

8.4 The control of gene expression (A-Level Only) - Gene technologies 2 – Questions

Q1.

(a) Plasmids are often used as vectors in genetic engineering.

(i) What is the role of a vector?

(1)

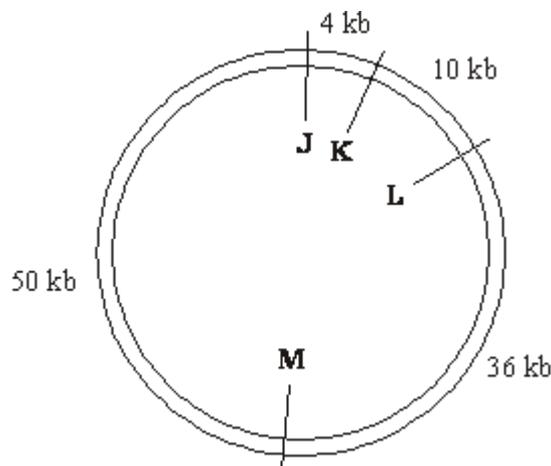
(ii) Describe the role of restriction endonucleases in the formation of plasmids that contain donor DNA.

(2)

(iii) Describe the role of DNA ligase in the production of plasmids containing donor DNA.

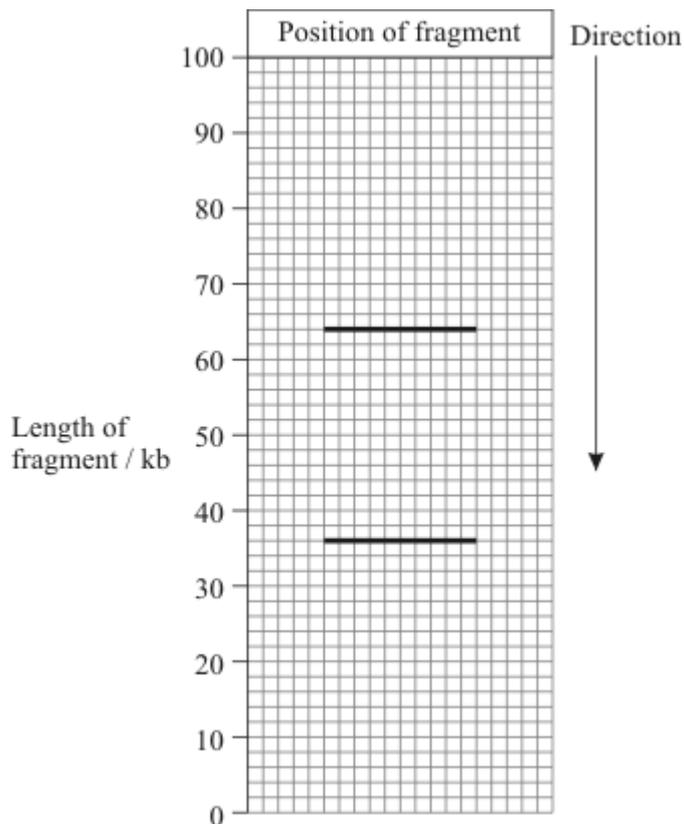
(1)

(b) There are many different restriction endonucleases. Each type cuts the DNA of a plasmid at a specific base sequence called a restriction site. The diagram shows the position of four restriction sites, **J**, **K**, **L** and **M**, for four different enzymes on a single plasmid. The distances between these sites is measured in kilobases of DNA.



1 kb = 1 kilobase

The plasmid was cut using only two restriction endonucleases. The resulting fragments were separated by gel electrophoresis. The positions of the fragments are shown in the chart below.



(i) Which of the restriction sites were cut?

(1)

(ii) Explain your answer.

(1)

(Total 6 marks)

Q2.

The polymerase chain reaction (PCR) can be used to produce large quantities of DNA. Describe how the PCR is carried out.

(Total 6 marks)

Q3.

A protein produced by a species of bacterium is toxic to caterpillars. The gene coding for this protein was removed and transferred into a crop plant.

- (a) (i) Describe how the gene could have been removed from the bacterial DNA.

(2)

- (ii) Many copies of the isolated gene were required. Name the process used in a laboratory to produce many copies of DNA from a small amount.

(1)

- (b) The gene was injected into isolated cells from the crop plant. These cells were then cloned and new plants grown from the cloned cells. Explain the advantage of inserting the gene into isolated plant cells rather than directly into cells within a whole plant.

(3)

(Total 6 marks)

Q4.

(a) Explain the reason for each of the following in the polymerase chain reaction (PCR).

(i) DNA is heated to 95 °C.

(1)

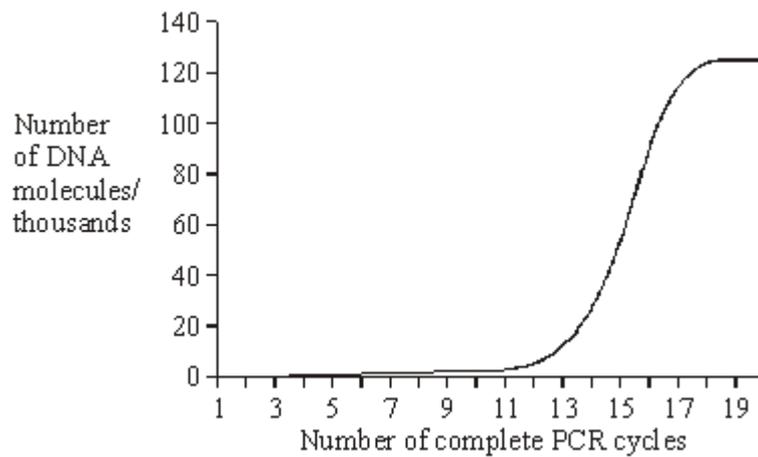
(ii) DNA polymerase used is heat-stable.

(1)

(iii) The reaction mixture is cooled to 40 °C.

(1)

(b) The graph shows the number of DNA molecules made using PCR, starting with one molecule.



(i) Explain the shape of the curve from cycles 1 to 16.

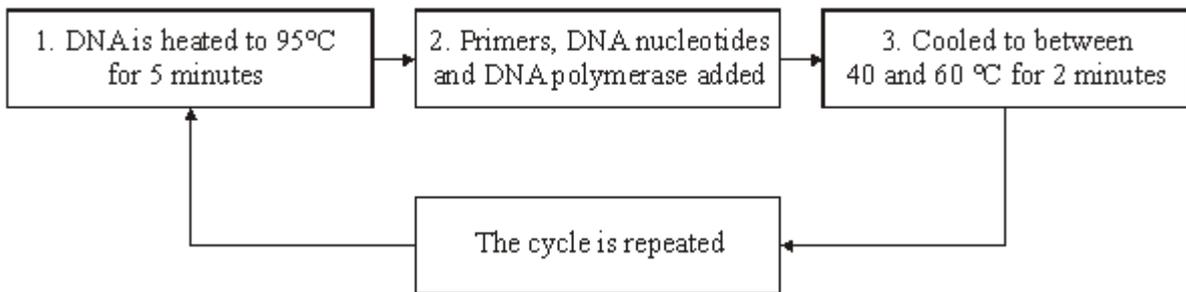
(2)

(ii) Suggest **one** explanation for the levelling out of the curve from cycles 17 to 20.

(2)
(Total 7 marks)

Q5.

The polymerase chain reaction is a process which can be carried out in a laboratory to replicate DNA. The diagram shows the main stages involved in the polymerase chain reaction.



(a) Explain why DNA is heated to 95 °C.

(1)

(b) What is the role of

(i) a primer in this process;

(1)

(ii) DNA polymerase?

(1)

(c) (i) How many DNA molecules will have been produced from one molecule of DNA after 6 complete cycles?

(1)

(ii) Suggest **one** use of the polymerase chain reaction.

(2)
(Total 8 marks)

Q7.

Read the following passage.

Malaria is a disease so deadly that it has devastated armies and destroyed great civilisations. It has been estimated that in the course of history malaria has been responsible for the death of one out of every two people who have ever lived. Even today, with all the advantages of modern technology, it is still responsible for some three million deaths a year.

5 The first half of the twentieth century was a time of hope for malarial control. The drugs chloroquine and proguanil had just been discovered and there seemed a real possibility of a malaria-free world. Unfortunately, this honeymoon ended almost as soon as it had started, with the emergence of drug-resistant parasite populations. Scientists now accept that whatever
10 new drug they come up with, it is likely to have a very limited effective life. As a result, they are increasingly looking at combinations of drugs.

The approach to malaria control which holds the best hope is the production of a vaccine. One of these is being developed by a researcher in South America. His vaccine is based on a small synthetic polypeptide called SPf66 which is dissolved in a saline solution and given as an injection. A series of early trials on human volunteers produced confusing results. In one trial
15 the effectiveness of the vaccine was claimed to be 80% while, in others, the results were statistically insignificant. Not only were the results inconclusive but the methods used were challenged by other scientists. In particular, the controls were considered inappropriate.

Another, possibly more promising, approach has been the development of a DNA-based vaccine. In theory, all that is required is to identify the DNA from the parasite which encodes
20 key antigens. Unfortunately, scientists have hit snags. Although they have succeeded in sequencing the human genome, the genome of the malarial parasite has created major difficulties. This is partly because of the very high proportion of the bases adenine and thymine. In some places these two bases average 80%, and on chromosomes 2 and 3 nearly 100% of the bases present are adenine and thymine. Because of this, it has proved impossible
25 to cut the relevant DNA with the commonly available restriction enzymes into pieces of a suitable size for analysis.

Use information from the passage and your own knowledge to answer the following questions.

(a) Explain how a resistant parasite population is likely to arise and limit the life of any new anti-malarial drug (lines 8 - 9).

(3)

- (b) A person has a 1 in 500 probability of being infected by a chloroquine-resistant strain of malarial parasite and a 1 in 500 probability of being infected by a proguanil-resistant strain. Use a calculation from these figures to explain why scientists are “increasingly looking at combinations of drugs” (lines 9 - 10).

(2)

- (c) (i) Explain why trials of the SPf66 vaccine needed a control.

(1)

- (ii) The controls for the SPf66 vaccine trials were considered inappropriate (line 17).

Suggest how the control groups in these trials should have been treated.

(2)

- (d) In some of the DNA of a malarial parasite, the proportion of adenine and thymine bases averages 80% (lines 22 - 23). In this DNA what percentage of the nucleotides would you expect to contain

(i) phosphate; _____

(ii) guanine? _____

(2)

- (e) (i) Use your knowledge of enzymes to explain why restriction enzymes only cut DNA at specific restriction sites.

(3)

- (ii) Restriction enzymes that can cut the DNA of chromosomes 2 and 3 produce pieces that are too small for analysis. Explain why these restriction enzymes produce small DNA fragments.

(2)

(Total 15 marks)

Q8.

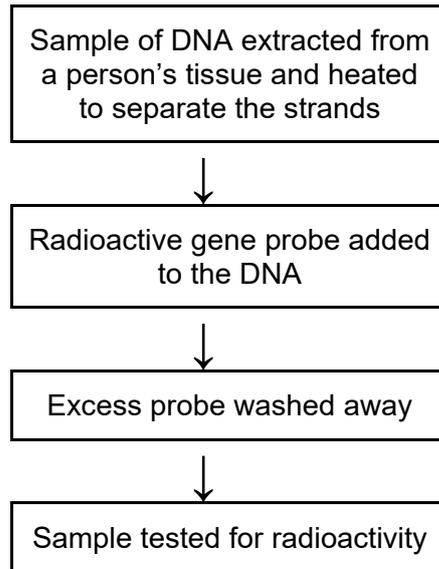
- (a) An antigen in a vaccine leads to the production of antibodies. Describe the part played by B lymphocytes in this process.

(4)

- (b) Hepatitis B vaccine contains a viral antigen produced by genetically modified bacteria. Describe how the isolated gene that codes for a protein in the virus's coat could be transferred to the bacterial cells.

Q9.

- (a) Cystic fibrosis can be caused by any one of several mutant alleles of the cystic fibrosis gene. The most common of these mutant alleles accounts for about 70% of cases of cystic fibrosis. The use of gene probes can identify individuals carrying this allele. Gene probes are single strands of DNA which are radioactively labelled. They have a base sequence that is complementary to a mutant allele. The main stages in using a gene probe are shown in the diagram.



Using the information given, explain how the use of a gene probe could enable the presence of a mutant allele of the cystic fibrosis gene to be detected.

(4)

- (b) Sheep have been genetically engineered to produce alpha-1-antitrypsin which is used to treat cystic fibrosis. Use your knowledge of this process to explain **one** argument for and **one** against using sheep in this way.

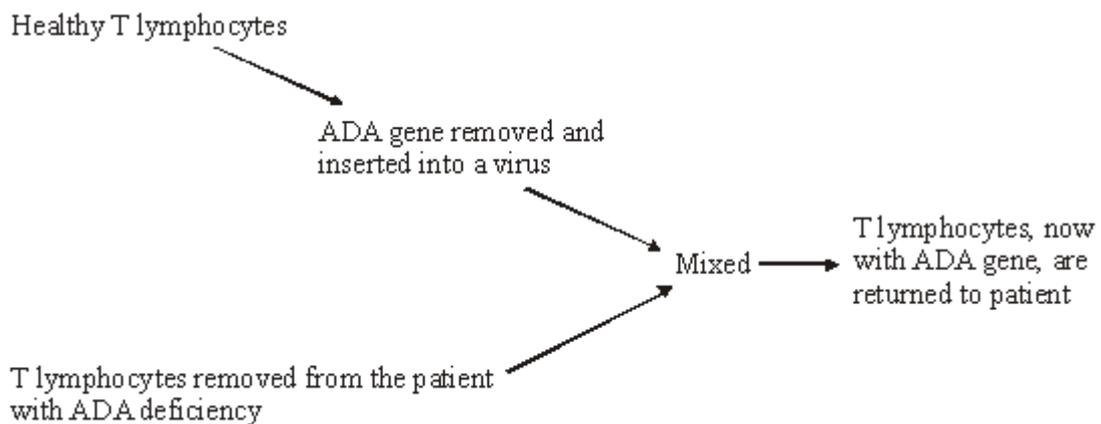
For

Against

(2)
(Total 6 marks)

Q10.

Gene therapy is used to treat the genetic disorder, ADA deficiency. Affected individuals are unable to produce the enzyme adenosine deaminase (ADA). Without this enzyme, T lymphocytes, a type of white blood cell, cannot provide immunity to infection. The diagram shows the processes involved in the treatment of ADA deficiency by gene therapy.



(a) What is meant by *gene therapy*?

(1)

(b) The ADA gene is inserted into a virus. Give **two** advantages of using a virus in gene therapy.

1. _____

2. _____

(2)

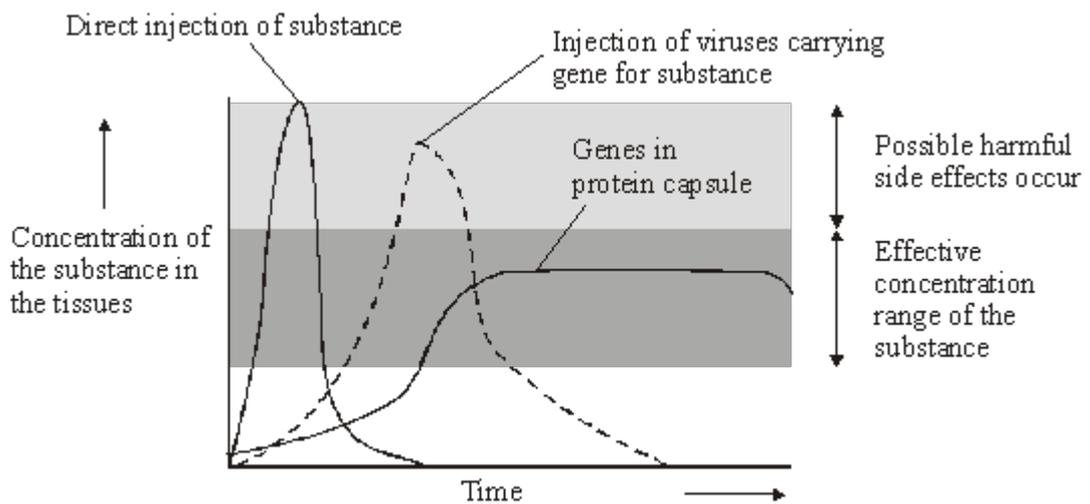
(c) Individuals who have been treated by this method of gene therapy do not pass on the ADA gene to their children. Explain why.

(6)

(b) In gene therapy, genes are introduced into a person who has defective genes which do not produce an important substance. Three experiments were done to compare techniques for introducing an important substance into a person with defective genes.

1. The substance was injected directly.
2. Harmless viruses carrying genes coding for the substance were injected.
3. The genes were put into a protein capsule which was inserted into the tissues.

The graph shows results of the experiments.



Takahiro Ochiya et al, *Biomaterials for Gene Delivery: Studies on Metastasis*, (National Cancer Centre, Research Institute, Tokyo, Japan) 1999

(i) Describe the results of the three experiments.

(3)

(ii) Using the information in the graph, suggest **one** advantage and **one** disadvantage of the capsule method compared to the others.

Advantage _____

Disadvantage _____

(2)

(Total 11 marks)

Q12.

- (a) Describe how a gene can be isolated from human DNA.

(2)

- (b) Describe how an isolated gene can be replicated by the polymerase chain reaction (PCR).

(4)

- (c) (i) Describe how a harmless virus, genetically engineered to contain a CFTR gene, can be used to insert the gene into a cystic fibrosis sufferer.

(2)

- (ii) A virus used in gene therapy has RNA as its genetic material and has an enzyme called reverse transcriptase. Inside a human cell, reverse transcriptase uses viral RNA to make viral DNA.

Explain why the enzyme is called *reverse transcriptase*.

(1)

(Total 9 marks)

Q13.

Scientists manufactured large quantities of human insulin using genetic engineering. They started by isolating mRNA from pancreas cells. From this they produced DNA which coded for insulin.

- (a) (i) Suggest **two** reasons why it was better to start with mRNA from pancreas cells rather than with the DNA from these cells.

1. _____

2. _____

(2)

- (ii) The scientists used two enzymes, **Enzyme 1** and **Enzyme 2**, to produce DNA from mRNA.

The reactions catalysed by these enzymes are shown below.

mRNA $\xrightarrow{\text{Enzyme 1}}$ single-stranded DNA

single-stranded DNA $\xrightarrow{\text{Enzyme 2}}$ double-stranded DNA

Name enzymes 1 and 2.

Enzyme 1 _____

Enzyme 2 _____

(2)

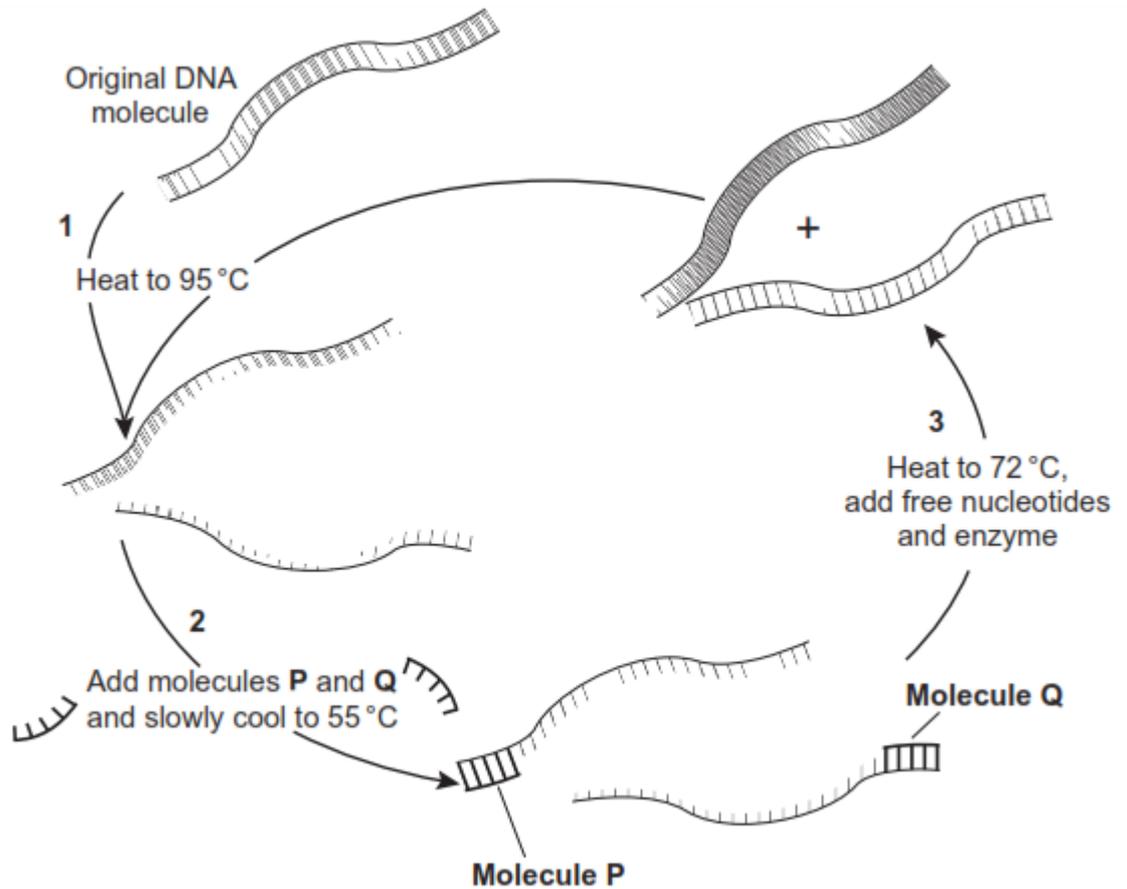
- (iii) In a double-stranded DNA molecule, the two strands are held together by weak bonds.

Name this type of bond.

(1)

- (b) The scientists used the polymerase chain reaction (PCR) to make copies of the DNA.

The diagram shows the stages of the PCR.



(i) **P** and **Q** are short lengths of single-stranded DNA.

What name is given to molecules such as **P** and **Q**?

(1)

(ii) The mixture is cooled from 95°C to 55°C at step 2.

Explain why.

(1)

(iii) Explain the function of molecules **P** and **Q**.

(Extra space) _____

(2)

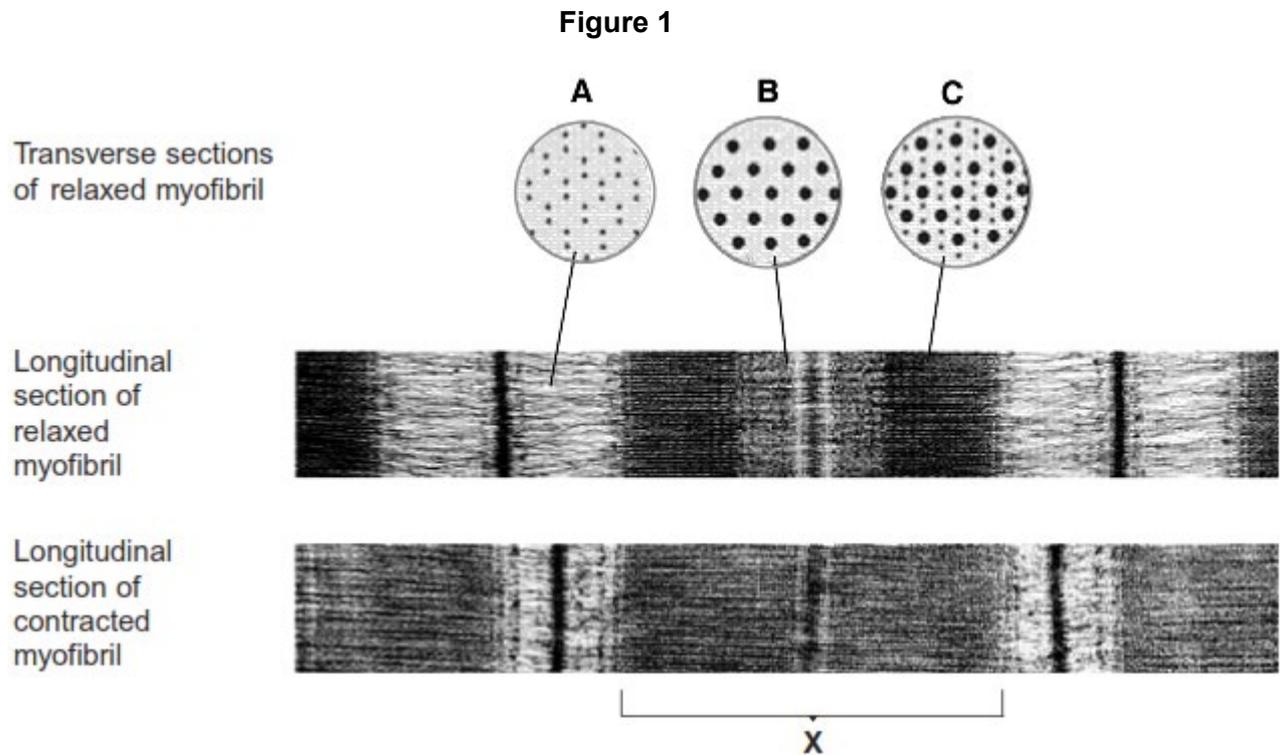
- (iv) How many copies of each original DNA molecule would be present after 5 cycles of PCR?

(1)

(Total 10 marks)

Q14.

Figure 1 shows sections through relaxed and contracted myofibrils of a skeletal muscle. The transverse sections are diagrams. The longitudinal sections are electron micrographs.



- (a) (i) The electron micrographs are magnified 40 000 times. Calculate the length of band **X** in micrometres. Show your working.

Length of band **X** = _____ μm

(2)

- (ii) Explain the difference in appearance between transverse sections **A** and **C** in **Figure 1**.

(1)

- (b) Explain what leads to the differences in appearance between the relaxed myofibril and the contracted myofibril.

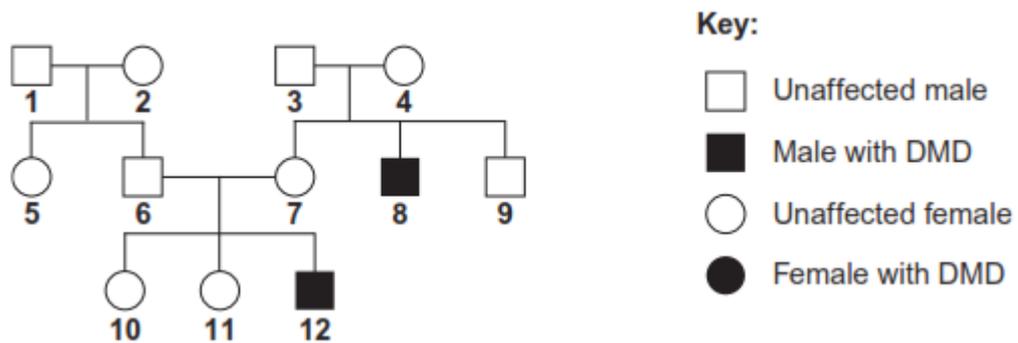
(Extra space)

(4)

- (c) Duchenne muscular dystrophy (DMD) is a condition caused by the recessive allele of a sex-linked gene. A couple have a son with DMD. They want to know the probability that they could produce another child with DMD. They consulted a genetic counsellor who produced a diagram showing the inheritance of DMD in this family.

This is shown in **Figure 2**.

Figure 2



The couple who sought genetic counselling are persons **6** and **7**.

- (i) Give the evidence to show that DMD is caused by a recessive allele.

(1)

- (ii) Give the numbers of **two** people in **Figure 2** who are definitely carriers of muscular dystrophy.

(1)

- (iii) Complete the genetic diagram to find the probability that the next child of couple **6** and **7** will be a son with muscular dystrophy. Use the following symbols:

X^D = normal X chromosome
 X^d = X chromosome carrying the allele for muscular dystrophy
 Y = normal Y chromosome

	6	7
<i>Parental phenotypes</i>	Unaffected	Unaffected
<i>Parental genotypes</i>	_____	_____
<i>Gametes</i>	_____	_____
<i>Offspring genotypes</i>	_____	
<i>Offspring phenotypes</i>	_____	
<i>Probability of having a son with DMD</i>	_____	

(4)

- (d) DMD is caused by a deletion mutation in the gene for a muscle protein called dystrophin. A deletion is where part of the DNA sequence of a gene is lost. People in different families may inherit mutations in different regions of this gene.

Scientists isolated the dystrophin gene from DNA samples taken from children **10**, **11** and **12**. They cut the gene into fragments using an enzyme. The scientists then used two DNA probes to identify the presence or absence of two of these fragments, called **F** and **G**. This allowed them to find the number of copies of each fragment in the DNA of a single cell from each child.

The table shows their results.

Child	Number of copies of gene fragment per cell	
	F	G

10 (unaffected girl)	2	1
11 (unaffected girl)	2	2
12 (boy with DMD)	1	0

- (i) The number of copies of gene fragments **F** and **G** shows that person **12** has DMD.
Explain how.

(1)

- (ii) The number of copies of gene fragments **F** and **G** shows that person **12** is male.
Explain how.

(2)

- (iii) The genetic counsellor examined the scientists' results. He concluded that person **10** is a carrier of DMD but her sister, **11**, is not.

Describe and explain the evidence for this in the table.

(Extra space) _____

(3)

- (e) Person **12** took part in a trial of a new technique to help people with DMD.
Doctors took muscle cells from person **12**'s father and grew them in tissue culture.
They suspended samples of the cultured cells in salt solution and injected them into

a muscle in person **12**'s left leg. They injected an equal volume of salt solution into the corresponding muscle in his right leg. Person **12** was given drugs to suppress his immune system throughout the trial.

Four weeks later, the doctors removed a muscle sample from near the injection site in each leg. They treated these samples with fluorescent antibodies. These antibodies were specific for the polypeptide coded for by gene fragment **G** of the dystrophin gene.

The results are shown in the table.

Location and treatment	Percentage of muscle fibres labelled with antibody
Left leg - injected with cultured cells suspended in salt solution	6.8
Right leg - injected with salt solution	0.0

- (i) Why was it necessary to treat person **12** with drugs to suppress his immune system?

(1)

- (ii) Explain why salt solution was injected into one leg and cultured cells suspended in salt solution into the other.

(1)

- (iii) This technique is at an early stage in its development. The doctors suggested that further investigations need to be carried out to assess its usefulness for treating people with DMD.

Explain why they made this suggestion.

(Extra space) _____

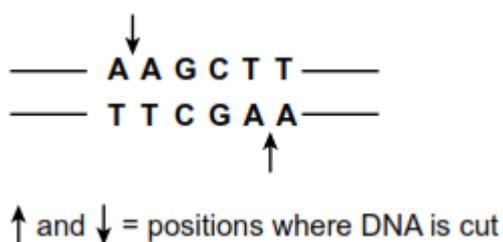
(4)

(Total 25 marks)

Q15.

*Hind*III is an enzyme that cuts DNA into smaller fragments.
The enzyme cuts DNA at the specific base sequence shown in **Figure 1**.

Figure 1



- (a) What general name is given to enzymes such as *Hind*III?

(1)

- (b) *Hind*III produces DNA fragments with sticky ends.

- (i) Use information from **Figure 1** to give the base sequence of one of these sticky ends.

(1)

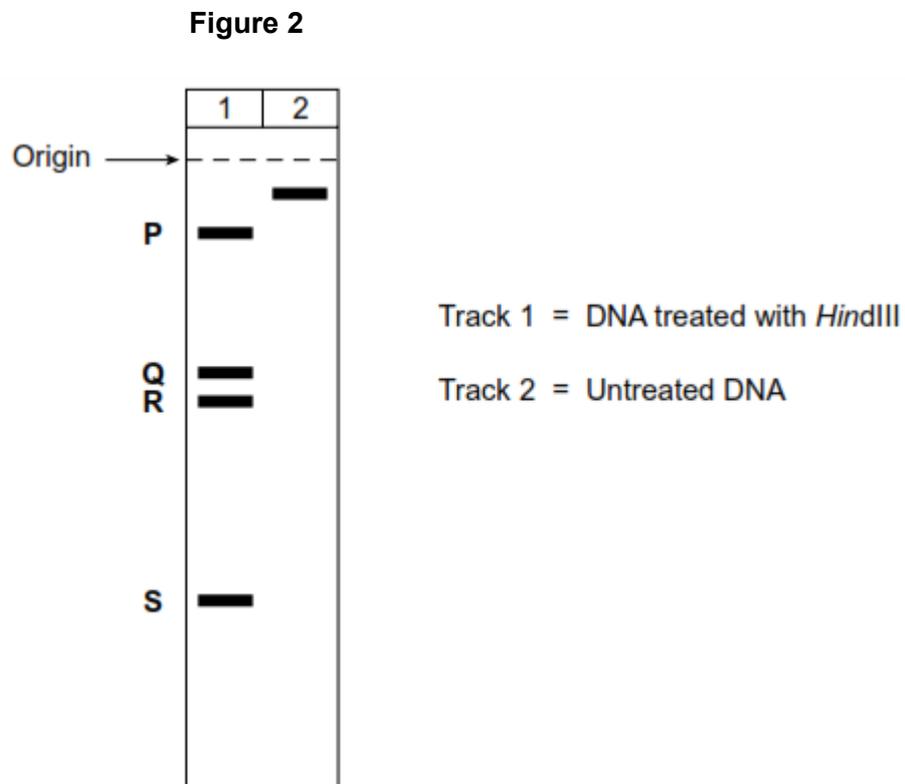
(ii) Sticky ends are useful in genetic engineering. Explain how.

(2)

(c) Scientists prepared a sample containing many identical molecules of DNA. The DNA molecules were linear (non-circular).

They divided the sample into two portions. They treated one portion with *Hind*III but did not treat the other portion. They then carried out gel electrophoresis on each portion.

The results are shown in **Figure 2**.



(i) The lengths of the fragments produced from the DNA treated with *Hind*III were 287, 1232, 1550 and 4943 base pairs. How many base pairs are there in fragment **P**?

P = _____ base pairs

(1)

(ii) How many times did the base sequence, **AAGCTT** occur in the DNA?
TTCGAA

(1)

- (iii) In a certain genetic condition, **one** of these **AAGCTT** sequences is changed.
TTCGAA

Predict what effect this would have on the appearance of the gel in Track 1 of **Figure 2**.

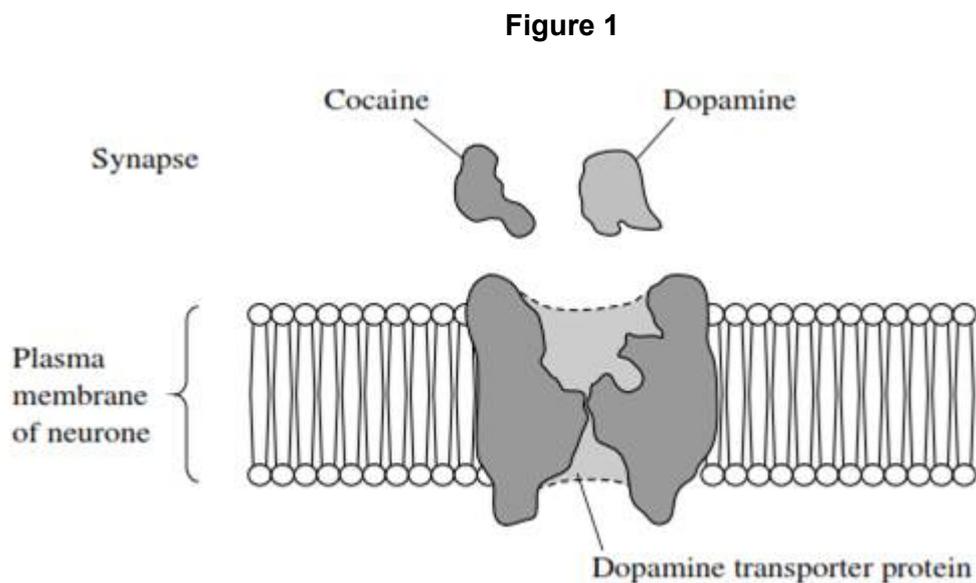
(2)
(Total 8 marks)

Q16.

Cocaine is a highly addictive and illegal drug.

The release of the neurotransmitter dopamine in specific synapses in the brain leads to feelings of pleasure. Dopamine is removed from synapses by dopamine transporter proteins in the plasma membrane of neurones. Cocaine binds to the dopamine transporter protein.

Figure 1 shows a dopamine transporter protein and molecules of cocaine and dopamine.



- (a) Using all of the information, suggest how cocaine leads to feelings of pleasure.

(Extra space)

(3)

- (b) (i) Scientists isolated a mutated gene for the dopamine transporter protein.

Name **one** method that the scientists could have used to produce many copies of the mutated gene in the laboratory.

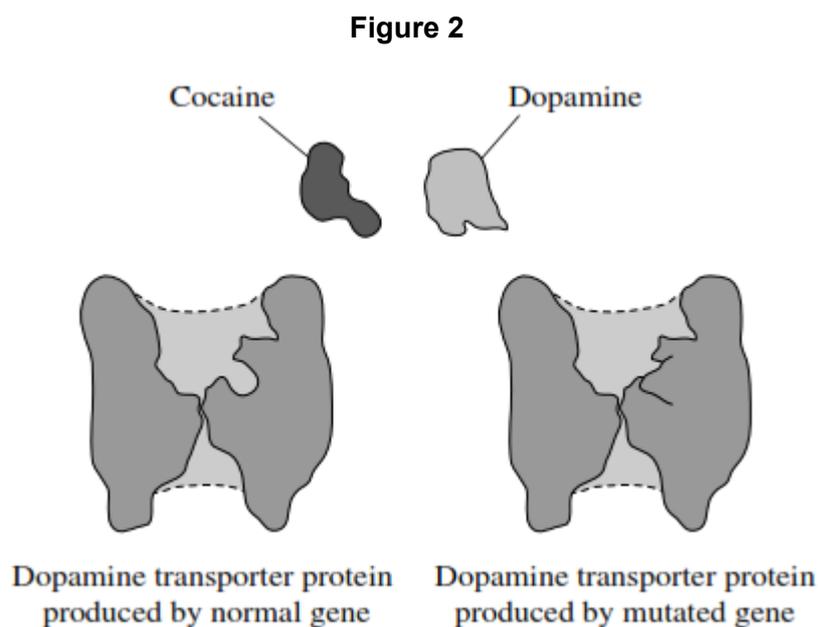
(1)

- (ii) Copies of the gene were then inserted into early embryos of mice. When these mice were born, samples of their DNA were tested using DNA probes to make sure that the mutated gene was present in the mice.

What is a DNA probe?

(2)

- (c) **Figure 2** shows dopamine transporter proteins produced from the normal gene and from the mutated gene.



Explain how the mutation leads to the production of a protein that transports dopamine but is **not** affected by cocaine.

(Extra space) _____

(3)
(Total 9 marks)