BIOCHEMISTRY: LS2101

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Syllabus

Introductory biochemistry: biological interactions.

Amino acids

•Protein structure and folding, Enzymology, Enzyme kinetics, and allostery. vitamins and coenzymes.

•Overview of techniques in protein purification.

•Nucleic acid structure.

•Introduction to intermediary metabolism: Glycolysis, TCA cycle, Electron transport

Glycolysis, TCA cycle, Electron transport

Glycolysis: Greek glykys, meaning “sweet,” and lysis, meaning “splitting”: Glucose to Pyruvate by Enzyma

CARBOHYDRATES: Most abundant biomolecules on earth Empirical Formula : $(CH_2O)_n$

Monosaccharides

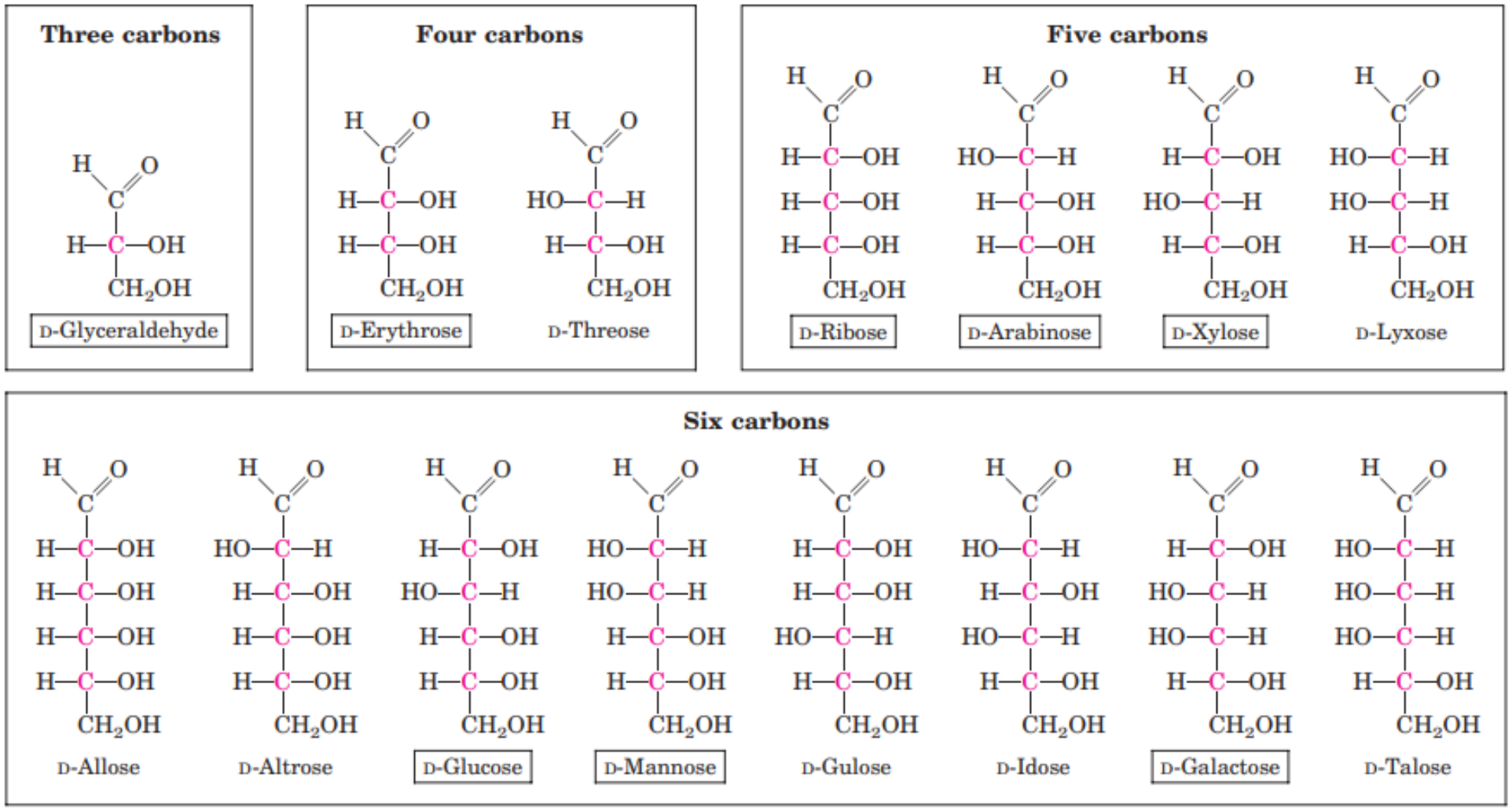
Glucose, Fructose,
Ribose

Oligosaccharides

Disaccharides
Cane Sugar

Polysaccharides
Starch

The oxidation of carbohydrates is the central energy-yielding pathway in most non-photosynthetic cells.


Three carbons
D-Glyceraldehyde
Four carbons
D-Erythrose
D-Threose
Five carbons
D-Ribose
D-Arabinose
D-Xylose
D-Lyxose
Six carbons
D-Allose
D-Altrose
D-Glucose
D-Mannose
D-Gulose
D-Idose
D-Galactose
D-Talose

D-Aldoses
(a)

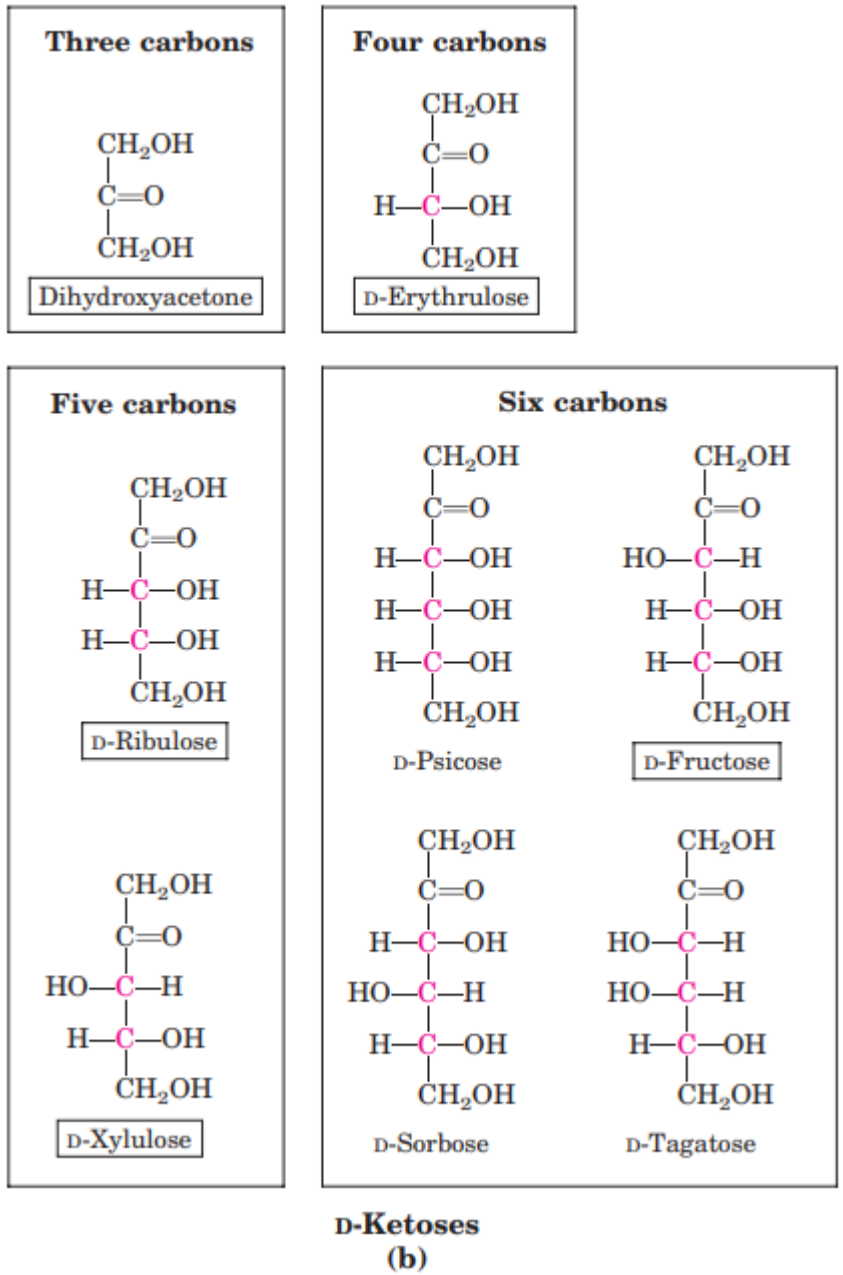


FIGURE 7–3 Aldoses and ketoses. The series of **(a)** D-aldoses and **(b)** D-ketoses having from three to six carbon atoms, shown as projection formulas. The carbon atoms in red are chiral centers. In all these D isomers, the chiral carbon *most distant from the carbonyl carbon* has the same configuration as the chiral carbon in D-glyceraldehyde. The sugars named in boxes are the most common in nature; you will encounter these again in this and later chapters.

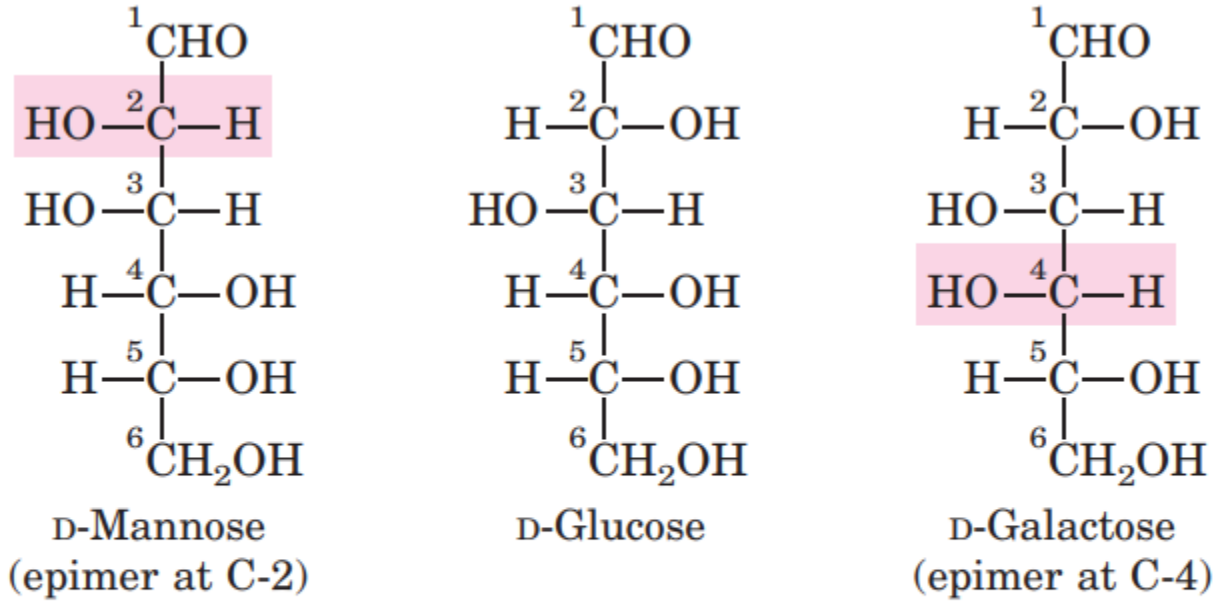
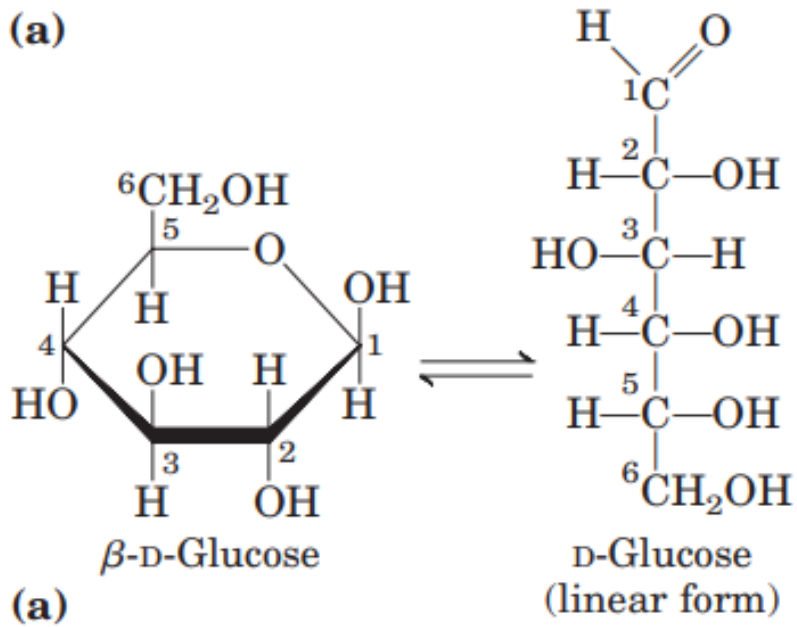


FIGURE 7–4 Epimers. D-Glucose and two of its epimers are shown as projection formulas. Each epimer differs from D-glucose in the configuration at one chiral center (shaded red).


(a)
$^6CH_2OH$
5
O
H
H
OH
4
1
OH
H
HO
H
3
2
H
OH
β-D-Glucose
(a)
H
O
1C
2
H—C—OH
HO—C—H
3
4
H—C—OH
5
H—C—OH
6CH2OH
D-Glucose
(linear form)

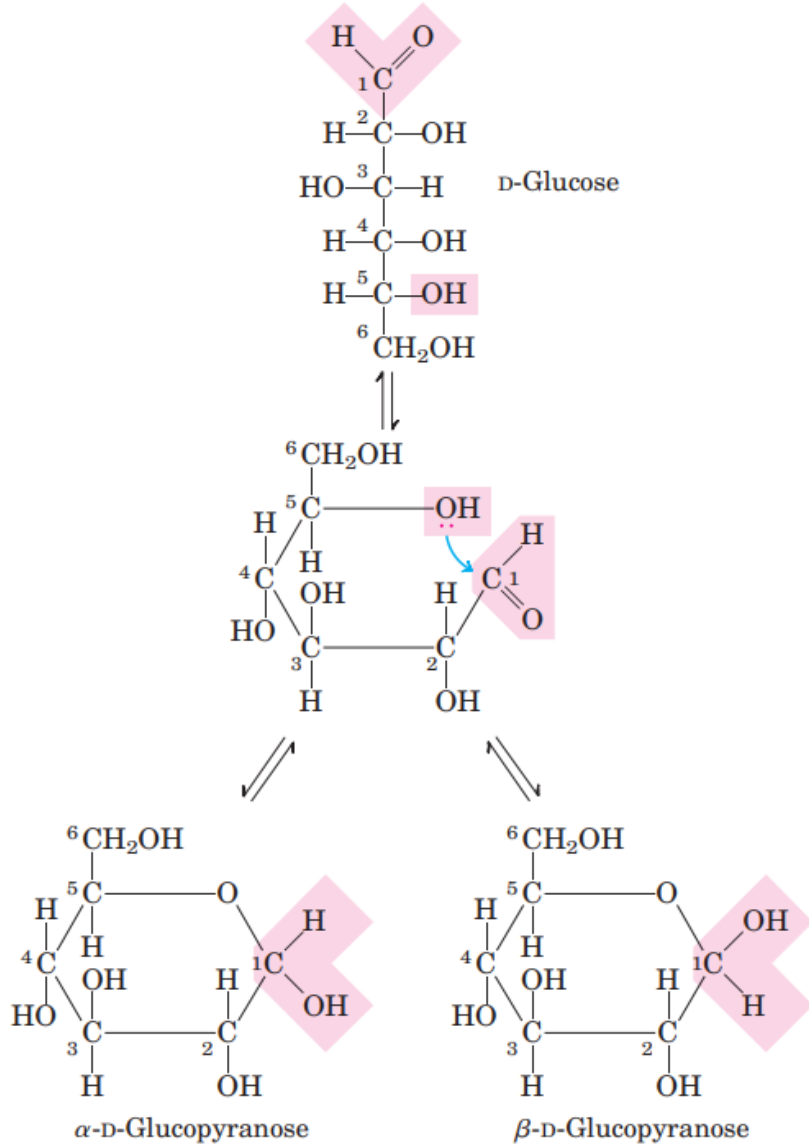


FIGURE 7–6 Formation of the two cyclic forms of D-glucose. Reaction between the aldehyde group at C-1 and the hydroxyl group at C-5 forms a hemiacetal linkage, producing either of two stereoisomers, the α and β anomers, which differ only in the stereochemistry around the hemiacetal carbon. The interconversion of α and β anomers is called mutarotation.

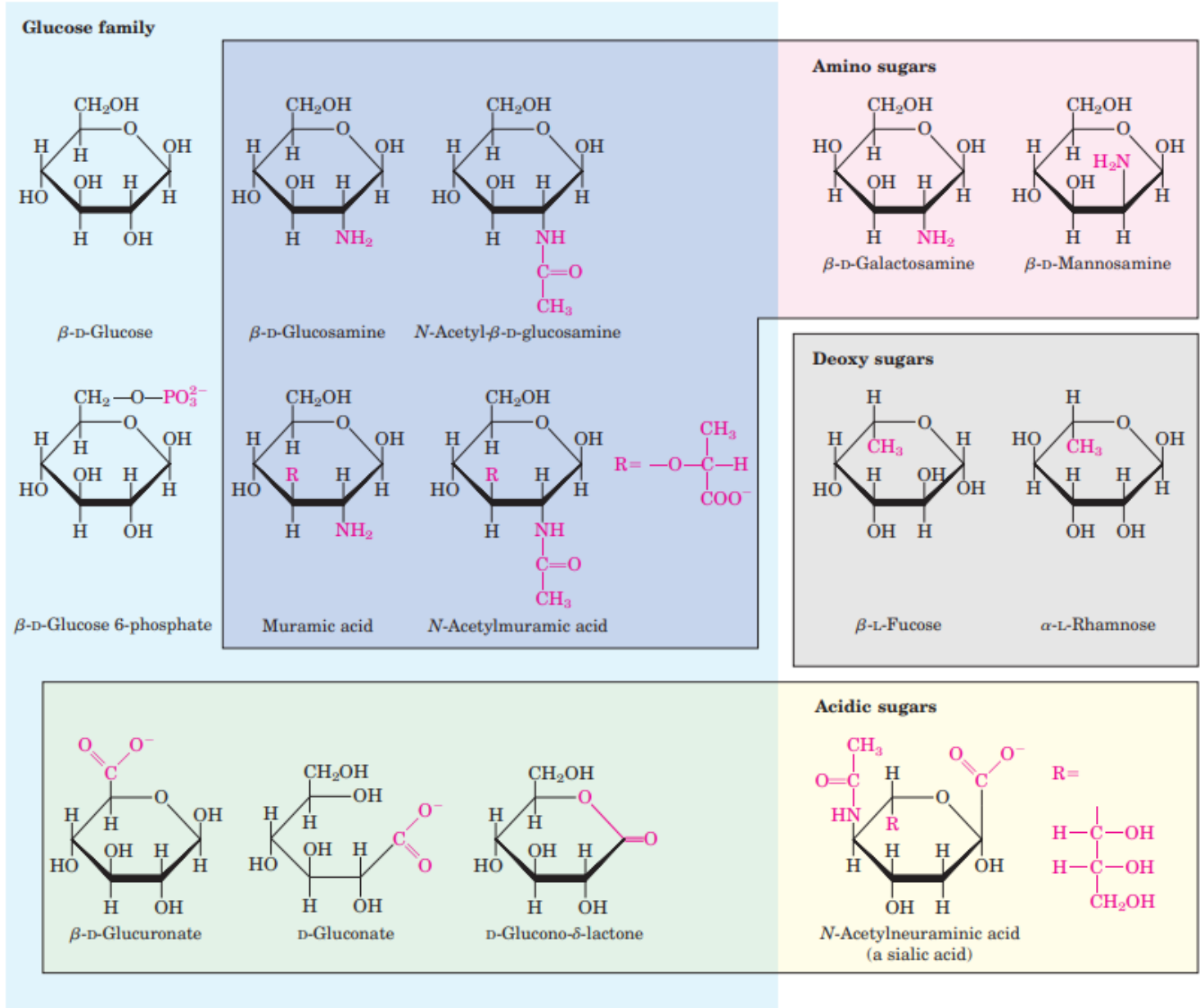


FIGURE 7–9 Some hexose derivatives important in biology. In amino sugars, an —NH_2 group replaces one of the —OH groups in the parent hexose. Substitution of —H for —OH produces a deoxy sugar; note that the deoxy sugars shown here occur in nature as the L isomers. The acidic sugars contain a carboxylate group, which confers a negative charge at neutral pH. D-Glucono-δ-lactone results from formation of an ester linkage between the C-1 carboxylate group and the C-5 (also known as the δ carbon) hydroxyl group of D-gluconate.

glycosidic bonds

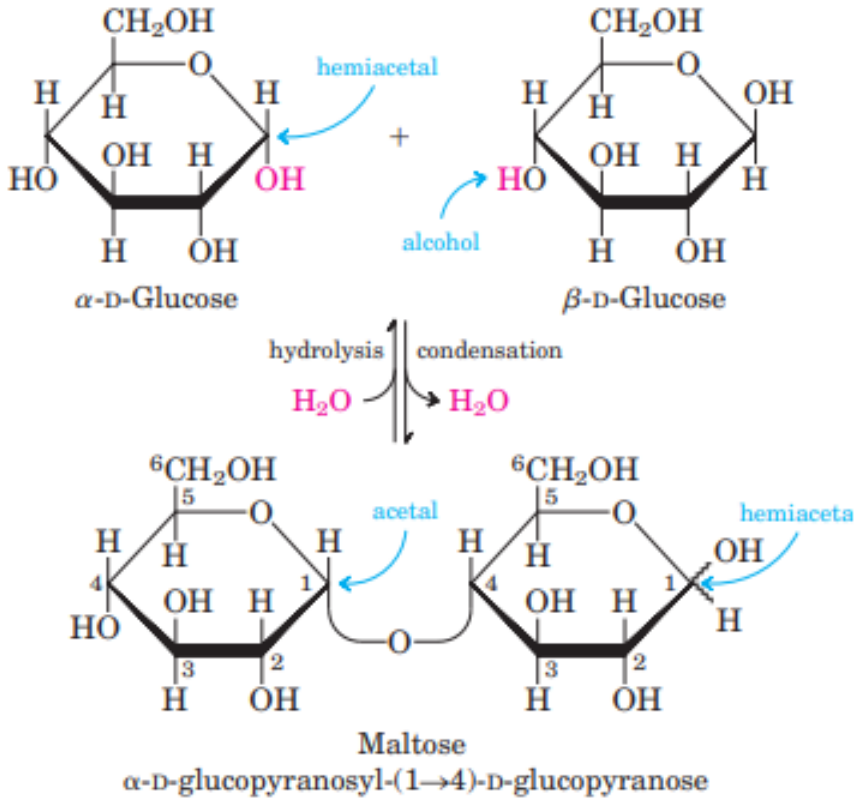
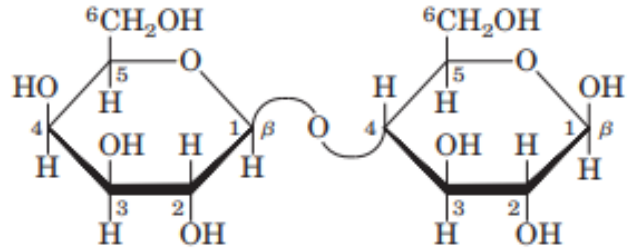
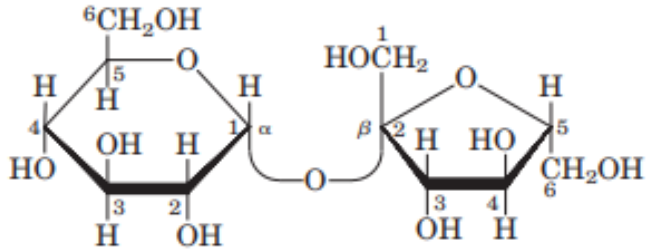


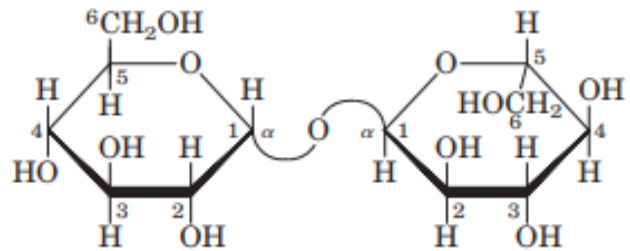
FIGURE 7–11 Formation of maltose. A disaccharide is formed from two monosaccharides (here, two molecules of D-glucose) when an —OH (alcohol) of one glucose molecule (right) condenses with the intramolecular hemiacetal of the other glucose molecule (left), with elimination of H_2O and formation of an *O*-glycosidic bond. The reversal of this reaction is hydrolysis—attack by H_2O on the glycosidic bond. The maltose molecule retains a reducing hemiacetal at the C-1 not involved in the glycosidic bond. Because mutarotation interconverts the α and β forms of the hemiacetal, the bonds at this position are sometimes depicted with wavy lines, as shown here, to indicate that the structure may be either α or β.



Lactose (β form)
β-D-galactopyranosyl-(1→4)-β-D-glucopyranose
Gal(β1→4)Glc



Sucrose
α-D-glucopyranosyl β-D-fructofuranoside
Glc(α1↔2β)Fru



Trehalose
α-D-glucopyranosyl α-D-glucopyranoside
Glc(α1↔1α)Glc

FIGURE 7–12 Some common disaccharides. Like maltose in Figure 7–11, these are shown as Haworth perspectives. The common name, full systematic name, and abbreviation are given for each disaccharide.

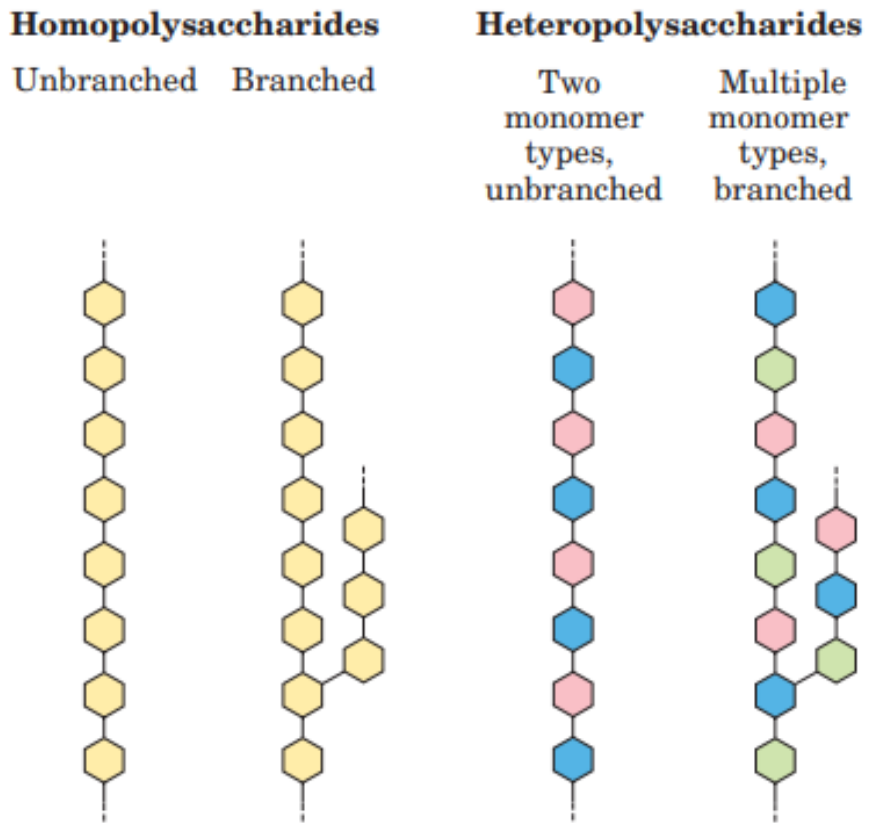


FIGURE 7–13 Homo- and heteropolysaccharides. Polysaccharides may be composed of one, two, or several different monosaccharides, in straight or branched chains of varying length.

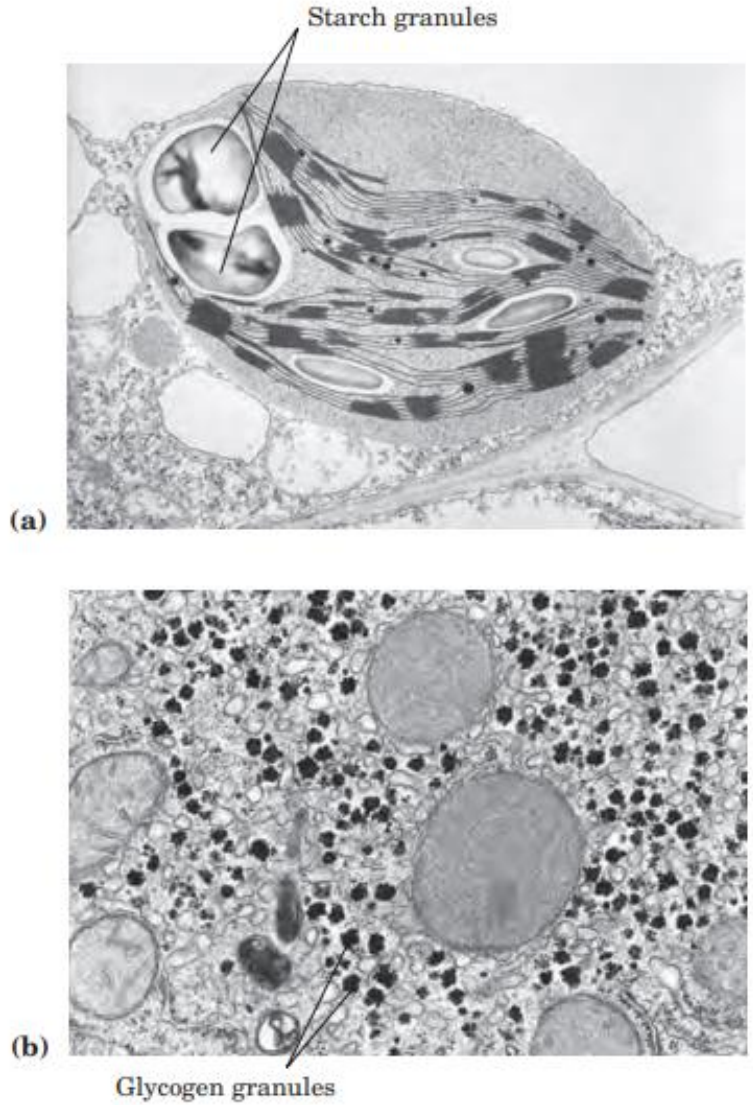


FIGURE 7–14 Electron micrographs of starch and glycogen granules. **(a)** Large starch granules in a single chloroplast. Starch is made in the chloroplast from D-glucose formed photosynthetically. **(b)** Glycogen granules in a hepatocyte. These granules form in the cytosol and are much smaller (~0.1 μm) than starch granules (~1.0 μm).

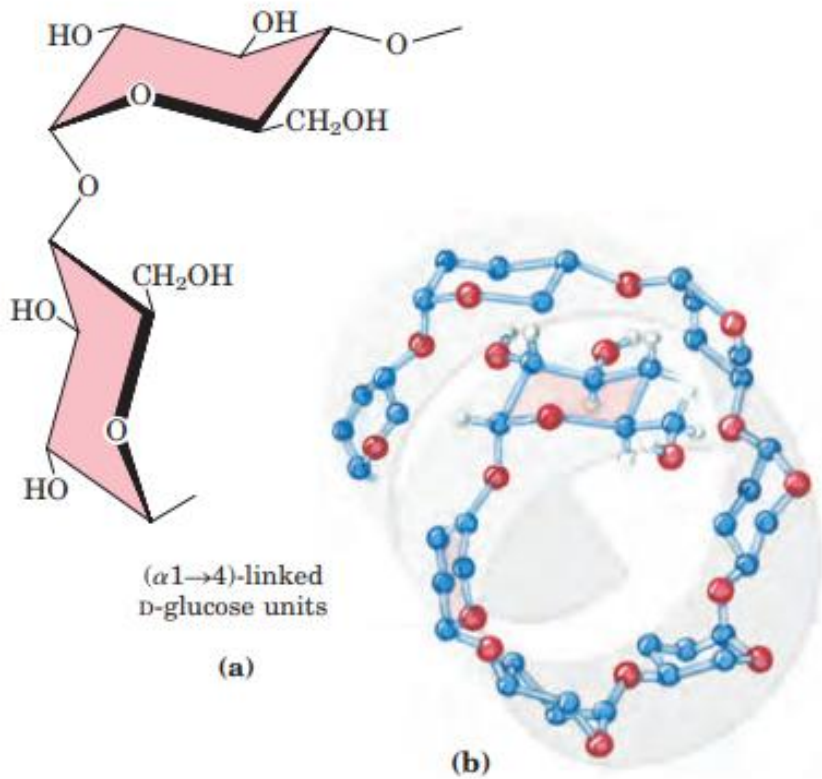


FIGURE 7–21 The structure of starch (amylose). (a) In the most stable conformation, with adjacent rigid chairs, the polysaccharide chain is curved, rather than linear as in cellulose (see Fig. 7–16). **(b)** Scale drawing of a segment of amylose. The conformation of (α1→4) linkages in amylose, amylopectin, and glycogen causes these polymers to assume tightly coiled helical structures. These compact structures produce the dense granules of stored starch or glycogen seen in many cells (see Fig. 7–14).

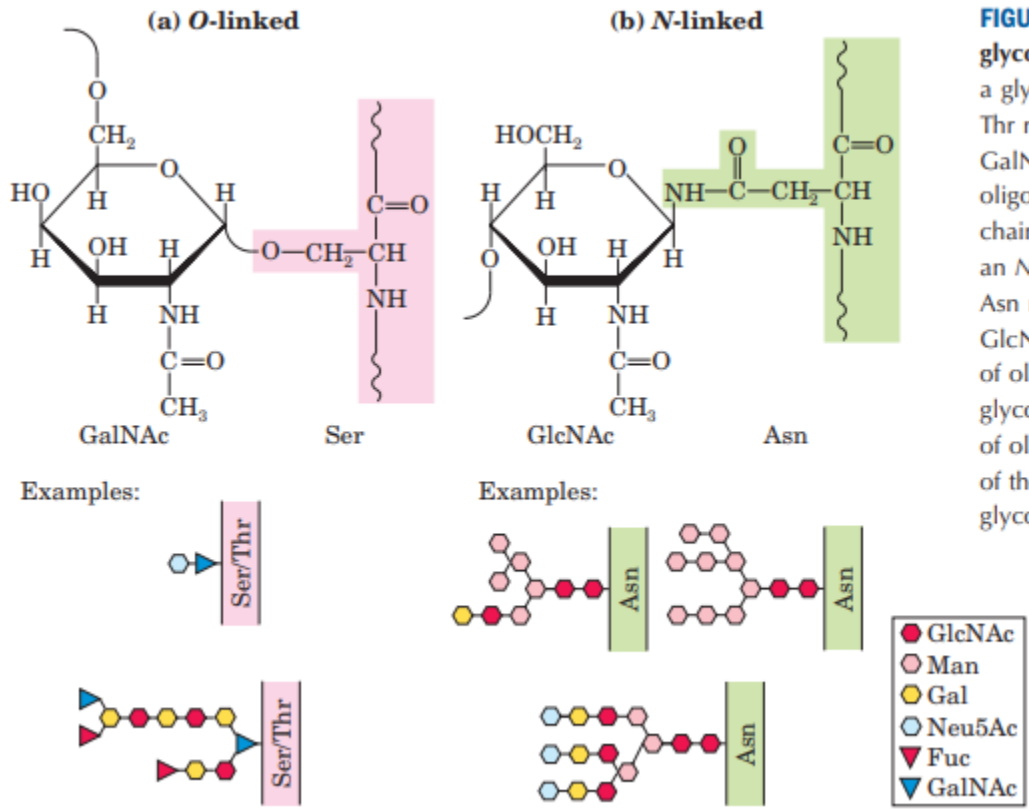


FIGURE 7–31 Oligosaccharide linkages in glycoproteins. **(a)** *O*-linked oligosaccharides have a glycosidic bond to the hydroxyl group of Ser or Thr residues (shaded pink), illustrated here with GalNAc as the sugar at the reducing end of the oligosaccharide. One simple chain and one complex chain are shown. **(b)** *N*-linked oligosaccharides have an *N*-glycosyl bond to the amide nitrogen of an Asn residue (shaded green), illustrated here with GlcNAc as the terminal sugar. Three common types of oligosaccharide chains that are *N*-linked in glycoproteins are shown. A complete description of oligosaccharide structure requires specification of the position and stereochemistry (α or β) of each glycosidic linkage.

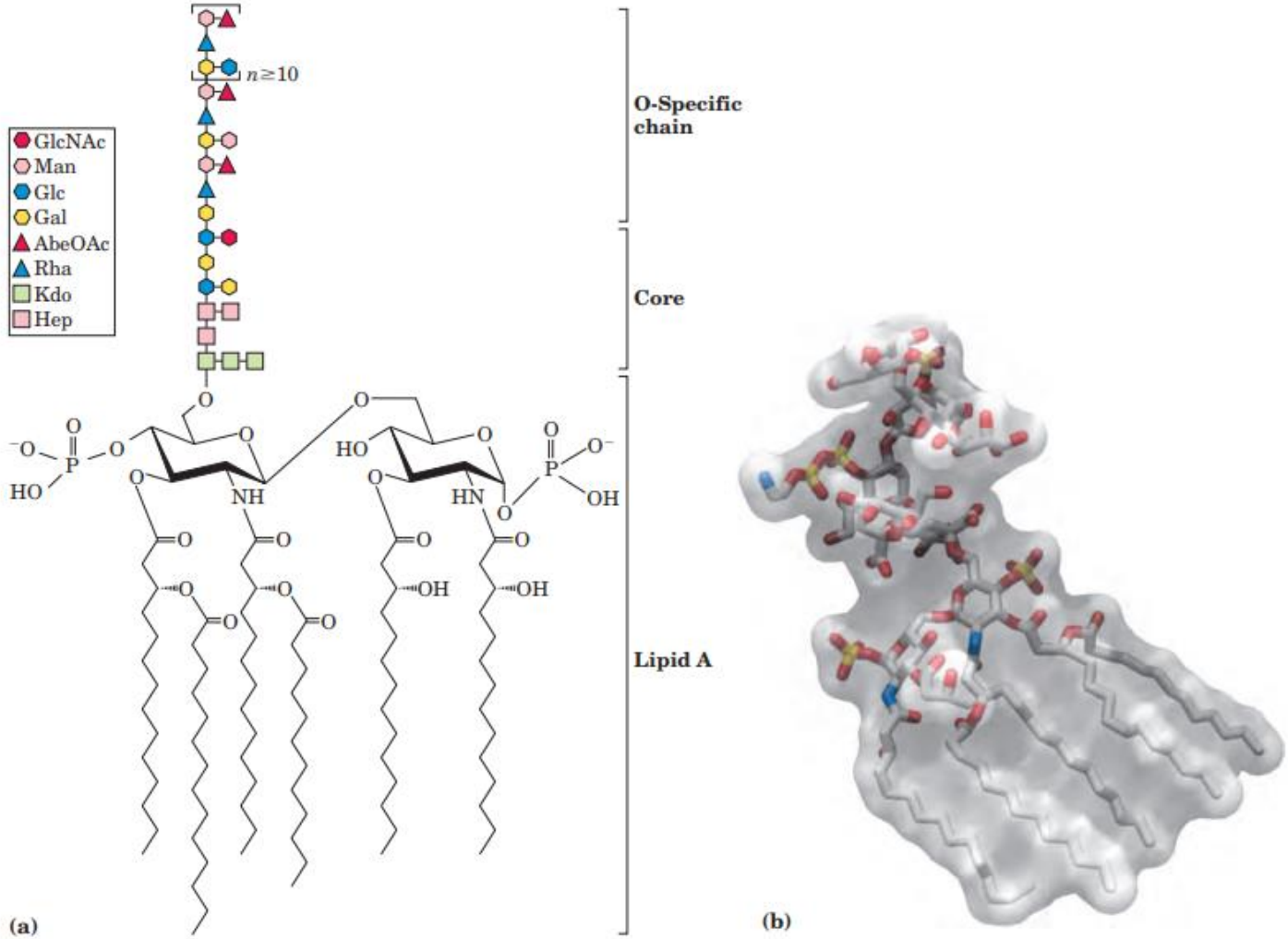


FIGURE 7–32 Bacterial lipopolysaccharides. (a) Schematic diagram of the lipopolysaccharide of the outer membrane of *Salmonella typhimurium*. Kdo is 3-deoxy-D-manno-octulosonic acid, previously called *keto*deoxy*o*ctonic acid; Hep is L-glycero-D-mannoheptose; AbeOAc is abequose (a 3,6-dideoxyhexose) acetylated on one of its hydroxyls. There are six fatty acids in the lipid A portion of the molecule. Different bacterial species have subtly different lipopolysaccharide structures, but they have in common a lipid region (lipid A), a core oligosaccharide, and an "O-specific" chain, which is the principal determinant of the serotype (immunological reactivity) of the bacterium. The outer membranes of the gram-negative bacteria *S. typhimurium* and *E. coli* contain so many lipopolysaccharide molecules that the cell surface is virtually covered with O-specific chains. **(b)** The stick structure of the lipopolysaccharide of *E. coli* is visible through a transparent surface contour model of the molecule. The position of the sixth fatty acyl chain was not defined in the crystallographic study, so it is not shown.

Glycolysis:

In glycolysis (from the Greek glykys, meaning “sweet,” and lysis, meaning “splitting”), a molecule of glucose is degraded in a series of enzyme-catalyzed reactions to yield two molecules of the three-carbon compound pyruvate.

During the sequential reactions of glycolysis, some of the free energy released from glucose is conserved in the form of ATP and NADH.

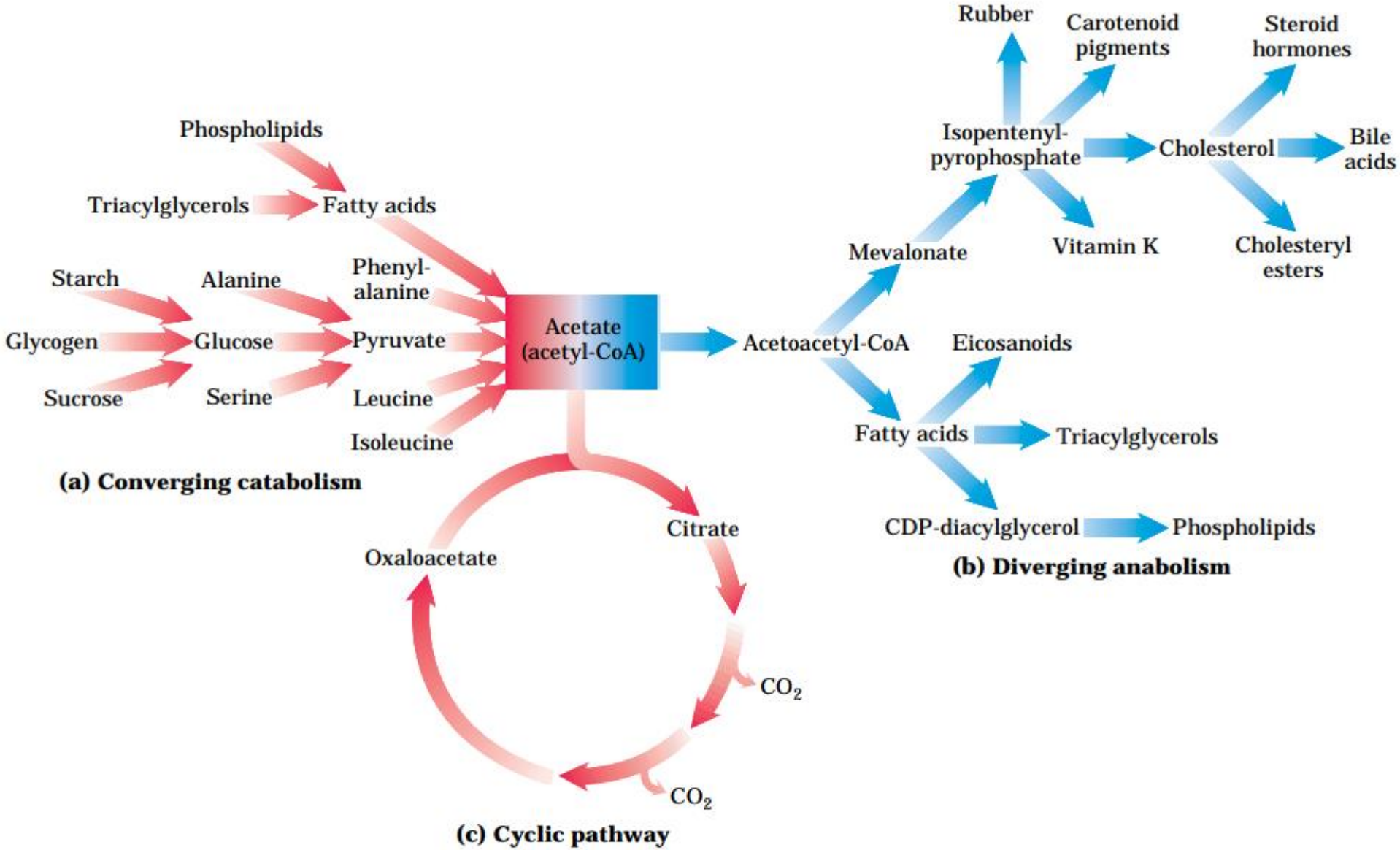


FIGURE 4 Three types of nonlinear metabolic pathways. **(a)** Converging, catabolic; **(b)** diverging, anabolic; and **(c)** cyclic, in which one of the starting materials (oxaloacetate in this case) is regenerated and reenters the pathway. Acetate, a key metabolic intermediate, is the breakdown product of a variety of fuels **(a)**, serves as the precursor for an array of products **(b)**, and is consumed in the catabolic pathway known as the citric acid cycle **(c)**.

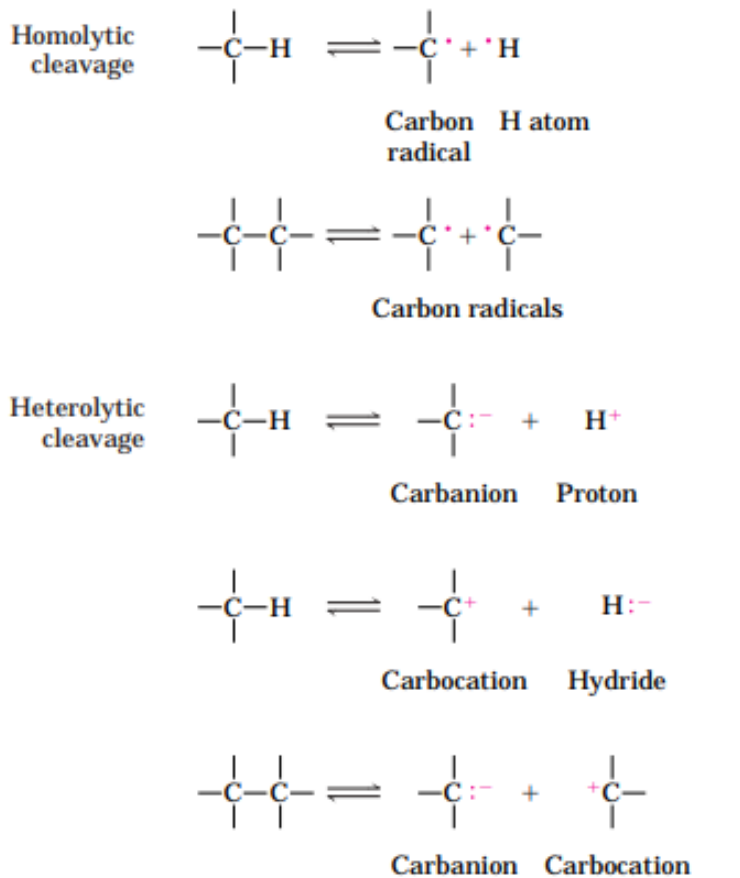


FIGURE 5 **Two mechanisms for cleavage of a C—C or C—H bond.** In homolytic cleavages, each atom keeps one of the bonding electrons, resulting in the formation of carbon radicals (carbons having unpaired electrons) or uncharged hydrogen atoms. In heterolytic cleavages, one of the atoms retains both bonding electrons. This can result in the formation of carbanions, carbocations, protons, or hydride ions.

Most of the reactions in living cells fall into one of **five general categories:**

(1) Oxidation-reductions;

(2) **Reactions that make or break carbon–carbon bonds**;

(3) **Internal rearrangements, isomerizations, and eliminations**;

(4) **Group transfers**; and

(5) **Free radical reactions**.

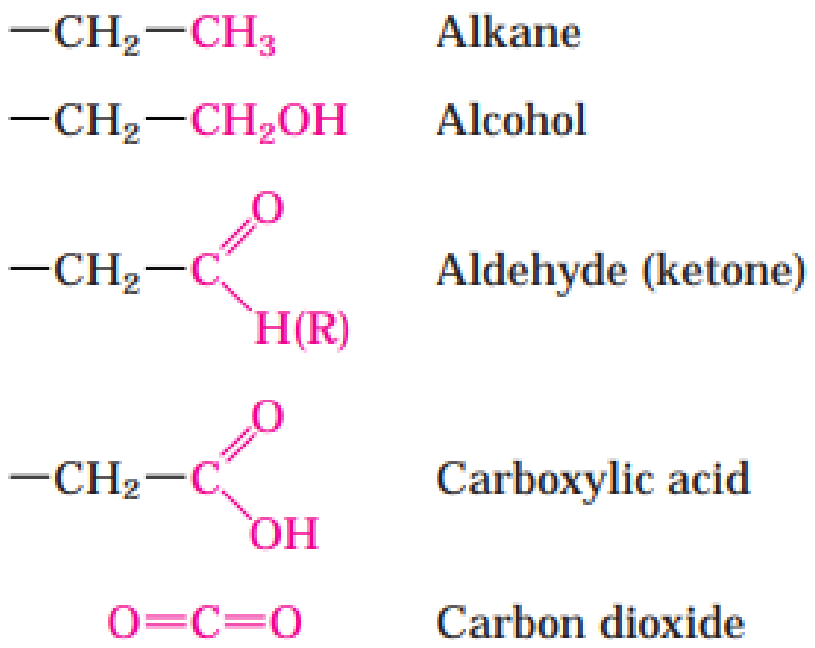


FIGURE 6 **The oxidation states of carbon in biomolecules.** Each compound is formed by oxidation of the red carbon in the compound listed above it. Carbon dioxide is the most highly oxidized form of carbon found in living systems.

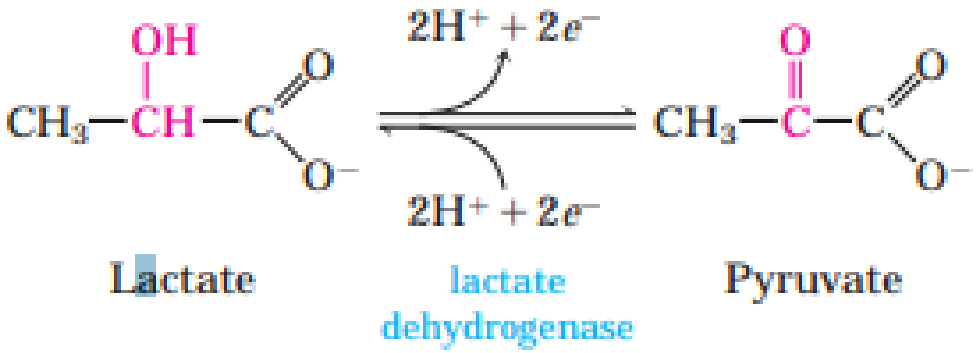


FIGURE 7 An oxidation-reduction reaction. Shown here is the oxidation of lactate to pyruvate. In this dehydrogenation, two electrons and two hydrogen ions (the equivalent of two hydrogen atoms) are removed from C-2 of lactate, an alcohol, to form pyruvate, a ketone. In cells the reaction is catalyzed by lactate dehydrogenase and the electrons are transferred to a cofactor called nicotinamide adenine dinucleotide. This reaction is fully reversible; pyruvate can be reduced by electrons from the cofactor. In Chapter 13 we discuss the factors that determine the direction of a reaction.

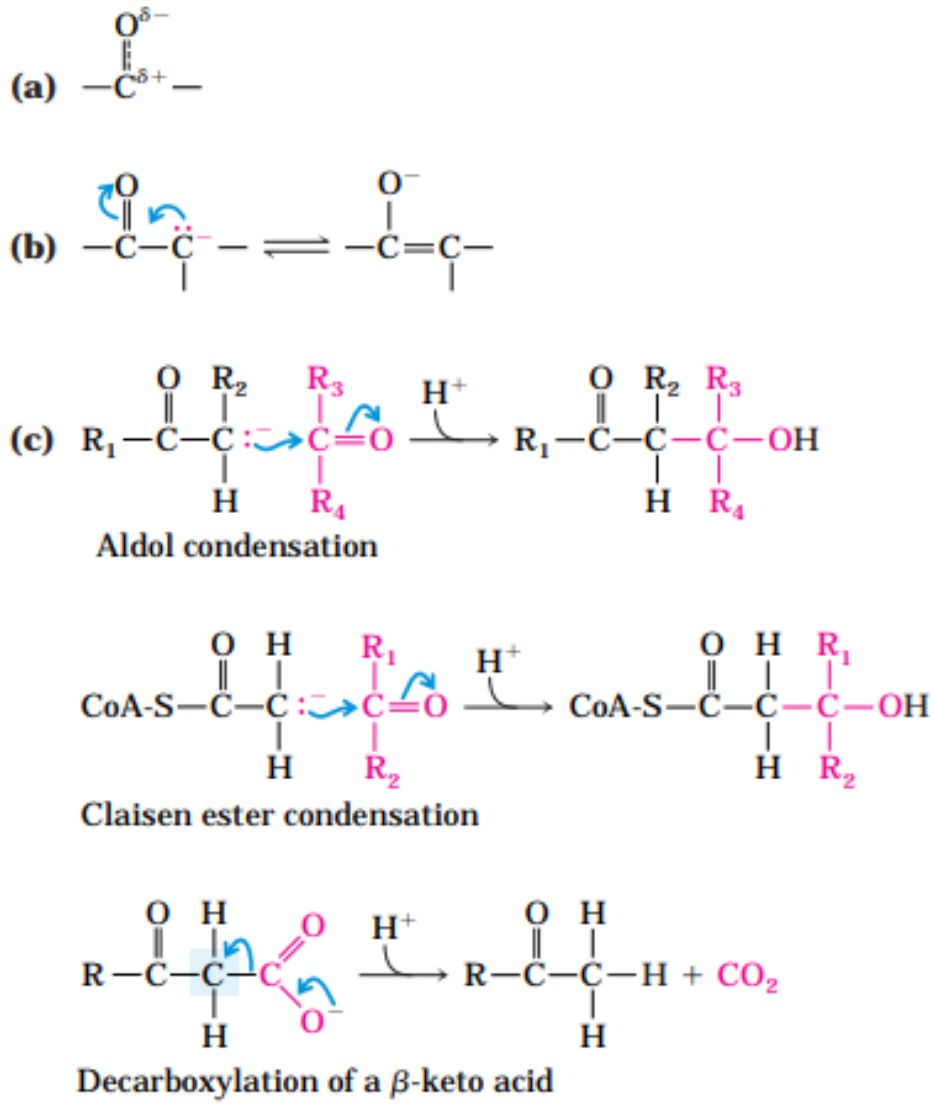


FIGURE 8 **Carbon–carbon bond formation reactions. (a)** The carbon atom of a carbonyl group is an electrophile by virtue of the electron-withdrawing capacity of the electronegative oxygen atom, which results in a resonance hybrid structure in which the carbon has a partial positive charge. **(b)** Within a molecule, delocalization of electrons into a carbonyl group facilitates the transient formation of a carbanion on an adjacent carbon. **(c)** Some of the major reactions involved in the formation and breakage of C—C bonds in biological systems. For both the aldol condensation and the Claisen condensation, a carbanion serves as nucleophile and the carbon of a carbonyl group serves as electrophile. The carbanion is stabilized in each case by another carbonyl at the carbon adjoining the carbanion carbon. In the decarboxylation reaction, a carbanion is formed on the carbon shaded blue as the CO_2 leaves. The reaction would not occur at an appreciable rate but for the stabilizing effect of the carbonyl adjacent to the carbanion carbon. Wherever a carbanion is shown, a stabilizing resonance with the adjacent carbonyl, as shown in **(a)**, is assumed. The formation of the carbanion is highly disfavored unless the stabilizing carbonyl group, or a group of similar function such as an imine, is present.

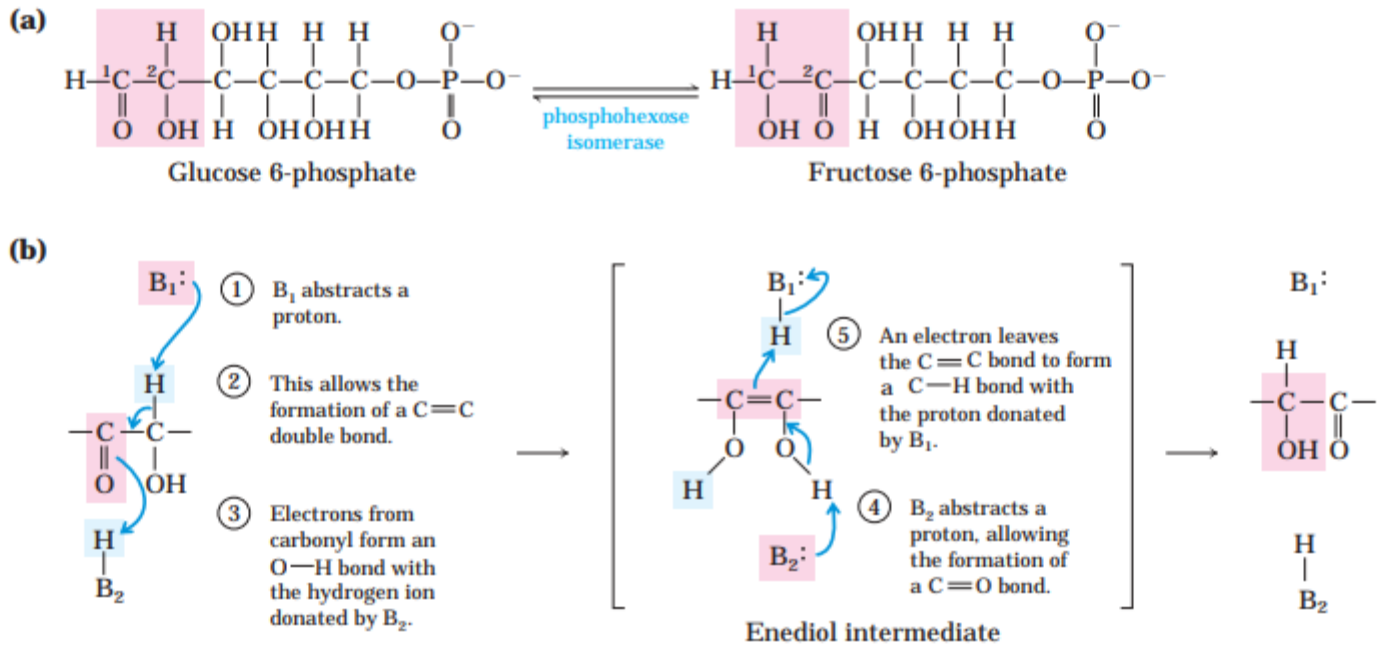


FIGURE 9 **Isomerization and elimination reactions. (a)** The conversion of glucose 6-phosphate to fructose 6-phosphate, a reaction of sugar metabolism catalyzed by phosphohexose isomerase. **(b)** This reaction proceeds through an enediol intermediate. The curved blue arrows represent the movement of bonding electrons from nucleophile (pink) to electrophile (blue). B_1 and B_2 are basic groups on the enzyme; they are capable of donating and accepting hydrogen ions (protons) as the reaction progresses.

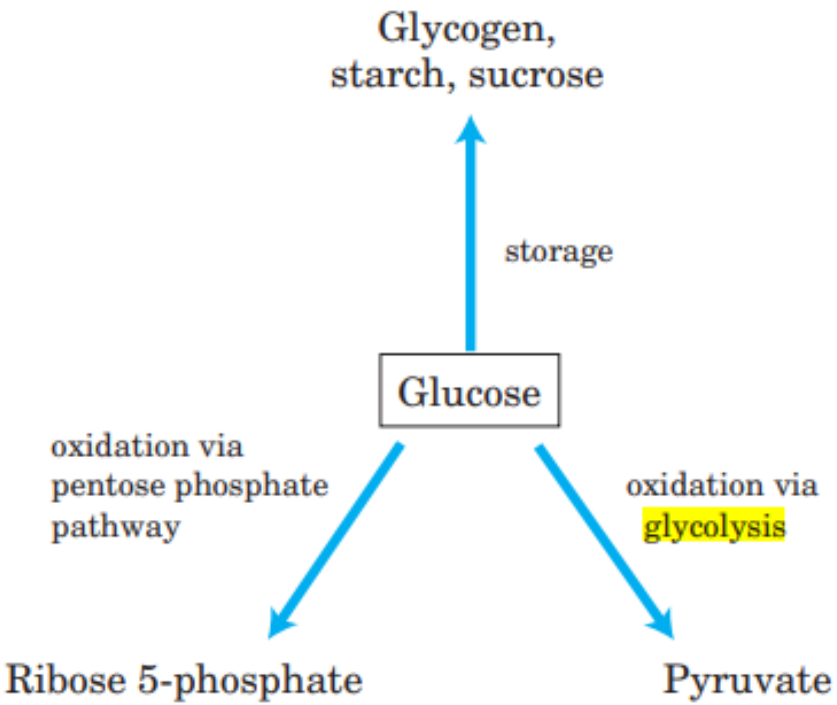


FIGURE 14–1 Major pathways of glucose utilization. Although not the only possible fates for glucose, these three pathways are the most significant in terms of the amount of glucose that flows through them in most cells.

Before Glycolysis:

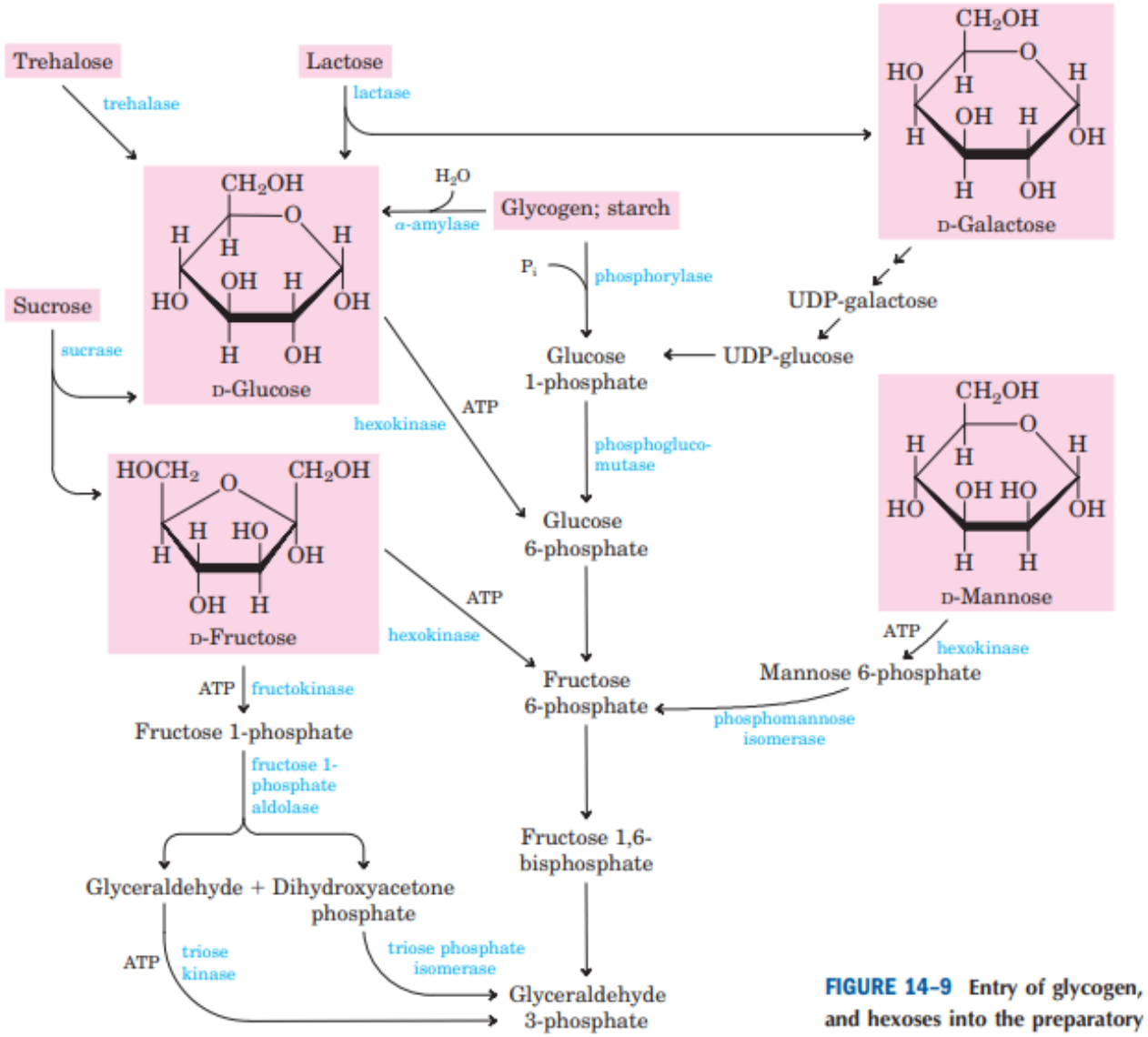
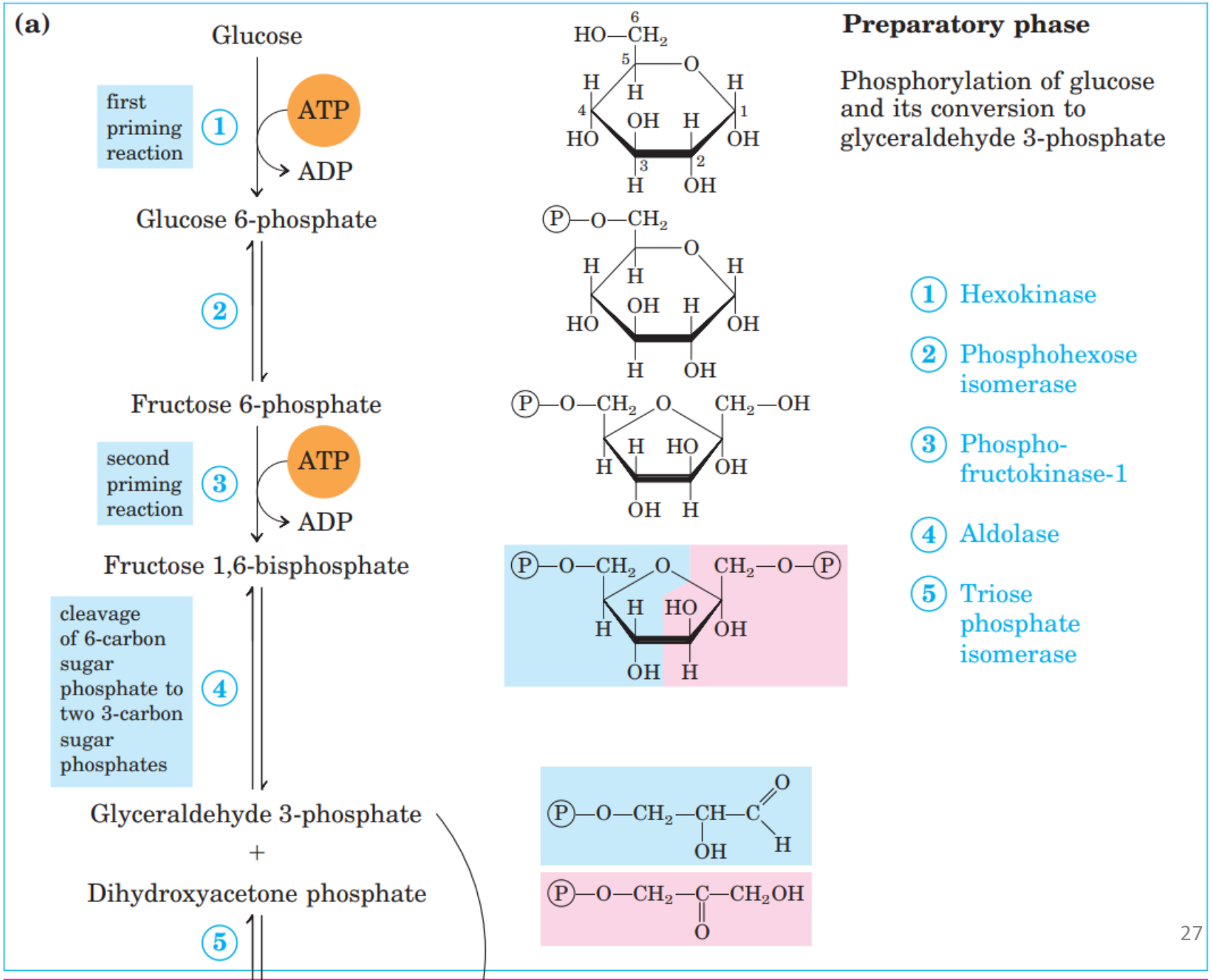
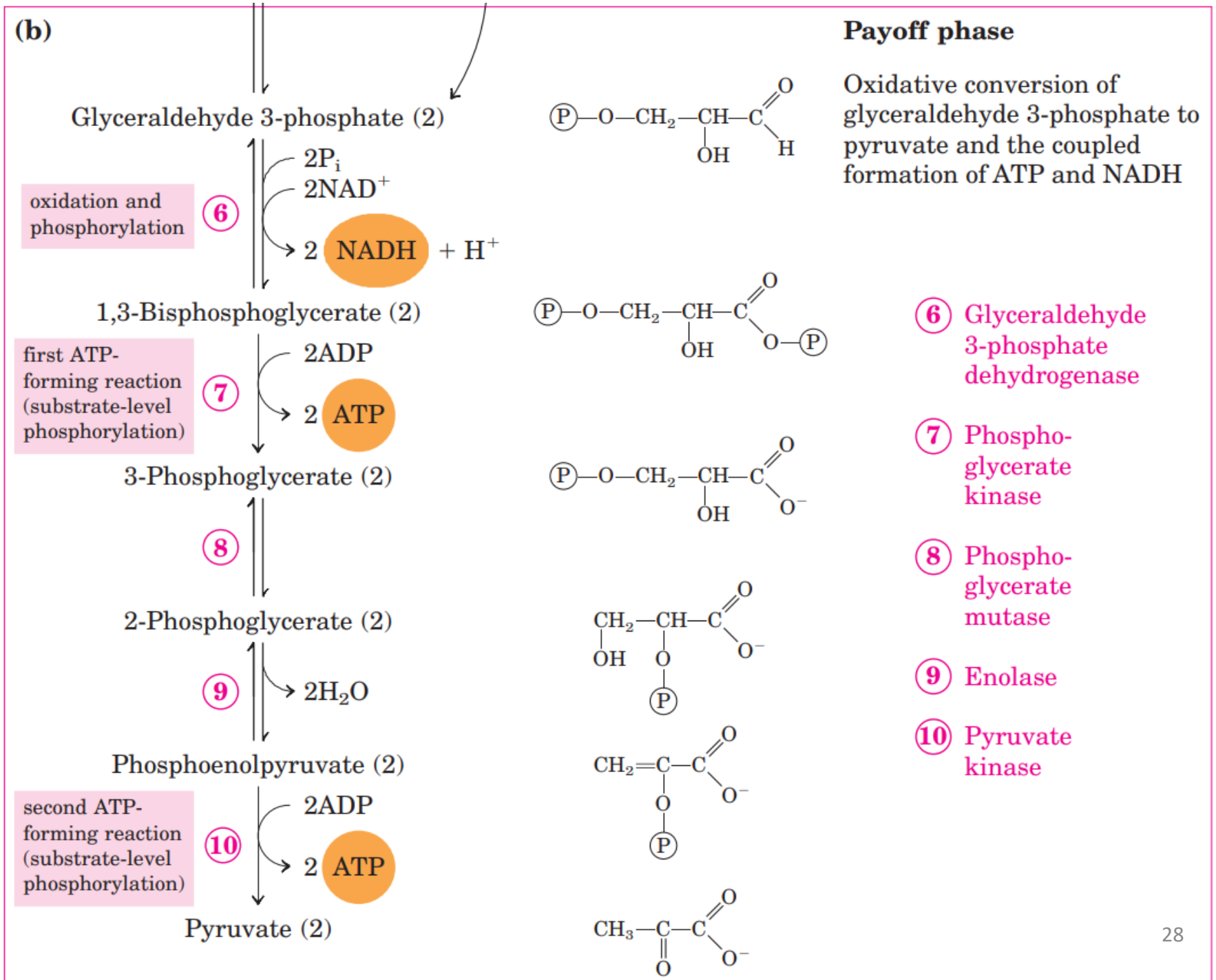


FIGURE 14–9 Entry of glycogen, starch, disaccharides, and hexoses into the preparatory stage of glycolysis.

Glycolysis: Phase 1



Glycolysis: Phase 2



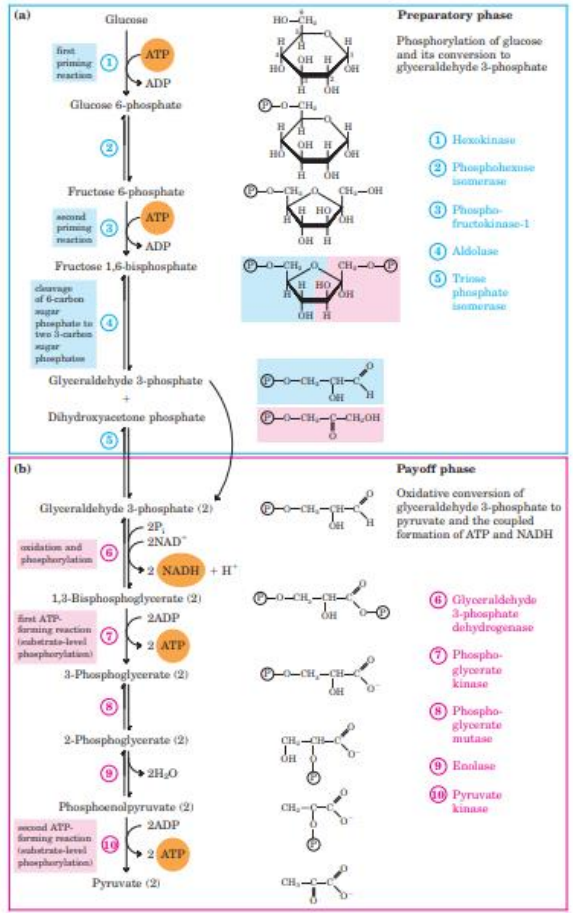
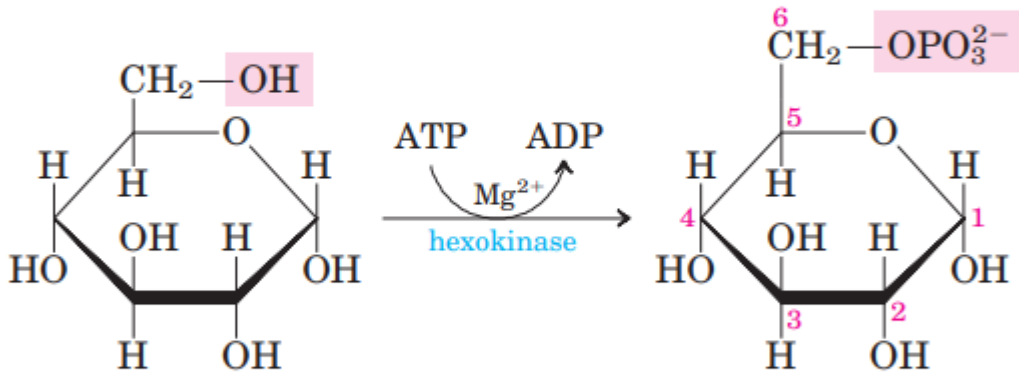


FIGURE 14–2 The two phases of glycolysis. For each molecule of glucose that passes through the preparatory phase **(a),** two molecules of glyceraldehyde 3-phosphate are formed; both pass through the payoff phase **(b).** Pyruvate is the end product of the second phase of glycolysis. For each glucose molecule, two ATP are consumed in the preparatory phase and four ATP are produced in the payoff phase, giving a net yield of two ATP per molecule of glucose converted to pyruvate. The numbered reaction steps are catalyzed by the enzymes listed on the right, and also correspond to the numbered headings in the text discussion. Keep in mind that each phosphoryl group, represented here as Ⓟ, has two negative charges ($—PO_3^{2-}$).

(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:



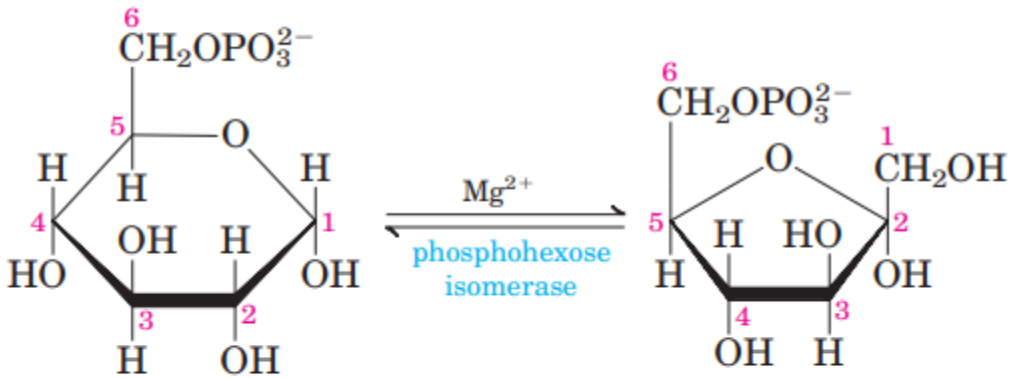
Glucose

Glucose 6-phosphate

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

(2) *Conversion of Glucose 6-Phosphate to Fructose 6-Phosphate*

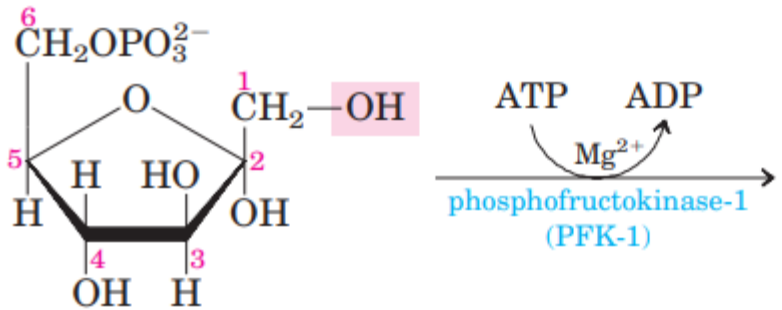
The enzyme **phosphohexose isomerase (phosphoglucose isomerase)** catalyzes the reversible isomerization of glucose 6-phosphate, an aldose, to **fructose 6-phosphate,** a ketose:



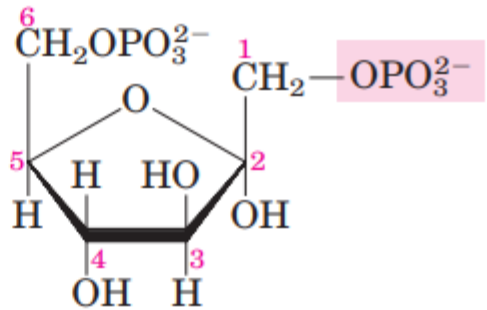
Glucose 6-phosphate — Fructose 6-phosphate

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

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:**



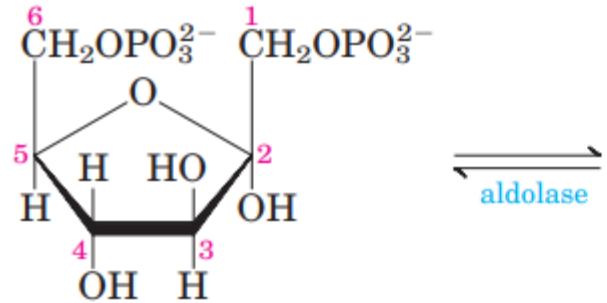
Fructose 6-phosphate



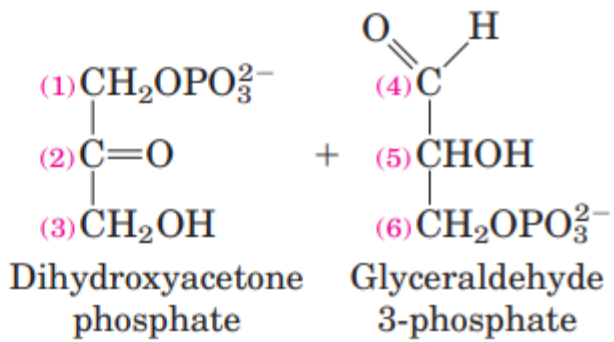
Fructose 1,6-bisphosphate

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

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:



Fructose 1,6-bisphosphate

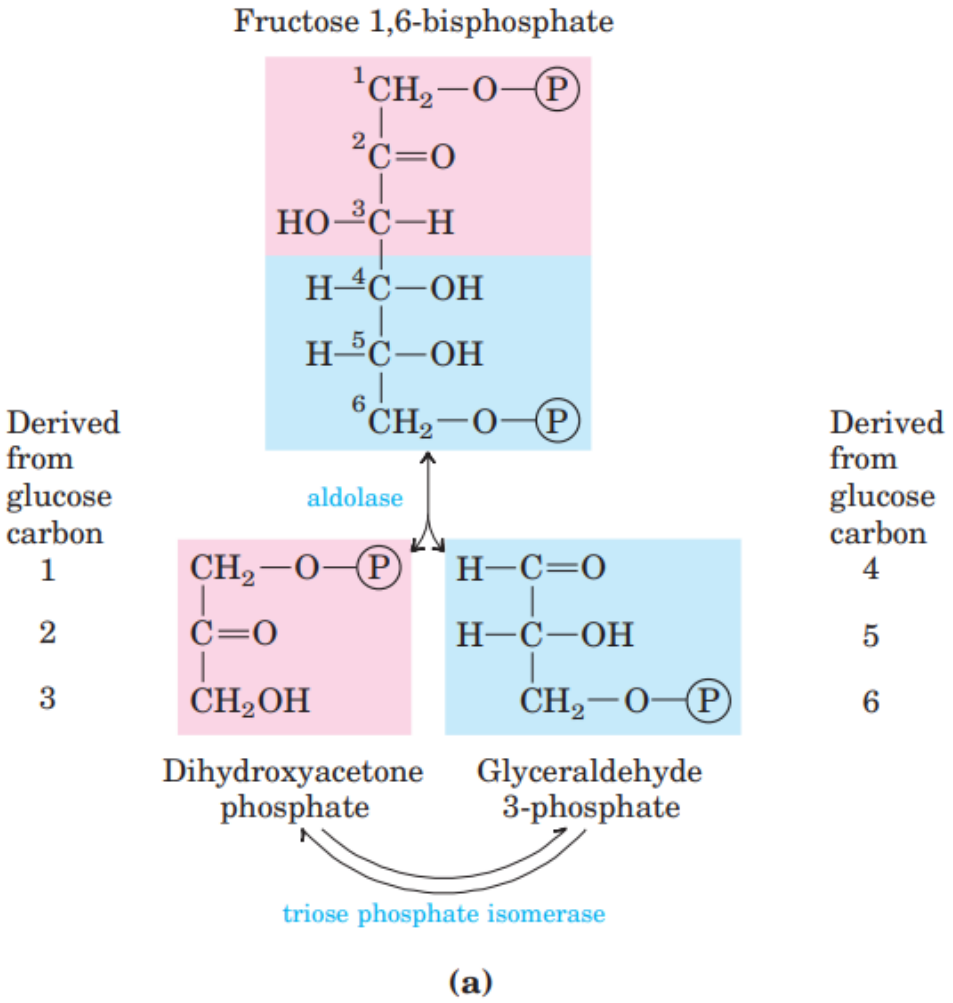


Dihydroxyacetone phosphate

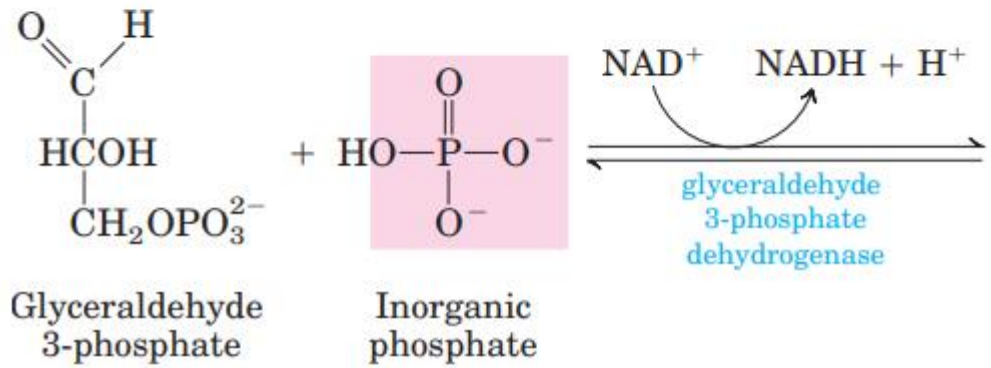
Glyceraldehyde 3-phosphate

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

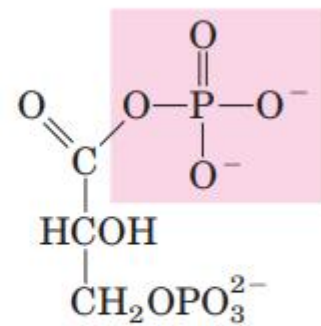
5 Interconversion of the Triose Phosphates



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:**



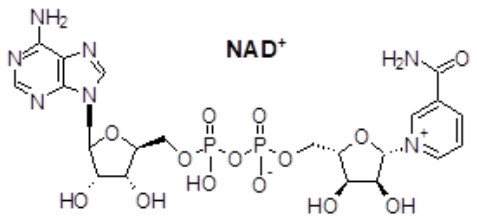
Glyceraldehyde 3-phosphate + Inorganic phosphate

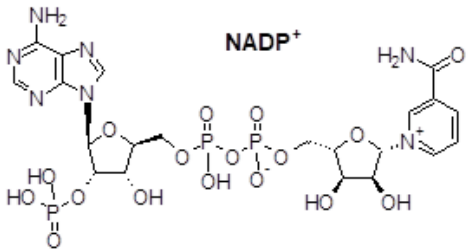


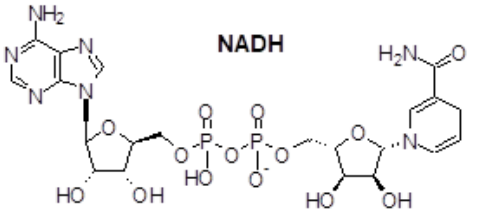
1,3-Bisphosphoglycerate

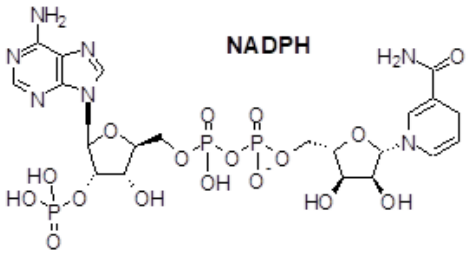
$\Delta G'^{\circ} = 6.3$ kJ/mol

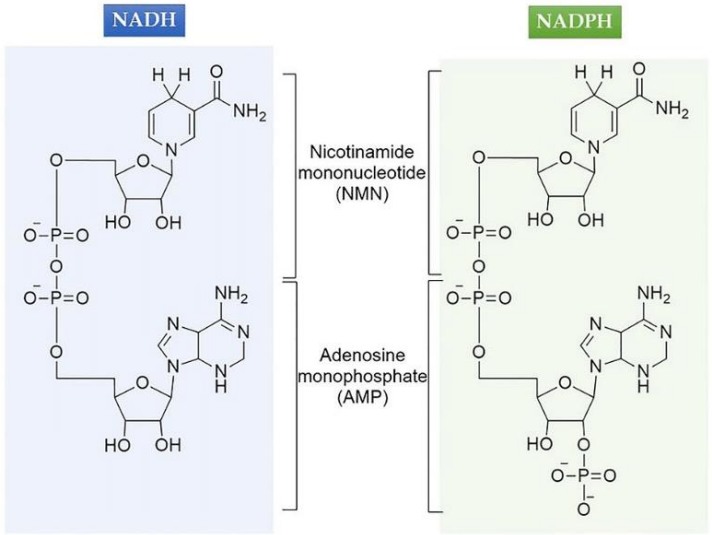
NADP: Nicotinamide Adenine Dinucleotide Phosphate

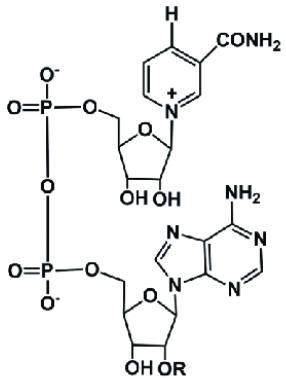


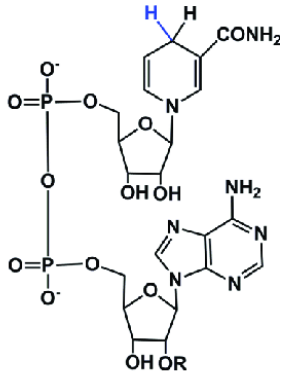


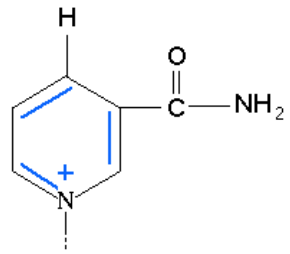


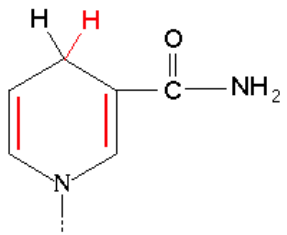


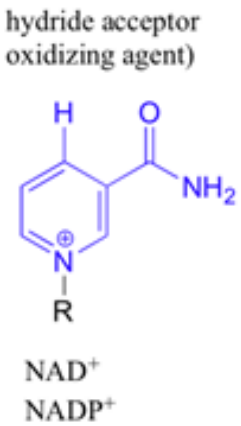


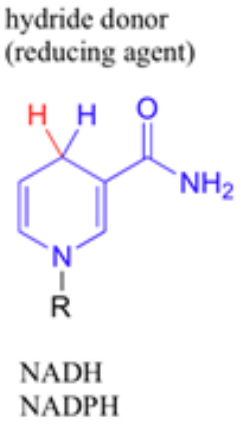


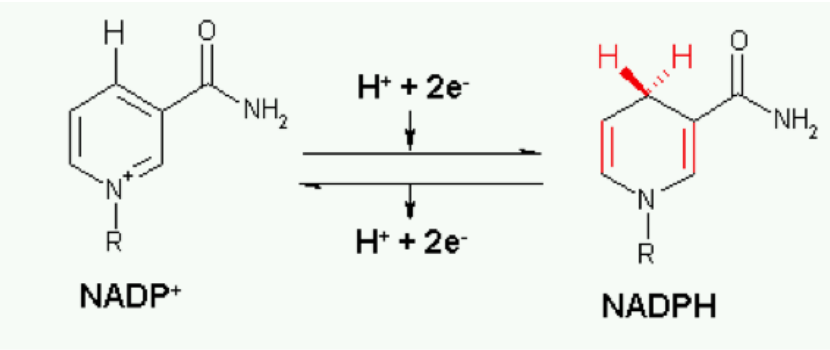












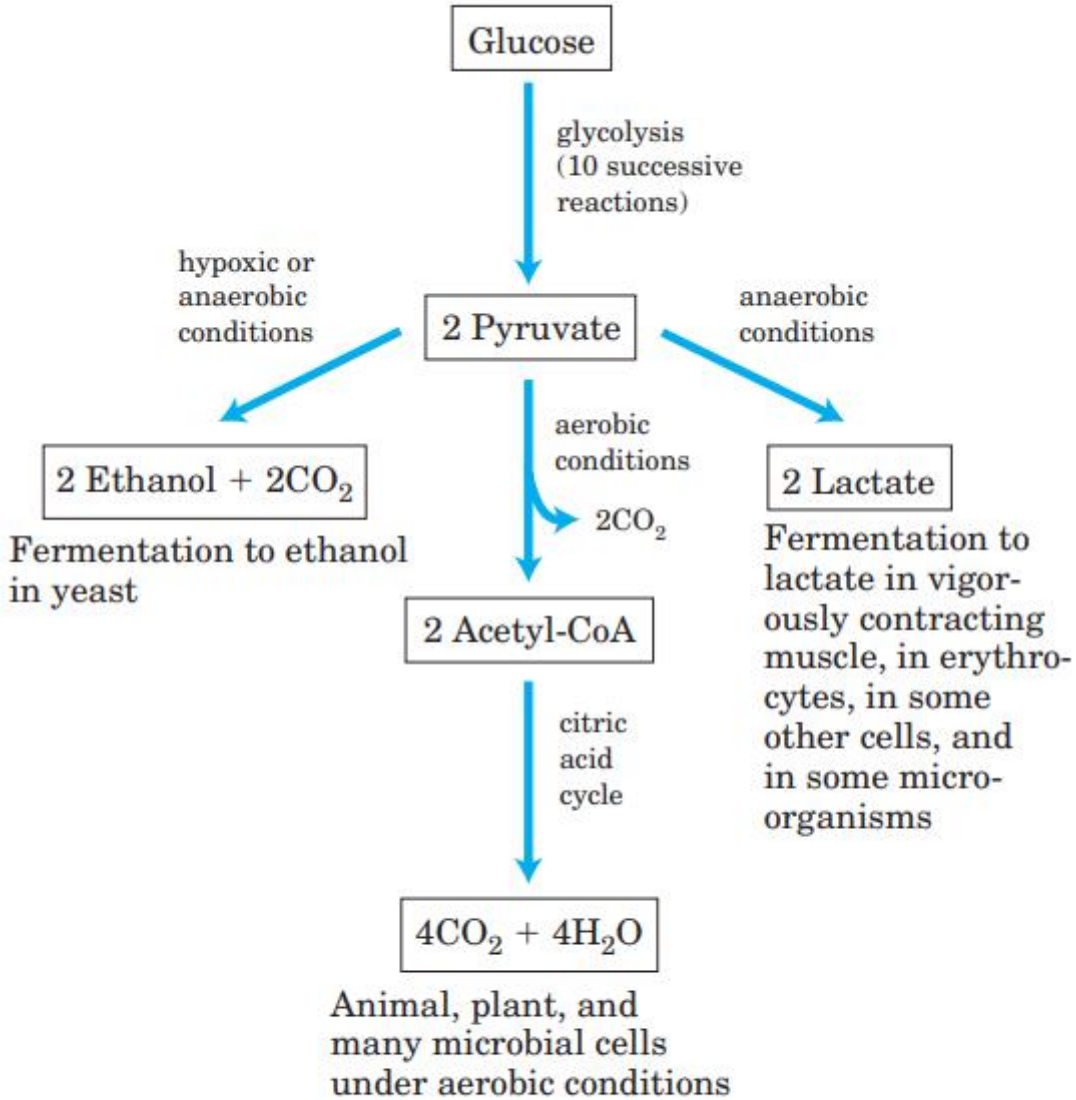


FIGURE 14–3 Three possible catabolic fates of the pyruvate formed in glycolysis. Pyruvate also serves as a precursor in many anabolic reactions, not shown here.

ATP Formation Coupled to Glycolysis During glycolysis some of the energy of the glucose molecule is conserved in ATP, while much remains in the product, pyruvate. The overall equation for glycolysis is

$$\text{Glucose} + 2\text{NAD}^+ + 2\text{ADP} + 2\text{P}_\text{i} \longrightarrow 2\ \text{pyruvate} + 2\text{NADH} + 2\text{H}^+ + 2\text{ATP} + 2\text{H}_2\text{O} \quad (14\text{–}1)$$

For each molecule of glucose degraded to pyruvate, two molecules of ATP are generated from ADP and P_i. We can now resolve the equation of glycolysis into two processes—the conversion of glucose to pyruvate, which is exergonic:

$$\text{Glucose} + 2\text{NAD}^+ \longrightarrow 2\ \text{pyruvate} + 2\text{NADH} + 2\text{H}^+ \quad (14\text{–}2)$$
$$\Delta G_1'^\circ = -146\ \text{kJ/mol}$$

and the formation of ATP from ADP and P_i, which is endergonic:

$$2\text{ADP} + 2\text{P}_\text{i} \longrightarrow 2\text{ATP} + 2\text{H}_2\text{O} \quad (14\text{–}3)$$
$$\Delta G_2'^\circ = 2(30.5\ \text{kJ/mol}) = 61.0\ \text{kJ/mol}$$

The sum of Equations 14–2 and 14–3 gives the overall standard free-energy change of glycolysis, $\Delta G_s'^\circ$:

$$\Delta G_s'^\circ = \Delta G_1'^\circ + \Delta G_2'^\circ = -146\ \text{kJ/mol} + 61.0\ \text{kJ/mol} = -85\ \text{kJ/mol}$$

Under standard conditions and in the cell, glycolysis is an essentially irreversible process, driven to completion by a large net decrease in free energy. At the actual intracellular concentrations of ATP, ADP, and P_i (see Box 13–1) and of glucose and pyruvate, the energy released in glycolysis (with pyruvate as the end product) is recovered as ATP with an efficiency of more than 60%.

What Next?

Energy Remaining in Pyruvate Glycolysis releases only a small fraction of the total available energy of the glucose molecule;

the two molecules of pyruvate formed by glycolysis still contain most of the chemical potential energy of glucose, energy that can be extracted by oxidative reactions in

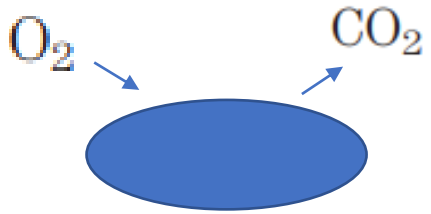
The citric acid cycle and

Oxidative phosphorylation

The Citric Acid cycle

Tricarboxylic acid (TCA) cycle or the Krebs cycle after its discoverer, Hans Krebs

Cellular respiration



Stage 1: Organic fuel molecules—glucose, fatty acids, and some amino acids—are oxidized to yield two-carbon fragments in the form of the acetyl group of **acetyl-coenzyme A** (acetyl-CoA).

Stage 2: The acetyl groups are fed into the **citric acid cycle**, which enzymatically oxidizes them to CO_2 ; the energy released is conserved in the reduced electron carriers **NADH** and $FADH_2$.

Stage 3: Here these reduced coenzymes are themselves oxidized, giving up protons (H^+) and electrons. The electrons are transferred to O_2— the final electron acceptor—via a chain of electron-carrying molecules known as the respiratory chain. In the course of electron transfer, the large amount of energy released is conserved in the form of ATP, by a process called **oxidative phosphorylation**.

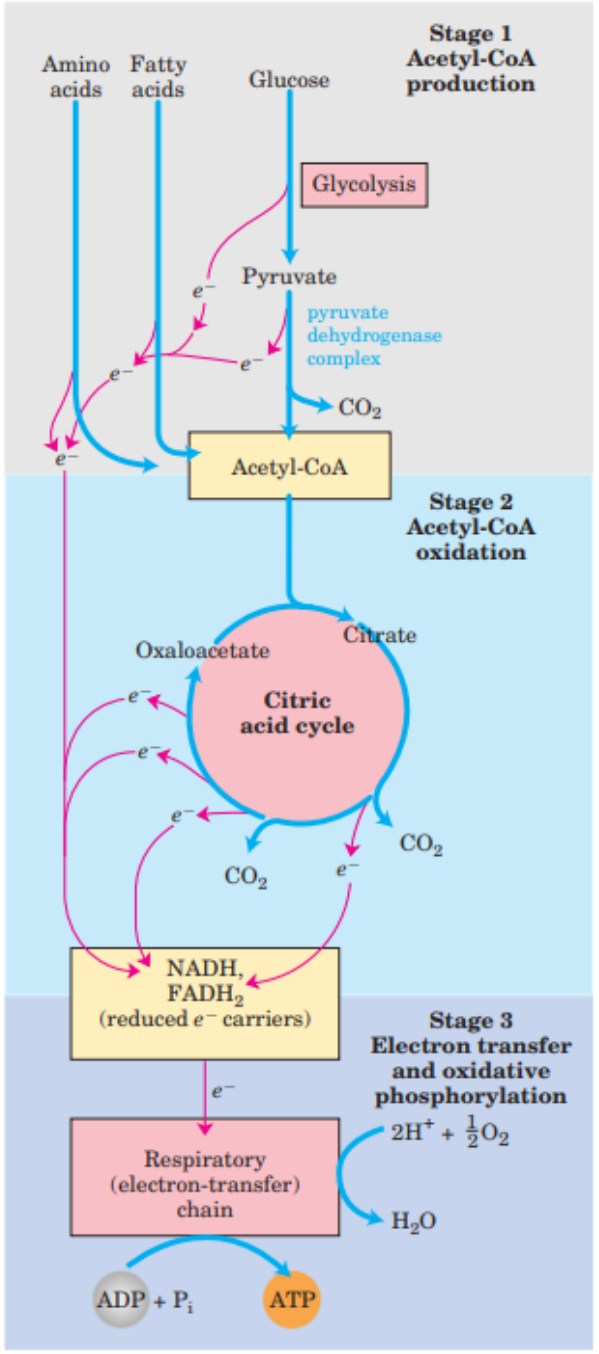


FIGURE 16–1 Catabolism of proteins, fats, and carbohydrates in the three stages of cellular respiration. Stage 1: oxidation of fatty acids, glucose, and some amino acids yields acetyl-CoA. Stage 2: oxidation of acetyl groups in the citric acid cycle includes four steps in which electrons are abstracted. Stage 3: electrons carried by NADH and $FADH_2$ are funneled into a chain of mitochondrial (or, in bacteria, plasma membrane–bound) electron carriers—the respiratory chain—ultimately reducing O_2 to H_2O. This electron flow drives the production of ATP.

Before The Citric Acid cycle

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

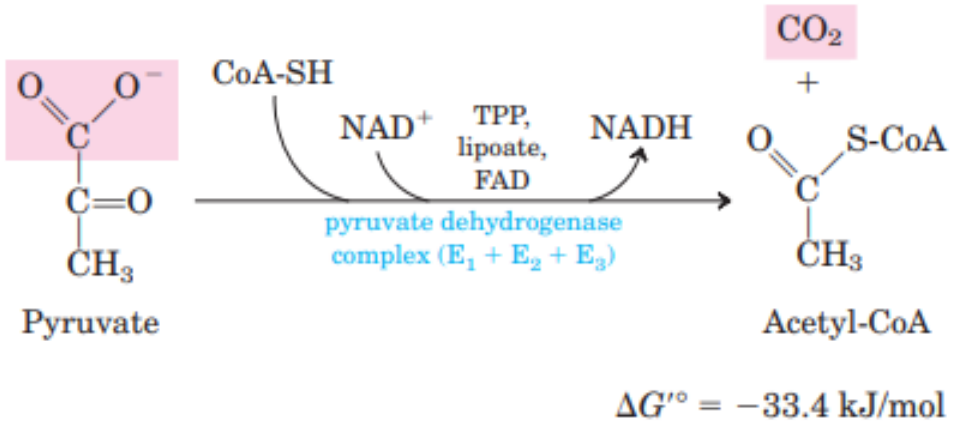


FIGURE 16–2 Overall reaction catalyzed by the pyruvate dehydrogenase complex. The five coenzymes participating in this reaction, and the three enzymes that make up the enzyme complex, are discussed in the text.

Pyruvate dehydrogenase (PDH) complex, a **cluster of enzymes**—multiple copies of each of three enzymes—located in the mitochondria of eukaryotic cells and in the cytosol of prokaryotes. Uses five cofactors.

There are two other enzyme clusters are also involved in subsequent steps.
Alpha-ketoglutarate dehydrogenase, of the **citric acid cycle** and the branched-chain **Alpha -keto acid dehydrogenase**, of the **oxidative pathways** of several amino

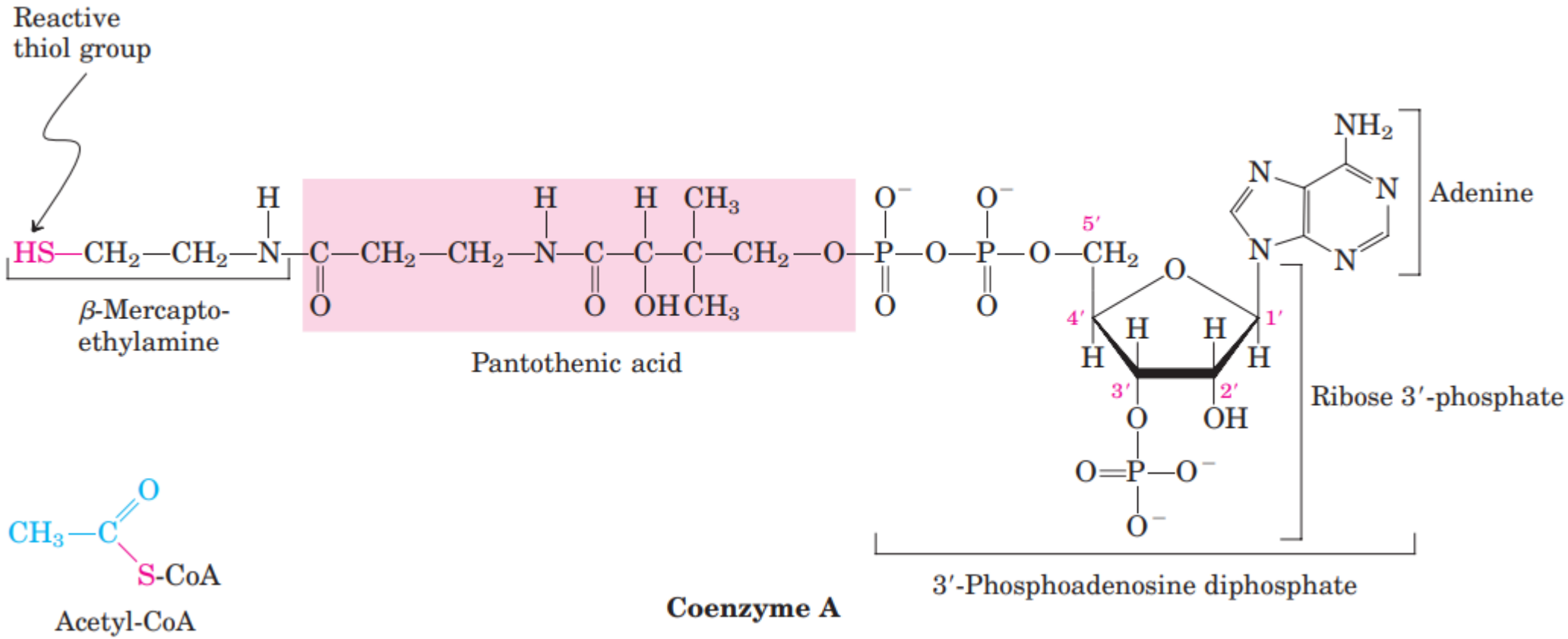


FIGURE 16-3 Coenzyme A (CoA). A hydroxyl group of pantothenic acid is joined to a modified ADP moiety by a phosphate ester bond, and its carboxyl group is attached to β-mercaptoethylamine in amide linkage. The hydroxyl group at the 3′ position of the ADP moiety has a phosphoryl group not present in free ADP. The —SH group of the mercaptoethylamine moiety forms a thioester with acetate in acetyl-coenzyme A (acetyl-CoA) (lower left).

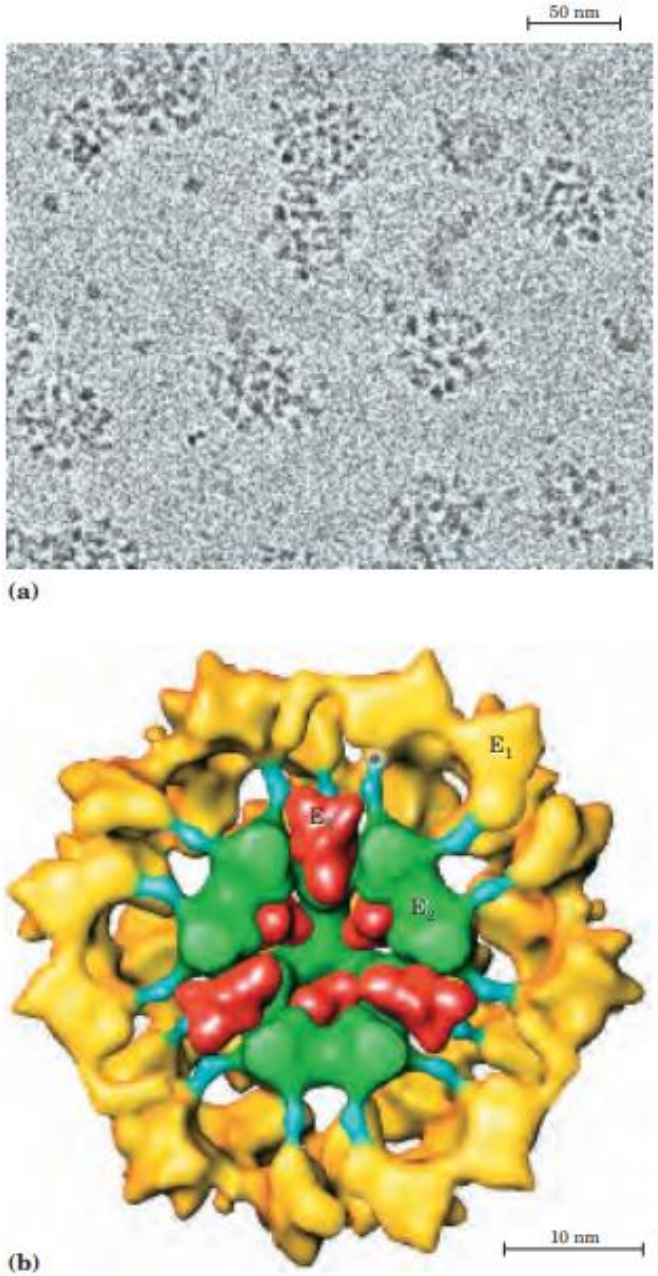


FIGURE 16–5 Structure of the pyruvate dehydrogenase complex **(a)** Cryoelectron micrograph of PDH complexes isolated from bovine kidney. In cryoelectron microscopy, biological samples are viewed at extremely low temperatures; this avoids potential artifacts introduced by the usual process of dehydrating, fixing, and staining. **(b)** Three-dimensional image of PDH complex, showing the subunit structure: E_1, pyruvate dehydrogenase; E_2, dihydrolipoyl transacetylase; and E_3, dihydrolipoyl dehydrogenase. This image is reconstructed by analysis of a large number of images such as those in **(a)**, combined with crystallographic studies of individual subunits. The core (green) consists of 60 molecules of E_2, arranged in 20 trimers to form a pentagonal dodecahedron. The lipoyl domain of E_2 (blue) reaches outward to touch the active sites of E_1 molecules (yellow) arranged on the E_2 core. A number of E_3 subunits (red) are also bound to the core, where the swinging arm on E_2 can reach their active sites. An asterisk marks the site where a lipoyl group is attached to the lipoyl domain of E_2. To make the structure clearer, about half of the complex has been cut away from the front. This model was prepared by Z. H. Zhou et al. (2001); in another model, proposed by J. L. S. Milne et al. (2002), the E_3 subunits are located more toward the periphery (see Further Read-

The Citric Acid cycle

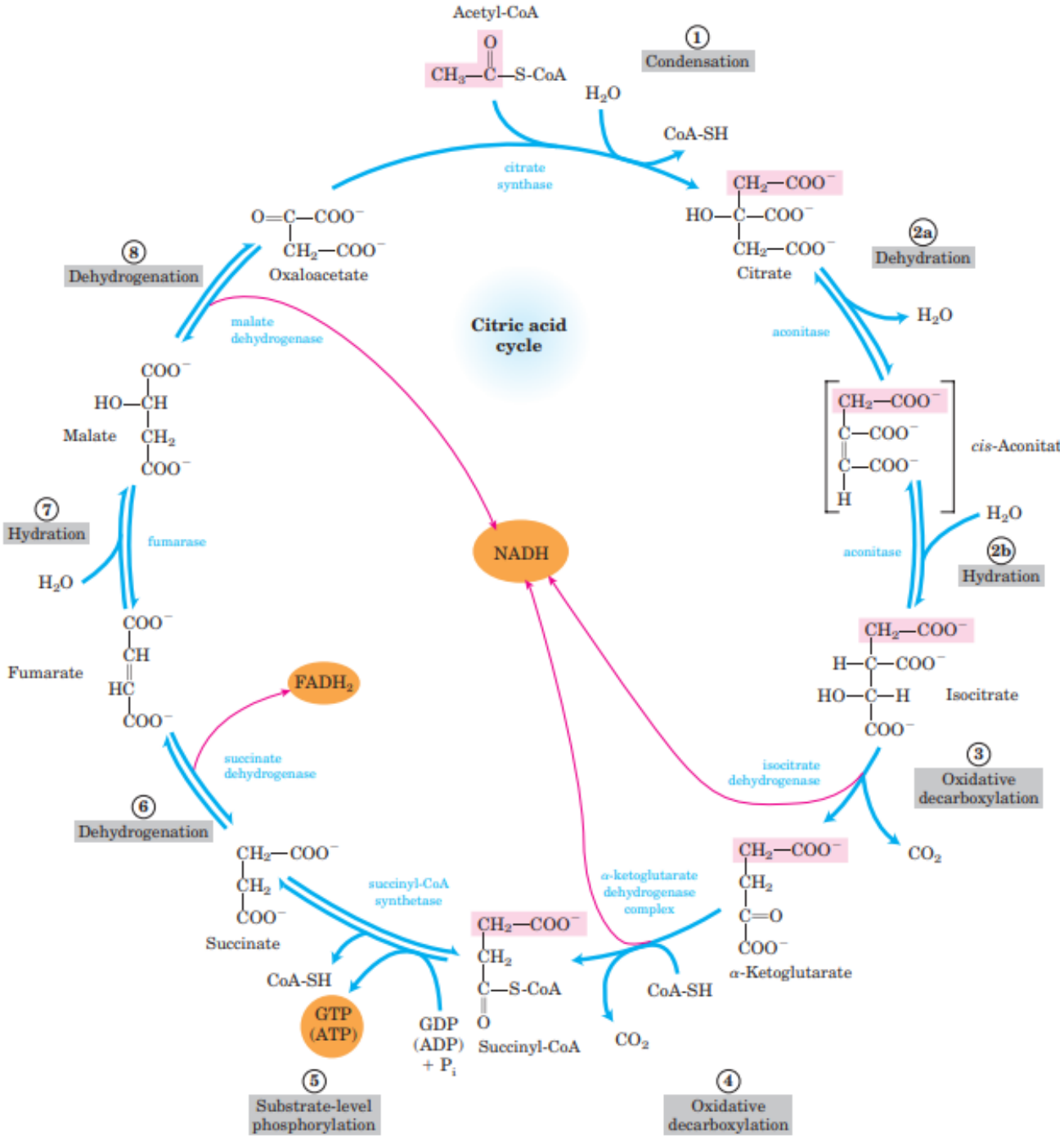
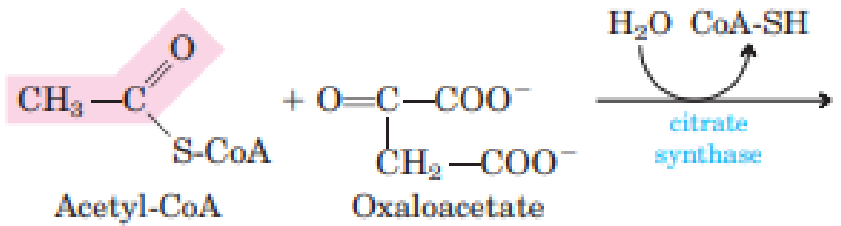
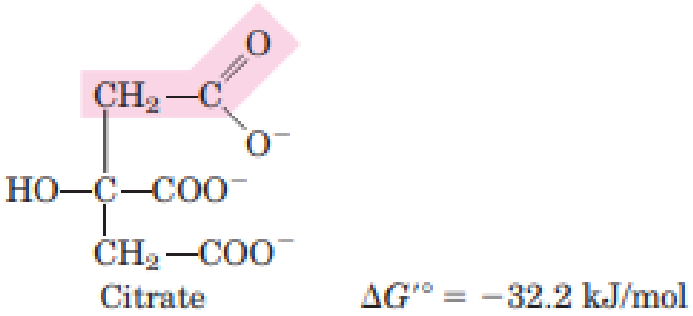


FIGURE 16–7 Reactions of the citric acid cycle. The carbon atoms shaded in pink are those derived from the acetate of acetyl-CoA in the first turn of the cycle; these are *not* the carbons released as CO_2 in the first turn. Note that in succinate and fumarate, the two-carbon group derived from acetate can no longer be specifically denoted; because succinate and fumarate are symmetric molecules, C-1 and C-2 are indistinguishable from C-4 and C-3. The number beside each reaction step corresponds to a numbered heading on pages 608–612. The red arrows show where energy is conserved by electron transfer to FAD or NAD^+, forming $FADH_2$ or NADH + H^+. Steps ①, ③, and ④ are essentially irreversible in the cell; all other steps are reversible. The product of step ⑤ may be either ATP or GTP, depending on which succinyl-CoA synthetase isozyme is the catalyst.

The Citric Acid Cycle Has Eight Steps

(1) *Formation of Citrate* The first reaction of the cycle is the condensation of acetyl-CoA with **oxaloacetate** to form **citrate,** catalyzed by **citrate synthase:**





$\Delta G'^{\circ} = -32.2$ kJ/mol

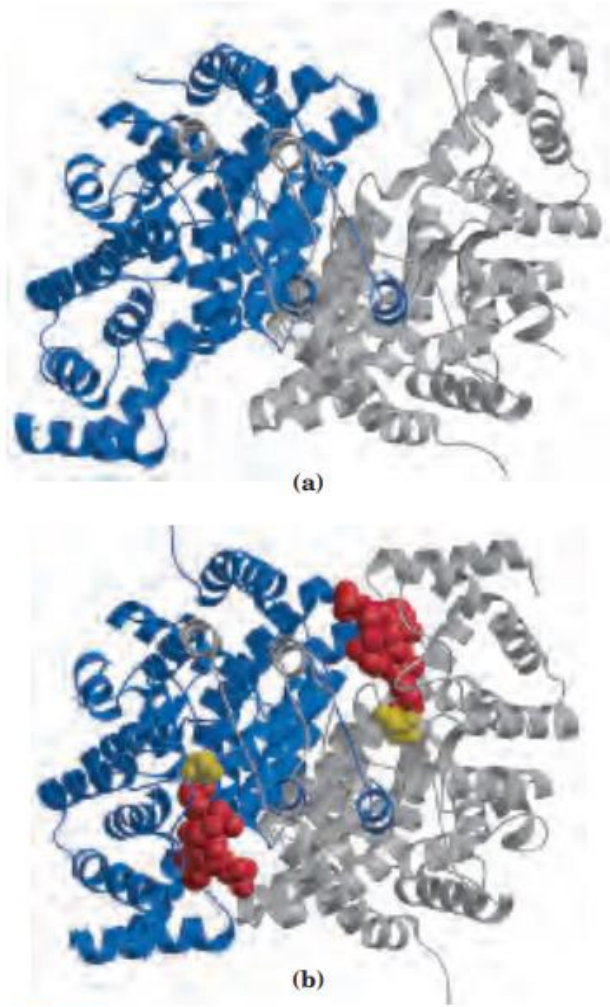
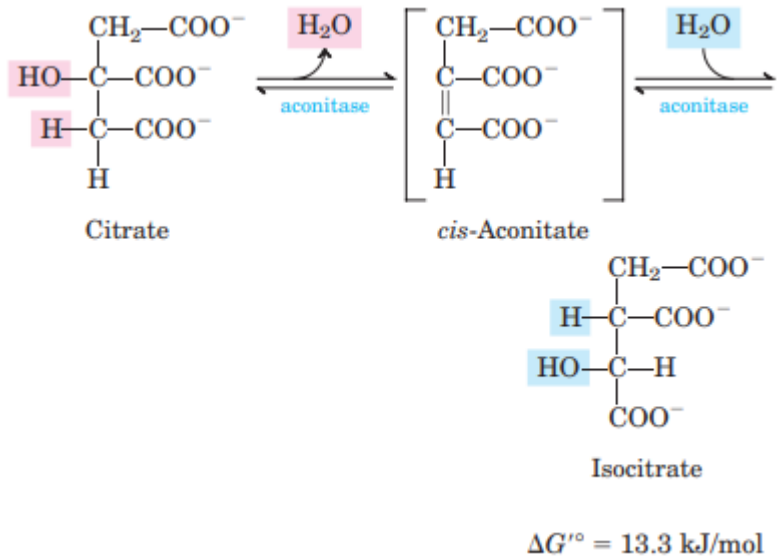
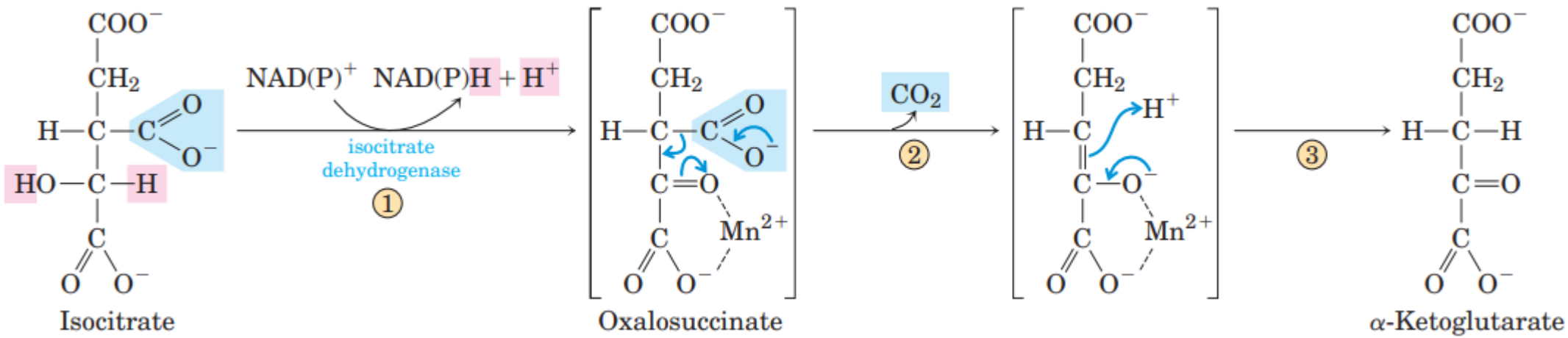


FIGURE 16–8 Structure of citrate synthase. The flexible domain of each subunit undergoes a large conformational change on binding oxaloacetate creating a binding site for acetyl-CoA. **(a)** open form of the enzyme alone (PDB ID 5CSC); **(b)** closed form with bound oxaloacetate (yellow) and a stable analog of acetyl-CoA (carboxymethyl-CoA; red) (derived from PDB ID 5CTS).

2: Formation of Isocitrate via cis-Aconitate The enzyme aconitase (more formally, aconitate hydratase) catalyzes the reversible transformation of citrate to isocitrate, through the intermediary formation of the tricarboxylic acid cis-aconitate

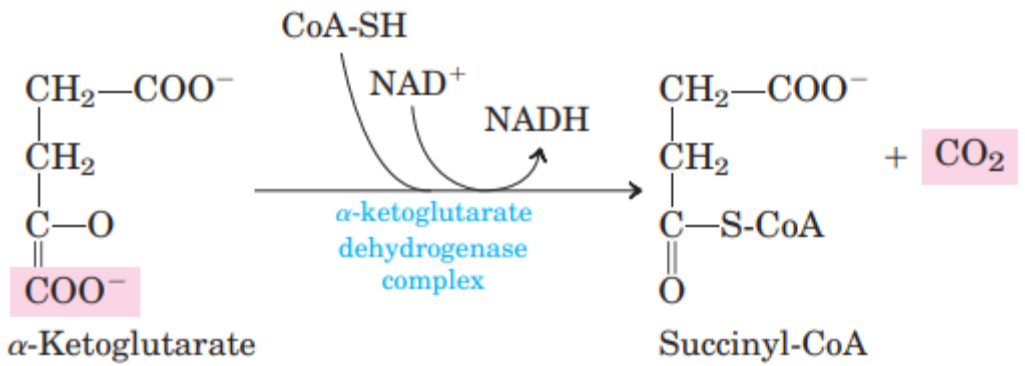


3: Oxidation of Isocitrate to Alpha -Ketoglutarate and CO2



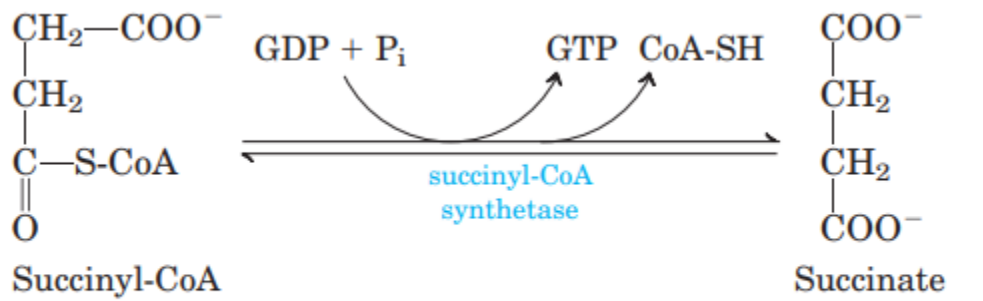
④ *Oxidation of α-Ketoglutarate to Succinyl-CoA and CO_2*

The next step is another oxidative decarboxylation, in which α-ketoglutarate is converted to **succinyl-CoA** and CO_2 by the action of the **α-ketoglutarate dehydrogenase complex;** NAD^+ serves as electron acceptor and CoA as the carrier of the succinyl group. The energy of oxidation of α-ketoglutarate is conserved in the formation of the thioester bond of succinyl-CoA:



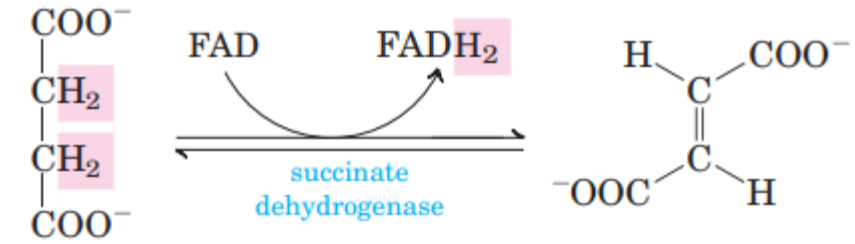
$\Delta G'^{\circ} = -33.5$ kJ/mol

5 *Conversion of Succinyl-CoA to Succinate* Succinyl-CoA, like acetyl-CoA, has a thioester bond with a strongly negative standard free energy of hydrolysis ($\Delta G'^\circ \approx -36$ kJ/mol). In the next step of the citric acid cycle, energy released in the breakage of this bond is used to drive the synthesis of a phosphoanhydride bond in either GTP or ATP, with a net $\Delta G'^\circ$ of only -2.9 kJ/mol. **Succinate** is formed in the process:



$\Delta G'^\circ = -2.9$ kJ/mol

⑥ *Oxidation of Succinate to Fumarate* The succinate formed from succinyl-CoA is oxidized to **fumarate** by the flavoprotein **succinate dehydrogenase:**

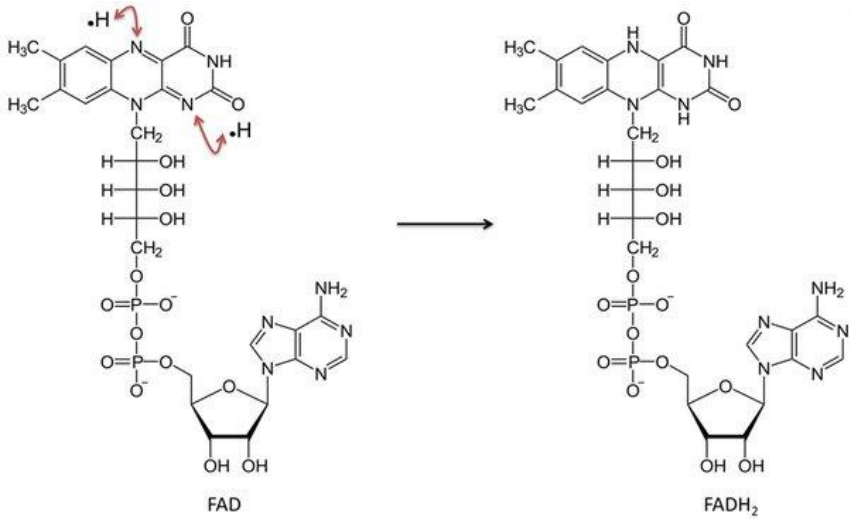


Succinate

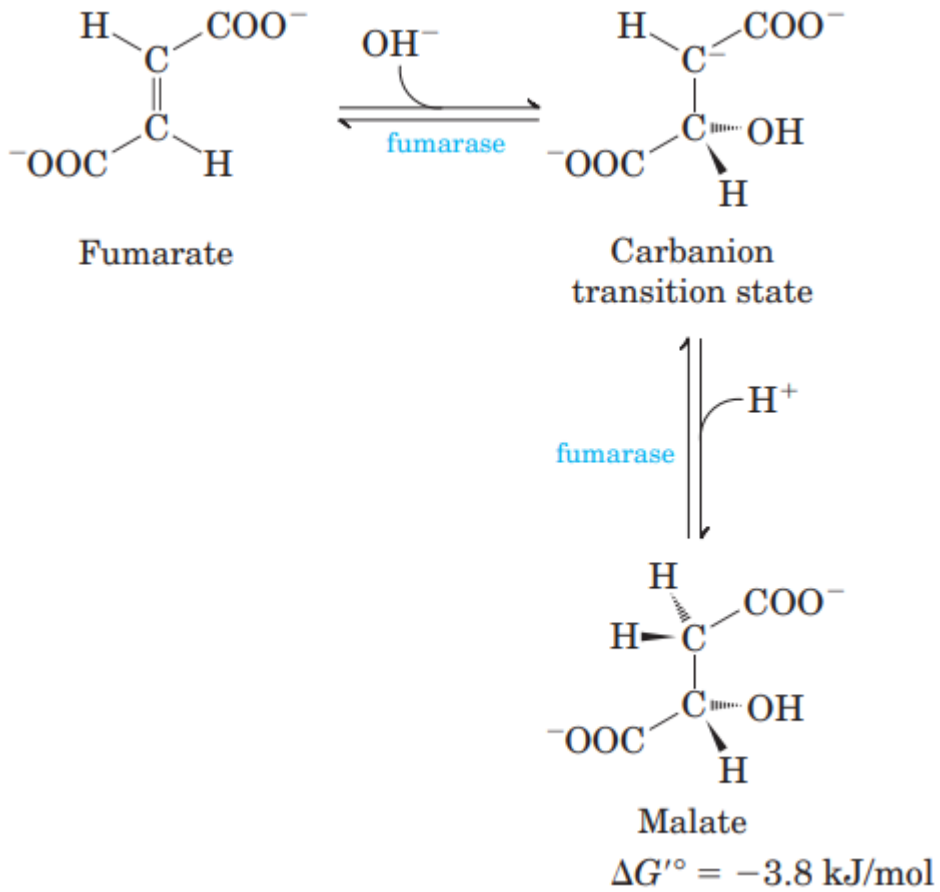
Fumarate

$\Delta G'^{\circ} = 0$ kJ/mol

FAD: Flavin adenine dinucleotide



7 *Hydration of Fumarate to Malate* The reversible hydration of fumarate to **L-malate** is catalyzed by **fumarase**



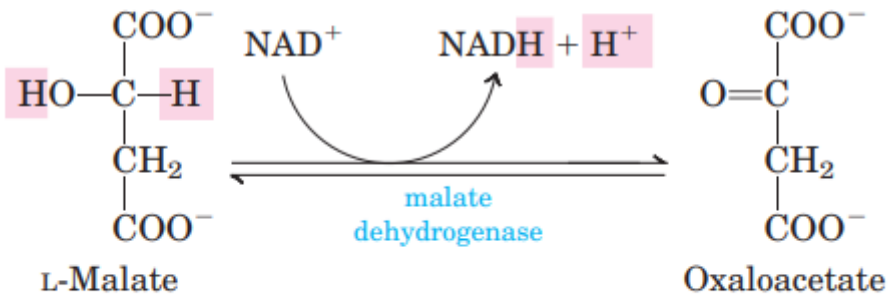
Fumarate

Carbanion transition state

Malate

$\Delta G'^{\circ} = -3.8$ kJ/mol

(8) *Oxidation of Malate to Oxaloacetate* In the last reaction of the citric acid cycle, NAD-linked **L-malate dehydrogenase** catalyzes the oxidation of L-malate to oxaloacetate:



L-Malate Oxaloacetate

$\Delta G'^{\circ} = 29.7$ kJ/mol

TCA Products:

Energy

+

Carbon dioxide

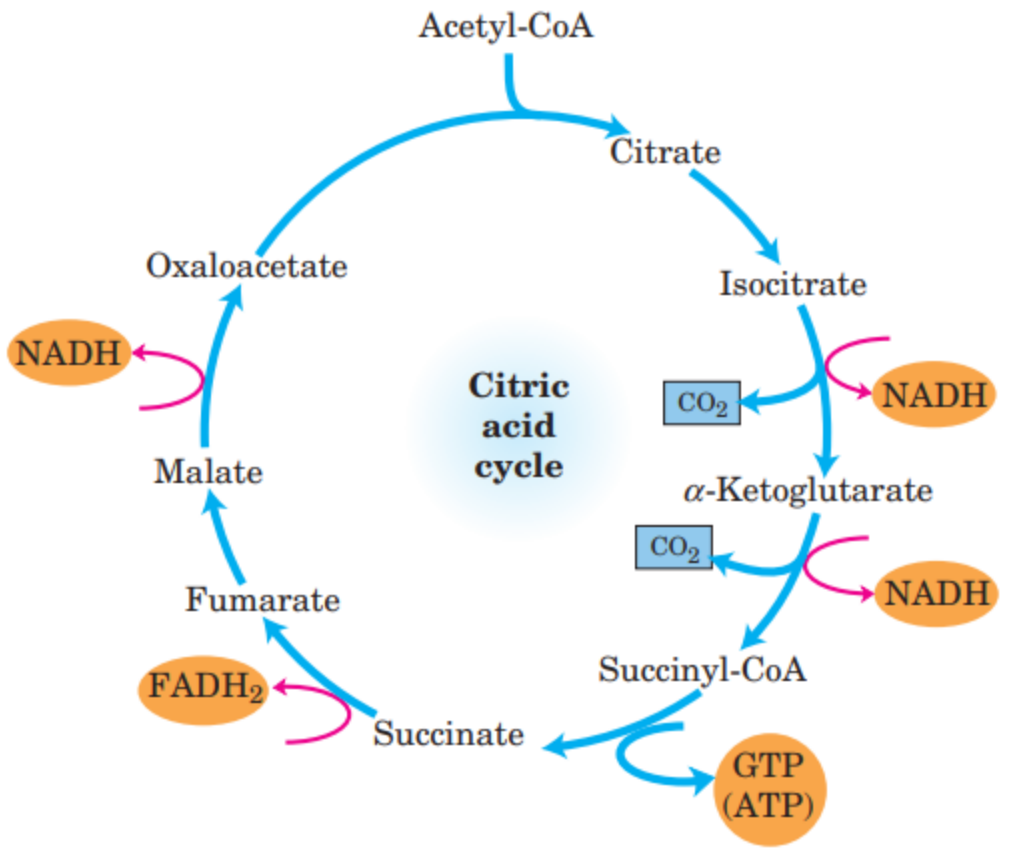


FIGURE 16–13 Products of one turn of the citric acid cycle. At each turn of the cycle, three NADH, one $FADH_2$, one GTP (or ATP), and two CO_2 are released in oxidative decarboxylation reactions. Here and in several following figures, all cycle reactions are shown as proceeding in one direction only, but keep in mind that most of the reactions are reversible (see Fig. 16–7).