Cambridge International Examinations
Cambridge International Advanced Subsidiary and Advanced Level

CANDIDATE NAME

CENTRE NUMBER  CANDIDATE NUMBER

BIOLOGY  9700/31
Paper 3 Advanced Practical Skills 1
May/June 2018
2 hours

Candidates answer on the Question Paper.
Additional Materials: As listed in the Confidential Instructions.

READ THESE INSTRUCTIONS FIRST

Write your Centre number, candidate number and name on all the work you hand in.
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.
DO NOT WRITE IN ANY BARCODES.

Answer all questions.

Electronic calculators may be used.
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.

For Examiner’s Use

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td></td>
</tr>
</tbody>
</table>

This document consists of 14 printed pages and 2 blank pages.
Before you proceed, read carefully through the whole of Question 1 and Question 2.

Plan the use of the two hours to make sure that you finish all the work that you would like to do.

If you have enough time, think about how you can improve the confidence in your results, for example by obtaining and recording one or more additional measurements.

You will gain marks for recording your results according to the instructions.

1 Milk contains proteins which are used to make cheese. During cheese making, bacteria are added to the milk. The bacteria change the pH of the milk to acidic, causing the proteins to coagulate (clot) forming curds. The curds are then used to make the cheese.

\[
\text{milk + bacteria} \rightarrow \text{proteins in milk coagulate (curds)}
\]

\[
\text{pH more acidic}
\]

Estimating the protein concentration in milk is important when making cheese.

You will need to:

- make simple (proportional) dilutions of the proteins in the milk, \( M \)
- carry out the biuret test on each concentration, to provide a measure of the concentration of proteins present in the milk
- carry out the biuret test on milk with an unknown concentration of protein, \( U \)
- estimate the concentration of milk protein in \( U \).

You are provided with the materials shown in Table 1.1.

**Table 1.1**

<table>
<thead>
<tr>
<th>labelled</th>
<th>contents</th>
<th>hazard</th>
<th>volume/cm³</th>
</tr>
</thead>
<tbody>
<tr>
<td>M</td>
<td>1.0% milk</td>
<td>none</td>
<td>40</td>
</tr>
<tr>
<td>U</td>
<td>milk with an unknown concentration of protein</td>
<td>none</td>
<td>20</td>
</tr>
<tr>
<td>K</td>
<td>5% potassium hydroxide solution</td>
<td>harmful irritant</td>
<td>20</td>
</tr>
<tr>
<td>C</td>
<td>0.15% copper sulfate solution</td>
<td>none</td>
<td>20</td>
</tr>
<tr>
<td>W</td>
<td>distilled water</td>
<td>none</td>
<td>100</td>
</tr>
</tbody>
</table>

It is recommended that you wear suitable eye protection.
If \( K \) comes into contact with your skin, wash it off immediately under cold water.
(a) You are required to make simple (proportional) dilutions of the proteins in the milk, $M$ (1.0%). Reduce the concentration by 0.2% between each successive dilution. You will also make a 0.1% concentration.

You will need to prepare 10 cm$^3$ of each concentration.

(i) Table 1.2 shows how to make up two of the concentrations you will use, 1.0% and 0.1%.

Decide which other concentrations of milk to prepare using simple (proportional) dilutions of $M$.

Complete Table 1.2 to show how you will prepare the other concentrations.

<table>
<thead>
<tr>
<th>volume of $M$ / cm$^3$</th>
<th>volume of distilled water, $W$ / cm$^3$</th>
<th>percentage concentration of milk</th>
</tr>
</thead>
<tbody>
<tr>
<td>10.0</td>
<td>0.0</td>
<td>1.0</td>
</tr>
<tr>
<td>1.0</td>
<td>9.0</td>
<td>0.1</td>
</tr>
</tbody>
</table>

Read step 1 to step 9 before proceeding.

1. Prepare all the concentrations of milk as shown in Table 1.2 in the test-tubes provided.

2. Put the bung into one of the test-tubes and invert it to mix well. Repeat with each of the test-tubes.

3. Label the spotting tile with the concentrations of milk prepared in step 1.

4. Use a pipette to put 2 drops of 1.0% milk into the labelled cavity on the tile.

Any milk remaining in the pipette should be put back into the test-tube so that as little of the milk as possible is removed from the test-tube. You will need this milk for step 13.

5. Repeat step 4 with each of the concentrations of milk.

6. Put 1 or 2 drops of $K$ into each of the concentrations of milk on the tile and mix.

7. Put 0.5 cm$^3$ of $C$ into each mixture on the tile, using the syringe labelled $C$.

8. Leave for 2 minutes for the colour to change.
9. Compare the colour with the standard colours in Fig. 1.1. Record the colour of the mixture in (a)(ii).

<table>
<thead>
<tr>
<th>violet</th>
<th>pale violet</th>
<th>blue</th>
</tr>
</thead>
</table>

Fig. 1.1

(ii) Record your results in an appropriate table for the known concentrations of milk, using only the standard colours shown in Fig. 1.1.

10. Put 2 drops of **U** onto the spotting tile.

11. Repeat step 6 to step 9 with **U**. Record the colour in (a)(iii).

(iii) State the colour for sample **U**.

............................................................[1]
Fig. 1.1 has only three colours. In (a)(ii) more than one of the known concentrations may match U. This means that an estimate of the concentration of milk protein in U may not be accurate.

(iv) Using the result in (a)(iii), state the known concentrations of milk protein where the colour result for U is the same.

.......................................................................................................................................[1]

(v) A significant source of error in this procedure is the difficulty of matching the colour to Fig. 1.1.

Complete Table 1.3 to suggest:
• an improvement to this procedure of matching the colour (dependent variable)
• one other significant source of error in this procedure
• an improvement to reduce this error.

Table 1.3

<table>
<thead>
<tr>
<th>significant source of error</th>
<th>improvement</th>
</tr>
</thead>
<tbody>
<tr>
<td>matching the colour may be inconsistent</td>
<td></td>
</tr>
</tbody>
</table>

another significant source of error

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

(b) A student suggested that another procedure to estimate the concentration of protein in milk could be to compare how much coagulation was produced in acidic conditions.

The student added acid to known concentrations of milk protein and observed a range of coagulation. The concentration of milk protein in an unknown sample could then be estimated more accurately.

You are provided with 1.0 mol dm⁻³ hydrochloric acid, labelled A.

It is recommended that you wear suitable eye protection.
If A comes into contact with your skin, wash it off immediately under cold water.
Read step 12 to step 16 before proceeding.

12. Put 10 cm$^3$ of U into a test-tube.

   In step 13, you will be adding A to the milk in the test-tubes, do not stir or mix A with the milk.

13. Put 2 cm$^3$ of A into each of the test-tubes containing the known concentrations of milk protein (from step 4) and U.

14. Observe the changes in the milk in all of the test-tubes for up to 1 minute.

15. If coagulation is not visible after 1 minute, you may need to carefully tilt each test-tube to move the milk and then observe the coagulation.

16. Using the observations from step 14 and step 15 and the key in Fig. 1.2, record the results in (b)(i).

   key
   +++++++ most coagulation
   ++++++  
   +++     
   ++      
   +       least coagulation

   Fig. 1.2

   (i) Prepare the space below and record the results for the known concentrations of milk protein using Fig. 1.2.
(ii) Using the key in Fig. 1.2, record the result for \( U \).

.........................................................................................[1]

(iii) Complete Fig. 1.3 to show the position on the line of each of the percentage concentrations of milk decided in Table 1.2.

Put the label \( U \) on Fig. 1.3 to show an estimate of the concentration of milk which provides a measure of the proteins in \( U \), using the result in (b)(ii).

0.0% \hspace{1cm} 1.0%

| percentage concentration of milk protein |

Fig. 1.3
(c) Milk proteins can also be coagulated using the enzyme rennet.

A student investigated the effect of temperature on the activity of rennet, shown by the percentage coagulation of the milk.

The results are shown in Table 1.4.

Table 1.4

<table>
<thead>
<tr>
<th>temperature /°C</th>
<th>percentage coagulation of the milk</th>
</tr>
</thead>
<tbody>
<tr>
<td>8.5</td>
<td>7</td>
</tr>
<tr>
<td>28.0</td>
<td>63</td>
</tr>
<tr>
<td>35.5</td>
<td>84</td>
</tr>
<tr>
<td>41.0</td>
<td>92</td>
</tr>
<tr>
<td>50.0</td>
<td>39</td>
</tr>
</tbody>
</table>

(i) Plot a graph of the data in Table 1.4 on the grid in Fig. 1.4.

*Use a sharp pencil for drawing graphs.*
(ii) Suggest explanations for the results between 35 °C and 45 °C.

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...........................................................................................................................................
...........................................................................................................................................[2]

(iii) If a student wanted to investigate the independent variable, pH, a number of experiments would need to be set up.

State the pH values you would select and describe how you would change the pH.

*pH values* .................................................................

*description* .................................................................

...........................................................................................................................................
...........................................................................................................................................
...........................................................................................................................................
...........................................................................................................................................
...........................................................................................................................................
...........................................................................................................................................
...........................................................................................................................................
...........................................................................................................................................
[1]

[Total: 22]
2  **J1** is a slide of a stained transverse section through a plant leaf.

You are not expected to be familiar with this specimen.

**a)** The specimen on **J1** has an irregular shape.

*Use a sharp pencil for drawings.*

**(i)** Draw a large plan diagram of the section of the leaf shown by the shaded area in Fig. 2.1.

*Use one ruled label line and label to identify the vascular bundle.*

---

**Fig. 2.1**

*You are expected to draw the correct shape and proportions of the different tissues.*
(ii) Observe the upper epidermis and palisade layer in the leaf (see Fig. 2.1).

Select one group of two adjacent, touching cells from the upper epidermis and one adjacent, touching cell from the palisade tissue below. Each cell must touch at least one of the other cells.

Make a large drawing of this group of three cells.

Use one ruled label line and label to identify the cell wall of one cell.

(iii) Observe the leaf on J1.

Suggest one observable feature of J1 which supports the conclusion that this plant grows in a dry environment.

Explain how this feature would prevent water loss.

feature ........................................................................................................................................

explanation ................................................................................................................................

................................................................................................................................................

{2}
(b) Fig. 2.2 shows a diagram 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 this stage micrometer is **0.1 mm**.

![Fig. 2.2](image)

(i) Complete Fig. 2.3, by using Fig. 2.2 to calculate the actual length of one eyepiece graticule unit.

Step 1

1 eyepiece graticule unit = __________ divided by __________ = __________ mm

Step 2

Convert the answer to a measurement with the unit most suitable for use in light microscopy.

_________ multiplied by __________ = __________

![Fig. 2.3](image)
Fig. 2.4 shows a photomicrograph of a transverse section through a leaf, taken using the same microscope with the same lenses as Fig. 2.2.

You are not expected to be familiar with this specimen.

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

Show all the steps in your calculation and use appropriate units.

............................................................ [2]
Fig. 2.5 is a photomicrograph of a transverse section of the same leaf as in Fig. 2.4.

Annotate Fig. 2.5 to describe two observable differences between the leaf in Fig. 2.5 and the leaf on J1:

- Draw label lines to two different features and use only the labels P and Q.
- Next to each letter, describe how each feature on the leaf in Fig. 2.5 differs from the leaf on J1.

Fig. 2.5