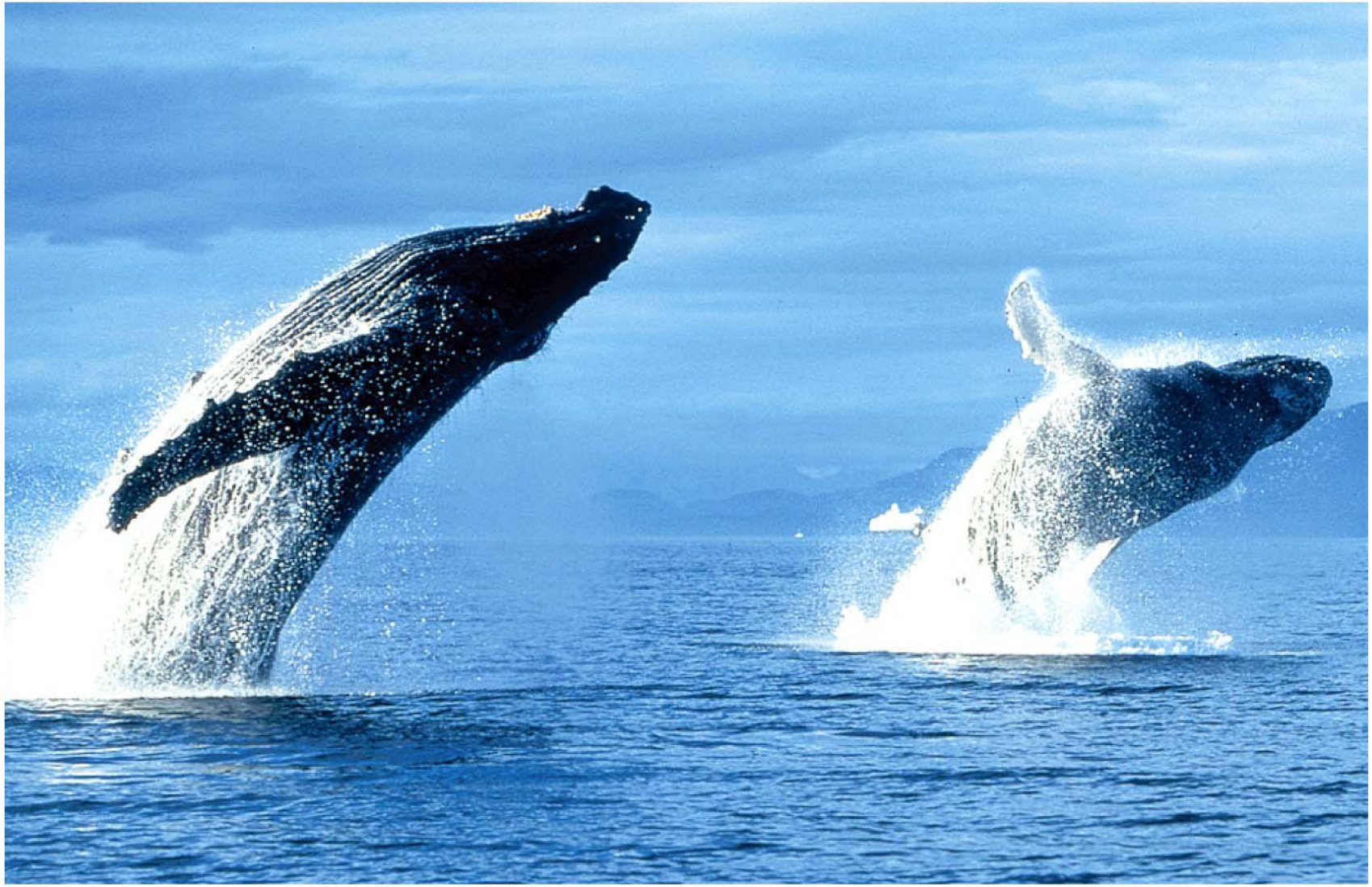
Lipolysis, β-oxidation & ketone body formation



Craig Wheelock

Questions? Comments?

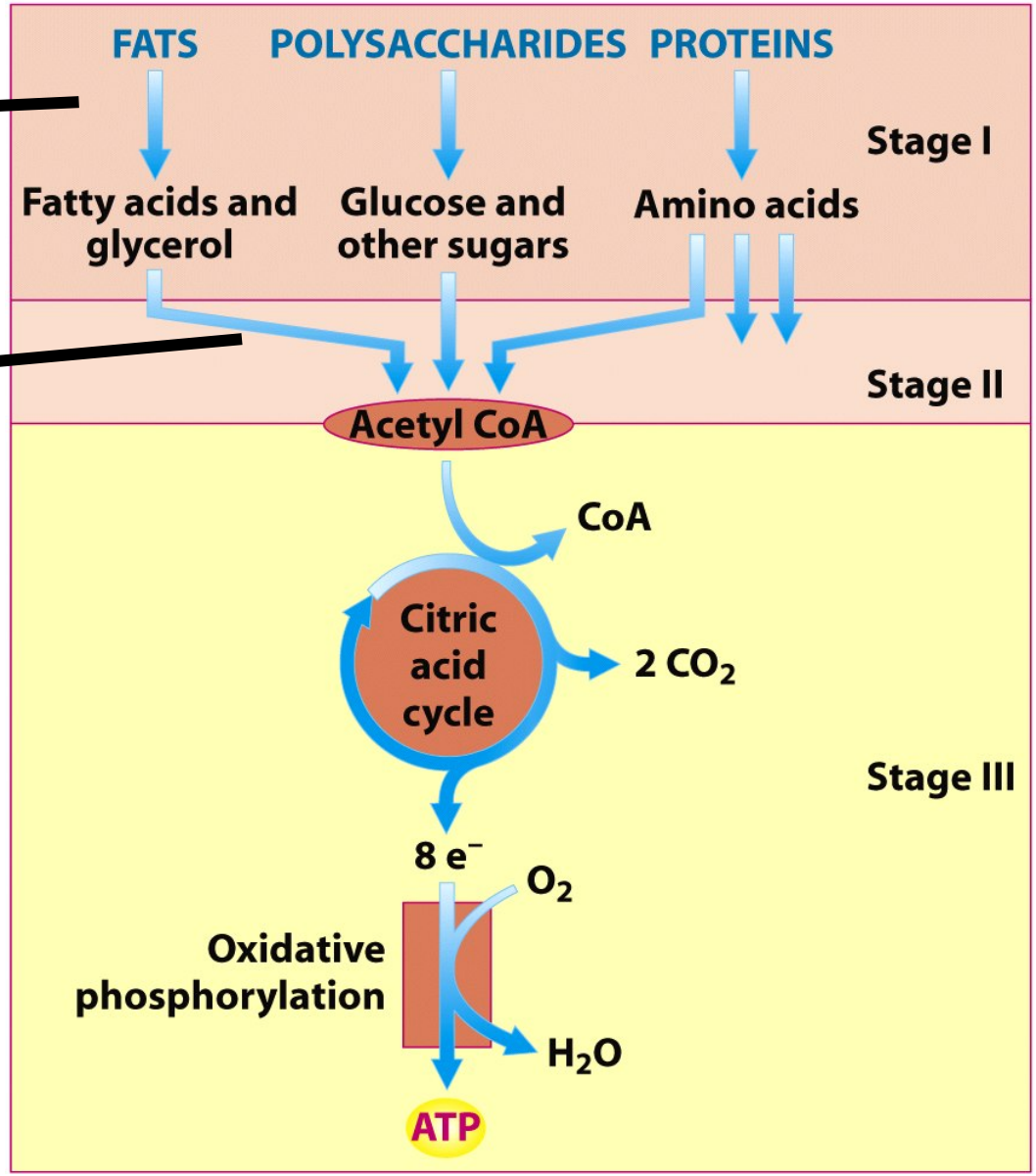
February 1st, 2008

craig.wheelock@ki.se

Catabolism's 3 stages

Lipolysis

β-oxidation



Catabolism is the set of metabolic pathways that break down molecules into smaller units & release energy

Catabolism's 3 stages

- **Stage 1 – food is broken down into smaller units - digestion**
- **Stage 2 – these molecules are degraded to simple units that play a central role in metabolism**
- **Stage 3 – ATP is produced from the complete oxidation of the acetyl unit of acetyl CoA**

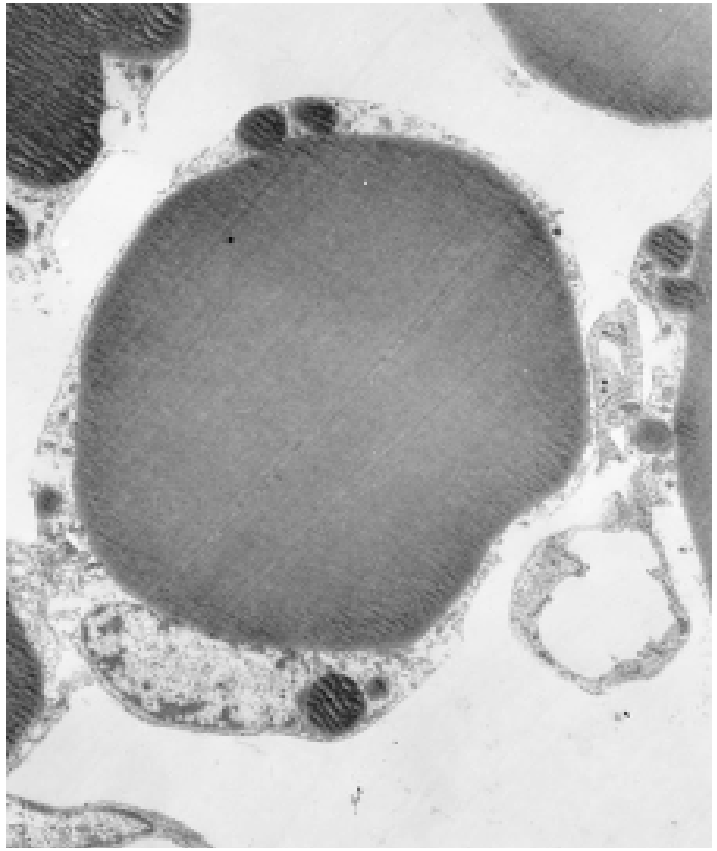
Lipids to be degraded come from 2 sources:

a) **Food**

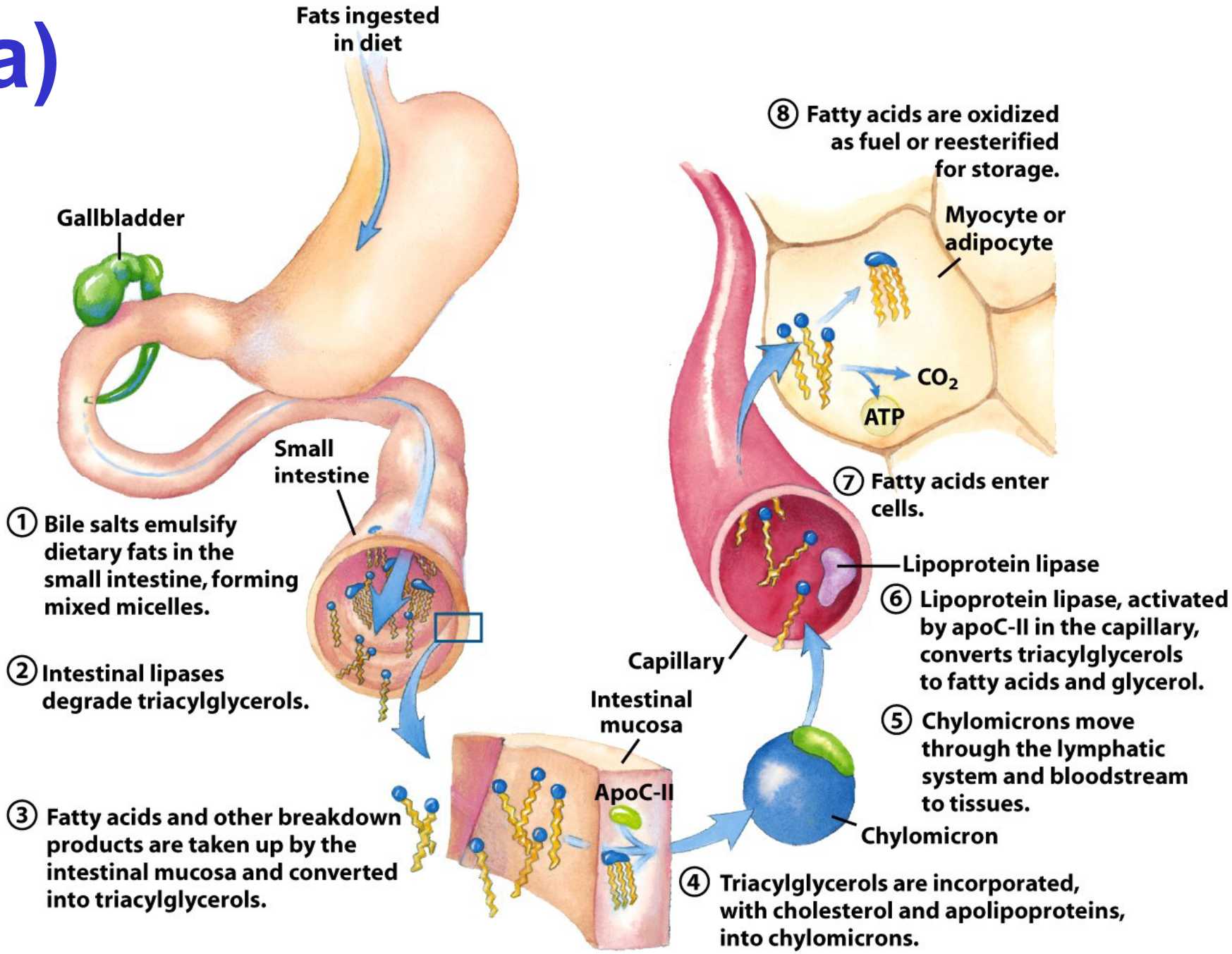
- adults eat 60-150 g/day
- >90% TAG

the rest:
cholesterol,
cholesterol esters, PL, FA.....

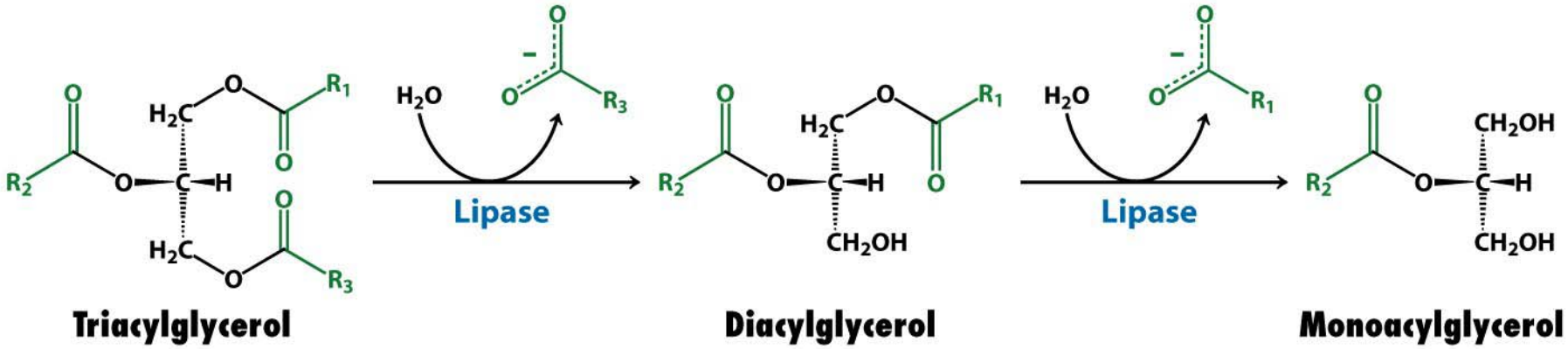
b) **Fat deposits**
(adipocytes)



a)


Fats ingested in diet
Gallbladder
Small intestine
① Bile salts emulsify dietary fats in the small intestine, forming mixed micelles.
② Intestinal lipases degrade triacylglycerols.
③ Fatty acids and other breakdown products are taken up by the intestinal mucosa and converted into triacylglycerols.
Intestinal mucosa
ApoC-II
④ Triacylglycerols are incorporated, with cholesterol and apolipoproteins, into chylomicrons.
⑤ Chylomicrons move through the lymphatic system and bloodstream to tissues.
Chylomicron
Capillary
⑥ Lipoprotein lipase, activated by apoC-II in the capillary, converts triacylglycerols to fatty acids and glycerol.
Lipoprotein lipase
⑦ Fatty acids enter cells.
⑧ Fatty acids are oxidized as fuel or reesterified for storage.
Myocyte or adipocyte
CO_2
ATP

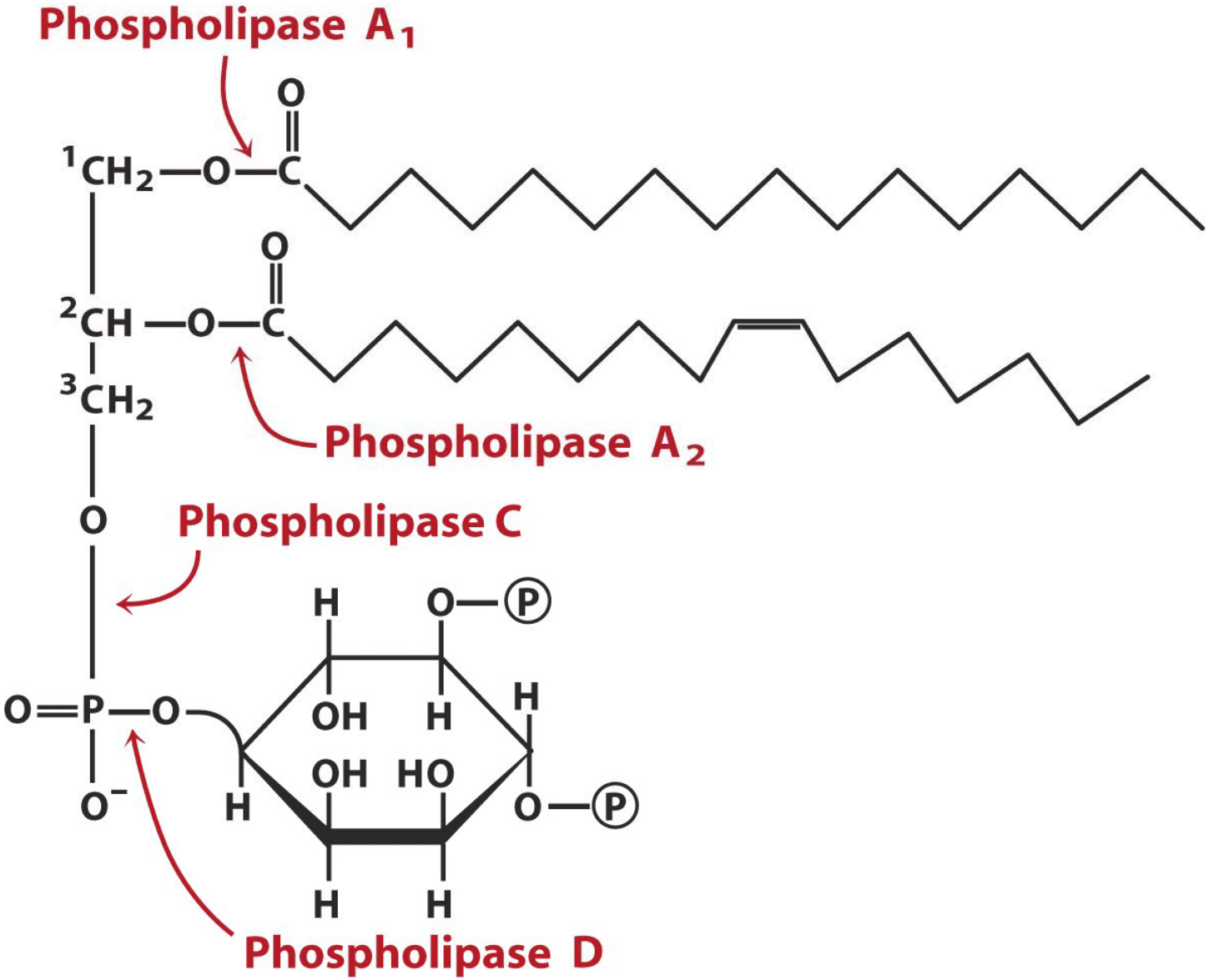
Pancreas lipases break down TAG to FFA and 2-MAG



Cholesterol esters are hydrolyzed to cholesterol and FFAs

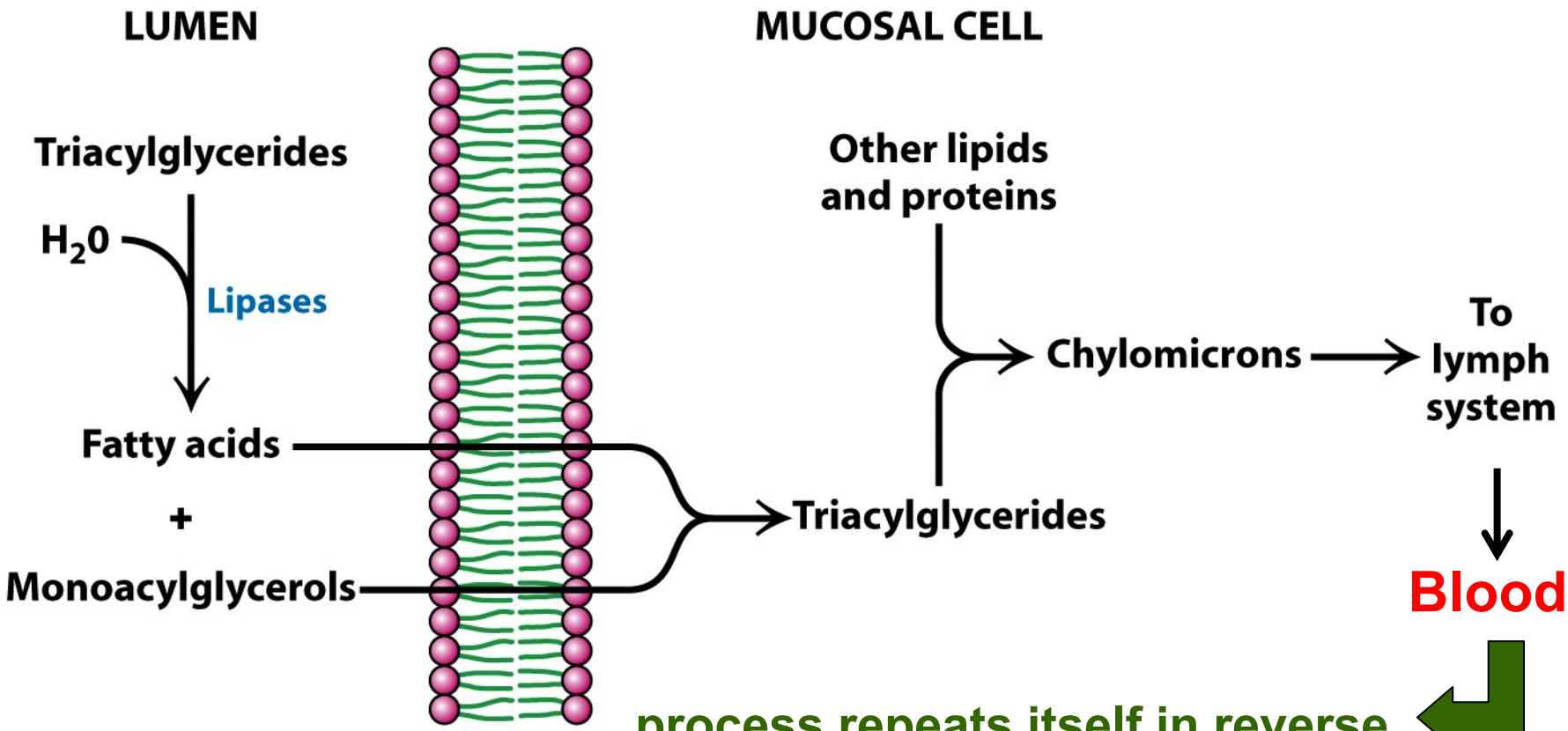
FAs are cleaved from PLs

Phospholipases



Dietary lipids are transported in chylomicrons

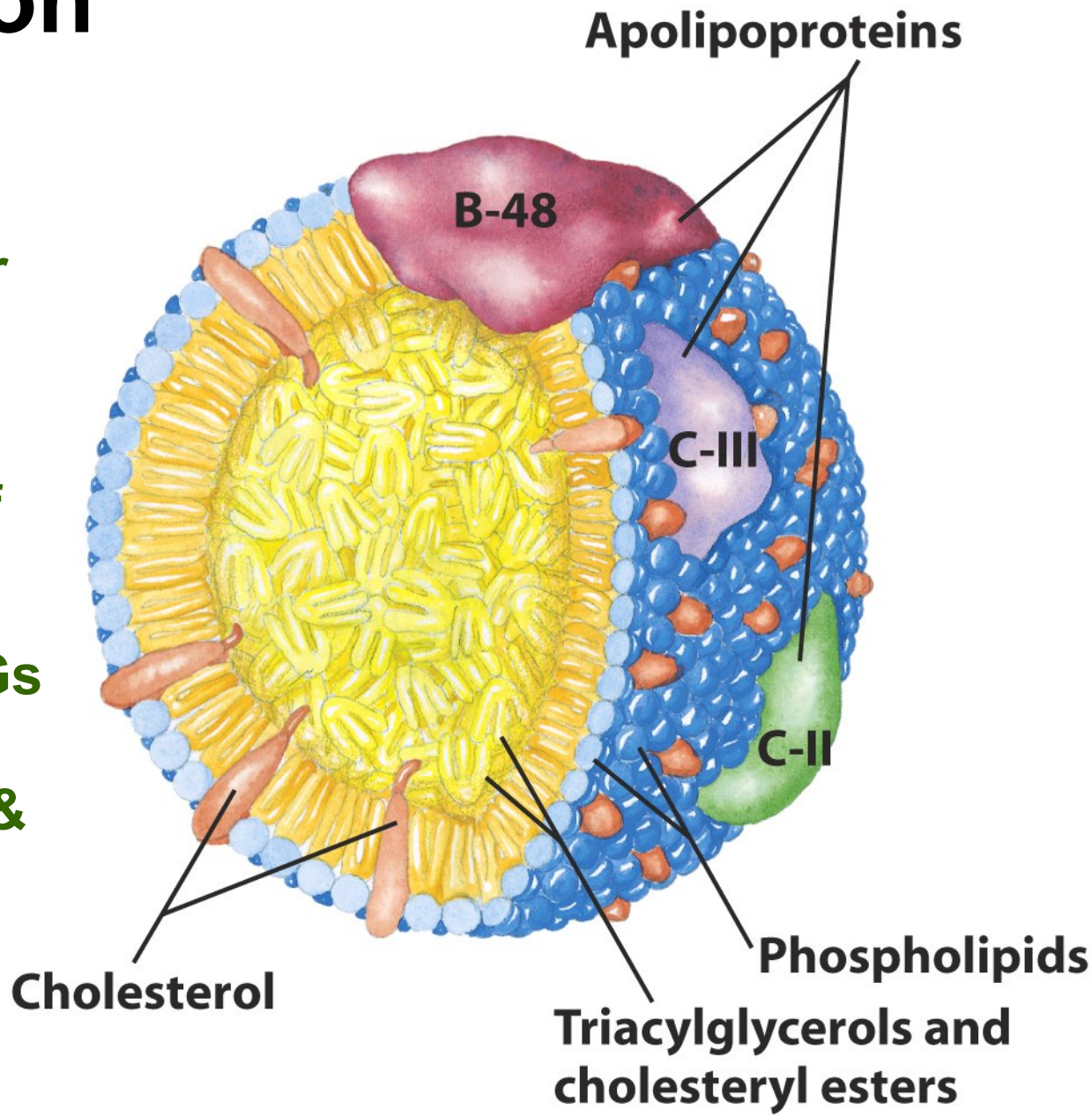
Lipoprotein transport particles (TAGs and apoliprotein B-48)



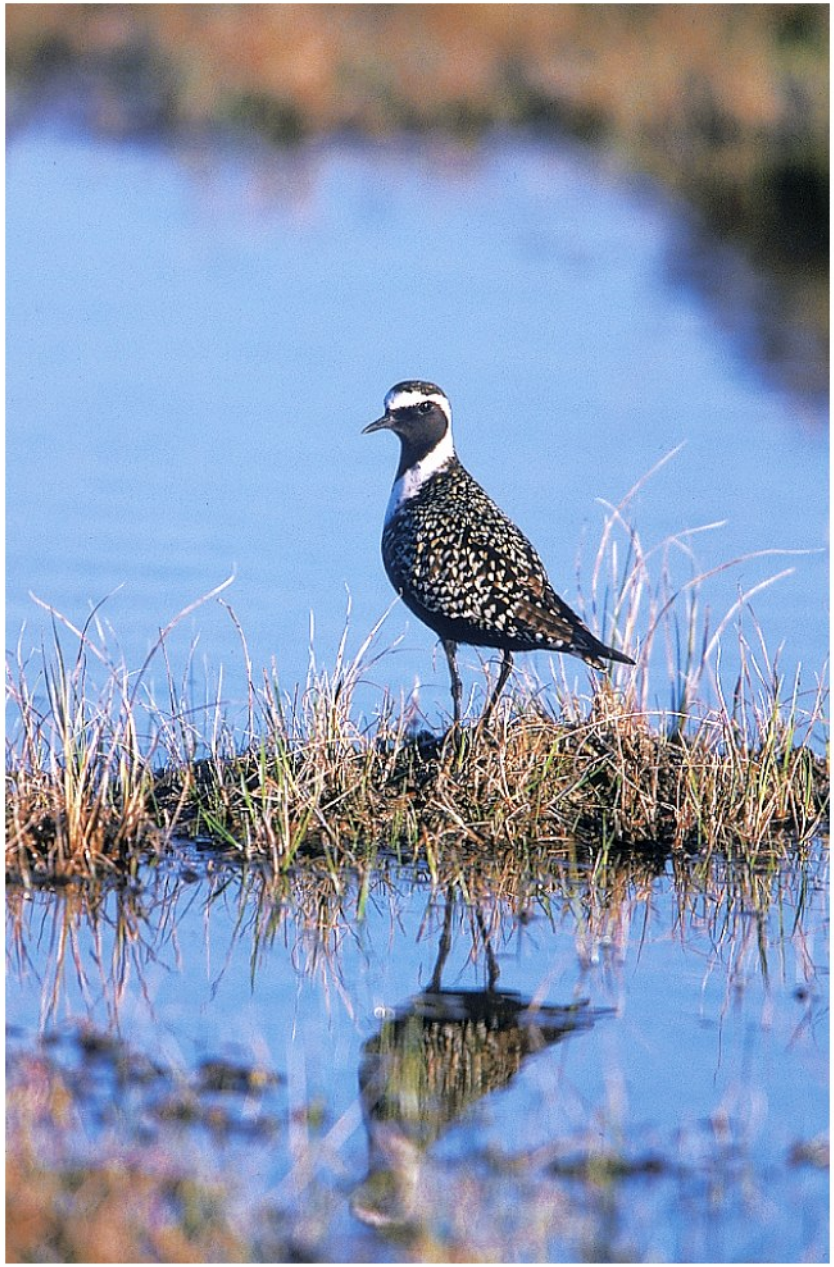
process repeats itself in reverse
Membrane-bound lipases
degrade TGs to FFAs and MAGs
for transport into tissue . . . .

Chylomicron

- large lipoprotein particles (diameter of 75 to 1,200 nm)
- created by the absorptive cells of the small intestine
- composed of TAGs (85%) & contain some cholesterol & cholesteryl esters

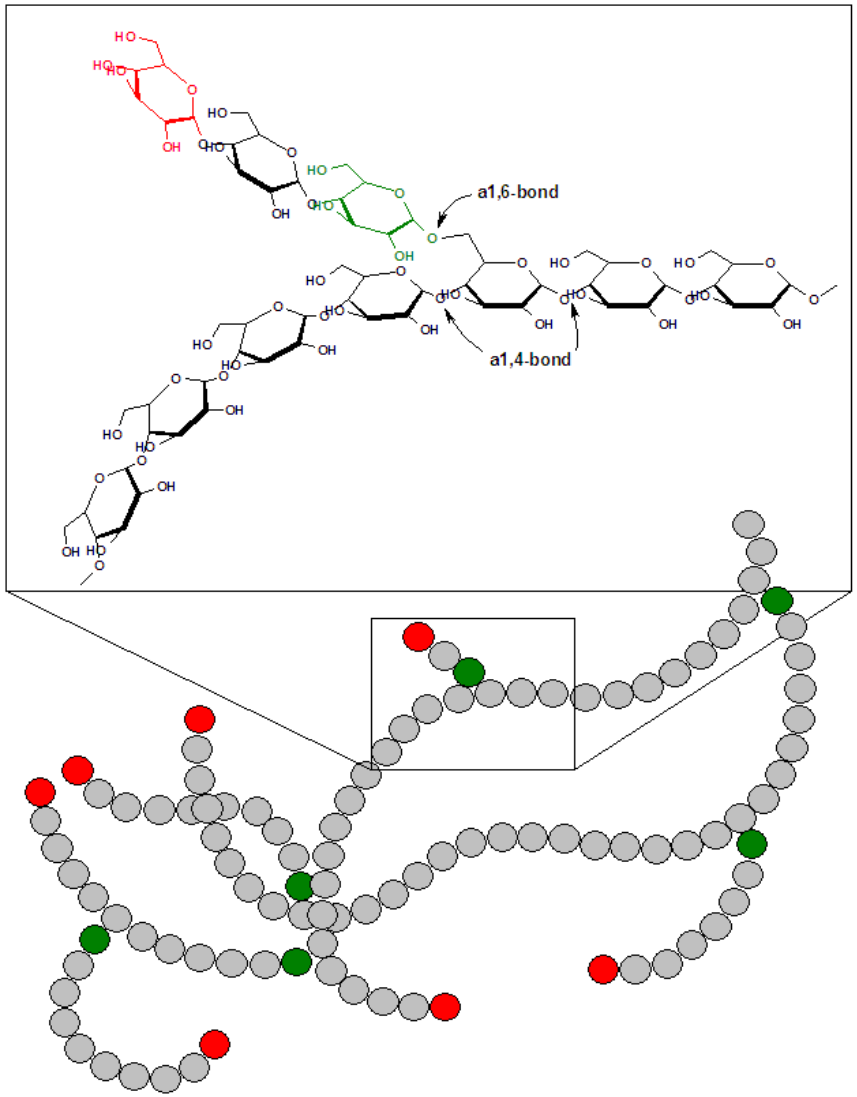


b) Stored fat as an energy source



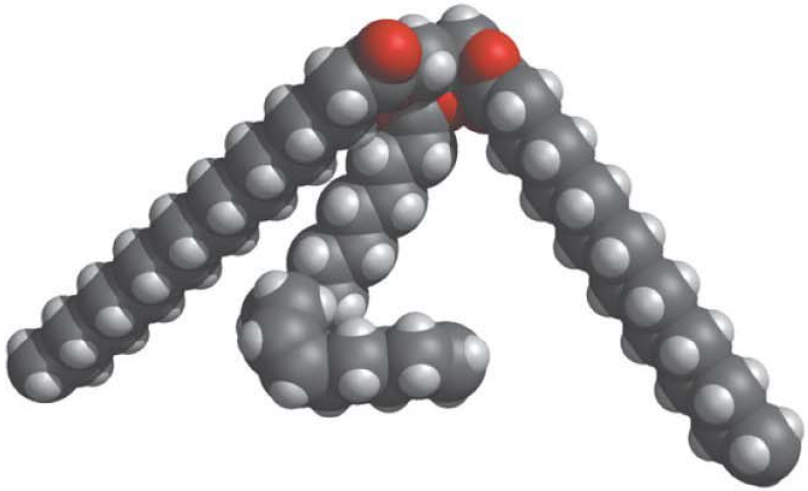
Energy depots

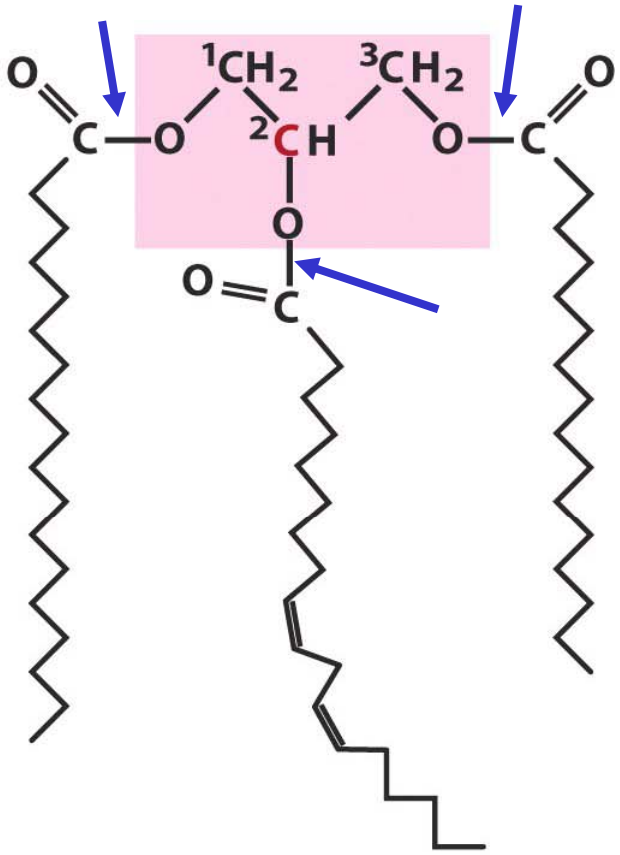
- Glycogen in muscle
  - 1200 kcal
- Glycogen in liver
  - 400 kcal
- Triacylglycerols in fat
  - 135,000 kcal
- Proteins (mainly muscle)
  - 24,000 kcal



Mobilization of stored fat:

a hormone sensitive lipase hydrolyzes TAG to FFA and glycerol



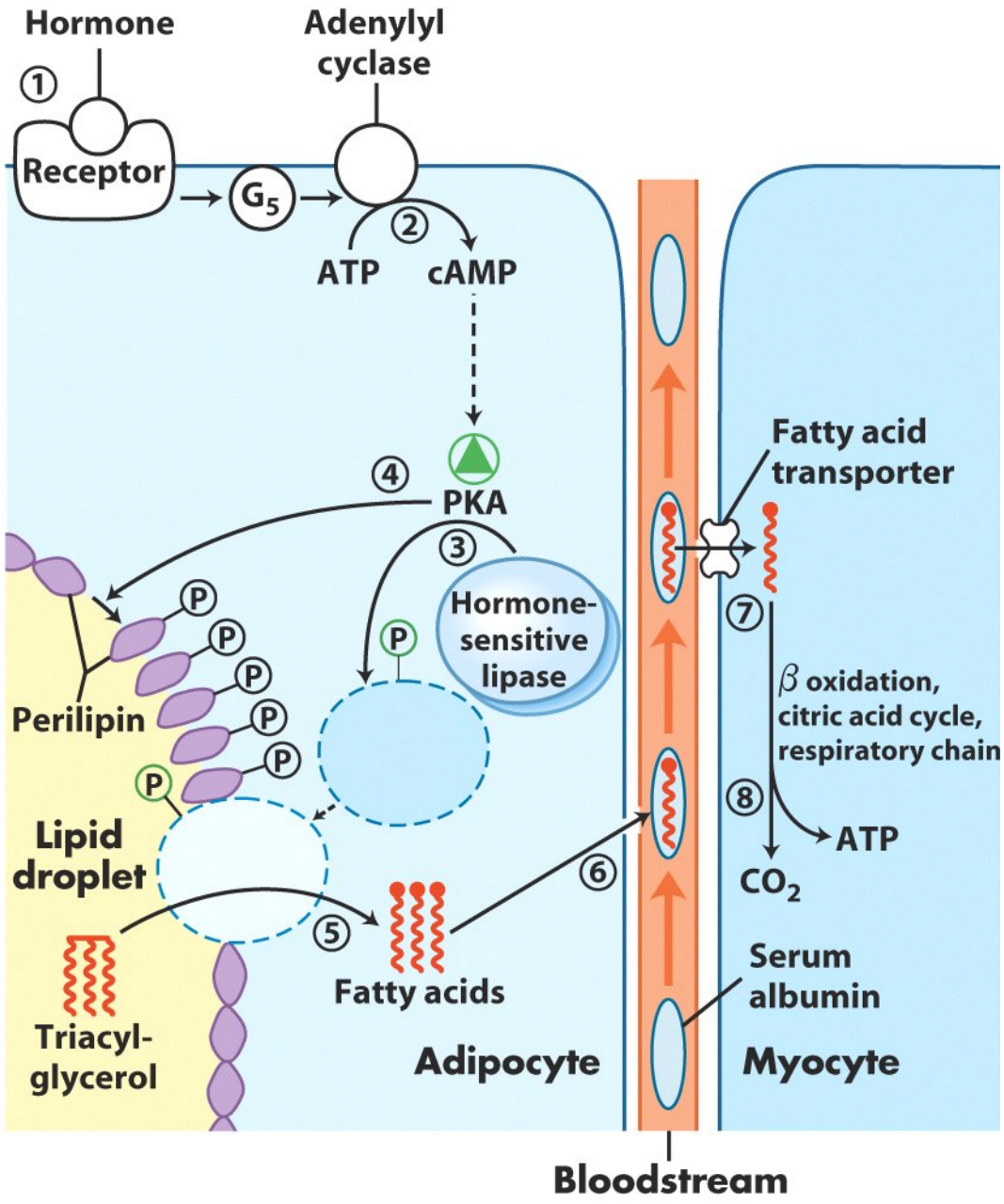


1-Stearoyl, 2-linoleoyl, 3-palmitoyl glycerol,
a mixed triacylglycerol

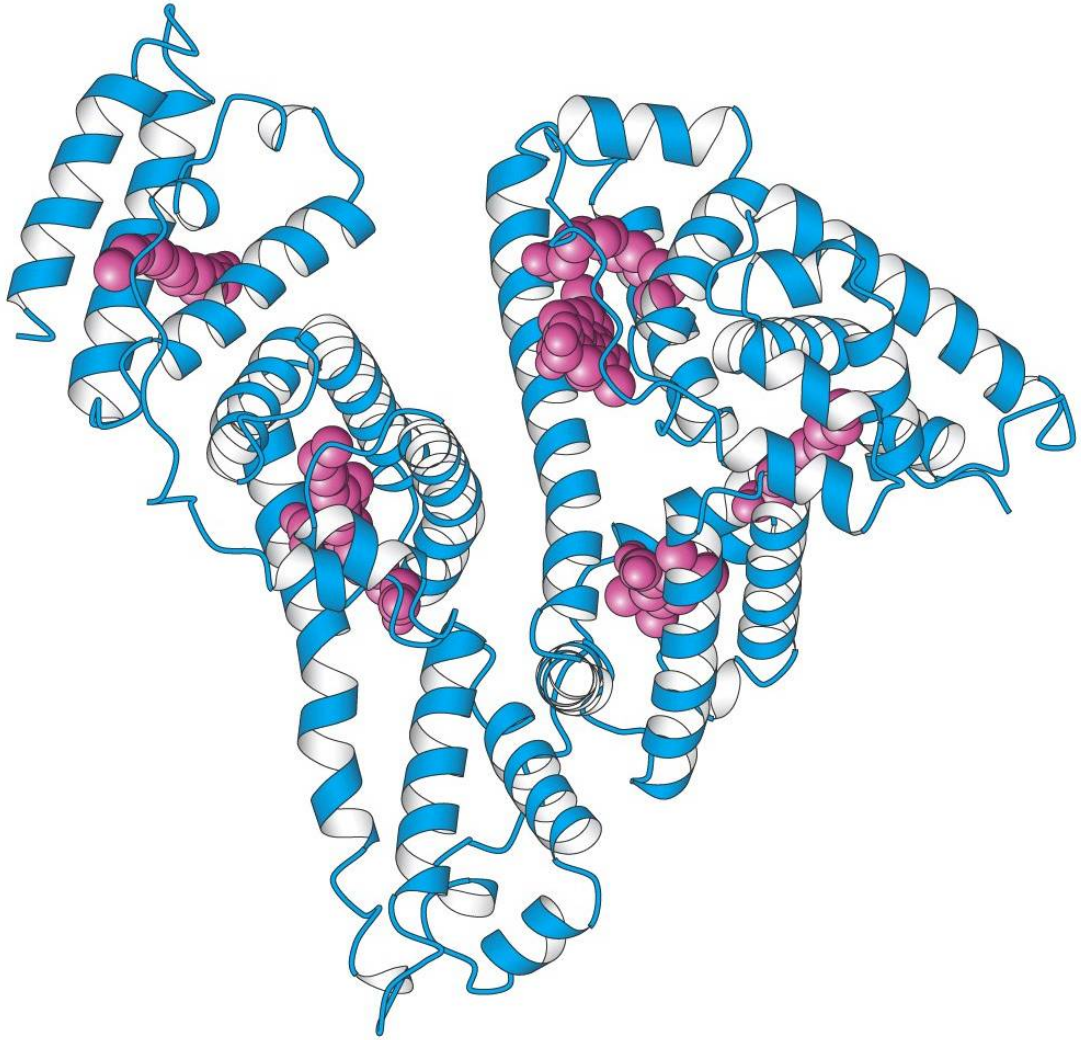
Hormonal regulation of lipolysis

epinephrine & glucagon

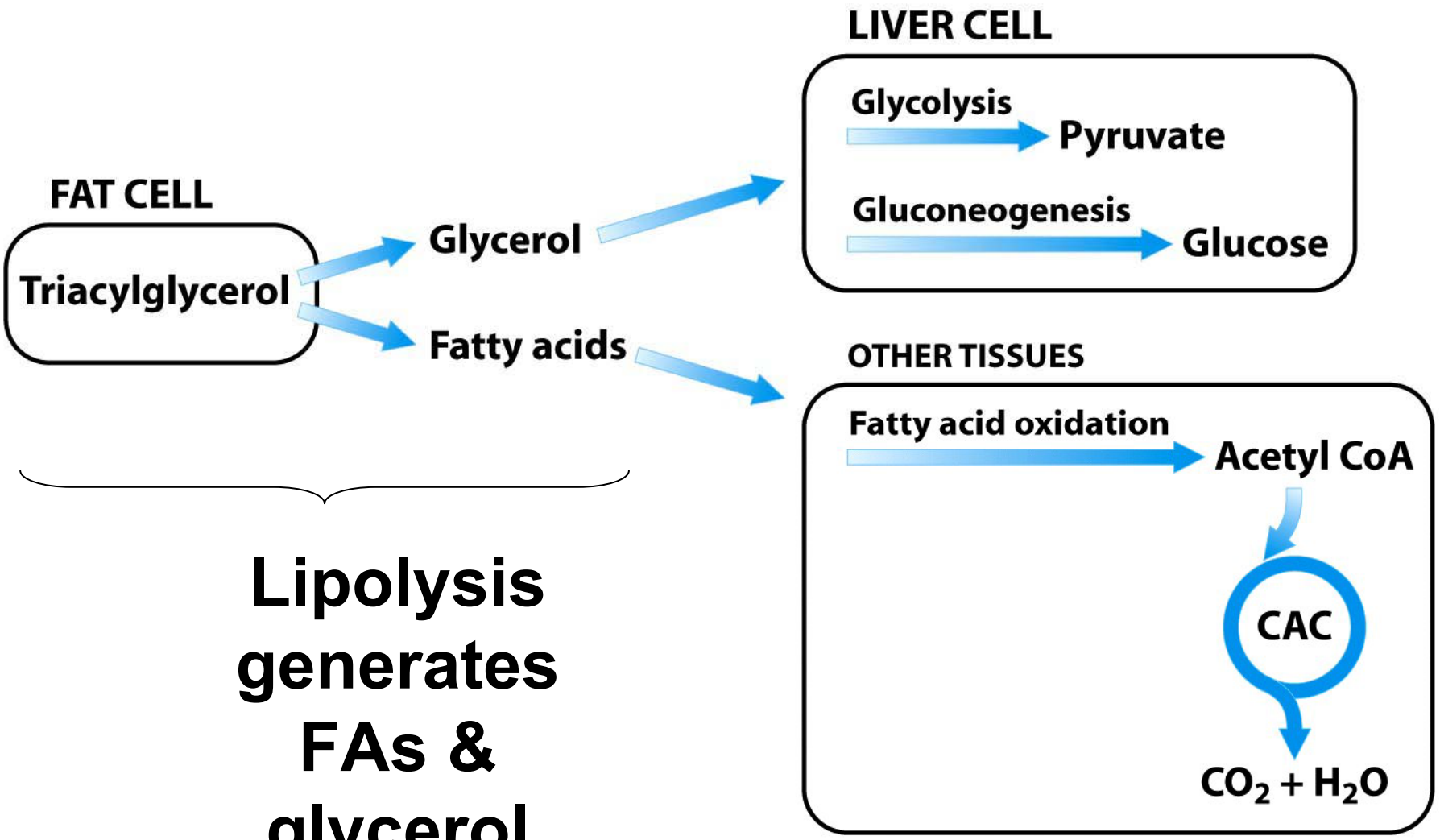
glucagon released when glucose levels are low, causes liver to convert glycogen to glucose



Albumin



transports FA in the blood to the target cell

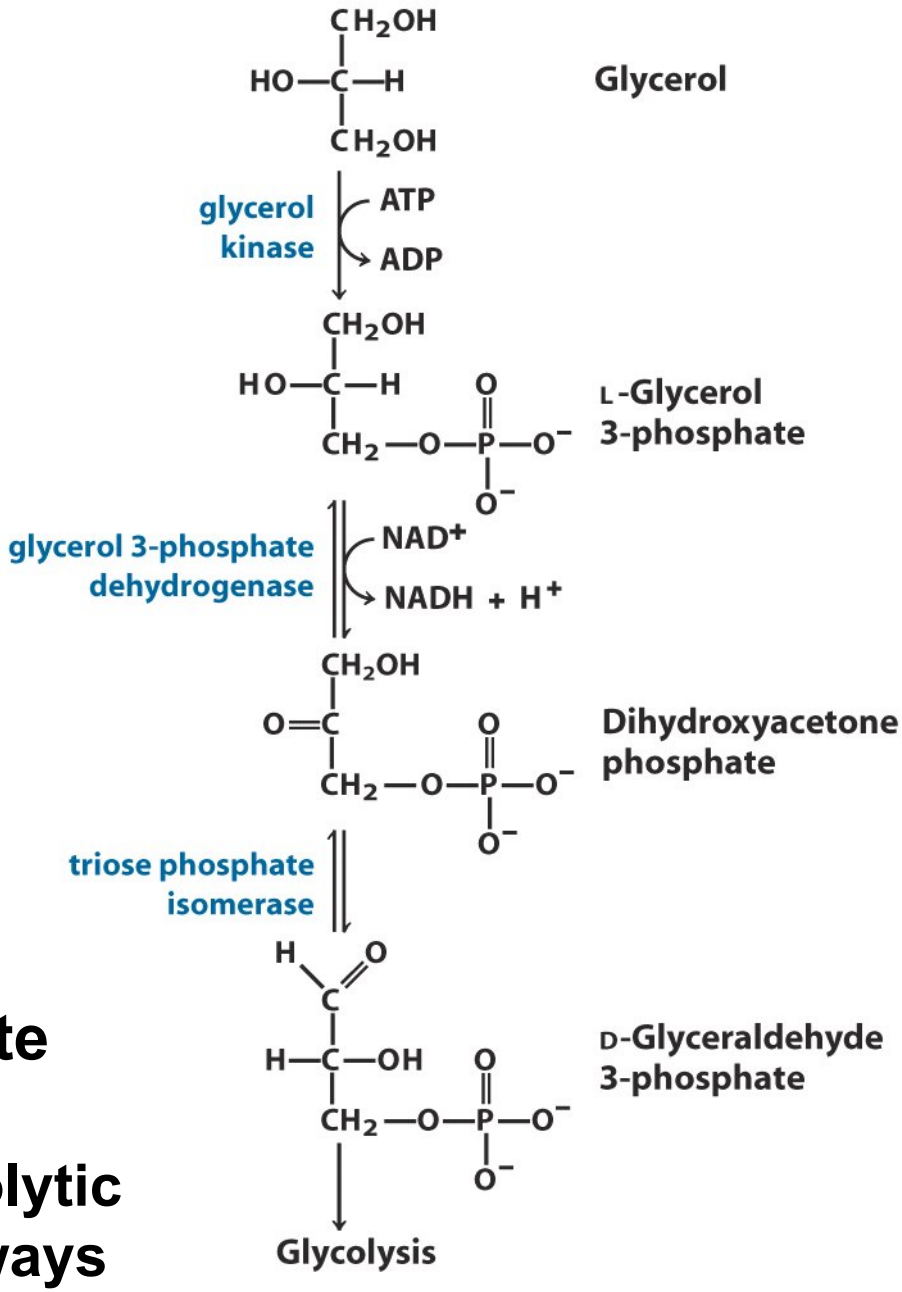


Lipolysis generates FAs & glycerol

CAC=citric acid cycle

Glycerol metabolism in the liver

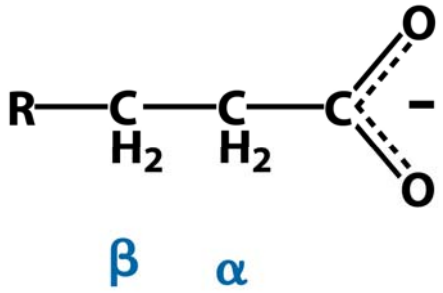
(only 5% of the energy content in TAG is found in glycerol)



glyceraldehyde-3-phosphate

intermediate in both the glycolytic and the gluconeogenic pathways

”β-oxidation” or FA degradation occurs in the mitochrondria



First → the FA is activated (FA-CoA is formed) on the outer mitochondrial membrane of Acyl-CoA-synthetase

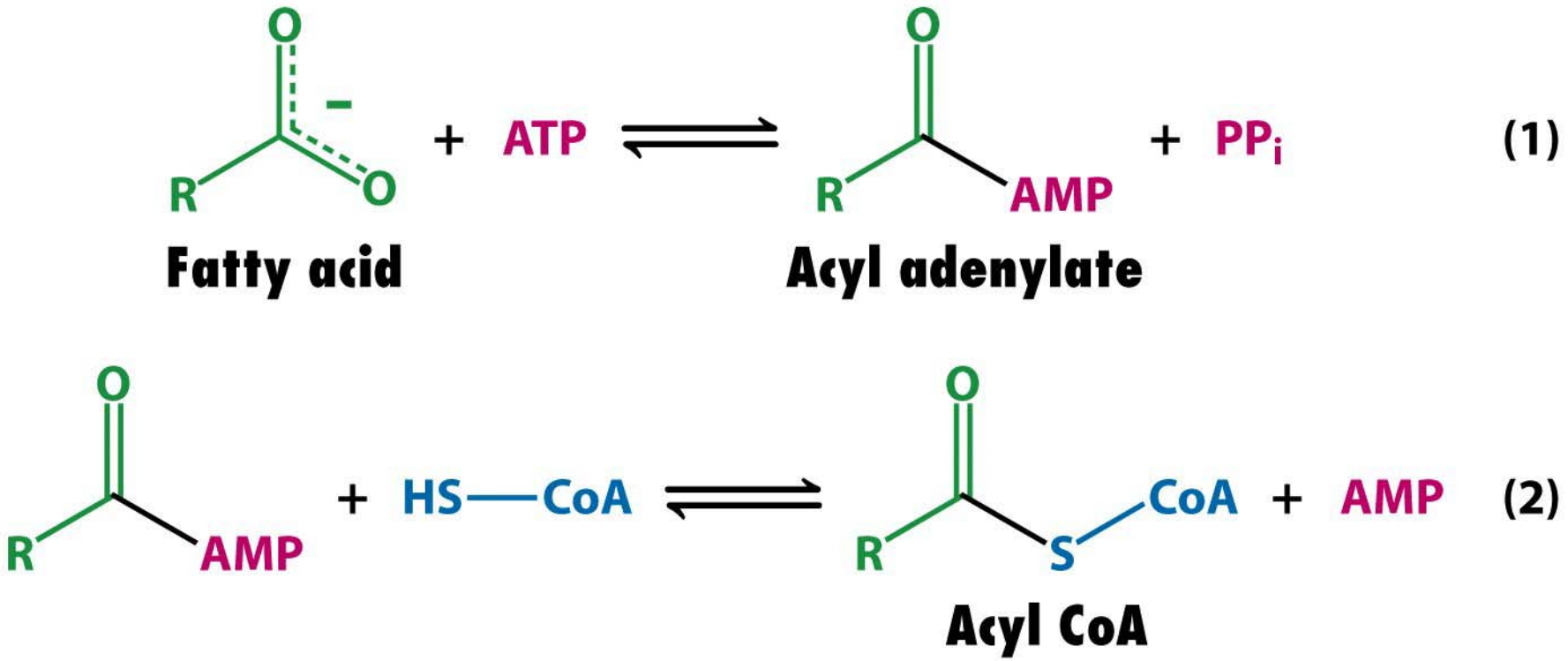
Second → FA-CoA is transported into the mitochondria with help of carnitine if ≥ C_{14}

OVERALL ENERGY PRODUCTION:

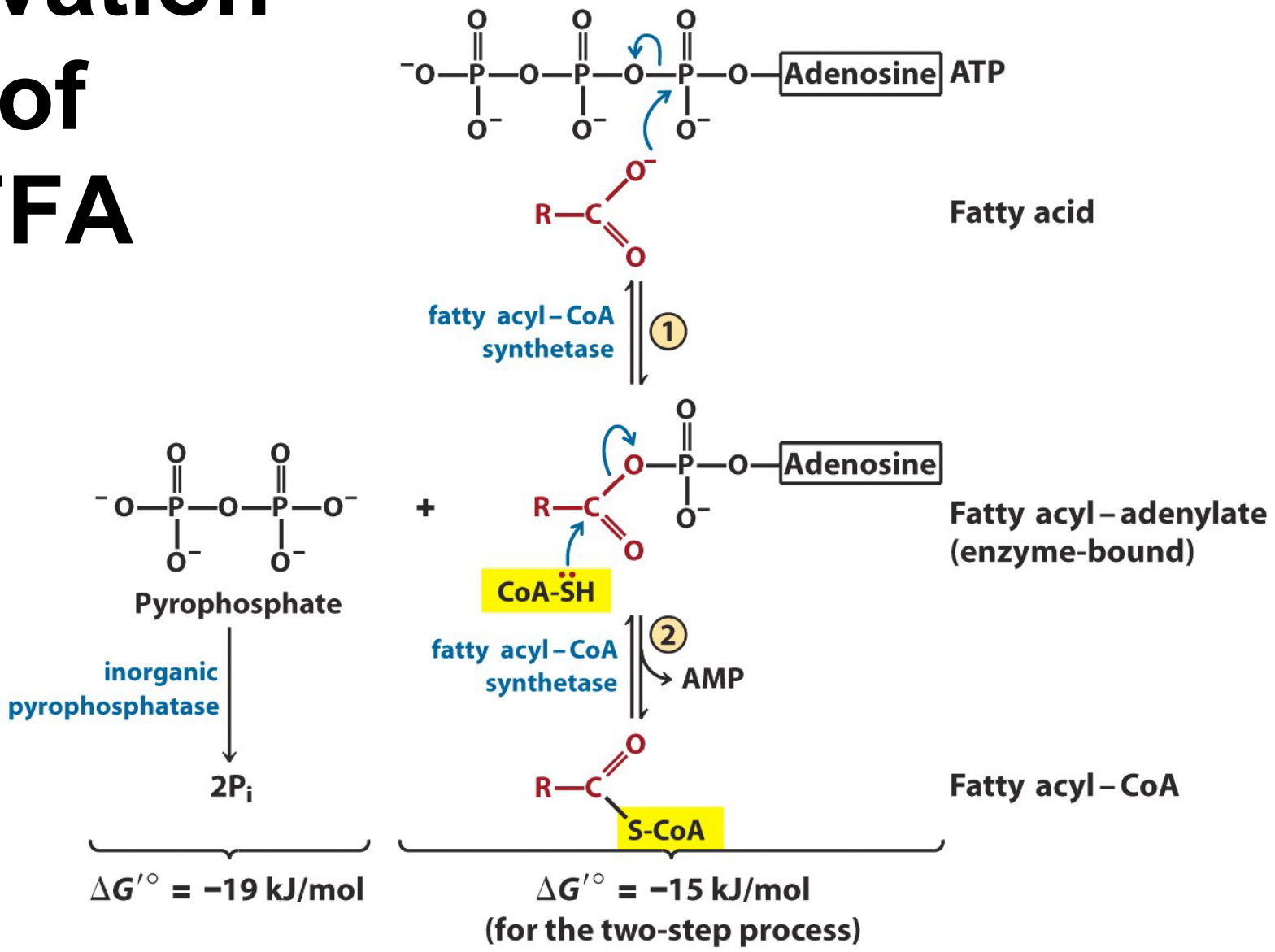
$$\text{Palmitoyl CoA} + 7\ \text{FAD} + 7\ \text{NAD}^+ + 7\ \text{CoA} + 7\ \text{H}_2\text{O} \longrightarrow 8\ \text{AcCoA} + 7\ \text{FADH}_2 + 7\ \text{NADH} + 7\text{H}^+$$

Acyl-CoA-synthetase activates FAs on the outer mitochondrial membrane

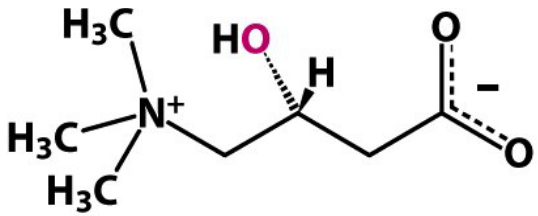
- FA + ATP ⇆ Acyl adenylate + PP_i
- $PP_i + H_2O \rightarrow 2\ P_i$
- Acyl adenylate + HS-CoA ⇆ Acyl-S-CoA + AMP

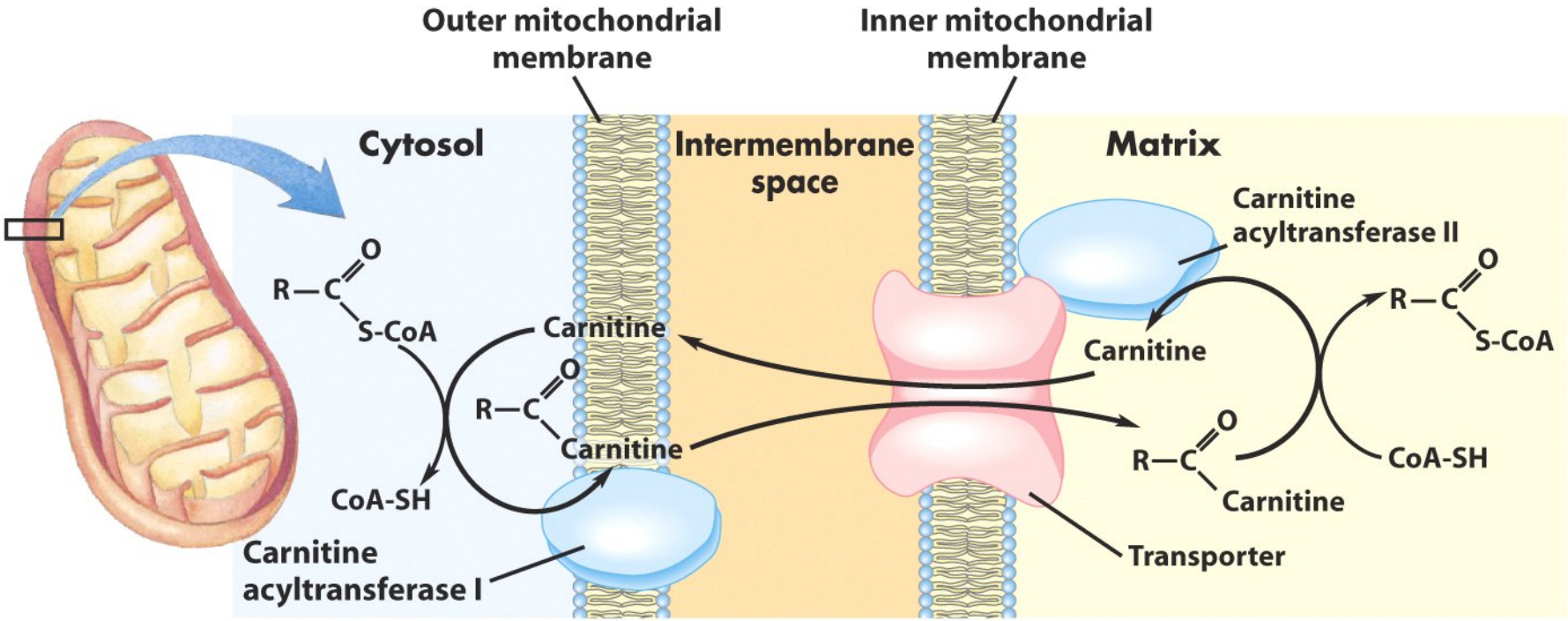


Activation of FFA

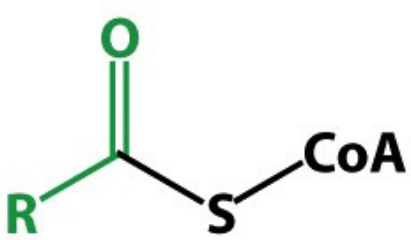


The carnitine shuttle

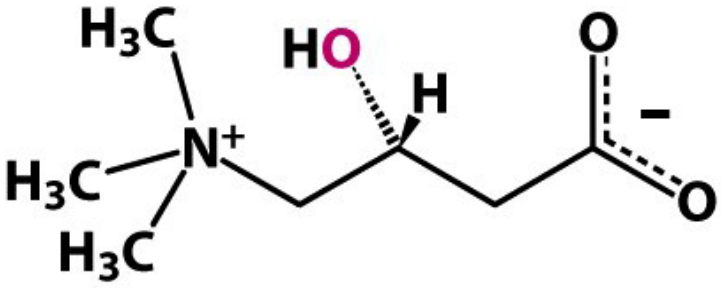




FA are activated on the outer mitochondrial membrane, but oxidized in mitochondrial matrix



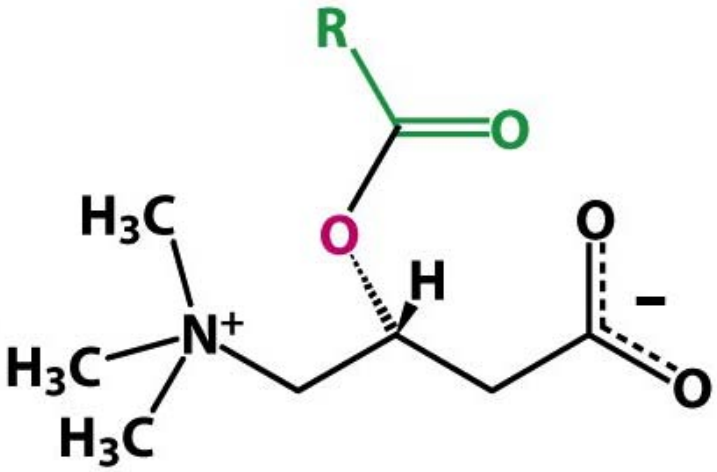
+



Acyl-CoA **Carnitine**

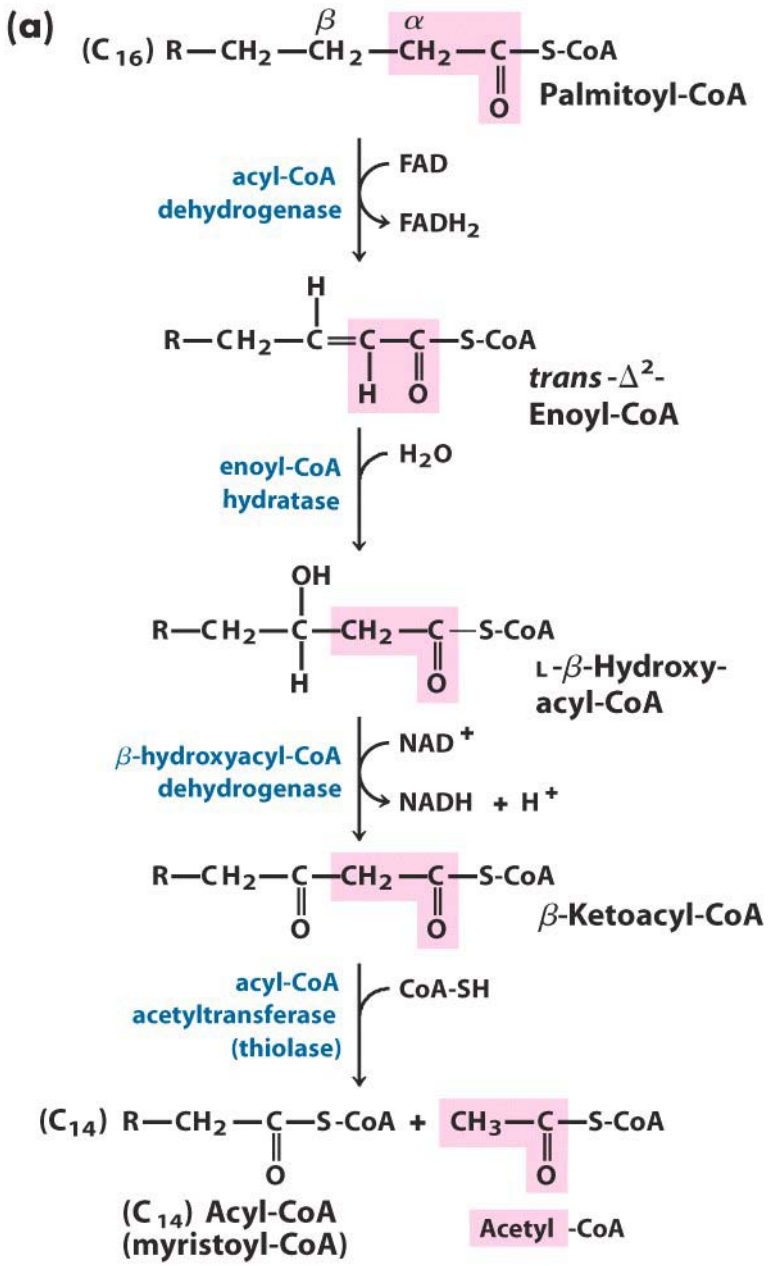
CAT 1 *CAT 2*

CAT = carnitine acyltransferase (AKA: CPT, carnitine acyltransferase)



+ HS—CoA

Acyl carnitine

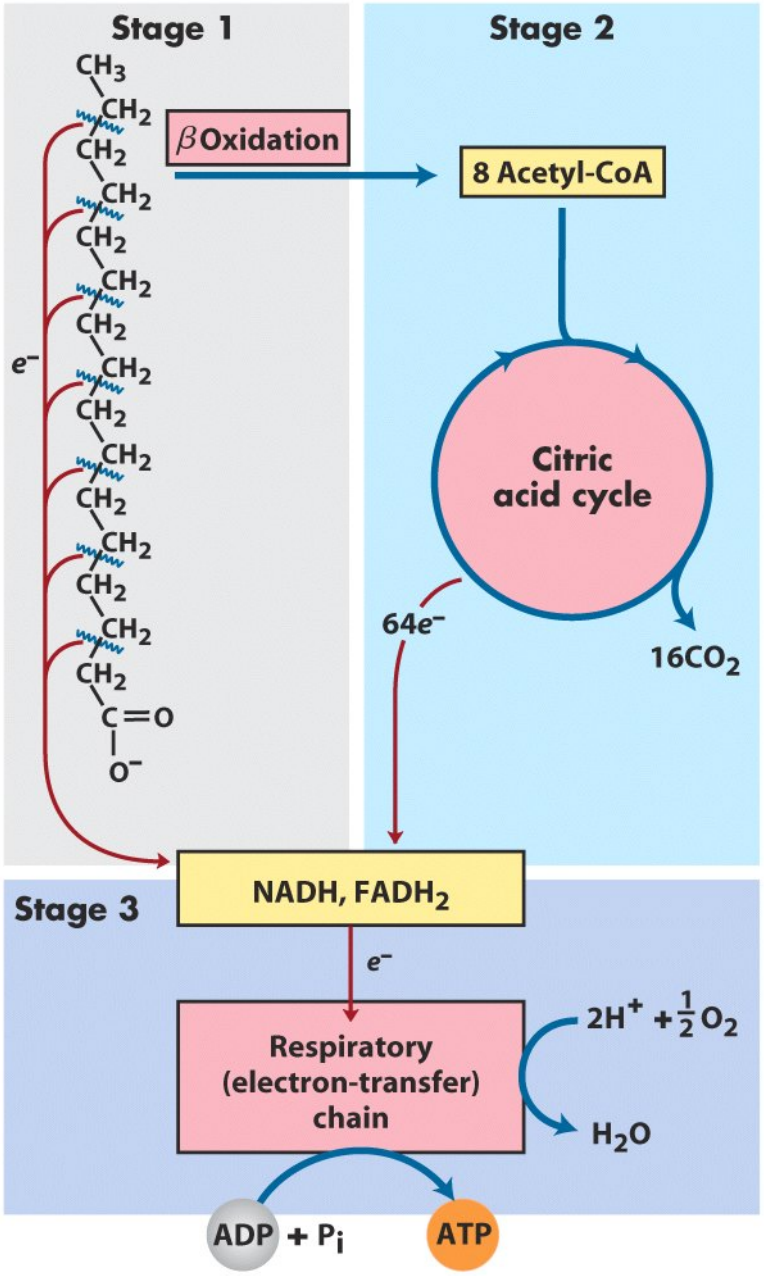


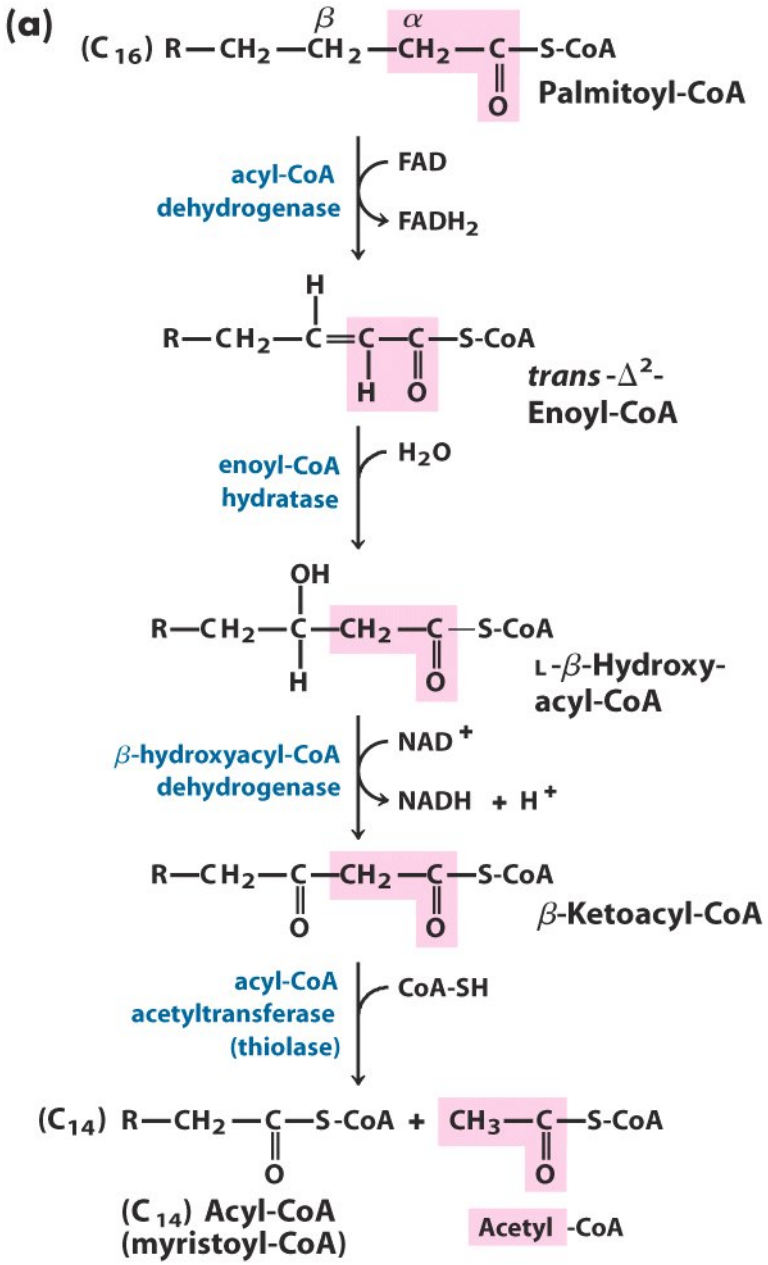
β-oxidationen has 4 stages and generates $FADH_2$, NADH and Ac-CoA

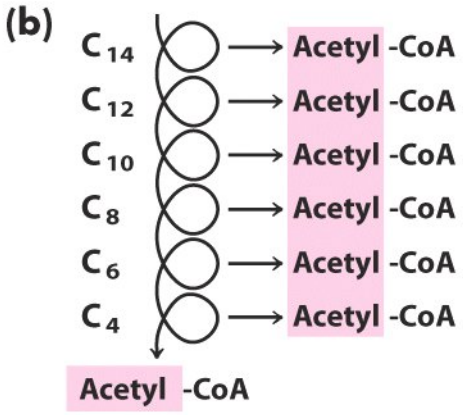
Ac-CoA
is further oxidized in the citric acid cycle

and

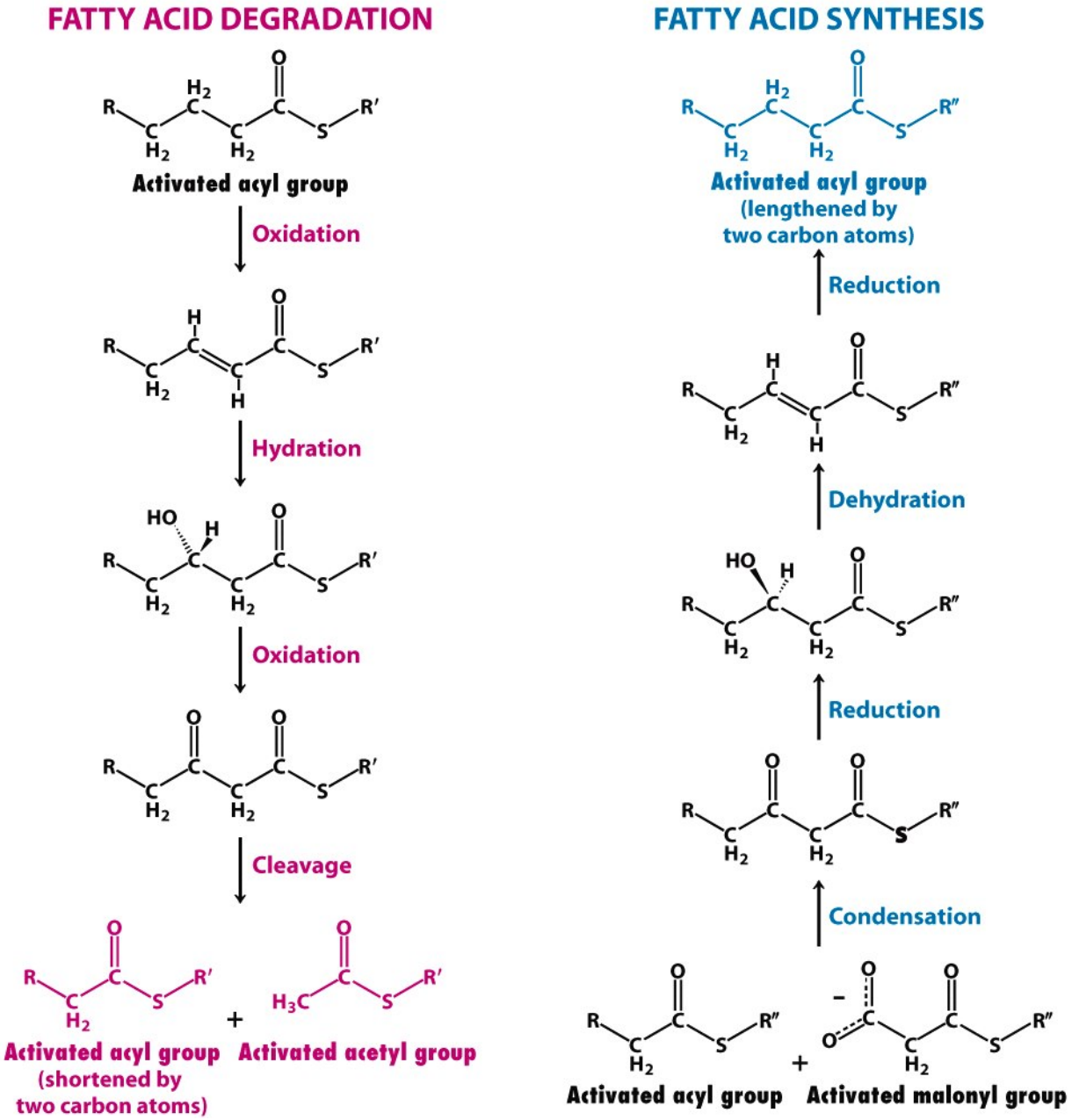
NADH and $FADH_2$
in the respiratory chain



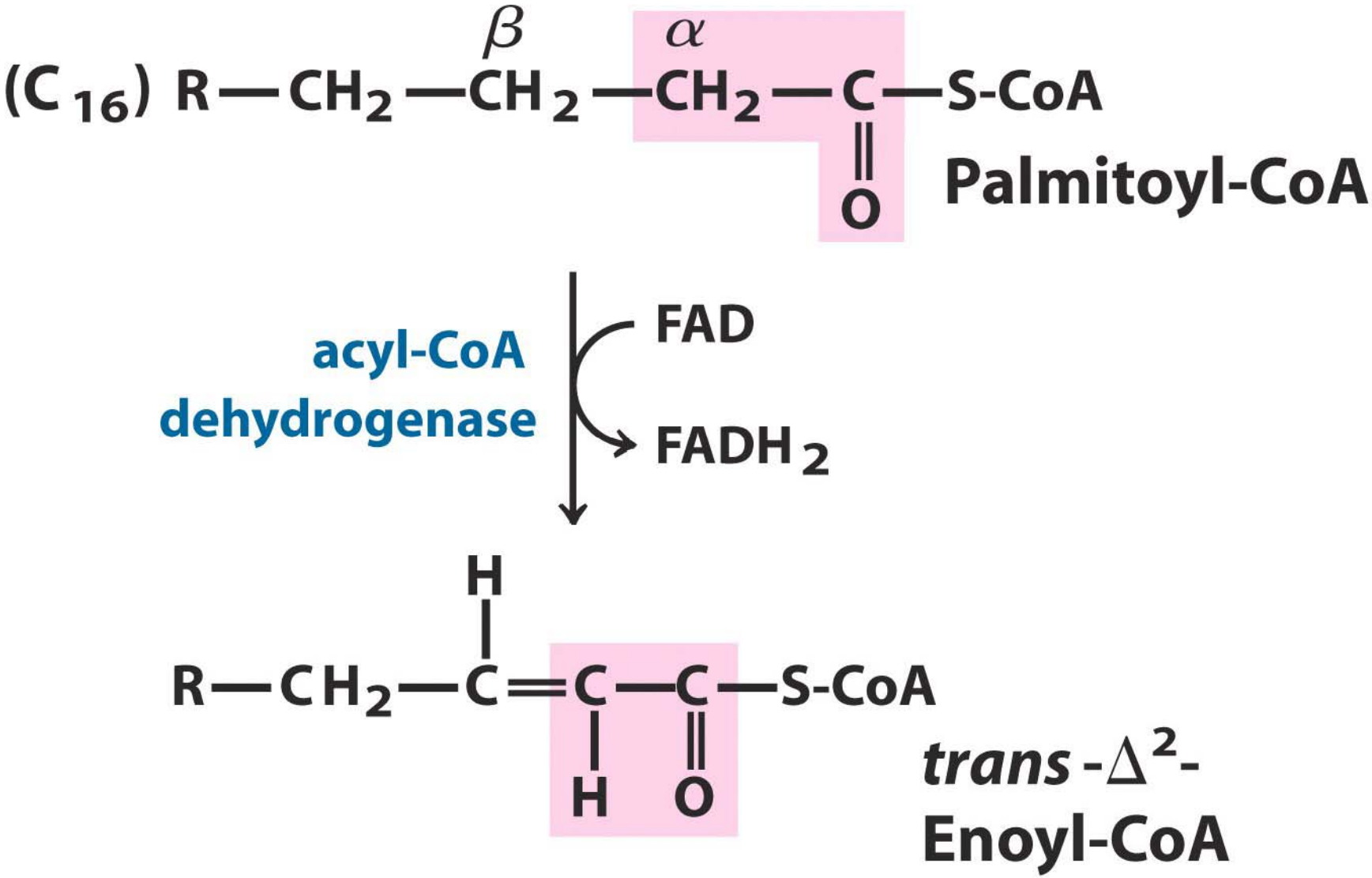




β-oxidation's 4 stages are repeated - each time 2 carbons are cleaved off

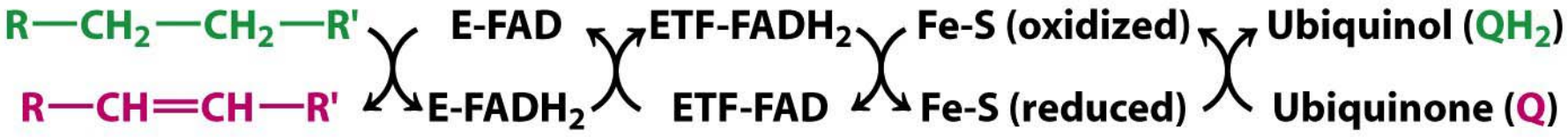

FATTY ACID DEGRADATION
Activated acyl group
Oxidation
Hydration
Oxidation
Cleavage
Activated acyl group
(shortened by
two carbon atoms)
Activated acetyl group
FATTY ACID SYNTHESIS
Activated acyl group
(lengthened by
two carbon atoms)
Reduction
Dehydration
Reduction
Condensation
Activated acyl group
Activated malonyl group

Stage 1: Oxidation

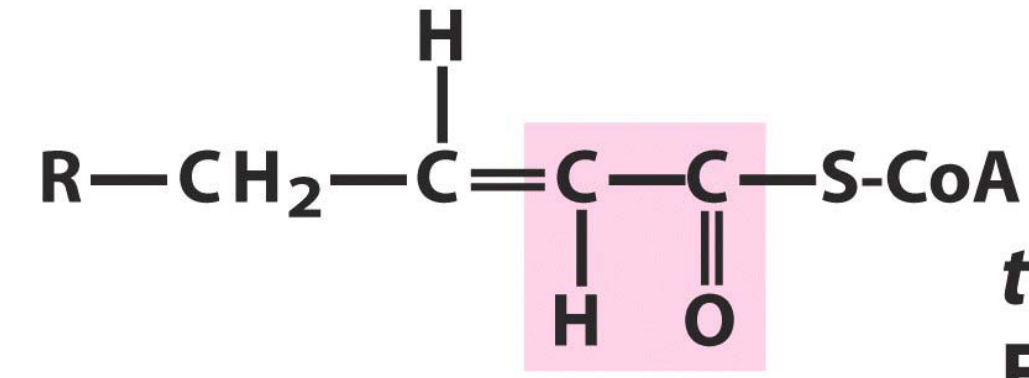


Acyl-CoA-dehydrogenase

- Forms a trans double bond between the α- and β-carbons
- 3 isozymes depending upon FA length:
  - 12-18 VL (very long) chain
  - 4-14 M (medium) chain
  - 4- 8 S (short) chain



Stage 2: Hydration

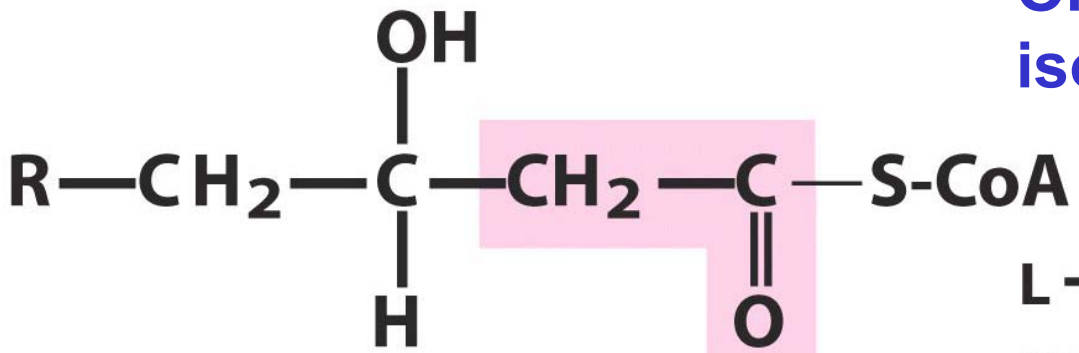


trans-Δ^2-Enoyl-CoA

enoyl-CoA hydratase

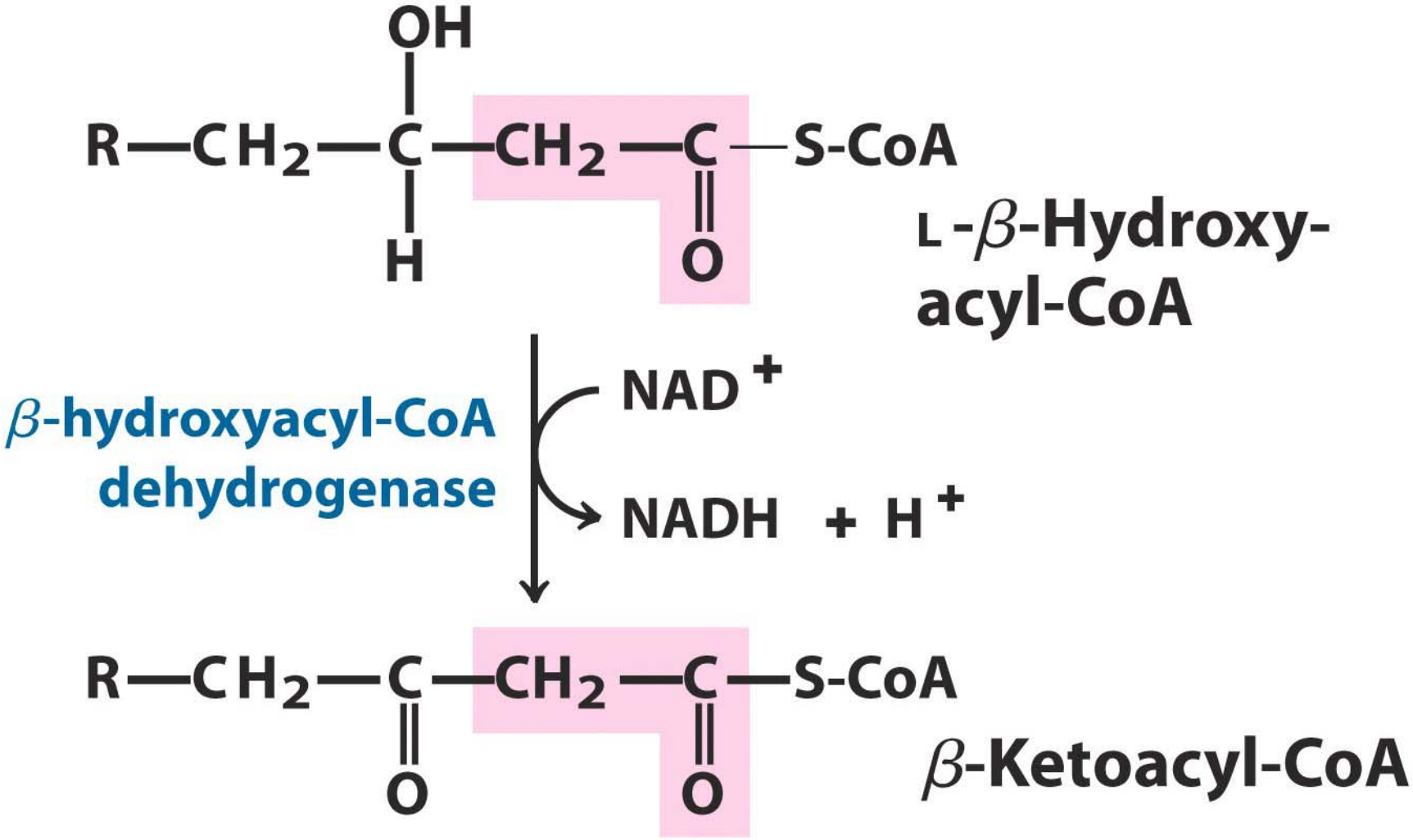
H_2O

Stereospecific! Only the L-isomer is formed

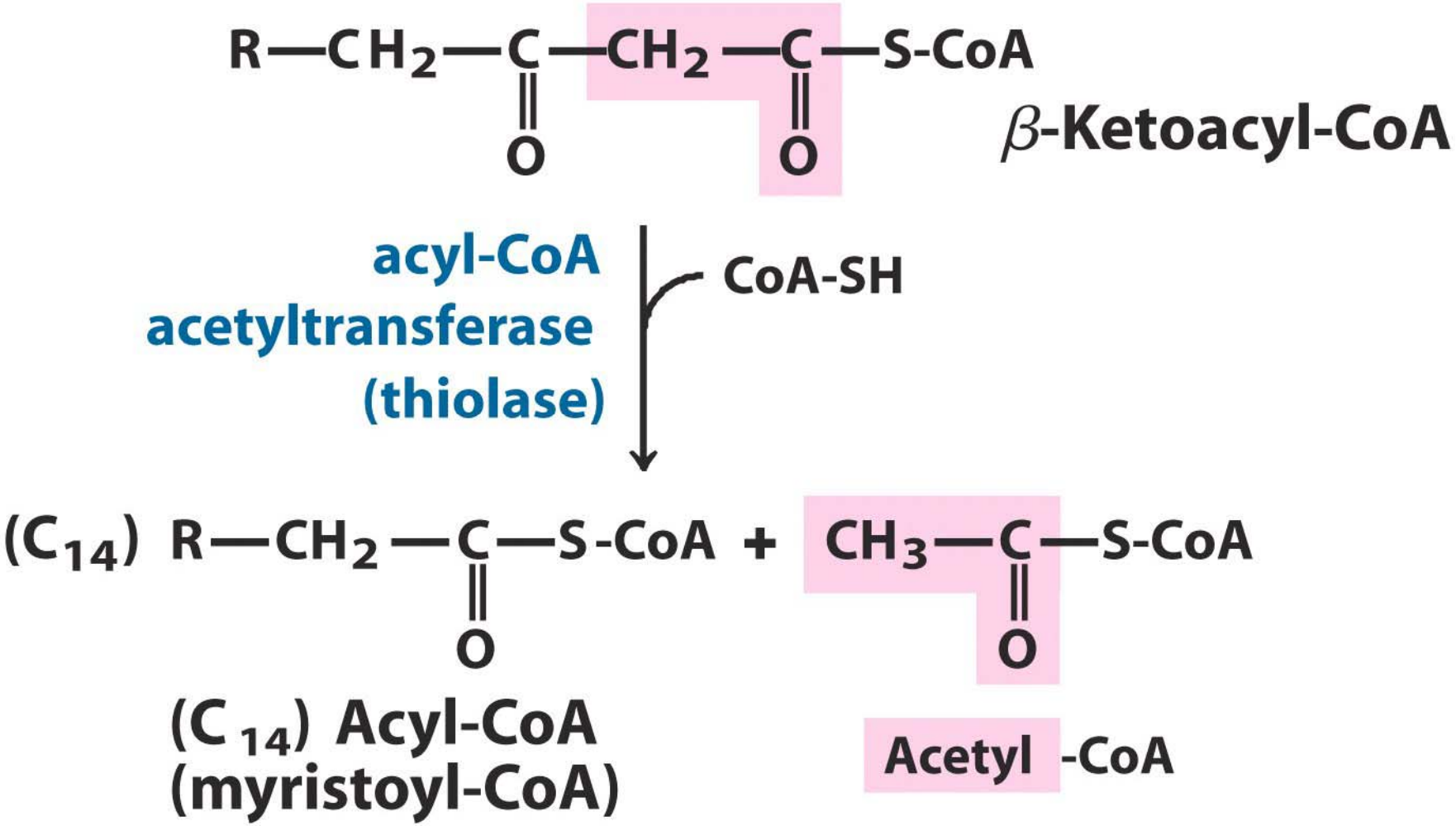


L-β-Hydroxy-acyl-CoA

Stage 3: Oxidation



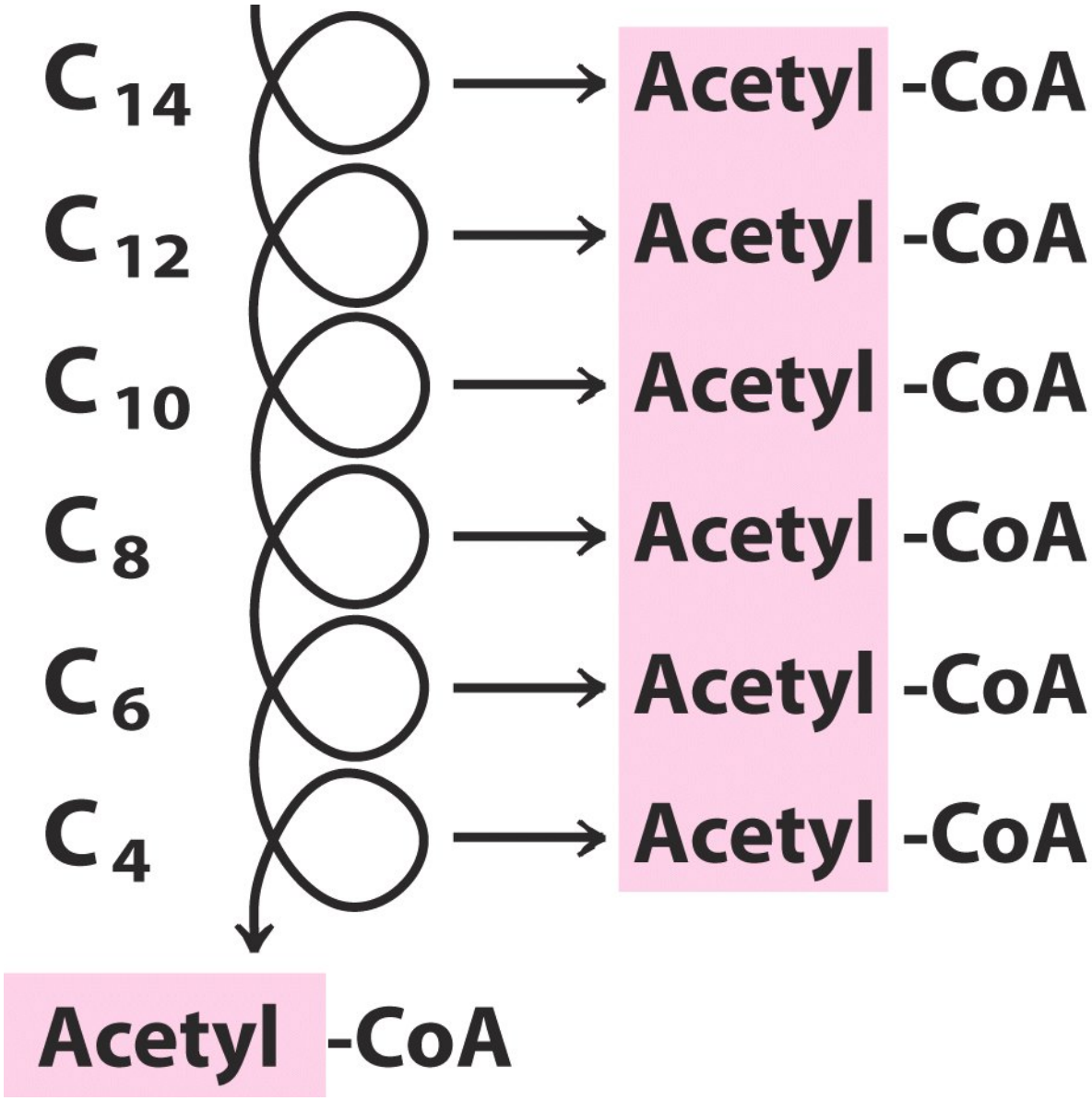
Stage 4: Thiolysis (cleavage of the α-β-bond)



Stages 2-4 use different enzymes depending on the FA chain length

≥ C12: enzyme complex in the inner membrane

< C12: 4 individual, soluble enzymes in the nearby matrix



Complete oxidation of palmitate yields 106 molecules of ATP

$7FADH_2$ = 10.5 ATP

7NADH = 17.5 ATP

8Ac CoA = 80 ATP

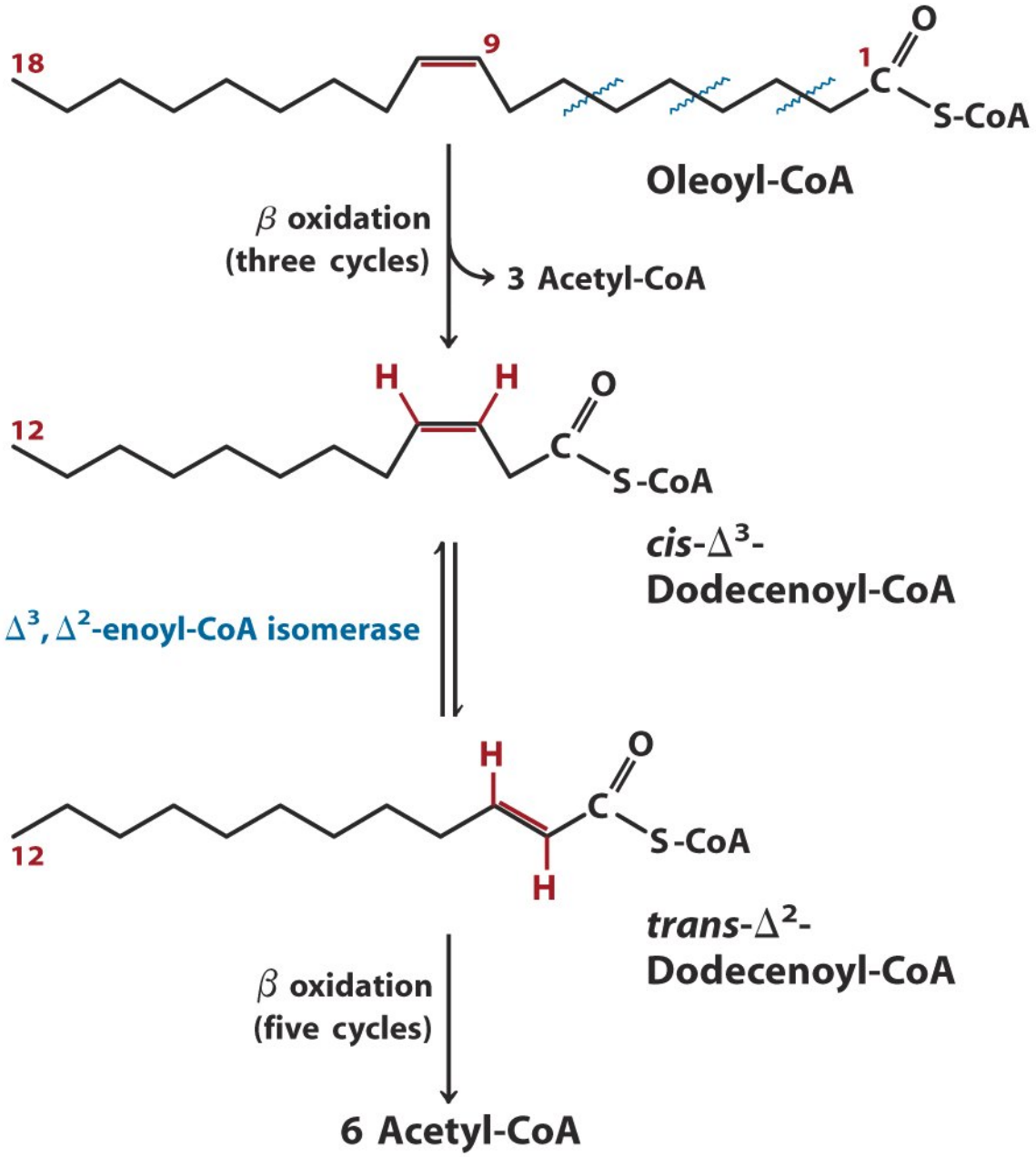
but 2 ATP lost in palmitate activation

TOTAL = 106 ATP

$$C_n\text{-acyl CoA} + FAD + NAD^+ + CoA + H_2O \longrightarrow C_{n-2}AcCoA + FADH_2 + NADH + H^+$$

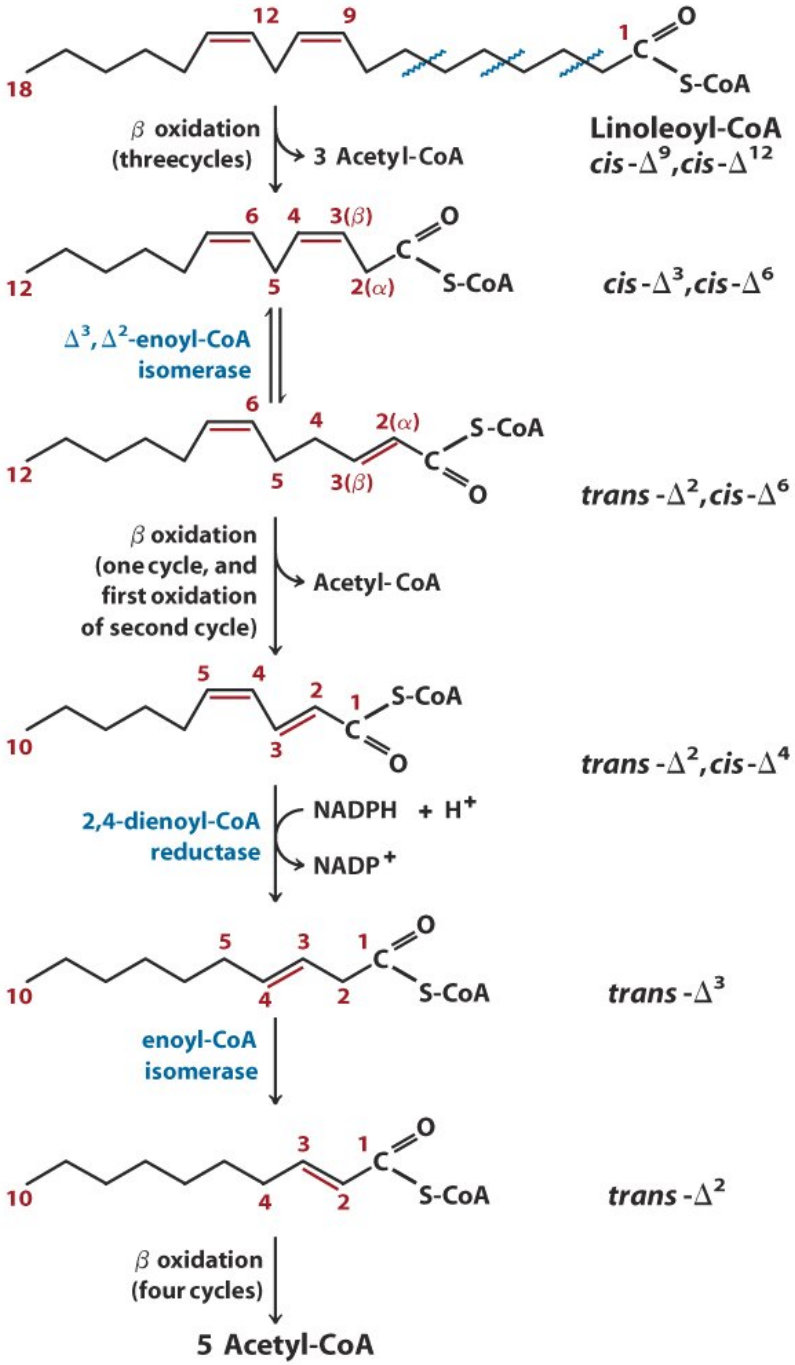
Oxidation of mono-unsaturated fatty acids (MUFAs)

- needs an isomerase

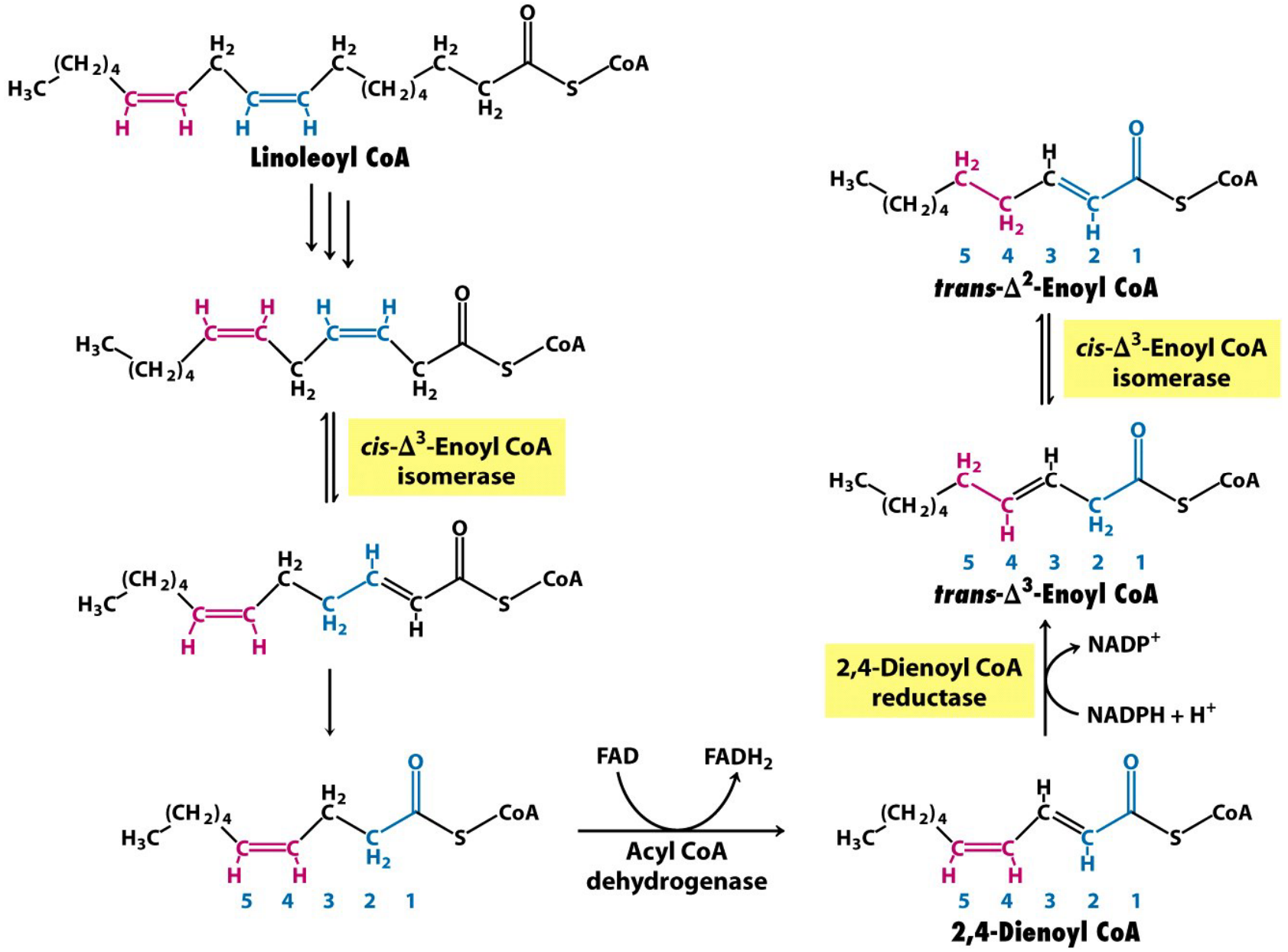


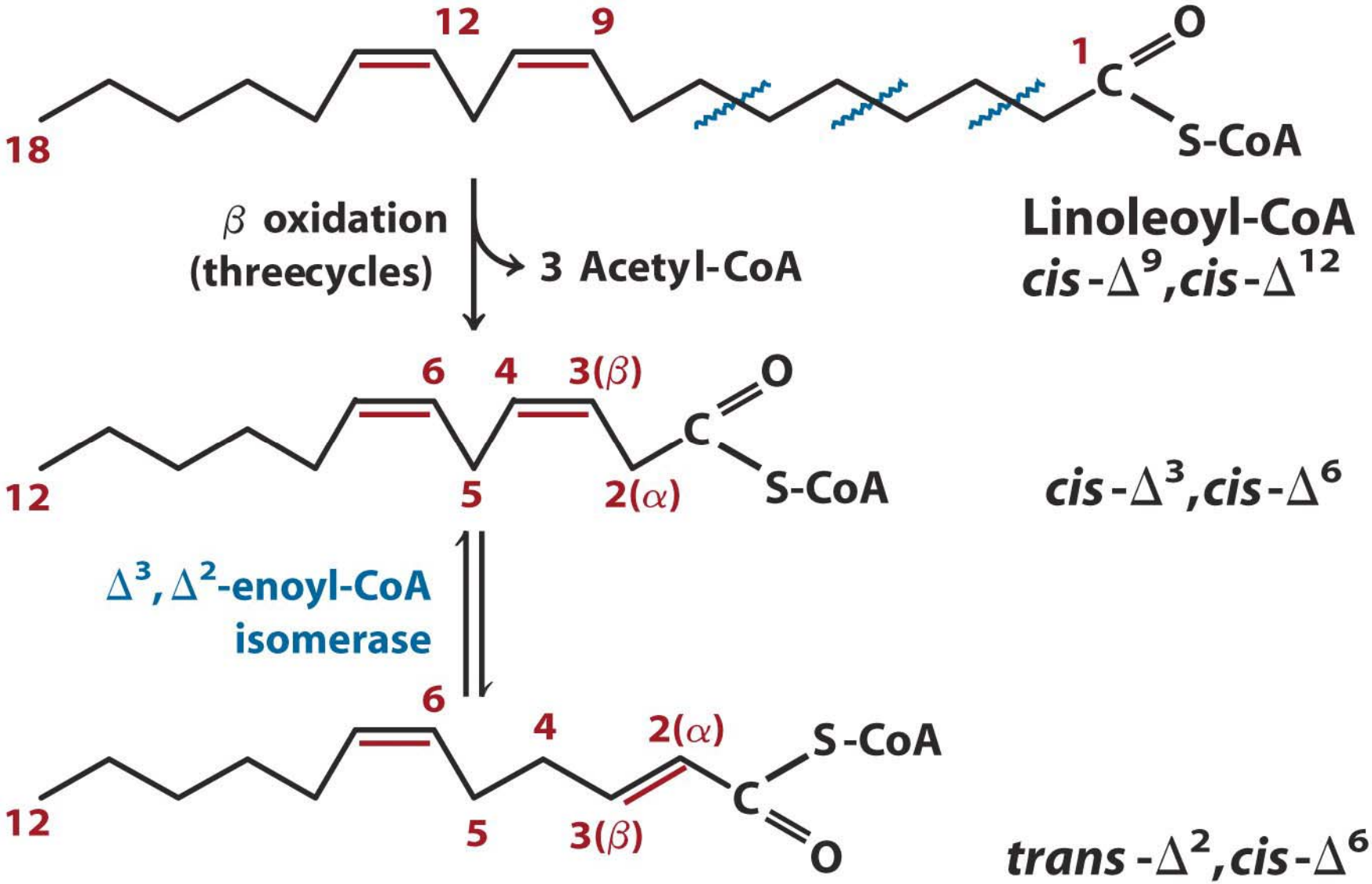
Oxidation of poly-unsaturated fatty acids (PUFAs)

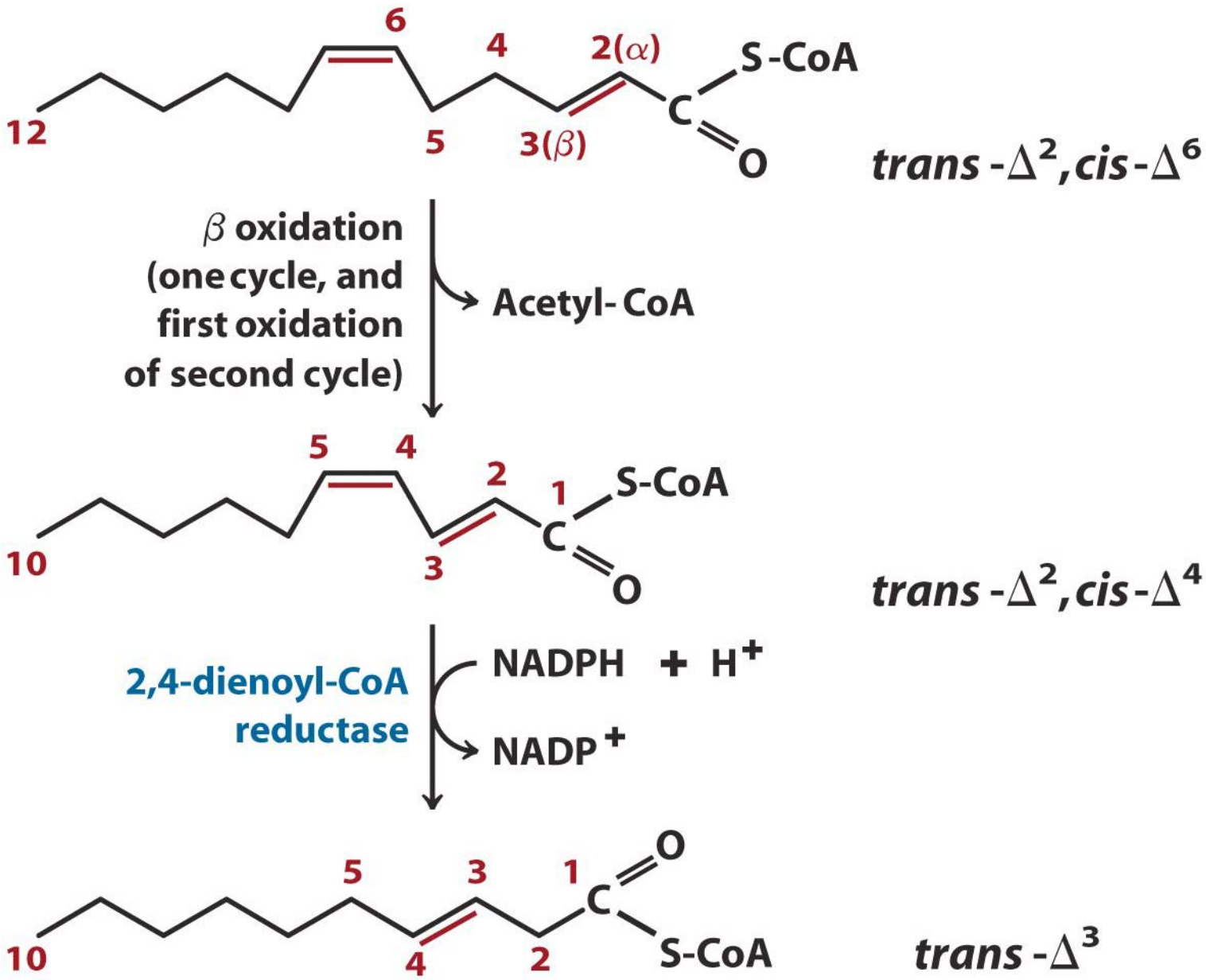
- needs both isomerase and reductase

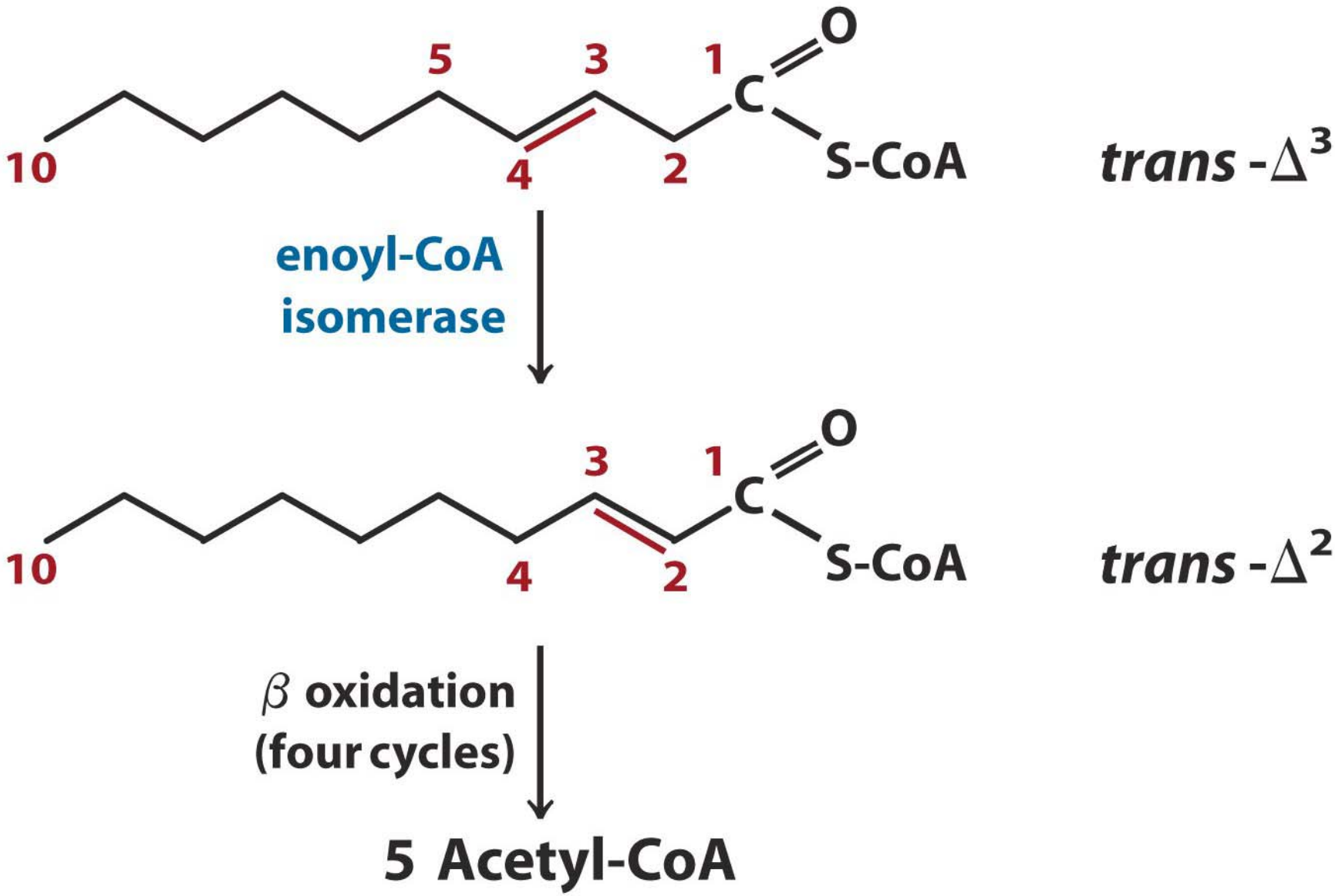


Oxidation of PUFAs
- needs both isomerase and reductase

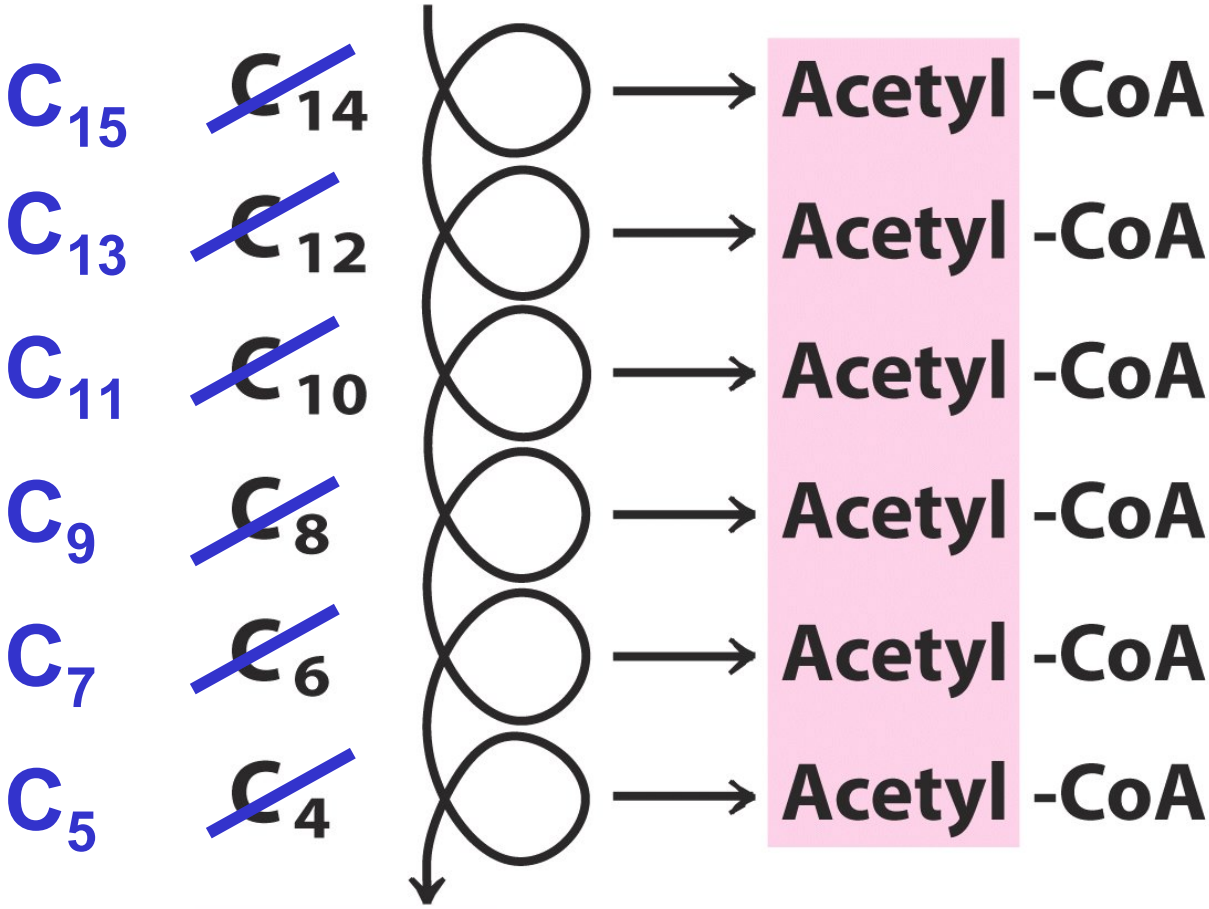


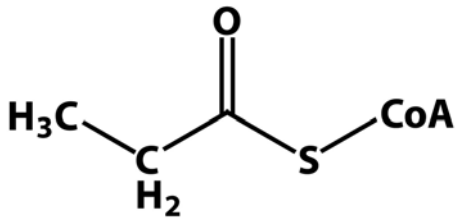

12
9
1
O
C
S-CoA
18
β oxidation
(threecycles)
3 Acetyl-CoA
Linoleoyl-CoA
cis-Δ9,cis-Δ12
6
4
3(β)
O
C
S-CoA
12
5
2(α)
cis-Δ3,cis-Δ6
Δ3, Δ2-enoyl-CoA
isomerase
6
4
2(α)
S-CoA
C
12
5
3(β)
O
trans-Δ2,cis-Δ6


6
4
2(α)
S-CoA
12
5
3(β)
C
O
trans -Δ2, cis -Δ6
β oxidation
(one cycle, and
first oxidation
of second cycle)
Acetyl- CoA
5
4
2
1
S-CoA
10
3
C
O
trans -Δ2, cis -Δ4
2,4-dienoyl-CoA
reductase
NADPH + H+
NADP+
5
3
1
O
10
4
2
C
S-CoA
trans -Δ3

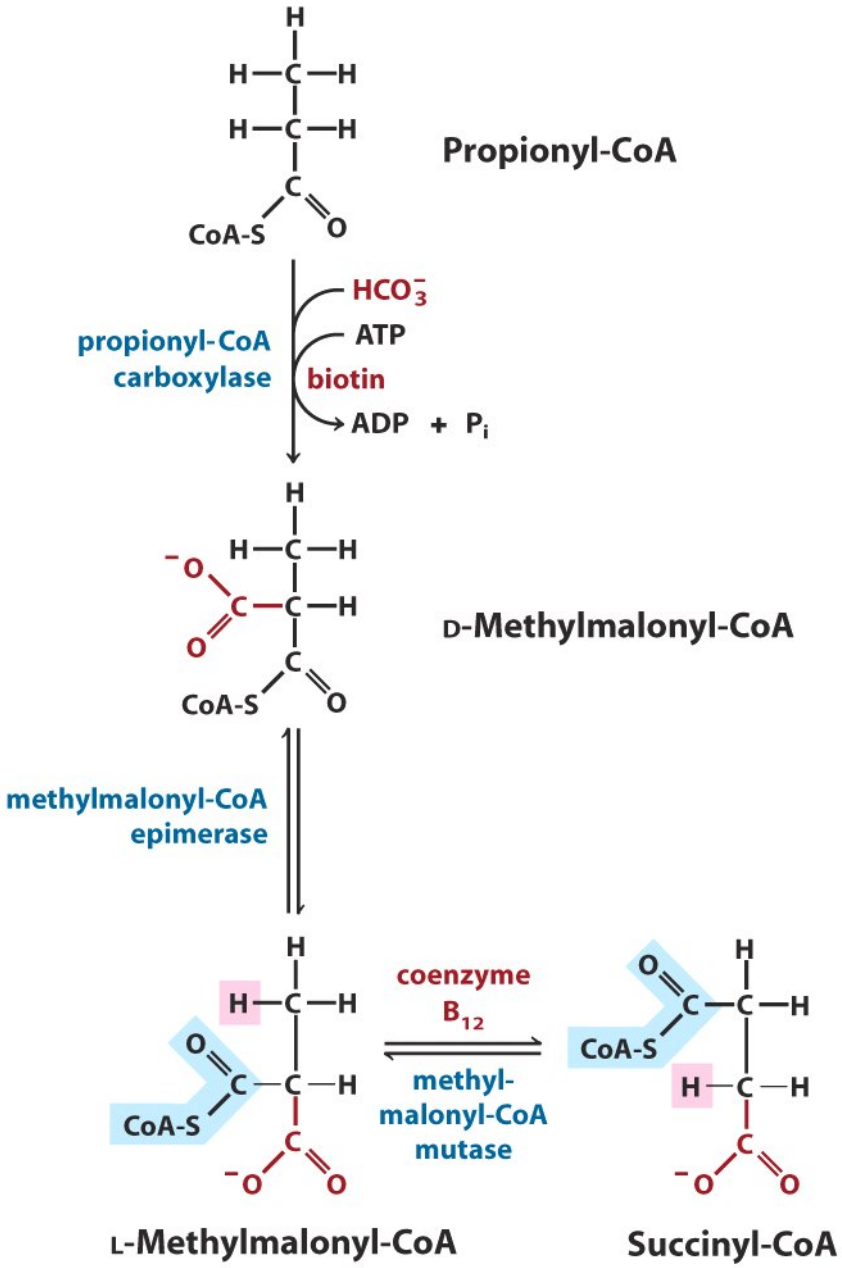

10
5
4
3
2
1
C
O
S-CoA
trans -Δ3
enoyl-CoA
isomerase
10
4
3
2
1
C
O
S-CoA
trans -Δ2
β oxidation
(four cycles)
5 Acetyl-CoA

Oxidation of odd chain odd chain FAs





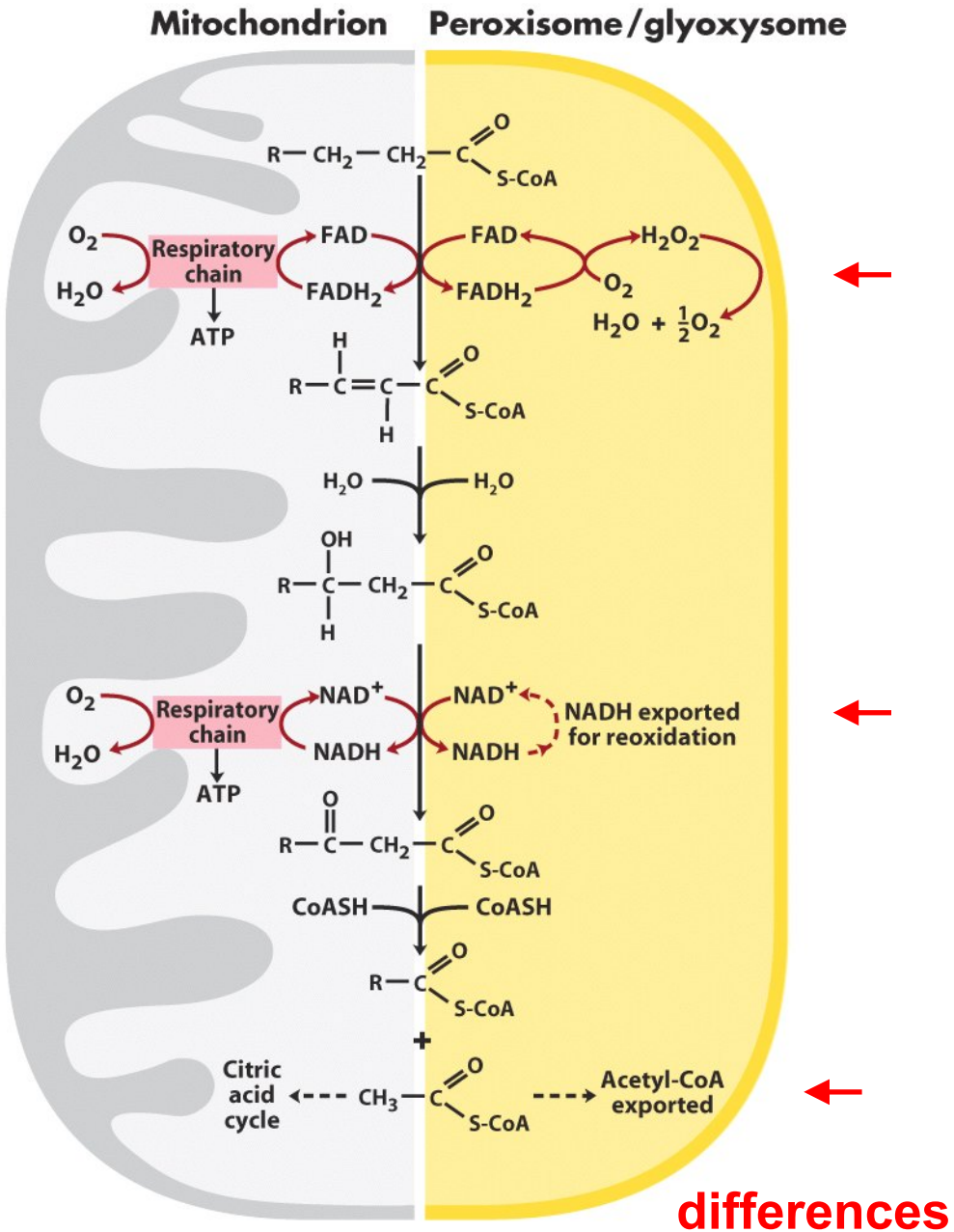
Oxidation of propionyl-CoA



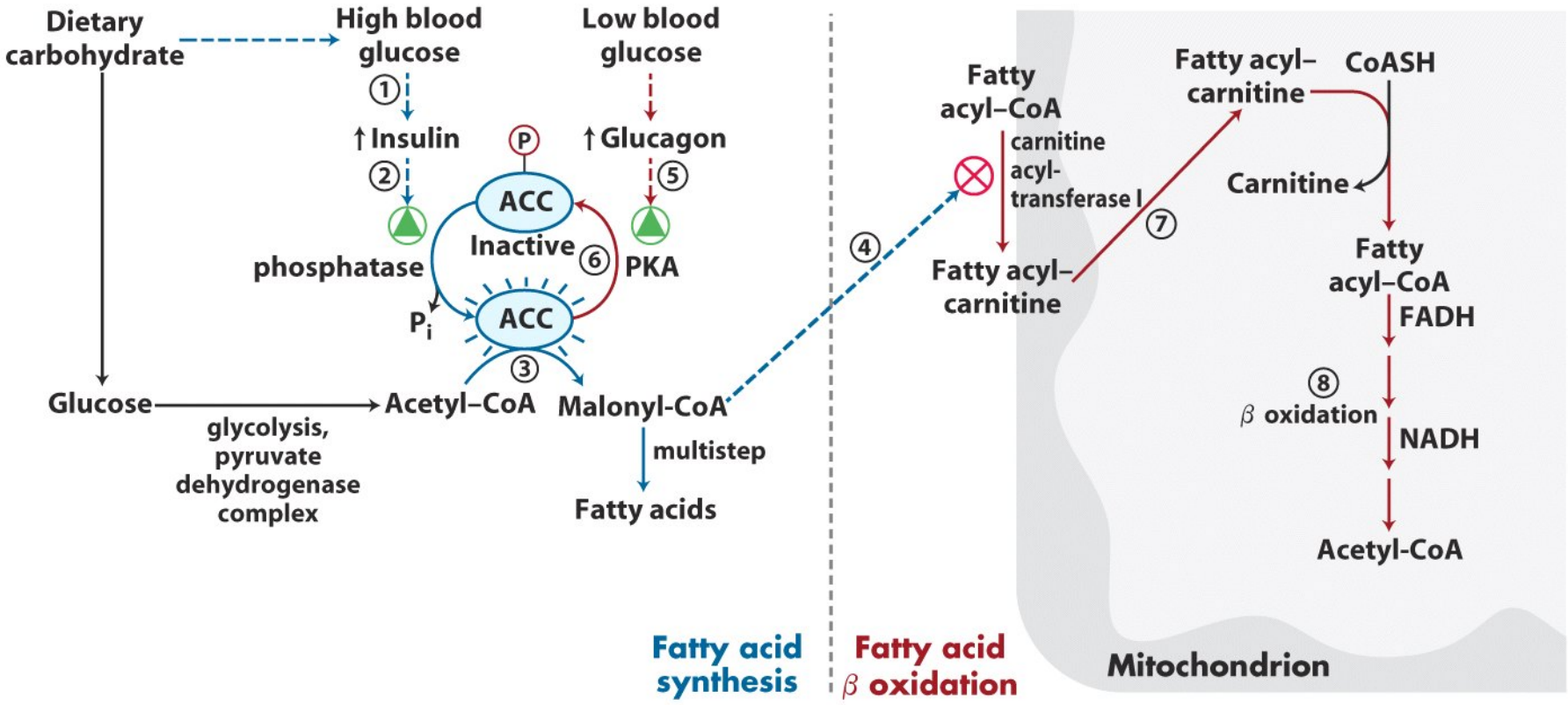
citric acid cycle

FA-oxidation also occurs in peroxisomes

- very long FA (but stops at octanoyl CoA)



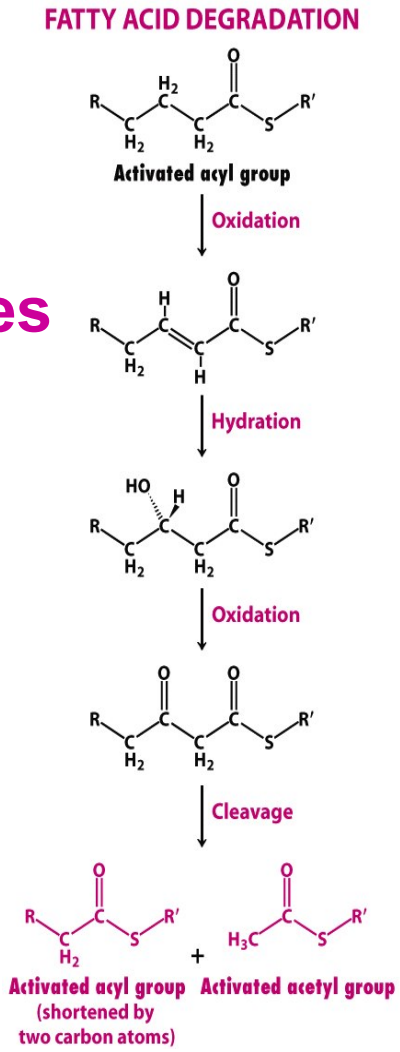
Regulation of FA metabolism

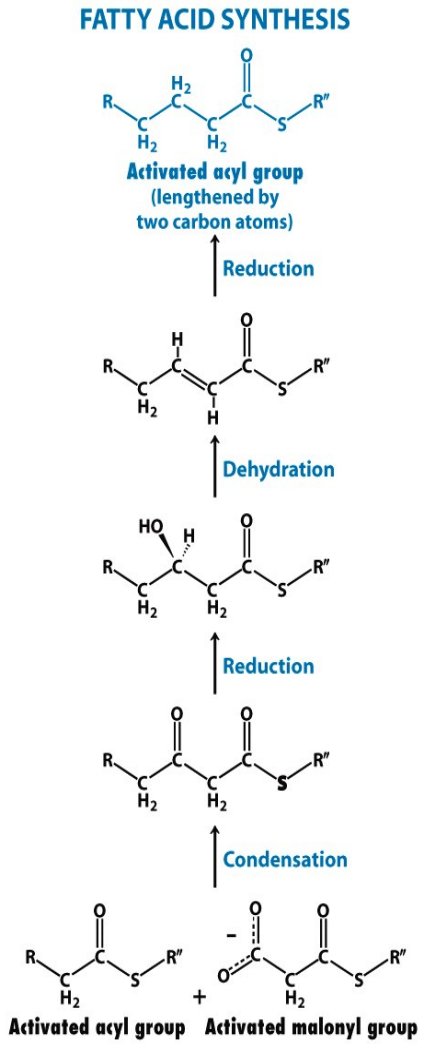


FA syntheisis and degration do not run at the same time

FA-degradation

- Mito matrix
- CoA
- Multiple enzymes
- Ac-CoA
- NAD^+, FAD

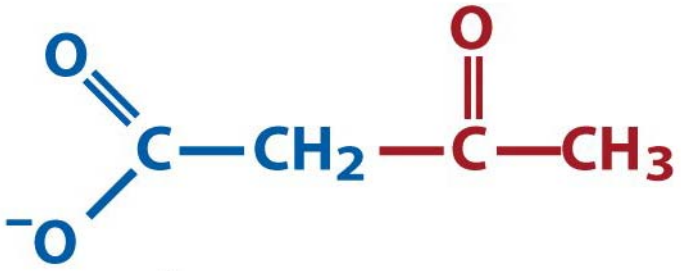




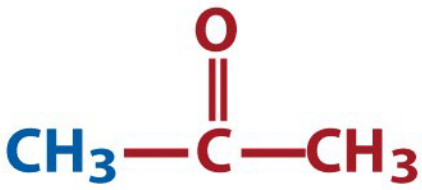
FA-synthesis

- Cytosol
- ACP
- Enzyme complex
- Mal-CoA
- NADPH

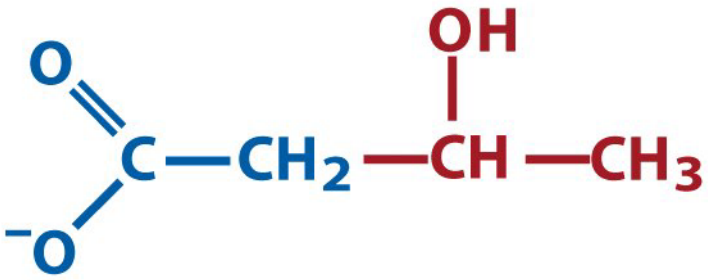
Ketone bodies



Acetoacetate



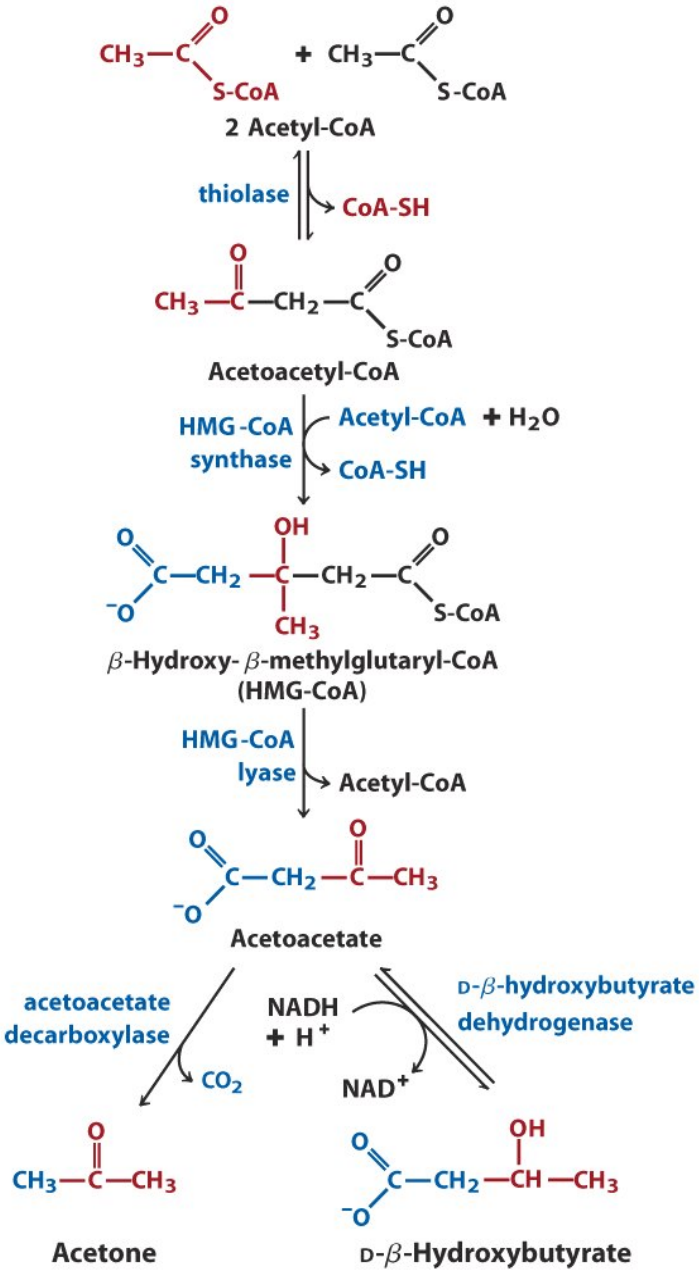
Acetone

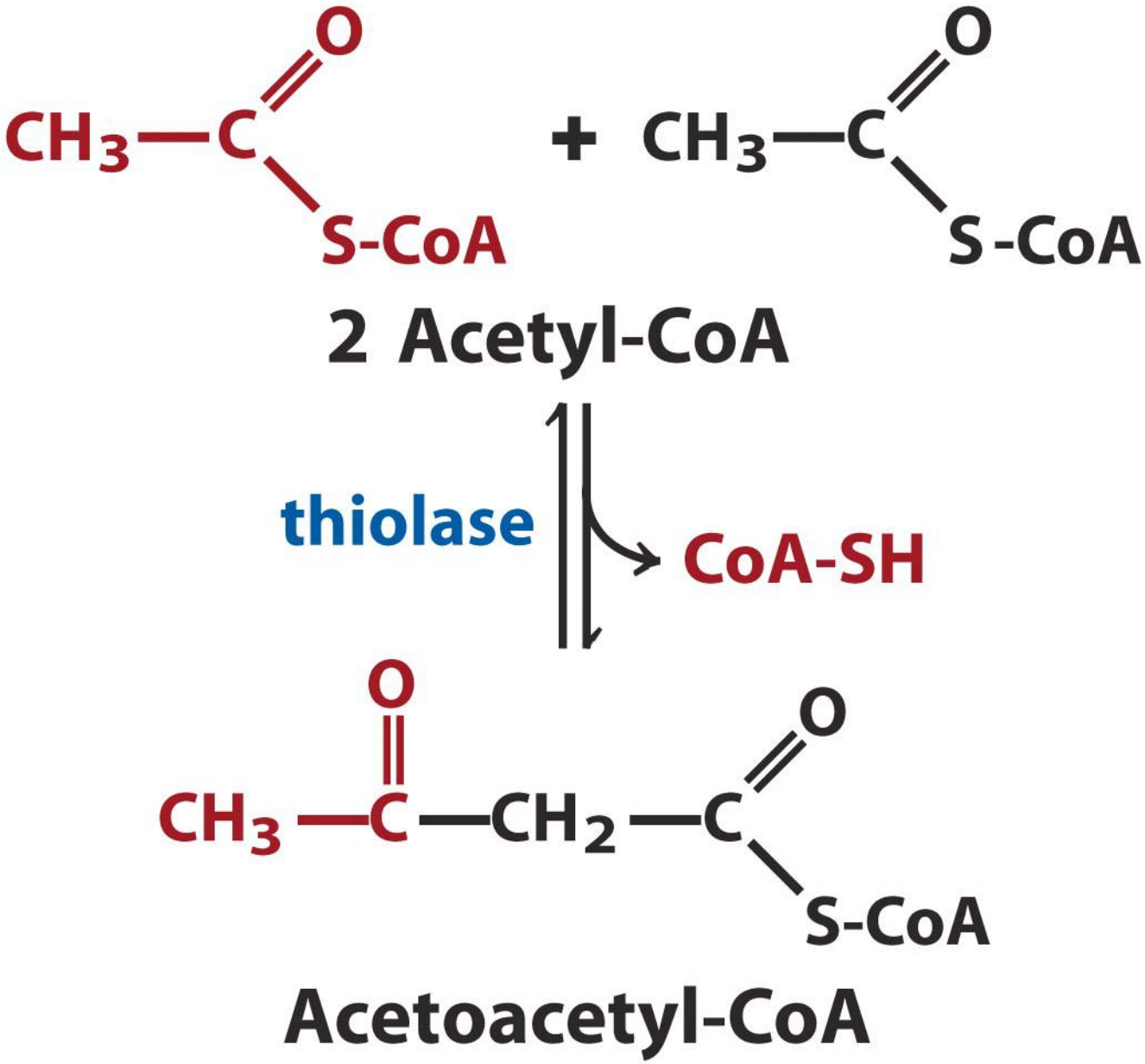


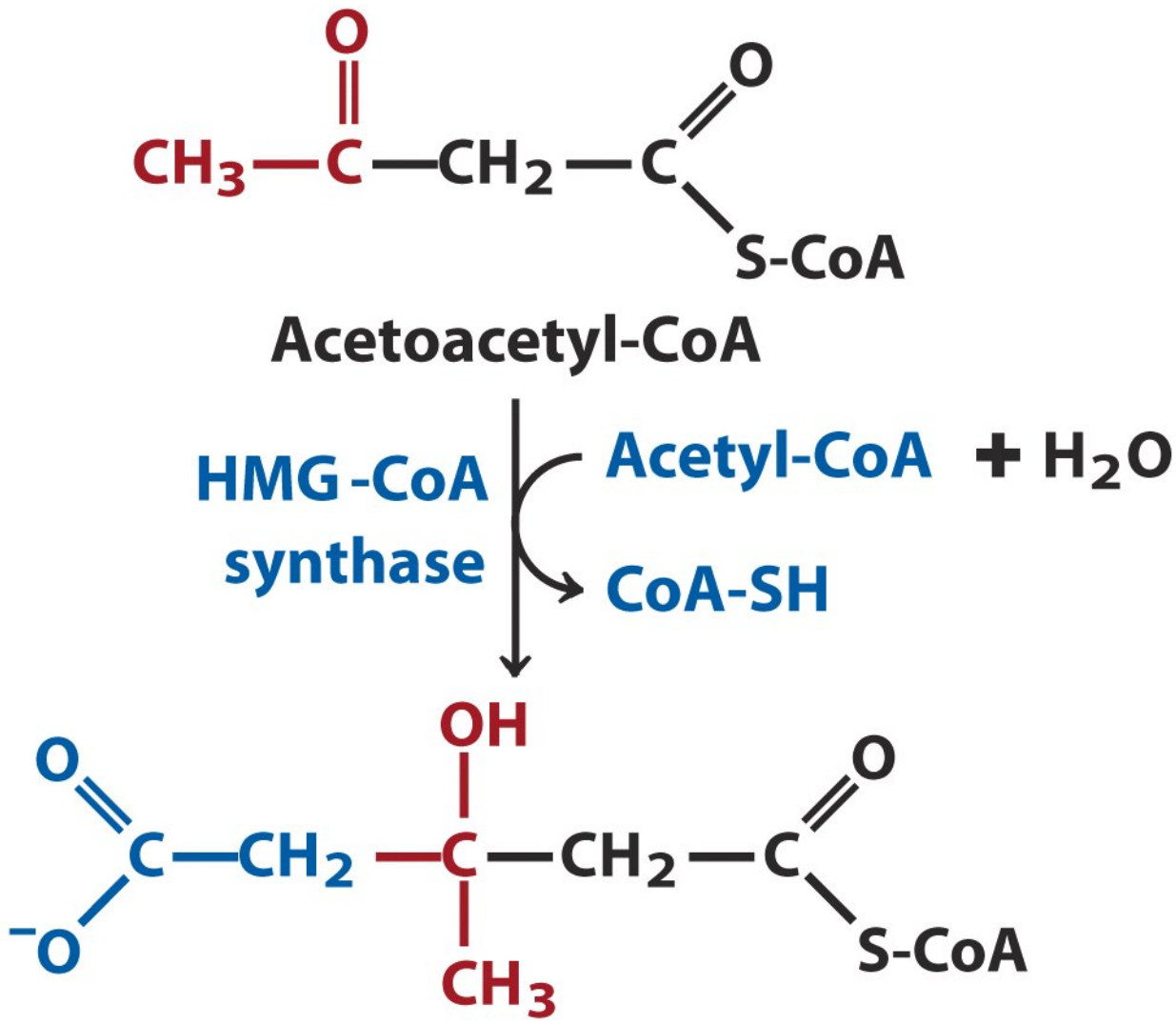
D-β-Hydroxybutyrate

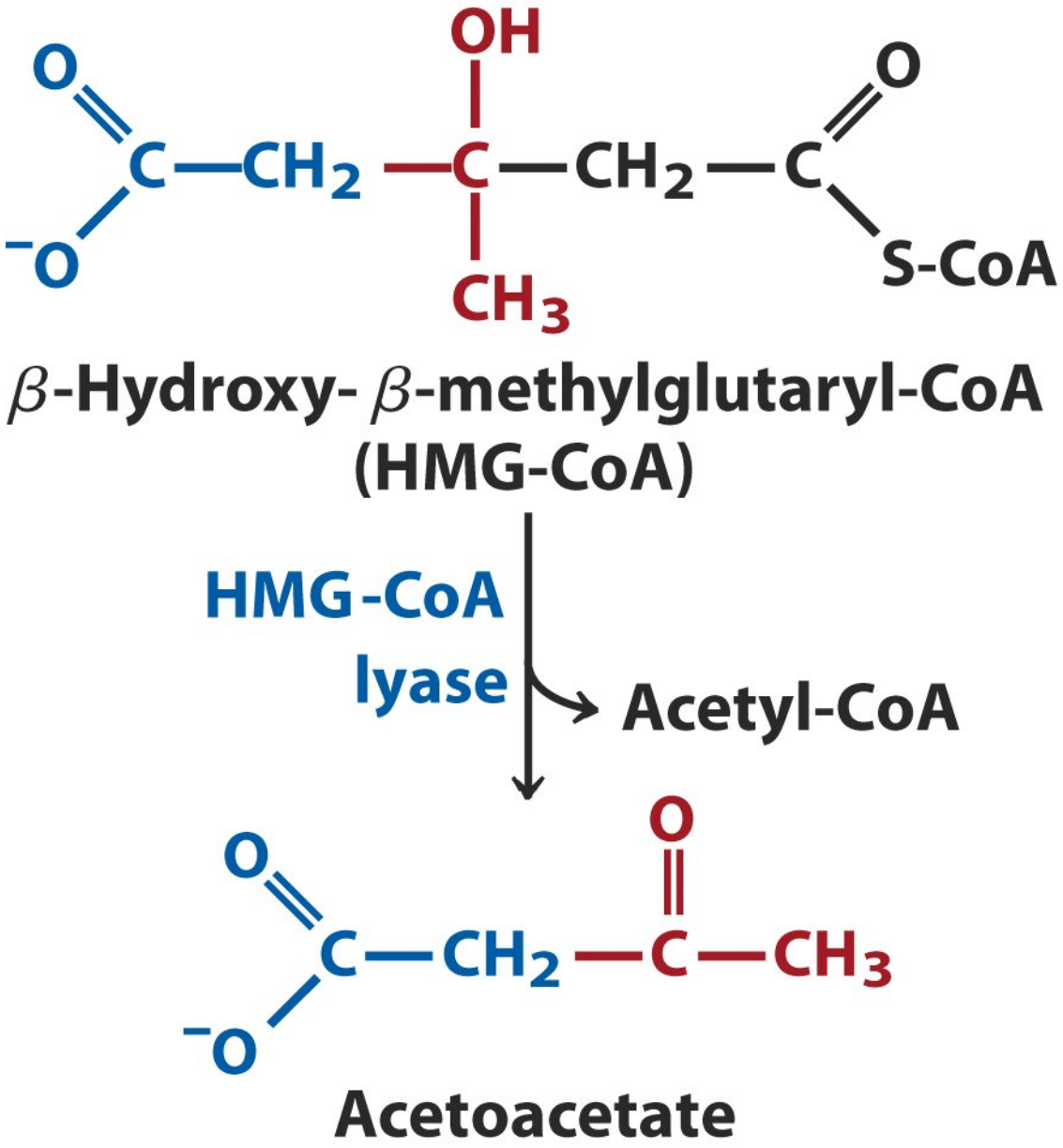
Formed in the liver (mitochondrial matrix) and is transported with the blood to other cells where it is used as fuel

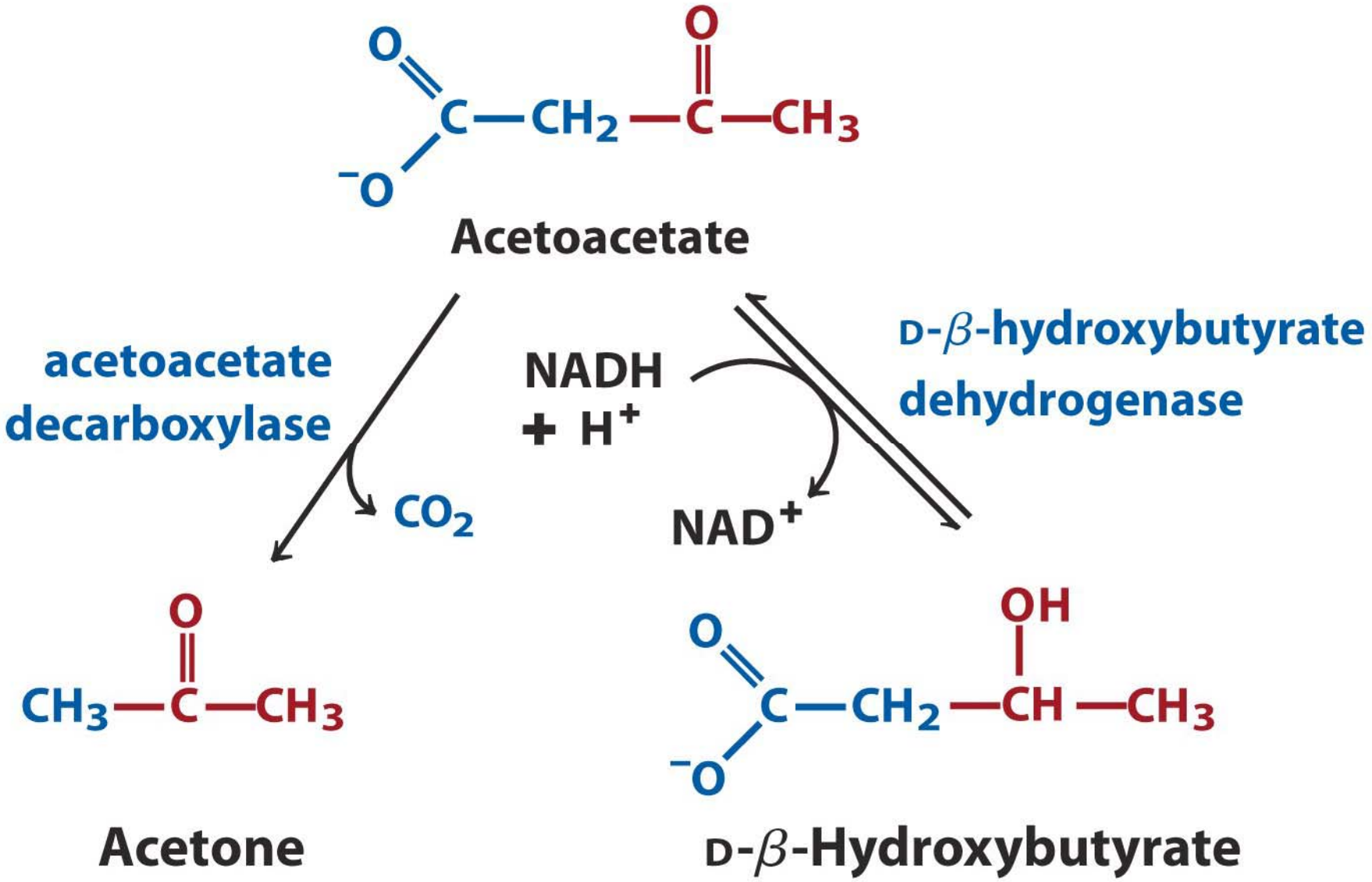
Ketone bodies are formed from a surplus of Ac-CoA




CH3
C
O
S-CoA
+
CH3
C
O
S-CoA
2 Acetyl-CoA
thiolase
CoA-SH
CH3
C
O
CH2
C
O
S-CoA
Acetoacetyl-CoA


O
CH3—C—CH2—C
O
S-CoA
Acetoacetyl-CoA
HMG-CoA synthase
Acetyl-CoA + H2O
CoA-SH
OH
O
C—CH2—C—CH2—C
O
⁻O
S-CoA
CH3
β-Hydroxy-β-methylglutaryl-CoA
(HMG-CoA)

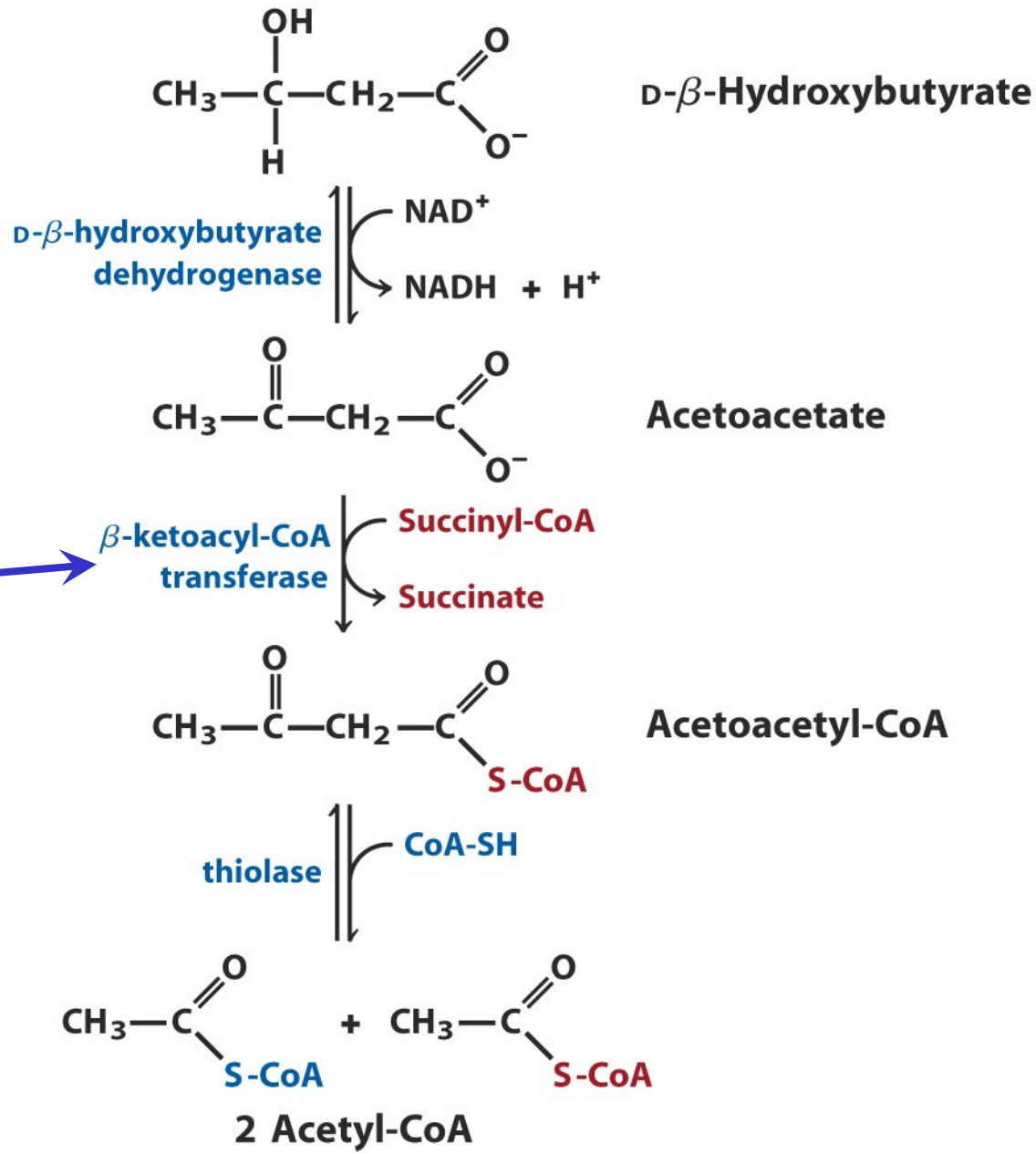

OH
O
C
CH2
C
CH2
C
O
S-CoA
−O
CH3
β-Hydroxy-β-methylglutaryl-CoA
(HMG-CoA)
HMG-CoA
lyase
Acetyl-CoA
O
O
C
CH2
C
CH3
−O
Acetoacetate


Acetoacetate
acetoacetate decarboxylase
CO_2
NADH + H^+
NAD^+
D-β-hydroxybutyrate dehydrogenase
Acetone
D-β-Hydroxybutyrate

Ketone bodies as energy sources

absent in the liver

citric acid cycle

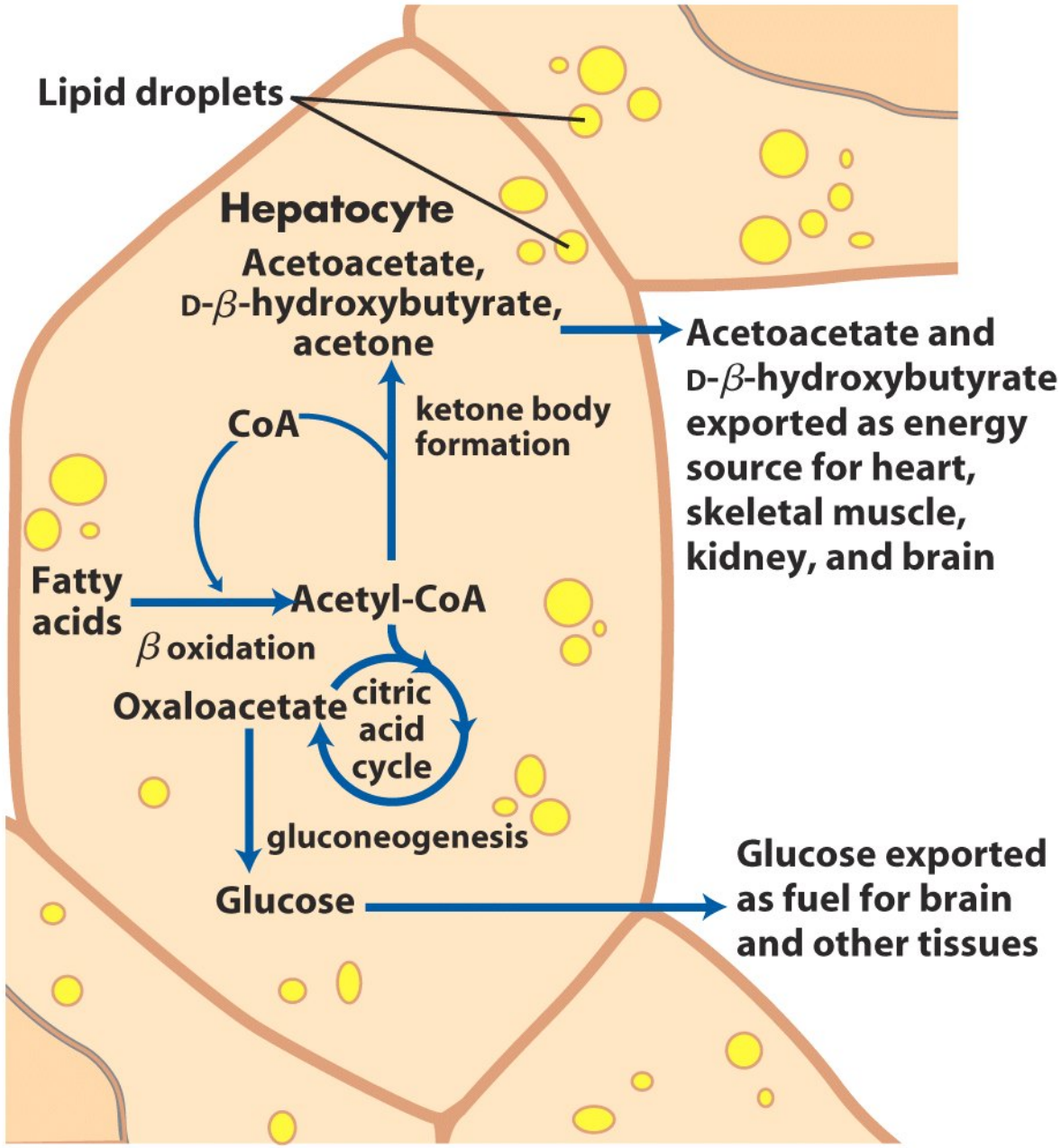


Functionality of ketone bodies

- **Water soluble**
  - **do not need lipoprotein or albumin**
- **Facilitate oxidation of FA even when Ac-CoA accumulates due to low activity in the citric acid cycle**
- **Liberate CoA when bound to Ac-CoA: FA oxidation can continue**
- **Important energy source for the brain under fasting conditions**

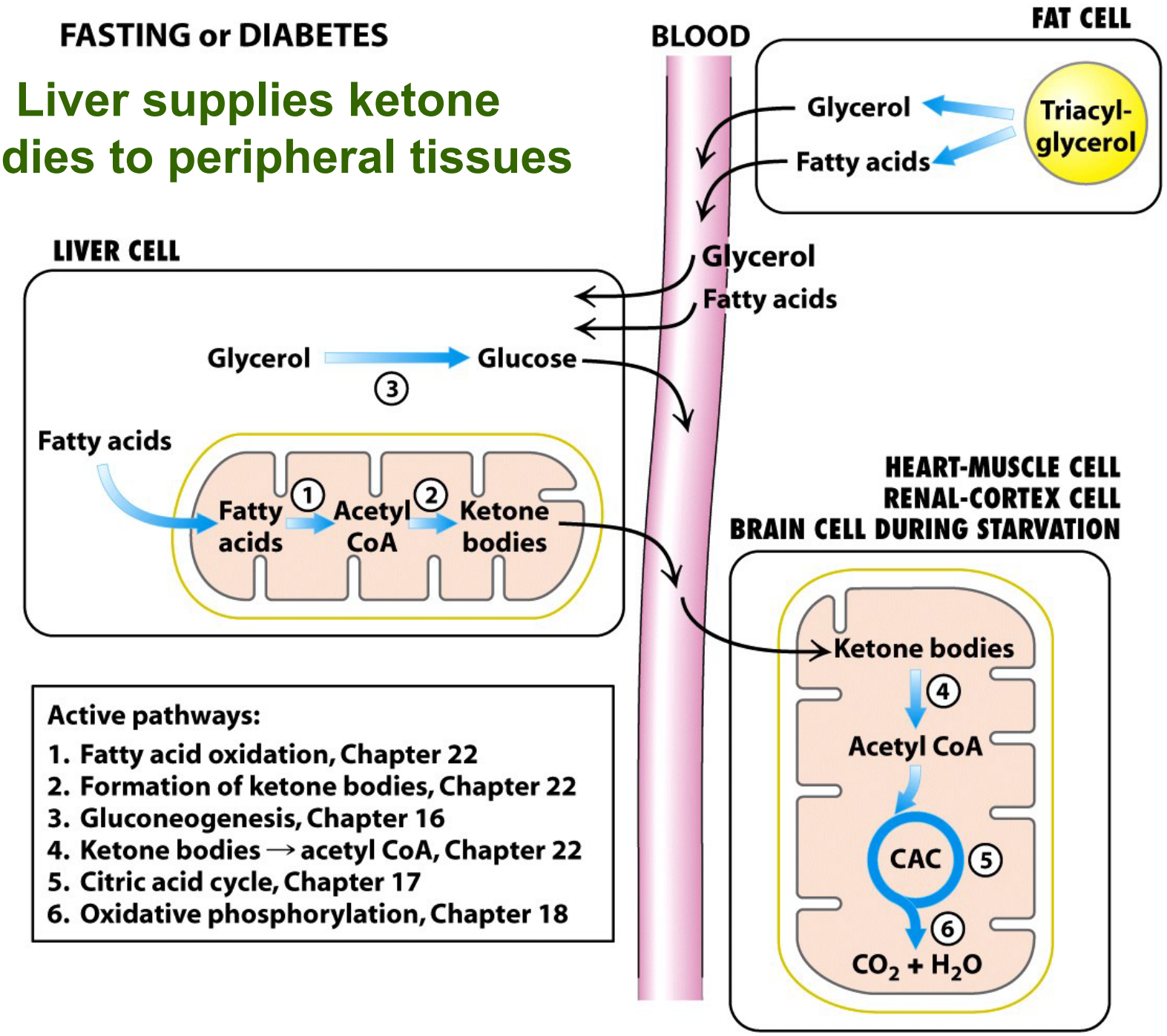
ketone bodies = water-soluble, transportable form of acetyl units

Fasting and diabetes lead to overproduction of ketone bodies



FASTING or DIABETES

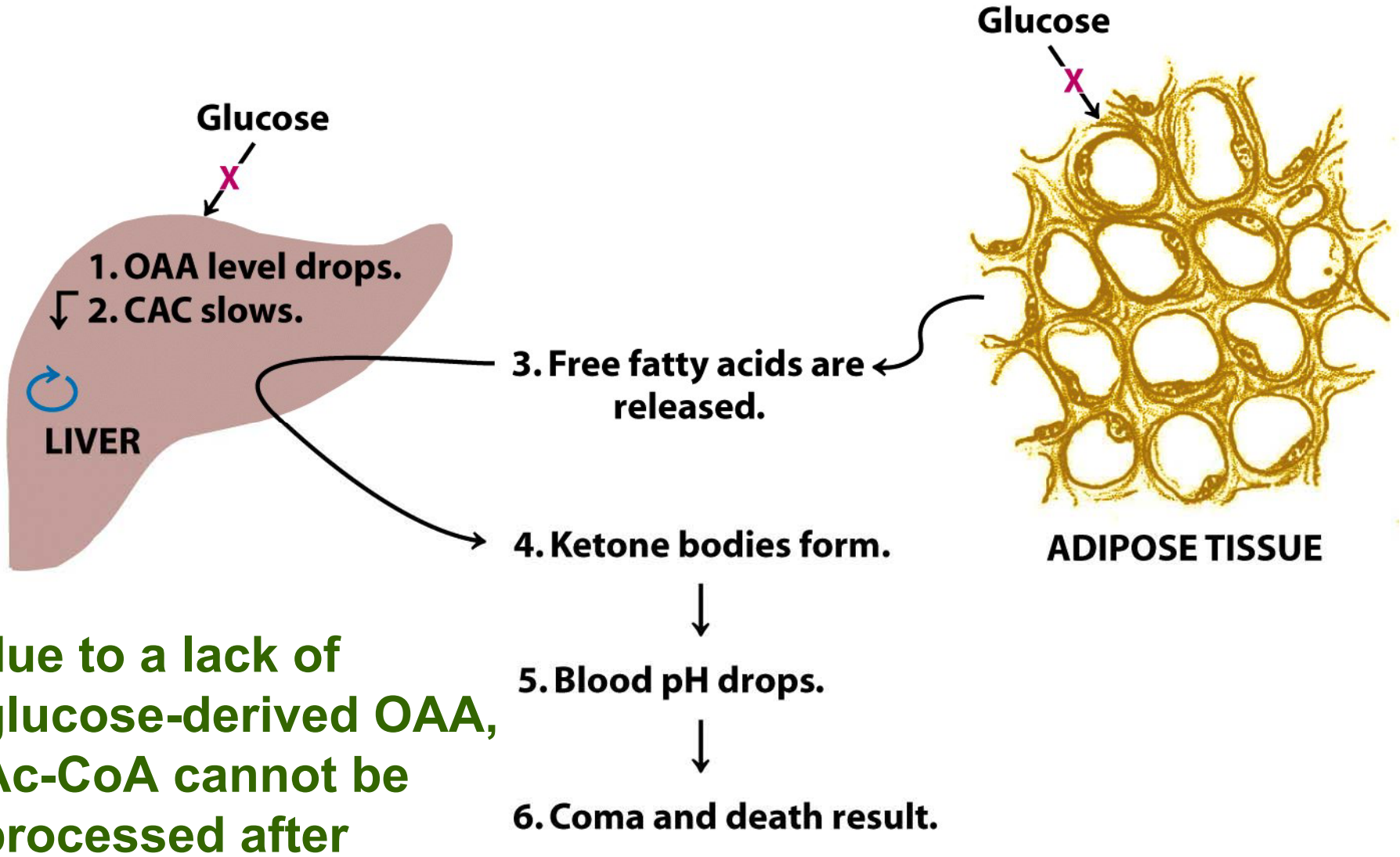
Liver supplies ketone bodies to peripheral tissues



Active pathways:

1. Fatty acid oxidation, Chapter 22
2. Formation of ketone bodies, Chapter 22
3. Gluconeogenesis, Chapter 16
4. Ketone bodies → acetyl CoA, Chapter 22
5. Citric acid cycle, Chapter 17
6. Oxidative phosphorylation, Chapter 18

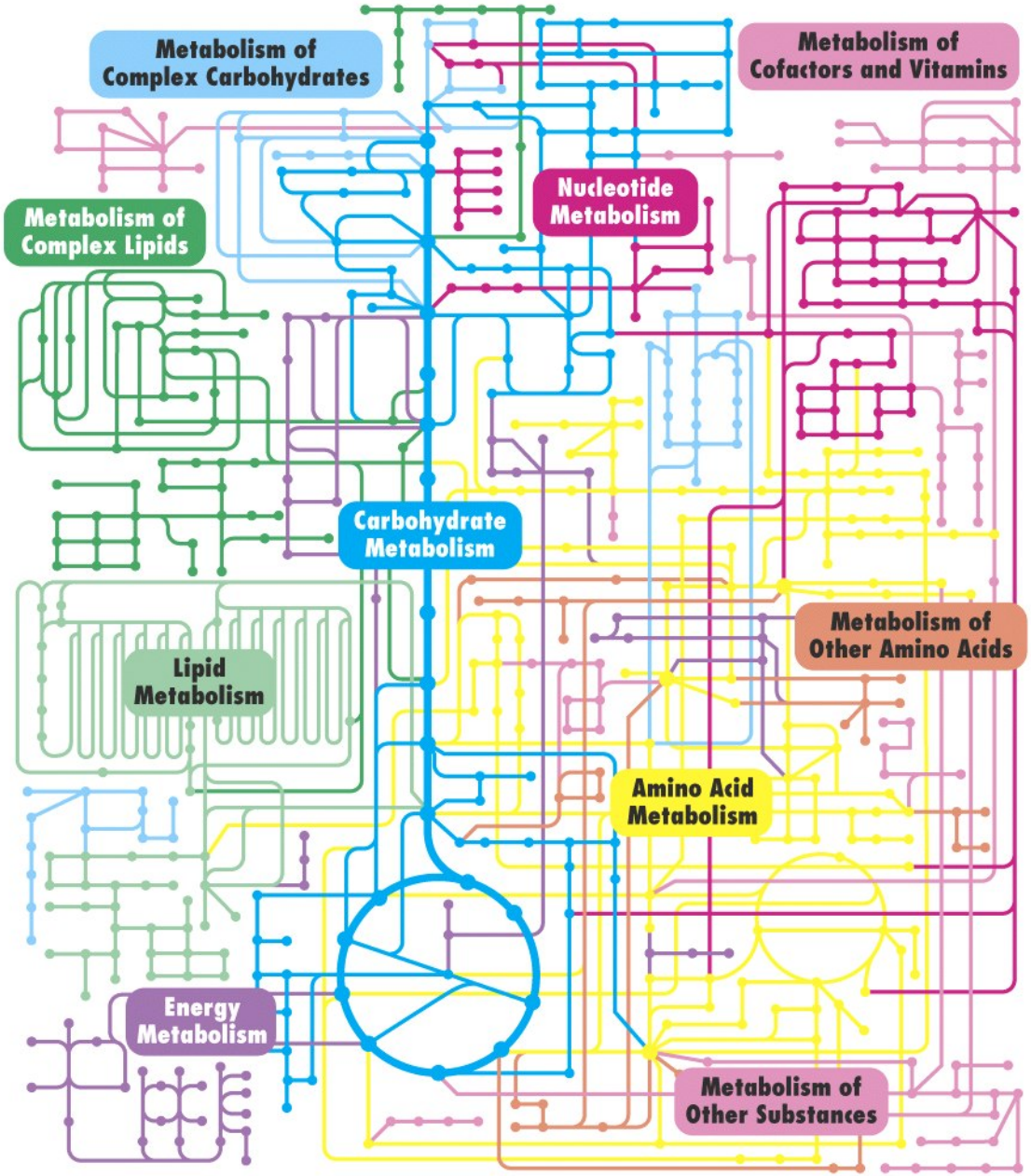
Diabetic ketosis results when insulin is absent



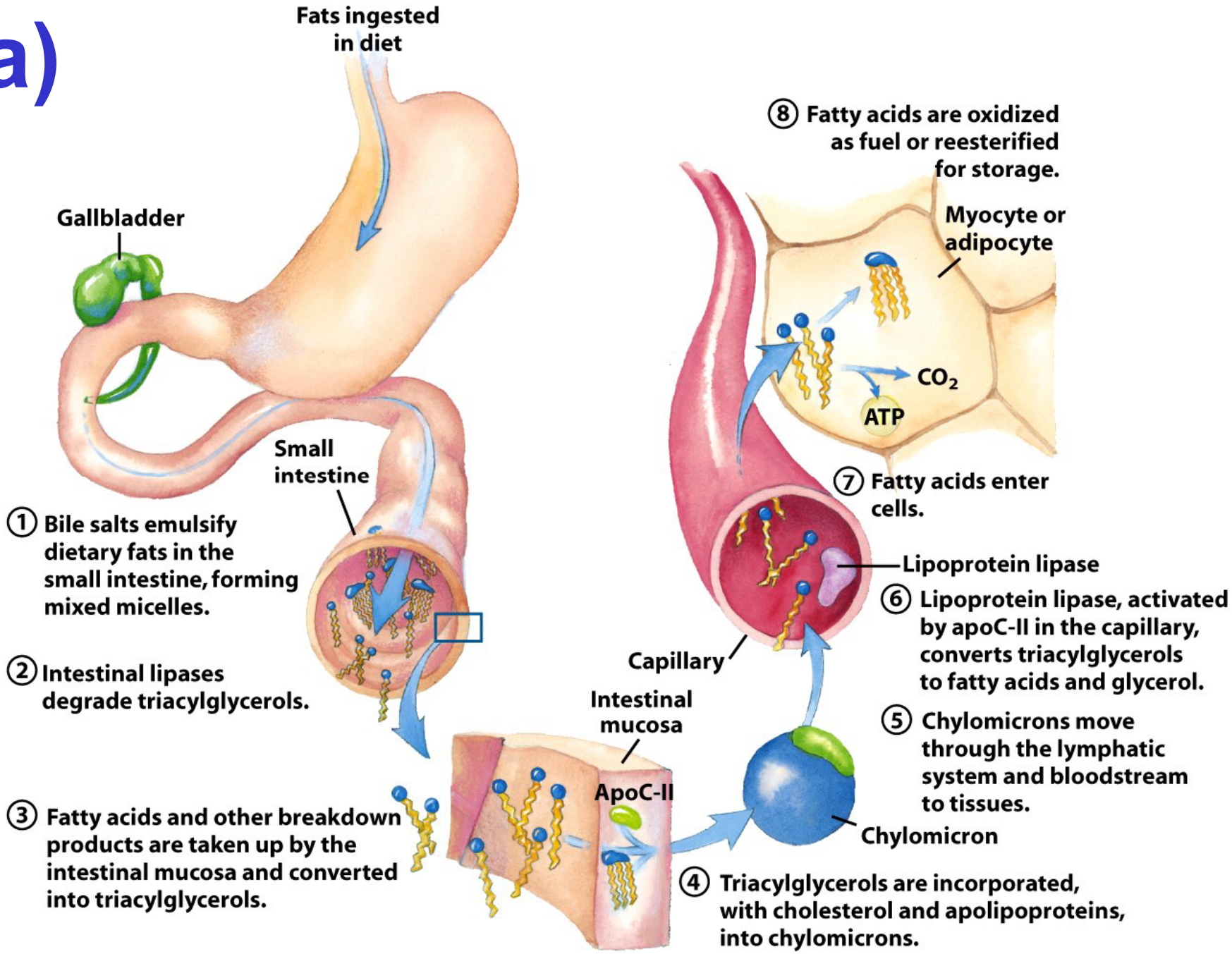
due to a lack of glucose-derived OAA, Ac-CoA cannot be processed after β-oxidation

OAA = oxaloacetate
CAC = citric acid cycle

So → What exactly have we learned?



a)


Fats ingested in diet
Gallbladder
Small intestine
① Bile salts emulsify dietary fats in the small intestine, forming mixed micelles.
② Intestinal lipases degrade triacylglycerols.
③ Fatty acids and other breakdown products are taken up by the intestinal mucosa and converted into triacylglycerols.
Intestinal mucosa
ApoC-II
④ Triacylglycerols are incorporated, with cholesterol and apolipoproteins, into chylomicrons.
⑤ Chylomicrons move through the lymphatic system and bloodstream to tissues.
Chylomicron
Capillary
⑥ Lipoprotein lipase, activated by apoC-II in the capillary, converts triacylglycerols to fatty acids and glycerol.
Lipoprotein lipase
⑦ Fatty acids enter cells.
⑧ Fatty acids are oxidized as fuel or reesterified for storage.
Myocyte or adipocyte
CO_2
ATP

Lipid Catabolism Summary

- TAGs are highly concentrated energy depots
- Lipases release FA from TAG
- Chylomicrons transport FA from small intestine to peripheral tissues
- FAs are synthesized and degraded by different pathways – essentially opposite
- Use of FAs as fuel requires 3 stages (catabolism)
- Oxidation of C16 FA = 106 ATPs
- Unsaturated and odd chain FAs require additional steps for degradation (isomerase, reductase)
- Ketone bodies are water-soluble transporters of Ac-CoA
- Ketone bodies supplied as energy during fasting

Useful links . . .

- http://www.cyberlipid.org/
- http://www.lipidlibrary.co.uk/
- http://www.lipidmaps.org/
- http://www.metabolomics.se/
  - (contains downloadable file of today's lecture under the section "Courses")