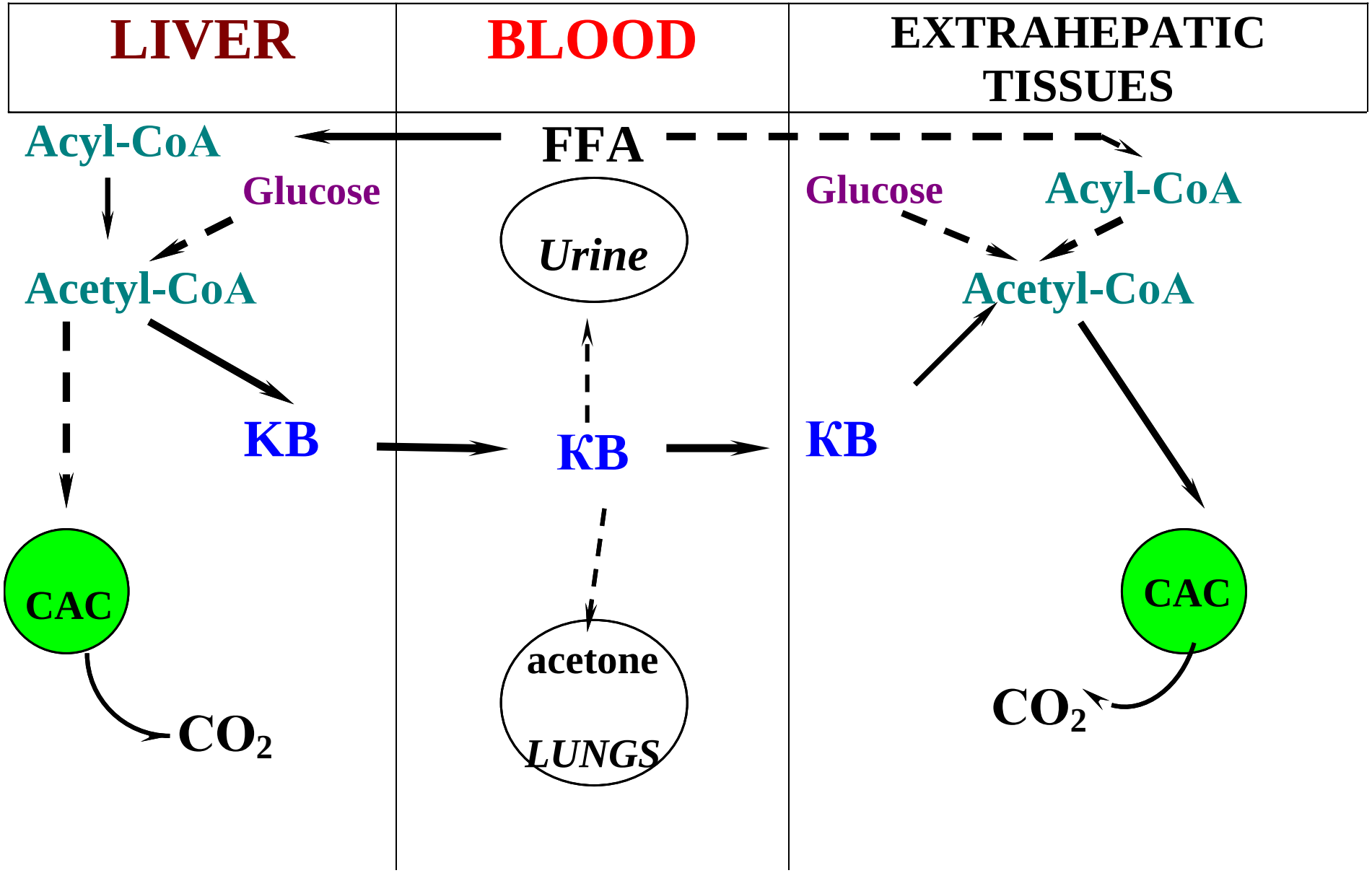


Ketone bodies can be regarded as a transport form of acetyl groups

Important sources of energy: heart muscle, renal cortex (preference to glucose, 1/3 of the energy)

brain - glucose is the major fuel but in starvation and diabetes brain uses acetoacetate




LIVER
BLOOD
EXTRAHEPATIC TISSUES
Acyl-CoA
FFA
Glucose
Urine
Glucose
Acyl-CoA
Acetyl-CoA
Acetyl-CoA
KB
KB
KB
CAC
CAC
acetone
LUNGS
CO_2
CO_2

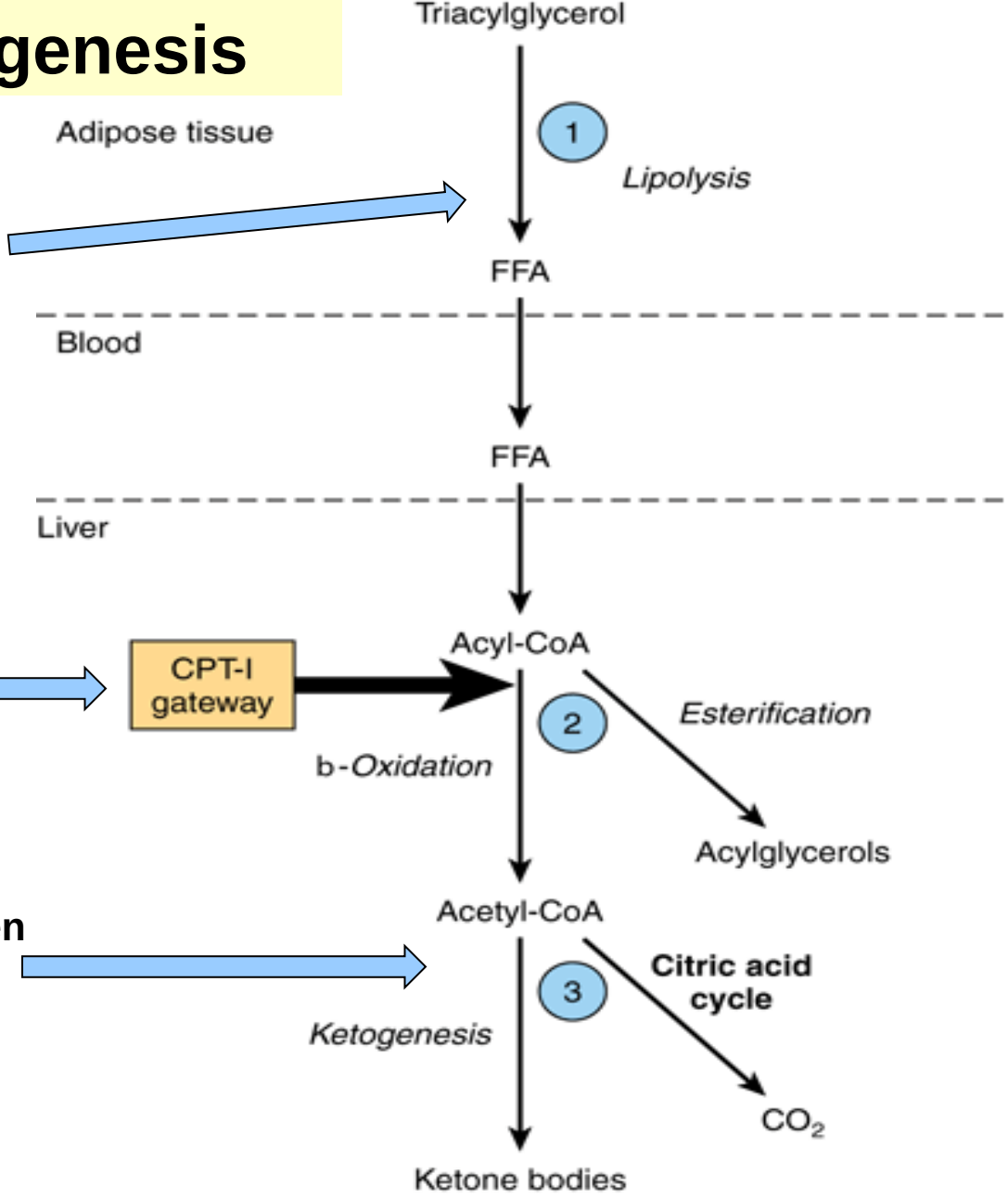
Regulation of ketogenesis

3 CRUCIAL STEPS

1. **Control of FFA mobilization from adipose tissue.**

2. **The activity of carnitine acyl transferase I in liver, which determines the proportion of FA flux that is oxidized.**

3. **Partition of acetyl-CoA between the pathway of ketogenesis and citric acid cycle.**



Source: Murray RK, Bender DA, Botham KM, Kennelly PJ, Rodwell VW, Weil PA: *Harper Illustrated Biochemistry, 29th Edition*: www.accessmedicine.com

Regulation of ketogenesis

	regulatory enzyme	*activation*	*inhibition*
1	**hormon sensitive lipase** *(lipolysis in fatty tissue)*	• ↑ ratio glucagon / insulin • catecholamines	• ↑ ratio insulin / glucagon
2	**carnitine acyltransferase I** *(transfer of fatty acids into mitochondria)*		• malonyl-Co A • ↑ ratio insulin / glucagon

3

The acetyl-CoA formed in β-oxidation is oxidized in the citric acid cycle, or it enters the pathway of ketogenesis to form ketone bodies.

As the level of serum FFA is raised, proportionately more FFA is converted to ketone bodies and less is oxidized via the citric acid cycle to CO_2.

A fall in the concentration of oxaloacetate, particularly within the mitochondria, can impair the ability of the citric acid cycle to metabolize acetyl-CoA and divert fatty acid oxidation toward ketogenesis.

Such a fall may occur because of an increase in the NADH/NAD^+ ratio caused by increased β-oxidation of fatty acids affecting the equilibrium between oxaloacetate and malate, leading to a decrease in the concentration of oxaloacetate, and when gluconeogenesis is elevated, which occurs when blood glucose levels are low.

The activation of pyruvate carboxylase, which catalyzes the conversion of pyruvate to oxaloacetate partially alleviates this problem, but in conditions such as starvation and untreated diabetes mellitus, ketone bodies are overproduced causing ketosis.

Concentration of Ketone Bodies

- **Concentration of total ketone bodies in the blood of well-fed individuals does not normally exceed**

  0.2 mmol/L (10-30 mg/L)

- ***Urine: loss via urine is usually less than***

  ***1 mg/day in* humans.**

Higher than normal quantities of ketone bodies present in the blood or urine constitute **ketonemia or ketonuria, respectively.**

The overall condition is called ketosis.

The basic form of ketosis occurs in **starvation** and involves depletion of available carbohydrate coupled with mobilization of FFA.

Ketosis occurs in **diabetes mellitus**

Nonpathologic forms of ketosis are found under conditions of high-fat feeding and after severe exercise in the postabsorptive state.

Acetoacetic and 3-hydroxybutyric acids are both moderately strong acids and are buffered when present in blood or other tissues.

However, their continual excretion in quantity progressively depletes the alkali reserve, causing

ketoacidosis.

This may be fatal in uncontrolled diabetes mellitus.

KETOSIS vs KETOACIDOSIS

THEY ARE NOT THE SAME!

KETOSIS

- **Low level** of ketones in the blood
- **Normal process** of the body
- **Safe function** of a low-carb, ketogenic diet

KETOACIDOSIS

- **Extremely high level** of ketones in blood
- **Can turn the blood acidic**, deadly if untreated
- **Occurs in diabetics** who don't take enough insulin or aren't well, people who are starving, or alcoholics

SYMPTOMS OF DIABETES KETOACIDOSIS (DKA)

high blood sugar levels and ketones in the urine

excessive thirst

urinating much more often and in larger amounts

sudden loss of weight

complaints of stomach pains or nausea

vomiting

signs of dehydration: dry mouth and tongue, sore throat, dark circles under the eyes

deep, heavy breathing

fruity-smelling breath

drowsiness leading in time to unconsciousness

FATTY ACID SYNTHESIS

There are few systems for fatty acid synthesis.

Extramitochondrial system: responsible for ***de novo*** synthesis of palmitic acid from acetyl-CoA.

Chain Elongation Systems:

Microsomal: present in microsomes which can lengthen existing fatty acid chains.

Mitochondrial: this system is mostly restricted to lengthening of an existing fatty acid of moderate chain-length. It operates under *anaerobiosis* and is favoured by a high $NADH/NAD^+$ ratio.

Extramitochondrial (Cytoplasmic) Synthesis of Fatty Acids: (*De Novo* Synthesis)

The synthesis takes place in cytosol.

Starting material is acetyl-CoA and synthesis always ends in formation of palmitic acid.

Materials Required for the Synthesis

Enzymes

– Acetyl-CoA carboxylase,

– Fatty acid synthase, a multienzyme complex

Coenzymes and cofactors:

Biotin, NADPH, Mn^{++}

CO_2

ATP

Sources of acetyl-CoA

- **Acetyl-CoA is mainly found in mitochondria and cannot pass out. It forms citrate by condensing with oxaloacetate. Citrate is transported out. Once in cytoplasm, an enzyme citrate lyase cleaves citrate to form acetyl-CoA and oxaloacetate.**
- **Carnitine-acetyl transferase may probably transfer acetyl group of acetyl-CoA to carnitine to form acetylcarnitine in mitochondria. After translocation to cytoplasm acetyl group may be transferred to CoA to make it acetyl-CoA.**

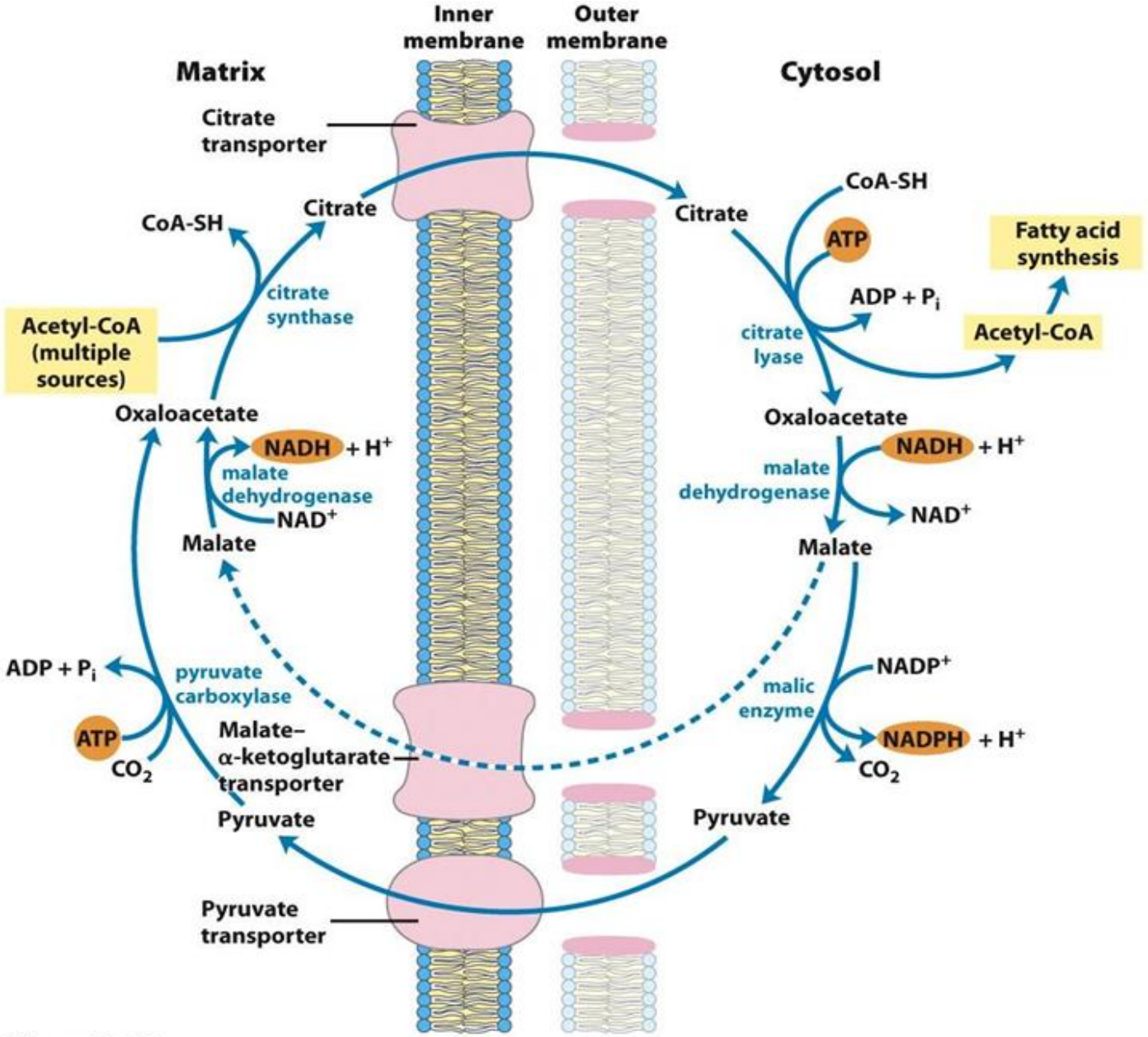


Figure 21-10

Lehninger Principles of Biochemistry, Fifth Edition

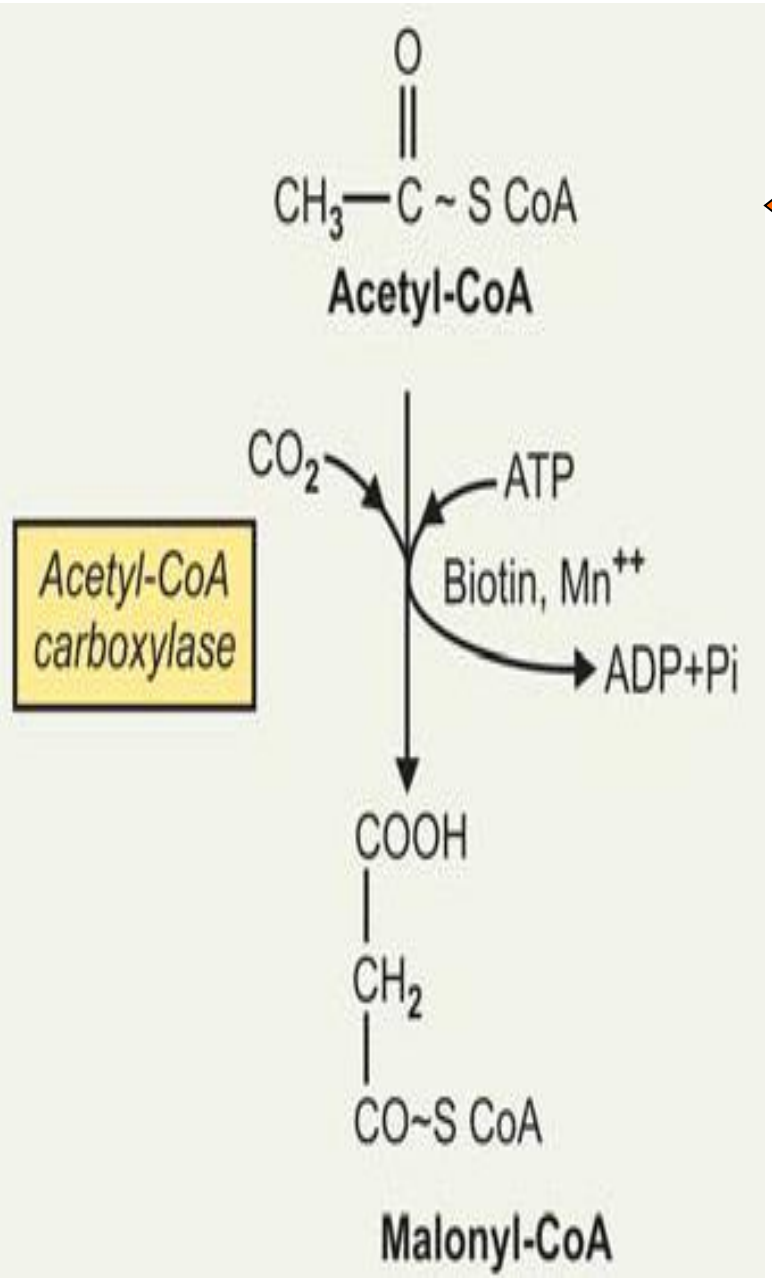
Sources of acetyl-CoA

- **Carnitine-acetyl transferase may probably transfer acetyl group of acetyl-CoA to carnitine to form acetylcarnitine in mitochondria. After translocation to cytoplasm acetyl group may be transferred to CoA to make it acetyl-CoA.**

Sources of NADPH

- **Pentose phosphate pathway is the main source of NADPH.**
- **Cytoplasmic enzyme called malic enzyme (NADP-malate dehydrogenase) catalazed the reaction in which malate is oxidatively decarboxylated to pyruvate and NADPH is produced.**
- **Cytoplasmic isocitrate dehydrogenase uses NADP as the coenzyme.**

Formation of malonyl CoA from acetyl-CoA



Reaction occurs in two steps:

Biotin-enzyme + ATP + HCO_3^-

↓

Carboxy-biotin-enzyme+ADP+P_i

Carboxy-biotin-enzyme + Acetyl-CoA

↓

Malonyl CoA + biotin-enzyme

The reaction is ***irreversible.***

Regulation

Acetyl-CoA carboxylase catalyses the rate limiting step in the *de novo* synthesis of fatty acids andprovides the earliest point at which control can be exerted.

The enzyme is inactivated by phosphorylation.

A: Insulin, CoA, Guanine nucleotides

I: Glucagon, adrenaline, acyl-CoA
Decrease in citrate concentration decreases acetyl-CoA carboxylase activity

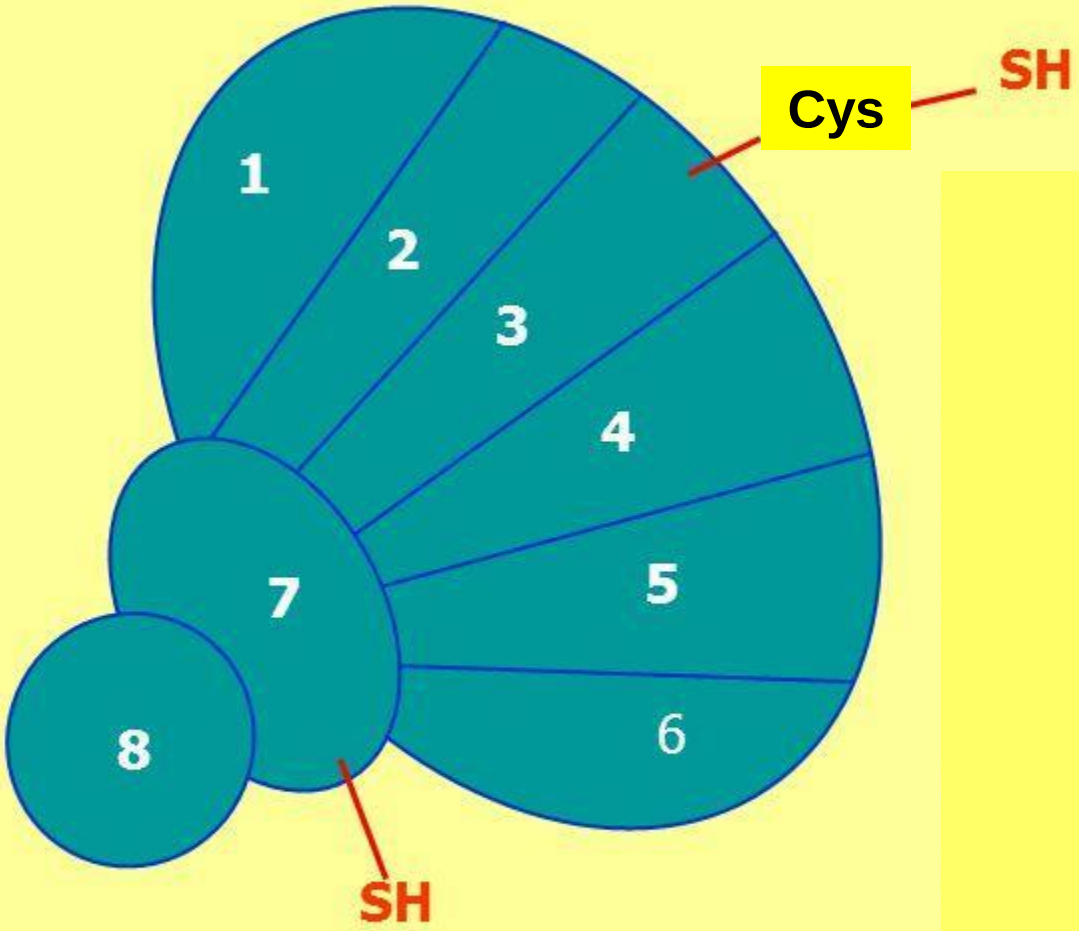
Fatty acid synthase

It is a **multienzyme complex.**

It is made up of an ellipsoid dimer of two identical polypeptide monomeric units,

arranged in a “head to tail” fashion

Structure of Fatty Acid Synthase



7:

Acyl Carrier Protein (ACP)

Fatty acid synthase

The ACP has an –SH group in the 4-phosphopantothene moiety, referred as ***pantothenyl-SH (Pan-SH)***

Another active –SH group present in the cysteine moiety of the enzyme ***ketoacyl synthase***, referred as ***cysteinyl-SH (Cys–SH).***

The “Pan-SH” of one monomeric unit is in close

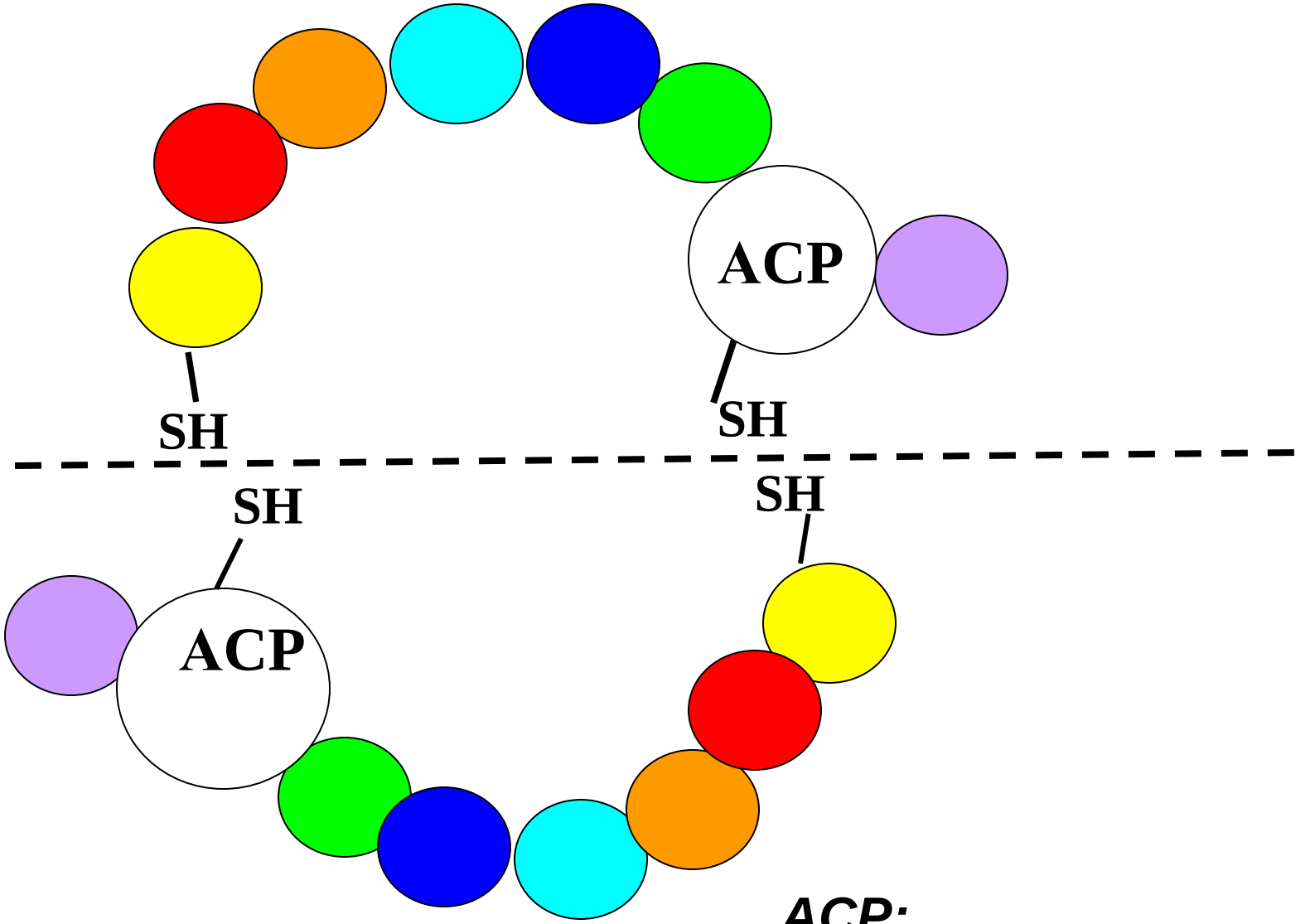
proximity to the “Cys-SH” group of other monomeric unit and vice-versa.

Sequence of domaines in primary structure of fatty acid syhthase monomer

- ***ketoacyl synthase,***
- ***malonyl/acetyl transacylase,***
- ***hydratase***
- ***enoyl reductase,***
- ***ketoacyl reductase,***
- ***ACP***
- ***thio-esterase (deacylase)***

Complex is functional only when the two monomeric units are in association with each other. The functional activity is lost when they are dissociated. In a dimer form, the complex jointly synthesises 2 molecules of palmitic acid simultaneously.

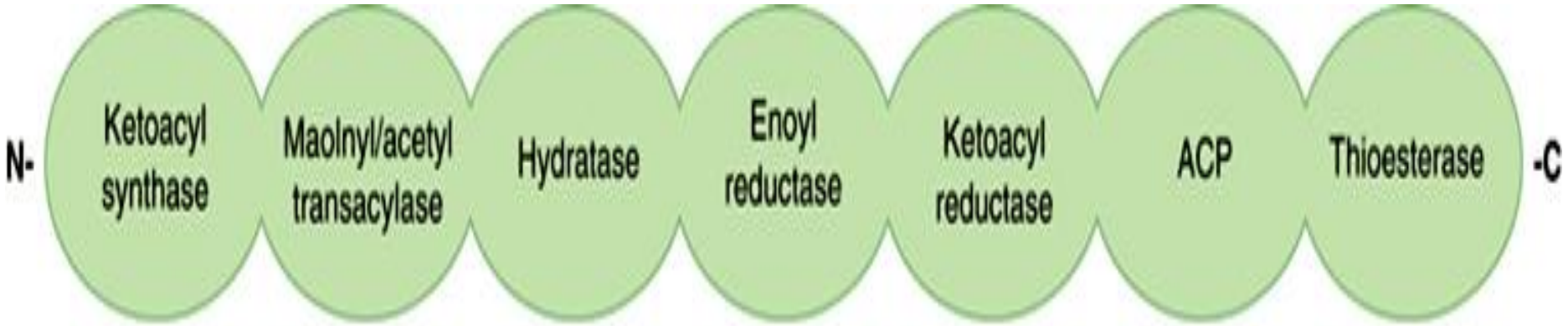
Fatty acid synthase

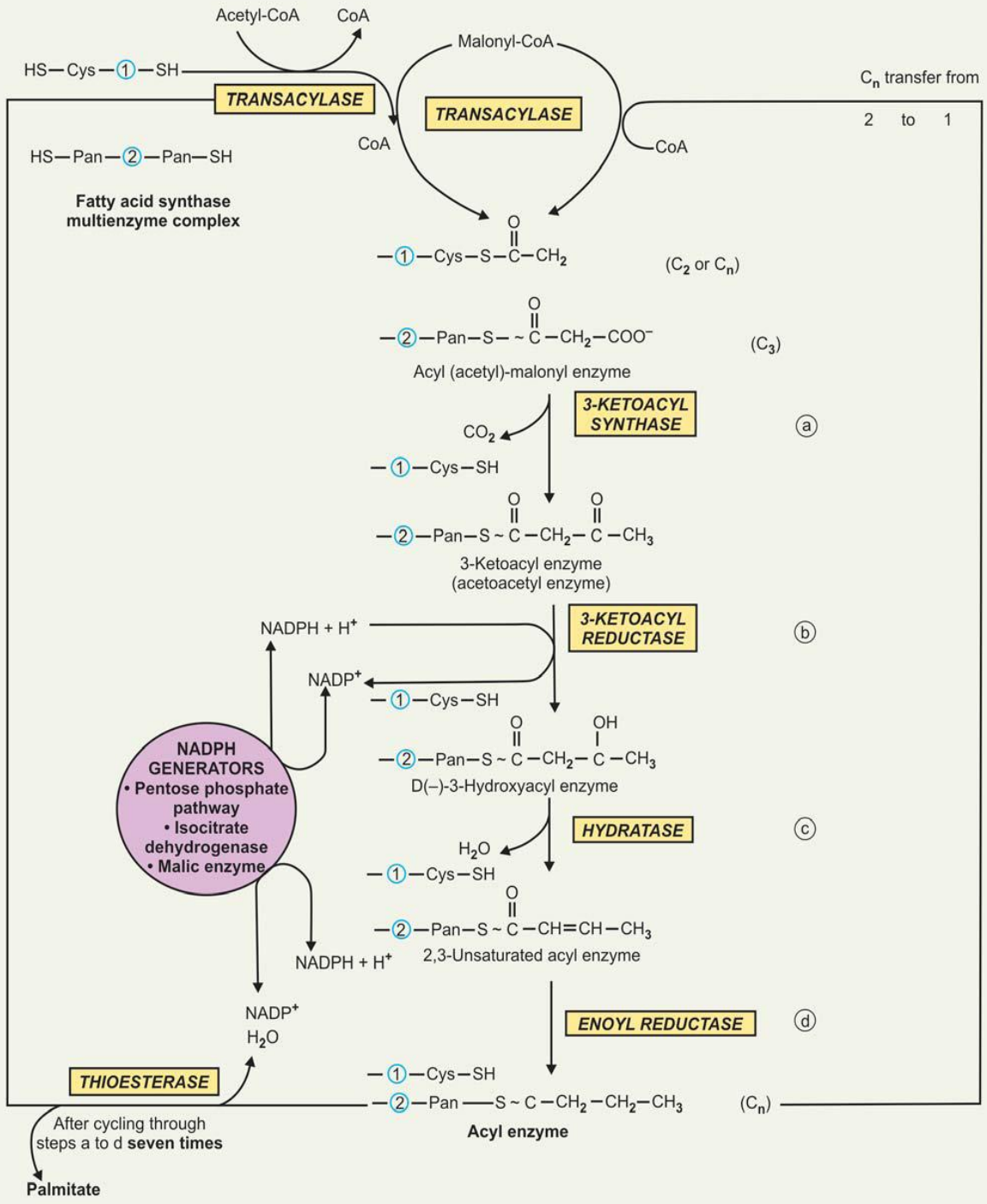


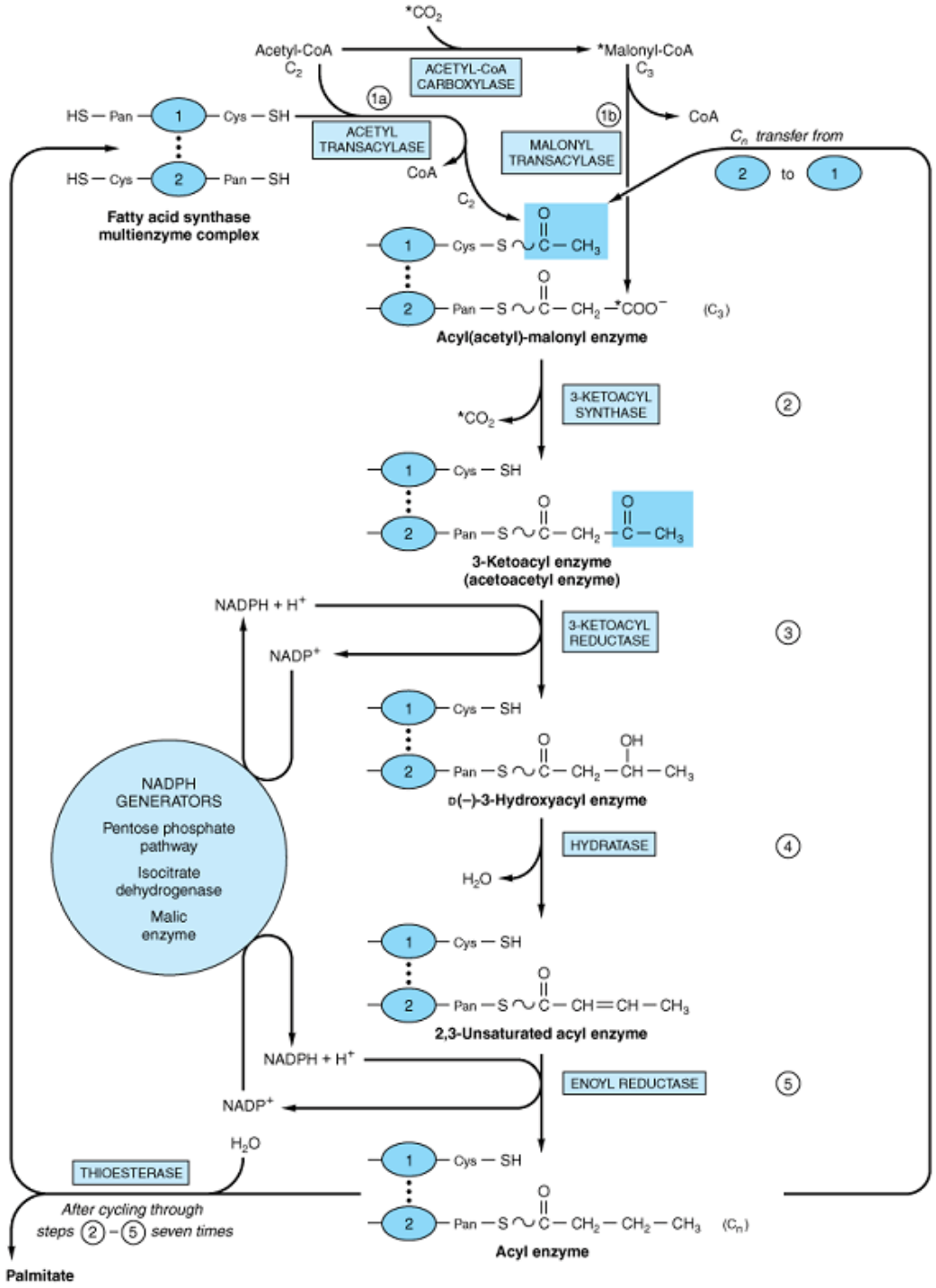
ACP:

Acyl Carrier Protein.

Sequence of enzyme domains in primary structure of fatty acid synthase monomer




Acetyl-CoA
CoA
Malonyl-CoA
HS—Cys—①—SH
TRANSACYLASE
TRANSACYLASE
C_n transfer from
2 to 1
CoA
CoA
HS—Pan—②—Pan—SH
Fatty acid synthase multienzyme complex
—①—Cys—S—C(=O)—CH_2
(C_2 or C_n)
—②—Pan—S—~C(=O)—CH_2—COO^-
(C_3)
Acyl (acetyl)-malonyl enzyme
3-KETOACYL SYNTHASE
a
CO_2
—①—Cys—SH
—②—Pan—S~C(=O)—CH_2—C(=O)—CH_3
3-Ketoacyl enzyme (acetoacetyl enzyme)
3-KETOACYL REDUCTASE
b
$NADPH + H^+$
$NADP^+$
—①—Cys—SH
—②—Pan—S~C(=O)—CH_2—C(OH)—CH_3
D(–)-3-Hydroxyacyl enzyme
NADPH GENERATORS
• Pentose phosphate pathway
• Isocitrate dehydrogenase
• Malic enzyme
HYDRATASE
c
H_2O
—①—Cys—SH
—②—Pan—S~C(=O)—CH=CH—CH_3
2,3-Unsaturated acyl enzyme
$NADPH + H^+$
$NADP^+$
H_2O
ENOYL REDUCTASE
d
THIOESTERASE
—①—Cys—SH
—②—Pan—S~C—CH_2—CH_2—CH_3
(C_n)
Acyl enzyme
After cycling through steps a to d seven times
Palmitate


*CO2
Acetyl-CoA
C2
*Malonyl-CoA
C3
ACETYL-CoA CARBOXYLASE
1a
1b
CoA
ACETYL TRANSACYLASE
MALONYL TRANSACYLASE
HS — Pan — 1 — Cys — SH
HS — Cys — 2 — Pan — SH
Fatty acid synthase multienzyme complex
Cn transfer from 2 to 1
C2
1 — Cys — S — C — CH3
2 — Pan — S — C — CH2 — *COO− (C3)
Acyl(acetyl)-malonyl enzyme
*CO2
3-KETOACYL SYNTHASE
2
1 — Cys — SH
2 — Pan — S — C — CH2 — C — CH3
3-Ketoacyl enzyme (acetoacetyl enzyme)
NADPH + H+
NADP+
3-KETOACYL REDUCTASE
3
1 — Cys — SH
2 — Pan — S — C — CH2 — CH(OH) — CH3
D(−)-3-Hydroxyacyl enzyme
NADPH GENERATORS
Pentose phosphate pathway
Isocitrate dehydrogenase
Malic enzyme
H2O
HYDRATASE
4
1 — Cys — SH
2 — Pan — S — C — CH=CH — CH3
2,3-Unsaturated acyl enzyme
NADPH + H+
NADP+
ENOYL REDUCTASE
5
H2O
THIOESTERASE
After cycling through steps 2 – 5 seven times
1 — Cys — SH
2 — Pan — S — C — CH2 — CH2 — CH3 (Cn)
Acyl enzyme
Palmitate

Biosynthesis of Triacylglycerols

Glucose

Dihydroxyacetone phosphate

Glycerol

NADH

NAD^+

ATP

ADP

Glycerol-3-phosphate

Acyl-CoA

Acyl-CoA

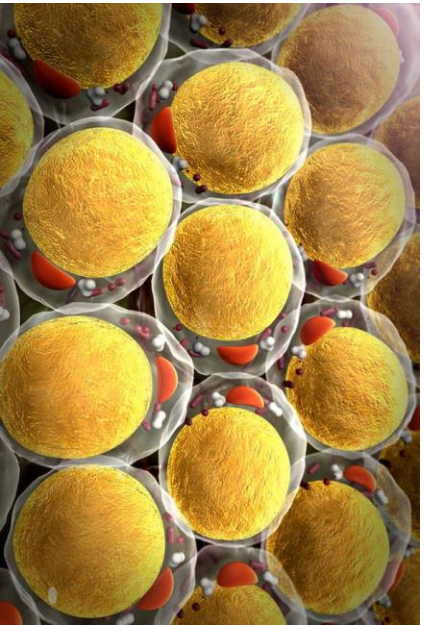
Phosphatidate

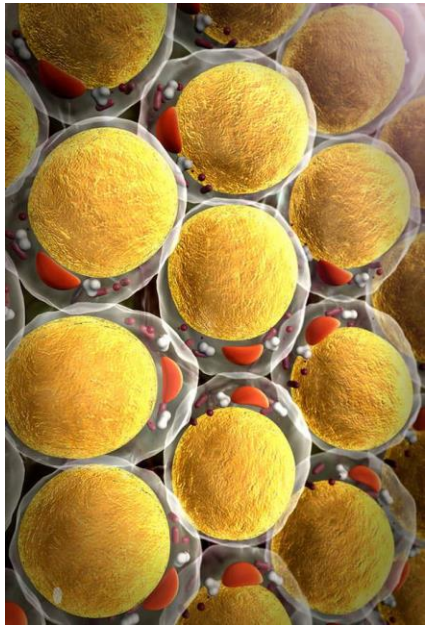
P_i

Diacylglycerol

Acyl-CoA

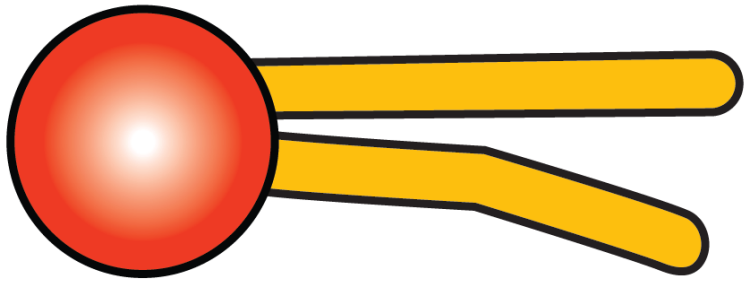
Triacylglycerol





Phospholipids

- Phospholipids are a specialized group of lipids performing a variety of functions.
- These include the membrane structure & functions, involvement in blood clotting & supply of arachidonic acid for the synthesis of prostaglandins.



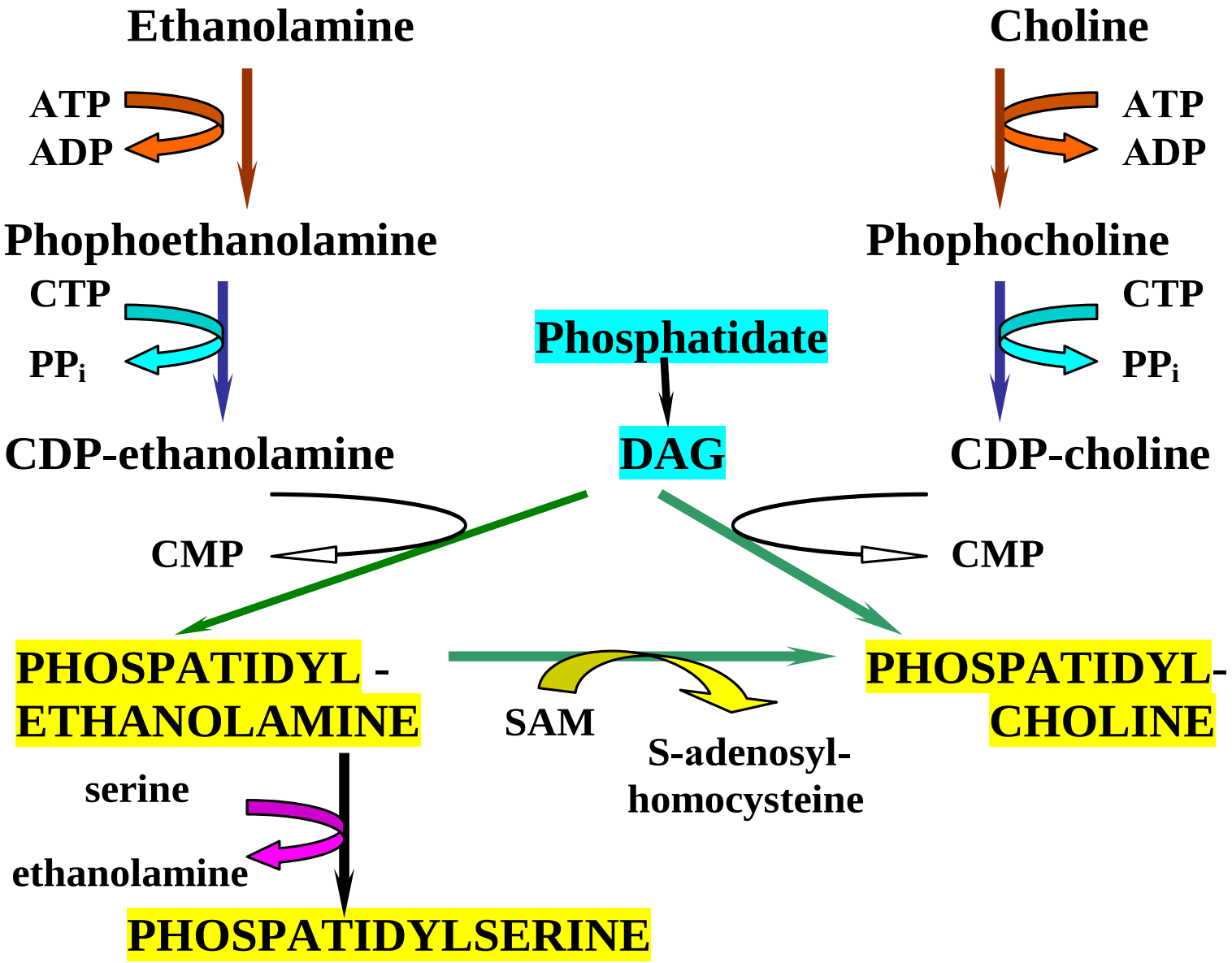
Synthesis of phospholipids

- Phospholipids are synthesized from phosphatidic acid & 1,2-diacylglycerol, intermediates in the production of triacylglycerols.
- Phospholipids synthesis occurs in the smooth endoplasmic reticulum.
- Inner mitochondrial membrane

Formation of lecithin & cephalin

- It occurs mainly in liver & brain.
- Choline & ethanolamine first get phosphorylated & then combine with CTP to form, CDP-choline & CDP-ethanolamine.
- Phosphatidylcholine (lecithin) is synthesized when CDP-choline combines with 1,2-diacylglycerol.

Biosynthesis of phospholipids



Synthesis of phosphatidylserine

- Phosphatidyl ethanolamine can exchange its ethanolamine group with free serine to produce phosphatidylserine.

Formation of phosphatidylinositol

- CDP-diacylglycerol produced from phosphatidic acid combines with inositol to form phosphatidyl inositol (PI).
- Phosphatidyl inositol contains arachidonic acid on carbon 2 of glycerol which serves as, a substrate for prostaglandin synthesis.
- PI is important for signal transmission across membranes.

PHOSPHOLIPID BIOSYNTHESIS

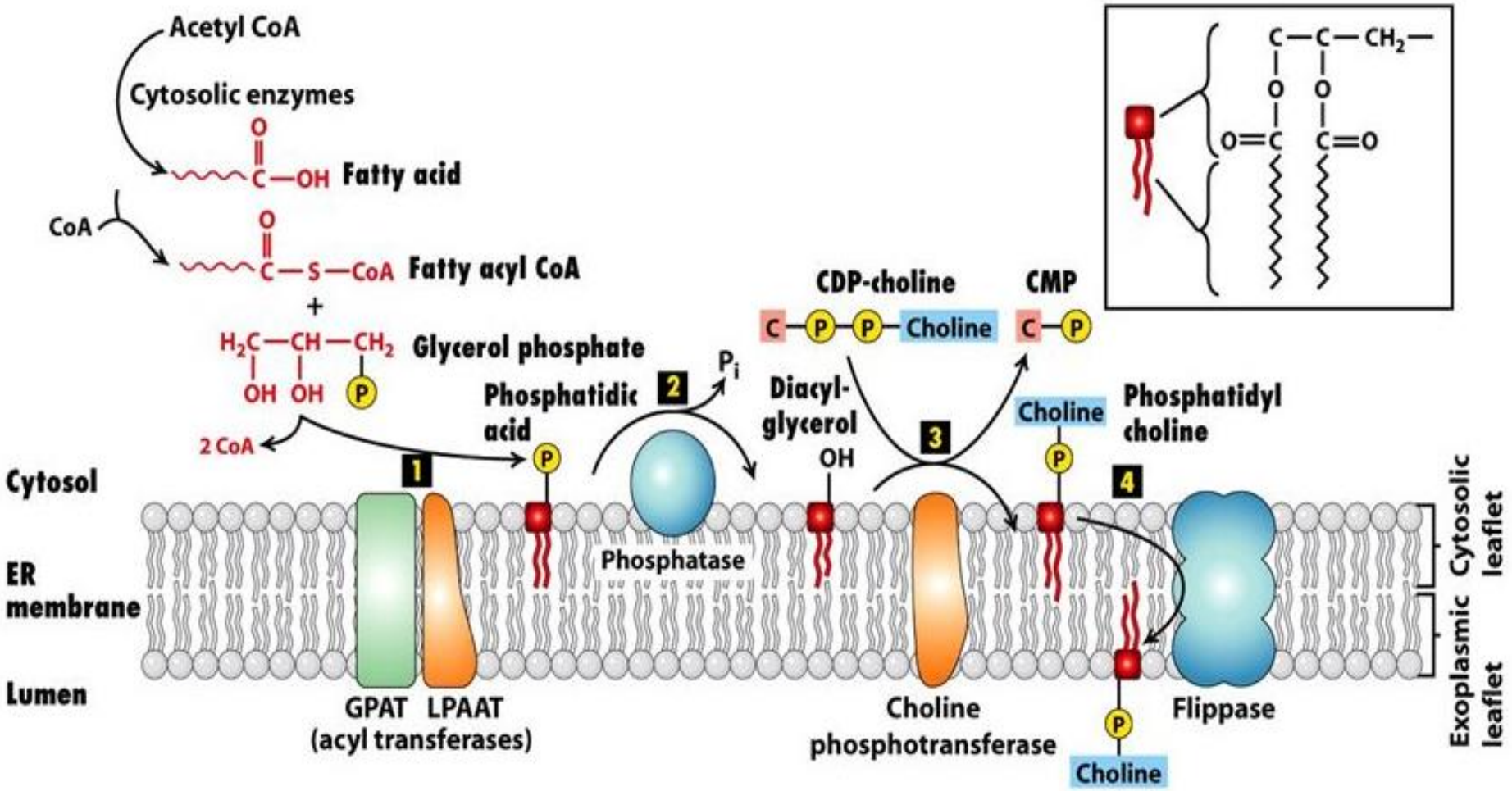


Figure 10-25
Molecular Cell Biology, Sixth Edition

The synthesis of phospholipids requires

- **glycerol**
- **fatty acids**
- **inorganic phosphates**
- **nitrogen bases (in particular, choline for synthesis of phosphatidylcholine)**

Fatty Infiltration of Liver

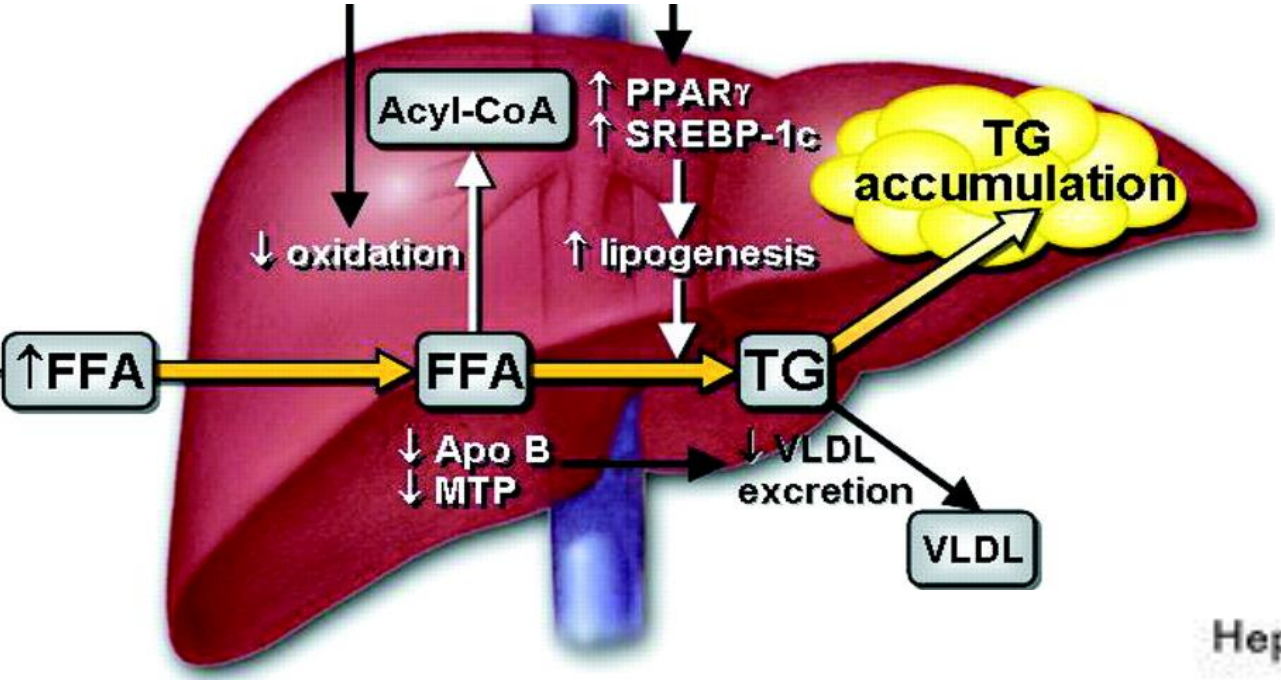
In insufficient synthesis of choline, or its short supply to the liver, the synthesis of phospholipids from neutral fat components becomes either impossible, or drastically decreased, which results in deposition of neutral fats in the liver.

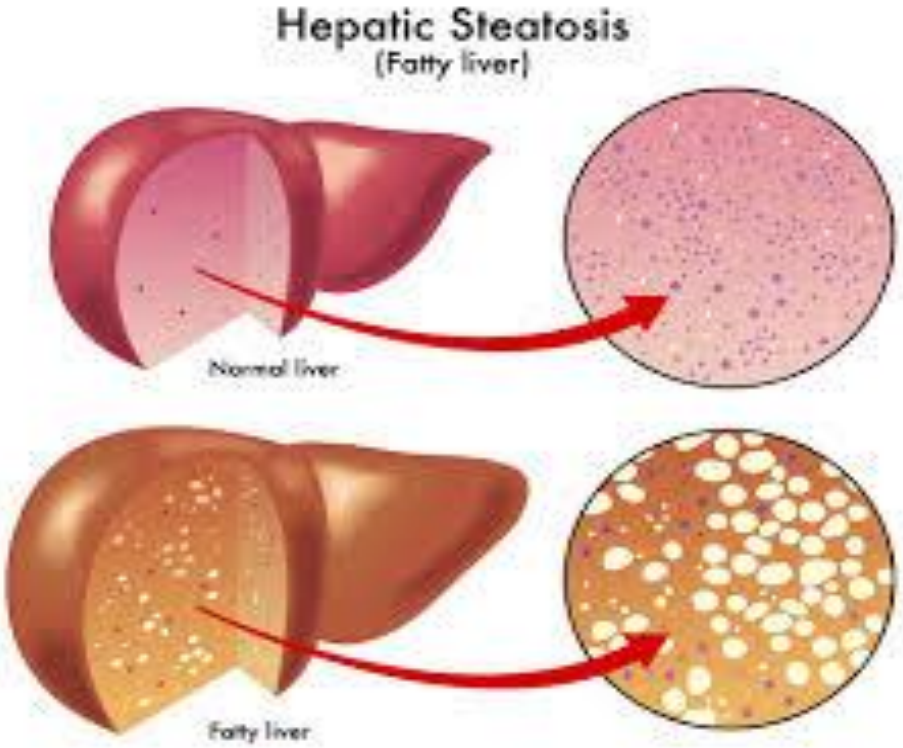
Such condition is referred to as

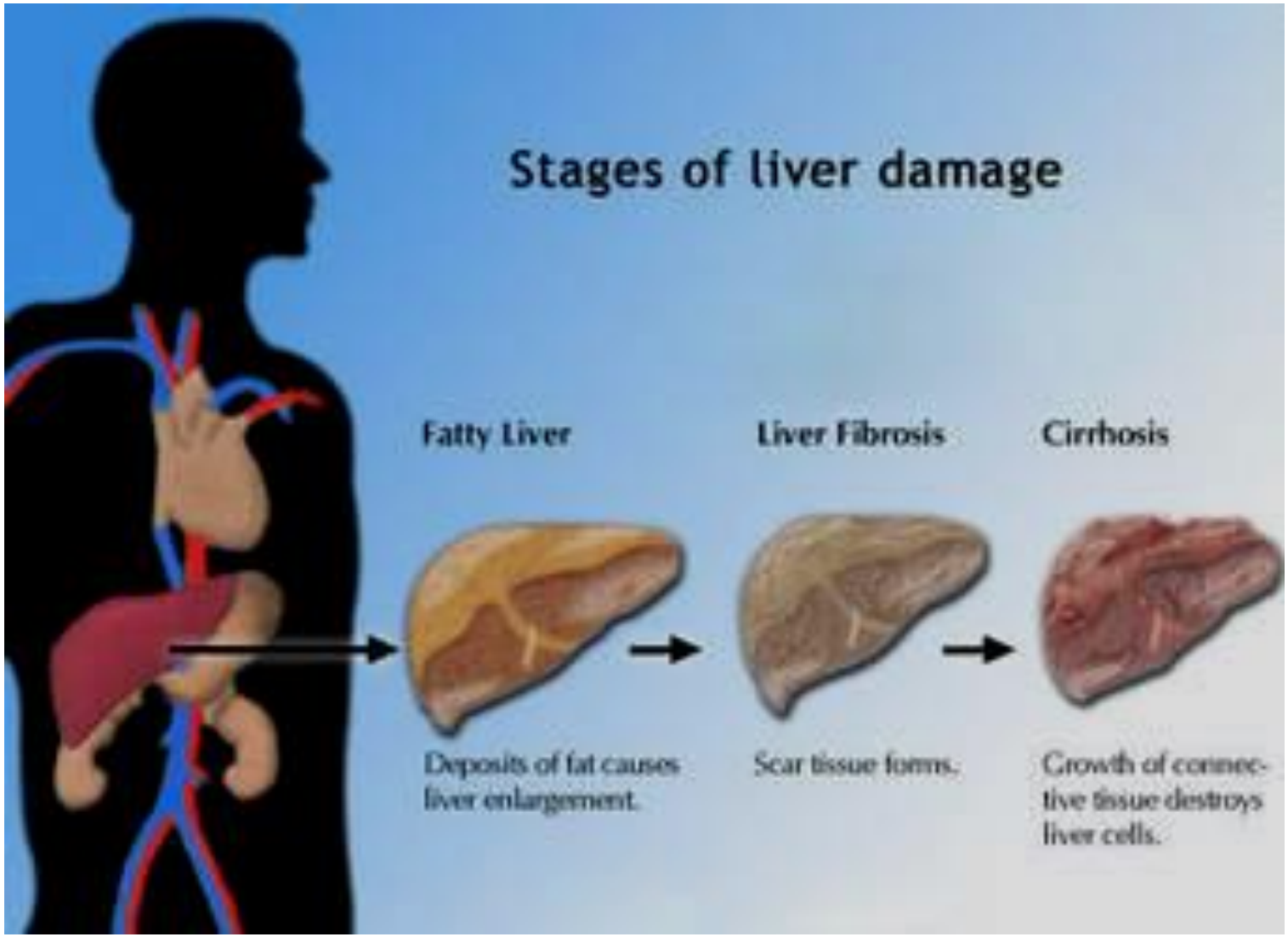
***fatty infiltration of liver*,**

which may subsequently develop into

a fatty degeneration of the liver (steatosis)


Acyl-CoA
↑ PPARγ
↑ SREBP-1c
TG accumulation
↓ oxidation
↑ lipogenesis
↑FFA
FFA
TG
↓ Apo B
↓ MTP
↓ VLDL excretion
VLDL


Hepatic Steatosis
(Fatty liver)
Normal liver
Fatty liver


Stages of liver damage
Fatty Liver
Liver Fibrosis
Cirrhosis
Deposits of fat causes liver enlargement.
Scar tissue forms.
Growth of connective tissue destroys liver cells.

In other words, the synthesis of phospholipids needs either choline, or compounds capable to act as methyl group donors and thus participate in the production of choline (for example, methionine)

Such compounds are known as

lipotropic agents

curd cheese is recommended in the diet as lipotropic agent, since its ingredient is casein, a protein whose molecule contains a large number of methionine residues