PYRUVATE DEHYDROGENASE COMPLEX (PDC)

- **Multienzyme complex:** have multiple enzyme unit to carry out different reaction in sequence

Metabolic fate of pyruvate

- Under aerobic conditions, pyruvate is converted to acetyl CoA which enters the TCA cycle to be oxidized to CO_2.
- ATP is generated.
- Glycolysis is taking place in cytoplasm.
- So pyruvate is generated in cytoplasm.
- This is transported into mitochondria by a pyruvate transporter.

Pyruvate Dehydrogenase Complex

- Inside the mitochondria, pyruvate is oxidatively decarboxylated to acetyl CoA by pyruvate dehydrogenase (PDH).
- It is a multi-enzyme complex with 5 co-enzymes & 3 apo-enzymes.

Coenzymes in PDH Complex

- Thiamine pyrophosphate (TPP)
- Co-enzyme A (CoA)
- FAD
- NAD^+
- Lipoamide
- The lipoic acid, also called thioctic acid has two sulphur atoms & 8 carbon atoms.
- It can accept or donate hydrogen atoms

Enzymes in PDH Complex

- Pyruvate Dehydrogenase (Enzyme 1)
- Dihydro Lipoyl Trans Acetylase (Enzyme 2)
- Dihydro Lipoyl Dehydrogenase (Enzyme 3)

Pyruvate Dehydrogenase (Enzyme 1)

- It catalyses oxidative decarboxylation.
- It requires TPP.
- Thiamine (B_1), a B-complex group vitamin is essential for utilization of pyruvate.
- The two carbon unit remains attached to the enzyme, as hydroxyethyl-TPP.

Dihydro Lipoyl Trans Acetylase (Enzyme 2)

- Hydroxyethyl group is oxidized to form an acetyl group and then transferred from TPP to lipoamide to form acetyl lipoamide.

Dihydro Lipoyl Dehydrogenase (Enzyme 3)

- The last step is the oxidation of lipoamide.
- At the end of the reaction the cofactors, namely TPP, Lipoamide & FAD are regenerated.
- $FADH_2$ transfers the reducing equivalents to NAD^+ to give NADH + H^+, which can pass through the ETC to give 3 ATP (6 ATP from 2 moles of pyruvate formed from glucose) by oxidative phosphorylation.

Pyruvate Is Oxidized to Acetyl-CoA and CO_2

The overall reaction catalyzed by the pyruvate dehydrogenase complex is an **oxidative decarboxylation,** an irreversible oxidation process in which the carboxyl group is removed from pyruvate as a molecule of CO_2

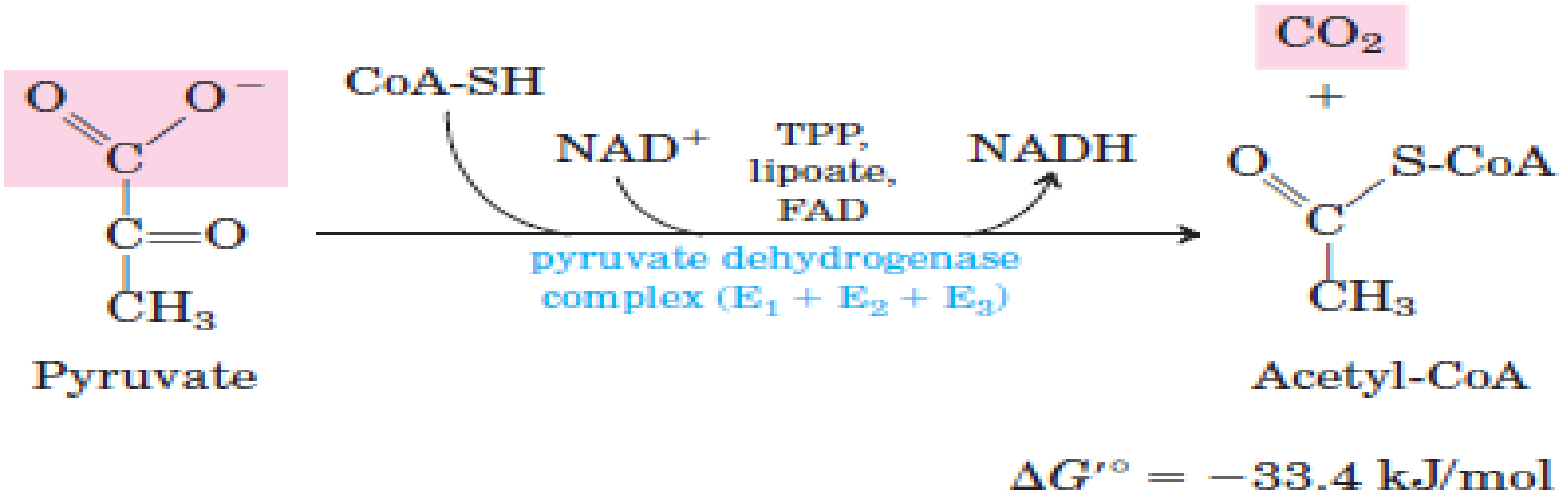
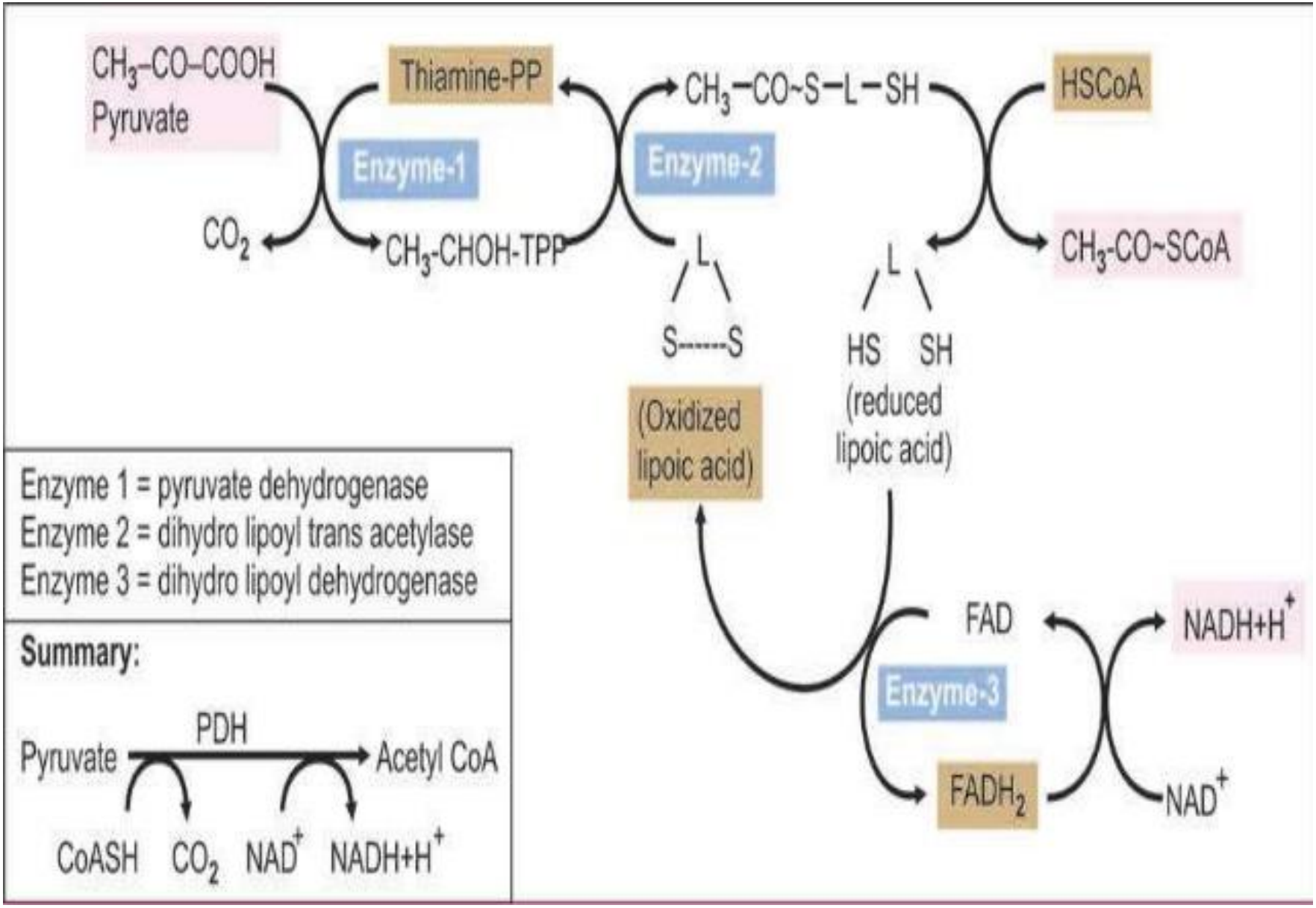


FIGURE 16–2 Overall reaction catalyzed by the pyruvate dehydrogenase complex. The five coenzymes participating in this reaction, and the three enzymes that make up the enzyme complex, are discussed in the text.

The Pyruvate Dehydrogenase Complex Consists of Three Distinct Enzymes

The PDH complex contains three enzymes—**pyruvate dehydrogenase** (E_1), **dihydrolipoyl transacetylase** (E_2), and **dihydrolipoyl dehydrogenase** (E_3)—each present in multiple copies. The number of copies of each enzyme and therefore the size of the complex varies among species. The PDH complex isolated from mammals is about 50 nm in diameter—more than five times the size of an entire ribosome and big enough to be visualized with the electron microscope (Fig. 16–5a). In the bovine enzyme, 60 identical copies of E_2 form a pentagonal dodecahedron (the core) with a diameter of about 25 nm (Fig. 16–5b). (The core of the *Escherichia coli* enzyme contains 24 copies of E_2.) E_2 is the point of connection for the prosthetic group lipoate, attached through an amide bond to the ε-amino group of a Lys residue (Fig. 16–4). E_2 has three functionally distinct domains (Fig. 16–5c): the amino-terminal *lipoyl domain,* containing the lipoyl-Lys residue(s); the central E_1- and E_3-*binding domain;* and the inner-core *acyltransferase domain,* which contains the acyltransferase active site. The yeast PDH complex has a single lipoyl domain with a lipoate attached, but the mammalian complex has two, and *E. coli* has three (Fig. 16–5c). The domains of E_2 are separated by linkers, sequences of 20 to 30 amino acid residues, rich in Ala and Pro and interspersed with charged residues; these linkers tend to assume their extended forms, holding the three domains apart.

The active site of E_1 has bound TPP, and that of E_3 has bound FAD. Also part of the complex are two reg-


CH3–CO–COOH
Pyruvate
Thiamine-PP
Enzyme-1
CO2
CH3-CHOH-TPP
CH3–CO~S–L–SH
Enzyme-2
L
S------S
(Oxidized lipoic acid)
HSCoA
CH3-CO~SCoA
L
HS SH
(reduced lipoic acid)
FAD
Enzyme-3
FADH2
NADH+H+
NAD+
Enzyme 1 = pyruvate dehydrogenase
Enzyme 2 = dihydro lipoyl trans acetylase
Enzyme 3 = dihydro lipoyl dehydrogenase
Summary:
Pyruvate
PDH
Acetyl CoA
CoASH
CO2
NAD+
NADH+H+

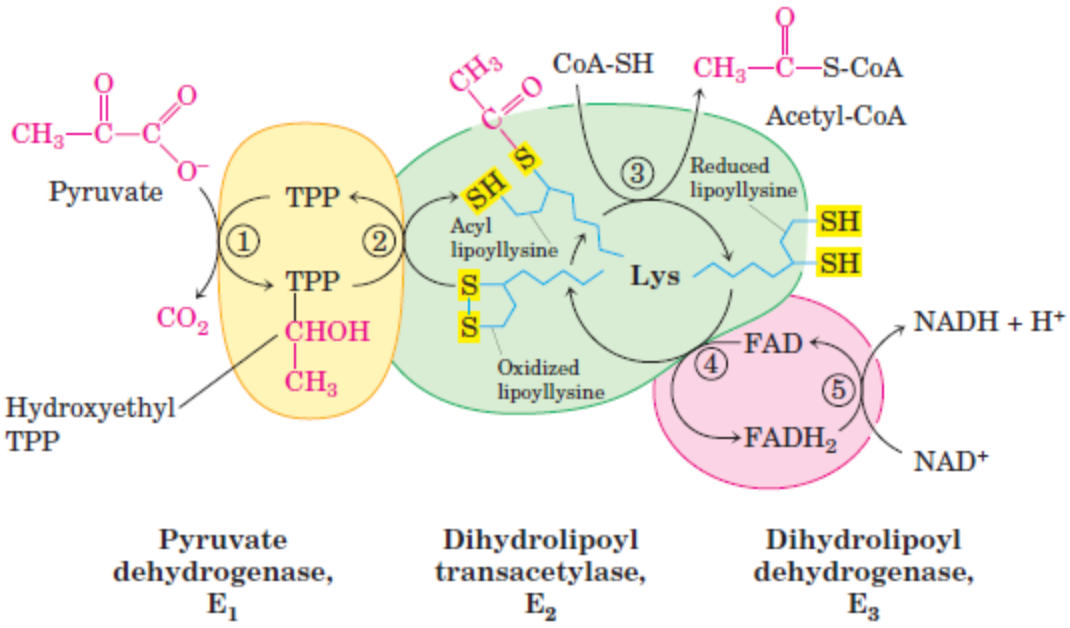


FIGURE 16–6 Oxidative decarboxylation of pyruvate to acetyl-CoA by the PDH complex. The fate of pyruvate is traced in red. In step ① pyruvate reacts with the bound thiamine pyrophosphate (TPP) of pyruvate dehydrogenase (E_1), undergoing decarboxylation to the hydroxyethyl derivative (see Fig. 14–13). Pyruvate dehydrogenase also carries out step ②, the transfer of two electrons and the acetyl group from TPP to the oxidized form of the lipoyllysyl group of the core enzyme, dihydrolipoyl transacetylase (E_2), to form the acetyl thioester of the reduced lipoyl group. Step ③ is a transesterification in which the —SH group of CoA replaces the —SH group of E_2 to yield acetyl-CoA and the fully reduced (dithiol) form of the lipoyl group. In step ④ dihydrolipoyl dehydrogenase (E_3) promotes transfer of two hydrogen atoms from the reduced lipoyl groups of E_2 to the FAD prosthetic group of E_3, restoring the oxidized form of the lipoyllysyl group of E_2. In step ⑤ the reduced $FADH_2$ of E_3 transfers a hydride ion to NAD^+, forming NADH. The enzyme complex is now ready for another catalytic cycle. (Subunit colors correspond to those in Fig. 16–5b.)

Regulation of PDH

- PDH is a good example for end product (acetyl CoA, NADH) inhibition.
- PDH is also regulated by phosphorylation & dephosphoryaltion.
- PDH is active as a dephosphoenzyme.
- PDH is inactive as a phosphoenzyme.
- PDH phosphatase activity is promoted by Ca^{2+}, Mg^{2+} & insulin (in adipose tissue).

Pyruvate Dehydrogenase Complex

Complex enzyme

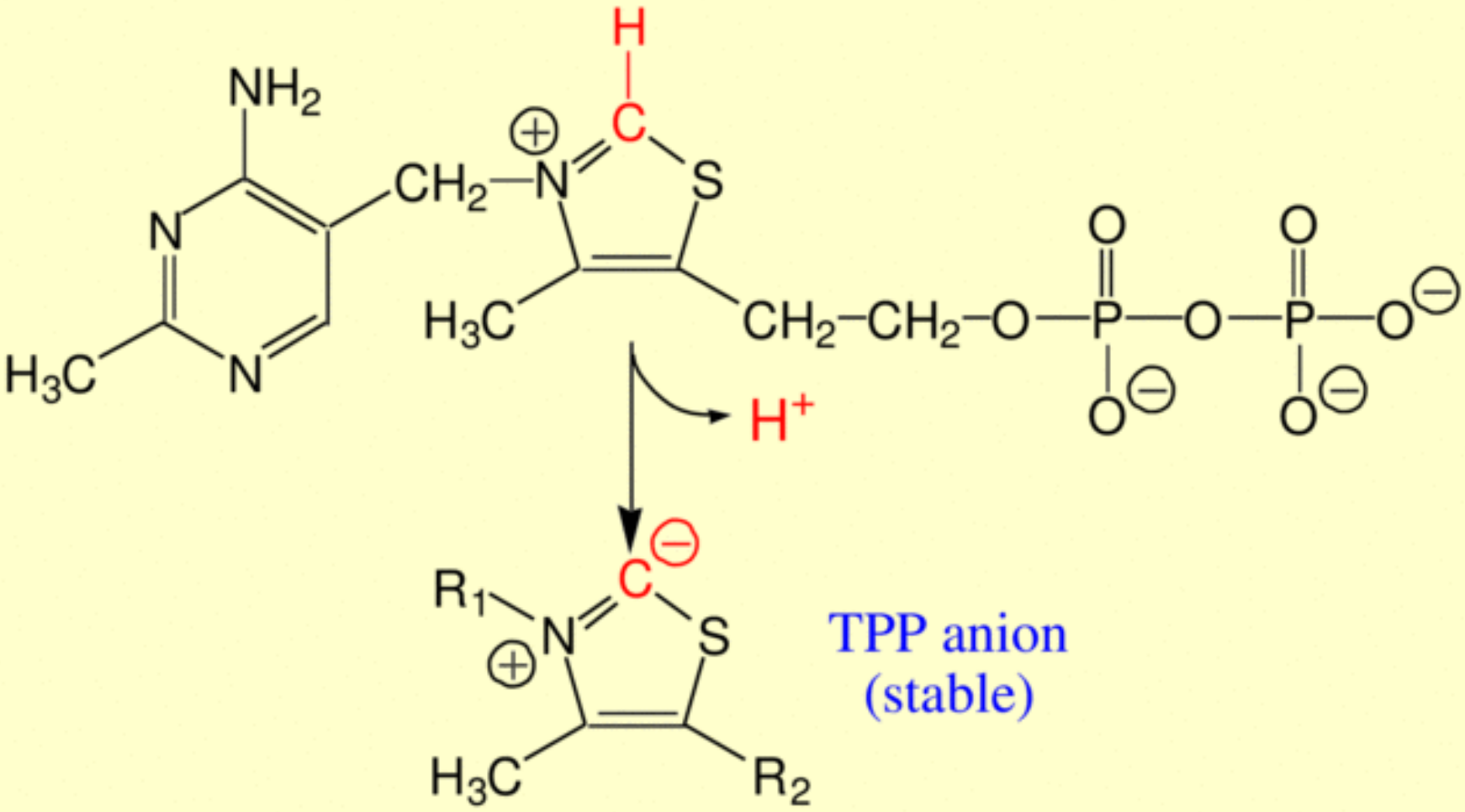
- **60 polypeptides of 3 kinds**

In mitocondrion matrix

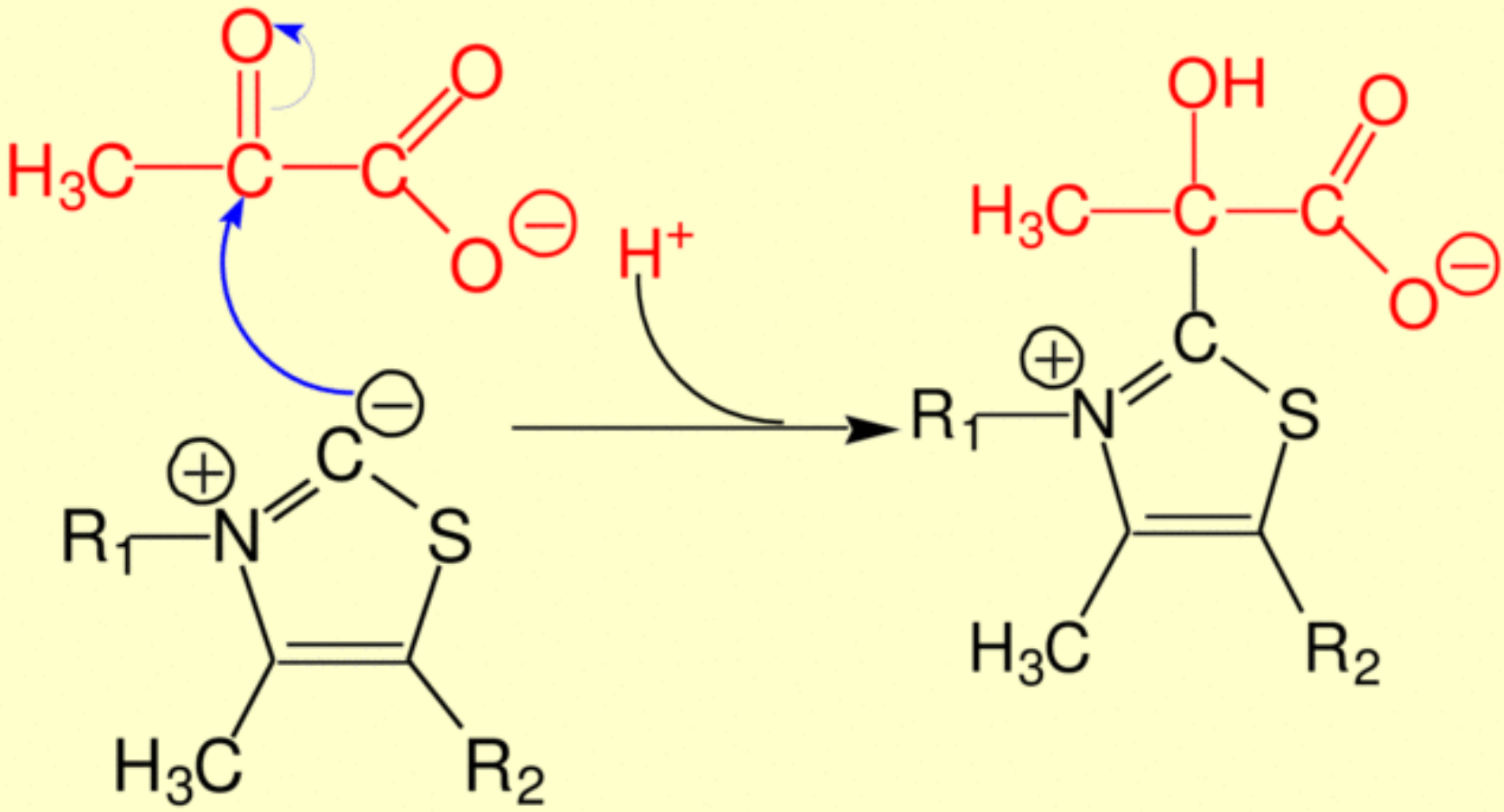
Regulated

- **Inhibited by NADH and GTP**
- **Stimulated by insulin**

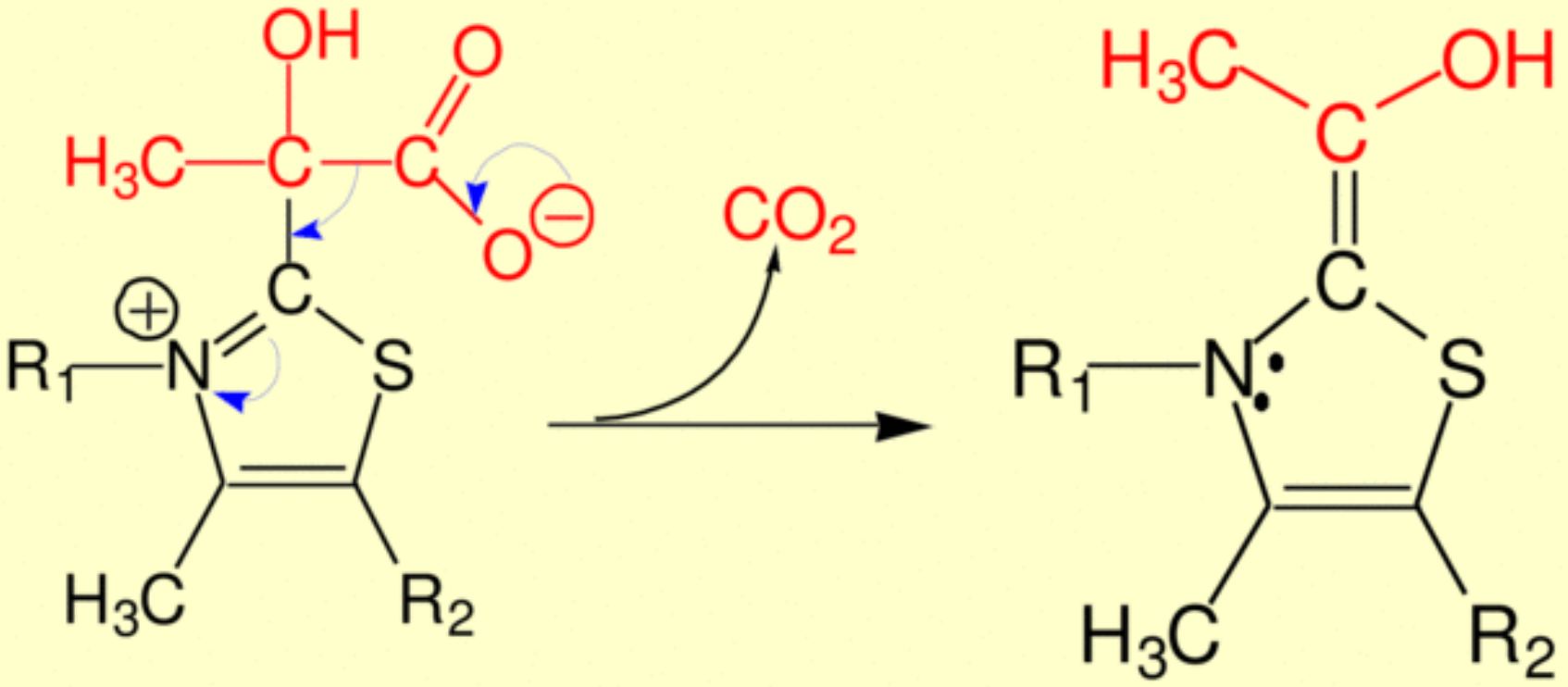
Thiamine Pyrophosphate (TPP)



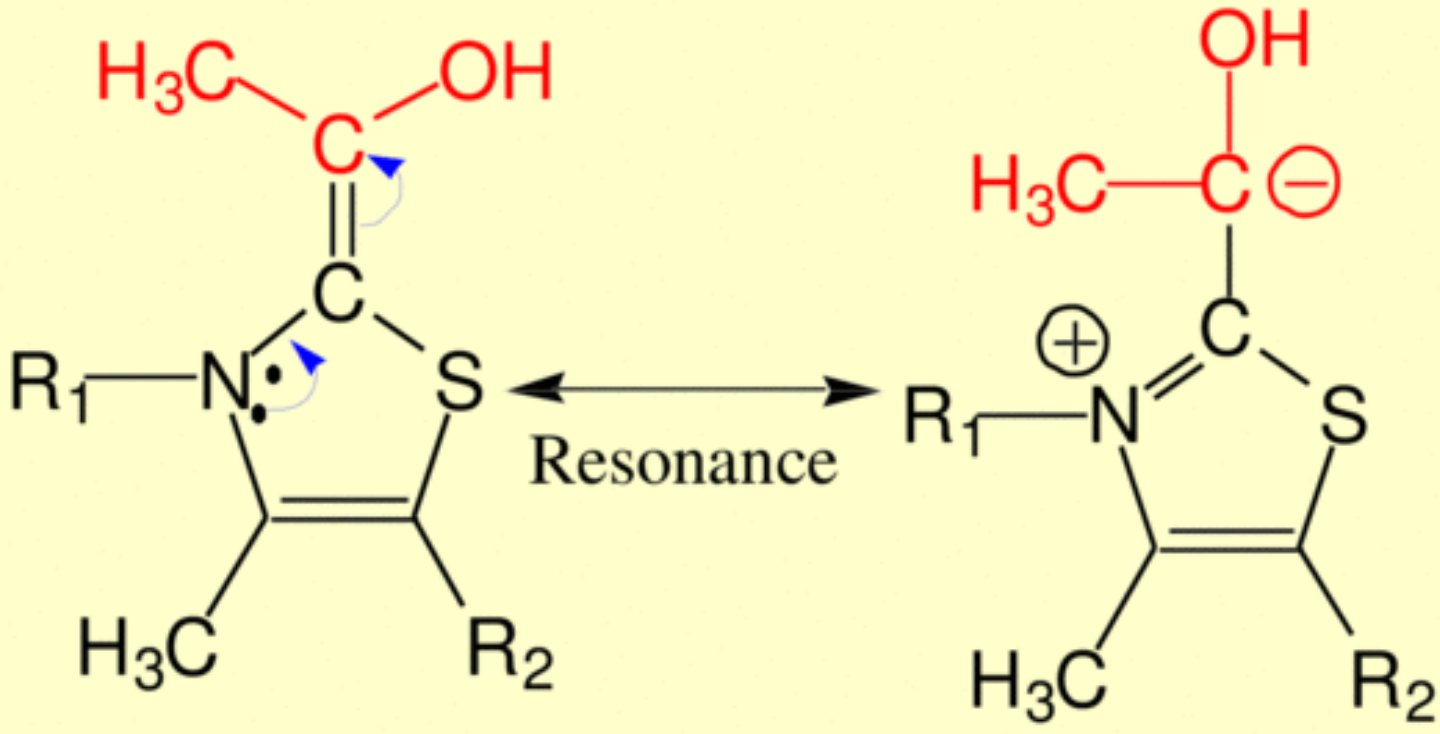
Pyruvate Dehydrogenase Mechanism



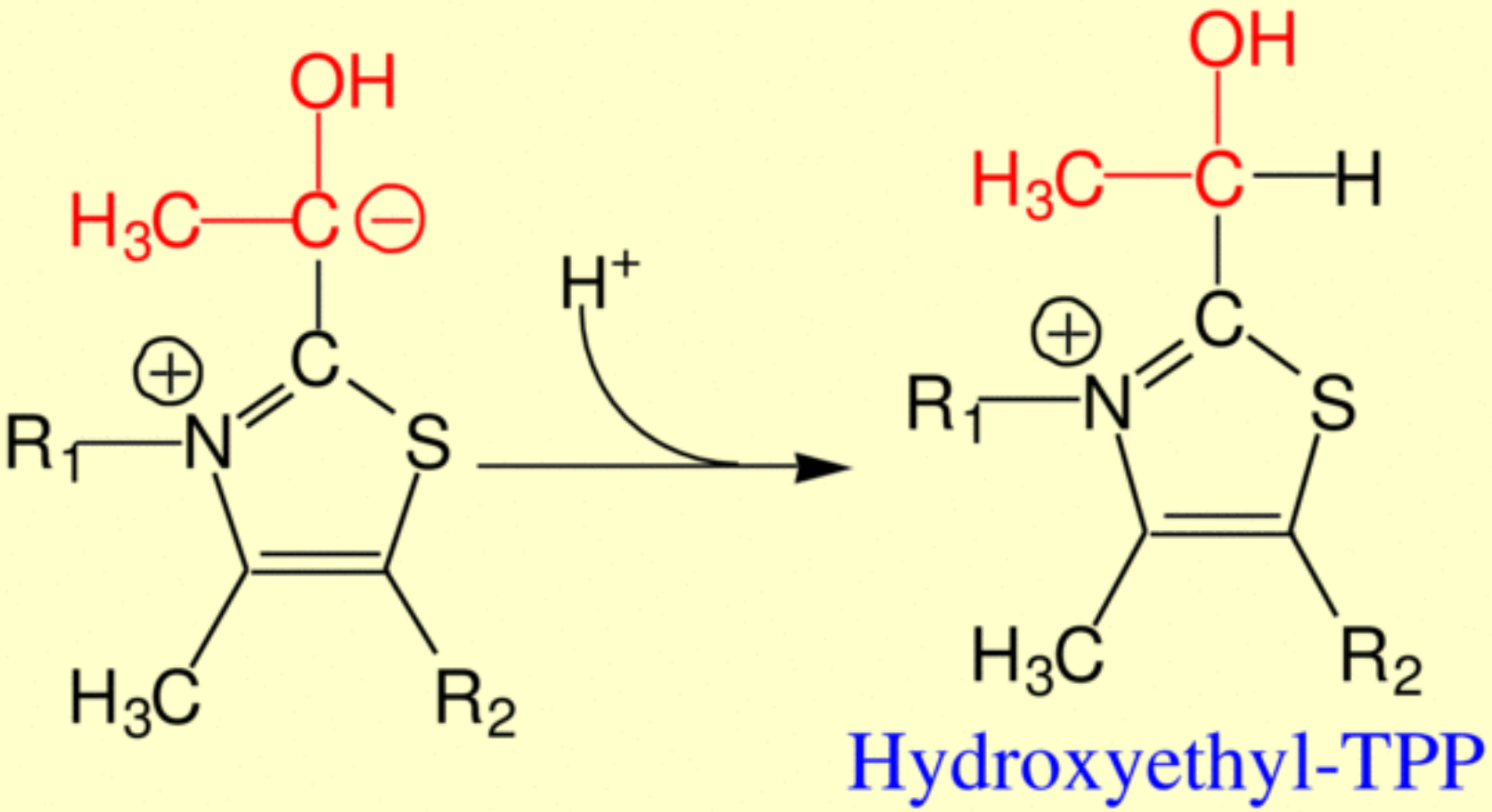
Pyruvate Dehydrogenase Mechanism



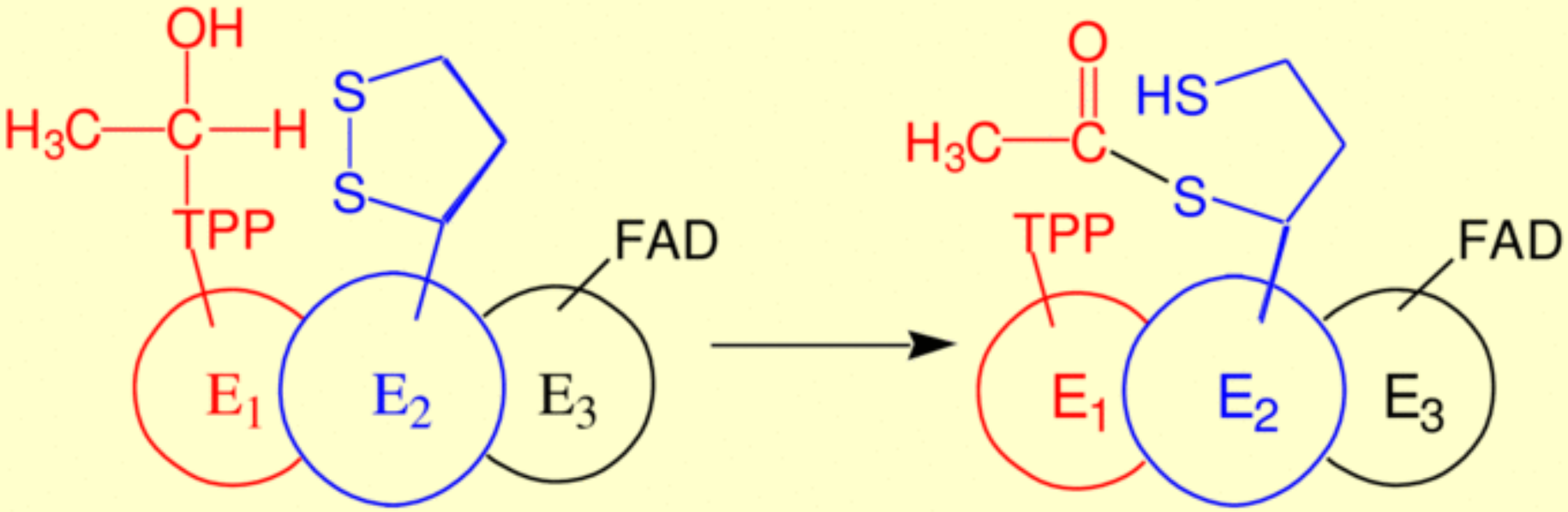
Pyruvate Dehydrogenase Mechanism



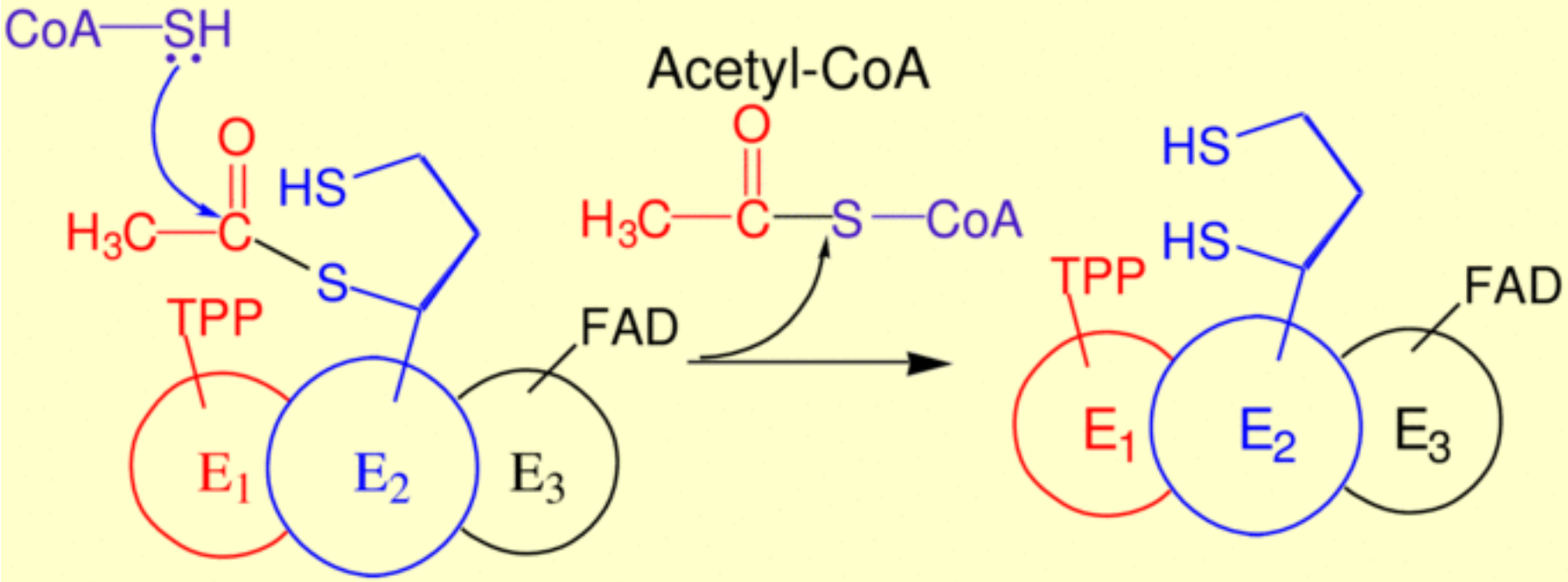
Pyruvate Dehydrogenase Mechanism



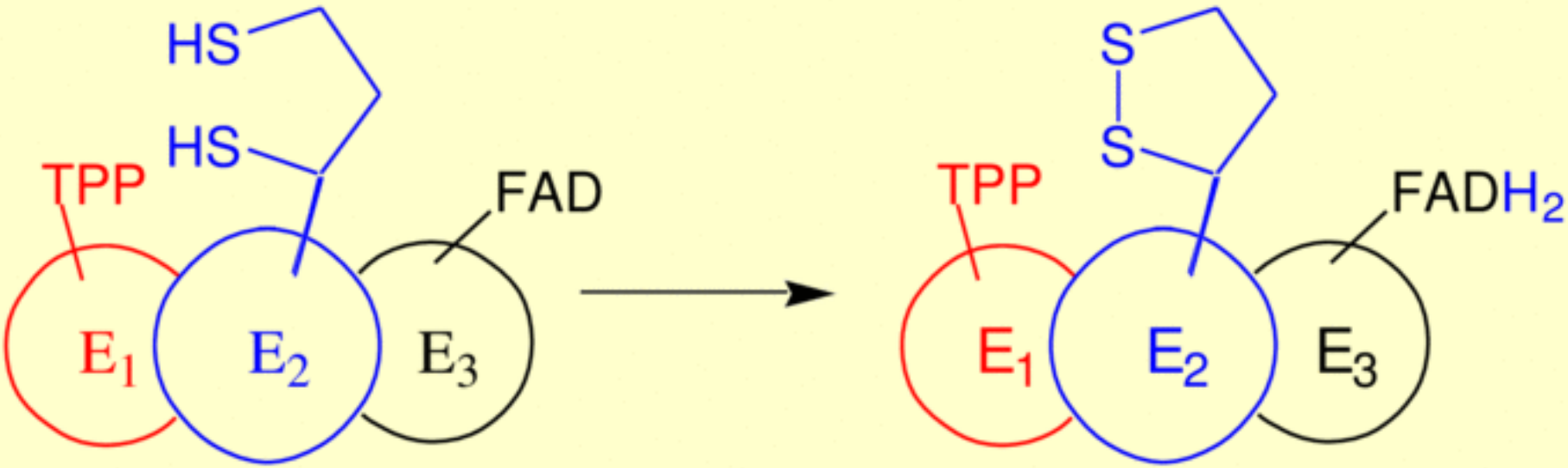
Pyruvate Dehydrogenase Mechanism



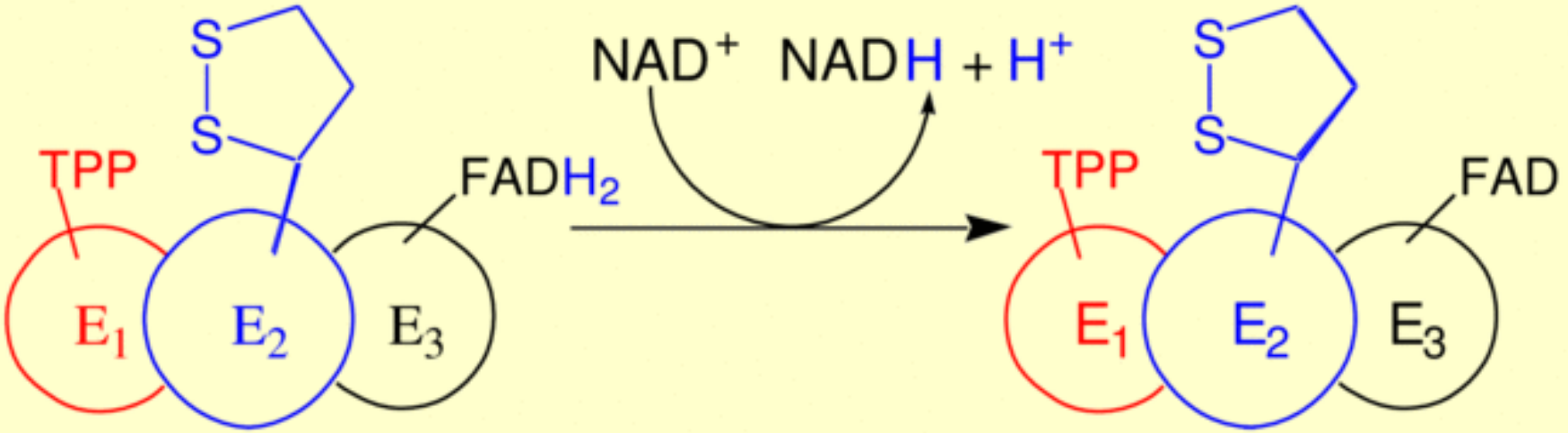
Pyruvate Dehydrogenase Mechanism



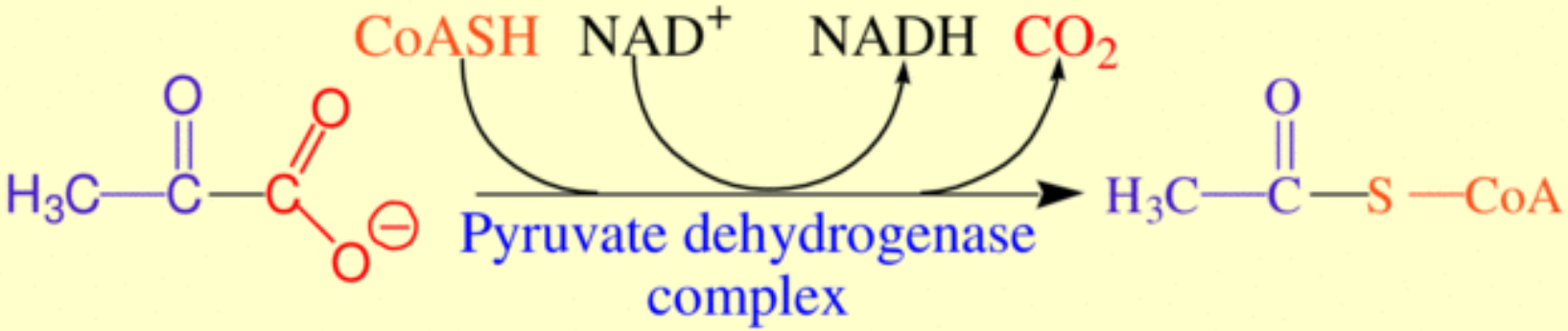
Pyruvate Dehydrogenase Mechanism



Pyruvate Dehydrogenase Mechanism



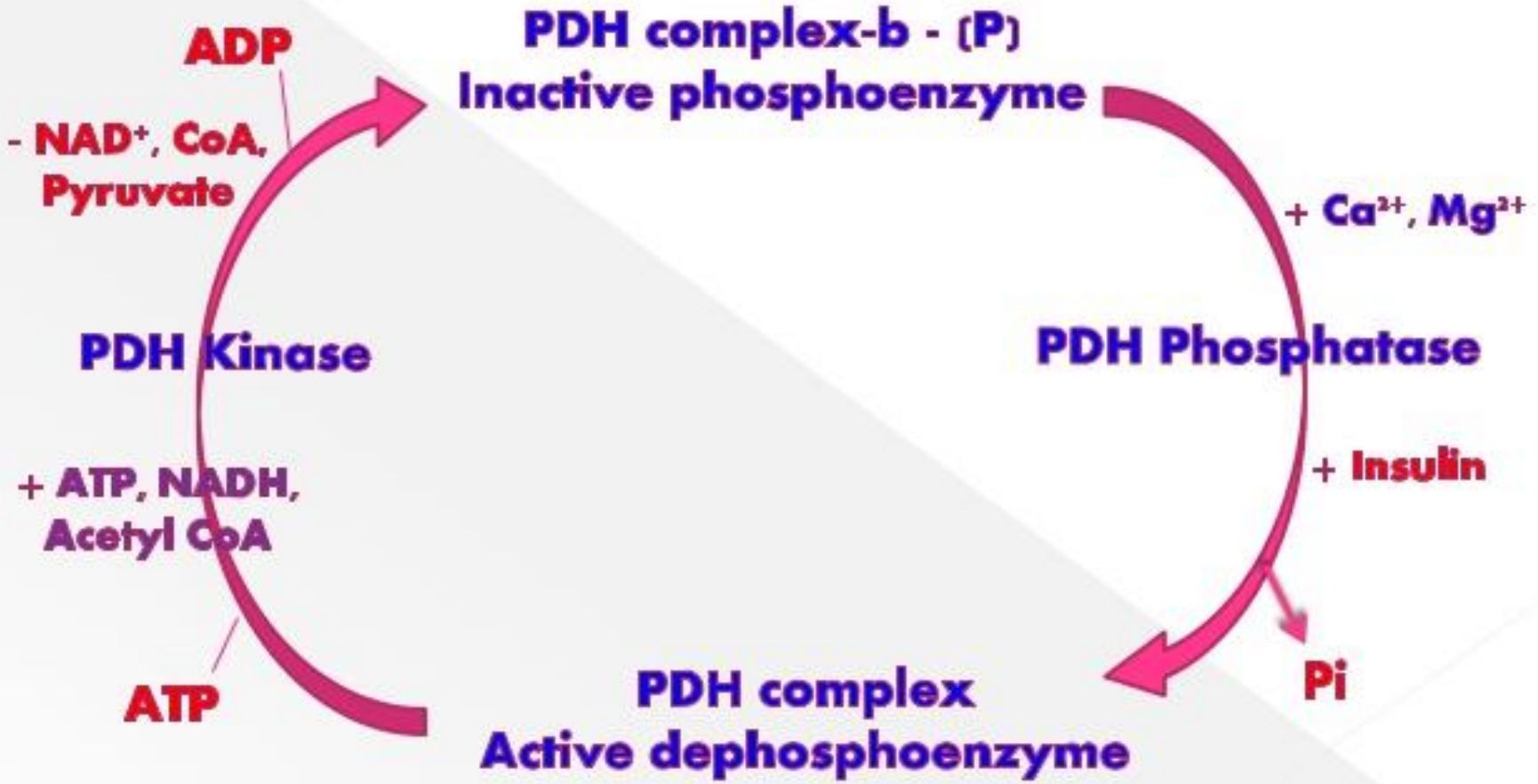
PDH: The Overall Reaction



Acetyl-CoA enter Krebs cycle

NADH passes $e^{\pm}$ to O_2

Regulation of PDH



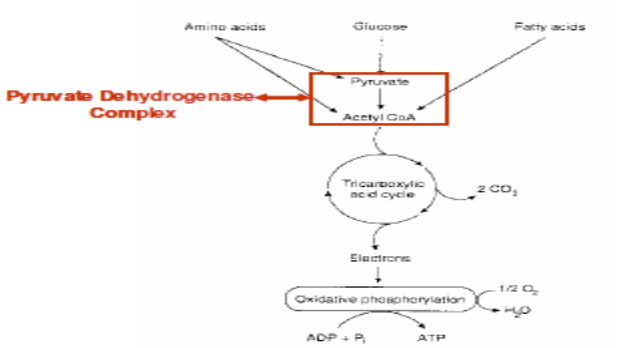
- Calcium released during muscle contraction stimulates PDH (by increasing phosphatase activity) for energy Production.
- PDH kinase (responsible to form inactive PDH) is promoted by ATP, NADH & acetyl CoA, while it is inhibited by NAD^+, CoA & pyruvate.
- In the presence of high energy signals (ATP, NADH), the PDH is turned off.

Biochemical importance of PDH

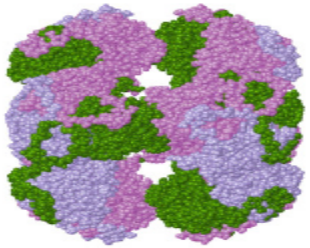
- Lack of TPP (Deficiency of B_1) inhibits PDH activity & causes accumulation of Pyruvate.
- In thiamine deficient alcoholics, pyruvate is rapidly converted to lactate, resulting in lactic acidosis.
- In patients with inherited deficiency of PDH, lactic acidosis (usually after glucose load) is observed.

CONVERSION OF PYRUVATE TO ACETYL CoA

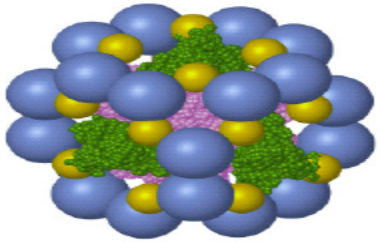
The pyruvate dehydrogenase complex LINKS GLYCOLYSIS TO THE TCA CYCLE! - also occurs in mitochondria



(a)



(b)



Structure of the pyruvate dehydrogenase (PDH) complex

- **Pyruvate dehydrogenase complex (PDH complex) is a multienzyme complex containing:**

  3 enzymes + 5 coenzymes + other proteins

  (+ ATP coenzyme as a regulator)

  E1 = pyruvate dehydrogenase

  E2 = dihydrolipoamide acetyltransferase

  E3 = dihydrolipoamide dehydrogenase

Table 16.1

Enzymes and coenzymes of the pyruvate dehydrogenase complex

Enzyme	Abbreviation	Coenzyme
Pyruvate dehydrogenase	E_1	Thiamine pyrophosphate (TPP)
Dihydrolipoyl transacetylase	E_2	Lipoamide, coenzyme A (CoASH)
Dihydrolipoyl dehydrogenase	E_3	Flavin adenine dinucleotide (FAD), nicotinamide adenine dinucleotide (NAD^+)

Table 16-1 Concepts in Biochemistry, 3/e

Overall reaction of pyruvate dehydrogenase complex

- **Multienzyme Complex (36 subunits!)**
- pyruvate + CoASH + NAD+ → acetyl-CoA + CO2 + NADH + H+