Introduction:

Think about a soap bubble, which consists of a thin, flexible membrane. This soapy membrane seals the inside air from the outside. Similarly, the cell membrane is a thin, flexible layer that seals the inside of the cell from its outside environment. The cell membrane encloses the contents of all cells, organelles and many cytoplasmic inclusions, and regulates what gets in and out. This is called selective permeability. Plasma membrane also acts as a dam for hydrophilic (water-soluble) molecules that cannot pass freely through cell membrane but it does not stop the diffusion of water. Remember about the diffusion that we have studied in previous chapter is the movement of particles from an area of higher concentration to an area of lower concentration. Regulation of water flow through plasma membrane as well as its selective permeability nature is necessary important factor in maintaining homoeostasis that is crucial for life processes. In this section, you will learn about the osmosis, osmotic pressure, osmotic equilibrium, Donnan equilibrium, and electro-osmosis.

Objectives: In this present module we will discuss and address the following objectives in detail:

1. What is Osmosis?
2. What Factors Controls Osmosis?
3. Define osmotic pressure and Osmotic equilibrium,
4. Principal of Donnan equilibrium,
5. What is electro-osmosis

Osmosis:
Greek word ‘osmos’ meaning ‘pushing’ i.e. Osmosis mean, push out a cell’s plasma membrane. It is spontaneous movement of solvent (e.g. water) through a semi permeable (i.e. selectively permeable) membrane across a solution gradient, from a region of lower solute concentration to a region of higher solute concentration in order to attain equilibrium is called osmosis. The net movement of solvent is called osmotic flow. This process does not require energy (ATP). There are many medical and health related problem are occurs due to disturbances in osmosis e.g. Diarrhea, cholera, edema and inflammation of tissue etc.

![Figure-1: During Osmosis, water diffuses across a selectively permeable membrane. Sugar molecules did not change on each side of the membrane but water molecules has changed in both side of the membrane.](image)

**What regulate Osmosis?**  If we add sugar in to water then water becomes sweeter as we add more sugar. When we placed stronger sugar solution with direct contact of weaker sugar solution or water then water molecules simultaneously starts diffusing in one direction and sugar molecules diffuse in other direction until equilibrium established. When we separated these two solutions by selectively permeable membrane water molecules diffusing from the region of higher water concentration to that of lower water concentration produce osmotic pressure. The water molecules continue to diffuse until it is equal concentration on both compartment of the membrane reached as shown in **Figure-1**.
Therefore we conclude that unequal distribution of solute particles, which generate a concentration gradient or osmotic pressure, is one factor that control osmosis.

**Osmotic Pressure:**

It is define as the extra pressure that is applied to a solution to prevent the passage of solvent in to the solution, when both are separated by a semipermeable membrane. As we know that semipermeable membranes do not let the solute pass through. A solvent will move to the side that is more concentrated to try to make each side more similar. Since there is a flow of solvents as shown in Figure-2, the height of each side changes, which generate pressure. Since the velocity is the same as that of a free molecule, the pressure will be the same as the pressure of an ideal gas of the same molecular concentration. Hence, the osmotic pressure $\pi$, is given by van’t Hoff formula, which is identical to the pressure formula of an ideal gas:

$$\pi = cRT$$

![Figure-2: Osmotic pressure](image)

Where,

$c$ is the molar concentration of the solute,
\[ R = 0.082 \text{ (liter-bar) / (deg-mol)}, \text{ is the gas constant, and} \]

\[ T \text{ is the temperature on the absolute temperature scale (Kelvin).} \]

Van’t Hoff received the first Nobel Prize in chemistry, in 1901, for his interpretation of osmosis. The osmotic pressure is directly proportional to the concentration of the solute molecules or ions. It is a colligative property, meaning that it does not depend on the solute type, or its molecular size, but only on its molar concentration.

**Units of osmotic pressure:** Osmole. One osmole is defining as the number of molecules in gram molecular weight of undissociated solute e.g. One gram molecular weight of glucose (180g) is called one osmole. However, one gram molecular weight of NaCl (58.5g) is equivalent to two osmoles, since NaCl ionizes to give two particles (Na\(^+\) and Cl\(^-\)). The osmotic pressure of biological fluids is frequently expressed as ‘milliosmoles’. The osmotic pressure of plasma is 280-300 milliosmoles/l.

**Osmotic Potential (Solute Potential)**

It is define as the potential of water molecules to move from a low concentrated (hypotonic) solution to a high concentrated (hypertonic) solution across a semi-permeable membrane. It is a relative value. If a membrane-bound, aqueous system with a solute particle concentration of 0.25 M is placed into an aqueous system with a particle concentration of 1.0 M, we say that the membrane-bound system has a high water potential because water would tend to flow, via osmosis, from the membrane-bound system into the system in which it was placed. Similarly, if we placed the same membrane-bound, aqueous system in an aqueous system with a particle concentration of 0.25 M, then we would say that the membrane-bound system has a low water potential because water would tend to flow into it from the surrounding environment. There would, of course, be zero water potential if both systems were isotonic. Osmotic potential of pure water \((\psi_\pi)\) is usually consider as of zero, and in this case, solute potential can never be positive. The association of solute concentration (in molarity) to solute potential is given by the van’t Hoff equation:

\[ (\psi_\pi) = -MiRT \]

Where;
\[ M = \text{concentration of the solute (molarity)} \]
\[ i = \text{van't Hoff factor} \]
\[ R = \text{i ideal gas constant, and} \]
\[ T = \text{absolute temperature (C+273)} \]

Osmotic potential has many important implications for living organisms. For example, when a solute particle is dissolved in pure water, water molecules are less likely to diffuse away via mechanism of osmosis towards low or no solute concentration. Thus solution has negative water potential than that of pure water. Furthermore, negative potential of solute will increase as the concentration of solute molecules increase. If a living cell is surrounded by a more concentrated solution, the cell will tend to lose water to the more negative water potential \( (\psi_w) \) of the surrounding environment. The effect of the force of osmotic potential on individual cells largely depends on whether or not the cell possesses a mechanism to withstand that force. It is crucially important to understand the how osmosis and osmotic potential affects cells.

This can be the case for marine organisms living in sea water and halophytic plants growing in saline environments. In the case of plant cells, which generally have low water potential because they are surrounded by hypotonic environments, absorb water until their cell wall prevents further osmosis. At this point they are said to be turgid. This is what happens when a stalk of celery “crisps up” when you place it in water, and is the preferred state of most plants. Most plants, however, have the ability to increase solute inside the cell to drive the flow of water into the cell and maintain turgor. This effect can be used to power an osmotic power plant. Animal cells don’t have cell walls and, indeed, if you place most animal’s cells in a markedly hypotonic environment, such as DW, water rushes into the cell and the cell explodes, this is called lysis. Erythrocytes that are placed in hypotonic environments and explode are said to have undergone hemolysis. Placing plant or animal cells in hypertonic environments increases the water potential of the cell, causing water to leave the cell. As water leaves, the cell shrivels; animal cells undergo crenation or “pruning,” whereas the membrane of plant cells shrinks back from the cell wall in a process called plasmolysis. Crenation and plasmolysis are generally fatal to the cell. Placing plant cells into an environment where the water potential is equal results in limp, flaccid plants; animal cells are normally in this kind of isotonic environment.
Cells in an isotonic (*isos* = “equal”) solution:
In an isotonic solution, the concentration of the dissolved solute particles in solution is the same as the concentration of dissolved substances inside the cell. The term isotonic is used when a cell is in direct contact with isosmotic solution (0.9% NaCl) that does not change the cell volume and thus cell tone and normal shape is maintained (Figure-3). Thus solutions that exert the same osmotic pressure are also called isoosmotic.

![Figure-3: A. Cell retained their normal shape in isotonic solution because water molecules move into and out of the cell at the same rate.](image)

Cells in hypotonic (*hypo* = “under”) solution:
In the hypotonic solution, the concentration of dissolved substances is lower in the solution outside the cell than the inside concentration of the cell. If a cell is placed into a solution

![Figure-4: Cell undergoes swelling in hypotonic solution due to osmosis.](image)
that has a higher water concentration than the cell, water tends to flow into the cell. In this case, we say that the solution in which the cell is placed is hypotonic and experience osmosis. Osmosis, unlike diffusion requires force. This force is due to interaction of solute particles which due to the Brownian motion interact with the cell membrane and get repelled, hence attaining a momentum and directed away from the cells, which in turn transfer the momentum to neighboring water molecule and drive them away from the membrane. The cells swells when they placed in hypotonic solution, which contains lower amount of solutes, making the water flow through the plasma membrane into the cell. The cell swells and its internal pressure increases. As the pressure increases inside the animal cells, the plasma membrane swells like the red blood cells as shown in Figure-4A. If the solution is extremely hypotonic, the plasma membrane may be unable to withstand this pressure and ultimately leading to cell burst.

In case of plant cells, which contain a rigid cell wall that supports the plasma membrane of the cell, they do not burst when in a hypotonic solution. As the pressure increases inside the cell, the plasma membrane is pressed against the cell wall, as shown in Figure-4B. So, instead of bursting the plant cell become firmer. That is the reason misting of water on fruits and vegetable help to keep them fresh.

**Cells in hypertonic (hyper = “over”) solution:**

In a hypertonic solution, the concentration of dissolved substances outside the cell is higher than the concentration inside the cell. Since, it has a higher solute concentration than the cell. Under hypertonic solution cells experience osmosis that cause water to flow out.

![Diagram](image)

Figure-5: Cell undergoes shrinkage in hypertonic solution due to osmosis.
Animal cells in a hypertonic solution i.e. one with higher osmolarity than the cytosol, the cell shrinks as shown in Figure-5A, because of decreased pressure in the cells. While plant cells in a hypertonic environment lose water, mainly from the vacuole. The plasma membrane and cytoplasm shrink away from the cell wall as shown in Figure-5B. Loss of water in a plant cell results in a drop in osmotic pressure and explains why plant wilt.

**Some naturally existing examples of osmosis:**

1. Vacuoles in the plant cell have a higher solute concentration, which is lower than the cell’s cytosol that in turn has higher solute concentration than the extracellular environment. So the water comes inside the cytosol and into the vacuole, but the cell doesn’t undergo osmotic burst as it has a rigid cell wall, which increases the pressure inside the cell, called turgor pressure but not the cell’s volume. This pressure pushes the cell wall.

2. In protozoan the cell doesn’t have cell wall, but the vacuole takes up all the water that comes inside the cell through osmosis and moves it out of the cell by integrating with the plasma membrane.

3. The frog’s oocyte and the eggs which have normal salt levels like the other cells protect themselves from osmotic rapture as they lack the water channel protein which are normally present in erythrocytes and the other cell type. These cells when injected with mRNA aquaporin protein, which is a protein of erythrocytes, shows swelling.
4. The cells of plant undergo cell division by expanding the cell’s vacuole by hormone induced mechanism for loosening of the cell wall.

5. In the case of diarrhea which leads to dehydration of the body due to excess of water getting out of the body as soon as we drink it. Getting antibiotics against the cholera or any other pathogen is not sufficient. The uptake of Na+ ions by the NA+/K+ ATPase and the glucose by glut2 receptors of the epithelial cells of intestine into the blood generating an osmotic gradient which leads to the flow of water into the blood, hence re-hydrating the body. So it is advisable to take sugar salt solution in case of diarrhea.

6. The opening and closing of the guard cells of the stomata is due K+ and Cl– ion influx causing the water to enter the cell, causing the cell to bulge outwards, causing it to open. The closing of Stomata is due to the revere process, by the efflux of ions and movement of the water outside. The opening and closing of the stomata is tightly regulated, when there is low level of CO₂ the water comes inside the guard cell, and if in the case of excess water evaporation the K+ ions moves outside the cell through a hormone regulation, which is formation of abscissic acid

**Reverse Osmosis (RO):** Prof. Reid and his colleagues gave the concept of reverse osmosis in 1950s. They demonstrated reverse osmosis desalination using cellulose acetate films. When solution is separated from its solvent by a semipermeable membrane, and hydrostatic pressure is applied increasingly on the solution side the permeation rate will decrease accordingly, and finally stop. The hydrostatic pressure at which permeation is
stop called the osmotic pressure, and permeation become in reverse direction at a greater hydrostatic pressure. This phenomenon is called reverse osmosis (RO). Reverse osmosis has a number of innate advantages like in food industries and desalination of seawater.

**Donnan Membrane Equilibrium:**

The Donnan membrane equilibrium is also known as the Donnan's effect, Donnan law or Gibbs–Donnan equilibrium named after the physicist Jossia Willard Gibbs. This is defined as the uneven charge distribution along one of the side of the membrane, as these charged can't diffuse through the membrane. Hence, creating a concentration gradient of the ions across the membrane that is then maintained by the diffusion of other permeable solutes, however, because of the solute 'p' which is not able to cross the membrane makes the side more concentrate due to its presence. So, the concentration of the impermeable solute remains high on one of the side even in the presence of equilibrium. This effect is known as Donnan membrane equilibrium.

![Donnan diagram](image)

**Figure-8: Donnan equilibrium:** a). Sodium and chloride ions separated by a membrane. b). Na⁺ P and Na⁺ Cl⁻ separated by a membrane: initial conditions. c). The final Donnan equilibrium conditions arising from (b).

The living cells contained impermeable anionic colloids, which can’t cross the membrane. These can be proteins or organic phosphates, creating donnan equilibrium in the interior of cell. To balance the environments, the water tries to move inside through the property of osmosis, but to protect the cells from rupturing, the ATP-dependent Na⁺ - K⁺ channels continuously pushes the Na⁺ ions out of the cell. Also due to the impermeability of the membrane for Na⁺ the cell prevent it to re-entering the cell, hence generating second donnan equilibrium. These two donnan effect balance each other and prevent the cells from osmotic burst. The salient feature of Donnan membrane equilibrium are:
1. The concentration of diffusible ions is influenced by the presence of non-diffusible ions across the membrane.

2. The concentration of oppositely charged ions (Na\(^+\)) is greater on the side of the membrane containing non-diffusible ions (P\(^-\)).

3. Concentration of the similarly charged ions (Cl\(^-\)) is higher on the side of the membrane not containing the non-diffusible ions (P\(^-\)).

4. The net concentration of total ions will be greater on the side of the membrane containing non-diffusible ions which is responsible to develop the osmotic pressure difference on either side of the membrane.

**Application of Donnan membrane Equilibrium:**

1. **Osmotic imbalance:** Donnan membrane equilibrium helps in to explain the osmotic pressure differences because it provides the detail information about the differential distribution of ions in different compartments of the body.

2. **Difference in the ionic concentrations of biological fluids:** Generally low concentration of inorganic cations (Na\(^+\), K\(^+\)) and higher concentration of anions (Cl\(^-\)) are present in lymph and interstitial fluids compared to plasma. This is attributed to the higher protein content in the plasma.

3. **Membrane hydrolysis:** Presence of non-diffusible ions influenced the acidic or alkaline nature on either side of a membrane due to relative strength of H\(^+\) and OH\(^-\) ions. This phenomenon is called membrane hydrolysis. Donnan membrane equilibrium explains the greater concentration of (H\(^+\)) ions in the gastric juice.

4. **Lower pH in RBC:** The pH of RBC is slightly lower (7.25) than that of plasma (7.4) due to negative charge of the haemoglobin of RBC is and, therefore that causes the accumulation of positively charged ions including H\(^+\).

**Electro-osmosis:**
The motion of the fluid in the capillary, when the electric field is applied is called electro-osmosis. This motion is directly proportional to the electric field and depends upon the material of capillary and surface of solution in contact with the wall of capillary. The motion
is due to electrical force on ions in the electrical double layer, a thin layer of ions that is located near a wall exposed to an aqueous solution.

The flow of the solution if uniform across the capillary then fluid velocity would be uniform far from the wall, but it would decay to zero at the wall over a length scale, ranging from approximately 0.5-200 nm. The 1st layer is surface charge depending upon the material of capillary used; generally borosilicate is used which provide negative charge to the membrane. This layer is also called Stern layer or Helmholtz layer. The second layer is diffuse layer and is made of the ions from the solution. It is the moving layer and depends upon the electric field applied across membrane. When electric potential is applied across the membrane the, the diffuse layer moves from one side of the membrane and drag the solution with itself. Electro-osmosis is defined in term of mobility and velocity-

\[ \text{Mobility } \mu \text{ is given by: } \mu_{\text{EOF}} = \frac{\varepsilon \zeta}{\eta} \]

Where, \( \zeta \) zeta, is the potential of the double layer on the capillary wall, this potential depends upon the ionic strength, as the pH varies, the zeta potential also varies. At high pH the silanol groups get deprotonated, increasing the net charge on capillary wall, hence the zeta potential increases. So the electro osmotic flow increases with increase in the pH this varies when pH varies from 2-12.

\[ \varepsilon = \text{dielectric constant,} \]
\[ \eta = \text{the viscosity of the solution. Units are kg/s m} \]

\[ \text{Velocity } = \text{It is the mobility multiplied by electric field, the ability to move in electric field. } \]

\[ V_{\text{EOF}} = F \mu_{\text{EOF}} \]
The flow induced by the pump shows the parabolic motion, as the flow is applied to the entire cross section, so the velocity of solution in the center is more than the velocity of solution in contact with the capillary wall due to the frictional force. But in the case of electro-osmotic flow A. the bulk flow is due to the primary action of cation near the surface, so the force is distributed throughout the surface. The flat flow is falls directly at the surface of capillary, due to the frictional force it encounters, but to a lower extent then the pump induced flow. The electro-osmotic flow results in compact peak then the broad peak, which is seen in the parabolic laminar flow, because of the analyte which is spread in large area.

**Applications of Osmosis:**

1. **Blood volume and Fluid balance:** Osmosis play a crucial role in regulation of blood volume as well as urine excretion. Osmosis principal is also significantly contributed to maintained fluid balance in different compartment of the body.

2. **RBCs and Fragility:** In isotonic solution (0.9% NaCl) RBCs volume remain unchanged and their shape is intact. While in hypertonic solution, cytoplasm of RBCs is shrinks due to outflow of water, this phenomenon is called ‘crenation’. While in hypotonic solution, RBCs undergoes ‘hemolysis’ due to entry of water that often caused due to rapture of plasma membrane. Increased fragility of RBC is observed in ‘hemolytic jaundice’ while it is decreased in certain anemia. Generally, RBCs of normal human blood begin to hemolysis in 0.45% NaCl and the hemolysis is almost complete in 0.33% NaCl. Osmotic fragility test is very crucial for RBCs is employed in laboratory for diagnostic purpose.

3. **Transfusion:** Normal saline (0.9% NaCl) or glucose (5%) or any other suitable combination like DNS are isotonic solution. In hospital, these isotonic solution are very commonly used for transfusion during treatment of dehydration, burn etc.

4. **Action of purgatives:** The Epson (MgSO₄.7H₂O) or Glauber’s (Na₂SO₄.10 H₂O) salts withdraw water from the body via osmosis, instead preventing the intestinal water absorption. Thus, the mechanism of purgative action is also based on osmotic phenomenon.

5. **Osmotic Diuresis:** High glucose concentration in blood is responsible for osmotic
diuresis resulting in the loss of water, electrolytes as well as glucose in the urine. This is the basis of polyuria observed in diabetes mellitus. Diuresis can be produced by administering compounds which are filtered but not reabsorbed by renal tubules e.g. mannitol.

Summary:

Osmosis is spontaneous movement of solvent (e.g. water) through a semi permeable (i.e. selectively permeable) membrane across a solution gradient, from a region of lower solute concentration to a region of higher solute concentration in order to attain equilibrium. There are many medical and health related problem are occurs due to disturbances in osmosis e.g. Diarrhea, cholera, edema and inflammation of tissue etc. Osmotic pressure or concentration gradient which is generated due to unequal distribution of solute particles is the key factor that control osmosis. Osmotic pressure $\pi$, is given by van’t Hoff formula; $\pi = cRT$. Osmole is the unit of osmotic pressure. Osmotic potential has many important implications for living organisms. The solutions that exert the same osmotic pressure are said to be isoosmotic. The term isotonic is used when a cell is in direct contact with an isosmotic solution. (0.9%NaCl) which does not change the cell volume and thus the cell tone is maintained. A solution with relatively greater osmotic pressure is referred to as hypertonic. On the other hand, a solution with relatively lower pressure is hypotonic. According to Donnan law, concentration of diffusible ions is influenced by the presence of non-diffusible ions across the membrane. It helps in to explain the osmotic pressure differences because it provides the detail information about the differential distribution of ions in different compartments of the body. electro-osmosis is define as the motion of the fluid in the capillary, when the electric field is applied. Briefly, osmosis is important phenomenon and play crucial role in maintain blood volume, RBCs, shapes, purgatives, during transfusion as well as in diuresis.