Cambridge International A Level

Cambridge International Examinations
Cambridge International Advanced Level

CANDIDATE NAME

CENTRE NUMBER

CANDIDATE NUMBER

1277471123

BIOLOGY **9700/51**

Paper 5 Planning, Analysis and Evaluation **May/June 2014**

1 hour 15 minutes

Candidates answer on the Question Paper.

No Additional Materials are required.

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Write in dark blue or black ink.
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Do not use staples, paper clips, highlighters, glue or correction fluid.
DO **NOT** WRITE IN ANY BARCODES.

Answer **all** questions.

Electronic calculators may be used.
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.

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1 **(a)** In an investigation to study genetic variation, DNA was obtained from four varieties of the same invertebrate species.

The following technique was used:

- DNA was digested using a number of different restriction enzymes to obtain fragments of between 200 – 700 base pairs in length, each of which have sticky ends.
- Known RNA sequences (RNA probes) were used to select DNA fragments with specific sticky ends and to separate them from the rest of the DNA.
- Multiple copies of the DNA fragments that hybridised with the DNA were made (DNA amplification).
- The fragments were separated by gel electrophoresis to give a genetic fingerprint called an amplified fragment length polymorphism (AFLP).

(i) Explain how RNA probes, used in this technique, select fragments of DNA.

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(ii) This technique is able to separate small DNA fragments that may differ by only one nucleotide. Suggest why this technique is used to study genetic variation within species.

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(b) Describe the main stages used in gel electrophoresis to separate and locate the amplified DNA fragments.

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.....[5]

(c) Three groups of students used gel electrophoresis to separate a set of DNA fragments from each of the four varieties.

State two variables that the students should standardise to ensure that their results from gel electrophoresis can be compared.

1. .....

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2. .....

.....[2]

(d) Three different groups of RNA probes, **A**, **B** and **C**, were used to select different DNA fragments from all four varieties of the invertebrate species, 1, 2, 3 and 4.
Gel electrophoresis was then used to separate the fragments.
Fig. 1.1 shows the results.

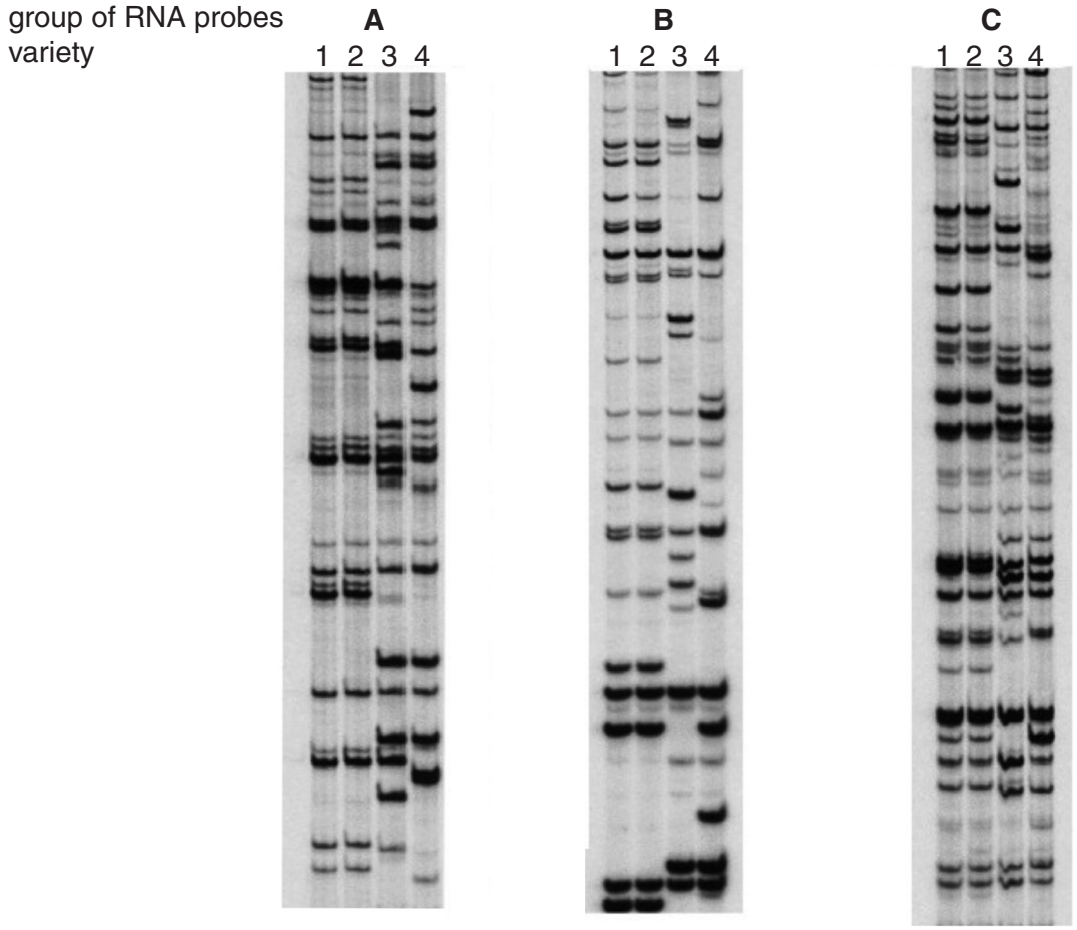


Fig. 1.1

(i) Fig. 1.1 shows that some DNA fragments are present in the genetic fingerprints of all four varieties, 1, 2, 3 and 4, when the different groups of RNA probes, **A**, **B** and **C**, are used.

On Fig. 1.1:
- draw an arrow (⟶) to indicate **one** DNA fragment found in all four varieties when RNA probes in group **A** were used.
- draw a second arrow for the RNA probes used in group **B**
- draw a third arrow for the RNA probes used in group **C**. [1]

(ii) State the varieties that appear to have the same genetic fingerprint. Give the evidence for your answer.

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.....[2]

[Total: 14]

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2 **(a)** Green manuring is a method of fertilising soil. One type of plant is grown for a certain period of time and then ploughed into the soil while the plants are still green.

A field investigation into the effect of green manuring on the yield of *Sorghum bicolor* was carried out using a legume. Different parts of the legume were used as green manure.

The *Sorghum* was grown in four types of trial plot as follows:

- no treatment (control)
- legume roots only ploughed into the soil
- legume shoots only ploughed into the soil
- both legume roots and shoots ploughed into the soil.

Fig. 2.1 shows the arrangement of trial plots used for the investigation.
A random number generator was used to locate each of the trial plots.

trial plots, 5 m × 5 m, separated by fences

control	shoots and roots	roots only	control	roots only	roots only	shoots and roots	shoots only
shoots and roots	roots only	shoots only	shoots only	control	shoots and roots	shoots only	control

Fig. 2.1

The trial plots were left for one month before the *Sorghum* grain was sown.

Fig. 2.2 shows how the grain was sown in each trial plot.

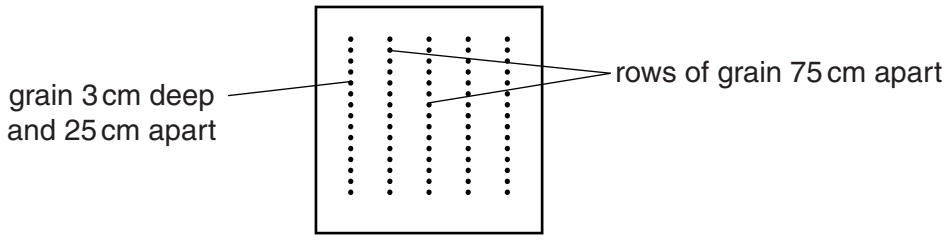


Fig. 2.2

(i) Identify two variables that have been standardised in this field investigation.

1. .....

2. .....[1]

(ii) Suggest two **abiotic** variables that **cannot** be standardised in this field investigation.

1. .....

2. .....[2]

(iii) Suggest why the plots used to grow the *Sorghum* were left for one month before the sowing of the grain.

.....

.....[1]

(iv) State how this experimental design helps to ensure the reliability of the results.

.....

.....[1]

(b) *Sorghum* from each of the trial plots was collected and the dry mass of the different parts of the plants determined. The mean dry mass of shoot, roots and grain was calculated.

Table 2.1 shows the results of this investigation.

Table 2.1

treatment	mean dry mass of *Sorghum* / kg per hectare			
	shoots	roots	grain	whole plant
no treatment (control)	3831	2486	398	6715
roots only	4773	2744	526	8043
shoots only	5645	3252	782	9679
roots and shoots	5923	3707	975	10605

(i) Calculate the percentage increase in dry mass of whole *Sorghum* plants as a result of fertilising the soil with legume shoots only. Show your working.

.....[2]

(ii) The increase in dry mass of grain caused by using legume roots as green manure is 128 kg per hectare.
Calculate the ratio of the increase in the mean dry mass of grain caused by using legume shoots in comparison to using legume roots only as green manure. Show your working.

.....[2]

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(c) Table 2.2 shows the results of *t*-tests that were carried out to compare the effect of the different types of green manure on the dry mass of *Sorghum.*

In each test the plants with added manure were compared with the control.

Table 2.2

treatment	P < 0.05			
	shoots	roots	grain	whole plant
legume roots only	not significant	not significant	not significant	not significant
legume shoots only	not significant	not significant	significant	significant
legume roots and shoots	significant	significant	significant	significant

(i) Suggest a null hypothesis for the statistical tests.

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.....[1]

(ii) State a reason why *t*-tests were used to find out if the effect of the different types of green manure on the dry mass of *Sorghum* was significant.

.....

.....[1]

(iii) Explain what is meant by:

statistically significant

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P < 0.05.

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.....[2]

(d) With reference to Table 2.1 and Table 2.2 only, state three conclusions that can be drawn from these results about the effect of using green manure on *Sorghum.*

1. .....

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2. .....

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3. .....

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.....[3]

[Total: 16]

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Cambridge International A Level

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CANDIDATE NAME	
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2847631226

BIOLOGY **9700/52**

Paper 5 Planning, Analysis and Evaluation **May/June 2014**

1 hour 15 minutes

Candidates answer on the Question Paper.

No Additional Materials are required.

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CAMBRIDGE International Examinations

1 **(a)** A group of students was given the task of planning a method to compare the effect of two different restriction enzymes on some DNA extracted from peas.

The students used the following procedure to extract and digest DNA from the peas.

- 3 g of sodium chloride was dissolved in 90 cm^3 of distilled water in a beaker.
- 10 cm^3 of liquid detergent was added to the salt solution and stirred.
- 50 g of peas were ground up using a glass rod and then added to the salty detergent solution.
- The beaker containing the mixture of peas and salty detergent solution was left in a water bath at 60°C for **exactly** 15 minutes.
- The mixture was then cooled in an ice-water bath for 5 minutes, stirring frequently.
- The mixture was filtered into another beaker.
- 10 cm^3 of filtrate was transferred into a boiling tube and 2–3 drops of a protease solution were added. This was left for a few minutes.
- Ice-cold ethanol was then poured down the side of the boiling tube to form a layer on top of the filtrate. This was left undisturbed for a few minutes to form a layer of precipitated DNA.
- The precipitated DNA was separated from the cold ethanol layer by twisting the DNA onto a glass hook.
- The DNA was mixed with 20 cm^3 EDTA buffer solution.
- 5 cm^3 samples of the EDTA buffer solution containing the DNA extract were placed into three separate test-tubes, 1, 2 and 3.
- Two different restriction enzymes, *Eco* RI and *Hin* dIII, were added to the samples as shown in Table 1.1.

Table 1.1

sample	1	2	3
restriction enzymes added	*Eco* RI	*Hin* dIII	*Eco* R1 and *Hin* dIII

- These were left for three hours to complete digestion of the DNA into fragments of different sizes.

Suggest a reason for each of the following steps in the procedure for extracting DNA.

(i) using detergent .....

.....[1]

(ii) leaving at 60°C .....

.....[1]

(iii) filtering the mixture .....

.....[1]

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(iv) adding protease .....

.....[1]

(b) The students used gel electrophoresis to separate the DNA fragments produced by the digestion with the restriction enzymes.

Describe the main stages used in gel electrophoresis that the students could use to separate and locate the DNA fragments.

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(c) **(i)** Identify a dependent variable that the student could measure.

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(ii) State two variables that the students should standardise to ensure that the results from the different samples shown in Table 1.1 can be compared.

1. .....

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2. .....

.....[2]

(d) Fig. 1.1 shows the results of the electrophoresis.

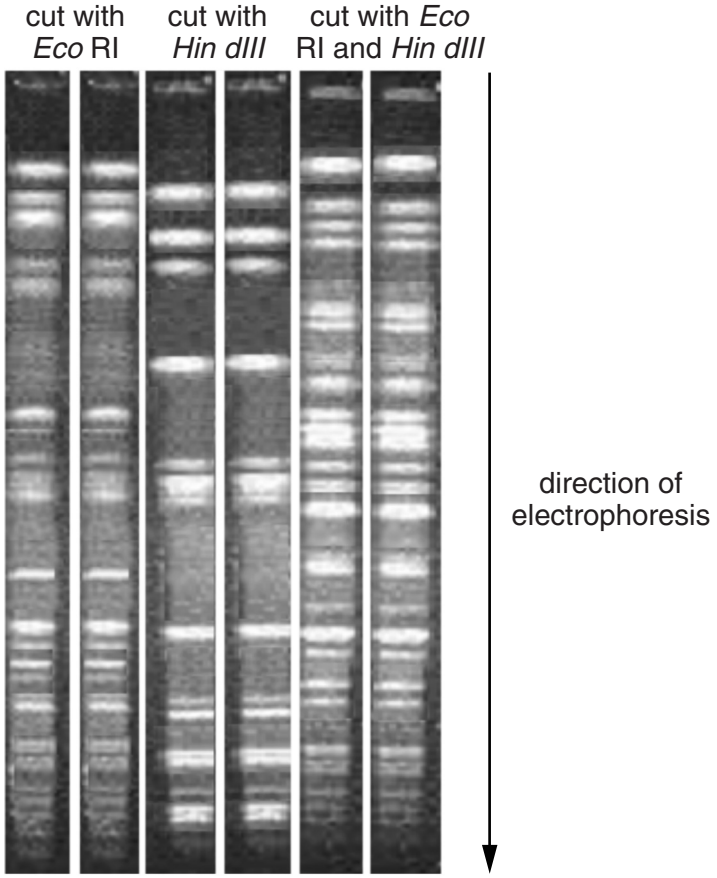


Fig. 1.1

The students concluded that the DNA of peas had more restriction sites for *Eco* RI than for *Hin* dIII and that some of the *Hin* dIII sites were within fragments produced by *Eco* RI.

(i) State **one** piece of evidence in Fig. 1.1 to support each of these conclusions:

there are more restriction sites for *Eco* RI than for *Hin* dIII

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there are some *Eco* RI sites within fragments produced by *Hin* dIII.

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(ii) Suggest **one** reason why the results in Fig. 1.1 may **not** support the student's conclusions.

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(e) Gel electrophoresis is also used in the sequencing of DNA.

A method used for sequencing DNA is described below.

- A DNA molecule is first heated so it separates into two strands.
- DNA polymerase is then used to synthesise DNA using normal nucleotides and modified nucleotides.
- The four different modified nucleotides are each labelled with a different coloured fluorescent dye.
- These are: **red** CTP **green** TTP **blue** GTP **yellow** ATP
- The addition of modified nucleotides to the DNA strand stops the addition of any more nucleotides.

Fig. 1.2 shows the formation of a three nucleotide fragment from normal nucleotides and a modified nucleotide.

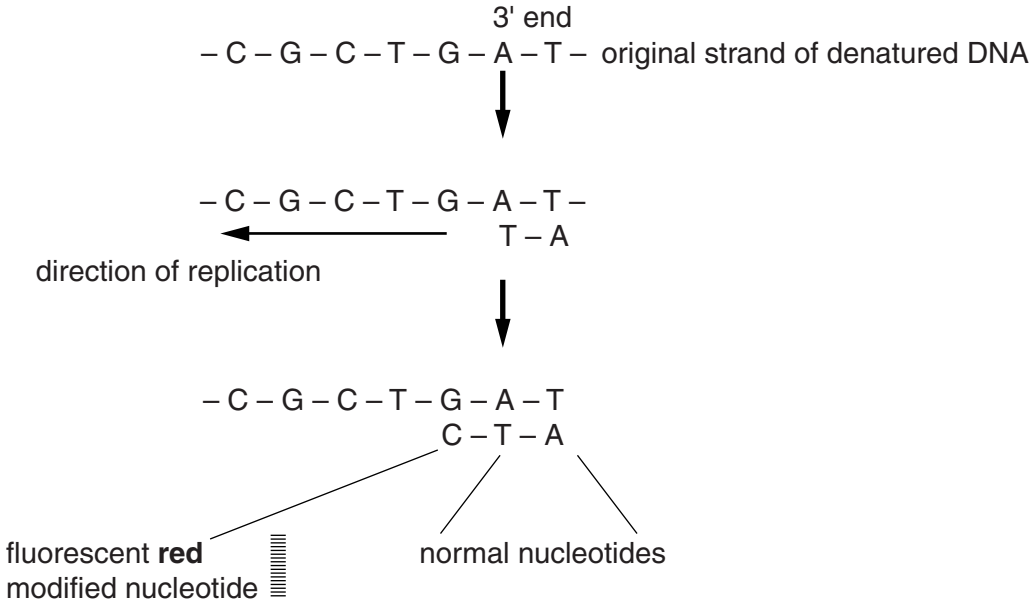


Fig. 1.2

- Fragments of different lengths are produced depending on the position of the modified nucleotides.
- The DNA fragments synthesised are separated by gel electrophoresis and a laser light used to identify the different colours.

Fig. 1.3 shows the results of sequencing part of a DNA molecule.

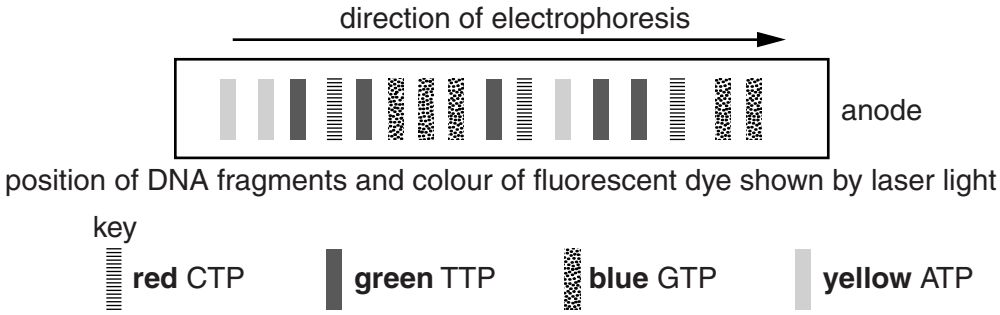


Fig. 1.3

(i) Use the information in Fig. 1.3 to work out the sequence of the DNA template, starting from the 3' end.

.....[1]

(ii) Explain how you worked out this sequence.

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[Total: 19]

2 **(a)** An investigation into the efficiency of nitrogen use by *Sorghum bicolor* was carried out in a glasshouse.

- Four varieties of *Sorghum,* **W**, **X**, **Y** and **Z**, were grown in pots.
- Eight replicates of each variety were watered using a nutrient medium containing all the plant requirements **except** nitrogen. These were the **low nitrogen** plants.
- Another eight replicates were watered with a nutrient medium containing all the required nutrients including nitrogen. These were the **high nitrogen** plants.
- The efficiency of nitrogen use was determined from the activity of three enzymes used in photosynthesis.

Fig. 2.1 shows the reactions in mesophyll cells catalysed by two of these enzymes.

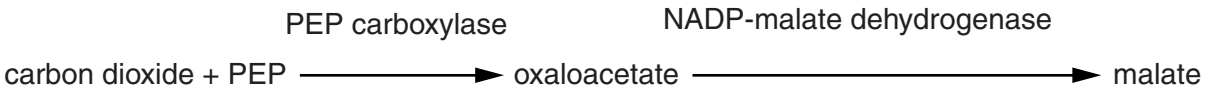


Fig. 2.1

In the bundle sheath cells malate is broken down to release carbon dioxide which is assimilated by the third enzyme, RuBP carboxylase/oxygenase (Rubisco).

(i) Identify the **two** independent variables in this investigation.

1. .....

2. .....[2]

(ii) Describe **one** way in which this experimental design helps to ensure the reliability of the results.

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.....[1]

(b) Table 2.1 shows the results of this investigation.

Table 2.1

	mean enzyme activity of leaf samples ± S_M / nmol min^{-1} mg^{-1} protein					
	PEP carboxylase		NADP-malate dehydrogenase		Rubisco	
variety	high nitrogen	low nitrogen	high nitrogen	low nitrogen	high nitrogen	low nitrogen
W	29.1 ± 8.3	41.1 ± 5.6	2662 ± 331	338 ± 20	13.1 ± 0.4	1.5 ± 0.6
X	17.1 ± 1.6	8.1 ± 0.9	1979 ± 161	2468 ± 256	16.4 ± 2.9	2.0 ± 0.2
Y	44.4 ± 6.0	10.8 ± 2.4	927 ± 126	1936 ± 146	19.8 ± 1.2	3.3 ± 0.3
Z	59.4 ± 2.8	7.5 ± 1.2	1477 ± 89	2547 ± 350	23.0 ± 1.2	2.4 ± 0.1

(i) Calculate the mean percentage decrease in activity of Rubisco between high and low nitrogen conditions for variety **Y**. Give your answer to the **nearest whole number**. Show your working.

.....% [2]

(ii) With reference to Table 2.1 explain what standard error (S_M) shows about the results of this investigation.

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(c) A number of *t*-tests were carried out to find out if the difference in activity of the enzymes was significantly different in the four varieties tested.

The tests carried out were between the following pairs of varieties:

W and **X**	**W** and **Y**	**W** and **Z**
X and **Y**	**X** and **Z**	**Y** and **Z**

(i) With reference to the activity of NADP-malate dehydrogenase in low nitrogen, shown in Table 2.1, identify the *t*-test(s) in which the difference in activity of NADP-malate dehydrogenase is **not** likely to be significant.

Show your answer by underlining the test(s). Give a reason for your answer.

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.....[2]

(ii) State why 14 degrees of freedom was used for each of the *t*-test calculations.

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(d) Using the information in Table 2.1, suggest which one of the varieties appears to be anomalous. Give a reason for your answer.

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[Total: 11]

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BIOLOGY **9700/53**

Paper 5 Planning, Analysis and Evaluation **May/June 2014**

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1 **(a)** In an investigation to study genetic variation, DNA was obtained from four varieties of the same invertebrate species.

The following technique was used:

- DNA was digested using a number of different restriction enzymes to obtain fragments of between 200 – 700 base pairs in length, each of which have sticky ends.
- Known RNA sequences (RNA probes) were used to select DNA fragments with specific sticky ends and to separate them from the rest of the DNA.
- Multiple copies of the DNA fragments that hybridised with the DNA were made (DNA amplification).
- The fragments were separated by gel electrophoresis to give a genetic fingerprint called an amplified fragment length polymorphism (AFLP).

(i) Explain how RNA probes, used in this technique, select fragments of DNA.

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.....[2]

(ii) This technique is able to separate small DNA fragments that may differ by only one nucleotide. Suggest why this technique is used to study genetic variation within species.

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(b) Describe the main stages used in gel electrophoresis to separate and locate the amplified DNA fragments.

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.....[5]

(c) Three groups of students used gel electrophoresis to separate a set of DNA fragments from each of the four varieties.

State two variables that the students should standardise to ensure that their results from gel electrophoresis can be compared.

1. .....

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2. .....

.....[2]

(d) Three different groups of RNA probes, **A**, **B** and **C**, were used to select different DNA fragments from all four varieties of the invertebrate species, 1, 2, 3 and 4.
Gel electrophoresis was then used to separate the fragments.
Fig. 1.1 shows the results.

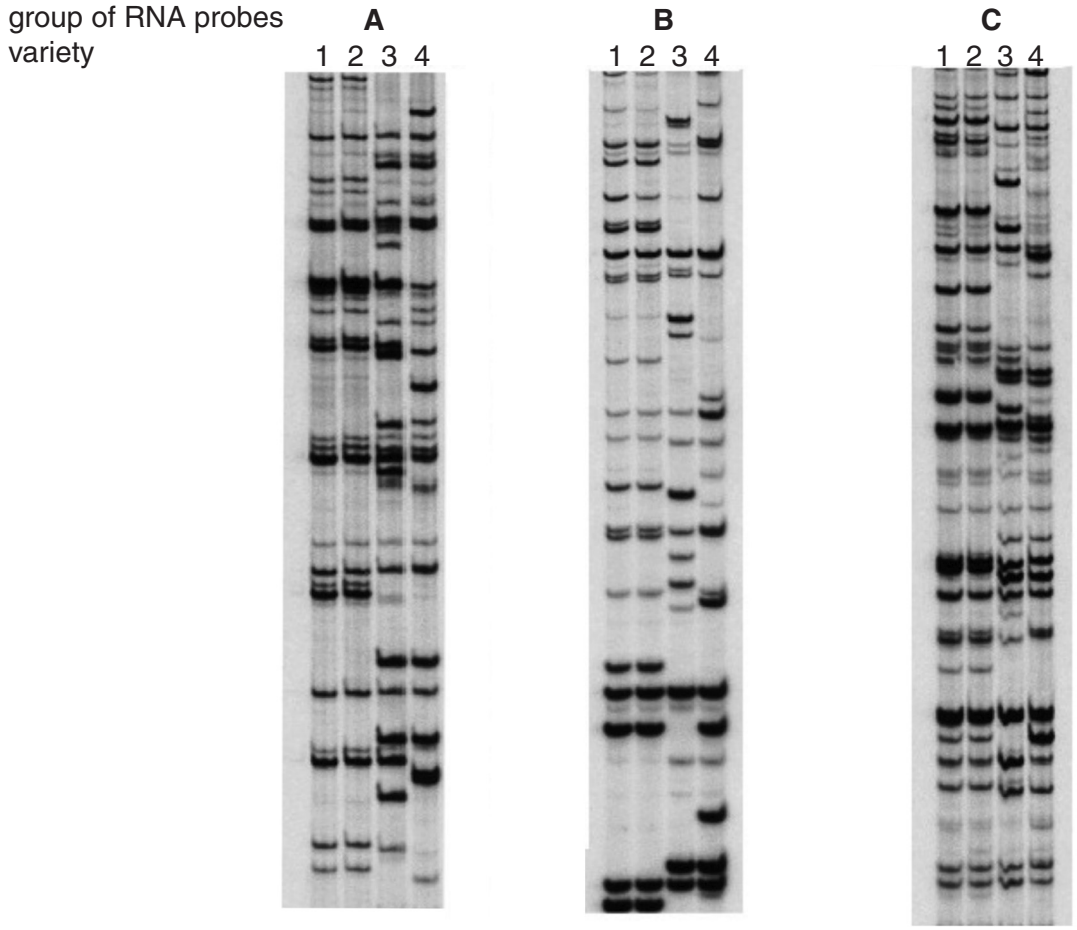


Fig. 1.1

(i) Fig. 1.1 shows that some DNA fragments are present in the genetic fingerprints of all four varieties, 1, 2, 3 and 4, when the different groups of RNA probes, **A**, **B** and **C**, are used.

On Fig. 1.1:
- draw an arrow (⟶) to indicate **one** DNA fragment found in all four varieties when RNA probes in group **A** were used.
- draw a second arrow for the RNA probes used in group **B**
- draw a third arrow for the RNA probes used in group **C**. [1]

(ii) State the varieties that appear to have the same genetic fingerprint. Give the evidence for your answer.

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.....[2]

[Total: 14]

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2 **(a)** Green manuring is a method of fertilising soil. One type of plant is grown for a certain period of time and then ploughed into the soil while the plants are still green.

A field investigation into the effect of green manuring on the yield of *Sorghum bicolor* was carried out using a legume. Different parts of the legume were used as green manure.

The *Sorghum* was grown in four types of trial plot as follows:

- no treatment (control)
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A random number generator was used to locate each of the trial plots.

trial plots, 5 m × 5 m, separated by fences

control	shoots and roots	roots only	control	roots only	roots only	shoots and roots	shoots only
shoots and roots	roots only	shoots only	shoots only	control	shoots and roots	shoots only	control

Fig. 2.1

The trial plots were left for one month before the *Sorghum* grain was sown.

Fig. 2.2 shows how the grain was sown in each trial plot.

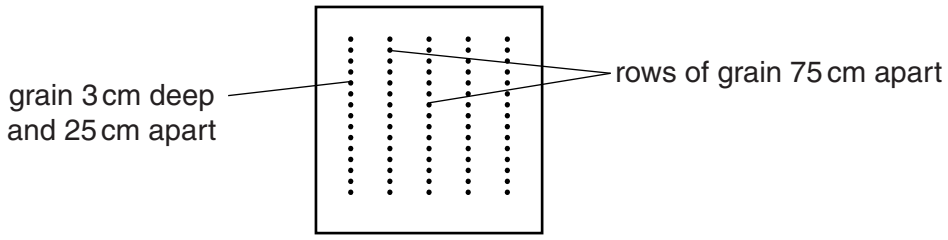


Fig. 2.2

(i) Identify two variables that have been standardised in this field investigation.

1. .....

2. .....[1]

(ii) Suggest two **abiotic** variables that **cannot** be standardised in this field investigation.

1. .....

2. .....[2]

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(iii) Suggest why the plots used to grow the *Sorghum* were left for one month before the sowing of the grain.

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.....[1]

(iv) State how this experimental design helps to ensure the reliability of the results.

.....

.....[1]

(b) *Sorghum* from each of the trial plots was collected and the dry mass of the different parts of the plants determined. The mean dry mass of shoot, roots and grain was calculated.

Table 2.1 shows the results of this investigation.

Table 2.1

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.....[2]

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Calculate the ratio of the increase in the mean dry mass of grain caused by using legume shoots in comparison to using legume roots only as green manure. Show your working.

.....[2]

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(c) Table 2.2 shows the results of *t*-tests that were carried out to compare the effect of the different types of green manure on the dry mass of *Sorghum.*

In each test the plants with added manure were compared with the control.

Table 2.2

treatment	P < 0.05			
	shoots	roots	grain	whole plant
legume roots only	not significant	not significant	not significant	not significant
legume shoots only	not significant	not significant	significant	significant
legume roots and shoots	significant	significant	significant	significant

(i) Suggest a null hypothesis for the statistical tests.

.....

.....[1]

(ii) State a reason why *t*-tests were used to find out if the effect of the different types of green manure on the dry mass of *Sorghum* was significant.

.....

.....[1]

(iii) Explain what is meant by:

statistically significant

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P < 0.05.

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.....[2]

(d) With reference to Table 2.1 and Table 2.2 only, state three conclusions that can be drawn from these results about the effect of using green manure on *Sorghum.*

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2. .....

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3. .....

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[Total: 16]

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