



CHAPTER

2

Biological reactions in microbial cells

CHAPTER FOCUS

- ❖ Why is an understanding of biological reactions important for the health professional?
- ❖ How does the structure of a biological compound relate to its function?
- ❖ What is the role of enzymes in biological processes?
- ❖ What are the principal pathways of energy production in microorganisms?
- ❖ How are different classes of biological molecules interconverted?
- ❖ What metabolic processes carried out by microorganisms are of use to humans?

INTRODUCTION

All living cells, from the smallest microorganisms to the most complex animal cells, undergo continual processes of breakdown, synthesis, replication and repair. **Metabolism** is the overall term used to describe these processes and it refers to all the different reactions that must occur in a cell in order for it to grow and reproduce. The pathways of metabolism are remarkably similar in all living cells. The differences that do occur usually reflect the availability of nutrients or the need for a cell to carry out a specialised function—which may be secretion, storage, structural support, energy for movement, or reproduction. Much of our knowledge of the processes that occur in human cells has been derived from research on the major metabolic pathways that occur in microbial cells.

Catabolism is the process of breakdown of complex molecules, usually with the release of energy. **Anabolism** refers to the synthesis of new or replacement molecules. This is an energy-requiring process. A **metabolic pathway** is a series of reactions in the process of metabolism.

Although metabolic processes are complex, consisting of thousands of chemical reactions, an understanding of the basic concepts of metabolism does not require an extensive knowledge of chemistry. The reactions are logical and can be understood in a simplified descriptive form. Many students are unnecessarily deterred from attempting to study the biochemistry of cells, as they do

not see its relevance to their work as health professionals. However, a knowledge of the processes that occur in all living cells will enable students to understand the chemistry underlying many of the phenomena they observe every day. For example:

- how pathogens invade, replicate and cause disease in a host
- why adequate nutrients are needed to provide energy for cellular activity, growth and repair
- why some microorganisms need a particular environment to survive
- how cells replicate and repair themselves
- the nature of the immune response
- the therapeutic use of drugs
- why some infections can be treated with antibiotics and others cannot
- how microorganisms are identified by biochemical tests
- how microbial reactions are used to produce compounds of use to humans.

This chapter assumes a basic knowledge of atoms and molecules, of ionic and covalent bonds, and some carbon chemistry. The metabolic pathways are described using diagrams, words and simple chemical formulae. Students can therefore study the reactions at a level suited to their chemistry background.

STRUCTURE OF BIOLOGICAL MOLECULES

All living matter is made up of a number of complex molecules containing the elements carbon, hydrogen, oxygen, nitrogen, phosphorus and sulfur, with lesser (trace) amounts of other elements. The way in which the atoms of these elements are arranged determines the structure and unique function of each molecule. The most important element is carbon, a small atom with four electrons in its outer shell, capable of forming four covalent bonds. Compounds containing carbon are called **organic compounds**. When carbon combines with other atoms it forms a molecule with a particular shape (**stereospecificity**) (see Figure 2.1). The configuration of atoms in the molecules of carbon-containing compounds gives rise to the enormous diversity we observe in biological molecules; it is also responsible for the specific shape and function of these molecules.

Functional groups

A number of specific structures are found joined to carbon in various organic compounds and are essential for the activity of these compounds. They are called 'active', or 'functional', groups. They include:

- Carboxylic acid—COOH in fatty acids (lipids), acetic acid (vinegar), amino acids (proteins)
- Amino—NH₂ in amino acids (proteins)

- Hydroxyl—OH in alcohols and glycerol (carbohydrates)
- Sulfhydryl—SH in proteins
- Organic phosphate—R-CH₂-OPO₄ in phospholipids and nucleic acids
- Ester linkage—RC(=O)OR in triglycerides.

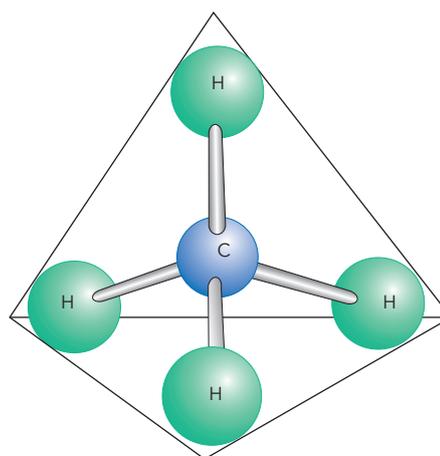


FIGURE 2.1

Structure of carbon-containing compounds

A carbon atom with four single bonds has this shape because of the direction of the bonds. The tetrahedral molecule of methane, CH₄, shown here, contains carbon bonded to four hydrogen atoms.

Another property conferred on biological molecules containing carbon is **stereoisomerism**. Because the carbon atom is linked to four other atoms, it often forms compounds which, although they have the same formula (composition), do not have the same structural configuration; they are mirror images and cannot be superimposed on each other (see Figure 2.2). This can be likened to a pair of gloves, where the right-hand glove cannot be substituted for the left because it has a different configuration.

Specificity

One of the most fascinating properties of biological molecules is their *specificity*—that is, the ability of a particular structure in a molecule to recognise and fit together with a complementary structure in another molecule. You may have experienced the frustration of having different pieces of electrical equipment that do not fit together—for example, having the wrong lead for a machine, or a plug and socket that do not ‘match’. A similar situation occurs in living cells. Biological molecules contain particular chemical groups in unique structures that confer certain properties on the molecules. The way in which these molecules interact forms the basis for the regulation of all life processes. If the structures of two molecules are compatible, then those molecules can combine and interact in the same way that an electric plug will fit into a compatible socket (see Figure 2.3). To take this analogy further, sometimes an ‘adaptor’ is needed to join two

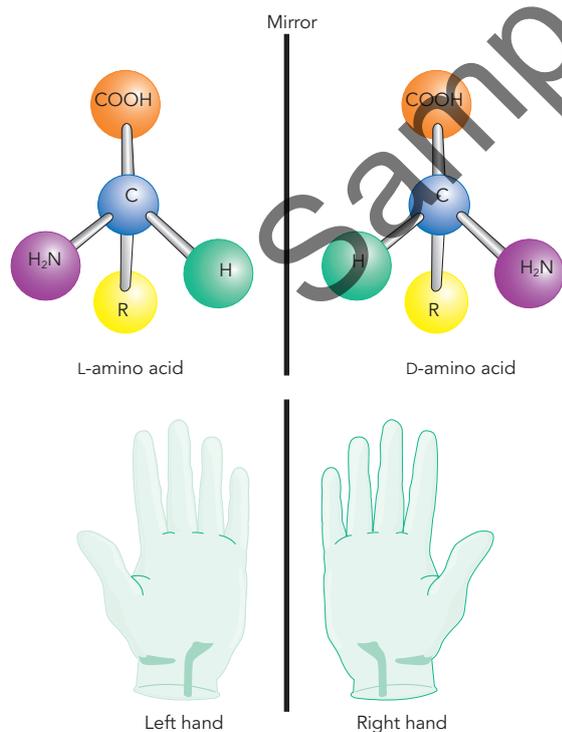


FIGURE 2.2

Stereoisomerism

Models of the L- and D- forms of an amino acid are mirror images and cannot be superimposed, just like the right- and left-hand gloves illustrated.

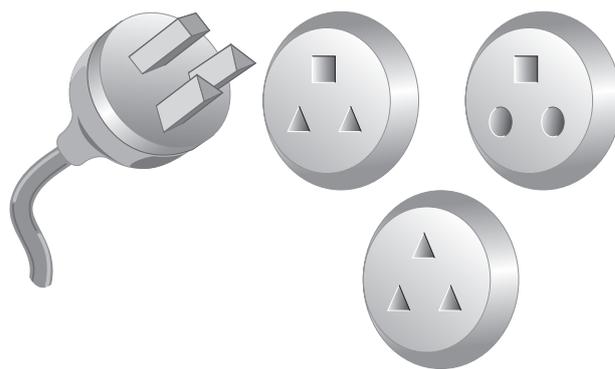


FIGURE 2.3

Specificity

The need for biological molecules to be the right shape in order to interact with each other is illustrated by this electrical plug and three different sockets.

pieces of equipment together. In biochemical terms, this adaptor molecule is called a **cofactor** or **coenzyme**.

The specific interaction between two compatible biological molecules determines which reactions can take place. It explains the unique mechanism of many important processes such as DNA replication, enzyme reactions, antigen–antibody interactions and the specificity of viral attack.

ENZYMES AND CHEMICAL REACTIONS

Metabolic processes involve thousands of chemical reactions. But what makes these reactions occur? A **catalyst** is defined as a substance that speeds up a chemical reaction by lowering the activation energy required for the reaction to take place. It takes part in the reaction but is itself unchanged.

What is **activation energy**? The simplest definition is that it is the amount of energy required to make a reaction occur. For example, if two substances such as hydrogen and oxygen are mixed, a reaction will not occur at a measurable rate. However, if some energy is applied (in the form of a spark or heat), the reaction occurs very rapidly. The necessary input of energy is called the activation energy. In industry, the amount of energy in the form of heat that needs to be added is often very great, but the addition of a catalyst (e.g. a metal) will allow the reaction to occur with a much lower energy input. In other words, *the catalyst lowers the activation energy*.

In biological systems the addition of large amounts of heat would destroy the cellular proteins, so an alternative way of providing the necessary energy has to be found. Living cells use **enzymes** which act as biological catalysts and lower the amount of energy (activation energy) required to get a reaction started. This is why enzymes are sometimes described as ‘speeding up a chemical reaction’ (see Figure 2.4).

Enzymes enable reactions to occur that might not otherwise happen; they give the reaction a ‘push’ so that it acquires a momentum of its own. They do this by providing a surface or site on which the reaction can take place, thereby allowing the reactant molecules to be held in close proximity to each other and increasing the efficiency of

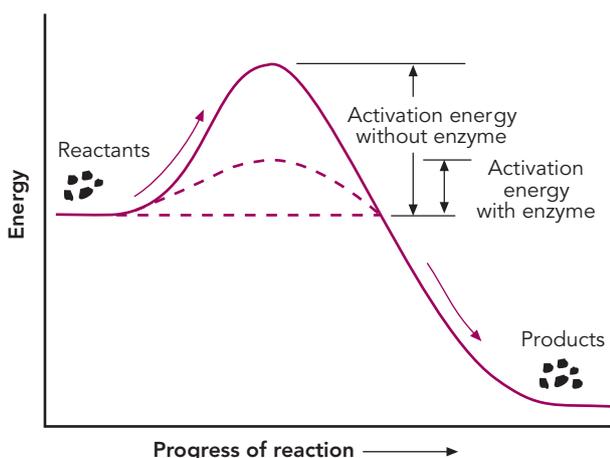


FIGURE 2.4

Activation energy

A chemical reaction cannot take place unless a certain amount of activation energy is available to start it. Enzymes lower the amount of activation energy needed to initiate a reaction. They thus make it possible for biologically important reactions to occur at the relatively low temperatures that living organisms can tolerate.

their interaction. All this is done at a temperature compatible with the normal activities of the cell.

Structure of enzymes

Enzymes are proteins produced in cells in response to the metabolic requirements of the cell. The genetic information needed for the synthesis of each particular enzyme is coded for in the DNA of the cell, together with information that enables the cell to synthesise all its other protein requirements (see Chapter 4). Each enzyme is able to act on a particular type of chemical compound, its **substrate**. The name of the enzyme is usually derived from the substrate it uses and the type of reaction that occurs. Most enzyme names end in *-ase*. For example, lactic dehydrogenase is an enzyme that removes hydrogen from its substrate, lactic acid.

Enzymes are proteins and are therefore made up of chains of amino acids. Each enzyme has its own unique sequence of amino acids and is folded in a certain way to give it a specific shape. Within this shape there is a particular location or area on the molecule called the **active site**. This site is rather like an electrical socket. It allows the attachment of a correspondingly shaped molecule or substrate. Once attached, this substrate can be modified or split; that is, it undergoes a metabolic reaction. It is then released and another substrate molecule takes its place and the reaction continues (see Figure 2.5).

There are hundreds of different enzymes in each cell. Every reaction that occurs in a living cell is controlled by a specific enzyme. Each enzyme has an active site specific for the shape of a particular substrate. Some enzymes contain both a protein and a non-protein component. The non-protein part, called a **cofactor** or **prosthetic group**, is necessary for enzyme activity. If the cofactor is an organic molecule, it is called a **coenzyme**. These coenzymes fit

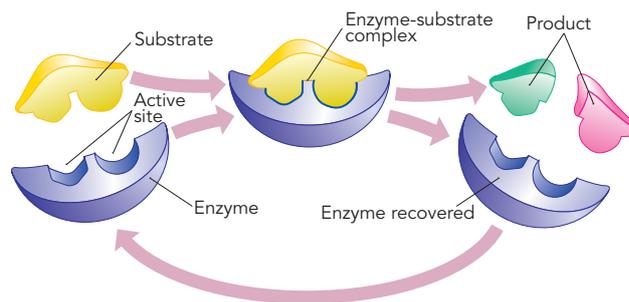


FIGURE 2.5

Enzyme-substrate complex

into the active site like an adaptor (see Figure 2.6). Some coenzymes can be synthesised by the cell. Others must be obtained from the food supply and are considered **essential nutrients**. Some enzymes require the presence of divalent metal ions as cofactors for activity, such as magnesium (Mg^{++}), calcium (Ca^{++}), manganese (Mn^{++}) and zinc (Zn^{++}).

Bacteria are usually able to synthesise all the coenzymes they require, whereas most animals need to receive them in their diet. They are called essential nutrients or **vitamins** (e.g. folic acid). These metabolic differences can be exploited when developing drugs to selectively inhibit the bacteria causing an infection without harming the host (human) cells. This topic is discussed further in Chapter 12.

Factors influencing enzyme activity

As mentioned before, enzymes usually react preferentially with a particular substrate. Sometimes, however, they may bind to a closely related compound, or **analogue**. When a compound with a structure similar to the substrate binds reversibly or irreversibly to the active site and prevents the real substrate reaching the enzyme, the activity of the

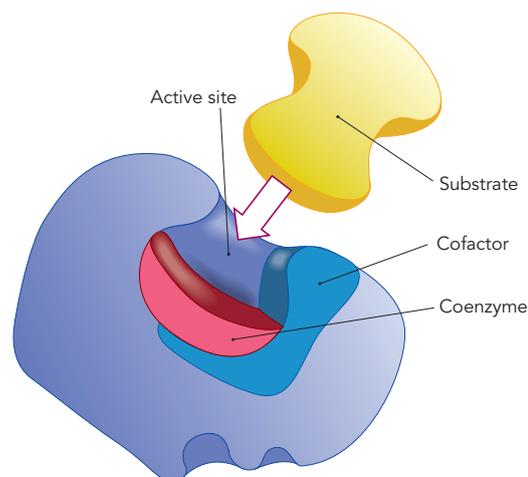


FIGURE 2.6

Components of an enzyme

Many enzymes require the protein portion of the enzyme as well as a cofactor (non-protein) portion for activity. Cofactors may be metal ions or organic molecules called coenzymes.

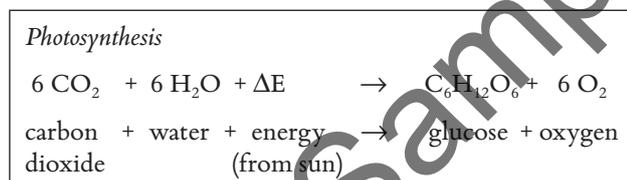
enzyme is inhibited, or an inactive product is formed. This property has been used to design nucleotide analogues, which are effective as antiviral drugs (see Chapter 12).

Enzymes have been found to function optimally at certain pH values and salt (ionic) concentrations. This is because a change in pH or ionic concentration may alter the electric charge on the protein molecule and therefore its shape. If the shape of the active site is altered, the substrate may not be able to attach. Temperature also affects the rate at which enzyme reactions take place. Bacteria therefore reproduce fastest at their optimum temperature.

It is important to remember all these factors when considering the metabolic reactions described below. Environmental factors can also exert some influence but, in general, microorganisms have shown themselves to be particularly adaptable to changes in their environment and can utilise a range of nutrients depending on what is available.

ENERGY PRODUCTION IN BIOLOGICAL SYSTEMS

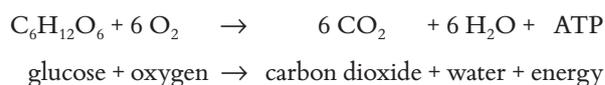
The basic metabolic process of life is the production of energy for use in other cellular processes. Initially, all energy comes from the sun, the radiant energy being trapped by the chlorophyll in the cells of green plants. In a process known as **photosynthesis**, the basic molecules of carbon dioxide (from the air) and water are converted to a simple carbohydrate molecule, glucose (containing six carbon atoms). In the process, oxygen is released. The removal of carbon dioxide from the atmosphere and the release of oxygen enables the survival of oxygen-requiring organisms such as humans, other animals and many microorganisms. This can be written as:



The energy from the sun that has been trapped and used to produce glucose is released when a molecule of glucose is broken down during metabolism. This complex process is called **oxidation**, or **respiration**, and consists of a number of interrelated biochemical pathways, including those of **glycolysis**, **fermentation** and **aerobic respiration**. Reactions that occur in the presence of oxygen are described as **aerobic**. Glycolysis and fermentation, which can occur in the absence of oxygen, are described as **anaerobic**. Many of these reactions require the involvement of various cofactors as well as enzymes, and result in the production of energy, which is trapped in a small molecule called **adenosine triphosphate (ATP)**. ATP is termed an 'energy-rich storage molecule' because the subsequent breakdown of ATP releases energy for use in other reactions in the cell.

In aerobic respiration (e.g. in muscle cells), glucose is completely broken down in a series of steps to form carbon dioxide and water, with the release of considerable amounts of energy. The reaction can be written as:

Respiration



It is obvious that this is the reverse process to photosynthesis.

There are many other reactions that take place, depending on the needs of the cell and the availability of oxygen. The reactions described in this chapter are of importance in microbial cells.

The energy released during the breakdown of the glucose molecule in living cells is used to form ATP. ATP is a small, water-soluble, biological energy-storage molecule which can diffuse around the cell and provide energy for other reactions in the cell. ATP consists of a molecule of the purine base, adenine, joined to the pentose (5-carbon) sugar, ribose, and then to three phosphate groups (see Figure 2.7). Each of these phosphate groups has a different 'bond energy'—that is, the amount of energy required to form or break the bond. The third phosphate bond, the 'triphosphate', is very labile; that is, it is readily broken with the release of a large amount of energy. It also requires a large amount of energy to form it. The triphosphate bond is therefore referred to as a 'high-energy bond'. The base-ribose unit is called a **nucleoside**, and the base-ribose-phosphate is a **nucleotide**. ATP is also one of the building blocks of the nucleic acids, RNA and DNA.

For a molecule of glucose to be broken down or metabolised, it must first acquire some energy, or be 'activated'. This occurs through the removal of one phosphate group

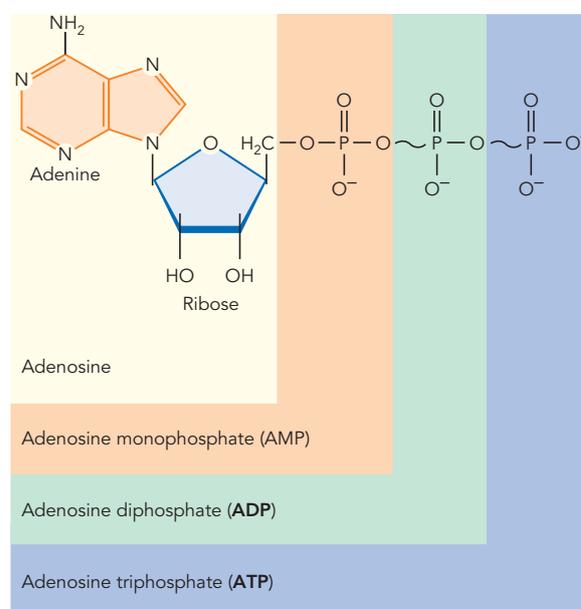


FIGURE 2.7

Structure of ATP

ATP is composed of the purine base, adenine, joined to the sugar, ribose and three phosphate groups. The addition of the third phosphate group to ADP requires a large amount of energy, which is released when the bond is broken.

from a molecule of ATP—that is, the conversion of ATP to ADP (adenosine diphosphate) and the transfer of the phosphate group to glucose:



The 'activated' glucose molecule (glucose-6-phosphate) is now ready to participate in other cellular reactions.

The processes that occur in living cells can be likened to those in a complex piece of machinery. Fuel (in the form of nutrients) is broken down (oxidised, metabolised) to provide energy to carry out the functions of the machine. Manufactured engines, however, waste a lot of energy as heat, whereas living cells are highly efficient and use sophisticated energy-capturing systems such as ATP and other complex molecules to conserve energy.

Energy requirements of microbial cells

Microbial cells require energy to carry out a range of activities necessary for their growth and reproduction. These activities include:

- the synthesis of lipids, carbohydrates, enzymes and other types of proteins
- the formation of the various structural components of the cell
- the repair and maintenance of the cellular environment
- the accumulation and storage of nutrients and the disposal of waste products
- the active transport of substances into and out of the cell
- the movement of flagella and cilia.

STRUCTURE OF BIOLOGICAL MOLECULES

We now consider in more detail the structure of the biological molecules that make up living cells—carbohydrates, lipids, proteins and nucleic acids—and briefly describe the metabolic pathways involved in their synthesis and breakdown. Detailed discussion of all these pathways is beyond the scope of this text. However, a brief description of the major classes of molecules is given here, together with the most common pathways of energy production and biosynthesis.

Structure of carbohydrates

Carbohydrates (sugars) are a group of compounds composed primarily of carbon, hydrogen and oxygen, and their breakdown is the major source of energy in cells. The most common sugar unit is glucose. Glucose is a **hexose**, a 6-carbon compound containing six hydroxyl (OH) groups. Because of the stereoisomerism of the carbon atom, these hydroxyl groups can be arranged in different ways, giving rise to different **isomers** of glucose. In the cell, glucose exists in a ring form with a three-dimensional shape (see Figure 2.8).

The most abundant sugars are the 6-carbon sugars, which include glucose and fructose. Another important group—the **pentoses**—contain five carbon atoms. The most important sugars in this group are **ribose** and **deoxyribose**, which occur in nucleic acids.

Monosaccharides, disaccharides and polysaccharides

Sugars are found in nature as single units (**monosaccharides**), double units (**disaccharides**) or polymers (**polysaccharides**). The monosaccharides include the hexoses and pentoses described above. Disaccharides and polysaccharides are made up of basic hexose units joined together. The way in which they are linked (i.e. the direction of the bond) is referred to as an α -linkage or a β -linkage and determines the shape of the molecule (see Figure 2.9).

The most common disaccharide is sucrose, which consists of one molecule of glucose and one of fructose joined together. There are a number of different polysaccharides which have quite different characteristics that are determined by their structure (linkage). Cellulose is a straight chain polymer and a major component of plant cells—providing fibrous structural support. Starch is a storage polysaccharide found in potatoes and other root vegetables. It provides an easily digested source of energy. Animal cells contain glycogen as their storage polysaccharide. Dextran is a polymer of the glucose isomer dextrose and is used clinically as a blood plasma substitute.

Complex polysaccharide compounds located on the outer walls of some cells are responsible for conferring antigenic properties on the cell.

Structure of lipids

Fats, or triglycerides, are lipids consisting of glycerol and long-chain fatty acids. **Glycerol** is a 3-carbon compound containing three hydroxyl (OH) groups. **Fatty acids** are long chains of carbon and hydrogen (usually 16 or 18 carbon atoms in length) with a single carboxyl (COOH) group at one end. Triglycerides consist of a glycerol molecule combined with three long-chain fatty acids joined by ester linkages (see Figure 2.10).

Another important group of lipid molecules is the **phospholipids**. These consist of glycerol esterified with two long-chain fatty acids. The third position is occupied by a phosphate group, which can also be joined to another organic molecule such as choline. Phospholipids are an integral component of all cell membranes (see Chapter 3).

In phospholipids, one fatty acid chain is replaced by a phosphate group linked to an organic compound such as choline.

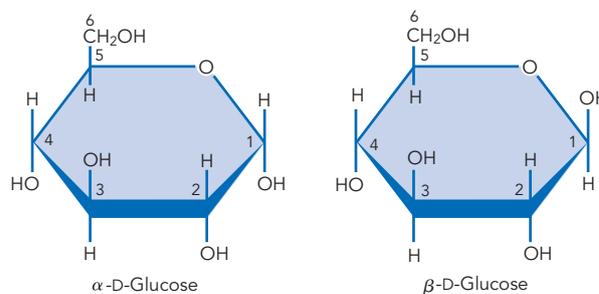
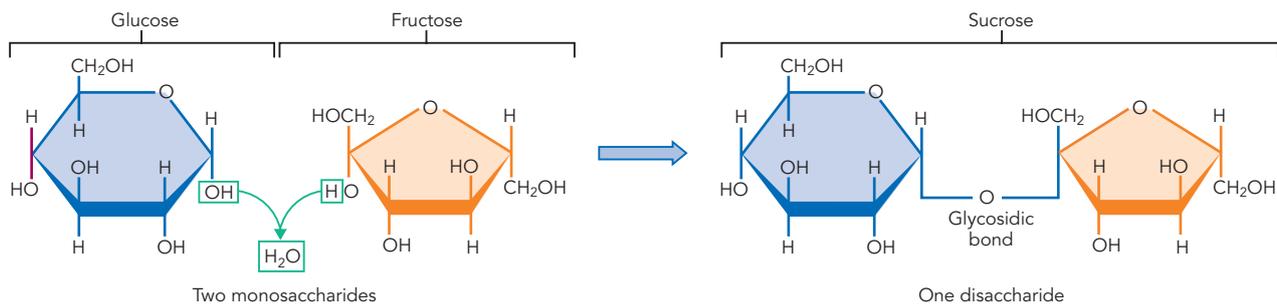


FIGURE 2.8

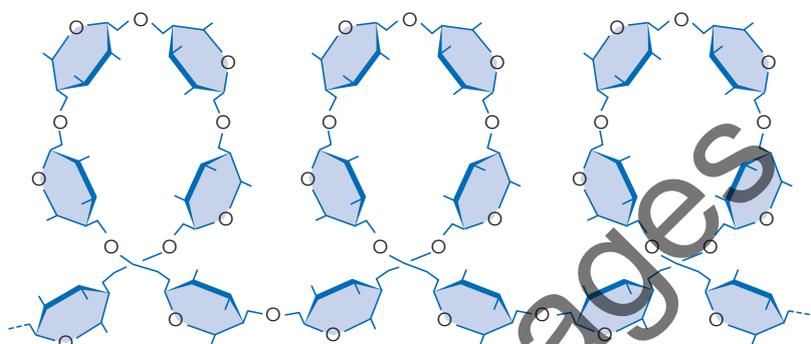
Ring structure of glucose

Sugars exist in biological molecules in a three-dimensional ring form.

(a) Formation of sucrose



(b) Structure of starch



(c) Structure of cellulose

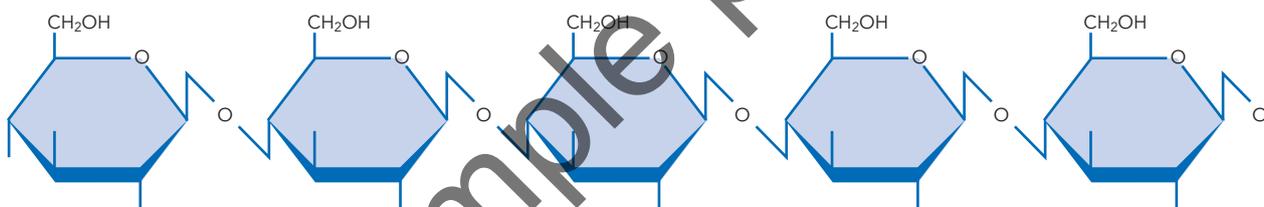


FIGURE 2.9

Structure of carbohydrates

(a) Two monosaccharides are joined to form a disaccharide by the removal of water and the formation of a glycosidic bond; (b) polysaccharides such as starch are formed by the linking of many monosaccharides in long chains; (c) structure of cellulose, β 1–4 linkage, gives rise to a straight-chain polymer which cannot be broken down by human enzymes.

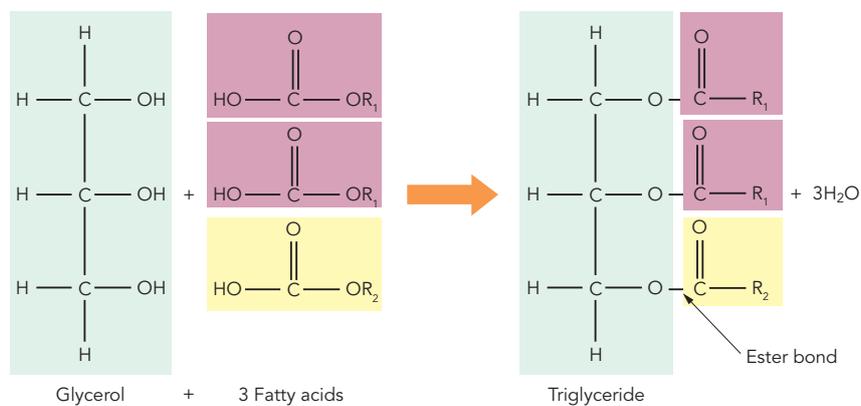


FIGURE 2.10

Structure of a triglyceride

Three long-chain fatty acids are combined with (esterified) a molecule of glycerol. The chain length and saturation of the fatty acids determine the properties of the lipid.

Structure of proteins

Proteins consist of chains of amino acids arranged in a specific sequence. There are 20 different naturally occurring amino acids that serve as the building blocks of proteins. Amino acids are a group of organic molecules that all contain an amino ($-\text{NH}_2$) group and a carboxyl ($-\text{COOH}$) group attached to various side-chain groups (usually designated R, R_1 , R_2 ,

etc.). The differences in the side-chain groups confer different properties on the amino acids. For example, glutamic acid has a carboxyl group in its side chain, so it is quite acidic; lysine has an amino group, so it is basic; cysteine has a sulfhydryl (SH) group, so it is capable of forming disulfide bridges (S–S) within the protein molecule. The structures of the 20 naturally occurring amino acids are shown in Figure 2.11.

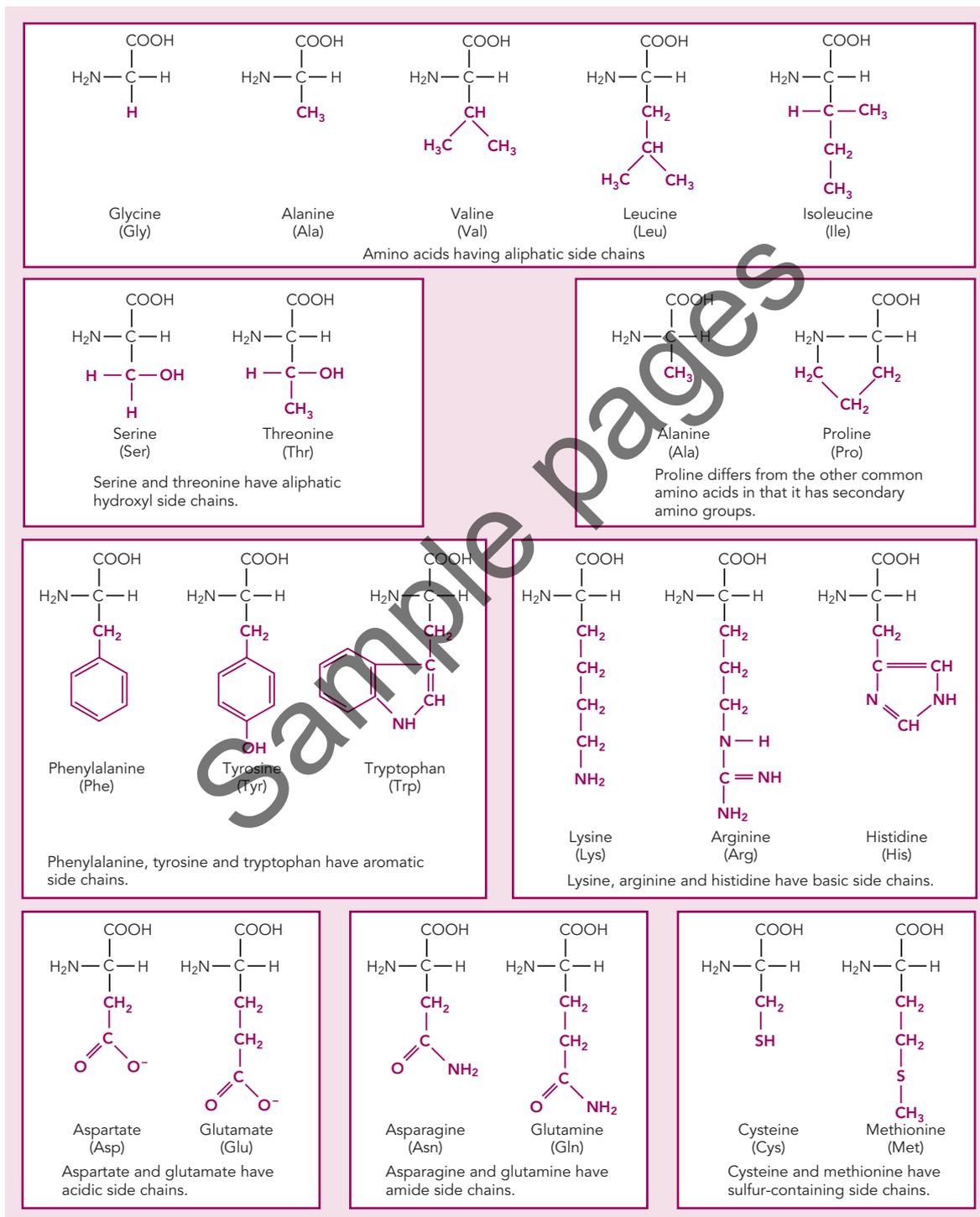


FIGURE 2.11

Structures of the 20 naturally occurring amino acids

The nature of the R side chain confers special properties on the amino acid molecule.

When two amino acids join together, they do so by the removal of a molecule of water from the amino and carboxyl groups of adjacent acids, forming a peptide bond (Figure 2.12). Short sequences of amino acids joined together are called **peptides**; longer ones are called **polypeptides**. Proteins are long polypeptide chains, folded and cross-linked to form compounds with a specific molecular structure.

The nature of the side chains of the amino acids affects the overall structure of the protein. The size, shape and charge of each amino acid determine the ability of the protein to fold and assume a particular conformation and activity (Figure 2.13). When this conformation is disrupted, the proteins are said to be 'denatured' and their function, or enzyme activity, may be lost.

BIOCHEMICAL PATHWAYS OF ENERGY PRODUCTION

Breakdown of polysaccharides

The ability of an organism to utilise different carbohydrates as an energy source depends on its ability to synthesise the

appropriate enzymes. For example, human saliva contains the enzyme amylase, which can break the α -linkages found between the glucose units in starch (see Figure 2.9(b), above). However, humans cannot synthesise the enzyme which breaks the β 1–4 linkage that occurs between the glucose molecules in cellulose (Figure 2.9(c)), and so they are unable to metabolise cellulose. Cellulose therefore acts as fibre, or roughage, in the human diet and aids in digestion and formation of faeces without providing any energy.

Many microorganisms, however, produce an enzyme which can break the β -linkages and release single glucose units. Ruminant animals such as cows rely on the bacteria present in their stomachs to break down cellulose, which is the primary polysaccharide in grass and other fibrous plants. The ability of soil microorganisms to break down cellulose is important for the decomposition of plant organic matter.

Starch, with its branched structure, is mainly a storage carbohydrate. It can be utilised by many organisms, including humans, as a source of energy. Enzymes in the mouth

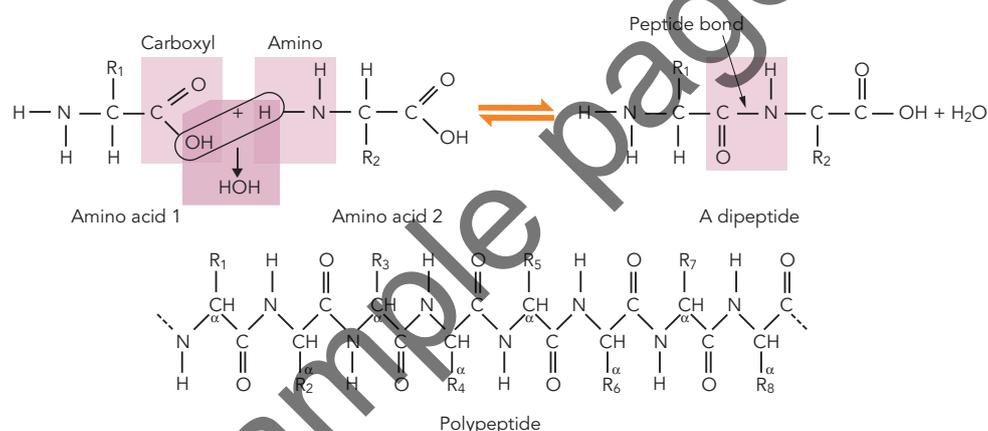


FIGURE 2.12

Synthesis of a peptide bond

Two amino acids join together to form a dipeptide. Water is eliminated from adjacent molecules to form the peptide bond.

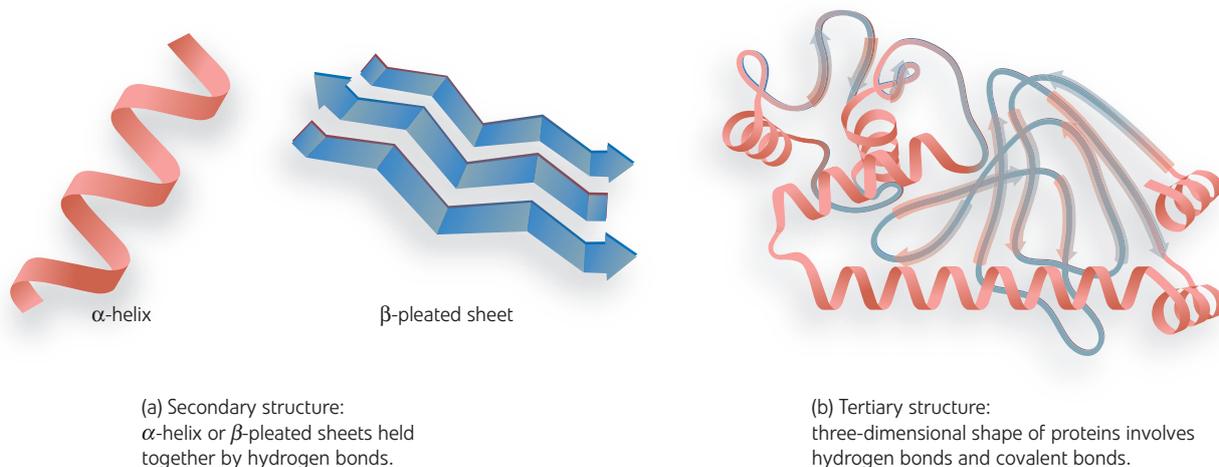


FIGURE 2.13

Structure of proteins

(salivary amylase) and small intestine (pancreatic amylase) break it down to the disaccharide, maltose, for digestion and absorption. Most microorganisms use carbohydrates as their primary source of energy. They are broken down by various reactions to single glucose units, which then enter the pathways described below.

Breakdown of glucose

This is a brief overview of the principal pathways of glucose catabolism in microorganisms. The first stage is called **glycolysis**, or the **Embden-Meyerhof pathway** (see Figure 2.14(a)). It consists of ten reactions, each catalysed by a different enzyme, and is essentially the same in all living cells,

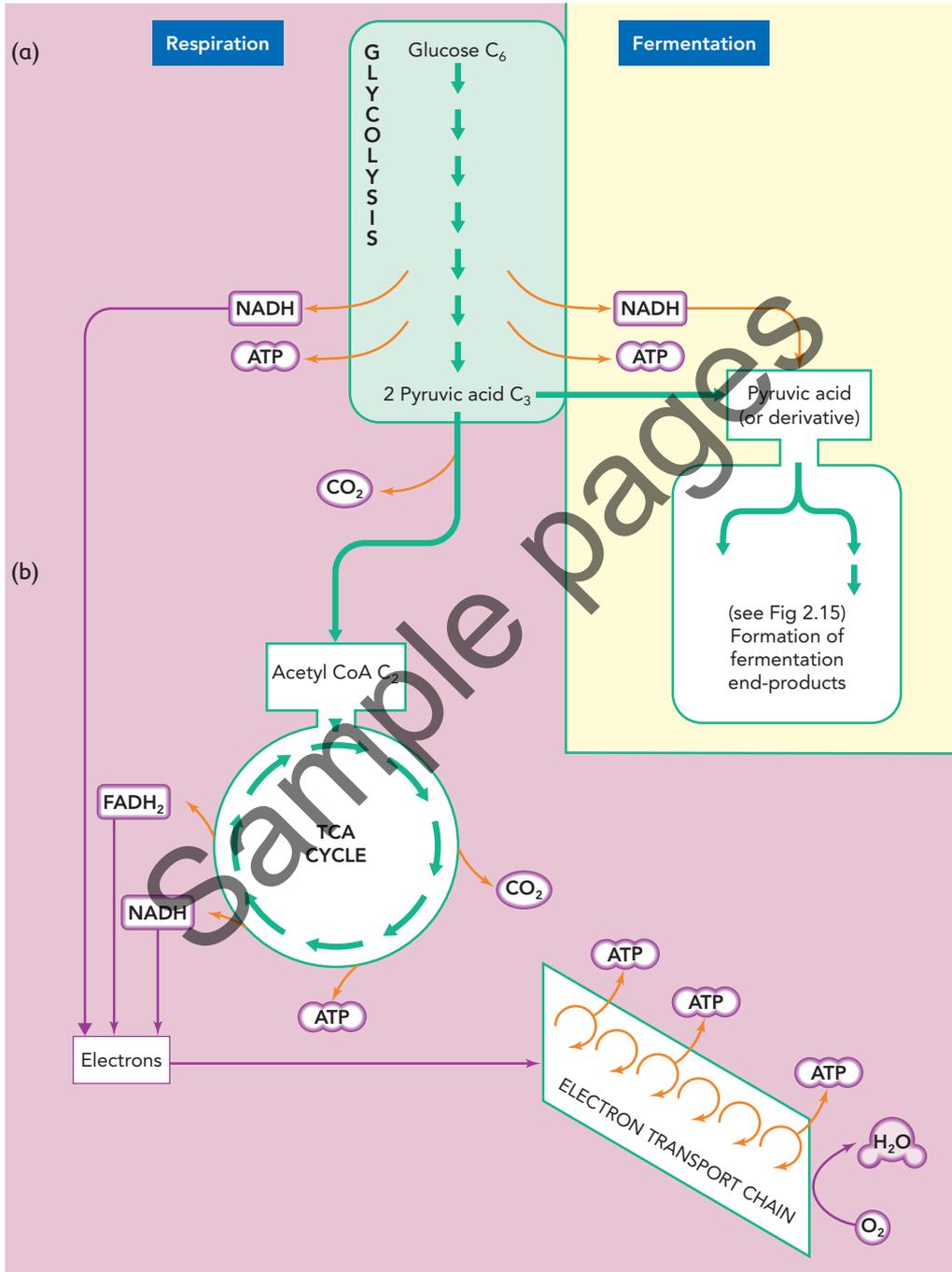


FIGURE 2.14

Overview of glucose metabolism

(a) Embden-Meyerhof pathway of glycolysis

One molecule of glucose gives rise to two molecules of pyruvic acid with the overall yield of two molecules of ATP.

(b) The reactions of the TCA cycle

Pyruvic acid from glycolysis is converted to the two-carbon intermediate acetyl CoA which enters the cycle. Each turn of the cycle results in the release of two molecules of CO_2 . ATP is produced by substrate-level phosphorylation and by oxidative phosphorylation via the electron transport chain.

plant, animal or microbial. It does not require the presence of oxygen. During this process, each molecule of glucose (six carbon atoms) is broken down to form two molecules of **pyruvic acid** (three carbon atoms), yielding only a small amount of energy. A number of other sugars, including fructose, can enter the glycolytic pathway and be converted to pyruvic acid, but the pathways involved are not described here.

Pyruvic acid is an important intermediate that can be further metabolised, either aerobically (respiration) or anaerobically (fermentation). Under aerobic conditions, **respiration** occurs, in which pyruvic acid is first converted to a two-carbon compound called **acetyl CoA**, which then undergoes a series of reactions known as the **Krebs cycle**, or **tricarboxylic acid (TCA) cycle**. This cycle leads to the formation of carbon dioxide and water, and large amounts of energy are released and stored as ATP. Acetyl CoA is a central intermediate that also occurs in the metabolism of lipids and proteins.

Respiration

Organisms that are capable of aerobic respiration oxidise pyruvic acid (three carbon atoms) to acetyl CoA (two carbon atoms), which then enters the Krebs, or tricarboxylic acid (TCA), cycle (Figure 2.14(b)). The TCA cycle provides a mechanism whereby molecules of acetyl CoA continuously enter the cycle and are broken down to carbon dioxide (CO₂) and water with the release of large amounts of energy in the form of ATP. A supply of inorganic phosphate (Pi) is necessary for these reactions to occur.

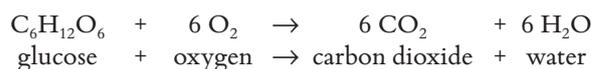
The intermediate steps require the transfer of electrons to the coenzyme, nicotinamide adenine dinucleotide (NAD), which is part of the electron transport chain used to convert the energy from these reactions into ATP. The TCA cycle also provides intermediate compounds that link into pathways involved in the synthesis of lipids, proteins and nucleic acids (see Figure 2.16, page 36).

Oxidative phosphorylation and the electron transport chain

In the presence of oxygen, pyruvic acid is oxidised via the TCA cycle with the release of large amounts of energy. The cell uses a process called **oxidative phosphorylation** to capture this energy in the storage molecule, ATP. The process involves the transfer of electrons from the reduced coenzyme, NADH, along a series of specialised carrier molecules located in the cell membranes to the final electron acceptor, molecular oxygen. This is known as the **electron transport chain**. It is a complex system comprising a series of steps in which each oxidation reaction is linked to a reduction reaction—that is, the energy released from one reaction is immediately used to carry out another reaction and to synthesise ATP.

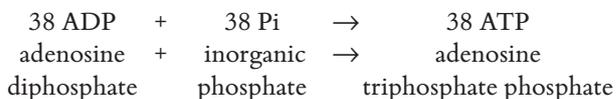
Summary of glucose oxidation

The overall process of the aerobic oxidation of one molecule of glucose can be written:



Each molecule of glucose produces 38 molecules of ATP.

The energy equation can be written:



Fermentation of pyruvic acid

Under anaerobic conditions pyruvic acid undergoes **fermentation**, which is the term used to describe catabolic reactions that occur in the absence of oxygen. The energy yield is low compared to respiration. In microorganisms a number of different fermentative pathways may be followed, some of which give rise to useful products (see Figure 2.15).

No single organism has all the enzymes required for all these pathways. In fact, most organisms use only one pathway. Microbiologists can use this property to identify a particular organism by characterising the end products they release into the environment from fermentation reactions. Conversely, pure cultures of specific microorganisms can be added to a particular substrate to produce a desired product. Many of the products of microbial fermentation are of great use to humans. For example, yeast (*Saccharomyces*) is added to grape juice to produce alcohol in winemaking; yeast is also added to bread to produce carbon dioxide, which causes the bread to rise; and lactobacilli are added to various milk products to induce 'souring' during the production of cheese and yoghurt. These processes are described in more detail later in this chapter (see Table 2.1, page 39).

Anaerobic respiration

Some microorganisms use inorganic substances as the final electron acceptor instead of oxygen. For example, some species of *Pseudomonas* and *Bacillus* can use the nitrate ion (NO₃⁻) as the acceptor, forming nitrite (NO₂), nitric oxide (NO) or nitrogen gas. Others use sulfate or carbonate. The amount of ATP formed is not usually as great as that produced when molecular oxygen is the final electron acceptor, but is quite significant.

The metabolic reactions described above are for glucose, which is the primary source of energy in most cells. However, lipids and proteins are also broken down with the release of energy and the formation of small molecules (building blocks) that can be used in other cellular processes.

Breakdown of lipids

Triglycerides are readily broken down by enzymes called **lipases**, which split the fatty acids from the glycerol molecule. The glycerol is then converted to dihydroxyacetone phosphate, an intermediate in glycolysis. The fatty acids are broken down by β -oxidation into two-carbon units of acetyl CoA which enter the TCA cycle. The metabolism of fats therefore yields large amounts of energy.

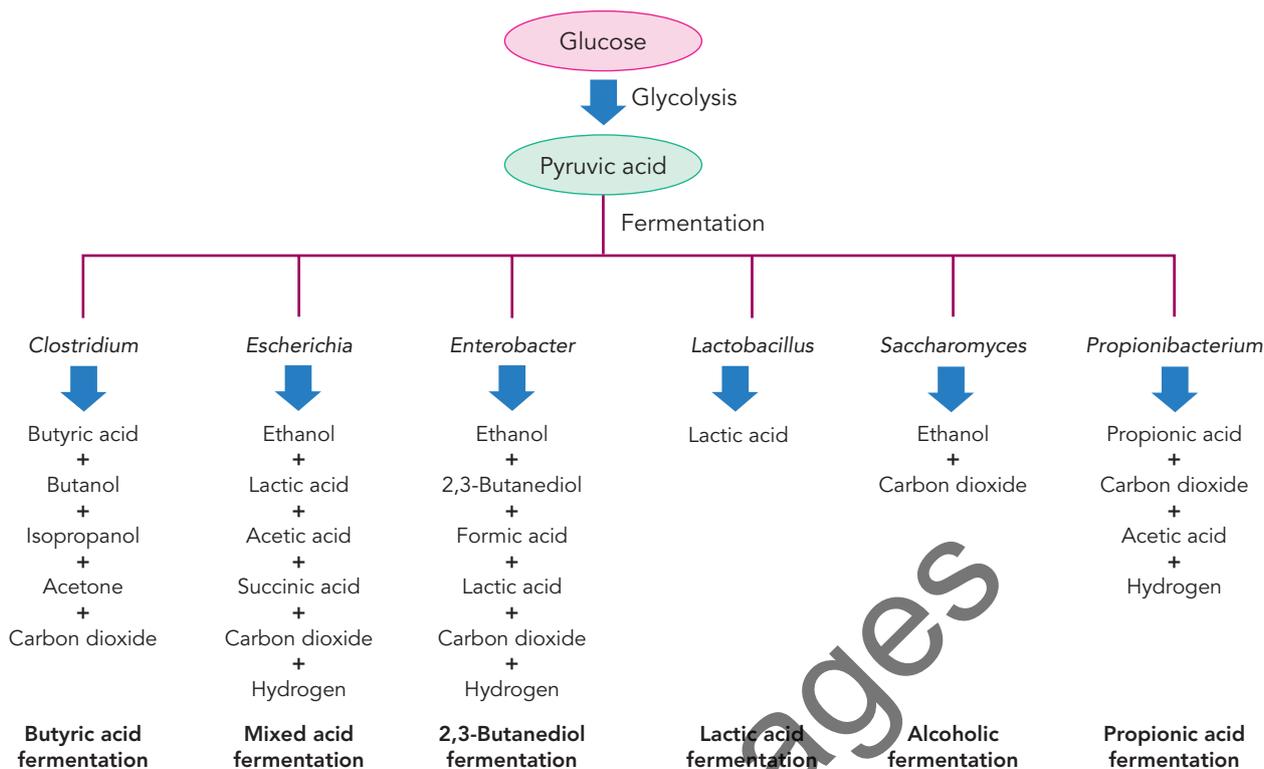


FIGURE 2.15

Major pathways of fermentation

Different microorganisms use characteristic fermentative pathways. The products of fermentation can be used to aid in identification of the microorganisms.

Breakdown of proteins

Proteins are broken down in most bacterial cells by proteolytic enzymes (called proteases or peptidases) to form smaller peptide fragments or single amino acids. The amino acids are reused for protein synthesis or are metabolised further. The removal of the amino group gives rise to compounds that can enter the TCA cycle and be broken down, producing energy in the form of ATP.

ANABOLISM—BIOSYNTHESIS OF CELLULAR COMPONENTS

The energy released in the reactions described above is used by the cell in a number of different ways. ATP is needed for the synthesis of the chemical components of the cell—that is, the carbohydrates, lipopolysaccharides, RNA and DNA, structural proteins and enzymes, the cell wall and the phospholipids of the cell membrane. The small building blocks of these complex macromolecules are activated by combining with ATP so that they have sufficient energy to enter their respective biosynthetic pathways.

Energy is also used for active transport of substances into and out of the cell and for the movement of flagella and cilia.

Biosynthesis of carbohydrates

Microorganisms utilise different pathways for the synthesis of carbohydrates, depending on the availability of nutrients and the particular needs of the organism.

Autotrophs are organisms that can use carbon dioxide from the air as their primary source of carbon; that is, they can live and reproduce without access to complex molecules. They are able to use energy from the sun during photosynthesis to synthesise glucose from carbon dioxide with the release of oxygen. These organisms include photosynthetic bacteria (cyanobacteria, green sulfur and purple sulfur bacteria), algae and green plants.

Heterotrophs, which include most bacteria, fungi and protozoa, must be provided with a source of organic carbon in their environment in order to synthesise glucose and larger polysaccharides. The intermediates in the TCA cycle, or other breakdown products from lipid or protein metabolism, can be a suitable source. Depending on the needs of the cell and the availability of energy in the form of ATP, various biosynthetic pathways are utilised which are the reverse of (or parallel to) those described for the breakdown of carbohydrates. However, some microorganisms have strict nutritional requirements; these are described in Chapter 3.

One important complex polysaccharide molecule that is synthesised only by prokaryotic cells is **peptidoglycan**, a compound that provides strength and rigidity to bacterial cell walls. Peptidoglycan is a complex molecule composed of sugar molecules cross-linked by peptide bridges (see Chapter 3).

Biosynthesis of lipids

Bacterial lipids are usually formed by the condensation of long-chain fatty acids with a molecule of glycerol to form triglycerides. The fatty acids are synthesised from units of acetyl CoA, which occur as a breakdown product of carbohydrate metabolism.

Biosynthesis of amino acids

Amino acids are required for the synthesis of proteins and also serve as precursors for the purine and pyrimidine bases which are the building blocks of the nucleic acids RNA and DNA. Amino acids are synthesised in microbial cells by the addition of an amino group ($-\text{NH}_2$) to various intermediates in the TCA cycle. The nitrogen may be derived from ammonium salts (NH_4^+) or nitrates (NO_3^-), or from nitrogen in the atmosphere. Some bacteria are capable of using atmospheric nitrogen to form nitrogenous compounds in a process called **nitrogen fixation**.

Biosynthesis of proteins

The biosynthesis of proteins occurs under the direction of the genetic material of the cell, the DNA. It requires the participation of RNA and other nucleotides and is described in detail in Chapter 4.

INTERRELATIONSHIP OF METABOLIC PATHWAYS

Living cells are in a constant dynamic equilibrium of breakdown, synthesis and repair. The metabolic pathways are interrelated so that, for example, acetyl CoA molecules formed from breakdown of lipids can be used in the TCA cycle to produce energy; intermediates in the TCA cycle can be converted to amino acids and then incorporated into proteins. The integration of these processes allows for the most efficient use of nutrients and energy. Some microorganisms can synthesise all their cellular requirements from simple organic or inorganic compounds. However, microorganisms show great diversity in their nutritional requirements and some require more complex molecules such as sugars, amino acids, and vitamins or cofactors. Some mutant strains of bacteria may have an absolute requirement for a particular compound (e.g. a fatty acid or amino acid). Figure 2.16 illustrates the interrelationship between the pathways of bacterial metabolism. It is beyond the scope of this text to examine all these pathways in detail, but it is important to remember that most microorganisms of medical importance are **heterotrophic**—that is, they use complex carbon-containing molecules as their primary nutritional source. This property is useful for the identification and control of potential pathogens.

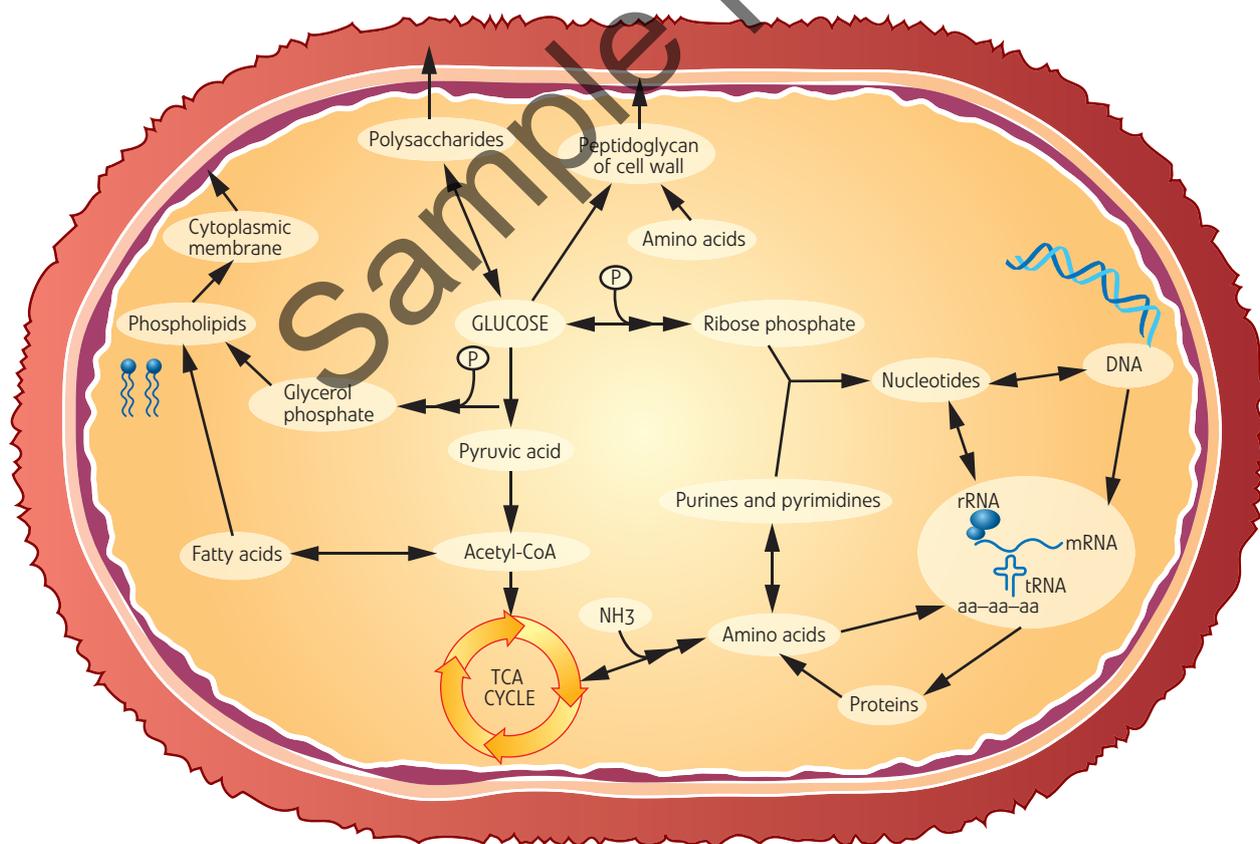


FIGURE 2.16

Interrelationship of metabolic pathways

Bacteria are able to synthesise all their cellular requirements when supplied with a simple carbon source, nitrogen (ammonia) and inorganic salts.

PRACTICAL APPLICATIONS OF MICROBIAL PROCESSES

The metabolic pathways described in this chapter represent only a small fraction of the reactions carried out by living cells. These reactions are controlled by specific enzymes produced by cells according to the information contained in their genes. In animal cells, the production of a set of enzymes by a cell is usually fixed, and reflects the function of that cell. If the required enzymes are not produced or are produced in the wrong amount, the workings of the cell are greatly affected. The measurement of the amount of enzyme produced by a cell can be used to diagnose whether a cell is functioning normally. For example, in humans, liver function tests measure the levels of different liver enzymes in blood to determine whether the liver is damaged by diseases such as hepatitis or cirrhosis.

In the microbial world, bacteria are able to adapt the metabolic pathways they use to the nutrients available. Because the bacterial cell is so small, it only synthesises enzymes for its immediate needs, even though it may carry the genetic information for other enzymes. When provided with alternative carbon sources it is often able to synthesise the enzymes necessary for metabolism of that compound. The metabolic pathways employed therefore reflect the environment in which the organism is generally found—for example, the sulfur bacteria occur in hot springs and synthesise enzymes that use sulfur compounds as electron acceptors.

The metabolic abilities of specific microorganisms are used by scientists in a number of areas including diagnostic microbiology, industry, scientific research and genetic engineering. The ability of microorganisms to break down large biological molecules is important for the decomposition of waste matter in the environment.

Diagnostic microbiology

In the laboratory most pathogens grow readily on plates made of a rich medium containing horse blood, ‘blood agar’, but in order to identify them they must be grown on additional media. The ability (or lack of it) to grow under different environmental conditions is used by microbiologists to differentiate between species of microorganisms that may appear morphologically identical. This is the principle behind the selective and differential media used in a diagnostic microbiology laboratory. A brief introduction is appropriate here to show how an understanding of metabolic pathways helps to identify different pathogens.

A **selective medium** is one to which one or more chemical compounds have been added to prevent the growth of certain microorganisms, but not others. The ‘compound’ is often something as simple as a high salt concentration. A **differential medium** is one to which some sort of indicator, usually a dye, has been added. This allows the clinical microbiologist to differentiate between various bacteria on the basis of chemical reactions that occur during growth.

Sometimes the medium is both differential and selective at the same time. For example, mannitol salt agar (MS agar) is used in the identification of a mixture of streptococci and different species of staphylococci. Staphylococci grow readily in the high salt concentration, whereas the growth of streptococci is inhibited. *Staphylococcus aureus* produces enzymes, which utilise mannitol (a sugar alcohol) and convert it to an acid that turns the pink indicator dye in the medium to yellow. *Staphylococcus epidermidis* appears similar under the microscope to *Staphylococcus aureus*, but does not metabolise mannitol, so there is no acid production and no colour change in the medium (see Figure 2.17).

A battery of different biochemical tests is used in the microbiology laboratory to assist in the identification of a particular microorganism, and many of these involve the measurement of an enzyme reaction. In effect, these tests detect the presence or absence of the enzyme required for a reaction similar to the ones described above, thus indicating the kind of organism that is present. Enzyme reactions that produce gases such as carbon dioxide or hydrogen can also be seen by the appearance of bubbles in the culture medium. Enzymic breakdown of sulfur compounds with the release of hydrogen sulfide is detected by reacting the gas with iron (ferric) compounds—as well as by its offensive smell!

Many commercial companies market diagnostic kits for the identification of microorganisms. These kits test for the presence of several different enzymes in a single specimen. Some of these reagents are very sensitive and can give a positive diagnosis when only a small amount of specimen is available. This topic is discussed further in Chapter 15.

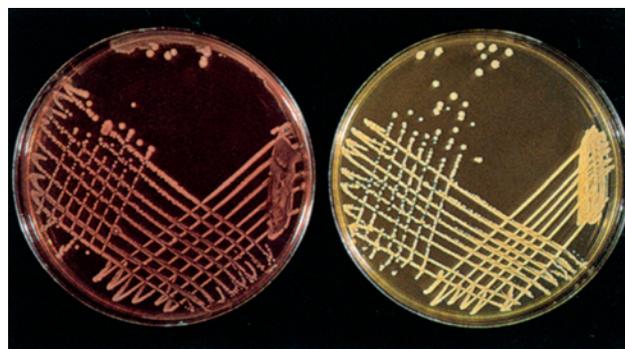


FIGURE 2.17

Mannitol-fermentation test

Growth on mannitol salt agar distinguishes different species of staphylococci. *Staphylococcus aureus* ferments mannitol, producing acid, which turns the indicator in the medium to yellow. *Staphylococcus epidermidis* does not produce a colour change.

Source: Dr Penny Bishop.

INDUSTRIAL APPLICATIONS OF MICROBIAL METABOLISM

Microorganisms are used in industry in a number of different ways—for example:

- as living cell cultures in the production of food
- in the preparation of purified enzymes
- as a source of primary or secondary metabolites.

Live microbial cells

Live cell preparations, such as yeasts, and lactic acid bacteria are widely used in the food industry. Fermentation of sugar by yeast cells under anaerobic conditions produces alcohol and carbon dioxide. For example:

- Wine is made from fermented grape juice. Wild yeast cells are found on grapes in nature. In practice, the wine they produce is of variable quality, so most winemakers add a pure culture of the yeast, *Saccharomyces ellipsoideus*, and control the fermentation by the amount of oxygen present. An understanding of metabolism can be used to achieve the desired product. When oxygen is low or absent, yeast fermentation produces mainly alcohol. When oxygen is available, carbon dioxide is also formed. When the alcohol concentration reaches 12–14 per cent the yeast cells die. Champagne is made by a secondary fermentation in which fresh yeast cells and sugar are added to still wine in a sealed bottle. The carbon dioxide produced cannot escape and remains as bubbles in the champagne.
- Beer is also a product of yeast fermentation. An extract of grains such as barley is used to make a sugar substrate, or 'malt'. Hops are added for flavour. Special strains of yeast, *Saccharomyces carlsbergensis* or *Saccharomyces cerevisiae*, are used.
- The leavening of bread also relies on the properties of the yeast cell. When added to the dough, yeast produces

alcohol and carbon dioxide gas. The carbon dioxide causes the bread to rise. When the bread is baked, the alcohol and carbon dioxide evaporate, leaving the distinctive holes in the bread. Sourdough bread is made by adding cultures of lactic acid bacteria to the dough to produce the distinctive 'sour' taste.

- Lactic acid bacteria produce lactic acid by fermentation of glucose and are responsible for the 'souring' of milk to produce cheese, yoghurt, buttermilk and other products. Various other bacteria and some fungi are added to produce intermediate compounds which impart a distinctive appearance and flavour to cheeses (e.g. different strains of the mould *Penicillium* are used for blue cheeses and camembert).
- Cultures of the nitrogen-fixing bacterium *Rhizobium* are frequently inoculated on to leguminous plant seeds to encourage the formation of root nodules where nitrogen fixation can take place.
- Mushrooms are cultivated for food.

Microbial enzymes

Microorganisms can also provide a source of enzymes for use in industry. Some chemical reactions are difficult to carry out by non-enzymic means. As explained earlier in this chapter, enzymes are biological catalysts that allow chemical reactions to occur more efficiently by lowering the activation energy—often giving a much higher yield of product for a lower cost. Specific enzyme proteins can be extracted from large-scale preparations of microorganisms, purified and used in the commercial production of

CASE HISTORY 2.1

Diagnosis of illness

A 10-year-old boy visited the GP's surgery complaining of a sore throat and looking flushed. On examination he was found to have a temperature of 38.4°C and his tonsils were red and swollen with areas of pus on them. The doctor also noticed that he had a number of infected sores on his arms and legs. His mother explained he had scratched some mosquito bites. The doctor took swabs from the boy's throat and also from the skin sores and sent them to the pathologist. He started the boy on some broad spectrum antibiotics.

In the microbiology laboratory, each specimen was cultured on blood agar and also on Mannitol salt agar. The skin specimen grew well on blood agar, and on mannitol salt it produced a yellowish colour. The throat specimen grew on blood agar, but there was no visible growth on mannitol salt agar.

Questions

1. What does this tell you about the bacteria present at each site?
2. What would be the preliminary identification of the organisms from each site?
3. Which infection do you think is most likely responsible for the boy's temperature (see Chapter 7)?
4. What further tests would be needed to decide whether the doctor had prescribed the best treatment (see Chapter 12)?

a particular compound. Examples are protein-digesting enzymes (proteases in laundry detergents) and glucose isomerase, which converts glucose to fructose for use in the confectionery industry. Another useful enzyme is penicillin acylase, used in the manufacture of semi-synthetic antimicrobial drugs.

Primary metabolites

Many different organic compounds are produced by microorganisms in the metabolic reactions outlined in this chapter. **Primary metabolites** are products formed in the major pathways of fermentation. These compounds can often be obtained in sufficient yields to be of use commercially. Some of the more important metabolites are described below.

- Citric acid, an intermediate in the TCA cycle, is widely used as a flavouring in foods and beverages.
- Sorbose, produced when the bacterium *Acetobacter* oxidises sorbitol, is used to make ascorbic acid (vitamin C).
- Acetic acid (vinegar) is a product of the oxidation of alcohol (one of the products of fermentation) by members of the genera *Acetobacter* and *Gluconobacter*. Sometimes the *Acetobacter* are an accidental contaminant of food and cause acid production or 'souring'. Vinegar can be produced commercially from the alcohol present in wine or any other product of alcoholic fermentation. Vinegar is used to preserve (pickle) food because most harmful bacteria do not tolerate acid conditions.
- Vitamins and growth factors are used as food supplements for humans and in animal feeds. Most vitamins are made commercially by chemical synthesis, but some are too complex and so are produced by microbial fermentation. These include vitamin B12 and riboflavin.

- Some amino acids are produced by microbial means. Chemical synthesis of amino acids usually results in the formation of a mixture of the D- and L-stereoisomers. Naturally occurring amino acids exist only the L- form. Using microorganisms to produce the L-amino acid means that a pure product can be obtained. Some useful amino acids are:

- the salt of glutamic acid, monosodium glutamate (MSG), used as a flavour enhancer
- phenylalanine and aspartic acid, which are components of the sweetener, aspartame
- lysine, an essential amino acid for humans, produced as a food supplement.

Some uses for products of microbial fermentation are listed in Table 2.1.

Secondary metabolites

Secondary metabolites are an interesting group of organic compounds, usually produced by microorganisms when nutrients have been depleted and the number of cells is no longer increasing rapidly. These compounds do not appear to be essential for growth or reproduction. The formation of these metabolites is limited to certain kinds of organisms and is very dependent on growth conditions. It is possible to select for specific strains of an organism in order to enhance the yield of the desired metabolite.

Antibiotics form one of the most important groups of secondary metabolites, and their discovery has had an enormous impact on the practice of medicine since the first commercial development of penicillin in the 1940s. More than 5000 antibiotic substances have been described, but most of them are too toxic for human use. Most commercially useful antibiotics are produced by filamentous fungi and bacteria of the actinomyces group (see Table 2.2). They are described more fully in Chapter 12.

TABLE 2.1 Some useful fermentation reactions

PRODUCT	USES	SUBSTRATE	MICROORGANISM
Lactic acid	Cheese, yoghurt Sauerkraut	Milk Cabbage	<i>Lactobacillus</i> spp.
Propionic acid and carbon dioxide	Swiss cheese	Milk	<i>Propionibacterium</i>
Ethanol	Beer Wine Fuel	Malt extract Grape juice Agricultural waste	<i>Saccharomyces cerevisiae</i> (yeast) <i>Saccharomyces ellipsoideus</i> (yeast) <i>Saccharomyces cerevisiae</i>
Acetic acid	Vinegar	Ethanol	<i>Acetobacter</i> (bacterium)
Glycerol	Industry/Food/Pharmaceutical	Molasses	<i>Saccharomyces cerevisiae</i>
Citric acid	Flavouring	Molasses	<i>Aspergillus</i> (fungus)
Sorbose	Vitamin C	Sorbitol	<i>Acetobacter</i>

TABLE 2.2 Commercial production of antibiotics

ANTIBIOTIC	MICROORGANISM	TYPE
Penicillin	<i>Penicillium chrysogenum</i>	Fungus
Cephalosporin	<i>Cephalosporium</i> spp.	Fungus
Bacitracin	<i>Bacillus subtilis</i>	Bacterium
Polymixin B	<i>Bacillus polymyxa</i>	Bacterium
Cycloheximide	<i>Streptomyces griseus</i>	Actinomycete
Streptomycin	<i>Streptomyces griseus</i>	Actinomycete
Erythromycin	<i>Streptomyces erythreus</i>	Actinomycete
Aminoglycosides	<i>Streptomyces</i> spp.	Actinomycete
Tetracycline	<i>Streptomyces rimosus</i>	Actinomycete

MICROORGANISMS AS TOOLS IN SCIENTIFIC RESEARCH

The metabolic processes that occur in microorganisms are very similar to those occurring in the cells of higher organisms. In fact, much of the scientific knowledge about the metabolism of animal cells was derived from laboratory research using bacteria. Bacteria are easy to grow in large numbers on a defined medium (i.e. controlled nutrients and growth conditions). They provide the research worker with a uniform population of cells, making it easier to interpret results. The simple genetic material (the DNA is all on one circular chromosome) provided the ideal system for early genetic mapping experiments. The techniques and knowledge obtained from this 'simple' system have now been adapted for use in more ambitious projects such as mapping of the DNA of the human genome. A knowledge of the processes of DNA replication in bacteria provided the scientist with a tool for manipulating the genetic information in cells—**genetic engineering**.

Genetic engineering

A very important use for microorganisms in recent years has been the production of specific compounds for medical use by the process of genetic engineering. The discovery and isolation of enzymes called restriction endonucleases—bacterial enzymes that were able to rupture strands of DNA at specific sites in the nucleic acid chain—allowed scientists to manipulate the structure of bacterial DNA. Scientists have developed methods of inserting genetic information into microbial cells in such a way as to direct the microbe to synthesise large amounts of a desired compound. Examples of genetic engineering include the production of human insulin, human growth hormone and

some vaccines. Genetic engineering is described in more detail in Chapter 4.

ENVIRONMENTAL USES FOR MICROORGANISMS

Microorganisms play an essential role in the environment in decomposition and recycling of nutrients.

Decomposition

The catabolic reactions carried out by many microorganisms are essential for the decomposition of organic matter such as plant material and sewage. The enzymes produced by microorganisms break down complex carbohydrates and other biological compounds, releasing small organic molecules into the soil. These molecules are a source of nutrients for growing plants.

Hydrocarbon metabolism

Hydrocarbons are organic compounds containing only hydrogen and carbon, and are usually insoluble in water. Two examples are oil and petroleum. The chemical breakdown of hydrocarbons is a slow process, requiring oxygen. Very few microorganisms can utilise hydrocarbons for growth. The exceptions are some strains of *Pseudomonas*, *Nocardia* and *Mycobacterium*, and some yeasts and moulds. Cultures of *Pseudomonas* have been used to help disperse oil spills. Usually, the hydrocarbon metabolism would proceed too slowly to be of use, but it was found that the addition of other nutritional requirements, nitrogen and phosphate to speed up metabolism and growth of the bacteria enabled the oil to be broken down and dispersed.

Microorganisms have been used in bioremediation projects such as those undertaken at the Sydney Olympic site (see Spotlight box: Bioremediation—environmental uses for bacteria).

Bioremediation—environmental uses for bacteria

When the Homebush site in Sydney was chosen for the 2000 Olympic Games, one of the problems confronting the organisers was the fact that for many years the area had been used to dump the city's waste. Wilson Park is a 12-hectare portion of Crown Land adjacent to the Olympic site. It was used for the production of town gas for Sydney between 1953 and 1974, and approximately 230 000 tonnes of hydrocarbon (tar) waste remained buried on the site (see Figure 2.18(a)).

A feature of the Sydney 2000 bid had been the guarantee of a 'green games', so the main task was to clean up the site to a level where the risks to humans and the ecology of the area could be contained.

A number of toxic volatile organic compounds such as benzene, toluene, ethyl benzene and xylene were emanating from the buried tar and were found in the groundwater on site. The challenge was to remove these compounds and make the area safe.

Three Sydney scientists were commissioned by the Olympic Coordination Authority to undertake the project. They found that there were high numbers of hydrocarbon-degrading bacteria present in the topsoil of the site. In the laboratory these bacteria were capable of completely metabolising benzene to carbon dioxide and water if simple plant fertilisers containing nitrogen and phosphorus were added to the cultures. The isolated bacteria included species of *Pseudomonas*, *Bacillus*, *Microbacterium*, *Actinomyces* and *Gordonia*. Some of these bacteria were similar to those used to clean up oil spills overseas.

The remediation project consisted of providing a healthy vegetated topsoil cover to act as a microbial biofilter. The enhanced bacterial activity in the soil was responsible for degrading the hydrocarbon gas pollutants and stopping their reaching the surface. In this way the park was made safe for human use once more (see Figure 2.18(b)).



FIGURE 2.18(a)

Aerial view of gasworks at Wilson Park, Silverwater, in 1962



FIGURE 2.18(b)

Wilson Park, 2000, after remediation and prior to Sydney Olympic Games

Source: Photos by D. Sheumack from D. Sheumack, M. Howe, B. Bicknell, M. Muir, J. Pym, E. Ling and K. Hughes 2000, Contamination assessment and bioremediation at Wilson Park. *Proceedings of the 15th International Clean Air and Environment Conference*, Sydney, Clean Air Society of Australia and New Zealand Inc., November 2000, Hanitro Pty Ltd, vol. 1: 191–98.

SUMMARY

- ❖ Metabolism is the overall term used to describe the chemical reactions of breakdown (catabolism), synthesis (anabolism) and repair that occur in living cells.
 - ❖ Organic compounds are compounds containing carbon; they exhibit stereoisomerism by forming four covalent bonds with other atoms.
- STRUCTURE OF BIOLOGICAL MOLECULES**
- ❖ Living matter is made up of complex molecules containing the elements carbon, hydrogen, oxygen, nitrogen, phosphorus and sulfur, and other, trace elements.
 - ❖ The arrangement of atoms within a molecule determines its shape and function.
 - ❖ The particular structure of biological molecules enables them to combine with complementary molecules. This is termed 'specificity'.

ENZYMES AND CHEMICAL REACTIONS

- ❖ Enzymes are proteins that act as biological catalysts to lower the activation energy of a reaction.
- ❖ The production of enzymes is under the control of the DNA of the cell.
- ❖ Enzymes have a particular shape, containing an active site that is specific for the substrate; some require a cofactor for activity.

ENERGY PRODUCTION IN BIOLOGICAL SYSTEMS

- ❖ Photosynthesis uses energy from the sun to convert carbon dioxide and water to glucose.
- ❖ Cells break down glucose and release energy by the processes of glycolysis, fermentation and respiration.
- ❖ ATP is a small, biological, energy-rich storage molecule.
- ❖ Breaking the bond of the terminal phosphate group of ATP releases large amounts of energy, used by the cell for essential processes.

STRUCTURE OF BIOLOGICAL MOLECULES*Structure of carbohydrates*

- ❖ Carbohydrates (sugars) are organic compounds made up of carbon, hydrogen and oxygen.
- ❖ The most common sugars are hexoses and pentoses; they may exist as monosaccharides, disaccharides or polysaccharides.

Structure of lipids

- ❖ Fats, or triglycerides, are composed of glycerol and long-chain fatty acids.

Structure of proteins

- ❖ There are 20 different naturally occurring amino acids.
- ❖ Proteins consist of chains of amino acids joined by peptide bonds in a specific sequence and folded into a particular shape.

BIOCHEMICAL PATHWAYS OF ENERGY PRODUCTION

- ❖ Energy is produced from the oxidation of carbohydrates by a process of glycolysis, followed by either fermentation or respiration.

- ❖ Fermentation occurs anaerobically and results in the formation of a number of useful products, depending on the microorganism.
- ❖ Respiration via the Krebs (TCA) cycle occurs in the presence of oxygen with the formation of carbon dioxide, water and large amounts of energy, which is stored as ATP.
- ❖ Proteins are broken down by proteolytic enzymes into amino acids, which can take part in other reactions or in the synthesis of new proteins.

ANABOLISM—BIOSYNTHESIS OF CELLULAR COMPONENTS

- ❖ Autotrophs are organisms that use carbon dioxide to synthesise glucose.
- ❖ Heterotrophs require a source of organic carbon to synthesise cellular components.
- ❖ Bacteria synthesise lipids from acetyl CoA and glycerol.
- ❖ Amino acids are synthesised by incorporation of nitrogen from nitrates or ammonia into the intermediates of the TCA cycle.
- ❖ Some bacteria can use atmospheric nitrogen to synthesise nitrates and ammonia.
- ❖ Proteins are formed in a complex series of reactions, directed by the nucleic acids, DNA and RNA.

INTERRELATIONSHIP OF METABOLIC PATHWAYS

- ❖ Microorganisms can use many different interrelated metabolic pathways to synthesise cellular requirements.

PRACTICAL APPLICATIONS OF MICROBIAL PROCESSES

- ❖ Many of the reactions carried out by microorganisms are of benefit to humans.
- ❖ Biochemical reactions are used to identify bacteria.
- ❖ Microorganisms and their products are used in the food industry.
- ❖ Genetic engineering involves the use of microorganisms to produce compounds for human use.

ENVIRONMENTAL USES FOR MICROORGANISMS

- ❖ Microbial metabolism is important for the decomposition of sewage and other organic matter.

STUDY QUESTIONS

1. What is meant by 'metabolism'?
2. Which elements make up biological molecules?
3. What name is given to compounds containing carbon?
4. (a) What name is given to the process in which carbon dioxide from the air is converted to glucose?
(b) What type of microorganisms can carry out this process?
5. How do microbial cells obtain energy for their synthetic reactions?
6. Why are enzymes called biological catalysts?
7. What is meant by the 'active site' of an enzyme?
8. Give two examples of naturally occurring monosaccharides, disaccharides and polysaccharides.
9. Which metabolic pathway produces most energy?
10. Which compound is the key intermediate that links different metabolic pathways?
11. Give three examples of fermentative processes that are of use to humans.
12. How do microorganisms contribute to the recycling of nutrients to the environment?

TEST YOUR UNDERSTANDING

1. Give three examples of specificity in biological systems.
2. Explain how the properties of the carbon atom contribute to the shape and function of biological molecules.
3. Describe the difference between autotrophic and heterotrophic organisms.
4. How can the biochemical properties of microorganisms be used to aid in their identification?

Sample pages