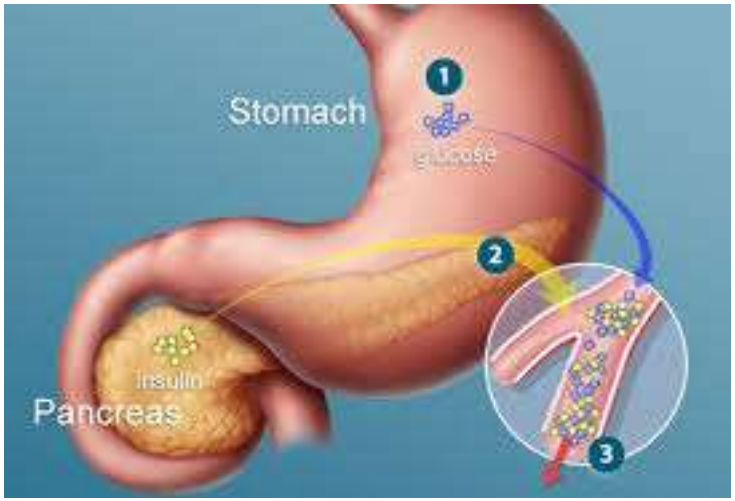
Glycolysis II



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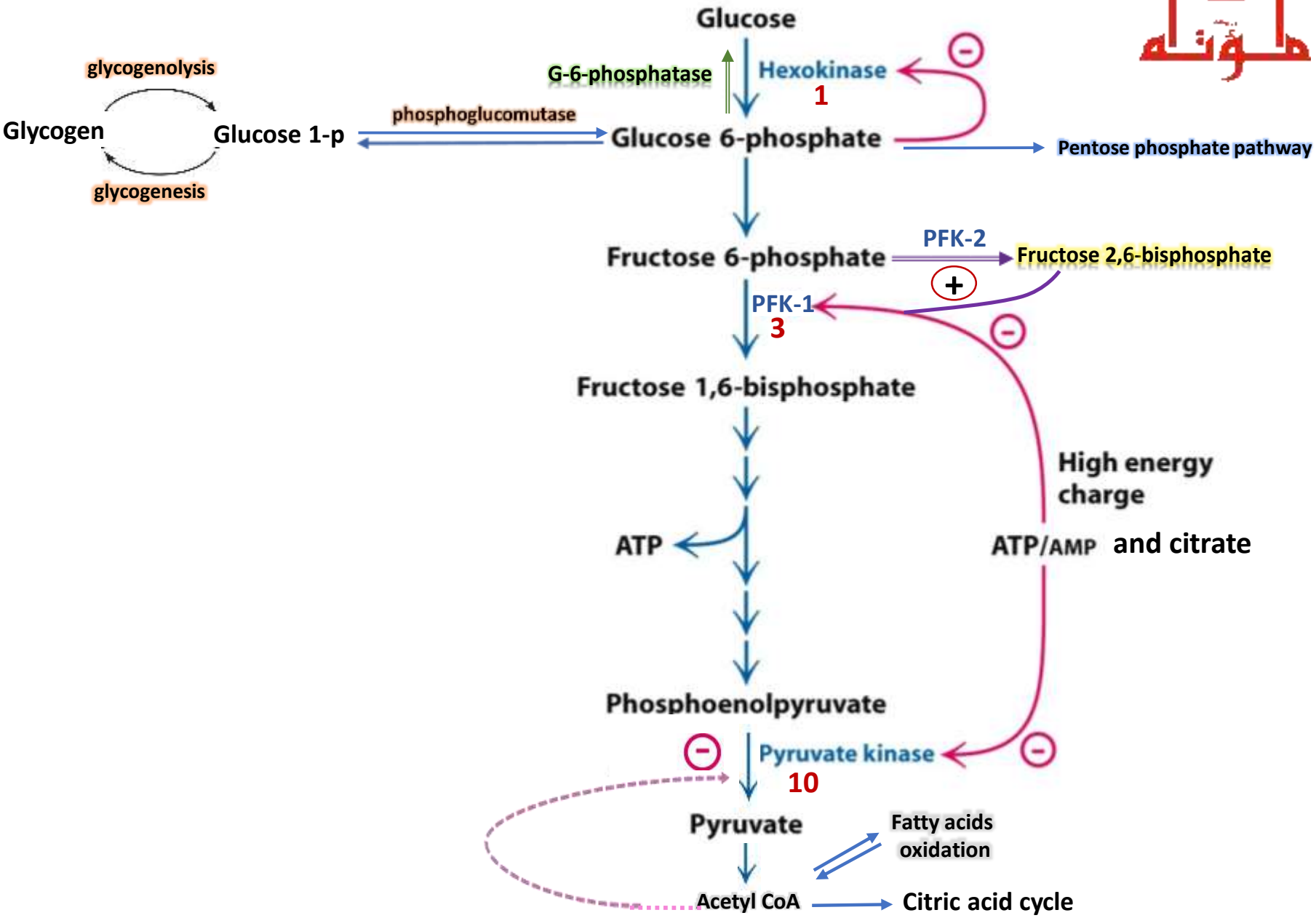
Glycolysis Regulation



Glycolysis Regulation

- Glycolysis can be controlled at 3 points:

1. Step 1 which is catalyzed by hexokinase enzyme (allosteric enzyme). Hexokinase isoforms (except glucokinase) are allosterically inhibited by excess G6P
2. Step 3 which is catalyzed by phosphofructokinase-1 enzyme. It is an allosteric enzyme. Two inhibitors are citrate and ATP whereas AMP and recently fructose 2,6-biphosphate (in liver) are activators. Actually this is the most important control point and it is considered as the main **rate-limiting step** in glycolysis
3. Step 10 which is catalyzed by pyruvate kinase enzyme. It is controlled by the level of ATP and Acetyl CoA (both are allosteric inhibitors). **Accumulated Acetyl CoA in the cytosol** is an indicator that the energy is now available from fat breakdown so no need to proceed in glycolysis

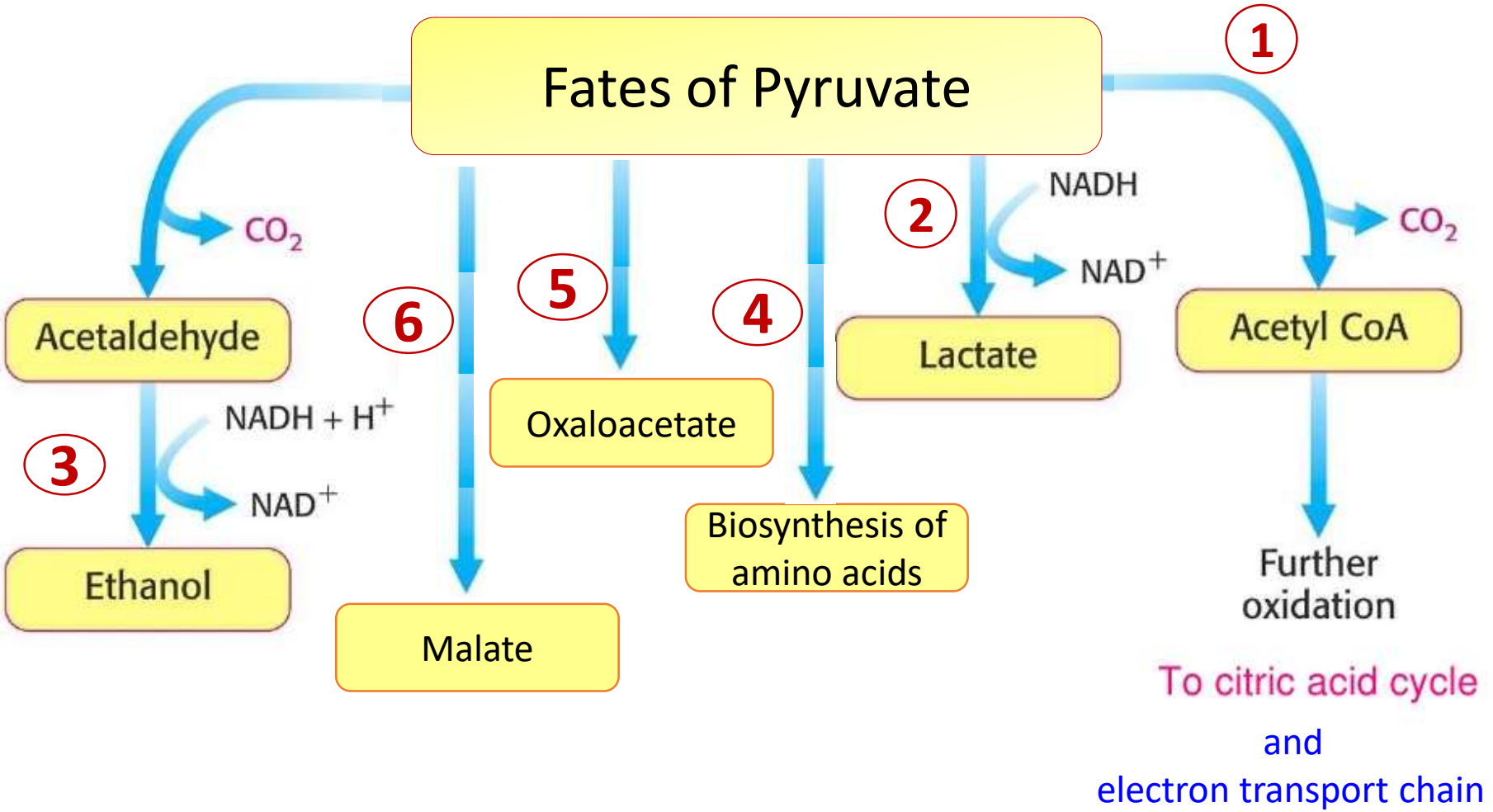
Fluoride as Inhibitor of Enolase

- Oral bacteria depends on the food debris or dietary sugars found on the tooth surface as a primary source of energy. Acids are produced through fermentation process (harmful)
- Fluoride is a competitive inhibitor of enolase enzyme catalyzing Step 9
- Drinking fluoridated water or using a toothpaste containing fluoride inhibit the oral bacteria enolase activity. Consequently, this disrupts the bacteria glycolytic pathway and prevents formation of dental caries

Fluoride as Inhibitor of Enolase

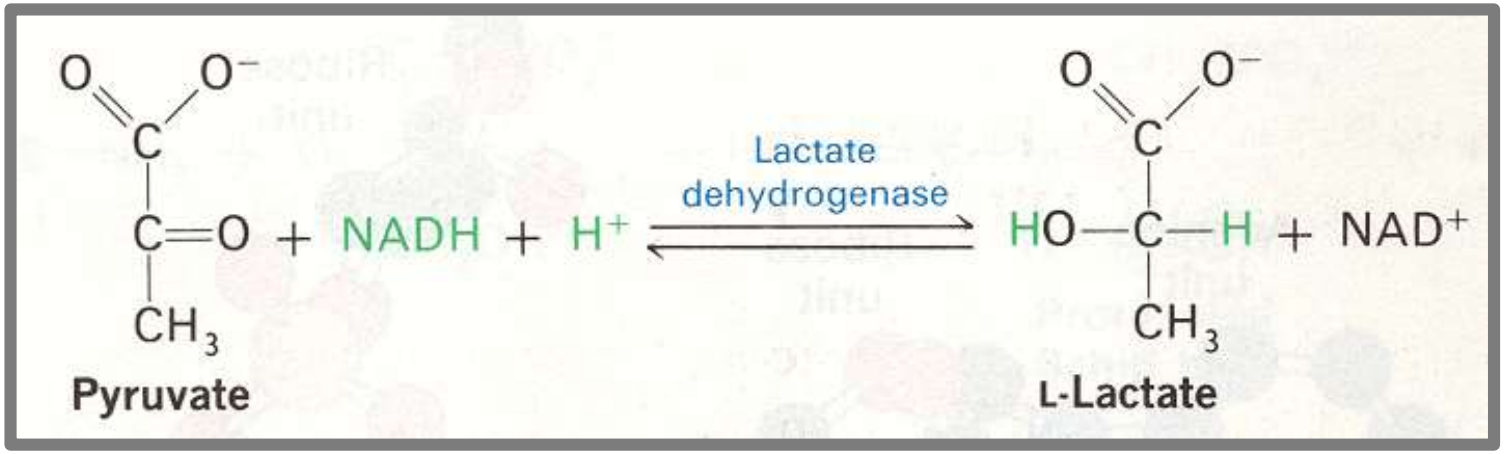
- Sodium fluoride is known to have antiglycolytic effect that inhibits glycolysis by erythrocytes
- NaF tubes (gray top) are widely used for blood collection for glucose measurement
- Fluoride-containing tubes are suitable for blood collection if there is a long delay in blood separation following collection (false negative result)

Metabolic Fates of Pyruvate



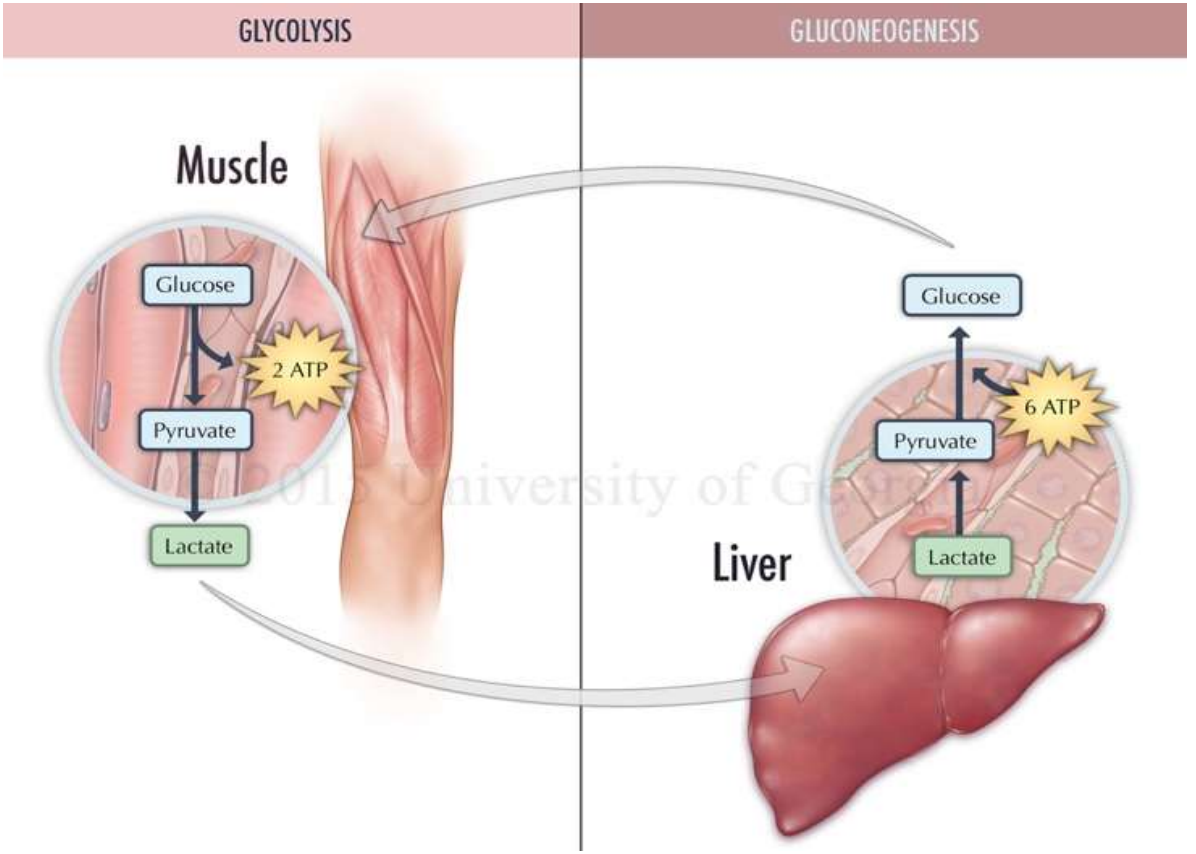
Metabolic Fates of Pyruvate

2. Lactic Acid Fermentation: **bacteria, RBCs and O_2-starved muscle cells**

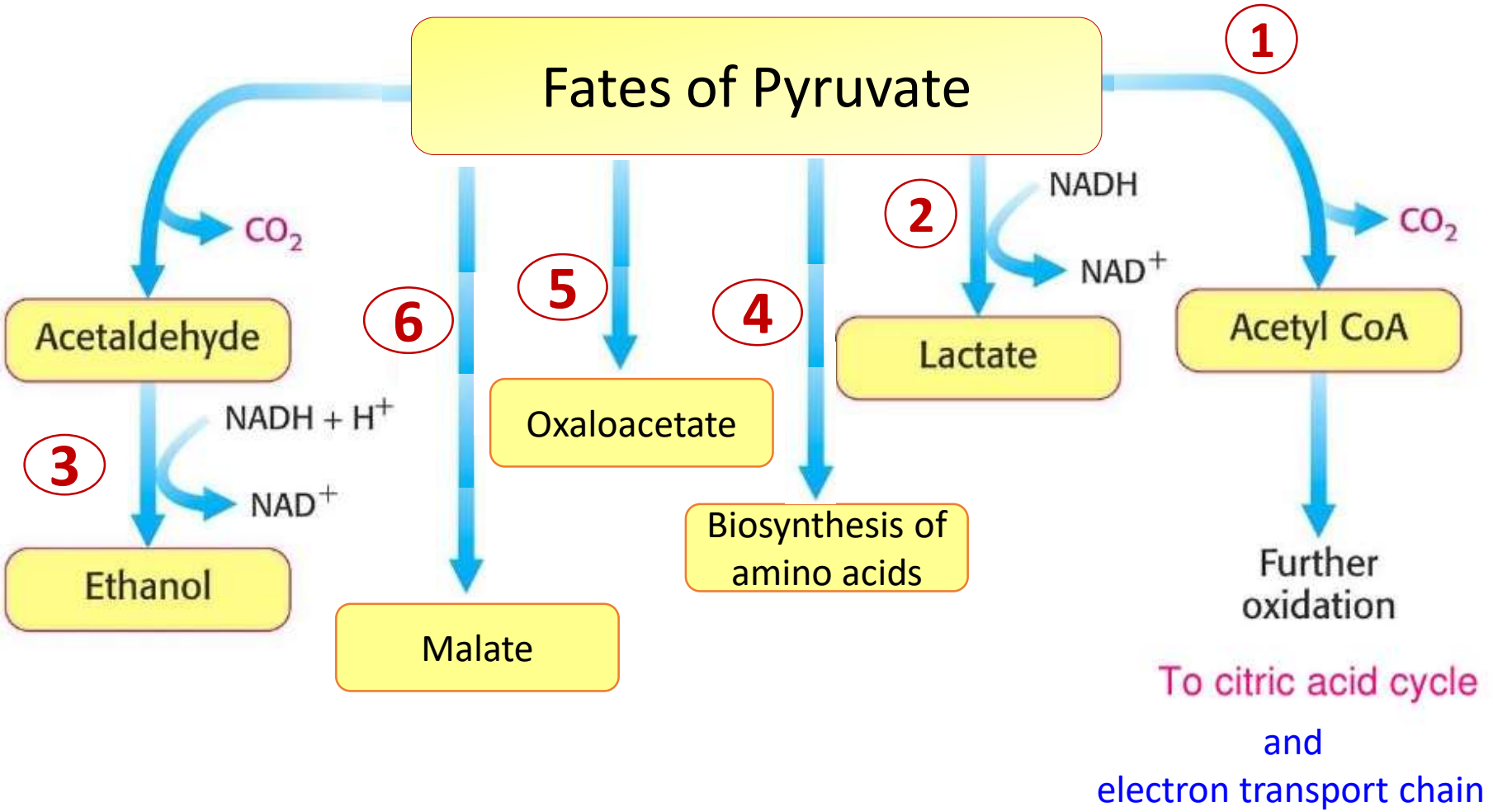


Cori Cycle

- Cori cycle "lactic acid cycle" is the metabolic pathway in which lactic acid produced in muscles during the time of oxygen depletion is converted back to glucose in the liver by gluconeogenesis process



Metabolic Fates of Pyruvate



Metabolic Fates of Pyruvate

1. In aerobic conditions, pyruvate is converted to acetyl CoA which enters the citric acid cycle for further oxidation to CO_2 followed by oxidative phosphorylation
2. In anaerobic conditions like in lactic acid bacteria and some human cells (e.g. RBCs and O_2-starved muscle cells), pyruvate is reduced to lactic acid with the concomitant oxidation of NADH to NAD^+ (lactic acid fermentation)
3. In anaerobic conditions like in some M.O's (e.g. yeast), pyruvate is converted to ethanol
4. Amino acid biosynthesis: pyruvate is a precursor of some amino acids like alanine

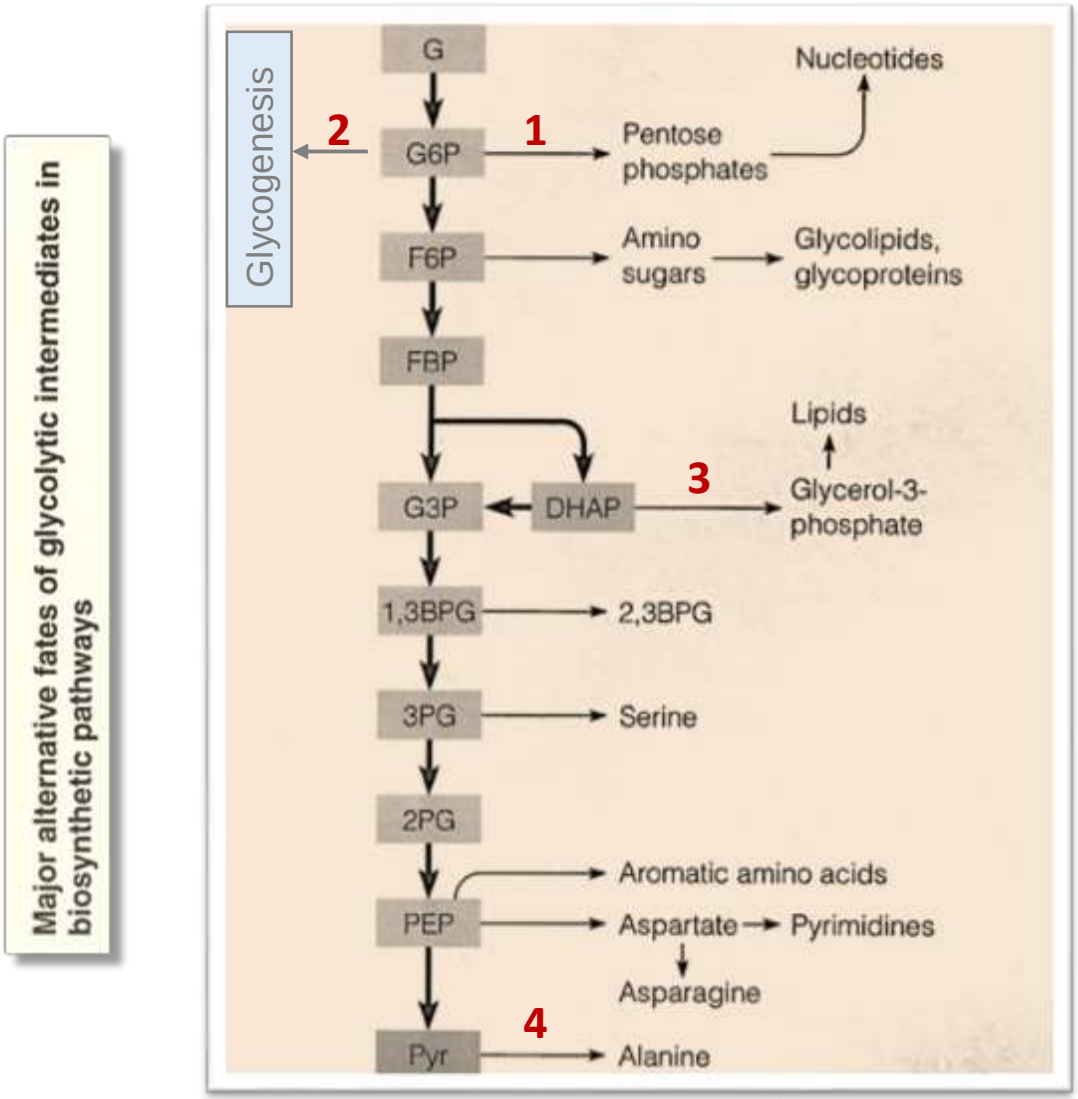
5. & 6. Pyruvate can be used for synthesis of oxaloacetate or malate (both are TCA cycle intermediates)

Glycolysis as Anabolic Pathway

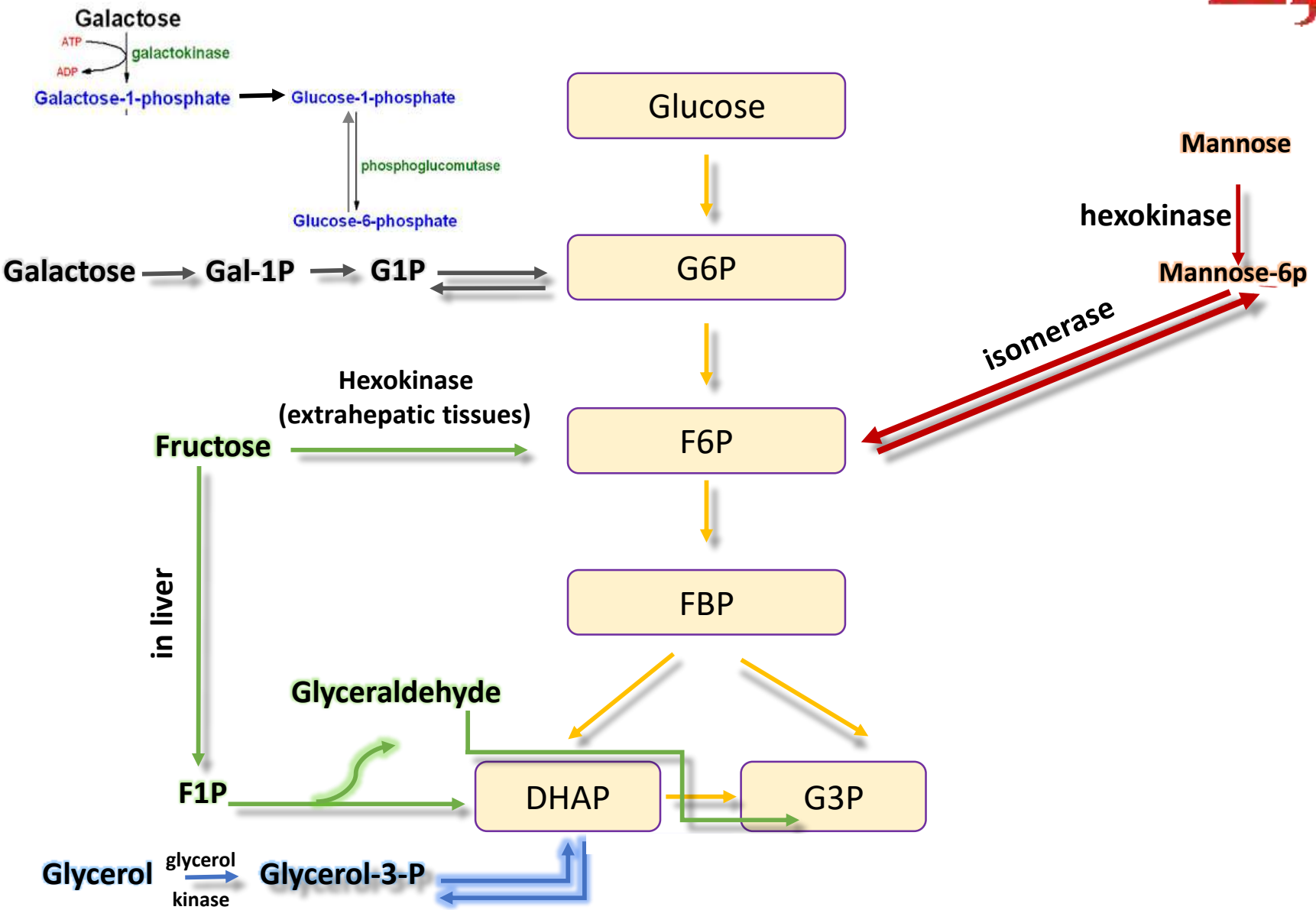
- Glycolysis acts as catabolic as well as anabolic pathway. Therefore, glycolysis is very important central metabolic pathway
- Glycolysis intermediates with biosynthetic roles:

1. Nucleotides biosynthesis: G6P is an initial substrate in pentose phosphate pathway (metabolic pathway which generates pentoses)
2. Glycogenesis via G6P
3. Lipids biosynthesis: DHAP is converted to glycerol
4. Amino acids biosynthesis: pyruvate as precursor of alanine

Glycolysis as Anabolic Pathway

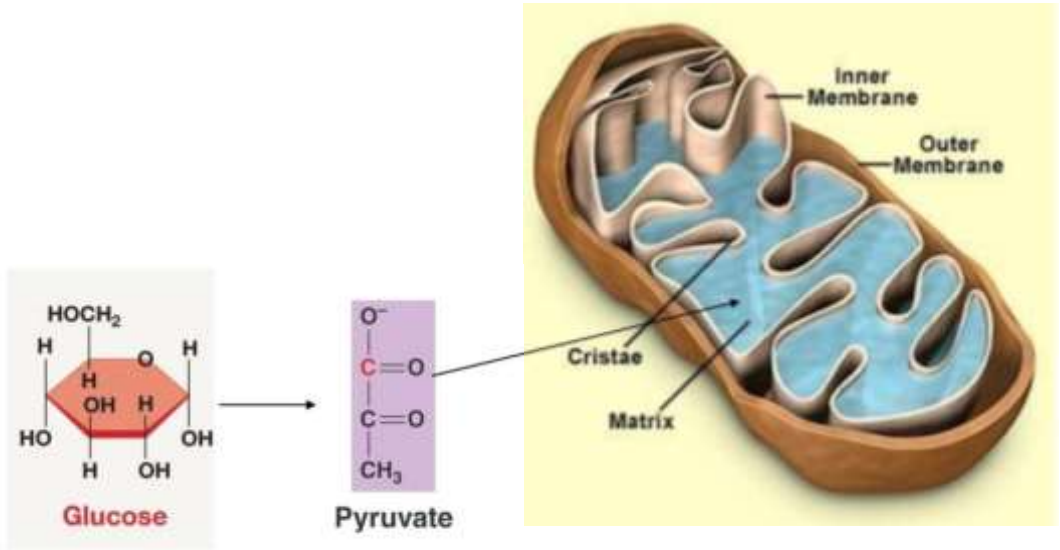


Other substrates enter Glycolysis

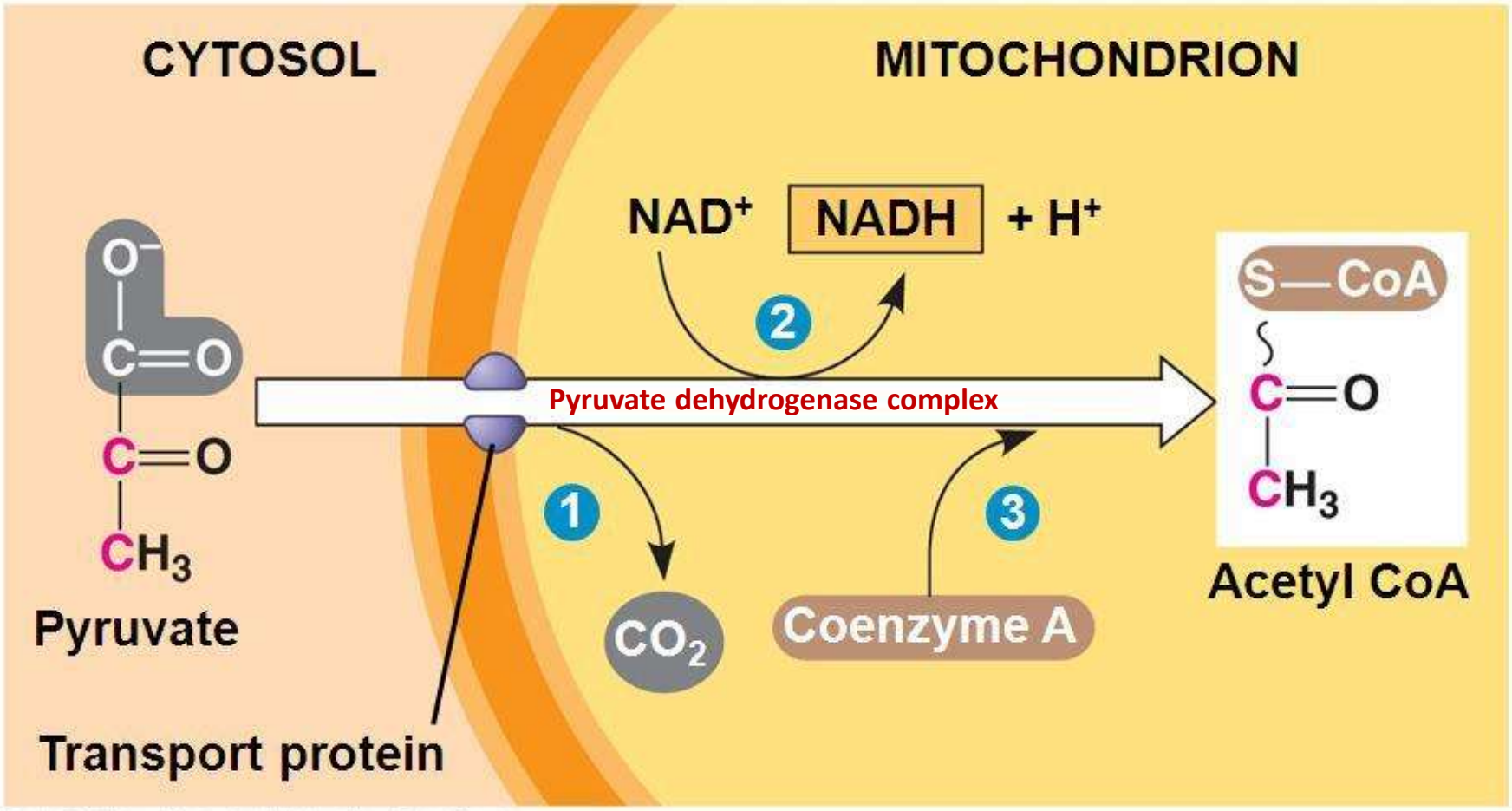


Acetyl CoA Formation

- In aerobic respiration, pyruvate (3C) joins the citric acid cycle after its conversion to acetyl CoA (2C)
- Citric acid cycle occurs in the mitochondrial matrix. Shuttling of pyruvate from the cytosol is facilitated by a transporter protein embedded in the inner mitochondrial membrane called pyruvate translocase



Acetyl CoA Formation

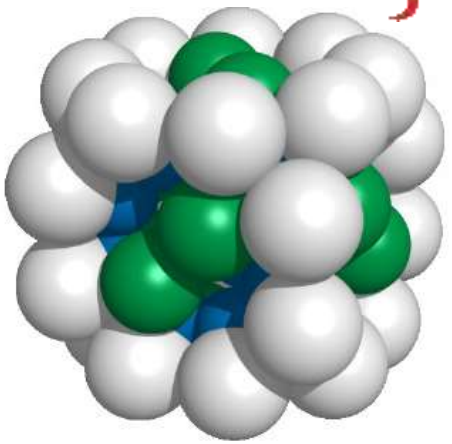


Acetyl CoA Formation

- Pyruvate dehydrogenase complex (PDC) catalyzes the **irreversible** oxidative decarboxylation of pyruvate into Acetyl CoA with the release of CO_2
- Energy-rich molecule “NADH” is also produced
- Coenzyme A (CoA) acts as acetyl group carrier due to its free sulfhydryl (–SH) end capable of forming thioester bond
- PDC is a multi-enzyme system consists of three catalytic enzymes and five coenzymes (three of them are prosthetic groups as they are tightly bound to their corresponding enzymes)

Pyruvate Dehydrogenase Complex

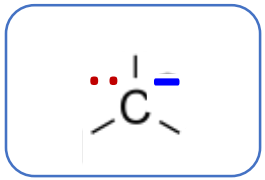
- **E1: pyruvate dehydrogenase**
- **E2: dihydrolipoamide transacetylase**
- **E3: dihydrolipoamide dehydrogenase**

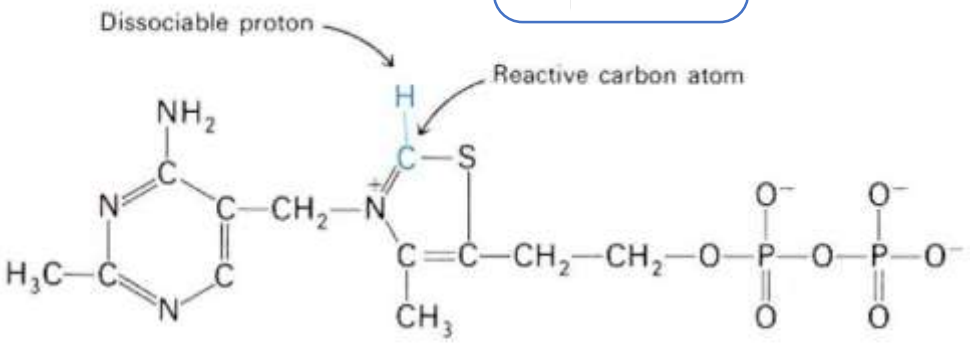


Coenzymes

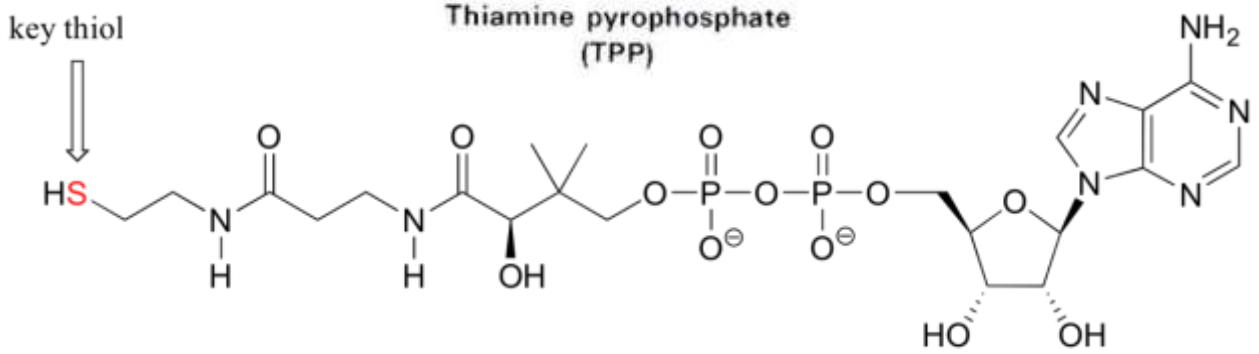
- Thiamine pyrophosphate (TPP) a prosthetic group of **E1**
- Lipoic acid (lipoamide) a prosthetic group of **E2**
- Flavin adenine dinucleotide (FAD) a prosthetic group of **E3**
- Coenzyme A (CoA or CoA-SH)
- Nicotinamide adenine dinucleotide (NAD^+)

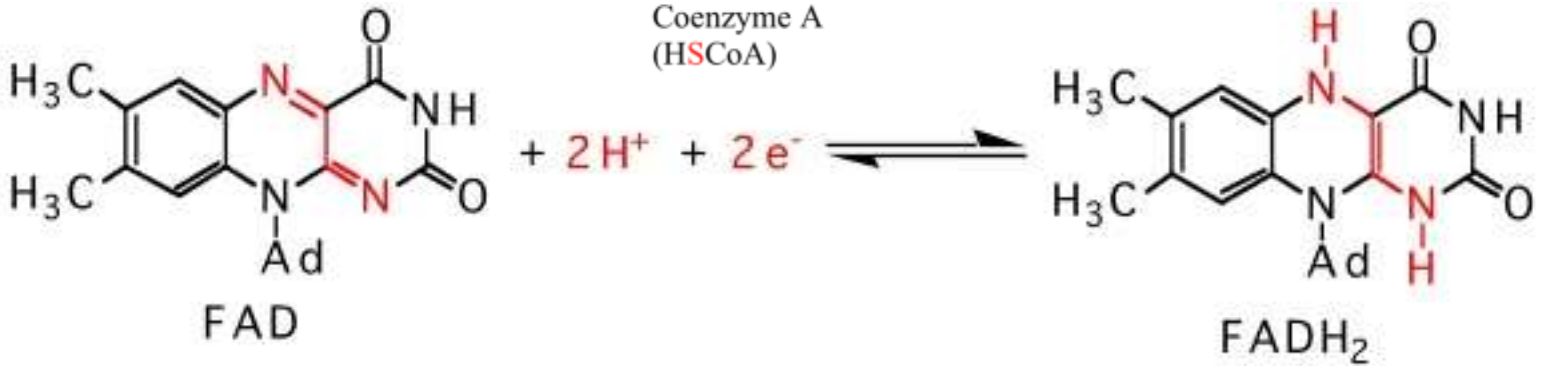
Coenzymes Structure



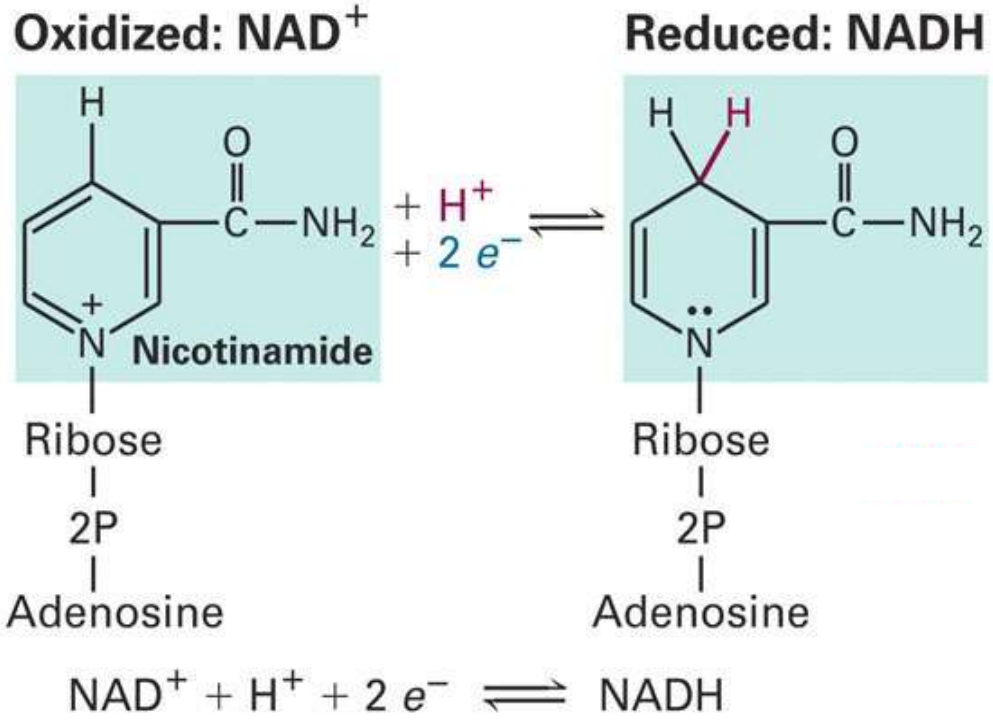


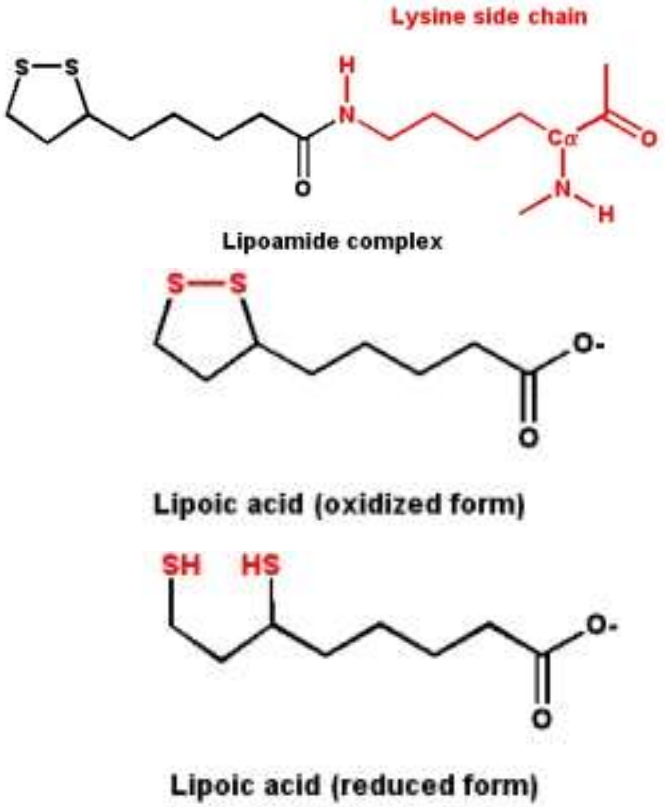
Thiamine pyrophosphate (TPP)



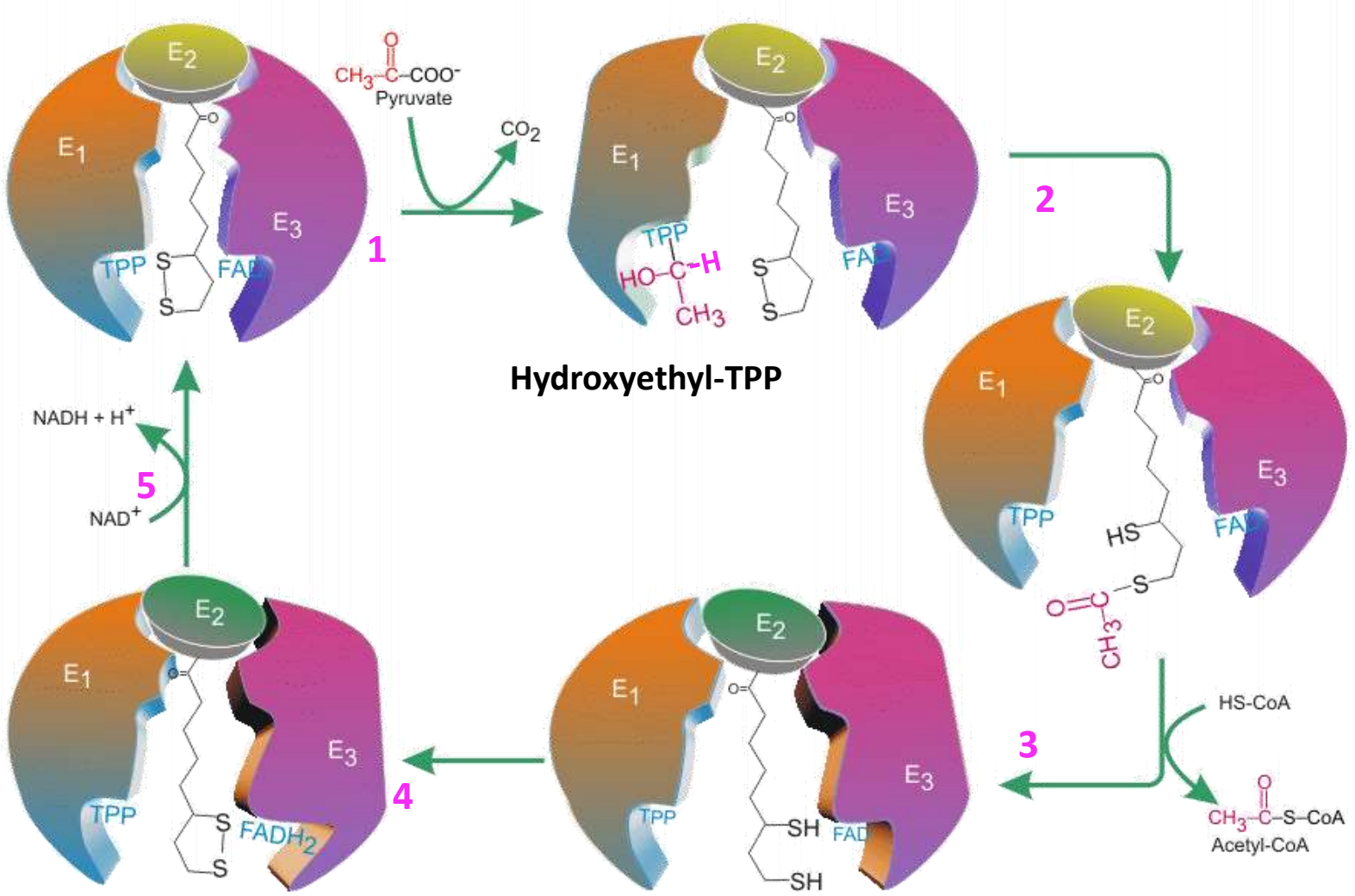


Coenzymes Structure





Pyruvate Dehydrogenase Complex



Mechanism of PDC

- The mechanism by which this complex catalyzes the reaction is complicated but the main processes involve (5 steps):

1. Decarboxylation of pyruvate and the release of CO_2 a reaction catalyzed by E1-TPP. The product of this reaction "Hydroxyethyl moiety" is a substrate for the next reaction
2. The transfer of Hydroxyethyl moiety from TPP of E1 to lipoic acid of E2. This step is mediated by an oxidation of Hydroxyethyl to acetyl group coupled with reduction of disulfide bond
3. Transfer of acetyl group from lipoamide to CoA forming thioester bond and consequently Acetyl CoA is produced

Mechanism of PDC

4. Regeneration of disulfide bond of lipoamide via FAD (E3 prosthetic group) which is reduced to FADH2
5. Regeneration of FAD by NAD^+ which is reduced to NADH with the electrons transferred during the reaction (originally from Hydroxyethyl oxidation)