

الجامعة التكنولوجية

قسم الهندسة الكيميائية

المرحلة الاولى

الكيمياء الاحيائية

م. علياء عصام

CHAPTER - 4

Amino Acids, Peptides & Proteins

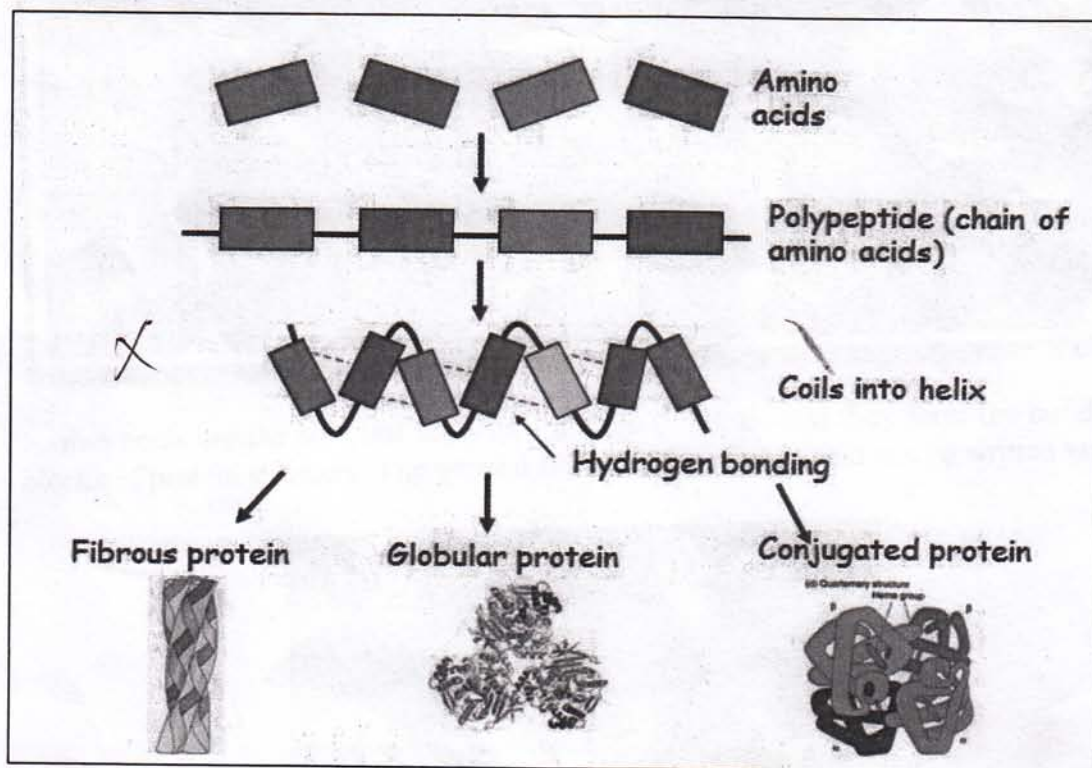
Introduction

Proteins are complex organic nitrogenous substances found in animal and plant tissues. The term protein is derived from Greek: Proteios means primary or holding first place.

Protein is the essential constituent of living cells.

Protein make up to 12% of the protoplasm. They are not only responsible for comprising the structure of the cell but are concerned with every function of the cell including those of respiration, catalysis of reactions by enzymes, transport of materials, regulation of metabolism, and defense actions.

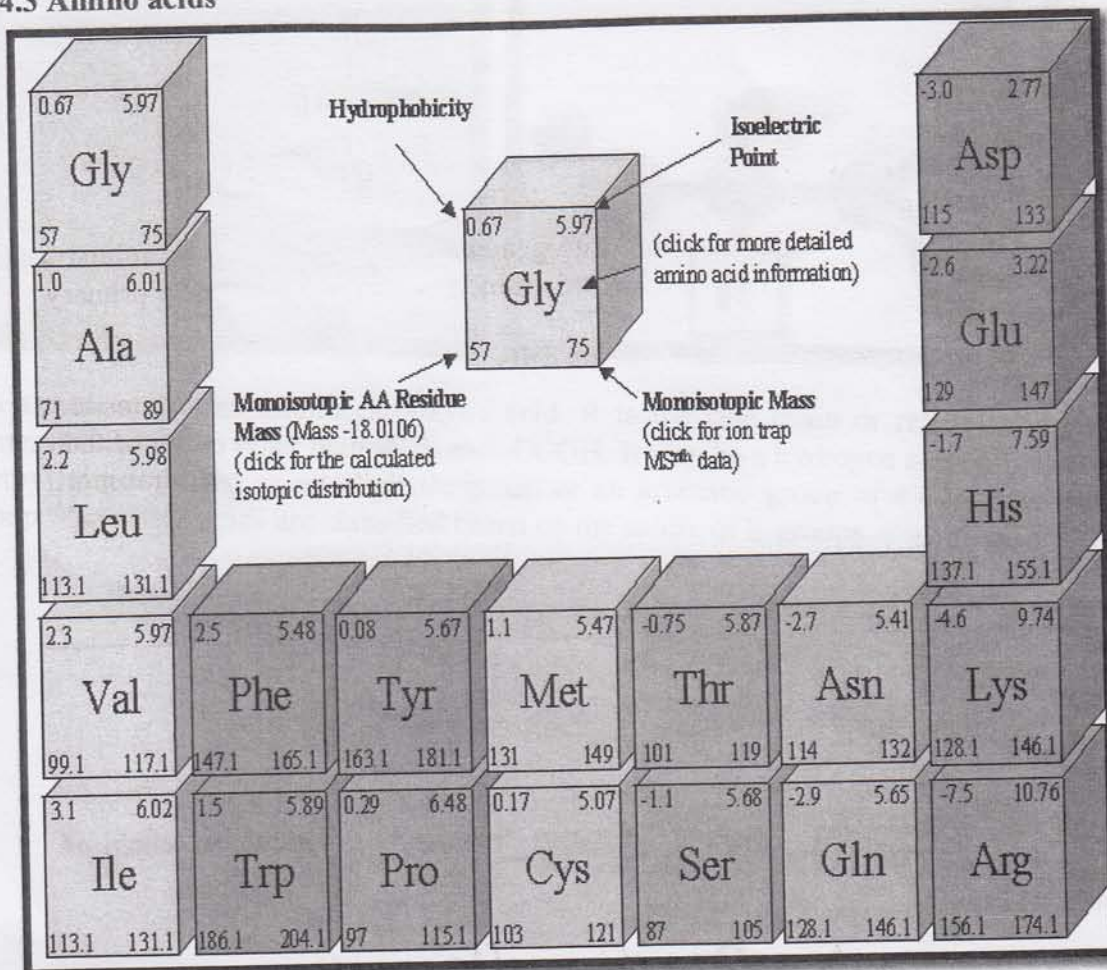
The foods rich in proteins are known as body building foods.



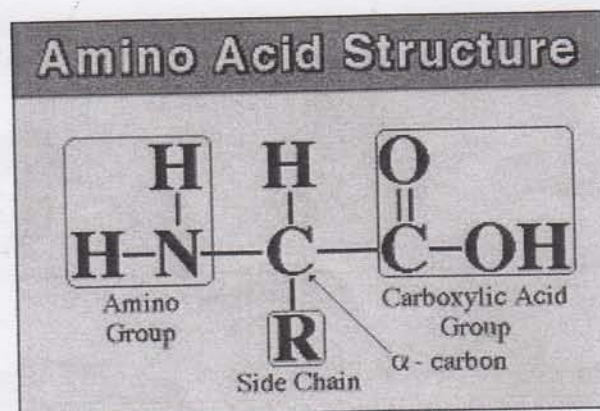
Sources of protein

Proteins are obtained from animal and plant sources. The animal sources of proteins include milk, egg, meat, fish, liver etc. Plant sources of proteins are pulses, nuts and cereals.

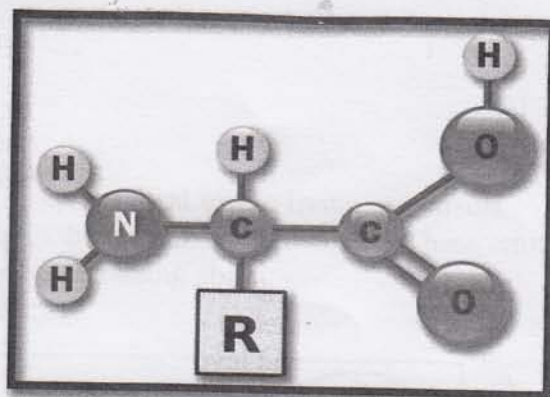
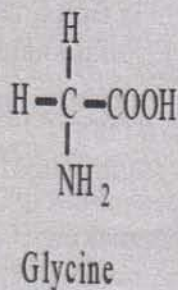
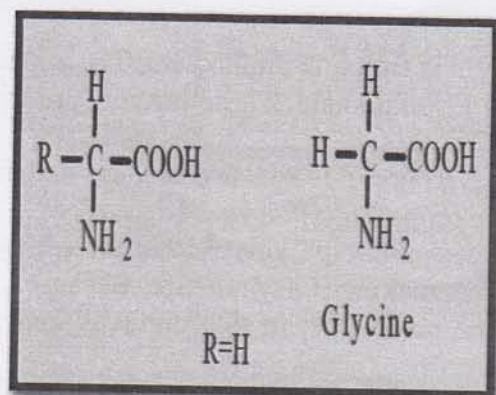
4.3 Amino acids



Amino acids are the simplest units of a protein molecule and they form the building blocks of protein structure. The general formula of an amino acid can be written as:



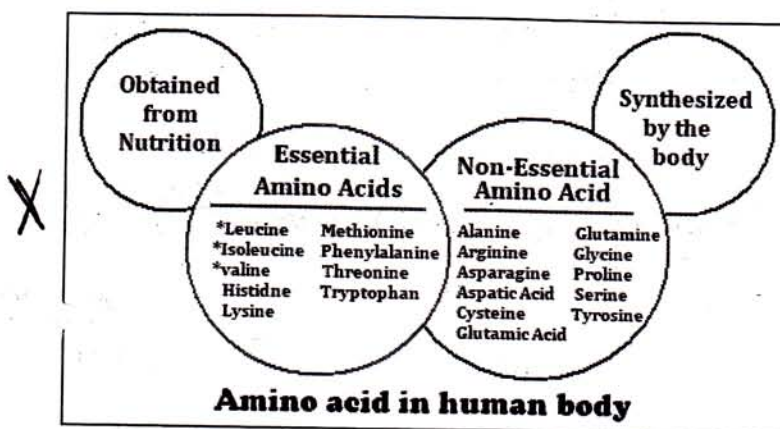
The general structure of α -amino acid in its un-ionized form.



An amino acid is an amino carboxylic acid. R is the side chain or residue and it represents the group other than $-\text{NH}_2$ and $-\text{COOH}$. It may be a hydrogen atom (H) or a methyl group ($-\text{CH}_3$) or an aliphatic group or an aromatic group or a heterocyclic group. The amino acids are classified based on the nature of R groups.

Essential amino acids

Certain amino acids cannot be synthesized by the living organisms. They must be compulsorily included in the diet for normal health. These amino acids are called essential amino acids. For human being about 10 amino acids are considered as essential:



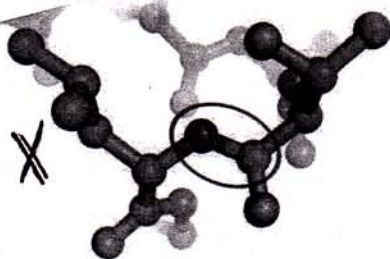
Non-essential amino acids

Certain amino acids can be synthesized in the cells from essential amino acids or from other compounds. So these amino acids need not be included in the diet. They are called non-essential amino acids.

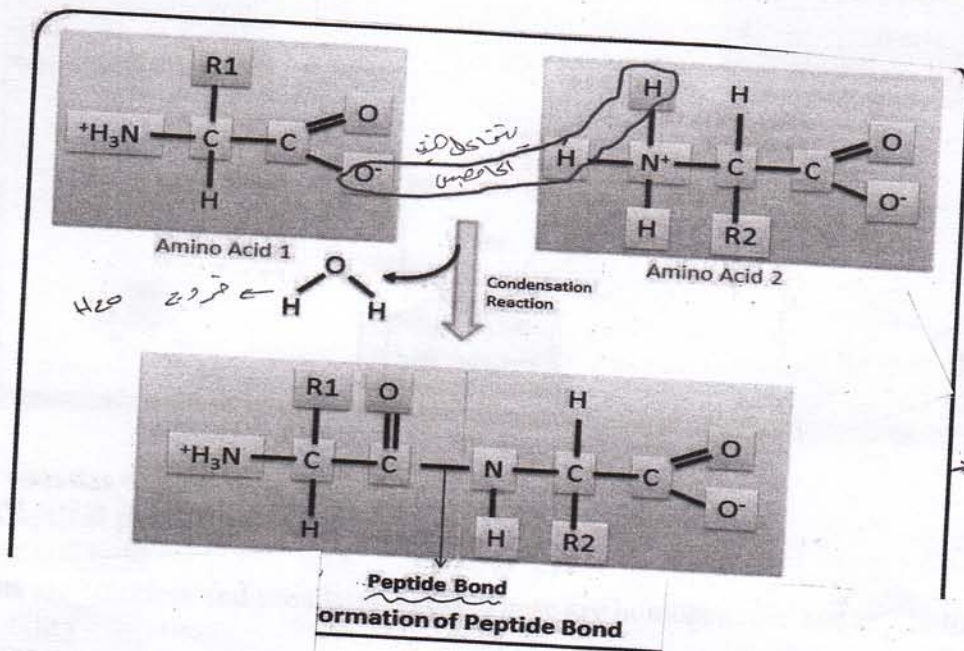
Non protein amino acids

Certain amino acids which do not exist in proteins are called non protein amino acids eg. Ornithine and β -alanine etc..

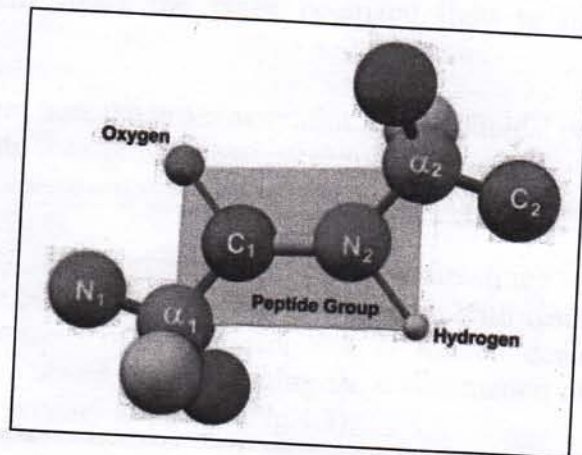
Peptide bonds



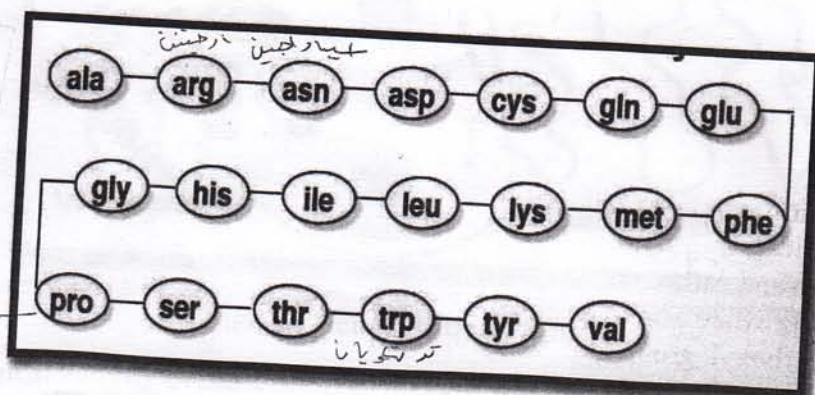
In proteins, amino acids are linked together by linkages called peptide bonds. The carboxyl group of one amino acid is joined to the α -amino group of another amino acid by a peptide bond.



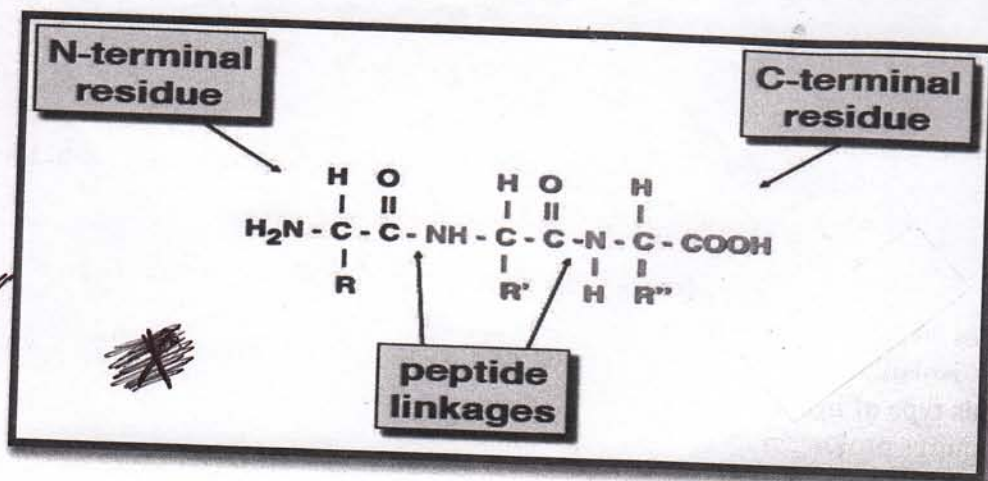
The peptide bond is also called as the amide bond. The two amino acids, joined by a peptide bond, constitute a dipeptide. The dipeptide is formed by simple condensation reaction.



Here is an example sequence of amino acids in a protein. It also shows the abbreviations of amino acids in a protein.



The product formed by a peptide bond is called a peptide. The compound formed by the linking of three amino acids is called as tripeptide.



4.4 Properties of proteins

4.4.1 Physical properties

1. Color and taste

Proteins are colorless and usually tasteless. These are homogeneous and crystalline.

2. Solubility

Solubility of proteins is influenced by pH. Solubility is lowest at isoelectric point and increased with increasing acidity or alkalinity.

3. Optical activity

All protein solutions rotate the plane polarized light to the left i.e. these are levorotatory.

4. Colloidal nature

Because of their giant size, the proteins exhibit many colloidal properties are:

- Their diffusion rate is extremely low.
- They may produce considerable light-scattering in solution, thus resulting in visible turbidity (Tyndall effect).

5. The comparatively weak forces responsible for maintaining secondary, tertiary and quaternary structure of proteins are readily disrupted with resulting loss of biologic activity. This disruption of native structure is termed denaturation. Physically, denaturation may be viewed as randomizing the conformation of a polypeptide chain without affecting its primary structure (Fig.4.3).

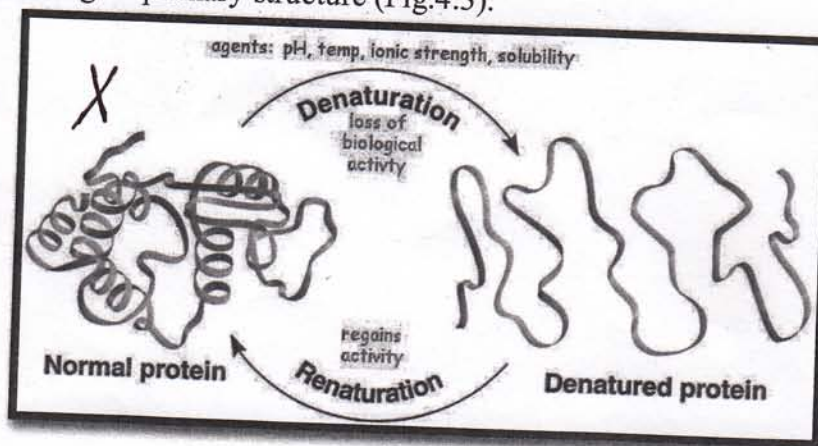


Fig. 4.3 Denaturation of protein.

The biological activity of most proteins is destroyed by exposure to strong mineral acids or bases, heat, urea, acetone, alcohol and ionic detergents etc. Denatured proteins are less soluble in water.

Enzymes & Kinetic enzymes

1 Introduction

All the enzymes are proteins and they are produced by the living cells. They act as biological catalysts. Enzymes catalyzed and enhance the rate of biochemical reactions occurring in various vital processes like breathing, digestion, pumping of heart, formation of body tissues, contraction of muscles, transport of ions across the plasma membranes etc. So without enzymes there is no life. They are inactive at 0°C and destroyed by moist heat at 100°C .

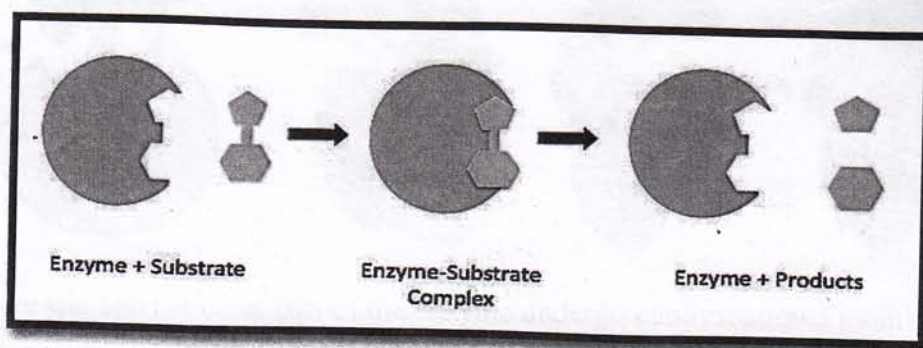
The substances on which the enzymes act are called as "Substrates".

Enzymes are highly specific in their action (i.e) an enzyme can act on a single or a small group of closely related substrates. During catalytic action, the enzymes do not undergo any permanent modification and regenerated at the end of the reaction. The general enzyme catalyzed reaction takes place as per the equation.



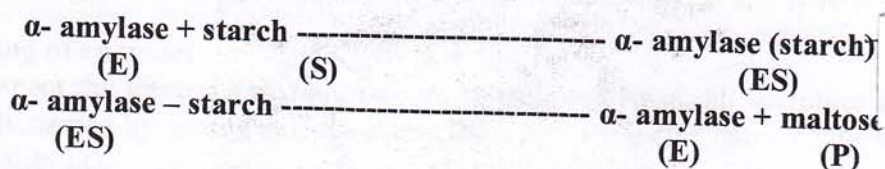
E - Enzyme; S = Substrate; ES - Enzyme-Substrate complex.

P = product. ES complex is an unstable and highly energized complex.



For example:

α -amylase acts on starch and produce maltose units. In this reaction α -amylase is the enzyme, starch is the substrate and maltose is the product.

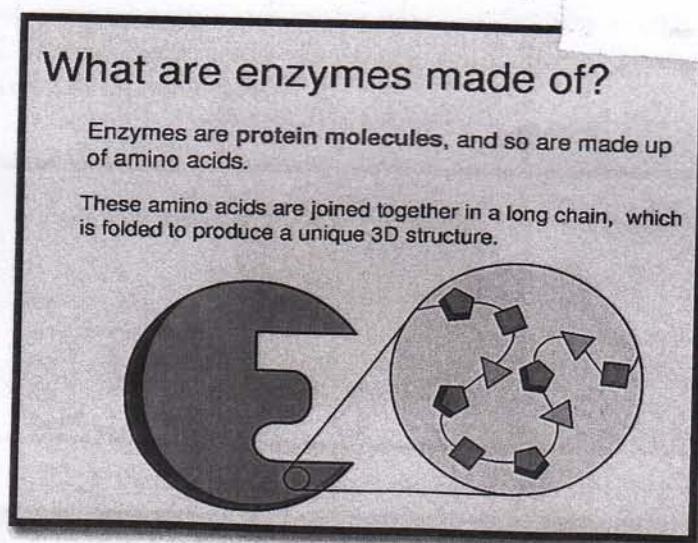


Most of the enzymes, synthesized by a living cell, function within that cell and hence are called as endo enzymes or intra cellular enzymes.

Some enzymes are liberated from the living cells after synthesis, secreted to the environment and function in extra cellular regions. These enzymes are called as exo enzymes or extra cellular enzymes

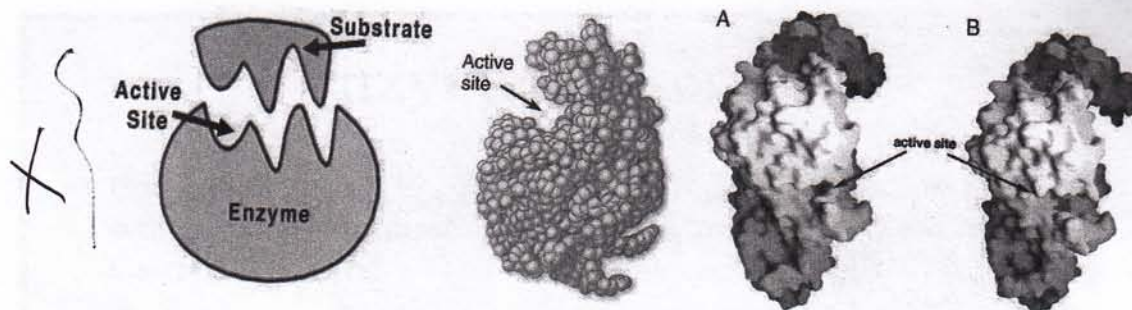
5.2 Chemical nature of enzymes

All the enzymes are proteins with no exception. Some enzymes are simple proteins i.e. their molecules consist of only amino acids.



Many enzymes are conjugated proteins and their molecules consist of Amino acids and a non-protein part (not made up of amino acids). The non-protein part of the enzyme is known as coenzyme or prosthetic group, without which the enzyme is inactive. The protein part of the enzyme (in conjugated type) is known as the apoenzyme. The coenzyme and the apoenzyme complex is called as holoenzyme.





The active site and the other part of the enzyme undergo conformational modification when they come in contact with the substrate. Koshland's induced fit hypothesis of enzyme-substrate interaction postulates that the active site of the enzyme consists of a number of 'active' contact amino acids which permit the substrate to come close to the reactive groups of the enzyme which thereupon undergoes a conformational change, binding the substrate firmly to the enzyme and promoting catalytic activity.

Naming of enzymes

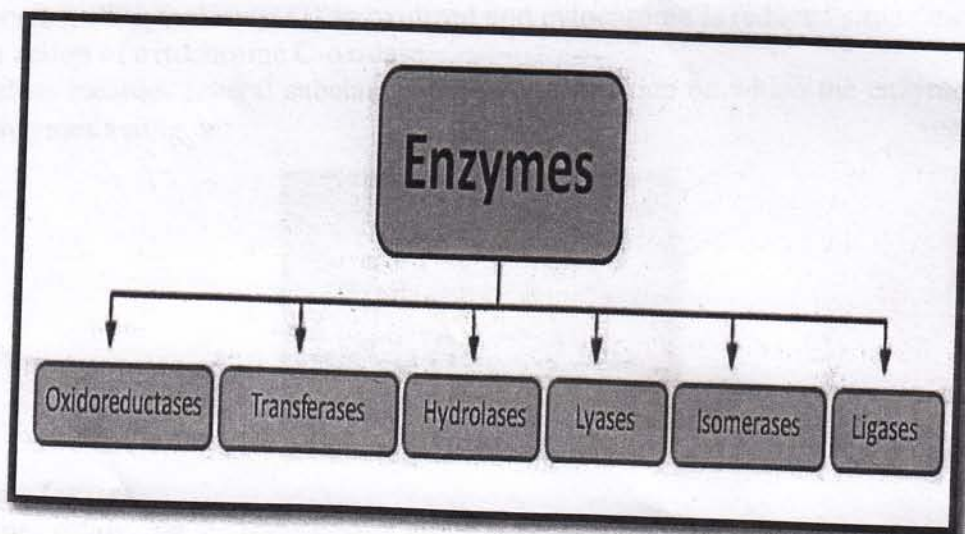
Except the enzymes ptyalin, pepsin, trypsin and renin, all the other enzymes are usually named by adding suffix - ase to the main part of the name of the substrate on which they act.

Examples:

Maltase acts on maltose
Lactase acts on lactose
Lipases act on lipids
Proteases act on proteins
Amylases act on starch (amylum)

Classification of enzymes

The most comprehensive system for the classification of enzymes was devised in 1961 by the Enzyme Commission of International Union of Biochemistry (IUB). The 6 major classes of enzymes are:

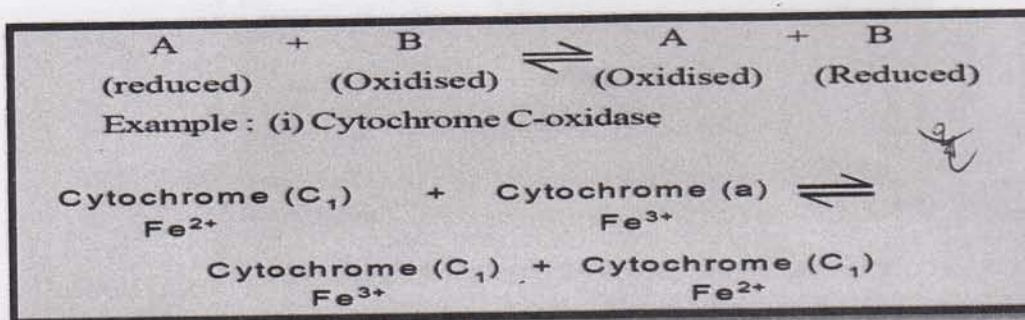


Enzymatic Action

- **Oxidoreductases (EC 1):** oxidation-reduction reactions, which entail the transfer of electrons from a substrate to another substrate
- **Transferases (EC 2):** move a chemical group from one compound to another compound
- **Hydrolases (EC 3):** Hydrolysis of a chemical bond
- **Lyases (EC 4):** formation of a double bond
- **Isomerases (EC 5):** catalyzes substrate to an isomeric form
- **Ligases (EC 6):** catalyzes a reaction that joins two substrates

1. Oxidoreductases

Enzymes catalyzing oxido-reduction reactions between two substrates A and B are called as oxido-reductases.



In this reaction cytochrome C1 is oxidized and cytochrome is reduced simultaneously by the action of cytochrome C-oxidase.

This class includes several subclasses based on the group on which the enzymes act. The enzymes acting on:

-CH -OH	(1.1)
-C=O	(1.2)
-C=CH	(1.3)
-CH-NH ₂	(1.4)
-CH-NH	(1.5)

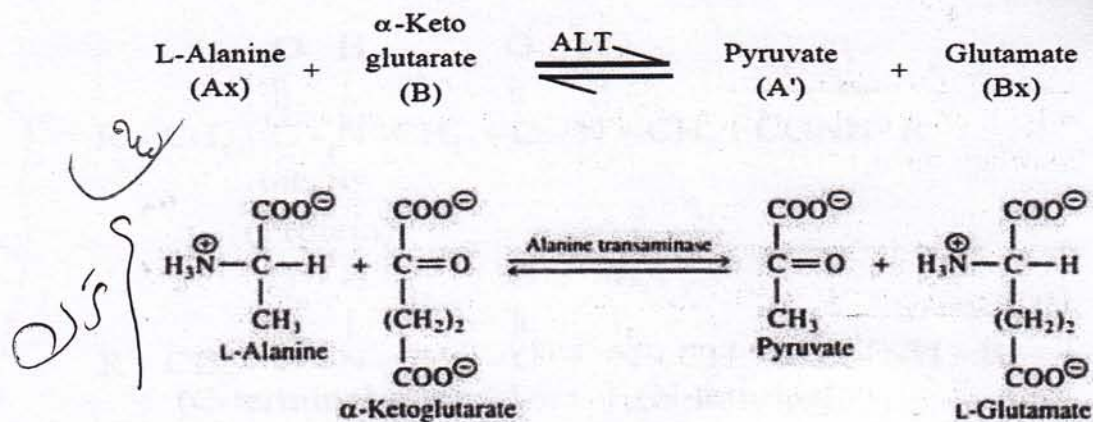
2. Transferases

Enzymes catalyzing the transfer of a group (x) from one substrate (AX) to another (B) are known as transferases.



Example:

The reaction catalyzed by alanine transaminase (ALT) is:



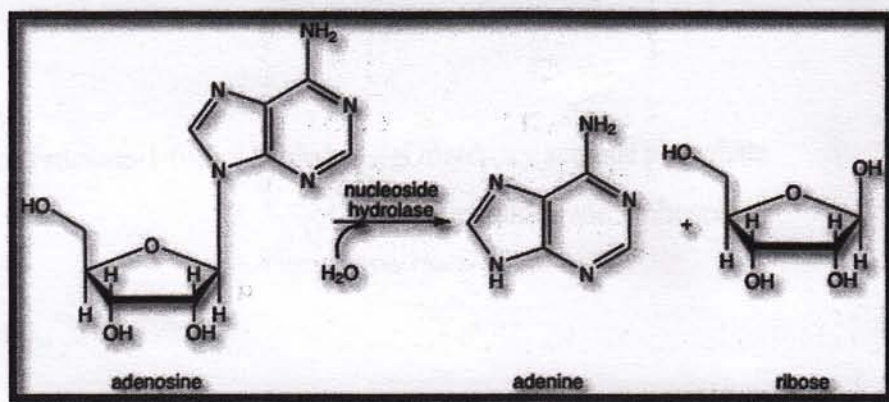
In this reaction the amino group from alanine is transferred to α -ketoglutarate to form glutamate.

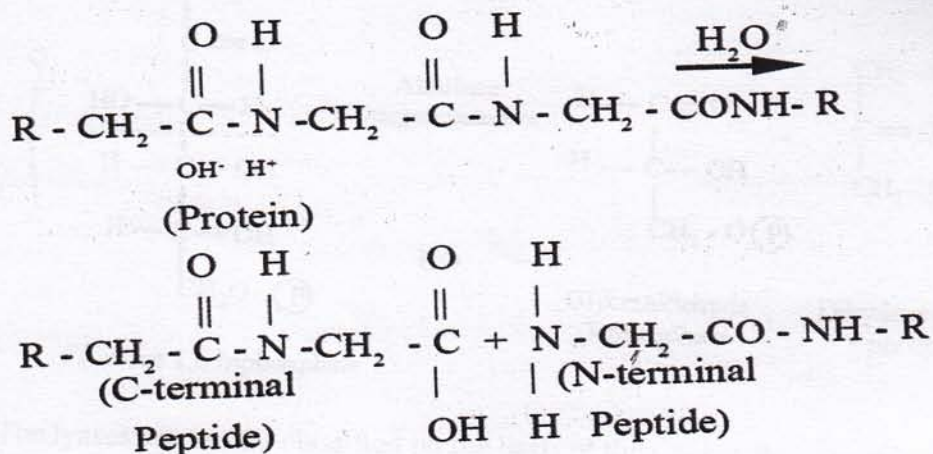
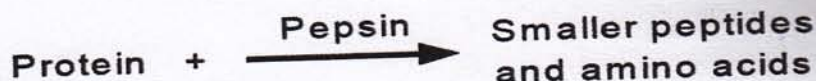
These enzymes are further divided into subclasses on the basis of nature of the group transferred.

- | | |
|--------------------------------------|-------|
| Transfer of (a) one carbon compounds | (2.1) |
| (b) aldehyde or ketonic groups | (2.2) |
| (c) acyl groups | (2.3) |
| (d) glycosyl groups | (2.4) |
| (e) Phosphate groups | (2.7) |
| (f) Sulphur containing groups | (2.8) |

3. Hydrolases

The hydrolases are those enzymes which catalyze hydrolysis reactions i.e the direct addition of water molecule (s) across the bond, which is to be cleaved. The substrate for these enzymes are esters, ethers, peptides and glycosides. Example: Pepsin. This enzyme is a gastro intestinal enzyme which is proteolytic in nature and involve in the hydrolysis of proteins present in the food.



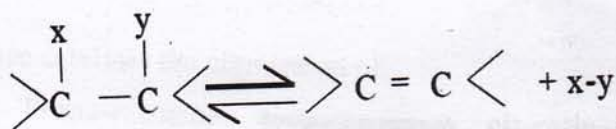


The hydrolases are divided into several subclasses, depending on the nature of the group or bond being hydrolyzed viz.

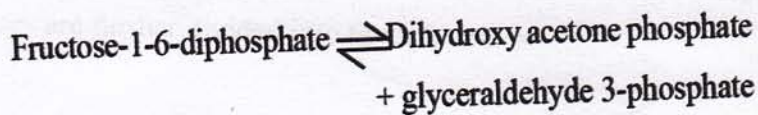
- (a) esterases etc. - hydrolyse ester bonds (3.1)
- (b) glycosidases - hydrolyse glycosidic bonds (3.2)
- (c) peptidases - hydrolyse peptide bonds (3.4)

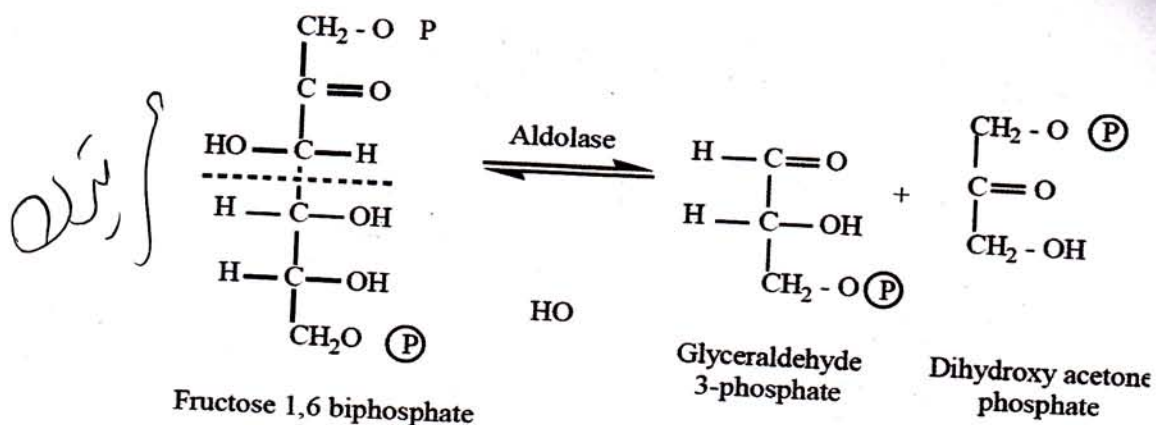
4. Lyases

The lyases are a smaller class of enzymes that catalyze the removal of a small molecule from a larger substrate molecule. Since the reactions are reversible, lyases may also be considered to catalyze the addition of small molecules to the substrate molecule.



Example:





The lyases are further classified on the basis of the linkage they attack viz., acting on:

- | | |
|------------------|-------|
| a. C-C bond | (4.1) |
| b. C-O bond | (4.2) |
| c. C-N bond | (4.3) |
| d. C-S bond | (4.4) |
| e. C-halide bond | (4.5) |

5. Isomerases

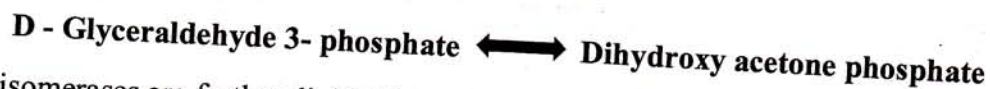
This class includes all enzymes which catalyze isomerization reactions i.e. interconversion of optical, geometrical or position isomers.

Example:

Retinene isomerase catalysis the conversion of:



Triose phosphate isomerase catalyses the conversion of:



The isomerases are further divided into the subclasses:

- | | |
|-------------------------|-------|
| A. racemases | (5.1) |
| B. epimerases | (5.2) |
| C. cis-trans isomerases | (5.3) |

6. Ligases

These enzymes are otherwise known as synthetases. They catalyze synthesis reactions by joining two molecules, coupled with the breakdown of a phosphate bond of adenosine triphosphate. ATP cleavage provides energy for the new bond formation.

Example:

Formation of malonyl CoA from acetyl CoA in the presence of acetyl CoA carboxylase.



The subclasses of ligases are based on the nature of bond formed in the product.
Formation of:

- a. C-O bond (6.1)
- b. C-S bond (6.2)
- c. C-N bond (6.3)
- d. C-C bond (6.4)

Class	Reaction type	Important subclasses
1 Oxidoreductases	<p>○ = Reduction equivalent</p> <p>A_{red} + B_{ox} ⇌ A_{ox} + B_{red}</p>	Dehydrogenases Oxidases, peroxidases Reductases Monooxygenases Dioxygenases
2 Transferases	<p>A-B + C ⇌ A + B-C</p>	C ₁ -Transferases Glycosyltransferases Aminotransferases Phosphotransferases
3 Hydrolases	<p>A-B + H₂O ⇌ A-H + B-OH</p>	Esterases Glycosidases Peptidases Amidases
4 Lyases ("synthases")	<p>A + B ⇌ A-B</p>	C-C-Lyases C-O-Lyases C-N-Lyases C-S-Lyases
5 Isomerases	<p>A ⇌ Iso-A</p>	Epimerases <i>cis trans</i> Isomerases Intramolecular transferases
6 Ligases ("synthetases")	<p>A + B + XTP ⇌ A-B + XDP</p> <p>X = A, G, U, C</p>	C-C-Ligases C-O-Ligases C-N-Ligases C-S-Ligases

Factors influencing enzyme activity

The activity of enzymes is markedly affected by several factors. These factors are:

1. Temperature
2. pH
3. Substrate concentration
4. Metal ions (activators)
5. Inhibitors
6. Enzyme concentration etc.

1 pH

All the enzymes have a particular pH at which their activity is maximal; above or below this pH the activity is low. The pH at which the enzyme shows maximum activity is known as optimum pH. Some of the enzymes and their optimum pH are:

- (a) Pepsin - 2.0
- (b) Amylase - 7.0
- (c) Trypsin - 8.5
- (d) Alkaline phosphatase - 9.9

Only in this optimum pH, ionization of active amino acids in enzymes and substrate are favored for ES complex formation. The pH activity relationship is shown in the (Fig. 5.1).

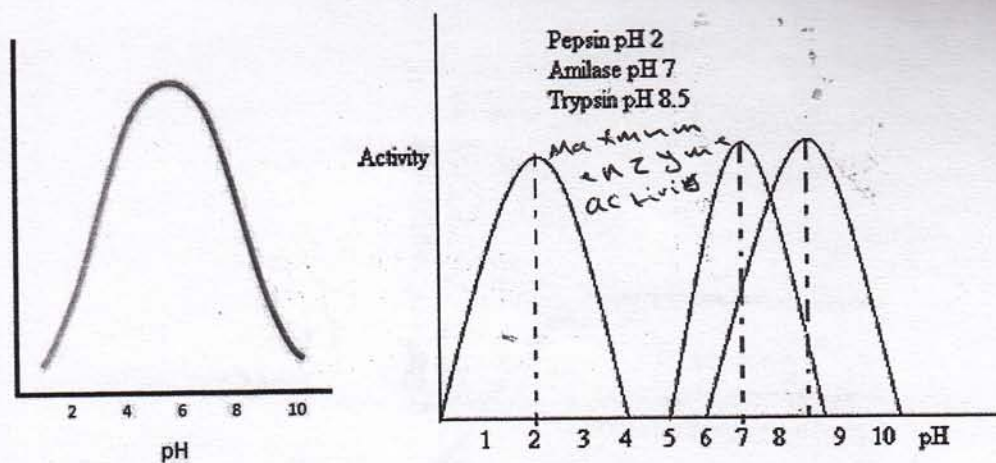


Fig. 5.1 The pH activity relationship.

2 Temperature

Rise in temperature causes increase in the rate of enzyme catalyzed reactions up to a certain temperature i.e. about 45°C . Above which the activity declines due to denaturation of enzymes (due to their protein nature). As the enzyme is denatured and inactivated, the reaction which it catalyzes slows down and ultimately stops. So the temperature at which the enzyme shows maximum activity is known as optimum temperature. The optimum temperature of most of the enzymes is found to be 37°C . The relationship of enzyme activity to temperature is shown below in Fig. 5.2:

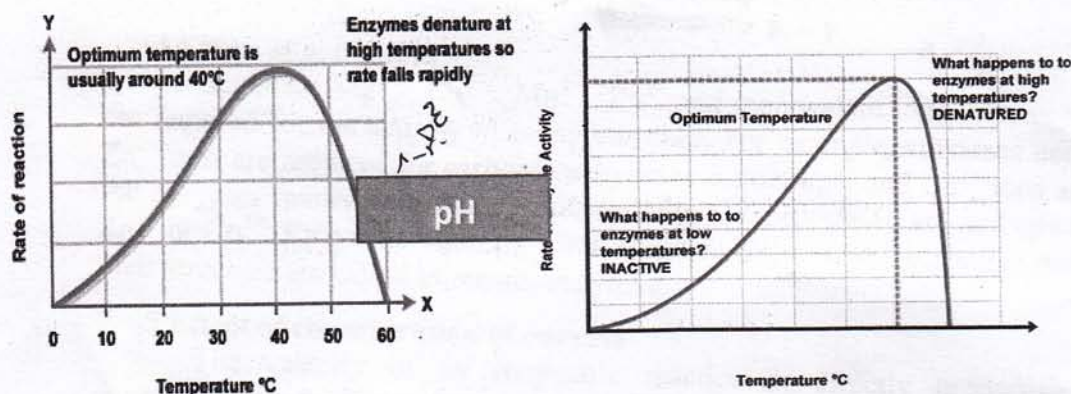
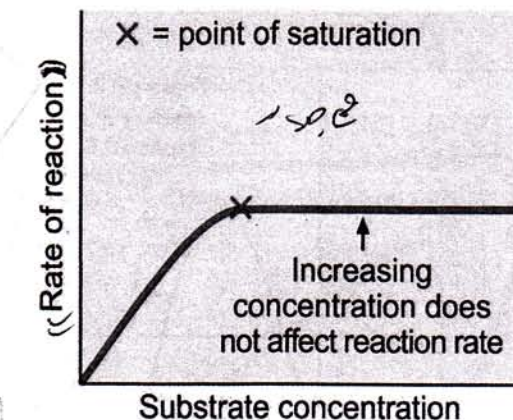


Fig. 5.2 The temperature activity relationship.

3 Substrate concentration

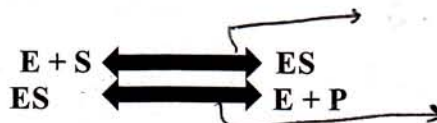
With a fixed amount of enzyme, the reaction rate is proportional to the concentration of substrate. But this is true up to a certain concentration after which the increase in concentration of substrate does not further increase the velocity of the reaction. ✓

Since the number of active sites on an enzyme molecule are limited, a stage will come when all of them have filled with the substrate molecules. This is known as saturation of enzyme. Now, since none of the active sites of the enzyme is free, further addition of the substrate molecule will not increase the product formation (Fig. 5.3).



Substrate concentration - activity relationship.

It was Michaelis and Menten in 1913, who proposed a successful explanation for the effect of substrate concentration on the enzyme activity. According to them the enzyme 'E', and the substrate 'S' combine rapidly to form a complex, the enzyme substrate complex 'ES'. The complex then breaks down relatively, slowly to form the product of the reaction. The enzyme regenerated can involve in another round of catalysis.



4 Effect of activators

Ions, like Mg^{2+} , Cu^{2+} , Mn^{2+} , Zn^{2+} and monovalent ions such as Na^+ and K^+ are required for the activity of many enzymes. For example, amylases need Cl^- ions, Zn^{2+} ions are required for carbonic anhydrase action, Fe^{2+} and Cu^{2+} ions are required for enzymes involved in redox reactions. Several peptidases are activated by Mn^{2+} , Zn^{2+} or Co^{2+} . Enzymes requiring metal ions or enzymes which contain metal ions in their structure are called as metalloenzymes.

5 Effect of concentration of enzyme

The velocity of an enzymatic reaction is directly proportional to the concentration of enzyme. In case the enzyme concentration is doubled then as much as twice active site become available to combine with the substrate, provided an excess of substrate is present and so the maximum velocity is also doubled. At a fixed concentration of the substrate a level is reached when all the substrate molecules are utilized and no more change in velocity of the reaction takes place (Fig. 5.4).

Enzyme Kinetics

For all enzymatic processes the rate of the reaction depends upon the concentration of the enzyme and its substrates, other conditions like temperature and pH being constant. Figure 6.1 shows the relationship between the substrate and product concentrations.

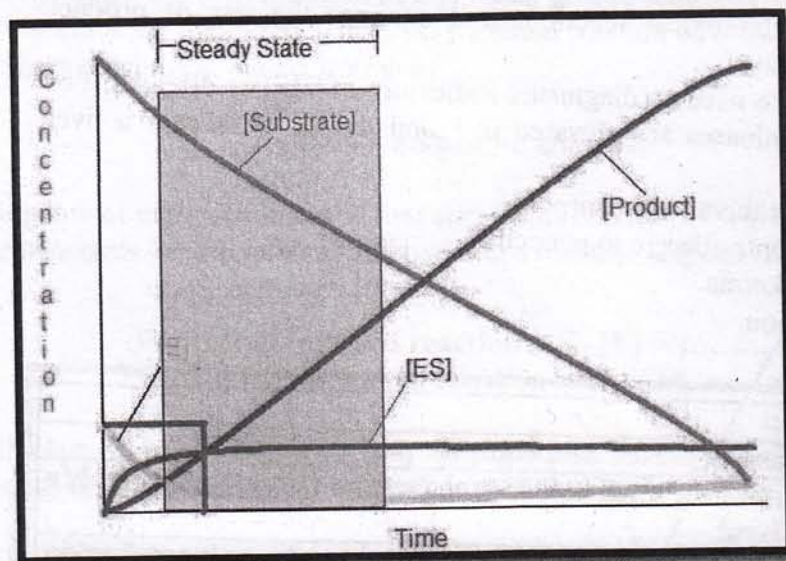
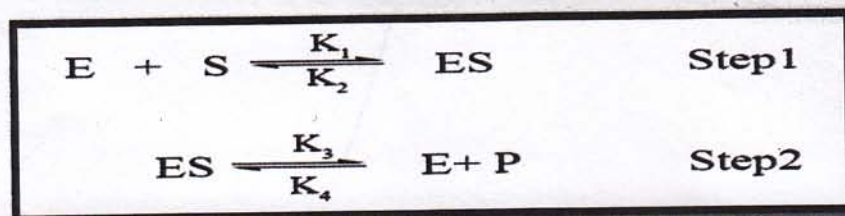


Fig .6.1 Relationship between substrate and product concentrations.

Although the velocity increases linearly with enzyme concentration, at constant enzyme concentration it increases hyperbolically as the substrate concentration increases. This indicates that the enzyme has a definite number of sites to combine with substrate. When all sites are occupied, no further rate enhancement occurs and the enzyme is saturated with the substrate.

Derivation of M - M Equation

Leonor Michaelis and Mand L. Menton in 1913 proposed a successful explanation for the effect of substrate concentration on the enzyme activity. According to them the enzyme E, and the substrate S combines rapidly to form a complex, the enzyme substrate complex ES. This complex then breaks down relatively and slowly to form the product P of the reaction. These sequence of reactions can be represented in the following equations.



Step 1: k_1 & k_2 are the rate constants of the forward and backward reactions.

Step 2: k_3 & k_4 are the rate constants of the forward and backward reactions respectively.

This is true only for the enzyme reactions which fulfill the following conditions:

- Only a single substrate and a single product are involved.
- The reaction proceeds essentially to completion.

- c. The concentration of the substrate is much greater than that of the enzyme in the system.
- d. An intermediate enzyme substrate complex is formed.
- e. The rate of decomposition of the substrate is proportional to the concentration of the enzyme substrate complex.

It is assumed that the concentration of S is much greater than that of E and that only initial velocities are measured, where only a small fraction of S has been converted. Under these conditions, concentration of:

P----- ES can be ignored.

Applying law of mass action to the first step of the reaction in which K_1 and K_2 are the rate constants for the forward and backward reaction respectively:

$$\text{The rate of forward reaction} = K_1 [E] [S] \dots\dots\dots (1)$$

$$\text{The rate of backward reaction} = K_2 [ES] \dots\dots\dots (2)$$

Applying law of mass action to the second step of the reaction in which k_3 and k_4 are the rate constants for the forward and backward reaction respectively,

$$\text{The rate of forward reaction} = K_3 [ES] \dots\dots\dots (3)$$

The rate of backward reaction can be neglected. The total enzyme in the system can be represented as:

$$[E_t] = [E] + [ES] \dots\dots\dots (4)$$

Where $[E]$ is the uncombined free enzyme concentration, $[ES]$ the enzyme substrate concentration and $[E_t]$ the total enzyme concentration.

The velocity of the overall reaction is:

$$V = K_3 [ES] \dots\dots\dots (5)$$

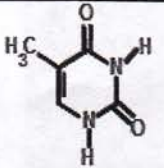
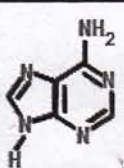
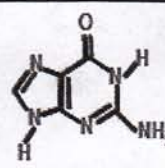
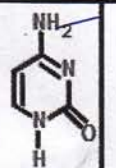
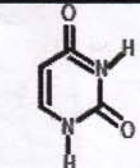
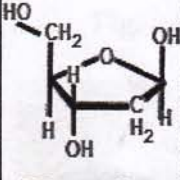
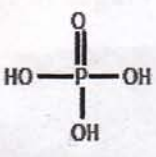
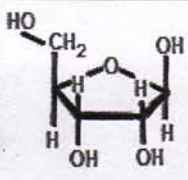
This is the actual rate equation for the overall reaction but it is not useful since neither K_3 nor $[ES]$ can be measured directly. It is assumed that the reaction proceeds at steady state where the rate of formation of $[ES]$ equals to the rate of degradation of $[ES]$. The rate of formation of ES, V_f is proportional to E and S as in any second order reaction.

NUCLEIC ACIDS

1 Introduction

Nucleic acids are colorless, complex, amorphous compounds made up of three units: purine and pyrimidine bases, sugar and phosphoric acid. The nucleic acids are of two types DNA and RNA. It is very important to know the structure of nucleic acids and their components.

Components of Nucleic Acids

	DNA only	DNA & RNA			RNA only
Nitrogen bases	 Thymine	 Adenine	 Guanine	 Cytosine	 Uracil
sugar & phosphate	 2-Deoxyribose	 Phosphate			 Ribose

Nucleic acids

Two types of nucleic acids are present in all mammalian cells.

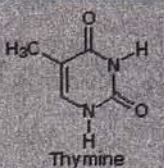
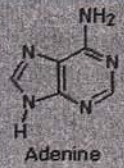
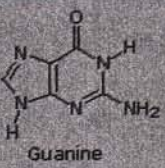
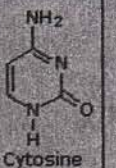
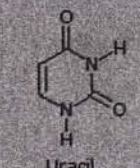
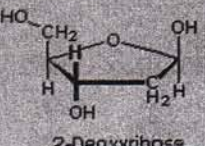
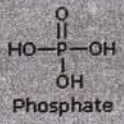
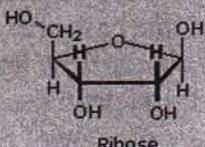
They are DNA - deoxy ribonucleic acid and RNA- ribonucleic acid.

DNA is present in the nucleus and mitochondria. RNA is present in the nucleus, ribosome and cytoplasm.

Nucleic acids are acidic substances containing nitrogenous bases, pentose sugar and phosphoric acid. Both DNA and RNA are polynucleotides. They are polymers of mononucleotides.

In nucleic acids, nucleotides are joined together by phosphodiester linkages.

Components of Nucleic Acids

	DNA only	DNA & RNA			RNA only
Nitrogen Bases	 Thymine	 Adenine	 Guanine	 Cytosine	 Uracil
Sugars & Phosphate	 2-Deoxyribose	 Phosphate			 Ribose

Nucleosides

A nucleoside is composed of purine or pyrimidine base and a pentose sugar. Two types of pentose sugar are present in nucleoside, they are ribose and deoxy ribose (Fig. .3). In the case of purine nucleosides, the sugar is attached to N-9 of the purine ring, whereas in pyrimidine nucleosides, the sugar is attached to N-1 of the pyrimidine ring. The type of linkage is N-glycosidic linkage (Fig. .4).

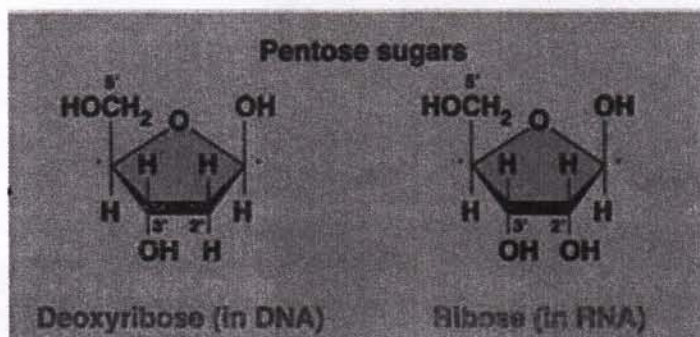


Fig. 3 Structure of sugars in nucleic acids

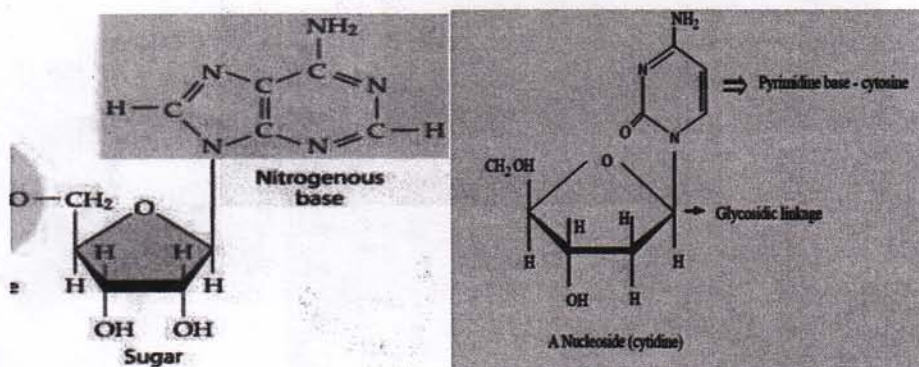
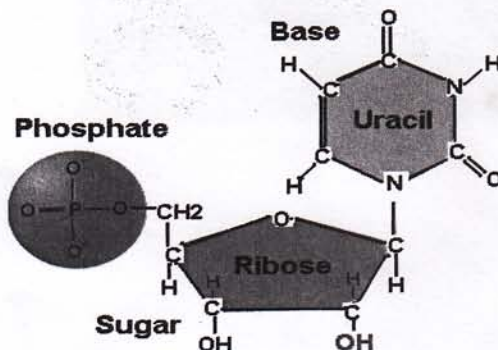


Fig. 4 Structure of a nucleoside

Nucleotides

Nucleotides are phosphorylated nucleosides usually one or two of hydroxyl groups of ribose (or) deoxyribose are phosphorylated. Thus a nucleotide has three structural components. They are nitrogenous base, sugar and phosphate. Phosphate is attached to ribose (or) deoxy ribose through an ester linkage (Fig. .5).



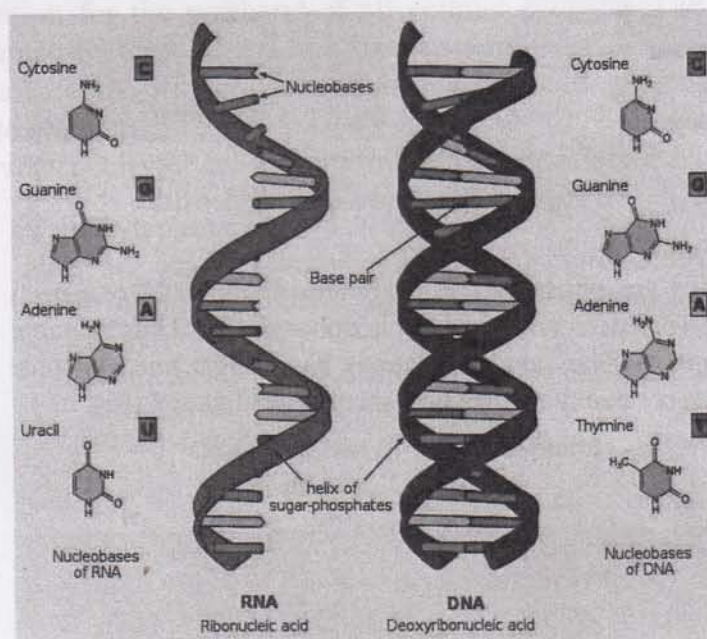
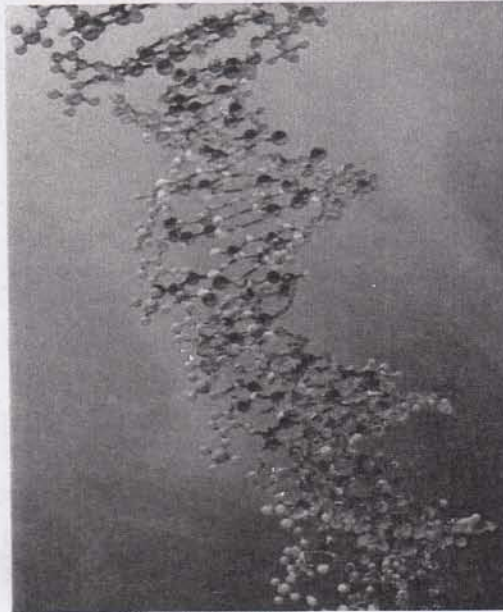
Nucleotide

Fig. .5 Structure of a nucleotide.

Structure of DNA

.1 Primary structure

Nucleotide sequence of a nucleic acid is known as its primary structure which confers individuality to the polynucleotide chain. Polynucleotide chain has direction. They are represented in 5'→3' and 3'→5' directions. Each polynucleotide chain has 2 ends. The 5' end carrying a phosphate group and 3' end carrying an unreacted hydroxyl group (Fig 7.6).



Functions of DNA

1. DNA is the genetic material of living organisms. It is the greatest super chip ever made by man.
2. DNA contain all the information required for the information of an individual organism.
3. The genetic information in DNA is converted to characteristic features of living organisms like color of the skin and eye, height, intelligence, ability to metabolize particular substance, ability to withstand stress, susceptibility to disease and ability to produce or synthesize certain substances.
4. DNA is the source of information for the synthesis of all cellular proteins. The segment of DNA that contain information for a protein is known as gene.
5. DNA is transmitted from parents to off springs and hence transmit genetic information from one generation to another.
6. The amount of DNA in any given species or cell is constant and is not affected by nutritional and metabolic states.

Structure of RNA

RNAs are present in the nucleus, ribosomes and cytoplasm of eukaryotic cells. They are involved in the transfer and expression of genetic information. They act as primer for DNA formation. Some act as enzymes and as coenzymes. RNA also function as genetic material for viruses.

RNAs are also polynucleotides. In RNA polymer, purine and pyrimidine nucleotides are linked together through phosphodiester linkages. The sugar present is ribose. The nitrogenous bases present in RNA are adenine and guanine (purine bases), uracil and cytosine pyrimidine bases). The nucleotides present in RNA are adenylic acid, guanylic acid, cytidylic acid and uridylic acid.

Purine $\begin{cases} \rightarrow A \\ \rightarrow G \end{cases}$

Pyrimidine $\begin{cases} \rightarrow U \\ \rightarrow C \end{cases}$

The Structure of RNA RiboNucleic Acid

- RNA is a polymer composed of RNA Nucleotides.

Each "Nucleotide" is made up of 3 components:

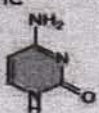
1. A phosphate group
2. A sugar - the sugar in RNA is Ribose.
3. A Nitrogenous Base

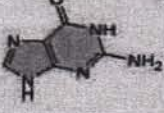
Held Together by "Covalent Bonds." These bonds are strong.... You wouldn't want your DNA falling apart

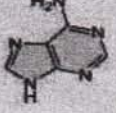
This is how I draw a Nucleotide

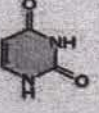
Phosphate
Covalent Bonds
Nitrogenous Base
Pentose Sugar

$P - S - B$

Cytosine  **C**

Guanine  **G**

Adenine  **A**

Uracil  **U**

Nucleobases of RNA

Nucleobases

Base pair

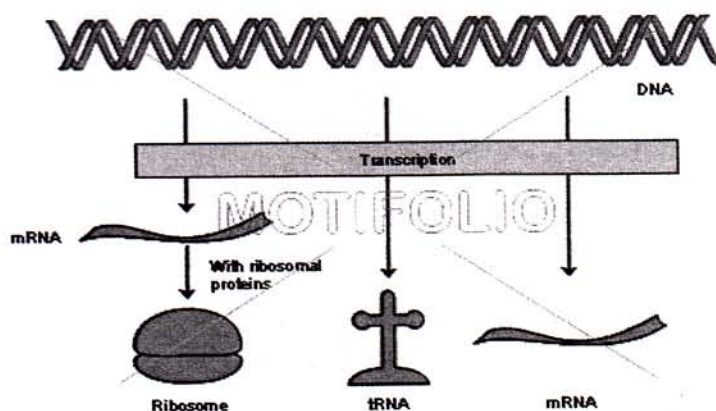
helix of sugar-phosphates

Types of RNA

There are mainly three types of RNAs in all prokaryotic and eukaryotic cells. They are:

- (1) Messenger RNA (mRNA)
- 2) Transfer RNA (t RNA)
- 3) Ribosomal RNA (r RNA). They differ from each other by size, formation and stability.

The transcription of the three types of RNA



1. Messenger RNA

It accounts for 1-5% of cellular RNA. They have a primary structure. They are single stranded linear molecules. They consist of 1000-10,000 nucleotides. They have a free or phosphorylated 3' and 5' end. They have different life span ranging from few minutes to days. mRNA molecules are capped at 5' end by methylated guanine triphosphate. Capping protects mRNA from nuclease attack. At 3' end a polymer of adenylate (poly A) is found as the tail. Poly A tail protects mRNA from nuclease attack.

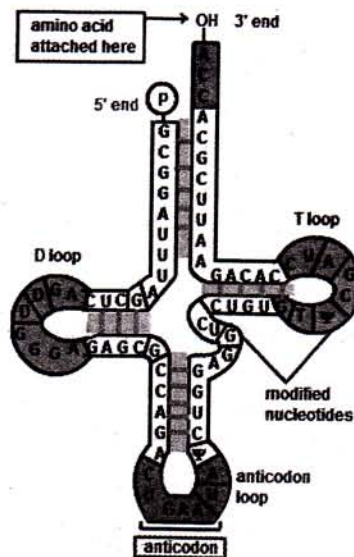
Intrastrand base pairing among complementary bases allows folding of the linear molecule. As a result, hairpin or loop like secondary structure is formed.

Functions

1. mRNA is a direct carrier of genetic information from the nucleus to the cytoplasm.
2. It contain information required for the synthesis of protein molecules.

2. Transfer RNA

It accounts for 10-15% of total cell RNA. They are the smallest of all the RNAs. Usually they consist of 50-100 nucleotides. They are single standard molecules. They contain unusual bases such as methylated adenine, guanine, cytosine and thymine, dihydrouracil and pseudouridine. These unusual bases are important for binding 6-RNA to intra chain base pairing. Further some bases are not involved in base pairing, resulting in loops and arms formation in tRNA. These folding in the primary structure generates a secondary structure



Secondary Structure of t-RNA.

Secondary structure of t-RNA is in the form of a clover leaf. The important feature of the clover-leaf structure are:

1. An acceptor arm with base sequence "CCA" 3'-OH of adenosine moiety of t-RNA.
2. An anticodon arm which recognizes codon on mRNA.
3. T_jC arm which contain unusual base cytosine.
4. D- arm which contain many dihydrouracil residues.

Functions

1. It is the carrier of amino acids to the site of protein synthesis.
2. There is at least one t-RNA molecule to each of 20 amino acids required for protein synthesis.

3. Ribosomal RNA

This accounts for 80% of the total cellular RNA. It is present in ribosomes. In ribosomes, r-RNA is found in combination with protein. It is known as ribonucleoprotein. The length of rRNA ranges from 100-600 nucleotides. rRNA molecules have a secondary structure. Intra strand base pairing between complementary bases generate double helical segments or loops.

Functions

1. They are required for the formation of ribosomes
2. They are involved in the initiation of protein synthesis.

Differences between DNA and RNA

	DNA	RNA
1.	Sugar moiety is deoxy ribose	Sugar moiety is ribose
2.	<u>Uracil</u> is absent	Thymine is usually absent
3.	Double stranded molecules both run in opposite directions	Single stranded molecules
4.	Sum of purine and pyrimidine bases are equal $G+C = A+T$	Sum of purine and pyrimidine bases are not equal. $G+C \neq A+T$.
5.	Bases are not modified	Bases are modified
6.	Resistant to alkali hydrolysis	Highly susceptible to alkali hydrolysis.
7.	No catalytic activity	Some are catalytically active.
8.	Mostly DNA is present in nucleus and also in mitochondria	present in nucleolus and cytoplasm

Biochemistry

Biochemistry is a branch of life science which deals with the study of chemical reactions that occur in living cells and organisms.

Life is a chemical process involving thousands of different reactions occurring in an organized manner.

It takes into account the studies related to the nature of the chemical constituents of living matter, their transformations in biological systems and the energy changes associated with these transformations.

The major objective of Biochemistry is the complete understanding of all the chemical processes associated with living cells at the molecular level (Biomolecules) found in

cells to determine their structures and to analyze how they function.

The human body is composed of major elements such as carbon, hydrogen, oxygen and nitrogen that combine to form a great variety of molecules called biomolecules.

The four major complex biomolecules are carbohydrates, proteins, lipids and nucleic acids.

It is very important to know the nature and functions of these biomolecules.

Biochemistry can be defined as: -

The study of molecules in living matter which is one of the famous sciences for these reasons: -

1. understanding the basic construction many part in biological science.
2. to increasing correlation of biochemical in many field like medicine and engineering like the study of enzymes and their effect on medical diagnosis and part of engineering projects that belong (belongs) to environmental engineering.

Bio molecules

Polymeric biomolecules

Protein, Amino Acid and Polysaccharide can be considered as polymers which composed from one monomer or more

Studies proved that 90% or more from the dried weight of living organisms is a polymer
it can be explained:—

1. Proteins :- They are Polymers consisting of Amino acid connected with each other by Peptide bonds. each Protein has two terminals N-terminal and C-terminal

2. Nucleic Acid: These are two types of nucleic acid

RNA ribosomal nucleic acid

DNA Deoxy ribosomal nucleic acid

They are Polymers produced from nucleotide linkage by bonds named Phosphodiester bond which is play a role of translating genetic information growth, improvement, Reproduction

4. Poly saccharide

Polymers consist of saccharide joined with each other by glycoside bonds Poly saccharide considered source of energy.

Poly saccharide stored as starch in plants, glycogen in animals and synthesized build in plants as cellulose.

LIPIDS

6.1 Introduction

Lipids form a group of organic compounds which are widely distributed in living organisms. Chemically they are esters of fatty acids or are capable of forming esters. The important lipids are triacylglycerol, phospholipids, sterols and fatty acids.

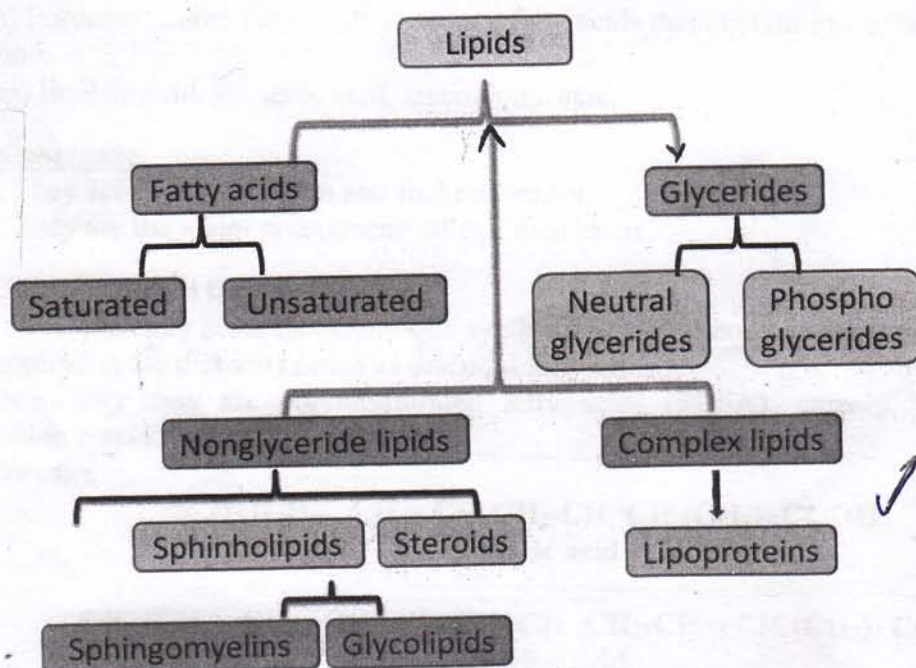
Lipids
(Greek, "lipos", fat)
Substances of biological origin that have the common property of being soluble in non-polar solvents.

6.2 Fatty acids

Fatty acids are carboxylic acid with hydrocarbon side chains. They are the simplest form of lipids and they are water soluble. They exist in the body either as free acids or fatty acyl esters such as triacylglycerol. The fatty acids are released from these lipids on hydrolysis by lipases.

6.2.1 Classification

Fatty acids may be divided into (1) saturated fatty acids and (2) unsaturated fatty acids.



6.2.1.1 Saturated fatty acids

These are fatty acids which do not contain double bonds. They have general formula $C_nH_{2n+1}COOH$ (Table 6.1).

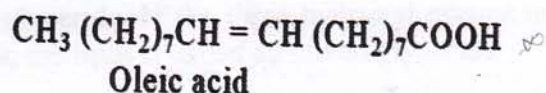
Table 6.1 Saturated fatty acids commonly found in natural fats.

Acid	Formula	Carbon atoms
Acetic	CH_3COOH	2
Propionic	$\text{C}_2\text{H}_5\text{COOH}$	3
Butyric	$\text{C}_3\text{H}_7\text{COOH}$	4
Caproic	$\text{C}_5\text{H}_{11}\text{COOH}$	6
Caprylic	$\text{C}_7\text{H}_{15}\text{COOH}$	8
Decanoic	$\text{C}_9\text{H}_{19}\text{COOH}$	10
Lauric	$\text{C}_{11}\text{H}_{23}\text{COOH}$	12
Myristic	$\text{C}_{13}\text{H}_{27}\text{COOH}$	14
Palmitic	$\text{C}_{15}\text{H}_{31}\text{COOH}$	16
Stearic	$\text{C}_{17}\text{H}_{35}\text{COOH}$	18
Arachidic	$\text{C}_{19}\text{H}_{39}\text{COOH}$	20
Behenic	$\text{C}_{21}\text{H}_{43}\text{COOH}$	22
Lignoceric	$\text{C}_{23}\text{H}_{47}\text{COOH}$	24

6.2.1.2 Unsaturated fatty acid

These are fatty acids which contain double bonds. They have general formula $(\text{C}_n\text{H}_{2n-1}\text{COOH})$. They are subdivided into:

- (a) Monounsaturated fatty acid: These are fatty acids containing one double bond.
(eg) Oleic acid.



- (b) Polyunsaturated fatty acid: These are fatty acids that contain more than one double bond.

(eg) linoleic acid, linolenic acid, arachidonic acid.

Importance

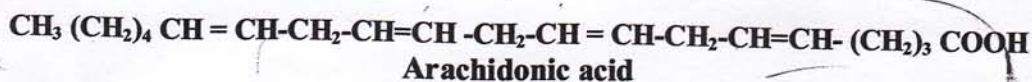
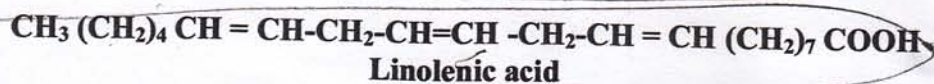
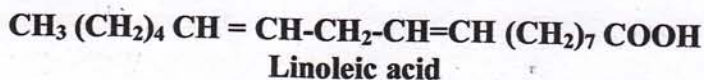
1. They act as energy stores and fuel molecules.
2. They are the major components of cell membrane.

6.2.1.3 Essential fatty acid (EFA)

The fatty acids that cannot be synthesized by the body and therefore should be supplied in the diet are known as essential fatty acids.

Chemically they are polyunsaturated fatty acids (PUFA), namely linoleic acid, linolenic acid and arachidonic acid.

Structure

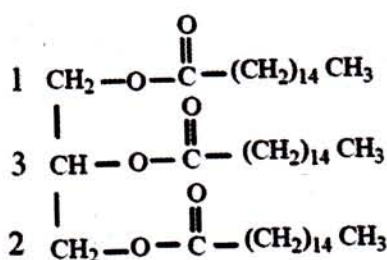


Functions

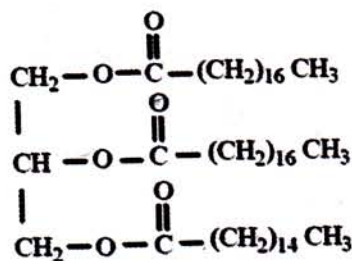
1. EFAs are required for the membrane structure and functions.
2. They are necessary for the maintenance of growth, reproduction and good health.
3. They are important for the transport of cholesterol, formation of lipoprotein and prevention of fatty liver.
4. They serve as precursor for prostaglandin biosynthesis.
5. They prolong clotting time and increase fibrinolytic activity.

6.3 Structure of triacylglycerol

Triacylglycerols are simple lipids in which glycerol backbone is esterified with three fatty acids. This form the major part of dietary lipids. They are stored in adipose tissue and serve as concentrated fuel reserve of the body.



Tripalmitin (simple triacyl glycerol)



Distearopalmitin (mixed triacylglycerol)

If the three hydroxyl groups are esterified with same type of fatty acid then the lipid is called as simple glyceride. If the three hydroxyl groups are esterified with different type of fatty acids, the lipid is called as mixed glyceride.

6.3.1 Properties

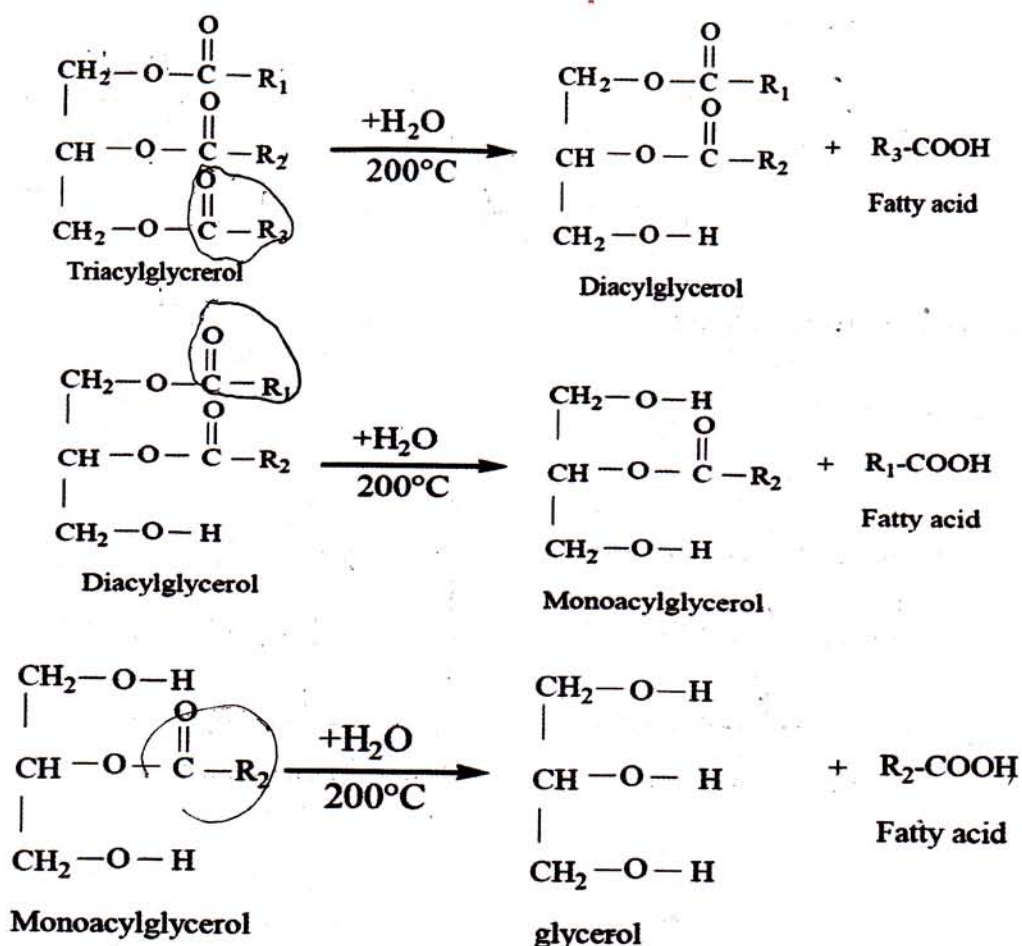
6.3.1.1 Physical

1. Triacylglycerols are nonpolar, hydrophobic molecules, insoluble in water, but soluble in organic solvents.
2. Specific gravity of fats is lower than water. Therefore fats and oils float on water.
3. Melting point of triacylglycerol is related to the chain length and degree of unsaturation of fatty acids. The longer the chain length, the higher the melting point and greater the number of double bonds, the lower the melting point.
4. They are tasteless, odorless, colorless and neutral in solution.
5. They are themselves good solvents for other fats.

6.3.1.2 Chemical properties

1. Hydrolysis:

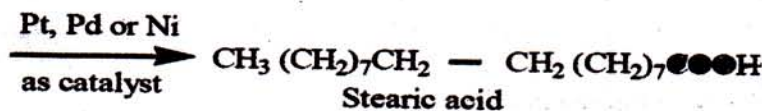
On boiling with water at 200°C, triacylglycerols are hydrolyzed to glycerol and fatty acids in a stepwise manner.



The reaction can also be catalyzed by the enzymes lipases.

2. Hydrogenation

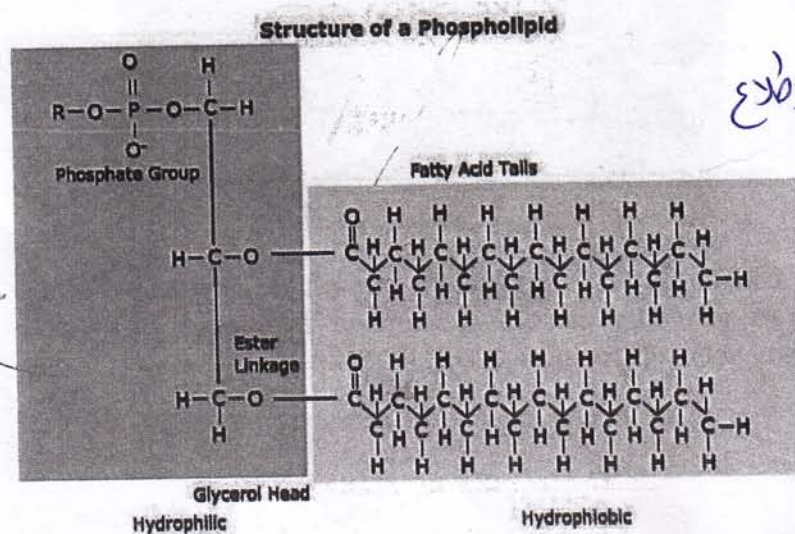
Hydrogenation of unsaturated fatty acids present in the fats, lead to the formation of saturated fats. Hydrogenation elevates the melting point. Thus an oil is converted to a solid fat.



This reaction is of great commercial importance since it permits transformation of inexpensive and unsaturated liquid vegetable fats into solid fats. The latter are used in the manufacture of candles, vegetable shortenings like Vanaspati and oleomargarine.

6.4 Phospholipids

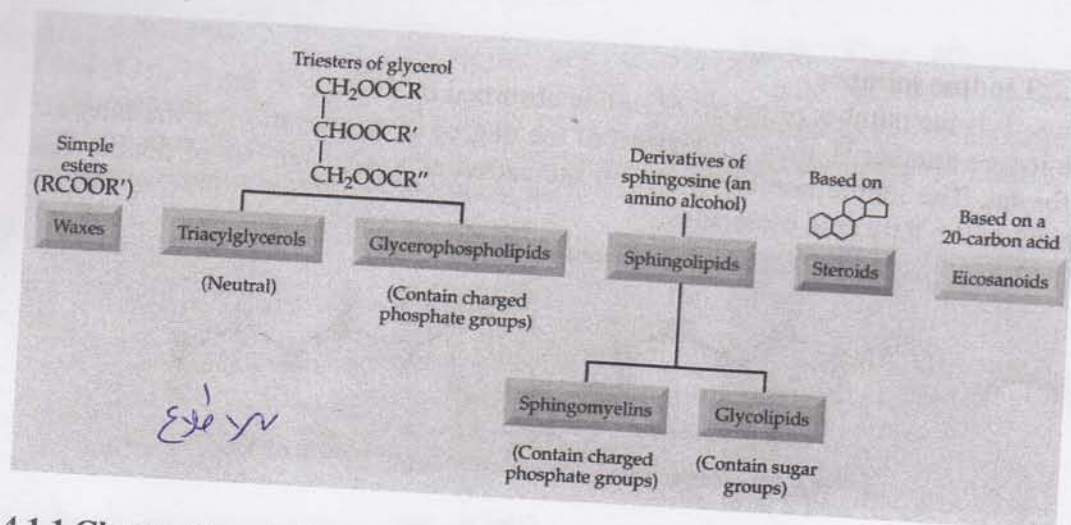
Phospholipids are compound lipids containing phosphoric acid in addition to fatty acid, alcohol and a nitrogenous base.



6.4.1 Classification

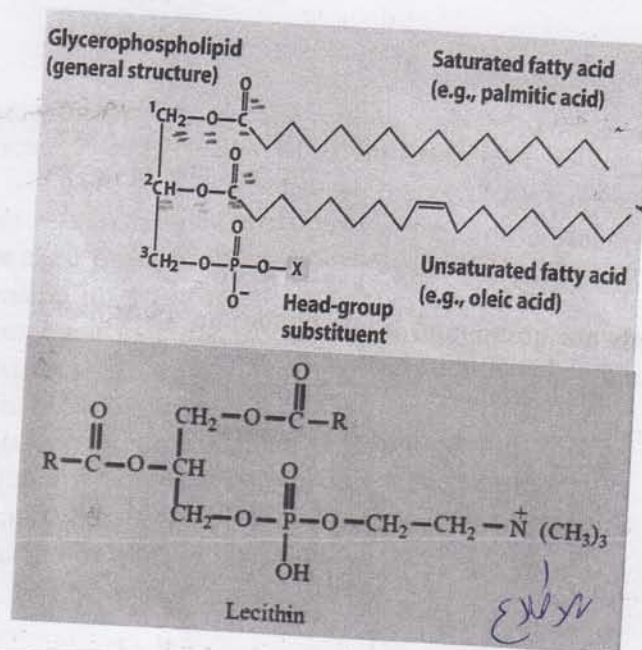
Phospholipids are classified into two types.

1. Glycerophospholipids (or) Phosphoglycerides that contain glycerol as alcohol.
2. Sphingophospholipids that contain sphingosine as alcohol.



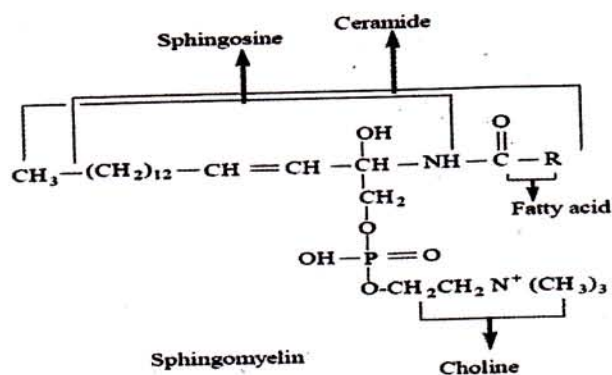
6.4.1.1 Glycerophospholipids

These are the major lipids that occur in biological membranes. They present in all plant and animal cells. They are abundantly present in heart, brain, kidney, egg yolk and soybean. The important glycerophospholipids are lecithin, cephalin, phosphatidyl inositol, cardiolipin and plasmalogen.



6.4.1.2 Spingophospholipids

These are present in plasma membrane and myelin sheath. They are amphipathic lipids having polar head and non-polar tail. They contain an amino alcohol called shingosine. It is attached to a fatty acid by an amide linkage to form ceramide. Ceramide is linked to phosphorylcholine to form sphingomyelin, which is an important member of sphingophospholipids.



6.4.4 Importance of phospholipids

1. They form the structural components of membrane and regulate membrane permeability.
2. They play an important role in cellular respiration.
3. They participate in the absorption of fat from the intestine.
4. They act as surface tension lowering agent.
5. They are essential components of bile where they act as detergents and help in the solubilisation of cholesterol.
6. They also participate in blood clotting.
7. They protect and insulate the neuronal fibers of myelin sheath.
8. They are involved in the interaction of hormones with receptors.
9. They can act as lipotropic agents and prevent fatty liver formation.
10. They help in the reverse transport of cholesterol.

6.5.1 Cholesterol

Cholesterol is exclusively found in animals and is the most abundant animal sterols. It is widely distributed in all cells and is a major component of cell membrane and lipoproteins. In human beings, it is very important to control the normal level of cholesterol in blood.

6.5.1.1 Structure

Cholesterol is a C_{27} ($C_{27}H_{46}O$) compound. It has one hydroxyl group at C_3 and a double bond between C_5 and C_6 . An aliphatic side chain is attached to C_{17} . Cholesterol contains a total of 5 methyl groups (Fig. 7.1).

Cholesterol is the precursor of various physiologically important compounds such as bile acids, vitamin-D, steroid hormones etc.

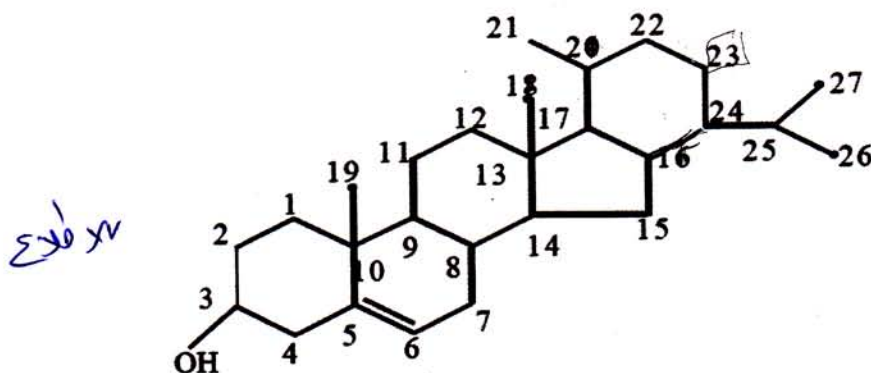


Fig. 7.1 Structure of cholesterol

6.5.1.2 Properties

1. They are white shining rhombic plate like crystals.
2. It is tasteless and odorless
3. It has a high melting point of 150°C .
4. It is insoluble in water and soluble in fat solvents.
5. It is a poor conductor of heat and electricity and serves as an insulator against electric charge. In brain, where it is present abundantly, it acts as an insulator against nerve impulse which are electrical in nature.
6. Cholesterol, when oxidized under suitable conditions, undergoes rapid oxidation to form a ketone-cholestenone.
7. The hydroxyl group of cholesterol readily forms ester with fatty acids, stearic acid etc.
8. It gives addition reactions such as hydrogenation and halogenation because of the presence of double bond.

6.5.1.3 Physiological importance of cholesterol

1. It is one of the essential constituents of cells.
2. It influences the permeability functions of the cell.
3. It controls the red cells from being easily hemolysis.
4. It performs defensive action.
5. It assists the formation of bile acids and bile salts, 7- dehydrocholesterol, vitamin D3, corticosteroid hormones, androgens, estrogens and progesterone.
6. It acts as an antagonist to phospholipid.

CHAPTER 2

Carbohydrates: Structure and function

Introduction

Carbohydrates are widely distributed in plants in which they are formed from carbon dioxide of the atmosphere and water by photosynthesis.

Plants use carbohydrates as the precursor for the synthesis of proteins, lipids and other organic compounds. Animals obtain their carbohydrates from plants.

Functions of carbohydrates

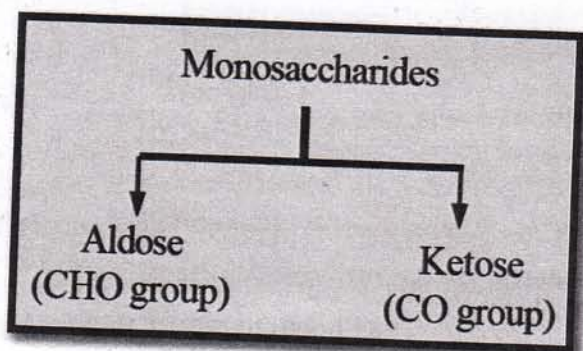
Carbohydrates have a variety of functions in the animal and human body.

1. They supply energy for body functions and for doing work.
2. They are structural components of many organisms.
3. They exert a sparing action on proteins.
4. They provide the carbon skeleton for the synthesis of some nonessential amino acids and fats.
5. Some carbohydrates are present as tissue constituents.
6. Starch forms main source of carbohydrates in the diet.

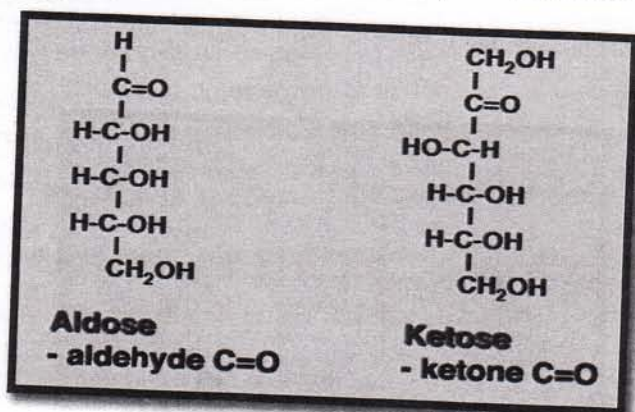
Classification of Carbohydrates.

3.3.1 Monosaccharides

Monosaccharaides are defined as polyhydroxy aldehydes or ketones, which cannot be further hydrolyzed to simple sugars. Monosaccharaides are divided into two groups according to their functional groups (Fig. 3.2).

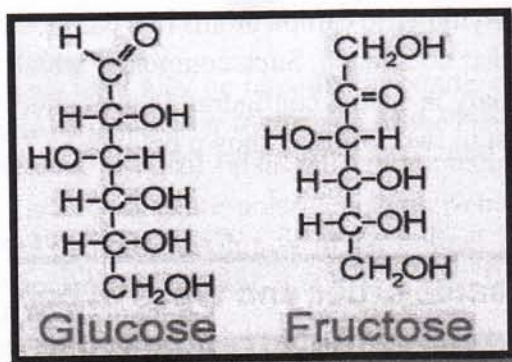


Classification of monosaccharides.



They are also classified based on the number of carbon atoms present in the monosaccharaides.

Carbon Atoms	General term	Aldehyde	Ketone
3	Triose	Aldotriose	Ketotriose
4	Tetrose	Aldotetrose	Ketotetrose
5	Pentose	Aldopentose	Ketopentose
6	Hexose	Aldohexose	Ketohexose
7	Heptose	Aldoheptose	Ketoheptose



Structure of glucose and fructose.

Glucose is a simple sugar. It is a monosaccharide. It cannot be hydrolyzed further. Glucose is an important sugar of blood.

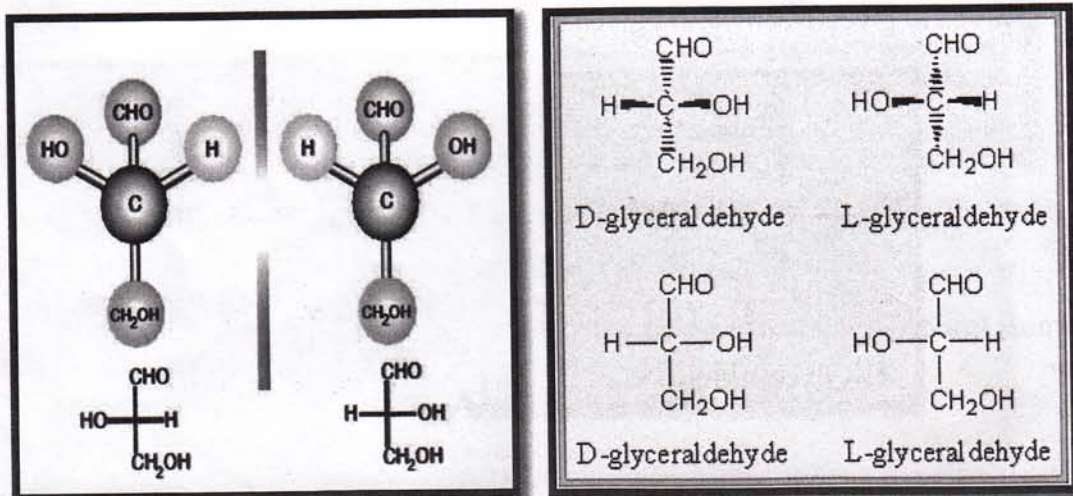
Human blood contains 60-100 mg of glucose in 100 ml of blood in fasting. It serves as the major metabolic fuel in cells and tissues.

Oxidation of glucose quickly provides energy for the cells. Hence, glucose is described as the chief source of energy.

The empirical formula of glucose is CH_2O and the molecular formula is $\text{C}_6\text{H}_{12}\text{O}_6$. The molecular formula of fructose is $\text{C}_6\text{H}_{12}\text{O}_6$. Glucose has aldehyde group whereas fructose has ketone group.

Physical properties of monosaccharaides

1. Color and shape: monosaccharide are colorless and crystalline compounds.
2. Solubility: they are readily soluble in water.
3. Taste: they have sweet taste.
4. Stereo isomerism.



The mutarotation is due to the existence of two optical isomers of glucose, namely α - D glucose with a specific rotation $+112.2^\circ$ and β - D glucose with a specific rotation $+18.7^\circ$.

α and β isomers are called as anomers and the carbon atom responsible for this is the anomeric carbon atom. Anomers are isomers differing in configuration of a particular carbon atom alone.

A freshly prepared aqueous solution of α - D glucose has a specific rotation of $+112.2^\circ$. When this solution is allowed to stand, the rotation falls to 52.7° and remains constant at this value. This gradual change in specific rotation is called mutarotation.

The value of mutarotation for α , D-glucose is $+59.5^\circ$. $(+112.2^\circ) - (52.7^\circ) = +59.5^\circ$.

A freshly prepared solution of β , D glucose has a rotation value of 18.7° . It also gradually increases and reaches the same final value of $+52.7^\circ$.

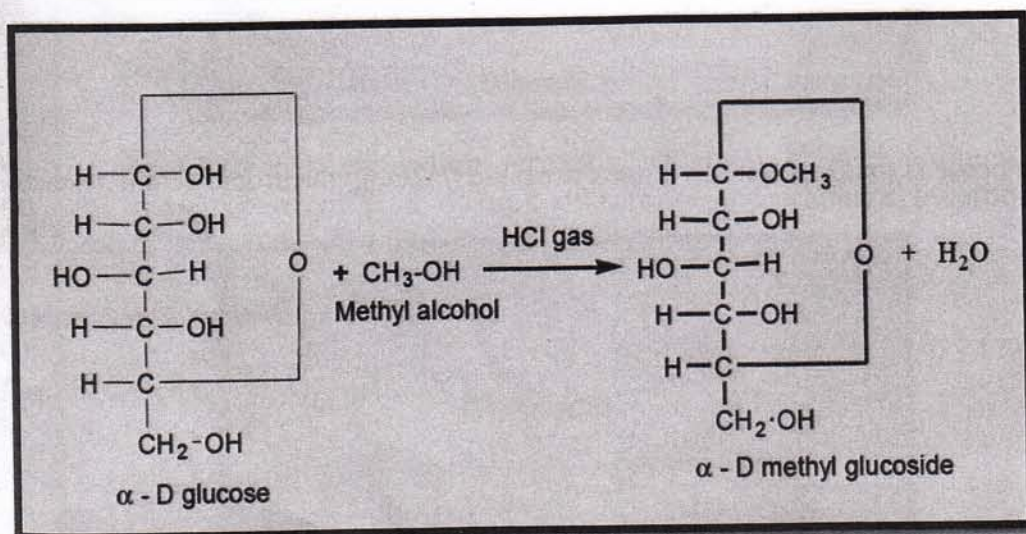
Chemical properties of glucose

Glucose contain active groups. The active groups are responsible for their chemical properties. There are three types of active groups in glucose. They are:

1. Glycosidic -OH group
2. Alcoholic -OH group
3. Aldehyde - C-H group
 $\begin{array}{c} \text{O} \\ \parallel \\ \text{C-H} \end{array}$

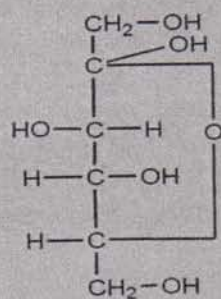
1. Glucoside formation

Glucose reacts with methanol in the presence of HCl and gives α and β glucoside. Glucoside formation is due to the reaction of alcohol with glucoside -OH group of glucose.

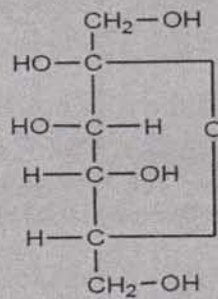


β -D glucose is forms β , D-methyl glucoside. In the same way, fructose forms fructoside.

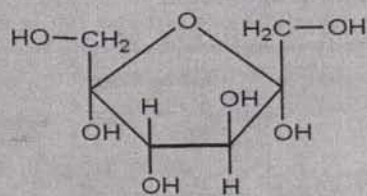
2. Fructose



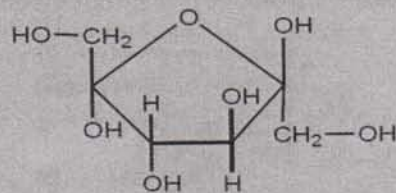
α D - Fructose



β D - Fructose



α D - Fructofuranose

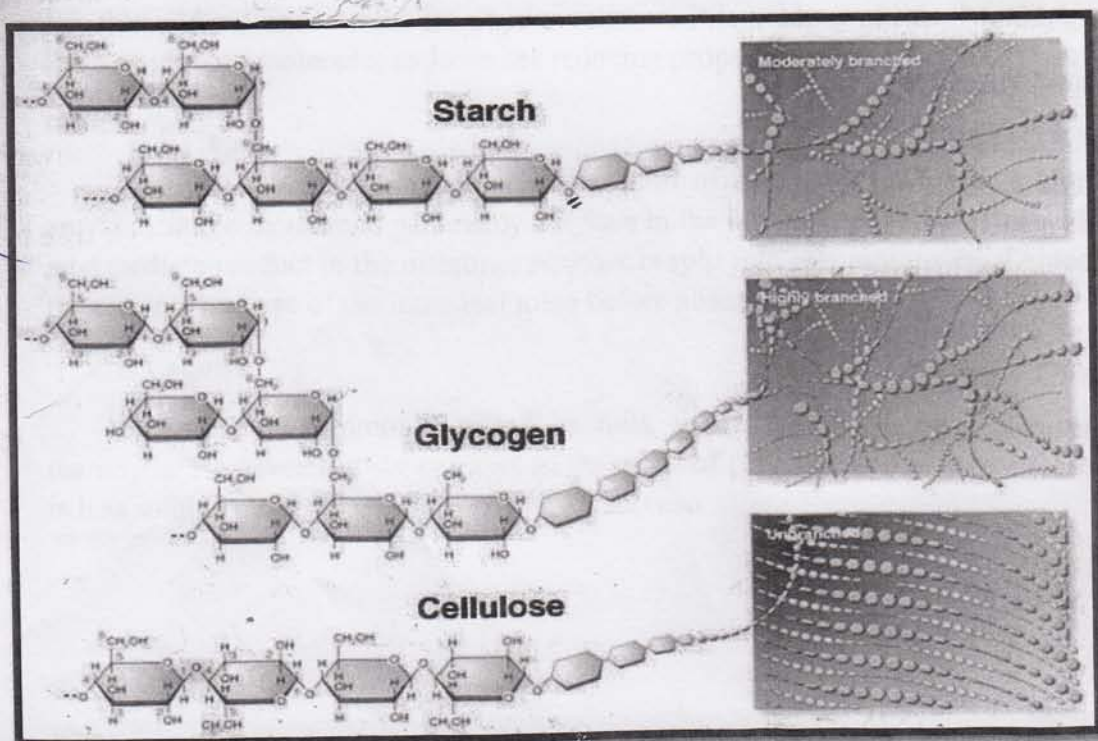


β D - Fructofuranose

Pyranose structure of fructose.

Disaccharides

Disaccharides are sugars containing two molecules of monosaccharaides. Disaccharides are formed by the condensation of two molecules of monosaccharaides with the elimination of one molecule of water.

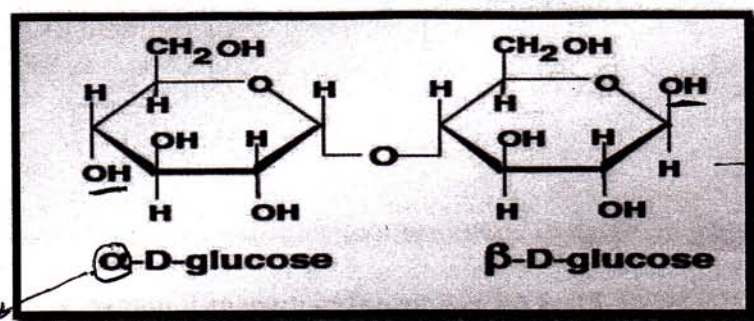


In disaccharides, monosaccharides are linked by the glycosidic bonds. The properties of the disaccharides depend to a great extent on the type of linkage. If the two potential aldehyde or ketone of both monosaccharides are involved in the linkage, the sugar will not exhibit reducing properties and will not be able to form osazones. eg. Sucrose.

But if one of them is not bound in this way, it will permit reduction and osazone formation by the sugars eg. Lactose and maltose which are known as reducing disaccharides.

Maltose

Maltose is composed of two glucose molecules combined by α -1,4 glycosidic linkage. It is commonly called malt sugar. Malt from sprouting barley is the major source of maltose. It is a rather sweet sugar and is highly soluble in water.



(α -1,4 glycosidic linkage)

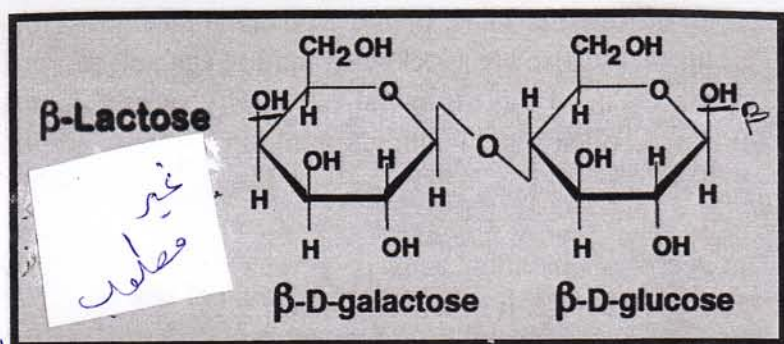
The structure of maltose shows that the potential aldehyde group of glucose -2 is blocked in the glycosidic linkage, whereas the potential aldehyde group of glucose -1 is free and can reduce alkaline copper solution. It is because of this free aldehyde in the first glucose molecule, maltose has reducing property.

Metabolism

Maltose is the end product of digestion of starch by the action of salivary amylase, in the mouth and pancreatic amylase in the intestine. Maltose is formed as an intermediate product in the intestine. Maltose is split into two molecules of glucose by the enzyme maltase of the intestinal juice before absorption.

Lactose

Lactose is commonly called as milk sugar. It is present in the milk of mammals. However lactose is found in the urine of pregnant and lactating women. It is less soluble in water and less sweet than sucrose.



(β, 1-4 glycosidic linkage)

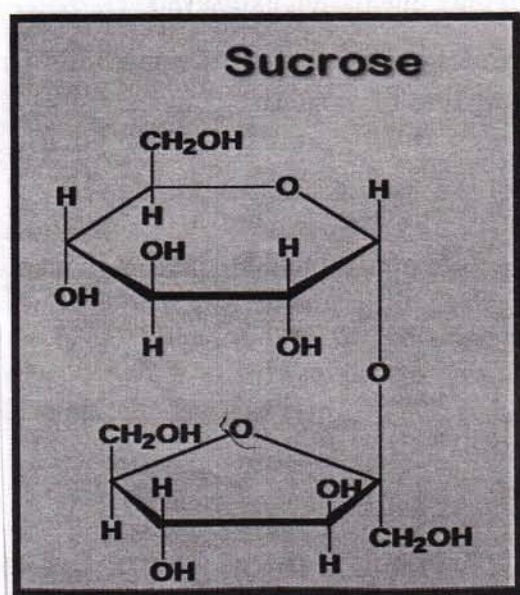
Just as in maltose, lactose has a free potential aldehyde group in the glucose molecule, not involved in the glycosidic linkage between glucose and galactose molecules. Whereas, the potential aldehyde group of galactose molecule is blocked in the linkage. Because of the presence of free aldehyde group in the glucose molecule, lactose can reduce Fehling's solution and is therefore a reducing sugar.

Metabolism

When lactose is hydrolyzed by acids or by the enzyme lactase, one molecule of glucose and one molecule of galactose are formed. The intestine of milk sucking infants has the enzyme lactase, which converts lactose into glucose and galactose. Then only it is absorbed in the body. Excess of lactose ingested into the body causes diarrhea, abnormal intestinal flow and colic pain. Lactose is not fermented by yeast.

Sucrose

Sucrose is ordinary "table sugar". It is also called as "cane sugar" as it can be obtained from sugar cane. It is widely distributed in sugar cane, beet root, pine apple, honey, carrot and ripe fruits.



Sucrose consists of one molecule of glucose and one molecule of fructose. The linkage between these molecules are formed between the aldehyde group of glucose

and the ketone group of fructose. Thus, both the potential aldehyde group of glucose and the ketone group of fructose are blocked in the linkage and sucrose has no free reducing group. On account of this structural peculiarity sucrose is a non-reducing sugar. It does not reduce Tollen's and Fehling's solutions and does not form osazone.

Metabolism

Sucrose on hydrolysis by dilute acids or the enzyme sucrase or invertase gives a mixture of glucose and fructose. It is called as invert sugar.

Inversion

Sucrose is dextrorotatory ($+62.5^\circ$) but its hydrolytic products are levorotatory because fructose has a greater specific levo-rotation than the dextrorotation of glucose. As the hydrolytic products invert the rotation, sucrose is known as invert sugar and the process is called as inversion. Honey contains plenty of 'invert sugar' and the presence of fructose accounts for the greater sweetness of honey.

Polysaccharides

Polysaccharides, which are also known as glycan composed of number of monosaccharide units. They represent condensation products of several molecules of simple sugars or monosaccharaides. Monosaccharaides are linked together by glycosidic bonds in polysaccharides. They form linear chain or branched chain molecules.

They contain only one type of monosaccharide units or many types of monosaccharide units. According to this nature polysaccharides are classified into two groups, homo polysaccharides and hetero polysaccharides.

Homo polysaccharides are composed of only one type of monosaccharaides. On hydrolysis they yield only one type of monosaccharaides Eg. starch, glycogen, cellulose etc. which yield only glucose on hydrolysis.

Hetero polysaccharides are composed of a mixture of monosaccharaides. On hydrolysis, they yield a mixture of monosaccharaides. Eg. Hyaluronic acid, Heparin, Muco polysaccharides.

Starch

This is the storage form of carbohydrate present in plants. They are abundantly found in root, stem, vegetables, fruits and cereals. The bulk of our diet which consists mainly of rice, wheat and vegetables is a good source of starch.

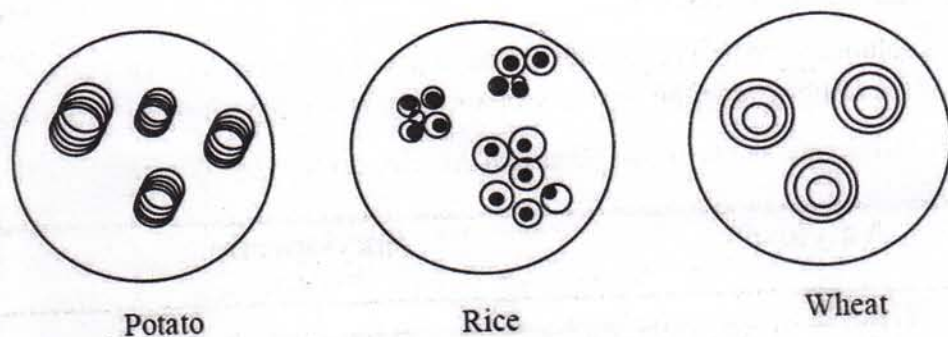
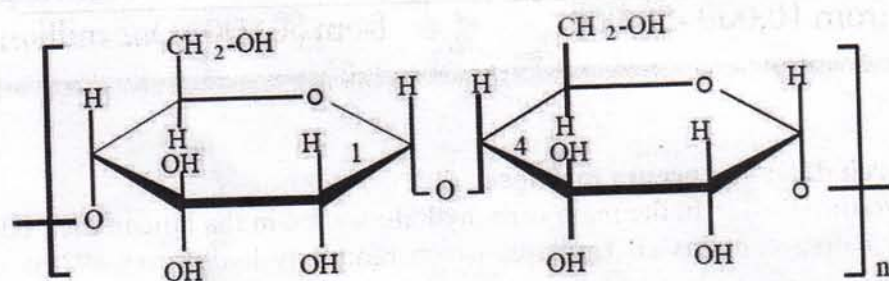


Fig. 3.7 Starch grains under microscope.

Starch occurs in the forms of grain which may be spherical or oval in shape. Microscopically, the starch grains are found to differ in size and shape according to their sources (Fig 3.7).

Starch is made up of two structurally different homo polysaccharide units. They are amylose and amylopectin. In amylose, glucose molecules are arranged in a linear form. Glucose molecules are arranged in a highly branched form in amylopectin.



Amylose has 1, 4 glycosidic linkages. The glycosidic - OH group of C1 in one glucose unit is joined to that of C4 of the next unit (Fig 3.8).

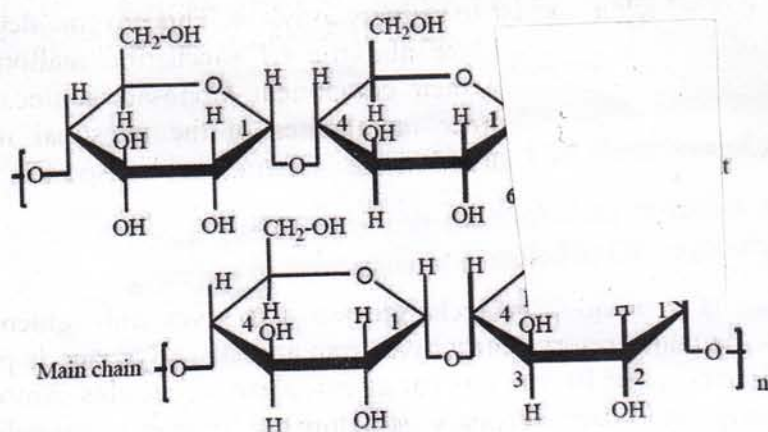


Fig. 3.9 Structure of Amylopectin (α 1-4 and α 1-6 glycosidic linkages).

... of branching has substituents at carbon atoms 1.

Glycogen

Glycogen is a homo polysaccharide since it gives only glucose units on hydrolysis. It is the major reserve carbohydrate in animals. Glycogen is present in all cells of skeletal muscle and liver and occur as cytoplasmic granules. Among plants, it is found in fungi and yeast. Primary structure of glycogen resembles that of amylopectin but glycogen is more highly branched, with branch points occurring every 8 to 12 glucose residues

(Fig. 1.10). Glycogen is readily available as immediate source of energy. During starvation glycogen is mobilized from the storage tissue and converted to glucose by the enzyme glycogen phosphorylase. Glucose formed like this is oxidized

to produce energy. The degree of branching is considerably more in glycogen when compared to starch.

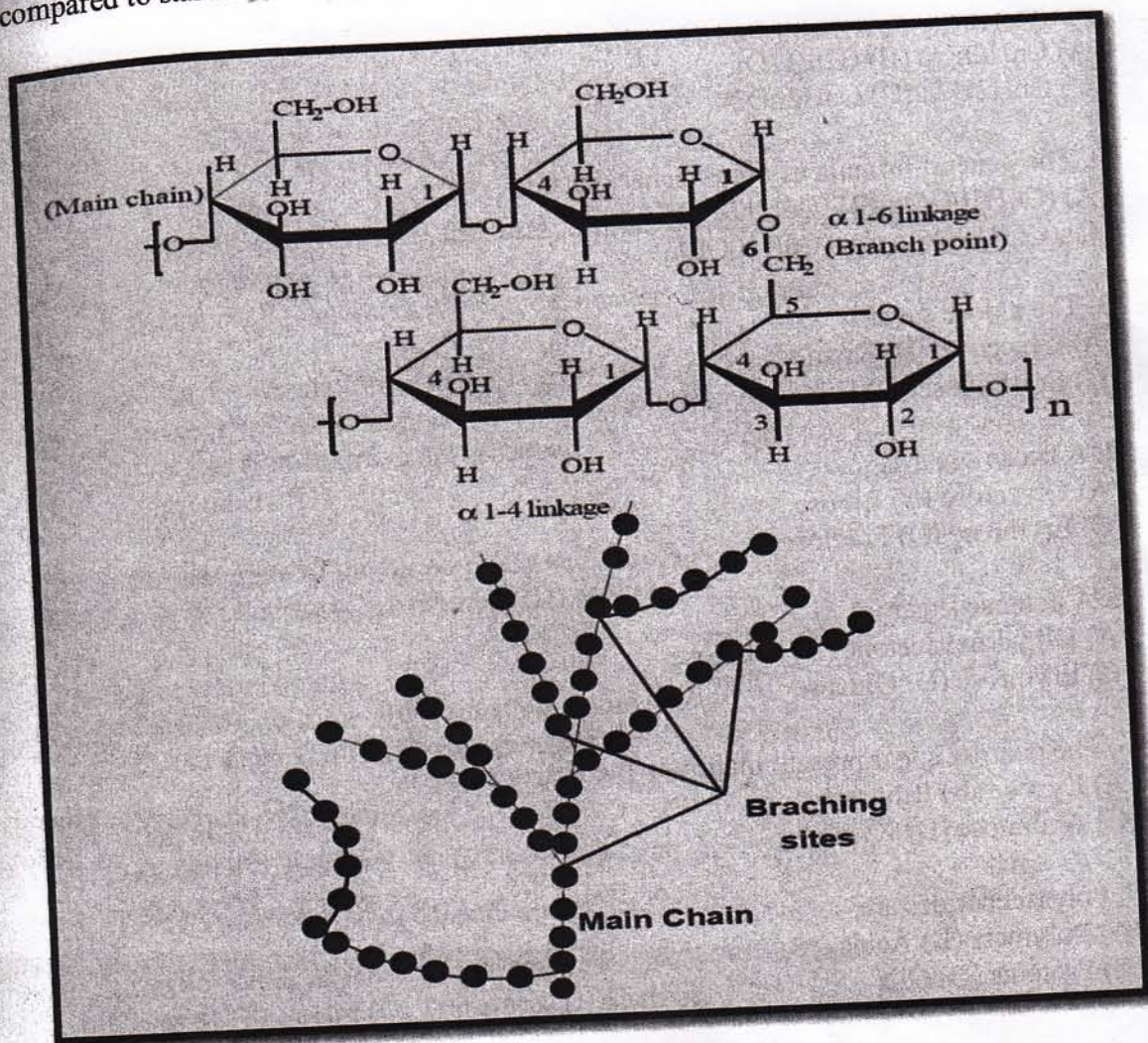


Fig. 1.10 Structure of glycogen.

Formation of glycogen from glucose is called as Glycogenesis and breakdown of glycogen to form glucose is called as Glycogenolysis.