KARPAGAM ACADEMY OF HIGHER EDUCATION

(Deemed to be University)
(Established Under Section 3 of UGC Act 1956)
Coimbatore – 641 021.
(For the candidates admitted from 2017onwards)

DEPARTMENT OF BIOCHEMISTRY

Subject	:	**BIOCHEMISTRY**	**Semester**	:	**I**
Subject code	:	**18MBU103**	**Class**	:	**B.Sc Microbiology**

Course objectives

- To provide the basics of biochemistry and its applications.
- To learn structures and functions of enzymes, proteins, carbohydrates, fats, process of metabolism and the molecular basis of the action of genes also form a part of biochemistry.
- It serves as a good research techniques and the ability to combine and analyze information.

Course

Unit I

Atoms and molecules, cell structure, cell organelles, developing membrane structure, transport of molecules, Beer and Lambert's Law, Colorimeter, Anabolism and catabolism and standard for energy change.

Unit II

Monosaccharides-families, stereo isomerism, epimers, Mutarotation and anomers. Forms of glucose and fructose, Fischer and Haworth projection. Sugar derivatives. Disaccharides- occurrence, concept of reducing and non-reducing sugars and Haworth projections. Polysaccharides-storage and structural polysaccharides.

Unit III

Classification and functions of lipids. Storage lipids- structure and function of fatty acids. Triacylglycerols. Saponification. Structural lipids- structure, functions and properties of phosphoglycerides and sphingolipids.

Unit IV

Classification and functions of proteins and amino acids, Structure of amino acids and concept of zwitterion. Ninhydrin reaction. Natural modifications of amino acids in proteins. Non protein amino acids, Oligopeptides: Structure and functions of glutathione, insulin and aspartame. Primary and Secondary structure of proteins- alpha helix, beta pleated sheet. Tertiary and quaternary structures of proteins. Human haemoglobin structure.

Structure and classification of enzymes, mechanism of action of enzymes. Km equation and enzyme activity. Allosteric enzyme and its mechanism. Multienzyme complex. Enzyme inhibition. Vitamins-classification and characteristics, sources and importance.

Unit V

Nucleic Acids-Purines & Pyrimidinesnucleotides, RNA, & DNA base pairing schemes, types of RNA: mRNA, rRNA, tRNA, aminoacyl tRNA synthetase, Secondary structure of DNA, Watson and Crick model. Denaturation of DNA keto-enol tautomerism and consequences.

SUGGESTED READINGS

1. Campbell, M.K. (2012) Biochemistry, 7th edition. Published by Cengage Learning.
2. Campbell, P.N., and Smith, A.D., (2011) Biochemistry Illustrated, 4th edition. Published by Churchill Livingstone.
3. Tymoczko, J.L., Berg, J.M., and Stryer, L. (2012) Biochemistry: A short course, 2nd edition. W.H.Freeman.
4. Berg, J.M., Tymoczko, J.L., and Stryer, L. (2011) Biochemistry, W.H.Freeman and Company. Nelson, D.L and Cox, M.M. (2008) Lehninger Principles of Biochemistry, 5th edition. W.H. Freeman and Company.
5. Willey, M.J., Sherwood, L.M., & Woolverton, C. J. (2013) Prescott, Harley and Klein's Microbiology. 9th edition. McGrawHill.

KARPAGAM ACADEMY OF HIGHER EDUCATION

(Deemed to be University)
(Established Under Section 3 of UGC Act 1956)
Coimbatore – 641 021.
(For the candidates admitted from 2017onwards)

DEPARTMENT OF BIOCHEMISTRY

Subject	:	**BIOCHEMISTRY**	**Semester**	:	**I**
Subject code	:	**18MBU103**	**Class**	:	**B.Sc Biochemistry**

LECTURE PLAN
DEPARTMENT OF BIOCHEMISTRY

Sl. No	LECTURE DURATION	TOPICS	BOOK REFERENCE	PAGE NO
		UNIT-I		
1	1	Atoms and Molecules	T1	1
2	2	Cell Structure, Cell organelles	T1	2-7
3	1	Developing membrane structure	T1	2-7
4	1	Transport of molecules	T2	2
5	1/2	Beer and Lambert's law	T1	76
6	1/2	Colorimeter	W1	
7	1	Anabolism and catabolism	T2	27
8	1	Standard free energy change	T2	23-26
9	1	Revision and possible question discussion		
Total: 9 hours				
		UNIT-II		
1	1	Families of Monosaccharide	T1	235-236
2	2	Stereoisomerism of Monosaccharide, epimers, Mutarotation and anomers	T1	237-238
3	1	Forms of glucose and fructose and Haworth projection formulae	T1	239
4	1	Sugar derivatives	T1	240
5	1	Disaccharides: concept of reducing and non-reducing sugars, Haworth projections	T1	239 ,243-249

6	1	Polysaccharides; storage polysaccharides, starch and glycogen	T1	244-247
7	1	Structural polysaccharides cellulose and chitin	T1	247-252
8	1	Revision and Possible QP discussion		
Total: 9 hours				
		UNIT-III		
1	1/2	Classification of lipids	T1	357-363
2	1/2	Functions of Lipids	T1	357-363
3	1	Storage lipids; structure of fatty acids	T1	343-346
4	1	Functions of storage lipids	T1	343-346
5	1	Triacylglycerols & Saponification	T1	346
6	2	Structural lipids-structure, functions and properties of phosphoglycerides	T1	350-352
7	1	Structure, functions and properties of sphingolipids	T1	349, 354
8	1	Revision and Possible QP discussion		
Total: 8 hours				
		UNIT-IV		
1	1	Classification and functions of proteins and amino acids	T1	74-76
2	1	Structure of amino acids and concept of zwitterions, Ninhydrin reaction	T1	72-79
3	1	Natural modifications of amino acids in proteins	T1	114-116
4	1/2	Non protein amino acids	T1	114-116
5	1	Oligopeptides: Structure and functions of glutathione, insulin and aspartame	T1	876-878, 439, 559
6	1/2	Primary and secondary structure of proteins-alpha helix, beta pleated sheet	T1	113-123
7	1/2	Tertiary and quaternary structures of proteins	T1	113-123
8	1	Human haemoglobin structure	T1	154-158
9	1/2	Structure and classification of enzyme	T1	183-185
10	1/2	Mechanism of action of enzymes	T1	186-194
11	1/2	Km equation and enzyme activity	T1	197-201
12	1	Allosteric enzyme and its mechanism	T1	220-223

13	1/2	Multi-enzyme complex	T1	223-225
14	1/2	Enzyme inhibition	T1	226-236
15	1	Vitamins-classification and characteristics-sources and importance	W2	
16	1	Revision and Possible QP discussion		
Total: 12 hours				
UNIT-V				
1	1	Nucleic acids-Purines & Pyrimidine nucleotides	T1	273-278
2	1	RNA & DNA base pairing schemes	R1	
3	2	Types of RNA: mRNA, rRNA, tRNA, aminoacyl tRNA synthetase	T1 T3	1008-1020 97-100
4	2	Secondary structure f DNA, Watson and Crick model	T1 T3	279-280 95-96
5	1	Denaturation of DNA keto-enol tautomerism and consequences	W3	
6	1	Revision and Possible QP discussion		
Total: 8 hours				
PREVIOUS YEAR END SEMESTER EXAMINATION QUESTION PAPER DISCUSSION				
1	1	Previous year ESE question paper discussion		
2	1	Previous year ESE question paper discussion		
Total: 2 hours				
Grand Total: 48 hours				

REFERENCES

T1	David L Nelson and Michael M. Cox (2008). Lehninger Principles of Biochemistry (5th edition), Freeman and Company.
T2	Berg JM, Tymoczko JL and Stryer L (2011). Biochemistry, W.H. Freeman and Company.
T3	Debu (2011). Fundamentals of Biochemistry (9th edition) New Central Book Agency, Calcutta
W1	https://www.azosensors.com/article.aspx?ArticleID=324
W2	https://caloriebee.com/nutrition/Vitamins-Their-functions-deficiency-symptoms-and-natural-sources
R1	Ananth P, Goldsmith G, Yathindra N (2013). An innate twist between crick's wobble and Watson-crick base pairs. RNA. 19:1038-1053.
W3	https://en.wikipedia.org/wiki/Keto%E2%80%93enol_tautomerism

UNIT-I

SYLLABUS

Atoms and molecules, cell structure, cell organelles, developing membrane, structure, transport of molecules, Beer and Lambert's Law, Colorimeter, Anabolism and catabolism and standard for energy change.

Atoms and Molecules

An atom is the smallest unit of matter that has the properties of an element. It is composed of a dense core called the nucleus and a series of outer shells occupied by orbiting electrons. Elements are substances consisting of one type of atom, for example Carbon atoms make up diamond, and also graphite. Pure (24K) gold is composed of only one type of atom, gold atoms.

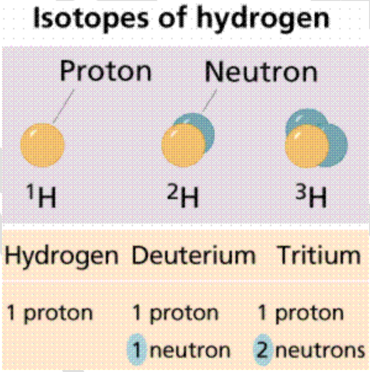
Subatomic particles were discovered during the 1800s. There are three main subatomic particles viz., Proton, Neutron and Electron.

- The **proton** is located in the center (or nucleus) of an atom, each atom has at least one proton. Protons have a charge of +1, and a mass of approximately 1 atomic mass unit (amu). Elements differ from each other in the number of protons they have, e.g. Hydrogen has 1 proton; Helium has 2.
- The **neutron** also is located in the atomic nucleus (except in Hydrogen). The neutron has no charge, and a mass of slightly over 1 amu.
- The **electron** is a very small particle located outside the nucleus. Because they move at speeds near the speed of light the precise location of electrons is hard to pin down. Electrons occupy orbital's, or areas where they have a high statistical probability of occurring. The charge on an electron is -1. Its mass is negligible (approximately 1800 electrons are needed to equal the mass of one proton).

Name	Charge	Location	Mass
Proton	+1	atomic nucleus	1.6726 X 10^{-27} kg
Neutron	0	atomic nucleus	1.6750 X 10^{-27} kg
Electron	-1	electron orbital	9.1095 X 10^{-31} kg

The **atomic number** is the number of protons an atom has. It is characteristic and unique for each element. The **atomic mass** (also referred to as the atomic weight) is the number of protons and neutrons in an atom. Atoms of an element that have differing numbers of neutrons (but a constant atomic number) are termed isotopes.

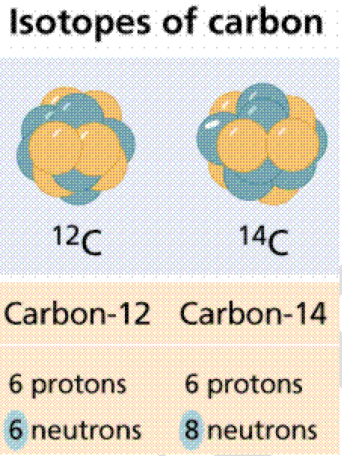
Example 1: Isotopes of Hydrogen



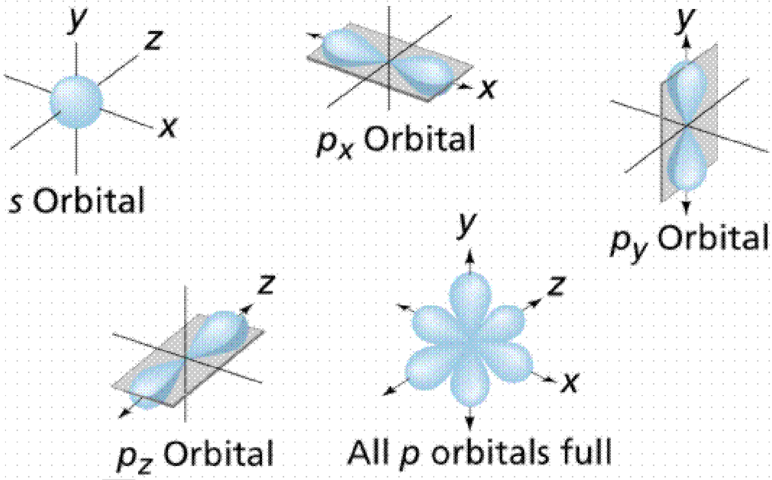
Note that each of these isotopes of hydrogen has only one proton. Isotopes differ from each other in the number of neutrons, not in the number of protons.

Some isotopes are radioisotopes, which spontaneously decay, releasing radioactivity. Other isotopes are stable. Examples of radioisotopes are Carbon-14 (symbol ^{14}C), and deuterium (also known as Hydrogen-2; ^2H). Stable isotopes are ^{12}C and ^1H.

Example 2: Isotopes of carbon



An orbital is also an area of space in which an electron will be found 90% of the time. Orbitals have a variety of shapes. Each orbital has a characteristic energy state and a characteristic shape. The *s* orbital is spherical. Since each orbital can hold a maximum of two electrons, atomic numbers above 2 must fill the other orbitals. The p_x, p_y, and p_z orbitals are dumbbell shaped, along the x, y, and z axes respectively.



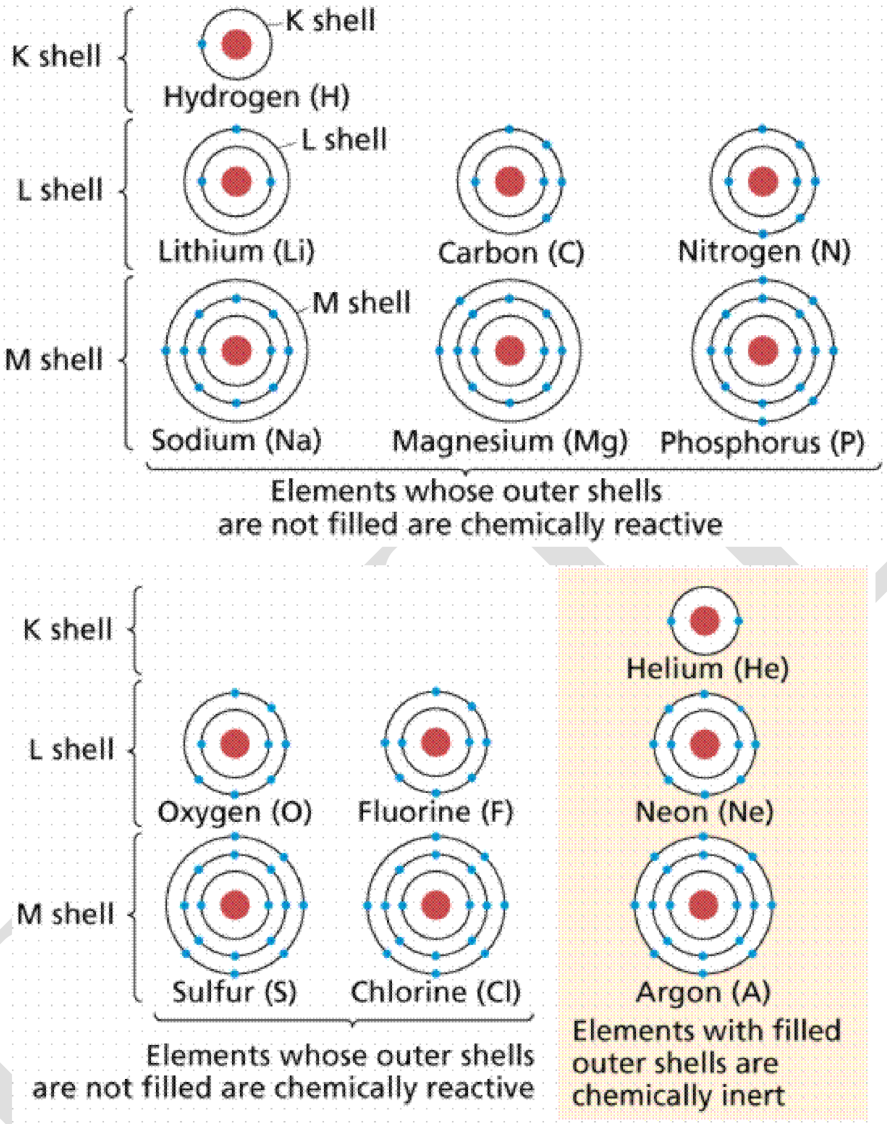
Energy levels (also referred to as electron shells) are located a certain "distance" from the nucleus. The major energy levels into which electrons fit, are (from the nucleus outward) K, L, M, and N. Sometimes these are numbered, with electron configurations being: $1s^2 2s^2 2p^1$, (where the first shell K is indicated with the number 1, the second shell L with the number 2, etc.). This nomenclature tells us that for the atom mentioned in this paragraph, the first energy level (shell) has two electrons in its *s* orbital (the only orbital it can have), and second energy level has a maximum of two electrons in its *s* orbital, plus one electron in its *p* orbital.

As a general rule, for the atoms we are likely to encounter in biological systems, atoms tend to gain or lose their outer electrons to achieve a Noble Gas outer electron shell configuration of two or eight electrons. The number of electrons that are gained or lost is characteristic for each element, and ultimately determines the number and types of chemical bonds atoms of that element can form.

Chemical Bonding

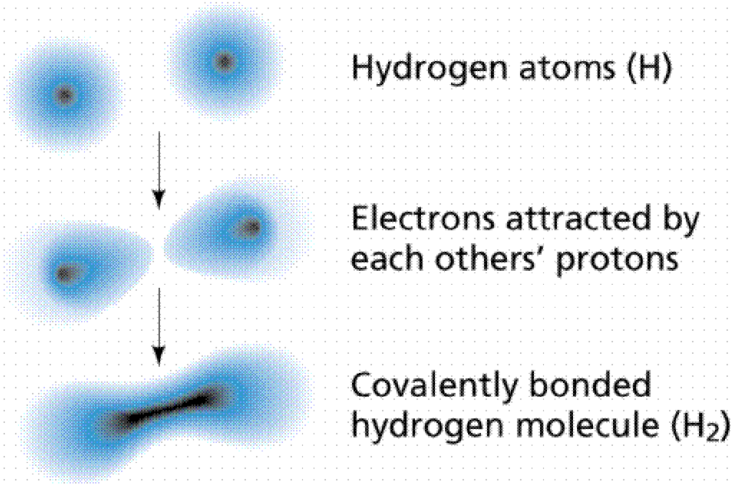
Ionic bonds

Ionic bonds are formed when atoms become ions by gaining or losing electrons. Chlorine is in a group of elements having seven electrons in their outer shells. Members of this group tend to gain one electron, acquiring a charge of -1. Sodium is in another group with elements having one electron in their outer shells. Members of this group tend to lose that outer electron, acquiring a charge of +1. Oppositely charged ions are attracted to each other, thus Cl^- (the symbolic representation of the chloride ion) and Na^+ (the symbol for the sodium ion, using the Greek word *natrium*) form an ionic bond, becoming the molecule sodium chloride. Ionic bonds generally form between elements in Group I (having one electron in their outer shell) and Group VII a (having seven electrons in their outer shell). Such bonds are relatively weak, and tend to disassociate in water, producing solutions that have both Na and Cl ions.

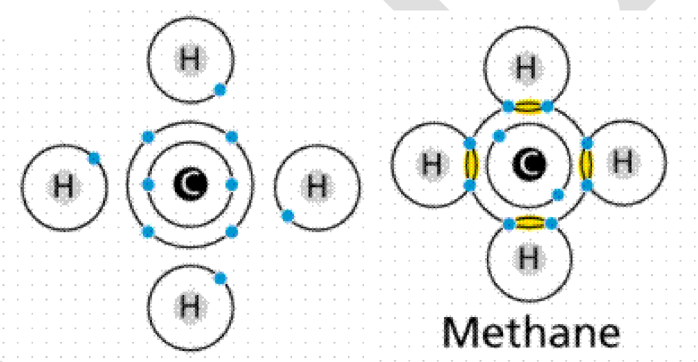


Covalent bonds

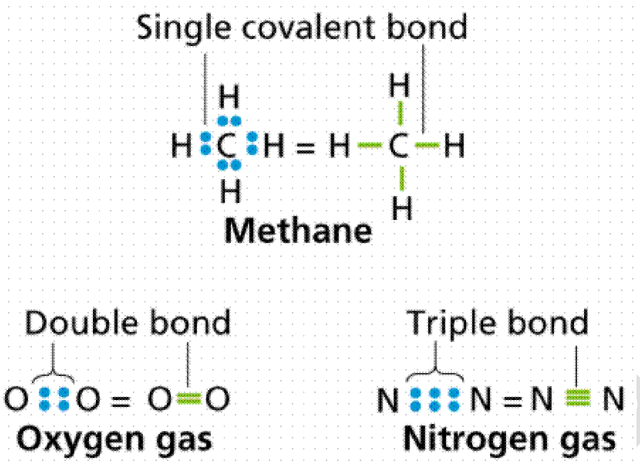
Covalent bonds form when atoms share electrons. Since electrons move very fast they can be shared, effectively filling or emptying the outer shells of the atoms involved in the bond. Such bonds are referred to as electron-sharing bonds. An analogy can be made to child custody: the children are like electrons, and tend to spend some time with one parent and the rest of their time with the other parent. In a covalent bond, the electron clouds surrounding the atomic nuclei overlap.



Carbon (C) is in Group IV a, meaning it has four electrons in its outer shell. Thus to become a "happy atom", Carbon can either gain or lose four electrons. By sharing the electrons with other atoms, Carbon can become a happy atom, alternately filling and emptying its outer shell, as with the four hydrogen's.



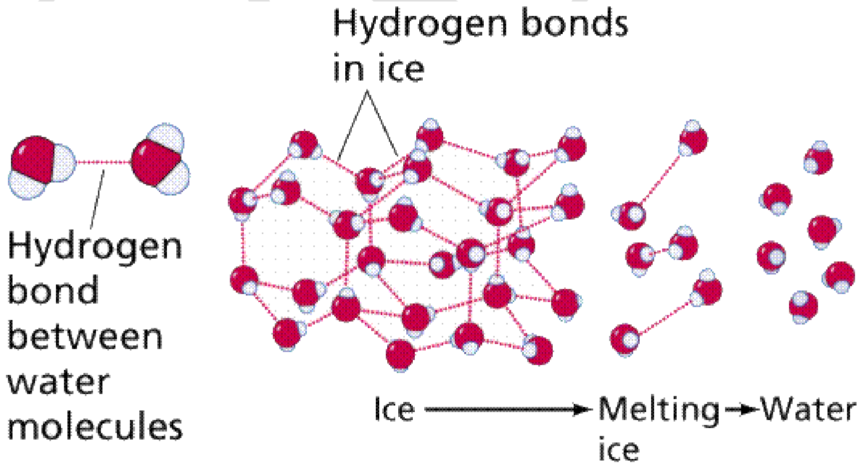
The molecule methane (chemical formula CH_4) has four covalent bonds, one between Carbon and each of the four Hydrogens. Carbon contributes an electron, and Hydrogen contributes an electron. The sharing of a single electron pair is termed a single bond. When two pairs of electrons are shared, a double bond results, as in carbon dioxide. Triple bonds are known, wherein three pairs (six electrons total) are shared as in acetylene gas or nitrogen gas. The types of covalent bonds are:



Hydrogen bonds

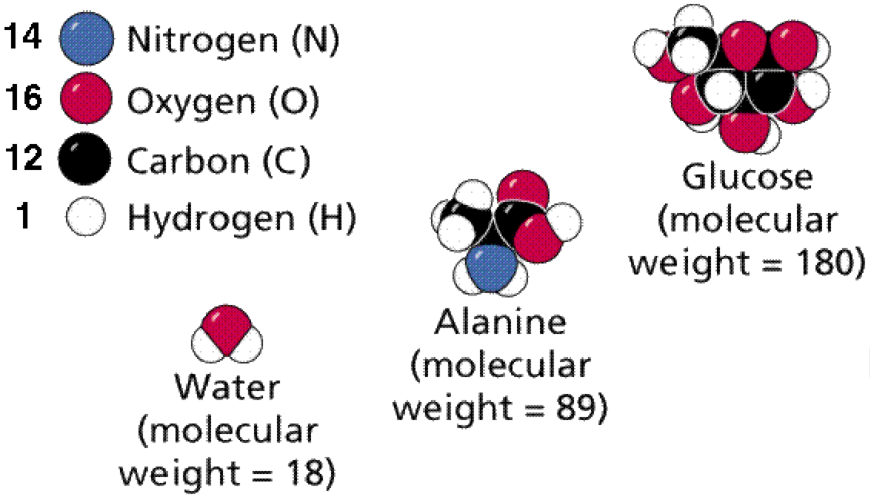
Hydrogen bonds result from the weak electrical attraction between the positive end of one molecule and the negative end of another. Individually these bonds are very weak, although taken in a large enough quantity, the result is strong enough to hold molecules together or in a three-dimensional shape.

Example: Formation of a hydrogen bond between the hydrogen side of one water molecule and the oxygen side of another water molecule.



Molecules

Molecules are compounds in which the elements are in definite, fixed ratios. Those atoms are held together usually by one of the three types of chemical bonds discussed above. For example: water, glucose, ATP. Mixtures are compounds with variable formulas/ratios of their components. For example: soil. Molecular formulas are an expression in the simplest whole-number terms of the composition of a substance. For example, the sugar glucose has 6 Carbons, 12 hydrogens, and 6 oxygens per repeating structural unit. The formula is written $C_6H_{12}O_6$.



Chemical reactions occur in nature, and some also can be performed in a laboratory setting. One chemical equation are linear representations of how these reactions occur. Combination reactions occur when two separate reactants are bonded together, e.g. A + B -----> AB. Disassociation reactions occur when a compound is broken into two products, e.g. AB -----> A + B.

The cell and cell theory

Soon after Anton van Leeuwenhoek invented the microscope, Robert Hooke in 1665 observed a piece of cork under the microscope and found it to be made of small compartments which he called “cells” (Latin cell = small room). In 1672, Leeuwenhoek observed bacteria, sperm and red blood corpuscles, all of which were cells. In 1831, Robert Brown, an Englishman observed that all cells had a centrally positioned body which he termed the nucleus.

The cell theory

In 1838 Matthias Schleiden and Theodor Schwann formulated the “Cell Theory.” The cell theory maintains that

- All organisms are composed of cells.
- Cell is the structural and functional unit of life, and
- Cells arise from pre-existing cells.

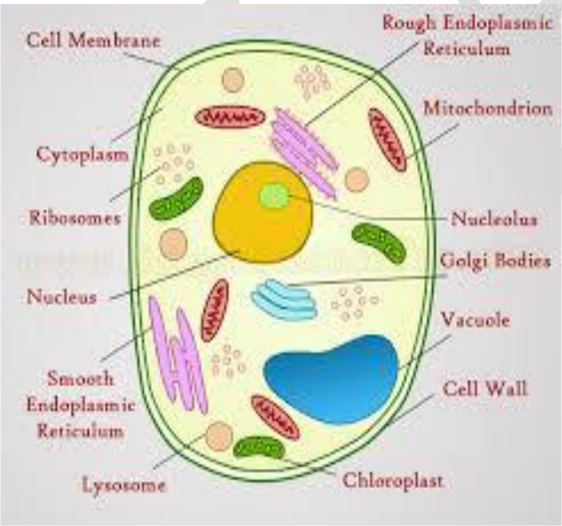
The cells vary considerably, in shape and size

Cell Structure, cell organelles and developing membrane structure

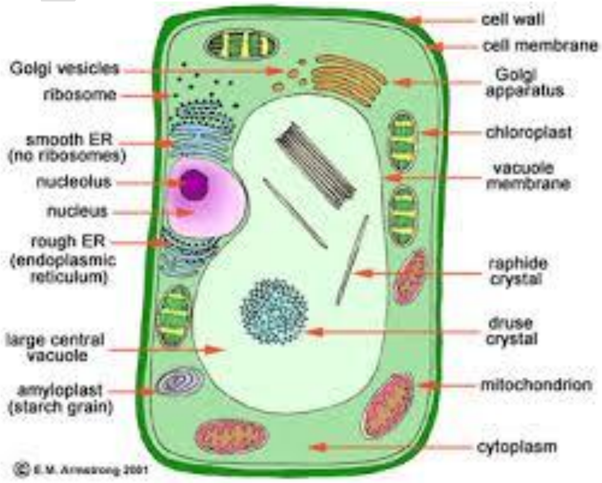
A cell may be defined as a unit of protoplasm bounded by a plasma or cell membrane and possessing a nucleus. Protoplasm is the life giving substance and includes the cytoplasm and the nucleus. The cytoplasm has in it organelles such as ribosomes, mitochondria, Golgi bodies plastids, lysosomes and endoplasmic reticulum. Plant cells have in their cytoplasm large vacuoles containing non-living inclusions like crystals, pigments etc. The bacteria have neither organelles nor a well formed nucleus. But every cell has three major components

- plasma membrane
- cytoplasm
- DNA (naked in bacteria and covered by a membrane in all other organisms

Animal Cell



Plant Cell



Plasma membrane

Each cell has a limiting boundary, the cell membrane, plasma membrane or plasmalemma. It is a living membrane, outermost in animal cells but next to cell wall in plant cells.

The plasma membrane is made of proteins and lipids and several models were proposed regarding the arrangement of proteins and lipids. The fluid mosaic model proposed by Singer and Nicholson (1972) is widely accepted.

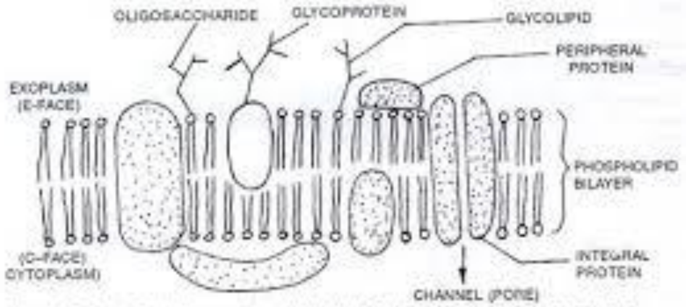


Figure 3.7 A simplified diagrammatic sketch of the Fluid-Mosaic Model of biomembrane (after Singer and Nicolson 1972).

According to the fluid mosaic model,

(i) The plasma membrane is composed of a lipid bilayer of phospholipid molecules into which a variety of globular proteins are embedded.

(ii) Each phospholipid molecule has two ends, an outer head hydrophilic i.e. water attracting, and the inner tail pointing centrally hydrophobic, i.e. water repelling

(iii) The protein molecules are arranged in two different ways:

   a. Peripheral proteins or extrinsic proteins: these proteins are present on the outer and inner surfaces of lipid bilayer.

   b. Integral proteins or intrinsic proteins: These proteins penetrate lipid bilayer partially or wholly.

Functions

(i) The plasma membrane encloses the cell contents.

(ii) It provides cell shape (in animal cells) e.g. the characteristic shape of red blood cells, nerve cells, bone cells, etc

(iii) It allows transport of certain substances into and out of the cell but not all substance, so it is termed selectively permeable.

Cell wall

In bacteria and plant cells the outermost cell cover, present outside the plasma membrane is the cell wall. Bacterial cell wall is made of peptidoglycan.

Structure

- Outermost non-living, layer present in all plant cells.
- Secreted by the cell itself.
- In plant, made of cellulose but may also contain other chemical substance such as pectin and lignin.
- The substance constituting the cell is not simply homogenous but it consists of fine threads or fibres called microfibrils.
- It may be thin (1 micron) and transparent as in the cells of onion peel. In some cases it is very thick as in the cells of wood.

Functions

- The cell wall protects the delicate inner parts of the cell.
- Being rigid, it gives shape to the cell.
- Being rigid, it does not allow distension of the cell, thus leading to turgidity of the cell that is useful in many ways.
- It freely allows the passage of water and other chemicals into and out of the cells.
- There are breaks in the primary wall of the adjacent cells through which cytoplasm of one cell remains connected with the other. These cytoplasmic strands which connect one cell to the other one are known as plasmodesmata.
- Walls of two adjacent cells are firmly joined by a cementing material called middle lamella made of calcium pectate.

Mitochondria and chloroplast - the energy transformers

Mitochondria (found in plant and animal cells) are the energy releasers and the chloroplasts (found only in green plant cells) are the energy trappers.

Mitochondria (Singular = mitochondrion)

Appear as tiny thread like structure under light microscope. Approximately 0.5 - 1.00 µm (micrometer) Number usually a few hundred to a few thousand per cell (smallest number is just one as in an alga (Micromonas).

Structure

The general plan of the internal structure of a mitochondria observed by means of electron microscope. It consists of the following parts:

- Wall made of double membrane
- The inner membrane is folded inside to form projections called cristae which project into the inner compartment called matrix.

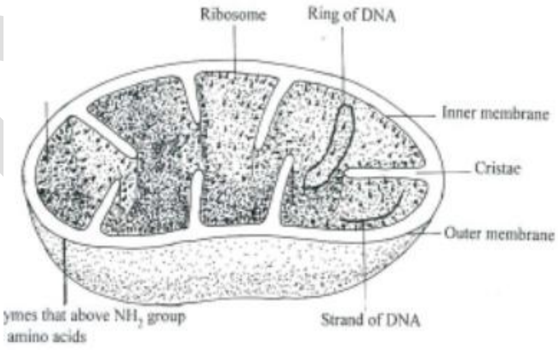
Function

Oxidizes pyruvic acid (breakdown product of glucose) to release energy which gets stored in the form of ATP for ready use. This process is also called cellular respiration.

Plastids

Plastids are found only in a plant cell. They may be colorless or with color. Based on this fact, there are three types of plastids.

(i) Leucoplast-white or colorless
(ii) Chromoplast – blue, red, yellow etc.
(iii) Chloroplast – green



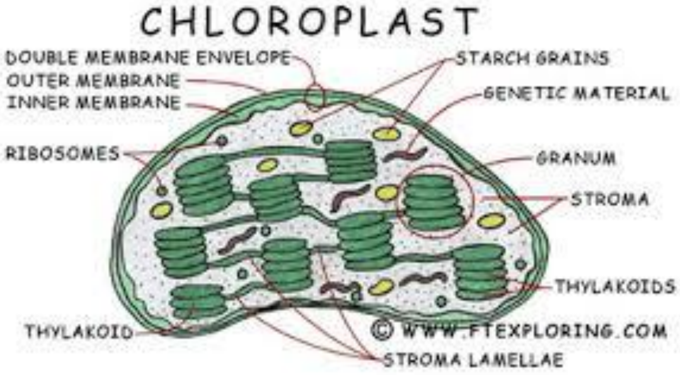
Chloroplast

- Found in all green plant cells in the cytoplasm.

- Number 1 to 1008
- Shape: Usually disc-shaped or spherical as in most plants around you. In some ribbon - shaped as in an alga spirogyra or cup - shaped as in other algae *Chlamydomonas*.
- Structure: the general plan of the structure of a single chloroplast is given below:
    - Wall made of double membrane i.e. outer membrane and inner membrane numerous stack-like (piles) groups or grana (singular = granum) are interconnected by lamellae.
    - Sac like structures called thylakoids placed one above the other constitute granum.
    - Inside of the chloroplast is filled with a fluid medium called stoma.

Function

Chloroplasts are the seat of photosynthesis (production of sugar, from carbon dioxide and water in the presence of sunlight).



Chloroplast versus mitochondria

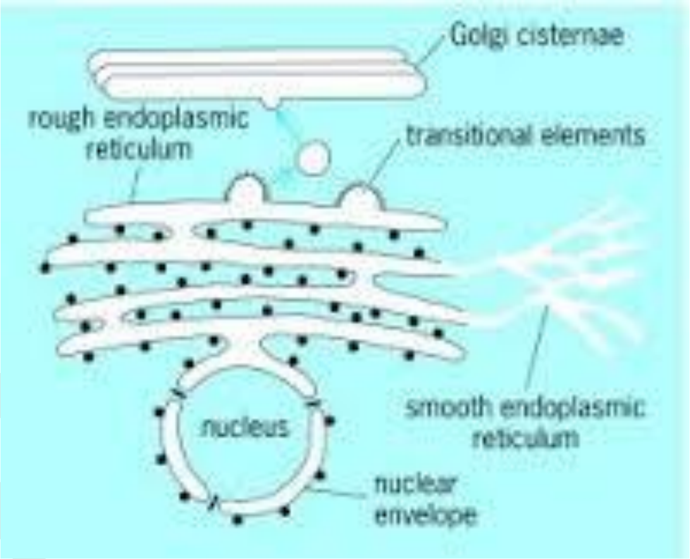
These two organelles are opposite to each other, one traps the solar energy locking it in a complex molecule (by photosynthesis), the other releases the energy by breaking the complex molecule (by respiration).

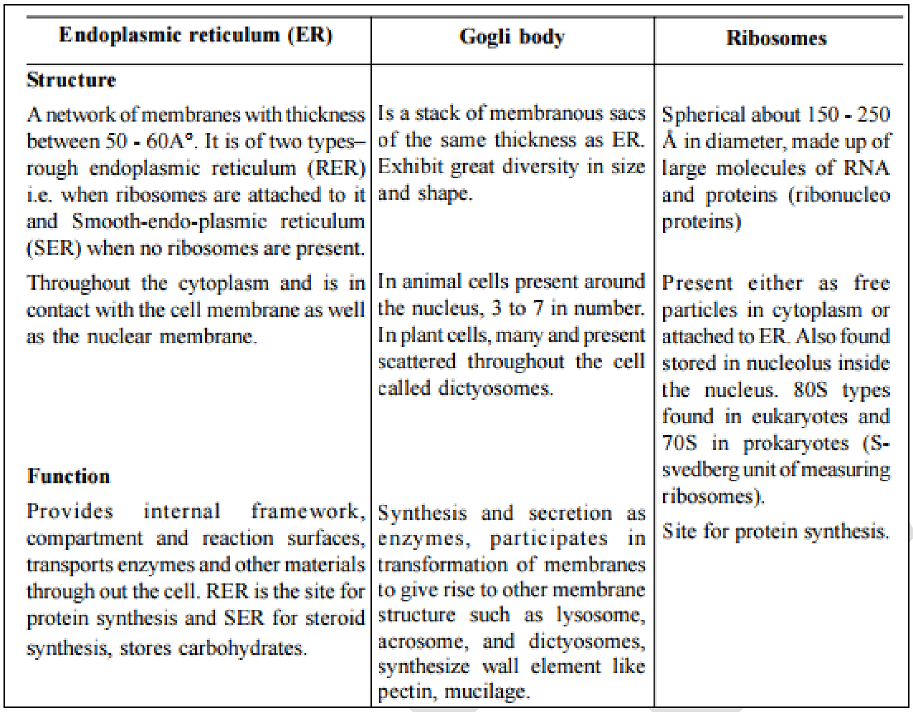
Similarities between mitochondria and chloroplasts

Both contain their own DNA (the genetic material) as well as their own RNA (for protein synthesis). Thus, they can self duplicate to produce more of their own kind without the help of nucleus. Since chloroplasts and mitochondria contain their own DNA the hereditary molecule and also their own ribosomes, they are termed semi-autonomous only because they are incapable of independent existence though they have ribosomes and DNA.

Endoplasmic reticulum (ER), Golgi body and ribosomes

Endoplasmic reticulum (ER) and Golgi body are single membrane bound structures. The membrane has the same structure (lipid-protein) as the plasma membrane but ribosomes do not have membranes Ribosomes are involved in synthesis of substances in the cell, Golgi bodies in secreting and the ER in transporting and storing the products. These three organelles operate together. The figure below shows the diagram of ER and Golgi body. Note the ribosomes present in ER.





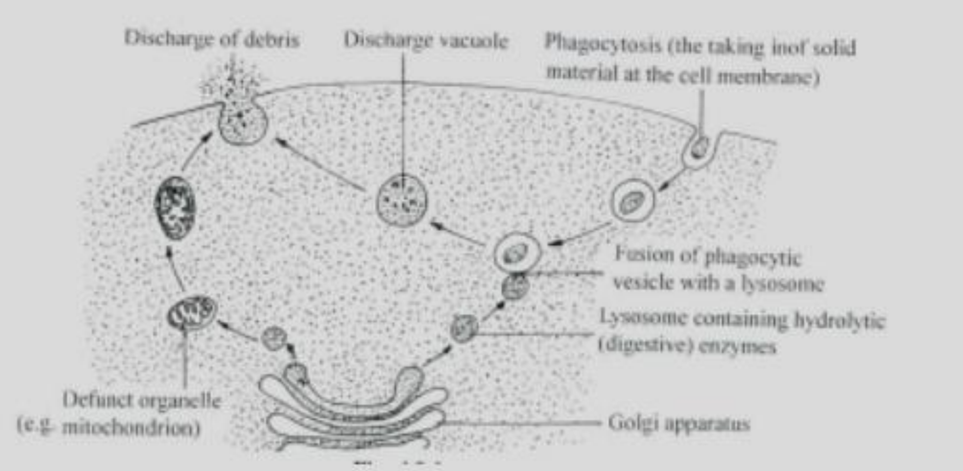
Endoplasmic reticulum (ER)	Gogli body	Ribosomes
Structure		
A network of membranes with thickness between 50 - 60A°. It is of two types– rough endoplasmic reticulum (RER) i.e. when ribosomes are attached to it and Smooth-endo-plasmic reticulum (SER) when no ribosomes are present.	Is a stack of membranous sacs of the same thickness as ER. Exhibit great diversity in size and shape.	Spherical about 150 - 250 Å in diameter, made up of large molecules of RNA and proteins (ribonucleo proteins)
Throughout the cytoplasm and is in contact with the cell membrane as well as the nuclear membrane.	In animal cells present around the nucleus, 3 to 7 in number. In plant cells, many and present scattered throughout the cell called dictyosomes.	Present either as free particles in cytoplasm or attached to ER. Also found stored in nucleolus inside the nucleus. 80S types found in eukaryotes and 70S in prokaryotes (S-svedberg unit of measuring ribosomes).
Function		
Provides internal framework, compartment and reaction surfaces, transports enzymes and other materials through out the cell. RER is the site for protein synthesis and SER for steroid synthesis, stores carbohydrates.	Synthesis and secretion as enzymes, participates in transformation of membranes to give rise to other membrane structure such as lysosome, acrosome, and dictyosomes, synthesize wall element like pectin, mucilage.	Site for protein synthesis.

The micro bodies (tiny but important)

These are small sac-like structures bounded by their membranes. These are of different kinds and the most important ones are lysosomes, peroxisomes and glyoxysomes.

Lysosomes (lysis = breaking down; soma = body)

Lysosomes are present in almost all animal cells and some non - green plant cells (Fig 4.9). They perform intracellular digestion.



Some main features of lysosomes are as follows

- Membranous sacs budded off from Golgi body.
- May be in hundreds in single cell.
- Contain several enzymes (about 40 in number)
- Materials to be acted upon by enzymes enter the lysosomes.
- Lysosomes are called "suicidal bags" as enzymes contained in them can digest the cell's own material when damaged or dead.
- Importance of intracellular digestion by the lysosomes
- Help in nutrition of the cell by digesting food, as they are rich in various enzymes which enable them to digest almost all major chemical constituents of the living cell.
- Help in defense by digesting germs, as in white blood cells.
- Help in cleaning up the cell by digesting damaged material of the cell.
- Provide energy during cell starvation by digestion of the cells own parts (autophagic, auto: self; phagos: eat up).
- Help sperm cells in entering the egg by breaking through (digesting) the egg membrane.
- In plant cells, mature xylem cells lose all cellular contents by lysosome activity.
- When cells are old, diseased or injured, lysosomes attack their cell organelles and digest them. In other words lysosomes are autophagic, i.e. self devouring.

Peroxisomes

Found both in plant and animal cells. Found in the green leaves of higher plants. They participate in oxidation of substrates resulting in the formation of hydrogen peroxide.

- They often contain a central core of crystalline material called nucleoid composed of urate oxidase crystals.
- These bodies are mostly spherical or ovoid and about the size of mitochondria and lysosomes. They are usually closely associated with E.R.
- They are involved in with photorespiration in plant cells.

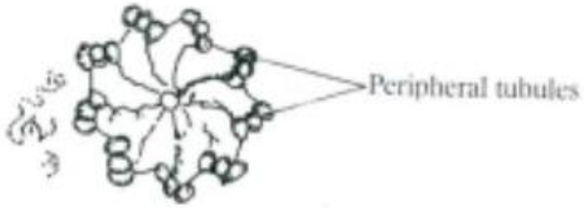
- They bring about fat metabolism in cells.

Glyoxysomes

- The micro bodies present in plant cells and morphologically similar to peroxisomes.
- Found in the cell of yeast and certain fungi and oil rich seeds in plants.
- Functionally they contain enzyme of fatty acid metabolism involved in the conversion of lipids to carbohydrates during germination.

Centriole

It is present in all animal cells, located just outside the nucleus. It is cylindrical, 0.5 µm in length and without a membrane. It has 9 sets of peripheral tubules but none in the centre. Each set has three tubules arranged at definite angles. It has its own DNA and RNA and therefore it is self duplicating. Function: Centrioles are involved in cell division. They give orientation to the 'mitotic spindle' which forms during cell division.



Nucleus

General structure of nucleus

- It is the largest organelle seen clearly when the cell is not dividing.
- It stains deeply, is mostly spherical, WBC have lobed nuclei.
- It is mostly one in each cell (uni-nucleate, some cells have many nuclei; (multinucleate).
- Double layered nuclear membrane enclosing nucleoplasm which contains chromatin network and a nucleolus.

Functions

- Maintains the cell in a working order.
- Co-ordinates the activities of organelles.

- Takes care of repair work.
- Participates directly in cell division to produce genetically identical daughter cells, this division is called mitosis.
- Participates in production of gametes through another type of cell division called meiosis.

The part of a nucleus are given here

Nuclear membrane

- Double layered membrane is interrupted by large number of pores.
- Membrane is made up of lipids and proteins (like plasma membrane) and has ribosomes attached on the outer membrane which make the outer membrane rough.
- The pores allow the transport of large molecules in and out of nucleus, and the membranes keep the hereditary material in contact with the rest of the cell.

Chromatin

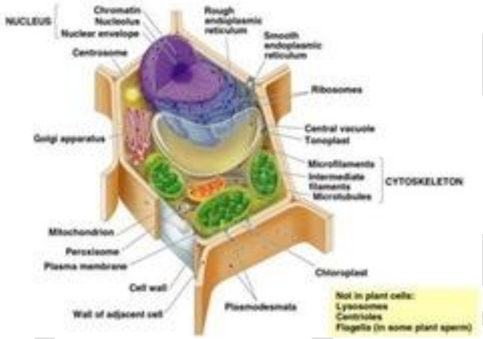
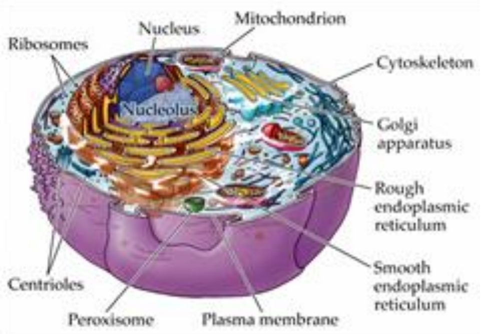
- Within the nuclear membrane there is jelly like substance (karyolymph or nucleoplasm) rich in proteins.
- In the karyolymph, fibrillar structures form a network called chromatin fibrils, which gets condensed to form distinct bodies called chromosomes during cell division. On staining the chromosomes, two regions can be identified in the chromatin material heterochromatin dark and autromaticn (light). Heterochromatin has less DNA and genetically less active than euchromatin which has more DNA and genetically more active.
- Number of chromosomes is fixed in an organism. During cell division chromosomes divide in a manner that the daughter cells receive identical amounts of hereditary matter.

Nucleolus

- Membraneless, spheroidal bodies present in all eukaryotic cells except in sperms and in some algae.

- Their number varies from one to few, they stain uniformly and deeply. It has DNA, RNA and proteins.
- Store house for RNA and proteins; it disappears during cell division and reappears in daughter cells.
- Regulates the synthetic activity of the nucleus.
- Thus nucleus and cytoplasm are interdependent, and this process is equal to nucleo–cytopalsmic interaction.

Differences between animal cell and plant cell

	Plant Cell		Animal Cell
1	A plant cell is usually larger in size.		An animal cell is comparatively smaller in size.
2	 **Plant Cell** It is enclosed by a rigid cellulose cell wall in addition to plasma membrane.		 **Animal Cell** It is enclosed by a thin, flexible plasma membrane only.
3	It cannot change its shape.		An animal cell can often change its shape.
4	Plastids are present. Plant cells exposed to sunlight contain chloroplast.		Plastids are usually absent.

5	A mature plant cell contains a large central vacuole.		An animal cell often possesses many small vacuoles.
6	Nucleus lies on one side in the peripheral cytoplasm.		Nucleus usually lies in the centre.
7	Centrioles are usually absent except in motile cells of lower plants.		Centrioles are practically present in animal cells
8	Lysosomes are rare.		Lysosomes are always present in animal cells.
9	Glyoxysomes may be present.		They are absent.
10	Tight junctions and desmosomes are lacking. Plasmodesmata are present.		Tight junctions and desmosomes are present between cells. Plasmodesmata are usually absent.
11	Reserve food is generally in the form of starch.		Reserve food is usually glycogen.
12	Plant cell synthesize all amino acids, coenzymes and vitamins required by them.		Animal cell cannot synthesize all the amino acids, co enzymes and vitamins required by them.
13	Spindles formed during cell divisions in anastral i.e. without asters at opposite poles.		Spindle formed during cell division is amphiastral i.e. has an ester at each pole.
14	Cytokinesis occurs by cell plate method.		Cytokinesis occurs by construction or furrowing.
15	Plant cell does not burst if placed in hypotonic solution due to the		Animal cell lacking contractile vacuoles usually burst, if placed in

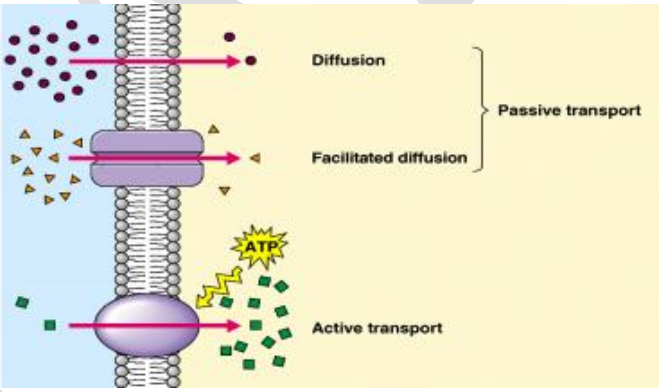
	presence of the cell wall.		hypertonic solution.

Transport of Molecules across Cell Membrane

Some endogenous substances and many drugs easily diffuse across the lipid bilayer. However, the lipid bilayer presents a formidable barrier to larger and more hydrophilic molecules (such as ions). These substances must be transported across the membrane by special proteins. The following are the types of transport:

There are two ways in which substances can enter or leave a cell:

1) Passive
    a) Simple Diffusion
    b) Facilitated Diffusion
    c) Osmosis (water only)
2) Active
    a) Molecules
    b) Particles



Simple Diffusion across the lipid bilayer

Since membranes are held together by weak forces, certain molecules can slip between the lipids in the bilayer and cross from one side to the other. This spontaneous process is termed diffusion. Diffusion is the movement of particles down their gradient. A gradient is any imbalance in concentration, and moving down a gradient just means that the particle is trying to be evenly distributed.

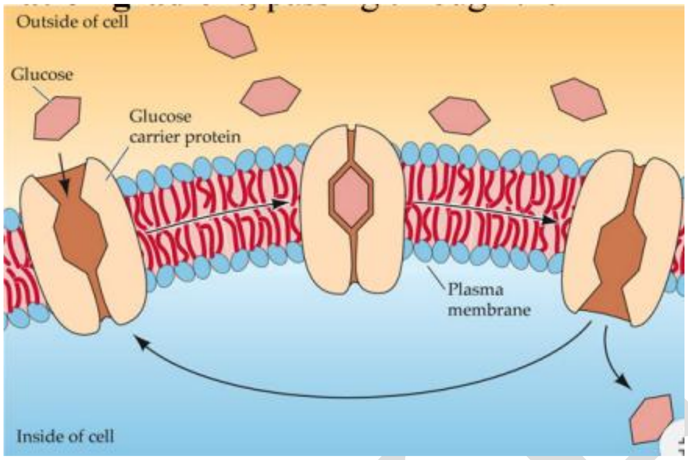
We call this evening-out moving “downhill”, and it doesn’t require energy. The molecule most likely to be involved in simple diffusion is water - it can easily pass through cell membranes.

Some major examples of diffusion in biology:

- Gas exchange at the alveoli — oxygen from air to blood, carbon dioxide from blood to air.
- Gas exchange for photosynthesis — carbon dioxide from air to leaf, oxygen from leaf to air.
- Gas exchange for respiration — oxygen from blood to tissue cells, carbon dioxide in opposite direction.
- Transfer of transmitter substance — acetylcholine from presynaptic to postsynaptic membrane at a synapse.
- Osmosis — diffusion of water through a semi-permeable membrane.

Facilitated Diffusion

This is the movement of specific molecules down a concentration gradient, passing through the membrane via a specific carrier protein. Thus, rather like enzymes, each carrier has its own shape and only allows one molecule (or one group of closely related molecules) to pass through. Selection is by size; shape; charge. Common molecules entering/leaving cells this way include glucose and amino-acids. It is passive and requires no energy from the cell. If the molecule is changed on entering the cell (glucose + ATP → glucose phosphate + ADP), then the concentration gradient of glucose will be kept high, and there will a steady one-way traffic.



When water undergoes simple diffusion, it is known as **osmosis**.

Osmosis is a special example of diffusion. It is the diffusion of water through a partially permeable membrane from a more dilute solution to a more concentrated solution – down the water potential gradient) Note: diffusion and osmosis are both passive, i.e. energy from ATP is not used. A partially permeable membrane is a barrier that permits the passage of some substances but not others; it allows the passage of the solvent molecules but not some of the larger solute molecules. Cell membranes are described as selectively permeable because not only do they allow the passage of water but also allow the passage of certain solutes.

The presence of particular solutes stimulates the membrane to open specific channels or trigger active transport mechanisms to allow the passage of those chemicals across the membrane.

Some major examples of osmosis

- Absorption of water by plant roots.
- Re-absorption of water by the proximal and distal convoluted tubules of the nephron.
- Re-absorption of tissue fluid into the venule ends of the blood capillaries.
- Absorption of water by the alimentary canal — stomach, small intestine and the colon.

There are 3 types of solutions that involve water and how they affect the cell. They are:

Hypertonic Solution: the solution the cell is placed in has less water than the cell

In a hypertonic solution, there is a higher concentration of water inside the cell than outside the cell. A hypertonic solution has more solute (salt, sugar, etc.) than the cell and this cause there to be less water in the solution. Water flows from an area of high concentration to an area of low and leaves the cell. This loss of water causes the cell to shrivel. In animal cells, the shriveling is called crenating. The red blood cells in the picture to the left have crenated.

In plant cells, plasmolysis occurs and the cell membrane shrinks away from the cell wall. Death will result in both cells.

Hypotonic Solution: the solution the cell is placed in has more water than the cell

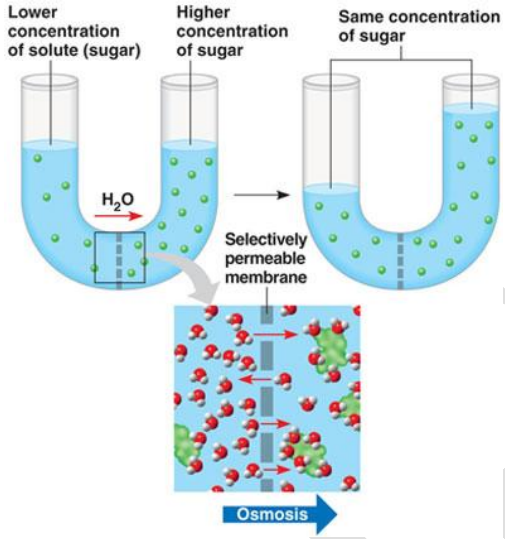
In a hypotonic solution, the solution contains a higher percentage of water than the cell. A hypotonic solution has less solute than the cell and this causes the solution to have more water than the cell. When a cell is placed in a hypotonic solution, water flows from an area of high concentration to an area of low and rushes into the cell. This causes the cell to expand and possibly burst. In animal cells, the cell bursts or will lyse, killing the cell.

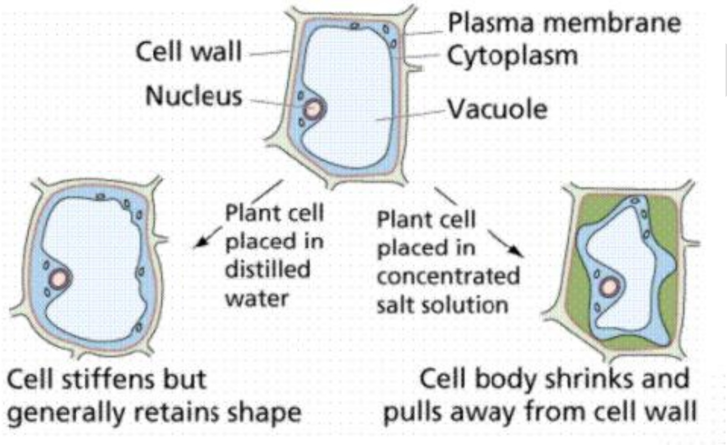
In plant cells, the cell membrane is pressed up against the cell wall but the cell wall does not allow the cell to expand anymore and the plant cell does not die.

Isotonic Solution: the solution the cell is placed in has equal amount of water as the cell

In an isotonic solution, there is the same percentage of water on the outside of the cell as the inside of the cell. An isotonic solution has the same amount of solute as the inside of the cell. Water moves at a constant rate in and out of the cell and the cell maintains its original shape.

In animal and plant cells, the cell keeps its shape when in an isotonic solution. Most cells live in an isotonic environment and they are able to maintain their shape and survive.





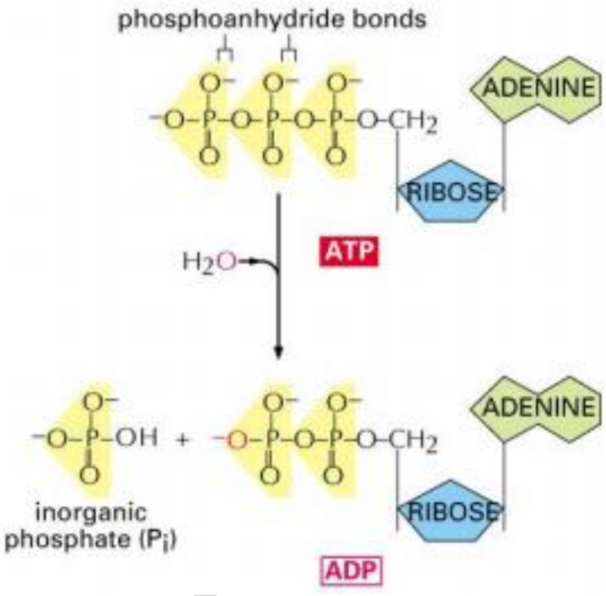
Active Transport

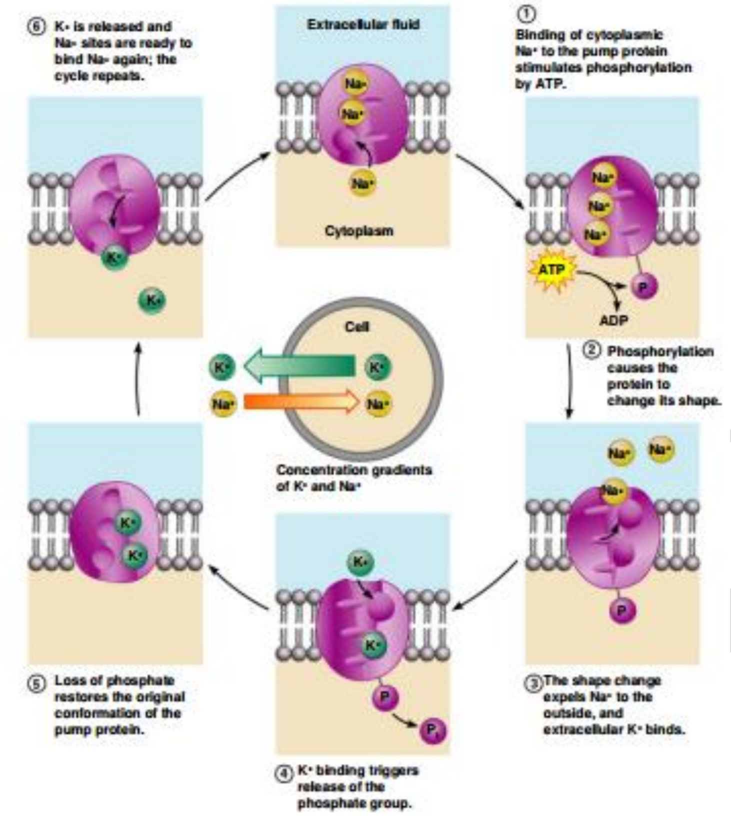
Transport method that moves particles from an area of low concentration to an area of high concentration or against a concentration gradient. Active transport relies on energy from the breakdown of ATP to move substances across the membrane. ATP, or adenosine triphosphate, is the main source of energy in the cell. The hydrolysis of the end phosphate group from an ATP molecule releases energy. The use of energy from ATP in active transport can be direct or indirect. Direct use of ATP is called primary active transport, and indirect use is called secondary active transport.

Primary active transport:

An example of primary active transport is the sodium-potassium pump or sodium-potassium ATPase (Na/K ATPase). The function of these pumps is to move 3 sodium ions from inside the cell to the extracellular fluid outside. It also pumps 2 potassium ions into the cell. So the concentration of K (potassium) is higher on the inside of the cell as it is on the outside and the concentration of Na (Sodium) is higher on the outside of the cell. Many cells use 1/3 to 1/2 of their ATP to run these ion pumps.

:





Secondary active transport

Secondary active transport does not use ATP. Instead it uses the energy stored from ion gradients. This is where an understanding of the sodium-potassium pump comes in handy. Let me explain. The Na/K ATPase moves Na actively by using ATP. It moves Na across its concentration gradient towards the outside of the cell, and in turn brings in K ions inside the cell.

As an example as to why this is important let's look at sugar, glucose. There is generally more glucose inside of the cell than outside. So, moving glucose across its gradient will generally take a lot of energy but not if we use sodium. Since we actively pump Na outside of the cell, the environment has more sodium than the inside of the cell.

This means that the sodium will want to move from outside the cell to inside. The cell uses this property to move glucose. It will allow sodium to re-enter the cell, but the sodium allows glucose to piggyback a ride into the cell alongside it. This is what is called co-transport and it uses a symporter.

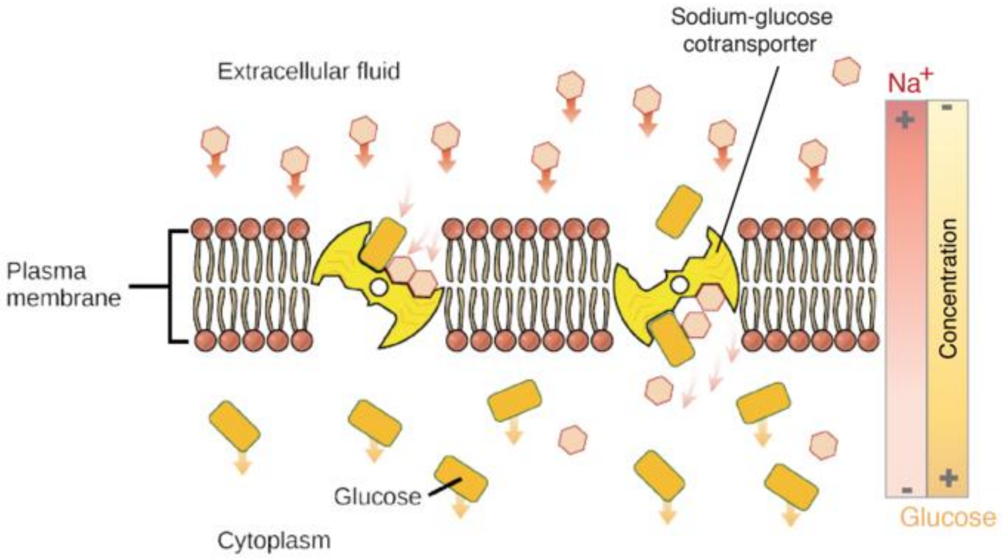
Symporter (Co-transport) is co-transport going in the same direction.

The other type of co-transport is exchange. This uses what is called an antiporter. The cell will move a solute of higher concentration outside the cell to inside the cell (Same example as Na). While the solute moves from higher to lower concentration, it generates energy. This is needed to move a molecule from the cell to outside of the cell, against its concentration gradient.

An example of this is Na/Ca exchanger (Sodium-Calcium). This is an example of an antiporter. It exchanged 3 sodium ions by moving it into the cell. For this exchange it will transport 1 calcium ion to the environment. This is similar to going to a store and exchanging money for items.

Antiporter (Exchange) is co-transport moving in the opposite direction.

The last type of port that uses the ion gradient is a **uniporter**. This is a simple port that moves an ion through a channel across its gradient. This is a type of diffusion and can be considered passive transport.



Differences between active and passive transport

	Active Transport	Passive Transport
Definition	Active Transport uses ATP to pump molecules AGAINST/UP the concentration gradient. Transport occurs from a low concentration of solute to high concentration of solute. Requires cellular energy.	Movement of molecules DOWN the concentration gradient. It goes from high to low concentration, in order to maintain equilibrium in the cells. Does not require cellular energy.
Types of Transport	Endocytosis, cell membrane/sodium-potassium pump & exocytosis	Diffusion, facilitated diffusion, and osmosis.
Functions	Transports molecules through the cell membrane against the concentration gradient so more of the substance is inside the cell (i.e. a nutrient) or outside the cell (i.e. a waste) than normal. Disrupts equilibrium established by diffusion.	Maintains dynamic equilibrium of water, gases, nutrients, wastes, etc. between cells and extracellular fluid; allows for small nutrients and gases to enter/exit. No NET diffusion/osmosis after equilibrium is established.
Types of Particles Transported	proteins, ions, large cells, complex sugars.	Anything soluble (meaning able to dissolve) in lipids, small monosaccharides, water, oxygen, carbon dioxide, sex hormones, etc.
	phagocytosis, pinocytosis, sodium/potassium pump, secretion	diffusion, osmosis, and facilitated diffusion.

	Active Transport	Passive Transport
Examples	of a substance into the bloodstream (process is opposite of phagocytosis & pinocytosis)	
Importance	In eukaryotic cells, amino acids, sugars and lipids need to enter the cell by protein pumps, which require active transport. These items either cannot diffuse or diffuse too slowly for survival.	It maintains equilibrium in the cell. Wastes (carbon dioxide, water, etc.) diffuse out and are excreted; nutrients and oxygen diffuse in to be used by the cell.

Beer and Lambert's Law

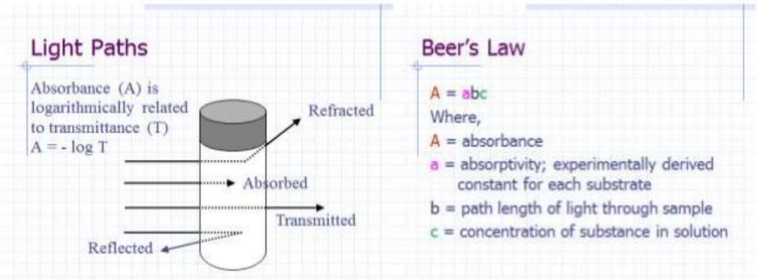
There are following possibilities when light is targeted to an object:

1. Transmission (if the object is transparent)
2. Absorption (if the object is liquid/gas or capable of absorption)
3. Reflection (if an opaque material is placed)
4. Refraction (if medium is changed during the path of light)
5. Scattering (if the surface is rough)

The Beer-Lambert law (also called the Beer-Lambert-Bouguer law or simply Beer's law) is the linear relationship between absorbance and concentration of an absorber of electromagnetic radiation. The general Beer-Lambert law is usually written as:

$$\mathbf{A = a_{\lambda} \cdot b \cdot c}$$

Where A is the measured absorbance, a_{λ} is a wavelength-dependent absorptivity coefficient, b is the path length, and c is the analyte concentration

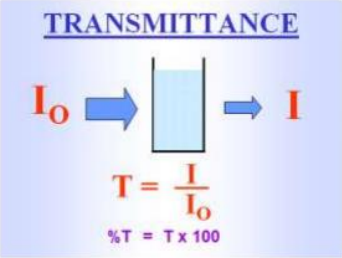


Light Paths

Absorbance (A) is logarithmically related to transmittance (T)
$A = - \log T$

Beer's Law

$A = abc$
Where,
A = absorbance
a = absorptivity; experimentally derived constant for each substrate
b = path length of light through sample
c = concentration of substance in solution

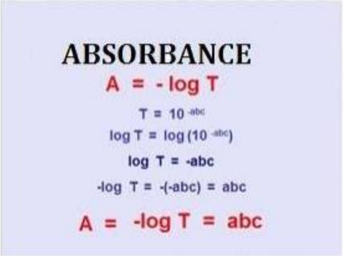


TRANSMITTANCE

$I_0 \rightarrow I$

$T = \frac{I}{I_0}$

$\%T = T \times 100$



ABSORBANCE

$A = - \log T$

$T = 10^{-abc}$

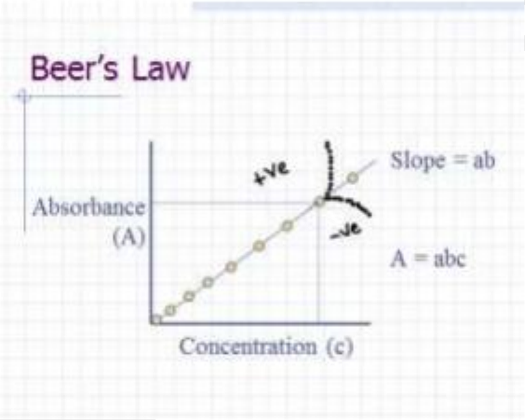
$\log T = \log(10^{-abc})$

$\log T = -abc$

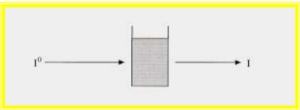
$-\log T = -(-abc) = abc$

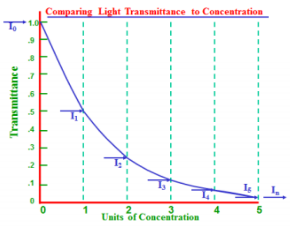
$A = -\log T = abc$

As given in the above figure, as the transmission increases, absorbance decreases exponentially.



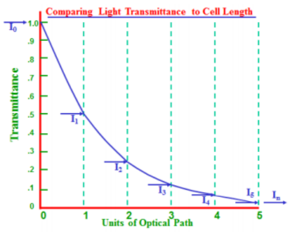
Beer's Law





Graph for Beer's Law

$I_t = I_o e^{-kc}$



Graph for Lambert's Law

$I_t = I_o e^{-kT}$

Colorimeter

A colorimeter is a device used to test the concentration of a solution by measuring its absorbance of a specific wavelength of light.

Principle

The colorimeter is based on Beer-Lambert's law, according to which the absorption of light transmitted through the medium is directly proportional to the medium concentration.

Instrumentation

The instrument use for colorimetry is colorimeter. This apparatus will comprise of the following parts:

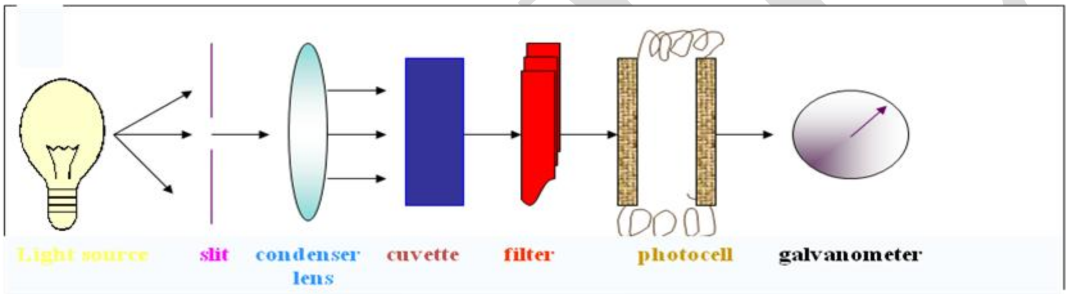
1. Light source
2. Filter (the device that selects the desired wavelength to obtain monochromatic light)
3. Cuvette chamber (the transmitted light passes through compartment wherein the solution containing the colored solution are kept in cuvette, made of glass or disposable plastic)
4. Detector (this is a photosensitive element that converts light into electrical signals)
5. Galvanometer (measures electrical signal quantitatively)

White light from a tungsten lamp passes through a slit, then a condenser lens, to give a parallel beam which falls on the solution under investigation contained in an absorption cell or cuvette. The cell is made of glass with the sides facing the beam cut parallel to each other.

Beyond the absorption cell is the filter, which is selected to allow maximum transmission of the color absorbed. If a blue solution is under examination, then red is absorbed and a red filter is selected.

The light then falls on to a photocell which generates an electrical current in direct proportion to the intensity of light falling on it.

This small electrical signal is increased by the amplifier which passes to a galvanometer of digital readout to give absorbance reading directly.



Anabolism and Catabolism

Metabolism is the set of life-sustaining chemical transformations within the cells of living organisms. These enzyme-catalyzed reactions allow organisms to grow and reproduce, maintain their structures, and respond to their environments. The word metabolism can also refer to all chemical reactions that occur in living organisms, including digestion and the transport of substances into and between different cells, in which case the set of reactions within the cells is called intermediary metabolism or intermediate metabolism.

The term metabolism is derived from the Greek – "Metabolismos" for "change", or "overthrow". The history of the scientific study of metabolism spans several centuries and has moved from examining whole animals in early studies, to examining individual metabolic reactions in modern biochemistry. The first controlled experiments in human metabolism were published by Santorio Santorio in 1614 in his book Ars de statica medicina. He described how he weighed himself before and after eating, sleep, working, sex, fasting, drinking, and excreting. He found that most of the food he took in was lost through what he called "insensible perspiration".

Catabolism is the metabolic process by which molecules are broken down and energy is produced. The body then receives the energy it needs to perform the most basic functions. Catabolism breaks down carbohydrates, amino acids, and lipids in the following stages.

- Step 1: Digestion

  First, large molecules are digested into smaller counterparts. This occurs for all molecules too large to be directly absorbed. ATP then pumps everything back into cells.
- Step 2: Energy Release Begins

  Acetyl coenzyme A further breaks down molecules, beginning the release of energy.
- Step 3: Energy Produced

  The molecules are oxidized, releasing all stored energy.

Carbohydrates are broken down into simple sugars, protein into amino acids, amino acids into keto acids, and fat lipids into free fatty acids and glycerol.

Anabolism is the process by which the body utilizes the energy released by catabolism to synthesize complex molecules.

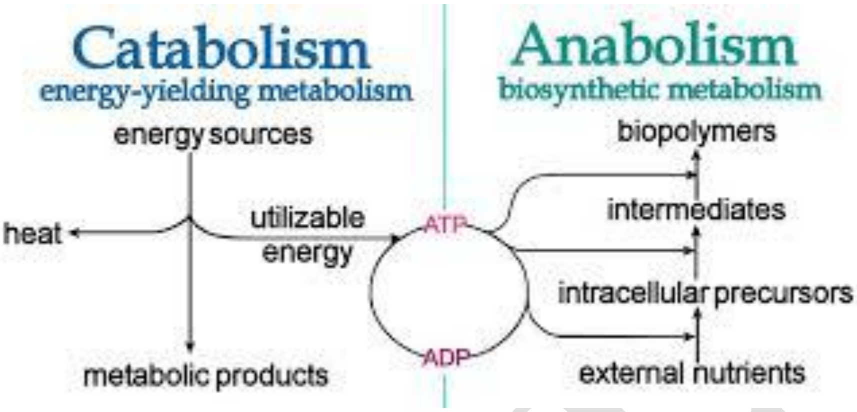
These complex molecules form cellular structures and act as the building blocks of the body. Though working in an opposite fashion to catabolism, anabolism similarly has 3 steps.

- Step 1: Precursor Production

  Precursors of complex molecules are produced, such as amino acids and monosaccharide’s.
- Step 2: Activation

  These precursors are activated into reactive forms by ATP.
- Step 3: Assembly

  Proteins, polysaccharides, lipids, and nucleic acids are assembled and ready for use.



Standard for energy change

The free energy change of a chemical process under standard state conditions, ΔG^o, can be determined four different ways:

- From Free Energies of Formation
- From Enthalpy Changes and Entropy Changes
- From Equilibrium Constants
- From Cell Potentials

Using Free Energies of Formation to Determine Standard State Free Energy Changes

If we know the standard free energy changes of formation, ΔG^o_f, of each species in a change we can determine the standard state free energy change, ΔG^o, for the change using the following equation:

$$\Delta G^o = \Sigma\ \Delta G^o_{f(products)} - \Sigma\ \Delta G^o_{f(reactants)}$$

Using Enthalpy Changes and Entropy Changes to Determine Standard State Free Energy Changes

If we know the enthalpy change, ΔH^o, and the entropy change, ΔS^o, for a chemical process, we can determine the standard state free energy change, ΔG^o, for the process using the following equation:

$$\Delta G^o = \Delta H^o - T\Delta S^o$$

In this equation T is the temperature on the Kelvin scale. In introductory courses we make the assumption that ΔH^o and ΔS^o, do not change as the temperature changes.

Using Equilibrium Constants to Determine Standard State Free Energy Changes

If we know the equilibrium constant, K_{eq}, for a chemical change (or if we can determine the equilibrium constant), we can calculate the standard state free energy change, ΔG^o, for the reaction using the equation:

$$\Delta G^0 = - RT \ln K_{eq}$$

In this equation

- $R = 8.314\ J\ mol^{-1}\ K^{-1}$ or $0.008314\ kJ\ mol^{-1}\ K^{-1}$.
- T is the temperature on the Kelvin scale.
- K_{eq} is the equilibrium constant at the temperature T.

Using Cell Potentials to Determine Standard State Free Energy Changes

If we know the standard state cell potential, E^o, for an electrochemical cell (or if we can determine the standard state cell potential), we can calculate the standard state free energy change, ΔG^o, for the cell reaction using the equation:

$$\Delta G^0 = -nFE^0$$

In this equation

- n is the number of moles of electrons exchanged in the cell reaction.
- F is 96.485 kJ $volt^{-1}$ $mole^{-1}$ (the "Faraday").

E^o is the cell potential under standard state conditions.

Karpagam Academy of Higher Education
Department of Biochemistry
I B.Sc., Microbiology
18MBU103 - Biochemistry

Question number	**Unit**	**Question**	Option I	Option II	Option III	Option IV	**Answer**
1	1	The most predominant chemical constituent of life	Water	protein	lipid	carbohydrate	**Water**
2	1	The cellular organelles regarded as the digestive tract of the cell	Nucleus	Golgi apparatus	mitochondria	endoplasmic reticulum	**endoplasmic reticulum**
3	1	Gases such as oxygen and carbon dioxide cross the plasma membrane by	secondary active transport	passive diffusion through the lipid bilayer	specific gas transport proteins	primary active transport	**passive diffusion through the lipid bilayer**
4	1	Which of the following is an example of primary active transport	Cl--HCO$_3$- exchange	Na$^+$ - H$^+$ exchange	Na$^+$-Ca^{2+} exchange	The Na$^+$, K$^+$ ATPase	**The Na$^+$, K$^+$ ATPase**
5	1	The sodium pump	Exchanges extracellular Na+ for intracellular K+	Is important for maintaining a constant cell volume	Can only be inhibited by metabolic poisons	Is an ion channel	**Is important for maintaining a constant cell volume**
6	1	A substance can only be accumulated against its electrochemical gradient by	Facilitated diffusion	Passage through ion channels	Diffusion through a uniport	Active transport	**Active transport**
7	1	The movement of molecules from an area of high concentration to an area of lower concentration is known as	Osmosis	Diffusion	Active Transport	Phagocytosis	**Diffusion**
8	1	What is the collective term for all of the chemical processes occurring within a cell?	Anabolism	catabolism	metabolism	synthesis	**metabolism**
9	1	Change in color of particular reactant can be detected in	Spectrometer	calorimeter	colorimeter	all of them	**colorimeter**
10	1	According to the Beer-Lambert Law, on which of the following	Distance that the light has travelled	Colour of the solution	Solution concentration	Extinction coefficient of	**Colour of the solution**

		does absorbance not depend?	through the sample			the sample	
11	1	What is the name of an instrument used to measure the absorbance of a coloured compound in solution?	Coulometer	Colourmeter	Colorimeter	Calorimeter	**Colorimeter**
12	1	The wavelength of an absorption is 495 nm. In what part of the electromagnetic spectrum does this lie?	Radiowave	Infrared	Ultraviolet-visible	Microwave	**Ultraviolet-visible**
13	1	Aqueous KMnO4 solutions are purple. A plot of absorbance against concentration is	linear with a positive gradient	non-linear	an exponential curve	linear with a negative gradient	**an exponential curve**
14	1	Ribosomes help in	Protein synthesis	Photosynthesis	Lipid synthesis	Respiration	**Protein synthesis**
15	1	Food is converted to energy in	Nucleus	Nucleolus	Chloroplast	Mitochondria	**Mitochondria**
16	1	Extra cellular DNA is found in	Chloroplast	Endoplasmic reticulum	Ribosomes	Nucleus	**Chloroplast**
17	1	Fluid mosaic model was given by	Robertson	Schwann	Dave Donson	Singer and Nicolson	**Singer and Nicolson**
18	1	The cellular organelles called “suicide bags” are	Lysosomes	Ribosomes	Nucleolus	Golgi’s bodies	**Lysosomes**
19	1	The power house of the cell is	Nucleus	Cell membrane	Mitochondria	Lysosomes	**Mitochondria**
20	1	The Golgi complex	Synthesizes proteins	Produces ATP	Provides a pathway for transporting chemicals	Forms glycoproteins	**Forms glycoproteins**
21	1	Plasma membrane is made up of	Protein, lipid, carbohydrate	Lipid, carbohydrate	Protein, lipid	Protein	**Protein, lipid, carbohydrate**
22	1	Plant cell is mainly composed of	Cellulose	Starch	Protein	Lipid	**Cellulose**
23	1	Anabolism and catabolism are types of	chemical reaction	chain reactions	metabolism	complex reactions	**metabolism**
24	1	Overall chemical reaction that takes place with in a cell are collectively called as	metabolism	anabolism	complex reaction	catabolism	**metabolism**

25	1	Study of chemical components as well as chemical processes that takes place in a living organism is	Microbiology	Biochemistry	Fresh water biology	Chemical biology	**Biochemistry**
26	1	Which of the following is a chemical link between catabolism and anabolism?	AMP	ADP	ATP	All of these	**ATP**
27	1	Tunnels which allow specific ions to pass through them are called	selectively permeable tunnels	permeable tunnels	both A and B	channel proteins	**channel proteins**
28	1	Type of transport which always involves a protein is	passive transport	active transport	lateral diffusion	flip flop	**active transport**
29	1	Mitochondrial DNA is	Circular double stranded	Circular single stranded	Linear double helix	None of these	**Circular double stranded**
30	1	Which of these is part of the cell membrane?	triglycerides	phospholipids	ATP	more than one of these	**phospholipids**
31	1	How do fat-soluble molecules normally get into a cell?	they dissolve in the fat layers of the membrane and enter the cell by diffusion	they pass through protein pores in the cell membrane	they are absorbed by phagocytosis	they never get in	**they dissolve in the fat layers of the membrane and enter the cell by diffusion**
32	1	The phospholipids are unusual molecules because:	they have hydrophilic regions	they have hydrophobic regions	they are triglycerides	both A and B	**they have hydrophobic regions**
33	1	Which of the following statements best describes the "fluid mosaic model" of the structure of the cell membrane?	two layers of protein with lipid layers between the protein layers	two layers of lipid with proteins between the lipid layers	a double layer of lipid molecules with protein molecules suspended in the layer	A single layer of protein on the outside and a single layer of lipids on the inside	**a double layer of lipid molecules with protein molecules suspended in the layer**
34	1	The movement of chloride ions from an area where chloride is concentrated to an area where chloride is less concentrated is which of these?	diffusion	active transport	osmosis	exocytosis	**diffusion**
35	1	If a cell has a solute concentration of 0.07% which of the solutions	0.01% solute	0.1% solute	1% solute	10% solute	**0.01% solute**

		would be hypotonic to the cell?					
36	1	Which of the following is necessary in order for osmosis to occur?	a permeable membrane	a semi-permeable membrane	an isotonic solution	ATP	**a permeable membrane**
37	1	Which of these are passive transport mechanisms?	osmosis	diffusion	phagocytosis	both A and B	**both A and B**
38	1	In an isotonic solution there would be:	no net movement of water	net movement of water into the cell	net movement of water out of the cell	bursting of the cell	**no net movement of water**
39	1	The sodium-potassium pump (which carries sodium out of a cell and potassium into a cell) is an example of:	active transport	endocytosis	exocytosis	passive transport	**active transport**
40	1	The process of a cell engulfing a solid object is:	phagocytosis	exocytosis	pinocytosis	diffusion	**phagocytosis**
41	1	What is likely to happen to a plant cell that is placed in pure water?	it becomes turgid	it becomes flaccid	it undergoes plasmolysis	it bursts	**it becomes turgid**
42	1	When a cell bursts due to osmosis, it is in a solution that is:	hypertonic	isotonic	hypotonic	either A or C	**hypotonic**
43	1	Why do plant cells behave differently to animal cells when placed in a hypotonic solution?	Plant cells are permeable to water	Plant cells do not carry out active transport	Plant cells contain a vacuole	Plant cells have a cell wall	**Plant cells have a cell wall**
44	1	Which of these equations is correct?	ATP + inorganic phosphate --> ADP	ADP + inorganic phosphate --> ATP	ATP + ADP --> inorganic phosphate	ATP + ADP --> organic phosphate	**ADP + inorganic phosphate --> ATP**
45	1	Atoms which have same number of protons but different number of neutrons are called	isotopes	isomers	spectators	allotropes	**isotopes**
46	1	To determine mass of other compound by comparing it with mass of carbon-12 atoms is	relative molecular mass	relative atomic mass	relative molecular radius	relative atomic radius	**relative atomic mass**
47	1	Sum of protons (p+) and neutrons (n0) in an atom is called its	atomic number	nucleon number	Avogadro's number	protonic identity	**nucleon number**
48	1	Chloride ion has number of	17	18	24	34	**17**

		protons of					
49	1	Smaller particles in atom are called	atomic particles	sub-atomic particles	smaller particles	neutral particles	**sub-atomic particles**
50	1	Number of protons and electrons in n atom is	different	same	average	constant	**same**
51	1	Electrons orbit around nucleus and bears	positive charge	negative charge	no charge	neutral charge	**negative charge**
52	1	Positively charged particle of atom is called	protons	neutrons	electrons	charges	**protons**
53	1	Neutrons carry	positive charge	negative charge	no charge	neutral charge	**no charge**
54	1	Total number of protons in an atom of each element is called its	atomic number	atomic mass	molecular mass	atomic scale	**atomic number**
55	1	Nucleus in an atom consists of	protons	neutrons	electrons	a and b	**a and b**
56	1	Molecules which contains fixed number of same type of atoms are molecules or	elements	compounds	mixtures	all of them	**elements**
57	1	If an atoms loses electron ion obtained is charged	positively	negatively	neutral	smaller	**positively**
58	1	If 3 Na+ ions pumped out of cell and 2 K+ pumped in to cell then number of ATP molecules hydrolyzed are	1	2	3	4	**1**
59	1	The filter color used to measure optical density of any blue color solution is	Blue	Yellow	Red	Green	**Blue**
60	1	The cuvette used for analysis of sample at UV range is	Glass cuvette	Quartz	Silica	All	**Quartz**

UNIT-II

SYLLABUS

Monosaccharides-families, stereo isomerism, epimers, mutarotation and anomers. Forms of glucose and fructose, Haworth projection. Sugar derivatives. Disaccharides-occurrence, concept of reducing and non-reducing sugars and Haworth projections. Polysaccharides-storage and structural polysaccharides.

Carbohydrates

A carbohydrate is a biological molecule consisting of carbon (C), hydrogen (H) and oxygen (O) atoms, usually with a hydrogen–oxygen atom ratio of 2:1 (as in water); in other words, with the empirical formula $C_m(H_2O)_n$ (where *m* could be different from *n*). Carbohydrates are hydrates of carbon; technically they are polyhydroxy aldehydes and ketones. Carbohydrates are also known as saccharides, the word saccharide comes from Greek word sakkron which means sugar.

Functions of Carbohydrate

All animals derive the major portion of their food calories from the different types of Carbohydrates in their diets. Most of the energy for the metabolic activities of the cell in all organisms is derived from the oxidation of Carbohydrate. Important functions of Carbohydrate are that of storing food, acting as a framework in body, performs are listed below.

Carbohydrate functions as Bio Fuel

Carbohydrate functions as an energy source of the body and acts as Bio fuel. Step wise details for the process of production of energy are discussed below.

- Polysaccharides such as starch and glycogen are first hydrolyzed by enzymes to Glucose.
- Glucose is the transported from one cell to another by blood in case of animals and cell sap in case of plants.
- Glucose is then oxidized to produce carbon dioxide and water.

- Energy is released in this process which is used for functioning of the cells.

Carbohydrate functions as Primary Source of Energy

The process of production of energy by carbohydrates is described in above steps. Now it is important to note, that fats and proteins can also be burned to provide energy but carbohydrate functions as primary source of energy. Fats are only burned if there is non availability of carbohydrates. When fat is burned in absence of carbohydrates, toxic compounds like called ketone bodies are produced. Accumulation of these ketone bodies over long period causes a condition called Ketosis. In this condition blood becomes unable to carry oxygen properly and this can be fatal. Thus, one of important function of carbohydrate is help burn fat properly.

Carbohydrate functions as storage food

Different forms of Carbohydrate are stored in living organism as storage food.

- Polysaccharide starch acts as storage food for plants.
- Glycogen stored in liver and muscles acts as storage food for animals.
- Insulin acts as storage food of dahlias, onion and garlic.

Thus carbohydrate performs the function of storing food.

Carbohydrate functions as framework in body

Different Carbohydrates especially Polysaccharides act as framework in living organism.

- Cellulose forms cell wall of plant cell along with hemicelluloses and Pectin
- Chitin forms cell wall of fungal cell and exoskeleton of arthropods
- Peptidoglycan forms cell wall of bacteria and cyanobacteria.

Thus carbohydrates function as contributing material to the cellular structure.

Carbohydrate functions as Anticoagulant

Heparin is a polysaccharide (carbohydrate) which acts as anticoagulant and prevents intravascular clotting.

Carbohydrate functions as Antigen

Many antigens are glycoprotein (which contains oligosaccharide) in nature and give immunological properties to the blood.

Carbohydrate functions as Hormone

Many Hormones like FSH (Follicular Stimulating Hormone which takes part in ovulation in females) and LH (Leteinizing Hormone) are glycoprotein and help in reproductive processes.

Carbohydrates provide raw material for industry

Carbohydrates are an important component of many industries like textile, paper, lacquers and breweries.

Other Functions

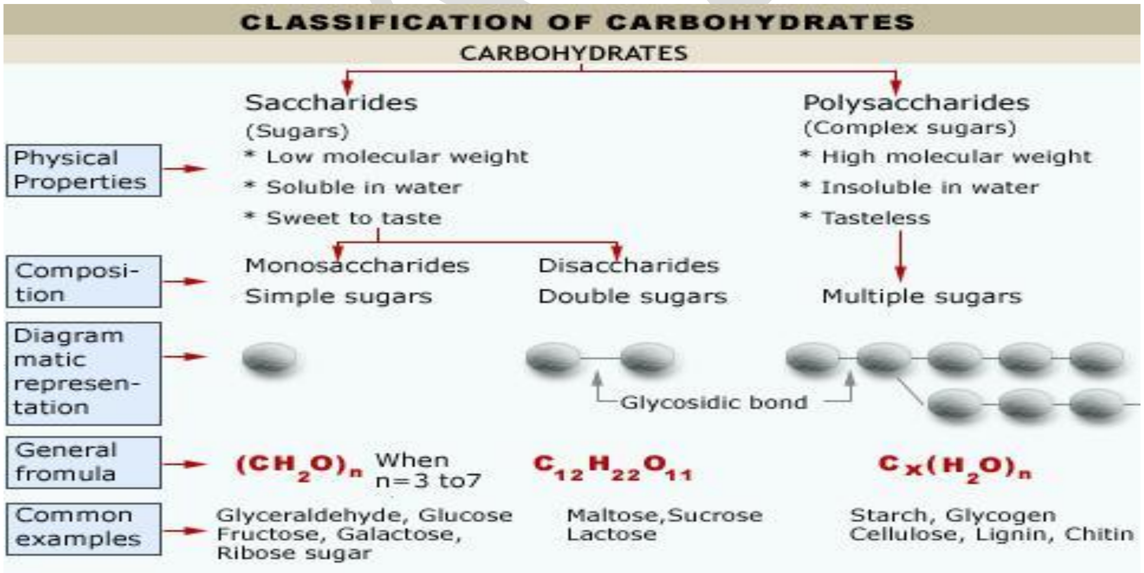
Agar is polysaccharide used in culture media, laxative and food.

Cellulose acts as roughage of food. It stimulates peristalsis movement and secretion of digestive enzymes.

Hyaluronic acid found in between joints acts as synovial fluid and provides frictionless movement.

Classes of carbohydrates

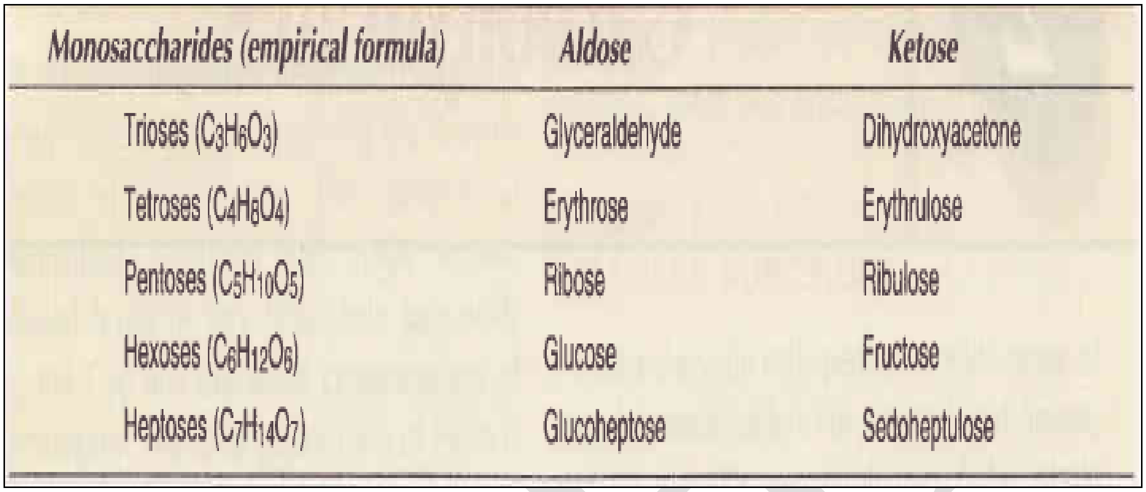
Carbohydrates are classified into three groups



Monosaccharides (From Greek, *mono*=one; *sakchron*=sugar)

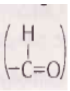
The following table shows the classification of monosaccharides based on the number of their carbon atoms, their general structure, and examples for each.

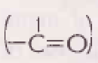
- They have the general formula $Cn(H_2O)_n$, and they cannot be further hydrolyzed.
- The monosaccharides are divided into different categories, based on the functional group and the number of carbon atoms.



Monosaccharides (empirical formula)	Aldose	Ketose
Trioses ($C_3H_6O_3$)	Glyceraldehyde	Dihydroxyacetone
Tetroses ($C_4H_8O_4$)	Erythrose	Erythrulose
Pentoses ($C_5H_{10}O_5$)	Ribose	Ribulose
Hexoses ($C_6H_{12}O_6$)	Glucose	Fructose
Heptoses ($C_7H_{14}O_7$)	Glucoheptose	Sedoheptulose

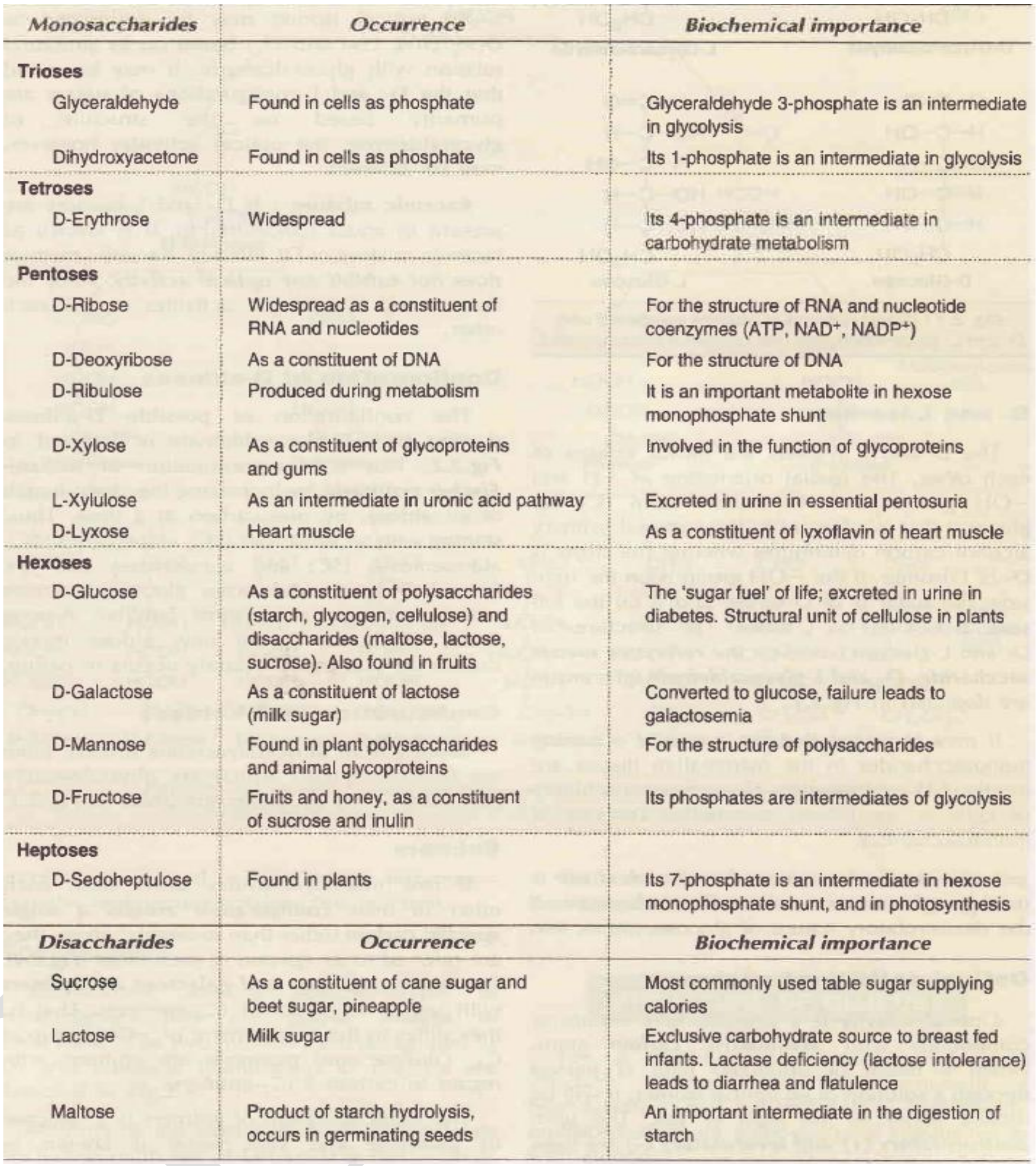
Classification of monosaccharide with selected examples

Aldoses: When the functional group in monosaccharides in aldehyde  $\left(\overset{\overset{H}{|}}{-C}=O\right)$ they are known as aldoses e.g. glyceraldehydes, glucose.

Ketoses: When the functional group is a keto  $\left(\overset{|}{-C}=O\right)$ group, they are referred to as ketoses

e.g. dihydroxyacetone, fructose.

- Based on the number of carbon atoms, the monosaccharides are regarded as trioses (3C), tetroses (4C), pentoses (5C), hexoses (6C) and heptoses (7C).
- These terms along with functional groups are used while naming monosaccharides.
- For instance, glucose is an aldohexose while fructose is a ketohexose.
- The common monosaccharides and disaccharides of biological importance are given.



Monosaccharides	*Occurrence*	*Biochemical importance*
Trioses		
Glyceraldehyde	Found in cells as phosphate	Glyceraldehyde 3-phosphate is an intermediate in glycolysis
Dihydroxyacetone	Found in cells as phosphate	Its 1-phosphate is an intermediate in glycolysis
Tetroses		
D-Erythrose	Widespread	Its 4-phosphate is an intermediate in carbohydrate metabolism
Pentoses		
D-Ribose	Widespread as a constituent of RNA and nucleotides	For the structure of RNA and nucleotide coenzymes (ATP, NAD^+, $NADP^+$)
D-Deoxyribose	As a constituent of DNA	For the structure of DNA
D-Ribulose	Produced during metabolism	It is an important metabolite in hexose monophosphate shunt
D-Xylose	As a constituent of glycoproteins and gums	Involved in the function of glycoproteins
L-Xylulose	As an intermediate in uronic acid pathway	Excreted in urine in essential pentosuria
D-Lyxose	Heart muscle	As a constituent of lyxoflavin of heart muscle
Hexoses		
D-Glucose	As a constituent of polysaccharides (starch, glycogen, cellulose) and disaccharides (maltose, lactose, sucrose). Also found in fruits	The 'sugar fuel' of life; excreted in urine in diabetes. Structural unit of cellulose in plants
D-Galactose	As a constituent of lactose (milk sugar)	Converted to glucose, failure leads to galactosemia
D-Mannose	Found in plant polysaccharides and animal glycoproteins	For the structure of polysaccharides
D-Fructose	Fruits and honey, as a constituent of sucrose and inulin	Its phosphates are intermediates of glycolysis
Heptoses		
D-Sedoheptulose	Found in plants	Its 7-phosphate is an intermediate in hexose monophosphate shunt, and in photosynthesis
Disaccharides	***Occurrence***	***Biochemical importance***
Sucrose	As a constituent of cane sugar and beet sugar, pineapple	Most commonly used table sugar supplying calories
Lactose	Milk sugar	Exclusive carbohydrate source to breast fed infants. Lactase deficiency (lactose intolerance) leads to diarrhea and flatulence
Maltose	Product of starch hydrolysis, occurs in germinating seeds	An important intermediate in the digestion of starch

Stereoisomerism of monosaccharides:

All the monosaccharides except dihydroxyacetone contain one or more asymmetric (chiral) carbon atoms and thus occur in optically active isomeric forms. The simplest aldose, glyceraldehyde, contains one chiral center (the middle carbon atom) and therefore has two different optical isomers, or **enantiomers**

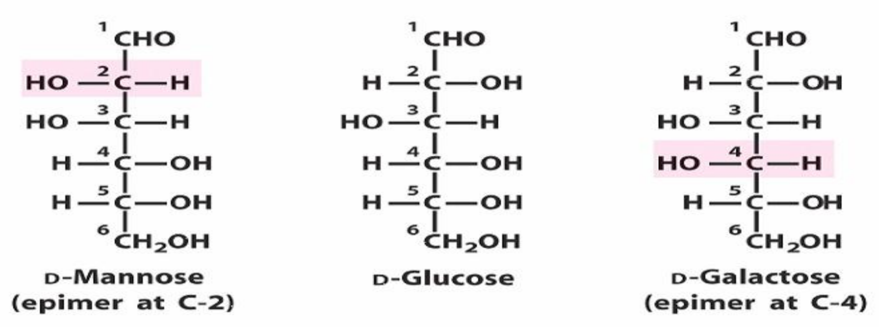
- Carbon 2 of glyceradehyde is a chiral center.

- There are thus 3 steroisomers of glyceraldehydes: D-glyceraldehyde and L-glyceraldehyde.
- By convention, sugars are written with the most oxidized carbon (i.e. aldehyde or ketone) at the top.
- The chiral center farthest from the most oxidized carbon determines if it is D or L.
- If the hydroxyl points to the left, then it is the L configuration if to the right then it is D.
- In general, only the D isomers are used biologically, but there are many exceptions to this generalization.
- Sugars can be conveniently written as Fischer projections to indicate stereochemistry.
- The most oxidized carbon is placed at the top and each carbon between it and the last carbon is a cross from which are appended the hydrogen and hydroxyl group.
- It makes a difference if the hydroxyl group is written to the or lift.
- It is important to recognize that a Fischer projection indicates the stereochemistry of each chiral center.
- One must imagine that the groups to the left and right (-H and –OH) are coming out the plane towards the viewer, while the substituent's above and below are out of the plane directed away from the viewer.
- In general, a molecule with n chiral centers can have $2n$ stereoisomers. Glyceraldehyde has $2^1 = 2$; the aldohexoses, with four chiral centers, have $2^4 = 16$ stereoisomers.

Epimers

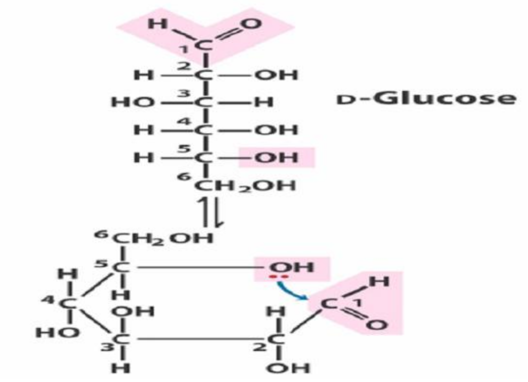
Carbohydrates that differ only in their stereochemistry at one position are called Epimers.

- Eg. Glucose and mannose (C-2)
- Glucose and galactose (C-4)



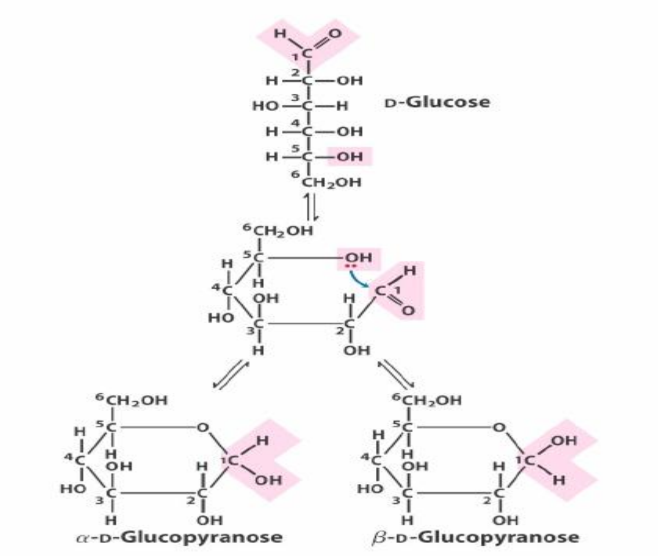
Mutarotation and anomers:

- In aqueous solution, D-glucose exists in one of 2 forms: α-D-glucose and ß-D-glucose.
- This is because Aldehydes can react with alcohols to form a hemiacetal.
- In this case, the hydroxyl oxygen attacking the molecule it is an intermolecular reaction, which results in formation of a ring.
- Rings with 6 members are the most stable, but 5-membered rings are possible.
- The oxygen that attacked the carbonyl carbon will be a member of the ring.
- The carbonyl oxygen is converted to a hydroxyl group in the process.
- The stereochemistry of this hydroxyl group is determined by the position of the carbonyl during the attack; it can be one of 2 possible configurations: α or ß.
- Six-member rings resemble pyran and are referred to as pyranosides.
- Five – member rings resemble furan and are referred to as furanosides.



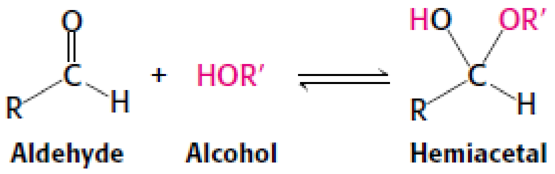
Anomers

- Isomeric forms of monosaccharides that differ only in their configuration about the hemiacetal or hemiketal carbon atom are called **anomers**. The hemiacetal (or carbonyl) carbon atom is called the **anomeric carbon**. The _ and _ anomers of D-glucose interconvert in aqueous solution by a process called **mutarotation**
- The aldehyde or ketone carbon is referred to as the anomeric carbon, as this is the chiral center that differs between 2 Anomers.
- For D-sugars the anomer has the hydroxyl group down in the Haworth projection and on the same side as the ring oxygen in the Fisher projection.

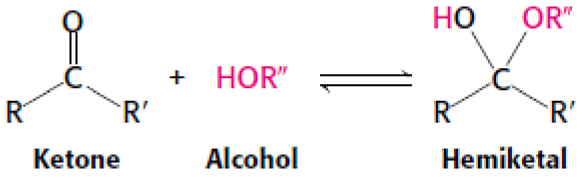


Haworth Projections

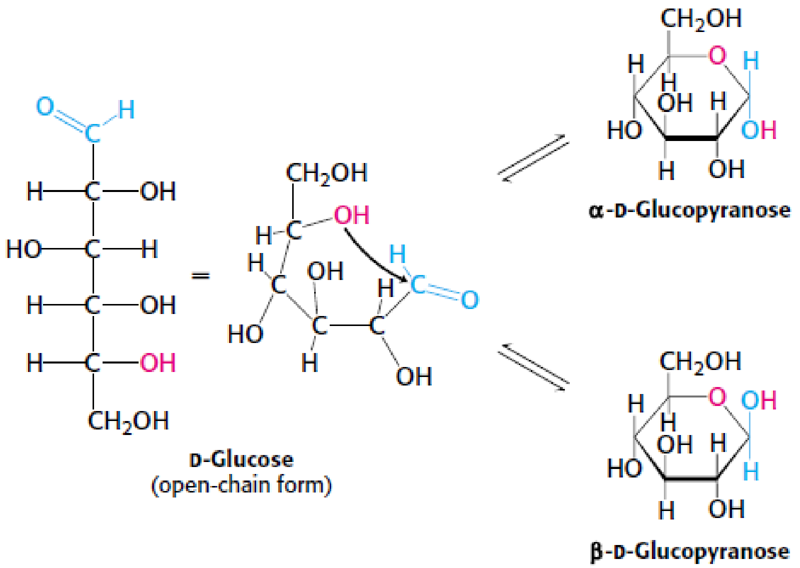
The predominant forms of ribose, glucose, fructose, and many other sugars in solution are not open chains. Rather, the open-chain forms of these sugars cyclize into rings. In general, an aldehyde can react with an alcohol to form a hemiacetal.

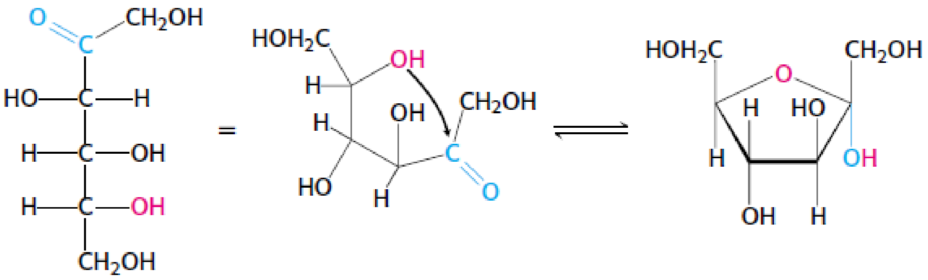


For an aldohexose such as glucose, the C-1 aldehyde in the open-chain form of glucose reacts with the C-5 hydroxyl group to form an intramolecular hemiacetal. The resulting cyclic hemiacetal, a six-membered ring, is called pyranose because of its similarity to pyran. Similarly, a ketone can react with an alcohol to form a hemiketal.



The C-2 keto group in the open-chain form of a ketohexose, such as fructose, can form an intramolecular hemiketal by reacting with either the C-6 hydroxyl group to form a six-membered cyclic hemiketal or the C-5 hydroxyl group to form a five-membered cyclic hemiketal. The five-membered ring is called a furanose because of its similarity to furan.





D-Fructose
(open-chain form)

α-D-Fructofuranose
(a cyclic form of fructose)

The depictions of glucopyranose and fructofuranose shown below are Haworth projections. In such projections, the carbon atoms in the ring are not explicitly shown. The approximate plane of the ring is perpendicular to the plane of the paper, with the heavy line on the ring projecting toward the reader. Like Fischer projections, Haworth projections allow easy depiction of the stereochemistry of sugars.

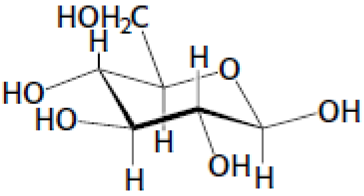
An additional asymmetric center is created when a cyclic hemiacetal is formed. In glucose, C-1, the carbonyl carbon atom in the open-chain form, becomes an asymmetric center. Thus, two ring structures can be formed: α-D-glucopyranose and β-D-glucopyranose. For D sugars drawn as Haworth projections, the designation α means that the hydroxyl group attached to C-1 is below the plane of the ring; β means that it is above the plane of the ring. The C-1 carbon atom is called the anomeric carbon atom, and α and β forms are called anomers. An equilibrium mixture of glucose contains approximately one-third α anomer, two-thirds β anomer, and <1% of the open-chain form.

The same nomenclature applies to the furanose ring form of fructose, except that α and β refer to the hydroxyl groups attached to C-2, the anomeric carbon atom. Fructose forms both pyranose and furanose rings. The pyranose form predominates in fructose free in solution, and the furanose form predominates in many fructose derivatives. Pentoses such as D-ribose and 2-deoxy-D-ribose form furanose rings, as we have seen in the structure of these units in RNA and DNA.

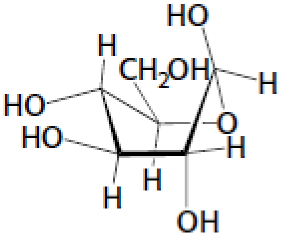
Chair and boat forms of glucose

The six-membered pyranose ring is not planar, because of the tetrahedral geometry of its saturated carbon atoms. Instead, pyranose rings adopt two classes of conformations, termed chair and boat because of the resemblance to these objects. In the chair form, the substituents on the ring carbon atoms have two orientations: axial and equatorial. Axial bonds are nearly perpendicular to the average plane of the ring, whereas equatorial bonds are nearly parallel to this plane. Axial substituents sterically hinder each other if they emerge on the same side of the ring (e.g., 1,3-diaxial groups). In contrast, equatorial substituents are less crowded. The chair form of β-D-glucopyranose predominates because all axial positions are occupied by hydrogen atoms. The bulkier –OH and –CH2OH groups emerge at the less-hindered periphery. The boat form of glucose is disfavored because it is quite sterically hindered.

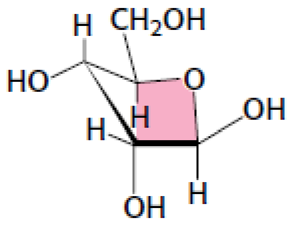
Furanose rings, like pyranose rings, are not planar. They can be puckered so that four atoms are nearly coplanar and the fifth is about 0.5 Å away from this plane. This conformation is called an envelope form because the structure resembles an opened envelope with the back flap raised. In the ribose moiety of most biomolecules, either C-2 or C-3 is out of the plane on the same side as C-5. These conformations are called C2- endo and C3-endo, respectively.



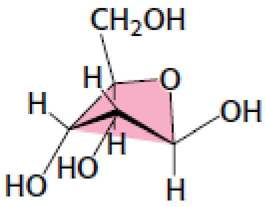
Chair form



Boat form



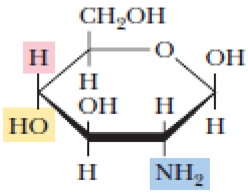
C_3-endo



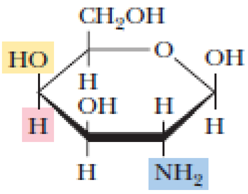
C_2-endo

Sugar Derivatives

Amino sugars, including D-glucosamine and D-galactosamine, contain an amino group (instead of a hydroxyl group) at the C-2 position. They are found in many oligosaccharides and polysaccharides, including chitin, a polysaccharide in the exoskeletons of crustaceans and insects.



β-D-Glucosamine



β-D-Galactosamine

Glucosamine

Glucosamine ($C_6H_{13}NO_5$) is an amino sugar and a prominent precursor in the biochemical synthesis of glycosylated proteins and lipids. Glucosamine is part of the structure of the polysaccharides chitosan and chitin, which compose the exoskeletons of crustaceans and other arthropods, as well as the cell walls of fungi and many higher organisms. Glucosamine is one of the most abundant monosaccharides. It is produced commercially by the hydrolysis of crustacean exoskeletons or, less commonly, by fermentation of a grain such as corn or wheat.

Glucosamine is naturally present in the shells of shellfish, animal bones, bone marrow, and fungi. D-Glucosamine is made naturally in the form of glucosamine-6-phosphate, and is the biochemical precursor of all nitrogen-containing sugars.[30]Specifically in humans, glucosamine-6-phosphate is synthesized from fructose 6-phosphate and glutamine by glutamine fructose-6-phosphate transaminase as the first step of the hexosamine biosynthesis pathway. The end-product of this pathway is uridine diphosphate N-acetylglucosamine (UDP-GlcNAc), which is then used for making glycosaminoglycans,proteoglycans, and glycolipids.

Galactosamine

Galactosamine is a hexosamine derived from galactose with the molecular formula $C_6H_{13}NO_5$. This amino sugar is a constituent of some glycoprotein hormones such as follicle-stimulating hormone (FSH) and luteinizing hormone (LH). Other sugar constituents of FSH and LH include glucosamine, galactose and glucose. Galactosamine is a hepatotoxic, or liver-damaging, agent that is sometimes used in animal models of liver failure.

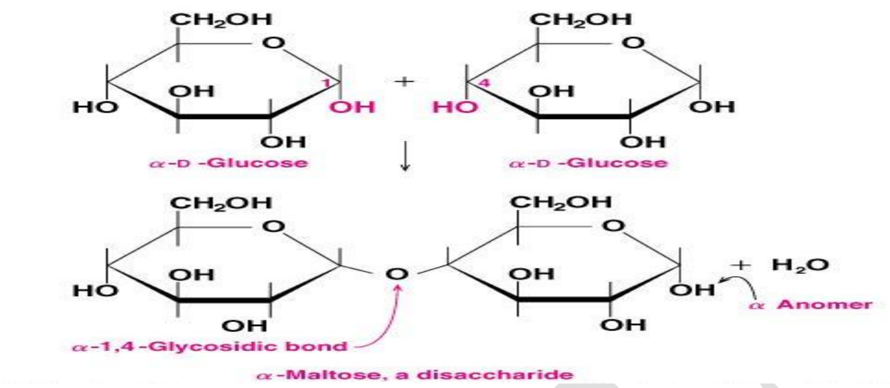
Disaccharides

- A disaccharide is formed when a hydroxyl group on one monosaccharide reacts with the anomeric carbon of another monosaccharide to form a glycosidic bond.
- Each disaccharide has a specific glycosidic linkage (depending on which hydroxyl reacts with which anomer).
- The three most common disaccharides are **maltose, lactose and sucrose.**
- When hydrolyzed using acid or an enzyme, the following monosaccharide are produced.
- The disaccharides are of two types
  1. Reducing disaccharides with free aldehyde or keto group e.g. maltose, lactose.
  2. Non-reducing disaccharides with no free aldehyde or keto group e.g. sucrose,

Maltose

Occurrence: Not occur in our body, but present in germinating cereals and malt; It is the breakdown product of starch

Structure: Maltose (malt sugar or corn sugar) is composed of two glucose molecules are joined through α-1,4 glycosidic linkage



Properties

- Because one of the glucose molecules is a hemiacetal (having a free aldehyde group) it can undergo mutarotation (Gradual change in specific rotation; Glucose if freshly prepared have sp rotation of +112· , but on standing gives a rotation of + 52.).
- It exist in α and β forms
- Since it is having a free aldehyde group, it reduce compounds and and so maltose is a reducing sugar.
- Reduce Fehling and Benedicts solution but not Barfoeds solution
- Forms osazone with phenyl hydrazine
- Maltose can be fermented by yeast to produce ethanol.
- Maltose is also used in cereals, candies and malted milk.

  Hydrolysis: Hydrolyzed by maltase present in alimentary canal; two glucose molecules are released upon hydrolysis

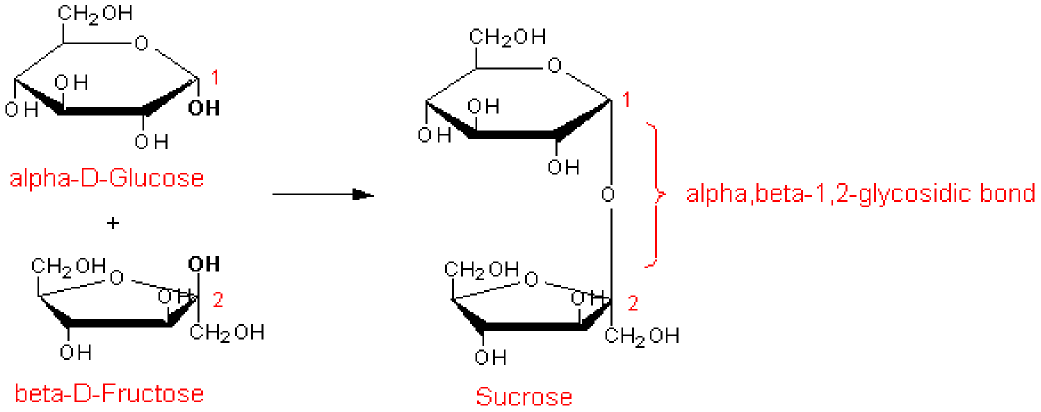
Sucrose

Occurrence

It is the sweetest of all the sugars; does not exist in our body, occur in cane sugar, pineapple, carrot root, sweet potato and honey. Sucrose is the most abundant disaccharide and is commercially produced from sugar cane and sugar beets.

Structure

Sucrose (table sugar) consists of one glucose molecule and one fructose molecule linked by an α,β-1,2-glycosidic bond.



- It is not having a free aldehyde or ketone group, so don't have mutarotation; does not exist in α and β forms. **Because the glycosidic bond in sucrose involves both** anomeric carbons, neither monosaccharide can undergo mutorotation, and so sucrose is not a reducing sugar.

Properties

- White crystalline solid powder; sparingly soluble in water
- The specific rotation of fructose is 66.5, but upon hydrolysis it is changed to -19.5. This because the hydrolyzed product, fructose, which is having more levorotary than the glucose. This reaction is called inversion and the sugar is called invert sugar.
- it does not reduce Fehling , Benedicts s and Barfoeds solution
- it cannot from crystals with phenyl hydrazine
- **Hydrolysis:**
- Hydrolyzed by sucrose present in alimentary canal; one glucose and one fructose molecules are released upon hydrolysis

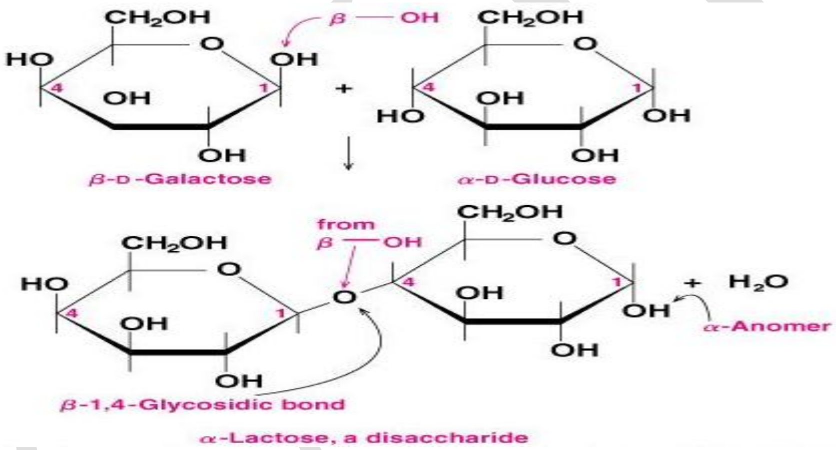
Lactose

Occurrence

- Present in human milk (9.8%) produced by mammary gland of human beings; It comes from milk products (about 4-5% of cow's milk).; also occur in urine during pregnancy.

Structure

- **Lactose** (milk sugar) consists of one glucose molecule and one galactose molecule linked by a β-1,4 glycosidic bond.



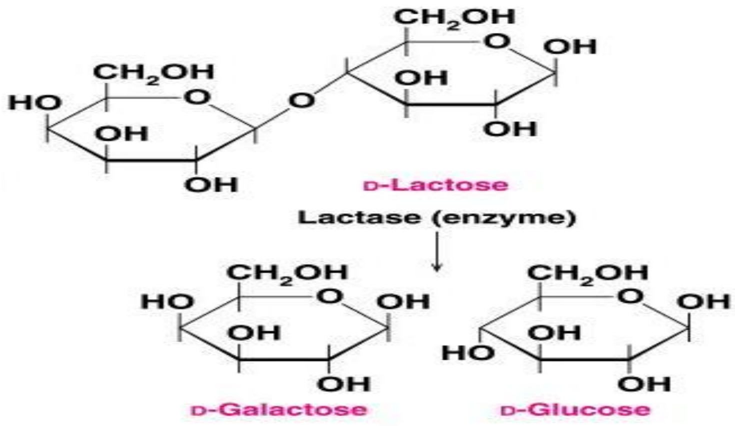
- Because the glucose is a hemiacetal, it can undergo mutorotation, and it is having a free aldehyde group, which reduce compounds and so lactose is a reducing sugar.

Properties

- White crystalline solid powder; sparingly soluble in water
- The specific rotation is + 55.2·
- Exist in α and β forms
- Reduce Fehling and Benedicts solution but not Barfoeds solution
- Forms osazone with phenyl hydrazine

Hydrolysis of Lactose

Hydrolyzed by lactase present in alimentary canal; one glucose and one galactose molecules are released upon hydrolysis.



- Some people don’t produce enough lactase, the enzyme that hydrolyzes lactose, and so can’t digest lactose.
- Many adults become lactose intolerant, and develop abdominal cramps, nausea and diarrhea.
- Lactase can be added to milk products (or taken as a supplement) to combat this problem

Polysaccharides

A **polysaccharide** is a polymer consisting of hundreds to thousands of monosaccharide joined together by glycosidic linkages.

They are further classified into

Homopolysaccharides

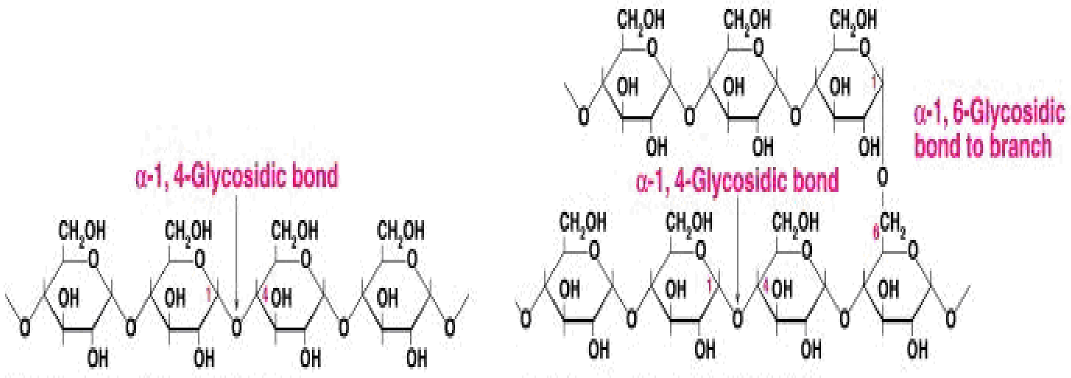
1. Storage polysaccharides: Eg-Starch (Plant); glycogen (animal)
2. Structural polysaccharides: Eg-Cellulose (Plant); Chitin (animal)

Heteropolysaccharides

1. Glycoproteins
2. Glycosaminoglycans

- Heparin/Heparin sulfate

- Chondroitin sulfate
- Keratin sulfate
- Hyaluronic acid
- Three biologically important polysaccharides are **starch, glycogen** and **cellulose** all three are polymers of D-glucose, but they differ in the type of glycosidic bond and/or the amount of branching
- Starch and glycogen are used for storage of carbohydrates
- Starch is found in plants and glycogen in animals
- The polymers take up less room than would the individual glucose molecules, so are more efficient for storage
- Cellulose is a structural material used in formation of cell walls in plants Plant Starch (Amylose and Amylopectin)



(a) Unbranched chain of amylose

(b) Branched-chain of amylopectin

Storage polysaccharides

Starch

- Half of the carbohydrate ingested by human is starch.
- It is the source of carbohydrates and fundamental source of energy.
- Starch is the carbohydrate reserve of plants which is the most important dietary source for higher animals, including man.

Occurrence: It is the storage form of carbohydrate in plants ; It is present in cereals, potato, and legumes, root, tubers, tubers, vegetables etc fruits. It is found as granules in cytoplasm of chloroplast

Structure

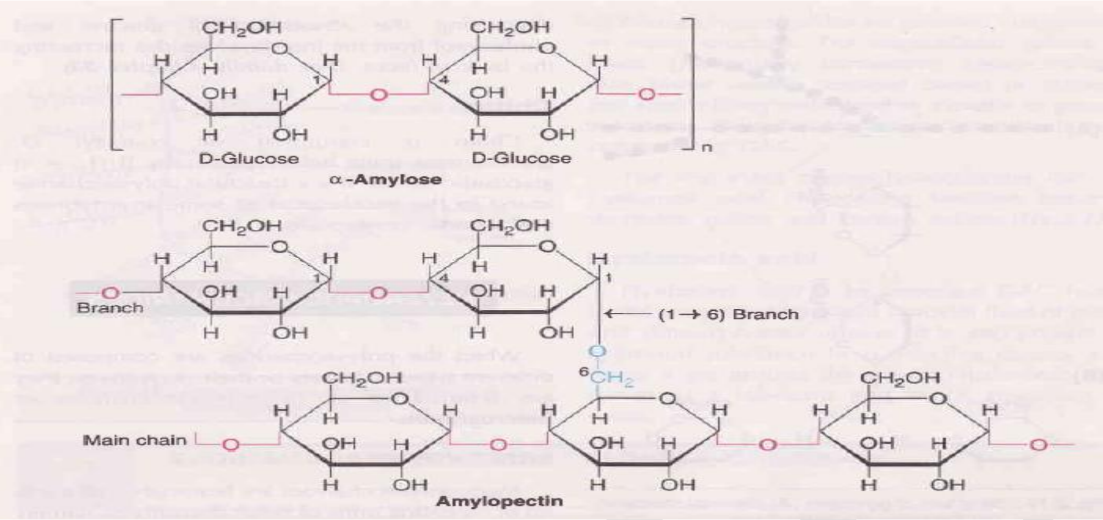
- Starch is a homopolymer composed of D-glucose units held by a-glycosidic bonds.
- It is known as glucosan or glucan.
- Starch consists of two polysaccharide components-water soluble amylose (15-20%) and a water insoluble amylopectin (80-85%).

Amylose (α- amylose)

It is a long unbranched polysaccharide; made of α- D glucose joined by α (1 → 4) glycosidic linkage. In starch it constitute about 15-20%. It is in the helical form and 6 glucose unit per turn. It have nearly 300-400 glucose units; molecular weight is 1000-50,000. It form blue color with iodine.

Amylopectin (β-amylose)

Amylopectin on the other hand, is a branched polysaccharide atleast 80 branch with an interval of 24-30 glucose units(20-30 glucose units per branch).It is made of α- D glucose joined by α 1,4 glycosidic linkage and the branch is established with α 1,6 glycosidic linkage(α (1 → 6) glycosidic bonds at the branching points and α (1 → 4) linkages everywhere). In starch it constitute about80-85%. It have nearly 300-5500 glucose units; molecular weight is 5,00,000. It form blue colour with iodine.



Structure of starch (α-amylose and amylopectin)

Properties of starch

White , soft powder, tasteless; insoluble in water; specific rotation is+196.

Hydrolysis

Starch is a glucosan, because it yields only glucose molecule on hydrolysis; with water it form hydrated micelle

- Starches are hydrolyzed by amylase (pancreatic or salivary) to liberate dextrins, and finally maltose and glucose units.
- Amylase acts specifically on a (1 → 4) glycosidic bonds.

α- amylase

Amylose ------→ Maltose + glucose

α- amylase attacks the α 1,4 glycosidic linkage. It is present in saliva

α- amylase/ β- amylase

Amylo pectin -------------→ Maltose + glucose

α- amylase attacks the α 1,4 glycosidic linkage. It is present in saliva. α 1,6 glycosidic linkage is attacked by as α 1,6 glucosidase

Starch with mineral acid gives glucose. This glucose reacts with iodine and give gradual change in colour ie., -blue-----------purple------------red------------none

Starch on partial hydrolysis yield dextrin which gives stiffness to cloths

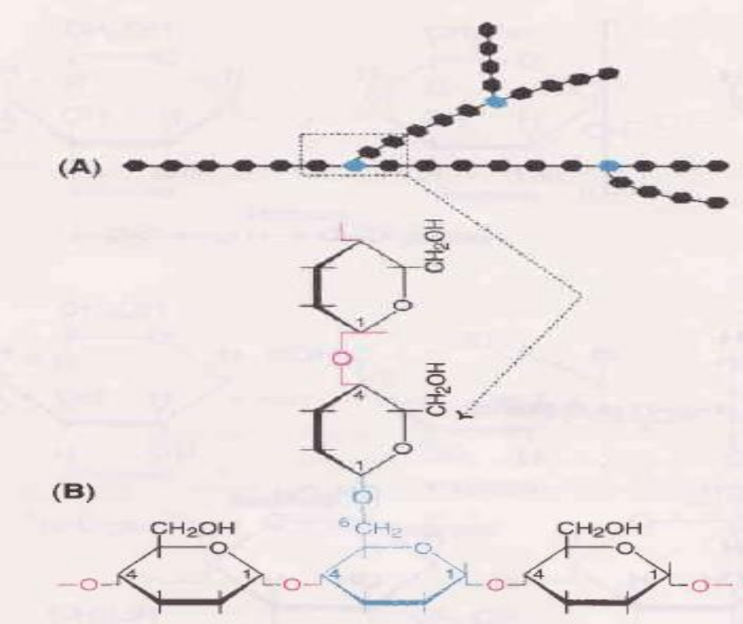
Glycogen

Glycogen is the carbohydrate reserve in animals, hence often referred to as animal starch. It is the reserve carbohydrate found in liver and muscle of animal and human beings

It is present in high concentration in liver, followed by muscle, brain etc. Liver have more glycogen (7% of its weight) than muscle. Glycogen is also found in plants that do not possess chlorophyll (e.g. yeast, fungi).

Structure

- The structure of glycogen is similar to that of amylopectin with more number of branches. It is a branched polymer of carbohydrate ; made of α-D glucose; Glucose is the repeating unit in glycogen joined together by α (1 → 4) glycosidic bonds, and α (1 → 6) glycosidic bonds at branching points, the branching is established by α 1,6 glycosidic linkage.
- The molecular weight (up to 1 x 10^8) and the number of glucose units (up to 5000-25,000) vary in glycogen depending on the source from which glycogen is obtained.



Structure of glycogen (A) General structure (B) Enlarged at a branch point

Properties

White, tasteless powder; readily soluble in water; Non reducing; give red color with iodine

Hydrolysis

On complete hydrolysis, glycogen yields glucose and maltose

Dextrin

This is formed by the partial (incomplete) hydrolysis of starch by salivary amylase; and also by dilute mineral acid and heat.

Inulin

It is a fructosan; made of repeating units of fructose. It is found in roots and tubers of dahlia and dandelions; it mainly used in assessing the kidney function.

Structural Polysaccharides

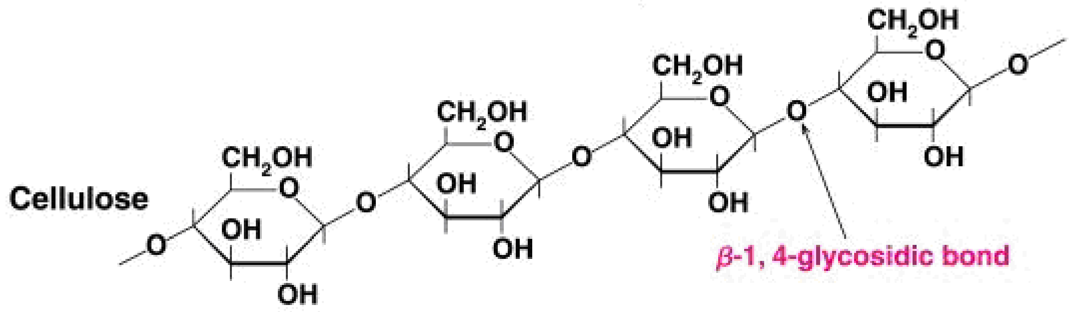
Structural polysaccharides are the polysaccharides that are found to form the structure of an organism.

Eg. Cellulose - in plants
Chitin - found in outer skeleton of insects and crabs
Lignin - wood

Cellulose

It is the most abundant of all biomolecule in biosphere. 50% carbon in vegetation is contributed by cellulose. In plant, it is the main constituent of supporting tissue. It is not present in animal.

- Cellulose is a polymer made with repeated glucose units bonded together by *beta*-linkages.
- The structural components of plants are formed primarily from cellulose.
- Wood is largely cellulose and lignin, while paper and cotton are nearly pure cellulose.



Properties

- Cellulose is insoluble in water. It does not change color when mixed with iodine. On hydrolysis, it yields glucose. It is the most abundant carbohydrate in nature.
- Fibrous, tough, white solid; insoluble in ordinary solvents and water; give no color with iodine.

Hydrolysis

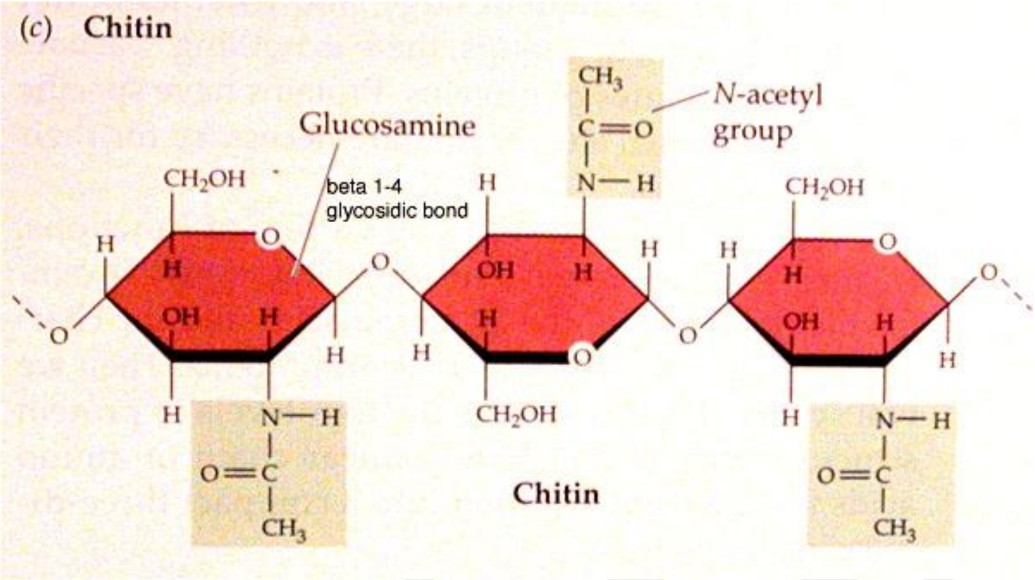
- Humans and many other animals lack an enzyme to break the *beta*-linkages, so they do not digest cellulose.
- Certain animals such as termites can digest cellulose, because bacteria possessing the enzyme are present in their gut.
- It is not acted upon by amylase in human intestine, so doesn't have any nutritive value. It adds bulk to the intestinal constituents and stimulates the peristaltic movement of bowel so it aids in relieving constipation.
- On complete hydrolysis by cellulase enzyme it yields α-D glucose. This enzyme is mainly present in termites, which are able to digest the wood.
- It is also hydrolyzed by acids such as sulfuric acid, nitric acid and sodium hydroxide.

Chitin

Chitin is a polysaccharide found in the outer skeleton of insects, crabs, shrimps, and lobsters and in the internal structures of other invertebrates.

Structure

It is a long-chain polymer of a *N*-acetylglucosamine, a derivative of glucose, Joined through ß(1-4) linked units of the amino sugar N-acetyl-glucosamine.



Properties

In its unmodified form, chitin is translucent, pliable, resilient, and quite tough. but in most invertebrates it occurs largely as a component of composite materials

Application

Chitin is the main source of production of chitosan, which is used in a number of applications, such as a flocculating agent, a wound healing agent, a sizing and strengthening agent for paper, and a delivery

Karpagam Academy of Higher Education
Department of Biochemistry
I B.Sc., Microbiology
18MBU103 - Biochemistry

Question number	**Unit**	**Question**	Option I	Option II	Option III	Option IV	**Answer**
1	2	Two monosaccharides are joined by	Peptide bond	Phosphodiester bond	Glycosidic bond	Hydrogen bond	**Glycosidic bond**
2	2	All the following are storage polysaccharides except	Starch	Cellulose	Dextran	Glycogen	**Dextran**
3	2	The glycosidic linkage between glucose molecule in maltose is	β 1 – 4	α 1 – 2	α 1 – 4	β 1 – 2	**α 1 – 4**
4	2	The glycosidic linkage between two glucose molecules in isomaltose is	α 1 – 4	β 1 – 4	α 1 – 6	β 1 – 6	**α 1 – 6**
5	2	Which of the following sugar give a positive result with Seliwanoff test	Sucrose	Glucose	Galactose	Mannose	**Sucrose**
6	2	Maltose is a disaccharide of______	Glucose and galactose	Glucose and glucose	Glucose and lactose	Fructose and lactose	**Glucose and glucose**
7	2	Glycosidic bond in sucrose is _________	α 1 – 4	β 1 – 4	α 1 – 2	β 1 – 2	**β 1 – 2**
8	2	Majority of the monosaccharides found in the human body are of …	L-type	D-type	DL-types	None of the above	**D-type**
9	2	Example of Epimers is	Glucose & Galactose	Glucose & Ribose	Mannose & Glucose	a & c	**a & c**
10	2	The end product of hydrolysis of “Starch” by amylase is …	Soluble starch	Glucose	Dextrins	Maltose	**Glucose**

11	2	Cellulose fibers resemble with the protein structure in the form of	ß-sheets	α-helices	ß-turns	None of these	**ß-sheets**
12	2	Hydrolysis of lactose yields	galactose and fructose	galactose and glucose	glucose and fructose	fructose and galactose	**galactose and glucose**
13	2	Boat and chair conformations are found	in pyranose sugars	in any sugar without axial -OH groups	in any sugar without equatorial -OH groups	only in D-glucopyranose	**in pyranose sugars**
14	2	Storage polysaccharide made by animals is	amylopectin	glycogen	cellulose	collagen	**glycogen**
15	2	The glycosaminoglycan which does not contain uronic acid is	Dermatan sulphate	Chondroitin sulphate	Keratan sulphate	Heparan sulphate	**Keratan sulphate**
16	2	Keratan sulphate is found in abundance in	Heart muscle	Liver	Adrenal cortex	Cornea	**Cornea**
17	2	Repeating units of hyaluronic acid are	N-acetyl glucosamine and D-glucuronic acid	N-acetyl galactosamine and D-glucuronic acid	N-acetyl glucosamine and galactose	N-acetyl galactosamine and L-iduronic acid	**N-acetyl glucosamine and D-glucuronic acid**
18	2	The approximate number of branches in amylopectin is	10	20	40	80	**80**
19	2	In amylopectin the intervals of glucose units of each branch is	10–20	24–30	30–40	40–50	**24–30**
20	2	The general formula for polysaccharide is	(C6H10O5)n	(C6H12O5)n	(C6H10O6)n	(C6H10O6)n	**(C6H10O5)n**
21	2	α-D-glucose and β -D-glucose are	Stereoisomers	Epimers	Anomers	Keto-aldo pairs	**Anomers**
22	2	The general formula of monosaccharides is	CnH2nOn	C2nH2On	CnH2O2n	CnH2nO2n	**CnH2nOn**
23	2	The aldose sugar is	Glycerose	Ribulose	Erythrulose	Dihydoxyacetone	**Glycerose**
24	2	A triose sugar is	Glycerose	Ribose	Erythrose	Fructose	**Glycerose**
25	2	A pentose sugar is	Dihydroxyacetone	Ribulose	Erythrose	Glucose	**Ribulose**
26	2	The pentose sugar present mainly in the heart muscle is	Lyxose	Ribose	Arabinose	Xylose	**Lyxose**

27	2	Polysaccharides are	Polymers	Acids	Proteins	Oils	**Polymers**
28	2	The number of isomers of glucose is	2	4	8	16	**16**
29	2	Two sugars which differ from one another only in configuration around a single carbon atom are termed	Epimers	Anomers	Optical isomers	Stereoisomer s	**Epimers**
30	2	Isomers differing as a result of variations in configuration of the —OH and —H on carbon atoms 2, 3 and 4 of glucose are known as	Epimers	Anomers	Optical isomers	Steroisomers	**Epimers**
31	2	The most important epimer of glucose is	Galactose	Fructose	Arabinose	Xylose	**Galactose**
32	2	α-D-glucose + 112.0 → + 52.50 ← + 19.0 β-D-glucose for glucose above represents	Optical isomerism	Mutarotation	Epimerisation	D and L isomerism	**Mutarotation**
33	2	Compounds having the same structural formula but differing in spatial configuration are known as	Stereoisomers	Anomers	Optical isomers	Epimers	**Epimers**
34	2	In glucose the orientation of the —H and —OH groups around the carbon atom 5 adjacent to the terminal primary alcohol carbon determines	D or L series	Dextro or levorotatory	α and β anomers	Epimers	**D or L series**
35	2	The sugar found in milk is	Galactose	Glucose	Fructose	Lactose	**Lactose**
36	2	Invert sugar is	Lactose	Sucrose	Hydrolytic products of sucrose	Fructose	**Hydrolytic products of sucrose**
37	2	Sucrose consists of	Glucose + glucose	Glucose + fructose	Glucose + galactose	Glucose + mannose	**Glucose + fructose**
38	2	The monosaccharide units are linked by 1 → 4 glycosidic linkage in	Maltose	Sucrose	Cellulose	Cellobiose	**Maltose**
39	2	Which of the following is a non-reducing sugar?	Isomaltose	Maltose	Lactose	Trehalose	**Trehalose**
40	2	Which of the following is a reducing sugar?	Sucrose	Trehalose	Isomaltose	Agar	**Isomaltose**
41	2	A dissaccharide formed by 1,1-glycosidic linkage between their monosaccharide units is	Lactose	Maltose	Trehalose	Sucrose	**Trehalose**
42	2	A polysacchharide which is often called animal starch is	Glycogen	Starch	Inulin	Dextrin	**Glycogen**
43	2	The homopolysaccharide used for intravenous	Agar	Inulin	Pectin	Starch	**Agar**

		infusion as plasma substitute is					
44	2	The polysaccharide used in assessing the glomerular fittration rate GFR) is	Glycogen	Agar	Inulin	Hyaluronic acid	**Inulin**
45	2	The constituent unit of inulin is	Glucose	Fructose	Mannose	Galactose	**Fructose**
46	2	The polysaccharide found in the exoskeleton of invertebrates is	Pectin	Chitin	Cellulose	Chondroitin sulphate	**Chitin**
47	2	Which of the following is a heteroglycan?	Dextrins	Agar	Inulin	Chitin	**Agar**
48	2	A positive Benedict’s test is not given by	Sucrose	Lactose	Maltose	Glucose	**Sucrose**
49	2	Starch is a	Polysaccharide	Monosaccharide	Disaccharide	None of these	**Polysaccharide**
50	2	A positive Seliwanoff’s test is obtained with	Glucose	Fructose	Lactose	Maltose	**Fructose**
51	2	Osazones are not formed with the	Glucose	Fructose	Sucrose	Lactose	**Sucrose**
52	2	The most abundant carbohydrate found in nature is	Starch	Glycogen	Cellulose	Chitin	**Cellulose**
53	2	The total Glucose in the body is ________ gms.	10–15	20–30	40–50	60–80	**20–30**
54	2	Whcih of the following features are common to monosaccharides?	Contain asymmetric centres	Are of 2 types – aldoses and ketoses	Tend to exist as ring structures in solution	Include glucose, galactose and raffinose	**Tend to exist as ring structures in solution**
55	2	The following examples are important heteropolysaccharides except	Amylopectin	Heparin	Peptidoglycan	Hyaluronic acid	**Amylopectin**
56	2	Glucosamine is an important constituent of	Homopolysaccharide	Heteropolysaccharide	Mucopolysaccharide	Dextran	**Mucopolysaccharide**
57	2	Glycogen is present in all body tissues except	Liver	Brain	Kidney	Stomach	**Brain**
58	2	Iodine test is positive for starch, dextrin and	Mucoproteins	Agar	Glycogen	Cellulose	**Glycogen**
59	2	The distinguishing test between monosaccharides and dissaccharide is	Bial’s test	Seliwanoff’s test	Barfoed’s test	Hydrolysis test	**Barfoed’s test**
60	2	Cane sugar is known as	Galactose	Sucrose	Fructose	Maltose	**Sucrose**

UNIT-III

SYLLABUS

Classification and functions of lipids, storage lipids-structure and function of fatty acids. Triacylglycerols. Saponification. Structural lipids-structure, functions and properties of phosphoglycerides and sphingolipids.

Lipids

Definition

- The lipids are heterogeneous group of compounds related to fatty acids.
- They constitute a broad group of naturally occurring molecules that include fats, waxes, sterols, fat-soluble vitamins (such as vitamins A, D, E, and K), monoglycerides, diglycerides, triglycerides, phospholipids, and others.

Biological significance

The main biological functions of lipids include

- Fat serve as an efficient sources of energy storage,
- Serve as insulating material
- Helps in blood clotting
- Serve as structural components of cell membranes, and as important signaling molecules.
- Lipoproteins and glycolipids are important for maintaining cellular integrity.

Classification of Lipids

- They are broadly classified into simple lipids, complex lipids, derived lipids and miscellaneous lipids based on their chemical composition.

Simple lipids

Esters of fatty acids with alcohols. These are mainly of two types

Fats and oils (triacylglycerols)

- These are esters of fatty acids with glycerol.
- The difference between fat and oil is only physical.

- Thus, oil is a liquid while fat is a solid at room temperature.

Waxes

- Esters of fatty acids (usually long chain) with alcohols other than glycerol.
- These alcohols may be aliphatic or alicyclic.
- Cetyl alcohol is most commonly found in waxes.

Complex (or compound) lipids

- These are esters of fatty acids with alcohols containing additional groups such as phosphate, nitrogenous base, carbohydrate, protein etc.
- They are further divided as follows

Phospholipids

- They contain phosphoric acid and frequently a nitrogenous base.
- This is in addition to alcohol and fatty acids.

(i) Glycerophospholipids: These phospholipids contain glycerol as the alcohol e.g., lecithin, cephalin.

(ii) Sphingophospholipids: Sphingosine is the alcohol in this group of Phospholipids e.g., sphingomyelin.

Glycolipids

- These lipids contain a fatty acid, carbohydrate and nitrogenous base.
- The alcohol is sphingosine; hence they are also called as glycosphingolipids.
- Glycerol and phosphate are absent e.g., cerebrosides, gangliosides.

Lipoproteins

- Macromolecular complexes of lipids with proteins.

Other complex lipids

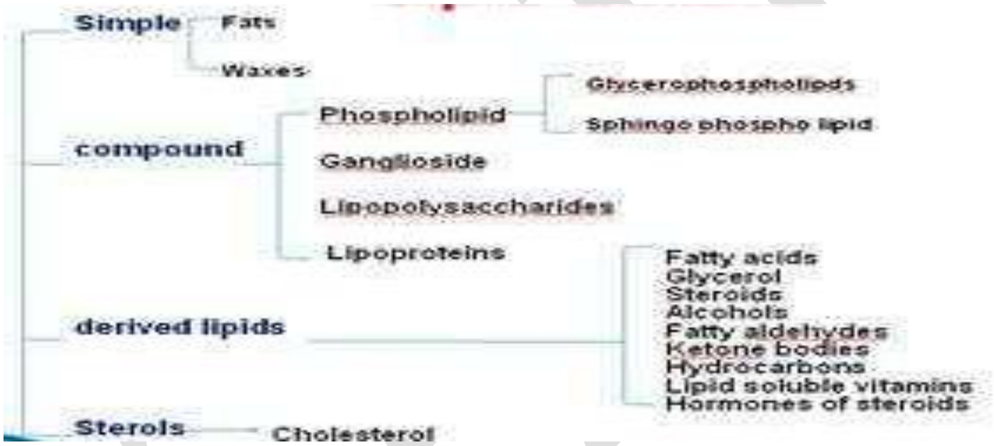
- Sulfolipids, amino lipids and lipopolysaccharides are among the other complex lipids.

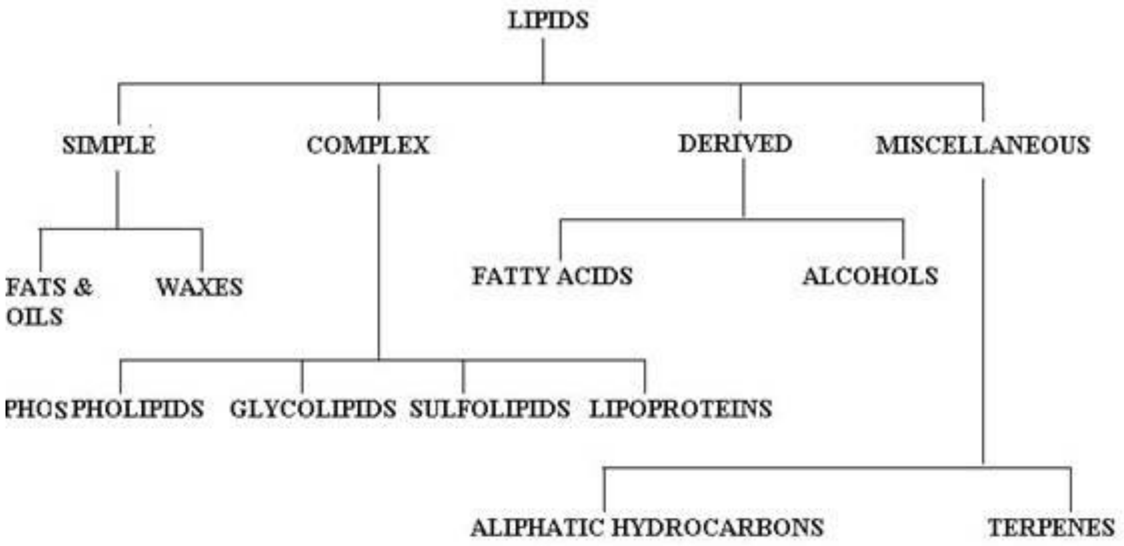
Derived lipids

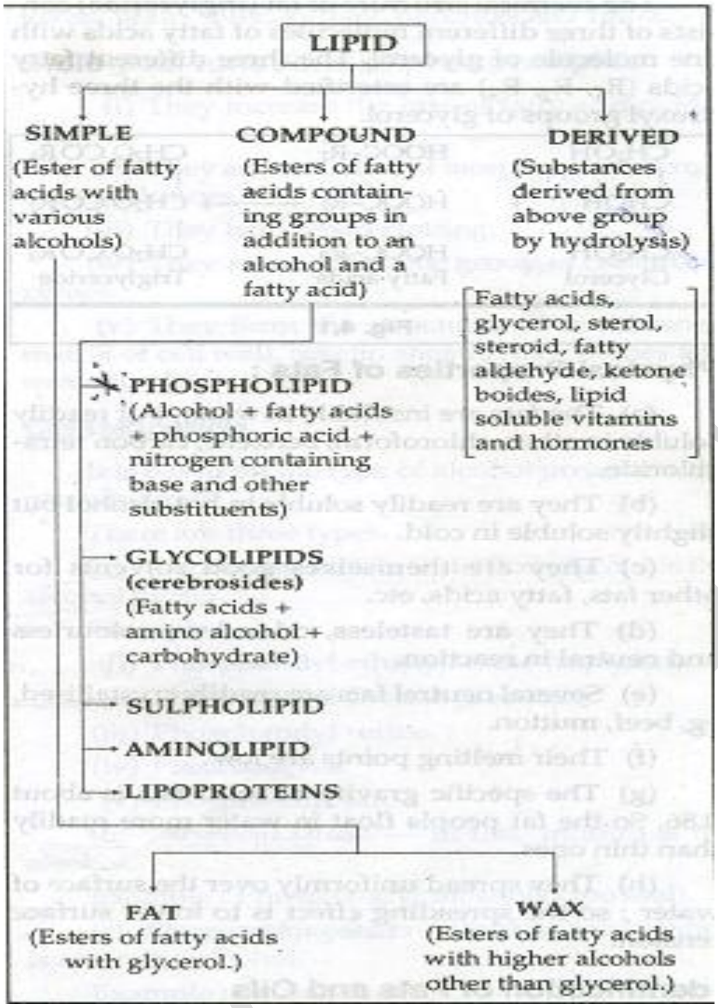
- These are the derivatives obtained on the hydrolysis of group 1 and group 2 lipids which possess the characteristics of lipids.
- These include glycerol and other alcohols, fatty acids, mono- and diacylglycerols, lipid (fat) soluble vitamins, steroid hormones, hydrocarbons and ketone bodies.

Miscellaneous lipids

- These include a large number of compounds possessing the characteristics of lipids e.g., carotenoids, squalene, hydrocarbons such as pentacosane (in bees wax), terpenes etc.




LIPIDS
SIMPLE
COMPLEX
DERIVED
MISCELLANEOUS
FATS & OILS
WAXES
FATTY ACIDS
ALCOHOLS
PHOSPHOLIPIDS
GLYCOLIPIDS
SULFOLIPIDS
LIPOPROTEINS
ALIPHATIC HYDROCARBONS
TERPENES



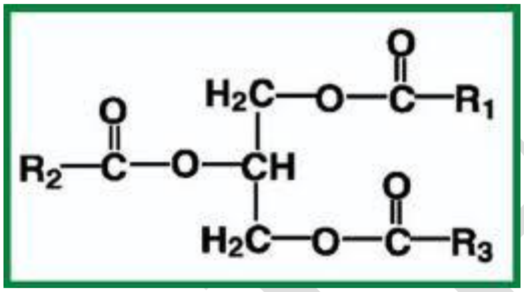
Simple Lipids

Fats

- Fat, any substance of plant or animal origin that is nonvolatile, insoluble in water, and oily or greasy to the touch. They are esters of fatty acids with glycerol.
- They are found in nature in large quantities. Fats are usually solid at ordinary temperatures, such as 25 °C (77 °F), but they begin to liquefy at somewhat higher temperatures.

- Chemically, fats are identical to animal and vegetable oils, consisting primarily of glycerides, which are esters formed by the reaction of three molecules of fatty acids with one molecule of glycerol.

  Eg-Triacylglycerol



  - They are the best reserve of food material in the human body.
  - They act as insulator for the loss of body heat.
  - They act as a padding material for protecting internal organs.
  - The chemical structure of fat (triglyceride) consists of three different molecules of fatty acids with one molecule of glycerol. The three different fatty acids (R_1, R_2, R_3) are esterified with the three hydroxyl groups of glycerol.

Physical and Chemical Properties

- Fats (and oils) may be divided into animal and vegetable fats according to source. Further, they may be classified according to their degree of unsaturation as measured by their ability to absorb iodine at the double bonds.
- This degree of unsaturation determines to a large extent the ultimate use of the fat.
- Liquid fats (i.e., vegetable and marine oils) have the highest degree of unsaturation, while solid fats (vegetable and animal fats) are highly saturated.
- Solid vegetable fats melting between 20 and 35 °C (68 and 95 °F) are found mainly in the kernels and seeds of tropical fruits.
- They have relatively low iodine values and consist of glycerides containing high percentages of such saturated acids as lauric, myristic, and palmitic.

- Fats are practically insoluble in water and, with the exception of castor oil, are insoluble in cold alcohol and only sparingly soluble in hot alcohol.
- They are soluble in ether, carbon disulfide, chloroform, carbon tetrachloride, petroleum benzin, and benzene. Fats have no distinct melting points or solidifying points because they are such complex mixtures of glycerides, each of which has a different melting point.
- Glycerides, further, have several polymorphic forms with different melting or transition points.
- Fats can be heated to between 200 and 250 °C (392 and 482 °F) without undergoing significant changes provided contact with air or oxygen is avoided.
- Above 300 °C (572 °F), fats may decompose, with the formation of acrolein (the decomposition product of glycerol), which imparts the characteristic pungent odour of burning fat.
- Hydrocarbons also may be formed at high temperatures.
- Fats are hydrolyzed readily.
- This property is used extensively in the manufacture of soaps and in the preparation of fatty acids for industrial applications.

Physical Properties of Fats

- The fats are insoluble in water, but readily soluble in ether, chloroform, benzene, carbon tetrachloride.
- They are readily soluble in hot alcohol but slightly soluble in cold.
- They are themselves good solvents for other fats, fatty acids, etc.
- They are tasteless, odourless, colorless and neutral in reaction,
- Several neutral fats are readily crystallized, eg, beef, mutton
- Their melting points are low.
- The specific gravity of solid fats is about 0.86. So the fat people float in water more readily than thin ones.

- They spread uniformly over the surface of water; so the spreading effect is to lower surface tension.

Chemical properties of fats

Hydrolysis

1. Hydrolysis of triacylglycerol takes place by lipases producing fatty acids and glycerol.
2. Phospholipases attack the ester linkage of phospholipids.

Saponification

- Boiling with an alcoholic solution of strong metallic alkali hydrolyzes triglycerides into glycerol and fatty acids are called saponification.
- The products are glycerol and the alkali salts of the fatty acids which are called soaps.
- Fats, phospholipids, glycolipids and waxes are called saponifiable lipid.
- Steroids, polyisoprenoids and higher alcohols are grouped as unsaponifiable lipids because they cannot give rise to soap.

Saponification number

- The number of milligrams of KOH is required to saponify 1 gram of fat or oil.
- The amount of alkali needed to saponify a given quantity of fat will be depended upon the number of -{:OOH group present. It is inversely proportional to the average molecular weight of the fatty acids in the fat i.e. the fats containing short chain fatty acids will have more -{:OOH groups per gram than long chain fatty acids and this will take up more alkali and hence will have higher saponification number.

Example: Butter containing a larger proportion of short chain fatty acids such as butyric and caproic acids, has relatively high saponification number 220 to 230.

Acid number

- The number of milligrams of KOH is required to neutralize the free fatty acids of 1 gram of fat.
- Significance: The acid number indicates the degree of rancidity of the given fat.

Iodine number

- This is the amount (in grams) of iodine absorbed by 100 grams of fat.
- This is the measure of the degree of unsaturation of a fat.
- 3. Significance: If the fat contains higher number of unsaturated fatty acids, it becomes essential for the protection of heart disease. These unsaturated fatty acids being combined with the cholesterol are oxidized in the liver producing bile acids, bile salts, Vitamin D, gonadotrophin hormones. They prevent atherosclerosis.

Acetyl number

- The number of milligrams of KOH required to neutralize the acetic acid obtained by saponification of 1 gram of fat after it has been acetylated.
- This is a measure of the number of hydroxy acid groups in the fat.

Polenske number

- The number of milliliters of 0.l (N) KOH required to neutralize the insoluble fatty acids from 5 grams of fat.

Reichert-Miessl number

- This is the same as the Polenske number except that the soluble fatty acids are measured by titration of the distillate obtained by steam distillation of the saponification mixture.
- Significance: It measures the amount of volatile soluble fatty acids.

Halogenation

- Chlorine, bromine and iodine atoms may be added to the double bonds of unsaturated fatty acids containing fats.

Rancidity

- Nearly all natural fats are oxidized when ex- posed to air, light, moisture, particularly, if warm, it develops an unpleasant odour and taste. The enzyme lipase which in the presence of moisture and temperature bring about hydrolysis rapidly.

- This happens so due to the formation of peroxides at the double bonds of unsaturated fatty acids.
- Vitamin E is an important natural antioxidant and prevents development of rancidity.

Soaps

- Soaps are metallic salts of fatty acids.
- Soaps are formed by adding alkalis to fatty acids.
- Soaps of unsaturated fatty acids are softer and more water soluble than those of saturated fatty acids.
- Potassium soap of an acid is more water soluble and softer than the sodium soap, calcium and magnesium soaps are far less soluble.

Compound lipids

Phospholipids

These are complex or compound lipids containing phosphoric acid, in addition to fatty acids, nitrogenous base and alcohol. Based on the type of alcohol present in the phospholipid they are classified into three types.

Glycerophosphatides - In this, glycerol is the alcohol group.

Example

- Phosphatidyl ethanolamine (cephalin).
- Phosphatidyl choline (Lecithin).
- Phosphatidyl serine.
- Plasmalogens.
- Phosphatidic acid.

Phosphoinositides - In this, inositol is the, alcohol.

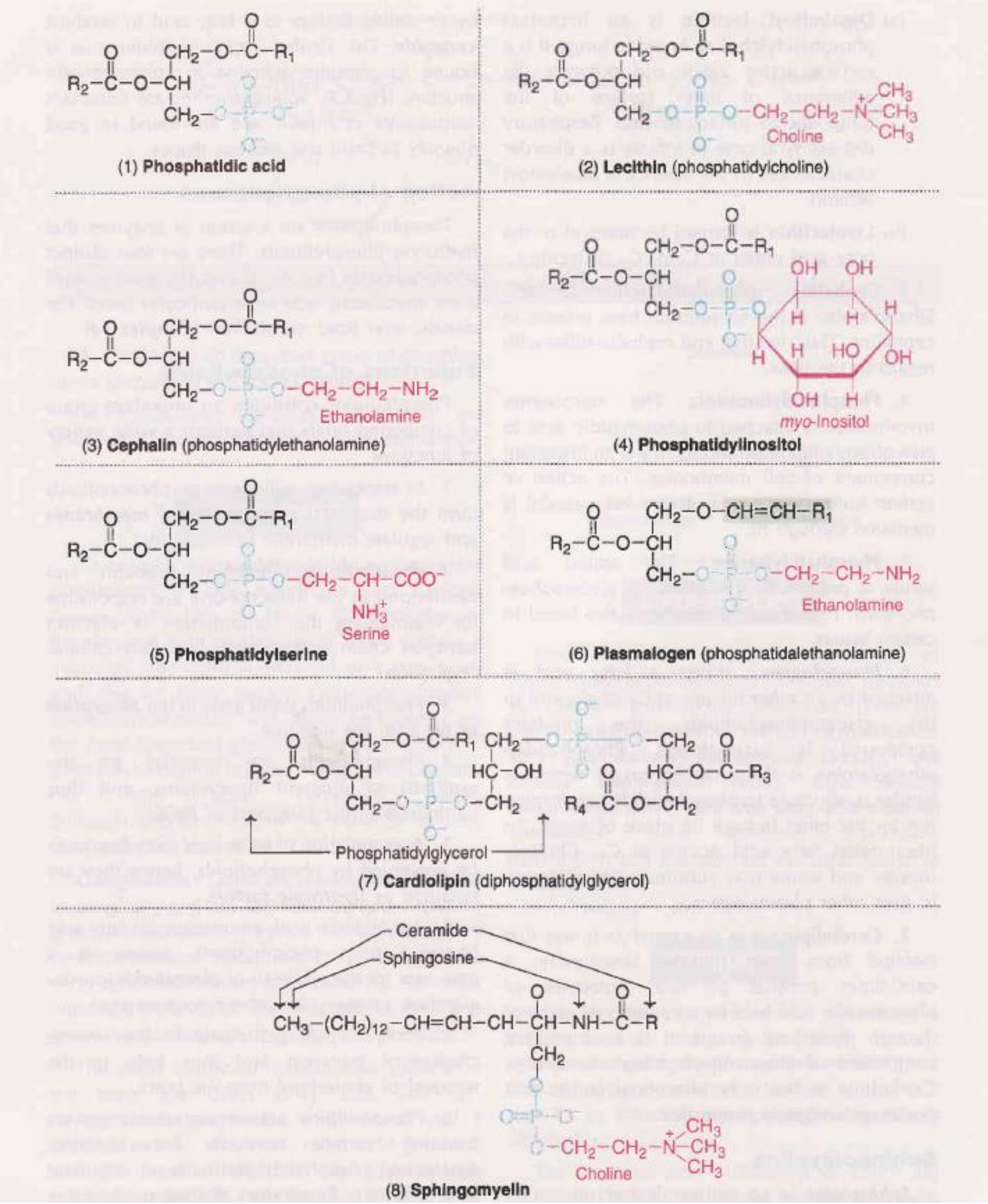
Example: Phosphatidyl inositol (Lipositol).

Phosphosphingosides - In this, sphingosine is an amino alcohol. .

Example: Sphingomyelin, ceramide.

Structure

- **Phosphatidic acid:** This is the simplest phospholipid. It does not occur in good concentration in the tissues.
- **Lecithins (phosphatidylcholine):** These are the most abundant group of phospholipids in the cell membranes.
- **Cephalins (phosphatidylethanolamine):** Ethanolamine is the nitrogenous base present in cephalins, thus lecithin and cephalin differ with regard to the base.
- **Phosphatidylinositol:** The steroisomer myo-inositol is attached to phosphatidic acid to give Phosphatidylinositol.
- **Phosphatidylserine:** The amino acid serine is present in this group of glycerophospholipids. Phosphatidylthreoninise also found in certain tissues.
- **Plasmalogens:** When a fatty acid is attached by an ether linkage at C1 of glycerol in the glycerophospholipids, the resultant compound is plasmalogen.
- **Cardiolipin**: It is so named as it was first isolated from heart muscle. Structurally, a cardiolipin consists of two molecules of phosphatidic acid held by an additional glycerol through phosphate groups.



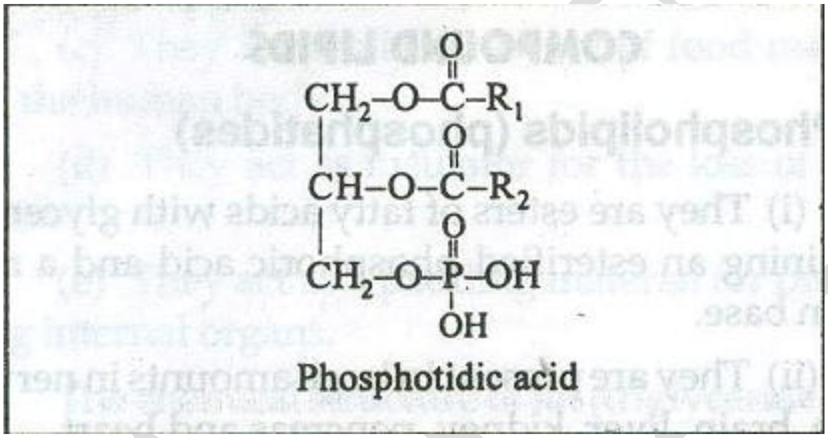
Structure of phospholipids

Phosphatidic acid and phosphat idyl glycerols

Phosphatidic acid is important as an intermediate in the synthesis of triacylglycerols and phospholipids.

Cardiolipin

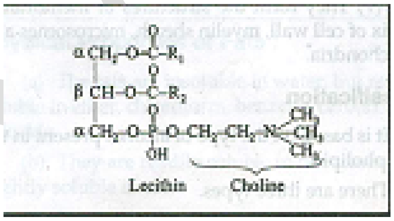
- It is formed from phosphatidyl glycerol.
- Chemically, it is diphosphatidyl glycerol.
- It is found in inner membrane of mitochondria and bacterial wall.



Phosphotidic acid

Lecithins (Phosphatidylcholine)

The lecithins contain glycerol and fatty acids, phosphoric acid and choline (nitrogenous base). Lecithins generally contain a saturated fatty acid at a position and an unsaturated fatty acid at β position. They can exist in aor β forms.



Physical Properties

- Lecithins are waxy, white substances but become brown soon when exposed to air.
- They are soluble in ordinary fat solvents except acetone.
- They decompose when heated.
- They constitute valuable agents for the emulsifications of fats and oils.

Chemical Properties of Lecithin

- When aqueous solution of lecithins is shaken with ~S04' choline is split off, forming phosphatidic acid.'
- When lecithins are boiled with alkalis or mineral acids, not only choline is split off; phosphatidic acid is further hydrolyzed to glycerophosphoric acid and 2 molecules of fatty acids.

Lecithin H_2SO4-+ Phosphatidic acid + cholin.

Phosphatidic acid -+ Glycerophosphoric acid + fatty acids (2 mol)'

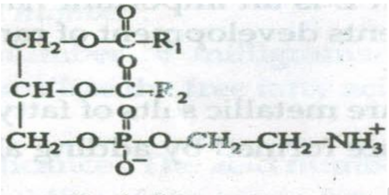
Physiological Functions of Lecithin

- It facilitates the combinations with proteins to from lipoproteins of plasma and cells.
- Acetylcholine formed from choline has an important role in the transmission of nervous impulses across synapses.
- Choline is the most important lipotropic agent as it can prevent formation of fatty liver.
- Lecithin lowers the surface tension of lung alveoli. Dipalmityllecithin is a major constituent of "lung surfactant" which prevents the adherence ql the inner surface of the alveoli of the lungs (preventing the collapse of the alveoli) by its surface tension lowering effect. The absence of this in the alveolar membrane of some premature infants causes the respiratory distress syndrome in them.
- It lowers the surface tension of water molecule and helps in the emulsification of fat.

Difference of Lecithin and Cephalin

Cadmium chloride compound of Cephalin is soluble but cadmium chloride compound of lecithin is insoluble.

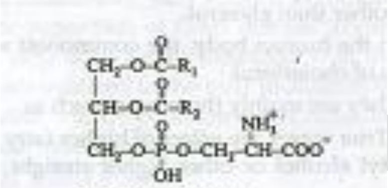
Cephalins (Phosphatidyl ethanplamine)



Cephalin

They always occur in the tissues in association with lecithins and are very similar in properties. The only difference is the nitrogenous base.

Phosphatidyl Serine

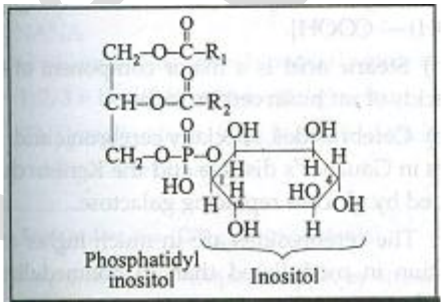


Phosphatidyl serine

A cephaline like phospholipid is found in tissues.

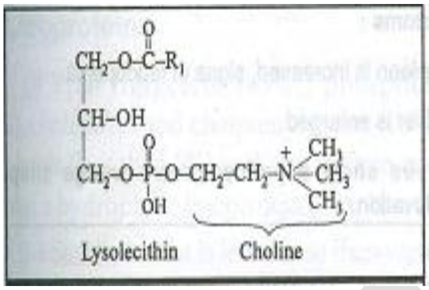
Phosphatidyl inositol (Lipositol or Phosphoinasitides)

Phosphatidyl~ inositol Inositol



- It acts as second messenger in Ca ++ de- pendent hormone action.
- Some signals must provide communication between the hormone receptor on the plasma membrane and intracellular Ca ++ reservoirs.
- They are more acidic than the other phospholipids.

Lysophospholipids:



- These are phosphoacylglycerols containing only one acyl radical in ex position eg, Lysolecithin.

Formation

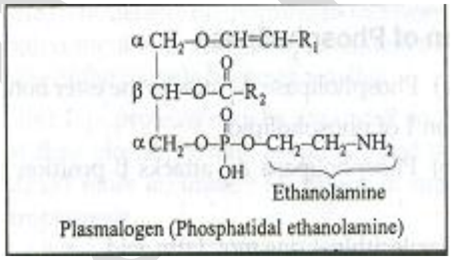
(a) By the action of phospholipase A_2.

(b) By interaction of lecithin and cholesterol in presence of the enzyme lecithin cholesterol acyl transferase, so lysolecithin and cholesterol ester are formed

Lecithin + cholesterol .t LCAT

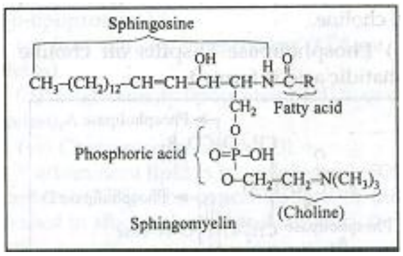
Lysolecithin + cholesterol ester.

Plasmalogens



- These are the contents of brain and muscle.
- Structurally, these resemble lecithins and cephalins but give a positive reaction when tested for aldehydes with Schiff's reagent (fuchsin sulfurous acid) after pretreatment of the phospholipid with mercuric chloride. Plasmalogen (phosphatidal ethanolamine)
- They possess an ether link in ex position instead of ester link. The alkyl radical is an unsaturated alcohol.

Sphingomyelins



- These are found in large quantities in brain and nerve tissue.
- The concentrations of these phospholipids are increased in Niemann-Pick disease in the liver and spleen.
- These contain sphingosine (18 carbon) (amino alcohol) fatty acid, phosphoric acid and choline. No glycerol is present.
- In sphingosine molecule -N~ group.· binds a fatty acid by an amide linkage to produce ceramide. When phosphate group is attached to ceramide it is called ceramide phosphate.
- When choline is split off from sphingomyelin, ceramide phosphate is left.

Clinical Aspect

- In Niemann-.Pick disease excess amount of sphingomyelin are deposited in brain, liver, spleen.
- It is a lipid storage disease (lipidoses) and hereditary. It is caused by the deficiency of enzyme sphingomyelinase.
- The clinical findings are:
  - (a) Enlarged liver and spleen.
  - (b) Mental retardation.
  - (c) Nervous system is affected.
  - (d) Anemia and leukocvtosis.

Action of Phospholipase

(a) Phospholipase A_1 attacks the ester bond in position 1 of phospholipid.

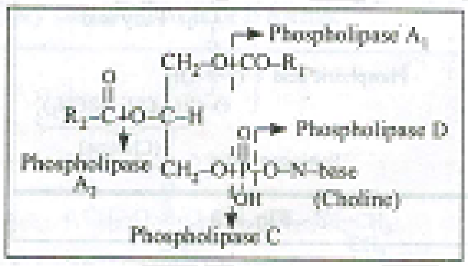
(b) Phospholipase A_2 attacks β position and form

Lysolecithin + one mol. fatty acid.

(c) Phospholipase B (lysophospholipase) attacks lysolecithin and hydrolyzes ester bond in a position and forms glyceryl phosphoryl choline + 1 mol fatty acid.

(d) Phospholipase C hydrolyzes phosphate ester bond and produces α, β diacyl glycerol + phosphoryl choline.

(e) Phospholipase D-splits off choline and phosphatidic acid is formed



Function of Phospholipids

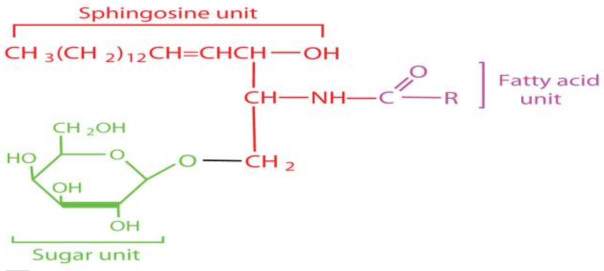
- Phospholipids constitute an important group of compound lipids that perform a wide variety of functions.
- In association with proteins, phospholipids form the structural components of membranes and regulate membrane permeability.
- Phospholipids (lecithin, cephalin and cardiolipin) in the mitochondria are responsible for maintaining the conformation of electron transport chain components, and thus cellular respiration.
- Phospholipids participate in the absorption of fat from the intestine.
- Phospholipids are essential for the synthesis of different lipoproteins, and thus participate in the transport of lipids.
- Accumulation of fat in liver (fatty liver) can be prevented by phospholipids, hence they are regarded as lipotropic factors.

- Arachidonic acid, an unsaturated fatty acid liberated from phospholipids, serves as a precursor for the synthesis of eicosanoids (prostaglandins, prostacyclinst, hromboxanes etc.,).
- Phospholipids participate in the reverse cholesterol transport and thus help in the removal of cholesterol from the body.
- Phospholipids act as surfactants (agents. lowering surface tension). For instance dipalmitoyl phosphatidylcholinies an important lung surfactant. Respiratory distress syndrome infants are associated with insufficient production of this surfactant.
- Cephalins, an important group of phospholipids participate in blood clotting.
- Phospholipids (phosphatidylinositol) are involved in signal transmission across membranes.

Glycolipids

Glycolipids are lipids with a carbohydrate attached. Their role is to provide energy and also serve as markers for cellular recognition. Eg: One type of glycolipid found in human red blood cells is involved in the ABO blood type antigens.

They contain an amino alcohol (Sphingosine) attached with an amide linkage to fatty acid and glycosidically to a carbohydrate moiety (Sugar, amino sugar, sialic acid).



Classification

They are classified into (i) Cerebrosides, (ii) Gangliosides.

Cerebrosides

- Cerebrosides contain galactose, a high molecular weight fatty acid and sphingosine. Therefore, they may also be classified as sphingolipids.

- They are the chief constituent of myelin sheath.
- They may be differentiated by the type of fatty acid in the molecule.

These are

- Kerasin-Containing lignoceric acid [CH_3 - (CH_2)22 - COOH].
- Cerebron-Containing a hydroxylignoceric acid (cerebronic acid).
- [CH3-(C~h1 - CH(OH) - COOH].
- Nervon-Containing an unsaturated homologue of lignoceric acid called nervonic acid. [CH_3 - {C~h- CH = CH - (CH_2)13 - COOH].
- Oxynervon-Containing hydroxynervonic acid [CH_3 - (CH_2)7 - CH = CH - (CH_2)12- CH(OH) - COOH].
- Stearic acid is a major component of the fatty acids of rat brain cerebrosides.
- Cerebrosides, specially cerebronic acid, increases in Gaucher's disease and the Kerasin characterized by glucose replacing galactose.
- The cerebrosides are in much higher concentration in medullated than in nonmedullated nerve fibers.

Clinical Aspect

Gaucher's disease

- The cerebroside content of the reticuloendothelial cell (spleen) is very high.
- In cerebroside molecule, the kerasin is characterised by glucose replacing galactose.
- The disease is caused by the deficiency of enzyme glucocerebrosidase.

Symptoms

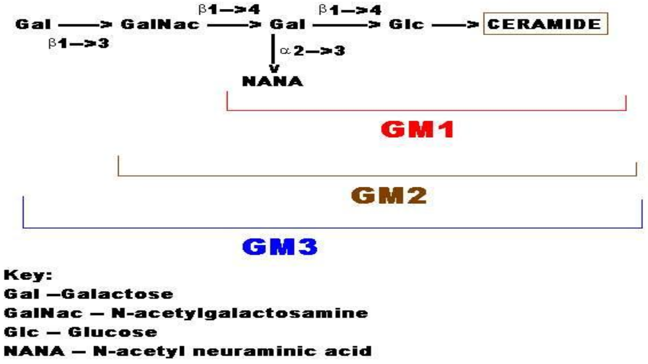
- Spleen is increased, signs of leucopenia.
- Liver is enlarged.
- Eyes show a yellow brown wedge shaped elevation.

Gangliosides

- These are glycolipids occurring in the brain.

- Gangliosides contain ceramide (sphingosine + fatty acids), glucose, galactose, N-acetylgalactosamine and sialic acid.
- Some gangliosides also contain dihydrosphingosine or Gangliosine in place of sphingosine.
- Most of the gangliosides contain a glucose, two molecules of galactose, one N-acetylgalactosamine and upto three molecules of sialic acid.

They are further classified into GM1, GM2, GM3. The following figure depict this.

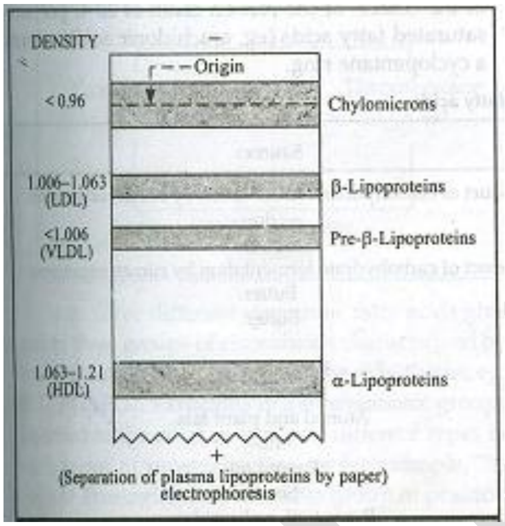


Lipo Proteins

Lipids are transported in blood as large macromolecules called lipoproteins. These are complexes with proteins. Free fatty acids are the exception, mainly binding to albumin.

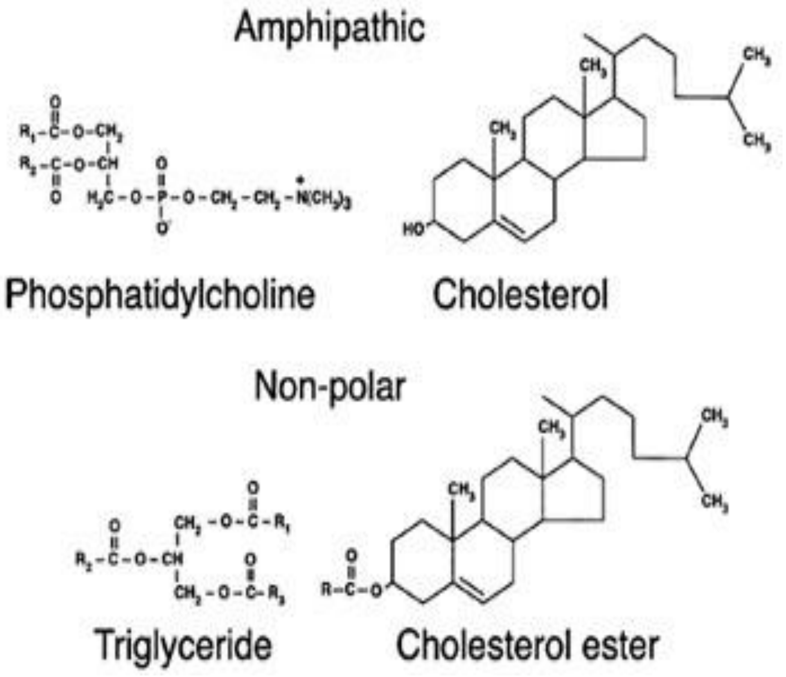
(i) Triacylglycerol (45%), phospholipids (35%), cholesterol and cholesteryl esters (15%), free fatty acids (less than 5%) and also protein combine to form a hydrophilic lipoprotein complex.

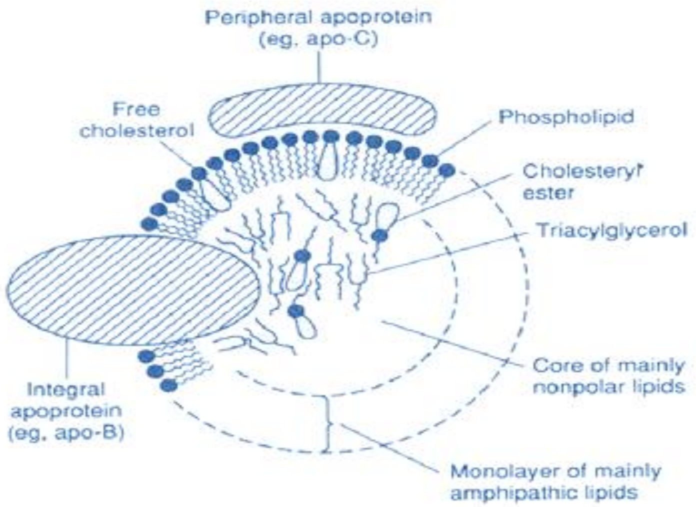
(ii) Since pure fat is less dense than Water, the proportion of lipid to protein in lipoproteins in plasma is separated by ultracentrifugation.



Structure of lipo protein

Hydrophobic lipids, triglycerides and phospholipids are within the lipoprotein core, with the polar portions of phospholipids and the water-soluble alcohol portion of free cholesterol projecting into the aqueous environment, causing solubilization of the lipoprotein.





Types of lipoproteins

Lipoprotein classes can be separated physico chemically, either by electrophoresis which uses surface charge or by ultracentrifugation which uses relative density. Four major groups of lipoproteins have been identified which are important physiologically and in clinical diagnosis in some metabolic disorders of fat metabolism.

(i) chylomicrons,
(ii) Very-low-density lipoprotein (VLDL),
(iii) Intermediate-density lipoprotein (IDL),
(iv) Low-density lipoprotein (LDL)
(v) High-density lipoprotein (HDL).

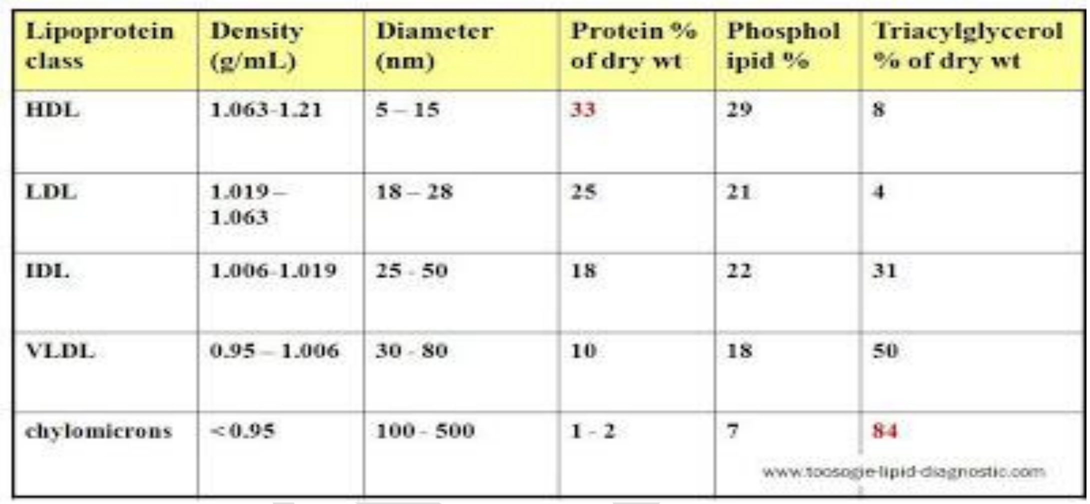
Predominant lipid is triacylglycerol (50%) and cholesterol (23%). The concentrations of these are increased in atherosclerosis and coronary thrombosis etc.

LDL: Predominant lipid is cholesterol (46%) and phospholipids (23%).Increase in atherosclerosis and coronary thrombosis, etc.

HDL: Predominant lipid is phospholipid (27%) and proteins (45%). The protein moiety lipoprotein is known as an apo protein which constitute nearly 60% of some HDL and 1% of chylomicrons. Many lipoproteins contain more than one type of apoprotein polypeptide.

The larger lipoproteins (such as chylomicrons and VLDL) consist of a lipid core of non-polar triacylglycerol and cholesteryl ester surrounded by more polar phospholipid, cholesterol and apoproteins.

The table gives the properties of different lipo proteins



Lipoprotein class	Density (g/mL)	Diameter (nm)	Protein % of dry wt	Phospholipid %	Triacylglycerol % of dry wt
HDL	1.063-1.21	5 – 15	33	29	8
LDL	1.019 – 1.063	18 – 28	25	21	4
IDL	1.006-1.019	25 - 50	18	22	31
VLDL	0.95 – 1.006	30 - 80	10	18	50
chylomicrons	<0.95	100 - 500	1 - 2	7	84

www.toosogie-lipid-diagnostic.com

Importance

- To transport and deliver the lipids to tissues.
- To maintain structural integrity of cell surface and sub cellular particles like mitochondria and microsomes.
- The β-lipoprotein fraction increases in severe diabetes mellitus, atherosclerosis etc. Hence determination of the relative concentrations of α and β-lipoproteins and pre-β- lipoproteins are of diagnostic importance.

Aminolipids

Phosphatidyl ethanolamine and serines are aminolipids and sphingomyelins and gangliosides contain substituted amino groups.

Sulpholipids (Sulphatides)

- These have been isolated from brain and other animal tissues.

- These are sulphate derivatives of the a1actosyl residue in cerebrosides.

Derived Lipids

Fatty Acids

- In chemistry, especially biochemistry, a fatty acid is a carboxylic acid with a long aliphatic tail (chain), which is either saturated or unsaturated.
- Most naturally occurring fatty acids have a chain of an even number of carbon atoms, because they are synthesized from 2-carbon units and are straight chain derivatives (from 4 to 28.)
- These are obtained by the hydrolysis of fats. Fatty acids are usually derived from triglycerides or phospholipids.
- When they are not attached to other molecules, they are known as "free" fatty acids.
- The straight chain may be saturated (containing no double bonds) or unsaturated (containing one or more double bonds).
- Carbon atoms of fatty acids are numbered from the carboxyl carbon (carbon No.1). The carbon atom adjacent to the carboxyl carbon (Carbon No. 2) is also known as the α-carbon. Carbon atom No. 3 is the β-carbon and the end methyl carbon is known as the γ-carbon.
- Various conventions are used for indicating the number and position of the double bonds, eg, Δ^9 indicates a double bond between carbon atoms 9 and 10 of the fatty acid.

Functions

- Fatty acids are important sources of fuel because, metabolized, they yield large quantities of ATP.
- Many cell types can use either glucose or fatty acids for this purpose.
- In particular, heart and skeletal muscle prefer fatty acids.
- The brain cannot use fatty acids as a source of fuel; it relies on glucose or ketone bodies

Types

1. Straight chain.
2. Branched chain.
3. Substituted (methyl substituted -cerebronic acid)
4. Cyclic (chaulmoogric acid) used in leprosy.

Straight chain

(a) Saturated {odd (less than 10 carbon atom)} & even (greater than 10 carbon atom)}.

(b) Unsaturated (odd & even). (Straight chain even number fatty acid is common)

Saturated Fatty Acids

General formula for saturated fatty acids is CnH2n+l COOE. Other higher fatty acids occur in waxes. A few branched-chain fatty acids have also been isolated from both plant and animal sources. Prostanoids include Prostaglandins (PC), and thromboxanes (TX).

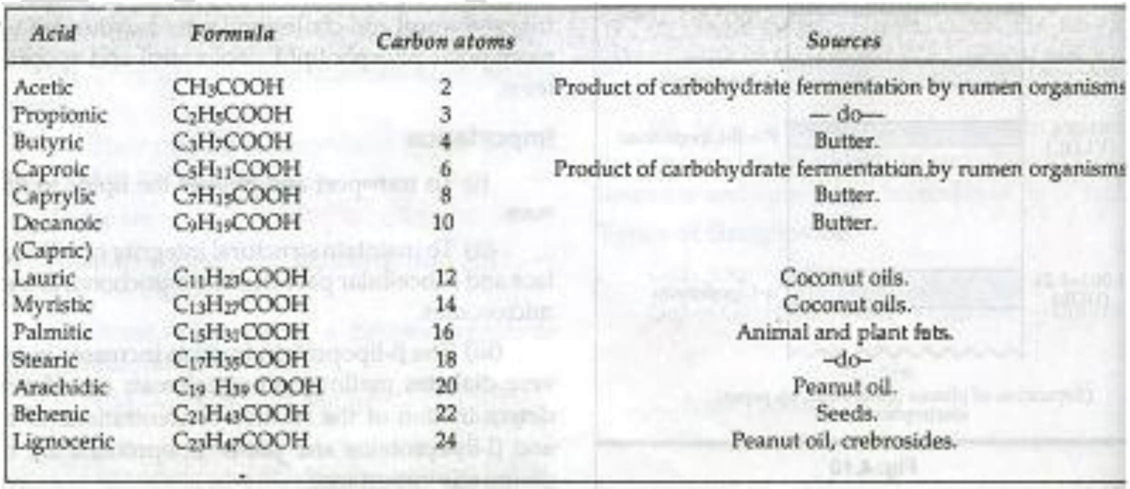
General characteristics of prostanoid

(a) All are 20 carbon compound.

(b) Trans double bond at 13 position.

(c) -OH group at 15 position.

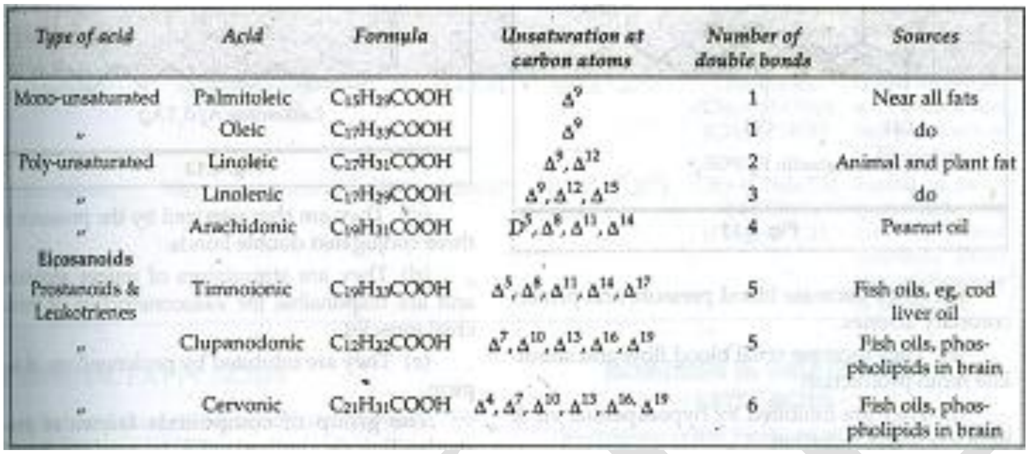
Saturated Fatty Acids



Acid	*Formula*	*Carbon atoms*	*Sources*
Acetic	CH_3COOH	2	Product of carbohydrate fermentation by rumen organisms
Propionic	C_2H_5COOH	3	— do—
Butyric	C_3H_7COOH	4	Butter.
Caproic	$C_5H_{11}COOH$	6	Product of carbohydrate fermentation.by rumen organisms
Caprylic	$C_7H_{15}COOH$	8	Butter.
Decanoic (Capric)	$C_9H_{19}COOH$	10	Butter.
Lauric	$C_{11}H_{23}COOH$	12	Coconut oils.
Myristic	$C_{13}H_{27}COOH$	14	Coconut oils.
Palmitic	$C_{15}H_{31}COOH$	16	Animal and plant fats.
Stearic	$C_{17}H_{35}COOH$	18	-do-
Arachidic	$C_{19} H_{39} COOH$	20	Peanut oil.
Behenic	$C_{21}H_{43}COOH$	22	Seeds.
Lignoceric	$C_{23}H_{47}COOH$	24	Peanut oil, crebrosides.

Unsaturated fatty acids

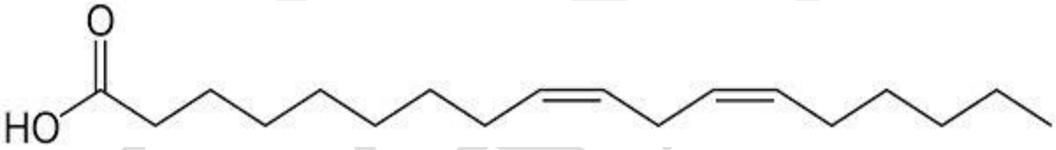
A.General formula C_nH2n-l COOH



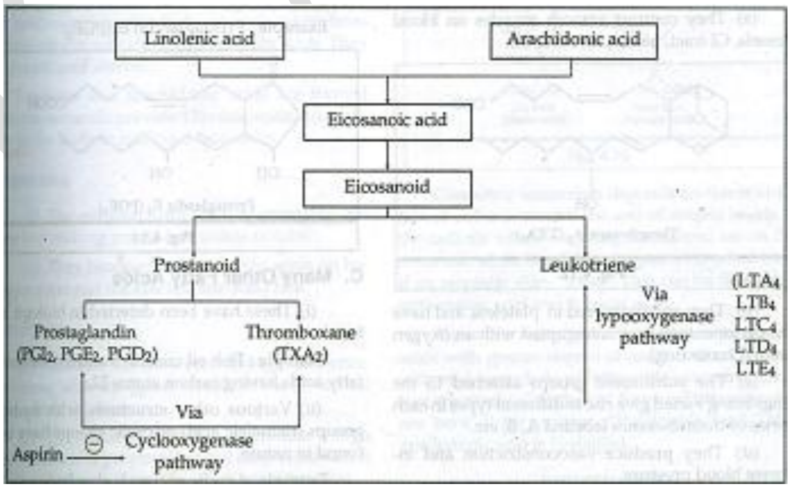
Type of acid	Acid	Formula	Unsaturation at carbon atoms	Number of double bonds	Sources
Mono-unsaturated	Palmitoleic	$C_{15}H_{29}COOH$	Δ^9	1	Near all fats
"	Oleic	$C_{17}H_{33}COOH$	Δ^9	1	do
Poly-unsaturated	Linoleic	$C_{27}H_{31}COOH$	Δ^9, Δ^{12}	2	Animal and plant fat
"	Linolenic	$C_{17}H_{29}COOH$	$\Delta^9, \Delta^{12}, \Delta^{15}$	3	do
"	Arachidonic	$C_{19}H_{31}COOH$	$D^5, \Delta^8, \Delta^{11}, \Delta^{14}$	4	Peanut oil
Eicosanoids Prostanoids & Leukotrienes	Timnodonic	$C_{19}H_{29}COOH$	$\Delta^5, \Delta^8, \Delta^{11}, \Delta^{14}, \Delta^{17}$	5	Fish oils, eg. cod liver oil
"	Clupanodonic	$C_{12}H_{32}COOH$	$\Delta^7, \Delta^{10}, \Delta^{13}, \Delta^{16}, \Delta^{19}$	5	Fish oils, phospholipids in brain
"	Cervonic	$C_{21}H_{31}COOH$	$\Delta^4, \Delta^7, \Delta^{10}, \Delta^{13}, \Delta^{16}, \Delta^{19}$	6	Fish oils, phospholipids in brain

- Fatty acids with one double bond are monounsaturated and those with 2 or more double bonds are collectively known as polyunsaturated fatty acids (PUFA).

  Eg: Linoleic acid



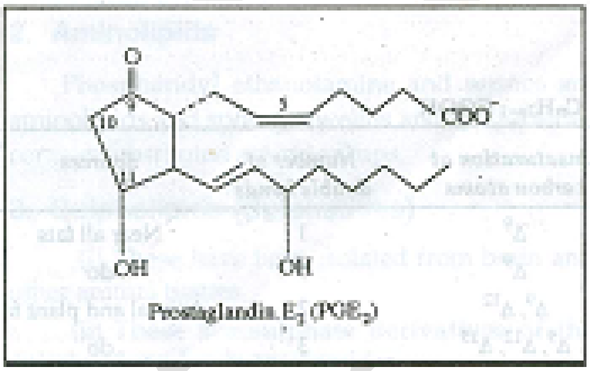
Classification



Three different eicosanoic fatty acids give rise to three groups of eicosanoids characterized by the number of double bonds in the side chains, eg, PG_1, PG_2, PG_3. Variations in the substituent groups attached to the rings give rise to different types in each series of prostaglandins, as for example, "E" type of Prostaglandin has a keto group in position9,whereas the "F" type has a hydroxyl group in this position,

Prostacycllns (PGI)

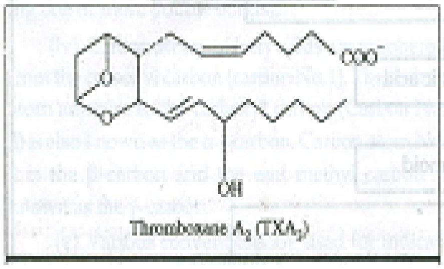
- They are formed in vascular endothelium and continually formed in heart. They are also formed in kidneys.
- They are formed from cyclic endoperoxide PGH_2 by the action of microsomal Prostacyclin synthetase.
- They inhibit platelet aggregation and gastric secretion from the pyloric mucosa.



Prostaglandin E_2 (PGE_2)

- They decrease blood pressure and protect coronary arteries.
- They increase renal blood flow and stimulate renin production.
- They are inhibited by hyperlipemia, vit. E deficiency and radiation.

Thromboxanes

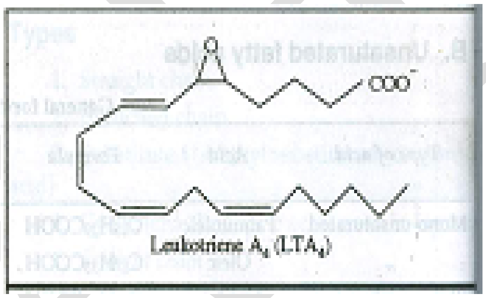


Thromboxane A_2 (TXA_2)

- They contract smooth muscles on blood vessels, GI tract, uterus, bronchioles.
- They are discovered in platelets, and have the cyclopentane ring interrupted with an oxygen atom (Oxane ring).
- The substituent groups attached to the rings being varied give rise to different types in each series of thromboxanes labelled A, B, etc.
- They produce vasoconstriction and in- crease blood pressure.
- They cause release of serotonin and calcium ion (Ca ++) from platelet granules.
- lmidazoles inhibit their synthesis.

Leukotrienes

- They are the third group of eicosanoid derivatives formed via the lipoxygenase pathway rather than cyclization of the fatty acid chain.

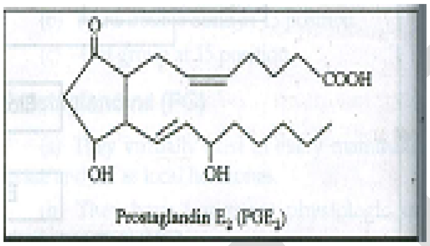


- They are first described in leukocytes.
- They are characterized by the presence of three conjugated double bonds.
- They are stimulators of mucus secretion and are responsible for vasoconstriction of bronchial muscles.
- They are inhibited by prolonged use of aspirin.
- The groups of compounds known as prostaglandins are synthesized from arachidonic acid in the body. They have pharmacologic and biochemical activity.

Prostaglandins (PG)

- They virtually exist in every mammalian tissue and act as local hormones.
- They have important physiologic anc pharmacologic activities.

- They are synthesized in vivo by cyclization of the center of the carbon chain of 20C polyunsaturated fatty acids (eg, arachidonic acid) to fom a cyclopentane ring.
- **Example:** Prostaglandin E_2(PGE_2)



Prostaglandin E_2 (PGE_2)

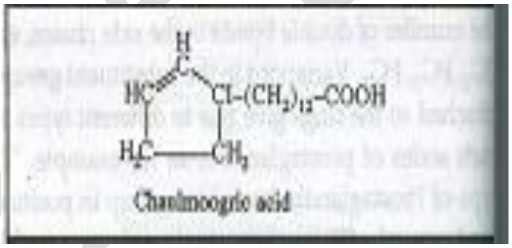
Many Other Fatty Acids

- These have been detected in biologic material.

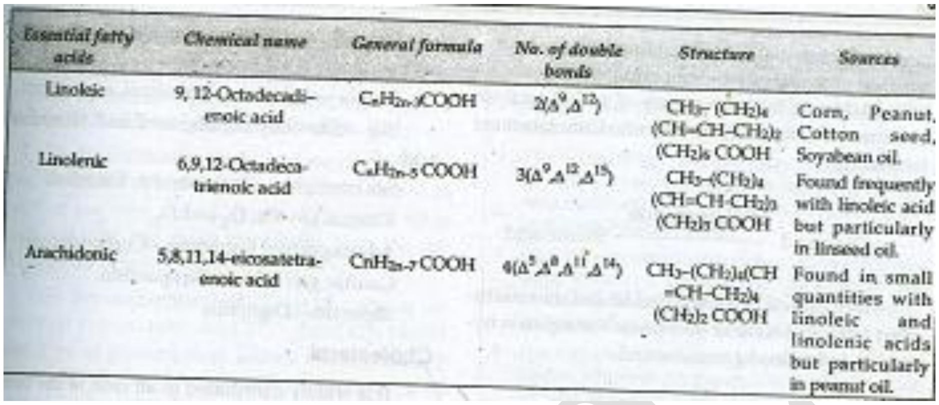
Example: Fish oil contain 5 and. 6 unsaturated fatty acids having carbon atoms 22.

- Various other structures with hydroxy groups (ricinoleic acid) or cyclic groups have been found in nature. .

Example of cyclic groups is chaulmoogric acid which was used many years ago in the treatment of leprosy.



Chaulmoogric acid



Essential fatty acids	Chemical name	General formula	No. of double bonds	Structure	Sources
Linoleic	9, 12-Octadecadi-enoic acid	$C_nH_{2n-3}COOH$	$2(\Delta^9,\Delta^{12})$	$CH_3-(CH_2)_4(CH=CH-CH_2)_2(CH_2)_6COOH$	Corn, Peanut, Cotton seed, Soyabean oil.
Linolenic	6,9,12-Octadeca-trienoic acid	$C_nH_{2n-5}COOH$	$3(\Delta^9,\Delta^{12},\Delta^{15})$	$CH_3-(CH_2)_4(CH=CH-CH_2)_3(CH_2)_3COOH$	Found frequently with linoleic acid but particularly in linseed oil.
Arachidonic	5,8,11,14-eicosatetra-enoic acid	$CnH_{2n-7}COOH$	$4(\Delta^5,\Delta^8,\Delta^{11},\Delta^{14})$	$CH_3-(CH_2)_4(CH=CH-CH_2)_4(CH_2)_2COOH$	Found in small quantities with linoleic and linolenic acids but particularly in peanut oil.

Essential Fatty Acids

Burr and Burr (1930) introduced the term "Essential Fatty Acids" (EF A) on the basis that they are essential for the growth and health of young albino rats. These polyunsaturated fatty acids which are not synthesized in the body but are taken from natural sources are called essential fatty acids. They are (mentioned above): linolenic and arachidonic acids are formed from linoleic acids provided linoleic acids are available in the body in sufficient quantities.

Properties

- The essential fatty acids of vegetable oils have low melting points and iodine number.
- They become saturated fatty acids on hydrogenation and the oils become solid fats.

Functions

- The essential fatty acids in high concentration along with the lipids constitute the structural elements of the tissues.
- The lipids of gonads also contain a high concentration of polyunsaturated fatty acids which suggest the importance of reproductive function.
- They effect the prolongation of clotting time and increase the fibrinolytic activity.
- They retard atherosclerosis being esterified and emulsified with cholesterol and are incorporated into lipoproteins for transport to the liver for further oxidation.
- They cure skin lesions.

- The deficiency of these acids in the diet of babies causes eczema.

Isomerim in Unsaturated Fatty Acids

Variations in the locations of the double bond in unsaturated fatty acid chains produce isomers. Oleic acid has 15 different positional isomers.

Geometric isomerism depends on the orientation of radicals around the axis of double bonds. If the radicals which are being considered are on the same side of the bond, the compound is called "cis", if on opposite side, "trans". This can be illustrated with maleic acid and fumaric acid.

There are more geometric isomers in case of acids with greater degree of unsaturation. The un- saturated long chain of fatty acids occurring in nature are nearly all in tbe 'cis' form and the molecules are "bent" at the position of the double bond. Thus, arachidonic acid is D-shaped.

Refined and Hydrogenated Oils

Refined oil: It is prepared in the following manner:

- Free fatty acids are removed by alkali treatment
- Colouring matter is removed by activated carbon.
- Odour is '~ov"" by superheated steam,

Essential fatty acids, or EFAs, are fatty acids that humans and other animals must ingest because the body requires them for good health but cannot synthesize them. The term "essential fatty acid" refers to fatty acids required for biological processes but does not include the fats that only act as fuel. Only two fatty acids are known to be essential for humans: alpha-linolenic acid (anomega-3 fatty acid) and linoleic acid (an omega-6 fatty acid).

Some other fatty acids are sometimes classified as "conditionally essential," meaning that they can become essential under some developmental or disease conditions; examples include docosahexaenoic acid (an omega-3 fatty acid) and gamma-linolenic acid(an omega-6 fatty acid).

The essential fatty acids start with the short chain polyunsaturated fatty acids (SC-PUFA):

- ω-3 fatty acids:
- α-Linolenic acid or ALA (18:3n3)
- ω-6 fatty acids:
- Linoleic acid or LA (18:2n-6)

These two fatty acids cannot be synthesized by humans because humans lack the desaturase enzymes required for their production.

They form the starting point for the creation of longer and more desaturated fatty acids, which are also referred to as long-chain polyunsaturated fatty acids (LC-PUFA):

ω-3 fatty acids

eicosapentaenoic acid or EPA (20:5n-3)
docosahexaenoic acid or DHA (22:6n-3)

ω-6 fatty acids

gamma-linolenic acid or GLA (18:3n-6)
dihomo-gamma-linolenic acid or DGLA (20:3n-6)
arachidonic acid or AA (20:4n-6)

ω-9 fatty acids are not essential in humans because they can be synthesized from carbohydrates or other fatty acids.

Functions of Essential Fatty Acids

Essential fatty acids have a ton of benefits in our body. They are

- They help with cellular development and the formation of healthy cell membranes, and they have actually been shown to block tumor formation in animals, as well as block the growth of human breast cancer cells.
- Essential fatty acids assist in the development and function of the brain and nervous system.
- Helps to regulate proper thyroid and adrenal activity.
- They play a role in thinning blood, which can prevent blood clots that lead to heart attacks and stroke.

- They also possess natural anti-inflammatory qualities that can relieve symptoms of both arthritis and other autoimmune system diseases.
- Essential fatty acids regulate blood pressure, immune responses and liver function, as well as help with blood clotting and breaking down cholesterol.
- Diet low in these fatty acids has been shown to create skin problems, including eczema, dandruff, split nails and brittle hair.

Hydrogenated oils

The refined oils are hydrogenated under optimum temperature and pressure with hydrogen in the" presence of nickel catalyst. . Unsaturated fatty acids are converted into saturated fatty' acids.

Hydrogenation
Oleic acid-------------------------Stearic acid

The liquid oil becomes solid fat and the unsaturated fatty acid content decreases. Vanaspati is hydrogenated refined groundnut oil.

Alcohols

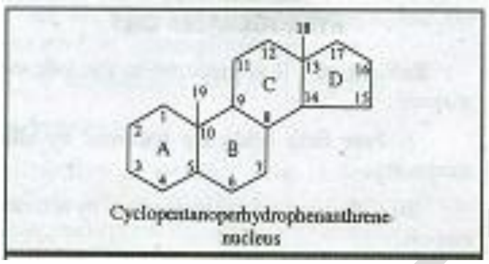
Alcohols found in lipid molecules include glycerol, cholesterol and higher alcohols (cetyl alcohol), usually found in the waxes.

The unsaturated alcohols are important pigments. Phytyl alcohol is a constituent of chlorophyll and lycophyll ($C_{40}H_{56}O_2$); a polyunsaturated dihydroxy alcohol occurs in tomatoes as a purple pigment.

Steroids

The steroids are often found in association with fat. They have a similar cyclic nucleus resembling phenanthrene (rings A, B, C) to which a cyclopentane ring (D) is attached. The parent substance is better designated as cyclopentano-perydrophen anthrene. The positions on the steroid nucleus are numbered as shown in the figure.

Methyl side chains occur typically at positions 10 and 13 (constituting C atoms 19 and 18). A side chain at position 17 is usual (as in cholesterol). *H* the compound has one or more hydroxyl groups and no carbonyl or carboxyl groups, it is a *sterol,* and the name terminates in-OL.



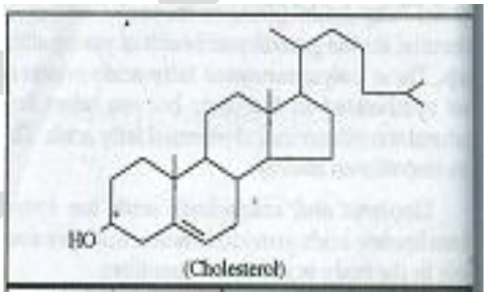
Cyclopentanope Rhydrophenanthrene. nucleus

Steroids may be divided in the following manner:

- Sterols=cholesterol, ergosterol, coprosterol.
- Bile acids-Glycocholic acid and taurocholic acid.
- Sex hormones-Testosterone, Estradiol.
- Vitamin D-Vit. 02 and 03 .
- Adrenocortical hormones-Corticosterone.
- Cardiac glycosides-Stropanthin.
- Saponins-Digitonin.

Cholesterol

It is widely distributed in all cells of the body. It occurs in animal fats but not in plant fats. Its structure is given below. The metabolism of cholesterol is discussed in the chapter of lipid metabolism



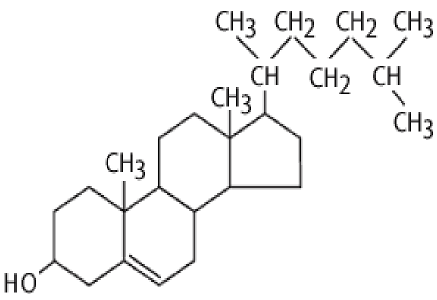
(Cholesterol)

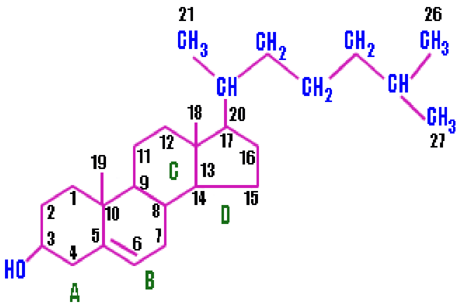
Cholesterol and significance

Cholesterol

- Cholesterol, exclusively found in animals, is the most abundant animal sterol.

- It is widely distributed in all cells and is a major component of cell membranes and lipoproteins.
- Cholesterol (Creek: chole-bile) was first isolated from bile.
- Cholesterol literally means 'solid alcohol from bile.'





Functions of Cholesterol

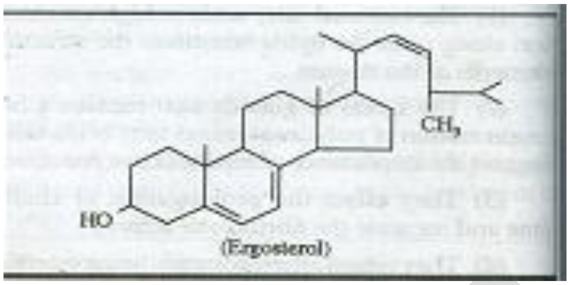
- Cholesterol is a poor conductor of heat and electricity, since it has a high dielectric constant.
- It is present in abundance in nervous tissues.
- It appears that cholesterol functions as an insulating cover for the transmission of electrical impulses in the nervous tissue.
- Cholesterol performs several other biochemical functions which include its role in membrane structure and function, in the synthesis of bile acids, hormones (sex and cortical) and vitamin D.

Erogesrol

(i) It occurs in ergot and yeast.

(ii) It is the precursor of vitamin 0.

(iii) It acquires antirachitic properties with the opening of ring B when irradiated with ultraviolet light.



(Ergosterol)

Coprosterol

It occurs in feces as a result of the reduction by bacteria in the intestine of the double bond between C_5, and C_6 of cholesterol.

Important Tests

(1) Greese spot test: A drop of oil placed over a piece of ordinary paper. A translucent spot is visible. This indicates the presence of fat.

(2) Emulsification test: 2 ml water is taken in one test tube and 2 ml of diluted bile salt solution in another test tube. Add 3 drops of the given oil to each test tube and shake vigorously. Note the stability of the emulsification formed.

(3) Saponification test: Take 10 drops of coconut oil in a test tube. Add 20 drops of 40% NaOH and 2 ml of glycerol to it. Gently boil for about 3 minutes until complete saponification occurs. If oil globules are visible, boiling must be continued. Divide the solution into 3 parts to carry the following experiments in test tube 1, 2, 3.

To test tube No. 1 add saturated solution of NaCl. Note that the soap separates out and floats to the surface (salting out process).

To test tube No.2 add a few drops of cone. HCl. An oily layer of the fatty acids rises to the surface.

To test tube No.3 add a few drops of $CaCl_2$ solution. The insoluble calcium soap is precipitated. . .

Unsaturation test

Add 10 drops of Huble's iodine reagent to 10 rnl of chloroform. The chloroform assumes a pink colour due to the free iodine. The solution is divided equally into three test tubes as (a), (b) and (c) and three types of oil are added.

Add the oil No. 1 to the test tube (a) drop by drop shaking the tube vigorously after each addition till the pink colour of the solution just disappears. The number of oil drops required are noted. The experiment is repeated by oil 2 and 3 adding to test tubes (b) and (c), respectively. The more the number of drops required to discharge the pink colour, the less is the unsaturation.

Colour Reactions to Detect Sterols

Liebermann-Burchard Reaction: A chloroform solution of a sterol when treated with acetic anhydride and sulphuric acid gives a green colour .This reaction is the basis of a colorimetric estimation of blood cholesterol.

Salkowski test: A red to purple colour appears when a chloroform solution 0; the sterol is treated with an equal volume of concentrated sulphuric acid. .

Clinical Orientation

- The high concentration of polyunsaturated fatty acids in the lipids of gonads are important in reproductive function. .
- The essential fatty acid deficiency causes swelling of mitochondrial membrane resulting. in the reduction in efficiency of oxidative phosphorylation producing increased heat.
- Docosahexenoic acid formed from dietary linolenic acids enhances the electrical response of the photoreceptors to illumination. Therefore; linolenic acid of the diet is essential for optimal vision.
- The deficiency of essential fatty acids causes skin lesions, abnormal pregnancy and lactation in adult females, fatty liver, kidney damage.
- The genetic deficiency of lecithin cholesterol acyl transferase (LCAT) .causes Norum's Disease.
- Sitosterol decreases the intestinal absorption of exogenous and endogenous cholesterol and thereby lowers the blood cholesterol level.
- The deficiency of the enzyme sphingomyelinase. Causes the large accumulations of

  sphingomyelins in brain, liver and spleen of children resulting in the Niemann-Pick disease with the symptoms of enlarged abdomen, liver, spleen and mental deterioration.

  - Absence of dipalmityl lecithin (DPL} in premature foetus produces respiratory distress syndrome (Hyaline-membrane disease).
  - The inherited Gaucher's Disease in infancy and childhood is caused by the deficiency of the enzyme glucocerebrosldase involving the large accumulations of glucocereqrosides (usually Kerasin) in the liver, spleen, bone marrow, and brain with the manifestations of weight loss, failure in growth, and progressive mental retardation.
  - The autosomal recessive Tay-Sach's Disease (GM_2 Gangliosidosis) results in the accumulation of large amounts of gangliosides in the brain and nervous tissues due to the absence of the enzyme hexosaminidase A with the association of progressive development of idiocy and blindness in infants soon after birth.
  - The inherited disorder Metachromatic Leukodystrophy (MLD) happens on the salfatide,

    formed from galactocerebroside, accumulation in various tissues owing to the deficiency of the enzyme sulfatase (Aryl salfatase) with the symptoms of weakness, ataxia, defects in locomotion, paralysis, difficulties in speech in children before three years of age and psychiatric manifestation including progressive dementia in adults.

- Obesity and atherosclerosis are distinctly related to the concentrations of cholesterol and polyunsaturated fatty acids in the body.

Karpagam Academy of Higher Education
Department of Biochemistry
I B.Sc., Microbiology
18MBU103 - Biochemistry

Question number	**Unit**	**Question**	Option I	Option II	Option III	Option IV	**Answer**
1	3	An example of a hydroxy fatty acid is	Ricinoleic acid	Crotonic acid	Butyric acid	Oleic acid	**Ricinoleic acid**
2	3	An example of a saturated fatty acid is	Palmitic acid	Oleic acid	Linoleic acid	Erucic acid	**Palmitic acid**
3	3	If the fatty acid is esterified with an alcohol of high molecular weight instead of glycerol, the resulting compound is	Lipositol	Plasmalogen	Wax	Cephalin	**Wax**
4	3	A fatty acid which is not synthesized in the body and has to be supplied in the diet is	Palmitic acid	Lauric acid	Linolenic acid	Palmitoleic acid	**Linolenic acid**
5	3	Essential fatty acid:	Linoleic acid	Linolenic acid	Arachidonic acid	All these	**All these**
6	3	The fatty acid present in cerebrosides is	Lignoceric acid	Valeric acid	Caprylic acid	Behenic acid	**Lignoceric acid**
7	3	The number of double bonds in arachidonic acid is	1	2	4	6	**4**
8	3	In humans, a dietary essential fatty acid is	Palmitic acid	Stearic acid	Oleic acid	Linoleic acid	**Linoleic acid**
9	3	A lipid containing alcoholic amine residue is	Phosphatidic acid	Ganglioside	Glucocerebroside	Sphingomyelin	**Sphingomyelin**

10	3	Cephalin consists of	Glycerol, fatty acids, phosphoric acid and choline	Glycerol, fatty acids, phosphoric acid and ethanolamine	Glycerol, fatty acids, phosphoric acid and inositol	Glycerol, fatty acids, phosphoric acid and serine	**Glycerol, fatty acids, phosphoric acid and ethanolamine**
11	3	In mammals, the major fat in adipose tissues is	Phospholipid	Cholesterol	Sphingolipids	Triacylglycerol	**Triacylglycerol**
12	3	Glycosphingolipids are a combination of	Ceramide with one or more sugar residues	Glycerol with galactose	Sphingosine with galactose	Sphingosine with phosphoric acid	**Ceramide with one or more sugar residues**
13	3	The importance of phospholipids as constituent of cell membrane is because they possess	Fatty acids	Both polar and nonpolar groups	Glycerol	Phosphoric acid	**Both polar and nonpolar groups**
14	3	In neutral fats, the unsaponificable matter includes	Hydrocarbons	Triacylglycerol	Phospholipids	Cholsesterol	**Hydrocarbons**
15	3	Higher alcohol present in waxes is	Benzyl	Methyl	Ethyl	Cetyl	**Cetyl**
16	3	Kerasin consists of	Nervonic acid	Lignoceric acid	Cervonic acid	Clupanodonic acid	**Lignoceric acid**
17	3	Gangliosides are complex glycosphingolipids found in	Liver	Brain	Kidney	Muscle	**Brain**
18	3	Unsaturated fatty acid found in the cod liver oil and containing 5 double bonds is	Clupanodonic acid	Cervonic acid	Elaidic acid	Timnodonic acid	**Timnodonic acid**
19	3	Phospholipid acting as surfactant is	Cephalin	Phosphatidyl inositol	Lecithin	Phosphatidyl serine	**Lecithin**
20	3	An oil which contains cyclic fatty acids and once used in the treatment of leprosy is	Elaidic oil	Rapeseed oil	Lanoline	Chaulmoogric oil	**Chaulmoogric oil**
21	3	Unpleasant odours and taste in a fat (rancidity	Lead	Copper	Tocopherol	Ergosterol	**Tocopherol**
22	3	Gangliosides derived from glucosylceramide contain in addition one or more molecules of	Sialic acid	Glycerol	Diacylglycerol	Hyaluronic acid	**Sialic acid**
23	3	’Drying oil’, oxidized spontaneously by atmospheric oxygen at ordinary	Coconut oil	Peanut oil	Rape seed oil	Linseed oil	**Linseed oil**

		temperature and forms a hard water proof material is					
24	3	Deterioration of food (rancidity	Cholesterol	Vitamin E	Peroxidation of lipids	Phenolic compounds	**Peroxidation of lipids**
25	3	The number of ml of N/10 KOH required to neutralize the fatty acids in the distillate from 5 gm of fat is called	Reichert-Meissel number	Polenske number	Acetyl number	Non volatile fatty acid number	**Reichert-Meissel number**
26	3	Molecular formula of cholesterol is	C27H45OH	C29H47OH	C29H47OH	C23H41OH	**C27H45OH**
27	3	The cholesterol molecule is	Benzene derivative	Quinoline derivative	Steroid	Straight chain acid	**Steroid**
28	3	Salkowski test is performed to detect	Glycerol	Cholesterol	Fatty acids	Vitamin D	**Cholesterol**
29	3	Palmitic, oleic or stearic acid ester of cholesterol used in manufacture of cosmetic creams is	Elaidic oil	Lanoline	Spermaceti	Chaulmoogric oil	**Lanoline**
30	3	Dietary fats after absorption appear in the circulation as	HDL	VLDL	LDL	Chylomicron	**Chylomicron**
31	3	Free fatty acids are transported in the blood	Combined with albumin	Combined with fatty acid binding protein	Combined with β - lipoprotein	In unbound free salts	**Combined with albumin**
32	3	Long chain fatty acids are first activated to acetyl-CoA in	Cytosol	Microsomes	Nucleus	Mitochondria	**Cytosol**
33	3	The enzyme acyl-CoA synthase catalyses the conversion of a fatty acid of an active fatty acid in the presence of	AMP	ADP	ATP	GTP	**ATP**
34	3	Carnitine is synthesized from	Lysine and methionine	Glycine and arginine	Aspartate and glutamate	Proline and hydroxyproline	**Lysine and methionine**
35	3	The enzymes of β-oxidation are found in	Mitochondria	Cytosol	Golgi apparatus	Nucleus	**Mitochondria**
36	3	Long chain fatty acids penetrate the inner mitochondrial membrane	Freely	As acyl-CoA derivative	As carnitine derivative	Requiring Na dependent carrier	**As carnitine derivative**
37	3	Dietary fibres are rich in	Cellulose	Glycogen	Starch	Proteoglycans	**Cellulose**

38	3	The end products of saponification:	glycerol	acid	soap	Both (A and (C	**Both (A and (C**
39	3	Triglycerides are	Heavier than water	Major constituents of membranes	Non-polar	Hydrophilic	**Non-polar**
40	3	Cerebronic acid is present in	Glycerophospholipids	Sphingophospholipids	Galactosyl ceramide	Gangliosides	**Galactosyl ceramide**
41	3	Acylsphingosine is also known as	Sphingomyelin	Ceramide	Cerebroside	Sulphatide	**Ceramide**
42	3	The highest phospholipids content is found in	Chylomicrons	VLDL	LDL	HDL	**HDL**
43	3	The major lipid in chylomicrons is	Triglycerides	Phospholipids	Cholesterol	Free fatty acids	**Triglycerides**
44	3	Number of carbon atoms in cholesterol is	17	19	27	30	**27**
45	3	The lipoprotein richest in cholesterol is	Chylomicrons	VLDL	LDL	HDL	**LDL**
46	3	The nitrogenous base in lecithin is	Ethanolamine	Choline	Serine	Betaine	**Choline**
47	3	All the following are omega-6-fatty acids except	Linoleic acid	α-Linolenic acid	γ-Linolenic acid	Arachidonic acid	**α-Linolenic acid**
48	3	All the following have 18 carbon atoms except	Linoleic acid	Linolenic acid	Arachidonic acid	Stearic acid	**Arachidonic acid**
49	3	A 20-carbon fatty acid among the following is	Linoleic acid	α -Linolenic acid	β -Linolenic acid	Arachidonic acid	**Arachidonic acid**
50	3	Predominant fatty acids in meat are	Saturated	Monounsaturated	Polyunsaturated	Mono and poly-unsaturated	**Saturated**
51	3	Cholesterol is present in all of the following except	Egg	Fish	Milk	Pulses	**Pulses**
52	3	Which of the following has the highest cholesterol content?	Meat	Fish	Butter	Milk	**Butter**
53	3	Cholesterol is a	Animal sterol	M.F. C27 H46O	5 methyl groups	All of these	**All of these**
54	3	Lieberman-Burchard reaction is performed to detect	Cholesterol	Glycerol	Fatty acid	Vitamin D	**Cholesterol**
55	3	Fatty acids are oxidized by	α -oxidation	β -oxidation	ω -oxidation	All of these	**All of these**
56	3	Which of the following is not an unsaturated fatty acid?	Oleic acid	Stearic acid	Linaoleic acid	Palmitic acid	**Stearic acid**

57	3	Calorific value of lipids per gm is	4 Kcal	8 Kcal	9 Kcal	None of these	**9 Kcal**
58	3	Saponification:	Hydrolysis of fats by alkali	Hydrolysis of glycerol by liposes	Esterification	Reduction	**Hydrolysis of fats by alkali**
59	3	In cephalin, choline is replaced by	Serine	Ethanolamine	Betaine	Sphingosine	**Ethanolamine**
60	3	A fatty acid not synthesized in man is	Oleic	Palmitic	Linoleic	Stearic	**Linoleic**

UNIT-IV

SYLLABUS

Classification and functions of proteins and amino acids, structure of amino acids and concept of zwitterions. Ninhydrin reaction. Natural modifications of amino acids in proteins. Non-protein amino acids, Oligopeptides: Structure and functions of glutathione, insulin and aspartame. Primary and secondary structure of proteins-alpha helix, beta pleated sheet. Tertiary and quaternary structures of proteins. Human haemoglobin structure.
Structure and classification of enzymes, mechanism of action of enzymes. Km equation and enzyme activity. Allosteric enzyme and its mechanism. Multienzyme complex, enzyme inhibition. Vitamins-classification and characteristics, sources and importance.

Proteins

Proteins are important macromolecules of the cells, formed by the polymerization of amino acids. Proteins are the mode of expression of the genetic information. They performs various biology functions in the cells, such as they act as the structural components of cells, enzymes, hormones, pigments, storage proteins and some toxins in the cells.

Classification of proteins

Proteins are classified based on the following criterions:

(1) Structure
(2) Composition
(3) Function

Structure

Based on the structure, proteins are classified into 3 groups

(a) Fibrous proteins
(b) Globular Proteins
(c) Intermediate proteins

Fibrous Proteins

- They are linear in shape

- Secondary structure is the most important functional structure of fibrous proteins
- Usually, these proteins do not have tertiary structures
- Physically fibrous proteins are very tough and strong
- They are insoluble in the water
- Long parallel polypeptide chains cross linked at regular intervals
- Fibrous proteins form long fibres or sheaths.

Functions of fibrous proteins

- Perform the structural functions in the cells

Example: Collagen, Myosin, Silk and keratin

Globular Proteins

- Globular proteins are spherical or globular in shape
- The polypeptide chain is tightly folded into spherical shapes
- Tertiary structure is the most important functional structure in globular proteins
- Physically they are soft than fibrous proteins.
- They are readily soluble in water.
- Most of the proteins in the cells belong to the category of globular proteins.

Functions

- Form enzymes, antibodies, and some hormones.

Example: Insulin, Haemoglobin, DNA polymerase and RNA polymerase.

Intermediate proteins

- Their structure is intermediate to linear and globular structures.
- They are short and more or less linear shaped proteins
- Unlike Fibrous proteins, they are soluble in water.

Functions:

- Blood clotting proteins

Example: Fibrinogen

Composition

They are broadly divided into two types

(a) Simple proteins

(b) Conjugated proteins

Simple proteins

Simple proteins composed of only amino acids

Proteins may be fibrous or globular

They possess relatively simple structural organization

Example: Collagen, Myosin, Insulin, Keratin

Conjugated proteins

- Conjugated proteins are complex proteins
- They contain one or more non-amino acid components.
- Here the protein is tightly or loosely bound to one or more non-protein parts.
- The non-protein parts of these proteins are called prosthetic groups.
- The prosthetic group may be metal ions, carbohydrates, lipids, phosphoric acids, nucleic acids and FAD.
- The prosthetic group is essential for the biological functions of these proteins.
- Conjugated proteins are usually globular in shape and are soluble in water.
- Most of the enzymes are conjugated proteins.

Based on the nature of prosthetic groups, the conjugated proteins are further classified as follows.

Phosphoprotein: Prosthetic group is phosphoric acid, Example-casein of milk, vitellin of egg yolk.

Glycoproteins: Prosthetic group is carbohydrates, Example- most of the membrane proteins, mucin (Component of saliva).

Nucleoprotein: Prosthetic group is nucleic acid, Example- proteins in chromosomes, structural proteins of ribosome.

Chromoproteins: Prosthetic group is pigment of chrome, Example: Haemoglobin, Phytochrome and Cytochrome.

Lipoproteins: Prosthetic group is lipids, Example: Membrane proteins.

Flavoproteins: Prosthetic group is FAD (Flavin Adenine Dinucleotide), Example: Proteins of Electron Transport System (ETS).

Metalloproteins: Prosthetic group is metal ions, Example: Nitrate reductase.

Functions

Structural proteins

- Form the component of the connective tissue, bone, tendons, cartilage, skin, feathers, nails, hairs and horn
- Most of them are fibrous proteins and are insoluble in water.

Example: Collagen, Keratin and Elastin

Enzymes

- They are the biological catalysts
- Enzymes reduce the activation energy of reactants and speed up the metabolic reactions in the cells.
- Most of them are globular conjugated proteins.

Hormones

- They include the proteinaceous hormones in the cells.

Example: Insulin, Glucagon

Respiratory pigments

- They are coloured proteins
- All of them are conjugated proteins and they contain pigments (chrome) as their prosthetic group.

Example: Haemoglobin, Myoglobin

Transport proteins

- They transport the materials in the cells
- They form channels in the plasma membrane
- They also form one of the components of blood and lymph in animals

Example: Serum albumin

Contractile proteins

- They are the force generators of muscles
- They can contract with the expense of energy from ATP molecules.

Example: Actin, Myosin

Storage proteins

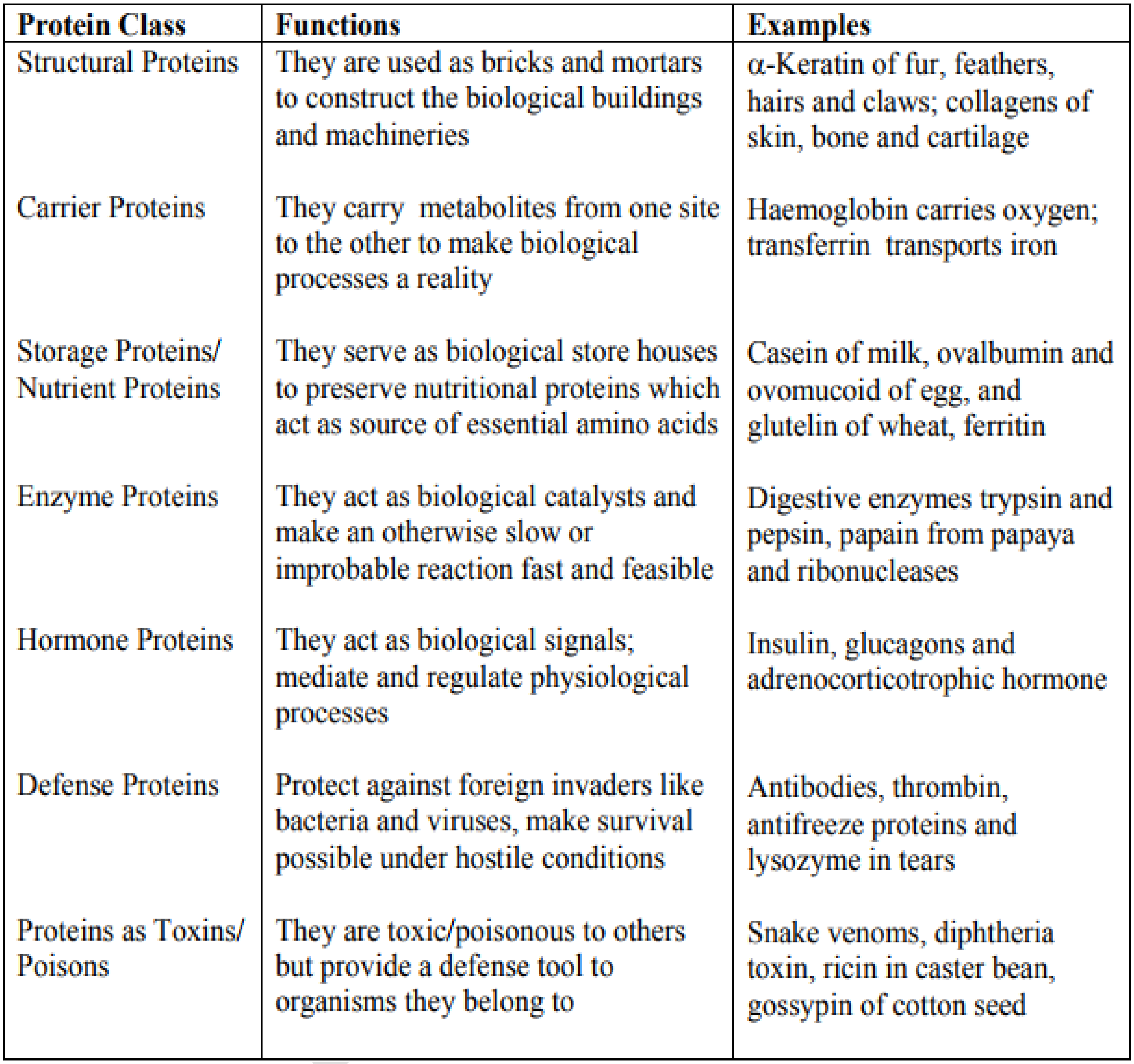
- They act as the store of metal ions and amino acids in the cells
- Found in seeds, egg and milk
- Abundantly seen in pulses (Legume seeds).

Example: Ferritin which stores iron, casein

Toxins

- They are toxic proteins

Example: Snake venom

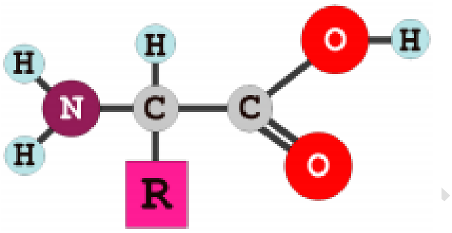


Protein Class	Functions	Examples
Structural Proteins	They are used as bricks and mortars to construct the biological buildings and machineries	α-Keratin of fur, feathers, hairs and claws; collagens of skin, bone and cartilage
Carrier Proteins	They carry metabolites from one site to the other to make biological processes a reality	Haemoglobin carries oxygen; transferrin transports iron
Storage Proteins/ Nutrient Proteins	They serve as biological store houses to preserve nutritional proteins which act as source of essential amino acids	Casein of milk, ovalbumin and ovomucoid of egg, and glutelin of wheat, ferritin
Enzyme Proteins	They act as biological catalysts and make an otherwise slow or improbable reaction fast and feasible	Digestive enzymes trypsin and pepsin, papain from papaya and ribonucleases
Hormone Proteins	They act as biological signals; mediate and regulate physiological processes	Insulin, glucagons and adrenocorticotrophic hormone
Defense Proteins	Protect against foreign invaders like bacteria and viruses, make survival possible under hostile conditions	Antibodies, thrombin, antifreeze proteins and lysozyme in tears
Proteins as Toxins/ Poisons	They are toxic/poisonous to others but provide a defense tool to organisms they belong to	Snake venoms, diphtheria toxin, ricin in caster bean, gossypin of cotton seed

Functions of proteins

Amino acids

Amino acids as the building blocks of proteins. Amino acids is defined as a molecule containing an amine group (-NH2), carboxyl group (-COOH) and the variable group denoted as R, different among different amino acids. R groups is also called the side chain, The overall amino acid formula can be represented as : R-CH(NH2)-COOH. An average molecular weight is about 135 daltons.



Classification of Amino acids

Amino acids are broadly classified into four types

- **(a) Non-Polar**
- **(b) Polar**
- **(c) Acidic**
- **(d) Basic**

Other type of classifications

- **(a) Essential**
- **(b) Non-essential**
- **(c) Semi-essential**

Non-polar Amino Acids

- The non-polar amino acids contain mostly hydrocarbon R groups that do not bear positive or negative charges.
- Non-polar (i.e., hydrophobic) amino acids play an important role in maintaining the three-dimensional structures of proteins, because they interact poorly with water.

Two types of hydrocarbon side chains are found in this group:

- **(a) Aromatic**
- **(b) Aliphatic**

Aromatic amino acids

- Aromatic amino acid contains aromatic ring in their structure
- Benzene is one of the simplest aromatic amino acid.

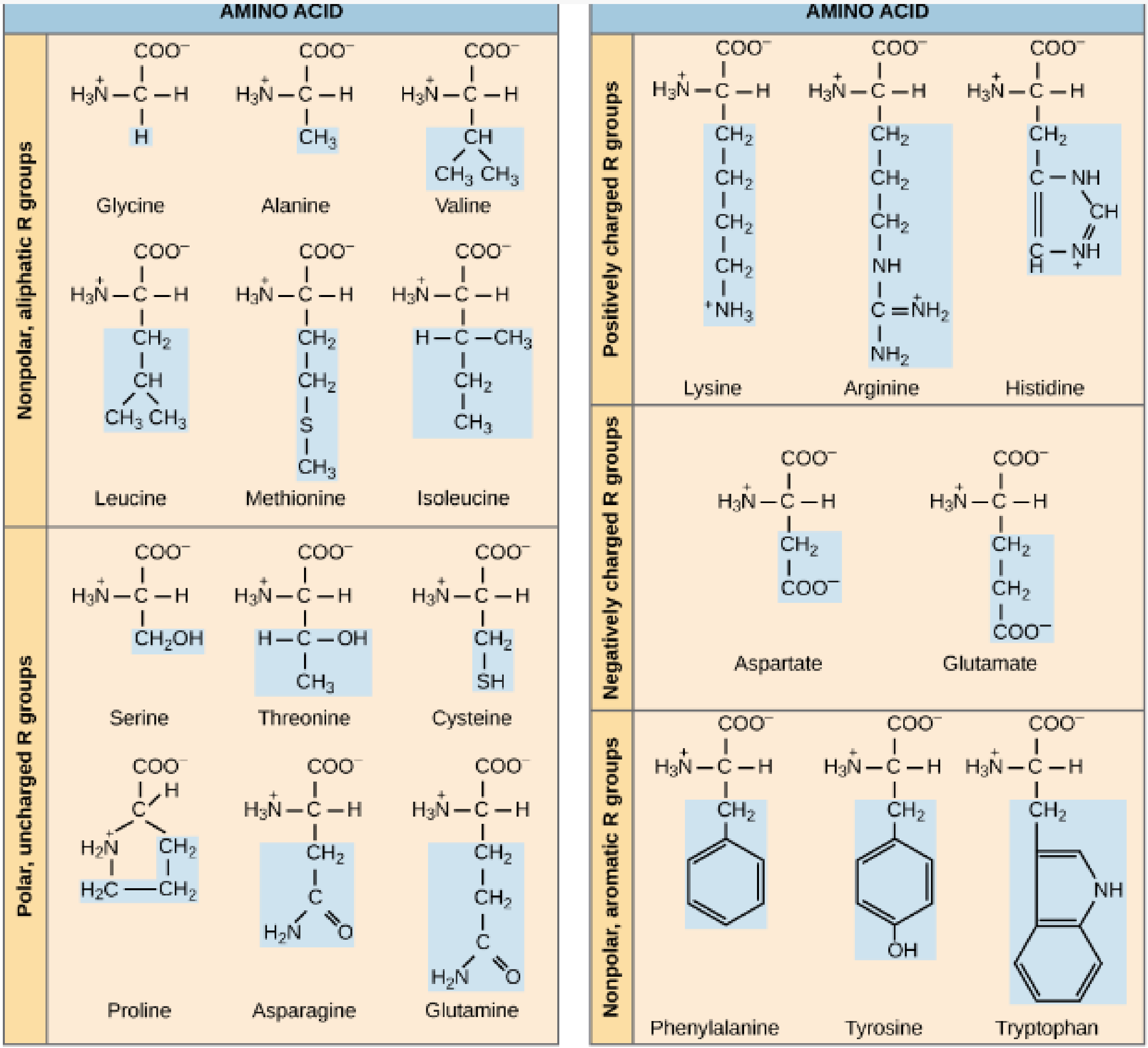
Aliphatic amino acids

- The term aliphatic refers to non-aromatic amino acids such as methane and cyclohexane.
- Phenylalanine and tryptophan contain aromatic ring structures.
- Glycine, alanine, valine, leucine, isoleucine, and proline have aliphatic R groups.
- A sulfur atom appears in the aliphatic side chains of methionine and cysteine. Methionine contains a thioether group (—S—CH3) in its side chain.

Polar Amino Acids

- Polar amino acids have functional groups capable of hydrogen bonding, they easily interact with water.
- Polar amino acids are described as hydrophilic, or “water-loving.”
- Serine, threonine, tyrosine, asparagine, and glutamine belong to this category.
- Serine, threonine, and tyrosine contain a polar hydroxyl group, which enables them to participate in hydrogen bonding, an important factor in protein structure.

- The hydroxyl groups serve other functions in proteins.



Acidic Amino Acids

- Two standard amino acids have side chains with carboxylate groups.

- Because the side chains of aspartic acid and glutamic acid are negatively charged at physiological pH, they are often referred to as aspartate and glutamate.

Basic Amino acids

- Basic amino acids bear a positive charge at physiological pH.
- They can therefore form ionic bonds with acidic amino acids.
- Lysine, which has a side chain amino group, accepts a proton from water to form the conjugate acid (—NH3).

Classification based on nutritional requirements

Essential amino acids:

These amino acids cannot be synthesized in the body and have to be present essentially in the diet.

Examples: Valine, Isoleucine, Leucine, Lysine, Methionine, Threonin, Tryptophan and Phenylalanine.

Semi-essential amino acids

These amino acids can be synthesized in the body but the rate of synthesis is lesser than the requirement (e.g. during growth, repair or pregnancy)

Example: Arginine and Histidine

Non-essential amino acids

These amino acids are synthesized in the body, thus their absence in the diet does not adversely affect the growth.

Example: Glycine, Alanine and the other remaining amino acids.

Physical Properties of amino acids

- Colourless
- Crystalline
- May be sweet (Glycine, Alanine, Valine)
- Tasteless (Leucine)
- Bitter (Arginine, Isoleucine).
- Soluble in water, acids, but insoluble in organic solvents.

- High melting point (More than 200° C).

Chemical Properties of amino acids

Reaction due to NH2 group

- Reaction with acids to form salt
- Reaction with nitrous acids to liberate Nitrogen
- Reaction with CO2 to form carbamino compounds

Reaction due to COOH group

- Reaction with strong alkalies to form salt
- Reaction with alcohols to form esters

Reaction due to both NH2 and COOH group

- Amino acids condense with each other by COOH group at one amino acids with NH2 of other amino acid to form peptide bond.

Functions of amino acids

Histidine

- Found in high concentrations in hemoglobin.
- Useful in treating anemia due to relationship to hemoglobin.
- Has been used to treat rheumatoid arthritis.
- Precursor to histamine.
- Associated with allergic response and has been used to treat allergy.
- Assists in maintaining proper blood pH.

Isoleucine

- Muscle tissue uses Isoleucine as an energy source.
- Required in the formation of hemoglobin.

Leucine

- Potent stimulator of insulin.
- Helps with bone healing.
- Helps promote skin healing.

- Modulates release of Enkephalins, which are natural pain-reducers.

Lysine

- Helps form collagen, the connective tissue present in bones, ligaments, tendons, and joints.
- Assists in the absorption of calcium.
- Essential for children, as it is critical for bone formation.
- Involved in hormone production.
- Lowers serum triglyceride levels.

Methionine

- Assists in breakdown of fats.
- Precursor of the amino acids Cysteine and Taurine.
- Helps reduce blood cholesterol levels.
- Antioxidant.
- Assists in the removal of toxic wastes from the liver.
- Helps prevent disorder of hair, skin, and nails due to sulfur and anti-oxidant activity.
- Required for synthesis of RNA and DNA.
- Natural chelating agent for heavy metals, such as lead and mercury.

Phenylalanine

- Precursor to the hormone, Thyroxin.
- Enhances mood, clarity of thought, concentration, and memory.
- Suppresses appetite.
- Major part of collagen formation.
- Powerful anti-depressant.
- Used in the treatment of Parkinson's Disease.

Threonine

- Required for formation of collagen.
- Helps prevent fatty deposits in the liver.

- Aids in production of antibodies.
- Can be converted to Glycine (a neurotransmitter) in the central nervous system.
- Acts as detoxifier.
- Needed by the GI (gastrointestinal) tract for normal functioning.

Tryptophan

- Precursor to the key neurotransmitter, serotonin, which exerts a calming effect.
- Effective sleep aid, due to conversion to serotonin.
- Effective in some forms of depression.
- Treatment for migraine headaches.
- Stimulates growth hormone.
- Tryptophan must compete with 5 other amino acids to pass through the blood-brain barrier and enter the brain. Those 5 are: tyrosine, phenylalanine, leucine, isoleucine, and valine and are called Large Neutral Amino Acids (LNAA).

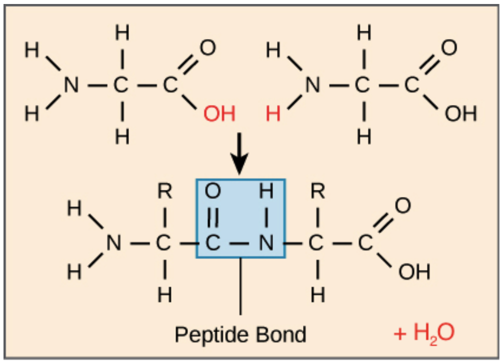
Valine

- Competes with Tyrosine and Tryptophan in crossing the blood-brain barrier.
- The higher the Valine level, the lower the brain levels of Tyrosine and Tryptophan.
- Actively absorbed and used directly by muscle as an energy source.

Peptide bonds

Each protein in your cells consists of one or more polypeptide chains. Each of these polypeptide chains is made up of amino acids, linked together in a specific order. The chemical properties and order of the amino acids are key in determining the structure and function of the polypeptide

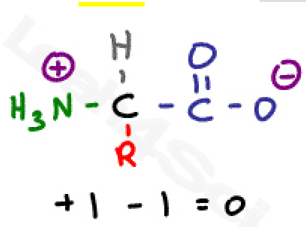
The amino acids of a polypeptide are attached to their neighbors by covalent bonds known as peptide bonds. Each bond forms in a dehydration synthesis (condensation) reaction. During protein synthesis, the carboxyl group of the amino acid at the end of the growing polypeptide chain reacts with the amino group of an incoming amino acid, releasing a molecule of water. The resulting bond between amino acids is a peptide bond.



At one end, the polypeptide has a free amino group, and this end is called the amino terminus (or N-terminus). The other end, which has a free carboxyl group, is known as the carboxyl terminus (or C-terminus).

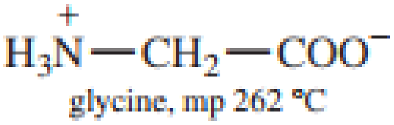
Concept of Zwitterions

Zwitterion is the dipolar form of an amino acid which occurs when H^+ ion is transferred from an acid group to an amine group.



The dipolar nature of amino acids gives them some unusual properties:

1. Amino acids have high melting points, generally over 200° C.



$$H_3\overset{+}{N}—CH_2—COO^-$$

glycine, mp 262 °C

2. Amino acids are more soluble in water than they are in ether, dichloromethane, and other common organic solvents.

3. Amino acids have much larger dipole moments than simple amines or simple acids.

4. Amino acids are less acidic than most carboxylic acids and less basic than most amines.

Ninhydrin reaction

The reaction between alpha-amino acid and ninhydrin involved in the development of color.

alpha-amino acid + ninhydrin ---> reduced ninhydrin + alpha-amino acid + H_2O

alpha-amino acid + H_2O ---> alpha-keto acid +NH_3

alpha-keto acid + NH_3 ---> aldehyde + CO_2

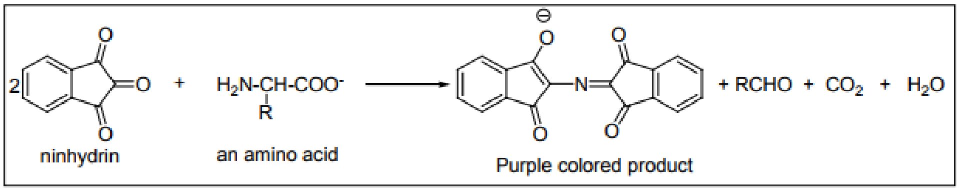
Step 1: it is an oxidative deamination reaction that removes two hydrogen from the alpha amino acid to yield an alpha-imino acid. Simultaneously, the original ninhydrin is reduced and loses an oxygen atom with the formation of a water molecule.

In Step 2: The NH group in the alpha imino acid is rapidly hydrolyzed to form an alpha keto acid with the production of an ammonia molecules.

Step 3: This alpha-keto acid further undergoes decarboxylation reaction under a heated condition to form an aldehyde that has one less carbon atom than the original amino acid. A carbon dioxide molecule is produced.

Further the overall, reaction becomes

alpha-amino acid + 2 ninhydrin ---> CO_2 + aldehyde + final complex(BlUE) + $3H_2O$



Non-Non-Protein amino acids

The amino acid which are not involved in the protein synthesis are called as non protein amino acids.

These non-protein amino acids are classified into two types

(a) Alpha

(b) Non-alpha

Alpha amino acids

- Ornithine
- Citrulline
- Thyroxine
- S-adenosylmethionine
- Homecysteine
- Ovathiol
- Azaserine

Non-Alpha amino acids

- Beta-alanine
- Beta-aminoisobutyric acid
- Gama-aminobutyric acid
- Aminolevulinic acid
- taurine

Alpha Amino acid

Ornithine

- Ornithine is precursors of polyamine
- Ornithine enters liver, mitochondria and participates in urea synthesis.

Citrulline

- Citrulline is intermediates in the biosynthesis of urea.
- L-ornithine transcarbamoylase catalyzes transfer of the carbamoyl group carbamoyl phosphate to ornithine, forming cirtulline and orthophosphate. While the reaction occurs in the mitochondrial matrix, both the formation of ornithine and the subsequent metabolism of citrulline take place in the cytosol.

Thyrosine

- Tyrosine forms norepinephrine and epinephrine and following iodination the thyroid hormones triiodothyronine and thyroxine.
- Use of measurement of blood thyroxine or thyroid stimulating hormone (TSH) in the neonatal diagnosis of congenital hypothyroisidm.
- The amino acid thyrosine is the starting point in the synthesis of the catecholamines and of the thyroid hormones tetraiodothyronine (thyroxine; T4) and triiodothyronine (T3).

Ovathiol

- Sulfur containing amino acid found in fertilized eggs, and acts as an antioxidant.

Azaserine

- Purine deficiency states, while rare in humans, generally reflect a deficiency of folic acid.
- Compounds that inhibit formation of tetrahydrofolates and therefore block purine synthesis have been used in cancer chemotherapy.
- Inhibitory compounds and the reactions they inhibit include, azaserine, diazanorleucine, 6-mercaptopurine and mycophenolic acid.

Non-alpha amino acids

Beta-Alanine and Aminoisobutyrate

- Alanine and aminoisobutyrate are formed during catabolism of the pyrimidines uracil and thymine.
- Traces of alanine also result from the hydrolysis of alanyl dipeptides by the enzyme carnosinase.
- Aminoisobutyrate also arises by transamination of methylmalonate semialdehyde, a catabolite of L-valine.
- The initial reaction of alanine catabolism is transamination to malonate semialdehyde.

- Subsequent transfer of coenzyme A from succinyl-CoA forms malonyl-CoA semialdehyde which is then oxidized to malonyl-CoA and decarboxylated to the amphibolic intermediate acetyl-CoA.

Beta-Alanyl Dipeptides

- The alanyl dipeptides carnosine and anserine (N-methylcarnosine) activate myosin ATPase chelate copper, and enhance copper uptake.
- Analyl-imidazole buffers the pH of anaerobically contracting skeletal muscle.
- Biosynthesis of carnosine is catalyzed by carnosine synthetase in a two-stage reaction that involves initial formation of an enzyme-bound acyl-adenylate of alanine and subsequenct transfer of the alanyl moiety to L-histidine.
- Hydrolysis of carnosine to alanine and L-histidine is catalyzed by carnosinase. The heritable disorder carnosinase deficiency is characterized by carnosinuria.
- Homocarnosine present in human brain at higher levels than carnosine is synthesized in brain tissue by carnosine synthetase. Serum carnosinase does not hydrolyze homocarnosine.
- Homocarnosinosis, a rare genetic disorder, is associated with progressive spastic paraplegia and mental retardation.

Gama-Aminobutyrate

- Gama-Aminobutyrate (GABA) functions in brain tissue as an inhibitory neurotransmitter by altering transmembrane potential differences.
- GABA is formed by decarboxylation of glutamate by L-glutamate decarboxylase.
- Trasamination of aminobutyrate forms succinate semialdehyde which can be reduced to hydroxybutyrate by L-lactate dehydrogenase,or be oxidized to succinate and thence via the citric and cycle to CO_2 and H_2O.
- A rare genetic disorder of GABA metabolism involves a defective GABA amino transferase, an enzyme that participate in the catabolism of GABA subsequent to its postsynpatic release in brain tissue.

- Defects in succinic semialdehyde dehydrogenase are responsible for another rare metabolic disorder of aminobutyrate catabolism characterized by 4-hydroxybutyric aciduria.

Structure and functions of Glutathione

Glutathione (GSH) is often referred to as the body's master antioxidant.

Composted of three amino acids

- Cysteine
- Glycine
- Glutamate

Glutathione can be found in virtually every cell of the human body.

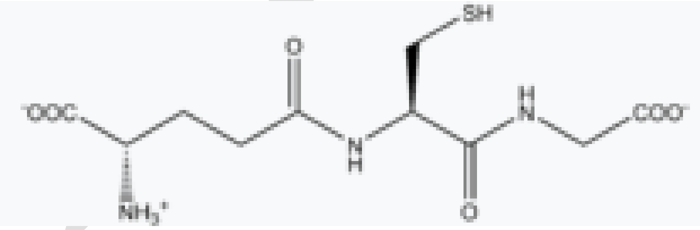
The highest concentration of glutathione is in the liver, making it critical in the body's detoxification process.

Glutathione is also an essential component to the body's natural defense system.

Viruses, bacteria, heavy metal toxicity, radiation, certain medications, and even the normal process of aging can all cause free-radical damage to healthy cells and deplete glutathione.

Glutathione depletion has been correlated with lower immune function and increased vulnerability to infection due to the liver's reduced ability to detoxify.

As the generation of free radicals exceeds the body's ability to neutralize and eliminate them, oxidative stress occurs.



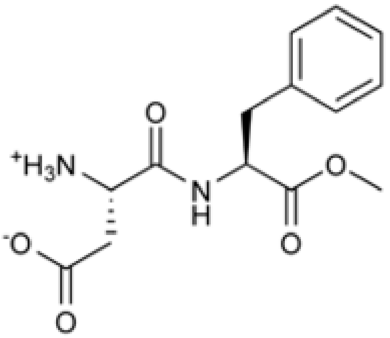
Functions of Glutathione

- It maintains levels of reduced glutaredoxin and glutathione peroxidase.
- It is one of the major endogenous antioxidants produced by the cells, participating directly in the neutralization of free radicals and reactive oxygen compounds

- Regulation of the nitric oxide cycle is critical for life, but can be problematic if unregulated
- It is used in metabolic and biochemical reactions such as DNA synthesis and repair, protein synthesis, prostaglandin synthesis, amino acid transport, and enzyme activation.
- Thus, every system in the body can be affected by the state of the glutathione system, especially the immune system, the nervous system, the gastrointestinal system, and the lungs.
- It has a vital function in iron metabolism.
- It has roles in progression of the cell cycle, including cell death.
- GSH levels regulate redox changes to nuclear proteins necessary for the initiation of cell differentiation.
- Differences in GSH levels also determine the expressed mode of cell death, being either apoptosis or cell necrosis.
- Manageably low levels result in the systematic breakage of the cell whereas excessively low levels result in rapid cell death.

Aspartame

- Aspartame is an artificial sweeter.
- Aspartame is 180 to 200 times sweeter than normal sugar.
- Aspartame is not suitable for baking because if often breaks down when heated and loses much of its wetness and at temperatures above 90° Fa component of it can covert to formaldehyde.



These aspartame is synthesized from two amino acids

- Aspartic acid
- Phenylalanine

Aspartame has the chemical formula of $C_{14}H_{18}N_2O_5$.

Upon ingestion, aspartame breaks down into several residual chemicals, including

- Aspartic acid
- Phenylalanine
- Methanol
- Formaldehyde

Methanol and Formaldehyde

- Approximately 10% of aspartame is broken down into methanol in the small intestine. Most of the methanol is absorbed and quickly converted into formaldehyde.
- High concentration, formaldehyde can kill cells and tissues.

Phenylalanine

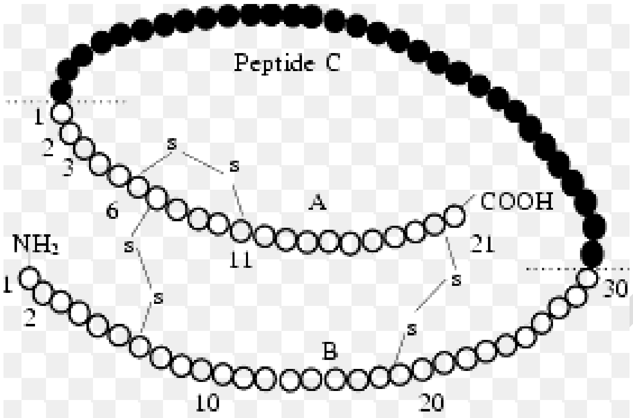
- One of the functional groups in aspartame is phenylalanine, which is unsafe for those born with phenylketonuria, a rare genetic conditions.
- Approximately 50% of aspartame is broken down into phenylalanine, which is completely safe for everyone except sufferers of phenylketonuria.

Aspartic acid

- Approximately 40 percent of aspartame is broken down into aspartic acid.
- Aspartic acid belongs to a class of chemicals that in high concentrations act as an excitotoxin, damage on brain and nerve cell.

Structure of Insulin

Insulin is a hormone secreted by the pancreas that regulates glucose levels in the blood. Without insulin, cells cannot use the energy from glucose to carry out functions within the body. Insulin was first discovered in 1921 by Frederick Grant Banting and Charles. The FDA approved insulin in 1939.



Insulin is composed of two peptide chains referred to as the A chain and B chain. A and B chains are linked together by two disulfide bonds, and an additional disulfide is formed within the A chain. In most species, the A chain consists of 21 amino acids and the B chain of 30 amino acids.

Although the amino acid sequence of insulin varies among species, certain segments of the molecule are highly conserved, including the positions of the three disulfide bonds, both ends of the A chain and the C-terminal residues of the B chain. These similarities in the amino acid sequence of insulin lead to a three dimensional conformation of insulin that is very similar among species, and insulin from one animal is very likely biologically active in other species. Indeed, pig insulin has been widely used to treat human patients.

Insulin molecules have a tendency to form dimers in solution due to hydrogen-bonding between the C-termini of B chains. Additionally, in the presence of zinc ions, insulin dimers associate into hexamers.

Functions of insulin

- Insulin is made in the pancreas by beta cells.
- After the body takes in food, these beta cells release insulin, which enables cells in the liver, muscles and fat tissues to take up glucose and either store it as glycogen or allow blood to transfer it to organs in the body for use as an energy source.
- This process stops the use of fat as a source of energy.

- When glucose levels are elevated in the blood, insulin is produced at higher rates by the pancreas in order to maintain normal sugar concentrations in the blood.
- Without insulin, the body cannot process glucose effectively and glucose begins to build up in the blood stream instead of being transported to different cells.
- In contrast with elevated levels of glucose in the blood, when there is a deficit of glucose available to the body, alpha cells in the pancreas release glucagon, a hormone that causes the liver to convert stored glycogen into usable glucose which is then released into the bloodstream.

Some of the effects of the insulin on the metabolism include:

- Controlling cell intake of substances like glucose in many organs like muscles and adipose tissues.
- Controlling amino acid uptake, thus increasing DNA replication and protein synthesis
- Altering the activity of enzymatic cells

Other Cellular effects of insulin include:

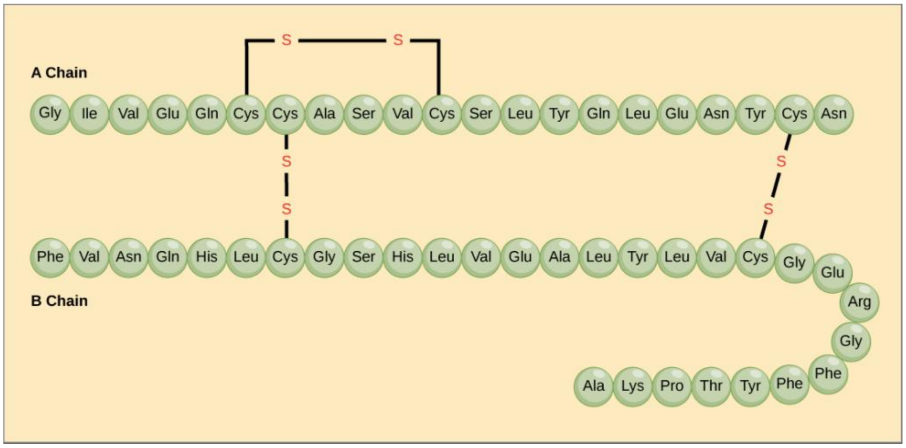
- Increasing synthesis of glycogen. Glycogen is a type of storage for glucose and is stored in the liver. Levels of blood glucose determine whether glucose is stored as glycogen or is excreted. Low levels of glucose cause the liver to excrete glucose, while higher levels of glucose allow glucose to be stored as glycogen.
- Increasing the synthesis and esterification of fatty acids. This is caused by the insulin causing fat cells to convert blood lipids to triglycerides. Esterification is caused when the insulin causes the adipose tissue to convert fats from fatty acid esters.
- Increasing the esterification of fatty 4. Decreasing protein breakdown (proteolysis) 5. Reducing lipolysis 6. Increasing uptake of substances like amino acid and potassium 7. Relaxing wall of arteries of muscles, which vasodilation 8. Increasing secretion of HCl into the stomach.

Protein structure

- Egg whites contain large amounts of proteins called albumins, and the albumins normally have a specific 3D shape, thanks to bonds formed between different amino acids in the protein. Heating causes these bonds to break and exposes hydrophobic (water-hating) amino acids usually kept on the inside of the protein. The hydrophobic amino acids, trying to get away from the water surrounding them in the egg white, will stick to one another, forming a protein network that gives the egg white structure while turning it white and opaque.
- The shape of a protein is very important to its function.
- To understand how a protein gets its final shape or conformation, we need to understand the four levels of protein structure: primary, secondary, tertiary, and quaternary.

Primary structure

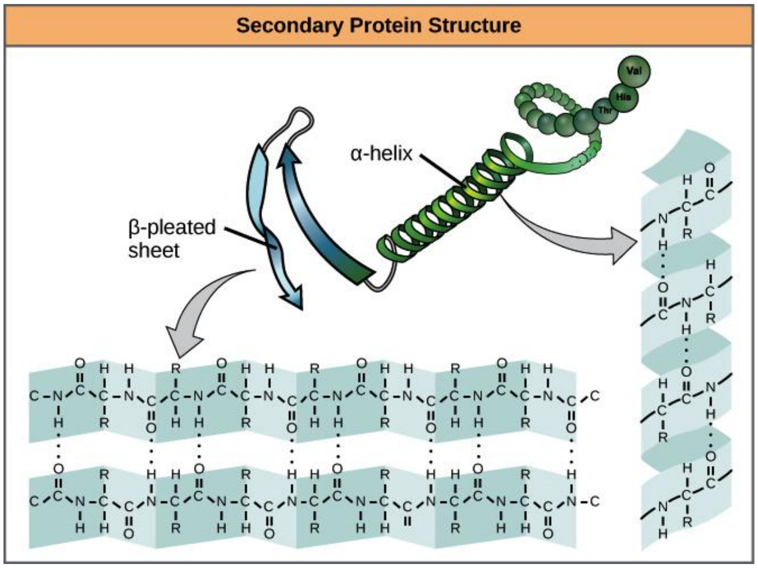
- The simplest level of protein structure, **primary structure**, is simply the sequence of amino acids in a polypeptide chain.
- For example, the hormone insulin has two polypeptide chains, A and B, shown in diagram below. (The insulin molecule shown here is cow insulin, although its structure is similar to that of human insulin.)
- Each chain has its own set of amino acids, assembled in a particular order.
- For instance, the sequence of the A chain starts with glycine at the N-terminus and ends with asparagine at the C-terminus, and is different from the sequence of the B chain.



- Insulin consists of an A chain and a B chain. They are connected to one another by disulfide bonds (sulfur-sulfur bonds between cysteines).
- The A chain also contains an internal disulfide bond. The amino acids that make up each chain of insulin are represented as connected circles, each with the three-letter abbreviation of the amino acid's name.

Secondary structure

- The next level of protein structure, **secondary structure**, refers to local folded structures that form within a polypeptide due to interactions between atoms of the backbone. (The backbone just refers to the polypeptide chain apart from the R groups – so all we mean here is that secondary structure does not involve R group atoms).
- The most common types of secondary structures are the α helix and the β pleated sheet. Both structures are held in shape by hydrogen bonds, which form between the carbonyl O of one amino acid and the amino H of another.

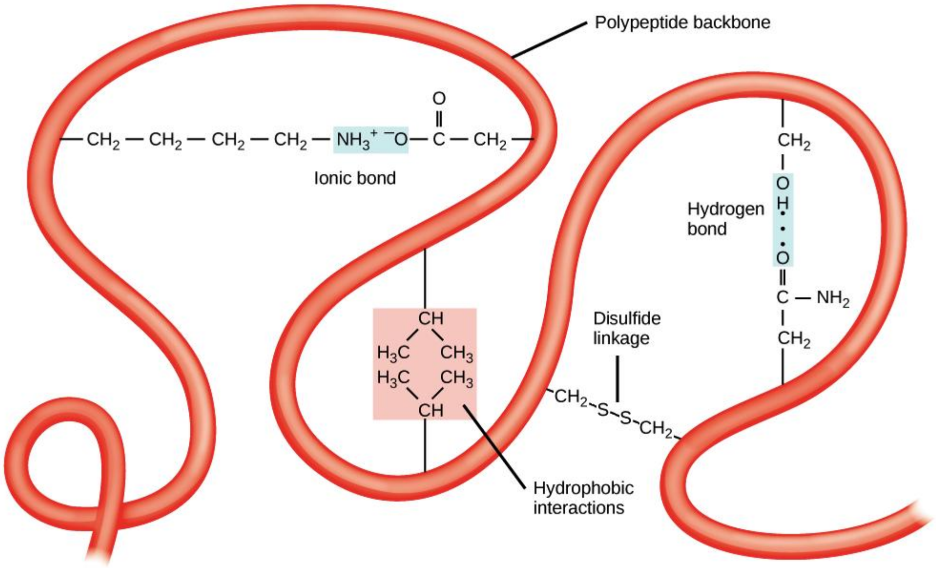


Secondary Protein Structure

- In an **α helix**, the carbonyl (C=O) of one amino acid is hydrogen bonded to the amino H (N-H) of an amino acid that is four down the chain. (E.g., the carbonyl of amino acid 1 would form a hydrogen bond to the N-H of amino acid 5).
- This pattern of bonding pulls the polypeptide chain into a helical structure that resembles a curled ribbon, with each turn of the helix containing 3.6 amino acids.
- The R groups of the amino acids stick outward from the α helix, where they are free to interact.
- In a **β pleated sheet**, two or more segments of a polypeptide chain line up next to each other, forming a sheet-like structure held together by hydrogen bonds.
- The hydrogen bonds form between carbonyl and amino groups of backbone, while the R groups extend above and below the plane of the sheet.
- The strands of a β pleated sheet may be **parallel**, pointing in the same direction (meaning that their N- and C-termini match up), or **antiparallel**, pointing in opposite directions (meaning that the N-terminus of one strand is positioned next to the C-terminus of the other).

Tertiary structure

- The overall three-dimensional structure of a polypeptide is called its **tertiary structure**. The tertiary structure is primarily due to interactions between the R groups of the amino acids that make up the protein.
- R group interactions that contribute to tertiary structure include hydrogen bonding, ionic bonding, dipole-dipole interactions.
- For example, R groups with like charges repel one another, while those with opposite charges can form an ionic bond. Similarly, polar R groups can form hydrogen bonds and other dipole-dipole interactions. Also important to tertiary structure are **hydrophobic interactions**, in which amino acids with nonpolar, hydrophobic R groups cluster together on the inside of the protein, leaving hydrophilic amino acids on the outside to interact with surrounding water molecules.
- Finally, there's one special type of covalent bond that can contribute to tertiary structure: the disulfide bond. **Disulfide bonds**, covalent linkages between the sulfur-containing side chains of cysteines, are much stronger than the other types of bonds that contribute to tertiary structure.



Quaternary structure

- Many proteins are made up of a single polypeptide chain and have only three levels of structure. However, some proteins are made up of multiple polypeptide chains, also known as subunits. When these subunits come together, they give the protein its **quaternary structure**.
- one example of a protein with quaternary structure: hemoglobin. hemoglobin carries oxygen in the blood and is made up of four subunits, two each of the α and β types. In general, the same types of interactions that contribute to tertiary structure (mostly weak interactions, such as hydrogen bonding and London dispersion forces) also hold the subunits together to give quaternary structure.

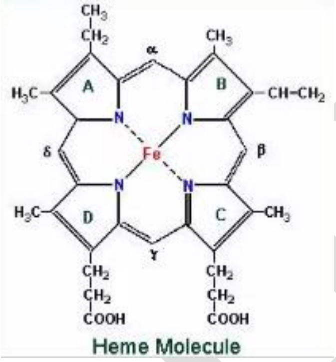
Hemoglobin

- Hemoglobin, a chromo protein, found exclusively in red blood cells is actually a conjugated protein containing heme as prosthetic group and globin as the protein part apoprotein.
- The normal concentration of Hb in an adult varies from 14.0 to 16.0 gm%.Approximately 90 mg/kg of Hb is produced and destroyed in the body every day.
- Hb has a molecular weight of about 67,000.
- Each gram of Hb contains 3.4 mg of iron.
- Heme is present as a prosthetic group in hemoglobin as well as in myoglobin, cytochromes, peroxidases, catalases and tryptophan pyrrolases etc.
- Heme is produced by the combination of iron with a porphyrin ring.
- The heme protion is alike in all forms of hemoglobin

Structure of Heme

- Heme is a derivative of porphyrin, porphyrins are cyclic compounds formed by the fusion of 4 pyrrole rings linked by methenyl bridges.
- Since an atom of iron is present heme is called ferroprotoporphyrin.

- These rings are names as I,II,III, IV and the bridges are names as alpha, beta, gamma and delta.
- Porphyrins contain side chains attached to each of the other four pyrrole rings.
- Different porphyrins vary in nature of the side chains that are attached to each of the pyrrole rings.



- Heme consists of one ferrous atom (Fe++) that is co-ordinated in the centre of the tetra pyrrole ring of protoporphyrin IX.
- The double bonds are resonating and therefore keep shifting in their position.
- When the ferrous atom in heme gets oxidized to ferric form. Hematin is formed, which loses the property of carrying oxygen and is brown in color, as compared to that of heme which is red in color.

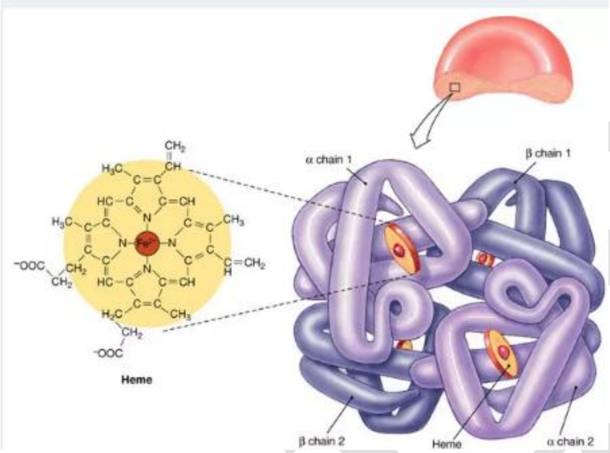
Structure of Globin

1. Different hemoglobins are produced during embryonic, fetal and adult life.
2. Each consists of a tetramer of globin polypeptide chains.
3. The major adult hemoglobin HbA has the structure $\alpha_2\beta_2$.

Polypeptide chains

- Each polypeptide chain contains heme in the heme pocket. Thus one Hb molecule contains 4 Heme units.
- The subunits of hemoglobin are arranged array with a tight spherical overall appearance and each individual polypeptide is folded in such a manner to maximize polar residues being on the exposed surface and non-polar interactions being

internal, making this large protein water soluble. The interior surface of the molecule lined with non-polar groups froms a hydrophobic pocket into which heme is inserted.



- The arrangement of polypeptides is held together by hydrogen bonding, hydrophobic interactions and multiple ionic interactions that take place at the contact points between subunits.
- These subunits interactions play a critical role in the binding of oxygen to hemoglobin.
- In the amino acid sequence of each polypeptide chain, certain residues appear to be critical to stability and function.
- Such residues are usually the same in α or β chains.
- The NH2 terimal valines of the beta chains are important in 2,3-BPG interactions. The C-terminal residues are important in the salt bridges.
- Each heme moiety can bind a single oxygen molecule, a molecule of hemoglobin can transport up to four oxygen molecules.
- Each heme unit holds an iron ion is such a way that the iron can interact with an oxygen molecule, forming oxyhemoglobin.
- Blood containg RBCs filled with oxyhemoglobin is bright red.

- The iron oxygen interaction is very weak; the two can easily be separated without damaging the heme unit or the oxygen molecules.
- The binding of an oxygen molecule to the iron in a heme unit is therefore completely reversible.
- A hemoglobin molecule in which the iron has separated from the oxygen molecule is called deoxyhemoglobin.

Primary structure of hemoglobin

- Normal alpha chain contains 141 AA residues in linear sequence.
- The non-alpha chains are all 146 amino acids in length; the beta chain begins with valine and histidine.
- The C-terminal residues are Tyrb145 and Hisb146.The delta chain differs from the beta chain in only 10 residues.
- The first eight residues are the C-terminal residues (127-146) are the same in the delta and beta chains. Tetramers of beta chains maybe found in a thalassemia.
- The gamma chain of fetal hemoglobin differs from the beta chain by 39 residues.
- The N-terminal residues of the gamma chain and beta chain are glycine and valine respectively, while the C-terminal residues.
- Try145 and His146 are the same as in gamma and beta chains. Appreciable quantities of free gamma are found in the red cells of some infants with a thalassemia, free gamma chains like beta chains can form homotetramers known as hemoglobin barts.

Secondary structure of hemoglobin

- About 75 percent of the amino acids in α or β chains are in a helical arrangement.
- All studied hemoglobins have a similar helical content.
- Eight helical arease lettered A to H, occur in the β chains.
- Hemoglobin nomenclature specifies that amino acids within helices are designated by the amino acid number and the helix letter, while amino acids between helices bear the number of the amino acid and the letters of the two helices. Thus residues

EF3 is the third residue of the segment connecting the E and F helices, while residues F8 is the eighth residue of the F helix. Alignment according to helical designation makes homology evident; residue F8 is the proximal heme-linked histidine and the histidine on the distal side of the heme is E7.

Tertiary structure

- The tertiary folding of each globin chain forms an approximate sphere. Tertiary folding fives rise to at least 3 functionally important characteristics of the hemoglobin molecules.
- Polar or charged side chains tend to be directed to the outside surface of the subunit and conversely, non-polar structure tend to the directed inwards. The effect of this is to make the surface of the molecule hydrophilic and the interior hydrophobic
- An open toped cleft in the surface of the subunit known as haem pocket iscreated.
- This hydrophobic cleft protects the ferrous ion from oxidation.
- The amino acids which form the inter-subunit bonds responsible for maintaining the quaternary structure and thus the function of the haemoglobin molecule are brought into the correct orientation to permit these bonds to form.

Quarternary structure

T-form

- The deoxy form of hemoglobin is called the "T" form or taut or tense form. In this form the two αβ dimmers interact through a network of ionic bonds and hydrogen bonds that constrain the movement of the polypeotide chains. The T form is the low oxygen affinity form of hemoglobin.

R form

- The binding of hemoglobin causes rupture of some the ionic bonds and hydrogen bonds between the αβ dimmers. This leads to a structure called "R" or relaxed form, in which the polypeptide chains have more freedom of movement. The R form is the high affinity form of hemoglobin.

Functions of hemoglobin

Hemoglobin as oxygen carrier

- The main function of hemoglobin is to carry oxygen from the lungs to all the tissues of the body. This is due to the affinity of hemoglobin for oxygen. When hemoglobin comes in contact with oxygen, it combines with it and form oxy-hemoglobin. This is a weak bond. When blood reaches to tissues, where oxygen is deficient, the bond is broken and oxygen diffuses out to tissues.

Hemoglobin as carbon dioxide carrier

- Some of carbon dioxide is transported from tissues to lungs through hemoglobin. Although the majority of it is transported via plasma but still it carries some of CO2 to lungs.

Color of blood

- The red color of blood is due to hemoglobin. When red blood cells are separated from the blood, the red color disappears. This means that the red color of blood is due to red blood cells. Hence the name red blood cells is given to it. And as we know that hemoglobin is present inside red blood cells, therefore it gives red coloration to RBCs

Buffering action

- Hemoglobin also acts as a buffer. Buffer means to resist change in pH.Blood has 7.4 pH and it remains in the narrow range. Because, if it changes the life of the person may be endangered. Therefore, hemoglobin plays very important role in keeping the pH of blood constant.

Erythrocyte metabolism

- Hemoglobin plays an important role in the modulation of erythrocyte metabolism.

Interaction with drugs

- Not only for oxygen, but hemoglobin act a very important role the transport of various drugs to their site of action.

Physiological active catabolites

- Hemoglobin is a source of various physiological active catabolites.

Karpagam Academy of Higher Education
Department of Biochemistry
I B.Sc., Microbiology
18MBU103 - Biochemistry

Question number	**Unit**	**Question**	Option I	Option II	Option III	Option IV	**Answer**
1	4	Proteins contain	Only L- α - amino acids	Only D-amino acids	DL-Amino acids	Both A) and B)	**Only L- α - amino acids**
2	4	The optically inactive amino acid is	Glycine	Serine	Threonine	Valine	**Glycine**
3	4	At neutral pH, a mixture of amino acids in solution would be predominantly:	Dipolar ions	Nonpolar molecules	Positive and monovalent	Hydrophobic	**Dipolar ions**
4	4	The true statement about solutions of amino acids at physiological pH is	All amino acids contain both positive and negative charges	All amino acids contain positively charged side chains	Some amino acids contain only positive charge	All amino acids contain negatively charged side chains	**All amino acids contain both positive and negative charges**
5	4	pH (isoelectric pH) of alanine is	6.02	6.6	6.8	7.2	**6.02**
6	4	Since the pK values for aspartic acid are 2.0, 3.9 and 10.0, it follows that the isoelectric (pH) is	3	3.9	5.9	6	**3**
7	4	Sulphur containing amino acid is	Methionine	Leucine	Valine	Asparagine	**Methionine**
8	4	All the following are sulphur containing amino acids found in proteins except	Cysteine	Cystine	Methionine	Threonine	**Threonine**
9	4	An aromatic amino acid is	Lysine	Tyrosine	Taurine	Arginine	**Tyrosine**
10	4	The functions of plasma albumin are	Osmosis	Transport	Immunity	both A)and B)	**Osmosis**

11	4	Amino acid with side chain containing basic groups is	2-Amino 5-guanidovaleric acid	2-Pyrrolidine carboxylic acid	2-Amino 3-mercaptopropanoic acid	2-Amino propanoic acid	**2-Amino 5-guanidovaleric acid**
12	4	An essential amino acid in man is	Aspartate	Tyrosine	Methionine	Serine	**Methionine**
13	4	Non essential amino acids	Are not components of tissue proteins	May be synthesized in the body from essential amino acids	Have no role in the metabolism	May be synthesized in the body in diseased states	**May be synthesized in the body from essential amino acids**
14	4	An example of polar amino acid is	Alanine	Leucine	Arginine	Valine	**Arginine**
15	4	The amino acid with a nonpolar side chain is	Serine	Valine	Asparagine	Threonine	**Asparagine**
16	4	A ketogenic amino acid is	Valine	Cysteine	Leucine	Threonine	**Cysteine**
17	4	An amino acid that does not form an α-helix is	Valine	Proline	Tyrosine	Tryptophan	**Proline**
18	4	An amino acid not found in proteins is	β-Alanine	Proline	Lysine	Histidine	**β-Alanine**
19	4	In mammalian tissues serine can be a biosynthetic precursor of	Methionine	Glycine	Tryptophan	Phenylalanine	**Glycine**
20	4	A vasodilating compound is produced by the decarboxylation of the amino acid:	Arginine	Aspartic acid	Glutamine	Histidine	**Histidine**
21	4	Biuret reaction is specific for	–CONH-linkages	–CSNH2 group	–(NH)NH2 group	All of these	**–CONH-linkages**
22	4	Sakaguchi’s reaction is specific for	Tyrosine	Proline	Arginine	Cysteine	**Arginine**
23	4	Million-Nasse’s reaction is specific for the amino acid:	Tryptophan	Tyrosine	Phenylalanine	Arginine	**Tyrosine**
24	4	Ninhydrin with evolution of CO2 forms a blue complex with	Peptide bond	α -Amino acids	Serotonin	Histamine	**α -Amino acids**
25	4	Which of the following is a dipeptide?	Anserine	Glutathione	Glucagon	β -Lipoprotein	**Anserine**
26	4	Which of the following is a tripeptide?	Anserine	Oxytocin	Glutathione	Kallidin	**Glutathione**
27	4	Casein, the milk protein is	Nucleoprotein	Chromoprotein	Phosphoprotein	Glycoprotein	**Phosphoprotein**
28	4	An example of phosphoprotein present in egg yolk is	Ovoalbumin	Ovoglobulin	Ovovitellin	Avidin	**Ovovitellin**

29	4	A simple protein found in the nucleoproteins of the sperm is	Prolamine	Protamine	Glutelin	Globulin	**Protamine**
30	4	Histones are	Identical to protamine	Proteins rich in lysine and arginine	Proteins with high molecular weight	Insoluble in water and very dilute acids	**Proteins rich in lysine and arginine**
31	4	The protein present in hair is	Keratin	Elastin	Myosin	Tropocollagen	**Keratin**
32	4	Both α-helix and β-pleated sheet conformation of proteins were proposed by	Watson and Crick	Pauling and Corey	Waugh and King	Y.S.Rao	**Pauling and Corey**
33	4	Each turn of α-helix contains the amino acid residues (number):	3.6	3	4.2	4.5	**3.6**
34	4	Distance traveled per turn of α−helix in nm is	0.53	0.54	0.44	0.48	**0.54**
35	4	Along the α-helix each amino acid residue advances in nm by	0.15	0.1	0.12	0.2	**0.15**
36	4	The number of helices present in a collagen molecule is	1	2	3	4	**3**
37	4	In proteins the α-helix and β-pleated sheet are examples of	Primary structure	Secondary structure	Tertiary structure	Quaternary structure	**Secondary structure**
38	4	The α-helix of proteins is	A pleated structure	Made periodic by disulphide bridges	A non-periodic structure	Stabilised by hydrogen bonds between NH and CO groups of the main chain	**A non-periodic structure**
39	4	Tertiary structure of a protein describes	The order of amino acids	Location of disulphide bonds	Loop regions of proteins	The ways of protein folding	**The ways of protein folding**
40	4	In a protein molecule the disulphide bond is not broken by	Reduction	Oxidation	Denaturation	X-ray diffraction	**Denaturation**
41	4	Denaturation of proteins results in	Disruption of primary structure	Breakdown of peptide bonds	Destruction of hydrogen bonds	Irreversible changes in the molecule	**Destruction of hydrogen bonds**

42	4	The enzyme trypsin is specific for peptide bonds of	Basic amino acids	Acidic amino acids	Aromatic amino acids	Next to small amino acid residues	**Basic amino acids**
43	4	Chymotrypsin is specific for peptide bonds containing	Uncharged amino acid residues	Acidic amino acids	Basic amino acid	Small amino acid residues	**Uncharged amino acid residues**
44	4	The end product of protein digestion in G.I.T. IS	Dipeptide	Tripeptide	Polypeptide	Amino acid	**Amino acid**
45	4	At isoelectric pH, an amino acid exists as	Anion	Cation	Zwitterion	None of these	**Zwitterion**
46	4	At a pH below the isoelectric point, an amino acid exists as	Cation	Anion	Zwitterion	Undissociated molecule	**Cation**
47	4	An amino acid having a hydrophilic side chain is	Alanine	Proline	Methionine	Serine	**Serine**
48	4	An amino acid that does not take part in α helix formation is	Histidine	Tyrosine	Proline	Tryptophan	**Proline**
49	4	Primary structure of a protein is formed by	Hydrogen bonds	Peptide bonds	Disulphide bonds	All of these	**Peptide bonds**
50	4	α-Helix is formed by	Hydrogen bonds	Hydrophobic bonds	Electrostatic bonds	Disulphide bonds	**Hydrogen bonds**
51	4	Aromatic amino acids can be detected by	Sakaguchi reaction	Millon-Nasse reaction	Hopkins-Cole reaction	Xanthoproteic reaction	**Xanthoproteic reaction**
52	4	Two amino groups are present in	Leucine	Glutamate	Lysine	Threonine	**Lysine**
53	4	During denaturation of proteins, all of the following are disrupted except	Primary structure	Secondary structure	Tertiary structure	Quaternary structure	**Secondary structure**
54	4	All the following are branched chain amino acids except	Isoleucine	Alanine	Leucine	Valine	**Alanine**
55	4	Millon’s test is for identification of	Tyrosine	Tryptophan	Proline	Arginine	**Tyrosine**
56	4	Hopkins-Cole test is for identification of	Tyrosine	Tryptophan	Arginine	Cysteine	**Tryptophan**
57	4	Collagen is very rich in	Glycine	Serine	Aspartic acid	Glutamic acid	**Glycine**
58	4	In glutathione (a tripeptide) is present apart from Glutamic acid and cysteine:	Serine	Glycine	Leucine	Phenyl alanine	**Glycine**
59	4	2-Amino 3-OH propanoic acid is	Glycine	Alanine	Valine	Serine	**Serine**

60	4	All amino acids have one asymmetric carbon atom, except	Arginine	Aspargine	Histidine	Glycine	**Glycine**
61	4	A Holoenzyme is	Functional unit	Apo enzyme	Coenzyme	All of these	**All of these**
62	4	Example of an extracellular enzyme is	Lactate dehydrogenase	Cytochrome oxidase	Pancreatic lipase	Hexokinase	**Pancreatic lipase**
63	4	Enzymes, which are produced in inactive form in the living cells, are called	Papain	Lysozymes	Apoenzymes	Proenzymes	**Proenzymes**
64	4	An example of ligases is	Succinate thiokinase	Alanine racemase	Fumarase	Aldolase	**Succinate thiokinase**
65	4	An example of lyases is	Glutamine synthetase	Fumarase	Cholinesterase	Amylase	**Fumarase**
66	4	Activation or inactivation of certain key regulatory enzymes is accomplished by covalent modification of the amino acid:	Tyrosine	Phenylalanine	Lysine	Serine	**Serine**
67	4	The enzyme which can add water to a carbon-carbon double bond or remove water to create a double bond without breaking the bond is	Hydratase	Hydroxylase	Hydrolase	Esterase	**Hydratase**
68	4	Fischer’s ‘lock and key’ model of the enzyme action implies that	The active site is complementary in shape to that of substance only after interaction.	The active site is complementary in shape to that of substance	Substrates change conformation prior to active site interaction	The active site is flexible and adjusts to substrate	**The active site is complementary in shape to that of substance**
69	4	From the Lineweaver-Burk plot of Michaelis-Menten equation, Km and Vmax can be determined when V is the reaction velocity at substrate concentration S, the X- expressed as axis experimental data are	1/V	V	1/S	S	**1/S**
70	4	A sigmoidal plot of substrate concentration	Michaelis-	Co-operative	Competitive	Non-	**Co-operative**

		([S] verses reaction velocity (V) may indicate	Menten kinetics	binding	inhibition	competitive inhibition	**binding**
71	4	The kinetic effect of purely competitive inhibitor of an enzyme	Increases Km without affecting Vmax	Decreases Km without affecting Vmax	Increases Vmax without affecting Km	Decreases Vmax without affecting Km	**Increases Km without affecting Vmax**
72	4	An inducer is absent in the type of enzyme:	Allosteric enzyme	Constitutive enzyme	Co-operative enzyme	Isoenzymic enzyme	**Constitutive enzyme**
73	4	In reversible non-competitive enzyme activity inhibition	Vmax is increased	Km is increased	Km is decreased	Concentration of active enzyme is reduced	**Concentration of active enzyme is reduced**
74	4	In competitive enzyme activity inhibition	The structure of inhibitor generally resembles that of the substrate	Inhibitor decreases apparent Km	Km remains unaffective	Inhibitor decreases Vmax without affecting Km	**The structure of inhibitor generally resembles that of the substrate**
75	4	In enzyme kinetics Vmax reflects	The amount of an active enzyme	Substrate concentration	Half the substrate concentration	Enzyme substrate complex	**The amount of an active enzyme**
76	4	In enzyme kinetics Km implies	The substrate concentration that gives one half Vmax	The dissocation constant for the enzyme substrate comples	Concentration of enzyme	Half of the substrate concentration required to achieve Vmax	**The substrate concentration that gives one half Vmax**
77	4	In a competitive enzyme activity inhibition	Apparent Km is decreased	Apparent Km is increased	Vmax is increased	Vmax is decreased	**Apparent Km is increased**
78	4	In non competitive enzyme activity inhibition, inhibitor	Increases Km	Decreases Km	Does not effect Km	Increases Km	**Does not effect Km**
79	4	The pH optima of most of the enzymes is	Between 2 and 4	Between 5 and 9	Between 8 and 12	Above 12	**Between 5 and 9**

80	4	Coenzymes are	Heat stable, dialyzable, non protein organic molecules	Soluble, colloidal, protein molecules	Structural analogue of enzymes	Different forms of enzymes	**Heat stable, dialyzable, non protein organic molecules**
81	4	Factors affecting enzyme activity:	Concentration	pH	Temperature	All of these	**All of these**
82	4	The normal serum GOT activity ranges from	3.0–15.0 IU/L	4.0–17.0 IU/L	4.0–60.0 IU/L	0.9–4.0 IU/L	**4.0–17.0 IU/L**
83	4	The normal GPT activity ranges from	60.0–250.0 IU/L	4.0–17.0 IU/L	3.0–15.0 IU/L	0.1–14.0 IU/L	**3.0–15.0 IU/L**
84	4	The normal serum acid phosphatase activity ranges from	5.0–13.0 KA units/100 ml	1.0–5.0 KA units/100 ml	13.0–18.0 KA units/100 ml	0.2–0.8 KA units/100 ml	**1.0–5.0 KA units/100 ml**
85	4	The normal serum alkaline phosphatase activity ranges from	1.0–5.0 KA units/100 ml	5.0–13.0 KA units/100 ml	0.8–2.3 KA units/100 ml	13.0–21.0 KA units/100 ml	**5.0–13.0 KA units/100 ml**
86	4	The isoenzymes LDH5 is elevated in	Myocardial infarction	Peptic ulcer	Liver disease	Infectious diseases	**Liver disease**
87	4	LDH1 and LDH2 are elevated in	Myocardial infarction	Liver disease	Kidney disease	Brain disease	**Myocardial infarction**
88	4	The pH optima for salivary analyse is	6.6–6.8	2.0–7.5	7.9	8.6	**6.6–6.8**
89	4	The pH optima for pancreatic analyse is	4	7.1	7.9	8.6	**8.6**
90	4	The substrate for amylase is	Cane sugar	Starch	Lactose	Ribose	**Starch**
91	4	Vitamin A or retinal is a	Steroid	Polyisoprenoid compound containing a cyclohexenyl ring	Benzoquinone derivative	6-Hydroxychromane	**Polyisoprenoid compound containing a cyclohexenyl ring**
92	4	β-Carotene, precursor of vitamin A, is oxidatively cleaved by	β-Carotene dioxygenase	Oxygenase	Hydroxylase	Transferase	**β-Carotene dioxygenase**
	4	Preformed Vitamin A is supplied by	Milk, fat and liver	All yellow vegetables	All yellow fruits	Leafy green vegetables	**Milk, fat and liver**
93	4	Fat soluble vitamins are	Soluble in alcohol	one or more Propene units	Stored in liver	All these	**All these**
94	4	The normal serum concentration of vitamin A	5–10	15–60	100–150	0–5	**15–60**

		in mg/100 ml is					
95	4	One manifestation of vitamin A deficiency is	Painful joints	Night blindness	Loss of hair	Thickening of long bones	**Night blindness**
96	4	Deficiency of Vitamin A causes	Xeropthalmia	Hypoprothrombinemia	Megaloblastic anemia	Pernicious anemia	**Xeropthalmia**
97	4	An important function of vitamin A is	To act as coenzyme for a few enzymes	To play an integral role in protein synthesis	To prevent hemorrhages	To maintain the integrity of epithelial tissue	**To maintain the integrity of epithelial tissue**
98	4	Retinal is a component of	Iodopsin	Rhodopsin	Cardiolipin	Glycoproteins	**Rhodopsin**
99	4	Richest source of Vitamin D is	Fish liver oils	Margarine	Egg yolk	Butter	**Fish liver oils**
100	4	Deficiency of vitamin D causes	Ricket and osteomalacia	Tuberculosis of bone	Hypthyroidism	Skin cancer	**Ricket and osteomalacia**
101	4	Vitamin K2 was originally isolated from	Soyabean	Wheat gram	Alfa Alfa	Putrid fish meal	**Putrid fish meal**
102	4	Vitamin synthesized by bacterial in the intestine is	A	C	D	K	**K**
103	4	The most important natural antioxidant is	Vitamin D	Vitamin E	Vitamin B12	Vitamin K	**Vitamin E**
104	4	Creatinuria is caused due to the deficiency of vitamin	A	K	E	D	**E**
105	4	The daily requirement of riboflavin for adult in mg is	0–1.0	1.2–1.7	2.0–3.5	4.0–8.0	**1.2–1.7**
106	4	The precursor of CoA is	Riboflavin	Pyridoxamine	Thiamin	Pantothenate	**Pantothenate**
107	4	FAD is a coenzyme for	Succinate dehydrogenase	Glycerol-3-phosphate dehydrogenase	Sphingosine reductase	All of these	**All of these**
108	4	Niacin contains a	Sulphydryl group	Carboxyl group	Amide group	All of these	**Carboxyl group**
109	4	Vitamin B12 is	Not stored in the body	Stored in bone marrow	Stored in liver	Stored in RE cells	**Stored in liver**
110	4	Daily requirement of vitamin C in adults is about	100 mg	25 mg	70 mg	100 mg	**70 mg**

111	4	Precursor of Vitamin A is	α-Carotene	β-Carotene	γ-Carotene	All of these	**All of these**
112	4	Conversion of β-carotene into retinal requires the presence of	β-Carotene dioxygenase	Bile salts	Molecular oxygen	All of these	**All of these**
113	4	Provitamins A include	Retinal	Retionic acid	Carotenes	All of these	**Carotenes**
114	4	Provitamin D3 is	Cholecalciferol	Ergosterol	7-Dehydrocholesterol	Ergocaliferol	**7-Dehydrocholesterol**
115	4	Ergosterol is found in	Animals	Plants	Bacteria	All of these	**Plants**
116	4	A water soluble form of vitamin K is	Phylloquinone	Farnoquinone	Menadione	None of these	**Menadione**
117	4	The non-protein part of rhodopsin is	Retinal	Retinol	Carotene	Repsin	**Retinal**
118	4	Antisterility vitamin is	Vitamin B1	Vitamin B2	Vitamin E	Vitamin K	**Vitamin E**
119	4	The sulphur-containing vitamins among the following B-Vitamin is	Thiamine	Riboflavin	Niacin	Pyridoxine	**Thiamine**

UNIT-V

SYLLABUS

Nucleic acids - Purines & Pyrimidines nucleotides, RNA & DNA base pairing schemes, types of RNA: mRNA, tRNA, aminoacyl tRNA synthetase, Secondary structure of DNA, Watson and Crick model. Denaturation of DNA keto-enol tautomerism and consequences.

Nucleic acids, which are relatively strong acids found in the nuclei of cells, were first isolated in 1869. The nucleic acids are polymers with molecular weights as high as 100,000,000 grams per mole. They can be broken down, or digested, to form monomers known as nucleotides.

There are three basic components of Nucleic acids

a) Nitrogenous bases – Purines and Pyrimidines
b) Sugar Moiety
c) Phosphate Molecule

Nitrogenous Bases

The nitrogenous bases are the derivatives of two parent compounds. They are

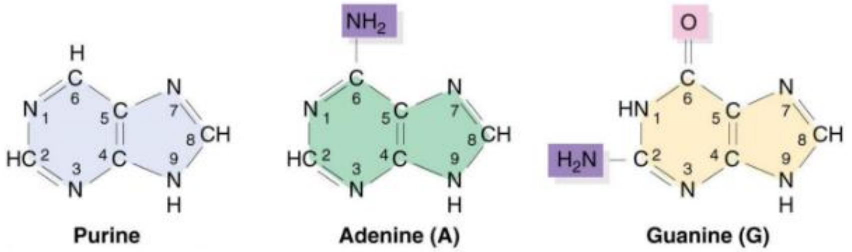
a) Purines
b) Pyrimidines

Purines

Purines bases are heterocyclic compounds consisting of a pyrimidines ring and an imidazole ring fused together. The two purine bases are

a) Adenine (6-amino purine): ($C_5H_5N_5$), found in both RNA and DNA, is a white crystalline purine base, with molecular weight 135.15 daltons and melting point 360 to 365 C.

b) Guanine (2-Amino-6-oxyPurine): ($C_5H_5ON_5$), also found in both DNA and RNA, is a colorless, insoluble crystalline substance, with MW=151.15 Daltons. It was first isolated from guano (bird manure), hence so named.

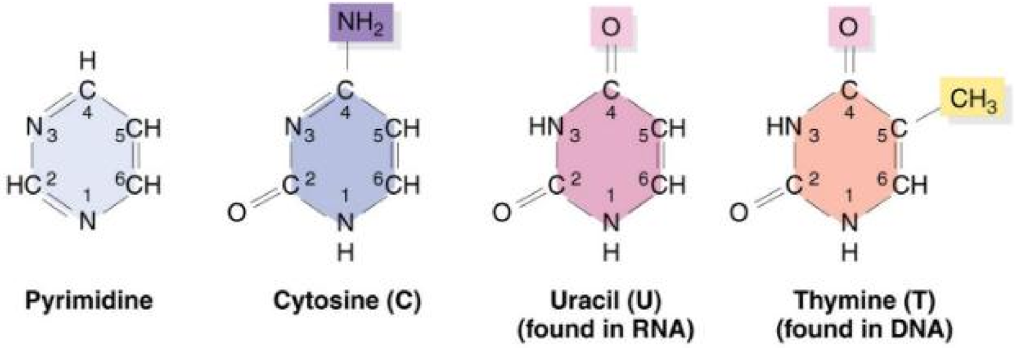


Pyrimidines: Pyrimidine bases consist of six membered ring with two nitrogen atoms. The pyrimidine bases are –

a) Cytosine (2-Oxy-4-amino pyrimidine): ($C_5H_6O2N_5$), found in both RNA and DNA, is a white crystalline substance, with MW=111.12 daltons and a melting point 320 to 325 C.

b) Thymine (2, 4-dioxy-5-methyl pyrimidine): ($C_5H_6O2N_2$), found in DNA molecules only, has MW=126.13 Daltons. It was first isolated from thymus, hence so named. Only rarely does thymine occur in RNA.

c) Uracil (2, 4-dioxy pyrimidine) ($C_4H_4O2N_2$), found in RNA molecules only, is a white, crystalline pyrimidine base with MW=112.10 daltons and a melting point 338 C. Only rarely does uracil occur in DNA.



Sugarmoiety

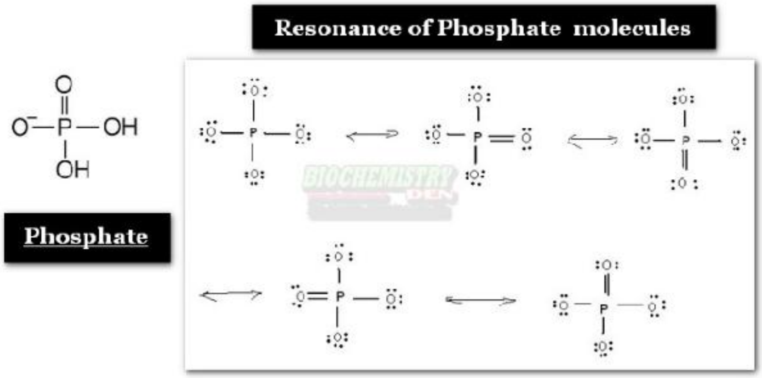
Pentose sugar is present in DNA & RNA. It is present in their "β-furanose" from (close five number ring) and of β-configuration. Two types of pentose sugars present in the nucleic acid.

a) Ribose (RNA)

b) 2-Deoxyribose (DNA)

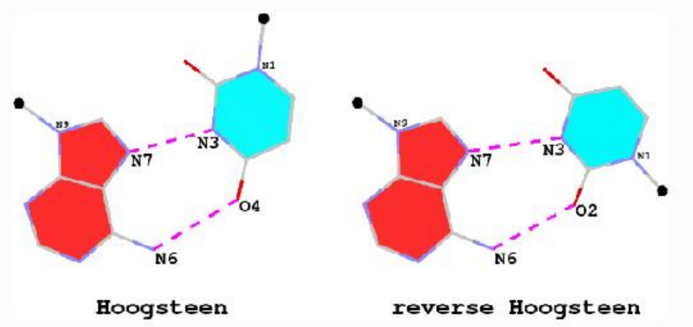
Phosphate

It contains the monovalent hydroxyl groups and one divalent oxygen atom all are linked to pentavalent phosphorous atom. The base is joined covalently (at N1 of pyrimidines and N9 of purines) and the phosphate is esterified to the 5’-carbon. The N-glycosyl bond is formed by removal of the elements of water (a Hydroxyl groups from pentose and Hydrogen atom from the base).



Hoogsteen and reverse Hoogsteen base pairs

The A·U (or A·T) Hoogsteen pair is a well-known type of base pair (bp), named after the scientist who discovered it. As shown in the Figure below (left), in the Hoogsteen bp scheme, adenine uses its N7 (acceptor) and N6 (donor) atoms at the major groove edge to form two H-bonds with the N3 (donor) and O4 (acceptor) atoms from uracil, respectively. Interestingly, if the uracil base ring is flipped around the N7(A)…N3(U) H-bond by 180 degrees, N6(A) now forms an H-bond with O2(U), i.e., N6(A)…O2(U): this pairing scheme is called the reverse Hoogsteen bp (right).



Types of RNA

The process of transcription creates RNA using a DNA template. First, the double-stranded DNA separates into its component strands. Proteins called promoters bind to the strand of DNA and function at strategic locations known as promoter sequences. The enzyme RNA polymerase attaches to the promoter-DNA complex and unwinds the DNA.

DNA contains the four base pairs adenine, cytosine, guanine and thymine; RNA contains the first three but substitute’s uracil in place of thymine. A single strand of DNA can serve as a template for countless identical molecules of RNA, using multiple copies of DNA polymerase. A termination sequence on the DNA strand indicates the stopping point for transcription.

Messenger RNA

Messenger ribonucleic acids (mRNAs) transfer the information from DNA to the cell machinery that makes proteins. Tightly packed into every cell nucleus, which measures just 10 microns in diameter, is a three-meter long double-stranded DNA “instruction manual” on how to build and maintain a human body. In order for each cell to maintain its structure and perform all of its functions, it must continuously manufacture cell-type specific parts (proteins). Inside each nucleus, a multi subunit protein called RNA polymerase II (RNAP II) reads DNA and simultaneously fabricates a “message” or transcript, which is called messenger RNA (mRNA), in a process called transcription. Molecules of mRNA are composed of relatively short, single strands of molecules made up of adenine, cytosine, guanine and uracil bases held together by a sugar phosphate backbone. When RNA polymerase finishes reading a section of the DNA, the pre-mRNA copy is processed to form mature mRNA and then transferred out of the cell nucleus. Ribosomes read the mRNA and translate the message into functional proteins in a process called translation. Depending on the newly synthesized protein's structure and function, it will be further modified by the cell, exported to the extra-cellular space, or will remain inside the cell. The diagram below shows transcription (DNA->RNA) taking place in the cell nucleus where RNAP is RNA polymerase II enzyme synthesizing RNA.

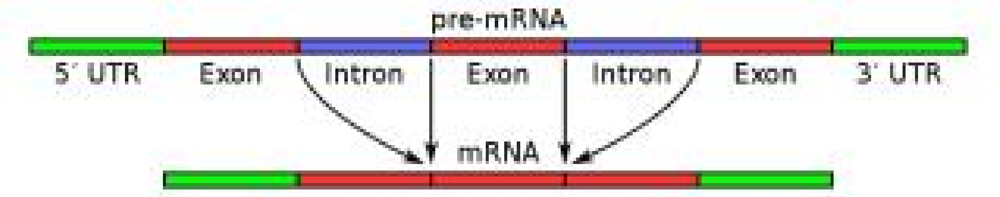
Precursor mRNA contains introns and exons. Introns are removed before translation, while exons code for the amino acid sequence of proteins. To make mature mRNA, the cell machinery removes "non-translatable" introns from the pre-mRNA, leaving only translatable exon sequences in the mRNA.

Types of mRNA

Pre-mRNA and hnRNA

Precursor mRNA (pre-mRNA) is the primary transcript of eukaryotic mRNA as it comes off the DNA template. Pre-mRNA is part of a group of RNAs called heterogeneous nuclear RNA (hnRNA). hnRNA refers to all single strand RNA located inside the nucleus of the cell where transcription takes place (DNA->RNA) and pre-mRNA form a large part of these ribonucleic acids. Pre-mRNA contains sequences that need to be removed or "spliced out" before being translated into a protein. These sequences can either be removed through the catalytic activity of the RNA itself, or through the action of a multi-protein structure called spliceosome. After this processing step, the pre-mRNA is considered as a mature mRNA transcript.

The diagram below describes the structure of pre-mRNA. Pre-mRNA includes introns and may or may not include the 5' cap and poly-adenylated 3' tail:



Monocistronic mRNA

A monocistronic mRNA molecule contains the exon sequences coding for a single protein. Most eukaryotic mRNAs are monocistronic.

Bicistronic mRNA

A bicistronic mRNA molecule contains the exon coding sequences for two proteins.

Polycistronic mRNA

A polycistronic mRNA molecule contains the exon coding sequences for multiple proteins. Most mRNA of bacteria and bacteriophages (viruses that live in bacterial hosts) are polycistronic.

Prokaryotic vs. Eukaryotic mRNA

Polycistronic prokaryotic mRNAs contain multiple sites for initiating and terminating protein synthesis. Eukaryotes have just one site for translation initiation and eukaryotic mRNAs are primarily monocistronic. Prokaryotes lack organelles and a well defined nuclear envelope, and therefore mRNA translation can be coupled with mRNA transcription in the cytoplasm. In eukaryotes, mRNA is transcribed on chromosomes in the nucleus, and after processing, is shuttled through nuclear pores and into the cytoplasm. Unlike prokaryotes, translation in eukaryotes takes place only after transcription has been completed. Prokaryotic mRNA is constantly degraded by ribonucleases, enzymes that cut RNA. For example, the half-life of mRNA in E. coli is approximately two minutes. Bacterial mRNAs are short-lived to allow for flexibility in adjusting to rapidly changing environmental conditions. Eukaryotic mRNAs are more metabolically stable. For example, precursors of mammalian red blood cells (reticulocytes), which have lost their nuclei, synthesize hemoglobin for several days by translating mRNAs that were transcribed when the nucleus was still present. Lastly, the mRNAs of prokaryotes undergo minimal processing. In eukaryotes, the pre-mRNA must undergo processing before being translated, involving the removal of introns, the addition of the 5' –cap as well as the 3' poly-adenylated tail before mature mRNA is formed and ready to be translated.

Functions of mRNA

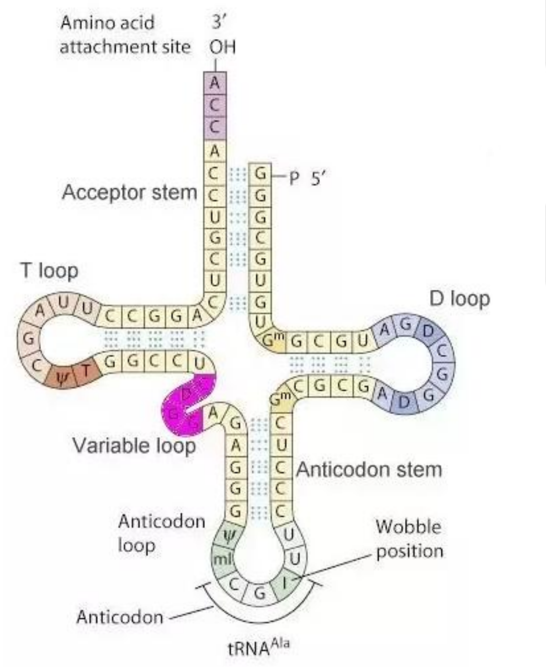
The primary function of mRNA is to act as an intermediary between the genetic information in DNA and the amino acid sequence of proteins. mRNA contains codons that are complementary to the sequence of nucleotides on the template DNA and direct the formation of amino acids through the action of ribosomes and tRNA. mRNA also contains multiple regulatory regions that can determine the timing and rate of translation. In addition, it ensures that translation proceeds in an orderly fashion because it contains sites for the docking of ribosomes, tRNA as well as various helper proteins.

Proteins produced by the cells play a variety of roles, either as enzymes, structural molecules or as transport machinery for various cellular components. Some cells are also specialized for secreting proteins, such as the glands that produce digestive enzymes or hormones which influence the metabolism of the entire organism.

mRNA Translation

mRNA can be translated on free ribosomes in the cytoplasm with the help of transfer RNA (tRNA) molecules and multiple proteins called initiation, elongation and termination factors. Proteins that are synthesized on free ribosomes in the cytoplasm are often used by the cell in the cytoplasm itself or targeted for use inside intracellular organelles. Alternatively, proteins that have to be secreted begin to be translated in the cytoplasm but as soon as the first few residues are translated, specific proteins transport the entire translation machinery to the membrane of the endoplasmic reticulum (ER). The initial few amino acids get embedded in the ER membrane and the rest of the protein is released into the interior space of the ER. The short sequence is removed from proteins that need to be secreted from the cell, while those intended for internal membranes retain that short stretch providing a membrane anchor.

tRNA or transfer RNA is used during translation to add the amino acids to the growing polypeptide being made by the ribosome during the production of new protein. tRNA has a hairpin loop structure shaped like a lower case t. tRNA also possess an anti-codon at the bottom of the looping structure that helps it match to the correct codon presented by the mRNA transcript acting as the "instruction manual" within the ribosome during translation. I've inserted an image below to help illustrate my explanation.



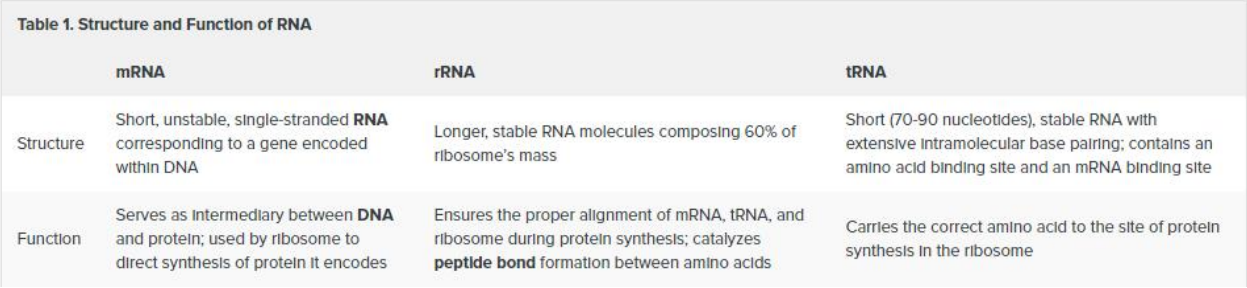


Table 1. Structure and Function of RNA

	mRNA	rRNA	tRNA
Structure	Short, unstable, single-stranded **RNA** corresponding to a gene encoded within DNA	Longer, stable RNA molecules composing 60% of ribosome's mass	Short (70-90 nucleotides), stable RNA with extensive intramolecular base pairing; contains an amino acid binding site and an mRNA binding site
Function	Serves as intermediary between **DNA** and protein; used by ribosome to direct synthesis of protein it encodes	Ensures the proper alignment of mRNA, tRNA, and ribosome during protein synthesis; catalyzes **peptide bond** formation between amino acids	Carries the correct amino acid to the site of protein synthesis in the ribosome

DNA —The Thread of Life: Watson-Crick Model, Characteristics

DNA is the largest macromolecule that represents the genetic material of the cell. Chemically, DNA is a double helix of two antiparallel polynucleotide chains. Each polynucleotide chain is a linear mixed polymer of four deoxyribotides i.e. deoxyadenylate, deoxyguanylate, deoxycytidylate and thymidylate.

Watson-Crick Model of DNA:

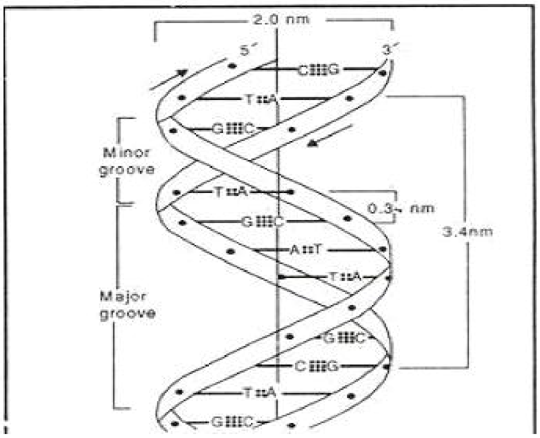
In 1953, J.D. Watson (an American biologist) and F.H.C. Crick (a British Physicist) proposed the three-dimensional model of physiological DNA (i. e B-DNA) on the basis of X-ray diffraction data of DNA obtained by Franklin and Wilkins. For this epoch-making discovery, Watson, Crick and Wilkins got Nobel Prize in medicine in 1962. Term DNA was given by Zaccharis.

The important features of Watson – Crick Model or double helix model of DNA are as follows:

1. The DNA molecule consists of two polynucleotide chains or strands that spirally twisted around each other and coiled around a common axis to form a right-handed double-helix.

2. The two strands are antiparallel i.e. they ran in opposite directions so that the 3′ end of one chain facing the 5′ end of the other.

3. The sugar-phosphate backbones remain on the outside, while the core of the helix contains the purine and pyrimidine bases.



4. The two strands are held together by hydrogen bonds between the purine and pyrimidine bases of the opposite strands.

5. Adenine (A) always pairs with thymine (T) by two hydrogen bonds and guanine (G) always pairs with cytosine (C) by three hydrogen bonds. This complimentarily is known as the base pairing rule. Thus, the two stands are complementary to one another.

6. The base sequence along a polynucleotide chain is variable and a specific sequence of bases carries the genetic information.

7. The base compositions of DNA obey Chargaff s rules (E.E. Chargaff, 1950) according to which A=T and G=C; as a corollary ∑ purines (A+G) = 2 pyrimidines (C+T); also (A+C) = (G+T). It also states that ratio of (A+T) and (G+C) is constant for a species (range 0.4 to 1.9) DNA. A Hypothetically untwisted DNA

8. The diameter of DNA is 20nm or 20 A. Adjacent bases are separated 0.34 nm or by 3.4 A along the axis. The length of a complete turn of helix is 3.4 nm or 34 A i.e. there are 10bp per turn. (B- DNA-Watson rick DNA)

9. The DNA helix has a shallow groove called minor groove (-1,2nm) and a deep groove called major groove (-2.2nm) across.

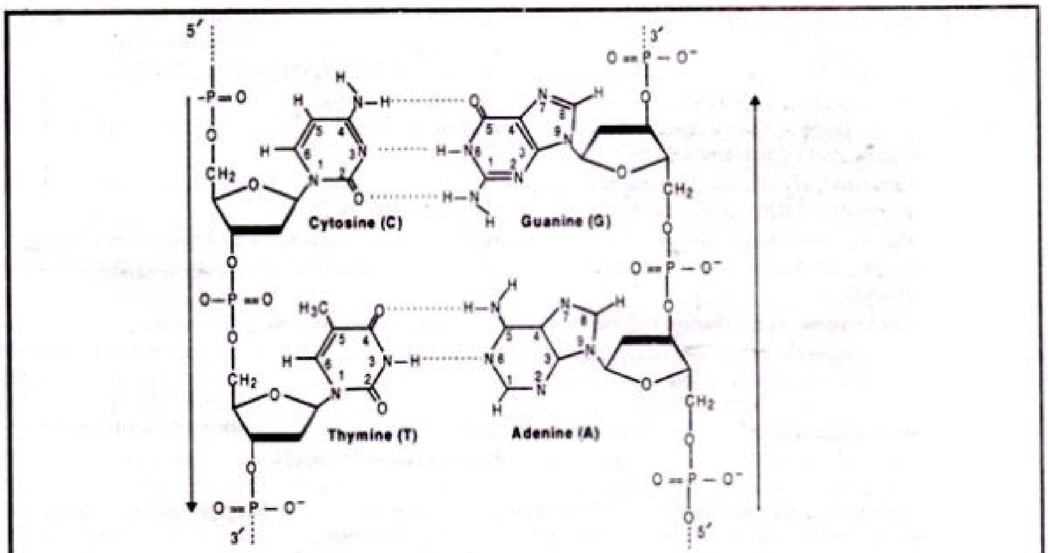


Figure 4.29 A part of DNA showing complementary base pairing. Three hydrogen bonds link cytosine & guanine, whereas two hydrogen bonds form between thymine and adenine.

Some other characteristics of DNA:

1. The amount of DNA per nucleus is constant in all the somatic cells of a given species.

2. The total amount of DNA in a haploid genome is a characteristic of each organism and is known as C-value.

3. Only a small fraction of DNA is functional in eukaryotes.

4. DNA is the chemical basis of heredity and is organized into genes or cistrons.

5. DNA replicates to form DNAs and transcribes to form RNAs.

6. DNA replication occurs in the S-phase of cell cycle.

7. DNA replication is semi conservative in which two daughter DNA molecules formed; each receives one of parental strand and one new strand.

8. One strand of DNA directs the synthesis called template strand, or antisense or non-coding strand. The other strand is called coding or non template of sense strand which has the same sequence as the RNA transcript except for T in place of U.

9. DNA has many repeated base sequences, some of which ire mobile.

10. DNA can easily undergo denaturation (melting) and renaturation with any change in pH, temperature and salt concentration. DNA with a high G+ G content are more resistant to thermal melting than A + T rich molecules.

11. DNA can be synthesized in vitro (in the laboratory).

12. DNA can be measured by the unit picogram (lpg= 10 g)

Biological Importance of DNA:

1. Hereditary material:

The genetic information stored in the nucleotide sequence of DNA helps in synthesis of specific proteins or polypeptides and transmit the information to daughter cells or offsprings. Thus, DNA is called as molecular blueprint or thread of life.

2. Autocatalytic role DNA:

DNA undergoes replication (self-duplication) in the S-phase of cell cycle. During the process each DNA strand of a double helix can act as template for the synthesis of daughter strand.

3. Heterocatalytic role:

During transcription any one strand of DNA acts as template for the synthesis of RNA. This is called the heterocatalytic role of RNA.

4. Variations:

DNA undergoes recombination its meiosis and occasional mutation (changes in nucleotide sequences) which creates variations in population and ultimately contributes to evolution.

5. DNA controls cellular metabolism, growth, and differentiation.

6. DNA finger printing (-DNA typing or profiling):

Each individual carries specific short nucleotide repeats which are called as minisatellites or VNTRs (Variable Number of Tandem Repeats). The VNTRs of two individuals are variable and forms the basis of DNA fingerprinting This technique is used to identify criminals, determine paternity, verification of immigrant etc.

7. Recombinant DNA technology (Genetic engineering):

It involves the artificial cleaving and rejoining DNA sequences from two or more organisms to create recombinant DNA. This technology is employed for production of genetically modified organisms (GMOs), genetically modified foods (GMFs), vaccines, hormones, enzymes, clones etc. It is also used for construction of probes (short polynucleotide chain attached to a radioactive or fluorescent marker) for diagnosis of diseases und curing hereditary diseases by replacing a faulty gene with a normal gene (gene therapy), formation of clones etc.

Denaturation of DNA:

Denaturation of DNA double helix takes place by the following denaturating agents:

(i) Denaturation by Temperature:

If a DNA solution is heated to approximately 90°C or above there will be enough kinetic energy to denature the DNA completely causing it to separate into single strands. This denaturation is very abrupt and is accelerated by chemical reagents like urea and formamide.

The chemicals enhance the aqueous solubility of the purine and pyrimidine groups. This separation of double helix is called melting as it occurs abruptly at a certain characteristic temperature called denaturation temperature or melting temperature (Tm).

It is defined as temperature at which 50% of the DNA is melted. The abruptness of the transition indicates that the DNA double helix is highly cooperative structure, held together by many reinforcing bonds. The melting of DNA can be followed spectrophotometrically by monitoring the absorbance of DNA at 260 nm. Tm is analogous to the melting point of crystal. The Tm value depends on the nature of the DNA.

If several samples of DNA are melted, it is found that the Tm is highest for those DNAs that contain the highest proportion of G—C. Actually the value is used to estimate the percentage of G—C in a DNA sample. In fact, the Tm of DNA from many species varies linearly with G—C content.

This relationship between Tm and G—C content arises due to guanine and cytosine form three hydrogen bonds when base paired, whereas adenine and thymine form only two.

Denaturation involves the following changes of the properties of DNA:

(a) Increase in Absorption of UV-Light:

If denaturation is followed spectrophotometrically by monitoring the absorbance of light at 260 nm, it is observed that the absorbance at 260 nm increases as the DNA become denatured, a phenomenon known as the hyperchromatic effect or hyperchromacity or hyperchromism. This is due to un-stacking of base pairs.

A plot of the absorbance at 260 nm against the temperature of a DNA solution indicates that little denaturation occurs below approximately 70°C, but further increases in temperature result in a marked increase in the extent of denaturation.

(b) Decrease in Specific Optical Rotation:

Double-stranded DNA shows a strong positive rotation which highly decreases with denaturation. This change is analogous to the change in rotation observed when the proteins are denatured.

(c) Decrease in Viscosity:

The solutions of native DNA exhibit high viscosity because of the relatively rigid double helical, long and rod like character of DNA molecule. Denaturation causes a marked decrease in viscosity.

Melting curve of DNA

If melted DNA is cooled it is possible to reassociate the separated strands, a process known as renaturation. However, a stable double-stranded molecule may be formed only if the complementary strands collide in such a way that their bases are paired precisely. But renaturation may not be precise if the DNA is very long and complex.

Thus the rate of renaturation (renaturation kinetics) can give information about the complexity of a DNA molecule. Complete denaturation is not a readily reversible process. If a heat-denatured DNA solution is cooled slowly (anneling) and hold the solution at about 25°C below Tm and above a concentration of 0.4M Na+ for several hours, some amount of

DNA (50-60%) is renatured. Rapid cooling does not reverse denaturation, but if the cooled solution is again heated and then cooled slowly, renaturation takes place.

(ii) Denaturation by Chemical Agents:

Denaturation of DNA double helix can also be brought about by certain chemical agents such as urea and formamide. These chemical reagents enhance the aqueous solubility of the purine and pyrimidine groups. The Tm value is lowered by the addition of urea. In 8M urea, Tm is decreased by nearly 20°C. DNA can be completely denatured by 95% formamide at room temperature only.

(iii) Effect of pH on Denaturation:

Denaturation also occurs at acidic and alkaline solutions in which ionic changes of the purine and pyrimidine bases can occur. In acidic solutions at pH values 2-3 the amino groups bind with protons and the DNA double helix is disrupted. Similarly, in alkaline solutions at pH 12, the enolic hydroxyl groups ionize, thus preventing the keto-amino hydrogen bonding.

Renaturation of DNA:

When preparations of double-stranded DNA are denatured and allowed to renature, the rate of renaturation can give valuable information about the complexity of the DNA if there are repetitive sequences in the DNA, it shows less complexity in comparison to its total length, but the complexity is equal to its total length if all sequences are unique.

The 1kb DNA fragments are denatured by heating above its Tm and then renatured at a temperature 10°C below the Tm and monitored either by decrease in absorbance at 260 nm (hypochromic effect), or by passing samples at intervals through a column of hydroxylapatite, which retains only double stranded DNAs, and estimating how much of the sample is retained.

The degree of renaturation after a given time depends on C0, the concentration of double stranded DNA prior to denaturation, and t, the duration of the renaturation in seconds. The concentration is measured in nucleotides per unit volume. In order to compare the rates of renaturation of different samples of DNA it is usual to measure C0 and the time taken for renaturation to proceed half way to completion, t1/2, and to multiply these values together to give a C0t1/2 value. The larger the C0t1/2, the greater the complexity of the DNA; hence λ DNA has a far lower C0t1/2 than does human DNA.

if the extent of renaturation is plotted against log C0t (known as Cot curve), it is observed that part of the DNA is renatured quite rapidly while the rest is very slow to renature. This indicates that some sequences have a higher concentration than others i.e., part of the genome consists of repetitive sequences.

These repetitive sequences can be separated from the single-copy unique DNA by passing the renaturating sample through a hydroxylapatite column early in the renaturation process, at a time which gives a low value of C0t. At this stage only the rapidly renaturating sequences will be double stranded, and will, therefore, bind to the column.

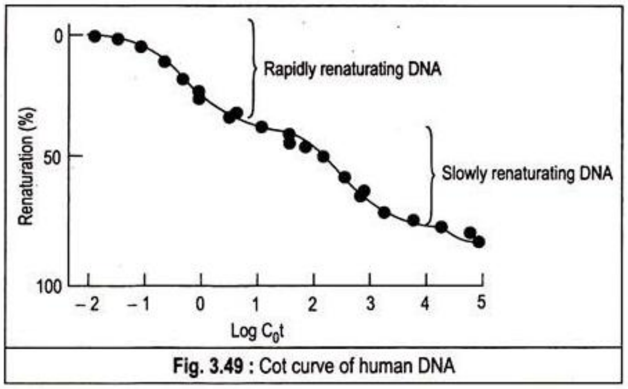
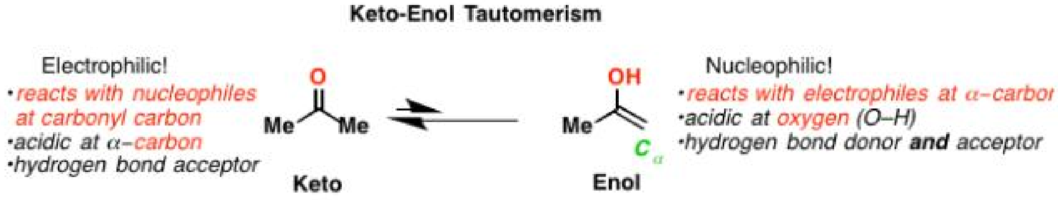


Fig. 3.49 : Cot curve of human DNA

Keto-enol tautomerism

Aldehydes and ketones are somewhat lycanthropic chemical species. Take acetone. It behaves as a garden-variety polar aprotic solvent, which makes it a useful medium for SN2 reactions; it reacts readily with nucleophiles like enolates, Grignards, and amines; and is several pKa units less acidic than alcohols (~20 vs. 16). This chemical behavior reflects the fact that it spends the vast majority of its time as a ketone, with an electrophilic carbonyl carbon. It's nice and stable. You use it to wash glassware with, or as paint thinner.



- *Equilibrium between isomers* (not resonance)
- Under most conditions, keto form is favored (6600:1 for acetone)
- *Important for aldehydes, ketones*, but not so much for carboxylic acids, esters and amides under normal conditions (>10 million : 1 keto: enol)

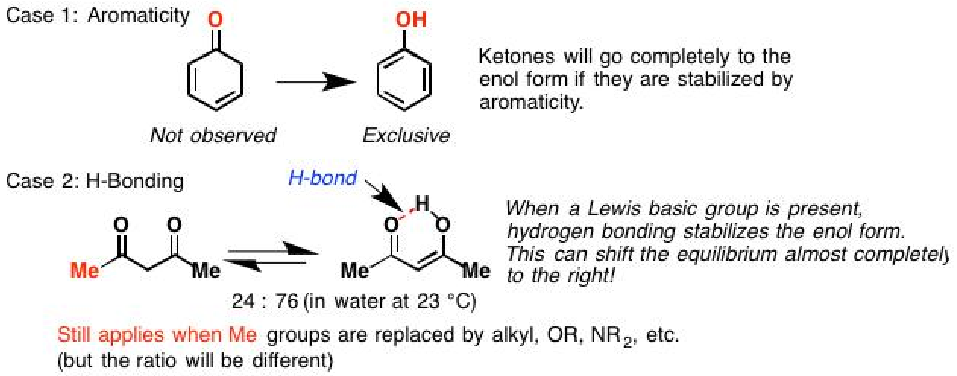
But every couple of blue moons (for acetone in water about 1/6600 th of the time at 23 °C) acetone undergoes a transformation to its alter ego, the enol form. And as its name suggests, the enol form – which is an isomer, not a resonance form – has the characteristics of both alkenes and alcohols: it can involve itself in hydrogen bonding via the OH group, it is acidic at oxygen, and it reacts with electrophiles (like aldehydes, for example, in the Aldol reaction). In short, the enol form differs from the keto form in its polarity, acidity, and nucleophilicity just like werewolves differ from ordinary folks in their copious body hair, nocturnal rambunctiousness, and peculiar dietary habits.

The reason for the equilibrium lying to the left is due to bond energies. The keto form has a C–H, C–C, and C=O bond whereas the enol has a C=C, C–O an O–H bond. The sum of the first three is about 359 kcal/mol (1500 kJ/mol) and the second three is 347 kcal/mol (1452 kJ/mol). The keto form is therefore more thermodynamically stable by 12 kcal/mol (48 kJ/mol).

Although the keto form is most stable for aldehydes and ketones in most situations, there are several factors that will shift the equilibrium toward the enol form. The same factors that stabilize alkenes or alcohols will also stabilize the enol form. There are two strong factors and three subtle factors.

1. Aromaticity. Phenols can theoretically exist in their keto forms, but the enol form is greatly favored due to aromatic stabilization.

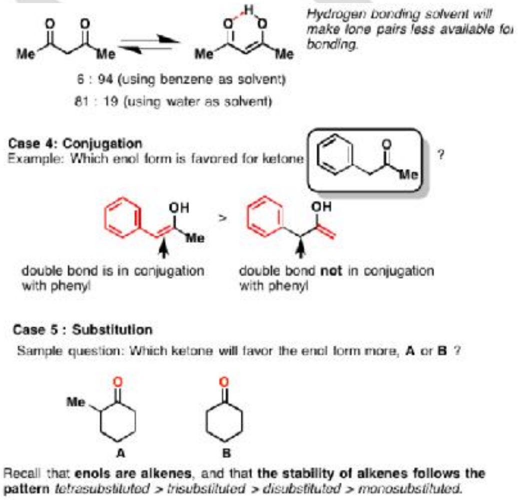
2. Hydrogen Bonding. Nearby hydrogen bond acceptors stabilize the enol form. When a Lewis basic group is nearby, the enol form is stabilized by internal hydrogen bonding.



3. Solvent. Solvent can also play an important role in the relative stability of the enol form. For example, in benzene, the enol form of 2,4-pentanedione predominates in a 94:6 ratio over the keto form, whereas the numbers almost reverse completely in water. What's going on? In a polar protic solvent like water, the lone pairs will be involved in hydrogen bonding with the solvent, making them less available to hydrogen bond with the enol form.

4. Conjugation. π systems are a little like Cheerios in milk: given the choice, they want to connect together than hang out in isolation. So in the molecule depicted, the more favorable tautomer will be the one on the left, where the double bond is a connected by conjugation to the phenyl.

5. Substitution. In the absence of steric factors, increasing substitution at carbon will stabilize the enol form. Enols are alkenes too – so any factors that stabilize alkenes, will stabilize enols as well. All else being equal, double bonds increase in thermodynamic stability as substitution is increased. So in the above example, the enol on the left should be the more stable one. As you might suspect, "all things being equal" sounds like a big caveat. It is – all else is rarely equal. But that's a topic for another day – or, more likely, another course.



Hydrogen bonding solvent will make lone pairs less available for bonding.

6 : 94 (using benzene as solvent)
81 : 19 (using water as solvent)

Case 4: Conjugation
Example: Which enol form is favored for ketone ?

double bond is in conjugation with phenyl

double bond **not** in conjugation with phenyl

Case 5 : Substitution
Sample question: Which ketone will favor the enol form more, **A** or **B** ?

Recall that **enols are alkenes**, and that **the stability of alkenes follows the pattern** *tetrasubstituted > trisubstituted > disubstituted > monosubstituted.*

Karpagam Academy of Higher Education
Department of Biochemistry
I B.Sc., Microbiology
18MBU103 – Biochemistry

Question number	**Unit**	**Question**	**Option I**	**Option II**	**Option III**	**Option IV**	**Answer**
1	5	The backbone of nucliec acid structure is contributed by	Hydrophobic forces	hydrogen bonds	phosphodiester linkages	ionic bonds	**phosphodiester linkages**
2	5	The pyrimidine base of the DNA is	cytosine	Guanine	Uracil	Adenine	**cytosine**
3	5	There are three hydrogen bonds between Cytosine and	Adenine	guanine	cytosine	thymine	**thymine**
4	5	An increased melting temperature for duplex DNA results from a high content of	Adenine & Guanine	Cytosine &thymine	Adenine &thymine	Cytosine &guanine	**Cytosine &guanine**
5	5	The base which is absent in RNA is	Cytosine	Uracil	thymine	Adenine	**thymine**
6	5	The following facts are true of all transfer (t)RNA except that	the 5' end is phosphorylated	they are single chains	methylated bases are found	the anticodon loop is identical	**the anticodon loop is identical**
7	5	Z-DNA was discovered by	Watson & Crick	Hoogsten	Chargaff	warg &Rich	**Hoogsten**
8	5	The base sequence at the end of all tRNA is	CCA	CAA	CCC	AAA	**CCA**
9	5	Deoxy ribose has no oxygen in	5' position	2' position	3' position	4' position	**2' position**
10	5	Among the RNA's which of the	rRNA	tRNA	mRNA	5SRNA	**mRNA**

		following is very unstable					
	5	Ribose is linked with purine by	C_1 to N_1	C_1 to N_9	C_5 to N_9	C_1 to N_7	**C_1 to N_9**
12	5	mRNA has complementary sequence of	C-DNA	t-RNA	RNA	r-RNA	**t-RNA**
13	5	The higher percentage of RNA found is	Nucleus	Mitochondria	microsomes	golgibodies	**Nucleus**
14	5	RNA is easily hydrolysed by base due to	presence of OH group in position 2 of the ribose	Differences in the bases	Low molecular weight	both a and b	**presence of OH group in position 2 of the ribose**
15	5	The best role of purine & pyrimidine nucleotides is to serve as the monomeric precursors of	RNA	DNA	Both	all of the above	**Both**
16	5	Melting temperature or Tm is high in DNA containing	rich in A, T pair	rich in G,C pair	Both in equal ratio	All	**rich in G,C pair**
17	5	The purine nucleotides act as the components of	FAD+	NAD+	NADP+	All	
18	5	When pancreatic deoxyribonuclease attacks DNA, the product obtained is	3’ phosphonucleotide	5’ phosphonucleotide	5’3’ phosphodiesters	2’3’ phosphodiesters	**5’3’ phosphodiesters**
19	5	The pyrimidine nucleotides act as the high energy intermediates	UDPG	ATP	ADP	GDP	**UDPG**
20	5	The chemical name of thymine	2-Oxy –4-amino pyrimidine	2,4-dioxy –5-methyl pyrimidine	2,4- dioxy pyrimidine	Purine	**2,4-dioxy –5-methyl pyrimidine**
21	5	S1 nuclease	degrades double stranded DNA	degrades single stranded DNA	degrades RNA & DNA hybrid	degrades single stranded RNA	**degrades single stranded DNA**
22	5	An increased melting temperature for duplex DNA results from a high content of	A+G	C+T	A+T	G+C	**G+C**
23	5	The strucutre of RNA is in the form of	Hairpin loop	clover-leaf like structure	double helical structure	alpha helix	**clover-leaf like structure**

24	5	6-Amino purine is	Guanine	adenine	cytosine	adenine	**adenine**
25	5	The chemical name of 2-amino – 6- oxy purine	Adenine	xanthine	guanine	Cytosine	**guanine**
26	5	The lactom form is the predominant tautomer of	uracil	cytosine	xanthine	Guanine	**uracil**
27	5	N7-methyl guanine has been found more recently in the nucleic acids of the cells of	Bacteria	yeast	mammals	plants	**mammals**
28	5	Hypoxanyhine and ribose contitute	Adenosine	inosine	guanosine	cytidine	**inosine**
29	5	Thymine and deoxy ribose form	deoxycytidine	deoxyadenosine	deoxythymidine	deoxywidine	**deoxythymidine**
30	5	The most abundant intracellular free nucletide	ATP	FAD	NAD+	NADP+	**ATP**
31	5	The concentration of ATP in living mammalian cells in mm is near	0.2	0.3	0.4	1	**1**
32	5	The intracellular cAMP concentration in are µm near	3	2	1	0.5	**1**
33	5	The epinelisation of galactose to glucose & vice versa take place by	UTP	CTP	GTP	ATP	**UTP**
34	5	The biosynthesis of phosphoglycerides in animal tissue requires	ATP	GTP	CTP	TPP	**GTP**
35	5	The chemical name 4-hydro-xypyrazole pyrimidines is used for	Thioguanine	mercaptopurine	Axatheeopurine	Allopurinol	**Allopurinol**
36	5	Transforming factor is	RNA	DNA	tRNA	r-RNA	**DNA**
37	5	Guanosine nucletide is held by the cytosine nucletide by the number of hydrogen bonds	1	2	3	4	**2**
38	5	Within single turn of DNA the number of base pairs exists	4	6	8	10	**10**
39	5	Each turn of DNA structure has a pitch in nm of	1.4	2.4	3.4	4.4	**3.4**
40	5	The double stranded DNA	Denaturation	Filteration	sedimentation	concentration	**Denaturation**

		molecule loses its viscosity upon					
41	5	DNA molecule contains the number of nucletides	800-4000	1000-6000	1200-8000	1600-9000	**1600-9000**
42	5	DNA is denatured by	heat	Acid	Alkali	All	**All**
43	5	Chromatin contains the number of repeating units in nm	10	15	20	25	**10**
44	5	Each RNA molecule contains the number of nucleotides	40-4000	50-5000	60-6000	70-7000	**60-6000**
45	5	RNA molecule has a molecular weight of	15000-30000	20000-35000	25000-40000	30000-50000	**30000-50000**
46	5	Each tRNA molecule contains the number of nucleotides	70	75	80	85	**75**
47	5	In every cell the number of tRNA molecules are atleast	10	15	20	25	**20**
48	5	All tRNA molecules have a common CCA sequence at the	3’ termini	5’ termini	3’5’ termini	All	**3’ termini**
49	5	In nearly all tRNA molecules there is a loop containing the nucleotides of	Cytidine	thymidine	ribothymidine & pseudothymidine	cytidine	**ribothymidine & pseudothymidine**
50	5	In tRNA molecules, there is another loop containing the minor base	Uracil	Dihydro uracil	cytosine	Dihydrocytosine	**Dihydro uracil**
51	5	Because DNA is a highly charged polyanion, its stability to heat	does not depend on hydrophobic interactions	increases with increasing salt	independent of G - C content	decreases with increasing salt	**increases with increasing salt**
52	5	The sugar in RNA is ___________ , the sugar in DNA is ___________	deoxyribose, ribose	ribose, deoxyribose	ribose, phosphate	ribose, uracil	**ribose, phosphate**
53	5	Thymidine	can participate in hydrophobic interactions due to its methyl group	is replaced by uracil in RNA	normally forms two hydrogen bonds with adenosine	all of the above	**is replaced by uracil in RNA**

54	5	A nucleotide consists of	a sugar, a base and a phosphate	a sugar and a phosphate	paired bases	a sugar, a base and three phosphates	**a sugar, a base and a phosphate**
55	5	Which of the following is found on RNA but not DNA?	Uracil	Deoxyribose	Phosphate	Adenine	**Uracil**
56	5	The most stabilizing force for nucleic acids is	hydrogen bonds	electrostatic bond	Van der Waals	conformational entropy	**hydrogen bonds**
57	5	___________ together to form an mRNA molecule.	Splicing	Introns	exons	transcription	**Introns**
58	5	Oligonucleotides are formed from 2 to 10 ___________.	strands	hydrogen bonds	nucleosides	nucleotides	**nucleotides**
59	5	The variable portion of DNA is the sequence of ___________ .	phosphoric acids	sugars	bases	phosphates	**bases**
60	5	Which pair is a complementary base pair?	C-G	A-G	A-C	G-G	**A-G**