INTRODUCTION TO BIOCHEMISTRY

Definition

The science that is concerned with the structures, interactions, and transformations of biological molecules. The chemistry of life

Biochemistry can be subdivided three principal areas

- Structural chemistry
- Metabolism
- The chemistry of processes and substances that store and transmit biological information (molecular genetics**)**

Biochemistry and Life

The cell is the fundamental unit of life

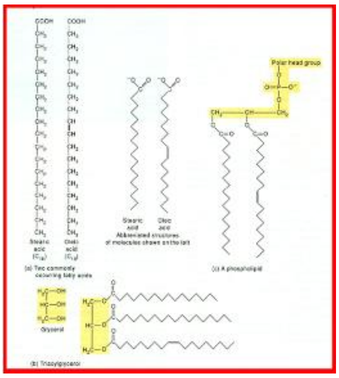
- Prokaryotes and eukaryotes
- Eukaryotic cells
- animal cells
- plant cells (chloroplasts and cell walls)

Cells are composed of:

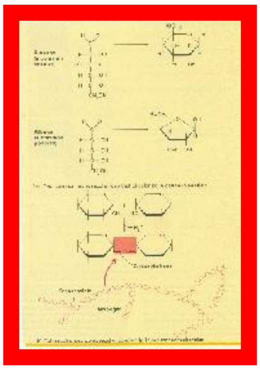
- Small molecules
- Macromolecules
- Organelles

Expect for water, most of the molecules found in the cell are macromolecules, can be classified into four different categories:

- Lipids
- Carbohydrates
- Proteins
- Nucleic acids
- ➢ Lipids are primarily hydrocarbon structures

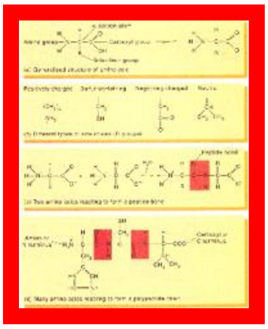


- ➢ Carbohydrates, like lipids, contain a carbon backbone, but they also contain many polar hydroxyl (-OH) groups and therefore very soluble in water.



- Proteins are the most complex macromolecules in the cell.
- They are composed of linear polymers called polypeptides, which contain amino acids connected by peptide bonds

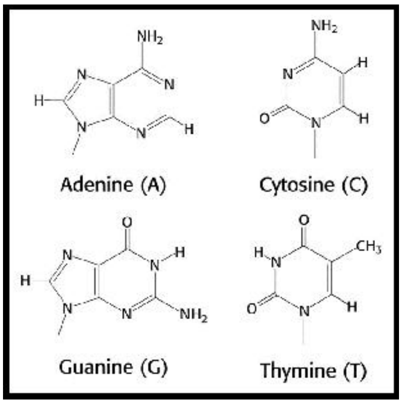
  Each amino acid contains a central carbon atom attached to four substituents
  - A carboxyl group
  - An amino group
  - A hydrogen atom
  - An R group



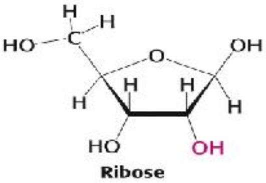
Nucleic acids are the large macromolecules in the cells.
They are very long linear polymers, called polynucleotides, composed of nucleotides
A nucleotide contains:

- A five-carbon sugar molecules
- One or more phosphate groups
- A nitrogenous base
- DNA: A, T, G, C
- RNA: A, U, G,C

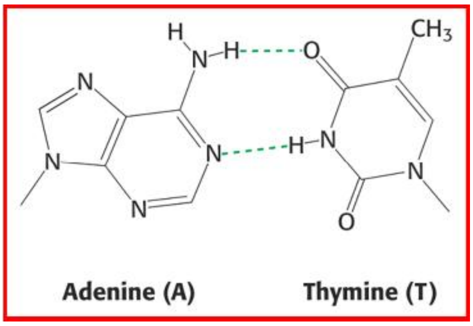
DNA Contain Four Bases

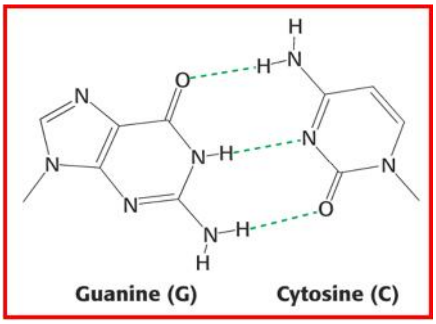


RNA

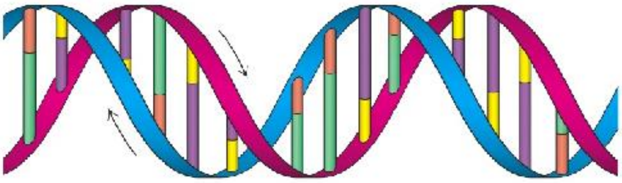


Watson-Crick base pairs





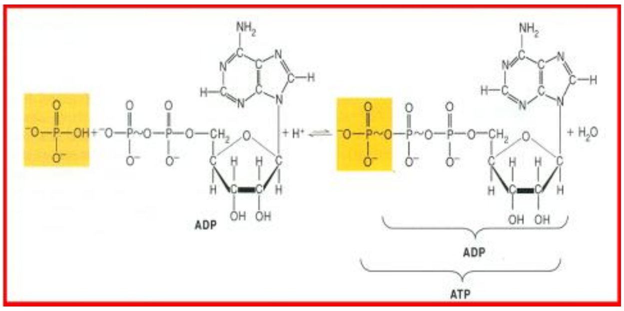
The Double Helix



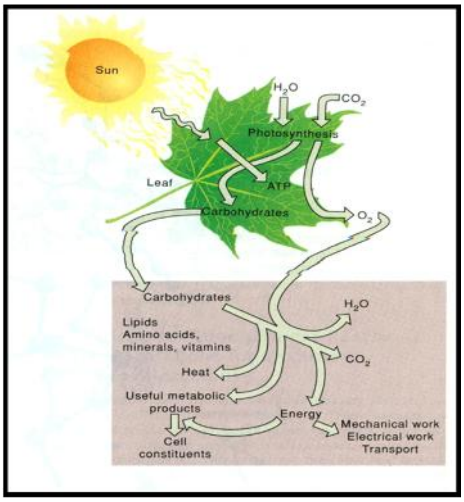
Biochemical Energy

- All cellular functions re quire energy.
- The most-important chemical form of energy in most cells is ATP, *adenosine 5'-triphosphate.*
- ATP $\longrightarrow$ ADP + P_i
- Most ATP synthesis occurs in chloroplasts and mitochondria

ADT and ATP Structures



Energy Transfer



Transfer of Information from DNA to Protein

CARBOHYDRATES

Occurrences

Carbohydrates are the most abundant organic compounds in the plant world. They act as storehouses of chemical energy (glucose, starch, glycogen); are the components of supportive structures in plants (cellulose), crustacean shells (chitin) and connective tissues in animals (acidic polysaccharides) and are essential components of nucleic acids (D-ribose and 2-deoxy-D-ribose). Carbohydrates make up about three fourths of the dry weight of plants.

The Nature of Carbohydrates

Carbohydrates are compounds of great importance in both the biological and commercial world They are used as a source of energy in all organisms and as structural materials in membranes, cell walls and the exoskeletons of many arthropods. Hydrates of carbon. All carbohydrates contain the elements carbon (C), hydrogen (H) and oxygen (O) with the hydrogen and oxygen being present in a 2 : 1 ratio

Carbohydrates are usually defined as

Polyhydroxy aldehydes or ketones, or substances that hydrolyze to yield polyhydroxy aldehydes or ketones”. (0r) Aldehyde or Ketone derivatives of polyhydroxy alcohols

Carbohydrates are aldehyde or ketone compounds with multiple hydroxyl groups

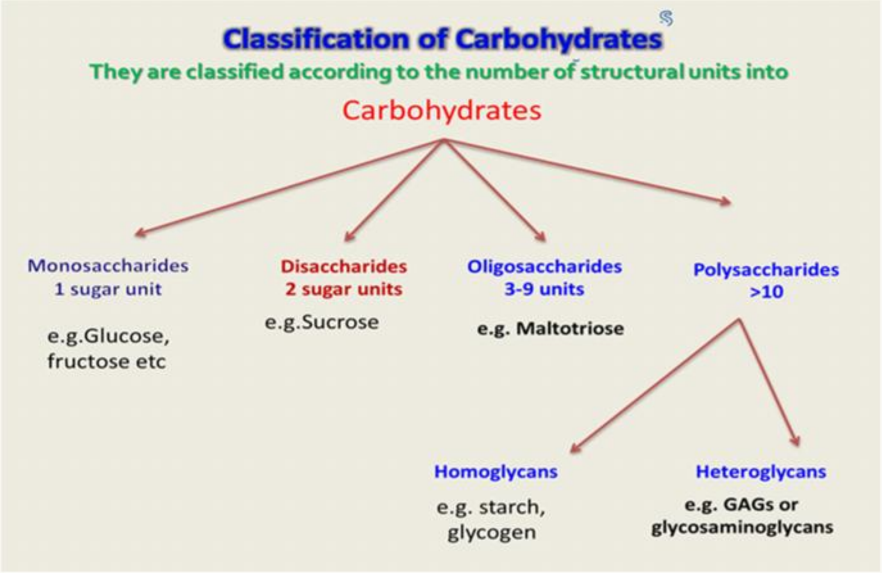
General molecular formula Cn (H2O)n

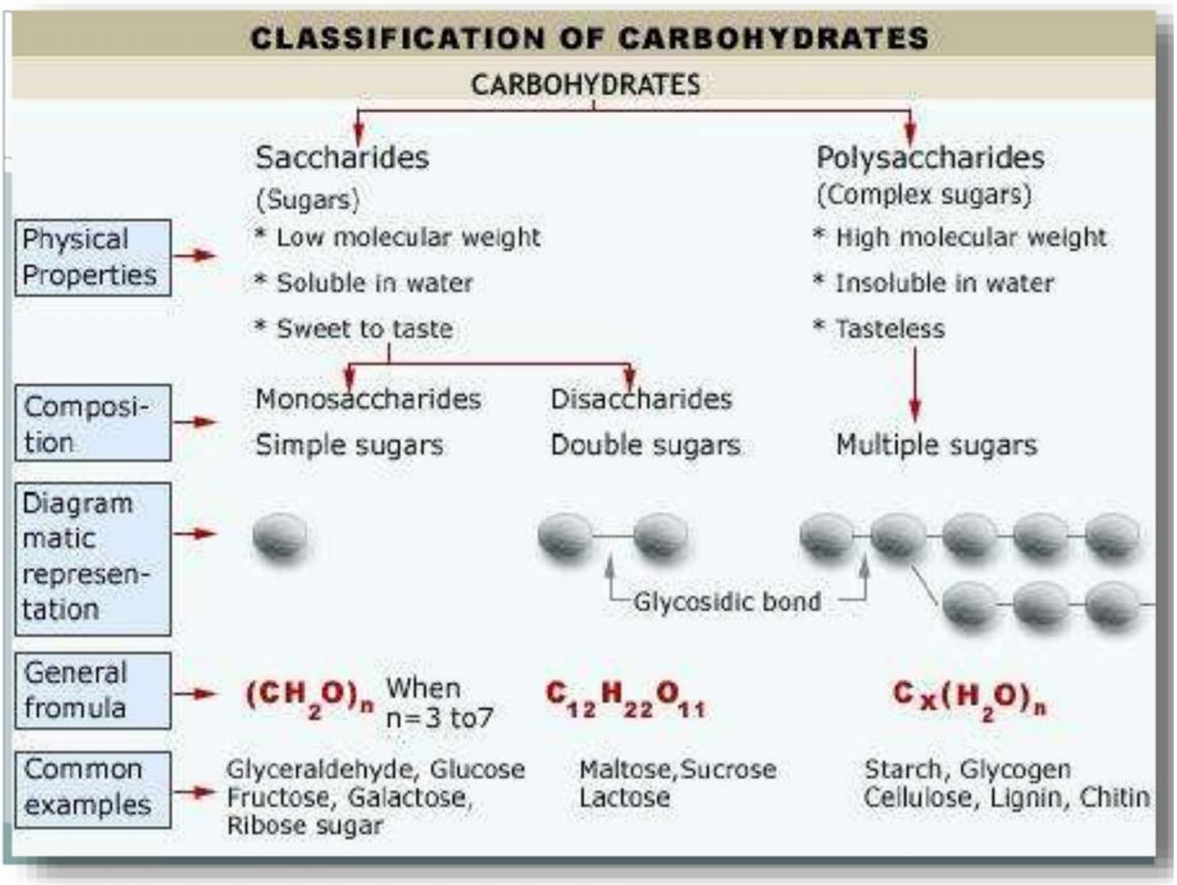
The term —carbohydrate comes from the observation that when you heat sugars, you get carbon and water (hence, hydrate of carbon).

Functions of carbohydrates

Main sources of ENERGY in body (4kcal/g)

– RBCs and Brain cells have an absolute requirement of carbohydrates
- Storage form of energy (starch and glycogen)
- Excess carbohydrate is converted to fat.
- Glycoproteins and glycolipids are components of cell membranes and receptors.
- Structural basis of many organisms.e.g. Cellulose in plants,
- exoskeleton of insects, cell wall of microbes,
- Mucopolysaccharides and ground substance in higher organisms.





CLASSIFICATION OF CARBOHYDRATES

CARBOHYDRATES

	Saccharides (Sugars)		Polysaccharides (Complex sugars)
Physical Properties	* Low molecular weight * Soluble in water * Sweet to taste		* High molecular weight * Insoluble in water * Tasteless
Composi-tion	Monosaccharides Simple sugars	Disaccharides Double sugars	Multiple sugars
Diagrammatic representation		Glycosidic bond	
General fromula	$(CH_2O)_n$ When n=3 to7	$C_{12}H_{22}O_{11}$	$C_x(H_2O)_n$
Common examples	Glyceraldehyde, Glucose Fructose, Galactose, Ribose sugar	Maltose,Sucrose Lactose	Starch, Glycogen Cellulose, Lignin, Chitin

MONOSACCHARIDES

Def : They are the simplest carbohydrate unites which cannot be hydrolysed to a simpler form They are classified into a) simple monosaccharides b) derived monosaccharides

Simple monosaccharides sub classified according to

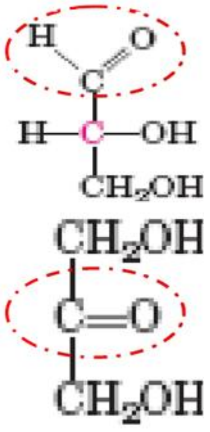
1-The number of carbon atoms present in their molecule and,

2- The type of carbonyl group they contain.

Derived monosaccharides include the derivatives of simple monosaccharides such as oxidation products, reduction products, substitution products and esters

* Nomenclature:

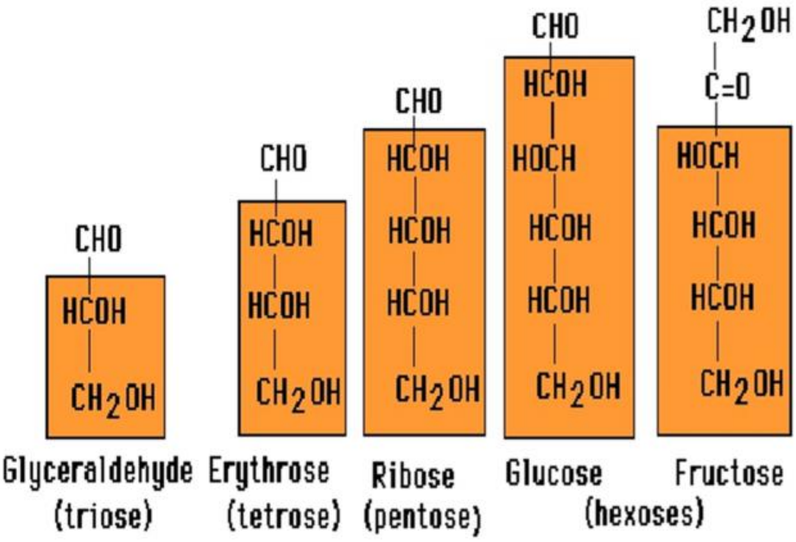
- 1- According to active group in the sugar:
  - If monosaccharide contains aldehyde group (CHO) → it's called aldose.
  - And if contain ketone group (c=o) → it's called ketose.



2.- **According to the number of carbon atoms (n):**

If sugar contains

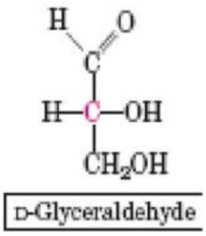
3 carbons → it’s called triose, 4c→ tetroses 5c→ pentose 6c→ hexose 7c→ heptoses

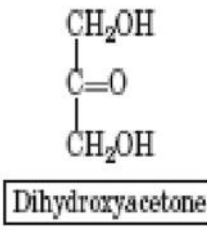


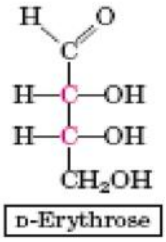
3- By combining the two methods, we find that

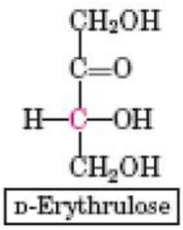
3c-Aldotriose	ketotriose
4c-Aldotetrose	Ketotetrose
5c-Aldopentise	Ketppentose
6c-Aldohexose	Ketohexose

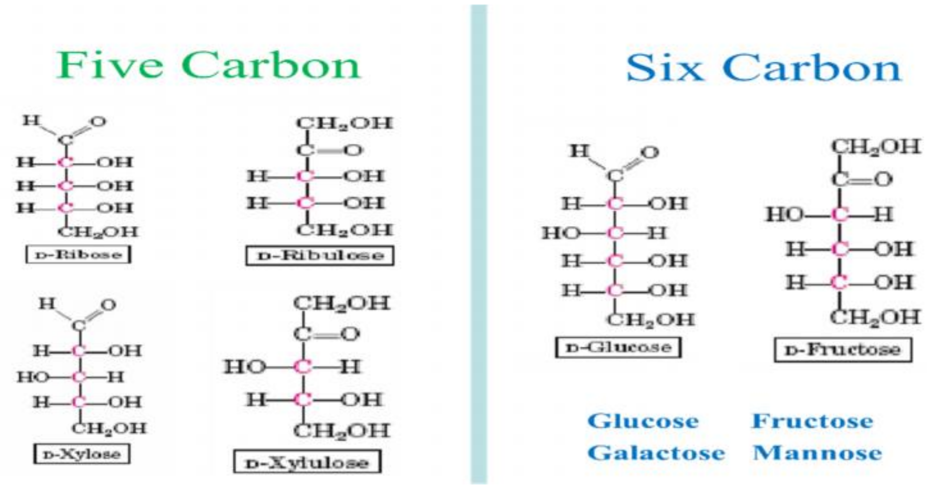
Three Carbon **Four Carbon**

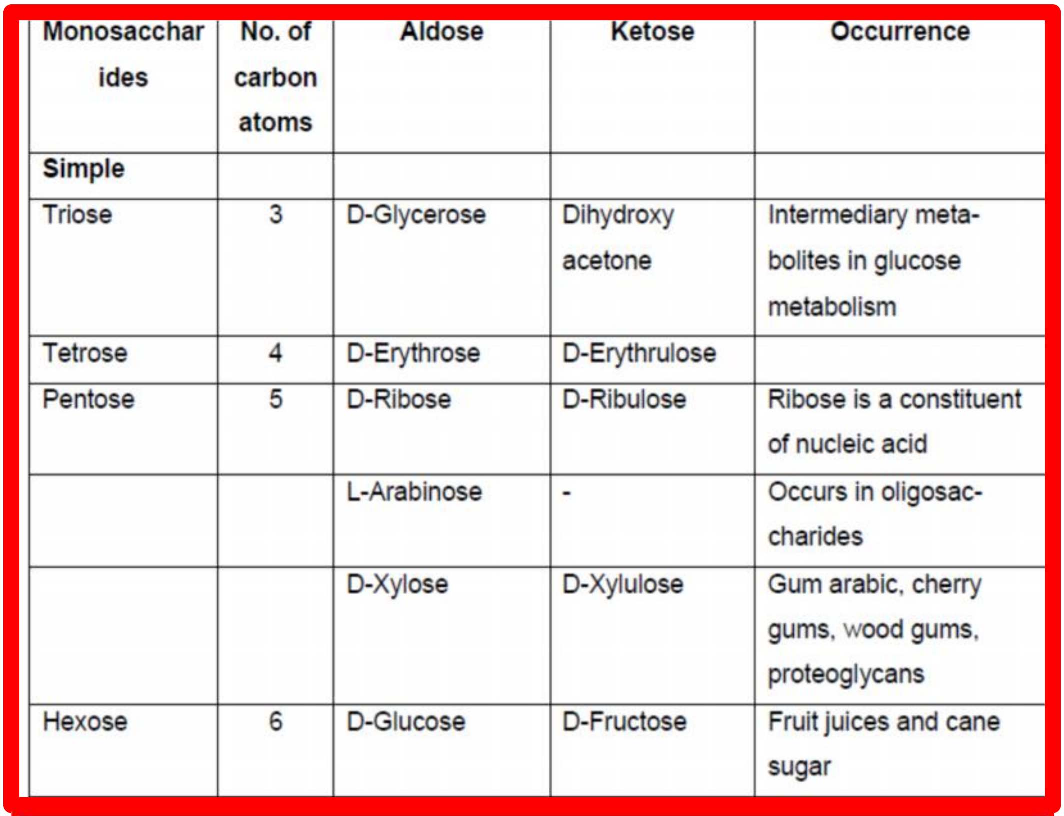


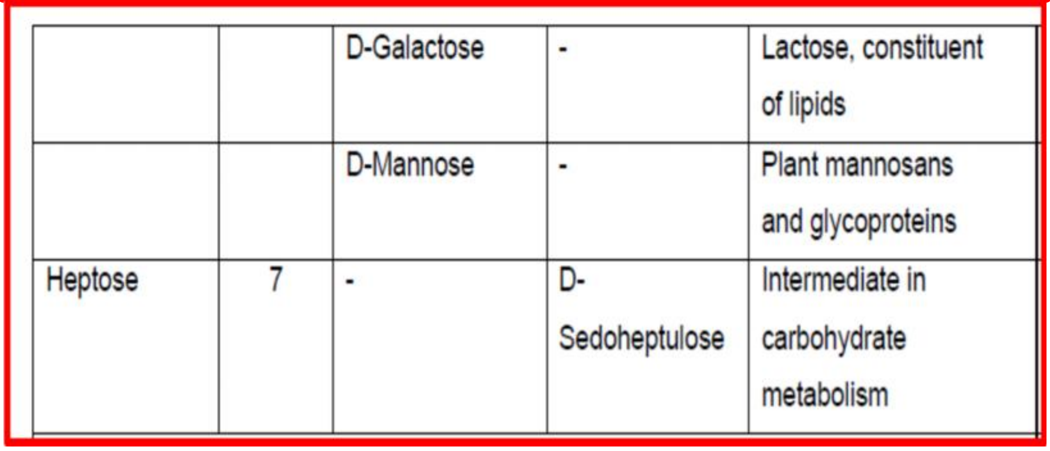




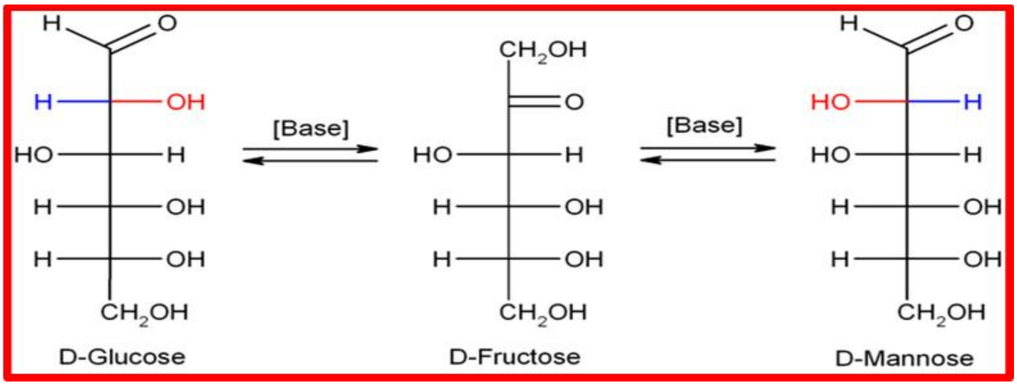


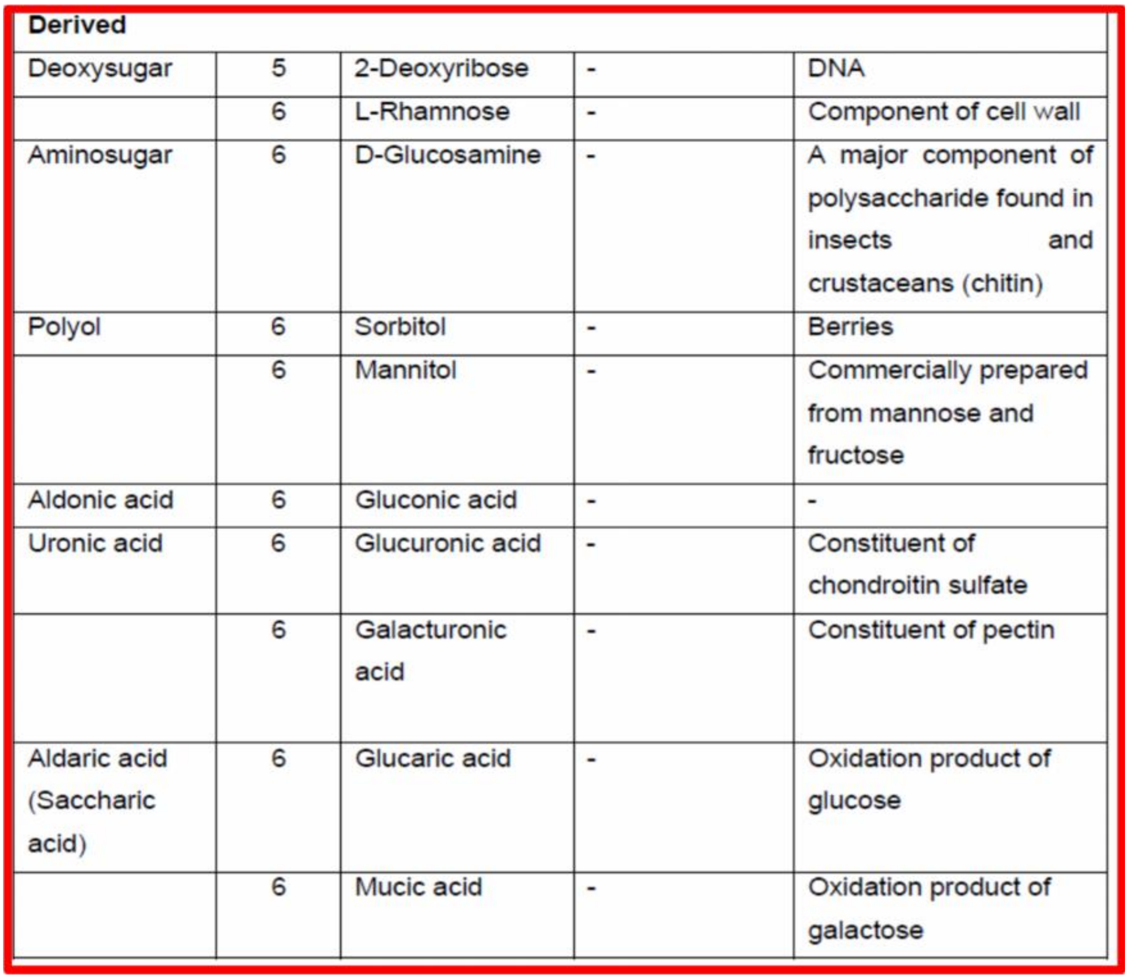






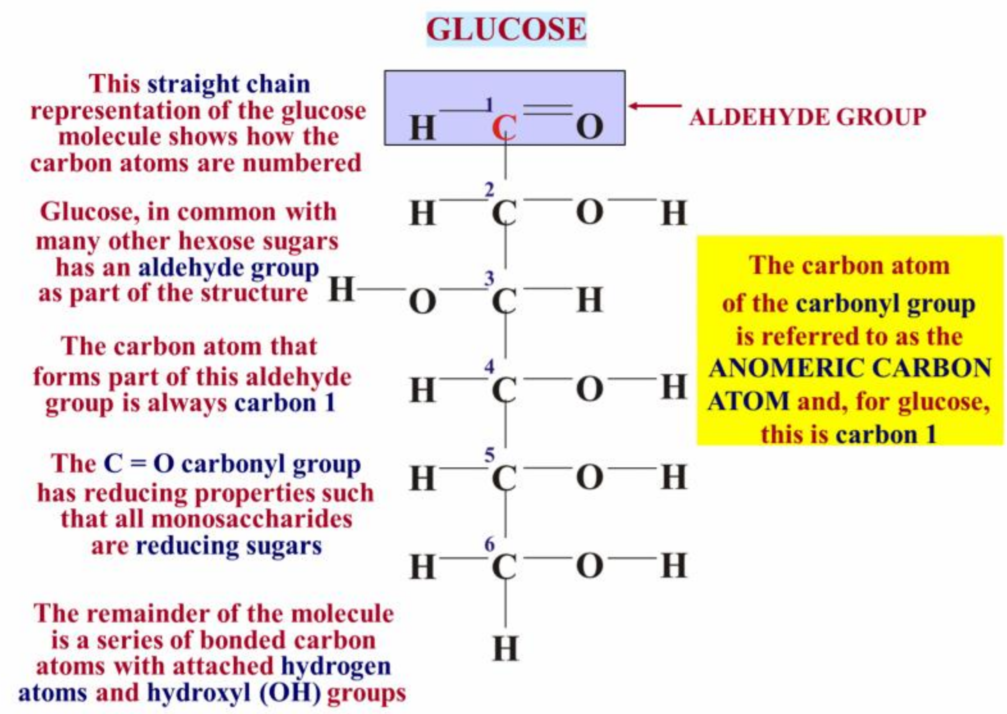
Monosaccharides	No. of carbon atoms	Aldose	Ketose	Occurrence
Simple				
Triose	3	D-Glycerose	Dihydroxy acetone	Intermediary metabolites in glucose metabolism
Tetrose	4	D-Erythrose	D-Erythrulose	
Pentose	5	D-Ribose	D-Ribulose	Ribose is a constituent of nucleic acid
		L-Arabinose	-	Occurs in oligosaccharides
		D-Xylose	D-Xylulose	Gum arabic, cherry gums, wood gums, proteoglycans
Hexose	6	D-Glucose	D-Fructose	Fruit juices and cane sugar
		D-Galactose	-	Lactose, constituent of lipids
		D-Mannose	-	Plant mannosans and glycoproteins
Heptose	7	-	D-Sedoheptulose	Intermediate in carbohydrate metabolism





Derived				
Deoxysugar	5	2-Deoxyribose	-	DNA
	6	L-Rhamnose	-	Component of cell wall
Aminosugar	6	D-Glucosamine	-	A major component of polysaccharide found in insects and crustaceans (chitin)
Polyol	6	Sorbitol	-	Berries
	6	Mannitol	-	Commercially prepared from mannose and fructose
Aldonic acid	6	Gluconic acid	-	-
Uronic acid	6	Glucuronic acid	-	Constituent of chondroitin sulfate
	6	Galacturonic acid	-	Constituent of pectin
Aldaric acid (Saccharic acid)	6	Glucaric acid	-	Oxidation product of glucose
	6	Mucic acid	-	Oxidation product of galactose

GLUCOSE



GLUCOSE

This straight chain representation of the glucose molecule shows how the carbon atoms are numbered

Glucose, in common with many other hexose sugars has an aldehyde group as part of the structure

The carbon atom that forms part of this aldehyde group is always carbon 1

The C = O carbonyl group has reducing properties such that all monosaccharides are reducing sugars

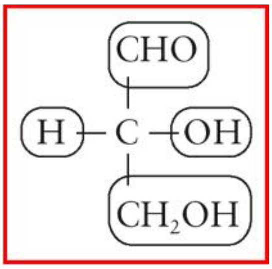
The remainder of the molecule is a series of bonded carbon atoms with attached hydrogen atoms and hydroxyl (OH) groups

ALDEHYDE GROUP

The carbon atom of the carbonyl group is referred to as the ANOMERIC CARBON ATOM and, for glucose, this is carbon 1

MONOSACCHARIDE STRUCTURES

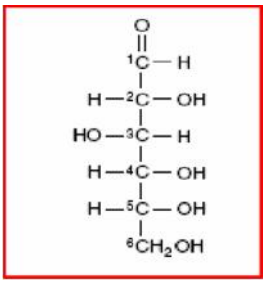
The simplest monosaccharide that possesses a hydroxyl group and a carbonyl group with an asymmetric carbon atom is the aldotriose -glyceraldehyde.(Reference carbohydrate) (A carbon is said to be asymmetric if four different groups or atoms are attached to it. The carbon is also called as a chiral center). Glyceraldehyde is considered as a reference compound and it exists in two optically active forms, D and L. A chiral object cannot be superimposed on its mirror image. A chiral carbon (Asymmetric carbon) is one that has four different groups attached to it.



1. The straight – chain (open chain) structural formula

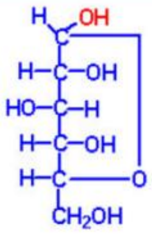
   Aldohexose- an account for some of the properties of glucose, but cannot explain some Reaction

   D-glucose



2. Cyclic structure

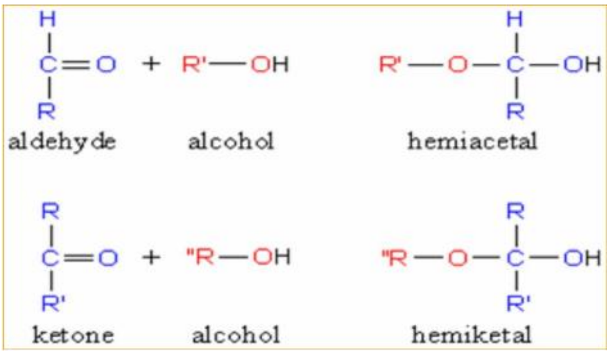
   When an aldehyde or a ketone group is present in a molecule that also possesses hydroxyl groups, an intramolecular arrangement may occur to form a hemiacetal or a hemiketal, respectively. This intramolecular hemiacetal or hemiketal is the basis for the cyclic structure of the sugars .Hence, Haworth (an English chemist) proposed a cyclic hemiacetal structure that accounts completely for its chemical properties



Cyclic Fischer Projection of □-D-Glucose

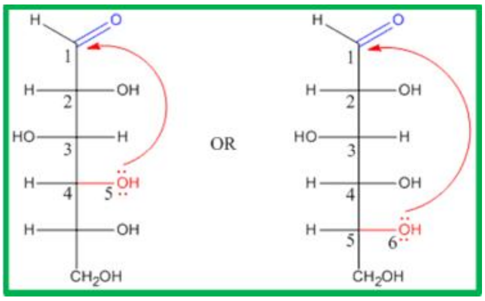
Haworth Projection of □-D-Glucose

Hemiacetals or Hemiketals



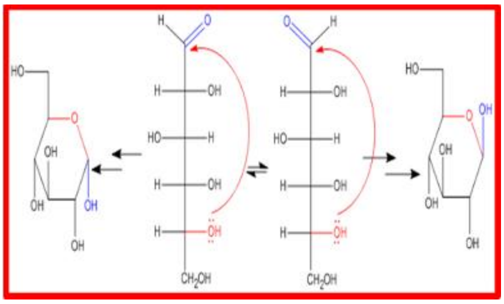
An aldehyde or ketone can react with an alcohol in a 1:1 ratio to yield a hemiacetal or hemiketal, respectively, creating a new chiral center at the carbonyl carbon

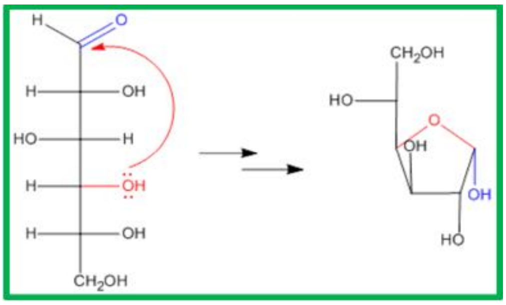
Two types of ring structures are possible, the five-membered furanose and the six membered pyranose ring if the carbonyl group interact with hydroxyl group. These names are derived from the parent compounds 'furan' and 'pyran'. The most common ring structure for aldohexoses is the pyranose ring structure that involves the first carbonyl carbon and the hydroxyl group attached to the fifth carbon. The furanose ring structure is formed by interaction of carbonyl carbon with the hydroxyl group attached to the fourth carbon. This furanose form is less stable than the pyranose structure and is not very common among Aldohexose

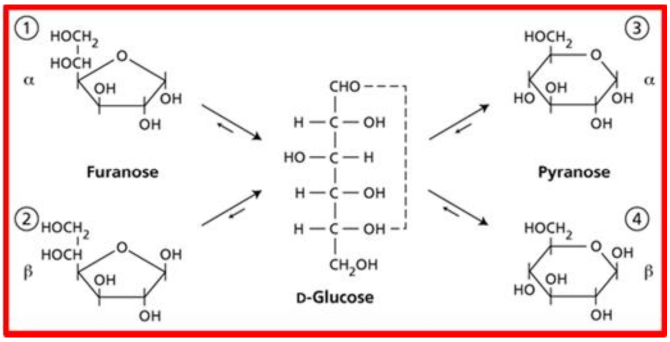


Six membered pyranose ring

Five membered furanose ring

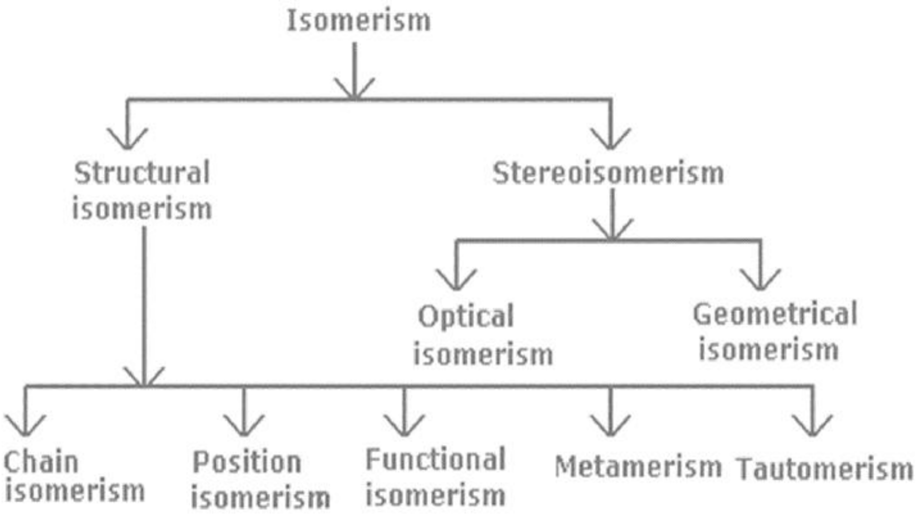






Isomerism

Isomers are different compounds that have the same molecular formula. Different compounds means that they have different physical properties (melting point, boiling point etc.). They may also have very different chemical properties depending on the type of isomerism present. It was coined by J.J. Berzelius for different compounds with same molecular formula

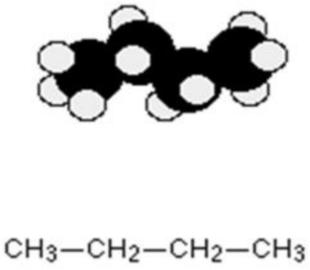


Structural isomers, in which the atoms are joined in a different order, so that they have different structural formulae

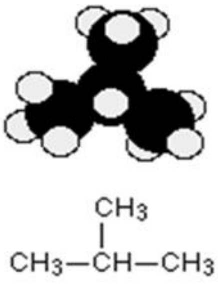
Types of structural isomerism

Chain isomerism

These isomers arise because of the possibility of branching in carbon chains. For example, there are two isomers of butane, C_4H_{10}. In one of them, the carbon atoms lie in a "straight chain" whereas in the other the chain is branched.



$CH_3-CH_2-CH_2-CH_3$



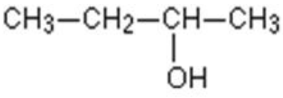
$$CH_3-\underset{}{\overset{\overset{\Large CH_3}{|}}{C}H}-CH_3$$

Position isomerism

In position isomerism, the basic carbon skeleton remains unchanged, but important groups are moved around on that skeleton.

$CH_3-CH_2-CH_2-CH_2-OH$

butan-1-ol

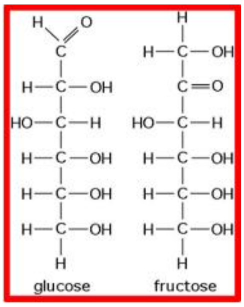


$$CH_3-CH_2-\underset{\underset{\Large OH}{|}}{C}H-CH_3$$

butan-2-ol

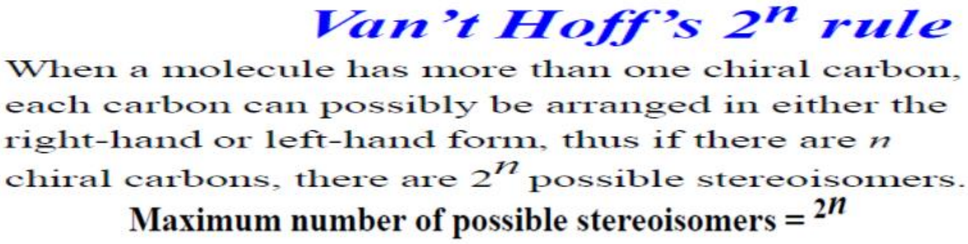
Functional Isomers

D-glucose and D-fructose differ in the position of carbonyl group (aldehyde and ketone group). These two compounds are functional isomer



Stereoisomerism or constitutional isomers

Another type of isomerism exhibited by compounds possessing asymmetric carbon atom like monosaccharides, is stereoisomerism. These stereoisomers differ in the spatial arrangement of atoms or groups. There are two types of stereoisomerisms - geometrical and optical isomerism.



Van't Hoff's 2^n rule

When a molecule has more than one chiral carbon, each carbon can possibly be arranged in either the right-hand or left-hand form, thus if there are n chiral carbons, there are 2^n possible stereoisomers.

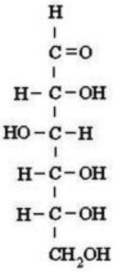
Maximum number of possible stereoisomers = 2^n

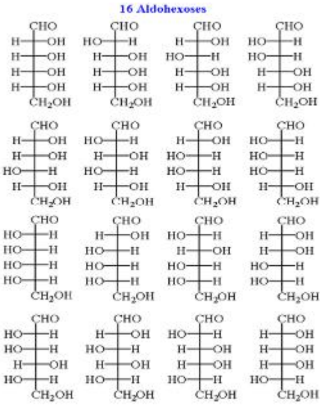
Geometric Isomerism

Geometric isomers (also called cis/trans isomers) are a type of stereoisomer resulting from a double bond or a ring structure. The double bond or ring in the structure means that not all bonds are free to rotate, giving rise to geometric isomers whose shapes cannot interconvert. Geometrical isomerism is not noticed among carbohydrates.

Optical isomerism

Optical isomers differ in the arrangement of atoms around an asymmetric carbon atom. The number of possible optical isomers can be calculated using the formula 2n where n=number of asymmetric carbon atoms. For example, glucose contains four asymmetric carbon atoms and the possible optical isomers of glucose are 24 = 16. Optical isomers are named like this because of their effect on plane polarised light.





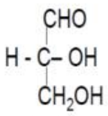
Enantiomers

Enantiomers are non- superimposable mirror images of each other. They differ in the ability to rotate the plane polarized light .A solution of one enantiomer rotates the plane of such light to the right, and a solution of the other to the left. D-glucose and L-glucose are examples of enantiomers.

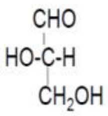
D and L isomers (Enantiomers)

Enantiomers :

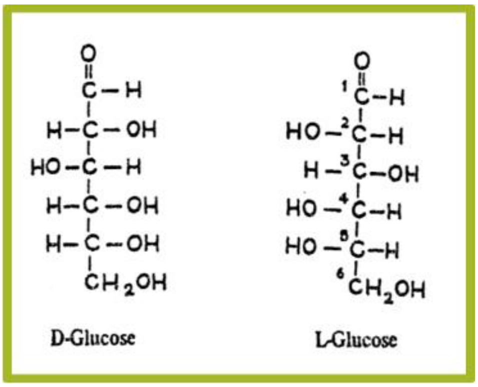
They are the mirror image of each others.

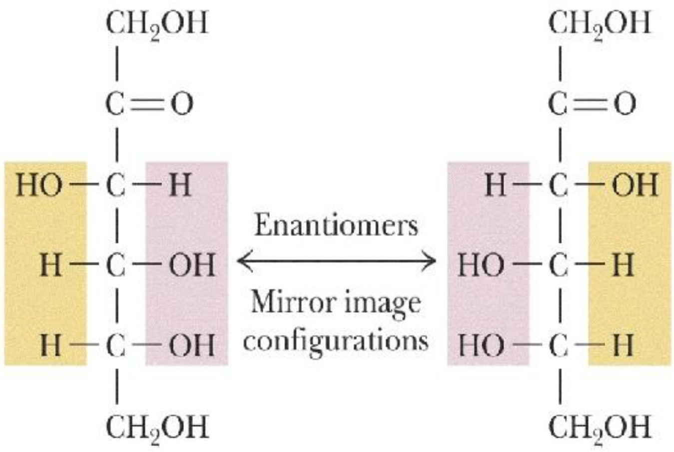


D-Glyceraldehyde



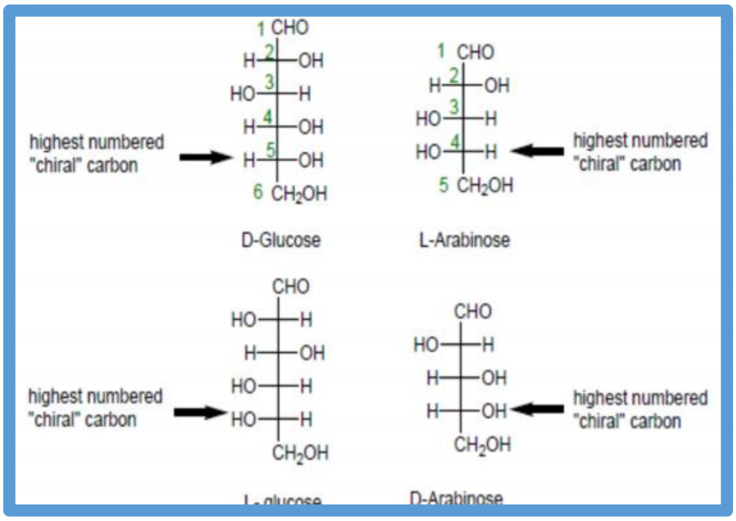
L-Glyceraldehyde





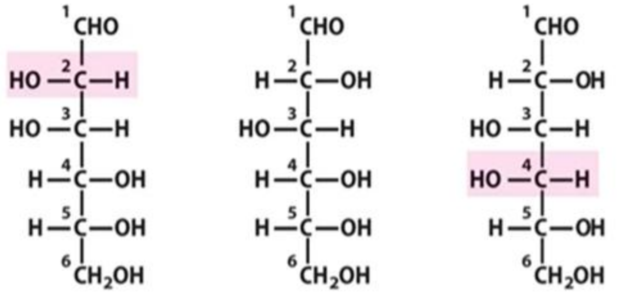
D-Fructose L-Fructose

Carbohydrates are designated as D- or L- according to the stereochemistry of the highest numbered chiral carbon of the Fischer projection. If the hydroxyl group of the **highest numbered chiral carbon** is pointing to the right, the sugar is designated as **D** (*Dextro*: Latin for *on the right side*). If the hydroxyl group is pointing to the left, the sugar is designated as **L** (*Levo*: Latin for *on the left side*). Most naturally occurring carbohydrates are of the D-configuration.



Epimers

Epimers are monosaccharides differing in configuration around a single carbon atom other than the carbonyl carbon. e.g. Mannose and glucose are epimers with respect to carbon 2. Galactose and glucose are epimers with respect to carbon 4.



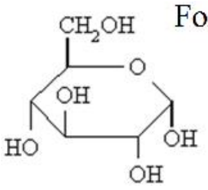
D-Mannose (epimer at C-2) D-Glucose D-Galactose (epimer at C-4)

D-Galactose is an epimer of D-glucose because the two sugars differ only in the configuration at C-4. D-Mannose is an epimer of D-glucose because the two sugars differ only in the configuration at C-2.

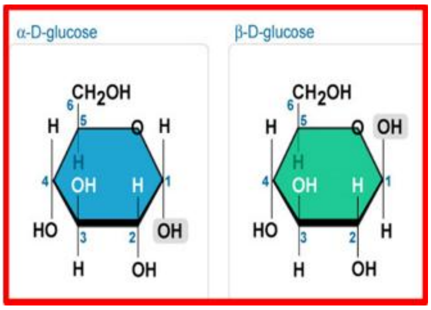
Anomers

When a molecule such as glucose converts to a cyclic form, it generates a new chiral centre at C-1.

The carbon atom that generates the new chiral centre (C-1) is called the anomeric carbon. Anomers are special cases — they are epimers that differ in configuration only at the anomeric carbon



For example, α-D-glucose and β-D-glucose are Anomers



The α form has the anomeric OH group at C-1 on the opposite side of the ring from the CH2OH group at C-5.

The β form has the anomeric OH group on the same side as the CH_2OH

Diastereomers

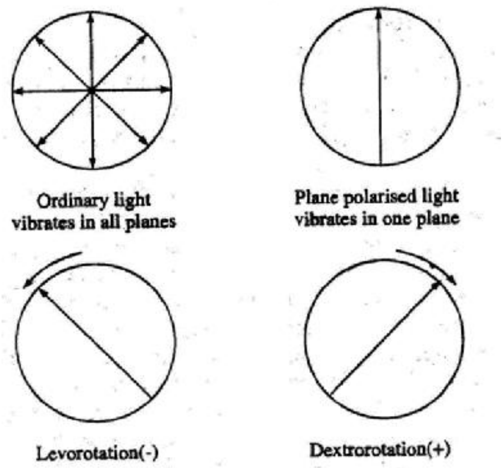
Diastereomers are stereoisomers that are not mirror images of each other. D-glucose, D-mannose, D-galactose and other members of aldohexose are diastereoisomers**.**

Stereoisomers with two or more stereocenters can be diastereomers. It is sometimes difficult to determine whether or not two molecules are diastereomers.

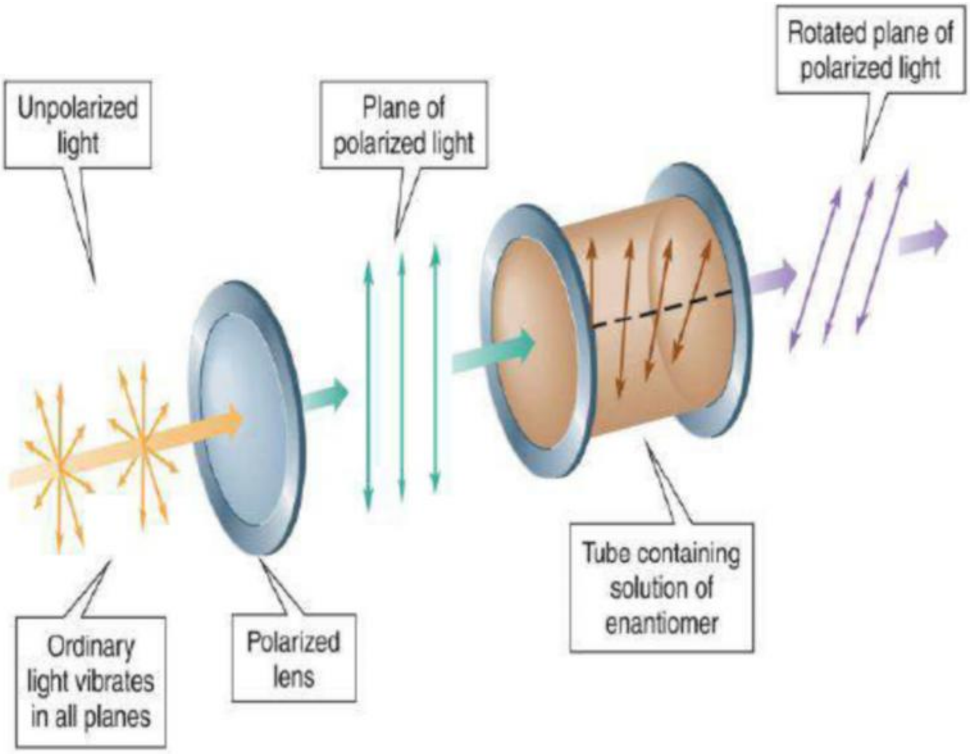
Optical activity

Molecules that are chiral can rotate the plane of polarized light. The property of rotating the plane of polarized light is called optical activity, A ray of ordinary light vibrates in all directions at right angles to the direction in which the ray is travelling. When this light is passed through a Nicol prism, the emerged light vibrates in only one direction and such light is called as a 'plane polarized light

When a beam of plane polarized light is passed through a sugar solution, that is optically active, the plane-polarized light will be rotated either to the right (clockwise) or to the left (anticlockwise). When the plane polarized light is rotated to the right, the compound is dextrorotatory and is written as (+). • If the plane polarized light is rotated to the left, the compound is levorotatory (-)



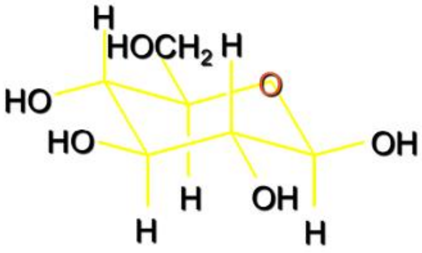
Property and rotation of plane polarised light



Mutarotation

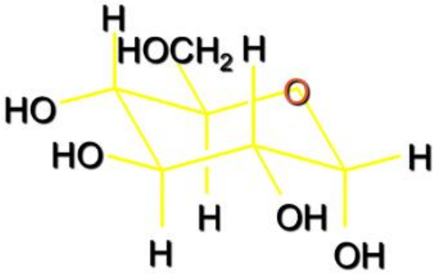
Mutarotation is a term given to the change in the observed optical rotation of a substance with time. Glucose, for example, can be obtained in either its a or b-pyranose form. The two forms have different physical properties such as melting point and optical rotation. When either form is dissolved in water, its initial rotation changes with time. Eventually both solutions have the same rotation. In Glucose solution, 2/3 of sugar exist as β form, & 1/3 as α form. Inter conversion of α & β forms is called MUTA ROTATION

Mutarotation of D-Glucose



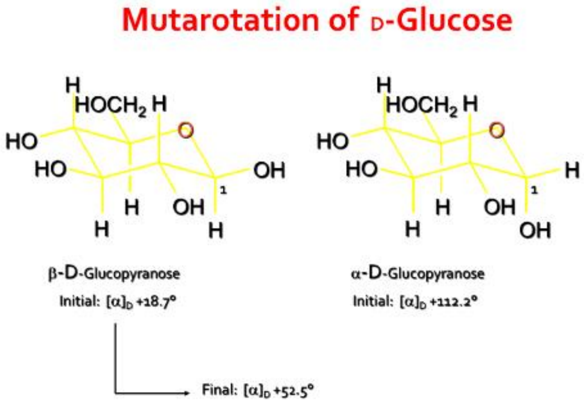
β-D-Glucopyranose

Initial: $[\alpha]_D$ +18.7°



α-D-Glucopyranose

Initial: $[\alpha]_D$ +112.2°

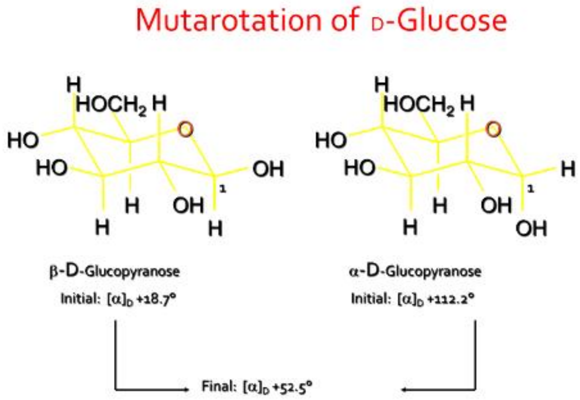


Mutarotation of D-Glucose

β-D-Glucopyranose
Initial: $[\alpha]_D$ +18.7°

α-D-Glucopyranose
Initial: $[\alpha]_D$ +112.2°

Final: $[\alpha]_D$ +52.5°

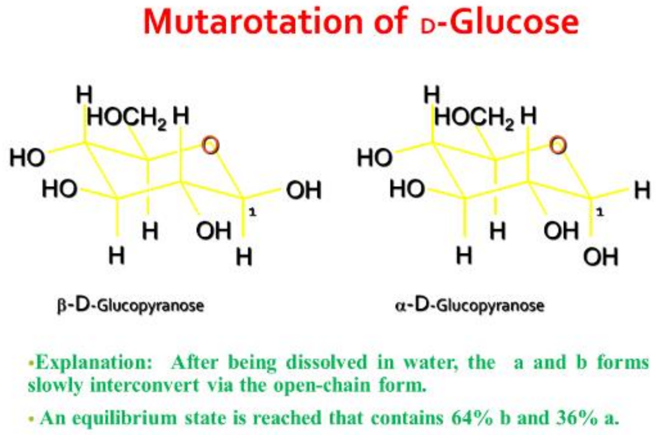


Mutarotation of D-Glucose

β-D-Glucopyranose
Initial: $[\alpha]_D$ +18.7°

α-D-Glucopyranose
Initial: $[\alpha]_D$ +112.2°

Final: $[\alpha]_D$ +52.5°



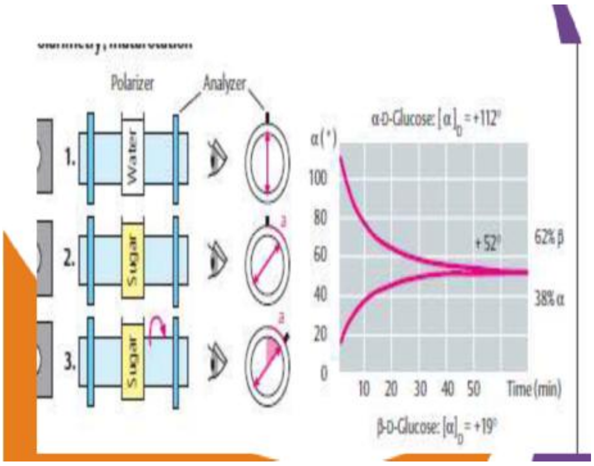
Mutarotation of D-Glucose

β-D-Glucopyranose

α-D-Glucopyranose

•Explanation: After being dissolved in water, the a and b forms slowly interconvert via the open-chain form.

• An equilibrium state is reached that contains 64% b and 36% a.



Oligosaccharides

These consist of 2 and up to 10 molecules of simple sugars and are hydrolysable. They are sub classified into di-, tri- and tetrasaccharides etc…, according to the number of molecules of simple sugars they yield on hydrolysis

Disaccharides

Disaccharides are sugars composed of two monosaccharides covalently bonded together by a glycosidic linkage. The most abundant disaccharides are sucrose, lactose and maltose. The disaccharides can be classified into homo disaccharides and hetero disaccharides

A) Homo disaccharides: are formed of the same monosaccharide units maltose, isomaltose, cellobiose and trehalose

B) Hetero disaccharides: are formed of different monosaccharide and include: sucrose, lactose

- Maltose, also known as malt sugar, is formed from two glucose molecules
- Lactose, or milk sugar, is a disaccharide formed when the monosaccharides glucose and galactose are joined
- Sucrose is common household sugar and is formed when the monosaccharides glucose and fructose bond

MALTOSE = GLUCOSE + GLUCOSE
LACTOSE = GLUCOSE + GALACTOSE
SUCROSE = GLUCOSE + FRUCTOSE

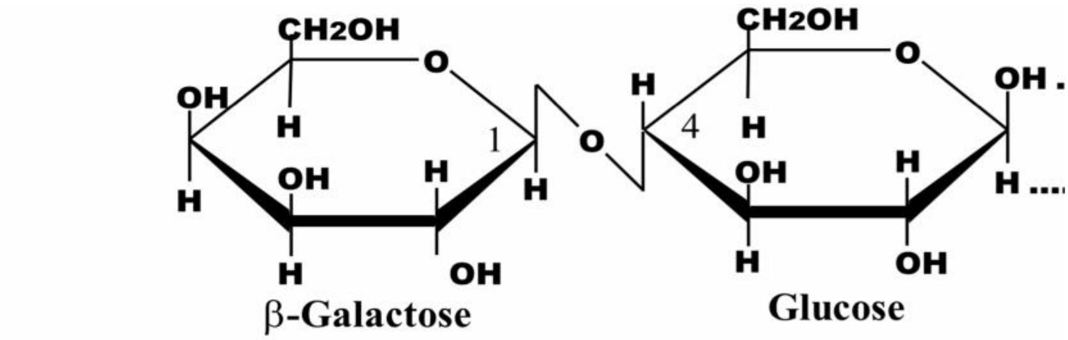
Lactose:

It is formed of β-galactose and α-glucose linked by β-1,4-glucosidic linkage

Contain free anomeric carbon so reducing sugar

Lactose is a reducing disaccharide found only in milk.

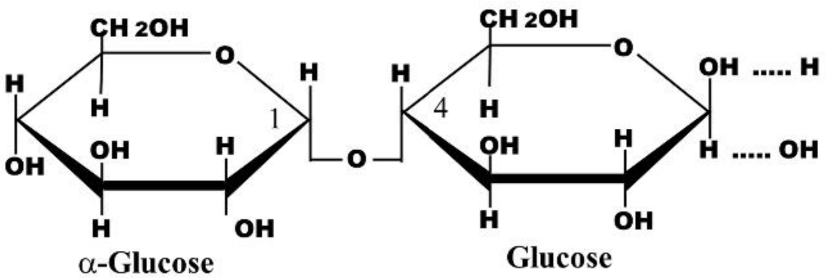
It is made up of galactose at the non-reducing end and glucose at the reducing end.



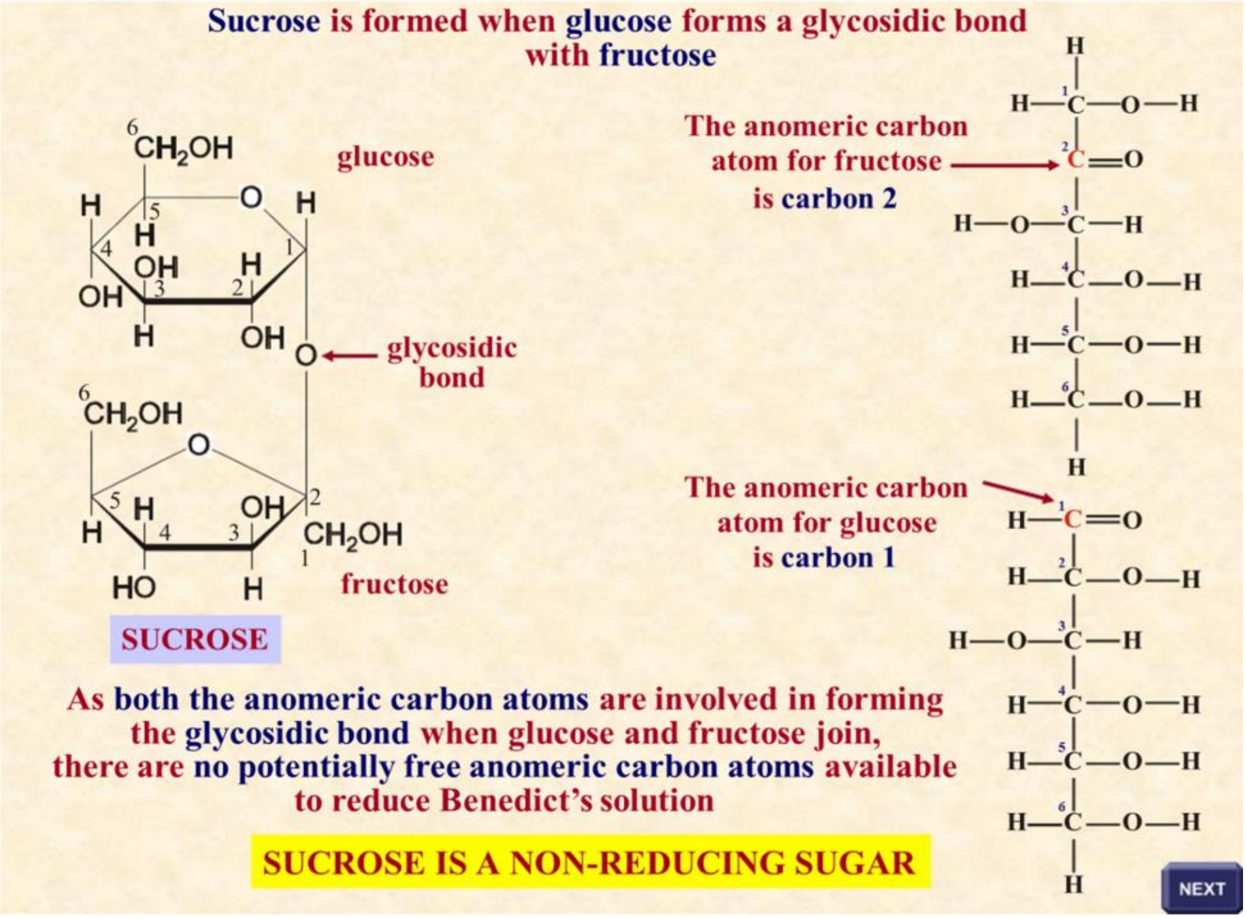
α– and β–Lactose

Maltose (malt sugar):

Maltose is a disaccharide made up of two glucose residue joined by a glycosidic linkage between C-1 of one glucose residue and C-4 of the other. The anomeric carbon atom of the second glucose is free and therefore maltose is a reducing sugar.



α– and β–Maltose



Sucrose is formed when glucose forms a glycosidic bond with fructose

The anomeric carbon atom for fructose is carbon 2

The anomeric carbon atom for glucose is carbon 1

SUCROSE

As both the anomeric carbon atoms are involved in forming the glycosidic bond when glucose and fructose join, there are no potentially free anomeric carbon atoms available to reduce Benedict's solution

SUCROSE IS A NON-REDUCING SUGAR

Sucrose, a sugar of commercial importance, is widely distributed in higher plants. Sugarcane and sugar beet are the sole commercial sources. It is made up of glucose and fructose. The anomeric carbon atom of glucose (C-1) and fructose (C-2) are involved in linkage and is therefore a non-reducing disaccharide. Sucrose is a major intermediate product of photosynthesis and it is the principal form in which sugar is transported from the leaves to other portions of plants via their vascular systems.

Invert sugar

The hydrolysis of sucrose when followed Polarimetrically the optical rotation changes from positive (dextro-) to negative (levo-). The dextrorotatory sucrose on hydrolysis yield levorotatory mixture of glucose and fructose. The levorotaion is due to the presence of fructose which is by itself more levorotatory (-92) than dextrorototary glucose (+52.2). This phenomenon is called inversion and the mixture of glucose and fructose is called invert sugar. This reaction is catalysed by the enzyme invertase. Invert sugar is sweeter than sucrose. Honey contains plenty of invert sugar and therefore is very sweet.

Polysaccharides

Polysaccharides are large polymers of the monosaccharides. Unlike monosaccharides and disaccharides, polysaccharides are either insoluble or form colloidal suspensions. The polysaccharides found in nature either serve a structural function (structural polysaccharides) or play a role as a stored form of energy (storage polysaccharides).

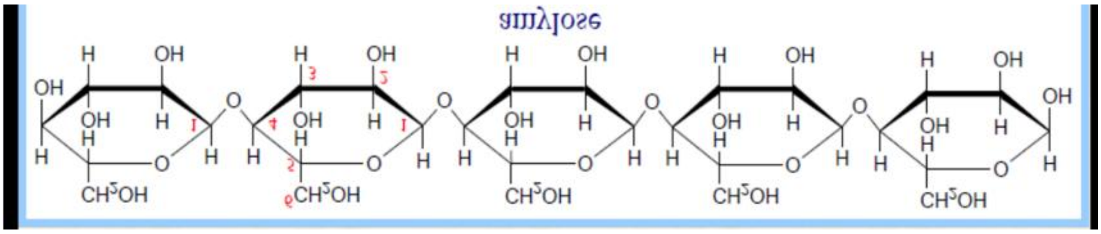
Storage polysaccharides (digestible polysaccharides)

The principal storage polysaccharides are STARCH AND GLYCOGEN. Starch is a polymer of alpha glucose and is, in fact, a mixture of two different polysaccharides – AMYLOSE AND AMYLOPECTIN

Starch

The principal food-reserve polysaccharide in the plant kingdom is starch. It forms the major source of carbohydrate in the human diet. Starch has been found in some protozoa, bacteria and algae. But the major source is plants where it occurs in the seeds, fruits, leaves, tubers and bulbs in varying amount from a few percent to over 74%. Starch is an alpha-glucan that has structurally distinct components called amylose and amylopectin. A third component referred as the intermediate fraction has also been identified in some starches. Starch molecules are organized into quasicrystalline macromolecular aggregates called granules. The shape of the granules are characteristics of the source of the starch. The two components, amylose and amylopectin, vary in amount among the different sources from less than 2% of amylose in waxy rice or waxy maize to about 80% amylose in amylomaize. The majority of starches contain 15 to 35% of amylose.

AMYLOSE



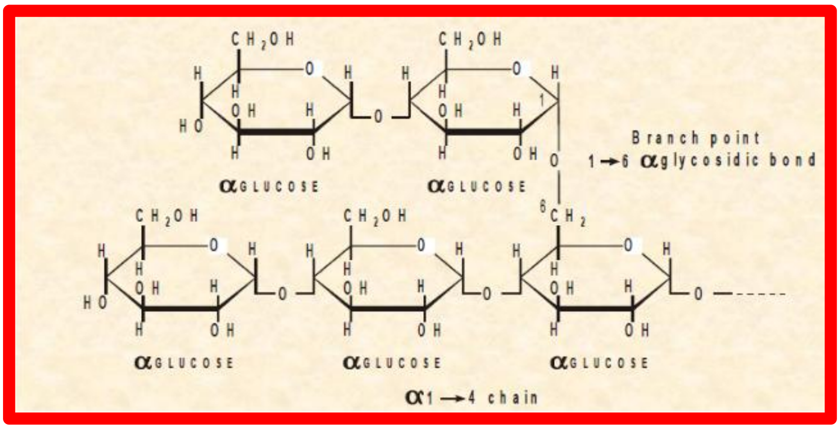
Amylose is formed by a series of condensation reactions that bond alpha glucose molecules together into a long chain forming many glycosidic bonds

Amylose is made up of α D-glucose units linked mostly in a linear way by α 1- 4 linkages

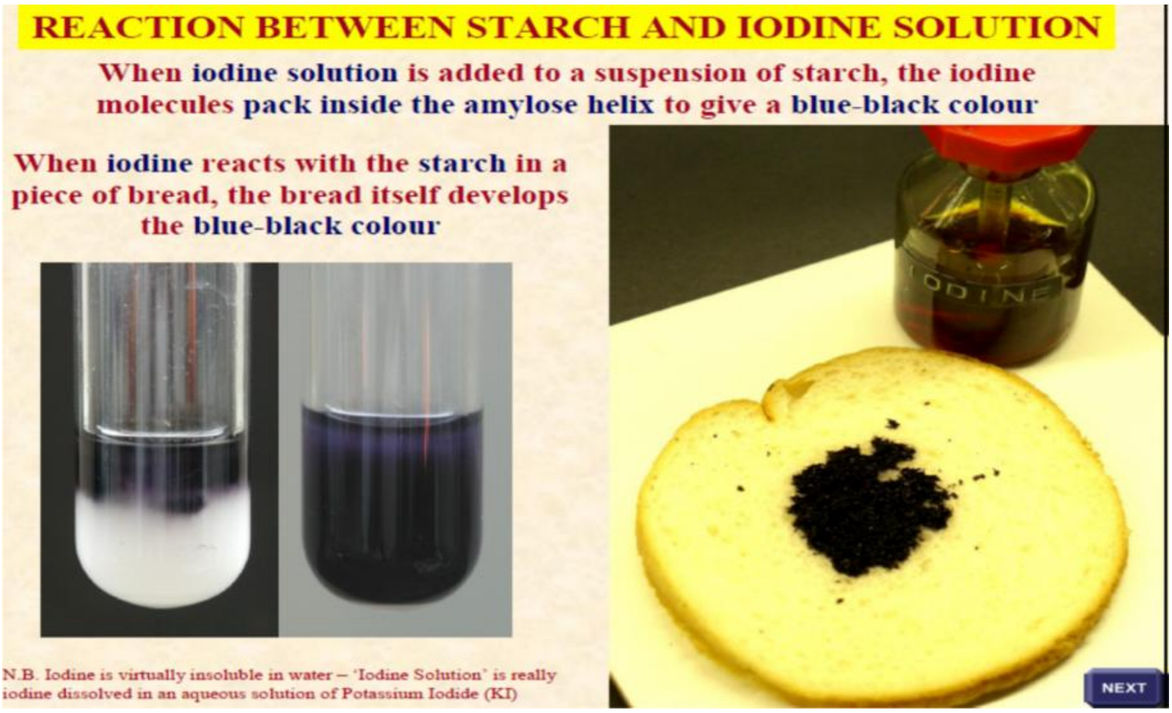
It has a molecular weight of 150,000 to 1,000,000 depending on its biological origin. It consists of a mixture of linear molecules with limited, long-chain branching involving α 1-6 Linkages. Amylose gives a characteristic blue color with iodine due to the ability of the iodine to occupy a position in the interior of a helical coil of glucose units

Amylopectin

Amylopectin consists of a straight chain of alpha glucose units with branch points occurring at approximately every twelth glucose unit along the straight chain. The branch point's form when carbon 6 of a glucose molecule in the straight chain forms a glycosidic bond with carbon 1 of a glucose molecule positioned above the chain.



This highly branched amylopectin molecule is wrapped around the amylose to make up the final starch molecule. This large insoluble molecule with branch points that allow for easy access for enzymes when breaking down the molecule, makes starch an ideal food storage compound. Amylopectin is a branched, water-insoluble polymer comprised of thousands of D-glucose residues. It contains 94-96% α 1-4 and 4-6% α 1-6 linkages. The molecular weight of amylopectin is in the order of 107 - 108. The amylopectin molecule is 100 - 150 A in diameter and 1200-4000 A long.

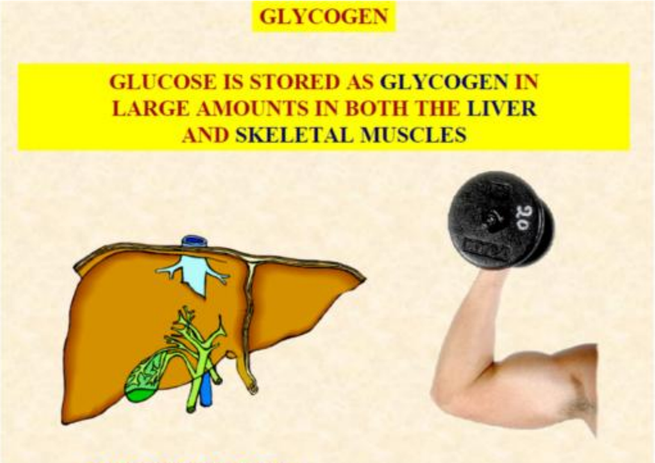


REACTION BETWEEN STARCH AND IODINE SOLUTION

When iodine solution is added to a suspension of starch, the iodine molecules pack inside the amylose helix to give a blue-black colour

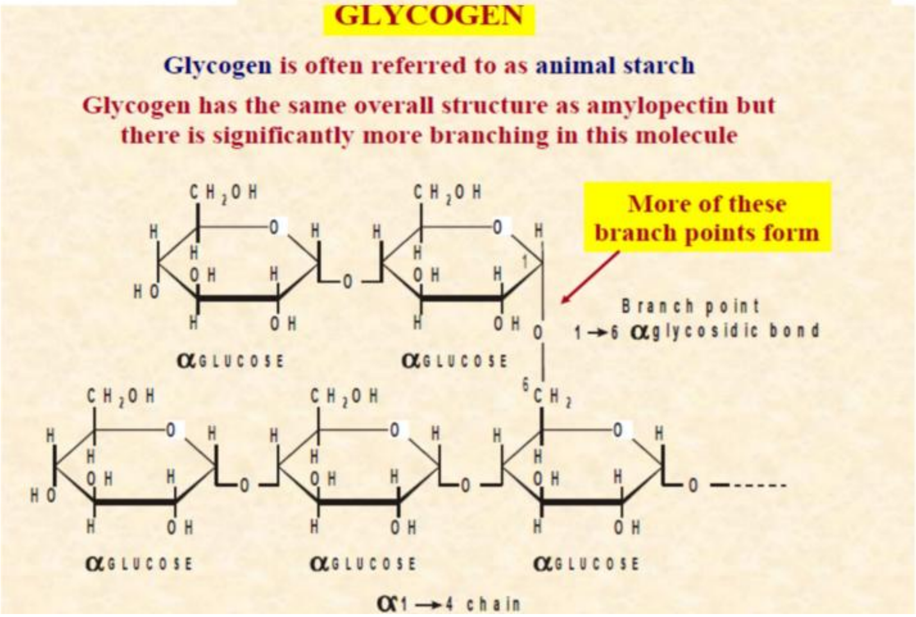
When iodine reacts with the starch in a piece of bread, the bread itself develops the blue-black colour

N.B. Iodine is virtually insoluble in water – 'Iodine Solution' is really iodine dissolved in an aqueous solution of Potassium Iodide (KI)



GLYCOGEN

GLUCOSE IS STORED AS GLYCOGEN IN LARGE AMOUNTS IN BOTH THE LIVER AND SKELETAL MUSCLES



GLYCOGEN

Glycogen is often referred to as animal starch

Glycogen has the same overall structure as amylopectin but there is significantly more branching in this molecule

More of these branch points form

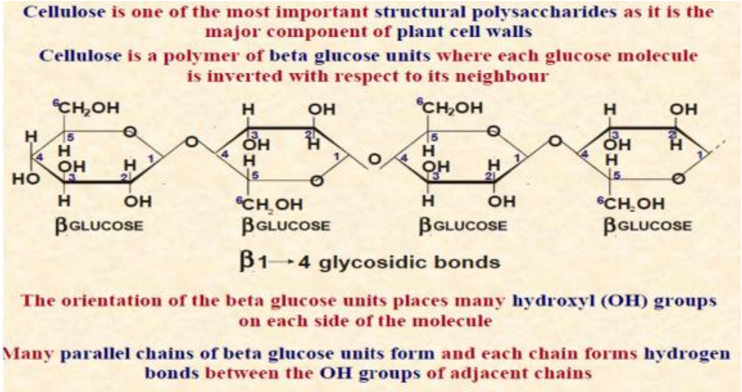
Branch point 1→6 α glycosidic bond

α1 → 4 chain

Inulin

Inulin is a non-digestible fructosyl oligosaccharide found naturally in more than 36000 types of plants. It is a storage polysaccharide found in onion, garlic, chicory, artichoke, asparagus, banana, wheat and rye. It consists of mainly, if not exclusively, of - 2->1 fructosyl-fructose links. A starting glucose moiety can be present, but is not necessary. Inulin is a soluble fibre that helps maintain normal bowel function, decreases constipation, lowers cholesterol and triglycerides. It is used for fat replacement and fibre enrichment in processed foods

STRUCTURAL POYSACCHARIDES (indigestible polysaccharides)



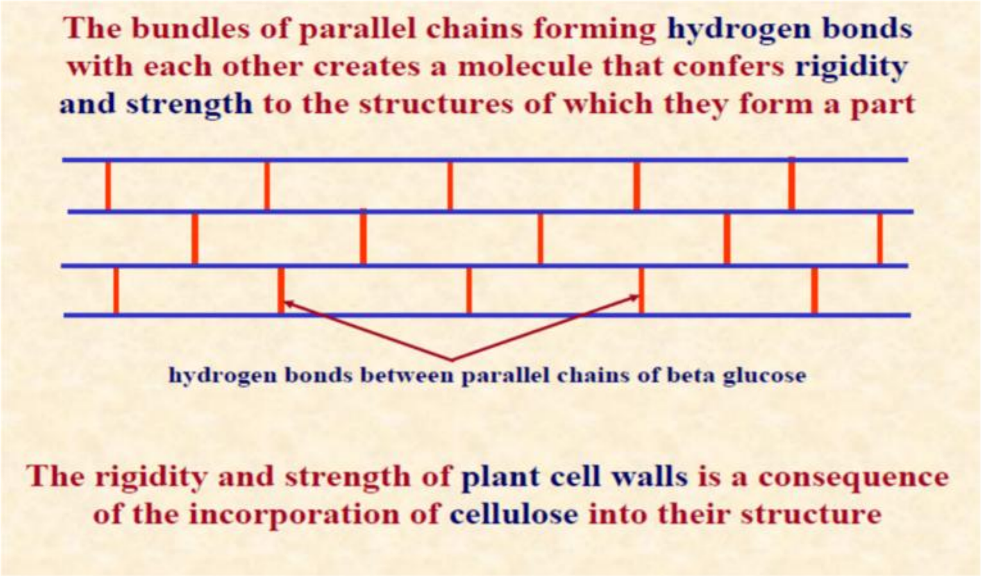
Cellulose is one of the most important structural polysaccharides as it is the major component of plant cell walls

Cellulose is a polymer of beta glucose units where each glucose molecule is inverted with respect to its neighbour

β1 → 4 glycosidic bonds

The orientation of the beta glucose units places many hydroxyl (OH) groups on each side of the molecule

Many parallel chains of beta glucose units form and each chain forms hydrogen bonds between the OH groups of adjacent chains


The bundles of parallel chains forming hydrogen bonds with each other creates a molecule that confers rigidity and strength to the structures of which they form a part

The rigidity and strength of plant cell walls is a consequence of the incorporation of cellulose into their structure

Cellulose is the most abundant organic substance found in nature. It is the principal constituent of cell walls in higher plants. It occurs in almost pure form (98%) in cotton fibres and to a lesser extent in flax (80%), jute (60-70%), wood (40-50%) and cereal straws (30-43%). It is linear, unbranched homoglycan of 10,000 to 15,000 D-glucose units joined by β 1- 4 linkages The structure of cellulose can be represented as a series of glucopyranose rings in the chair conformation.

Pectin- It is in fruits of many plants. The constituent monosaccharide is ά-D – galactouronic acid

Chitin- Most abundant in nature after cellulose, found in fungi and anthropods. The constituent monosaccharides are N- Acetyl- D- glucosamine. It is non- reducing sugar

Properties of carbohydrate

Monosaccharides

1. **Reaction with alcohol** – The Glycosidic OH group of mutarotating sugars reacts with alcohol to form ά and β glycosides or acetals. Thus glucose form glucosides and fructose from fructosides
2. **Reaction with acetic anhydride-** The Glycosidic and alcoholic OH group of monosaccharides and disaccharides react with acetylating agents to form acetate derivatives called esters
3. **Oxidation with acids**- Only the aldehydes group of sugar is oxidized to produce monocarboxylic acid with bromine water, while with nitric acid both aldoses and ketoses react to form dicarboxylic acid
4. **Oxidation with metal hydroxides**- Metal hydroxides like copper hydroxide oxidize free aldehydes or keto group of mutarotating monosaccharides and at the same time they themselves reduce to free metal

$$\text{Reducing sugar} + Cu^{2+} \longrightarrow \text{oxidized sugar} + 2\,Cu^{+}$$

$$2Cu^{+} + 2OH^{-} \longrightarrow \underset{\text{Yellow}}{2Cu.OH} \longrightarrow \underset{\text{Red}}{Cu_2O + H_2O}$$

This sugar is active ingredient in Fehling’s, Benedict’s and Barfoed’s reagent

5. **Reduction**- The sugars undergo reduction with sodium amalagam to form corresponding alcohols. Glucose yields sorbitol and fructose yields mixture of sorbitol and mannitol. With strong acids, it undergo reduction to form levulinic acid.

6. **Reaction with hydrogen cyanide**- (Kilani synthesis). It forms cyanohydrins

7. Reaction with **alanine**– The aldehydes group of carbohydrate condenses with the amino group of alanine to form Schiff's base.

8. **Reaction with Phenyl hydrazine –** reaction of monosaccharides with phenyl hydrazine yields osazone

9. **Fermentation**- Monosaccharides are readily fermented by yeast to form alcohol

Disaccharides

1. Sucrose- it is dextrorotatory, a non- reducing sugar, does not exhibit mutarotation, does not form osazone and fermentable
2. Lactose- It is dextrorotatory, a reducing sugar, exhibit mutarotation, form osazone and non-fermentable
3. Maltose- It is dextrorotatory, a reducing sugar, exhibit mutarotation, form osazone and fermentable
4. Cellobiose- It is a reducing sugar, exhibit mutarotation

Polysaccharides

1. Starch- It is made up of amylose and amylopectin. Amylose gives blue colour with iodine, amylopectin gives purple with iodine and dextrorotatory,
2. Glycogen- It is a non-reducing sugar, gives red colour with iodine, white powder, fairly stable in hot alkali
3. Cellulose- It gives no colour with iodine, fibrous, tough, white solid
4. Inulin- It is a non-reducing sugar, gives no colour with iodine

5) Chitin- It is non-reducing sugar

Physical Properties of Carbohydrates

a) Monosaccharides

1) Crystaline compound
2) Sweet to taste
3) Soluble in water
4) Need to be digested before absorbed in blood stream

b) Disaccharides

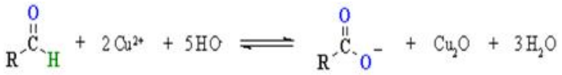
1) Crystaline compound
2) Sweet to taste
3) Soluble in water
4) Need to be digested before absorbed in blood stream

c) **Polysaccharides**

1) Amorphous compound
2) Not soluble in water
3) No sweet taste
4) Form colloidal suspension and need to be digested before absorbed

Reducing property of sugars

Sugars are classified as either reducing or non-reducing depending upon the presence of potentially free aldehyde or keto groups. The reducing property is mainly due to the ability of these sugars to reduce metal ions such as copper or silver to form insoluble cuprous oxide, under alkaline condition



Any carbohydrate which is capable of being oxidized and causes the reduction of other substances without having to be hydrolyzed first is known as reducing sugar. The aldehyde group of aldoses is oxidized to carboxylic acid. This reducing property is the basis for qualitative (Fehling's, Benedict's, Barfoed's tests) and quantitative reactions. All monosaccharides are reducing. In the case of oligosaccharides, if the molecule possesses a free aldehyde or ketone group it belongs to reducing sugar (maltose and lactose). The carbohydrates which are unable to be oxidized and do not reduce other substances are known as non- reducing sugars. If the reducing groups are involved in the formation of glycosodic linkage. the sugar belongs to the non- reducing group (trehalose, sucrose, raffinose and stachyose). The reason that sucrose is a non-reducing sugar is that it has no free aldehyde or keto group. Additionally, its anomeric carbon is not free and can't easily open up its structure to react with other molecules.

Reaction with acids

Monosaccharides are generally stable to hot dilute mineral acids though ketoses are appreciably decomposed by prolonged action. Heating a solution of hexoses in a strong non-oxidising acidic conditions, hydroxyl methyl furfural is formed. The hydroxymethyl furfural from hexose is usually oxidized further to other products .When phenolic compounds such as resorcinol, naphthol or anthrone are added, mixture of coloured compounds are formed. The Molisch test used for detecting carbohydrate in solution is based on this principle. When conc.H_2SO_4 is added slowly to a carbohydrate solution containing naphthol, a pink color is produced at the juncture

Reaction with alkali

Dilute alkali

Sugars in weak alkaline solutions undergo isomerization to form 1,2-enediols followed by the formation of a mixture of sugars**.**

Strong alkali

Under strong alkaline conditions sugar undergo caramelization reactions

Monosaccharides (Simple sugars)	**Oligosaccharides**	**Polysaccharides (Glycans)**
Low molecular weight carbohydrates and cannot be hydrolysed further	Contain 2-10 monosaccharides joined by glycosidic bonds. Low molecular weight carbohydrates which can be hydrolysed by enzymes or acids to yield monosaccharides	Contain many monosaccharides joined by glycosidic bonds. They can be hydrolysed by enzymes or acids.
Crystalline, soluble in water, and sweet in taste.	Powdery or crystalline, soluble in water and sweet in taste	Insoluble in water, tasteless, linear or branched
Classified into triose, tetrose, pentose, hexose and heptose depending upon the number of carbon atoms. They may be either aldoses or ketoses depending upon whether they contain a free aldehyde or ketone group, respectively	Classified into disaccharide, trisaccharide, tetrasaccharide and pentasaccharide depending upon the number of monosaccharides they contain.	Classified into homoglycans and heteroglycans depending upon the kind of monosaccharides present. Depending upon the function, they are classified as storage and structural polysaccharides.

CARBOHYDRATE

FILL UP THE BLANKS

1. Starch is classified under ________
2. Isomerism is a common property due to the presence of _______ and structural arrangement
3. Cellulose is a polysaccharide made up of glucose and the linkages are ____
4. Plant gums are ___
5. Starch is a polymer of ______
6. Amylopectin has branched chained ______ units
7. ___ is the sugar present in milk
8. The storage polysaccharides of animal tissue is ________
9. Isomerism and optical activity property of carbohydrate is due to ______
10. _____ is a non-reducing sugar
11. The change in the optical activity of sugar solution is known as _____
12. The carbon atom to which four different atoms or groups are attached is called _____ carbon atom
13. The plant gums are the Heteropolysaccharides containing several units of ______
14. Gluconic acid is a _____ acid
15. The group that confers sweet taste to glucose is ______
16. Freshly prepared glucose solution has a specific rotation of _____
17. The sugar used for silvering the mirror is ______
18. Fructose is the hydrolytic product of ________
19. _______ is a non-reducing disaccharide sugar
20. The reagent used to form osazone with sugars is __
21. Carbohydrates consists of ______ elements in the ratio 1:2:1
22. The general formula of monosaccharide is _____
23. Galactose is ___ type of monosaccharide
24. Oligosaccharides on hydrolysis give _____ number of monosaccharides
25. The monosaccharide glucose has got __ functional group
26. The monosaccharide having keto group is known as _____

27 The isomers having similar molecular formula but different structural formula is called as ___

28 Stereoisomers are grouped into ______ and _________

29 Number of optical isomers of the compound depends on number of ________

30 In a carbohydrate, If H is in the left and OH is on the right, It is designated by_____

31 Number of optical isomers present in glucose is ______

32 If the compound causes rotation of the polarized light to the left, it is said to be ______

33 In a mixture where optical activity is zero, such a mixture is called as _____

34 Mutarotation is undergone by ______ sugars only

35 Pyranose rings has got ______ type of linkage by oxygen atom

36 Hexoses are formed by hydrolysis of _______

37 Glucose on reaction with bromine water yields _____

38 Fructose undergo reduction in the presence of H catalyst to form ________

39 Reaction between glucose and alanine results in the formation of ______

40 Monosaccharide on reduction with phenyl hydrazine results in the formation of ____

41 Glucose is converted to ethyl alcohol on fermentation by the action of _____

42 Any carbohydrate which is capable of being oxidized and causes the reduction of other substances is called as _______

43 Test for identifying the reducing property of sugar is found by________

44 ____ is an example of reducing disaccharide sugar

45 Sucrose on hydrolysis yields ____ and _____

46 ____ enzyme is involved in the hydrolysis of sucrose

47 Glucose and glucose combination results in the formation of ______

48 Lactose is composed of one molecules of ____ and ___

49 Lactose is also called as _______

50 Amylose and Amylopectin are components of _____-

51 Glycogen is a _____

52 Starch on reaction with iodine gives_____ colour

53 Reducing centre in aldose sugar lies at ______

54 Reducing sugar in ketose sugar lies at _______

55 Fruit sugar is chemically called as _______

56 _____ is an example of homopolysachharides

57 Glucose carries ______ asymmetric carbon atoms

58 Sucrose has specific rotation of _____

59 Carbohydrates are polyhydroxy ______-

60 Cellulose is a polysaccharide made of glucose and the linkages are _______

61 Hydrolysis of sucrose by the invertase yields ___ and ____

62 Compounds possessing a free aldehydes or ketone group and two or more hydroxyl groups are ______

63 The shift or change in the optical activity of a particular carbohydrate is known as ____

64 Inulin is polymer of ______ occurring in composite plant

65 Monosaccharides having six carbon atoms are called as ___

66 Compound containing ά 1-6 linkage with 2000 to 3000 glucose units is called as ___

67 General formula for carbohydrate is ______

68 An example of triose is ___

69 The cane sugar is made up of ____- and ______

70 ____is an aldopentose while _____ is a ketohexose sugar

71 The milk sugar______ is made up of glucose and _____

72 Invert sugar is also called as _______

73 Glucose and galactose are the constituents of _______

74 Starch is made up of ____ sub units

75 ___is an optically inactive triose

76 The ability of sugars to form esters indicate the presence of ____group in their molecule

77 _______ reaction is used to distinguish between aldose and ketoses
78 Fructose on reduction with sodium amalgam yields mixture of ______
79 ____ is an isomer of mannose
80 Molisch test with sugar develops ___ colour
81 Sucrose is a________ sugar
82 Isomerit compounds have _________ molecular formula
83 Starch contains __________ units
84 During germination starch is converted to ________
85 Maltose is a disaccharide formed by the linkage of ___________
86 Pentosans have the molecular formula ______
87 Starch on hydrolysis yields _______
88 Fructose is a______ sugar (other than fruit)
89 Optical isomerism is caused by________
90 Non- reducing disaccharides is________
91 Cellulose is a polysaccharide containing a _________
92 Aldose containing the group _______
93 The number of molecules of water eliminated when the two molecules of monosaccharide react is______
94 Disaccharide that is formed by the union of two of the same monosaccharide is_____
95 The fraction of starch that give blue colour with iodine is _______
96 The sugar units derived on the hydrolysis of starch is_______
97 Ribose is a._______ sugar
98 The animal sugar is called as _________
99 Amylopectin content in starch is_________
100 The structural polysaccharide is_______
101 Reducing lactose sugar has got ___ type linkage
102 Sucrose is commercially obtained from_______
103 Invertase is an enzyme which hydrolyses________
104 Xylose is a carbohydrate occurs in______
105 The shift or change in the optical activity of particular carbohydrates is called as_____
106 Levulose is commonly named as_________
107 Dextrin is one of the hydrolysed products of ______
108 Fructose is a ________
109 An example of homopolysachharides is _______
110 Amylopectin contains _________
111 Glycosidic bond is formed between ________
112 ______ Biomolecules simply refers as “Staff of life”
113 In carbohydrates a special functional groups present ______ and _____
114 Simplest carbohydrate is _______
115 Examples of Epimers is _______
116 _________ will answer Molisch test
117 The red precipitate formed when glucose is heated with “Benedict’s reagent” is…

118 _______ Sugar exhibits inversion of optical rotation on heating with dilute acid ….
119 The end product of hydrolysis of “Starch” by amylase is …
120 Example for “Fructosan” is ……..
121 The reagent used for distinguishing a reducing monosaccharide from a reducing disaccharide is …
122 Fructose and Glucose can be distinguished by ….
123 Concept of tetrahedral carbon atom” are first introduces by …
124 The general formula for carbohydrate is _________
125 _______ is an example of aldotriose
126 The glycosidic linkage between glucose in maltose is _____
127 A ketose pentose will have _______ stereoisomers
128 ____________ is an example epimers
129 Hydrolysis of glycoside bond involves _________of glycosidic bonds
130 Most complex also most abundant carbohydrates that are found in nature are ________
131 The most common monomer of carbohydrate is _______
132 Carbohydrates occur naturally in _____ form
133 Different ways to represent a carbohydrate molecule are straight chain, Fischer and _______
134 _________ representation of a carbohydrate can a hemiketal or hemiacetal be observed
135 Cell wall in plants is made of _________ carbohydrate
136 _________ test is used for testing reducing sugar
137 D & L Designation can be used to _______ molecules
138 A straight chain hexose sugar forms ____ type of ring
139 Casein contained in milk is a/an _________
140 Complete hydrolysis of cellulose gives _______
141 The change in optical rotation of freshly prepared solution of glucose is known as ____
142 _____ compounds, when heated at 483k turns to caramel
143 Methyl α-D glucoside and methyl –β-D glucoside are________
144 Number of chiral carbon atoms in β-D- (+) -glucose is _______
145 _______ monosaccharides is present as five membered cyclic structure (furanose structure)
146 A polymer of fructose is ______
147 _________ test is undertaken to differentiate between Glucose and Fructose
148 A 0.5 mol dm3 solution of sucrose was heated at 80 °C for 5 min with Benedict’s reagent. The resulting colour is _____
149 The relationship between glucose, mannose and galactose is ________
150 All carbohydrate foods are composed of the __________ elements

Answers

Sl.No	Answer	Sl.No	Answer
1	Storage polysaccharide	34	Reducing
2	Asymmetric carbon atom	35	C1-C5
3	1-4 Glycosidic linkage	36	Cane sugar
4	Natural polysaccharide	37	Gluconic acid
5	Glucose	38	Sorbitol and Mannitol
6	1-6 Glycosidic linkage	39	Schiff's base
7	Lactose	40	Osazone
8	Glycogen	41	Yeasts
9	Asymmetric carbon atom	42	Reducing sugar
10	Sucrose	43	Fehling's solution
11	Mutarotation)	44	Lactose/maltose
12	Asymmetric	45	Glucose and fructose
13	Glucose	46	Invertase
14	Monobasic acid	47	Maltose
15	Aldehyde	48	Glucose and Galactose
16	+ 112.2	49	Milk sugar
17	Glucose	50	Starch
18	Sucrose	51	Non-reducing sugar
19	Sucrose	52	Blue
20	Phenyl Hydrazine	53	Carbon No.1
21	C, H, O	54	Carbon No 2
22	$C_nH_{2n}O_n$	55	Fructose
23	Hexoses	56	Starch and Cellulose
24	2 to 10	57	4
25	Aldehydes	58	$+ 66.5^0$
26	Ketoses/ Fructose	59	Aldehydes or Ketones
27	Structural isomers	60	β- 1-4 Glycosidic linkage
28	Geometrical, optical	61	Glucose and Fructose
29	Asymmetric carbon atom	62	Monosaccharide
30	D- form	63	Mutarotation
31	16	64	Inulin
32	Levorotatory	65	Hexoses
33	Racemic	66	Amylopectin
Sl.No	Answer	Sl.No	Answer
67	$(CH_2O)_n$	100	Pectin
68	Glyceraldehyde's	101	C1-C4
69	Glucose and Fructose	102	Sugarcane
70	Ribose, Fructose	103	Sucrose
71	Lactose and Galactose	104	Flower of plants

72	Sucrose	105	Mutarotation
73	Lactose	106	Fructose
74	2, Amylose and Amylopectin	107	Sucrose
75	Glyceraldehydes	108	Ketohexoses
76	Alcohol	109	Cellulose
77	Mild oxidant- HOBr	110	ά – 1-4 and ά-1-6 linkage
78	Sorbitol and Mannitol	111	Two monosaccharides
79	D- Glucose	112	Carbohydrate
80	Violet	113	CHO and C=O
81	Non- reducing sugar	114	Glyceraldehyde
82	same	115	D glucose and D- mannose D Glucose and D galactose
83	Glucose + Glucose	116	Carbohydrate
84	Maltose	117	Cuprous oxide
85	Glucose + Glucose	118	sucrose
86	(C5H8O4)n	119	Maltose
87	Dextrin	120	Inulin
88	Levose sugar	121	Barfoed’s
89	Asymmetric carbon atom	122	Seliwanoff
90	Sucrose	123	Kekulé (1862)
91	Linear chain of beta D glucose units	124	$C_m(H_2O)_n$
92	CHO	125	glyceraldehyde,
93	One	126	α(1→4)
94	Maltose	127	Eight
95	Amylose	128	Glucose and mannose
96	ά D glucose	129	breakdown
97	Pentose	130	Starch
98	Glycogen	131	glucose
99	80-85%	132	D
Sl.No	Answer	Sl.No	Answer
133	Haworth	138	Pyranose
134	Ring	139	lactose
135	Cellulose	140	D-glucose units.
136	Fehling	141	Mutarotation
137	chiral	142	Glucose
143	Anomers	147	Seliwanoff
144	5	148	Cherry red
145	Glucose	149	Epimers
146	Inulin	150	C H O

DEFINITION

1. **Carbohydrate:** Carbohydrates are chemically defined as polyhydroxy aldehydes or ketones, their derivatives and their polymers. (or) Aldehyde or Ketone derivatives of polyhydroxy alcohols
2. **Monosaccharides :** They are the simplest carbohydrate unites which cannot be hydrolysed to a simpler form
3. **Oligosaccharides:** They contain two to ten monosaccharide units joined by glycosidic linkages that can be easily hydrolysed**.**
4. **Polysaccharides:** They are high molecular weight polymers containing more than ten monosaccharides.
5. **Chiral or Asymmetric carbon**: A chiral carbon (Asymmetric carbon) is one that has four different groups attached to it.
6. **Isomers:** Isomers are different compounds that have the same molecular formula
7. **Structural isomers: T**he atoms are joined in a different order, so that they have different structural formulae
8. **Functional Isomers: D**iffer in the position of carbonyl group D-glucose and D-fructose (aldehyde and ketone group). These two compounds are functional isomer
9. **Stereoisomerism or constitutional isomers:** These stereoisomers differ in the spatial arrangement of atoms or groups
10. **Geometric Isomerism:** Geometric isomers (also called cis/trans isomers) are a type of stereoisomer resulting from a double bond or a ring structure
11. **Optical isomerism:** Optical isomers differ in the arrangement of atoms around an asymmetric carbon atom**.**
12. **Enantiomers:** Enantiomers are non- superimposable mirror images of each other. D-glucose and L-glucose are examples of enantiomers.
13. **Epimers:** Epimers are monosaccharides differing in configuration around a single carbon atom other than the carbonyl carbon**.** Mannose and glucose are epimers with respect to carbon 2. Galactose and glucose are epimers with respect to carbon 4
14. **Anomers:** When a molecule such as glucose converts to a cyclic form, it generates a new chiral centre at C-1. The carbon atom that generates the new chiral centre (C-1) is called the anomeric carbon
15. **Homopolysachharides: W**hen made from a single kind of monosaccharide. Eg starch, cellulose, inulin, glycogen, chitin
16. **Heteropolysaccharides: They** are made up of more than one type of monosaccharides. Eg. Hemicellulose
17. **Diastereomers:** Diastereomers are stereoisomers that are not mirror images of each other. D-glucose, D-mannose, D-galactose
18. **Optical activity:** Molecules that are chiral can rotate the plane of polarized light. The property of rotating the plane of polarized light is called optical activity.
19. **Mutarotation:** Mutarotation is the change in the optical rotation because of the change in the equilibrium between two anomers, when the corresponding stereocenters interconvert. Inter conversion of α & β forms is called MUTA ROTATION

20. **Maltose:** Maltose, also known as malt sugar, is formed from two glucose molecules
21 **Lactose** : It is formed of b-galactose and a-glucose linked by b-1,4-glucosidic linkage
22 **Sucrose**: Sucrose is formed when glucose forms a glycosidic bond with fructose
23 **Invert sugar:** The dextrorotatory sucrose on hydrolysis yield levorotatory mixture of glucose and fructose. This phenomenon is called inversion and the mixture of glucose and fructose is called invert sugar
24 **Starch:** Starch is a polymer of alpha glucose and is, in fact, a mixture of two different polysaccharides – Amylose and Amylopectin
25 **Amylose:** Amylose is formed by a series of condensation reactions that bond alpha glucose molecules together into a long chain forming many glycosidic bonds
26 **Amylopectin**: Amylopectin consists of a straight chain of alpha glucose units with branch points occurring at approximately every twelth glucose unit along the straight chain
27 **Cellulose**: It is a polymer of beta glucose units where each glucose molecule is inverted with respect to its neighbour
28 **Reducing sugar**: A reducing sugar is any sugar that is capable of acting as a reducing agent because it has a free aldehyde group or a free ketone group
29 **Non-Reducing Sugar**: The carbohydrates which are unable to be oxidized and do not reduce other substances are known as non- reducing sugars.

Write short notes on:

1) Cellulose:

It is structural polysaccharide. It is a long straight chain polysaccharide consisting of many β – D glucose units joined together by (1, 4) Glycosidic linkages. Because it consists of only of glucose units, it is known as glucosans or glucans. It occurs in the cell wall of plants and molecular weight ranges from 200,000 to 2, 000,000. It is fibrous, tough, white solid, insoluble in water, gives no colour with iodine, lack sweetness and has no nutritive value

2) Starch

It is reserve food materials of higher plants and is found in cereals, potatoes etc. It consists of two components namely amylose and Amylopectin. Amylose is a long unbranched chain (ά -1-4 linkage) with 15-20% content and Amylopectin branched chain It is composed of linearly linked alpha 1,4 linked glucose units (coiled into tubular sections) with occasional alpha 1-6 Glycosidic bonds which provide branching points. Each amylopectin molecule may contain 100,000-200,000 glucose units, and each branch is about 20 or 30 glucose units in length, so that these molecules are bushy and nearly spherical in shape with 80-85% content in starch. Hydrolysis of starch leads to formation of ά – D glucose units by diastase enzyme. Synthesis of starch involves simultaneous synthesis of amylose and amylopectin. Amylose are formed by elimination of water from Glycosidic OH group of one ά- D glucose molecule and alcoholic OH group on carbon 4 of the adjacent glucose. Amylopectin in addition to ά -1-4 linkage like amylose, it has many sided chain attached to basic chain by 1, 6 glucosidic *linkage*

3) **Sucrose**

It is also called as table sugar, cane sugar, beet sugar. It is mainly found in plants. It is present in varying amounts in different parts of the plant. It is non- reducing sugar because Glycosidic bond blocks reducing group of both monosaccharides units. On hydrolysis, it yields one molecule of glucose and one molecule of fructose.

$$\underset{\text{Sucrose}}{C_{12}H_{22}O_{11}} + H_2O \longrightarrow \underset{\text{Glucose}}{C_6H_{12}O_6} + \underset{\text{Fructose}}{C_6H_{12}O_6}$$

Hydrolysis is done by enzyme invertase or dilute acids. The glucose and fructose are produced with a in the optical rotation from positive to negative which is called as inversion.

$$\underset{+\,66.5}{C_{12}H_{22}O_{11} + H_2O} \longrightarrow \underset{+\,52.5}{C_6H_{12}O_6} + \underset{-\,92.0}{C_6H_{12}O_6}$$

Sucrose is fermented with yeast, soluble in water, slightly soluble in alcohol, ether. It is colourless. Invert sugar is sweeter than sucrose itself.

4.) **Isomerism**

It was coined by J.J. Berzelius for different compounds with same molecular formula. Isomer is formed only in the presence of asymmetric carbon atom. It is classified into structural and Stereoisomers. Structural isomerism (Similar molecular formula with different structural formula) has three types namely chain, functional and positional. Stereoisomers (Similar structural and molecular formula but different spatial arrangement.) have two types namely geometrical and optical. Number of optical isomers in a compound depends on number of asymmetric carbon atoms. Number of optical isomers is equal to 2n where n refers to number of asymmetric carbon atoms. When light is passed through optically active solution, it can be rotated either left or right side. A compound rotating the plane of polarized light to the right is called as dextrorotatory and designated as D or (+). If the compound causes rotation of the polarized light to the left is called as levorotatory and indicated by L or (-).

5) **Monosaccharides**

These are simplest of carbohydrates and are known as sugars. These are the building blocks of complex carbohydrates. These cannot be hydrolysed. These are sweet tasting, crystalline and soluble in water. They have a potential aldehydes or keto group and hence are reducing sugars in nature. Aldehydes group are known as aldoses or aldose sugar. Monosaccharides having keto group are known as ketoses or keto sugar. Depending up on the number of carbon atoms, monosaccharides are classified into triose (Glyceraldehyde’s), tetroses (Erythroses), pentoses(ribose), Hexoses(glucose, fructose) and heptoses (Sedoheptulose). Monosaccharides exhibit gradual change in specific rotation called as Mutarotation

6) **Optical activity**

Optical activity takes place mainly due to the presence of asymmetric carbon atom. Optical isomers differ from each other in the disposition of the various atoms or groups of atoms in space around the asymmetric carbon atom. It is referred to as mirror image of each other. Number of optical isomers in any compound depends on the number of asymmetric carbon atoms. According to the rule of Le-bell Van’ts Hoff total number of optical isomers of a compound will be equal to 2^n where n refers to number of asymmetric carbon atoms When a beam of light is allowed to pass through optically active solution, it can be rotated to the right side (Dextrorotatory- D(+)) and can be rotated to the left side(Levorotatory L (-)). When equal amounts of D and L type of isomers are present, the resulting mixture is optically inactive which is called as racemic mixture. When a monosaccharide is dissolved in water, the optical rotating power of solution gradually changes until reaches a constant value which is referred to as Mutarotation.

7) **Oxidation of hexose sugar**

Monosaccharides on oxidation under proper condition form different products eg. aldoses may form monobasic acid or dibasic saccharic acid or monobasic uronic acid

A) **Production of adonic acids**: Aldoses when oxidized in the presence of bromine water, al Aldehyde group is converted into carboxyl group

$$CHO\text{-}(CHOH)_4\text{-}CH_2OH \;+\; HoBr \longrightarrow COOH\text{—}(CHOH)_4 - CH_2OH + HBr$$

Glucose Bromine water Gluconic acid

Ketoses are not readily oxidized by bromine water

B) **Saccharic acid** — The aldoses and ketoses undergo oxidation in the presence of Nitric acid to convert to carbonyl to form saccharic acid or dibasic acid. Eg. D-glucose, D-galactose and D-mannose are converted to D-glucoric acid, D-galactosaccharic acid and D-mannaric acid

$$CHO\text{-}(CHOH)_4\text{-}CH_2OH + HNO_3 \longrightarrow COOH\text{-}(CHOH)_4\text{—}COOH$$

Glucose Glucoric acid

While fructose is oxidized to tartaric acid, glycolic acid and trihydroxy glutaric acid

$$CH_2OH\text{-}C{=}O\text{-}(CHOH)_3\text{-}CH_2OH + HNO3 \longrightarrow COOH\text{-}(CHOH)_3\text{-}COOH$$

Fructose Trihydroxy glutaric acid

$COOH\text{-}(CHOH)_2\text{–}COOH$

Tartaric acid

$CH_2OH\text{-}COOH$

Glycolic acid

8) **Lactose**

It is composed of one molecule of glucose and on molecule of galactose. It is also called as milk sugar. Lactose in human milk is 60% and in cow’s milk about 4.5%. It is formed in mammary glands. It is dextrorotatory. The ά and β forms has specific rotation of + 90 and + 35 respectively and equimolar has + 52.5. It is less soluble in water, less sweet than sucrose. Upon the action of lactose, lactose yields glucose and galactose. It is a reducing sugar and has C_1-C_4 Glycosidic linkage. So it reduces Fehling’s solution.

9) **Asymmetric carbon atom.**

The carbon atom whose four valencies are satisfied by four different groups is called as asymmetric carbon atom. The presence of asymmetric carbon atom in the carbohydrate makes possible for the formation of isomers in them. The arrangement of the groups in different patterns in Stereoisomers always takes place around asymmetric carbon atom. Number of possible optical isomers in any compound depends upon the number of asymmetric carbon atom. Total number of optical isomers of a compound is given by the formula 2n where n is number of asymmetric carbon atom. Presence of asymmetric carbon atom confers optical activity to it. Accordingly D and L types of compound are present.

10) **Classification of carbohydrate**

Depending upon their complexity and behaviour on hydrolysis, the carbohydrates are classified into three categories

a) Monosaccharides b) Oligosaccharides c) Polysaccharides

Monosaccharides based on number of carbon atoms present in it, it is classified as follows 1) Triose – Glyceraldehydes 2) Tetroses- Erythroses 3) Pentoses- Riboses 4) Hexoses- Glucoses/ Fructoses 5) Heptoses - Sedoheptuloses

Oligosaccharides are classified based on number of monosaccharides formed on hydrolysis. Number of monosaccharides formed ranged from 2 to 10. Accordingly a) two – disaccharides b) three- trisaccharides c) four- tetrasaccharides etc.,

Disaccharides based on the linkage is classified into 1) reducing sugar and 2) non- reducing sugar

Reducing sugar has got two types of linkage a) C1-C4 (maltose and lactose) and C1- C6 (gentibiose and melibiose) and non- reducing sugar has C1-C1 linkage (sucrose)

Polysaccharides are classified into a) Homoglycans (Cellulose) b) Heteroglycans (Hemi cellulose). Based on their functional aspect, polysaccharides are classified into a) Nutrient / digestible/storage – (Starch, glycogen and inulin) b) Structural/indigestible (Cellulose/ pectin/chitin).

11) **Mutarotation**

When a monosaccharide is dissolved in water, the optical rotatory power of the solution gradually changes until a constant value is attained. A freshly prepared aqueous solution of ά D glucose for instance has a specific rotation of + 112.2 and when this solution is allowed to stand, the rotation falls to + 52..7 and remains constant. This gradual change in specific rotation is known as mutarotation. All reducing sugars undergo mutarotation.

12) **Reducing and non-reducing sugar**

Any carbohydrate which is capable of being oxidized and causes the reduction of other substances without having to be hydrolysed first is known as reducing sugar. The carbohydrates which are unable to be oxidized and do not reduce other substances are known as non- reducing sugars. All free monosaccharides having free aldehydes or ά hydroxy ketonic groups are capable of being oxidized and cause the reduction of the other substances is reducing sugars. Fehling's solution and Benedict's solution are used to carry out the oxidation to check for reducing sugar. The sucrose is non- reducing sugar because the Glycosidic bond blocks reducing groups of both the monosaccharides units. Reducing sugar will exhibit mutarotation. Reducing sugar will have C1-C4 linkage while non-reducing sugar will have C1-C1 linkage. Reducing sugar are hemiacetal, do form osazones with phenyl hydrazines, form oximes with hydroxylamine. But non- reducing sugar are acetal, do not form osazone and oximes

13) **Polysaccharides**

It is formed by combination of many monosaccharides joined together by Glycosidic linkages. They are also called as glycans. It is classified into Homoglycans (made up of one kind of monosaccharides) and Heteroglycans (made up of different kinds of monosaccharides). Polysaccharides are grouped based on their functional aspect, polysaccharides are classified into a) Nutrient/ digestible/storage – (Starch, glycogen and inulin) b) Structural/indigestible (Cellulose/ pectin/chitin). The starch is made up of amylose and amylopectin and is reserve food materials for higher plants. Glycogen is the major reserve food in animals and often called as animal starch. Cellulose is a long straight chain polysaccharides consisting of many β- D glucose units joined together by ά -1-4 Glycosidic linkage. Hemi cellulose is made up of pentoses and hexoses. It is long chained but shorter chains with branched ones.

ESSAY TYPES

1. What are the main types of carbohydrates found in plants? Add a note on their importance in plant metabolism

The main type of carbohydrates found in plants are monosaccharides (triose, tetroses, pentoses, hexoses and heptoses), oligosaccharides (disaccharides and trisaccharides) and polysaccharides (storage polysaccharides- starch, glycogen) and structural polysaccharides (cellulose, chitin). The metabolism of carbohydrates is of utmost importance to organisms individually and collectively. Fundamentally all organic food stuffs are ultimately derived from the synthesis of carbohydrates through photosynthesis. The catabolism of carbohydrates provides the major share of energy requirements for maintainenance of life and work function. Carbohydrates act as an energy reservoirs and serve architectural function and are important constituents of nucleic acids. Plant contains higher amount of carbohydrates compared to animals. The carbohydrates present in grains, tubers, roots are referred to as starch and forms a stable food.

2) Describe the mechanism of biosynthesis and degradation of sucrose

Synthesis of sucrose

Synthesis of sucrose in plants may take place in three different ways

1) From glucose –1-phosphate and fructose in the presence of the enzyme Phosphorylase. Eg. In bacteria

$$\text{Glucose- 1-Phosphate + Fructose} \longrightarrow \text{Sucrose + Pi}$$

2) From UDPG (Urdine Di- phosphate glucose) and fructose in the presence of the enzyme sucrose synthetase eg. In higher plants

$$\text{UDPG + Fructose} \xrightarrow[\text{Sucrose synthetase}]{} \text{UDP + Sucrose}$$

3) From UDPG and fructose- 6-phosphate in the presence of enzyme sucrose Phosphate synthetase. eg, in higher plants

$$\text{UDPG + fructose -6-phosphate} \xrightarrow[\text{Sucrose phosphate synthetase}]{} \text{UDP + sucrose phosphate}$$

Sucrose phosphate is hydrolysed in the presence of phosphatase enzyme to yield sucrose

$$\text{Sucrose Phosphate} \xrightarrow[\text{Phosphatase}]{} \text{Sucrose + phosphate}$$

Break down of sucrose

Sucrose is broken down or hydrolysed to yield glucose and fructose in the presence of the enzyme invertase or sucrase. The reaction is irreversible

$$\text{Sucrose} + H_2O \xrightarrow[\text{Invertase}]{} \text{Glucose + Fructose}$$

3) **Describe the properties of carbohydrate**

Monosaccharides

1) **Reaction with alcohol –** The Glycosidic OH group of mutarotating sugars reacts with alcohol to form ά and β glycosides or acetals. Thus glucose form glucosides and fructose from fructosides
2) **Reaction with acetic anhydride-** The Glycosidic and alcoholic OH group of monosaccharides and disaccharides react with acetylating agents to form acetate derivatives called esters
3) **Oxidation with acids-** Only the aldehydes group of sugar is oxidized to produce monocarboxylic acid with bromine water, while with nitric acid both aldoses and ketoses react to form dicarboxylic acid
4) **Oxidation with metal hydroxides-** Metal hydroxides like copper hydroxide oxidize free aldehydes or keto group of mutarotating monosaccharides and at the same time they themselves reduce to free metal

$$\text{Reducing sugar} + Cu^{2+} \longrightarrow \text{oxidized sugar} + 2\,Cu^{+}$$

$$2Cu^{+} + 2OH^{-} \longrightarrow \underset{\text{Yellow}}{2Cu.OH} \longrightarrow \underset{\text{Red}}{Cu_2O + H_2O}$$

This sugar is active ingredient in Fehling’s, Benedict’s and Barfoed’s reagent

5) **Reduction-** The sugars undergo reduction with sodium amalagam to form corresponding alcohols. Glucose yields sorbitol and fructose yields mixture of sorbitol and mannitol. With strong acids, it undergo reduction to form levulinic acid.

$\longrightarrow$

6) **Reaction with hydrogen cyanide-** (Kilani synthesis). It forms cyanohydrins

7) **Reaction with alanine** – The aldehydes group of carbohydrate condenses with the amino group of alanine to form Schiff's base.

8) **Reaction with Phenyl hydrazine** – reaction of monosaccharides with phenyl Hydrazine yields osazone

9) **Fermentation-** Monosaccharides are readily fermented by yeast to form alcohol

Disaccharides

1) **Sucrose**- it is dextrorotatory, a non- reducing sugar, does not exhibit mutarotation, does not form osazone and fermentable
2) **Lactose**- It is dextrorotatory, a reducing sugar, exhibit mutarotation, form osazone and Non-fermentable
3) **Maltose**- It is dextrorotatory, a reducing sugar, exhibit mutarotation, form osazone and fermentable
4) **Cellobiose**- It is a reducing sugar, exhibit mutarotation

Polysaccharides

1) **Starch**- It is made up of amylose and amylopectin. Amylose gives blue colour with Iodine, amylopectin gives purple with iodine and dextrorotatory,
2) **Glycogen**- It is a non-reducing sugar, gives red colour with iodine, white powder, Fairly stable in hot alkali
3) **Cellulose**- It gives no colour with iodine, fibrous, tough, white solid
4) **Inulin**- It is a non-reducing sugar, gives no colour with iodine
5) **Chitin-** It is non-reducing sugar

4) **Compare the structural features of amylose, cellulose and chitin**

Sl.No	Amylose	Cellulose	Chitin
1	Formed by elimination of a molecule of water from Glycosidic OH group of one ά- D glucose and alcoholic OH group on carbon 4 of the adjacent ά- D glucose	Formed by eliminating a molecule of water from Glycosidic OH on carbon atom 1 of one β- D glucose and the alcoholic OH group on carbon 4 of the adjacent β- D glucose	The alcoholic OH group on carbon atom 2 of β- D glucose units is replaced by N- acetylamino group
2	The linkage in amylose is ά -1-4 linkage	It is regarded as an anhydride of β- D glucose	It is linear polymer of N- acetyl- D_ glucosamine units joined together by β- 1-4 glucosidic linkage

5) **Enumerate the important monosaccharides and their occurrence**

The important monosaccharides are a) Triose b) Tetroses c) Pentoses d) Hexoses e)

Heptoses

1) **Triose –** Glyceraldehydes. The simplest compound having three carbon atoms. It is colourless, sweet, crystalline and soluble in water, insoluble in ether, cannot be hydrolysed and formed in the plants during glycolysis

2**)** **Tetroses**- Erythroses. This has four carbon atoms, colourless, crystalline, soluble in water, insoluble in ether. It is produced in plants in photosynthesis in presence of transketolase from fructose -6- phosphate

3) **Pentoses**- Riboses and Ribulose. They have 5 carbon atoms. In plants they are found in combined state. It reduces Fehling's solution and gives Molisch's test. They are not fermentable. Ribose is an aldopentoses and ribulose is keto pentoses. It is used in the formation of RNA. Arabinose is colourless, sweet in taste. It can be obtained by the hydrolysis of gum Arabic, peach gum and cherry gum. It reduces Fehling's solution. Xylose is aldosugar and xylulose is ketonoic form and is formed in photosynthesis. It is colourless, crystalline, optically inactive. It is also formed by hydrolysis of wood gum or xylose.

4) **Hexose**s- Glucose/ fructose- These sugars are six carbon atoms and cannot be hydrolysed. Glucose is called as dextrose formed by the hydrolysis of cane sugar, glucosides, starch and cellulose etc., It is needle shaped crystals, anhydrous. Fructose is a keto sugar formed in equal quantity with glucose by the hydrolysis of cane sugar, soluble in hot absolute alcohol and ether. Mannose is prepared by hydrolysing mannane found in ivory nuts.

5) **Heptoses –** Glucoheptoses and Sedoheptulose. They are seven sugar carbon atom. It is ketosugar and formed in photosynthesis

6) Discuss the four important chemical reactions of carbohydrates

1) **Oxidation with acids:**

a) **Mild oxidizing agent (HOBr**). Only the aldehydes group is oxidized by bromine water to produce monocarboxylic acid. Ketoses do not react with bromine water

$$\underset{\text{Glucose}}{CHO\text{-}(CHOH)_4\text{-}CH_2OH} + HOBr \longrightarrow \underset{\text{Gluconic acid}}{COOH\text{ - }(CHOH)_4\text{-}CH_2OH} + HBr$$

b) **Strong acids (HNO$_3$)**: Both aldoses and ketoses react with nitric acid to form dicarboxylic acid

$$\underset{\text{Glucose}}{CHO\text{-}(CHOH)_4\text{-}CH_2OH} + HNO_3 \longrightarrow \underset{\text{Glucaric acid}}{COOH\text{ - }(CHOH)_4\text{-}COOH} + H_2O$$

c) **Oxidation with metal hydroxide**: Metal hydroxide Cu(OH)$_2$ oxidize the free aldehydes and keto group and reduce itself to form free metal

$$\text{Reducing sugar} + \underset{\text{Blue}}{2\,Cu^{2+}} \longrightarrow \text{Oxidized sugar} + 2\,Cu^{2+}$$

$$2\,Cu^{2+} + 2OH^- \longrightarrow \underset{\text{Yellow}}{2CuOH} \longrightarrow \underset{\text{Red}}{Cu_2O} + H_2O$$

2) **Reduction:** The sugars may be reduced in various ways depending upon the type of reducing sugars used.

a) With sodium amalgam: The monosaccharides are reduced to form alcohol by treating it with sodium amalgam. Glucose yields sorbitol and fructose yields mixture of sorbitol and mannitol

$$\underset{\text{Glucose}}{CHO\text{-}(CHOH)_4\text{-}CH_2OH} + \text{Na amalgam} \longrightarrow \underset{\text{Sorbitol}}{CH_2OH\text{-}(CHOH)_3\text{-}CH_2OH}$$

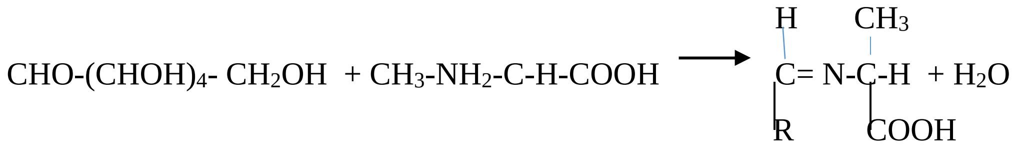
$$\underset{\text{Fructose}}{CH_2OH\text{-}C{=}O\text{-}(CHOH)_4\text{-}CH_2OH} + 4H \longrightarrow \left.\begin{array}{c} \underset{\text{Sorbitol}}{CH_2OH\text{-}(CHOH)_3\text{-}CH_2OH} \\ \underset{\text{Mannitol}}{CH_2OH\text{-}(CHOH)_3\text{-}CH_2OH} \end{array}\right\}$$

b) **With strong mineral acid**- Hexoses on reaction with acid undergo reduction to form 5-hydroxy methyl furfural which is on further heating form levulinic acid

c) **With dilute alkali**: Glucose fructose and mannose are interconvertible in weak alkaline solution such as calcium hydroxide and barium hydroxide at low temperature

3) **Reaction with Phenyl hydrazine**: One mole of aldose reacts with one mole of phenyl hydrazine to form one molecule of hydrazones and hydrazones is oxidized to form aldohydrazones and finally osazone. Osazone is noticed in monosaccharides and disaccharides.

4) **Reaction with alanine.** The aldehydes group of carbohydrates condenses with amino group of alanine to form Schiff's base



$$CHO\text{-}(CHOH)_4\text{-}CH_2OH + CH_3\text{-}NH_2\text{-}C\text{-}H\text{-}COOH \longrightarrow \underset{R}{\overset{H}{C}}{=}N\text{-}\underset{COOH}{\overset{CH_3}{C}}\text{-}H + H_2O$$

7) **Write a detailed note on polysaccharides with examples**

Polysaccharides are also compound sugars and yield more than 10 molecules of monosaccharides on hydrolysis. These may be further classified depending up on whether monosaccharides molecules produced as a result of the hydrolysis of polysaccharides of the same type (homo polysaccharides) or if different types (Hetero polysaccharides). The general formula $(C_6H_{10}O_5)x$. Based on the functional aspect, polysaccharides are grouped in to

a) **Nutrients/digestible polysaccharides** – These act as a metabolic reserve of monosaccharides in plant and animals eg. Starch, glycogen

b) **Structural/ indigestible polysaccharides**- These serve as rigid mechanical structures in plants and animals- cellulose, pectin and chitin

1) **Starch**- Major reserve food in higher plants. It consists of ά-D –glucose and made up of amylose and amylopectin. Amylose gives blue colour with iodine; amylopectin gives purple colour with iodine. It is dextrorotatory. It yields glucose units on hydrolysis. Hence it is a glucosans

2) **Glycogen**- It is major source of food of animals. It is stored in liver and muscles of animals. The constituent monosaccharides are ά-D –glucose. It si non-reducing sugar, gives red colour with iodine. It yields glucose units on hydrolysis, hence a glucosans

3) I**nulin** – It is found in tubers and roots. The constituent monosaccharide is β- D-glucose. It is non-reducing sugar, gives no colour with iodine, yields fructose on hydrolysis, hence fructosans

4) **Cellulose**- It is widely distributed in plants. The constituent monosaccharide is is β-D-glucose., gives no colour with iodine , yields glucose units on hydrolysis, hence glucosans

5) **Pectin**-It is in fruits of many plants. The constituent monosaccharide is ά-D –galactouronic acid

6) **Chitin**- Most abundant in nature after cellulose, found in fungi and anthropods. The constituent monosaccharides are N- Acetyl- D- glucosamine. It is non- reducing sugar.

8) What re polysaccharides? Differentiate between amylose and amylopectin

Polysaccharides are formed by the combination of many monosaccharides joined together by Glycosidic linkages. They are also known as glycans and are again classified into Homoglycans and Heteroglycans. These polysaccharides which are made up of only kind of monosaccharides are known as Homoglycans. While those which are made up of two or more kinds of monosaccharides are known as hetero glycans. Polysaccharides are hydrolysed either by enzymes or mineral acid. The most common examples of polysaccharides are starch, cellulose, glycogen, Chitin and inulin

Amylose	Amylopectin
1) It contains 15-20% in starch	1) It contains 80-85% in starch
2) It is straight chain compound of about 100-700 glucose units	2) It is branched chain compound of about 300-5500 glucose units
3) The molecular weight is 10000 to 50000	The molecular weight is 50000-1000000
4) It has ά-1-4 linkage	It has ά-1-4 linkage and ά- 1-6 linkage
5) It does not form paste and readily soluble in water	It is not readily dispersed in water and form paste
6) It gives blue colour with iodine	It gives red colour with iodine
7) Enzymatic hydrolysis of amylose with amylase yields maltose	On incomplete hydrolysis yields disaccharide isomaltose

9) **Distinguish between mono, Oligo and Polysaccharides**

Monosaccharides	**Oligosaccharides**	**Polysaccharides**
1) It is simplest sugar eg. Glucose	It is also sugar, but it may be non-reducing or reducing sugar eg., sucrose, maltose	It is no reducing sugar eg. Starch, glycogen
2) It contains generally up to 9 carbon atoms	It contains generally 12 to 36 carbon atoms	It contains more number of carbon atoms
3) They contain carbonyl group and show the properties of aldehydes and ketones	Do not contain carbonyl group and does not show reaction with aldehydes or ketone group	Do not contain carbonyl group and does not show reaction with aldehydes and ketone group
4) They are colorless, crystalline and sweet	They are generally colorless, crystalline and sweet	They are colorless, amorphous and tasteless
5) Soluble in water	Soluble in water	Insoluble in water
6) They are optically active	They are optically active	They are optically inactive
7) They have free or potential aldehydes or ketone group	No	No
8) It cannot be hydrolyzed	It can be hydrolyzed to yield 2-10 molecules of monosaccharides	It can be hydrolyzed to yield more than 10 molecules of monosaccharides

10) **Discuss the importance/significance of carbohydrates**

A**) In plants and animals**

1) It is structural materials of plants
2) It is reserve food material as starch in tubers, roots and grains
3) Sucrose is present in the nectar of flowers and in fruits
4) Carbohydrate on oxidation yields energy which is utilized by plants for various physiological processes

B) **For human beings**

1) The starches and sugar are the main food easily digestible and oxidisable to provide energy for various physiological activities
2) Carbohydrates present in seeds such as rice, maize, rye, barley are utilized for the production of alcoholic beverages
3) Carbohydrates derivatives such as glucosides form important drugs and other medicines for various diseases
4) Carbohydrates particularly cellulose and its derivatives are used in the production of artificial silk, paper and plastics

5) Blood contains glucose as sugar. Blood glucose are removed by muscles and older tissues and form glycogen which provides energy on oxidation. Mammary glands form milk sugar

11) What are the important tests for carbohydrates?

1) **Fehling's solution test**: Mix Fehling's solution A and B in equal amounts in a test tube. Now add equal volume of glucose solution and boil it till brick red precipitate is produced. The Fehling's solution contains cupric sulfate, sodium hydroxide and sodium potassium tartrate. When solution is mixed with glucose and boiled, the aldehydes group of glucose is oxidized and cupric salt is reduced to cuprous oxide giving red precipitate

$$C_6H_{12}O_6 + 2\ \underset{\text{Blue}}{Cu(OH)_2} \longrightarrow C_6H_{12}O_7 + \underset{\text{Red}}{Cu_2O} + H_2O$$

2) **Benedict's test**: Mix 3 ml of Benedict's solution and one ml of glucose solution and warm them gently. A reddish brown colour of cuprous oxide is produced. On warming the colour of the mixture turn from blue to green and from green to reddish brown. Benedict's solution contains blue colour copper sulfate. When alkaline solution of glucose is added to this and warmed, the aldehydes group of glucose is oxidized and blue colored copper sulfate is changed to green colored copper hydroxide and ultimately copper hydroxide changed to reddish brown cuprous oxide

3) **Molisch's Test**: Sugar and their polymers give characteristic colour in the presence of strong sulfuric acid and ά naphthol. The sugars in the presence of acids undergo dehydration to form furfural and the violet colour is produced by the condensation of aldehydes and phenol

4) **Seliwanoff test**: It is a chemical test which distinguishes between aldose and ketose sugar. Sugars containing keto group when heated with HCl and resorcinol, bright red colour is produced

PROTEINS

The word "Protein" was coined by J.J. Berzelius in 1838 and was derived from the Greek word "Proteios" meaning the ‘first rank’.

Introduction:

- Most abundant organic molecules of the living system
- Its fundamental basis of structures and function of life.
- 50 % of dry weight of every cell
- It’s a polymer of L α-amino acids.
- 300 different amino acids occur in nature –only 20 as standard amino acids.
- 21st amino acid added - Seleno cysteine

Definition

Proteins are organic complex nitrogenous compounds of high molecular weight, formed of C, H, O, N [N= 16%].

They are formed of a number of amino acids linked together by peptide linkage [-CO-NH-].

The carboxylic group of the first amino acid units with the amino group of the second amino acid and so on.

General Properties of Proteins

- Proteins are substances of high molecular weight.
- Proteins form colloidal solution and having its same properties as:
  Tyndall effect & Brownian movement
- Proteins are non-dialyzable due to their large molecules.
- Proteins are amphoteric which liable to react with acid and alkali.
- Each protein has its own isoelectric point.
- Protein acts as a buffer solution which resists the change of its pH by addition of acid or alkali.
- Denaturation

Biological Importance of Proteins

- They provide the body with nitrogen, sulfur, and some vitamins.
- Formation of enzymes and protein hormones.
- Formation of supporting structures in the body as bone, cartilage, skin, nails, hair and muscles.
- They enter in the formation of buffer system of the blood.
- They enter in the formation of haemoglobin
- They include plasma proteins, which carry hormones, minerals and lipids (in the form of lipoprotein complex).
- They enter in formation of antibodies (immunoglobulins

CLASSIFICATION OF PROTEINS

Proteins are classified on the basis of

– Chemical nature and solubility

- Simple • Conjugates • Derived

– Function

• Structural • Enzyme or catalytic • Transport • Hormonal
• Contractile • Storage • Genetic • Defence • Receptor

– Nutritional Importance

• Complete • partially incomplete • Incomplete

Classification of proteins according to their solubility and composition

I- Simple proteins:

i.e. on hydrolysis gives only amino acids

as albumin , globulins, glutellin, Prolamines , Protamines, Histones, Albuminoids (Scleroproteins).

Albumins

- Albumins are readily soluble in water, dilute acids and alkalies coagulated by heat.
- Seed proteins contain albumin in lesser quantities.
- Albumins may be precipitated out from solution using high salt concentration, a process called 'salting out'.
- They are deficient in glycine.
- Serum albumin and ovalbumin (egg white) are examples

Globulins

- Globulins are insoluble or sparingly soluble in water, but their solubility is greatly increased by the addition of neutral salts such as sodium chloride.
- These proteins are coagulated by heat.
- They are deficient in methionine.
- Serum globulin, fibrinogen, myosin of muscle and globulins of pulses are examples.

Prolamins

- Prolamins are insoluble in water but soluble in 70-80% aqueous alcohol.
- Upon hydrolysis they yield much proline and amide nitrogen, hence the name prolamin.
- They are deficient in lysine.
- Gliadin of wheat and zein of corn are examples of Prolamins.

Glutelins

Glutelins are insoluble in water and absolute alcohol but soluble in dilute alkalies and acids. They are plant proteins e.g., glutenin of wheat.

Histones

Histones are small and stable basic proteins

- They contain fairly large amounts of basic amino acid, histidine.
- They are soluble in water, but insoluble in ammonium hydroxide.
- They are not readily coagulated by heat.
- They occur in globin of haemoglobin and nucleoproteins

Protamines

Protamines are the simplest of the proteins.

- They are soluble in water and are not coagulated by heat.
- They are basic in nature due to the presence of large quantities of arginine.

- Protamines are found in association with nucleic acid in the sperm cells of certain fish.
- Tyrosine and tryptophan are usually absent in protamines.

Albuminoids

- These are characterized by great stability and insolubility in water and salt solutions.
- These are called Albuminoids because they are essentially similar to albumin and globulins.
- They are highly resistant to proteolytic enzymes.
- They are fibrous in nature and form most of the supporting structures of animals.
- They occur as chief constituent of exoskeleton structure such as hair, horn and nails.

ii. Conjugated or compound proteins

- ➢ These are simple proteins combined with some non-protein substances known as prosthetic groups.
- ➢ The nature of the non-protein or prosthetic groups is the basis for the sub classification of conjugated proteins.

Its again of 6 types

1. Phosphoproteins: These are proteins conjugated with phosphate group. Phosphorus is attached to OH group of serine or threonine. e.g. Casein of milk and vitellin of yolk.
2. Lipoproteins: These are proteins conjugated with lipids. Functions: a - help lipids to transport in blood b - Enter in cell membrane structure helping lipid soluble substances to pass through cell membranes
3. Glycoproteins: proteins conjugated with sugar (carbohydrate e.g. – Mucin , Some hormones such as erythropoietin, present in cell membrane structure blood groups**.**
4. Nucleoproteins: These are basic proteins (e.g. histones) conjugated with nucleic acid (DNA or RNA). e.g. a- chromosomes : are proteins conjugated with DNA
   b- Ribosomes : are proteins conjugated with RNA
5. Chromoprotein: prosthetic group is colored in nature. Ex: Haemoglobins, cytochromes
6. . Metallo protein: it contains metal ions such as Fe, Co, Zn, Cu, Mg,

iii. Derived proteins

- Denatured or degraded product of simple or conjugated protein by the action of acids, alkalies or enzymes
- They include two types of derivatives, primary-derived proteins and secondary-derived proteins

Its of 2 types

– Primary derived protein: denatured or coagulated or first hydrolysed product of proteins. They are

• Cogulatedproteins: denatured protein produced by agents such as heat, acids, alkalies

• Proteans: earliest product of protein hydrolysis by enzymes, dilute acids, alkalies etc. Insoluble in water

• Metaprotein: second stage of protein hydrolysis obtained by treatment with slightly stronger acids and alkalies

Secondary protein derivatives:

These are the hydrolytic products of proteins

- Proteoses:

Result from partial hydrolysis of proteins.

- Peptones:

Result from further hydrolysis of proteases.

Soluble in H_2O.

- Peptides:

Resulting from further hydrolysis of peptones.

- Amino acids

Protein -----> Protean ------- Metaprotein

Proteoses ------> Peptones ------->Peptides ------ amino acids

Classification of proteins based on function

Catalytic proteins – Enzymes

- The most striking characteristic feature of these proteins is their ability to function within the living cells as biocatalysts.
- These biocatalysts are called as enzymes.
- Enzymes represent the largest class.
- Nearly 2000 different kinds of enzymes are known, each catalysing a different kind of reaction.
- They enhance the reaction rates a million fold

Regulatory proteins – Hormones

These are polypeptides and small proteins found in relatively lower concentrations in animal kingdom but play highly important regulatory role in maintaining order in complex metabolic reactions e.g., growth hormone, insulin etc.

Protective proteins – Antibodies

- These proteins have protective defence function.
- These proteins combine with foreign protein and other substances and fight against certain diseases.
- e.g., immunoglobulin.
- These proteins are produced in the spleen and lymphatic cells in response to foreign substances called antigen.
- The newly formed protein is called antibody which specifically combines with the antigen which triggered its synthesis thereby prevents the development of diseases.
- Fibrin present in the blood is also a protective protein

Storage proteins

It is a major class of proteins which has the function of storing amino acids as nutrients and as building blocks for the growing embryo.

Storage proteins are source of essential amino acids, which cannot be synthesized by human beings.

The major storage protein in pulses is globulins and Prolamins in cereals. In rice the major storage protein is glutelins. Albumin of egg and casein of milk are also storage proteins.

Transport proteins

- Some proteins are capable of binding and transporting specific types of molecules through blood.
- Haemoglobin is a conjugated protein composed of colourless basic protein, the globin and ferroprotoporphyrin or haem.
- It has the capacity to bind with oxygen and transport through blood to various tissues.
- Myoglobin, a related protein, transports oxygen in muscle.
- Lipids bind to serum proteins like albumin and transported as lipoproteins in the blood

Toxic proteins

- Some of the proteins are toxic in nature.
- Ricin present in castor bean is extremely toxic to higher animals in very small amounts.
- Enzyme inhibitors such as trypsin inhibitor bind to digestive enzyme and prevent the availability of the protein.
- Lectin, a toxic protein present in legumes, agglutinates red blood cells.
- A bacterial toxin causes cholera, which is a protein.
- Snake venom is protein in nature

Structural proteins

These proteins serve as structural materials or as important components of extra cellular fluid. Examples of structural proteins are myosin of muscles, keratin of skin and hair and collagen of connective tissue.

Carbohydrates, fats, minerals and other cellular components are organized around such structural proteins that form the molecular framework of living material.

Contractile proteins

Proteins like actin and myosin function as essential elements in contractile system of skeletal muscle.

Secretary proteins

Fibroin is a protein secreted by spiders and silkworms to form webs and cocoons.

Classification based on size and shape

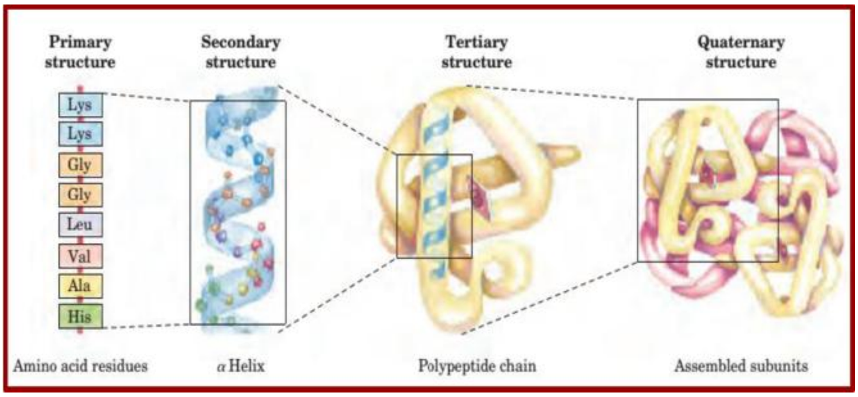
Based on size and shape, the proteins are also subdivided into globular and fibrous proteins

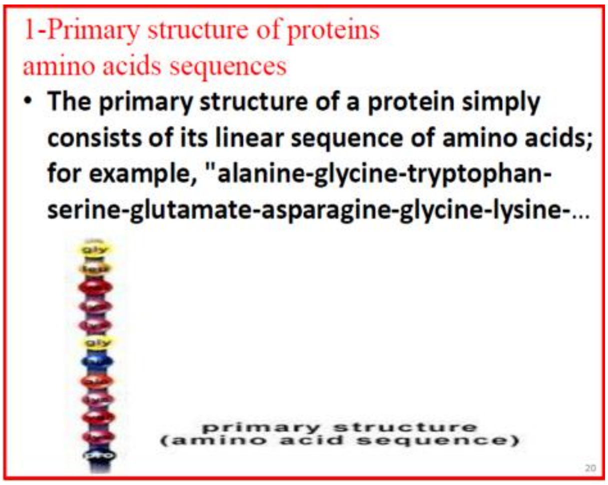
- Globular proteins are mostly water-soluble and fragile in nature e.g., enzymes, hormones and antibodies.
- Fibrous proteins are tough and water-insoluble.
- They are used to build a variety of materials that support and protect specific tissues, e.g., skin, hair, fingernails and keratin

Structures of Protein

Conformation of a protein refers to the three-dimensional structure in its native state. There are many different possible conformations for a molecule as large as a protein. A protein can perform its function only when it is in its native condition. Due to the complexity of three-dimensional structures, the structure of protein is discussed at different levels of its organization. Proteins are formed of a large number of amino acid linked together by peptide bonds (polypeptide chain**).**

There are four orders of protein structures

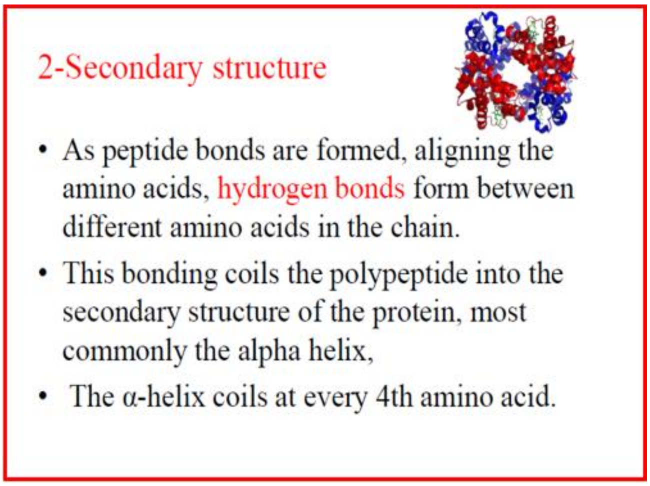




1-Primary structure of proteins
amino acids sequences

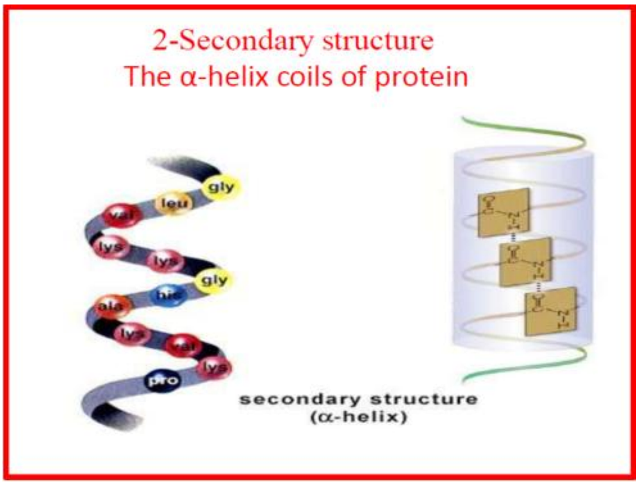
- **The primary structure of a protein simply consists of its linear sequence of amino acids; for example, "alanine-glycine-tryptophan-serine-glutamate-asparagine-glycine-lysine-...**

primary structure
(amino acid sequence)



2-Secondary structure

- As peptide bonds are formed, aligning the amino acids, hydrogen bonds form between different amino acids in the chain.
- This bonding coils the polypeptide into the secondary structure of the protein, most commonly the alpha helix,
- The α-helix coils at every 4th amino acid.

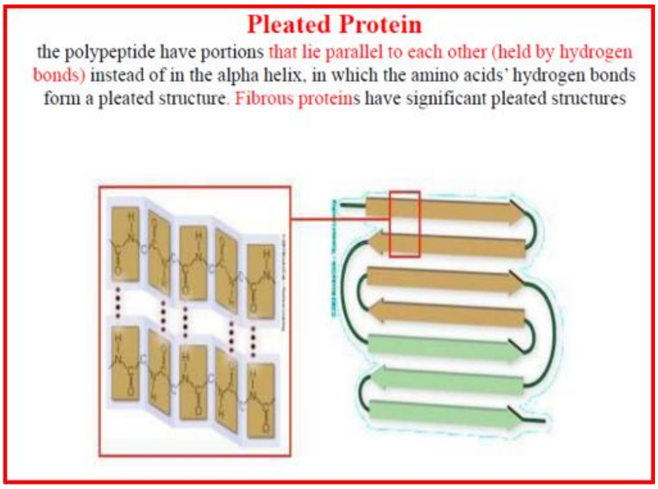


2-Secondary structure
The α-helix coils of protein

secondary structure
(α-helix)

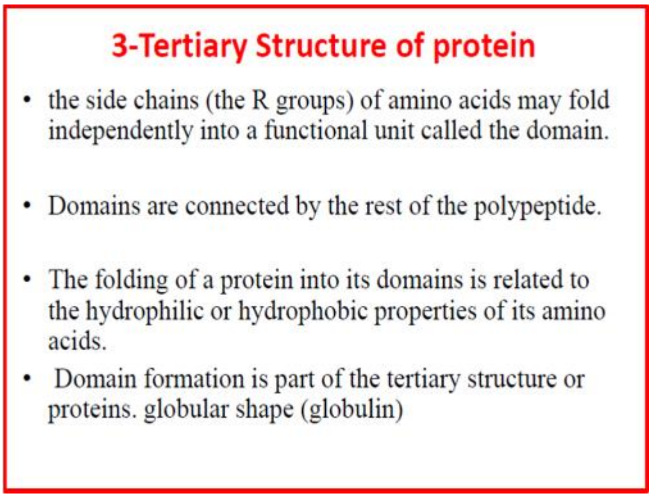
Alpha Helix

α helix is twisted by an equal amount about each α-carbon. The formation of the α-helix is spontaneous and is stabilized by H-bonding between amide nitrogen and carbonyl carbons of peptide bonds spaced four residues apart. This orientation of H-bonding produces a helical coiling of the peptide backbone such that the R-groups lie on the exterior of the helix and perpendicular **to its axis.**



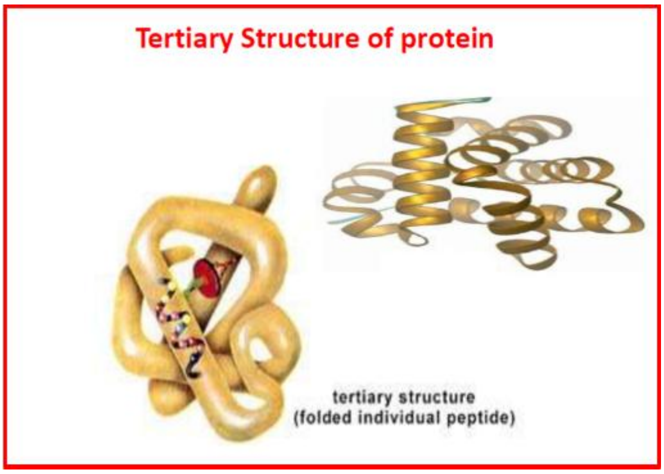
Pleated Protein

the polypeptide have portions that lie parallel to each other (held by hydrogen bonds) instead of in the alpha helix, in which the amino acids' hydrogen bonds form a pleated structure. Fibrous proteins have significant pleated structures



3-Tertiary Structure of protein

- the side chains (the R groups) of amino acids may fold independently into a functional unit called the domain.
- Domains are connected by the rest of the polypeptide.
- The folding of a protein into its domains is related to the hydrophilic or hydrophobic properties of its amino acids.
- Domain formation is part of the tertiary structure or proteins. globular shape (globulin)



Tertiary Structure of protein

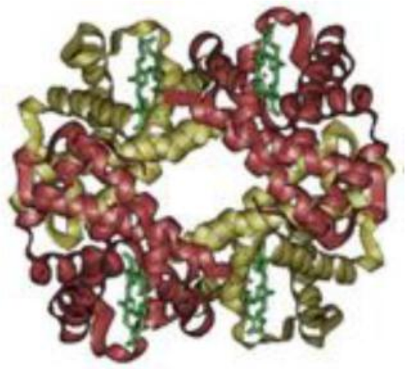
tertiary structure
(folded individual peptide)

5 kind of bonds stabilize tertiary structure

- van der Waals interaction (between neighbouring atoms) Van derWaals forces include attractions and repulsions between atoms, molecules, and surfaces
- H-bonds within the chains or between chains
- hydrophobic interactions (between non-polar)
- ionic interactions (between oppositely charged groups)
- Disulphide linkages, the SH groups of two neighbouring cysteine's form –s=s bond known as disulphide linkage. (covalent bond**)**

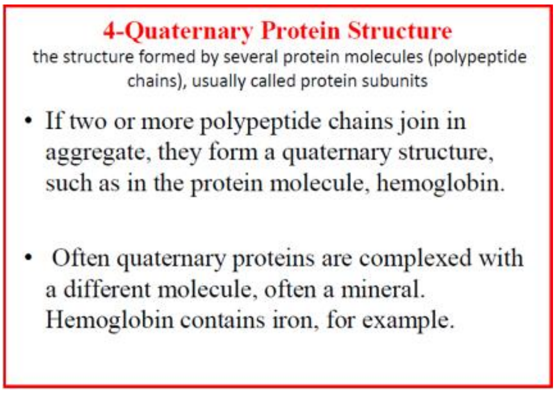
Quaternary Structure

- Majority of proteins are composed of single polypeptide chains
- Some of protein consists of 2 or more polypeptide chain which may be identical or different
- Such protein are termed as oligomers and poses quaternary structures.
- Proteins with identical Proteins oligomers.-homo subunits are termed Homodimers
- Proteins containing several distinct polypeptide chains are termed heterodimer
- Quaternary structure refers to a functional protein aggregate (organization) formed by Interpolypeptide linkage of subunits or polypeptide chains**.**



Myoglobin has no quaternary structure

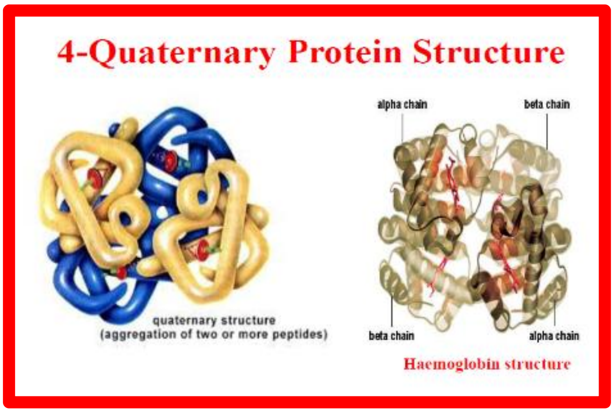
Haemoglobin molecule, which consists of **four separate polypeptide chains**, exhibits quaternary structure.



4-Quaternary Protein Structure

the structure formed by several protein molecules (polypeptide chains), usually called protein subunits

- If two or more polypeptide chains join in aggregate, they form a quaternary structure, such as in the protein molecule, hemoglobin.
- Often quaternary proteins are complexed with a different molecule, often a mineral. Hemoglobin contains iron, for example.



4-Quaternary Protein Structure

Haemoglobin structure

Properties of Protein

Physical

- Pure proteins are generally tasteless, though the predominant taste of protein hydrolysates is bitter.
- Pure proteins are odourless.
- Because of the large size of the molecules, proteins exhibit many properties that are colloidal in nature.
- Proteins, like amino acids, are amphoteric and contain both acidic and basic groups.
- They possess electrically charged groups and hence migrate in an electric field.
- Many proteins are labile and readily modified by alterations in pH, UV radiation, heat and by many organic solvents
- The absorption spectrum of protein is maximum at 280 nm due to the presence of tyrosine and tryptophan, which are the strongest chromophores in that region

Denaturation of Protein

- ✓ The comparatively weak forces responsible for maintaining secondary, tertiary and quaternary structure of proteins are readily disrupted with resulting loss of biological activity.
- ✓ This disruption of native structure is termed denaturation
- ✓ it is a change in native state (physical, chemical, and biological properties) of proteins without destruction of their peptide linkages ,but destruction of secondary bonds leading to unfolding protein molecule

Denaturating agents:

Physical: High temperature, high pressure, X-ray, ultraviolet rays-mechanical agitation.,

Chemical: Strong acids, strong alkalies organic solvents, heavy metals

Results of denaturation

Physical:

- Decrease solubility, increase viscosity and can not be crystallized.

Chemical:

- Unfolding of the protein molecule.
- Destruction of some subsidiary hydrogen bonds.
- Exposure of some groups as (SH) of cystiene.

Biological:

- Loss of activity, if it is hormone or enzyme.
- Loss of antigen antibody reaction (allergic manifestation). Easily digested

Chemical properties

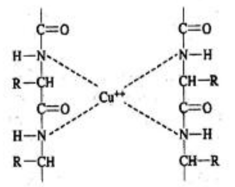
Colour reactions of proteins

1. The colour reactions of proteins are of importance in the qualitative detection and quantitative estimation of proteins and their constituent amino acids.
2. Biuret test is extensively used as a test to detect proteins in biological materials

Biuret reaction

A compound, which is having more than one peptide bond when treated with Biuret reagent, produces a violet colour.

This is due to the formation of coordination complex between four nitrogen atoms of two polypeptide chains and one copper atom



Violet coloured coordination complex

Coordination complex with peptide bonds and copper

Xanthoproteic reaction

Addition of concentrated nitric acid to protein produces yellow colour on heating, the colour changes to orange when the solution is made alkaline. This is due to the nitration of the phenyl rings of aromatic amino acids.

Hopkins-Cole reaction

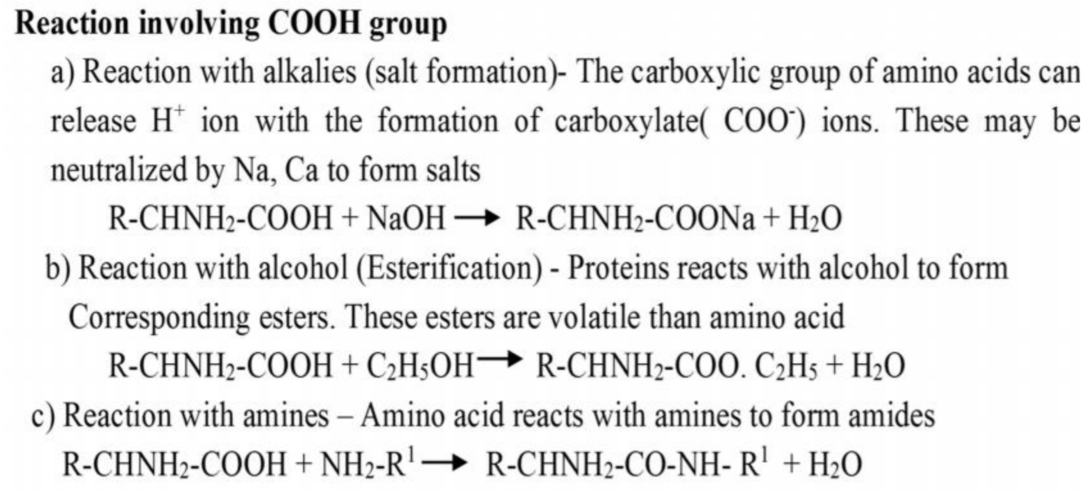
Indole ring of tryptophan reacts with glacial acetic acid in the presence of concentrated sulphuric acid and forms a purple coloured product

Hydrolysis:

a) By acidic agents- Proteins up on hydrolysis with concentrated HCl at 100- 110oC for 6 to 20 hours yield amino acid in the form of their hydrochlorides

b) By alkaline agents – Proteins may also be hydrolyzed by 2N NaOH

c) By proteolytic enzymes- Certain proteolytic enzymes like pepsin and trypsin hydrolyze the proteins under certain conditions of temperature and acidity



Reaction involving COOH group

a) Reaction with alkalies (salt formation)- The carboxylic group of amino acids can release H^+ ion with the formation of carboxylate(COO^-) ions. These may be neutralized by Na, Ca to form salts

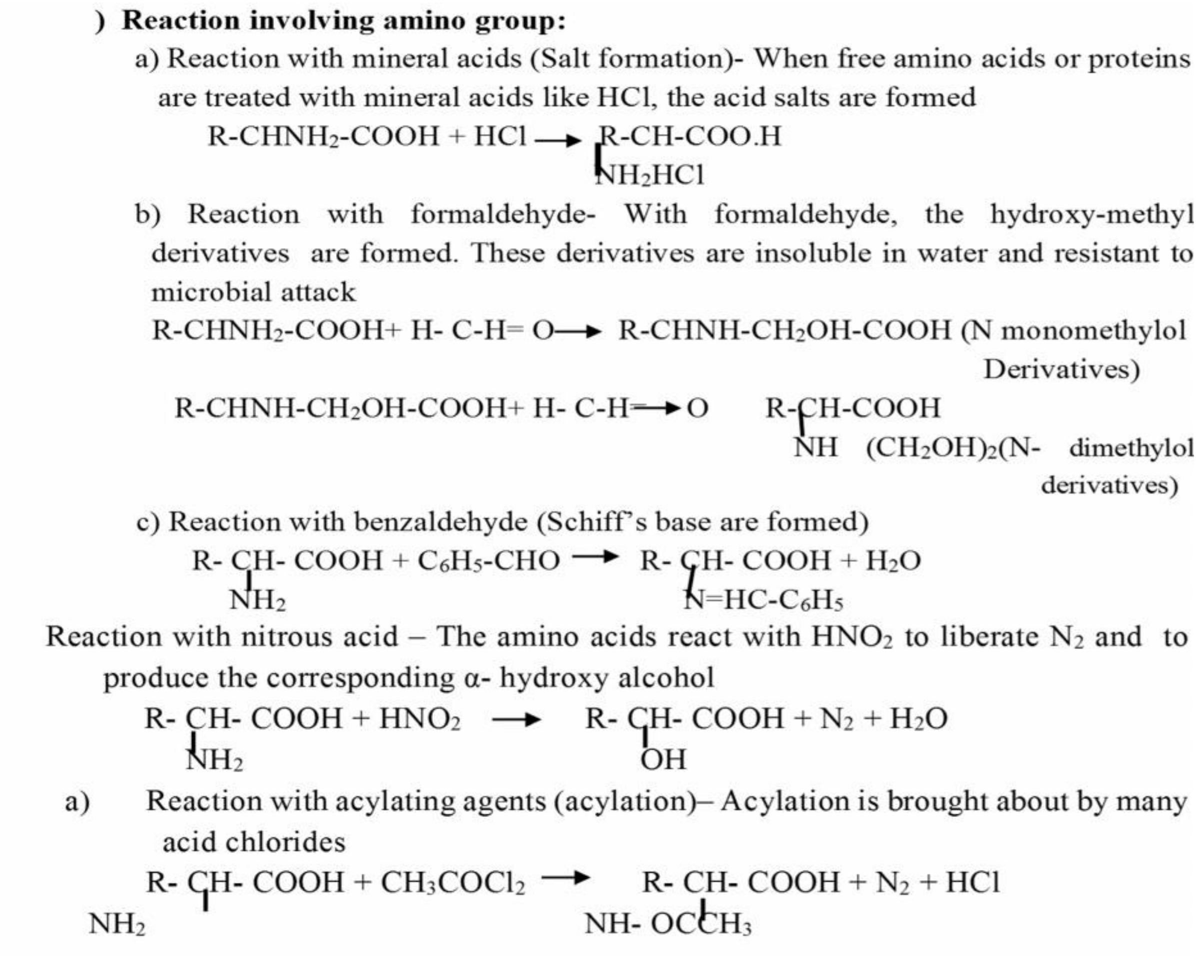
$$R\text{-}CHNH_2\text{-}COOH + NaOH \rightarrow R\text{-}CHNH_2\text{-}COONa + H_2O$$

b) Reaction with alcohol (Esterification) - Proteins reacts with alcohol to form Corresponding esters. These esters are volatile than amino acid

$$R\text{-}CHNH_2\text{-}COOH + C_2H_5OH \rightarrow R\text{-}CHNH_2\text{-}COO.\ C_2H_5 + H_2O$$

c) Reaction with amines – Amino acid reacts with amines to form amides

$$R\text{-}CHNH_2\text{-}COOH + NH_2\text{-}R^1 \rightarrow R\text{-}CHNH_2\text{-}CO\text{-}NH\text{-}\ R^1 + H_2O$$



) Reaction involving amino group:

a) Reaction with mineral acids (Salt formation)- When free amino acids or proteins are treated with mineral acids like HCl, the acid salts are formed

$$R\text{-}CHNH_2\text{-}COOH + HCl \rightarrow \underset{NH_2HCl}{R\text{-}CH}\text{-}COO.H$$

b) Reaction with formaldehyde- With formaldehyde, the hydroxy-methyl derivatives are formed. These derivatives are insoluble in water and resistant to microbial attack

$$R\text{-}CHNH_2\text{-}COOH + H\text{-}\ C\text{-}H{=}\ O \rightarrow R\text{-}CHNH\text{-}CH_2OH\text{-}COOH$$ (N monomethylol Derivatives)

$$R\text{-}CHNH\text{-}CH_2OH\text{-}COOH + H\text{-}\ C\text{-}H \rightarrow O \quad R\text{-}\underset{NH\ (CH_2OH)_2}{CH}\text{-}COOH$$ (N- dimethylol derivatives)

c) Reaction with benzaldehyde (Schiff's base are formed)

$$R\text{-}\ \underset{NH_2}{CH}\text{-}\ COOH + C_6H_5\text{-}CHO \rightarrow R\text{-}\ \underset{N=HC\text{-}C_6H_5}{CH}\text{-}\ COOH + H_2O$$

Reaction with nitrous acid – The amino acids react with HNO_2 to liberate N_2 and to produce the corresponding α- hydroxy alcohol

$$R\text{-}\ \underset{NH_2}{CH}\text{-}\ COOH + HNO_2 \rightarrow R\text{-}\ \underset{OH}{CH}\text{-}\ COOH + N_2 + H_2O$$

a) Reaction with acylating agents (acylation)– Acylation is brought about by many acid chlorides

$$R\text{-}\ \underset{NH_2}{CH}\text{-}\ COOH + CH_3COCl_2 \rightarrow R\text{-}\ \underset{NH\text{-}\ OCCH_3}{CH}\text{-}\ COOH + N_2 + HCl$$

Reaction involving both COOH and NH_2 groups

a) Ninhydrin test- It is powerful oxidizing agent and causes oxidative decarboxylation of α-amino acids producing CO2, NH3 and aldehyde with one less carbon atom than the parent amino acids. The reduced Ninhydrin then react with the liberated NH3 forming blue colored complex

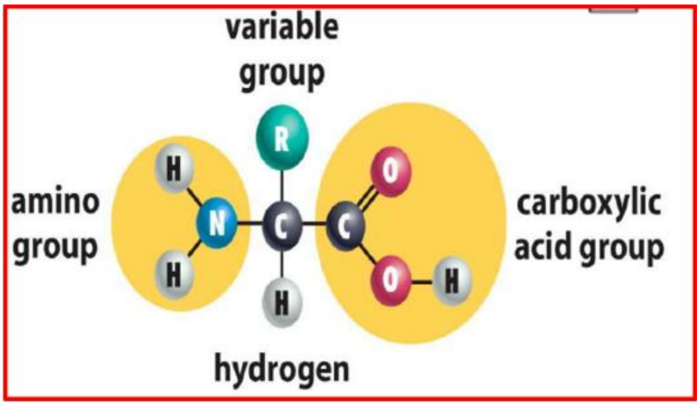
Why is protein folding?

Protein folding is the process by which a protein structure assumes its functional shape or conformation. All protein molecules are heterogeneous unbranched chains of amino acids. By coiling and folding into a specific three-dimensional shape they are able to perform their biological function

Protein folding is a spontaneous process directed by the amino acid sequence of each individual protein. Protein folding occurs in a cellular compartment called the endoplasmic reticulum. This is a vital cellular process because proteins must be correctly folded into specific, three-dimensional shapes in order to function correctly. Unfolded or misfolded proteins contribute to the pathology of many diseases

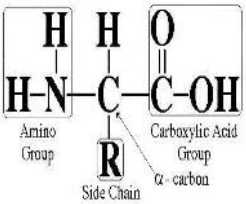
Protein folding is “the final step in the decoding of genetic information

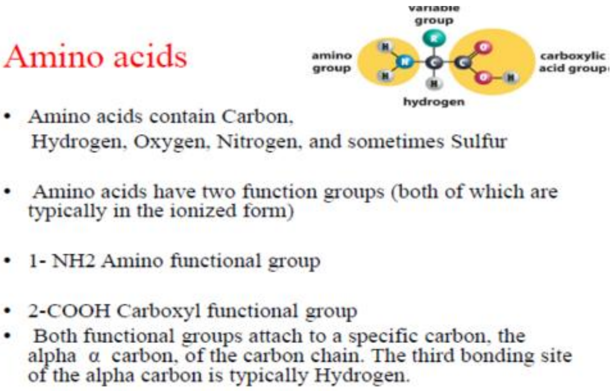
AMINO ACID



General Structure of Amino Acid

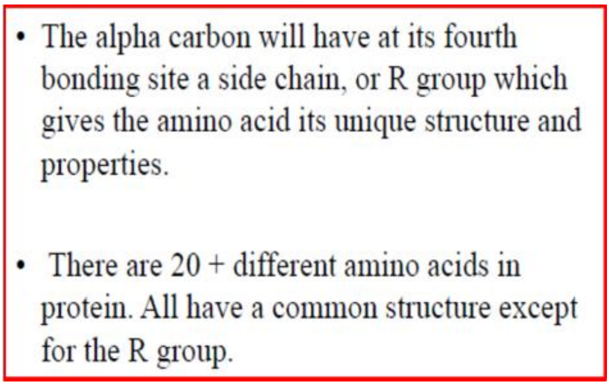
Each amino acid (except proline) has a carboxyl group, an amino group and a distinctive side chain bonded to the alpha carbon atom. At physiological pH the carboxyl group is dissociated forming the negatively charged carboxylate ion (-COO-), and the amino group is protonated (-NH_3^+)





Amino acids

- Amino acids contain Carbon, Hydrogen, Oxygen, Nitrogen, and sometimes Sulfur
- Amino acids have two function groups (both of which are typically in the ionized form)
- 1- NH2 Amino functional group
- 2-COOH Carboxyl functional group
- Both functional groups attach to a specific carbon, the alpha α carbon, of the carbon chain. The third bonding site of the alpha carbon is typically Hydrogen.



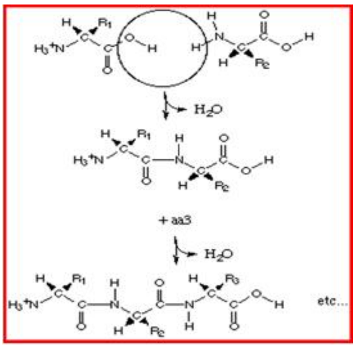
- The alpha carbon will have at its fourth bonding site a side chain, or R group which gives the amino acid its unique structure and properties.
- There are 20 + different amino acids in protein. All have a common structure except for the R group.

Bond Formation

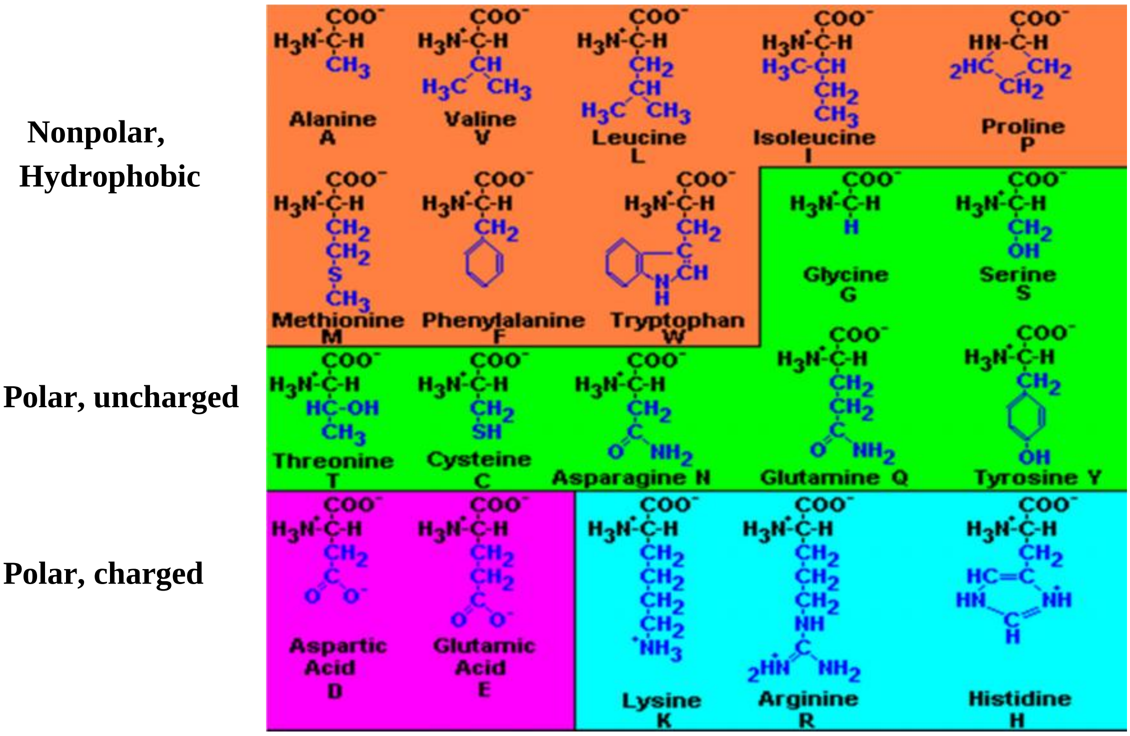
Linking two amino acids together

Definitions (N-terminal, C-terminal,

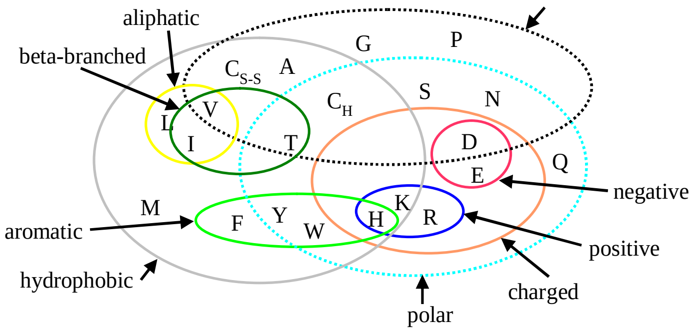
Polypeptide backbone, amino acid residue, side chains)



20 Amino Acids



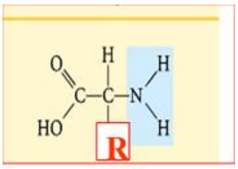
Amino Acid Classification



Amino acids can be classified in 4 ways:
1 .Based on structure
2. Based on the side chain characters
3. Based on nutritional requirements
4. Based on metabolic fate

Based On Structure

Aliphatic amino acids

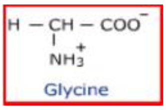


They are classified in three broad categories

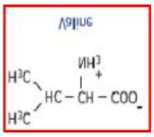
A- Monobasic, monocarboxylic amino acids i.e. neutral or uncharged**:**

They are further subdivided in 5 groups:

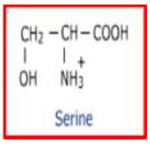
a. Simple amino acids-example: glycine, alanine



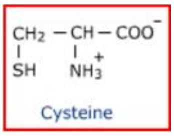
b. Branched chain amino acids-e.g.: valine, leucine, isoleucine



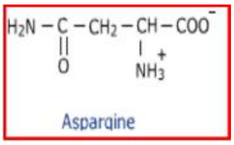
c. Hydroxyl group containing amino acids-e.g.: serine, threonine



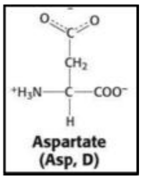
d. Sulphur containing amino acids-e.g.: cysteine, cystine, methionine

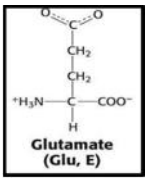


e. Amide group containing amino acids-e.g.- asparagine, glutamine

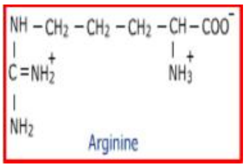


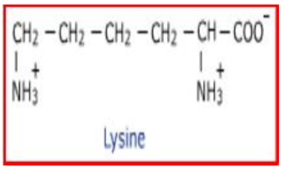
B) Mono amino dicarboxylic acid
Example: aspartic acid, glutamic acid



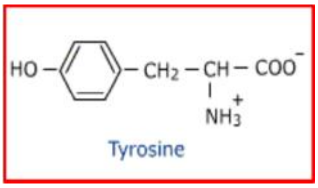


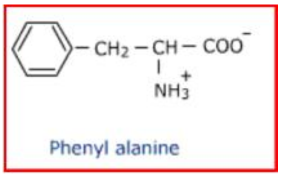
C) Di /poly amino mono carboxylic acid
Example: lysine, arginine



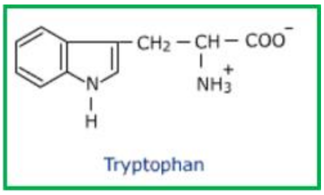


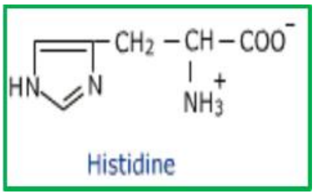
2) Aromatic amino acids
Phenyl alanine and tyrosine



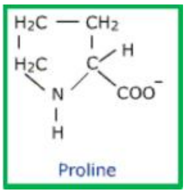


3) Heterocyclic amino acids
Tryptophan and Histidine





4) Imino acids
Proline



5) Derived amino acids (non-protein amino acid) Non-α-amino acids
e.g.: β-alanine, γ-amino butyric acid (GABA), δ-amino Levulinic acid
Derived and Incorporated in tissue proteins:
e.g.: Hydroxy-proline, hydroxy-lysine
Derived but not incorporated in tissue proteins:
e.g.: Ornithine, Citrulline, Homocysteine, Arginosuccinic acid

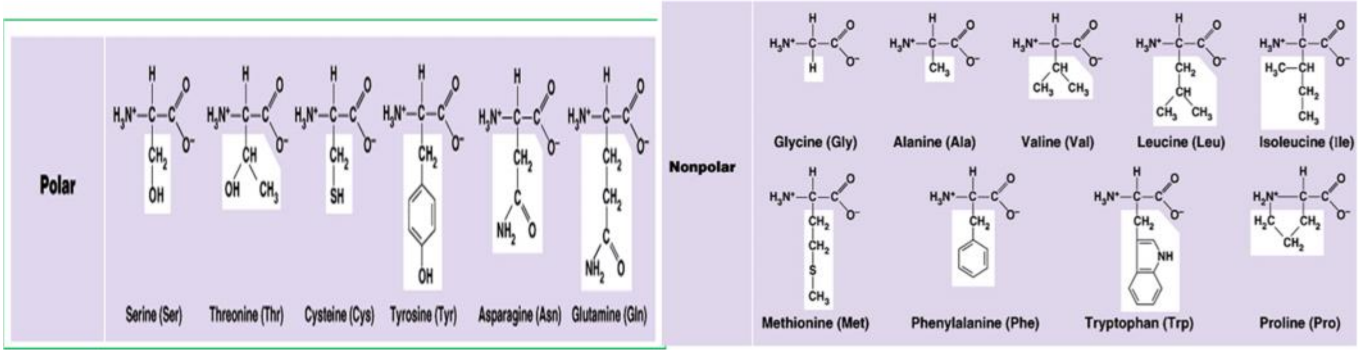
II- Classification according to polarity of side chain (R):

A- **Polar amino acids**: in which R contains polar hydrophilic group so can forms hydrogen bond with H2O. In those amino acids, R may contain:

1- OH group: as in serine, threonine and tyrosine
2- SH group: as in cysteine
3- Amide group: as in glutamine and asparagine
4- NH_2 group or nitrogen act as a base (basic amino acids): as lysine, arginine and histidine
5- COOH group (acidic amino acids): as aspartic and glutamic.

B- Non polar amino acids:

R is alkyl hydrophobic group which can't enter in hydrogen bond formation. 9 amino acids are non-polar (glycine, alanine, valine, leucine, isoleucine, phenyl alanine, tryptophan, proline and methionine)



III- Nutritional classification:

1- Essential amino acids: These amino acids can't be formed in the body and so, it is essential to be taken in diet. Their deficiency affects growth, health and protein synthesis.

2- Semi essential amino acids: These are formed in the body but not in sufficient amount for body requirements especially in children.

Summary of essential and semi essential amino acids:

Villa HM = Ten Thousands Pound

V= valine i= isoleucine l= lysine l= leucine

A = arginine* H= histidine* M= methionine

T= tryptophan Th= threonine P= phenyl alanine

*= arginine and histidine are semi essential

3- Non essential amino acids: These are the rest of amino acids that are formed in the body in amount enough for adults and children. They are the remaining 10 amino acids.

IV- Metabolic classification: according to metabolic or degradation products of amino acids they may be:

1- Ketogenic amino acids: which give ketone bodies. Lysine and Leucine are the only pure ketogenic amino acids.

2- Mixed ketogenic and glucogenic amino acids: which give both ketone bodies and glucose. These are: isoleucine, phenyl alanine, tyrosine and tryptophan.

3- Glucogenic amino acids: Which give glucose. They include the rest of amino acids. These amino acids by catabolism yields products that enter in glycogen and glucose formation

Amphoteric properties of amino acids: that is they have both basic and acidic groups and so can act as base or acid.

Neutral amino acids (monobasic, monocarboxylic) exist in aqueous solution as "Zwitter ion" i.e. contain both positive and negative charge. Zwitter ion is electrically neutral and can't migrate into electric field.

Isoelectric point (IEP) = is the pH at which the Zwitter ion is formed. e.g IEP of alanine is 6

Chemical properties of amino acids:

1- Reactions due to COOH group:

Salt formation with alkalis, ester formation with alcohols, amide formation with amines and decarboxylation

2-Reactions due to NH_2 group: deamination and reaction with Ninhydrin reagent.

Ninhydrin reagent reacts with amino group of amino acid yielding blue colored product. The intensity of blue color indicates quantity of amino acids present. Ninhydrin can react with imino acid like proline and hydroxy proline but gives yellow colour

3. Reaction due to side chain (R)
   a) Million reaction : for Tryosine it gives red colour
   b) Rosenheim reaction : for tryptophan, it gives violet ring
   c) Pauly reaction: for imidazole ring of histidine , give yellow to reddish product
   d) Sakagushi test: for guanido group of arginine and gives red colour
   e) Lead sulphide test (sulfur test): sulfur containing amino acid gives brown colour

:

Properties of amino acids

Physical

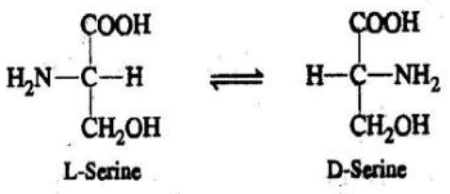
- Amino acids are white crystalline substances.
- Most of them are soluble in water and insoluble in non-polar organic solvents (e.g., chloroform and ether).
- Aliphatic and aromatic amino acids particularly those having several carbon atoms have limited solubility in water but readily soluble in polar organic solvents.
- They have high melting points varying from 200-300oC or even more.
- They are tasteless, sweet or bitter.
- Some are having good flavour. Sodium glutamate is a valuable flavouring agent and is used in the preparation of certain dishes and sauces.

Amphoteric nature of amino acids

- Amino acids are amphoteric compounds, as they contain both acidic (COOH) and basic (NH_2) groups.
- They can react with both alkalies and acids to form salts.
- In acid solution amino acids carry positive charges and hence they move towards cathode in an electric field.
- In alkaline solution, the amino acids carry negative charges and therefore move towards anode.
- When an amino acid is dissolved in water, it exists as inner salt carrying both positive and negative charges.
- This occurs as a result of dissociation of carboxyl group to release the H+ ion, which passes from the carboxyl to the amino group.
- The amino acids possessing both positive and negative charges are called Zwitter ions
- The zwitterion reacts as an acid with a base by liberating a proton (H+) from the NH3+ group and as a result possesses a net negative charge.
- On the other hand, zwitterions reacts with an acid as base, combining with the proton (H+) of the acid resulting in the formation of a compound having a net positive charge.
- These reactions are reversible.
- The pH at which the amino acid has no tendency to move either towards positive or negative electrode is called isoelectric pH or isoelectric point.
- At isoelectric pH, the amino acid molecule bears a net charge of zero

Isomerism

All amino acids except proline, found in protein are α-amino acids because NH_2 group is attached to the α-carbon atom, which is next to the COOH group. Examination of the structure of amino acids reveals that except glycine, all other amino acids possess asymmetric carbon atom at the alpha position. Because of the presence of asymmetric carbon atom, amino acids exist in optically active forms



Structure of D- and L-Serine

'D' and 'L' do not refer to the optical rotation, but to the steric configuration of amino group to the right and left side of the carboxyl group. The direction of optical rotation of amino acid is indicated by the symbol + or -, which follows the designation 'D' or 'L'. The steric configuration and optical rotation of an amino acid may be simultaneously expressed as D (+) or D (-) and L (+) or L (-). L-forms are more common than D-forms and most of the naturally occurring amino acids are L-amino acids

Peptides and Proteins

20 amino acids are commonly found in protein.

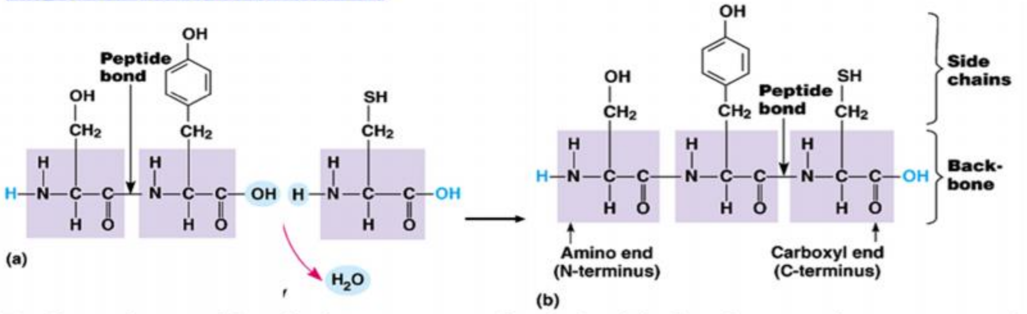
These 20 amino acids are linked together through “peptide bond forming peptides and proteins (what’s the difference?).

-The chains containing less than 50 amino acids are called “peptides”, while those containing greater than 50 amino acids are called “proteins”.

Peptide bond formation:

α-carboxyl group of one amino acid (with side chain R1) forms a covalent peptide bond with α-amino group of another amino acid (with the side chain R2) by removal of a molecule of water. The result is : Dipeptide (i.e. Two amino acids linked by one peptide bond). By the same way, the dipeptide can then forms a second peptide bond with a third amino acid (with side chain R3) to give Tripeptide. Repetition of this process generates a polypeptide or protein of specific amino acid sequence.

Peptide bond formation:



- Each polypeptide chain starts on the left side by free amino group of the first amino acid enter in chain formation . It is termed (N- terminus).
- Each polypeptide chain ends on the right side by free COOH group of the last amino acid and termed (C-terminus).

Examples on Peptides:

1- Dipeptide (tow amino acids joined by one peptide bond):
Example: **Aspartame** which acts as sweetening agent being used in replacement of cane sugar. It is composed of aspartic acid and phenyl alanine.

2- Tripeptides (3 amino acids linked by two peptide bonds).
Example: **GSH** which is formed from 3 amino acids: glutamic acid, cysteine and glycine. It helps in absorption of amino acids, protects against hemolysis of RBC by breaking H_2O_2 which causes cell damage.

3- octapeptides: (8 amino acids)
Examples: Two hormones; oxytocine and vasopressin (ADH).

4- polypeptides: 10- 50 amino acids: e.g. Insulin hormone

FILL UP THE BLANKS

1. The basic functional group of protein is_______
2. Amino acid lowers a net charge of zero at _______
3. Water soluble proteins are_________
4. The S containing amino acids are____,___ and ______
5. Phenyl alanine is a _____ amino acid
6. _____ is an amino acid found in thyroid secretions
7. _____ is an example of basic amino acid_______
8. ______ is a factor for converting nitrogen to protein
9. ____ is an aromatic amino acid
10. The essential amino acid is _______
11. _______ enzyme splits peptides of proteins
12. The linkage formed when two molecules of amino acids linked together is called as ______
13. _______ is a salt soluble protein in cereals
14. The average content of nitrogen in protein is _____
15. The building blocks of proteins are ______
16. An example of mono-dicarboxylic acid is ______
17. Proteins when treated with dilute copper sulfate and made alkaline with sodium hydroxide develop violet colour. This is due to ______
18. Amino acids soluble in water and in ammonia are known as _____
19. Methionine is an amino acid and also contain
20. Proteins contain NH_2 and COOH are called _______
21. Example of fibrous proteins is _____
22. two essential amino acids are _______ and ______
23. The pH at which amino acid has no net charge is called _______
24. Albumin is an example for ____
25. Globulin is an example for ______
26. The bond responsible for the secondary structure of proteins is _____
27. Proteins are least soluble at__ point
28. _____ is an example of pyrimidine bases
29. The condensation products of nucleotides are called _______
30. The purine bases are _______ and _______
31. The type of RNA which gets involved in the transform of amino acid in protein synthesis is ______
32. The nitrogenous base present in cephalin is _______
33. Nucleotide is composed of nucleoside and ______
34. The alcohol soluble proteins are known as _____
35. Nucleotide is _____form of nucleoside
36. _____ is a nitrogenous base present only in DNA and not in RNA
37. In double helical DNA, the number of adenine is equal to ______which are connected by _____ bonds

38. ______ is the only amino acid lacking an asymmetric carbon atom
39. In amino acid, the 2nd and 4th carbon atoms from COOH group are called _____ and _____ respectively
40. Interpolypeptide H- bonding results in ______structure
41. The amino acid which break helical structures is_____
42. Histones contain a high proportion of ______ amino acid and are present in _______
43. _______ and _______ are proteins which are example of primary structure
44. The type of structure found in silk and wool are _____and ______
45. _______ and _______are proteins which are example of quaternary structure
46. _________ is the type of configuration found in amino acids
47. ______ and _______are the colors produced in Millon’s test and biuret test
48. The protein content is very high in ____and low in ______
49. All naturally occurring amino acids belong to ______ series
50 Tyrosine is a _________
51 The limiting amino acid in cereals is _______
52 Nucleotide consists of _______
53 Peptide linkage is present in _______
54 The building blocks of proteins are_______
55 Methionine is a ______
56 The primary function of protein is to provide________
57 Globulin is a ________
58 The neutral amino acid has _________
59 At isoelectric point, the protein solution has_______
60 Aspartic acid is a ________
61 The methionine is limiting in _________
62 Adenosine is a _____
63 DNA contains two polynucleotide chains running in ________
64 The first nucleotide derivatives acts as precursor for the synthesis of carbohydrates is ______
65 Prolamins are soluble in _________
66 Tryptophan belongs to the amino acid group of________
67 Among the three types of RNA present in the living system, the tRNA represents about_______
68 Amphoteric nature of proteins is because of both _________ and _______
69 RNA is usually occurs_______
70 An example for conjugate protein is ______
71 Prolamins are a group of plant _____ protein
72 Uracil base has the related compounds nucleoside and nucleotide as_________
73 Nucleic acid consists of________ sugar
74 Simple proteins on hydrolysis yield ________
75 Amides ($RCONH_3$) are derivatives of _________
76 An example Heterocyclic amino acid is_______

77 Amino acids having both amino and carboxylic group is known as_____
78 Albumins is a ________ protein
79 The covalent linkage in the primary structure of protein is_________
80 Xantro proteins test is used to find out________ amino acid
81 L-aspartyl-L-phenyl alanine is an example of________
82 Alcohol soluble proteins are known as________
83 Glutathione is having ________ peptide bond
84 Cytochrome is an example for ________
85 Pyrimidines bases of RNA are _________ and _________
86 Trypsin is a _______
87 The nitrogenous base absent in RNA is __________
88 Glycine is a optically ______ molecule
89 Nitroprusside test identifies ________
90 Biuret is a _____test for proteins
91 Proteins on complete hydrolysis yield a group of simple organic compounds of low molecular weight called as ______
92 The non-amino acid portion of a conjugate protein is called as ______
93 __________ an example of phospho proteins
94 Cytochromes are example of _____
95 __________ is a transport protein
96 The first amino acid isolated was________
97 ______ is a non-protein amino acid
98 The secondary structure of proteins is stabilized by_______
99 _________ represents the quaternary structure of proteins
100 The common name for 6 amino purine is________
101 Ribosome is an example for _______ proteins
102 Egg albumin is an example for______
103 The amino acid which is most likely to disturb alpha helix is _______
104 Amino acid can form disulphide bonds is ________
105 The repeating units in protein is _________
106 The primary structure of proteins represents ______
107 Peptide bond is a ________
108 The first protein sequenced by Fredrick sanger is ________
109 A dipeptide bond has ________
110 Myoglobin is a ____
111 Alpha helix has___________
112 The 3D structure of protein can be determined by _______ and _______
113 _________ is both glycogenic and ketogenic amino acid
114 _______- amino acid is precursor for niacin
115 The first amino acid formed in a polypeptide chain is _______
116 The amino acid having buffering capacity is _________
117 The 21st amino acid is _______

118 The limiting amino acid in pulses is _______

119 All the amino acids found in protein except Proline contains ____

120 Ionic state of amino acids depends upon_____

121 _________ has an imino (>NH) group instead of amino group (-NH_2)

122` Compound that give negative test with Ninhydrin and positive test with benedict’s Solution.is ____

123 Proteins when heated with concentric acid give a yellow color is provided by ___test

124 Gene is a segment of ________

125 Nucleic acids are polymers of ______

126 The reason for double helical structure of DNA is operation of _______

127 The base is present in RNA but not in DNA is ________

128 Each polypeptide in a protein has amino acids linked with each other in a specific sequence. This sequence of amino acids is said to be _________

129 In fibrous proteins, polypeptide chains are held together by______

130 DNA and RNA contain four bases each._________ base is not found in RNA

131 .Amino acids and Protein tests include ________

132 Two amino acids of the standard 20 contain sulfur atoms. They are___ and _____

133 Example of non-protein amino acid is ________

134 Amino acid which synthesizes many hormones is: _______

135 An amino acid not found in proteins is_____

136 Sakaguchi’s reaction is specific for________

137 The most dominant amino acid present in proteins is ___

138 GAG triplet codon and is a __________

139 Beta pleated sheet is a _______ protein structure

140 The partial or complete disorganization of a protein’s three-dimensional shape is called _____

ANSWER

Sl.No	Answer	Sl.No	Answer
1	Amino	34	Prolamins
2	Iso electric point	35	Phosphorylated
3	Albumins	36	Thymine
4	Methionine, Cystine, Cysteine	37	Thymine, H bonding
5	Aromatic	38	Glycine
6	Insulin	39	β,€
7	Lysine, Arginine	40	Secondary
8	6.25	41	Proline
9	Phenyl alanine,Tryosine	42	Basic, Nuclei
10	Histidine	43	Myoglobin, Insulin
11	Trypsin, Pepsin	44	α- secondary, β-secondary
12	Peptide	45	Haemoglobins, Myoglobin

13	Albumins	46	L type
14	16%	47	Red, violet
15	Amino acids	48	Pulses, Cereals
16	Aspartic acid and glutamic acid	49	Aliphatic
17	Biuret test	50	Aromatic amino acid
18	Protamines	51	Lysine
19	Sulphur	52	Nucleoside + H_2PO_4
20	Simple proteins	53	Proteins
21	Collagens/ Elastin/ Keratins	54	Amino acids
22	Histidine ,Lysine	55	S containing amino acid
23	Isoelectric point	56	Amino acids
24	Salt soluble	57	Water insoluble proteins
25	Storage	58	One NH_2 and one COOH group
26	H bonding	59	Zero charge
27	Isoelectric	60	Dicarboxylic acid
28	Thymine/ Cytosine/ Uracil	61	Pulses
29	Nucleic acid	62	Adenine + ribose
30	Adenine, Guanine	63	Opposite
31	t-RNA	64	UDPG
32	Serine/Ethanolamine	65	60-90% alcohol
33	Orthophosphoric acid	66	Heterocyclic amino acid
67	15%	106	Sequence of amino acids
68	Both basic and acidic properties	107	Covalent bond
69	single stranded helix	108	Insulin
70	Nucleoproteins	109	two amino acids
71	Simple	110	hemeproteins
72	Uridine and uridylic acid	111	Coil of amino acid chain
73	Four pentose	112	NMR Spectroscopy and X-Ray Crystallography
74	α amino acid	113	Isoleucine, Phenylalanine, Tryptophan, Tyrosine and Threonine.
75	Amino acids	114	Tryptophan
76	Proline	115	Methionine
77	Zwitter ions	116	Histidine
78	Simple proteins	117	seleno cystenine
79	Peptide bond	118	Lysine
80	Aromatic amino acid	119	Amino
81	Dipeptide	120	pH
82	Prolamins	121	NH_2
83	Penta	122	A monosaccharide
84	Chromo proteins	123	Xanthoproteic acid
85	Cytosine and Uracil	124	DNA
86	Carboxy peptidase	125	nucleotide
87	Thiamine	126	hydrogen bonding
88	Inactive	127	uracil

89	S-amino acid	128	Amide bond
90	General	129	disulphide cross bridges
91	Amino acid	130	Thymine
92	Prosthetic group	131	Six
93	Casein	132	Methionine and cysteine
94	Haemoproteins	133	carnitine, GABA, Levothyroxine)
95	Carrier protein	134	Tyrosine
96	Glycine	135	8
97	Beta alanine	136	Test for arginine
98	H bonding	137	Glycine
99	Haemoglobins	138	Protein
100	Adenine	139	Tertiary
101	Nucleoprotein	140	Denaturation
102	Storage protein		
103	Proline		
104	cysteine		
105	Amino acids with different R		

DEFINITIONS

1. Protein: Any of a class of nitrogenous organic compounds which have large molecules composed of one or more long chains of amino acids and are an essential part of all living organisms,
2. Amino acid: A simple organic compound containing both a carboxyl (—COOH) and an amino (—NH2) group.
3. Essential amino acid: An essential amino acid, or indispensable amino acid, is an amino acid that cannot be synthesized de novo (from scratch) by the organism, and thus must be supplied in its diet
4. Peptide bond: A peptide bond is a chemical bond formed between two molecules when the carboxyl group of one molecule reacts with the amino group of the other molecule, releasing a molecule of water
5. Zwitterion: Zwitterion is the dipolar form of an amino acid which occurs when H + ion is transferred from an acid group to an amine group.
6. Simple proteins: Simple proteins yield on hydrolysis, only amino acids.
7. Salting out: Process where albumins may be precipitated out from solution using high salt concentration
8. Conjugated protein: These are simple proteins combined with some non-protein substances known as prosthetic groups
9. Denaturation: Denaturation is a process in which proteins or nucleic acids lose the quaternary structure, tertiary structure and secondary structure which is present in their native state, by application of some external stress or compound such as a strong acid or base, a concentrated inorganic salt, an organic solvent
10. Biuret reaction: A compound, which is having more than one peptide bond when treated with Biuret reagent, produces a violet colour. This is due to the formation of coordination complex between four nitrogen atoms of two polypeptide chains and one copper atom

11. Amphoteric nature of proteins: Amino acids due to the presence of their ionizable α-amino and α-carboxylic group can act sometimes as acids and sometimes as bases depending on the pH of their media.
12. Genetic code: The means by which DNA and RNA molecules carry genetic information in living cells.
13. Nucleotides: A compound consisting of a nucleoside linked to a phosphate group. Nucleotides form the basic structural unit of nucleic acids such as DNA.
14. Nucleoside: A compound (e.g. adenosine or cytidine) consisting of a purine or pyrimidine base linked to a sugar.
15. DNA: Deoxyribonucleic acid, a self-replicating material which is present in nearly all living organisms as the main constituent of chromosomes. It is the carrier of genetic information.
16. RNA: Ribonucleic acid, a nucleic acid present in all living cells. Its principal role is to act as a messenger carrying instructions from DNA for controlling the synthesis of proteins

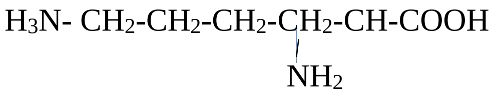
WRITE SHORT NOTES ON:

1. **Essential Amino Acids**

   Essential amino acids are those which cannot be synthesized in the body in adequate amounts and must be supplied from outside as part of food. The presence of these amino acids is essential for growth of the young and maintenance of the adult. The essential amino acids are 1) lysine 2) Tryptophan 3) Histidine 4) Leucine 5) Phenyl alanine 6) Iso-leucine 7) Threonine 8) Methionine 9) Valine and 10) Arginine

2. **Basic Amino Acids**

   Based on the composition of the side chain, the twenty amino acids may be grouped in to 8 categories. Out of this is basic amino acids where it possess amino group in side chain eg., Lysine and arginine Basic amino acids are polar and positively charged at pH values below their pK_a's, and are very hydrophilic. Even though the basic amino acids are almost always in contact with the solvent, the side chain of lysine has a marked hydrocarbon character, so it is often found near the surface, with the amino group of the side chain in contact with solvent.



$$H_3N\text{- }CH_2\text{-}CH_2\text{-}CH_2\text{-}\underset{\overset{|}{NH_2}}{CH_2}\text{-}CH\text{-}COOH$$

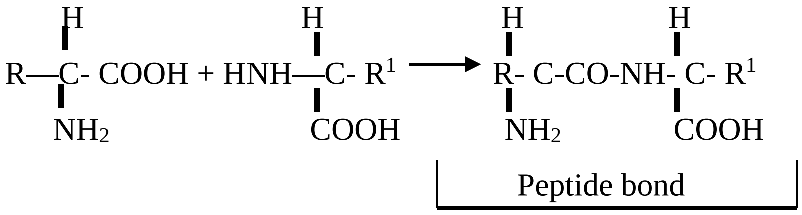
Lysine (Lys)

3. **Biological properties of Proteins**

   Proteins are of utmost significance to biological systems. These are most critical to life and perform various functions. Many proteins act as a catalyst to enhance the rate of chemical reactions. The fibrous proteins act as structural proteins holding the skeletal elements. Proteins also perform transport functions. Various proteins are known to be hormone which regulates the growth of the plants and animals. Some proteins like snake venom act as toxic one which degrade enzymes. Regulatory proteins regulate cellular or metabolic activities. Proteins defend against other organisms. Proteins provide nutrition to growing embryos and store ion which act as storage proteins**,**

4. Peptide bonds

The amino acid unit is linked together through the carboxyl and amino group to produce the primary structure of the protein chain. The bond between two adjacent amino acids is a special type of amide bond known as the peptide bond and the chain thus formed is called as peptide chain. A single peptide bond is formed when two amino acids involved in a reaction and the carboxyl group of one amino acid react with the amino group of another amino acid, with elimination of one molecule of water. Depending up on the number of amino acid involved in the reaction, the compound is known as Dipeptide (two amino acid with one peptide bond), a tripeptide (three amino acid with two peptide bond) or a polypeptide (many (n…) amino acids with n-1 peptide bonds) where n is number of amino acids. When a polypeptide chain is formed, one free amino and one carboxyl group is left at the two different ends. The free amino end is called as N terminal and the free carboxyl end is called as C-terminal



$$R-\overset{H}{\underset{NH_2}{C}}-COOH + HNH-\overset{H}{\underset{COOH}{C}}-R^1 \longrightarrow \underbrace{R-\overset{H}{\underset{NH_2}{C}}-CO-NH-\overset{H}{\underset{COOH}{C}}-R^1}_{\text{Peptide bond}}$$

Dipeptide

5) Nucleic acids

The nucleic acids are biopolymers of high molecular weight with mono nucleotide as their repeating units. The nucleic acid contains carbon, hydrogen, oxygen, nitrogen and phosphorus. The nucleic acids are of considerable importance in biological systems. They are two types 1) RNA (Ribose nucleic acid) 2) DNA (Deoxyribose nucleic acid). The basic chemical subunits of the nucleic acids are nucleotides. The nucleotides are made of 3 compartments 1) A heterocyclic ring containing nitrogen and nitrogenous base 2) a five carbon pentose sugar 3) A phosphate group. The bases found in nucleic acid are of two types- Purines and Pyrimidines. Adenine and guanine are purines and cytosine and Uracil and thymine are pyrimidines bases. The nucleotides found in nucleic acids are much fewer in number than the amino acids. Ribose nucleic acid is also of common occurrence in plants as well as animals. It is of three types 1) r-RNA,2) m-RNA 3) t-RNA .RNA is intimately associated with protein synthesis and are found chiefly in nucleolus in the nucleus. (Ribosomes and cytoplasm). Besides this, RNA also occurs in mitochondria and chloroplast. All plant virus contains RNA. DNA which is the seat of all the hereditary characters is chiefly found in chromatin in the nucleus where it is associated with proteins called as histones. Some DNA is also found in mitochondria and chloroplast. DNA is a poly nucleotide having a specific sequence of deoxyribonucleotide units covalently joined through 3', 5'phosphodiester bonds, two of the polymers wind around each other, like the outside and inside rails of winding staircase (double helical compound).

6 **Lipoproteins**

It is a conjugate protein. They are complexes of lipids and proteins. The common lipids which are found as prosthetic groups are lecithin and cephalin. They are insoluble in water and are found in the membrane, nucleus and lamellas of the chloroplast. A lipid protein aggregate that serves to carry water- insoluble lipids in the bood. Egg yolk contains lipoproteins.

7) **RNA**

Ribose nucleic acid (RNA) is a single stranded structure consisting of only one polynucleotide chain. RNA consists of the following bases 1) Adenine and Uracil 2) Guanine and Cytosine. The ratio of purines and pyrimidines bases is not 1:1. The pentose sugar is β-D-ribose. The size of the RNA molecule is very small in comparison to the DNA molecule. Molecular weight of RNA may range from several thousands to some lakhs. There are 3 types of RNA's in plant cells.

a) Messenger RNA (mRNA)

Molecular weight of mRNA is higher among different types of RNA's. m-RNA is synthesized in nucleolus and after taking genetic information from DNA goes into cytoplasm and helps in the formation of specific protein. Sequence of 3 bases or nucleotides in m-RNA molecule constitutes a codon.

b) **Ribosomal RNA(r-RNA)**

r-RNA is found in Ribosomes which act as template for the synthesis of protein

c) **Transfer RNA (t-RNA)**

Basic structure of all t-RNA molecules is on the clover leaf pattern. T-RNA are found in cytoplasm and consists of only about 80 bases. t-RNA contains many unusual bases and nucleosides. There are different t-RNA molecules with specific anticodons to pick up specific amino acids. However many t-RNA may be specific to a particular amino acids are single t-RNA species may recognize several amino acids.

8) **Chemical properties of proteins**

1) **Hydrolysis:**

a) By acidic agents- Proteins up on hydrolysis with concentrated HCl at 100- 110^{o}C for 6 to 20 hours yield amino acid in the form of their hydrochlorides

b) By alkaline agents – Proteins may also be hydrolyzed by 2N NaOH

c) By proteolytic enzymes- Certain proteolytic enzymes like pepsin and trypsin hydrolyze the proteins under certain conditions of temperature and acidity

2) **Reaction involving COOH group**

a) **Reaction with alkalies (salt formation**) - The carboxylic group of amino acids can release H^+ ion with the formation of carboxylate (COO^-) ions. These may be neutralized by Na, Ca to form salts

$R-CHNH_2-COOH + NaOH \rightarrow R-CHNH_2-COONa + H_2O$

b) **Reaction with alcohol (Esterification**) - Proteins reacts with alcohol to form Corresponding esters. These esters are volatile than amino acid

$R-CHNH_2-COOH + C_2H_5OH \rightarrow R-CHNH_2-COO.\ C_2H_5 + H_2O$

c) **Reaction with amines** – Amino acid reacts with amines to form amides

$R-CHNH_2-COOH + NH_2-R^1 \rightarrow R-CHNH_2-CO-NH-R^1 + H_2O$

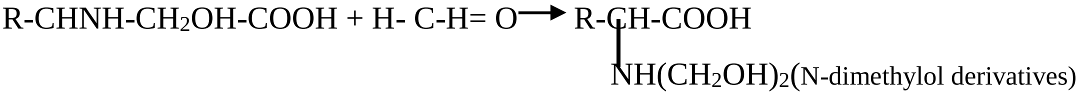
3) **Reaction involving amino group**:

a) Reaction with mineral acids (Salt formation)- When free amino acids or proteins are treated with mineral acids like HCl, the acid salts are formed

$R-CHNH_2-COOH + HCl \rightarrow R-\underset{\substack{|\\ NH_2HCl}}{C}H-COO.H$

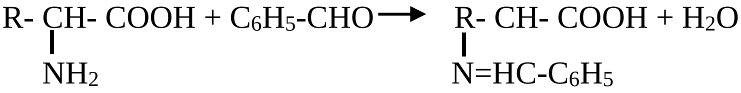
b) **Reaction with formaldehyde**- With formaldehyde, the hydroxy-methyl derivatives are formed. These derivatives are insoluble in water and resistant to microbial attack

$R-CHNH_2-COOH + H-C-H=O \rightarrow R-CHNH-CH_2OH-COOH$ (N monomethylol derivatives)



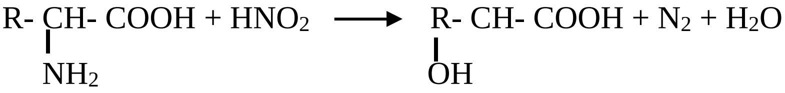
$R-CHNH-CH_2OH-COOH + H-C-H=O \rightarrow R-\underset{\substack{|\\ NH(CH_2OH)_2}}{C}H-COOH$ (N-dimethylol derivatives)

c) **Reaction with benzaldehyde** (Schiff's base are formed)



$R-\underset{\substack{|\\ NH_2}}{C}H-COOH + C_6H_5-CHO \rightarrow R-\underset{\substack{|\\ N=HC-C_6H_5}}{C}H-COOH + H_2O$

d) **Reaction with nitrous acid** – The amino acids react with HNO_2 to liberate N_2 and to produce the corresponding α- hydroxy alcohol



$R-\underset{\substack{|\\ NH_2}}{C}H-COOH + HNO_2 \rightarrow R-\underset{\substack{|\\ OH}}{C}H-COOH + N_2 + H_2O$

e) **Reaction with acylating agents (acylation**) – Acylation is brought about by many acid chlorides

4) **Reaction involving both COOH and NH_2 groups**

a) **Ninhydrin test**- It is powerful oxidizing agent and causes oxidative decarboxylation of α- amino acids producing CO_2, NH_3 and aldehyde with one less carbon atom than the parent amino acids. The reduced Ninhydrin then react with the liberated NH_3 forming blue colored complex

9) **Conjugate proteins**

These proteins on hydrolysis yields amino acid plus some non-amino acid part called as the prosthetic group. Depending up on the nature of the prosthetic group associated with them, conjugate proteins are classified as follows.

a) Nucleoproteins (histones)- These are associated with nucleic acids
b) Glycoproteins- These are associated with some carbohydrates
c) Chromoprotein – These proteins are associated with some coloring matter eg. Chlorophyll, carotenoids
d) Lipoproteins – These are associated with lipids or fatty substances eg., cephalin, lecithin
e) Iron-prophyrin proteins – These are associated with iron-prophyrin compound eg., cytochromes
f) Simple metal containing proteins- These are associated with some metals directly eg., ferredoxin
g) Flavoprotein – These are associated with some Flavin compounds eg., FAD

10) **Color reaction of proteins**

a) **Biuret test**: compounds having peptide bonds produce a purple color on reaction with alkaline copper sulfate solution. The color deepens as the number of peptide bonds increases and proteins produce blue- violet
b) **Xanthoproteic test**: Yellow color develops on boiling proteins with concentrated nitric acid due to the presence of benzene rings. Thus reactions is due to the nitration of the phenyl rings (tyrosine)
c) **Millon's test**: Red color develops when proteins are heated with silver nitrate in nitric acid
d) **Nitroprusside test:** Red color develops with sodium Nitroprusside in dilute ammonia (Cysteine specific)
e) **Hopkins- Cole test**: Violet rings develop on addition of concentrated sulphuric acid at the junction of proteins and glyoxylic acid solution

11) **Classification of proteins based on solubility**

Simple proteins are those which on hydrolysis yields only amino acids. On the basis of solubility property, simple proteins are classified as

a) Albumins – soluble in water and salt solution
b) Globulins- sparingly soluble in water but soluble in salt solution
c) Prolamins- soluble in 70-80% alcohol but insoluble in water and absolute alcohol
d) Glutelins- Insoluble in water, salt and alcohol but soluble in acid and alkali
e) Scleroproteins- insoluble in aqueous solvents (found in animals only)

12) Distinguish between RNA and DNA

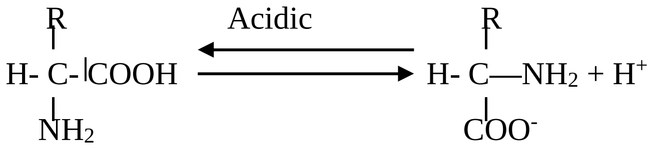
Sl. no	DNA	RNA
1	Visually double stranded structure	Single stranded structure
2	Bases are adenine, guanine, Uracil and cytosine	Bases are adenine, guanine, thymine and cytosine
3	Pentose sugar is β-D-Ribose	Pentose sugar is β-D-2 deoxy ribose
4	Found only in the chromatin of the cell nucleus	Found in cell cytoplasm
5	Never present in free state	May be present in free state
6	It is alkali stable	It is alkali labile
7	It act as template for its synthesis	It does not act as template for its synthesis
8	It undergo mutation	It does not undergo mutation
9	It is usual genetic material	It is genetic material for some viruses only
10	It stains green with dye pyronins	It stained red with pyronins

13) Secondary structures

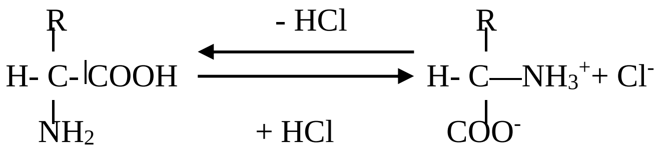
Polypeptide chain of the protein molecule is held in a coiled or helical shape by hydrogen bonds which are established in between peptide linkages. This coiled or helical shape of polypeptide chain constitutes the α- helix or secondary structure of proteins. Based on the nature of hydrogen bonding, two regular types of secondary structure in proteins- alpha helical and beta pleated sheet. Although hydrogen bonds are very weak but when they are present in very large number all along the backbone of the polypeptide chain, they reinforce one another to stabilize the helical structure eg., silk and wool

14) Amphoteric nature of proteins

Proteins act as acid or alkali or both. These migrate in an electric field and the direction of migration depends on the net charge possessed by the molecule. The net charge is influenced by the pH value. Each protein has a fixed value of isoelectric point at which it will move in an electric field. Isoelectric point is the pH value at which the number of cations and anions are equal. Thus, at isoelectric point, the net charge of a protein is always zero. But the total charge of proteins at this point is always maximum. The proteins are dipolar ions or Zwitter ions. At pH value less than the isoelectric point, the proteins will have net positive charge and as a cation will migrate towards negative pole. Similarly, at pH value higher than isoelectric point, the proteins will have a net negative charge and as a anion will move towards positive pole



$$H\text{- }\overset{R}{\underset{NH_2}{C}}\text{-}COOH \;\underset{\longrightarrow}{\xleftarrow{\text{Acidic}}}\; H\text{- }\overset{R}{\underset{COO^-}{C}}—NH_2 + H^+$$



$$\underset{NH_2}{\overset{R}{\text{H-C-}}}\text{COOH} \underset{+\,HCl}{\overset{-\,HCl}{\rightleftharpoons}} \underset{COO^-}{\overset{R}{\text{H-C}}}\text{—}NH_3^+ + Cl^-$$

Basic

15) **Genetic code**

The language of the genes. The set of triplet code words in DNA (or m-RNA) is the code for the amino acids of proteins. Of the 64 possible codons, 61 are the codes for the amino acids and the remaining being termination codons that are not translated. All the genetic information is encoded in DNA which produces hereditary characters in all the living beings. The sequence of 3 nucleotides in polynucleotide chains of DNA molecule is called as triplet code. In m-RNA molecule, this sequence of 3 nucleotides is complimentary to the sequence of nucleotides in DNA and is called as codon. There are different codons for different amino acids. The latter are incorporated in a particular protein through specific t-RNA molecules with specific anticodons. Anticodon is complimentary to codon. A list of all the codons which specify amino acid constitutes the coding dictionary or genetic code. Genetic code can degenerate. It is not overlapping. The first and second letters of codon are more important than third in specifying an amino acid.

16) **Denaturation of proteins**

Denaturation refers to the changes in the properties of a protein. In other words, it is the loss of biological activity. Denaturation may be brought about by a variety of agent's viz., physical agents (heat, cooling and freezing, rubbing, UV ray) and chemical agents (ionizing radiation, organic solvents). Denaturation mainly leads to the unfolding of the peptide chain, thus causing disorganization of the internal structure of proteins. Denaturation of proteins causes decrease in solubility, cessation of biological activity, decrease in size and shape of molecule, change in optical rotation in the direction of increased levorotations, alteration in the surface tension and loss of antigenicity. Some proteins, when denatured cannot be brought back to the original state. Such cases Denaturation is irreversible. The process of regaining normal protein properties by denatured protein is called as Denaturation or refolding.

17. **Solubility of proteins**

The solubility of proteins is markedly influenced by pH. Solubility is lowest at isoelectric point and increases with increasing acidity or alkalinity. This is because when the protein molecules exists as either cations or anions, repulsive forces between ions are high since all the molecules possess excess charges of the same sign. Thus, they will be more soluble than in the isoelectric state. Globulins are sparingly soluble in water but their solubility is greatly increased by the addition of neutral salts like NaCl. It is referred to as salting in effect. Proteins are precipitated from aqueous solution by high concentration of neutral salts. This is the salting out process. Divalent and trivalent ions are more effective than univalent ions.

ESSAY TYPES

1 **How are proteins classified based on solubility and functions?**

Proteins are classified based on solubility as follows

a) **Albumins**- It is soluble in water and dilute solutions of acids, bases and salts. It is precipitated with neutral salts, coagulated by heat. It is widely distributed in nature and more abundant in seeds.

b) **Globulins-** It is widely distributed in nature, insoluble in water, soluble in salt solution

c) **Glutelins**- Isolated from plant seeds, insoluble in water, salt solution but soluble in acid and alkali, coagulated by heat eg., glutenin(Wheat)

d) **Prolamins**- It is insoluble in water and dilute salt solutions but soluble in dilute acids and alkalies and also in 60-80% alcohol solution, not coagulated by heat eg., Hordein of oats

e) **Protamines**- These are basic proteins and occur almost entirely in animals. It is soluble in water, not coagulated by heat, form salt with acid and alkali

f) **Scleroproteins**- it occur almost entirely in animals. It is insoluble in water , dilute solutions of acid, bases and salts, 60-80 % alcohol

Proteins based on function

1. **Enzyme proteins**- It is highly specialized protein having catalytic activity- the enzymes. It catalyzes a variety of reactions. Most of the enzymes are globular proteins eg., urease, amylase and catalase
2. **Structural proteins**- It is inert to biochemical reactions. The cell wall and primary fibrous constituents of the cell have structural proteins eg collagen, keratin and fibroin
3. **Transport or carrier proteins –** Certain proteins in animals are involved in the transport of many essential biological factors to various parts of the organisms eg., Haemoglobins
4. **Nutrient or storage proteins**. Ovalbumin is the major protein of egg white. The milk protein casein stores amino acids. The seeds of many plants store nutrient proteins required for the growth of the germinating seedlings
5. **Contractile or motile proteins**- Some protein endow cell and organisms with the ability to contract to change shape or to move about eg., actin and myosin
6. **Defense proteins –** Many proteins defend organisms against invasion by other species or protect from injury eg., Antibodies
7. **Regulatory proteins**- Some proteins help regulate cellular or physiological activity. Among them are many hormones eg., insulin- regulate sugar metabolism
8. **Toxic proteins**- Some proteins acts as a toxic substance eg., snake venom

2) **Describe the protein biosynthesis process or describe activation, initiation, elongation and termination steps in protein synthesis**

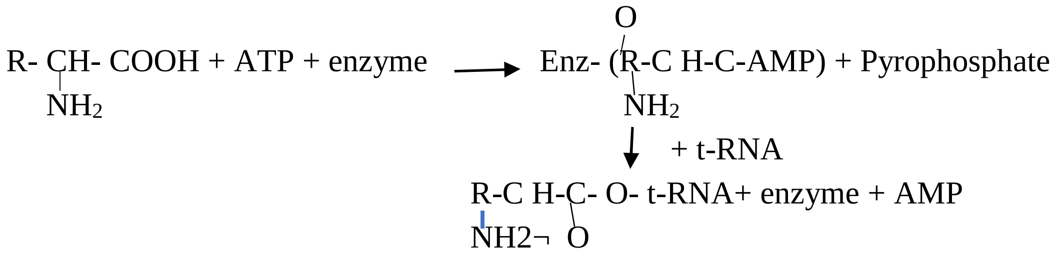
Protein synthesis in plants is under the direct control of DNA. The various steps involved in protein synthesis in plants is as follows

1) DNA in the nucleus directs the synthesis of m-RNA and provides with it with necessary genetic information in the form of codons for the formation of specific proteins. This process is called as **transcription.** m-RNA is synthesized in the presence of enzyme RNA polymerase

2) m-RNA molecules moves into the cytoplasm where it causes the formation of specific t-RNA molecules having specific anticodons complementary to codons

3) m-RNA becomes associated with the ribosomes which act as template for protein synthesis. At this template energy is supplied by GTP (Guanosine phosphate)

4) t-RNA molecules picks up a specific amino acid by its C_C_A end after the amino acid has been activated by ATP in the presence of specific enzyme (activation of amino acid)



$$\underset{NH_2}{R\text{- }\underset{|}{C}H}\text{- COOH + ATP + enzyme} \longrightarrow \text{Enz- (}\underset{NH_2}{\overset{O}{R}}\text{-C H-C-AMP) + Pyrophosphate}$$

$$\downarrow \quad \text{+ t-RNA}$$

$$\underset{NH_2}{\text{R-C H}}\text{-}\underset{O}{C}\text{- O- t-RNA+ enzyme + AMP}$$

Amino acid attached to t-RNA

5) In bacteria E coli the 70S ribosomes dissociates into 30S + 50S subunits

6) 30 S subunits of the ribosomes recognizes the 5^1 terminal end of m-RNA from where the protein synthesis ie., the formation of polypeptide chain starts.

7) 30S subunits also recognize t-RNA amino acid complex which is then transferred to 50S subunit

8) 30S and 50S subunits of the ribosomes unit in the presence of Mg^{2+} ions and become associated with m-RNA

9) t-RNA amino acid complex attached to the ribosome is placed opposite to the specific codon on m-RNA molecule due to the presence of its complementary anticodons in t-RNA molecule

10) Ribosomes and the m-RNA move relative to each other. When ribosome reach second codon, another specific t-RNA amino acid complex is attached to the ribosomes so that its anticodon is placed opposite its complementary codon

11) A peptide bond is established between the carboxylic and amino group of two amino acid in the presence of the enzyme peptide synthetase. The preceding t-RNA molecule is released into the cytoplasm to become charge again with amino acid

12) In the same way, during the movement of the m-RNA and ribosome, a number of specific amino acids are added one after another into the growing polypeptide chain

13) Polypeptide chain remain attached to the 50S subunit till it is complete

14) Termination of the polypeptide chain takes place when ribosomes come over the non-sense codon in m-RNA molecule during its movement against m-RNA. Nonsense or chain termination codons are UAA, UAG, UGA

15) Polypeptide chain produced on the same m-RNA may be of one or more types depending upon whether m-RNA is monocistronic and polycistronic
16) After formation of polypeptide chains, the m-RNA disintegrates and ribosome again dissociates into its subunits which are cycled back into the cytoplasm
17) The process in which a particular nucleotide sequence on m-RNA is translated into a particular amino acid sequence with the help of the ribosomes is called as **translation**

3) Describe amino acid classification with one example in each group

a) **On the basis of the composition of the side chain or R group**

1) **Simple amino acids –** No functional group in the side chain eg., Glycine, alanine
2) **Hydroxy amino acids**- These contain hydroxy group in their side chain- eg. Serine and Threonine
3) **S containing amino acids**- These possess a sulfur atom in the side chain. Eg. Methionine, Cystine
4) **Acidic amino acids**- These have carboxyl group in the side chain eg. Aspartic acid, glutamic acid
5) **Amino acid amides**- These are derivatives of acidic amino acids in which one of the carboxyl group has been transformed into an amide group eg. Asparagine, Glutamine
6) **Basic amino acids**- These possess an amino group in the side chain eg. Lysine, arginine
7) **Heterocyclic amino acids-** These amino acids have their side chain a ring which possess at least one atom other than cation- Tryptophan, Histidine
8) **Aromatic amino acid-** They have benzene ring in the side chain eg. Phenylalanine, Tryosine

b) **On the basis of number of amino and carboxylic group**

1) **Mono amino-mono carboxylic amino acid**- eg., Glycine, alanine
2) **Mono amino dicarboxylic amino acid** –eg. Aspartic acid, glutamic acid
3) **Diamino mono carboxylic amino acid**- eg, lysine, arginine

c) **On the basis of the polarity of the side chain or R group**

1) **Amino acid with non-polar R group**- This group includes five amino acids with aliphatic R groups (alanine, valine, leucine, isoleucine, and Proline), two with aromatic rings (Phenylalanine, Tryptophan) and one S containing amino acid (Methionine). The R group in this category of amino acid is hydrocarbon in nature, hence hydrophobic

2) **Amino acids with polar but uncharged R group**- The r groups of these amino acids are more soluble in water ie., more hydrophilic. This category includes 7 amino acids- glycine, serine, Threonine, tyrosine, cysteine, asparagines, and glutamine. The polarity of these amino acids may be due to either a hydroxyl group of a sulfide group or an amide group.

c) **Amino acids with positively charged R groups**- These are diamino mono carboxylic acids, in other words, their side chain contains an extra amino group which imparts basic properties to them. Eg., Lysine , arginine and Histidine

4) **Describe in detail about different types of ribonucleic acids**

Ribonucleic acids (RNA) are along unbranched macromolecule consisting of nucleotides joined by 3'-5' phosphoester bonds. The number of ribonucleotides in RNA ranges from as few as 75to many thousands. Three general types of RNA are found a) Ribosomal b) Transfer c) Messenger

a) Ribosomal RNA- It is the most stable form of RNA and is found in Ribosomes. It has the highest molecular weight, Ribosomal RNA is the most abundant of all types of RNA's and makes up about 80% of the total RNA of a cell. Ribosomal RNA represents about 40 to 60% of the total weight of ribosomes. r-RNA is found in ribosomes which act as template for the synthesis of proteins

b) Messenger RNA (m-RNA). Molecular weight of m-RNA is higher among different types of RNA. M-RNA is synthesized in nucleolus and after taking genetic information from DNA goes into the cytoplasm and helps in the formation of specific proteins. Sequence of 3 bases or nucleotides in m-RNA molecules constitutes a codon

c) Transfer RNA or soluble RNA(t-RNA or s-RNA)

These are comparatively very small with a molecular weight of about 25000. Basic structure of all t-RNA molecules is of clover leaf pattern. t-RNA are found in cytoplasm and consisted of about 80 bases. It constitutes 15-20% of total RNA. t-RNA contain many unusual bases and nucleosides. All t=RNA molecules contain guanine (G) at 5 'end and 3 end always in base sequence. During protein synthesis these ends in fact picks up the amino acids and transfer it to the growing polypeptide chain and hence these RNA's are called t-RNA. These t-RNA is also called as s-RNA or soluble RNA because they are soluble in IM NaCl. There are different t-RNA molecules with specific anticodons to pick up specific amino acids. However many t-RNAs may be specific to a particular amino acid or a single t-RNA may be recognized several amino acids.

5) **Write on chemistry of nucleotides**.

Nucleic acid is unbranched long chain polymers of nucleotides. Each nucleotide consist of 3 parts

a) A purine or pyrimidine base
b) An aldopentose sugar
c) Orthophosphoric acid

Nitrogenous bases:

Two types of nitrogenous bases are found in all nucleic acid. These are derivatives of purines and pyrimidines

a) Pyrimidines derivatives- The common pyrimidine derivatives found in nucleic acid are

1) Uracil- found in RNA only, white and crystalline
2) Thymine- found in DNA only
3) Cytosine- found both in RNA and DNA, white, crystalline

b) Purine derivatives- These are all derived from their parent compound purines which contain six membered pyrimidine ring fused to the five member imidazole ring.

1) Adenine- It is found in both RNA and DNA, white, crystalline base

2) Guanine- It is found in both RNA and DNA, colorless, insoluble, crystalline

Pentose sugar

The two types of nucleic acids are distinguished primarily by the basis of the 5 carbon keto sugar or pentose which they possess. One possesses D-2- deoxy ribose, hence the name DNA, while the other contain D- ribose, hence the name RNA. Both these sugars in nucleic acids are present in the furanose form and are β configuration. The two sugars may be different by means of specific color reactions. Ribose reacts with orcinol in hydrochloric acid solution containing ferric chloride. Deoxy ribose reacts with diphenylamine in acid solution. An important property of the pentoses is their capacity to form esters with phosphoric acid

Phosphoric acid

The molecular formula of phosphoric acid is H_3PO_4. Combination of a base and pentose sugar is called as **nucleosides.** If one of the C atom of the pentose sugar in nucleoside is phosphorylated is called as **nucleotide.** The nucleotides are joined together by phosphorus eater links between carbon number 3 and 5 of the pentose sugar to form long polynucleotide chains that constitute the nucleic acid.

6) **Describe double helix of DNA**

Watson and Crick (1953) proposed a model for the structure of DNA molecule. The DNA molecule is a double helix structure consisting of two long polynucleotide chains coiled round each other around an imaginary axis and running opposite to each other. Each polynucleotide chain consists of thousands of nucleotide units. The back bone of the two helices of polynucleotide chains consists of Deoxy ribose phosphates while the bases are present on the inner side. The bases of the one polynucleotide chain are complementary to the bases of other polynucleotide chain and are joined together by hydrogen bonds. The base pairing is very specific. The complementary bases are 1) Adenine and Thymine 2) Guanine and Cytosine. The distance between two subsequent bases in the polynucleotide chain is 3.4 A^o .Each turn of the two polynucleotide chain is completed after 10 bases. The distance between the axis and the sugar phosphate region is about $10A^o$.

7. **Classify the proteins giving suitable examples for each group**

A) **Classification based on the shape of protein molecules**:

1) **Globular or Corpuscular proteins**- They have an axial ratio of less than 10. Hence possess a relatively spherical or ovoid shape. Usually soluble in water, acid, bases, salts or alcohol. Eg. Amylase, urease

2) **Fibrous proteins**- These have axial ratio greater than 10. Hence resemble long fibers or ribbons in shape, These are mainly of animal origin and insoluble in all common solvents eg., Collagen

B) **Based on composition and solubility**

Based on composition, proteins are classified into simple, conjugate and derived proteins

1) **Simple proteins**- This group includes proteins containing amino acid as structural components. On decomposition with acids, these liberate constituent amino acids.

Based on solubility, simple proteins are classified into

a) Albumins- It is soluble in water and dilute solutions of acids, bases and salts. It is precipitated with neutral salts, coagulated by heat. It is widely distributed in nature and more abundant in seeds

b) Globulins- It is widely distributed in nature, insoluble in water, soluble in salt solution

c) Glutelins- Isolated from plant seeds, insoluble in water, salt solution but soluble in acid and alkali, coagulated by heat eg., glutenin(Wheat)

d) Prolamins- It is insoluble in water and dilute salt solutions but soluble in dilute acids and alkalies and also in 60-80% alcohol solution, not coagulated by heat eg., Hordein of oats

e) Protamines- These are basic proteins and occur almost entirely in animals. It is soluble in water, not coagulated by heat, form salt with acid and alkali

f) **Scleroproteins**- it occur almost entirely in animals. It is insoluble in water; dilute solutions of acid, bases and salts, 60-80 % alcohol

2) **Conjugate Proteins**:

These proteins on hydrolysis yield amino acids plus some non- amino acid part called as the prosthetic group. Depending upon the nature of the prosthetic group associated with them, conjugate proteins are grouped into

1. Nucleoproteins (histones)- These are associated with nucleic acids
2. Glycoproteins- These are associated with some carbohydrates
3. Chromoprotein – These proteins are associated with some coloring matter eg.

Chlorophyll, carotenoids

4. Lipoproteins – These are associated with lipids or fatty substances eg., cephalin, lecithin
5. Iron-prophyrin proteins – These are associated with iron-prophyrin compound eg., cytochromes
6. Simple metal containing proteins- These are associated with some metals directly eg., ferredoxin
7. Flavoproteins – These are associated with some flavin compounds eg., FAD

3) **Derived proteins**

These are derived proteins resulting from the action of heat, enzymes or chemical reagents

a) **Primary derived- The size of the proteins are not altered materially**

1) Proteans- Insoluble in water eg., Myosan
2) Metaproteins- Insoluble in water, soluble in acid and alkali
3) Coagulated proteins- insoluble in water, produced by action of heat or alcohol eg, coagulated egg white

b) **Secondary derived-** These are derivatives of proteins in which the hydrolysis has certainly occurred

1) Proteoses- soluble in water, coaguable in heat eg, albumose
2) Peptones- Soluble in water, non- coaguable by heat produced by action of dilute acids
3) Polypeptides- These are combinations of two or more amino acid units

8) **Enumerate the physical and chemical properties of proteins**

A) **Physical properties**

1) Taste and color- Proteins are colorless, homogenous and crystalline
2) Shape and size- Simple crystalloid, spherical to long fibrillar structures
3) Molecular weight- It has large molecular weight ranging from 5×10^3 to 1×10^6
4) Colloidal nature- Because of their giant size, the proteins exhibit many colloidal properties
5) Denaturation- It refers to the changes in the properties of a protein
6) Amphoteric nature- They act as acid and alkali. Thus the proteins are dipolar or zwitter ions
7) Ion binding capacity- Being Amphoteric in nature, the proteins can form salts with both cations and anions based on their net charge.
8) Solubility - The solubility is markedly influenced by pH. It is lowest at isoelectric point and increases with acidity or alkalinity.
9) Optical activity- All the protein solution rotate the plane of polarized light to the left (levorotatory)

B) **Chemical properties**

1) Hydrolysis:

a) By acidic agents- Proteins up on hydrolysis with concentrated HCl at 100- 110^{o}C for 6 to 20 hours yield amino acid in the form of their hydrochlorides

b) By alkaline agents – Proteins may also be hydrolyzed by 2N NaOH

c) By proteolytic enzymes- Certain proteolytic enzymes like pepsin and trypsin hydrolyze the proteins under certain conditions of temperature and acidity

2) Reaction involving COOH group

a) Reaction with alkalies (salt formation)- The carboxylic group of amino acids can release H^+ ion with the formation of carboxylate(COO^-) ions. These may be neutralized by Na, Ca to form salts

$R\text{-}CHNH_2\text{-}COOH + NaOH \longrightarrow R\text{-}CHNH_2\text{-}COONa + H_2O$

b) Reaction with alcohol (Esterification) - Proteins reacts with alcohol to form Corresponding esters. These esters are volatile than amino acid

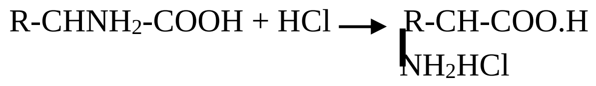
$R\text{-}CHNH_2\text{-}COOH + C_2H_5OH \longrightarrow R\text{-}CHNH_2\text{-}COO.\ C_2H_5 + H_2O$

c) Reaction with amines – Amino acid reacts with amines to form amides

$R\text{-}CHNH_2\text{-}COOH + NH_2\text{-}R^1 \longrightarrow R\text{-}CHNH_2\text{-}CO\text{-}NH\text{-}R^1 + H_2O$

3) Reaction involving amino group:

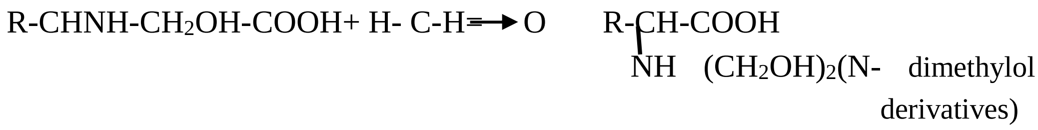
a) Reaction with mineral acids(Salt formation)- When free amino acids or proteins are treated with mineral acids like HCl, the acid salts are formed



$$R\text{-}CHNH_2\text{-}COOH + HCl \longrightarrow \underset{\displaystyle NH_2HCl}{\underset{|}{R\text{-}CH}}\text{-}COO.H$$

b) Reaction with formaldehyde- With formaldehyde, the hydroxy-methyl derivatives are formed. These derivatives are insoluble in water and resistant to microbial attack

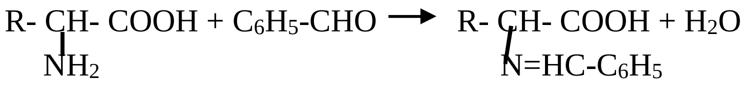
$R\text{-}CHNH_2\text{-}COOH + H\text{-}C\text{-}H{=}O \longrightarrow R\text{-}CHNH\text{-}CH_2OH\text{-}COOH$ (N monomethylol Derivatives)



$$R\text{-}CHNH\text{-}CH_2OH\text{-}COOH + H\text{-}C\text{-}H{=}O \longrightarrow R\text{-}\underset{\displaystyle NH\ (CH_2OH)_2}{\underset{|}{CH}}\text{-}COOH$$

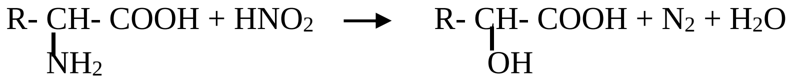
(N- dimethylol derivatives)

c) Reaction with benzaldehyde (Schiff's base are formed)



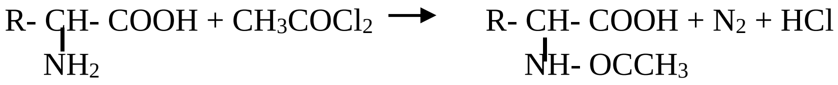
$$R\text{-}\underset{\displaystyle NH_2}{\underset{|}{CH}}\text{-}COOH + C_6H_5\text{-}CHO \longrightarrow R\text{-}\underset{\displaystyle N{=}HC\text{-}C_6H_5}{\underset{|}{CH}}\text{-}COOH + H_2O$$

d) Reaction with nitrous acid – The amino acids react with HNO_2 to liberate N_2 and to produce the corresponding α- hydroxy alcohol



$$R\text{-}\underset{\displaystyle NH_2}{\underset{|}{CH}}\text{-}COOH + HNO_2 \longrightarrow R\text{-}\underset{\displaystyle OH}{\underset{|}{CH}}\text{-}COOH + N_2 + H_2O$$

f) Reaction with acylating agents (acylation)– Acylation is brought about by many acid chlorides



$$R\text{-}\underset{\displaystyle NH_2}{\underset{|}{CH}}\text{-}COOH + CH_3COCl_2 \longrightarrow R\text{-}\underset{\displaystyle NH\text{-}OCCH_3}{\underset{|}{CH}}\text{-}COOH + N_2 + HCl$$

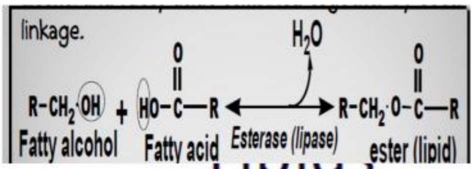
4) Reaction involving both COOH and NH_2 groups

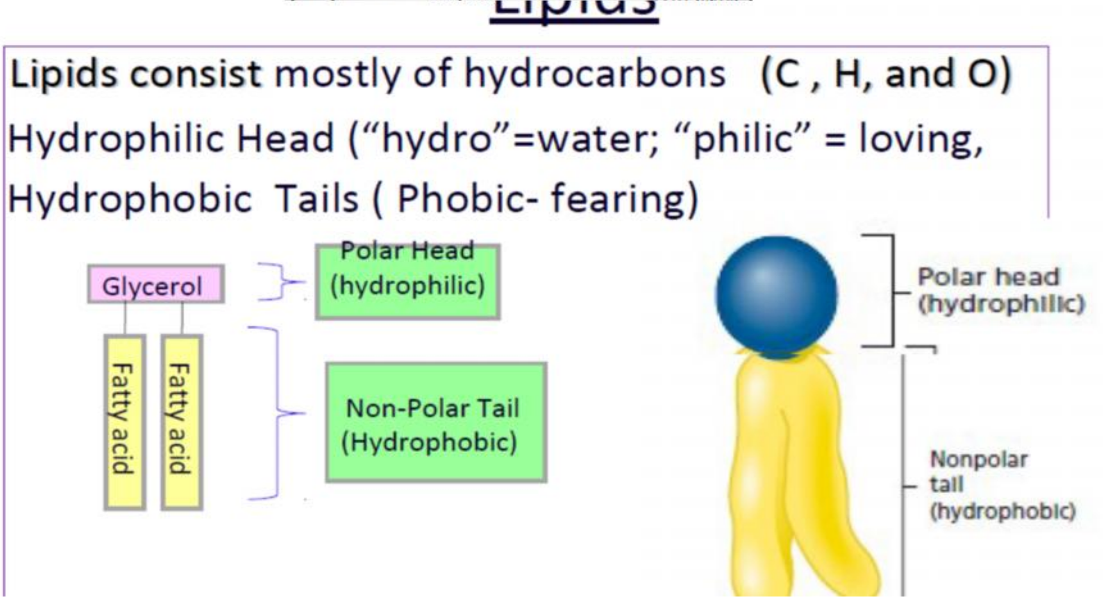
a) Ninhydrin test- It is powerful oxidizing agent and causes oxidative decarboxylation of α- amino acids producing CO_2, NH_3 and aldehyde with one less carbon atom than the parent amino acids. The reduced Ninhydrin then react with the liberated NH_3 forming blue colored complex.

LIPIDS

Definition

Lipids are organic compounds formed mainly from alcohol and fatty acids combined together by ester linkage. Lipids are naturally occurring molecules from plants or animals that are soluble in nonpolar organic solvents





Lipids consist mostly of hydrocarbons (C , H, and O)

Hydrophilic Head ("hydro"=water; "philic" = loving,

Hydrophobic Tails (Phobic- fearing)

Lipids are insoluble in water, but soluble in fat or organic solvents (ether, chloroform, acetone, benzene). Lipids include oils, waxes, fats and related compounds. They are widely distributed in nature both in plants and in animals

Biological Importance of Lipids

They are more palatable and storable to unlimited amount compared to carbohydrates. They have a high-energy value (25% of body needs) and they provide more energy per gram than carbohydrates and proteins but carbohydrates are the preferable source of energy. (9, 4.4 Kcal/g). Supply the essential fatty acids that cannot be synthesized by the body. Supply the body with fat-soluble vitamins (A, D, E and K). They are important constituents of the nervous system. Tissue fat is an essential constituent of cell membrane and nervous system. It is mainly phospholipids in nature that are not affected by starvation.

Stored lipids "depot fat" is stored in all human cells acts as:

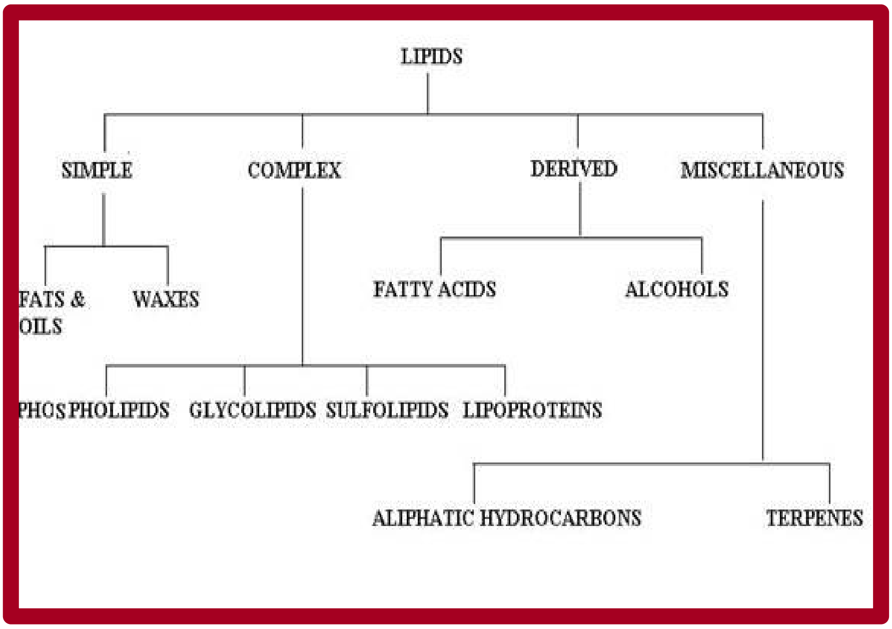
- A store of energy.
- A pad for the internal organs to protect them from outside shocks.
- A subcutaneous thermal insulator against loss of body heat.

Lipoproteins: which are complex of lipids and proteins, are important cellular constituents that present both in the cellular and subcellular membranes.

Cholesterol: enters in membrane structure and is used for synthesis of adrenal cortical hormones, vitamin D3 and bile acids.

Lipids: provide bases for dealing with diseases such as obesity, atherosclerosis, lipid-storage diseases, essential fatty acid deficiency, respiratory distress syndrome

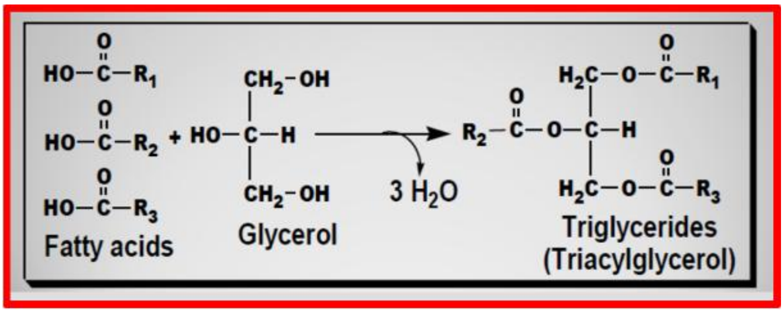
CLASSIFICATION OF LIPIDS



Simple Lipids

A) Neutral fats or oils (Triglycerides)

Esters of fatty acids with glycerol and monohydric alcohols. Depending upon the constituent alcohols they are further subdivided into fats or oils and waxes. Fats, also termed as triacylglycerol are esters of fatty acids with glycerol e.g. Plants- vegetable oils; Animals-ghee and butter. Waxes are esters of fatty acids and alcohols other than glycerol e.g., Plant wax-carnauba wax;



Types of triglycerides

a) Simple triglycerides:

If the three fatty acids connected to glycerol are of the same type, the triglyceride is called simple triglyceride

b) Mixed Triglycerides

If the three fatty acids connected to glycerol are of different type, the triglyceride is called mixed triglyceride

Natural fats are mixture of mixed triglycerides with a small amount of simple triglycerides. The commonest fatty acids in animal fats are palmitic, stearic and oleic acids. The main difference between fats and oils is for oils being liquid at room temperature, whereas, fats are solids. This is mainly due to presence of larger percentage of unsaturated fatty acids in oils than fats that has mostly saturated fatty acids

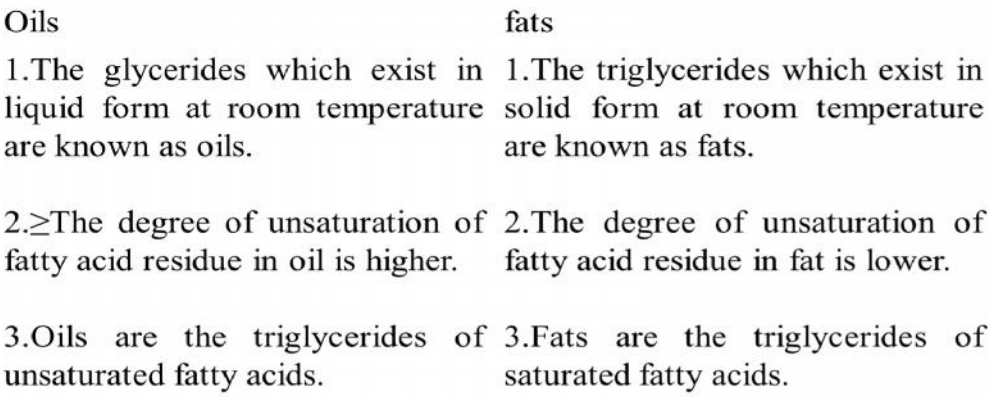
B-Waxes

Waxes are solid simple lipids containing a monohydric alcohol (with a higher molecular weight than glycerol) esterified to long-chain fatty acids. Examples of these alcohols are palmitoyl alcohol, cholesterol, vitamin A or D. Waxes are insoluble in water, but soluble in fat solvents and are negative for Acrolein test. Waxes are not easily hydrolyzed as the fats and are indigestible by lipases and are very resistant to rancidity. Thus they are of no nutritional value

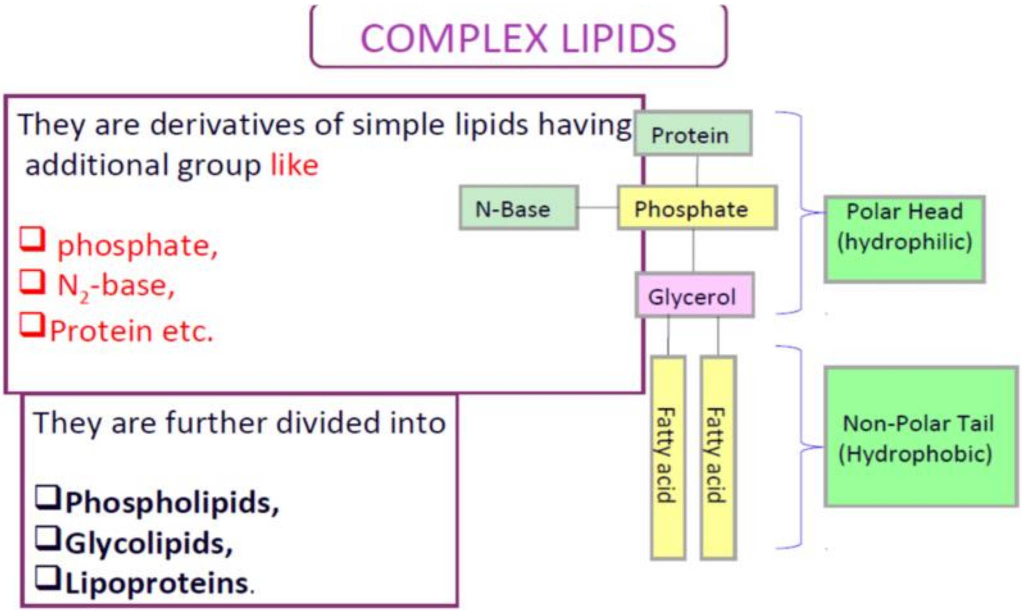
Physical properties of fat and oils

Freshly prepared fats and oils are colourless, odourless and tasteless. Any color, or taste is due to association with other foreign substances, e.g., The yellow color of body fat or milk fat is due to carotene pigments (cow milk). Fats have specific gravity less than 1(one) and, therefore, they float on water. Fats are insoluble in water, but soluble in organic solvents as ether and benzene. Melting points of fats are usually low, but higher than the solidification point,

Difference between fat and oil



Oils	fats
1.The glycerides which exist in liquid form at room temperature are known as oils.	1.The triglycerides which exist in solid form at room temperature are known as fats.
2.≥The degree of unsaturation of fatty acid residue in oil is higher.	2.The degree of unsaturation of fatty acid residue in fat is lower.
3.Oils are the triglycerides of unsaturated fatty acids.	3.Fats are the triglycerides of saturated fatty acids.



COMPLEX LIPIDS

They are derivatives of simple lipids having additional group like

❑ phosphate,
❑ N_2-base,
❑ Protein etc.

They are further divided into

❑ **Phospholipids,**
❑ **Glycolipids,**
❑ **Lipoproteins.**

They are lipids that contain additional substances, e.g., sulfur, phosphorus, amino group, carbohydrate, or proteins beside fatty acid and alcohol.

Compound or conjugated lipids are classified into the following types according to the nature of the additional group

a) Phospholipids
b) Glycolipids.
c) Lipoproteins
d) Sulfolipids and amino lipids

A-Phospholipids

Phospholipids or phosphatides are compound lipids, which contain phosphoric acid group in their structure

Every animal and plant cell contains phospholipids. The membranes bounding cells and subcellular organelles are composed mainly of phospholipids. Important role in signal transduction across the cell membrane. They are source of polyunsaturated fatty acids for synthesis of eicosanoids.

Sources: They are found in all cells (plant and animal), milk and egg yolk in the form of lecithins.

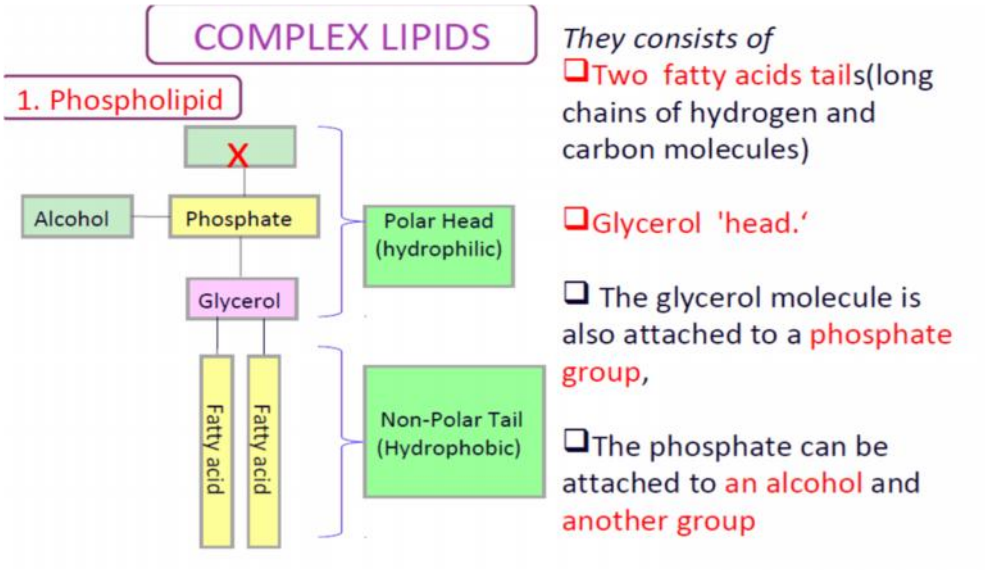
Structure: phospholipids are composed of:

Fatty acids (a saturated and an unsaturated fatty acid).

Nitrogenous base (choline, serine, threonine, or ethanolamine).

Phosphoric acid.

Fatty alcohols (glycerol, inositol or sphingosine



They consists of

❑Two fatty acids tails(long chains of hydrogen and carbon molecules)

❑Glycerol 'head.'

❑ The glycerol molecule is also attached to a phosphate group,

❑The phosphate can be attached to an alcohol and another group

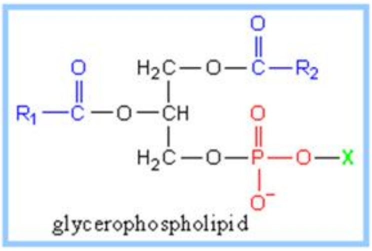
Classification of Phospholipids

Are classified into 2 groups according to the type of the alcohol present into two types

A-Glycerophospholipids: They are regarded as derivatives of phosphatidic acids that are the simplest type of phospholipids

Phosphatidic acids: those lipids which on hydrolysis give rise to one molecule of glycerol and phosphoric acid and two molecules of fatty acids

1. Lecithins
2. Cephalins.
3. Plasmalogens.
4. Inositides.
5. Cardiolipin



Lecithins

Lecithins are glycerophospholipids that contain choline as a base beside phosphatidic acid. They exist in 2 forms a- and b- lecithins. The common fatty acids in lecithins are stearic, palmitic, oleic, linoleic, linolenic, or arachidonic acids.

Cephalins

Cephalins resemble lecithins in structure except that choline is replaced by ethanolamine, serine or threonine amino acids. Certain cephalins are constituents of the complex mixture of phospholipids, cholesterol and fat that constitute the lipid component of the lipoprotein.

Inositides

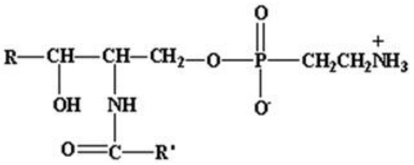
They are similar to lecithins or cephalins but they have the cyclic sugar alcohol, inositol as the base. They are formed of glycerol, one saturated fatty acid, one unsaturated fatty acid, phosphoric acid and inositol

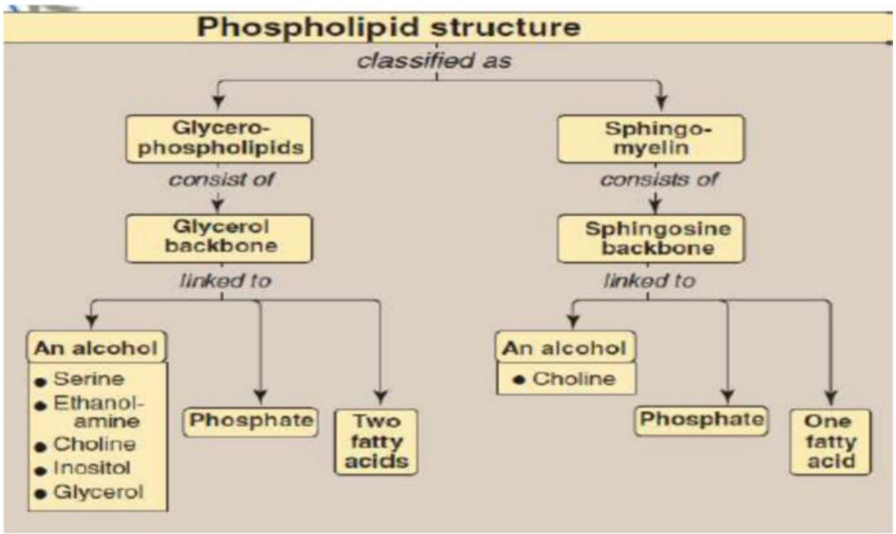
B-Sphingo phospholipids

Sphingophospholipids are found in the seeds of several plant species.

Sphingomyelins

Sphingomyelins are found in large amounts in brain and nerves and in smaller amounts in lung, spleen, kidney, liver and blood. Sphingomyelins differ from lecithins and cephalins in that they contain sphingosine as the alcohol instead of glycerol, they contain two nitrogenous bases: sphingosine itself and choline





B-Glycolipids

Glycolipids are structurally characterised by the presence of one or more monosaccharide residues and the absence of a phosphate. They are lipids that contain carbohydrate residues with sphingosine as the alcohol and a very long-chain fatty acid (24 carbon series). The monosaccharides commonly attached are D-glucose, D-galactose or N-acetyl D-galactosamine

C-Lipoproteins

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Protein molecules associated with triacylglycerol, cholesterol or phospholipids are called lipoproteins The protein part of lipoprotein is known as apoprotein. Lipoproteins occur in milk, egg-yolk and also as components of cell membranes

Structural lipoproteins

These are widely distributed in tissues being present in cellular and subcellular membranes

Transport lipoproteins:

These are the forms present in blood plasma. They are composed of a protein called apolipoprotein and different types of lipids. (Cholesterol, cholesterol esters, phospholipids and triglycerides.

sulpholipids

Sulfolipids are a class of lipids which possess a sulfur-containing functional group. The predominant fatty acid present in sulpholipid is linolenic acid. The sulpholipid is mostly present in chloroplasts, predominantly in the membranes of thylakoid. Plant sulfolipid is found in the photosynthetic membranes of plastids and provides negative charge in the thylakoid membrane where it is thought to stabilize photosynthetic complexes

Derived lipids

They occur as such or are released from the other two major groups because of hydrolysis that is are the building blocks for simple and complex lipids

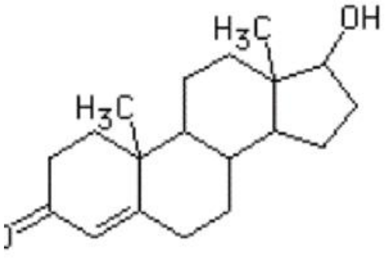
They include fatty acid and alcohol , Fatty soluble vitamin A,D, E and K, hydrocarbon and sterols.

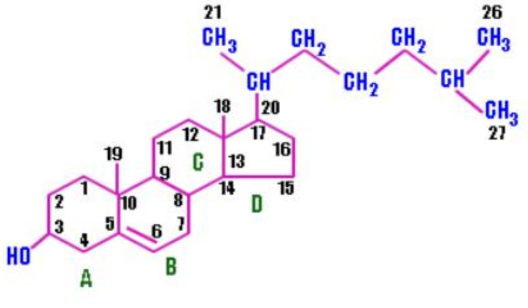
Steroids

Steroids constitute an important class of biological compounds

The steroids do not contain fatty acids but are included in lipids as they have fat like properties

Composition: Contain a characteristic arrangement of 3 Cyclohexane ring, 1 Cyclopentane ring, a total of 17-carbon atoms in four fused Carbon ring.





Classification

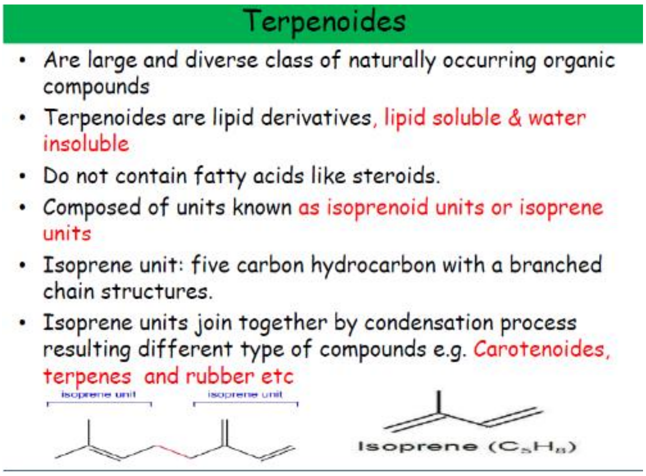
Sterol

- Also known as steroid alcohols
- occur naturally in plants, animals, and fungi, with the most familiar type of animal sterol being cholesterol
- TYPES:
- Phytosterols – plant sterol (campesterol, sitosterol, and stigmasterol)
- blocks cholesterol absorption sites in the human intestine, thus helping to reduce cholesterol in humans
- Zoosterol – animal sterol (cholesterol)
- Ergosterol – sterol present in the cell membrane of fungi

Examples of steroids:

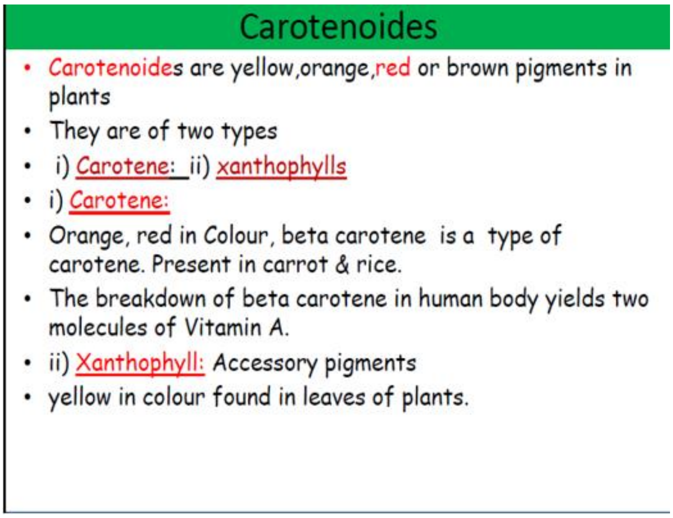
I) Cholesterol:

- An important component of animal cell membrane.
- Precursor molecule of all hormones such as aldosterone, sex hormone and Vitamin D
- Aldosterone helps to regulate Na+ ions in the blood
- Sex hormones e.g. testosterones, progesterone, oestrogens help to maintain male and female characteristics



Terpenoides

- Are large and diverse class of naturally occurring organic compounds
- Terpenoides are lipid derivatives, lipid soluble & water insoluble
- Do not contain fatty acids like steroids.
- Composed of units known as isoprenoid units or isoprene units
- Isoprene unit: five carbon hydrocarbon with a branched chain structures.
- Isoprene units join together by condensation process resulting different type of compounds e.g. Carotenoides, terpenes and rubber etc



Carotenoides

- Carotenoides are yellow,orange,red or brown pigments in plants
- They are of two types
- i) Carotene: ii) xanthophylls
- i) Carotene:
- Orange, red in Colour, beta carotene is a type of carotene. Present in carrot & rice.
- The breakdown of beta carotene in human body yields two molecules of Vitamin A.
- ii) Xanthophyll: Accessory pigments
- yellow in colour found in leaves of plants.

Plant Fatty Acids

They are basic building blocks of lipids (simplest lipids). Fatty acids are carboxylic acids with a long hydrocarbon chain attached

Definition:

Aliphatic mono-carboxylic acids that are mostly obtained from the hydrolysis of natural fats and oils.

The general formula R-(CH2)n-COOH and mostly have straight chain (a few exceptions have branched and heterocyclic chains). "n" is mostly an even number of carbon atoms (2-34)

Structure of fatty acids

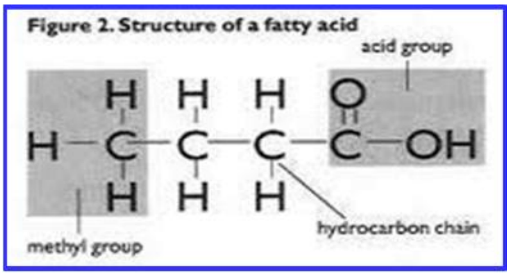
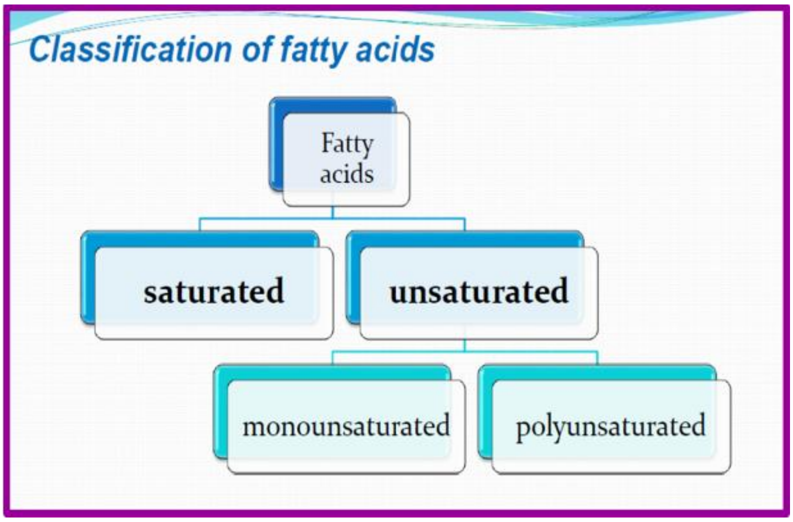


Figure 2. Structure of a fatty acid

A fatty acid consists of a hydrophobic hydrocarbon chain with a terminal carboxyl group

All of the lipid molecules in cell membranes are amphipathic (or amphiphilic) because they have both polar (Hydrophilic) and non –polar (hydrophobic) portions in their structure



1-Saturated Fatty Acids

a. No double bonds with 2-24 or more carbons.
b. Solid at RT except if they are short chained.
c. May be even or odd numbered.
d. Molecular formula, CnH2n+1COOH

Saturated Fatty acids could be:

A-Short chain Saturated F.A. (2-10 carbon).

a - Short chain Saturated volatile F.A.(2-6 C).
b - Short chain Saturated non-volatile F.A.(7-10 C).

A- Saturated- short chain Volatile short -chain Fatty acids:

Liquid in nature and contain 1-6 C water-soluble volatile at room temperature
e.g., acetic, butyric, & caproic acids.

Acetic F.A. (2 C) CH_3-COOH.

Butyric F.A. (4 C) CH_3-$(CH_2)_2$-COOH.

Caproic F.A. (6C) CH_3-(CH_2)4-COOH

Non -volatile short - chain fatty acids:

Solids at room temperature, contain 7-10 carbons. Water-soluble, non-volatile at RT include caprylic and capric F.A.

Caprylic (8 C) CH_3-$(CH_2)_6$-COOH.

Capric (10 C) CH_3-$(CH_{2)8}$-COOH

B - Saturated - Long -chain fatty acids:

<10 carbon atoms.

In hydrogenated oils, animal fats, butter and coconut and palm oils.

Non-volatile, Water-insoluble

E.g. palmitic, stearic, & lignoceric F.A.

- Palmitic (16 C) CH_3-$(CH_2)_{14}$-COOH
- Stearic (18 C) CH_3-$(CH_2)_{16}$-COOH
- Lignoceric (24C) CH_3-$(CH_2)_{22}$-COOH

2 **–Unsaturated Fatty Acids**

- Contain double bond

A. monounsaturated: they contain one double bond.
(CnH2n-1 COOH)

B. polyunsaturated; they contain more the one double bond
(CnH2n-# COOH).

A-Mono unsaturated fatty acids:

1-Palmitoleic acid:

It is found in all fats. It is C16:1Δ9, i.e., has 16 carbons and one double bond located at carbon number 9 and involving carbon 10.

CH_3-(CH_2)5CH = CH-(CH_2)7 –COOH

2-Oleic acid

Is the most common fatty acid in natural fats?

It is C18:1Δ9, i.e., has 18 carbons and one double bond located at carbon number 9 and involving carbon 10.

$CH_{3-(}CH_2)_7$- CH=CH – $(CH_2)_7$-COOH

B **-Poly unsaturated fatty acids**

Definition:

They are essential fatty acids that cannot be synthesized in the human body and must be taken in adequate amounts in the diet. They are required for normal growth and metabolism

Source: vegetable oils such as corn oil, linseed oil, peanut oil, olive oil, cottonseed oil, soybean oil and many other plant oils, cod liver oil and animal fats.

Deficiency: Their deficiency in the diet leads to nutrition deficiency disease.

Symptoms: poor growth and health with susceptibility to infections, dermatitis, decreased capacity to reproduce, impaired transport of lipids, fatty liver, and lowered resistance to stress.

1-Linoleic:

C18: 2D 9, 12.

It is the most important since other essential fatty acids can be synthesized from it in the body.

$CH_3-(CH_2)_4-CH = CH-CH_2-CH= CH- (CH_2)_7-COOH$

2-Linolenic acid:

C 18:3D 9, 12, 15,

In corn, linseed, peanut, olive, cottonseed and soybean oils.

$CH_3-CH_2-CH=CH-CH_2-CH=CH-CH_2- CH=CH-(CH_2)_7- COOH$

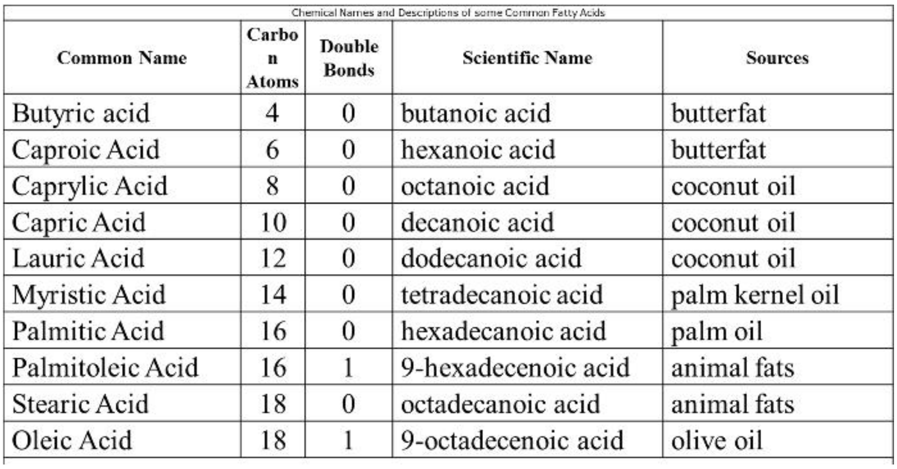
3-Arachidonic acid:

C20: 4D 5, 8, 11, 14.

It is an important component of phospholipids in animal and in peanut oil from which prostaglandins are synthesized.

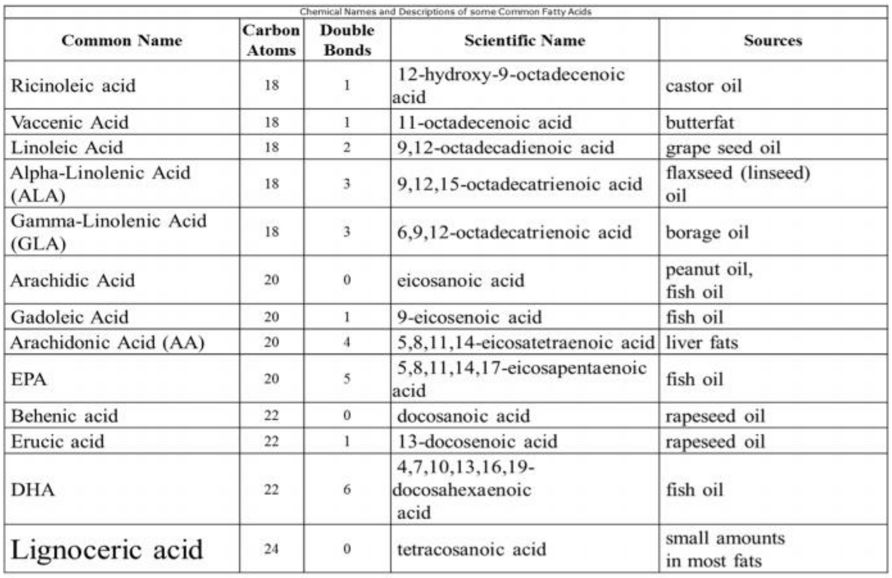
$CH_3-(CH_2)_4-CH=CH-CH_2-CH=CH-CH_2- CH=CH-CH_2-CH=CH-(CH_2)_3-COOH$

Common Fatty Acids



Chemical Names and Descriptions of some Common Fatty Acids

Common Name	Carbon Atoms	Double Bonds	Scientific Name	Sources
Butyric acid	4	0	butanoic acid	butterfat
Caproic Acid	6	0	hexanoic acid	butterfat
Caprylic Acid	8	0	octanoic acid	coconut oil
Capric Acid	10	0	decanoic acid	coconut oil
Lauric Acid	12	0	dodecanoic acid	coconut oil
Myristic Acid	14	0	tetradecanoic acid	palm kernel oil
Palmitic Acid	16	0	hexadecanoic acid	palm oil
Palmitoleic Acid	16	1	9-hexadecenoic acid	animal fats
Stearic Acid	18	0	octadecanoic acid	animal fats
Oleic Acid	18	1	9-octadecenoic acid	olive oil



Chemical Names and Descriptions of some Common Fatty Acids

Common Name	Carbon Atoms	Double Bonds	Scientific Name	Sources
Ricinoleic acid	18	1	12-hydroxy-9-octadecenoic acid	castor oil
Vaccenic Acid	18	1	11-octadecenoic acid	butterfat
Linoleic Acid	18	2	9,12-octadecadienoic acid	grape seed oil
Alpha-Linolenic Acid (ALA)	18	3	9,12,15-octadecatrienoic acid	flaxseed (linseed) oil
Gamma-Linolenic Acid (GLA)	18	3	6,9,12-octadecatrienoic acid	borage oil
Arachidic Acid	20	0	eicosanoic acid	peanut oil, fish oil
Gadoleic Acid	20	1	9-eicosenoic acid	fish oil
Arachidonic Acid (AA)	20	4	5,8,11,14-eicosatetraenoic acid	liver fats
EPA	20	5	5,8,11,14,17-eicosapentaenoic acid	fish oil
Behenic acid	22	0	docosanoic acid	rapeseed oil
Erucic acid	22	1	13-docosenoic acid	rapeseed oil
DHA	22	6	4,7,10,13,16,19-docosahexaenoic acid	fish oil
Lignoceric acid	24	0	tetracosanoic acid	small amounts in most fats

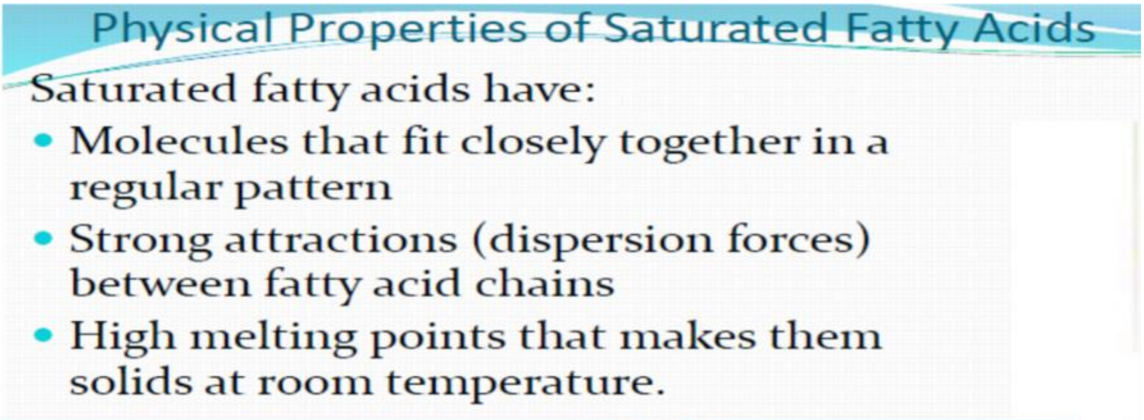
Unusual fatty acids

The unusual fatty acids are found only in few individual species or genus or a whole family.

Castor bean (Ricinus communis) seed oil isrich in ricinoleic acid (90%) which is 12-hydroxy oleic acid $CH_3(CH_2)_5\text{-}CH(OH)\text{-}CH_2\text{-}CH{=}CH\text{-}(CH_2)_7\text{-}COOH$.

Rape seed (Brassica napus) is rich in Erucic acid
(cis-13- docosenoic acid)
$CH_3(CH_2)_7\text{-}CH{=}CH\text{-}(CH_2)_{11}\text{-}COOH$).

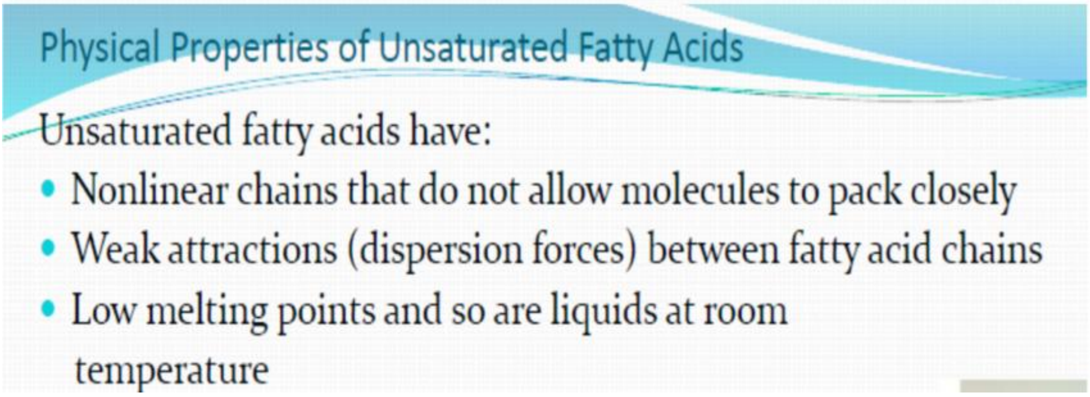
Hydnocarpic and chaulmoogricacids are found in chaulmoogra oil which is used in the treatment of leprosy



Physical Properties of Saturated Fatty Acids

Saturated fatty acids have:

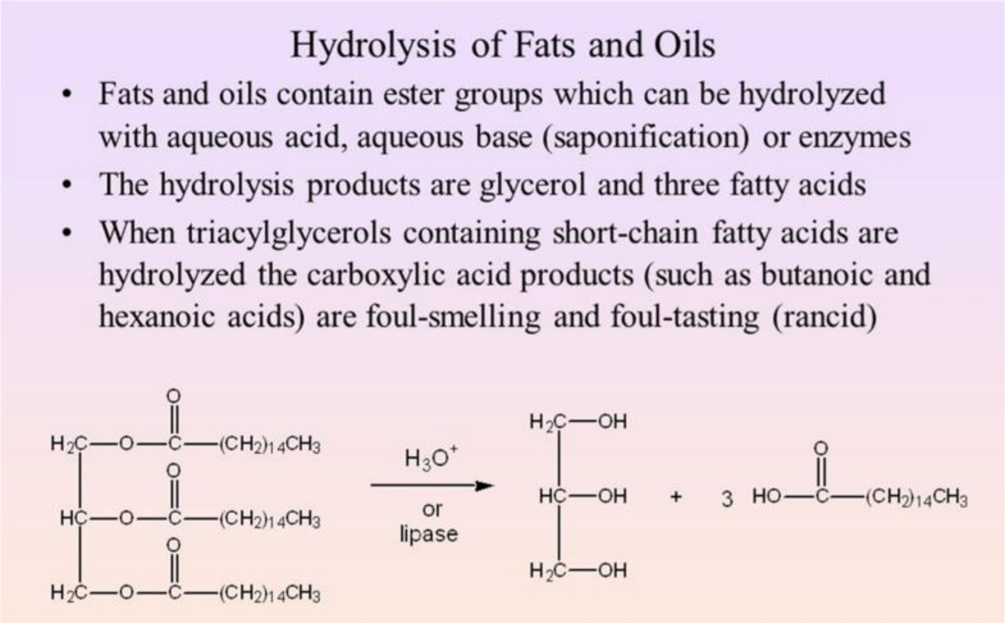
- Molecules that fit closely together in a regular pattern
- Strong attractions (dispersion forces) between fatty acid chains
- High melting points that makes them solids at room temperature.



Physical Properties of Unsaturated Fatty Acids

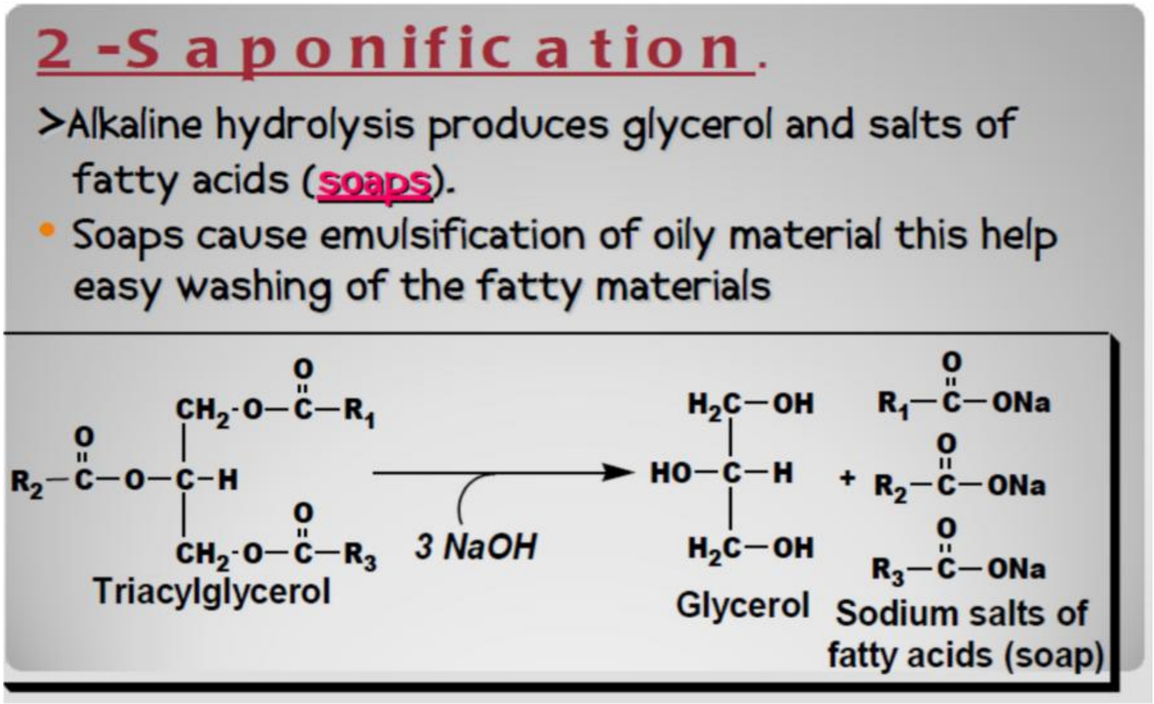
Unsaturated fatty acids have:

- Nonlinear chains that do not allow molecules to pack closely
- Weak attractions (dispersion forces) between fatty acid chains
- Low melting points and so are liquids at room temperature



Hydrolysis of Fats and Oils

- Fats and oils contain ester groups which can be hydrolyzed with aqueous acid, aqueous base (saponification) or enzymes
- The hydrolysis products are glycerol and three fatty acids
- When triacylglycerols containing short-chain fatty acids are hydrolyzed the carboxylic acid products (such as butanoic and hexanoic acids) are foul-smelling and foul-tasting (rancid)



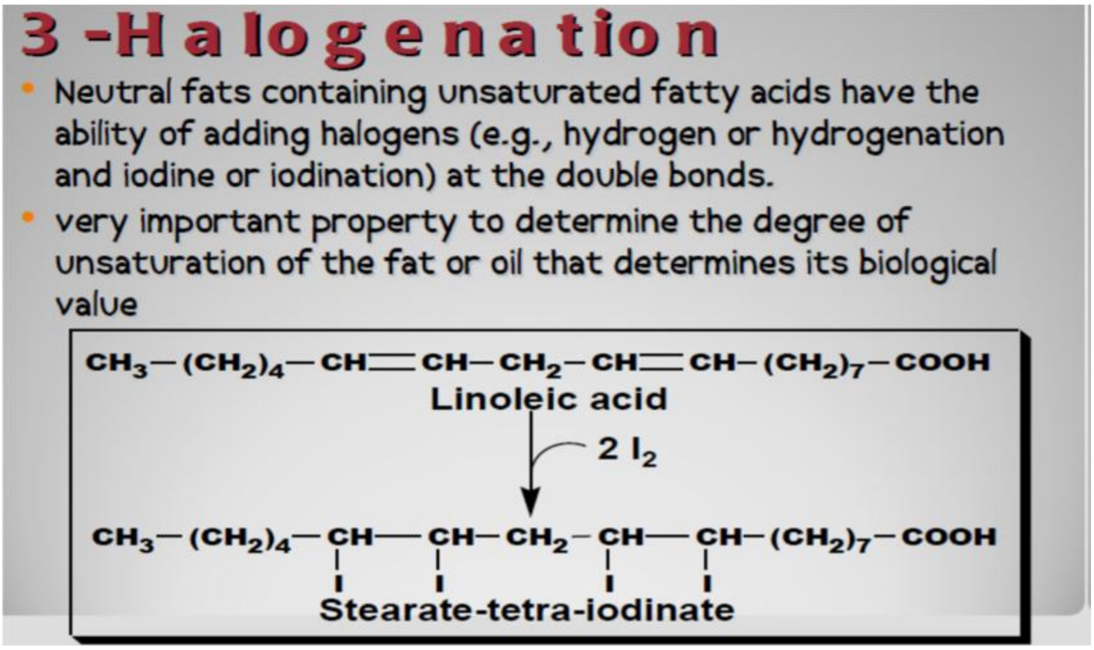
2 -Saponification.

>Alkaline hydrolysis produces glycerol and salts of fatty acids (soaps).

- Soaps cause emulsification of oily material this help easy washing of the fatty materials

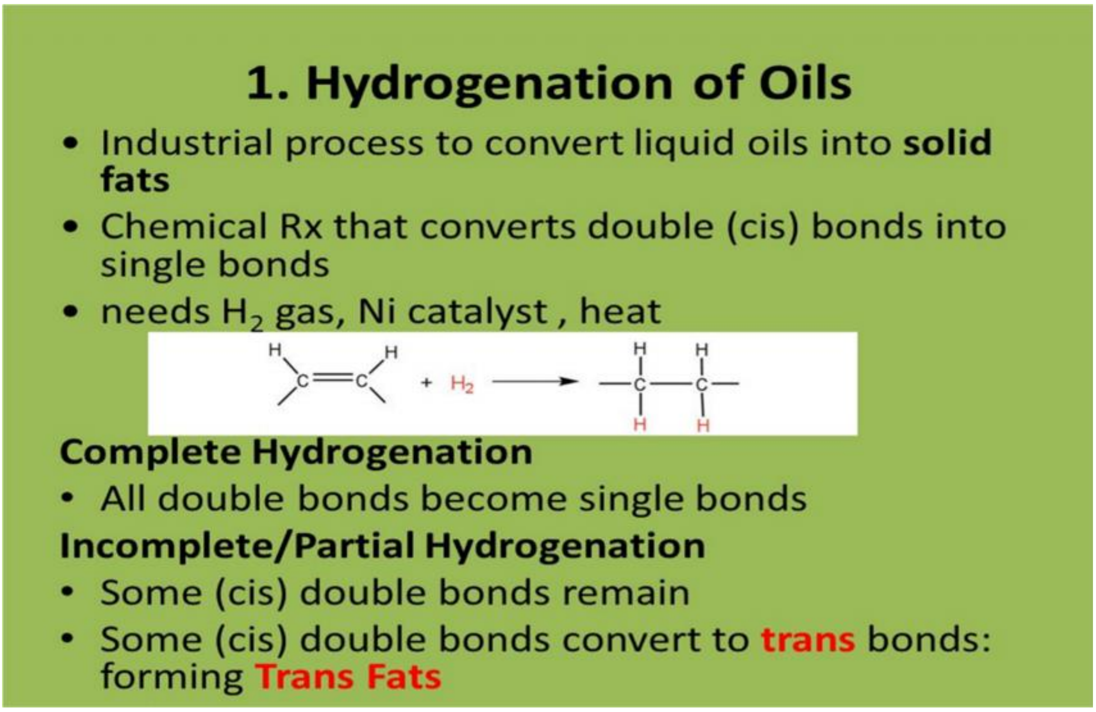
The alkali salt of fatty acid resulting from saponification is soap.

- The soaps we use for washing consists of Na or K salts of fatty acids like palmitic, stearic and oleic acid.
- The potassium soaps are soft and soluble whereas the sodium soaps are hard and less soluble in water



3 -Halogenation

- Neutral fats containing unsaturated fatty acids have the ability of adding halogens (e.g., hydrogen or hydrogenation and iodine or iodination) at the double bonds.
- very important property to determine the degree of unsaturation of the fat or oil that determines its biological value



1. Hydrogenation of Oils

- Industrial process to convert liquid oils into **solid fats**
- Chemical Rx that converts double (cis) bonds into single bonds
- needs H_2 gas, Ni catalyst , heat

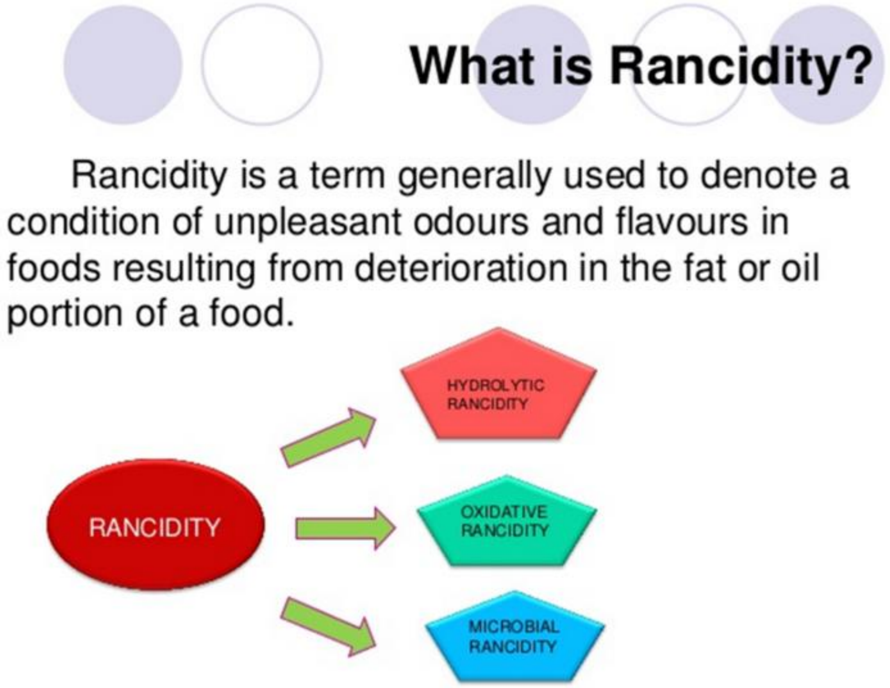
Complete Hydrogenation

- All double bonds become single bonds

Incomplete/Partial Hydrogenation

- Some (cis) double bonds remain
- Some (cis) double bonds convert to **trans** bonds: forming **Trans Fats**

The degree of unsaturation of the fatty acids present in triacylglycerol determines whether a fat is liquid or solid at room temperature. The presence of more unsaturated fatty acids lower the melting point. The presence of highly unsaturated fatty acids makes the oil more susceptible to oxidative deterioration. The objective of hydrogenation is to reduce the degree of unsaturation and to increase the melting point of the oil. The oil can be selectively hydrogenated by careful choice of catalyst and temperature. Hydrogenation of unsaturated fats in the presence of a catalyst is known as hardening.



What is Rancidity?

Rancidity is a term generally used to denote a condition of unpleasant odours and flavours in foods resulting from deterioration in the fat or oil portion of a food.

Saturated fats resist rancidity more than unsaturated fats that have unsaturated double bonds.

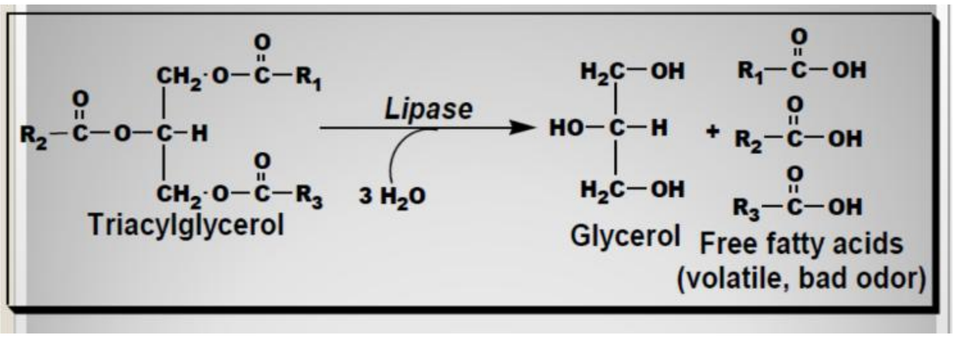
Definition

Physico-chemical change development of unpleasant odor or taste or abnormal color particularly on aging exposure to atmospheric oxygen, light, moisture, bacterial or fungal contamination and/or heat.

1-Hydrolytic rancidity:

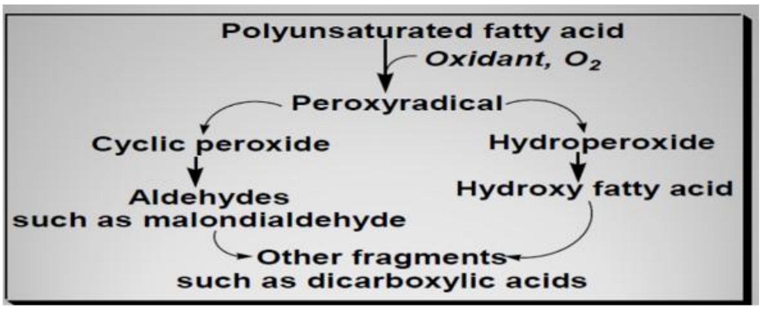
From slight hydrolysis of the fat by lipase. Bacterial contamination leading to the liberation of free fatty acids and glycerol at high temp and moisture .Volatile short-chain fatty acids have unpleasant odor. Caused by the breaking down of a lipid into its component fatty acids and glycerol. C-O-CO-R + H_2O → C-O-H + HO-CO-R

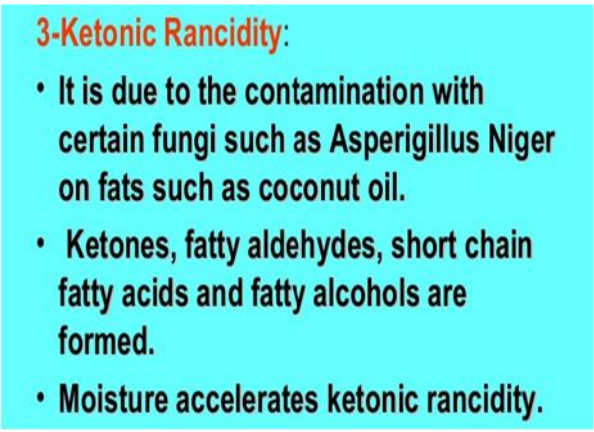
The water present in the food and the high temperature will increase the rate of hydrolysis to fatty acids



2-Oxidative Rancidity

Oxidation of fat or oil by exposure to oxygen, light and/or heat producing peroxide derivatives e.g., peroxides, aldehydes, ketones and dicarboxylic acids that are toxic and have bad odor. due to oxidative addition of oxygen at the unsaturated double bond of unsaturated fatty acid of oils.



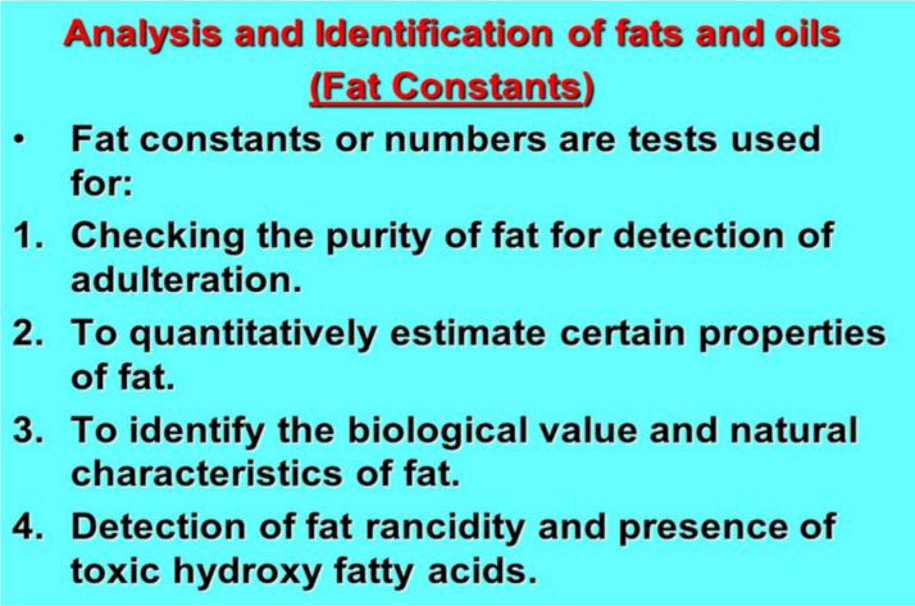


3-Ketonic Rancidity:

- It is due to the contamination with certain fungi such as Asperigillus Niger on fats such as coconut oil.
- Ketones, fatty aldehydes, short chain fatty acids and fatty alcohols are formed.
- Moisture accelerates ketonic rancidity.

- **Prevention of rancidity is achieved by:**

1. Avoidance of the causes (**exposure to light, oxygen, moisture, high temperature and bacteria or fungal contamination**). By keeping fats or oils in well-closed containers in cold, dark and dry place (i.e., **good storage conditions).**
2. Removal of catalysts such as lead and copper that catalyze rancidity.
3. Addition of **anti-oxidants** to prevent peroxidation in fat (i.e., rancidity). They include phenols, naphthols, tannins and hydroquinones. **The most common natural antioxidant is vitamin E that is important *in vitro* and *in vivo*.**

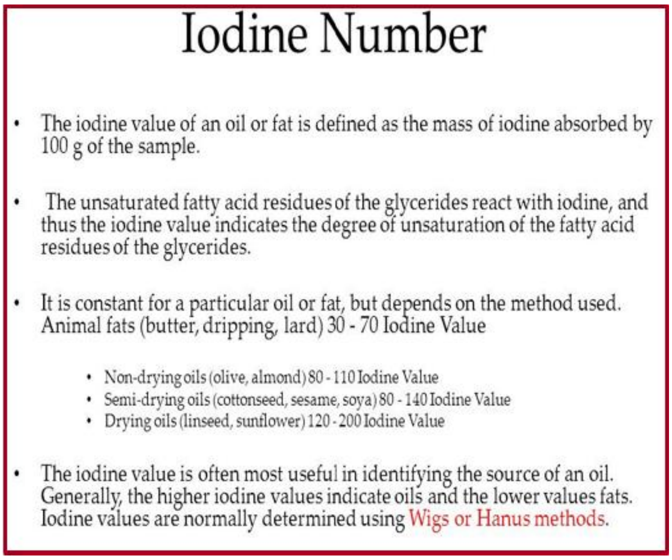


Analysis and Identification of fats and oils

(Fat Constants)

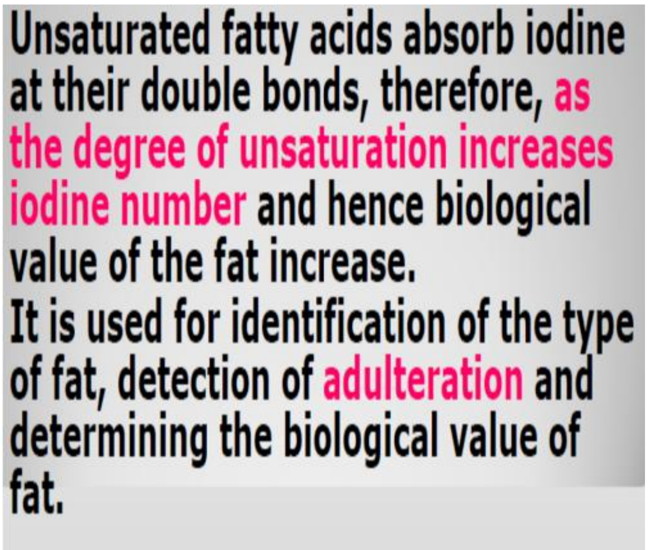
- **Fat constants or numbers are tests used for:**

1. **Checking the purity of fat for detection of adulteration.**
2. **To quantitatively estimate certain properties of fat.**
3. **To identify the biological value and natural characteristics of fat.**
4. **Detection of fat rancidity and presence of toxic hydroxy fatty acids.**



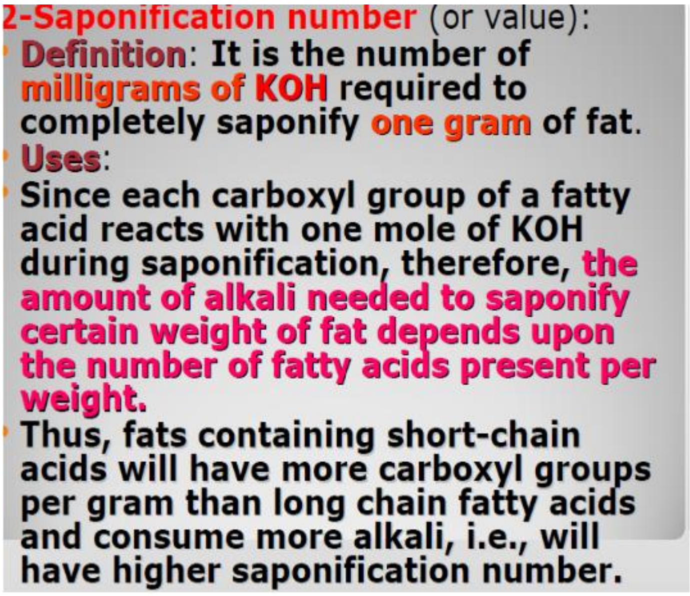
Iodine Number

- The iodine value of an oil or fat is defined as the mass of iodine absorbed by 100 g of the sample.
- The unsaturated fatty acid residues of the glycerides react with iodine, and thus the iodine value indicates the degree of unsaturation of the fatty acid residues of the glycerides.
- It is constant for a particular oil or fat, but depends on the method used. Animal fats (butter, dripping, lard) 30 - 70 Iodine Value
  - Non-drying oils (olive, almond) 80 - 110 Iodine Value
  - Semi-drying oils (cottonseed, sesame, soya) 80 - 140 Iodine Value
  - Drying oils (linseed, sunflower) 120 - 200 Iodine Value
- The iodine value is often most useful in identifying the source of an oil. Generally, the higher iodine values indicate oils and the lower values fats. Iodine values are normally determined using Wigs or Hanus methods.



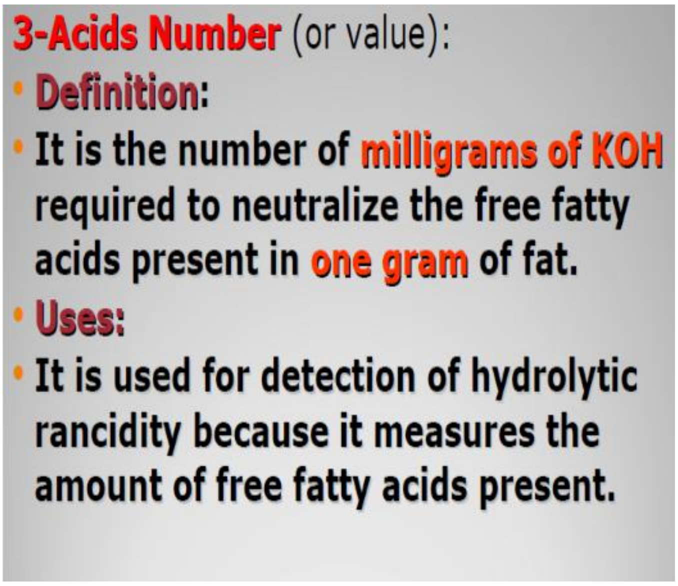
Unsaturated fatty acids absorb iodine at their double bonds, therefore, as the degree of unsaturation increases iodine number and hence biological value of the fat increase.

It is used for identification of the type of fat, detection of adulteration and determining the biological value of fat.



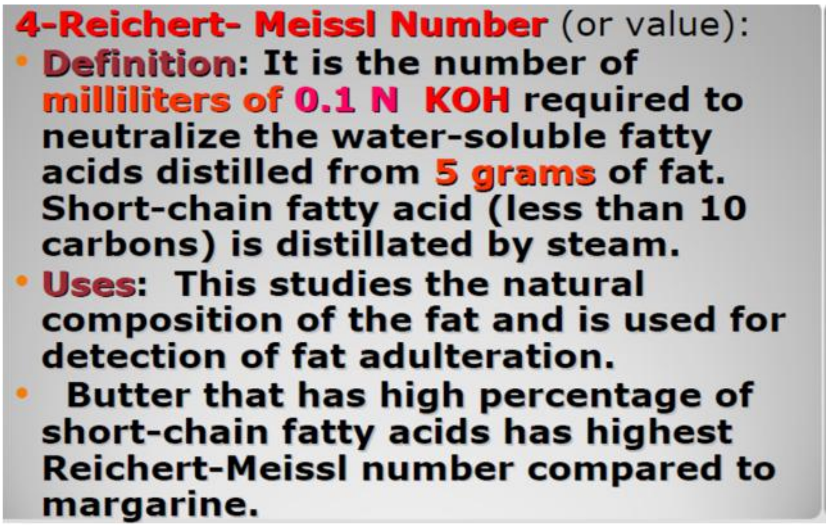
2-Saponification number (or value):

- **Definition**: **It is the number of milligrams of KOH required to completely saponify one gram of fat.**
- **Uses**:
- **Since each carboxyl group of a fatty acid reacts with one mole of KOH during saponification, therefore, the amount of alkali needed to saponify certain weight of fat depends upon the number of fatty acids present per weight.**
- **Thus, fats containing short-chain acids will have more carboxyl groups per gram than long chain fatty acids and consume more alkali, i.e., will have higher saponification number.**



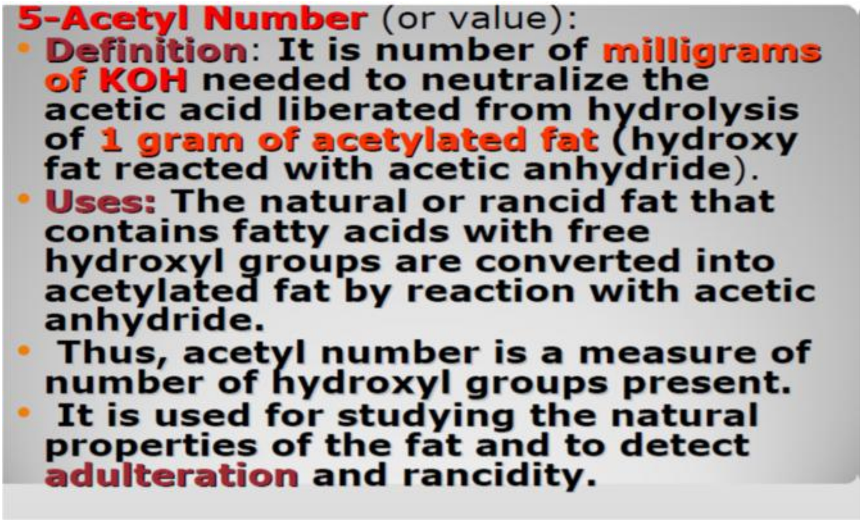
3-Acids Number (or value):

- **Definition:**
- **It is the number of milligrams of KOH required to neutralize the free fatty acids present in one gram of fat.**
- **Uses:**
- **It is used for detection of hydrolytic rancidity because it measures the amount of free fatty acids present.**



4-Reichert- Meissl Number (or value):

- **Definition**: It is the number of **milliliters of 0.1 N KOH** required to neutralize the water-soluble fatty acids distilled from **5 grams** of fat. Short-chain fatty acid (less than 10 carbons) is distilled by steam.
- **Uses**: This studies the natural composition of the fat and is used for detection of fat adulteration.
- Butter that has high percentage of short-chain fatty acids has highest Reichert-Meissl number compared to margarine.



5-Acetyl Number (or value):

- **Definition**: It is number of **milligrams of KOH** needed to neutralize the acetic acid liberated from hydrolysis of **1 gram of acetylated fat** (hydroxy fat reacted with acetic anhydride).
- **Uses:** The natural or rancid fat that contains fatty acids with free hydroxyl groups are converted into acetylated fat by reaction with acetic anhydride.
- Thus, acetyl number is a measure of number of hydroxyl groups present.
- It is used for studying the natural properties of the fat and to detect **adulteration** and rancidity.

FILL UP THE BLANKS

1. At room temperature, the liquid fat contains______
2. The chemical process used to in the manufacture of Vanaspati is ______
3. Saponification is ______ process
4. Hydrogenation of oils gives _______
5. Diester linkage are seen in _____-
6. The building blocks of fats are_______ and _______
7. Sterols contain characteristic cyclic nucleus made up of ____ six member ring and _____ five member ring
8. On hydrolysis, fat gives _____ and ________
9. Hydrolytic rancidity of fat is due to the production of ______
10. An example of essential fatty acid is _______
11. The solid fat contains _______ fatty acid
12. _______ is the chemical process involved in the manufacture of soap
13. The solid alcohol present in the plant is known as ______

14. Arachidonic acid contain _______ number of double bonds
15. Among the three compounds, carbohydrate, lipid and proteins, ______ has more calorific value
16. The higher the unsaturated fatty acid content in oil, ______ will be the iodine value
17. Among oils, ______ has the highest viscosity
18. Vegetable oils are converted to solid fats by the addition of ____ to the double bonds of the unsaturated fatty acids
19. Iodine number of coconut is ______
20. _______ is an example of antioxidant
21. Fat constant _______ is used to measure the free fatty acids in fat
22. Arachidic acid is saturated non-volatile fatty acids found mostly in _____oil
23. The bile of animals contain the steroid namely _____
24. Example of unsaturated fatty acids are _____ and _____-
25. Lecithin is an example of _____
26. Triacyl glycerol is the ester of _____ with glycerol
27. An example of plant sterol is _______
28. Iodine number indicates the degree of ______in a fatty acid
29. A neutral lipid contains _______ as alcohol
30. ____ and _____ are polyunsaturated fatty acids
31. _______ is used as an indicator in iodine value estimation and the end point is _____
32. The most abundant saturated and unsaturated fatty acids are _____ and _____
33. A fatty acid derivatives reacts with a carbohydrate and a ______ base to form ____lipid
34. The two types of glycolipids occurring in nature are______ and _____
35. As chain length of saturated fatty acid increases the ______ and ______ also increases
36. Chemical reaction of fatty acid due to COOH are ____ and _____
37. Oxidative and hydrolytic rancidity can be known by measuring _____ and ____
38. Naturally occurring fatty acids have ______ configuration
39. Fatty acids with less than 10 carbon atoms are ____ and those with more than 10 are_______
40. Metal salts of fatty acids are called _______
41. Functional group in fatty acid is called _____group
42. When fats are hydrolyzed the cleavage occurs at _____linkages
43. The processes involved in the drying of oils are ____ and _____
44. _____ and ______ are the most common fatty acids present in drying oils
45. The general formula of saturated fatty acid is ______ and unsaturated fatty acid is ____
46. The structural formula of mixed triglyceride is _____
47. Tocopherol is another name for _______
48. _____ is an important sterol which is converted to vitamin D on irradiation by sunlight
49. Simple lipids constitute the major groups ____ and _______
50. _______ is a fat solvent generally used for the extraction of lipids
51. Oxidative rancidity is observed more frequently in ____fat than in ____ fat
52. Hydrolytic rancidity of butter is caused by the action of ____ due to secretion by_____-

53. The antioxidants present in the vegetable oil is ____
54. ____ test forms the basis of presence of glycerol in fat molecule
55. Quantity of free fatty acid present in a fat is indicated by_____
56. Number of double bonds present in fatty acid molecule is noticed by_____
57. The acetyl number is a measure of the number of _______groups in the fat
58. Saponification number is highest in ______
59. The iodine number in linseed oil is ______
60. _____ amount of calories is provided by fat
61. _______ is referred to as essential fatty acid
62. Oils and fats are _____
63. Iodine value measures the extent of _____present in fats and oils
64. _____is used to estimate the amount of linoleic and linolenic acids in fats or oils
65. The development of off flavour in fats is known as ________
66. The enzyme responsible for conversion of fats into fatty acid and glycerol is ____
67. The essential fatty acid present in vegetable fat is _____
68. The fatty acid which contain double bond in its structure is referred to as ____
69. Palmitic acid is a ____fatty acid
70. The unsaturated fatty acid have _____melting point
71. Double bond is observed in ______fatty acid
72. Unsaturated fatty acids containing fats show _____isomerism due to presence of double bond
73 In the manufacture of soap, the __________ process is involved
74 Richart-Meissel number measures__________ fatty acids
75 The process of hydrogenation of fatty acid yields _______
76 Waxes are __________ lipids
77 Saponification process yields ___________
78 Phospholipid contains ____, ______ and ________
79 Iodine number indicates degree of ________
80 Hydrogenation is_________ hydrogen
81 Manufacture of Vanaspati involves________ process
82 Capric acid occurs in ________ oil
83 The iodine value of groundnut oil ranges from _________ to ____
84 Complex compounds of fatty acid with carbohydrates are called _______
85 At room temperature oils are ________
86 Iodine number is ____ required to neutralize _______of fat
87 Lecithin is a __________ lipid
88 Esters containing fatty acids, alcohol, phosphoric acid and choline are called ____
89 Fat is estimated by ______ method
90 Soaps are products of________
91 Ergosterol is an example of _______
92 Oleic acid is an example of __________ fatty acid
93 When unsaturation is introduced in stearic acid the melting point _______

94 Arachidonic acid is present in _________

95 Fat is ester of fatty acid with _______

96 The name lipid was suggested by _____

97 Nitrogenous base in lecithin is _________

98 Choline is very _____ in reaction

99 Cephalin is a ______ lipid

100 An example of phospholipids is _______

101 _________ is an antioxidant to prevent oxidative rancidity in vegetable oils

102 ________ is common solvent

103 The most abundant member in the group of complex lipid is ______

104 The development of off flavors due to Aspergillus Niger is called as_____

105 The unsaturated fatty acid containing 4 double bond is________

106 The richest source of essential fatty acid is__________

107 The essential PUFA is ________

108 Examples of monosaturated fatty acid is _________

109 Number of double bond in arachidonic acid is _______

110 The fats and oils are rich in __________ and __________ fatty acids

111 __________ fatty acid has16 carbon atoms

112 __________ is a hydroxyl fatty acid

113 _______ storage form of lipid

114 In which form are most lipids found ______

115 The three-carbon "backbone" found in all triglycerides is called: _______

116 A glycerol backbone with one or two fatty acids plus another group, possibly a phosphorus group, produces a: ________

117 Triglycerides that contain one or more double covalent bonds between carbon atoms of their fatty acids are called __________

118 Bubbling hydrogen gas through polyunsaturated vegetable oil will cause the oil to become more__________ and more_______

119 Chemically, fats and oils are _________

120 Palmitoleic acid (16:1) classified as __________ fatty acid.

121 _________ is classified as sterol

122 Most commonly occurring fatty acid in nature is ________

123 In most naturally occurring monounsaturated fatty acids, the double bond will be placed between _______

124 The number of OH group in fatty acids can be expressed as________

125 The degree of unsaturation of lipids can be measured as _________

ANSWERS

Sl.No	Answer	Sl.No	Answer
1	Unsaturated	34	Galactosyldyglyceride, sulpholipids
2	Hydrogenation	35	Carbon and Hydrogen

3	Alkali hydrolysis	36	hydrolysis and Saponification
4	Solid fats- ghee	37	Peroxide value and Lipase enzyme
5	Compound lipid	38	Monocarboxylic with even number of carbon
6	Fatty acid and glycerol	39	Unsaturated and saturated
7	Pyranose, Furanose	40	Soap
8	fatty acid and glycerol	41	COOH
9	Lipase	42	Fatty acid + glycerol
10	Linoleic/Oleic/Linolenic	43	oxidation and cross linking
11	Saturated	44	α linolenic and linoleic
12	Saponification	45	$CH_2(CH_2)_n\ R-CH=CH(\ CH_2)_n-COOH$
13	Glycerol	46	$CH_3-OOC-R_1$ | $CH-OOC-R_2$ | $CH_2-OOC-R_3$
14	4	47	vitamin E
15	Lipids	48	Ergosterol
16	Greater	49	Fats and waxes
17	Castor oil	50	Ether
18	hydrogen	51	Animal fats, vegetable fat
19	7-10	52	Lipase and microbes
20	Tocopherol	53	Tocopherol
21	Acid value	54	Acrolein test
22	peanut/groundnut	55	Acid number
23	Cholic	56	Iodine number
24	Oleic and Linoleic	57	OH
25	Phosphoglyceride	58	Coconut
26	3 fatty acids	59	170-195
27	Ergosterol	60	9 Kcal
28	Unsaturation	61	PUFA
29	Glycerol	62	Simple
30	Arachidonic and Erucic	63	Unsaturation
31	Starch and blue to colourless	64	Acid value
32	Palmitic acid, Oleic/linoleic acid	65	Rancidity
33	Sulphur, Sulpholipid	66	Lipase
67	Linoleic	100	Lecithin
68	Unsaturated	101	Tocopherols
69	Saturated	102	Ethyl alcohol
70	Low	103	Sphingolipids
71	Unsaturated	104	Ketonic rancidity

72	Cis-Trans	105	Arachidonic acid
73	Saponification	106	Linseed oil
74	Soluble	107	Linoleic acid
75	Dalda	108	Oleic acid
76	Simple	109	4
77	Soap	110	Saturated, unsaturated
78	Fatty acid and glycerol and H_3PO_4	111	Palmitoleic acid
79	Degree of unsaturation	112	Ricinoleic acid
80	Addition	113	Cholesterol or triglycerides
81	Hydrogenation	114	triglycerides
82	Coconut oil	115	Glycerol
83	135-194	116	Phospholipid
84	Glycolipids	117	Unsaturated fatty acid
85	Liquids	118	Saturated , solid
86	Gram iodine, 100 g	119	Esters
87	Phospholipids	120	Monosaturated
88	Phospholipids	121	Cholesterol
89	Saponification	122	Lauric , oleic
90	Fat and alkali	123	Two carbon atoms
91	Plant sterol	124	Acetyl number
92	Unsaturated fatty acid	125	Iodine value
93	Decreases		
94	Peanut		
95	Glycerol		
96	Bloor		
97	Choline		
98	Neutral		
99	Phospho		

WRITE SHORT NOTES ON:

1) **Oxidative rancidity:**

Oils containing highly unsaturated fatty acids are spontaneously oxidized by atmospheric oxygen at ordinary temperature. The oxidation takes place slowly and results in the formation of short chain fatty acids (C4- C10) and aldehydes which give a rancid taste and oil or to the fats. This type of rancidity is called as oxidative rancidity and is due to a reaction called auto oxidation. Oxidative rancidity is observed more frequently in animal fats than in vegetable fats. This is due to the presence of natural antioxidants in vegetable fats such as tocopherol, phenols, and napthols which check autoxidation. Vitamin E is therefore sometimes added to foods to prevent rancidity. The action of antioxidants is opposed by a group of compounds called as pro oxidants present in fats and oils which accelerate the oxidation of the parent compound and these compounds are formed during the processing and refining of fats eg. Copper, nickel, lactic acid**.**

1) **Phospholipids**

It is most abundant membrane lipids, As the name implies, phospholipids contain phosphorus in the form of phosphoric acid. In phospholipids, two of the OH groups in glycerol are linked to fatty acid and third OH is linked to phosphoric acid. Phospholipids are classified into a) Phosphoglycerates b) Phosphoinositides c) Phosphosphingosides. Phosphoglycerates are the major phospholipids found in membrane and contain two fatty acid molecules esterified to the first and second OH groups of glycerol and third OH forms an ester linkage with phosphoric acid. Lecithin, Cephalin and plasmologens belong to this group. Phosphoinositide is found in brain tissues and are considerable importance because of their role in transport processes in cells. It is also called as glycolipids since it has much carbohydrate residues. Phosphosphingosides are commonly found in nerve tissues and apparently lack in plants and microbes. This differ from other phospholipids in their lack of glycerol and the presence of another nitrogenous sphingosins besides choline in place of glycerol.

3. **Sterols**

It is derived lipids. It does not contain fatty acids. So they are non saponifiable. All steroids may be considered as derivatives of a fused and fully saturated ring system called as steranes. Important steroids are sterols, bile acids and hormones. Among the sterols, cholesterol is important (C27) found in animal fat followed by ergosterol (C28) found in yeast which is precursor for vitamin D and tanosterol (C30) found in wool fat required for biosynthesis of cholesterol. The bile acids include cholic acid (C29) present in intestine and gall bladder required for absorption of fats and chenodeoxy cholic acid. The hormones include progesterone (C21) found in corpus luteumm Deoxy corticosterone found in aldrenal coster testosterone (C19) found in testes and esteradiol (C18) present in ovaries.

4. **Chemical constants of oils/ fat or Quantitative test of fats or oils**

The chemical determination on the nature of fatty acids and the number of hydroxyl groups present in the fat molecule is referred to as chemical constants which includes the following

1) **Acid value**- It is the number of milligrams of KOH required to neutralize I gram of the oil or fat. It gives about the amount of free acid present in fat or oil

2) **Saponification value**- It is the number of milligrams of KOH required to neutralize the fatty acid resulting from the complete hydrolysis of I gram of fat or oil. Higher the molecular weight of fat or oil, smaller the Saponification value.

3) **Iodine value**- number of grams of iodine taken up by 100 grams of fat or oil. It gives an idea on degree of unsaturation

4) **RM value**- Number of milliliter of 0.1N KOH solution required to neutralize the volatile soluble acids obtained by the hydrolysis of 5 g of fat. It is used for finding the purity of butter or ghee. It is lower for adulterated ghee than pure ghee

5) **Polenske number**- Number of milliliter of 0.1N KOH required to neutralize the insoluble fatty acid obtained from 5 g fat

6) **Acetyl number**- It is number of millilitres of 0.1N KOH required to neutralize the acetic acid obtained by Saponification of 1 g fat after it has been acetylated. It is the measure of number of OH groups in fat. Castor oil has got the highest acetyl number (146) because of high content of hydroxy ricinoleic acid in it

5. **Rancidity**

Oils and fats on long storage in contact with heat, light, air and moisture develop an unpleasant odor. Such oils and fats are known as rancid oils and fats. The rancidity develops due to certain chemical changes taking place in fat. The changes include

1) **Enzymatic hydrolysis**- In presence of enzymes and microbes, the fats and oils form bad smell lowering fatty acids

2) **Air oxidation of unsaturated fatty acids**- During air oxidation, the unsaturated fatty acid portion of fats are oxidized at the site of the double bonds into aldehyde and ketones with unpleasant odor

3) **β oxidation of saturated fatty acids**- The saturated fatty acids undergoes β oxidation followed by decarboxylation to form ketones of unpleasant Odor

6. **Functions of lipids/importance of lipids**

1) Fats serve reserve food in seeds
2) Oils are used by human beings for various purposes
3) Lipids provides the structural frame work to the living tissues of plants and animals
4) Lipids provides as the prime fuel reserve for metabolism and provides more energy than carbohydrates and proteins
5) Fats provides a bridge in filling the gap between water soluble and insoluble phases
6) Waxes give a protective covering on the upper surface of leaves, stems and fruits
7) Certain oils like castor oil, mustard oil, clove oils and coconut oils are used medicinally
8) Other oils are used in the preparation of soaps and vegetable ghee
9) Lipids acts as carrier of natural fat soluble vitamins A D E K
10) The sex hormones, adreno corticoids, cholic acids and vitamin D are all synthesized from cholesterol
11) Phospholipids play an important role in the absorption and translocation of fatty acids
12) The fats are characterized for their high insulating capacity

7. **Compound lipids**

The compound lipids consists of phospholipids and glycolipids

A) Phospholipids:

Phospholipids are those lipids which contains phosphorus. Phosphorus is present in the form of esterified phosphoric acid. Based on the alcoholic component present, the phospholipids are classified into

1) Phosphoglyceride (glycerol containing) - It is the largest and most wide spread class. It is again subdivided into the following groups

i) Phosphatidyl cholines – lecithin, cephalin
ii) Phosphatidyl ethanolamine
iii) Phosphatidyl series
iv) Plasmologens

v) Phosphatidyl glycerols

2) Phosphoinositides (Inositol containing) – It is made up of glycerol, fatty acid, phosphoric acid and cyclic hexahydric alcohol mesoinositol. It is divided into i) Mono ii) Di. Mono is found widely in both plants and animals while Di occur only in brain tissues

3) **Phosphosphingosides-** It contains a alcohol phytosphingosine in plants and sphingosine in animals. It differs from phytosphingosine in having double bond

B) Glycolipids

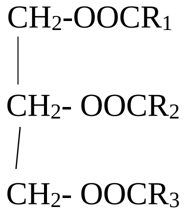
Glycolipids are those lipids which contain carbohydrates. They have been isolated from different plant sps. Galactosyl diglycerides and sulpholipids form the major Glycolipids in plants. Galactosyl diglycerides may be mono or di and it has been isolated from chloroplast of many plants. The plant sulpholipid was discovered by the use of radioactive sulfur by Benson in the leaves of higher plants. It is made up of glycerol, fatty acid and sugar quinovose which bears S containing group. The plant sulpholipids contains sulphonic acid

8) Lipoproteins

Lipoproteins are complexes which contain lipids and proteins in association. It is most important complexes because in the living being they are found in the form of soluble lipid drops and build the plasma membranes and membranes of various cell organelles. The membranes of these cell –organelle possess a number of enzymes for biochemical reactions. The lipoproteins of the membranes may be water soluble or fat soluble. They help in the entry or exit of water and fat soluble compounds and also maintain the concentration and equilibrium for such compounds in the cell. The lipoproteins complexes are affected by agents that influence the purity of proteins such as heat, pH, and other chemicals. Lipoproteins provide surface for biochemical reactions and energy production and also help in the transportation of lipids and proteins to the various parts of the plant or animal body.

9. Fats and oils

It is simple lipids. They are called as triglycerides. It is the most abundant of all lipids. Chemically triglycerides are esters of glycerol and 3 fatty acids molecules.

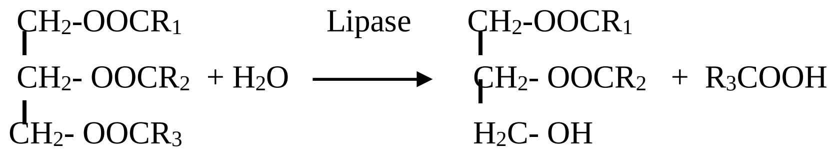


$$\begin{array}{l} CH_2\text{-}OOCR_1 \\ | \\ CH_2\text{- }OOCR_2 \\ | \\ CH_2\text{- }OOCR_3 \end{array}$$

In triglycerides, all the three fatty acids molecules may be identical or may be different. All the fat contains glycerol in common but differs in amount and types of fatty acids. Triglycerides which contain saturated fatty acids are solid at room temperature and are called as fats. Other triglycerides which contain unsaturated fatty acids are liquid at room temperature and are called as oils. Oils can be converted to solid fats by saturating the fatty acids. This process is utilized in the manufacture of vegetable ghee from oils. The animal fat such as meat, milk and eggs contain high amount of saturated fatty acids and high melting point. So it is solid in state. The plant fats contains high amount of polyunsaturated fatty acids and low melting point, so it is liquid at state.

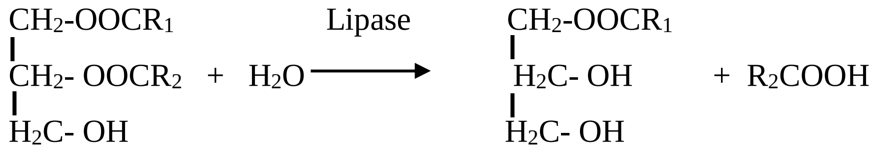
10. **Action of lipase enzyme**

The fats are hydrolyzed by the enzyme lipases to yield fatty acid and glycerol. The lipases catalyses this reaction at a slightly alkaline pH (7.5-8.6) in a step wise manner. The fats are first split into produce diglycerides, part of these are then split to monoglycerides. Finally part of the monoglycerides split to yield fatty acids and glycerol



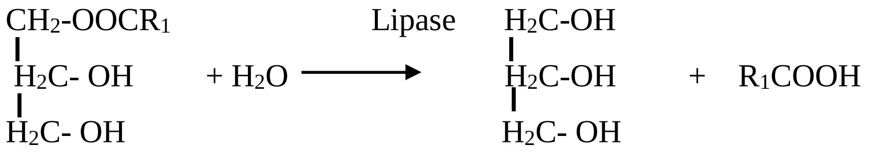
$$\begin{array}{l} CH_2\text{-}OOCR_1 \\ | \\ CH_2\text{-}OOCR_2 \\ | \\ CH_2\text{-}OOCR_3 \end{array} + H_2O \xrightarrow{Lipase} \begin{array}{l} CH_2\text{-}OOCR_1 \\ | \\ CH_2\text{-}OOCR_2 \\ | \\ H_2C\text{-}OH \end{array} + R_3COOH$$

Triglycerides **diglycerides** **fatty acid**



$$\begin{array}{l} CH_2\text{-}OOCR_1 \\ | \\ CH_2\text{-}OOCR_2 \\ | \\ H_2C\text{-}OH \end{array} + H_2O \xrightarrow{Lipase} \begin{array}{l} CH_2\text{-}OOCR_1 \\ | \\ H_2C\text{-}OH \\ | \\ H_2C\text{-}OH \end{array} + R_2COOH$$

Diglycerides **Monoglycerides** **fatty acid**

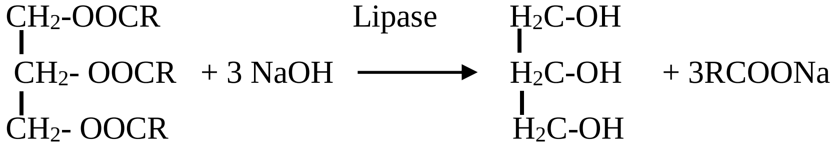


$$\begin{array}{l} CH_2\text{-}OOCR_1 \\ | \\ H_2C\text{-}OH \\ | \\ H_2C\text{-}OH \end{array} + H_2O \xrightarrow{Lipase} \begin{array}{l} H_2C\text{-}OH \\ | \\ H_2C\text{-}OH \\ | \\ H_2C\text{-}OH \end{array} + R_1COOH$$

Monoglycerides **Glycerol** **fatty acid**

11) **Saponification**

Hydrolysis of fat by alkali such as NaOH or KOH leads to the formation of sodium or potassium salts of fatty acid. These salts are known as soaps and process of its formation is known as Saponification. The soaps are of two types a) hard b) soft. Hard soap such as bar soaps is sodium salts of higher fatty acids. Soft soaps are the potassium salts of higher fatty acids and marketed as semisolids or pastes



$$\begin{array}{l} CH_2\text{-}OOCR \\ | \\ CH_2\text{-}OOCR \\ | \\ CH_2\text{-}OOCR \end{array} + 3\,NaOH \xrightarrow{Lipase} \begin{array}{l} H_2C\text{-}OH \\ | \\ H_2C\text{-}OH \\ | \\ H_2C\text{-}OH \end{array} + 3RCOONa$$

Fat Glycerol Na salts of fatty acid (Soap)

12. **Hydrogenation of fats**

Oils containing unsaturated fatty acids can be hydrogenated in the presence of high temperature, pressure and finally divided nickel. Unsaturated fatty acids either free or combined in lipids react with gaseous hydrogen to yield the saturated fatty acids. The addition of hydrogen takes place at the C-C double bonds. Thus one mole of oleic, linoleic or linolenic acid react with 1,2 or 3 moles of hydrogen respectively to form Stearic acid .

$$CH_3\,(CH_2)_7CH\,(CH_2)_7COOH + H_2 \xrightarrow[\text{Catalyst}]{\text{Pu/Pd/Ni}} CH_3\,(CH_2)_7CH_2\,\text{–}CH_2(\,CH_2)_7COOH$$

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

13. **Physical properties of fats**

    1) **State –** Fats containing saturated fatty acid are solid and fats containing unsaturated fatty acids are liquid at room temperature
    2) **Color, Odor and Taste-** Fats are colorless, odorless and has bland taste
    3) **Solubilit**y- Fats are soluble in organic solvents like ether, chloroform, benzene and insoluble in water
    4) **Melting point**- Saturated glycerides containing fat require high temperature for melting and unsaturated glycerides containing fats require lower melting point.
    5) **Specific gravity –** The specific gravity of fats is less than 1 and therefore they float on the water surface
    6) **Geometric isomerism-** Unsaturated fatty acids containing fats show cis-trans isomerism due to presence of double bond
    7) I**nsulation-** They are bad conductors of heat
    8) **Surface tension –** Fats redues surface tension

14. **Chemical properties**

    1) **Hydrolysis**- Fats undergo hydrolysis when they are treated with mineral acids, alkalies, or fat splitting enzyme lipase or hydrolases to yield glycerol and fatty acid
    2). **Hydrogenation-** Oils containing unsaturated fatty acids can be hydrogenated in the presence of high temperature, pressure and finally divided nickel. By this oils are converted to solid fats
    3) **Hydrogenolysis-** Oils and fats are converted to glycerol and a long chain aliphatic alcohol when excess of hydrogen is passed through them under pressure and in the presence of copper chromium catalyst. The splitting of fat by hydrogen is called Hydrogenolysis
    4) **Halogenation**- When unsaturated fatty acids are treated with halogens such as iodine and chlorine, they take up halogens at the double bond site. This process of taking iodine is called as halogenation and it is an indication of unsaturation.
    5) **Rancidity**- Oils and fats on long storage in contact with heat, light, air and moisture develop an unpleasant odor, such oils and fats are called as rancid fats and oils.
    6) **Emulsification**- The process of breaking of large size fat molecules in to smaller ones is known as emulsification

ESSAY TYPES

1. **What are essential fatty acids? Define different chemical constants and their significance in relation to fatty acid composition in fats and oils**

   Essential fatty acids are group of polyunsaturated fatty acids produced by plants, but not by humans, required in the human diet. Following are the chemical constants which give valuable information about the chemical nature of fatty acids and number of hydroxyl groups present in the fat molecule

1. **Acid value**- It is the number of milligrams of KOH required to neutralize I gram of the oil or fat. It gives about the amount of free acid present in fat or oil
2. **Saponification value**- It is the number of milligrams of KOH required to neutralize the fatty acid resulting from the complete hydrolysis of I gram of fat or oil. Higher the molecular weight of fat or oil, smaller the Saponification value.
3. **Iodine value**- number of grams of iodine taken up by 100 grams of fat or oil. It gives an idea on degree of unsaturation
4. **RM value**- Number of milliliter of 0.1N KOH solution required to neutralize the volatile soluble acids obtained by the hydrolysis of 5 g of fat. It is used for finding the purity of butter or ghee. It is lower for adulterated ghee than pure ghee
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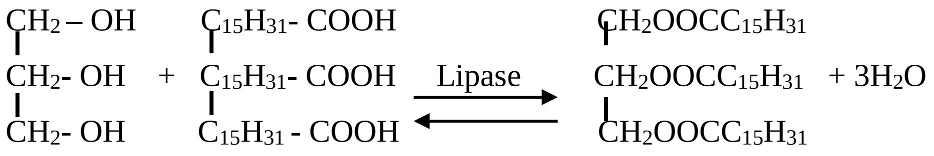
2. Write on neutral lipids and their important chemical reactions

The neutral lipids or triglycerides are esters of the trihydric alcohol, glycerol and fatty acids. In nature, three molecules of fatty acids combines with one molecule of glycerol with the elimination of three molecules of water. The enzyme lipase catalyses the reaction



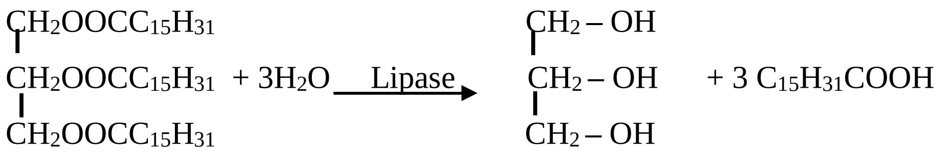
$$\begin{array}{l} CH_2-OH \\ | \\ CH_2-OH \\ | \\ CH_2-OH \end{array} + \begin{array}{l} C_{15}H_{31}-COOH \\ | \\ C_{15}H_{31}-COOH \\ | \\ C_{15}H_{31}-COOH \end{array} \overset{\text{Lipase}}{\rightleftharpoons} \begin{array}{l} CH_2OOCC_{15}H_{31} \\ | \\ CH_2OOCC_{15}H_{31} \\ | \\ CH_2OOCC_{15}H_{31} \end{array} + 3H_2O$$

Glycerol **Palmitic acid** **Tripalmitic acid (fat)**

In triglycerides, all the three fatty acids molecules may be identical or may be different. All the fats contain glycerol in common, but differ in amount and types of fatty acids. The triglycerides which contain saturated fatty acids are solid at room temperature and are called fats. Other triglycerides which contain unsaturated fatty acids are liquid at room temperature are called oils. Most animal fats are rich in saturated fatty acid while plant fats contain a large proportion of unsaturated fatty acids.

The important chemical reactions are

1) **Hydrolysis**- Fats undergo hydrolysis when they are treated with mineral acids, alkalies, or fat splitting enzyme lipase or hydrolases to yield glycerol and fatty acid.



$$\begin{array}{l} CH_2OOCC_{15}H_{31} \\ | \\ CH_2OOCC_{15}H_{31} \\ | \\ CH_2OOCC_{15}H_{31} \end{array} + 3H_2O \xrightarrow{\text{Lipase}} \begin{array}{l} CH_2-OH \\ | \\ CH_2-OH \\ | \\ CH_2-OH \end{array} + 3\ C_{15}H_{31}COOH$$

Tripalmitic acid (fat) Glycerol Palmitic acid

2). **Hydrogenation-** Oils containing unsaturated fatty acids can be hydrogenated in the presence of high temperature, pressure and finally divided nickel. By this oils are converted to solid fats

3) **Hydrogenolysis**- Oils and fats are converted to glycerol and a long chain aliphatic alcohol when excess of hydrogen is passed through them under pressure and in the presence of copper chromium catalyst. The splitting of fat by hydrogen is called Hydrogenolysis

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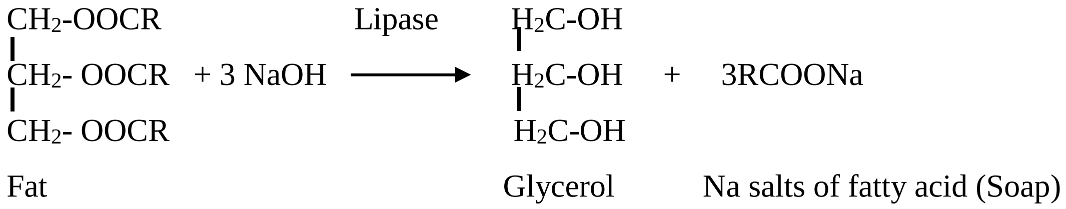
6) **Emulsification**- The process of breaking of large size fat molecules in to smaller ones is known as emulsification

7) **Saponification**- Hydrolysis by alkali such as NaOH or KOH leads to formation of sodium or potassium salts of fatty acids called as soaps and process of its formation is known as Saponification



$$\begin{array}{l} CH_2\text{-}OOCR \\ | \\ CH_2\text{-}OOCR \\ | \\ CH_2\text{-}OOCR \end{array} + 3\,NaOH \xrightarrow{\text{Lipase}} \begin{array}{l} H_2C\text{-}OH \\ | \\ H_2C\text{-}OH \\ | \\ H_2C\text{-}OH \end{array} + 3RCOONa$$

Fat Glycerol Na salts of fatty acid (Soap)

3. **Explain the classification of lipids with suitable examples**

Bloor (1943) classified the lipids based on the chemical composition

A) Simple lipids or homolipids

a) Fats and oils (Triglycerides) → Simple → tripalmitin
→ Mixed

b) Waxes → Sperm whale wax

B) Compound lipid or heterolipids

Phospholipids → Phosphoglycerides- Lecithin, cephalin
→ Phosphoinositides-
→ Phosphosphingosides

C) Derived Lipids → Steroids
→ Terpens- Mono, Di, tri, tetra and poly
→ Carotenoids- lycopene, carotenes, xanthophylls

4. **List the important properties of fatty acids**

   Physical properties of fats

   1) State – Fats containing saturated fatty acid are solid and fats containing unsaturated fatty acids are liquid at room temperature
   2) Color, Odor and Taste- Fats are colorless, odorless and has bland taste
   3) Solubility- Fats are soluble in organic solvents like ether, chloroform, benzene and insoluble in water
   4) Melting point- Saturated glycerides containing fat require high temperature for melting and unsaturated glycerides containing fats require lower melting point.
   5) Specific gravity – The specific gravity of fats is less than 1 and therefore they float on the water surface
   6) Geometric isomerism- Unsaturated fatty acids containing fats show cis-trans isomerism due to presence of double bond
   7) Insulation- They are bad conductors of heat
   8) Surface tension – Fats redues surface tension

   Chemical properties

   1) Hydrolysis- Fats undergo hydrolysis when they are treated with mineral acids, alkalies, or fat splitting enzyme lipase or hydrolases to yield glycerol and fatty acid

   2). Hydrogenation- Oils containing unsaturated fatty acids can be hydrogenated in the presence of high temperature, pressure and finally divided nickel. By this oils are converted to solid fats

   3) Hydrogenolysis- Oils and fats are converted to glycerol and a long chain aliphatic alcohol when excess of hydrogen is passed through them under pressure and in the presence of copper chromium catalyst. The splitting of fat by hydrogen is called Hydrogenolysis

   4) Halogenation- When unsaturated fatty acids are treated with halogens such as iodine and chlorine, they take up halogens at the double bond site. This process of taking iodine is called as halogenation and it is an indication of unsaturation.

   5) Rancidity- Oils and fats on long storage in contact with heat, light, air and moisture develop an unpleasant odor, such oils and fats are called as rancid fats and oils.

   6) Emulsification- The process of breaking of large size fat molecules in to smaller ones is known as emulsification

5. **List the saturated fatty acids with their molecular formula and suitable examples**

   1) Butyric acid- $CH_3(CH_2)_2COOH$- butter
   2) Caproic acid- $CH_3(CH_2)_4COOH$ – Butter, palm oil, coconut oil
   3) Caprylic acid- $CH_3(CH_2)_6COOH$- Palm oil, coconut oil
   4) Capric acid - $CH_3(CH_2)_8COOH$ – Palm oil, coconut oil
   5) Lauric acid - $CH_3(CH_2)_{10}COOH$ – Lauraceae, palm oil, coconut oil
   6) Myristic acid - $CH_3(CH_2)_{12}COOH$ – Seed fats, butter
   7) Palmitic acid - $CH_3(CH_2)_{14}COOH$ – peanut oil, palm oil
   8) Stearic acid - $CH_3(CH_2)_{16}COOH$ – Plant and animal fat
   9) Arachidic acid - $CH_3(CH_2)_{18}COOH$- Peanut oil
   10) Lerotic acid- $CH_3(CH_2)_{24}COOH$ – Beewax

Definition

1. **Lipids:** Lipids are chemically heterogeneous group of compounds that are insoluble in water but soluble in non-polar solvents such as chloroform
2. **Fat**: Any of a group of natural esters of glycerol and various fatty acids, which are solid at room temperature and are the main constituents of animal and vegetable fat
3. **Oil:** An oil is any neutral, nonpolar chemical substance that is a viscous liquid at ambient temperatures and is both hydrophobic and lipophilic
4. **Fatty acid**: A carboxylic acid consisting of a hydrocarbon chain and a terminal carboxyl group, especially any of those occurring as esters in fats and oils.
5. **Saturated fatty acid**: A saturated fat is a type of fat in which the fatty acid chains have all or predominantly single bonds
6. **Unsaturated fatty acid**: An unsaturated fat is a fat or fatty acid in which there is at least one double bond within the fatty acid chain
7. **Essential fatty acid**: An unsaturated fatty acid that is essential to human health, but cannot be manufactured in the body. Abbreviated EFA
8. **Triglyceride:** An ester formed from glycerol and three fatty acid groups. Triglycerides are the main constituents of natural fats and oils.
9. **Simple lipids:** Lipids containing only fatty acids and glycerol or long chain alcohols
10. (Monohydric) are called as simple lipids which include fats, oils and waxes
11. **Compound lipids:** Esters of fatty acids with alcohol and possess additional groups also. These group of lipids include glycerophospholipids, sphingo phospholipids, glycolipids, sulpholipids and lipoproteins
12. **Lecithin**: Lecithin contains glycerol, fatty acids, phosphoric acid and a nitrogenous base, choline
13. **Sterols:** Any of a group of naturally occurring unsaturated steroid alcohols, typically waxy solids
14. **Rancidity**: Development of disagreeable odour and taste in fat or oil upon storage is called rancidity.
15. **Hydrolytic rancidity:** Hydrolytic rancidity refers to the Odor that develops when triglycerides are hydrolysed and free fatty acids are released**.**
16. **Oxidative rancidity**: The unsaturated fatty acids are oxidised at the double bonds to form peroxides, which then decompose to form aldehydes and acids of objectionable odour and taste.
17. **Ketonic rancidity**: Some moulds (Penicillium and Aspergillus spp.) attack fats containing short-chain fatty acids and produce ketones with a characteristic odour and taste. Butter, coconut, and palm kernel oils are most susceptible.
18. **Hydrogenation:** Unsaturated fatty acids may be converted to saturated fatty acids by the relatively simple hydrogenation reaction.
19. **Hardening:** Hydrogenation of unsaturated fats in the presence of a catalyst is known as hardening.
20. **Saponification number:** It is defined as milligrams of KOH required to saponify 1 gm of fat or oil.

21. **Iodine Number:** It is defined as the number of grams of iodine taken up by 100 grams of fat or oil.
22. **Reichert-Merisel number: N**umber of millilitres of 0.1 N alkali required to neutralise
23. the soluble volatile fatty acids contained in 5 gm of fat.
24. **Polanski number:** Number of millilitres of 0.1 N potassium hydroxide solution required to neutralise the insoluble fatty acids (not volatile with steam distillation) obtained from 5 gm of fat.
25. **Acetyl number**: Amount in millilitres of potassium hydroxide solution required to neutralise the acetic acid obtained by saponification of 1 gm of fat or oil after acetylation.
26. **Acid number/Acid value**: The milligram of potassium hydroxide required to neutralise
27. the free fatty acids present in one gram of fat or oil.
28. **Iodine value:** Measure of the degree of unsaturation of an oil, fat, or wax; the amount of iodine, in grams, that is taken up by 100 grams of the oil, fat, or wax**.**
29. **Peroxide value**: The peroxide value is defined as the amount of peroxide oxygen per 1 kilogram of fat or oil

METABOLISM

Metabolism, the sum of the chemical reactions that take place within each cell of a living organism and that provide energy for vital processes and for synthesizing new organic material

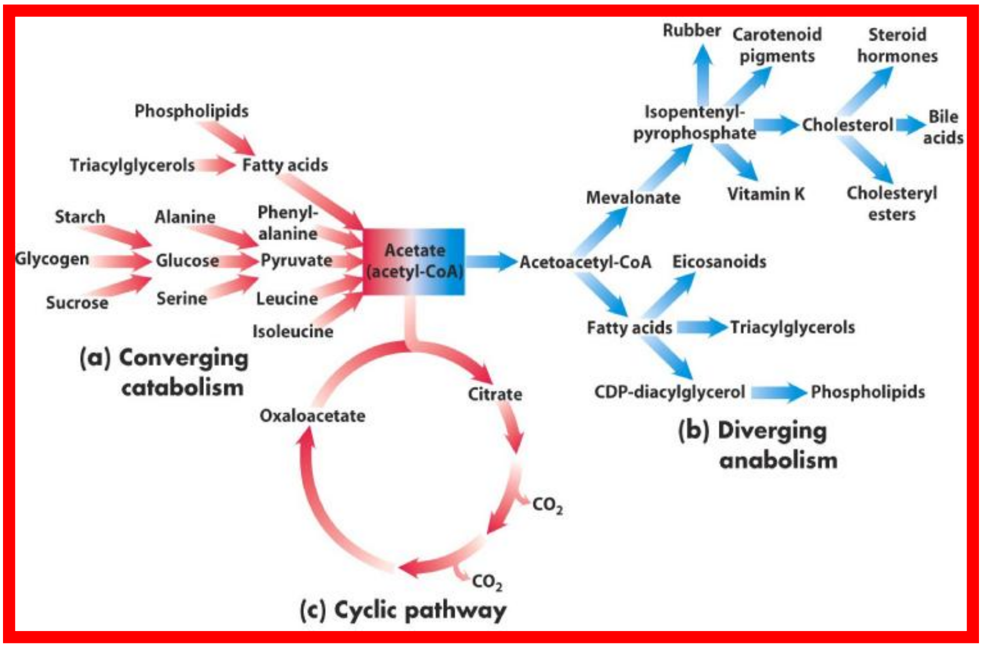
Two types of metabolic reactions take place in the cell: 'building up' (anabolism) and 'breaking down' (catabolism

Catabolism is the breaking down of things - a series of chemical reactions that break down complex molecules into smaller units; catabolic processes usually release energy. the breaking down of organic matter for example, the breaking down of glucose to pyruvate, by cellular respiration,

Anabolism: The build-up of complex organic molecules from simpler ones, reactions are called anabolic or biosynthetic. They involve dehydration synthesis (release water) and are endergonic. The building up of components of cells such as proteins and nucleic acids

The chemical reactions of metabolism are organized into metabolic pathways, in which one chemical is transformed through a series of steps into another chemical, by a sequence of enzymes. Enzymes are crucial to metabolism because they allow organisms to drive desirable reactions that require energy that will not occur by themselves, by coupling them to spontaneous reactions that release energy. Enzymes act as catalysts that allow the reactions to proceed more rapidly. Enzymes also allow the regulation of metabolic pathways in response to changes in the cell's environment or to signals from other cells.

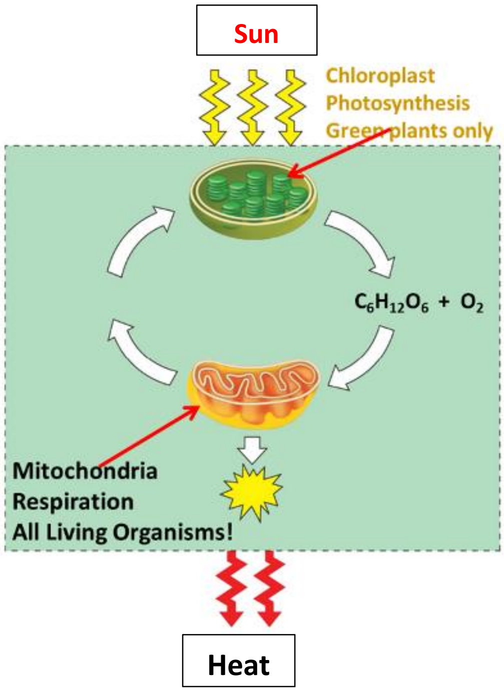
Connection between catabolic and anabolic pathways



The Relationship between Photosynthesis and Respiration

Photosynthesis generates the oxygen and glucose used by the mitochondria of eukaryotes as fuel for: cellular respiration. Cellular respiration breaks down glucose into simpler substances and releases the stored energy. Some of this energy is used to make ATP from ADP. Some of this energy is lost as heat. The waste products of respiration, carbon dioxide and water are the raw materials for photosynthesis.

H. IMPORTANT NOTE: While only green plants carry out photosynthesis, ALL living things carry out respiration

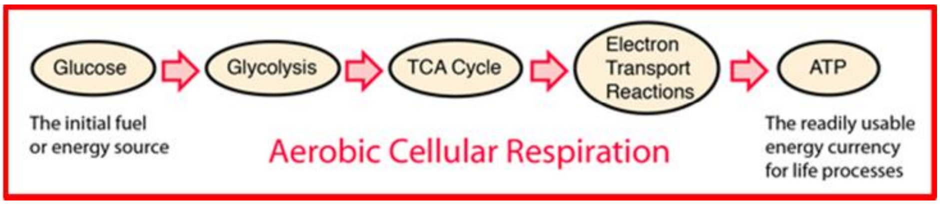


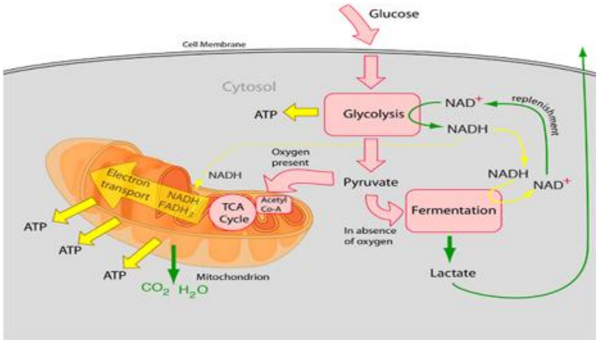
CARBOHYDRATES

Cellular Respiration

The term cellular respiration refers to the biochemical pathway by which cells release energy from the chemical bonds of food molecules and provide that energy for the essential processes of life. All living cells must carry out cellular respiration. It can be aerobic respiration in the presence of oxygen or anaerobic respiration

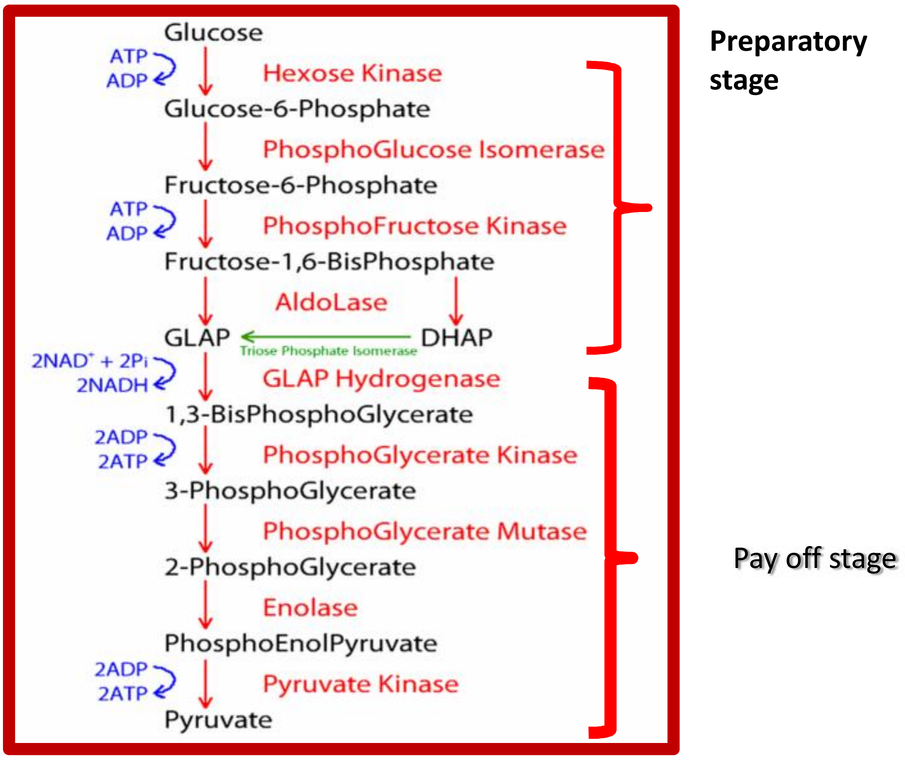
The energy currency of these cells is ATP, and one way to view the outcome of cellular respiration is as a production process for ATP.





Carbohydrate metabolism is a fundamental biochemical process that ensures a constant supply of energy to living cells. The most important carbohydrate is glucose, which can be broken down via glycolysis, enter into the Kreb's cycle and oxidative phosphorylation to generate ATP.

Further important pathways in carbohydrate metabolism include the pentose phosphate pathway (conversion of hexose sugars into pentoses), glycogenesis (conversion of excess glucose into glycogen, stimulated by insulin), glycogenolysis (conversion of glycogen polymers into glucose, stimulated by glucagon) and gluconeogenesis (.a metabolic pathway that results in the generation of glucose from non-carbohydrate carbon substrates such as lactate, glycerol, and glucogenic amino acids.



Definition:

Glycolysis can be defined as the sequence of reactions for the breakdown of Glucose (6-carbon molecule) to two molecules of pyruvic acid (3-carbon molecule) under aerobic conditions; or lactate under anaerobic conditions along with the production of small amount of energy

In glycolysis, also referred to as the Embden-Meyerhof-Parnas pathway, each glucose molecule is split and converted to two three-carbon units (pyruvate).

It is derived from Greek word glycose -sweet or sugar, lysis- dissolution.

Site: Cytosolic fraction of cell

Glycolysis literally means "splitting sugars" and is the process of releasing energy within sugars. In glycolysis, glucose (a six carbon sugar) is split into two molecules of the three-carbon sugar pyruvate. This multi-step process yields two molecules of ATP (free energy containing molecule), two molecules of pyruvate, and two "high energy" electron carrying molecules of NADH. Glycolysis can occur with or without oxygen

Glycolysis, which consists of 10 reactions, occurs in two stages:

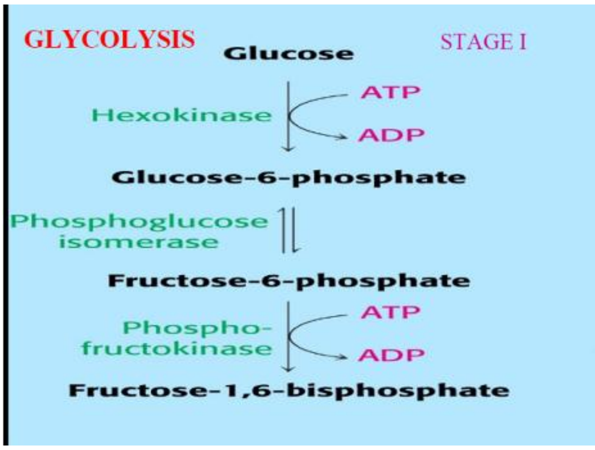
(Preparatory stage and Pay off stage)

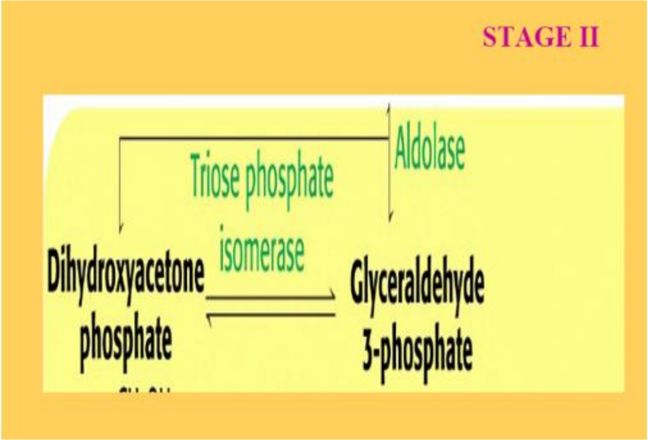
1 Glucose is phosphorylated twice and cleaved to form two molecules of glyceraldehyde-3-phosphate (G-3-P). The two ATP molecules consumed during this stage are like an investment, because this stage creates the actual substrates for oxidation in a form that is trapped inside the cell.

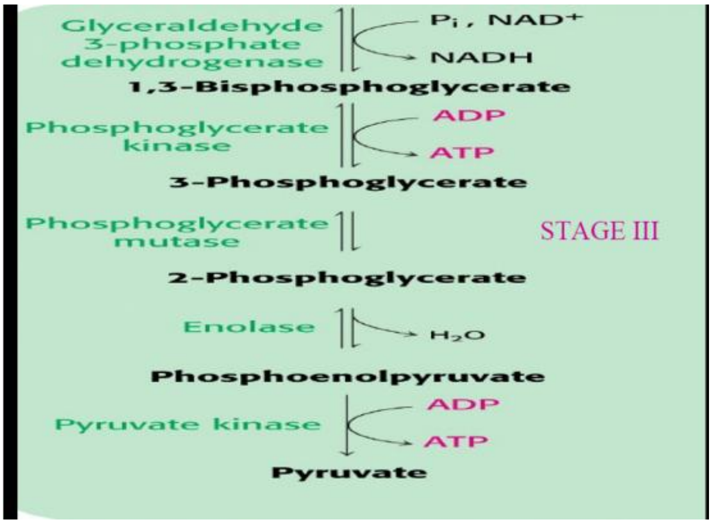
2 Glyceraldehyde-3-phosphate is converted to pyruvate. Four ATP and two NADH molecules are produced. Because two ATP were consumed in stage 1, the net production of ATP per glucose molecule is 2.

The glycolytic pathway can be summed up in the following equation:

D-Glucose + 2 ADP + 2 Pi + 2 NAD → 2 pyruvate + 2 ATP + 2 NADH + 2H + $2H_2O$







Step 1

The enzyme hexokinase phosphorylates (adds a phosphate group to) glucose in the cell's cytoplasm. In the process, a phosphate group from ATP is transferred to glucose producing glucose 6-phosphate.

Glucose (C6H12O6) + hexokinase + ATP → ADP + Glucose 6-phosphate ($C_6H_{13}O_9P$)

Step 2

The enzyme phosphoglucoisomerase converts glucose 6-phosphate into its isomer fructose 6-phosphate. Isomers have the same molecular formula, but the atoms of each molecule are arranged differently.

Glucose 6-phosphate ($C_6H_{13}O_9P$) + Phosphoglucoisomerase → Fructose 6-phosphate ($C_6H_{13}O_9P$)

Step 3

The enzyme phosphofructokinase uses another ATP molecule to transfer a phosphate group to fructose 6-phosphate to form fructose 1, 6-bisphosphate.

Fructose 6-phosphate ($C_6H_{13}O_9P$) + phosphofructokinase + ATP → ADP + Fructose 1, 6-bisphosphate ($C_6H_{14}O_{12}P_2$

Step 4

The enzyme aldolase splits fructose 1, 6-bisphosphate into two sugars that are isomers of each other. These two sugars are dihydroxyacetone phosphate and glyceraldehyde phosphate.

Fructose 1, 6-bisphosphate ($C_6H_{14}O_{12}P_2$) + aldolase → Dihydroxyacetone phosphate ($C_3H_7O_6P$) + Glyceraldehyde phosphate ($C_3H_7O_6P$)

Step 5

The enzyme triose phosphate isomerase rapidly inter-converts the molecules dihydroxyacetone phosphate and glyceraldehyde 3-phosphate. Glyceraldehyde 3-phosphate is removed as soon as it is formed to be used in the next step of glycolysis.

Dihydroxyacetone phosphate ($C_3H_7O_6P$) → Glyceraldehyde 3-phosphate ($C_3H_7O_6P$)

Step 6

The enzyme triose phosphate dehydrogenase serves two functions in this step. First the enzyme transfers a hydrogen (H-) from glyceraldehyde phosphate to the oxidizing agent Nicotinamide adenine dinucleotide (NAD+) to form NADH. Next triose phosphate dehydrogenase adds a phosphate (P) from the cytosol to the oxidized glyceraldehyde phosphate to form 1, 3-bisphosphoglycerate. This occurs for both molecules of glyceraldehyde 3-phosphate produced in step 5.

A. Triose phosphate dehydrogenase + 2 H- + 2 NAD+ → 2 NADH + 2 H+

B. Triose phosphate dehydrogenase + 2 P + 2 glyceraldehyde 3-phosphate ($C_3H_7O_6P$) → 2 molecules of 1,3-bisphosphoglycerate ($C_3H_8O_{10}P_2$)

Step 7

The enzyme phosphoglycerokinase transfers a P from 1,3-bisphosphoglycerate to a molecule of ADP to form ATP. This happens for each molecule of 1,3-bisphosphoglycerate. The process yields two 3-phosphoglycerate molecules and two ATP molecules.

2 molecules of 1, 3-bisphoshoglycerate ($C_3H_8O_{10}P_2$) + phosphoglycerokinase + 2 ADP → 2 molecules of 3-phosphoglycerate ($C_3H_7O_7P$) + 2 ATP

Step 8

The enzyme phosphoglyceromutase relocates the P from 3-phosphoglycerate from the third carbon to the second carbon to form 2-phosphoglycerate.

2 molecules of 3-Phosphoglycerate ($C_3H_7O_7P$) + phosphoglyceromutase → 2 molecules of 2-Phosphoglycerate ($C_3H_7O_7P$)

Step 9

The enzyme enolase removes a molecule of water from 2-phosphoglycerate to form phosphoenolpyruvate (PEP). This happens for each molecule of 2-phosphoglycerate

2 molecules of 2-Phosphoglycerate ($C_3H_7O_7P$) + enolase → 2 molecules of phosphoenolpyruvate (PEP) ($C_3H_5O_6P$

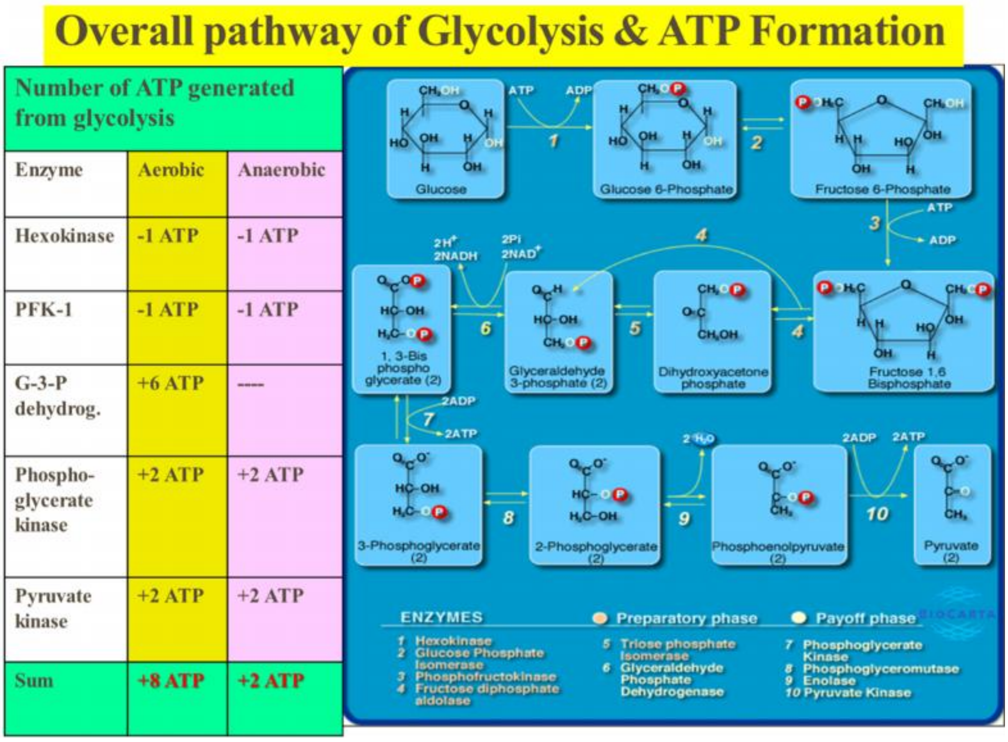
Step 10

The enzyme pyruvate kinase transfers a P from PEP to ADP to form pyruvate and ATP. This happens for each molecule of phosphoenolpyruvate. This reaction yields 2 molecules of pyruvate and 2 ATP molecules.

2 molecules of phosphoenolpyruvate ($C_3H_5O_6P$) + pyruvate kinase + 2 ADP → 2 molecules of pyruvate ($C_3H_3O_3$-) + 2 ATP

The glycolytic pathway can be summed up in the following equation:

D-Glucose + 2 ADP + 2 Pi + 2 NAD → 2 pyruvate 2 ATP + 2 NADH + 2H + $2H_2O$

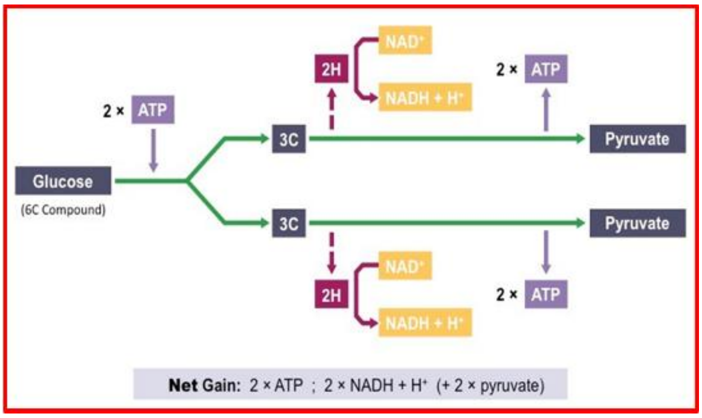


Overall pathway of Glycolysis & ATP Formation

Number of ATP generated from glycolysis

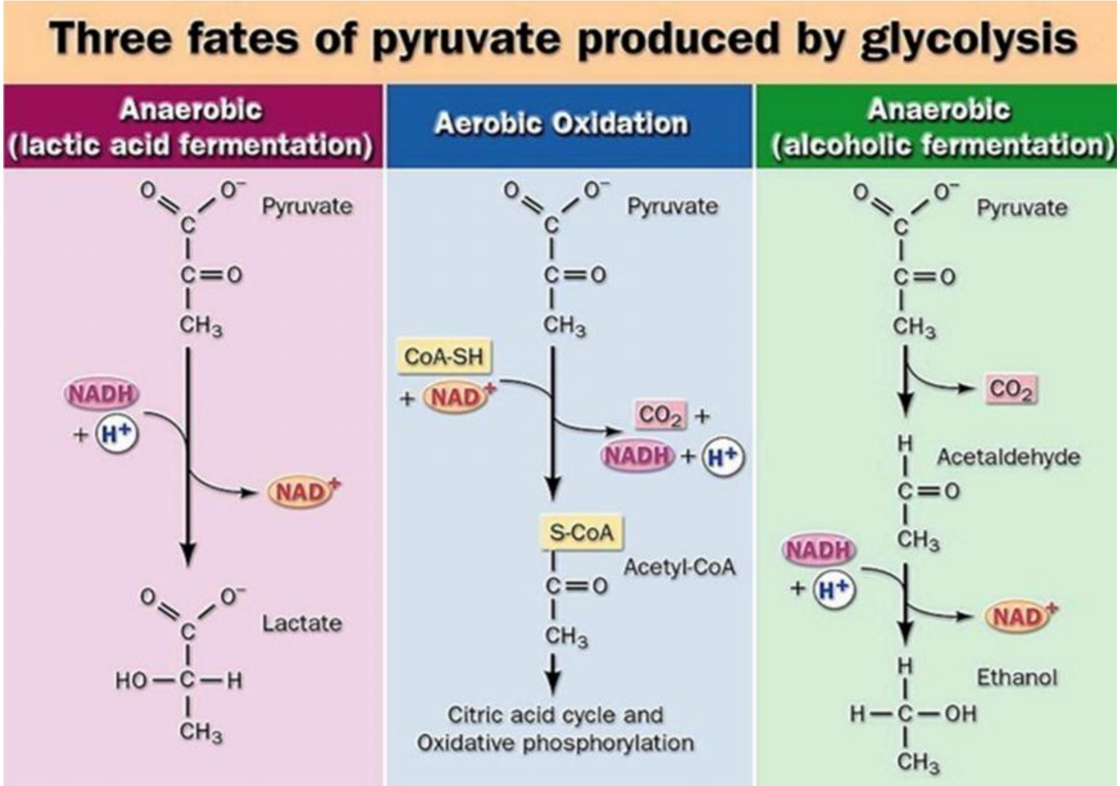
Enzyme	Aerobic	Anaerobic
Hexokinase	-1 ATP	-1 ATP
PFK-1	-1 ATP	-1 ATP
G-3-P dehydrog.	+6 ATP	----
Phospho-glycerate kinase	+2 ATP	+2 ATP
Pyruvate kinase	+2 ATP	+2 ATP
Sum	+8 ATP	+2 ATP

Overview of Glycolysis



Advantages and Disadvantages of Glycolysis

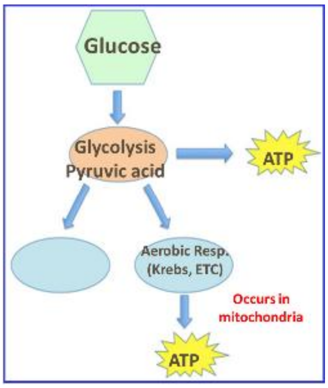
Glycolysis only produces a gain of 2 ATP per molecule of glucose, but the process is so fast that 1000's of ATP are produced in just a few milliseconds. Another advantage is that glycolysis does not require oxygen Energy can be produced for the cell even if no oxygen is present. Disadvantage: If the cell relied only on glycolysis for ATP production, the cell would quickly run out of NAD^+ to accept the hydrogen electrons. Without NAD+, the cell cannot keep glycolysis going and ATP production would stop. To keep glycolysis going, the NADH must deliver their high-energy cargo of electrons to another pathway, and then return to glycolysis to be used again



The Fate of Pyruvic Acid – What happens to it?

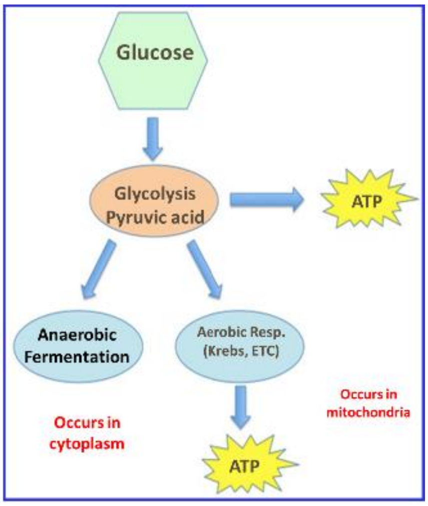
If oxygen is present:

In the presence of oxygen, the pyruvic acid will enter the mitochondria and undergo aerobic respiration. Aerobic respiration includes the stages known as the kerb cycle and the electron transport chain. Aerobic respiration will yield many more ATP than glycolysis.

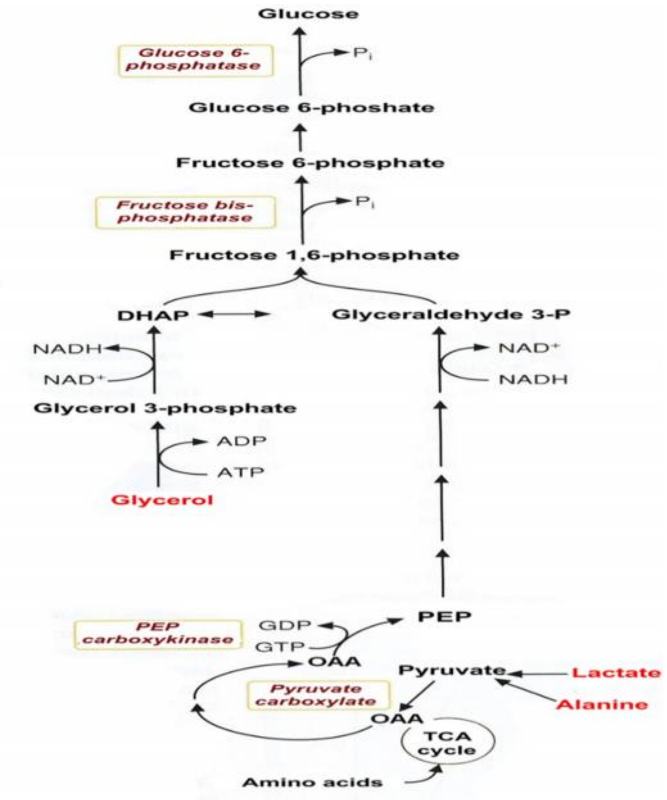


If no oxygen is present

In the absence of oxygen, the pyruvic acid will enter the anaerobic pathways of fermentation. Fermentation yields no additional ATP and Occurs in the cytoplasm



Gluconeogenesis



What are the two major stages of aerobic respiration?

- Aerobic respiration has two major stages:
- The Krebs cycle
- The electron transport chain

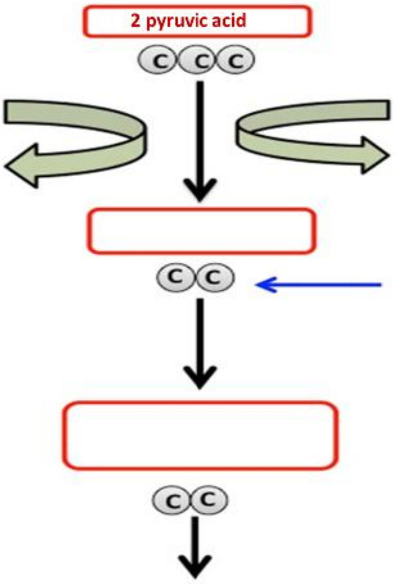
The Krebs cycle occurs in the matrix of the mitochondria and the electron transport chain occurs along the cristae membranes. At the end of glycolysis, about 90% of the chemical energy that was available in the glucose molecule is still unused. This energy is locked in: the high-energy electrons of pyruvic acid.

The Bridge Reactions:

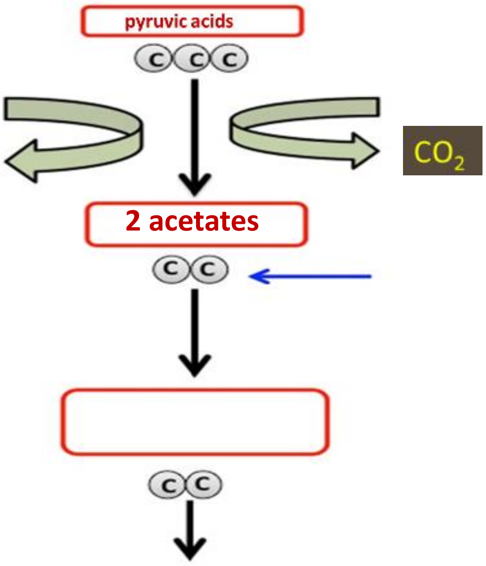
As the pyruvic acid enters the mitochondria, the following reaction occurs.

Steps in the Bridge Reaction:

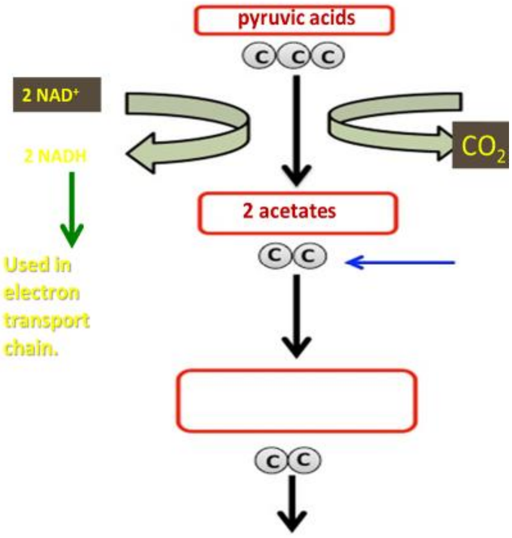
1) Pyruvic acid enters the mitochondria.



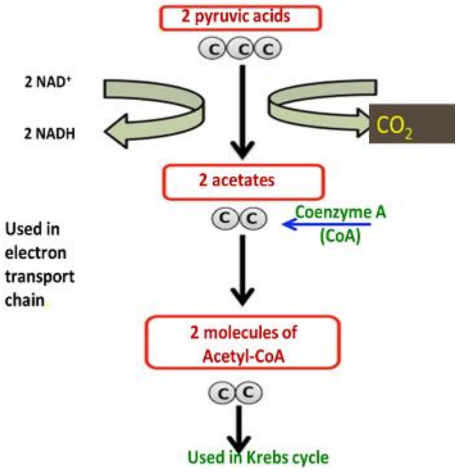
2) The 3-C Pyruvic acid is converted to 2-C acetate. This is accomplished by removing a molecule of CO_2 from each molecule of pyruvic acid. The carbon dioxide is released in air released into the air.



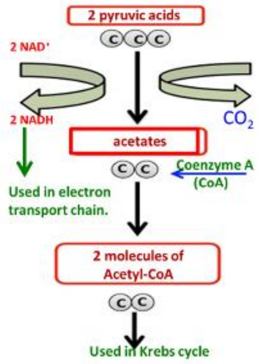
3) For each pyruvic acid that is converted to acetate, one molecule of NAD^+ is converted to NADH



4) Co enzyme attaches to acetate to form acetyl CoA . The acetyl CoA will be released into Kerb cycle



5) This reaction is often referred to as “bridge reaction”.
It is the bridge between: the cytoplasm and the
Mitochondria anaerobic and aerobic respiration
Glycolysis and the Krebs cycle.
Oxidative decarboxylation of pyruvic acid to Acetyl CoA

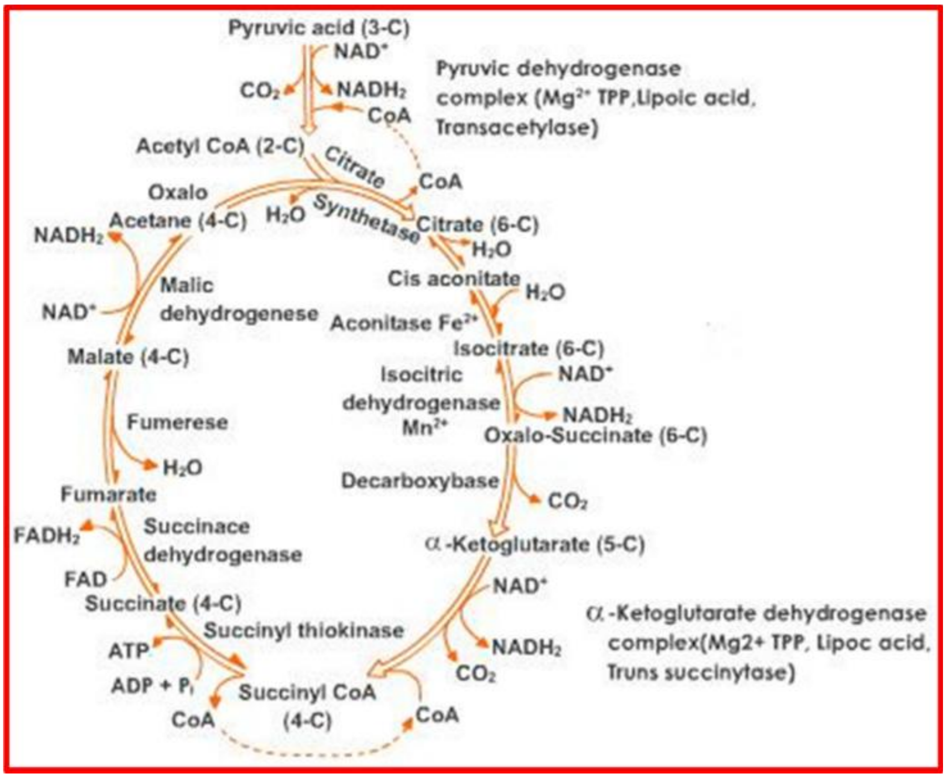


The Krebs cycle

A. The Krebs cycle is a biochemical pathway that uses the acetyl CoA molecules from the bridge reaction to produce: hydrogen atoms, ATP, carbon dioxide.

B. This set of reactions occurs in the matrix of the mitochondria.

C. Krebs cycle is so named to honour Hans Krebs. He was a German – British scientist who was largely responsible for working out the pathway in the 1930’s



a) Formation of citric acid

The acetyl- CoA combines with oxaloacetic acid to form citric acid. It contains 6 carbon atoms. This reaction is catalysed by an enzyme called citric acid synthetase.

b) Dehydration

Citric acid undergoes dehydration and forms cis-aconitic acid. This reaction is catalysed by the enzyme aconitase

c) Hydration

The aconitic acid is hydrated and it forms isocitric acid. This reaction is catalysed by the enzyme aconitase

d) Dehydrogenation I

Isocitric acid undergoes dehydrogenation in the presence of isocitric acid dehydrogenase to form Oxalo succinic acid. In this reaction 2 hydrogen atoms are released. They are accepted by NAD+ to form NADH. $NAD^+ + 2H \longrightarrow NADH + H^+$

e) Decarboxylation

The Oxalo succinic acid undergoes decarboxylation to form a-ketoglutaric acid. This reaction is catalysed by decarboxylase. In this reaction one CO2 is eliminated. Hence the a-ketoglutaric acid has only 5 carbon atoms

f) Oxidative decarboxylation

During oxidative decarboxylation a-ketoglutaric acid is converted into succinyl CoA. This reaction is catalysed by a-ketoglutaric acid dehydrogenase. Two hydrogen atoms are released and they are transferred to NAD. The NAD is converted into NADH. In the next step, the succinyl CoA is decarboxylated to succinic acid. This step is catalysed by succinic acid Thiokinase. CoA is liberated.

g) Oxidation

Succinic acid is oxidised to fumaric acid by the removal of 2 hydrogen atoms. The reaction is catalysed by succinic acid dehydrogenase. The hydrogen atoms are accepted by FAD and it forms $FADH_2$

Hydration

Fumaric acid undergoes hydration to form malic acid. This reaction is catalysed by fumarase

Dehydrogenation

It is the final step in Krebs cycle. Oxalo acetic acid is regenerated from malic acid by a process of dehydrogenation. This reaction is catalysed by malic acid dehydrogenase in the presence of NAD. The 2 hydrogen atoms removed are accepted by NAD and it forms NADH. The oxaloacetic acid formed in the above reaction condenses with the acetyl CoA to form citric acid again and thus the cycle is repeated

D. **Summary of the Krebs cycle**

NAD^+ and FAD are electron carrier. NAD^+ and FAD will deliver high energy electrons of hydrogen to electron transport chain

2. What is the total amount of CO2, ATP, NADH, and FADH2 that is produced during one turn of the Krebs cycle?

a) 2 CO_2
b) 1 ATP
c) 3 NADH
d) 1 $FADH_2$

The above totals are for one molecule of pyruvic acid.

Now remember that during glycolysis, **glucose** was broken down into two molecules of **pyruvic acid**. Therefore, one glucose molecule causes **two** turns of the **Kreb's cycle**

What is the total amount of CO_2, ATP, NADH, and $FADH_2$ that is produced per molecule of glucose in the Krebs cycle?

a) 4 CO_2

b) 2 ATP

c) 6 NADH

d) 2 $FADH_2$

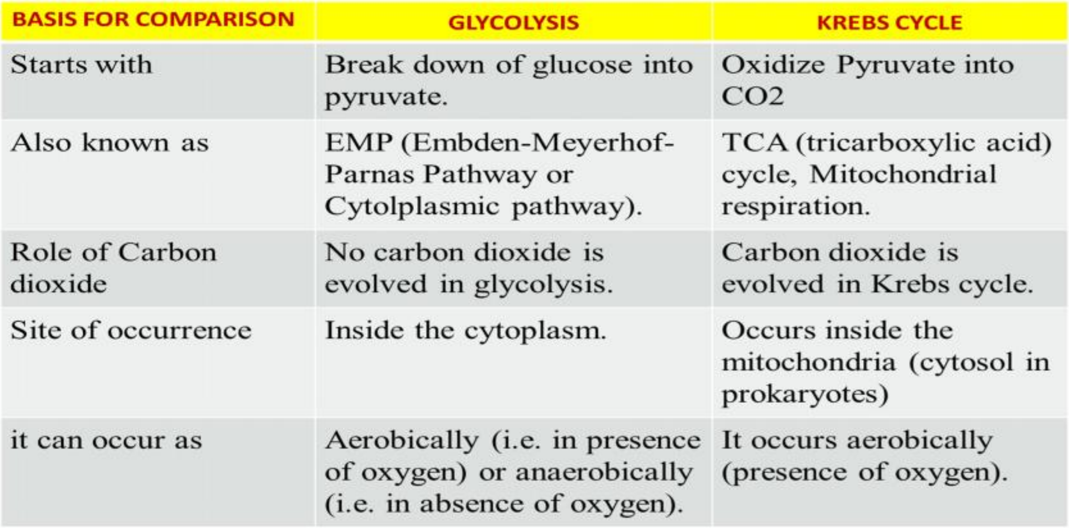
What happens to each of these products?

The carbon dioxide is released when you exhale.

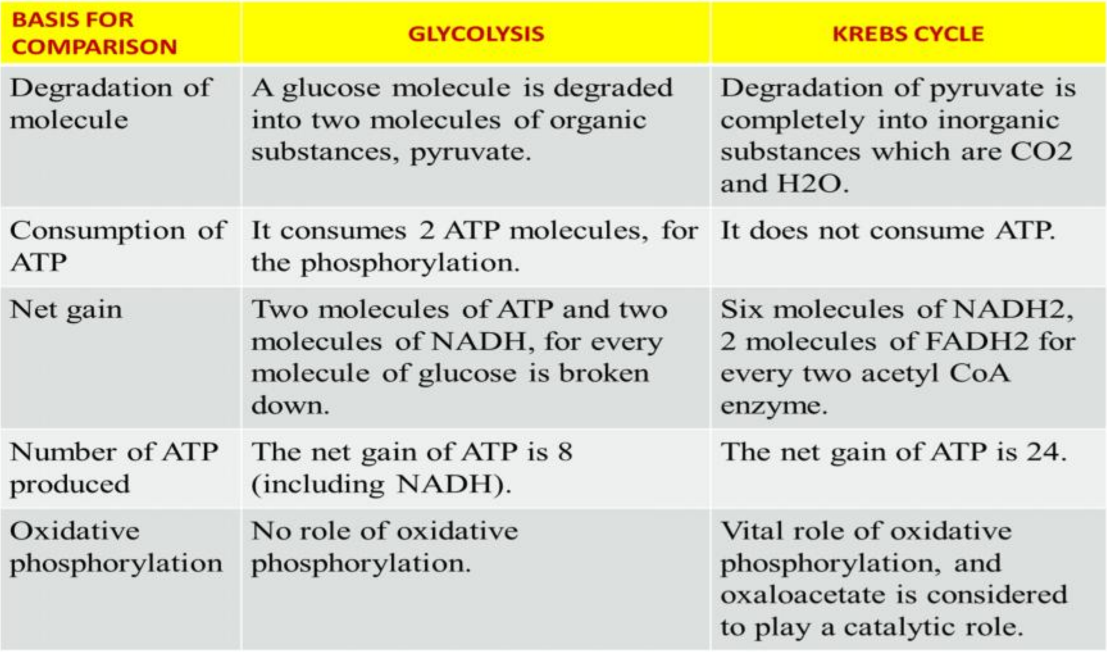
The ATP is used for cellular activities.

The NADH and the $FADH_2$ will be used in the next stage to generate huge amounts of ATP. Most of the energy contained in the original glucose molecule still has not been transferred to ATP. This transfer of energy will occur in the next step, the electron transport chain

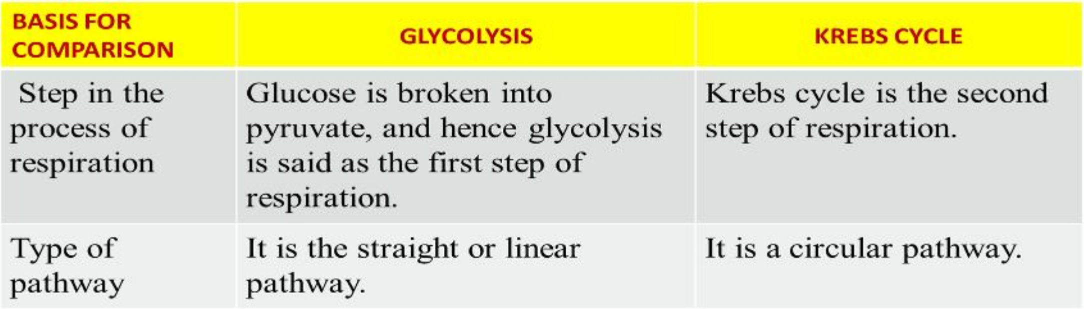
Key Difference between Glycolysis and Krebs Cycle



BASIS FOR COMPARISON	GLYCOLYSIS	KREBS CYCLE
Starts with	Break down of glucose into pyruvate.	Oxidize Pyruvate into CO2
Also known as	EMP (Embden-Meyerhof-Parnas Pathway or Cytolplasmic pathway).	TCA (tricarboxylic acid) cycle, Mitochondrial respiration.
Role of Carbon dioxide	No carbon dioxide is evolved in glycolysis.	Carbon dioxide is evolved in Krebs cycle.
Site of occurrence	Inside the cytoplasm.	Occurs inside the mitochondria (cytosol in prokaryotes)
it can occur as	Aerobically (i.e. in presence of oxygen) or anaerobically (i.e. in absence of oxygen).	It occurs aerobically (presence of oxygen).



BASIS FOR COMPARISON	GLYCOLYSIS	KREBS CYCLE
Degradation of molecule	A glucose molecule is degraded into two molecules of organic substances, pyruvate.	Degradation of pyruvate is completely into inorganic substances which are CO2 and H2O.
Consumption of ATP	It consumes 2 ATP molecules, for the phosphorylation.	It does not consume ATP.
Net gain	Two molecules of ATP and two molecules of NADH, for every molecule of glucose is broken down.	Six molecules of NADH2, 2 molecules of FADH2 for every two acetyl CoA enzyme.
Number of ATP produced	The net gain of ATP is 8 (including NADH).	The net gain of ATP is 24.
Oxidative phosphorylation	No role of oxidative phosphorylation.	Vital role of oxidative phosphorylation, and oxaloacetate is considered to play a catalytic role.



BASIS FOR COMPARISON	GLYCOLYSIS	KREBS CYCLE
Step in the process of respiration	Glucose is broken into pyruvate, and hence glycolysis is said as the first step of respiration.	Krebs cycle is the second step of respiration.
Type of pathway	It is the straight or linear pathway.	It is a circular pathway.

Electron Transport Chain

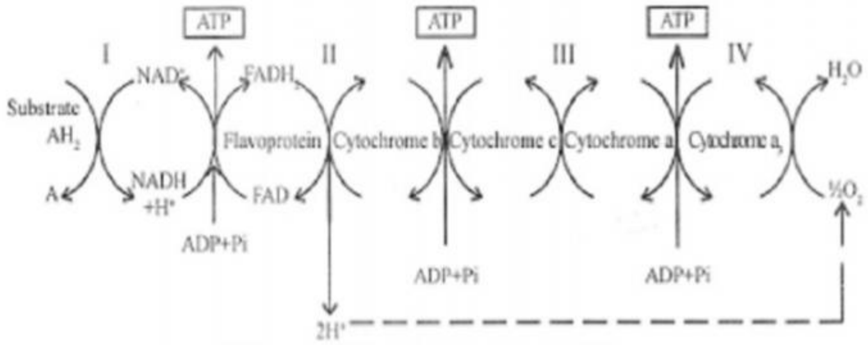
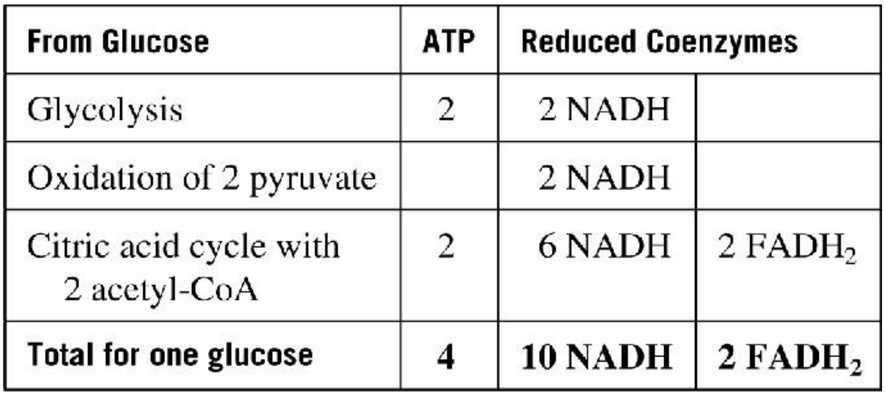


Fig. Electron transport system

The reduced coenzymes NADH and FADH2 produced from glycolysis, oxidation of pyruvate, and the citric acid cycle are oxidized to provide the energy for the synthesis of ATP.

In electron transport or the respiratory chain,

Hydrogen ions and electrons from NADH and $FADH_2$ are passed from one electron acceptor or carrier to the next until they combine with oxygen to form H_2O. The energy released during electron transport is used to synthesize ATP from ADP and Pi during oxidative phosphorylation



From Glucose	**ATP**	**Reduced Coenzymes**	
Glycolysis	2	2 NADH	
Oxidation of 2 pyruvate		2 NADH	
Citric acid cycle with 2 acetyl-CoA	2	6 NADH	2 $FADH_2$
Total for one glucose	**4**	**10 NADH**	**2 $FADH_2$**

In the electron transport system, there are five protein complexes, which are numbered I, II, III, IV, and V. two electron carriers, coenzyme Q and cytochrome c, attached to the inner membrane of the mitochondrion, carry electrons between these protein complexes bound to the inner membrane. In electron transport, the oxidation of NADH and $FADH_2$ provides hydrogen ions and electrons that eventually react with oxygen to form water.

In complex I,

Electron transport begins when hydrogen ions and electrons are transferred from NADH to complex I. loss of hydrogen from NADH regenerates NAD+ to oxidize more substrates in oxidative pathways such as the citric acid cycle. Hydrogen ions and electrons are transferred to the mobile electron carrier CoQ, forming CoQH2. CoQH2 carries electrons from complexes I and II to complex III.

Complex II consists of the enzyme succinate dehydrogenase from the citric acid cycle. In complex II, CoQ obtains hydrogen and electrons directly from $FADH_2$. This produces CoQH2 and regenerates the oxidized coenzyme FAD, which becomes available to oxidize more substrates.

$$\mathrm{FADH_2} + \mathrm{CoQ} \longrightarrow \mathrm{FAD} + \mathrm{CoQH_2}$$

Complex II consists of the enzyme succinate dehydrogenase from the citric acid cycle.

In complex II, CoQ obtains hydrogen and electrons directly from FADH2 and becomes CoQH2. two electrons are transferred from the mobile carrier CoQH2 to a series of iron-containing proteins called cytochromes. electrons are then transferred to two cytochrome c, which can move between complexes III and IV.

At complex IV, Four electrons from four cytochrome c are passed to other electron carriers. Electrons combine with hydrogen ions and oxygen (O_2) to form two molecules of water. energy is used to pump H+ from the mitochondrial matrix into the intermembrane space, further increasing the hydrogen ion gradient.

$$\boxed{4e^- + 4\mathrm{H}^+ + \mathrm{O}_2 \longrightarrow 2\mathrm{H_2O}}$$

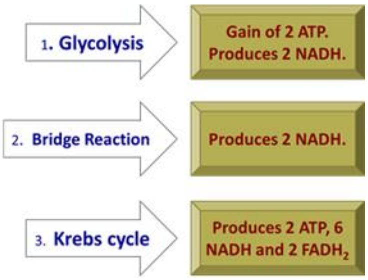
The final stage of aerobic respiration is the electron transport chain, which is located on the inner mitochondrial membrane The inner membrane is arranged into folds (cristae), which increases the surface area available for the transport chain. The electron transport chain releases the energy stored within the reduced hydrogen carriers in order to synthesise ATP. This is called oxidative phosphorylation, as the energy to synthesise ATP is derived from the oxidation of hydrogen carriers

Oxidative phosphorylation occurs over a number of distinct steps:

- Proton pumps create an electrochemical gradient (proton motive force)
- ATP synthase uses the subsequent diffusion of protons (chemiosmosis) to synthesise ATP
- Oxygen accepts electrons and protons to form water

ATP Accounting

Let’s summarize what has happened prior to the electron transport chain



. Glycolysis 2ATP
Krebs cycle 2ATP
Electron Transport Chain 34 ATP

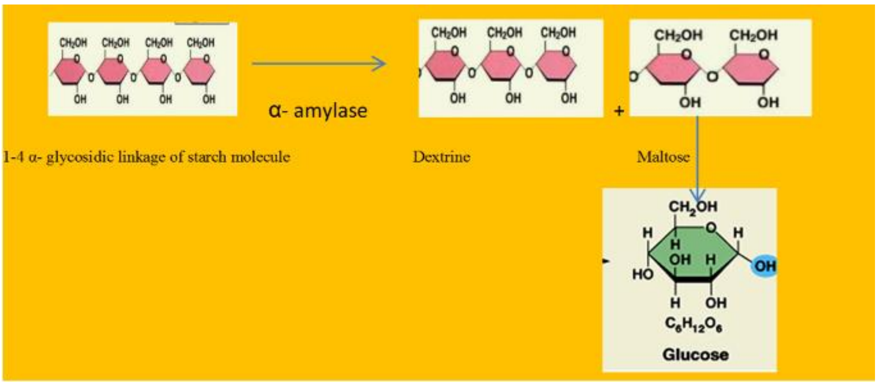
One molecule of glucose has produced 38 ATP.

Only about 40% of the energy contained in the glucose molecule has been converted to ATP. The remaining 60% is given off as heat

BREAKDOWN OF STARCH BY AMYLASES

Molecules of starch consist of links of glucose polymers formed by glycosidic bonds. The enzyme amylase breaks glycosidic bonds and turns the starch into glucose molecules. Amylase is an enzyme present in human saliva designed to break down starch present in foods like potatoes, rice and cereal grains. The main effect of amylase on starch is to break it down into simple sugars, which are used as an immediate energy source for the body. Another source of amylase production is the pancreas, which catalyzes the breakdown of dietary starch in the body for energy use.

Starch $\xrightarrow{\text{amylase}}$ dextrine+maltose $\xrightarrow{\text{amylase}}$ 2 glucose

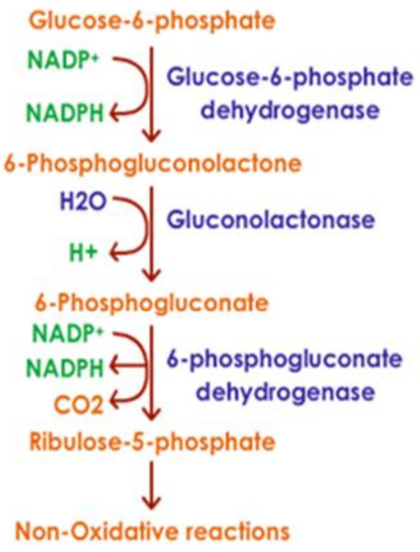


Pentose Phosphate Pathway

Pentose phosphate pathway is an alternative pathway to glycolysis and TCA cycle for oxidation of glucose.

It is a shunt of glycolysis. It is also known as hexose monophosphate (HMP) shunt or phosphogluconate pathway. It occurs in cytoplasm of both prokaryotes and eukaryotes. Pentose phosphate pathway starts with glucose and it is a multi-steps reaction. Breakdown of carbohydrates (glucose) takes place in the body by glycolysis followed by tricarboxylic acid cycle (Kreb's cycle) resulting in yield of energy in the form of ATP. Glucose can alternatively also undergo a different pathway to produce other products required by the cells. One of these alternate pathway is the pentose phosphate pathway or also called as hexose monophosphate pathway in which oxidation of glucose 6-phosphate takes place to produce pentoses. The fate of glucose whether to undergo glycolysis or the hexose monophosphate pathway is decided by the relative concentrations of NADP+ and NADPH.

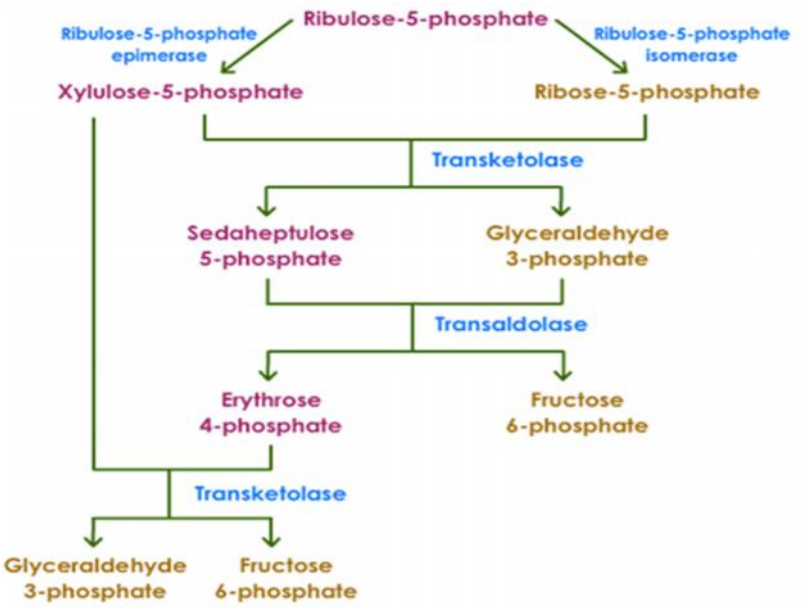
This metabolic pathway is fragmented into two phases taking place in the cytosol as all the enzymes required are present there: the oxidative and the non-oxidative phase



The pathway is also known as Phosphogluconate pathway, since the first compound formed is Phosphogluconate

Oxidative Stage of Pentose Phosphate Pathway

Glucose-6-phosphate + 2 NADP $^+$- ribulose-5-phosphate + CO2+ 2 NADPH + 2H$^+$



Non-Oxidative Stage of Pentose Phosphate Pathway

Glucose is phosphorylated to glucose-6-phosphate as in EMP pathway. Glucose-6-phosphate is oxidized to 6 phosphogluconate with NADP+ is reduced to NADPH (Glucose -6- phosphate dehydrogenase). 6-phosphogluconate is converted to ketopentose sugar- ribulose-5-P with the liberation of one molecule of CO_2 for every sugar molecule coenzyme NADP is reduced to NADPH2 (6-phosphogluconate dehydrogenase). Ribulose-5-phosphate is changed to aldopentose sugar Ribose-5-P(phosphoribose isomerase). Two molecules of ribose-5-P combine to form one molecule of Sedoheptulose and another glyceraldehyde -3- phosphate (Transketolase). The 3 carbon atom chain from Sedoheptulose is linked with 3 carbon atom of 3-phosphoglyceraldehyde with the formation of hexose phosphate- fructose-6- P (Transaldolase). A second molecule of fructose-6-P is formed by linking 4 carbon atom

(Erythrose-4-P) from Sedoheptulose with 2 carbon fragment pentose molecules (Xylulose-5-P) along with the formation of 3- phosphoglyceraldehyde (Transketolase) Two molecules of 3- phosphoglyceraldehyde like to form fructose 1-6 diphosphate which after dephosphorylation form fructose -6-P

Thus with the formation of fructose-6-P, the cycle is complete in which 6 molecules of glucose enter, but only one of them is completely oxidized liberating 6 molecules of CO_2 and the rest are five reformed with 12 molecules of $NADPH_2$

6 Glucose-6-P + 12 NADP+ + $7H_2O$ $\longrightarrow$ 6-Fructose-6-P + $6CO_2$ + $12NADPH_2$ + H_3PO_4

$NADPH_2$ undergo oxidation by atmospheric O_2 via electron transport system where 3 molecules of ATP are produced per $NADPH_2$ oxidized. Thus the oxidation of 12 $NADPH_2$ molecules will produce 12 x 3 = 36 ATP molecules. Therefore this pathway is as efficient as EMP Kreb's cycle.

Significance

It provides alternative route for carbohydrate breakdown and provision of energy

It provides ribose sugar for synthesis of nucleic acid

It plays important role in fixation of CO2 in photosynthesis through ribulose -5-P

AMINO ACID METABOLISM

The catabolic pathways for the 20 amino acids vary considerably, but all amino acids are degraded to one of seven metabolites:

- pyruvate,
- α ketoglutarate,
- succinyl-CoA,
- fumarate,
- oxaloacetate,
- acetyl CoA, or
- Acetoacetate.
- What is the fate of each of these metabolites?

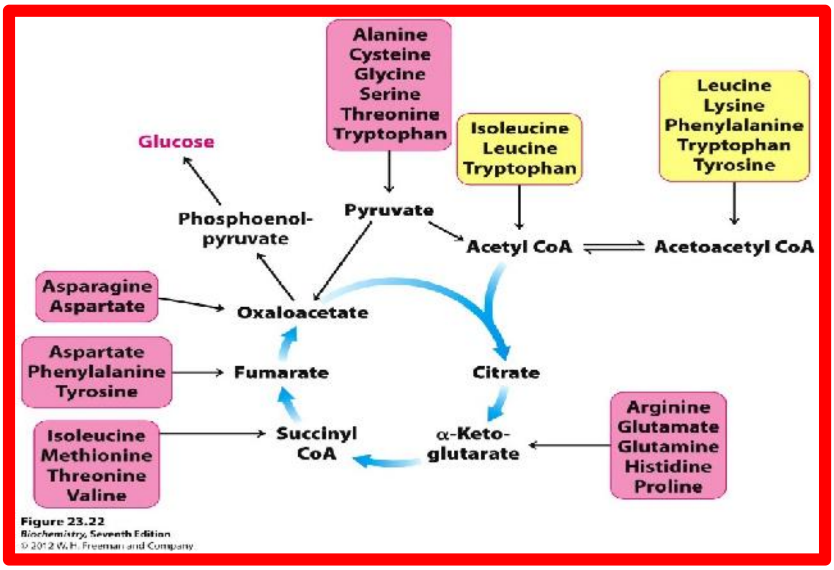


Figure 23.22
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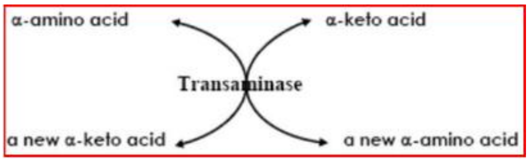
Catabolic Pathway of Amino Acids

- Transamination
- Deamination
- Transamidination
- Transamidation
- Decarboxylation

1. Transamination

Transamination means transfer of amino group from α-amino acid to α-keto acid with formation of a new α-amino acid and a new –α keto acid. All amino acids can be transaminated except lysine, threonine, proline and hydroxy proline. All transamination reactions are reversible. It is catalysed by aminotransferases (transaminases). It needs pyridoxal phosphate as a coenzyme



Examples of transaminases

A. Alanine transaminase B. Aspartate transaminase C. Glutamate transaminase

Deamination

Deamination means the removal of amino group from α-amino acid in the form of ammonia with formation of α-keto acid

Deamination may be oxidative or non-oxidative

A. **Oxidative deamination**

It is catalysed by one of the following enzymes:

1. L-1. Amino acid oxidases
2. D-amino acid oxidases
3. Glutamate dehydrogenase

B. **Non-oxidative deamination**

It is catalysed by one of the following enzymes:

1. Dehydratases
2. Desulfhydrases

Most of the naturally occurring α-amino acids are catabolized by transamination with α-ketoglutaric acid followed by deamination of the produced glutamic acid, a condition called trans deamination

Transamidination

Transamidination means the transfer of amidine group from a donor molecule to an acceptor molecule It is catalyzed by transamidinase enzyme

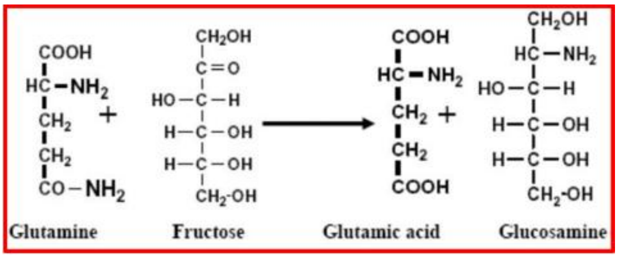
An example of transmidination reaction is the transfer of amidine group from arginine (donor) to glycine (acceptor) in creatine biosynthesis

Transamidation

Transamidation means transfer of amide group nitrogen from a donor molecule to an acceptor molecule. It is catalyzed by transamidase enzyme. Examples of transmidation reaction include:

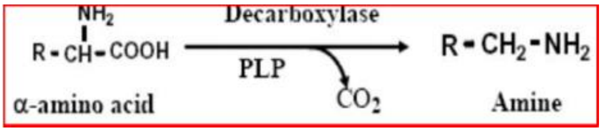
Transfer of amide nitrogen from glutamine (donor) to fructose (acceptor) 1.to form glucosamine

Glucosamine biosynthesis



Decarboxylation

Decarboxylation means removal of CO_2 from amino acid with formation of corresponding amines. It is catalyzed by decarboxylase enzyme. It needs pyridoxal phosphate as a coenzyme

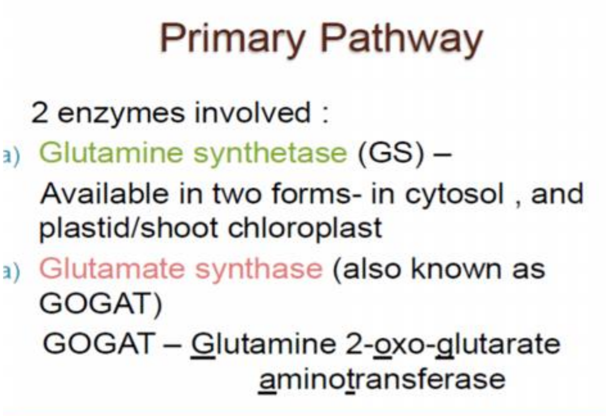


Ammonia assimilating enzymes

Ammonia Assimilation: Conversion of ammonia generated from nitrate assimilation or photorespiration into amino acid.

2 pathways –

i. Primary Pathway ii. Alternative Pathway

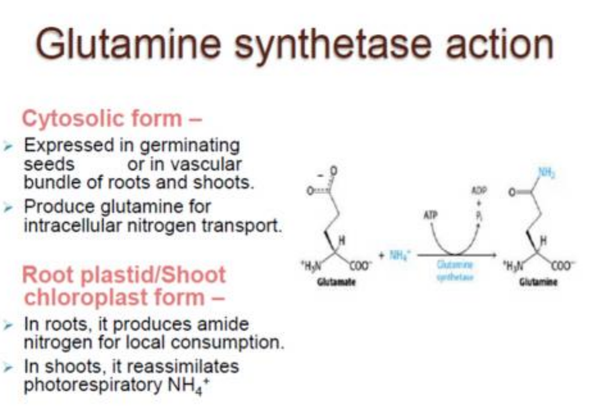


Primary Pathway

2 enzymes involved :

a) Glutamine synthetase (GS) –
Available in two forms- in cytosol , and plastid/shoot chloroplast

a) Glutamate synthase (also known as GOGAT)
GOGAT – Glutamine 2-oxo-glutarate aminotransferase



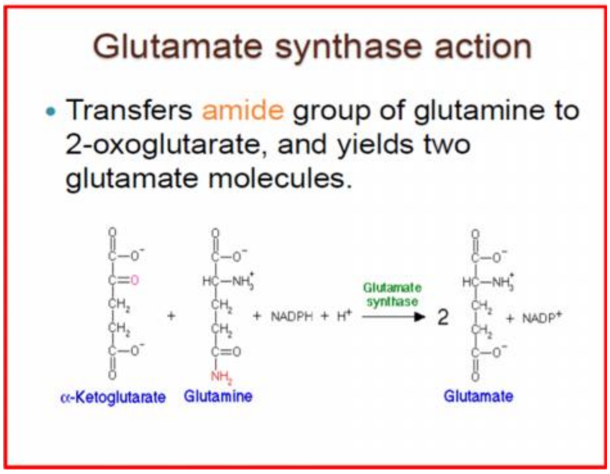
Glutamine synthetase action

Cytosolic form –

- Expressed in germinating seeds or in vascular bundle of roots and shoots.
- Produce glutamine for intracellular nitrogen transport.

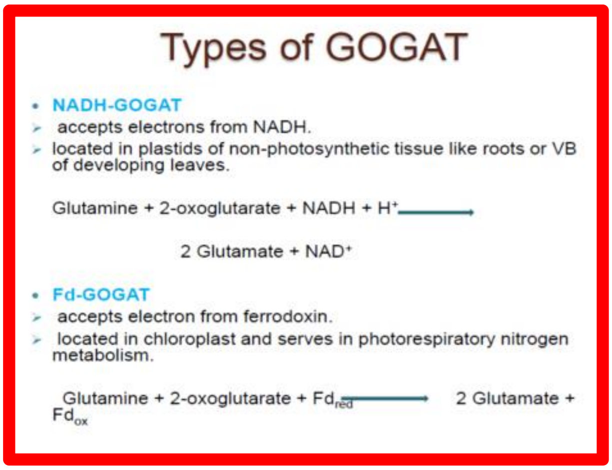
Root plastid/Shoot chloroplast form –

- In roots, it produces amide nitrogen for local consumption.
- In shoots, it reassimilates photorespiratory NH_4^+



Glutamate synthase action

- Transfers amide group of glutamine to 2-oxoglutarate, and yields two glutamate molecules.



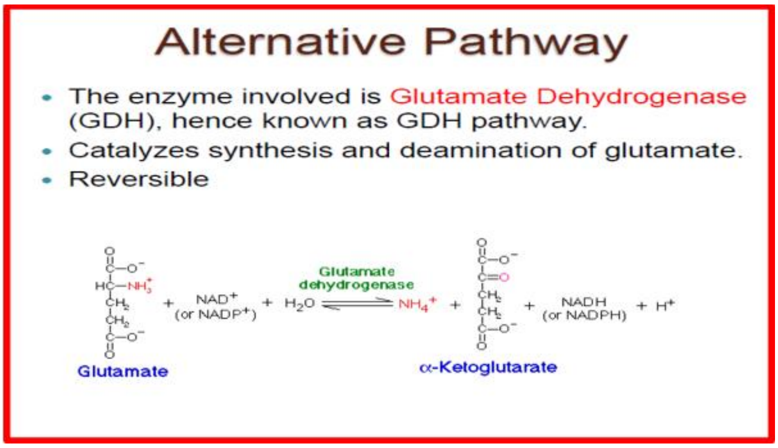
Types of GOGAT

- NADH-GOGAT
  - accepts electrons from NADH.
  - located in plastids of non-photosynthetic tissue like roots or VB of developing leaves.

Glutamine + 2-oxoglutarate + NADH + $H^+ \longrightarrow$ 2 Glutamate + NAD^+

- Fd-GOGAT
  - accepts electron from ferrodoxin.
  - located in chloroplast and serves in photorespiratory nitrogen metabolism.

Glutamine + 2-oxoglutarate + $Fd_{red} \longrightarrow$ 2 Glutamate + Fd_{ox}



Alternative Pathway

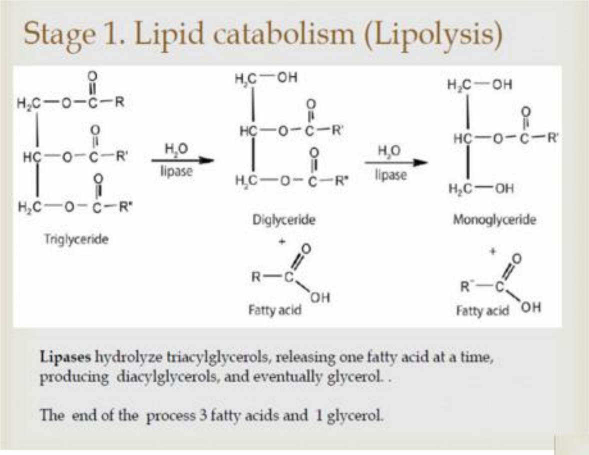
- The enzyme involved is Glutamate Dehydrogenase (GDH), hence known as GDH pathway.
- Catalyzes synthesis and deamination of glutamate.
- Reversible

LIPID METABOLISM

Lipid metabolism is the synthesis and degradation of lipids in cells. Lipid metabolism is the break down or storage of fats for energy; these fats are obtained from consuming food and absorbing them or they are synthesized by an animal's liver.

Phospholipase: An enzyme that catalyzes the splitting of a phospholipid molecule by the addition of water. Also called a lecithinase.

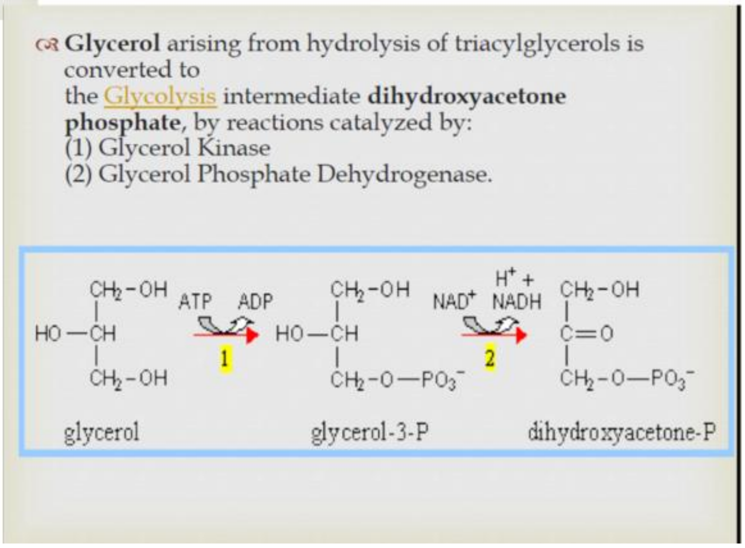
There are a number of phospholipases. They are grouped on the basis of the specific chemical bond they split. The phospholipases include phospholipase A1 (PLA1), phospholipase A2 (PLA2), phospholipase C, and phospholipase D.



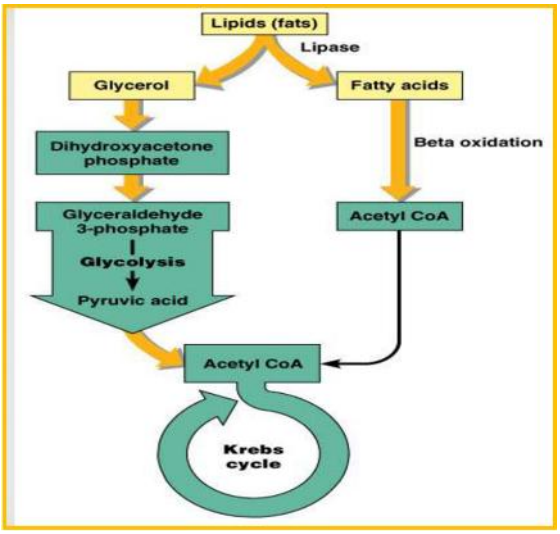
Stage 1. Lipid catabolism (Lipolysis)

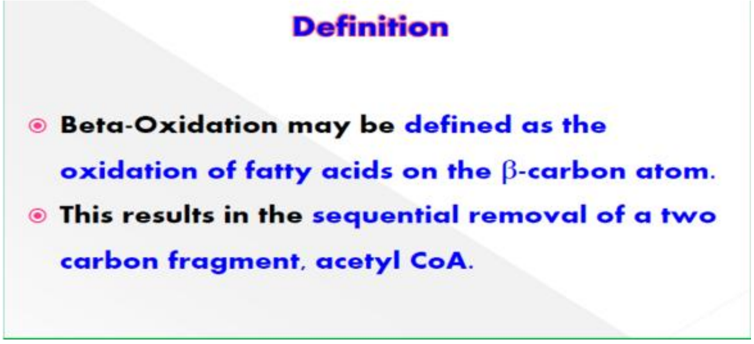
Lipases hydrolyze triacylglycerols, releasing one fatty acid at a time, producing diacylglycerols, and eventually glycerol. .

The end of the process 3 fatty acids and 1 glycerol.



- **Glycerol** arising from hydrolysis of triacylglycerols is converted to the Glycolysis intermediate **dihydroxyacetone phosphate**, by reactions catalyzed by:
  (1) Glycerol Kinase
  (2) Glycerol Phosphate Dehydrogenase.





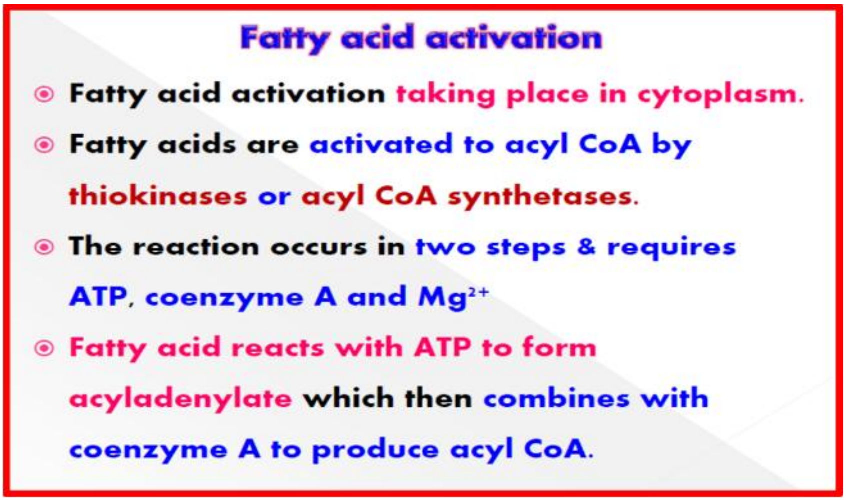
Definition

- **Beta-Oxidation may be defined as the oxidation of fatty acids on the β-carbon atom.**
- **This results in the sequential removal of a two carbon fragment, acetyl CoA.**

β oxidation is the chief process by fatty acid degradation in plants. This is one of the methods of oxidation of fatty acid. The β oxidation mechanism is well established for saturated fatty acid. The β oxidation takes place in mitochondria and involves sequential removal of 2 C in the form of acetyl CoA from the carboxyl end of the fatty acid. This is called as β oxidation because β –C of the fatty acid is oxidized during the process.

Stages of fatty acid oxidation

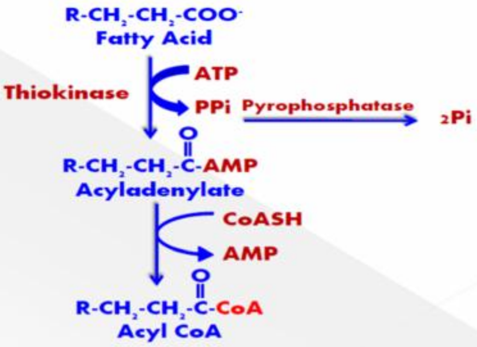
1. Activation of fatty acids takes place on the outer mitochondrial membrane
2. Transport into the mitochondria
3. Degradation to two-carbon fragments (as acetyl CoA) in the mitochondrial matrix (b-oxidation pathway



Fatty acid activation

- **Fatty acid activation taking place in cytoplasm.**
- **Fatty acids are activated to acyl CoA by thiokinases or acyl CoA synthetases.**
- **The reaction occurs in two steps & requires ATP, coenzyme A and Mg^{2+}**
- **Fatty acid reacts with ATP to form acyladenylate which then combines with coenzyme A to produce acyl CoA.**

Activation of fatty acid to Acyl CoA



$R\text{-}CH_2\text{-}CH_2\text{-}COO^-$
Fatty Acid
Thiokinase
ATP
PPi
Pyrophosphatase
2Pi
$R\text{-}CH_2\text{-}CH_2\text{-}\overset{O}{\overset{\|}{C}}\text{-}AMP$
Acyladenylate
CoASH
AMP
$R\text{-}CH_2\text{-}CH_2\text{-}\overset{O}{\overset{\|}{C}}\text{-}CoA$
Acyl CoA

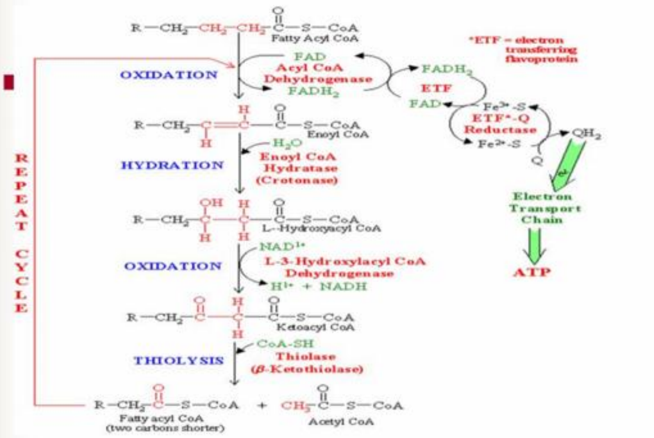
Transport of Acyl CoA into Mitochondria

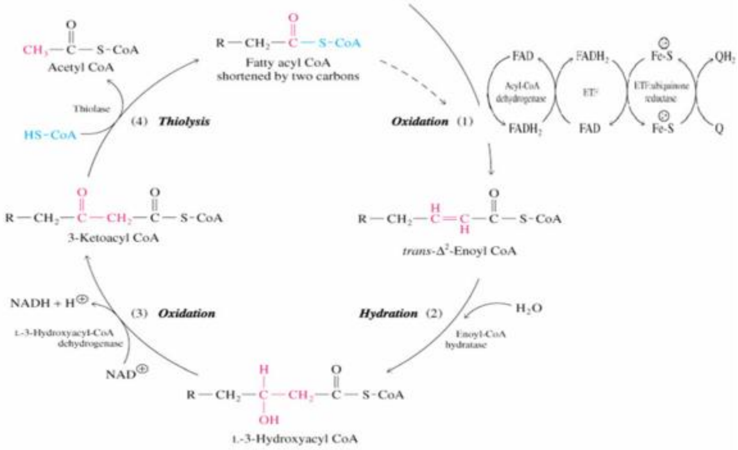
- The inner mitochondrial membrane is impermeable to fatty acids.
- A specialized carnitine carrier system (carnitine shuttle) operates to transport activated fatty acids from cytosol to the mitochondria.

β-Oxidation Proper

- Each cycle of β –oxidation, liberating a two carbon unit-acetyl CoA, occurs in a sequence of four reactions
  1. Oxidation
  2. Hydration
  3. Oxidation
  4. Cleavage

Beta-Oxidation of Fatty Acids



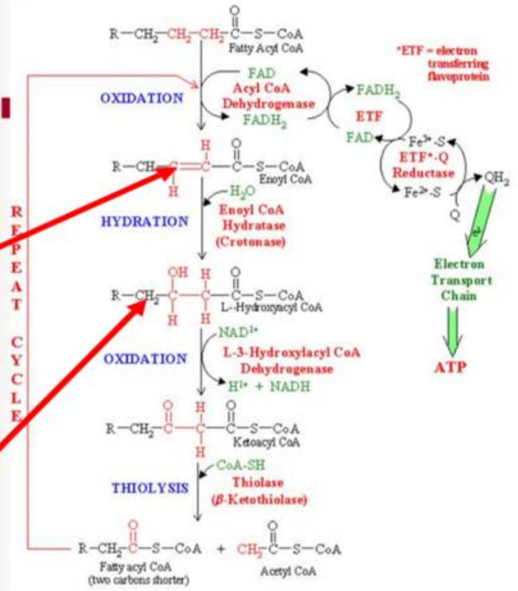


1.Oxidation

- Acyl CoA undergoes dehydrogenation by an FAD-dependent flavoenzyme, acyl CoA dehydrogenase.
- A double bond is formed between α & β carbons (i.e., 2 & 3 carbons) **Enoyl CoA**

2. Hydration:

- Enoyl CoA hydratase brings about the hydration of the double bond to form β -hydroxyacyl CoA.

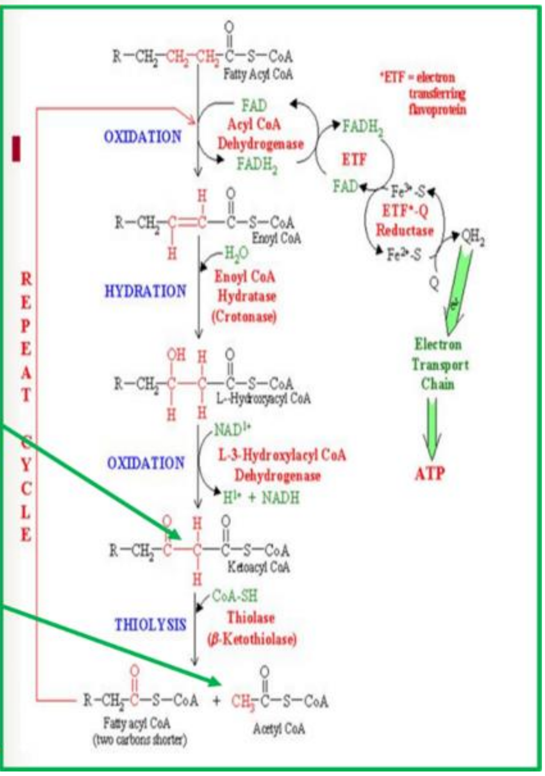


3. Oxidation

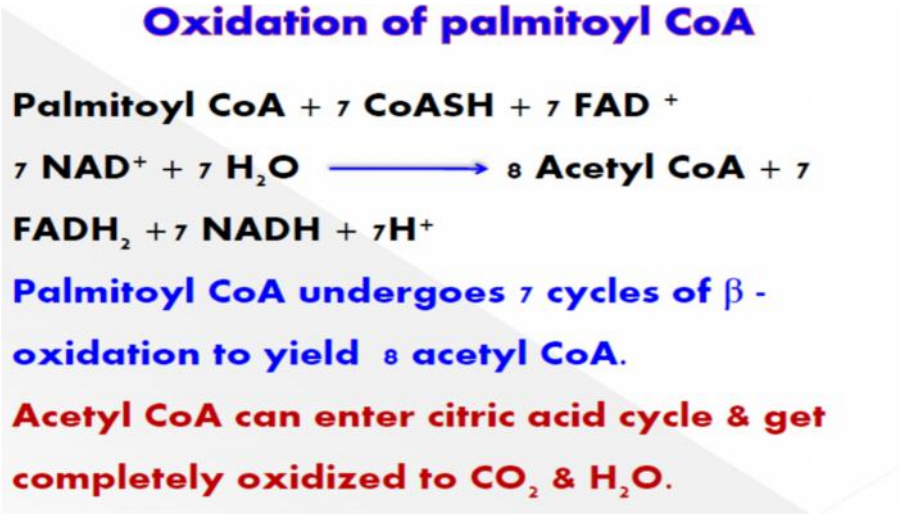
- β-Hydroxyacyl CoA dehydrogenase catalyses the second oxidation & generates NADH.
- The product formed is β-ketoacyl CoA.

4. Cleavage

- The final reaction in β -oxidation is the liberation of a 2 carbon fragment, acetyl CoA from acyl CoA.
- This occurs by a thiolytic cleavage catalysed by β-ketoacyl CoA thiolase (or thiolase).



- The new acyl CoA, containing two carbons less than the original, reenters the β-oxidation cycle.
- The process continues till the fatty acid is completely oxidized.

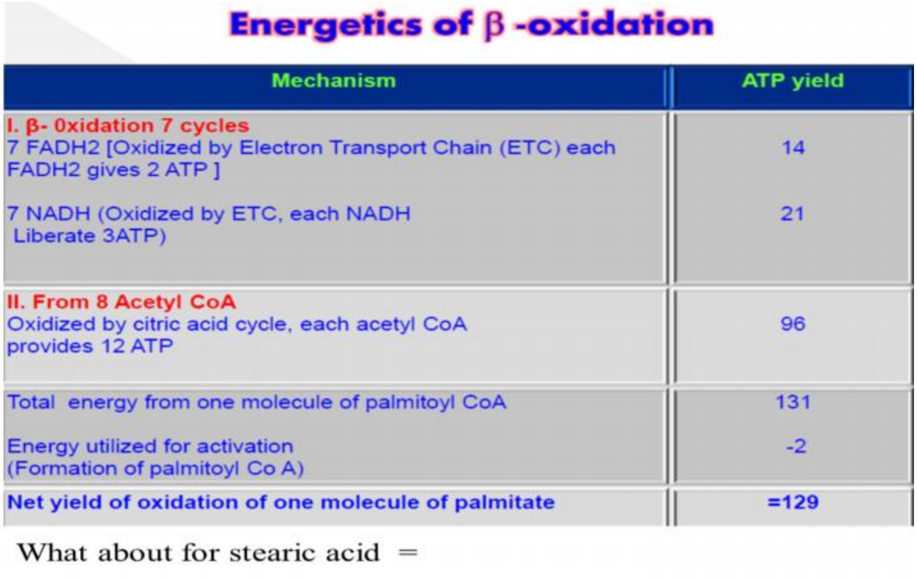


Oxidation of palmitoyl CoA

$$\text{Palmitoyl CoA} + 7\ \text{CoASH} + 7\ \text{FAD}^+ \; 7\ \text{NAD}^+ + 7\ H_2O \longrightarrow 8\ \text{Acetyl CoA} + 7\ \text{FADH}_2 + 7\ \text{NADH} + 7H^+$$

Palmitoyl CoA undergoes 7 cycles of β - oxidation to yield 8 acetyl CoA.

Acetyl CoA can enter citric acid cycle & get completely oxidized to CO_2 & H_2O.



Energetics of β -oxidation

Mechanism	ATP yield
I. β- 0xidation 7 cycles 7 FADH2 [Oxidized by Electron Transport Chain (ETC) each FADH2 gives 2 ATP]	14
7 NADH (Oxidized by ETC, each NADH Liberate 3ATP)	21
II. From 8 Acetyl CoA Oxidized by citric acid cycle, each acetyl CoA provides 12 ATP	96
Total energy from one molecule of palmitoyl CoA	131
Energy utilized for activation (Formation of palmitoyl Co A)	-2
Net yield of oxidation of one molecule of palmitate	=129

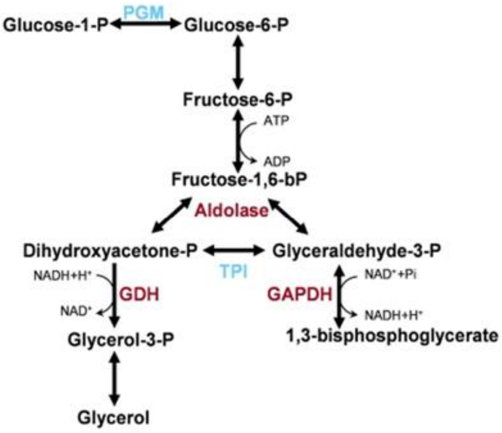
What about for stearic acid =

BIOSYNTHESIS OF FAT

Synthesis of glycerol

Glycerol portion of the fat is synthesized from Dihydroxy acetone phosphate which is produced from fructose 1-6 diphosphate in the presence of Aldolase and cofactor Zn2+ and Cu2+, as an intermediate product in glycolysis. It is done in 2 steps

- Dihydroxy acetone phosphate is reduced to ά - glycerophosphate by enzyme ά- glycerophosphate dehydrogenase and coenzyme NADH2
- ά – Glycerophosphate is then hydrolyzed by phosphatase to liberate phosphoric acid and glycerol

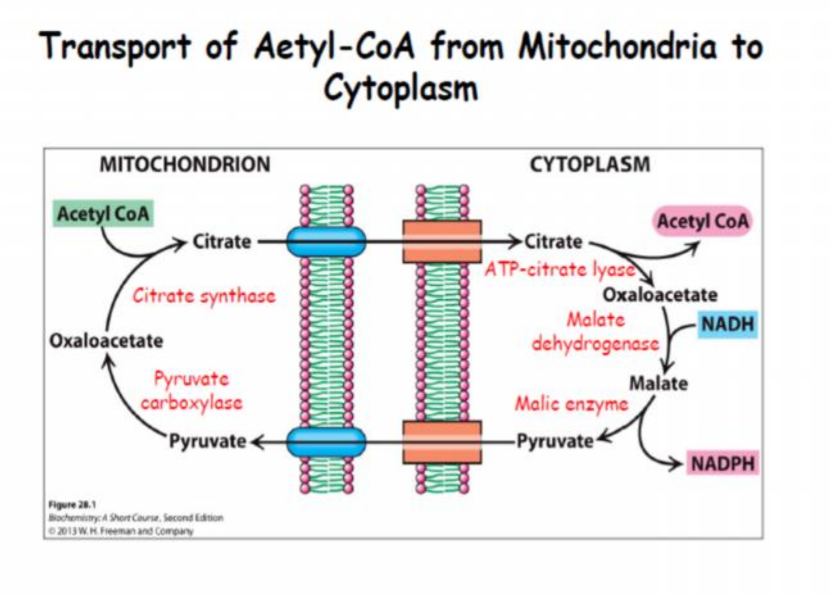


Ii. Fatty acid synthesis

Fatty acid synthesis is the creation of fatty acids from acetyl-CoA and NADPH through the action of enzymes called fatty acid synthases. This process takes place in the cytoplasm of the cell. Most of the acetyl-CoA which is converted into fatty acids is derived from carbohydrates via the glycolytic pathway. Fatty acid synthesis is somewhat of a complicated cytoplasmic pathway

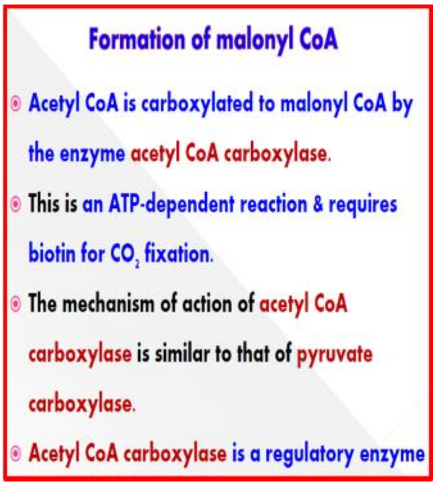
Fatty Acid Synthesis

Acetyl CoA is produced in the mitochondria by the oxidation of pyruvate, fatty acids, degradation of carbon skeleton of certain amino acids & from ketone bodies. Mitochondria are not permeable to acetyl CoA Acetyl CoA condenses with oxaloacetate in mitochondria to form citrate. Citrate is freely transported to cytosol by tricarboxylic acid transporter. In cytosol it is cleaved by ATP citrate lyase to liberate acetyl CoA & oxaloacetate. Oxaloacetate in the cytosol is converted to malate. Malic enzyme converts malate to pyruvate. NADPH & CO_2 are generated in this reaction. Both of them are utilized for fatty acid synthesis



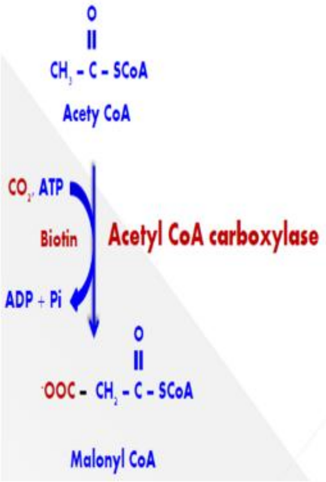
Transport of Aetyl-CoA from Mitochondria to Cytoplasm

Acetyl CoA, the starting point with two keto acid carbons, is first converted to malonyl CoA with three keto acid carbons



Formation of malonyl CoA

- Acetyl CoA is carboxylated to malonyl CoA by the enzyme acetyl CoA carboxylase.
- This is an ATP-dependent reaction & requires biotin for CO_2 fixation.
- The mechanism of action of acetyl CoA carboxylase is similar to that of pyruvate carboxylase.
- Acetyl CoA carboxylase is a regulatory enzyme



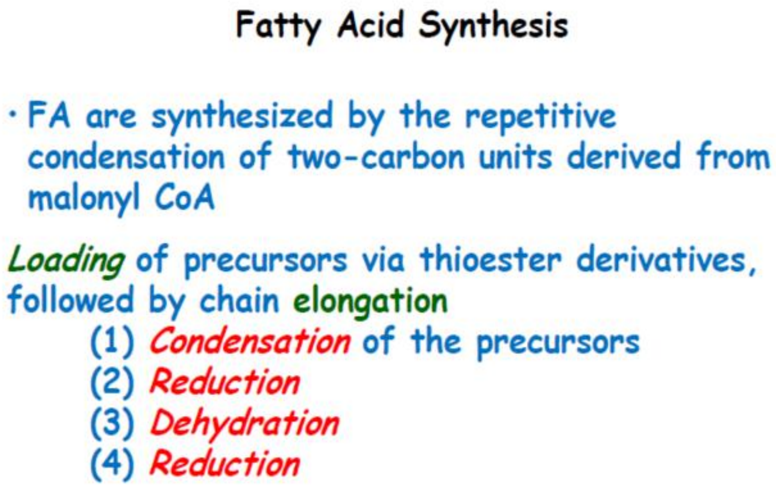
The elongation phase of fatty acid synthesis starts with the formation of acetyl ACP and malonyl ACP. Acetyl transacylase and malonyl transacylase catalyze these reactions.

$$\boxed{\begin{array}{l}\text{Acetyl CoA} + \text{ACP} \rightleftharpoons \text{acetyl ACP} + \text{CoA}\\ \text{Malonyl CoA} + \text{ACP} \rightleftharpoons \text{malonyl ACP} + \text{CoA}\end{array}}$$

Acetyl ACP and malonyl ACP react to form acetoacetyl ACP. The acyl-malonyl ACP condensing enzyme catalyzes this condensation reaction.

$$\boxed{\text{Acetyl ACP} + \text{malonyl ACP} \longrightarrow \text{acetoacetyl ACP} + \text{ACP} + \text{CO}_2}$$

In the condensation reaction, a four-carbon unit is formed from a two carbon unit and a three-carbon unit, and CO_2 is released.



Fatty Acid Synthesis

- FA are synthesized by the repetitive condensation of two-carbon units derived from malonyl CoA

Loading of precursors via thioester derivatives, followed by chain *elongation*

(1) *Condensation* of the precursors

(2) *Reduction*

(3) *Dehydration*

(4) *Reduction*

Malonyl CoA reacts with another molecule of acetyl CoA in the presence of fatty acid synthetase and coenzyme $NADPH_2$ to form coenzyme-A derivative of butyric acid (4 C atoms)

$$\textbf{Malonyl CoA} + \textbf{Acetyl CoA} + \textbf{2NADPH}_2 \qquad \textbf{Butryl CoA} + \textbf{CoA}+ \textbf{CO}_2+ \textbf{H}_2\textbf{O}+ \textbf{2NADP}$$

Butryl CoA in the next step will combine with malonyl CoA to form CoA derivative of fatty acid containing 6 C atoms. This process will be repeated till coenzyme A derivative of long chain fatty acid (which may contain up to 16-18 C atoms-palmitic acid) is produced. For example This process is then repeated six times, each time a malonyl CoA molecule is added, each time CO_2 is released, and each time the product is processed into a 2-carbon longer fatty acid inked to ACP After six repeated cycles, palmitoyl (16-carbon fatty acid)-ACP, a typical fatty acid is generated . This is treated with a thioesterase to remove ACP and the fatty acid palmitic acid is released. The enzyme involved is fatty acid synthetase is not simple but a complex of many enzymes and an acyl carrier protein called ACP is used for catalyze the reaction. The synthesis of fatty acid involves 3 categories namely initiation reaction, chain elongation reaction and termination reaction

Condensation of fatty acids and glycerol

The fats or triglycerides are synthesized not from glycerol and free fatty acids but from ά-glycerophosphate and CoA derivatives of fatty acid

The glycerol is phosphorylated to form L-ά – glycerophosphate ά – glycerophosphate undergo condensation with 2 molecules of acyl CoA to form ά- phosphatidic acid (Acyltransferase). ά-phosphatidic acid undergo dephosphorylation in the presence of phosphatase to form ά, β-diglyceride

Condensation of one molecule of an acyl –CoA with free hydroxyl group of the ά, β-diglyceride takes place in the presence of enzyme diglyceride acyl transferase to form triglyceride

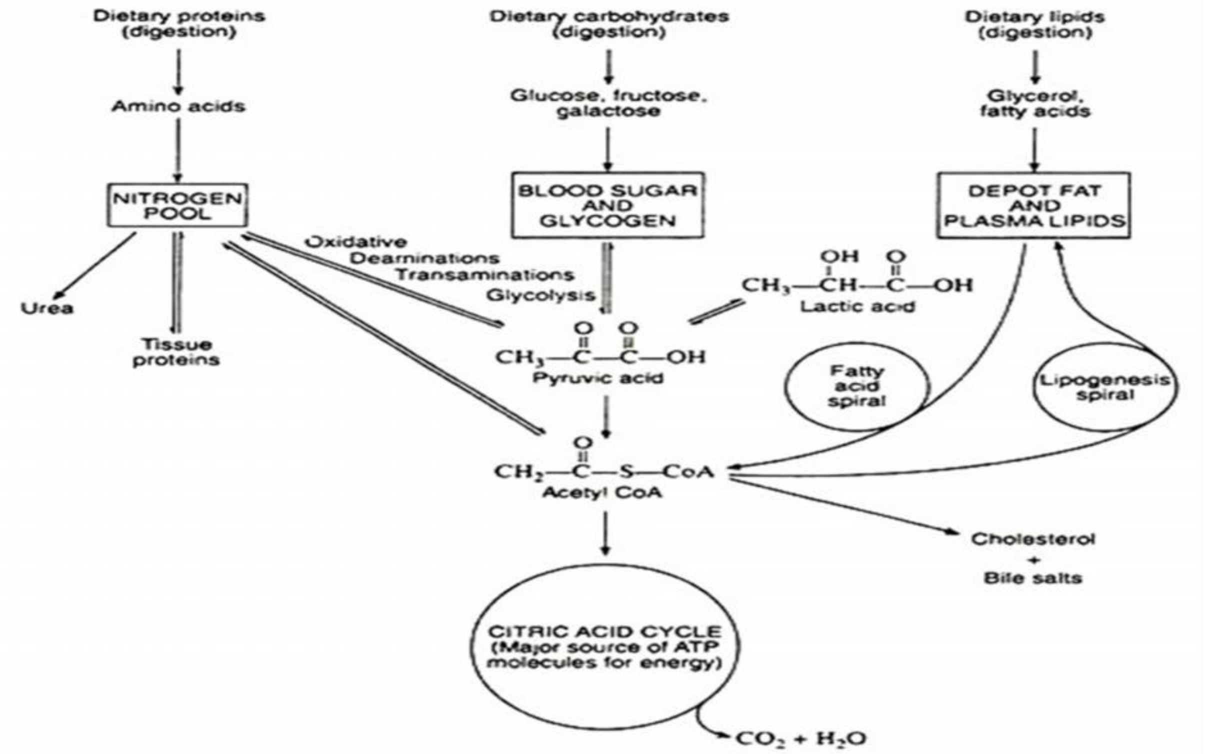


Fig. 8.91 : Interrelationships of metabolic pathways

FILL UP THE BLANKS

1) The citric acid cycle reaction takes place in ____ cell
2) Adenosine triphosphate has two _______ phosphate
3) Number of ATP molecules produced when one molecule of glucose gets oxidized to CO2 and water are______
4) The initiating codon in protein synthesis is _______
5) The type of RNA which gets involved in the transfer of amino acid in protein synthesis is ___
6) Carotenoids give maximum absorption at _______ nm
7) NADPH is produced is ___ pathway
8) Complete oxidation of a glucose molecule yields ______ ATP molecules in nett
9) The other names of citric acid cycle are _______ and _______
10) In double helical DNA, the number of adenine is equal to ______ which are connected by _______ bonds
11) In non- cyclic electron transport, molecular O2 is released by photo oxidation of _____

12) Cyclic photophosphorylation takes place only in ______

13) Non- Cyclic photophosphorylation takes place both in ______ and _______

14) The first stable product of dark reaction of photosynthesis is _______

15) C4 plants belongs to ______ family

16) CO2 fixation in C3 plants follow _____ pathway

17) CO2 fixation in C4 plants follow _____ pathway

18) Number of ATP molecules produced in every cycle of β –oxidation of fatty acid is ______

19) Number of ATP molecules produced for complete oxidation of acetyl CoA into H2O in kreb cycle is ______

20) Complete oxidation of a glucose molecule results in the gain of _____ ATP molecules

21) The two phases in which photosynthesis can takes place is _____ and ________

22) The primary CO_2 acceptor in Calvin cycle is _______

23) The first stable compound in C4 cycle is ________

24) One molecule of glucose after oxidation liberates _____

25) The process of conversion of glucose to pyruvic acid is known as ______

26) Aerobic oxidation of pyruvic acid into CO_2 and H_2O takes place through ______ cycle

27) The product formed from pyruvic acid under anaerobic condition is ______

28) The glycolysis pathway of oxidation of glucose to pyruvic acid was studied by _______ scientists

29) The process of glycolysis takes place in _____ and does not require ______ at any stage

30) All the reactions of Kreb’s cycle takes place inside the_____

31) The common pathway for the metabolism of carbohydrates, fats and proteins is ______

32) ____ act as a connecting link between glycolysis and Kreb’s cycle

33) Synthesis of ATP during oxidation of coenzymes in electron transport system of aerobic respiration is called ________

34) Biosynthesis of protein in the plant is done under the direction of ____

35) The process of formation of mRNA from DNA is known as ________

36) The process of formation of protein from the language available in the form of mRNA is known as ______

37) The terminating codon to stop the chain formation is ______-

38) The main pathway of saturated fatty acid synthesis in plants, animals is through ______

39) Biosynthesis of triglycerides is through condensation of _______ and _______

40) Oxidation of fatty acid takes place through __ and _______

41) The end product in the β- oxidation of fatty acid is ______

42) Number of ATP molecules produced during β- oxidation of fatty acid depends on _______

43) One molecule of ATP is equal to _____ calories

44) Biosynthesis of fatty acid in plant, animal or bacteria takes place through ______ pathway

45) Number of ATP molecules produced on complete oxidation of acetyl CoA to CO2 and H2O is equal to ________

46) The dominant pathway of oxidation of fatty acid is _____
47) In non-cyclic photophosphorylation, molecular oxygen is released through _____
48) ______ process act as a link between glycolysis and kreb's cycle
49) The two phases that occur is during photosynthesis is _____ and______-
50) The two basic mechanisms in the synthesis of amino acids are ______ and _____
51) The first amino acid produced during biosynthesis of amino acids is ____
52) The transfer of amino group of amino acid to carboxyl group of keto acid is called ______
53) The ammonia assimilating enzymes are______, _______ and _______
54) An example of carotenoids is ______
55) The coenzyme used in biosynthesis of fatty acid is ________
56) The coenzyme required for transamination is_________
57) Number of ATP molecules formed when one molecule Palmitic acid get oxidized to CO_2 and water is_______
58) Synthesis of carbohydrates in green plants using sunlight, CO_2 and water is known as _____
59) Carbon dioxide fixation in C_3 plants occurs through________
60) The number of ATP molecules produced on complete oxidation of glucose is______
61) The end product is glycolysis is________
62) The common pathway for the metabolism of fats, carbohydrates and amino acid is____
63) The process of glycolysis takes place in______
64) The citric acid cycle takes place in ______ cell
65) The terminal product in the glycolysis pathway is________
66) In glycolysis, one molecule of glucose produces _____ molecules of pyruvic acid
67) The oxidation of pyruvic acid in aerobic condition takes place through________
68) Oxidative phosphorylation takes place inside ________
69) The process of formation of mRNA from DNA is known as______
70) The mechanism of reduction of NADP into NADPH + H+ may be called as______
71) The mechanism of production of ATP molecules from ADP and inorganic phosphate (iP) is called as _________
72) Oxidation of glucose-6-phosphate to 6-phospho gluconic acid without entering glycolysis is called as _____
73) _______ cycle is the final common pathway for oxidation of fuel molecules
74) Glycolysis takes place by a linear sequence of ______ number of enzyme catalyzed reaction
75) Citric acid cycle proceeds in a cyclic way by _____ number of enzyme catalyzed reaction
76) The fatty acid Stearic acid to break down completely into CO_2 and H_2O , the number of cycle of β oxidation needed is_____
77) The acetyl CoA to convert into CO_2 and H_2O has to enter______
78) Reaction between pyruvic acid and NH_3 will give_________
79) The first Amino acid synthesized by reductive amination_______
80) ______ is the process which forms link between glycolysis and kreb cycle
81) The carrier of citric acid cycle is ______

82) NADPH is produced in _______ pathway
83) The four membered aldose sugar phosphate formed in HMP shunt pathway is ___
84) During glycolysis, Fructose 1, 6 diphosphate is decomposed by the enzyme______
85) Under anaerobic conditions the glycolysis of one mole of glucose yields ______moles of ATP.
86) Tricarboxylic acid cycle to be continuous requires the regeneration of _______
87) The number of molecules of ATP produced by the total oxidation of acetyl CoA in TCA cycle is ______
88) Acetyl CoA is not used for the synthesis of _______
89) During each cycle of β-oxidation ________ carbon atom is removed from ______end of fatty acid
90) In deamination, amino acid is converted in to ________
91) Transamination is catalyzed by ________
92) Process of breakdown of amino acids to α keto acids is called __________
93) The key enzyme in the regulation of fatty acid synthesis is _______
94) Fatty acid synthesis takes place in _________
95) Number of ATPs are formed during complete oxidation of palmitate is _______
96) ______ link glycolysis and β-oxidation to citric acid cycle

ANSWER

Sl.No	Answer	Sl.No	Answer
1	Mitochondria	34	DNA
2	Terminal	35	Transcription
3	38	36	Translation
4	AUG	37	UAA/UAG/UGA
5	tRNA	38	Malonyl CoA
6	45	39	fatty acid and glycerol
7	Cyclic Photophosphorylation	40	α, β
8	38	41	Acetyl CoA
9	Kreb cycle (TCA cycle)	42	Number of carbon atoms
10	Thymine, Hydrogen	43	7600
11	H_2O	44	Malonyl CoA
12	PS-1	45	12
13	PS-1, PS-11	46	β
14	3 phosphoglyceric acid	47	Photo oxidation of H_2O
15	Gramineae	48	Oxidative decarboxylation
16	Calvin cycle	49	Light, dark
17	Hatch- Slack	50	reductive amination, transamination
18	3	51	Glutamic acid
19	12	52	Transamination
20	38	53	GDH, GS, GOGAT

21	Light reaction, dark reaction	54	β-carotene
22	Ribulose diphosphate	55	NADPH
23	Oxaloacetic acid	56	Pyridoxal phosphate
24	673	57	130
25	Glycolysis	58	Photosynthesis
26	Kreb’s cycle	59	Calvin Cycle
27	Alcohol	60	38
28	Embden, Meyerhoff and Parnes	61	Pyruvic acid
29	Cytoplasm , Oxygen	62	Kreb’s cycle
30	Mitochondria	63	Cytoplasm
31	Kreb’s cycle	64	Mitochondria
32	Acetyl CoA	65	Pyruvic acid
33	Oxidative phosphorylation	66	Two

Sl.No	Answer	Sl.No	Answer
67	Kreb cycle	82	Pentose phosphate
68	Mitochondria	83	Erythrose
69	Transcription	84	Aldolase
70	Electron transport system	85	2
71	Photophosphorylation	86	Oxaloacetate
72	Pentose phosphate pathway	87	12
73	TCA cycle	88	Glucose
74	10	89	2, carbonyl
75	12	90	keto acid
76	8	91	aminotransferase
77	TCA cycle	92	transamination
78	Phenylalanine	93	acetyl CoA carboxylase
79	Glutamic acid	94	cystol
80	Oxidative decarboxylation	95	129
81	oxaloacetate	96	Acetyl CoA

WRITE SHORT NOTES ON:

1. β-Oxidation

Long chain fatty acids may have up to 16-18 atoms are broken down by the process of β-oxidation which ultimately produces the active 2 C units. B oxidation is the chief process of fatty acid degradation in plants. B oxidation takes place in mitochondria and involves sequential removal of 2 C in the form of acetyl CoA molecules from the carboxyl end of the fatty acid .This is called β- oxidation because β- C of the fatty acid is oxidized during the process. The various steps include as follows

Fatty acid → Fatty acyl- CoA → α-β- unsaturated acyl CoA → β hydroxy acyl CoA β keto acyl CoA → Fatty acyl CoA → Acetyl-CoA

The acetyl CoA produced again reenters the β oxidation spiral at step 2 losing further 2 C units. This sequence is continued until whole molecule is degraded. Thus for each turn of β- oxidation generates 5 ATP molecules except in the first turn where one ATP molecule is consumed resulting it in net gain of 4 ATP molecules.

2. **Synthesis of sucrose**

Sucrose is synthesized in plants by two reactions involving UDP-D glucose. In the first reaction sucrose synthetases catalyzes the transfer of glucose residue of UDP- glucose to fructose with the formation of sucrose and UDP

$$\text{UDP-D-Glucose} + \text{D-fructose} \xleftrightarrow{\text{sucrose synthetase}} \text{Sucrose} + \text{UDP}$$

In the second reaction, sucrose phosphate synthetase catalyzes the reaction of UDP-D-glucose and D-Fructose -6-P to form sucrose phosphate and UDP. Sucrose phosphate undergo dephosphorylation to sucrose in the presence of phosphatases

$$\text{UDP-D-Glucose} + \text{D-Fructose -6-P} \xrightarrow{\text{Sucrose synthetase}} \text{Sucrose –P} + \text{UDP}$$

$$\text{Sucrose –P} \longrightarrow \text{Sucrose} + \text{Pi}$$

3. **Bioenergetics of glucose oxidation**

In glucose oxidation, one molecule of glucose gives 2 molecules of pyruvic acid. In this four molecules of ATP are formed (2 ATP during conversion of 1-3 diphospho glyceric acid to 3 phosphoglyceric acid and 2 ATP during conversion of 2 phopshoenol pyruvic acid to pyruvic acid while 2 ATP molecules are consumed during phosphorylation, Thus during glycolysis there is net gain of 2 ATP molecules(4 ATP- 2 ATP = 2 ATP). Two molecules of NAD are reduced to two molecules of NADH$_2$ which later on oxidized aerobically to yield six molecules of ATP (one NAD molecule after oxidation produces 3 molecules of ATP). Thus the total gain of ATP molecules during glycolysis in presence of O$_2$ will be increased to eight instead of two. The energy of glucose become stored partly in ATP molecules and partly in ATP molecules and partly in NADH$_2$ molecules

3. **Chlorophyll**

Chlorophyll is photosynthetic pigments. Chlorophyll is insoluble in water and can be extracted only with organic solvents. Chlorophyll consists of a, b, c, d, e. Chlorophyll a is present in all the photosynthesizing plants, chlorophyll b is present in higher plants and green algae, chlorophyll c present in diatoms and brown algae, chlorophyll d in some red algae. Chlorophylls are magnesium porphyrin compounds. The porphyrin rings consists of four pyrol rings joined together by CH bonds. A long chain of C atoms called as phytol chain is attached to porphyrin rings. Molecular formula of chlorophyll a is $C_{55}H_{12}O_5N_4Mg$ and Chl b $C_{55}H_{70}O_5Mg$. Chl a and b consists of Mg porphyrin head which is hydrophilic and a phytol tail which lipophilic. The two chlorophyll differ with respect to Chl a -CH$_2$

group and Chl b –CHO group in 3rd carbon atom in II pyrol ring. Chlorophyll is formed from proto chl in light,

4. **C_4 plants**

   C_4 plants leaves possess special anatomy called Kranz type. The chloroplasts in C_4 plants are dimorphic present in mesophyll cells of normal type and chloroplast in bundle sheath. PEP Carboxylase enzyme occurs in mesophyll cells. C_4 cycle is performed in mesophyll cells. C_4 plants possess two types of CO_2 acceptor 1) Phosphoenol pyruvate (in mesophyll cells) 2) Ribulose diphosphate (in bundle sheath cells). C_4 plants are found in subtropical and tropical regions. They grow fast at high temperature and in more light intensities. Therefore C_4 plants are called efficient plants. In C_4 plants, the O_2 has no inhibitory effect. They lack photo respiration.

5. **Dark reaction**

   It is also known as Blackman's reaction or thermo chemical reaction or path of carbon in photosynthesis. It does not require light i.e., it takes place in presence and absence of light. It mainly occurs in the stroma portion of chloroplast. The different steps in dark reaction were studied by Calvin and coworkers. Calvin represented different steps of dark reaction in the form of cycle. Therefore, it is called as Calvin cycle. The dark reaction of photosynthesis is purely enzymatic and slower than the primary photochemical reaction. The various steps in Calvin cycle are as follows

   Six molecules of CO_2 combine with ribulose 1, 5- diphosphate to form 12 molecules of 3 phosphoglyceric acid → 1, 3 diphosphoglyceric acid → 3 phosphoglyceraldehyde →

   Dihydroxyacetone phosphate + 3 phosphoglyceraldehyde → Fructose 1, 6 diphosphate

   → Fructose 6 P + 3 phosphoglyceraldehyde → Xylulose 5 P (2) + Erythrose 4 P (2) + 3 phosphoglyceraldehyde(2) → Sedoheptulose 1,7 diphosphate

   → Sedoheptulose – 7- phosphate + 3- phosphoglyceraldehyde → Xylulose – 5-P + ribose- 5-P → Ribulose -1, 5- diphosphate. Thus completing the cycle.

7. **Transamination**

   This term denotes the reversible transfer of the amino group from an amino acid to a ά-keto acid. There is no liberation of NH_3 unlike during deamination. Transamination is a very important reaction for the formation of amino acid as well as the transformation they undergo. Transaminases are universally distributed and large in number, but two of them are particularly abundant in animal tissues, they catalyse the following two reactions

   L-Glutamic acid + Oxalo acetic acid → ά –ketoglyceric acid + L-aspartic acid

   L-Glutamic acid + pyruvic acid → ά –ketoglyceric acid + L-alanine

   Glutamic acid is a major participant in transamination reaction. Transamination is a transfer of primary amino group of a donor ά- amino acid to an acceptor ά – amino acid.

Transamination can take place on a primary amino group situated at the end of a chain of amino acid.

8. **Oxidative Phosphorylation**

   Synthesis of ATP during oxidation of coenzyme in electron transport system of aerobic respiration is called oxidative phosphorylation. It occurs during respiration and takes place inside mitochondria. It occurs inside the F_1 particles present on the inner membrane of cristae of mitochondria. It requires molecular oxygen for terminal oxidation. Phosphorylation occurs in electron transport system where pigments are not involved. ATP molecules are released into the cytoplasm which is used for various metabolic processes. The complete oxidation of one molecule of glucose in aerobic condition produces 38 ATP molecules (30 ATP molecules in Kreb’s cycle and 8 ATP molecules during glycolysis.

9. **Significance of Kreb’s cycle**

   Kreb’s cycle occupies a central and very important place in the metabolism of plants. It provides energy in the form of ATP molecules through oxidative phosphorylation for various metabolic activities. It is directly related with nitrogen metabolism (ά- keto glutaric acid, an intermediate product of Kreb’s cycle, is the first acceptor molecule of NH_2 in forming of amino acid, the glutamic acid). It is from the glutamic acid that various transamination reaction begins to form different amino acid which ultimately condenses to form proteins. It is also intimately related to fat metabolism (Dihydroxy acetone phosphate produced in glycolysis may be converted into glycerol via ά-glycerophosphate and vice versa.). Glycerol is important constituent of fats. After β- oxidation, fatty acid gives rise to active 2-C units, the acetyl CoA which may enter Kreb’s cycle. Other metabolic processes are related to Kreb’s cycle through its intermediates in one or the other way.

10. **Oxidative decarboxylation of pyruvate**

   Oxidative decarboylation is completed in several steps catalyzed by a enzyme complex pyruvate dehydrogenase. The enzyme complex includes enzyme- pyruvic acid decarboxylase, dihydroxy lipoyl transacetylase, dihydrolipoyl dehydrogenase, coenzymes- thiamine pyrophosphate (TPP), lipoic acid, coenzyme A and NAD. Different steps of oxidative decarboxylation involves

   1) Pyruvic acid is converted to acetaldehyde by the enzyme pyruvic acid decarboxylase
   2) Acetaldehyde is converted into active acetaldehyde
   3) Active acetaldehyde reacts with lipoic acid to form acetyl lipoic acid by the enzyme dihydroxylipoyl transacetylase
   4) Acetyl lipoic acid reacts with coenzyme A resulting in to formation of acetyl CoA and reduced lipoic acid by enzyme dihydrolipoyl dehydrogenase

$$\underset{\text{Pyruvic acid}}{CH_3CO.COOH} + \underset{\text{Coenzyme A}}{Co.A.SH} + NAD \longrightarrow \underset{\text{Acetyl Coenzyme A}}{CH_3CO.S.CoA} + CO_2 + NADH_2$$

11. **Ammonia assimilating enzymes**

Rhizobium during symbiosis fixes the atmospheric nitrogen in the root nodules of the plant. During free living state, bacteria must be able to assimilate nitrogen for their own growth. There are two possible primary pathways for ammonia assimilation into amino acid which depends on bacteria active in a particular species. availability of ammonia and energy. In higher concentration of ammonia, assimilation is catalyzed by glutamate dehydrogenase. The reaction is as follows

$$\text{ά-ketoglutarate} + NH_4^+ \xrightarrow[\text{NADH}]{\text{Glutamate (LDH) dehydrogenase}} \text{L- Glutamate}$$

In the case of limiting ammonia concentration, it occurs with the ATP driven glutamate synthatase and glutamate synthase system

L- Glutamate + NH_4 + ATP Glutamate Synthetase (GS) Glutamine + ADP + iP

$$\text{L- Glutamine} + \text{ά-ketoglutarate} \xrightarrow[\text{NADPH}]{\text{Glutamine Synthase (GOGAT)}} \text{2L-Glutamate}$$

In all legumes, glutamine synthatase (GS) appears to be primarily responsible for the assimilation of ammonia produced by nodule nitrogenase fixation nitrogen. The assimilation of ammonia by GS requires glutamate as substrate for the production of glutamine. Substrate glutamate may be produced by glutamate synthase (GOGAT) which transfer amide nitrogen from glutamine to L- ά-ketoglutarate to produce two glutamate, the substrate of GS. Thus these enzymes GS and GOGAT may work in a cycle that yields glutamate. Glutamate dehydrogenase (GDH) has the capacity to assimilate $NH_4{}^+$ in legume nodules. However it is usually considered to be secondary to the GS_GOGAT cycle in assimilating ammonia.

CARBOHYDRATE METABOLISM

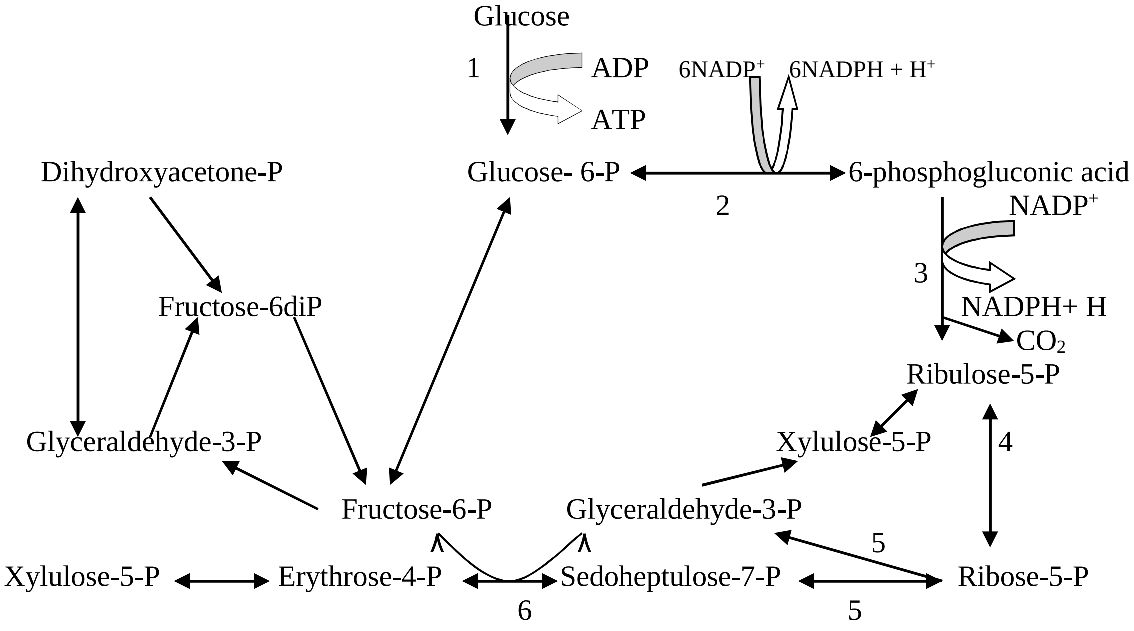
1. **Enumerate the reactions sequence of pentose phosphate pathway. What is its significance?**

Glycolysis is the principal route of the conversion of carbohydrates into pyruvic acid in many biology systems. It has been observed that inhibitors such as iodoacetate, fluorides, aresenates etc. inhibit certain steps in glycolysis. This has led to discovery of alternate route of carbohydrate breakdown in plants. One such alternative route is pentose phosphate pathway. It involves the oxidation of glucose- 6—phosphate to 6- phosphogluconic acid which is converted into pentose phosphates. It is directly oxidized without entering glycolysis, hence it is also called direct oxidation pathway or hexose monophosphate unit

1) Glucose is phosphorylated to glucose-6-phosphate as in EMP pathway
2) Glucose-6-phosphate is oxidized to 6 phosphogluconate with $NADP^+$ is reduced to NADPH(Glucose -6- phosphate dehydrogenase)

3) 6-phosphogluconate is converted to ketopentose sugar- ribulose-5-P with the liberation of one molecule of CO$_2$ for every sugar molecule coenzyme NADP is reduced to NADPH$_2$ (6-phosphogluconate dehydrogenase)

4) Ribulose-5-phosphate is changed to aldopentose sugar Ribose-5-P(phosphoribose isomerase)



Pentose- Phosphate Pathway

5) Two molecules of ribose-5-P combine to form one molecule of Sedoheptulose and another glyceraldehyde -3- phosphate (Transketolase)

6) The 3 carbon atom chain from Sedoheptulose is linked with 3 carbon atom of 3-phosphoglyceraldehyde with the formation of hexose phosphate- fructose-6-P(Transaldolase)

7) A second molecule of fructose-6-P is formed by linking 4 carbon atom (Erythrose-4-P) from Sedoheptulose with 2 carbon fragment pentose molecules(Xylulose-5-P) along with the formation of 3- phosphoglyceraldehyde(Transketolase)

8) Two molecules of 3- phosphoglyceraldehyde like to form fructose 1-6 diphosphate which after dephosphorylation form fructose -6-P

Thus with the formation of fructose-6-P, the cycle is complete in which 6 molecules of glucose enter, but only one of them is completely oxidized liberating 6 molecules of CO$_2$ and the rest are five reformed with 12 molecules of NADPH$_2$

$6\ \text{Glucose-6-P} + 12\ \text{NADP}^+ + 7\text{H}_2\text{O} \longrightarrow 6\text{-Fructose-6-P} + 6\text{CO}_2 + 12\text{NADPH}_2 + \text{H}_3\text{PO}_4$

NADPH$_2$ undergo oxidation by atmospheric O$_2$ via electron transport system where 3 molecules of ATP are produced per NADPH$_2$ oxidized. Thus the oxidation of 12 NADPH$_2$ molecules will produce 12 x 3 = 36 ATP molecules. Therefore this pathway is as efficient as EMP Kreb’s cycle.

Significance

1) It provides alternative route for carbohydrate breakdown and provision of energy
2) It provides ribose sugar for synthesis of nucleic acid
3) It plays important role in fixation of CO_2 in photosynthesis through ribulose -5-P

2 **Explain cyclic and non –cyclic photophosphorylation and their importance in photosynthesis**

The mechanism of reduction of NADP into NADPH + H+ may be called as electron transport system in photosynthesis while the mechanism of production of ATP from ADP and inorganic P with the help of light energy is called photophosphorylation. Two types of photophosphorylation is present namely non-cyclic and cyclic photophosphorylation.

A) Non- Cyclic photophosphorylation.

Pigment system 1 and II are involved in non-cyclic photophosphorylation

1. The pigment molecules of PS I absorb light of higher wavelength and become photo excited and energy is trapped by Chl a 700(P700) and gets excited and loses its outer valence electron to primary acceptor(X) of PS1
2. The reduced unknown substance X (P430) then reduces ferredoxin (Fd) by ferredoxin reducing substance(FRS)
3. The electrons from ferredoxin then pass to NADP+ to reduce it by the enzyme Ferredoxin NADP- reductase
4. The reduced NADP+ participate in CO2 assimilation
5. Simultaneously PS II also absorb photons and get photo excited and energy is transferred to reaction center Chl a 673 which takes part in primary photochemical reaction. The Chl a 673 oxidized water to free O2 and reduce unknown substance Q
6. Photo excited P673 emits outer valence electron and electron is trapped by Q substance which is primary acceptor of PS II
7. Reaction center of PS II on transferring its electron to Q becomes oxidized and oxidized pigment molecule is reduced by trapping electrons from H_2O and this step is known as photo oxidation of water and result in evolution of molecular O_2

$$2\,H_2O \longrightarrow 4e^- + 4\,H^+ + O_2$$

8. Molecular oxygen and 4H+ are released inside the thylakoid membrane
9 The reduced Q or C550, transfer electrons to cytochrome b550
10. Reduced cyt b559 transfers electrons to plastoquinone (PQ) which is H+ carrier
11. Reduced PQ then transfers electrons to cytochrome causing the reduction and oxidation of PQ and ADP is converted to ATP
12. The reduced Cyt F transfers its electron to plastocyanin (PC) and PC is reduced and Cyt F is oxidized
13. The reduced PS transfers electron to PS-1. The PS is oxidized and P700 is reduced In the above scheme, electron ejected from PS II did not return to its place of origin., instead it was passed on PS1. Similarly the electrons emitted from PS 1 did not cycle

back but was used to reduce NADP+ to NADPH + H+. Hence it is called as non-cyclic photophosphorylation

B) Cyclic photophosphorylation

Cyclic photophosphorylation takes place only in PS1 and formation of NADPH2 does not take place. This situation is created by blocking the activity of PS II through certain inhibitors. Under such conditions, only PS 1 is active, photolysis of water does not take place, blockage of non-cyclic ATP formation cause a drop in CO2 assimilation in dark reaction and shortage of oxidized NADP. When P700 molecule in PS 1 gets oxidized by absorbing photons of light, ejected electron is captured by ferredoxin via FRS. From ferredoxin, the electrons cannot be drained off to dark reaction due to shortage of NADP and ultimately it falls back to P700 through number of intermediate electron carriers like cytochrome b6, cytochrome f7 plastocyanin. At two places during electron transport, i.e. between ferredoxin and cytochrome b6¬ and cytochrome f7 there is phosphorylation of one ADP molecule to form one ATP molecule. Since in the above electron transport system, the electron which was emitted from P700 molecule is cycled back, the process is called as cyclic photophosphorylation.

3. **Explain Calvin cycle/ enumerate the CO_2 assimilation pathway in Calvin cycle/ Calvin cycle and its significance**

The dark reaction of photosynthesis is purely enzymatic slower than the primary photochemical reaction. It takes place in stroma portion of the chloroplast and is independent of light. The conversion of CO2 to carbohydrate with the help of assimilatory power (NADPH2 + ATP) in dark reaction of photosynthesis is most thoroughly studied. The different steps of dark reaction were studied by Calvin and Coworkers and represented in the form of cycle is known as Calvin cycle

The Importance of the Calvin Cycle

1. PGAL, the product of the Calvin Cycle can be converted into all sorts of other molecules.
2. Glucose phosphate is one result of PGAL metabolism; it is a common energy molecule.
3. Glucose phosphate is combined with fructose to form sucrose used by plants.
4. Glucose phosphate is the starting point for synthesis of starch and cellulose.
5. The hydrocarbon skeleton of PGAL is used to form fatty acids and glycerol; the addition of nitrogen forms various amino acids

Various steps are

1) The CO_2 is accepted by ribulose 1-5 Diphosphate to form 6 carbon compound ,being unstable converted into 2 molecules of 3 phosphoglyceric acid by hydrolysis and dismutation
2) 3-Phosphoglyceric acid is reduced to 3 phosphoglyceraldehyde by enzyme triose phosphate dehydrogenase

3) Some molecules of 3 phosphoglyceraldehyde isomerizes into Dihydroxy acetone phosphate and both of which join together in the presence of Aldolase to form fructose 1-6 diphosphate
4) Fructose 1-6 diphosphate is converted into fructose 6- phosphate in the presence of phosphatase
5) Fructose 6-P is converted into sucrose

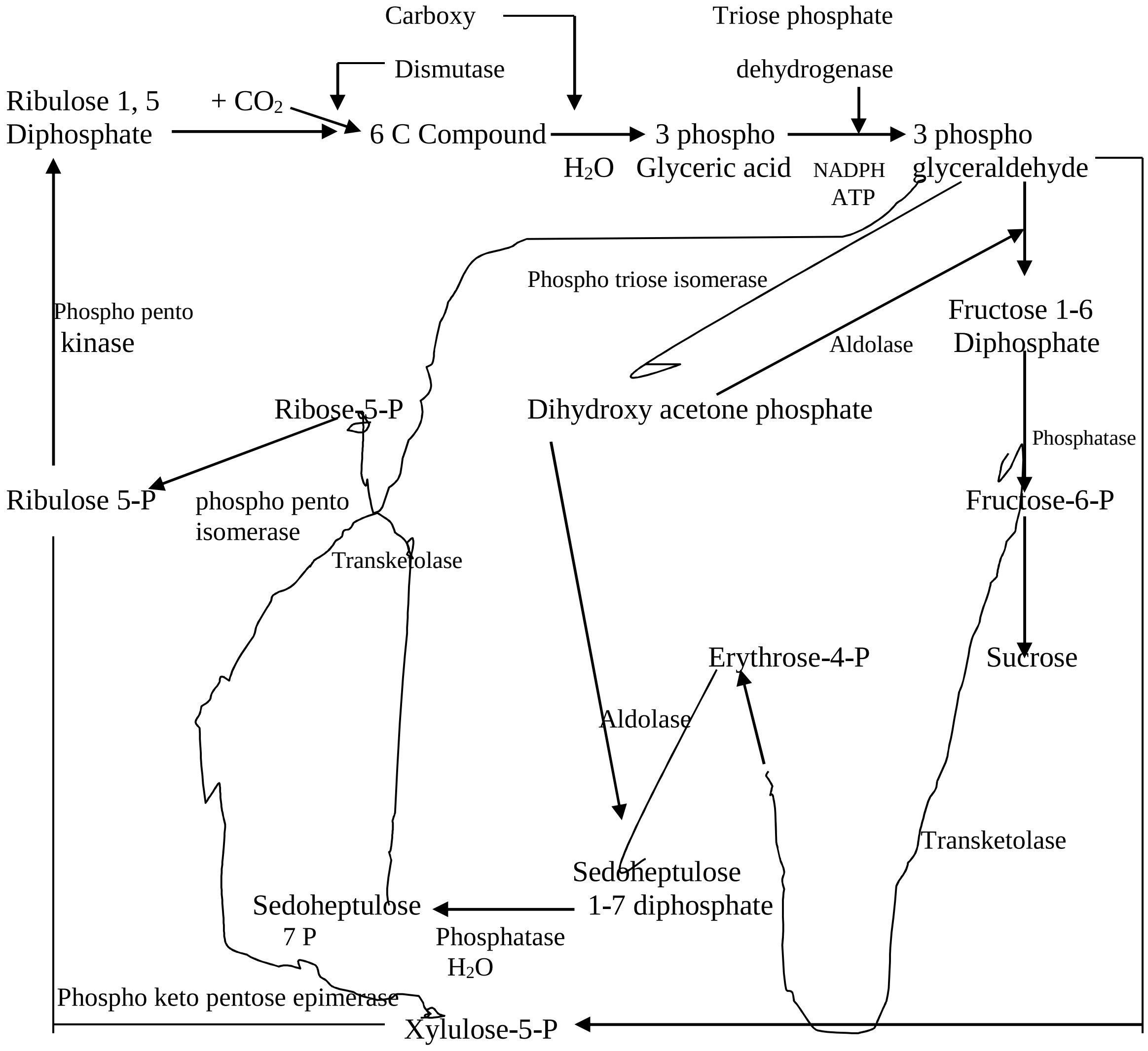


Fig: Calvin cycle

6) Some molecules of 3 phosphoglyceraldehyde reacts with fructose 6-P in the presence of transketolase to form Erythrose -4-P (4 C atom sugar) and Xylulose -5-P (5 C atom sugar)
7) Erythrose -4-P reacts with Dihydroxy acetone phosphate in the presence of Aldolase to form Sedoheptulose 1-7 diphosphate(7 C atom)
8) Sedoheptulose 1-7 diphosphate is converted to Sedoheptulose 7-phosphate by phosphatase

9) Sedoheptulose 7 P reacts with 3 phosphoglyceraldehyde in the presence of transketolase to form Xylulose -5-P and Ribose -5-P (5 carbon atom)
10) Xylulose -5-P is converted into ribulose 5-P in the presence of phospho keto pentose epimerase
11) Ribose -5-P is converted into ribulose 5 phosphate in the presence of phospho pentose isomerase
12) Ribulose -5-P is converted into Ribulose 1-5 diphosphate in the presence of phospho pentose kinase and ATP, thus completing the cycle
    Because first visible product of this cycle is 3 phosphoglyceric acid which is 3 C Compound, **Calvin cycle is known as C_3 pathway**

4. **Why C_4 plants are more efficient than C_3 plants?**
    1. In C_4 plants, it increases the photosynthetic yield two or three times more than C_3 Plants.
    2. In C_4 plants, it performs a high rate of photosynthesis even when the stomata are nearly closed
    3. It increases the adaptability of C_4 plants to high temperature and light intensities
    4. It increases the rate of CO_2 fixation at 25-30^o C in C_4 plants as compared to C_3 plants
    5. It reduced the rate of photo respiration at 25-30^{o}C
    6. C_4 plants leaves possess special type of anatomy called Kranz type
    7. Chloroplast in C_4 leaves are dimorphic
    8. PEP carboxylase enzyme occur in mesophyll cells
    9. C_4 cycle is performed in mesophyll cells
    10. They possess two types of CO_2 acceptor -1) Phosphoenol pyruvate 2) Ribulose diphosphate
    11. First stable compound formed is oxaloacetic acid
    12. C_4 plants are found in tropical and sub-tropical regions
    13. C_4 plants, the O_2 has no inhibitory effect. Hence C_4 plants are called as efficient plants

5. **Write the structure of chlorophyll and carotenes. Explain their role in photosynthesis**

    Chlorophylls and carotenoids are insoluble in water and can be extracted only with organic solvents. Carotenoids include carotenes and xanthophylls. Different pigments absorb light of different wavelength and show characteristic absorption peaks. Chlorophylls are of different types. Chlorophyll a is present in all photosynthezing plants, chlorophyll b in higher plants and algae, chlorophyll c in diatoms and brown algae, chlorophyll d in some red algae.

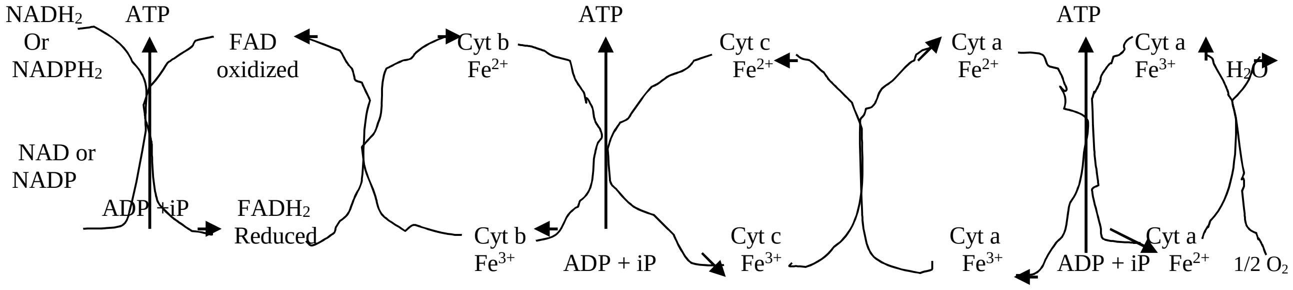
    Chlorophylls are magnesium porphyrin compounds. The porphyrin ring consists of four pyrrol rings joined together by CH bridges. A long chain of C atoms called as phytol chain is attached to porphyrin ring. Both chlorophyll a and b have Mg porphyrin head which is hydrophilic and a phytol tail which is lipophilic. Chlorophyll is formed from proto chlorophyll in light.

Carotenes consist of an open chain conjugated double bond system ending on both sides with ionone rings. They are hydrocarbon. Different carbons differ only in their arrangement of their molecules in space

Chlorophyll is vital for photosynthesis, which allows plants to absorb energy from light. Chlorophyll molecules are specifically arranged in and around photosystems that are embedded in the thylakoid membranes of chloroplasts. In these complexes, chlorophyll serves two primary functions. The function of the vast majority of chlorophyll (up to several hundred molecules per Photosystem) is to absorb light and transfer that light energy by resonance energy transfer to a specific chlorophyll pair in the reaction center of the photosystems.

The two currently accepted photosystem units are Photosystem II and Photosystem I, which have their own distinct reaction center chlorophylls, named P680 and P700, respectively.[3] These pigments are named after the wavelength (in nanometers) of their red-peak absorption maximum. The identity, function and spectral properties of the types of chlorophyll in each photosystem are distinct and determined by each other and the protein structure surrounding them. Once extracted from the protein into a solvent (such as acetone or methanol),[4][5][6] these chlorophyll pigments can be separated in a simple paper chromatography experiment and, based on the number of polar groups between chlorophyll a and chlorophyll b, will chemically separate out on the paper. The function of the reaction center chlorophyll is to use the energy absorbed by and transferred to it from the other chlorophyll pigments in the photosystems to undergo a charge separation, a specific redox reaction in which the chlorophyll donates an electron into a series of molecular intermediates called an electron transport chain. The charged reaction center chlorophyll ($P680^+$) is then reduced back to its ground state by accepting an electron. In Photosystem II, the electron that reduces $P680^+$ ultimately comes from the oxidation of water into O_2 and H^+ through several intermediates. This reaction is how photosynthetic organisms such as plants produce O_2 gas, and is the source for practically all the O_2 in Earth's atmosphere. Photosystem I typically works in series with Photosystem II; thus the $P700^+$ of Photosystem I is usually reduced, via many intermediates in the thylakoid membrane, by electrons ultimately from Photosystem II. Electron transfer reactions in the thylakoid membranes are complex, however, and the source of electrons used to reduce $P700^+$ can vary. The electron flow produced by the reaction center chlorophyll pigments is used to shuttle H^+ ions across the thylakoid membrane, setting up a chemiosmotic potential used mainly to produce ATP chemical energy; and those electrons ultimately reduce $NADP^+$ to NADPH, a universal reductant used to reduce CO_2 into sugars as well as for other biosynthetic reductions.

6. **Write the components of electron transport chain in sequence and indicate sites of ATP molecules.**



The respiratory breakdown of glucose in the presence of oxygen is an oxidative process. During aerobic respiration, simple carbohydrates and intermediates are oxidized. Each oxidative step involves release of a pair of hydrogen atom (2H) which dissociate into two protons ($2H^+$) and two electrons($2e^-$). The pair of hydrogen atom ($2H^+ + 2e^-$) released in each oxidative step of Kreb's cycle do not combine directly with oxygen but pass through a series of coenzymes and cytochromes which form electron transport chain before reacting with O_2 to form H_2O. During the transfer of hydrogen atoms from one coenzyme to another coenzyme a large amount of energy is released which is picked up by ADP to form ATP with the help of inorganic phosphate. Electron transport system is made up of coenzymes NAD^+ and $NADP^+$, FAD, coenzyme Q, cytochromes b, C_1,C_2, a, a_1. The transfer of electrons in all the compounds takes place first in NAD^+ or $NADP^+$ and later FAD. Three ATP molecules are produced for each NADH + H^+ or NADPH + H^+ molecules. Only two ATP molecules are produced for each $FADH_2$ molecules. The reduction of various cytochromes requires only electrons and no protons. The formation of one molecule of water require $½ O_2 + 2e^- + 2H^+$, while reduction of one molecule of O_2 requires $4e^- + 4H^+$. The reduction and oxidation of coenzymes and cytochromes takes place in sequence and step wise because in electron transport system, they are arranged according to their redox potential. The first coenzyme (NAD^+) possesses low redox potential while last cytochrome (cyt a_2) is highest. This transfer of electrons proceeds from compounds with low redox potential to those in high redox potential. Complex oxidations of glucose molecules need 38 ATP molecules. Because huge energy is generated in mitochondria, it is referred to as power house of the cell wall

7. **How acetyl CoA is oxidized to CO_2 in TCA cycle/ describes Kreb's Cycle. What is its significance?**

Pyruvic acid produced during glycolysis enters into Kreb's cycle for further oxidation. Kreb's cycle is also called as citric acid cycle or tricarboxylic acid cycle. It takes place in mitochondria, where all the necessary enzymes s are found in cristae

1. Pyruvic acid reacts with CoA and NAD and oxidatively decarboxylated and is converted into acetyl CoA
2. Acetyl CoA condenses with oxaloacetic acid in the presence of condensing enzyme

3. Citric acid is dehydrated in the presence of aconitase to form Cis-aconitic acid
4. Cis-aconitic acid reacts with water to form iso-citric acid
5. Iso-citric acid is oxidized to Oxalo Succinic acid in the presence of iso-citric dehydrogenase. Coenzyme II NADH is reduced
6. Oxalic Succinic acid is decarboxylated in the presence of Oxalo-Succinic decarboxylase to form ά- ketoglutaric acid
7. ά- ketoglutaric acid reacts with CoA and NAD in the presence of ά- ketoglutaric acid dehydrogenase to form succinyl CoA
8. Succinyl CoA react with water molecule to form Succinic acid, GDP is converted to GTP
9. Succinic acid is oxidized to fumaric acid in the presence of Succinic dehydrogenase. FAD is reduced
10. Fumaric acid reacts with H_2O in the presence of Fumarase to form malic acid
11. Malic acid is oxidized to form oxaloacetic acid in the presence of malic dehydrogenase NAD is reduced. Thus cycle is repeated with another molecule of acetyl CoA.

Significance of Kreb's Cycle

Kreb's cycle occupies a central and very important place in the metabolism of plants. It provides energy in the form of ATP molecules through oxidative phosphorylation for various metabolic activities. It is directly related with nitrogen metabolism (ά- keto glutaric acid, an intermediate product of Kreb's cycle , is the first acceptor molecule of NH_3 in forming of amino acid, the glutamic acid. It is from the glutamic acid that various transamination reactions begin ultimately condensing to form proteins. It is also intimately related to fat metabolism (Dihydroxy acetone phosphate produced in glycolysis may be converted into glycerol via ά- glycerophosphate and vice versa). Glycerol is important constituent of fats. After β-oxidation, fatty acid give rise to active 2-C units, the acetyl CoA which may enter the Kreb's cycle. Other metabolic processes are related to Kreb's cycle through its intermediates in one or other way.

8. **Enumerate the reaction sequence from glucose to pyruvic acid/ Explain glycolysis**

1) Glucose react with ATP in the presence of hexokinase to form Glucose -6-P
2) Glucose-6-P is isomerised to fructose-6-P by phosphohexoisomerase
3) Fructose-6-P react with ATP in the presence of phosphohexokinase to form Fructose 1-6-diphosphate
4) Fructose 1-6-Diphosphate is converted into two trioses 3-Phosphoglyceraldehyde and dihydroxy acetone phosphate in the presence of aldolase
5) 3-Phosphoglyceraldehyde react with H_3PO_4 to form 1,3- diphosphoglyceraldehyde
6) 1,3- diphosphoglyceraldehyde in the presence of triose phosphate dehydrogenase and coenzyme NAD to form 1,3 diphosphoglyceric acid
7) 1,3- diphosphoglyceric acid reacts with ADP in the presence of phosphoglyceric trans phosphorylase to form 3 phosphoglyceric acid and ATP

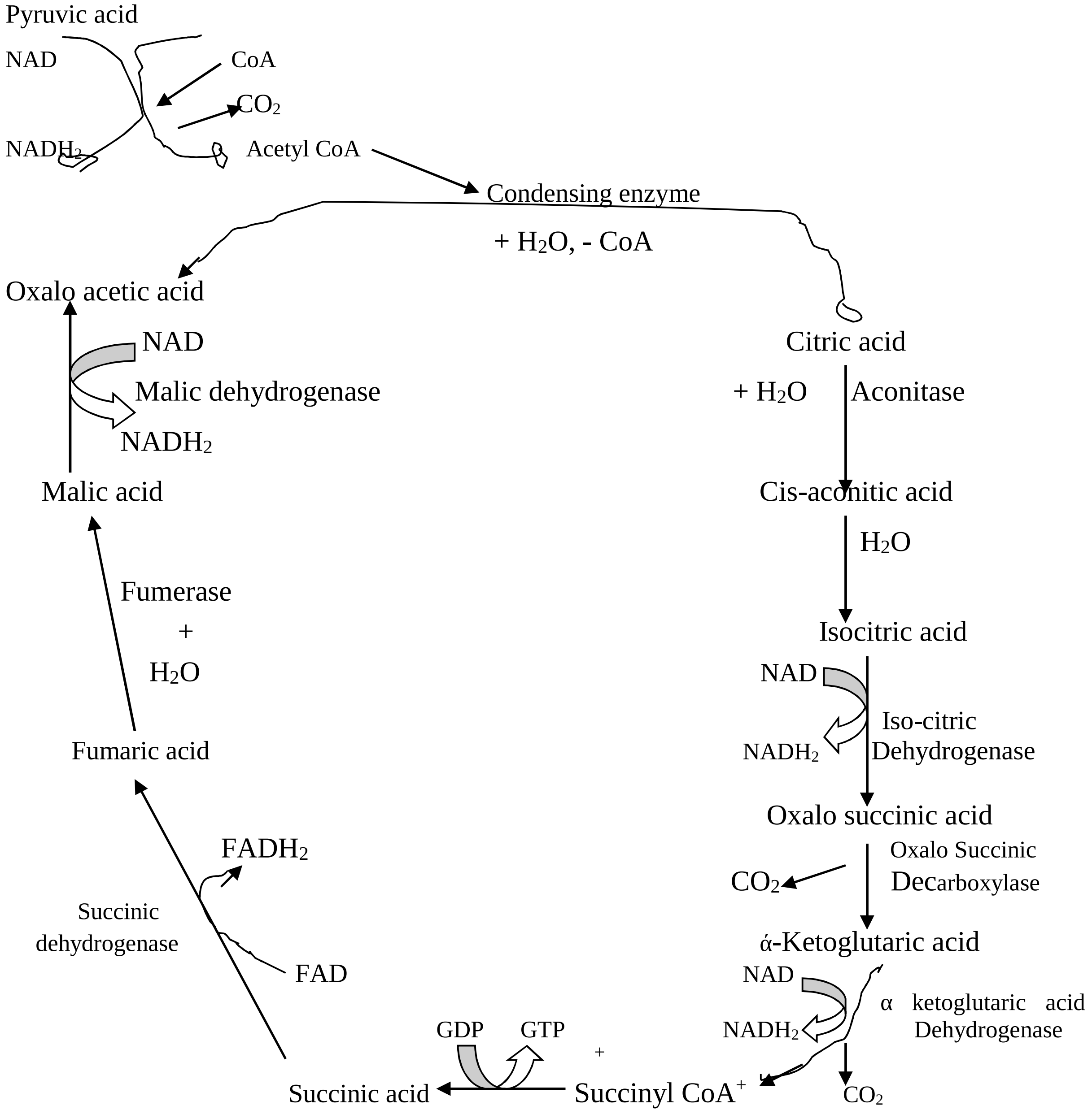


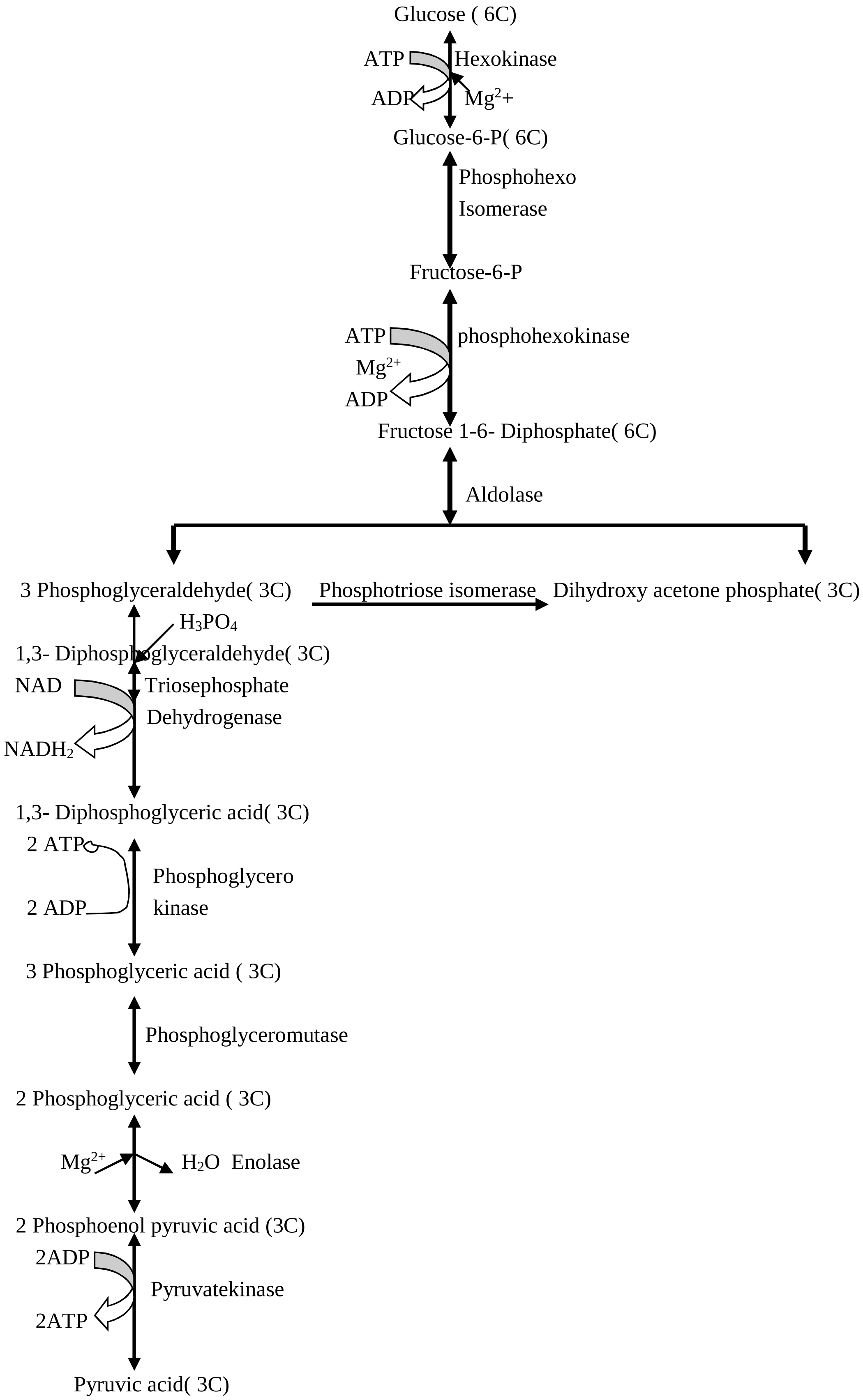
Fig. TCA Cycle

8) 3- phosphoglyceric acid is isomerised into 2-phosphoglyceric acid in the presence of phosphoglyceromutase

9) 2- phosphoglyceric acid is converted into 2 phosphoenol pyruvic acid in the presence of enolase

10. - 2-phosphoenol pyruvic acid reacts with ADP to form pyruvic acid and ATP

` The Pyruvic acis so formed unger go either aerobic decpmosition or anarobic decomposition depending on the availability of oxygen. In the absence of oxygen, pyruvic acid is converted into alcochol(alcoholic fermentation) or lactic acid (lactic acid fermentation). In the presence of oxygen, pyruvic acid undergo oxidation by entering Kreb's cycle.


Glucose (6C)
ATP
Hexokinase
ADP
Mg2+
Glucose-6-P(6C)
Phosphohexo
Isomerase
Fructose-6-P
ATP
phosphohexokinase
Mg2+
ADP
Fructose 1-6- Diphosphate(6C)
Aldolase
3 Phosphoglyceraldehyde(3C)
Phosphotriose isomerase
Dihydroxy acetone phosphate(3C)
H3PO4
1,3- Diphosphoglyceraldehyde(3C)
NAD
Triosephosphate
Dehydrogenase
NADH2
1,3- Diphosphoglyceric acid(3C)
2 ATP
Phosphoglycero
kinase
2 ADP
3 Phosphoglyceric acid (3C)
Phosphoglyceromutase
2 Phosphoglyceric acid (3C)
Mg2+
H2O Enolase
2 Phosphoenol pyruvic acid (3C)
2ADP
Pyruvatekinase
2ATP
Pyruvic acid(3C)

9. **Discuss the synthesis of starch**

Synthesis of starch involves the simultaneous synthesis of amylose (with ά (1,4) glycosidic linkage) and amylopectin (with (ά 1,6 glycosidic linkage), two important constituent of starch

A) **Synthesis of amylose**

Synthesis of amylose may take place by any one of the following way

1) Amylose can be synthesized in the presence of enzyme starch phosphorylase from glucose 1-phosphate and an acceptor molecule containing of about 3 to 20 glucose units together by ά (1-4) glycosidic linkage

$$\text{n (Glucose -1-phosphate + acceptor)} \xleftrightarrow[\text{Starch phosphorylase}]{\hspace{3em}} \text{Amylose + n(iP)}$$

1) Formation of ά(1-4) glycosidic linkage may also take place in the presence of enzyme UDPG trans glycosylase(amylose synthatase) by their transfer of glucose from UDPG (uridine diphosphate glucose) to an acceptor molecule consisting of 2 to 4 or more glucose units joined together by ά(1-4) glycosidic linkages

$$\text{UDPG + Acceptor} \xleftrightarrow{\text{Amylose synthetase}} \text{UDP + ά(1-4) glucosyl- acceptor}$$

2) Formation of ά (1-4) glycosidic linkage leading to the synthesis of amylose may also take in the presence of D-enzyme by the transfer of two or more glucose units from malto dextrin to a variety of acceptors such as malto triose

B) **Synthesis of amylopectin (ά 1, 6 glycosidic linkages)**

It takes place in the presence of Q-enzyme by the transfer of small chains of glucose units joined together by ά (1-4) glycosidic linkages to an acceptor molecule consisting of at least four ά (1-4) linked glucose units. The (ά 1, 6 glycosidic bonds established between C_1 of the terminal glucose units of the donor molecule and C_4 of one of the glucose unit of the receptor.

FAT METABOLISM

1) **How is Palmitic acid is biosynthesized?**

1) The main pathway of saturated fatty acid synthesis in plants and animals takes place through malonyl CoA pathway. Long chain saturated fatty acid is synthesized in plants from active two carbon units, the acetyl CoA. Synthesis of fatty acids from $CH_3CO.CoA$ takes place step by step. In each step fatty acid chain is increased by two carbon atoms

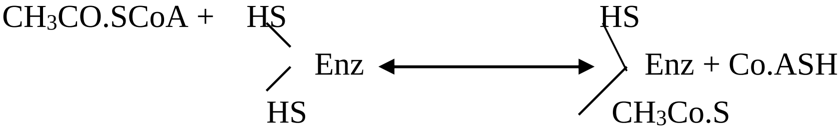
$$\text{Acetyl CoA} + CO_2 \xrightarrow{\text{Acetyl CoA Carboxylase}} \text{Malonyl- CoA + ADP + iP}$$

2) Malonyl CoA reacts with another molecule of acetyl CoA in the presence of fatty acid synthetase and coenzyme $NADPH_2$ to form coenzyme-A derivative of butyric acid (4 C atom)

$$\text{Malonyl CoA + Acetyl CoA} + 2NADPH_2 \rightarrow \text{Butryl CoA + CoA} + CO_2 + H_2O + 2NADP$$

3) Butryl CoA in the next step will combine with malonyl CoA to form CoA derivative of fatty acid containing 6 C atoms. This process will be repeated till coenzyme A derivative of long chain fatty acid (which may contain up to 16-18 C atoms-palmitic acid) is produced The enzyme involved is fatty acid synthetase is not simple but a complex of many enzymes and an acyl carrier protein called ACP is used for catalyze the reaction

The biosynthesis of saturated fatty acid involves 3 steps 1) Initial reaction when acetyl CoA transfers its acetyl group to one of the SH group of multi enzyme complex fatty acid synthetase



$$CH_3CO.SCoA + \begin{matrix} HS \\ HS \end{matrix} > Enz \longleftrightarrow \begin{matrix} HS \\ CH_3Co.S \end{matrix} > Enz + Co.ASH$$

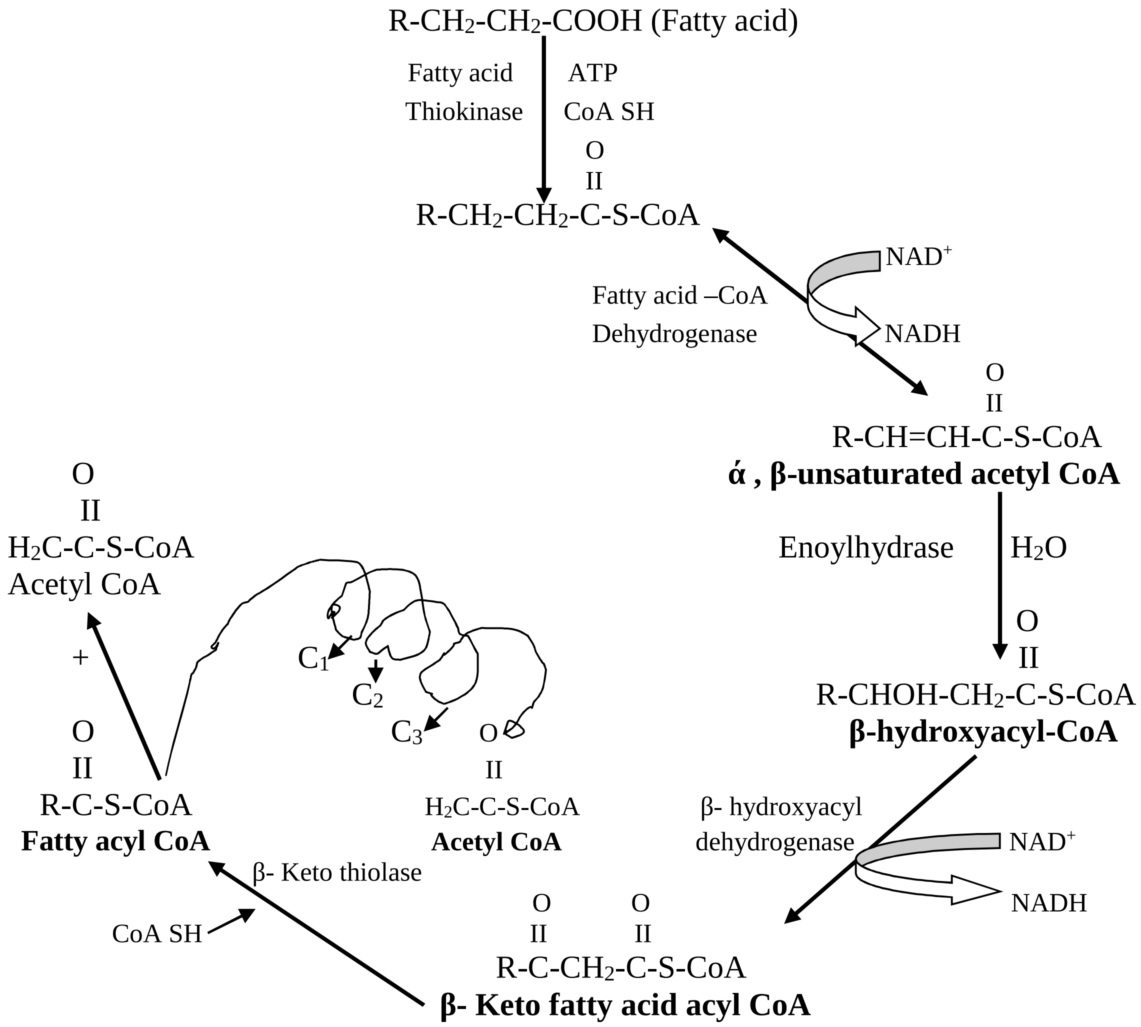
2) Chain elongation reaction- Here six different reactions are involved a) Malonyl transfer b) Condensation 3) Reduction 4 Dehydration 5) Reduction 6) Acyl transfer

3) Termination reaction- When the fatty acid residue has attained a desired length, the chain elongation stops at a reaction (reduction) and cycle is not repeated.

2. **Explain β oxidation and its importance/ How Palmitic acid is is oxidized through β oxidation**

β oxidation is the chief process by fatty acid degradation in plants. This is one of the methods of oxidation of fatty acid. The β oxidation mechanism is well established for saturated fatty acid. The β oxidation takes place in mitochondria and involves sequential removal of 2 C in the form of acetyl CoA from the carboxyl end of the fatty acid. This is called as β oxidation because β –C of the fatty acid is oxidized during the process. Various steps of β oxidation are

1. Activation of fatty acid in the presence of ATP and enzyme Thiokinase, Co.ASH is consumed and CoA derivative of fatty acid is produced
2. Two hydrogen atom are removed between ά, β –C atoms and a trans ά, β unsaturated fatty acid acyl CoA is formed which catalyzed by FAD containing enzyme acyl CoA dehydrogenase
3. Addition of water molecule across the double bond to form β-hydroxyacyl CoA in the presence of enoylhydrase
4. β-hydroxy acyl-CoA is dehydrogenated in the presence of NAD, specific β-hydroxyacyl-CoA dehydrogenase to form β-keto fatty acyl CoA
5. β keto acyl CoA is cleaved by enzyme β-keto acyl thiolase to form 2C unit of acetyl CoA and a fatty acyl-CoA molecule which is shorter by two carbon atoms than when entered the β- oxidation spiral



β-Oxidation spiral of fatty acid

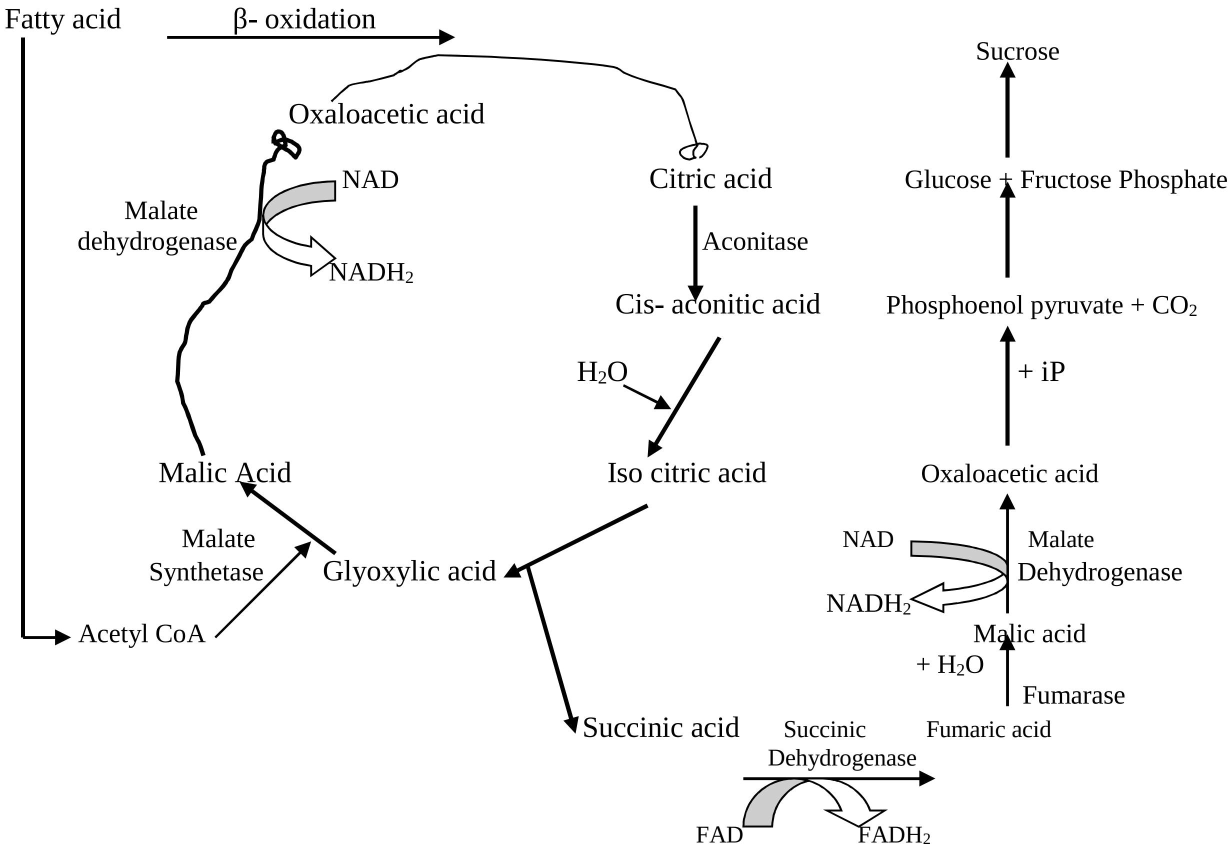
The fatty acyl CoA produced again reenters the β- oxidation of fatty acid may be completely oxidized to CO_2 and H_2O through TCA cycle and may also be converted into carbohydrate s by glyoxylate cycle. This conversion takes place only in case of plants. In each turn of β- oxidation generates 5 ATP molecules and however in the first turn there is compensation of 1 ATP in the first step, hence there will be only 4 ATP molecules. Complete oxidation of one acetyl CoA molecule in TCA cycle to CO_2 and H_2O will result 12 ATP molecules.

Importance of β- oxidation is that sufficient amount of utilizable energy is produced for example. Complete oxidation of palmitic acid produced through β- oxidation and TCA cycle produces 130 ATP molecules. Each ATP molecule represents a gain of about 7600 calories of free energy. Thus the complete oxidation of one molecule of palmitic acid by β- oxidation and TCA cycle will show a gain of 7600 x 136 = 9, 88,000 calories of utilizable energy.

3. **Explain the chemistry of seed germination/ Explain glyoxylate cycle**

Many fatty seeds convert their fats into carbohydrates through glyoxylate cycle. This conversion takes place as the seeds germinate, where fat content is decreased with a simultaneous increase in sucrose.

1 Acetyl CoA produced after β – oxidation of fatty acid condenses with Oxalo acetic acid to form citric acid
2. Citric acid is dehydrated to produce cis-aconitic acid in the presence of aconitase
3. Cis-aconitic acid reacts with one molecule of H_2O to form iso-citric acid
4. Iso-citric acid is broken down into glyoxylic acid and Succinic acid by the enzyme iso citratase
5. Glyoxylic acid combines with acetyl CoA in the presence of malate synthetase to produce malic acid
6. Malic acid is oxidized into oxaloacetic acid in the presence of malic dehydrogenase and coenzyme NAD
7. Succinic acid is converted into Oxalo acetic acid through fumaric acid and malic acid
8. Oxaloacetic acid produced is decarboxylated in the presence of inosine triphosphate to form Phosphoenol pyruvic acid
9. Phosphoenol pyruvic acid by the reverse reaction of glycolysis is converted into glucose and fructose phosphate
10. Finally glucose and fructose phosphate are converted into sucrose



Glyoxylate Cycle

The significance of glyoxylate cycle is that the germination of fatty seeds, the fats which are insoluble is hydrolyzed into fatty acid and glycerol. Fatty acids after β- oxidation produce acetyl CoA units which synthesis sucrose which is supplied to different growing regions of the young germinating seedlings. Those microorganisms which can grow on ethyl alcohol or acetates as a sole source of energy and carbon, make use of this cycle in synthesizing longer carbon chains

4. Give brief note on fat synthesis

Fats are synthesized in plants through following steps

1. **Synthesis of glycerol**

Glycerol portion of the fat is synthesized from Dihydroxy acetone phosphate which is produced from fructose 1-6 diphosphate in the presence of Aldolase and cofactor Zn^{2+} and Cu^{2+}, as an intermediate product in glycolysis. It is done in 2 steps

1) Dihydroxy acetone phosphate is reduced to ά - glycerophosphate by enzyme ά-glycerophosphate dehydrogenase and coenzyme $NADH_2$
2) ά – glycerophosphate is then hydrolyzed by phosphatase to liberate phosphoric acid and glycerol

2. **Synthesis of fatty acids**

Long chain saturated fatty acids are synthesized in plants from active 2 Carbon atom units acetyl CoA. Synthesis of fatty acids from acetyl CoA takes place step by step. In each step the fatty acid chain is increased by two carbon atoms.

$\text{Acetyl CoA} + CO_2 \xrightarrow{\text{Acetyl CoA Carboxylase}} \text{Malonyl- CoA} + \text{ADP} + \text{iP}$

2) Malonyl CoA reacts with another molecule of acetyl CoA in the presence of fatty acid synthetase and coenzyme $NADPH_2$ to form coenzyme-A derivative of butyric acid (4 C atoms)

$\text{Malonyl CoA} + \text{Acetyl CoA} + 2NADPH_2 \rightarrow \text{Butryl CoA} + \text{CoA} + CO_2 + H_2O + 2NADP$

3) Butryl CoA in the next step will combine with malonyl CoA to form CoA derivative of fatty acid containing 6 C atoms. This process will be repeated till coenzyme A derivative of long chain fatty acid (which may contain up to 16-18 C atoms-palmitic acid) is produced

The enzyme involved is fatty acid synthetase is not simple but a complex of many enzymes and an acyl carrier protein called ACP is used for catalyze the reaction. The synthesis of fatty acid involves 3 categories namely initiation reaction, chain elongation reaction and termination reaction

3. **Condensation of fatty acids and glycerol**

The fats or triglycerides are synthesized not from glycerol and free fatty acids but from ά- glycerophosphate and CoA derivatives of fatty acid

1) The glycerol is phosphorylated to form L-ά – glycerophosphate
2) ά – glycerophosphate undergo condensation with 2 molecules of acyl CoA to form ά-phosphatidic acid (Acyltransferase)
3) ά- phosphatidic acid undergo dephosphorylation in the presence of phosphatase to form ά, β- diglyceride

4) Condensation of one molecule of an acyl –CoA with free hydroxyl group of the ά, β-diglyceride takes place in the presence of enzyme diglyceride acyl transferase to form triglyceride

PROTEIN METABOLISM

1. **What are the different amino acids that constitute proteins in plants? Give an account of their biosynthesis**

Plant protein consists of over 20 different types of amino acids. Amino acids are grouped into three groups

a) **Aliphatic amino acids**
   1) Mono amino monocarboxylic amino acids- alanine, valine, leucine
   2) Sulfur containing amino acid- Methionine, cystine cysteine
   3) Mono amino dicarboxylic amino acids- Aspartic acid, asparagine, glutamic acid, glutamine
   4) Basic amino acid- lysine, arginine, histidine

b) Aromatic amino acid – Phenylalanine, tyrosine

c) Heterocyclic amino acid –tyrptophan, proline, hydroxy proline

Biosynthèses of Amino acids

Amino acids are synthesized in plants in the following ways

1) Inorganic nitrogen in the form of NH_3 reacts with ά- ketoglutaric acid (an intermediate of Kreb’s cycle) in the presence of enzymes glutamic dehydrogenase and reduced coenzyme $NADPH_2$ to form n amino acid , the glutamic acid

$$COOH\text{-}CH_2\text{-}CH_2\text{-}C{=}O\text{-}COOH + NADPH_2 + NH_3 \xrightarrow[\text{Dehydrogenase}]{\text{Glutamic}} COOH\text{-}CH_2\text{-}CH_2\text{-}CHNH_2\text{-}COOH + NADP + H_2O$$

ά- Ketoglutaric acid Glutamic acid

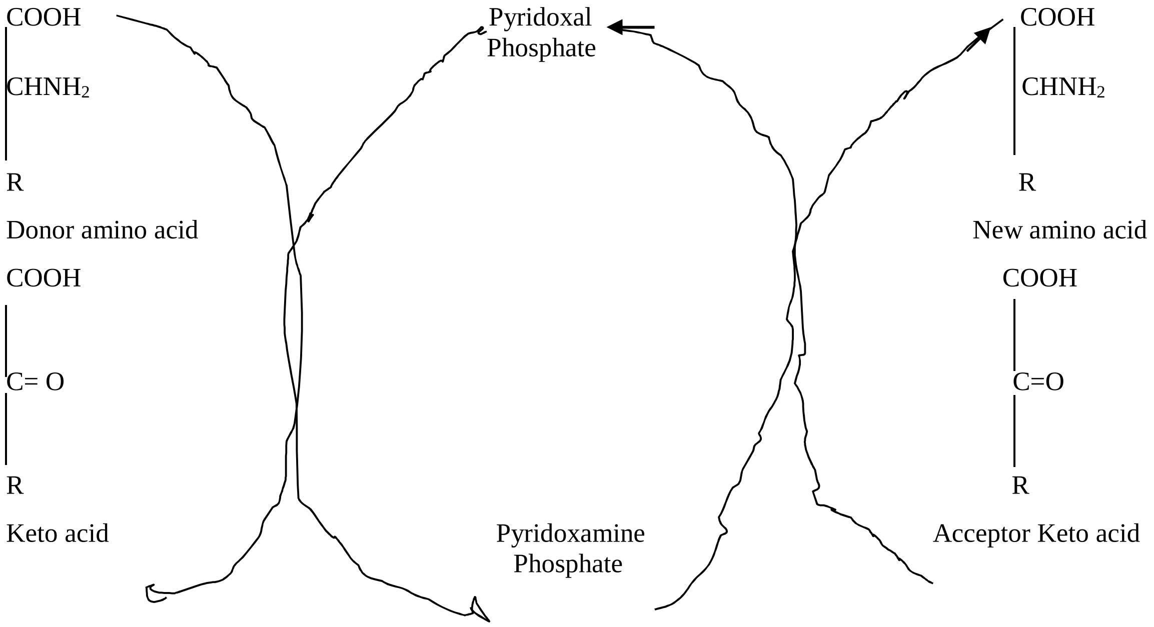
It is called as reductive amination since the above process involve conversion of the inorganic nitrogen (NH_3) into organic nitrogen (amino acid) is accompanied by aminations and reduction at the keto group of the organic acid

2) **Transamination**

The various other amino acids which ultimately condense to form proteins are produced by transamination reactions involving the transfer of amino group from glutamic acid to the keto position of the corresponding keto acid. Amino group from other amino acid except glutamic acid may also be transferred to other keto acids forming corresponding amino acids. Transamination reaction takes place in the presence of enzymes transaminases which require coenzyme pyridoxal phosphate

The coenzyme pyridoxal phosphate act as carrier of amino group. It picks up the amino group from the donor amino acid and is converted into Pyridoxamine phosphate. The latter transfer this amino group to the acceptor keto acid forming a new amino acid and itself is converted into pyridoxal phosphate for example

Glutamic acid + Oxalo acetic acid → ά- ketoglutaric acid + aspartic acid
Glutamic acid + Pyruvic acid → ά- ketoglutaric acid + ά- alanine
Aspartic acid + pyruvic acid → oxaloacetic acid + ά- alanine



2. Explain how proteins undergo oxidation?

The oxidation of proteins involves two steps 1) Hydrolysis or degradation of proteins to amino acids 2) oxidation of amino acids

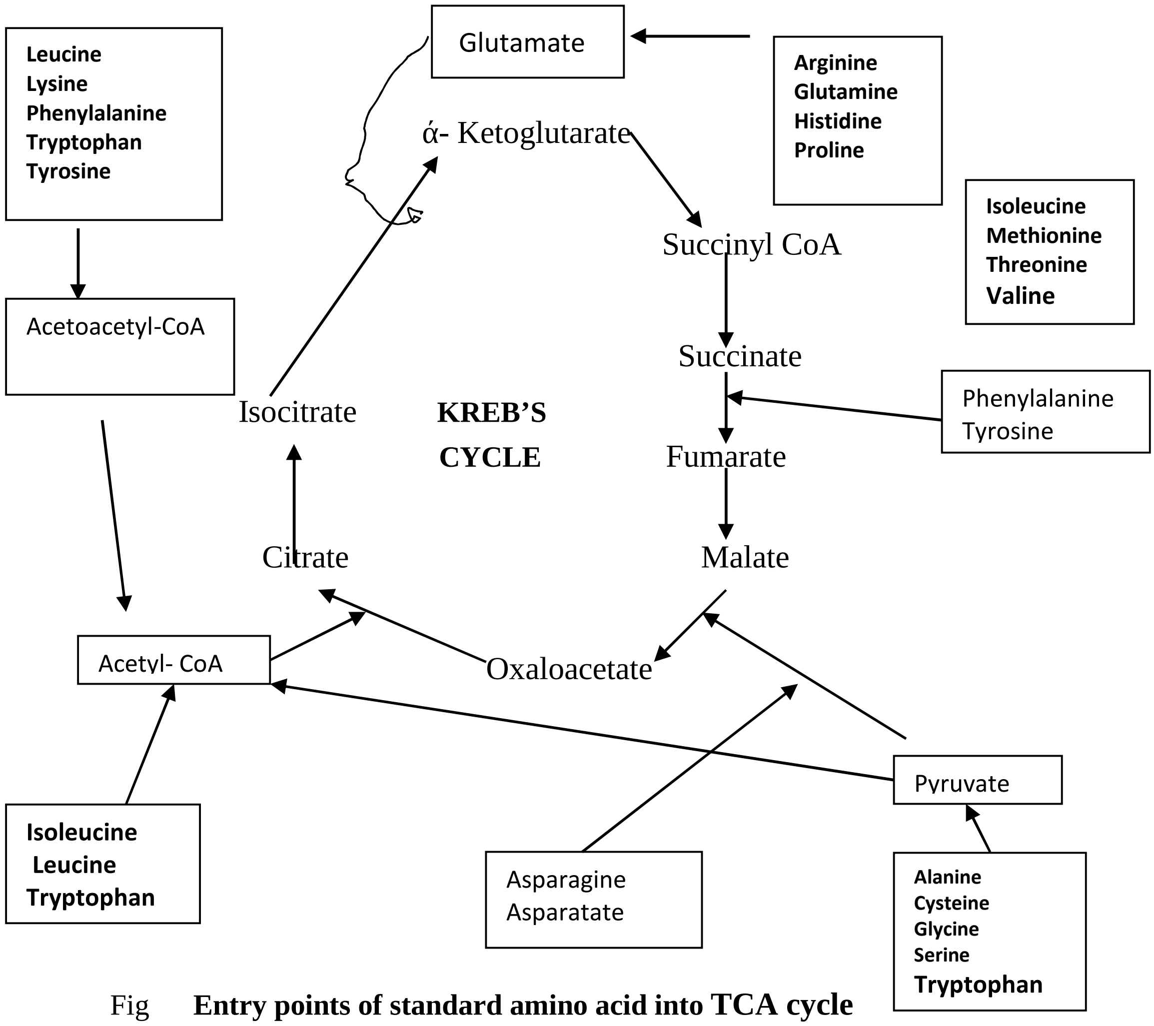


Fig **Entry points of standard amino acid into TCA cycle**

The hydrolysis of proteins can be done by acids, alkalies or enzymes. The hydrolysis of proteins by enzyme is called as enzymolysis and the enzymes are called as proteolytic enzymes. The enzyme peptidases attack on peptide linkage of polypeptides and produce amino acids. Twenty standard amino acids which make up protein have twenty different pathways for its degradation. The twenty catabolic pathways converge to form only five products all of which enter the citric acid cycle. All or part of the carbon skeletons of ten of the amino acid are finally broken down to acetyl CoA, five amino acids are converted to ά- ketoglutaric acid , four into succinyl CoA, two into fumarate and two into oxaloacetate. The strategy of amino acid degradation is to form major metabolic intermediates that can be converted into glucose or to be oxidized by the citric acid cycle.

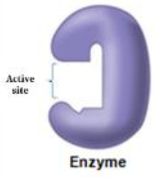
Definition

1. **Glycolysis:** Glycolysis is the sequence of reactions that convert glucose into pyruvate with the concomitant trapping of the energy as ATP.
2. **Citric acid cycle:** The citric acid cycle (CAC) – also known as the tricarboxylic acid (TCA) cycle or the Krebs cycle – is a series of chemical reactions used by all aerobic organisms to release stored energy through the oxidation of acetyl-CoA derived from carbohydrates, fats, and proteins into carbon dioxide and chemical energy in the form of adenosine triphosphate (ATP)
3. **Hexose Monophosphate Shunt**: A sequence of metabolic reactions by which NADPH is synthesized, together with ribose phosphate, part of the synthesis of nucleic acids
4. **Gluconeogenesis:** Gluconeogenesis (GNG) is a metabolic pathway that results in the generation of glucose from certain non-carbohydrate carbon substrates
5. **Glycogenesis:** Glycogenesis is the process of glycogen synthesis, in which glucose molecules are added to chains of glycogen for storage.
6. G**lyoxylate cycle.** The glyoxylate cycle, a variation of the tricarboxylic acid cycle, is an anabolic pathway occurring in plants, bacteria, protists, and fungi. The glyoxylate cycle centers on the conversion of acetyl-CoA to succinate for the synthesis of carbohydrates
7. **Electron transport Chain**: the stepwise transfer of electrons from one carrier molecule, as a flavoprotein or a cytochrome, to another along the respiratory chain and ultimately to oxygen during the aerobic production of ATP.
8. **Oxidative phosphorylation:** The synthesis of ATP by phosphorylation of ADP for which energy is obtained by electron transport and which takes place in the mitochondria during aerobic respiration
9. **Alpha oxidation: A**lpha-Oxidation is a process in which fatty acids are shortened by one carbon atom. The alpha-oxidation sequence of 3-methyl-branched fatty acids starts with an activation to the corresponding CoA-ester. Subsequently this acyl-CoA-ester undergoes a 2-hydroxylation by the peroxisomal phytanoyl-CoA hydroxylase (PAHX).
10. **β-Oxidation of fatty acids:** Beta-oxidation is the catabolic process by which fatty acid molecules are broken down in the cytosol in prokaryotes and in the mitochondria in eukaryotes to generate acetyl-CoA, which enters the citric acid cycle, and NADH and FADH2, which are co-enzymes used in the electron transport
11. **Transamination:** a chemical reaction that transfers an amino group to a keto acid to form new amino acids.
12. **Oxidative deamination:** An amino acid is converted into the corresponding keto acid by the removal of the amine functional group as ammonia and the amine functional group is replaced by the ketone group**.**
13. **Non-oxidative deamination:** Amino acids such as serine and histidine are deaminated non-oxidatively

ENZYMES

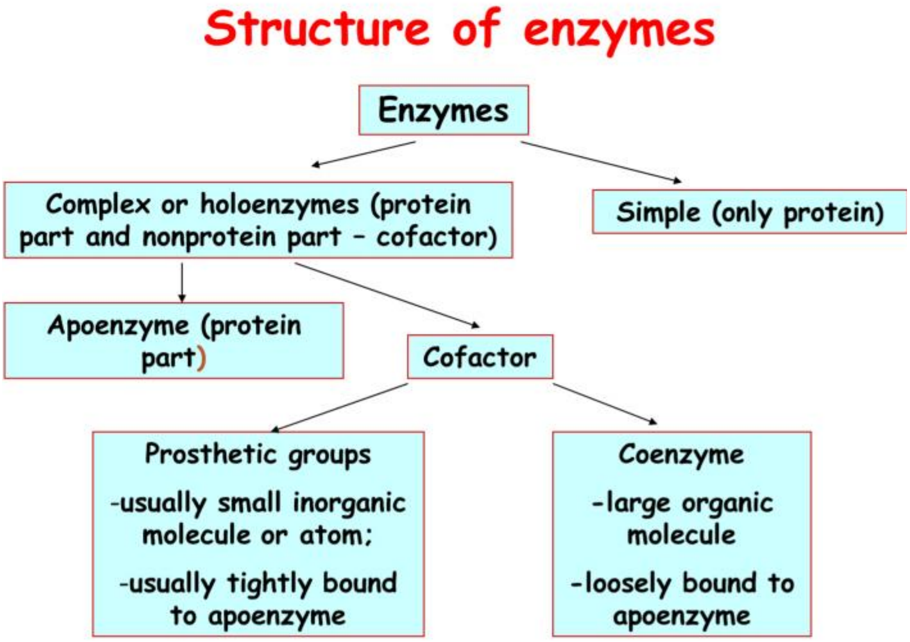
What is an enzyme?

Globular protein which functions as a biological catalyst, speeding up reaction rate by lowering activation energy without being affected by the reaction it catalyse.



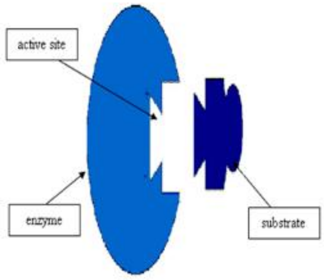
Characteristics

1. Enzymes speed up the reaction by lowering the activation energy of the reaction.
2. Their presence does not effect the nature and properties of end product.
3. They are highly specific in their action that is each enzyme can catalyze one kind of substrate.
4. Small amount of enzymes can accelerate chemical reactions.
5. Enzymes are sensitive to change in pH, temperature and substrate concentration.
6. Turnover number is defined as the number of substrate molecules transformed per minute by one enzyme molecule.
7. Catalase turnover number = 6 x10^6/min

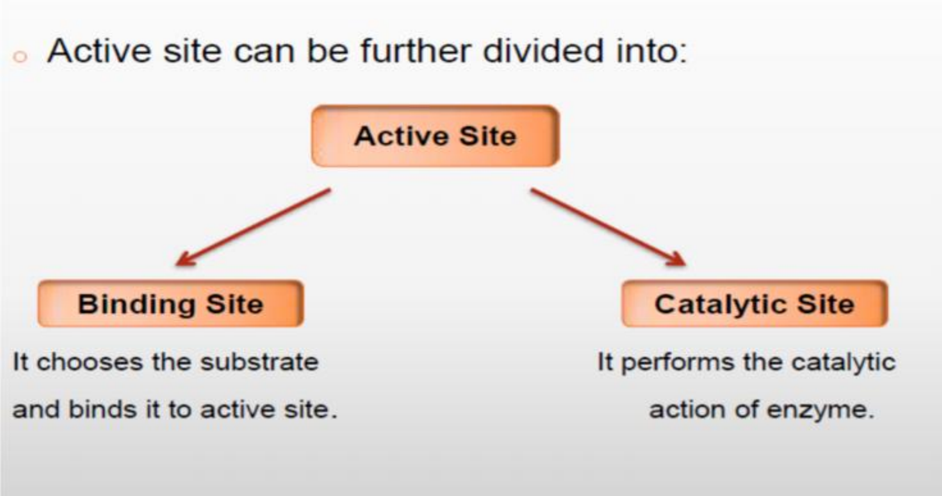


Active site

The active site of an enzyme is the region that binds substrates, co-factors and prosthetic groups and contains residue that helps to hold the substrate. Active sites generally occupy less than 5% of the total surface area of enzyme. Active site has a specific shape due to tertiary structure of protein. A change in the shape of protein affects the shape of active site and function of the enzyme.



Enzymes are highly specific for the type of the reaction they catalyze and for their substrate



- Active site can be further divided into:

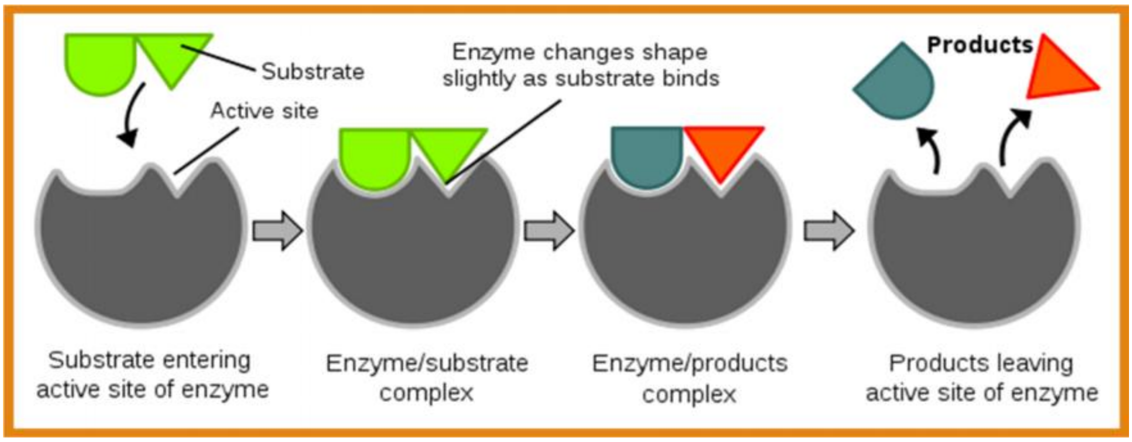
Active Site

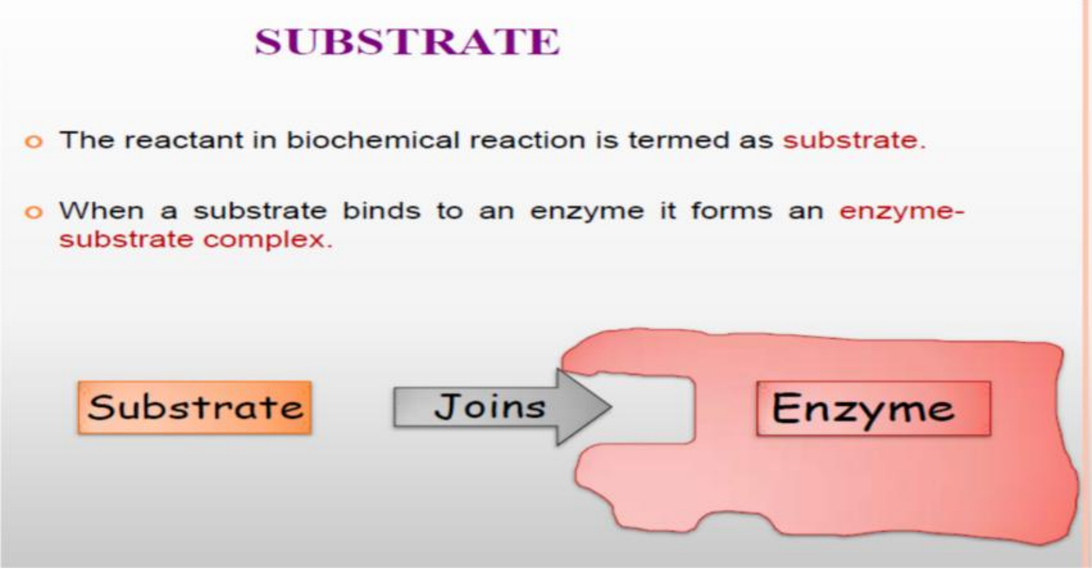
Binding Site

It chooses the substrate and binds it to active site.

Catalytic Site

It performs the catalytic action of enzyme.





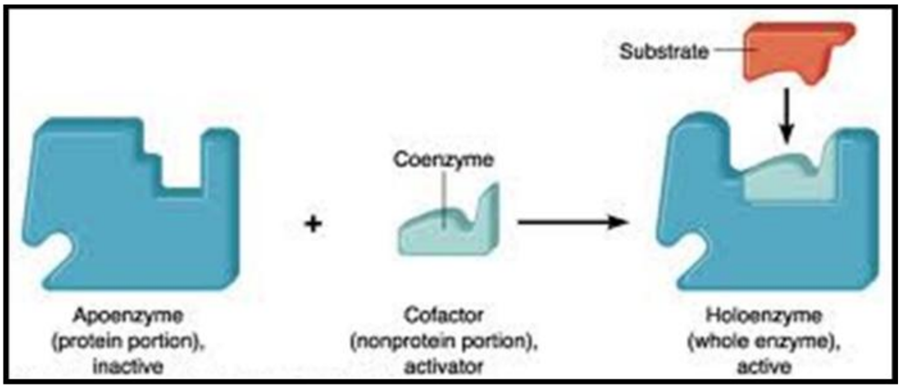
SUBSTRATE

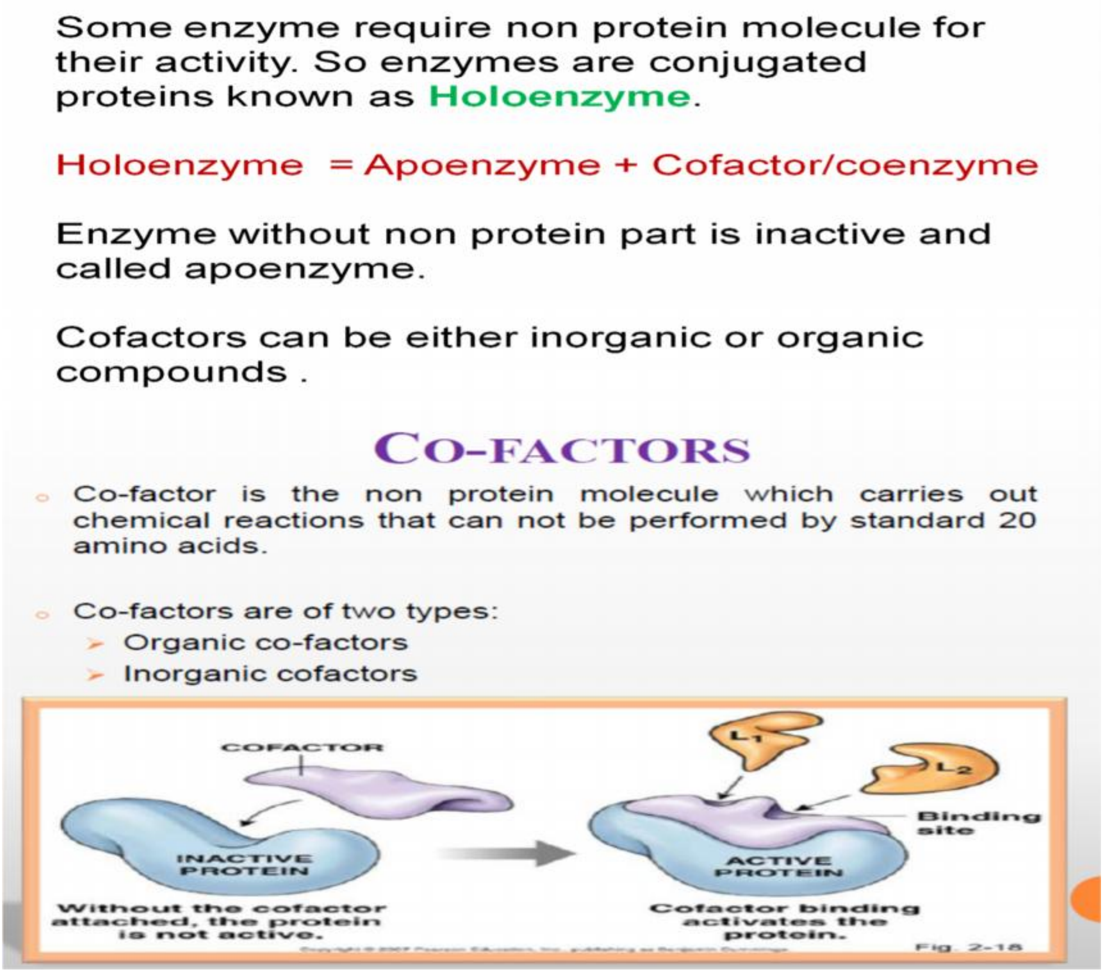
- The reactant in biochemical reaction is termed as substrate.
- When a substrate binds to an enzyme it forms an enzyme-substrate complex.

Enzymes are highly specific with varying degrees of specifity.

- Absolute specifity – they act on one substrate and only on that substrate.
- Stereospecifity – such enzymes that can detect the difference between optical isomers (mirror images) and select only one of such isomers.
- Reaction specifity – enzymes that catalyze certain types of reactions.
- Group specifity – enzymes that catalyzes a group of substances that contain specific compounds

APOENZYME and HOLOENZYME





Some enzyme require non protein molecule for their activity. So enzymes are conjugated proteins known as Holoenzyme.

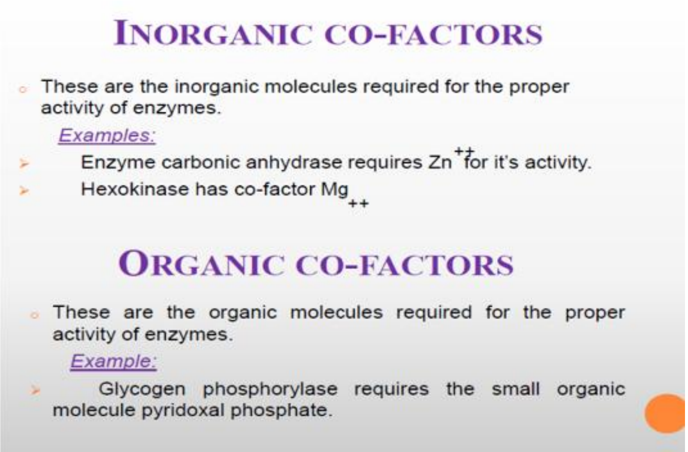
Holoenzyme = Apoenzyme + Cofactor/coenzyme

Enzyme without non protein part is inactive and called apoenzyme.

Cofactors can be either inorganic or organic compounds .

CO-FACTORS

- Co-factor is the non protein molecule which carries out chemical reactions that can not be performed by standard 20 amino acids.
- Co-factors are of two types:
  - Organic co-factors
  - Inorganic cofactors



INORGANIC CO-FACTORS

- These are the inorganic molecules required for the proper activity of enzymes.

Examples:

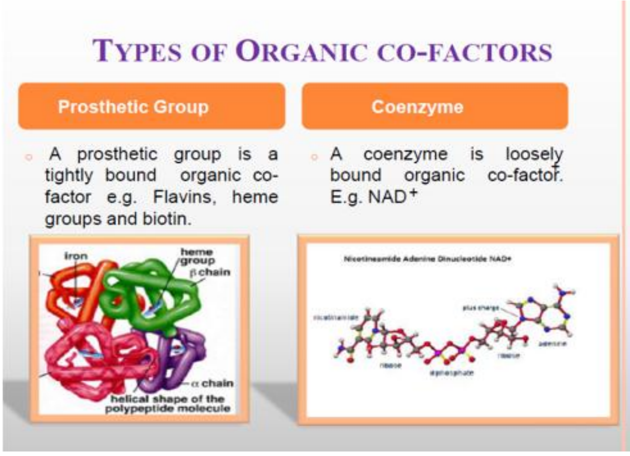
- Enzyme carbonic anhydrase requires Zn^{++} for it's activity.
- Hexokinase has co-factor Mg_{++}

ORGANIC CO-FACTORS

- These are the organic molecules required for the proper activity of enzymes.

Example:

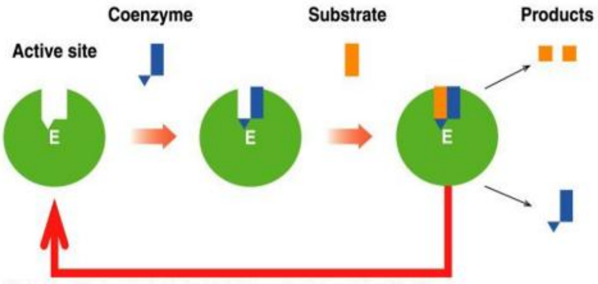
- Glycogen phosphorylase requires the small organic molecule pyridoxal phosphate.



TYPES OF ORGANIC CO-FACTORS

Prosthetic Group	Coenzyme
A prosthetic group is a tightly bound organic co-factor e.g. Flavins, heme groups and biotin.	A coenzyme is loosely bound organic co-factor. E.g. NAD^+

Coenzymes are small organic molecules that are often required to prepare the active site for proper substrate binding and/or participate in catalysis. Because they are not destroyed during the reaction, coenzymes are only required in small quantities



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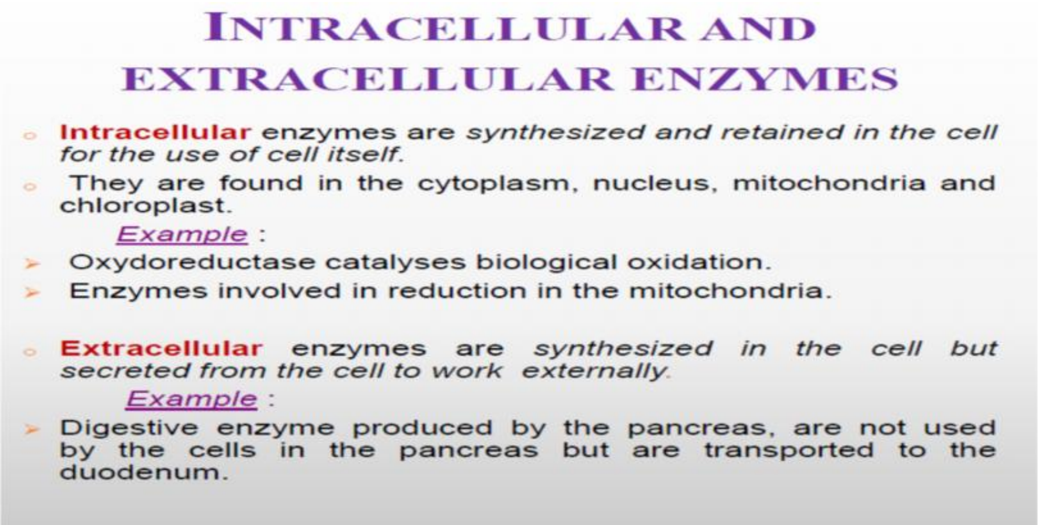


INTRACELLULAR AND EXTRACELLULAR ENZYMES

- **Intracellular** enzymes are *synthesized and retained in the cell for the use of cell itself.*
- They are found in the cytoplasm, nucleus, mitochondria and chloroplast.

Example :

- Oxydoreductase catalyses biological oxidation.
- Enzymes involved in reduction in the mitochondria.

- **Extracellular** enzymes are *synthesized in the cell but secreted from the cell to work externally.*

Example :

- Digestive enzyme produced by the pancreas, are not used by the cells in the pancreas but are transported to the duodenum.

Classification and Nomenclature of Enzymes

Naming Enzymes

- The name of an enzyme in many cases end in –ase
  For example, sucrase catalyzes the hydrolysis of sucrose
- The name describes the function of the enzyme
  For example, oxidases catalyze oxidation reactions
- Sometimes common names are used, particularly for the digestion enzymes such as pepsin and trypsin
- Some names describe both the substrate and the function
  For example, alcohol dehydrogenase oxides ethanol

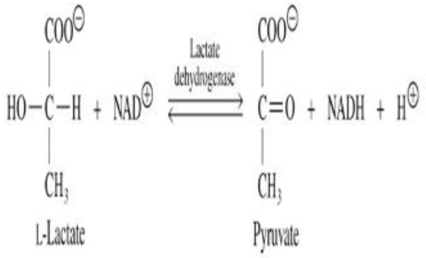
Enzymes are classified into six functional Classes (EC number Classification) by the International Union of Biochemists (I.U.B.). on the Basis of the Types of Reactions That They Catalyze

EC 1. Oxidoreductases
EC 2. Transferases
EC 3. Hydrolases
EC 4. Lyases
EC 5. Isomerases
EC 6. Ligases

EC 1. Oxidoreductases

Biochemical Activity:

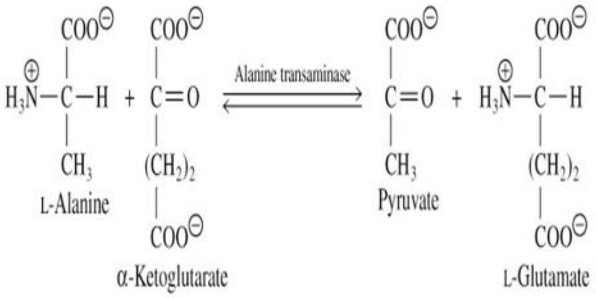
Catalyse Oxidation/Reduction Reactions Act on many chemical groupings to add or remove hydrogen atoms. Examples: Lactate dehydrogenase. Glucose Oxidase. Peroxidase. Catalase.
Phenylalanine hydroxylase.



EC 2. Transferases

Biochemical Activity:

Transfer a functional groups (e.g. methyl or phosphate) between donor and acceptor molecules. Examples: Transaminases (ALT & AST). Phosphotransferases (Kinases). Transmethylases.
Trans peptidases. Transacylase.
Catalyze group transfer reactions

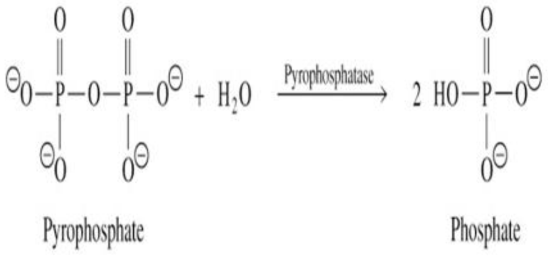


(5.2)

EC 3.Hydrolases

Biochemical Activity:

Catalyse the hydrolysis of various bonds Add water across a bond.
Examples: Protein hydrolysing enzymes (Peptidases).Carbohydrases (Amylase, Maltase, Lactase). Lipid hydrolysing enzymes (Lipase).Deaminases. Phosphatases.



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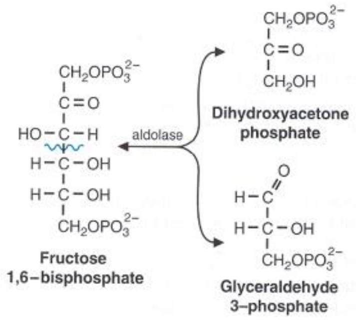
EC 4. Lyases

Biochemical Activity:

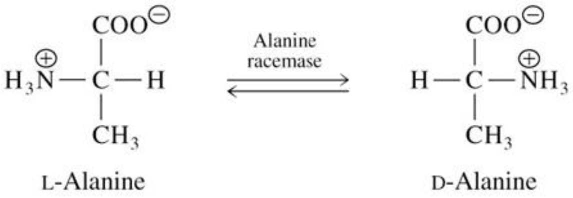
Cleave various bonds by means other than hydrolysis and oxidation .Add Water, Ammonia or Carbon dioxide across double bonds, or remove these elements to produce double bonds. Examples: Fumarase. Carbonic anhydrase**.**



EC 5.Isomerases

Biochemical Activity:

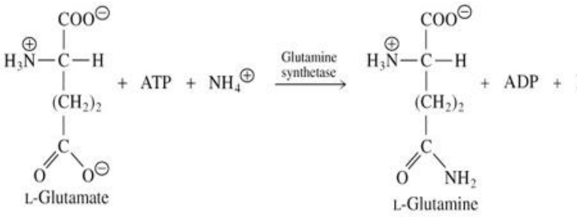
Catalyse isomerization changes within a single molecule. Carry out many kinds of isomerization: L to D isomerizations .Mutase reactions (Shifts of chemical groups). Examples: Isomerase. Mutase



EC 6. Ligases

Biochemical Activity:

Join two molecules with covalent bonds Catalyse reactions in which two chemical groups are joined (or ligated) with the use of energy from ATP. Examples: Acetyl~CoA Carboxylase. Glutamine synthetase



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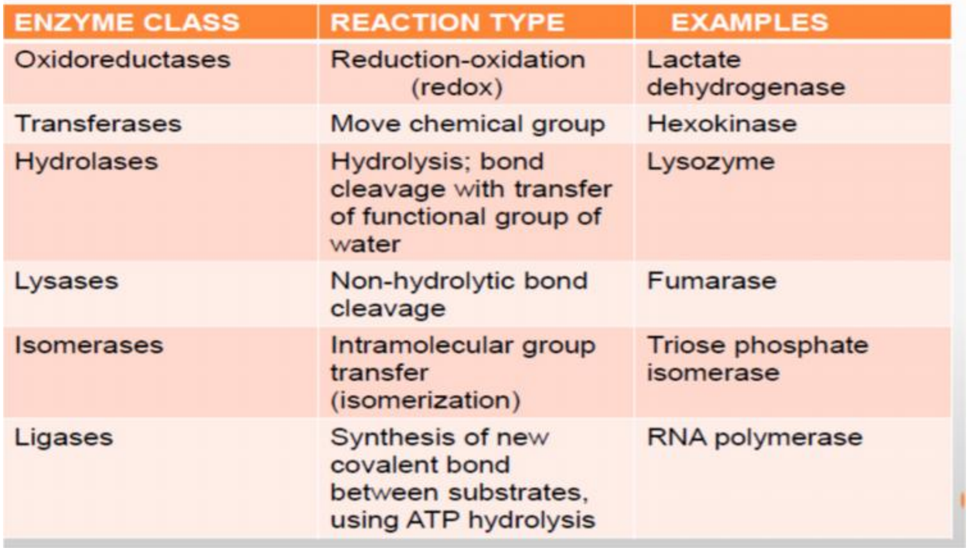
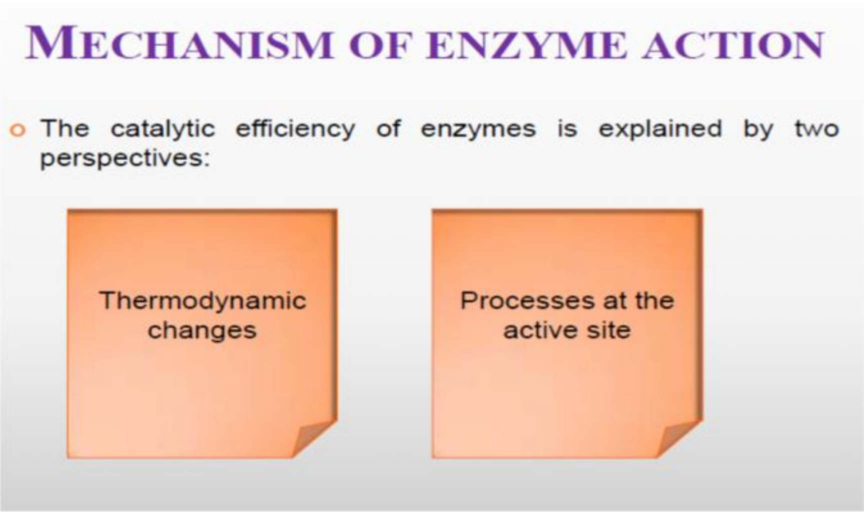


ENZYME CLASS	REACTION TYPE	EXAMPLES
Oxidoreductases	Reduction-oxidation (redox)	Lactate dehydrogenase
Transferases	Move chemical group	Hexokinase
Hydrolases	Hydrolysis; bond cleavage with transfer of functional group of water	Lysozyme
Lysases	Non-hydrolytic bond cleavage	Fumarase
Isomerases	Intramolecular group transfer (isomerization)	Triose phosphate isomerase
Ligases	Synthesis of new covalent bond between substrates, using ATP hydrolysis	RNA polymerase

Mechanism of Action of Enzymes



MECHANISM OF ENZYME ACTION

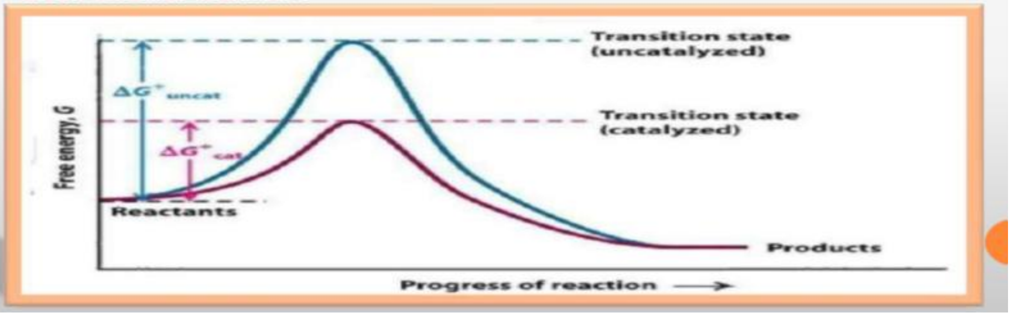
- The catalytic efficiency of enzymes is explained by two perspectives:

Thermodynamic changes

Processes at the active site

THERMODYNAMIC CHANGES

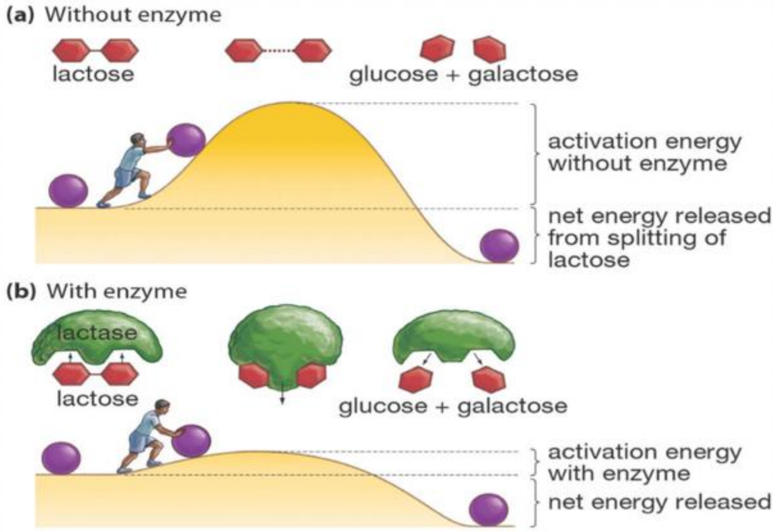
- All chemical reactions have energy barriers between reactants and products.
- The difference in transitional state and substrate is called *activational barrier*.

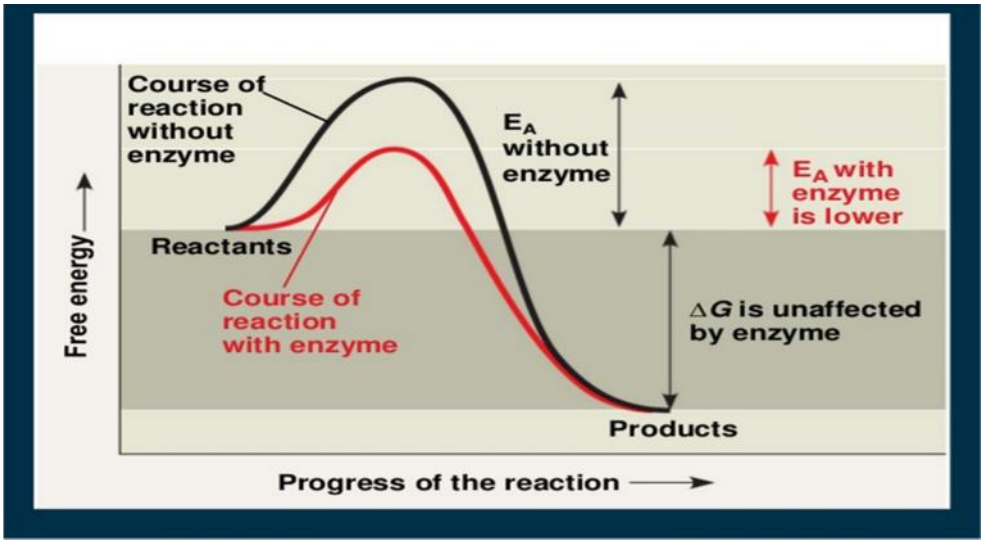


THERMO-DYNAMIC CHANGES OVERVIEW

- Enzymes increase reaction rates by decreasing the Activation energy

Enzymes Lower a Reaction's Activation Energy





The enzymatic reactions takes place by binding of the substrate with the active site of the enzyme molecule by several weak bonds.

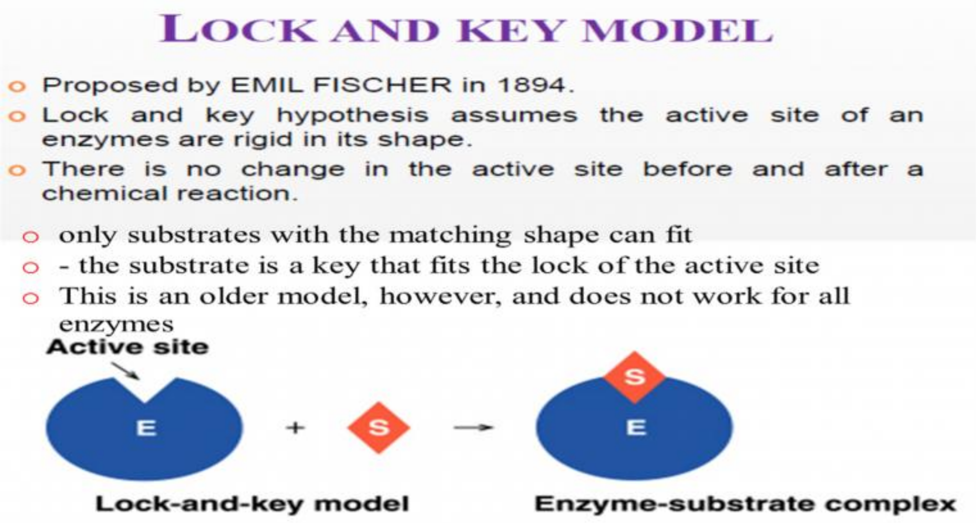
E + S ‹--------› ES --------› E + P

Formation of ES complex is the first step in the enzyme catalyzed reaction then ES complex is subsequently converted to product and free enzyme.

Enzyme-Substrate Interactions:

Formation of Enzyme substrate complex by:

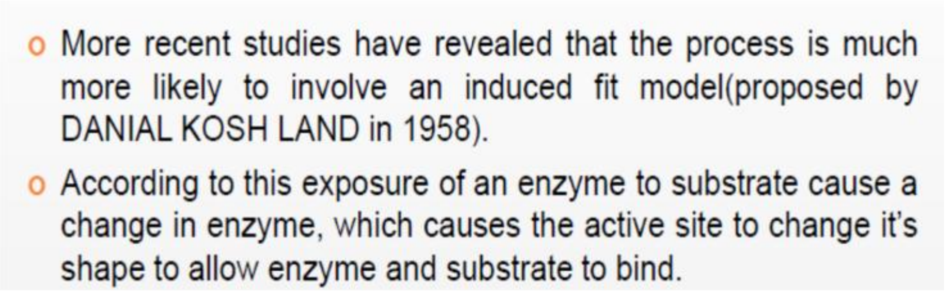
- Lock-and-Key Model
- Induced Fit Model



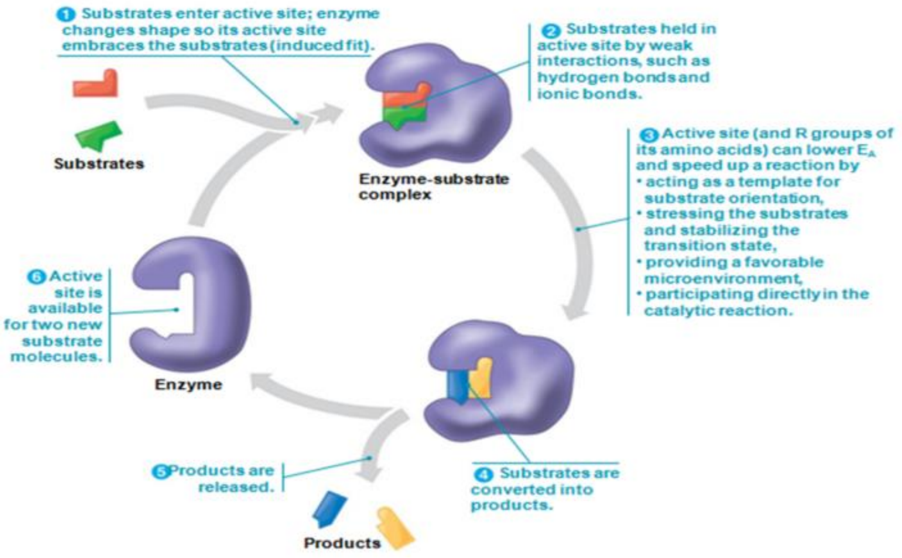
LOCK AND KEY MODEL

- Proposed by EMIL FISCHER in 1894.
- Lock and key hypothesis assumes the active site of an enzymes are rigid in its shape.
- There is no change in the active site before and after a chemical reaction.
- only substrates with the matching shape can fit
- - the substrate is a key that fits the lock of the active site
- This is an older model, however, and does not work for all enzymes

Induced Fit Model



- More recent studies have revealed that the process is much more likely to involve an induced fit model(proposed by DANIAL KOSH LAND in 1958).
- According to this exposure of an enzyme to substrate cause a change in enzyme, which causes the active site to change it's shape to allow enzyme and substrate to bind.



Enzyme Catalyzed Reactions

When a substrate (S) fits properly in an active site, an enzyme-substrate (ES) complex is formed:

$$E + S \leftrightarrows ES$$

Within the active site of the ES complex, the reaction occurs to convert substrate to product (P):

$$ES \rightarrow E + P$$

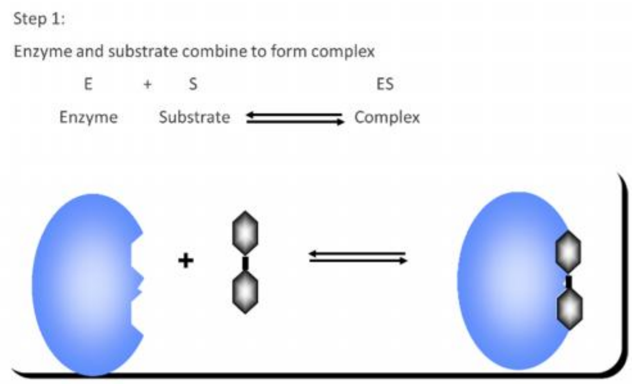
The products are then released, allowing another substrate molecule to bind the enzyme

- This cycle can be repeated millions (or even more) times per minute

The overall reaction for the conversion of substrate to product can be written as follows:

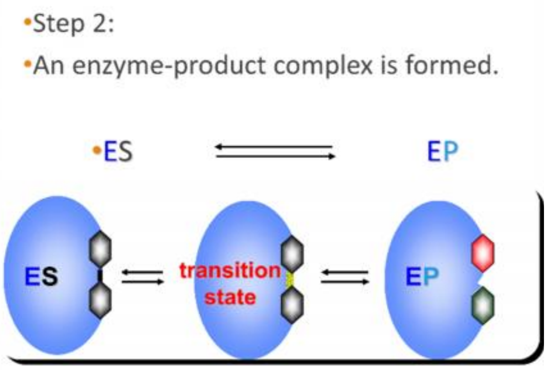
$$E + S \leftrightarrows ES \rightarrow E + P$$

Enzyme-substrate complex



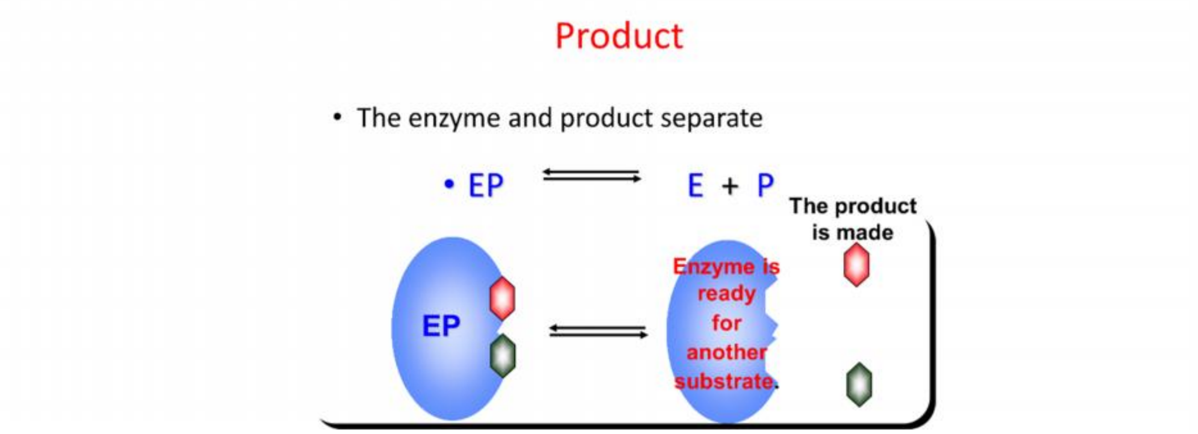
Step 1:

Enzyme and substrate combine to form complex



•Step 2:

•An enzyme-product complex is formed.



Product

• The enzyme and product separate

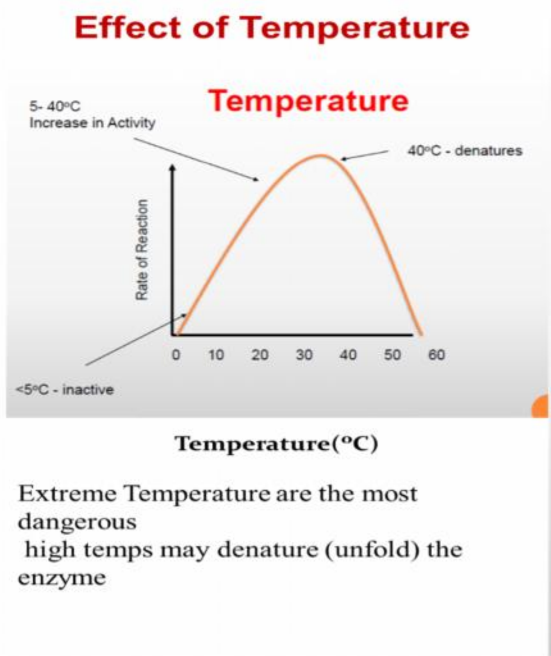
What Affects Enzyme Activity?

Three factors:

1. Environmental Conditions
2. Cofactors and Coenzymes
3. Enzyme Inhibitors

.1. Environmental Conditions

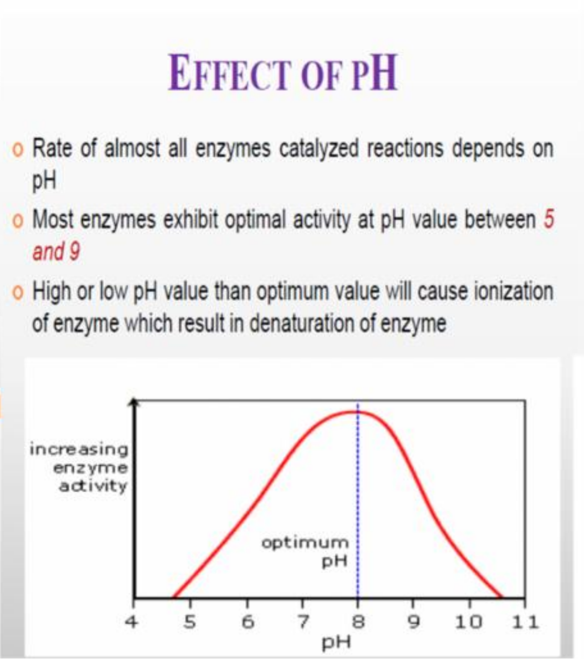
- Temperature, Hydrogen ion concentration (pH), Substrate concentration
- Enzyme concentration, Products of the reaction, Presence of activator/inhibitor,
- Allosteric effects. Time



Effect of Temperature

Temperature(°C)

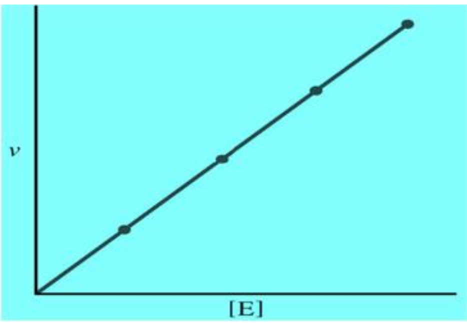
Extreme Temperature are the most dangerous
high temps may denature (unfold) the enzyme



EFFECT OF PH

- Rate of almost all enzymes catalyzed reactions depends on pH
- Most enzymes exhibit optimal activity at pH value between *5 and 9*
- High or low pH value than optimum value will cause ionization of enzyme which result in denaturation of enzyme

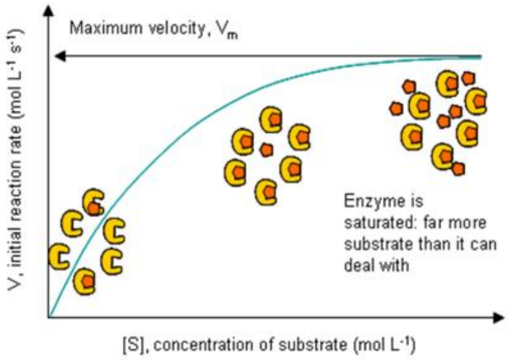
Rate of the reaction or velocity is directly proportional to the Enzyme Concentration when sufficient substrate is present

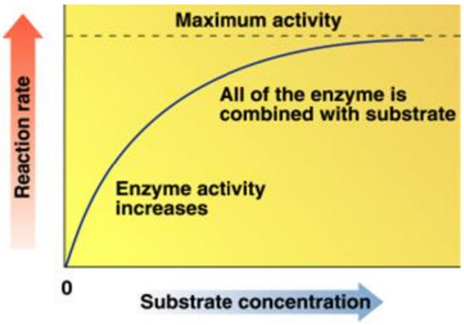


Accumulation of Product in a reaction causes inhibition of enzyme activity.

Substrate Concentration and Reaction Rate

The rate of reaction increases as substrate concentration increases (at constant enzyme concentration). Maximum activity occurs when the enzyme is saturated (when all enzymes are binding substrate**)**





Enzyme Kinetics

- Study of reaction rate and how they changes in response to change in experimental parameter is known as **kinetics**.

Michaelis- Menten Kinetics

The model involves one substrate molecule,

$$E + S \underset{k_{-1}}{\overset{k_1}{\longleftrightarrow}} ES \xrightarrow{k_2} E + P$$

Where

- S is the substrate
- E is the enzyme
- K_1, k_{-1} and k_2 are the rate constants

- The mathematical equation that defines the quantitative relationship between the rate of an enzyme reaction and the substrate concentration is the *Michaelis-Menten equation*:

$$V_0 = \frac{V_{max}\,[S]}{K_m + [S]}$$

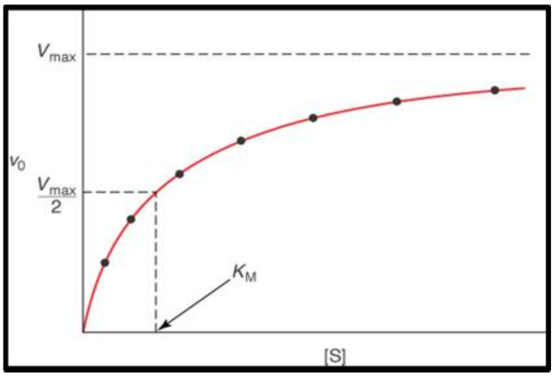
V_0 is the observed velocity at the given [S]

K_m is the Michaelis-Menten constant

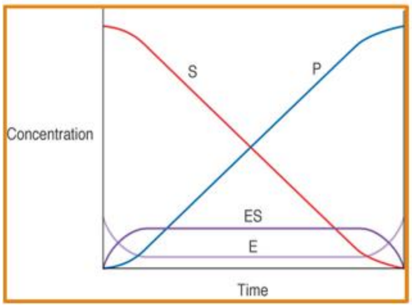
$K_m = (K_{-1} + K_2) / K_1$

V_{max} is the maximum velocity at saturating [S] conc.

Graphical determination of K_M



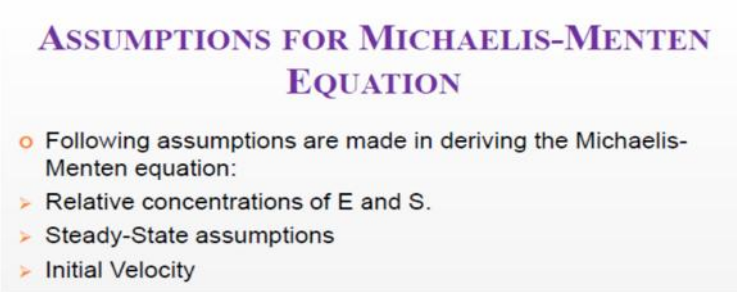
Changes in concentration for a simple enzyme-catalyzed reaction



ES remains constant while S is converted to P.Here all the substrate is converted to P

What is Km and Vmax?

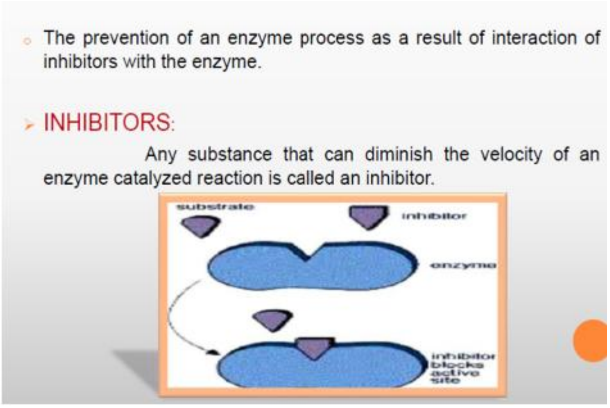
Km is the concentration of substrate which permits the enzyme to achieve half Vmax. An enzyme with a high Km has a low affinity for its substrate, and requires a greater concentration of substrate to achieve Vmax."



ASSUMPTIONS FOR MICHAELIS-MENTEN EQUATION

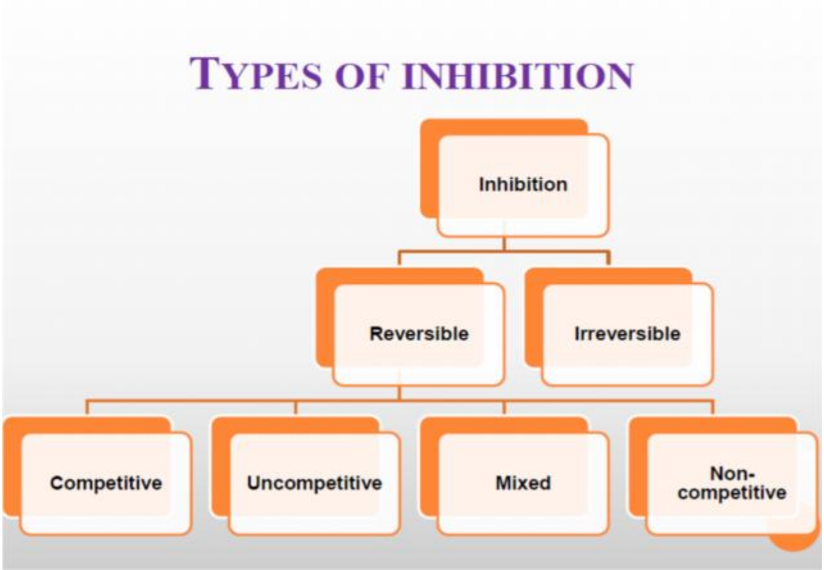
- Following assumptions are made in deriving the Michaelis-Menten equation:
  - Relative concentrations of E and S.
  - Steady-State assumptions
  - Initial Velocity

Enzyme Inhibition



- The prevention of an enzyme process as a result of interaction of inhibitors with the enzyme.
- INHIBITORS:

  Any substance that can diminish the velocity of an enzyme catalyzed reaction is called an inhibitor.



TYPES OF INHIBITION

Inhibition

Reversible

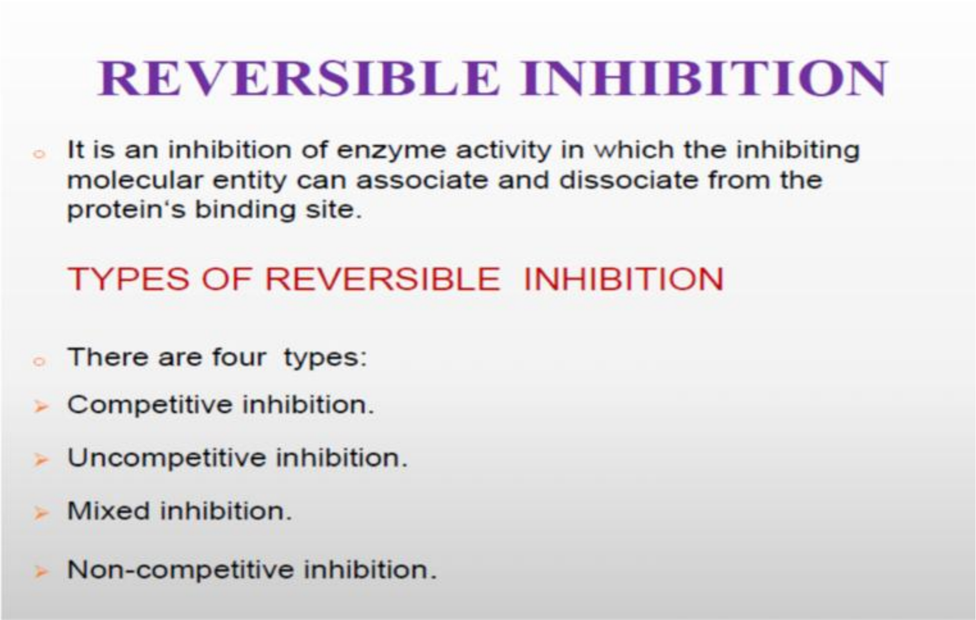
Irreversible

Competitive

Uncompetitive

Mixed

Non-competitive

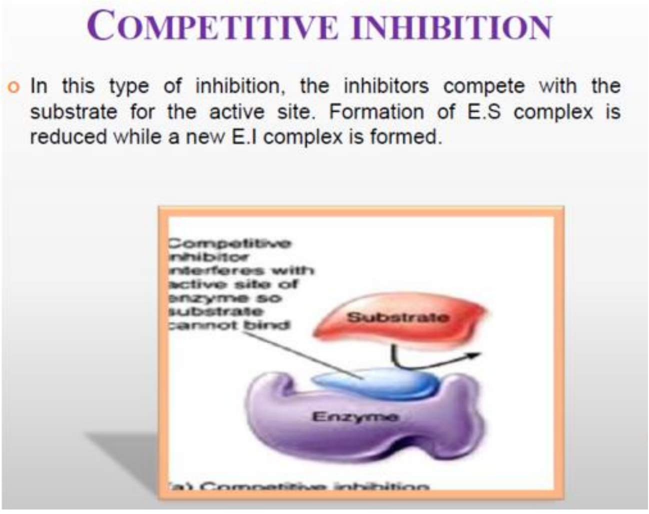


REVERSIBLE INHIBITION

- It is an inhibition of enzyme activity in which the inhibiting molecular entity can associate and dissociate from the protein's binding site.

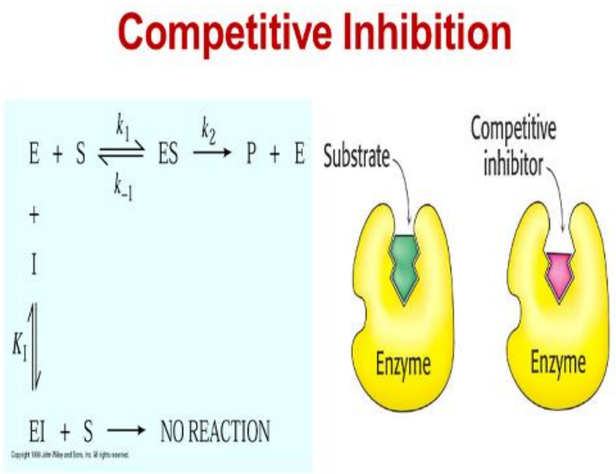
TYPES OF REVERSIBLE INHIBITION

- There are four types:
- Competitive inhibition.
- Uncompetitive inhibition.
- Mixed inhibition.
- Non-competitive inhibition.



COMPETITIVE INHIBITION

- In this type of inhibition, the inhibitors compete with the substrate for the active site. Formation of E.S complex is reduced while a new E.I complex is formed.



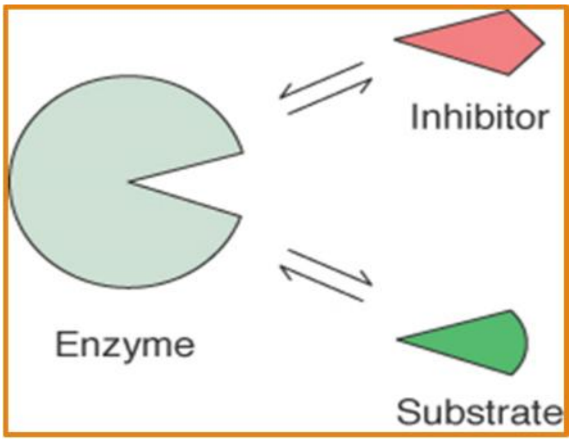
Competitive Inhibition

$$E + S \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} ES \xrightarrow{k_2} P + E$$

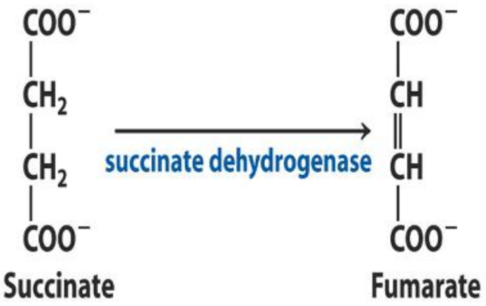
$$+ \; I \; \underset{K_I}{\rightleftharpoons}$$

$$EI + S \longrightarrow \text{NO REACTION}$$

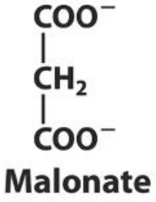
Competitive enzyme inhibition



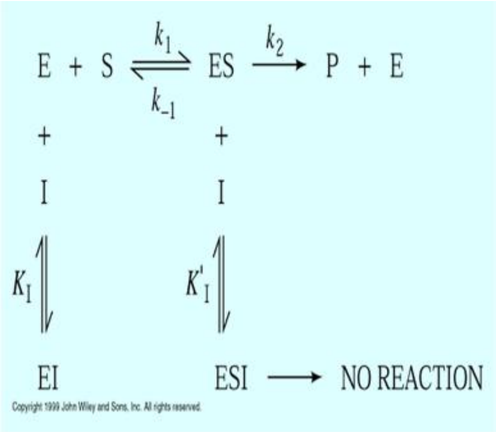
Succinate dehydrogenase

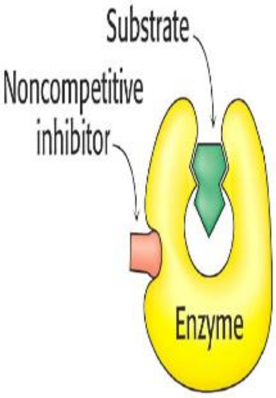


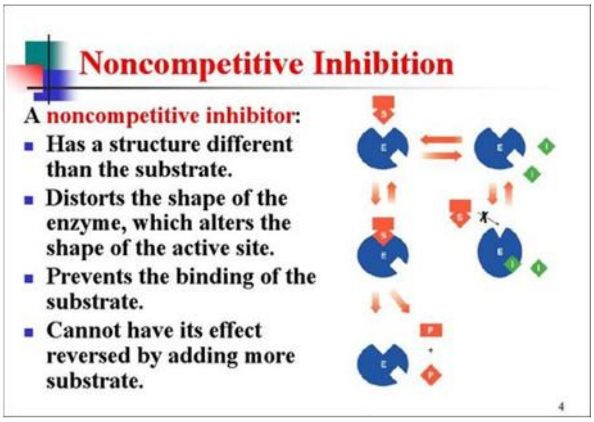
Malonate is a competitive
Inhibitor



Non-Competitive Inhibition







Noncompetitive Inhibition

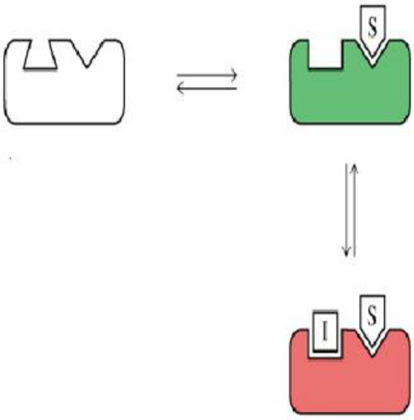
A **noncompetitive inhibitor**:

- Has a structure different than the substrate.
- Distorts the shape of the enzyme, which alters the shape of the active site.
- Prevents the binding of the substrate.
- Cannot have its effect reversed by adding more substrate.

UNCOMPETITIVE INHIBITION

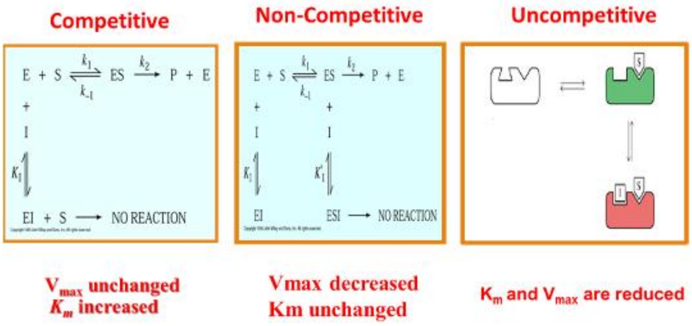
- In this type of inhibition, inhibitor does not compete with the substrate for the active site of enzyme instead it binds to another site known as *allosteric* site.

Un-competitive Inhibition

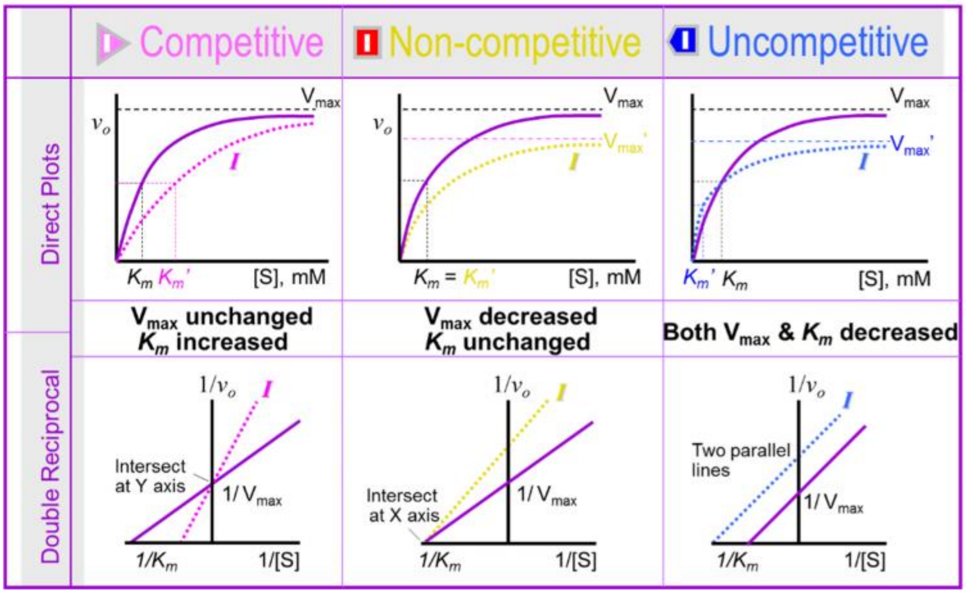


Un-competitive Inhibition

Binds only to the enzyme-substrate complex. Does not have the capacity to bind to the free enzyme. Not overcome by increasing substrate concentration. Both the Km and Vmax are reduced. Only binds when the ES complex is formed ii) reduces the amount of ES available, and thus less [S] to create the half of ES --> apparent lower Km iii) reduced the amount of available ES that aren't bound to the ESI, so at the high [S] concentrations, we have Vmax also lowering



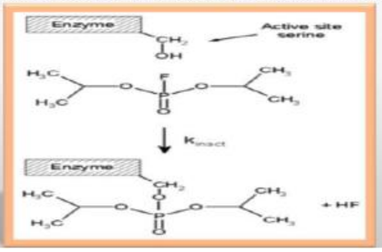
Enzyme Inhibition (Plots)



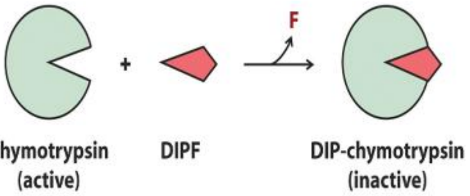
	Competitive	Non-competitive	Uncompetitive
	V_{max} unchanged **K_m increased**	**V_{max} decreased** **K_m unchanged**	**Both V_{max} & K_m decreased**

IRREVERSIBLE INHIBITION

- This type of inhibition involves the *covalent attachment* of the inhibitor to the enzyme.
- The *catalytic activity* of enzyme is completely lost.
- It can only be restored only by synthesizing molecules.



Irreversible inhibition



Diisopropylphosphofluoridate (DIPF) is an irreversible inhibitor of the enzyme chymotrypsin

ACTIVATION

- Activation is defined as the conversion of an inactive form of an enzyme to active form which processes the metabolic activity.

TYPES OF ACTIVATION

- Activation by co-factors.
- Conversion of an enzyme precursor.

ACTIVATION BY CO FACTORS

- Many enzymes are activated by co-factors.

Examples:

- *DNA polymerase* is a holoenzyme that catalyzes the polymerization of de -oxyribonucleotide into a DNA strand. It uses Mg- ion for catalytic activity.
- *Horse liver dehydrogenase* uses Zn- ion for it's activation.

Isoenzyme

Isoenzymes or isozymes are multiple forms of same enzyme that catalyze the same chemical reaction. These enzymes usually display different kinetic parameters (e.g. different KM values), or different regulatory properties. The existence of isozymes permits the fine-tuning of metabolism to meet the particular needs of a given tissue or developmental stage. Isozymes are usually the result of gene duplication, but can also arise from polyploidisation or nucleic acid hybridization. An example of an isozyme is glucokinase, a variant of hexokinase which is not inhibited by glucose 6-phosphate. Isozymes (and allozymes) are variants of the same enzyme.

Zymogen

Zymogens (proenzymes) are inactive forms of enzyme. They are activated by removal of peptide sections. Digestive enzymes are produced as zymogens, and are then activated when needed. For example, proinsulin is converted to insulin by removing a 33-amino acid peptide chain. The digestive enzymes must be stored as zymogens because otherwise they would damage the pancreas

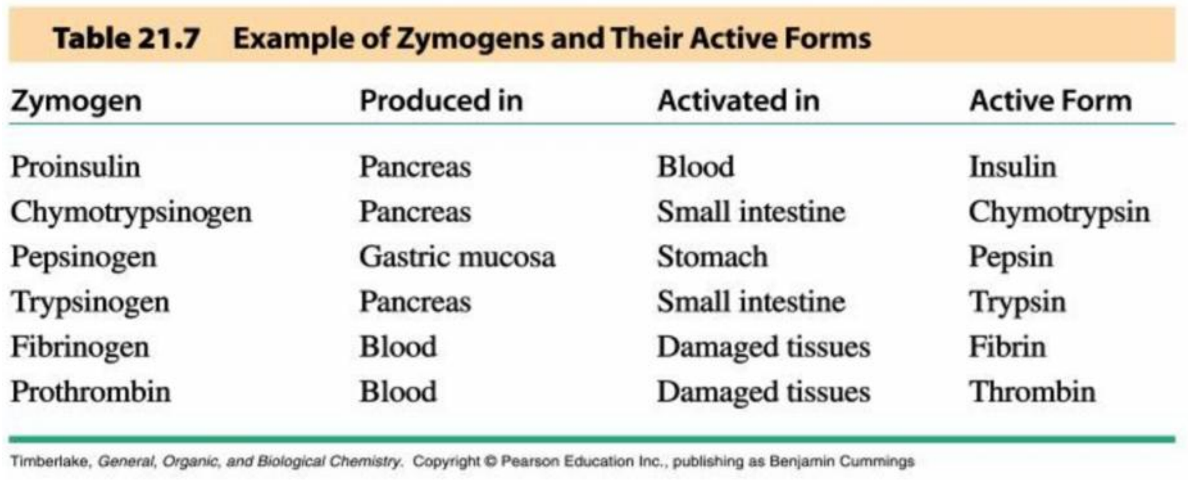


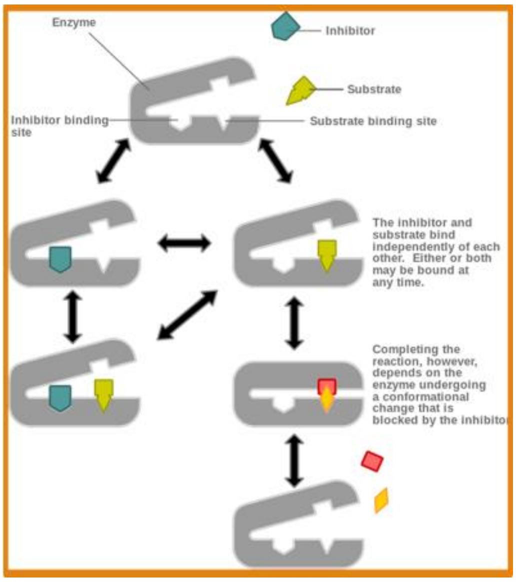
Table 21.7 Example of Zymogens and Their Active Forms

Zymogen	Produced in	Activated in	Active Form
Proinsulin	Pancreas	Blood	Insulin
Chymotrypsinogen	Pancreas	Small intestine	Chymotrypsin
Pepsinogen	Gastric mucosa	Stomach	Pepsin
Trypsinogen	Pancreas	Small intestine	Trypsin
Fibrinogen	Blood	Damaged tissues	Fibrin
Prothrombin	Blood	Damaged tissues	Thrombin

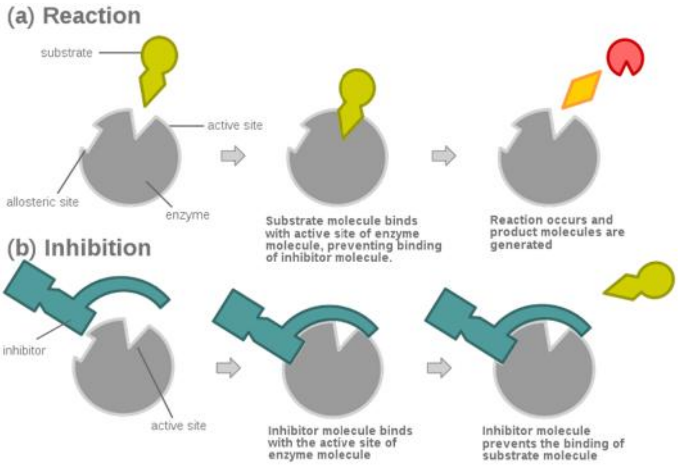
Timberlake, *General, Organic, and Biological Chemistry*. Copyright © Pearson Education Inc., publishing as Benjamin Cummings

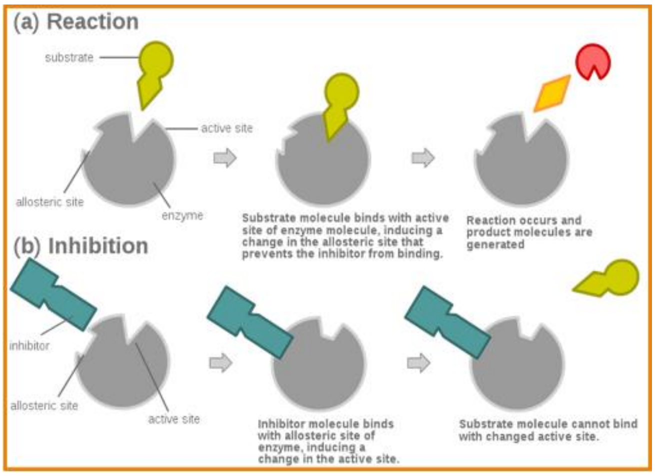
Allosteric Enzyme

Some of the enzymes possess additional sites, known as allosteric sites (Greeek; allo-other) besides the active site. Such enzymes are known as allosteric enzyme. The allosteric sites are unique places on the enzyme molecules; Allosteric enzyme have one or more allosteric sites Allosteric sites are binding sites distinct from an enzyme active site or substrate binding site Molecule that bind to allosteric sites are called effector or modulator. Effector may be positive or negative, this effector regulate the enzyme activity. The enzyme activity is increased when a positive allosteric effector binds at the allosteric site known as activator site. On the other hand, negative allosteric effector bind at the allosteric site called inhibitor site



and inhibit the enzyme activity. Binding to allosteric sites alter the activity of the enzyme. An allosteric inhibitor binds to the enzyme, inducing it to assume an inactive form. All non-competitive inhibition is allosteric inhibition, but not all allosteric inhibition is non-competitive inhibition because certain forms of allosteric inhibition can prevent the substrate from binding to the active site.





FILL UP THE BLANKS

1. _______ is one of the enzyme involved in carbohydrate metabolism
2. Enzymes are_____-
3. NADPH is an universal ______ source
4. Enzymes which break the linkage between C_C bond and C-N bond are_______
5. An example of Co enzyme is ______
6. ______ enzyme splits peptides of proteins
7. The zymogens are inactive enzymes precursor is converted into an active enzyme by__ compounds
8. The enzyme which catalyst the transfer of hydrogen from one molecule to another is called _____
9. The enzyme that hydrolyze sucrose is _____-
10. Enzymes are located in _______
11. Vitamin B1 and B6 form coenzymes with ____ and _______
12. Enzymes is a ____ which _______ the activation energy of a reaction
13. _______ is a metallo enzyme
14. The coenzymes of niacin are ____ and ______-
15. Enzymes which are present inside the living cells and act within is called as ______
16. The prosthetic group of complex proteins enzyme is called as ______
17. Apo enzyme and coenzyme put together is called as _______
18. The enzyme that catalyses hydrolysis reactions is known as ______
19. _____ enzymes hydrolyze lipids
20. _____ is an example of oxidation-reduction enzymes
21. An enzyme which catalyse L- alanine to D-alanine is ______

22. ____ is an example of coenzyme

23. Multiple forms of the enzyme which catalyze the same reaction is called as _____-

24. _____ is an example of simple protein enzyme

25. Catalase is an example of __ enzyme

26. Coenzymes accounts for about ____% of the entire enzymes molecule

27. In Apo enzyme, if the prosthetic group includes the metal ions, then it is called as ____

28. The catalytic power of an enzyme is measured by_______

29. Number of substrate molecules converted into products per unit time, when the enzyme is fully separated with substrate is known as _______

30. The specific region of the enzyme on which substrates binds is known as _____

31. _____ proposed enzyme substrate complex theory about mechanism of enzyme action

32. Emil Fischer proposed _____model regarding the action of enzymes substrate

33. The catalytic power of certain enzymes is reversibly altered by certain inorganic and organic molecules called ______

34. The inactive forms of enzymes are called ______

35. Compounds which convert the enzymes into inactive substances and adversely affect enzymatically catalyzed reaction is known as ______-

36. The inhibitors due to a compound which is totally different structure from the substrate of the enzymes is called as ______

37. Malonic acid which inhibit the activity of the enzyme sucrose dehydrogenase is referred to as _____-inhibitor

38. Lock and Key hypothesis on mechanism of enzyme action was proposed by______

39. ______ is a fat soluble vitamins

40. _____- is a water soluble vitamins

41. Irradiation of ergosterol results in the formation of ______

42. Evening blindness is caused by the deficiency of _______

43. In 1912 _____ coined the term vitamin

44. __________ is another anti beri- beri factor

45. ______ is a water soluble vitamin present in cow's milk

46 Isomerases involve in ______

47 Enzymes which add or remove phosphoric acid is called as_______

48 Carboxylase is _________

49 Enzyme is a _______

50 In arresting the activities of enzymes, the non- competitive enzyme inhibitor may combine ________ with enzyme

51 The protein part of enzyme is called as _________

52 Invertase is an enzyme which hydrolyze _________

53 Trypsin is a _______

54 The coenzyme required for transamination is _______

55 The coenzyme used in the biosynthesis of fatty acid is _________

56 Succinate dehydrogenase is a _______

57 An example of plant proteolytic enzyme is_______

58 Non-protein part in complex protein enzyme is known as______
59 β-carotene is precursors of _________
60 Ergosterol is a precursor of_______
61 An example of carotenoids is________
62 Enzymes fast up the velocity of a biochemical reaction by _____
63 Km values are not altered by _____________ type of inhibitor
64 The low value of Km indicates_________
65 Zinc is present in ___ enzymes
66 An allosteric modulator influences enzyme activity by______
67 The enzyme which catalyse R-CH2OH + O2 → R-CHO + H2O2 is _______
68 The enzyme that catalyse Glucose + ATP → Glucose -6-phosphate + ADP is ______
69 The typical saturation curve for an enzyme catalyzed reaction is sigmoidal. This indicates that the enzyme is _________
70 Michaelis menten equation is given by _______
71 Both Vmax and Km values are altered in _________ inhibition
72 A competitive inhibitor of an enzyme ____ Km and ______Vmax
73 A competitive inhibitor binds to ______
74 The inactive precursor of an active enzyme is called __________
75 The inhibition of succinate dehydrogenase by malonate is an example for _____
76 The uncompetitive inhibitor binds with_________
77 Isoenzymes are generally separated by ________method
78 Non-competitive inhibitor binds with________
79 Sigmoidal graph indicates _________
80 The coenzyme present in transaminase is ________
81 The low value of Km indicates _______
82 According to IUB enzymes are classified into _________ classes
83 The conversion of ATP to cAMP is catalyzed by______
84 Substrate binding site on the enzyme is called as___
85 Urease is a _______ type of enzyme
86 .J.B.Sumner isolated the _____ enzyme
87 Enzymes bringing about hydrolysis of esters and peptide bonds are_______
88 Most modern hypothesis about enzyme action is called _______
89 Fitting of substrate in active site is called as ________
90 In case of induced fit model _______ changes its shape slightly
91 Substrate binds to specific region of enzyme called ______
92 Enzymes do not change the _____
93 Catalytic region in which small portion of molecules are involved in catalysis is called__
94 Regulators of enzymatic reactions are classified as _______ and ____
95 Model proposed by Emil Fischer is known as________
96 Kind of molecules at which enzymes act are classified as ____
97 Apoenzyme plus co factor is known as ______-

98 Majority of enzymes are inactive at ______ temperature

99 Substance that accelerates the rate of a chemical reaction is called a(an)_______

100 Haemoglobins has _________ structure

101 The key regulated enzymes in glycolysis are ___, ______and __

Sl.No	Answer	Sl.No	Answer
67	Alcohol oxidase	100	Quaternary
68	Hexokinase	101	Hexokinase, Phosphofructokinase, Pyruvate kinase
69	Allosteric		
70	$V_0 = V_{max} \frac{[S]}{[S] + K_M}$		
71	Uncompetitive		
72	Increases , no effect		
73	Substrate		
74	Zymogen		
75	Competitive inhibitor		
76	Enzyme substrate complex		
77	electrophoresis and ion-exchange chromatograph		
78	Enzyme substrate complex		
79	Allosteric regulation		
80	pyridoxal-phosphate,		
81	High substrate affinity		
82	Six		
83	adenylyl cyclase		
84	Active		
85	Hydrolase		
86	Urease		
87	Hydrolase		
88	Induce fit		
89	Lock and key		
90	enzyme		
91	Active site		
92	Eq		
93	Active site		
94	Activator and inhibitor		
95	Deduction		
96	Substrate		
97	Holoenzyme		
98	Above 40°C		
99	Catalyst		

WRITE SHORT NOTES ON:

1. **Enzyme as a catalyst**

   Most of the chemical reactions in the cell take place in the presence of specific proteinaceous substances called as enzyme. It is referred to as biological catalyst. The enzymes do not alter the chemical equilibrium point of a reversible reaction but only the speed of the reaction is changed. It is produced from the living cells and being protainaceous in nature cannot last indefinitely in a reaction system as they often become damaged or inactivated by the reaction they catalyze. They must be replaced constantly by further synthesis in the body. Most individual enzymes are very specific in that they act either on a single or at most on some structurally related substrates. Since the enzymes are highly specific as to the reaction they catalyze, names of the enzymes are suitably changed for example hydrolyases (hydrolysis), isomerases (isomerization), oxidases (oxidation) , dehydrogenases(dehydrogenation), transminases (transamination) , transaldolases (translocation) etc.,

2. **Co-enzyme**

   Certain enzymes in addition to their protein structure have a non-protein group attached to them. The non-protein part of the enzyme is called as prosthetic group or co enzymes. The coenzymes are small molecular weight, organic, dialyzable, thermostable compounds required for the catalytic activity of one or a group of enzymes. Some of the common examples of coenzymes are NAD+, NADP, FAD, FMN CoA, Pyridoxals. Coenzymes are momo or dinucleotides. Most of the coenzymes are the members of vitamin B complex group. Based on the reaction in which they participate coenzymes are classified as hydrogen transferring group_. The coenzymes usually occur in living cells in low concentrations. The coenzyme is as essential a reactant as the substrate that is activated by the enzyme

   Substrate (A) + Coenzyme Enzyme (A) Substrate (A) derivative + Coenzyme derivatives
   Substrate (B) + coenzyme derivative Enzyme (B) Substrate (B) - derivative + Coenzyme

3. **Isoenzymes**

   Many enzymes occur is more than one molecular form in the same species, in the same tissue or even in the same cell. In such cases, the different forms of the enzyme catalyze the same reaction but since they possess different kinetic properties and different amino acid composition, they can be separated by appropriate techniques such as electrophoresis. Such multiple forms of the enzymes are called Isoenzymes. It is widespread in nature. Over a hundred enzymes are known to be of isoenzymic nature and consequently occur in two or more molecule form eg. Lactic dehydrogenase which catalyze the reaction of pyruvate to lactate, occurs, occurs in five different forms

   Lactate + NAD+ LDH Pyruvate + NADH + H+

4. **Competitive Inhibition**

   There are certain products which inhibit the enzyme activity which are known as enzyme inhibitors. Enzyme inhibitors are of two types namely irreversible and reversible. Reversible inhibition is of three types viz., competitive, non-competitive and uncompetitive inhibitors. Competitive inhibitors depend on the fact the inhibitors have structural similarity with the substrate both of which compete for the same active site of the enzyme. The inhibition is removed by increasing the concentration of the substrate. An example of competitive inhibition is the inhibition of succinic dehydrogenase by malonic acid. Inhibition of the activity of this enzyme is due to the structural similarity between malonic acid and succinic acid.

5. **How does enzyme work?**

   There is physical contact between enzyme and substrate. As only a specific key fits in a particular lock to open it, similarly a specific substrate combines with the active site of the specific enzyme to form enzyme substrate complex. This enzyme substrate complex break after the substrate has been converted into the products and the enzyme is again set free. The lock and key theory is supported from the study of the competitive inhibition of the enzyme activity. Another theory is induced fit theory proposed by Koshland. According to this theory when a suitable substrate approached the active site of the enzyme and as it forms the enzyme substrate complex, substrate induces some configurational changes in the enzyme. As a result, the attractive groups form a complementary structure so that the catalytic group of the active site is in proximity of the bond to be broken. After the suitable enzyme substrate complex has been formed, the substrate molecule held by the hydrogen bonds is converted into the products and the enzyme is set free.

6. **Specificity of an enzyme**

   The enzyme is specific in their action. The specificity lies in the fact that they may act.

   1) **Absolute specificity** – Some enzyme are capable of acting on only one substrate eg. Urease act only on urea
   2) **Group specificity**- Some enzymes are capable of catalysing the reaction of a structurally related group of compounds eg. Lactic dehydrogenase catalyze the interconversions of pyruvic acid and lactic acid
   3) **Optical activity**- A particular enzyme will react with only one of the two optical isomers eg. Arginase act only on L arginine and not on its D- isomer
   4) **Geometrical specificity** – Some enzymes exhibit specificity towards the cis and trans forms eg., Fumarase catalyze the inter conversion

7. **Factors influencing enzyme action**

   1) **Temperature**- The activity of enzyme is optimum at normal body temperature. At low temperature, enzyme activity is minimum. An increase in temperature up to certain limit increases the enzyme activity. Maximum being at about 45oC after which enzyme activity is retarded.
   2) p**H-** Enzymes is active only over a limited range of pH
   3) **Water**- In absence pf water, the enzyme activity is suppressed. Proper hydration of the cells is necessary for the enzyme activity
   4) **Substrate concentration**- Enzyme activity increases with the substrate concentration till the active sites of the enzyme are saturated with substrate
   5) **Enzyme concentration**- Increase in concentration of the enzyme will increase the rate of reaction catalysed by it provided there is enough concentration of substrate
   6) **Inhibitors** – Presence of inhibitors in the reaction mixture inhibits the activity of the enzymes partially or completely depending up on the nature of the inhibitors.

8. **Lock and key hypothesis**

   Emil Fischer proposed the lock and key hypothesis to explain the mode of action of enzyme. Interaction of substrate and enzyme is visualized in terms of lock and key model. Union between the substrate and the enzyme takes place at the active site more or less in a manner in which a key fits a lock and results in the formation of an enzyme substrate complex. Enzyme substrate complex depends on a reciprocal fit between the molecular structure of the enzyme and the substrate. This complex is unstable and almost immediately this complex decomposes to produce the end products of the reaction and to regenerate the free enzyme.

9 Inhibitors

There are certain products which inhibit the enzyme activity. These are known as enzyme inhibitors. During the reactions, the active sites of the enzymes are filled up with these substances instead of substrate molecules. Thus, the activity of the enzyme is lot. There are two types of enzyme inhibitors viz., reversible and irreversible. Irreversible inhibition results from the formation of a stable enzyme inhibitor (EI) complex which results in complete inhibition of the enzyme. Reversible inhibition is of three types – competitive inhibition, non-competitive inhibition and uncompetitive inhibition. Incompetitive inhibition,, inhibitor competes with the true substrate for the active site in the enzyme which is removed by increasing the concentration of substrate. In non-competitive inhibitions, the inhibitors react with enzyme to reduce catalytic activity without preventing the formation of enzyme substrate complex. In case of uncompetitive inhibitions, the inhibitor is thought to combine with forms of the enzyme, but they do not combine actually with the substrate. In the case of allosteric inhibition, the inhibitor is structurally different from substrate is bound at a site other than the active site of the enzyme. This alters the configuration of the enzyme protein and thereby prevents it from binding of the substrate.

10. **Allosteric enzyme**

    It is found that when a series of reaction is catalysed by a number of enzymes in sequence, the accumulation of the final end product may cause inhibition in the activity of the first enzyme of the series. This inhibition due to a compound which is totally different in structure from the substrate of the enzyme is called as allosteric inhibition or feedback inhibition and such as enzyme is called as allosteric enzyme. This type of inhibition takes place due to presence of allosteric sites on the surface of the of the allosteric enzyme away from the active site. The final end product molecule fits in the allosteric site and in some way brings about a change in shape of the enzyme so that the active site of the enzyme becomes unfit for making complex with the substrate. The allosteric inhibition id reversible. When the concentration of the final end product in the cell falls, it leaves the allosteric site and the activity of the allosteric enzyme is restored.

11. **Active site**

    Since the substrate molecules are comparatively much smaller than the enzyme molecules, there should be some specific regions on the enzyme for binding with the substrate. Such specific regions are variously called as active site or catalytic site or substrate sites. The active site present in different enzymes has the following common feature

a) Active site occupies very small portion of the enzyme molecule
b) The active site is neither a point or a line or even a plane in a 3D entity
c) The arrangement of atoms in the active site is well defined resulting in the marked Specificity of the enzymes
d) The active site binds the substrate molecules by relatively weak force
e) The active site in the enzyme molecules are grooves or cervices from which water is ``largely excluded

12. **Enzyme modifiers**

The catalytic activity of certain enzymes is reversibly altered by certain inorganic and organic molecules called modifiers. Those molecules which increase the enzyme activity are called positive modifiers or activators and those which decrease the enzyme activity are negative modifiers or inhibitors. Many metals act both as positive and negative modifiers, whereas certain organic molecules retard enzyme activity, thus acting as negative modifiers. The inorganic modifiers (enzyme activators) are mostly metals. Certain enzymes apart from a requirement of coenzymes also need a metal ion for full activity. Removal of metal often results in partial or total loss of enzyme activity. The activity is however restored by replacing the metal ions. Such metal ions include K+, Cu+, Fe2+, Mg2+, Ca2+, Mn2+, Zn2+ etc., . For eg, Fe, Cu and Mo are required in oxido reduction reaction. Mg is needed for phosphate reaction. The mechanism on how metal ion brings about activation include as direct participation in catalysis, formation of metallo substrate, formation of metalloenzymes. Alteration of equilibrium constant and conformational change in the enzyme. Organic modifiers (enzyme inhibitors) are of two types reversible and irreversible. Reversible inhibition include competitive, non-competitive and uncompetitive.

ESSAY TYPES

1. **Explain in detail about classification of enzymes**

Enzymes may be classified according to the type of reaction that they catalyze

a) **Oxido reductases**- These enzymes catalyze reaction in which one substrate is oxidize, acting as hydrogen donor, and another substrate is reduced. The Oxido reductases include the dehydrogenases which convert single bonds to double bonds and the oxidases which user oxygen as the oxidant. Others include peroxidases which use H_2O_2 as the oxidant, the hydrolases which introduce hydroxyl group and the oxygenases which introduce molecular oxygen in place of a double bond in a substrate
b) **Transferases**- Enzymes in this class transfer one carbon group (methyl aldehyde or Ketonic groups, or phosphoryl/ or amino group and so on from one substrate to another eg., transaminases, transaldolases
c) **Hydrolases-** It catalyze the hydrolytic (water adding) cleavage of C-O, C-N, C-C and other single bonds. The class includes peptidases, esterases, glycosidaes, phosphatases
d) **Lyases**- These enzymes cleave bonds without the addition of water, but by elimination reactions to form double bonds or rings. It also catalyzes the reverse reaction. Usually C-C, C==O , C=N bonds are acted upon eg., decarboxylases, aldolases, dehydralases

e) **Isomerases**- Racemases, epimerases cis- trans isomerases, intramolecular oxido reductases , mutases which alter the structure, but not the atomic composition of substrates

f) **Ligases-** enzyme in this class, also known as synthetases, couple the hydrolysis of a pyrophosphate in ATP or other triphosphate to a second reaction in which two molecules are joined eg., RNA ligase will form a new phosphodiester bonds

2) Discuss the biochemical action of vitamins as coenzyme

Vitamins are organic substances which cannot be synthesized by some organisms and must therefore be supplied to them regularly out in small quantities in order to ensure a normal metabolism. Certain vitamins, notably water soluble vitamins and particularly the B group vitamins are coenzymes or essential constituents of coenzymes. It must be noted that the same compounds often serve as coenzymes for all living organisms, but vitamins only for some of them.

1) Nicotinamide Adenine dinucleotide (NAD)- It is coenzyme of various dehydrogenases. It binds the hydrogen originating from a substrate AH_2 and transforms into NADH+ H^+

2) Nicotinamide Adenine dinucleotide phosphate (NADP) – It is also coenzyme of some dehydrogenases very similar to NAD. It differs only by an additional phosphate residue esterified on the hydroxyl in 2' of the ribose to adenine. It is a hydrogen acceptor

3) Flavin nucleotides- Flavin mononucleotides (FMN) and Flavin adenine dinucleotide (FAD) are also coenzymes of dehydrogenases often called flavour proteins. These two coenzymes derive from riboflavin or vitamin B2. They are involved in the process of cell oxidation by fixation- reversible of 2H. Succinate dehydrogenases is an eg. of FAD enzyme

4) Ferro prophyrin- These ferro prophyrin coenzymes are associated with cytochromes

5) Lipoic acid- This coenzyme acts as hydrogen transporter in the oxidative decarboxylation reactions, particularly in that of pyruvic acid

6) Thiamine pyrophosphate (TPP) - The pyrophosphoric ester of thiamine or vitamin B1 is an integral part of the active site of Carboxylase. It is a transporter of aldehyde, particularly acetaldehyde. It is a coenzyme of decarboxylases of α- keto acids

7) Coenzyme A- Coenzyme A derives from B group vitamins, the pantothenic acid. Coenzyme A is important in the oxidation of fatty acid as well as in biosynthesis of fatty acid

8) Tetrahydrofolic acid- It is the active form of a vitamin folic acid. It is coenzyme transporter of C1 units which behaves as a substrate and is involved in a large number of processes like biosynthesis of purine ribonucleotides, glycine-serine interconversions.

9) Biotin- It is coenzyme of different carboxylation reactions. It is involved in the carboxylation of acetyl coenzyme A into malonyl coenzyme A in the synthesis of fatty acid.

10) Pyridoxal phosphate- It is coenzyme derivative of pyridoxine or vitamin B6. It is an integral part of the active site of various enzymes involved in the metabolism of amino acids, transaminases, amino acid decarboxylases

11) Cobamide coenzymes- It is derivative of cobalamine or vitamin B12. It is involved in various isomerisation reaction especially transfer of carboxyl group in the case of isomerisation of methyl malonyl CoA to succinyl Coenzyme A

3) Discuss the chemical nature and properties of enzymes. State briefly how an enzyme exerts its catalytic activity?

Chemical nature

The enzymes are essentially proteins and possess properties characteristic to these. Enzyme is considered as a protein with catalytic properties due to its power of specific activation. Enzymes contain C, H, N and S. It has amphoteric property. It undergoes Denaturation and form antibodies. Chemically the enzyme may be divided into two categories

1) Simple- protein enzymes- They contain simple proteins eg., Urease, amylase

2) Complex- protein enzymes- Thèse contain conjugale proteins. They have protein part called Apoenzyme and non-protein part called prosthetic group associated with the protein part. The two parts constitute what is called as holoenzyme eg. Catalase, cytochrome the activity. The activity of an enzyme depends on the fact that non-proteinaceous prosthetic group is intimately associated with the protainaceous Apoenzyme

Conjugate-protein enzyme $\rightleftarrows$ protein part + prosthetic group

Holoenzyme $\rightleftarrows$ Apoenzyme + coenzyme

Coenzymes are thermostable, dialyzable organic compounds and account for 1% of the entire enzyme molecule

Properties of enzyme

2) Catalytic power- Only small amount of enzyme is enough to convert large quantity of the substrate into products. Their turn over number ie., the number of substrate molecules converted by one molecule of the enzyme per second when its active site is saturated with substrate

3) Specificity – Enzymes are very specific in their action. A particular enzyme usually acts on a particular substrate to catalyze a particular type reaction

4) Reversibility- In most of the cases, the reactions catalyzed by the enzymes are reversible depending upon the requirement of the cell. But in some cases, there are separate enzymes for forward and backward reactions

5) Thermolabile- The enzymes are very sensitive to heat. They are inactivated at very low temperature. At a very temperature (60-70^{o}C) usually they are destroyed.

6) Inhibitors- Enzymes are sensitive to inhibitors, while some inhibitors may be partially inhibit their activity; other inhibitors destroy them permanently and inhibit activity.

7) Colloidal nature- The enzymes are of colloidal dimensions and present large surfaces for reactants in water to facilitate the enzyme reaction

Mode of action of enzyme is explained by two theories

2) Collision theory- Some enzymes need water to break the bond, while others require compounds of low molecular weight to make the enzyme active. The enzyme like lipases, chymotrypsin, phosphatases generally catalyze the reaction following way

$$\underset{\text{Enzyme}}{EH} + \underset{\text{Substrate}}{Rx} \longrightarrow \underbrace{ER + Hx}_{\text{Intermediates}}$$

$$ER + H_2O \longrightarrow ROH + EH$$

On summing up $Rx + H_2O \longrightarrow ROH + Hx$

Other enzymes like hexokinases, Co A Transferases catalysed the reaction in the following manner:

$$E + AB \longrightarrow E\text{-}A + B$$
$$E\text{-}A + C \longrightarrow E + AC$$

The enzyme (E) first combines with A part of the compound (AB) and conserves the bond energy. It reacts with (C) compound to form (AC). Water molecules here do not react with (E-A) complex and the energy of the bond is conserved to be used for the synt

Enzyme-substrate complex theory- It was proposed by Michalis and Menton

Enzyme + Substrate → Enzyme – substrate complex

Enzyme- substrate complex → Enzyme + product

The substrate combines with the enzyme and leaves the enzyme surface in the form of reaction products. Enzyme substrate complex is formed as intermediates which are decomposed rapidly into the enzyme and the product molecules of a new compound (AC).

4 **Describe in detail the various factors which affect the enzyme activity**

The rate of an enzyme- catalysed reaction is influenced by the following environmental factors

1) **Enzyme concentration**- Enzyme molecules are larger than the substrate molecules. They possess many active sites in which the substrate molecules get attached at the time of reaction. The reaction proceeds until all the active sites of the enzyme molecules are filed by the substrate molecules.

2) **Substrate concentration**- If the enzyme concentration is fixed and the substrate concentration is relatively higher, the velocity of the reaction is increased to its maximum. If the substrate concentration is relatively low at a particular enzyme concentration, the reaction proceeds, but at slower rate because the active sites of only few enzyme molecules become saturated with the substrate molecules. An increase in the substrate increase the rate of reaction and if the substrate concentration is increased beyond a limit, when all the active sites of the enzyme molecules have become saturated, the reaction will become constant without accelerating the velocity.

3) **Temperature.-**The rates of enzyme catalysed reaction are increased as the temperature is raised. The enzyme catalysed reaction show the increase in velocity between 25-35oC. At 0oC, the enzyme become inactivated, but not destroyed. At 60 to

70oC, the enzymes are inactivated and destroyed. It is due to coagulation and Denaturation.

4) **Hydrogen concentration**- Hydrogen ion concentration of an enzyme solution has a marked effect on its activity. Every enzyme act at a particular pH. The pH at which the rate of reaction is maximum is called as optimum pH range. If the pH value is increased or decreased at either side of the optimum pH range, the rate of reaction usually decreases. At extreme pH the enzymes are denatured and inactivated. The hydrogen ion concentration also alters the ionization, solubility of the substrates, inhibitors, activators and absorbed ions

5) **Effect of ions**- The cations like Mg2+, Ca2+, Mn2+, Zn2+,Na+ or K= play important role in the activity of certain enzymes. These enzymes on the absence of a particular cation remain inactive.

6) **Accumulation of products**- The accumulation of products of enzyme catalyzed reaction may increase the rate of reverse reaction. The enzymes become inactive due to accumulation of these products on the surface of the enzyme molecules itself. They may also change the pH of the enzyme solution.

5) Write an account on the occurrence and nomenclature of enzymes

Every cell synthesizes its own enzymes with the help of genes. The synthesized enzymes are then distributed to the different parts of the cell and ultimately to the different parts of the plant or animal body. The enzymes that are produced within the cell for metabolic activities are known as endoenzymes and those which act away from the site of synthesis are called exoenzymess

Since the enzymes are specific for a particular reaction, they are named according to the substrate on which they act or on the nature of reaction they catalysed. The most common methods for naming them is suffix ase at the end of the name of the substrate attached. Thus peptide is attacked by peptidases, lipid by lipase, and urea by urease

The international commission established a numerical system of classification

1) Each enzyme has a systematic code number (EC) of four digits
2) The first digit of the four figures indicate the main class
3) The second digit indicates the subclass
4) The third digit indicates the subdivision of sub class
5) The fourth digit designates the aerial number of the specific enzyme in the fourth sub –class
6) In the system of nomenclature, enzyme commission recommends both systematic and trivial names for enzymes for eg, EC.1, 1, 1, 1 stands for the enzyme alcohol dehydrogenase EC stands for the enzyme commission
    1. Stands for oxido reductases
    1.1. Stands for enzyme which utilizes substrate as CHOH (alcohol group)
    1.1.1. Stands for the enzymes which utilizes NAD as an acceptor

6). Give a brief note on the practical application of enzymes

1) Wine manufacturing- Papain is used in brewing industry as a stabilizer for chill proof beer

2) Cheese making- An enzyme lipase is added to cheese for imparting flavour to it.
3) Candy making- Invertase helps preventing granulation of sugars in soft cantered candies
4) Bread whitening- Lipoxygenase is used for whitening of bread
5) Clarifying fruit juice- Pectic enzymes is used for clarifying the fruit juice
6) Destaining and desizing fabrics- Alcalase is used for removing stains due to glue, gelatin or starch.
7) Wound healing- Proteolytic enzymes are used to alleviate skin diseases, bed sores
8) Analysing biochemical- Uricase and urease are employed in the determination of uric acid and urea respectively
9) Diagnosing hypertension- Radio immunoassay procedure is used for diagnosing cases of hypertension by employing a proteolytic enzymes secreted by the kidneys.
10) Syrup manufacturing- Immobilized glucose isomerase is being used in the production of high fructose core syrup

7) **Write a detailed account on enzyme inhibition**

There are certain products which inhibit the enzyme activity. These are known as enzyme inhibitors. During the reaction, the active sites of the enzymes are filled up with theses inhibitors instead of substrate molecules. Thus the activity of the enzymes is lost. There are two types of enzyme inhibition 1) irreversible 2) reversible. Irreversible inhibition results from the formation of a stable enzyme inhibitor (EI) complex which results in complete inhibition of the enzyme. Reversible inhibition is of three types a) Competitive inhibition b) Non- competitive c) Uncompetitive

A) **Competitive inhibition**- this type of inhibition depends on the fact the inhibitor competes with the true substrate for the active site of the enzyme. The inhibition is retrieved by increasing the concentration of the substrate eg. The inhibition of succinic dehydrogenase by malonic acid since molecular structure of succinic acid is very similar to malonic acid. Because of this similarity in structure, the enzyme can react with both to form complex. However, only the enzyme- succinic acid complex decomposes to yield a reaction product

$$E + Iki \xleftrightarrow{KI} EI$$

Where E is the enzyme, I is inhibitor (malonic acid0, Ki inhibitor association constant and EI enzyme inhibitor complex.

B) **Non- competitive inhibition**- In non- competitive inhibition, the inhibitor reacts with the enzyme to reduce catalytic activity without preventing the formation of enzyme – substrate complex. The affinity of the enzyme for substrate is not reduced, but maximum velocity of the reaction is reduced eg. Fluoride ion inhibits enolase and thiocyanate ion inhibit fumarase. Inhibition cannot overcome by increasing substrate concentration,

C) **Uncompetitive inhibition**- The inhibitor is thought to combine with forms of the enzyme, but they do not combine actually with the substrate. These forms of the enzyme are known as substrate non- combining forms and cannot then be converted back to the substrate combining forms of the enzyme. This type of inhibition is not relieved by increasing the concentration of the substrate.

D) **Allosteric inhibition**- The inhibitor is structurally quite different from the substrate, is bound at asite other than the active site of the enzyme. This binding of the inhibitor alters the conformation of the enzyme protein and thereby prevents it from binding to the substrate. Since the inhibitors bind at a site other than the active site of the enzyme, they are called allosteric effectors and phenomenon is called as allosteric effect.

8) **Explain the mechanism and co enzymatic action with suitable examples**

Mechanism of enzyme action

The mechanism of enzyme action is explained by collision theory and enzyme substrate complex theory

1) **Collision theory**- Some enzymes need water to break the bond, while others require compounds of low molecular weight to make the enzyme active. The enzyme like lipases, chymotrypsin, phosphatases generally catalyze the reaction following way

$$\underset{\text{Enzyme}}{EH} + \underset{\text{Substrate}}{Rx} \longrightarrow \underset{\text{Intermediates}}{ER + Hx}$$

$$\underline{ER + H_2O \longrightarrow ROH + EH}$$

On summing up $Rx + H_2O \longrightarrow ROH + Hx$

Other enzymes like hexokinases, CoA Transferases catalysed the reaction in the following manner:

$E + AB \longrightarrow E\text{-}A + B$

$E\text{-}A + C \longrightarrow E + AC$

The enzyme (E) first combines with A part of the compound (AB) and conserves the bond energy. It reacts with (C) compound to form (AC). Water molecules here do not react with (E-A) complex and the energy of the bond is conserved to be used for the synthesis of a new compound (AC).

2) Enzyme-substrate complex theory- It was proposed by Michaelis and Menton

Enzyme + Substrate ⟷ Enzyme – substrate complex

Enzyme- substrate complex ⟶ Enzyme + product

The substrate combines with the enzyme and leaves the enzyme surface in the form of reaction products. Enzyme substrate complex is formed as intermediates which are decomposed rapidly into the enzyme and the product molecules.

Mechanism of Co enzyme action

The coenzyme is as essential a reactant as the substrate that is activated by the enzyme. The following equation represents the reaction of the coenzyme with the substrate

Substrate (A) + Co enzyme $\xrightarrow{\text{Enzyme A}}$ Substrate (A) derivatives+ Co enzyme derivatives

The coenzyme is regenerated and again becomes available as an essential component for the above reaction

Substrate (B) + co enzyme derivative $\xrightarrow{\text{Enzyme B}}$ Substrate (B) derivative + coenzyme

The function of coenzyme in the enzymatic reactions is thus to assist in their cleavage of the substrate by acting as an acceptor for one of the cleavage products.

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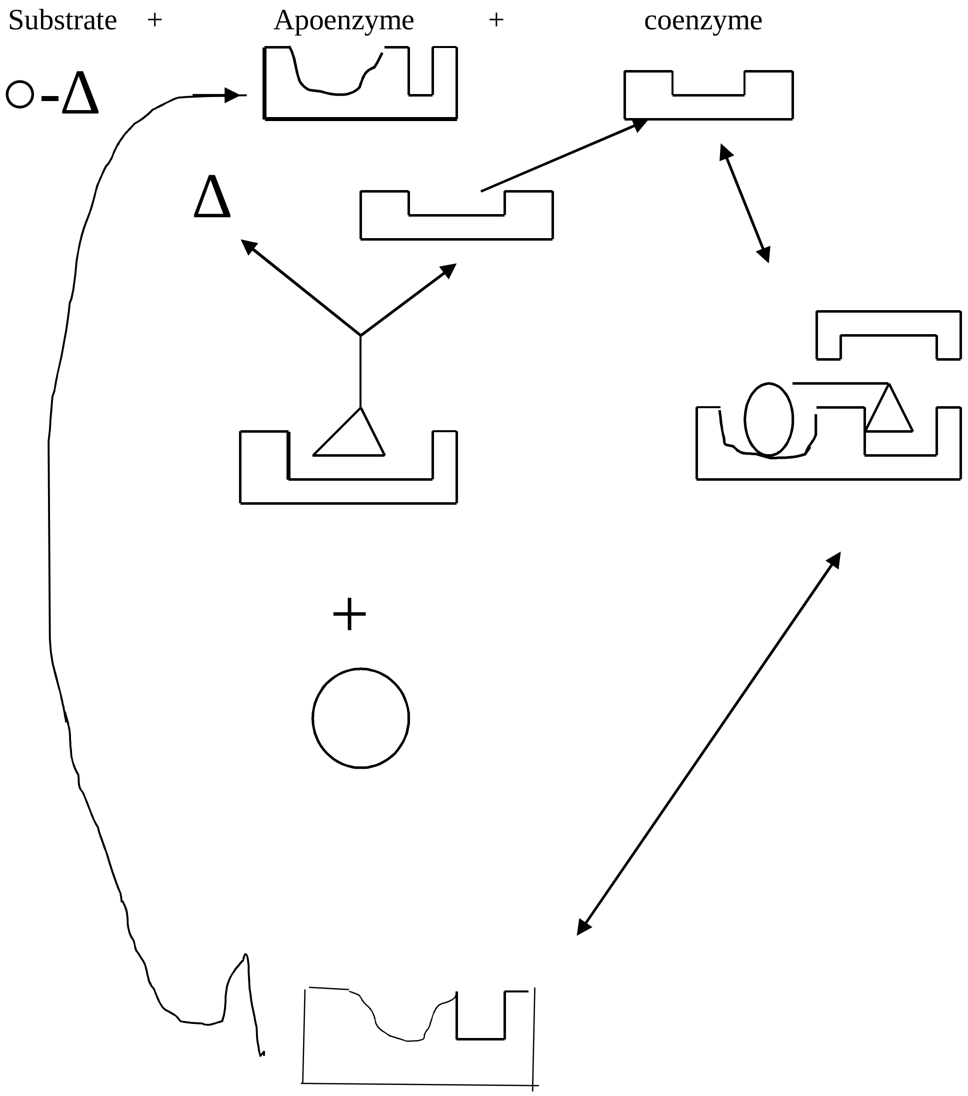
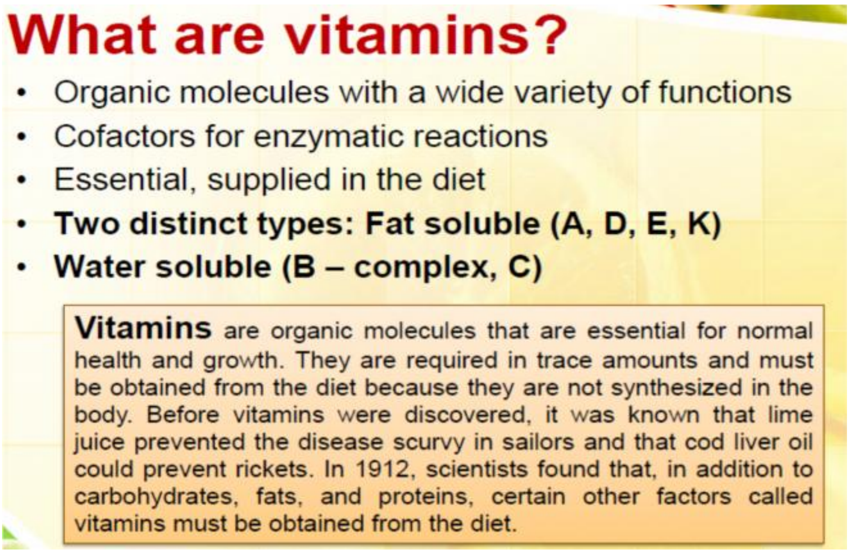


Fig1. Function of coenzymes

For eg. Dehydrogenases utilize NAD+ or NADP. Their functions is to transfer the hydrogen nuclei with two electrons from the substrate, thus oxidizing it

Substrate + NAD+ + Enzyme → oxidized substrate + NADH + H+

VITAMINS



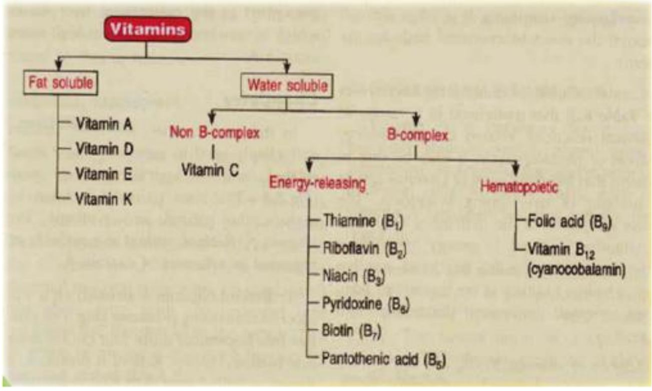
What are vitamins?

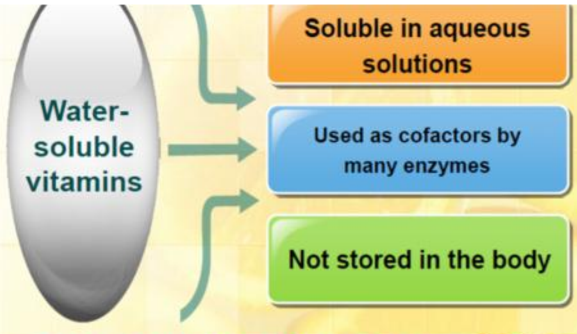
- Organic molecules with a wide variety of functions
- Cofactors for enzymatic reactions
- Essential, supplied in the diet
- **Two distinct types: Fat soluble (A, D, E, K)**
- **Water soluble (B – complex, C)**

Vitamins are organic molecules that are essential for normal health and growth. They are required in trace amounts and must be obtained from the diet because they are not synthesized in the body. Before vitamins were discovered, it was known that lime juice prevented the disease scurvy in sailors and that cod liver oil could prevent rickets. In 1912, scientists found that, in addition to carbohydrates, fats, and proteins, certain other factors called vitamins must be obtained from the diet.

Why are they good for us?

- Greater need due to worse environment
- Improve immunity
- Prevent ilnesses
- Slower aging





Water-soluble vitamins

- Soluble in aqueous solutions
- Used as cofactors by many enzymes
- Not stored in the body

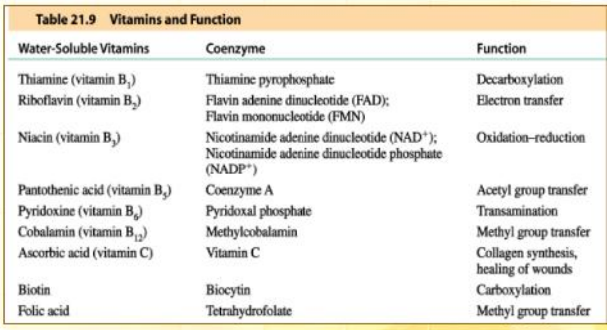
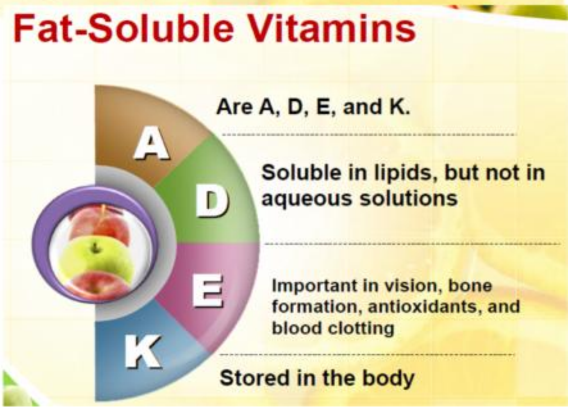


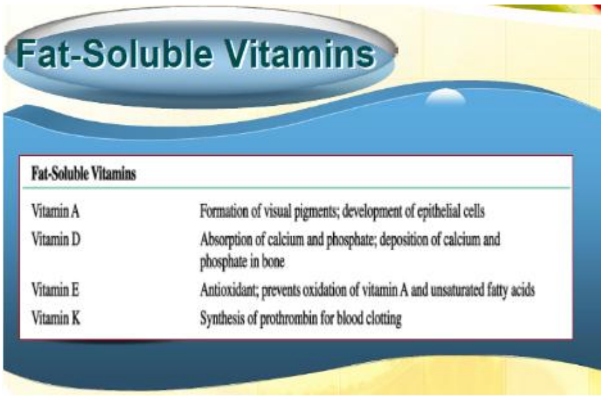
Table 21.9 Vitamins and Function

Water-Soluble Vitamins	Coenzyme	Function
Thiamine (vitamin B_1)	Thiamine pyrophosphate	Decarboxylation
Riboflavin (vitamin B_2)	Flavin adenine dinucleotide (FAD); Flavin mononucleotide (FMN)	Electron transfer
Niacin (vitamin B_3)	Nicotinamide adenine dinucleotide (NAD^+); Nicotinamide adenine dinucleotide phosphate ($NADP^+$)	Oxidation–reduction
Pantothenic acid (vitamin B_5)	Coenzyme A	Acetyl group transfer
Pyridoxine (vitamin B_6)	Pyridoxal phosphate	Transamination
Cobalamin (vitamin B_{12})	Methylcobalamin	Methyl group transfer
Ascorbic acid (vitamin C)	Vitamin C	Collagen synthesis, healing of wounds
Biotin	Biocytin	Carboxylation
Folic acid	Tetrahydrofolate	Methyl group transfer



Fat-Soluble Vitamins

- Are A, D, E, and K.
- Soluble in lipids, but not in aqueous solutions
- Important in vision, bone formation, antioxidants, and blood clotting
- Stored in the body

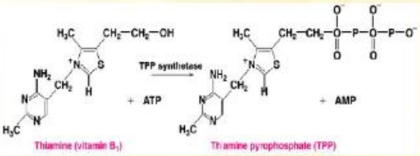


Fat-Soluble Vitamins

Fat-Soluble Vitamins	
Vitamin A	Formation of visual pigments; development of epithelial cells
Vitamin D	Absorption of calcium and phosphate; deposition of calcium and phosphate in bone
Vitamin E	Antioxidant; prevents oxidation of vitamin A and unsaturated fatty acids
Vitamin K	Synthesis of prothrombin for blood clotting

Thiamine (Vitamin B$_1$)

- Was the first B vitamin identified.
- Is part of the coenzyme thiamine pyrophosphate(TPP).



- TPP coenzyme is required by enzymes in the decarboxylation of α-keto carboxylic acids.
- Deficiency results in beriberi (fatigue, weight loss, and nerve degeneration).

Riboflavin (Vitamin B$_2$)

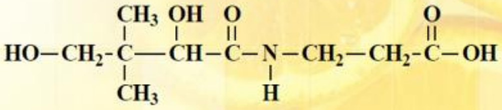
- Made of the sugar alcohol *ribitol* and *flavin*
- Part of the coenzymes flavin adenine dinucleotide (FAD) and flavin mononucleotide (FMN)
- Needed for good vision and healthy skin

Niacin (Vitamin B$_3$)

- Is part of the coenzyme nicotinamide adenine dinucleotide (NAD^+) involved in oxidation-reduction reactions.
- Deficiency can result in dermatitis, muscle fatigue, and loss of appetite.
- Is found in meats, rice, and whole grains.

Pantothenic Acid (Vitamin B$_5$)

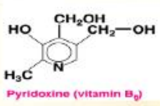
- Is part of coenzyme A needed for energy production as well as glucose and cholesterol synthesis.
- Deficiency can result in fatigue, retarded growth and anemia.
- Part of coenzyme A
- Reduces stress, prevents tiredness
- Necessary for formation of glycogen, fatty acids, steroid hormones
- Sources: whole grains, and vegetables ,eggs,liver, heart, yeast

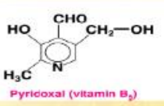


$$HO-CH_2-\underset{CH_3}{\overset{CH_3}{C}}-\overset{OH}{CH}-\overset{O}{\overset{\|}{C}}-\underset{H}{N}-CH_2-CH_2-\overset{O}{\overset{\|}{C}}-OH$$

Pyridoxine (Vitamin B$_6$)

- Pyridoxine and pyridoxal are two forms of vitamin B$_6$, which are converted to the coenzyme pyridoxal phosphate (PLP).


Pyridoxine (vitamin B$_6$)


Pyridoxal (vitamin B$_6$)


Pyridoxal phosphate (PLP)

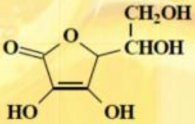
- PLP is required in the transamination of amino acids and decarboxylation of carboxylic acids.
- Deficiency of pyridoxine may lead to dermatitis, fatigue, and anemia.

Cyanocobalamin

- Formation of red blood cells
- Maintenance of neuro tissue
- Cure of neuro diseases, anemia
- Sources: dairy products, meat, poultry, sea products

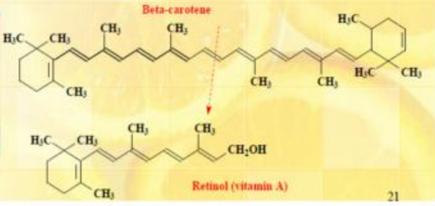
Ascorbic Acid (Vitamin C)

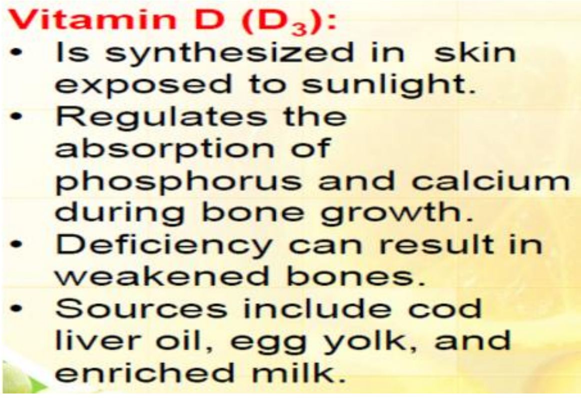
- Is required in collagen synthesis.
- Deficiency can lead to weakened connective tissue, slow-healing wounds, and anemia.
- Is found in Indian gooseberries, blueberries, citrus fruits, tomatoes, broccoli, red and green vegetables.



Vitamin A

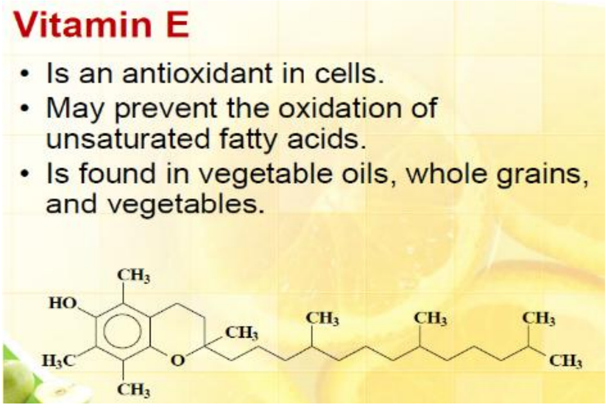
- Vitamin A is obtained from meats and beta-carotenes in plants.
- Beta-carotenes are converted by liver enzymes to vitamin A (retinol).





Vitamin D (D_3):

- Is synthesized in skin exposed to sunlight.
- Regulates the absorption of phosphorus and calcium during bone growth.
- Deficiency can result in weakened bones.
- Sources include cod liver oil, egg yolk, and enriched milk.



Vitamin E

- Is an antioxidant in cells.
- May prevent the oxidation of unsaturated fatty acids.
- Is found in vegetable oils, whole grains, and vegetables.

Vitamin E

Tocoferols

- Antioxidant (lipid protection)
- improves immunity
- Cures muscle, heart and skin diseases, burns
- Sources: green vegetables, vegetable oil, corn, eggs, bread, dairy products, peas, beans

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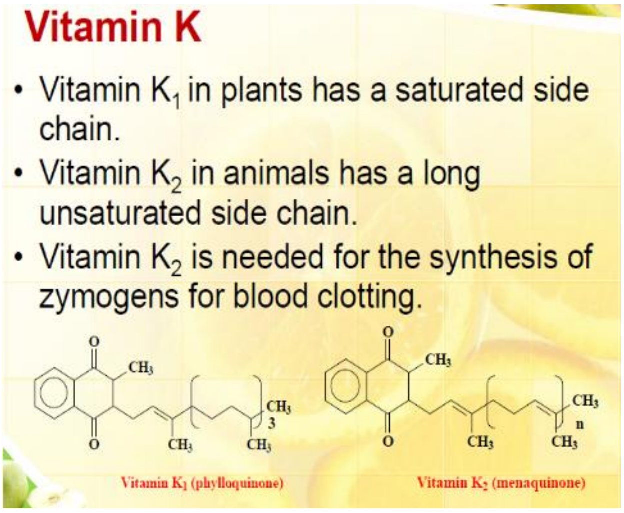


Vitamin K

- Vitamin K_1 in plants has a saturated side chain.
- Vitamin K_2 in animals has a long unsaturated side chain.
- Vitamin K_2 is needed for the synthesis of zymogens for blood clotting.

Vitamin K_1 (phylloquinone)

Vitamin K_2 (menaquinone)

Fill up the Blanks

1. The fat soluble vitamins are A , D, ___ and ____
2. The water soluble vitamin ae ____ and ______
3. Vitamin A is also called as________
4. Thiamine is also called as ________
5. Vitamin B2 is called as _______
6. Niacin is also called by _______
7. Ascorbic acid is referred as ________
8. Vitamin D is also called as ________
9. Vitamins often act as ________
10. A steroid vitamin is _______

11. Ascorbic acid act as a ______ agent
12. The first person to discover vitamins was ________
13. The cobalt containing vitamin is _______
14. Vitamin C is considered as __________ in water
15. The vitamin that act as coenzyme is ______
16. The most important function of vitamins is to serve as ________
17. Among the vitamins, ________ show greater deficiency in body
18. Several vitamins have strong ________ potential
19. Vitamins which are not stored in body are called_____
20. Vitamin which doesn't act as a coenzyme is_______

ANSWER

Sl.No	Answer	Sl.No	Answer
1	E and K	11	Reducing
2	B and C	12	Funk
3	Retinol	13	Vitamin B12
4	Vitamin B1	14	Soluble
5	Riboflavin	15	Vitamin B complex
6	Vitamin B3	16	coenzyme
7	Vitamin C	17	Water soluble
8	Calciferol	18	Antioxidant
9	Co enzyme	19	Water soluble
10	D	20	Vitamin C

Essay type

1) General characteristics of vitamins

a. Vitamins occur in plants, animals as well as microorganisms, i.e. it has widespread occurrence.
b. The plants and many microbes can synthesize all the vitamins, whereas only a few vitamins are synthesized in animals.
c. Human body can synthesize some vitamins. For eg. Vitamin A from precursor carotene and Vitamin D from ultraviolet irradiation of 7-dehydrocholesterol. Some members of Vitamin B- complex are synthesized by micro-organisms present in the intestinal tract. Vitamin C is also synthesized in some animals such as rat.
d. All the cells of the body store vitamins to some extent.
e. Vitamins are partly destroyed and partly excreted.
f. Vitamins are non-antigenic.
g. Most of the vitamins are coenzymes, however few are haematopoietic.
h. Vitamins carry out functions in very low concentration; hence total daily requirement is very small.
i. Vitamins are effective when taken orally.

j. A lack of one or more vitamins (avitaminosis) leads to characteristic deficiency symptoms in humans as well as in animals.

2). Function of vitamins

A vitamin is an organic compound and an essential nutrient that an organism requires in limited amounts. Vitamins have diverse biochemical functions.

1. Hormone-like functions as regulators of mineral metabolism (Vitamin D)
2. Regulators of cell and tissue growth and differentiation (Vitamin A)
3. As antioxidants (e.g., vitamin E and sometimes vitamin C)
4. Enzyme cofactors (tightly bound to enzyme as a part of prosthetic group, coenzymes (vitamins, the B complex). In this role, vitamins may be tightly bound to enzymes as part of prosthetic groups: For example, biotin is part of enzymes involved in making fatty acids

3) Classification of vitamins

The body needs 13 vitamins for normal health. These include vitamins A, C, D, E, K and the B complex (thiamine, riboflavin, niacin, pantothenic acid, biotin, vitamin B 6, B 12 and folate. Each of these vitamins provides various functions for the body. Under normal circumstances, a person can obtain all of the vitamins he needs from eating a well-balanced diet.

Vitamins are classified several different ways, according to how they travel through your body and the various roles they play in keeping you healthy (Vitamins are classified by their biological and chemical activity)**.** Vitamins are classified primarily as to solubility. Some are soluble in fat and some in water. Vitamins that are fat soluble are stored in the body and can accumulate. Water soluble vitamins are flushed out by the kidneys.

1- Fat soluble vitamins:
  a) Vit. A (Retinol, Retinal, Retinoic acid)
  b) Vit. D (Vit. D3: Cholecalciferol, Vit. D2: Ergocalciferol)
  c) Vit. E (α-Tocopherol)
  d) Vit. K (Vit. K1: Phylloquinones, Vit. K2: Menaquinones)

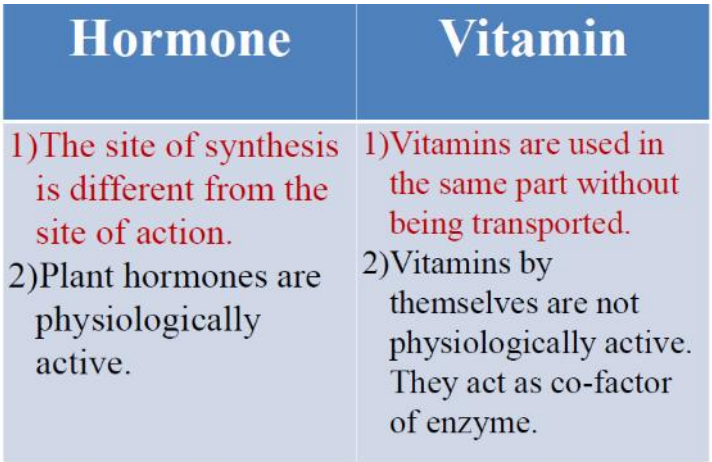
2-Water soluble vitamins:
  (A) Vit. B group
    B1 (Thiamin)
    B2 (Riboflavin)
    B3 (Niacin)
    B5 (Pantothenic acid)
    B6 (Pyridoxine)
    B9 (Folic acid)
    B12 (Cyanocobalamin)
  B) Vit. C : (l-Ascorbic acid)
  (C) Vit. H: (Biotin)

Definition

1. **Vitamins:** Any of a group of organic compounds which are essential for normal growth and nutrition and are required in small quantities in the diet because they cannot be synthesized by the body.
2. **Water soluble vitamins**: A vitamin that can dissolve in water. Water-soluble vitamins are carried to the body's tissues but are not stored in the body. Water-soluble vitamins are found in fruit, vegetables and grains.
3. **Fat soluble vitamins:** A vitamin that can dissolve in fats and oils. Fat-soluble vitamins are absorbed along with fats in the diet and can be stored in the body’s fatty tissue. They come from plant and animal foods or dietary supplements Vitamins A, D, E, and K are fat-soluble.

PLANT HORMONES

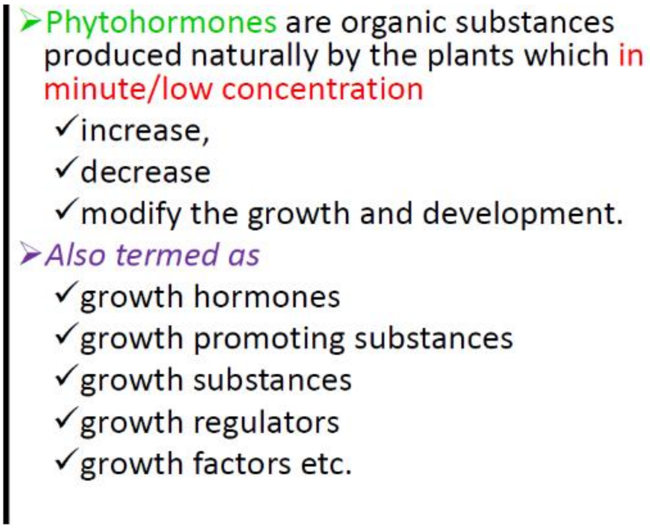
Plant‘s growth and development are under the control of two sets of internal factors. Nutritional factors such as supply of carbohydrates, protein, fats and others constitute the raw materials required for growth. Proper utilization of these raw materials is under the control of certain chemical messengers which can be classified into hormones and vitamins



Hormone	Vitamin
1)The site of synthesis is different from the site of action. 2)Plant hormones are physiologically active.	1)Vitamins are used in the same part without being transported. 2)Vitamins by themselves are not physiologically active. They act as co-factor of enzyme.

The term hormone is derived from a greek root “hormao” which means “to stimulate”(Beylis and Starling, 1902). Thimann (1948) suggested using the term “phytohormone” for hormones of plant

A hormone is a naturally produced chemical synthesized in one part of the plant and then travels to another part where it effects growth and development. Hormone- gr. to excite. Organic substances produced in small amounts that regulate and coordinate metabolism, growth, and morphogenesis



- Phytohormones are organic substances produced naturally by the plants which in minute/low concentration
  - ✓increase,
  - ✓decrease
  - ✓modify the growth and development.
- *Also termed as*
  - ✓growth hormones
  - ✓growth promoting substances
  - ✓growth substances
  - ✓growth regulators
  - ✓growth factors etc.

Plant Growth regulators

Plant growth regulators (PGR) refers to natural or synthetic substances influence the growth and development

IAA (Auxin) – both natural and synthetic

IBA (Auxin) – Always synthetic

All plant hormones are plant growth regulators, but all plant growth regulators are not plant hormones

Six Major Classes of Plant Hormones

Auxins

Leaf primordia, young leaves, developing seeds.

Polarly (unidirectionally) and nonpolarly.

Cytokinins

Root tips.

From roots to shoots via xylem.

Ethylene

Most tissues in response to stress.

Diffusion from site of synthesis.

Abscisic acid

Mature leaves and roots, seeds.

From leaves in phloem and from the roots in the xylem.

Gibberellins

Young tissues of the shoot and developing seeds.

Xylem and phloem.

Brassinosteroids

Young tissues and throughout the plant.

They act locally.

Classification of PGR

a) **On the basis nature of function**

Growth promoting hormones / growth regulators: Increase the growth of the plants

Eg Auxin, Gibberellin and cytokinins

b) Growth inhibiting hormones/ growth retardant

Inhibit the growth of the plant

Eg, ABA, ethylene

Auxin

Derived from the greek word “ auxein” means “to grow/increase”. Auxins may be defined as growth promoting substances which promote growth along the vertical axis when applied in low concentration to the shoot of the plant

The idea of existence of auxin was proposed by Charles Darwin (1880) in his book “The power of movements in plants”. Coleoptiles of canary grass to unilateral light and observed it to bend towards light. He covered the coleoptiles tip with tin foil or cut it off and observed that coleoptiles did not bend towards unilateral light. Concluded some stimulus is transmitted from upper to the lower part which induced bending of the coleoptiles

Auxin – promotes plant growth causes elongation of developing cells. Increases the concentration of hydrogen ion in primary walls, in turns, loosens cellulose fibers which increase cell wall plasticity. In response, turgor pressure causes the cell expand, thus generating growth. It is produced at the tips of shoot and roots. Active phototropism and geotropism. Active in leaves, fruits and germinating seeds

Auxin Provides Chemical Signals That Communicate Information Over Long Distances

- Apical Dominance- inhibitory influence of the apical bud upon lateral buds.

Auxin Plays a Role in the Differentiation and Regeneration of Vascular Tissue

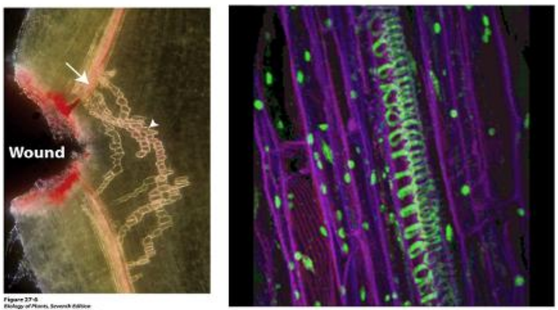


Figure 27-6
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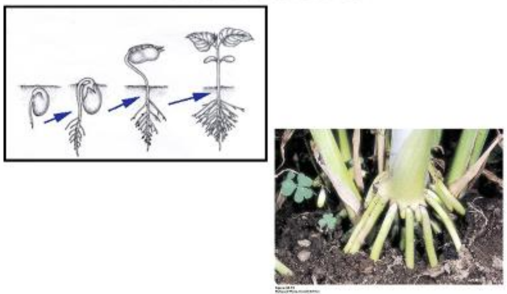
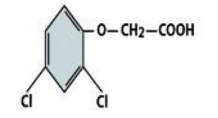
Auxin Promotes Fruit Development

Auxin Promotes the Formation of Lateral and Adventitious Roots



Auxin Prevents Abscission

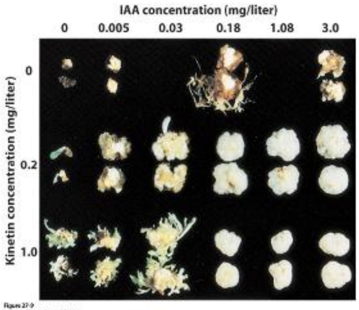
Synthetic Auxins Kill Weeds- Herbicides



2,4-Dichlorophenoxyacetic acid (2,4-D)

Cytokinins

Group of hormones that stimulate cytokinesis (cell division). Produce mainly in the roots and transported throughout the plant. Stimulates the growth of lateral buds, thus weakening apical dominance.



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Cytokinins Promote Cell Division

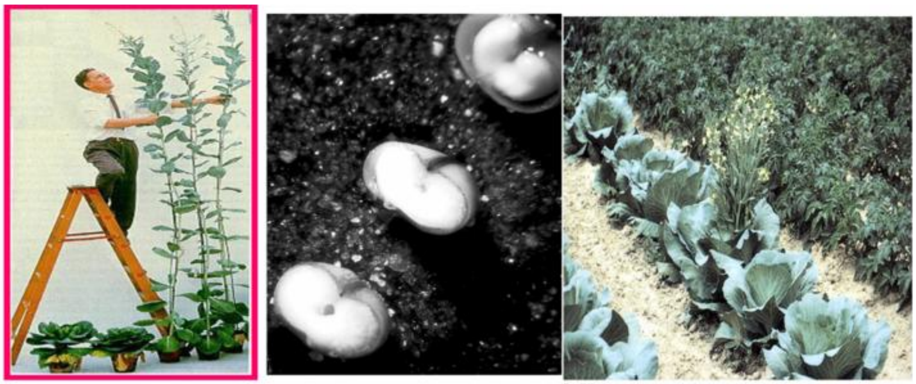
Cytokinins Delay Leaf Senescence

Cytokinins Promote the Growth of Lateral Buds

Gibberellins (gibberelic acid- GA)

Promotes cell growth; Synthesized in young leaves, roots, and seeds. Involved in the promotion of fruit development and seed germination. High concentrations of GA causes rapid elongation of stems called bolting.

It was discovered by Japanese plant pathologist Kurosawa in the year 1928. Rice plants was infected by fungus *Gibberella Fujikurai* showed excessive cell elongation. The symptom is called Bakane disease. Chemical was extracted and purified and named as Gibberellic acid. Now there are 80 type of gibberelic acid. The most common is GA_3 .



Gibberellin (GA) causes dwarf mutants to grow tall

Breaking seed dormancy

Bolting

Ethylene

Gas- C_2H_4

Promotes the ripening of fruit. Involved in stimulating the production of flowers. In combination with auxin, ethylene inhibits the elongation of roots, stems, and leaves, and influences leaf abscission, the aging and dropping of leaves.

Ethylene Plays a Role in Fruit Ripening

Abscisic acid (ABA)

Discovery: F.T. Addicott and his associates discovered abscisic acid in the early 1960s in the process of studying abscission in cotton. Bud Dormancy -- This is influenced by cytokinins and auxin-induced synthesis of ethylene.

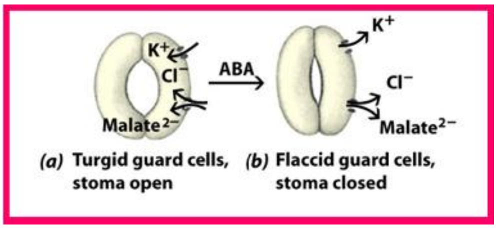
Abscisic Acid (ABA) Prevents Seed Germination

Seed Dormancy -- Abscisic acid delays seed germination in many plants. Has some effect on induction and maintenance of dormancy in general. Induces seeds to synthesize storage proteins. Inhibits the effect of gibberellins on stimulating de novo synthesis of a-amylase

Abscisic Acid Plays a Role as a Root-to-Shoot Signal

Biomass production by plants is regulated by water availability. A limited water supply in the soil is sensed by roots and communicated to the shoot to allow timely adjustment of the plant's transpiration. Abscisic acid plays a pivotal role in adjustment of plants to abiotic stress conditions abscisic acid (ABA) is the long-distance signal that communicates water stress from the root to the shoot.

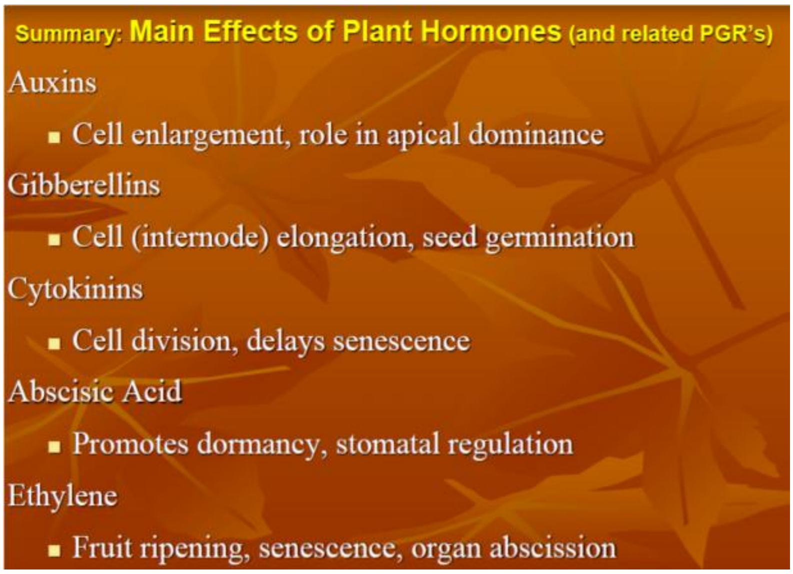
Abscisic Acid is Responsible for Stomatal Closure



(a) Turgid guard cells, stoma open (b) Flaccid guard cells, stoma closed

Closure of Stomata --

Large amounts of abscisic acid in the leaves causes the stomata to close which helps the plant conserve water during droughts. Reactions can be instigated within minutes of spraying. commercially it is used in fields when droughts threaten.



Summary: Main Effects of Plant Hormones (and related PGR's)

Auxins
- Cell enlargement, role in apical dominance

Gibberellins
- Cell (internode) elongation, seed germination

Cytokinins
- Cell division, delays senescence

Abscisic Acid
- Promotes dormancy, stomatal regulation

Ethylene
- Fruit ripening, senescence, organ abscission

Fill up the blanks

1) The term auxin was coined by ______
2) The precursor of indole acetic acid (natural auxin) is _______
3) _______ is the gaseous hormone
4) The rooting hormone is ______
5) The auxin which is used as weedicide is _______
6) _________ is synthetic auxin
7) Nodule formation is induced by _____
8) Bud initiation in shoots in promoted by_______
9) Bolting of some rosette plants is promoted by______
10) Regulators of plants growth are produced in ___________
11) Synthetic hormones used for ripening is _____
12) Combination of hormones which cause apical dominance and bolting in plants is ______ and _______
13) Maximum seed germination occurs when seeds are treated with ______
14) _________ is growth inhibiting hormones
15) Auxin promotes growth in plants by________
16) Plants synthesize auxin from the amino acid_______
17) This plant hormone promotes stem elongation and enzyme production in developing seeds is ____
18) _______ is produced by aging fruit
19) The hormone promotes lateral bud dormancy is _______
20) Plant hormone inhibits the effects of other hormones is ____
21) Plant hormone causing, abscission of leaves, senescence, bud dormancy and inhibition of cell division is _______
22) A dwarf plant can be induced to reach normal height by the application of: _______
23) Removal of the tip of a plant stimulates lateral growth AND the plant gets bushier, because the tip of a stem produces _______
24) Auxins are produced at or near the tips of roots and stems in areas called______
25) Any substance produced in one part of the plant that affects another part is a ________
26) A ripe Avocado will cause other avocados to ripen through the release of ______
27) Seedless fruits can be produced by the application of auxin or:_______
28) Extraordinary growth in the height of a plant would be caused by: _____
29) Substances that stimulate cell division and cause dormant seeds to sprout are:_____
30) Ethylene differ from other plant hormones as it is ____
31) A high concentration of auxins can inhibit plant growth. Many of these compounds could be used as ______
32) Plant hormone which saves the crops from falling is _____
33) Plant hormone which helps in formation of proteins and RNA is ____
34) The dormancy of the plant is broken by application of ________
35) Hormone which help in the cell division and development in the presence of auxin is ____

36) The hormone which increases the cambium activity in the wooden plants is _____

37) Apical dominance is affected by the hormone _________

38) Delay in senescence is caused by the spray of ________

39) Abscission is prevented by ______

40) High concentration of ethylene is present in ________

41) The precursor of ethylene is _______

42) All the climatic fruits except_____ respond to ethylene application

43) _______ hormone promotes release of ethylene

44) ABA act as antagonistic to _______

45) Hormones promote stem elongation and stimulate enzyme production in germinating seeds is ________

46) Plant hormones stimulate cell division in the presence of auxin, promote chloroplast development and bud formation, and delay leaf aging is ___

47) The site of gibberellic acid production in the plant is __________

48) At the cellular level, hormone important in triggering cell division is _______

49) _____ can be sprayed on peach trees in the spring to cause fruit or flowering thinning.

50) Sprouting of potatoes is inhibited by__________

51) _________ is referred to as stress hormone

52) Gibberellin is named after the fungus _______

ANSWER

Sl.No	Answer	Sl.No	Answer
1	F W Went	27	Gibberellins
2	Tryptophan	28	Auxin
3	Ethylene	29	Cytokinins
4	NAA	30	Gas
5	2,4 D	31	Herbicides
6	NAA, 2,4 D and 2,4,5 T	32	Auxin
7	IAA	33	Cytokinins
8	Auxin	34	Gibberellins
9	Gibberellins	35	Cytokinins
10	Phytohormones	36	Gibberellins
11	Ethephon	37	IAA
12	Auxin , Gibberellins	38	Abscisic Acid
13	Gibberellins	39	Ethylene
14	Abscisic acid and Ethylene	40	Ripening fruits
15	Stem elongation	41	Methionine
16	Tryptophan	42	Grapes
17	Gibberellins	43	Abscisic Acid
18	Ethylene	44	Gibberellic acid
19	Abscisic Acid:	45	Gibberellins
20	Abscisic Acid:	46	Cytokinins

21	Abscisic Acid:	47	Leaf
22	Auxin	48	Cytokinins
23	Auxin	49	Abscisic Acid:
24	Apical meristems	50	2,4 D
25	Hormone	51	Abscisic Acid:
26	Ethylene	52	Gibberella fujikoroi

Essay type

1, **Plant hormone or plant growth regulator**

Thimann (1948) designated the plant hormones by the term ‘phytohormones’ in order to distinguish them from animal hormones. Johannes van Overbeek (1950) defined plant hormones as “organic compounds which regulate plant physiological process regardless of whether these compounds are naturally occurring and/or synthetic ; stimulating and/or inhibitory ; local activators or substances which act at a distance from the place where they are formed.” Plant hormones, also known as phytohormones are chemicals that regulate plant growth. In plants, these are produced by cells in one area of the plant, such as leaves, stems or roots and then transported to a different area of the plant in order for them to have a response. Plant hormones shape the plant, affecting seed growth, time of flowering, the sex of flowers, senescence of leaves, and fruits. They affect which tissues grow upward and which grow downward, leaf formation and stem growth, fruit development and ripening, plant longevity, and even plant death. Hormones are vital to plant growth, and, lacking them, plants would be mostly a mass of undifferentiated cells. So they are also known as growth factors or growth hormones. Plants lack glands that produce and secrete hormones. Instead, each cell is capable of producing hormones. Hormones are transported within the plant using localised movement and cytoplasmic streaming within cells and slow diffusion. Phloem and Xylem are vascular tissues that also help in the transportation of hormones from one part of the plant to another. Plant hormones can be classified into five major categories, some of which are made up of many different chemicals that can vary in structure from one plant to the next. The classifications is based on chemical structural similarities and their effects on plant physiology. Each class has positive as well as inhibitory function and work in tandem with each other, interplaying to affect growth regulation. Abscisic acid, Auxins, Cytokinins, Ethylene and Gibberellins

2. **Role of Auxin in plants**

Kögl and Haagen-Smit (1931) introduced the term ‘auxin’ (auxeinG = to grow or to increase) for designating those plant hormones which are especially concerned with cell enlargement or the growth of the shoots. An auxin may, thus, be defined as “an organic substance which promotes growth (i.e., irreversible increase in growth) along the longitudinal axis when applied in low concentrations to shoots of the plants freed as far as practicable from their own inherit growth promoting substances. Auxin is a plant phytohormone involved in practically every dimension of plant development, including responses to light and gravity, organ patterning, vascular development and regulating intrinsic growth and environmental responses in both shoot and root architecture. The auxin in plant shoots cause them to grow away from gravity or upwards, while the auxins in roots cause them to grow towards gravity or downwards. There are four auxins that exist in nature and are synthesized by plants. Since their discovery, more auxins have been derived from existing ones and others have been synthesized in the laboratory. The most prevalent of the natural auxins is indole-3-acetic acid (IAA), which is produced by algae, plants, bacteria, and fungi. IAA thickens the cambium layer of plants by actually enlarging xylem cells. Another agriculturally significant auxin is indole-3-butyric acid (IBA), a synthetic form of which is used in a variety of products available to boost propagation rates. If you have used a rooting hormone products on your cuttings to make clones, it is highly likely you were using IBA to initiate adventitious root production.

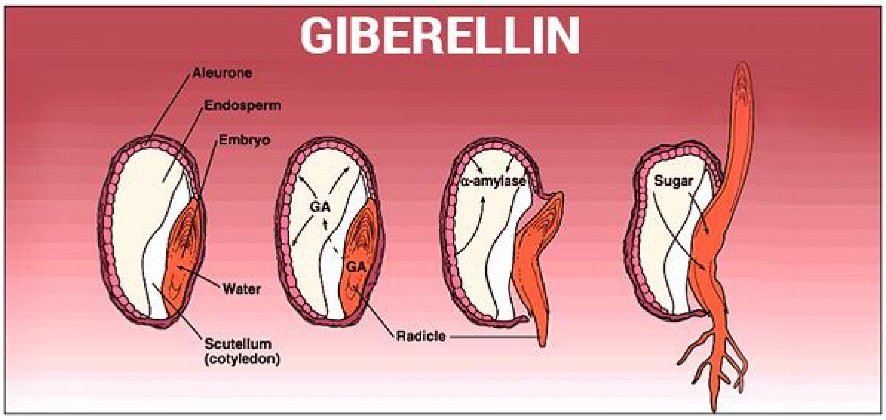
Functions

1) **Eradication of weeds**: Auxins are being used as weedicides mainly due to their selective herbicidal nature ,toxic residues of auxins disappear from soil very soon.2,4-D is used to destroy broad leaved divots(mostly weeds).However auxins like 2,4-D and 2,4,5-T are nontoxic to narrow leaved plants i.e monocots.
2) **Root initiation**: Application of auxins activates root initials. Rapid adventitious root formation is absolutely essential in cuttings for their successful development into new plants in vegetative propagation. Auxins like IAA, IBA, NAA,2,4-D when applied in low concentration from 10–1000ppm on cuttings initiate adventitious root formation.
3) **Flower initiation**: Auxins normally inhibit flowering. However in litchi and pineapple (Annanus sativus) auxins like 2, 4 -D and NAA have been found to promote flowering.
4) **Production of parthenocarpic fruits**: The conversion of ovary into fruit had been believed mainly due to the activity of auxins.
5) **Apical dominance and dormancy**: Auxins recommended for increasing the dormancy period in potato tubers is methyl ester of naphthalene acetic acid.
6) **Enzymatic activity**: the auxins stimulate the activity of enzymes (conjugated with aspartic acid).They show inhibitory effect on isoenzymes like peroxidase in tobacco.
7) To prevent premature fruit drop: Auxins have been successfully employed for checking premature fall of fruits because their application prevent the formation of abscission layer

3) Role of gibberellin on plants

The gibberellins are weakly acidic phytohormones which help in longitudinal growth of stem. .In 1926, a plant pathologist Kurosawa, discovered the causative agent of the disease (bakanae disease) as a fungus Gibberella fujikoroi. Yamuta and Sumuki (1938) demonstrated that some substance secreted by this fungus was responsible for the elongation and they later isolated this substance and named it gibberellin A3 or gibberellic acid (GA). There are more than 70 gibberellins isolated. They are GA1, GA2, GA3 and so on. The GA3 Gibberellic acid is the most widely studied plant growth

Gibberellins are chemically different from auxins, in that they contain gibbane ring system with specific biological properties
Gibberellins are the plant growth regulators involved in regulating the growth and influencing different developmental processes which include stem elongation, germination, flowering, enzyme induction, etc.



Functions of Gibberellins

Stem elongation:

Gibberellins cause stem elongation and leaf expansion. It is believed that certain types of dwarfness are due to gibberellin deficiency. But it has no effect on roots.

Bolting

Gibberellin induces stem elongation in rosette plants. Cabbage is a rosette plant with profuse leaf growth and retarded intermodal length. Just prior to flowering, internodes elongate enormously. This is called bolting. Bolting can be induced artificially by the application of gibberellins under normal conditions.

Seed Germination

Some seeds that are sensitive to light such as tobacco and lettuce exhibit poor germination in the absence of sunlight. Germination begins rapidly if the seeds are exposed to the sunlight. If the seeds are treated with gibberellic acid, the light requirement can be overcome

Breaking of seed dormancy

Gibberellins break dormancy of buds and tubers. But in root tubers it inhibits the development of the root tuber.

Parthenocarpy

Gibberellins cause parthenocarpic in apple and pear.

Increasing Fruit Size

Gibberellins along with auxin, control the growth and development of fruits.

Flowering and sex expression

Gibberellins control flowering in long day plants. Gibberellins promote the production of male flowers, either in place of female flowers in monoecious plants or in genetically female plants such as cucurbits.

Fruit growth and parthenocarpy

Increased yield (larger size) and better shape of grapes is obtained by treating the fruit bunches with GA. It induces parthenocarpy in apples, pears, tomatoes and cucumbers.

Delayed ripening

Gibberellins delay fruit maturity and senescence in lemons, oranges and cherries. This helps in storing the fruits.

Flowering

Gibberellins help in the flowering of many long day plants

4. **Role of Cytokinins in plants**

Cytokinins are a group of hormones that promote cell division in plant roots and shoots and the growth of buds. These hormones have been found in all complex plants as well as mosses, fungi, and bacteria. There are about 200 different natural and synthetic cytokinins known to botanists today. Most cytokinins are produced in the meristem of the roots. Meristem is the name for a region of tissue within the plant that actively promotes cell division. In other words, the meristem is any place that's still growing (like the tip of the roots or the top of the stem). Once the cytokinins has been produced in the roots, it travels up the xylem, or vascular tissue, to other parts of the plant where continued growth takes place (such as young leaves, developing fruits, and seeds). Cytokinins increase cell division by stimulating the production of proteins needed for mitosis. Cytokinins help in cytokinesis

Functions of Cytokinins:

1. **Cell Division:**
   Cytokinins are essential for cytokinesis though chromosome dou- bling can occur in their absence. In the presence of auxin, cytokinins bring about division even in permanent cells.
2. **Cell Elongation:**
   Like auxin and gibberellins, cytokinins also cause cell elongation.
3. **Morphogenesis**:
   Both auxin and cytokinins are essential for morphogenesis or dif- ferentiation of tissues and organs. Buds develop when cytokinins are in excess while roots are formed when their ratios are reversed
4. **Differentiation**:
   Cytokinins induce formation of new leaves, chloroplasts in leaves, lateral shoot formation and adventitious shoot formation. They also bring about lignification and differentiation of inter-fascicular cambium
5. **Senescence (Richmond-Lang Effect):**
   Cytokinins delay the senescence of leaves and other organs by mobilisation of nutrients
6. **Apical Dominance**:
   Presence of cytokinins in an area causes preferential movement of nutrients towards it. When applied to lateral buds, they help in their growth despite the presence of apical bud. They thus act antagonistically to auxin which promotes apical dominance.

7. **Seed Dormancy**:
   Like gibberellins, they overcome seed dormancy of various types, including red light requirement of Lettuce and Tobacco seeds.
8. **Resistance:**
   Cytokinins increase resistance to high or low temperature and disease.
9. **Phloem Transport**:
   They help in phloem transport.
10. **Accumulation of Salts:**
    Cytokinins induce accumulation of salts inside the cells.
11. **Flowering:**
    Cytokinins can replace photoperiodic requirement of flowering in certain cases.
12. **Sex Expression**:
    Like auxins and ethylene, cytokinins promote femaleness in flowers.
13. **Parthenocarpy:**
    Crane (1965) has reported induction of parthenocarpy through cytokinins treatment

5. **Role of Abscisic acid (ABA) in plants**

Abscisic acid (ABA) is often referred to as a inhibitory rather than stimulatory hormone. It is involved in the closure of stomata, bud and seed dormancy and is known to inhibit other hormonal actions. F.T. Addicott and his associates discovered abscisic acid in the early 1960s in the process of studying abscission in cotton. Abscisic acid owes its names to its role in the abscission of plant leaves. Plants also have hormones that stimulate processes that are necessary for them to live. Abscisic acid is a plant hormone involved in many developmental plant processes, such as dormancy and environmental stress response. Abscisic acid is produced in the roots of the plant as well as the terminal buds at the top of the plant. It is also called as stress hormone because the production of hormone is stimulated by drought, water logging and other adverse environmental conditions. Abscisic acid is known as dormin as it induces dormancy in buds, underground stems and seeds. Abscisic acid is a mildly acidic dextrorotatory cis sesquiterpenes growth hormone.

Functions of Abscisic Acid:

1. **Bud Dormancy**
   Abscisic acid induces dormancy of buds towards the approach of winter.
2. **Seed Dormancy**
   It is mainly caused by abscisic acid. Dormancy allows seeds to tolerate desiccation and extremes of temperature better. The buds as well as seeds sprout only when abscisic acid is overcome by gibberellins. Because of its action in inducing dormancy, abscisic acid or ABA is also named as dormin.
3. **Stoppage of Cambium Activity**
   Formation of abscisic acid stops mitosis in vascu- lar cambium towards the approach of winter.
4. **Abscission**
   Abscisic acid promotes abscission of flowers and fruits.

5. **Leaf Senescence**

Its excessive presence stops protein and RNA synthesis in the leaves and hence stimulates their senescence (leaf fall is actually promoted by ethylene)

6. **Transpiration**

During desiccation and other stresses, abscisic acid is rapidly synthesised. The inhibitor causes closure of stomata and hence prevents transpiration.

7**. Resistance**

Abscisic acid increases resistance of plants to cold and other types of stresses. It is, therefore, also known as stress hormone.

8. **Starch Hydrolysis**

Abscisic acid inhibits gibberellin mediated amylase formation during germination of cereal grains.

9. **Flowering:**

In small quantities, Abscisic acid is known to promote flowering in some short day plants, e.g., Strawberry, Black Currant.

10. **Parthenocarpy**:

ABA has been found to induce parthenocarpic development in Rose.

11. **Rooting:**

Rooting of stem cuttings is promoted in some cases by abscisic acid, e.g., Bean, Ivy, Poinsettia (= Euphorbia pulcherrima)

12. **Membrane Potential**:

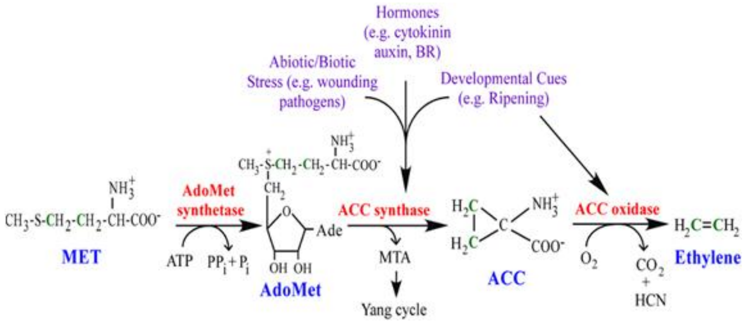
ABA induces a positive surface potential on cell membrane.

13. **Controlled Growth:**

It is antagonist to gibberellins and counteracts the effect of other growth promoting hormones (auxins and cytokinins) and therefore, keeps their activity under check. By controlling growth, ABA plays an important role in seed development and seed maturation. Normally it inhibits seed germination, growth of excised embryos, growth of Duckweed and other plants.

6. Role of ethylene in plants

Ethylene (ripening hormone), unlike the rest of the plant hormone compounds is a gaseous hormone. Ethylene is produced in all higher plants and is produced from methionine in essentially all tissues. Production of ethylene varies with the type of tissue, the plant species, and also the stage of development. The mechanism by which ethylene is produced from methionine is a 3 step process. Plants synthesize ethylene in response to stress to drought stress, heat stress, biotic stress, wounding and flooding



In 1901, Dimitry Neljubov observed that dark grown pea seedlings in the laboratory exhibited symptoms that were later termed the triple response. The first indication that ethylene is a natural product of plant tissues was published by H. H. Cousins in 1910. In 1934, R. Gane and others identified ethylene chemically as a natural product of plant metabolism, and because of its dramatic effects on plant it was classified as a hormone.

Ethylene can easily be synthesized in all plant organs such as roots, stems, leaves, tubers, fruits and seeds. It is highest in senescing tissues and ripening fruits. Within the plant organs, ethylene formation is mainly located in peripheral tissues. Ethylene is biologically active at low concentration (less than 1 ppm). Ethylene can easily pass through plasma membrane into the cell, easily diffuse within the plant, and flushed out of plant tissues through intercellular spaces

Ethylene is regarded as a multifunctional phytohormone that regulates both growth, and senescence. It promotes or inhibits growth and senescence processes depending on its concentration, timing of application, and the plant species. Often considered an 'aging' hormone due to its role in accelerating such developmental processes as ripening, senescence, and abscission, Ethylene will cause a wide range of effects in plants, depending on the age of the plant and how sensitive the plant is to ethylene.

Functions of Ethylene:

1) **Growth:**

   Ethylene inhibits longitudinal growth but stimulates transverse or horizontal growth and swelling of axis

2) **Senescence:**

   It hastens the senescence of leaves and flowers

3) **Abscission**:

   Abscission of various parts (leaves, flowers, fruits) is stimulated by ethylene which induces the formation of hydrolases.

4) **Apical Dominance:**

   Ethylene promotes apical dominance and prolongs dormancy of lateral buds

5) **Breaking of Dormancy**:

   It breaks the dormancy of buds, seeds and storage organs.

6) **Root Initiation**:

   In low concentration ethylene helps in root initiation, growth of lateral roots and root hairs. This increases the absorption surface of the plant roots.

7) **Fruit Ripening:**

   It aids in ripening of climacteric fruits and dehiscence of dry fruits. Climacteric fruits are fleshy fruits which show a sudden sharp rise of respiration rate at the time of ripening (respiratory climacteric). They are usually transported in green or unripe stage. Ethylene is used to induce artificial ripening of these fruits, e.g., Apple, Mango, Banana, etc.

8) **Flowering:**

   It stimulates flowering in Pineapple and related plants as well as mango though in other cases the gaseous hormone causes fading of flowers. This helps in synchronizing fruit set.

9) **Sex Expression**:

   Like auxins and cytokinins, ethylene has a feminizing effect on sex expression. The genetically male plants of Cannabis can be induced to produce female flowers in the presence of ethylene. The number of female flowers and hence fruit is enhanced in monoecious plants like Cucumber.

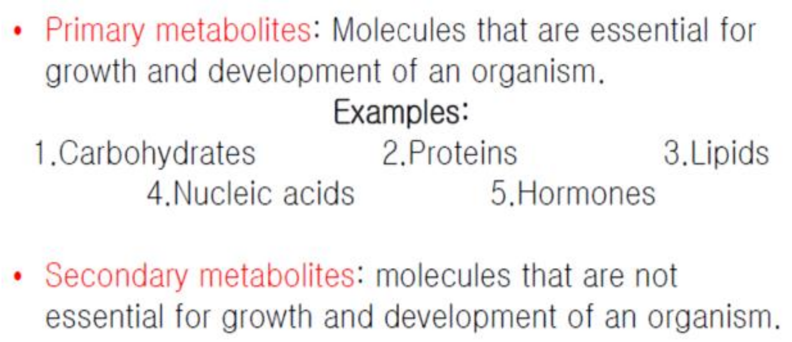
10) **Epinasty** (bending of stems)

Definitions

1. Plant Hormones: Plant hormones (also known as phytohormones) are chemicals that regulate plant growth. (or) Any of various hormones produced by plants that control or regulate germination, growth, metabolism, or other physiological activities
2. Auxin: A plant hormone which causes the elongation of cells in shoots and is involved in regulating plant growth.
3. Gibberellins: Any of a group of plant hormones that stimulate stem elongation, germination, and flowering.
4. Cytokinins: any of a class of plant hormones, produced by the roots and traveling upward through the xylem, that promote tissue growth and budding and, on application, retard plant senescence. Abscisic acid: Abscisic acid is a plant hormone involved in many developmental plant processes, such as dormancy and environmental stress response.
5. Ethylene: Ethylene is a regulator of seed germination, seedling growth, leaf and petal abscission, organ senescence, stress responses, and pathogen responses.
6. Growth retardants: Natural growth inhibitors are regulating substances which retard such processes as root and stem elongation, seed germination, and bud opening.

SECONDARY METABOLITES

Metabolites

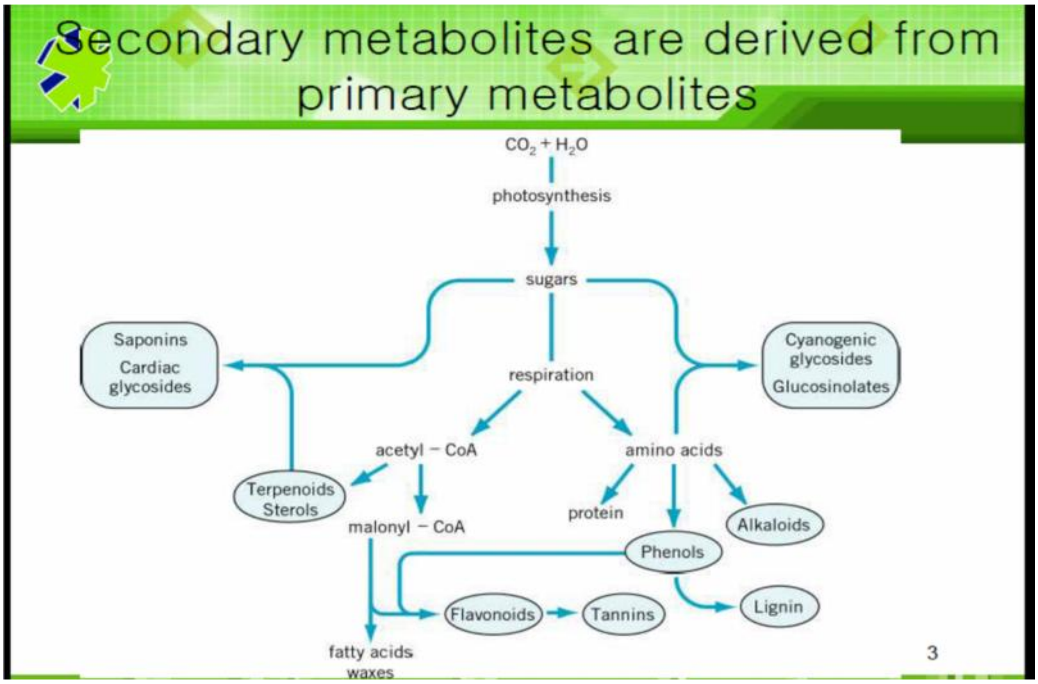


- Primary metabolites: Molecules that are essential for growth and development of an organism.

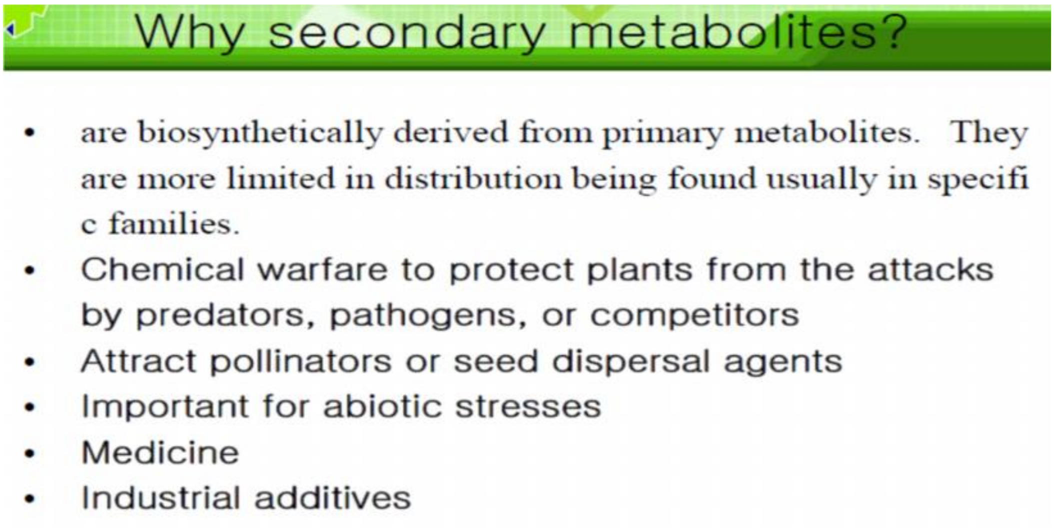
Examples:

1.Carbohydrates 2.Proteins 3.Lipids
4.Nucleic acids 5.Hormones

- Secondary metabolites: molecules that are not essential for growth and development of an organism.



Secondary metabolites are derived from primary metabolites



Why secondary metabolites?

- are biosynthetically derived from primary metabolites. They are more limited in distribution being found usually in specific families.
- Chemical warfare to protect plants from the attacks by predators, pathogens, or competitors
- Attract pollinators or seed dispersal agents
- Important for abiotic stresses
- Medicine
- Industrial additives

Organic compounds produced by the plants which have no direct role in the growth and development are called as secondary metabolites.

These secondary compounds produced by plants are grouped into five major groups.

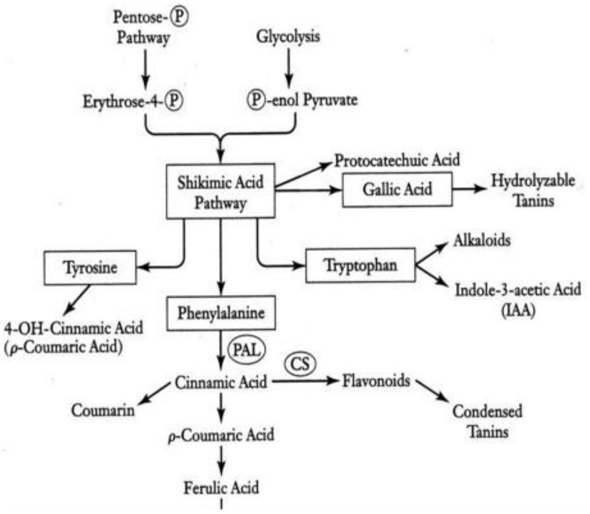
1. Phenolics
2. Terpenoids
3. Alkaloids
4. Special nitrogen metabolites
5. Cuticular compounds

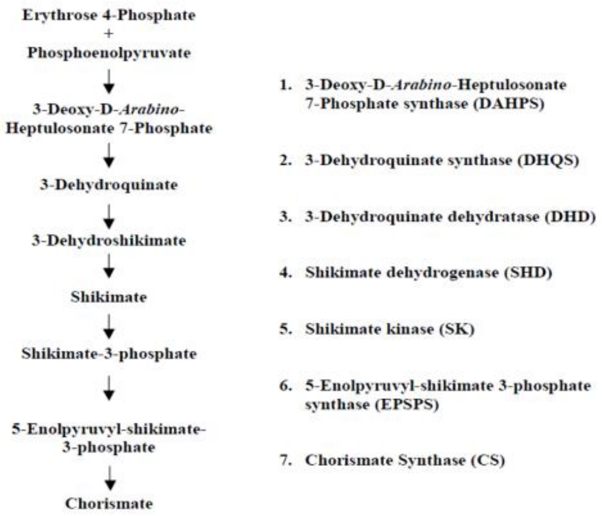
Phenolics

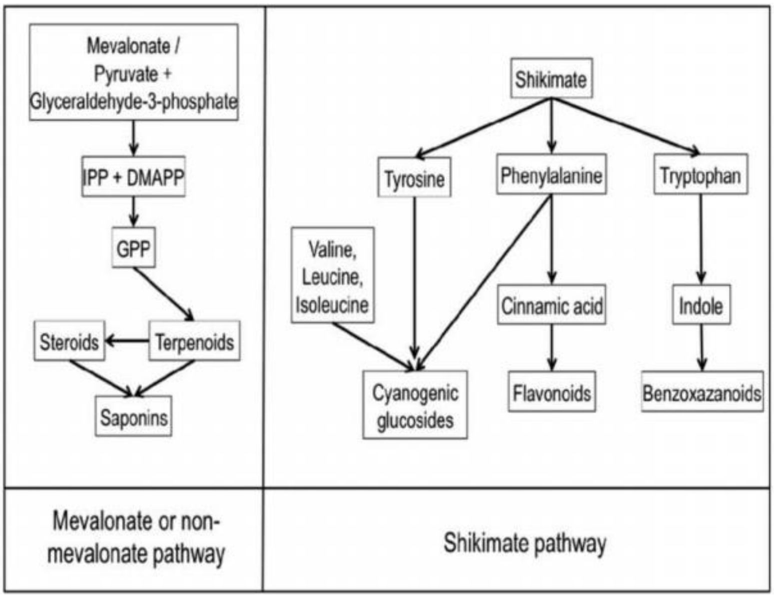
Phenol

Phenolics are a group of compounds characterized by at least one aromatic ring bearing one or more hydroxyl groups. Phenolic compounds from plants are one of largest group of secondary plants constituents synthesized by fruits, vegetables, teas, cocoa and other plants that possess certain health benefits. They are characterized by the antioxidant, anti-inflammatory, anti-carcinogenic and other biological properties, and may protect from oxidative stress and some diseases.. Phenolics range from simple, low molecular-weight, single aromatic-ringed compounds to large and complex tannins and derived polyphenols. They can be classified based on the number and arrangement of their carbon atoms and are commonly found conjugated to sugars and organic acids. Phenolics can be classified into two groups: the flavonoids and the non-flavonoids. Flavonoids are polyphenolic compounds comprising fifteen carbons, with two aromatic rings connected by a three-carbon bridge. They are the most numerous of the phenolics and are found throughout the plant kingdom. They are present in high concentrations in the epidermis of leaves and the skin of fruits and have important and varied roles as secondary metabolites. Plants produce a large variety of secondary compounds containing a phenol group. Most of the thousands of phenolics known to date are of plant origin. These phenolic compounds are synthesized via two different routes: the shikimate pathway and the acetate-malonate pathway, and thus represent a heterogeneous group. The shikimate pathway participates in the synthesis of most plant phenolics, whereas the malonate pathway is of less significance in higher plants, although it is an important source of phenolic products in fungi and bacteria.

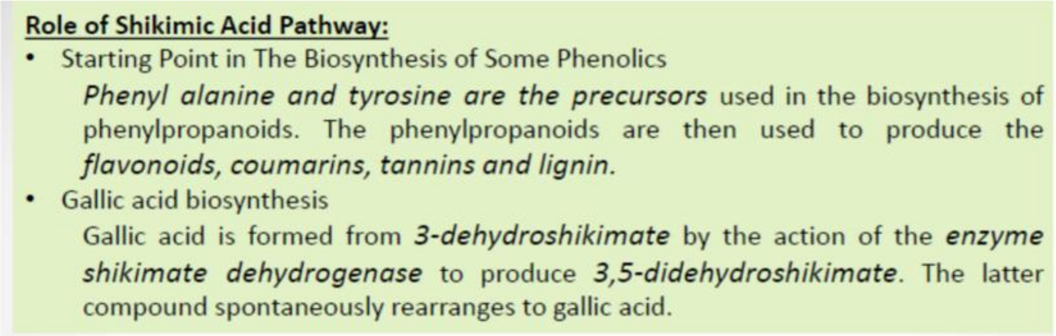
The shikimate pathway is defined as seven metabolic steps beginning with the condensation of phosphoenolpyruvate (PEP) and Erythrose 4-phosphate (Ery4P) and ending with the synthesis of chorismate. It is the common route leading to the production of three aromatic amino acids: phenylalanine (Phe), tyrosine (Tyr) and tryptophan (Trp).Shikimic acid is a tri-hydroxy cyclohexene carboxylic acid important in biosynthesis of so many compounds that the shikimate pathway is named after it. Shikimic acid, more commonly known as its anionic form shikimate, is an important biochemical intermediate in plants and microorganisms







The key starting materials are phosphoenolpyruvate (PEP) and Erythrose 4P derived from glycolysis and pentose phosphate pathways, respectively. These two compounds condense to produce a six carbon cyclic compound with one carbon (COOH) side chain namely shikimate. Then shikimate is phosphorylated and condensed with another molecule of PEP to produce a cyclic compound containing a three carbon and one carbon side chains. This is finally converted to aromatic amino acids phenylalanine and tyrosine. These amino acids are deaminated followed by hydroxylation at different carbon atoms in the aromatic ring to form cinnamic acid derivatives. These cinnamic acid derivatives are utilised for the synthesis of different phenolic compounds.

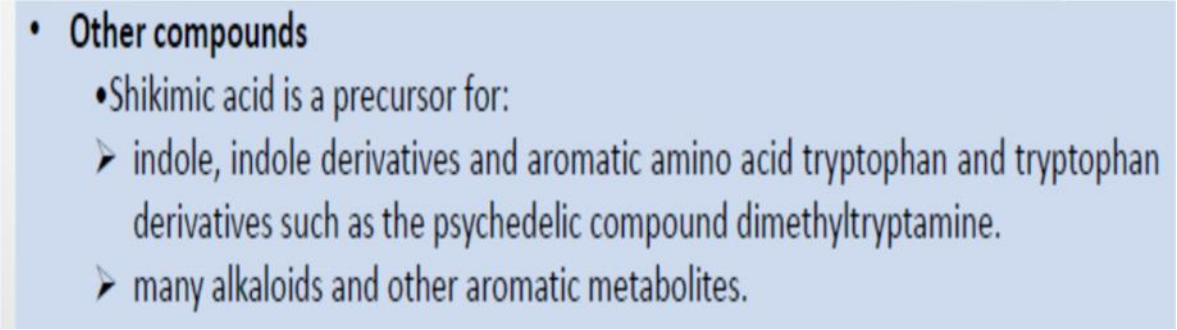


Role of Shikimic Acid Pathway:

- Starting Point in The Biosynthesis of Some Phenolics

  Phenyl alanine and tyrosine are the precursors used in the biosynthesis of phenylpropanoids. The phenylpropanoids are then used to produce the *flavonoids, coumarins, tannins and lignin.*
- Gallic acid biosynthesis

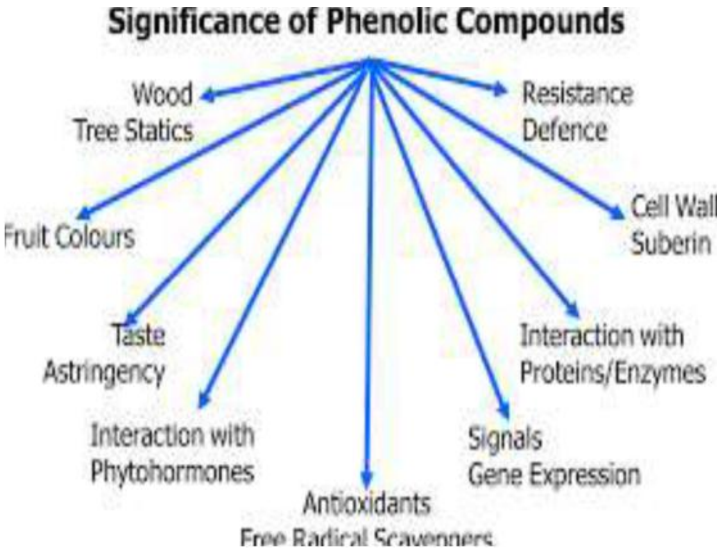
  Gallic acid is formed from ***3-dehydroshikimate*** by the action of the ***enzyme shikimate dehydrogenase*** to produce ***3,5-didehydroshikimate***. The latter compound spontaneously rearranges to gallic acid.



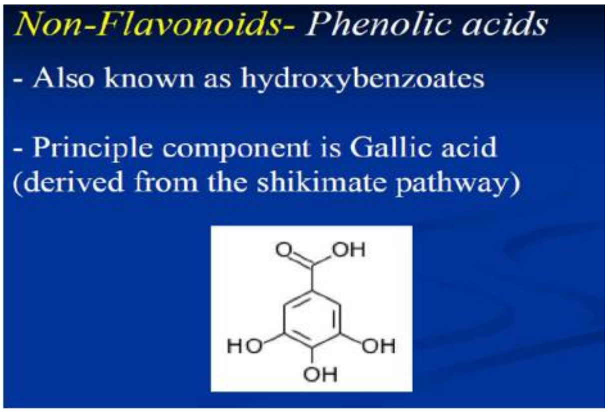
- Other compounds
  - Shikimic acid is a precursor for:
    - indole, indole derivatives and aromatic amino acid tryptophan and tryptophan derivatives such as the psychedelic compound dimethyltryptamine.
    - many alkaloids and other aromatic metabolites.

Functions of phenolics

Phenolics are of great importance as cell wall components. Cell wall structures such as lignins, cutins and suberins, which provide mechanical support and function as barriers against microbial attack. The flavonoids and anthocyanins contribute to flower and fruit colours. Attracting insects and animals to the plant for pollination and seed dispersal Phenolics also play a defensive role in plants by protecting against predators. Tannins and phenolic resins at the plant surface are effective feeding deterrents. Phenolic compounds also produce allelopathic effect. Phenolics also function as signal molecules in the interaction between nitrogen fixing bacteria and leguminous plants. Phenolic compounds function as effective antioxidants. Polyphenolics are important in foodstuffs, wines and herbal teas because of their astringent taste . Polyphenolics were used as tanning agents in leather industries. Anthocyanins have considerable potential in the food industry as safe and effective food additives



Significance of Phenolic Compounds



Non-Flavonoids- Phenolic acids

- Also known as hydroxybenzoates

- Principle component is Gallic acid (derived from the shikimate pathway)

What Are Alkaloids?

These are commonly applied to basic nitrogenous compounds of plant origin that are physiologically active

Characteristics

They are bitter in taste. Derived from amino acids. The amino acids that are most often serve as alkaloidal precursors are: phenylalanine, tyrosine, tryptophan, histidine, anthranilic acid, lysine and ornithine. Alkaloids form double salts with compounds of mercury, gold, platinum and other heavy metals. These salts are obtained as precipitate which are microcrystals. Insoluble or sparingly soluble in water, but the salts formed on reaction with acids are usually freely soluble. Most are crystalline solids although a few are amorphous. Alkaloids constitute a very large group of secondary metabolites, with more than 12,000 substances isolated. A huge variety of structural formulas, coming from different biosynthetic pathways. Alkaloids are produced by a large variety of organisms, such as bacteria, fungi, animals but mostly by plants as secondary metabolites. Most of them are toxic to other organisms and can be extracted by acid-base. The term alkaloid was coined in 1819 by the pharmacist W. Meisner and meant simply, alkali like.

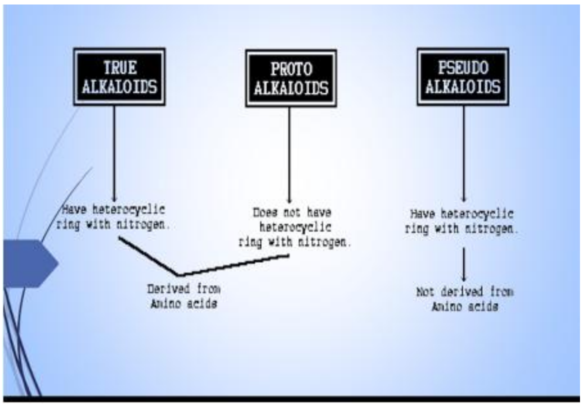
Sources and Occurrence of Alkaloids

Alkaloids can occur in plant kingdoms; among the angiosperms,

• Leguminosae, • Papaveraceae, • Ranunculaceae, • Rubiaceae, • Solanaceae,

• Berberidaceae are outstanding alkaloid-yielding plants

Types of Alkaloids



TRUE ALKALOIDS

PROTO ALKALOIDS

PSEUDO ALKALOIDS

Have heterocyclic ring with nitrogen.

Does not have heterocyclic ring with nitrogen.

Have heterocyclic ring with nitrogen.

Derived from Amino acids

Not derived from Amino acids

True Alkoloids

True alkoloids derived from amino acids. Heterocyclic ring with nitrogen. Highly reactive substances in low doses also. Bitter taste with white appearance. Form water soluble salts examples: cocaine, morphine, nicotine, dopamine etc

Proto-Alkaloid

General Characteristics Features:

- Have no nitrogen as part of heterocyclic ring
- Derived from amino acids like Phenylalanine, Tyrosine
- Physiologically active compounds
- Examples: Adrenaline, Ephedrine, Colchicines, Mescaline

Pseudo Alkaloids

Alkaloid-like compounds that do not originate from amino acids. This group includes terpene-like and steroid-like alkaloids, as well as purine-like alkaloids such as caffeine, theobromine, theacrine and theophylline. These do not show many of the typical characters of alkaloids but give the standard qualitative tests for alkaloid.

Functions

- They may act as protective against insects and herbivores due to their bitterness and toxicity.
- They are, in certain cases, the final products of detoxification (waste products).
- Source of nitrogen in case of nitrogen deficiency
- They, sometimes, act as growth regulators in certain metabolic systems.
- They may be utilized as a source of energy in case of deficiency in carbon dioxide assimilation
- Muscle relaxant, Pain killers, tranquilizers, Mind altering drugs, Chemotherapy

TERPENOIDS

Terpenoids are hydrocarbons of plant origin of the general formula $(C_5H_8)n$ as well as their oxygenated, hydrogenated and dehydrogenated derivatives.

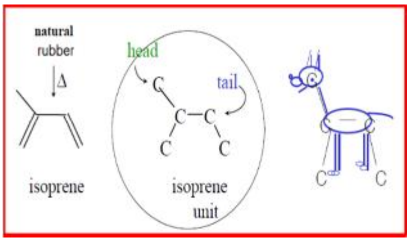
Terpenes - class of >20,000 compounds containing carbon atoms in multiples of five

Terpenoids - oxygen-containing terpenes (alcohols, ketones, aldehydes)

The name "terpene" is derived from the word "turpentine"

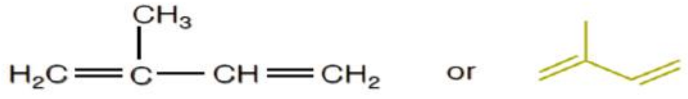
Terpenes and terpenoids are the primary constituents of the essential oils of many types of plants and flowers.

The chemist Leopold Ruziicka (born 1887) showed that many compounds found in nature were formed from multiples of five carbons arranged in the same pattern as an isoprene molecule (obtained by pyrolysis of natural rubber).

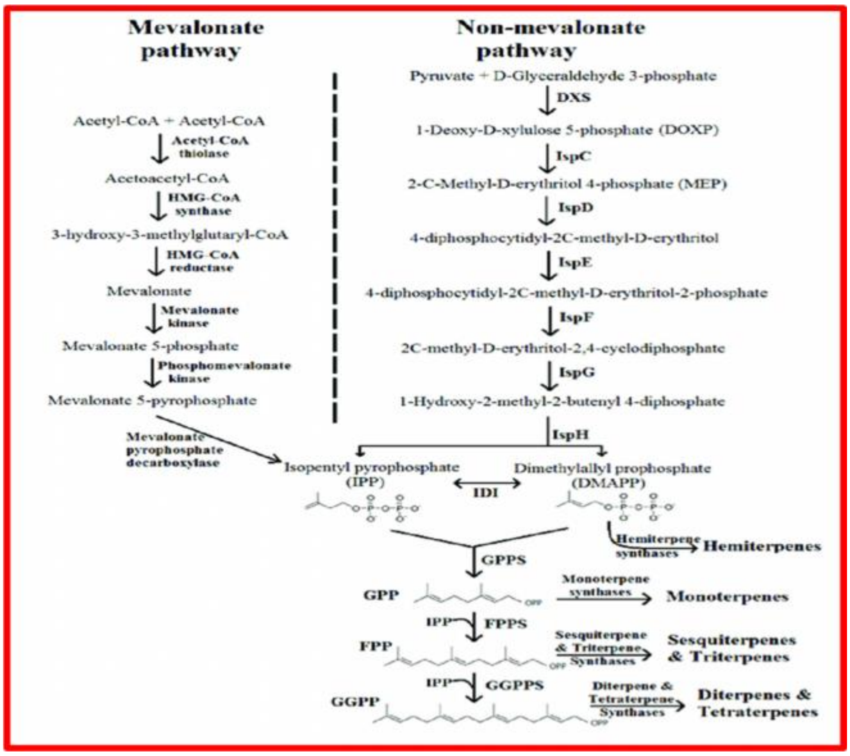


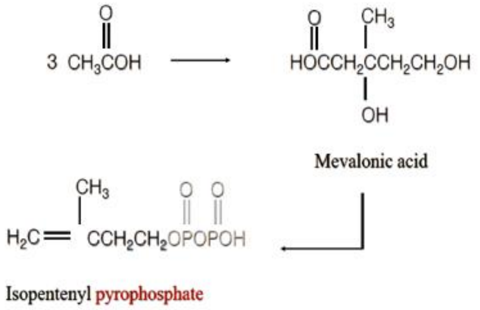
He called these compounds “terpenes”.

- The isoprene units in terpenes do not come from isoprene.
- They come from isopentenyl pyrophosphate.
- Isopentenyl pyrophosphate (5 carbons) comes from acetate (2 carbons) via mevalonate (6 carbons).
  - Terpenes are natural products that are structurally related to isoprene.


$H_2C{=}C(CH_3){-}CH{=}CH_2$

Isoprene
(2-methyl-1,3-butadiene)





ISOPRENE C5 is the basic unit of the terpenoids
Production in Plants: * Flowers * Leaves * Fruit

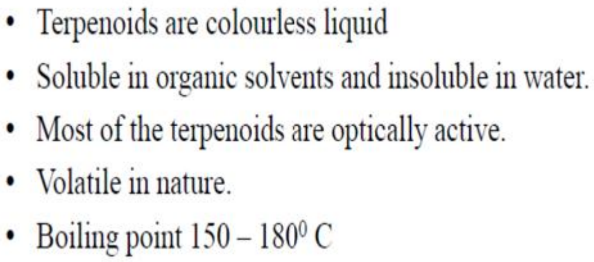
Examples of terpenoids

Geraniol, Citronellol, Farnesol

Limonene, Citral

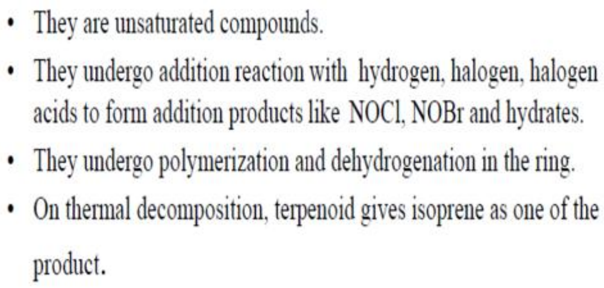
Eugenol

Physical properties



- Terpenoids are colourless liquid
- Soluble in organic solvents and insoluble in water.
- Most of the terpenoids are optically active.
- Volatile in nature.
- Boiling point 150 – 180^0 C

Chemical properties

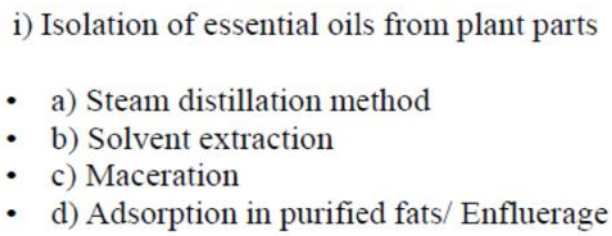


- They are unsaturated compounds.
- They undergo addition reaction with hydrogen, halogen, halogen acids to form addition products like NOCl, NOBr and hydrates.
- They undergo polymerization and dehydrogenation in the ring.
- On thermal decomposition, terpenoid gives isoprene as one of the product.

Biological Role (volatile and non-volatile)

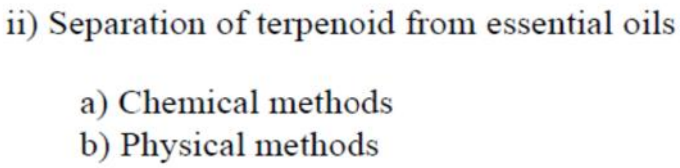
- Flavour, fragrance, scent
- Antibiotics
- Hormones
- Membrane lipids
- Insect attractants
- Insect antifeedants

Isolation of terpenoids



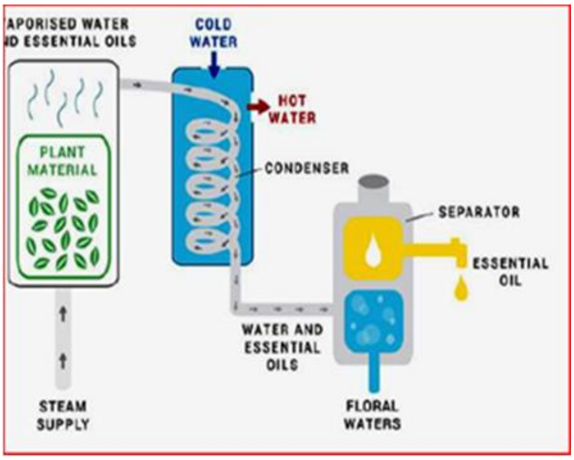
i) Isolation of essential oils from plant parts

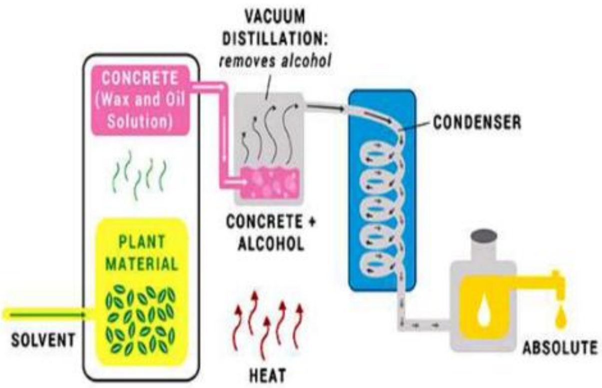
- a) Steam distillation method
- b) Solvent extraction
- c) Maceration
- d) Adsorption in purified fats/ Enfluerage

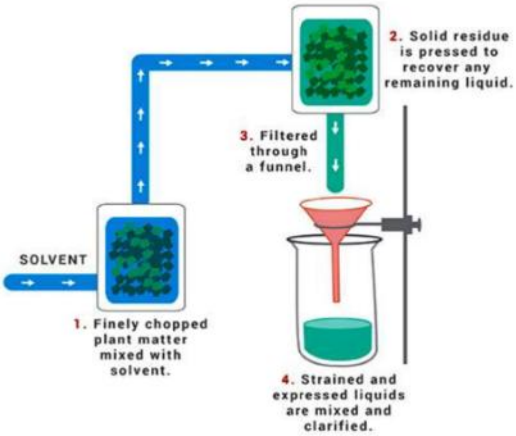


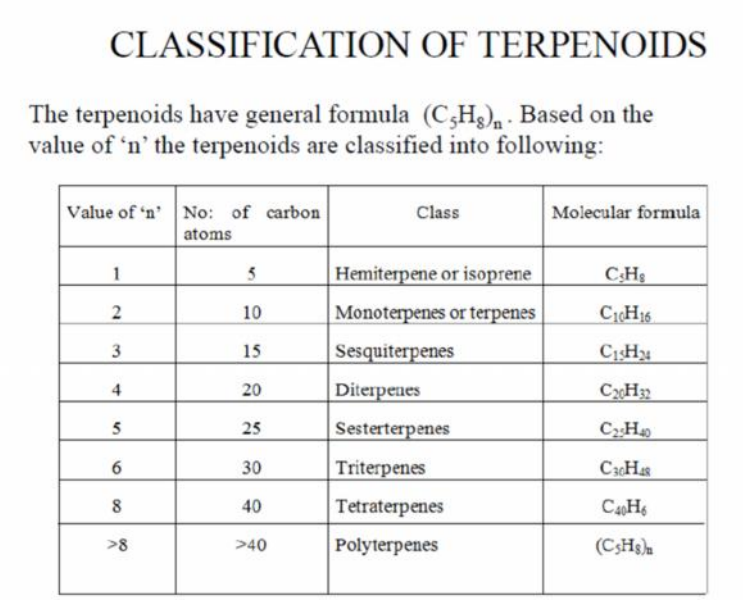
ii) Separation of terpenoid from essential oils

a) Chemical methods
b) Physical methods





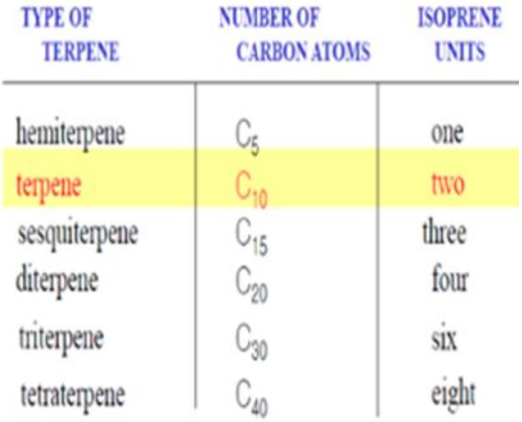




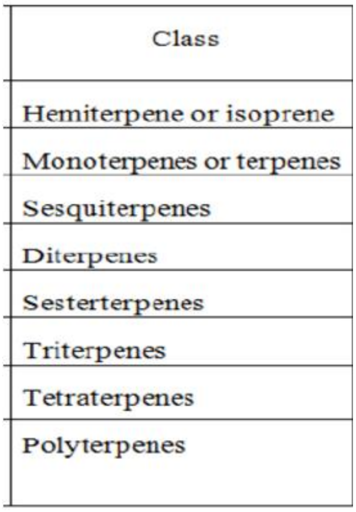
CLASSIFICATION OF TERPENOIDS

The terpenoids have general formula $(C_5H_8)_n$. Based on the value of ‘n’ the terpenoids are classified into following:

Value of ‘n’	No: of carbon atoms	Class	Molecular formula
1	5	Hemiterpene or isoprene	C_5H_8
2	10	Monoterpenes or terpenes	$C_{10}H_{16}$
3	15	Sesquiterpenes	$C_{15}H_{24}$
4	20	Diterpenes	$C_{20}H_{32}$
5	25	Sesterterpenes	$C_{25}H_{40}$
6	30	Triterpenes	$C_{30}H_{48}$
8	40	Tetraterpenes	$C_{40}H_6$
>8	>40	Polyterpenes	$(C_5H_8)_n$



TYPE OF TERPENE	NUMBER OF CARBON ATOMS	ISOPRENE UNITS
hemiterpene	C_5	one
terpene	C_{10}	two
sesquiterpene	C_{15}	three
diterpene	C_{20}	four
triterpene	C_{30}	six
tetraterpene	C_{40}	eight

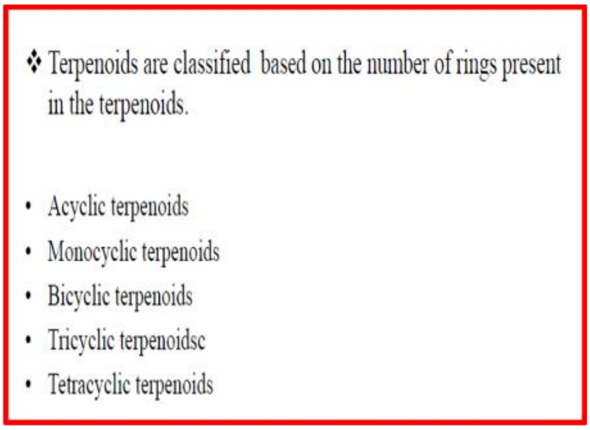


Class
Hemiterpene or isoprene
Monoterpenes or terpenes
Sesquiterpenes
Diterpenes
Sesterterpenes
Triterpenes
Tetraterpenes
Polyterpenes

Menthol, camphor, eugenol
Pheromones, Lemon grass, citral

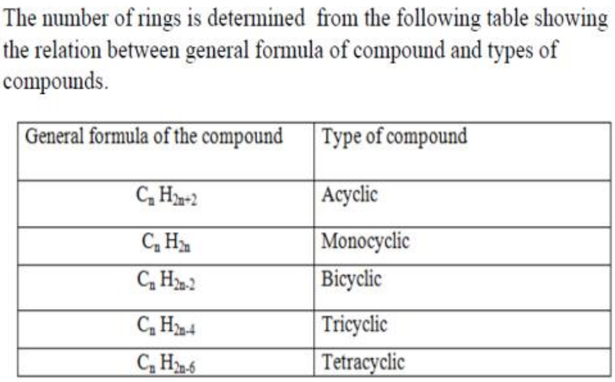
Vitamin A

Carotenoids
Rubber



❖ Terpenoids are classified based on the number of rings present in the terpenoids.

- Acyclic terpenoids
- Monocyclic terpenoids
- Bicyclic terpenoids
- Tricyclic terpenoidsc
- Tetracyclic terpenoids



The number of rings is determined from the following table showing the relation between general formula of compound and types of compounds.

General formula of the compound	Type of compound
$C_n H_{2n+2}$	Acyclic
$C_n H_{2n}$	Monocyclic
$C_n H_{2n-2}$	Bicyclic
$C_n H_{2n-4}$	Tricyclic
$C_n H_{2n-6}$	Tetracyclic

Functions

a. Plants employ Terpenoid metabolites for a variety of basic functions in growth and development, majority of terpenoids for more specialized chemical interactions and protection in the abiotic and biotic environment.

b. Traditionally, plant-based terpenoids have been used by humans in the food, pharmaceutical, and chemical industries, and more recently have been exploited in the development of biofuel products.

c. Plants have direct and indirect defence responses when they are attacked by herbivores or infected by fungal and bacterial pathogens. Direct defences include physical structures, such as trichomes and thorns, and the accumulation of chemical or biochemical compounds that have antibiotic activities or toxicities

d. Phytoalexins are low-molecular-weight compounds that are produced as part of the plant defence system. In many plant species diterpenes and sesquiterpenes act as phytoalexins.

Fill up the blanks

1. Molecules found in all plant cells and are necessary for the life of the plant is ____
2. Created through the mevalonic acid pathway; they are composed of isoprene units is ____

3. Created through the shikimic acid pathway; it contains a hydroxyl group attached to an aromatic ring ___
4. They are modifications of amino acids; they are nitrogenous compounds that are bases is ___
5. Monoterpenoids consists of ________ carbon
6. Number of isoprene units in carotenoids is _______
7. Chlorophyll is an example of _______________
8. The best example of Monoterpenoids is_______
9. Terpenoids made out of _________
10. The molecular formula for isoprene is ______
11. Essential oils are types of ___________
12. Members of secondary metabolites include ____, _____ and ¬¬¬¬¬¬¬¬¬¬¬
13. Terpenes are derived biosynthetically from units of ________, which has the molecular formula
14. Organic compounds produced by the plants which have no direct role in the growth and Development are called as __________
15 Compounds characterized by at least one aromatic ring bearing one or more hydroxyl Groups is known as _____
16 Phenolic compounds are biosynthesised through ____ pathway
17 The ______ and ______ contribute to flower and fruit colours
18 Phenolic compoundso produce _____ effect on succeeding crop
19 Element from the periodic table is found in all Alkaloids is ______
20 ______ number of alkaloids known to exists
21 Compounds that contain a fully unsaturated six carbon ring linked to an oxygen are called ____.
22 Alkaloids are naturally occurring compounds which contain ....in their molecules.
23 Derivatives of five carbon isoprene units is _____
24 The secondary metabolites are the end product of _________
25 The role of secondary metabolites is mainly ___
26 Group of compounds characterized by at least one aromatic ring bearing one or more hydroxyl groups is _________
27 Phenolics can be classified into two groups: the ____ and the _____
28 Flavonoids are present in _______ and _______ of plants
29 _____ coined the term terpenes
30 All terpenoids are classified based on number of _________
31 _______ are formed by monoterpenes (C10), with two isoprene units
32 The amino acids that are most often serve as alkaloidal precursors are ________, _________, ______, _____________, ________ and ________
33 All names of alkaloids should end in ____
34 The key starting materials are ______ for Shikimate pathway
35 True alkaloids derived from ____
36 Phenolics are uncommon in ______

37 In plants, phenols are present largely in ______

38 The member of the non-flavonoids is _______

39 _____ consist of 10-C atoms or two isoprene residues

40 Both mono and sesquiterpenoids have common source is _______

41 ____ are low-molecular-weight compounds that are produced as part of the plant defence system

ANSWER

Sl.No	Answer	Sl.No	Answer
1	Primary metabolite	22	N atoms
2	Terpenoids	23	Terpenoids
3	Phenolics	24	Primary metabolism
4	Alkaloids	25	Defence
5	Ten	26	Phenolics
6	8	27	Flavonoids, non-flavonoids
7	Diterpenoid	28	Fruits and flower
8	Menthol	29	Leopold Ruziicka
9	isoprene	30	Five carbon
10	C_5H_8	31	Menthol/ linalool/ geraniol / caryophyllene
11	Terpenes	32	Phenylalanine / tyrosine / tryptophan / histidine / anthranilic acid / lysine and ornithine
12	Phenolics, alkaloid, terpenoids	33	"-ine"
13	C_5H_8	34	phosphoenolpyruvate (PEP) and Erythrose 4P
14	Secondary metabolites	35	Amino acids
15	Phenolics	36	bacteria, fungi, and algae
16	Shikimate	37	Vascular plants
17	Flavonoids and anthocyanin	38	Gallic acid
18	Allelopathic	39	Monoterpenes
19	N	40	Essential oils
20	10000	41	Phytoalexins
21	Phenolics		

Short notes

1) **Secondary metabolites**

A substance essential to the metabolism of a particular organism or to a particular metabolic process is called metabolites. A plant cell produces two types of metabolites: primary metabolites involved directly in growth and metabolism (carbohydrates, lipids and proteins), and secondary metabolites considered as end products of primary metabolism and not involved in metabolic activity (alkaloids, phenolics, sterols ,steroids, essential oils, lignin's and tannins etc).They acts as defence chemicals. Their absence does not cause bad effects in the plants. Organic compounds produced by the plants which have no direct role in the growth and development are called as secondary metabolites. There are about 100,000 secondary compounds that are produced by the plants and the structures of more than 15000 alkaloids, 30000 terpenes, several thousand phenyl propanoids, 1000 flavoniods, 500 quinones, 700 polyacetylenes and 800 non-protein amino acids have already been characterised. Secondary metabolites are frequently produced at highest levels during a transition from active growth to stationary phase. Secondary metabolites are biosynthetically derived from primary metabolites. They are more limited in distribution being found usually in specific families. Chemical warfare to protect plants from the attacks by predators, pathogens, or competitors. Attract pollinators or seed dispersal agents. Important for abiotic stresses, Medicine and Industrial additives. The four major classes of secondary metabolites are alkaloids, glycosides, phenolics, Terpenoid

2) **Phenolics**

Phenolics are a group of compounds characterized by at least one aromatic ring bearing one or more hydroxyl groups. Phenolic compounds from plants are one of largest group of secondary plants constituents synthesized by fruits, vegetables, teas, cocoa and other plants that possess certain health benefits. They are characterized by the antioxidant, anti-inflammatory, anti-carcinogenic and other biological properties, and may protect from oxidative stress and some diseases. Phenolics range from simple, low molecular-weight, single aromatic-ringed compounds to large and complex tannins and derived polyphenols. They can be classified based on the number and arrangement of their carbon atoms and are commonly found conjugated to sugars and organic acids. Phenolics can be classified into two groups: the flavonoids and the non-flavonoids. These phenolic compounds are biosynthesised through shikimate pathway. Flavonoids are polyphenolic compounds comprising fifteen carbons, with two aromatic rings connected by a three-carbon bridge. They are the most numerous of the phenolics and are found throughout the plant kingdom. They are present in high concentrations in the epidermis of leaves and the skin of fruits and have important and varied roles as secondary metabolites

3. Terpenoids

The chemist Leopold Ruziicka showed that many compounds found in nature were formed from multiples of five carbons arranged in the same pattern as an isoprene molecule (obtained by pyrolysis of natural rubber). He called these compounds "terpenes".Terpenoids are the largest and most diverse family of natural products, ranging in structure from linear to polycyclic molecules and in size from the five-carbon hemiterpenes to natural rubber, comprising thousands of isoprene units. All terpenoids are synthesized through the condensation of isoprene units (C5) and are classified by the number of five-carbon units present in the core structure. They are polymeric isoprene derivatives and synthesized from acetate via the mevalonic acid pathway. During their formation, the isoprene units are linked in head and tail fashion. The number of units incorporated into a particular terpene serves as a basis for their classification. Many flavour and aromatic molecules, such as menthol, linalool, geraniol and caryophyllene are formed by monoterpenes (C10), with two isoprene units, and sesquiterpenes (C15), with three isoprene units. Other bioactive compounds, such as diterpenes (C20), triterpenes (C30) and tetraterpenes (C40) show very special properties

4. **Classification of Terpenes**
    1 Hemiterpenes consist of a single isoprene unit.
    2. Monoterpenes consist of two isoprene units and have the molecular formula $C_{10}H_{16}$. Examples of monoterpenes are: geraniol, limonene and terpineol.
    3. Sesquiterpenes consist of three isoprene units and have the molecular formula $C_{15}H_{24}$. Examples of sesquiterpenes are: humulene, farnesenes, farnesol.
    4. Diterpenes are composed of four isoprene units and have the molecular formula $C_{20}H_{32}$. They derive from geranylgeranyl pyrophosphate. Examples of diterpenes are cafestol, kahweol, cembrene and taxadiene (precursor of taxol)
    5. Triterpenes consist of six isoprene units and have the molecular formula $C_{30}H_{48}$.
    6. Tetraterpenes contain eight isoprene units and have the molecular formula $C_{40}H_{64}$ Biologically important tetraterpenes include the acyclic lycopene, the Monocyclic gamma-carotene, and the bicyclic alpha- and beta-carotenes.

5. **Alkaloids**

    Alkaloids are defined as basic compounds synthesized by living organisms containing one or more heterocyclic nitrogen atoms, derived from amino acids (with some exceptions) and pharmacologically active. The class name is directly related to the fact that nearly all alkaloids are basic (alkaline) compounds. Alkaloids constitute a very large group of secondary metabolites, with more than 12,000 substances isolated. A huge variety of structural formulas, coming from different biosynthetic pathways. Alkaloids are produced by a large variety of organisms, such as bacteria, fungi, animals but mostly by plants as secondary metabolites. Most of them are toxic to other organisms and can be extracted by acid-base. There are many different ways of classifying alkaloids; here we use a system based mainly on either the type of ring structure or the botanical taxa in which the alkaloids are found. By this method, there are some sixteen major groups of alkaloids. They are bitter in taste. Derived from amino acids. The amino acids that are most often serve as alkaloidal precursors are: phenylalanine, tyrosine, tryptophan, histidine, anthranilic acid, lysine and ornithine. Alkaloids can occur in plant kingdoms; among the angiosperms, Leguminosae, Papaveraceae, Ranunculaceae, Rubiaceae, Solanaceae, Berberidaceae are outstanding alkaloid-yielding plants. All names of alkaloids should end in “-ine” A prefix or suffix is added to the name of a principal alkaloid from the same source. (Quinine, quinidine, hydroquinine)

6 **Shikimate pathway**

Shikimate pathway is an important pathway in plants through which many secondary plant products are synthesised. The key starting materials are phosphoenolpyruvate (PEP) and Erythrose 4P derived from glycolysis and pentose phosphate pathways, respectively. These two compounds condense to produce a six carbon cyclic compound with one carbon (COOH) side chain namely shikimate. Then shikimate is phosphorylated and condensed with another molecule of PEP to produce a cyclic compound containing a three carbon and one carbon side chains. This is finally converted to aromatic amino acids phenylalanine and tyrosine. These amino acids are deaminated followed by hydroxylation at different carbon atoms in the aromatic ring to form cinnamic acid derivatives. These cinnamic acid derivatives are utilised for the synthesis of different phenolic compounds.

Essay types

1. **Functions of phenolics**

1. Phenolics are of great importance as cell wall components. They form part of cell wall structures such as lignins, cutins and suberins, which provide mechanical support and function as barriers against microbial attack
2. The flavonoids and anthocyanins contribute to flower and fruit colours. This is important for attracting insects and animals to the plant for pollination and seed dispersal.
3. Phenolics also play a defensive role in plants by protecting against predators.
4. Simple phenolic acids, polyphenolics like tannins and phenolic resins at the plant surface are effective feeding deterrents
5. Phenolics are accumulated as post-inflectional low molecular compounds called phytoalexins as a result of microbial attack.
6. Among the phenolic phytoalexins, hydroxycoumarins and hydroxycinnamate conjugates contribute to disease resistance mechanism in plants.
7. Phenolic compounds also produce allelopathic effect. A well-known compound from Juglans species is juglone which is highly toxic for a wide range of plants. It occurs in the plant as a non-toxic glucoside and is made active by deglucosylation and oxidation after leaching from the leaves into the soil.
8. Phenolics also function as signal molecules in the interaction between nitrogen fixing bacteria and leguminous plants
9. These plants exude flavonoids which act selectively in Rhizobia as inducers of nodulation gene transcription.

10 Salicylic acid is strongly implicated as a signal molecule which induces active defence responses in several plant species against many types of pathogens.

11 Recently, it has been shown that phenolic compounds function as effective antioxidants.

12 Polyphenolics are important in foodstuffs, wines and herbal teas because of their astringent taste.

13 Plants rich in polyphenolics were used as tanning agents in leather industries.

14 Phenolic pigments (anthocyanins, flavones etc) of fruits are most widespread food colours occurring in fruit juices, wines and jams.

15 Anthocyanins have considerable potential in the food industry as safe and effective food additives.

2. **Explain the occurrence, classification and function of alkaloids**

Alkaloids are commonly applied to basic nitrogenous compounds of plant origin that are physiologically active. The names of these molecules tend to end in the suffixes –ine or –in. Many are derived from amino acids, but others result from modification of various classes of molecules including polyphenols, terpenes, or steroids. Alkaloids are produced by a large variety of organisms, such as bacteria, fungi, animals but mostly by plants as secondary metabolites. Most of them are toxic to other organisms and can be extracted by acid-base. The class name is directly related to the fact that nearly all alkaloids are basic (alkaline) compounds. Alkaloids constitute a very large group of secondary metabolites, with more than 12,000 substances isolated. Dicots are richer in alkaloids than Monocots. Families rich in Alkaloids: Apocynaceae, Rubiaceae, Solanaceae and Papaveracea. Families free from Alkaloids: Rosaceae, Labiatae

There are many different ways of classifying alkaloids; here we use a system based mainly on either the type of ring structure or the botanical taxa in which the alkaloids are found.

a) **True Alkaloids**

True alkaloids derived from amino acids. Heterocyclic ring with nitrogen. Highly reactive substances in low doses also .Bitter taste with white appearance. Form water soluble salts Examples: cocaine, morphine, nicotine, dopamine

b) **Proto-Alkaloids**

Which contain nitrogen and also originate from amino acids. Examples include mescaline, adrenaline and ephedrine

c) **Pseudo alkaloids**

Alkaloid-like compounds that do not originate from amino acids. This group includes terpene-like and steroid-like alkaloids, as well as purine-like alkaloids such as caffeine, theobromine, theacrine and theophylline.

Functions

1) They may act as protective against insects and herbivores due to their bitterness and toxicity.
2) They are, in certain cases, the final products of detoxification (waste products).
3) Source of nitrogen in case of nitrogen deficiency
4) They, sometimes, act as growth regulators in certain metabolic systems.
5) They may be utilized as a source of energy in case of deficiency in carbon dioxide assimilation
6) Muscle relaxant, Pain killers, tranquilizers, Mind altering drugs, Chemotherapy

2. **Explain the occurrence, classification and function of phenols**

Compounds that contain a fully unsaturated six carbon ring linked to an oxygen are called phenolics These are secondary natural metabolites produced in plants biogenetically from either the shikimate/phenylpropanoid pathway which directly provide phenylpropanoid and which fulfil a very broad physiological role in plants. Plant phenolics are a chemically heterogeneous group of nearly 10,000 individual compounds: Some are soluble only in organic solvents, some are water-soluble carboxylic acids and glycosides, and others are large, insoluble polymers. Plant phenolics are synthesized by several different routes and thus constitute a heterogeneous group from a metabolic point of view. Two basic pathways are involved: the shikimic acid pathway and the malonic acid pathway. The shikimic acid pathway participates in the biosynthesis of most plant phenolics. The malonic acid pathway, although an important source of phenolic secondary products in fungi and bacteria, is of less significance in higher plants.

Phenolic compounds are the most widely distributed secondary metabolites, ubiquitously present in the plant kingdom, even if the type of compound present varies according to the phylum under consideration. Phenolics are uncommon in bacteria, fungi, and algae. Bryophytes are regular producers of polyphenols including flavonoids, but it is in the vascular plants that the full range of polyphenols is found. It is estimated that about 2% of all carbon photosynthesized by plants is converted into flavonoids or closely related compounds

Phenolics can be mainly classified into 2 groups: a) The flavonoids b) the non-flavonoids

The Flavonoids

Largest group of phenols: 4500, -Flavonoids are polyphenolic compounds comprising fifteen carbons, with two aromatic rings connected by a three-carbon bridge. They are the most numerous of the phenolics and are found throughout the plant kingdom. They are present in high concentrations in the epidermis of leaves and the skin of fruits and have important and varied roles as secondary metabolites

Major role in plants: color, pathogens, light stress

Very often in epidermis of leaves and fruit skin and Potential health promoting compounds – antioxidants

They can be divided into anthocyanins, flavones and flavonols.

The non-flavonoids

The main non-flavonoids of dietary significance are the C6–C1 phenolic acids, most notably gallic acid, which is the precursor of hydrolysable tannins, the C6–C3 hydroxycinammates and their conjugated derivatives, and the polyphenolic C6–C2–C6 stilbenes

Functions

The highly ordered interactions between plants and their biotic and abiotic environments have been a major driving force behind the emergence of specific natural products. In this connection, the accumulation of phenolics in plant tissues is considered a common adaptive response of plants to adverse environmental conditions, therefore increasing evolutionary fitness.

a) Plant phenolics are considered to have a key role as defence compounds when environmental stresses, such as high light, low temperatures, pathogen infection, herbivores, and nutrient deficiency, can lead to increased production of free radicals and other oxidative species in plants. A growing body of evidence suggests that plants respond to these biotic and abiotic stress factors by increasing their capacity to scavenge reactive oxygen species.

b) Plant growth depends on the supply of recycled nutrients; external nutrient inputs. Polyphenols, phenolic compounds can directly affect the composition and activity of decomposer communities, thus influencing the rates of decomposition and nutrient cycling.

c) Different types of soluble polyphenols, such as ferulic acid, gallic acid, or flavonoids, have been found to either stimulate or inhibit spore germination and hyphal growth of saprotrophic fungi.

d) Plant mycorrhizal infection, nutrient uptake, and plant growth can be impaired by specific phenolics released by competitors in a process referred to as allopathy. Flavonoids present in the root exudates of a variety of leguminous plants activate the rhizobium genes responsible for the nodulation process and might be responsible for vesicular–arbuscular mycorrhiza colonization.

e) Nodule formation is initiated by the host plant roots exuding phenolic flavonoid compounds into the rhizosphere.

f) Anthocyanins represent a class of flavonoids providing the red and blue/purple colors familiar in many flowers and fruits. These compounds are synthesized as visual cues, to attract pollinators and other animals for seed dispersal, as well as molecular cues protecting plants from various stress conditions, and are stored in the acidic vacuole of specialized cells.

g) Plants produce a broad range of phenolic metabolites that serve a dual function of both repelling and attracting different organisms in the plant's surroundings. The role of plant phenolics in chemo ecology, especially on the feeding behaviour of herbivores, has been recognized

3. **Explain the occurrence, classification and function of terpenoids**

Terpenoids, also known as isoprenoids, are the most numerous and structurally diverse natural products found in many plants Terpenoids are the largest and most diverse family of natural products, ranging in structure from linear to polycyclic molecules and in size from the five-carbon hemiterpenes to natural rubber, comprising thousands of isoprene units. Terpenes are hydrocarbons resulting from the condensation of several 5-carbon isoprene units. Plant terpenoids are used extensively for their aromatic qualities and play a role in traditional herbal remedies. Terpenoids contribute to the scent of eucalyptus, the flavors of cinnamon, cloves, and ginger, the yellow color in sunflowers, and the red color in tomatoes. Well-known terpenoids include citral, menthol, camphor, salvinorin A in the plant Salvia divinorum, the cannabinoids found in cannabis, ginkgolide and bilobalide found in Ginkgo biloba, and the curcuminoids found in turmeric and mustard seed.

Classification

Most natural Terpenoid hydrocarbon have the general formula $(C_5H_8)n$. Terpenes are classified into many categories based on the no. of carbon atoms and iso- prene residues present in their structure

i) Monoterpenes. They consist of 10-C atoms or two isoprene residues.
ii) Sesquiterpenes. These contain 15-C atoms or three isoprene residues.
iii) Diterpenes. These contain 20-C atoms or four isoprene residues.
iv) Triterpenes. These consist of 30-C atoms or six isoprene units.
v) Tetraterpenes. These consist of 40-C atoms or eight isoprene residues.
vi) Polyterpenes. These consist of large number of isoprene residues.

Both mono and sesquiterpenoids have common source i.e essential oils

General properties of Terpenoids

1. Most of the terpenoids are colourless, fragrant liquids which are lighter than water and volatile with steam. A few of them are solids e.g. camphor. All are soluble in organic solvent and usually insoluble in water. Most of them are optically active.
2. They are open chain or cyclic unsaturated compounds having one or more double bonds. Consequently they undergo addition reaction with hydrogen, halogen, acids, etc. A number of addition products have antiseptic properties.
3. They undergo polymerization and dehydrogenation
4. They are easily oxidized nearly by all the oxidizing agents. On thermal decomposition, most of the terpenoids yields isoprene as one of the product

Functions

a) Plants employ Terpenoid metabolites for a variety of basic functions in growth and development majority of terpenoids for more specialized chemical interactions and protection in the abiotic and biotic environment.

b) Traditionally, plant-based terpenoids have been used by humans in the food, pharmaceutical, and chemical industries, and more recently have been exploited in the development of biofuel products.

c) Plants have direct and indirect defence responses when they are attacked by herbivores or infected by fungal and bacterial pathogens. Direct defences include physical structures, such as trichomes and thorns, and the accumulation of chemical or biochemical compounds that have antibiotic activities or toxicities

d) Phytoalexins are low-molecular-weight compounds that are produced as part of the plant defence system. In many plant species diterpenes and sesquiterpenes act as phytoalexins.

Definition

1. **Secondary metabolites:** Organic compounds produced by the plants which have no direct role in the growth and development are called as secondary metabolites
2. **Phenolics:** Phenolics are a group of compounds characterized by at least one aromatic ring bearing one or more hydroxyl groups
3. **Alkaloids**: Alkaloids are basic N containing heterocyclic compounds derived from higher plants often having marked physiological activity.
4. **Terpenoid :** Any of a large class of organic compounds including terpenes, diterpenes, and sesquiterpenes. They have unsaturated molecules composed of linked isoprene units, generally having the formula (C_5H_8)