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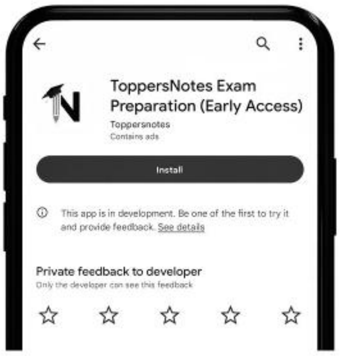

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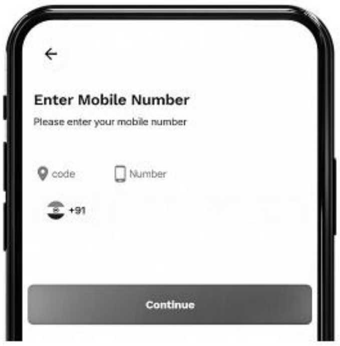

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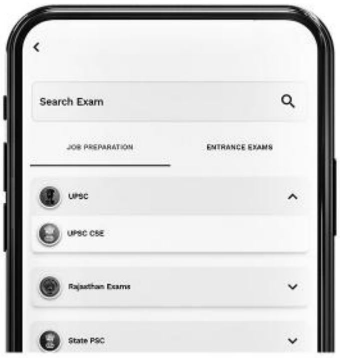

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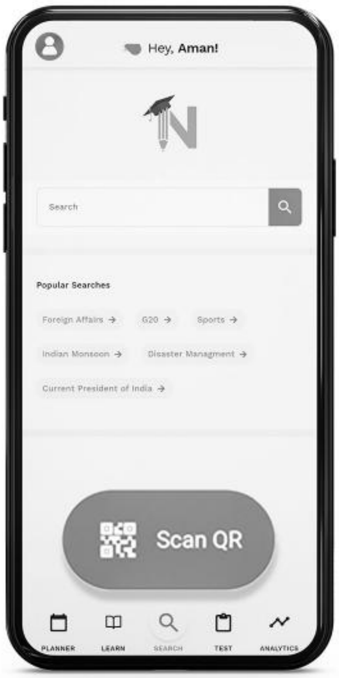

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CHAPTER 1

Biomolecules

CHAPTER OUTLINE

- The Tissues Introduction
- Lipids
- Polysaccharides
- Metabolism
- Enzymes
- Types of Biomolecules
- Proteins
- Nucleic Acids (DNA and RNA)
- Living State

INTRODUCTION

- Biomolecules form the basic structural constituents of a living cell.
- They include organic and inorganic compounds.

Major complex biomolecules of cells and their functions

Biomolecules	Building block	Functions
	Carbohydrates	
Polysaccharide (glycogen)	Monosaccharide (glucose)	Storage
Proteins	Amino acids	Fundamental basis of structure and function of cell.
Lipids	Fatty acids and glycerol	Storage and structural components of membrane
	Nucleic acids	
Deoxyribo – nucleic acid	Deoxyribonuc – leotides	Transmission of hereditary information
Ribonucleic acid	Ribonucleotides	Protein biosynthesis

Average Composition of Cells

- Water: 70-90%
- Protein: 10-15%
- Carbohydrates: 3%
- Lipids: 2%
- Nucleic acids: $5-7\%$
- Ions: 1%

Analysis of Chemical Composition

- Chemical analysis is used to determine the sorts of organic molecules (carbon-containing compounds) found in biological tissues.
- Obtaining living tissue
- Slurry was obtained by grinding in trichloroacetic acid.
- **Filtrate** (acid-soluble fraction containing biomacromolecules) and **retentate** are obtained from the slurry.
- Thousands of organic substances were discovered in the filtrate.
- Separation techniques that are used to separate one component from another.
- Analytical techniques were used to discover the molecular formula and likely structure.
- Biomolecules are all carbon-containing compounds.

Analysis of Inorganic Compounds

- Living tissue was extracted.
- It is dried to remove all of the water, and the remaining material provides the dry weight.
- The dry material is incinerated.
- All organic and gaseous compounds will be eliminated, leaving only "ash".
- Many inorganic elements, such as Ca, Mg, S, P, and others, as well as inorganic compounds ($SO_4{}^{2-}$, $PO_4{}^{3-}$ etc), etc., are found in ash.

Primary and Secondary Metabolites

PRIMARY METABOLITES VERSUS SECONDARY METABOLITES

Primary metabolites	Secondary metabolites
Primary metabolites perform physiological functions in the body like intrinsic functions	Secondary metabolites are derivatives of primary metabolites
Directly involved in the growth, development, and reproduction	Play a role in ecological functions
Formed during the growth phase due to the energy metabolism	Formed near the stationary phase of growth
Produced in large quantities and extraction is easy	Produced in small quantities and extraction is difficult
Same in all plants	Unique to different plant species
A part of the basic molecular structure of an organism	Not a part of the basic molecular structure of an organism
Ethanol, lactic acid, nucleotides and vitamins are examples	Pigments, antibiotics, and drugs are examples

Examples of Secondary Metabolite

- Pigments: Carotenoids, Anthocyanins etc.
- Alkaloids: Morphine, Codeine etc.
- Terpenoides: Monoterpenes, Diterpenes etc.
- Essential oils: Lemon grass oil etc.
- Toxins: Abrin, Ricin etc.
- Lectins: Concanavalin A.
- Drugs: Vinblastin, curcumin etc.
- Polymeric substances: Rubber, gums, cellulose etc.

TYPES OF BIOMOLECULES

Based on the molecular weight and solubility, biomolecules are divided into **two categories**: **Biomicromolecules and biomacromolecules**

I. Biomicromolecules

- They are simple small-molecule compounds with a low molecular weight (less than 1000 Da), great solubility, and a simple shape.
- They are discovered in acid soluble pools.
- It consists of amino acids, carbohydrates, nitrogen bases, lipids, and other substances.

Amino acids

- Amino acids are chemical molecules that serve as the foundation for proteins.
- Nature contains up to 300 amino acids. Only 20 of these are known as standard amino acids, which are typically found in proteins.
- Protein and amino acid incorporation is governed by DNA/mRNA triplet codes.
- Non-coded amino acids can also be found in proteins. These amino acids are known as rare amino acids.
- Through changes, rare amino acids are produced from coded amino acids. For example, hydroxyproline from proline, hydroxylysine from lysine, and so on.
- Amino acid structure and properties: A typical amino acid is composed of an amino group (-NH_2), an acid group (-COOH), a hydrogen atom (H), and a variable group I. The amino group (-NH_2) is basic in nature, whereas the carboxyl group (-COOH) is acidic, and both are connected to the same carbon atom (α -carbon).
- They are represented by the general formula:

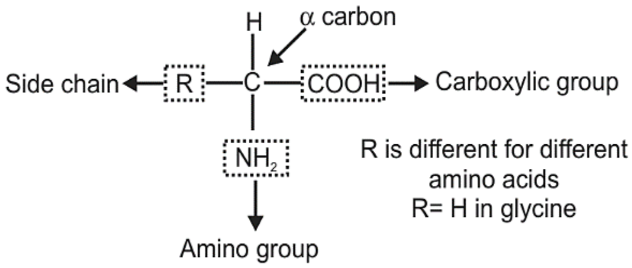


Figure: α-amino acid

- Based on R group, there are 21 amino acids.
- When R is H, Glycine. Glycine is the simplest amine acid.
- When R is CH_3, alanine.
- The chemical and physical properties of amino acids depend upon the amino group, carboxyl group and R group.
  - More carboxyl group- acidic amino acid
  - More amino group-basic amino acid
  - Equal amino and carboxylic group- neutral amino acid.
- Amphoteric substances are amino acids. They have both a basic and an acidic group.
- Some amino acids have ionizable -NH_2 and -COOH groups. As a result, the structure of amino acids alters in different pH solutions.

$$H_3^+N-\overset{R}{\overset{|}{C}}H-COOH \rightleftarrows H_3^+N-\overset{R}{\overset{|}{C}}H-COO^- \rightleftarrows H_2N-\overset{R}{\overset{|}{C}}H-COO^-$$

Zwitter ionic form

Isoelectric Point

- The amino acid's isoelectric point is defined as the point at which a molecule exists as a zwitter ion with no net charge.
- As a result, the molecule is electrically neutral at this location, with maximum solubility and the least buffer capacity. The isoelectric point (PI) of all amino acids is not the same.
- The nature of the 2olymeriz groups of amino acids determines the PI value of protein.
- PI can be computed by obtaining the average pKa values of 2olymeriz groups.

Amino acids are classified into seven categories based on their structure and reaction:

(i) **Acidic amino acids**: Amino acids with an additional carboxyl group. Glutamate (glutamic acid), for example, and aspartate (aspartic acid).

(ii) **Basic amino acids**: Amino acids containing an extra amino group that do not produce amides. For example, arginine and lysine.

(iii) Neutral amino acid: A non-cyclic hydrocarbon chain amino acid containing one amino group and one carboxylic group. For example, glycine, alanine, and valine.

(iv) Sulphur-containing amino acids: Sulphur-containing amino acids. Cysteine and methionine are two examples.

(v) Alcoholic amino acids are amino acids with an alcoholic or hydroxyl group. Serine and threonine are two examples.

(vi) Aromatic amino acids are cyclic amino acids with a straight side chain including carboxylic and amino groups. For example, phenylalanine, tryptophan, or tyrosine.

(vii) Heterocyclic amino acid: An amino acid with a nitrogen ring structure. For example, histidine and proline.

Based on nutritional requirements, amino acids are grouped into two classes:

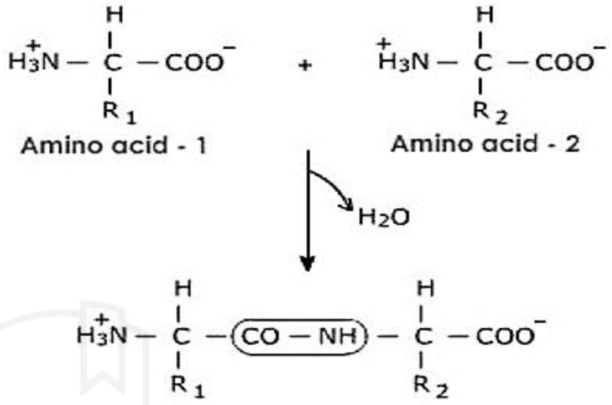
(a) Essential amino acids

(b) Non-essential amino acids

Essential amino acids	**Non-essential amino acids**
Amino acids which cannot be synthesized by the body and therefore, need to be supplied through the diet.	Amino acids that can be synthesized in our body to meet the biological needs.
<u>Examples</u>: Arginine, valine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, and tryptophan Arginine and histidine are called semi-essential amino acids as they can be partly synthesized in our body.	<u>Examples</u>: Glycine, alanine, serine, cysteine, aspartic acid, glutamic acid, aspargine, glutamine, tyrosine, and proline

Peptide Bond Formation

- When two amino acids mix, one amino acid's amino group unites with the carboxyl group of the second amino acid.
- This results in the creation of peptide bonds.
- The interaction of one molecule's amino group and another's carboxyl group results in the removal of a water molecule and the formation of an amide (-CO-NH-) bond.



Polypeptides

- Polypeptides are molecules that contain more than ten amino acids. They are created by the sequential ordering of amino acids.
- Oligopeptides are relatively shorter peptides, whereas polypeptides are longer polymers.
- Proteins are polypeptides with a molecular mass more than 10,000 that include more than 100 amino acids.

LIPIDS

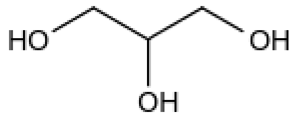
- **Lipids** are **esters** of **fatty acids** with alcohol that are insoluble in water but soluble in a variety of nonpolar organic solvents such as ether, benzene, and chloroform.
- Lipids form **colloidal complexes** and disperse uniformly in water as minute droplets.
- The **complex** is referred to as an **emulsion**, and the **basic components** of **all lipids** are **fatty acids**.

Fatty acids

- Fatty acids are organic acids that have hydrocarbon chains that end in a -COOH group that is connected to an R-group.

- The R group could be methyl ($-CH_3$), ethyl ($-C_2H_5$) (1C to 19C)
  - Palmitic acid, for example, has 16 carbons ($CH_3-(CH_2)_{14}COOH$)
  - Arachidonic acid has 20 carbons.
- Lipids contain fatty acids as well as glycerol.

Structure of glycerol (trihydroxy propane)



- Lipids are classified as monoglycerides, diglycerides, or triglycerides based on the number of glycerols.
- Triglyceride is a glyceride that is made up of a single molecule of glycerol that has been esterified with three fatty acids. It's mostly found in vegetable oils and animal fat.

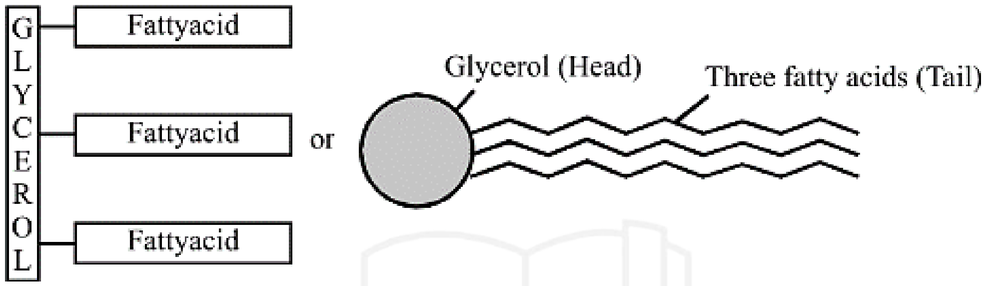
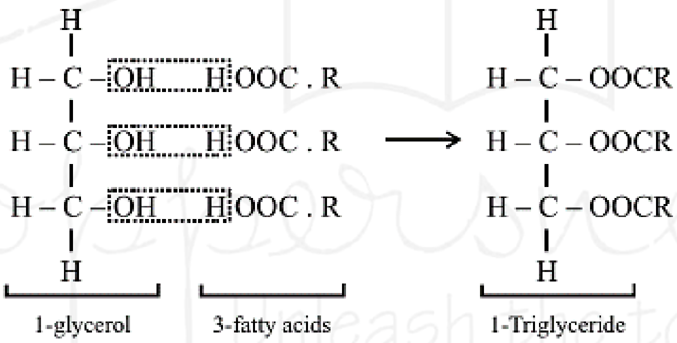


Fig. 9.2 : Triglyceride (neutral fat) molecule



- Fatty acids are of two types:

Saturated Fats	**Unsaturated Fats**
Contains a single bond.	Contains at least one double bond.
Not to be consumed more than 10 percent of total calories per day.	Not to be consumed more than 30 percent of total calories per day.
Excessive consumption leads to heart diseases.	Good for consumption, but excessive may increase cholesterol.
Increases low-density lipoproteins (LDL), which is called as bad cholesterol.	Increases High-density lipoprotein (HDL), which is commonly known as good cholesterol and also reduce low-density lipoproteins (LDL).
Would not spoil quickly.	Spoil quickly.
Foods sources of saturated fats are whole milk, butter, cheese, margarine, coconut oil, vegetable oil, meat, peanut, fried foods, etc.	Foods sources of unsaturated fats are walnuts, flax, avocado, sunflower oil, soybean oil, fish oil, canola oil, red meat, etc.
High melting point.	Low melting point.
Solid state in room temperature.	Liquid state in room temperature.

- Saturated fats have general formula of $C_nH_{2n}O_2$. E.g. Palmitic acid, stearic acid
- Unsaturated Fatty acids: The general formula is $C_nH_{2n-2}O_2$.
  E.g.
  - Oleic acid (with one double bonds, $C_{18}H_{34}O_2$)
  - Linoleic acid (with double bonds, $C_{18}H_{32}O_2$)
  - Linolenic acid (with three double bonds, $C_{18}H_{30}O_2$)
  - Arachidonic acid (with four double bonds, $C_{20}H_{32}O_2$)

Classification of Lipids

Lipids are classified into three types:

(a) Simple lipids
(b) Compound lipid
(c) Derived lipids

(a) Simple fats: These are fatty acid esters with various alcohols. They are divided further into fats and waxes.

(i) Fats: Fats are glycerol esters of fatty acids. The liquid form of a fat is known as oil. Simple lipids are referred to as fats in mammals and oils in plants.
Example: Triacyl glycerol

(ii) Waxes: Waxes are long chain monohydric alcohol fatty acid esters.
Examples: Cholesterol ester, myricyl palmitate, and cetyl palmitate

(b) Compound lipids: These are fatty acid esters with alcohol that contain other groups in addition to the alcohol and the fatty acid. They are further classified as follows:

(i) Phospholipids (Phosphatides): Phospholipids (Phosphatides) are esters of fatty acids with glycerol that contain an esterified phosphoric acid and a nitrogen base. These lipids are abundant in nerve tissue, brain, liver, kidney, pancreas, and heart.

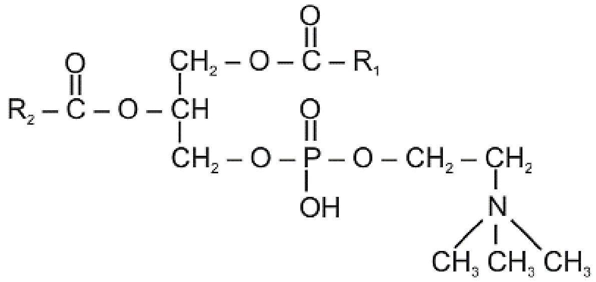


Figure: Phospholipid (Lecithin)

(ii) Glycolipids are lipids that contain a carbohydrate moiety as well as an amino alcohol.

(iii) Lipoproteins – Lipoproteins are formed when lipids such as triacyl glycerol, phospholipids, cholesterol and cholesteryl esters, and free fatty acids mix in specific proportions with protein to create a hydrophilic lipoprotein complex.
Examples: Chylomicrons, very low-density lipoprotein (VLDL), low density lipoprotein (LDL), and high density lipoprotein (HDL). The protein moiety of lipoprotein is referred to as apoprotein.

(c) Derived lipids: These are lipids that are created by hydrolyzing simple and compound lipids. They are either lipid-like substances (such as sterols) or lipid derivatives (such as terpenes and prostaglandins)

Functions of Fats

- Fats stored in adipose tissue provide an efficient source of energy.
- Fats serve as a thermal insulator in the subcutaneous tissue and around the internal organs.
- They also act as an electrical insulator against nerve impulse transmission.
- Cholesterol and phospholipids are essential components of cell membranes.
- Lipoproteins and glycolipids preserve the integrity and permeability of cells.

- Fats are the primary source of fat-soluble vitamins.
- Platelet phosphatides are involved in the mechanism of blood coagulation.

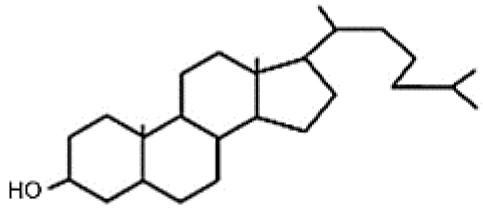


Figure: Cholestrol

SUGARS (CARBOHYDRATES)

- Carbohydrates that are sweet and water-soluble are sugars.
- They consist of C, H, and O in the proportions 1:2:1.
- Examples: Glucose, Ribose etc.

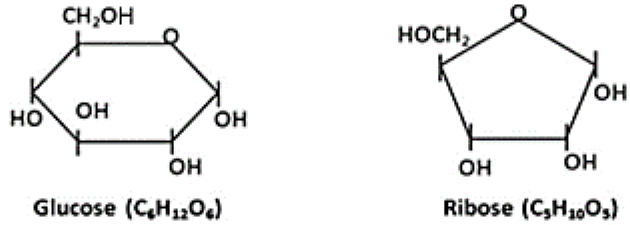


Figure: Sugars (Carbohydrates)

Nitrogenous bases

- Nitrogenous bases are carbon compounds with heterocyclic rings.
- Examples: Adenine, Thiamine, Guanine.
- It includes purines and pyrimidines.
  Purine = Adenine (A) and Guanine (G)
- Pyrimidines = Cytosine (C), Thymine (T) and Uracil (U)

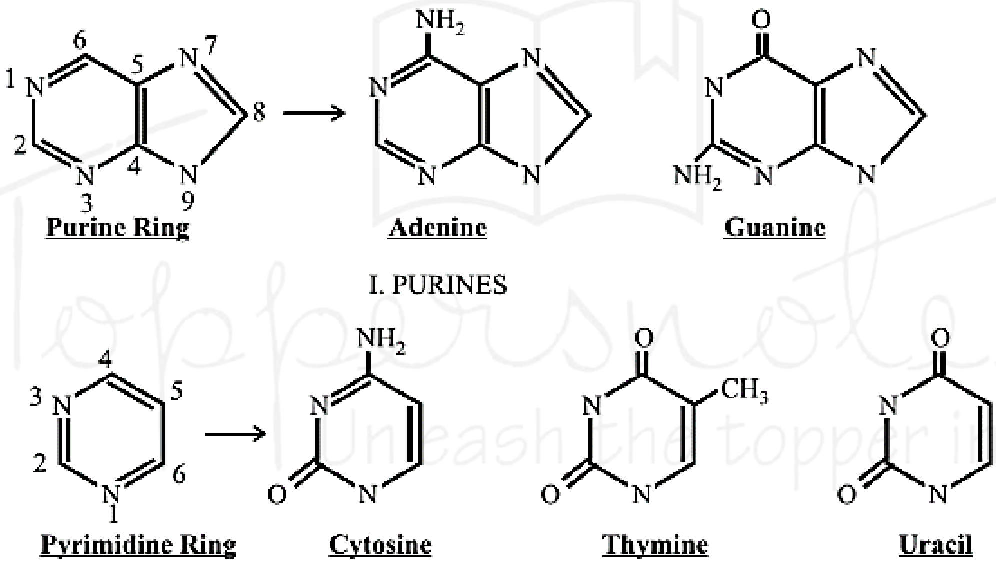


Figure: II. Pyrimidines

Nucleoside and nucleotide

Nucleoside: Nitrogen base + Sugar
Adenine + Sugar = Adenosine
Guanine + Sugar = Guanosine
Cytosine + Sugar = Cytidine
Thymine + Sugar = Thymidine
Uracil + Sugar = Uridine

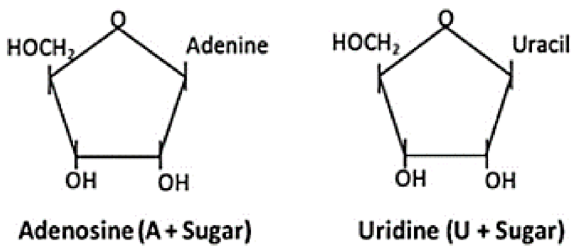


Figure: Nucleosides

Nucleotide: Nitrogen base + Sugar + phosphate.
Adenine + sugar + phosphate = Adenylic acid
Guanine + sugar + phosphate = Guanylic acid
Cytosine + sugar + phosphate = Cytidylic acid
Thymine + sugar + phosphate = Thymidylic acid
Uracil + sugar + phosphate = Uridylic acid

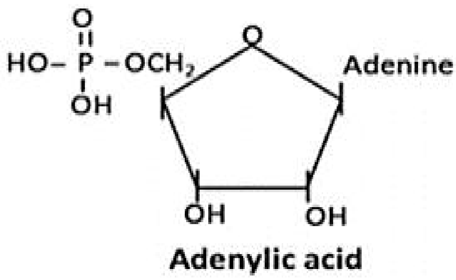


Figure: Nucleotide

- Nucleotides are heterocyclic compounds.
- Nucleic acids (DNA & RNA) are made up of nucleotides.

II. Biomacromolecules

- Biomacromolecules are large, complex chemicals with a high molecular weight (greater than 1000Da, with the exception of lipids), limited solubility, and intricate conformation.
- They exist in the acid-insoluble fraction. They produce colloidal complexes typically and are always organic.
- Acid insoluble fraction includes
  - Proteins
  - Nucleic acids
  - Polysaccharides
  - Lipids
- The lipids' molecular weight does not exceed 800Da. However, it belongs to the acid-insoluble fraction because lipids are structured like cell membranes.
- When a tissue is ground, these membranes rupture and form water-insoluble vesicles; therefore, lipids are not strictly macromolecules.
- The **acid insoluble fraction** contains cytoplasmic and organelle macromolecules.

PROTEINS

- Proteins are polymers with a high molecular weight.
- Proteins are linear heteropolymers of amino acids that are folded in many different ways.
- The linear amino acid polymers are known as polypeptides. Peptide bonds interconnect the linear chains of amino acids.
- A protein composed of two or more polypeptides is termed multimeric.
- Peptide bond is a covalent bond formed when the -COOH group of one amino acid reacts with the $-NH_2$ group of the subsequent amino acid by dehydrating (releasing a molecule of water).

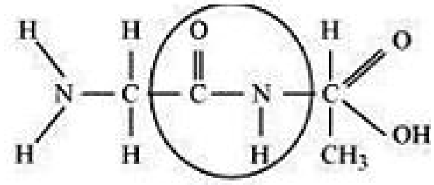


Figure: Peptide bond

- Proteins are the most abundant organic molecules of the living system and form the fundamental basis of structure and function of life.
- They contain carbon, hydrogen, nitrogen, oxygen and polymer.

Structure of Protein

Proteins are macromolecules composed of amino acids that have been polymerized. Proteins are divided into four structural levels.

(a) Primary structure: In a polypeptide chain, it is the linear sequence of amino acids. It describes the sequence of amino acids, or the positional information contained within a protein. The chain's left end contains the first amino acid (N-terminal amino acid). Right end has last amino acid (C-terminal amino acid).

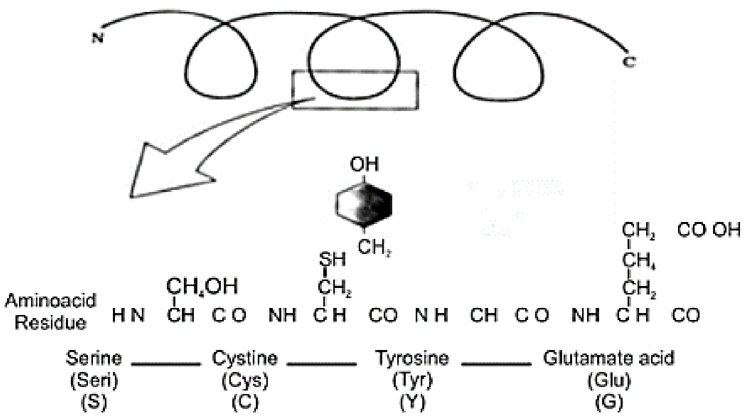


Figure: Primary structure of a portion of a hypothetical protein

(b) Secondary structure:

- The polypeptide chain is coiled to create a three-dimensional secondary structure. Only right handed helices are present.
- There are three different forms of secondary structures: the -helix, the -pleated structure, and the collagen helix.
  (i) In **α-helix**, the polypeptide chain is right-handedly spirally wound. Hydrogen bonds between two amino acids help to stabilize the helix. Such as keratin, myosin, the epidermis, and fibrin.
  (ii) In **β -pleated secondary structure**, two or more polypetides are hydrogen-bonded together. In place of a fiber or filament, a sheet is produced in α -helix. β -keratin, silk fibroin, etc.
  Three stands or polypeptides are entwined around one another in collagen helix.

(c) Tertiary Structure

- The helical polypeptide chain is further wound and folded to create a complex tertiary structure. It provides a three-dimensional view of proteins. Proteins require tertiary structure for many biological activities.
- During the coiling of polypeptide, various types of bonds are observed. There are covalent bonds, ionic or electrostatic bonds, hydrogen bonds, van der Waals interactions, and hydrophobic bonds.

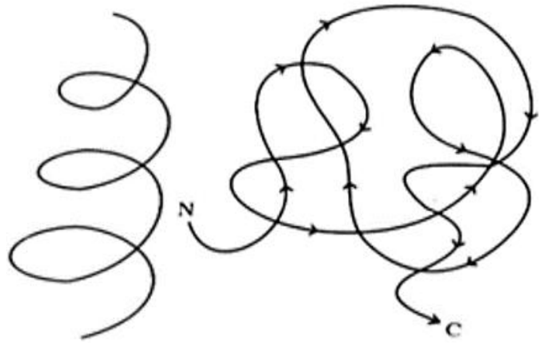


Figure: (a) Secondary structure (b) Tertiary structure

Tertiary structure bonds are readily shattered by high temperature, drastic changes in pH, and high-energy radiation. Denaturation is the process of tertiary structure disintegration.

(d) Quaternary structure: The quaternary structure is composed of multiple polypeptide chains. For instance, Hb has four subunits (2 α subunits and 2 β subunits).

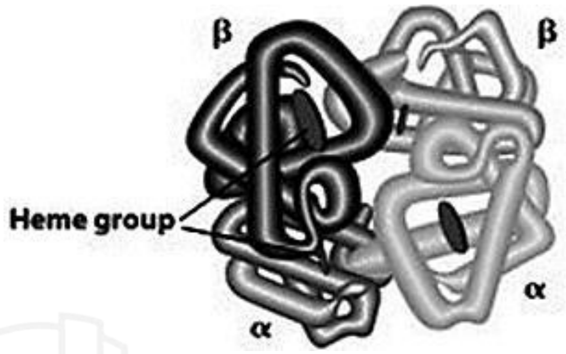


Figure: Quaternary structure

Types of Proteins

On the basis of their molecular shape

Fibrous protein	**Globular proteins**
Thread like molecules, which lie side by side to form fibres.	Molecules are folded into compact units to form spheroidal shapes
Held together by hydrogen bonds.	Spherical in shape.
Generally insoluble in water.	Soluble in water.
Examples: Fibroin in silk, collagen in tendons, α – keratin in skin, hair, nails etc.	Example: Haemoglobin

On the basis of constitution

Simple proteins	Conjugated proteins	Derived proteins
Made up of amino acids only. On hydrolysis yield only α-amino acids.	Proteins composed of simple proteins combined with non-protein part called as prosthetic groups.	Proteins derived from the simple and conjugated proteins by the action of acids, alkalies or enzymes.
Examples: Albumin, globulin.		Examples: Proteoses, peptones and peptides.

Functions of Protein

- Proteins are important for growth and tissue repair.
- They aid in the transport of nutrients across cell membranes (e.g., GLUT-4 enables glucose transport into the cell).
- They serve as intercellular ground substance (for instance, collagen).
- They function as antibodies to combat infectious organisms. They function as receptors (such as receptors for scent, taste, and hormones).
- Some are hormones (such as insulin) that regulate a variety of physiological processes.
- Enzymes are biocatalyst-functioning proteins.
- Proteins, such as trypsin, contribute to blood coagulation via thrombin, fibrinogen, and other protein factors.

POLYSACCHARIDES

- Polysaccharides are condensation polymers in which monosaccharides are bound together by **glycosidic linkage**. Thus, they are known as **nonsugars**.
- They serve two essential functions: structural and energy storage.
- Glycosidic bond in polysaccharides: the bond formed when individual monosaccharides are linked by dehydration between two carbon atoms.
- Normal formation occurs between carbon atoms 1 and 4 of adjacent monosaccharide units.

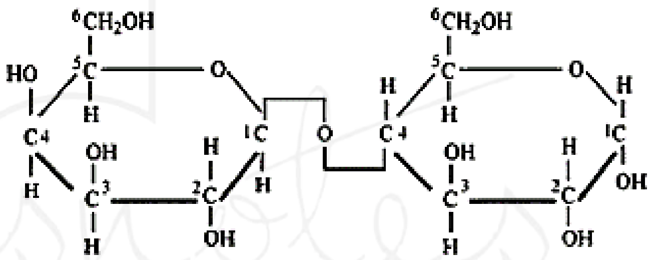


Figure: Glycosidic bond

- The right end of a polysaccharide is known as the reducing end, while the left end is referred to as the non-reducing end.

Polysaccharides are of two types based on their composition:
(a) Homopolysaccharides
(b) Heteropolysaccharides

Homopolysaccharides	Heteropolysaccharides
Carbohydrates which are formed by 1olymerization of only one type of monosaccharide monomers.	Carbohydrates which are formed by condensation of two or more monosaccharides or their derivatives.
<u>Examples</u>: Starch (polymer of glucose), Cellulose (polymer of glucose), Inulin (polymer of fructose), Glycogen (polymer of glucose)	Examples: Glucosamine, N-acetyl galactosamine, chitin (Exoskeleton of arthropods)

Polysaccharides are of three main types: **Storage, structural** and **mucopolysaccharides**.

(a) Food storage polysaccharides: These polysaccharides are utilized as dietary reserves. E.g. glycogen and starch

(i) Glycogen: α – D Glucose polymerizes to form glycogen. It is an animal storage polysaccharide. It is specifically known as animal starch. Mostly stored in the liver and the muscles.

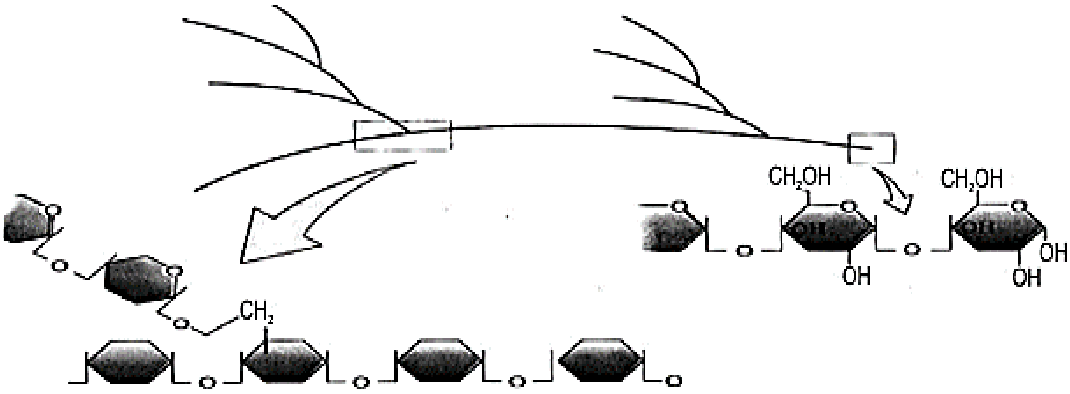


Figure: Diagrammatic representation of a portion of glycogen

(ii) Starch: It consists of amylase and amylopectin heteropolymers. It acts as a source of energy storage for plants.

Starch forms helical secondary structures and can contain iodine molecules within the helical portion; the blue starch-iodine complex forms the basis of the confirmatory test for starch detection.

(iii) Inulin: It is the fructose polymer.

(b) Structural Polysaccharide: These polysaccharides contribute to the formation of the structural framework of the cell walls of plants and the skeletons of animals. Chitin and cellulose are examples.

(i) Cellulose is the glucose homopolymer. It comprises the plant cell wall, paper pulp, and cotton fibers, among others. Cellulose chains are unbranched and linear. The successive glucose molecules are connected via 1- 4β bonds.

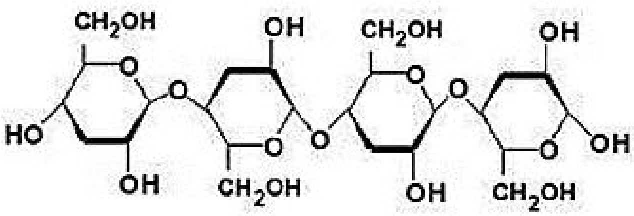


Figure: Structure of cellulose

(ii) Chitin: It is the second most prevalent organic substance. It is a heteropolysaccharide that is a structural component of fungal cell walls and arthropod exoskeletons. Not glucose but nitrogen-containing glucose derivatives known as N-acetyl glucosamine is the fundamental unit of chitin. Monomers are connected via 1-4 β linkages.

(c) Mucopolysaccharides are viscous substances containing acidic or amino-terminated polysaccharides derived from galactose, mannose sugar derivatives, and uronic acids. These substances are called glycosamino glycans (GAG). Hyaluronic acid, chondroitin sulfate, and heparin are essential mucopolysaccharides.

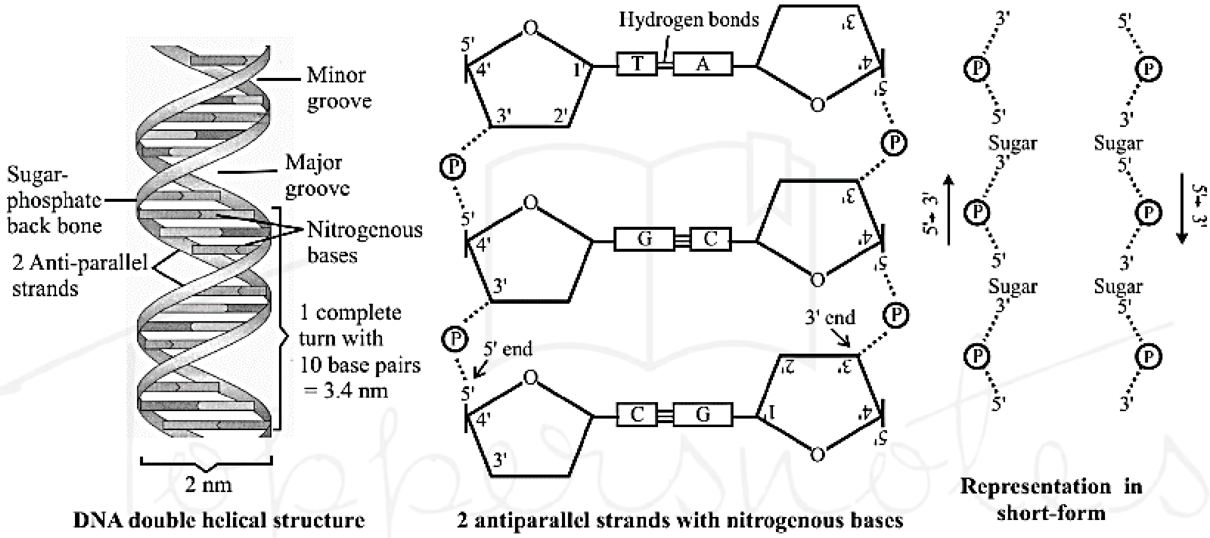
They are found within the cell walls of plants, outside the cells of bacteria, blue-green algae, and numerous aquatic organisms. Mucopolysaccharide functions as a layer of cement between cells, connective tissues, and cartilages.

NUCLEIC ACIDS (DNA AND RNA)

- Nucleic acids are composed of nucleotide polymers.

- In addition to a phosphate group, a nucleotide contains a sugar and heterocyclic nitrogenous bases.
- There are two varieties of nitrogenous bases:
  - Adenine and guanine are purines.
  - The three pyrimidines are thiamine, cytosine, and uracil.
- Sugar comes in two forms:
  - RNA is composed of ribose, which has an additional oxygen atom at the carbon -2 position.
  - Deoxyribose is what makes up DNA.
- A phosphate molecule connects the 3'-carbon atom of one sugar nucleotide to the 5'-carbon atom of the sugar nucleotide that follows.
- The phosphodiester bond is a covalent bind between phosphate and two sugar molecules. These compounds constitute the sugar-phosphate backbone of nucleic acids.
- DNA and RNA are both forms of nucleic acid.

Secondary Structure of DNA (Watson – Crick Model)



Salient features of the Watson and Crick model are as follows:

- DNA exists as a double helix. The 2 polynucleotide strands are arranged anti-parallely (one to 5 'to 3'and the other is 3′ to 5′).
- The backbone of DNA is formed by the sugar-phosphate chain.
- Nitrogen base pairs form the steps of DNA.
- Nitrogen bases include Adenine (A), Guanine (G), Thymine (T) and Cytosine (C).
- A pairs with $T (A = T)$ by 2 hydrogen bonds.
- G pairs with $C (G \equiv C)$ by 3 hydrogen bonds.
- One full turn of helical strand have 10 steps (10 base pairs)
- Length of one full turn is 34Å (i.e. 3.4Å for each step).
- At each step the strand turns 36°(360° for a full turn).
- The two polynucleotide chains are 20Å apart.

METABOLISM

- Metabolism refers to the sum of all biochemical reactions occurring within a biological system.
  - CO_2 removal from amino acids to produce amine
  - Elimination of the amino group from a nucleotide base
  - Hydrolysis of a glycosidic bond

- Metabolic pathways are a series of linked reactions (multistep chemical reactions) that occur during metabolism.
- Metabolic pathways are comparable to urban automobile traffic.
- Similar to automobile traffic, the flow of metabolites through metabolic pathways has a distinct rate and direction. This metabolic flux is referred to as the dynamic state of body components.
- Metabolic Pathways are 2 Types

Anabolic pathways	**Catabolic pathways**
Biosynthetic pathway	Degradation pathway
Simpler molecules form complex structures.	Complex molecules become simple structures (degradation)
Involves consumption of energy	Involves release of energy
Examples: Formation of acetic acid from cholesterol, assembly of amino acids to protein, photosynthesis etc.	Examples: Formation of lactic acid from glucose (glycolysis), respiration etc.

- Chemical bonds are used to store the energy released during catabolism. This bond energy is utilized for biosynthetic, osmotic, and mechanical work when required.
- Bond energy in adenosine triphosphate (ATP) is the most significant form of energy in living systems.

THE LIVING STATE

- In order to perform work, a system must be in a non-equilibrium stable state.
- A system in equilibrium cannot perform work.
- Consequently, an individual's life processes are constant efforts to evade equilibrium.
- Avoiding equilibrium state requires the input of energy provided by metabolism.
- Therefore, without metabolism, a living state is not possible.
- Living organisms exist in a stable state characterized by the concentration of biomolecules in their body, which is a non-equilibrium state.

ENZYMES

- Enzymes are complicated macromolecules with a high molecular weight.
- In a cell, they catalyze biochemical reactions. They facilitate the disintegration of large molecules into smaller molecules or the combination of two smaller molecules into a larger molecule. Thus, they are known as biocatalysts.
- Enzymes do not initiate a chemical reaction. However, they contribute to its acceleration.
- Enzymes affect the rate but not the direction of biochemical reactions.
- The majority of enzymes have a high turnover rate. The turnover number of an enzyme is the number of molecules per minute that the enzyme acts upon. A high enzyme turnover rate increases the reaction's efficacy.
- Enzyme actions are specific.
- With increasing temperature, enzyme activity decreases.
- They are most active at the optimal pH range of 6-8.The velocity of an enzyme increases as its substrate concentration rises, eventually reaching its utmost velocity.
- All enzymes are proteins but all proteins are not enzymes.
- Ribozymes are nucleic acids (RNA) with enzyme-like properties.
- Enzymes have primary, secondary, and tertiary structure, like all proteins. The substrate fits into crevices (pockets) dubbed 'active site' in the tertiary structure of an enzyme.
- Inorganic catalysts operate at elevated temperatures and pressures. However, enzymes are harmed by elevated temperatures. (>40°C). The enzymes of thermophilic organisms are stable at elevated temperatures (up to 80-90 C).

- **Carbonic anhydrase** is the most rapidly acting enzyme. It accelerates the subsequent reaction by a factor of 10 million.
- In an hour, only 200 molecules of H_2CO_3 are produced in the absence of enzyme. About 600,000 molecules are formed per second in the presence of carbonic anhydrase.

Process of Enzyme Action

- **Substrate (S)** is the substance that is transformed into **product (P)** by the action of an **enzyme (E).**
- First, the substrate "S" binds to the active site of the enzyme "E" This results in the formation of a "ES" enzyme-substrate complex.
- During the state in which a substrate is bound to an enzyme, a new structure of the substrate is formed during the transition state. It is the structure between the substrate and the finished product.
- Ultimately, the structure of the substrate is transformed into the structure of the product, and the product is then discharged from the active site.
- The transition state has greater energy and less stability than the product state.
- **Activation energy** is the difference between the **average energy content** of "**S**" and its **transition state.**

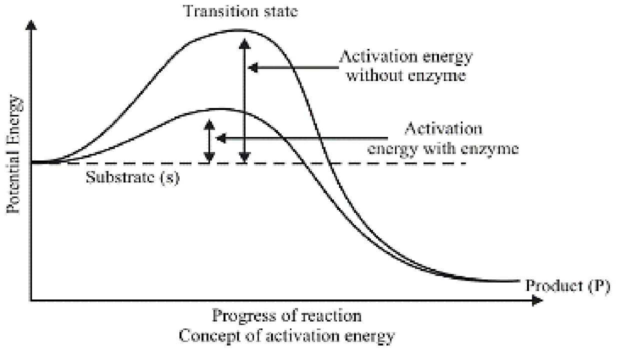


Figure: Concept of activation energy

Catalytic Cycle of Enzyme

- First of all, the substrate binds to the active site of enzyme $(E + S)$
- This induces some changes in enzymes so that the substrate is tightly bound with active site of enzyme (ES).
- The active site breaks chemical bonds of the substrate (EP).
- The enzyme releases the products and the free enzyme is ready to bind to other molecules of the substrate (E + P).

$$E + S \leftrightarrow ES \rightarrow EP \rightarrow E + P$$

The pathway of this transformation must go through the so-called ***transition state structure***.

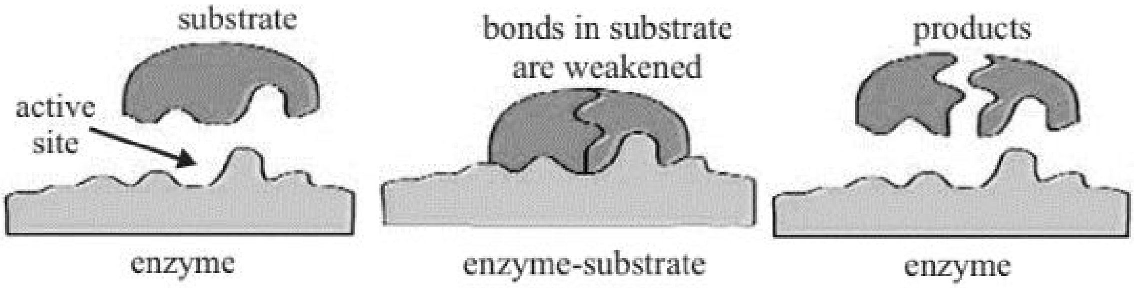
Mode of Enzyme Action

There are two distinct explanations for the mode of enzyme action: the lock and key and induced-fit hypotheses-

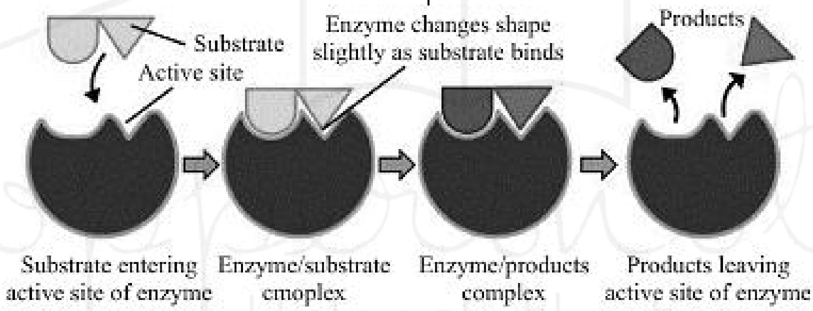
1. **Lock and Key Hypothesis:**
    - It was proposed in 1894 by **Emil Fischer**.
    - According to this, the **active sites of enzymes have a geometry that precisely complements the shape of the substrate**.
    - The enzyme molecule functions by chemically bonding with the substrate molecule to form an enzyme-substrate complex. The enzyme's tertiary structure includes a unique pocket or site where substrate molecules can attach and interact.
    - This results in an interaction between the active sites of the enzyme and the reactive sites of the substrate.
    - Now, the enzyme degrades the substrate into products. Initially, the products remain attached to the enzyme for a brief period, forming an enzyme product complex.
    - The products are then liberated from the enzyme molecule. The enzyme is now able to accept a new substrate molecule. Thus, the same enzyme can be utilized repeatedly.
    - According to this model, an enzyme is capable of catalyzing a **reverse reaction**.

$$S + E \rightleftharpoons ES \rightleftharpoons EP \rightleftharpoons E - P$$

S	E	ES	EP	E – P
Substrate	Enzyme	Enzyme substrate complex	Enzyme product complex	Enzyme Product



2. **Induced Fit Hypothesis:**
   - It was proposed in 1959 by Daniel Koshland.
   - In the presence of a specific substrate, the morphology of active sites changes to become complementary.
   - Between the substrate and the products, there exists a highly unstable transition state.
   - When substrate molecules bind to an enzyme molecule, the active site undergoes a change to precisely fit the transition state (induced fit).
   - This induced alignment maintains the substrates at the proper angle for the reaction to occur.



Factors Affecting Enzyme Action

Changes in environmental conditions can affect the activity of an enzyme by altering the tertiary structure of the protein.

These include temperature, pH, changes in substrate concentration, and the binding of particular compounds.

(a) pH: enzymes are pH-sensitive. Each enzyme is at its most active at the optimal pH. Below and beyond the optimal value, activity decreases.

(b) Temperature: Low temperature maintains the enzyme in a state of temporary inactivity, whereas high temperature destroys enzymatic activity because heat denatures proteins. In general, all enzymes function optimally at body temperature.

(c) Enzyme Concentration: The rate of a reaction is directly proportional to the concentration of an enzyme. An increase in enzyme concentration will increase the reaction rate up to a certain point, after which the reaction rate will remain constant. Increasing the concentration of an enzyme increases the number of active sites.

(d) Concentration of substrate: An increase in substrate concentration increases the enzyme's activity until all of its active sites are saturated with substrate molecules. Consequently, the substrate molecules occupy the active sites vacated by the products and are unable to accelerate the reaction rate further.