

**BIOLOGY**

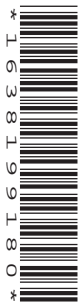
**9700/33**

Advanced Practical Skills 1

**May/June 2014**

**CONFIDENTIAL INSTRUCTIONS**

**Great care should be taken to ensure that any confidential information given, including the identity of material on microscope slides where appropriate, does not reach the candidates either directly or indirectly.**



If you have any problems or queries regarding these Instructions, please contact CIE  
by e-mail: info@cie.org.uk  
by phone: +44 1223 553554  
by fax: +44 1223 553558  
stating the Centre number, the nature of the query and the syllabus number quoted above.

This document consists of **11** printed pages and **1** blank page.

### Instructions for preparing apparatus

These Instructions give details of the apparatus required by each candidate for each experiment in this paper. A summary of the questions that will be presented to the candidates is included, where appropriate, to allow the Biology teacher to test the apparatus appropriately.

**No access to the question paper is permitted in advance of the examination.**

If a candidate breaks any of the apparatus, or loses any of the material supplied, the matter should be rectified and a note made in the Supervisor's Report.

Candidates must be provided with a microscope with:

- Eyepiece lens,  $\times 10$  (equal to 16 mm or  $\frac{2}{3}$ " )
- Low-power objective lens,  $\times 10$  (equal to 16 mm or  $\frac{2}{3}$ " )
- High-power objective lens,  $\times 40$  (equal to 4 mm or  $\frac{1}{6}$ " )
- Eyepiece graticule fitted within the eyepiece and visible in focus at the same time as the specimen.

To avoid confusion, Cambridge request that only the lenses specified above are fitted in the microscopes to be used in the examination. Any lenses which are **not**  $\times 10$  or  $\times 40$  should be removed or replaced.

Each candidate must have sole, uninterrupted, use of the microscope for at least 55 minutes.

Supervisors are advised to remind candidates that **all** substances in the examination should be treated with caution. Pipette fillers and safety goggles should be used where necessary.

In accordance with the COSHH (Control of Substances Hazardous to Health) Regulations, operative in the UK, a hazard appraisal of the examination has been carried out.

The following codes are used where relevant.

**C** = corrosive substance

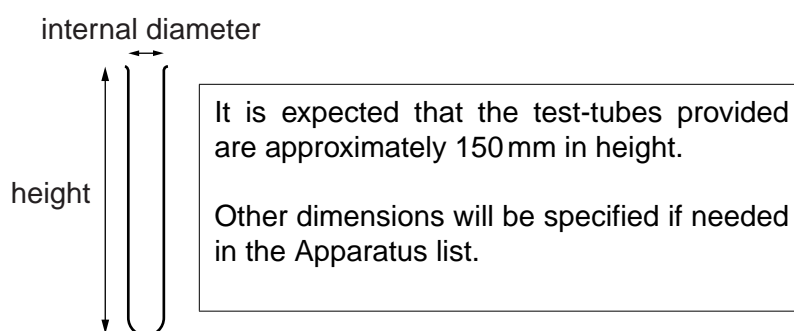
**H** = harmful or irritating substance

**T** = toxic substance

**F** = highly flammable substance

**O** = oxidising substance

**N** = harmful to environment



Centres are reminded that they are **not** permitted to open the question paper envelopes before the examination. Centres should also refer to the Handbook for Centres.

If there are any difficulties with any aspect of setting up this practical examination that the Centre is not able to resolve, it is essential for Centres to contact the Product Manager as soon as possible by **e-mail** to [info@cie.org.uk](mailto:info@cie.org.uk), by **fax** to +44 1223 553558 or by **phone** to +44 1223 553554.

## Confidential Instructions

Each candidate will require:

### For both questions

- mm ruler.
- 7.0% yeast cell suspension (see below).

### Question 1

- Solutions and reagents provided to the candidates should be supplied in a suitable beaker, or container, for removal of the solution using a syringe. More of the solutions and reagents should be available if requested by candidates.
- Clean test-tubes and syringes are needed for each candidate.
- Fresh **Y**, **C** and **H** are needed for each candidate.
- All solutions and reagents should be disposed of according to local safety regulations.

Summary of solutions and reagents:

| labelled | contents  | hazard          | volume /cm <sup>3</sup> |
|----------|---|-----------------|-------------------------|
| <b>Y</b> | 7.0% yeast cell suspension  | none            | at least 50             |
| <b>C</b> | 0.01 mol dm <sup>-3</sup> calcium hydroxide coloured blue with bromothymol blue indicator | [H]<br>irritant | at least 70             |
| <b>H</b> | 0.05 mol dm <sup>-3</sup> hydrochloric acid   | [H]<br>irritant | at least 30             |

**It is advisable to wear safety glasses/goggles when handling chemicals.**

Preparation of solutions and reagents:

- (i) **Y**, at least 50 cm<sup>3</sup> of a 7.0% yeast cell suspension with glucose added **10 to 15** minutes before the candidates start Question 1 (also the starting yeast cell suspension for Question 2). As the yeast cell suspension will froth, it should be prepared in a large container.

7.0g of dried yeast (**for baking**) is added to 80 cm<sup>3</sup> of warm distilled water, stirred and made up to 100 cm<sup>3</sup> with warm distilled water. This should be kept at a temperature between 35 °C and 40 °C.

This is sufficient for 2 candidates.

**10 to 15** minutes before the candidates start Question 1 add the glucose.

Sprinkle 20 g of glucose, a little at a time, onto the surface of the yeast cell suspension, stirring continuously. Keep warm between 35 °C and 40 °C until needed.

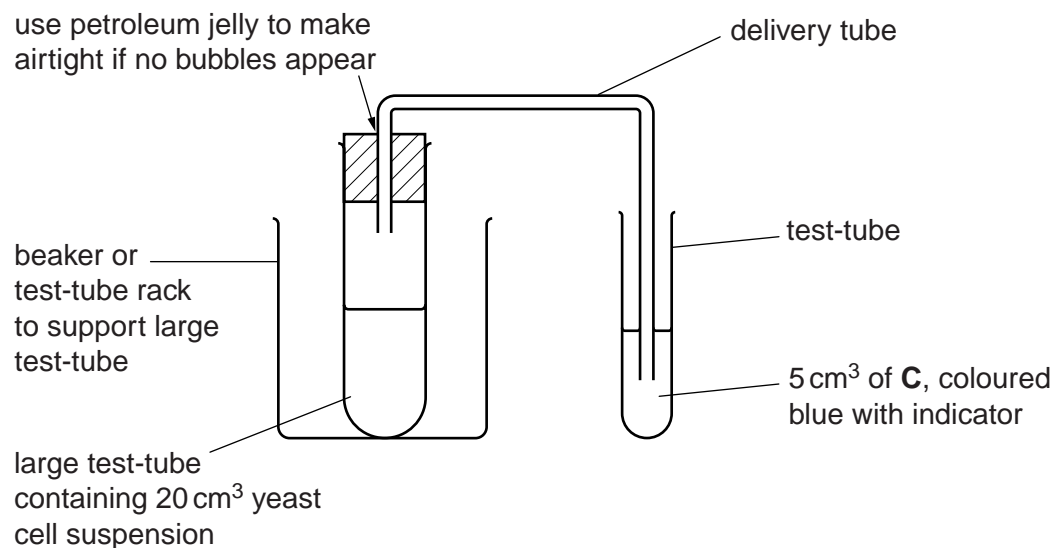
When ready to put into beakers for each candidate, it is suggested that the yeast cell suspension is decanted into a new beaker or container leaving the froth behind.

50 cm<sup>3</sup> of the yeast cell suspension should be given to each candidate in a beaker or container, labelled **Y**. The beaker or container needs to be large enough to hold at least 250 cm<sup>3</sup> since the yeast will froth.

It is **essential** to try the yeast well **before** the examination to make sure that it will become active. (Do **not** use Brewer's yeast as this does not always work actively enough in the time).

**To test the activity of the yeast cell suspension**, set up the apparatus as in Fig. 1.1.

**C** should remain blue after three minutes. If the blue colour is removed then dilute the yeast cell suspension and repeat.



**Fig. 1.1**

**[H] (ii) C**, at least 70 cm<sup>3</sup> of 0.01 mol dm<sup>-3</sup> calcium hydroxide, in a beaker or container, labelled **C**.

This should be coloured blue by putting approximately 5 cm<sup>3</sup> of bromothymol blue indicator into the 70 cm<sup>3</sup> of **C**.

**C** is prepared by dissolving 0.74 g of calcium hydroxide in 500 cm<sup>3</sup> of distilled water, mixing well then making up to 1000 cm<sup>3</sup> with distilled water. This makes a solution which may remain cloudy. (If you do not have a balance which measures to 0.01 g then 0.7 g of calcium hydroxide is acceptable.)

This is sufficient for 14 candidates.

**To prepare bromothymol blue indicator:**

- Put 0.1 g of bromothymol blue into a beaker or container.
- Put 16 cm<sup>3</sup> of 0.01 mol dm<sup>-3</sup> sodium hydroxide solution into the same beaker and mix well to dissolve the bromothymol blue.
- Make up to 250 cm<sup>3</sup> with distilled water.

**To prepare 0.01 mol dm<sup>-3</sup> sodium hydroxide solution:**

- Using forceps, put 4 g of sodium hydroxide into a beaker or container with 80 cm<sup>3</sup> of distilled water.
- Mix well to dissolve.
- Make up to 100 cm<sup>3</sup> with distilled water. This is the stock solution of 1 mol dm<sup>-3</sup> sodium hydroxide solution.
- Put 1 cm<sup>3</sup> of this stock solution into a beaker or container and make up to 100 cm<sup>3</sup> with distilled water.

**[H] (iii) H**, at least 30 cm<sup>3</sup> of 0.05 mol dm<sup>-3</sup> hydrochloric acid in a beaker or container, labelled **H**.

This is sufficient for 1 candidate.

(Safety: Acid **must** be added to water. Water must **not** be added to acid.)

To dilute 1.00 mol dm<sup>-3</sup> hydrochloric acid, add 5 cm<sup>3</sup> of the acid to 95 cm<sup>3</sup> of distilled water in a beaker or container.

Apparatus for each group of candidates should be clean.

Syringe needles are **not** required and must **not** be given to candidates.

| Apparatus for each candidate   | Quantity | ✓ |
|--|----------|---|
| 10 cm <sup>3</sup> syringe with the means to wash it out   | 2        |   |
| 2 cm <sup>3</sup> or 3 cm <sup>3</sup> syringe with the means to wash it out   | 2        |   |
| Glass rod  | 1        |   |
| Beaker or container (approximately 400 cm <sup>3</sup> ) with tap water, labelled <b>For washing</b>   | 1        |   |
| Beaker or container, labelled <b>For waste</b>   | 1        |   |
| Paper towels   | 8        |   |
| Test-tubes – to hold approximately 25 cm <sup>3</sup>  | 6        |   |
| Test-tubes – with an internal diameter between approximately 22 mm and 24 mm and to hold more than 40 cm <sup>3</sup> and less than 50 cm <sup>3</sup> | 1        |   |
| Bung (to give an airtight fit with the larger test-tube) with delivery tube to go into the smaller test-tubes as shown in Fig. 1.1                     | 1        |   |
| Test-tube rack(s) or container(s) to hold six small test-tubes and one large test-tube   | 1        |   |
| Petroleum jelly to be available on request   |          |   |
| Stop-clock or timer showing seconds  | 1        |   |
| Glass marker pen   | 1        |   |
| Safety goggles/glasses   | 1        |   |

During the examination, the Supervisor (**not** the Invigilator) should, **out of the sight of the candidates**, carry out **Question 1** using the same solutions and reagents as the candidates. The results for **1(a)(iv)** should be written in the Supervisor's Report (on pages 11 and 12), not on a spare question paper.

The Supervisor's Report and the candidates' seating plan should be enclosed with the candidates' scripts.

Please ensure that if the scripts are in several packets a copy of the Supervisor's Report and the candidates' seating plan are enclosed with each packet of scripts.

The Invigilator should **not** carry out **Question 1**.

**Question 2**

- Yeast cell suspensions provided to the candidates should be supplied in a container, suitable for the removal of a drop of suspension using a glass rod or teat pipette.

Prepare a 7.0% yeast cell suspension as described for Question 1. **The glucose should be added to the yeast 10 to 15 minutes before the candidates start Question 2. Each candidate should have fresh yeast cell suspension.**

Put 50 cm<sup>3</sup> of 7.0% yeast suspension into a beaker or container and make up to 100 cm<sup>3</sup> with warm distilled water. The yeast needs to be actively frothing.

This makes the 3.5% yeast suspension needed for the samples **S2** and **S3**.

**S1 could be prepared the day before and stored in a refrigerator.**

Summary of solutions and reagents:

| labelled  | contents                                 | hazard         | volume /cm <sup>3</sup> |
|-----------|--|----------------|-------------------------|
| <b>S1</b> | 3.5% <b>boiled</b> yeast cell suspension | none           | at least 5              |
| <b>S2</b> | 3.5% active yeast cell suspension        | none           | at least 5              |
| <b>S3</b> | mixture of <b>S1</b> and <b>S2</b>       | none           | at least 5              |
| <b>M</b>  | 1% methylene blue solution               | [H]<br>harmful | at least 5              |

**It is advisable to wear safety glasses/goggles when handling chemicals.**

Each candidate will require:

- (i) **S1**, at least 5 cm<sup>3</sup> of 3.5% **boiled** yeast cell suspension in a small container, labelled **S1**. **For example:** put 20 cm<sup>3</sup> of 3.5% yeast cell suspension into a container and put this into a boiling water-bath for 10 minutes.

To check that **all** the yeast cells are dead:

- place a drop of suspension onto a microscope slide
- add 1 drop of methylene blue as made up below
- mix with a glass rod and leave for 5 minutes
- add a coverslip and observe using the high-power objective lens of the microscope. Nearly all the cells should be blue. If this is not the case then boil for an extra 5 minutes. Repeat if necessary.

**S1 could be prepared the day before and stored in a refrigerator.**

This is sufficient for 4 candidates.

- (ii) **S2**, at least 5 cm<sup>3</sup> of 3.5% yeast cell suspension (active) as diluted above from active 7.0% yeast suspension.
- (iii) **S3**, at least 5 cm<sup>3</sup> of 3.5% yeast cell suspension made up of equal volumes of **S1** and **S2**.  
**For example**, put 10 cm<sup>3</sup> of **S1** into a small container. Add 10 cm<sup>3</sup> of **S2** to the small container and mix well.

This is sufficient for 4 candidates.

- [H] (iv) **M**, at least 5 cm<sup>3</sup> of freshly prepared 1% methylene blue solution in a container with a pipette, labelled **M**.

This is prepared by dissolving 1.0 g of methylene blue and 0.6 g of sodium chloride in 80 cm<sup>3</sup> of distilled water and making it up to 100 cm<sup>3</sup> with distilled water.

This is sufficient for 20 candidates.

(Safety: Be careful not to inhale the powder. If methylene blue comes into contact with your skin, rinse with cold water.)

Apparatus for each group of candidates should be clean.

| Apparatus for each candidate   | Quantity | ✓ |
|--|----------|---|
| Microscope slides and coverslips   | 3        |   |
| Pipette, teat to remove samples from containers  | 2        |   |
| Glass rod  | 1        |   |
| Mounted needle or seeker   | 1        |   |
| Beaker or container, (about 200 cm <sup>3</sup> ) with tap water, labelled <b>for washing</b>  | 1        |   |
| Beakers or container, labelled <b>for waste</b>  | 1        |   |
| Glass marker pen   | 1        |   |
| Paper towel  | 6        |   |
| Stop-clock or stopwatch or sight of a clock to time five minutes   | 1        |   |
| Microscope with: <ul style="list-style-type: none"> <li>• Low-power objective lens, ×10 (equal to 16 mm or <math>\frac{2}{3}</math>" )</li> <li>• High-power objective lens, ×40 (equal to 4 mm or <math>\frac{1}{6}</math>" )</li> <li>• Eyepiece lens, ×10 (equal to 16 mm or <math>\frac{2}{3}</math>" )</li> <li>• Eyepiece graticule fitted within the eyepiece and visible in focus at the same time as the specimen.</li> </ul> | 1        |   |

To avoid confusion, Cambridge request that only the lenses specified above are fitted in the microscopes to be used in the examination. Any lenses which are **not** ×10 or ×40 should be removed or replaced.

Each candidate must have sole, uninterrupted use of the microscope for 55 minutes.



**MATERIALS TO BE SUPPLIED BY CAMBRIDGE**

- (i) Question papers.

**RETURN OF EXAMINATION MATERIALS TO CAMBRIDGE**

There are **no** materials to return to Cambridge, only question papers, Supervisor's Report forms and seating plan.

**SUPERVISOR'S REPORT and SEATING PLAN**

The Teacher responsible for the examination is asked to fill in the Supervisor's Report in these Confidential Instructions. For Centres where more than one script packet is used, there must be a copy of the completed Supervisor's Report and the candidates' seating plan in each script packet.

These Supervisors' Reports are essential in order to allow the Examiners to assess all candidates as fairly as possible and should always be completed by every Centre.

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**This form should be completed and sent to the Examiner with the scripts.**

**SUPERVISOR'S REPORT ON PRACTICAL BIOLOGY A LEVEL**

**May/June 2014**

*The Supervisor or Teacher responsible for the subject should provide the following information.*

1. Was any difficulty experienced in providing the necessary materials? If so, give brief details.
  
2. Give details of any difficulties experienced by particular candidates, giving names and candidate numbers. Reference should be made to:
  - (a) difficulties arising from faulty specimens or microscopes;
  - (b) accidents to apparatus or materials;
  - (c) assistance provided in case of colour-blindness;
  - (d) any other information that is likely to assist the Examiner, especially if this cannot be discovered from the scripts.

All other cases of individual hardship, e.g. illness or disability, should be reported direct to CIE on the normal 'Special Consideration Form' as detailed in the Handbook for Centres.

3. During the examination, the Supervisor should, **out of sight of the candidates**, carry out **Question 1** using the same solutions and reagents as the candidates. The results for **1(a)(iv)** should be written in the Supervisor's Report which should be enclosed with the candidates' scripts. If the scripts are in several packets, please ensure that a copy of the Supervisor's Report is enclosed with each packet of scripts. The Invigilator should **not** carry out **Question 1**.



Results for **Question 1(a)(iv)**

Temperature of examination room .....°C

- 4. Enclose a seating plan of work benches with the scripts, giving details of the candidate numbers of the places occupied by the candidates for each session. Use separate paper for this.

**Declaration** (to be signed by the Principal or the Examinations Officer)

The preparation of this practical examination has been carried out so as to maintain fully the security of the examination.

Signed .....

Name (in block capitals) .....

Centre number (of enclosed scripts) .....

Centre name .....

If scripts are required by CIE to be despatched in more than one envelope, it is essential that a copy of the relevant Supervisor’s Report and the appropriate seating plan(s) are sent inside **each envelope**.

