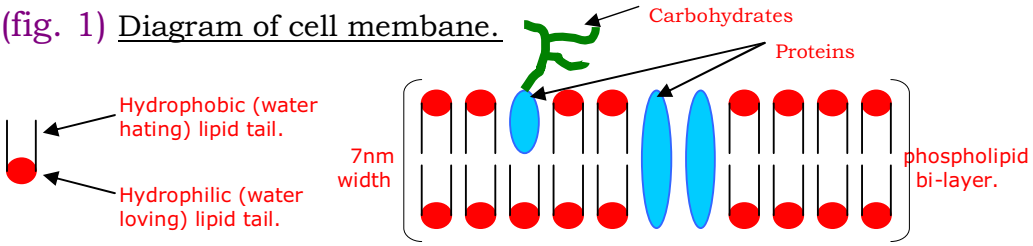
Investigation to find the water potential of a plant cell.

Introduction: -

Theory:

Plant cells are made up of a phospholipid bilayer (fig. 1). There are hydrophilic heads, which face the liquid medium in and around the cells. A hydrophobic tail attached to the head, which face inwards. These tails are non-polar. Non-polar molecules like oxygen can diffuse through the membrane easily. Water, despite being a polar molecule, can also diffuse rapidly across the membrane because it is so small. Big polar molecules and ions can not pass straight through the membrane, e.g. Na+, Cl- and glucose. These molecules can only pass the membrane through protein channels, which only allow specific molecules in.

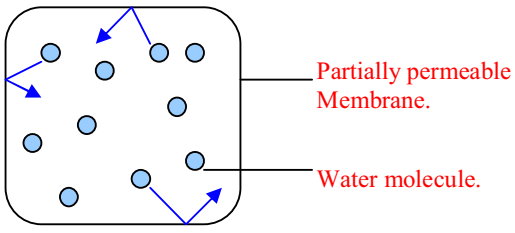
(fig. 1) Diagram of cell membane.



Water molecules posses kinetic energy in that, when together in liquid or gas form, they move about very rapidly in random directions. When water is surrounded by a membrane the water molecules will tend to hit it as they move randomly, and in doing so, they will generate pressure (fig.2). This pressure is known as water potential. The more times the water molecules hit the membrane in a unit of time then the higher the pressure, i.e. the higher the water potential.

(fig. 2)

Diagram showing Water potential.



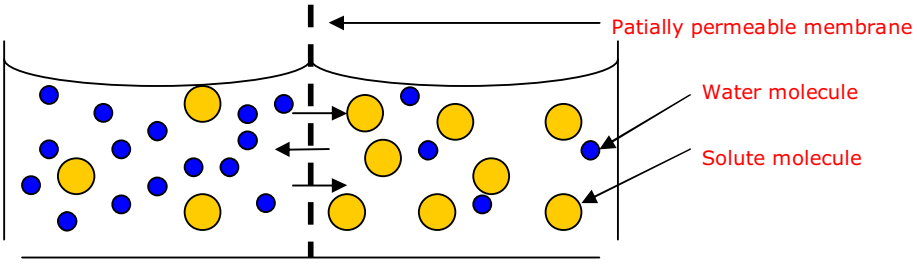
Water molecules generate pressure as they collide with a partially permeable membrane.

One definition for **water potential** could be 'the tendency for water molecules to move from one place to another- with or without a net movement'. Water always moves from an area of high water

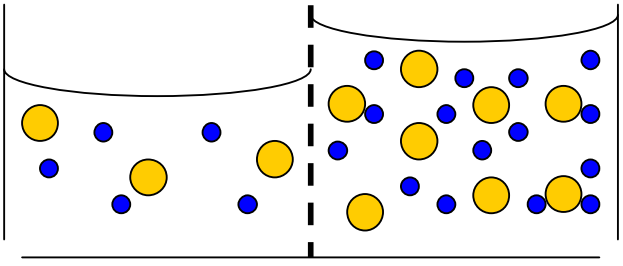
potential to an area of low water potential, meaning it always moves down its water potential gradient. There will be no net movement of water molecules when equilibrium is reached, meaning that the water potential in one area is the same as in the other area. The symbol for water potential is ψ, the Greek letter psi, and this is measured in units of pressure, kilopascals (Kpa).

The highest water potential is distilled (pure) water, measured at 0. If any solute e.g. sucrose, is placed in the distilled water, it will lower the 'purity' of the water and, therefore, lower the water potential. This means that the water potential will be less than zero, thus a solution with any solutes in it, will have a negative water potential. **Solute potential**, ψ**s** is the amount that a solute lowers the water potential, meaning that solute potential is always negative. In (fig. 3a), it can be seen that two solutions are separated by a partially permeable membrane. In solution A, there are lots of water molecules but not so many solute molecules. In solution B, there are not so many water molecules but there are lots of solute molecules. Because the solute molecules are too big to pass the partially permeable membrane, the water molecules move via osmosis. Because solution B has a higher solute to water ratio than solution A, solution B has a lower water potential. And because water moves down its water potential gradient, from a region of high water potential to a region of lower water potential, the net movement of the water molecules will be from A to B. In (fig.3b), the solutions have reached an equilibrium where there will be no net movement across the partially permeable membrane

(fig. 3a) Showing the movement of water molecules in a solution across a partially permeable membrane.



(fig. 3b) The equilibrium of the two solutions.



Pressure potential (ψp) is another factor that affects water potential. Plant cells have a very strong cell wall made up of cellulose. This means that if the cell were placed in distilled water, it would not burst (as would an animal cell placed in distilled water, because animal cells only have the double phospholipid or semi-permeable membrane with no cell wall).

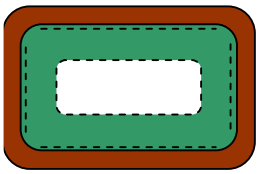
So if a plant cell is dopped in to distilled water, water will begin to enter the cell because plant cells generally take up solutes from the soil through their roots, meaning the cell will have a lower water potential than the distilled water, and water will go down its concentration gradient. The plant cell will grow turgid exerting a pressure inside the cell (fig.4a). This is pressure potential and forces water back out of the cell. Because water potential is described as to move water molecules form one place to another, pressure potential must increase the water potential.

In a plant cell, the water potential can be expessed as the following equation: $\psi = \psi s + \psi p$

If the plant cell was placed in a concentrated sugar solution then water would be taken out of the cell, going down its concentration gradient. The cell will become flacid and the protoplast will pull away fom the cell wall causing the cell to be plasmolysed. The cell will not be damaged too much because the rigid cell wall will keep the cells shape (fig.4b).

(fig. 4a) A turgid plant cell.

Turgid cell showing partially permeable membranes as dotted lines.



Cell wall- freely permeable

Plasma membrane - partially permeable

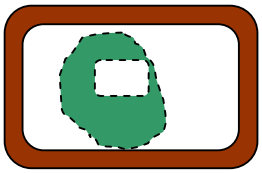
Cytoplasm

Tonoplast- patially permeable

Vacuole

(fig. 4b) A flacid plant cell.

Fully plasmolysed cell



Cell wall

External solution has passed throughthe cell wall and is still in contact with the protoplast.

Protoplast has shrunk away from the cell wall - the cell is fully polymolysed.

Vacuole

Cytoplasm

Osmosis is the process that water passes through the membranes of the cells. It can be described as 'The movement of water molecules from a region of high water potential to a region of low water potential through a partially permeable membrane.' The two diagrams, (fig. 3a & b) show how osmosis works.

Aim: -

The aim of this experiment is to find the water potential of a plant cell. By using the theory above, it can be possible to find the exact water potential by plotting a graph.

Prediction: -

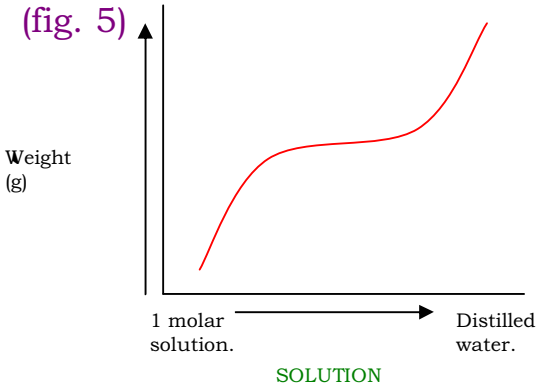
As we have seen from the theory above, plant cells will allow water molecules to pass through the membane of the cell without trouble, but big molecules such as glucose, can not diffuse through the membrane. Water will go down its water potential gradient from a region of high water potential to a region of low water potential. Distilled water has a very high water potential at 0Kpa. Any solution with solutes in it make the water potential less e.g. a 1 molar solution of glucose has an osmotic pessure of –3500Kpa.

From all of this, we can make an accurate prediction to what we think will happen to the potato cells. When the potato cylinder is placed in the strongest 1 mol sugar solution, the potato cylinder will decrease in size and mass because the solution will have a lower water potential than the potato cylinder. This means that the water will move down its water potential gradient, in this case, out of the cell, into the solution. The cell will become plasmolysed until equilibrium is reached.

When the potato cylinder is placed in the pure, distilled water, the potato cylinder will increase in mass and size because the solution has a higher water potential than the potato cylinder. The fluid will enter the cell through osmosis going down its water potential gradient, until pressure potential inside the cell forces water back into the solution and equilibrium is reached.

Between these two solutions will be the exact water potential of the potato cylinder. When it has reached equilibrium there will be no net movement of water and no change in size or mass in the potato cylinder. By drawing a graph, the exact water potential can be found. Fig. 5 shows what this graph may look like. We will change the solutions around by adding concentrations of either different amounts of distilled water to a 1 molar solution or different concentrations of solvent to distilled water. This means that it will be easier to plot the graph and find the exact water potential.

(fig. 5)



Predicted graph.

This is an 'S' shaped line o sigmoid curve.

A table to show the variables that must be controlled.

Variable.	Why it must be controlled.	How it will be controlled.
Temperature.	At higher temps., the water molecules move faster than at lower temps., i.e. diffusion will take place quicker	I shall do the experiment at room temp., so if there is a change in temp., all the specimens will experience the same change.
Surface area.	The more surface area there is the more area for diffusion to take place and the faster diffusion can occur.	I shall cut the potato with a cork borer and cut the cyliders to the same length (nearest mm) so they all have the same suface area.
Concentration gadient.	The bigger the difference in concentrations, the more molecules pass through the membrane, which means a bigger net rate of diffusion.	The concentration of the water and glucose solutions will be the variable I will change in the experiment. ★
Cells water potential.	Two different potatoes may contain cells with slightly different water potentials, which could give anomolous results.	I shall take all my potato samples from the same potato.
Measurements.	Wrong measurments of the different molar solutions could mean the osmotic potential will be wrong and my results will not be accurate.	I will measure the solutions to the greatest degree of accuracy using precise equipment.
Time.	Enough time must be given to let osmosis happen and all the specimens must be given the exact same time or some will be able to take up or release more water than others which will change my results.	I can time the potato cylinders by using a stop clock.

★ This will be my variable in the experiment and I will change the concentrations of the solutions by using the information on page 6.

A table to show how the concentrations of the solutions can be made.

End concentration. (Mol dm)	% of distilled water.	% of 1M glucose solution.	Osmotic potential. (Kpa)
1.00	0	100	-3500
0.75	25	75	-2370
0.50	50	50	-1450
0.25	75	25	-680
0.00	100	0	0

The concentations which are above are the ones which I will use in the test. I feel these will give me enough results to plot a graph and find the water potential of the potato cylinders.

I may have to make up the one molar solution in order to obtain the rest of my solutions. This can be done by taking the formula for glucose which is C□H□□O□. and from this I can work out the molecular mass and then from this, I can work out the amount of glucose in grams needed in a litre of distilled water which will make up a 1 molar solution:

C□H□□O□: = (12 x 6) + (1 x 12) + (6 x 16) =180

This means that 180g of glucose is needed to be added to a litre of distilled water to make a 1 molar solution.

Method: -

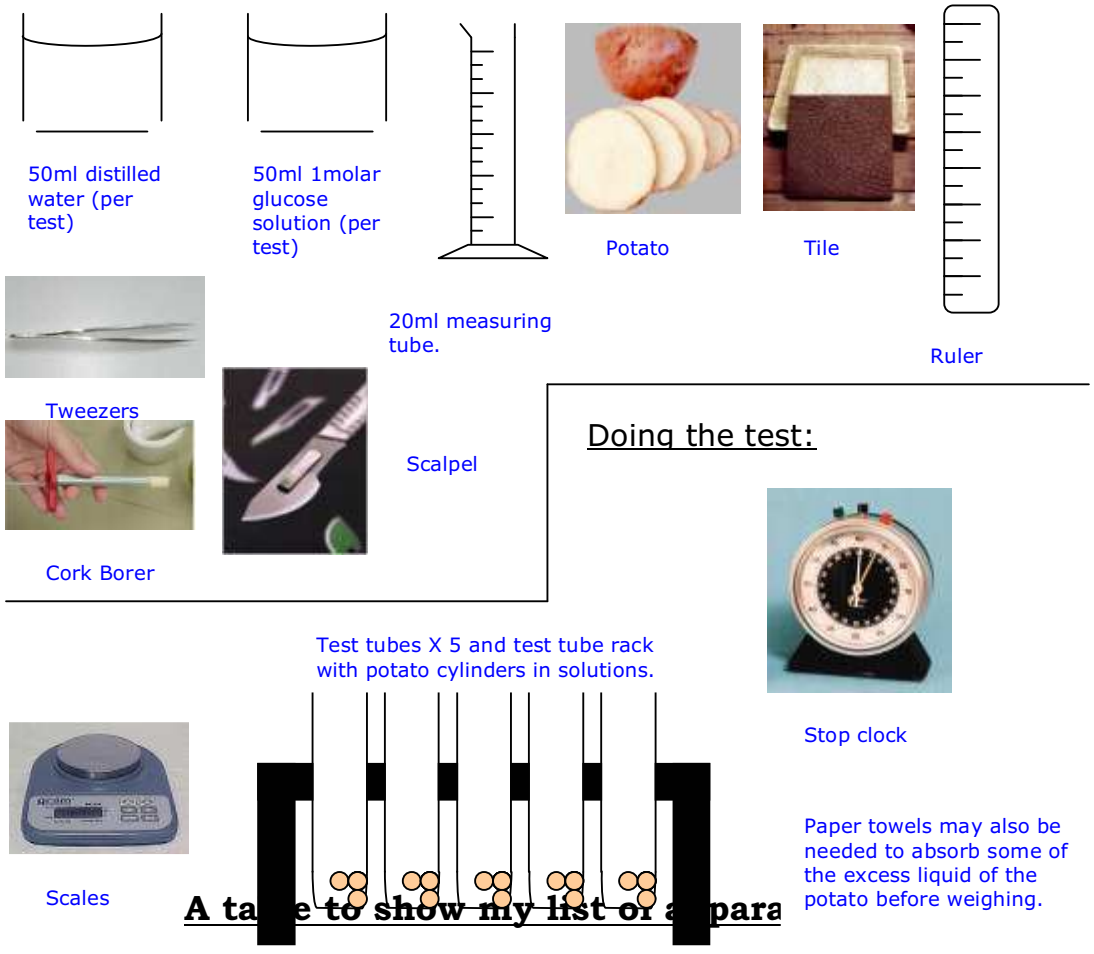
- Make up a 1 mol solution of glucose and then using distilled water, make up 0.25, 0.5 and 0.75 molar solutions of glucose.
- For each solution, measure out 20ml into three different test tubes and also 20ml of 1 mol solution and distilled water into a further two test tubes.
- Cut out 15 cylinders of potato using a cork borer, trim to 5mm in length.
- Record the weight of the potato cyliders to 2 d.p, in groups of three (making sure that these groups of three are put in the same solution) before putting them in the solution.
- Place the groups of three potato cyliders in the 5 solutions leaving 3 mins between each so that it

gives me time to measure the weight for each set of potato cyliders when taken out of the solution, as shown in (fig. 6)

- Leave the potato cyliders in the solution fo 30 mins, timed with a stop watch.
- Take the potato cyliders from the solutions and then remove the excess liquid with a paper towel.
- Weigh the three potato cyliders from each solution to 2 d.p and record the figures against their starting weights.
- Calculate the difference in weight of the potato cyliders and plot these figures on a graph.
- Then read off the water potential of the potato from the graph where there is no weight change.
- Repeat the process three times in order to get an average, and to reduce the effect of an anomaly on the whole test.

Diagram of my apparatus: -

(fig. 6) Preparation apparatus:



A table to show my list of apparatus

Item	Quantity	Concentration and volume.
Potato	1 large one	---------
Cork borer	1	---------
Electric scales	1 (accurate to 2 d.p)	---------
Test tubes	5 (per test)	---------
Scalpel	1	---------
Ruler	1	---------
Clay tile	1	---------
tweezers	1 pair	---------
Test tube rack	1	---------
measuring cylinder	1	20ml
Paper towels	However many needed	---------
Stop watch	1	---------
Distilled water	---------	50ml (per test)
1 mol glucose solution	---------	50ml (per test)
Graph paper	1	---------

A table to show reasons for choice of apparatus.

Item	What is it used for?	Reason for choice.
Potato	This is where the plant cells will come from.	Potato lets water in and out easily and this is easy to measure.
Cork borer	Cutting the potato into identical cyliders.	All the cylinders will have the same suface area.
Electric scales	To weigh the potato before and after test.	Gives precise reading to 2 d.p.
Test tubes	To hold the different solutions and potato cylindes.	Because the solution may not cover the potato in a shallow dish.
Scalpel	To cut the potato to 5mm lengths.	Very precise cutting tool, damaging cell walls to minimum.
Ruler	To measure the length of potato cylinder.	Gives an accurate length of potato cylinder.
Clay tile	To cut the potato on.	Hard surface to cut on

tweezers	To hold the potato cylinder in place for cutting.	Hold the potato firmly so a bad cut will not happen, and minimize damaging to cell walls.
Test tube rack	To hold the test tubes.	Holds test tubes upright, and contents are visible.
measuring cylinder	To measue out my solutions	This is an accurate way of measuring.
Paper towels	To remove excess solution off the potato cylinder before weighing.	They soak up liquid well, and do not damage the piece of potato, nor leave any residue thereon.
Stop watch	To time how long the potato cylinder have been in the solutions.	Very accurate way of measuring time.
Distilled water	To make up solutions.	----------
1 mol glucose solution	To make up solutions.	----------
Graph paper	To plot the results of the experiment.	An easy and accurate way visual of doing it.

Preliminary tests: -

In my preliminary test, I used the same sized potato cylinder, all weighing 0.5g. I put these potato cylinders into 5 solutions, 1 mol of glucose solution, 0.25, 0.50 and 0.75 mol of glucose solution and one of distilled water. Here are the results:

Weight g 1.0M solution	Weight g 0.75M solution	Weight g 0.5M solution	Weight g 0.25M solution	Weight g distilled solution
0.4	0.4	0.5	0.5	0.5
0.4	0.7	0.4	0.5	0.7
0.3	0.5	0.5	0.5	0.6

These results seem fairly reasonable although I was expecting to weights to change more considerably. This may be because I only left these samples in for ten minutes rather the thirty that I will do in the experiment.

I found using the cork borer a lot easier to use than I thought it would be, but cutting the cylinders with the scalpel to exactly the right length was hard. The concentrations that I have used are good and easy to make up. However, I feel they do not give me a wide enough range for the experiment, so I will use the following, 0, 0.2, 0.4, 0.6, 0.8 and 1 molar solutions. I think at least three tests will have to be done to get an average, if not more, so that anomaly can be recognised like the one in this preliminary investigation (highlighted).

A table to show how the new concentrations of the solutions can be made

End concentration. (Mol dm)	% of distilled water.	% of 1M glucose solution.
0.00 molar solution	100 (20ml)	0 (0ml)
0.20 molar solution	80 (16ml)	20 (4ml)
0.04 molar solution	60 (12ml)	40 (8ml)
0.06 molar solution	40 (8ml)	60 (12ml)
0.08 molar solution	20 (4ml)	80 (16ml)
1.00 molar solution	0 (0ml)	100 (20ml)

Safety precautions: -

- Care most be taken with glasswear to prevent any breakages.
- Spillages of the solutions must be mopped up quickly.
- Care must be taken when using the sharp cork borer by using it on a hard surface like a tile.
- Care must be taken when using the sharp scalpel, and also using it on a hard surface like a tile.
- Attention must be given to the experiment at all times so that optimum results are obtained safely.

References: -

- Cambridge advanced sciences – Biology 1
- Biology – Martin Rowland.
- Advanced Biology – Principals & Applications.
- Biology dictionary – Alan Clamp.

I only used these sources as they are reliable. 'The Cambridge advanced sciences – Biology 1' was published by our exam board so must have the correct things for this examination. 'Biology – Martin Rowland' and 'Advanced Biology – Principals & Applications', are both from two different biologists but share the same content as in 'The Cambridge advanced sciences – Biology' 1' but in more detail. This shows that they all link to each other in their content despite being written by different people so the information must be accurate to modern knowledge in the subject. The 'Biology dictionary – Alan Clamp' has many definitions which also appear to

be the same in the other books so this source must be as reliable as the rest.