H

JURONG JUNIOR COLLEGE
JC 2 PRELIMINARY EXAMINATIONS
Higher 2

CANDIDATE NAME

CLASS

BIOLOGY

9744/01

Paper 1 Multiple Choice

15 September 2017

1 hour

Additional Materials: Multiple Choice Answer Sheet

READ THESE INSTRUCTIONS FIRST

Write in soft pencil.
Do not use staples, paper clips, glue or correction fluid.
Write your name and class on the Answer Sheet in the spaces provided unless this has been done for you.

There are **thirty** questions on this paper. Answer **all** questions. For each question there are four possible answers **A**, **B**, **C** and **D**.
Choose the **one** you consider correct and record your choice in **soft pencil** on the separate Answer Sheet.

Read the instructions on the Answer Sheet very carefully.

Each correct answer will score one mark. A mark will not be deducted for a wrong answer.
Any rough working should be done in this booklet.
The use of an approved scientific calculator is expected, where appropriate.

1 An actively growing cell is supplied with radioactive amino acids.

Which cell component would first show an increase in radioactivity?

A Golgi body

B mitochondrion

C nucleus

D rough endoplasmic reticulum

2 When mucus is secreted from a goblet cell in the trachea, these events take place.

1 addition of carbohydrate to protein

2 fusion of the vesicle with the plasma membrane

3 secretion of a glycoprotein

4 separation of a vesicle from the Golgi body

What is the sequence in which these events take place?

A 1 → 4 → 2 → 3

B 1 → 4 → 3 → 2

C 4 → 1 → 2 → 3

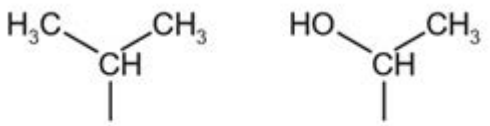
D 4 → 1 → 3 → 2

3 Which combination is found in a prokaryotic cell?

	endoplasmic reticulum	DNA	RNA	nucleus
A	X	✓	X	X
B	✓	X	X	✓
D	X	✓	✓	X
C	X	X	✓	✓

4 Threonylvaline is a dipeptide formed from the two amino acids, valine and threonine. A peptide bond forms between the amine group of valine and carboxyl group of threonine.

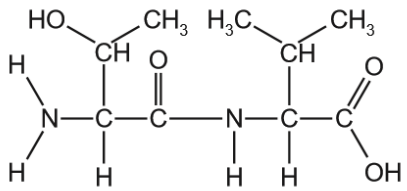
The side-chains (R groups) of the two amino acids are shown.



valine threonine

Which molecular structure is threonylvaline?

A



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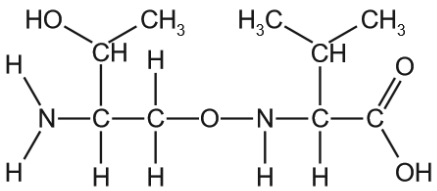

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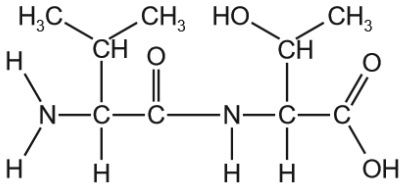

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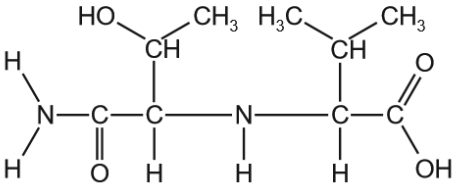


5 Which roles of the cell surface membrane are a result of the properties of the phospholipids?

1 to allow cytokinesis to occur in mitotic cell division

2 to allow entry and exit of oxygen and carbon dioxide

3 to allow the phagocytosis of a bacterium into a cell

A 1, 2 and 3

B 1 and 2 only

C 1 and 3 only

D 2 and 3 only

6 Which set of statements correctly describes haemoglobin?

	1	2	3	4
A	four polypeptide chains, each containing a haem group	iron ions can associate with oxygen forming oxyhaemoglobin	in each chain, hydrophobic R groups of amino acids point towards the centre of the molecule	at 50 % saturation, two oxygen molecules are transported by the molecule
B	polypeptide chains interact to produce a globular chain	each chain contains a haem group of amino acids surrounding an iron ion	consists of two identical alpha chains and two identical beta chains	each chain can transport an oxygen molecule
C	polypeptide chains interact to produce an almost spherical molecule	an iron ion is present within each haem group	quaternary structure has two alpha chains and two beta chains	each molecule can transport a total of four oxygen atoms
D	polypeptide chains produce a loose helical shape, which folds to form a spherical molecule	iron ions in the molecule can bind reversibly with oxygen	in each chain, hydrophobic R groups of amino acids surround the iron ion	each molecule can transport a total of eight oxygen atoms

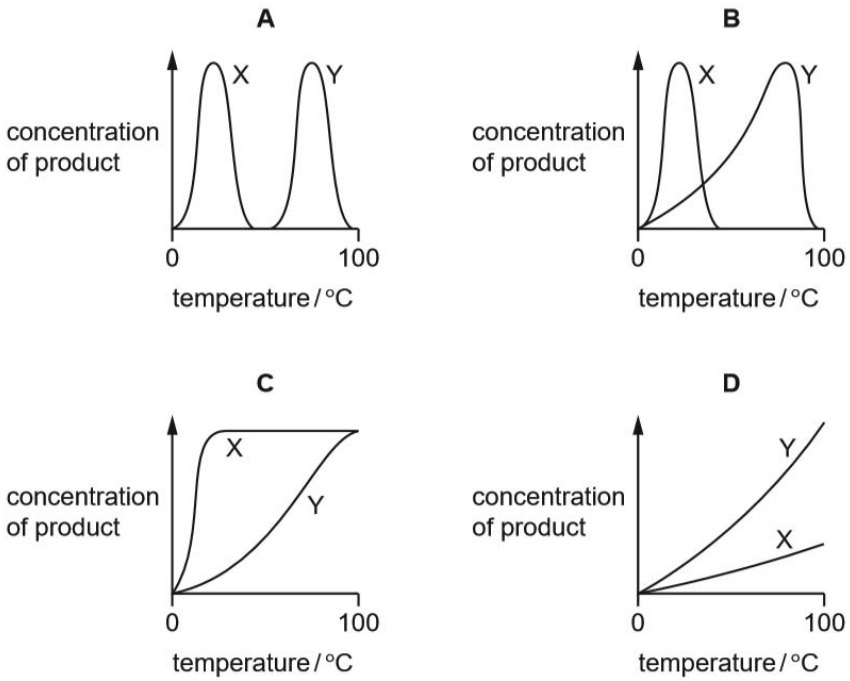
7 Two enzymes, X and Y, were used in an experiment.

Enzyme X was from bacteria that live in rivers and lakes at temperatures from 5°C to 20°C.

Enzyme Y was from bacteria that live in hot water springs at temperatures from 40°C to 85°C.

The experiment measured the concentration of product produced by each enzyme at temperatures between 0°C and 100°C after 5 minutes.

Which graph shows the results?



8 Within its own environment a particular cell line cannot be induced to produce a cell from a different cell line.

Which statement explains this?

A Genes not required for a particular cell line are methylated.

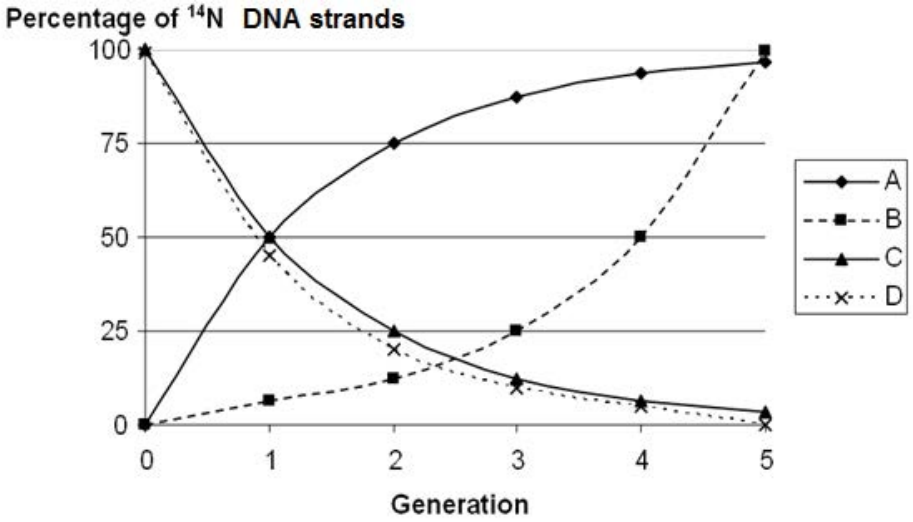
B Genes not required for a particular cell line are removed by enzymes.

C Only pre-mRNA that is required for a particular cell line is processed.

D Stem cells have only the genes required for their particular cell line.

9 Bacteria were cultured in a medium containing heavy nitrogen (^{15}N) until all DNA was labelled. These bacteria were then grown in a medium containing only normal nitrogen (^{14}N) for 5 generations. The percentage of ^{14}N DNA strands in each generation was estimated.

Which curve provides evidence that DNA replication is semi-conservative?



10 An unidentified single-stranded molecule was described as having the following features.

- complementary base pairing along some of its length
- an area that can attach to a ribosome
- a site to which a specific amino acid attaches

What is the unidentified molecule?

A ribosomal RNA

B messenger RNA

C RNA polymerase

D transfer RNA

11 Some antibacterial drugs can affect the synthesis of proteins.

antimicrobial drug	rifampicin	streptomycin	tetracycline
mode of action	binds to RNA polymerase	genetic code misread during translation	prevents binding of tRNA to ribosome

Which is the correct set of immediate effects of these drugs?

antimicrobial drug	rifampicin	streptomycin	tetracycline
A	defective protein synthesised	mRNA does not bind to ribosome	amino acids not added to growing chain
B	mRNA not synthesised	defective protein synthesised	amino acids not added to growing chain
C	mRNA not synthesised	mRNA does not bind to ribosome	transcription prevented
D	transcription prevented	defective protein synthesised	mRNA does not bind to ribosome

12 Which statement about prokaryotes and chloroplasts is correct?

A Prokaryotes and chloroplasts have circular DNA where genes carrying the code for cell walls are located.

B Prokaryotes and chloroplasts have 70S ribosomes that are the sites for translation and polypeptide synthesis.

C Prokaryotes and chloroplast have an outer membrane and a separate inner, folded membrane where ATP synthesis occurs.

D Prokaryotes and chloroplast have double-stranded linear DNA where genes carrying coded information are located.

13 Human immunodeficiency virus (HIV) is a retrovirus. After infecting a host cell, viral DNA is produced which is incorporated into the DNA of the host cell. The modified host genome now codes for the production of new HIV particles.

Which could be used as a potential treatment to slow down the spread of HIV?

1 inhibitors of restriction endonucleases

2 inhibitors of reverse transcriptase

3 restriction endonucleases

4 reverse transcriptase

A 1 and 4 only

B 1 only

C 2 and 3 only

D 2 only

14 Which of the following does not occur during bacterial conjugation?

A direct contact between donor and recipient cells

B shortening of the pilus

C unidirectional transfer of both DNA strands

D enzymatic cleavage of one strand at the origin of transfer

15 Transcriptional control in eukaryotic cells can be accomplished at several levels.

What may be involved in such control?

1 The same combination of DNA binding proteins regulate the activity of all genes.

2 Enhancers may be involved in the promotion as well as regulation of gene transcription.

3 Phosphorylation of transcriptional factors by a kinase may occur.

4 Enhancers may be some distance from the promoter sites they control.

A 1, 2, 3 and 4

B 1, 2 and 3 only

C 1, 3 and 4 only

D 2, 3 and 4 only

16 Multiple copies of a wanted DNA fragment can be made by the polymerase chain reaction (PCR).

Which description of this procedure is not correct?

A After 'n' turns of the PCR cycle, up to 2^n copies of the wanted DNA are produced.

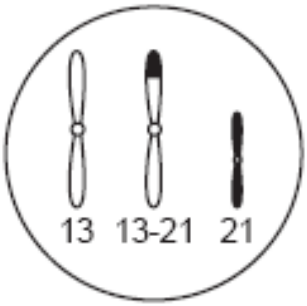
B Using a heat-stable enzyme, such as Taq polymerase, means that the enzyme does not lose activity over time.

C Using an enzyme with a high optimum temperature allows DNA polymerisation above the annealing temperature.

D Using specific primers means that only the wanted DNA is replicated.

17 Down's syndrome can be caused by a trisomy of chromosome 21, but can also result from the translocation of chromosome 21 into chromosome 13, forming a single chromosome 13-21.

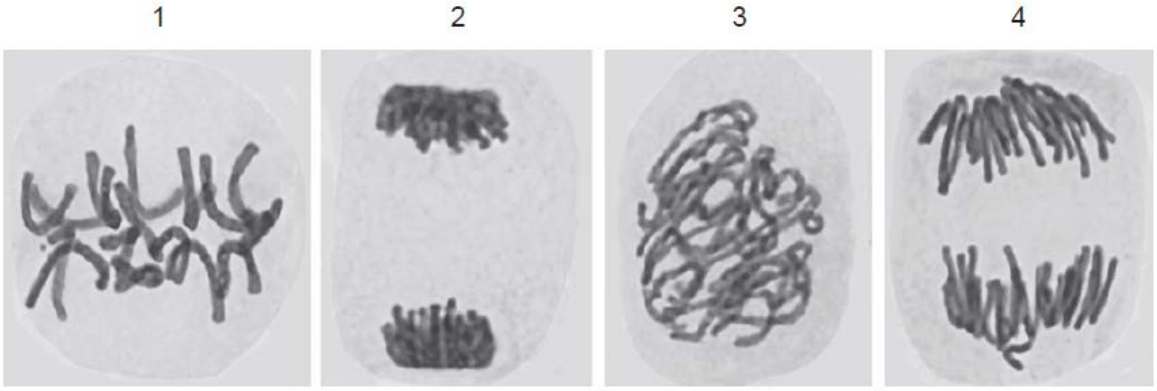
The diagram shows chromosomes 13 and 21 in the nucleus of a diploid (2n) testis cell from a phenotypically normal male carrier of a 13-21 translocation. This cell has a chromosome number of 45.



Which is **not** a likely outcome of fertilisation of normal oocytes by sperm from this male?

	chromosomes in sperm	embryo
A	13 and 21	2n =46 normal phenotype
B	13-21	2n =45 normal phenotype
C	13-21 and 21	2n =46 Down's syndrome
D	13-21 and 21	2n =47 Down's syndrome

18 The photomicrographs show cells in various stages of the cell cycle.



Which cells contain twice as many DNA molecules as a cell from the same organism after cytokinesis?

A 1, 2, 3 and 4

B 1, 2 and 4 only

C 1 and 3 only

D 2 and 4 only

19 Gene mutations in either the *BRCA1* or the *BRCA2* genes are responsible for the majority of hereditary breast cancer in humans.

The proteins produced by the two genes migrate to the nucleus where they interact with other proteins, such as those produced by the tumour suppressor gene, *p53*, and the DNA repair gene, *RAD51*.

Which combination of gene activity is most likely to result in breast cancer?

	gene		
	BRCA1 or *BRCA2*	*p53*	*RAD51*
A	✓	✓	✓
B	✓	✓	✗
C	✓	✗	✓
D	✗	✗	✗

key

✓ = gene produces normal protein

✗ = gene produces abnormal protein or no protein

20 What maximum number of different genotypes and phenotypes are possible among the children of a mother with blood group A and a father with blood group B?

	genotype	phenotype
A	2	2
B	2	4
C	4	4
D	4	2

21 A test cross resulted in these recombinants:

$$\frac{tB}{tb} \qquad \frac{Tb}{tb}$$

Which was the parental test cross?

A $\frac{TB}{tb} \times \frac{tb}{tb}$

B $\frac{TB}{tB} \times \frac{tb}{Tb}$

C $\frac{Tb}{tB} \times \frac{tb}{tb}$

D $\frac{TB}{tb} \times \frac{TB}{tb}$

22 Isolated chloroplasts, suspended in buffer solution, are often used to study the light dependent stage of photosynthesis.

During this stage, electrons (e^-) are transferred by carriers and provide energy so that a proton (H^+) gradient can be formed. Protons diffuse through membrane proteins that are linked to synthase enzymes.

Three compounds that can be added to isolated chloroplasts are:

1 DCMU, which inactivates a carrier that accepts electrons from photosystem II

2 DCPIP, which can act as a final electron acceptor

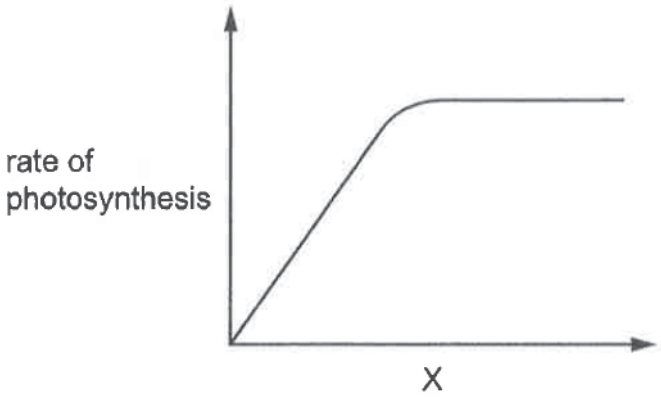
3 ammonium hydroxide solution, which absorbs protons

Which compounds, when added separately to isolated chloroplasts, would allow the light dependent stage of photosynthesis to occur and which would inhibit it?

	allow	inhibit
A	1	2 and 3
B	1 and 3	2
C	2	1 and 3
D	2 and 3	1

23 The rate of photosynthesis in pondweed was measured when one variable was changed and all others were standardised.

The graph shows the rate of photosynthesis at different values of a variable, X.



Which variables could be represented by X?

1 carbon dioxide availability

2 light intensity

3 oxygen availability

4 temperature

5 leaf area exposed to direct light

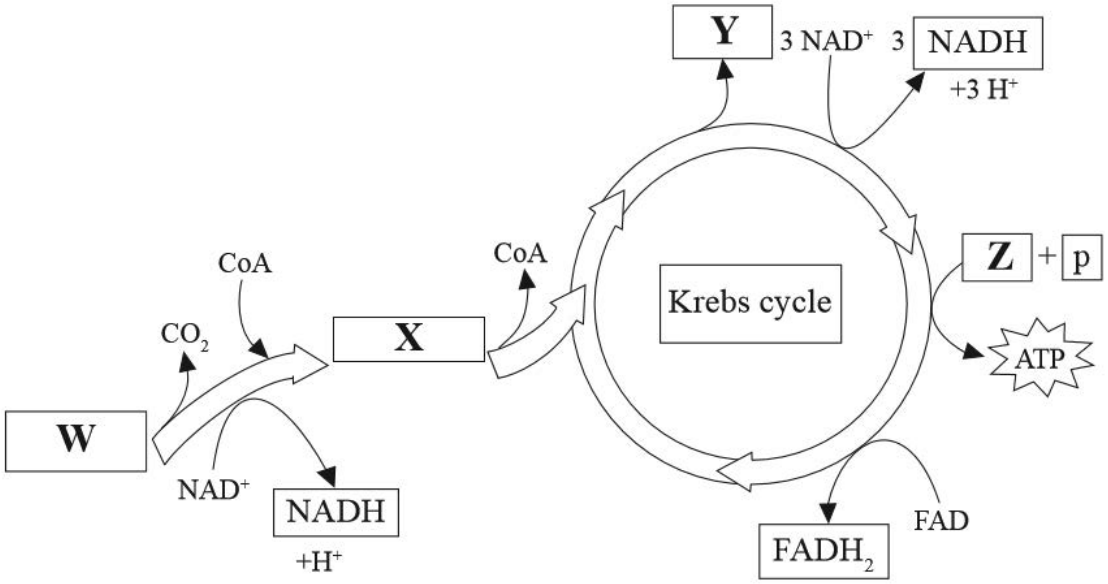
A 1, 2 and 5

B 1 and 2 only

C 2, 4 and 5

D 3 and 4

24 The diagram below shows the link reaction and stages of the Krebs cycle. Which molecules are represented by the letters W, X, Y and Z?



	W	X	Y	Z
A	acetyl CoA	carbon dioxide	ADP	pyruvate
B	pyruvate	acetyl CoA	carbon dioxide	ADP
C	ADP	carbon dioxide	acetyl CoA	pyruvate
D	acetyl CoA	pyruvate	carbon dioxide	ADP

25 The function of phosphatases in signal transduction is to

A move the phosphate group of the transduction pathway to the next molecule of a series.

B prevent a protein kinase from being reused when there is another extracellular signal.

C amplify the second messengers such as cAMP. / amplify the transduction signal so it affects multiple transducers.

D inactivate protein kinases and turn off the signal transduction.

26 Which statements are acceptable parts of Darwinian evolutionary theory?

1 Advantageous behaviour acquired during the lifetime of an individual is likely to be inherited.

2 In competition for survival, the more aggressive animals are more likely to survive.

3 Species perfectly adapted to a stable environment will continue to evolve.

4 Variation between individuals of a species is essential for evolutionary change.

A 1, 2 and 4 only

B 2 and 3 only

C 3 and 4 only

D 4 only

27 Before the settlement of California in the 1800s, the elk population was very large. By about 1900 there were only a few dozen elk left.

Owing to protection, there are now about 3000 elk living in a small number of isolated herds.

Unfortunately, some of the elk in all the herds have difficulty grazing due to a shortened lower jaw.

Which statements best explain this?

1 The early settlers only hunted elk that could graze.

2 There was a mutation affecting jaw size in one of the herds.

3 There is random mating within each herd.

4 The current elk population demonstrates a founder effect.

5 There was directional selection favouring short jaws.

A 1, 2 and 4 only

B 2, 3 and 5 only

C 2 and 5 only

D 3 and 4 only

28 Darwin's view of the process of evolution to form new species (speciation) has been reinforced by more recent discoveries in genetics and cell biology.

In this view, which sequence of events is considered most likely to lead to speciation?

A	adaptation to population	→	competition and predation leading to natural selection	→	behavioural isolation	→	sympatric speciation
B	adaptation to population	→	competition and predation leading to natural selection	→	behavioural isolation	→	allopatric speciation
C	competition and predation leading to natural selection	→	geographical isolation	→	adaptation of isolated populations	→	sympatric speciation
D	competition and predation leading to natural selection	→	geographical isolation	→	adaptation of isolated populations	→	allopatric speciation

29 Apart from the ABO blood groups, humans can also be Rhesus positive or Rhesus negative.

People with the Rhesus antigen are Rhesus positive. When a Rhesus negative person is given Rhesus positive blood in a transfusion there is no problem. However, a second transfusion of Rhesus positive blood to this Rhesus negative person will result in a reaction between the two types of blood.

Which statements explain this?

1 A Rhesus negative person naturally has anti-Rhesus antibodies.

2 Exposure to Rhesus antigen causes anti-Rhesus antibody production.

3 B-cells make anti-Rhesus antibodies.

4 Anti-Rhesus antibody production begins after the second exposure.

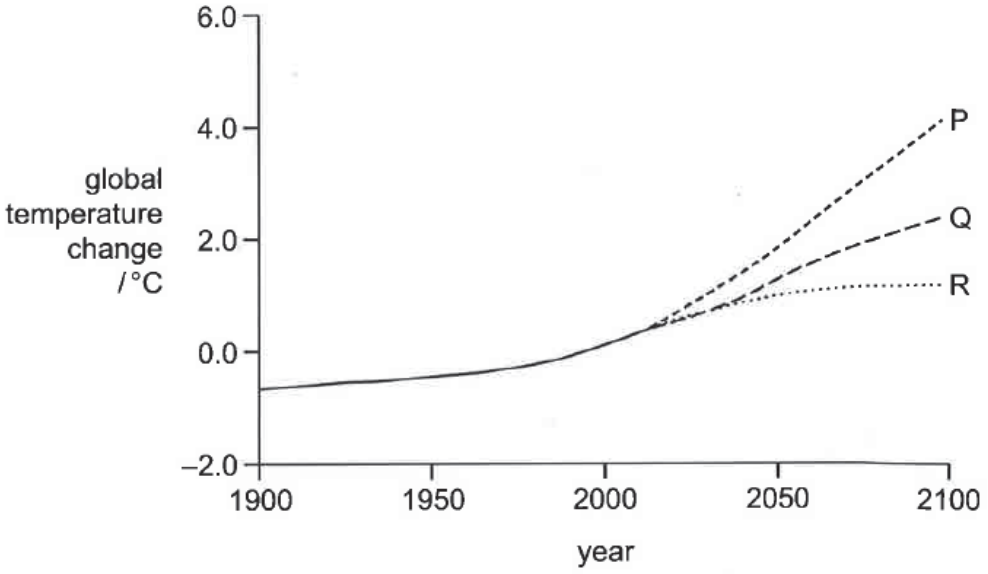
A 1, 2, 3 and 4

B 1 and 2 only

C 2 and 3 only

D 3 and 4 only

30 The graph shows the predicted change in global temperatures using three different models, P, Q and R. Model Q assumes that no new factors act to influence the rate of climate change.



The predictions based on models P and R can be explained using some of the following statements.

1 An increased global temperature and reduced rainfall will lead to an increase in forest fires.

2 Permanently frozen soil and sediment in the Arctic will begin to thaw as global temperatures rise.

3 Rising sea temperatures will cause increased growth of photosynthetic algae.

4 Rising sea temperatures will reduce the solubility of greenhouse gases in the oceans.

Which of these statements support predictions P and R?

	statements that support prediction P	statements that support prediction R
A	1, 2 and 4	3
B	1 and 3	2 and 4
C	2	1, 3 and 4
D	3 and 4	1 and 2

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JURONG JUNIOR COLLEGE
JC 2 PRELIMINARY EXAMINATIONS
Higher 2

CANDIDATE NAME

CLASS

BIOLOGY 9744/02

Paper 2 Structured Questions **25 August 2017**

Candidates answer on the Question Paper. **2 hours**

No Additional Materials are required.

READ THESE INSTRUCTIONS FIRST

Write your name and class in the spaces at the top of this page.
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.

Answer **all** questions in the spaces provided on the Question Paper.

The use of an approved scientific calculator is expected, where appropriate.
You may lose marks if you do not show your working or if you do not use appropriate units.

The number of marks is given in brackets [ ] at the end of each question or part question.

For Examiner's Use	
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7	
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Total	

This document consists of **26** printed pages and **4** blank pages.

[Turn over

Answer **all** questions.

1 The plasma membrane Ca^{2+} ATPase (PMCA) is a vital transport protein that regulates the amount of Ca^{2+} within eukaryotic cells. In humans, there is a very large transmembrane electrochemical gradient of Ca^{2+} driving the entry of Ca^{2+} into cells, yet low intracellular concentrations of Ca^{2+} are maintained by PMCA.

Fig. 1.1 shows two conformations that PMCA interconverts between, depending on whether Ca^{2+} is bound.

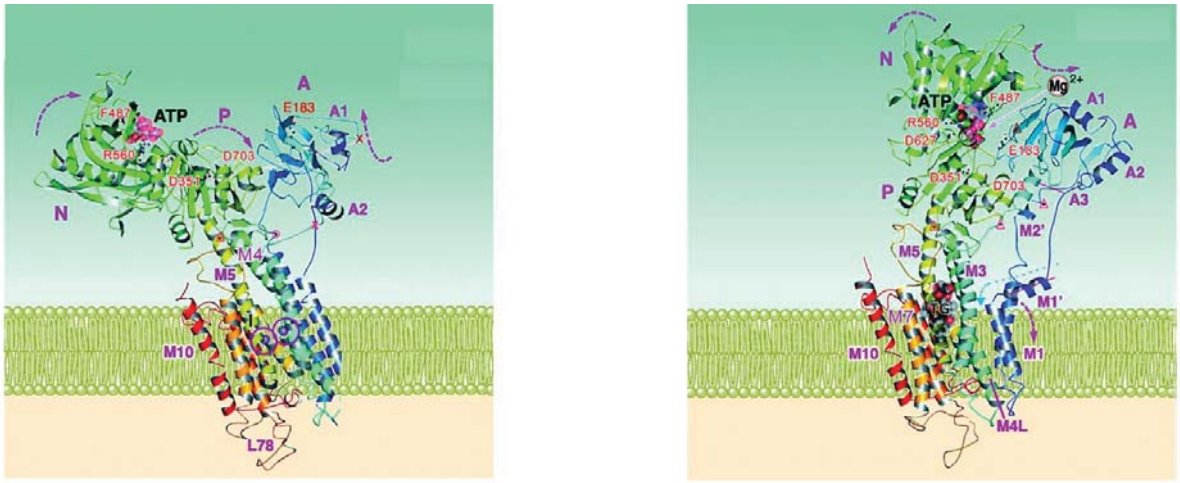

Fig. 1.1

(a) Explain why Ca^{2+} cannot freely cross the plasma membrane. [2]

(b) Describe how low intracellular concentrations of Ca^{2+} are maintained by PMCA. [3]

Various forms of PMCA are expressed in different cell types, including erythrocytes (red blood cells). Erythrocytes rely on Ca^{2+} dependent signalling during their differentiation from hematopoietic stem cells in the bone marrow. The process of erythropoiesis (production of mature red blood cells) is illustrated in Fig. 1.2.

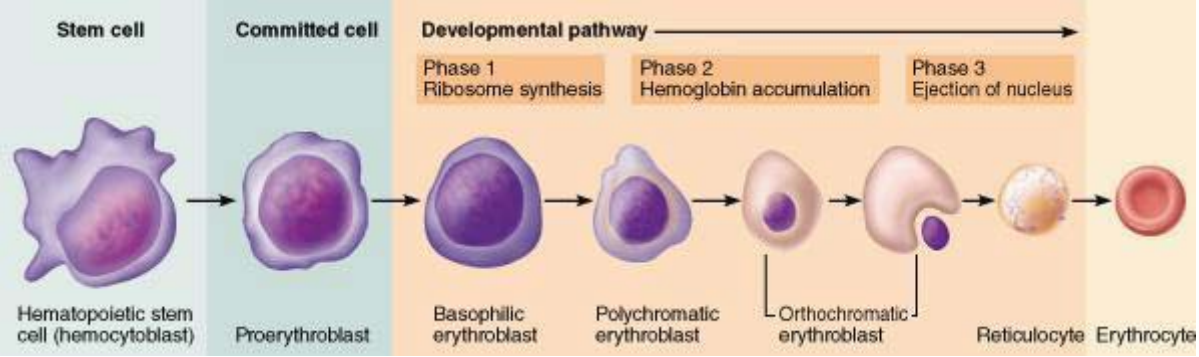

Fig. 1.2

(c) Describe the potency of these hematopoietic stem cells and explain their normal functions. [4]

In addition to the loss of the nucleus, cellular organelles such as the endoplasmic reticulum, Golgi apparatus and mitochondria are also lost from the reticulocytes shown in Fig. 1.2.

(d) Outline the structure of the endoplasmic reticulum and Golgi apparatus in typical eukaryotic cells. [2]

(e) Since mature mammalian erythrocytes lack mitochondria, suggest how these cells derive energy from glucose. [1]

_____

_____

[Total: 12]

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2 Fig. 2.1 shows two animal cells in different stages of the mitotic cell cycle.

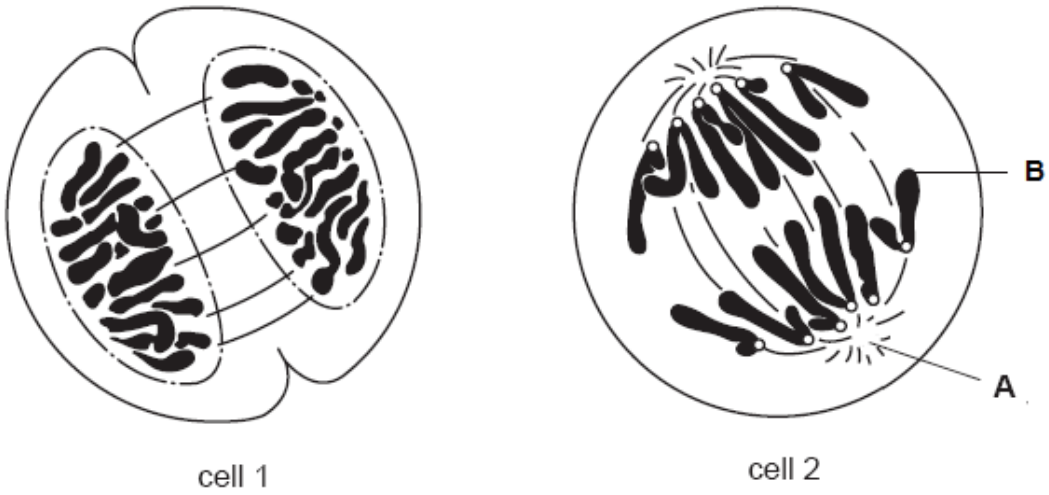

Fig. 2.1

(a) With reference to cell 1,

(i) identify the stage of nuclear division taking place. [1]

(ii) describe the events occurring at this stage. [2]

(b) A pair of rod-like structures can be found in region A. Outline the roles of these structures during mitosis in animal cells. [2]

(c) Region B of the chromatid contains non-coding repetitive nucleotide sequences.

(i) Account for the progressive shortening of these sequences after repeated rounds of DNA replication. [2]

(ii) Describe two functions of these non-coding DNA in eukaryotes. [2]

(d) In 2006, Yamanaka and his colleagues demonstrated in an experiment with mice that induced pluripotent stem (iPS) cells could be produced by genetically reprogramming fully differentiated adult cells. There has been evidence to suggest that these iPS cells exhibit high telomerase reverse transcriptase (TERT) activity and are capable of dividing indefinitely.

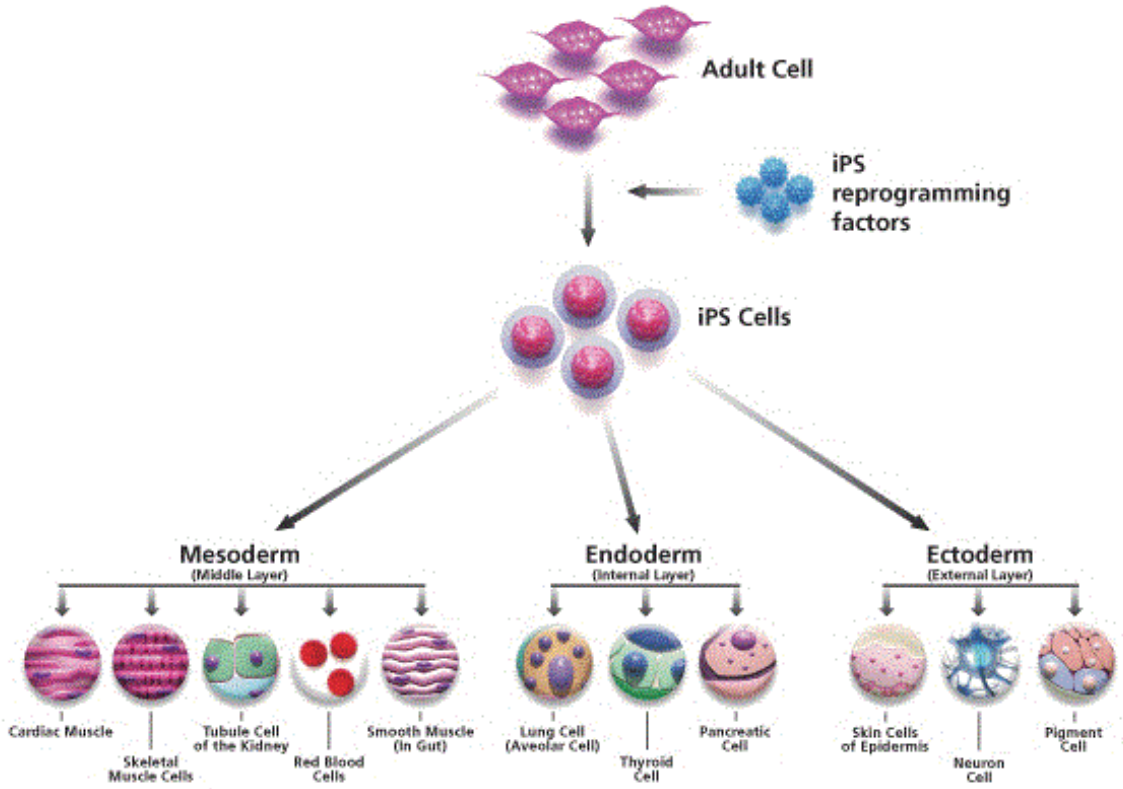

Fig. 2.2

(i) Discuss the role of TERT in enabling the iPS cells to divide indefinitely. [2]

(ii) Suggest an advantage of using iPS cells in research and medical applications. [1]

[Total: 12]

3 Antibiotic resistance is rising to dangerously high levels in all parts of the world. A growing list of infections – such as pneumonia, tuberculosis, blood poisoning and gonorrhoea – are becoming harder, and sometimes impossible, to treat as antibiotics become less effective.

Some bacteria are naturally resistant to certain types of antibiotics. However, bacteria may also become resistant either by a genetic mutation or by acquiring resistance from another bacterium.

(a) Outline the process of how a bacterium is able to acquire resistance from another bacterium. [3]

More than 2 million Americans each year are infected by antibiotic-resistant bacteria, and at least 23,000 die annually from those infections. Antibiotic-resistant bacteria have become a global health crisis and alternative treatments such as Phage Therapy are being considered for combating bacterial infections.

Phage Therapy involves the targeted application of bacteriophages that, upon encounter with specific pathogenic bacteria, can infect and kill them. Phages are currently being used therapeutically to treat bacterial infections that do not respond to conventional antibiotics.

Fig. 3.1 is an electron micrograph showing a phage infecting a bacterium during Phage Therapy.

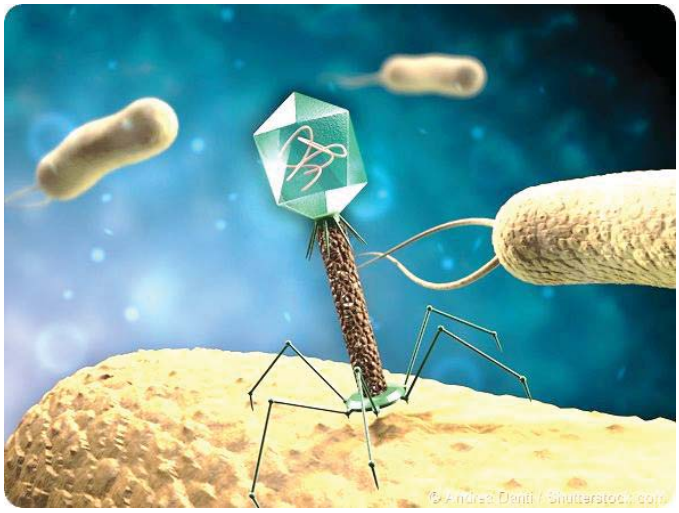

Fig. 3.1

(b) Suggest the reproductive cycle of a phage used for Phage Therapy. [1]

(c) Describe how a structural feature of the phage allows for targeted application to specific pathogenic bacteria. [2]

(d) Explain how the use of phages can prevent the spread of bacterial infection. [2]

(e) Suggest characteristics of phages that make them attractive therapeutic agents. [2]

[Total: 10]

4 Sickle cell anaemia is a recessive genetic disease caused by a mutation that commonly occurs in the DNA, resulting in hydrophobic valine replacing hydrophilic glutamic acid at the 6th amino acid position of the β chain.

(a) State the type of mutation that commonly occurs to result in sickle cell anaemia. [1]

(b) Describe the effects of this change in amino acids on the red blood cells of an individual with the disease. [4]

To detect if individuals are afflicted with sickle cell anaemia, restriction fragment length polymorphism (RFLP) analysis can be carried out using gel electrophoresis and Southern Blotting. Restriction enzymes are used to digest the DNA before RFLP analysis and the mutation removes a recognition site of the restriction enzyme *MstII,* as shown in Fig. 4.1. The enzyme's recognition sites on the normal allele and the mutant allele are shown by arrows.

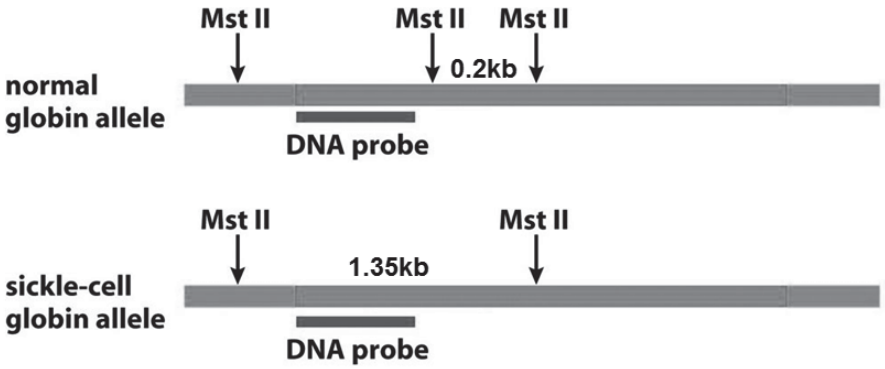

Fig. 4.1

(c) Draw, on Fig. 4.2, the expected band patterns produced by DNA from individuals with sickle cell anaemia. [1]

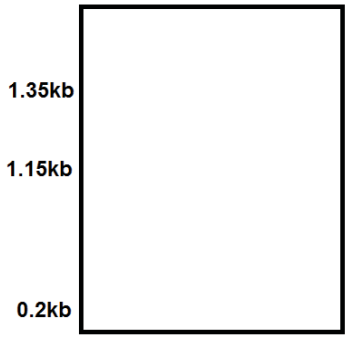

Fig. 4.2

(d) Suggest why it is necessary to carry out Southern Blotting after gel electrophoresis. [2]

(e) Outline the role of the DNA probe in Southern Blotting. [1]

[Total: 9]

5 Fig. 5.1 shows awned and awnless rice strains. A long awn is one of the distinct morphological features of wild rice species. It is a long needle-like appendage that is thought to aid in seed dispersal and prevent predation by animals.

The genes *DROOPING LEAF (DL)* and *OsETTIN2 (OsETT2)* are involved in awn formation. Genetic analysis experiments indicate that *DL* and *OsETT2* act independently in awn formation.

Fig. 5.1

A cross between pure-breeding awned and awnless strains produced awned plants in F1.

The F1 plants were then self-pollinated.

In the F2 generation, 658 awned plants and 48 awnless plants were produced.

This control of awn development is an example of epistasis resulting in a ratio that is close to 15:1.

(a) Define the term *locus*. [2]

(b) Explain the term epistasis in this context. [3]

(c) Use the symbols A, a and B, b to draw a genetic diagram to explain the results shown in the F2 generation. [4]

A chi-squared test was carried out on the results of the second cross.

Table 5.1

phenotype	observed number (O)	expected number (E)	$\frac{(O-E)^2}{E}$
awn	658		
awnless	48		
total number	706	706	x^2 =

(d) Complete the five missing values in Table 5.1. [3]

(e) Table 5.2 shows part of the table of probabilities for the chi-squared test.

Table 5.2

degrees of freedom	probability								
	0.995	0.975	0.9	0.5	0.1	0.05	0.025	0.01	0.005
1	.000	.000	0.016	0.455	2.706	3.841	5.024	6.635	7.879

Use Table 5.2 and your calculated value for the chi-squared test to find the probability that the observed ratio of phenotype does not deviate significantly from the expected ratio. [1]

(f) State what conclusions may be drawn from the probability found in (e). [2]

[Total: 15]

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6 Fig. 6.1 shows some stages in mammalian respiration.

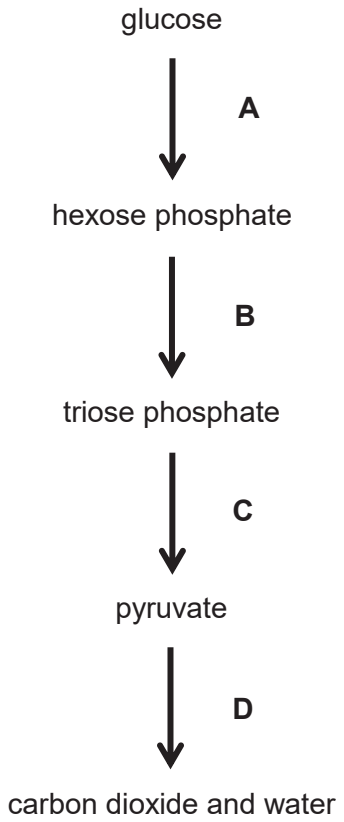

Fig. 6.1

(a) Name the processes taking place during Stage D and state precisely where they occur. [3]

(b) Intermediates produced at the end of Stages B and C are important in the conversion of carbohydrates to lipids such as triglycerides. Some of the triose phosphate can be converted into glycerol-3-phosphate, while pyruvate can undergo further reactions to form intermediates required for the synthesis of fatty acids.

(i) Describe the formation of triglycerides. [3]

(ii) State two roles of triglycerides in living organisms. [2]

(c) The first reaction in Stage A is catalysed by the enzyme hexokinase. It has been observed that hexokinase is bound to the outer mitochondrial membrane in muscle cells which undergo high rates of glycolysis.

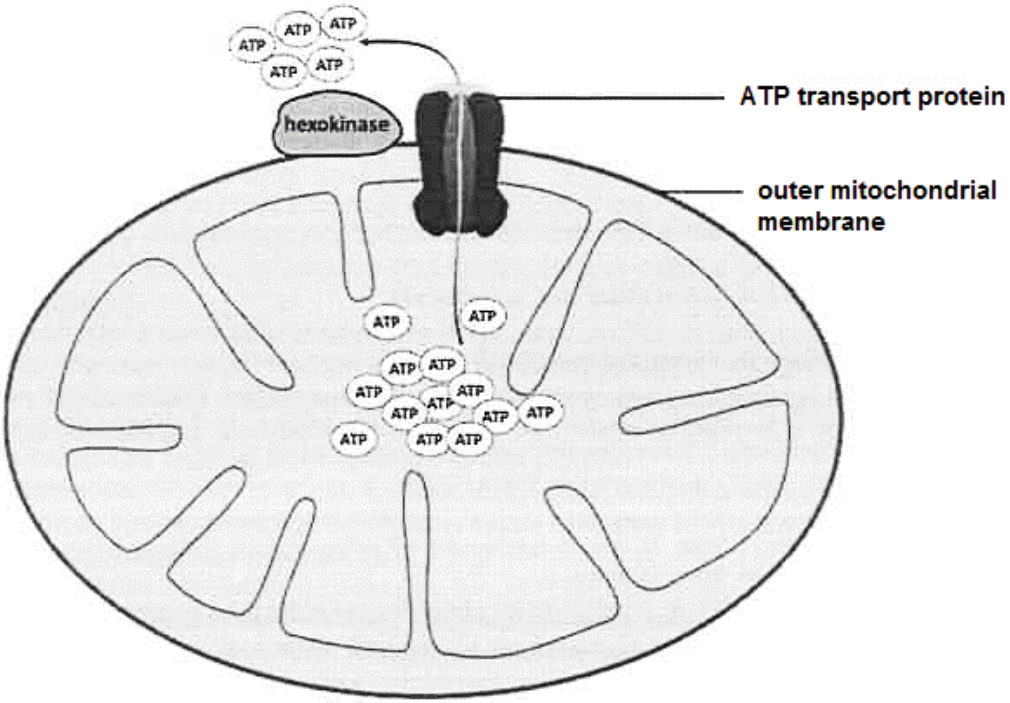

Fig. 6.2

With reference to the role of mitochondria and Fig. 6.2, suggest how the association of hexokinase with mitochondria can lead to high rates of glycolysis. [2]

Fig. 6.3 shows an electron micrograph of a mitochondrion.

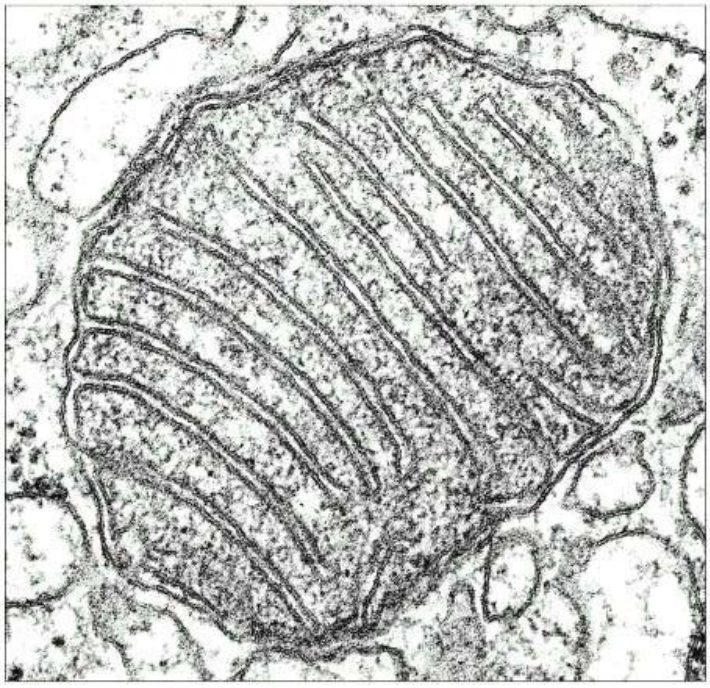

Fig. 6.3

(d) With reference to features visible in Fig. 6.3, outline how the structure of the mitochondrion is adapted for its function. [2]

[Total: 12]

7 Fig. 7.1 shows a flower of *Lilium polyphyllum*, a lily that grows in the Himalayan mountains. This species is cross-pollinated by insects.

Fig. 7.1

(a) Plants of this species that grow at low altitudes produce flowers 60 days before the plants of the same species that grow at high altitudes. Scientists think that plants of *L. polyphyllum* growing at high altitudes may evolve into a new species.

Explain how natural selection could lead to the evolution of a new species of lily. [5]

(b) In order for natural selection to occur a population must show phenotypic variation. Explain why variation is important in natural selection. [2]

(c) Fungi were often classified as different species according to their visible reproductive structures. *Penicillium dodgei* and *Eupenicillium brefeldianum* were classified as different species because they had different types of spores.

However, recently it was recognised that the spores of *P. dodgei* were asexual spores, while those of *E. brefeldianum* were sexual spores. A comparison of the DNA of these two fungi shows that they are the same species. This fungus is now known as *Penicillium brefeldianum*.

Outline how DNA analysis can show that *P. dodgei* and *E. brefeldianum* are the same species. [2]

(d) Describe the advantages of using DNA analysis in determining homology between *P. dodgei* and *E. brefeldianum*. [3]

[Total: 12]

8 Fig. 8.1 shows part of the immune response to the first infection by a bacterial pathogen that has entered the body through the lining of a bronchiole. J and K are stages in the immune response.

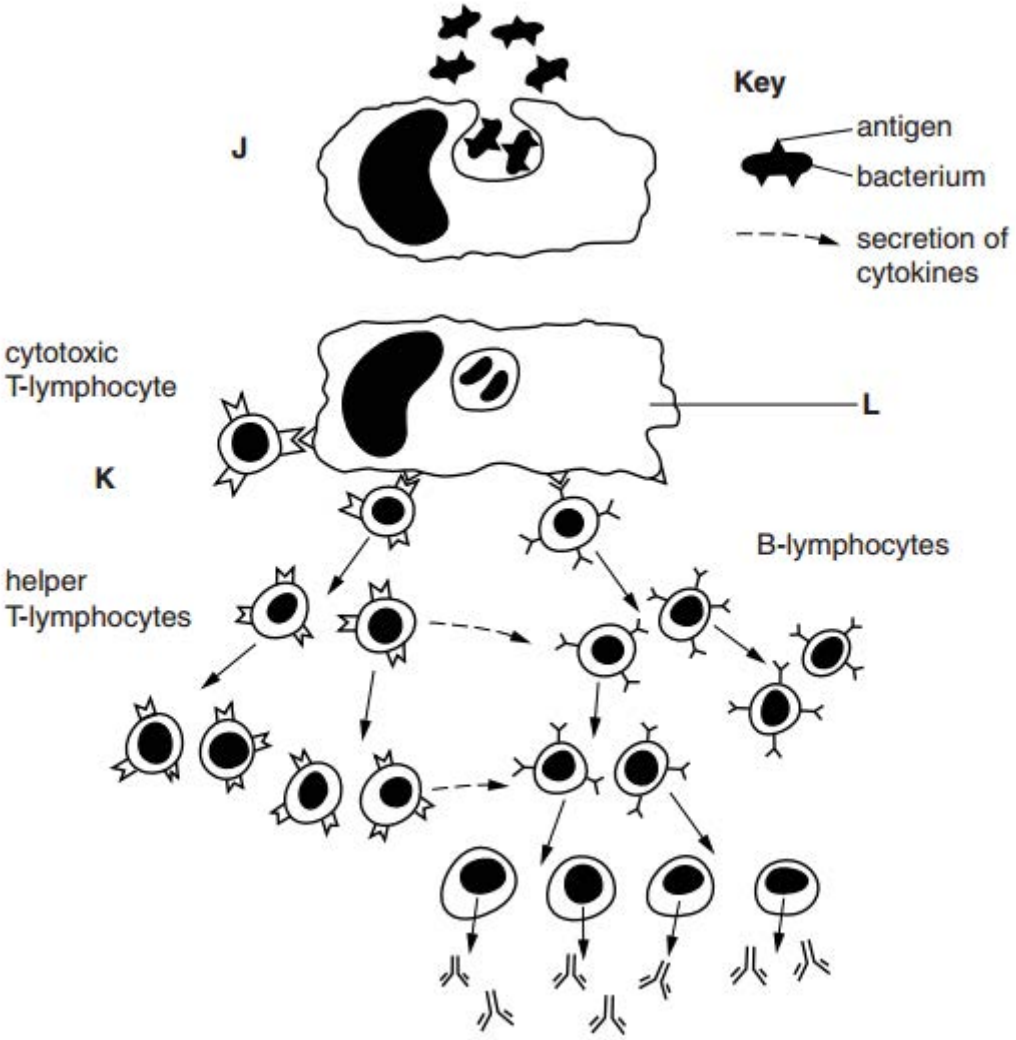

Fig. 8.1

(a) (i) State the process happening at stage J. [1]

(ii) Explain the role of cell L at stage K in the immune response. [2]

(b) With reference to Fig. 8.1, explain how the response to a second infection by this bacterial pathogen differs from the first. [3]

B-lymphocytes have antibodies located on their external surface. When B-lymphocytes become plasma cells they then secrete antibodies.

Fig. 8.2 shows how the enzyme papain digests an antibody to obtain three fragments.

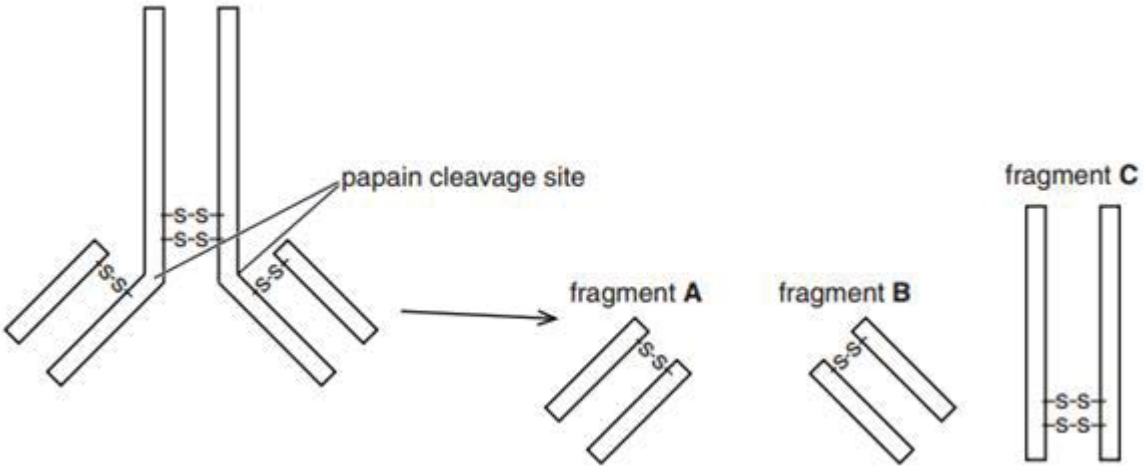

Fig. 8.2

(c) The three fragments, A, B and C still retain their ability to function.

State the function of:

(i) fragments A and B [1]

(ii) fragment C. [1]

(d) There are various ways in which the effectiveness of immune responses can be reduced.

Suggest how each of the following reduces the effectiveness of an immune response.

(i) Some pathogens are covered in cell surface membranes from their host. [1]

(ii) B-lymphocytes do not mature properly and do not recognise any antigens. [1]

[Total: 10]

9 Reef-building corals are marine invertebrates closely related to jellyfishes and are found in shallow, clear tropical seas. The corals secrete an exoskeleton of calcium carbonate that becomes the underlying structure of the coral reef.

Zooxanthellae are a group of unicellular photosynthetic algae that live inside the cells of reef-building corals. The relationship is beneficial to both the zooxanthellae and the coral.

(a) Evidence shows that the relationship between zooxanthellae and reef-building corals has evolved by free-living algae invading corals that did not contain algae. [1]

(i) Corals that do not need zooxanthellae can live at a greater depth that reef-building corals.
Explain why. [3]

(ii) Suggest how the zooxanthellae may benefit in two ways from their association with the corals. [2]

Under conditions of environmental stress, the relationship between the reef-building corals and zooxanthellae can break down. Loss of zooxanthellae and the subsequent whitening that occurs, as shown in Fig. 9.1, is known as coral bleaching. Coral bleaching can lead to the death of the coral.

Fig. 9.1

(b) State one reason why permanent loss of zooxanthellae can lead to death of the coral. [1]

(c) One type of environmental stress that can cause coral bleaching is an increase in sea temperature.

Suggest why areas of sea with reef-building corals are particularly susceptible to increased temperature as a result of global climate change. [2]

[Total: 8]

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JURONG JUNIOR COLLEGE
JC 2 PRELIMINARY EXAMINATIONS
Higher 2

CANDIDATE NAME

CLASS

INDEX NUMBER

BIOLOGY 9744/03

Paper 3 Long Structured and Free-response Questions **11 September 2017**

Candidates answer on the Question Paper. **2 hours**

No Additional Materials are required.

READ THESE INSTRUCTIONS FIRST

Write your class, index number and name in the spaces at the top of this page.
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.

Section A
Answer **all** questions in the spaces provided on the Question Paper.

Section B
Answer any **one** question in the spaces provided on the Question Paper.
Circle the question number of the question attempted.

The use of an approved scientific calculator is expected, where appropriate.
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	
Section A	
1	
2	
3	
Section B	
4 / 5	
Total	

This document consists of **19** printed pages and **3** blank pages.

[Turn over

Section A

Answer **all** the questions in this section.

1 The *lac* operon is an operon required for the uptake and metabolism of lactose in *Escherichia coli* and many other bacteria. Glucose is the preferred carbon source for most bacteria, as glucose requires fewer steps and less energy to break down than lactose. However, if lactose is the only sugar available, the *E. coli* uses it as an energy source and the *lac* operon allows for the effective digestion of lactose when glucose is not available.

To use lactose, the bacteria must express the *lac* operon genes, which encode key enzymes for lactose uptake and metabolism. Fig. 1.1 shows the results of an experiment carried out to determine the effects of adding lactose on the expression of some of the genes involved in the breakdown of lactose.

The initial gene expression was measured by determining the mRNA produced at time 0. This was taken as 10% (black bars). All the other values are relative to this initial value taken every 30 seconds over the next 2 minutes.

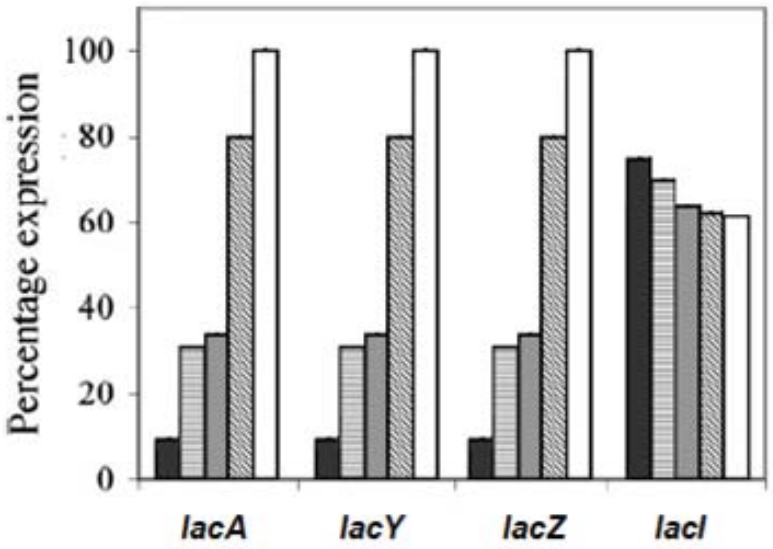

Fig. 1.1

(a) Suggest why operons are necessary in bacteria. [2]

(b) Using data from *lacA*, *lacY* and *lacZ* in Fig. 1.1 and your knowledge of how different types of operons are regulated, explain how lactose is able to control the expression of these genes. [4]

It has been suggested that not all genes involved in lactose hydrolysis are organised into one single operon.

(c) Use evidence from Fig. 1.1 to support the statement above. [3]

(d) A series of mutations was introduced into the *lac* operon, resulting in the inversion of the operator and the promoter regions.

Suggest the effect on the transcription of the *lac* genes when lactose is absent. [2]

Owing to *E. coli's* rapid growth rate, *E. coli* has been an expression host of choice in the biotechnology industry for large-scale production of anti-freeze proteins (AFPs). AFPs is a class of polypeptides that help to stop ice forming inside the Arctic and Antarctic fishes thus permitting their survival in sub-zero environments.

In the Arctic and Antarctic, environmental temperatures can reach low to freezing levels. These fishes indigenous to these habitats are presented with potential desiccation, which can lead to potentially detrimental challenges such as decreased enzymatic rates and freezing. Besides hindering cellular processes, sub-zero temperatures induce ice crystals formation, which can lead to cell death by rupturing cells either physically or through osmotic pressure changes.

Commercially, there appears to be countless applications for AFPs:

- as additives to frozen foods to lengthen the shelf life
- for incorporation with the genome of the raw foods to retard ice crystal growth
- to prevent damage to agricultural crops by increasing freeze tolerance of crop plants and extending the harvest season in cooler climates
- for introduction into ice cream and yogurt products to allow the production of very creamy, dense, reduced fat ice cream with fewer additives.

(e) Outline how the genome of *E. coli* and the genome of the fish are similar and how they are different. [4]

(f) Anti-freeze glycoprotein (AFGP) is one type of anti-freeze protein. Messenger RNA coding for AFGP is translated at a ribosome to produce a polypeptide. Describe how this polypeptide is then processed to make AFGP. [4]

Some fish produce another anti-freeze protein, called AFP II. The tissues of these fish were tested for the presence of AFP II and the mRNA coding for AFP II. The results are shown in Table 1.1.

Table 1.1

molecule	present in
AFP II protein	all tissues
AFP II mRNA	liver tissue only

(g) Explain the distribution of the AFP II protein and AFP II mRNA. [4]

(h) With reference to named examples, describe the roles performed by proteins involved in transport in fishes. [2]

Table 1.2 shows Earth's ice ages over the last 850 million years.

Table 1.2

Ice age	Time / millions of years ago
Quaternary	0 to 2.6
Karoo	260 to 360
Andean-Saharan	420 to 460
Cryogenian	630 to 850

Fig. 1.2 shows how the number of families of fishes has changed over time.

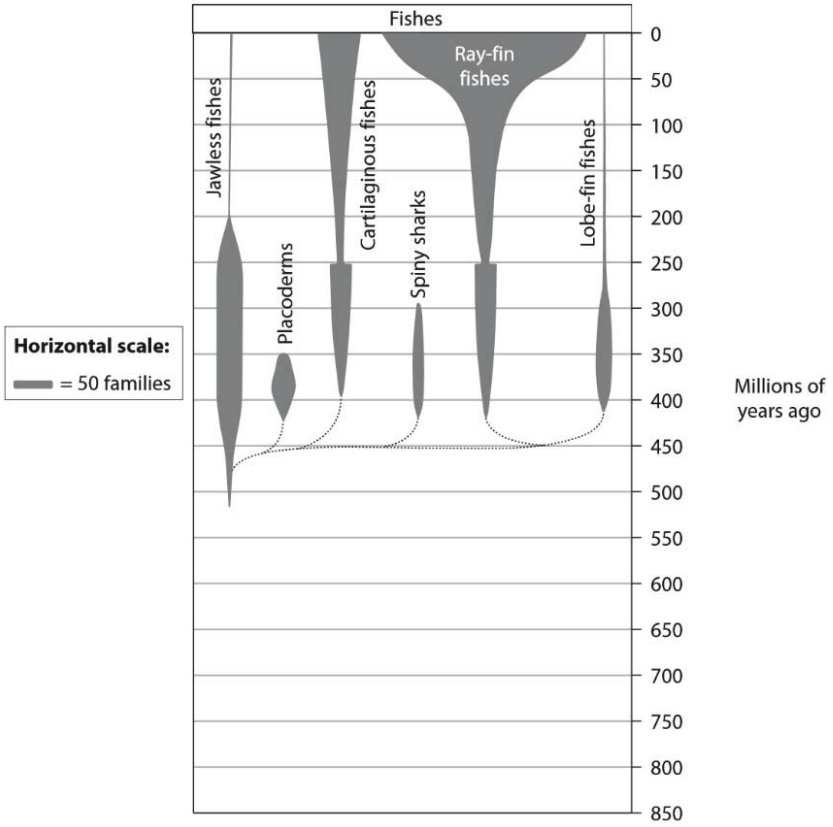

Fig. 1.2

(i) Many different types of AFPs are produced by ray-fin fishes. Analyse the data to explain when these ray-fin fishes are likely to have evolved the ability to produce AFPs. [2]

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[Total: 27]

2 Measles is a highly contagious, serious disease caused by *Morbillivirus*, a single-stranded enveloped RNA virus. Envelope glycoproteins mediate transmission of the virus into host cells in the human respiratory tract. Once inside the host cell, the viral RNA genome is transcribed into mRNA, which undergoes translation to manufacture viral proteins. These viral proteins function to form capsid proteins for new viruses which eventually leave the host cell.

(a) With reference to the information given, outline how viruses challenge the concept of what is considered living. [2]

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In the 1980s, measles caused an estimated 2.6 million deaths each year, and the disease remains one of the leading causes of death among young children globally.

The number of cases of measles is reported to the World Health Organisation (WHO) by countries throughout the world so that global data is collected.

Fig. 2.1 shows the global data collected between January 2008 and December 2012.

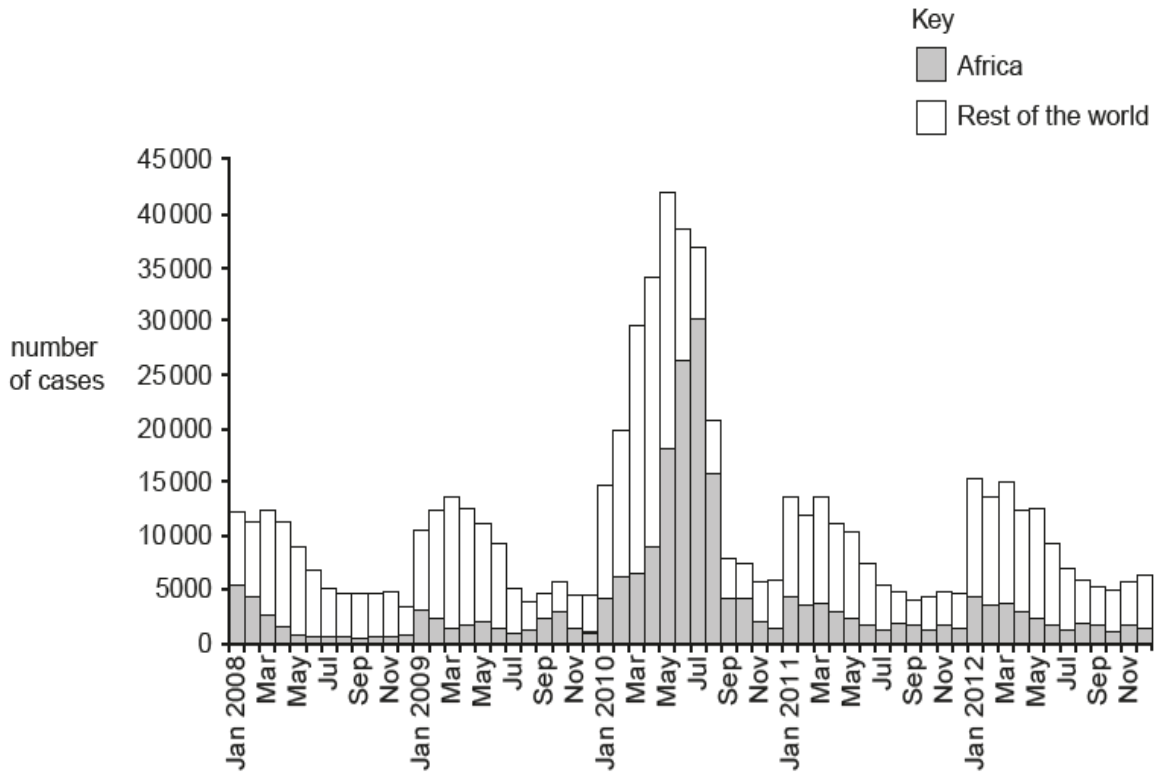

Fig. 2.1

(b) Use the data in Fig. 2.1 to describe the pattern shown in the number of cases of measles reported to the WHO between January 2008 and December 2012. [3]

(c) Routine measles vaccination for children, combined with mass immunisation campaigns in countries with high case and death rates, are key public health strategies to reduce global measles deaths. By 2016, about 85% of the world's children received one dose of the measles vaccine by their first birthday, and the global push to improve vaccine coverage resulted in a 79% reduction in deaths.

(i) State precisely the type of immunity gained by receiving a measles vaccine. [1]

(ii) Outline one benefit of vaccination. [1]

(iii) Outline one risk of vaccination. [1]

(d) Unlike measles for which an effective vaccine has been developed, it has been extremely difficult to design an effective vaccine against malaria. Malaria is a disease caused by the parasite *Plasmodium falciparum*. *P. falciparum* multiplies in liver cells of the host before emerging after 9-30 days wrapped in the liver cell surface membrane. They enter red blood cells, multiply and then cause rupture of the host cells, resulting in the release of more parasites every 36-48 hours, in a manner that has some similarity to that of viruses.

Use the information given and your own knowledge to suggest why it has been extremely difficult to design an effective vaccine against malaria. [2]

(e) Another infectious disease, Tuberculosis (TB), is one of the top ten causes of death worldwide. Name the bacterium that causes TB and describe how TB is transmitted. [3]

[Total: 13]

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3 Dengue fever is a disease spread by a particular species of mosquito, *Aedes aegypti.* The incidence of this disease and the numbers of this species of mosquito have increased dramatically in recent years, spreading beyond the tropics. This has been attributed to global warming.

(a) Explain how global warming has resulted in the spread of dengue beyond the tropics. [2]

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In an attempt to reduce the numbers of *A. aegypti*, male mosquitoes infected with the *Wolbachia* bacteria have been produced and released into the wild to mate with females. *Wolbachia* naturally occurs in up to 60% of all insect species, but not in *A. aegypti. Wolbachia* induces a conditional sterility that occurs within the mosquitoes due to cytoplasmic incompatibility, shown in Fig. 3.1, a concept first introduced in a paper published by Dr. Hannes Laven in 1967.

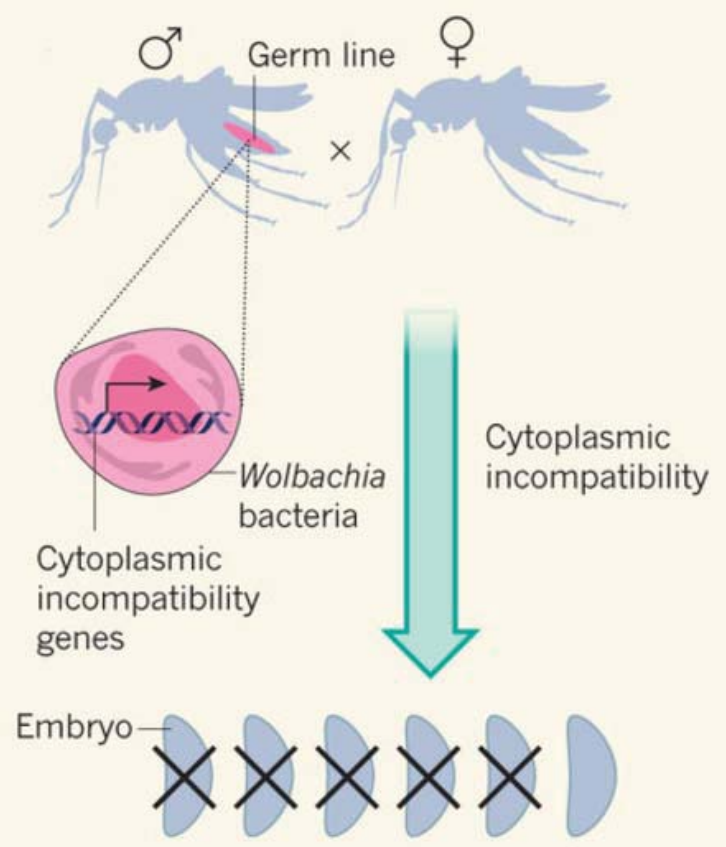

Fig. 3.1

(b) The cytoplasmic incompatibility genes are DNA in nature. DNA is a double helix consisting of two polynucleotide strands held together by phosphodiester bonds between the adjacent nucleotides. Each strand contains a sugar-phosphate backbone and hydrogen bonds are formed between the complementary strands via complementary base pairing.

Describe two other structural features of DNA. [2]

There are three possible matings between the male and female mosquitoes in the wild as shown in Table 3.1.

Table 3.1

	male	female	results
cross 1	infected	uninfected	lay eggs that are not viable and do not hatch
cross 2	infected	infected	infected offspring
cross 3	uninfected	infected	infected offspring

Embryonic development aborts when sperm from an infected male fertilises an uninfected egg and the paternal genome does not contribute to the development of the embryos that are not viable.

(c) Based on the information provided and Cross 1, suggest how *Wolbachia* induces sterility. [1]

In 1970, Erich Jost repeated Laven's earlier work.

(d) Explain why repeating the work of others is an important part of science research. [2]

In 2016, hundred thousands of male mosquitos with *Wolbachia* were released at 3 selected sites in Singapore: Braddell Heights, Nee Soon East, and Tampines West.

(e) State why releasing such large numbers of male mosquitoes did not immediately increase the risk of transmission of dengue fever in these estates. [1]

_____

_____

Another method employed in Australia involves the release of both male and female mosquitoes with *Wolbachia* into the wild. An advantage of this method is that there is no need for further releases of mosquitoes with *Wolbachia.*

(f) With reference to Table 3.1, explain why there is no need for further releases with this method. [2]

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_____

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[Total: 10]

Section B

Answer **one** question in this section.

Write your answers on the lined paper provided at the end of this Question Paper.

Your answers should be illustrated by large, clearly labelled diagrams, where appropriate.

Your answers must be in continuous prose, where appropriate.

Your answers must be set out in parts **(a)**, **(b)** etc., as indicated in the question.

4 **(a)** Describe the reproductive cycle of the influenza virus and explain how new strains of the virus may arise as a result of mutation. [13]

(b) Describe the process of transduction and its advantages to prokaryotes. [12]

[Total: 25]

5 **(a)** Describe the roles of the proteins involved in the process of DNA replication and compare the advantages of PCR with the advantages of DNA replication. [13]

(b) Outline the structure of G-protein linked receptor and describe the action of glucagon on liver cells in the regulation of blood glucose concentration. [12]

[Total: 25]

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JURONG JUNIOR COLLEGE
JC 2 PRELIMINARY EXAM
Higher 2

CANDIDATE NAME

CLASS

BIOLOGY

9744/04

Paper 4 Practical

15 August 2017

CCONFIDENTIAL INSTRUCTIONS

2 hours 30 minutes

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

This document consists of 7 printed pages.

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Question 1

Preparation list

	Apparatus/Reagents/Chemicals	Quantity per student
1	2,6-dichlorophenolindophenol (DCPIP) in a specimen tube with a cap, labelled **D**.	3 cm^3
2	Leaf extract in a specimen tube, labelled **L**, in a beaker containing ice	10 cm^3
3	Graduated 1 cm^3 Pasteur pipette	1
4	1 cm^3 syringes	2
5	6 cm × 15 cm filters, folded longitudinally to form a 'tent', of the following colours: purple, labelled **P** (approx. 425 nm) blue, labelled **B** (approx. 450 nm) green, labelled **G** (approx. 525 nm) orange, labelled **O** (approx. 625 nm) red, labelled **R** (approx. 675 nm) Filters should be labelled in one corner.	1 of each colour
6	White tile – 10 cm × 10 cm	1
7	Length of aluminium foil, large enough to wrap around the specimen tube containing the leaf extract	1 piece
8	Square piece of aluminium foil, large enough to form a lid for the specimen tube containing the leaf extract	1 piece
9	Bench lamp with 60 W filament bulb (or equivalent)	1
10	Stop clock or stopwatch	1
11	30 cm ruler	1
12	Glass rod	1
13	Safety glasses/goggles	1 pair
14	250 cm^3 beaker containing distilled water, labelled **for washing**	1
15	250 cm^3 beaker, labelled **for waste**	1
16	Paper towels	5
17	Disposable gloves	1 pair

Instructions for preparation

Buffer solution, pH 7.5

4.5g disodium hydrogenphosphate ($Na_2HPO_4.12H_2O$)
1.7g potassium dihydrogenphosphate (KH_2PO_4)
500cm^3 water

Dissolve the disodium hydrogenphosphate and potassium dihydrogenphosphate in 450cm^3 distilled or deionised water.
When the solids have completely dissolved make the volume up to 500 cm^3 with distilled or deionised water.
Check the pH of the buffer and adjust if necessary. Adding potassium dihydrogenphosphate will lower pH, adding disodium hydrogenphosphate will increase pH.

2,6-dichlorophenolindophenol (DCPIP) solution, labelled **D**

0.2 g DCPIP
0.9 g potassium chloride

Dissolve in 250 cm^3 of buffer solution.
This can be made before the examination and stored in a refrigerator.
The concentration being used by candidates does not constitute a hazard.

Extraction solution

13.7 g sucrose
0.1 g potassium chloride

Dissolve in 100 cm^3 of buffer solution.
Refrigerate until needed.

Leaf extract, labelled **L** and with hazard symbol for irritant

This should be prepared on the day on which the practical is to be carried out.

Use approximately 15 g spinach leaves (or any soft, dark green leaves) for each 100 cm^3 of the extraction solution.

Use scissors to cut the midrib and any large veins from the leaves. Cut the remaining leaf material into small pieces and add to the extraction solution. Use a liquidiser or blender to separate and disrupt the cells. Filter the leaf extract through muslin or fine mesh fabric to remove leaf debris. Store the filtrate in a beaker in a refrigerator before dispensing to candidates.

This should be provided to students in a beaker of ice.

Filters

These should be obtained from a photographic supplier and should allow light of the approximate wavelengths required to pass through without large differences in overall absorption.

Lee Filters (obtainable from www.hwarta.com) provide a range of suitable filters other than the codes stated in the preparation list.

	Specified Codes	**Examples of Alternatives**
purple	136	052, 170
blue	140	144, 117
green	139	124
orange	105	021
red	164	022

Question 2

Preparation list

Each candidate must have sole, uninterrupted use of a microscope for 1 hour 15 minutes only.

For each candidate:

- the microscope must be set up on the low-power objective lens
- the slide must not be left in the stage or n the microscope

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^3) with tap water, labelled **for washing**	1	
Beakers or container, labelled **for waste**	1	
Glass marker pen	1	
Paper towel	6	
Stop-clock or stopwatch or sight of a clock to time five minutes	1	
Microscope with: • Low-power objective lens, × 10 (equal to 16 mm or $\frac{2}{3}$") • High-power objective lens, × 40 (equal to 4 mm or $\frac{1}{6}$") • Eyepiece lens, × 10 (equal to 16 mm or $\frac{2}{3}$") • Eyepiece graticule fitted within the eyepiece and visible in focus at the same time as the specimen.	1	

- 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. **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.**

As the yeast cell suspension will froth, it should be prepared in a large container.

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

10 to 15 minutes before the candidates start Question 2 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.

Put 50 cm^3 of the 7.0% yeast suspension (that you have prep above) into a beaker or container and make up to 100 cm^3 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^3**
S1	3.5% boiled yeast cell suspension	none	at least 5
S2	3.5% active yeast cell suspension	none	at least 5
S3	mixture of S1 and S2	none	at least 5
M	1% methylene blue solution	[H] 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^3 of 3.5% boiled yeast cell suspension in a small container, labelled **S1**. For example: put 20 cm^3 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^3 of 3.5% yeast cell suspension (active) as diluted above from active 7.0% yeast suspension.

(iii) S3, at least 5 cm^3 of 3.5% yeast cell suspension made up of equal volumes of **S1** and **S2**. For example, put 10 cm^3 of S1 into a small container. Add 10 cm^3 of **S2** to the small container and mix well.

This is sufficient for 4 candidates.

(iv) **M**, at least 5 cm^3 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^3 of distilled water and making it up to 100 cm^3 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.

Question 3

No materials are required for this question.

H

JURONG JUNIOR COLLEGE
JC 2 PRELIMINARY EXAMINATIONS
Higher 2

CANDIDATE NAME

CLASS

BIOLOGY 9744/04

Paper 4 Practical **15 August 2017**

2 hours 30 minutes

Candidates answer on the Question Paper.

Additional Materials: As listed in the Confidential Instructions.

READ THESE INSTRUCTIONS FIRST

Write your name and class on all the work you hand in.
Give details of the practical shift and laboratory, where appropriate, in the boxes provided.
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.

Answer **all** questions in the spaces provided on the Question Paper.

The use of an approved scientific calculator is expected, where appropriate.
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.

Shift
Laboratory

For Examiner's Use	
1	
2	
3	
Total	

This document consists of **17** printed pages.

[Turn over

Answer **all** questions.

1 During the light dependent stage of photosynthesis, hydrogen ions and electrons are transferred to hydrogen acceptor molecules, including NADP.

DCPIP (2,6-dichlorophenolindophenol) is a blue dye, which acts as a hydrogen ion and electron acceptor. As DCPIP accepts hydrogen ions or electrons it is reduced and becomes colourless.

You are required to investigate the effect of different wavelengths of light on the rate of the light dependent stage of photosynthesis in a leaf extract containing chloroplasts.

You are provided with:
- a leaf extract in buffered solution, labelled **L**, in a beaker with ice,
- DCPIP solution, labelled **D**,
- filters that allow light of specific wavelengths to pass through, as shown in Table 1.1.

Table 1.1

colour	label	wavelength / nm
purple	P	425
blue	B	450
green	G	525
orange	O	625
red	R	675

The leaf extract is an irritant. It is recommended that you wear safety goggles/glasses and gloves.

Proceed as follows.

1. Stir the leaf extract, **L**, using the glass rod.

2. Use a syringe to draw up 0.5 cm^3 of **L**.

3. Wipe the outside of the syringe to remove any liquid.

4. Put the syringe at the centre of a white tile. This will be used as a colour standard.

5. You are required to add enough DCPIP solution, **D**, to change the colour of the remaining leaf extract, **L**. The change in colour must be sufficient to be observable in the 0.5 cm^3 sample transferred to a syringe in step **8**.
   - Using a Pasteur pipette, put about 0.5 cm^3 of DCPIP solution, **D**, into the remaining leaf extract, **L**, in the specimen tube.
   - Shake the specimen tube gently so that the colour spreads evenly.
   - Tilt the specimen tube and view the colour against a white background.
   - If there is no noticeable colour change, add DCPIP solution, **D**, drop by drop until a noticeable colour change is achieved.

6. Immediately wrap the specimen tube containing the mixture of **L** and **D** in foil. Cover the specimen tube with a foil lid, as shown in Fig. 1.1. This should be easy to remove to obtain the mixture of **L** and **D**. Put back the covered specimen tube containing the mixture of **L** and **D** in the beaker with ice.

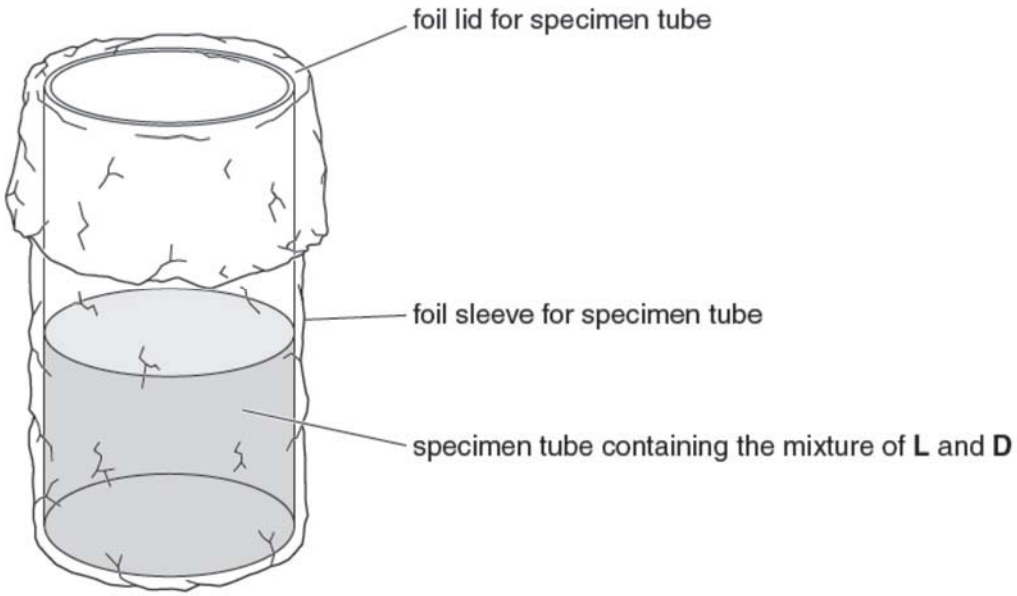


Fig. 1.1

7. Place the bench lamp 10 cm from the syringe on the white tile. Do **not** switch the lamp on.

   The next steps have to be carried out very quickly one after another, so read steps 8–15 and refer to Fig. 1.2 before proceeding.

8. Remove the foil lid and use a clean syringe to draw up 0.5 cm^3 of the mixture of **L** and **D** in the specimen tube. Replace the foil lid immediately.

9. Wipe the outside of the syringe and place it next to the colour standard on the white tile. This is the test syringe.

10. Immediately cover both syringes with the purple filter, **P**, as shown in Fig. 1.2 on page 4.

11. Switch on the bench lamp and immediately start a stopwatch or stop clock.

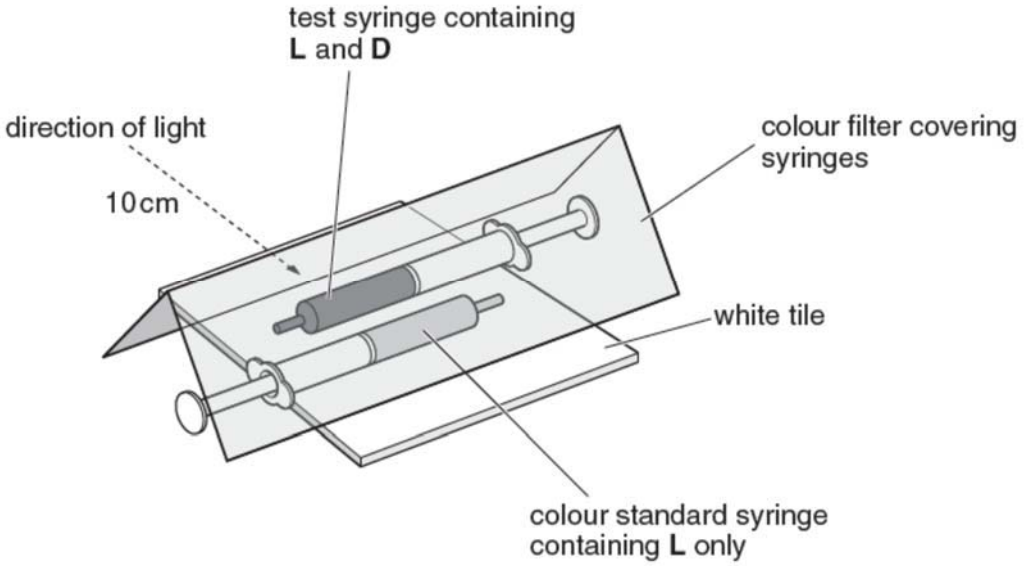


Fig. 1.2

12. In the space provided in **(a)**, record the time taken for the colour in the test syringe to match that of the colour standard. If the colour does not match after 300 seconds then record 'more than 300'.

13. Switch off the bench lamp.

14. Expel the contents of the test syringe into the beaker labelled **waste**. Rinse the syringe.

15. Repeat steps **8–14** using each of the four remaining coloured filters in turn (blue, green, orange and red).

(a) Record these results in a suitable table in the space provided to show the effect of wavelength on the time to decolourise DCPIP. [3]

(b) (i) Give one reason to explain why the leaf extract was kept on ice. [1]

(ii) State why the leaf extract containing DCPIP was kept covered by foil. [1]

(iii) Describe a suitable control that could have been set up for this investigation. [1]

(c) Suggest why the rate of photosynthesis is different at different wavelengths. [2]

(d) Suggest two significant sources of error in this experiment and describe two corresponding improvements that could be made to reduce the effects of these errors. [4]

In another similar investigation, a student collected leaves from two varieties of the same species of a garden plant that has different coloured leaves.

Variety A dark red leaves
Variety B green and white striped leaves

The student made a chloroplast extract from the leaves of each variety and measured the rate of photosynthesis for each extract in different wavelengths of light.

Table 1.2 shows the rates of photosynthesis calculated by the student from her results.

Table 1.2

wavelength of light / nm	rate of photosynthesis / s^{-1}	
	source of chloroplasts	
	dark red leaf	green and white striped leaf
425	0.097	0.081
450	0.071	0.063
525	0.023	0.023
625	0.030	0.039
675	0.057	0.058

(e) Use the grid provided to plot line graphs showing the effect of wavelength on the rate of photosynthesis. [4]

(f) Use your graph to estimate the rate of photosynthesis at a wavelength of 430 nm in plants with dark red leaves. [1]

(g) Explain how light of wavelength 430 nm leads to the decolourisation of DCPIP. [1]

(h) The photosynthetic pigments of the leaves from the two varieties of plants were extracted and were separated by two-way chromatography. The pigments were first separated by one solvent and then separated again by a second solvent at right angles to the first solvent. Fig. 1.3 shows the results for the two different varieties.

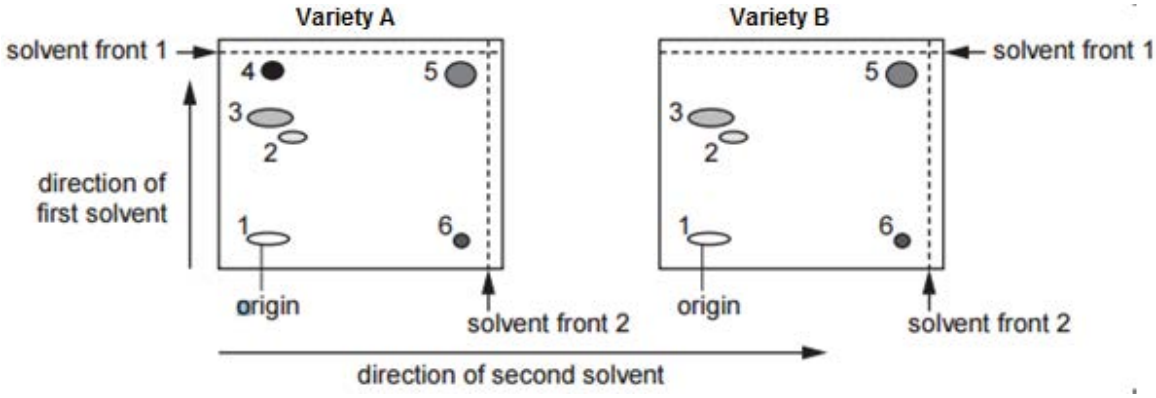


Fig. 1.3

Different photosynthetic pigments absorb different wavelengths of light. Table 1.3 shows some information about the pigments, P, Q, R, S and T, found in the 2 varieties, including the wavelength of light at which maximum light absorption occurs.

Table 1.3

pigment	wavelength of light / nm	Rf value	
		solvent 1	solvent 2
P	620	0.20	0.89
Q	545 and 547	0.60	0.29
R	420 and 660	0.65	0.11
S	490	0.91	0.19
T	430 and 645	0.82	0.92

$$Rf = \frac{\text{distance moved by pigment}}{\text{distance moved by solvent front}}$$

One of the varieties lacks one of the pigments.
Using the information in Table 1.3 and Fig. 1.3:

(i) identify the variety that lacks one of these pigments and state the letter of the missing pigment. [1]

(ii) state the evidence that supports your answer to (i). [2]

[Total: 21]

2 Methylene blue stains dead cells blue.
Living cells are not stained blue so they will appear white or clear.

You are provided with:
- methylene blue solution, M, (handle carefully as it will stain your skin)
- suspensions of yeast cells, labelled S1, S2 and S3.

Each suspension, S1, S2 and S3 has been heated for ten minutes at 45°C or 80°C or 100°C.

You are required to:
- use the microscope to observe the colour of the yeast cells from S1, S2 and S3, after M has been added
- record your observations by using annotated drawings of three yeast cells from each of S1, S2 and S3
- identify the temperature at which each of S1, S2 and S3 was heated.

1. Label three microscope slides S1, S2 and S3.

2. Place one drop of S1 onto slide S1 and add one drop of M. Mix carefully using a glass rod. (If M comes into contact with your skin rinse with cold water.)

3. Repeat step 2 with S2 and S3.

4. Leave for five minutes.

5. Add a coverslip to each slide.

6. Use the paper towel to dry off any excess liquid around the coverslip.

7. Use the microscope to observe the yeast cells on each slide, then select cells which you can draw and annotate to describe the effect of the methylene blue, M.

(a) (i) Prepare the space below and record your observations by:

- making drawings of three cells from each of the slides in the boxes provided
- annotating your drawings to describe the effect of methylene blue, M on the cells. [4]

S1

S2

S3

(ii) Use your observations to identify the temperature that was used to heat each of the suspensions S1, S2 and S3.
Complete the table. [1]

suspension	temperature / °C
S1	
S2	
S3	

(iii) Explain how you identified the yeast cells that had been heated at 100 °C. [1]

(b) Using the eyepiece graticule fitted in the eyepiece lens of your microscope, and the stage micrometer, find the actual length, in μm, of one of the yeast cells that you have drawn in S2.

Show the measurements that you made and your working. [3]

Actual length of a yeast cell = ……..... μm

(c) Draw a straight line on your drawing across the yeast cell to show where you took your measurement. [1]

Use your knowledge of the actual size of the yeast cell to calculate the magnification of your drawing. [1]

(d) The yeast *Rhodotorula glutinis* produces an enzyme, α-arabinofuranosidase, that could be used in the production of compounds to enhance the flavour and smell of fruit juices. The effect of the initial pH of the culture medium on the growth rate of this yeast was tested. Three continuous culture systems were set up, each with a different initial pH. The cultures were sampled at hourly intervals for 20 hours at each pH. The mean growth rate was then calculated.

The mean growth rates with their standard deviations are shown in Table 2.1.

Table 2.1

pH	mean growth rate / arbitrary units h^{-1}
4.0	0.156 $\pm$ 0.001
5.2	0.197 $\pm$ 0.013
7.0	0.037 $\pm$ 0.011

A *t*-test was carried out on the results for pH 4.0 and pH 5.2 and gave the value,

$$t = 2.4$$

The degree of freedom is 38.

Based on the findings of the *t*-test, a student concluded that pH 5.2 was optimum for the production of the enzyme α-arabinofuranosidase by *R. glutinis*. Suggest two reasons why this conclusion may not be valid. [2]

(e) A student carried out *t*-test on the results to compare the lengths of yeast cells when grown in different media.

A number of *t*-test was carried out to find out if, after 70 minutes, the difference in mean yeast cell length is significant:

1. between medium A and medium B $t = 2.50$
2. between medium A and medium C $t = 3.56$
3. between medium B and medium C $t = 1.94$

Table 2.2 shows the critical values for the *t*-test.

The number of degrees of freedom is 18.

Table 2.2

degrees of freedom	10	12	14	16	18	20	22	24	26	28	30	40	50	60
probability 0.05	2.23	2.18	2.14	2.12	2.10	2.09	2.07	2.06	2.06	2.05	2.04	2.02	2.01	2.00
probability 0.01	3.17	3.06	2.98	2.92	2.88	2.85	2.82	2.80	2.78	2.76	2.75	2.70	2.68	2.66

State what conclusions can be drawn about the significance of the differences in mean lengths from the three values of t given above. [3]

(f) Fig. 2.1 is a photomicrograph of a stained transverse section through part of a plant leaf. This plant species is native to part of Asia.

You are not expected to have studied this leaf.

Fig. 2.1

Draw a large plan diagram of the part of the leaf shown in Fig. 2.1. On your diagram, use a ruled label line and label to show the vascular bundle. [4]

[Total: 20]

3 A number of plant tissues are coloured because the cells contain chemicals called betacyanins.

You are provided with beetroot which contains betacyanins which colour the beetroot red.

In this experiment, you will test the effect of two different alcohols – methanol and ethanol on beetroot membranes. Ethanol is found in alcoholic beverages. Methanol, sometimes referred to as wood alcohol, can cause blindness and death.

If beet membranes are damaged, the red pigment will leak out into the surrounding environment. The intensity of color in the environment should be proportional to the amount of cellular damage sustained by the beet.

Plan an investigation to find out whether or not betacyanin leakage for beetroot occurs at the same intensity using ethanol and methanol.

You must use:
- beetroot
- 40% ethanol
- 40% methanol
- colourimeter and cuvette

You may select from the following apparatus and use appropriate additional apparatus:
- normal laboratory glassware e.g. test-tubes, boiling tubes, beakers, measuring cylinders, graduated pipettes, glass rods etc.
- stopwatch
- distilled water
- white tile
- scalpel
- forceps

Your plan should:
- have a clear and helpful structure such that the method you use is able to be repeated by anyone reading it
- be illustrated by relevant diagram(s), if necessary, to show, for example, the arrangement of the apparatus used
- identify the independent and dependent variables
- describe the method with the scientific reasoning used to decide the method so that the results are as accurate and repeatable as possible
- include layout of results tables and graphs with clear headings and labels
- use the correct technical and scientific terms
- include reference to safety measures to minimise any risks associated with the proposed experiment.

[Total: 14]

H

JURONG JUNIOR COLLEGE
JC 2 PRELIMINARY EXAMINATIONS
Higher 2

CANDIDATE NAME

CLASS

BIOLOGY

9744/01

Paper 1 Multiple Choice

15 September 2017

1 hour

Additional Materials: Multiple Choice Answer Sheet

READ THESE INSTRUCTIONS FIRST

Write in soft pencil.
Do not use staples, paper clips, glue or correction fluid.
Write your name and class on the Answer Sheet in the spaces provided unless this has been done for you.

There are **thirty** questions on this paper. Answer **all** questions. For each question there are four possible answers **A**, **B**, **C** and **D**.
Choose the **one** you consider correct and record your choice in **soft pencil** on the separate Answer Sheet.

Read the instructions on the Answer Sheet very carefully.

Each correct answer will score one mark. A mark will not be deducted for a wrong answer.
Any rough working should be done in this booklet.
The use of an approved scientific calculator is expected, where appropriate.

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

[Turn over

1 An actively growing cell is supplied with radioactive amino acids.

Which cell component would first show an increase in radioactivity?

A Golgi body

B mitochondrion

C nucleus

D rough endoplasmic reticulum

2 When mucus is secreted from a goblet cell in the trachea, these events take place.

1 addition of carbohydrate to protein

2 fusion of the vesicle with the plasma membrane

3 secretion of a glycoprotein

4 separation of a vesicle from the Golgi body

What is the sequence in which these events take place?

A 1 → 4 → 2 → 3

B 1 → 4 → 3 → 2

C 4 → 1 → 2 → 3

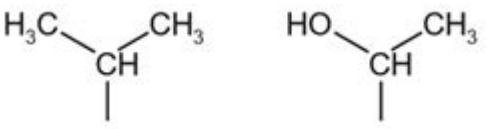
D 4 → 1 → 3 → 2

3 Which combination is found in a prokaryotic cell?

	endoplasmic reticulum	DNA	RNA	nucleus
A	X	✓	X	X
B	✓	X	X	✓
C	X	✓	✓	X
D	X	X	✓	✓

4 Threonylvaline is a dipeptide formed from the two amino acids, valine and threonine. A peptide bond forms between the amine group of valine and carboxyl group of threonine.

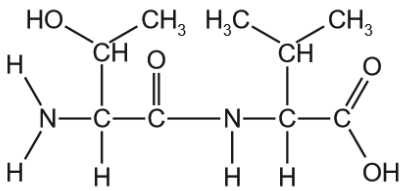
The side-chains (R groups) of the two amino acids are shown.



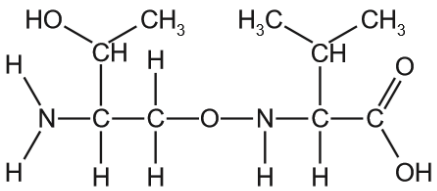
valine threonine

Which molecular structure is threonylvaline? **ANSWER: A**

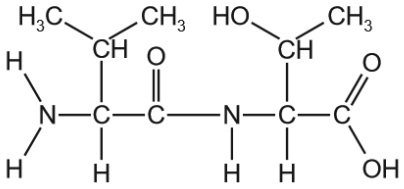
A



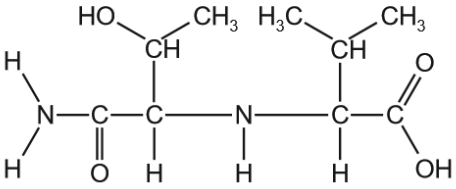
B



C



D



5 Which roles of the cell surface membrane are a result of the properties of the phospholipids?

1 to allow cytokinesis to occur in mitotic cell division

2 to allow entry and exit of oxygen and carbon dioxide

3 to allow the phagocytosis of a bacterium into a cell

A 1, 2 and 3

B 1 and 2 only

C 1 and 3 only

D 2 and 3 only

6 Which set of statements correctly describes haemoglobin?

	1	2	3	4
A	four polypeptide chains, each containing a haem group	iron ions can associate with oxygen forming oxyhaemoglobin	in each chain, hydrophobic R groups of amino acids point towards the centre of the molecule	at 50 % saturation, two oxygen molecules are transported by the molecule
B	polypeptide chains interact to produce a globular chain	each chain contains a haem group of amino acids surrounding an iron ion	consists of two identical alpha chains and two identical beta chains	each chain can transport an oxygen molecule
C	polypeptide chains interact to produce an almost spherical molecule	an iron ion is present within each haem group	quaternary structure has two alpha chains and two beta chains	each molecule can transport a total of four oxygen atoms
D	polypeptide chains produce a loose helical shape, which folds to form a spherical molecule	iron ions in the molecule can bind reversibly with oxygen	in each chain, hydrophobic R groups of amino acids surround the iron ion	each molecule can transport a total of eight oxygen atoms

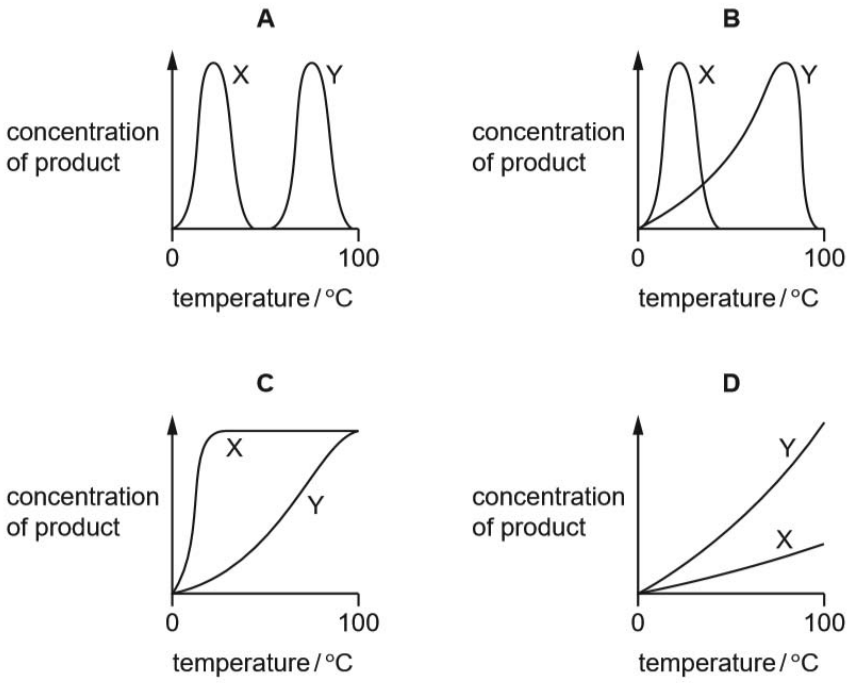
7 Two enzymes, X and Y, were used in an experiment.

Enzyme X was from bacteria that live in rivers and lakes at temperatures from 5°C to 20°C.

Enzyme Y was from bacteria that live in hot water springs at temperatures from 40°C to 85°C.

The experiment measured the concentration of product produced by each enzyme at temperatures between 0°C and 100°C after 5 minutes.

Which graph shows the results? **ANSWER: B**



8 Within its own environment a particular cell line cannot be induced to produce a cell from a different cell line.

Which statement explains this?

A Genes not required for a particular cell line are methylated.

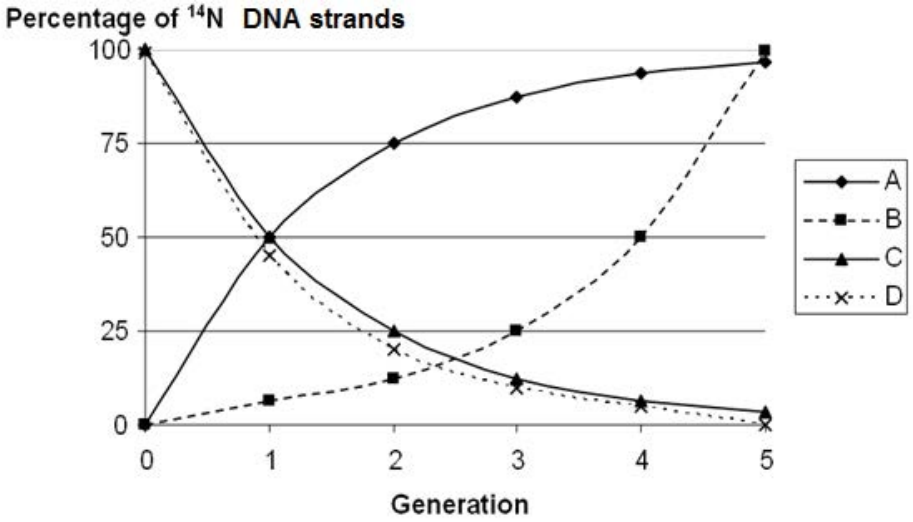
B Genes not required for a particular cell line are removed by enzymes.

C Only pre-mRNA that is required for a particular cell line is processed.

D Stem cells have only the genes required for their particular cell line.

9 Bacteria were cultured in a medium containing heavy nitrogen (^{15}N) until all DNA was labelled. These bacteria were then grown in a medium containing only normal nitrogen (^{14}N) for 5 generations. The percentage of ^{14}N DNA strands in each generation was estimated.

Which curve provides evidence that DNA replication is semi-conservative? **ANSWER: A**



10 An unidentified single-stranded molecule was described as having the following features.

- complementary base pairing along some of its length
- an area that can attach to a ribosome
- a site to which a specific amino acid attaches

What is the unidentified molecule?

A ribosomal RNA

B messenger RNA

C RNA polymerase

D transfer RNA

11 Some antibacterial drugs can affect the synthesis of proteins.

antimicrobial drug	rifampicin	streptomycin	tetracycline
mode of action	binds to RNA polymerase	genetic code misread during translation	prevents binding of tRNA to ribosome

Which is the correct set of immediate effects of these drugs?

antimicrobial drug	rifampicin	streptomycin	tetracycline
A	defective protein synthesised	mRNA does not bind to ribosome	amino acids not added to growing chain
B	mRNA not synthesised	defective protein synthesised	amino acids not added to growing chain
C	mRNA not synthesised	mRNA does not bind to ribosome	transcription prevented
D	transcription prevented	defective protein synthesised	mRNA does not bind to ribosome

12 Which statement about prokaryotes and chloroplasts is correct?

A Prokaryotes and chloroplasts have circular DNA where genes carrying the code for cell walls are located.

B Prokaryotes and chloroplasts have 70S ribosomes that are the sites for translation and polypeptide synthesis.

C Prokaryotes and chloroplast have an outer membrane and a separate inner, folded membrane where ATP synthesis occurs.

D Prokaryotes and chloroplast have double-stranded linear DNA where genes carrying coded information are located.

13 Human immunodeficiency virus (HIV) is a retrovirus. After infecting a host cell, viral DNA is produced which is incorporated into the DNA of the host cell. The modified host genome now codes for the production of new HIV particles.

Which could be used as a potential treatment to slow down the spread of HIV?

1 inhibitors of restriction endonucleases

2 inhibitors of reverse transcriptase

3 restriction endonucleases

4 reverse transcriptase

A 1 and 4 only

B 1 only

C 2 and 3 only

D 2 only

14 Which of the following does not occur during bacterial conjugation?

A direct contact between donor and recipient cells

B shortening of the pilus

C unidirectional transfer of both DNA strands

D enzymatic cleavage of one strand at the origin of transfer

15 Transcriptional control in eukaryotic cells can be accomplished at several levels.

What may be involved in such control?

1 The same combination of DNA binding proteins regulate the activity of all genes.

2 Enhancers may be involved in the promotion as well as regulation of gene transcription.

3 Phosphorylation of transcriptional factors by a kinase may occur.

4 Enhancers may be some distance from the promoter sites they control.

A 1, 2, 3 and 4

B 1, 2 and 3 only

C 1, 3 and 4 only

D 2, 3 and 4 only

16 Multiple copies of a wanted DNA fragment can be made by the polymerase chain reaction (PCR).

Which description of this procedure is not correct?

A After 'n' turns of the PCR cycle, up to 2^n copies of the wanted DNA are produced.

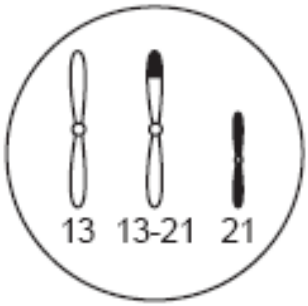
B Using a heat-stable enzyme, such as Taq polymerase, means that the enzyme does not lose activity over time.

C Using an enzyme with a high optimum temperature allows DNA polymerisation above the annealing temperature.

D Using specific primers means that only the wanted DNA is replicated.

17 Down's syndrome can be caused by a trisomy of chromosome 21, but can also result from the translocation of chromosome 21 into chromosome 13, forming a single chromosome 13-21.

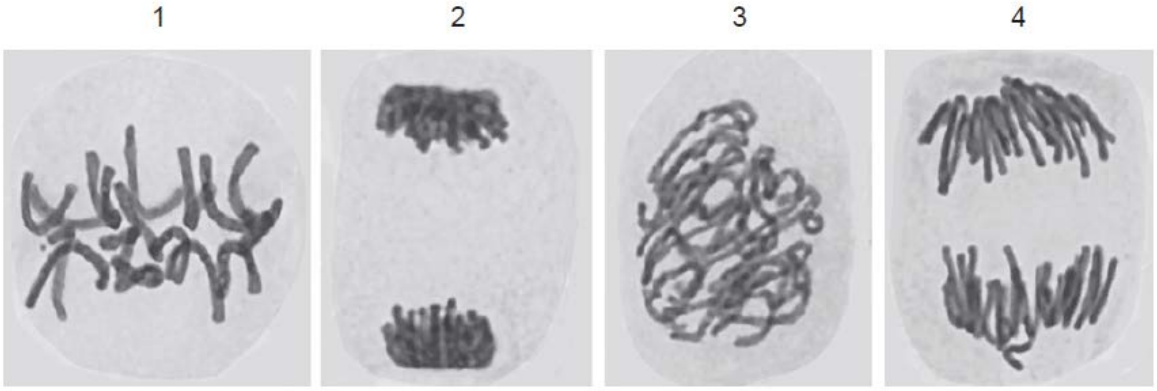
The diagram shows chromosomes 13 and 21 in the nucleus of a diploid (2n) testis cell from a phenotypically normal male carrier of a 13-21 translocation. This cell has a chromosome number of 45.



Which is **not** a likely outcome of fertilisation of normal oocytes by sperm from this male?

	chromosomes in sperm	embryo
A	13 and 21	2n =46 normal phenotype
B	13-21	2n =45 normal phenotype
C	13-21 and 21	2n =46 Down's syndrome
D	13-21 and 21	2n =47 Down's syndrome

18 The photomicrographs show cells in various stages of the cell cycle.



Which cells contain twice as many DNA molecules as a cell from the same organism after cytokinesis?

A 1, 2, 3 and 4

B 1, 2 and 4 only

C 1 and 3 only

D 2 and 4 only

19 Gene mutations in either the *BRCA1* or the *BRCA2* genes are responsible for the majority of hereditary breast cancer in humans.

The proteins produced by the two genes migrate to the nucleus where they interact with other proteins, such as those produced by the tumour suppressor gene, *p53*, and the DNA repair gene, *RAD51*.

Which combination of gene activity is most likely to result in breast cancer? **ANSWER: D**

	gene		
	BRCA1 or *BRCA2*	*p53*	*RAD51*
A	✓	✓	✓
B	✓	✓	✗
C	✓	✗	✓
D	✗	✗	✗

key

✓ = gene produces normal protein

✗ = gene produces abnormal protein or no protein

20 What maximum number of different genotypes and phenotypes are possible among the children of a mother with blood group A and a father with blood group B?

	genotype	phenotype
A	2	2
B	2	4
C	4	4
D	4	2

21 A test cross resulted in these recombinants:

$$\frac{tB}{tb} \qquad \frac{Tb}{tb}$$

Which was the parental test cross?

A $\frac{TB}{tb} \times \frac{tb}{tb}$

B $\frac{TB}{tB} \times \frac{tb}{Tb}$

C $\frac{Tb}{tB} \times \frac{tb}{tb}$

D $\frac{TB}{tb} \times \frac{TB}{tb}$

22 Isolated chloroplasts, suspended in buffer solution, are often used to study the light dependent stage of photosynthesis.

During this stage, electrons (e^-) are transferred by carriers and provide energy so that a proton (H^+) gradient can be formed. Protons diffuse through membrane proteins that are linked to synthase enzymes.

Three compounds that can be added to isolated chloroplasts are:

1 DCMU, which inactivates a carrier that accepts electrons from photosystem II

2 DCPIP, which can act as a final electron acceptor

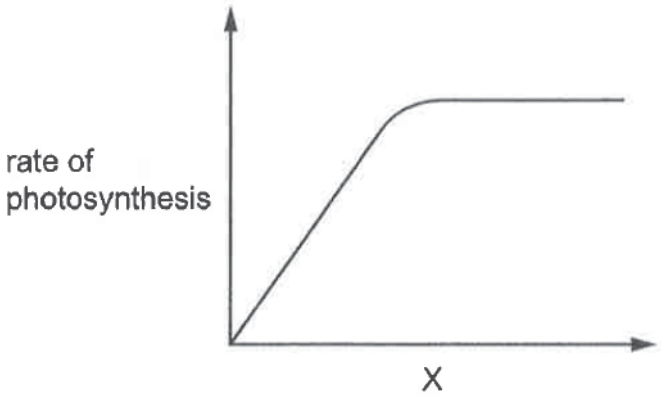
3 ammonium hydroxide solution, which absorbs protons

Which compounds, when added separately to isolated chloroplasts, would allow the light dependent stage of photosynthesis to occur and which would inhibit it?

	allow	inhibit
A	1	2 and 3
B	1 and 3	2
C	2	1 and 3
D	2 and 3	1

23 The rate of photosynthesis in pondweed was measured when one variable was changed and all others were standardised.

The graph shows the rate of photosynthesis at different values of a variable, X.



Which variables could be represented by X?

1 carbon dioxide availability

2 light intensity

3 oxygen availability

4 temperature

5 leaf area exposed to direct light

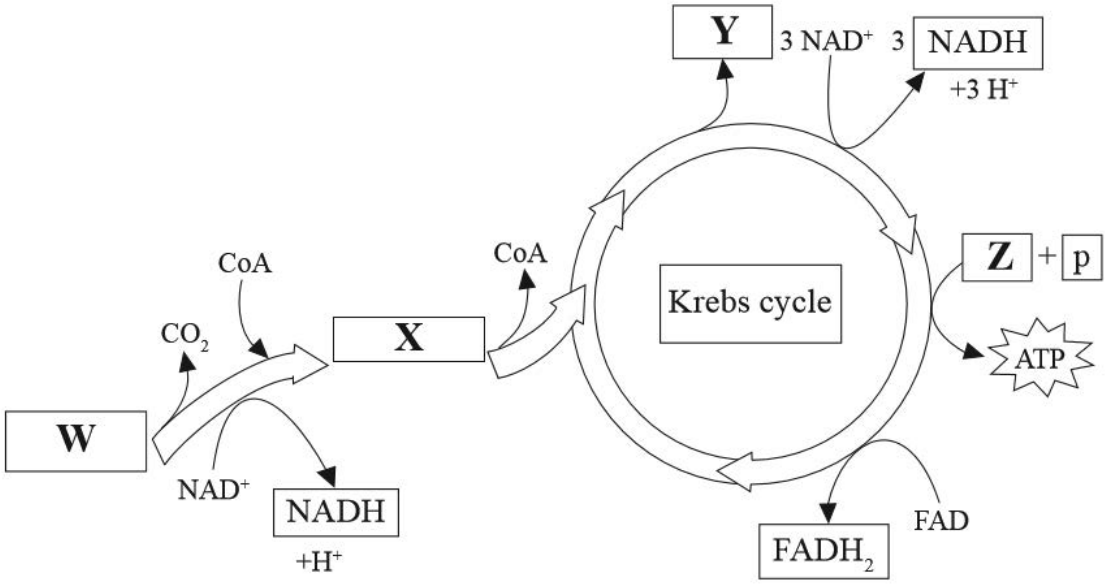
A 1, 2 and 5

B 1 and 2 only

C 2, 4 and 5

D 3 and 4

24 The diagram below shows the link reaction and stages of the Krebs cycle. Which molecules are represented by the letters W, X, Y and Z?



	W	X	Y	Z
A	acetyl CoA	carbon dioxide	ADP	pyruvate
B	pyruvate	acetyl CoA	carbon dioxide	ADP
C	ADP	carbon dioxide	acetyl CoA	pyruvate
D	acetyl CoA	pyruvate	carbon dioxide	ADP

25 The function of phosphatases in signal transduction is to

A move the phosphate group of the transduction pathway to the next molecule of a series.

B prevent a protein kinase from being reused when there is another extracellular signal.

C amplify the second messengers such as cAMP. / amplify the transduction signal so it affects multiple transducers.

D inactivate protein kinases and turn off the signal transduction.

26 Which statements are acceptable parts of Darwinian evolutionary theory?

1 Advantageous behaviour acquired during the lifetime of an individual is likely to be inherited.

2 In competition for survival, the more aggressive animals are more likely to survive.

3 Species perfectly adapted to a stable environment will continue to evolve.

4 Variation between individuals of a species is essential for evolutionary change.

A 1, 2 and 4 only

B 2 and 3 only

C 3 and 4 only

D 4 only

27 Before the settlement of California in the 1800s, the elk population was very large. By about 1900 there were only a few dozen elk left.

Owing to protection, there are now about 3000 elk living in a small number of isolated herds.

Unfortunately, some of the elk in all the herds have difficulty grazing due to a shortened lower jaw.

Which statements best explain this?

1 The early settlers only hunted elk that could graze.

2 There was a mutation affecting jaw size in one of the herds.

3 There is random mating within each herd.

4 The current elk population demonstrates a founder effect.

5 There was directional selection favouring short jaws.

A 1, 2 and 4 only

B 2, 3 and 5 only

C 2 and 5 only

D 3 and 4 only

28 Darwin's view of the process of evolution to form new species (speciation) has been reinforced by more recent discoveries in genetics and cell biology.

In this view, which sequence of events is considered most likely to lead to speciation?

A	adaptation to population	→	competition and predation leading to natural selection	→	behavioural isolation	→	sympatric speciation
B	adaptation to population	→	competition and predation leading to natural selection	→	behavioural isolation	→	allopatric speciation
C	competition and predation leading to natural selection	→	geographical isolation	→	adaptation of isolated populations	→	sympatric speciation
D	competition and predation leading to natural selection	→	geographical isolation	→	adaptation of isolated populations	→	allopatric speciation

29 Apart from the ABO blood groups, humans can also be Rhesus positive or Rhesus negative.

People with the Rhesus antigen are Rhesus positive. When a Rhesus negative person is given Rhesus positive blood in a transfusion there is no problem. However, a second transfusion of Rhesus positive blood to this Rhesus negative person will result in a reaction between the two types of blood.

Which statements explain this?

1 A Rhesus negative person naturally has anti-Rhesus antibodies.

2 Exposure to Rhesus antigen causes anti-Rhesus antibody production.

3 B-cells make anti-Rhesus antibodies.

4 Anti-Rhesus antibody production begins after the second exposure.

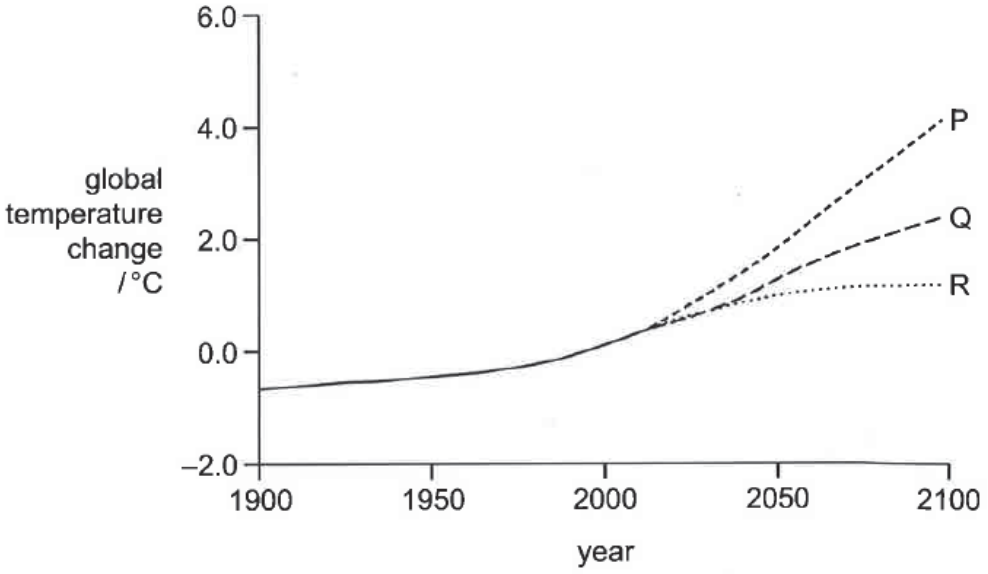
A 1, 2, 3 and 4

B 1 and 2 only

C 2 and 3 only

D 3 and 4 only

30 The graph shows the predicted change in global temperatures using three different models, P, Q and R. Model Q assumes that no new factors act to influence the rate of climate change.



The predictions based on models P and R can be explained using some of the following statements.

1 An increased global temperature and reduced rainfall will lead to an increase in forest fires.

2 Permanently frozen soil and sediment in the Arctic will begin to thaw as global temperatures rise.

3 Rising sea temperatures will cause increased growth of photosynthetic algae.

4 Rising sea temperatures will reduce the solubility of greenhouse gases in the oceans.

Which of these statements support predictions P and R?

	statements that support prediction P	statements that support prediction R
A	1, 2 and 4	3
B	1 and 3	2 and 4
C	2	1, 3 and 4
D	3 and 4	1 and 2

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JURONG JUNIOR COLLEGE
JC 2 PRELIMINARY EXAMINATIONS
Higher 2

CANDIDATE NAME

CLASS

BIOLOGY 9744/02

Paper 2 Structured Questions **25 August 2017**

Candidates answer on the Question Paper. **2 hours**

No Additional Materials are required.

READ THESE INSTRUCTIONS FIRST

Write your name and class in the spaces at the top of this page.
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.

Answer **all** questions in the spaces provided on the Question Paper.

The use of an approved scientific calculator is expected, where appropriate.
You may lose marks if you do not show your working or if you do not use appropriate units.

The number of marks is given in brackets [ ] at the end of each question or part question.

For Examiner's Use	
1	
2	
3	
4	
5	
6	
7	
8	
9	
Total	

Answer **all** questions.

1 The plasma membrane Ca^{2+} ATPase (PMCA) is a vital transport protein that regulates the amount of Ca^{2+} within eukaryotic cells. In humans, there is a very large transmembrane electrochemical gradient of Ca^{2+} driving the entry of Ca^{2+} into cells, yet low intracellular concentrations of Ca^{2+} are maintained by PMCA.

Fig. 1.1 shows two conformations that PMCA interconverts between, depending on whether Ca^{2+} is bound.

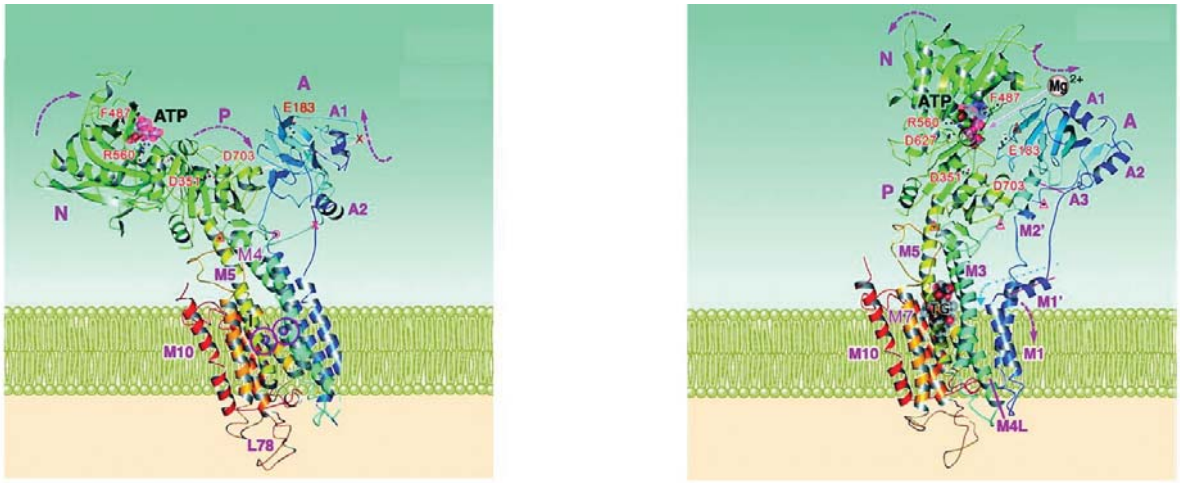

Fig. 1.1

(a) Explain why Ca^{2+} cannot freely cross the plasma membrane. [2]

1. **(Hydrophobic) non-polar fatty acids make up the hydrophobic core of the plasma membrane, therefore the membrane is impermeable to Ca^{2+};;**
2. **Ca^{2+} are <u>charged</u> and are <u>repelled by the hydrophobic core</u> of the plasma membrane, thus Ca^{2+} cannot cross the plasma membrane freely;;**

(b) Describe how low intracellular concentrations of Ca^{2+} are maintained by PMCA. [3]

1. **Ca^{2+} <u>recognises and binds to the specific binding site</u> of PMCA/the carrier protein;; (extra pt)**
2. **PMCA <u>undergoes a conformational change</u> and releases Ca^{2+} to the extracellular/other side of the membrane;;**
3. **Ca^{2+} is transported across the plasma membrane <u>via active transport</u>;;**
4. **<u>against a concentration gradient</u>;;**
5. **ATP is hydrolysed/required;;**

Various forms of PMCA are expressed in different cell types, including erythrocytes (red blood cells). Erythrocytes rely on Ca^{2+} dependent signalling during their differentiation from hematopoietic stem cells in the bone marrow. The process of erythropoiesis (production of mature red blood cells) is illustrated in Fig. 1.2.

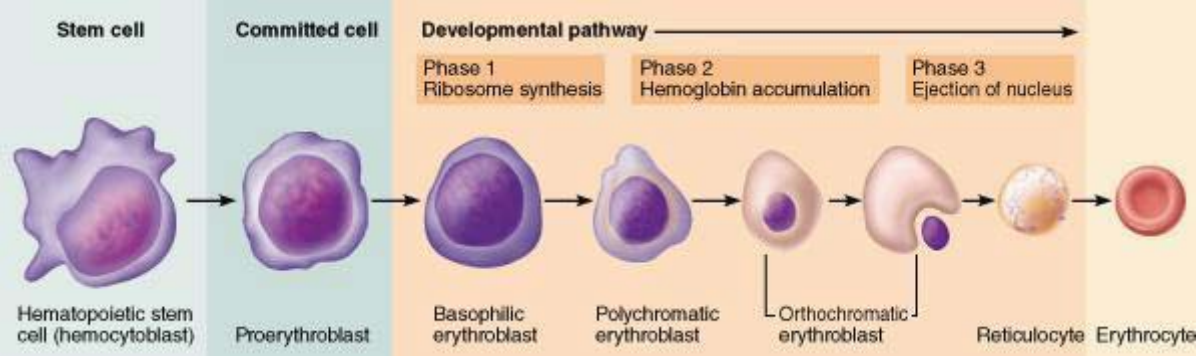

Fig. 1.2

(c) Describe the potency of these hematopoietic stem cells and explain their normal functions. [4]

1. **These hematopoietic/blood stem cells are multipotent;;**
2. **They have the ability to differentiate into a limited range of cell types and so are not pluripotent or totipotent;;**

3. **To maintain and repair the specific tissue (blood) where they are found/where they reside (by replacing worn-out or damaged cells);;**
4. **Can undergo differentiation to form the different types of blood cells – red blood cells and white blood cells (e.g. B lymphocytes, T lymphocytes, natural killer cells, macrophages, platelets etc.);;**
5. **To replace worn-out or damaged red blood cells and white blood cells;;**
6. **Will constantly divide to replace cells such as the red blood cells that are worn out in three to four months;;**

In addition to the loss of the nucleus, cellular organelles such as the endoplasmic reticulum, Golgi apparatus and mitochondria are also lost from the reticulocytes shown in Fig. 1.2.

(d) Outline the structure of the endoplasmic reticulum and Golgi apparatus in typical eukaryotic cells. [2]

1. **The (rough) endoplasmic reticulum consists of a network of sheets called cisternae;;**
   OR
2. **The (smooth) endoplasmic reticulum consists of a network of tubules/tubes called cisternae**
3. **The Golgi apparatus consists of a stack of flattened membrane-bound sacs called cisternae (together with a system of associated vesicles called Golgi vesicles);;**

(e) Since mature mammalian erythrocytes lack mitochondria, suggest how these cells derive energy from glucose. [1]

1. **Mature erythrocytes can still obtain <u>ATP</u> (via substrate level phosphorylation) <u>from glycolysis</u>;;**

[Total: 12]

2 Fig. 2.1 shows two animal cells in different stages of the mitotic cell cycle.

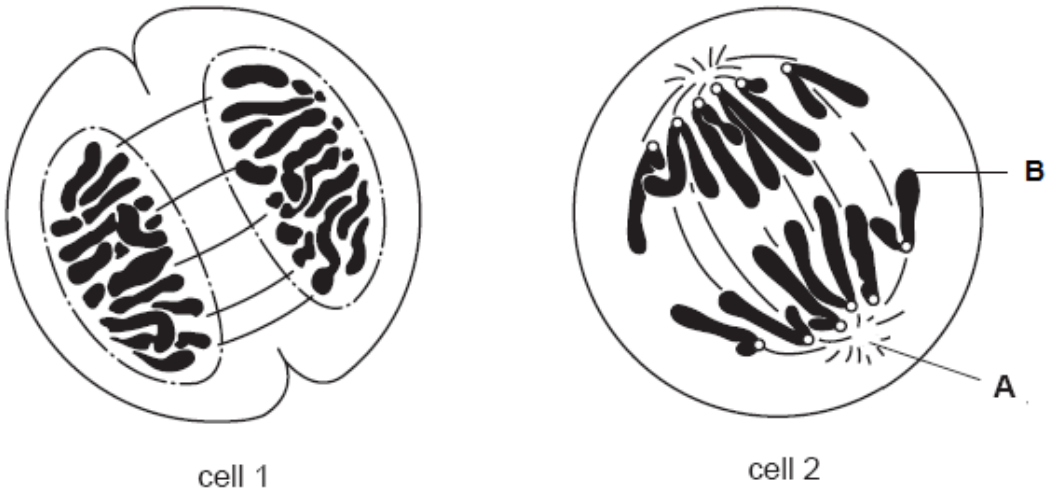

Fig. 2.1

(a) With reference to cell 1,

(i) identify the stage of nuclear division taking place. [1]

1. **Telophase;;**

(ii) describe the events occurring at this stage. [2]

1. **Daughter chromosomes pulled by spindle fibres attached to centromeres reach opposite poles of the cell;;**
2. **Spindle fibres disintegrate;;**
3. **Nuclear envelope reforms around the chromatin (accept: chromosomes) in each daughter cell;;**
   (any 2)

(b) A pair of rod-like structures can be found in region A. Outline the roles of these structures during mitosis in animal cells. [2]

1. **(The two pairs of) centrioles move to the opposite poles of the cell (during prophase) and determine the polarity of the cell;;**
2. **Centrioles act as the microtubule-organising centre (MTOC) – the centrioles produce spindle fibres at the poles towards the equator of the cell;;**
3. **Centrioles organise the synthesis of spindle fibres which lead to the separation of chromatids during cell division;;**

(c) Region B of the chromatid contains non-coding repetitive nucleotide sequences.

(i) Account for the progressive shortening of these sequences after repeated rounds of DNA replication. [2]

1. **DNA polymerase can only add DNA nucleotides to the free 3' (-OH) end of an existing strand;;**
2. **With the removal of the RNA primer from the 5' end of the newly synthesised DNA strand, there is no free 3' (-OH) end for DNA polymerase to add (free) nucleotides to,;;**
   OR
3. **With the removal of the RNA primer from the 5' end of the newly synthesised DNA strand, the RNA primer at the 5' end of the (newly synthesised) DNA strand is removed but not replaced;;**
4. **This results in a daughter strand that is shorter than the parental/template DNA strand.**

(ii) Describe two functions of these non-coding DNA in eukaryotes. [2]

1. **Telomeres ensure genes are not lost or eroded due to the end replication problem with each round of DNA replication, preventing loss of important genetic information;;**
2. **Telomeres protect and stabilise the terminal ends of chromosomes by forming a loop which confers stability to linear chromosomes by preventing accidental fusion of the single-stranded end of one chromosome to the single-stranded end of another chromosome via complementary base pairing;;**
   OR
3. **Telomeres protect and stabilise the terminal ends of chromosomes. Telomeric DNA is bound by specific telomere-specific binding proteins which protect the chromosomal ends from degradation by exonucleases;;**
4. **Telomeres that are critically short trigger apoptosis (programmed cell death);;**
5. **Telomeres allow their own extension, by providing an attachment point for the correct positioning of telomerase;;**
   (any 2)

(d) In 2006, Yamanaka and his colleagues demonstrated in an experiment with mice that induced pluripotent stem (iPS) cells could be produced by genetically reprogramming fully differentiated adult cells. There has been evidence to suggest that these iPS cells exhibit high telomerase reverse transcriptase (TERT) activity and are capable of dividing indefinitely.

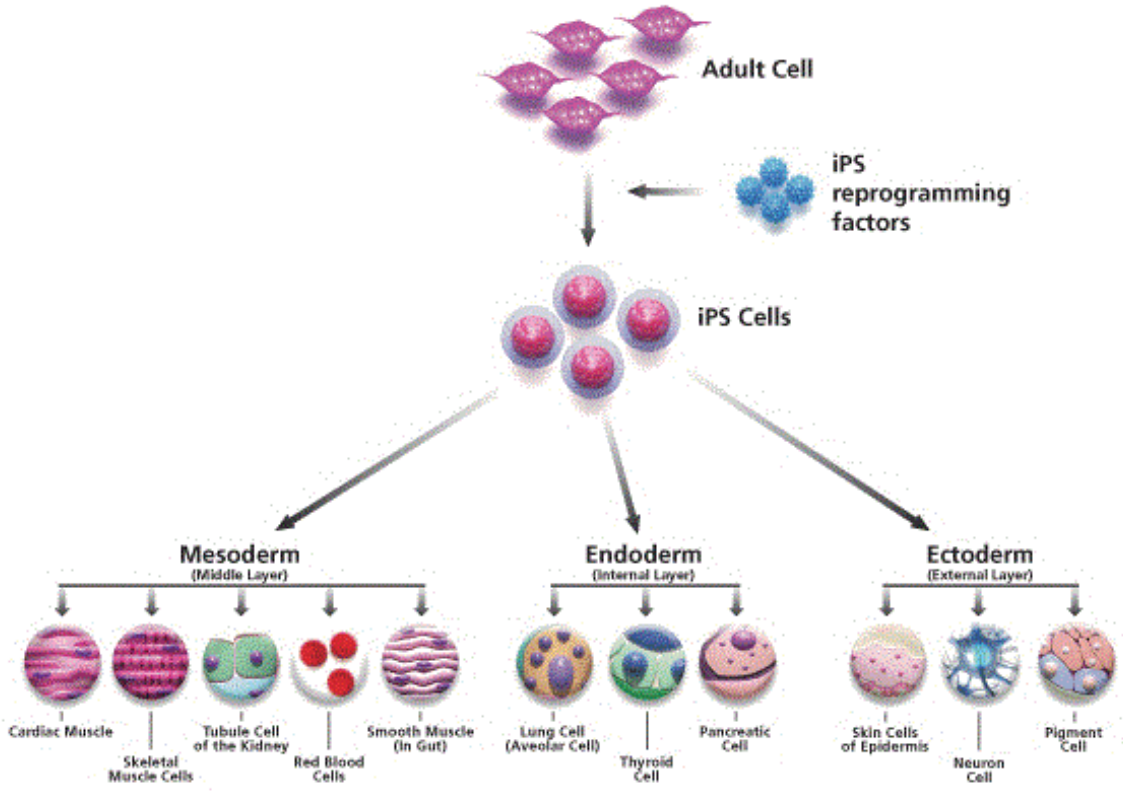

Fig. 2.2

(i) Discuss the role of TERT in enabling the iPS cells to divide indefinitely. [2]

1. **TERT catalyses the <u>elongation of telomeres / maintain length of telomeres / maintain number of telomere repeat sequences</u> by adding telomere repeat sequences to the 3' end of the DNA strand/telomeres;;**
2. **Therefore, the cells do not enter replicative senescence / apoptosis is not triggered;;**

(ii) Suggest an advantage of using iPS cells in research and medical applications. [1]

1. **Since iPS cells are derived from adult cells, the use of iPS cells overcomes ethical issues pertaining to the destruction of human embryos as a source of (pluripotent) stem cells;;**
   OR
2. **Since iPS cells can be derived from adult cells of the patient, the use of iPS cells presents no/low risk of immune rejection;;**

[Total: 12]

3 Antibiotic resistance is rising to dangerously high levels in all parts of the world. A growing list of infections – such as pneumonia, tuberculosis, blood poisoning and gonorrhoea – are becoming harder, and sometimes impossible, to treat as antibiotics become less effective.

Some bacteria are naturally resistant to certain types of antibiotics. However, bacteria may also become resistant either by a genetic mutation or by acquiring resistance from another bacterium.

(a) Outline the process of how a bacterium is able to acquire resistance from another bacterium. [3]

1. **bacterial conjugation;;**
2. **F^+ cell/donor bacterial cell with F factor produces sex pilus to attach itself to F^- cell/recipient cell;;**
3. **A temporary cytoplasmic mating bridge is formed between the two bacterial cells which allows F^+ cell to transfer its F plasmid containing the antibiotic resistance gene to the F^- cell (by rolling circle mechanism);;**

Or

4. **bacterial transformation;;**
5. **A bacterium takes up foreign DNA containing antibiotic resistance gene;;**
6. **The foreign DNA is incorporated into bacterium's own DNA via homologous recombination/through crossing over with a homologous region found on the bacterial chromosome;;**

More than 2 million Americans each year are infected by antibiotic-resistant bacteria, and at least 23,000 die annually from those infections. Antibiotic-resistant bacteria have become a global health crisis and alternative treatments such as Phage Therapy are being considered for combating bacterial infections.

Phage Therapy involves the targeted application of bacteriophages that, upon encounter with specific pathogenic bacteria, can infect and kill them. Phages are currently being used therapeutically to treat bacterial infections that do not respond to conventional antibiotics.

Fig. 3.1 is an electron micrograph showing a phage infecting a bacterium during Phage Therapy.

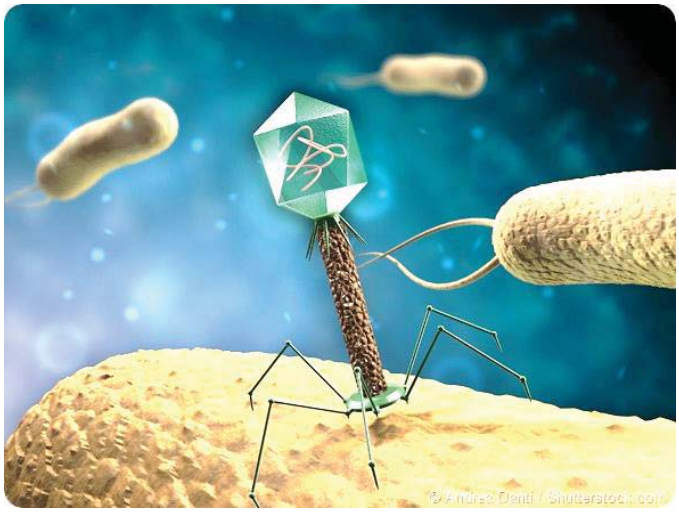

Fig. 3.1

(b) Suggest the reproductive cycle of a phage used for Phage Therapy. [1]

- **lytic cycle**

(c) Describe how a structural feature of the phage allows for targeted application to specific pathogenic bacteria. [2]

1. **attachment sites on its tail fibres;;**
2. **<u>complementary in shape to specific receptor sites</u> on the specific host bacterial cell wall, <u>recognise and adsorb</u> to specific receptor sites on the specific host bacterial cell wall;; *(mark for 'specific receptor sites on the specific host bacterial cell wall' once)***

(d) Explain how the use of phages can prevent the spread of bacterial infection. [2]

1. **enzymes coded by the genome of the phage shuts down the bacterium's macromolecular (i.e. DNA, RNA and protein) synthesis;;**
   OR
2. **phage nucleases hydrolyse the bacterial chromosome;;**
   OR
3. **lysozyme breaks down peptidoglycan cell wall;;**
4. **new bacteria cells cannot be synthesized;;**
5. **lysis (death) of host cell occurs upon the release of new phage particles;;**

(e) Suggest characteristics of phages that make them attractive therapeutic agents. [2]

1. **highly specific / more specific than antibiotic;;**
2. **very effective in lysing targeted pathogenic bacteria;;**
3. **typically harmless;;**
4. **will not develop resistance;;**
5. **rapidly modifiable to combat the emergence of newly arising bacterial threats;;**

[Total: 10]

4 Sickle cell anaemia is a recessive genetic disease caused by a mutation that commonly occurs in the DNA, resulting in hydrophobic valine replacing hydrophilic glutamic acid at the 6th amino acid position of the β chain.

(a) State the type of mutation that commonly occurs to result in sickle cell anaemia. [1]

1. **(single) base-pair substitution;;**

(b) Describe the effects of this change in amino acids on the red blood cells of an individual with the disease. [4]

1. **This results in a change in the tertiary structure/3D conformation of haemoglobin to produce haemoglobin S (HbS) instead of HbA;;**
2. **This decreases the solubility of deoxygenated HbS and at low oxygen concentration, hydrophobic areas of different HbS would stick together;;**
3. **HbS molecules will polymerise and precipitate out of solution to form rigid fibres;;**
4. **the change from HbA to HbS would result in changes to the shape of the red blood cells from circular biconcave shape to become sickle shape;;**

To detect if individuals are afflicted with sickle cell anaemia, restriction fragment length polymorphism (RFLP) analysis can be carried out using gel electrophoresis and Southern Blotting. Restriction enzymes are used to digest the DNA before RFLP analysis and the mutation removes a recognition site of the restriction enzyme *MstII,* as shown in Fig. 4.1. The enzyme's recognition sites on the normal allele and the mutant allele are shown by arrows.

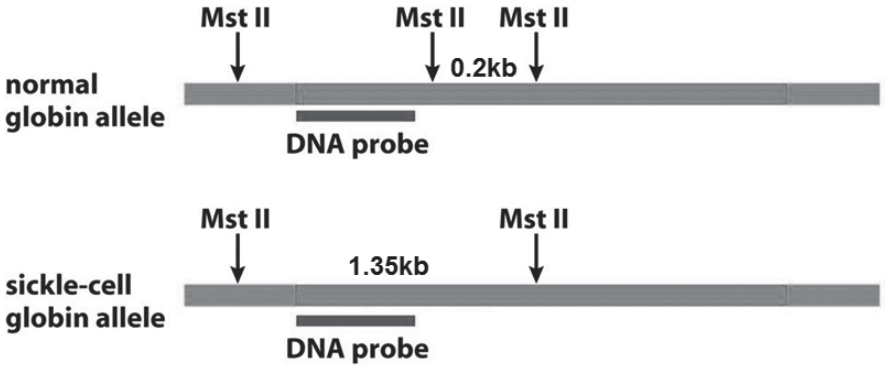

Fig. 4.1

(c) Draw, on Fig. 4.2, the expected band patterns produced by DNA from individuals with sickle cell anaemia. [1]

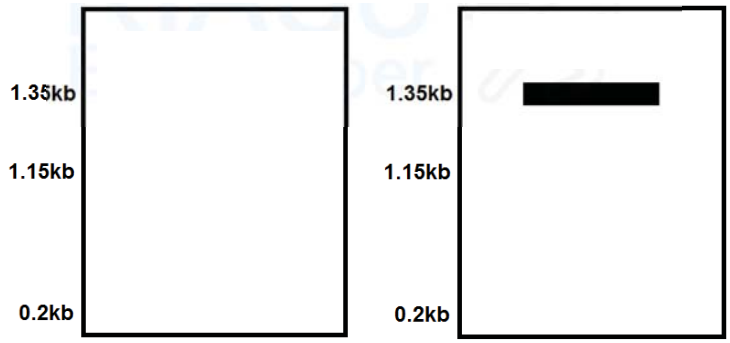

Fig. 4.2

(d) Suggest why it is necessary to carry out Southern Blotting after gel electrophoresis. [2]

1. **After gel electrophoresis is carried out to separate DNA fragments according to size, there might be <u>too many bands to be distinguished individually</u>;;**
2. **Hence, Southern blot is usually carried out to <u>identify the DNA fragment of interest</u>;;**

(e) Outline the role of the DNA probe in Southern Blotting. [1]

1. **The probe is single-stranded to hybridise/complementary base pair with DNA fragments/specific nucleotide sequences on the nitrocellulose paper;;**
2. **The probe is radioactively-labelled so that the DNA fragments will show up as bands after autoradiography/on the photographic X-ray film;;**

[Total: 9]

5 Fig. 5.1 shows awned and awnless rice strains. A long awn is one of the distinct morphological features of wild rice species. It is a long needle-like appendage that is thought to aid in seed dispersal and prevent predation by animals.

The genes *DROOPING LEAF (DL)* and *OsETTIN2 (OsETT2)* are involved in awn formation. Genetic analysis experiments indicate that *DL* and *OsETT2* act independently in awn formation.

Fig. 5.1

A cross between pure-breeding awned and awnless strains produced awned plants in F1.

The F1 plants were then self-pollinated.

In the F2 generation, 658 awned plants and 48 awnless plants were produced.

This control of awn development is an example of epistasis resulting in a ratio that is close to 15:1.

(a) Define the term *locus*. [2]

1. **the position of a gene;;**
2. **on a chromosome or within a DNA molecule;;**

(b) Explain the term epistasis in this context. [3]

1. **The awn formation is controlled by two genes occupying different loci;;**
2. **when either gene has a dominant allele at the gene loci;; (it hides the effect of the other gene)**
   OR
3. **a copy of dominant allele at either gene results in awn development;;**
4. **homozygous for the recessive allele at both genes results in awnless plants/character;;**
   (Such gene interaction is also known as duplicate dominant epistasis)

(c) Use the symbols A, a and B, b to draw a genetic diagram to explain the results shown in the F2 generation. [4]

F1 phenotypes: **awned plants** **X** **awned plants**

F1 genotypes: **AaBb** **X** **AaBb** ;;

Gametes **AB** **Ab** **AB** **Ab** ;;

aB **ab** **aB** **ab**

Fertilization

	AB	**Ab**	**aB**	**ab**
AB	**AABB**	**AABb**	**AaBB**	**AaBb**
Ab	**AABb**	**AAbb**	**AaBb**	**Aabb**
aB	**AaBB**	**AaBb**	**aaBB**	**aaBb**
ab	**AaBb**	**Aabb**	**aaBb**	**aabb**

F2 genotype : **AABB** **AABb** **AaBB** **AaBb** **Aabb** **AAbb** **aaBB** **aaBb** : **aabb** ;;

F2 phenotype : **awned plants** : **awnless plants** ;;

F2 phenotypic ratio : **15** : **1**

A chi-squared test was carried out on the results of the second cross.

Table 5.1

phenotype	observed number (O)	expected number (E)	$\frac{(O-E)^2}{E}$	
awn	658	**661.9**	**0.0229793**	;;
awnless	48	**44.1**	**0.339381**	;;
total number	706	706	x^2 = **0.3623603**	;;

(d) Complete the five missing values in Table 5.1. [3]

(e) Table 5.2 shows part of the table of probabilities for the chi-squared test.

Table 5.2

degrees of freedom	probability								
	0.995	0.975	0.9	0.5	0.1	0.05	0.025	0.01	0.005
1	.000	.000	0.016	0.455	2.706	3.841	5.024	6.635	7.879

Use Table 5.2 and your calculated value for the chi-squared test to find the probability that the observed ratio of phenotype does not deviate significantly from the expected ratio. [1]

- **Value of p is between 0.5 - 0.9**

(f) State what conclusions may be drawn from the probability found in (e). [2]

(Value of p is between 0.5 - 0.9, more than p = 0.05)

1. **Do not reject the null hypothesis, there is no significant difference between the observed and the expected ratio;;**
2. **The observed ratio of phenotype does not deviate significantly from the expected ratio, any deviation from the expected is due to chance;;**

[Total: 15]

6 Fig. 6.1 shows some stages in mammalian respiration.

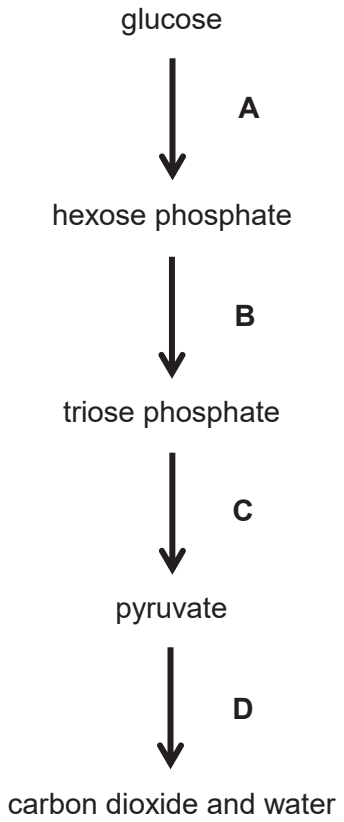

Fig. 6.1

(a) Name the processes taking place during Stage D and state precisely where they occur. [3]

1. **Link reaction – mitochondrial matrix;;**
2. **Krebs cycle – mitochondrial matrix;;**
3. **Oxidative phosphorylation – inner mitochondrial membrane;;**

(b) Intermediates produced at the end of Stages B and C are important in the conversion of carbohydrates to lipids such as triglycerides. Some of the triose phosphate can be converted into glycerol-3-phosphate, while pyruvate can undergo further reactions to form intermediates required for the synthesis of fatty acids.

(i) Describe the formation of triglycerides. [3]

1. **A triglyceride is formed by condensation reactions between 1 glycerol and 3 fatty acids;;**
2. **Each of glycerol's hydroxyl/–OH groups condenses with the carboxyl/ –COOH group of a fatty acid;;**
3. **In each condensation reaction, one water molecule is removed, resulting in the formation of an ester bond/linkage;;**

(ii) State two roles of triglycerides in living organisms. [2]

1. **Triglycerides serve as a good energy source;;**
2. **Triglycerides are a weight efficient means for organisms to store energy / serve as good energy storage molecules;;**
3. **Triglycerides serve as a good source of metabolic water;;**
4. **Triglycerides are good thermal insulators that reduce / prevent excessive heat loss from the body;;**
5. **Provide buoyancy to aquatic animals;;**
   (any 2)

(c) The first reaction in Stage A is catalysed by the enzyme hexokinase. It has been observed that hexokinase is bound to the outer mitochondrial membrane in muscle cells which undergo high rates of glycolysis.

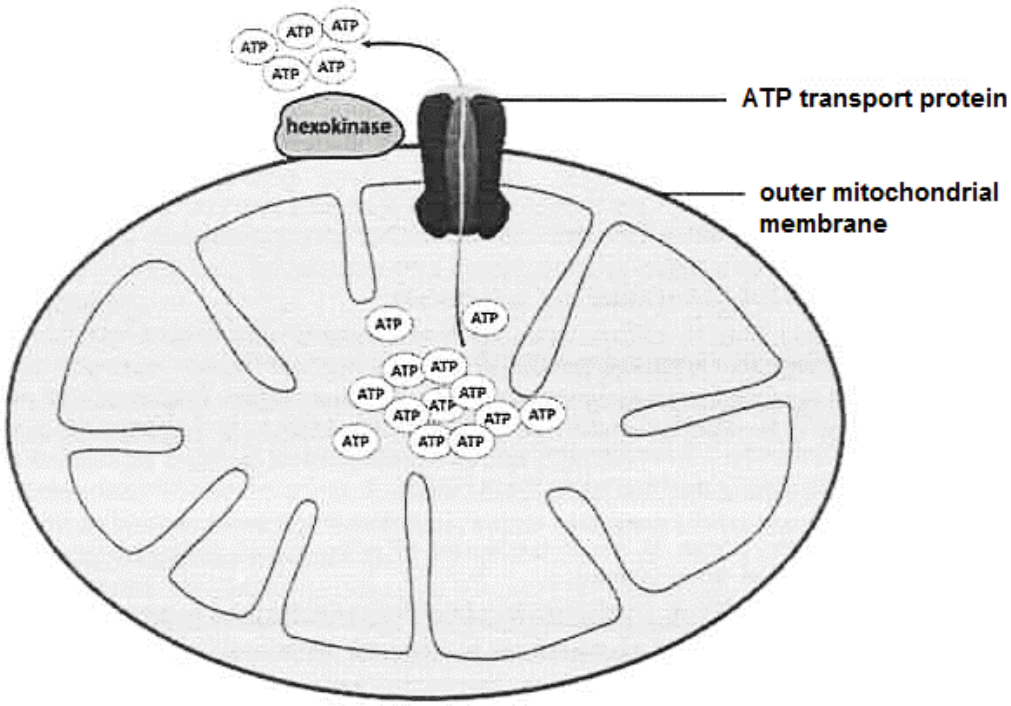

Fig. 6.2

With reference to the role of mitochondria and Fig. 6.2, suggest how the association of hexokinase with mitochondria can lead to high rates of glycolysis. [2]

1. **Mitochondria are the site of aerobic respiration to synthesise ATP;;**
2. **Due to the close proximity of hexokinase to the mitochondria, ATP produced by the mitochondria can easily be used by hexokinase to phosphorylate glucose;; increasing the rate of glycolysis.**

Fig. 6.3 shows an electron micrograph of a mitochondrion.

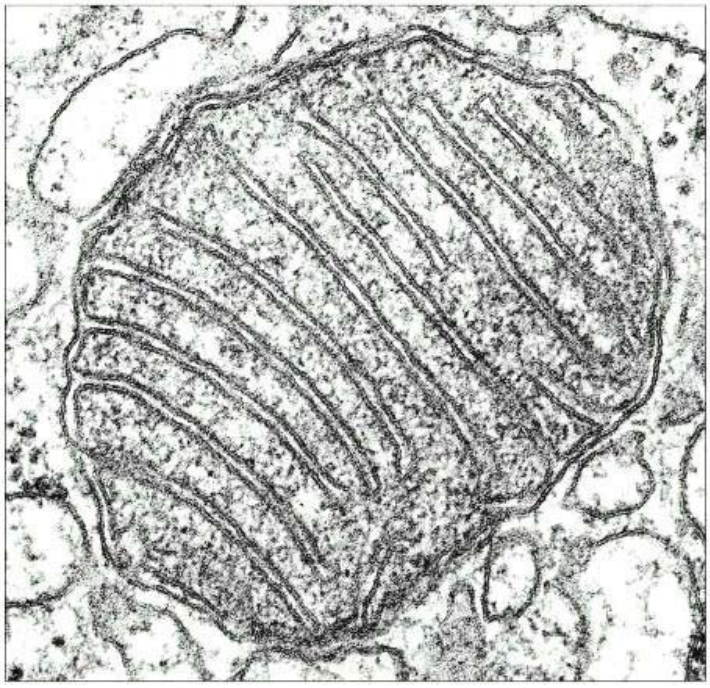

Fig. 6.3

(d) With reference to features visible in Fig. 6.3, outline how the structure of the mitochondrion is adapted for its function. [2]

1. **The inner mitochondrial membrane is <u>highly folded</u>, providing <u>a large surface area</u> where stalked particles, enzymes and electron carriers of the electron transport chain (ETC) *(any 1 e.g.)* needed for <u>aerobic respiration</u> can be located;;**
2. **The mitochondrion is enclosed by <u>double membranes</u> separated by (an extremely narrow fluid-filled space) intermembrane <u>space</u>, allowing for <u>compartmentalisation</u> within the mitochondrion / specialised metabolic pathways to take place in different areas;;**

[Total: 12]

7 Fig. 7.1 shows a flower of *Lilium polyphyllum*, a lily that grows in the Himalayan mountains. This species is cross-pollinated by insects.

Fig. 7.1

(a) Plants of this species that grow at low altitudes produce flowers 60 days before the plants of the same species that grow at high altitudes. Scientists think that plants of *L. polyphyllum* growing at high altitudes may evolve into a new species.

Explain how natural selection could lead to the evolution of a new species of lily. [5]

1. **The subpopulations / low and high altitudes of *L. polyphyllum* become physiologically isolated leading to sympatric speciation;;**
2. **The subpopulations did not interbreed (reproductive isolation due to differing flowering times) and thus gene flow was disrupted (resulting in different species);;**
3. **The subpopulations were exposed to different environments and were thus subjected to different selection pressures;;**
4. **Since there was variation within the subpopulations due to spontaneous mutation,;;**
5. **individuals with favourable characteristics were at a selective advantage and can survive to maturity, (undergo fertilisation / mate), reproduce and passed on their favourable alleles / genes (R:traits) to their offspring (or vice versa);;**
6. **Over successive generations, evolutionary changes occurred independently in each subpopulation;;**
7. **New species of lily have thus arisen by descent with modifications from ancestral species by accumulation of modifications as the population of lily adapt to the new environment;;**

(b) In order for natural selection to occur a population must show phenotypic variation. Explain why variation is important in natural selection. [2]

1. **Variation describes the differences in characteristics shown by individuals belonging to the same species due to presence of different alleles in the individuals;;**
2. **Variation is the raw material, presence of different alleles leading to difference in characteristics, for natural selection to act on;;**
3. **Resulting in differential reproductive success;;**

(c) Fungi were often classified as different species according to their visible reproductive structures. *Penicillium dodgei* and *Eupenicillium brefeldianum* were classified as different species because they had different types of spores.

However, recently it was recognised that the spores of *P. dodgei* were asexual spores, while those of *E. brefeldianum* were sexual spores. A comparison of the DNA of these two fungi shows that they are the same species. This fungus is now known as *Penicillium brefeldianum*.

Outline how DNA analysis can show that *P. dodgei* and *E. brefeldianum* are the same species. [2]

1. **(Homologous DNA sequences) have few differences in DNA bases / sequences;;**
2. **Homologous DNA sequences are identical / more similar in length;;**
3. **Genes are same;;**

(d) Describe the advantages of using DNA analysis in determining homology between *P. dodgei* and *E. brefeldianum*. [3]

1. **Unambiguous and objective. A, T, G, C are easily recognized / one cannot be confused with another. They are not dependent on subjective judgements / observations involving qualitative differences;;**
2. **Quantifiable and can be converted to numerical form and open to statistical and analysis;;**
3. **Homologous regions of DNA from different species provides many points of comparison as each nucleotide position is a point of comparison;; (Each nucleotide position along a stretch of DNA represents an inherited character in the form of one of four DNA bases.)**

[Total: 12]

8 Fig. 8.1 shows part of the immune response to the first infection by a bacterial pathogen that has entered the body through the lining of a bronchiole. J and K are stages in the immune response.

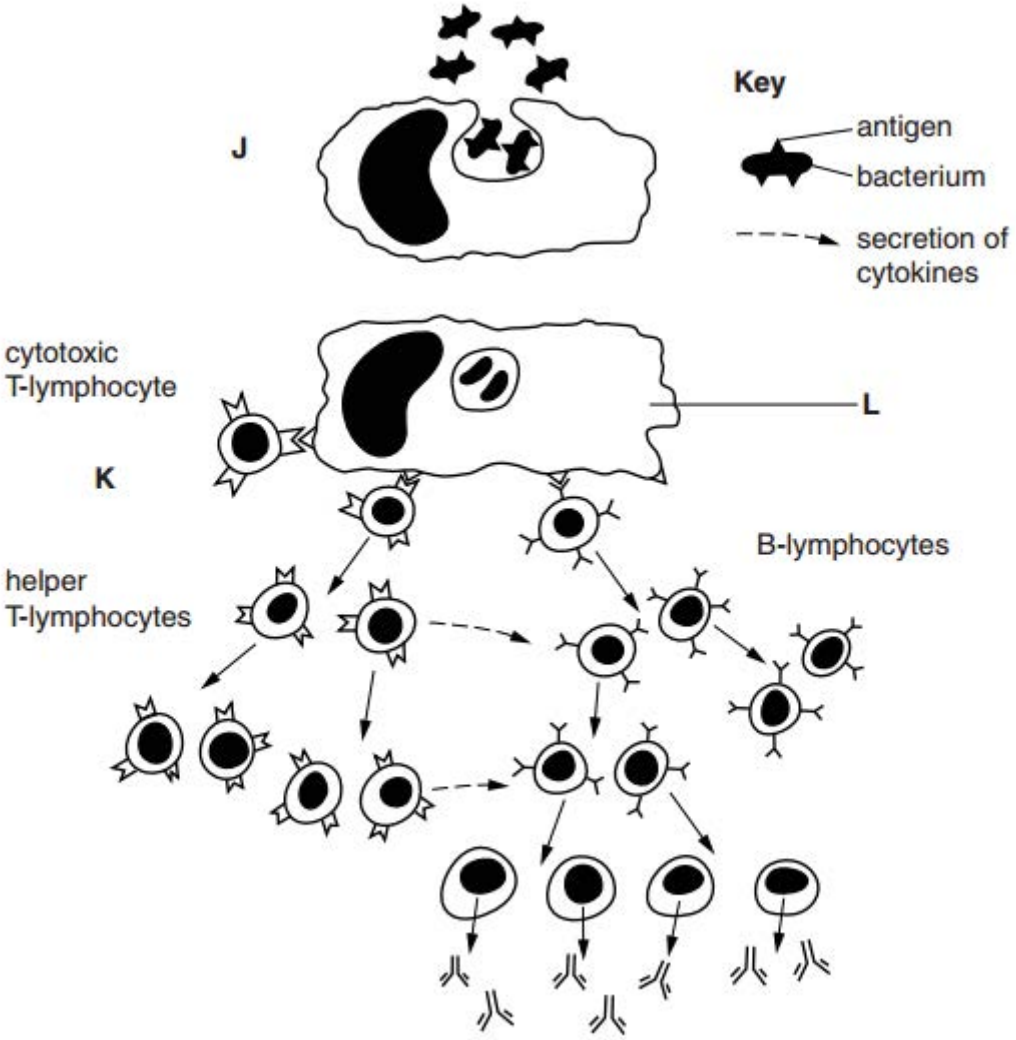

Fig. 8.1

(a) (i) State the process happening at stage J. [1]

1. **Phagocytosis / endocytosis;;**

(ii) Explain the role of cell L at stage K in the immune response. [2]

1. **Digestion of bacteria to destroy bacteria;;**
2. **Antigen presentation on cell surface;;**
3. **Clonal selection / activation of specific B / T-lymphocytes;;**

(b) With reference to Fig. 8.1, explain how the response to a second infection by this bacterial pathogen differs from the first. [3]

1. **Memory (B / T) cells <u>quickly divide by mitosis to form large numbers</u> of effector B / T-lymphocytes (e.g. B-lymphocytes and helper / cytotoxic T-lymphocytes) upon re-exposure to the same antigen;;**
   OR
2. **There is an <u>increased in number</u> of helper / cytotoxic T-lymphocytes specific for this pathogen;;**
3. **There is faster response (for second and subsequent contacts);;**
4. **which increases chances of coming across more pathogens / APCs;;**
5. **<u>Faster production</u> of B-lymphocytes / plasma cells / antibodies / helper T-lymphocytes / cytotoxic T-lymphocytes / cytokines;;**
6. **<u>Greater concentration</u> of antibodies or greater numbers of B / plasma cells;;**
7. **Pathogen removed / killed faster;;**
8. **Person does not become ill / no symptoms;;**

B-lymphocytes have antibodies located on their external surface. When B-lymphocytes become plasma cells they then secrete antibodies.

Fig. 8.2 shows how the enzyme papain digests an antibody to obtain three fragments.

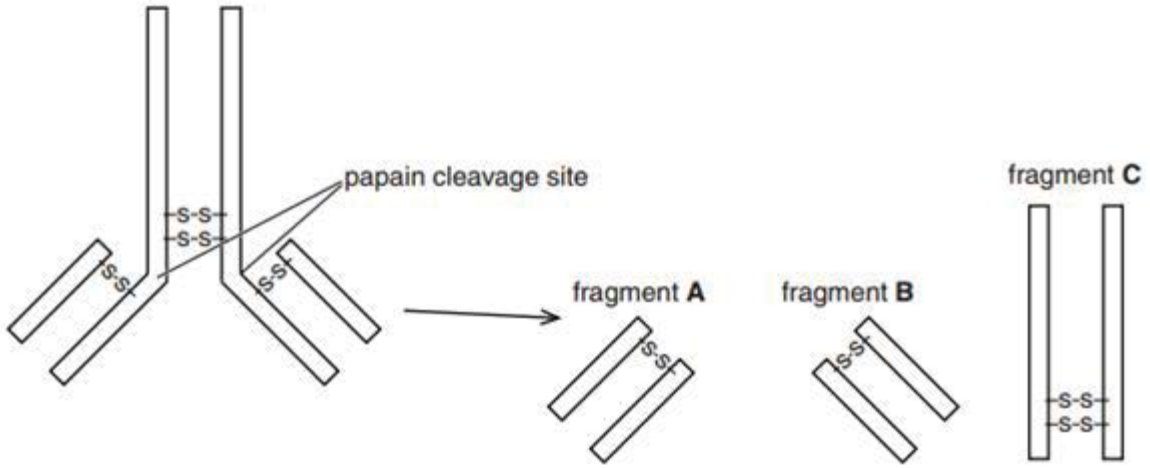

Fig. 8.2

(c) The three fragments, A, B and C still retain their ability to function.

State the function of:

(i) fragments A and B. [1]

1. **Antigen binding sites / bind to antigen / both bind to same (type of) antigen;;**

(ii) fragment C. [1]

1. **Binding to phagocyte / monocyte / macrophage / neutrophil / B-lymphocyte / named cell type with Fc receptor;;**

(d) There are various ways in which the effectiveness of immune responses can be reduced.

Suggest how each of the following reduces the effectiveness of an immune response.

(i) Some pathogens are covered in cell surface membranes from their host. [1]

1. **Pathogens not recognised as non-self / foreign (or vice versa);;**

(ii) B-lymphocytes do not mature properly and do not recognise any antigens. [1]

1. **No antibodies / plasma cells / memory B cells produced;;**
2. **No humoral response;;**
3. **No antigen presentation by B-lymphocytes;;**

[Total: 10]

9 Reef-building corals are marine invertebrates closely related to jellyfishes and are found in shallow, clear tropical seas. The corals secrete an exoskeleton of calcium carbonate that becomes the underlying structure of the coral reef.

Zooxanthellae are a group of unicellular photosynthetic algae that live inside the cells of reef-building corals. The relationship is beneficial to both the zooxanthellae and the coral.

(a) Evidence shows that the relationship between zooxanthellae and reef-building corals has evolved by free-living algae invading corals that did not contain algae. [1]

(i) Corals that do not need zooxanthellae can live at a greater depth that reef-building corals.
Explain why. [3]

1. **Corals without zooxanthellae do not need to rely on light;;**
2. **The corals may have different feeding methods;;**
3. **Reef-building corals with zooxanthellae need light for them to photosynthesise;;**
4. **As depth increases, less light penetration/more light absorbed by the water;;**

(ii) Suggest how the zooxanthellae may benefit in two ways from their association with the corals. [2]

1. **The corals provide zooxanthellae with carbon dioxide for photosynthesis;;**
2. **Protection from predation;;**
3. **Protection from extreme conditions;;**
4. **The corals provide a physical support for zooxanthellae to absorb light;;**
5. **Nitrogen from the coral's nitrogenous waste supports algal growth;;**

Under conditions of environmental stress, the relationship between the reef-building corals and zooxanthellae can break down. Loss of zooxanthellae and the subsequent whitening that occurs, as shown in Fig. 9.1, is known as coral bleaching. Coral bleaching can lead to the death of the coral.

Fig. 9.1

(b) State one reason why permanent loss of zooxanthellae can lead to death of the coral. [1]

1. **Loss of major food source/decreased source of food;;**
2. **Less organic compounds (e.g. sugars);;**
3. **Loss of protective algal layer for corals from harmful effects of sunlight;;**

(c) One type of environmental stress that can cause coral bleaching is an increase in sea temperature.

Suggest why areas of sea with reef-building corals are particularly susceptible to increased temperature as a result of global climate change. [2]

1. **Coral reefs grow in shallow/tropical seas;;**
2. **Shallow water heats up more rapidly/surface waters are warmer than deeper water;;**
3. **Temperature increases may be greater near the equator;;**

[Total: 8]

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JURONG JUNIOR COLLEGE
JC 2 PRELIMINARY EXAMINATIONS
Higher 2

CANDIDATE NAME

CLASS

INDEX NUMBER

BIOLOGY 9744/03

Paper 3 Long Structured and Free-response Questions **11 September 2017**

Candidates answer on the Question Paper. **2 hours**

No Additional Materials are required.

READ THESE INSTRUCTIONS FIRST

Write your class, index number and name in the spaces at the top of this page.
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.

Section A
Answer **all** questions in the spaces provided on the Question Paper.

Section B
Answer any **one** question in the spaces provided on the Question Paper.
Circle the question number of the question attempted.

The use of an approved scientific calculator is expected, where appropriate.
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	
Section A	
1	
2	
3	
Section B	
4 / 5	
Total	

This document consists of **22** printed pages and **1** blank page.

[Turn over

Section A

Answer **all** the questions in this section.

1 The *lac* operon is an operon required for the uptake and metabolism of lactose in *Escherichia coli* and many other bacteria. Glucose is the preferred carbon source for most bacteria, as glucose requires fewer steps and less energy to break down than lactose. However, if lactose is the only sugar available, the *E. coli* uses it as an energy source and the *lac* operon allows for the effective digestion of lactose when glucose is not available.

To use lactose, the bacteria must express the *lac* operon genes, which encode key enzymes for lactose uptake and metabolism. Fig. 1.1 shows the results of an experiment carried out to determine the effects of adding lactose on the expression of some of the genes involved in the breakdown of lactose.

The initial gene expression was measured by determining the mRNA produced at time 0. This was taken as 10% (black bars). All the other values are relative to this initial value taken every 30 seconds over the next 2 minutes.

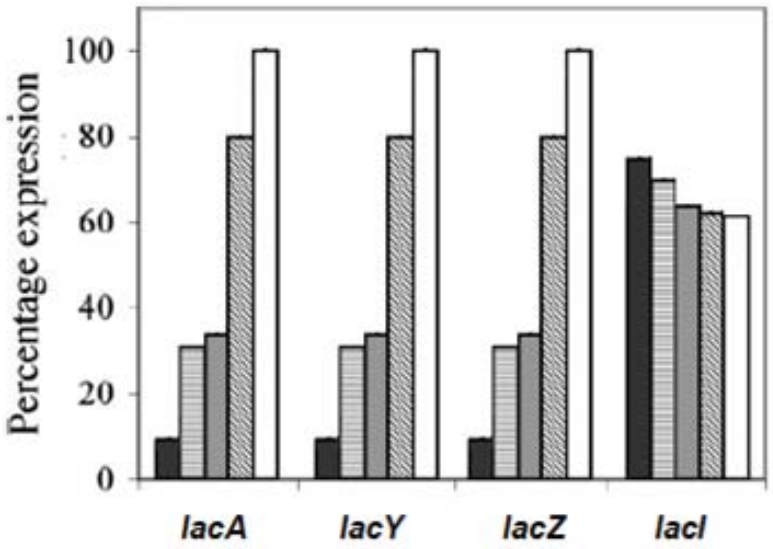


Fig. 1.1

(a) Suggest why operons are necessary in bacteria. [2]

1. **Operons allow for the <u>simultaneous regulation of related genes</u> of related functions which are involved in the same metabolic activity;;**
2. **Genes coding for the proteins/enzymes of a single biochemical pathway / of related functions / same metabolic activity, (such as the catabolism of carbohydrates) are <u>grouped together</u> into an operon for <u>easier control</u>;;**
   OR
3. **Operons allow/enable the simultaneous regulation of related genes to <u>adapt/in response to environmental changes</u>;;**
4. **The genes on the operons are only expressed when required / the bacteria only produce the proteins/enzymes that are required to increase efficiency / conserve/prevent the waste of energy and resources;;**

5. **Thus the bacteria are at a <u>selective advantage</u>;;**

(b) Using data from *lacA*, *lacY* and *lacZ* in Fig. 1.1 and your knowledge of how different types of operons are regulated, explain how lactose is able to control the expression of these genes. [4]

1. **Gene expression increased from 10% at 0 min to 100% at 2 min;;**
2. **Operon is turned on / need to be turned on / a inducible system where expression of *lacA*, *lacY* and *lacZ* are inactive / not constitutively active (10% at time 0);;**
3. **lactose binds to the repressor (and act as a inducer) / allolactose binds to the allosteric site of the lac repressor, the lac repressor protein changes to inactive conformation, thus the repressor protein is unable to bind to the operator;;**
4. **RNA polymerase is able to recognise and bind to the promoter, and transcription of the (structural) genes of the operon led to formation of mRNA;;**
   (resulting in an increase in gene expression)

It has been suggested that not all genes involved in lactose hydrolysis are organised into one single operon.

(c) Use evidence from Fig. 1.1 to support the statement above. [3]

1. ***lacA*, *lacY* and *lacZ* respond to lactose (inducer) to the same extent;;**
2. **percentage expression for *lacA*, *lacY* and *lacZ* increases from 10% at 0s to 100% at 2 min;;**
3. **suggesting that *lacA*, *lacY* and *lacZ* are under the control of the same promoter and operator;;**
   OR
4. ***lacI* does not respond significantly to lactose in the same period of time;;**
5. **percentage expression for *lacA*, *lacY* and *lacZ* increases from 10% at 0s to 100% at 2 min compared to *lacI* which decreases from 80% at 0s to 60% at 2 min (data tbu);;**
6. **suggesting that *lacI* must be found in a different region of the bacterial chromosome, under the control of a different promoter;;**
   OR
7. **Gene expression of *lacA*, *lacY* and *lacZ* increase whereas *lacI* decrease;;**
8. **percentage expression for *lacA*, *lacY* and *lacZ* increases from 10% at 0s to 100% at 2 min compared to *lacI* which decreases from 80% at 0s to 60% at 2 min (data tbu);;**
9. **suggests that *lacI* is not transcribed simultaneously, *lacI* may be controlled by a different promoter;;**
   Mark once for promoter

(d) A series of mutations was introduced into the *lac* operon, resulting in the inversion of the operator and the promoter regions.

Suggest the effect on the transcription of the *lac* genes when lactose is absent. [2]

1. **In the absence of lactose, lac repressor protein binds to the operator region, which is now upstream of the promoter / lac repressor protein does not physically block RNA polymerase;;**
   Accept ref to the effect of the inversion;;
2. **Hence, RNA polymerase can bind to the promoter, transcription of *lac* genes is now always on / cannot be turned off;;**

Owing to *E. coli's* rapid growth rate, *E. coli* has been an expression host of choice in the biotechnology industry for large-scale production of anti-freeze proteins (AFPs). AFPs is a class of polypeptides that help to stop ice forming inside the Arctic and Antarctic fishes thus permitting their survival in sub-zero environments.

In the Arctic and Antarctic, environmental temperatures can reach low to freezing levels. These fishes indigenous to these habitats are presented with potential desiccation, which can lead to potentially detrimental challenges such as decreased enzymatic rates and freezing. Besides hindering cellular processes, sub-zero temperatures induce ice crystals formation, which can lead to cell death by rupturing cells either physically or through osmotic pressure changes.

Commercially, there appears to be countless applications for AFPs:

- as additives to frozen foods to lengthen the shelf life
- incorporation with the genome of the raw foods to retard ice crystal growth
- to prevent damage to agricultural crops by increasing freeze tolerance of crop plants and extending the harvest season in cooler climates
- introduction into ice cream and yogurt products to allow the production of very creamy, dense, reduced fat ice cream with fewer additives.

(e) Outline how the genome of *E. coli* and the genome of the fish are similar and how they are different. [4]

Similar:

1. **both have double-stranded DNA;**

Differences (max 3)

	Feature	Prokaryotic	Eukaryotic	
1.	**Linearity/ circularity**	**Circular / ~~Looped~~ chromosome**	**Linear chromosomes**	**;;**
2.	**Association with proteins**	**Naked / Associated with H-NS proteins (nucleoid-associated proteins)**	**Associate with histones and scaffolding proteins**	**;;**
3.	**Introns**	**Absence of introns**	**Presence of introns**	**;;**
4.	**Genome Size / Number of genes**	**Small / Fewer genes present**	**Large / Many more genes present**	**;;**
5.	**Number of chromosomes**	**Single chromosome**	**Multiple chromosomes**	**;;**
6.	**Origin of replication**	**One per chromosome**	**Many per chromosome**	
7.	**Operons**	**Presence of operons**	**Absence of operons**	

AVP: (pt 5-7) [max 1]
Extra: prokaryotic, in cytoplasm / not membrane bound vs eukaryotic, membrane bound/in nucleus;

(f) Anti-freeze glycoprotein (AFGP) is one type of anti-freeze protein. Messenger RNA coding for AFGP is translated at a ribosome to produce a polypeptide. Describe how this polypeptide is then processed to make AFGP. [4]

1. **In the rER, the AFGP polypeptide will <u>coil and fold</u> into (the geometrically regular) secondary structures/α-helix and β-pleated sheet (held together by hydrogen bonding between C=O and –NH groups of the amino acids);;**
   OR
   The AFGP polypeptide will undergo <u>further bending, coiling, folding</u>, to form the (specific) <u>tertiary structure/3D conformation</u>;;
2. **<u>biochemical modification/ glycosylation</u> takes place / AFGP may be modified by enzymes in the ER lumen that <u>add carbohydrate chains</u> to them;;**
   OR
3. **the protein is released into the lumen of the Golgi body for further modification, sorting and packaging into vesicles;;**
4. **The AFGP is <u>packaged into transport vesicles / Golgi vesicles and transported</u> towards the Golgi body / other parts of the cell;;**
5. **movement from rER to GA;;**

Some fish produce another anti-freeze protein, called AFP II. The tissues of these fish were tested for the presence of AFP II and the mRNA coding for AFP II. The results are shown in Table 1.1.

Table 1.1

molecule	present in
AFP II protein	all tissues
AFP II mRNA	liver tissue only

(g) Explain the distribution of the AFP II protein and AFP II mRNA. [4]

1. **AFP II gene/allele is activated only in liver cells / deactivated in cells other than liver cells;;**
   ALLOW "switched on/off"
2. **activation could be due to DNA demethylation/histone acetylation/activator protein binding to enhancer;;**
   OR
3. **deactivation could be due to DNA methylation/histone deacetylation/repressor protein binding to silencer;;**
4. **transcription and translation/protein synthesis of AFP II occurs/takes place only in liver cells;;**
   Ref to liver cells required only once if context / chain of argument is clear.
5. **the protein/AFP II is secreted from liver cells / transported around the body, presence of protein;;**
6. **AFP II in all tissues prevents freezing/ice forming in all parts of the body;;**

(h) With reference to named examples, describe the roles performed by proteins involved in transport in fishes. [2]

1. **haemoglobin/ myoglobin, for oxygen transport/ storage;;**
2. **membrane carriers/ channels, with role in passive transport;;**
3. **example of carrier/ channel protein, with specific role;;**
   - **GLUT/glucose transporter for glucose transport / aquaporin for water molecules transport / H^+ channel in stalked particle**
4. **membrane pumps/ AW with role in active transport;;**
5. **example of pump/ AW with role;;**
   - **Na^+/K^+ pump for transporting Na^+ (out of the cell) and K^+ (into the cell), H^+ pump along ETC**
6. **further example of membrane transport protein with contrasting role;;**
7. **electron carriers/electron transport chain, components for chemiosmosis/ ATP synthesis/redox reactions;;**

Table 1.2 shows Earth's ice ages over the last 850 million years.

Table 1.2

Ice age	Time / millions of years ago
Quaternary	0 to 2.6
Karoo	260 to 360
Andean-Saharan	420 to 460
Cryogenian	630 to 850

Fig. 1.2 shows how the number of families of fishes has changed over time.

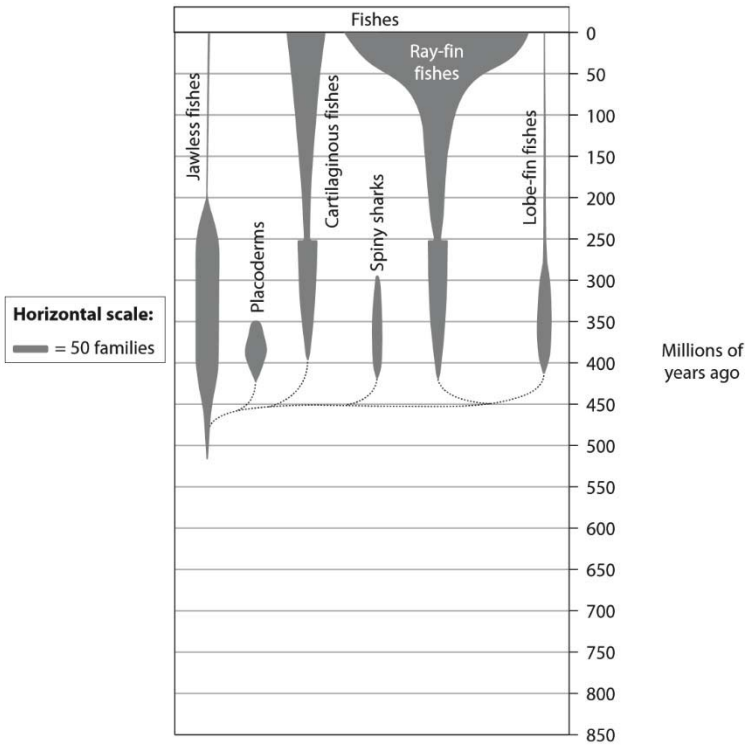


Fig. 1.2

(i) Many different types of AFPs are produced by ray-fin fishes. Analyse the data to explain when these ray-fin fishes are likely to have evolved the ability to produce AFPs. [2]

1. **(sea) ice is a selection pressure for AFPs / AFPs are advantageous (only) when there is (sea) ice;;**
   ALLOW AFPs allow fish to survive the ice age
   OR
2. **so AFPs are likely to have appeared/increased in frequency during an ice age;;**
   OR
3. **the only ice ages since the existence of the ray-fin fish are the Quaternary and Karoo;;**
4. **therefore ray-fin fish producing AFPs are likely to have evolved in the last 2.6 million years / between 260 and 360 million years ago;;**
   ALLOW during the Karoo / Quaternary (ice age)

[Total: 27]

2 Measles is a highly contagious, serious disease caused by *Morbillivirus*, a single-stranded enveloped RNA virus. Envelope glycoproteins mediate transmission of the virus into host cells in the human respiratory tract. Once inside the host cell, the viral RNA genome is transcribed into mRNA, which undergoes translation to manufacture viral proteins. These viral proteins function to form capsid proteins for new viruses which eventually leave the host cell.

(a) With reference to the information given, outline how viruses challenge the concept of what is considered living. [2]

1. **Viruses are acellular and do not contain cytoplasm or cellular organelles;;**
2. **Viruses contain only one type of hereditary material / either DNA or RNA but never both;;**
3. **Viruses must replicate using the host cell's (metabolic machinery) ribosomes, amino acids, enzymes, nucleotides, energy – any 1 e.g.;;**
   OR
4. **Outside of host cells, viruses do not carry out any metabolism / grow or divide;;**
   OR
5. **The new viral components are synthesised and assembled within the infected host cell;;**
   (any 2)

In the 1980s, measles caused an estimated 2.6 million deaths each year, and the disease remains one of the leading causes of death among young children globally.

The number of cases of measles is reported to the World Health Organisation (WHO) by countries throughout the world so that global data is collected.

Fig. 2.1 shows the global data collected between January 2008 and December 2012.

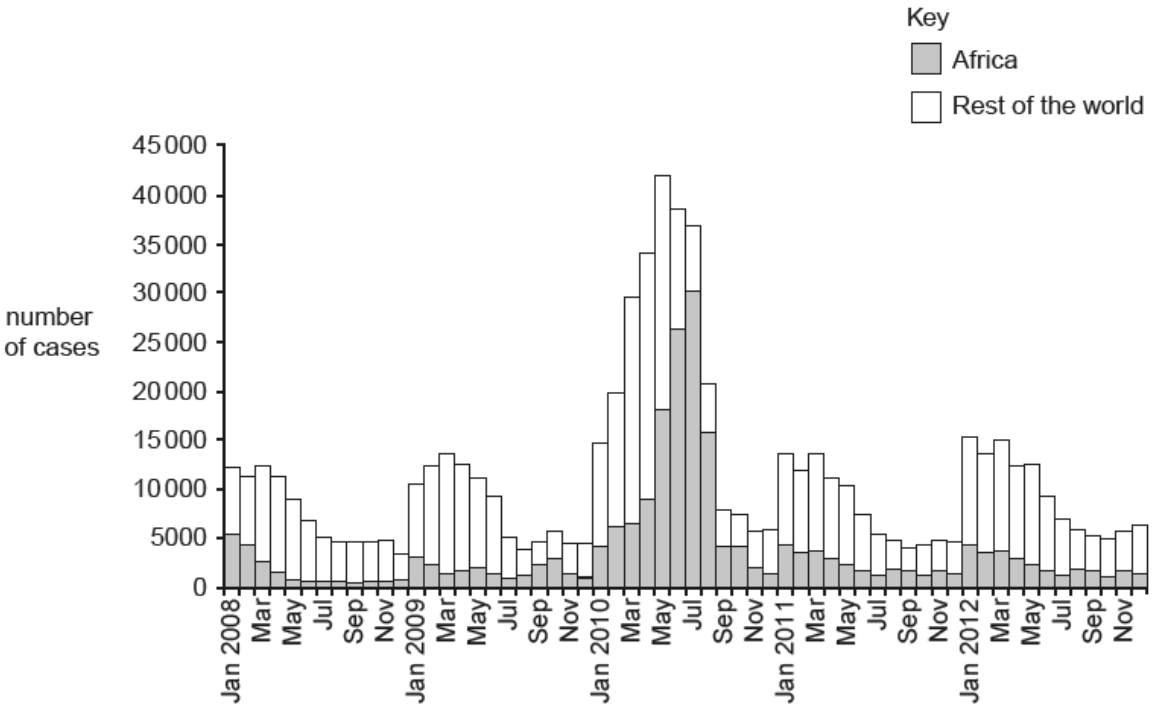


Fig. 2.1

(b) Use the data in Fig. 2.1 to describe the pattern shown in the number of cases of measles reported to the WHO between January 2008 and December 2012. [3]

1. **Number of cases fluctuated between 2008 to 2012 / in all years;;**
2. **Number of cases tends to be higher at the beginning of each year (than at the end of each year), except in 2010;;**
3. **Number of cases in the rest of the world are greater than in Africa each year (or vice versa), except in 2010;;**
4. **Number of cases (much) higher in 2010;;**
5. **with the highest peak/highest total number of *quote number between 41 875 - 42 143* cases in (May) 2010;;**
   OR
6. **with the highest number of 30 000 cases in Africa in (July) 2010;;**
7. **Epidemic lasted longer in 2010;;**
8. **There has been 5 *(accept: 4 since no data before Jan 2008)* outbreaks/epidemics between Jan 2008 and Dec 2012;;**

(c) Routine measles vaccination for children, combined with mass immunisation campaigns in countries with high case and death rates, are key public health strategies to reduce global measles deaths. By 2016, about 85% of the world's children received one dose of the measles vaccine by their first birthday, and the global push to improve vaccine coverage resulted in a 79% reduction in deaths.

(i) State precisely the type of immunity gained by receiving a measles vaccine. [1]

1. **(Adaptive) Acquired active immunity;;**

(ii) Outline one benefit of vaccination. [1]

Benefits of Vaccination

1. **Protect the individual against disease by conferring immunity to the individual without prior exposure to a specific pathogen;;**
2. **Confer lifelong immunity to the individual by providing secondary immune response against subsequent encounter with the same pathogen;;**
3. **Confer herd immunity to unvaccinated individuals in a community (e.g. pregnant women and people with allergies/weakened immune system), reducing possibility of transmission between individuals / unvaccinated individuals have a very low risk of becoming infected;;**
4. **Contribute to the elimination / eradication of infectious diseases within human population (if pathogen relies only on human as host);;**

(iii) Outline one risk of vaccination. [1]

Risks

1. **Side effects/adverse reactions after vaccination have been observed in small numbers of individuals (e.g. life-threatening allergic reaction, fainting and rashes etc.);;**
2. **Some individuals are more susceptible to vaccination risks than others (e.g. individuals with weakened immune systems);;**
3. **(for live attenuated vaccines) Pathogen used in vaccines may regain its virulence and cause disease;;**

(d) Unlike measles for which an effective vaccine has been developed, it has been extremely difficult to design an effective vaccine against malaria. Malaria is a disease caused by the parasite *Plasmodium falciparum*. *P. falciparum* multiplies in liver cells of the host before emerging after 9-30 days wrapped in the liver cell surface membrane. They enter red blood cells, multiply and then cause rupture of the host cells, resulting in the release of more parasites every 36-48 hours, in a manner that has some similarity to that of viruses.

Use the information given and your own knowledge to suggest why it has been extremely difficult to design an effective vaccine against malaria. [2]

1. **The parasite <u>mostly stays inside host cells</u>, thus <u>evading the host immune system</u>;;**
2. **The parasite <u>mostly stays inside host cells/uses liver cell membrane as 'covering'</u> and is therefore <u>disguised as 'self'/non-foreign</u>;;**
3. **Difficulty in designing a vaccine which <u>stimulates both cell-mediated and humoral responses</u> to bring about parasite clearance and provide future immunity against the parasite;;**
4. **<u>Diversity/large degree of variation/change in parasite antigens</u> such that existing <u>antibodies</u> targeting the old antigens <u>bind poorly/cannot recognise and bind</u> to the new antigenic sites, allowing parasites with new antigens to evade immunological memory against the original parasite;;**
   (Any 2)

(e) Another infectious disease, Tuberculosis (TB), is one of the top ten causes of death worldwide. Name the bacterium that causes TB and describe how TB is transmitted. [3]

1. ***Mycobacterium tuberculosis*;; *(penalise for spelling)***
2. **The bacteria are transmitted from person to person through fine <u>aerosol droplets</u>;; (accept: airborne)**
3. **formed when an <u>infected person</u> with the active disease <u>sneezes/coughs/ breathes</u> (accept: spits/talks);;**
   OR
4. **droplets are <u>inhaled by an uninfected person</u>;;**

[Total: 13]

3 Dengue fever is a disease spread by a particular species of mosquito, *Aedes aegypti.* The incidence of this disease and the numbers of this species of mosquito have increased dramatically in recent years, spreading beyond the tropics. This has been attributed to global warming.

(a) Explain how global warming has resulted in the spread of dengue beyond the tropics. [2]

1. **Global warming results in <u>increase in geographical/distribution range of *A. aegypti*</u> as they have <u>increased survival in its new location</u> (that used to be too cold) / are extending increasingly into temperate zones;;**

2. **Also results in <u>increase in number of mosquito vectors</u> as <u>higher temperatures lead to increased metabolism / accelerates their development</u>;;**
   OR
3. **Increases activity of female mosquitoes and reduce the incubation time for them to become infectious / transmit DENV;;**
   OR
4. **Rate of viral replication within vector will increase and extrinsic incubation period (before DENV becomes transmissible to another host) will shorten;;**

In an attempt to reduce the numbers of *A. aegypti*, male mosquitoes infected with the *Wolbachia* bacteria have been produced and released into the wild to mate with females. *Wolbachia* naturally occurs in up to 60% of all insect species, but not in *A. aegypti. Wolbachia* induces a conditional sterility that occurs within the mosquitoes due to cytoplasmic incompatibility, shown in Fig. 3.1, a concept first introduced in a paper published by Dr. Hannes Laven in 1967.

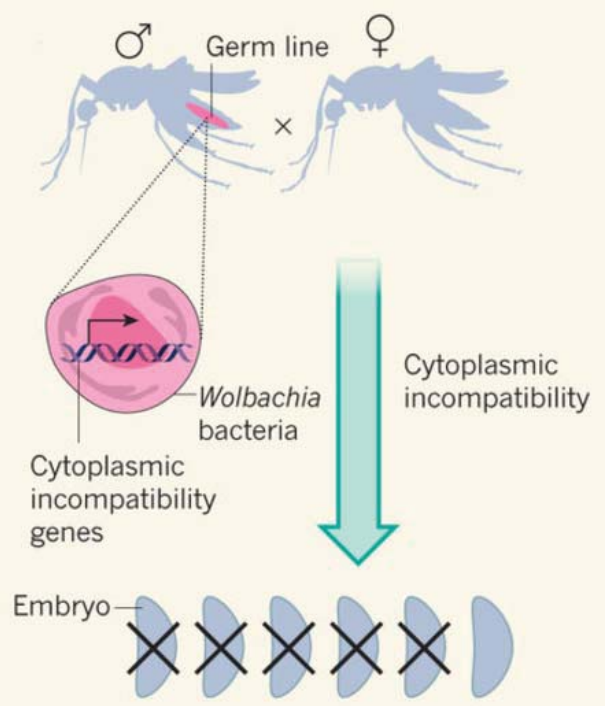


Fig. 3.1

(b) The cytoplasmic incompatibility genes are DNA in nature. DNA is a double helix consisting of two polynucleotide strands held together by phosphodiester bonds between the adjacent nucleotides. Each strand contains a sugar-phosphate backbone and hydrogen bonds are formed between the complementary strands via complementary base pairing.

Describe two other structural features of DNA. [2]

1. **The two strands coil around each other in a right-handed double helix;;**
2. **The strands are antiparallel/run in opposite directions/one strand runs in the 5' to 3' direction while the complementary strand runs in the 3' to 5' direction;;**
3. **Each nucleotide comprising of a deoxyribose, a phosphate group and one of the four nitrogenous bases - Adenine, Thymine, Cytosine or Guanine;;**
4. **The nitrogenous bases are arranged as side groups of the chains (oriented toward the central axis);;**
5. **The width between the 2 sugar-phosphate backbones is constant at 2nm, equals to the width of 1 base pair i.e. 1 purine + 1 pyrimidine;;**
6. **One complete turn of the double helix measures 3.4nm in length and comprises 10 base pairs;;**

There are three possible matings between the male and female mosquitoes in the wild as shown in Table 3.1.

Table 3.1

	male	female	results
cross 1	infected	uninfected	lay eggs that are not viable and do not hatch
cross 2	infected	infected	infected offspring
cross 3	uninfected	infected	infected offspring

Embryonic development aborts when sperm from an infected male fertilises an uninfected egg and the paternal genome does not contribute to the development of the embryos that are not viable.

(c) Based on the information provided and Cross 1, suggest how *Wolbachia* induces sterility. [1]

1. **Cytoplasmic incompatibility genes in *Wolbachia* are transcribed and translated to form <u>proteins</u> / *Wolbachia* secretes a (DNA binding) <u>protein</u> that <u>binds to the sperm/paternal DNA/chromosome</u>;;**

In 1970, Erich Jost repeated Laven's earlier work.

(d) Explain why repeating the work of others is an important part of science research. [2]

1. **To test/check results/findings;;**
2. **(if results support) to build scientific consensus / increase confidence (A: reliability, R: accuracy) in the findings;;**
3. **(if results different) to revise/refine the theory/model/hypothesis;;**

In 2016, hundred thousands of male mosquitos with *Wolbachia* were released at 3 selected sites in Singapore: Braddell Heights, Nee Soon East, and Tampines West.

(e) State why releasing such large numbers of male mosquitoes did not immediately increase the risk of transmission of dengue fever in these estates. [1]

1. **Only females spread dengue;;**
2. **Males do not bite / feed on blood;;**

Another method employed in Australia involves the release of both male and female mosquitoes with *Wolbachia* into the wild. An advantage of this method is that there is no need for further releases of mosquitoes with *Wolbachia.*

(f) With reference to Table 3.1, explain why there is no need for further releases with this method. [2]

1. **Infected female mosquitoes can mate with both uninfected and infected males to produce infected offspring;;**
2. **Passing the bacteria from generation to generation;;**
3. **Over time, the percentage of mosquitoes carrying *Wolbachia* grows until it remains high without the need for further releases;;**

[Total: 10]

Section B

Answer **one** question in this section.

Write your answers on the lined paper provided at the end of this Question Paper.

Your answers should be illustrated by large, clearly labelled diagrams, where appropriate.

Your answers must be in continuous prose, where appropriate.

Your answers must be set out in sections **(a)**, **(b)** etc., as indicated in the question.

4 (a) Describe the reproductive cycle of the influenza virus and explain how new strains of the virus may arise as a result of mutation. [13]

Adsorption

1. **The haemagglutinin glycoproteins (on the viral envelope) recognise and bind to (sialic acid containing) receptors on the host cell plasma membrane;;**

Penetration and Uncoating

2. **The influenza virus enters the host cell by receptor-mediated endocytosis;;**
3. **whereby the host cell membrane invaginates (and pinches off), engulfing / placing the virus in an endocytic vesicle;;**
4. **Acidification causes the viral envelope to fuse with the endocytic vesicle membrane;;**
5. **releasing the nucleocapsid into the cytoplasm of the cell;;**
6. **The capsid is then degraded by cellular enzymes, releasing the viral nucleic acid;;**

Replication

7. **The viral genome functions as the template for synthesis of complementary RNA strand (cRNA);;**
8. **using the viral RNA-dependent RNA polymerase (and host RNA nucleotides in the nucleus);;**
9. **This cRNA acts as a template for the synthesis of new copies of viral RNA (genome);;**
10. **and functions as mRNA which undergoes translation to produce viral proteins (e.g. capsid proteins and glycoproteins for the viral envelope);;**

Maturation

11. **The glycoproteins synthesised (by ribosomes on the ER) are transported to the (host cell's) plasma membrane via vesicles and incorporated into the plasma membrane;;**
12. **The capsid is assembled around the viral RNA genome and the RNA-dependent RNA polymerase;;**

Release

13. **The new influenza viruses bud off from the host cell's plasma membrane / ref. to budding, resulting in the new viruses acquiring their lipid bilayer from the plasma membrane of the host cell, with viral proteins and glycoproteins embedded;;**
14. **Neuraminidase catalyses the cleavage of sialic acid residues from haemagglutinin, facilitating the release of the newly replicated viruses (for the next round of infection);;**

[max 11m]

Antigenic drift

15. **Point mutations in haemagglutinin gene in influenza virus occur during the replication of viral RNA;;**
16. **Accumulation of mutations results in slight changes to the shape of haemagglutinin;;**
17. **leading to antigenic drift;;**
18. **Shape of the new haemagglutinin is now complementary to other membrane receptors found on new types of host cells;;**

QWC
Good spread of knowledge communicated without ambiguity to include:
at least 1 MP from each of the 2 sections – reproductive cycle of virus (pt 1-14) and antigenic drift (pt 15-18);;

(b) Describe the process of transduction and its advantages to prokaryotes. [12]

1. **Transduction is the process by which bacterial DNA / genes is transferred from one bacterium (host cell) to another (recipient cell) via a bacteriophage;;**

2. **Generalised transduction requires infection of a bacterium by a virulent bacteriophage (and any random portion of the bacterial DNA transferred);;**
3. **During the assembly of the newly replicated phage genome within the phage capsid, a small piece of the host cell's degraded DNA gets mistakenly packaged within the capsid (defective phage);;**

4. **Specialised transduction requires infection of a bacterium by a temperate bacteriophage (and only bacterial genes adjacent to the integrated prophage transferred);;**
5. **When a temperate phage enters into lytic cycle from lysogenic cycle / spontaneous induction occurs;;**
6. **Small region of the bacterial DNA that was adjacent to the prophage is excised;;**
7. **The phage-host hybrid DNA is packaged within a capsid (defective phage);;**

8. **The defective phages infect / attach to other bacterial cells and inject the piece of host bacterial DNA into the newly infected bacterial cell cytoplasm;;**

9. **(Generalised & Specialised transduction) Foreign bacterial DNA is incorporated into the recipient / bacterial cell's chromosome through homologous recombination;;**
   OR
10. **(Generalised & Specialised transduction) If there is sufficient homology between the DNA fragments and bacterial chromosome, crossing over occurs, and segments of the original chromosomal DNA will be replaced;;**

11. **(Specialised Transduction) Phage-host hybrid DNA integrates into the recipient cell's chromosome, as phage enters the lysogenic cycle;;**

Advantages

12. **New alleles can then be incorporated into the bacterial genome of recipient cell;;**
13. **Recipient cell will subsequently express new characteristics / show change in phenotype;;**
14. **Generate genetic diversity / variation;;**
15. **(Example of new allele) Gain antibiotic resistant alleles from other bacteria;;**
16. **(How it confers selective advantage) Antibiotics less effective against the bacteria;;**
17. **Thus the bacteria is at a selective advantage;;**

QWC:
scientific argumentation exemplified by:
two or more advantages to prokaryotes (pt 12-17) linked coherently to the correct stage of the process;;

5 (a) Describe the roles of the proteins involved in the process of DNA replication and compare the advantages of PCR with the advantages of DNA replication. [13]

1. **Helicase causes the DNA molecule to unwind and unzip;;**
2. **hydrogen bonds between complementary bases break, causing the 2 parental DNA strands to separate;;**
3. **single-strand DNA binding proteins bind to the 2 separated parental DNA strands;;**
4. **to stabilise the single-stranded DNA formed;;**
5. **Primase catalyses the formation of a short RNA primer (– the start of a new strand in the 5' to 3' direction);;**
6. **DNA polymerase (then binds to the RNA primer and) adds nucleotides to the free 3' end of the RNA primer/existing strand;;**
7. **DNA polymerase catalyses the formation of phosphodiester bonds between the nucleotides;;**
8. **RNA nucleotides of all the RNA primers are replaced with DNA nucleotides by another DNA polymerase;;**
9. **DNA ligase seals the gaps between the DNA fragments;;**
10. **by catalysing the formation of phosphodiester bonds between adjacent nucleotides (to form a continuous strand);;**

Similarities

11. **Both processes allow for the production of large amounts of DNA;;**
12. **PCR can be fully automated as Taq polymerase can withstand high temperatures without being denatured, DNA replication also do not require replacement of enzymes / OWTTE;;**
13. **It is easy to set up and use a thermal cycler for PCR, DNA replication also takes place in the nucleus easily;;**

Differences

14. **PCR has high sensitivity and can amplify sequences from minute amounts of target DNA while replication require the entire template/cannot amplify sequences from minute amounts;;**
15. **Millions of copies of target DNA can be obtained in a relatively short period of time/in a few hours in PCR while DNA replication requires a longer period/10 to 12 hours;;**
16. **PCR is robust and can amplify specific sequences from material in which the DNA is badly degraded/embedded in a medium while DNA replication cannot replicate badly degraded DNA;;**
17. **PCR is a specific process which amplifies only target sequences while DNA replication results in the entire DNA sequence being replicated;;**

QWC:
Scientific argumentation exemplified by:
Two or more direct comparisons of the advantage of PCR and/or DNA replication, each clearly set out to show its similarity with, or difference between, **both** PCR and DNA replication;;

(b) Outline the structure of G-protein linked receptor and describe the action of glucagon on liver cells in the regulation of blood glucose concentration. [12]

Structure of G-protein linked receptor

1. **G-protein linked receptor consist of a single polypeptide chain coiled and folded into a tertiary structure;;**
2. **held together by hydrogen bonds, ionic bonds, disulfide bonds and hydrophobic interactions;;**
3. **comprised of seven transmembrane α-helices;;**
4. **has different binding sites for signal molecule/ligand and G protein;;**
   OR
5. **extracellular part/domain of G-protein linked receptor serve as the binding site for (specific) signal molecule/ligand;;**
6. **intracellular parts/domain of G-protein linked receptor serves as binding site for G-protein;;**
   pt 4 or pt5&6: award once
7. **embedded in and span the plasma membrane, held by weak hydrophobic interactions;;**
8. **Non-polar R groups of amino acid residues on the receptor form hydrophobic interactions with non-polar hydrocarbon tails of the membrane phospholipid molecules;;**
9. **The extracellular parts of G-protein linked receptors may be glycosylated (as they serve as the binding site for ligands);;**

Action of glucagon on liver cells

10. **Glucagon recognises and binds to the specific binding site of G-protein linked receptor on the liver cell membrane;;**
11. **and induces a conformational change in the receptor;;**
12. **The receptor now binds to G protein and activates it;; (A molecule of GTP replaces the GDP on the G protein)**
13. **The activated G-protein dissociates from the receptor and activates adenyl cyclase;;**
14. **Adenyl cyclase catalyses the conversion of ATP to cyclic AMP (cAMP);;**
15. **The cAMP then acts as a second messenger;;**
16. **and triggers downstream signalling events/phosphorylation cascade such that glycogen phosphorylase is activated;;**
17. **Glycogen phosphorylase will catalyse the breakdown of glycogen to glucose/glycogenolysis;;**
18. **Glucagon also stimulates an increase in the rate of conversion of amino acids and glycerol to glucose/ gluconeogenesis;;**
19. **so that the blood glucose concentration increases and return back to normal levels.**

QWC
Good spread of knowledge communicated without ambiguity to include:
at least 2 MP from each of the 2 sections – structure of G-protein linked receptor (pt 1-9) and action of glucagon on liver cell (pt 10-18);;

H

JURONG JUNIOR COLLEGE
JC 2 PRELIMINARY EXAMINATIONS
Higher 2

CANDIDATE NAME

CLASS

BIOLOGY 9744/04

Paper 4 Practical **15 August 2017**

2 hours 30 minutes

Candidates answer on the Question Paper.

Additional Materials: As listed in the Confidential Instructions.

READ THESE INSTRUCTIONS FIRST

Write your name and class on all the work you hand in.
Give details of the practical shift and laboratory, where appropriate, in the boxes provided.
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.

Answer **all** questions in the spaces provided on the Question Paper.

The use of an approved scientific calculator is expected, where appropriate.
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.

Shift
Laboratory

For Examiner's Use	
1	
2	
3	
Total	

Answer **all** questions.

1 During the light dependent stage of photosynthesis, hydrogen ions and electrons are transferred to hydrogen acceptor molecules, including NADP.

DCPIP (2,6-dichlorophenolindophenol) is a blue dye, which acts as a hydrogen ion and electron acceptor. As DCPIP accepts hydrogen ions or electrons it is reduced and becomes colourless.

You are required to investigate the effect of different wavelengths of light on the rate of the light dependent stage of photosynthesis in a leaf extract containing chloroplasts.

You are provided with:
- a leaf extract in buffered solution, labelled **L**, in a beaker with ice,
- DCPIP solution, labelled **D**,
- filters that allow light of specific wavelengths to pass through, as shown in Table 1.1.

Table 1.1

colour	label	wavelength / nm
purple	P	425
blue	B	450
green	G	525
orange	O	625
red	R	675

The leaf extract is an irritant. It is recommended that you wear safety goggles/glasses and gloves.

Proceed as follows.

1. Stir the leaf extract, **L**, using the glass rod.

2. Use a syringe to draw up 0.5 cm^3 of **L**.

3. Wipe the outside of the syringe to remove any liquid.

4. Put the syringe at the centre of a white tile. This will be used as a colour standard.

5. You are required to add enough DCPIP solution, **D**, to change the colour of the remaining leaf extract, **L**. The change in colour must be sufficient to be observable in the 0.5 cm^3 sample transferred to a syringe in step **8**.
   - Using a Pasteur pipette, put about 0.5 cm^3 of DCPIP solution, **D**, into the remaining leaf extract, **L**, in the specimen tube.
   - Shake the specimen tube gently so that the colour spreads evenly.
   - Tilt the specimen tube and view the colour against a white background.
   - If there is no noticeable colour change, add DCPIP solution, **D**, drop by drop until a noticeable colour change is achieved.

6. Immediately wrap the specimen tube containing the mixture of **L** and **D** in foil. Cover the specimen tube with a foil lid, as shown in Fig. 1.1. This should be easy to remove to obtain the mixture of **L** and **D**. Put back the covered specimen tube containing the mixture of **L** and **D** in the beaker with ice.

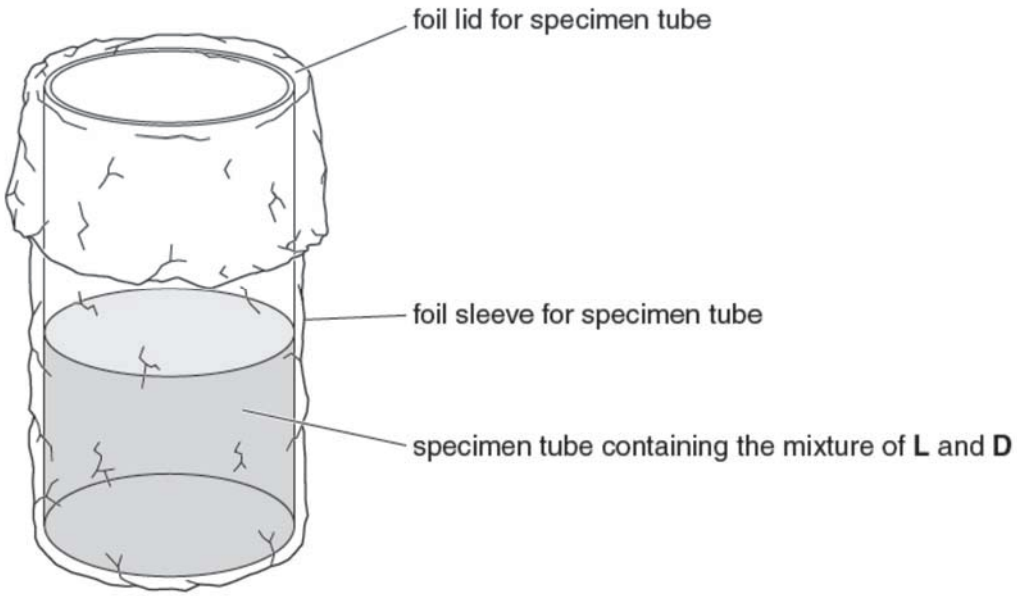


Fig. 1.1

7. Place the bench lamp 10 cm from the syringe on the white tile. Do **not** switch the lamp on.

   The next steps have to be carried out very quickly one after another, so read steps 8–15 and refer to Fig. 1.2 before proceeding.

8. Remove the foil lid and use a clean syringe to draw up 0.5 cm^3 of the mixture of **L** and **D** in the specimen tube. Replace the foil lid immediately.

9. Wipe the outside of the syringe and place it next to the colour standard on the white tile. This is the test syringe.

10. Immediately cover both syringes with the purple filter, **P**, as shown in Fig. 1.2 on page 4.

11. Switch on the bench lamp and immediately start a stopwatch or stop clock.

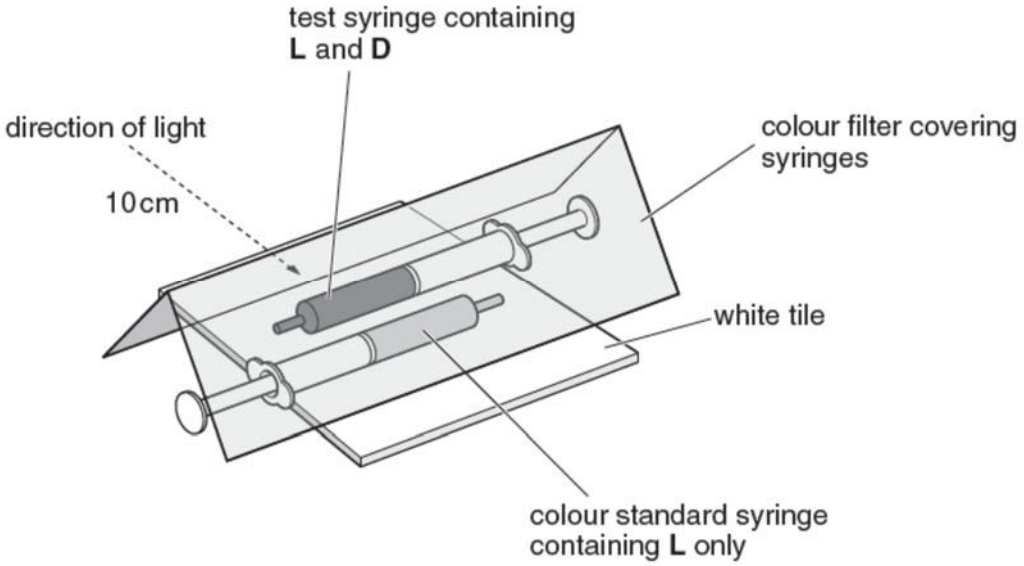


Fig. 1.2

12. In the space provided in **(a)**, record the time taken for the colour in the test syringe to match that of the colour standard. If the colour does not match after 300 seconds then record 'more than 300'.

13. Switch off the bench lamp.

14. Expel the contents of the test syringe into the beaker labelled **waste**. Rinse the syringe.

15. Repeat steps **8–14** using each of the four remaining coloured filters in turn (blue, green, orange and red).

(a) Record these results in a suitable table in the space provided to show the effect of wavelength on the time to decolourise DCPIP. [3]

Table showing the effect of wavelength on the time to decolourise DCPIP

Wavelength / nm	Time to decolourise DCPIP / s
425 (purple)	113
450 (blue)	241
525 (green)	more than 300
625 (orange)	155
675 (red)	142

1. **Suitable column / row headings with correct units;;**
2. **Recording time in whole number of seconds;;**
3. **Record 425 nm (purple) is fastest and 525 nm (green) is slowest or 'more than 300' and results recorded for all five filters;;**

(b) (i) Give one reason to explain why the leaf extract was kept on ice. [1]

1. **To prevent the enzymes in the extract from damaging the chloroplasts;;**

(ii) State why the leaf extract containing DCPIP was kept covered by foil. [1]

1. **To prevent photosynthesis / light dependent reaction occurring and decolourising the DCPIP;;**

(iii) Describe a suitable control that could have been set up for this investigation. [1]

1. **Description of a tube containing DCPIP and water / boiled leaf extract only;;**
2. **Description of a tube containing DCPIP and leaf extract in the dark;;**

(c) Suggest why the rate of photosynthesis is different at different wavelengths. [2]

1. **Some wavelengths are used more effectively than others for photosynthesis or uses data to make the same point, e.g. purple / red / orange wavelengths are the most effective;;**
2. **Chlorophyll absorbs some wavelengths of light more than others or uses the data to make the same point, e.g. chlorophyll absorbs purple / blue wavelengths and orange / red / long wavelengths;;**
3. **Some wavelengths release more H^+ ions / electrons than others, e.g. purple / red / orange wavelengths;;**

(d) Suggest two significant sources of error in this experiment and describe two corresponding improvements that could be made to reduce the effects of these errors. [4]

1. **Difficulty in matching the colour of the standard by eye;;**
2. **Use a colorimeter;;**

3. **Difficulty in observing the colour all the time because of keeping the filter in place;;**
4. **Filter in front of the syringes so colour can be seen from behind;;**

5. **Leakage of light through the ends of the folded filter;;**
6. **Remove all other light sources;;**

7. **There is warming up / increase in temperature as experiment proceeds due to the heating effect of the lamp;;**
8. **Use of a heat filter or cool light source;;**

In another similar investigation, a student collected leaves from two varieties of the same species of a garden plant that has different coloured leaves

Variety A dark red leaves
Variety B green and white striped leaves

The student made a chloroplast extract from the leaves of each variety and measured the rate of photosynthesis for each extract in different wavelengths of light.

Table 1.2 shows the rates of photosynthesis calculated by the student from her results.

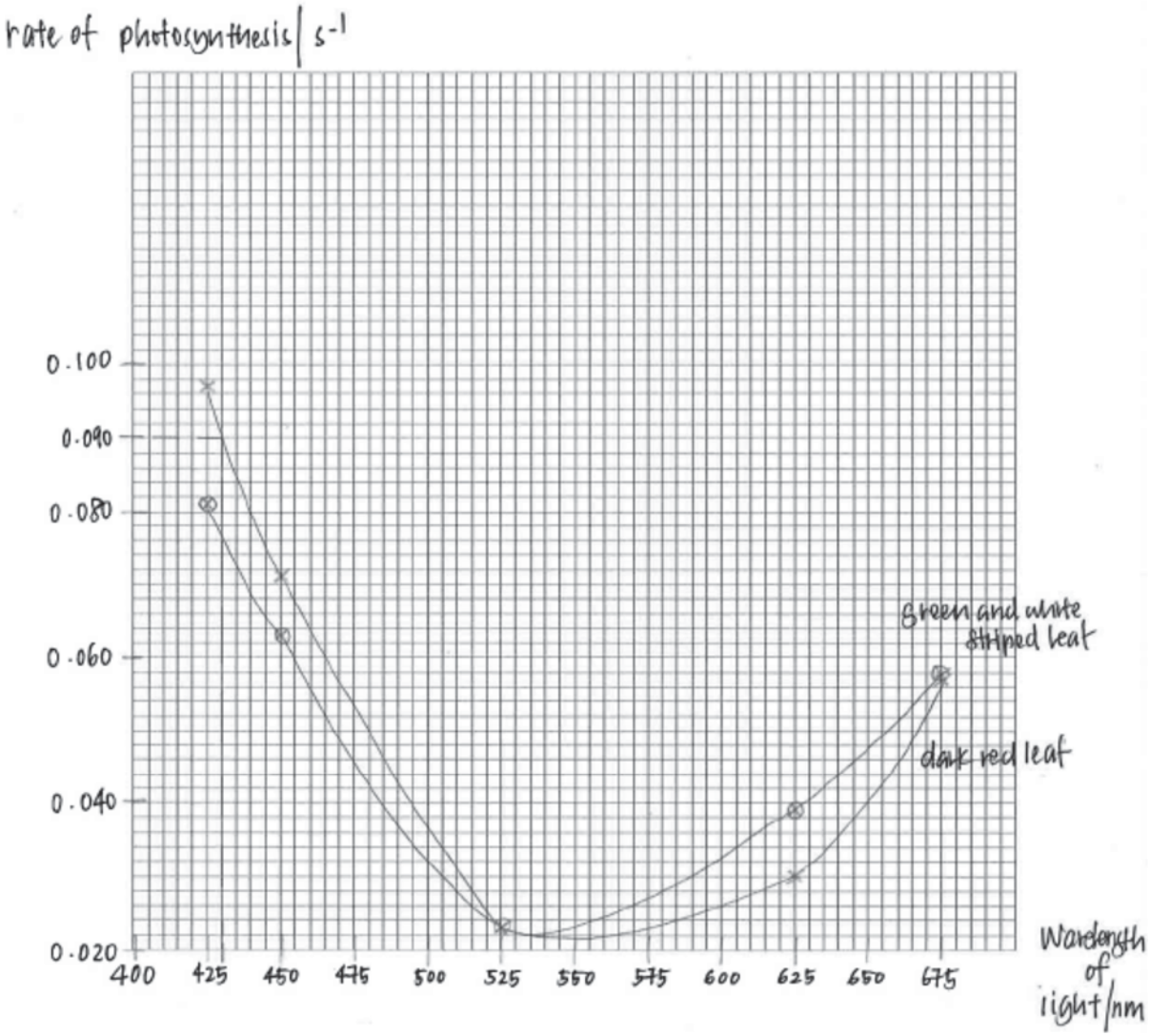
Table 1.2

wavelength of light / nm	rate of photosynthesis / s^{-1}	
	source of chloroplasts	
	dark red leaf	green and white striped leaf
425	0.097	0.081
450	0.071	0.063
525	0.023	0.023
625	0.030	0.039
675	0.057	0.058

(e) Use the grid provided to plot line graphs showing the effect of wavelength on the rate of photosynthesis. [4]

1. **Axes correctly labelled with correct units;;**
2. **Scaled appropriately with ascending scale and equidistant intervals, with a scale such that plotted points occupy at least 50% of the graph paper in both the x and y directions;;**
3. **All values from student's calculations of rate plotted correctly to ± half a small square on the graph paper provided;;**
4. **Line graph showing best-fit line + 2 clearly labelled graph drawn;;**

Graph of rate of photosynthesis s^{-1} against wavelength of light / nm



(f) Use you graph to estimate the rate of photosynthesis at a wavelength of 450 nm in plants with dark red leaves. [1]

1. **0.090 s^{-1};;**

(g) Explain how light of wavelength 450 nm leads to the decolourisation of DCPIP. [1]

1. **Light energy causes release of electrons from chlorophyll / from photolysis of water and electrons reduce the DCPIP;;**

(h) The photosynthetic pigments of the leaves from the two varieties of plants were extracted and were separated by two-way chromatography. The pigments were first separated by one solvent and then separated again by a second solvent at right angles to the first solvent. Fig. 1.3. shows the results for the two different varieties.

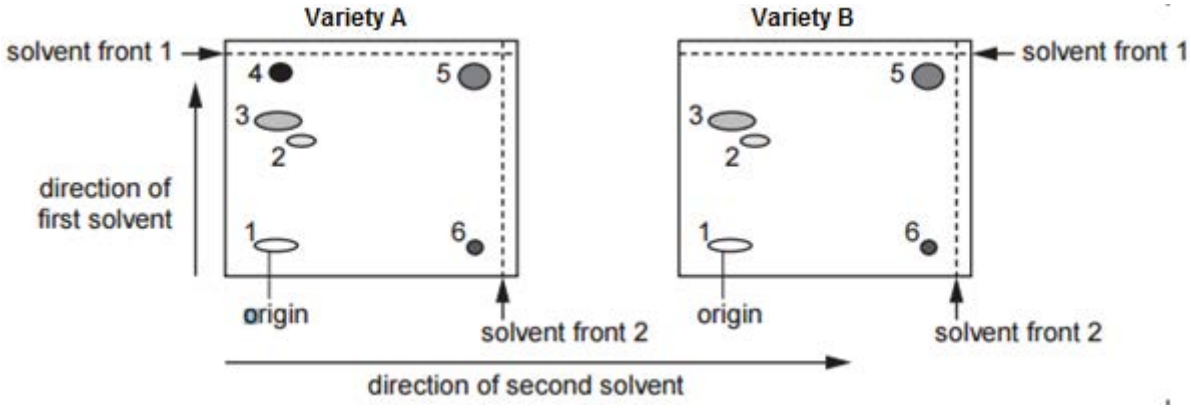


Fig. 1.3

Different photosynthetic pigments absorb different wavelengths of light. Table 1.3 shows some information about the pigments, P, Q, R, S and T, found in the 2 varieties, including the wavelength of light at which maximum light absorption occurs.

Table 1.3

pigment	wavelength of light / nm	Rf value	
		solvent 1	solvent 2
P	620	0.20	0.89
Q	545 and 547	0.60	0.29
R	420 and 660	0.65	0.11
S	490	0.91	0.19
T	430 and 645	0.82	0.92

$$Rf = \frac{\text{distance moved by pigment}}{\text{distance moved by solvent front}}$$

One of the varieties lacks one of the pigments.
Using the information in Table 1.3 and Fig. 1.3:

(i) identify the variety that lacks one of these pigments and state the letter of the missing pigment. [1]

1. **Variety B and pigment S;;**

(ii) state the evidence that supports your answer to (i). [2]

1. **Chromatogram for Variety B has a pigment / spot / number 4 missing;**
2. **At about Rf 0.91 (in solvent 1) / Rf 0.19 (in solvent 2) / it has the highest Rf in solvent 1 / a low Rf in solvent 2;;**

[Total: 22]

2 Methylene blue stains dead cells blue.
Living cells are not stained blue so they will appear white or clear.

You are provided with:
- methylene blue solution, M, (handle carefully as it will stain your skin)
- suspensions of yeast cells, labelled S1, S2 and S3.

Each suspension, S1, S2 and S3 has been heated for ten minutes at 45°C or 80°C or 100°C.

You are required to:
- use the microscope to observe the colour of the yeast cells from S1, S2 and S3, after M has been added
- record your observations by using annotated drawings of three yeast cells from each of S1, S2 and S3
- identify the temperature at which each of S1, S2 and S3 was heated.

1. Label three microscope slides S1, S2 and S3.

2. Place one drop of S1 onto slide S1 and add one drop of M. Mix carefully using a glass rod. (If M comes into contact with your skin rinse with cold water.)

3. Repeat step 2 with S2 and S3.

4. Leave for five minutes.

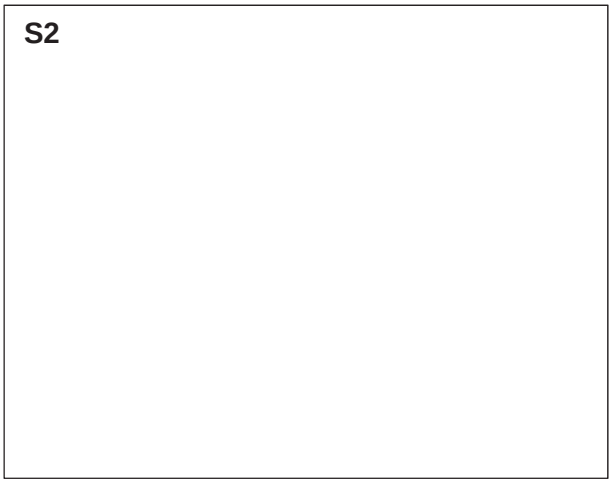
5. Add a coverslip to each slide.

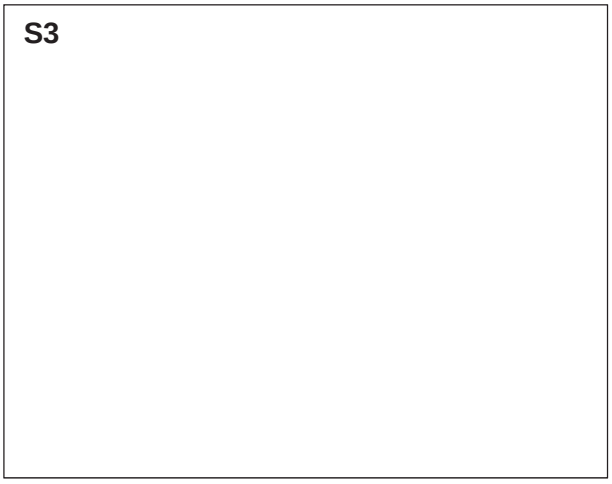
6. Use the paper towel to dry off any excess liquid around the coverslip.

7. Use the microscope to observe the yeast cells on each slide, then select cells which you can draw and annotate to describe the effect of the methylene blue, M.

(a) (i) Prepare the space below and record your observations by:
- making drawings of three cells from each of the slides in the boxes provided
- annotating your drawings to describe the effect of methylene blue, M on the cells. [4]

S1





1. **At least 9 separate cells in total drawn in boxes S1, S2 and S3 + size at least <u>10mm across smallest cell</u>, in any box;;**
2. **<u>Drawing and quality</u>: Do not give mark for any shading, any ruled lines, or any line is too thick, has any feathery or dashed lines or gap in line, has any overlaps;;**
3. **Drawn only <u>3 cells</u> in each of the three boxes;;**
4. **At least one colour stated for each of the cells in the boxes S1, S2 and S3;;**

(ii) Use your observations to identify the temperature that was used to heat each of the suspensions S1, S2 and S3.
Complete the table. [1]

suspension	temperature / °C
S1	**100**
S2	**45**
S3	**80**

(iii) Explain how you identified the yeast cells that had been heated at 100 °C. [1]

1. **Yeast cells blue + therefore inactive or dead;;**

(b) Using the eyepiece graticule fitted in the eyepiece lens of your microscope, and the stage micrometer, find the actual length, in μm, of one of the yeast cells that you have drawn in S2.

Show the measurements that you made and your working. [3]

1. **shows division of stage micrometer measurement by number of eyepiece graticule divisions;;**
2. **shows measurement of yeast cell from S2 in eyepiece graticule divisions;;**
3. **(conversion of measurement from S2, in eyepiece graticule divisions, to) correct answer to calculation in μm;;**

x40 objective

20 graticule units	**=**	**10 micrometer divisions**
	=	**10 x 0.01 mm**
	=	**0.10 mm = 100 μm**
So 1 graticule unit	**=**	**5.00 μm (3 sf);;**

Length of a yeast cell = _____ of graticule units;;

Length of a yeast cell = _____ μm;;

Actual length of a yeast cell = …….... μm

(c) Draw a straight line on your drawing across the yeast cell to show where you took your measurement. [1]

Use your knowledge of the actual size of the yeast cell to calculate the magnification of your drawing. [1]

1. **Magnification of drawing = Length of the drawing / actual length of guard cells**

= _____ ;;

(d) The yeast *Rhodotorula glutinis* produces an enzyme, α-arabinofuranosidase, that could be used in the production of compounds to enhance the flavour and smell of fruit juices. The effect of the initial pH of the culture medium on the growth rate of this yeast was tested. Three continuous culture systems were set up, each with a different initial pH. The cultures were sampled at hourly intervals for 20 hours at each pH. The mean growth rate was then calculated.

The mean growth rates with their standard deviations are shown in Table 2.1.

Table 2.1

pH	mean growth rate / arbitrary units h^{-1}
4.0	0.156 $\pm$ 0.001
5.2	0.197 $\pm$ 0.013
7.0	0.037 $\pm$ 0.011

A t-test was carried out on the results for pH 4.0 and pH 5.2 and gave the value,

$$t = 2.4$$

The degree of freedom is 38.

Based on the findings of the *t*-test, a student concluded that pH 5.2 was optimum for the production of the enzyme α-arabinofuranosidase by *R. glutinis*. Suggest two reasons why this conclusion may not be valid. [2]

1. **Only three pH values tested / only 2 pH values (used for *t*-test);;**
2. **No data between pH 4 and 5.2 / 5.2 and 7;;**
3. **Only growth measured;;**
4. **Yield of enzyme might be higher at different pH than optimum growth;;**

(e) A student carried out *t*-test on the results to compare the lengths of yeast cells when grown in different media.

A number of *t*-test was carried out to find out if, after 70 minutes, the difference in mean yeast cell length is significant:

1. between medium A and medium B $t = 2.50$
2. between medium A and medium C $t = 3.56$
3. between medium B and medium C $t = 1.94$

Table 2.2 shows the critical values for the *t*-test.

The number of degrees of freedom is 18.

Table 2.2

degrees of freedom	10	12	14	16	18	20	22	24	26	28	30	40	50	60
probability 0.05	2.23	2.18	2.14	2.12	2.10	2.09	2.07	2.06	2.06	2.05	2.04	2.02	2.01	2.00
probability 0.01	3.17	3.06	2.98	2.92	2.88	2.85	2.82	2.80	2.78	2.76	2.75	2.70	2.68	2.66

State what conclusions can be drawn about the significance of the differences in mean lengths from the three values of t given above. [3]

1. **Critical value (at p >0.05) is 2.10;;**
2. **A + B (1) and A + C (2) have values greater than the critical value / 2.10 / p<0.05**
   OR
   B + C (3) has value less than critical value / 2.10 / p>0.05;;
3. **A + B / A + C results are significant / not due to chance / caused by an environmental factor**
   OR
   B + C results are not significant / due to chance;;
4. **(A + B / A + C) Differences due to chance is less than 1 in 20 / 0.05 probability;;**
5. **A + C (also) significant at >p = 0.01;;**

(f) Fig. 2.1 is a photomicrograph of a stained transverse section through part of a plant leaf. This plant species is native to part of Asia.

You are not expected to have studied this leaf.

Fig. 2.1

Draw a large plan diagram of the part of the leaf shown in Fig. 2.1. On your diagram, use a ruled label line and label to show the vascular bundle. [4]

1. **At least 2 lines for upper epidermis and 2 lines for lower epidermis + one enclosed area + size at least 80mm for depth of midrib + no shading;;**
2. **No cells + one enclosed area (vascular bundle);;**
3. **Correct proportion of vascular bundle in relation to distribution of tissues in midrib;;**
4. **Uses label line and label to vascular bundle;;**

Low power / Plan drawing of transverse section through part of a plant leaf

[Total: 20]

3 A number of plant tissues are coloured because the cells contain chemicals called betacyanins.

You are provided with beetroot which contains betacyanins which coloured the beetroot red.

In this experiment, you will test the effect of two different alcohols – methanol and ethanol on beetroot membranes. Ethanol is found in alcoholic beverages. Methanol, sometimes referred to as wood alcohol, can cause blindness and death.

If beet membranes are damaged, the red pigment will leak out into the surrounding environment. The intensity of color in the environment should be proportional to the amount of cellular damage sustained by the beet.

Plan an investigation to find out whether or not betacyanin leakage for beetroot occurs at the same intensity using ethanol and methanol.

You must use:
- beetroot
- 40% ethanol
- 40% methanol
- colourimeter

You may select from the following apparatus and use appropriate additional apparatus:
- normal laboratory glassware e.g. test-tubes, boiling tubes, beakers, measuring cylinders, graduated pipettes, glass rods etc.
- stopwatch
- distilled water
- white tile
- scalpel
- forceps

Your plan should:
- have a clear and helpful structure such that the method you use is able to be repeated by anyone reading it
- be illustrated by relevant diagram(s), if necessary, to show, for example, the arrangement of the apparatus used
- identify the independent and dependent variables
- describe the method with the scientific reasoning used to decide the method so that the results are as accurate and repeatable as possible
- include layout of results tables and graphs with clear headings and labels
- use the correct technical and scientific terms
- include reference to safety measures to minimise any risks associated with the proposed experiment.

[Total: 14]

(a) Independent and dependent variables

1. **State and describe independent variable;;**
2. **State and describe dependent variable;;**

- Independent variable is concentration of alcohols – 0%, 10%, 20%, 30%, 40% ethanol and methanol.

- Dependent variable is absorbance.

(b) Controlled/fixed/standardised, variables to improve accuracy or reliability

3. **Identifies at least two variables to be controlled;;**
4. **Describes how two identified variables are controlled;;**

- Size / shape / number of beetroot pieces used.
  - Use a fixed number of pieces for each experiment.
- Source / age of beetroot pieces
  - All beetroot pieces must come from the same beetroot.
- Volume of alcohol used
  - 10 cm^3 used for each experiment.
- Duration
  - 5 min for each experiment.
- pH
  - Use a constant volume of pH buffer for all experiments.
- Temperature
  - Use a constant temperature of 35°C by placing test-tubes in a thermostatically controlled water bath.

(c) Scientific reasoning

5. **Describe the method with the scientific reasoning used to decide the method so that the results are as accurate and repeatable as possible;;**

- Reference to phospholipids / proteins of membrane being affected + Explanation of how membrane component is affected e.g. movement or solubility of phospholipids / denaturation of proteins leading to effect on the permeability of the membrane (cell or vacuole);;
- Suitable trend suggested for stated factor e.g. Increasing alcohol concentration increase membrane permeability + Result in more betacyanins / pigment / red colour leaking from the cells / vacuoles;;
- Membrane permeability can be measured by measuring the extent of leakage of pigment using a colourimeter to give an absorbance value;;
  (Max 2)

(d) Describe the experimental procedure (method)

6. **Plans suitable method to vary the concentration of ethanol and methanol;;**

- Describe simple dilution methods + Table showing how to dilute 40% ethanol and methanol to get 10%, 20%, 30%.

7. **Plans suitable method to get equal amounts of beetroot pieces for ethanol / methanol and experiment;;**
8. **Wash and rinse beetroot in distilled water to remove any pigments which may leak out during cutting;;**

1. Cut beetroot to cubes of 1cm by 1cm by 1cm (or any reasonable fixed sizes).
2. Wash and rinse beetroot in distilled water to remove any pigments which may leak out during cutting.
3. Place 5 beetroot cubes (any reasonable numbers) into a test tube containing 10 cm^3 of 10% ethanol.

9. Specifies how to ensure all beetroot pieces are exposed to the alcohols / are submerged in the alcohols;;

4. Carefully agitate the tube to make sure that the beetroot pieces do not stick together / Ensure that all beetroot pieces are submerged in the alcohols.

10. Plans a suitable procedure that involves monitoring the betacyanins leakage in response to different concentration of alcohols over a set period of time;;
11. Devises a method that uses tubes, beetroot pieces, ethanol and methanol;;
12. Specifies method of monitoring and recording betacyanins leakage;;

5. Place test-tube in a waterbath / thermostatically controlled waterbath maintained at 35-40˚C (to choose only one temperature).
6. Immediately start timing for 5 minutes using a stopwatch.
7. After 5 minutes, transfer 1 cm^3 of solution from test-tube to a cuvette and measure the colour intensity using a colorimeter. Record the absorbance values.

8. Repeat step 1-7 to obtain another 2 readings.
9. Repeat step 1-8 for 20%, 30%, 40% and 0% (control) ethanol.
10. Repeat entire experiment twice.
11. Repeat step 1-10 for the different concentrations of methanol.
12. Plot a graph of absorbance / A.U. against concentration of ethanol and methanol / %.

(e) Draw and annotate relevant diagram(s) – marking points may be credited from diagrams. e.t.c.

(f) Ensuring reliability and accuracy
13. Describe how to ensure reliability (replicates and repeats);;
14. Describe how to ensure accuracy (more or wider range of concentrations used);;

(Reliability)
- Repeat two more times with different / new beetroot pieces and alcohols.

(Accuracy)
- Repeating with more or wider range of alcohol concentrations.

For teacher perusal
- Insufficient number of values of independent variables e.g. only at 0%, 10%, 20%, 30% and 40%, so lack of results between intervals, changes could be missed so making conclusions about trends are less valid.
  Modification: Include intermediate concentrations within the range e.g. 5%,15% ,25%, 35%.
- Insufficient range of independent variables (0-40%), lack of results beyond the range investigated could make it difficult to identify a trend or pattern.
  Modification: extend the range e.g. 0-60%.

(g) Recording

15. Shows how results are to be presented in the form of tables with independent (alcohols concentration) and dependent variables (absorbance) in appropriate columns / rows;;

Table showing absorbance value with increase concentration of ethanol

Concentration of ethanol / %	Absorbance value / A.U.
0	
10	
20	
30	
40	

Table showing absorbance value with increase concentration of methanol

Concentration of methanol / %	Absorbance value / A.U.
0	
10	
20	
30	
40	

(h) Risks/safety

16. Refers to hazards and precaution;;

Risks	Safety Precautions
Thermometer and test tube breakage.	Remove the thermometer / test tube from the water bath immediately after use and place it in a safe place e.g. drawer / test tube rack, to prevent it from rolling onto the floor.
Scalpel are sharp objected that should be handle carefully.	Place the sharp objects away from the main work area after use.
Ethanol and methanol are highly flammable.	Ensure that there is no naked flame nearby.
Methanol can cause blindness when splashed into the eyes.	Wear goggles and wash eyes immediately with cold water when methanol splashed into the eyes.

(i) Control

17. refers to control experiment without alcohols;;

- $10cm^3$ of distilled water to replace $10cm^3$ alcohols.

(j) Finding if pigment leakage occurs at the same intensity using ethanol and methanol

18. Description that if the 2 graphs do not superimposed on each other then pigment leakage do not occur at the same intensity (vice versa);;