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 candidate 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 or correction fluid.
● Do not write on any bar codes.
● You may use a calculator.
● You should show all your working and use appropriate units.

INFORMATION

● The total mark for this paper is 40.
● The number of marks for each question or part question is shown in brackets [ ].

This document has 16 pages. Blank pages are indicated.
1 The enzyme amylase hydrolyses starch to produce a reducing sugar.

You will investigate the diffusion of reducing sugar from a mixture of starch and amylase through dialysis (Visking) tubing. Dialysis tubing acts as a partially permeable membrane.

You are provided with the materials shown in Table 1.1.

<table>
<thead>
<tr>
<th>labelled</th>
<th>contents</th>
<th>hazard</th>
<th>volume / cm³</th>
</tr>
</thead>
<tbody>
<tr>
<td>E</td>
<td>2.0% amylase solution</td>
<td>irritant</td>
<td>20</td>
</tr>
<tr>
<td>S</td>
<td>1.0% starch solution</td>
<td>none</td>
<td>20</td>
</tr>
<tr>
<td>W</td>
<td>distilled water</td>
<td>none</td>
<td>150</td>
</tr>
<tr>
<td>T</td>
<td>15 cm length of dialysis tubing in distilled water</td>
<td>none</td>
<td>–</td>
</tr>
</tbody>
</table>

If E comes into contact with your skin, wash off immediately under cold water.

It is recommended that you wear suitable eye protection.

Fig. 1.1 shows the apparatus you will set up for this investigation.
Carry out step 1 to step 9.

step 1 Tie a knot in the dialysis tubing as close as possible to one end, so that the end is sealed.

step 2 To open the other end, wet the dialysis tubing and rub the tubing gently between your fingers and thumb.

step 3 Put 5.0 cm$^3$ of $S$ into the dialysis tubing.

step 4 Add 1.0 cm$^3$ of $E$ into the dialysis tubing.

step 5 Rinse the outside of the dialysis tubing by dipping it into the water in the beaker labelled $T$.

Look carefully at Fig. 1.1. This has been set up so that the volume of water covering the dialysis tubing is as small as possible. The part of the dialysis tubing containing the mixture is on the bottom of the beaker.

step 6 Put the dialysis tubing into the beaker labelled $X$. Use a paper clip to hold the dialysis tubing in place, as shown in Fig. 1.1.

step 7 In this step, you will use a syringe to measure the volume of distilled water, $W$, needed to fill beaker $X$ up to the level shown in Fig. 1.1.

Use a syringe to put $W$ into beaker $X$ up to the level shown in Fig. 1.1. Record in (a)(i) the volume of $W$ added.

(a) (i) State the volume of $W$ that you added to beaker $X$ in step 7.

\[
\text{volume of } W = \text{................................................... } \text{cm}^3 \ [1]
\]

Between step 8 and step 9, you will be leaving the apparatus for 20 minutes. Use this time to continue with other parts of Question 1.

step 8 Leave the apparatus for 20 minutes.

step 9 After leaving the apparatus for 20 minutes, remove the dialysis tubing from beaker $X$ and put the dialysis tubing into the container labelled For waste. Do not throw away the solution in beaker $X$. This solution will be needed in step 13.
You will:

- prepare a range of known concentrations of reducing sugar by serial dilution of the 1.0% reducing sugar solution, \( R \)
- carry out the Benedict’s test on the known concentrations of reducing sugar and the solution in beaker \( X \) collected in step 9
- use the results to estimate the concentration of reducing sugar in the solution in beaker \( X \).

You are provided with distilled water, \( W \), and the materials shown in Table 1.2.

**Table 1.2**

<table>
<thead>
<tr>
<th>labelled</th>
<th>contents</th>
<th>hazard</th>
<th>volume / cm(^3)</th>
</tr>
</thead>
<tbody>
<tr>
<td>( R )</td>
<td>1.0% reducing sugar solution</td>
<td>none</td>
<td>25</td>
</tr>
<tr>
<td>Benedict’s</td>
<td>Benedict’s solution</td>
<td>harmful</td>
<td>30</td>
</tr>
</tbody>
</table>

If Benedict’s comes into contact with your skin, wash off immediately under cold water.

It is recommended that you wear suitable eye protection.

You will need to carry out a **serial** dilution of the 1.0% reducing sugar solution, \( R \), to reduce the concentration by **half** between each successive dilution.

You will need to prepare **four** concentrations of reducing sugar in addition to 1.0% reducing sugar solution, \( R \).

After the serial dilution is completed, you will need to have 10 cm\(^3\) of each concentration available to use.

(ii) Complete Fig. 1.2 to show how you will prepare your serial dilution.

Fig. 1.2 shows the first two beakers you will use to make your serial dilution. You will need to draw **three** additional beakers.

For each beaker, add labelled arrows to show:

- the volume of reducing sugar solution transferred.
- the volume of distilled water, \( W \), added.

Under each beaker, state the concentration of reducing sugar solution.
20 cm$^3$ of 1.0% reducing sugar solution, R

10 cm$^3$ of 1.0% reducing sugar solution to use

0 cm$^3$ of W
step 10 Set up a boiling water-bath ready for step 12.

step 11 Prepare the concentrations of reducing sugar solution, as decided in (a)(ii), in the beakers provided.

step 12 Carry out the Benedict’s test on each of the concentrations of reducing sugar solution. Test each solution separately. For each concentration, use 2.0 cm$^3$ of the reducing sugar solution and 2.0 cm$^3$ of Benedict’s solution.

Record in (a)(iii) the time taken for the first appearance of any colour change. If there is no colour change after 120 seconds, record as ‘more than 120’.

(iii) Record your results in an appropriate table.

(iv) Describe one significant source of error when carrying out step 12.

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...........................................................................................................................................
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........................................................................................................................................... [1]
step 13 Carry out the Benedict’s test on the solution in beaker \( X \) from step 9. Use 2.0 cm\(^3\) of the solution in beaker \( X \) and 2.0 cm\(^3\) of Benedict’s solution.

Record in (a)(v) the time taken for the first appearance of any colour change. If there is no colour change after 120 seconds, record as ‘more than 120’.

(v) State the time taken for the first appearance of any colour change in step 13.

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time taken .......................................................... [1]
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(vi) Complete Fig. 1.3 to show:

- the positions of each of the percentage concentrations of reducing sugar solution
- an estimate of the concentration of reducing sugar in the solution in beaker \( X \), using the letter \( X \).

![Diagram of percentage concentration of reducing sugar]  
**Fig. 1.3**  
[2]

(vii) Describe how you could modify this procedure to obtain a more accurate estimate of the concentration of reducing sugar in the solution in beaker \( X \).

Do not include the use of a colorimeter in your answer.

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........................................................................................................................................... [3]
A student repeated step 1 to step 9 at a range of different temperatures. All other variables were kept constant.

The student measured the concentration of reducing sugar in the solution surrounding the dialysis tubing using a different method. The student used a chemical that reacts with reducing sugar to produce a coloured solution. The higher the concentration of reducing sugar, the greater the light absorbance of the solution.

The student used a colorimeter to measure the absorbance of light by the coloured solution.

The results are shown in Table 1.3.

Table 1.3

<table>
<thead>
<tr>
<th>temperature / °C</th>
<th>light absorbance</th>
</tr>
</thead>
<tbody>
<tr>
<td>30</td>
<td>0.900</td>
</tr>
<tr>
<td>41</td>
<td>1.450</td>
</tr>
<tr>
<td>49</td>
<td>1.575</td>
</tr>
<tr>
<td>59</td>
<td>1.100</td>
</tr>
<tr>
<td>70</td>
<td>0.650</td>
</tr>
</tbody>
</table>
(i) Plot a graph of the data shown in Table 1.3 on the grid in Fig. 1.4.

Use a sharp pencil.

Fig. 1.4

(ii) Explain why the light absorbance at 70 °C is less than the light absorbance at 49 °C.

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...........................................................................................................................................
...........................................................................................................................................
........................................................................................................................................... [3]

[Total: 23]
2. J1 is a slide of a stained transverse section through a plant leaf.

(a) (i) Draw a large plan diagram of the region of the leaf on J1 indicated by the shaded area in Fig. 2.1. Use a sharp pencil.

Use one ruled label line and label to identify the vascular bundle.
(ii) Observe the upper epidermis of the leaf on J1. The position of the upper epidermis is shown in Fig. 2.2.

Select a group of four adjacent cells in the upper epidermis.

Each cell must touch at least one other cell.

- Make a large drawing of this group of four cells.
- Use one ruled label line and label to identify the cell wall of one cell.
(b) Fig. 2.3 shows a photomicrograph of a stage micrometer scale that is being used to calibrate an eyepiece graticule.

One division, on either the stage micrometer scale or the eyepiece graticule, is the distance between two adjacent lines.

The length of one division on the stage micrometer in Fig. 2.3 is 0.10 mm.

![Figure 2.3](image)

(i) Calculate the actual length of one eyepiece graticule unit shown in Fig. 2.3.

Give your answer in micrometers (µm).

Show your working.

actual length = ........................................... µm [3]
Fig. 2.4 shows a photomicrograph of a transverse section through a different leaf from **J1**. This was taken with the same microscope and the same lenses used to take the photomicrograph in Fig. 2.3.

Fig. 2.4

(ii) Use the calibration of the eyepiece graticule unit from (b)(i) to calculate the actual length of the plant leaf from **X** to **Y** in Fig. 2.4.

Show your working.
(iii) Fig. 2.5 is a photomicrograph of a transverse section of the same leaf as shown in Fig. 2.4.

**Fig. 2.5**

Identify two observable differences, other than size and colour, between the leaf in Fig. 2.5 and the leaf on J1.

Record these two observable differences in Table 2.1.

**Table 2.1**

<table>
<thead>
<tr>
<th>feature</th>
<th>Fig. 2.5</th>
<th>J1</th>
</tr>
</thead>
</table>

[2]

[Total: 17]