

**ADVANCED GCE  
BIOLOGY**

Microbiology and Biotechnology

**MONDAY 28 JANUARY 2008**

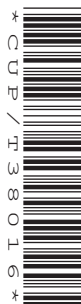
**2805/04**

Morning

Time: 1 hour 30 minutes

Candidates answer on the question paper.

**Additional materials:** Electronic calculator  
Ruler (cm/mm)



Candidate  
Forename

Candidate  
Surname

Centre  
Number

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Candidate  
Number

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**INSTRUCTIONS TO CANDIDATES**

- Write your name in capital letters, your Centre Number and Candidate Number in the boxes above.
- Use blue or 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.
- Do **not** write outside the box bordering each page.
- Write your answer to each question in the space provided.

**INFORMATION FOR CANDIDATES**

- The number of marks for each question 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.

**For Examiner's Use**

Qu.	Max.	Mark
1	20	
2	18	
3	19	
4	8	
5	20	
6	5	
<b>TOTAL</b>	<b>90</b>	

This document consists of **18** printed pages and **2** blank pages.

Answer **all** the questions.

- 1 (a) Table 1.1 shows some different features of microorganisms.

**Table 1.1**

microorganism	type of microorganism	prokaryotic	cell wall	mitochondria	autotrophic nutrition
<i>Escherichia coli</i>	bacterium	✓	✓	✗	✗
<i>Saccharomyces</i>					
<i>Fusarium graminearum</i>					
<i>Chlamydomonas</i>	protocist				✓
<i>Nitrobacter</i>					
<i>Plasmodium falciparum</i>			✗	✓	

key: ✓ = feature present ✗ = feature absent

- (i) Complete all the blank boxes in Table 1.1. [6]  
 (ii) Some of the microorganisms in Table 1.1 do not exhibit autotrophic nutrition.

Explain why they are not considered to be autotrophs.

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

- (iii) State the main component of the cell wall of any **two** of the organisms which have a tick in the 'cell wall' column.

In each case name the organism and the cell wall component.

1. organism .....  
    cell wall component .....  
 2. organism .....  
    cell wall component .....[2]

- (b) Three students, **X**, **Y** and **Z**, were asked to determine the nature of the cell wall of *E. coli* using the Gram staining technique. The students were reminded of the main steps of the procedure:

Step 1: Prepare a smear on a clean microscope slide.  
 Step 2: Pass the smear through a low Bunsen flame.  
 Step 3: Add crystal violet solution and wash off with iodine solution.  
 Step 4: Add alcohol.  
 Step 5: Add carbol fuschin solution or safranin solution and air dry.  
 Step 6: Add a cover slip and view under the microscope.

- (i) Student **X** followed the instructions carefully and had a successful outcome.  
 Student **Y** did not carry out Step 2.  
 Student **Z** did not carry out Step 5.

Describe **and** explain the observations made by the three students in Step 6.

Student **X** .....

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

Student **Y** .....

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

Student **Z** .....

.....  
 .....  
 .....  
 .....[6]

(ii) Write a risk assessment for the Gram staining procedure.

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..... [4]

[Total: 20]

**5**  
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- 2 (a) Fig. 2.1 shows a time course for penicillin production in a fed-batch culture. The curve representing the cell biomass is incomplete.

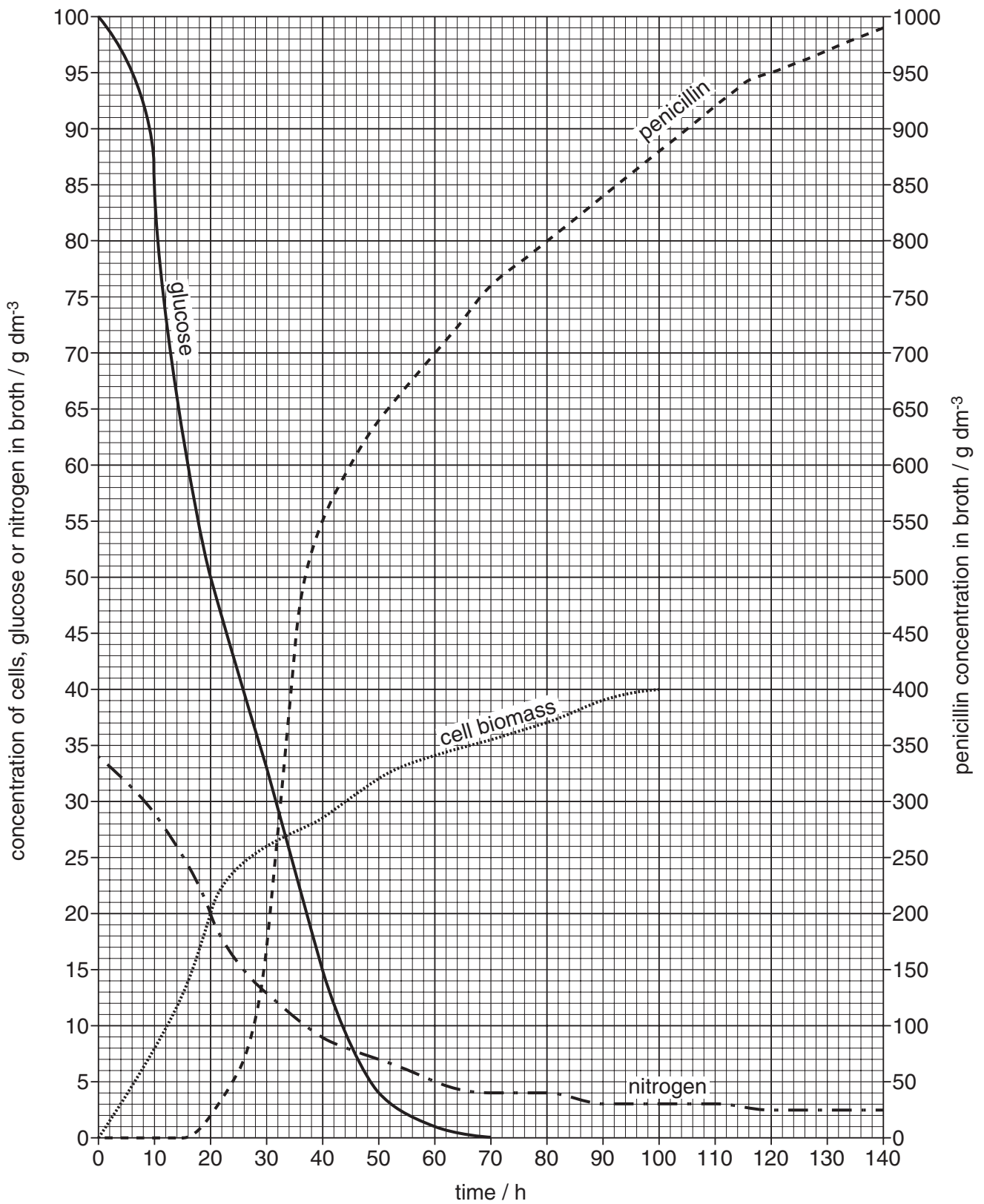


Fig. 2.1

- (i) Complete the cell biomass curve in Fig. 2.1 using the data in Table 2.1.

**Table 2.1**

time / h	cell biomass / g dm <sup>-3</sup>
110	41.5
120	42.0
130	42.5
140	43.0

[1]

- (ii) Calculate the maximum rate of **penicillin** production in g dm<sup>-3</sup> h<sup>-1</sup>. Show your working.

Answer = .....g dm<sup>-3</sup> h<sup>-1</sup> [2]

- (iii) With reference to the relevant curves in Fig. 2.1, describe **and** explain the changes in concentration of **glucose** and **nitrogen**.

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.....[4]

- (b) Penicillin is a secondary metabolite.

Explain whether the data in Fig. 2.1 provide evidence to support this statement.

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.....[2]

Compare the advantages **and** disadvantages of batch and continuous fermentation.

You should refer in your answer to production, operating conditions, fermenter design and potential problems that can occur.

[illegible]



[Total: 18]

- 3 Robert Koch, a German physician, discovered the causes of the diseases anthrax and tuberculosis.

Koch and his colleagues were also responsible for a number of other important advances in microbiology.

- (a) Koch examined the blood of animals infected with anthrax and identified rod-shaped microorganisms,  $1\text{ }\mu\text{m} \times 4\text{ }\mu\text{m}$  in size. He also observed the formation of resistant dormant stages, known as endospores. These endospores caused anthrax when injected into healthy mice.

- (i) A cell with a diameter of 0.1 mm is only just visible to the naked eye.

State whether Koch would have been able to see the microorganism that causes anthrax without the use of a microscope. Justify your answer.

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

- (ii) State the phase of a population growth curve during which endospore **formation** is likely to occur. Explain your answer.

phase .....  
 explanation .....  
 .....[2]

- (b) Koch developed techniques that,

- enabled microorganisms to be cultured on solid media;
- separated the cells so that individual colonies developed.

Initially, he used boiled, sliced potatoes as the solid medium. The inoculated slices of potato were incubated under glass bell jars.

- (i) Suggest why Koch was successful in culturing only some types of microorganism using potato slices.

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

- (ii) Explain the advantage of incubating potato slices under glass bell jars.

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

- (c) Koch subsequently improved his methods to identify disease-causing microorganisms. Fluid from a patient with a disease was cultured on the surface of a solid gelatin medium. Individual colonies of different types of microorganism were obtained.

- (i) Koch inoculated cells from each of the different colonies separately into small mammals.

Explain how this enabled him to identify which microorganism had caused the disease.

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.....[2]

- (ii) Koch assumed that an individual colony resulted from a single cell.

Explain whether or not he was justified in making this assumption.

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.....[2]

- (iii) Koch was advised to use agar, instead of gelatin, to solidify the medium. The gelatin medium melted at temperatures above 28°C and was digested by some of the microorganisms growing on it.

Explain why agar, rather than gelatin, is a more suitable agent to solidify the medium.

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.....[3]

(d) Koch's technique of isolating pure cultures from a mixture is still used.

Outline how you would obtain isolated colonies from a tube of broth culture that contains a mixture of microorganisms. Assume,

- that the work area is disinfected;
- an updraft of warm air is provided by a Bunsen burner flame;
- you are provided with an inoculating loop;
- you have access to sterile Petri dishes containing sterile nutrient agar;
- the dishes will be partially taped before incubation.

You may use the space below for diagrams.

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[4]

[Total: 19]

**13**  
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- 4 In this question, one mark is available for the quality of use and organisation of scientific terms.

Fig. 4.1 shows the secondary **aerobic** treatment of sewage using the activated sludge method.

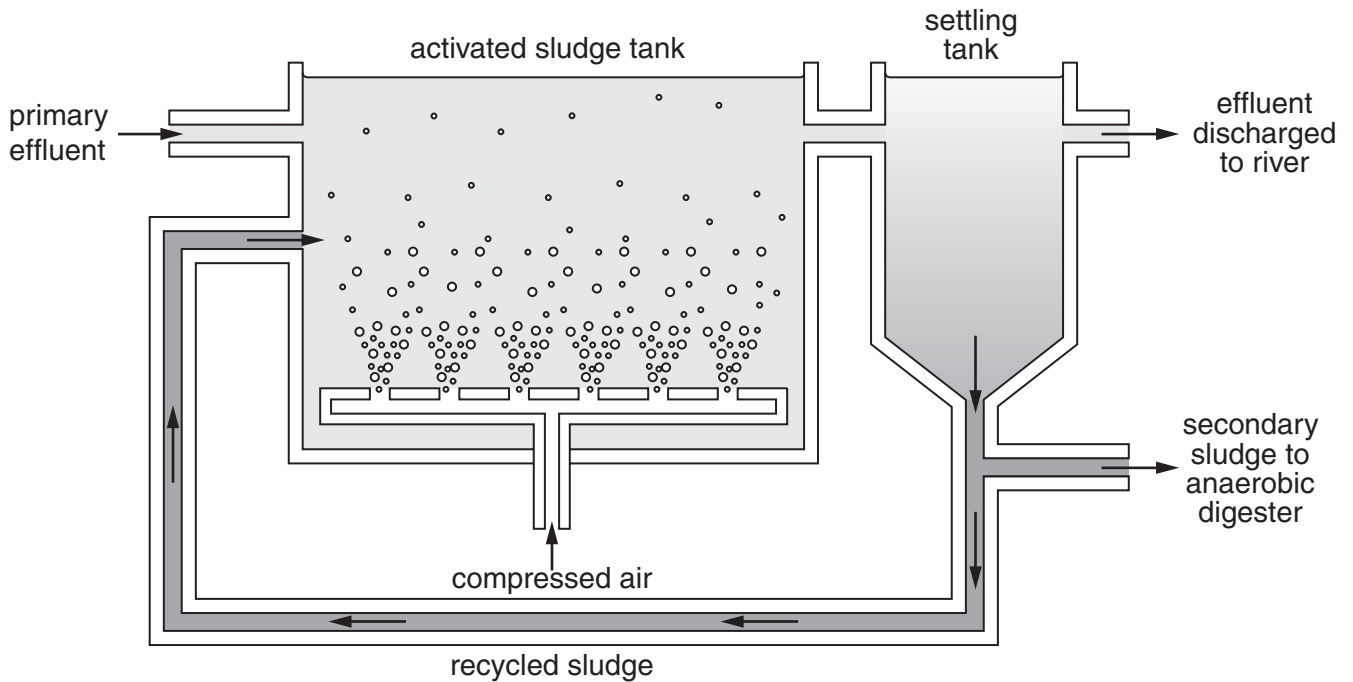


Fig. 4.1

Describe the role of microorganisms in this process.

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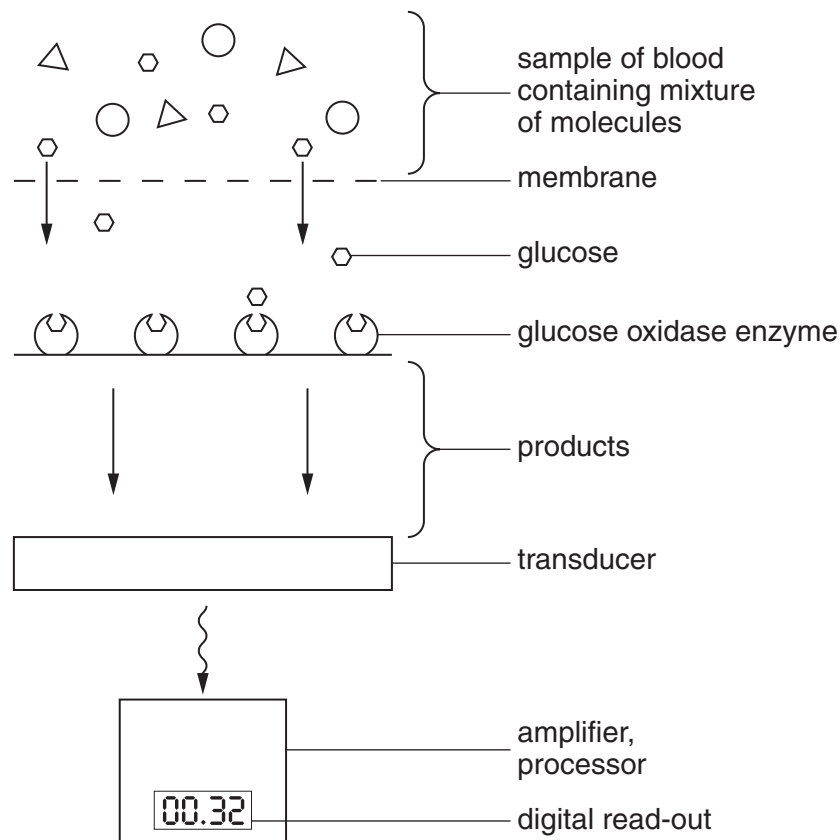
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[Total: 8]

- 5 (a) The use of biosensors for the detection of chemicals, such as glucose and organophosphates, has proved to be a reliable alternative to other methods.

Glucose biosensors, first developed in the 1980s, enable diabetics conveniently and easily to monitor their blood glucose. Fig. 5.1 shows the key components of one type of glucose biosensor.



**Fig. 5.1**

The development of organophosphate biosensors has benefits for health care and environmental monitoring.

With the increasing demand for food production, the use of organophosphates as insecticides to protect crops has increased. However, organophosphates can remain in the environment and are potentially toxic to humans and other animals.

- (i) Explain how water and food supplies may contain organophosphates.

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.....[2]



- (ii) Many organophosphates are irreversible inhibitors of acetylcholinesterase.

Explain why this makes them harmful to human health.

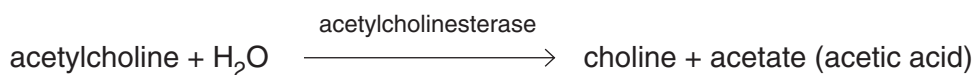
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.....[2]

- (iii) The equation below summarises the reaction catalysed by acetylcholinesterase.



Design a simple biosensor using acetylcholinesterase to detect the presence of a harmful organophosphate in a sample of river water. Include explanations for each element of your design.

You may wish to draw a labelled diagram in the space below to help your answer.

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.....[5]

- (iv) Suggest the advantages of organophosphate biosensors compared with other detection methods.

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.....[3]

- (b) Using genetic modification, crop plants resistant to the herbicide glyphosate can be produced. Glyphosate does not act on acetylcholinesterase but inhibits other enzyme systems.

- (i) Explain why a different biosensor to that used in (a)(iii) and (iv) would need to be developed to detect glyphosate in a sample of river water.

.....

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.....[2]

- (ii) Suggest **one** advantage and **one** disadvantage of producing glyphosate-resistant crop plants.

advantage .....

.....

disadvantage .....

.....[2]

- (c) Crop plants can be genetically modified for glyphosate resistance by using a restriction endonuclease and DNA ligase.

Describe the roles of these two enzymes in genetic manipulation.

restriction endonuclease .....

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DNA ligase .....

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.....[4]

[Total: 20]

- 6 Fig. 6.1 represents a poster produced by a student. The poster contains information about the use of microorganisms as a food source. Unfortunately, each box contains a mistake.

For each box, circle the mistake and write the correct word in the space provided. The first one has been done for you.

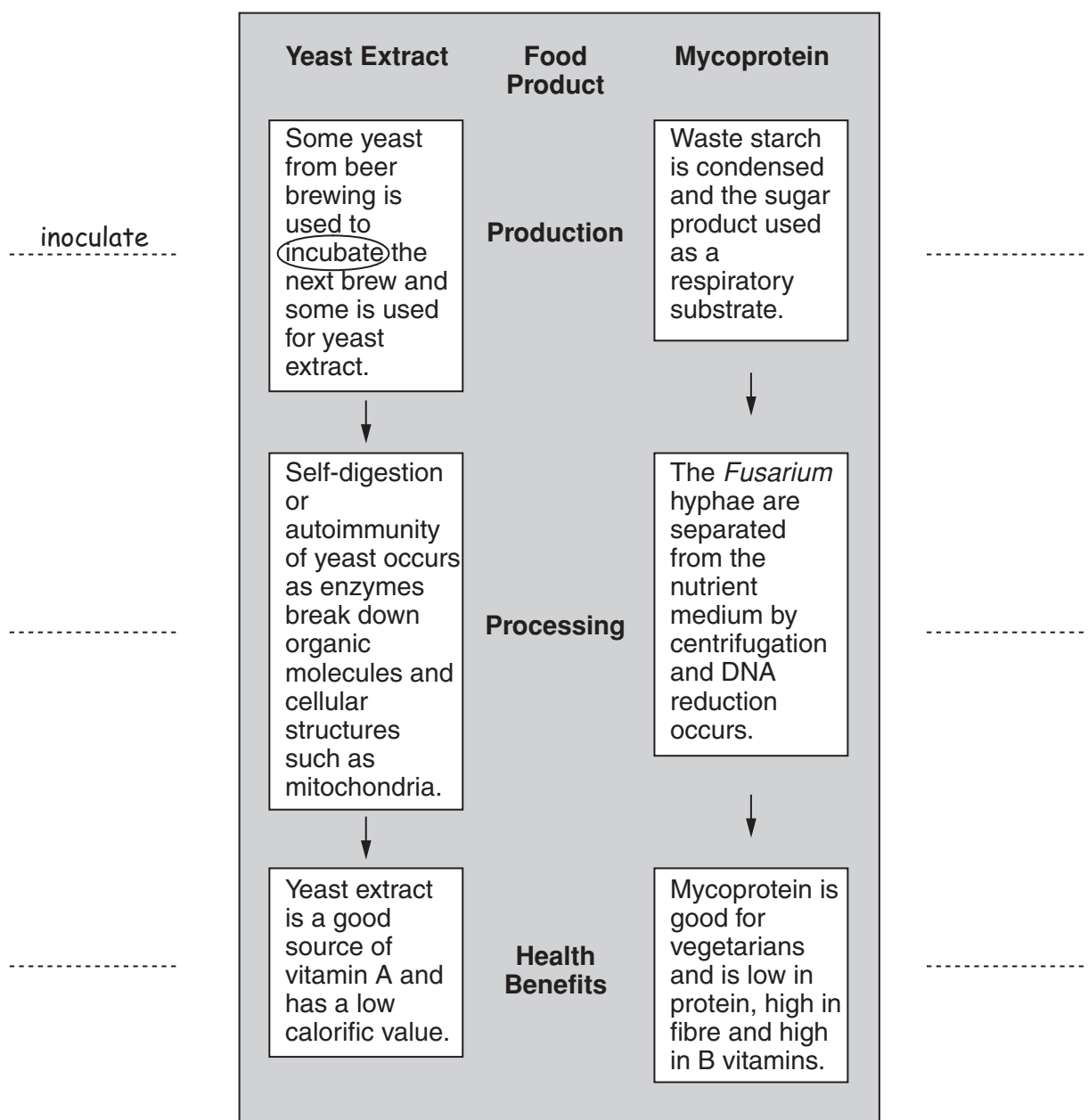


Fig. 6.1

[5]  
[Total: 5]

## END OF QUESTION PAPER

Copyright Acknowledgements:

Fig. 2.1 Adapted from Queener, S. and Swartz, R. (1979) *Penicillins: biosynthetic and semisynthetic*, in A. H. Rose (ed.), *Economic Microbiology, Volume 3: Secondary Products of Metabolism*, p. 35, Academic Press Inc.

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