<u>Investigating the water potential of plant material</u>

Aim: To discover the water potential of plant material (potato and apple).

Introduction:

Water potential/osmosis is the ability of water molecules to move from an area of high concentration to that of a lower concentration. Using this theory one can find the water potential of plant material by comparing it to solutions of a known water potential, i.e. sucrose solution. By discovering if the sucrose solutions have either higher or lower water potential than the plant material.

This can be found by putting a sample of plant material into a specified concentration of sucrose solution, and then seeing if the net movement of water molecules is into or out of the sample. If the net movement were to be into the sample (this can be calculated by a change in the mass of the sample), the sucrose solution would have a higher water potential (greater water concentration) than the sample. This is because osmosis states that the net movement of water molecules is from an area of high water potential to that of a low water potential.

Once a set of results of results has been obtained they can be plotted onto a graph. From this one will see at what concentration of sucrose solution there would be no net movement of water molecules i.e. where the line of best fit crosses the x-axis. Then by using the conversion graph calculate the water potential of potato.

Why use sucrose? Sucrose is a naturally occurring disaccharide in plants; therefore it is unlikely to affect the chemical properties of the potato's cells. Sucrose is also a safe material; therefore little precautionary measures need to be taken. One only needs to wear a lab coat to protect ones clothing, goggles are not needed. Sucrose is widely available and hence has been widely tested. The water potential of various concentrations have been found, this means that by finding the line of best fit, the point at which the line intersects the x-axis, the plant material will have an equal water potential to that concentration of sucrose.

Firstly a set of concentrations of the sucrose solution needs to be chosen. Preliminary tests were done to see at what concentration there was a net movement of water out of the potato. The tests showed that when using a sucrose solution with a concentration of 0.5$moldm^{-3}$ there was movement of water out of the potato. However the potato was only left in the solution for around 25 minutes, this was obviously not long enough, however from these preliminary tests one can hypothesise that the water potential of potato will be less than that of 0.5moldm^{-3}. This is because if there was movement of water molecules out of the potato after a minimal time span like 25 minutes, given more time one would expect a greater net movement. Therefore a set of concentrations between 0-0.6moldm^{-3} was chosen.

Also from these preliminary tests it was decided that the plant material should be left for a number of hours in order to get measurable change in mass. However it should not be left for days as this may see the growth of mould or other micro organisms, this would probably affect the results as they would decompose the plant material and make the results unreliable. Therefore it was decided they should be left for 6 hours, as this was the longest they could be left, unless they were left over night. In leaving them for 6 hours this should produce a measurable change in mass.

Prediction:

One would expect potato to have a higher water potential than apple. This is due to what their function is in the natural world. A potato is a tuba and its primary job is to produce runners so the plant can expand and produce more plants. It stores starch, which is insoluble in water as it is a huge molecule. Hence resulting in starch having no affect on the water potential of potato. Other molecules such as salts and soluble minerals affect the water potential of water.

Apple however is a fruit and is designed to be eaten and is therefore sweet and appetising. This is because the organism that eats the apple will then drop the apple's seeds far and wide. The sweetness is from sugar-sucrose which is a disaccharide made from the joining of two monosaccharides-fructose and glucose, which is soluble in water because it is a relatively small molecule and a polar molecule. This means that one end is δ^+ and the other δ^-, which are the attracted to the opposite charge of the polar water molecule. The sugar therefore affects the water potential of apple and one would expect it to be lower than that of potato, as one would expect there to be a greater number of solutes in apple than potato.

Preliminary tests showed that potato contained little or no sugar, but was full of starch. In contrast apple contained no trace of starch, but was full of sugar. To test for starch potassium-iodide solution was added to the sample of plant material. When testing for sugars add benedicts solution to the aqueous/suspended sample and heat gently. The sample will start to turn from green through to brick red depending on the amount of reducing sugar present. Sucrose is not a reducing sugar and needs to be broken down into its two monosaccharides-fructose and glucose, which are reducing sugars. This is achieved by hydrolysing the glycosidic bond between the two monosaccharides, use hydrochloric acid, but then add an alkali such as sodium hydrogen carbonate to neutralise the sample, then add benedicts solution and heat as before and one will see the sample change to green through to a reddish colour as apple is rich in sugars.

Equipment List:

Volumetric flask	Boiling tubes	Conical flasks
Pipettes	Boiling tube racks	Tile
Pipette fillers	Cork borer (4mm diameter)	Electronic balance

Pipette fillers		Knife

Method:

1) Make up the sucrose solutions; one needs to dilute the primary sucrose solution of 1.0 moldm^{-3}. Use the table below to make up 0.20dm^{-3} of the varying concentrations of sucrose:

Concentration moldm^{-3}	Volume of 1.0 moldm^{-3} sucrose	Volume of distilled water dm^{-3}
0.00	0.00	0.20
0.10	0.02	0.18
0.20	0.04	0.16
0.30	0.06	0.14
0.40	0.08	0.12
0.50	0.10	0.10
0.60	0.12	0.08

Use a pipette or syringe to obtain the required amount of 1.0 moldm^{-3}, put into a volumetric flask of 0.20dm^{-3}. Fill up the flask with de-ionised water until the mark is reached, the bottom of the meniscus should be inline with the mark.

2) Use a pipette up take up 0.03dm^{-3} of the sucrose solution and put into a boiling tube, this is so that the sample of potato will be completely covered. It is now ready for the potato to be put in.

3) Plunge 4mm cork borer into the plant material. Then cut the material into 20mm lengths. Weigh each piece to get a 'mass before' result.

4) Put one piece of plant material into each boiling tube of sucrose solution, leave for 6 hours.

5) After 6 hours take samples out of each boiling tube. Remove all the liquid on the outside of the plant material. This needs to be removed so one is only weighing the liquid inside the potato sample.

6) Weigh the samples and work out how much it has lost or gained in mass. Then calculate the percentage change of each sample of potato, the average percentage change for each concentration, and find the standard deviation of the results. Finally plot the results onto a graph and calculate the line of regression, to find where the line crosses the x-axis and thus what concentration of sucrose has an equal water potential to the plant material.

Justification of Method and Choice of Equipment:

Once the solutions have been made up they need to be kept refrigerated and covered to prevent evaporation. Evaporation will cause the volume of water to reduce and thus increase the concentration of the sucrose. Thus making the test void, as the concentration of the sucrose solution will not be known.

The equipment, which has been made reference to above, was chosen for its accuracy in comparison to a measuring cylinder for example. This is because measuring cylinders are quite wide, therefore the meniscus is wide and there is a greater chance for inaccuracy. Whilst volumetric flasks and pipettes are very thin at where the mark is, thus reducing the chance of inaccuracy. If one were to make up each $0.03dm^{-3}$ separately there is also another chance of error and inaccuracy that is why it is better to make up a batch of each solution. Each test will be repeated five times to check for accuracy of results.

The samples of plant material should all be of the same surface area and around the same mass, in order to make the test fair. This is because if there were to be a difference in surface area, there would not be a constant area of potato directly exposed to the sucrose solution. Thus resulting in a different amount of water molecules being able to either exit or enter the potato (membrane) at one time. A similar mass is required because if there were major differences in mass there maybe a difference in the amount of water in that sample of potato and they may have very different water potentials to the other samples of potato. To get the same surface area a cork borer will be used and the potato sample will be cut to 20mm in length.

Using standard deviation will show the range from the average result, therefore a smaller standard deviation the more reliable the result. Purely finding the average is not enough to show reliability, as anomalous results can drastically change the average and thus make it unreliable. The line of regression is calculated because it a very accurate line of best fit. It will produce a line far more accurate than purely using ones eye to estimate, thus making the point at which the x-axis is intersected accurate.

Safety:

In general this is a fairly low risk investigation. Sucrose is purely a sugar solution, therefore it is not an irritant or explosive. Hence there being no reason to wear safety goggles. A lab coat would be advisable to protect ones clothing from spillages. A lot of equipment is made from glass; therefore care must be taken to prevent these from falling and smashing. If such an accident were to happen simply use a dustpan and brush to clean up (not hands). Keep stools under tables when not in use and clear up any spillages quickly particularly those on the floor.

Analysis

From observing the graphs one can see that potato has a water potential of –640KPa. Whilst apple has a water potential of either –840KPa or –1040KPa. Therefore the prediction that apple would have a lower water potential than potato was correct. The reason for this is that an apple is sweet and hence has sugar inside its flesh. Sugar can dissolve in water and hence reduces the water potential of the liquid inside the flesh of the apple. The sweetness is to attract animals and birds to eat it, they will then spread the seeds of the apple so new apple trees can grow. Whilst a potato contains starch, which is not soluble in water, it therefore will not affect the water potential of the flesh of the potato.

A negative percentage change in mass, showed that the water potential of the sucrose solution outside of the plant material less than that inside the plant material. Hence resulting in the water molecules net movement being out of the sample, therefore a loss in mass would be logical. A positive percentage change in mass showed that the water potential of the sucrose outside the plant material was greater than that inside the plant material. Therefore the net movement of the water molecules was into the sample, a gain in mass would also be logical. This has previously been proved, the laws of osmosis state 'the net movement of water molecules from an area of high water potential to an area of low water potential'.

The graphs show the basic concept of as the concentration of sucrose gets higher the smaller the positive change in percentage mass and the greater the negative change in percentage mass. This due to the water potential of the sucrose solution getting lower, and hence resulting in either a decreasing net movement of water molecules into the plant material, or an increasing net movement of water molecules out of the plant material. The latter is because the water potential of the sucrose solution is getting increasingly smaller than that of the plant material. Whilst the former takes place because the water potential of the sucrose solution is getting ever closer to that of the plant material.

Evaluation

The results obtained from the potato investigation all seem fairly consistent. The averages that were plotted onto the graph all seem to be fairly close to the line of best fit. However there was one result, which should be classed as anomalous, that was the first result for the 0.1moldm^{-3}. It had a change in mass of 22.47%, which is a huge gain in mass in comparison to the other results obtained. The result seems very similar to those obtained from the 0.0 moldm^{-3}. That sample of potato could therefore have been left in water rather than the 0.1moldm^{-3} sucrose solution. This would have been a fairly easy mistake to make, as both solutions were clear, transparent solutions, and would have been put into the boiling tubes at similar times.

In contrast to the potato investigation the apple investigation had a number of anomalous results. The second and fifth results obtained from the 0.2moldm^{-3} sucrose solution showed a loss in mass whilst the other result showed a gain in mass. To prove that the two referred to results were anomalous and not the other three, the set of results obtained from the 0.4moldm^{-3} sucrose solution showed less of a loss of mass than the two identified anomalous results.

The 0.3$moldm^{-3}$ sucrose solution results are all very varied numbers. They are all anomalous results and hence should be ignored[1]. The first and possibly the second result from the 0.4$moldm^{-3}$ seem to be anomalous also.

There are a lot of anomalous results in the apple investigation. The reasons for this are endless. However from considering how the investigation was carried out the most credible explanation is due to the fact these tests were done on two separate days, as there was not enough time to set up all of them at the same time. Unfortunately due to various reasons such as teacher absence the remaining tests were not carried out until a week later. The apple that had originally been used the previous week was used a second time. However after a week spent in a box the apple had dried out. The reason for this was that the cut parts of the apple had lost moisture through evaporation. The surface of the cut apple would therefore have had a lower water potential than the rest of the apple, resulting in osmosis and water moving to the areas of lower water potential, until the water potential was even. However this would not have happened as water would have continued to evaporate and so would osmosis in an attempt to even out the water potential of the apple. The final result is an apple with little water (moisture) and a very low water potential.

Reliability cannot be proved by simply repeating the test a number of times, only if the results collected are similar can they be declared as reliable. This can be shown mathematically by using a process known as standard deviation. Standard deviation shows how reliable the mean is by showing how close the results are to it, therefore the smaller the standard deviation the more reliable the results. One can see from the results table of the potato investigation that the standard deviation is fairly small with the exception of 0.1$moldm^{-3}$ sucrose solution. The apple investigation's standard deviations showed that 0.2$moldm^{-3}$ and 0.3$moldm^{-3}$ sucrose solution tests were also unreliable.

Improvement to the procedure of the investigation would be:

- Do all the tests on the same day.
- If not possible cover plant material in cling film and keep refrigerated, this will help in the retention of moisture (water).
- Ensure that the samples mass only varies by a few hundredths of a gram.

If there had been time the apple investigation would have been repeated, as there were a number of anomalous results.

[1] Reason for second graph, which omitted this result.