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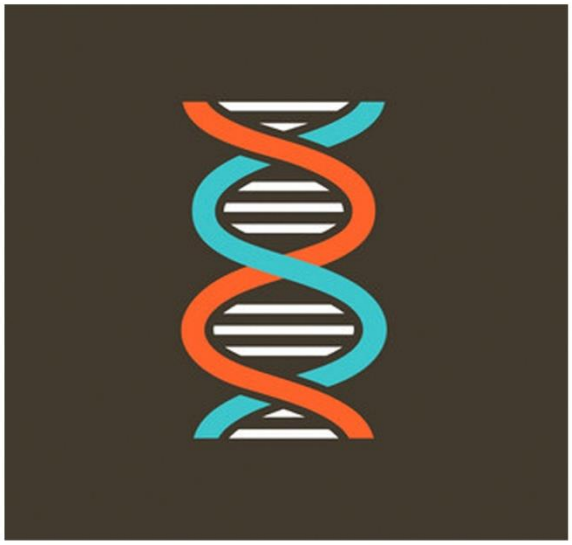
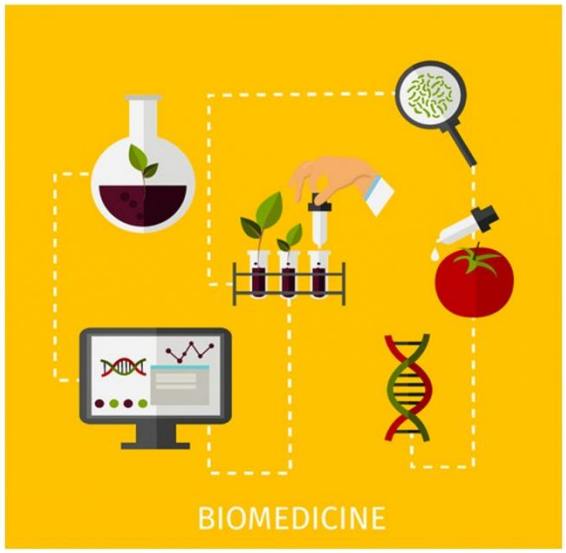
FUNDAMENTALS OF PLANT BIOCHEMISTRY AND BIOTECHNOLOGY
BIOCHEM-231

IMPORANT QUESTIONS FROM PREVIOUS YEAR EXMINATION





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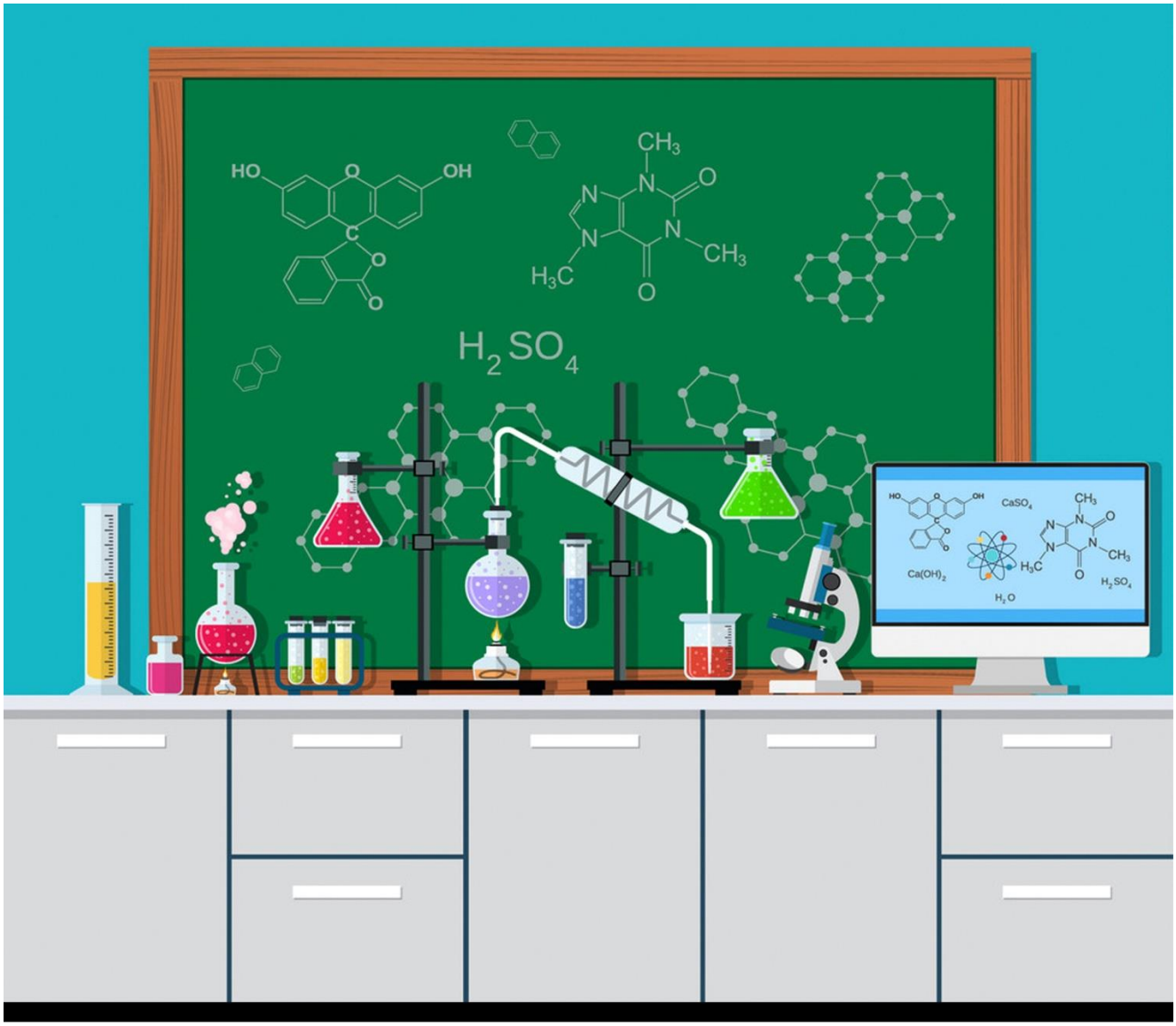

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Q.1.Define Biochemistry & Give the scope and importance of Biochemistry In Agriculture.

Ans: Biochemistry: Biochemistry may be defined as a science concerned with chemical nature and chemical behavior of the living matter.

Scope and Importance of Biochemistry in Agriculture:

1) To evaluate nutritive value of cereals, pulses, poultry and cattle feed.

2) Development and exploitation of better genotypes.

3) Removal and inactivation of toxic or anti nutritional factors present in food grains in general and grain legumes in particular by breeding and chemical treatments. e.g. BOAA in Lakh dal, Trypsin inhibitors of soybean, Aflatoxins of groundnut.

4) Food preservation and processing technology and post-harvest physiology of fruit crops and vegetables and their nutritional quality.

5) Biochemistry of disease and pest resistance.

6) Biochemistry of drought resistance, proline and hydroxyproline imparts drought resistance to Jowar.

7) Formulation of balanced diet

8) Use of nonconventional sources of protein foods viz., single cell proteins, fish protein concentrates, mushrooms and leaf proteins.

9) Developments in the field of inter mediatory metabolism i.e. synthesis and degradation of constituents of living tissues.

Q.2 Define Biomolecules. Write down the important biomolecules of life.

Ans: Biomolecules: An organic compound normally present as an essential component of living organism.

Important Biomolecules of life:

1) Water: Being the universal solvent and major constituents (60%) of any living body without which life is impossible. It acts as a media for the physiological and biochemical reactions in the body itself. Maintain the body in the required turgid condition.

2) Carbohydrates: It is very important for source of energy for any physical body function.

3) Proteins: These are very important from body maintenance point of view,helps in tissue, cell formation.

4) Lipids: These are very important from energy source as well as human nutrition point of view.

5) Nucleic Acids: Nucleic acids are very important as DNA carries the hereditary information and RNA helps in protein formation for the body.

6) Enzymes: Enzymes are simple or combined proteins acting as specific catalysts and activates the various biochemical and metabolic processes within the body.

Q.3 Define Carbohydrates & classify monosaccharides with suitable examples.

Ans: Carbohydrates are defined as polyhydroxy aldehydes or polyhydroxy ketones and the substances which yield these derivatives on hydrolysis.

1) Mono saccharides:

This are simple sugars that are cannot be hydrolyzed into smaller units. Depending upon no. of carbon in a unit, mono saccharides are subdivided into a dioses to decoses. More common subclasses of mono saccharides are:

Aldoses:	**Ketoses:**
Aldotrioses e.g. Glycerose,	Ketotrioses e.g Dihydroxyacetone,
Aldotertroses e.g. Erythrose,	Ketotetroses e.g Erythrulose,
Aldopentoses e.g. Ribose,	Ketopentoses e.g Ribulose,
Aldohexoscs e.g. Glucose, Galatose	Ketohexoses e.g. Fructose,
Aldoheptose e.g. Glucoheptose.	Ketoheptose e.g. Scdoheptulose.

Q.4 Give the Classification of Carbohydrates. And write the functions of carbohydrates.

Ans:

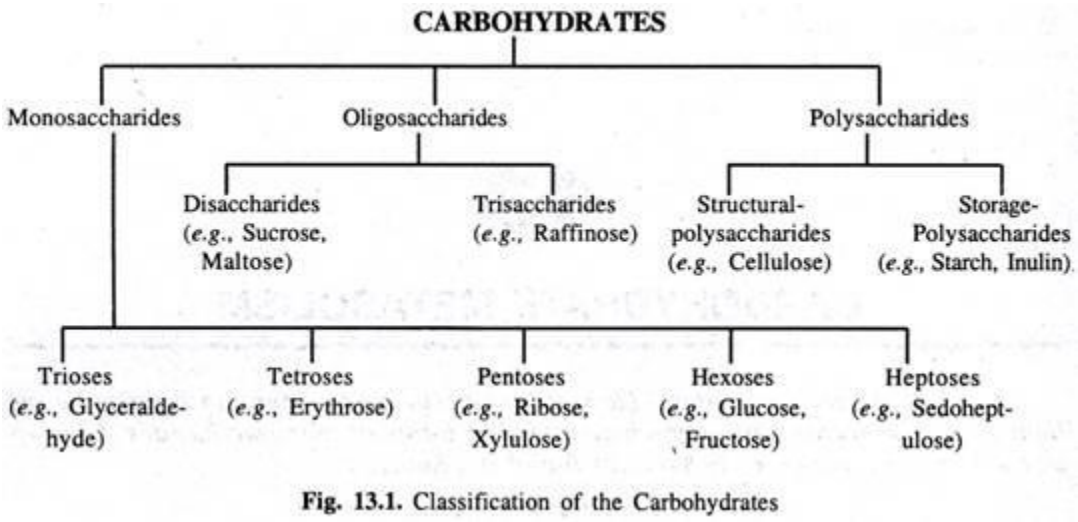


Fig. 13.1. Classification of the Carbohydrates

1) **Mono saccharides:** Simple sugars are cannot be hydrolyzed into smaller units. Depending upon no. of carbon in a unit, mono saccharides are subdivided into a dioses to decoses.
2) **Oligosaccharide:** Oligosaccharides are polymers of mono saccharides containing two to ten residues accumulate in vacuole while polysaccharides in plastids.
3) **Polysaccharides:** Polysaccharides are polymeric anhydrides of mono saccharides. The long chain polymers are either straight chain or branched. They are also called glycanes.

Functions of Carbohydrates:

i) Supply energy **ii)** Stored energy for future use **iii)** Structural constituents **iv)** Proteins sparing action **v)** Necessary for oxidation of protein and fat **vi)** Necessary for synthesis of nonessential amino acids **vii)** Conserve water and electrolyte **viii)** Beneficial effect on micro flora.

Q.5 Define amino acids. Classify them and write down Functions and properties of Amino acids.

Ans: Amino acids are organic acids which contain both basic (amino - NH_2) and acidic (carboxyl COON) groups .

Classification of Amino Acid:

1.Based on Composition they are Classified as:

1) Aliphatic mono amino monocarboxylic acids: e.g. glycine, alanine, valine, leucine, isolucine

2) Aromatic amino acids: e.g. phenylalanine, tyrosine and tryptophan

3) Hydroxy amino acids: e.g. serine, threonine

4) Acidic or dicarboxylic acids: e.g. Aspartic acid and glutamic acids

5) Basic amino acids: e.g. Lysine, arginine and histidine.

6) Sulphur containing amino acids: e.g. methionine, cysteine

7) Secondary amino acids: e.g. proline and hydroxyproline

8) Non protein amino acid: e.g. Aminobutyrate, homoserine and cystathionine in plants.

2.Based on their presence or absence in proteins:

Amino acids are classified as protein amino acids and non protein amino acids.

a) Protein amino acids: - Amino acids that are used for synthesis of proteins are called protein amino acids. Eg lucine, proline.

b) Non protein amino acids: Eg:- beta alanine, hydroxy proline, N- acetyl glutamic acid etc

3. Based on requirement to the body

Essential : Valine, Leucine.

Non essential: Proline,Glycine.

4. Based on polarity of the side chains: This is the most accepted form of classification of amino acids which is based on polarity and hydrophobic nature of R groups.

a) Nonpolar or hydrophobic: Valine, Leucine.

b) Polar uncharged amino acids: Serine, Threonine.

c) Polar amino acids with positively charged side chains: Lysine, Histidine

d) Polar amino acids with negatively charged side groups: Aspartic acid and Glutamic acid.

Properties of Amino Acids:

Color less, crystalline, tufts of slender needles (Tyrosine) to hexagonal plates (cystine). Taste varies from sugar sweet (glycine, alanine) through tasteless (Tyrosine) to bitter (arginine). All amino acids except glycine contain at least one asymmetric carbon atom ornithine needles (tyrosine) to hexagon plates (cysteine)

Functions of Amino Acids:

i) Formation of Proteins **ii)** Maintenance of Tissues **iii)** Formation of Enzymes, Hormones and Antibodies.

Q.6 Define Protein. Classify them on the basis of their composition.

Ans: Proteins are polymeric compounds of the monomeric units of amino acid.

A) Based on Composition:

i) Simple Proteins

ii) Conjugated Proteins

iii) Derived proteins

i) Simple Proteins: Classified according to solubility

a) Albumins

b) Globulins

c) Glutelins

d) Histories

e) Protamine

f) prolamines

g) Scleroproteins

ii) Conjugated Proteins: Contain amino acid + prosthetic group.

a) Glycoproteins

b) Chromoproteins

c) Lipoproteins

d) Nucleoproteins

e) Phosphoprotein

iii) Derived Proteins: Derivatives of proteins due to action of heat, enzymes, or chemical reagents.

a) Primary Derived

b) Secondary Derived

Q.7 **Write down the functions and properties of Proteins.**

Ans: Properties of proteins:

1. U.V absorption: Proteins absorb U.V radiation at 280 nm because of the presence of aromatic amino acids like tryptophan and tyrosine. This property is used in estimation of proteins.

2. Isoelectric point: Isoelectric point is also called as isoelectric pH. This is the pH at which the number of positive and negative charges is equal in the protein and they are electrically neutral. Solubility of proteins is least at isoelectric pH.

3. Zwitterions: Proteins contain both positive and negative charges and hence they are called as zwitterions. Amino acids will act as zwitterions as they can donate a proton and forms cation.

4. Immunological properties: Proteins exhibit a special property called immunological property, which is useful in defense mechanism. When ever any antigen enters into the body, immediately body releases a special class of proteins called as defense antibodies. The interaction of antigen and antibody to form the antigen-anti body complex is called immune reaction.

5. Denaturation: It is a physical change in which there is a collapse of protein structure. Due to denaturation, there is a decrease in solubility and loss of biological activity of proteins.

6. Protein folding: Many proteins fold to their native conformation on their own by self assembly. However several other accessory proteins help in this process.

7. Solubility: Protein solubility is influenced by pH, heavy metals, salts and organic solvents.

Functions of Proteins:

1.Few hormones are examples of this class of proteins that are responsible for the regulation of many processes in organisms. Eg: Insulin.

2. The proteins are involved in transporting some chemical compounds and ions. Eg: Haemoglobin.

3.The proteins are involved in the defense mechanism of the cell. Eg: Gamma globulins.

4.Proteins are involved in maintaining the structure of other biological components like cells and tissues. Eg: Collagen, elastin.

5. Proteins are involved in contraction of the tissues. Ex: Actin and myosin are responsible for muscular motion.

6. Proteins contain energy, which can be released during various metabolic processes in the organism. Ex: Egg ovalbumin, milk casein

7. The proteins act as receptor molecules. They are responsible for signal detection and translation into other type of signal.

Q.8 Define Nucleic Acids and give the hydrolytic products of RNA & DNA.Differnciate RNA & DNA.

Ans: Nucleic acids are high molecular weight polymers which store and transfer genetic material from generation to generation.

Hydrolytic Products Of RNA & DNA:

Sr. No.	Components	RNA	DNA
1	Pentose Sugar	D - Ribose	D - 2 Deoxyribose
2	Acid	Phosphoric Acid	Phosphoric Acid
3	Nitrogen Bases		
	a) Purines	Adenine, Guanine	Adenine, Guanine
	b) Pyrimidine	Cytosine, Uracil	Cytosine, Uracil

S.N.	DNA	RNA
1.	DNA stands for Deoxyribonucleic Acid. The sugar portion of DNA is 2-Deoxyribose.	RNA stands for Ribonucleic Acid. The sugar portion of RNA is Ribose.
2.	The helix geometry of DNA is of B-Form (A or Z also present).	The helix geometry of RNA is of A-Form.
3.	DNA is a double-stranded molecule consisting of a long chain of nucleotides.	RNA usually is a single-strand helix consisting of shorter chains of nucleotides.
4.	The bases present in DNA are adenine, guanine, cytosine and thymine.	The bases present in RNA are adenine, guanine, cytosine and uracil.
5.	DNA is self-replicating.	RNA is synthesized from DNA on an as-needed basis.
6.	Base Pairing :AT (adenine-thymine)GC (guanine-cytosine).	Base Pairing :AU (adenine-uracil)GC (guanine-cytosine).
7.	Purine and Pyrimidine bases are equal in number.	There is no proportionality in between the number of Purine and Pyrimidine bases.
8.	DNA is susceptible to UV damage.	Compared with DNA, RNA is relatively resistant to UV damage.
9.	Hydrogen bonds are formed between complementary nitrogen bases of the opposite strands (A-T, C-G).	Base pairing through hydrogen bonds, occurs in the coiled parts.
10.	DNA is found in the nucleus of a cell and in mitochondria.	Depending on the type of RNA, this molecule is found in a cell's nucleus, its cytoplasm, and its ribosome.
11.	DNA can't leave the nucleus.	RNA leaves the nucleus (mRNA).
12.	The C-H bonds in DNA make it fairly stable, plus the body destroys enzymes that would attack DNA. The small grooves in the helix also serve as protection, providing minimal space for enzymes to attach.	The O-H bond in the ribose of RNA makes the molecule more reactive, compared with DNA. RNA is not stable under alkaline conditions, plus the large grooves in the molecule make it susceptible to enzyme attack.
13.	Renaturation after melting is slow.	It is quite fast.
14.	DNA is only two types: intra nuclear and extra nuclear.	Three different types of RNA: m-RNA, t-RNA and r-RNA.

15.	Its quantity is fixed for cell.	The quantity of RNA of a cell is variable.
16.	It is long lived.	Some RNAs are very short lived while others have somewhat longer life.
17.	**Functions:**Long-term storage of genetic information; transmission of genetic information to make other cells and new organisms.	**Functions:**Used to transfer the genetic code from the nucleus to the ribosomes to make proteins. RNA is used to transmit genetic information in some organisms and may have been the molecule used to store genetic blueprints in primitive organisms.

Q.9 Define Lipids. Classify them on the basis of product of Hydrolysis.

Ans: Chemically lipids are defined as esters of glycerol and fatty acids or as the triglycerides of fatty acids

Classification on the basis of product of Hydrolysis:

1. Simple lipids 2. Compound Lipids 3. Derived Lipids

1.Simple Lipids: On hydrolysis gives fatty acids and alcohol (trihydric or monohydric)

Oils: Unsaturated fatty acid + glycerol.

Fats: Saturated fatty acids + glycerol,

Waxes: Fatty acids + mono or dihydric alcohol.

Simple glyceride: Contains same fatty acids. .

Mixed glyceride: Contains different fatty acids.

2. Compound Lipids: On hydrolysis gives phosphoric acid, various sugars, sphingosine, ethanolamine and serine in addition to fatty acids and glycerol.

a) Phospholipid b) Glycolipids c) Sphingophospholipids

3. Derived Lipids

Hydrolytic products of simple and compound lipids

i) Alcohols: Glycerol and other sterol ii) Fatty acids iii) Terpenoids

Q.10 Give the Significance of Lipids.

Ans: 1. Lipids act as reservoir of energy in biological systems. Being more reduced than carbohydrates, lipids can store more energy. The most important storage form of lipids is the triacyl glycerols stored in the oil bodies in plant seeds and adipose tissues in animals.

2. Lipids act as the major components of biological membranes. The most Important class of lipids in this regard is the amphipathic phospholipids with a small hydrophilic head and a long hydrophobic tail arranged in a bilayer form.

3. Some lipids act as members of electron transport system in inner mitochondrial membrane viz. ubiquinone and also phosphorylation systems in thylakoid membrane.

4. Lipids act as carriers of sugars viz. dolichol in the biosynthesis of glycoproteins.

5. Lipids materials are used for the biosynthesis of certain hormones in animals & plants.

6. Lipids in the form of bile acids (e.g. cholic acid) help in the digestion and absorption of other lipids.

7. Triacyl glycerols act as heat insulating materials.

Q.11 Define Enzymes. Give the Classification Of Enzymes.

Ans: Enzyme: Catalytically active protein of biological origin or organic catalyst produced by living cells.

Classification Of Enzymes:

International Union Of Biochemistry (IUB) gives this Clasification

CLASS	DESIGNATION	FUNCTION
EC1	Oxidoreductases	catalyze oxidation/reduction reactions
EC2	Transferases	transfer a functional group (e.g. a methyl or phosphate group)
EC3	Hydrolases	catalyze the hydrolysis of various bonds
EC4	Lyases	cleave various bonds by means other than hydrolysis and oxidation
EC5	Isomerases	catalyze isomerization changes within a single molecule
EC6	Ligases	join two molecules covalent bonds.

cbnm .

Q.12. What are the properties of Enzymes.

Ans: Properties of enzymes.-

Some important properties of enzymes are given below,

1) Colloidal nature: Because of the large size, the enzyme molecules possess extremely low rates of diffusion and form colloidal systems in water. Being colloidal in nature enzymes are non dialyzable, although some contain dialyzable or dissociable component in the form of co-enzyme.

2) Catalytic nature: Enzyme act catalytically and accelerates the rate of the chemical reactions occuring in plant and animal tissues. They participate in reaction but at the end of reaction, they recovered as such without undergoing any change.

3)High molecular weight: Enzyme molecules are of giant size. Their molecular weight range from 12, 000 to over 1 million. Therefore, they are very large as compared to substrate or functional group they act upon.

4) Specificity of enzyme action: Enzymes are specific in their action. Their specificity lies in the fact that they may act-

Absolute specificity: Some enzymes are capable of acting on only one substrate. e.g.urease acts only on urea to produce ammonia and CO2.

Group specificity: Some enzymes are capable of catalyzing the reaction of structurally related group of compounds. It may bond dependent or group dependent. e.g. lactate dehydrogenase (LDH) catalyses the inter conversion of pyruvic acid and lactic acid and also number of other structurally related compounds.

Optical specificity: Some enzymes react with only one of the two optical isomers. e.g. arginase acts only on L-arginine and not on its D-isomer (D-arginine).

Geometrical specificity: Some enzymes exhibit specificity towards cis and trans forms. e.g.fumarase acts on fumaric acid(transform) and not on maleic acid which is cis isomer of fumaric acid.

5) Heat sensitivity (ThermoIability): Enzymes are very sensitive to heat. The rate of an enzyme action increases with every rise in temperature of 10°C upto 60°C. Above 60°C the enzyme coagulate and become inactivated. This is because of irreversible change in their chemical structure.

6) Reversibility of reaction : The enzymes are capable of bringing about reversion in a chemical reaction. For e.g Lipase catalyze the synthesis of fat (from glycerol and fatty acids) & it can also hydrolyze the fat into their components.

7) pH sensitivity: The-pH value or the H+ ion concentration of medium controls the activity of an enzyme to a great extent. Usually maximum enzyme activity is obtained at or near the iso electric point of the enzymes.

Q.13 Define Enzyme immobilization .Enlist the Methods and Explain Entrapment method.

Ans.: Enzyme immobilization may be defined as confining the enzyme molecules to a distinct phase from the one in which the substrates and the products are present; this may be achieved by fixing the enzyme molecules to or within some suitable material. It is critical that the substrates and the products move freely in and out of the phase to which the enzyme molecules are confined.

Methods of Immobilization: The various methods used for immobilization of enzymes may be grouped into the following four types:

(i) Adsorption

(ii) Covalent Bonding

(iii)Entrapment

(iv) Membrane Confinement.

(*) Entrapment: In this approach, enzyme molecules are held or entrapped within suitable gels or fibers and there may or may not be covalent bond formation between the enzyme molecules and the matrix. A non-covalent entrapment may be viewed as putting the enzyme molecule in a molecular cage just as a caged bird / animal. When covalent binding is also to be generated, the enzyme molecules are usually treated with a suitable reagent.

Q.14 Explain the Beta oxidation of Fatty acid.

Ans: Beta oxidation of fatty acids: Mechanism of Beta oxidation of fatty acids was proposed by Knoop in 1904. It takes place in mitochondrial matrix.

* B-oxidation of fatty acid is the degradation of fatty acid into the 2 Carbon compound. *Carboxyl group at C_1 of fatty acid is activaed by the attachment of CoA. This allows stepwise oxidation of fatty acyl group at C_3 (Beta carbon). This catabolic mechanism known as Beta oxidation of fatty acid..

Sequence reactions in B-oxidation:

1)Activation of fatty acids.

2)Dehydrogenation of acyl CoA to enol CoA.

3)Hydration of enol CoA to Beta Hydroxyacyl CoA.

4)Oxidation of Beta hydroxy acyl CoA to Beta ketoacyl CoA.

5)Thiolysis of Beta ketoacyl CoA (i.e. formation of 2 carbon compounds, acyl-CoA and acytyl-CoA)

Step 1: Activation of fatty acids: Fatty acids are activated by activation of carboxyl gruop at C_1 by the attachment of CoA and Results is the Formation of acyl-CoA.

Step 2 : Dehydrogenation of acyl-CoA: Acyl-CoA is oxidized by the enzyme acyl-CoA dehydrogenase to produce an enol CoA with a trans double bond between C_2 & C_3. There is a formation $FADH_2$.

Step 3:Hydration of enol-CoA: A mole of water is added to a double bond to form Beta hydroxyacyl CoA. This reaction is catalyzed by the enzymes enol-CoA hydratase.

Step 4: Oxidation of Beta hydroxyacyl-CoA: Beta hydroxyacyl-CoA is dehydrogenate (or oxidized) to form beta Ketoacyl-CoA by the action of enzyme B-hydroxyacyl-CoA dehydrogenase. NAD^+ is the electron acceptor & thus NADH is formed.

Step 5: Thiolysis of Beta ketoacyl-CoA: Thiolysis is a splitting by thiol(-SH) group. This reaction brings the cleavage of Beta ketoacyl CoA by the thiol group of second mole of

CoA. Which yields acetyl-CoA & acyl CoA. The reaction is catalyzed by the enzyme ketothiolase or thiolase. Acetyl CoA enters in citric acid cycle and acyl-CoA undergoes another Beta-Oxidation Cycle.

Q.15 Define Photo-phosphorylation. Give the Type of it & Differentiate between Cyclic and non-cyclic photophosphorylation.

Ans: It is the formation of a phosphate derivative of a biomolecule, usually by enzymatic transfer of a phosphate group from ATP.

Types of Phosphorylation:

1) Photophosphorylation or photosynthetic phosphorylation.

a) Cyclic photophosphorylation

b) Noncyclic photophosphorylation

2) Substrate level photophosphorylation.

3) Oxidative phosphorylation.

Comparison of cyclic and noncyclic photophosphorylation

S.No.	Cyclic photophosphorylation	Noncyclic photophosphorylation
(1)	No oxygen is given off (Anoxygenic).	Oxygen is given off (Oxygenic).
(2)	No water is consumed.	Water is used up.
(3)	Only one light-trapping system (Photosystem 1) is involved.	Two light-trapping systems (Photosystem 1 and II) are involved.
(4)	No NADPH synthesized.	NADPH synthesized
(5)	Last electron acceptor is P_{-}	Last electron acceptor is NADP.
(6)	The system is found dominantly in bacteria.	The system is dominant in green plants.
(7)	The process is not inhibited by DCMU.	The process is stopped by use of DCMU

Q.16 Explain Cyclic photophosphorylation.

Ans: Cyclic photophosphorylation - ATP synthesis driven by cyclic electron flow through photosystem I. During cyclic photophosphorylation, chlorophyll molecules absorb a photon of light energy and become exicted. As a result, the energy of photon get converted into the energy of electron. This electron now shelled from chlorophyll and it is taken by ferrdoxin & then transfer to cytochromes. From cytochromes it again reaches to the chlorophyll molecule from which it was expelled. During this travel of electron, the free energy is utilized for the synthesis of ATP.

Q.17 Write down the Factors affecting Enzyme Activity

Ans: Factors Affecting Enzyme Activity:

1) PH

2) Temperature

3) Substrate Concentration

4) Enzyme Concentration

5) Concentration of any Activator Present

6) Concentration of any Inhibitor Present

7) Ionic Strength

8) Redox Potential

9) Concentration or Reaction Products

(For Detailed Answer refer to ssac354 notes)

Factor affecting enzyme activity:

1) pH: Enzymes are sensitive to pH change. For every enzyme, there is an optimum pH at which it function best. Some enzyme act best in alkaline medium (trypsin- pH 9.0) while some prefer acidic medium (pepsin-pH 2.0). For most biological enzymes the optimum pH is around 7.4.

2) Temperature: The enzyme activity increases with temperature reaches a maximum, then the activity declines with further increase in temperature. The enzymes of high animals (warm blooded) exhibit an optimum temperature around 370C. At temperature 0^0C and 100^0C, the rate of enzyme catalyzes reaction is nearly zero.

3) Substrate concentration: Increase in substrate concentration, increases the rate of reaction at beginning. Further, increase in substrate concentration dose not increases the rate of reaction or the velocity of reaction is constant

4) Enzyme concentration: With the increase in enzyme concentration, the rate of formation of product is increased. When the concentration of enzyme is much lower than the substrate, the rate of enzyme catalyzed reaction is directly depend on the enzyme concentration. The reaction rate is increased, as the concentration of enzyme increased.

5) Concentration of activator present: When enzymes are simple proteins and some of them contains inactive precursors called proenzyme or zymogens, the activation is brought about by participation of another factor known as co-factor. The activation may be the effect of an inorganic ion such as Na or K on the rate of reaction.

6) Concentration of inhibitor present: They slow down the rate of reaction. The most potent poisons of living organisms exert their action by inhibiting enzymes. For e.g. cyanide inhibit cytochrome oxidase.

7) Ionic strength: As the ionic strength is increased further, the solubility of enzyme begin to decrease. At high ionic strength, a protein enzyme may be precipitate from solution and the effect is called salting out.

8) Concentration of reaction products: Accumulation of reaction products causes lowering of the enzyme activity. This is prevented by nature by prompt removal of the products from the site of formation. For e.g. absorption of the products of digestion from gastro intestinal tract into blood stream.

Q.18 Define Biotechnology. Give the Application of biotechnology in Agriculture.

Ans: Biotechnology- Bio means life and technology means the application of knowledge for practical use ie., the use of living organisms to make or improve a product.

Applications of biotechnology in agriculture (plants)

A. Crop Improvement

 Plants with built in resistance to pest and Diseases.

 Plants with built in tolerance to environmental conditions

 Improved color and quality

B. Pharmaceuticals

 Plants that produce edible vaccines

C. Food

 Improved taste and nutrition

 Improved handling qualities

D. Industrial

 plants that produce plastics, fuels, and other products

 plants for environmental cleanup

E. Other

 pesticides made from naturally-occurring microorganisms and insects

Applications of biotechnology in agriculture (animals)

A. Food

 Increased milk production

 growth hormones in farm-raised fish that result in earlier market-ready fish

B. Pharmaceuticals

 Animals engineered to produce human proteins for drugs, including insulin

and vaccines

C. Breeding

 Disease tolerance

 Exact copies of desired stock

 Increased yields

D. Health

 Microorganisms introduced into feed for beneficial purposes

 Diagnostics for disease and pregnancy detection

 Animals engineered to produce organs suitable for transplantation into

Humans.

Q.19. What is PCR technique? Give its applications.

Ans: Polymerase chain reaction (PCR) is a method widely used in molecular biology to make multiple copies of a specific DNA segment. PCR was developed by Kary Mullis.

Steps for PCR Techniques:

There are three main stages:

Denaturing – when the double-stranded template DNA is heated to separate it into two single strands.

Annealing – when the temperature is lowered to enable the DNA primers to attach to the template DNA.

Extending – when the temperature is raised and the new strand of DNA is made by the Taq polymerase enzyme.

These three stages are repeated 20-40 times, doubling the number of DNA copies each time. A complete PCR reaction can be performed in a few hours, or even less than an hour with certain high-speed machines. After PCR has been completed, a method called electrophoresis can be used to check the quantity and size of the DNA fragments produced.

Applications of PCR:

1. For various agro product development.

2. In identification of different cultivars of rice.

3. For gene discovery and cloning.

4. Seed quality Control.

5. It has great spectrum of genetic research.

6. To determine specific location of gene.

7. in diagnosis of genetic Disorder.

8. Development of GM crops.

Q.21 What are the types of tissue culture?

Ans:

Embryo Culture: Embryo culture is the type of tissue culture that involves the isolation of an embryo from a given organism for in vitro growth. Embryo culture may involve the use of a mature of immature embryo. Whereas mature embryos for culture are essentially obtained from ripe seeds, immature embryo (embryo rescue) involves the use of immature embryos from unripe/hybrid seeds that failed to germinate. In doing so, the embryo is ultimately able to produce a viable plant. For embryo culture, the ovule, seed or fruit from which the embryo is to be obtained is sterilized, and therefore the embryo does not have to be sterilized again. Salt sucrose may be used to provide the embryo with nutrients. The culture is enriched with organic or inorganic compounds, inorganic salts as well as growth regulators.

Callus Culture : Callus - This is the term used to refer to unspecialized, unorganized and a dividing mass of cells. A callus is produced when explants (cells) are cultured in an appropriate medium - A good example of this is the tumor tissue that grows out of the wounds of differentiated tissues/organs. In practice, callus culture involves the growth of a callus (composed of differentiated and non- differentiated cells), which is the followed by a procedure that induces organ differentiation. For this type of tissue culture, the culture is often sustained on a gel medium, which is composed of agar and a mixture of given macro and micronutrients depending on the type of cells. Different types of basal salt mixtures such as murashige and skoog medium are also used in addition to vitamins to enhance growth.

Organ Culture: Organ culture is a type of tissue culture that involves isolating an organ for in vitro growth. Here, any organ plant can be used as an explant for the culture process (Shoot, root, leaf, and flower). With organ culture, or as is with their various tissue components, the method is used for preserve their structure or functions, which allows the organ to still resemble and retain the characteristics they would have in vivo. Here, new growth (differentiated structures) continues given that the organ retains its physiological features. As such, an organ helps provide information on patterns of growth, differentiation as well as development. There are number of methods that can be used for organ culture. These include;

1.Plasma clot method 2.Raft method 3.Agar gel method 4. Grid method.

Cell Suspension Culture: The growing of individual cells that have been obtained from any kind of explant tis-sue or callus referred to as cell suspension culture. These are initiated by transferring pieces of tissue explant/callus into liquid medium (without agar) and then placed them on a gyratory shaker to provide both aeration and dispersion of cells. Like callus culture, the cells are also sub-cultured into new medium. Cell suspension cultures may be done in batch culture or continuous culture system. In the later system, the culture is continuously sup-plied with nutrients by the inflow of fresh medium with subsequent draining out of used medium but the culture volume is constant. This culture method is mainly used for the synthesis of specific metabolite or for biomass production.

Anther Culture: An important aspect of plant tissue culture is the haploid production by another cul-ture or pollen culture which was first established **by Guha and Maheswari** (1964, 1966) in Datura. During the last few decades, much progress has been made in different crops like rice, wheat, maize, mustard, pepper and others. The anthers bearingthe uni-nucleate microspores are selected and allowed to grow in medium to

produce callus from the pollen mass. Then the triggering of these androgenic calli is directed to produce the embryos and haploid plants are developed from these androgenic embryos. The anther culture can be done with the isolated anthers on solid medium where anther wall will break open and the androgenic calli will be formed from the pollen. In pollen culture, microspores of uni--nucleate stage are collected in liquid media and can be grown in suspension culture. In suspension, the uni-nucleate pollens may give rise to calli mass or the globular mass from which the plants can be raised either through embryogenic or organogenic path-way.

Pollen Culture: Microspore or the immature pollen can be used as the explant to get the haploid plants directly. For pollen or microspore culture the flower buds are collected, surface sterilized and the anther lobes are dissected out from the flower buds as before. Then the anther lobes are squeezed with the help of a scalpel within a tube or small beaker to col-lect the microspore or pollen in nutrient media. Then the anther tissue debris is removed by filtering the suspension through a nylon sieve with a diameter slightly larger than the pollen size (40µ-100µ) allowing the microspore only to pass through it. Then the microspore-suspension washed and concentrated to a plating density. The microspores obtained are then mixed with an appropriate culture medium at a density of 103— 104 microspore/ml, and plated in small petriplate. To ensure good aeration, the layer of liquid in the dish should be as thin as possible, and sealed with 'parafilm' to avoid dehydration. The responsive pollen will divide and form embryos or calli which directly or indirectly will form the haploid plantlet. By following the method of sub-culturing the whole plant suitable for soil transfer can be obtained.

Advantages of pollen culture:

i) The explants i.e., microspores or pollens are all haploid cells.

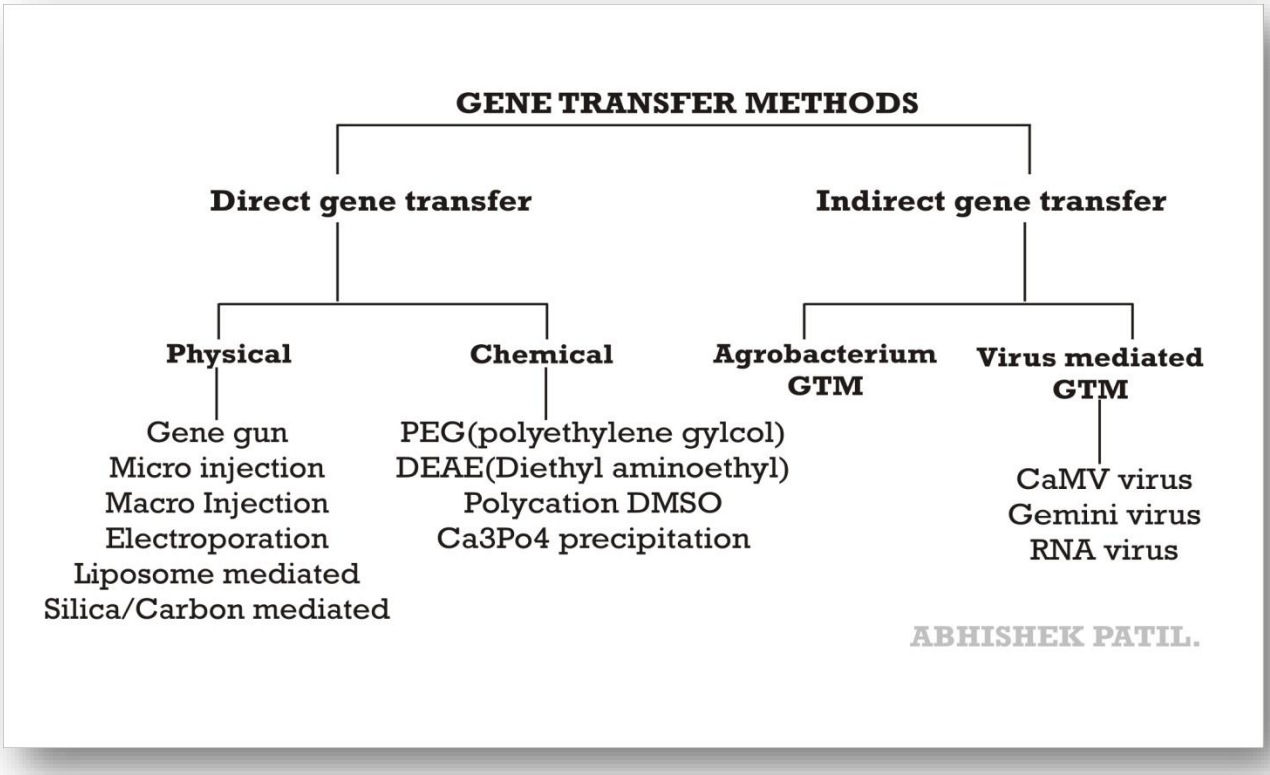
(ii) The sequence of androgenesis can be observed starting from a single cell.

(iii) The microspores are ideal for uptake, transformation and mutagenic studies, and the microspores are evenly exposed to chemicals and physical mutagens.

(iv) Higher yields of plants/anther could be obtained.

Q.22 What are the different methods for gene transfer? Describe Gene gun method.

Ans:



The gene gun method is a method used for genetically modifying plants.

John Cornell Sanford, Klein and colleagues developed the "Biolistic Particle Delivery System" or so-called "gene gun".

The gene gun method delivers extra DNA directly into a plant's nucleus. The method is also commonly called particle acceleration or microprojectile bombardment. The gene gun can be used on seedlings or tissue culture cells. Prior to injecting the DNA into the plant tissue via the gene gun, either microscopic gold or tungsten particles are liberally coated with many hundreds of copies of genes. The particles are then forced into the nucleus with the gene gun.

Advantages of Gene Gun Method

- The gene gun is highly effective at modifying the DNA and genetics of plant cells.
- Genetic modification in plants can make them resistant to drought, disease, and pests. It also helps create a more nutritious edible plant with higher levels of proteins.
- Method is easy to use, rapid and versatile
- Transient or stable expression is possible
- Small amounts of nucleic acids and few cells are required for efficient transformation
- Large DNA fragments may be transferred as well as small interfering RNA's for gene silencing

Disadvantages :

Cause cell damage.

Frequency of transformation is low.

Integration is low.

Requirement of equipment and personal skill.

Q.23. Write a short note on Organogenesis.

Ans: Organogenesis: In plant tissue culture, organogenesis means genesis of organs like shoots, roots, leaves, flowers, etc.

Types of organogenesis:

Caulogenesis:

Type of organogenesis by which only adventitious shoot bud initiation take place in the callus tissue.

Rhizogenesis:

Type of organogenesis by which only adventitious root formation takes place in the callus tissues.

CauloRhizogenesis:

It is the comination of both caulogenesis and rhizogenesis.

Q.24.What is Embryo rescue? Write down its significance?

Ans: Embryo rescue is a process carried out in distant (interspecific or intergeneric) hybridization where endosperm development is poor, shortening of breeding cycle, etc.

In the embryo culture technique which is the nutritional relationship between the embryo and endosperm is restored by providing the artificial medium to induce and complete growth of hybrid embryos is called as embryo rescuing.

Significance:

- It is used to overcome embryo abortion.
- Used to overcome seed dormancy.
- It enables transfer of resistant genes for pests and diseases and various environmental stresses into the cultivated species.
- Used to produce not only interspecific hybrids but also intergeneric hybrids.

Q.25. What is gene cloning? Enlist the steps involved in gene cloning.

Ans: Gene cloning is the act of making copies of a single gene.

Steps for Gene Cloning:

1) Identification & Isolation of DNA Fragments to be Cloned.

2) Amplification of specific gene through PCR.
3) Cutting & Joining of DNA.
4) Insertion of Isolated DNA into the Suitable Vector to Form the Recombinant DNA.
5) Introduction of the Recombinant DNA into a Suitable Organism known as Host:
6) Selection of the Transformed Host Cells and Identification of the Clone Con-taining the Gene of Interest
7) Multiplication/Expression of the Introduced Gene in the Host.

Q.26. What is somaclonal variation? Give its use in crop improvement.

Ans: Somaclonal Variation**:** According to Larkin and Scowcroft (1981), "Somaclonal variation is the genetic variability which is regenerated during tissue culture" or plant variants derived from any form of cell or tissue cultures.

Use in Crop Improvement:

1. Somaclonal variation and gametoclonal variation represent useful source of introducing genetic variations that could be of value to plant breeders.

2. Single gene mutation in nuclear or organelle genome may give the best available variety in vitro that has a specific character.

3. Gametoclonal variation, induced mostly by meiotic recombination during the sexual cycle of F1 hybrid, results in trasngressive segregation to uncover unique gene combination.

4. Various cell lines selected in vitro may prove potentially applicable to agriculture and industry like resistance to herbicide, pathotoxin, salt or aluminium.

5. Variability in cell cultures has played a useful role in synthesis of secondary metabolites on a commercial scale.

6. Technique employed for Somaclonal and gametoclonal variation are relatively easier than recombinant DNA technique.

Somaclonal variants for agronomically desirable traits in several crop plants have been raised from tissue culture. Some examples of Somaclonal variation in crop plants as well as in some horticulturally important plants are given below:

Rice: High modification for seed weight, seed proteins percentage, tiller number, panicle length and time of flowering. At IRRI, mutants were observed for many characters such as panicle, grain, and leaf morphology and tiller arrangement.

Wheat: Variations were manifested for gliadin proteins in seed, grain colour, plant height, heading date and yield.

Maize: Plants regenerated from selected cell lines were resistant both to T-toxin and to infection to Drechslera maydis causing southern leaf blight. Cytoplasmic male sterile lines are very sensitive to the T-toxin produced by Drechslera maydis.

Potato: Somaclonal variants were selected for resistance to Phytopthora infestans and to its multiple races and resistance to early blight.

Tomato: Somaclones were isolated with variant phenotypes, such as recessive mutation for male sterility, resistance to Fusarium oxysporium, jointless pedicel , tangerine virescent leaf, flower and fruit colour.

Sugarcane: Somaclonal variants have been isolated by different workers for cane yield, sugar yield and resistance to smut disease caused by Ustilago scitamini, downey mildew caused by Helminthosporium sacchari.

Q.27 Write down the properties of buffer,pH, Water.

Ans:

Properties of buffer:

Since Good buffers are often used in research involving living cells, they are required to be non-toxic to the cells used in the experiment.

Good buffer is also resistant to non-enzymatic degradation by other components of the setup.

Good buffers will not pass through cell membranes.

Good buffers have a high solubility in water, since most biological systems naturally use water as their solvent. Also, the solubility level of Good buffers in organic solvents such as fats and oils is low.

Good buffer is in the range corresponding to a pH range of 6 to 8.

Properties of pH:

The pH scale measures how acidic or basic a substance is.

The pH scale ranges from 0 to 14.

A pH of 7 is neutral.

A pH less than 7 is acidic.

A pH greater than 7 is basic.

The pH scale is logarithmic and as a result, each whole pH value below 7 is ten times more acidic than the next higher value.

Properties of Water:

Water (H_2O) is a polar inorganic compound.

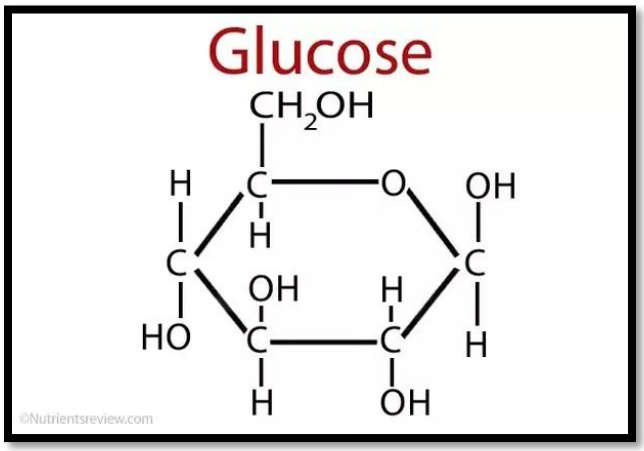
Water has no color.

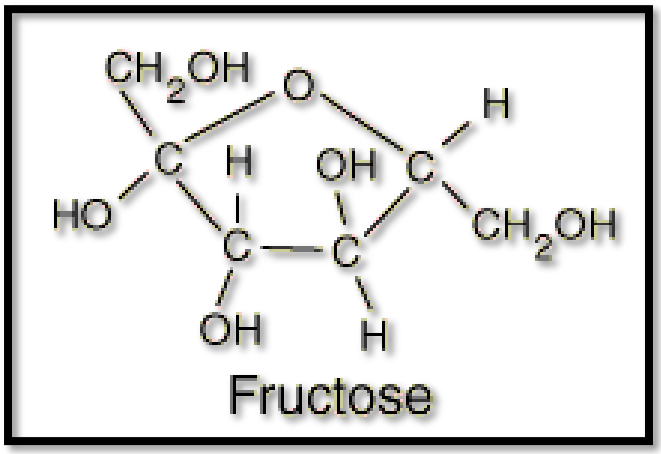
Water has no odor.

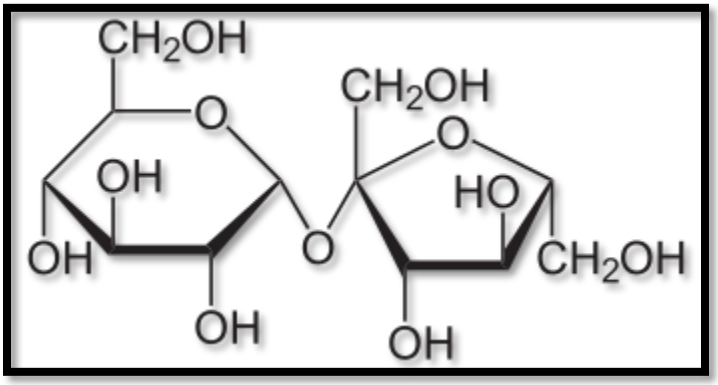
It is Universal Solvent.

Water has properties of both acid and base.

STURCTURES OF MOLECULES







SUCROSE

THANK YOU