

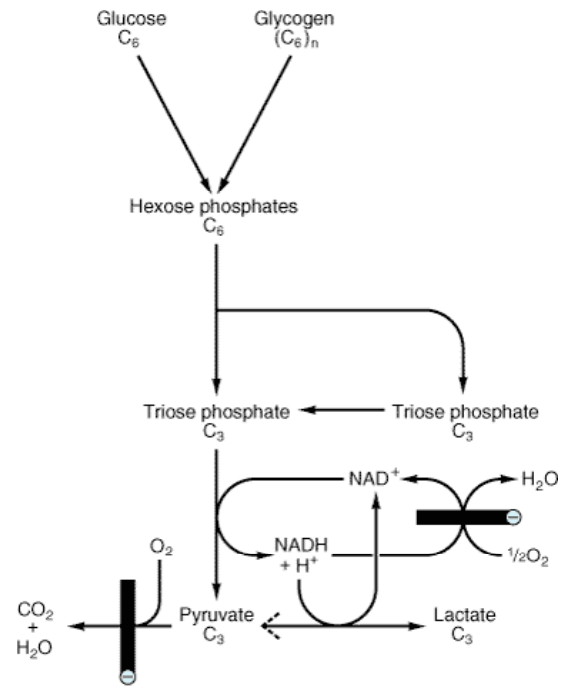
Glycolysis, Gluconeogenesis, and the Pentose Phosphate Pathway
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I. Important enzymes:

The “irreversible” enzymes of glycolysis are those that involve ATP. In gluconeogenesis, the reverse of glycolysis, these steps must be catalyzed by other enzymes (what are they called?) to.

1. Hexokinase:

- irreversible,
- **uses** 1 ATP per Glucose
- low K_m (high affinity for Glu)
- Hexokinase is in all cells, and its isozyme *Glucokinase* (aka Hexokinase IV) is found in the liver. Glucokinase has a higher K_m and primarily is active after meals (high Glu concentration in liver).
- allosteric inhibition by product (Glu-6-Ph)



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2. Phosphofructokinase- I:

- Irreversible
- Uses 1 ATP per Glu
- ***major rate regulator: at this point, the product has to continue in glycolysis (Glu and Glu-6-Ph can have other fates until this point, ie glycogen synthesis or pentose phosphate pathway)
- allosteric regulation: negative: ATP, citrate¹
 positive: AMP, ADP, fructose-2,6-bisphosphate

3. Pyruvate Kinase:

- Irreversible,
- Makes 2 ATP per Glu (1 per PEP): transfers phosphoryl group from PEP to ADP, producing ATP.
- Allosteric regulation: negative: (a) ATP, (b) acetyl-CoA, (c) fatty acids, (d) alanine²
 Positive: (a) Fructose-1,6-bisphosphate

4. Glyceraldehyde 3-phosphate dehydrogenase:

- This is REVERSIBLE, but plays a crucial role in glycolysis → this is the first enzyme that results in the formation of an energy-rich product!
- Oxidation by NAD⁺ (to NADH) and subsequent addition of phosphate (PO₄)₃-
- Mechanism: tetramer (look at structure), -SH group of active site is oxidized to a thiol ester, which is then *phosphorylated*; inorganic phosphate (Pi) is added, forming 1,3-bisphosphoglycerate, and the —SH group is reconstituted.
- NAD⁺ must be regenerated in order to allow glycolysis to continue. In anaerobic environments, this is achieved by reduction of pyruvate to lactate or ethanol (in yeast).

II. Mnemonic for remembering the glycolysis intermediates:

¹Produced in Citric Acid Cycle

² Product of transamination of pyruvate

Like I said in section, you should try to understand why each step has to occur, look at what is happening and how the names of the intermediates and enzymes reflect the structure and function. But in case you get stuck, here is a mnemonic:

Goodness (Glucose)
Gracious, (Glucose-6-P)
Father (Fructose-6-P)
Franklin (Fructose-1,6-diP)
Did (Dihydroxyacetone-P)
Go (Glyceraldehyde-P)
By (1,3-Biphosphoglycerate)
Picking (3-phosphoglycerate)
Pumpkins (2-phosphoglycerate)
(to)
PrEPare (Phosphoenolpyruvate [PEP])
Pies (Pyruvate)

III. Practice problems:

1. The conversion of 1 mol of fructose 1,6-bisphosphate to 2 mol of pyruvate by the glycolytic pathway results in a net formation of:
 - A) 1 mol of NAD^+ and 2 mol of ATP.
 - B) 1 mol of NADH and 1 mol of ATP.
 - C) 2 mol of NAD^+ and 4 mol of ATP.
 - D) 2 mol of NADH and 2 mol of ATP.
 - E) 2 mol of NADH and 4 mol of ATP.

2. Which of these cofactors participates directly in most of the oxidation-reduction reactions in the fermentation of glucose to lactate?
 - A) ADP
 - B) ATP
 - C) FAD/FADH_2
 - D) Glyceraldehyde 3-phosphate
 - E) NAD^+/NADH

3. The steps of glycolysis between glyceraldehyde 3-phosphate and 3-phosphoglycerate involve all of the following *except*:
 - A) ATP synthesis.
 - B) catalysis by phosphoglycerate kinase.
 - C) oxidation of NADH to NAD^+ .
 - D) the formation of 1,3-bisphosphoglycerate.
 - E) utilization of P_i .

5. Which of the following statements is *incorrect*?
 - A) Aerobically, oxidative decarboxylation of pyruvate forms acetate that enters the citric acid cycle.
 - B) In anaerobic muscle, pyruvate is converted to lactate.
 - C) In yeast growing anaerobically, pyruvate is converted to ethanol.
 - D) Reduction of pyruvate to lactate regenerates a cofactor essential for glycolysis.
 - E) Under anaerobic conditions pyruvate does not form because glycolysis does not occur.

5. The ultimate electron acceptor in the fermentation of glucose to ethanol is:
- A) acetaldehyde.
 - B) acetate.
 - C) ethanol.
 - D) NAD⁺.
 - E) pyruvate.
6. Which of the following compounds *cannot* serve as the starting material for the synthesis of glucose via gluconeogenesis?
- A) acetate
 - B) glycerol
 - C) lactate
 - D) oxaloacetate
 - E) α-ketoglutarate
7. An enzyme used in both glycolysis and gluconeogenesis is:
- A) 3-phosphoglycerate kinase.
 - B) glucose 6-phosphatase.
 - C) hexokinase.
 - D) phosphofructokinase-I.
 - E) pyruvate kinase.
8. Which one of the following statements about gluconeogenesis is *false*?
- A) For starting materials, it can use carbon skeletons derived from certain amino acids.
 - B) It consists entirely of the reactions of glycolysis, operating in the reverse direction.
 - C) It employs the enzyme glucose 6-phosphatase.
 - D) It is one of the ways that mammals maintain normal blood glucose levels between meals.
 - E) It requires metabolic energy (ATP or GTP).
9. All of the following enzymes involved in the flow of carbon from glucose to lactate (glycolysis) are also involved in the reversal of this flow (gluconeogenesis) *except*:
- A) 3-phosphoglycerate kinase.
 - B) aldolase.
 - C) enolase.
 - D) phosphofructokinase-I.
 - E) phosphoglucoisomerase.
10. The metabolic function of the pentose phosphate pathway is:
- A) act as a source of ADP biosynthesis.
 - B) generate NADPH and pentoses for the biosynthesis of fatty acids and nucleic acids.
 - C) participate in oxidation-reduction reactions during the formation of H₂O.
 - D) provide intermediates for the citric acid cycle.
 - E) synthesize phosphorus pentoxide.
11. The oxidation of 3 mol of glucose by the pentose phosphate pathway may result in the production of:
- A) 2 mol of pentose, 4 mol of NADPH, and 8 mol of CO₂.
 - B) 3 mol of pentose, 4 mol of NADPH, and 3 mol of CO₂.
 - C) 3 mol of pentose, 6 mol of NADPH, and 3 mol of CO₂.

- D) 4 mol of pentose, 3 mol of NADPH, and 3 mol of CO₂.
 E) 4 mol of pentose, 6 mol of NADPH, and 6 mol of CO₂.

12. Briefly describe the possible metabolic fates of pyruvate produced by glycolysis in humans, and explain the circumstances that favor each.

Ans: Under aerobic conditions, pyruvate is oxidized to acetyl-coA and passes through the citric acid cycle. Under anaerobic conditions, pyruvate is reduced to lactate to recycle NADH to NAD⁺, allowing the continuation of glycolysis.

13. In the conversion of glucose to pyruvate via glycolysis, all of the following enzymes participate. Indicate the order in which they function by numbering them.

- 1 hexokinase
4 triose phosphate isomerase
2 phosphohexose isomerase
6 enolase
5 glyceraldehyde 3-phosphate dehydrogenase
7 pyruvate kinase
3 phosphofructokinase-I

Which of the enzymes represents a major regulation point in glycolysis? ANS: Phosphofructokinase

Which catalyzes a reaction in which ATP is produced? Ans: pyruvate kinase

Which catalyzes a reaction in which NADH is produced? Ans: glyceraldehyde-3-phosphate dehydrogenase

14. The conversion of glyceraldehyde 3-phosphate to dihydroxyacetone phosphate is catalyzed by triose phosphate isomerase. The standard free-energy change (ΔG°) for this reaction is -7.5 kJ/mol. Draw the two structures. Define the equilibrium constant for the reaction and calculate it using only the data given here. Be sure to show your work. ($R = 8.315$ J/mol·K; $T = 298$ K)

Ans: See Fig. 15-4, p. 535.

$$K_{eq}' = \frac{[\text{glyceraldehyde 3-phosphate}]}{[\text{dihydroxyacetone phosphate}]}$$

$$\Delta G^\circ = -RT \ln K_{eq}'$$

$$\ln K_{eq}' = \frac{-\Delta G^\circ}{RT}$$

$$\ln K_{eq}' = \frac{-(-7,500 \text{ J/mol})}{(8.315 \text{ J/mol}\cdot\text{K})(298 \text{ K})} = 3.027$$

$$K_{eq}' = 20.6$$

(See also Chapter 14.)

IV. Following the Fate of Carbons:

2. If glucose labeled with ¹⁴C in C-1 were fed to yeast carrying out the ethanol fermentation, where would the ¹⁴C label be in the products? Ans: In C-2 (methyl group) of ethanol only

3. In an anaerobic muscle preparation, lactate formed from glucose labeled in C-3 and C-4 would be labeled in: Ans: only the carboxyl carbon atom.
4. Glucose labeled with ^{14}C in C-1 and C-6 gives rise in glycolysis to pyruvate labeled in: its methyl carbon.
5. In an anaerobic muscle preparation, lactate formed from glucose labeled in C-2 would be labeled in: only the carbon atom carrying the OH

Study Questions:

1. What does the process of glycolysis accomplish in terms of ATP manufacture and end product formation?
2. Where does glycolysis occur (in which types of cells, and where in the cell?) and what are the destinations in the cell for its end products?
3. Is hexokinase reversible or irreversible? Does it have a relatively high or low K_m ? What is the function of its isozyme, glucokinase?