Core practical 8: Investigate the effect of environmental conditions on water uptake in a plant shoot

Objectives

- Know how to determine water uptake in a leafy shoot
- Be able to investigate the effect of environmental conditions on water uptake

Safety

- Care should be taken not to break the capillary tubing when pushing the cutting into the connecting tubing.
- Students should let you know if they find the plant sap irritating to their skin.
- Ensure the room is well ventilated to remove small amounts of noxious gases that are given off by some plant materials.

Specification links

- Practical techniques 1, 2, 3, 8
- CPAC 1a, 2a–2d, 4a, 4b, 5b

Procedure

In this investigation water uptake by a plant shoot is measured under different environmental conditions using a bubble potometer. The potometer may be a simple length of capillary tubing or may have a reservoir to allow it to be refilled easily (see **fig A** in the Student sheet). When using this method, it can be assumed that the plant shoot takes up an equal volume of water to that lost by transpiration, so that transpiration rate is determined indirectly.

A number of different environmental conditions could be tested, including the following.

- Light: use a lamp at different distances and a light meter to record intensity.
- Air movement: use a fan at different distances or speed settings.
- Temperature: use an incubator set to different temperatures.
- Humidity: increase humidity by covering the shoot with a clear plastic bag.

Before starting your investigation, choose one environmental condition as your independent variable. Write how you will change this variable. Consider how other variables will be controlled. Using the same shoot for each condition will ensure that variables relating to the plant shoot, such as leaf area and stomatal density, will remain constant.

Procedure for using the potometer (steps 1 to 5 may have been done for you already):

1. Lay the capillary tube and rubber connector under water and fill both parts.
2. Carefully select a leafy shoot which has a stem as close in diameter to that of the rubber connector as possible. Keep the end of the shoot under water at all times while you select and trim it as necessary.
3. Quickly insert the shoot into the rubber connector, making sure the fit is as tight as possible. Carry out this step under water.
4. Firmly clamp the capillary tube to the stand with the shoot at the top end. Place the bottom end of the capillary tube into the beaker of water. Two people may need to carry out this step to avoid the leafy shoot becoming detached.
5. Smear petroleum jelly around the join between the shoot and the rubber connector to ensure an airtight seal. Dry the leaves by blotting them gently with paper towels.
6. Leave the apparatus for 5 minutes to allow water to be drawn up into the end of the capillary tube. A small air bubble should be present at the end of the capillary tube from the time spent out of water in step 4. If there is no air bubble, the tube can quickly be removed from the water and a piece of paper towel can be used to soak a little water from the open end. When the tube is replaced in the beaker of water a small air bubble should be visible.
7. Time the movement of this air bubble along the capillary tube for a set distance (e.g. 2 cm). Alternatively, measure the distance moved by the bubble in a set time (e.g. 5 minutes). Record your results in a suitable table.

8. Repeat the readings using different environmental conditions as instructed by your teacher. Reset the air bubble to the start each time if necessary. Your teacher will explain how to do this for the equipment that you are using. Allow the shoot to settle in the new conditions for 2–3 minutes before testing.
9. When your investigation is complete, work out the leaf area of your shoot by drawing around each leaf on graph paper and counting up the squares.

Notes on procedure

- Much time and frustration can be saved if steps 1–5 are carried out for the students in advance. It may be necessary to designate a different environmental condition to be tested by each group to spread out demands on equipment.
- If possible, use one lesson to demonstrate the use of the potometer and a second to investigate the effect of environmental differences.
- The tubing must be filled with water before the practical starts.
- If air comes into contact with exposed xylem vessels, air will enter the xylem in the shoot and an airlock will form, which will prevent water uptake. The stem must be re-trimmed underwater.
- The connection between the shoot and the rubber tubing must be airtight. Small plastic cable ties or a twist of garden wire can be used to gently secure the connection if necessary.
- Ensure students dry the leaves and keep them dry throughout the procedure.
- Check that students do not allow the shoot to become detached from the tubing.
- Students may need help introducing an air bubble into the bottom of the capillary tube.
- In step 7, timing the movement of the air bubble for a set distance along the capillary tube is probably best as this avoids the air bubble being drawn up too far and reaching the shoot.
- Resetting of the air bubble may not be needed for every new test. It is easily achieved if a potometer with a water reservoir is being used. If not, applying a paper towel to the end of the capillary tubing should draw water and the bubble back towards the start. If this does not work, keeping the tube end underwater while gently squeezing the rubber connecting joint may have the desired effect.
- Students often need help converting between units, especially when working with areas or volumes. Remind students that 1 cm^2 = 100 mm^2 and that 1 cm^3 = 1000 mm^3, for example.

Answers to questions

1. Without an airtight seal, water will not be drawn up the capillary tube and no air bubble movement will be possible. Water movement in the capillary tube and in the xylem relies on cohesive forces between water molecules. Air bubbles prevent the cohesive forces and stop the upward tension being transmitted through the entire water column.
2. Limitations may include:
   - factors that influence transpiration and are difficult to control, such as the size of the shoot, the number of leaves on the shoot, the total surface area of the leaves, leaves that have not been dried fully or differences in density of stomata between leaves
   - factors that reduce the accuracy of the equipment, such as the difficulty in making a seal between the shoot and the apparatus.
3. The limitations may have the following effects.
   - Transpiration rate will increase if a larger leaf surface area is used or if there are more leaves present. Wet leaves reduce the diffusion gradient for water.
   - The lack of an airtight seal will slow air bubble movement and may stop transpiration altogether. This will produce a lower value for the water uptake rate.

4. The limitations could be reduced in the following ways.
   - Shoots should be the same size, with the same number of leaves. Leaf area should be measured so that water loss per unit area can be calculated.
   - The seal between the shoot and the apparatus must be as airtight as possible. This may be achieved by using a flexible material and adding a sealant.
   - Repeating the experiment a large number of times will give a measure of reliability and will allow a mean to be calculated.

Sample data

Air movement/fan setting	Water uptake/mm^3 min^{-1}			
	Repeat 1	Repeat 2	Repeat 3	Mean
off	3.1	3.2	2.8	3.0
low	6.2	5.5	5.9	5.9
medium	8.1	7.8	7.8	7.9
high	9.9	9.9	9.7	9.8

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Safety

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All the maths you need

- Recognise and make use of appropriate units in calculations.
- Recognise and use expressions in decimal and standard form.
- Use an appropriate number of significant figures.
- Substitute numerical values into algebraic equations using appropriate units for physical quantities.
- Solve algebraic equations.
- Calculate the circumferences, surface areas and volumes of regular shapes.

Equipment

- large leafy shoot – the end must be cut and kept under water
- capillary tubing with a short rubber connecting tube attached at one end
- grease pencil or fine marker pen
- ruler
- clamp stand
- 250 cm^3 or 500 cm^3 beaker
- stop clock
- fan, or dark cupboard, lamp and light meter, or plastic bag, or incubators
- petroleum jelly
- small plastic cable ties or thin garden wire
- sharp scissors

Diagram

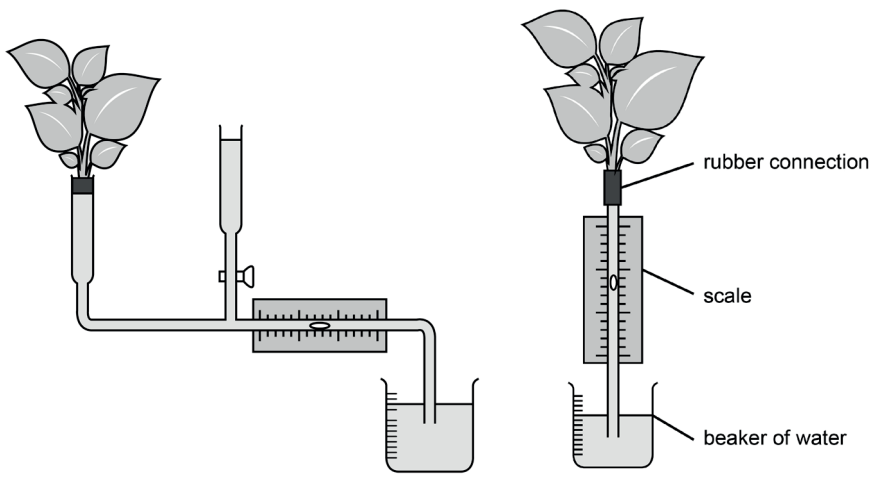

fig A Two types of potometer

Procedure

In this investigation water uptake by a plant shoot is measured under different environmental conditions using a bubble potometer. The potometer may be a simple length of capillary tubing, or may have a reservoir to allow it to be refilled easily (see **fig A**). When using this method, it can be assumed that the plant shoot takes up an equal volume of water to that lost by transpiration, so that transpiration rate is determined indirectly.

A number of different environmental conditions could be tested, including the following.

- Light: use a lamp at different distances and a light meter to record intensity.
- Air movement: use a fan at different distances or speed settings.
- Temperature: use an incubator set to different temperatures.
- Humidity: increase humidity by covering the shoot with a clear plastic bag.

Before starting your investigation, choose one environmental condition as your independent variable. Write how you will change this variable. Consider how other variables will be controlled. Using the same shoot for each condition will ensure that variables relating to the plant shoot, such as leaf area and stomatal density, will remain constant.

Procedure for using the potometer (steps 1 to 5 may have been done for you already):

1. Lay the capillary tube and rubber connector under water and fill both parts.
2. Carefully select a leafy shoot which has a stem as close in diameter to that of the rubber connector as possible. Keep the end of the shoot under water at all times while you select and trim it as necessary.
3. Quickly insert the shoot into the rubber connector, making sure the fit is as tight as possible. Carry out this step under water.
4. Firmly clamp the capillary tube to the stand with the shoot at the top end. Place the bottom end of the capillary tube into the beaker of water. Two people may need to carry out this step to avoid the leafy shoot becoming detached.
5. Smear petroleum jelly around the join between the shoot and the rubber connector to ensure an airtight seal. Dry the leaves by blotting them gently with paper towels.
6. Leave the apparatus for 5 minutes to allow water to be drawn up into the end of the capillary tube. A small air bubble should be present at the end of the capillary tube from the time spent out of water in step 4. If there is no air bubble, the tube can quickly be removed from the water and a piece of paper towel can be used to soak a little water from the open end. When the tube is replaced in the beaker of water a small air bubble should be visible.
7. Time the movement of this air bubble along the capillary tube for a set distance (e.g. 2 cm). Alternatively, measure the distance moved by the bubble in a set time (e.g. 5 minutes). Record your results in a suitable table.
8. Repeat the readings using different environmental conditions as instructed by your teacher. Reset the air bubble to the start each time if necessary. Your teacher will explain how to do this for the equipment that you are using. Allow the shoot to settle in the new conditions for 2–3 minutes before testing.
9. When your investigation is complete, work out the leaf area of your shoot by drawing around each leaf on graph paper and counting up the squares.

Analysis of results

1. Your initial results will be in terms of distance moved by the bubble. Convert your results into the volume of water taken up as follows. First measure the radius of the hole in the capillary tubing. Then use the formula for the volume of a cylinder $v = \pi r^2 h$ to calculate the volume of water. In this case, h is the distance moved by the bubble.
2. Calculate the rate of water uptake as volume taken up per unit of time using the formula:
$$\text{rate of water uptake} = \frac{\text{volume of water taken up}}{\text{time taken for the bubble to move}}$$
. Units are typically in $mm^3\,min^{-1}$.
3. To compare different shoots, the leaf area of each shoot must be taken into account. This is done by converting results into volume of water per unit of leaf area per unit time, e.g. $mm^3\,m^{-2}\,hour^{-1}$. Divide your water uptake rate results by the leaf area from your measurements. Express your results in suitable units.

Learning tips

- The experiment measures the rate of water uptake. As this is proportional to the rate of transpiration it can be taken as a measure of transpiration rate if we assume that the rate of water loss equals the rate of water uptake. To measure water loss directly, loss in mass could be measured.

Questions

1. Why is it necessary to form an airtight seal?
2. The limitations of an investigation are factors that reduce the accuracy and reliability of results. They may arise from variables that are difficult to control. What are the limitations of this procedure?
3. What effect will these limitations have on your data?
4. Suggest ways of reducing the limitations to give more accurate and reliable data.

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Safety

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- Ensure the room is well ventilated to remove small amounts of noxious gases that are given off by some plant materials.

Equipment per student/group	Notes on equipment
large leafy shoot – the end must be cut and kept under water. The cut shoots should be kept under water in a bucket or sink until needed.	To save time it is very beneficial if the potometers are set up for students in advance and kept in water to avoid bubbles developing in the capillary tube. See steps 1–5 of the Student sheet for guidance. Choose leafy shoots with stem diameters that will give a tight fit in the rubber tubing; this is vital for the success of the practical. Woody stems such as beech or privet are best as they are easy to fit into the potometer. Cuttings should be at least 15 cm long.
capillary tubing with a short rubber connecting tube attached at one end	Capillary tubing with a bore diameter of 1 mm is suitable.
grease pencil or fine marker pen	Pens suitable for marking the capillary tubing
ruler	One per group
clamp stand	One per group
250 cm^3 or 500 cm^3 beaker	One per group
stop clock	One per group
fan	To allow air movement to be varied
dark cupboard, lamp and light meter	To allow light levels to be varied
plastic bag	To allow humidity to be increased
incubators	To allow temperature to be varied
petroleum jelly	To seal joints
small plastic cable ties or thin garden wire	Use to gently secure the seal between rubber tubing and plant shoot if necessary.
sharp scissors	To trim the shoot under water. Blades must be sharp to avoid crushing the xylem vessels.

Notes