



Oxford Cambridge and RSA

Wednesday 18 June 2025 – Morning

A Level Biology B (Advancing Biology)

H422/03 Practical skills in biology

Time allowed: 1 hour 30 minutes



You must have:

- a ruler (cm/mm)

You can use:

- a scientific or graphical calculator



Please write clearly in black ink. **Do not write in the barcodes.**

Centre number

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Candidate number

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First name(s)

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Last name

StemUp Model Solution

INSTRUCTIONS

- Use black ink. You can use an HB pencil, but only for graphs and diagrams.
- Write your answer to each question in the space provided. If you need extra space use the lined pages at the end of this booklet. The question numbers must be clearly shown.
- Answer **all** the questions.
- Where appropriate, your answer should be supported with working. Marks might be given for using a correct method, even if your answer is wrong.

INFORMATION

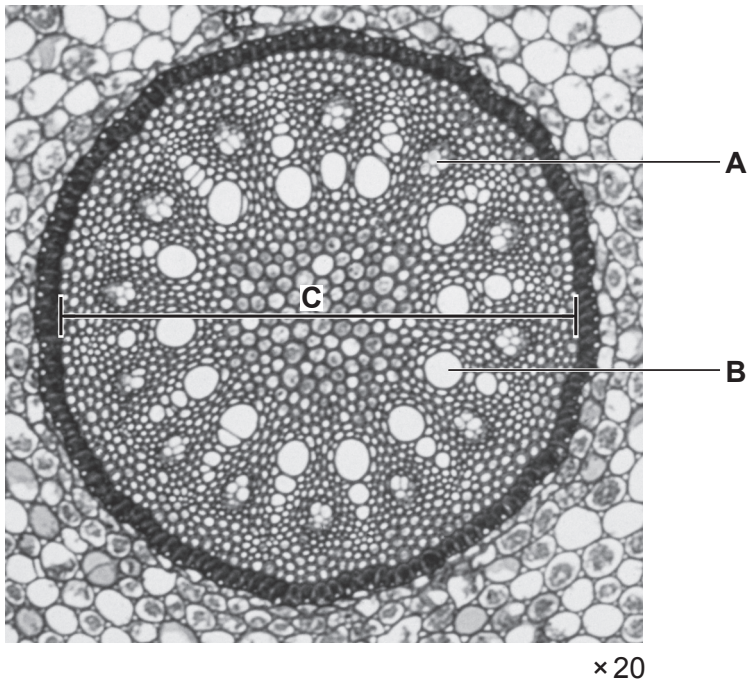
- The total mark for this paper is **60**.
- The marks for each question are shown in brackets [].
- Quality of extended response will be assessed in questions marked with an asterisk (*).
- This document has **20** pages.

ADVICE

- Read each question carefully before you start your answer.

1 Two functions of the roots of plants are to take up water from the soil and to store starch.

(a) The photomicrograph shows a transverse section of a root in a monocotyledonous plant.



(i) The cells labelled **A** and **B** are two different types of cell in the vascular tissue of the root.

Identify the types of cell labelled **A** and **B**.

A *sieve tube element (phloem)*

B *xylem vessel*

[2]

(ii) The diameter of one region of the root is represented by the line **C** in the image.

The magnification used to produce the image was $\times 20$.

Calculate the actual diameter of **C**.

Give your answer in mm.

image diameter of C = 68 mm

actual = image $\div$ magnification = 68 mm $\div$ 20 = 3.4 mm

Diameter = *3.4* mm [2]

(iii) One risk when dissecting plant tissues, such as roots, is the potential for injury when using sharp instruments.

State **one other** health and safety risk that should be considered before dissecting plant tissues.

Plant tissue or sap could cause an allergic reaction (skin irritation), so wear

non-latex gloves.

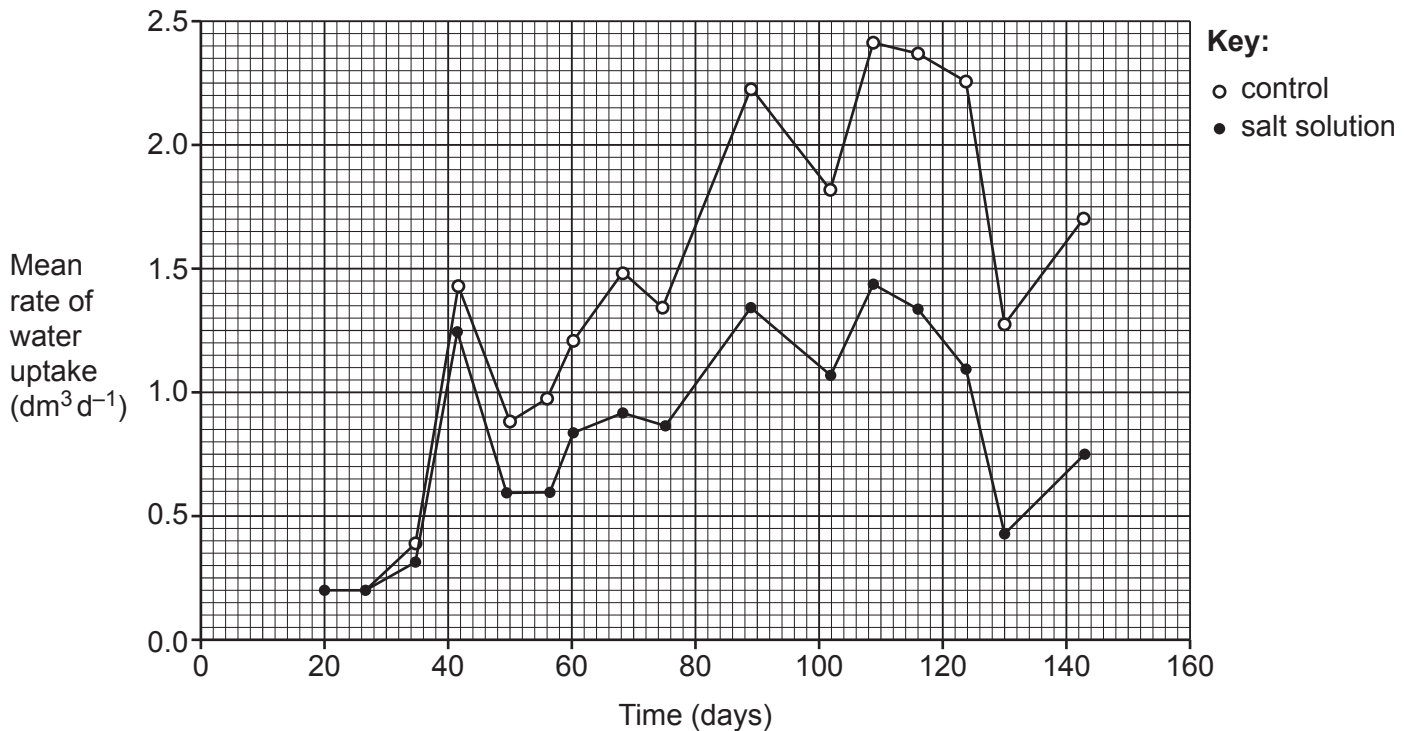
[1]

- (b) A scientist investigates how the rate of water uptake by roots is affected by the presence of salt (NaCl) in the surrounding soil.

This is the method they use:

- Grow 110 tomato plants in pots of soil in a greenhouse.
- Add 50 mmol dm^{-3} salt solution to the soil of 55 plants, every day.
- The other 55 plants are a control group.
- Calculate the rate of water uptake per day ($\text{dm}^3 \text{ d}^{-1}$) by the roots of each plant.

The results of the investigation are shown in the graph.



- (i) State how the group of control plants would be set up differently to the group of plants given salt solution **and** explain the purpose of this control group.

..Add an equal volume of distilled water to the soil of each control plant every.....
 ..day, instead of the salt solution.....
 ..This allows comparison, to show that any effect observed is only a result of.....
 ..the salt solution..... [2]

- (ii) Calculate the largest percentage difference in water uptake between the plants in the control group and the plants given salt solution.

Use your answer to complete the sentence.

largest difference is on day 124: control = 2.25, salt = 1.10

$$(2.25 - 1.10) \div 1.10 \times 100 = 104.5\% = 105\%$$

On day 124..... the water uptake of the control plants was 105.....% greater than the water uptake of the plants given the salt solution. [2]

(iii) Suggest what conclusions can be made from the results shown in the graph.

Water was still taken up even when salt was present, and in both groups the mean rate of water uptake fluctuates during the experiment.
Up to about day 42 there was little difference (salt had no effect between days 20 and 26), but after day 26 the salt reduced the mean rate of water uptake. [2]

(c) The roots of plants can store starch in high concentrations.

Outline how the starch concentration of a sample of root tissue could be estimated using colorimetry.

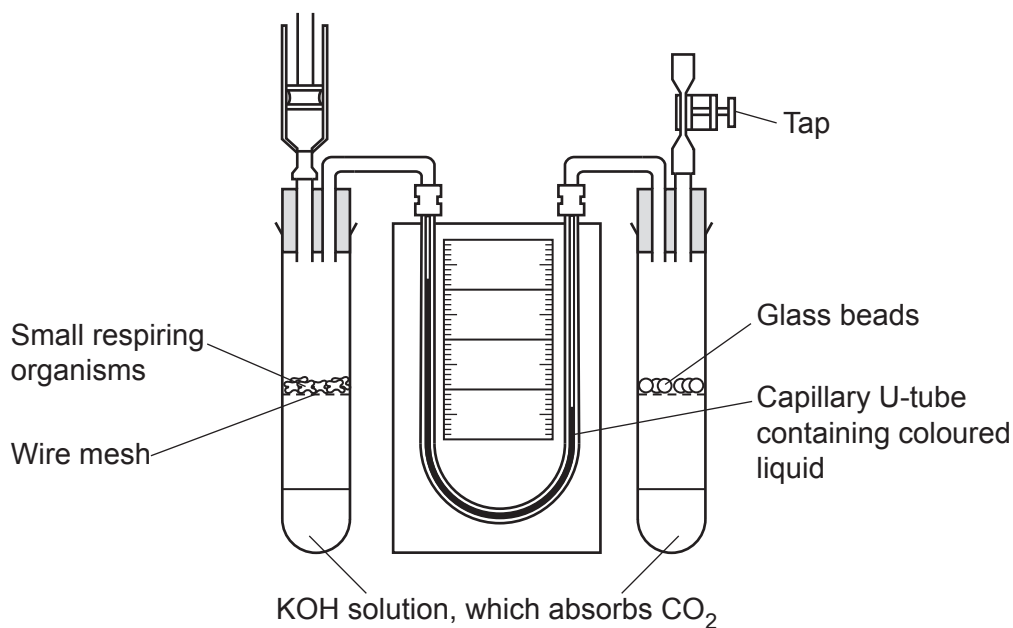
Grind a known mass of root in 50 cm³ of distilled water to make a starch solution.
Add iodine in potassium iodide solution, using the same volume and concentration for the sample and each standard.
Calibrate the colorimeter with a blank, select a red filter and measure absorbance.
Serially dilute a known starch solution to make standards and plot a calibration curve of absorbance against concentration.
Read the sample concentration off the calibration curve. [4]

5
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- 2 Respirometers can be used to measure the respiratory quotient and the rate of respiration of organisms.

The diagram shows a respirometer that can be used for these measurements.



The oxygen (O_2) used by organisms for respiration can be estimated by measuring the distance moved by the coloured liquid in the capillary U-tube over a set time period. The volume of O_2 can then be calculated.

- (a) A student measures the volume of O_2 used and the volume of carbon dioxide (CO_2) produced per minute in four species of bean.

The results are shown in the table.

Species of bean	Volume of O_2 used in one minute (mm^3)	Volume of CO_2 produced in one minute (mm^3)
A	0.063	0.050
B	0.038	0.027
C	0.061	0.065
D	0.072	0.052

- (i) Describe how the values for the volume of CO_2 produced in one minute would be measured and calculated using a respirometer.

Remove the KOH solution from the respirometer and replace it with an equal volume of water, so CO_2 is no longer absorbed.
Record the distance moved by the coloured liquid over 1 minute, both with KOH and with water.
Change caused by CO_2 production = distance moved with KOH present minus distance moved without KOH.
Calculate the volume using $\pi r^2 l$, where r is the radius of the capillary tube.

[3]

- (ii) Calculate the RQ of **species A**, using the values in the table.

$$\begin{aligned} \text{RQ} &= \text{CO}_2 \text{ produced} \div \text{O}_2 \text{ used} \\ &= 0.050 \div 0.063 = 0.7937 \end{aligned}$$

RQ = 0.794 [1]

- (iii) The student thinks the results for one of the species are anomalous.

State which species has anomalous results **and** explain why the student thinks the results are anomalous.

Species C, because its RQ is greater than 1: $0.065 \div 0.061 = 1.07$.
The other species have RQs of approximately 0.7 to 0.8 (A = 0.79, B = 0.71, D = 0.72), so C does not fit the pattern.

[2]

- (b)* Plan an investigation to test the effect of temperature on the **rate of respiration** in germinating beans and obtain valid results.

You do **not** need to include details of how to set up or use a respirometer in your plan.

Use at least five temperatures within a suitable range, e.g. 10, 15, 20, 25, 30 and 35 °C (up to 40 °C), each set using a thermostatically controlled water bath, checked with a thermometer.

Use the same species and variety of bean, of the same age and stage of germination (e.g. soaked for 3 days), and the same mass of beans in each respirometer.

Let each respirometer equilibrate in the water bath for the same time (10 minutes) before each test, then measure the O_2 used over a set time (e.g. 10 minutes) and calculate the rate of respiration per unit mass ($\text{mm}^3 O_2 \text{ g}^{-1} \text{ min}^{-1}$).

Carry out 3 replicates at each temperature and calculate the mean rate, identifying any anomalies.

Use Spearman's rank to test for a correlation between temperature and rate, or an unpaired t-test to compare the mean rate at two temperatures.

Safety: KOH is corrosive (medium risk), so wear goggles and gloves; broken [6]

Extra answer space if required.

glass can cut (low risk), so use a broken glass bin; hot water can scald (low risk). Wash and cover any cut, rinse skin with water and seek first aid.

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.....

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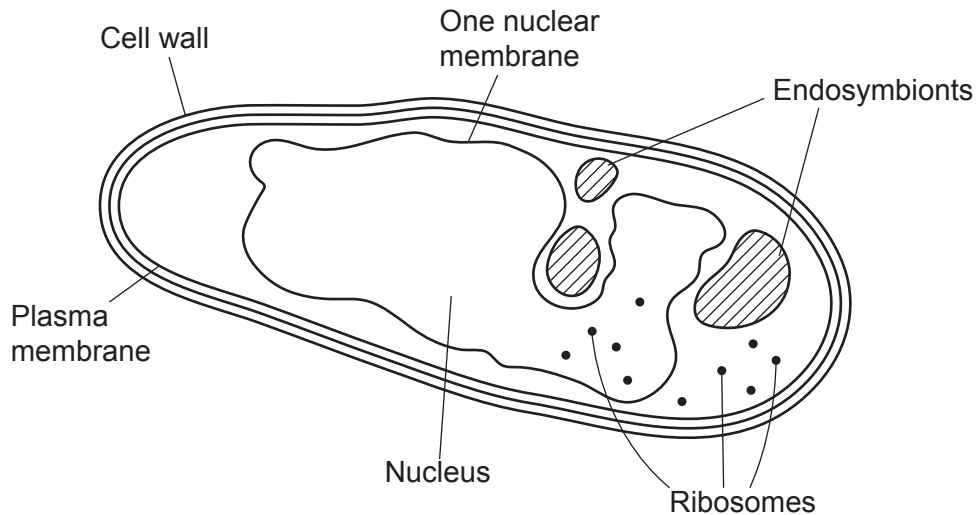
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- 3 *Parakaryon myojinensis* is a species of single-celled organism that was discovered in 2012.

P. myojinensis is unique among species that have been discovered because it has features of both eukaryotes and prokaryotes. It also has features that are neither eukaryotic nor prokaryotic.

A student uses a sharp pencil to draw a *P. myojinensis* cell from an electron micrograph.

A representation of the student's drawing of *P. myojinensis* is shown in the image.



Endosymbionts are organisms that can also be found in some eukaryotic and prokaryotic cells.

- (a) State **two** ways in which the student could improve their drawing.

1 *Do not use shading or hatching; draw clear, single, continuous lines.*

2 *Use ruled, horizontal label lines and add a scale bar or magnification.*

[2]

- (b) The student concludes that 'although it does not seem to have mitochondria, *P. myojinensis* is more similar to a eukaryote than a prokaryote'.

Discuss the evidence from the image that supports **and** does **not** support this conclusion.

..Supports: a nucleus is present, as in eukaryotes.....
 ..Does not support: eukaryotes have other membrane-bound organelles such as
 ..endoplasmic reticulum and Golgi apparatus, but none are present.....
 ..Eukaryotes do not have ribosomes in the nucleus, and they have a double
 ..membrane (nuclear envelope), but this cell has only one nuclear membrane.....
 [3]

- (c) Suggest what the absence of mitochondria indicates about the process of respiration in *P. myojinensis*.

..It may rely on anaerobic respiration, so all its ATP is produced in glycolysis in
 ..the cytoplasm.....
 [1]

- (d) The student is interested in whether the cell wall of *P. myojinensis* is made from peptidoglycan or a different substance.

State how to test for the presence of peptidoglycan in the cell wall of *P. myojinensis*.

..Use Gram staining: add crystal violet stain, then potassium iodide (Gram's
 ..iodine) solution.....
 ..Wash with ethanol to remove unbound crystal violet, then add a counterstain
 ..(safranin). A purple colour shows that peptidoglycan is present.....
 [2]

(e)* *Rhizobium* species are other examples of single-celled organisms.

Describe how to culture *Rhizobium*, in vitro.

Your answer should include steps that would reduce the risk of contamination of the culture.

Disinfect the workspace with Virkon solution and work aseptically next to a Bunsen flame, which creates a convection current away from the workspace. Select a pink root nodule (healthy) from a leguminous plant, cutting it off with a sterilised scalpel. Wash and sterilise the nodule surface, then crush it in a little sterile distilled water to make a *Rhizobium* suspension. Prepare an agar mixture containing mannitol (respiratory substrate), yeast extract (nitrogen source), magnesium sulfate (essential ions) and a pH buffer. Boil it, then cool and pour into a sterile Petri dish and allow it to set. Add 1 cm³ of *Rhizobium* suspension with a sterile pipette and spread it with a sterile glass spreader. Partially seal the Petri dish with tape to prevent entry of unwanted microorganisms, place it upside down in an incubator and incubate at 25 °C for several days (not above 30 °C, to avoid culturing pathogens). Risk assessment: cuts from scalpels (medium risk), so cut away from the body; skin irritation from plant material (low risk), so wear non-latex gloves.

[6]

Extra answer space if required.

4 Mutations in some genes are associated with an increased risk of developing cancer.

(a) The table lists three genes associated with cancer: *Myc*, *p53* and *Ras*.

Complete the table by putting a tick (✓) in the appropriate box or boxes on each line to show whether a gene has the feature.

Feature	<i>Myc</i>	<i>p53</i>	<i>Ras</i>
A mutation that causes a loss of function that can lead to cancer.		✓	
It is a proto-oncogene.	✓		✓

[1]

(b) The polymerase chain reaction (PCR) can be used to detect alleles that are associated with cancer.

(i) Complete the table to show the temperatures used in the three stages of a PCR cycle and the processes that occur at each temperature.

Temperature (°C)	Process
.....95.....	Separation of DNA strands
55	..annealing of primers to the template strands.....
.....72.....	DNA polymerase attaches nucleotides to the new strands

[2]

(ii) In theory, the number of DNA molecules can increase exponentially during PCR.

Calculate the theoretical maximum number of DNA molecules that could be present after 15 cycles of PCR, assuming that the process started with one DNA molecule.

Give your answer as a $\log_{10}$ value.

$$2^{15} = 32\,768$$

$$\log_{10} 32\,768 = 4.515$$

Number of DNA molecules = ...10^{4.515}..... [2]

- (iii) Suggest **one** reason why the theoretical maximum number of DNA molecules might **not** be achieved.

Reagents such as DNA nucleotides or primers may be used up before all cycles of PCR are complete.

[1]

- (iv) A version of PCR known as quantitative PCR (qPCR) is used to detect alleles that are associated with an increased risk of cancer.

qPCR involves:

- extracting mRNA (transcribed from the target allele) from a cell
- converting the mRNA to complementary DNA (cDNA)
- conducting PCR on the cDNA sample using *Taq* DNA polymerase, allele-specific primers and fluorescent probes.

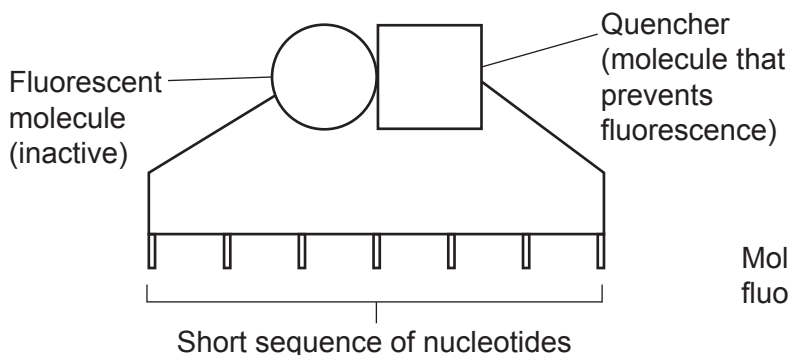
State the name of the enzyme that converts mRNA to cDNA.

reverse transcriptase

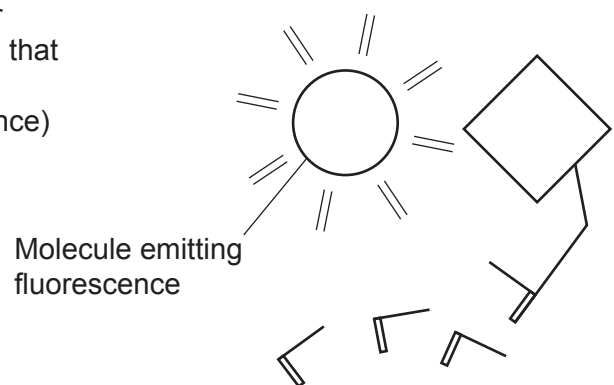
[1]

- (v) The diagram shows a fluorescent probe used in qPCR.

Before the probe has bound to cDNA



After the action of *Taq* DNA polymerase



Suggest how fluorescent probes can be used in qPCR to estimate the concentration of cDNA derived from a cancer-associated allele.

*The probe sequence is complementary to the target allele. Fluorescence is detected only when *Taq* polymerase synthesises a new strand, separating the quencher. Fluorescence is proportional to cDNA concentration, so fluorescence after a set number of cycles estimates the initial cDNA concentration.*

[2]

- (c) Electrophoresis is another technique that can be used to detect cancer-associated alleles.

CYP17 is a gene that has been linked to some cancers.

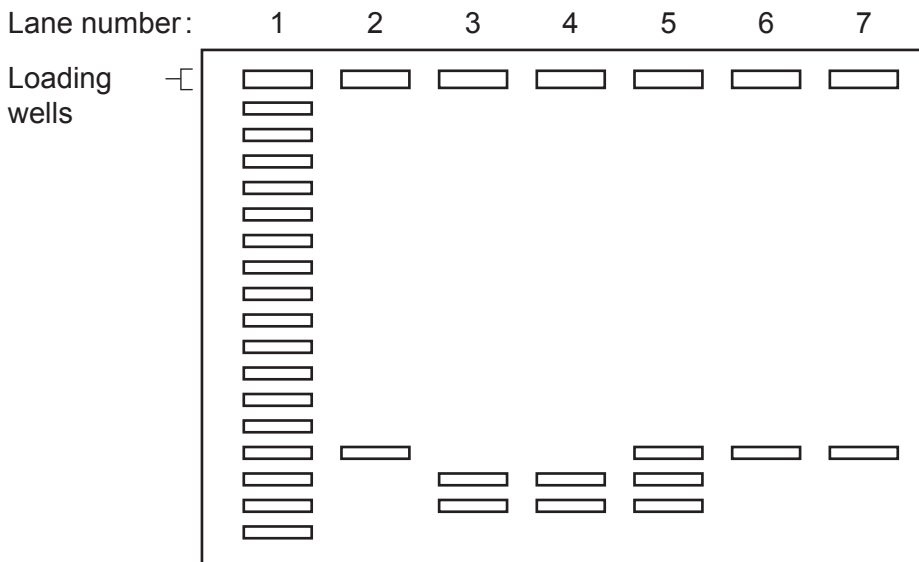
- The normal *CYP17* allele does not increase the risk of cancer.
- A mutant *CYP17* allele increases the risk of some cancers.

Restriction enzymes are used to produce fragments of the *CYP17* alleles for analysis in electrophoresis.

Restriction enzymes:

- produce one fragment when the normal *CYP17* allele is present
- produce two smaller fragments when the mutant *CYP17* allele is present.

The diagram shows the results of electrophoresis of DNA samples from 6 people. Lanes 2 to 7 in the diagram show only DNA fragments from *CYP17* alleles.



- (i) Samples containing DNA fragments are placed in the loading wells of the electrophoresis box before beginning electrophoresis.

State **one other** substance that should be added to the box before beginning electrophoresis **and** explain the purpose of this substance.

..Running..buffer..solution.....

..To maintain a constant pH and conduct the current, creating an electric field.....

..that allows the DNA fragments to move through the gel.....

.....

..... [2]

(ii) State **and** explain the purpose of the DNA fragments in lane 1 in the diagram.

It is a DNA ladder: marker DNA fragments of known length (size).

These are used to compare the distance travelled by the sample fragments, to

determine the size of the sample fragments.

..... [2]

(iii) State the lane numbers that indicate:

- a genotype that is homozygous for the normal *CYP17* allele
- a genotype that is homozygous for the mutant *CYP17* allele.
- a heterozygous genotype for the *CYP17* gene

Homozygous normal ..2, 6, 7.....

Homozygous mutant ..3, 4.....

Heterozygous ..5.....

[2]

- 5 Scientists analysed the effect of alcohol consumption by parents on the risk of a child being born with microcephaly.

Microcephaly is defined as a head circumference more than two standard deviations below the mean at a particular age.

The scientists obtained information on the number of units of alcohol consumed per week by both parents:

- during the three months before conception
- during the first three months of the mother's pregnancy.

The scientists compared the risk of the newborn baby having microcephaly when parents consumed:

- no alcohol
- more than 5 units of alcohol per week.

They performed statistical tests to test the hypothesis that there was a greater risk of microcephaly when parents consumed more than five units of alcohol per week.

The p values from the statistical tests are shown in the table.

	p value	
	Mother consuming >5 units per week	Father consuming >5 units per week
Before pregnancy	0.078	0.048
In the first three months of pregnancy	0.508	0.246

- (a) Describe the conclusions that can be made from the p values in the table.

Before pregnancy, there is a significantly greater risk of microcephaly when
fathers consume >5 units per week ($p = 0.048 < 0.05$), but no significant
increase in risk when mothers do ($p = 0.078 > 0.05$).
In the first three months of pregnancy, alcohol consumption has no significant
effect on the risk for either parent ($p > 0.05$). [2]

- (b) Suggest what additional information is needed to increase confidence in the conclusions.

A larger sample size, and how the information on alcohol use was obtained

(e.g. self-reported)

Whether confounding factors were controlled, e.g. smoking, drug use, age of

parents, maternal diet and folic acid intake

[2]

END OF QUESTION PAPER

[illegible]

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