RIVER VALLEY HIGH SCHOOL
YEAR 6
PRELIMINARY EXAMINATION II

CANDIDATE NAME									
CENTRE NUMBER	S					INDEX NUMBER				

H2 BIOLOGY **9648/03**

Paper 3 Application Paper **20 Sep 2016**

2 hours

Additional Materials: Answer Paper

READ THESE INSTRUCTIONS FIRST

Write your index number and name on all the work you hand in.
Write in dark blue or black pen.
You may use a HB pencil for any diagrams or graphs.
Do not use staples, paper clips, glue or correction fluid.
DO **NOT** WRITE IN ANY BARCODES.

Section A
Answer **all** questions in the spaces provided on the question paper.

Section B
Answer **all** question on the answer paper provided.

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	**/ 13**
2	**/ 13**
3	**/ 14**
4	**/ 12**
Section B	
5	**/ 20**
Total	**/ 72**

This Question Paper consists of **16** printed pages.

Section A (80 marks)

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

1 The coat colour of Norwegian cattle is mainly determined by the distribution of two pigments: red and black. Both pigments are produced by the action of the enzyme tyrosinase, in cells called melanocytes. A low level of activity of the enzyme leads to the production of red pigment, while a high activity of the enzyme produces black pigment.

The activity of the enzyme is increased by the melanocyte stimulating hormone (MSH), which binds to the MSH receptor. The receptor is encoded by gene **E**, which has three alleles, $\mathbf{E^D}$, $\mathbf{E^A}$ and **e**. $\mathbf{E^D}$ and $\mathbf{E^A}$ each encodes a receptor with different activity. No receptor is produced by the recessive allele, **e**.

Alleles $\mathbf{E^D}$ and $\mathbf{E^A}$ differs by a single base substitution; while alleles **e** and $\mathbf{E^A}$ differs by a single base deletion.

(a) Explain how a mutation at $\mathbf{E^A}$ allele may result in the protein encorded by **e** allele. [3]

DNA was extracted from the frozen semen of six bulls with different genotypes at the E locus. The DNA from each animal was separately digested with two different restriction enzymes, **P** and **Q**.

The products of each digestion were separated on a gel and the banding pattern shown in **Fig. 1.1**.

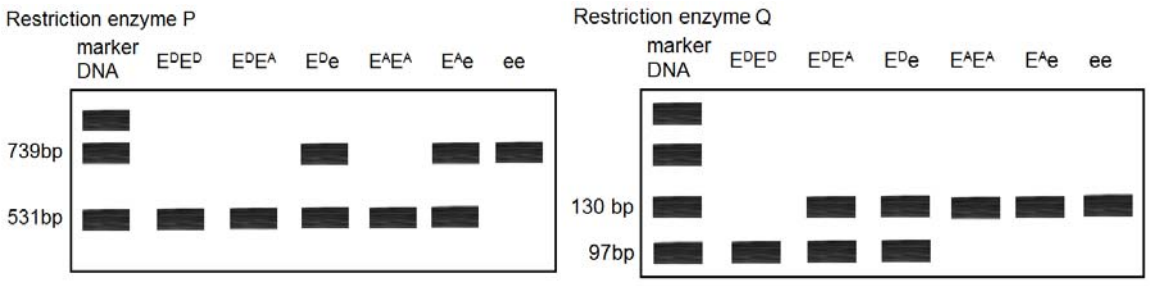

Fig. 1.1

(b) **(i)** Explain why the banding patterns from the same genotype are different when a different restriction enzyme is used. [2]

(ii) Explain the role of marker DNA. [2]

(iii) State which genotype(s) can be identified by using each of the two restriction enzymes. [2]

P:

Q:

Bovine somatotropin (BST) is an animal hormone used to increase milk production in dairy cows. BST is naturally produced in the cow pituitary gland in small quantity and is used to regulate metabolic processes. With the advent of biotechnology, *BST* gene can be cloned in *Escherichia coli*. The bacteria are grown in bioreactors to produce BST, which is purified to produce hormone for injection.

Fig. 1.2 shows the plasmid map of the BGH1 plasmid, used for transformation of *BST* gene into *E.coli*. Sal 1 restriction enzyme was used to cut the BGH1 plasmid for insertion of *BST* gene.

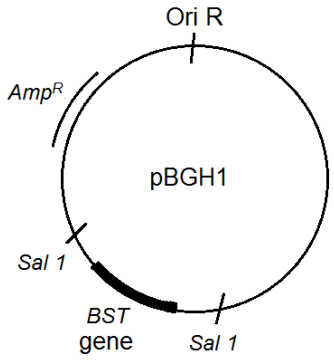

Fig. 1.2

(c) **(i)** Describe the natural role of Sal 1 restriction enzyme. [2]

(ii) Explain how Amp^R gene facilitates *BST* gene cloning. [2]

[Total: 13]

2 Omega-3 fatty acids are polyunsaturated fats that are often marketed as health supplements to prevent cholesterol deposits in arterial walls.

In the wild, omega-3 fatty acids are produced by marine algae and accumulate in small fishes that feed on them. These small fishes are then fed to farmed fishes to increase their omega-3 fatty acid content. Farmed fishes high in omega-3 fatty acids serve as a good source for omega-3 fatty acid extraction for the health supplement industry.

A new method to produce omega-3 fatty acids involves insertion of seven genes of the marine algae into oilseed plant, *Camelina sativa.* The resulting seed pods contain as much as 200 milligrams of omega-3 fatty acids in a single tablespoon of 'fish oil' extracted. Currently, this 'fish oil' is fed to farmed fishes, in replacement of small fishes.

The effect of human consumption of 'fish oil' from genetically modified *C. sativa* seed pods is still under investigation.

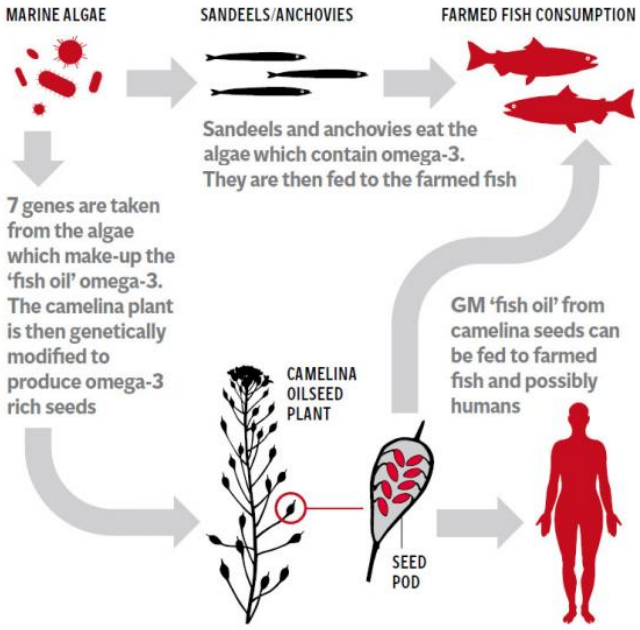

Fig. 2.1

Source: http://www.independent.co.uk/news/science/new-gm-cereal-crop-produces-fish-oil-in-its-seeds-10372772.html

(a) A Ti plasmid containing the seven genes from marine algae is constructed and introduced into *A. tumefaciens* before infecting *C. sativa* plant cells. Describe the properties of Ti plasmids that make them suitable as vectors in this process. [2]

(b) Suggest why crop plants like *C. sativa* are better candidates for genetic modification than fishes. [2]

(c) **(i)** With reference to **Fig 2.1**, suggest two benefits of using genetically modified *C. sativa* as an omega-3 fatty acid source for farmed fishes. [2]

(ii) Describe an ethical and a social implication associated with genetically modified *C. sativa* 'fish oil'. [2]

The effectiveness of genetically modified *C. sativa* seed pods in preventing cholesterol deposition in arteries was investigated using rodents, to evaluate if they can be used to substitute fish oil from farmed fishes. Effectiveness is evaluated by analysing the levels of total omega-3 fatty acids available in blood and the diameter of rodents' arteries.

Rodents were divided into three groups of 10 and were subjected to respective treatment for a period of six months as shown below.

Group A: High fat diet enriched with 'fish oil' from genetically modified *C. sativa* seed pods

Group B: High fat diet enriched with fish oil from farmed fishes fed with small fishes

Control: High fat diet without fish oil

The results are summarised in the **Table 2.1**.

Table 2.1

	Group A	**Group B**	**Control**
Total omega-3 fatty acids level/ $mmolL^{-1}$	33.7 ± 0.3	27.1 ± 0.1	20.0 ± 0.3
Cross section of artery after 6 months			

(d) With reference to **Table 2.1**, comment on whether genetically modified *C. sativa* is a more effective substitute. [3]

(e) Describe how genetic engineering is used to increase quality of another named crop plant. [2]

[Total: 13]

3 Severe combined immunodeficiency disease (SCID) is caused by a severe genetic defect often found in newborns. The condition must be diagnosed and treated quickly to prevent serious complications. However, doctors continue to struggle with often ineffective treatment options.

In recent studies, researchers found that blood stem cells may effectively treat SCID caused by a deficiency in the adenosine deaminase (ADA) gene. The ADA gene is critical for the proper functioning of the immune system.

(a) Explain how an individual with SCID caused by ADA deficiency inherited this condition. [2]

(b) **(i)** Describe the normal functions of adult stem cells obtained from the bone marrow and stem cells obtained from the zygote. [2]

(ii) Explain how the properties of blood stem cells allow for them to effectively treat SCID. [3]

A trial was carried out on an ADA-SCID patient of age 7 months. Blood stem cells were collected from the patient's bone marrow, transduced with a retroviral vector containing the therapeutic ADA allele, and injected back to the patient. The patient received 8.8 x 10^6 blood stem cells per kg body weight, containing 25% successfully transduced cells in culture.

Fig. 3.1 shows the total T cell count in the blood sample of this patient.

These cells were then isolated and analysed for presence of ADA mRNA in the cytoplasm. **Fig. 3.2** shows the percentage of cells carrying ADA mRNA.

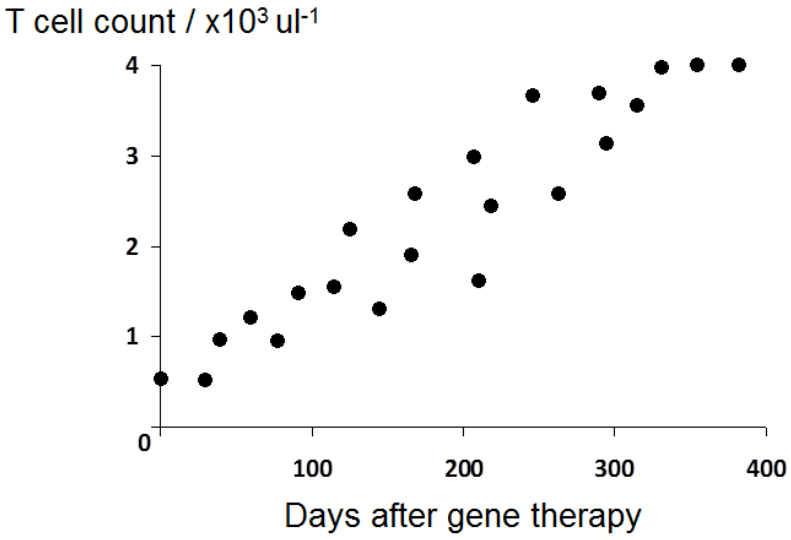

Fig. 3.1

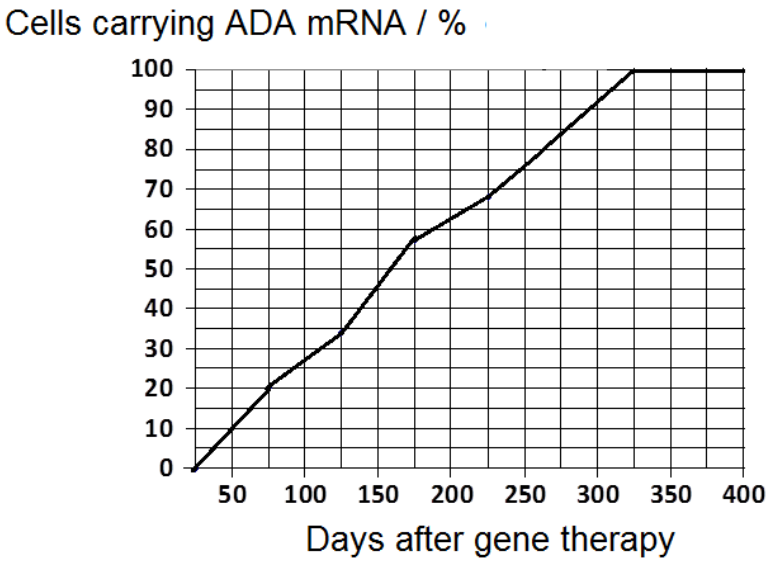

Fig. 3.2

(c) **(i)** Suggest a medical advantage of using patient's own blood stem cells rather than stem cells from a healthy donor. [1]

(ii) Explain why a retroviral vector was chosen for this trial. [2]

(d) With reference to **Fig. 3.1** and **3.2**, account for the success of this gene therapy in treatment of SCID, 350 days after the therapy. [4]

[Total: 14]

4 Planning question

The use of β-galactosidase for the hydrolysis of lactose in milk is a promising enzyme application in the food processing industry. A large fraction of the human population is lactose intolerant, due to the low levels of β-galactosidase present in the intestine. This causes difficulty in digesting milk products. Therefore lactose hydrolysis, which lowers lactose concentration in milk, allows the lactose intolerant population to consume milk.

Milk products containing lactose may have different pH. In order for β-galactosidase to work, its optimal pH for catalytic activity must coincide with the pH of the milk product. Hence, β-galactosidase is extracted from different sources due to their difference in optimal pH. Sources of β-galactosidase include mould species such as *Aspergillus niger*, and yeast species such as *Saccharomyces cerevisiae*.

ONPG, can be used to study the catalytic function of β-galactosidase as shown in the following reaction:

ONPG is a colourless solution while ONP produced from the reaction is a yellow solution. The reaction can be stopped by adding sodium carbonate solution in 1:1 ratio. By comparing the colour intensity of the resulting product to a colour standard, the concentration of ONP at the end of the reaction can be determined.

Using this information and your own knowledge, design an experiment to determine the respective optimal pH at which β-galactosidase of *Aspergillus niger* and *Saccharomyces cerevisiae* work.

You must use:

- 10% ONPG solution
- *A. niger* β-galactosidase
- *S. cerevisiae* β-galactosidase
- pH buffers for different pH
- 10% ONP solution
- 10% sodium carbonate solution
- Distilled water
- Stopwatch

You may select from the following apparatus:

- Normal laboratory glassware e.g. test-tubes, beakers, measuring cylinders, graduated pipette, glass rods, etc.
- Droppers
- Eye protection
- Gloves

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 diagrams, if necessary,
- identify the independent and dependent variables,
- describe the method with scientific reasoning used to decide the method so that the results are as accurate and reliable as possible
- show how you will record your results and the proposed layout of results tables and graphs,
- use the correct technical and scientific terms,
- include reference to safety measures to minimize any risks associated with the proposed experiment.

[Total: 12]

Section B (20 marks)

Answer **all** question.

Write your answers on the separate answer paper provided.
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.

A **NIL** return is necessary if you have not attempted this section.

5. **(a)** Explain how restriction fragment length polymorphism (RFLP) analysis facilitates the construction of a genomic linkage map. [6]

(b) Plant tissue culture techniques allows for aseptic growth of excised plant parts *in-vitro*. Describe how this is carried out. [9]

(c) *"The Human Genome Project was one of the great feats of exploration in history… (giving) us the ability, for the first time, to read nature's genomic blueprint for building a human being." – National Human Genome Research Institute.* [5]

Discuss the main objectives of this project.

[Total: 20]