

**ADVANCED GCE****BIOLOGY**

Microbiology and Biotechnology

2805/04

Candidates answer on the question paper

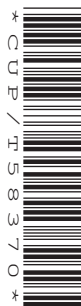
OCR Supplied Materials:

- Insert (inserted)

Other Materials Required:

- Electronic calculator
- Ruler (cm/mm)

Monday 26 January 2009
Morning

Duration: 1 hour 30 minutes

Candidate Forename		Candidate Surname	
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Centre Number						Candidate Number				
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INSTRUCTIONS TO CANDIDATES

- Write your name clearly in capital letters, your Centre Number and Candidate Number in the boxes above.
- Use black ink. Pencil may be used for graphs and diagrams only.
- Read each question carefully and make sure that you know what you have to do before starting your answer.
- Answer **all** the questions.
- Do **not** write in the bar codes.
- Write your answer to each question in the space provided, however additional paper may be used if necessary.

INFORMATION FOR CANDIDATES

- The number of marks is given in brackets [] at the end of each question or part question.
- The total number of marks for this paper is **90**.
- You will be awarded marks for the quality of written communication where this is indicated in the question.
- You may use an electronic calculator.
- You are advised to show all the steps in any calculations.
- This document consists of **20** pages. Any blank pages are indicated.

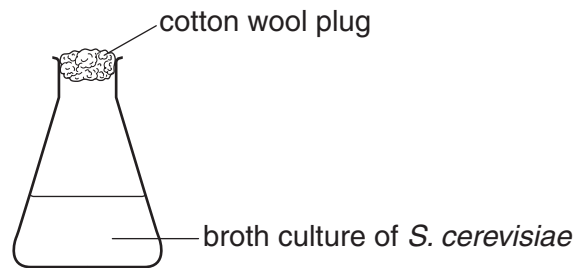
FOR EXAMINER'S USE		
Qu.	Max.	Mark
1	19	
2	18	
3	20	
4	10	
5	9	
6	14	
TOTAL	90	

Answer **all** the questions.

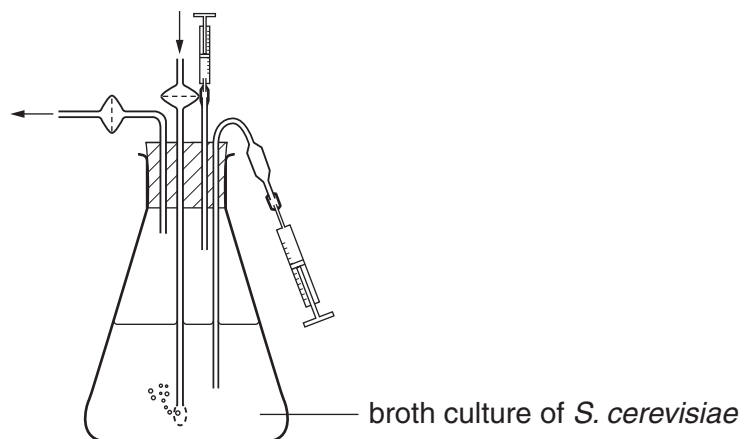
- 1 (a) *Saccharomyces cerevisiae* is used in beer production. A student investigated population growth in *S. cerevisiae* under **aerobic** conditions. The investigation involved a scaling-up procedure.

Fig. 1.1 shows the three stages used to scale-up from a flask containing a broth culture of *S. cerevisiae* to a 10 dm³ bench fermenter.

stage 1: flask



stage 2: bioreactor



stage 3: bench fermenter

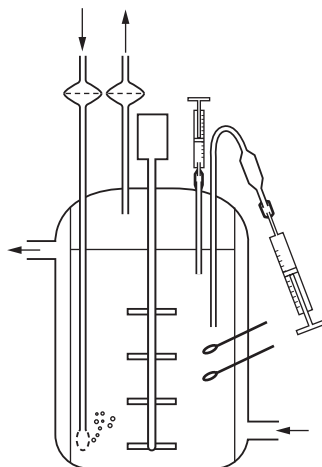


Fig. 1.1

- (i) The student was supplied with a sterile boiling tube containing a stock broth culture of a pure strain of *S. cerevisiae*. The tube was sealed with a cotton wool plug.

Describe how the student inoculated the flask shown in **stage 1** of Fig. 1.1.

Include in your answer an explanation for the aseptic technique used.

..... [4]

- (ii) For each of **stages 2** and **3**:
- describe **one** addition or modification that has been made to the apparatus **from the previous stage** and
 - explain why the addition or modification has been made.

stage 2 addition or modification

.....

explanation

.....

.....

stage 3 addition or modification

.....

explanation

.....

.....

..... [4]

- (iii) In **stage 3**, the student removed samples of the culture medium at timed intervals in order to obtain a count of yeast cells using a haemocytometer.

Table 1.1 lists the steps in the procedure for using a haemocytometer.

State **one** reason for carrying out each of the steps. The first has been completed for you.

Table 1.1

step in procedure	reason for carrying out the step
carry out a ten-fold serial dilution (decimal dilution) to obtain a 10^{-3} dilution	<i>dilutes the sample otherwise there will be too many yeast cells to count accurately.</i>
agitate the test-tube containing the 10^{-3} dilution before removing a sample
wait five minutes after adding the sample to the haemocytometer before counting the number of yeast cells
count cells in a triple-lined square and include cells touching the middle line of the north and west sides only
count cells in a number of triple-lined squares

[4]

- (b) Describe what happens in the **fermentation** stage of beer production.

Details of milling, mashing and boiling are **not** required.

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..... [5]

- (c) Beer production is carried out using batch culture in the fermentation stage.

A number of commercial brewers have been investigating the use of continuous culture.

Explain what is meant by the term *continuous culture*.

.....

.....

.....

.....

.....

..... [2]

[Total: 19]

2 Fig. 2.1, **on the insert**, shows diagrams of five different microorganisms.

- (a) Complete Table 2.1 by naming **and** stating **one** function for each of the structures labelled **A** to **E** on Fig. 2.1.

Table 2.1

	name of structure	function of structure
A		
B		
C		
D		
E		

[5]

- (b) Using the information provided on Fig. 2.1, complete Table 2.2 below.

Table 2.2

size	name of organism
largest organism	
smallest organism	

[3]

- (c) (i) With reference to Fig. 2.1, state the kingdom to which *Chlamydomonas* belongs.

..... [1]

- (ii) Suggest why *Chlamydomonas* has been classified into the kingdom stated in 2(c)(i).

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

- (d) Describe the nature and organisation of the genetic material inside,

- (i) bacterial cells, such as *Escherichia coli*;

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

- (ii) viruses, such as bacteriophage lambda (λ phage);

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.....
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.....

- (iii) fungi, such as *Saccharomyces cerevisiae*.

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

[7]

[Total: 18]

- 3** Plant tissue culture has many commercial applications. These include the production of large numbers of genetically identical plants and the culturing of plant cells in a fermenter for the production of secondary products.

(a) Early attempts to culture plant tissue on a culture medium were not always successful.

This culture medium contained a range of substances that were known to be requirements for plant cell growth and division. These included:

- elements, present as mineral ions
- a sugar suitable for plant cells to use.

(i) Magnesium, phosphorus, calcium, sulphur and potassium are elements that would have been present as mineral ions in the culture medium.

Choose **two** elements from the list above and state **one** use in tissue culture for each of the elements chosen.

element

use

.....

element

use

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

(ii) Explain why a sugar was included in the culture medium.

.....

.....

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

(iii) Investigations have shown that the original culture medium lacked key components needed for successful plant cell growth and division.

State the term given to plant chemicals that stimulate cell growth and division.

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

(iv) Some investigators reported successful growth using root tips.

Suggest **one** reason why plant tissue culture was more successful using organs such as root tips.

.....

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

- (b)** In many stages of plant tissue culture strict aseptic technique needs to be used.

Air flow hoods are used in laboratories that carry out tissue culture techniques.

Outline the main features of an air flow hood **and** explain the benefits of its use in plant tissue culture.

..... [5]

- (c) State why it is an **advantage** that plants produced by plant tissue culture techniques are genetically identical.

..... [1]

- (d) In this question, one mark is available for the quality of use and organisation of scientific terms.

A flow diagram of one technique used to produce a large number of plants is shown in Fig. 3.1.

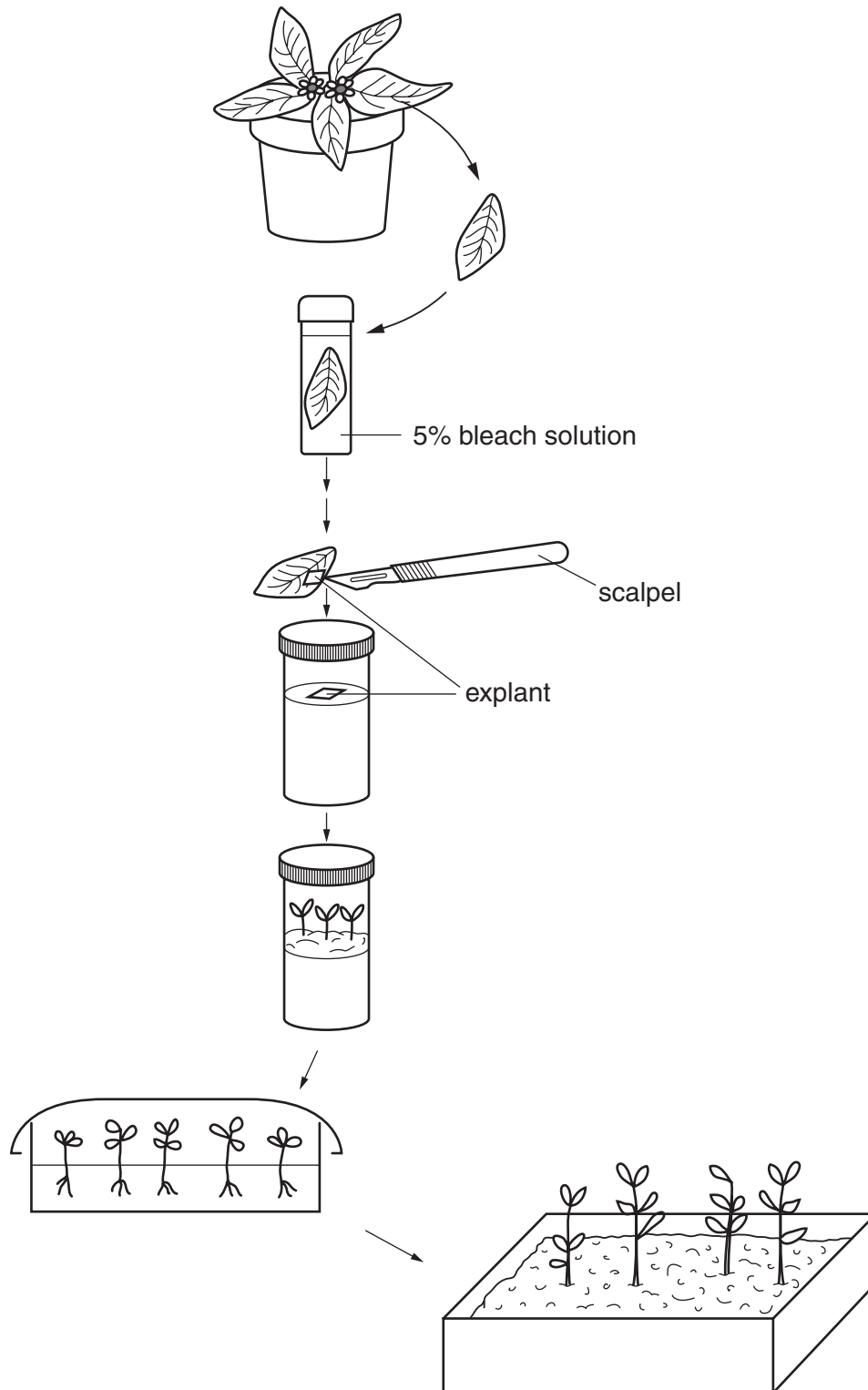


Fig. 3.1

With reference to Fig. 3.1, describe the technique of plant tissue culture.

You may annotate Fig. 3.1 to aid your answer.

..... [7]

Quality of Written Communication [1]

[Total: 20]

Turn over

- 4 Insulin and human growth hormone (HGH) are proteins of medical importance. They can be produced on a large scale using *Escherichia coli*.

- (a) Explain the reasons for using microorganisms, such as *E. coli*, in the large-scale production of insulin and HGH.

.....

.....

.....

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..... [3]

- (b) Table 4.1 summarises the sequence of events in the production of HGH by *E. coli*.

Complete Table 4.1 by adding **one** relevant statement in each of the empty boxes.

Table 4.1

description of event	explanation
obtain copies of gene with 'sticky ends'	the gene codes for the synthesis of HGH, the desired product
	acts as a vector for the transfer of the gene into the host
use restriction endonuclease enzyme	
mix vector and gene	
	completes gene splicing by sealing the sugar-phosphate backbone
	to obtain transformed or recombinant host <i>E. coli</i> cells
screen for successfully transformed cells	
	to obtain large amounts of HGH for extraction and purification

[7]

[Total: 10]

- 5 (a) Many people with diabetes rely on the use of glucose biosensors to give them quantitative values that enable them to calculate correct insulin doses for treatment.

A type of glucose biosensor has been developed that can be implanted under the skin and is connected to a monitoring device that displays glucose levels. The biosensor incorporates immobilised glucose oxidase.

The source of glucose oxidase is a fungus, *Aspergillus niger*.

- (i) Suggest **one** advantage of using an implantable glucose biosensor rather than a traditional portable glucose biosensor.

.....
 [1]

- (ii) Safety tests have been performed to see whether glucose oxidase can leach out of the biosensor.

Suggest why it is important to confirm that no leaching occurs.

.....

 [1]

Trastuzumab, used in the treatment of tumours, is an example of a humanised monoclonal antibody. Humanised antibody is a mouse-human hybrid antibody that is the product of genetic engineering.

The gene coding for the antibody is expressed by cells grown in culture and the antibody is secreted into the culture medium.

Trastuzumab binds to HER2 receptors found in large numbers on the surface of certain tumour cells. One result of this binding is that the cell ceases to grow and divide.

- (b) Explain the likely consequences of using mouse antibodies rather than humanised antibodies.

.....

 [3]

- (c) (i) Suggest why Trastuzumab can be more beneficial than chemotherapy for the treatment of some tumours.

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

- (ii) Suggest why Trastuzumab is not always the recommended treatment for other types of tumour.

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

- (d) Most polypeptides that are produced in mammalian cells undergo glycosylation; this is the addition of a sugar group to form a glycoprotein. Glycosylation takes place in the endoplasmic reticulum or Golgi body following translation at the ribosome.

Antibodies require glycosylation in order to function properly.

Suggest, with reasons, whether *E. coli* or mammalian cells would be better as a host for the production of humanised antibody.

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

[Total: 9]

Explain the advantages of enzyme immobilisation in manufacturing industries.

..... [7

Turn over

- (b) Figs. 6.1, 6.2 and 6.3 show the activity of free enzyme in solution compared to the activity of the same enzyme when immobilised.

A time course for starch hydrolysis using free and immobilised glucoamylase enzyme

(100% conversion represents the complete hydrolysis of starch to glucose).

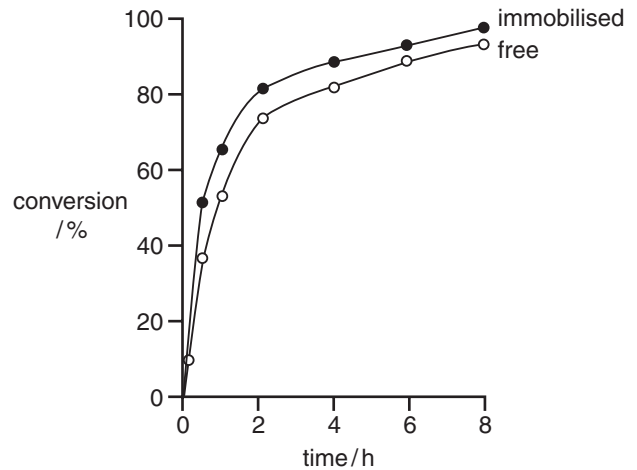


Fig. 6.1

The effect of pH on activity of lysozyme, an enzyme which lyses some bacterial cells by disrupting the peptidoglycan layer.

key

method 1 = chemically bonded

method 2 = adsorbed

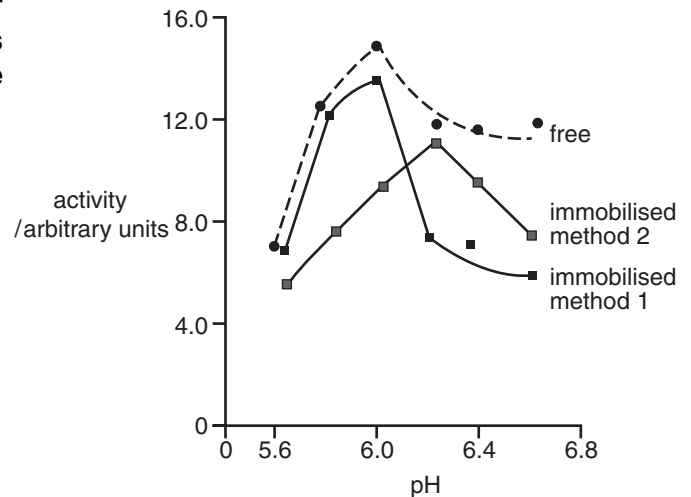


Fig. 6.2

The effect of pH on activity of glucose isomerase, an enzyme that is used in the production of high fructose syrups.

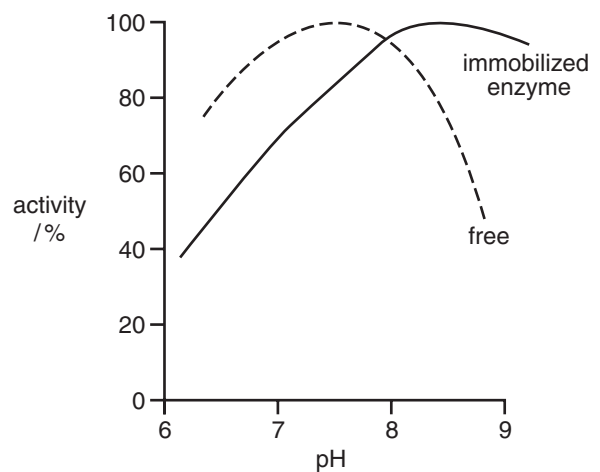


Fig. 6.3

Describe **and** comment on the data shown in Figs. 6.1, 6.2 and 6.3.

Fig. 6.1

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Fig. 6.2

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Fig. 6.3

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..... [6]

[Total: 14]

END OF QUESTION PAPER

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Copyright Acknowledgements:

- Fig. 6.1 Data source: L A Nakhapetyan & I I Menyailova, *Immobilized amyloglycosidase: preparation, properties and application for starch hydrolysis*, 1980
Fig. 6.2 Data source: Brown et al, *An organic milieu in immobilized enzyme synthesis and catalysis*, 1974
Fig. 6.3 Data source: B Atkinson, *Biochemical engineering and biotechnology handbook*, p590, Macmillan Publishers Ltd, 1991

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