TAMPINES MERIDIAN JUNIOR COLLEGE

JC2 PRELIMINARY EXAMINATION

CANDIDATE NAME **Suggested Answers**

CIVICS GROUP

H2 BIOLOGY **9744**

Paper 4 Practical **24 August 2023**
2 hours 30 minutes

Candidates answer on the Question Paper.

READ THESE INSTRUCTIONS FIRST

Do not open this booklet until you are told to do so.
Write your name, civics group and 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 and graphs.
Do not use staples, paper clips, glue or correction fluid/tape.

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

The use of scientific calculator is expected, where appropriate.
You may lose marks if you do not show your working 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.

Shift
Laboratory

For examiner's Use	
1	/ 17
2	/ 16
3	/ 22
Total	**/ 55**

Question 1

You should read through the whole of this question carefully and then plan your use of time to make sure that you finish all that you would like to do.

Yeast cells are able to respire both aerobically and anaerobically using glucose as a substrate.

You are required to investigate the effect of temperature on the rate of respiration of yeast using a respirometer.

A simple respirometer can be made by attaching a length of plastic tubing to a 10 cm^3 syringe, using a short section of rubber tubing. The respirometer may be supported with a retort stand and clamp.

Fig. 1.1 shows a simple respirometer containing a yeast suspension.

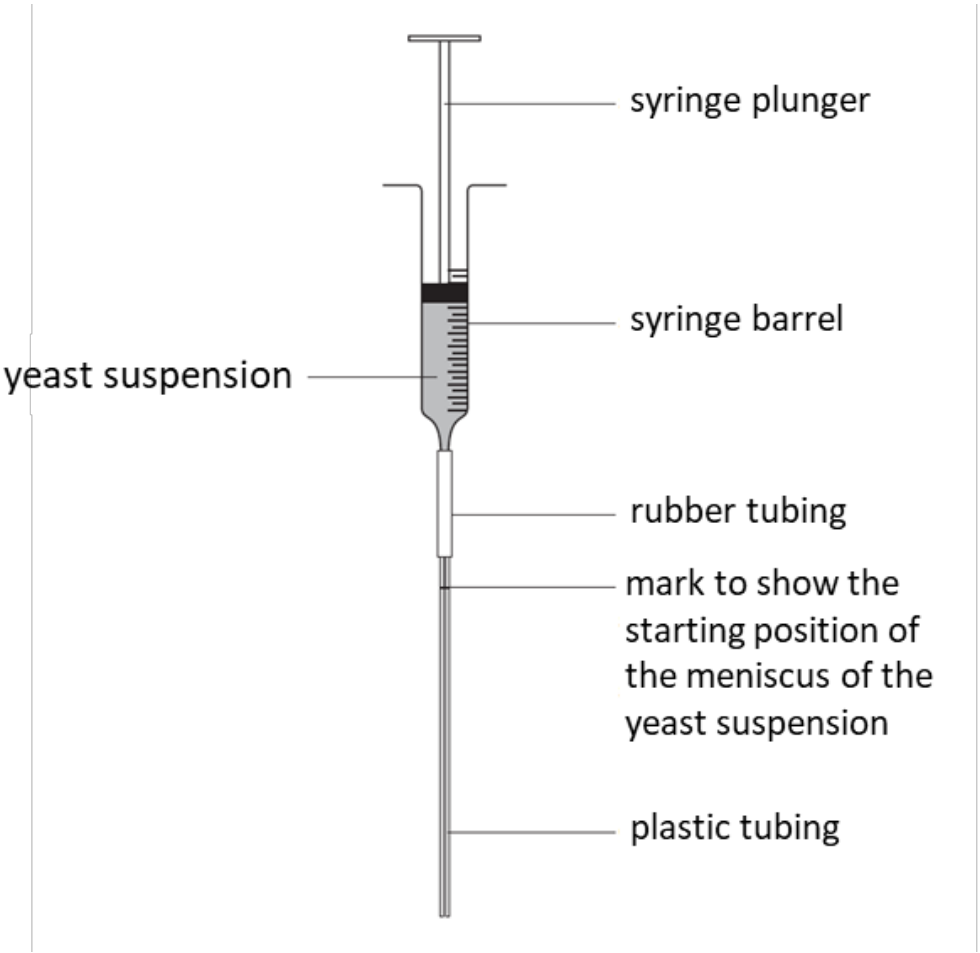


Fig. 1.1

In order to introduce the yeast suspension into the simple respirometers, the following steps are used:

- Remove a sample of the yeast suspension in 20% glucose solution and place it into a boiling tube.
- Make sure that the plunger is pushed fully into the barrel of the syringe.
- Insert the plastic tubing into the yeast suspension in a boiling tube.
- Withdraw the plunger until the yeast suspension reaches the 5 cm^3 mark on the syringe.
- Lift the respirometer so that the glass tubing comes out of the yeast suspension.
- Keep the apparatus vertical. Slowly withdraw the plunger so that the yeast suspension rises up the plastic tubing until the suspension is near to the top of the plastic tubing (See Fig. 1.1).
- Mark the start position of the meniscus of the yeast suspension.

Consider a strategy to allow you to compare the effect of temperature on the rate of respiration of yeast, making use of the apparatus provided.

You will need to decide on a suitable range of temperature for this investigation.

You are provided with:

- simple respirometers
- a suspension of yeast cells in a 20% glucose solution that has been maintained at 35°C for at least 15 minutes before the start of the examination
- water at 30°C
- water at 40°C
- crushed ice
- hot water at 70°C
- a thermometer
- a stopwatch
- a retort stand.

Determine the rate of respiration of yeast by measuring the movement of the meniscus down the plastic tubing.

(a) Describe how you will use the apparatus and materials provided to determine the effect of temperature on the rate of respiration. [10]

Your plan should:

- identify the independent and dependent variables.
- Identify the variables you will need to control.
- include a control.
- have a clear and helpful structure such that the method you use is able to be repeated by anyone reading it, including scientific reasoning behind procedure.
- use the correct technical and scientific terms.
- include reference to safety measures to minimise any risks associated with proposed experiment.

There is no need to describe how to set up respirometer in your methods (It has been described on page 3).

1. **Independent & dependent Variables [compulsory – 1 mark each]**:

   (a) Independent variable: **Temperature / °C**

   (b) Dependent variable: **Distance moved by meniscus in 1 min / mm**
   OR
   Time taken for meniscus to move 100mm /s

2. **Variables to be kept constant and how**: **(any 1)**

Concentration of yeast /% or Volume of yeast /cm^3	Obtain yeast suspension from same stock suspension (e.g.20%). Use a syringe to draw fixed volume(e.g. 20cm^3) of yeast suspension
Diameter of glass tubing /mm	Use the same glass tubing / same batch of glass tubings
Depends on the dependent variable: Duration of experiment /s Or Distance moved by the meniscus /mm	 Use a stopwatch to ensure fixed duration e.g.1 min Use a ruler to ensure fixed distance of *e.g. 100* mm
AVP	

Rationale: These factors affect the distance moved by the meniscus/time take for meniscus to move (depends on dependent variable) and hence must be kept constant to ensure the accuracy of the experiment.

3. **[preliminary test]** Using the yeast suspension given, set up the respirometer according to given instructions, and allow the reaction to proceed and observe for 2 min. The meniscus should move down the glass tubing. This proves that the yeast is respiring. Proceed with the experiment.

4. **Stir the yeast suspension thoroughly** with **a glass rod** [Accept: shake to mix] to ensure a homogenous suspension.

5. Remove 20 cm^3 of yeast suspension using syringe in 20% glucose solution and place it into 5 different boiling tubes respectively.
   [Accept: if student mention other suitable volumes (more than 5cm^3 or pour yeast suspension to certain level, e.g. fill ½ or 2/3 the boiling tube with yeast suspension]

6. Prepare **5** water bath of different temperatures (**10, 20, 30, 40, 50 °C**) using the iced and hot water provided, measured by **thermometer**. [Accept other temperatures of **regular intervals**, between **0 to 60°C**]

7. **Equilibrate** yeast suspension by placing a boiling tube in 10°C **water-bath** [any other temp] for **2 min** [Accept: specified timing that is 1min or more].

8. After 2 min, set up the respirometer with the given instructions (on page 2). **Mark the start position for the meniscus**, using a marker.

9. **[Extra pt]:** Withdraw the plunger slightly, to allow the yeast suspension to be **higher than the meniscus**.

10. Ensure that the respirometer **remains vertical** throughout the experiment.

Either

11. (a) Immediately start time using a **stopwatch** (when the yeast suspension reached the 'start' point) and allow the reaction of proceed for **1 min** [accept 30s to 2min].

    (b) After 1 min, mark the end position of the meniscus and record the **distance moved** by the meniscus (between start and stop mark), using **a ruler**.

OR

12. (a) Use a **ruler** to **measure 20 cm** [Accept: 5cm to 30cm] from start position and mark the end position.

    (b) Start the **stopwatch** and **measure the time taken** for yeast suspension to move 20cm.

13. Repeat steps 2 to 5 using **fresh yeast suspension at 10 °C**, to obtain **3 replicates**, to calculate average, so as to **minimize errors and improve accuracy**. ‘

    Repeat steps 2 to 5 for the other 4 temperatures.

    Repeat the entire experiment one more time to **ensure reproducibility**.

14. **Control [compulsory]**: **Replace yeast suspension with equal volume of distilled water** at 35°C / at every temperature. This is to show that **any movement of the meniscus** is **due to the production of carbon dioxide during respiration of yeast**.

15. **Risk and precautions [compulsory]:**

    - Yeast is an irritant to the skin / eyes. Wear gloves / goggles to protect skin / eyes.
    - Hot water may scald skin. Use mittens, cloth to handle receptacle with hot water.

[Max 10]

(b) Carry out the method you proposed in **(a)** to determine the rate of respiration of yeast at **30°C**. There is no need to carry out replicates for this question.

Present your result and workings clearly. [2]

Distance moved by meniscus in 1 min = ……**d**….. mm

Rate of respiration = d / 1 min = …….. **mm min^{-1}**

……OR……..

Time taken for meniscus to move (e.g) 100 mm = ……**t**…… s

Rate of respiration = **100 mm / t s** = …….. **mm s^{-1}**

- 1m for correct dp and unit of distance moved/ time taken
- 1m for correct working of rate of respiration + correct answer calculated + units (mm min^{-1} / cm min^{-1} / mm s^{-1} / cm s^{-1})

(c) Explain why the meniscus of the yeast suspension moved down the plastic tubing. [3]

- During respiration, **carbon dioxide is produced** …
- during (oxidative) **decarboxylation** in **link reaction** / **Krebs cycle** of aerobic respiration / **alcoholic fermentation** of anaerobic respiration.
- ***Idea that*** The gas produced displaced the yeast suspension in the syringe, pushing / forcing the yeast suspension out via the glass tubing.

(d) Using your theoretical knowledge, sketch a curve on Fig. 1.2 to show how you would expect the rate of respiration of yeast to change as temperature changes.

Label the axes. [2]

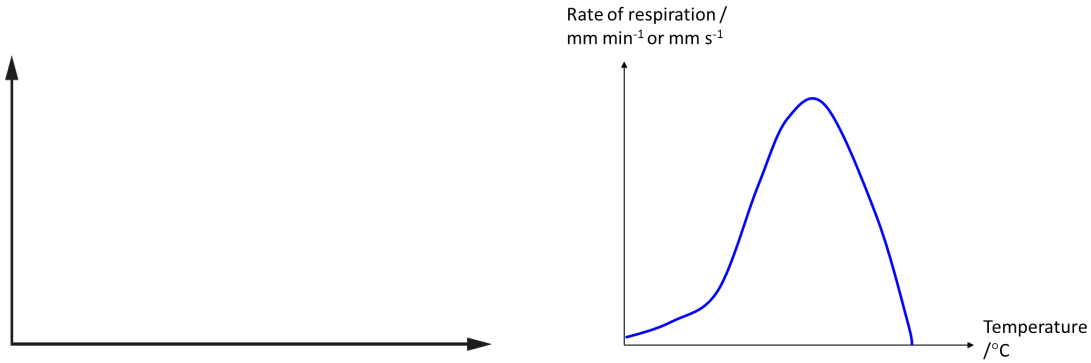


Fig. 1.2

- 1m for correct axes **with units**
- 1m graph of correct shape. (A: if graph did not touch 0)

[Total: 17]

Question 2
Enzyme **E** catalyses the hydrolysis of starch. Starch can be found on the surface of some types of paper. When dipped in iodine, these papers turned dark blue / black.

The end-point of this hydrolysis catalysed by Enzyme **E** is the change in colour of stained paper from blue/black to light blue or white.

You are required to:

- make different concentrations of the enzyme solution, **E,** by simple dilution.
- investigate the effect of different concentrations of **E**, by finding the time taken for the dark blue / black colour of a square paper to change to light blue or white.

You are provided with:

Solutions	Contents	Hazard	Concentration /%	Volume / cm^3
E	*Enzyme solution*	Irritant Harmful	1.0	50
W	Distilled water	None	-	50
0.01% iodine (for Question 2)	Iodine in potassium iodide solution	Irritant Harmful	0.01	-

(a) (i) Decide on the concentrations of **E** you will use in your investigation.

You will need to make up to 10 cm^3 of each concentration of **E**.

Use the space provided below to show:

- the concentrations of **E**
- the volumes of **E**
- the volumes of **W**. [2]

Concentration of **E** /%	Volume of **E** /cm^3	Volume of **W** /cm^3
1.0	10	0
0.8	8	2
0.6	6	4
0.4	4	6
0.2	2	8

- 5 concentrations with **regular** interval, including 1% **E**
- Correct volumes of E and W added to make up to 10 cm^3

You are advised to read steps **1** to **11** before proceeding.

Proceed as follows:

1. Prepare the concentrations of enzyme solution as stated in **(a)(i)** with the beakers provided.

2. Label 5 test tubes and add the concentrations of **E** into the respective test-tubes, to a depth of 2 cm.

3. Using a scalpel, make a single cut, 1 cm long, in the end of a wooden splint as shown in Fig. 2.1.

4. Fit a piece of 1 cm x 1 cm paper into the cut as shown in Fig. 2.2.

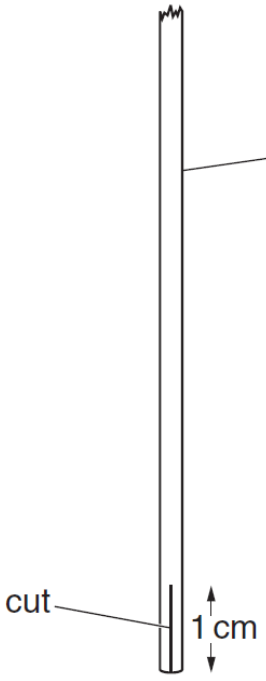


Fig. 2.1

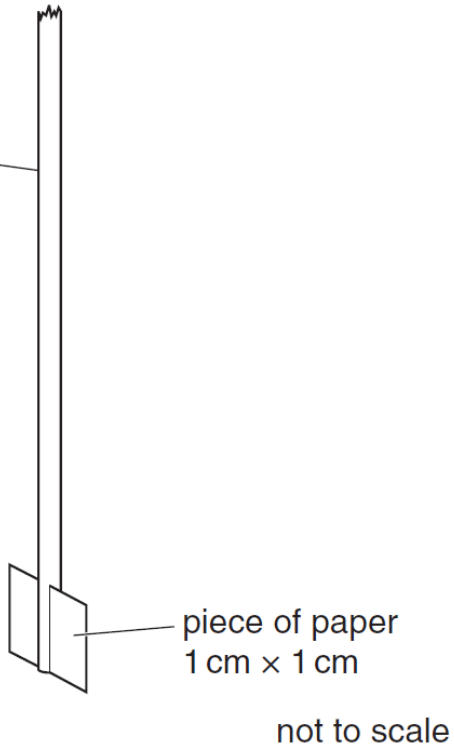


Fig. 2.2

5. Put the piece of paper into the iodine solution, labelled **0.01% iodine (for Question 2)**, as shown in Fig. 2.3, for at least 30 seconds so that the paper is evenly stained.

   This should be used as the colour standard.

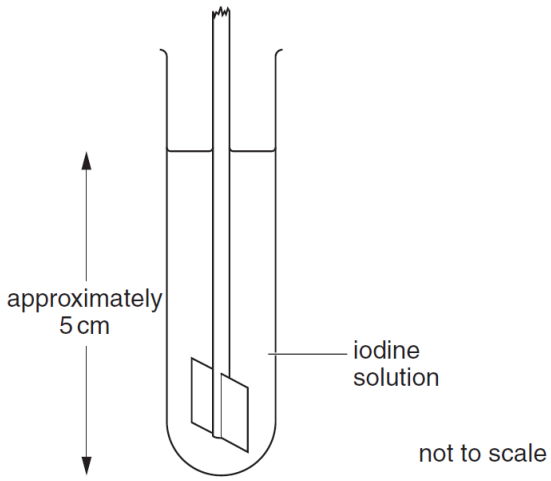


Fig. 2.3

6. Repeat steps **4** to **5** with another wooden splint.

7. Put this splint and its piece of stained paper into the test-tube containing **E**, as shown in Fig. 2.4.

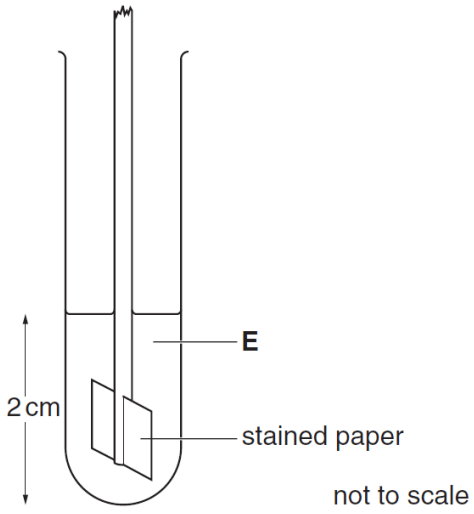


Fig. 2.4

8. Start timing.

9. Occasionally mix the contents by moving the test-tube.

10. Record the time taken for the stained paper to reach the end-point.

    If the end-point is not reached at three minutes, stop timing. Record this as 'more than 180'.

11. Repeat steps **4** to **10** with each of the other concentrations of **E**.

(ii) Record your results in an appropriate format in the space below. [3]

Concentration of **E** /%	Time taken to reach end-point /s
1.0	30 / 37
0.8	38 / 48
0.6	42 / 82
0.4	54 / 85
0.2	62 / 89

- Correct headings with units (mark to be deducted if units is repeated in the cells of table)
- Record time to whole seconds/ 1 d.p. or 'more than 180'
- Correct trend: Highest concentration of E, has the shortest time

(iii) Explain the result that you have obtained. [3]

1. **[compulsory trend & data citation]** As enzyme concentration increases **from 0.2% to 1.0%,** the **time taken to reach end-point decreases from 62s to 30s**.

2. Increasing enzyme concentration, **increases the frequency of effective collision between enzyme/amylase and substrate/starch**.

3. This **increases** the **rate of enzyme-substrate complexes** formed.

4. Hence, **time taken for starch to completely hydrolyse decreases** / **rate of starch hydrolysis increases**, hence time to reach end-point decreases.

(b) You are also provided with an unknown concentration of the enzyme, labelled **U**.

(i) Carry out the same experiment to find out the time taken for the stained paper to reach end-point with **U**.

State the time taken in seconds for the time taken to reach end-point with **U**. [1]

***U = 0.6%**

……**0.4 to 0.8%** ……
[Accept: if no units stated]

Complete Fig. 2.5 by:

- **labelling the position** on the line for **each of the prepared percentage** concentrations of enzyme, **E**. [1]
- putting the label **U** on Fig. 2.5 to show an estimate of the concentration of enzyme in **U**. [1]

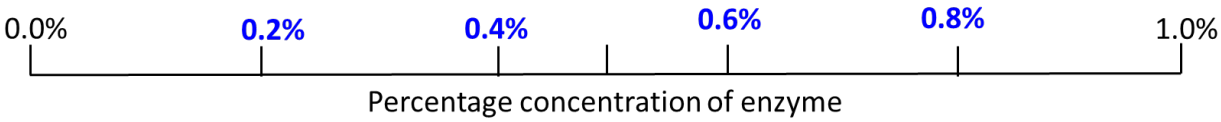


Fig. 2.5

- correct proportionate positions of all prepared concentration [ecf according to concentrations prepared by students] – no mark if students changed concentration here suddenly.
- Accurate estimation of enzyme concentration in **U**, according to student's result.

(c) Fig. 2.6 shows the ruler used by a student in step **7** to measure the depth of **E**.

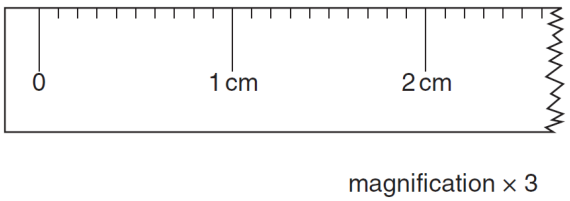


Fig. 2.6

The uncertainty or the precision of an instrument is given as half the smallest division of that instrument.

State the uncertainty of measurement using this ruler. [1]

- **0.5 mm** / **0.05cm**

(d) Identify **two** significant sources of error in your investigation. [2]

- Difficult to determine colour change / end-point of iodine colour.
- The reaction occurs too quickly for accurate measurement of time taken.
- The range of enzyme concentration used (0.2 – 1.0%) is too narrow.
- The 1cm x 1cm papers are not stained equally.
- The time given to stain the 1 cm x 1 cm papers are not kept constant.
- The 1cm x 1cm papers may drop off from the splint / stick to the sides of the test-tube.
- AVP
  [Reject: no control.] → Qn about control was asked earlier.

(e) Suggest how you would make **two** improvements to this investigation. [2]

- Carry out experiment with more enzyme concentration of **wider range** (e.g. 0.2% to 2.0%), to allow trend obtained to be **more reliable**.
- Stain each piece of paper for an equal amount of time, e.g 45 seconds / Stain all papers for an equal amount of time before cutting into pieces.
- Use a wider receptacle (e.g. boiling tube) to reduce the chance of paper sticking to the sides of the wall.
- AVP
  [Reject: have a control.] → Qn about control was asked earlier.

[Total: 16]

Question 3

(a) **Iodine** solution and **methylene blue** solution are used as stains for biological material.

You are required to:

- observe the effect of using different stains, **iodine** solution and **methylene blue** solution, on thin sections of plant material, **S**
- record observations of the cells and their cell contents.

Iodine solution and **methylene blue** solution will stain your skin. Handle the stained plant material with forceps.

If any methylene blue comes into contact with your skin, wash it off immediately with water.

1. Label two microscope slides, **S1** and **S2**.

2. Put **one or two drops** of **iodine** solution onto slide **S1** and **one or two drops** of **methylene blue** solution onto slide **S2**.

3. You are given plant material, **S**. Cut one end of **S** to expose fresh layer of the plant material using a knife, as shown in Fig. 3.1.

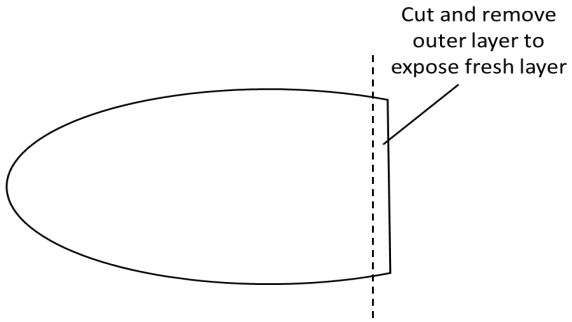


Fig. 3.1

4. Using a scalpel, cut **two very thin** sections of the plant material, **S**, from the newly exposed end.

5. Put each of the thin sections onto the drops of stain on slides **S1** and **S2** respectively, as shown in Fig. 3.2.

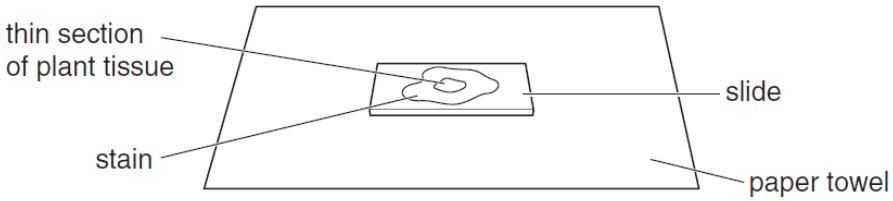


Fig. 3.2

6. Put a few more drops of **iodine** solution onto the plant section on **S1** and put a few more drops of **methylene blue** solution onto the plant section on **S2**. Make sure the **iodine** solution and **methylene blue** solution cover each of the sections.

7. Place a coverslip onto each slide. The coverslip may not lie flat.

8. Use the paper towel to dry off any excess liquid around the coverslip,

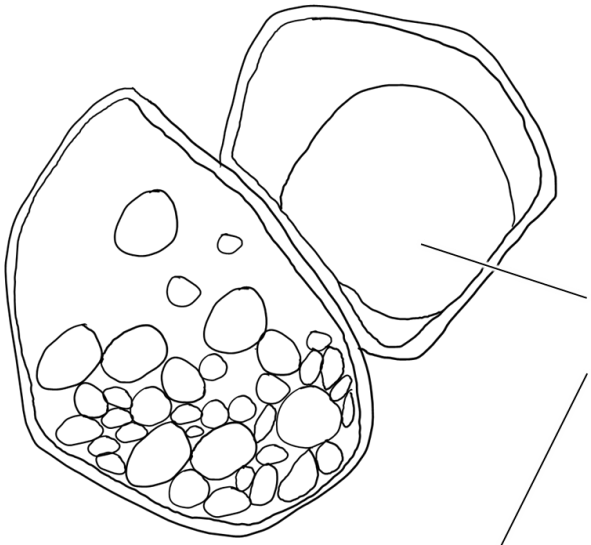
9. **(a)** View the slides using the microscope. Look for the **thinnest** part of the section, which may be at the edge, so that the cells and their contents can be observed.

   (i) From each slide **S1** and **S2**, make large, labelled drawings of two adjacent touching cells and their cell contents.

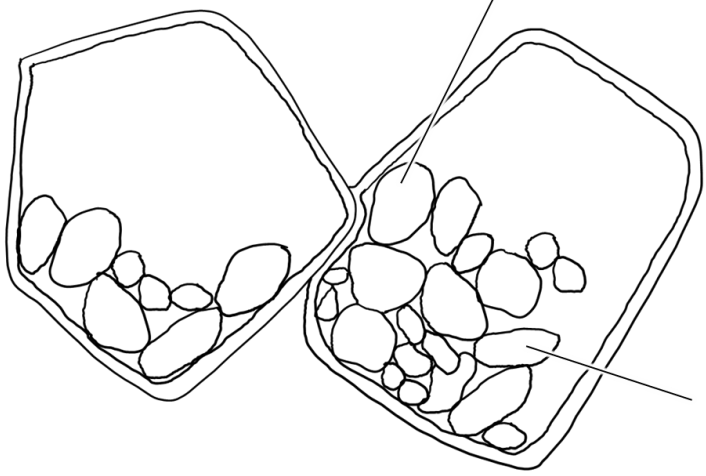
   (ii) On your drawing, label one starch grain.

   (iii) Annotate your drawings to describe one observable difference between **S1** and **S2**.

S1



S2



(ii)

- Starch grains in **S1** is **stained black/blue-black** while starch grains in **S2** is **clear/white/light blue**.
- ***Idea that:*** In S1, starch grains are seen as single mass while in S2, individual starch grains are more distinct.

Marking points:

1. Large drawing (at least ½ of given area) + 2 cells with the correct shape touching each other
2. Quality of drawing → smooth lines (clear, sharp, unbroken lines, no ruled lines, no lines thicker than 1mm, no 'tail') + No Shading
3. Labelled cell structures (Cell wall, cell surface membrane and cytosol/cytoplasm)(No nucleus)
4. At least 3 starch grains drawn in each cell (A: 1 large mass of starch grain in S1) + starch grains are of uneven sizes / drawn as irregular area inside cell.
5. Cell wall drawn as double line (for each cell – no 'sharing' of cell wall) with appropriate proportion
6. Label with a **ruled** label line **one** starch grain only (penalize without using of ruler to label)
7. Describe one observable difference between **S1** and **S2**.
   - Starch grains in **S1** is **stained black/blue-black** while starch grains in **S2** is **clear/white/light blue**.
   - ***Idea that:*** In S1, starch grains are seen as single mass while in S2, individual starch grains are more distinct.
   - In S1, more starch grains could be seen than in S2.

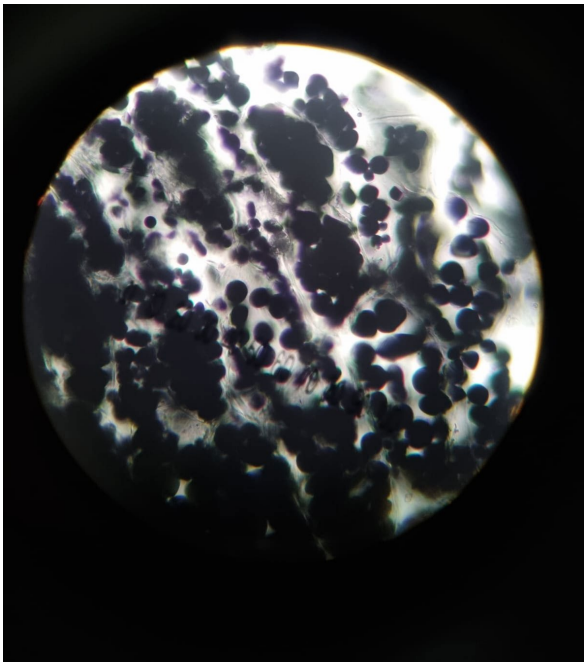
ERRORS:

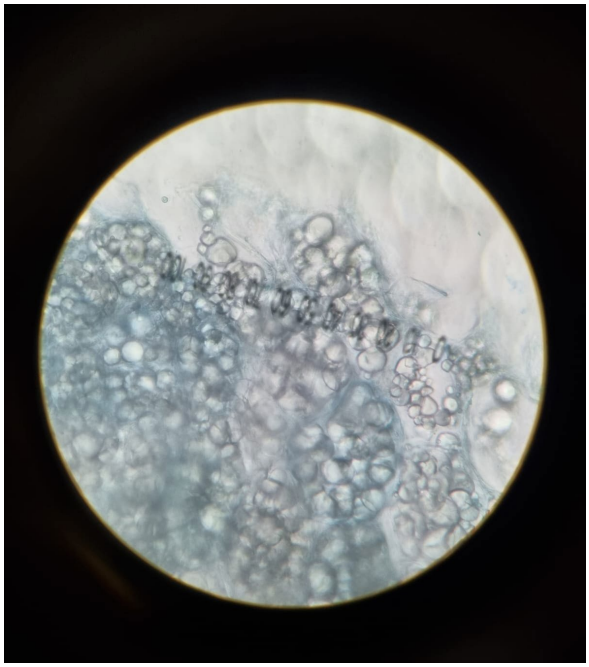
✗ *"S1 cells are larger than S2 (vice versa)" "S1 and S2 cells have different cell shapes"*

✓ **They are the same type of cells obtained from the same sweet potato sample!**

✗ *"S1 cells are stained darker while S2 are stained lighter"*

✓ **VAGUE answer**





(b) **M1** is a slide of a stained transverse section through a plant stem.

(i) Draw a large plan diagram of the region of the stem of **M1** indicated by the boxed region in Fig. 3.3. Use a sharp pencil.

Use a ruled label line and label to identify the epidermis. [5]

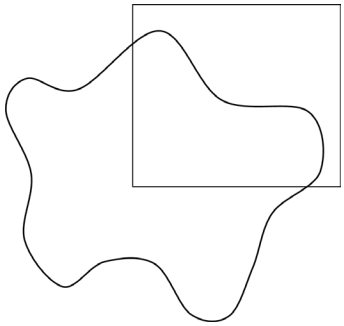
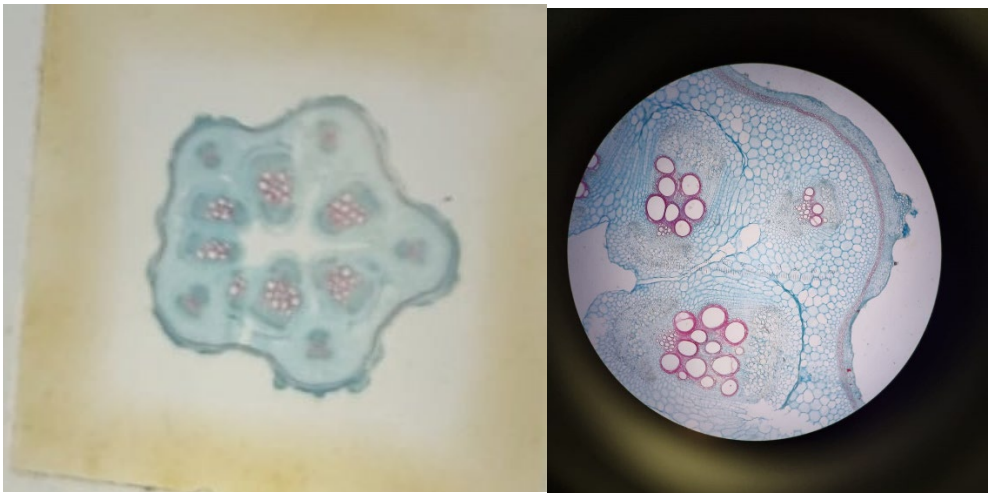
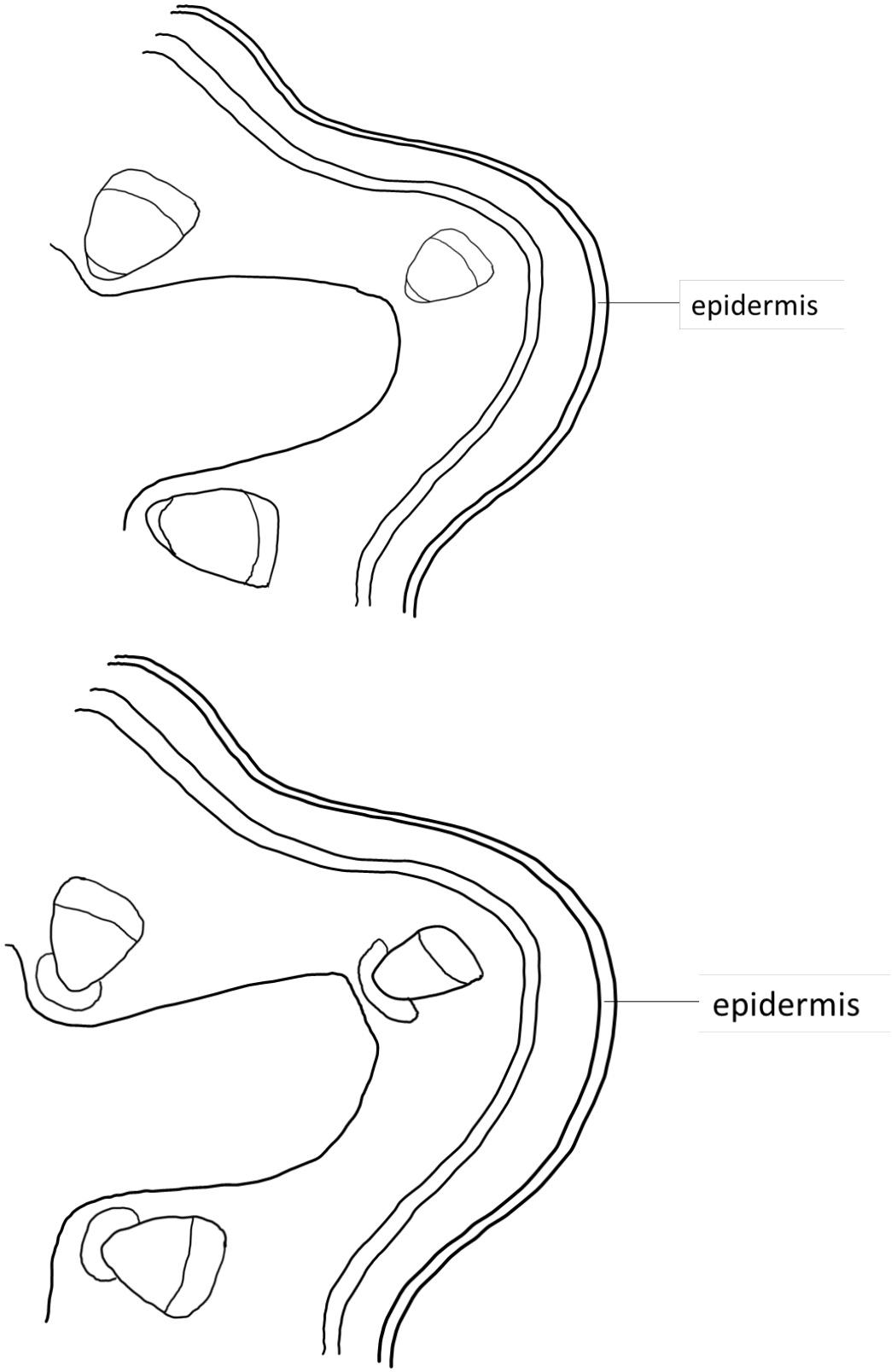


Fig. 3.3

1. Large plan drawing of the correct section (at least ½ of given area) + no cells drawn with smooth lines + no 'closing' of drawing
2. Quality of drawing → smooth lines (clear, sharp, unbroken lines, no ruled lines, no lines thicker than 1mm, no 'tail' or overlapping regions)
3. Drawing of epidermis (with 2 lines) + label with ruled label line
4. 4 other layers (innermost layer is the hollow region)
5. At least 2 vascular bundles drawn + each with at least 3 layers → middle layer (xylem) having the largest proportion (accept 4)




epidermis
epidermis

(iv) Fig. 3.4 shows a photomicrograph of a transverse section of a different stem from **M1**.

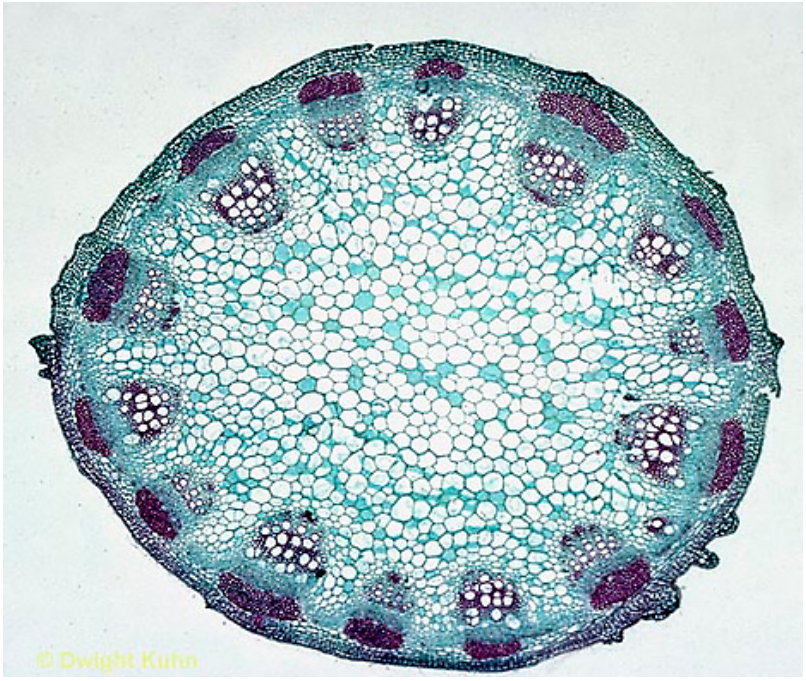


Fig. 3.4

Identify **two** observable differences, other than size and colour, between the stem section on **M1** and the stem section on Fig. 3.4.

Record the observable differences in Table 3.1. [2]

Table 3.1

feature	M1	Fig. 3.4
Shape	Star shape	Circular / oval
Proportion of vascular bundle to leaf section	Larger proportion	Smaller proportion
Number of vascular bundle	Less	More
Central region	Hollow / one large air Space	Comprises of cells / not hollow / no large air space
Arrangement of vascular bundle	Irregular arrangement	Arrange orderly in a circular manner

(c) In an investigation the concentration of sugars in a plant were measured over 24 hours. For the first eight hours, the plant was in the dark, then it was placed in the light for the remaining 16 hours.

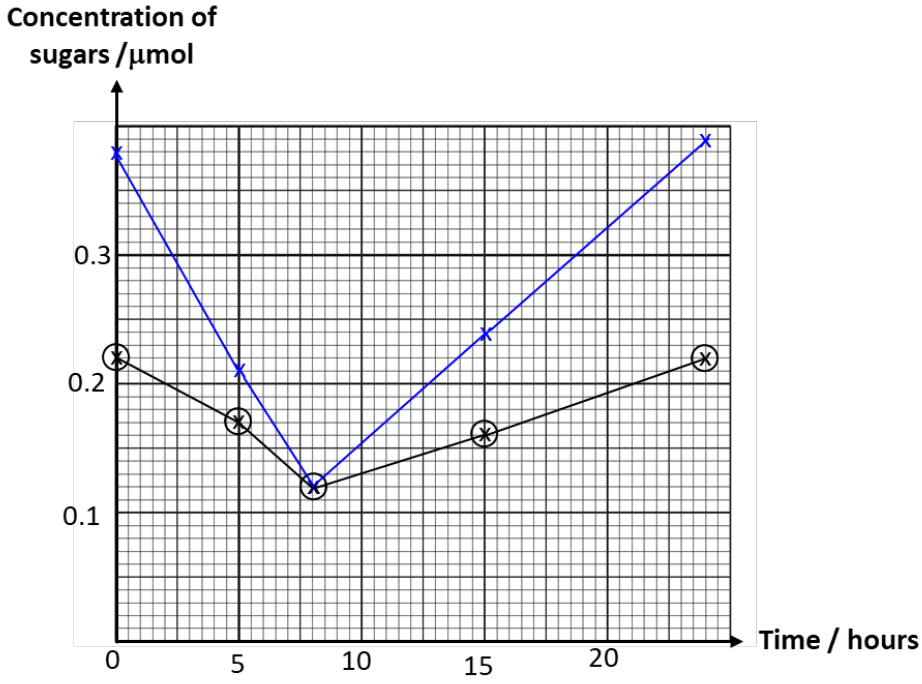
The concentrations of sugars were measured in samples taken from leaves and from phloem sieve tubes. Phloem functions to transport sugar from the leaves to the rest of the plant.

Table 3.2 shows the results of this investigation. [4]

Table 3.2

time / hours	concentration of sugars / mmol	
	in leaves	in phloem sieve tubes
0	0.38	0.22
5	0.21	0.17
8	0.12	0.12
15	0.24	0.16
24	0.39	0.22

(i) Use the grid provided to show the changes in concentration of sugars with time. [4]



- Correct axes with units
- Accurately plotted points for both graphs
- 2 graphs → 2 different symbols used plot + Graph joined point to point
- Appropriate scale + graph occupy at least half of graph area

(ii) Describe the trend for the concentration of sugars **in the leaves**. Suggest an explanation for the trend. [4]

1. **[describe - compulsory]** Concentration of sugars in leaves **decreases from 0.38 to 0.12 μmol** for the **first 8 hours** / **in the dark** then **increases from 0.12 to 0.39 μmol** in the **8 to 24 hours** / **in light**.

2. In the **dark**, **sugars** are **used for respiration**, and no photosynthesis takes place to produce sugars, hence concentration decrease.

3. In **light**, **concentration of sugars produced during photosynthesis** is **higher than** the concentration of sugars **used in respiration**, hence concentration increases.

4. In the presence of light, **light dependent reaction** occurs to produce **NADPH and ATP** which are **used in Calvin cycle / light independent reaction**.

5. In **Calvin cycle**, **glycerate-3-phosphate** is **reduced** by NADPH and ATP to form **glyceraldehyde-3-phosphate**, first sugar.

[must account for decrease and increase]

[Total: 22]