

**TEMASEK JUNIOR COLLEGE**  
**2023 JC2 PRELIMINARY EXAMINATION**  
**Higher 2**



CANDIDATE  
NAME

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CENTRE  
NUMBER

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INDEX  
NUMBER

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**BIOLOGY**

**9744/04**

Paper 4 Practical

**30 August 2023**

**2 hour 30 minutes**

Candidates answer on the Question Paper.

Additional Materials: As listed in the Confidential Instructions.

**READ THESE INSTRUCTIONS FIRST**

Write your name, CG, Centre number, index number on all the work you hand in.  
Give details of the practical shift and laboratory, where appropriate, in the boxes provided.

Write in dark blue or black pen.

You may use an HB pencil for any diagrams or graphs.

Do not use staples, paper clips, glue or correction fluid.

Answer **all** questions in the spaces provided on the Question Paper.

The use of an approved scientific calculator is expected, where appropriate.  
You may lose marks if you do not show your work or if you do not use appropriate units.

At the end of the examination, fasten all your work securely together.  
The number of marks is given in brackets [ ] at the end of each question or part question.

<b>Shift</b>
<b>Laboratory</b>

For Examiner's Use	
1	/ 17
2	/ 17
3	/ 21
Total	/ 55

Answer **all** questions.

- 1 (a) You will investigate the effect of different temperatures on the permeability of the cell surface membrane of beetroot cells.

Beetroot is a vegetable that contains a red pigment in its cells.

When beetroot tissue is put into water, the red pigment can move out of the cells through the cell surface membrane, changing the water to a red colour.

You are provided with:

labelled	contents	hazard	volume / cm <sup>3</sup>
<b>B</b>	4 pieces of beetroot	none	-
<b>W</b>	distilled water	none	100.0

The red pigment in beetroot cells can stain clothing and skin. Use blunt forceps to handle beetroot tissue and, if any pigment comes into contact with your skin, wash it off immediately under cold water.

It is recommended that you wear suitable eye protection.

You will need to:

- put beetroot tissue in water at different temperatures
  - leave the beetroot tissue in the water for a period of time
  - record the intensity of colour in the water at each temperature.
- (i) The temperature range that you will use must include a minimum temperature of 25°C and a maximum temperature of 65°C.

State **other** temperatures that you will use in your investigation.

[1]

**35°C, 45°C, 55°C**

- **Must have at least 3 other temperatures between the range**
- **Must have regular intervals**


*Read step 1 to step 13 before proceeding.*

**Proceed as follows.**

- 1 Cut the beetroot pieces into 2 mm thick discs. You will need at least 25 discs.
- 2 Put all the discs that you have cut into the beaker labelled **D** and cover them with approximately 30 cm<sup>3</sup> distilled water, **W**.
- 3 Stir with a glass rod.
- 4 Pour off the water into the beaker labelled **For waste**, leaving the discs in the beaker labelled **D**.
- 5 Put all the discs on a paper towel and blot them to remove excess water.

- 6 Set up and maintain a water-bath at 25°C, using the beaker labelled **water-bath**. The water-bath will be needed in step 9.
- 7 Put five discs into **one** test-tube.
- 8 Put 10 cm<sup>3</sup> of distilled water, **W**, into the test-tube with the five discs.
- 9 Put this test-tube into the water-bath for four minutes. You will need to maintain the water-bath at the correct temperature throughout these four minutes.
- 10 After four minutes, remove the test-tube with the discs from the water-bath.
- 11 Pour the water from this test-tube into a second test-tube, leaving the discs in the first test-tube.
- 12 Repeat step 1 to step 11 until all of the temperatures stated in **(a)(i)** have been tested, finishing with the maximum temperature of 65°C.
- 13 Observe the colour intensity of the water in each of the test-tubes in the test-tube rack.
- 14 Using only the symbols shown in Table 1.1 to represent intensity of colour, decide the intensity of colour in each of the test-tubes in the test-tube rack. Record your results in **(a)(ii)**.

Table 1.1

intensity of colour	symbol
dark red	++++++
 decreasing intensity of red colour	+++++
	++++
	+++
	++
	+
no colour	

- (ii) Record your results in an appropriate table, using only the symbols shown in Table 1.1.

Temperature / °C	Intensity of colour
25	+
35	++
45	+++
55	++++
65	+++++

**CH – Column heading with units, no units in table**

**D – record data for all 5 temperatures (ecf (a)(i)), no repeated data**

**Tr – Temperature increase, intensity increase (look at 25, and 65)**

**P – only symbols**

[4]

[TURN OVER

- (iii) Use your knowledge of cell surface membranes to explain the results that you recorded in (a)(ii). [3]


General ideas:

1. Reference to phospholipid bilayer / fatty acid tails
2. Reference to proteins denatured
3. Increased fluidity of membrane

1. Cell surface membrane is made up of a phospholipid bilayer
2. As temperature increases, more proteins in the membrane will be denatured,
3. Membrane will be more fluid, allowing more pigment to exit the cell.

- (iv) Identify **three** significant sources of error in this investigation. [3]

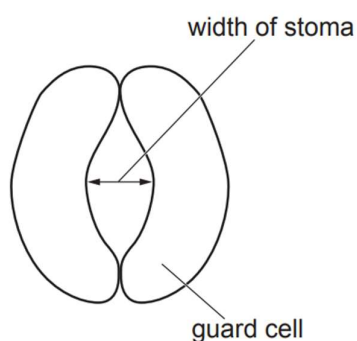
- (v) Suggest how you would make improvements to reduce **two** of the sources of error identified in (a)(iv). [2]

Source of error [3]	Improvement [2]
<p>1. Difficult to identify <u>intensity</u> of colour, subjective and prone to human error.</p> <p>Note: ½ mark if “intensity” is missing.</p>	<p>1. Use a colorimeter to detect intensity of colour</p>
<p>2. Difficult to maintain temperature throughout the experiment.</p> <p>R: Temperature not maintained (step 9).</p>	<p>2. Use thermostatically-controlled water-bath.</p>
<p>3. Not all pigment removed from beetroot tissue.</p> <p>These pigments would diffuse into the distilled water before the experiment resulting in inaccuracy.</p>	<p>3. Wash beetroot several times until no red colour in water.</p>
<p>4. Difficult to accurately cut 2 mm thick discs resulting in discs with different total surface area.</p>	<p>4. Use vernier caliper to measure 2 mm.</p> <p>Accept: Prepare thicker discs (e.g., 5mm) which are easier to measure and cut accurately.</p>
<p>5. Beetroot pieces were prepared from <u>different beetroot</u> in which the amount of pigments might differ from each other.</p>	<p>5. Ensure beetroot pieces provided were from the <u>same beetroot</u>.</p> 

6. Difficult to identify intensity of colour, subjective and prone to human error	6. Use a colorimeter to detect intensity of colour
7. Difficult to maintain temperature	7. Use thermostatically-controlled water-bath
8. Not all pigment removed from beetroot tissue before experiment	8. Wash beetroot several times until no red colour in water
9. Difficult to accurately cut 2 mm thick discs	9. Use vernier caliper to measure 2 mm

- (b) A scientist investigated changes in the mean width of stomata in the leaves of a plant growing in hot, dry conditions. The scientist measured the widths of stomata at different times of day, from dawn to midnight.

Fig. 1.1 shows where the scientist measured the width of each stoma.



**Fig. 1.1**

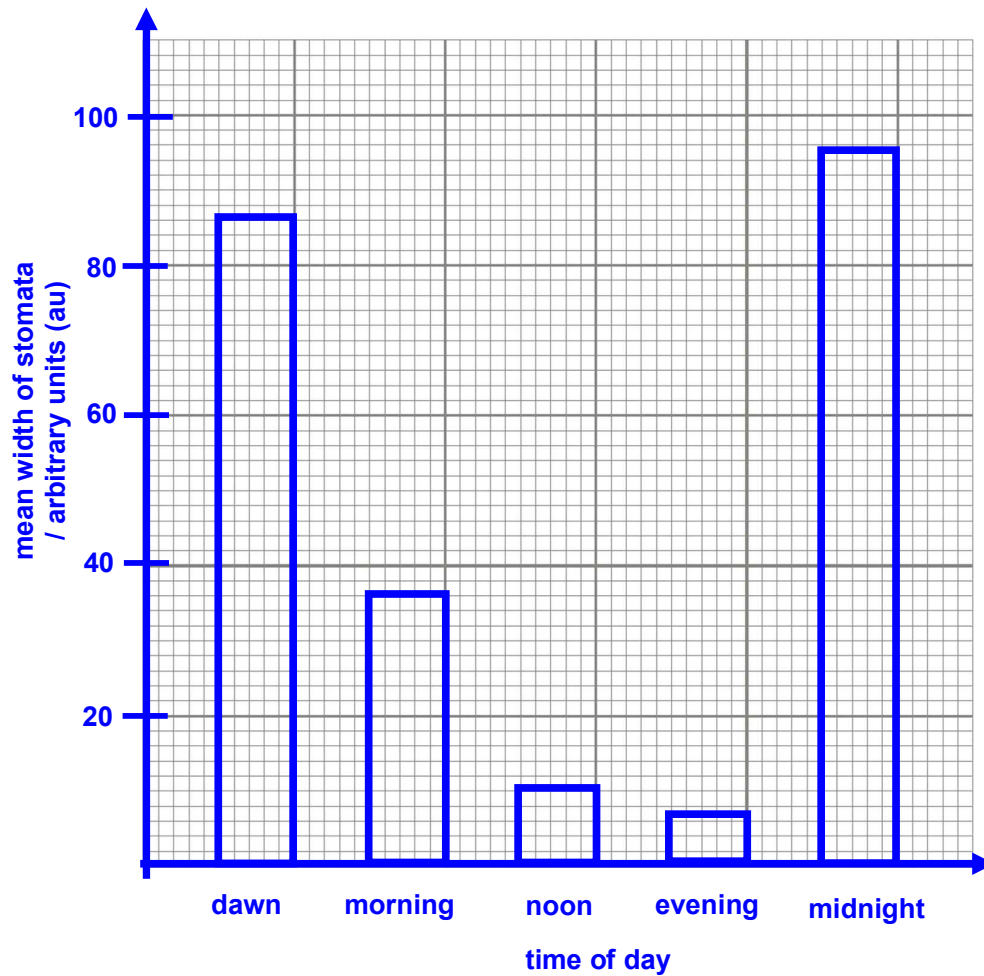
The scientist calculated the mean width of stomata for each time of day.

The results are shown in Table 1.2.

**Table 1.2**

time of day	mean width of stomata / arbitrary units (au)
dawn	86
morning	36
noon	10
evening	6
midnight	95

Use the grid provided to plot a graph in an appropriate form to display the data shown in Table 1.2. [4]



1. **S:**  
(x-axis) even width of bars;  
(scale on y-axis) label every 10 squares;
2. **P:**  
correct plotting of five bars;  
bars in order of table;  
equal spacing between bars;
3. **A:**  
(x-axis) time of day;  
(y-axis) mean width of stomata / au;
4. **BF:**  
five separate bars;  
bars drawn with even lines;  
labelled appropriately;  
**R:** shading of bars

[Total: 17]

- 2 (a) A group of students were given two species of plants, Species **A** and Species **B**. Their teacher also shared that one species is classified as a shade plant while the other is classified as a sun plant.

By measuring the rate of decolorization of DCPIP, describe how the students could design an experiment to investigate which species belongs to which category.

You must use:

- 50 g of leaves from Species **A** and Species **B**,
- 5 cm<sup>3</sup> of DCPIP solution,
- blender,
- 50 cm<sup>3</sup> cold extraction buffer solution,
- capillary tubes,
- aluminium foil,
- card tent,
- lamp which emits very little heat,
- timer, e.g. stopwatch
- ruler

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

- normal laboratory glassware e.g. test-tubes, beakers, measuring cylinders, graduated pipettes, glass rods, etc.
- syringes.

Your plan should:

- have a clear and helpful structure such that the method you use is able to be repeated by anyone reading it
- be illustrated by relevant diagram(s), if necessary
- identify the independent and dependent variables
- describe the method with the scientific reasoning used to decide the method so that the results are as accurate and repeatable as possible
- include layout of results tables and graphs with clear headings and labels
- use the correct technical and scientific terms
- include reference to safety measures to minimise any risks associated with the proposed experiment.

[13]

#### [IV, DV]

1. Independent variable: Light intensity / distance between lamp and plant extract + DCPIP [1/2]
2. Dependent variable: Time taken for decolorization of DCPIP [1/2]

#### [Hypothesis / Trend]

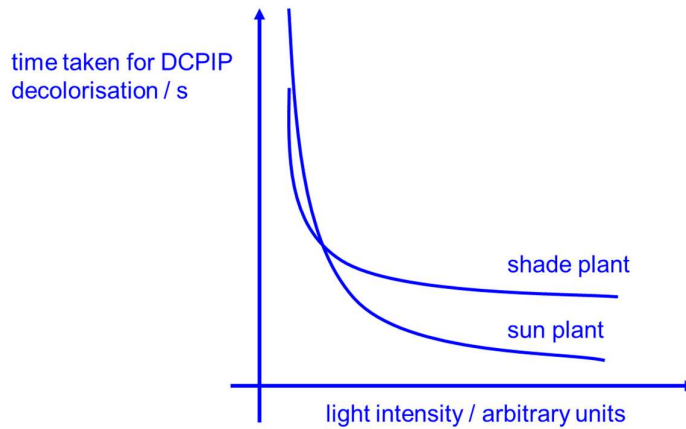
3. As light intensity increases, the time taken for decolorization of DCPIP decreases. [1]

If the species belongs to the sun plant category,

4. At low light intensity, the time taken for decolorization should be slower than the other species. [1/2]
5. At high light intensity, the time taken for decolorization should be faster than the other species. [1/2]

[TURN OVER

6. [Graph] [1]  
(A: x-axis is distance between lamp and plant extract)



7. [Data Table] [1]  
(A: IV is distance between lamp and plant extract / mm)

Light intensity / a.u.	Time taken for decolorization of DCPIP / s							
	Species A				Species B			
	Trial 1	Trial 2	Trial 3	Average	Trial 1	Trial 2	Trial 3	Average

[Vary IV – light intensity] – max 2

8. Place the lamp so that it shines along the surface of the bench. [1/2]
9. Place a piece of paper in front of the lamp and ensure the edge is 50 mm from the lamp, using a ruler. [1/2]  
(A: Use ruler to ensure distance between lamp and plant extract is constant)
10. Using a ruler, measure and mark on the paper 5 different distances, 100mm, 200mm, 300mm, 400mm, 500mm. [1]
11. Place the card tent over the piece of paper such that light from lamp is allowed to pass along the paper but not other light sources. [1/2]

[Constant Variable – chloroplast isolation] – max 2

12. Carefully remove any large veins using a scalpel. Discard the veins and retain the green leaf material. [1/2]
13. Cut the leaves of Species A into four small pieces, each about 3 cm by 3 cm. [1]  
(A: keep mass of leaves (after removal of vein) constant using mass balance)
14. Place the leaves in a blender. [1/2]
15. Using a syringe, add 3 cm<sup>3</sup> of cold buffer solution to the blender. [1/2]
16. Turn on the blender for 10 seconds to extract chloroplast. [1/2]  
(R: Chopping and grinding leaves – not consistent)
17. Transfer leaf extract to a Petri dish.
18. Immediately cover Petri dish with an aluminium foil. [1/2]



**[Constant Variable – amount of extract in each capillary tube and DCPIP]**

19. Using a syringe, add 1 cm<sup>3</sup> of blue DCPIP solution to the leaf extract in the Petri Dish and mix the liquids using a glass rod. [1/2]
20. Immediately cover with aluminium foil. [1/2]
21. Stand the ends of a capillary tube in the leaf extract-DCPIP mixture until the tube is filled to 5 mm mark. [1/2]
22. Immediately cover capillary tube with aluminium foil. [1/2]

**[Data Collection]**

23. Place the capillary tube on the 100mm mark on the piece of paper, under the card tent. [1/2]
24. Switch on the light and immediately start the stopwatch. [1/2]
25. Every 30 seconds, quickly remove the card tent and stop the stopwatch when decolorization is complete. Record the time taken for decolorization. [1/2]

**[Repeat for other IV]**

26. Repeat steps 21 – 25 to be placed on the 200mm, 300mm, 400mm, 500mm mark. [1/2]

**[Repeat for reliability]**

27. Repeat steps 12 – 26 two more times with fresh samples of Species A, cold buffer solution and DCPIP. [1]

**[Repeat for other species]**

28. Repeat steps 12 – 27 with species B instead of species A. [1/2]

**[Rate]**

29. Calculate rate by taking 1/time taken to decolorize. [1/2]

**[Conclusion]**

30. The species with more time taken at low light intensity and lesser time taken at high light intensity is the sun plant while the other is a shade plant. [1]

**[Risk]**

31. Medium risk: DCPIP, cold buffer solution are irritants. Wear goggles and gloves.
32. High risk: Prevent touching lamp with wet hands to prevent electrocution.

- (b) The students also found a published investigation on the effect of light intensity on stomatal density in the species *Lycopersicon esculentum*.

Two plants of *Lycopersicon esculentum* were selected. One was grown in high light intensity and the other was grown in low light intensity.

The results are shown in Table 2.1.

**Table 2.1**

leaf number	high light intensity			low light intensity		
	number of stomata $\times 10^3$		leaf area / cm <sup>2</sup>	number of stomata $\times 10^3$		leaf area / cm <sup>2</sup>
	upper surface	lower surface		upper surface	lower surface	
1	1634	3131	496	18	1277	160
2	1482	5072	509	10	906	115
3	1865	6365	637	14	1398	171
mean	1660	4856	547	14	1194	149

- (i) Calculate the percentage decrease in mean leaf area for leaves grown in low light intensity compared with those grown in high light intensity.

Your answer should be expressed as a whole number.

**73%** [1]

- (ii) The scientists who carried out the published investigation concluded that:

plants grown in higher light intensity have higher stomatal density **only** on the upper surface of the leaves compared to plants grown in lighter light intensity.

Evaluate whether the data in Table 2.1 supports this conclusion.

[3]

**Support [max 2]:**

1. Stomatal density on upper surface is higher in plants grown in high light intensity
2. Difference in mean stomatal density on upper surface (between the two light intensities) is greater than the difference between the lower surface
3. Stomatal density on lower surface is similar / very close
4. Leaf 1 stomatal density on lower surface is less than in plants grown in low light intensity
5. Calculate pair of stomatal densities (for 1 to 4)

**Do not support [max 2]:**

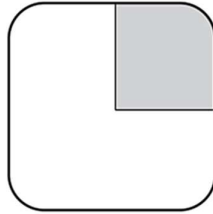
1. Mean stomatal density, on lower / both, surface(s) is higher in plants grown in high light intensity
2. For 2 out of 3 leaves (leaf 2 and 3), stomatal density is not higher only on the upper surface / is higher on lower / both, surface(s)
3. QF: Calculate pair of stomatal densities (for 1 to 2)
4. No statistical analysis done
5. Data only for one species

[Total: 17]

3 **P1** is a slide of a stained transverse section through a plant stem.

You are not expected to be familiar with this specimen.

- (a) Set up the microscope so that you can observe different tissues in the area of the stem on **P1** shown by the shaded region in Fig. 3.1.



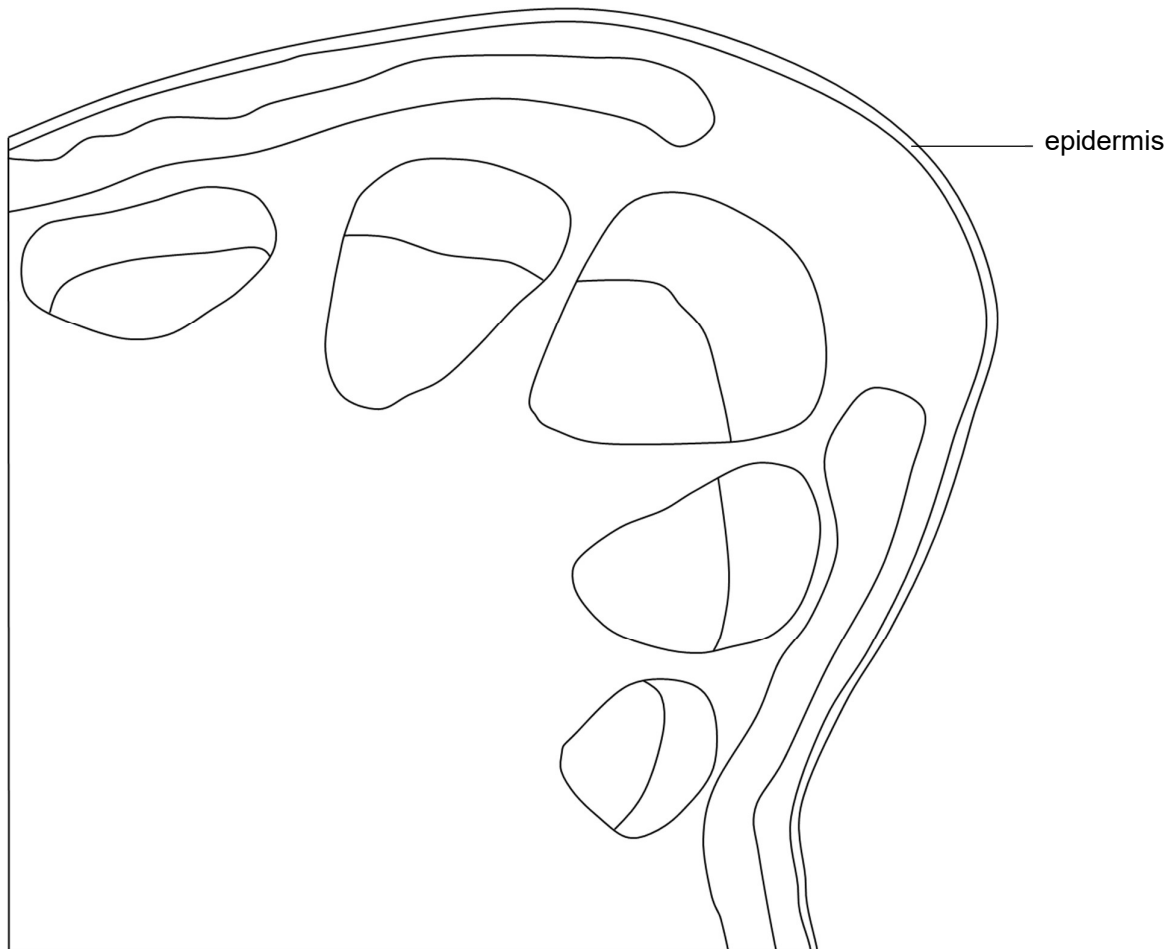
**Fig. 3.1**

*You are required to use a sharp pencil for drawings.*

- (i) Draw a large plan diagram of the area of the stem on **P1** shown by the shaded region in Fig. 3.1.

Your drawing should show the correct shapes and proportions of the different tissues.

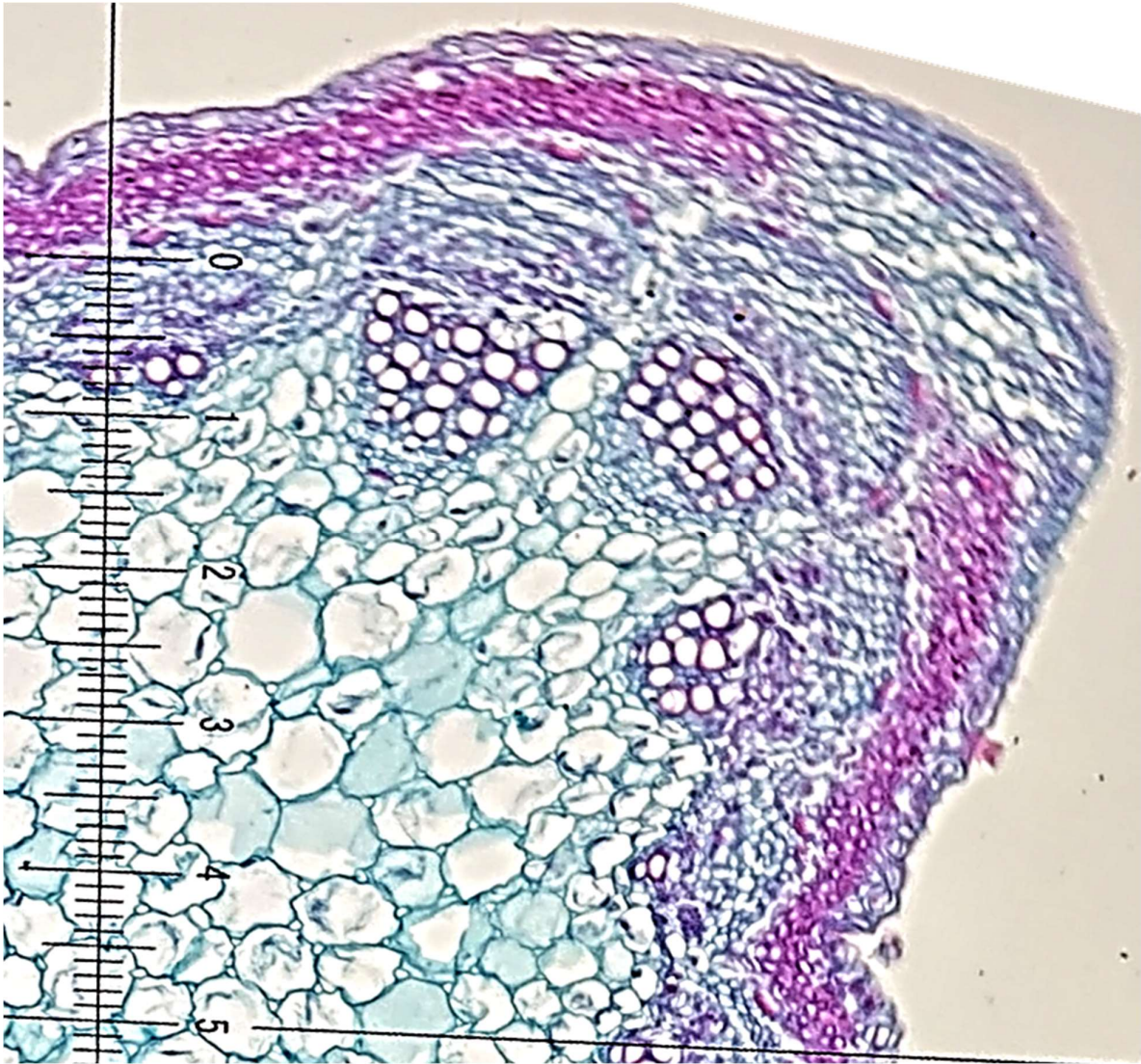
Use **one** ruled label line and label to identify the epidermis.



[5]

**[TURN OVER]**

M1	<ol style="list-style-type: none"> <li>1. clear, sharp, unbroken lines</li> <li>2. no shading</li> <li>3. Must <u>NOT</u> use ruler for any part of the drawing (except labelling line AND border lines)</li> </ol>	<b>Reject</b> <ul style="list-style-type: none"> <li>- if drawn over the print of question</li> <li>- feathery lines</li> <li>- overlaps or gaps</li> <li>- any lines thicker than 1mm</li> <li>- border line <u>CANNOT</u> project beyond epidermis</li> </ul>
M 2	<ol style="list-style-type: none"> <li>1. Must be correct section</li> <li>2. Must have 1 vertical and 1 horizontal border line drawn with ruler</li> <li>3. Border at least 100 mm</li> </ol>	
M 3	<ol style="list-style-type: none"> <li>1. no cells drawn</li> <li>2. use 4 horizontal lines which show 4 different layers of tissue</li> <li>4. Correct thickness of epidermis (3 mm)</li> <li>5. Dark purple layer is not continuous and should not be connected at the corner of the stem</li> </ol>	
M 4	<ol style="list-style-type: none"> <li>1. 1 large vascular bundle of correct shape, correct proportion and positioned at the corner of the stem</li> <li>2. Must show at least 4 smaller vascular bundles (2 on each side of the larger vascular bundle)</li> <li>3. Must draw a horizontal line to divide vascular bundle into xylem and phloem</li> <li>3. Proportion of xylem is about equal proportion to phloem</li> </ol>	<b>Reject any incomplete vascular bundles</b> <b>Cambium is not clearly visible. Do not need to indicate.</b>
M 5	<ol style="list-style-type: none"> <li>1. correct label with label line to epidermis</li> <li>2. use ruler to draw label line</li> </ol>	<b>Reject if labelled within drawn area</b>

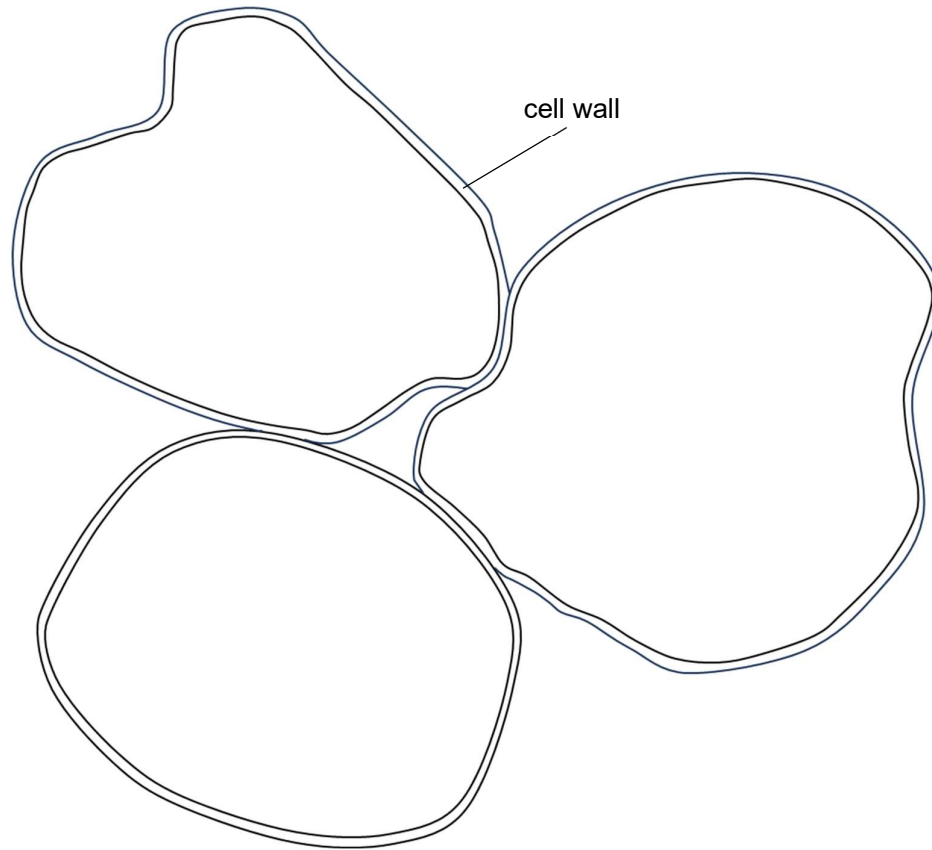


(ii) Observe the cells in the central tissue of the stem on **P1**.

Select **three** adjacent, touching cells of the central tissue.

Each cell that you select must touch at least two of the other cells.

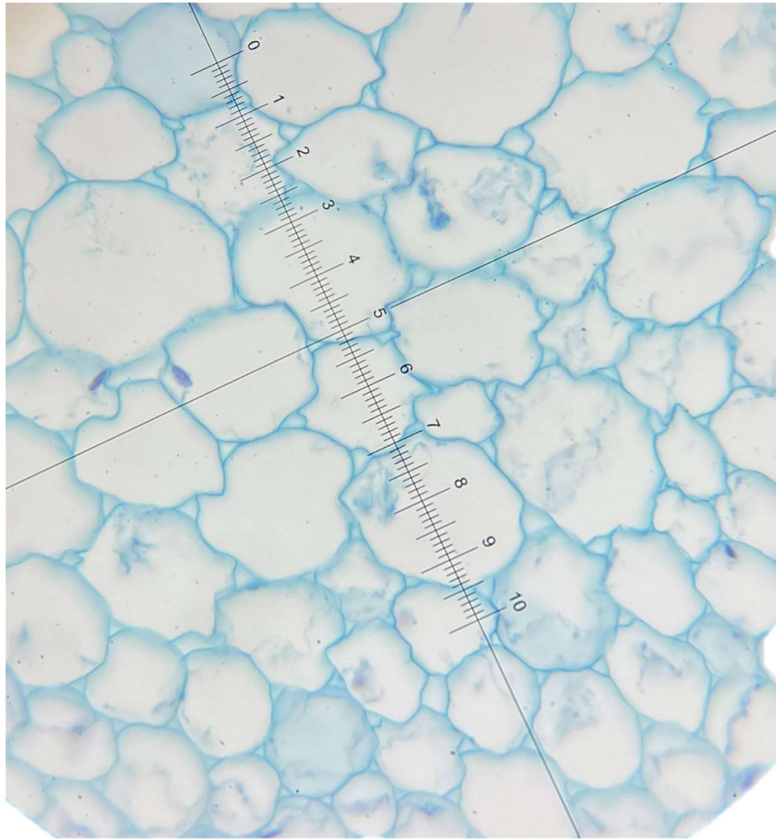
- Make a large drawing of this group of **three** touching cells.
- Use **one** ruled label line and label to identify the cell wall of **one** cell.



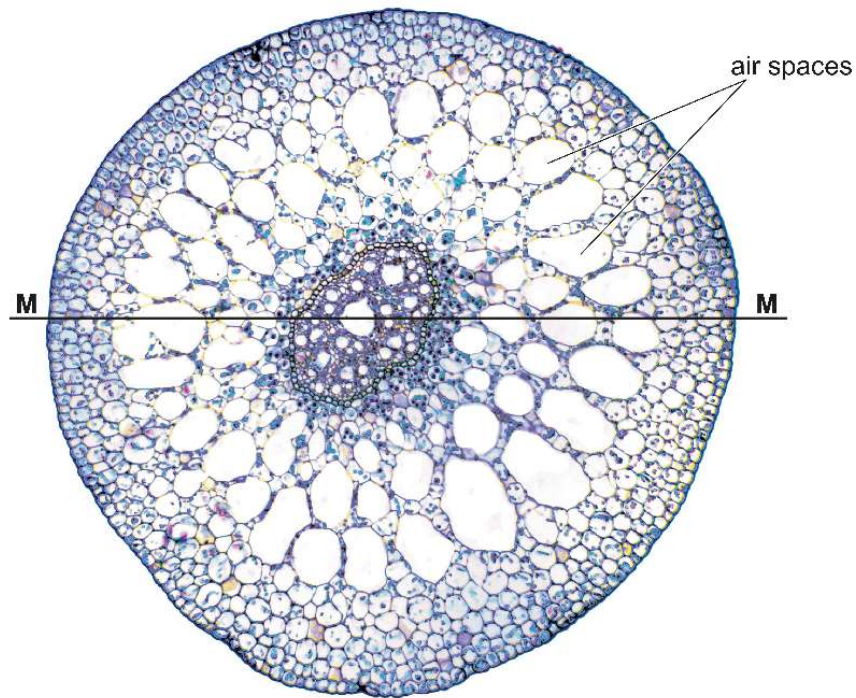
[5]

M 1	<ol style="list-style-type: none"> <li>1. clear, sharp, unbroken lines</li> <li>2. NO shading</li> <li>3. Largest cell 50 mm at widest part</li> </ol>	<b>Reject</b> <ul style="list-style-type: none"> <li>- if drawn over the print of question</li> <li>- feathery lines</li> <li>- overlaps or gaps or tails</li> <li>- any lines more than 1 mm</li> <li>- Cells must be large</li> </ul>
M 2	<ol style="list-style-type: none"> <li>1. Cell walls drawn as double lines.</li> <li>2. Separated by a space of 2 mm</li> </ol>	
M 3	<ol style="list-style-type: none"> <li>1. 3 cells drawn</li> <li>2. each cell of the group must touch at least two other cells</li> <li>3. Slightly rounded in shape</li> </ol>	
M 4	<ol style="list-style-type: none"> <li>1. Shows differences either in shape or size</li> <li>2. At least one intercellular space</li> </ol>	Do not give mark if EM organelles (e.g. mitochondria) included
M 5	<ol style="list-style-type: none"> <li>1. correct label with label line to cell wall</li> <li>2. label cell wall</li> </ol>	<b>Reject</b> if label within drawn area





- (b) Fig. 3.2 is a photomicrograph of a stained transverse section through a stem of a different type of plant. This plant is adapted to live submerged in water.



**Fig. 3.2**

- (i) Calculate the total area, in  $\text{mm}^2$ , of the image of the stem shown in Fig. 3.2.
- Assume that the stem is circular in cross-section.
  - Measure the diameter of the image of the stem along line **M–M**.
  - Show your working.

The formula for calculating the area of a circle is:

$$\text{area} = 3.14 \times r^2 \quad (r = \text{radius})$$

$$\text{diameter of the stem} = 97 \text{ mm (A: 96 – 98)}$$

$$\text{radius} = 97 \div 2 = 48.5 \text{ mm}$$

$$\text{area} = 3.14 \times 48.5^2 = 7\,386 \text{ mm}^2$$

[1] – correct diameter of the stem and units

[1] – value for the radius squared and multiplied by 3.14

[1] – correct answer

$$\text{area} = 7\,386 \text{ mm}^2 [3]$$



- (ii) In Fig. 3.2, the tissue that contains air spaces has an area in the image of 4500 mm<sup>2</sup>.

Use your answer in (b)(i) to calculate the percentage area of the stem shown in Fig. 3.2 that contains air spaces.

percentage area of stem that contains air space =  $4\,500 \div 7\,386 = 60.9\%$   
(accept ecf)

Answer to appropriate number of decimal places (whole no / 1 d.p.)

percentage area = 60.9 % [1]

- (iii) Describe how a more accurate measurement of the diameter of the stem can be obtained using the microscope. Include any additional apparatus that you might need. [3]

1. Calibrate eyepiece graticule with stage micrometer [1]
2. Align zero of eyepiece graticule with zero marking on stage micrometer. [1/2]
3. Locate another marking on graticule that match stage micrometer. [1/2]
4. length of 1 eyepiece division = length of stage micrometer division divided by total no. of eyepiece division. [1/2]
5. Measure radius using eyepiece graticule. [1/2]

Also accept:

6. Repeat measurement of radius to calculate average [1/2]

- (iv) The presence of a large number of air spaces in the stem is one adaptation of the plant shown in Fig. 3.2 for living submerged in water.

Suggest a function of these air spaces.

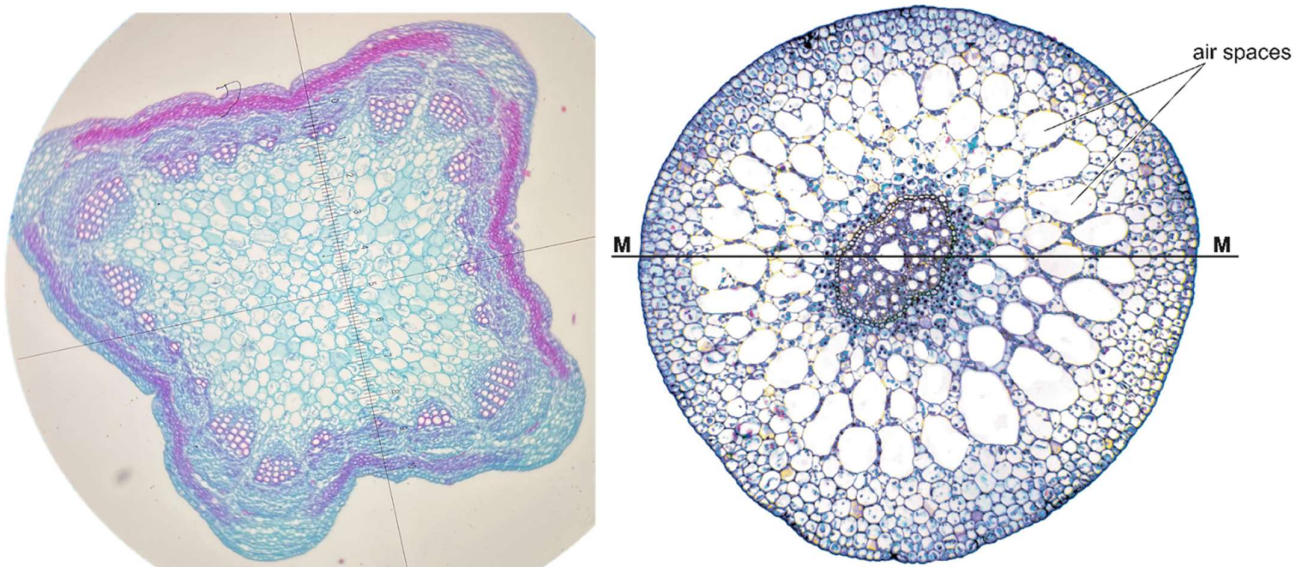
[1]

1. Provide buoyancy to allow stem to be kept upright so that the leaves are nearer to water surface.
2. Stores oxygen for cellular respiration
3. Stores carbon dioxide for photosynthesis
4. Allow rapid transport of gases such as oxygen or carbon dioxide from one part of the plant to another.

- (v) One difference between the stem shown in Fig. 3.2 and the stem on slide **P1** is the presence of air spaces.

Identify **other** observable differences between the stem shown in Fig. 3.2 and the stem on slide **P1**.

Prepare the space below to record **two** of these observable differences.



	Slide P1	Fig. 3.2
Location of vascular tissues	vascular bundles around in a ring near the edge of the stem / near epidermis  Reject: near the sides or corner of the stem	Vascular tissues are in the middle of the stem
Type of cells at the center of the organ	Center of the stem is filled with thin-walled cells / parenchyma	Center of the stem is filled with vascular tissue
Shape of cross-section of stem	Squarish shape	Circular shape
Amount of vascular tissue	More	Less
Pith	Present	Absent
Stele	Absent	Present

[3]

[Total: 21]