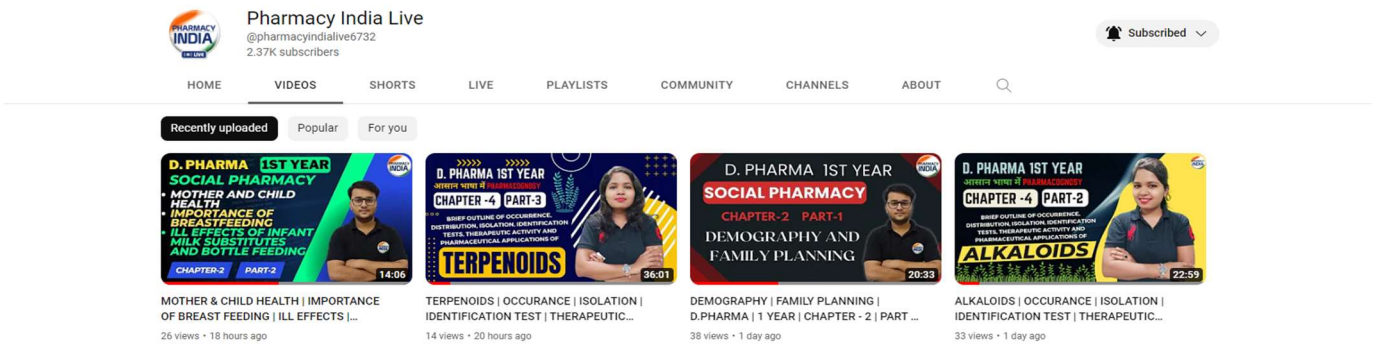
BIOCHEMISTRY AND CLINICAL PATHOLOGY

MODEL PAPER – 1

Syllabus to be covered in this module are-

- ❖Chapter-1 Introduction to Biochemistry
- ❖Chapter-2 Carbohydrates
- ❖Chapter-3 Proteins
- ❖Chapter-4 Lipids



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Questions

Long Questions-

Ques.1Mention the qualitative analysis of Carbohydrates.

Ques.2 Discuss in detailed about structure of proteins.

Ques.3 Write in detailed about proteins.

Ques.4 Define lipids. Give the classification of lipids.

Ques.5 Explain in detailed about chemical properties of lipids.

Ques.6 Give the Haworth Representation of the structure of glucose, fructose, and galactose.

Short Questions

Ques.1 What are protein deficiency treatments.

Ques.2 Give the biological role amino acids.

Ques.3 What are the qualitative test for proteins.

Ques.4 Give the diseases related to malnutrition of proteins.

Ques.5 Give the biological functions of lipids.

Ques.6 Mention the specific test for cholesterol.

Ques.7 Define lipoproteins. Mention the types and functions of lipoproteins.

Ques.8 Write a short note on role of lipids.

Ques.9 Write down the characteristics of lipids.

Ques.10 Write a short note on polysaccharides.

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Long Answers

Ques.1Mention the qualitative analysis of Carbohydrates.

Ans- Qualitative Analysis of Carbohydrates

Based on the reactivity with Tollen's, Benedict's or Fehling's reagent, carbohydrates are classified as:

Reducing sugars

Carbohydrates that can reduce Tollen's, Benedict's or Fehling's reagents are called reducing sugars (sugar with free aldehyde or ketone group). All monosaccharides and most of the disaccharides are reducing sugars. Some examples are Maltose and Lactose.

Non-reducing sugars

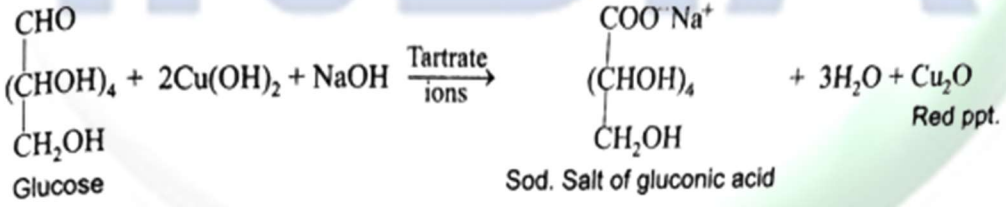
Carbohydrates that cannot reduce Tollen's, Benedict's or Fehling's reagents are called non-reducing sugars. Sucrose is a non-reducing sugar. Some Important Tests for the Detection of Carbohydrates.

Molisch's test

Molisch's reagent is 10% alcoholic solution of a-naphthol. This is a common chemical test to detect the presence of carbohydrates. Carbohydrates undergo dehydration by sulphuric acid to form furfural (furfuraldehyde) that reacts with a-naphthol to form a violet-coloured product.

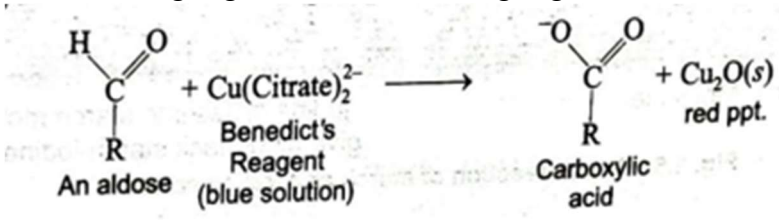
Fehling's test

This is an important test to detect the presence of reducing sugars. Fehling's solution A is copper sulphate solution and Fehling's solution B is potassium sodium tartrate. On heating, carbohydrate reduces deep blue solution of copper (II) ions to red precipitate of insoluble copper oxide.



Benedict's test

Benedict's test distinguishes reducing sugar from non-reducing sugar. Benedict's reagent contains blue copper (II) ions ions) that are copper (1) ions (Cu, carbohydrates. These precipitate as red (copper (I) oxide.



(Cu2+, cupric reduced to cuprous ions) by ions form coloured cuprous

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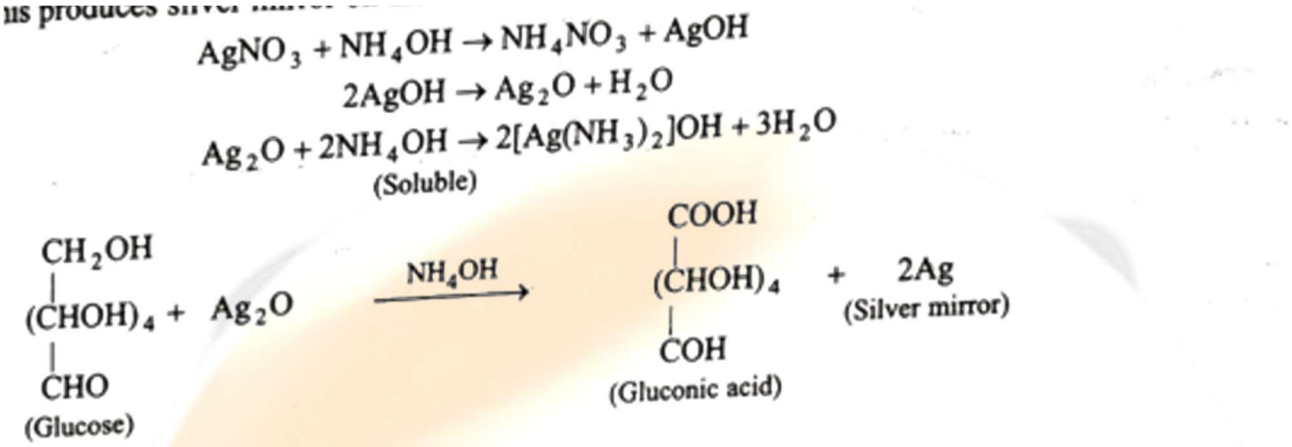
Tollen's test

Tollen's reagent is ammonical silver nitrate solution. On reacting with carbohydrate elemental silver is precipitating out of the solution, occasionally onto the inner surface of the reaction vessel. This produces silver mirror on the inner wall of the reaction vessel.



Iodine Test

Iodine test is used to detect the presence of starch. Iodine is not much soluble in water so iodine ed solution is prepared by dissolving iodine in water in presence of potassium iodide. Todine dissolved in an aqueous solution of potassium iodide reacts with starch to form a starch/iodine complex which gives characteristics blue-black colour to the reaction mixture.

Barfoed's Test

Barfoed's reagent, cupric acetate in acetic acid is slightly acidic and is balanced so that it can only be reduced by monosaccharides but not less powerful reducing sugars. Disaccharides may also react with this reagent, but the reaction is much slower when compared to monosaccharides. Perform this test with glucose, maltose, and sucrose.

Procedure:

To 1-2 mL of Barfoed's reagent, add an equal volume of sugar solution. Boil for 5 min. in a water bath and allow to stand.

Seliwanoff's Test:

Seliwanoff's Test distinguishes between aldose and ketose sugars. Ketoses are distinguished from aldoses via their ketone/aldehyde functionality. If the sugar contains a ketone group, it is a ketose and if it contains an aldehyde group, it is an aldose. This test is since, when heated, ketoses are more rapidly dehydrated than aldoses. This test with glucose, fructose, maltose, and sucrose.

Procedure:

Heat 1 mL of sugar solution with 3 mL. Seliwanoff's reagent (0.5 g resorcinol per HCI) in boiling water.

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In less than 30 seconds, a red color must appear for ketoses. ◆ Upon prolonged heating, glucose will also give an appreciable color.

Bial's Test

Bial's Test is to determine the presence of pentoses (5C sugars). The components of this reagent are resorcinol, HCI, and ferric chloride. In this test, the pentose is dehydrated to form furfural and the solution turns bluish and a precipitate may form. Perform this test with ribose and glucose.

Procedure:

To 5 mL of Bial's reagent, add 2-3 drops of sugar solution and boil. Upon boiling, note the green-blue color formed.

Ques.2 Discuss in detailed about structure of proteins.

Ans- Structure of Proteins

Most proteins fold into unique 3-dimensional structures. The shape into which a protein naturally folds is known as its conformation.

The sequence of amino acids in a protein is called its primary structure. Within a chain the atoms are held together by covalent bonds. Each protein has its own characteristic sequence of amino acids.

Three types of bonding can happen within a protein molecule (intramolecular bonding) and between protein molecules (intermolecular bonding):

- Hydrogen bonds
- Covalent bonds
- Ionic bonds

Protein chains arrange themselves to maximise the intra- and intermolecular bonding. The structure when protein chains are held in place is called the **secondary structure.** This may be:

- helical, e.g., keratin (the protein found in hair), or
- pleated sheet, e.g. fibroin (the protein found in silk)

These structures are held in place by hydrogen bonds.
Protein chains may fold into a globular shape. This is the tertiary structure of a protein. These globular proteins include enzymes and immunoglobins. The structures are held in place by hydrogen bonds, disulphide bridges and ionic bonds. Finally, some proteins have a quaternary structure. These contain more than one protein chain.

Examples are insulin and haemoglobin.

Proteins are naturally occurring polypeptides. They:

- contribute to the mechanical structure of animals, including humans, e.g., keratin in hair and fingernails, and fibrous proteins such as collagen in tendons

- enable animals to move, e.g., myosin in muscle.

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- facilitate transport of smaller molecules around animals' bodies, e.g., haemoglobin.

- control the types and rates of chemical reactions in living things, then they are called enzymes, e.g., amylase. are important components of the human immune system, e.g., immunoglobins.

Primary structure

- The primary structure is the amino acid sequence of the proteins, their lowest level of organization, it is unique and genetically determined.
- It may consist of 40 to over 4,000 amino acid residues and it determines the three-dimensional structure of the protein itself, which in turn determines its function.
- The polypeptide chain has polarity because its two ends are different: one has a free amino group and is called NH_2-terminus or amino-terminus, the other a free carboxyl group, and is called COOH-terminus or carboxyl-terminus.

Secondary structure

The long protein chains there are regions in which the chains are organized into regular structures known as alpha-helices (alpha-helixes) and beta-pleated sheets. These are the secondary structures in proteins. These secondary structures are held together by hydrogen bonds. Although the hydrogen bonds are always between CO and H-N groups, the exact pattern of them is different in an alpha-helix and a beta-pleated sheet.

The alpha-helix-In an alpha-helix, the protein chain is called like a loosely-coiled spring. Beta-pleated sheets-In a beta-pleated sheet, the chains are folded so that they lie alongside each other. They are antiparallel to each other.

Tertiary Structure

The overall three-dimensional shape of an entire protein molecule is the tertiary structure. The protein molecule will bend and twist in such a way as to achieve maximum stability or lowest energy state.

The tertiary structure will have a single polypeptide chain "backbone" with one or more protein secondary structures, the protein domains. Amino acid side chains may interact and bond in several ways.

Quaternary structure

Quaternary structure is the arrangement of more than one protein molecule in a multi-subunit complex.

Quaternary structure refers to the association of multiple individual protein chains into a single protein with multiple subunits. The arrangement of the subunits gives rise to a stable structure. In

- The subunits may be identical or different quaternary protein structure:

- When they are different, each subunit tends to have a different function.

The final shape of the protein complex is once again stabilized by various interactions, including hydrogen-bonding, disulphide-bridges and salt bridges.

Ques.3 Write in detailed about proteins.

Ans- Definition

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Proteins are large biomolecules, or macromolecules, consisting of one or more long chains of amino acid residues. Proteins perform a vast array of functions within organisms, including catalysing metabolic reaction.

A linear chain of amino acid residues is called a polypeptide. A protein contains at least one long polypeptide. Short polypeptides, containing less than 20-30 residues, are rarely considered to be proteins, and are commonly called peptides, or sometimes oligopeptides. The individual amino acid residues are bonded together by peptide bonds and adjacent amino acid residues.

Characteristics and Properties of Proteins

- Proteins are organic substances; they are made up of nitrogen, oxygen, carbon and hydrogen and sulphur.
- Proteins are the most important biomolecules; they are the fundamental constituent of the cytoplasm of the cell.
- Proteins are the structural elements of body tissues. Proteins are made up of amino acids.
- Proteins give heat and energy to the body and aid in building and repair.
- Only small amounts of proteins are stored in the body as they can be used up quickly on demand.
- Proteins are considered as the bricks, they make up bones, muscles, hair and other parts of the body.

Classification of Proteins

Proteins are grouped based on their composition and structure:

Based on Composition

(i) Simple proteins

(ii) Conjugated proteins

(iii) Derived proteins

(i) Simple proteins or holoproteins: These proteins are made of only one type of amino acid, as structural component, on decomposition with acids, they liberate constituent amino acids.

They are mostly globular type of proteins.

They are further sub-classified according to solubility as follows:

(a) Protamines and histones: These proteins occur only in animals and are basic proteins. They possess simple structure and low molecular, are water soluble and are not coagulated by heat They are strongly basic in character due to the high content of lysine, arginine. Examples: Protamines- salmine, clupine, cyprinine; Histones-nucleohistones, globin.

(b) Albumins: They are widely distributed in nature, mostly seen in seeds. They are soluble in

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water and dilute solutions of acids, bases, and salts. Examples; Leucosine, legumeline, serum albumin.

(c) Globulins: They are of two types, pseudoglobulins which are soluble in water, other is euglobulins which are insoluble in water. They are coagulated by heat. Examples Pseudoglobulin, serum globulin, glycinine etc.

(d) Scleroproteins or Albuminoids: These occur mostly in animals and are commonly known as animal skeleton proteins, they are insoluble in water, and in dilute solution of acids, based and

(ii) Conjugated or complex or hetero-proteins: These are proteins that are made of salts amino acids and other organic compounds. The non-amino acid group is termed as prosthetic group.

Complex proteins are further classified based on the type of prosthetic group present.

Metalloproteins: These are proteins linked with various metals. Examples: casein, collagen, ceruloplasmin.

Chromoproteins: These are proteins that are coupled with a coloured pigment. Examples myoglobin, hemocyanin, cytochromes, flavoproteins.

Glycoproteins and Mucoproteins: These proteins contain carbohydrates as the prosthetic group. Examples: Glycoproteins egg albumin, serum globulins, serum albumins; Mucoproteins-Ovomucoid, mucin.

Phosphoproteins: These proteins are linked with phosphoric acid. Example: casein. Lipoproteins: Proteins forming complexes with lipids are lipoproteins. Examples lipovitellin, lipoproteins of blood.

Nucleoproteins: These are compounds containing nucleic acids and proteins. Examples: Nucleoproteins, nucleohistones, nuclein.

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

Derived proteins are of two types, primarily derived proteins and secondary derived proteins. The primary derived proteins are derivatives of proteins, in which the size of the protein molecule is not altered materially, while in secondary derived proteins, hydrolysis occurs, as a result the molecules are smaller than the original protein.

Based on Solubility

They are grouped under two categories as globular and fibrous:

(i) Globular Proteins: Globular proteins have axial ratio less than 10. They are compactly folded, coiled and possess a relatively spherical or ovoid shape. They are usually soluble in water and in aqueous media. Examples: Insulin, plasma albumin, globulin.

(ii) Fibrous Proteins: These proteins have axial ratio more than 10, hence, they resemble long ribbons or fibrous in shape. They are mostly found in animals, and are not soluble in water or in solution of dilute acids. Fibrous proteins aid in protection and structural support. Examples: Collagen, Keratin, Elastins, Fibroin.

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Ques.4 Define lipids. Give the classification of lipids.

Ans- Lipids are naturally occurring organic compounds, commonly known as oils and fats. Lipids occur throughout the living world in microorganisms, higher plants, and animals and also in all cell types. Lipids contribute to cell structure, provide stored fuel, and also take part in many biological processes.

Lipids are naturally occurring hydrophobic molecules. They are heterogeneous group of compounds related to fatty acids. They include fats, oils, waxes, phospholipids, etc. They make up about 70% of the dry weight of the nervous system. Lipids are crucial for the healthy functioning of the nerve cells. Lipids are greasy or oily organic substances; lipids are sparingly soluble in water and are soluble in organic solvents like chloroform, ether, and benzene.

Classification of Lipids

(1) Simple Lipids

Simple lipids are esters of fatty acid linked with various alcohols.

(a) Fats and oils (triglycerides, triacylglycerols)

These esters of fatty acid have glycerol, a trihydroxy alcohol. Fat is solid at room temperature, while oil is in liquid form. Triglycerides are abundant and constitute about 98 percent of all dietary lipids. The rest consists of cholesterol, its esters and phospholipids. Unlike carbohydrates, which can be stored only for a short time in the body, triglycerides are stored in the body in large amounts as body fat, which can last for years.

An average man weighing about 70 kg, has at least 10 to 20 percent of his body weight in lipid most of which is triacylglycerol. This is found in adipose (fat) tissue, as well as all other organs of the body. Body fat is a reservoir of chemical energy. Simple lipids are the esters of fatty acids with various alcohols.

(b) Simple Triglycerides: Simple triglycerides are one in which three fatty acids radicals a similar or are of the same type. Example: Tristearin, Triolein.

(c) Mixed Triglycerides are one in which the three fatty acids radicals are different from each other. Example: distearo-olein, dioleo-palmitin.

(d) Waxes are the esters of fatty acids with high molecular weight monohydroxy alcohols. Example: Beeswax, Carnauba wax.

Waxes are long-chain saturated and unsaturated fatty acid esters with monohydroxy alcohols, which have high molecular weight.

(2) Compound lipids: (Complex lipids) Heterolipids

These are another classification of lipids. Heterolipids are fatty acid esters with alcohol and additional groups. On hydrolysis gives phosphoric acid, various sugars, sphingosine, ethanolamine, and serine in addition to fatty acids and glycerol. These heterolipids are further classified as:

(a) Phospholipids (phosphatids)

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(i) Phospholipids or Phosphatids are compounds containing fatty acids and glycerol in addition to a phosphoric acid, nitrogen bases and other substituents. They usually possess one hydrophilic head and two non-polar tails. They are called polar lipids and are amphipathic in nature.
Phospholipids can be phosphoglycerides, phosphoinositides and phosphosphingosides.

(ii) Phosphoglycerides are major phospholipids, they are found in membranes. It contains fatty acid molecules which are esterified to hydroxyl groups of glycerol. The glycerol group also forms an ester linkage with phosphoric acid. Example: Lecithin, Cephalins.

(iii) Phosphoinositides are said to occur in phospholipids of brain tissue and soybeans. They play important role in transport processes in cells.

(iv) Phosphosphingosides are commonly found in nerve tissue. Example: sphingomyelins.

(b) Glycolipids (cerebrosides) Glycolipids are fatty acids

Glycolipids also include some compounds like sulfolipids, gangliosides, and sulfatids which are structurally-related. with carbohydrates and nitrogen but without phosphoric acid.

These cerebrosides are important constituents of the brain and other tissues. They consist of at least one sugar unit, so they are also called glycosphingosides. They are like phospholipids because they have a hydrophobic region, with a polar region and two long hydrocarbon tails. Like phospholipids, glycolipids form lipid bilayers that are self-sealing and form the structure of cellular membranes.

(3) Derived Lipids

These substances are derived by hydrolysis from compound and simple lipids. These fatty acids include alcohols, mono- and diglycerides, carotenoids, steroids, and terpenes. Hydrolytic products of simple and compound lipids **(i) Alcohols Glycerol and other sterol**

(ii) Fatty acids (a) Steroids: The steroids are biological compounds that are some of the most studied types of fat.

They contain no fatty acids and unlike fats, are no saponifiable (cannot be hydrolysed to yield soap). → Cholesterol is a well-studied lipid, because of its strong correlation with the incidence

(i) Cholesterol:

- ❖ Cholesterol is a well-studied lipid, because of its strong correlation with the incidence cardiovascular disease.
- ❖ It is an important component of cell membranes and plasma lipoproteins, and is an important precursor of many biologically important substances like bile acids and steroid hormones.
- ❖ It is abundant in nerve tissues and is associated with gallstones.
- ❖ Dietary cholesterol is found in saturated fats of animals (as butter and lard), but vegetable oils do not contain cholesterol.
- ❖ Only a small portion of your body cholesterol comes from the diet. Most of it is produced in the body.

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- Eating unsaturated fatty acids from vegetable oil helps lower blood cholesterol levels by reducing cholesterol synthesis in the body.

- However, eating saturated fats from animal fat elevates blood cholesterol and triglycerides and reduce the ratio of your good to bad cholesterol.

(b) Terpenes in majority are found in plants. Example: Natural rubber, gernoil, etc.

(c) Carotenoids are tetraterpenes. They are widely distributed in both plants and animals. They are exclusively of plant origin. Due to the presence of many conjugated double bonds, they are colored red or yellow. Example: Lycopene, carotenes, Xanthophylls.

(iii) Terpenoids

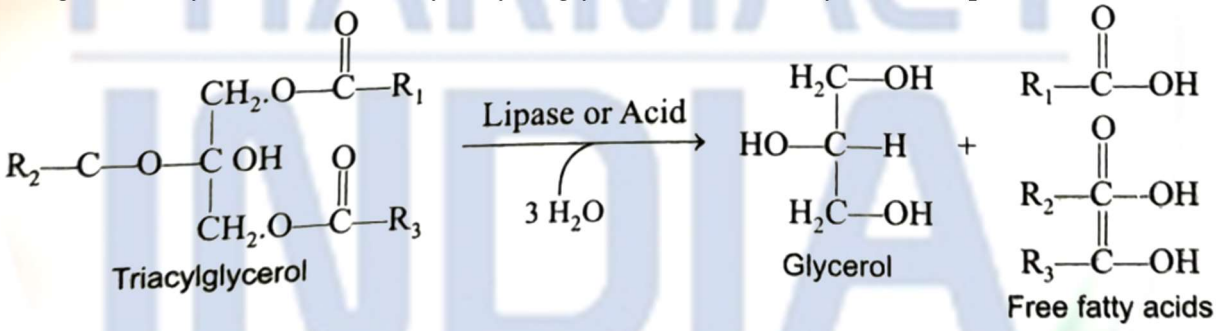
Ques.5 Explain in detailed about chemical properties of lipids.

Ans- CHEMICAL PROPERTIES OF LIPIDS

1. Hydrolysis:

- They are hydrolysed into their constituents (fatty acids and glycerol) by the action of super-heated steam, acid alkali or enzyme (e.g., lipase of pancreas).

- During their enzymatic and acids hydrolysis glycerol and free fatty acids are produced.



2. Hydrogenation

Unsaturated fatty acids may be converted to saturated fatty acids by the relatively simple hydrogenation reaction. Recall that the addition of hydrogen to an alkane (unsaturated) result in an alkane (saturated). A simple hydrogenation reaction is: alkene plus hydrogen yields an alkane

$$H_2C\text{-}CH_2+H_2=CH_3CH_3$$

Vegetable oils are commonly referred to as "polyunsaturated". This simply means that there are several double bonds present. Vegetable oils may be converted from liquids to solids by the hydrogenation reaction

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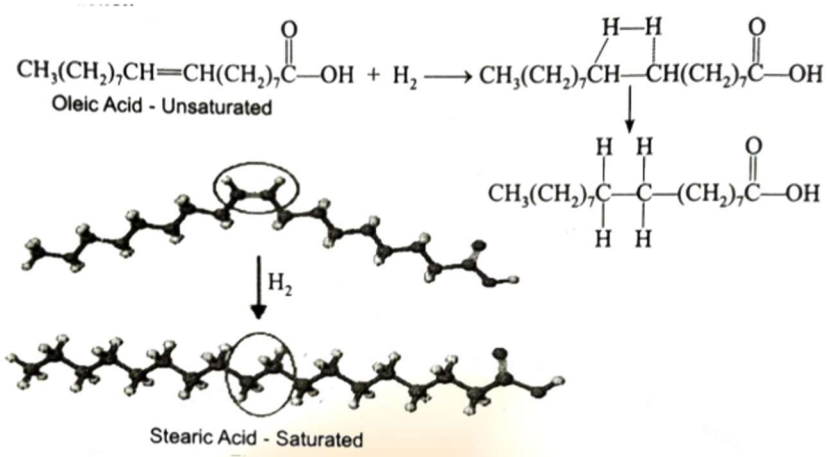
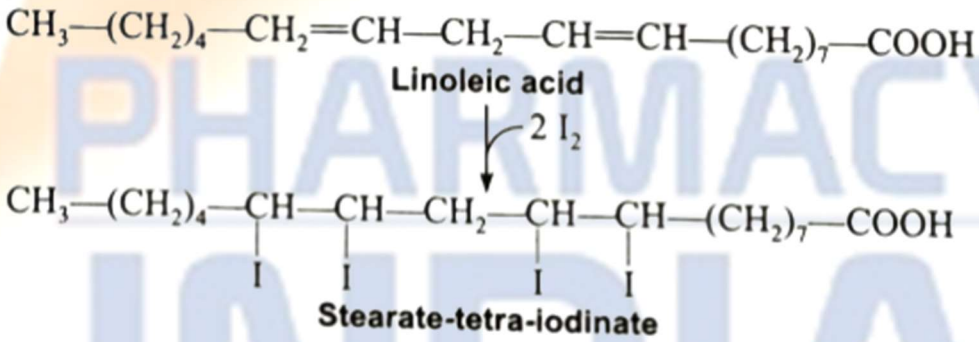


3. Halogenation

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



4. Saponification

- Saponification is the hydrolysis of fats or oils under basic conditions to form glycerol and the salt of the corresponding fatty acid.
- Saponification literally means "soap making". It is important to the industrial user to know the amount of free fatty acid present, since this determines in large measure the refining loss.
- Saponification (alkaline hydrolysis) is an important aspect of carotenoid analysis in foods where it is particularly effective for removing colourless contaminating lipid material and for destroying chlorophyll if present.

The saponification number is the number of milligrams of potassium hydroxide required to neutralize the fatty acids resulting from the complete hydrolysis of Ig of fat. It gives information concerning the character of the fatty acids of the fat - the longer the carbon chain, the less acid is liberated per gram of fat hydrolysed.

It is also considered as a measure of the average molecular weight (or chain length) of all the fatty acids present. The long chain fatty acids found in fats have low saponification value because they have a relatively fewer number of carboxylic functional groups per unit mass of the fat and therefore high molecular weight.

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5. Rancidity Oxidative and hydrolytic Rancidity

Rancidity is the complete or incomplete oxidation or hydrolysis of fats and oils when exposed to air, light, moisture or by bacterial action, resulting in unpleasant taste and odour. Specifically, it is the hydrolysis or autoxidation of fats into short-chain aldehydes and ketones which are objectionable in taste and odour. When these processes occur in food, undesirable odours and flavors can result. In certain cases, however, the flavors can be desirable (as in aged cheeses).

Three pathways for rancidification are:

Hydrolytic-Hydrolytic rancidity refers to the odour that develops when triglycerides are hydrolysed and free fatty acids are released.

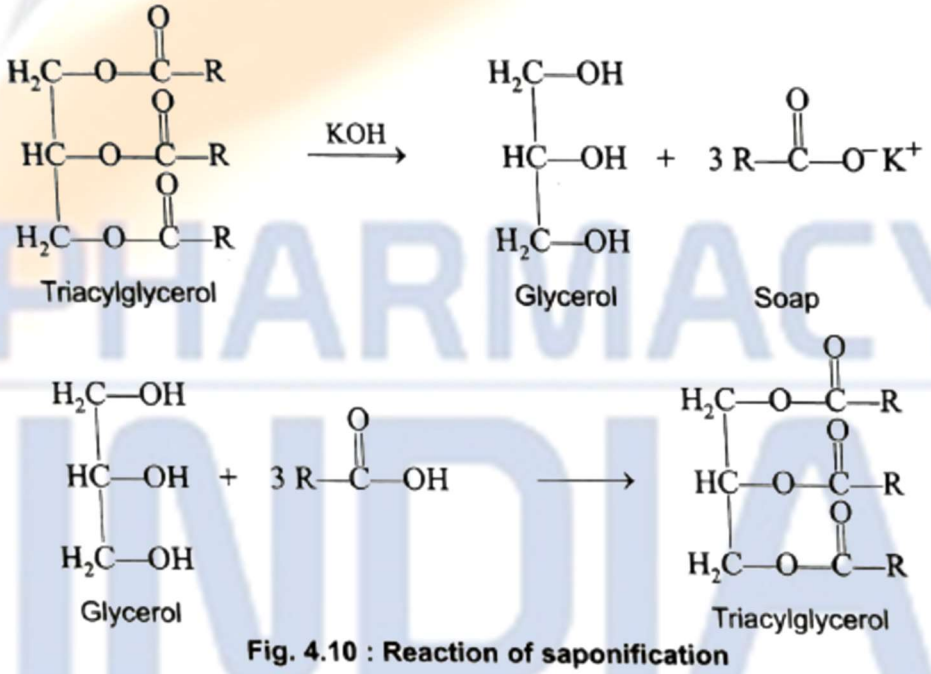

Fig. 4.10 : Reaction of saponification

Oxidative-oxidative rancidity is associated with the degradation by oxygen in the air. The double bonds of an unsaturaed fatty acid can be cleaved by free-radical reactions involving molecular oxygen. This reaction causes the release of malodorous and highly volatile aldehydes and ketones.

Microbial-Microbial rancidity refers to a process in which microorganisms, such as bacteria or molds, use their enzymes such as lipases to break down fat. This pathway can be prevented by sterilization.

6. lodine value

lodine value, also called lodine Number, in analytical chemistry, is the 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. Saturated oils, fats and waxes take up no iodine; therefore, their iodine value is zero; but unsaturated oils, fats and waxes take up iodine. (Unsaturated compounds contain molecules with double or triple bonds, which are very reactive toward iodine.) The more iodine is attached, the higher is the iodine value, and the more reactive, less stable, softer, and more susceptible to oxidation and rancidification is the oil, fat, or wax.

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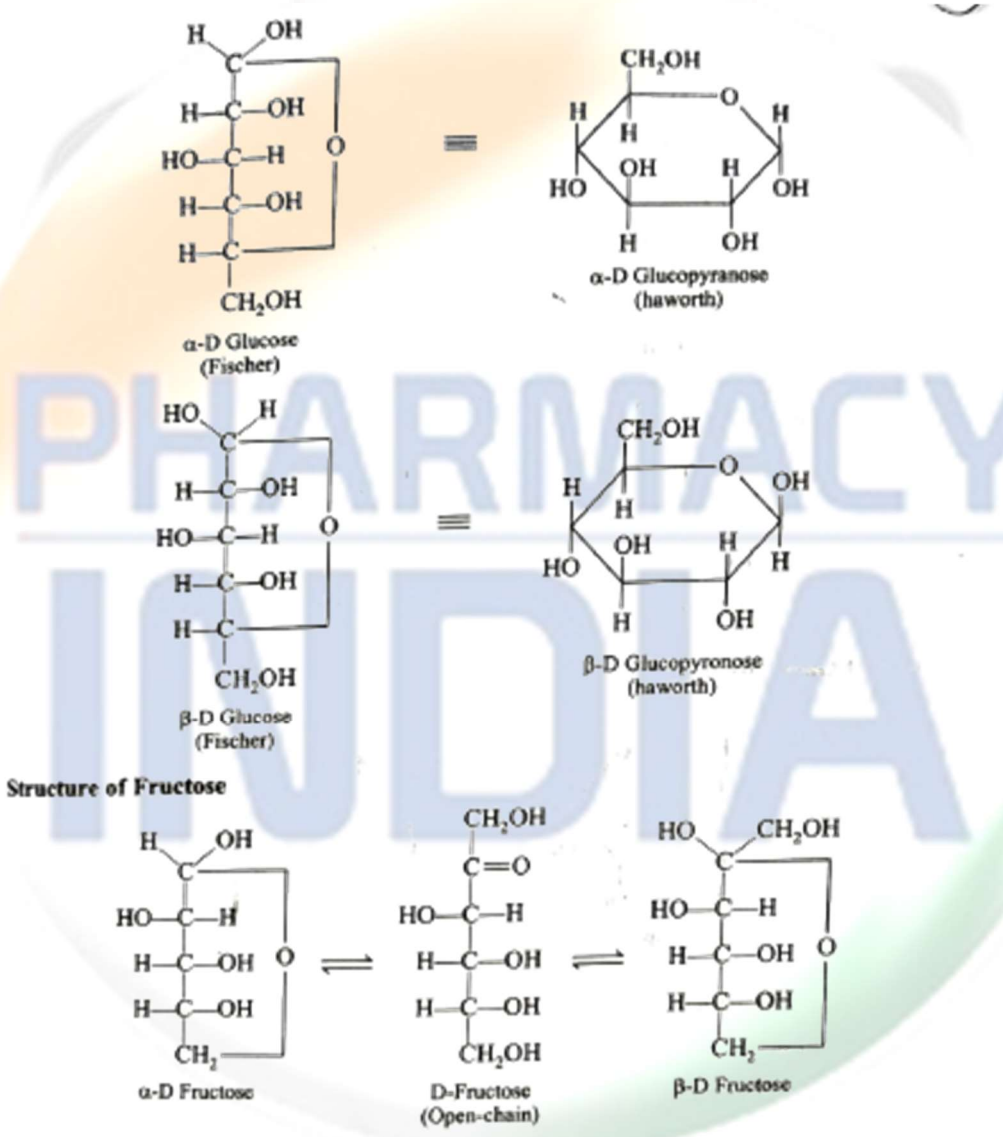
Ques.6 Give the Haworth Representation of the structure of glucose, fructose, and galactose.

Ans- Ring Form (Haworth Representation)

In Haworth formula, all the OH groups on the right in Fischer formula are directed below the plane of the ring, while these on the left go above the plane. The terminal CH,OH projects above the plane of the ring.



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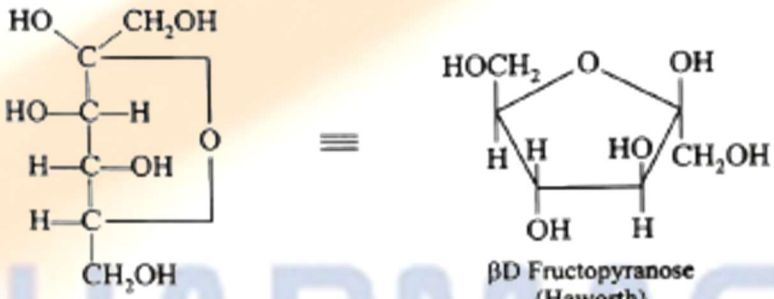
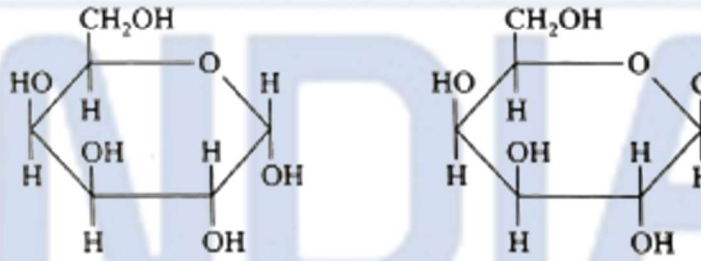
α-D Fructose (Fischer) ≡ αD Fructopyranose (Haworth)



β-D Fructose (Fischer) ≡ βD Fructopyranose (Haworth)

Structure of Galactose



α-D Galactopyranose β-D Galactopyranose

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Short Answers

Ques.1 What are protein deficiency treatments.

Ans- Protein Deficiency Treatments

1. Protein Supplements

The use of supplements depends on the extent of the condition as well as the effect on the deficiency. Supplementary nutrition is an addition to food intake, which enhances the dietary intake. The elements are crucial for efficient protein supply to the body.

2. Protein Rich Foods

Consumption of foods enriched with proteins is essential. These come in different varieties and should be taken daily for a healthy well-built body. Daily protein intake depends on the body's needs, and it varies from adults, children, and sick people.

There are high protein diets recommended for acute cases of protein deficiencies. This is an effective way to rebuild muscles, and it is also an effective solution for weight loss programs.

- High protein diets include meals enriched with foods like eggs, meats, peanuts, milk, chicken, sea foods, soy products, and fish.
- For vegetarians, proteins from vegetables like legumes and nuts are ideal.
- There are protein rich snacks that can be used to provide the necessary supply of protein. These include tasty tuna sandwich, sprout salads and soy products. Protein synthesis occurs on ribosomes.

Ques.2 Give the biological role amino acids.

Ans- Biological Role of Amino Acids

Amino acids play an important role in performing several biological and chemical functions in different parts of our body, including building, and repairing of tissues, the formation and function of enzymes, food digestion, the transportation of molecules etc.

Role of Essential Amino Acids

- Phenylalanine helps in maintaining a healthy nervous system and in boosting memory power.
- Valine acts as an important component in promoting muscle growth.
- Tryptophan is involved in the production of Vitamin B, and serotonin hormones.
- Serotonin plays a vital role in maintaining our appetite, regulating sleep, and boosting our moods.
- Leucine is involved in promoting protein synthesis and growth hormones.

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- Lysine is necessary for promoting the formation of antibodies, hormones, and enzymes.
- Histidine is involved in synthesizing red blood cells (RBCs) and white blood cells (WBCs).

Role of Non-Essential Amino Acids

- Alanine removes toxins from our body.
- Cysteine acts as antioxidant.
- Glutamine is necessary for the synthesis of DNA and RNA. Glycine acts as a neurotransmitter.
- Glutamic acid involved in the development and functioning of the human brain.
- Arginine helps in detoxification in the kidneys, healing wounds and maintaining immune system.
- Tyrosine plays a vital role in the production of the thyroid hormones.
- Serine helps in promoting muscle growth.
- Aspartic acid plays a major role in metabolism.
- Protein is mainly involved in the repairing of the tissues in the formation of collagen.

Ques.3 What are the qualitative test for proteins.

Ans- Qualitative Test for Proteins

1. Millon's reaction

The reaction is due to the presence of the hydroxyphenyl group, C_6H_5OH in the amino acid molecule; and any phenolic compound which is unsubstituted in the 3,5 positions such as tyrosine, phenol and thymol will give the reaction. Solutions of nitric acid containing mercuric nitrate reacts with phenols, producing red colors or yellow precipitates which react with nitric acid to form red solution.

Add 3 to 4 drops of Millon's reagent to 5 ml of test solution. Mix and bring the mixture gradually to a boiling point by heating over a small flame. Development of red color is due to the presence of protein. Excess of reagent should however be avoided since it may produce a yellow color which is not a positive reaction.

2. Xanthoproteic reaction

This reaction is due to the presence in the amino acid molecule of the phenyl group- C_6H_5, with which the nitric acid forms certain nitro modifications. The amino acids which are of special importance in this connection is those of tyrosine and tryptophan, Phenylalanine does not respond to this test as it is ordinarily preformed.

Nitration of the aromatic rings in Tyrosine and Tryptophan, with concentrated HNO_3, produce a yellow color.

Tyrosine or Tryptophan + con. HNO_3 → Yellow color

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Phenylalanine does not produce the color because the benzene ring is not activated for nitration.

Add 1 ml of conc. Nitric acid to 2 to 3 ml of test solution in a test tube. A white precipitate forms, which upon heating turns yellow and finally dissolves, imparting to the solution a yellow color, cool the solution and carefully add ammonium hydroxide or sodium hydroxide in excess. Note that the yellow color deepens into an orange.

3. Biuret test

It is a general test used for detecting the presence of proteins and peptides.

The Biuret test is given by those substances whose molecules contain two carbamoyl (-$CONH_2$) groups joined either directly or through a single atom of nitrogen or carbon. Similar substances which contain $CSNH_2$ -$C(NH)NH_2$, or -CH_2NH_2 in place of the $CONH_2$ group also respond to the test. It follows from this fact that substance which are non-protein in character but which contain the necessary groups will respond to the biuret test.

To 2 to 3 ml of test solution in a test tube add an equal volume of 10% sodium hydroxide solution, mix thoroughly, and add a 0.5% copper sulphate solution drop by drop, mixing between drops, until a purplish-violet or pinkish-violet color is produced.

4. Ninhydrin reaction

This test gives positive results with proteins, peptones, peptides, amino acids, and other primary amines, including ammonia.

- Amino acids (that have a-amino group) react with ninhydrin to form blue colored complex.
- This color is due to liberate NH_3 with ninhydrin.
- Ninhydrin is used to locate the a-amino acid in paper chromatography as a blue to purple spots.
- Also, permits the quantitative estimation of a-amino acid and peptides in column chromatography.

Proline gives yellow color due to lack of a-amino group. To 5 ml of dilute test solution, which must be approximately between pH 5 and pH 7 (a few drops of pyridine or a few crystals of sodium acetate may be used to adjust the pH), add 0.5 ml of 0.1 % ninhydrin, heat to boiling for one to two minutes, and allow to cool. A blue color develops if the test is positive.

Ques.4 Give the diseases related to malnutrition of proteins.

Ans- Diseases Related to Malnutrition of Proteins

- Proteins are substances that are part of cells, tissues, and organs throughout body, according to the centres for disease control.
- Protein deficiency is common among people who live in developing countries, those who live in impoverished communities in developed countries and in the elderly who lack access to nutritious food.

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- Protein deficiency also affects people who are born with a genetic disorder to produce certain proteins, and people with diseases that cause them to lose appetite and experience muscle breakdown.

1. Marasmus

Marasmus is a disease caused by a severe deficiency of protein and calories that affect infants and very young children, often resulting in weight loss and dehydration. Marasmus can develop into starvation and cause fatality caused by a lack of essential nutrients. People with marasmus appear bony with little muscle tissue.

2. Kwashiorkor

Lack of proteins from carbohydrates sources like rice, yams, and bananas causes kwashiorkor. This is a severe malnutrition disease common in older children. The University of Maryland Medical center explains that symptoms of the illness include a swollen stomach due to fluid retention. It also has symptoms common to marasmus such as irritability, diarrhoea, fatigue, limited growth, and cognitive development as well as mental health.

3. Protein Deficiency Symptoms

Lack of Protein has signs and symptoms depicted in the body changes. In case you experience any of the symptoms or you notice someone with the signs, it is advisable to seek immediate medical attention.

Some of these symptoms include:

- Weak and sore muscles Increased water retention
- Flakiness, dry skin, and rashes
- Lethargy
- Weight loss
- Anxiety
- Nausea
- Deep line formations around the toes and nails Stubborn wounds that do not heal
- Constant headaches
- Insomnia
- Moody feelings
- Blackouts
- Depression
- Skin ulcers

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- Bed sores
- Skin color changes

Ques.5 Give the biological functions of lipids.

Ans- Biological Function of Lipids

Storage of Energy

- Most lipids in biological systems function either as a source of stored metabolic energy or as structural matrices and permeability barriers in biological membranes.

Hormonal Roles

Very small amounts of special lipids act as both intracellular messengers and extracellular messengers such as hormones and pheromones.

- **Insulation:** Both thermal (triglycerides) and electrical (sphengolitrics).
- **Protection of internal organs** e.g., triglycerides and waxes.
- **Structural Components of cells** e.g., Phospholipids and cholesterol.

Types of lipids

- **Triglycerides:** Function as a long-term energy source in animals' fats and plants (Oils).
- **Phospholipids:** Structural component of cell membranes.
- **Steroids:** Act as hormones in plants and animals, and is a structural component of animal cell membranes (Cholesterol).
- **Waxes:** Act as a protective layer against water loss in plant leaves and animal skin
- **Carotenoids:** Light absorbing accessory pigment in plants (involved in Photo Glycolipids: Complexes of carbohydrate and lipid that function as cell receptor and cell recognition molecules.

Ques.6 Mention the specific test for cholesterol.

Ans- Specific Test for Cholesterol

1. Physical Test for Cholesterol: Cholesterol is a non-saponifiable lipid because it does not a contain any fatty acid, so if cannot produce soap. It contains basic steroidal nucleus, fat soluble and is found only in animals. It appears in the form of white crystals.

2. Salkowsk Reaction:

2 ml solution of cholesterol in chloroform

+

2 ml Conc. H_2SO_4 and mix carefully

↓

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The upper layer of chloroform becomes red and lower layer of

sulphuric acid changes to yellow with green fluorescence.

3. Liber man-Burchard's test

2 ml solution of cholesterol in chloroform
+
10 drops of acetic anhydride
+
+2 drops of H_2SO, mix carefully
↓

The rose red color is obtained which rapidly changes to blue and finally to bluish green.

4. Rosenhein Reaction

2 ml solution of cholesterol in chloroform
+
Trichloroacetic acid
+
Pink to Red color
↓

This reaction is also shown by Ergosterol and other sterols.

Ques.7 Define lipoproteins. Mention the types and functions of lipoproteins.

Ans- Lipoproteins

- Lipoproteins are special particles made up of droplets of fats surrounded by a single layer of Phospholipid molecules.
- Phospholipids are molecules of fats which are attached to a Phosphoros containing group. They have both polar and non-polar ends. (Amphipathic).

Types of Lipoproteins

1. Chylomicrons: These are the largest and least dense of the lipoproteins, with the highest triglyceride content. They consist of a protein component synthesized in the liver, which wraps around diet derived cholesterol and fats.

2. Very low-density lipoprotein (VLDL): This is composed of protein, fats and cholesterol synthesized in liver. It is associated with 5 different apoproteins. It is converted to IDL and LDL by the removal of apoproteins.

3. Intermediate density lipoprotein (IDL): It is created by the metabolism of VLDL.

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4. Low density lipoprotein (LDL): This is the last VLDL remnant and contains chiefly cholesterol.

5. High density lipoprotein (HDL): This has the highest protein; lipid ratio, and so is the densest. High HDL levels are associated with lowered risk of cardiovascular disease.

Functions of lipoproteins: Lipoproteins play essential roles in the body, specifically in:

(i) The absorption of and transport of lipids in the small intestine.
(ii) Transporting lipids from the liver to tissues.
(iii) Transferring lipids from tissues to the liver, also known as reverse cholesterol transport.
(iv) During reverse cholesterol transport, the body removes excess cholesterol from the tissues and brings it back to the liver. Then, the gallbladder, may remove it from the body, (or the body redistributes it)

Ques.8 Write a short note on role of lipids.

Ans- Role of Lipids

Fats have received a lot of bad publicity, and it's true that eating large amounts of fried foods and other "fatty foods can lead to weight gain and cause health problems. However, fats are essential to the body and have several important functions.

1. Waxes

Waxes are another biologically important category of lipids. Wax covers the feathers of some aquatic birds and the leaf surfaces of some plants, where its hydrophobic (water-repelling) properties prevent water from sticking to, or soaking into, the surface.

2. Phospholipids

Phospholipids are major components of the plasma membrane. Like fats, they are typically composed of fatty acid chains attached to a backbone of glycerol. Instead having three fatty acid tails, however, phospholipids generally have just two, and the third carbon of the glycerol backbone is occupied by a modified phosphate group. Different phospholipids have different modifiers on the phosphate group, with choline (a nitrogen-containing compound) and serine (an amino acid) being common examples.

3. Steroids

- Steroids are another class of lipid molecules, identifiable by their structure of four fused rings. Although they do not resemble the other lipids structurally, steroids are included in lipid category because they are also hydrophobic and insoluble in water.

- All steroids have four linked carbon rings and several of them, like cholesterol, also have a short tail.

- Many steroids also have an -OH functional group attached at a particular site, as shown for cholesterol below; such steroids are also classified as alcohols, and are thus called sterols.

Cholesterol, the most common steroid, is mainly synthesized in the liver and is the precursor to many steroid hormones.

- These include the sex hormones testosterone and estradiol, which are secreted by the gonads (testes and ovaries).

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- Cholesterol also serves as the starting material for other important molecules in the body, including vitamin D and bile acids, which aid in the digestion and absorption of fats from dietary sources.
- It is also a key component of cell membranes, altering their fluidity and dynamics.

Ques.9 Write down the characteristics of lipids.

Ans- Characteristics of Lipid are

- Lipids are relatively insoluble in water.
- They are soluble in non-polar solvents, like ether, chloroform, methanol. Lipids have high energy content and are metabolized to release calories.
- Lipids also act as electrical insulators; they insulate nerve axons.
- Fats contain saturated fatty acids; they are solid at room temperatures. Example, animal fats.
- Plant fats are unsaturated and are liquid at room temperatures,
- Pure fats are colourless, they have extremely bland taste.
- The fats are sparingly soluble in water and hence are described are hydrophobic substances.
- They are freely soluble in organic solvents like ether, acetone, and benzene. The melting point of fats depends on the length of the chain of the constituent fatty acid and the degree of unsaturation.
- Geometric isomerism, the presence of double bond in the unsaturated fatty acid of the lipid molecule produces geometric or cis-trans isomerism.
- Fats have insulating capacity; they are bad conductors of heat.
- Emulsification is the process by which a lipid mass is converted to several small lipid droplets. The process of emulsification happens before the fats can be absorbed by intestinal walls.
- The fats are hydrolysed by the enzyme lipases to yield fatty acids and glycerol.
- The hydrolysis of fats by alkali is called saponification. This reaction results in the formation of glycerol and salts of fatty acids called soaps.
- Hydrolytic rancidity is caused by the growth of microorganisms which secrete enzymes like lipases. These split fats into glycerol and free fatty acids.

Ques.10 Write a short note on polysaccharides.

Ans- Polysaccharides

- A long chain of monosaccharides linked by glycosidic bonds is known as a polysaccharide (poly- "many").

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- The chain may be branched or unbranched and may contain different types of monosaccharides.
- The molecular weight of a polysaccharide can be quite high, reaching to Daltons or more if enough monomers are joined.
- Starch, glycogen, cellulose, and chitin are some major examples of polysaccharides important in living organisms.
- Starch is the stored form of sugars in plants.
- Is made up of a mixture of two polysaccharides, amylose, and amylopectin (both polymers of glucose).
- Plants can synthesize glucose using light energy gathered in photosynthesis, and the excess glucose, beyond the plant's immediate energy needs, is stored as starch in different plant parts, including roots and seeds.

In starch, the glucose monomers are in the α form.

- **Amylose** consists entirely of unbranched chains of glucose monomers connected by a-glycosidic linkages between C-1 of one glucose. Unit and C-4 of the next glucose unit. The number of D-Glucose units in amylose ranges from 60-300.
- **Amylopectin:** Has a branched-chain structure. It is composed of chains of 25 to 30 D-glucose units. These chains are in turn connected to each other by 1,6 linkages. → The number of D-glucose units in amylopectin ranges from 300 to 6000.

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Very Short Answers

1. Which is a monosaccharide?
(a) Glucose
(b) Galactose
(c) Fructose
(d) All of these

2. Which is a disaccharide?
(a) Sucrose
(b) Lactose
(c) Maltose
(d) All of these

3. Glucose is:
(a) Disaccharide
(b) Monosaccharide
(c) Trisaccharide
(d) Polysaccharide

4. Glucose contains:
(a) One-CHO group
(b) 5-OH group
(c) 4 sec alcoholic group
(d) All of these

5. Sucrose is a:
(a) Disaccharide
(b) Monosaccharide
(c) Trisaccharide
(d) Polysaccharide

6. Common table sugar is:
(a) Glucose
(b) Sucrose
(c) Fructose
(d) Lactose

7. Which of the following sugar yields glucose only on Hydrolysis?
(a) Lactose
(b) Maltose
(c) Sucrose
(d) Fructose

8. The aldose and Ketose is differentiated by the following reagent:
(a) Br_2 water
(b) Fehling's Solution
(c) Tollen's reagent
(d) None of these

9. Which is a polysaccharide?
(a) Starch

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(b) Cellulose
(c) Glycogen
(d) All of these

10. Starch gives on hydrolysis:
(a) D-Galactose
(b) D-Glucose
(c) Fructose
(d) Lactose

11. The formula (CHO), is for:
(a) Starch
(b) Starch and cellulose
(c) Cellulose
(d) None of these

12. The sugar which is not a disaccharide in the following is:
(a) Lactose
(b) Galactose
(c) Sucrose
(d) Maltose

13. Glycogen is a:
(a) Monosaccharide
(b) Galactose
(c) Polysaccharide
(d) Maltose

14. Inulin (Homopolysaccharide) is used for clinical purposes in:
(a) Clearance tests
(b) Fehling test
(c) Blood test
(d) None of these

15. Essential components of milk is:
(a) Lactose
(b) Maltose
(c) Sucrose
(d) Fructose

16. α-amino acids may have
(a) One-NH_2 and one-COOH groups
(b) One-NH_2 and two-COOH groups
(c) Two-NH_2 and one-COOH groups
(d) All of these.

17. Proteins contain:
(a) Only α -amino acids
(b) Only β -amino acids
(c) Both α and β amino acids.
(d) α-β-γ amino acids

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18. Which of the following is an essential amino acid?
(a) Glycine
(b) Leucine
(c) Glutamic acid
(d) Valine

19. Proteins are:
(a) Polymers of ethylene
(b) Polyamides
(c) c-amino carboxylic acids
(d) Styrene polymer

20. The linear arrangement of amino acid units in proteins structure is called:
(a) Primary structure
(b) Tertiary
(c) Secondary structure
(d) Quaternary structure

21. The a-Helix is a common form of:
(a) Primary structure
(b) Secondary structure
(c) Tertiary structure
(d) None of these

22. The a-Helix structure is hold in a coiled conformation partially due to:
(a) H-bonding
(b) Optical activity
(c) Delocalisation
(d) α-bond

23. On hydrolysis, proteins give:
(a) Amino acids
(b) Alcohols
(c) Fatty acids
(d) Carboxylic acids

24. Amino Acids are linked together by a..... in protein molecules;
(a) Peptide bonds
(b) H-bonds
(c) Amide bonds
(d) None of these

25. One example of a non-essential amino acid is:
(a) Valine
(b) Histidine
(c) Glycine
(d) Arginine

26. Biuret reaction is specific for:
(a)-CONH bond
(b) -CSNH, group
(c)-$NH.NH_2$ group

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(d) All of these

27. Sakaguche's reaction is specific for:
(a) Tyrosine
(b) Protein
(c) Arginine
(d) Cysteine.

28. The protein present in Hair is:
(a) Keratin
(b) Elastin
(c) Myasin
(d) Tropocollagen

29. The most abundant protein in manuals is:
(a) Albumin
(b) Haemoglobin
(c) Collagen
(d) Elastin

30. In Nitroprusside test, amino acid cysteine produces a:
(a) Red color
(b) Blue color
(c) Yellow color
(d) Purple color

31. Protein present in haemoglobin has the structure known as:
(a) Primary
(b) Secondary
(c) Tertiary
(d) Quaternary

32. Fats are abundantly found in:
(a) Vegetative tissue
(b) Reproductive tissue
(c) Both (a) & (b)
(d) None of these

33. Natural lipids are easily soluble in:
(a) Oil
(b) Mercury
(c) Water
(d) None of these

34. Identify unsaturated fatty acids from the following:
(a) Linoleic acid
(b) Oleic acid
(c) Palmitoleic acid
(d) All of these

35. Liquid form of triglycerides at ordinary room temperature are called:
(a) Oils

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(b) Solid
(c) Fats
(d) None of these

36. Hydrolysis of fats by alkalies into fatty acids and glycerol is called:
(a) Coagulation
(b) Saponification
(c) Suspension
(d) Colloidal

37. The fats and oils are respectively rich in:
(a) Unsaturated fatty acids
(b) Saturated fatty acids
(c) Saturated and unsaturated fatty acids
(d) None of these

38. Which is a phospholipid?
(a) Lecithin
(b) Cholesterol
(c) Sterol
(d) Steroid

39. Essential fatty acids are:
(a) linoleic acid
(b) arachidonic acid
(c) linolenic acid
(d) Steroid

40. Examples of monosaturated fatty acids are:
(a) Oleic acid
(b) Arachi donic acid
(c) Palmitic acid
(d) Linolenic acid

41. High content of triglycerides are present in:
(a) LDL
(b) HDL
(c) VLDL
(d) Chylomicrons

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