## **Biochemistry**

## Solution

*Definition :* It is a homogeneous mixture of two or more components constituting one phase only. Solution = Solvent + Solute.

Part of Solution : Solution has two parts -

- a. Solvent : The substance present in larger amount (more than 50%) in a solution is called solvent.
- b. Solute : The substance present in lesser amount (less than 50%) in a solution is called solute.

#### **Classification of Solution**

- A. On the basis of their physical appearance :
  - i. *Saturated solution*: A saturated solution is one in which there is an equilibrium between the solution and undissolved solute at the temperature of the solution. If there is addition of solute it will remain undissolve.
  - ii. Unsaturated solution : An unsaturated solution is one in which the solvent is capable of dissolving more of the solute at a definite temperature of the solution.
  - iii. Supersaturated solution : A solution containing undissolved solute particles at a definite temperature of the solution. Some solutes, form a saturated solution and others remain undissolved.

## B. On the basis of their permeability through the cell membrane :

- i. *Colloidal solution*: If the solute particles can not pass through the semipermeable membrane; then the solution is called colloidal solution e.g. Aggregation of albumin solution, plasma, dextran etc.
- ii. Crystalloid solution : If the solute paticles can pass through the semipermeable membrane then the solution is called crystalloid solution i.e cane suger solution, normal saline, dextrose saline, cholera saline etc.

#### C. On the basis of their concentration :

i.

*Percent solution*: Percent is used in expression of concentrate has the usual meaning of per hundred.

- a. *Percent by weight (wt /wt)*: This indicates the gm of solute per 100 gm of solution i.e 38% HCl by weight.
- b. *Percent by volume (wt./V):* This indicates the gms of solute per 100 ml of solution i.e 5% NaCl in water.
- c. *Percent by volume (V/V)* : This indicates the milliliters of solute per 100 ml of solution i.e gm of solute/100 ml eg. 5% DA.
- Molar solution : It is a solution which contain 1 gm molecular weight of solute per liter of solution. Gram molecular weight of HCl = 36.5 gm.

So, molar solution of HCl contain 36.5 gm/ liter.

- Molal solution : It is a solution which contain 1 gm molecular wt. of solute in 1000 gm of solvent. Molal solution of HC1 contains 36.5 gm/kg.
- iv. Normal solution : It is a solution which contain 1 gm equivalent wt. of solute per liter of solution. Gm. eq. wt. of  $H_2SO_4 = 49$ . So, normal solution of  $H_2SO_4$  contain = 49 gm/ liter of solution.
- v. Osmolar solution : It is a solution which contains one osmole of sloute per liter of solution.
- D. On the basis of tonicity : It is used to describe the effective osmotic pressure of a solution relative to plasma.
  - i. *Isotonic*: Solution which have the same osmotic pressure as that of plasma. E.g. 0.9% NaCl or 5% glucose soln. is isotonic to plasma.
  - ii. *Hypertonic* : A solution which has higher osmotic pressure than that of plasma .
  - iii. *Hypotonic* : A solution which has lower osmotic pressure than that of plasma.

### Different forms of solution

- 1. Liquid (solute) with liquid (solvent) e.g. Bromine water.
- 2. Solid (solute) with liquid (solvent) e.g Sugar in water.
- 3. Gas (solute) with liquid (solvent) e.g. Soda water.
- Gas (solute) with gas (solvent) i.e. Air. It is a mixture of O<sub>2</sub>, N<sub>2</sub>, CO<sub>2</sub> etc.
- 5. Solid (solute) with solid (solvent) i.e brass. It is a mixture of Cu & Zn.

#### Preparation of a solution in the Laboratory

- Apparatus required :
- i. Analytical balance
- ii. Funnel
- iii. Volumetric flask with stop cork
- iv. Beaker
- v. Stirer.

*Procedure :* At first the required solute is measured by an analytical balance. Then take it in a beaker. Add some solvent to the beaker and mix it thoroughly by a stirer. Then transfer the solution by the help of funnel to the volumetric flask. Repeat the procedure for several times so that almost all the solvent can be transfered to the volumetic flask. Stop the flask by stop cork.

## Osmole

It is the amount of solute in gms. which gives rise to an osmotic pressure of 22.4 standard atmosphere, when dissolved in one liter of solution.

One osmole is the number of particles in a gram molecular weight of undissociated solute. So, 180 gm of glucose in solution will be 1 osmol of glucose as the molecules do not dissociate.

If a solute like sodium chloride, two ions dissociate in solution. So 58.5 gm. (1 gm mol) of sodium chloride will be 2 osmole.

## 4.2 Physiology

Osmol = Molecular weight of a solute in gm

The number of ions or particles which dissociates in solution

Osmolality : Osmoles per kg of water is called osmolality.

Molarity : Number of moles /L e.g mmol/L.

Normality : Number of equivalent/L e.g meq/L.

#### Osmolarity

Osmoles per liter of solution is called osmolarity i.e number of osmole/L (mosm/L).

- a. Example : Plasma osmolarity = 280 300 mosm/L.
  - i. Nacl : 92 % (crystaloid)
  - ii. Others : 8 % (other crystaloid = 7.5%; colloid = 0.5%).

( Plasma protein= 0.5% = 1.5 mosm/LColloidal osmotic pressure= 25 mm of HCrystalloid osmotic pressure= 298.5 mosmm/L)

*B. Relation* between osmolality to osmotic pressure : Osmotic pressure (mm of Hg) = 19.3 x osmolality (mosm/liter).

c. Osmolar gap :

Osmolar gap = Measured osmolarity - calculated osmolarity = 0 - 12 mosm/L

Osmolar gap increases in :

- 1. Gross hyperproteinaemia
- 2. Gross hyperlipidaemia
- 3. Unmeasured osmotic solutes in plasma eg. manitol, alcohol etc.
- d. Consequences of osmotic disequilibrium :
  - 1. *Cellular dehydration* : may leads to cerebral dehydration. Upto 20% cellular dehydration can be tolerated by body itself- for ordinay cell and upto 10% cellular dehydration can be tolerated by brain cell.
  - 2. Cellular over hydration : may leads to oedema.

#### Iso-osmotic

Any two solution having the same osmotic pressure or osmolarity are called iso-osmotic to each other.

#### Concentration

The concentration of a solution means that the weight of the substance dissolved in unit volume of solution.

## Standard Solution

It is that solution whose strength and concentration is known. Standard solution are :

- i. Molar solution
- ii. Molal solution
- iii. Normal solution
- iv. Osmolar solution
- v. Percent solution.

#### Molecular weight

Molecular weight of any compound means the atomic weight of the component elements in their proper proportion in that compound.

e.g. molecular weight of NaCl = 23 + 35.5 = 58.5.

## Gram molecular weight

Molecular weight expressed in gm is known as gram molecular weight e.g. gram molecular weight of NaCl = 58.5 gm.

## Equivalent weight

The equivalent weight of a reactive unit is that weight which combines with or replace one atomic weight of hydrogen (1.008; or same reacting value of hydrogen.

Equivalent weight = molecular weight / valency

#### Gram equivalent weight

- 1. Equavalent weight expressed in gm is known as gram equavalent weight.
- 2. *Milli-equivalent* weight : It is one thousandth of the gm-equavalent weight.

#### **Normal Saline**

0.9% NaCl solution is called normal saline.

#### Atomic weight

The atomic weight of an element is a number which express how many times the weight of atom of the element is greater than the weight of one atom of hydrogen.

- Atomic weight = Weight of 1 atom of element/weight of 1 atom of hydrogen.
- One gram molecular weight of any substance contains 6.023 x 10<sup>23</sup> (Avogadro number) particles. Thus 180 gm of glucose, 58.5 gm of Nacl represents 6.023 x10<sup>23</sup> particles.

## **Collloids & Crystaloid**

#### Colloid

Definition: Colloid may be defined as a substance in which the solute particles are proportionately larger than solvent and is slowly diffusible and is incapable to pass through a semipermeable membrane. or

Colloids are the substances which by virtue of their molecular size are slow diffusible rather than soluble in water and are incapable of passing through a semi-permeable membrane.

#### Example :

- 1. Proteins (plasma protein)
- 2. Polysaccharides (starch, gelatin, glycogen)
- 3. Lipids
- 4. DNA & RNA etc.

#### Parts of colloidal solution :

- a. *Dispersed phase* : The solute part of the colloidal solution is called dispersed phase. It is discontinuous phase.
- b. *Dispersion media* : The solvent part of the colloidal soln. is called dispersion media. It is a continuous phase.

#### **Classification of colloids**

According to the physical state of the dispersed phase in relation to dispersion media a colloid may remains in two states

- a. Sol : Here the colloidal solution remains in liquid state, the dispersed phase is solid and the disperson media is liquid e.g milk.
- b. *Gel*: When the colloidal solution remain in solid state, the dispersed phase is liquid and the dispersion media is solid then it is called gel e.g butter.

## Types of colloidal solution

#### Colloidal solution fall into two classes -

- a. *Emulsoid or lypophilic* : Here both the dispersed phase and dispersion media are liquid, and the solute has a great affinity for the solvent eg. milk.
- b. *Suspensoid or lypophobic* : Here the dispersed phase is solid and the dispersion media is liquid and there is no affinity between solute and solvent eg. antacid syrup.

#### Some colloid & crystalloid solutions

Colloid	Crystalloid
i. Plasma.	i. Normal saline
ii. Dextran etc	ii. Dextrose saline.
	iii. Cholera saline etc.

## Process of separation of colloid

Separation of colloids may be done by -

- 1. Dialysis
- 2. Isoelectric precipitation.
- 3. Salting out
- 4. Electrophoresis
- 5. Adsorption

- 6. Ultracentrifugation
- 7. Ultrafiltration.

## Properties of colloids :

- A. General properties :
  - 1. Variable colour. Example : Gold solution- red, silver solution- grey, sulphate solution- colourless etc.
  - 2. Variable shape : usually spherical.
  - 3. Size : 1 200 mµ
  - Low osmotic pressure : Plasme colloidal osmotic pressure = 28 - 32 mm of Hg.
  - 5. Imbibition of water : I gm of albumin (colloid) can hold 17 ml of H<sub>2</sub>0.
  - 6. By dialysis crystalloid can be separated from colloid.
  - 7. Solution can be converted into sol or gel state.
- B. Optical properties :
  - 1. *Brownian movement* : The continuous, hapazard, irregular, zigzag movement of colloidal particles is known as Brownian movement. That can be seen under the ultramicroscope.
    - a. *Reason of motion* : The pushing effect of solvent molecule.
    - b. *Rate of motion* : Depends on the size of colloid molecules. The smaller the molecule, the greater the movement.
  - 2. Tyndall phenomenon : When a beam of light is passed through a colloidal solution & is observed from right angle, the colloidal particles become visible due to the dispersion of light by the colloid particles. This is known as Tyndall phenomenon.

#### C. Electrical properties :

- 1. *Electrophoresis* : Colloidal particles carry electrical charges on surface. When an electric current is passed through the colloidal solution, positively & negatively charged particles move to the opposite poles. This is known as electrophoresis.
  - a. Rate of electrophoresis depends upon :
    - i. Size &
    - ii. Amount of charge.
- 2. *Isoelectric pH* : It is the pH at which the colloids exist in solution as 'Zwitter ion' containing equal number of positive & negative charges on their surface. So the net charee is zero. At pH above the isoelectric pH, they will exist as anion and at pH below their isoelectric pH, they will exist as cation.
- D. Other properties :
  - a. Interfacial tension : Colloid particles exert great interfacial tension which results in irnbibition of water and adsorption of many substances by them.
  - b. Donnan membrane equilibrium
  - c. Sol to jel or jel to sol formation
  - d. Coagulation & salting out.

## 4.4 Physiology

#### Functions of colloids in our body :

- 1. Some of the biological fluids are colloidal solution e.g. blood, milk, CSF etc.
- 2. Biological compounds as colloidal particles : protein, lipid, polysaccharides.
- 3. Blood coagulation : when blood is clotted, it forms a gelly like substance.
- 4. Fat digestion and absorptjon : bile in the intestine emulcifies fat.
- 5. Format ion of urine.

## Importance of colloids

- 1. Essential constituents of cell cytoplasm.
- 2. Exert colloidal osmotic pressure required for the fluid exchange in the capillary.
- 3. Tissue fluid is imbeded by colloids and in this way a good deal of water remains stored in the body.
- Colloidal sate is responsible for the so called selective absorption or selective membrane permeabilities of certain ions.

## Crystalloid

1. *Definition* : A crystalloid may be defined as a substance which can pass through a semi-permiable membrane & cannot be separated by ultracentrifugation.

*Example* : Electrolytes (Na<sup>+</sup>, K<sup>+</sup>, Ca<sup>++</sup> etc), minerals, vitamins, glucose, amino acid, fatty acid.

## Properties of crystalloids :

1.	Size	:	Less than 1 mµ
2.	Solute	:	Are smaller than the solvent particles
3.	Permeability	:	Can pass through the semi-permeable membrane.
4.	Physical form	:	Crystaltic and can be crystalised from solution.
5.	Visibility	:	Can not be seen under ultramicro- scope
6.	Dialysability	;	Dialysable
7.	Osmotic pressure	10	High
8.	Optical activity	:	Don't exhibit
9.	Movement	:	Molecular
10.	Conversion	:	Cannot be converted into sol & gel state.
11.	Separation	1	Can not be separated. by ultrafiltra- tion & ultracentrifugation.

## Differentiate colloids from crystalloids

7	<i>Fraits</i>	Colloid	Crystalloid
1.	Size	1-200 mµ	Less than 1 mµ
2.	Solute	are larger than the solvent particles	are smaller than the solvent particles
3.	Permeability	Cannot pass through the semi-permeable membrane.	Can pass

4.	Physical form	Amorphous and has no definite shape solution.	Crystaltic and can be crystalised from
5.	Visibility	Colloid particles can be seen under ultramicroscope	Can not be seen.
6.	Dialysability	Non dialysable	dialysable
7.	Osmotic pressure	Low	High
8.	Optical activity	Exhibit	Don't exhibit
9.	Movement	Brownian	Molecular
10.	Conversion	Solution can be con- verted into sol & gel state.	Cannot be converted
11.	Separation	Can be seperated by ultrafiltration & ultracentrifugation	Can not be separated
12.	Example	Plasma protein, lipid, starch, glycogen, DNA & RNA.	Electrolytes, minerals, vitamins, glucose, amino acid, fatty acid.

#### Differentiate Emulsoid & Suspensoid :

Emulsoid	Suspensoid
1. Here both dispersed phase and dispersion liquid.	<ol> <li>Here dispersed phase is solid &amp; the media are dispersion media is liquid.</li> </ol>
<ol> <li>The solute have a great affinity for the solvent.</li> </ol>	<ol><li>There is no affinity between solute and solvent.</li></ol>
3. Emulsoid is reversible.	3. Suspensoid is irreversible.
<ol> <li>It is stable colloidal solution.</li> </ol>	<ol> <li>It is less stable than emulsoid.</li> </ol>
<ol> <li>Well defined under micros- cope &amp; ultramicroscope</li> </ol>	<ol> <li>Invisible under microscope &amp; ultramicroscope</li> </ol>
<ol> <li>Not easily precipitated by salts.</li> </ol>	<ol> <li>Easily precipitated by salts.</li> </ol>
7. Particles surrounded by the shell absorbed water	7. Particles not surrounded by water shell
<ol> <li>Lower surface tension and much higher viscosity than the solvent.</li> </ol>	<ol> <li>Surface tension and higher viscosity of suspensoids are rearly the same as those of the solvent.</li> </ol>
Differnce between sol and gel	ie i schoen
Sol	Gel
<ol> <li>Colloid particles bind loosely with one another.</li> </ol>	<ol> <li>Colloid particles bind firmly with one another.</li> </ol>
<ol> <li>Here dispersion media is continous.</li> </ol>	<ol> <li>Here dispersion media is formed by hydrated colloid particles.</li> </ol>

## **Acid Base Balance**

#### Electrolytes

Electrolytes are the compound which can be dissociated into anion and cation on an electric current being passed. e. g Acid, base.

#### Non-Electrolytes

The compound which can not be dissociated into anions and cations on an electrical current being passed. e.g Cane sugar soln.

#### lons

Ions are the charged particles, which moves towards the positive pole (anode) or negative pole (cathod). They are formed when electrical current being passed through a solution of electrolytes.

- a. Ions moves toward the positive pole is called anion.
- b. Ions moves toward the negative pole is called cation.

#### Differnce between ions & electrolyte :

Ions	Electrolyte
1. lons are charged particles.	1. Electrolytes are neutral.
<ol> <li>lons does not gives rise to electrolyte.</li> </ol>	<ol> <li>Electrolytes gives up ions when placed in a electric field.</li> </ol>
<ol> <li>Ions moves towards the opposite pole.</li> </ol>	3. No such movement occurs.
<ol> <li>Pathway of ions can be traced by electrophoresis.</li> </ol>	<ol> <li>Electrolytes does not give such electrophoresis.</li> </ol>

#### Particles

Particles may be the molecule or fraction of molecule or aggregation of molecules.

Molecules e.g glucose. Fraction of molecule e.g in NaCl - Na & Cl. Aggregation of molecules e.g albumin.

#### Hydrophilic

If the dispersed phase of colloidal solution is liquid and the dispersion media is water then it is called hydrophilic. Here the solute has a great affinity for water.

#### Hydrophobic

If the dispersed phase of colloidal solution is solid and the dispersion media is water then it is called hydrophobic. Here the solute has no affinity for water and repeal each other.

## Acid

*Definition* : Acid is the substance, which produces hydrogen ion  $(H^+)$  when dissolved in water e.g HCl, H<sub>2</sub>CO<sub>3</sub>.

- i. It is proton (H<sup>+</sup>) donar
- ii. A solution is acid because there are more H<sup>+</sup> than OH<sup>-</sup>.

#### Types of Acid :

#### There are two types of acid

i. Strong acid : The acids which can dissociated fully

(strongly) when dissolve in water e.g HCl,  $H_2SO_4$  etc. Usually all the inorganic acids are strong acids except  $H_2CO_3$ .

ii. Weak acid : The acids which can dissociated partially (weakly), when dissolve in water e.g lactic acid, acetic acid,  $H_2CO_3$  etc. Usually all the organic acids are weak acids.

Concentrated acid : Acid in molecular form.

Dilute acid : Acid in ionized form.

Difference	between	strong	and	weak	acid
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	Strong acid	Weak acid
1.	It dissociates quickly and strongly.	<ol> <li>It dissociates slowly and partially.</li> </ol>
2.	Here conjugate base is weak.	2. Here conjugate base is strong.
3.	It is irreversible in reaction.	3. It is reversible in reaction.
4.	Usually these are inorganic acid.	<ol> <li>Usually these are organic acid.</li> </ol>

#### Properties of acid

- 1. It produces hydrogen ion in aqueous solution.
- 2. It produces salt and water with alkali
- 3. It converts blue litmus into red litmus.
- 4. It is sour in taste.

#### Strength of an acid

Strength of an acid is determined by

- 1. The strength of the conjugate base.
- 2. The basic strength of the solvent.
- 3. The dielectric constant of the solvent.

#### Acidity

The state of being acid or acid content of a fluid is known as acidity.

Types of acidity :

- 1. Actual acidity : This is the concentration of H<sup>+</sup> ion in a solution. It is spoken as pH.
- Free acidity : This is the amount of free acid which is present in the solution, not in combination with other compound.
- Combined acidity : Amount of acid present in combination with organic compound (e.g. proteins) and that present in acid salts.
- 4. *Total acidity :* Amount of free acid plus that present in combination. Free acid+ combined acid= total acid.
- 5. *Titrable acidity*: Amount of total ionizable hydrogen both ionized and non-inoized, determined by titration against a base. The concentration of H<sup>+</sup>ions in a solution plus those available for ionization although not actually ionized at the time.

## Base

Definition : The substance which can receive hydrogen ion (H+)

## 4.6 Physiology

#### is called base e.g OH<sup>-</sup>, Cl<sup>-</sup>, HCO<sub>3</sub><sup>-</sup> etc.

- i. It is proton (H<sup>+</sup>) acceptor.
- ii. A solution is alkaline because there is more hydroxyl ion than H<sup>+</sup>.

## Types of base :

- i. Strong base : The base which ionized completely in solution and produces a lot of hydroxyl ion (OH<sup>-</sup>) e.g NaOH, KOH etc.
- Weak base : The base which ionized partially in solution and produces small amount of hydroxyl ion (OH<sup>-</sup>) e.g. NH<sub>4</sub>OH.

## Conjugate base

The electro-negative portion of acid which receives hydrogen ion or proton to form acid is known as conjugate base.

- \* The conjugate base of strong acid is very weak. So strong acid dissociate very quickly and strongly.
- \* The conjugate base of the weak acid is strong. So the weak acid dissociate weakly and partially.

#### Alkali

The substance which can donate OH<sup>-</sup> ion in aqueous solution is called alkali.

Example : NaOH Na<sup>+</sup> + OH<sup>-</sup>. Here NaOH is alkali but hydroxyl ion (OH<sup>-</sup>) is base.

## Properties of alkali :

- 1. It produces hydroxyl ion in aqueous solution.
- 2. It produces salt and water with acid.
- 3. It convert red litmus into blue.

#### Difference between acid and alkali

	Acid		Alkali
1.	It produces hydrogen ion in water.	1.	It produces hydroxyl ion in water.
2.	It produces salt and water with alkali.	2.	It produces salt and water with acid.
3.	It converts blue litmus into red.	3.	It converts red litmus into blue.

Basic : Relating to a base of any kind is called basic.

**Basicity**: The state of being base or base content of a fluid is known as basicity.

Alkaline : Relating to an alkali, having the reaction of an alkali, called alkaline.

Alkalinity : The state of being alkali is known as alkalinity.

Alkalinuria : The passage of alkaline urine.

Alkalizer : An agent that neutralizes acid, called alkalizer.

#### рн

pH is the negative logarithm of hydrogen ion concentration to the base 10.

$$pH = -\log(H+)$$

 $= \log (1/H^+)$ 

(P indicates the negative logarithm to base 10)

So there is inverse relationship between the pH and hydrogen ion concentration. If hydrogen ion concentration increases, PH decreases and vice versa.

Normal body pH	:	7.35 - 7.45 (average 7.4)
Clinical safe pH	:	7.3 - 7.5
pH compitable to life	:	6.8 - 7.8
If the pH is above the 7.8	:	Death occurs due to tetany and cardiac arrest.

#### рн 7:

$$H^{+} = 10^{-7} \text{ mole/L}$$
  
= 10<sup>-7</sup> x 10<sup>9</sup>  
= 10<sup>2</sup>  
= 100 nmol/L

*pH*6:

 $H^{+} = 10^{-6} \text{ mole/L}$ 

 $= 10^{-6} \times 10^{9}$ 

$$= 10^3$$

= 1000 nmol/L

Significance of pH :

It is the measure of extent of acidity or alkalinity of a solution.

Importance of pH :

- 1. Maintenance of enzyme activity
- 2. Maintenance of cellular viability
- 3. Maintenance of neuromuscular irritability.

## **Optimum pH**

The P<sup>H</sup> at which most of the physiological enzymatic reaction occurs is known as optimum pH.

#### pH Scale

p

Sorensen introduced the simple pH scale. The degree of acidity or alkalinity can be measured on a scale of P<sup>H</sup> unit.

pH Scale	:	0 - 14	
рн 7	:	neutral.	
pH less than 7	:	acidic.	
H more than 7	:	alkaline.	

pH7: H<sup>+</sup> = 10<sup>-7</sup> mole/L = 10<sup>-7</sup> x 10<sup>9</sup> = 10<sup>2</sup> = 100 nmol/L

pH 6: H<sup>+</sup> = 10<sup>-6</sup> mole/L = 10<sup>-6</sup> x 10<sup>9</sup> = 10<sup>3</sup> = 1000 nmol/L

Expression of acidity :

рН	H <sup>+</sup> concentration (nmole/L)
6.8	160
7.3	50
7.35	45
7.4	40
7.45	35
7.5	30
7.8	16

Salient features of pH scale :

- 1. For unit change of pH, (H<sup>+</sup>) will change inverssly 10 times and (OH<sup>-</sup>) will chane directly 10 times.
- 2. At neutrality (H<sup>+</sup>) is equal to 10<sup>-7</sup> moles/L and neutral pH will be 7.
- 3. At 25°C Kw = 10-14 and so pH + p OH = 14
- Acidity means (H<sup>+</sup>) more than (OH<sup>-</sup>) and alkalinity means (OH<sup>-</sup>) more than (H<sup>+</sup>).
- 5 Even an extreme acidic solution contain some amount of OH<sup>-</sup> ions and even an extreme alkaline solution contain some amount of H<sup>+</sup> ions.

## Q. What do you mean by $P^H$ of a solution = 7?

#### Ans.

- i. It is the solution contains  $10^{-7}$  gmeq./L of hydrogen ion (H<sup>+</sup>)
- ii. It is the pH of water.
- iii. It means the solution is neutral.

## Calculation of $P^H$ of a solution :

For the calculation of pH,  $(H^+)$  ion concentration must be ascertained. If the hydrogen ion concentration of a solution is known then the pH of a solution can be calculated by using the formula-

 $pH = log (1/H^+).$ 

# Determination of pH of water with the help of law of mass action :

Water dissociates to a small but definite extent, depending on the temperature. i. e.  $HOH = H^+ + OH^-$ .

The higher the temperature, greater will be the dissociation.

According to the law of mass action, at equilibrium-

$$\frac{(H^+) \times (OH^-)}{(HOH)} = K (Constant)$$

But ordinarily, very few water molecules in a mass of water may get dissociated; So, the vast mass of undissociated water may taken to remain constant.

Thus -

$$\frac{(H^+) \times (OH^-)}{(HOH)} = K$$
  
or  $(H^+) \times (OH^-) = (HOH) \times K$ 

 $= K_1 \times K$  $= K_W.$ 

Here  $K_W$  is ion product or dissociation constant of water at particular temperature. At room temperature (25<sup>o</sup>C) the value of  $K_W = 1 \times 10^{-14} \text{ gmEq./Liter.}$ 

As dissociation of each molecule of water yields equal numbers of H<sup>+</sup> and OH<sup>-</sup>

Q. Why water is taken as standard for comparison of pH? Ans.

- i. Water Dissociates to a small but definite extent, depending on the temperature.
- ii. It produces same amount of H+ and OH- when dissociate.
- iii. It is neutral in nature.
- iv. It is stable and do not change physically and chemically for a long range of variation.

So, Water is taken as standard for comparison of pH.

#### рОН

It is the negative logarithm of (OH<sup>-</sup>) ion concen-tration of a solution. It is useful in calculating the pH of an alkaline solution.

#### Safe guard of pH

- 1. 1st line
  - i. Buffers within seconds
  - ii. Respiratory system minute to hours.
- 2. 2nd line :
  - i. Kidney system hours to days.

Metabolic acid base production :

- 1. Volatile acid (CO<sub>2</sub>) : 12 18 mole/day
- 2. Non-volatile acid : 50 100 meq/day (1 meq/kg/day)
- 3. Total acid : 1240 meq/day
- 4. Total base : 1160 meq/day
- 5. NET acid : 80 meq/day.

Routes of acid excretion :

- 1. Lungs : volatile acid
- Kidney : non-volatine acid.

N.B. Buffers are giving important protection of our pH system from production to excretion.

## Buffer

Definition : Buffer is the mixture of weak acid and it's salt with strong bases (B<sup>-</sup>/HB)

#### Example :

- NaHCO3 / H2CO3 i.
- KHb / H-Hb or KHbO2/ H-HbO2 ii.
- Na2HPO4 / NaH2PO4 ii.
- iv. Prot. / H-prot.
- NH<sub>3</sub>/NH<sub>4</sub>+ ٧.

## It depends upon-

pK of the buffer. a.

Concentration of buffer substance. b

salt (anti-acid compert) pH of a buffer solution,  $pH = pK + \log R$ acid (anti-base compert)

Composition of buffer : It is a pair of weak acid and its salt with a strong base.

#### Importance of buffer :

- It maintain the constant hydrogen ion concentration of the body for optimum cellular activity.
- ii. It is important in maintenance of normal acid-base balance of the body.
- iii. It is important for the optimum activity of enzyme.

## **Buffer** solution

A solution of two or more chemical compounds which has the ability to accept either the (H<sup>+</sup>) or (OH<sup>-</sup>) ions, in the solution, thus maintaining the pH of the solution relatively constant, despite the addition of considerable quantity of acid and base.

## Physiological buffer :

These are the buffers present in the body and maintains the pH of body fluid at a constant level, despite the addition of acid and alkali produced during metabolic activity is called physiological buffer. Example : H<sub>2</sub>CO<sub>3</sub>/HCO<sub>3</sub><sup>-</sup>.

## Buffer system of the body

Buffer system of the body are (according to severity) :

1. Plasma buffer	rs :
------------------	------

	i.	Bicarbonate buffer	•	NaHCO3 / H2CO3
	ii.	Protein buffer	:	Prot. / H-Prot.
	ii.	Phosphate buffer	:	Na <sub>2</sub> HPO <sub>4</sub> / NaH <sub>2</sub> PO <sub>4</sub>
2.	RB	C buffers :		
	i.	Haemoglobin buffer	:	KHb / H-Hb <i>or</i> KHbO <sub>2</sub> / H-HbO <sub>2</sub>
	ii.	Bicarbonate buffer		NaHCO <sub>3</sub> / H <sub>2</sub> CO <sub>3</sub>
	iii.	Phosphate buffer		Na <sub>2</sub> HPO <sub>4</sub> / NaH <sub>2</sub> PO <sub>4</sub>
3.	Blo	od buffers :		
	i.	Bicarbonate buffer	:	NaHCO <sub>3</sub> / H <sub>2</sub> CO <sub>3</sub>
	i.	Haemoglobin buffer	:	KHb / H-Hb
				or KHbO <sub>2</sub> / H-HbO <sub>2</sub>

	-	
111.	Protein	hutter
111.	riotem	ounci

- iv. Phosphate buffer
- 4. ECF buffers :
  - Bicarbonate buffer i. ii. Phosphate buffer
  - iii. Protein buffer
- 5. ICF buffers :

i.	Protein buffer	:	Prot. / H-Prot.
ii.	Phosphate buffer	:	Na <sub>2</sub> HPO <sub>4</sub> / NaH <sub>2</sub> PO <sub>4</sub>

- iii. Bicarbonate buffer
- - a. Acting throught the respiratory system :
    - i. Oxidised and reduced haemoglobin buffer system
    - ii. HCO3<sup>-</sup>/H2CO3 buffer system.
  - b. Acting through urinary system :
    - Phosphate buffer system (NaH2PO4 / Na2H PO4) 1.
    - iv. Ammonia buffer system (NH<sub>3</sub> / NH<sub>4</sub><sup>+</sup>).

#### **Buffering capacity**

Means the capability of resisting the change in pH by accepting either the  $(H^+)$  or  $(OH^-)$  ions.

#### Total body buffering capacity :

1000 meq H+/OH- or 12-15 meq/kg.

- i. Bicarbonate buffer (NaHCO<sub>3</sub> / H<sub>2</sub>CO<sub>3</sub>) : 55 %
- ii. Haemoglobin buffer (KHb / H-Hb or : 35 % KHbO<sub>2</sub>/H-HbO<sub>2</sub>)
- iii. Protein buffer (Prot. / H-Prot.) : 5%
- iv. Phosphate buffer (Na2HPO4 / NaH2PO4) 5 %

#### Body buffering activity :

- ECF buffer : Quick action, low capacity-1.
  - i. Bicarbonate buffer (NaHCO3 / H2CO3) : 86 %
  - ii. Protein buffer (Prot. / H-Prot.) + : 14 % Phosphate buffer (Na2HPO4 / NaH2PO4)
- ICF buffer : Delayed effect, high capacity, affect plasma 1. K<sup>+</sup> level.
  - i. Protein buffer (Prot. / H-Prot.) + : 64 % Phosphate buffer (Na2HPO4 / NaH2PO4)
  - ii. Bicarbonate buffer (NaHCO<sub>3</sub> / H<sub>2</sub>CO<sub>3</sub> ) : 36 %

#### рК

Is the negative logarithm of dissociation constant.

pK of body buffers :

i.	NaHCO <sub>3</sub> / H <sub>2</sub> CO <sub>3</sub>		6.1
	KHb / H-Hb or KHbO2/ H-HbO2		
	Na <sub>2</sub> HPO <sub>4</sub> / NaH <sub>2</sub> PO <sub>4</sub>	:	6.8
iv.	Prot. / H-prot.	:	6.5 - 7
٧.	$NH_3 / NH_4^+$	:	9.0

Buffering capacity will maximum when, pK = pH

NaHCO<sub>3</sub> / H<sub>2</sub>CO<sub>3</sub> : Na<sub>2</sub>HPO<sub>4</sub> / NaH<sub>2</sub>PO<sub>4</sub> : Prot. / H-Prot.

Na2HPO4 / NaH2PO4

Prot. / H-Prot.

- - : NaHCO<sub>3</sub> / H<sub>2</sub>CO<sub>3</sub>
- Systemic buffers :

#### Q. When will be the pH = pK?

Ans. We know that  $pH = pK + \log (Salt/Acid)$ . If the ratio of salt to acid is 1:1 (i.e. concentration of salt and

acid is same). Then the pH of the resultant mixture would be equal to pK.

#### Mode of buffer action :

- i. Conversion of strong acid into weak acid.
- ii. Conversion of strong base into neutral salt.

#### Mechanism of buffer action :

Buffer posses the capability of resisting the change in pH, by accepting either the  $(H^+)$  or  $(OH^-)$  ion.

So when strong acid such as HCl is added in bicarbonate buffer system ( $H_2CO_3/NaHCO_3$ ), then  $NaHCO_3$  neutralizes it, and strong acid is converted into weak acid.

 $HCl + NaHCO_2 = NaCl + H_2CO_3$  (weak acid)

Similary when a strong base such as NaOH is added in same buffer system ( $H_2CO_3/NaHCO_3$ ), then  $H_2CO_3$  converted the NaOH into neutral salt.

 $H_2CO_3 + NaOH = NaHCO_3$  (Neutral salt. +  $H_2O$ )

## Differnce between bicarbonate and phosphate buffer

Trait	Bicarbonate buffer	phosohate buffer
1. Composition	NaHCO <sub>3</sub> / H <sub>2</sub> CO <sub>3</sub>	(Na <sub>2</sub> HPO <sub>4</sub> / NaH <sub>2</sub> PO <sub>4</sub>
2. Conc. ratio in blood	20/1	4/1
3. Blood conc.	26-28 mmol/L High concentration	1.0 mmol/L Low concentration
<ol> <li>Linked up with</li> </ol>	Respiratory & kidney system	Kidney system
5. Defence	1st line of defence	less effective
б. рК	It is weaker than phosphate buffer because its pk is away fromthe pH of ECF	It is stronger than bicar bonate buffer because its pk is nearer to the pH of ECF
7. Importance	In ECF because of greater concentration	In ICF because of greater concentration.

#### Importance of bicarbonate buffer :

It plays an important role in the maintanence of acid-base balance because the concentration of each of the two element of the system can be regulated.

- i. CO<sub>2</sub> is regulated by respiratory system.
- ii. Bicarbonate ion by the kidney.

#### Bone buffer

- i. Alkaline calcium salt of bone
- ii. Capacity : 35000 meq H+ (adult)
- iii.  $Ca_3(PO_4)_3 + Ca^{++} + H_2O \iff Ca_{10}(PO_4)_6(OH)_2 + H^+$

Hydroxy appatite crystal

*Hydroxy appatite crystal*  $\rightarrow$  pick up proton (H<sup>+</sup>).

iv. Chronic acidosis :

In chronic renal failure  $\rightarrow$  bone buffer acts to maintain H<sup>+</sup> concentration.

- a. During alkalosis : by releasing proton (H<sup>+</sup>)
- b. During acidosis : by picking up proton (H<sup>+</sup>)

#### Total buffer base

Total buffer base of blood = 48 meg/L

- i. HCO3<sup>-</sup> : 28 meq/L
- ii. Hb<sup>-</sup> : 15 meq/L
- ii. Others : 5 meg/L

Base excess (BE) :

```
Base excess = Observe buffer base (HCO<sub>3</sub><sup>-</sup> concentration) -
normal buffer base (normal HCO<sub>3</sub><sup>-</sup>
concentration)
```

Normal value of base excess : -2 to +2 meq/L i. +ve base excess : Metabolic alkalosis ii. -ve base excess : Metabolic acidosis

N.B. Base component of a buffer fight against proton (H<sup>+</sup>)

#### Acid base disorder

Types of acid base disorder :

- 1. Simple acid base disorder :
  - i. Metabolic
    - a. Acidosis (decrease HCO3-)
    - b. Alkalosis (increase HCO3<sup>-</sup>)
  - ii. Respiratory
    - a. Acidosis (increase PCO<sub>2</sub>)
    - b. Alkalosis (decrease PCO<sub>2</sub>)
- 2. *Complex acid base disorder* : If both component vary. Any combination of 4 simple acid base disorder, but all four can not persist in a body.

#### Acid base parameters

- 1. Classical acid base parameters
  - i. pH (7.4)
  - ii. PCO<sub>2</sub> (40 mm of Hg)
- 2. Non classical acid base parameters
  - i. Plasma anion gap
  - ii. Electrolytes.

Plasma anion gap :

Law of neutrality-

Cations = Anions

- i.e  $Na^+ + K^+ + unmeasured cations (UC)$ 
  - = Cl<sup>-</sup> + HCO<sub>3</sub><sup>-</sup> + unmeasured anions (UA)
  - $(Na^+ + K^+) (Cl^- + HCO_3^-)$ 
    - = Unmeasured anions unmeasured cations
    - = UA UC
    - = Anion gap

UC : Ca++, Mg++, gama globulin

UA : Plasma protein, SO<sub>4</sub>, PO<sub>4</sub>, lactate, others

## Normal value : 12 (+/-) 4 meq/L.

#### Causes of increased anion gap :

- i. Renal failure
- ii. Lactic acidosis
- iii. Ketoacidosis
- iv. Intoxication of aspirin, alcohol poisoning.

*N.B.* End product of alcohol metabolism  $\rightarrow$  organic acid  $\rightarrow$  anion of that organic acid  $\rightarrow$  increase anion gap).

## Compensatory mechanism of acid baes disorder

1. Metabolic acidosis :

$$=\frac{decreased HCO_3}{PCO_2}$$

Compensation

2

→ pH becomes normal, but total body CO<sub>2</sub> decreases due to \* hyperventilation (Respiratory sysrem managed)

2. Metabolic alkalosis :

$$= \frac{increased HCO_3}{PCO_2}$$

Compensation

increased HCO<sub>3</sub>

- > pH becomes normal,
- but total body CO<sub>2</sub> increases due to \* *hypoventilation* (Respiratory sysrem managed)
- 1. Respiratory acidosis :

$$= \frac{HCO_3}{increased PCO_2}$$

Compensation =

- $\frac{\text{increase HCO}_3^-}{\text{increased PCO}_2}$
- → pH becomes normal, but total body CO<sub>2</sub> increases due to \* HCO<sub>3</sub><sup>-</sup> generation by the kidney (Kidney sysrem managed).
- 4. Respiratory alkalosis :

$$= \frac{\text{HCO}_3^-}{\text{decreased PCO}_2}$$

Compensation

$$\frac{\text{decrease HCO}_3}{\text{decreased PCO}_2}$$

 $\rightarrow$  pH becomes normal,

- but total body  $CO_2$  decreases due to \* *increase*  $HCO_3^-$  *excretion* by
  - the kidney
  - (Kidney sysrem managed).

#### 4 sequele occurs :

- 1. Total body CO2 decreases in
  - i. Metabolic acidosis
  - ii. Respiratory alkalosis
  - iii. Metabolic acidosis + Respiratory alkalosis.
- 2. Total body CO2 increases in
  - i. Metabolic alkalosis
  - ii. Respiratory acidosis.
- Total body CO<sub>2</sub> normal with acid baes disorder ini. Metabolic acidosis + Respiratory acidosis
  - ii. Metabolic alkalosis + Respiratory alkalosis.
- Total body CO<sub>2</sub> normal with acid baes disorder ini. Metabolic acidosis + Metabolic alkalosis.

## Laws & Equation

#### Law of mass action :

The velocity of reversible reaction is directly proportional to the product of the concentration of the reacting mass at constant temperature.

Chemical reaction :

HOH 
$$\langle V_1 \rangle$$
 H<sup>+</sup> + OH<sup>-</sup>

According to the law of mass action the velocity  $(V_1)$  of Ist reaction is proportional to (HOH) ie.

$$V_1 \propto (HOH)$$
 or  
 $V_1 = K_1 (HOH)$  .....

Here, K<sub>1</sub> is constant.

When velocity (V2) of 2nd reaction is considered

$$V_2 \propto (H^+) \times (OH^-)$$
 o

 $V_2 = K_2 (H^+) x (OH^-)$  ..... ii

Here, K<sub>2</sub> is constant.

The velocity of reactions i & ii are equal as the reaction occur at equilibrium position i.e.

$$V_{1} = V_{2}$$
  
i.e.  
$$K_{1}(HOH) = K_{2}(H^{+}) \times (OH^{-})$$
  
or 
$$\frac{K_{1}}{K_{2}} = \frac{(H^{+}) \times (OH^{-})}{HOH} = K$$
  
or 
$$\frac{(H^{+}) \times (OH^{-})}{HOH} = K = \frac{\text{Dissociated water}}{\text{Undissociated water}}$$

Here, K is equilibrium constant.

#### Importance of Law of mass action :

It is used to determine -

- i. The p<sup>H</sup> of a solution.
- ii. The hydrogen ion concentration (H<sup>+</sup>).
- iii. The dissociation constant.
- iv. Handerson-hasselbalch reaction.

#### Handerson-Hasselbalch Equation

Definition: The relationship between pH, pK and the concentration of an acid and its salt can be stated by a convenient equation -

$$p^{H} = p^{K} + \log \frac{\text{salt}}{\text{acid}}$$

This equation is called Handerson-Hasselbalch equation.

Here, pK is the negative logarithm of dissociated constant .

*Explanation* : When considered a pure solution of a weakly dissociated acid at equilibrium stage.

Such as HB  $\langle H^+ + B^- \rangle$  (B<sup>-</sup> = Base)

According to the low of mass action :

 $\frac{(H^+) \times (B^-)}{(HB)} = K \qquad (K = equilibrium constant)$ 

So,

 $(H^+) \times (B^-) = K (HB)$  (Cross multiplication)

$$(H^{+}) = K \frac{(HB)}{(B-)}$$
(Divided each side by B<sup>-</sup>)

So, 
$$\text{Log}(\text{H}^+) = \log \frac{\text{K}(\text{HB})}{(\text{B}^-)}$$

or, 
$$\log (H^+) = \log K + \log \frac{(HB)}{(B^-)}$$

So, 
$$-\log (H^+) = -\log K - \log \frac{(HB)}{(B^-)}$$

or, 
$$-\log(H^+) = -\log K + \log \frac{(B^-)}{(HB)}$$

or, 
$$pH = pK + \log \frac{(B^-)}{(HB)}$$

$$p^{H} = pK + \log \frac{Salt}{acid}$$

#### Importance of Handerson-Hasselbalch equation :

- 1. It is used to determined the pH of a buffer solution.
- 2. For determination of the relative concentration of salt and acid required.
  - a. pK = pH, when the two constituents are present in equal concentration.

- b. pH of buffer solution is determined by the ratio of salt and acid.
- c. The buffering power of a substance is highest when pH = pK.

## **Donan Equilibrium**

*Definition*: It is the condition of unequal distribution of diffusible ions across the semipermeable membrane due to the presence of non diffusible anion on one side of the semipermeable membrane.

Importance of Donnan equilibrium :

- 1. It explains how difference of electrical potential can be established on two sides of a membrane.
- 2. It may also involved in absorption and secretion.
- 3. In maintenance of differential concentration between the various compartments of the body.
- 4. Phenomenon of chloride shift can also be explained from this standpoint.

*Explanation* : Consider two ionized solutions a and b filling compartments of constant volume and separated by a partially permeable membrane. It was shown theoretically by Gibbs and confirmed experimentally by Donnan-that, *at equilibrium* :

1. Each solution will be electrically neutral- it's total charges on cations will equal those on anion.

2. The product of the diffusible ion on one side of the membrane will equal the product of the diffusible ions on the other.

(Diffusible cations)<sub>a</sub> x (Diffusible anions)<sub>a</sub>

= (Diffusible cations)<sub>b</sub> x (Diffusible anions)<sub>b</sub>

From which it follows that -

(Diffusible cations) <sub>a</sub>		(Diffusible anions)b
(Diffusible cations) <sub>b</sub>	=	(Diffusible anions) <sub>a</sub>

Consider that in our simple system sodium chloride  $(Na^+Cl^-)$  is present in solution *a* and *b* but that only *a*, contains a salt  $(Na^+X^-)$ ; where  $X^-$  is an undiffusible anion unable to cross the membrane.

Thus -

	Na+	Na+	
Solution (a)	Cl <sup>-</sup>	Cl <sup>-</sup>	Solution (b)
	X-		

The penetrating ions (Na<sup>+</sup> and Cl<sup>-</sup>) diffuse until equilibrium is attained. The two criteria established above will hold, namely.

$$(Na^{+})_{a} = (Cl^{-})_{a} + (X^{-})_{a}$$
$$(Na^{+})_{b} = (Cl^{-})_{b}$$

Electrically Neutral

and

## 4.12 Physiology

$(Na^+)_a \times (C1^-)_a$	=	(Na <sup>+</sup> ) <sub>b</sub> x (C1 <sup>-</sup> ) <sub>b</sub>	
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or,  $\frac{(Na^+)_a}{(Na^+)_b} = \frac{(C1^-)_b}{(C1^-)_a}$ 

From these relationships it follows that-

$$(Na^+)_a > (Cl^-)_a$$

and therefore-(Na<sup>+</sup>)<sub>2</sub>

$$Na^+)_a > (Na^+)_b$$

$$(Cl^-)_a < (Cl^-)_b$$

and

 $(Na^+)_a = (Cl^-)_a + (X^-)_a > (Na^+)_b + (Cl^-)_b$ 

So,

- 1. Cation concentration on the side containing non-diffusible anion will greater that on the other side.
- 2. Anion (diffusible) concentration will be more on the side without non-diffusible anion.
- Side containing non-diffusible ion will have more negativity.

(Ref. wright's)

Products & ratio

of diffusible ions

## Indicator

*Definition* : Indicator is the weak acid or weak base which indicates the acid-base reaction by changing their colour in an ionized and unionized form. It also indicates the end point of a reaction.

They react by changing their colour in an ionized and unionized form.

Some Common idicators with pH:

	Indicator	pH range	Colour change
1.	Topfer's reagent	2.9 - 4.0	Red to yellow
	Methyl red Litmus	4.2 - 6.3 4.5 - 8.3	Red to yellow Red to blue
4.	Phenopthalein	8.3-10.0	Colourless to Red
5.	Phenol red	6.6 - 8.3	Yellow to red

Use of indicator :

- i. It is used to determine the pH of a solution.
- ii. It also used in the laboratory for the titration, to indicate the end point of an acid-base reaction.

#### Q. How indicator is chosen?

Ans. Indicator is chosen according to the pH of media and the pK of indicator.

## Titration

Titration is the process by which the concentration of a solution is determined against (by) a known solution with the help of indicator.

*Importance of titration :* The process is used in the laboratory to know the strength and concentration of unknown solution.

## **Biological Oxidation**

*Oxidation* : Addition of oxygen or removal of hydrogen or removal of electron with gaining valency is called oxidation or chemical oxidation.

*Reduction :* Loss of  $O_2$  and addition of H<sup>+</sup> or electrons is known as reduction.

*N.B.* The enzymic actions are activated by the presence of some accessory substances. When these accessory substances are single ions (Mg<sup>+</sup>,  $PO_4^-$ ), they are called *co-factors*, but when they are complex non-protein organic molecules, they are known as *co-enzymes*.

*Biological Oxidation* : Oxidation in the tissue cells is called biological oxidation.

Difference between oxidation and biological oxidation : Oxidation occurs only one step of reaction. Biological oxidation occurs through several steps of reaction.

#### **Oxido-reductases**

Enzymes that carry out biological oxidation are known as oxido-reductases. They are classified into five groups.

 Oxidase : They catalyses the removal of H<sub>2</sub> but uses only O<sub>2</sub> as hydrogen acceptor.

Example : Cytochrome oxidase. phenolase, lactase etc.

2. Aerobic dehydrogenase : They catalyses the removal of  $H_2$  but uses either  $O_2$  or methyle blue, (artificial substances) as hydrogen aceptor.

Example : D-amino-acid oxidase, L-amino-acid oxidase

- 3. Anaerobic dehydrogenase : They catalyses the removal of hydrogen but can not use O<sub>2</sub> as hydrogen acceptor.
- Hydroperoxidase : These enzymes use H<sub>2</sub>O<sub>2</sub> as their substrate.
- 5. Oxygenase : They catalyzes the direct transfer and incorporation of oxygen into substrate molecule.

## Q. Why many steps of reaction is required for biological oxidation?

Ans. If biological oxidation occurs through one step of reaction , tremendous energy is released which is not tolerable for the cell and it will destroy the cell. So many steps of reaction is required.

## Q. Where ATP is formed in biological oxidation reaction?

Ans. In three sites ATP is formed :

- i. Between NADH<sub>2</sub> and FAD, where NADH<sub>2</sub> released  $H_2$  which received by FAD.
- ii. Between the cytochrom B and C
- iii. Between the cytochrome A and A<sub>3</sub>.

#### Oxidative phosphorylation

The transfer of hydrogen to flavoprotein is associated with the formation of ATP from ADP and the further transfer along the flavoprotein-cytochrome system generates 2 more molecules of ATP per pair of proton transfered. This production of ATP coupled to oxidation is called oxidative phosphorylation.

## Enzyme

*Definition* : Enzymes are protein catalists that increase rate of reactions without themselves being changed in the overall process.

(Ref. Lippincott's illustrated reviews of Biochemistry 2nd, P-47)

*or* Enzyme are organic catalysts, produced by the living cells. They only accelarates the reaction not decrease the reaction.

But inorganic catalyst increases or decreases the reaction. Enzymes are protein in nature.

#### Properties of enzymes :

i. Enzymes are heat liable.

- ii. Enzymes are made up of protein.
- iii. It is not dialysable.
- iv. Enzymes remain unchanged after reaction.
- v. Specificity of enzyme.

#### Q. Why enzymes are not dialysable?

Ans. Enzymes are made up of proteins. As proteins can not dialysed. So enzymes are not dialysable.

#### Enzymes specificity :

The enzymes are specific in their reaction. One enzymes can act only on a particular type of substrate just as key fits for a particular lock. e.g. amylase acts on starch, lipase acts on lipids etc.

Stability of enzyme depends upon :

- і. рн
- ii. Temperature
- iii. Concentration of solution.

#### Mechanism of enzyme action :

According to Michaelis and Menten-

- i. Enzyme (E) molecule first combine with the substrate (S) to form transient enzyme-substrate complex (ES).
- ii. It is a loose complex, but not a proper compound.
- iii. The complex soon dissociates to liberate a new substance (i.e. product, P) and the enzyme remains unchanged.

E + S <----> ES ----> E + P

#### Q. Why enzyme produces continually?

Ans. After reaction enzyme becomes old and is not sufficient for further reaction. Then renewal of enzyme or new set of enzyme are must required. For this reason enzymes are produced continually.

*Substrate* : Substance which may be acted upon by an enzyme are called substrate of that enzyme.

#### Classification of enzyme :

Enzymes are broadly classified into six major classes according to the types of reaction catalyzed by them. These are -

 Oxidoreductases : Catalyze oxidation-reduction reactions. Example : Lactate dehydrogenase- Oxidised lactic acid to pyruvic acid.

- 2. *Transferases* : Enzymes of this class catalyze the transfer of a particular group (C, N or P-containing group) from a molecule of one substrate to a molecule of another substrate.

Example :

- i. Transaminase
- ii. Serine hydroxy-methyl transferase etc.

3. *Hydrolases* : These enzymes catalyze the breakdown of their substrates by hydrolysis i.e cleavage of bonds by addition of water.

Example :

- i. Amylase, lipase, pepsin, \*urease etc which causes hydrolysis of food stuffs.
- ii. Urea  $\checkmark$  CO<sub>2</sub> + 2H<sub>2</sub>O
- 4. *Lyases* : Catalyze cleavage of C-C, C-S and certain C-N bonds.

Example :

- i. Carbonic anhydrase catalyses  $H_2CO_3$  to split it into water and carbondioxide.
- ii. Pyruvate decarboxylase :
   Pyruvate Acetaldehyde + CO<sub>2</sub>
- Isomerases : These are the enzyme that catalyzes the interconversion of different isomers. Example :
  - i. *Glucose phosphate isomerase-* which converts Glucose-1-P into Glucose -6-P and vice versa.
  - ii. Methylmalonyl CoA mutase : Methylmalonyl CoA Succinyl CoA
- Ligases: The enzymes that are capable of binding together of two molecules are known as ligases or synthetase i.e catalyze formation of bonds between carbon and O, S, N coupled to hydrolysis of high-energy phosphates.

Example :

- i. Citrate synthetase
- ii. Pyruvate carboxylaase :

Pyruvate +  $CO_2$   $\checkmark$  Oxaloacetate

(Ref. Lippincott's illustrated reviews of Biochemistry 2nd, P-47)

#### **Zymase secretion**

Some extracellular enzymes secreted in active form called zymase secretion.

Example : Ptyaline of saliva.

#### Zymogen secretion

Many enzymes are quite inactive when they are freshly secreted, called zymogen secretion. They require to be activated by other substances.

Example : Pepsinogen, trypsinogen etc.

#### Anti-enzyme

The substances, produced as a result of repeated injection of

certain enzymes in the serum, which prevents the normal activity of particular enzyme.

Example : Antipepsin, Antitripsin etc.

#### Isoenzyme

Iso-enzymes are the multiple forms of a same enzyme. They are physically distinct form of the same catalytic activity. Thus they catalyze the same reaction. So they are called isoenzyme (*also called isozymes*).

*Example* : Creatinine kinase (CK), lactate dehydrogenase (LDH), Glutamate:oxaloacetate transaminaes (GOT). These are commonly used in the diagnosis of *myocardial infarction*.

Many isoenzymes contain different subunits in various combinations. For example, creatine kinase occurs as three isoenzymes-

- i. CK1 = BB
- ii. CK2 = MB
- iii.  $CK_3 = MM$ .

#### Importance :

- i. Diagnosis of myocardial infarction :
  - a. CK2 : Following a myocardial infarction, this isoenzymes appears approximately 4 to 8 hours following onset of chest pain, and reaches a peak in activity at approximately 24 hours.
  - b. Lactate dyhydrogenase : Activity is also elevated in plasma following an infarction, peaking 3 to 6 days after the onset of symptoms. LDH activity is thus of diagnostic value in patients admitted more than 48 hours after the infarction- a time period when plasma CK2 may provide equivacal results.

(Ref. Lippincott's illustrated reviews of Biochemistry 2nd, P-59)

#### Enzyme modifier

The factors which influence or modifies the activity of the enzyme is called enzyme modifier and the reaction is called the enzyme modification reaction. There are two types of modifiers

- i. *Positive modifiers* : The factors which increases the catalytic activity of the enzyme. *Example* : Fe++, Mg++ etc.
- ii. *Negative modifiers* : The factors which decreases the catalytic activity of the enzyme.

#### Factors influence the enzyme activity

- 1. *Enzyme concentration* : More is the enzyme, more is the reaction and more will be the velocity of reaction.
- 2. Substrate concentration : Up to a certain point if the substrate concentration is increased, the speed of enzyme activity is also increased.
- 3.  $P^H$ : Which influences tremendously the chemical reaction of enzyme activity.
- 4. *Temperature* : Within limitation, more is the temperature more is the reaction. (30-40)°C works very efficiently.
- 5. *Influence of metabolic ions* : Many enzymes are inactive unless a cation (magnesium, calcium, zinc etc) is present.
- 6. Effect of hormones : Many hormones can influence the

enzyme activity. For example epinephrine and glucagone enhance the production of cyclic AMP in the liver cells, the cyclic AMP inturn promotes the convertion of enzyme phosphorylase to its active form.

7. Presence or absence of co-enzyme or co-factors.

## Types of enzyme inhibitor

Enzyme inhibitor falls in two groups :

- 1. *Compititive inhibitor*: Those which has structural similarity with the substrate molecule for the active site of the enzyme.
- 2. *Non compititive inhibitor :* Those which are attached not to the active site of the enzyme but to some other site of the enzyme.

#### Co- Enzyme

*Dfinition* : Co-enzymes are a group of chemical substance which are independent in their function. They are the non-protein prosthetic part of the enzyme.

Example : NAD+, FAD+, NADP+, CoA etc.

The properties of co-enzyme are :

- i. Heat stable
- ii. Dialysable
- iii. Non-colloidal
- iv. Non protein.

#### Apo-enzyme

The protein part of an enzyme is called apoenzyme.

#### Holoenzyme

The composite enzyme molecule consisting of the apo-enzyme and the co-enzyme is called holoenzyme.

## **Co-factors**

Co-factors are the divalent metalic ions. These are accessory substance, non-protein in nature which are neccessary for accelerate the enzyme reaction in addition to co-enzyme. Ex.  $Mg^{++}$ ,  $Ca^{++}$  etc.

#### Co-enzyme A

The co-enzyme form of pantothanic acid is called co-enzyme A. It is biologically important as it carries or donates acetyl group with sulfhydril group.

#### Difference between the enzyme & co-enzyme

Enzyme	Co-enzyme
<ol> <li>Enzymes are organic catalyst.</li> </ol>	<ol> <li>Co-enzymes are accessory substances.</li> </ol>
2. It is protein in nature.	2. It is non protein in nature.
3. Enzyme is not dialysable.	3. Co-enzyme is dialysable.
<ol> <li>Heat and pH liable.</li> </ol>	4. Heat and pH stable.
5. Enzyme can act itself.	5. It is required by an enzyme for its catalytic reaction.

## Osmosis

#### Definition :

Migration of solvent from the solution of lower concentration to the higher concentration, when they are seperated by a semipermeable membrane is *called osmosis*.

Rate of osmosis depends upon:

- i. Difference in osmotic pressure of the solution.
- ii. Permeability of the membrane.
- iii. Electrical potential across the membarane and charge upon the wall and pore of the membrane.

#### Osmotic pressure :

#### Definition :

The force (pressure) by which the osmosis occurs is called osmotic pressure.

Physiological importance :

- i. Absorption from the intestine.
- ii. Exchange in the capillary bed.
- iii. Regulation of urine formation.
- iv. Contineous osmotic exchange between plasma and red cells.
- v. Clinical use : Injection of saline are given by way of treatment.
- Q. On which factors osmotic pressure depends and why?

#### Ans.

Osmotic pressure depends upon the total number of the particles per unit volume but not upon the size of the molecules.

Because each particle in a solution exerts, on the average, the same amount of pressure against the membrane.

## Q. Do you think crystaloid particles exert more osmotic pressure than colloid particles, why?

#### Ans.

Yes, crystaloid particles exert more osmotic pressure. Because, ionisable particle exert more osmotic pressure than non-ionised particles.

Q Why ionise particle exert more Osmotic pressure than nonionised particles?

#### Ans.

A substance, which ionises has more number of osmotic particle, so osmotic pressure of ionised substance is more than of non-ionised substance.

#### Relation between osmolality to osmotic pressure

At nomal body temperature, 1 milliosmole per liter concentration is equivalent to 19.3 mm Hg of osmotic pressure.

So, osmotic pressure (mm Hg) = 19.3 x osmolality (milli osmole /L).

## Diffusion

*Definition* : The continual movement of molecules in a solution from a higher concentration to lower concentration is called diffusion.

*Diffusion depends* : Diffusion depends upon the weight and size of the molecules.

Physiological importance of diffusion :

- i. Admixture of food stuffs with digestive juice.
- ii. Absorption from the intestine.
- iii. Exchange between plasma and red cells.
- iv. Exchange in the capillary bed.
- v. Admixture of gases in the lungs.

#### Factors affect the rate of diffusion :

- 1. *Concentration gradient* : The greater the concentration difference, the greater is the rate of diffusion.
- 2. *Molecular diameter* : The less the diameter of the molecule, the greater is the rate of diffusion.
- 3. *Distance between the area :* The shorter is the distance, the greater is the rate of diffusion.
- 4. *Cross section*: The greater the cross section of the chamber in which the diffusion is taking place, the greater is the rate of diffusion.
- 5. *Temperature* : The greater the temperature, the greater is the rate of diffusion.

 $Diffusion \ rate = \underbrace{\frac{\text{Concentration difference x cross sectional area x temperature}}_{\text{Molecular weight x distance}}$ 

## Types of diffusion :

- i. *Simple diffusion*: Some lipid soluble substances (O<sub>2</sub>, CO<sub>2</sub>, alcohol, and fatty acids. can easily diffuse through the cell membrane, called simple diffusion. Here carrier is not required.
- ii. Facilitated diffusion : Some lipid insoluble substance (glucose etc) can diffuse through the lipid matrix of cell membrane with the help of carrier mechanism called facilitsated diffusion. Here carrier is required.

#### Factors essential for facilitated diffusion

- 1. Concentration gradient
- 2. Amount of carrier
- 3. Enzyme.
- 4. The rapidity with which the chemical reaction involved in the mechanism takes place.

#### Difference between the simple and facilitated diffusion

Simple diffusion	Facilitated diffusion
1. Carrier is not needed.	1. Carrier must be present.
2. Rate of diffusion is almost exactly proprtional to the difference of the concen- tration of particle on the two side of the membrane.	2. For a carrier transported substance, rate of diffusion approches a maximum, as the concentration of the substance increase.

#### Difference between the diffusion & osmosis

Diffusion	Osmosis
<ol> <li>Movement of the solute particles take place.</li> </ol>	<ol> <li>Movement of the solvent particle take place.</li> </ol>
<ol> <li>Movement of solute occ- urs from higher to lower. concentration.</li> </ol>	<ol> <li>Movement of solvent from lower to higher concentration.</li> </ol>
3. Semipermeable membrane may or may not present.	<ol> <li>Semipermeable membrane must be present.</li> </ol>
<ol> <li>It depends upon the wt. &amp; size of the solute perticles.</li> </ol>	<ol> <li>It depends upon the number of particles per unit volume of solution.</li> </ol>

## Transport

*Difinition :* It is the bio-physiochemical phenomenon by which different substance are transported from one place to other place in our body.

#### Types of transport :

- 1. Active Trnasport
- 2. Passive Transport

#### **Active Transport**

The movement of substance across the cell membrane against the concentration gradient with active expenditure of energy by the help of carrier called active transport.

The energey is derived from ATP and the carriers are present in the cell membrane.

Each type of carrier transports only a specific substance.

The gradient may be :

- i. Electrical.
- ii. Pressure.
- iii. Concentration
- iv. Speed of flow.

Active transport generally requires :

- i. Active expenditure of energy
- ii. Special carrier system
- iii. Difference of concentration gradient
- iv. Enzymes.

#### Types of active transport -

- i. Primary active transport.
- ii. Secondary active transport.
  - a. Co-transport
  - b. Counter transport.

Active transport is divided into two types according to the source of the energy used to cause the transport. They are called primary active transport and secondary active transport.

(Ref. Guyton & Hall- 11th Edition; Page-52)

#### **Primary Active Transport**

In primary active transport, the energy is derived directly from the breakdown of adenosine triphosphate (ATP) or some other high energy phosphate compound. Transport depends on carrier proteins that penetrate through the membrane ie Na<sup>+</sup>-K<sup>+</sup> pump.

(Ref. Guyton & Hall-11th Edition; Page-53)

## Na<sup>+</sup>-K<sup>+</sup> Pump

Sodium-potassium pump refers to the mechanism that transports sodium ions out of the cells to the exterior and at the same time pumps potassium ions from the outside to the inside. Here two  $K^+$  ions are transferred for each three Na<sup>+</sup> ions.

#### Explanation :

The carrier protein of the N<sup>+</sup>-K<sup>+</sup> pump is a complex of two separate globular proteins, a larger one (molecular weight 100,000) and a smaller one (molecular weight 45,000). The function of the smaller protein is not known. The larger protein has three specific features that are important for function of the pump :

- i. It has three receptor sites for binding sodium ions on the portion of the protein that protrudes to the interior of the cell.
- ii. It has two receptor sites for potassium ions on the outside.
- iii. The inside portion of this protein adjacent to or near to the sodium binding sites has ATPase activity.

When 3 Na<sup>+</sup> ions bind on the inside of the carrier protein and 2  $K^+$  ions on the outside, the ATPase function becomes activated. This then leaves 1 molecule of ATP, splitting it to ADP and liberating a high energy phosphate bond of energy. This energy is then believed to cause a conformational change in the protein carrier molecule, extruding the sodium ions to the outside and potassium ions to the inside.

Importance of Na<sup>+</sup>-K<sup>+</sup> pump : This pump is responsible for-

- i. Maintaining the sodium and potassium concentration difference across the cell membrane
- ii. Establishing a negative electrical potential inside the cells.
- iii. The pump is so imprtant to many different functioning systems of the body such as to nerve & muscle fibres for transmission of impulses, various glands for the secretion of different substances, and all the cells of the body to prevent cellular swelling.

(Ref. Guyton & Hall- 11th Edition; Page-53, 54)

**Primary active transport of calcium**: Calcium ions are normaly maintained at extremely low concentration in the intracellular cytosol of virtually all cells in the body, at concentration about 10,000 times less than that in the extracellular fluid. This is achieved mainly by two primary active transport calcium pumps. *One* is in the cell membrane and pumps calcium to the outside of the cell. The *other* pumps calcium ions into one or more of the internal vesicular organelles of the cell such as into the sarcoplasmic reticulum of muscle cell and into the mitochondria in all cells. In each of these instances, the carrier protein penetrates the membrane from side to side and also serves as an ATPase, having the same capability to cleave ATP as the ATPase sodium carrier protein. The difference is that this protein has a highly specific binding site for calcium instead of sodium.

(Ref. Guyton & Hall- 11th Edition; Page-54)

#### Primary active Transport of Hydrogen ions :

At two places in the body are important primary active transport systems for hydrogen ions. They are -

- 1. In the gastric glands of the stomach .
- 2. In the late distal tubules and cortical collecting ducts of the kidneys.

In the gastric glands, the deep-lying parietal cells have the most potent primary active mechanism for transporting hydrogen ions of any part of the body. This is the basis for secreting hydrochloric acid in the stomach digestive secretions. At the secretory side of the parietal cells, the hydrogen ion concentration can be increased as much as a million fold and then released in association with chloride ions in the form of hydrochloric acid.

In the renal tubules there are special intercalated cells in the late distal tubules and cortical collecting ducts that also transport hydrogen ions by primary active transport. In this case, large amounts of hydrogen ions are secreted into the tubules to eliminate them from the body for the purpose of controlling the blood hydrogen ion concentration. The hydrogen ions can be secreted against a concentration gradient of about 900 fold.

At many other points in the body, hydrogen ions are transported by secondary active transport, but in these instances, they usually are transported against far less concentration gradients, such as 4 to 1 up to 10 to 1.

(Ref. Guyton & Hall- 11th Edition; Page-54)

#### Secondary active transport

In secondary active transport, the energy is derived secondarily from energy that has been stored in the form of ionic concentration differences between the two sides of a membrane, created in the first place by primary active transport. Transport depends on carrier proteins that penetrate through the membrane.

Two types :

- 1. Co-transport
- 2. Counter transport.

(Ref. Guyton 10th Edition; Page-49)

#### Co-transport :

When sodium ions are transported out of cells by primary active transport, a large concentration gradient of sodium usually develops very high concentration outside the cell and very low concentration inside. This gradient represents a storehouse of energy because the excess sodium outside the cell membrane is always attempting to diffuse to the interior. Under the appropriate conditions, this diffusion energy of sodium can literally pull other substances along with the sodium through the cell membrane. This phenomenon is called co-transport; it is one form of secondary active transport. For sodium to pull another substance along with it, a coupling mechanism is required. This is achieved by means of still another carrier protein in the cell membrane. The carrier in this instance serves as an attachment point for both the sodium ion and the substance to be co-transported. Once they both are attached, a conformational change occurs in the carrier protein, and the energy gradient of the sodium ion causes both the sodium ion and the other substance to be transported together to the interior of the cell.

#### (Ref. Guyton & Hall- 11th Edition; Page-54)

**Co-Transport of Glucose and Amino Acids along with Sodium** *Glucose and many amino acids* are transported into most cells against large concentration gradients; the mechanism of this is entirely by the co-transport mechanism. The transport carrier protein has two binding sites on its exterior side, one for sodium and one for glucose. Also, the concentration of sodium ions is very high on the outside and very low inside, which provides the energy for the transport. A special property of the transport protein is that the conformational change to allow sodium movement to the interior will not occur until a glucose molecule also attaches. But when they are both attached, the conformational change takes place automatically, and both the sodium and the glucose are transported to the inside of the cell at the same time. Hence, this is a sodium-glucose co-transport mechanism.

Sodium co-transport of the amino acids occurs in the same manner as for glucose, except that it uses a different set of transport proteins. Five amino acid transport proteins have been identified, each of which is responsible for transporting one subset of amino acids with specific molecular characteristics.

Sodium co-transport of glucose and amino acids occurs especially in the epithelial cells of the intestinal tract and renal tubules to aid in the absorption of these substances into the blood.

#### (Ref. Guyton & Hall- 11th Edition; Page-54)

Other important co-transport mechanisms in at least some cells include co-transport of chloride ions, iodine ions, iron ions, and urate ions.

(Ref. Guyton & Hall- 11th Edition; Page-54)

#### Counter Transport

In counter-transport, sodium ions again attempt to diffuse to the interior of the cell because of their large concentration gradient. However, this time, the substance to be transported is on the inside of the cell and must be transported to the outside. Therefore, the sodium ion binds to the carrier protein where it projects through the exterior surface of the membrane, whereas the substance to be counter-transported binds to the interior projection of the carrier protein. Once both have bound, a conformational change occurs again, with the energy of the sodium ion moving to the interior causing the other substance to move to the exterior.

#### (Ref. Guyton & Hall-11th Edition; Page-54)

Sodium Counter-Transport of Calcium and Hydrogen ions : Two especially important counter-transport mechanisms are sodium-calcium counter-transport and sodium-hydrogen counter-transport. Calcium counter-transport occurs in all or almost all cell membranes, with sodium ions moving the interior and calcium ions, to the exterior, both bound to the same transport protein in a counter-transport mode. This is in addition to primary active transport of calcium that occurs in some cells.

Sodium-hydrogen counter-transport occurs in several tissues. An especially important example is in the proximal tubules of the kidneys, where sodium ions move from the lumen of the tubule to the interior of the tubular cells, whereas hydrogen ions are counter transported into the lumen. This mechanism is not nearly so powerful for concentrating hydrogen ions as is the primary active transport of hydrogen that occurs in some of the more distal renal tubules, but it can transport such large numbers of hydrogen ions that it is nevertheless a key to hydrogen ion control in the body fluids.

(Ref. Guyton & Hall- 11th Edition; Page-55)

Other important Counter-Transport Mechanism :Other counter-transport mechanisms include cation exchane of calcium or sodium ions on one side of the membrane for magnesium or potassium ions on the other side and anion exchanges of chloride ions moving in one direction for bicarbonate ions or sulfate ions moving in the other direction.

(Ref. Guyton 10th Edition; Page-47,49)

#### **Passive transport**

The movement of substance across the cell membrane along the concentration gradient is called passive transport.

#### The gradients are-

- i. Osmotic pressure
- ii. Hydrostatic pressre
- iii. Gradient of concentration and potential.

#### Tyses :

- i. Diffusion.
- ii. Osmosis.
- iii Filtration.
- iv. Flow due to gravity.

#### Difference between the active & passive transport

Active	Passive
<ol> <li>Movement occurs against concentration gradient.</li> </ol>	1. Movement occurs towards concentration gradient
2. Energy is required.	2. Energy is not required.
<ol> <li>It depends upon the availability of enzyme</li> </ol>	3. It depends upon concentration gradient,
and carrier.	hydrostatic pressure and osmotic pressure.
4. Membrane always necessary.	<ol> <li>Membrane may or may not present.</li> </ol>

#### Filtration

It is the process by which undissolved particles are separated from a liquid through a membrane as a result of mechanical force (filtering force).

#### Filtering force :

- i. Gravity.
- ii. Hydrostatic pressure.

#### Filtering membrane :

- i. Cloth
- ii. Filter paper
- iii. Membrane

#### Filtration depends on :

- i. Pressure difference across the filtering membrane.
- ii. Pore size of the membrane
- iii. Osmotic pressure of the fluid to be filtered

#### Improtance :

- i. Absorption from the small intestine.
- ii. Passage of water, salts, food stuff etc. from the blood stream to the tissue fluid.
- iii. Formation of urine
- iv. Formation of CSF.

#### Ultrafiltration

It is the process by which small crystalloid molecules are separated through a filtering membrane is called ultrafiltration. Such filtration takes place in the separation of plasma proteins from other plasma solutes in the formation of urine.

#### Dialysis

*Definition*: The process by which the more diffusible materials (crystaloid) can be separated from non-diffusible materials (colloid), called dialysis.

Importance of dialysis : Dialysis plays an important role in-

- i. Absorption of food stuff from GIT : During absorption from the intestine bigger food particles are held back.
- ii. Fluid exchange between blood capillaries and interstitial space : When blood passes through the capillaries the bigger particles albumin, globulin etc. are not allowed to pass into the tissue fluid and are held back into the blood.
- iii. In addition, waste products of metabolism accumulates in the body in "renal failure"; dialysis of blood is done through the artificial kidney to remove these waste products clinically.

#### Surface Tension

*Definition* : It is the tension which develops on the free surface of the liquid and gases. It is due to the inter-attracting forces in between atoms or molecules.

Importance of Surface tension :

- i. The globular shape of an oil drop in water and fat particles in the milk is due to surface tension.
- ii. Bile salts reduce the surface tension of fat converting into an emulsion.
- iii. The formation of cell membrane is partly due to surface tension of cell cytoplasm.

## Substance decreases the surface tension :

- i. Bile salts
- ii. Ammonia

iii. Organic substances

## iv. Strong mineral acids.

Substance increases the surface tension : Inorganic salts generally raise the surface tension.

## Interfacial tension

It may be defined as the surface tension exist at the surface of contact (interface) between two immiscible liquids and between a solid and a liquid.

#### Absorption

It is the process by which the substances enter into the cell, called absorption.

#### It depends upon -

- i. Carrier
- ii. Surface area of absorbing membrane.

#### Adsorption

It is the process by which the substances tends to the surface of the cell not into the cell.

Adsorbate : Any substance that adsorbed is called adsorbate.

Adsorbent : A substance that adsorbs is called adsorbent.

## Adsorption occurs due to -

- i. Surface tension
- ii. Residual valency
- iii. Electrical state.

## Importance of adsorption.

- i. Enzyme action.
- ii. Transport of substance in blood
- iii. The combination between toxin and anti-toxin, by which they neutralized each other is an adsorption phenomenon.

#### Difference between the adsorption & absorption.

#### Adsorption

Absorption

1. It is a process of penitration

across the membrane.

- It is a peculiar form of combination in which substance adhere together on their surface.
- 2. It is reversible. 2. It is not reversible.
- . 3. No chemical reaction occurs. 3. Chemical reaction occurs.

#### Isotopes

*Definition* : The elements having the same atomic number but different atomic weight is called isotopes.

*Example* :  $H_2O$ ,  $D_2O$  etc.

It depends upon the number of neutron.

- Hydrogen have two isotopes :
- i. Duterium.
- ii. Tritium.

Hydrogen, duterium and tritium having same atomic number 1, but having different atomic weight respectively as 1, 2 and 3.

## Classification of isotopes :

Isotopes are of two types :

- 1. *Natural Isotopes :* The isotopes which remain as mixture in the all natural sources and can be isolated by careful fractionation. Example : H<sub>2</sub>O, D<sub>2</sub>O.
- 2. *Radio active Isotopes :* The isotopes which break down constantly into more stable atoms. Ex. Uranium, radi-um etc. It can be prepared artificially by bombardment with a cyclotron or by uranium pile.

## Atomic weight

It is the number of weight bearing particles (proton & neutron) present in the atom.

#### **Atomic Number**

It is the number of proton present in the atom. Weight bearing particles : Proton & neutorn. Weight loss particles : Electrone.

#### Physiological importance of isotopes

A. By dilution of radioactivity added :

- i. Measurement of red cell volume with <sup>51</sup>Cr lebelled red blood corpuscles.
- ii. Determination of plasma volume with <sup>131</sup>I lebelled serum albumin.
- iii. Determination of total body water with <sup>131</sup>I.
- B. Study based on distribution of particular isotopes :
  - i. Uptake of <sup>131</sup>I by thyroid gland in thyroid function test.
  - ii. Uptake of <sup>32</sup>P by malignant tissue in its tracing in the body.
- C. As a radiation source in medicine.
  - i. <sup>131</sup>I in the treatment of hyperthyroidism and thyroid cancer.
  - ii. <sup>32</sup>P in the treatment of leukaemia & polycythemia vera.

#### Atom

It is the finnest particle of an element which does not posses all the properties but take part in chemical reaction.

#### Molecule

It is the finnest particle of an element or compound which posseses all the properties but can not take part in chemical reaction.

**Isober** : The elements having the same atomic weight but different atomic number is called Isober.

**Valency**: The combining power of one atom of an element (or a radicale), that of the hydrogen atom being the unit of comparison.

#### Half life

The half life of a radioactive isotope is the time taken by the radiation to come down to half the original strength.

The stability of a radio-active isotope is measured by its half life.

## Difference bewtween atom & molecule

Ate	2 <i>m</i>	Molecule
1.	Smallest particle of an element.	<ol> <li>Smallest particle of an element or compound.</li> </ol>
2.	Cna not exist in the free state.	2. Can exist in free state.
3.	Can take part in chemical reaction.	3. Can not take part in chemical reaction.

## Isomer & Isomerism

#### Isomer

*Definition* : It is the state of compounds having same number of atoms but different atomical arrangement.

**Isomerism**: It is the phenomenon where different chemical compounds having same molecular formula but differ in the atomic arrangement of the molecule.

#### Types of Isomer :

a. *Sterio-isomer* : Compounds which are identical in composition and differ only in spatial configuration are called sterio-isomers.

Two such isomers of glucose; one of which is the mirror immage of other are : L-Glucose and D-Glucose.

b. *Optical isomerm* : It is a phenomenon where isomers show different optical activities i.e. rotating the plain of polarised

light beam due to presence of asymmetric carbon atoms.

- i. *Dextro-rotatory* : A compound which causes rotation of polarised light to right or clock-wise is called dextro-isomers or dextro-rotatory.
- ii. *Levo-rotatory* : A compound which causes rotation of polarized light to left or anti-clock wise is called levo-isomer or levo-rotatory.
- *Epimer*: When isomerism occurs around a single carbon atom then it is called epimer.
   Example: Galactose and glucose are epimer to each other.

#### Asymmetric atom

A atom to which four different atom or groups of atoms are attached is called asymmetric atom.

Example : Glyceraldehyde.

#### Specific rotation

A constant rotation is called specific rotation.

#### Mutarotation

*Muta* means change and *rotation* means rotated form, So change in specific rotation is called mutarotation.

When an aldohexose is first dissolved in water, its optical rotation gradually changes until a constant rotation characteristic of sugar is reached due to changes of alpha to beta form and vice versa. This phenomenon is known as mutarotation.

## Carbohydrate

*Definition:* Carbohyrates are polyhydric alcohol having aldehydes or keton groups or compounds which yeilds those derivatives on hydrolysis. Here hydrated carbon is present. Example : Glucose, Fructose, Starch etc.

#### Sources of carbohydrate

Carbohydrates are widely distributed in nature. There are three main sources-

- 1. Sugars : Mono & Disaccharides- Glucose, fructose, galactose, sucrose, lactose etc.
- 2. Starch : Present in cereals, roots, tuber, plant stems etc.
- 3. Cellulose : Vegetable, cereals, fibrous lining of fruits.

#### Classification

Classified into four major groups according to the number of simple sugar unit present in the molecule. Such as -

A. Monosaccharides (simple sugar) : Those carbohydrates which can not be hydrolysed further into a simpler form. They contain only one unit of simple sugar. Example : Glucose, Fructose etc.

## Monosaccharides are subdivided into-

- 1. Trioses, tetroses, penoses, hexoses or heptoses according to the number of carbon atom present in the molecule.
- 2. Aldoses and ketoses, according to the nature of reducing group (aldehyde or ketone group) they contain.

#### Classification of monosaccharides

		Aldoses	Ketoses
a.	Trioses	Glycerose	Dihydroxyacetone
b.	Tetroses	Erythrose	Erythrulose
c.	Pentoses	Ribose	Ribulose
d.	Hexoses	Glucose	Fructose.

B. *Disaccharides* : Those which yield two molecules of same or different monosaccharides when hydrolysed.

*Example : Sucrose* (Glucose + Fructose), *Maltose* (Glucose + Glucose), *Lactose* (Glucose + Galactose).

#### Types :

- 1. Reducing : Maltose, Lactose.
- 2. Non-reducing : Sucrose.
- C. *Oligosaccharides* : Those which yield 3-10 monosaccharide units on hydrolysis.

Example : Rabinose, Raffinose etc.

#### D. Polysaccharides :

Those which yield an indefinetely larger number (more than 10 melecules) of monosaccharides on hydrolysis.

Example : Starches, dextrins, glycogen etc.

#### Classification of polysaccharide :

- 1. According to the nature of constituents
  - a. *Pentosans* : Those, made up of pentose units. Example : Inulin etc.
  - b. *Hexosans*: Those, made up of hexose units. Example : Starch, dextrin etc.
- 2. According to the nature of content
  - a. *Homopolysaccharides* : When they are made up of same kinds of monosaccharides. Example : Starch, dextrin, cellulose, glycogen etc.
  - b. *Heteropolysaccharides*: When they are made up of two or more kinds of monosaccha-rides or their derivatives.
     Example : Heparin, hyaluronic acid, chondroitin

sulphate etc.

#### Difference between aldose and ketose :

	Aldose	Ketose
1.	Aldose derived from primary alcohol.	<ol> <li>Ketose derived from secondary alcohol.</li> </ol>
2.	Aldose contains aldehyde group.	<ol> <li>Ketose contains Ketone group.</li> </ol>

#### Sugar Unit :

The sugar containing 3-7 carbon atom is called sugar unit.

#### Starch

- 1. *Definition* : It is the most important food source of carbohydrate and is found in cereals, potatoes, legumes and others.
- Structure : It is formed of a α-glucosidic chain. This produces only glucose on hydrolysis. So, it is called glucosan.

It has 2 parts :

- i. Amylose (15-20%) : non branched hellical structure.
- ii. Amylopectin (80-90%) : consists of branched chains composed of 24-30 glucose residues united by 1→ 4 linkages in the chains and by 1→ 6 linkages at the branch points.

#### Glycogen

- 1. *Definition* : It is the storage form of poly saccharide in animal body.
- 2. Structure : It has a branched structure with straight chains of  $11-18-\alpha$ -D-glucopyranose residues with branching by means of a  $(1 \rightarrow 4)$  glucosidic linkage.

#### Cellulose

- 1. Definition : It is the main carbohydrate component of plant.
- 2. Structure : It consists of  $\beta$ -D-glucopyranose units linked by  $\beta$  (1 $\rightarrow$  4) bonds to form long, straight chains strengthened by cross linked hydrogen bonds. It is not digested by human body due to absence of hydrolase which break down  $\beta$  linkage.

3. Importance : It is an important source of bulk in the diet, thereby stimulating peristalsis and elimination of stool.

## Glycosides

Glycosides are compounds formed from a condensation between a monosaccharide, or monosaccharide residue and the hydroxyl group of a second compound that may or may not (incase of glycone) be another monosac charide. *Example*: cardiac glycoside.

**Deoxy sugar :** Are those in which a hydroxyl group attached to the ring structure has been replaced by a hydrogen atom. *Example* : Deoxyribose.

Amino sugar : Sugars containing an amino group are called amino sugars.

*Example* : D-glucosamine, D-Galactosamine, and D-mannosamine etc.

(Ref. Herper's)

## Q. Which one is the most important in monosaccharide and polysaccharide?

- a. In Monosaccharide : Hexoses are the most important class of monosaccharides. They are widely distributed in nature either in free form or in combination with other molecules. Biologically important hexoes are glucose fructose, galactose and manose, among them glucose is most important.
- b. In polysaccharide : Glycogen is the most important plysaccharide for the human being because via the pathway of glycogenolysis it liberate glucose units for ready energy.

#### Important dietary carbohydrate :

- 1. Monosaccharides : Glucose, fructose
- 2. Disaccharides : Maltose, sucrose, lactose
- 3. Polysaccharides : Starch, glycogen, cellulose.

## Properties of carbohydrate

- 1. They are colourless crystaline compounds having a sweet taste.
- Chemically they are derivatives of polyhydric alcohols. The aldoses are derived from primary alcohols and ketoses from secondary alcohols.
- 3. *Formation of ester* : By virtue of alcohol group they easily form esters with acids.
- 4. *Reducing power*: By virtue of the aldehyde or ketone groups, the simple sugars are powerful reducing agent.
- 5. *Isomerism*: Due to the presence of asymmetric carbon atoms in the molecule, monosaccharides may remain in many isomeric forms.
- 6. *Optical rotation :* Simple sugars rotates in the plane of polarised light and therefore may exists in either dextro or levo forms.
- 7. *Condensation :* Simple sugars condense and form bigger carbohydrate molecules e.g polysaccharides.
- 8. Osazone formation : All reducing sugars condense with phenylhydrazine and produce osazone compounds.
- 9. Reduction : The monosaccharides may be reduced to their

corresponding alcohols by reducing agents such as sodium amalgam.

- 10. *Ring structures of sugars :* Sugars not only remain as straight chain compounds but may also remain in the form of rings.
- 11. Fermentation by yeast and other micro-organisms.

#### Functional (biomedical) importance of carbohydrate

- i. Ready source of energy : 4.1 Kcal/gm of carbohydrate.
- ii. It also constitutes the structural material of the organism.
- iii. Acts as important storage of food material of the body i. glycogen.
- iv. Actively concerned with protein and fat metabolism i.e protein and fat metabolized on the back ground of carbohydrate.
- v. Highly specific function :
  - a. Ribose : major constituents of nucleic acids
  - b. Lactose : sugar of milk
  - c. Fructose : give energy to sperm.
- vi. It gives nutrition to the nervous tissue & retina.
- vii. It prevents ketosis.
- viii. Carbohydrate is a protein sparer, that is carbohydrate is burned in preference to the burning of protein.
- ix. Antibodies, clotting factors & peptide hormones contain oligosaccharides.
- Diseases associated with carbohydrate are- diabetes mellitus, galactosaemia, glycogen storage diseases and lactose intolerance.
- xi. Sugar solution is used as I.V fluid.
- xii. Some drugs are chemically glycosides- are used as drug i.e cardiac glycosides, streptomycin.

#### L and D series of the sugars

The orientation of the H and OH group around the carbon atom just adjecent to the terminal primary alcohol (e.g. carbon atom 5 in glucose) determines the family to which the sugar belongs. When the "OH" group on this carbon is on the right, the sugar is the member of D-series, when it is on the left, it is a member of L-series.

#### Q. Why sucrose is a non reducing sugar?

Ans. Sucrose contains glucose and fructose which are linked together through their reducing groups and as a result of this, no reducing group is free and hence it does not respond the test. So sucrose is non reducing.

 ${\bf Glucosan}$  : Glucosan are those which yield only glucose on hydrolysis.

Example : Startch, Glycogen. Dextrin etc.

Fructosan : Fructosan are those which yield only fructose on hydrolysis.

*Example* : Inulin.

Inversion of sucrose : Inversion of sucrose reffers to that

phenomenon when the dextrorotatory property of sucrose solution changes into levorotatory form because of the hydrolysis of sugar into glucose and fructose.

#### Sucrose is an invert sugar - Explain?

Sucrose is dextrorotatory but on hydrolysis it yields a crude mixture (Glucose + Fructose) which is often called invert sugar because the strongly levorotatory fructose inverts the previous dextrorotatory action of sucrose. So sucrose is called invert sugar.

#### **Reducing sugars**

Are those which posses free aldehyde or ketone group called reducing sugar.

*Example* : All monosaccharides, maltose, lactose etc.

#### Properties of monosaccharide

- 1. These are white (colourless) crystaline compound having sweet taste.
- 2. Condensation into polysaccharide.
- 3. They have reducing property due to presence of aldehyde or ketone groups.
- 4. They can produce esters due to presence of alcohol groups.
- 5. Properties not dependent on alcohol or aldehyde or ketone groups
  - a. Formation of alcohol.
  - b. Formation of osazone sugar.
  - c. Formation of glycosides.
  - d. Optical rotation and isomerism.
  - e. Fermentation by yeast or bacteria.
  - f. Formation of amino sugar.
  - g. Acetylation.
  - h. Oxidation.

#### Glucose

#### Properties :

- i. Glucose or grap sugar is the most important of the monosaccharides because of its utility. It is sometimes called dextrose due to its dextrorotatory property.
- ii. Glucose is present in the body as glucosc phosphate, in milk sugar as lactose (glucose & galactose), in combination with proteins as glycoproteins, glucosamine.
- Some polysaccharides such as glycogen (liver and muscle), starch (plant), dextrin and cellulose (supporting structure of plant), yield glucose on hydrolysis.
- iv. When it is completely burn to  $CO_2$  and  $H_2O$  energy is evolved.
- v. Structure of glucose: Glucose may be present in two forms
  - a. A straight chain compound (L & D form).
  - b. A cyclic compound ( a & b form).

#### Dis-similarities between glucose and fructose

	Glucose		Fructose.
1.	Aldose.	1.	Ketose.
2.	Functional group lst carbon atom.	2.	Functional group 2nd carbon atom.
3.	Seliwanoff test (-ve) negative.	3.	Seliwanoff test (+ve) positive.

## Enediol :

t is the active form of that sugar when treated with alkali, it acts as a strong reducing agent. It is formed by enolization process.

## Lipids

*Definition :* Are ester like compounds of fatty acids which are insoluble in water but soluble in fat solvents, e.g. chloroform, ether, benzene etc.

## **Properties of lipid** :

- A. Physical :
  - 1. *Solubility* : They are insoluble in water but readily soluble in fat solvents i.e ether, chloroform, benzene etc.
  - 2. They are tasteless, odorless, colourless and neutral in reaction.
  - 3. *Melting point*: Different fats have different melting points. Thus beef fat melts at 49.5°C, human fats melts at about 17°C.
  - 4. Specific gravity : It is less than 1 i.e lighter than water.
  - 5 Neutral fats are non volatile and may be crystalized.
  - 6. *Spreading and surface tension*: When a liquid fat is poured on water, it spreads uniformly over the surface of water and reduces the surface tension of water.

#### B. Chemical :

- 1. *Hydrolysis* : By boiling with acids or alkalis or by applying superheated steam fats can be completely hydrolysed into fatty acids and glycerol.
- 2. *Rancidity*: Rancidity is a chemical changes that results in unpleasant odors and tastes in a fat. This is due to partial hydrolysis causing liberation of volatile fatty acids e.g Butyric acid from butter.
- 3. *Hydrogenation:* Unsaturated fatty acids are usually liquid at room temperature and commonly called oils. These oils can take hydrogen at their double bond in presence of catalyst (nickle) and converted into solid fats. This process is called hydrogenation or hardening of fats.
- Oxidation : Unsaturated fatty acids when react with oxygen to form a number of substances including aldehydes and ketones which may react further to form complex resinous compound.

#### **Classification of lipid**

Lipids are classified into -

- A. Simple lipid : Esters of fatty acids with various alcohols.
  - *Neutral fats*: Esters of fatty acids with glycerol e.g oil, triglyceride.
  - ii. *Waxes*: Esters of fatty acids with higher alcohols other than glycerol e.g cholesterol ester, vitamin D ester.
- B. Compound lipids : Esters of fatty acids with alcohols and containing other groups e.g PH<sub>4</sub>, sugar, protein, nitrogenous substances etc.

Example : Phospholipids, glycolipids, sulpholipid, lipoprotein etc.

- C. Derieved lipid : Substances derieved from hydrolysis of simple lipid and compound lipid are called derieved lipid.
   e.g, saturated and unsaturated fatty acids, steroids etc.
  - i. Fatty acids :
    - a. Saturated fatty acid : Having no double bond e.g stearic acid, palmitic acid
    - b. Unsaturated fatty acid
      - 1. Monounsaturated fatty acid e.g oleic acid
      - Polyunsaturated fatty acid (Ecosanoids- having 20 carbon atoms)
        - \* Protanoids : Protaglandin, protacyclins, thromboxanes
        - Leukotriens & lipoxins
  - ii. Steriods :
    - a. Hormones : Glucocorticoids, mineralocorticoids, cortical sex hormones
    - b. Steriods : Cholesterol
    - c. Bile acids.

#### **Biological** importance of lipid

- i. Lipids are the most concentrated source of energy. Their calorie value is 9.3 kcal/gm.
- ii. Lipids provide essential fatty acids.
- iii. Lipid acts as reserved food materials.
- iv. Lipid is a structural component of the cell membrane.
- v. Lipid acts as a insulating material.
- vi. Lipid protects the internal organ. e.g kidney etc.

## Fat

Are the esters of fatty acid with glycerol. It is solid at ordinary temperature.

## Oil

Unsaturated fatty acids are usually liquid at room temperature and commonly called oils.

#### Difference between fats and lipids :

Fats	Lipids
1. Fats are esters of fatty acids with glycerol.	<ol> <li>Lipids are ester like com pound of fatty acids.</li> </ol>
<ol> <li>Trihydric alcohol</li></ol>	<ol><li>Alcohol present is</li></ol>
present.	more than trihydric.
3 lt contains short chain	<ol> <li>It contain long chain</li></ol>
(upto 10-12 c) of fatty	(more than 12 c) of
acids.	fatty acid.

#### Iodine number

Means the number of grams of iodine absorbed by 100 gm of fat.

*Importance :* It indicates the degree of unsaturation in the fat. Higher the iodine number the more will be the degree of unsaturation.

**Saponification** : Hydrolysis of fat by an alkali is called saponification.

Saponification number : Means the amount of KOH in mg require to saponify completely 1 gm of fat.

*Importance :* It gives an idea, about average chain length of the fatty acids present in the fat.

## Alkali hydrolysis :

The process of saponification is called alkali hydrolysis.

#### Soap :

After saponification glycerol is released and fatty acid will produce salts with alkalis. These salts are known as soap.

#### Acid number :

It may be defined as mg of KOH required to neutralize the free fatty acid present in 1 gm of fat.

#### Fatty acid

Definition : Fatty acids are obtained from hydrolysis of fats.

#### Classification :

- A. On the basis of saturation : Pl see above
- B. On the basis of carbon atom :
  - 1. *Long chain fatty acids :* Fatty acids containing more than 10 carbon atoms.
  - 2. *Short chain fatty acids :* fatty acid containing less than 10 carbon atoms.

#### Fatty acids of physiological importance :

- 1. Palmatic acid : 16 Carbon atom (Saturated)
- 2. *Stearic acid* : 18 Carbon atom (Saturated)
- 3. Oleic acid : 18 Carbon atom (Unsaturated).

#### Essential fatty acids

Essential fatty acids are those which cannot be synthesized in our body hence are supplied in the diet.

#### Example :

- i. Linoleic acid
- ii. Linolenic acid
- ii. Arachidonic acid.

#### Properties :

- 1. Essential faty acid of vegetable oils have low melting point & iodine number.
- They become saturated fatty acids on hydrogenation and the oils bbbecomes solid fats.

#### Importance :

- i. Essential fatty acids reduces the plasma cholesterol concentration, thus it can reduce the coronary artery diseases.
- ii. Precursor of eicosanoids (prostaglandin, thromboxane, leukotrienes)
- iii. For synthesis of structural lipid.
- iv. Concerned with optimum gonadal activity and it is directly related to sterility.
- v. The deficience of essential fatty acid in the diet of baby cause eczema.

### Q. Why essential fatty acids are called so?

Ans. They play great role in proper functioning of many metabolic processes and are not synthesized in the body in quantities sufficient to meet the daily requirement, there fore it must be supplied in diet. So it is called essential fatty acids.

*Steroids* : Substances possessing cyclopentanoperhydrophenanthrene nucleus are called steroids.

#### Phospholipid

- 1. *Definition* : Phospholipids are composed of fatty acids, glycerol, phosphoric acid and in most cases a nitrogenous base.
- 2. Example:
  - a. Phosphatidyl inositol
  - b. Phosphatidylglycerls
  - c. Phosphatidyl serine
  - d. Phosphatic acid
  - e. Phosphatidylethanolamine
  - f. Sphingomyelins
  - g. Lysophospholipids
  - h. Plasmalogen.
- 3. Distribution : Widely present in the
  - a. Nervous tissue
  - b. Brain
  - c. Liver
  - d. Kidney
  - e. Pancrease
  - f. Heart etc.
- 4. Importance :
  - a. Takes part in the synthesis of biological membranes
  - b. Helps in blood coagulation
  - c. In bile, it keeps the cholesterol in solution
  - d. In the form of surfactant, it help in expansion of lung
  - e. Act as a second messenger of different hormones
  - f. Phospholipids of myelin sheath provides insulation.

#### Lipoprotein

- 1. *Definition* : Lipoproteins are conjugated proteins consisting of simple proteins combined with lipid components-triglycerides, cholesterol, and phospholipids.
- 2. Classification:
  - a. Chylomicron
  - b. Very low density lipoprotein VLDL
  - c. Low density lipoprotein LDL
  - d. High density lipoprotein HDL
- 3. Normal blood values :
  - a. LDL : <150 mg/dl
  - b. HDL : 55 65 mg/dl

#### Clinical importance :

1. LDL : Transport cholesterol from liver to peripheral tissues. It may be responsible for atherosclerosis.

- HDL : Transport cholesterol from peripheral tissues to liver. It act as a scavanger. Thus it can protect coronary heart disease.
- 3. VLDL : Transport exogenous triglycerides from liver to peripheral tissues for energy.
- 4. Chylomicron : Transport exogenous triglycerides from intestine to liver through lymphatic circulation.

#### Apo-lipoprotein

1. *Definition* : The protein moiety of a lipoprotein is known as an apo-lipoprotein. It is proteins or polypeptides.

It constitute about 70% of some HDL and 1% of chylomicrons. Some apolipoproteins are integral and cannot be removed, whereas others are free to transfer to other lipoprotein

2. Types with distribution :

Apo $A_1$ and $A_2$	HDL
Аро В <sub>48</sub>	Chylomicrons
Аро В <sub>100</sub>	VLDL, LDL
Apo C <sub>1</sub> , C <sub>2</sub> , C <sub>3</sub>	Not native to any lipoprotein but in almost all lpoproteins
Apo E	Not native to any lipoprotein but in almost all lpoproteins.

- 3. Functions of lipoproteins :
  - a. Structural component of lipoprotein
  - b. Act as enzyme co-factors e.g apo-C-II for lipoprotein lipase.
  - c. Act as a ligands e.g binding and recognition of lipoprotein with corresponding receptors.

## Cholesterol

- 1. *Definition* : It is a complex monohydric alcohol and steroid in nature.
- 2. Cholesterol turnover : 1 gm/day
  - a. Input : 1 gm/day

	<b>D</b> 1	20 C C C C C C C C C C C C C C C C C C C		15	1
1	Endogenous	prroduction	500	mg/	da
	Diraogenous	prioduction	200	mg/	ua

ii. Dietary : 500 mg/day

Food : Egg, milk, liver, brain, skin etc.

- b. Output : 1 mg/day
  - i. Bile salt : 500 mg/day
  - ii. Faecal neutral steroid : 400 mg/day
  - iii. Skin desquamation : 100 mg/day

### Importance :

- 1. Essential constituent of all cells
- 2. Increase the cell membrane permeability
- 3. Precuesors of bbile acids, adrenocortical hormones, sex hormones, vitamin D, and cardiac glycosides
- 4. Increase in plasma cholesterol level (hypercholesterolemia) may cause atherosclerosis.

## Lipid profile

Different types of lipids are present in plasma of blood in different amounts. It is knnown as lipid profile.

1.	Triglycerides	:	<150	mg %
2.	Cholesterol	2	150 - 22	0 mg %
3.	HDL	:	>40	mg %
4.	LDL	1	<130	mg %
5.	LDL : HDL	:	2.5 : 1	
6.	Cholesterol : HDL	:	3.5 : 1	

#### Prostaglandin

- 1. *Definition* : Prostaglandins are 20 carbone hydroxy fatty acids containing a five membered ring.
- Synthesis : Membrane phospholipids

Phospholipase

Arachidonic acid

Cyclooxygenase

#### Prostaglandins

- 3. Distribution :
  - a. Lungs
  - b. Kidney
  - c. Thyroid
  - d. Spleen
  - e. Brain
  - f. Iris
  - g. Endometrium etc.
- 4. Functions:
  - a. Causes vasodilation  $\rightarrow$  decreases blood pressure
  - b. It mediates inflammation
  - c. It causes pain sensation
  - d. It causes contraction of muscle
  - e. It Inhibits gastric secretion
  - f. It takes part in spermatogenesis.

## Protein

Definition : Protein is complex organic nitrogenous compound composed of amino acids linked together by peptide bonds. Sources : Meats, fish, milk, egg, pulses and vegetables etc.

#### Classification

Protein is classified into three groups (broad based types) -

- A. Simple protein : The protein which gives only amino acid on hydrolysis is called simple protein. These are pure protein and have no nonprotein group in the molecules. Example : Albumins, globulins etc.
- B. Conjugated protein : These contains amino acid units with one or more additional nonprotein (prosthetic) group or groups.

Example : DNA, RNA, Hb etc.

C. Derieved protein : These proteins are not found in nature as such. They are as products of hydrolysis of the native protein molecules. The gradual stages are as follows: Protein  $\rightarrow$  Proteoses  $\longrightarrow$  Peptones  $\longrightarrow$  Peptides  $\longrightarrow$ 

> polpeptide.

Polar or salt linkage : When two amino acids join together without forming peptide linkage, they are known to form polar linkage.

#### On the basis of source :

- 1. First class proptein : Proteins of high biological value which contain all the essential aminoacids. Source : Animal sources such as meat, fish, egg, milk.
- Second class protein : Protein of low biological value 2. which are poor in essential amino acids. Source : Plant sources such as vegetables- pulses, cereals, leans, nuts, oil seed etc.

#### On the basis of biological function :

- Catalytic 1. : enzymes
- 2. Contractive : actin, myosin
- 3. Hormonal : Insulin
- 4. Structural : collagen, elastin
- 5. Transport : albumin, Hb, lipoprotein, transferrin
- : calmodulin 6. Regulatory
- 7. Gene regulatory : histones
- 8 Protective : fibrin.

Zwitterion : At a certain pH the number of positive charge is equal to the number of negative charge and protein remains as zwitterion form.

Denaturation of protein : Means unfolding of the protein molecule.

#### Chromoproteins :

- i. Haemoglobin
- Myoglobin ii.
- iii. Cytochrome
- iv. Catalase.

#### Properties of protein :

- I. General properties :
  - Nature 1.

2

- : Colloidal Taste : Generally tasteless
- Odour : Odourless
- 3.
- 4. Molecular weight : High
- 5. Crystalysation : Globular proteins mostly crystalline.
- Solubility : Different for different proteins. They are 6. generally soluble in water & salt solution and insoluble in fat solvent.
- 7. Acidity & alkalinity : All proteins can behave like acids or bases depending on whether the medium is basic or acidic.
- 8. Isoelectric point : Each protein has a particular isoelectric point (pH) at which it can be precipitated.
- 9. Diffusibility : Most proteins cannot pass through the membranes of the cells.
- 10. Protein has no melting point
- 11. Osmotic pressure
- 12. Viscosity.
- II. Chemical properties :
  - 1. Proteins are polymers of amino acids.
  - 2. They are composed of C, H, O, N & sometimes S, P.
  - 3. Denaturation: Proteins undergo a change by the application of heat, addition of acid/alkali and some organic solvents. This is called denaturation of protein.
  - Coagulation : Proteins undergo coagulation by heat and 4. strong acid.
  - 5. Precipitation : Proteins undergo precipitation by metalic salt & alkaline reagent.
  - They give colour reaction-6.
    - i. Biuret test
    - ii. Millons test
    - iii. Xanthoprotein test
  - 7. Hydration.

#### Biomedical importance of protein :

- Structural component of cell membranes, organelles & i. cytoskeletal structures.
- Transport of O<sub>2</sub> by Hb or electrone by the cytochromes. ii.
- Acts as carrier e.g Na+-glucose co transporter. iii.
- iv. Maintenance of fluid balance by serum albumin.
- They help in formation of enzymes & many hormones. V.
- vi. The dietary protein regulates the supply of nitrogen & sulphur.
- vii. They perform hereditary transmission by nucleoprotein.
- viii. They help in blood clotting by prothrombin & fibrinogen.
- Some proteins are present as storage e.g ferritin is stored ix. being formed by protein & iron.
- They help in movement of muscle fibre by actin & Χ. myosin.
- Source of energy : 4.2 Kcal/gm. xi.

xii. Diseases associated with protein maturation are Dire consequences, Ehlers-Danlos syndrome & scurvy.

#### Amino Acid

*Definition*: Amino acids are organic acid in which one or more hydrogen atoms are replaced by amino  $(-NH_2)$  group. Thus it contains at least a free amino  $(-NH_2)$  group, and a carboxyl (-COOH) group.

In an  $\alpha$  amino acid, both amino and carboxyl groups are attached to the same ( $\alpha$ ) carbon atom.

#### Classification of amino acid :

Amino acids are classified into two catagories :

#### A. According to their functional group

- 1. *Neutral amino acids* : monoamino-monocarboxylic acid i.e glycin, alanine etc.
  - i. Aliphatic series
    - a. Normal series i.e glycine
    - b. Hydroxy series i.e serine
    - c. Sulphar containing i.e methionine
  - ii. Aromatic series
    - a. Normal series i.e tryptophan
    - b. Hydroxy series i.e tyrosine
- 2. Acidic amino acids : monoamino dicarboxylic acid i.e aspartic acid, glutamic acid etc.
- 3. *Basic amino acids* : diaminomono-carboxylic acid i.e agrinine, lysine etc.
- Imino acids : containing imino group but no amino group) i.e proline.
- B. Accroding to their biological value :
  - a. Essential amino acids : i.e isoleucine, valine etc.
  - a. Semi-essential amino acids : i.e arginine, histidine
  - b. Non-essential amino acids : i.e glycine, alanine, asparate, glutamate etc.

#### Essential amino acid

Essential amino acids are those which can not be synstheized by the body but needed for the growth of the body and, hence they have to be supplied in the diet. There are eight essential amino acids.

#### Example :

- 1. Valine
- 2. Leucine
- 3. Isoleucine
- 4. Lysine
- 5. Threonine
- 6. Methionine
- 7. Phenylalanine.
- 8. Tryptophan

## Non essential amino acids

Non essential amino acids are those which are synthesized by the body.

## Example :

- i. Glycine.
- ii. Alanine.
- iii. Serine.
- iv. Cysteine.
- v. Asparagine.
- vi. Tyrosine.
- vii. Aspartic acid.
- viii. Glutamic acid.
- ix. Proline
- x. Glutamine.

#### Semiessential amino acids

Those amino acids which are partly synthesized by the body are called semiessential amino acids.

Example : arginine, histidine.

## Nucleoprotein

- 1. *Definition* : Nucleoproteins are conjugate proteins containing neuclic acid as non protein prosthetic group and a base protein like protamines or histones.
- 2. Example :
  - i. Nucleoprotamine
  - ii. Nucleohistones
- 3. Distribution :
  - i. In ribosomes of all living cells
  - ii. Virus consists entirely of nucleoproteins.
- 4. Compositions of nucleoproteins :
  - A. Basic proteins (protamines or histones)
  - B. Nucleic acid (DNA, RNA)
    - i. Phosphoric acid
    - ii. Nucleosides
      - a. Bases
        - 1. Purine : adenine, guanine
        - 2. Pyrimidine : cytosine, thymine (DNA)
        - uracil (RNA)
      - b. Sugar :
        - 1. D-ribose (RNA)
        - 2. 2-Deoxy D-ribose (DNA).

#### Nucleic acids

Nucleic acids are high molecular weight polyanionic molecules composed of nucleotides i.e nucleic acids are polynucleotides

Types : It is two types -

- i. Deoxyribose nucleic acid DNA
- ii. Ribose nucleic acid RAN.

#### Component of nucleic acid :

#### Nucleic acid :

- i. Phosphoric acid
- ii. Nucleosides
  - a. Bases

- 1. Purine
  - Major :
  - \* Adenine
  - \* Guanine
  - Intermediate :
  - \* Xanthhine
  - \* Hypoxanthine
  - 2. Pyrimidine
    - \* Cytosine
    - \* Thymine (DNA)
    - \* Uracil (RNA)
- b. Sugar
  - D-ribose (RNA)

2-Deoxy D-ribose (DNA).

#### Nitrogenous base

Nitrogenous bases are of two types -

- i. *Purine base* : Adenine and guanine both are present in DNA & RNA.
- ii. *Pyrimidine base* : Cytosine and uracil are present in RNA. Where as cytosine along with thiamine are present in DNA.

#### Nucleotides

Nucleotides are the monomer units or building blocks of nucleic acids - serve multiple additional functions. They form a part of many coenzymes and serve as donors of phosphoryl groups (i.e ATP or GTP), of sugars ( i.e UDP- or GDP sugars), or of lipid (i.e CDP-acylglycerol).

These are the structural unit of nucleic acid containing a *purine or pyrimidine* base attached to a sugar- *ribose or deoxyribose* by a a glycosidic linkage and combined with *phosphoric acid*.

#### Example :

Nucleotide	Base	Nucleosides
1. Adenosine monophosphate- AMP	Adenine	Adenosine
2. Guanosine monophosphate- GMP	Guanine	Guanosine
3. Uridine monophosphate- UMP	Uracil	Uridine
4. Cytidine monophosphate- CMP	Cytosine	Cytidine
5. Thymidine monophosphate-TMP	Thymine	Thymidine

(Ref. Harper's Illustrated Biochemistry 26th ed, Page-286,288)

#### Importance of nucleotides :

- 1. They are the building block or monomer of nucleic acids
- 2. They can act as a co-enzymes i.e NAD, FAD, FMN
- 3. They can act as intracellular second messenger i.e cAMP, cGMP etc.
- 4. They can store and transfer energy.
- 5. They participate in synthetic process of different biomolecules i.e UDP- in glycogen synthesis
- 6. They participate in different phosphorylation process.

#### Nucleosides

Sugar-base combination is called nucleosides. A nucleosides is composed of a purine or a pyrimidine base to which a sugar (D-ribose or 2-deoxy D-robose) is attached in  $\beta$ -linkage at N<sub>9</sub> or N<sub>1</sub> respectively.

Example with composition :

	a. Nucleosides conta Purine bases	ining Base	Sugar
1.	Adenosine	Adenine	D-ribose
2.	Deoxy adenosine	Adenine	2-Deoxy ribose
3.	Guanosine	Guanine	D-ribose
4.	Deoxy guanosine	Guanine	2-Deoxy ribose
	b. Nucleosides contai Pyrimidine bases :		Sugar
1.	Uridine	Uracil	D-ribose
2.	Cytidine	Cytosine	D-ribose
4.	Deoxy cytidine	Cytosine	2-Deoxy ribose
5.	Deoxy thymidine	Thymine	2-Deoxy ribose

Lipoprotein (Described in lipid section)

#### DNA

It is a complex protein of high molecular weight consisting of deoxyribose, phosphoric acid, and four bases (2 purines and 2 pyrimidines). DNA is found mainly in the chromosomes of the cell nucleus.

Components of DNA :

- i. Sugar : 2-deoxy-D-ribose
- ii. Acid moiety : Phosphoric acid
- iii. Bases :
  - a. Purines : Adenine & Guanine
  - b. Pyrimidines : Cytosine & Thymine

#### Function of DNA :

The genetic information store in the nucleotide sequence of DNA serves two purposes-

- i. It is the source of information for the synthesis of all protein molecules of the cell & organism.
- It provides the information inherited by daughter cells or offspring.

(Ref. Guyton 11th Edition)

#### RNA

RNA (Ribonucleic acid) is a polymer of purine & pyrimidine ribonucleotides linked together by 3, 5, - phosphodiaster bridge. RNA is found mainly in the cytoplasm & nucleolus.

Components of RNA :

- i. Sugar : D-ribose
- ii. Acid moiety : Phosphoric acid
- iii. Bases :
  - a. Purines : Adenine & Guanine
  - b. Pyrimidines : Cytosine & uracil

#### 4.30 Physiology

#### Types of RNA :

- 1. *Messenger RNA (m RNA)*: Which carries the genetic code to the cytoplasm for controlling the formation of the proteins.
- 2. *Transfer RNA (t RNA) :* Which transports activated amino acids to the ribosomes to be used in assembling the protein molecules.
- 3. *Ribosomal RNA (r RNA) :* Which along with about 75 different proteins, forms the ribosomes, the physical and chemical structure on which protein molecules are actually assembled.

Functions of RNA: It is involved in protein synthesis.

(Ref. Guyton 11th Edition; Page 31)

## Transcription

Assembly of the RNA molecule from activated nucleotides using the DNA strand as a template- the process is called transcription.

(Ref. Guyton & Hall-11th edition; Page-33)

Assembly of the RNA molecule is accomplished by DNA under the influence of the enzyme RNA polymerase.

#### Translation

The process of formation of proteins on the ribosomes is called translation.

(Ref. Guyton & Hall-1th Edition; page-33)

Trait	DNA	RNA
1. Bases	1. Adenine, guanine, cytosine and thymine.	1. Adenine, guanine, cytosine and uracil.
2. Sugar moiety	2. Deoxyribose.	2. D-ribose
3. Structure	3. Have 2 strands of antiparallel double helical molecule.	3. Single-stranded molecules.
4. Situation	4. Solely intranuclear and mitrochondrial.	4. Both extra & intranuclear (cytoplasm & nucleolus)
5. Functions	<ol> <li>Carry genetic information from one generation of cells to the next &amp; undergo mutation.</li> </ol>	<ol> <li>Carry no genetic information &amp; undergo no mutation.</li> </ol>
	<ul> <li>ii. Concerned with source of information protein synthesis.</li> </ul>	ii. Concerned with protein synthesis.