Chapter 4

Cellular Respiration

Review

Cellular respiration is the first major topic for which you apply your knowledge of chemistry. For the most part, however, the chemistry is descriptive—that is, you won't have to solve chemical equations or even memorize structural formulas. Instead, you need to provide names of major molecules (usually just the reactants and the products), describe their sequence in a metabolic process, and most important, describe how the process accomplishes its metabolic objectives.

Energy Basics

Energy is required to do work—to do things, to move things, to put things together. How energy does work follows two laws of thermodynamics:

- The **first law of thermodynamics** states that the total amount of energy in the universe remains constant. Energy cannot be created or destroyed, but it can be converted from one form to another. These forms of energy include **kinetic energy** (energy of motion) and **potential energy** (stored energy, such as that in a chemical bond or in something elevated in a gravitational field).
- The **second law of thermodynamics** states that when energy is converted from one form to another, some energy is "lost." The word "lost" is not intended to mean "gone" (that would be a violation of the first law of thermodynamics). Instead, it means that not all of the energy gets passed from one usable form to another, that some of the energy becomes unusable or unable to do work. The unusable energy is usually in the form of heat. Further, as additional energy conversions occur, more energy becomes unusable, things become disorganized, and disorder or randomness increases. That disorder is called **entropy.** Because energy is constantly moving from one form to another, a consequence of the second law of thermodynamics is that entropy increases in the universe.

Energy conversions are usually discussed within the context of a **system,** such as a chemical reaction, a cell, a multicellular organism, or a planet like Earth. It can be a **closed system,** where only energy transfers among specific items are considered, or it can be an **open system,** in which exchanges of energy with the surroundings are included. Living things and the earth are open systems because they receive energy from sunlight.

In an **exergonic** chemical reaction, there is a net *release* of energy. These reactions can occur spontaneously, that is, without an input of energy. When glucose is broken down to CO_2 and H_2O during cellular respiration, for example, energy that is released from the reaction is stored in ATP, a source of energy for metabolic reactions:

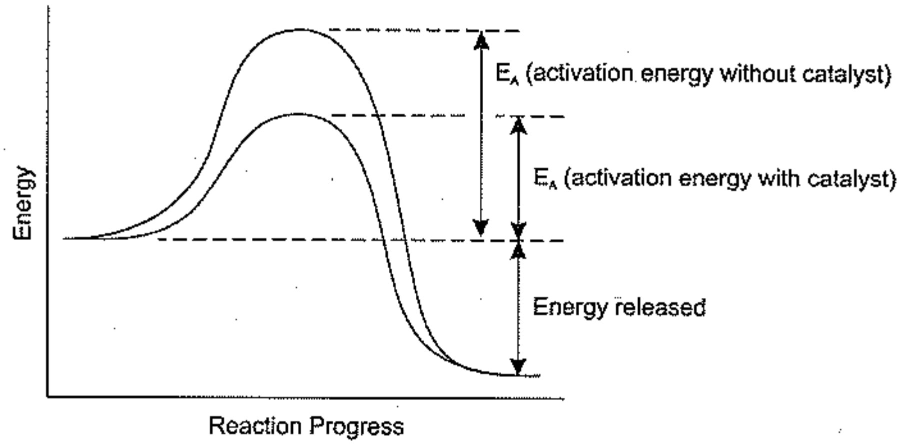
$$C_6H_{12}O_6 + 6\,O_2 \rightarrow 6\,CO_2 + 6\,H_2O + ATP$$

In an **endergonic** reaction, energy must be *added* to the reaction for it to occur. In photosynthesis, for example, sunlight provides the energy necessary for the production of glucose from CO_2 and H_2O:

$$6\,CO_2 + 6\,H_2O + \text{sunlight} \rightarrow C_6H_{12}O_6 + 6\,O_2$$

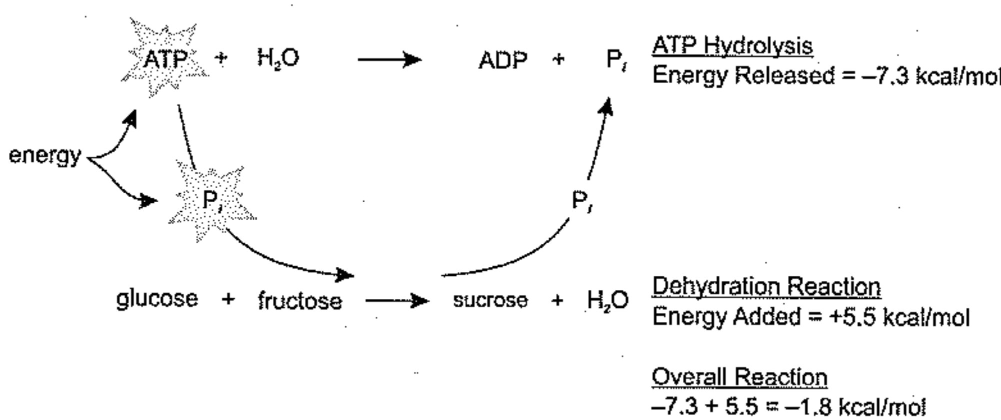
Note that the overall equation for photosynthesis is the reverse of that for respiration, except that the form of energy is different (ATP vs. sunlight).

Even though an exergonic reaction *can* occur spontaneously, it may not. Before most reactions can occur, **activation energy** is required to contort or destabilize the reactants. The necessary activation energy can be lowered by the presence of a catalyst (such as a metal ion or an enzyme). However, the overall energy of a reaction—the energy released or the energy required—is not changed by the presence of a catalyst (Figure 4-1).



Activation Energy for an Exergonic Reaction
Figure 4-1

Most metabolic reactions are endergonic and require an input of energy. That energy usually comes from the hydrolysis of adenosine *tri*phosphate (ATP) to adenosine *di*phosphate (ADP). The hydrolysis of ATP (an *exergonic* reaction) is **coupled** with the *endergonic* metabolic reaction. The coupling usually involves the transfer of energy with one of the inorganic phosphates (P_i) from ATP to one of the reactants, as shown in Figure 4-2. Coupled reactions with ATP (and with the help of specific enzymes) are typical of most endergonic metabolic reactions. Note that added energy is expressed as a positive number, whereas released energy is expressed as a negative number.



A Coupled Reaction
Figure 4-2

In order for life to persist, living things require a constant input of energy. That energy is used to maintain order in opposition to the entropy that increases as a result of chemical reactions. Without an input of energy, entropy increases, cells deteriorate, and death follows. **Photosynthesis,** the process of incorporating energy from sunlight into carbohydrates, and **respiration,** the process of extracting energy from those carbohydrates, provide the energy that allows cells to maintain order, minimize entropy, and remain alive.

Generating ATP

Phosphorylation is the process of adding energy and an inorganic phosphate to ADP to make ATP:

$$\text{energy} + P_i + \text{ADP} \rightarrow \text{ATP}$$

There are two basic mechanisms of phosphorylation in cells:

1. **Substrate level phosphorylation** occurs when a phosphate group *and* its associated energy are transferred to ADP to form ATP. The *substrate* molecule (a molecule with the phosphate group) donates the high energy phosphate group. Such phosphorylation occurs during glycolysis, the initial breakdown process of glucose (discussed later in this chapter).
2. **Oxidative phosphorylation** occurs when a phosphate group is added to ADP to form ATP, but the energy for the bond does not accompany the phosphate group. Instead, electrons give up energy for generating ATP during each step of a process, where electrons are transferred from one molecule (electron carrier) to another in a chain of reactions.

Cellular respiration is the ATP-generating process that occurs in cells. As previewed earlier, energy is extracted from energy-rich glucose to form ATP. The chemical equation describing this process is:

$$C_6H_{12}O_6 + 6\ O_2 \rightarrow 6\ CO_2 + 6\ H_2O + \text{energy}$$

$C_6H_{12}O_6$ is glucose, but sometimes you see CH_2O or $(CH_2O)_n$. These are general formulas for glucose or any carbohydrate.

When oxygen is available, most cells will generate ATP by **aerobic respiration.** There are three steps that occur in this ATP generating pathway:

1. **Glycolysis**
2. **Krebs cycle** (citric acid cycle)
3. **Oxidative phosphorylation**

When oxygen is not available, cells will generate ATP by **anaerobic respiration.** There are two slightly different pathways for this process:

1. **Alcohol fermentation**
2. **Lactic acid fermentation**

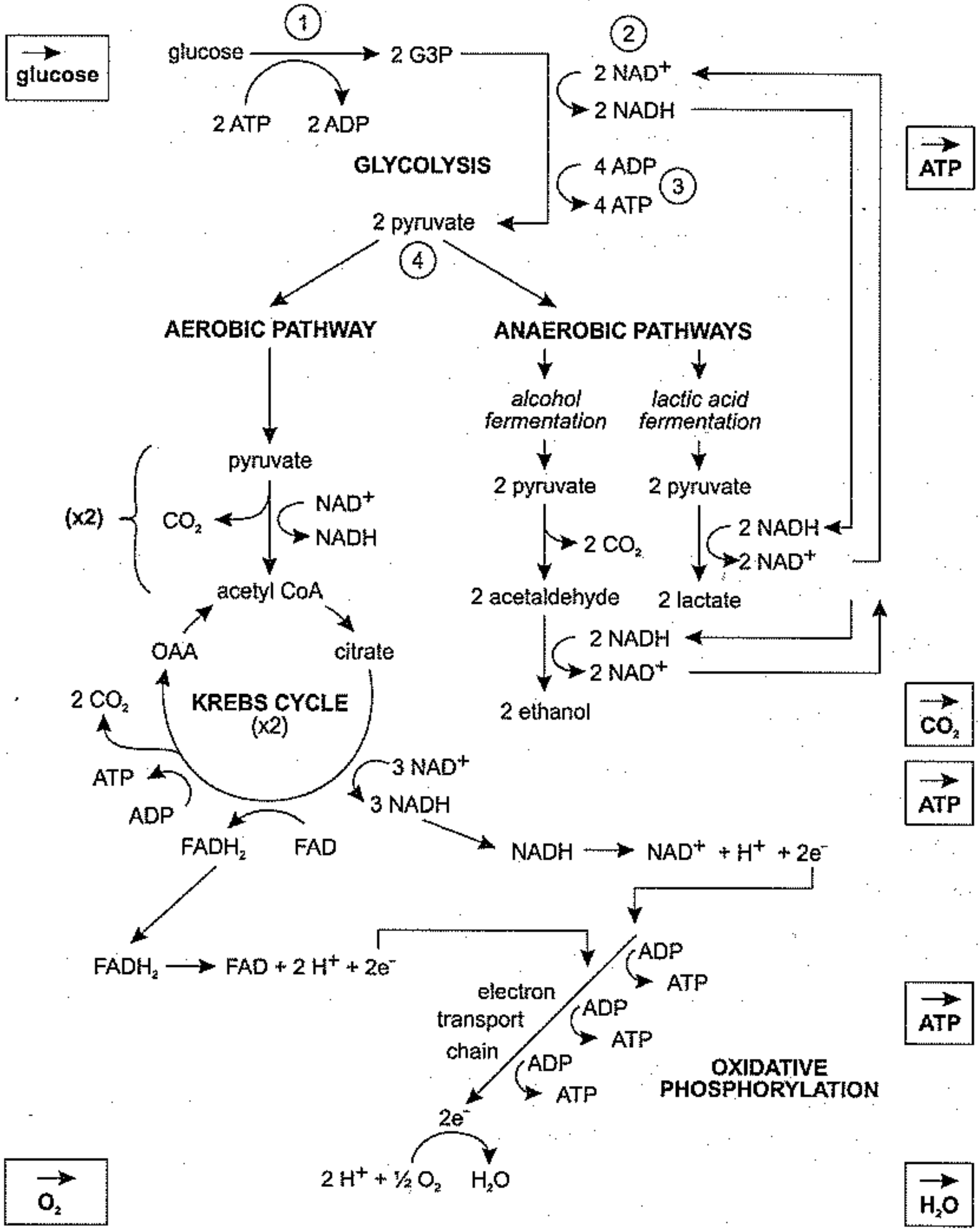
Each of these processes is discussed in detail in the following section. Refer to Figure 4-3. In addition to obtaining energy directly from glucose, other carbohydrates, such as starch and glycogen, can be hydrolyzed to glucose, and sucrose can be hydrolyzed to glucose and fructose. So they all end up being glucose or fructose and enter the glycolytic (glycolysis) pathway.

Proteins can also be a source of energy. Proteins that are eaten are digested to amino acids before they are absorbed into the bloodstream. Body proteins, if necessary, can be hydrolyzed to amino acids. Amino groups ($-NH_2$) are first stripped from the amino acids and then excreted as waste. The remainders of the amino acids are then enzymatically converted to various substances that enter intermediate steps of glycolysis or the Krebs cycle.

Fats are storage molecules for energy so they, too, can be sources of energy. Glycerol and fatty acids are obtained from fats by hydrolysis or by the digestion of fats that are eaten. After enzymatic conversions, glycerol enters glycolysis and fatty acids enter the Krebs cycle (as acetyl CoA).

Glycolysis

Glycolysis is the decomposition (lysis) of **glucose** (glyco) to **pyruvate** (or **pyruvic acid**). The steps are summarized here and in Figure 4-3.



Respiration
Figure 4-3

For each molecule of glucose:

1. **2 ATP are added.** The first several steps require the input of ATP. This changes glucose in preparation for subsequent steps.
2. **2 NADH are produced.** NADH, a *coenzyme,* is an electron carrier when NAD^+ combines with two energy-rich electrons and H+ (obtained from an intermediate molecule during the breakdown of glucose). As a result, NADH is an energy-rich molecule.
3. **4 ATP are produced by substrate-level phosphorylation.**
4. **2 pyruvate are formed.**

In summary, glycolysis takes 1 glucose molecule and turns it into 2 pyruvate, 2 NADH, and a net of 2 ATP (made 4 ATP, but used 2 ATP). The process occurs in the cytosol.

The Krebs Cycle

The Krebs cycle details what happens to pyruvate, the end product of glycolysis. Although the Krebs cycle is described for 1 pyruvate, remember that glycolysis produces 2 pyruvate. In Figure 4-3, the "× 2" next to the pyruvate and the Krebs cycle is a reminder to multiply the products of this cycle by 2 to account for the products of a single glucose.

The processing of pyruvate can be summarized as follows:

1. **Pyruvate to acetyl CoA.** In a step leading up to the actual Krebs cycle, pyruvate combines with coenzyme A (CoA) to produce acetyl CoA. In that reaction, 1 NADH and 1 CO_2 are also produced.
2. **Krebs cycle: 3 NADH, 1 $FADH_2$, 1 ATP, 2 CO_2.** The Krebs cycle begins when acetyl CoA combines with OAA (oxaloacetate) to form citrate. There are seven intermediate products. Along the way, 3 NADH, 1 $FADH_2$, and 1 ATP are made, and 2 CO_2 are released. $FADH_2$, like NADH, is a coenzyme, accepting electrons during a reaction.

The CO_2 produced by the Krebs cycle is the CO_2 animals exhale when they breathe.

Oxidative Phosphorylation

Oxidative phosphorylation is the process of producing ATP from NADH and $FADH_2$. Electrons from NADH and $FADH_2$ pass along an **electron transport chain (ETC).** The chain consists of proteins that pass these electrons from one carrier protein to the next. Some carrier proteins, such as the **cytochromes,** include nonprotein parts containing iron. Along each step of the chain, the electrons give up energy used to phosphorylate ADP to ATP. NADH provides electrons that have enough energy to generate about 3 ATP, while $FADH_2$ generates about 2 ATP.

The final electron acceptor of the electron transport chain is **oxygen.** The ½ O_2 accepts the two electrons and, together with 2 H^+, forms water.

One of the carrier proteins in the electron transport chain, **cytochrome c,** is so ubiquitous among living organisms that the approximately 100-amino-acid sequence of the protein is often compared among species to assess genetic relatedness.

Mitochondria

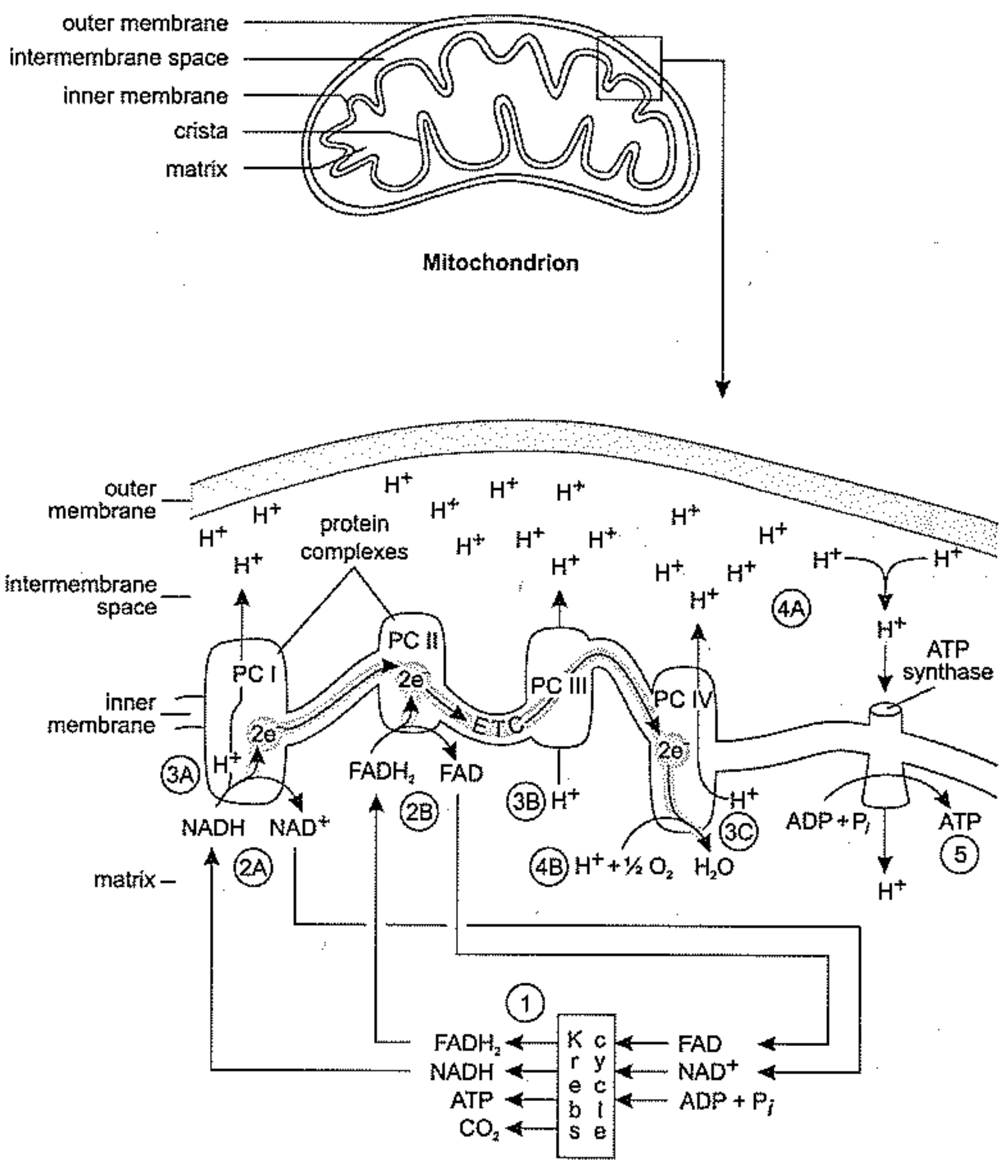
The two major processes of aerobic respiration, the Krebs cycle and oxidative phosphorylation, occur in mitochondria. There are four distinct areas of a mitochondrion, as follows (Figure 4-4):

1. **Outer membrane.** This membrane, like the plasma membrane, consists of a double layer of phospholipids.
2. **Intermembrane space.** This is the narrow area between the inner and outer membranes. H^+ ions (protons) accumulate here.

3. **Inner membrane.** This second membrane, also a double phospholipid bilayer, has convolutions called **cristae** (singular, **crista**). Oxidative phosphorylation occurs here. Within the membrane and its cristae, the electron transport chain, consisting of a series of protein complexes, removes electrons from NADH and $FADH_2$ and transports H^+ ions from the matrix to the intermembrane space. Some of these protein complexes are indicated in Figure 4-4 (PC I, PC II, PC III, and PC IV). Another protein complex, **ATP synthase,** is responsible for the phosphorylation of ADP to form ATP.
4. **Matrix.** The matrix is the fluid material that fills the area inside the inner membrane. The Krebs cycle and the conversion of pyruvate to acetyl CoA occur here.

Chemiosmosis in Mitochondria

Chemiosmosis is the mechanism of ATP generation that occurs when energy is stored in the form of a *proton concentration gradient* across a membrane. A description of the process during oxidative phosphorylation in mitochondria follows (Figure 4-4):



Chemiosmosis in Mitochondria

Figure 4-4

1. **The Krebs cycle produces NADH and $FADH_2$ in the matrix.** In addition, CO_2 is generated and substrate-level phosphorylation occurs to produce ATP.
2. **Electrons are removed from NADH and $FADH_2$.** Protein complexes in the inner membrane remove electrons from these two molecules (2A, 2B). The electrons move along the electron transport chain, from one protein complex to the next (shown as a shaded strip within the inner membrane).
3. **H^+ ions (protons) are transported from the matrix to the intermembrane compartment.** Protein complexes transport H^+ ions from the matrix, across the inner membrane, and to the intermembrane space (3A, 3B, 3C).
4. **A pH and electrical gradient across the inner membrane is created.** As H^+ are transferred from the matrix to the intermembrane space, the concentration of H^+ *increases* (pH decreases) in the intermembrane space (4A) and *decreases* in the matrix (pH increases). The concentration of H^+ in the matrix decreases further as electrons at the end of the electron transport chain (PC IV) combine with H^+ and oxygen to form water (4B). The result is a proton gradient (equivalent to a pH gradient) and an electric charge (or voltage) gradient. These gradients are potential energy reserves in the same manner as water behind a dam is stored energy.
5. **ATP synthase generates ATP.** ATP synthase, a channel protein in the inner membrane, provides a pathway for the protons in the intermembrane compartment to flow back into the matrix. As the protons are drawn though the channel by the voltage and pH gradients, the protons lose energy to the ATP synthase. ATP synthase uses the energy to generate ATP from ADP and P_i. It is similar to how a dam generates electricity when water passing through turbines forces them to turn.

How Many ATP?

How many ATP can *theoretically* be made from the energy released from the breakdown of 1 glucose molecule? In the cytoplasm, 1 glucose produces 2 NADH, 2 ATP, and 2 pyruvate during glycolysis. When the 2 pyruvate (from 1 glucose) are converted to 2 acetyl CoA, 2 more NADH are produced. From 2 acetyl CoA, the Krebs cycle produces 6 NADH, 2 $FADH_2$, and 2 ATP. If each NADH produces 3 ATP during oxidative phosphorylation, and $FADH_2$ produces 2 ATP, the total ATP count from 1 original glucose molecule appears to be 38. However, this number is reduced to 36 because the 2 NADH that are produced in the cytoplasm during glycolysis must be transported into the mitochondria for oxidative phosphorylation. The transport of NADH across the mitochondrial membrane reduces the net yield of each NADH to only 2 ATP (Table 4-1). However, the total yield from 1 glucose molecule *actually* hovers around 30 ATP due to variations in mitochondria efficiencies and competing biochemical processes.

Table 4-1

Process	Location of Process	Type of Phosphorylation	$FADH_2$ Produced	NADH Produced		ATP Yield
glycolysis	cytosol	substrate-level				2 ATP
glycolysis	cytosol	oxidative		2 NADH	=	4 ATP
pyruvate to acetyl CoA	mitochondrial matrix	oxidative		2 NADH	=	6 ATP
Krebs cycle	mitochondrial matrix	substrate-level				2 ATP
oxidative phosphorylation	mitochondrial inner membrane	oxidative		6 NADH	=	18 ATP
oxidative phosphorylation	mitochondrial inner membrane	oxidative	2 $FADH_2$		=	4 ATP
Total						**36 ATP**

A balance sheet accounting for ATP production from glucose by aerobic respiration. Total ATP production is theoretically 36 ATP for each glucose processed.

Anaerobic Respiration

What if oxygen is not present? If oxygen is not present, no electron acceptor exists to accept the electrons at the end of the electron transport chain. If this occurs, then NADH accumulates. After all the NAD^+ have been converted to NADH, the Krebs cycle and glycolysis both stop (both need NAD^+ to accept electrons). When this happens, no new ATP is produced, and the cell may soon die.

Anaerobic respiration is a method cells use to escape this fate. Although the two common metabolic pathways, **alcohol** and **lactic acid fermentation,** are slightly different, the objective of both processes is to replenish NAD^+ so that glycolysis can proceed once again. Anaerobic respiration occurs in the cytosol alongside glycolysis.

Alcohol Fermentation

Alcohol fermentation (or sometimes, just **fermentation**) occurs in plants, fungi (such as yeasts), and bacteria. The steps, illustrated in Figure 4-3, are as follows:

1. **Pyruvate to acetaldehyde.** For each pyruvate, 1 CO_2 and 1 acetaldehyde are produced. The CO_2 formed is the source of carbonation in fermented drinks like beer and champagne.
2. **Acetaldehyde to ethanol.** The important part of this step is that the energy in NADH is used to drive this reaction, releasing NAD^+. For each acetaldehyde, 1 ethanol is made and 1 NAD^+ is produced. The ethanol (ethyl alcohol) produced here is the source of alcohol in beer and wine.

It is important that you recognize the objective of this pathway. At first glance, you should wonder why the energy in an energy-rich molecule like NADH is removed and put into the formation of ethanol, essentially a waste product that eventually kills the yeast (and other organisms) that produce it. The goal of this pathway, however, does not really concern ethanol, but the task of freeing NAD^+ to allow glycolysis to continue. Recall that in the absence of O_2, all the NAD is bottled up in NADH. This is because oxidative phosphorylation cannot accept the electrons of NADH without oxygen. The purpose of the fermentation pathway, then, is to release some NAD^+ for use by glycolysis. The reward for this effort is 2 ATP from glycolysis for each 2 converted pyruvate. This is not much, but it's better than the alternative—0 ATP and imminent cellular death.

Lactic Acid Fermentation

Only one step occurs in lactic acid fermentation (Figure 4-3). A pyruvate is converted to lactate (or lactic acid); in the process, NADH gives up its electrons to form NAD^+. As in alcohol fermentation, the NAD^+ can now be used for glycolysis. In humans and other mammals, most lactate is transported to the liver, where it is converted back to glucose when surplus ATP is available.

Review Questions

Multiple-Choice Questions

The questions that follow provide a review of the material presented in this chapter. Use them to evaluate how well you understand the terms, concepts, and processes presented. Actual AP multiple-choice questions are often more general, covering a broad range of concepts, and often more lengthy. For multiple-choice questions typical of the exam, take the two practice exams in this book.

Directions: Each of the following questions or statements is followed by four possible answers or sentence completions. Choose the one best answer or sentence completion.

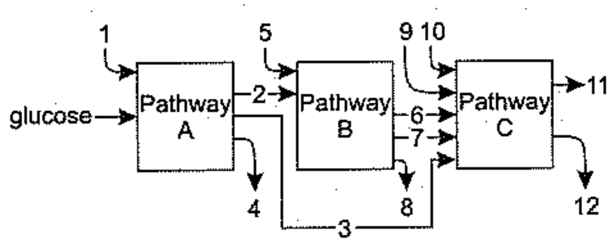
1. What is the value of the alcohol fermentation pathway?

- **A.** It produces ATP.
- **B.** It produces lactate (or lactic acid).
- **C.** It produces ADP for the electron transport chain.
- **D.** It replenishes NAD^+ so that glycolysis can produce ATP.

2. What is the purpose of oxygen in aerobic respiration?

- **A.** Oxygen accepts electrons at the end of an electron transport chain.
- **B.** Oxygen is necessary to carry away the waste CO_2.
- **C.** Oxygen is used in the formation of sugar molecules.
- **D.** The oxygen molecule becomes part of the ATP molecule.

Questions 3–7 refer to the following diagram. The three boxes represent the three major biosynthetic pathways in aerobic respiration. Arrows represent net reactants or products.



3. Arrow 2 is

- **A.** O_2
- **B.** ATP
- **C.** $FADH_2$
- **D.** pyruvate

4. Arrows 4, 8, and 12 could all be

- **A.** NADH
- **B.** ATP
- **C.** H_2O
- **D.** $FADH_2$

5. Arrows 3 and 7 could both be

A. NADH
B. ATP
C. H_2O
D. $FADH_2$

6. Arrow 9 could be

A. O_2
B. ATP
C. H_2O
D. FAD

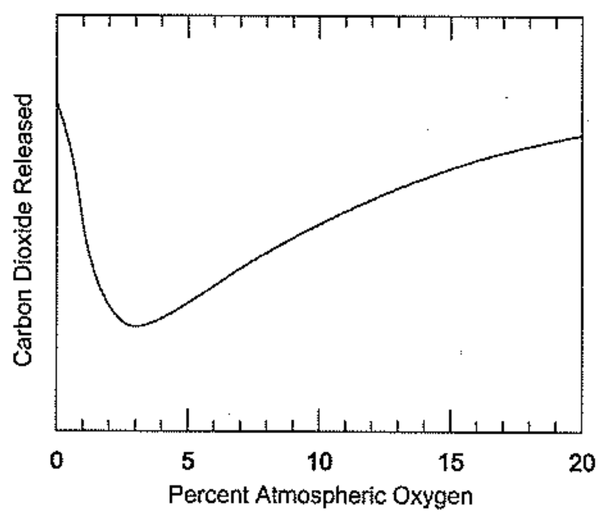
7. Pathway B is

A. oxidative phosphorylation
B. photophosphorylation
C. the Krebs cycle
D. glycolysis

8. Which of the following sequences correctly indicates the potential ATP yield of the indicated molecules from greatest ATP yield to least ATP yield?

A. pyruvate, ethanol, glucose, acetyl CoA
B. glucose, pyruvate, acetyl CoA, NADH
C. glucose, pyruvate, NADH, acetyl CoA
D. glucose, $FADH_2$, NADH, pyruvate

Questions 9–10 refer to the following graph that shows the amount of CO_2 that is released by plant cells at various levels of atmospheric oxygen.



9. At levels of atmospheric O_2 below 1%, the amount of CO_2 released is relatively high. This is probably because

A. The Krebs cycle is very active.
B. O_2 is being converted to H_2O.
C. Alcohol fermentation is occurring.
D. There are insufficient amounts of coenzyme A.

10. As levels of atmospheric O_2 increase beyond 5%, the amounts of CO_2 released increase. This is probably a direct result of

- **A.** an increase in glycolytic activity
- **B.** a greater availability of appropriate enzymes
- **C.** an increase in Krebs cycle activity
- **D.** an increase in atmospheric temperature

11. All of the following processes produce ATP EXCEPT:

- **A.** glycolysis
- **B.** the Krebs cycle
- **C.** lactic acid fermentation
- **D.** oxidative phosphorylation of NADH

12. Chemiosmosis describes how ATP is generated from ADP. All of the following statements conform to the process as it occurs in mitochondria EXCEPT:

- **A.** H^+ accumulates in the area between the membrane of the cristae and the outer membrane of the mitochondrion.
- **B.** A voltage gradient is created across the cristae membranes.
- **C.** A proton gradient is created across the cristae membranes.
- **D.** Electrons flowing through the ATP synthase channel protein provide the energy to phosphorylate ADP to ATP.

13. After strenuous exercise, a muscle cell would contain increased amounts of all of the following EXCEPT:

- **A.** ADP
- **B.** CO_2
- **C.** lactate (or lactic acid)
- **D.** glucose

14. All of the following statements about cellular respiration are true EXCEPT:

- **A.** Some of the products from the breakdown of proteins and lipids enter the Krebs cycle.
- **B.** If oxygen is present, water is produced.
- **C.** The purpose of oxygen in aerobic respiration is to donate the electrons that transform $NAD^+ + H^+$ to NADH.
- **D.** Lactate or ethanol is produced when oxygen is unavailable.

15. All of the following processes release CO_2 EXCEPT:

- **A.** the Krebs cycle
- **B.** alcohol fermentation
- **C.** oxidative phosphorylation
- **D.** the conversion of pyruvate to ethanol

Free-Response Questions

The AP exam has long and short free-response questions. The long questions have considerable descriptive information that may include tables, graphs, or figures. The short questions are brief but may also include figures. Both kinds of questions have four parts and generally require that you bring together concepts from multiple areas of biology.

The questions that follow are designed to further your understanding of the concepts presented in this chapter. Unlike the free-response questions on the exam, they are narrowly focused on the material in this chapter. For free-response questions typical of the exam, take the two practice exams in this book.

Directions: The best way to prepare for the AP exam is to write out your answers as if you were taking the exam. Use complete sentences and do *not* use outline form or bullets. You may use diagrams to supplement your answers, but be sure to describe the importance or relevance of your diagrams.

1. In the process of alcohol fermentation, 2 NADH molecules are converted to 2 NAD^+ as energy from the NADH is used to drive the formation of ethanol. Explain why there is a need to add energy to a process whose purpose is to extract energy from glucose.

2. The mitochondrion has two phospholipid-bilayer membranes: an outer membrane and an inner membrane. Explain why two membranes are necessary.

3. Describe the Krebs cycle and oxidative phosphorylation. Specifically address the following:
   a. ATP and coenzyme production
   b. the locations where these biosynthetic pathways occur
   c. chemiosmotic theory

4. Describe, at the molecular level, how aerobic respiration extracts energy from each of the following:
   a. starches and other carbohydrates
   b. proteins
   c. lipids

5. a. Explain, at the molecular level, why many organisms need oxygen to maintain life.
   b. Explain, at the molecular level, how some organisms can sustain life in the absence of oxygen.

Answers and Explanations

Multiple-Choice Questions

1. **D.** In the absence of oxygen, all of the NAD^+ gets converted to NADH. With no NAD^+ to accept electrons from the glycolytic steps, glycolysis stops. By replenishing NAD^+, alcohol fermentation allows glycolysis to continue.

2. **A.** At the end of the electron transport chain in oxidative phosphorylation, ½ O_2 combines with 2 electrons and 2 H^+ to form water.

3. **D.** You should review aerobic respiration by identifying each arrow: Pathway A is glycolysis, Pathway B is the Krebs cycle, and Pathway C is oxidative phosphorylation. Arrow 1: ADP or NAD^+; arrow 2: pyruvate; arrow 3: NADH; arrow 4: ATP; arrow 5: ADP, NAD^+, or FAD; arrows 6 and 7: $FADH_2$ and NADH (either one can be 6 or 7); arrow 8: ATP or CO_2; arrows 9 and 10: O_2 and ADP (either one can be 9 or 10); and arrows 11 and 12: H_2O and ATP (either one can be 11 or 12).

4. B. ATP is produced in the glycolytic pathway (glycolysis), the Krebs cycle, and by oxidative phosphorylation.

5. A. Arrow 3 represents the NADH produced in glycolysis (Pathway A) and is used in oxidative phosphorylation (Pathway C). In addition, NADH could also be represented by arrow 7, a product of the Krebs cycle (Pathway B). Arrow 7 could also represent $FADH_2$, but $FADH_2$ cannot be represented by arrow 3. Thus, only NADH can be represented by both arrows 3 and 7. If arrow 7 represents NADH, then arrow 6 represents $FADH_2$.

6. A. Arrow 9 could represent the O_2 that accepts the electrons after they pass through the electron transport chain in oxidative phosphorylation. Arrow 9 could also be ADP, but ADP is not among the answer choices.

7. C. Pathway B represents the Krebs cycle. The Krebs cycle uses the energy in pyruvate (arrow 2) to generate $FADH_2$ and NADH (arrows 6 and 7).

8. B. Each of these molecules has the potential to produce the following amounts of ATP: glucose, 36 ATP; pyruvate, 15 ATP; acetyl CoA, 12 ATP; NADH, 3 ATP (or 2 ATP if they originate in glycolysis); and $FADH_2$, 2 ATP. The metabolic pathway that breaks down ethanol to H_2O and CO_2 in the human liver is variable. However, answer choice A can be eliminated without knowing how many ATP molecules ethanol can yield because glucose produces more ATP than does pyruvate.

9. C. When O_2 is absent (or very low), anaerobic respiration (alcohol fermentation) is initiated. Alcohol fermentation releases CO_2. Photosynthesis, which would consume CO_2 to produce glucose, is obviously not occurring. This indicates that the plant activity illustrated by the graph is occurring at night (or during a heavily clouded day).

10. C. CO_2 is produced in the Krebs cycle. As in the previous question, the production of CO_2, rather than its consumption, indicates that photosynthesis is not occurring and that the plant activity is taking place at night.

11. C. Lactic acid fermentation, the conversion of pyruvate to lactate, removes electrons from NADH to make NAD^+. No ATP is generated by this step.

12. D. Protons, not electrons, pass through ATP synthase as they move down the proton gradient. An electrical gradient or voltage is produced by the greater number of positive charges (from the protons) in the intermembrane space relative to the number of positive charges inside the crista membrane.

13. D. During strenuous exercise, glucose is broken down to pyruvate. Aerobic respiration produces CO_2. Anaerobic respiration, which would occur during strenuous exercise, would increase lactate formation. Exercise would also consume ATP, producing ADP and P_i.

14. C. The purpose of O_2 is to *accept* the electrons at the end of the electron transport chain in oxidative phosphorylation. The electrons combine with O_2 and H to form water. Products from the breakdown of lipids and proteins are converted to pyruvate, acetyl CoA, or intermediate carbon compounds used in the Krebs cycle.

15. C. Oxidative phosphorylation describes the transfer of electrons from NADH and $FADH_2$ to electron acceptors that pump H^+ across the inner mitochondrial membrane. Oxygen is required as the final electron acceptor of these electrons. However, no CO_2 is involved. In contrast, all the remaining answer choices describe processes that release CO_2. Note that answer choices B and D describe the same process.

Free-Response Questions

1. In the absence of O_2, oxidative phosphorylation and ATP generation cannot proceed and NADH accumulates. As a result, there is no NAD^+ available for glycolysis. Fermentation regenerates NAD^+ from NADH, and this NAD^+ can be used in glycolysis to generate ATP.

2. The two membranes create an intermembrane space between them, where H^+ (protons), transported from the matrix, accumulate. This creates a proton gradient, which drives the movement of protons through ATP synthase back into the matrix. As protons pass through the ATP synthase channel, ATP is generated from ADP and P_i.

3. **a.** The Krebs cycle and oxidative phosphorylation are the oxygen-requiring processes involved in obtaining ATP from pyruvate. Pyruvate is derived from glucose through glycolysis, a process that does not require oxygen.

Before pyruvate enters the Krebs cycle, it combines with coenzyme A. During this initial reaction with coenzyme A, 2 electrons and 2 H^+ removed from pyruvate combine with NAD^+ to form 1 NADH + H^+. NADH is a coenzyme storing enough energy to generate 3 ATP in oxidative phosphorylation. A CO_2 molecule is also released. The end product of this reaction is acetyl CoA. To begin the Krebs cycle, acetyl CoA combines with oxaloacetate (OAA) to form citrate, releasing the coenzyme A component. A series of reactions then occurs that generates 3 molecules of the coenzyme NADH (from NAD^+), 1 molecule of the coenzyme $FADH_2$ (from FAD), and 1 ATP (from ADP + P_i) for each molecule of acetyl CoA that enters the Krebs cycle. The last product in the series of reactions, OAA, is the substance that reacts with acetyl CoA; thus, the Krebs reactions sustain a cycle.

Energy from the coenzymes NADH and $FADH_2$ is extracted to make ATP in oxidative phosphorylation. For each of these coenzymes, 2 electrons pass through an electron transport chain, passing through a series of protein carriers (some are cytochromes, such as cytochrome c). During this passage, 3 ATP are generated for each NADH originating in the Krebs cycle. $FADH_2$ generates 2 ATP. At the end of the electron transport chain, O_2 accepts the electrons (and 2 H^+) to form water. NAD^+ and FAD can be used again to receive electrons in the Krebs cycle. The total number of ATP generated from a single pyruvate is 15 ATP.

b. The Krebs cycle and oxidative phosphorylation occur in the mitochondria. The Krebs cycle occurs in the matrix of the mitochondria. The protein carriers for the electron transport chain are embedded in the inner mitochondrial membranes, called the cristae. Thus, oxidative phosphorylation occurs in these cristae membranes.

c. Chemiosmosis describes how ATP is generated from ADP + P_i. During oxidative phosphorylation, H^+ (protons) are deposited on the outside of the cristae, between the cristae and the outer membrane. The excess number of protons in this intermembrane space creates a pH and electric gradient. The gradient provides the energy to generate ATP as protons pass back into the matrix through ATP synthase, a channel protein in the cristae.

Note that the answer to each part of the question is labeled a, b, or c. Answering each part separately helps you to organize your answer and helps the grader recognize that you addressed each part of the question.

4. *This question is similar to question 1. One major difference is that you need to describe glycolysis in detail. You also need to correctly connect glycolysis with the Krebs cycle and ATP production because glycolysis produces 2 pyruvates. The explanation in part 1 is for the production of only 1 pyruvate. The second major difference is that you need to include a discussion of how starches, proteins, and lipids are tied to respiration. That part of the answer follows.*

a. Starches are polymers of glucose. Various enzymes break down starches and other carbohydrates to glucose. Disaccharides like sucrose are catalyzed to glucose and fructose. Once these carbohydrates are broken down to glucose, the glucose enters the glycolytic pathway. Fructose undergoes some intermediate steps and enters the glycolytic pathway after a couple of steps. Glycolysis then breaks down glucose, obtaining ATP and pyruvate. Pyruvate then enters the Krebs cycle, yielding more ATP, NADH, and $FADH_2$. NADH and $FADH_2$ then generate ATP during oxidative phosphorylation.

b. Lipids are hydrolyzed to glycerol and fatty acids. Both of these components undergo enzymatic reactions that eventually produce acetyl CoA. Acetyl CoA, in turn, begins the Krebs cycle, which generates NADH, $FADH_2$, and ATP.

c. Proteins are hydrolyzed to amino acids. Each of the various amino acids produces different products when broken down. Some of these products are converted to acetyl CoA; others are converted to OAA or other Krebs cycle intermediates. NH_3 is a toxic waste product from amino acid breakdown and is exported from the cell.

5. a. This question is the same as question 4, except there is an additional focus on the function of O_2. For this question, then, it is especially important that you state that the purpose of O_2 is to accept electrons at the end of the electron transport chain in oxidative phosphorylation. Then describe the consequences if oxygen is not present—no ATP, no oxidative phosphorylation, and no Krebs cycle.

   b. This question requires that you describe lactic acid fermentation and alcohol fermentation, specifically indicating that the function of these two processes is to regenerate NAD^+ so that glycolysis can continue and produce 2 ATP for each glucose.