

Introduction

When two aqueous solutions of different concentrations are connected by a membrane that is permeable to water molecules but not to solute molecules, osmosis, or water diffusion, occurs (Fig. 1). Diffusion is the net movement of molecules from a region of higher concentration to a region of lower concentration of these molecules. We say that the molecules move along the concentration gradient. As the diffusion continues, the concentration gradient decreases until the concentrations become equal and diffusion stops. Diffusion is a spontaneous and irreversible process. Spontaneous, because it occurs without any participation of external forces. Irreversible, because the net flux of particles is always along the concentration gradient and the solution will never spontaneously separate into two solutions with different concentrations. When diffusion involves a solvent, such as water, we speak of osmosis. Remember that the concentration of a solution means the concentration of a substance dissolved in water. In a highly concentrated solution, the concentration of water is low, and in a low concentration solution, there is a high concentration of water. In Fig. 1, the concentration of water is greater in the left cylinder and lower in the right cylinder, so we will observe the flow of water into the right cylinder. As a result of osmosis, the volume of the solution on the left side of the membrane will decrease and the volume of the solution on the right side will increase. The concentrations of solutions also change, the left solution becomes more concentrated and the right solution becomes less concentrated.

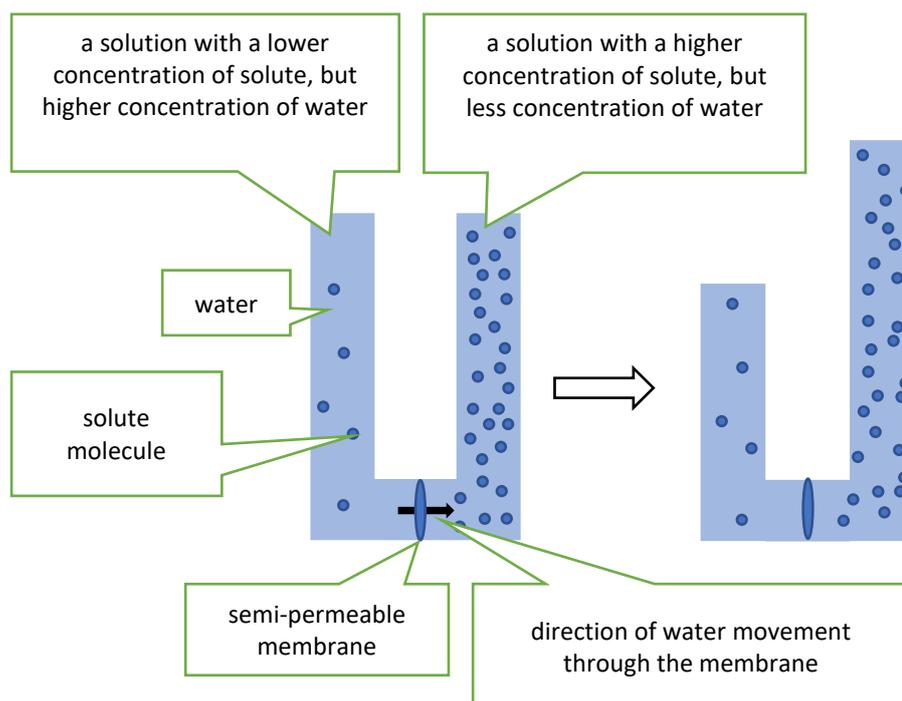


Fig. 1

The cause of diffusion (and osmosis) is the chaotic movement of all molecules in a solution. Since there are more water molecules on the left side of the membrane than on the right (Fig. 1), statistically more water molecules will move from left to right than from right to left. To understand this, imagine that there are 1000 water molecules on the left side of the membrane and 100 water molecules on the right side (Fig. 2). Each molecule can make a move to the right or to the left with equal probability (as, for example, flipping a coin gives the same probability that heads or tails will come up). So it is most likely that of the 1,000 molecules on the left, half, i.e. 500, will move to the right and pass through

the membrane. Of the 100 molecules on the right side, half, i.e. 50, will move to the left and pass through the membrane. On the left, the number of particles will decrease by 450. On the right, the number of particles will increase by 450. It turns out that we have a net flow of 450 molecules from left to right, and the number of molecules on both sides has become equal. We would say that the concentration of water on both sides of the membrane is equal.

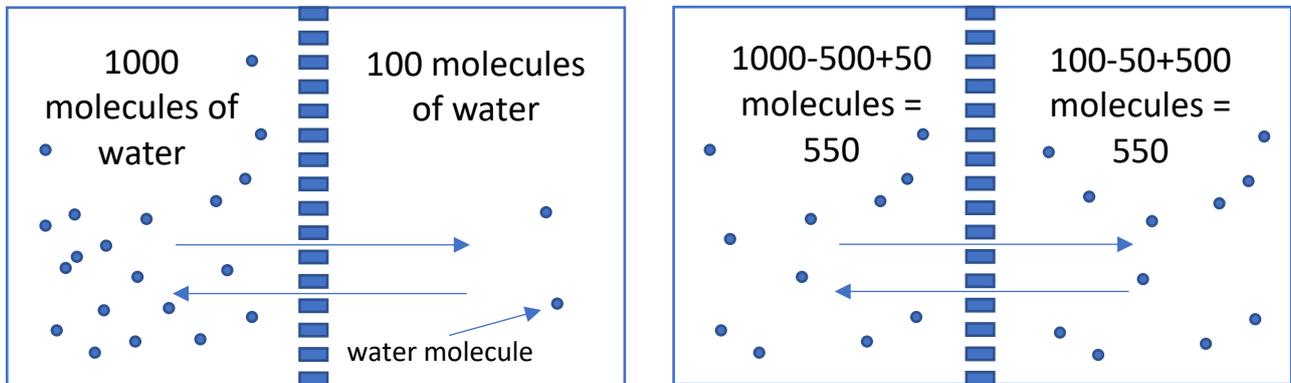


Fig. 2

In real solutions, there are a huge number of molecules, and each molecule can move in any direction.

To mathematically describe the phenomenon of osmosis, we will introduce the concept of osmotic pressure. The osmotic pressure is equal to the pressure that would have to be applied to a solution to stop water from flowing through the membrane. If our aqueous solutions in Fig.1 were in cylinders equipped with movable pistons, then by pressing the piston on the right side we could stop the flow of water. The pressure exerted by the piston on the surface of the solution in this cylinder would be equal to the osmotic pressure created by the difference in water concentrations on both sides of the membrane. Osmotic pressure can be described by the van't Hoff equation :

$$\pi = (C_1 - C_2)RT$$

π – osmotic pressure [Pa = N/m²]

C_1, C_2 – molar concentrations of solute on both sides of the membrane [mol/l]

R – gas constant ($= 8,31 \frac{J}{mol \cdot K}$)

T – absolute temperature of the solution [K]

When there is pure water on one side, the van't Hoff equation will be :

$$\pi = CRT$$

C – molar concentration of solute on another side [mol/l].

The van't Hoff equation has a serious limitation - it can only be used for very dilute solutions, so osmotic pressures are usually not calculated. In practice, a measuring device called an osmometer is used. Osmometers measure osmolality, i.e. the concentration of substances dissolved in water, expressed as the number of moles of osmotic active substances dissolved in 1 liter of water. The unit of osmolality is Osm.

Osmosis plays an important role in the physiology of living organisms. Water is essential for the proper functioning of cells. The cell membrane is permeable to water but impermeable to most

components of the cytoplasm. Thus, water can flow freely into the cell, causing it to swell, or flow out, which causes the cell to shrink. The extracellular solution, in relation to the intracellular solution, may be:

- isotonic - has the same osmotic pressure
- hypotonic - has a higher osmotic pressure
- hypertonic - has a lower osmotic pressure .

A cell placed in a hypotonic solution will swell, and in a hypertonic solution it will shrink (Fig. 3).

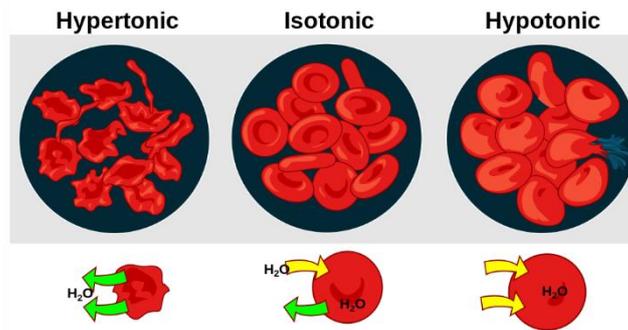


Fig. 3 from Wikipedia

Performing the experiment

The experiment consists in measuring the air pressure in two cylinders separated by a semi-permeable membrane. One cylinder will hold pure water, the other a concentrated sucrose solution. Changes in pressure will be caused by a change in the volume of liquid in both cylinders due to osmosis.

1. Connect the Dual Pressure Sensor to 550 Universal Interface:



2. Run the computer and open "Osmosis" file. You will see the window:



3. Change the sampling interval of data to 2 minutes.
4. Prepare 40 ml of 2 M sucrose solution.

The component of a solution that is present in the largest amount is known as the solvent. Any chemical species mixed in the solvent is called a solute, and solutes can be gases, liquids, or solids. The molarity or molar concentration of a solute (C) is defined as the number of moles of solute (n) per unit of volume (V) of solution (not solvent!):

$$C = \frac{n}{V}$$

The unit of molarity is M (pronounced "molar"). 1 M = 1 mol/1 litre. The mole is defined as exactly $6.02214076 \times 10^{23}$ elementary entities. Depending on the nature of the substance, an elementary entity may be an atom, a molecule, an ion, an ion pair, or a subatomic particle such as a proton. The weight of 1 mole of substance is called the molecular weight (μ). The number of moles (n) in a mass (m) can be calculated from the equation:

$$n = \frac{m}{\mu}$$

To prepare 40 ml of 2 M (2 mol/litre) sucrose solution we have to weigh the right amount of sucrose. The molecular weight of sucrose μ is equal 342 g/mol. We can calculate the mass of sucrose (m) from the equation:

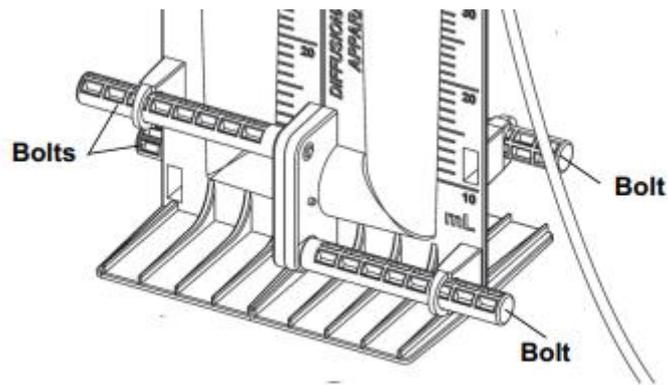
$$C = \frac{m}{\mu V}$$

Remember to convert the unit of volume (V) from "millilitre" to "litre" before the calculation. Use electronic scale to weigh the sucrose and prepare the solution in the beaker. The sucrose must be completely dissolved in water.

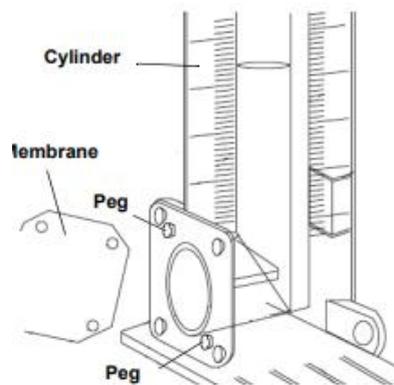
5. Prepare the Diffusion-Osmosis Apparatus and perform data recording .

**Do not touch the semi-permeable membrane with your hands.
Use tweezers!**

- Unscrew the four blue bolts and separate the two Diffusion-Osmosis Apparatus cylinders.



- The semi-permeable membrane is kept in distilled water in petrie dish. Take it out carefully with tweezers and line up two of the four corner holes on the membrane with the two raised pegs on one of the cylinders.



- Carefully place the other cylinder against the membrane so that the two raised pegs on the second cylinder go into the two membrane holes. Check that:
 - the membrane is securely in place between the two O-rings and pulled tightly across the space, with no folds or gaps;
 - all four raised pegs are inserted into the membrane holes;
 - the two alignment tabs near the top of the cylinders are aligned.
- Screw in the four bolts carefully. Ensure that they are tight but do not use much force. The two cylinders should be tightly secured to each other, with the membrane separating the space between the two cylinders.
- The two cylinders of the apparatus are labelled #1 and #2. Fill cylinder #1 with 40 ml of distilled water. Fill cylinder #2 with 40 ml of sucrose solution. Tilt the apparatus to remove air bubbles from the membrane.
- Place a blue cap onto the first cylinder, press it down, and turn the cap to seal the cylinder. Seal the second cylinder with the other cap.
- Attach pressure sensor #1 to cylinder #1 and attach pressure sensor #2 to cylinder #2.

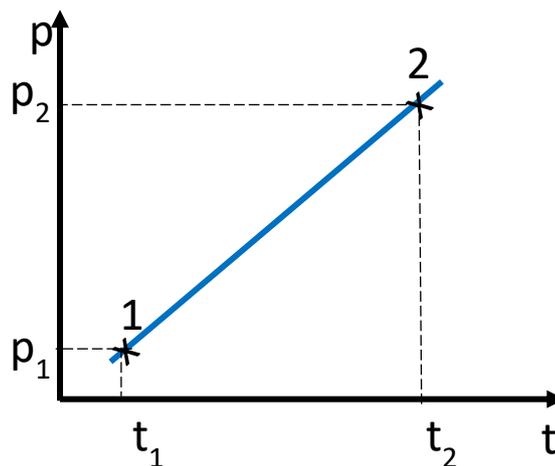
- Start recording data (click the green circle at the bottom of the SPARKvue window). Allow the processes of osmosis to occur within the apparatus undisturbed for 1 hour. Read and record pressure of water and sucrose every 2 minutes in Table 1.
- After 1 hour stop recording data (click the red square at the bottom of the SPARKvue window). Remove the caps, empty the cylinders and carefully dismantle the apparatus, rinse the membrane in distilled water and put it back in the petri dish. Rinse all the cylinders in distilled water.

6) Analyze the data.

- Calculate the Differential Pressure (Δp) and record in Table 1.
- Draw three graphs representing the rate of pressure change:
 - $p_{\text{water}}=f(t)$,
 - $p_{\text{sucrose}}=f(t)$,
 - $\Delta p=f(t)$.
- Fit the straight line $y=ax+b$ to the experimental points in the graphs. Calculate the slopes for each of them.
 - Manually:

Mark two points on the straight line as close as possible to the beginning and end of the straight line. From the x and y axes read the coordinates of the selected points ($t_1, p_1 ; t_2, p_2$). Calculate the slope a from the formula:

$$a = \frac{p_2 - p_1}{t_2 - t_1}$$



- Using an Excel spreadsheet:

In the sheet, in the first column, write the time (t), in the second – pressure (p) . Select cells with numbers. In the Menu, choose to Insert a chart. Select the scatter plot without lines and close the chart selection window with the Finish button. Right-click any measurement point on the chart and select Add trend line. Choose Linear type, and in Options, select Display equation on the chart. Close the

window with the OK button (in OpenOffice the trendline is called a regression curve). The function of the form $y = ax + b$ appears on the graph. Read the slope of the line.

Questions for discussion

1. What is the final change in the pressure in both cylinders?
2. Is the change in pressure equivalent on each side of the apparatus? In other words, is the loss of pressure on one side the same as the gain of pressure on the other side? Why or why not?
3. Compare the slopes for distilled water and sucrose solution. Use your observations to explain the relationship between solution concentration and the rate of osmosis.
4. Identify at least two parts of the Diffusion-Osmosis Apparatus that model osmosis in multicellular organisms, and explain whether the apparatus more closely models plant cells or animal cells.