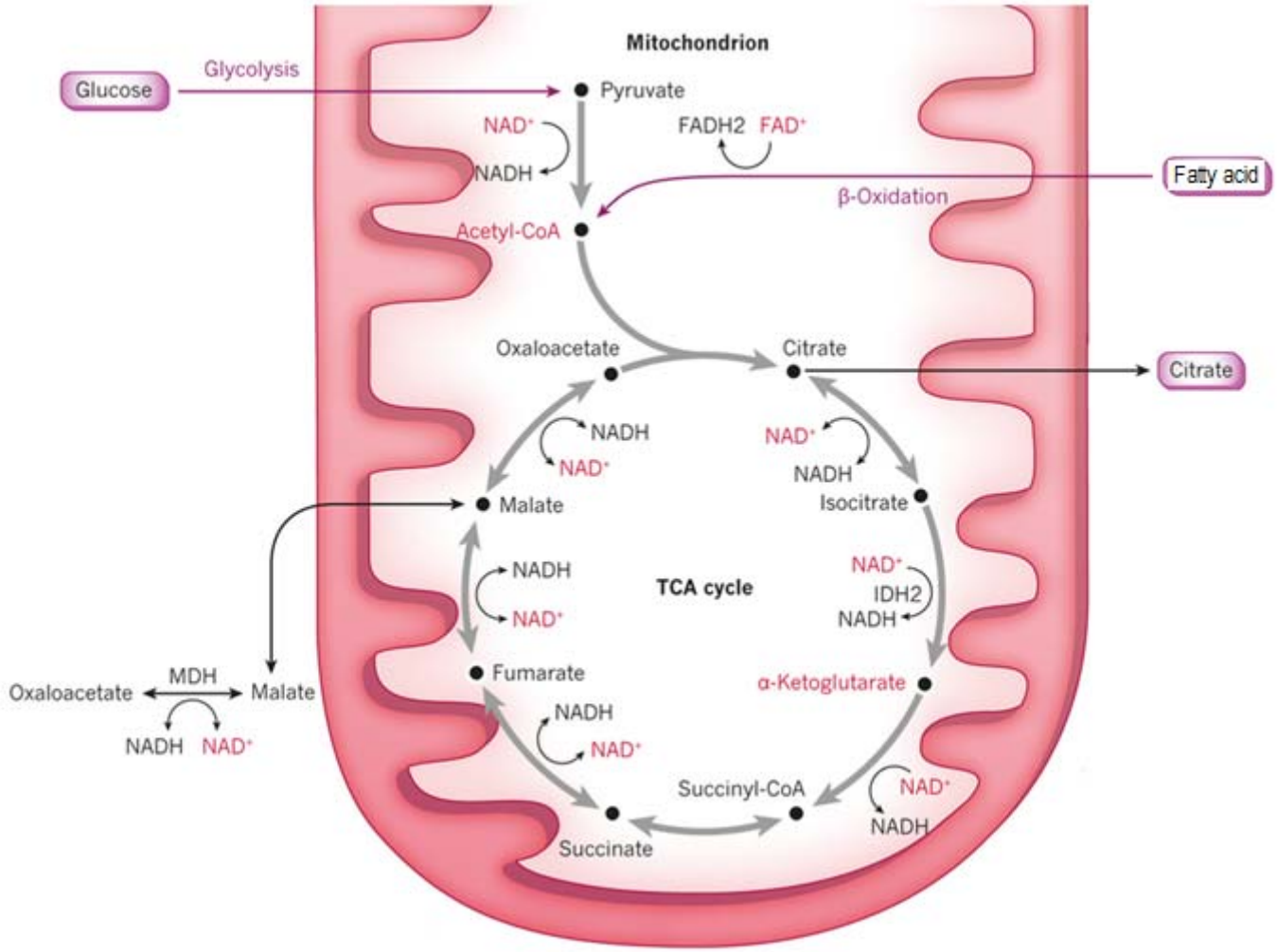
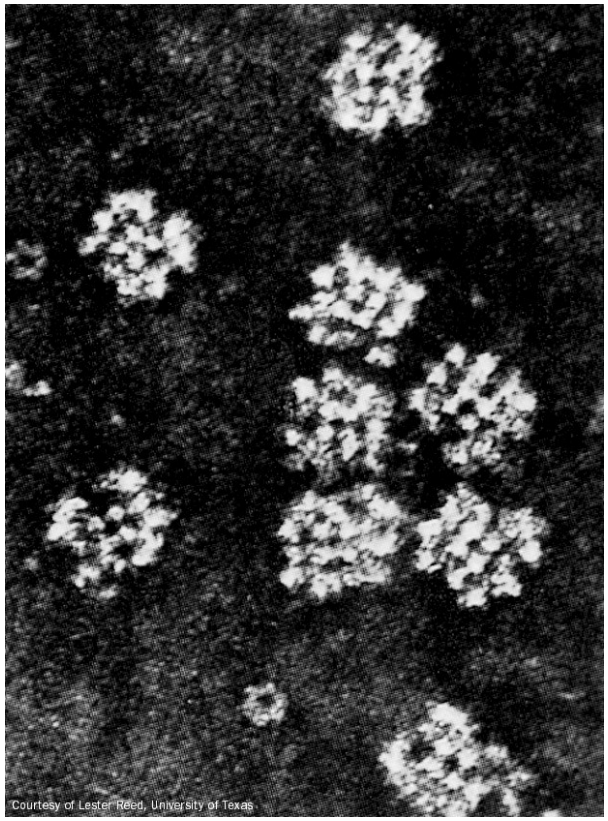
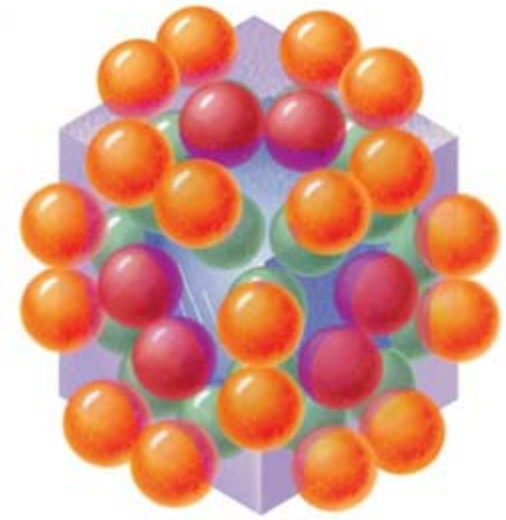
Metabolism II



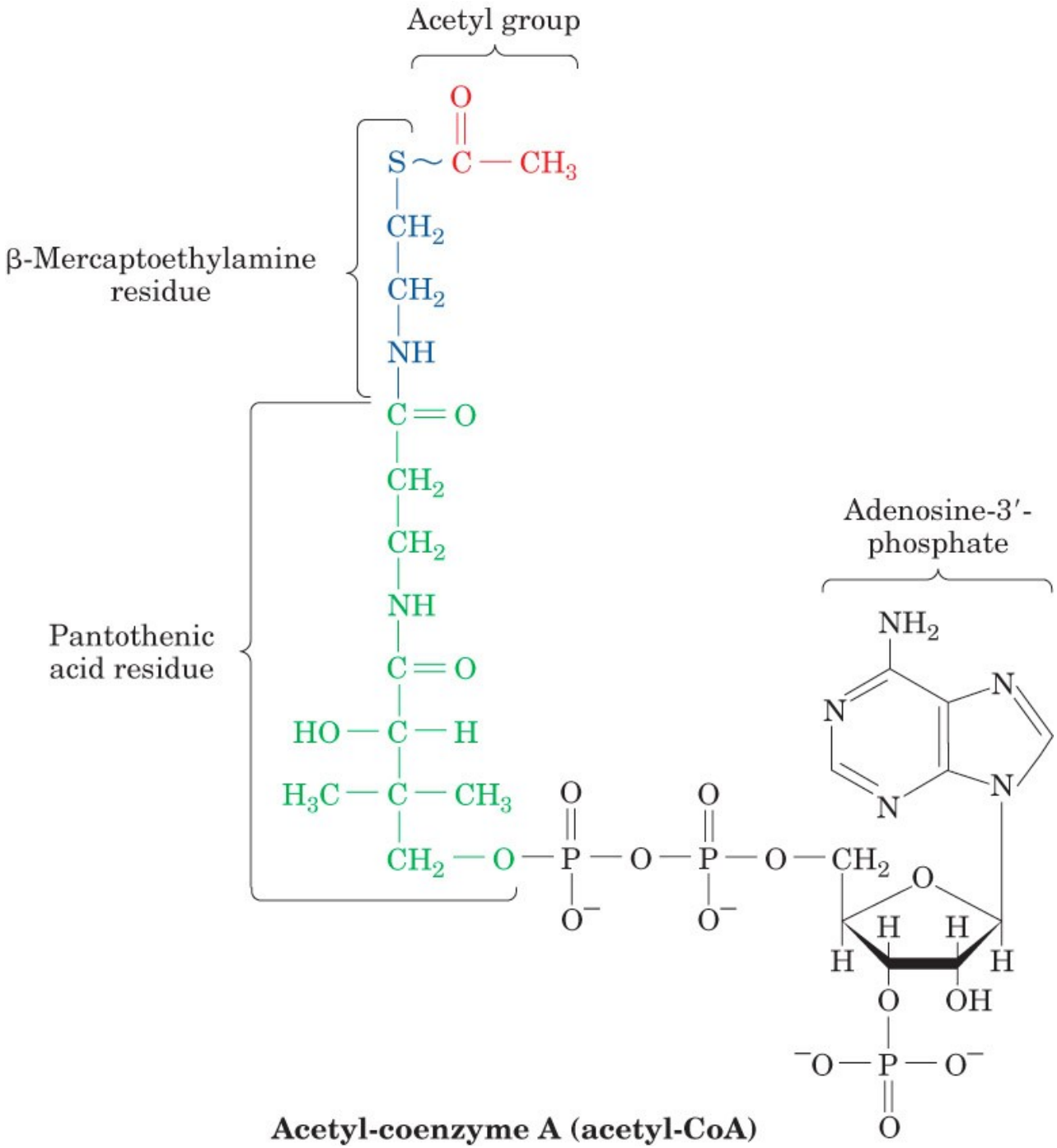
Aim: understand the principles of TCA cycle, electron transport chain, oxidative phosphorylation, metabolism of fatty acid

$$\text{Pyruvate} + \text{CoA} + \text{NAD}^+ \xrightarrow{\text{PDC}} \text{Acetyl-CoA} + \text{CO}_2 + \text{NADH} + \text{H}^+$$

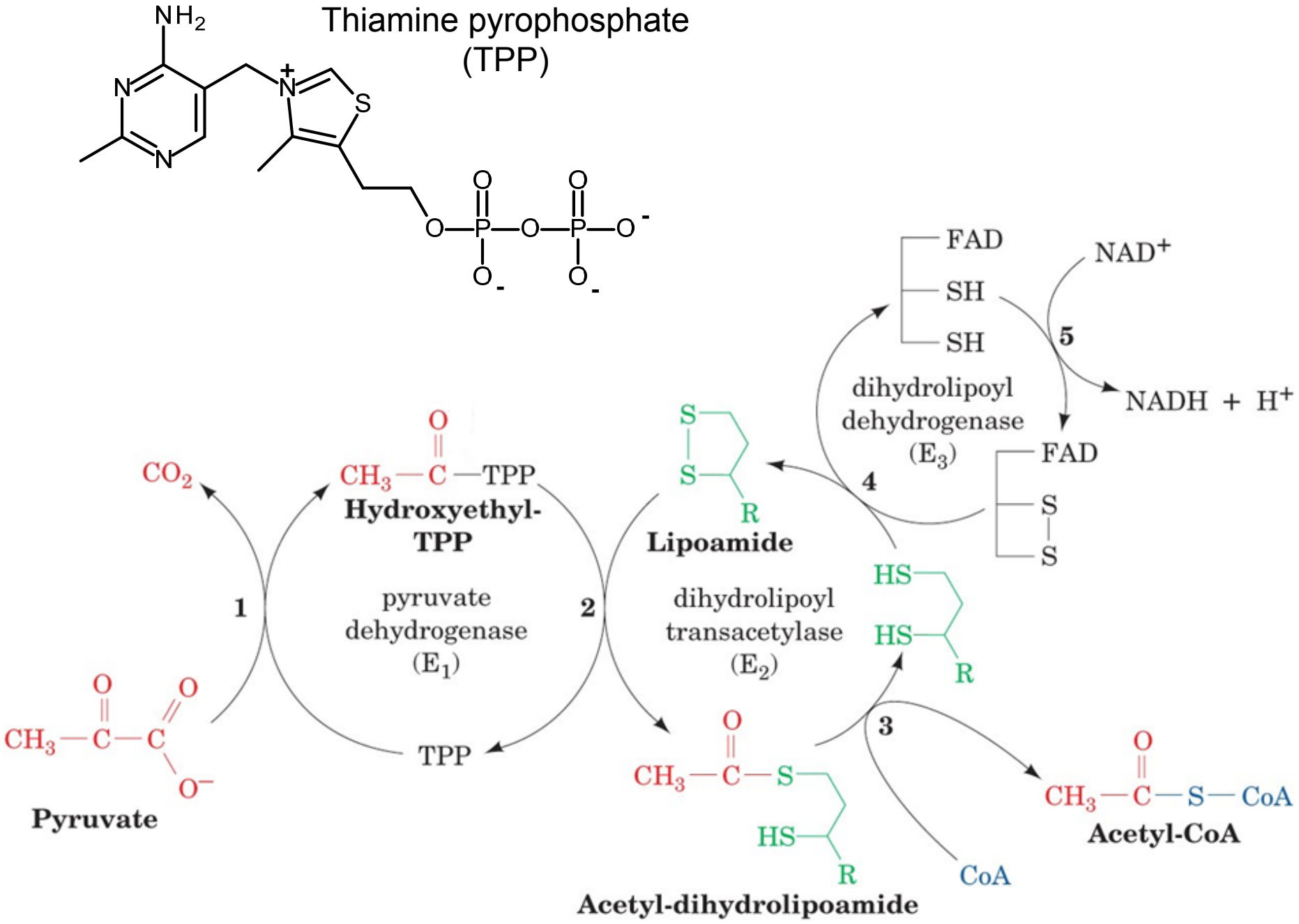




Pyruvate dehydrogenase complex (PDC)

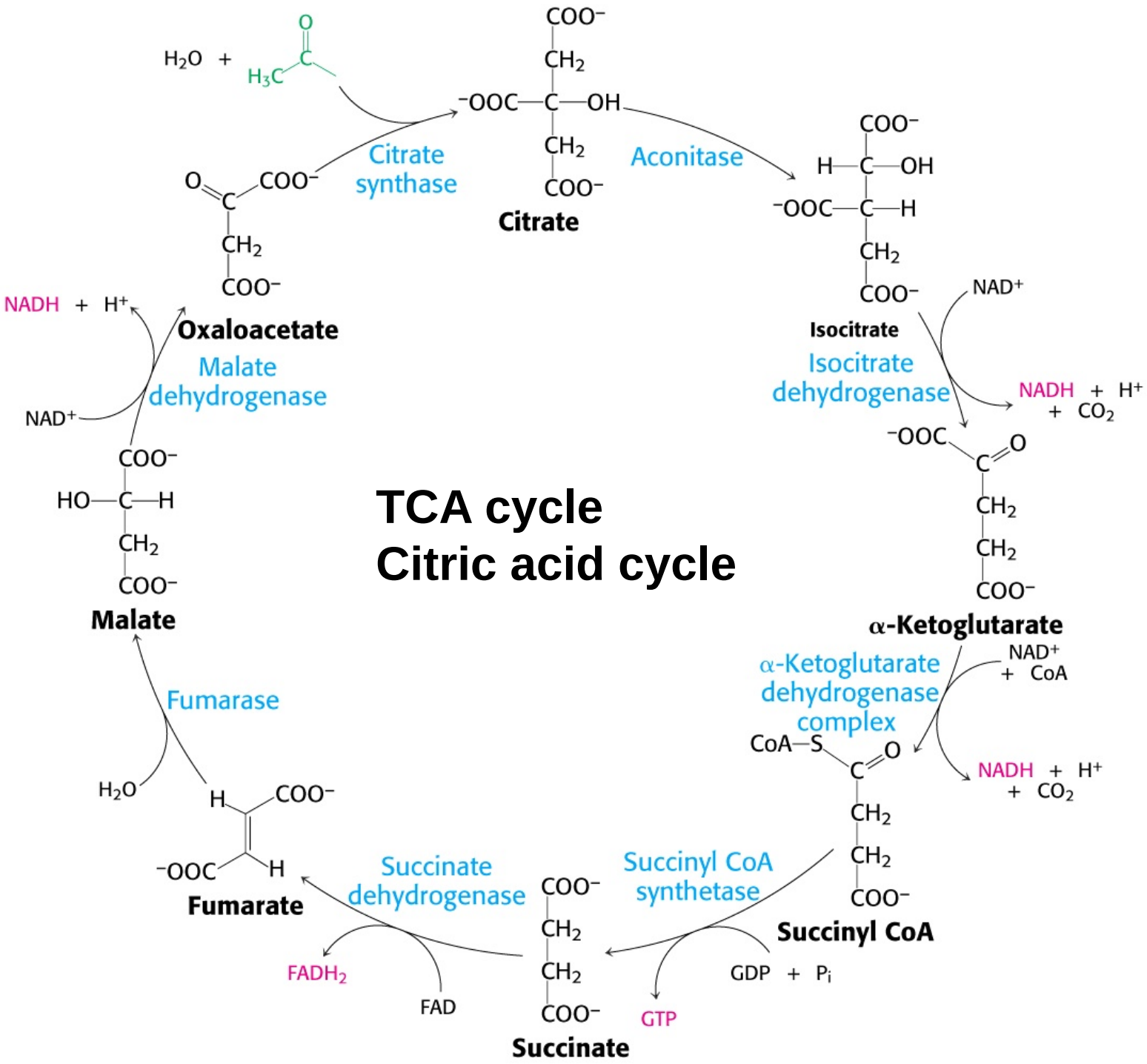


Acetyl-coenzyme A (acetyl-CoA)


Thiamine pyrophosphate
(TPP)
Pyruvate
CO2
Hydroxyethyl-
TPP
pyruvate
dehydrogenase
(E1)
1
TPP
Lipoamide
dihydrolipoyl
transacetylase
(E2)
2
Acetyl-dihydrolipoamide
CoA
3
Acetyl-CoA
FAD
SH
SH
dihydrolipoyl
dehydrogenase
(E3)
4
FAD
NAD+
5
NADH + H+

Tricarboxylic acid cycle (TCA cycle)

- What are the functions of TCA cycle?
- What are the reactions of TCA cycle?
- How does mitochondrion look like?

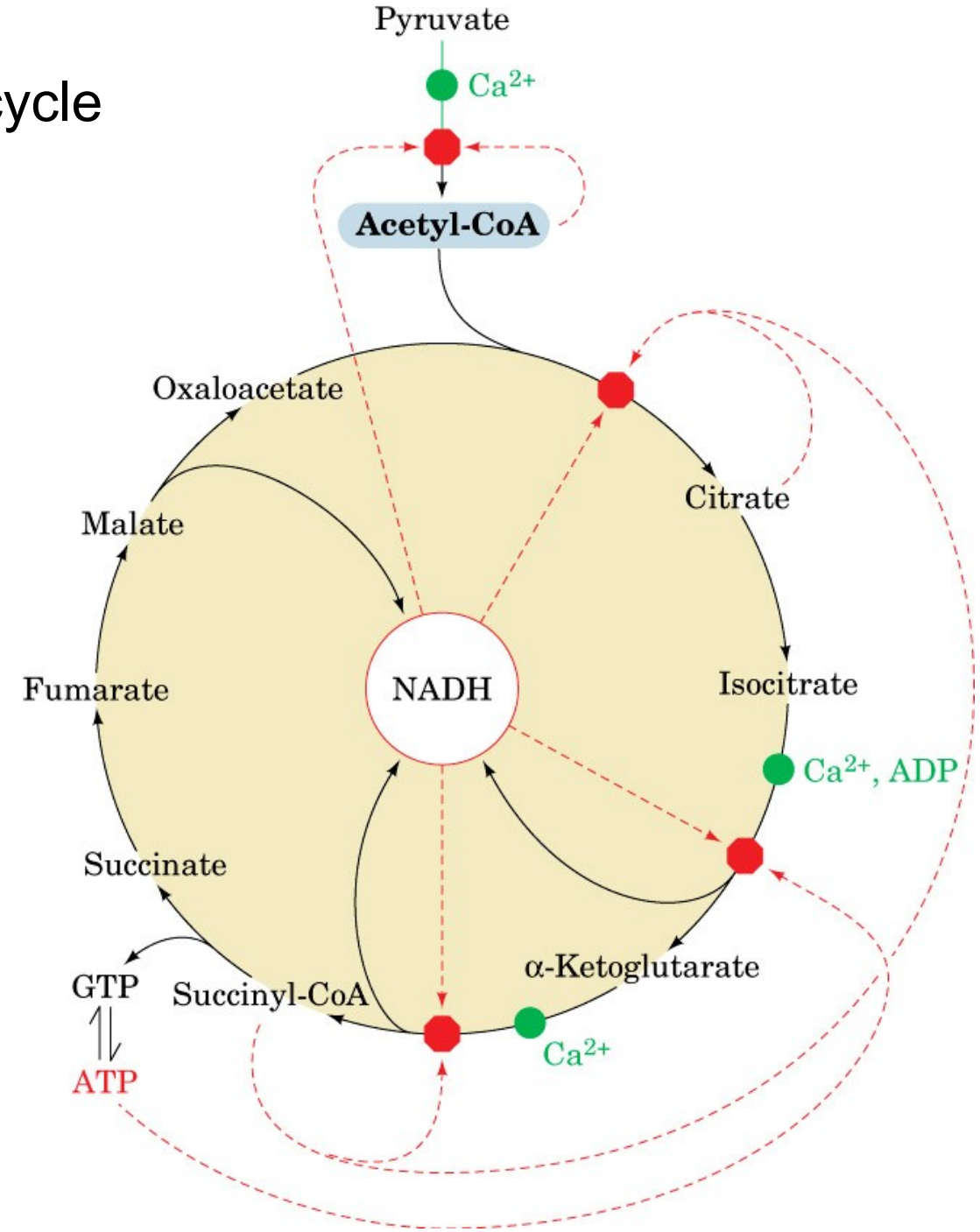

TCA cycle
Citric acid cycle
Citrate
Aconitase
Isocitrate
Isocitrate dehydrogenase
α-Ketoglutarate
α-Ketoglutarate dehydrogenase complex
Succinyl CoA
Succinyl CoA synthetase
Succinate
Succinate dehydrogenase
Fumarase
Fumarate
Malate
Malate dehydrogenase
Oxaloacetate
Citrate synthase

Summary

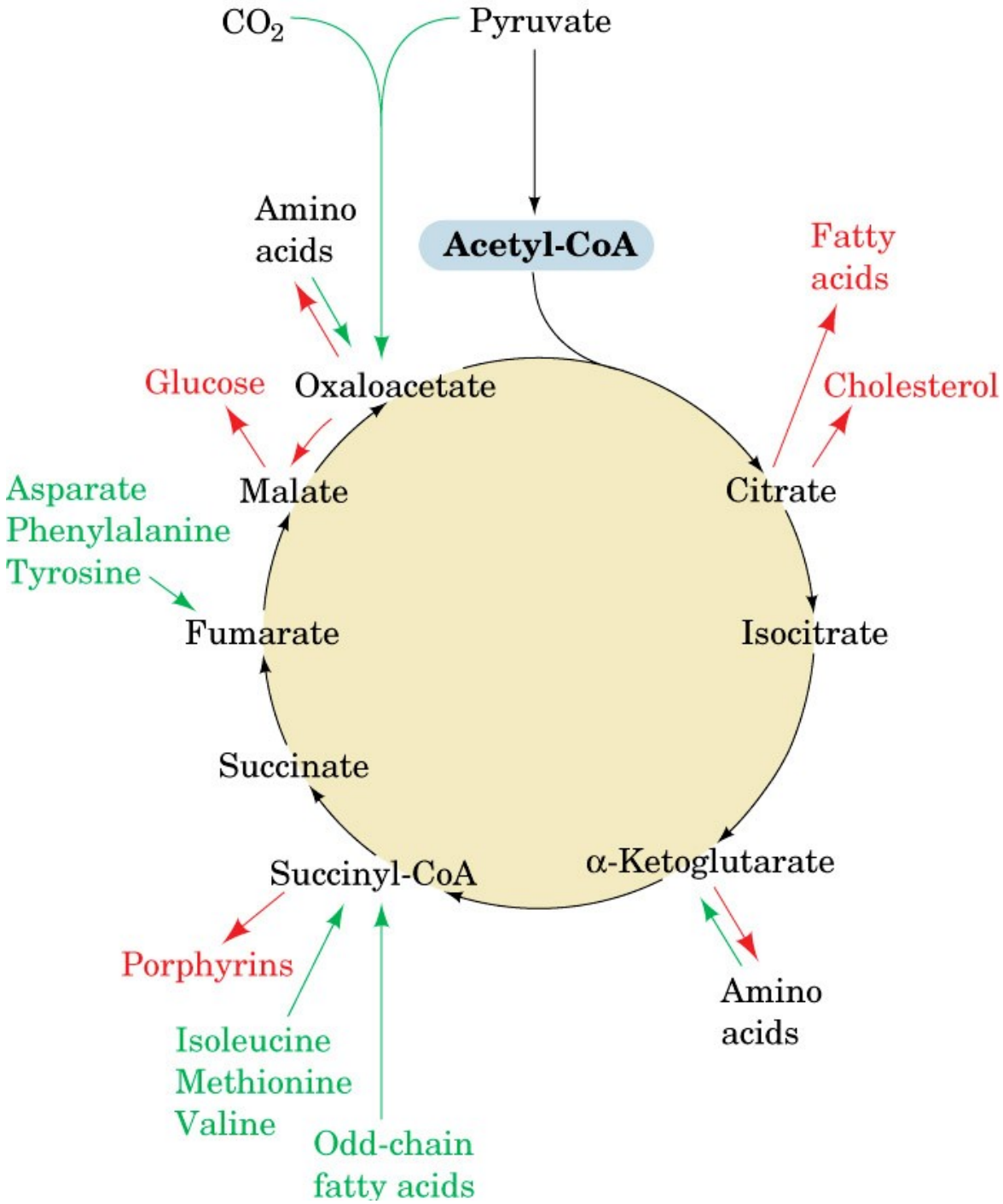
For each acetyl-CoA molecule oxidized, TCA cycle generates:

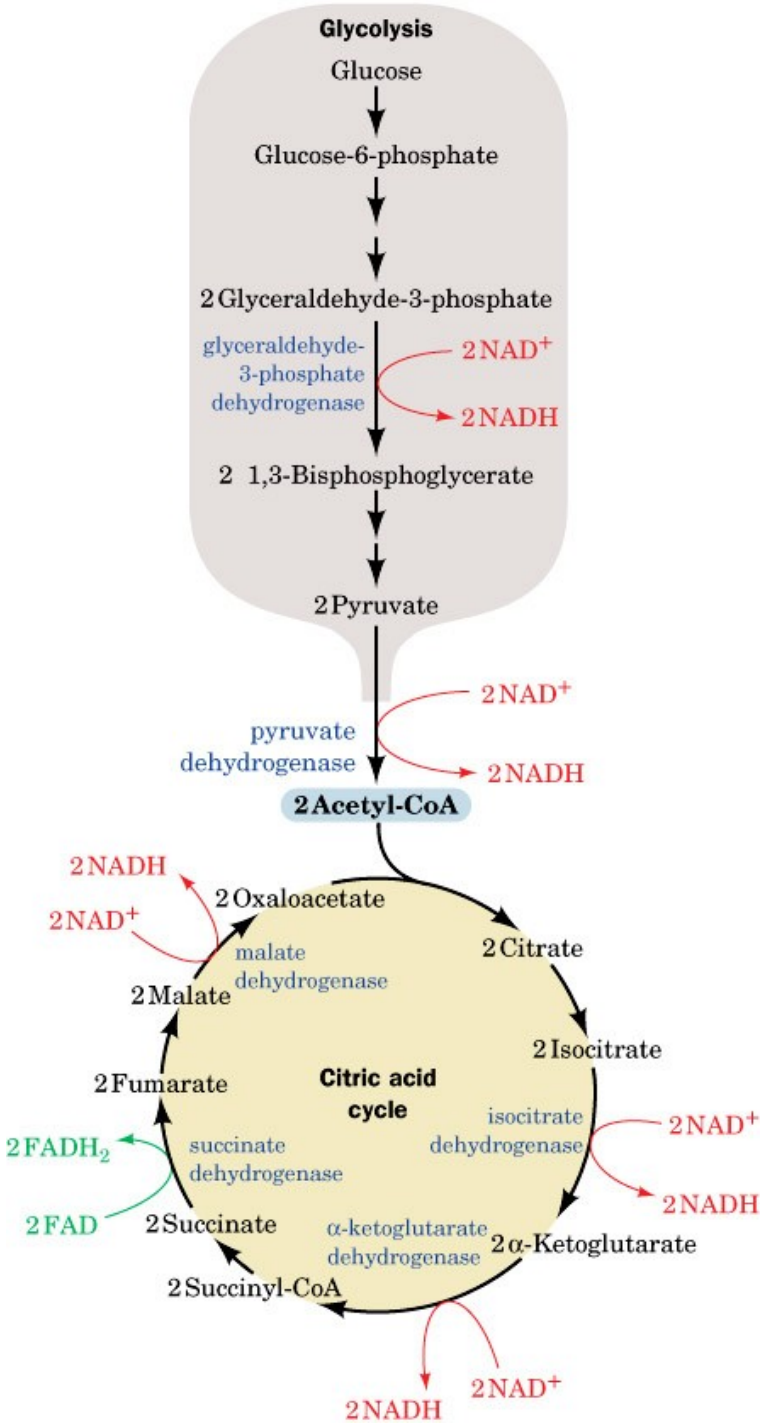
- 2 molecules of CO_2
- 3 molecules of NADH
- one $FADH_2$
- one GTP

Regulation of TCA cycle



Amphibolic function of TCA

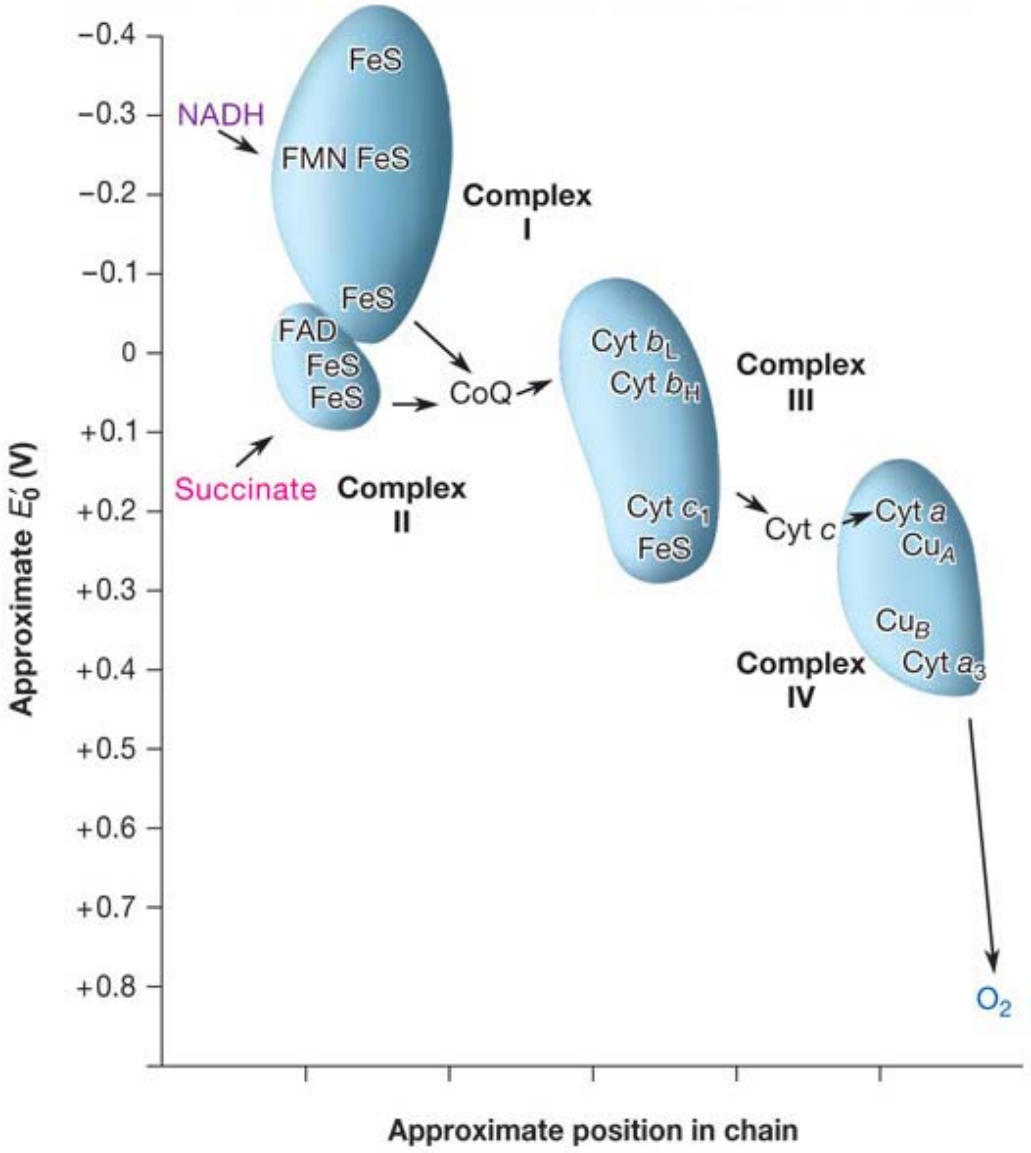



Glycolysis
Glucose
Glucose-6-phosphate
2 Glyceraldehyde-3-phosphate
glyceraldehyde-3-phosphate dehydrogenase
2 NAD+
2 NADH
2 1,3-Bisphosphoglycerate
2 Pyruvate
pyruvate dehydrogenase
2 NAD+
2 NADH
2 Acetyl-CoA
2 Oxaloacetate
2 NADH
2 NAD+
malate dehydrogenase
2 Malate
2 Citrate
2 Isocitrate
Citric acid cycle
isocitrate dehydrogenase
2 NAD+
2 NADH
2 α-Ketoglutarate
α-ketoglutarate dehydrogenase
2 Succinyl-CoA
2 NADH
2 NAD+
2 Succinate
2 FADH2
succinate dehydrogenase
2 FAD
2 Fumarate

Electron Transport Chain (ETC)

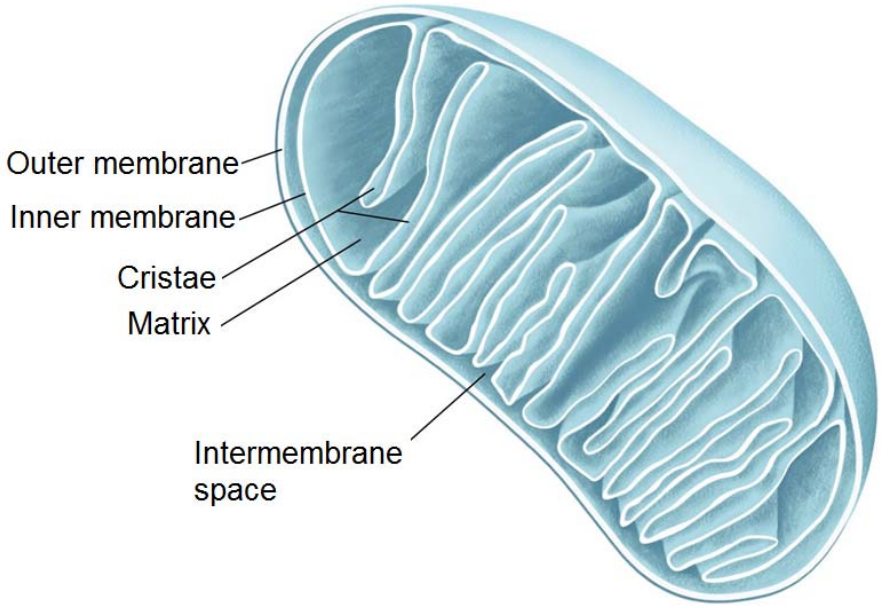
Series of electron carriers that operate together to transfer electrons from NADH and $FADH_2$ to a terminal electron acceptor

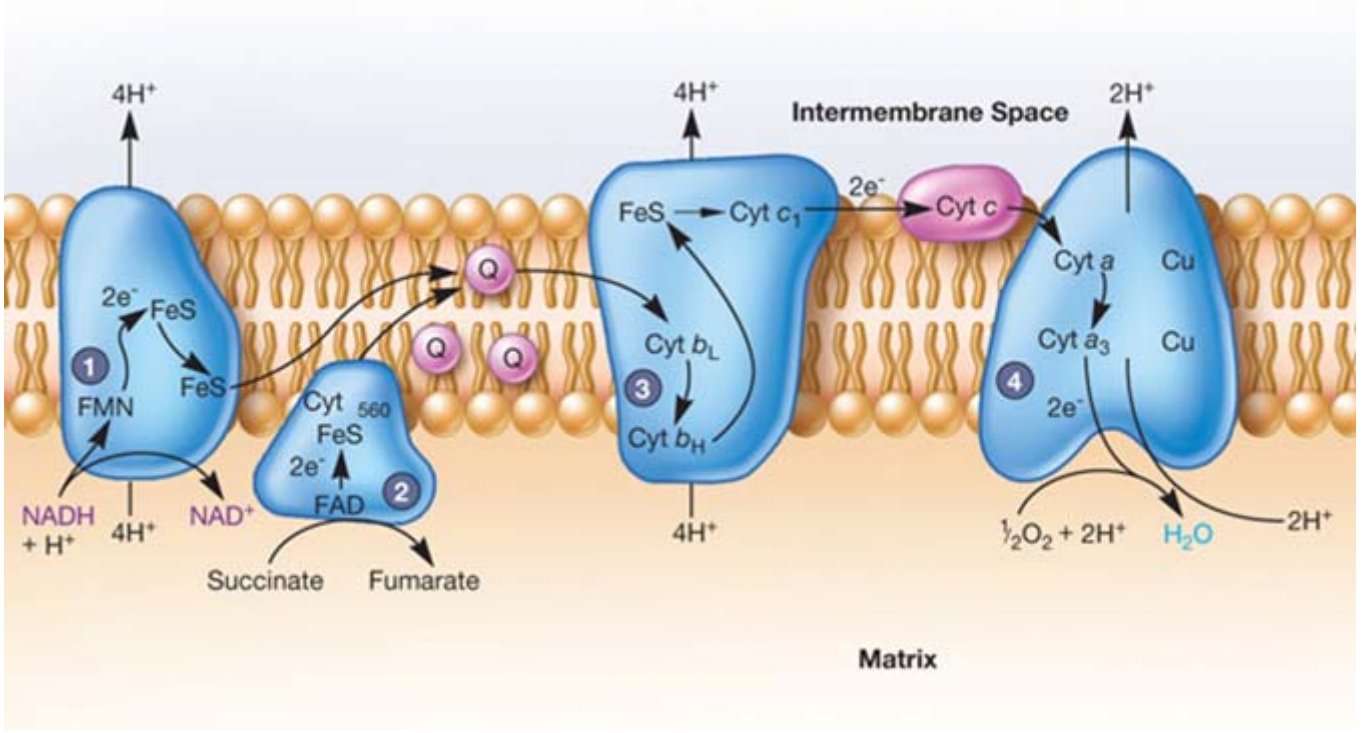
Electrons flow from carriers with more negative E_0 to carriers with more positive E_0

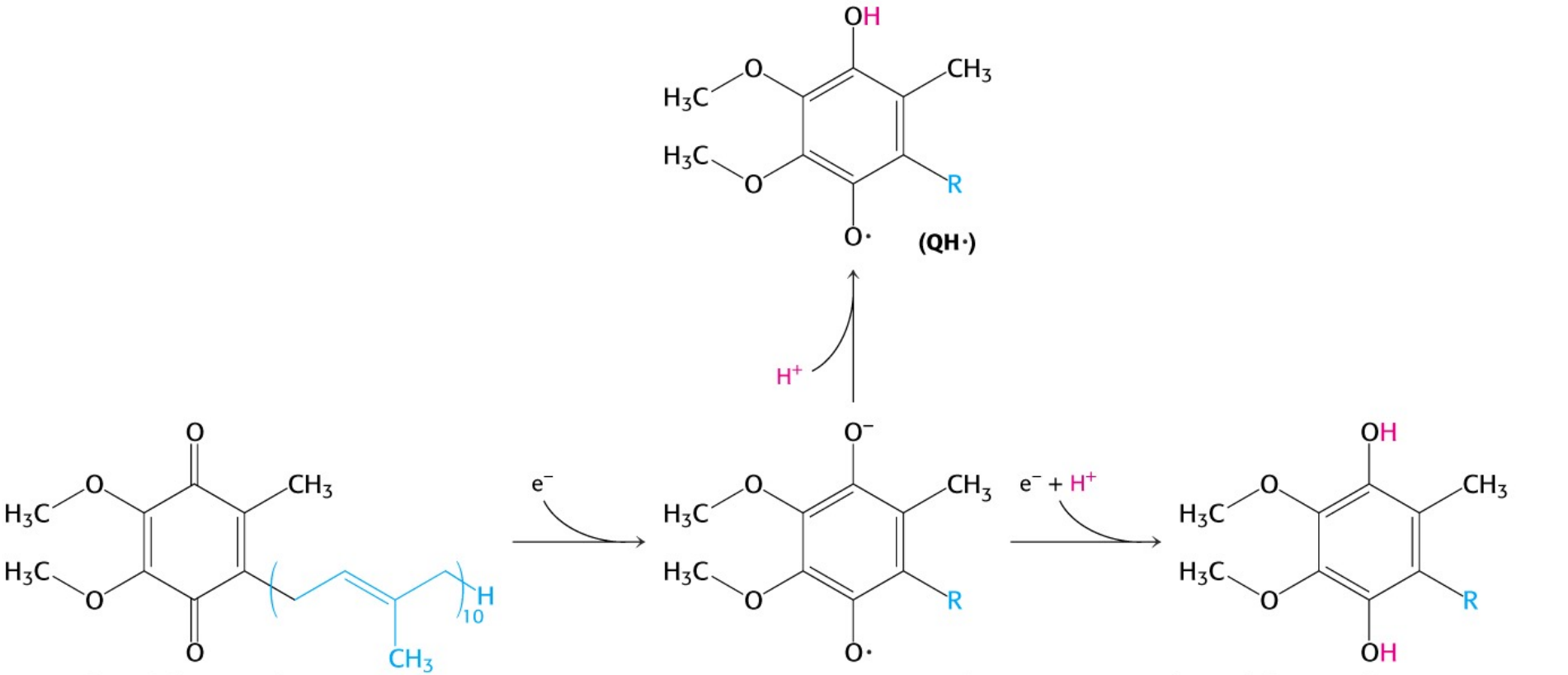


As electrons are transferred, energy is released.

In eucaryotes, ETC is located in the inner mitochrondrial membrane



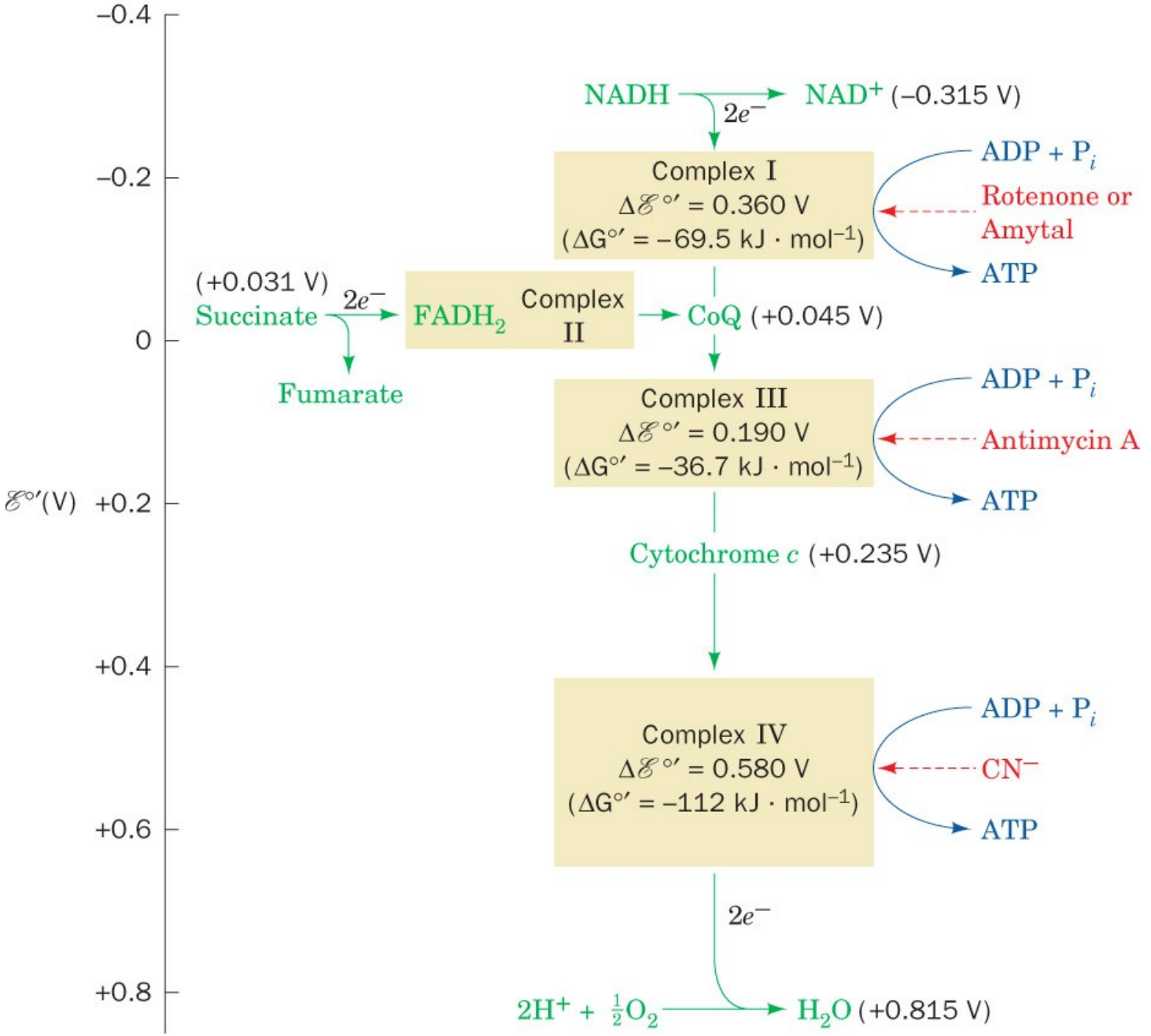




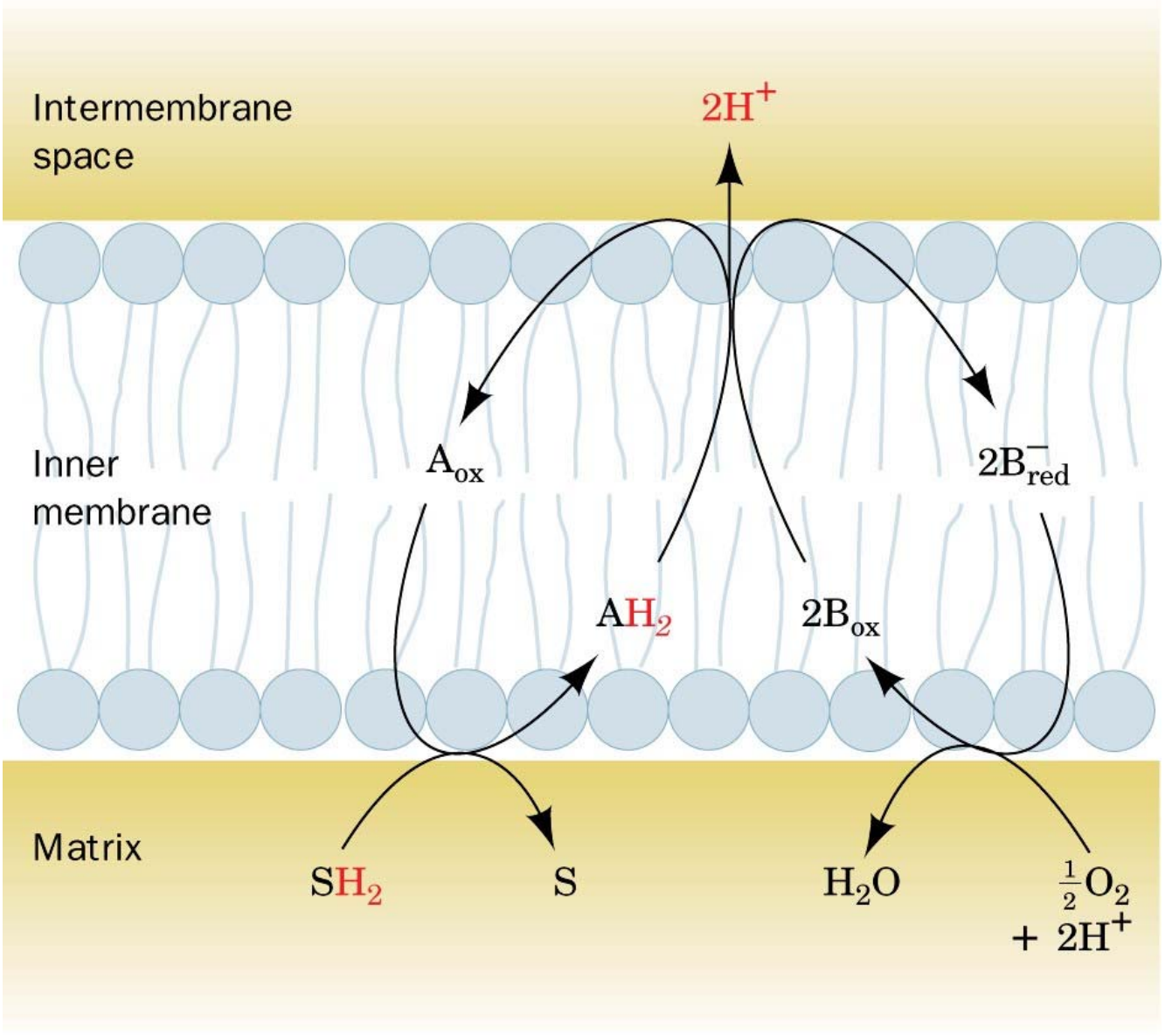
Oxidized form of coenzyme Q (Q, ubiquinone)

Semiquinone intermediate ($Q^{\cdot-}$)

Reduced form of coenzyme Q (QH_2, ubiquinol)


−0.4
−0.2
0
+0.2
+0.4
+0.6
+0.8
$\mathscr{E}^{\circ\prime}$(V)
NADH
$2e^-$
NAD$^+$ (−0.315 V)
Complex I
$\Delta\mathscr{E}^{\circ\prime}$ = 0.360 V
($\Delta G^{\circ\prime}$ = −69.5 kJ · mol^{-1})
ADP + P_i
Rotenone or Amytal
ATP
(+0.031 V)
Succinate
$2e^-$
FADH$_2$
Complex II
CoQ (+0.045 V)
Fumarate
Complex III
$\Delta\mathscr{E}^{\circ\prime}$ = 0.190 V
($\Delta G^{\circ\prime}$ = −36.7 kJ · mol^{-1})
ADP + P_i
Antimycin A
ATP
Cytochrome c (+0.235 V)
Complex IV
$\Delta\mathscr{E}^{\circ\prime}$ = 0.580 V
($\Delta G^{\circ\prime}$ = −112 kJ · mol^{-1})
ADP + P_i
CN$^-$
ATP
$2e^-$
2H$^+$ + $\frac{1}{2}$O$_2$
H$_2$O (+0.815 V)

The redox loop mechanism for electron transport–linked H^+ translocation



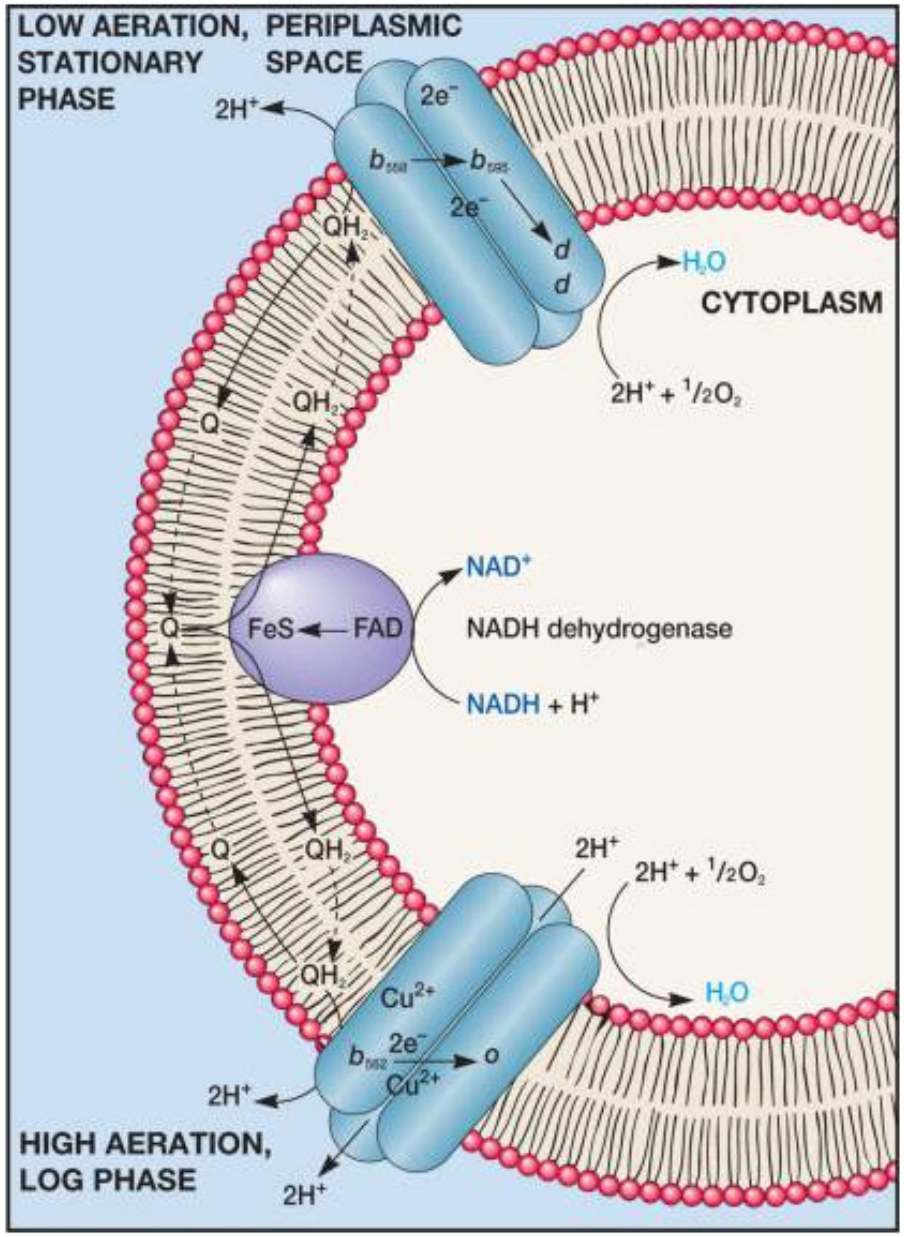
Procaryotic ETCs

Located in plasma membrane

Some resemble mitochondrial ETC, but many are different

- different electron carriers
- may be branched
- may be shorter
- may have lower P/O ratio

Electron Transport Chain of *E. coli*



stationary phase and low aeration

Branched pathway

log phase and high aeration

Oxidative Phosphorylation

Peter Mitchell

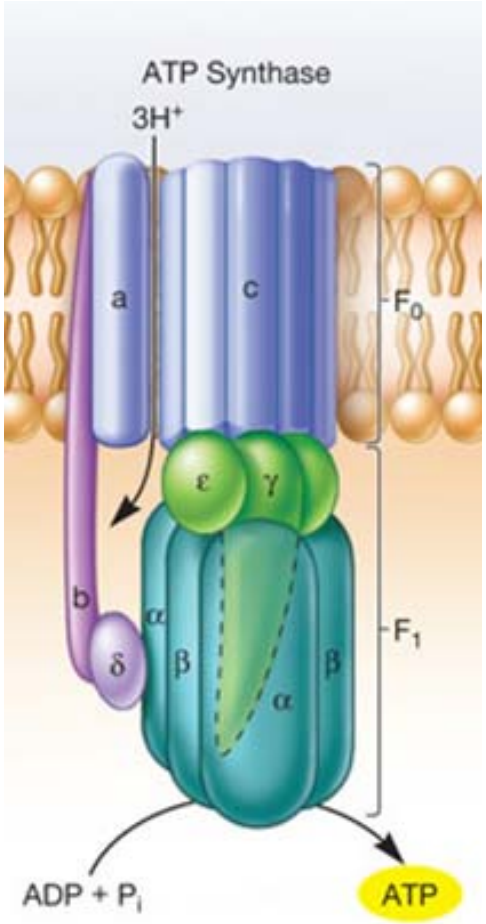
Process by which ATP is synthesized as the result of electron transport

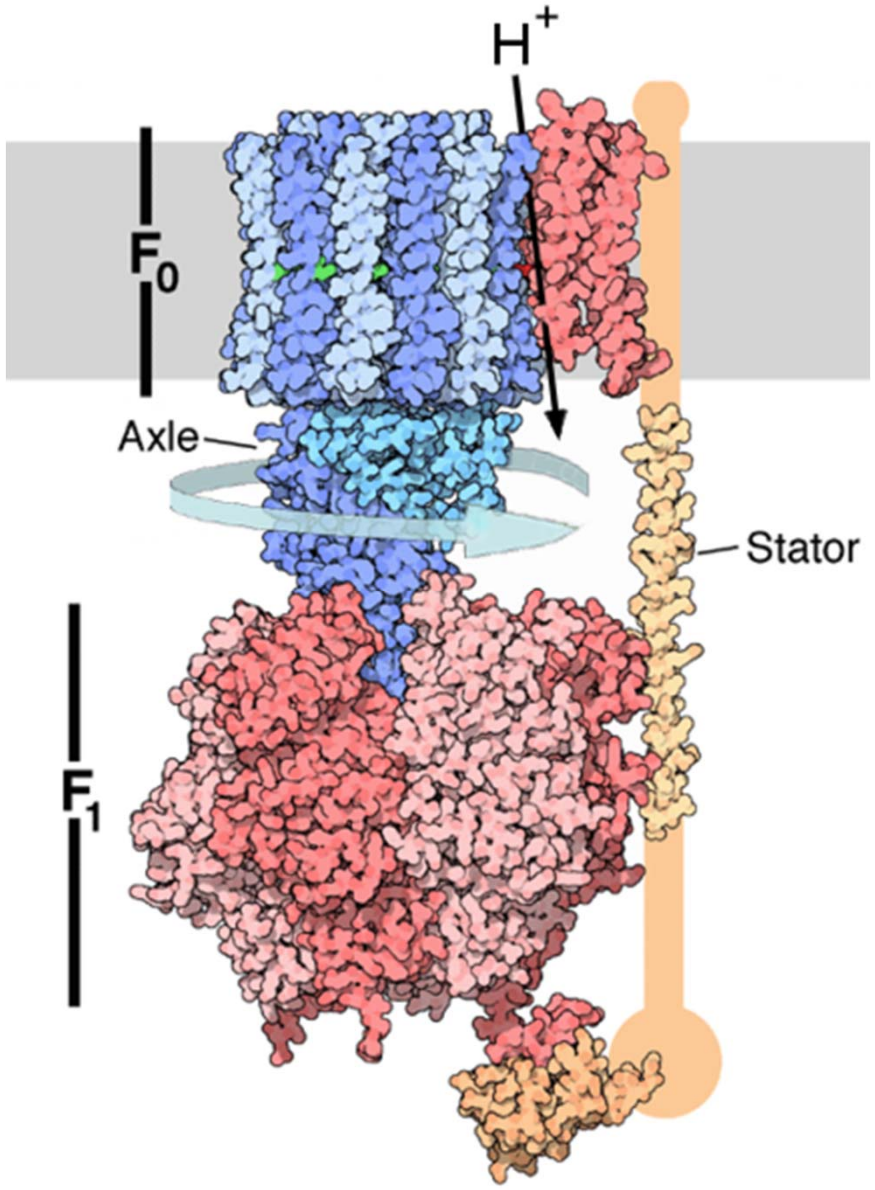
Proton motive force (PMF) drives ATP synthesis

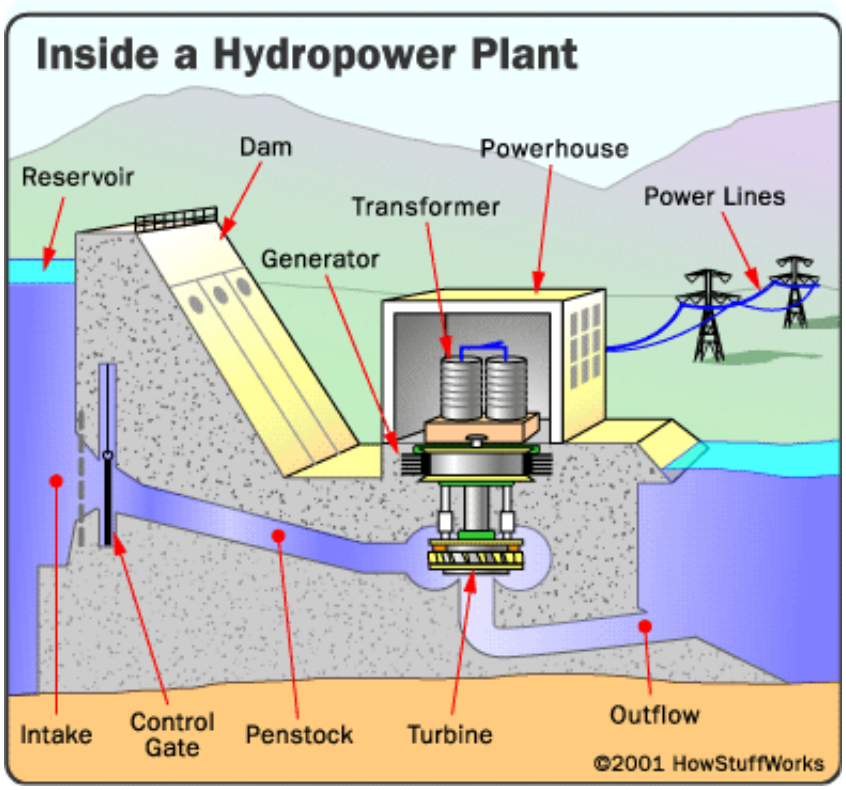
Diffusion of protons back across membrane (down gradient) drives formation of ATP

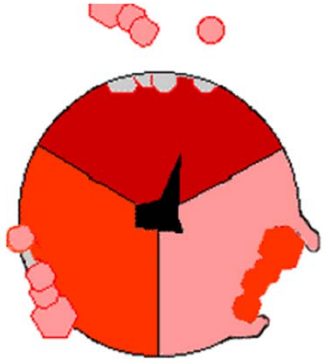
ATP synthase

- enzyme that uses PMF down gradient to catalyze ATP synthesis

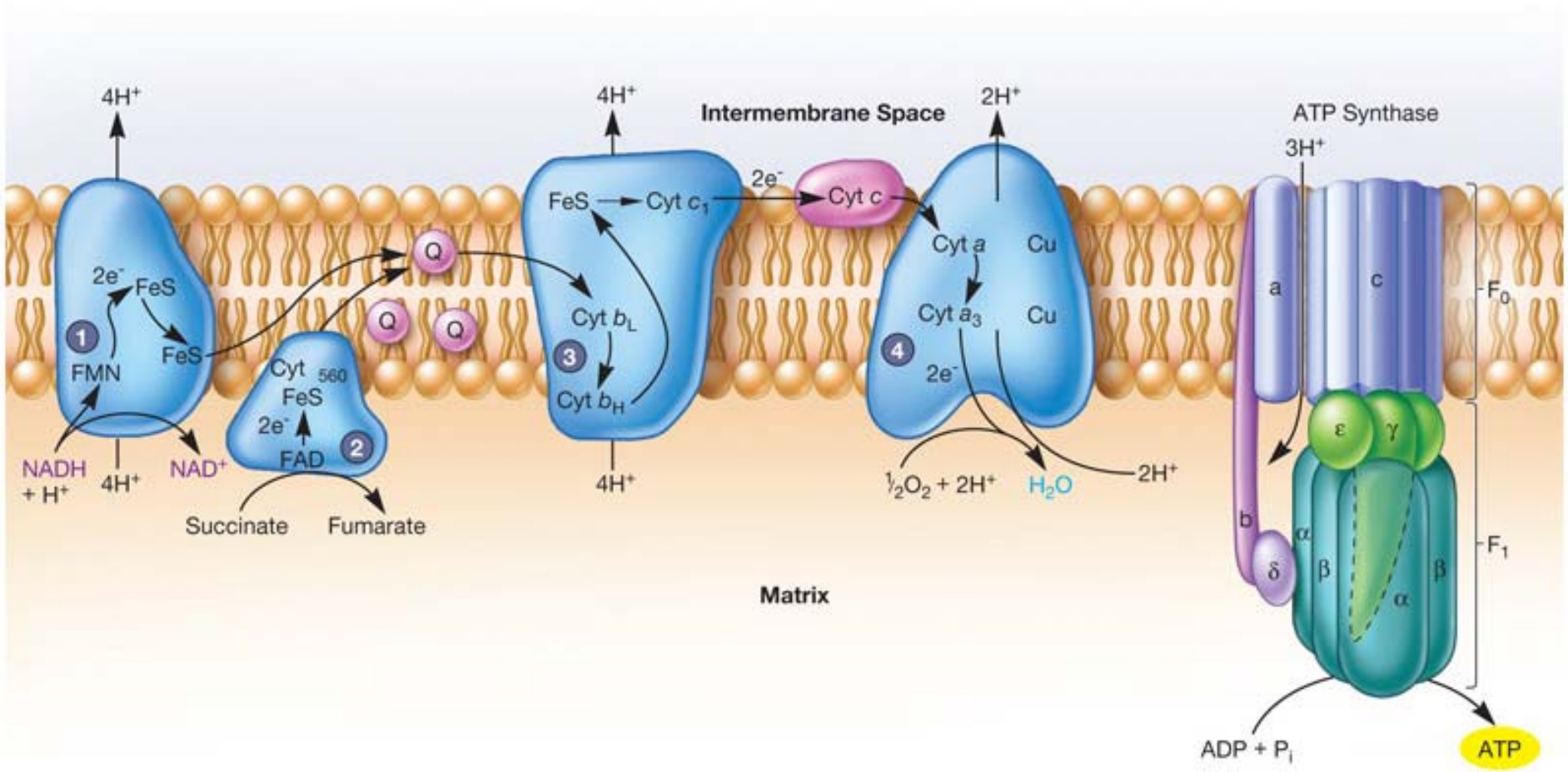



H+
F0
Axle
Stator
F1

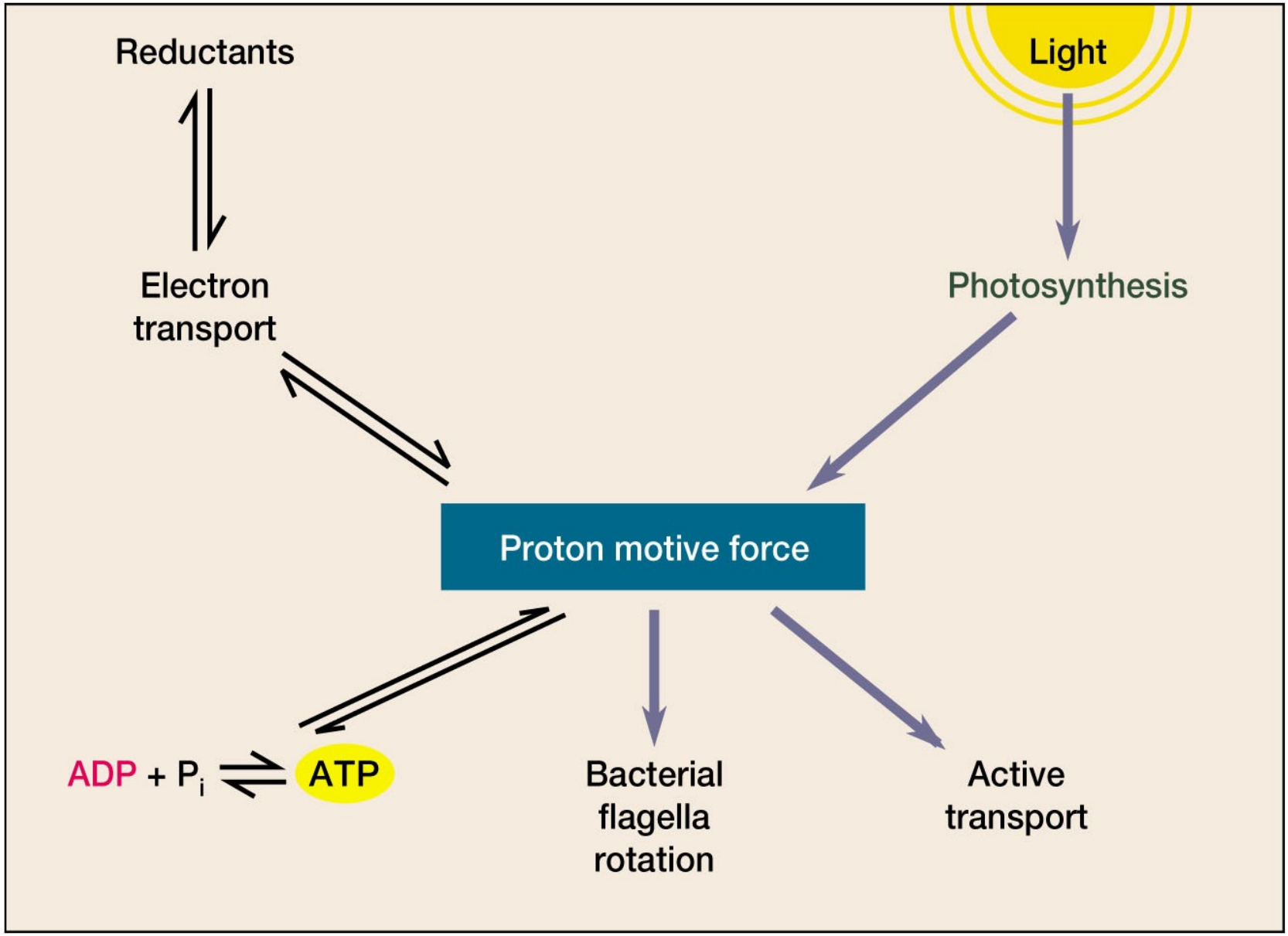

Inside a Hydropower Plant
Reservoir
Dam
Powerhouse
Transformer
Power Lines
Generator
Intake
Control Gate
Penstock
Turbine
Outflow
©2001 HowStuffWorks



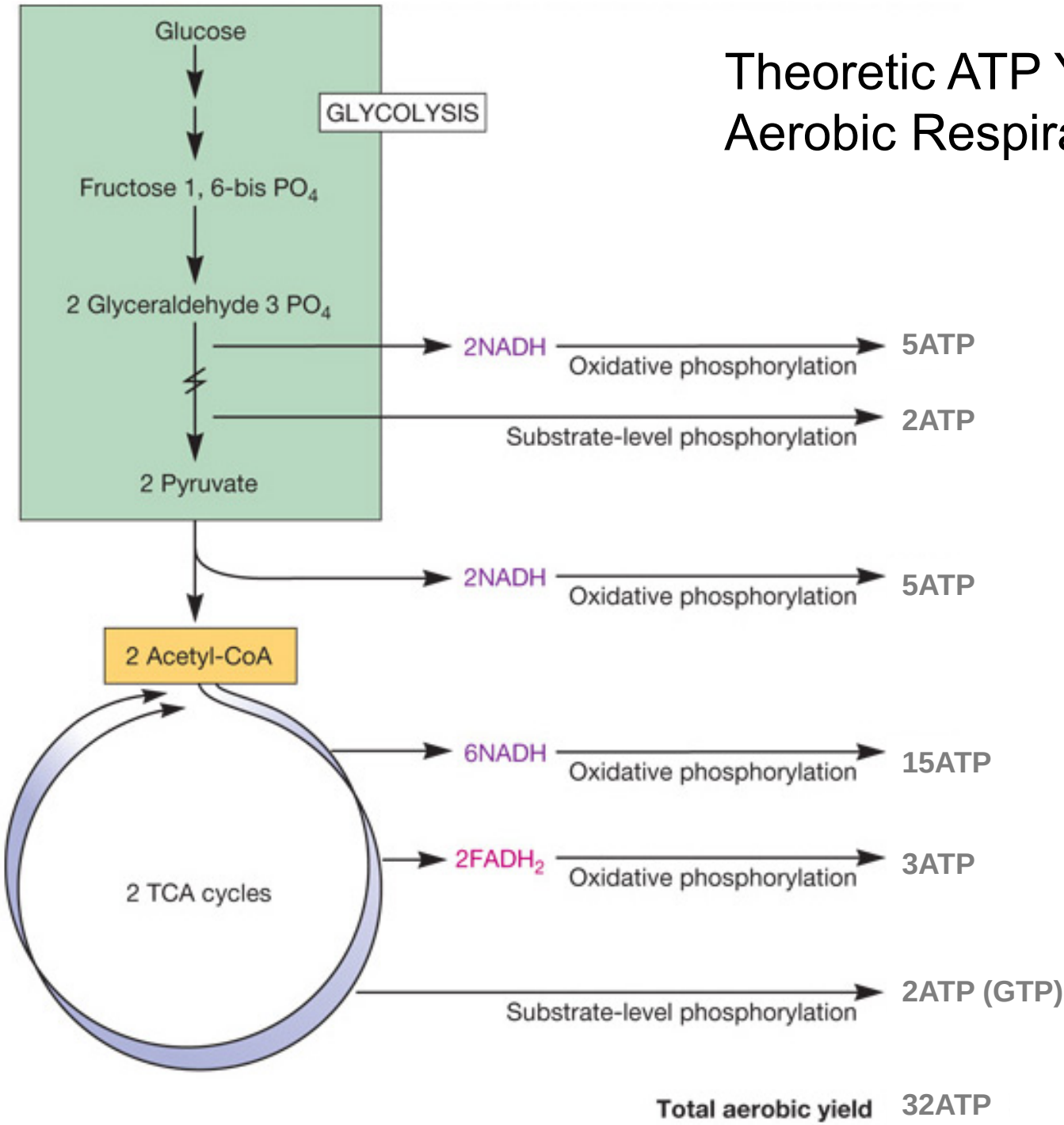
ETC, PMF and Oxidative phosphorylation



Importance of PMF



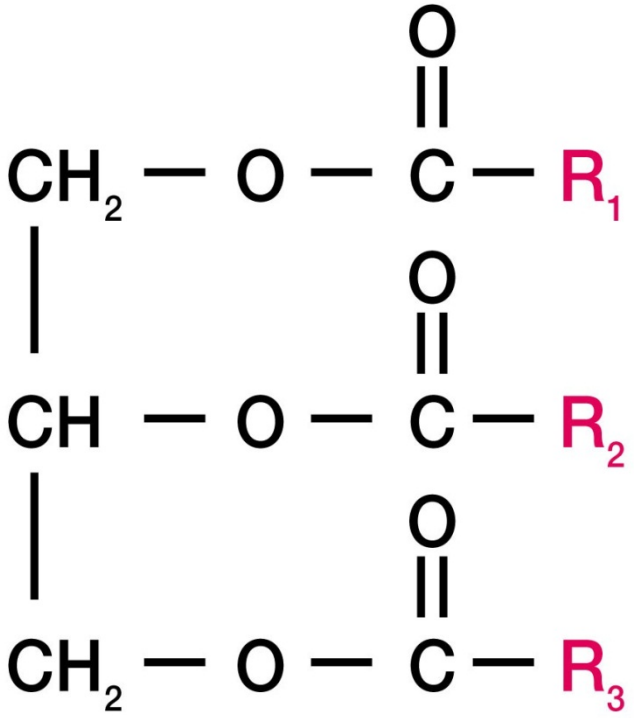
Theoretic ATP Yield from Aerobic Respiration



Lipid catabolism

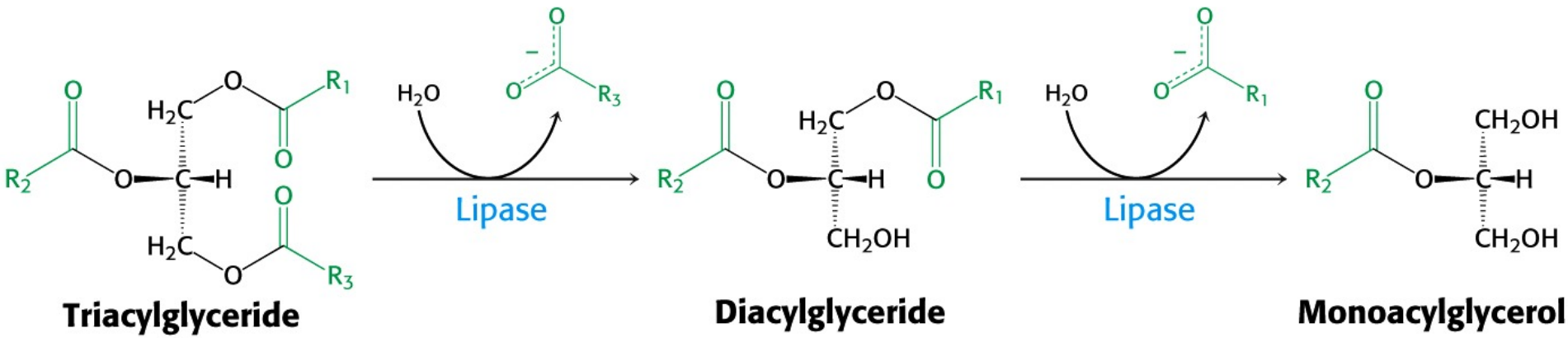
Triglycerides

- common energy sources
- hydrolyzed to glycerol and fatty acids by lipases
  - glycerol degraded via glycolytic pathway
  - fatty acids often oxidized via β-oxidation pathway

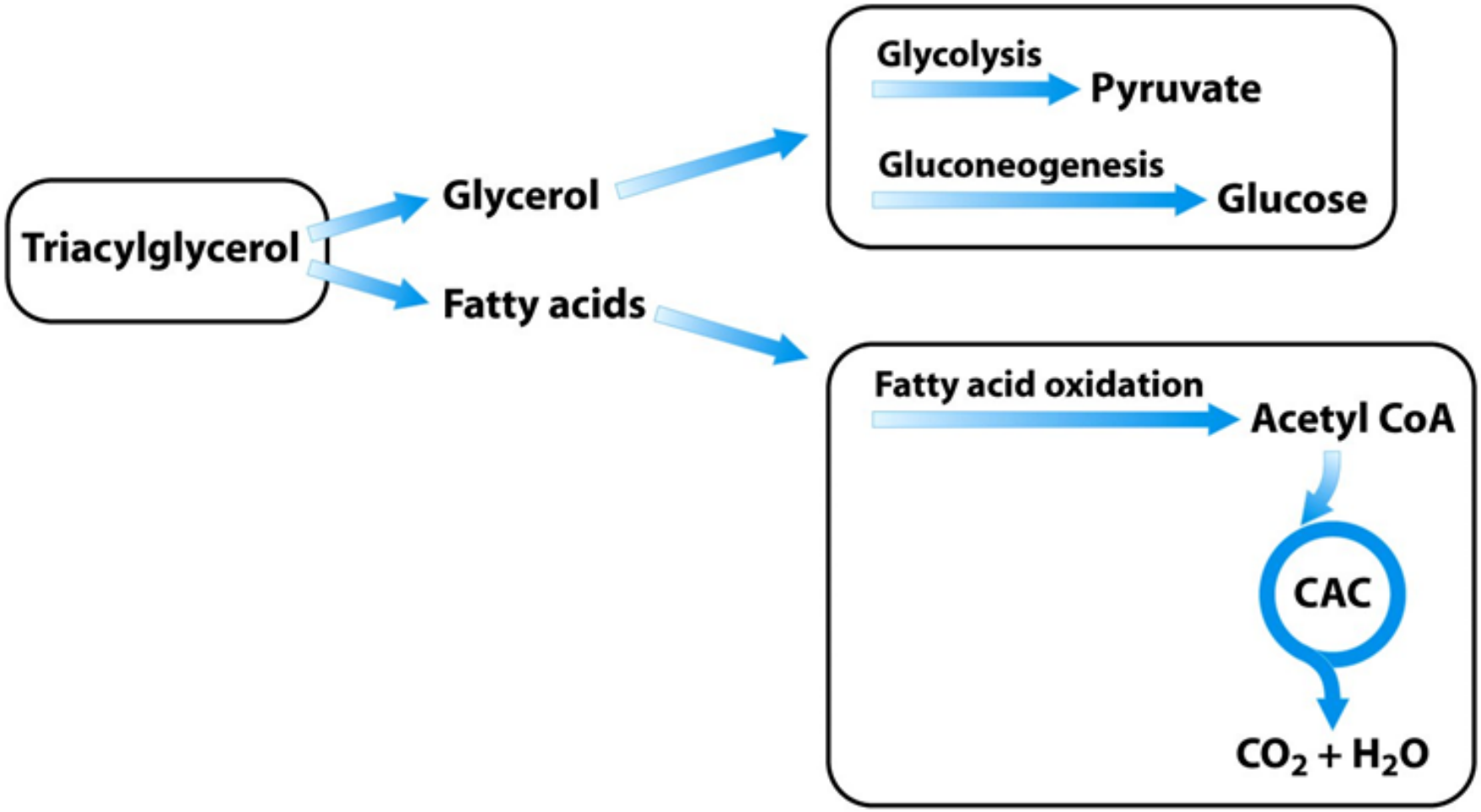


Lipase reactions

The enzyme lipase catalyses the hydrolysis of two of the fatty acids in the intestine.



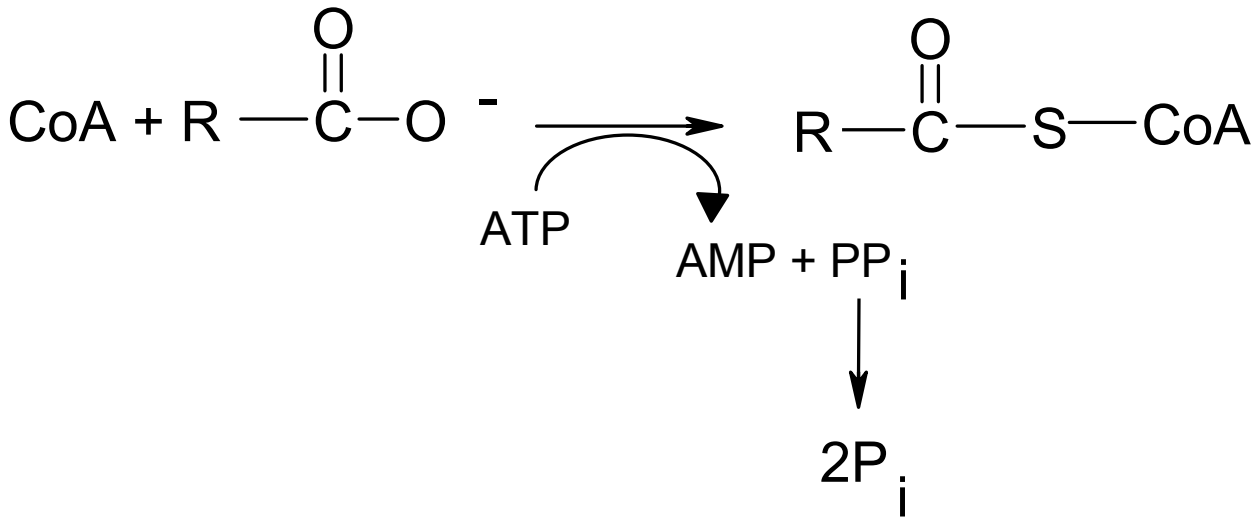
Triacylglycerol is hydrolyzed to fatty acids and glycerol, which can be used in other cells



Degradation of fatty acid has three steps:

1. Activation
2. Transport into mitochondrion
3. β-oxidation

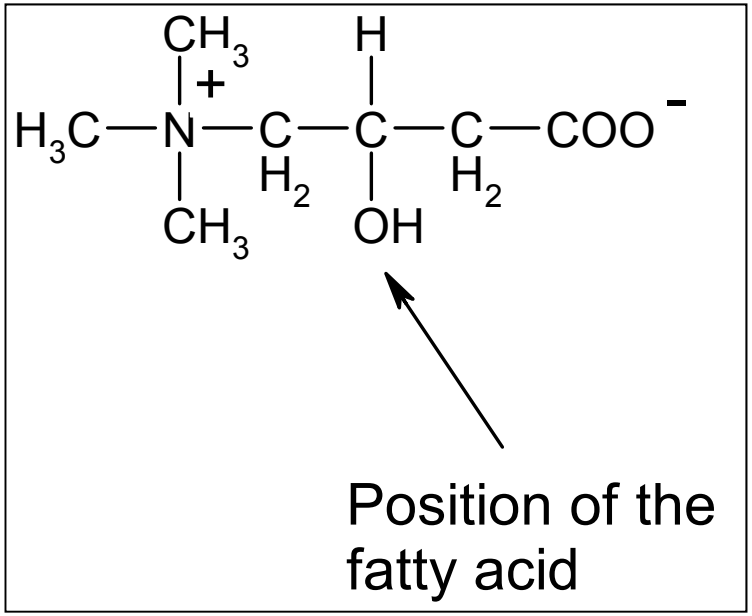
Activation of the fatty acid consists of esterification to CoA at the expense of ATP

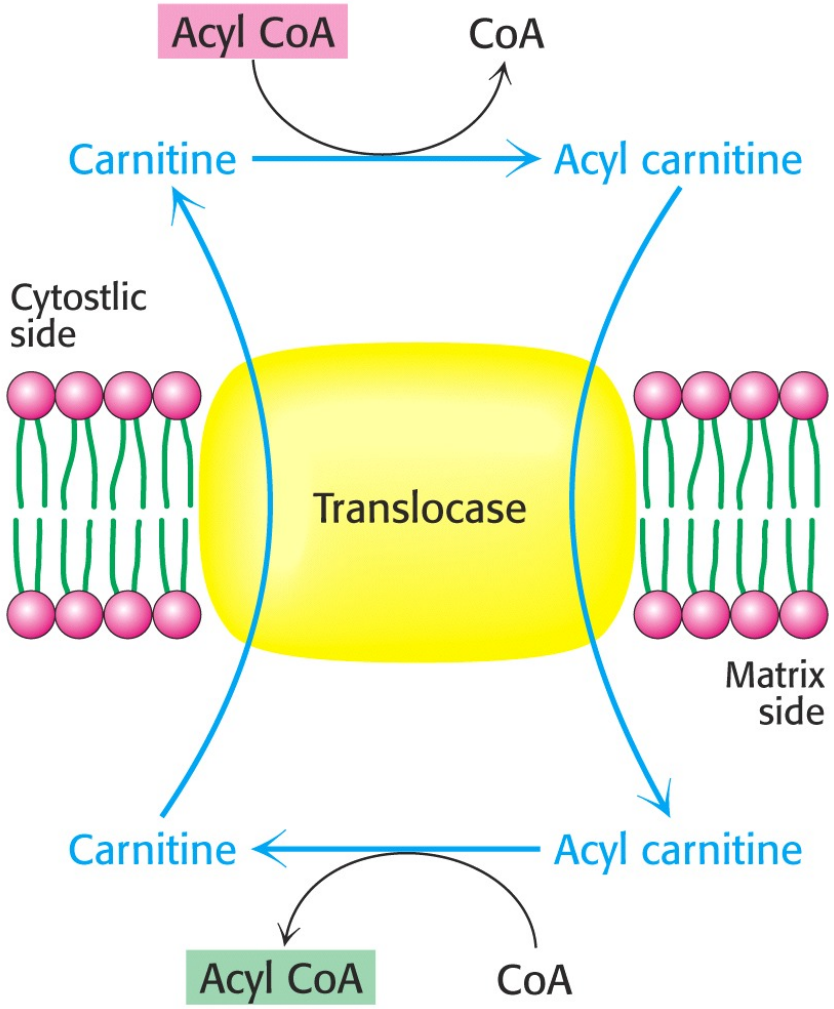


Transport of fatty acid into mitochondrion

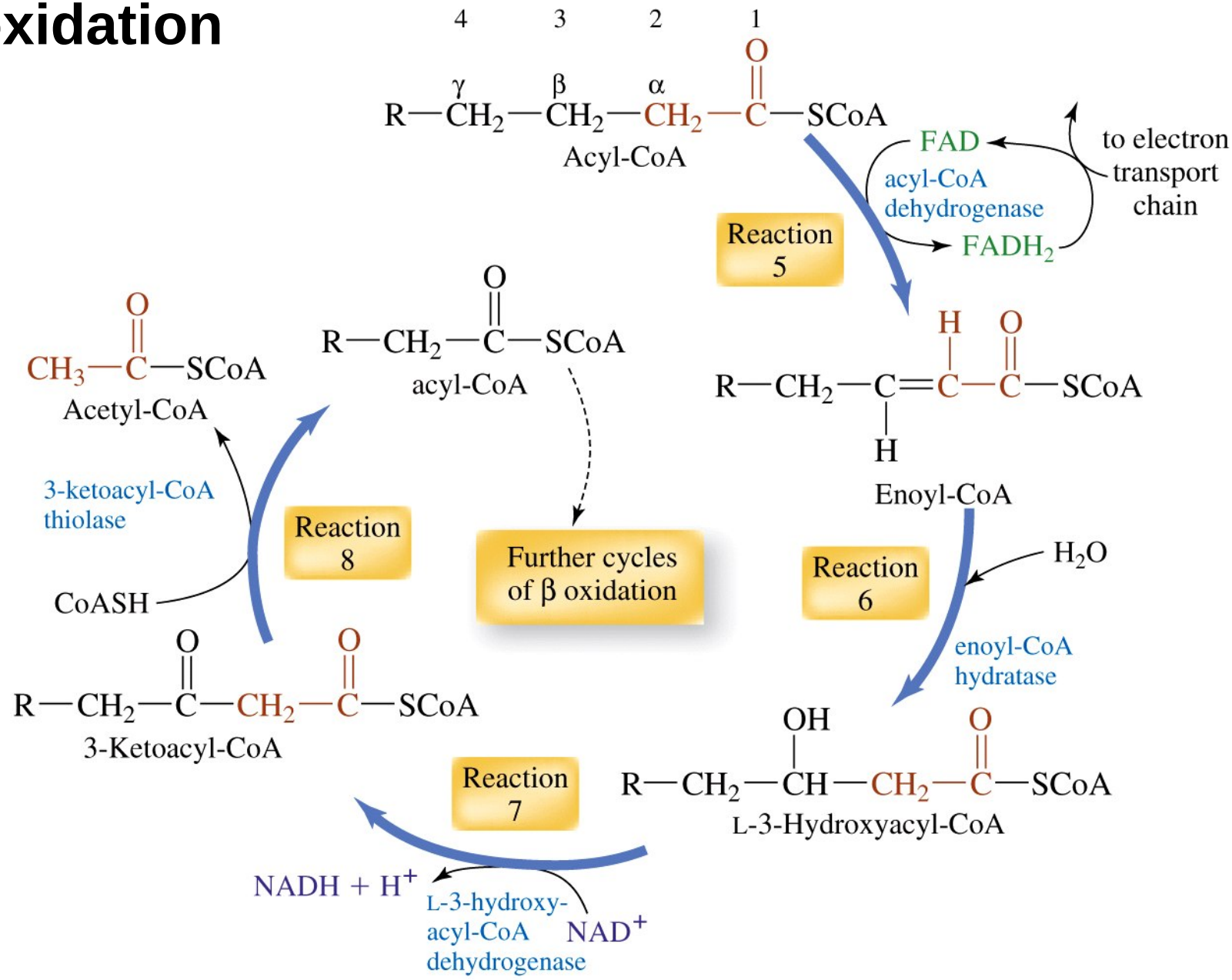
The acyl group is transported into mitochondrion using carnitine. A translocase brings in acyl-carnitine and expels carnitine. Acyl-CoA is regenerated inside mitochondrion.

Carnitine





β-oxidation


4 3 2 1
γ β α
R—CH2—CH2—CH2—C—SCoA
Acyl-CoA
FAD
acyl-CoA dehydrogenase
FADH2
to electron transport chain
Reaction 5
R—CH2—C—SCoA
acyl-CoA
H O
R—CH2—C=C—C—SCoA
H
Enoyl-CoA
H2O
Reaction 6
enoyl-CoA hydratase
OH O
R—CH2—CH—CH2—C—SCoA
L-3-Hydroxyacyl-CoA
Reaction 7
NADH + H+
L-3-hydroxy-acyl-CoA dehydrogenase
NAD+
O O
R—CH2—C—CH2—C—SCoA
3-Ketoacyl-CoA
CoASH
3-ketoacyl-CoA thiolase
Reaction 8
CH3—C—SCoA
Acetyl-CoA
Further cycles of β oxidation

How much ATP can be formed by oxidation of fatty acid?

It is now possible to calculate the amount of ATP formed during complete oxidation of a fatty acid, e.g. palmitic acid

8 Acetyl-CoA in the tricarboxylic acid cycle: 8 x 3 = 24 NADH
8 x 1 = 8 $FADH_2$
8 x 1 = 8 GTP

β-Oxidation yields: 7 NADH
7 $FADH_2$

In total: 7 + 24 = 31 NADH corresponds to 31 x 2,5 = 77,5 ATP
7 + 8 = 15 $FADH_2$ corresponds to 15 x 1,5 = 22,5 ATP
8 GTP corresponds to 8 ATP

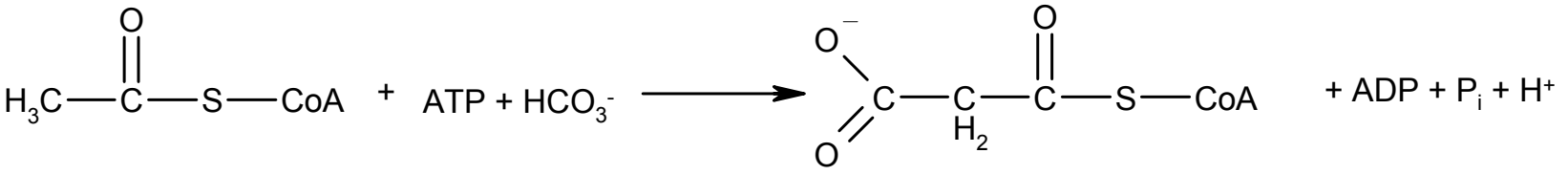
Summarised: 8 + 77,5 + 22,5 = 108 ATP

Two ATP are used during the initial activation of the fatty acid
The total ATP-yield will be **106**

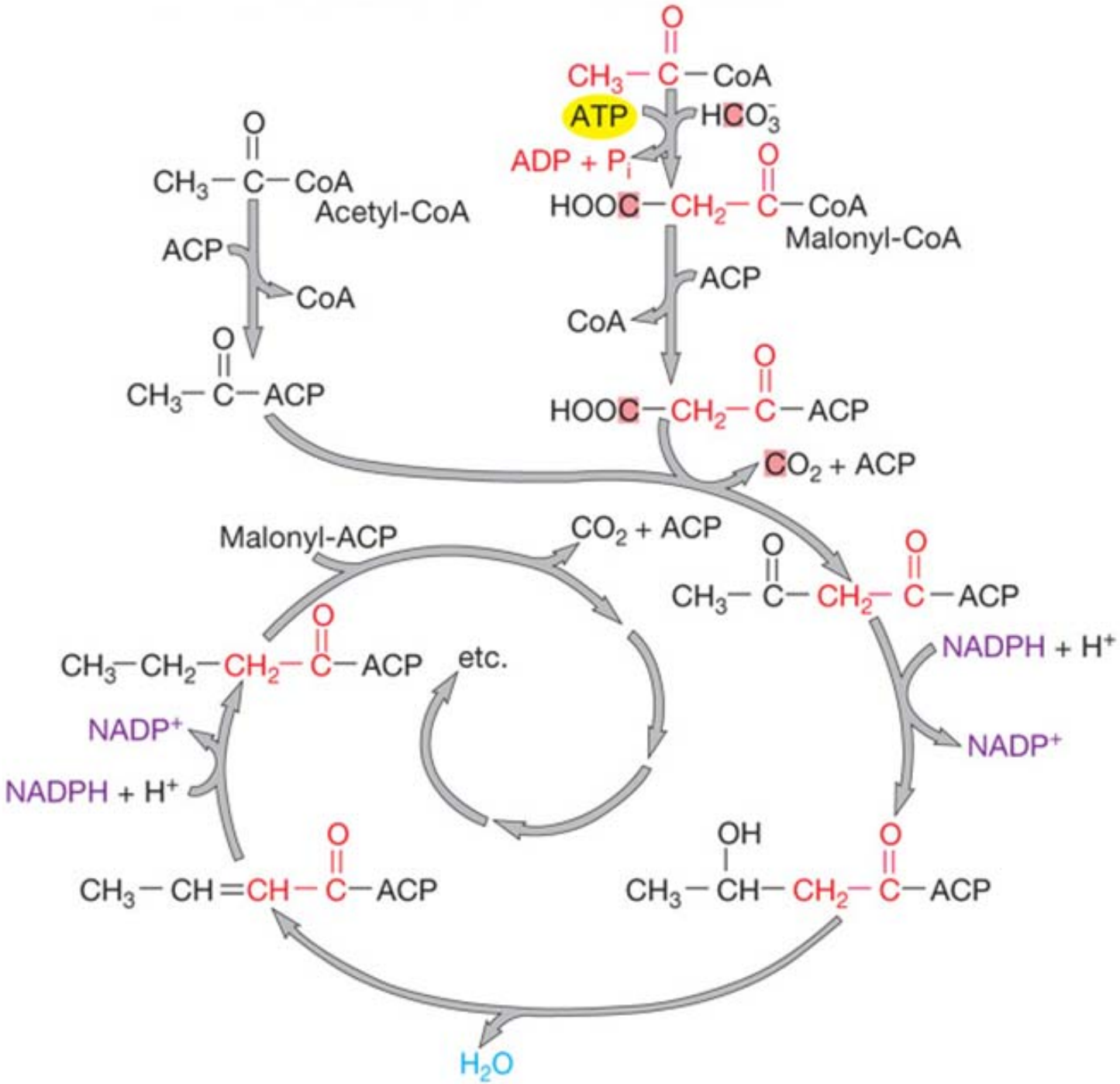
Fatty acid synthesis

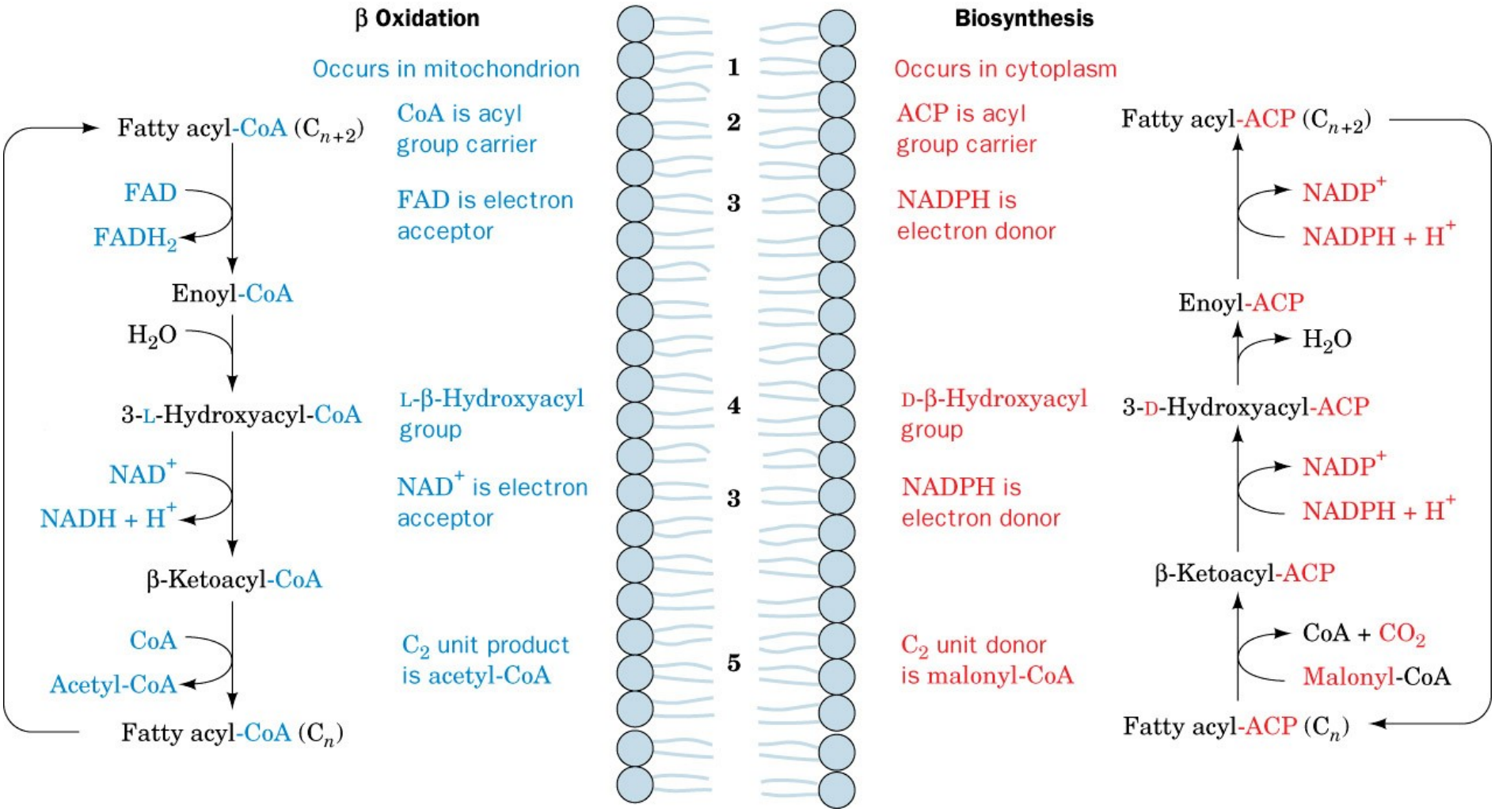
- Fat is an efficient way to store energy
- It is usually surplus of carbohydrates that are converted to fat
- The reactions are in principle a reversed β-oxidation

Synthesis of fatty acid starts with the carboxylation of acetyl-CoA to malonyl-CoA. This is the committed step, malonyl CoA can only be used for synthesis of fatty acid.



Acetyl-CoA **Malonyl-CoA**


CH3—C—CoA
ATP
HCO3−
ADP + Pi
HOOC—CH2—C—CoA
Malonyl-CoA
Acetyl-CoA
ACP
CoA
CH3—C—ACP
HOOC—CH2—C—ACP
CO2 + ACP
Malonyl-ACP
CO2 + ACP
CH3—C—CH2—C—ACP
NADPH + H+
NADP+
CH3—CH2—CH2—C—ACP
etc.
NADP+
NADPH + H+
CH3—CH=CH—C—ACP
CH3—CH—CH2—C—ACP
OH
H2O


β Oxidation
Occurs in mitochondrion
Fatty acyl-CoA (C_{n+2})
CoA is acyl group carrier
FAD
$FADH_2$
FAD is electron acceptor
Enoyl-CoA
H_2O
3-L-Hydroxyacyl-CoA
L-β-Hydroxyacyl group
NAD^+
$NADH + H^+$
NAD^+ is electron acceptor
β-Ketoacyl-CoA
CoA
Acetyl-CoA
C_2 unit product is acetyl-CoA
Fatty acyl-CoA (C_n)
1
2
3
4
3
5
Biosynthesis
Occurs in cytoplasm
ACP is acyl group carrier
NADPH is electron donor
D-β-Hydroxyacyl group
NADPH is electron donor
C_2 unit donor is malonyl-CoA
Fatty acyl-ACP (C_{n+2})
$NADP^+$
$NADPH + H^+$
Enoyl-ACP
H_2O
3-D-Hydroxyacyl-ACP
$NADP^+$
$NADPH + H^+$
β-Ketoacyl-ACP
CoA + CO_2
Malonyl-CoA
Fatty acyl-ACP (C_n)

Summary

- Pyruvate dehydrogenase complex bridges glycolysis and TCA cycle
- TCA cycle is the central hub of metabolism, 8 steps, produces 3 NADH + 1 $FADH_2$ + 1 GTP
- Electron transport chain - proton motive force - oxidative phosphorylation
- Lipid metabolism: β-oxidation of fatty acid
- Lipid metabolism: fatty acid synthesis, NADPH as cofactor