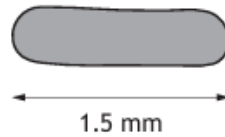


Microbe Growth past papers

1. The diagram represents a species of archaea as viewed under a microscope.



(1 mm = 1000 μm)

The actual length of this cell was 3 micrometres (μm).

The microscope had a total magnification of

- A $\times 5$
- B $\times 50$
- C $\times 500$
- D $\times 5000$

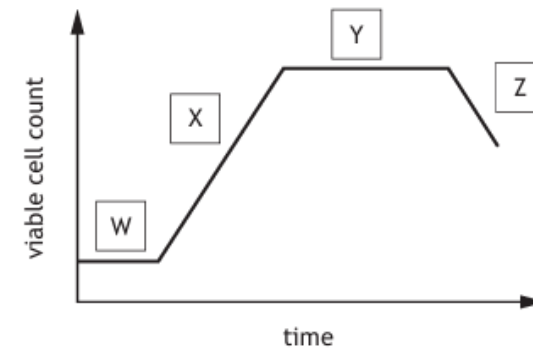
2. Which statement about culturing micro-organisms is **not** correct?

- A Some micro-organisms can use light as an energy source.
- B All micro-organisms require a chemical substrate as an energy source.
- C Sterility, pH, temperature, and oxygen levels can be monitored.
- D Some micro-organisms can synthesise their own amino acids and vitamins.

3. In which of the following domains of life are microorganisms found?

- A Bacteria only
- B Archaea only
- C Bacteria and archaea only
- D Bacteria, archaea and eukaryotes

4. The graph shows the phases of growth in a bacterial culture.



Which statement is correct?

- A X is the log phase and enzymes are being induced.
- B Z is the death phase where total cell count is decreasing.
- C Y is the stationary phase and nutrients are starting to run out.
- D W is the lag phase and secondary metabolites are being produced.

5. Which of the following statements about micro-organisms is correct?

- A All micro-organisms require amino acids in their growth medium.
- B Micro-organisms use a variety of metabolic substrates.
- C Micro-organisms include bacteria and archaea only.
- D Light can be used as an energy source by all micro-organisms grown in culture.

6. Which statement describes an event that occurs during the lag phase of microbial growth?

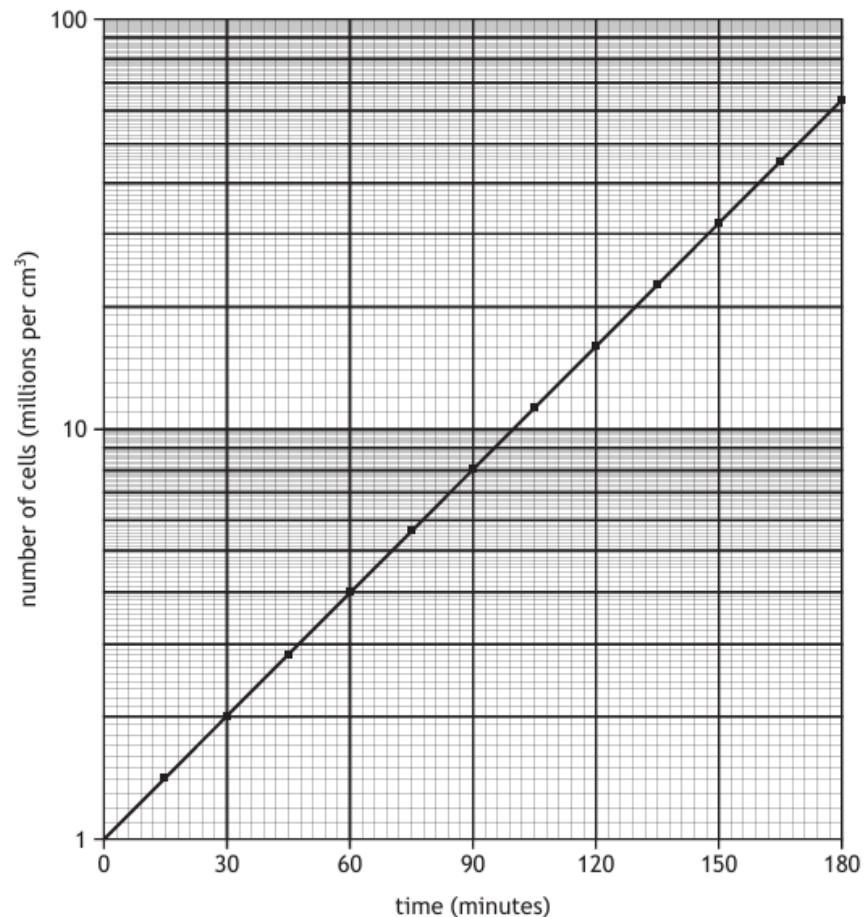
- A Secondary metabolites are produced
- B Certain enzymes are induced
- C Most rapid growth occurs due to plentiful nutrients
- D The culture medium becomes depleted of nutrients

Microbe Growth past papers

7

Yeast cells were cultured in a growth medium and the number of cells were counted at regular intervals over a period of 180 minutes.

The semi-logarithmic graph shows the number of cells per cm^3 of culture medium during this period.



How many yeast cells were present in the culture after 2 hours?

- A 1.6 million per cm^3
- B 10.6 million per cm^3
- C 16.0 million per cm^3
- D 70.0 million per cm^3

8

The following list relates to growth phases in a culture of the fungus *Penicillium chrysogenum*.

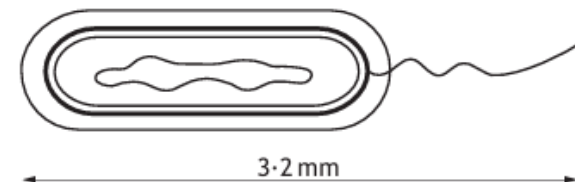
1. Growth is most rapid
2. Nutrients are completely depleted
3. Enzymes are induced
4. Antibiotics are produced

Which row in the table identifies the growth phases of this culture?

	Lag phase	Log phase	Stationary phase	Death phase
A	3	2	4	1
B	1	2	3	4
C	3	1	4	2
D	4	1	3	2

9

The diagram shows a bacterial cell that has been magnified 800 times.



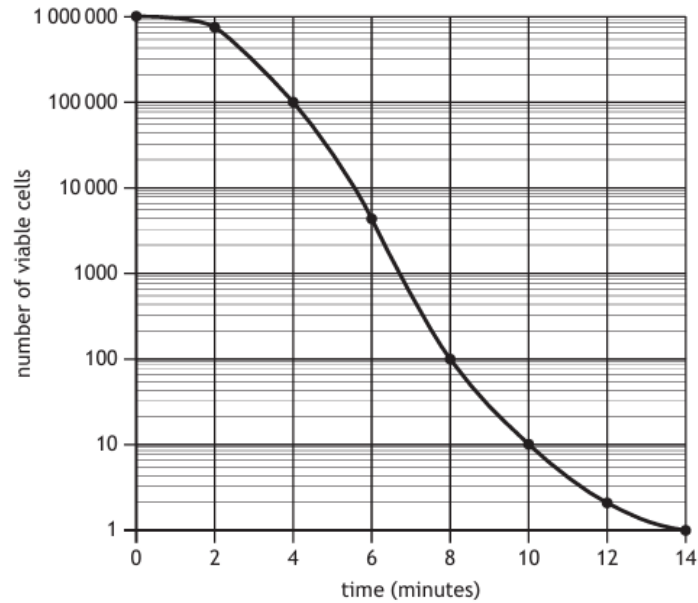
The length of the cell in micrometres (μm) is

- A 0.004
- B 0.04
- C 0.4
- D 4.0

Microbe Growth past papers

- 10 Bacterial cells were exposed to disinfectant for 14 minutes. Every 2 minutes a sample was taken and the number of viable cells counted.

The results are shown in the graph.



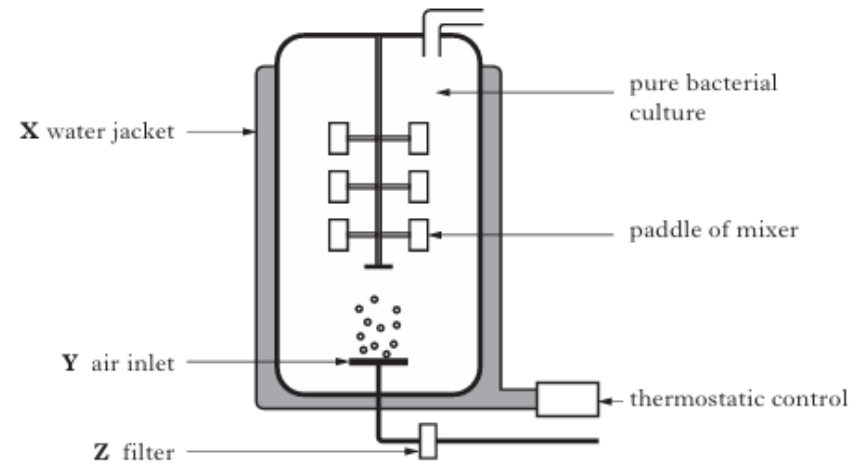
Calculate the percentage decrease in viable cells after being exposed to disinfectant for 6 minutes.

- A 99.60
- B 99.87
- C 996 000.00
- D 998 700.00

- 11 Buffers are added to culture media to

- A provide vitamins
- B maintain the pH
- C provide fatty acids
- D maintain the temperature.

- 12 The diagram below shows a fermenter.



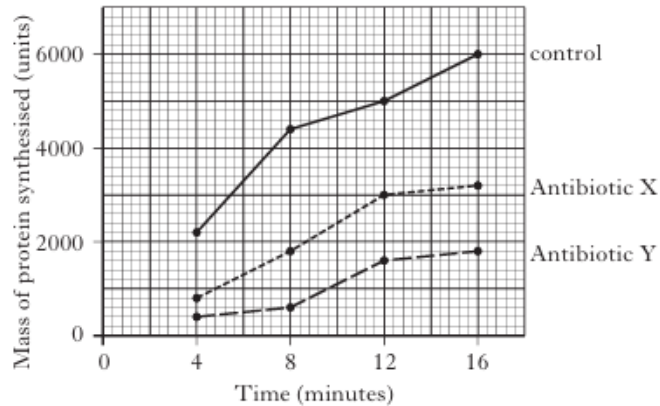
Which line in the table below matches correctly the parts of the fermenter labelled X, Y and Z with the functions they are involved in?

Function of fermenter parts			
	<i>maintaining temperature</i>	<i>controlling oxygen levels</i>	<i>maintaining sterility</i>
A	X	Y	Z
B	X	Z	Y
C	Y	X	Z
D	Y	Z	X

Microbe Growth past papers

- 13 An experiment was carried out to compare the effect of two antibiotics, X and Y, on the rate of protein synthesis in bacterial cells.

The results are shown in the graph below.

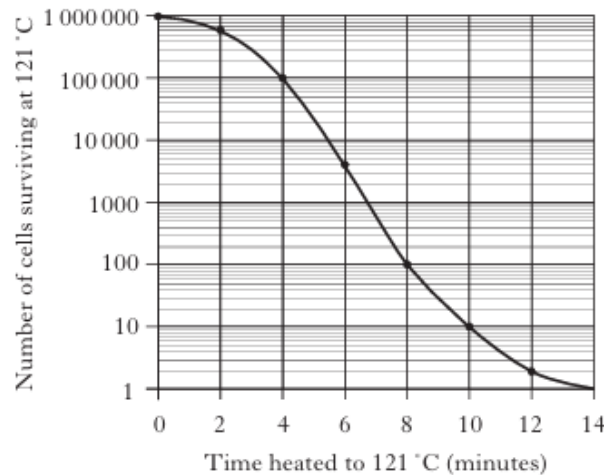


Which of the following conclusions from the graph is valid?

- A Antibiotic X was less effective than antibiotic Y in inhibiting bacterial protein synthesis.
- B Antibiotic Y was less effective than antibiotic X in inhibiting bacterial protein synthesis.
- C Bacterial protein synthesis was inhibited to the greatest extent without antibiotics.
- D The rate of protein synthesis was greatest between 12 and 16 minutes in all cases.

- 14 Bacterial cells were heated to 121 °C for increasing lengths of time to determine the number of live cells left after treatment.

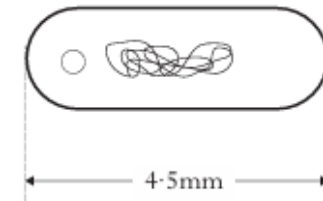
The graph below shows the number of bacterial cells which survived.



How many cells survive after 6 minutes?

- A 1300
- B 4000
- C 5000
- D 5500

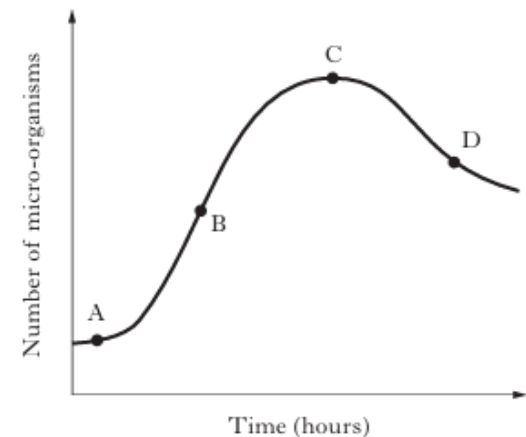
- 15 The diagram below shows an image of a bacterial cell that has been magnified 1500 times.



What is the actual length of the cell in micrometres (μm)?

- A 0.003
- B 3.000
- C 6750
- D 6.750

- 16 The graph below shows a typical growth pattern for micro-organisms in culture.



At which point on the graph are most enzymes being induced to metabolise available substrates?

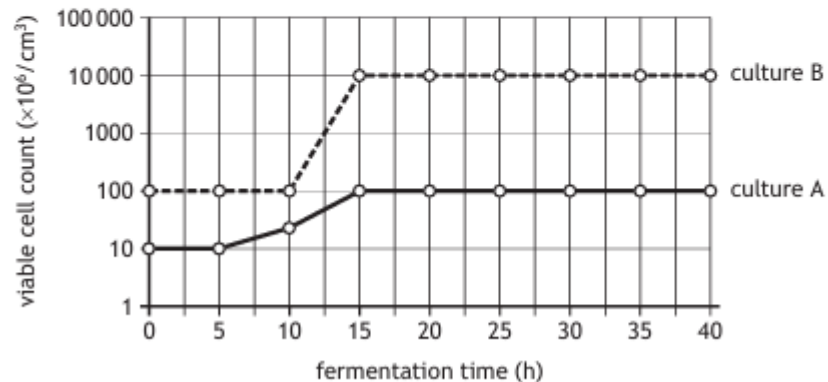
Microbe Growth past papers

- 1 The bacterium *Bacillus subtilis* (*B. subtilis*) produces antibiotics.

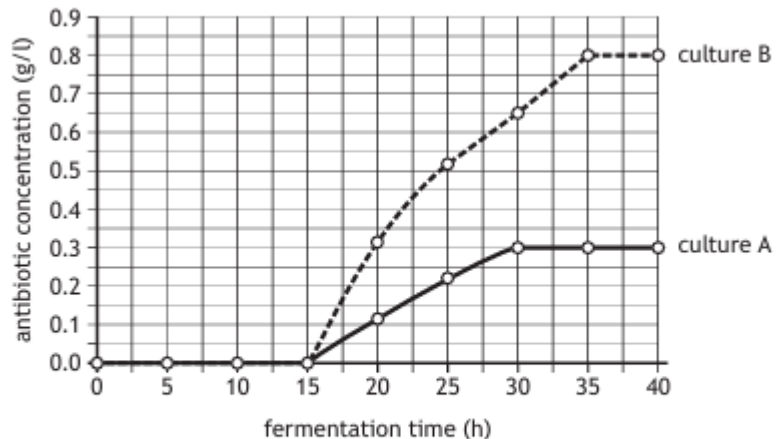
A study was carried out to compare the viable cell count and antibiotic concentration of two cultures of *B. subtilis* (A and B) grown under different conditions.

The results are shown in the graphs.

Graph 1



Graph 2



- (a) Using information in Graph 1 state the duration of the lag phase in culture A.

_____ hours

- b) Antibiotics are secondary metabolites.

- (i) Use evidence from both graphs to support this statement.

1

- (ii) Describe an ecological advantage to *B. subtilis* of producing antibiotics in its natural environment.

1

- c) Name a culture condition that would be controlled when producing antibiotics in a fermenter.

1

- 2 The bacteria *Streptomyces* is a microorganism found in soil. It produces a secondary metabolite, the antibiotic streptomycin, which kills other microorganisms. *Streptomyces* live in close association with plant roots. These plants produce soluble carbohydrates which are released into the soil through their roots.

- (i) Name the growth phase during which streptomycin is produced.

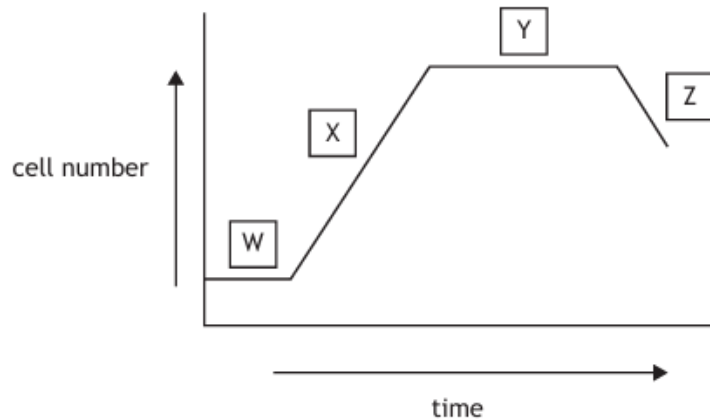
1

- (ii) Explain the advantage to *Streptomyces* of producing an antibiotic such as streptomycin.

1

Microbe Growth past papers

- 3 A growth curve in a culture of bacteria is shown in the diagram.



- (a) In culturing bacteria it is important to control the culture conditions in the growth medium.

Name one condition which should be controlled.

- (b) (i) Name the phase in which secondary metabolites such as antibiotics are produced.

Describe the ecological advantage of this to bacteria in the wild.

Phase _____

Advantage _____

- (ii) State the letter which indicates a region of the graph in which most enzymes are being induced to metabolise the available substrate.

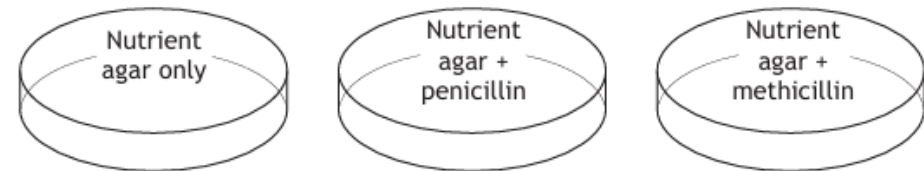
Letter _____

- (iii) State **one** reason for the decrease in number of cells at phase Z.

- 4 *Staphylococcus aureus* (*S.aureus*) is a species of bacteria that lives on human skin. This species of bacteria can cause infections if it enters the body through a wound. *S.aureus* infections can be treated with antibiotics such as methicillin and penicillin.

Infections can be caused by a strain of *S.aureus* called MRSA which is resistant to methicillin and penicillin and is becoming more common.

Samples were taken from a patient suspected of having a bacterial infection. The samples were used to inoculate plates of agar as shown in the diagram below.

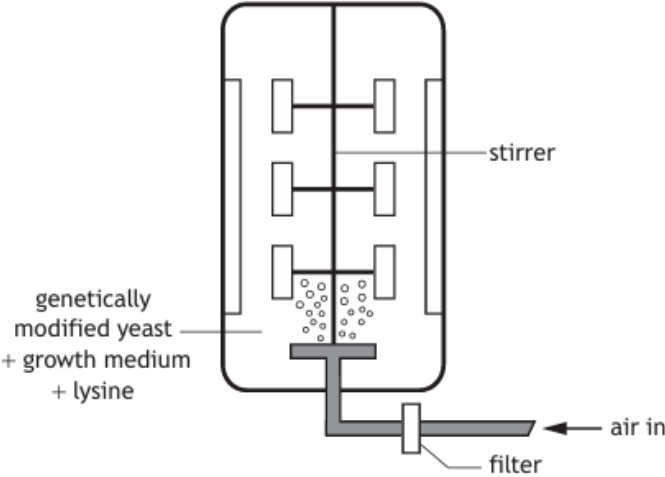


- (i) Predict the results if the cause of the bacterial infection was MRSA.

- (ii) The nutrient agar contained specific amino acids required for protein synthesis. Suggest **one other** type of complex compound that the nutrient agar may have contained.

Microbe Growth past papers

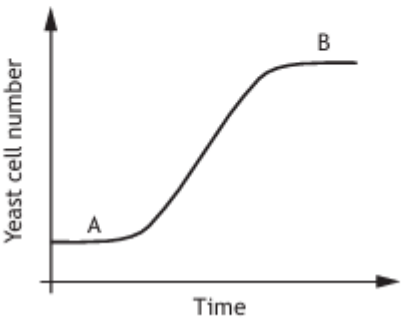
- 5 The diagram shows genetically modified yeast growing in a fermenter in a medium to which the amino acid lysine has been added.



- (a) (i) Name the process for which the yeast cells need the amino acid lysine. 1
- _____
- (ii) The fermenter contains 5.5 litres of growth medium.
Calculate the mass of lysine which should be added to the medium to give a concentration of 300 mg/l. 1
- Space for calculation*
- _____ mg
- (iii) The air entering the fermenter passes through a filter to prevent contamination.
Explain why it is necessary to prevent contamination of the culture. 1
- _____
- _____

- (a) (continued)
- (iv) The optimum pH for yeast growth is 4.5.
Suggest how this pH could be maintained in the fermenter. 1
- _____

- (b) Some phases of a growth curve of yeast culture are shown.



- Complete the table by selecting growth phase A or B. Name the chosen phase and describe an event which occurs during that phase of growth. 2

Letter	Phase of growth	Description

- (c) Describe a safety mechanism used to prevent the survival of genetically modified microorganisms in the external environment. 1
- _____
- _____

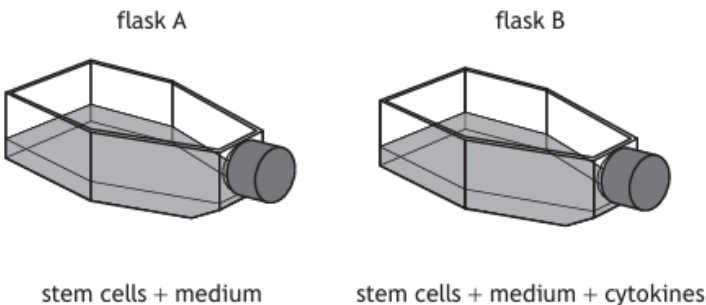
Microbe Growth past papers

Marks

6 When culturing stem cells, substances called cytokines can be included in the culture medium.

An investigation was carried out to determine the effect of cytokines on the growth of stem cells.

Cultures of stem cells were set up as shown.



The cells were cultured for 10 days and the cell count was recorded every 2 days.

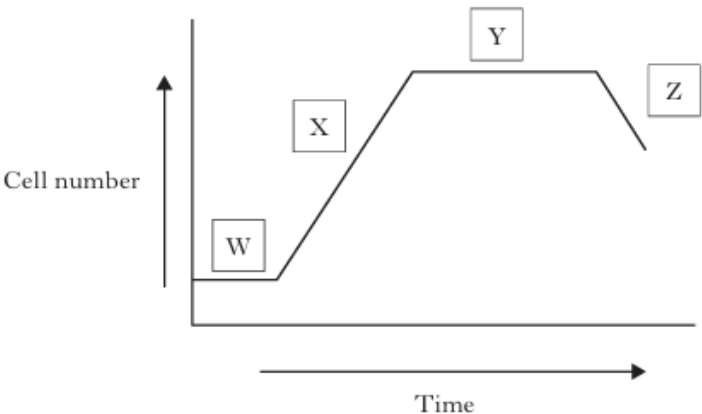
The results are shown in the table.

Day	Cell count (thousand cells per cm ³)	
	Medium only	Medium + cytokines
2	22	36
4	50	130
6	330	760
8	520	1800
10	13	8200

The cell count was a viable cell count.

Use evidence from the table to support this statement.

7 A growth curve in a culture of bacteria is shown in the diagram below.



(a) In culturing bacteria it is important to control the pH of the culture medium. Describe how the pH of a culture medium can be controlled.

1

(b) (i) Name each of the phases of growth indicated by letters on the graph.

W _____

X _____

Y _____

Z _____

2

(ii) Give the letter which indicates a region of the graph in which most enzymes are being induced to metabolise the available substrate.

Letter _____

1

(iii) Give **one** reason for the decrease in number of cells at phase Z.

1

1

Microbe Growth past papers

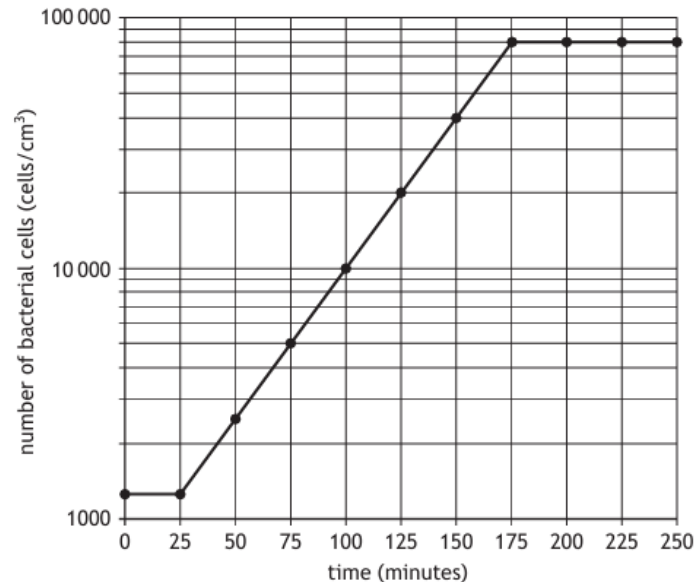
8

Some species of bacteria can be grown in fermenters to produce vinegar. The bacteria convert ethanol in the growth medium into vinegar.

An investigation was carried out to determine the relationship between the number of bacterial cells and the concentrations of ethanol and vinegar over 250 minutes.

The results are shown in Graph 1 and Graph 2.

Graph 1



a) Using information in Graph 1 state the duration of the log phase.

1

_____ minutes

b) Using information from Graph 2, explain why there is no further increase in vinegar concentration after 175 minutes.

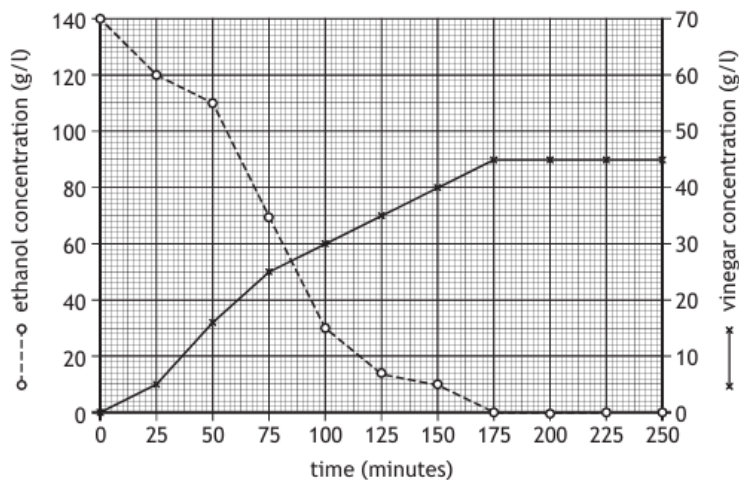
1

c) Using information from Graph 1 and Graph 2, identify the concentration of vinegar when the number of bacteria is 40 000 cells per cm³.

1

_____ g/l

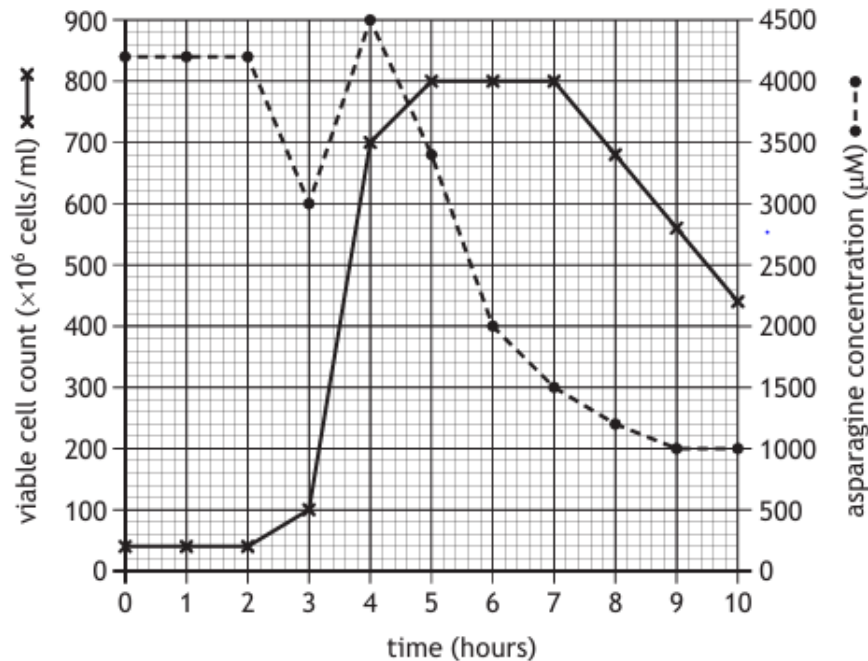
Graph 2



Microbe Growth past papers

- 9 Growth media used to culture the bacteria *E. coli* often contain the amino acid asparagine even though *E. coli* can produce this amino acid.

The graph shows the viable cell count of *E. coli* and the asparagine concentration in a culture grown over a period of 10 hours.



- (a) (i) State the asparagine concentration when the viable cell count was 100×10^6 cells/ml.

1

_____ μM

- (ii) Calculate the simplest whole number ratio of the asparagine concentration at 4 hours to that at 10 hours.

1

Space for calculation

_____ : _____
4 hours 10 hours

- (b) (i) Using the information given, suggest why there is a rapid increase in asparagine concentration between 3 and 4 hours.

2

- (ii) Explain the changes in the viable cell count between 7 and 10 hours.

1

- (c) (i) State why *E. coli* requires the amino acid asparagine for growth.

1

- (ii) Bacteria require other complex molecules apart from amino acids for biosynthesis.

Name another complex molecule that could be added to the growth medium.

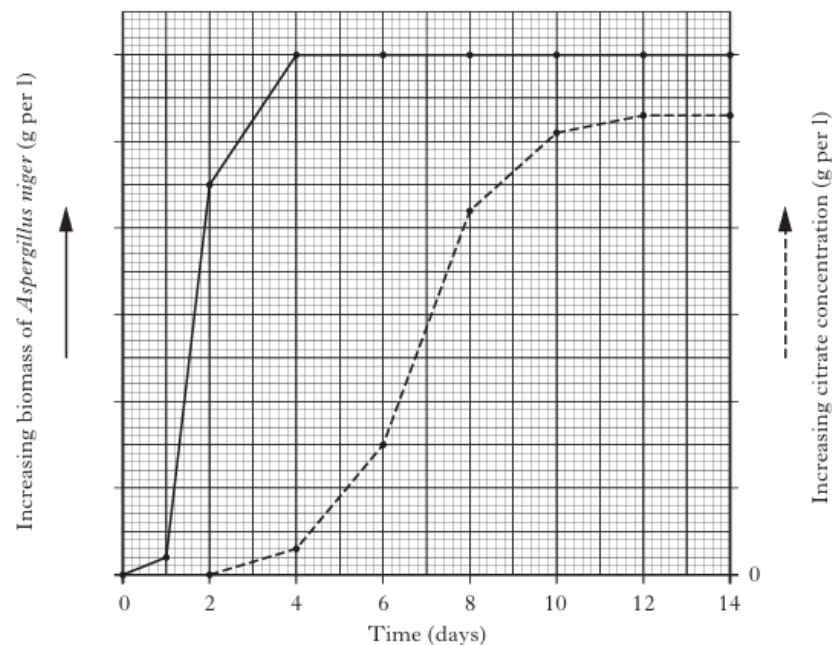
1

Microbe Growth past papers

- 10 Citrate is used in the food industry as a flavouring agent. Large quantities of citrate are produced in fermenters by the fungus *Aspergillus niger*.

The fungus was added to a growth medium and grown in a fermenter for 14 days.

The graph below shows the changes in biomass of the fungus and in the concentration of citrate over the growth period.



- (a) Identify the time period during which the log (exponential) phase of the growth of *Aspergillus niger* occurs.

Tick (✓) the correct box.

☐

Day 0 – 1

☐

Day 1 – 4

☐

Day 4 – 12

☐

Day 12 – 14

1

- (b) Suggest **one** reason why citrate was not produced until day 2.

1

- c) Industrial fermentation at optimum temperature and pH is used to make useful products.

Apart from enzyme inhibitors, give an example of a substance which could be added to the fermenter to give increased yield of the desired product.

1

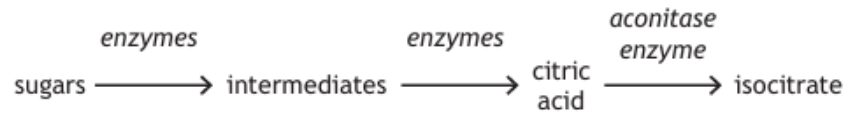
- 11 Write notes on the phases of growth of micro-organisms.

7

Microbe Growth past papers

- 12 The fungus *Aspergillus niger* (*A.niger*) is used to produce citric acid in fermenters using sugars as substrates.

Citric acid is an intermediate in a metabolic pathway as shown.



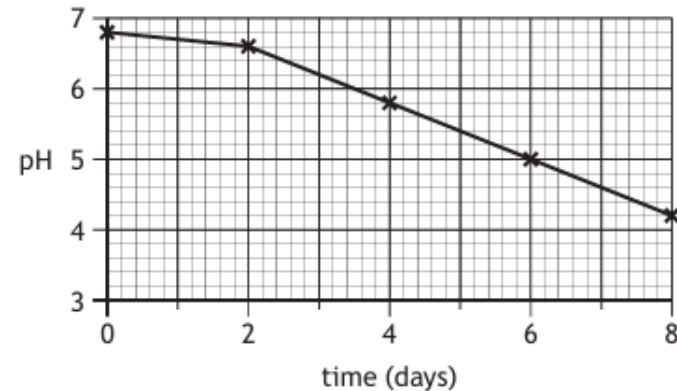
- (a) The aconitase enzyme requires iron to function.
Explain why the growth medium used to produce citric acid should not contain iron.

1

- (b) The optimum temperature for citric acid production by *A.niger* is 30 °C.
Explain why less citric acid would be produced if the temperature in the fermenter was reduced.

1

A.niger was grown in a fermenter over an 8 day period and the pH was measured every 2 days. The results are shown in the graph.



- (i) Predict the pH at 10 days. 1

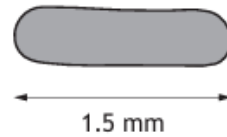
- (ii) Suggest a reason for the change in the pH observed. 1

- (iii) State a variable, other than temperature or pH, that should be monitored and controlled in this process. 1

- (iv) Explain why it was necessary to sterilise the fermenter before *A.niger* was added. 2

Microbe Growth past papers

1. The diagram represents a species of archaea as viewed under a microscope.



(1 mm = 1000 μm)

The actual length of this cell was 3 micrometres (μm).

The microscope had a total magnification of

- A $\times 5$
- B $\times 50$
- ☒ C $\times 500$
- D $\times 5000$

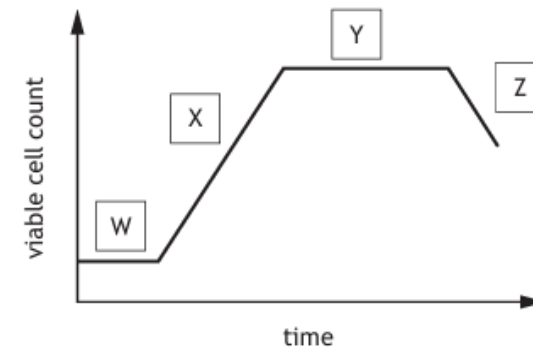
2. Which statement about culturing micro-organisms is **not** correct?

- A Some micro-organisms can use light as an energy source.
- ☒ B All micro-organisms require a chemical substrate as an energy source.
- C Sterility, pH, temperature, and oxygen levels can be monitored.
- D Some micro-organisms can synthesise their own amino acids and vitamins.

3. In which of the following domains of life are microorganisms found?

- A Bacteria only
- B Archaea only
- C Bacteria and archaea only
- ☒ D Bacteria, archaea and eukaryotes

4. The graph shows the phases of growth in a bacterial culture.



Which statement is correct?

- A X is the log phase and enzymes are being induced.
- B Z is the death phase where total cell count is decreasing.
- ☒ C Y is the stationary phase and nutrients are starting to run out.
- D W is the lag phase and secondary metabolites are being produced.

5. Which of the following statements about micro-organisms is correct?

- A All micro-organisms require amino acids in their growth medium.
- ☒ B Micro-organisms use a variety of metabolic substrates.
- C Micro-organisms include bacteria and archaea only.
- D Light can be used as an energy source by all micro-organisms grown in culture.

6. Which statement describes an event that occurs during the lag phase of microbial growth?

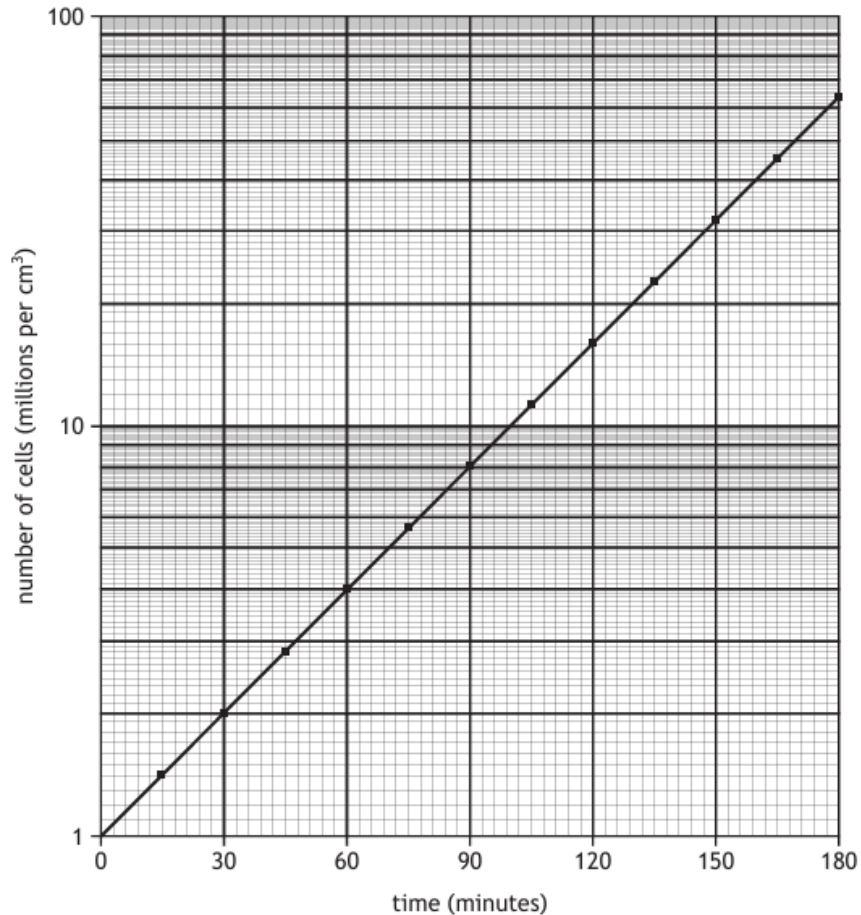
- A Secondary metabolites are produced
- ☒ B Certain enzymes are induced
- C Most rapid growth occurs due to plentiful nutrients
- D The culture medium becomes depleted of nutrients

Microbe Growth past papers

7

Yeast cells were cultured in a growth medium and the number of cells were counted at regular intervals over a period of 180 minutes.

The semi-logarithmic graph shows the number of cells per cm^3 of culture medium during this period.



How many yeast cells were present in the culture after 2 hours?

- A 1.6 million per cm^3
- B 10.6 million per cm^3
- C 16.0 million per cm^3**
- D 70.0 million per cm^3

8

The following list relates to growth phases in a culture of the fungus *Penicillium chrysogenum*.

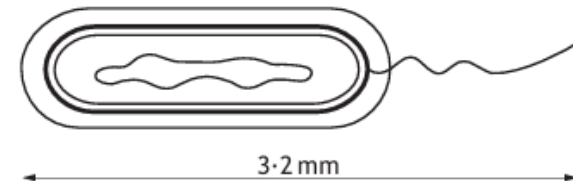
1. Growth is most rapid
2. Nutrients are completely depleted
3. Enzymes are induced
4. Antibiotics are produced

Which row in the table identifies the growth phases of this culture?

	Lag phase	Log phase	Stationary phase	Death phase
A	3	2	4	1
B	1	2	3	4
C	3	1	4	2
D	4	1	3	2

9

The diagram shows a bacterial cell that has been magnified 800 times.



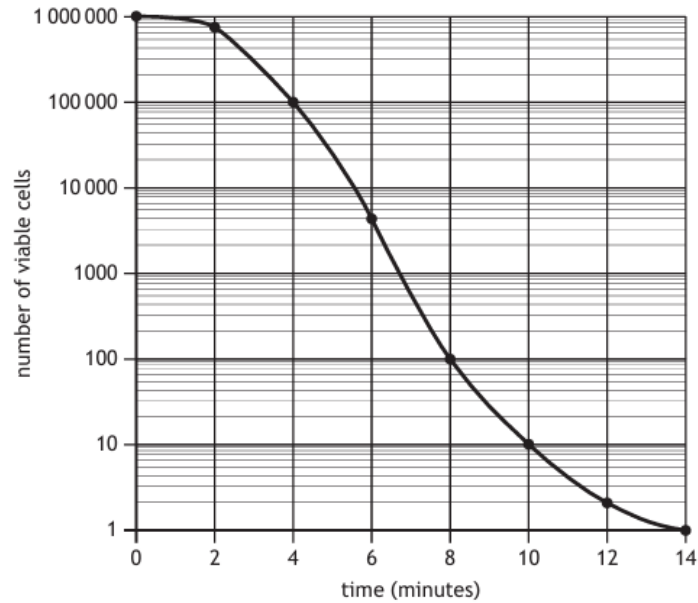
The length of the cell in micrometres (μm) is

- A 0.004
- B 0.04
- C 0.4
- D 4.0**

Microbe Growth past papers

- 10 Bacterial cells were exposed to disinfectant for 14 minutes. Every 2 minutes a sample was taken and the number of viable cells counted.

The results are shown in the graph.



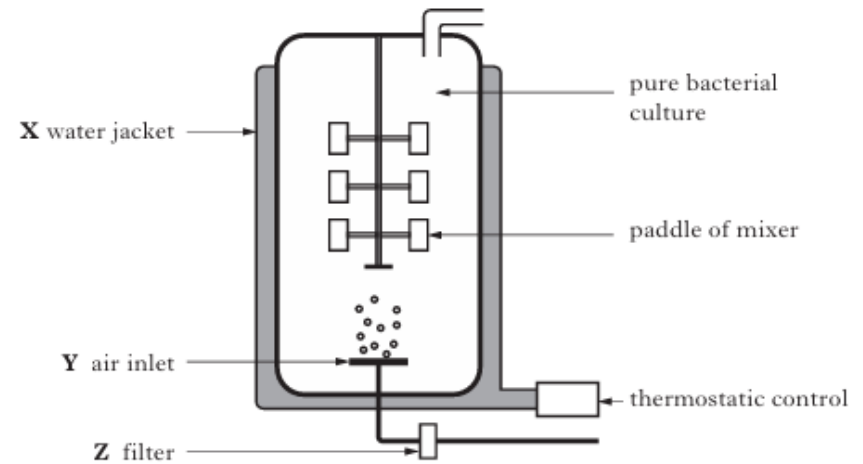
Calculate the percentage decrease in viable cells after being exposed to disinfectant for 6 minutes.

- A 99.60
B 99.87
C 996 000.00
D 998 700.00

- 11 Buffers are added to culture media to

- A provide vitamins
B maintain the pH
C provide fatty acids
D maintain the temperature.

- 12 The diagram below shows a fermenter.



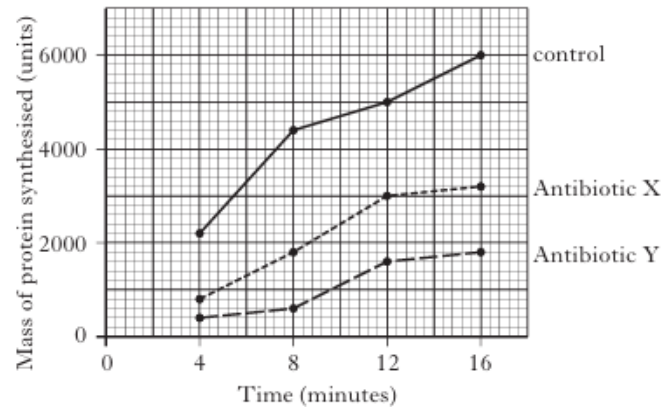
Which line in the table below matches correctly the parts of the fermenter labelled X, Y and Z with the functions they are involved in?

Function of fermenter parts			
	<i>maintaining temperature</i>	<i>controlling oxygen levels</i>	<i>maintaining sterility</i>
A	X	Y	Z
B	X	Z	Y
C	Y	X	Z
D	Y	Z	X

Microbe Growth past papers

- 13 An experiment was carried out to compare the effect of two antibiotics, X and Y, on the rate of protein synthesis in bacterial cells.

The results are shown in the graph below.

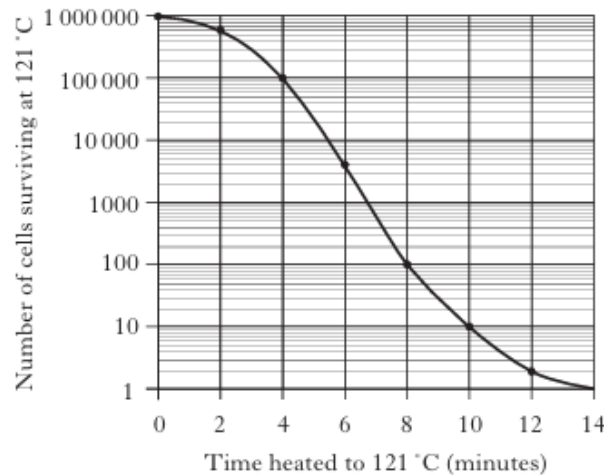


Which of the following conclusions from the graph is valid?

- ☒ A Antibiotic X was less effective than antibiotic Y in inhibiting bacterial protein synthesis.
- ☐ B Antibiotic Y was less effective than antibiotic X in inhibiting bacterial protein synthesis.
- ☐ C Bacterial protein synthesis was inhibited to the greatest extent without antibiotics.
- ☐ D The rate of protein synthesis was greatest between 12 and 16 minutes in all cases.

- 14 Bacterial cells were heated to 121 °C for increasing lengths of time to determine the number of live cells left after treatment.

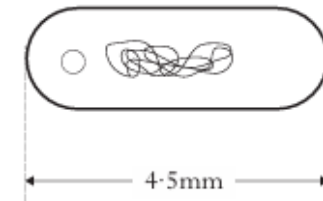
The graph below shows the number of bacterial cells which survived.



How many cells survive after 6 minutes?

- ☒ A 1300
- ☐ B 4000
- ☐ C 5000
- ☐ D 5500

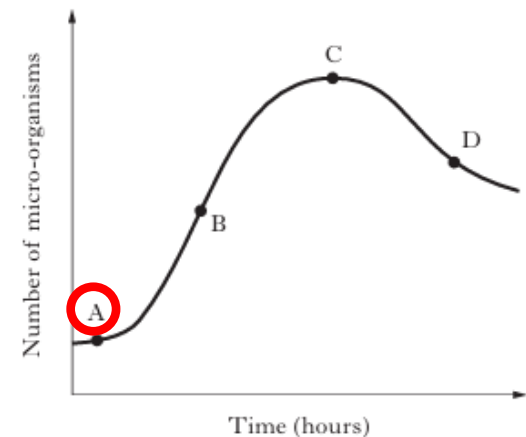
- 15 The diagram below shows an image of a bacterial cell that has been magnified 1500 times.



What is the actual length of the cell in micrometres (μm)?

- ☐ A 0.003
- ☒ B 3.000
- ☐ C 6750
- ☐ D 6.750

- 16 The graph below shows a typical growth pattern for micro-organisms in culture.



At which point on the graph are most enzymes being induced to metabolise available substrates?

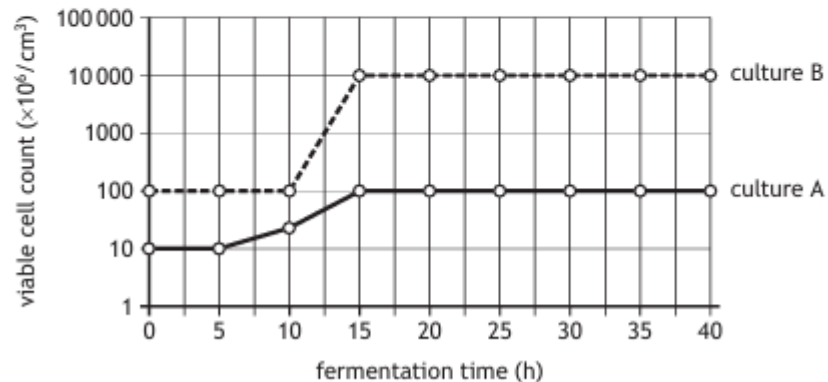
Microbe Growth past papers

- 1 The bacterium *Bacillus subtilis* (*B. subtilis*) produces antibiotics.

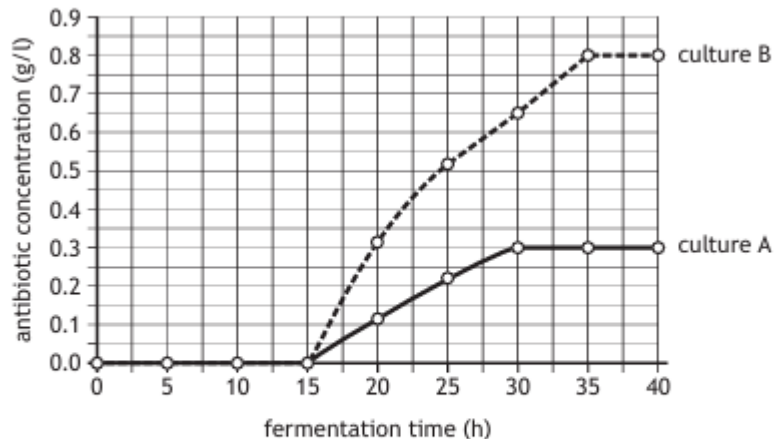
A study was carried out to compare the viable cell count and antibiotic concentration of two cultures of *B. subtilis* (A and B) grown under different conditions.

The results are shown in the graphs.

Graph 1



Graph 2



- (a) Using information in Graph 1 state the duration of the lag phase in culture A.

5

hours

- (b) Antibiotics are secondary metabolites.

- (i) Use evidence from both graphs to support this statement.

Antibiotic is only produced in the stationary phase.

- (ii) Describe an ecological advantage to *B. subtilis* of producing antibiotics in its natural environment.

Outcompetes other microbes

- (c) Name a culture condition that would be controlled when producing antibiotics in a fermenter.

Sterility/temperature/oxygen concentration/pH

- 2 The bacteria *Streptomyces* is a microorganism found in soil. It produces a secondary metabolite, the antibiotic streptomycin, which kills other microorganisms. *Streptomyces* live in close association with plant roots. These plants produce soluble carbohydrates which are released into the soil through their roots.

- (i) Name the growth phase during which streptomycin is produced.

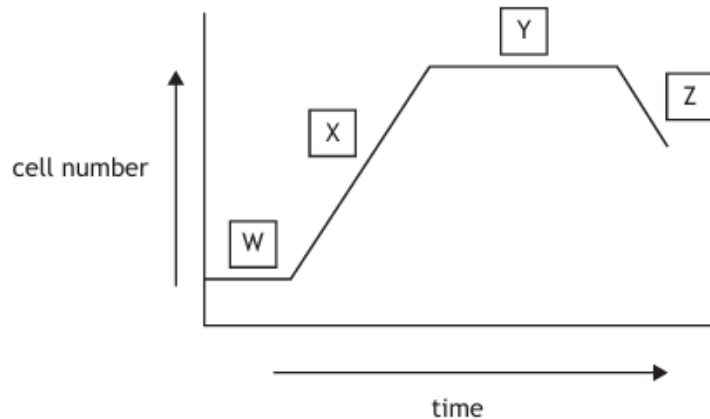
stationary

- (ii) Explain the advantage to *Streptomyces* of producing an antibiotic such as streptomycin.

Outcompetes other microbes

Microbe Growth past papers

- 3 A growth curve in a culture of bacteria is shown in the diagram.



- (a) In culturing bacteria it is important to control the culture conditions in the growth medium.

Name one condition which should be controlled.

Sterility/pH/temperature/oxygen concentration

- (b) (i) Name the phase in which secondary metabolites such as antibiotics are produced.

Describe the ecological advantage of this to bacteria in the wild.

Phase Stationary

Advantage Outcompetes other microbes.

- (ii) State the letter which indicates a region of the graph in which most enzymes are being induced to metabolise the available substrate.

Letter W

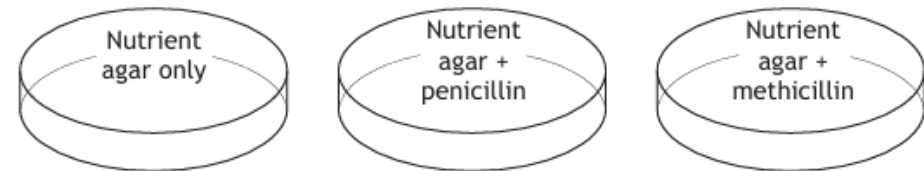
- (iii) State **one** reason for the decrease in number of cells at phase Z.

Nutrients ran out/toxic metabolites build up

- 4 *Staphylococcus aureus* (*S.aureus*) is a species of bacteria that lives on human skin. This species of bacteria can cause infections if it enters the body through a wound. *S.aureus* infections can be treated with antibiotics such as methicillin and penicillin.

Infections can be caused by a strain of *S.aureus* called MRSA which is resistant to methicillin and penicillin and is becoming more common.

Samples were taken from a patient suspected of having a bacterial infection. The samples were used to inoculate plates of agar as shown in the diagram below.



- (i) Predict the results if the cause of the bacterial infection was MRSA.

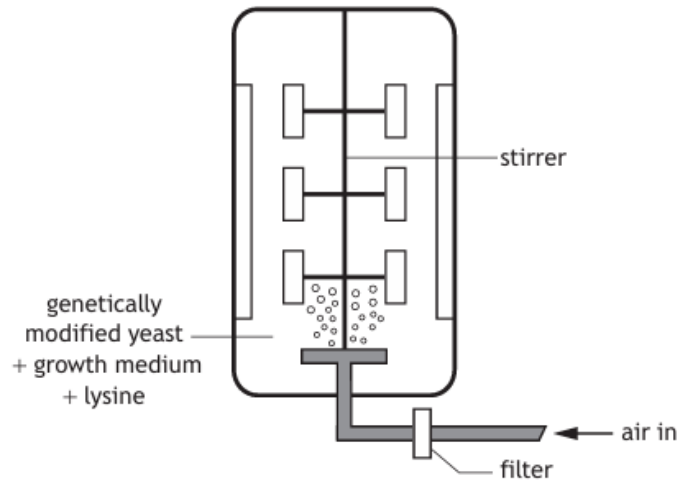
MRSA/bacterial growth on ALL plates

- (ii) The nutrient agar contained specific amino acids required for protein synthesis. Suggest **one other** type of complex compound that the nutrient agar may have contained.

Vitamins OR fatty acids

Microbe Growth past papers

- 5 The diagram shows genetically modified yeast growing in a fermenter in a medium to which the amino acid lysine has been added.



- (a) (i) Name the process for which the yeast cells need the amino acid lysine.

Protein synthesis

- (ii) The fermenter contains 5.5 litres of growth medium.

Calculate the mass of lysine which should be added to the medium to give a concentration of 300 mg/l.

Space for calculation

$$5.5 \times 300\text{mg}$$

1650 mg

- (iii) The air entering the fermenter passes through a filter to prevent contamination.

Explain why it is necessary to prevent contamination of the culture.

Prevents other microbes using up nutrients

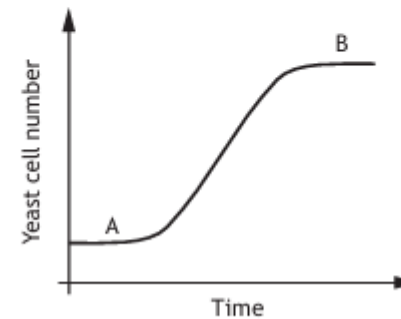
- (a) (continued)

- (iv) The optimum pH for yeast growth is 4.5.

Suggest how this pH could be maintained in the fermenter.

Using a buffer

- (b) Some phases of a growth curve of yeast culture are shown.



Complete the table by selecting growth phase A or B. Name the chosen phase and describe an event which occurs during that phase of growth.

Letter	Phase of growth	Description
A OR B	lag stationary	enzymes induced nutrients running out OR secondary metabolites being produced

- (c) Describe a safety mechanism used to prevent the survival of genetically modified microorganisms in the external environment.

Introduce genes in vector that prevents growth of microbe outside lab

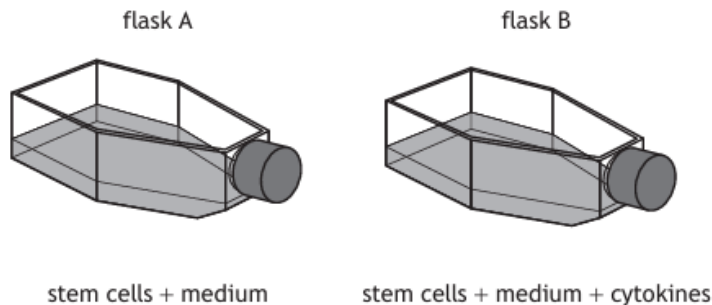
Microbe Growth past papers

Marks

- 6 When culturing stem cells, substances called cytokines can be included in the culture medium.

An investigation was carried out to determine the effect of cytokines on the growth of stem cells.

Cultures of stem cells were set up as shown.



The cells were cultured for 10 days and the cell count was recorded every 2 days.

The results are shown in the table.

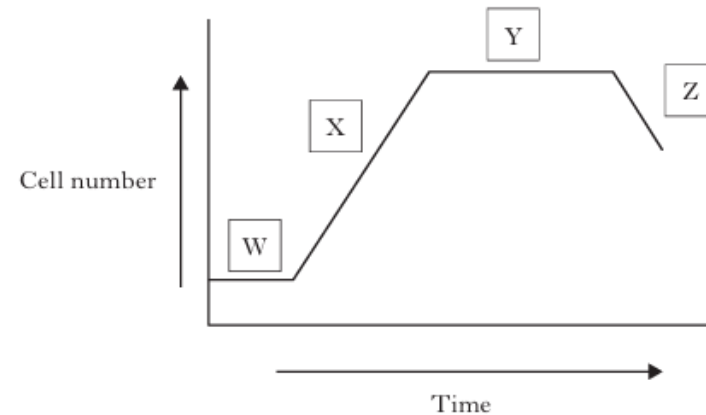
Day	Cell count (thousand cells per cm ³)	
	Medium only	Medium + cytokines
2	22	36
4	50	130
6	330	760
8	520	1800
10	13	8200

The cell count was a viable cell count.

Use evidence from the table to support this statement.

The cell count decreases after day 8

- 7 A growth curve in a culture of bacteria is shown in the diagram below.



- (a) In culturing bacteria it is important to control the pH of the culture medium. Describe how the pH of a culture medium can be controlled.

Adding buffer

1

- (b) (i) Name each of the phases of growth indicated by letters on the graph.

W Lag

X Log

Y Stationary

Z Death

2

- (ii) Give the letter which indicates a region of the graph in which most enzymes are being induced to metabolise the available substrate.

Letter W

1

- (iii) Give **one** reason for the decrease in number of cells at phase Z.

Nutrients ran out OR toxic metabolites have accumulated.

1

Microbe Growth past papers

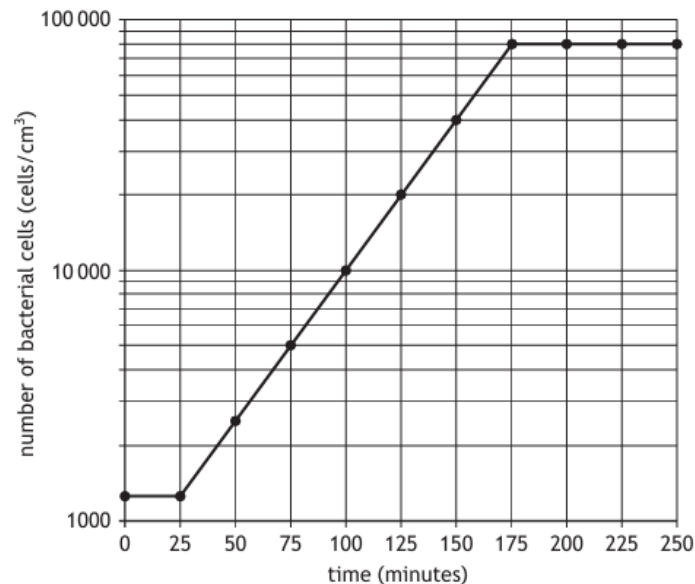
8

Some species of bacteria can be grown in fermenters to produce vinegar. The bacteria convert ethanol in the growth medium into vinegar.

An investigation was carried out to determine the relationship between the number of bacterial cells and the concentrations of ethanol and vinegar over 250 minutes.

The results are shown in Graph 1 and Graph 2.

Graph 1



a) Using information in Graph 1 state the duration of the log phase.

1

150 minutes

b) Using information from Graph 2, explain why there is no further increase in vinegar concentration after 175 minutes.

1

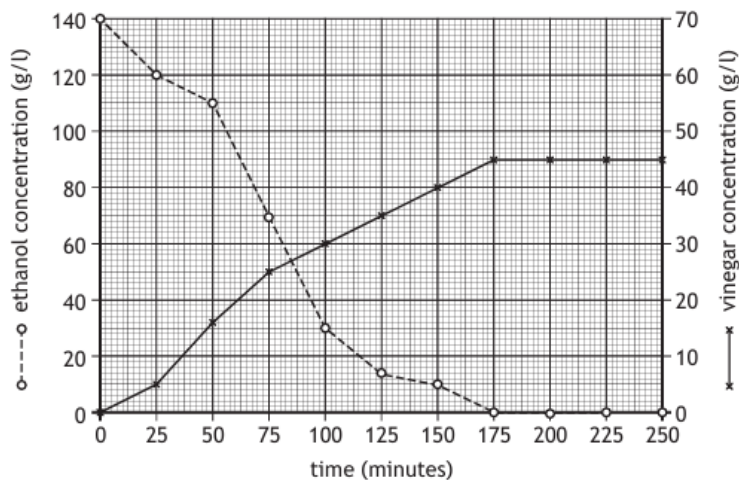
All ethanol has been used up/is at zero

c) Using information from Graph 1 and Graph 2, identify the concentration of vinegar when the number of bacteria is 40 000 cells per cm³.

1

40 g/l

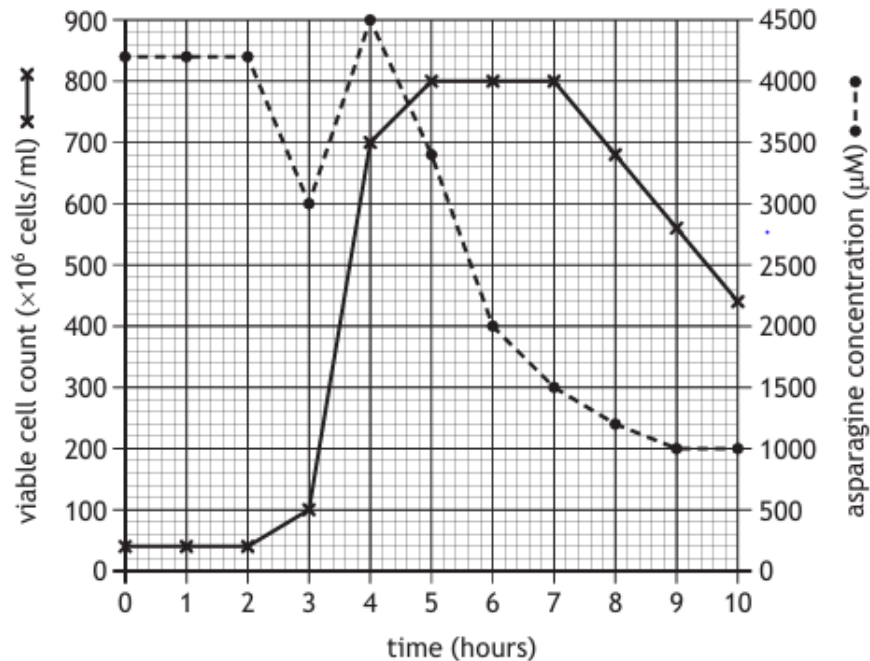
Graph 2



Microbe Growth past papers

- 9 Growth media used to culture the bacteria *E. coli* often contain the amino acid asparagine even though *E. coli* can produce this amino acid.

The graph shows the viable cell count of *E. coli* and the asparagine concentration in a culture grown over a period of 10 hours.



- (a) (i) State the asparagine concentration when the viable cell count was 100×10^6 cells/ml.

3000 μM

- (ii) Calculate the simplest whole number ratio of the asparagine concentration at 4 hours to that at 10 hours.

Space for calculation

9 : 2
4 hours : 10 hours

- (b) (i) Using the information given, suggest why there is a rapid increase in asparagine concentration between 3 and 4 hours.

More E Coli/bacteria (1)

E coli produces asparagine (1)

- (ii) Explain the changes in the viable cell count between 7 and 10 hours.

Toxic metabolites are accumulating OR nutrients running out

- (c) (i) State why *E. coli* requires the amino acid asparagine for growth.

To make protein / for protein synthesis

- (ii) Bacteria require other complex molecules apart from amino acids for biosynthesis.

Name another complex molecule that could be added to the growth medium.

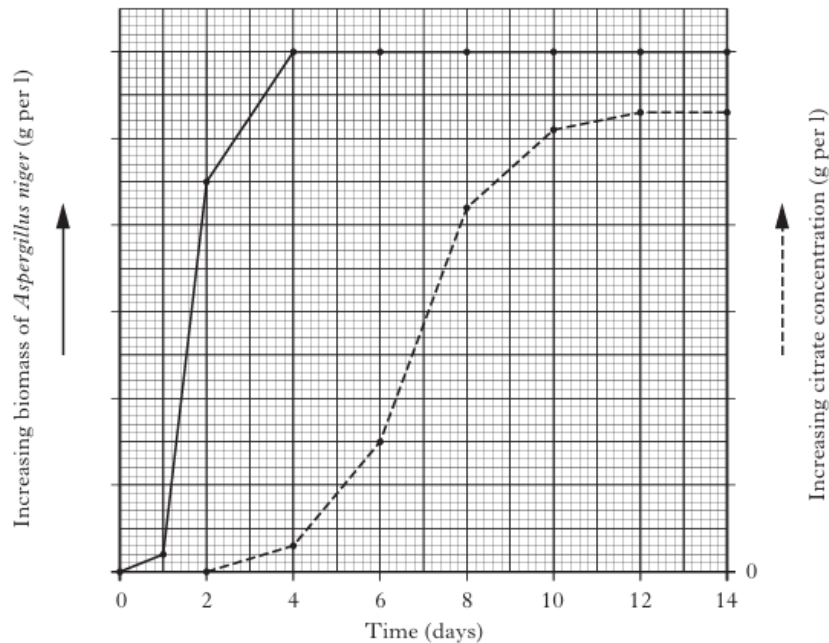
Fatty acids OR Vitamins (NOT glucose/carbohydrates etc)

Microbe Growth past papers

- 10 Citrate is used in the food industry as a flavouring agent. Large quantities of citrate are produced in fermenters by the fungus *Aspergillus niger*.

The fungus was added to a growth medium and grown in a fermenter for 14 days.

The graph below shows the changes in biomass of the fungus and in the concentration of citrate over the growth period.



- (a) Identify the time period during which the log (exponential) phase of the growth of *Aspergillus niger* occurs.

Tick (✓) the correct box.

☐

Day 0 – 1

☐

Day 1 – 4

☐

Day 4 – 12

☐

Day 12 – 14

1

- (b) Suggest **one** reason why citrate was not produced until day 2.

Enzymes being induced first.

1

- C) Industrial fermentation at optimum temperature and pH is used to make useful products.

Apart from enzyme inhibitors, give an example of a substance which could be added to the fermenter to give increased yield of the desired product.

Fatty acids/amino acids/vitamins

1

- 11 Write notes on the phases of growth of micro-organisms.

7

- all 4 phases named: lag, log/exponential, stationary and death
- lag phase when enzymes are induced

OR

- lag phase when DNA replicates
- log/exponential phase when fast/most growth occurs
- log/exponential phase when nutrients are plentiful
- stationary phase when nutrients are being depleted/used up

OR

- toxic metabolites/toxins produced/build up in stationary phase
- secondary metabolites/antibiotics are produced/build up in stationary phase
- antibiotics/secondary metabolites confer ecological advantage

OR

- antibiotics/secondary metabolites reduce/remove competition from other microorganisms
- death phase occurs due to toxic accumulation of metabolites/nutrients have run out
- viable cell count is only living micro-organisms

OR

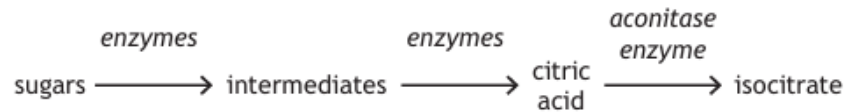
total cell count is living and dead micro-organisms

Any 7

Microbe Growth past papers

- 12 The fungus *Aspergillus niger* (*A.niger*) is used to produce citric acid in fermenters using sugars as substrates.

Citric acid is an intermediate in a metabolic pathway as shown.



- (a) The aconitase enzyme requires iron to function.

Explain why the growth medium used to produce citric acid should not contain iron.

1

Citric acid will be turned into isocitrate

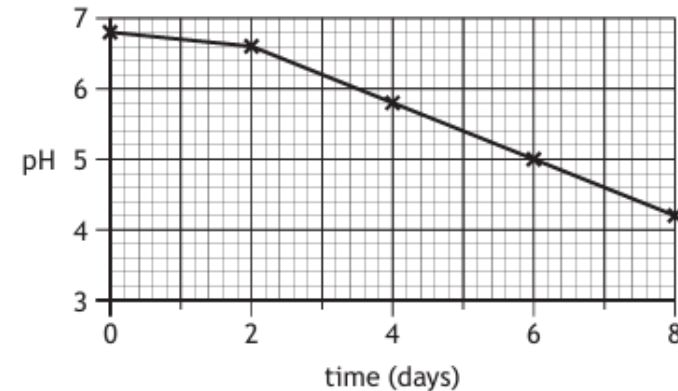
- (b) The optimum temperature for citric acid production by *A.niger* is 30 °C.

Explain why less citric acid would be produced if the temperature in the fermenter was reduced.

1

Lower enzyme activity

A.niger was grown in a fermenter over an 8 day period and the pH was measured every 2 days. The results are shown in the graph.



- (i) Predict the pH at 10 days.

1

3.4

- (ii) Suggest a reason for the change in the pH observed.

1

Citric acid produced OR no buffer added

- (iii) State a variable, other than temperature or pH, that should be monitored and controlled in this process.

1

Sterility/oxygen concentration OR Sugar concentration

- (iv) Explain why it was necessary to sterilise the fermenter before *A.niger* was added.

2

To kill other microbes (1)

which would compete with A niger (1)