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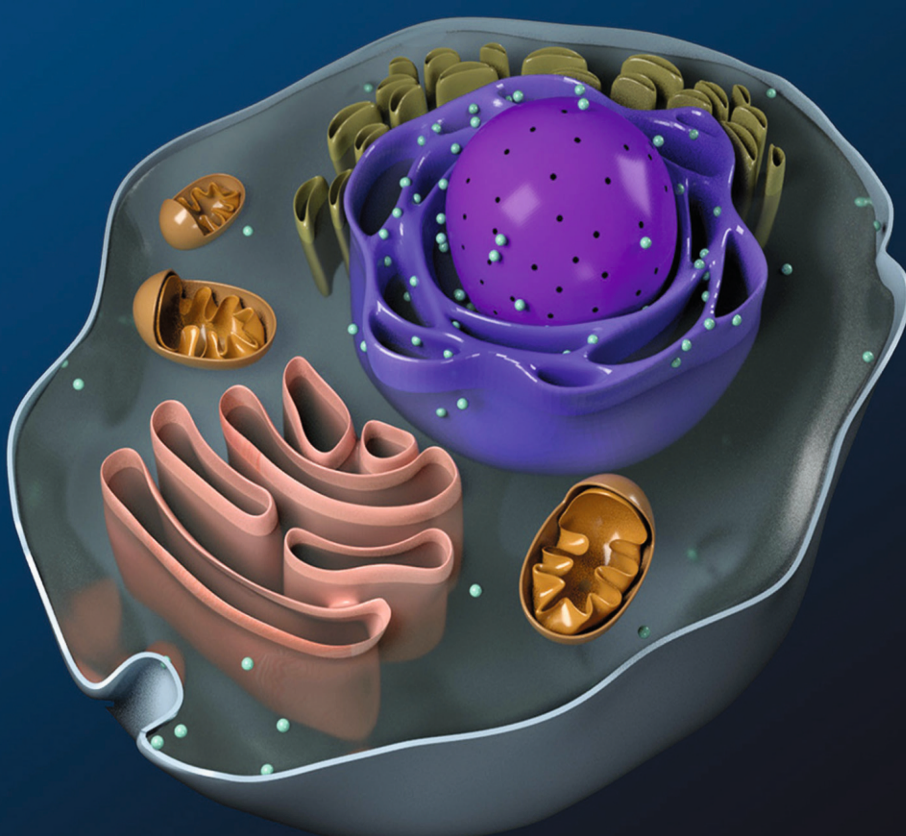
BIOTECHNOLOGY

A PROBLEM APPROACH

Sixth Edition



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Revised and Updated

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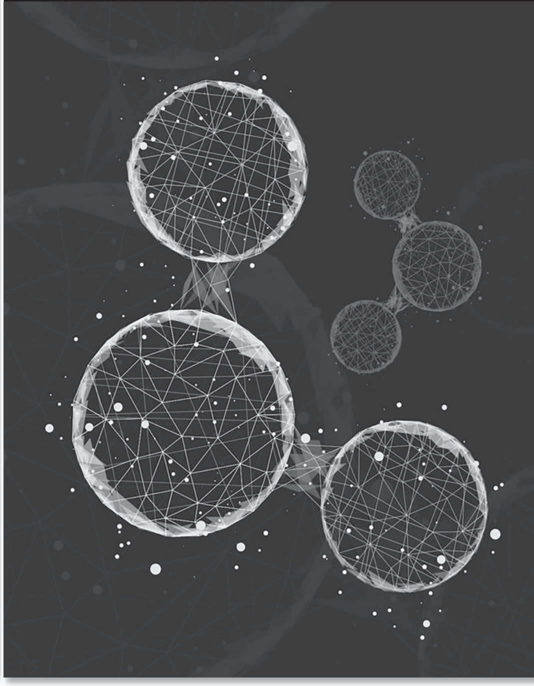
Chapter 1

Chemical foundations of life



Biological processes in living organisms follow the laws of physics and chemistry, so in order to understand how biological systems work, it is important to understand the underlying physics and chemistry. Therefore, the fundamentals of chemistry are important to understand biological processes. In this section, we have described some important chemical concepts necessary to understand cellular chemistry.

1.1 Matter and Energy

Matter is anything that has mass and occupies space. All bodies consist of matter. Matter can exist in three physical states – solid, liquid and gas. **Solids** have definite volume and definite shape. **Liquids** have definite volume but not the definite shape. They take the shape of the container in which they are placed. **Gases** have neither definite volume nor definite shape. They completely occupy the container in which they are placed.

Law of conservation of matter or mass

In a closed system, the total quantity of matter during a chemical reaction or during a physical change remains constant over time. In other words, matter cannot be created or destroyed in a chemical reaction or during a physical change; it can only be rearranged or transformed from one form to another.

Energy is defined as the capacity to do work or produce heat. Energy can be classified into two principal types: kinetic energy and potential energy. A body in motion possesses energy because of its motion. Such energy is called **kinetic energy**. The energy a body possesses because of its position, condition, or composition is called **potential energy**.

Law of conservation of energy

Energy cannot be created or destroyed in a chemical reaction or in a physical change. It can only be converted from one form to another.

Atom and molecules

Matter is composed of one or more elements. The smallest particle of an element is called an **atom**. Atoms consist principally of three fundamental sub-atomic particles: *electrons*, *protons*, and *neutrons*.

Chapter 2

Biomolecules and Catalysis

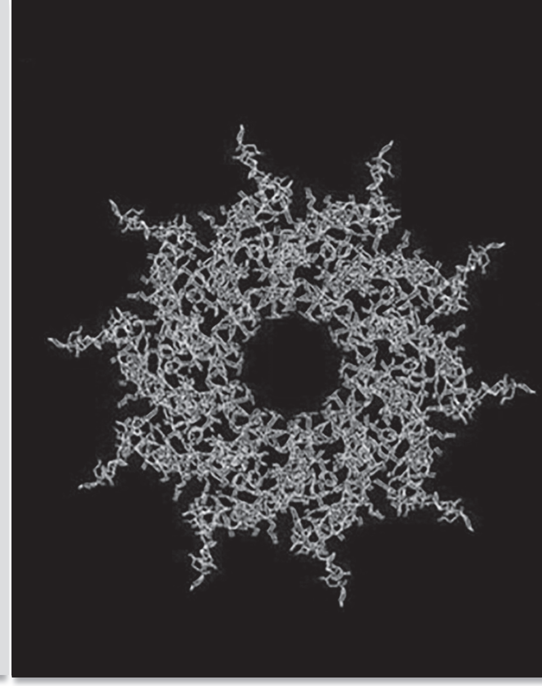
Biomolecules are carbon-based organic compounds that are produced by living organisms. Most biomolecules can be regarded as derivatives of hydrocarbons, with hydrogen atoms replaced by a variety of functional groups that confer specific chemical properties on the molecule. These molecules consist of a relatively small number of elements. Approximately 25 naturally occurring chemical elements are found in biomolecules, and most of these elements have a relatively low atomic number. In terms of the percentage of the total number of atoms, hydrogen, oxygen, nitrogen, and carbon together makeup over 99% of the mass of most cells. Biomolecules include both small as well as large molecules. The **small biomolecules** are low molecular weight (less than 1000) compounds which include sugars, fatty acids, amino acids, nucleotides, vitamins, hormones, neurotransmitters, primary and secondary metabolites. Sugars, fatty acids, amino acids, and nucleotides constitute the four major families of small biomolecules in cells. Each of these small biomolecules is composed of a small set of atoms linked to each other in a precise configuration through covalent bonds. **Large biomolecules** which have high molecular weight are called *macromolecules* and mostly are polymers of small biomolecules. These macromolecules are proteins, carbohydrates, and nucleic acids.

Small biomolecules	**Macromolecules**
Sugars	Polysaccharides
Amino acids	Polypeptides (proteins)
Nucleotides	Polynucleotides (nucleic acids)
Fatty acids	

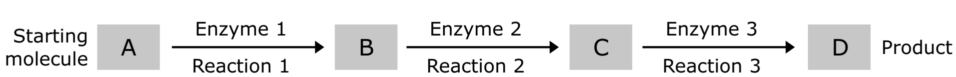
Nucleic acids and proteins are **informational macromolecules**. Proteins are polymers of amino acids and constitute the largest fraction (besides water) of cells. The nucleic acids, DNA and RNA, are polymers of nucleotides. They store, transmit, and translate genetic information. The polysaccharides, polymers of simple sugars, have two primary functions. They serve as energy-yielding fuel stores and as extracellular structural elements.

Chapter 3

Metabolism



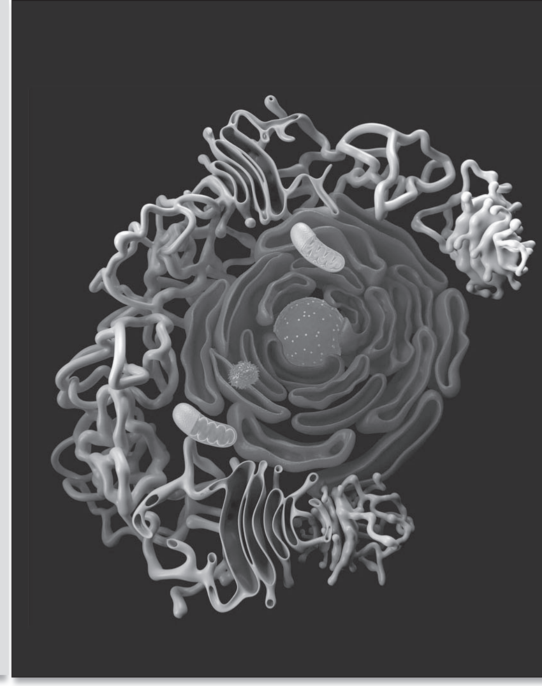
All cells function as biochemical factories. Within the living cell, biomolecules are constantly being synthesized and transformed into some other biomolecules. This synthesis and transformation constantly occur through enzyme-catalyzed chemical reactions. More than a thousand chemical reactions take place in a cell. Most of these chemical reactions do not occur in isolation but are always linked to some other reactions. All the interconnected chemical reactions occurring within a cell are called **metabolism** (derived from the Greek word for a *change*). Metabolism serves two fundamentally different purposes: 1. Generation of energy to drive vital functions and 2. Synthesis of biological molecules. The precursor is converted into a product during metabolic processes through a series of metabolic intermediates called **metabolites**. Cell metabolism is organized by enzymes. Enzyme-catalyzed reactions are connected in series so that the product of one reaction becomes the starting material, or substrate, for the next. The series of enzyme-catalyzed reactions transform substrates into end products through many specific chemical intermediates constitutes a **metabolic pathway**. Metabolism is sometimes referred to as **intermediary metabolism**. The term *intermediary metabolism* is often applied to the enzyme-catalyzed reactions that extract chemical energy from nutrient molecules and use it to synthesize and assemble cell components. The flow of metabolites through the metabolic pathway has a definite rate and direction. Metabolism is highly organized and regulated. Metabolic pathways are regulated through control of (1) the amounts of enzymes, (2) their catalytic activities, and (3) the availability of substrates. In multicellular organisms, the metabolic activities of different tissues are also regulated and integrated by growth factors and hormones that act from outside the cell.



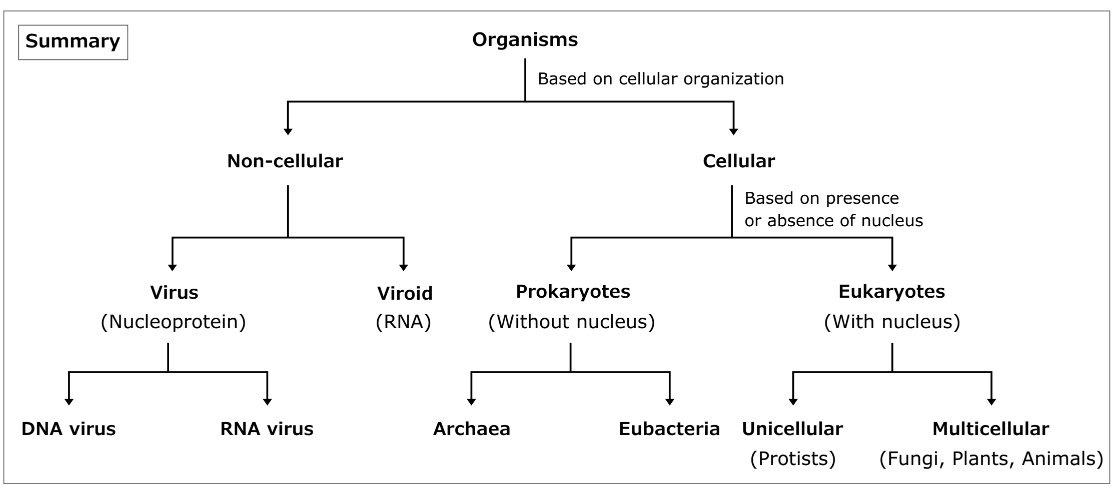
Metabolism consists of energy-yielding and energy-requiring reactions. The oxidation of carbon compounds is an important source of cellular energy. An energy currency common to all life forms, ATP, links energy-releasing pathways with energy-requiring pathways. ATP serves as the principal immediate donor of free energy in biological systems rather than as a long-term storage form of free energy.

Chapter 4

Cell Biology



A great diversity of organisms are present on the earth. These organisms can be classified into two broad categories- **cellular organisms** and **non-cellular organisms**. Cellular organisms belong to three distinct domains of life. These domains are bacteria and archaea and eukarya. Bacteria and archaea are prokaryotes. All eukaryotic organisms belong to domain eukarya which includes protists, fungi, plants and animals. Both prokaryotes and eukaryotes are cellular organisms. Viruses and viroids are non-cellular organisms because they lack cell or cell-like structure.

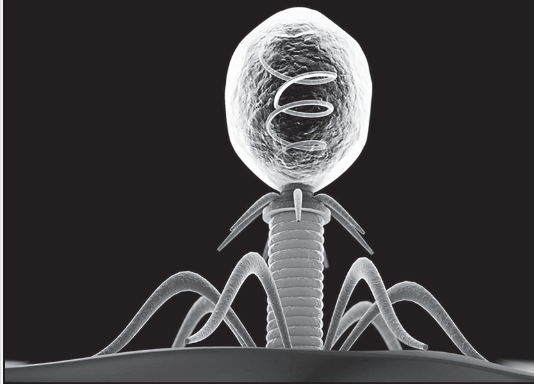


4.1 What is a cell?

The basic structural and functional unit of cellular organisms is the *cell*. It is an aqueous compartment bound by cell membrane, which is capable of independent existence and performing the essential functions of life. All cellular organisms consist of cell(s). Cellular organisms may be **unicellular** or **multicellular**. Unicellular organisms are made up of one cell. They are simple organisms and perform all functions in single cell. Most of prokaryotes are unicellular in nature. Multicellular organisms

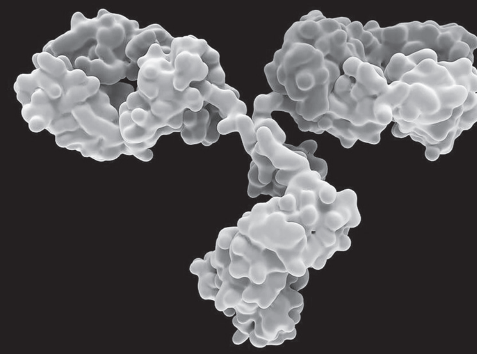
Chapter 5

Prokaryotes and Viruses



Prokaryotes (*pro* means before and *karyon* means kernel or nucleus) are cellular organisms that include two domains– **Bacteria** (sometimes referred to as *true bacteria* or *eubacteria*) and **archaea** (also termed as *archaebacteria* or *archaeobacteria*). The term *bacteria* or *eubacteria* refers to those that belong to the domain Bacteria, and the term *archaea* is used to refer to those that belong to domain Archaea. *The informal name 'bacteria' is occasionally used loosely in the literature to refer to all the prokaryotes, and care should be taken to interpret its meaning in any particular context*. Prokaryotic organisms are usually microscopic, single-celled organisms that have a relatively simple structure – neither nucleus nor unit membrane-bound organelles. Prokaryotes can be distinguished from eukaryotes in terms of their cell structure and molecular make-up. Prokaryotic cells have a simpler internal structure than eukaryotic cells. Although many structures are common to both cell types, some are unique to prokaryotes. Most prokaryotic cells lack extensive, complex internal membrane systems. The major distinguishing characteristics of prokaryotic and eukaryotic cells are as follows:

Feature	**Prokaryotic cells**	**Eukaryotic cells**
Membrane-bound nucleus	Absent	Present
DNA complexed with histone	Absent	Present
Number of chromosomes	One (mostly)	More than one
Mitosis and meiosis	Absent	Present
Sterol (in plasma membrane)	Absent, except *Mycoplasma*	Present
Ribosome	70S (cytosol)	80S (cytosol)
Unit-mem. bound organelle	Absent	Present
Cell wall	Present in *most* of prokaryotic cells. In eubacteria, it is made up of peptidoglycan.	Made up of cellulose in plant and chitin in fungi. Absent in animal cells.



Chapter 6

Immunology

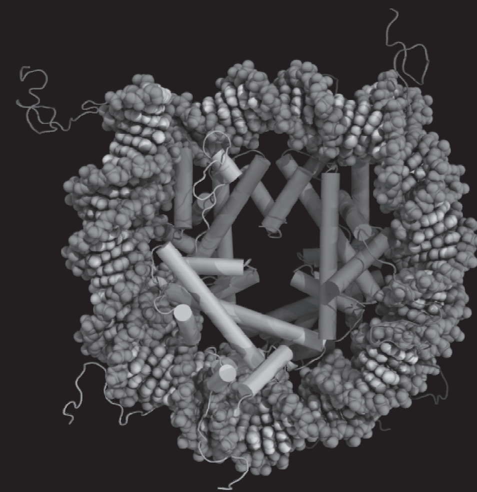
Immunology is the science that is concerned with immune response to foreign challenges or simply, study of the body's defense against infection. It addresses the questions such as how does the body defend itself against infection, when an infection does occur, how does the body eliminate the pathogens and how does long-lasting immunity to many infectious diseases develop? The ability of an organism to resist infections by pathogens or state of protection against foreign organisms or substances is called **immunity** (derived from Latin term *immunis*, meaning 'exempt'). The array of cells, tissues and organs which carry out this activity constitute the **immune system**. The immune response is a complex process and is divided into two categories — **innate** (or **native**) and **adaptive** (or **acquired**) immunity. *Innate immunity* is a general, non-specific immune response which presents in all individuals at all times. In contrast to innate immunity, *adaptive immunity* is highly specific to the particular pathogen that induced it. It develops during the lifetime of an individual as a response to infection and adaptation to the infection. Thus, when a given pathogen is new to the host, it is initially recognized by the innate immune system and then the adaptive immune response is activated. Innate immunity is the most ancient form of defense, found in most multicellular organisms, while adaptive immunity is a recent evolutionary phenomenon, having arisen in vertebrates. Thus, vertebrates are protected by both innate immunity and adaptive immunity.

6.1 Innate immunity

Innate immunity is present since birth, evolutionarily primitive and is relatively nonspecific. It provides the *early defense* against pathogens, before adaptive immune responses can develop. It is not specific to any one pathogen but rather acts against all foreign molecules and pathogens. It also does not rely on previous exposure to a pathogen and response is functional since birth and has no memory.

Components of the innate immune system

An organism can adopt three strategies to deal with the threat posed by pathogens (the agents that are capable of causing infectious diseases): *avoidance*, *resistance* and *tolerance*. **Avoidance**



Chapter 7

Genetics

All living organisms reproduce. Reproduction results in the formation of offspring of the same kind. However, the resulting offsprings need not and, most often, do not completely resemble the parents. Several characteristics may differ between individuals belonging to the same species. These differences are termed **variations**. The mechanism of transmission of characters, resemblances, and differences from the parental generation to the offspring is called **heredity**. The scientific study of heredity and variations is known as **genetics** (from the Greek word genno = give birth). The word 'genetics' was first suggested by prominent British scientist William Bateson. Genetics can be divided into three areas: *classical genetics, molecular genetics*, and *evolutionary genetics*. **Classical genetics** is concerned with the basic principles of heredity and how traits are passed from one generation to the next. It also addresses the relationship between chromosomes and heredity and the arrangement of genes on chromosomes. **Molecular genetics** covers the chemical nature of the gene and how genetic information is replicated and expressed, i.e., cellular processes of replication, transcription, and translation. **Evolutionary genetics** is the study of how genetic variation leads to evolutionary change. It is concerned with the evolution of genome structure, the genetic basis of speciation and adaptation, and genetic change in response to evolutionary processes such as natural selection, genetic drift, mutation, and gene flow in populations.

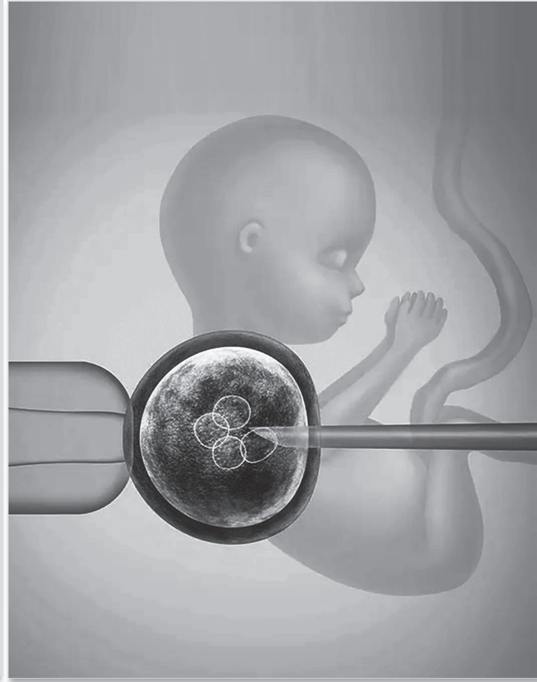
Classical genetics

7.1 Mendel's principles

Gregor Johann Mendel (1822–1884), known as the *father of genetics*, was an Austrian monk. He conducted a series of experiments using pea plants and showed that traits are passed from parents to offspring in predictable ways. By quantitative data analysis of results, he concluded that each trait in the pea plant is controlled by a pair of factors and that during gamete formation, members of a gene pair separate from each other. In 1865, he published the results of *hybridization experiments* titled *Experiments on Plant Hybridization* in a journal '*The proceeding of the Brunn*

Chapter 8

Recombinant DNA technology



Recombinant DNA technology (also known as genetic engineering) is the set of techniques that enable the DNA from different sources to be identified, isolated and recombined so that new characteristics can be introduced into an organism. The invention of recombinant DNA technology—the way in which genetic material from one organism is artificially introduced into the genome of another organism and then replicated and expressed by that other organism—was largely the work of Paul Berg, Herbert W. Boyer and Stanley N. Cohen, although many other scientists also made important contributions to the new technology as well. Paul Berg developed the first recombinant DNA molecules that combined DNA from the SV40 virus and lambda phage. Later in 1973, Herbert Boyer and Stanley Cohen develop recombinant DNA technology, showing that genetically engineered DNA molecules may be cloned in foreign cells.

One important aspect of recombinant DNA technology is **DNA cloning**. It is a set of techniques that are used to design recombinant DNA molecules and to direct their replication within host organisms. The use of the word 'cloning' refers to the method used to generate identical DNA molecules.

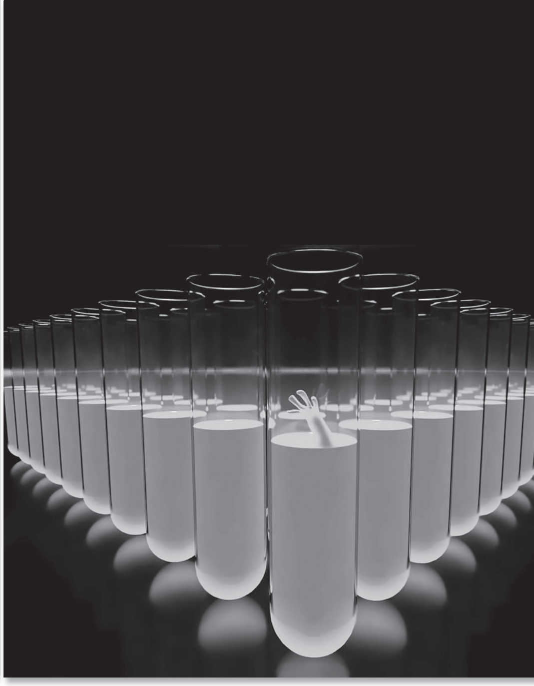
8.1 DNA cloning

DNA cloning is the production of a large number of identical DNA molecules from a single ancestral DNA molecule. The essential characteristic of DNA cloning is that the desired DNA fragments must be *selectively amplified,* resulting in a large increase in copy number of selected DNA sequences. In practice, this involves multiple rounds of DNA replication catalyzed by a DNA polymerase acting on one or more types of the template DNA molecule. Essentially two different DNA cloning approaches are used: *Cell-based* and *cell-free DNA cloning*.

Cell-based DNA cloning: This was the first form of DNA cloning to be developed and is an *in vivo* cloning method. The first step in this approach involves attaching foreign DNA fragments *in vitro* to DNA sequences, capable of independent replication. The recombinant DNA fragments are then transferred into suitable host cells where they can be propagated selectively. The essence of cell-based DNA cloning involves the following steps:

Chapter 9

Bioprocess Engineering



Bioprocess engineering is a specialization of chemical engineering that deals with the design and development of equipment and processes for the manufacturing of products such as food, pharmaceuticals and polymers from biological materials. It uses the capabilities of organisms in industrial, medical, environmental or agricultural processes in order to produce useful biological materials. Application of bioprocess engineering includes:

- Design and operation of fermentation systems,
- Development of food processing systems,
- Application and testing of product separation technologies,
- Design of instrumentation to monitor and
- Control biological processes and much more.

Bioprocess engineers work at the frontiers of biological and engineering sciences to *bring engineering to Life* through the conversion of biological materials into other forms needed by mankind. One of the main tasks of a bioprocess engineer is to control and maintenance of a biological processes such as the production of beverages, pharmaceuticals, antibiotics, enzymes, biochemicals, food processing and biological waste treatment. These processes require a well-designed growth environment to obtain the maximum yield of the product and consequently, these conditions need to be carefully controlled. Environmental design comprises the determination of the environment of the process, while fermentation engineering provides the means for meeting those requirements.

9.1 Concept of material and energy balance

Concepts of material (or mass) and energy balance is important in process engineering. The term 'balance' implies that the masses and energy entering and leaving the system should be equal. The *law of conservation of mass* provides the theoretical framework for mass balances. Similarly, the principle underlying all energy balance calculations is the *law of conservation of energy*, which states that energy can be neither created nor destroyed.