MINISTRY OF EDUCATION, SINGAPORE
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CAMBRIDGE INTERNATIONAL EDUCATION
General Certificate of Education Advanced Level

CANDIDATE NAME	

CENTRE NUMBER	S					INDEX NUMBER				

0123456789

BIOLOGY **9477/04**

Paper 4 Practical **For examination from 2026**

SPECIMEN PAPER **2 hours 30 minutes**

You must answer on the question paper.

You will need: The materials and apparatus listed in the confidential instructions

INSTRUCTIONS
- Answer **all** questions.
- Use a black or dark blue pen. You may use an HB pencil for any diagrams or graphs.
- Write your name, centre number and index number in the boxes at the top of the page.
- Write your answer to each question in the space provided.
- Do **not** use an erasable pen. Do **not** use correction fluid or tape.
- Do **not** write on any bar codes.
- You may use an approved calculator.
- Write the details of the shift and laboratory in the boxes provided.

INFORMATION
- The total mark for this paper is 50.
- The number of marks for each question or part question is shown in brackets [ ].

Shift
Laboratory

For Examiner's Use	
1	
2	
3	
Total	

This document has **18** pages.

Singapore Examinations and Assessment Board

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Answer **all** questions.

1 In this question you will investigate the effect of light intensity on the rate of photosynthesis.

(a) Sketch a fully labelled graph to show the expected relationship between the rate of photosynthesis and light intensity, as light intensity increases.

Explain the shape of your graph.

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.... [4]

In your investigation, light intensity will be controlled using five filters, **F1**, **F2**, **F3**, **F4** and **F5**. The percentage light intensity given by each filter when used with a standard light source is shown in Table 1.1.

Table 1.1

filter	percentage light intensity
F1	100.0
F2	70.0
F3	50.0
F4	25.0
F5	12.5

You are provided with:

- several pieces of pondweed (*Cabomba* sp.) in a beaker of 1% sodium hydrogencarbonate solution, labelled **P**, illuminated by a bench lamp
- five filters, labelled **F1**, **F2**, **F3**, **F4** and **F5**, as shown in Table 1.1
- a 20 cm^3 syringe attached to a capillary tube by plastic tubing with an air-tight seal
- ethanol, in a small specimen tube with a cap labelled **E**
- 50 cm^3 of 1% sodium hydrogencarbonate solution to which a small amount of detergent has been added, in a beaker labelled **D**.

Ethanol is flammable and harmful. Wear eye protection and wash off any splashes on skin with tap water.

Sodium hydrogencarbonate is a source of dissolved carbon dioxide.

Read through steps 1 to 14 and prepare a table to record your results in **1(b)** before starting the investigation.

Proceed as follows.

Steps 1–9 outline the procedure to assemble the apparatus to measure the rate of photosynthesis of the pondweed. The apparatus is shown in Figure 1.1.

step 1 Use a retort stand and clamp to support the barrel of the 20 cm^3 syringe (with the plunger removed), so that the end of the capillary tube is over an empty beaker. Ensure that the positioning of the clamp is as high as possible on the syringe barrel so that it will **not** block light from reaching the pondweed.

step 2 Observe the pieces of pondweed in **P** to identify the piece that is releasing oxygen bubbles from its cut stem most rapidly. Request new material if there are no pieces that are actively bubbling.

step 3 Put the actively bubbling piece of pondweed into the barrel of the syringe and fill the syringe with the solution from **P**, as shown in Figure 1.1. Use a finger to block the open end of the capillary tube while filling the barrel of the syringe. Tap the barrel to help release any bubbles trapped near the base of the syringe.

step 4 Remove your finger from the open end of the capillary tube and insert the plunger into the barrel of the syringe. Gently push the plunger down about 1 cm until some of the solution is forced out of the end of the capillary tube and there are no air bubbles in the capillary tube.

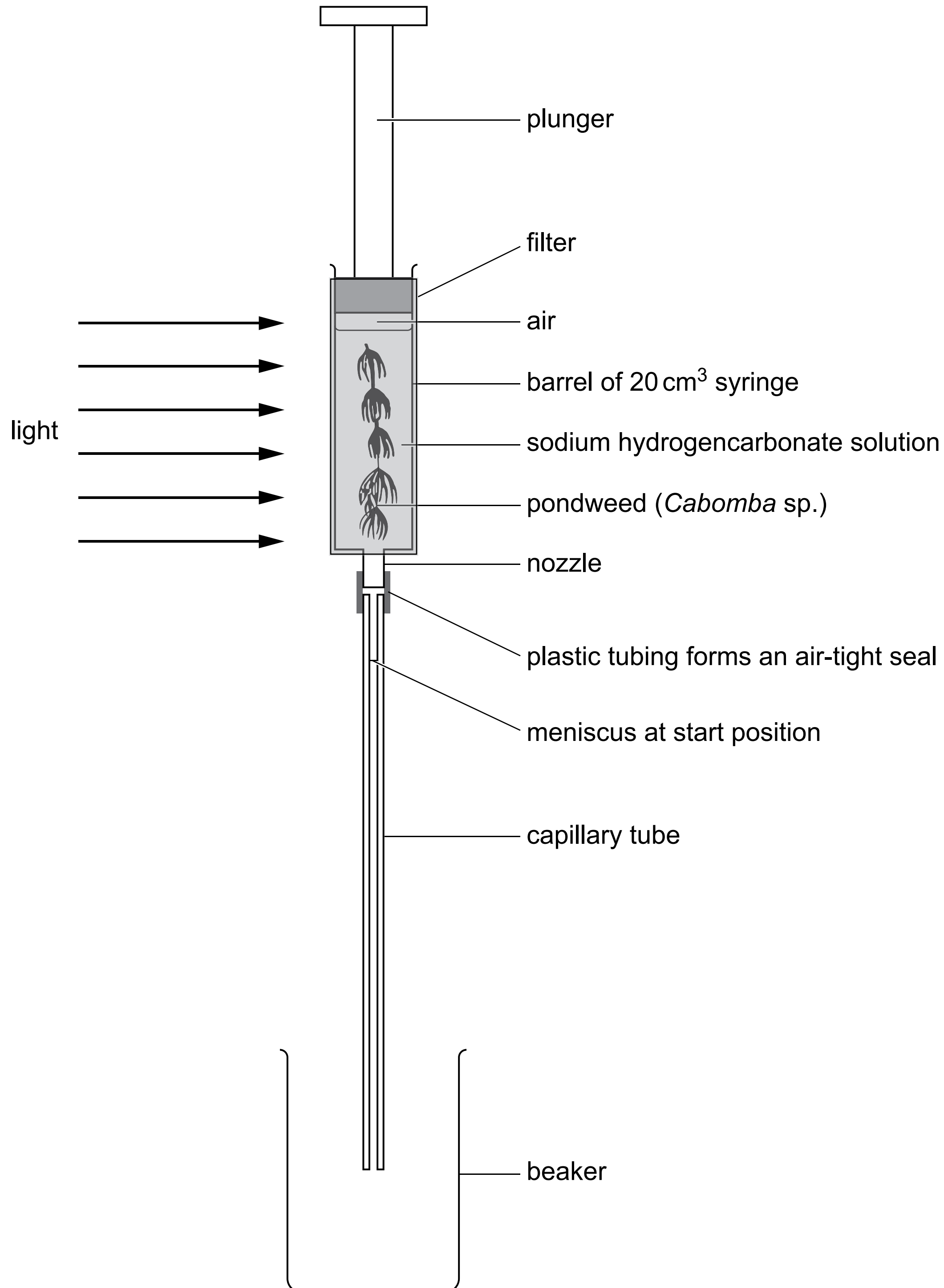

Figure 1.1

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step 5 Replace the empty beaker with the beaker containing solution **D** and adjust the clamp so that the open end of the capillary tube is below the surface of solution **D**. Carefully use the plunger to draw just enough of solution **D** into the capillary tube so that it reaches the nozzle of the syringe. This coats the inner surface of the capillary tube with detergent, which helps to ensure a smooth flow of liquid through the capillary tube during subsequent steps. Remove the beaker containing solution **D** and replace with the empty beaker.

step 6 Wipe the end of the capillary tube dry with a paper towel. Use the plunger to slowly draw up the column of liquid in the capillary tube until there is a clear meniscus a little below the plastic tubing, as shown in Figure 1.1. This is the start position. Mark the start position with a marker pen.

step 7 Position a lamp so that it will shine directly onto the syringe containing the pondweed. The lamp should be as close as possible to the pondweed, but no closer than 10 cm. The lamp should remain in this position throughout the investigation.

step 8 Switch on the lamp. From this point on, do **not** switch off the lamp. The pondweed should be maintained under constant illumination.

step 9 Carefully wrap filter **F1** around the barrel of the syringe and secure in place using a piece of Blu Tack™ or sticky tape on the side opposite to the light source. Leave the apparatus for two minutes to equilibrate.

step 10 If, after two minutes, the meniscus has moved down the capillary tube, continue from step 11. If the meniscus is **not** moving, select a new piece of pondweed that is actively bubbling and re-set the apparatus by repeating steps 1 to 9.

step 11 Use the plunger of the syringe to draw the meniscus back up to the mark for the starting position. Start a stopwatch and, after two minutes, mark the final position of the meniscus with a marker pen. Record the distance moved by the meniscus in two minutes in the table prepared in **1(b)**.

If the flow of liquid through the capillary tube becomes jerky when taking measurements, carefully push the plunger down until there are no air bubbles in the capillary tube. Then repeat steps 5 and 6 to reset the level of the meniscus to the start position.

step 12 Carefully remove filter **F1** and clean off the final position mark on the capillary tube (from step 11) using ethanol, **E**.

step 13 Repeat step 9 with filter **F2** followed, after two minutes, by step 11. Step 10 should **not** be repeated. After recording the result for filter **F2** in the table provided in **1(b)**, remove filter **F2** and clean off the final position mark on the capillary tube using ethanol, **E**.

step 14 Repeat step 13 with filters **F3**, **F4**, and **F5** in turn.

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(b) Record your results at each light intensity in a suitable form in the space below.

[5]

(c) **(i)** Use the grid on the page opposite to display your results from **1(b)**. [4]

(ii) Discuss what these results suggest about the relationship predicted in **1(a)**.

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.... [2]

(d) Suggest the limiting factor for photosynthesis that is acting when filter **F5** is used.

Explain your answer.

limiting factor ....

explanation ....

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[1]

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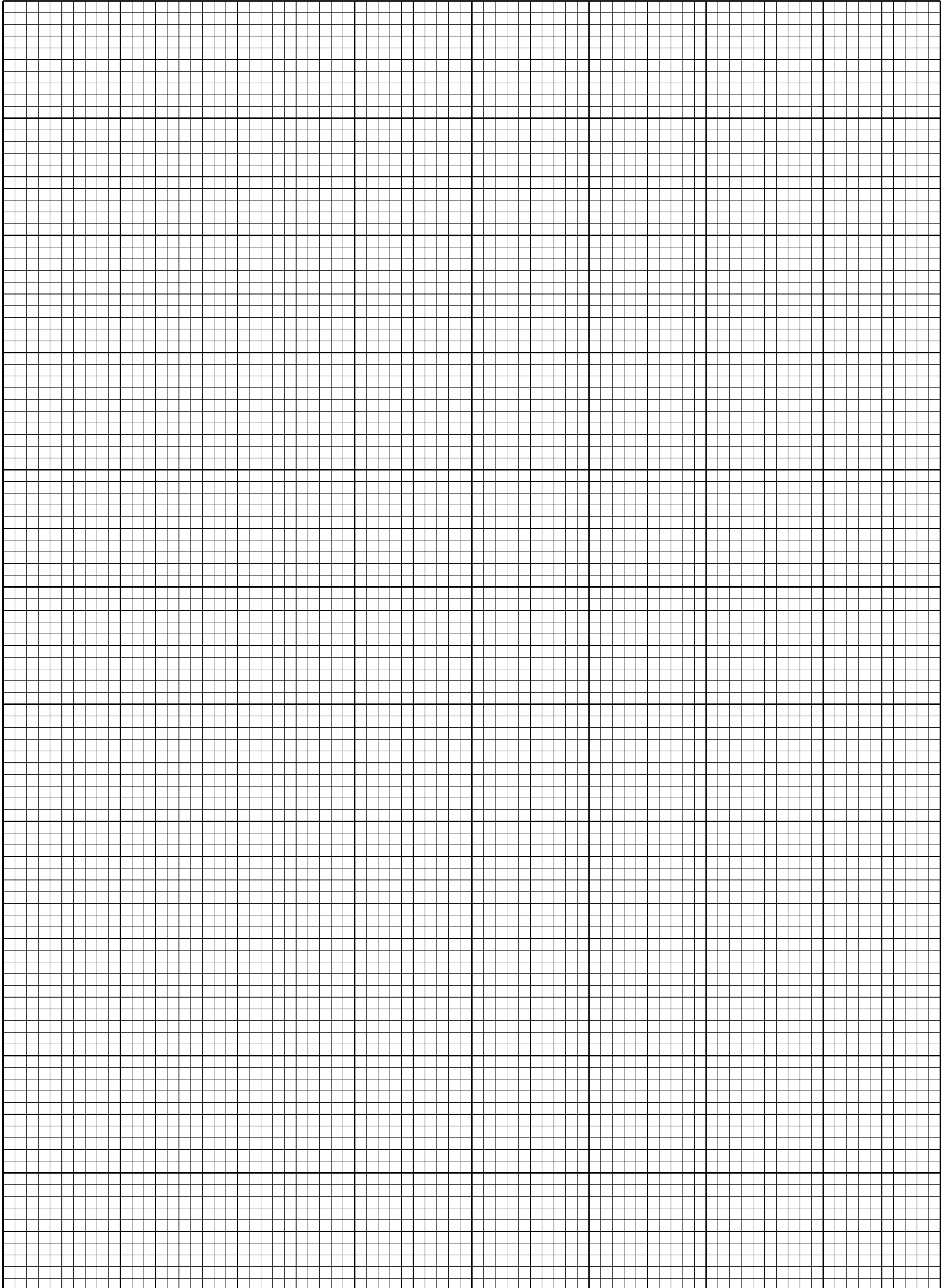

(e) Suggest how adding sodium hydrogencarbonate solution to the pondweed in the syringe increases the validity of the conclusions.

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.... [2]

(f) One way to increase confidence in the conclusions of this investigation would be to repeat the experiment several times.

Describe **two** other modifications to the method that would increase confidence in the conclusions **and** explain how these modifications would achieve this.

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.... [2]

[Total: 20]

2 During this question you will require access to a microscope and slide **S1**.

You will investigate the density of stomata on the lower surface of a leaf.

You are provided with a leaf, labelled **L**.

Proceed as follows.

step 1 Cover a small area (about 1 cm^2) of the lower epidermis of **L** with a thin layer of clear nail varnish. Make sure that you do **not** cover an area with large veins. Repeat this **three** more times on different areas of the lower epidermis to ensure that you have spare material if necessary.

step 2 Put **L** to one side for **at least** twenty minutes to allow the nail varnish to dry.

step 3 Work through parts **2(f)(i)** and **2(f)(ii)** while you wait for the nail varnish to dry or go on to other parts of the Question Paper as appropriate.

step 4 After allowing the nail varnish to dry for **at least** 20 minutes, carefully use a single-sided razor blade or the blade of a fine scalpel to lift one edge of the layer of nail varnish. You may then use blunt forceps to gently peel the layer of nail varnish from the leaf. If the layer does **not** come off in one piece, select the largest piece that is available. Transfer the layer to a microscope slide. Gently place a coverslip on the thin layer. No water is required.

(a) Examine the slide using a microscope and identify the stomata. Observe the thin layer using the three objective lenses and choose the lens that is **most** suitable for counting the number of stomata in the field of view.

Show which objective lens you have decided to use by placing a tick (✓) in the appropriate box and give a reason for your choice.

☐ low-power objective lens (×4)

☐ medium-power objective lens (×10)

☐ high-power objective lens (×40)

reason ....

....

.... [1]

(b) **(i)** Using the objective lens selected in **2(a)**, complete Table 2.1 so that in **2(b)(ii)** the **mean** number of stomata in a field of view can be calculated.

Count all the stomata for which the **centre** of the stomatal pore is visible in the field of view.

Table 2.1

number of stomata in a field of view

[1]

(ii) Calculate the **mean** number of stomata per field of view for **L**, using your data from Table 2.1.

Show your working.

mean number of stomata per field of view for **L** = ...
[2]

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(c) Focus the objective lens that you used for counting stomata on the stage micrometer provided.

(i) Measure the radius of the field of view and record this radius in mm.

radius, r, of field of view = ... mm [1]

(ii) Calculate the area of the field of view and record this area in mm^2.

area of circle = πr^2
$\pi = 3.14$

area of field of view = ... mm^2 [1]

(d) Using your results from **2(b)(ii)** and **2(c)(ii)**, calculate the mean density of stomata for **L** and record this density as number of stomata per mm^2.

mean density of stomata for **L** = ... per mm^2 [1]

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(e) Leaves that are **not** regularly exposed to full sunlight often show adaptations to relatively low light intensities. Such leaves are known as shade leaves. Leaves that grow in full sunlight and are adapted to relatively high light intensities are called sun leaves.

One adaptation of leaves to differing light intensities may be shown by the density of stomata on the leaves.

A student recorded the density of stomata for shade and sun leaves of a different species of plant and carried out a statistical test to determine whether there was a significant difference between the mean stomatal density for these two types of leaves.

(i) State a statistical test that could have been used to determine whether the difference in mean stomatal density between the sun and shade leaves is significant.

.... [1]

(ii) A summary of the student's results is shown in Table 2.2.

Table 2.2

density of stomata / mm^{-2}		**significance of difference**
shade leaves	**sun leaves**	
286	325	$p < 0.05$

Comment on what these results show **and** suggest an explanation for any pattern.

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.... [4]

(f) **(i)** Slide **S1** is a microscope slide of the lower epidermis of a leaf of *Allium*. Examine the slide under the high-power objective lens of your microscope and locate a stomatal pore.

Make a detailed, labelled drawing in the space below of the stomatal pore, its guard cells and three adjacent epidermal cells.

[5]

(ii) Using the eyepiece graticule fitted in the eyepiece lens of your microscope and the stage micrometer, determine the actual length, in micrometres (μm), of one of the guard cells that you have drawn.

Show the measurements that you made and your working.

length of guard cell = ... μm
[3]

[Total: 20]

3 All green plants photosynthesise in the light, taking in carbon dioxide and releasing oxygen. They also respire continuously, taking in oxygen and releasing carbon dioxide. The light intensity at which photosynthesis and respiration occur at the same rate, so that there is no net gas exchange, is called the compensation point.

Compensation points can be investigated using hydrogencarbonate indicator solution. This is harmless to living organisms but changes colour over a range of concentrations of carbon dioxide due to changes in pH, as shown in Table 3.1.

Table 3.1

percentage concentration of carbon dioxide	colour
0.04 (normal atmospheric)	red
falling below 0.04	turns purple
rising above 0.04	turns yellow

Monitoring the colour of hydrogencarbonate indicator solution in sealed vessels containing living plant material can show whether carbon dioxide is being taken in or given out.

Ivy plants produce both shade and sun leaves depending on where the leaves develop. A single ivy plant produces sun leaves at the top where the leaves are in direct sunlight and produces shade leaves lower down where light intensity is reduced.

Using this information and your own knowledge, plan an investigation to find the light intensity at which shade leaves and sun leaves from an ivy plant reach their compensation points.

In your plan you must use:

- hydrogencarbonate indicator solution
- sun and shade leaves from ivy.

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You may select from the following apparatus and plan to use appropriate additional apparatus:

- normal laboratory glassware, e.g. test-tubes, boiling tubes, beakers, measuring cylinders, graduated pipettes, glass rods, etc.
- syringes
- timer, e.g. stopwatch
- bungs
- bench lamp with 9 W LED bulb.

Your plan should:

- have a clear and helpful structure such that the method you use is able to be repeated by anyone reading it
- describe how results will be collected so that results are as accurate and repeatable as possible
- be illustrated by relevant diagram(s), if necessary
- identify the dependent variable and the independent variable
- identify the variables you will need to control
- use the correct technical and scientific terms.

You can assume that a risk assessment for this investigation has already been carried out so there is no need to include reference to safety measures to minimise any risks associated with the proposed experiment. [10]

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