16th March

Biology Coursework

Aim

To investigate and find the water potential of baking potatoes and sweet potatoes in (N/mm^2) using various strength solutions of sucrose (mol)

Hypothesis

I believe that the baking potatoes will have a less negative water potential. In the experiment they will not increase in mass at a lower of sucrose molarity than the sweet potatoes.

Baking potatoes contain more water than sweet potatoes. This is clearly obvious, cut a baking potato into cubes and it releases a lot of water, more so than sweet potatoes.

Water potential determines the rate of osmosis. Water movement form a high concentration to a low one is the means for testing the aim. Distilled water and other strength solutions will flow into the sweet potato more readily than into the baking potato. This is because baking potatoes have more water, and thus a less negative water potential. Sweet potatoes have less water content and will therefore have a more negative water potential.

Both potatoes will have a lower water potential then the distilled water and other sugar solutions. As water diffuses into their cells they will increase in mass. The weight difference will be converted into a percentage so that the water potential can be determined.

When plotted on a graph, percentage change of mass against solution molarity, the line of best fit will cross the x-axis. At this point the potatoes will not have increase in mass, their mechanical pressure will offset the solute potential. At this point there would be no diffusion gradient and osmosis would cease. The potatoes would have the same water potential as their surroundings.

Using tables I can determine what the water potential is in kPa or N/mm^2 by means of comparison using the graph obtained by this experiment. (See appendices i)

Obviously the sweet potato and the baking potato would have different water potentials. Their lines of best fit would cross at different points. Baking potatoes would be closer to the origin, as its water potential is less negative and will correspond to a less negative water potential in the sucrose solution.

Background Information

Osmosis is a special case of diffusion, specifically related to water molecules acting as a solvent with a solute, in this experiment the solute would be sucrose.

Osmosis is the diffusion of water molecules from a region of high concentration to a region of low concentration through a partially permeable membrane, in our case a lipid bi-layer.

To explain osmosis, we will take the common experiment of concentrated sucrose solution in dialysis tubing, suspended in distilled water.

Free water molecules have diffused into the sucrose solution form a region of high concentration (outside the bag) to a low one (inside the bag). Large slow moving hydrated sucrose is mostly retained within the dialysis tubing. Sucrose does diffuse outwards, but at a slower rate than water.

Water is bi-polar since oxygen is high electronegative.
Sucrose is somewhat bi-polar, with –OH groups having two separate areas of positive and negative charge. These naturally attract the opposite counterpart in the water molecule. They do not form a permanent bond but cause the water to "follow" the sucrose in a hydrated shell.

Different liquids containing water have a measure called water potential (Ψ psi) to show the tendency for water molecules to enter or leave that solution by osmosis.

Pure water has the highest water potential, set at zero kPa. If you were to dissolve a solute into distilled water, you would lower its water potential. As a result solutions at atmospheric pressure have negative values set in kPa. Water diffuses from a more positive water potential to a more negative one, for example 0 to –10kPa, -5 to –70kPa, or 2 to-5kPa.

Water potential is determined by the presence of dissolved solutes (solute potential) and the actual pressure acting on the water (pressure potential) such as when a cell is turgid. Water potential is calculated using the formula:

Water Potential = Solute Potential – Pressure Potential

$$\Psi = \Psi_s - \Psi_p$$

Solute potential is negative, since the solute dissolved lowers the potential below zero, as already explained.

Pressure potential however is positive. If a pressure is applied to distilled water, its pressure potential increases. Hydrostatic pressure (pressure potential) usually is positive but can be negative such as in xylem where the water is under tension so the pressure potential returns a negative value. In plant cells the force of the water pushing against the cell wall creates a positive pressure potential causing the cell to become turgid.

If we return to the sucrose experiment and let it continue, the bag will eventually become turgid.

If the bag is suspended in fresh distilled water, it will increase in mass as it gains water. Because the bag acts like a cell wall, it cannot stretch further and pressure begins to build up eventually rising out to cancel out the solute potential. Osmosis "ceases" since both inside and outside the bag have the same water potential, but in actual fact water molecules are entering and exiting the bag at the same rate.

Every cell in a plant has a lipid bi-layer surrounding its content. It's hydrophobic/hydrophilic nature makes it theoretically impermeable to water molecules. So how does water get into a cell to affect its water potential?

It is generally accepted that the cell membrane is a "fluid mosaic", that is the lipid molecules are free to orient as they like. Water can sometimes find gaps in-between various molecules embedded in the surface such as glycoproteins.

Also cells have protein lined pores that selectively allow some compounds through. These water channels occur in huge quantities all over the cell membrane's surface.

When a cell is in water of the same water potential as itself, there is no net water movement, even though water molecules are moving in and out of the cell. The two sets of water are said to be isotonic.

If there is no net water movement into cells, then they will not increase in mass. If we were to replace the surrounding cells with a solution of sucrose, you can test which solution will not cause the cells to increase in mass. If you know the water potential of the solution then you can find the water potential of the cell.

When the external water potential of the cell is less negative than the cell, more dilute, water flows into the cell. In this case the outside solution is said to be hypotonic. The cell sap becomes diluted, and the volume begins to expand and push against the cell wall. A pressure potential develops and eventually causes the rate of osmosis to cease.

The cell walls help prevent the cell from haemolysing and destroying the cell. The cell is now described as turgid. Being turgid does not cause damage to the cell and it will eventually return back to its normal size. This case is most likely to happen during the experiment when the solvent used has a molarity of 0.0 or 0.2 mol of sucrose (pure water or very dilute sucrose). In these cases the potato slices would have put on their maximum weight.

The reverse can also happen. If the surrounding solution is more negative (called hypertonic) then the water flows out of the cell. The cell sap would become more concentrated and the cell membrane would pull away from the walls, apart form where cytoplasmic connections occurred.

Cells in this condition have become flaccid. Cells that are flaccid are not damaged in the long run and return to normal once they receive water. This case will probably occur at molarities of 1.0 or 0.8 mols of sucrose, since they will have a more negative water potential than the potato cells. The slices in this case would lose weight.

The baking potato and the sweet potato have different compositions and these factors will determine the outcome of the experiment because the amount of impurities in the potato will govern its water potential.

Potatoes have 76g of water for every 100 grams, whereas sweet potatoes only have 70g of water every 100g. This already shows that baking potatoes have more water and therefore a less negative water potential than the drier sweet potatoes.

Preliminary Experiment

The aim of this preliminary experiment was to best determine how to carry out the method in the real experiment, not to gather results. I only used one type of potato, the baking potato, to test the method since the sweet potatoes would not show another way of carrying out the practical.

My method was to rinse all the equipment out with distilled water three times. Then I cut a cylinder out of the baking potato using a borer. Then I cut thirty-six slices of roughly the same width using a scalpel on a white tile, boring out more cylinders when needed. I then labelled six test tubes with 0.0, 0.2, 0.4, 0.6, 0.8, and 1.0. I then put 10 ml of distilled water into the 0.0 test tube using a burette, 8 ml into the 0.2 tube, 6 ml into the 0.4 tube, 4 ml into the 0.6 tube and 2 ml of distilled water into the 0.8 test tube. Once this is complete, you need to re-rinse the burette with distilled water. Then, put 10 ml of 1.0 mol sucrose into the 1.0 test tube, 8 ml into the 0.8 tube, 6 ml into the 0.6 tube, 4 ml into the 0.4 tube and 2 ml of 1.0 mol sucrose into the 0.2 tube. Shake each test tube well, covering it and then inverting it three times. You should now have sucrose concentrations equal to that of their labels. Record the combined mass of six potato slices and place them into the 1.0 mol test tube. After three minutes, drain away the solution and dry the potato slices gently on some tissue, then reweigh. Take another six slices and do the same for each of the different strength solutions of sucrose, taking six fresh slices every time.

The equipment used were: digital scales, baking potato, borer, sucrose 1.0 mol, scalpel, white tile, test tubes, test tube rack, pipette and distilled water.

Results:

Water (ml)	Sucrose (ml)	Molarity	Initial Weight (g)	Final Weight (g)	Percent change in mass (%)
0	10	1.0	2.12	1.97	-7.1
2	8	0.8	1.98	1.88	-5.1
4	6	0.6	2.17	2.16	-0.5
6	4	0.4	2.20	2.29	+4.1
8	2	0.2	2.22	2.35	+5.9
10	0	0.0	1.71	2.16	+26.3

I feel that the results generally went well, except the last value which was a bit more extreme than I thought it would be. While I was carrying out the practical I became aware that when the potato slices were placed into the test tubes they did not always show the same surface area, something which might affect results since the amount of contact between cells and water determines how much osmosis takes place, more surface area, more area for water to diffuse into cells. In a bid to correct this, I will use a mounting needle, so that as much surface area is in contact with the water. Also I might leave the potato slices in their test tubes for five minutes so that as much water as possible can enter the cell. Also I will repeat the experiment in total twice more to improve reliability (three times baking potato, three times sweet potato). In addition, I could also take the mean temperature of the room, because temperature differences have an affect on proteins and their hydrogen bonds (protein water channels), or I could submerge the test tube containing solutions into a water bath at a

reasonable temperature like 30°C. This would be a better temperature, as it would be more stable than fluctuations in the room temperature. Also, they protein water channels would benefit more and function more efficiently at a temperature greater than 25°C.

Method

Place a thermometer in the room (away from sunlight) and record its temperature every five minutes. The thermometer is there to get a mean temperature reading throughout the practical to allow for variances which could affect the protein pores in the potato cells. The thermometer must be out of sunlight because that could give a biased reading. Rinse out all components (not electronic like the stop clock or the thermometer) three times to remove impurities that could affect the accuracy of the solutions. Rinsing the equipment three times dilutes impurities by a greater factor than just doing it once.

Using a borer cut out a whole cylinder of potato out of the baking potato. Using a white tile as a backboard, cut 108 slices of potato, taking more cylinders out of the potato then needed, of roughly the same width using a scalpel. Take care with the scalpel as it is much sharper than a normal knife. The white tile will protect the surface you are working on. The 108 slices will be sufficient for the entire experiment. 36 slices will be required for the different solutions, six to each test tube. The remainder will allow for two extra repeats. The potato slices must be the same width and diameter to have the approximately same surface area, this is a primary factor over differences in mass as they are generated into percentages later on. The surface area will affect how much water can enter the total volume of the potato slice.

Label six test tubes with the values given below.

Mix together the quantities of distilled water and 1.0 mol sucrose solution as directed below.

Label	Add Distilled Water Volume (ml)	Add 1.0 Mol Sucrose Volume (ml)
0.0	10	0
0.2	8	2
0.4	6	4
0.6	4	6
0.8	2	8
1.0	0	10

You must cover then shake each test tube to mix up the sucrose and water, this will give an even water potential. The values of the mixtures give the molarity of the solution as said on the label. These molarities give a wide range of possible readings, sufficient enough to give a proper reading.

Weigh six potato slices on a digital set of scales, taking care to press the rest button before and after using it. Then carefully impale them on a mounting needle, making sure you do not stab yourself or cause the potato slices to break up. Space the slices out so there is a gap in between each of them on the needle. You need six slices to give a large enough mass change for the scales to recognise. Weighing scales in the lab will not show a small mass difference if you use one slice. You need to weigh the potatoes on digital scales because they are very accurate. You need to reset them because there may be residue left behind on the scales that could cause future

inaccurate readings. The slices need to be spaced out so that maximum surface area is shown. You cannot use broken slices because they will have more surface area than a normal slice.

Submerge the mounting needle (needle down) into the 0.0 mol solution. Then submerge the test tube into a water bath at 30°C. After 5 minutes remove it, roll it gently on some tissue to dry it, then remove the slices onto the reset weighing scale. Record the new weight on a suitable table (see results for an example). Dispose of the used potato slices. Using some tissue, dry the scales and then press the reset button. The water bath will succeed in keeping the temperature stable and accurate and will allow the protein channels to let water in quicker than at room temperature, this would show a more noticeable change on the weighing scales than simply a small 0.1% increase/decrease. Five minutes gives sufficient time for the water to enter the cell. Drying the potato slices removes excess water that was not absorbed. Pressing the reset button on the scales prevents inaccuracy, as does drying the weighing plate it and then pressing the reset button.

You must repeat the last two paragraphs until you have done all the possible molarities once, using fresh or rinsed with distilled water equipment three times equipment. Using fresh equipment will prevent inaccuracy

Once you have done each molarity once, repeat them each again two more times, making up new solutions, so they provide more reliable results, since doing a practical inaccurate three times in highly unlikely. By this time all 108 slices would have been used up, provided you did everything without breaking any slices.

After using all 108 slices of baking potato, take a fresh or rinsed borer and cut out whole cylinders of sweet potato. Taking a fresh or rinsed scalpel and a fresh or rinsed white tile as a backboard cut out 108 slices of sweet potato.

Repeat the experiment just as you did with the baking potato, making various solutions as outlined in the table and impaling six slices to every mounting needle.

Controlled Variables

The independent variable is the molarity of the sucrose solution
The dependent variable is the mass change in grams
The controlled variables are the surface area of the potato slices (to a certain extent)
the number of slices

Apparatus

Pipette
Digital scales
Sweet potato
Baking potato
Borer
Sucrose 1.0 mol
Distilled water
Scalpel
White tile
Mounting needle
Stop Clock
Water Bath

Safety and Ethical Issues

The biggest issue of concern would be the scalpel. It is sharper than a knife and carries the added risk of a carrying an infection, something which distilled water would not stop. The advice would be to refrain from moving it too far away from the site of the practical, and when using it to operate in safe and calm manner. You could even go as far to cover the blade with a double layer of tissue, not the handle, to prevent damage even by a bit.

The white tile would protect the desktop as the tile is impervious to the scalpel. The workbench would be damaged otherwise without the tile.

The mounting needle could also cause concern. You could easily stab yourself or someone else. The same advice for the scalpel would apply to the mounting needle, not to move it around and to wrap it up in tissue when not used.

You should also be careful not to burn your hands in the water bath as its water can scold at the wrong temperature.

There are no ethical implications with this test since it deals with plants.

Results

Mean Temp 32.0°C

Baking Potato

Molarity (mol)	Initial weight (g)				Final weight (g)				% Change in Mass			
	A	B	C	Average	A	B	C	Average	A	B	C	Average
0.0	0.84	0.75	0.80	0.80	0.90	0.80	0.86	0.85	7.1	6.7	7.5	7.1
0.2	0.76	0.83	0.86	0.82	0.81	0.87	0.91	0.86	6.6	4.8	5.8	5.7
0.4	0.87	0.85	0.76	0.83	0.88	0.89	0.78	0.85	1.1	4.7	2.6	2.8
0.6	0.73	0.90	0.76	0.80	0.72	0.92	0.76	0.80	-1.4	2.2	0.0	0.3
0.8	0.79	0.77	0.73	0.76	0.75	0.77	0.72	0.75	-5.1	0.0	-1.4	-2.1
1.0	0.83	0.73	0.91	0.82	0.77	0.70	0.88	0.78	-7.2	-4.1	-3.3	-4.9

Sweet Potato

Molarity (mol)	Initial weight (g)				Final weight (g)				% Change in Mass			
	A	B	C	Average	A	B	C	Average	A	B	C	Average
0.0	0.82	0.81	0.78	0.80	0.86	0.91	0.86	0.88	4.9	12.3	10.3	9.2
0.2	0.73	0.75	0.77	0.75	0.77	0.83	0.85	0.82	5.5	10.7	10.4	8.8
0.4	0.75	0.74	0.69	0.73	0.78	0.77	0.75	0.77	4.0	4.1	8.7	5.6
0.6	0.73	0.74	0.65	0.71	0.75	0.77	0.69	0.74	2.7	4.1	6.2	4.3
0.8	0.70	0.71	0.73	0.71	0.72	0.74	0.73	0.73	2.9	4.2	0.0	2.4
1.0	0.76	0.74	0.75	0.75	0.75	0.72	0.73	0.73	-1.3	-2.7	-2.7	-2.2

Conclusion

From my graph, I can clearly see that my hypothesis is correct. The baking potatoes came to the same water potential as the sucrose at a lower concentration than the baking potatoes (0.62 for baking potatoes against 0.92mol for sweet potatoes). If I were to extrapolate these results on the osmotic potential graph, I would get values of -2N/mm^2 for baking potatoes and –3N/mm^2 for sweet potatoes, again proving my hypothesis by showing that baking potatoes have a less negative water potential than sweet potatoes.

I ended up with this result because as mentioned in the background section, baking potatoes have a higher water content (76g out of every100g) than sweet potatoes. As a result it has a less negative water potential because its contents are more dilute. Also it has less contents than the sweet potato, 21.0g of carbohydrates against 22.0g in sweet potatoes, in other words its solute potential would be less negative (assuming that the pressure potential is the same at the start of the experiment for both types of potato). This means that water would not have entered the baking potato as fast as it did in the sweet potato because baking potatoes have a shallower concentration gradient than sweat potatoes. This explains the different positions of the two lines of best fits on the graph, sweet potatoes put on more weight because more water was entering its cells at a faster rate for the same amount of time as baking potatoes.

The results start up at a high percentage change in mass at 0.0 mol because the potatoes are in a pure water solution. By the laws of osmosis, water would diffuse into them. This is also true of the 0.2 mol value, it is very dilute and would probably have a minor negative number compared to other solutions, it is very dilute. The 0.4 value has more of a negative number but still not on the scale of the potatoes so water diffuses into them and the slices would increase in mass. At 0.6 mol the water potential is extremely close to that of the baking potato so osmosis slows to a snails pace and the slices do not increase in mass, their percentage mass does not change. At higher concentrations of sucrose, the baking potato has a less negative water potential so water flows out of the cell and the percentage mass would go down. Meanwhile at 0.6 mol, the water potential is still not as negative as that of the sweet potato. The osmosis rules still say that the water should go into the cells. As a result the mass goes up. At approximately 0.9 mol the water potential of the sweet potato and the surrounding sucrose solution are the same. Osmosis ceases and there is no net gain of weight. At higher concentrations the water potential of the sucrose solution is more extreme than that of the potato and water would flow out of the cells, causing a percentage decrease in mass.

The first half of the experiment, testing to find the percentage mass change in baking potatoes went incredibly well. The results I obtained do not significantly deviate from the line of best fit indicating that the experiment was relatively accurate.

However in the second half of the experiment when finding the percentage mass change for sweet potato, the results deviate more from the line of best fit more. The 1.0 mol result is even anomalous, it does not quite follow the line and seems to be lower than it really should be.

The temperature was not that precise, it was supposed to be 30°C but was instead 32.0°C. I do not think this had a major impact on the experiment since it was kept constant

Evaluation

The anomaly in the experiment at 1.0 mol during the sweet potato test could have upset the result of the conclusion. The graph depends on the line of best fit and this is easily influenced by extreme values. The true result of the 1.0 mol test could move the line of best fit to the right or to the left, thereby changing the reading molarity reading and the subsequent reading off the osmotic potential graph. This could also be said of the other results of the sweet potato test, as they do not follow an accurate line like the baking potato test. This could also change the position of the line of best fit.

Anomalous results could be caused by laziness or fatigue, or because the equipment was not washed well enough with distilled water before use. Also, if you do not keep wiping the scales and resetting it then the weights might be recorded wrongly. When making solutions it is important to check volumes in the pipette by looking at the level of the meniscus on a level plane and not at an angle, this could give differing volumes and inaccurate concentrations.

The limitations of the experiment were that the slices were not kept for sufficiently long periods of time in their solutions, and this prevented the maximum effect of osmosis from being carried out and thus a more representative percentage mass change. In subsequent investigations, the time may be extended to seven and a half minutes to allow for extra diffusion of water. More accurate readings for mass change would give a more accurate value for osmotic potential.

You could also find the optimum temperature to submerge the slices in, because the efficiency of the protein based water channels is dependent on their hydrogen bonds which are easily affected by temperature changes. A preliminary experiment could find this out by using a sucrose solution of 0.4 mol and trying different temperatures to see which one put on the most weight, which would therefore be the optimum temperature as the protein channels are working their best. Finding the best temperature would give more true values for mass change as the cells would be working at their best. This in turn would give a better osmotic potential value that was more accuracy.

Appendices (i)

Molarity (mol)	Osmotic Potential (N/mm2)
0.05	-0.13
0.10	-0.26
0.15	-0.41
0.20	-0.54
0.25	-0.68
0.30	-0.86
0.35	-0.97
0.40	-1.12
0.45	-1.28
0.50	-1.45
0.55	-1.62
0.60	-1.80
0.65	-1.98
0.70	-2.18
0.75	-2.37
0.80	-2.58
0.85	-2.79
0.90	-3.00
0.95	-3.25
1.00	-3.50

Bibliography

- M B V Roberts Biology A Functional Approach
  Students Manual

- Cleegs & Maklean Advanced Biology Principles and Applications
  Second Addition

- Mary Jones & Geoff Jones AS Biology Module 1 : Molecules & Cells
  Student Support Materials for Edexcel

- A E Bender & D A Bender Food Tables