



Knowledge... Everywhere

Chemistry

Bio-Molecules

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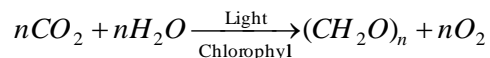


All living bodies are composed of several lifeless substances which are present in their cells in a very complex but highly organized form. These are called biomolecules. Some common examples are carbohydrates, proteins, enzymes, nucleic acids, lipids, amino acids, fats etc:

Living organisms → Organs → Tissues → Cells → Organelles → Biomolecules.

1. Carbohydrates.

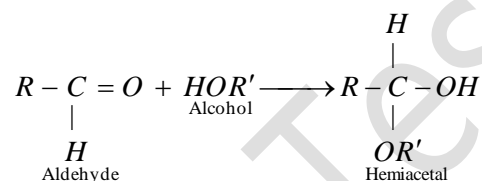
The carbohydrates are naturally occurring organic substances. They are present in both plants and animals. The dry mass of plants is composed of 50 to 80% of the polymeric carbohydrate cellulose. Carbohydrates are formed in the plants by photosynthesis from carbon dioxide and water.



Animals do not synthesise carbohydrates but depends on plants for their supply.

(1) **Definition:** "Carbohydrates are defined as a class of compounds that include polyhydric aldehydes or polyhydric ketones and large polymeric compounds that can be broken down (hydrolysed) into polyhydric aldehydes or ketones."

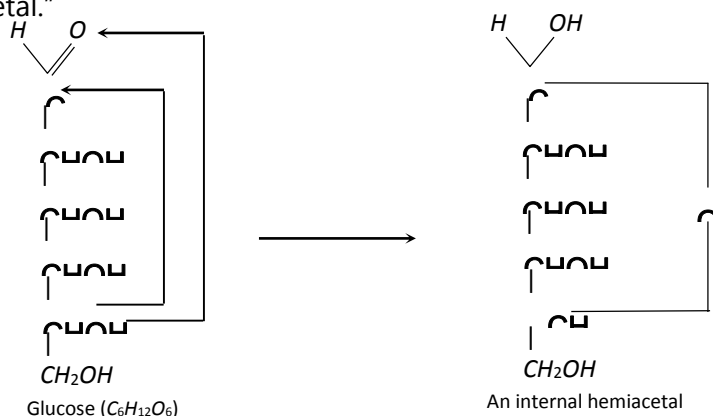
Carbohydrates contain $>C=O$ and $-OH$ groups. A carbonyl compound reacts with an alcohol to form hemiacetal.



In carbohydrates, the carbonyl group combine with an alcoholic group of the same molecules to form an internal hemiacetal thus the correct definition of carbohydrates is as follows



"A polyhydroxy compound that has an aldehydic or a ketonic functional group either free or as hemiacetal or acetal."



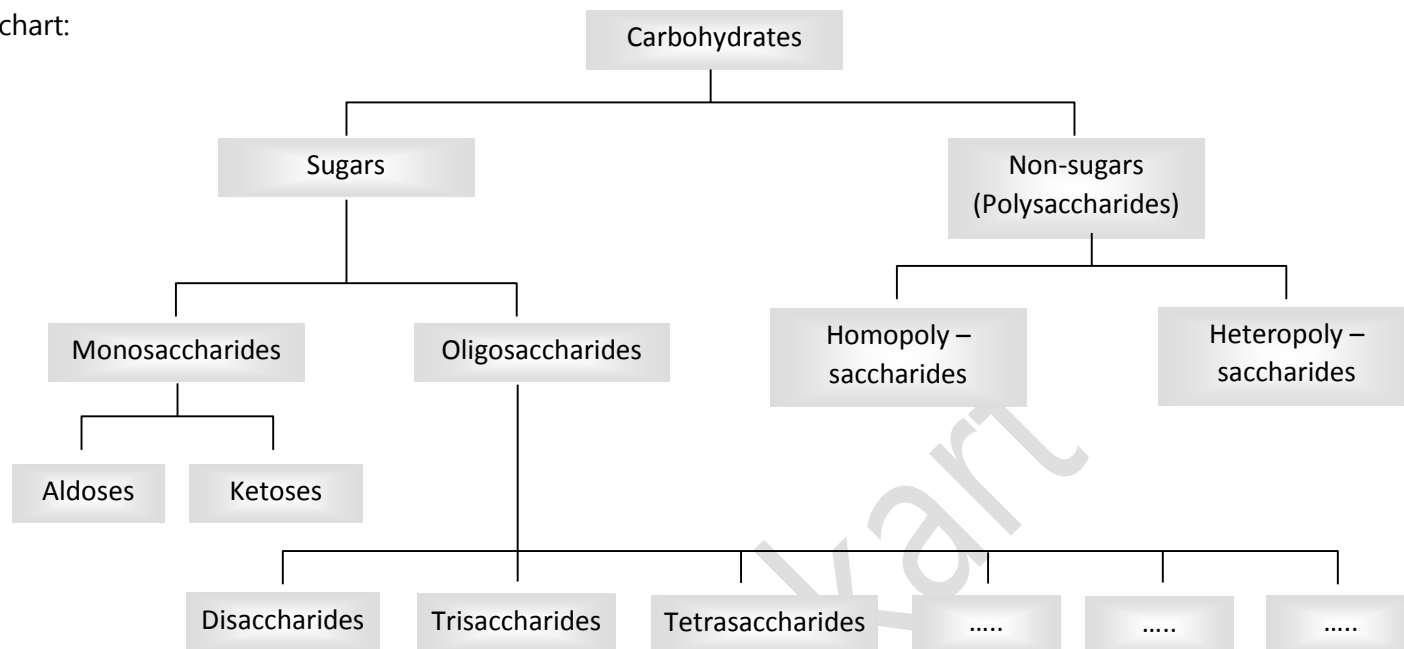
In general, carbohydrates are white solids, sparingly soluble in organic solvents and (except certain polysaccharides) are soluble in water. Many carbohydrates of low molecular masses have a sweet taste.

(2) **Nomenclature:** The name of simpler carbohydrates end is -ose. Carbohydrate with an aldehydic structure are known as aldoses and those with ketonic structure as ketoses. The number of carbon atom in the molecule is indicated by Greek prefix.

Number of carbon atoms in the molecule	Aldose	Ketose
3	Aldotriose	Ketotriose
4	Aldotetrose	Ketotetrose
5	Aldopentose	Ketopentose
6	Aldohexose	Ketohexose
7	Aldoheptose	Ketoheptose

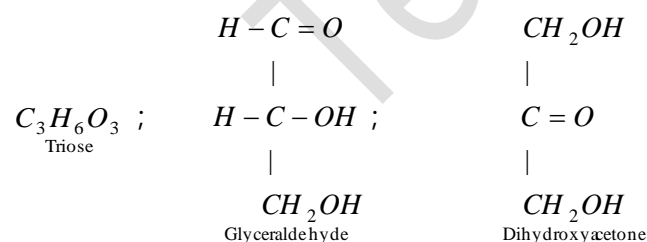


(3) **Classification:** The complete classification of carbohydrates may be depicted in short in the following chart:



2. Monosaccharides.

These are the simplest one unit non-hydrolysable sugars. They have the general formula $C_nH_{2n}O_n$ where n varies from 3 to 9 carbon atoms. About 20 monosaccharides occur in nature. The simplest are trioses ($n=3$)



The most important naturally occurring monosaccharides are pentoses and hexoses. A common pentose is ribose and two common hexoses are glucose and fructose.

Except ketotriose {dihydroxyacetone}, all aldose and ketoses {monosaccharides} contain asymmetric carbon atoms and are optically active. Number of isomers depend upon the number of asymmetric carbon atom in the molecules of monosaccharide and is derived by the formula 2^n where n is the number of asymmetric carbon atoms in the molecules.



Number of carbon atoms		No. of asymmetric carbon atoms	No. of isomers
Trioses 3	$\left\{ \begin{array}{l} \text{CH}_2\text{OHCHOHCHO} \\ \text{CH}_2\text{OHCOCH}_2\text{OH} \end{array} \right.$	1	2
		–	–
Tetroses 4	$\left\{ \begin{array}{l} \text{CH}_2\text{OH}(\text{CHOH})_2\text{CHO} \\ \text{CH}_2\text{OHCHOHCOCH}_2\text{OH} \end{array} \right.$	2	4
		1	2
Pentoses 5	$\left\{ \begin{array}{l} \text{CH}_2\text{OH}(\text{CHOH})_3\text{CHO} \\ \text{CH}_2\text{OH}(\text{CHOH})_2\text{COCH}_2\text{OH} \end{array} \right.$	3	8
		2	4
Hexoses 6	$\left\{ \begin{array}{l} \text{CH}_2\text{OH}(\text{CHOH})_4\text{CHO} \\ \text{CH}_2\text{OH}(\text{CHOH})_3\text{COCH}_2\text{OH} \end{array} \right.$	4	16
		3	8

Class	Molecular formula	Structural formula	Examples
		Aldoses	
Aldotrioses	$\text{C}_3\text{H}_6\text{O}_3$	$\text{CH}_2\text{OHCHOHCHO}$	Glyceraldehyde
Aldotetroses	$\text{C}_4\text{H}_8\text{O}_4$	$\text{CH}_2\text{OH}(\text{CHOH})_2\text{CHO}$	Erythrose, Threose
Aldopentoses	$\text{C}_5\text{H}_{10}\text{O}_5$	$\text{CH}_2\text{OH}(\text{CHOH})_3\text{CHO}$	Arabinose, Ribose, Xylose, Lyxose
Aldohexoses	$\text{C}_6\text{H}_{12}\text{O}_6$	$\text{CH}_2\text{OH}(\text{CHOH})_4\text{CHO}$	Glucose, Galactose, Mannose, Allose, Talose, Gulose, Idose, etc.
		Ketoses	
Ketotrioses	$\text{C}_3\text{H}_6\text{O}_3$	$\text{CH}_2\text{OHCO}\cdot\text{CH}_2\text{OH}$	Dihydroxyacetone
Ketotetroses	$\text{C}_4\text{H}_8\text{O}_4$	$\text{CH}_2\text{OH}\cdot\text{CO}\cdot\text{CHOH}\cdot\text{CH}_2\text{OH}$	Erythrulose
Ketopentoses	$\text{C}_5\text{H}_{10}\text{O}_5$	$\text{CH}_2\text{OH}\cdot\text{CO}\cdot(\text{CHOH})_2\text{CH}_2\text{OH}$	Ribulose, Xylulose
Ketohexoses	$\text{C}_6\text{H}_{12}\text{O}_6$	$\text{CH}_2\text{OH}\cdot\text{CO}(\text{CHOH})_3\text{CH}_2\text{OH}$	Fructose, Sorbose, Tangatose, Psicose

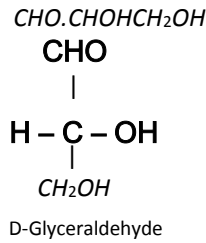
(1) **D and L-designation:** By **convention**, a molecule is assigned D-configuration if the –OH group attached to the carbon adjacent to the –CH₂OH group (last chiral carbon) is on the right hand side irrespective of the position of other groups. On the other hand, the molecule is assigned L-configuration if the –OH group attached to the carbon adjacent to the –CH₂OH group is on the left.

However, it may be noted that D- and L- do not represent dextrorotatory or laevorotatory. The optical activity of the molecule is represented by (+) and (–) which represent the direction of rotation of plane polarized light whether dextrorotatory or laevorotatory.

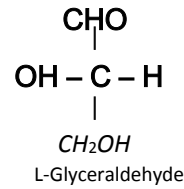


(2) **Configuration:** Configuration of Monosaccharides

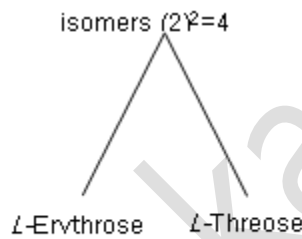
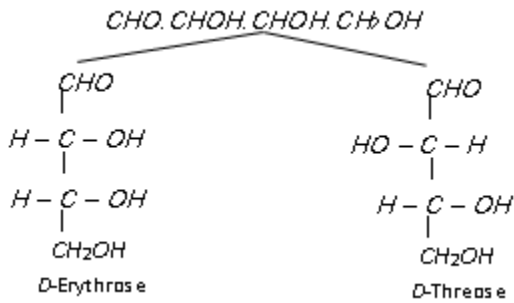
(i) **Aldotriose:**



isomers $(2)^1 = 2$

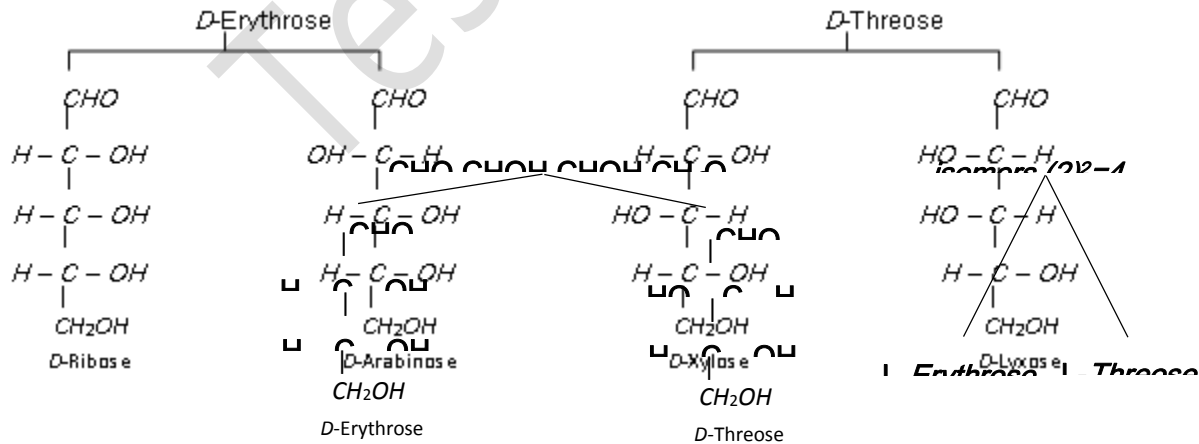


(ii) **Aldotetrose :**



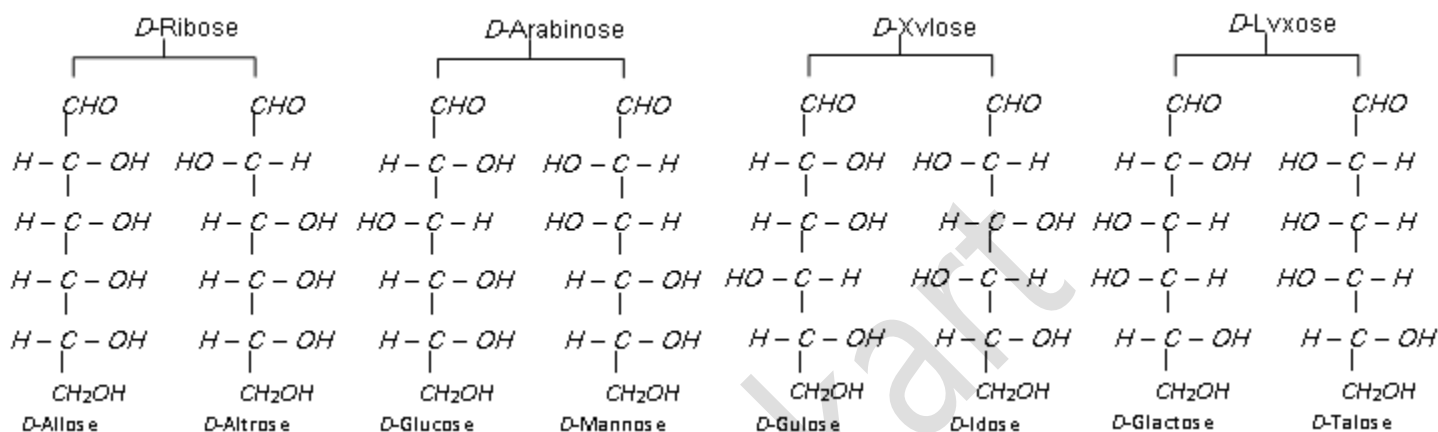
So it has four isomers, i.e., D, L-Erythrose and D, L-Threose.

(iii) **Aldopentose:** CHO.CHOH.CHOH.CHOH.CH₂OH, isomers $(2)^3 = 8$



So aldopentoses has eight isomers, i.e., D- and L-Ribose, D- and L-Arabinose, D- and L-Xylose and D, L-Lyxose

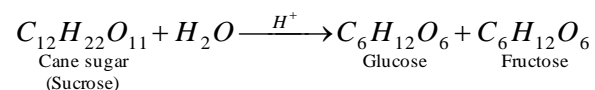
(iv) **Aldohexose:** $\text{CHO} \cdot (\text{CHOH})_4 \text{CH}_2\text{OH}$, isomers $(2)^4 = 16$



(3) **Glucose; $\text{C}_6\text{H}_{12}\text{O}_6$; Aldo-hexose:** Glucose is known as **dextrose** because it occurs in nature as the optically active dextrorotatory isomer. It is also called grape sugar as it is found in most sweet fruits especially grapes. It is present in honey also. It is essential constituent of human blood. The blood normally contains 65 to 110 mg of glucose per 100 mL (hence named Blood sugar). In combined form, it occurs in cane sugar and polysaccharides such as starch and cellulose. It is also present in various glycosides like **amygdalin** and **salicin**.

(i) **Preparation**

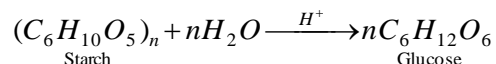
(a) Laboratory method



Note: HCl (dil.) is used for hydrolysis. Glucose being much less soluble in alcohol than fructose separates out by crystallising on cooling.



(b) Manufacture: It is obtained on a large scale by the hydrolysis of starch (corn starch or potato starch) with dilute sulphuric acid or hydrochloric acid.



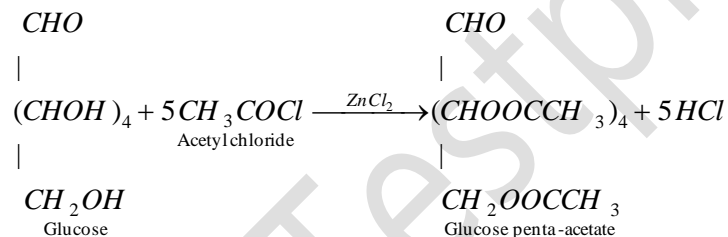
A thin paste of starch is boiled with dilute acid till the hydrolysis is complete. The excess of acid is neutralised with chalk (calcium carbonate) and the filtrate containing glucose is decolourised with animal charcoal. The solution is concentrated and evaporated under reduced pressure. Glucose is obtained in crystalline form.

(ii) **Physical properties:** It is a colourless crystalline solid, melts at $146^\circ C$. It is readily soluble in water. From aqueous solution, it separates as a crystalline monohydrate ($C_6H_{12}O_6 \cdot H_2O$) which melts at $86^\circ C$. It is sparingly soluble in alcohol but insoluble in ether. It is less sweet (three-fourth) than cane sugar. It is optically active and the ordinary naturally occurring form is (+) glucose or dextro form. It shows **mutarotation**.

(iii) **Chemical properties:** Glucose is a polyhydroxy aldehyde i.e. aldohexose. It has five - OH groups and one aldehydic group. It shows characteristics of hydroxyl and aldehydic group. Important chemical reaction of the glucose are the following:

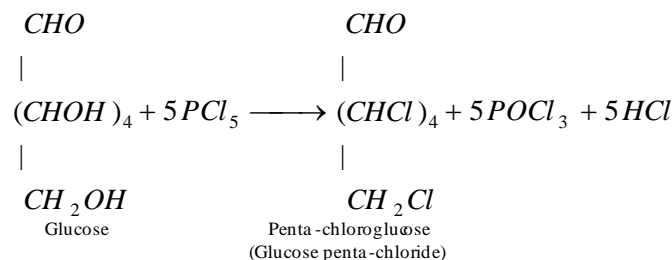
(a) Alcoholic reaction (Reaction due to -OH group)

- Reaction with acid chlorides and acid anhydride



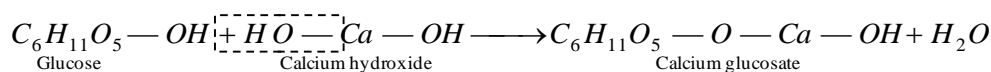
This shows that a molecule of glucose contains 5 - OH groups.

- Reaction with PCl_5

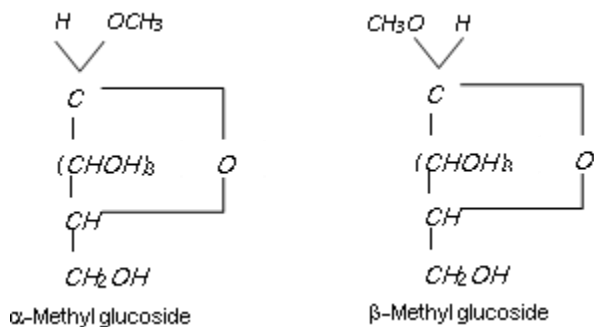
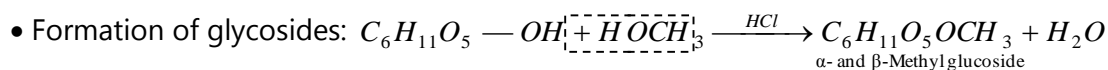


- Reaction with metallic hydroxides:



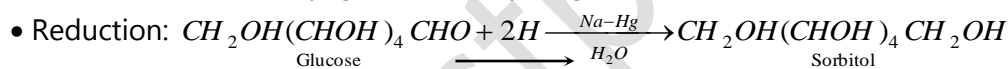


Note: Glucose behaves as a weak acid. Instead of $Ca(OH)_2$ we can take other metallic hydroxide like $Ba(OH)_2, Sr(OH)_2, Cu(OH)_2$ etc to form glucosate which is soluble in water.



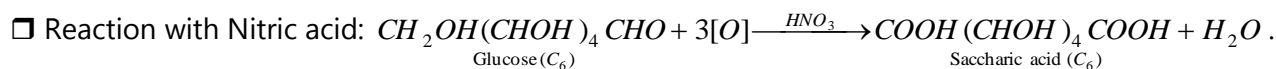
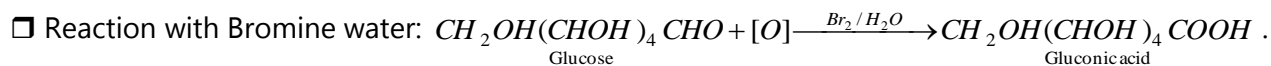
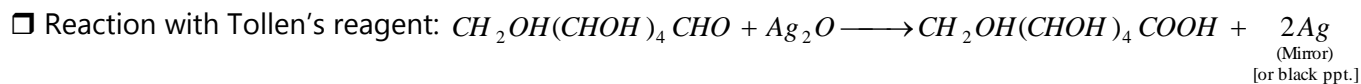
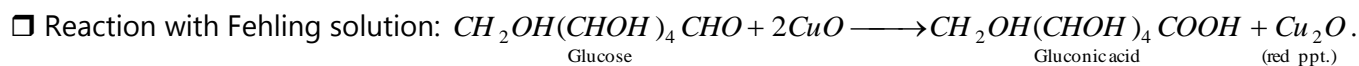
This reaction shows the presence of ring structure in glucose.

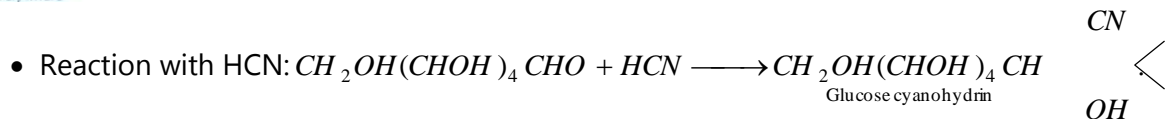
(b) Reactions of carbonyl group (Aldehydic group)



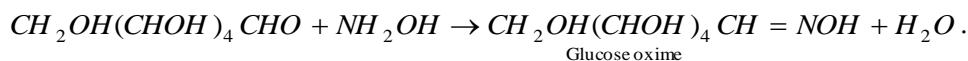
On prolonged heating with concentrated HI and red phosphorus at $110^\circ C$, glucose forms a mixture of 2-iodohexane and n-hexane.

• Oxidation

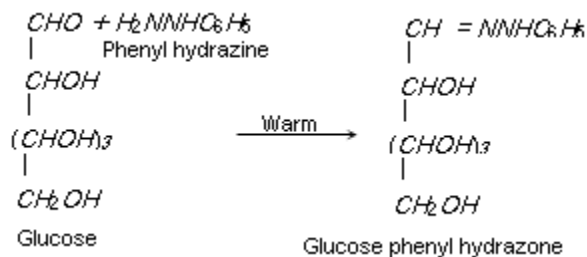




- Reaction with hydroxyl amine

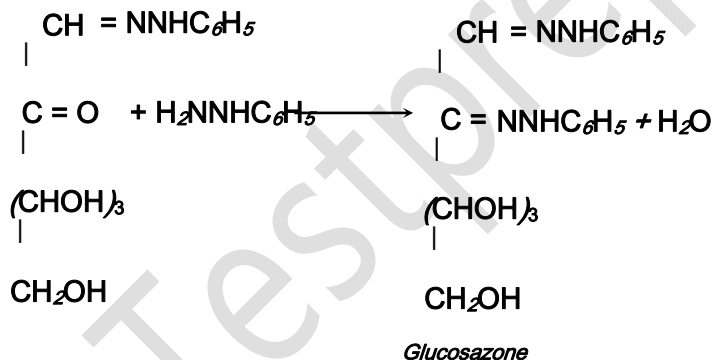


- Reaction with Phenyl hydrazine (Fischer's mechanism): When warmed with excess of phenyl hydrazine, glucose first forms phenylhydrazone by condensation with – CHO group.

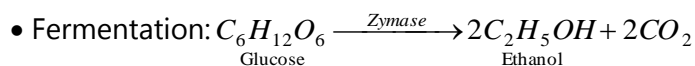


The adjacent –CHOH group is then oxidized by a second molecule of phenyl hydrazine.

The resulting carbonyl compounds reacts with a third molecule of phenyl hydrazine to yield glucosazone.



(c) Miscellaneous reactions

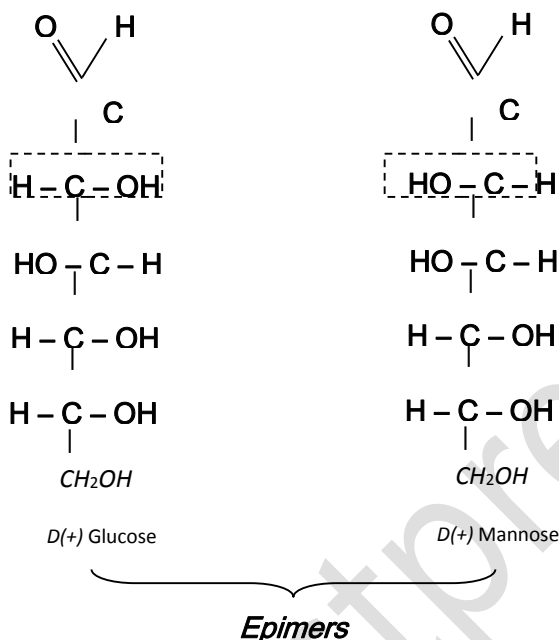
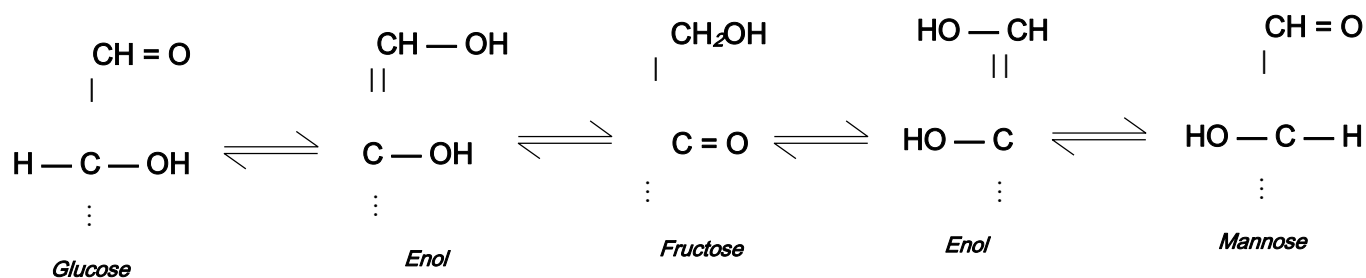


- Dehydration: When heated strongly or when treated with warm concentrated sulphuric acid, glucose is dehydrated to give a black mass (sugar carbon).

- Reaction with alkalis: When warmed with concentrated alkali, glucose first turns yellow; then brown and finally gives a resinous mass.

A dilute solution of glucose, when warmed with dilute solution of alkali, some glucose is converted into fructose and mannose. **D-glucose** and **D-mannose** are **epimers**.

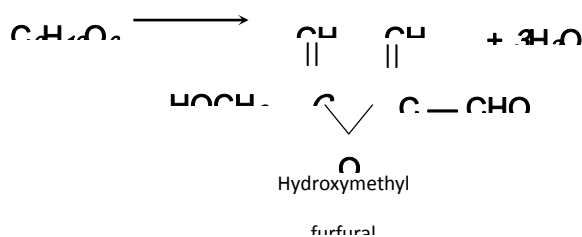




- Action of concentrated hydrochloric acid



On treatment with conc. HCl, glucose can also form hydroxymethyl furfural.



This on acid treatment gives laevulic acid



(iv) **Uses**

- (a) In the preservation of fruits and preparation of jams and jellies.
- (b) In the preparation of confectionary and as a sweetening agent.
- (c) As a food for patients, invalids and children.
- (d) In the form of calcium glucosate as medicine in treatment of calcium deficiency.
- (e) As a reducing agent in silvering of mirrors.
- (f) As a raw material for alcoholic preparations.
- (g) In industrial preparation of vitamin-C.
- (h) In the processing of tobacco.
- (i) As an intravenous injection to the patients with lower glucose content in blood.

(v) **Test of glucose**

- (a) When heated in a dry test tube, it melts, turns brown and finally black, giving a characteristic smell of burnt sugar.
- (b) When warmed with a little conc. H_2SO_4 , it leaves a charred residue of carbon.
- (c) When it is boiled with dilute $NaOH$ solution, it first turns yellow and then brown.
- (d) Molisch's test: This is a general test for carbohydrates. A drop or two of alcoholic solution of α -naphthol is added to 2mL of glucose solution. 1 mL of concentrated H_2SO_4 is added carefully along the sides of the test tube. The formation of a violet ring, at the junction of two liquids confirms the presence of a carbohydrate.
- (e) Silver mirror test: A mixture of glucose and ammonical silver nitrate is warmed in a test tube. Appearance of silver mirror on the inner walls confirms glucose.
- (f) Fehling's solution test: A little glucose is warmed with Fehling's solution. A red precipitate of cuprous oxide is formed.
- (g) Osazone formation: Glucose on heating with excess of phenyl hydrazine in acetic acid gives a yellow crystalline compound, m.pt. $205^\circ C$.

(vi) **Structure of glucose:** The structure of glucose has been established as follows

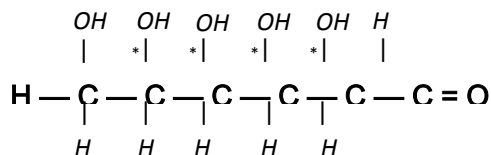
(a) Open chain structure: It is based on the following points:

- Elemental analysis and molecular mass determination show that the molecular formula of glucose is $C_6H_{12}O_6$.
- Glucose on complete reduction with HI and red phosphorus finally gives n-hexane. This indicates that it contains a straight chain of six carbon atoms.
- It reacts with acetic anhydride and forms penta-acetate derivative. This shows the presence of five hydroxyl groups each linked to a separate carbon atom as the molecule is stable.

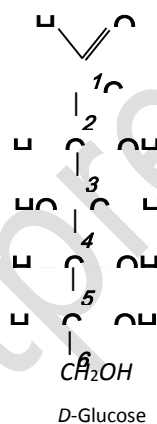


- Glucose combines with hydroxyl amine to form a monoxime. It also combines with one mole of HCN to form a cyanohydrin. These reactions indicate the presence of a carbonyl group, $>C=O$, in the molecule.
- Mild oxidation of glucose with bromine water gives gluconic acid. This shows the presence of an aldehyde group.

On the basis of above observations, the following open chain structure can be written for glucose.



There are four asymmetric carbon atoms marked by asterisks (*) in the molecule. This representation is incomplete, because a definite configuration to these asymmetric centres has not been assigned. The configuration of D-glucose was proved by **Emil Fischer**. The structure of D-glucose as elucidated by Emil Fischer is,



Evidence against open chain structure: The open chain formula of glucose accounts for most of the reactions satisfactorily but fails to explain the following

- Even though an aldehyde group is present, the glucose does not react with $NaHSO_3$ and NH_3 .
- Glucose does not give the Schiff's test for aldehydes.
- Glucose does not react with Grignard reagents.
- Glucose penta-acetate does not react with hydroxyl-amine.
- Two isomeric methyl glucosides (α and β) are obtained by heating glucose with methyl alcohol in presence of dry HCl gas.



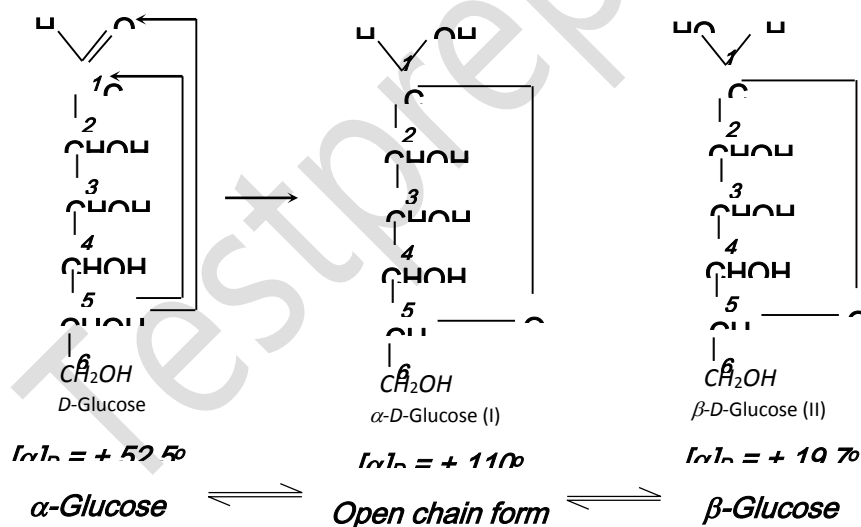
□ Glucose exists in two stereoisomeric forms (α and β). α -glucose with specific rotation $+110^\circ$ is obtained by crystallizing glucose from alcohol or acetic acid solution, whereas β -glucose with specific rotation $+19.7^\circ$ is obtained by crystallizing glucose from pyridine solution.

□ An aqueous solution of glucose shows **mutarotation**, i.e., its specific rotation gradually decreases from $+110^\circ$ to $+52.5^\circ$ in case of α -glucose and increases from $+19.7^\circ$ to $+52.5^\circ$ in case of β -glucose.

All these observations indicate that free aldehydic group is not present in the molecule.

(b) Cyclic structure of glucose: D-glucose exists in two optically active forms known as α -D-glucose and β -D-glucose.

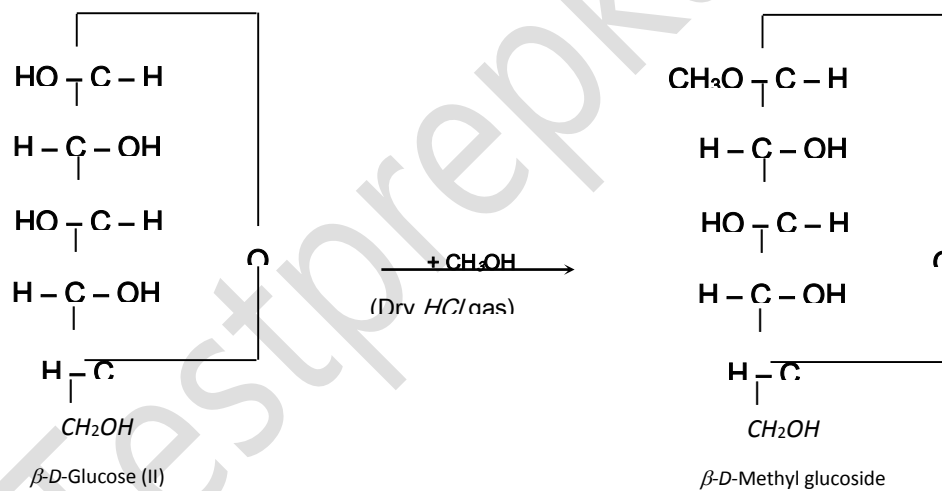
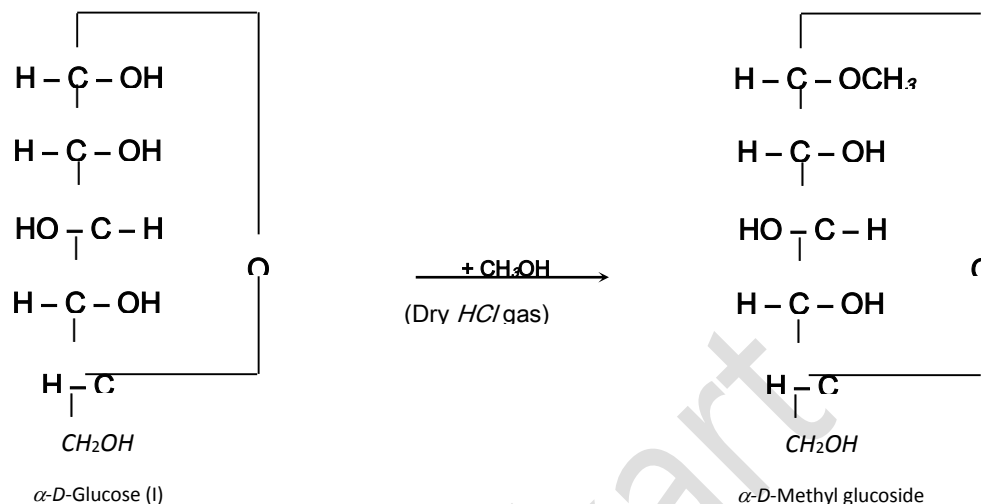
α -D-glucose has specific rotation of $+110^\circ$ and β -D-glucose has specific rotation of $+19.7^\circ$. The two isomers are interconvertible in aqueous solution. The equilibrium rotation is $+52^\circ$. The equilibrium mixture has 36% α -glucose, 64% β -glucose. Glucose forms a stable cyclic hemiacetal (according to Fischer) between $-CHO$ group and the $-OH$ group of the fifth carbon atom in pyranose structure. In this process first carbon atom becomes asymmetric giving two isomers (I) and (II) which differ only in the configuration of the first asymmetric carbon.



Carbon-1 in both configuration (I) and (II) is called an anomeric carbon atom. Due to anomeric carbon, glucose exists in two forms. Both the forms have different physical properties and are called anomers. The ring structure explains all the reactions of glucose. The objections against the open chain structure of glucose have also been satisfactorily explained, e.g.,



- α - and β -glucose on treatment with CH_3OH in presence of dry HCl gas forms α - and β -methyl glucosides respectively.



- **No reaction with NH_3 and $NaHSO_3$:** The glucose ring is not very stable. It is easily broken up by **strong reagents** like HCN , NH_2OH and $C_6H_5NHNH_2$, etc., to give the intermediate aldehyde form, which reacts with them just like an aldehyde.

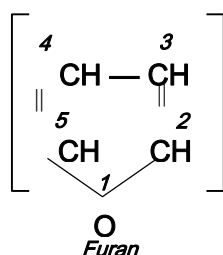
But weak reagents like NH_3 and $NaHSO_3$ are unable to open the chain and cannot react with it. This explains the inability of glucose to form aldehyde ammonia and bisulphite compound.

- **It explains mutarotation :** Ordinary glucose is α -glucose, with a fresh aqueous solution has specific rotation, $[\alpha]_D +110^\circ$. On keeping the solution for some time; α -glucose slowly changes into an equilibrium mixture of α -glucose (36%) and β -glucose (64%) and the mixture has specific rotation $+52.5^\circ$.

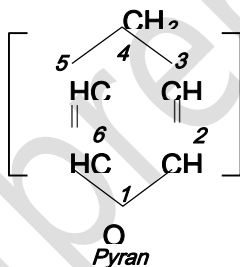


Similarly a fresh aqueous solution of β -glucose having specific rotation, $[\alpha]_D +19.7^\circ$, on keeping (standing) gradually changes into the same equilibrium mixture (having, specific rotation $+52.7^\circ$). So an aqueous solution of glucose shows a physical property, known as **mutarotation**, i.e., a change in the value of specific rotation (muta=change; rotation = specific rotation) is called **mutarotation**.

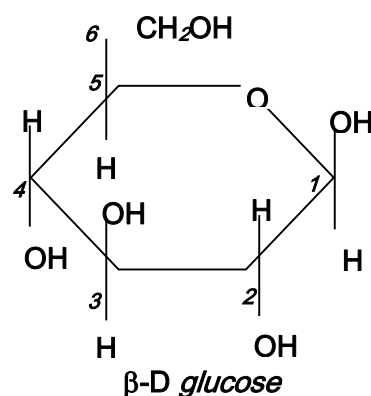
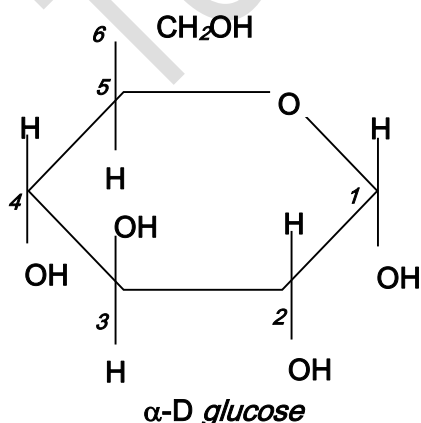
• **Methods for determining the size of rings:** Fischer and Tollen's proposed that the ring or the internal hemiacetal is formed between C^1 and C^4 . It means the ring is **Furan type** or 5-membered ring; this is called **Furanose structure**,



However according to Haworth and Hirst the ring is formed between C^1 and C^5 . It means the ring is **Pyran type** or 6-membered ring, this is called **Pyranose structure**.



The two forms of D-glucose are also shown by **Haworth** projection formula which are given below;

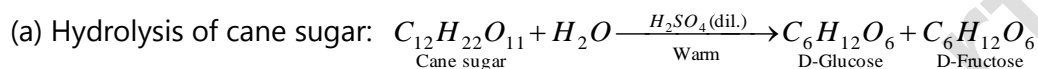


The above projection formulae show that the six membered ring is planar but actually the ring has a chain structure similar to cyclohexane.

In Haworth formula all the OH groups on the right in Fischer's formula are directed below the plane of the ring while those on the left go above the plane. The terminal CH_2OH projects above the plane of the ring.

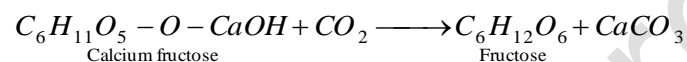
(4) **Fructose, fruit sugar $C_6H_{12}O_6$, Ketohehexose:** It is present in abundance in fruits and hence is called **fruit sugar**. It is also present in cane sugar and honey alongwith glucose in combined form. The polysaccharide **inulin** is a polymer of fructose and gives only fructose on hydrolysis. Since naturally occurring fructose is laevorotatory, it is also known as **laevulose**.

(i) **Preparation**



The solution having equal molecules of D-glucose and D-fructose is termed **invert sugar** and the process is known as **inversion**.

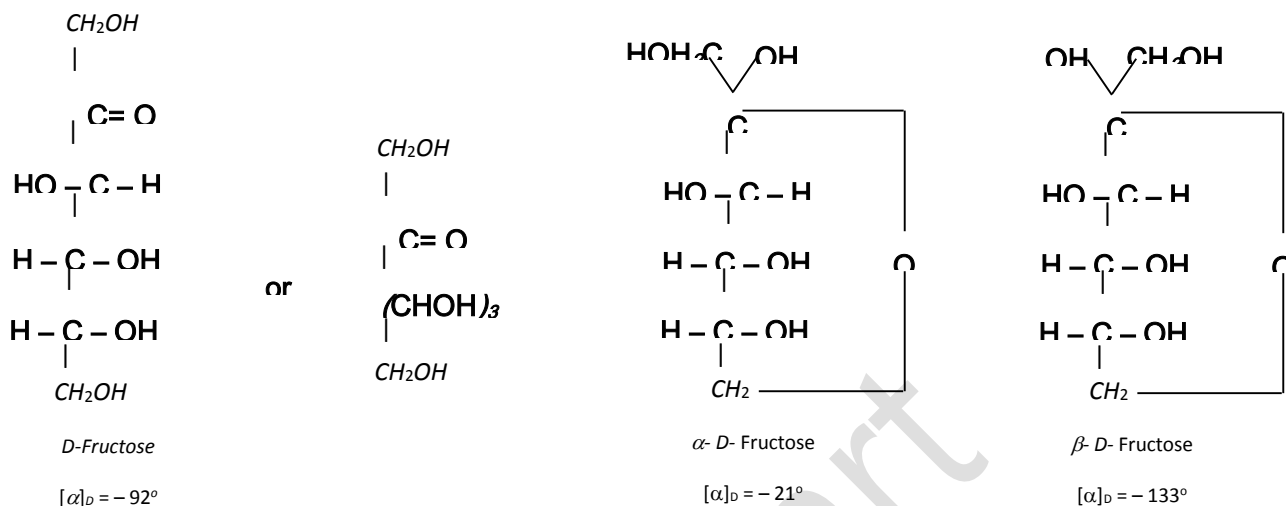
Note: The excess of sulphuric acid is neutralized by adding milk of lime. A little more of lime is added which converts both glucose and fructose into calcium glucosate and calcium fructose respectively.



(ii) **Properties:** The anhydrous fructose is a colorless crystalline compounds. It melts at $102^\circ C$. It is soluble in water but insoluble in benzene and ether. It is less soluble in water than glucose. It is the sweetest* of all sugars and its solution is laevorotatory. Like glucose, it also shows **mutarotation**.



Fructose is a pentahydroxy ketone and its open-chain and closed-chain structures can be represented as:



(5) Comparison between glucose and fructose

Property	Glucose	Fructose
Molecular formula	C ₆ H ₁₂ O ₆	C ₆ H ₁₂ O ₆
Nature	Polyhydroxy aldehyde.	Polyhydroxy ketone
Melting point	146°C	102°C
Optical activity of natural form	Dextrorotatory	Laevorotatory
With ethyl alcohol	Almost insoluble	More soluble
Oxidation (a) With bromine water (b) With nitric acid	Gluconic acid Saccharic acid (Glucaric acid)	No reaction Mixture of glycollic acid, tartaric acid and trihydroxy glutaric acid
Reduction	Sorbitol	Mixture of sorbitol and mannitol
Calcium hydroxide	Forms calcium glucosate, soluble in water	Forms calcium fructosate, insoluble in water
Molisch's reagent	Forms a violet ring	Forms a violet ring
Fehling's solution	Gives red precipitate	Gives red precipitate
Tollen's reagent	Forms silver mirror	Forms silver mirror
Phenyl hydrazine	Forms osazone	Forms osazone
Resorcinol + HCl (dil.) (Selivanoff's test)	No colouration	Gives red or brown colour or precipitate
Freshly prepared ammonium molybdate sol. + few drops of acetic acid (Pinoff's test).	Light blue colour	Bluish green colour on heating
Alcoholic α-naphthol + HCl (conc.) (Furfural test)	No colouration	A purple colour (violet) on boiling

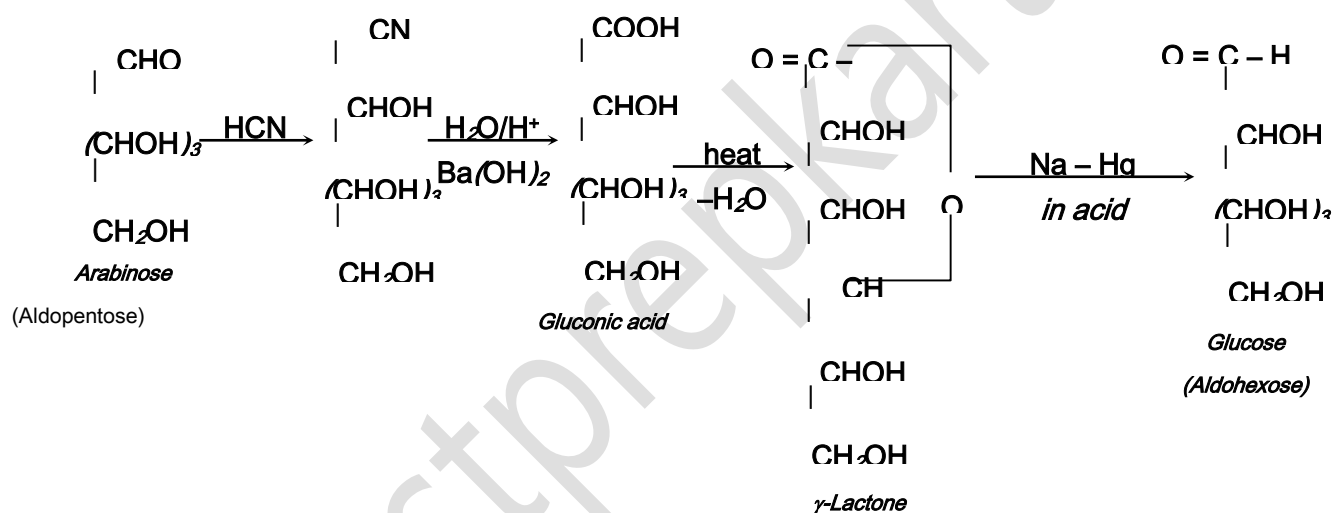


Note: Fructose gives reactions similar to glucose. The difference in properties is due to the fact that it contains a ketonic group while glucose contains an aldehydic group.

(6) **Interconversions**

(i) **Chain Lengthening of Aldoses (Killiani-Fischer synthesis):** The conversion of an aldose to the next higher member involves the following steps:

- (a) Formation of a cyanohydrin.
- (b) Hydrolysis of – CN to – COOH forming aldonic acid.
- (c) Conversion of aldonic acid into lactone by heating.
- (d) The lactone is finally reduced with sodium amalgam or sodium borohydride to give the higher aldose.

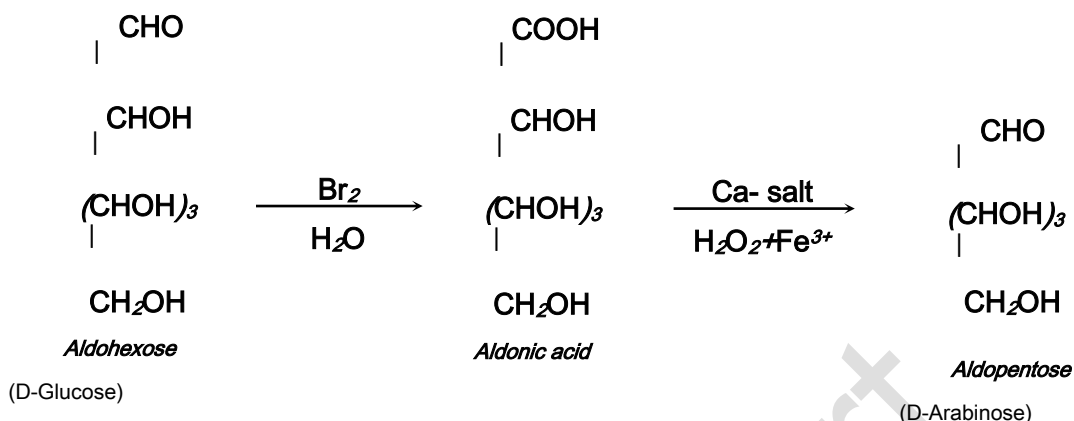


(ii) **Chain Shortening of Aldoses (Ruff Degradation)**

- (a) An aldose can be converted to the next lower member by **Ruff Degradation**. It involves two steps:
 - Oxidation of the aldose to aldonic acid by using bromine water.

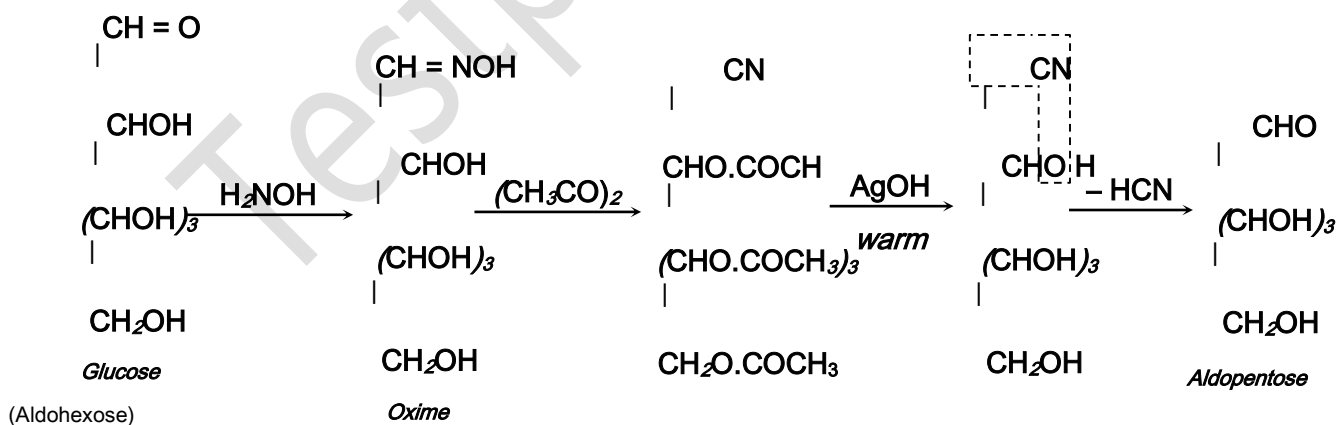


- The aldonic acid is treated with CaCO_3 to give the calcium salt which is then oxidised by Fenton's reagent ($\text{H}_2\text{O}_2 + \text{ferric sulphate}$) to form the next lower aldose.



(b) By Wohl's method: It involves the following steps

- Formation of oxime with hydroxyl amine.
- Heating of oxime with acetic anhydride undergoes dehydration into cyano compound, whereas the hydroxyl groups get acetylated.
- The acetyl derivative is warmed with ammoniacal silver nitrate which removes the acetyl group by hydrolysis and eliminates a molecule of HCN.

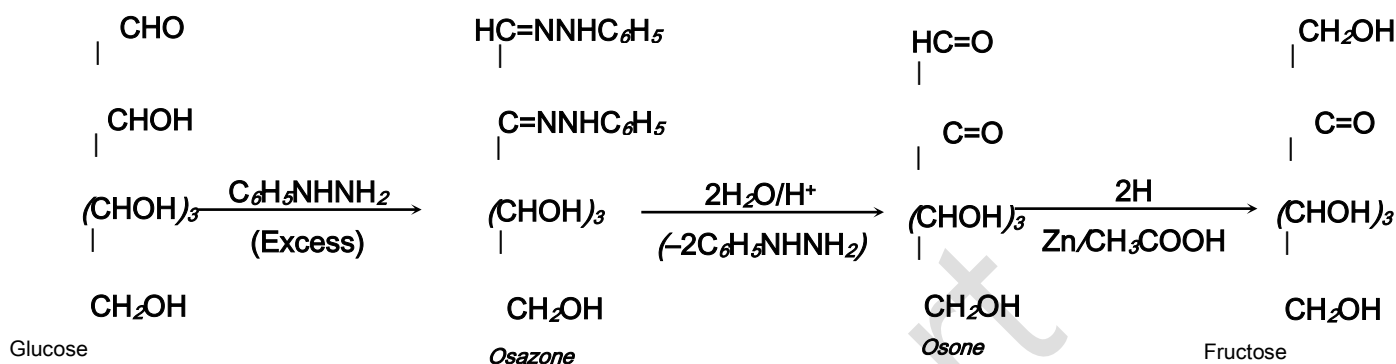


(iii) **Conversion of an Aldose to the isomeric Ketose:** Three steps are involved:

(a) Treatment of aldose with excess of phenyl hydrazine to form osazone.

(b) Hydrolysis of osazone with dil. HCl to form osone.

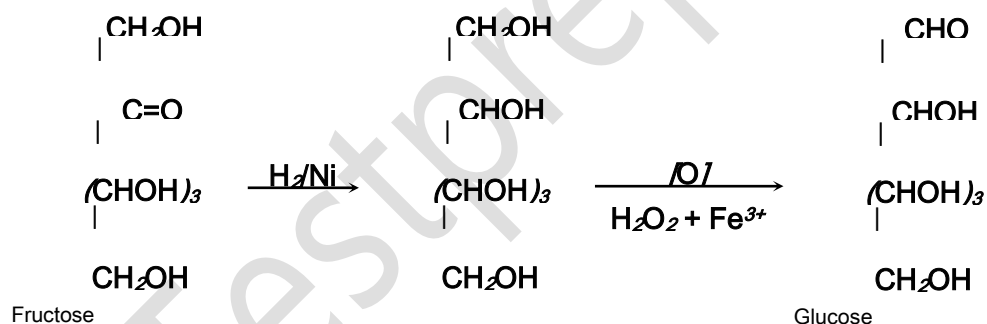
(c) Reduction of osone with zinc and acetic acid to form ketose.



(iv) **Conversion of a Ketose to the isomeric Aldose:** Two steps are involved,

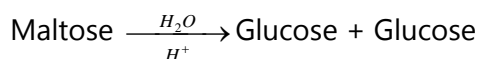
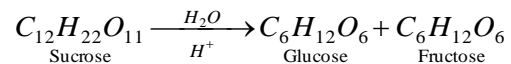
(a) Reduction of a ketose with H_2 / Ni to form polyhydric alcohol.

(b) Oxidation with Fenton's reagent to form aldose.



3. Disaccharides.

The disaccharides yield on hydrolysis two monosaccharides. Those disaccharides which yield two hexoses on hydrolysis have a general formula $C_{12}H_{22}O_{11}$. The hexoses obtained on hydrolysis may be same or different.



The hydrolysis is done by dilute acids or enzymes. The enzymes which bring hydrolysis of sucrose, lactose and maltose are invertase, lactase and maltase, respectively. Out of the three disaccharides, sucrose (cane-sugar) is the most important as it is an essential constituent of our diet.

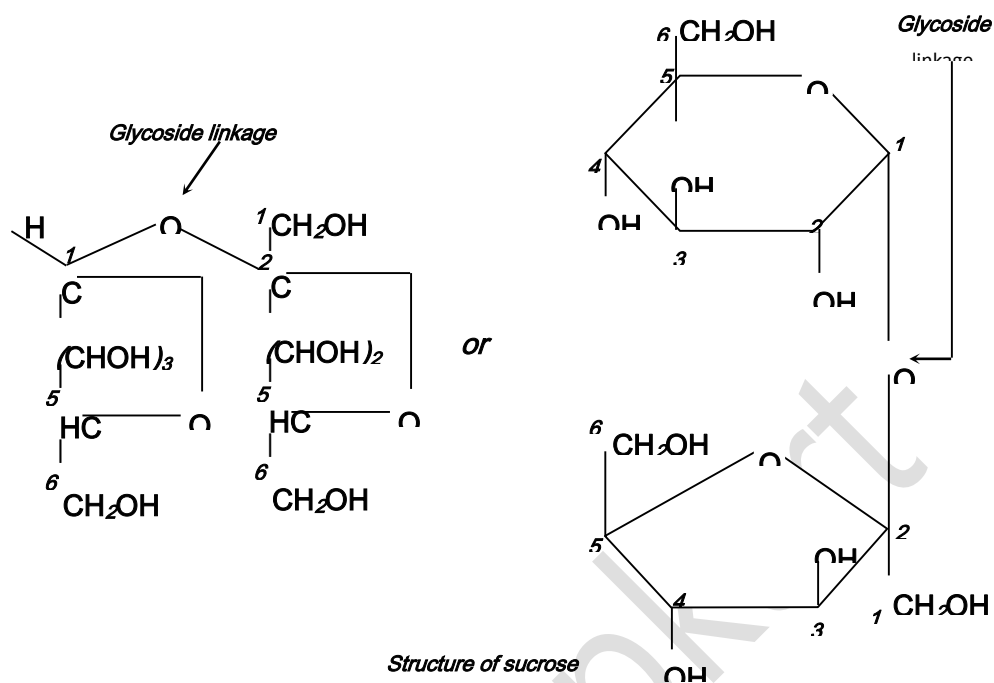
In disaccharides, the two monosaccharides are joined together by glycoside linkage. A glycoside bond is formed when hydroxy group of the hemiacetal carbon of one monosaccharide condenses with a hydroxy group of another monosaccharide giving – O– bond.

(1) **Sucrose; Cane-sugar [$C_{12}H_{22}O_{11}$]:** It is our common table sugar. It is obtained from sugar cane and sugarbeets. It is actually found in all photosynthetic plants.

(i) **Properties:** It is a colorless, odorless, crystalline compound. It melts at 185 – 186°C. It is very soluble in water, slightly soluble in alcohol and insoluble in ether. It is dextrorotatory but does not show **mutarotation**. It is a non-reducing sugar as it does not reduce Tollen's or Fehling's reagent. Sucrose, on heating slowly and carefully, melts and then if allowed to cool, it solidifies to pale yellow glassy mass called 'Barley sugar'. When heated to 200°C, it loses water to form brown amorphous mass called Caramel. On strong heating, it chars to almost pure carbon giving smell of burnt sugar.



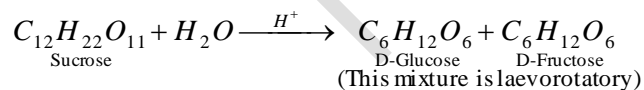
It is composed of α -D-glucopyranose unit and a β -D-fructofuranose unit. These units are joined by α - β -glycosidic linkage between C -1 of the glucose unit and C -2 of the fructose unit.



(ii) **Uses**

- (a) As a sweetening agent for various food preparations, jams, syrups sweets, etc.
- (b) In the manufacture of sucrose octa-acetate required to denature alcohol, to make paper transparent and to make anhydrous adhesives.

(2) **Inversion of cane-sugar:** The hydrolysis of sucrose by boiling with a mineral acid or by enzyme invertase, produces a mixture of equal molecules of D-glucose and D-fructose.



Sucrose solution is dextrorotatory. Its specific rotation is $+66.5^\circ$. But on hydrolysis, it becomes laevorotatory. The specific rotation of D-glucose is $+52^\circ$ and of D-fructose is -92° .

Therefore, the net specific rotation of an equimolar mixture of D-glucose and D-fructose is.

$$\frac{+52^\circ - 92^\circ}{2} = -20^\circ$$



Thus, in the process of hydrolysis of sucrose, the specific rotation changes from $+66.5^\circ$ to -20° , i.e., from dextro it becomes laevo and it is said that inversion has taken place. The process of hydrolysis of sucrose is thus termed as **inversion of sugar** and the hydrolysed mixture having equal molar quantities of D-glucose and D-fructose is called **invert sugar**. The enzyme that brings the inversion is named as **invertase**.

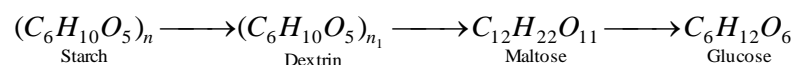
(3) Distinction between glucose and sucrose

Test	Glucose	Sucrose
With conc. H_2SO_4 in cold	No effect	Charring occurs and turns black
Molisch's reagent	Violet ring is formed	Violet ring is formed
With NaOH	Turns yellow	No effect
With Tollen's Solution	Gives silver mirror	No effect
With Fehling's solution	Gives red precipitate of Cu_2O	No effect
On heating with phenyl hydrazine	Gives yellow precipitate of glucosazone	No effect, i.e., does not form osazone
Aqueous solution + resorcinol + HCl (conc.)	No effect	Reddish-brown precipitate which dissolves in ethanol.

4. Polysaccharide: [Starch and cellulose].

Polysaccharides are polymers of monosaccharides. The most important polysaccharides are starch and cellulose. They have a general formula $(C_6H_{10}O_5)_n$. Starch (Amylum) is most widely distributed in vegetable kingdom. It is found in the leaves, stems, fruits, roots and seeds. Concentrated form of starch is present in wheat, corn, barley, rice, potatoes, nuts, etc. It is the most important food source of carbohydrates.

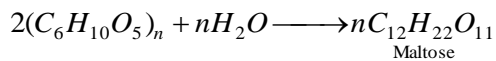
(1) **Starch and its derivatives:** Starch is a white amorphous substance with no taste or smell. When heated to a temperature between $200 - 250^\circ C$, it changes into dextrin. At higher temperature charring occurs. When boiled with dilute acid, starch ultimately yields glucose.



Both n and n_1 , are unknown, but n is believed to be greater than n_1 .

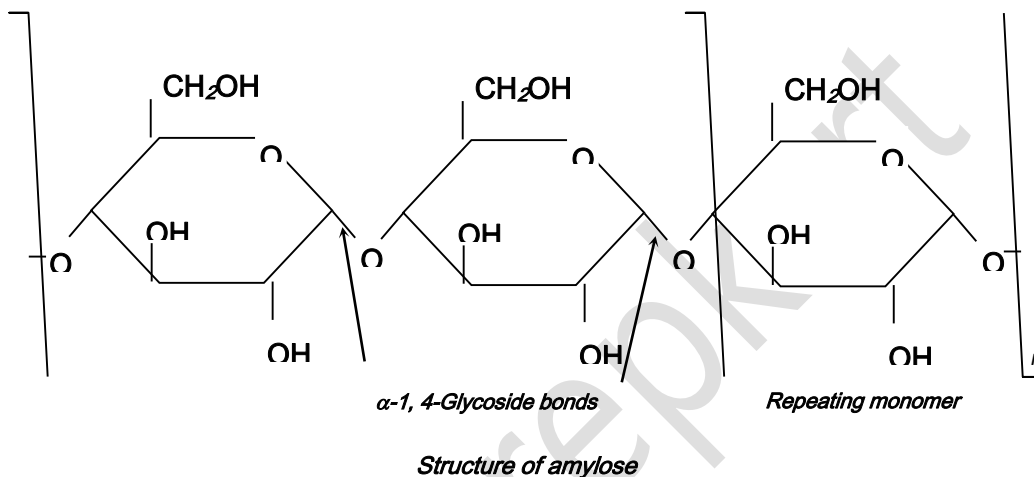
When treated with enzyme, diastase, it yields maltose.





Starch solution gives a blue colour with a drop of iodine which disappears on heating to $75 - 80^\circ C$ and reappears on cooling. The exact chemical nature of starch varies from source to source. Even the starch obtained from same source consists of two fractions (i) amylose and (ii) amylopectin.

Amylose is a linear polymer while amylopectin is a highly branched polymer. Both are composed of α -D-glucose units linked by glycosidic linkages. The number of D-glucose units in amylose range from 60 – 300. It is soluble in hot water, Amylopectin consists of D-glucose units from 300 – 600. It is insoluble in water.



Uses: Starch and its derivatives are used

- (i) As the most valuable constituent of food as rice, bread, potato and corn-flour, etc.
- (ii) In the manufacture of glucose, dextrin and adhesives (starch paste).
- (iii) In paper and textile industry.
- (iv) In calico printing as a thickening agent for colors.
- (v) Nitro starch is used as an explosive.
- (vi) Starch-acetate is a transparent gelatin like mass and is used mainly for making sweets.

Distinction between glucose, sucrose, starch



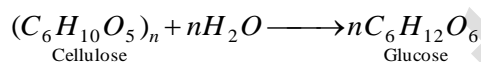
Test	Glucose	Sucrose	Starch
With iodine solution	No effect	No effect	Blue colour
With Fehling's solution	Gives red precipitate	No effect	No effect
With Tollen's reagent	Gives silver mirror	No effect	No effect
With phenyl hydrazine	Forms yellow osazone	No effect	No effect
Solubility in water	Soluble	Soluble	Insoluble
Taste	Sweet	Sweet	No taste

(2) **Cellulose and its uses:** It is found in all plants and so is the most abundant of all carbohydrates. It is the material used to form cell walls and other structural features of the plants. Wood is about 50% cellulose and the rest is lignin. Cotton and paper are largely composed of cellulose.

Pure cellulose is obtained by successively treating cotton, wool, flax or paper with dilute alkali, dilute HCl or HF . This treatment removes mineral matter, water, alcohol and ether. Cellulose is left behind as a white amorphous powder.

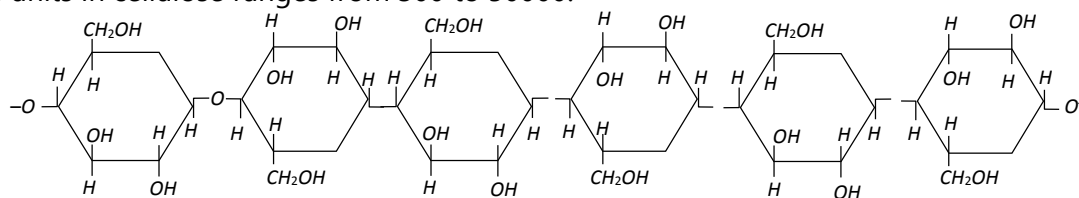
Cellulose is insoluble in water and in most of the organic solvents. It decomposes on heating but does not melt. It dissolves in ammonical copper hydroxide solution (Schwitzer's reagent). Cellulose also dissolves in a solution of zinc chloride in hydrochloric acid.

When it is treated with concentrated H_2SO_4 in cold, it slowly passes into solution. The solution when diluted with water, a starch like substance amyloid is precipitated and is called **parchment paper**. When boiled with dilute H_2SO_4 , it is completely hydrolysed into D-glucose.



The cattle, goats and other ruminants can feed directly cellulose (grass, straw, etc.) as they have digestive enzymes (cellulases) capable of hydrolysing cellulose into glucose. Man and many other mammals lack the necessary enzymes in their digestive tract and thus cannot use cellulose as food stuff.

Cellulose is a straight chain polysaccharide composed of D-glucose units which are joined by B-glycosidic linkages between C-1 of one glucose unit and C-4 of the next glucose unit. The number of D-glucose units in cellulose ranges from 300 to 50000.



Structure of cellulose

Uses: Cellulose is used



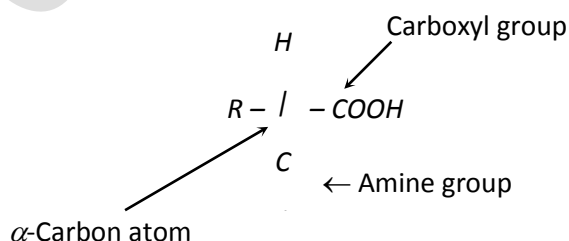
- (i) As such in the manufacture of cloth (cotton), canvas and gunny bags (jute) and paper (wood, bamboo, straw, etc.)
- (ii) In the form of cellulose nitrates for the manufacture of explosives (gun-powder), medicines, paints and lacquers. The cellulose nitrates with camphor yield celluloid which is used in the manufacture of toys, decorative articles and photographic films.
- (iii) In the form of cellulose acetate for the manufacture of rayon (artificial silk) and plastics.

5. Proteins and amino acids.

Proteins: Proteins are a class of biologically important compounds. They are crucial to virtually all processes in living systems. Some of them are hormones which serve as chemical messengers that coordinate certain biochemical activities. Insulin, for example, controls the level of sugar in the blood stream. Some proteins serve to transport the substances through the organism. Haemoglobin, for instance, carries oxygen in blood stream and delivers to different parts of the body. α -keratin, serves as a major constituent of hairs, nails and skin, while collagen is the prime constituent of tendons. Proteins are also found in toxins (poisonous materials) as well as in antibiotics.

Amino acids: An amino acid is a bifunctional organic molecule that contains both a carboxyl group, $-COOH$, as well as an amine group, $-NH_2$. They are classified as acidic basic or neutral according to number of amine and carboxyl groups in a molecule. Neutral amino acids contain only one amine and one carboxyl group. They are further classified according to the position of amine group in relation to carboxyl group into α -, β -, γ - and δ -amino acids. Out of these α -amino acids are most important as they are building blocks of bio-proteins.

In an α -amino acid, the amine group is located on the carbon atom adjacent to the carboxyl group (the α -carbon atom). The general structure of the α -amino acids is represented as



R may be alkyl, aryl or any other group.

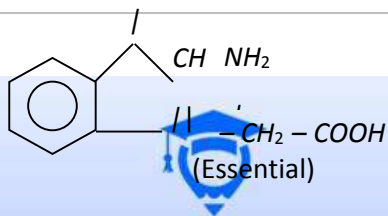
The proteins differ in the nature of R-group bonded to α -carbon atom. The nature of R-group determines the properties of proteins. There are about 20 amino acids which make up the bio-proteins. Out of these 10 amino acids (non-essential) are synthesised by our bodies and rest are essential in the



diet (essential amino acids) and supplied to our bodies by food which we take because they cannot be synthesised in the body. The α -amino acids are classified into the following four types.

Amino acids with non-polar side chain: Examples are:

Name	Structure	Three letter symbol	One letter code
Glycine	$\begin{array}{c} \text{NH}_2 \\ \\ \text{CH}_2 \\ \\ \text{COOH} \end{array}$	Gly	G
Alanine	$\begin{array}{c} \text{NH}_2 \\ \\ \text{CH}_3\text{CH} \\ \\ \text{COOH} \end{array}$	Ala	A
Valine	$\begin{array}{c} \text{NH}_2 \\ \\ (\text{CH}_3)_2\text{CH}-\text{CH} \\ \\ \text{COOH} \\ \text{(Essential)} \end{array}$	Val	V
Leucine	$\begin{array}{c} \text{NH}_2 \\ \\ (\text{CH}_3)_2\text{CH}-\text{CH}_2\text{CH} \\ \\ \text{COOH} \\ \text{(Essential)} \end{array}$	Leu	L
Isoleucine	$\begin{array}{c} \text{NH}_2 \\ \\ \text{C}_2\text{H}_5-\text{CH}-\text{CH} \\ \quad \\ \text{H} \quad \text{CH}_3 \\ \text{(Essential)} \end{array}$	ILE	I
Phenyl alanine	$\begin{array}{c} \text{NH}_2 \\ \\ \text{C}_6\text{H}_5\text{CH}_2\text{CH} \\ \\ \text{COOH} \\ \text{(Essential)} \end{array}$	PHE	F
Proline	$\begin{array}{c} \text{H}_2\text{C} \quad \text{CH}_2 \\ \quad \\ \text{H}_2\text{C} \quad \text{CHCOOH} \\ \quad \\ \text{N} \\ \\ \text{H} \end{array}$	Pro	P
Amino acids with polar but neutral side chain: Examples are			
Name	Structure	Three letter symbol	One letter code
Tryptophan	$\begin{array}{c} \text{H} \\ \\ \text{CH} \quad \text{NH}_2 \\ \\ \text{C}_6\text{H}_5 \\ \\ \text{CH}_2-\text{COOH} \\ \text{(Essential)} \end{array}$	Trp.	W

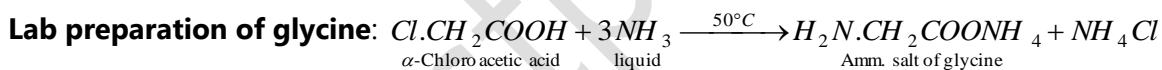
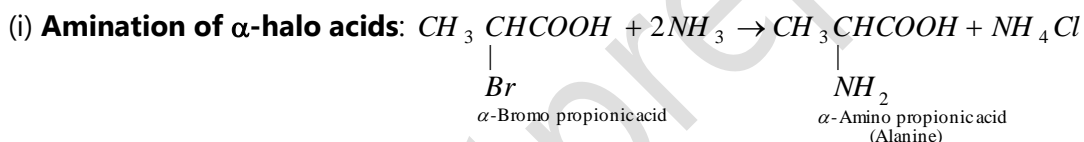


Serine	$\text{HO-CH}_2\text{-CH} \begin{cases} \text{NH}_2 \\ \text{COOH} \end{cases}$	Ser	S
Threonine	$\text{CH}_3\text{CHOH-CH} \begin{cases} \text{NH}_2 \\ \text{COOH} \end{cases}$ (Essential)	Thr	T
Tyrosine	$\text{HO} \begin{array}{c} \text{C}_6\text{H}_4 \\ \text{CH}_2\text{-CH-COOH} \end{array} \begin{cases} \text{NH}_2 \\ \end{cases}$	Tyr	Y
Cysteine	$\text{HS-CH}_2\text{-CH} \begin{cases} \text{NH}_2 \\ \text{COOH} \end{cases}$	Cys	C
Methionine	$\text{CH}_3\text{-S-CH}_2\text{-CH}_2\text{-CH} \begin{cases} \text{NH}_2 \\ \text{COOH} \end{cases}$ (Essential)	Met	M
Asparagine	$\text{H}_2\text{N} \begin{array}{c} \text{O} \\ \parallel \\ \text{C} \end{array} \text{-CH}_2\text{-CH} \begin{cases} \text{NH}_2 \\ \text{COOH} \end{cases}$	Asn	N
Glutamine	$\text{H}_2\text{N} \begin{array}{c} \text{O} \\ \parallel \\ \text{C} \end{array} \text{-CH}_2\text{-CH}_2\text{-CH} \begin{cases} \text{NH}_2 \\ \text{COOH} \end{cases}$	Gln	Q
Amino acids with acidic side chains : Examples are			
Aspartic acid	$\text{HOOC-CH}_2\text{-CH} \begin{cases} \text{NH}_2 \\ \text{COOH} \end{cases}$	Asp	D
Glutamic acid	$\text{HOOC-CH}_2\text{-CH}_2\text{CH} \begin{cases} \text{NH}_2 \\ \text{COOH} \end{cases}$	Glu	E

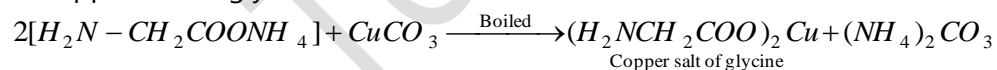


Amino acids with basic side chains : Examples are			
Lysine	$\begin{array}{c} \text{H}_2\text{N}(\text{CH}_2)_4\text{CH} \\ \text{(Essential)} \end{array} \begin{array}{l} \diagup \text{NH}_2 \\ \diagdown \text{COOH} \end{array}$	Lys	K
Arginine	$\begin{array}{c} \text{NH} \\ \diagdown \\ \text{H}_2\text{N} \end{array} \text{C} \cdot \text{NH} \cdot (\text{CH}_2)_3\text{CH} \begin{array}{l} \diagup \text{NH}_2 \\ \diagdown \text{COOH} \end{array}$ (Essential)	Arg	R
Histidine	$\begin{array}{c} \text{HC} \\ \diagdown \\ \text{N} \\ \diagup \\ \text{CH} \\ \diagdown \\ \text{NH} \end{array} \text{C} - \text{CH}_2 - \text{CH} \begin{array}{l} \diagup \text{NH}_2 \\ \diagdown \text{COOH} \end{array}$ (Essential)	His	H

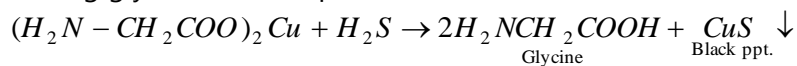
(1) Methods of preparation of α -amino acids



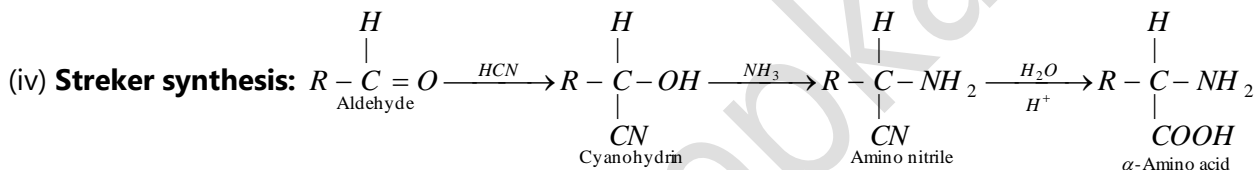
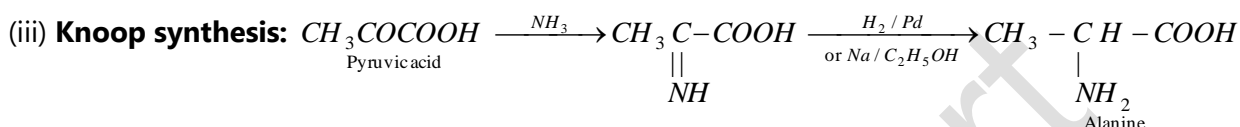
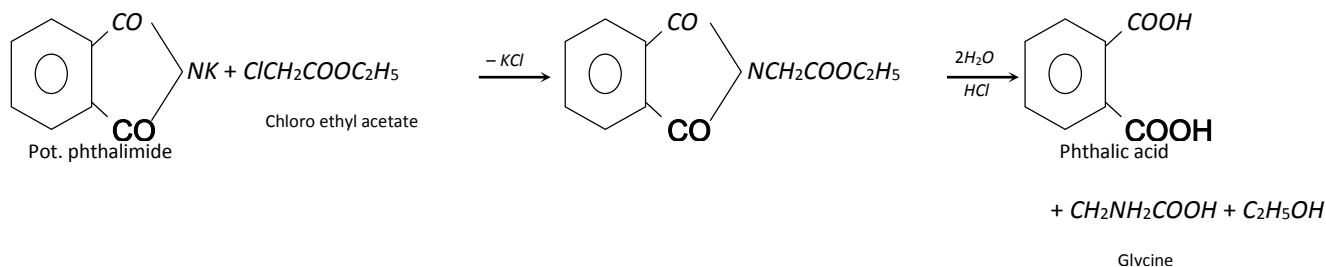
The ammonium salt so obtained is boiled with copper carbonate and cooled when blue colour needles of copper salt of glycine are obtained.



It is now dissolved in water and H_2S is passed till whole of the copper precipitates as copper sulphide leaving glycine as the aqueous solution.



(ii) **Gabriel phtalimide synthesis**



(v) **From natural protein:** Natural proteins are hydrolysed with dil. HCl or H_2SO_4 at $250^\circ C$ in an autoclave when a mixture of α -amino acids is obtained. This mixture is esterified and the various esters are separated by fractional distillation. The esters are then hydrolysed into respective α -amino acids.

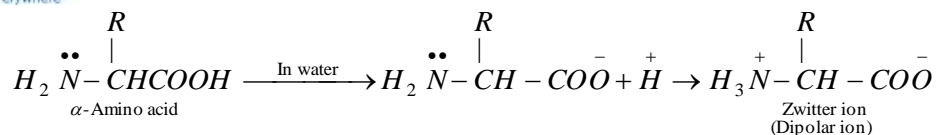
(2) **Physical properties**

(i) Amino acids are colourless, crystalline substances having sweet taste. They melt with decomposition at higher temperature (more than $200^\circ C$). They are soluble in water but insoluble in organic solvents.

(ii) Except glycine, all the α -amino acids are optically active and have an asymmetric carbon atom (α -carbon atom). Hence, each of these amino acids can exist in two optical isomers. In proteins, however, only one isomer of each is commonly involved.

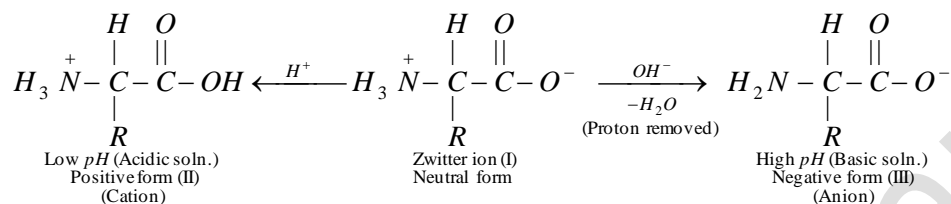
(iii) **Zwitter ion and isoelectric point:** Since the $-NH_2$ group is basic and $-COOH$ group is acidic, in neutral solution it exists in an internal ionic form called a **Zwitter ion** where the proton of $-COOH$ group is transferred to the $-NH_2$ group to form **inner salt**, also known as **dipolar ion**.





The Zwitter ion is dipolar, charged but overall electrically neutral and contains both a positive and negative charge.

(3) **Chemical properties:** Amino acids are amphoteric in nature. Depending on the pH of the solution, the amino acid can donate or accept proton.



When an ionized form of amino acid is placed in an electric field, it will migrate towards the opposite electrode. Depending on the pH of the medium, following three things may happen

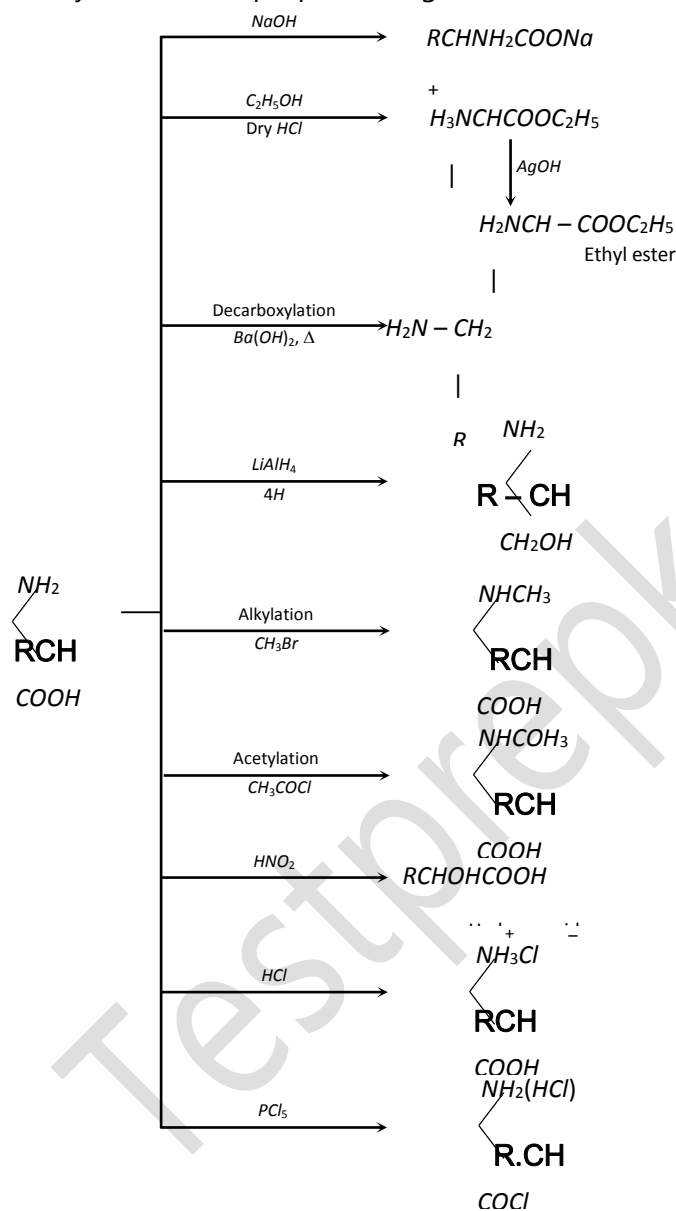
- In acidic solution (low pH), the positive ion moves towards cathode.
- In basic solution (high pH), the negative ion moves towards anode.
- The Zwitter ion does not move towards any of the electrodes.

The intermediate pH at which the amino acid shows no tendency to migrate towards any of the electrodes and exists the equilibrium when placed in an electric field is known as isoelectric point.

This is characteristic of a given amino acid and depends on the nature of R-linked to α -carbon atom.



(i) α -amino acids show the reactions of $-\text{NH}_2$ group, $-\text{COOH}$ groups and in which both the groups are involved. A summary of chemical properties is given below:



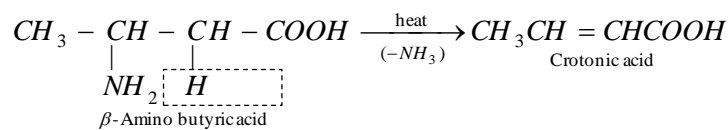
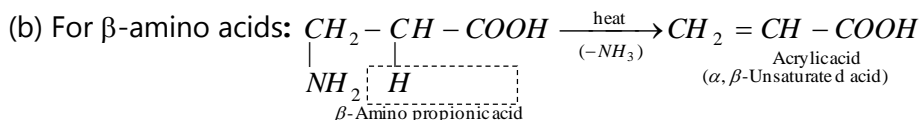
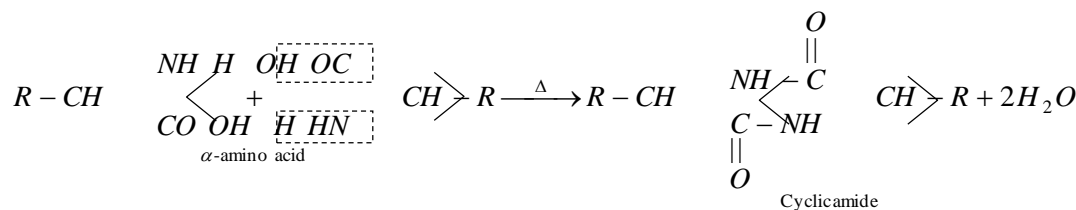
Note: Proline is the only natural α -amino acid which is a secondary amine.

□ Only achiral α -amino acid found in protein is glycine.

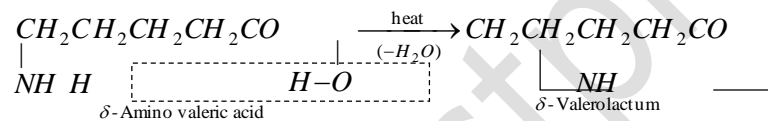
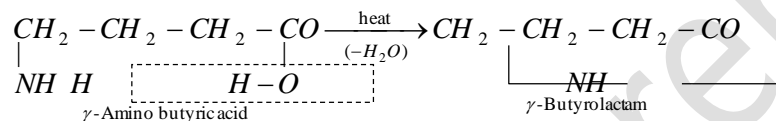


(ii) **Action of heat**

(a) For α -amino acids

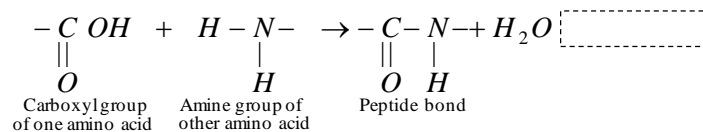


(c) For γ and δ amino acids



These lactams have stable five or six membered rings.

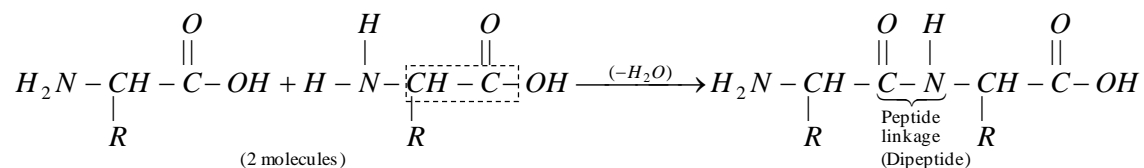
(iii) **Formation of proteins-peptide bond:** Proteins are formed by joining the carboxyl group of one amino acid to the α -amino group of another amino acid. **The bond formed between two amino acids by the elimination of a water molecule is called a peptide linkage or bond.** The peptide bond is simply another name for amide bond.



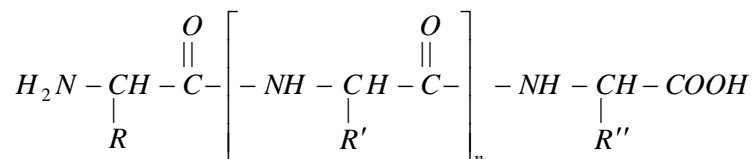
The product formed by linking amino acid molecules through peptide linkages, $-CO-NH-$, is called a **peptide**. Peptides are further designated as di, tri, tetra or penta peptides accordingly as they contain



two, three, four or five amino acid molecules, same or different, joined together in the following fashions.

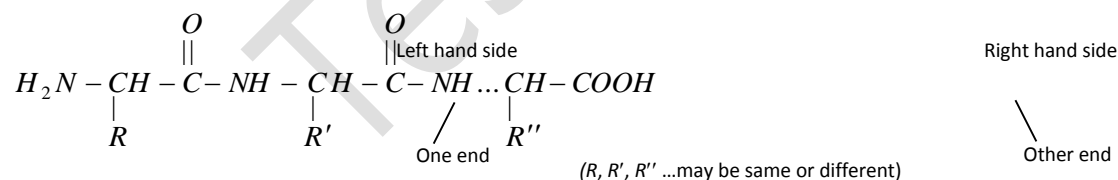


When the number of amino molecules is large, the product is termed polypeptide which may be represented as,



(4) **Composition:** Composition of a protein varies with source. An approximate composition is as follows: Carbon 50-53%; hydrogen 6-7%; oxygen 23-25%; nitrogen 16-17%; Sulphur about 1%. Other elements may also be present, e.g., phosphorus (in nucleoproteins), iodine (in thyroid proteins) and iron (in haemoglobin).

(5) **Structure of proteins:** The structure of proteins is very complex. **The primary structure of a protein refers to the number and sequence of the amino acids in its polypeptide chains** (discussed in the formation of proteins). The primary structure is represented beginning with the amino acid whose amino group is free (the N-terminal end) and it forms the one end of the chain. Free carboxyl group (C-terminal end) forms the other end of the chain.



Side chains may have basic groups or acidic groups as $-NH_2$ in lysine and $-COOH$ in aspartic acid. Because of these acidic and basic side chains, there are positively and negatively charged centres. Though the peptide linkage is stable, the reactivity is due to these charged centres in the side chains. Primary structure tells us nothing about the shape or conformation of the molecule. Most of the bonds in protein molecules being single bonds can assume infinite number of shapes due to free rotation about single bonds. However, it has been confirmed that each protein has only a single three dimensional conformation.



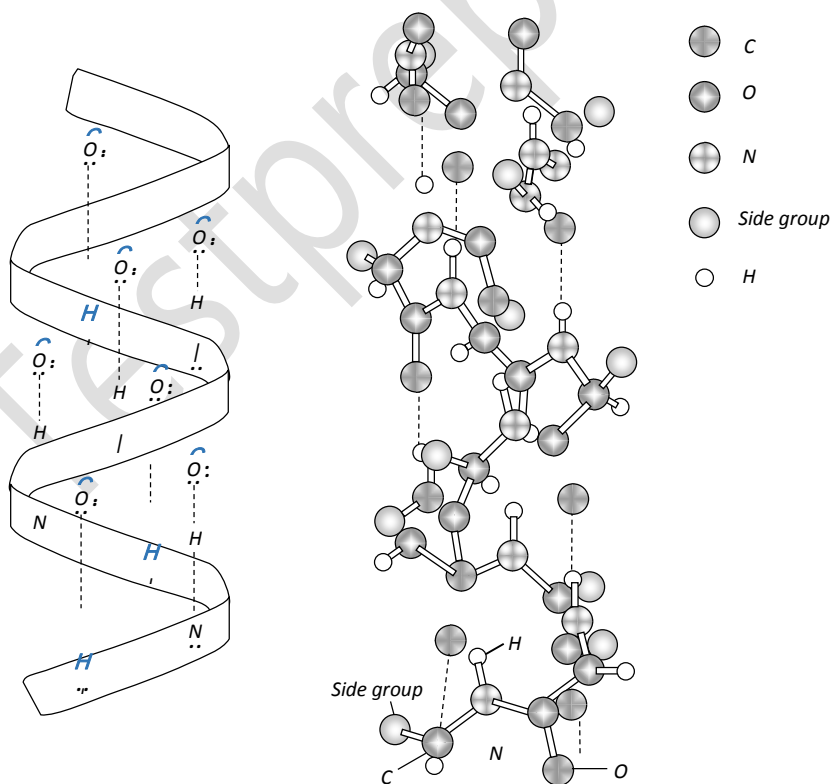
The fixed configuration of a polypeptide skeleton is referred to as the secondary structure of a protein. It gives information:

- About the manner in which the protein chain is folded and bent;
- About the nature of the bonds which stabilise this structure.

Secondary structure of protein is mainly of two types

(i) **α -helix** : This structure is formed when the chain of α -amino acids coils as a right handed screw (called α -helix) because of the formation of **hydrogen bonds** between amide groups of the same peptide chain, i.e., NH group in one unit is linked to carbonyl oxygen of the third unit by hydrogen bonding. This hydrogen bonding between different units is responsible for holding helix in a position. The side chains of these units project outward from the coiled backbone.

Such proteins are elastic, i.e., they can be stretched. On stretching weak hydrogen bonds break up and the peptide chain acts like a spring. The hydrogen bonds are reformed on releasing the tension. Wool and hair have α -helix structure.

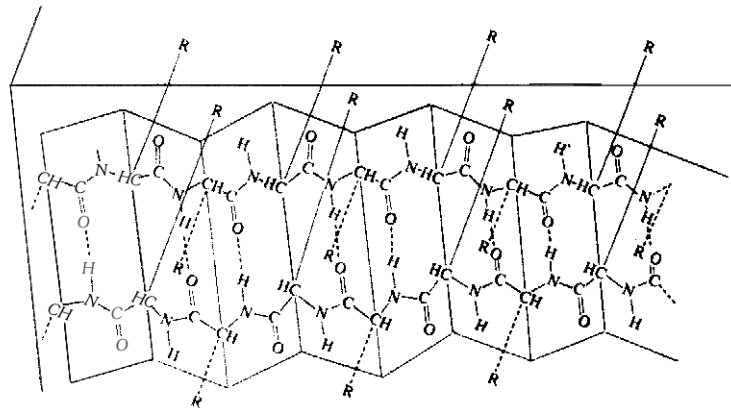


(a) Representation of a polypeptide chain in an α -helical configuration.

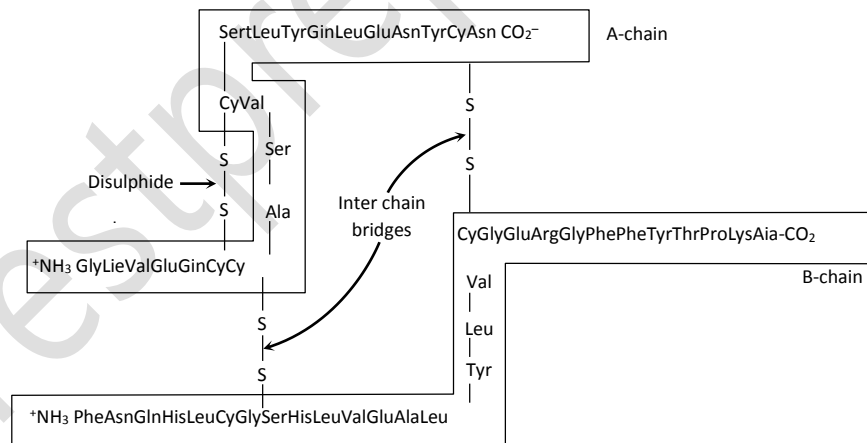
(b) Stabilization of an α -helical configuration by hydrogen bonding. The shaded spheres represent carbon-atoms or residues (R) of amino acids.



(ii) **β -pleated sheet:** A different type of secondary structure is possible when polypeptide chains are arranged side by side. The chains are held together by a very large number of hydrogen bonds between C = O and NH of different chains. Thus, the chains are bonded together forming a sheet. These sheets can slide over each other to form a three dimensional structure called a beta pleated sheet. Silk has a beta pleated structure.

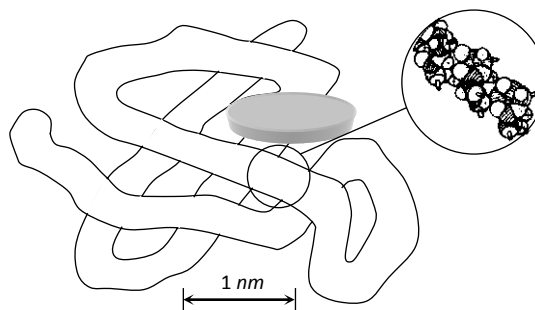


The beta pleated sheet structure of proteins



Tertiary Structure of proteins

Further folding and bending of secondary structure is called the tertiary structure of proteins



Tertiary structure of proteins



Globular proteins possess tertiary structure. In general globular proteins are very tightly folded into a compact spherical form.

(6) **Classification of proteins:** According to chemical composition, proteins are divided into two classes

(i) **Simple proteins:** Simple proteins are composed of chains of amino acid units only joined by peptide linkages. These proteins on hydrolysis yield only mixture of amino acids. Examples are

Egg albumin, serum globulins, glutelin in wheat, coryzenin in rice, tissue globulin, etc.

(ii) **Conjugated proteins:** The molecules of conjugated proteins are composed of simple proteins and non-protein material. The non-protein material is called **prosthetic group** or **cofactor**. These proteins on hydrolysis yield amino acids and non-protein material. Examples are

Mucin in saliva (prosthetic group, carbohydrate), casein in milk (prosthetic group, phosphoric acid), haemoglobin in blood (prosthetic group, iron pigment), etc.

According to molecular shape, proteins are divided into two types

(i) **Fibrous proteins:** These are made up of polypeptide chains that run parallel to the axis and are held together by strong hydrogen and disulphide bonds. They can be stretched and contracted like a thread. These are usually insoluble in water. Examples are: α -keratin (hair, wool, silk and nails); myosin (muscles); collagen (tendons, bones), etc.

(ii) **Globular proteins:** These have more or less spherical shape (compact structure). α -helices are tightly held up by weak attractive forces of various types: Hydrogen bonding, disulphide bridges, ionic or salt bridges. These are usually soluble in water. Examples are: Insulin, pepsin, haemoglobin, cytochromes, albumins, etc.

Proteins can also be classified on the basis of their function

Protein	Function	Examples
Enzymes	Biological catalysts, vital to all living systems.	Trypsin, pepsin.
Structural proteins	Proteins that hold living systems together.	Collagen.
Hormones	Act as messengers.	Insulin.
Transport proteins	Carry ions or molecules from place to another in the living system.	Haemoglobin.
Protective proteins (antibiotics)	Destroy any foreign substance released into the living system.	Gamma globulin.



(7) General and physical characteristic of proteins

(i) Most of them (except chromoproteins) are colorless, tasteless, and odorless. Many are amorphous but few are crystalline. They are nonvolatile and do not have a sharp melting point.

(ii) Most of them are insoluble in water and alcohol. But many of them dissolve in salt solutions, dilute acids and alkalis. Some proteins such as keratins (skin, hair and nails) are completely insoluble.

(iii) Protein molecules are very complex and possess very high molecular masses. They are hydrophilic colloids which cannot pass through vegetable or animal membrane. On addition of sodium chloride, ammonium sulphate magnesium sulphate, etc., some proteins are precipitated. The precipitate can be filtered and redissolved in water.

(iv) The solution of proteins are optically active. Most of them are laevorotatory. The optical activity is due to the presence of asymmetric carbon atoms in the constituent α -amino acids.

(v) **Isoelectric point:** Every protein has a characteristic isoelectric point at which its ionisation is minimum. Like amino acids, proteins, having charged groups (NH_3^+ and COO^-) at the ends of the peptide chain, are amphoteric in nature. In strong acid solution, protein molecule accepts a proton while in strong basic solution it loses a proton. **The pH at which the protein molecule has no net charge is called its isoelectric point.** This property can be used to separate proteins from mixture by electrophoresis.

(vi) **Denaturation:** The structure of the natural proteins is responsible for their biological activity. These structures are maintained by various attractive forces between different parts of the polypeptide chains. The breaking of these forces by a physical or a chemical change makes the proteins to lose all or part of their biological activity. This is called denaturation of proteins. The denaturing of proteins can be done by adding chemicals such as acids, bases, organic solvents, heavy metal ions, or urea. It can also be done with the help of heat and ultraviolet light. Denaturation can be irreversible or reversible. In irreversible denaturation, the denatured protein does not return to its original shape. For example, the heating of white of an egg (water soluble) gives a hard and rubbery insoluble mass.



(8) Chemical properties

(i) **Salt formation:** Due to presence of both $-NH_2$ and $-COOH$ groups in proteins, they form salts with acids and bases. Casein is present in milk as calcium salt.

(ii) **Hydrolysis:** The simple proteins are hydrolysed by acids, alkalies or enzymes to produce amino acids. Following steps are involved in the hydrolysis and the final product is a mixture of amino acids.

Protein \rightarrow Proteose \rightarrow Peptone \rightarrow Polypeptide \rightarrow Simple peptide \rightarrow Mixture of amino acids

(iii) **Oxidation:** Proteins are oxidised on burning and putrefaction. The products include amines, nitrogen, carbon dioxide and water. The bad smell from decaying dead animals is largely due to the formation of amines by bacterial oxidation of body proteins.

(9) Test of proteins

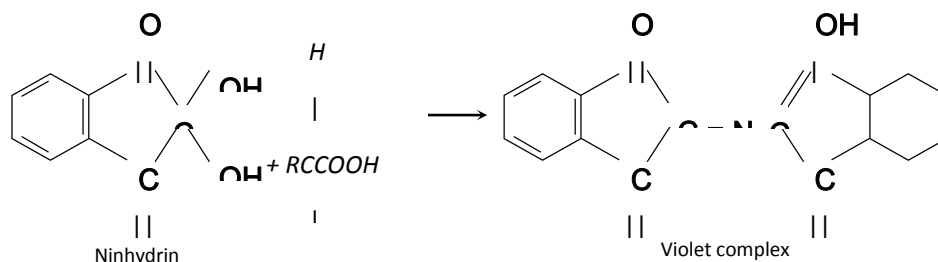
(i) **Biuret test:** On adding a dilute solution of copper sulphate to alkaline solution of protein, a violet colour is developed. This test is due to the presence of peptide ($-CO-NH-$) linkage.

(ii) **Xanthoproteic test:** Some proteins give yellow colour with concentrated nitric acid (formation of yellow stains on fingers while working with nitric acid in laboratory). The formation of yellow colour is due to reaction of nitric acid with benzenoid structures. Thus, when a protein solution is warmed with nitric acid a yellow colour may be developed which turns orange on addition of NH_4OH solution.

(iii) **Millon's test:** When millon's reagent (mercurous and mercuric nitrate in nitric acid) is added to a protein solution, a white precipitate which turns brick red on heating, may be formed. This test is given by proteins which yield tyrosine on hydrolysis. This is due to presence of phenolic group.

(iv) **Ninhydrin test: This test is given by all proteins.** When a protein is boiled with a dilute solution of ninhydrin, a violet colour is produced.

(v) **Nitroprusside test:** Proteins containing $-SH$ group give this test. When sodium nitroprusside solution is added to proteins having $-SH$ group, a violet colour is developed.



(vi) **Molisch's test:** This test is given by those proteins which contain carbohydrate residue. On adding a few drops of alcoholic solution of α -naphthol and concentrated sulphuric acid to the protein solution, a violet ring is formed.

(vii) **Hopkins-Cole test:** On adding concentrated sulphuric acid down the side containing a solution of protein and glyoxalic acid, a violet colour is developed.

(10) **Uses**

(i) Proteins constitute as essential part of our food. Meat, eggs, fish, cheese provide proteins to human beings.

(ii) **In textile:** Casein (a milk protein) is used in the manufacture of artificial wool and silk.

(iii) **In the manufacture of amino acids:** Amino acids, needed for medicinal use and feeding experiments, are prepared by hydrolysis of proteins.

(iv) **In industry:** Gelatin (protein) is used in food products, capsules and photographic plates. Glue (protein) is used as adhesive and in sizing paper. Leather is obtained by tanning the proteins of animal hides.

(v) **In controlling body processes:** Haemoglobin present in blood is responsible for carrying oxygen and carbon dioxide. Hormones (proteins) control various body processes.

(vi) **As enzymes:** Reactions in living systems always occur with the aid of substances called enzymes.

Enzymes are proteins produced by living systems and catalyse specific biological reactions.

Important enzymes are

Enzymes	Reaction catalysed
Urease	Urea \rightarrow CO ₂ + NH ₃
Invertase	Sucrose \rightarrow Glucose + Fructose
Maltase	Maltose \rightarrow 2 Glucose
Amylase	Starch \rightarrow n Glucose
Pepsin	Proteins \rightarrow Amino acids
Trypsin	Proteins \rightarrow Amino acids
Carbonic anhydrase	H ₂ CO ₃ \rightarrow H ₂ O + CO ₂



6. Nucleic acids.

In every living cell there are found nucleo-proteins which are made up of proteins and natural polymers of great biological importance called nucleic acids. Nucleic acids are complex compounds of carbon, hydrogen, oxygen, nitrogen and phosphorus. They play an essential role in transmission of the hereditary characteristics and biosynthesis of proteins. The genetic information coded in nucleic acids programmes the structure of all proteins including enzymes and thereby all metabolic activity of living organisms.

Two types of nucleic acids are found in biological systems, these are

Deoxyribonucleic acid (DNA) and

Ribonucleic acid (RNA)

The nucleic acid was first isolated by Friedrich Miescher in 1868 from the nuclei of pus cells and was named nuclein. The term nuclein was given by Altman.

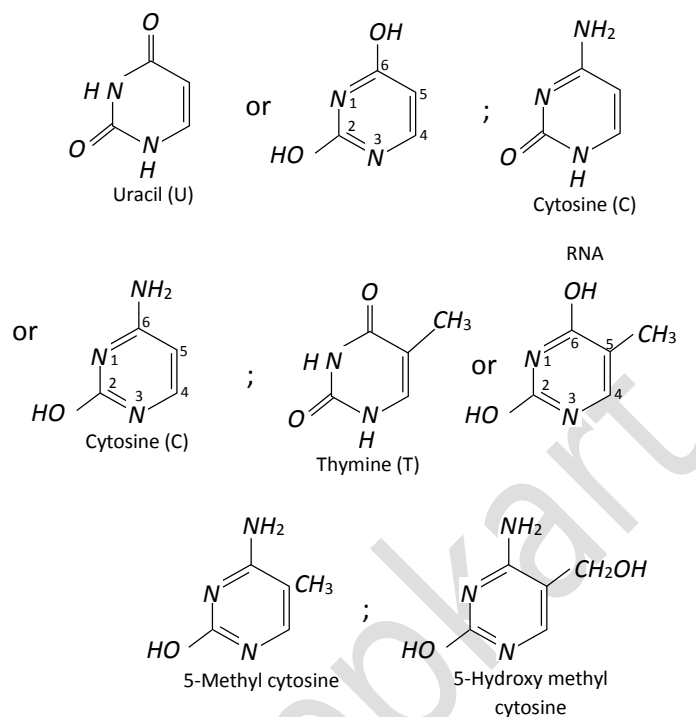
(1) **Composition: Nucleic acids like proteins and carbohydrates are polymers.** The simple units that make up the nucleic acid are called **nucleotides**. Nucleotides are themselves composed of following three simple molecules.

(i) **Nitrogenous base:** These are heterocyclic organic compound having two or more nitrogen atoms in ring skeleton. These are called bases because the lone pairs of electrons on the nitrogen atoms make them as Lewis bases.

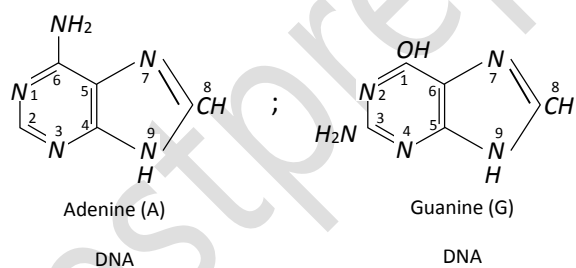


Their structures are given below

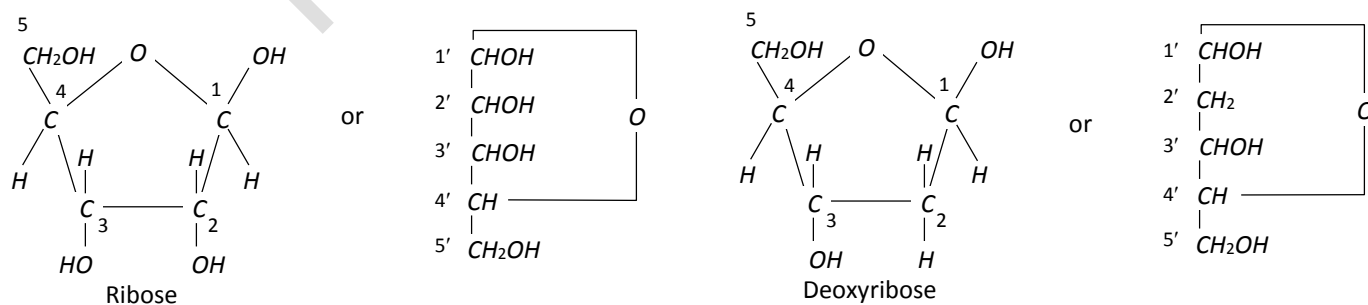
(a) Pyrimidine derivatives



(b) Purine derivatives



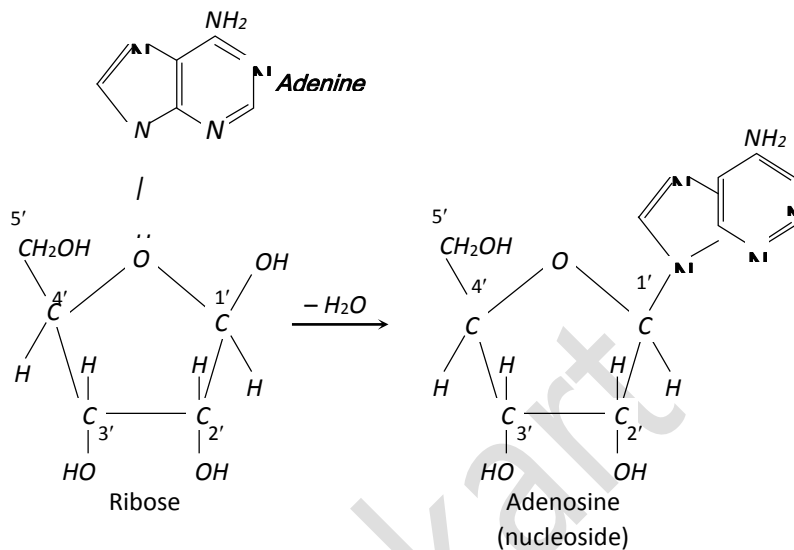
(ii) **Five carbon sugar (Pentose):** In RNA, the sugar is ribose where as in DNA, the sugar is deoxyribose.



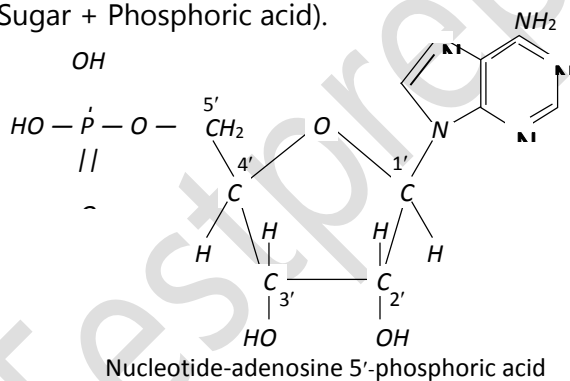
Both differ only at carbon atom 2' in the ring.



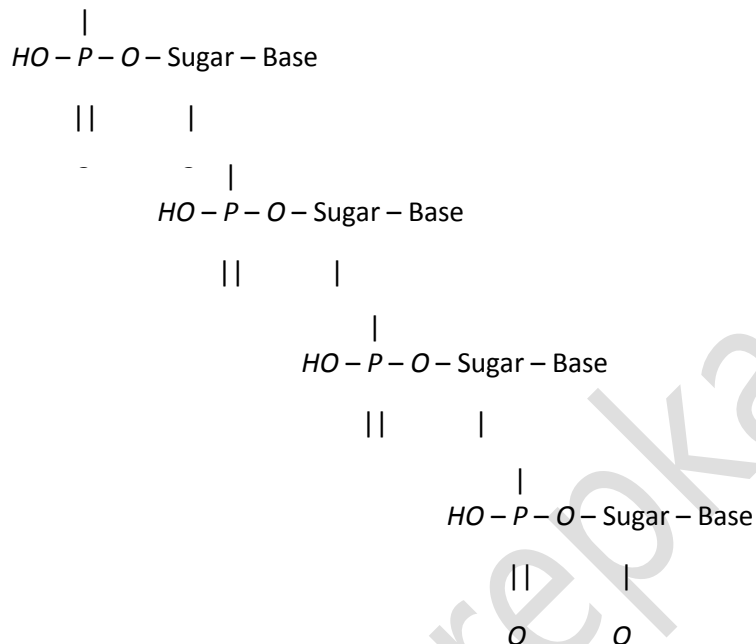
(iii) **Phosphoric acid, H_3PO_4** : Phosphoric acid forms esters to $-OH$ groups of the sugars to bind nucleotide segments together. A molecule called **nucleoside** is formed by condensing a molecule of the base with the appropriate pentose. (i.e., Base + Sugar).



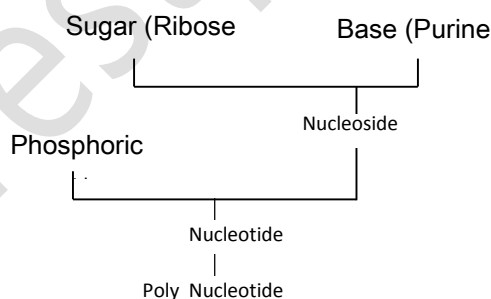
A **nucleotide** results when the nucleoside combined with phosphoric acid mainly at carbon 5' of the pentose. (i.e., Base + Sugar + Phosphoric acid).



This nucleotide is the building block of both DNA and RNA. The nucleic acids are condensation polymers of the nucleotide monomers and are formed by the creation of an ester linkage from phosphoric residue on one nucleotide to the hydroxy group on carbon 3' in the pentose of the second nucleotide. The result is a very long chain possessing upto a billion or so nucleotides units in DNA.



Thus, the formation of a nucleic acid can be summarised in the following general way



RNA nucleotides

Base	+	Ribose	→	(Nucleoside)	+	Phosphoric acid	→	Nucleotide
Adenine	+	Ribose	→	(Adenosine)	+	Phosphoric acid	→	Adenylic acid
Guanine	+	Ribose	→	(Guanosine)	+	Phosphoric acid	→	Guanylic acid
Cytosine	+	Ribose	→	(Cytidine)	+	Phosphoric acid	→	Cytidylic acid
Uracil	+	Ribose	→	(Uridine)	+	Phosphoric acid	→	Uridylic acid

DNA nucleotides

Adenine	+	Deoxy ribose sugar	+	Phosphate	→	Adenosine phosphate
Guanine	+	Deoxy ribose sugar	+	Phosphate	→	Guanosine phosphate
Cytosine	+	Deoxy ribose sugar	+	Phosphate	→	Cytosine phosphate
Thymine	+	Deoxy ribose sugar	+	Phosphate	→	Thymidine phosphate



(2) **Structure:** The sequence of bases along the DNA and RNA chain establishes its primary structure which controls the specific properties of the nucleic acid. An RNA molecule is usually a single chain of ribose-containing nucleotides. DNA molecule is a long and highly complex, spirally twisted, double helix, ladder like structure. The two polynucleotide chains or strands are linked up by hydrogen bonding between the nitrogenous base molecules of their nucleotide monomers. Adenine (purine) always links with thymine (pyrimidine) with the help of two hydrogen bonds and guanine (purine) with cytosine (pyrimidine) with the help of three hydrogen bonds. Hence, the two strands extend in opposite directions, i.e., are antiparallel and complimentary. The following fundamental relationship exist.

Note: Thymine combines only with deoxyribose sugar and uracil only with ribose sugar. Other bases can combine with either of the two sugars.

- The sum of purines equals the sum of pyrimidines.
- The molar proportion of adenine equals to that of thymine.
- The molar proportion of guanine equals to that of cytosine.

The double helix is 20 Å. It completes a spiral at every 10 nucleotide pairs at a length of 34 Å. Sequences of monomers (nucleotides) may present innumerable variations. Evidently, innumerable varieties of DNA exist in the organism.

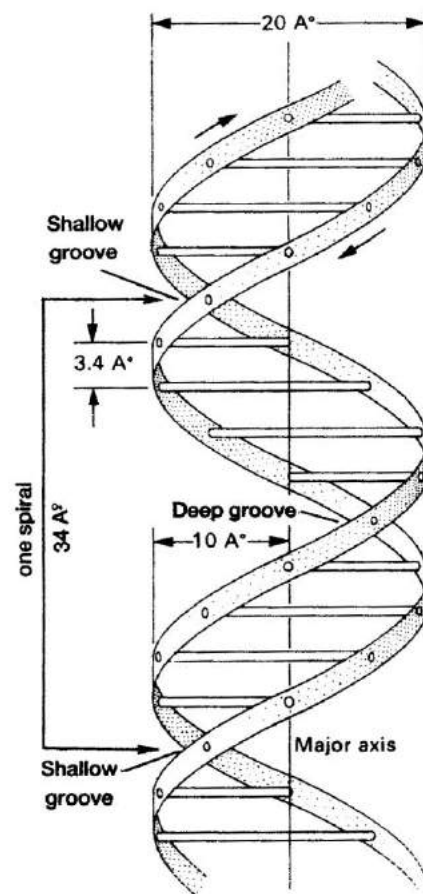


Fig : Helical structure of DNA

Watson, Crick and Witkins were awarded Noble prize in 1962 for suggesting the structure of DNA.

Difference between DNA and RNA

DNA	RNA
It has a double helix structure.	It has a single helix structure.
Sugar unit is deoxyribose.	Sugar unit is Ribose.
Base units are adanine, guanine, thyamine and cytosine.	It contains uracil base instead of thyamine, other bases being same as those in DNA.
Responsible for inheritance of character.	It is responsible for protein synthesis.



(3) **Functions of nucleic acid:** Nucleic acid have two important functions

(i) **Replication and**

(ii) **Protein synthesis**

(i) **Replication:** The genetic information for the cell is contained in the sequence of the bases A, T, G and C (adenine, thymine, guanine and cytosine) in the DNA molecule. The sequence of bases in one chain of the double helix controls the sequence in other chain. The two chains fit together like a hand and a glove. They separate and about the hand is formed a new glove, and inside the glove is formed a new hand. Thus, the pattern is preserved in the two new molecules of DNA.

[If one strand of DNA has the sequence ATGCTTGA, then the sequence of complimentary strand will be TACGAACT].

(ii) **Synthesis of proteins:** The DNA contains the genetic code and directs protein synthesis through RNA. The double helix of DNA partially uncoils and about the individual strands are formed chains of RNA. The new chains contain ribose instead of deoxyribose and the base sequence is different which is determined by DNA, i.e., opposite each adenine of DNA, there appears on RNA a uracil; opposite guanine, cytosine; opposite thymine, adenine, opposite cytosine, guanine. Thus, AATCAGTT on DNA becomes UUAGUCAA on RNA.

One kind of RNA, called messenger RNA, carries a message to the ribosome, where protein synthesis actually takes place. At the ribosome, messenger RNA calls up a series of transport RNA molecules, each of which is loaded with a particular amino acid. The order in which the transport RNA molecules are called (–the sequence in which the amino acids are arranged to form the protein chain) depends upon the sequence of bases along the messenger RNA chain. Thus GAU is the code for aspartic acid; UUU, phenyl alanine; GUG, valine. There are 64-three letter code words (codons) and only 20-odd amino acids, so that more than one codon call the same amino acid.

The relation between the nucleotide triplets and the amino acids is called Genetic code. Nirenberg, Hollay and Khorana presented the genetic code for which they were awarded Noble prize in 1968.

(4) **Mutation:** A mutation is a chemical or physical change that alters the sequence of bases in DNA molecule. Anything that causes mutation is called **mutagen**. A mutation results from ultraviolet light, ionisation radiations, chemicals or viruses. The changes in sequence of bases in DNA are repaired by special enzymes in the cell. If it is not, the protein produced has no biological activity and the cell dies.



These mutations often prove harmful and give rise to symptoms that cause diseases. Sickle-cell anaemia is one such example. Such disease is passed on from one generation to the next generation.

7. Lipids.

Lipids are constituents of plants and tissues which are insoluble in water but soluble in organic solvents such as chloroform, carbon tetrachloride, ether or benzene. They include a large variety of compounds of varying structures such as oils and fats; phospholipids, steroids, etc. Lipids are mainly made of carbon, hydrogen and oxygen. The number of oxygen atoms in a lipid molecule is always small as compared to the number of carbon atoms. Sometimes small amounts of phosphorus, nitrogen and sulphur are also present. They have a major portion of their structure like a hydrocarbon (aliphatic or fused carbon rings). Lipids serve as energy reserve for use in metabolism and as a major structural material in cell membranes for regulating the activities of cell and tissues.

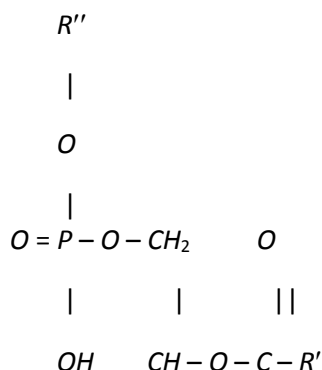
Simple lipids are esters of glycerol with long chain monocarboxylic acids which can be saturated or unsaturated. These are generally called glycerides of fats and oils. Waxes are esters of fatty acids with certain alcohols, not glycerol. Fats and oils have biological importance but waxes have no value as these are not digested.

The functions of triglycerides are the following

- (1) They are energy reserves in the cells and tissues of living system. When digested, triglycerides are hydrolysed to fatty acids and glycerol.
- (2) Catabolism of fatty acids form acetyl-coenzyme-A. Most of the energy of fatty acids is converted into ATP.
- (3) Acetyl coenzyme is the starting material for the synthesis of many compounds.
- (4) Fats deposited beneath the skin and around the internal organs minimise loss of body heat and also act as cushions to absorb mechanical impacts.



Another very important class of lipids are the phospholipids. These are polar lipids and like the fats, are esters of glycerol. In this case, however, only two fatty acid molecules are esterified to glycerol, at the first and second carbon atom. The remaining end position of the glycerol is esterified to a molecule of phosphoric acid, which in turn is also esterified to another alcohol. This gives a general structure.



The alcoholic compound linked to phosphoric group may be choline, ethanol, amine, serine or inositol. The phosphate groups forms a polar end, i.e., hydrophilic (water-attracting) and the two fatty acid chains constitute the non-polar tail, i.e., hydrophobic (water repelling). This structure gives the phospholipids good emulsifying and membrane forming properties.

Cell membranes are composed of phospholipids and proteins in about equal, proportion. The phospholipids in the membrane appear to be arranged in a double layer or bilayer in which the non-polar tails face each other, thereby exposing the polar heads to the aqueous environment on either side of the membrane. Proteins found in the membrane are embedded in the mosaic formed by the lipids. Phospholipids facilitate the transport of ions and molecules in and out of the cell and regulate the concentration of molecules and ions within the cell. They provide structural support for certain proteins. The above mentioned lipids are mainly straight chain compounds. There is a third class of lipids which are not straight chain compounds. They are called **Sterols**.

The sterols are composed of fused hydrocarbon rings and a long hydrocarbon side chain. Cholesterol is most important compound of this class and is found in animals only. It exists either free or as ester with a fatty acid. Cholesterol is also the precursor of hormones. Cholesterol and its esters are insoluble in water. So they are deposited in the arteries and veins if the blood cholesterol rises. This leads to high blood pressure and heart diseases. Cholesterol is a part of animal cell membrane and is used to synthesis steroid hormones, vitamin-D and bile salts.



8. Energy cycle or metabolism.

A cell has small molecules (micromolecules) as well as large molecules (macromolecules). The chemical reactions of a living organism can be divided into main two types

(1) The chemical reactions by which the large molecules are constantly broken down into smaller ones are called **catabolism**.

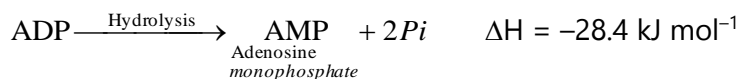
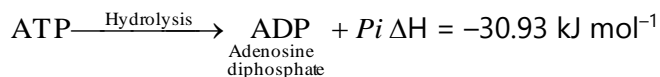
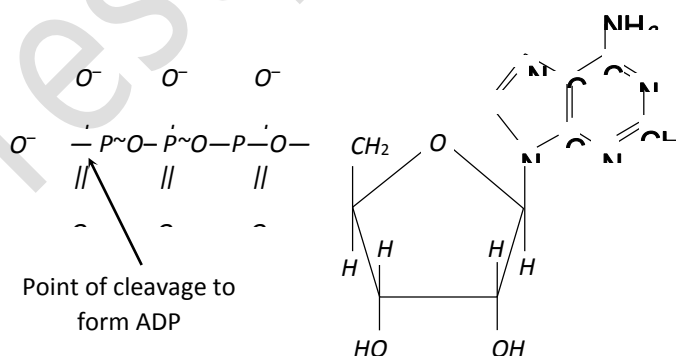
(2) The chemical reactions by which the macromolecules are synthesised within the cell are called **anabolism**.

The two processes i.e., degradation and synthesis are collectively called **metabolism**. Catabolism reactions are usually accompanied by release of energy whereas anabolism reactions require energy to occur.

The primary energy found in living cells is chemical energy, which can be easily stored, transferred and transformed. For this, the living cells contain a chemical compound called **adenosine triphosphate** (ATP). It is regarded as **energy currency** of living cells because it can trap, store and release small packets of energy with ease.

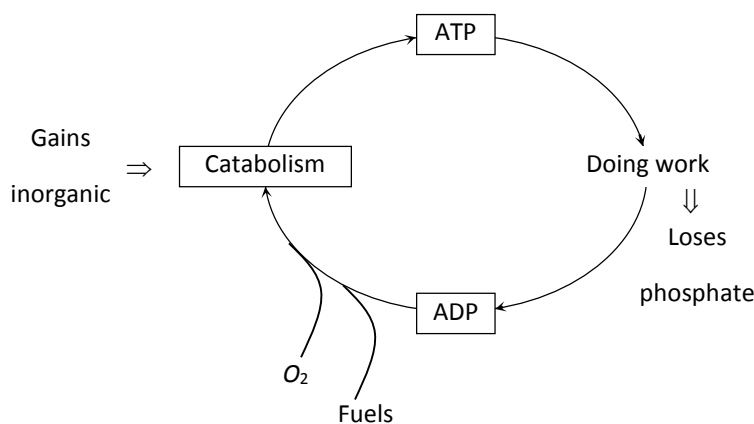
ATP consists of a purine base called **adenine** linked to a five carbon sugar named **ribose** which is further attached to **three molecules of phosphate**.

ATP is energy rich molecule, this is because of the presence of four negatively charged oxygen atom very close to each other. These four negatively charged o-atoms experience very high repulsive energy.



ADP can change to ATP in the presence of inorganic phosphate. This process is called **phosphorylation**.

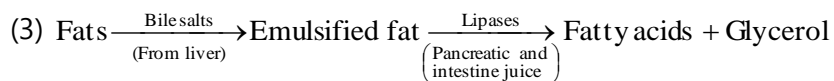
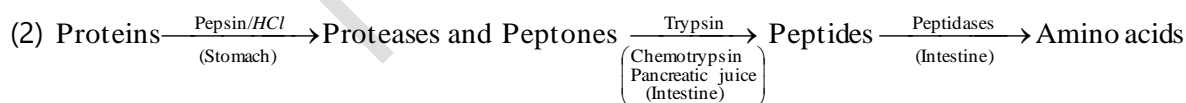




9. Digestion of food.

Digestion is the process by which complex constituents of food are broken down into simple molecules by a number of enzymes in mouth, stomach and small intestine. The simple molecules thus formed are absorbed into blood stream and reach various organs.

Raw food may be taken as such or after cooking. It is chewed in the mouth and swallowed when it passes through a long passage in the body called alimentary canal. During this passage it gets mixed with various enzymes in different parts of the alimentary canal. The carbohydrates, proteins and fats are converted into simpler forms which are then carried by blood to different parts of the body for utilisation. Digestion of food can be summarised in the following form



After digestion, there are present glucose, amino acids, fatty acids along with vitamins and mineral salts. Undigested food and secretions are pushed forward into the rectum from where these are excreted.



10. Vitamins.

In addition to air, water, carbohydrates, proteins, fats and mineral salts, certain organic substances required for regulating some of the body processes and preventing certain diseases are called **vitamins**. These compounds cannot be synthesized by an organism. These vitamins are required in small amounts and deficiency of any one causes one disease or the other. Thus, vitamins are essential constituents of our diet. Several of these vitamins are known and are designated as A, B, C, D, E and so on. Many of these are now synthesized on commercial scale. On the basis of solubility, the vitamins are divided into two groups.

(1) Fat soluble; Vitamin A, D, E and K.

(2) Water soluble; Vitamin B and C.

Name	Sources	Functions	Effects of deficiency
Water soluble vitamins			
B ₁ (Thiamine or Aneurin) (C ₁₂ H ₁₈ N ₄ SOCl ₂)	Rice polishings, wheat flour, oat meal, eggs, yeast, meat, liver, etc.	Major component of co-enzyme co-carboxylase required for carbohydrate and amino acid metabolism.	Beri-beri , loss of appetite and vigour, constipation, weak heartbeat, muscle atrophy, even paralysis.
B ₂ or G (Riboflavin or Lactoflavin) (C ₁₇ H ₂₀ N ₄ O ₆)	Cheese, eggs, yeast, tomatoes, green vegetables, liver, meat, cereals, etc.	Combines with phosphoric acid to form coenzyme FAD essential for oxidative metabolism.	Cheilosis , digestive disorders, burning sensations in skin and eyes, headache, mental depression, scaly dermatitis at angles of nares, corneal opacity, etc.
B ₃ (Pantothenic acid) (C ₉ H ₁₇ O ₅ N)	All food; more in yeast, liver, kidneys, eggs, meat, milk, sugarcane, groundnut, tomatoes.	Important component of Co-A required for oxidative metabolism.	Dermatitis , in cocks; greying of hairs, retarded body and mental growth, reproductive debility.



B ₅ or P-P (Nicotinic acid or Niacin) C ₆ H ₅ NO ₂ (C ₅ H ₄ N-COOH)	Fresh meat, liver, fish, cereals, milk, pulses, yeast, etc.	Active group in coenzyme NAD required for oxidative metabolism.	Pellagra , dermatitis, diarrhoea, dementia, muscle atrophy, inflammation of mucous membrane of gut.
B ₆ (Pyridoxine or Adermin) (C ₈ H ₁₁ O ₃ N)	Milk, cereals, fish, meat, liver, yeast synthesised by intestinal bacteria.	Important coenzyme required in protein and amino acid metabolism.	Dermatitis, anaemia , convulsions, nausea, insomnia, vomiting, mental disorders, depressed appetite.
Vit. H (Biotin) (C ₁₀ H ₁₆ N ₂ O ₃ S)	Yeast, vegetables, fruits, wheat, chocolate, eggs, groundnut synthesised by intestinal bacteria.	Essential for fat synthesis and energy production.	Skin lesions , loss of appetite, weakness, hairfall, paralysis.
Folic acid group	Green vegetables, soyabean, yeast, kidneys, liver, synthesised by intestinal bacteria.	Essential for synthesis of DNA and maturation of blood corpuscles.	Retarded growth, anaemia .
B ₁₂ (Cyanocobalamine) (C ₆₃ H ₈₈ O ₁₄ N ₁₄ PCo)	Meat, fish, liver, eggs, milk synthesised by intestinal bacteria.	Required for chromosome duplication and formation of blood corpuscles.	Retarded growth, pernicious anaemia
Vit. C (Ascorbic acid) (CH ₈ O ₆)	Lemon, orange and other citrus fruits, tomatoes, green vegetables, potatoes, carrots, pepper, etc.	Essential for formation of collagen, cartilage, bone, teeth, connective tissue and RBCs and for iron metabolism.	Wound-healing and growth retarded, scurvy , breakdown of immune defence system , spongy and bleeding gums , fragile blood vessels and bones , exhaustion, nervous breakdown, high fever.
Fat soluble vitamins			
Vit. A (Retinol or Axerophthol) (C ₂₀ H ₃₀ O)	Synthesised in cells of liver and intestinal mucous membrane from carotenoid pigments found in milk, butter,	Essential for synthesis of visual pigments; growth and division of epithelial cells.	Xerophthalmia-keratinized conjunctive and opaque and soft cornea. Stratification and keratinization in epithelia of skin, respiratory



	kidneys, egg yolk, liver, fish oil, etc.		passages, urinary bladder, ureters and intestinal mucosa, night-blindness, impaired growth, glandular secretion and reproduction.
Vit. D (Ergocalciferol), (Sunshine vitamin) $C_{28}H_{44}O$ and cholecalciferol	Synthesised in skin cells in sunlight from 7-dehydro-cholesterol also found in butter, liver, kidneys, egg yolk, fish oil, etc.	Regulates absorption of calcium and phosphorus in intestine, mineral deposition in bones and teeth.	Rickets with osteomalacia; soft and fragile teeth.
Vit. E group Tocopherols (α, β, γ) ($C_{29}H_{50}O_2$)	Green vegetables, oil, egg yolk, wheat, animal tissues.	Essential for proper spermatogenesis, pregnancy, lactation and muscular function.	Sterility (impotency) and muscular atrophy.
Vit. K (Phylloquinone) ($C_{31}H_{46}O_2$)	Carrots, lettuce, cabbage, tomatoes, liver, egg yolk, cheese; synthesized by colon bacteria.	Essential for synthesis of prothrombin in liver, which is required for blood clotting.	Haemorrhages , excessive bleeding in injury, poor coagulation of blood.



11. Hormones.

Hormones are the chemical substances secreted by ductless glands, which influence and control biological reactions. Some important hormones along with their source and function are being given below.

Hormone	Source	Functions
Steroid hormones		
Testosterone (Androgens)	Testis	Regulates development of reproductive male organs.
Estrogene and progesterons	Ovary (Uterus)	Female sex hormones: control normal functioning of female sex organs.
Cortisone and related hormones	Adrenal cortex	Regulates the metabolism of fats, proteins, carbohydrates and mineral salts.
Amine hormones		
Adrenalino (Epinephrine)	Adrenal medulla	Increase the pulse rate and blood pressure: reduces glucose from glycogen and fatty acids from fats.
Thyroxine	Thyroid	Stimulates rate of oxidative metabolism and regulates general growth and development.
Peptide hormones		
Oxytocino	Posterior pituitary	Causes contraction of some smooth muscle. Also causes contraction of uterus during child birth.
Vasopressin	Posterior pituitary	Inhibits excretion of water from the body by way of urine.
Insulin	Pancreas	Decrease blood glucose level.
Glucogen	Pancreas	Elevates blood glucose level.

Product	Applications
Human insulin hormone	Treatment of diabetes
Interferon	Antiviral agent
Growth hormones	Treatment of abnormal growth related diseases
Tissue plasminogen factor	Dissolving unwanted blood clots
Blood clotting factor VIII	Treatment of haemophilias
Vaccines	Against various infective diseases

