Glycolysis

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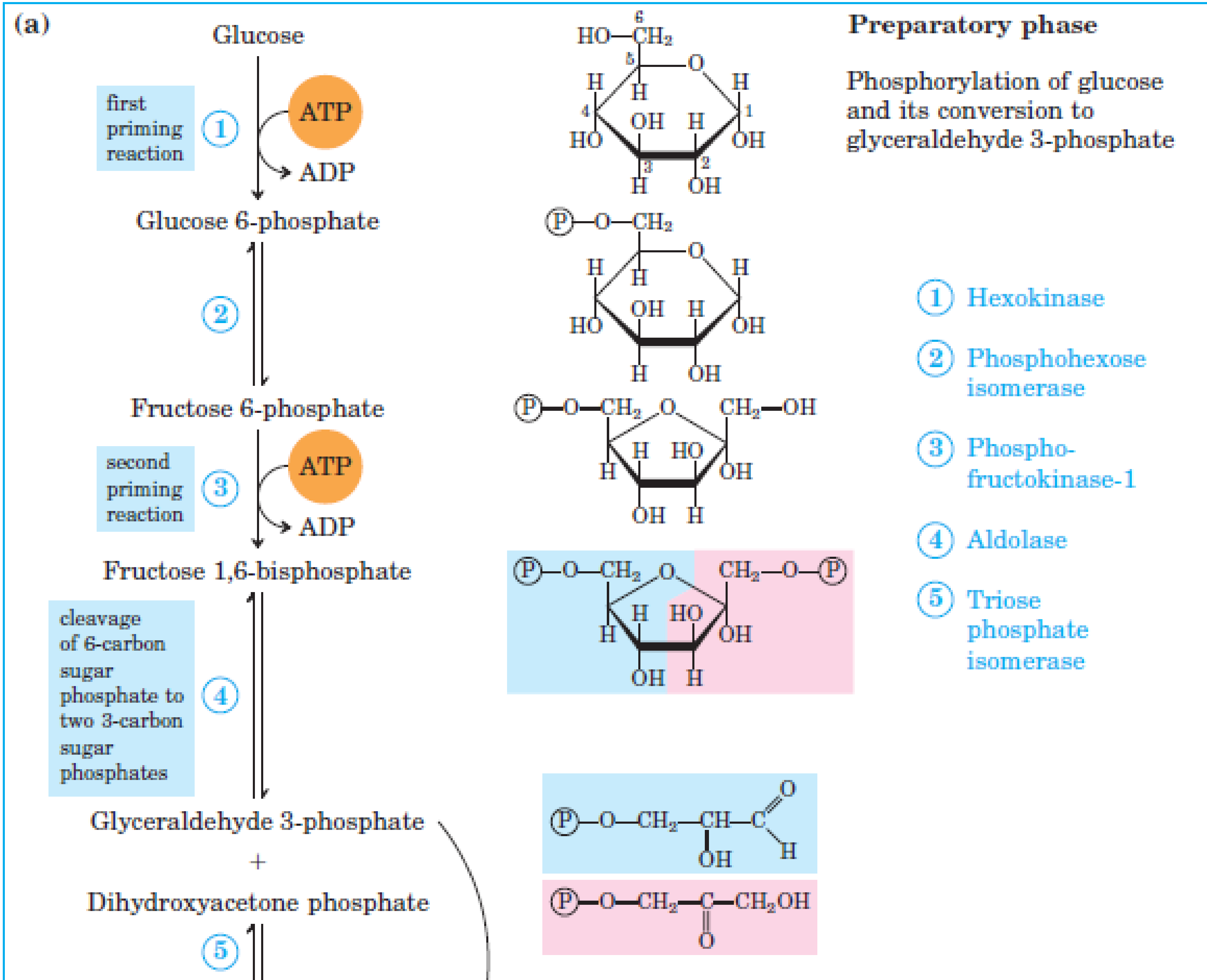
Glycolysis

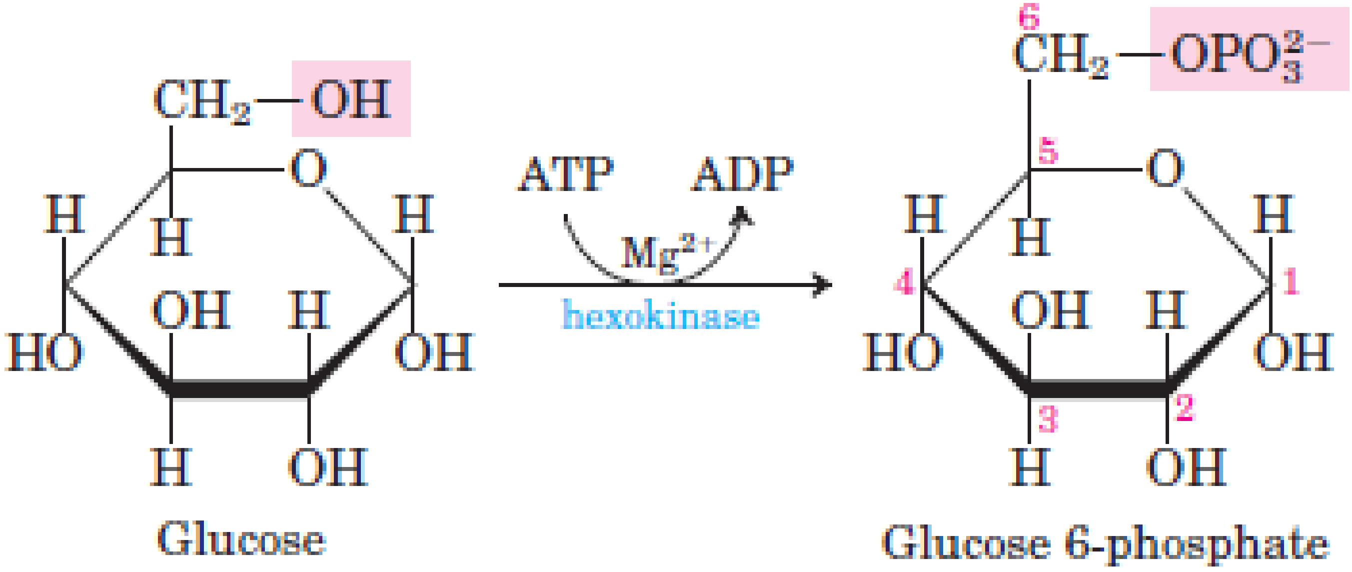
- The Embden-Meyerhof or glycolytic pathway is undoubtedly the most common pathway for glucose degradation to pyruvate in stage two of catabolism.
- It is found in all major groups of microorganisms and functions in the presence or absence of O_2.
- Glycolysis is located in the cytoplasmic matrix of procaryotes and eucaryotes.
- The pathway as a whole may be divided into two parts.
- In the initial six-carbon stage **(Preparatory stage)**, glucose is phosphorylated twice and eventually converted to fructose 1,6-bisphosphate.
- This preliminary stage does not yield energy; in fact, two ATP molecules are expended for each glucose.
- These initial steps "prime the pump" by adding phosphates to each end of the sugar.
- The phosphates will soon be used to make ATP.



(a)
Preparatory phase
Phosphorylation of glucose and its conversion to glyceraldehyde 3-phosphate
Glucose
first priming reaction
1
ATP
ADP
Glucose 6-phosphate
2
Fructose 6-phosphate
second priming reaction
3
ATP
ADP
Fructose 1,6-bisphosphate
cleavage of 6-carbon sugar phosphate to two 3-carbon sugar phosphates
4
Glyceraldehyde 3-phosphate
+
Dihydroxyacetone phosphate
5
1 Hexokinase
2 Phosphohexose isomerase
3 Phospho-fructokinase-1
4 Aldolase
5 Triose phosphate isomerase

Preparatory Phase

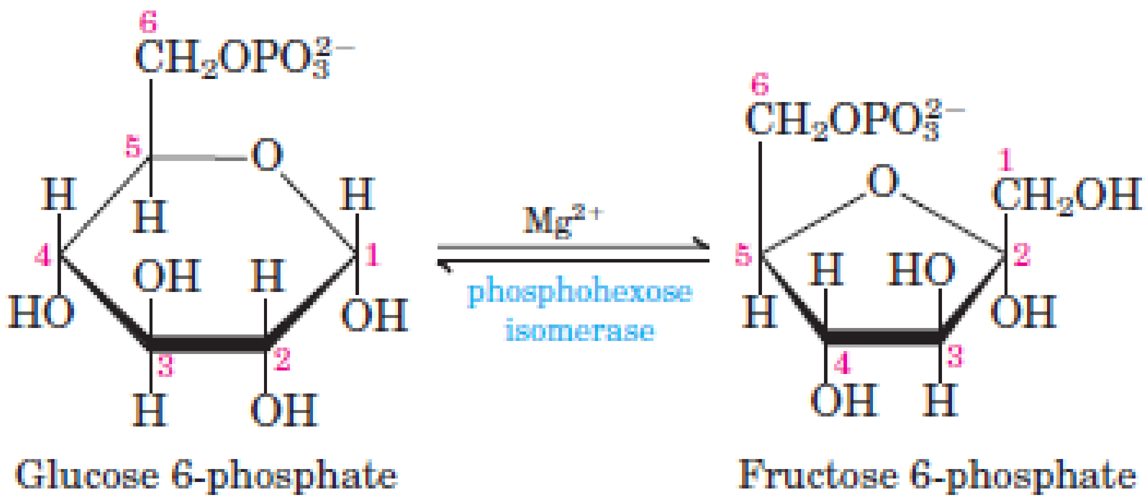
① ***Phosphorylation of Glucose*** In the first step of glycolysis, glucose is activated for subsequent reactions by its phosphorylation at C-6 to yield **glucose 6-phosphate,** with ATP as the phosphoryl donor:



$\Delta G'^{\circ} = -16.7$ kJ/mol

… Preparatory Phase

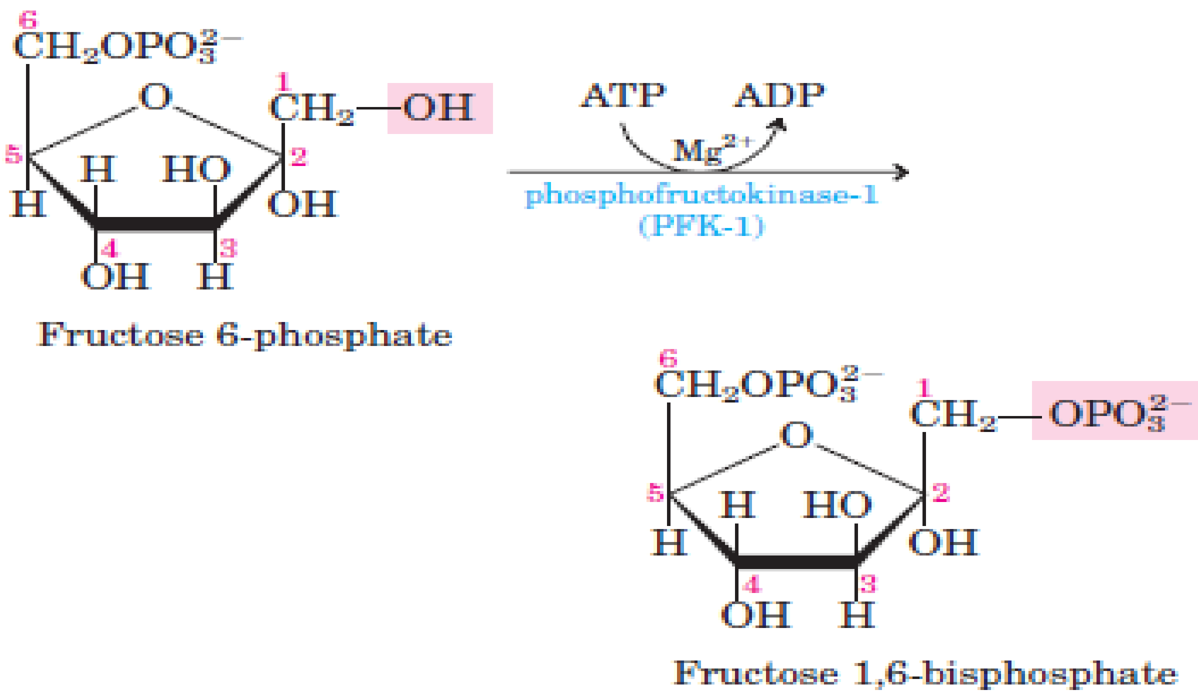
(2) *Conversion of Glucose 6-Phosphate to Fructose 6-Phosphate*
The enzyme **phosphohexose isomerase (phosphoglucose isomerase)** catalyzes the reversible isomerization of glucose 6-phosphate, an aldose, to **fructose 6-phosphate,** a ketose:



$\Delta G'^{\circ} = 1.7$ kJ/mol

… Preparatory Phase

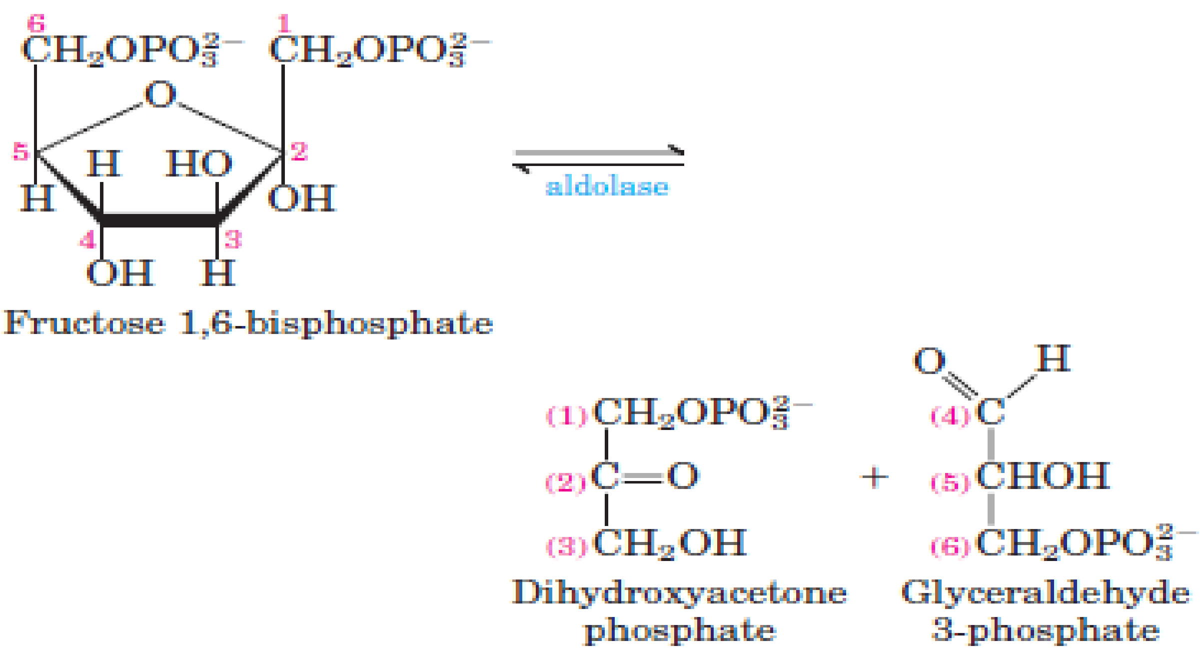
③ ***Phosphorylation of Fructose 6-Phosphate to Fructose 1,6-Bisphosphate*** In the second of the two priming reactions of glycolysis, **phosphofructokinase-1 (PFK-1)** catalyzes the transfer of a phosphoryl group from ATP to fructose 6-phosphate to yield **fructose 1,6-bisphosphate:**



$$\Delta G'^{\circ} = -14.2 \text{ kJ/mol}$$

… Preparatory Phase

④ ***Cleavage of Fructose 1,6-Bisphosphate*** The enzyme **fructose 1,6-bisphosphate aldolase,** often called simply **aldolase,** catalyzes a reversible aldol condensation (p. 485). Fructose 1,6-bisphosphate is cleaved to yield two different triose phosphates, **glyceraldehyde 3-phosphate,** an aldose, and **dihydroxyacetone phosphate,** a ketose:

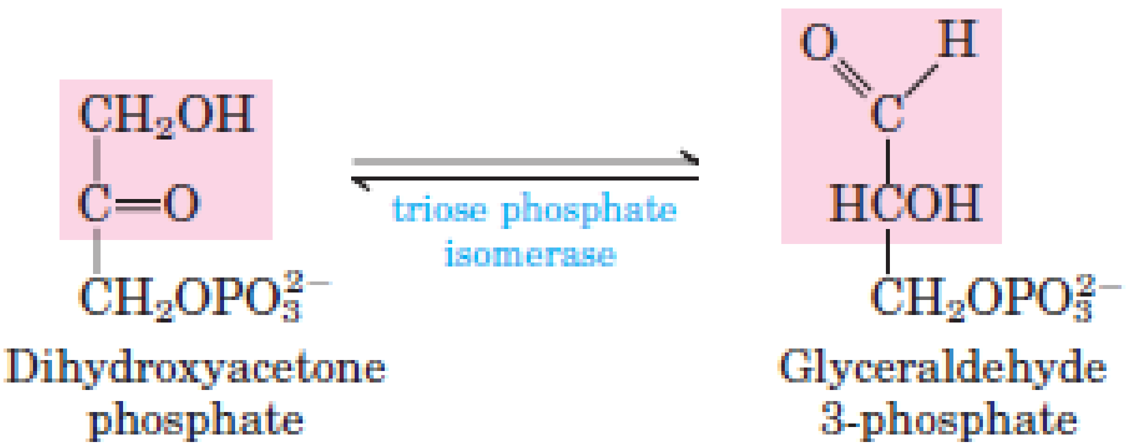


$\Delta G'^{\circ} = 23.8$ kJ/mol

… Preparatory Phase

(5) Interconversion of the Triose Phosphates

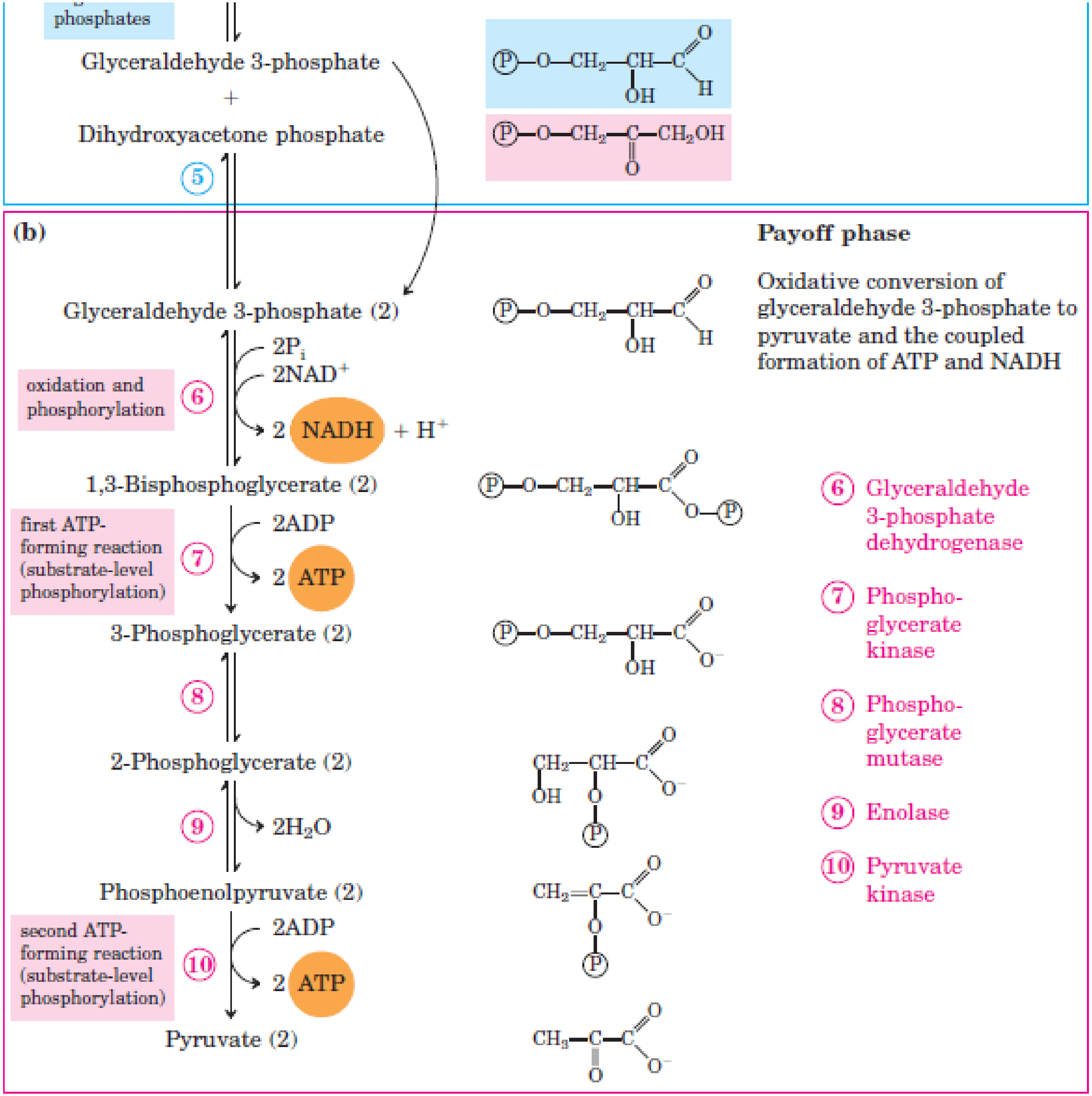
- *Only one of the* two triose phosphates formed by aldolase, glyceraldehyde 3-phosphate, can be directly degraded in the subsequent steps of glycolysis.
- The other product, dihydroxyacetone phosphate, is rapidly and reversibly converted to glyceraldehyde 3-phosphate by the fifth enzyme of the sequence, triose phosphate isomerase:



$\Delta G'^{\circ} = 7.5\ kJ/mol$

Payoff Phase

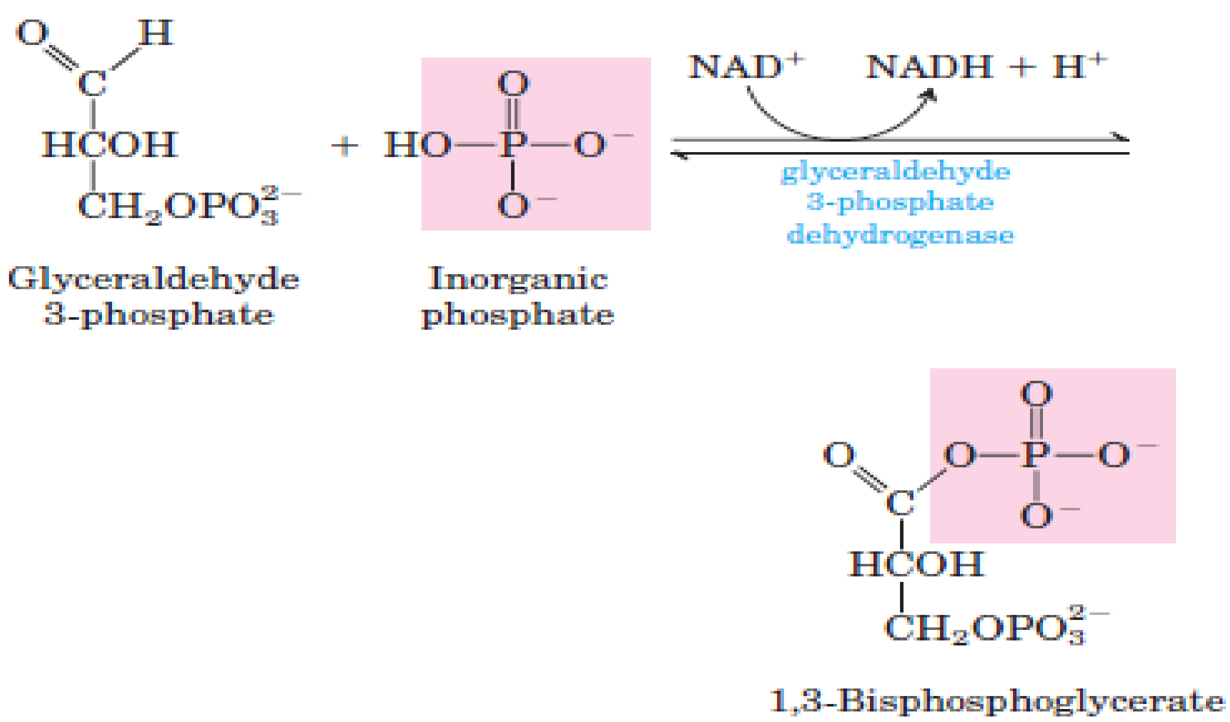
- The three-carbon stage **(payoff stage)** of glycolysis begins when the enzyme fructose 1,6-bisphosphate aldolase catalyzes the cleavage of fructose 1,6-bisphosphate into two halves, each with a phosphate group.
- One of the products, glyceraldehyde 3-phosphate, is converted directly to pyruvate in a five-step process.
- Because the other product, dihydroxyacetone phosphate, can be easily changed to glyceraldehyde 3-phosphate, both halves of fructose 1,6-bisphosphate are used in the three-carbon stage.



phosphates
Glyceraldehyde 3-phosphate
+
Dihydroxyacetone phosphate
5
(b)
Payoff phase
Oxidative conversion of glyceraldehyde 3-phosphate to pyruvate and the coupled formation of ATP and NADH
Glyceraldehyde 3-phosphate (2)
2P_i
2NAD+
2 NADH + H+
oxidation and phosphorylation
6
1,3-Bisphosphoglycerate (2)
first ATP-forming reaction (substrate-level phosphorylation)
7
2ADP
2 ATP
3-Phosphoglycerate (2)
8
2-Phosphoglycerate (2)
9
2H2O
Phosphoenolpyruvate (2)
second ATP-forming reaction (substrate-level phosphorylation)
10
2ADP
2 ATP
Pyruvate (2)
6 Glyceraldehyde 3-phosphate dehydrogenase
7 Phosphoglycerate kinase
8 Phosphoglycerate mutase
9 Enolase
10 Pyruvate kinase

...Payoff Phase

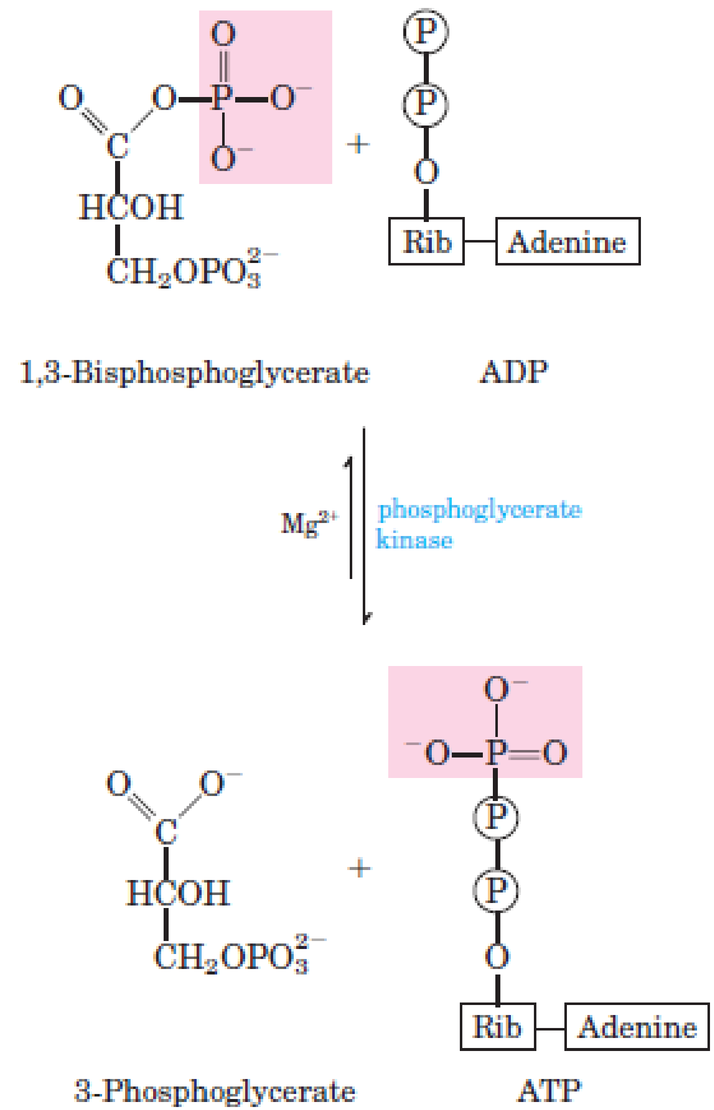
⑥ *Oxidation of Glyceraldehyde 3-Phosphate to 1,3-Bisphosphoglycerate* The first step in the payoff phase is the oxidation of glyceraldehyde 3-phosphate to **1,3-bisphosphoglycerate,** catalyzed by **glyceraldehyde 3-phosphate dehydrogenase:**



$\Delta G'^{\circ} = 6.3$ kJ/mol

...Payoff Phase

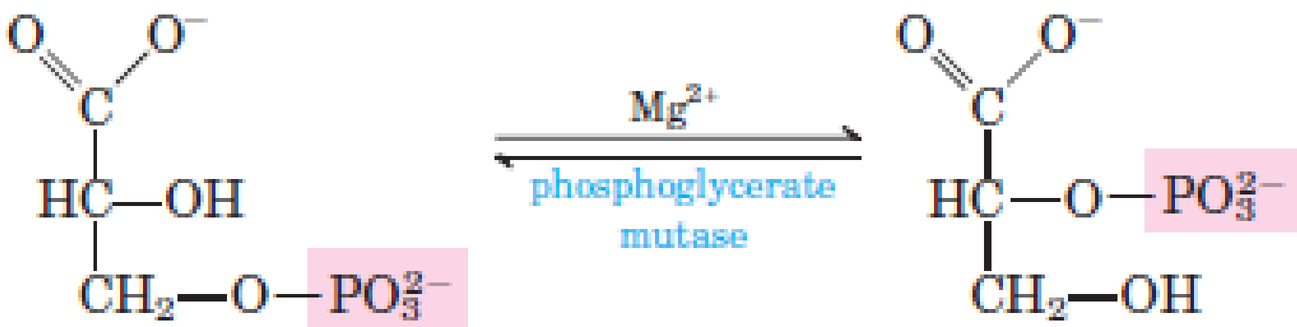
⑦ *Phosphoryl Transfer from 1,3-Bisphosphoglycerate to ADP* The enzyme **phosphoglycerate kinase** transfers the high-energy phosphoryl group from the carboxyl group of 1,3-bisphosphoglycerate to ADP, forming ATP and **3-phosphoglycerate:**



$\Delta G'^{\circ} = -18.5$ kJ/mol

...Payoff Phase

⑧ *Conversion of 3-Phosphoglycerate to 2-Phosphoglycerate*
The enzyme **phosphoglycerate mutase** catalyzes a reversible shift of the phosphoryl group between C-2 and C-3 of glycerate; Mg^{2+} is essential for this reaction:

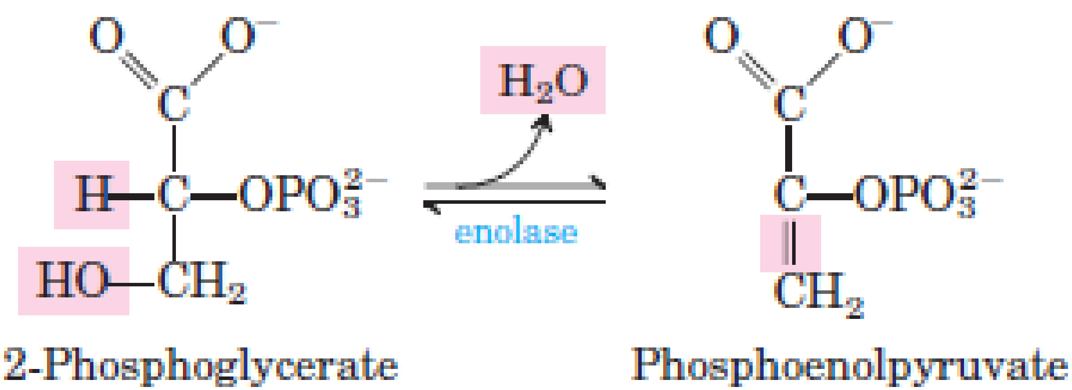


3-Phosphoglycerate 2-Phosphoglycerate

$\Delta G'^{\circ} = 4.4$ kJ/mol

...Payoff Phase

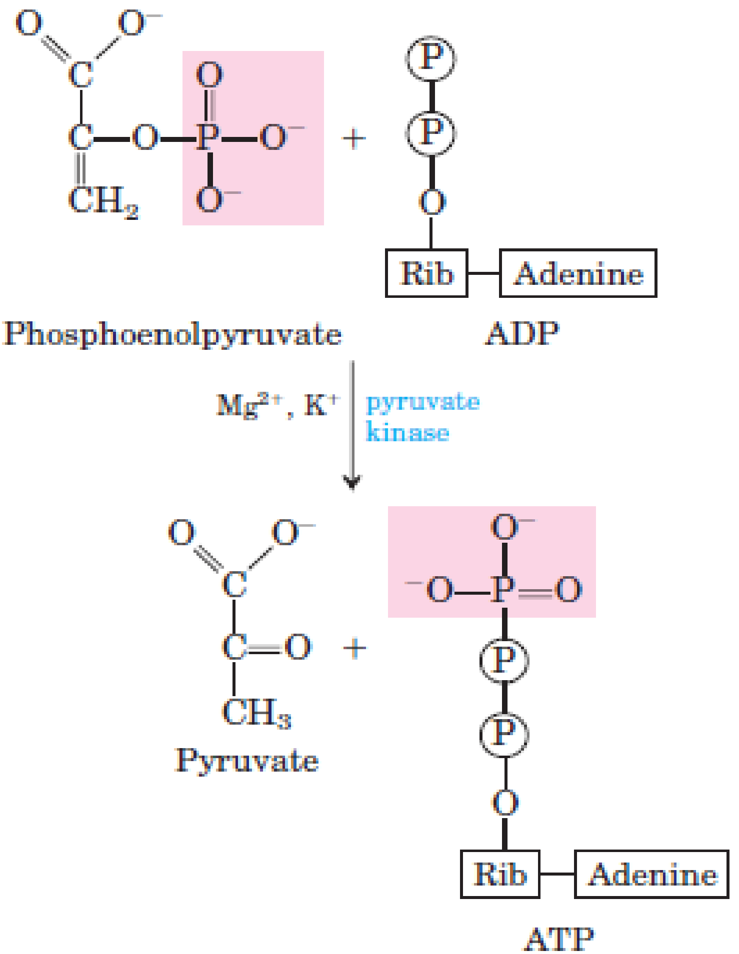
⑨ *Dehydration of 2-Phosphoglycerate to Phosphoenolpyruvate* In the second glycolytic reaction that generates a compound with high phosphoryl group transfer potential, **enolase** promotes reversible removal of a molecule of water from 2-phosphoglycerate to yield **phosphoenolpyruvate (PEP):**



$\Delta G'^{\circ} = 7.5$ kJ/mol

...Payoff Phase

(10) ***Transfer of the Phosphoryl Group from Phosphoenolpyruvate to ADP*** The last step in glycolysis is the transfer of the phosphoryl group from phosphoenolpyruvate to ADP, catalyzed by **pyruvate kinase,** which requires K^+ and either Mg^{2+} or Mn^{2+}:



$\Delta G'^{\circ} = -31.4$ kJ/mol

Substrate level phosphorylation

- Glyceraldehyde 3-phosphate is first oxidized with NAD as the electron acceptor, and a phosphate is simultaneously incorporated to give a high-energy molecule called 1,3-bisphosphoglycerate.
- The high energy phosphate on carbon one is subsequently donated to ADP to produce ATP.
- This synthesis of ATP is called substrate-level phosphorylation because ADP phosphorylation is coupled with the exergonic breakdown of a high-energy substrate molecule.
- A somewhat similar process generates a second ATP by substrate-level phosphorylation.
- The phosphate group on 3-phosphoglycerate shifts to carbon two, and 2-phosphoglycerate is dehydrated to form a second high-energy molecule, phosphoenolpyruvate.
- This molecule donates its phosphate to ADP forming a second ATP and pyruvate, the final product of the pathway.

Net Yield

- In the six-carbon stage two ATPs are used to form fructose 1,6-bisphosphate.
- For each glyceraldehyde 3-phosphate transformed into pyruvate, one NADH and two ATPs are formed.
- Because two glyceraldehyde 3-phosphates arise from a single glucose (one by way of dihydroxyacetone phosphate), the three-carbon stage generates four ATPs and two NADHs per glucose.
- Subtraction of the ATP used in the six-carbon stage from that produced in the three-carbon stage gives a net yield of two ATPs per glucose.
- Thus the catabolism of glucose to pyruvate in glycolysis can be represented by the following simple equation.

$$\text{Glucose} + 2\text{ADP} + 2\text{P}_i + 2\text{NAD}^+ \longrightarrow 2\text{ pyruvate} + 2\text{ATP} + 2\text{NADH} + 2\text{H}^+$$

Regulation

- The required adjustment in the rate of glycolysis is achieved by a complex interplay among ATP consumption, NADH regeneration, and allosteric regulation of several glycolytic enzymes—including hexokinase, PFK-1, and pyruvate kinase.
- Phosphofructokinase-1 is a regulatory enzyme, one of the most complex known.
- It is the major point of regulation in glycolysis.
- The activity of PFK-1 is increased whenever the cell's ATP supply is depleted or when the ATP breakdown products, ADP and AMP (particularly the latter), are in excess.
- The enzyme is inhibited whenever the cell has ample ATP and is well supplied by other fuels such as fatty acids.
- In some organisms, fructose 2,6-bisphosphate (not to be confused with the PFK-1 reaction product, fructose 1,6-bisphosphate) is a potent allosteric activator of PFK-1.

Questions

- Write an essay on EMP pathway or glycolysis.
- What are the regulatory steps of glycolysis?