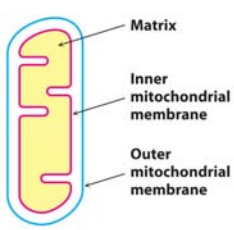
Preparation for Citric Acid Cycle

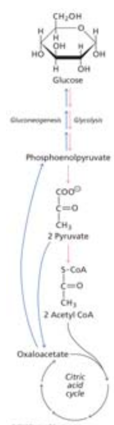
Chapter 18

Stryer Short Course

Overview

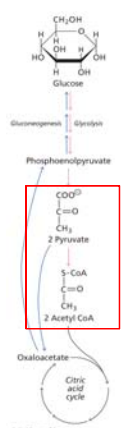
- Compartmentalization
  - Glycolysis: Cytosol
  - Citric Acid Cycle: mitochondria

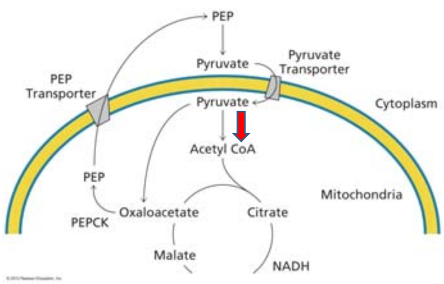




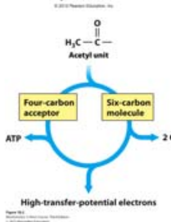
Bridge

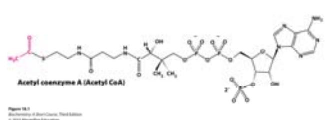
- Glycolysis
- Pyruvate dehydrogenase complex
  - **Commitment of carbon away from carbohydrates**
- Citric acid cycle





The Preparation Reaction





Pyruvate Dehydrogenase Complex

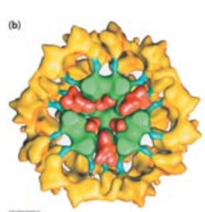
- Three distinct enzymes—in a massive complex
- Five chemical steps

Table 18.1 Pyruvate dehydrogenase complex of *E. coli*

Enzyme	Abbreviation	Number of chains	Prosthetic group	Reaction catalyzed
Pyruvate dehydrogenase component	E_1	24	TPP	Oxidative decarboxylation of pyruvate
Dihydrolipoyl transacetylase	E_2	24	Lipoamide	Transfer of acetyl group to CoA
Dihydrolipoyl dehydrogenase	E_3	12	FAD	Regeneration of the oxidized form of lipoamide

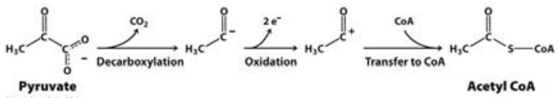
Abbreviations: TPP, thiamine pyrophosphate; FAD, flavin adenine dinucleotide.

(b)



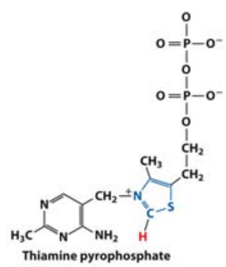
Formal Reactions

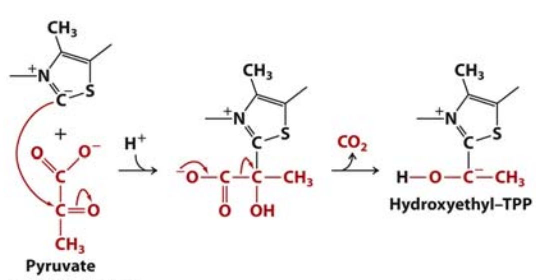
- Where are the problems in this formal reaction?
- Cofactors used to overcome problems!



Pyruvate Dehydrogenase (E_1)

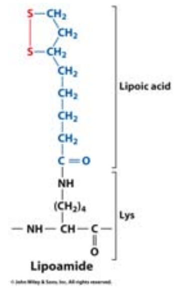
- TPP cofactor: Decarboxylation of α carboxyketones
- Stabilization of "acyl anion"
- Draw mechanism of decarboxylation

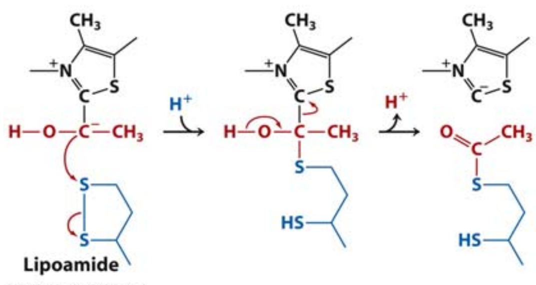




Dihydrolipoamide Acyltransferase (E_2)

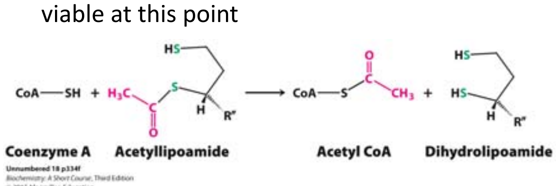
- Transfer catalyzed by E1
- Serves as an linker to "swing" substrate through subunits
- Mechanism of redox





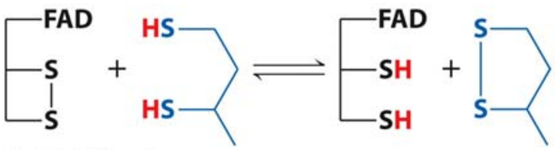
Step 3: transfer

- Maintenance of high energy bond
- Acetyl CoA product is made
- Lipoamide still reduced—not catalytically viable at this point

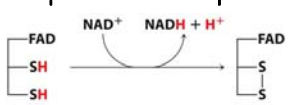


Dihydrolipoamide dehydrogenase (E_3)

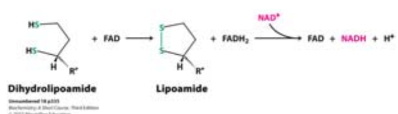
- Redox of prosthetic $FAD/FADH_2$
- Still not a regenerated catalyst!



Step 5: NADH produced

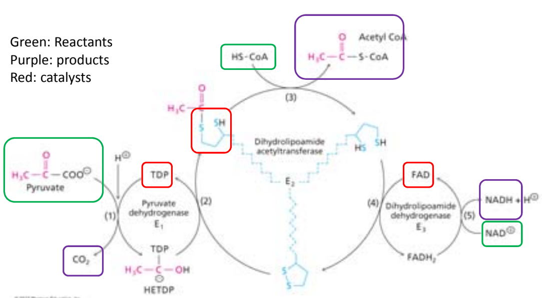


- Catalytic prosthetic group is restored by stoichiometric NAD+
- Overall redox:

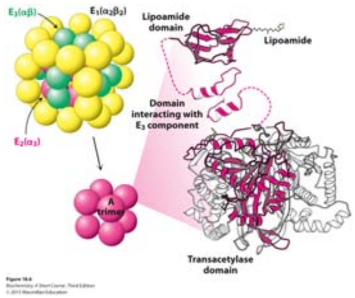


Oxidation energy of one carbon atom used to produce high energy thioester and NADH

Green: Reactants
Purple: products
Red: catalysts



Complex Structure



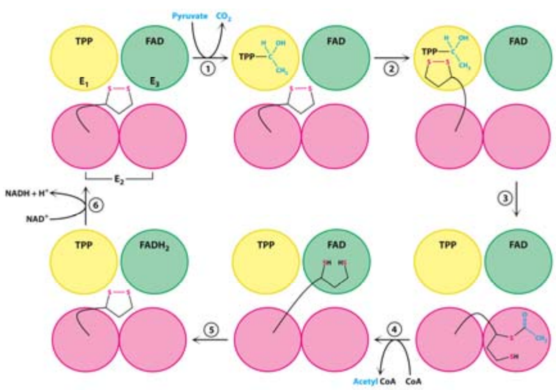
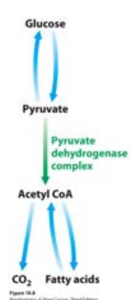


Figure 18.7
Biochemistry: A Short Course, Third Edition
© 2015 Macmillan Education

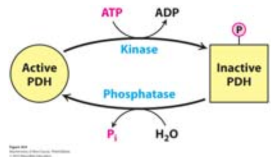
Regulation

- Metabolically irreversible
- Local regulation conserves glucose
- Pyruvate Dehydrogenase Complex inhibited by product
  - NADH
  - Acetyl CoA
- When these are at high concentration in mitochondria, pyruvate is not wasted



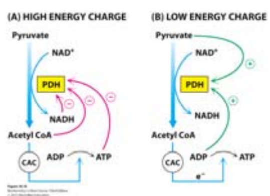
Covalent Regulation

- Kinase and phosphatase activity built into core of PDH
  - Kinase inactivates PDH
  - Phosphatase activates PDH
- Activity controlled by
  - Local energy charge
  - Hormones



Energy Charge Regulation

- High energy charge activates PDH kinase
- Low energy charge activates PDH phosphatase



Hormone Regulation

- α-Adrenergic receptor
  - Liver
  - Phosphoinositol pathway
  - Activates PDH phosphatase
  - More acetyl CoA
  - Purpose: Energy
- Insulin
  - Liver, adipose
  - Insulin receptor
  - Activates PDH phosphatase
  - More acetyl CoA
  - Purpose: store as fatty acids

Related Pathologies

- PDH can be inactivated by
  - Genetic defect of PDH phosphatase
  - cancer enhancement of PDH kinase
  - Beriberi (TPP deficiency)
  - Arsenic (binds lipamide)
- Results in
  - No entrance from glucose into citric acid cycle
  - Glucose metabolized anaerobically
  - Especially bad for nervous system, which can only make acetyl CoA from glucose (others can use fats to make acetyl CoA)