Cambridge Assessment International Education

Cambridge International AS & A Level

CANDIDATE NAME

CENTRE NUMBER | | | | | |

CANDIDATE NUMBER | | | | |

3997498716

BIOLOGY **9700/52**

Paper 5 Planning, Analysis and Evaluation **February/March 2023**

1 hour 15 minutes

You must answer on the question paper.

No additional materials are needed.

INSTRUCTIONS

- Answer **all** questions.
- Use a black or dark blue pen. You may use an HB pencil for any diagrams or graphs.
- Write your name, centre number and candidate number in the boxes at the top of the page.
- Write your answer to each question in the space provided.
- Do **not** use an erasable pen or correction fluid.
- Do **not** write on any bar codes.
- You may use a calculator.
- You should show all your working and use appropriate units.

INFORMATION

- The total mark for this paper is 30.
- The number of marks for each question or part question is shown in brackets [ ].

This document has **20** pages. Any blank pages are indicated.

1 Potatoes are underground organs made by the potato plant, *Solanum tuberosum*.

Fig. 1.1 shows a potato plant.

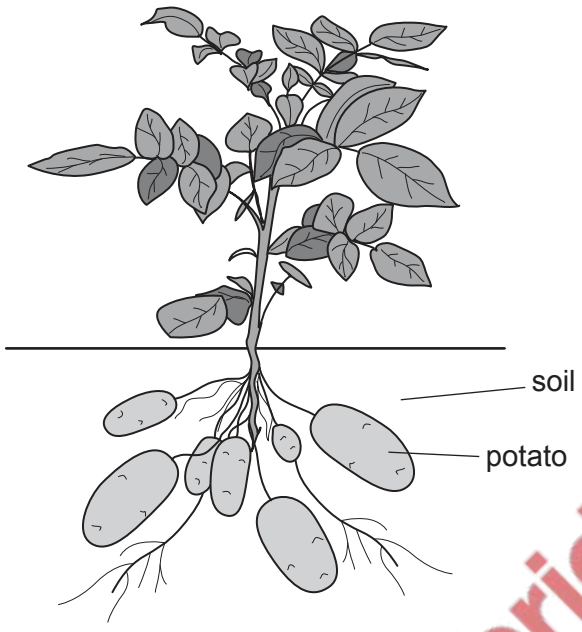

Fig. 1.1

Potatoes contain high quantities of starch and are a popular food. After harvesting, potatoes are stored. During storage, the starch content of potatoes gradually decreases due to the breakdown of starch molecules into glucose.

A glucose assay using a Benedict's test and a colorimeter can be used to determine the concentration of glucose in potatoes.

The glucose assay uses these steps:

- The outer skin is removed from a potato.
- The potato is cut into small pieces that are then ground into a pulp using a mortar and pestle.
- The potato pulp is filtered to remove most of the solids and obtain potato juice.
- Excess Benedict's solution is added to the potato juice.
- The mixture is heated in a water-bath at 90 °C for five minutes. During this five-minute period, copper ions (Cu^{2+}) in the Benedict's solution react with glucose in the potato juice to form an insoluble precipitate.
- The mixture is filtered to remove the precipitate. The filtrate, which is blue, contains copper ions that did not react with the glucose in the potato juice while the mixture was being heated.
- A sample of the blue filtrate is transferred to a colorimeter tube (cuvette).
- A colorimeter is used to measure the absorbance of the blue filtrate.
- A calibration curve is used to determine the glucose concentration of the potato juice from the measurement of absorbance.

Fig. 1.2 shows one type of colorimeter.

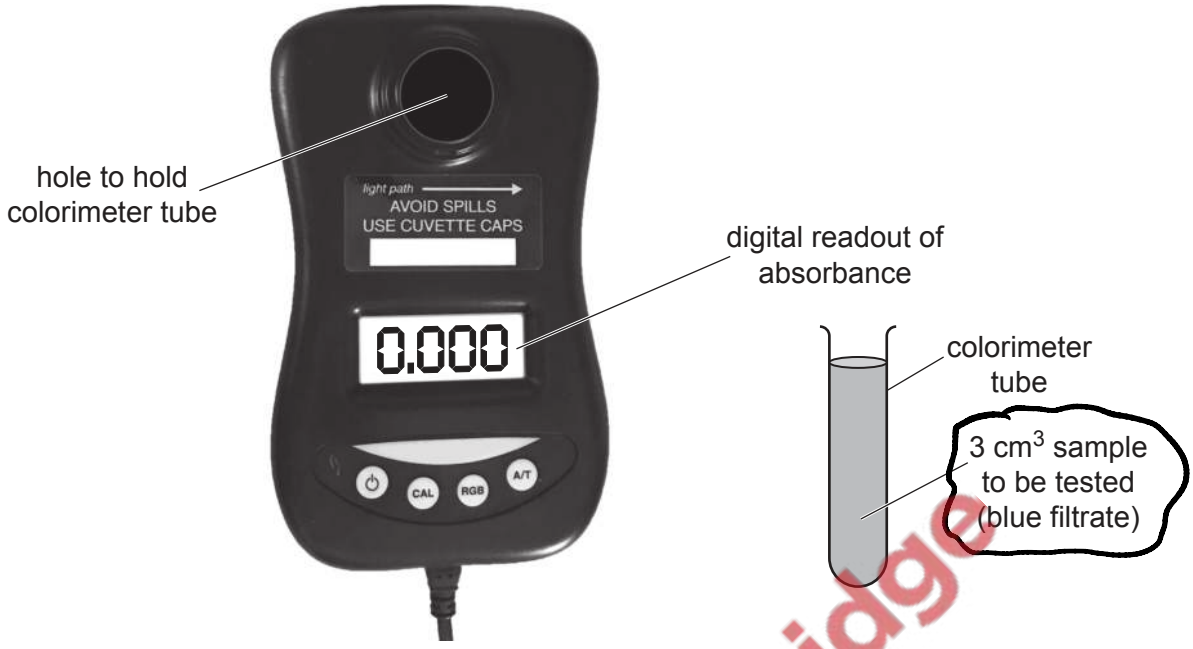

Fig. 1.2

A clean colorimeter tube containing the sample to be tested is placed in the colorimeter. Light is passed through the sample and the absorbance of light by the sample is measured.

(a) (i) Outline how colorimeters should be prepared **before** carrying out measurements on samples so that correct absorbance readings in the glucose assay are obtained.

Measure the absorbance of a colorimeter tube containing distilled water & set this absorbance to zero. [1]

dilutions → simple 2·0, 1·5, 1·0, 0·5
serial → constant dilution factor
2·0
0·2
0·02
0·002

To determine the concentration of glucose in potato juice using the glucose assay, a calibration curve needs to be produced. This involves using the glucose assay to obtain absorbance readings for standard glucose solutions of known concentrations.

(ii) A student was given a 2.0% stock solution of glucose from which to prepare a range of standard glucose solutions of known concentrations.

State the glucose concentrations the student could use to produce a calibration curve **and** describe how **20 cm^3** of each solution should be prepared by **proportional** dilution of the 2.0% stock solution of glucose.

simple.

You may use a table to show your answer.

glucose concentrations to use: 2.0%, 1.5%, 1.0%, 0.5%, 0.0%

how to prepare:

$C_1V_1 = C_2V_2$

(2) $V_1 = (1.5)(20)$

glucose conc. / %	Vol. of stock sol. / cm^3	Volume of distilled H_2O / cm^3
2.0	20	0
1.5	15	5
1.0	10	10
0.5	5	15
0.0	0	20

[3]

(iii) Sketch, on Fig. 1.3, a graph of the expected calibration curve that the student would obtain.

Include the axis labels.

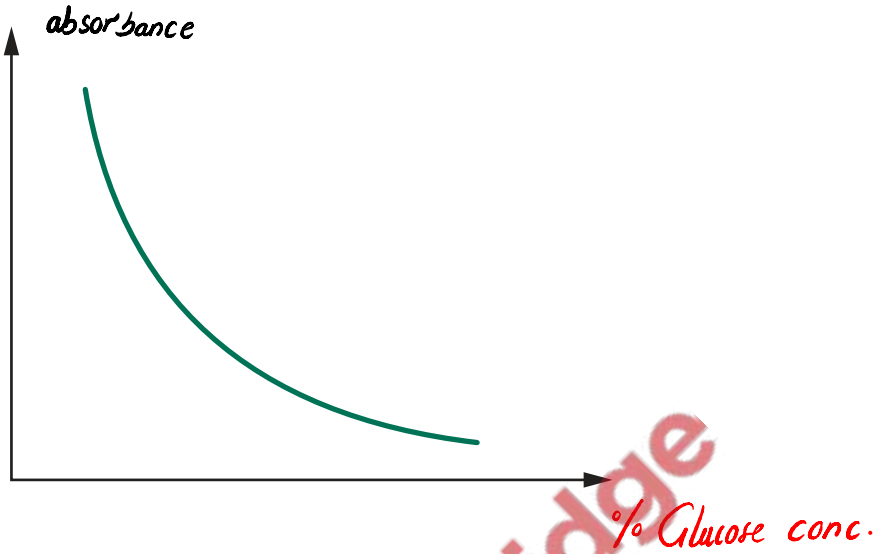

Fig. 1.3

[2]

(b) The student decided to investigate the effect of storage time on the glucose concentration of potatoes.

(i) Identify the **dependent** variable in this investigation.

Glucose concentration of the potato tissue [1]

(ii) The student was provided with freshly harvested potatoes, standard laboratory apparatus and a colorimeter.

Describe a method, using the glucose assay, that the student could use to investigate the effect of **storage time** on the glucose concentration of potatoes.

Your method should be set out in a logical order and be detailed enough to allow another person to follow it.

Details of how to carry out the glucose assay and how to prepare and use the colorimeter should **not** be included.

- use the same variety of potatoes
- Store the potatoes in similar storage condition which include
  - Low temperature
  - Low humidity
- Choose an appropriate range of storage times
  e.g. Every week for 5 consecutive weeks.
- at each storage time, potatoes are grounded to extract 2-3 cm^3 of Potato juice
- Measure absorbance using a Colorimeter
- Take atleast three readings at each storage time and calculate the mean
- Wear gloves to avoid irritation due to Benedict's Solution. [6]

Question 1 continues on page 8.

When potatoes are cooked at high temperatures to make potato chips, glucose and amino acids react together. This reaction is known as the Maillard reaction and is responsible for the orange-brown colour of potato chips.

When glucose reacts with the amino acid asparagine, acrylamide is made.

This reaction is shown in Fig. 1.4.

$$\text{asparagine} + \text{glucose} \xrightarrow[\text{cooking} > 120\,^{\circ}\text{C}]{\text{high temperature}} \text{acrylamide}$$

Fig. 1.4

Low concentrations of acrylamide are produced when potatoes are cooked at high temperatures to make potato chips. High concentrations of acrylamide are toxic to humans and can increase the risk of certain cancers developing.

Exposing raw potatoes to gamma radiation decreases the acrylamide concentration of potato chips.

Some scientists investigated the effect of gamma radiation on the composition of raw potatoes.

- Four bags of one variety of potato were each exposed to a different dose of gamma radiation: $0\,J\,kg^{-1}$, $50\,J\,kg^{-1}$, $100\,J\,kg^{-1}$ and $150\,J\,kg^{-1}$.
- Three samples of raw potatoes from each bag were then analysed chemically to determine the concentrations of glucose, protein and volatile nitrogen compounds as a percentage of dry mass. Volatile nitrogen compounds are present in potatoes due to the breakdown of some proteins and amino acids, such as asparagine.
- Statistical tests were carried out on the data to assess whether the gamma radiation dose affected the concentrations of glucose, protein and volatile nitrogen compounds in the raw potatoes.

Data from the chemical analysis and the statistical tests are shown in Table 1.1.

Table 1.1

gamma radiation dose / J kg^{-1}	mean composition of the raw potato sample as a percentage of dry mass		
	glucose	protein	volatile nitrogen compounds
0	0.83	10.92	1.14×10^{-2}
50	0.86	11.04	0.75×10^{-2}
100	0.83	10.08	0.68×10^{-2}
150	0.88	10.96	0.40×10^{-2}
statistical significance of differences between raw potato samples exposed to different doses of gamma radiation	not significant at $p < 0.05$ ✓	not significant at $p < 0.05$ ✓	significant at $p < 0.05$

~ breakdown of Asparagine.

(c) The scientists suggested that gamma radiation reduces the acrylamide content of potato chips by affecting asparagine.

Explain how the information provided **and** the data in Table 1.1 support this view.

Volatile nitrogen compounds decrease significantly with increase in gamma radiation dose. Which indicates decrease in asparagine so less acrylamide is made.

[2]

(d) In a further study, the scientists investigated whether gamma radiation **and** a hot water treatment reduce the acrylamide concentration of potato chips.

- Four bags of one variety of potato were each exposed to a different dose of gamma radiation: 0 J kg^{-1}, 50 J kg^{-1}, 100 J kg^{-1} or 150 J kg^{-1}.
- After exposure to gamma radiation, the potatoes from each bag were cut into 1 cm × 1 cm × 5 cm blocks.
- Potato blocks from each bag were given two different treatments.

- **treatment 1**: 100 g of potato blocks were cooked in 1 dm^3 of sunflower oil at 170 °C for five minutes. gamma radiation only. 0·5 dm^3

treatment 2: 100 g of potato blocks were heated in 500 cm^3 water at 85 °C for five minutes, removed from the water and then cooked in 1 dm^3 of sunflower oil at 170 °C for five minutes. gamma rad. & HOT WATER

The scientists measured the acrylamide concentration of the potato chips after these treatments.

This procedure was repeated a further five times to allow statistical tests to be carried out on the results.

The results of the investigation are summarised in Table 1.2.

Table 1.2

gamma radiation dose / J kg^{-1}	mean acrylamide concentration of potato blocks / µg kg^{-1}	
	treatment 1	treatment 2
0	4551	1768
50	3629	1487
100	3310	1339
150	2073	1010

(i) Using the data in Table 1.2, calculate the percentage decrease in the mean acrylamide concentration of potato blocks given treatment 2 compared to potato blocks given treatment 1, for a gamma radiation dose of 0 J kg^{-1}.

Show your working.

$$\frac{1768 - 4551}{4551} \times 100$$

percentage decrease = 61 % .....

[2]

The results in Table 1.2 are shown as a graph in Fig. 1.5. Error bars have been added to show 95% confidence intervals (95% CI).

No OVERLAPPING

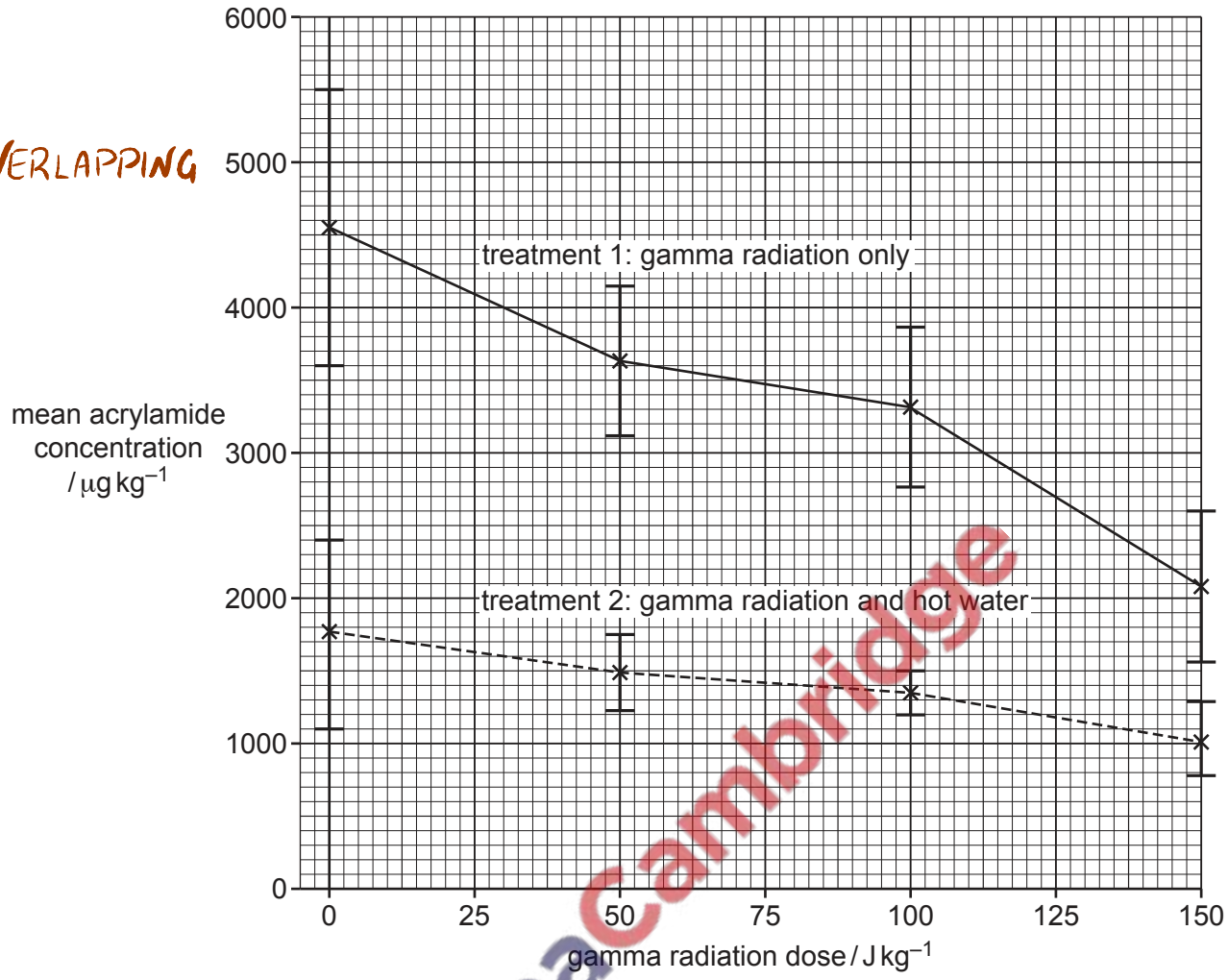

Fig. 1.5

The scientists concluded that a combination of gamma radiation **and** hot water treatment is the most effective way to reduce the acrylamide concentration of potato chips.

(ii) Explain how the information shown in Table 1.2 and Fig. 1.5 supports, or does **not** support, this conclusion.

SUPPORT

treatment 2 decreases the acrylamite concentration in potato blocks more than treatment 1

the non overlapping error bars indicate that treatment II is significantly better than treatment 1.

NOT SUPPORT

hot water treatment alone might be safer.

[3]

[Total: 20]

2 Rice, *Oryza sativa*, is an important food crop. Rice plants are wind pollinated. Pollen containing the male gamete is transferred by the wind to female reproductive organs of rice plants. Fertilisation and grain formation then occur.

Weedy rice is a wild form of rice that grows in fields of cultivated rice. Weedy rice competes with cultivated rice for resources. Weedy rice plants are taller than cultivated rice plants and produce a low yield of rice grains.

Several genetically modified (GM) varieties of cultivated rice have been developed.

One concern about the use of GM rice is gene flow from GM rice plants to weedy rice. Gene flow occurs when the wind carries pollen from GM rice plants to weedy rice plants.

Some scientists investigated gene flow from herbicide-resistant GM rice plants to weedy rice plants. The GM rice plants had a gene for herbicide resistance. Weedy rice plants do not have the gene for herbicide resistance.

The scientists wanted to test the hypothesis that:

> Gene flow from GM rice to weedy rice decreases as the distance between the GM rice crop and weedy rice increases.

The scientists planted GM rice and weedy rice in a field, as shown in Fig. 2.1.

Fig. 2.1 also shows that the wind normally blows in a north–west (NW) direction.

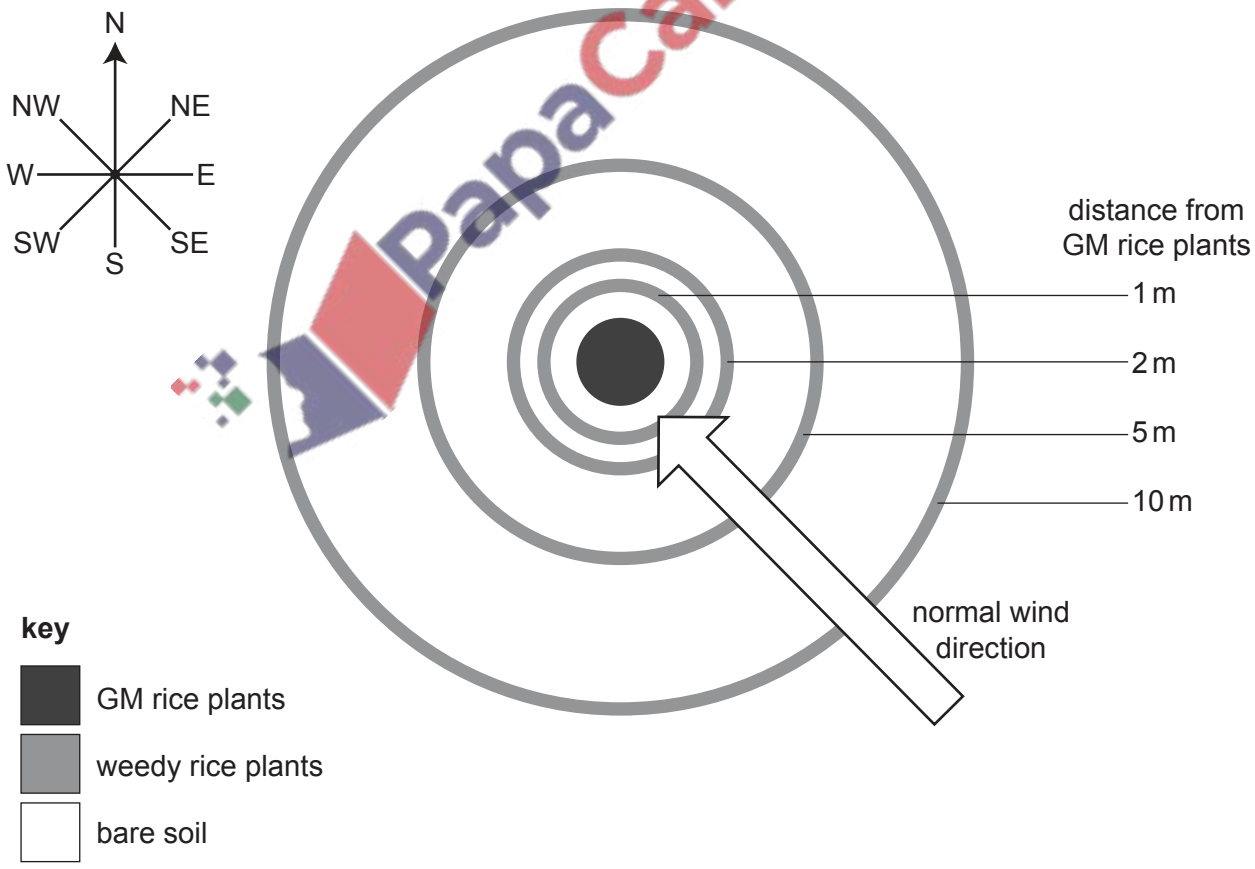

Fig. 2.1

After the plants had been pollinated and the rice grains had developed, the grains were collected from the **weedy** rice plants **only**.

- Grains were collected from all weedy rice plants growing 1 m from the GM rice plants in the directions N, NE, E, SE, S, SW, W and NW.
- Grains were also collected in the same directions from the GM rice plants at distances of 2 m, 5 m and 10 m.
- Approximately 1000 grains from each collection point were planted and germinated in controlled conditions in a glasshouse.
- The plants were grown for three weeks and then tested to determine whether they had the gene for herbicide resistance.

Fig. 2.2 shows young rice plants three weeks after germination.

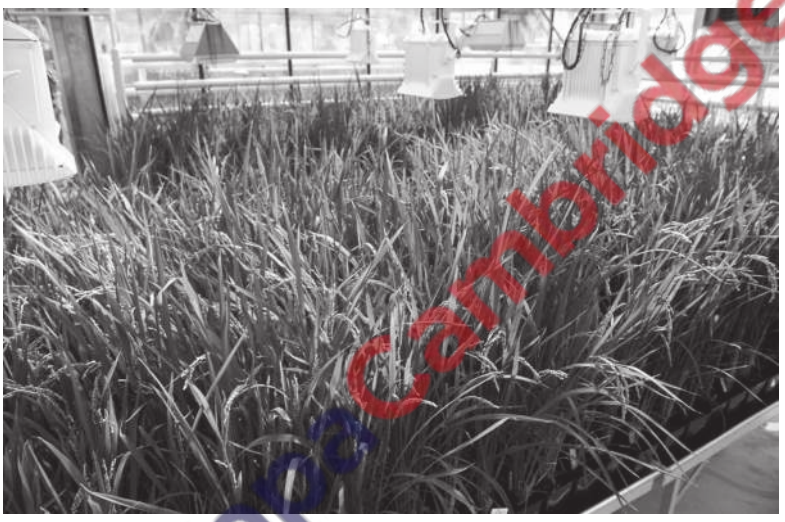

Fig. 2.2

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

distance from the GM rice plants

Direction of wind blow from GM rice plants [1]

(ii) Some of the young weedy rice plants had the gene for herbicide resistance. This gene had been carried to the parents of these plants in pollen from the GM rice plants.

Outline a method the scientists could use in the glasshouse to determine how many of the young weedy rice plants had the gene for herbicide resistance.

Your method should **not** require the extraction of nucleic acids.

Apply herbicide spray to rice plants and count the number of plants that survive.

..... [2]

(iii) The scientists calculated the percentage of young weedy rice plants that had the gene for herbicide resistance. This percentage was used as a measure of gene flow.

The percentage gene flow recorded at each NW collection point is shown in Fig. 2.3.

Fig. 2.3 also shows the percentage gene flow recorded at each distance from the GM rice plants at all the other collection points combined.

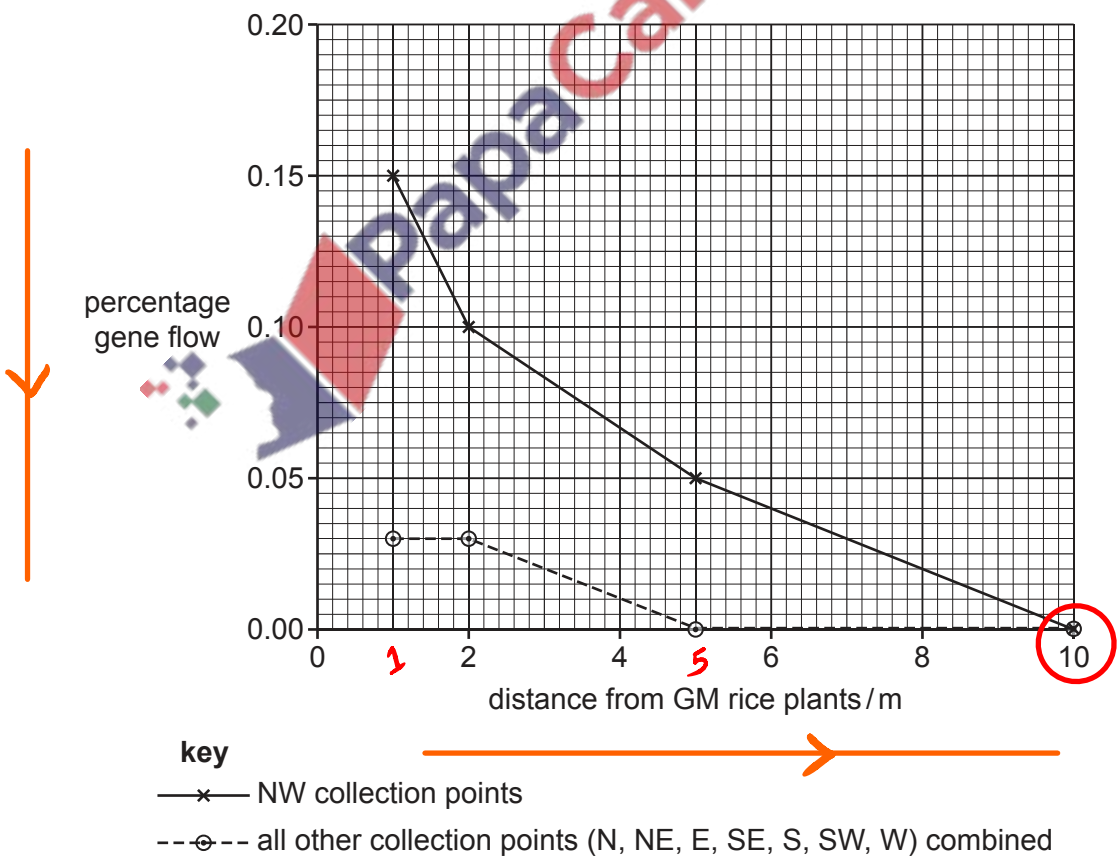

Fig. 2.3

State **two** conclusions that can be made from the data shown in Fig. 2.3.

As the distance from the GM rice plant increases, the percentage gene flow decreases

Percentage gene flow is 0 at 10m

..... [2]

(b) Weedy rice plants have a gene that results in increased height. GM rice plants do **not** have this gene. This gene allows gene flow from weedy rice plants to GM rice plants to be investigated.

In a second study, the scientists investigated gene flow from herbicide-resistant GM rice plants to weedy rice plants **and** gene flow from weedy rice plants to herbicide-resistant GM rice plants.

The scientists planted GM rice and weedy rice in areas next to each other in a field.

After the plants had been pollinated and the rice grains had developed, 1000 grains were collected from weedy rice plants at each of eight sampling sites **and** 1000 grains were collected from GM rice plants at each of eight sampling sites.

Plants were grown from the grains that had been collected and tested to determine the percentage gene flow between the weedy rice plants and the GM rice plants at each site.

Table 2.1 shows the results of the investigation.

Table 2.1

	percentage gene flow from GM rice plants to weedy rice plants	percentage gene flow from weedy rice plants to GM rice plants
	0.044	0.136
	0.013	0.136
	0.025	0.273
	0.019	0.235
	0.044	0.239
	0.025	0.242
	0.063	0.244
	0.057	0.273
mean:	0.036	0.222

(i) Complete Table 2.1 by calculating the mean percentage gene flow from GM rice plants to weedy rice plants.

Mean ~ 0.036

[1]

(ii) The scientists decided to carry out a *t*-test to compare the percentage gene flow from GM rice plants to weedy rice plants with the percentage gene flow from weedy rice plants to GM rice plants.

The scientists stated the null hypothesis:

> There is no difference between the percentage gene flow from GM rice plants to weedy rice plants and the percentage gene flow from weedy rice plants to GM rice plants.

The scientists calculated the value of *t* as **9.043**.

Table 2.2 shows the probability table for the *t*-test.

Table 2.2

degrees of freedom	critical values	
	$p = 0.05$ (5%)	$p = 0.01$ (1%)
12	2.179	3.055
13	2.160	3.012
14	2.145	2.977
15	2.131	2.947
16	2.120	2.921
17	2.110	2.898
18	2.101	2.878

$df = n_1 + n_2 - 2$

$8 + 8 - 2$

$df = 14$

$t = 9.043$

Explain, with reference to Table 2.2, what can be concluded from the analysis of the data collected by the scientists.

Critical t at $p = 0.05$ is 2.145

Calculated t is greater than critical t => $p < 0.05$

null hypothesis is rejected as

there is a significant difference in the percentage gene flow from GM rice plants to weedy rice plants and from weedy rice plants to GM rice plants

..... [3]

(iii) The results of this investigation were published in a scientific paper.

A student who read the paper concluded that the results showed there were no reasons to be concerned about gene flow from GM plants to wild plants.

Suggest **one** reason why the results of this investigation should **not** be interpreted in this way.

No long term data.

Investigation carried out only once.

..... [1]

[Total: 10]

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Cambridge Assessment International Education

Cambridge International AS & A Level

CANDIDATE NAME									
CENTRE NUMBER						CANDIDATE NUMBER				

7875730187

BIOLOGY **9700/53**

Paper 5 Planning, Analysis and Evaluation **October/November 2022**

1 hour 15 minutes

You must answer on the question paper.

No additional materials are needed.

INSTRUCTIONS

- Answer **all** questions.
- Use a black or dark blue pen. You may use an HB pencil for any diagrams or graphs.
- Write your name, centre number and candidate number in the boxes at the top of the page.
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- You should show all your working and use appropriate units.

INFORMATION

- The total mark for this paper is 30.
- The number of marks for each question or part question is shown in brackets [ ].

This document has **16** pages. Any blank pages are indicated.

1 Antibiotic resistance in bacteria is a global problem that has caused scientists to research into antibacterial substances other than antibiotics. Honey has properties that make it a good antibacterial substance. For example, honey contains hydrogen peroxide, which is known to kill bacteria.

The most effective honey tested so far for antibacterial activity is Manuka honey. It contains less hydrogen peroxide than many other types of honey, but it does contain an antibacterial compound, methylglyoxal (MGO), which is not found in other types of honey.

A student decided to investigate the effect of two antibacterial substances on the bacterium *Bacillus subtilis*, which respires aerobically:

- MGO in Manuka honey
- an antibiotic solution used in cell cultures to prevent contamination.

The student wanted to find the lowest concentration of each antibacterial substance that would kill or inhibit the growth of *B. subtilis*.

(a) The student used a broth culture for the investigation. To make a broth culture, a small quantity of *B. subtilis* is added to a clear nutrient solution. A fresh (newly made) broth culture of *B. subtilis* is also clear.

Fig. 1.1 is a diagram of a fresh broth culture of *B. subtilis*.

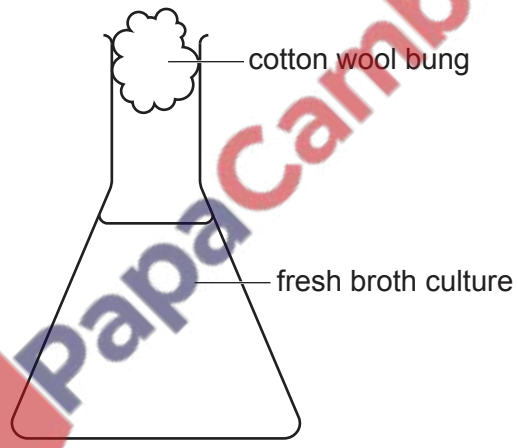

Fig. 1.1

(i) A sterile cotton wool bung was used in the top of the flask containing the fresh broth culture of *B. subtilis* to protect the culture from contamination.

Explain why it is better to use a sterile cotton wool bung in a flask containing broth culture of *B. subtilis*, rather than using a sterile rubber bung.

To enable oxygen to pass through for aerobic respiration [1]

(ii) Before comparing the two antibacterial substances, the student carried out a trial experiment.

The student transferred a sample of fresh broth culture to a culture tube and incubated the tube at 25 °C for 24 hours.

Fig. 1.2 summarises the results of the trial experiment.

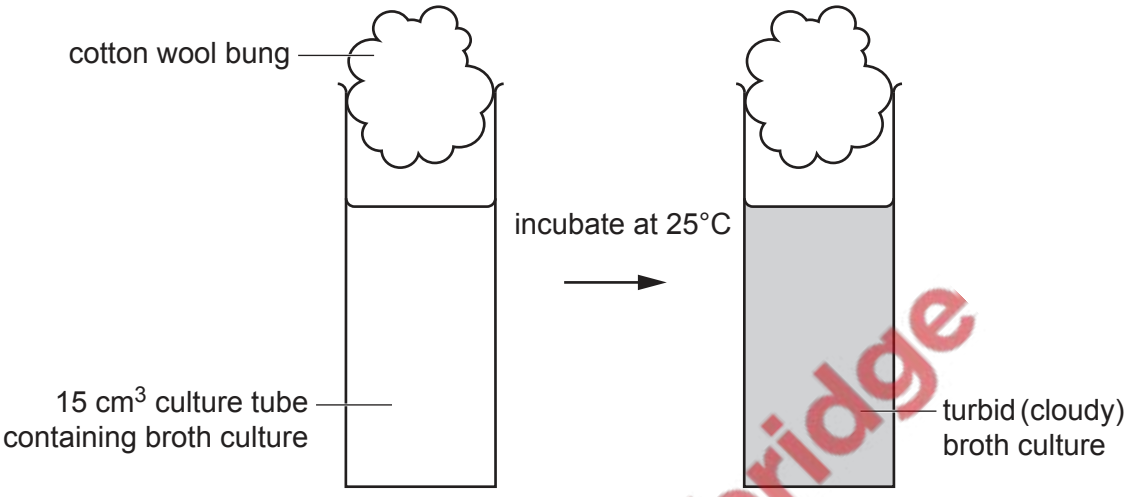

Fig. 1.2

The student decided that turbidity of the broth culture is a measure of bacterial population growth (bacterial growth).

Explain how this concept can be used in an investigation to measure the extent of bacterial growth.

the greater the turbidity, the greater is the growth of bacteria

..... [1]

(b) The student decided to test the antibiotic solution before testing the Manuka honey.

In addition to normal laboratory apparatus and materials, the student was provided with:

- a fresh broth culture of *B. subtilis*
- a clear antibiotic stock solution
- nutrient solution to dilute the antibiotic stock solution
- 15 cm^3 flat-bottomed glass culture tubes with sterile cotton wool bungs
- a choice of graduated pipettes to measure volumes accurately: 0.2 cm^3, 2.0 cm^3, 10.0 cm^3, 25.0 cm^3.

The student:

- prepared dilutions of the antibiotic stock solution and added a volume of each to different culture tubes
- added a volume of fresh broth culture of *B. subtilis* to each culture tube
- incubated the culture tubes in an incubator
- allowed time for bacterial growth to occur and then checked each culture tube
- recorded and analysed the results
- decided on the lowest concentration of antibiotic solution that appeared to kill or inhibit the growth of *B. subtilis*.

Outline a control for this part of the investigation.

use nutrient solution instead of antibacterial solution.

..... [1]

(c) In the next part of the investigation, the student used a stock solution of Manuka honey. The student remembered that hydrogen peroxide could be present but could not think of a way to break down the hydrogen peroxide to remove it from the solution.

Describe how the student can improve the investigation by removing hydrogen peroxide from the Manuka honey solution **and** explain why this improvement makes the results more valid.

Use peroxidase to breakdown H_2O_2

This ensures that bacterial killing is due to MGO in Manuka Honey.

..... [2]

(d) The student was provided with a stock solution of Manuka honey containing an MGO concentration of 600 $\mu g\,cm^{-3}$. This was a clear solution, labelled '100% honey'.

The same apparatus and materials were available.

Describe how the student could prepare a 10% solution of honey using the stock solution.

Construct a table to show how the dilution is made for the 10% solution and the other concentrations that the student could use.

Add 90cm³ of nutrient solution to a beaker containing 10cm³ of 100% honey to make 100cm³ of 10% solution of honey.

Space for table.

Conc/%	Vol. of 10% sol./cm³	Volume of nutrient sol/cm³
10	10	0
8	8	2
6	6	4
4	4	6
2	2	8

[2]

(e) State the independent variable **and** dependent variable for the part of the investigation involving Manuka honey solution.

independent variable ..... Concentration of Manuka Honey solution .....

dependent variable ..... turbidity .....

[2]

(f) Predict the results the student would expect when investigating the effect of Manuka honey on *B. subtilis*.

Explain the reasoning behind the prediction.

The greater the % conc. of Manuka honey solution, the more clear is the solution. This is because MGO in Manuka honey kills the bacteria.

[2]

(g) Describe how the student could determine the lowest concentration of Manuka honey solution that would kill or inhibit the growth of *B. subtilis*.

- Do **not** repeat any detail given in **(d)** of how to prepare the different concentrations of Manuka honey solution
- Do **not** give details of using aseptic technique (techniques to prevent contamination of the student, the environment or other people).

Your method should be set out in a logical way and be detailed enough to let another person follow it.

Use the same volume of broth cultures in all culture tubes

Use the same volume of Manuka honey solution in all culture tubes

Ensure that the broth culture and honey solution are properly mixed together.

Incubate at 25°C for 24 hours

measure the turbidity using a colorimeter

Determine the minimum concentration at which the solution is clear.

Repeat readings at each concentration of Manuka honey solution and determine the mean turgidity

Wear Gloves to avoid allergic reaction to Manuka Honey.

[5]

(h) MRSA, methicillin-resistant *Staphylococcus aureus*, is an example of antibiotic resistance in bacteria. There is evidence that medical-grade Manuka honey is effective in treating wounds infected with MRSA. This honey has been sterilised by gamma irradiation and filtered to remove contaminants.

A study was carried out to see if another type of honey, Germania honey, is as effective as Manuka honey in killing bacteria removed from wounds of 50 people with MRSA.

Five different concentrations of each type of honey were compared. The concentrations were numbered 1 to 5, with 1 being the highest concentration and 5 the lowest concentration.

At the concentrations where there was no visible growth in a broth culture of *S. aureus*, the researchers transferred samples onto nutrient agar plates containing no antibacterial substance. Incubation of these plates confirmed that there was no bacterial growth.

The results were analysed using the chi-squared (χ^2) test.

Table 1.1 shows the results of the study and the statistical analysis using the χ^2 test.

Table 1.1

concentration of honey M=Manuka G=Germania	number of cultures with bacterial growth	number of cultures with no bacterial growth	χ^2 value	significant
M1	2	48	5.005	yes
G1	9	41		
M2	5	45	13.306	Yes
G2	21	29		
M3	11	39	14.923	Yes
G3	30	20		
M4	43	7	3.052	No
G4	48	2		
M5	47	3	1.042	no
G5	49	1		

(i) State a null hypothesis for the investigation.

There is no significant difference in the effect of Manuka and Germania honey in killing bacteria

..... [1]

(ii) Table 1.2 shows some critical values for χ^2 at different probabilities.

Table 1.2

degrees of freedom	probability						
	0.99	0.95	0.90	0.10	0.05	0.01	0.001
1	0.0002	0.0039	0.0158	2.706	3.841	6.635	10.827
2	0.0201	0.1026	0.2107	4.605	5.991	9.210	13.815

Use Table 1.2 to decide whether the χ^2 values for concentrations 2, 3 and 4 in Table 1.1 are significant or not significant. Write your decision in the final column of Table 1.1:

- write **yes** if the value is significant
- write **no** if the value is **not** significant

[1]

(iii) This study compared the effectiveness of the two honey varieties in treating wounds infected with MRSA.

State the conclusions that can be made from the results and statistical analysis of this study.

Manuka Honey is more effective than Germania because the difference in results is significant at conc. 1, 2 and 3.

[2]

[Total: 20]

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2 Holstein cattle have been selectively bred for their high milk production.

The seed of the cotton plant, *Gossypium* spp. is a good source of fibre, protein and carbohydrate. To improve milk production, whole cottonseed or cottonseed meal can be added to livestock feed. Cottonseed meal is cottonseed that has been processed by grinding.

Fig. 2.1**A** shows whole cotton seed and Fig. 2.1**B** shows cottonseed meal.

A **B**

Fig. 2.1

Free gossypol is a toxin found in cotton seeds. Some of this free gossypol binds to protein to form bound gossypol. This occurs during cottonseed meal production and in the rumen (forestomach) of the cow during microorganism fermentation.

Free gossypol is easily absorbed. Bound gossypol is absorbed less easily and is not toxic.

An investigation was carried out on the effect of different dry-matter diets on lactating (milk-producing) Holstein cows:

- 30 healthy cows were fed on the same cottonseed-free diet for 14 days.
- The cows were then divided equally into five groups, **A** to **E**, and fed on one of five experimental diets for 42 days.
- The quantity of milk produced each day was recorded.
- The cows had no visible signs of illness during the 42 days.

For each diet, Table 2.1 shows:

- the cottonseed content and total gossypol (free and bound) content
- the mean daily dry matter taken in during feeding (intake)
- the mean daily lactation performance (milk yield).

Table 2.1

SE = standard error

group	description of dry matter diet	cottonseed content / percentage of dry matter		total gossypol in diet / mg kg^{-1}	dry matter intake kg d^{-1} SE = 1.4	milk yield / kg d^{-1} SE = 1.3
		whole cottonseed	cottonseed meal			
A	cottonseed replaced by soybean meal	0	0	0	24.8	27.6
B	whole cottonseed	15.0	0	1039.7	23.6	29.7
C	cottonseed meal	0	7.0	900.1	23.2	27.9
D	mixed 1 (whole cottonseed and cottonseed meal)	7.5	3.5	959.7	22.6	28.7
E	mixed 2 (whole cottonseed and cottonseed meal)	15.0	7.0	1922.0	24.0	32.6

The differences in the dry matter intake for groups **A** to **E** were **not** statistically significant.

(a) Explain why the cows were all fed on the same cottonseed-free diet for 14 days.

to ensure all the cows are standardised before investigation

..... [1]

(b) Using the information provided and the results shown in Table 2.1, a student made some statements about the effect of feeding different diets to lactating Holstein cows.

(i) The student stated that if the investigation is repeated, the groups of cows will produce milk in this order:

(highest quantity) **E** → **B** → **D** → **C** → **A** (lowest quantity).

With reference to Table 2.1, explain why the statement made by the student is **not** supported by the data.

The milk yields in all groups are very close to each other

Standard Error overlaps implying no significant difference in the milk yields.

no statistical test carried to determine if the results are significantly different.

[3]

(ii) Scientists are developing cotton plants that are genetically modified to produce seeds lacking gossypol. Table 2.1 suggests that there may be a link between total gossypol in the diet and milk yield.

The student stated that repeating the investigation with no gossypol in diets **B** to **E** would result in a lower milk yield for each group.

Explain whether or not Table 2.1 provides enough evidence to support the statement made by the student.

As the gossypol content increases, the milk yield also increases.

At 1922 $mg\,kg^{-1}$ gossypol → 32.6 $kg\,d^{-1}$ milk yield

At 900.1 $mg\,kg^{-1}$ gossypol → 27.9 $kg\,d^{-1}$ milk yield

[2]

(c) Higher concentrations of gossypol absorbed into the circulation can cause an illness in cows known as gossypol toxicity. Low concentrations of gossypol can be detoxified in the liver.

(i) During the investigation, the gossypol intake for each cow was measured and analysed.

At the end of the investigation (day 42), the blood plasma gossypol concentration of each cow was also measured.

Fig. 2.2 shows the free gossypol intake for each cow, plotted against the concentration of gossypol in plasma on day 42.

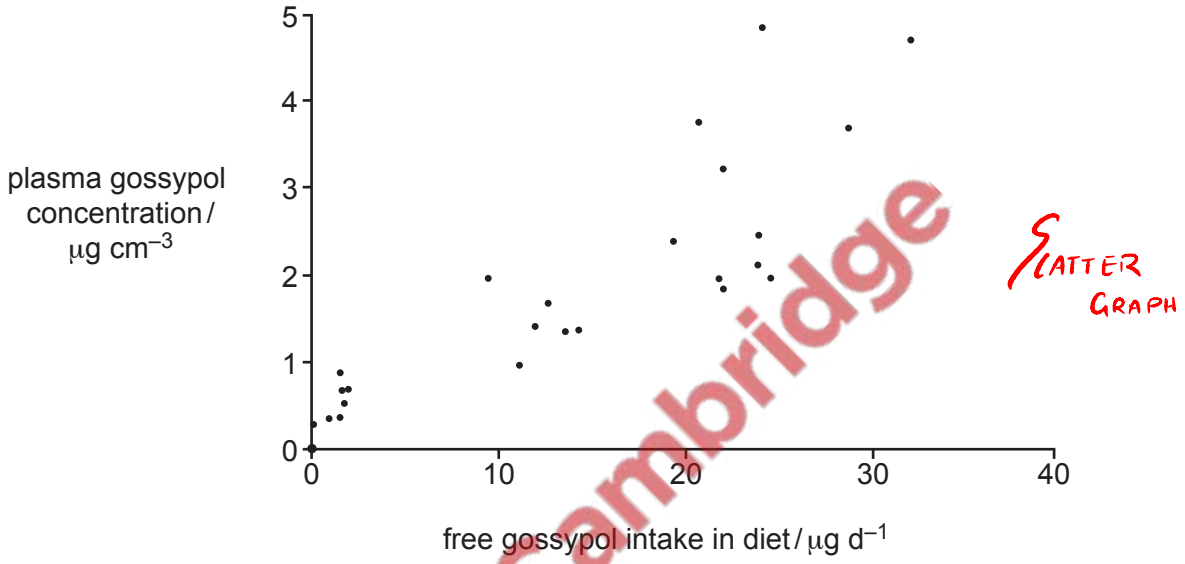

Fig. 2.2

With reference to Fig. 2.2, explain whether there is a relationship between free gossypol intake and plasma gossypol concentration on day 42.

.....

Positive Correlation

.....

.....

..... [1]

(ii) Gossypol produced by cotton plants can be found in two forms, the (–) isomer and the (+) isomer. The (–) isomer has higher biological activity within cells and is much more toxic than the (+) isomer. Both forms can bind protein to become bound gossypol.

Table 2.2 summarises the mean daily gossypol intakes for the five different experimental groups of lactating Holstein cows.

Table 2.2

group	description of dry matter diet	total gossypol intake / $g\,d^{-1}$	free gossypol intake / $g\,d^{-1}$	bound gossypol intake / $g\,d^{-1}$	(–) isomer intake / $g\,d^{-1}$
A	cottonseed replaced by soybean meal	0.0	0.0	0.0	0.0
B	whole cottonseed	23.5	22.4	1.1	9.4
C	cottonseed meal	21.0	1.5	19.5	8.4
D	mixed 1 (whole cottonseed and cottonseed meal)	21.9	12.1	9.8	8.8
E	mixed 2 (whole cottonseed and cottonseed meal)	45.7	25.2 ✓	20.5	18.3 ✓

The investigators knew that different diets presented different levels of risk of causing gossypol toxicity in cows. For example, the diet for group **A** cows did not present any risk of toxicity.

With reference to Table 2.2, discuss the extent to which the different experimental diets for groups **B**, **C**, **D** and **E** present a risk of causing gossypol toxicity.

Highest risk of toxicity is in Group E.

The risk decreases in the order, E → B → D → C

The greater the intake of free gossypol, the greater is the absorption of (-) isomer, and greater is the toxicity

[3]

[Total: 10]

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