2 Amylase is an enzyme that breaks down starch into maltose.

(a) A student investigated the breakdown of starch into maltose. The results are shown in **Fig. 2.1**.

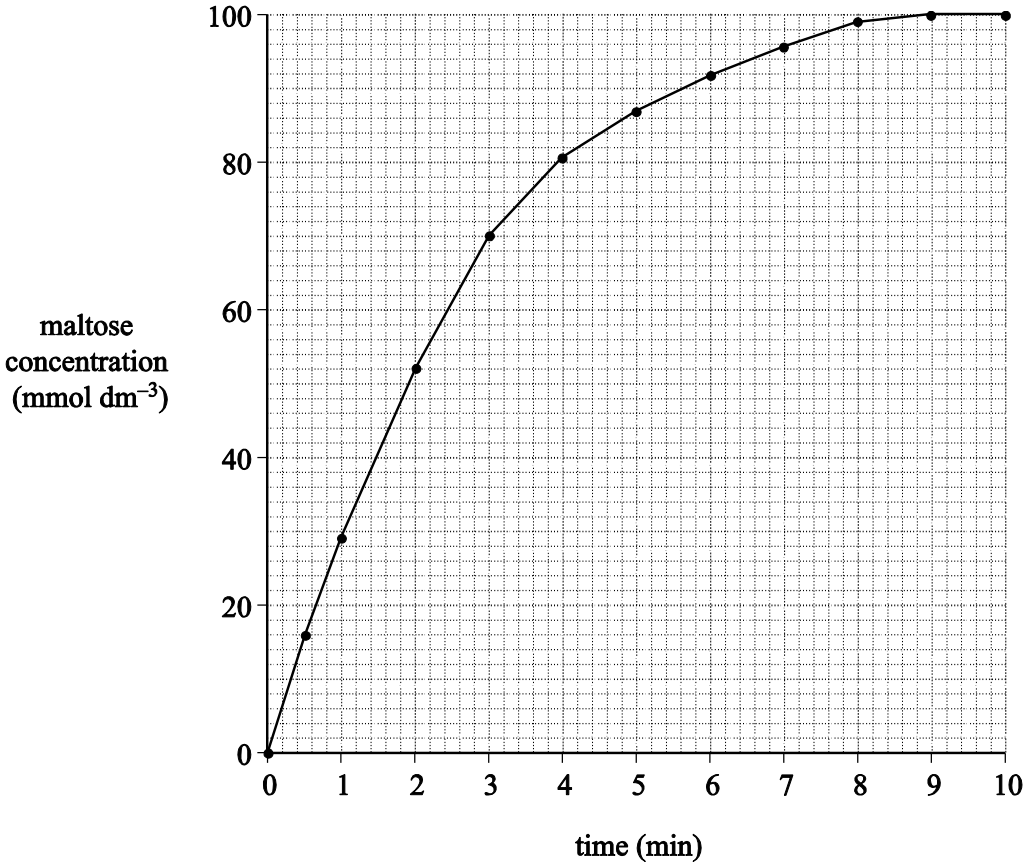


Fig. 2.1

(i) Calculate the rate of maltose production over the first 30 s.

Use appropriate units.

Answer…… **[2]**

(ii) How would this calculated rate differ from the 'true' initial rate of reaction?
Explain your answer.

……

……

……

……

…… **[3]**

(b) The student conducted a further investigation using the same enzyme and substrate.

- A range of substrate concentrations was used.
- The investigation was repeated in the presence of an inhibitor of amylase activity extracted from kidney beans.

Fig. 2.2 shows a sketch of the student's results.

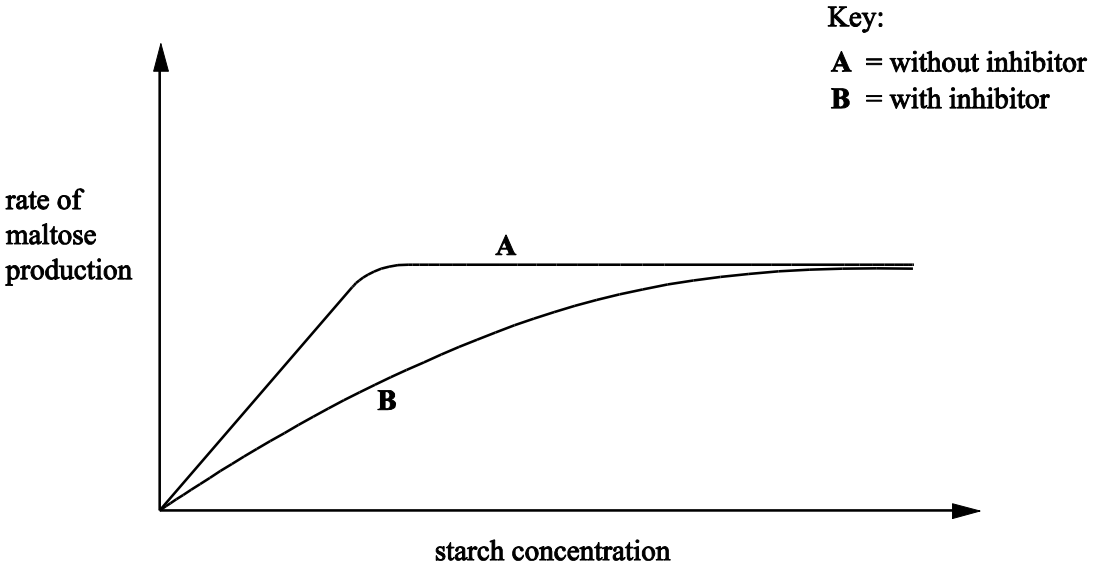


Fig. 2.2

(i) Explain the mechanism by which the extract from the kidney bean inhibited the amylase.

……

……

……

……

…… **[3]**

(ii) What evidence from the graph supports your answer to part **(i)**?

……

…… **[1]**

(c) The student then investigated the effect of pH on the activity of the amylase.

This was the method used,

- Tubes containing starch and amylase were set up in a range of pH buffer solutions.
- The same concentration of starch and amylase were used each time.
- A small sample of the solution was removed and tested for the presence of starch at 20 s intervals.
- The procedure was repeated three times and a mean was calculated for each pH.

The student presented the results in **Table 2.1**.

pH	4	5	6	7	8	9
Mean amylase activity (% of maximum)	27	68	96	100	50	29

Table 2.1

(i) Another student wanted to replicate the investigation.

Refine the method, by giving additional information, so that reproducible results would be obtained.

……...

……...

……...

…… **[3]**

(ii) Explain, with reference to bonding, why amylase activity is low at pH 4.

……...

……...

……...

……...

……...

…… **[4]**

(iii) The student concluded that the optimum pH for amylase was pH 7.

A teacher made the following statement:

*'The results in **Table 2.1** provide only weak support for the conclusion that the optimum pH for amylase is pH 7.0'*

Evaluate the statement **and** suggest an improvement to the student's procedure that would support the conclusion more strongly.

Evaluation

……...

……...

……...

Improvement

……...

……...

……...

[3]

(d) Amylase activity is increased in the presence of chloride ions.

State the name given to any inorganic ion that increases the activity of an enzyme.

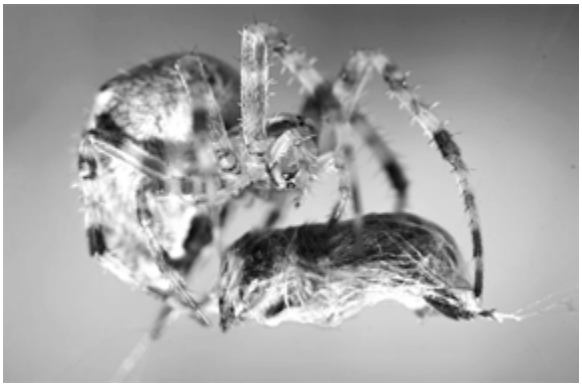
…… **[1]**

Question			Answer	Marks	Guidance
2	(a)	(i)	32 mmol dm^{-3} min^{-1}	2	**ALLOW** mmol dm^{-3} / min' or 'mmol dm^{-3} per , min / minute **ALLOW** 0.53 mmol dm^{-3} / s
		(ii)	(initial rate likely to be) greater *because* higher concentration of, substrate / amylose, molecules (at start) more chance of, substrate / AW, entering <u>active site</u>	3	**ALLOW** 'starch'
	(b)	(i)	*three from* competes (with substrate) / competitive enters / fits in / binds to / blocks, active site prevents substrate from entering active site (binds to active site) temporarily	3	
		(ii)	(at high substrate concentration) rate approaches rate in absence of inhibitor	1	**IGNORE** idea that increased substrate concentration overcomes the inhibition as answer must refer to evidence from the graph.
	(c)	(i)	*three from* specify volume of starch and amylase to be added to the tubes specify volume (in ml) of the solution that should be removed for testing stir before taking the sample test with iodine all carried out at same temperature	3	

Question			Answer	Marks	Guidance
		(ii)	*four from* ionic / hydrogen, bonds, disrupted / broken (by) high concentration of, hydrogen ions / H^+ tertiary structure / shape of active site, changed substrate no longer fits into active site (enzyme) denatured	4	**IGNORE** active site denatured.
		(iii)	*Evaluation, two from* idea that optimum could be anywhere between pH 6 and pH 8 only one value between pH 6 and pH 8 tested idea that shape of data implies optimum less than pH 7 *Improvement* repeat at more pH values between 6 and 8	3	
	(d)		cofactor	1	**IGNORE** coenzyme.
			Total	20	

8 Spiders inject a mixture of digestive enzymes into the body of their prey and feed on the products of this digestion.

The photograph shows a spider with its prey.



(a) State why enzymes are described as biological catalysts.

(1)

(b) One of the enzymes injected into the prey is called arazyme. Arazyme is a protease enzyme that can break down collagen.

(i) Compare and contrast the molecular structure of collagen and an enzyme such as arazyme.

(4)

(ii) The hydrolysis of collagen results in a decrease in pH. The indicator phenolphthalein is pink at pH 8 and becomes colourless at lower pH values.

Devise an investigation to determine the effect of arazyme concentration on the rate of hydrolysis of collagen.

(4)

DO NOT WRITE IN THIS AREA

DO NOT WRITE IN THIS AREA

DO NOT WRITE IN THIS AREA

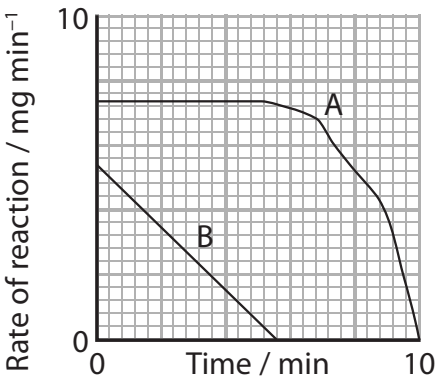
(c) The effect of substrate concentration on the rate of an enzyme reaction was investigated.

Two substrate solutions, A and B, were used. Solution A had a higher concentration than solution B. The optimum pH for the enzyme was pH 7.

The product lowered the pH of the solution.

The rate of reaction was determined at 1-minute intervals for 10 minutes.

The graph shows the results of this investigation.



It was concluded that the product reduces the activity of the enzyme.

Comment on the validity of this conclusion.

(4)

(Total for Question 8 = 13 marks)

TOTAL FOR PAPER = 80 MARKS

Question Number	Answer	Additional Guidance	Mark
8 (a)	An answer that makes reference to the following: • (proteins which) reduce activation energy of biological reactions (1)	ALLOW increase rate of biological reactions ALLOW references to {in cells / in living organisms}	**(1)**

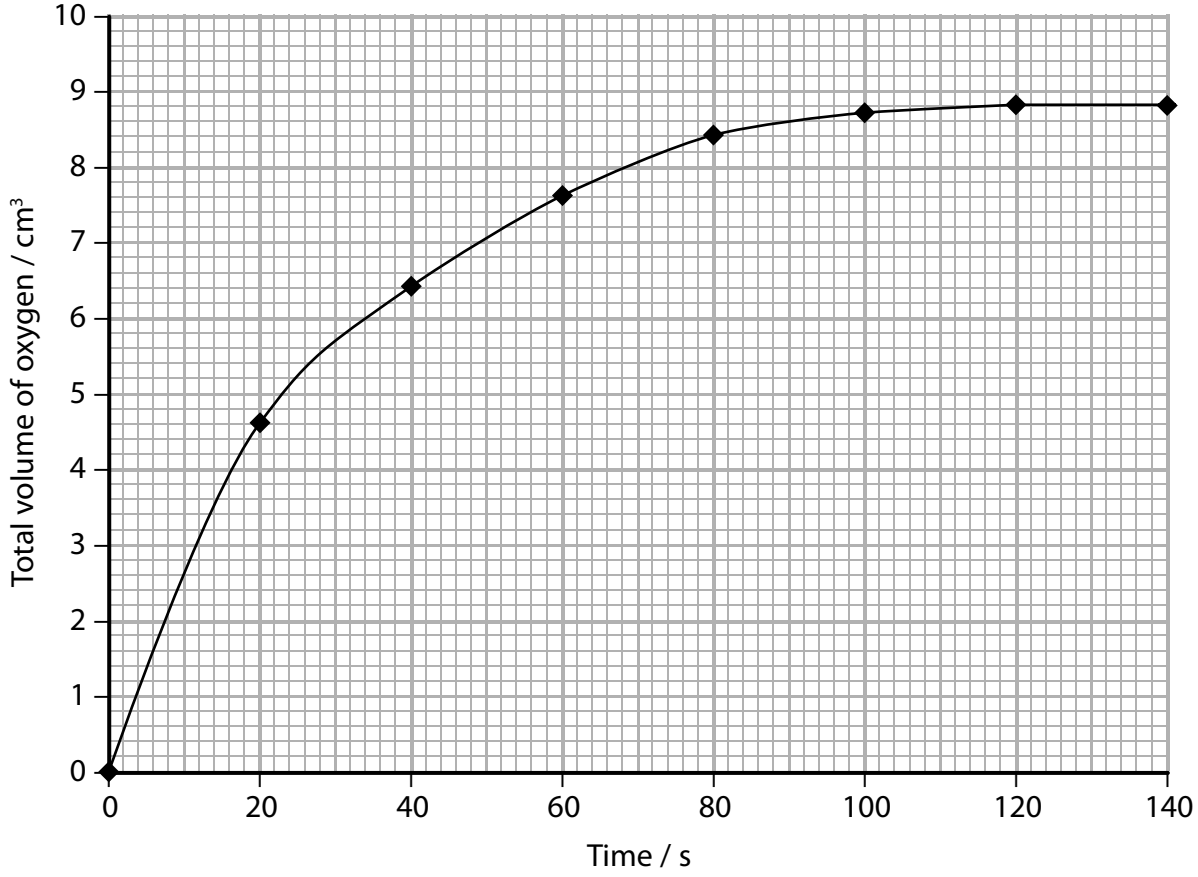
Question Number	Answer	Additional Guidance	Mark
8 (b)(i)	An answer that makes reference to four of the following: Similarities • both are chains of amino acids joined by peptide bonds (1) • (both have) bonds involved in holding molecule in its three dimensional shape (1) Differences • arazyme is folded into {compact / tertiary} structure whereas collagen has long (parallel) chains with cross links (1) • arazyme has an active site whereas collagen does not (1) • arazyme has some {hydrophilic groups / amino acids} on surface whereas collagen does not (1)	ALLOW a correct named bond ALLOW arazyme is a globular protein whereas collagen is a fibrous protein ALLOW collagen only has (hydrophobic groups / amino acids} on surface whereas arazyme does not	**(4)**

Question Number	Answer	Additional Guidance	Mark
8 (b)(ii)	An answer that makes reference to the following: • at least five different concentrations of arazyme solution (1) • a controlled variable (1) • measuring of the dependent variable (1) • time taken (for phenolphthalein) to decolourise (1) • {replicates / repeats} for each arazyme concentration to allow calculation of mean values (1)	ALLOW 0% arazyme e.g. volume of enzyme / volume of substrate / concentration of substrate / temperature / volume of indicator e.g. colorimeter / compare to colour standard	**(4)**

Question Number	Answer	Additional Guidance	Mark
8 (c)	An answer that makes reference to the following: • the conclusion is not valid (1) • as pH would reduce faster {at higher substrate concentrations / in graph A} (1) • (however) rate of reaction for A did not decrease immediately (1) • lower pH would result in the denaturation of the enzyme and reduction in the rate of reaction (1)	ALLOW the product does not reduce the activity of the enzyme ALLOW converse for solution B ALLOW more product is produced in solution A, but the rate of reaction decreases slower than in solution B ALLOW converse for solution B	**(4)**

10 Catalase is an enzyme present in many tissues of most living organisms, but can be found in high concentrations in liver cells. Its role is to break hydrogen peroxide down into oxygen and water. Hydrogen peroxide is produced by cells and is very harmful if it is not broken down.

A student carried out an investigation into the action of catalase. Some liver was chopped into small pieces, and added to hydrogen peroxide. The volume of oxygen gas produced was recorded and a graph was drawn.



(a) (i) Calculate the initial rate of reaction.

(3)

Answer .....

(ii) Analyse the graph to explain the change in the total volume of oxygen produced over the course of reaction.

(4)

(iii) Draw on the graph the line you would expect if the student repeated the investigation with the same concentration of hydrogen peroxide but with double the mass of liver.

(2)

(b) Describe how transcription is involved in the synthesis of an enzyme.

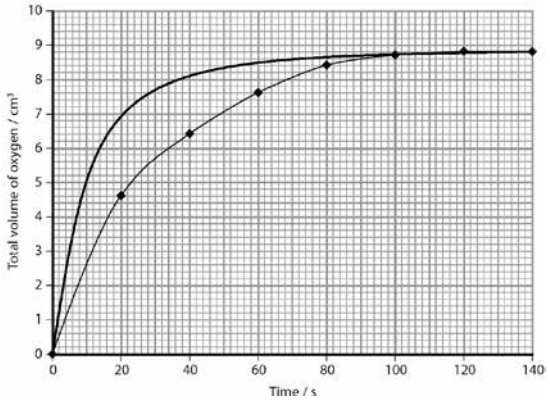
(4)

(Total for Question 10 = 13 marks)

TOTAL FOR PAPER = 80 MARKS

Question Number	Acceptable Answer	Additional guidance	Mark
10(a)(i)	suitable time interval chosen (in range 0 to 20 s, must be on straight line portion) (1) change calculated e.g. 4.6cm^3 ÷ 20 seconds (1) ans = 0.23 cm^3 s $^{-1}$ (1)	Allow full marks for correct answer, no working	**(3)**

Question Number	Acceptable Answer	Additional guidance	Mark
10(a)(ii)	An explanation that makes reference to the following: • initial rate of reaction {is fast / shows positive correlation} because {substrate / hydrogen peroxide} is not limiting (1) • therefore there are many {collisions between enzyme and substrate / enzyme substrate complexes} (1) • between {20s and 100s} the total volume produced slows because there is {less substrate / fewer collisions / fewer enzyme substrate complexes} (1) • no oxygen produced after 100s because reaction has stopped because {substrate used / substrate is limiting} (1)		**(4)**

Question Number	Acceptable Answer	Additional guidance	Mark
10(a)(iii)	Steeper initial gradient (1) Reaches a plateau faster (1)  		**(2)**

Question Number	Acceptable Answer	Additional guidance	Mark
10(b)	A description that makes reference to the following: • DNA {unzips / unwinds} and hydrogen bonds between complementary strands broken (1) • the {antisense / coding / template} strand used for mRNA synthesis (1) • RNA polymerase used to join RNA nucleotides (1) • complementary base pairing of A with U, not T (1)		**(4)**

(Total for Question 10 = 13 marks)

6 Enzymes control biochemical pathways.

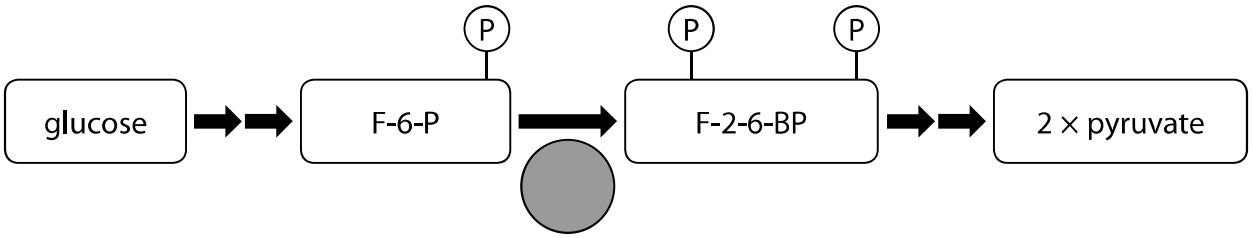
Phosphofructokinase is an enzyme involved in controlling the rate of glycolysis.

(a) State what is meant by the term enzyme.

(2)

(b) Phosphofructokinase is an enzyme that uses ATP to convert fructose-6-phosphate (F-6-P) into fructose-2,6-bisphosphate (F-2,6-BP).

The conversion of F-6-P by this enzyme is a rate-determining step in glycolysis. This is shown in the diagram.



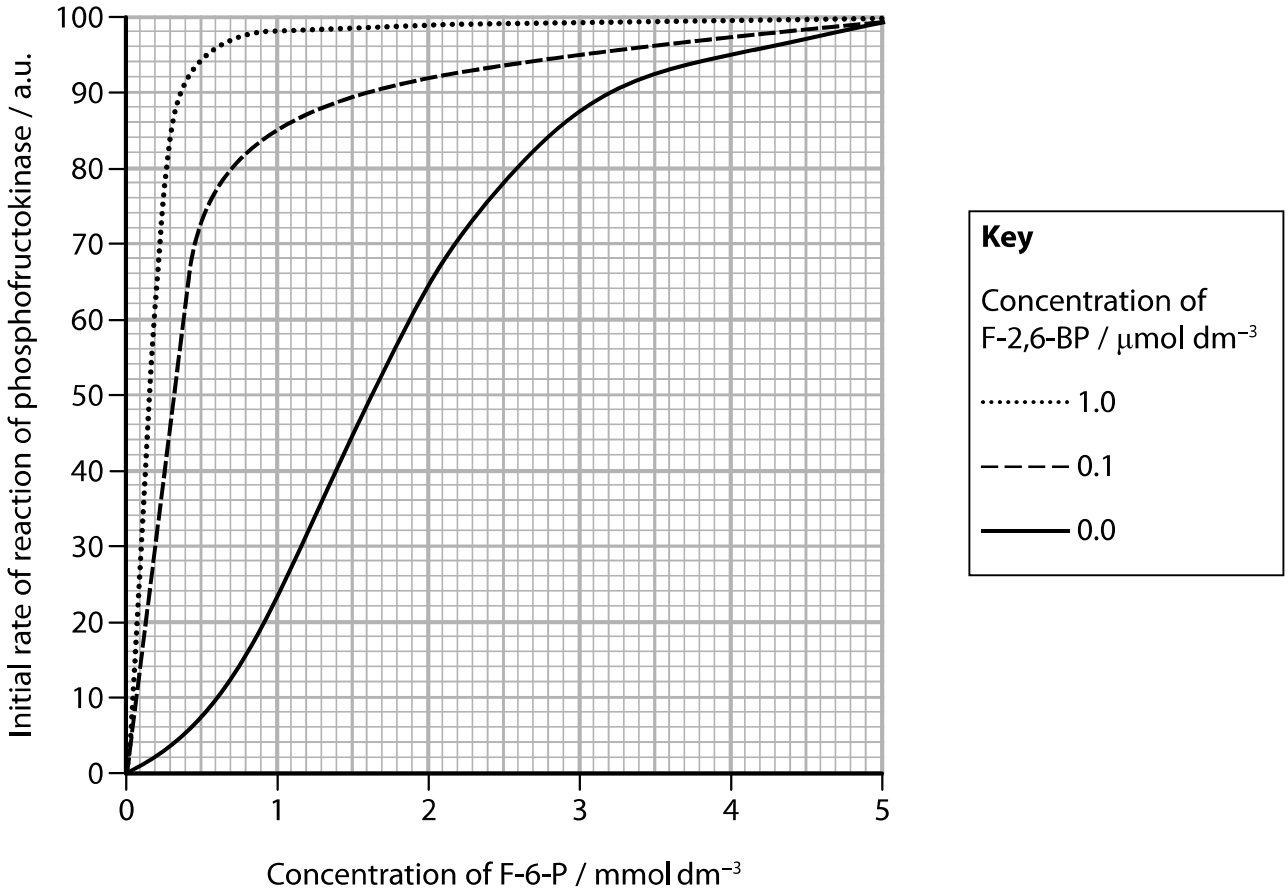
(i) Explain why ATP is required for this reaction.

(3)

(ii) The effect of substrate concentration on the initial rate of reaction of phosphofructokinase was investigated.

This investigation was repeated with the addition of two concentrations of F-2,6-BP.

The graph shows the results of this investigation.



Comment on the effects of F-6-P and F-2,6-BP concentrations on the rate of glycolysis.

(3)

(c) Glycolysis is inhibited by acidic conditions.

Devise an investigation to determine the effect of acidic conditions on the initial rate of reaction of phosphofructokinase.

(4)

(Total for Question 6 = 12 marks)

Question Number	Answer	Additional guidance	Mark
5(b)(iii)	An answer the makes reference to two of the following: • do not have allele conferring ability to respond to gravity / only have alleles that confer a lack of response to gravity (1) • (so) do not produce {IAA / auxin} (1) • (therefore) lack of stimulation of cell elongation on side of stem facing downwards (1)	ALLOW cells present in the stem fail to detect gravity ALLOW there is no auxin present	**(2)**

Question Number	Answer	Additional guidance	Mark
6(a)	An answer the makes reference to two of the following: • {biological / protein} catalyst (1) • lowers the activation energy (for a reaction) (1) • increasing the rate of reaction (1)		**(2)**

Question Number	Answer	Additional guidance	Mark
6(b)(i)	An explanation that makes reference the following: • hydrolysis of ATP (1) • provides energy for the reaction (1) • provides phosphate group for phosphorylation of F-6-P (1)	ALLOW as the reaction requires energy ALLOW provides {phosphate / Pi} that is added to F-6-P	**(3)**

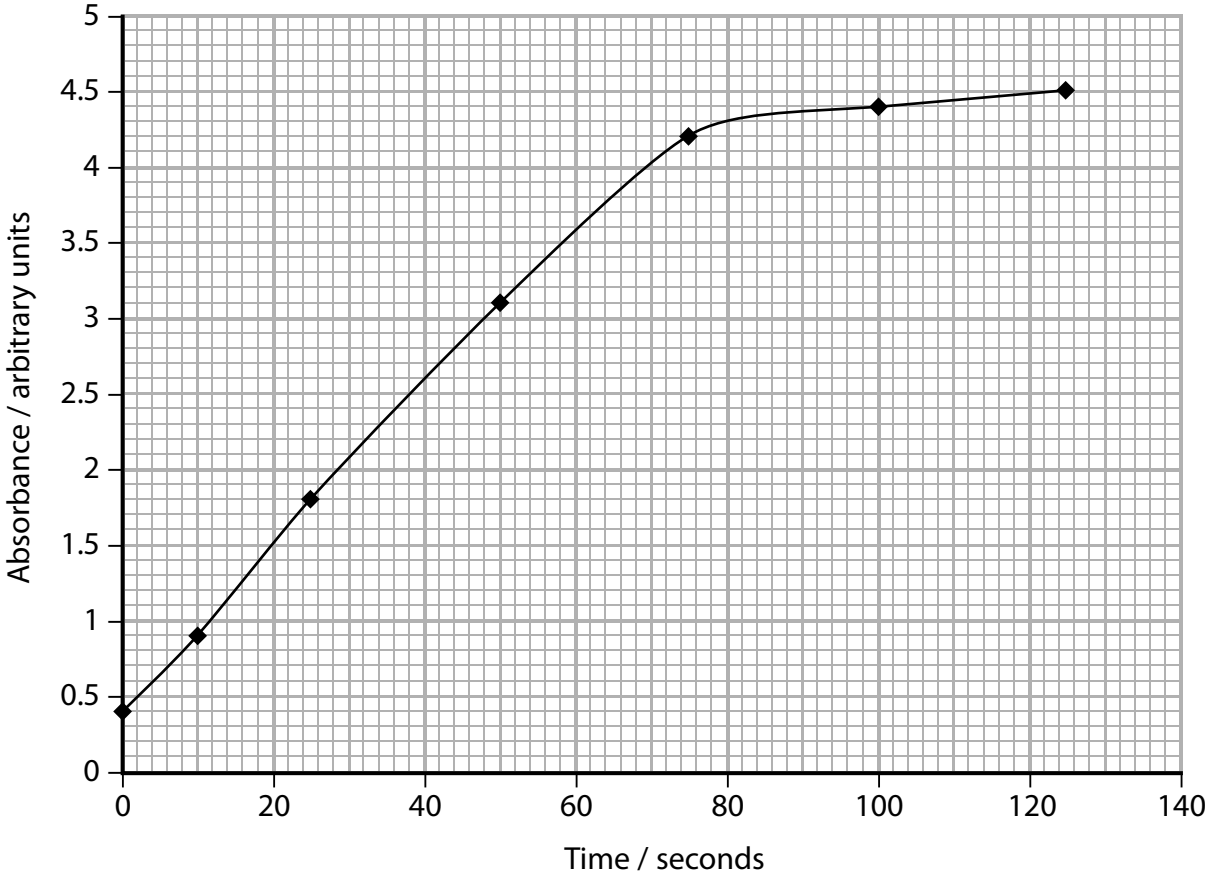
Question Number	**Answer**	**Additional guidance**	**Mark**
6(b)(ii)	An answer that makes reference to three of the following: • as concentration of { F-6-P / F-2,6-BP } increases so does the (initial) rate of reaction of the phosphofructokinase (1) • an increasing in the concentration of { F-6-P / F-2,6-BP } will increase the rate of glycolysis (1) • up to a maximum (rate) (1) • increasing the concentration of F-2,6-BP reduces the concentration of F-6-P required to achieve the maximum rate of glycolysis (1)	ALLOW 'enzyme' for 'phosphofructokinase' ALLOW F-2,6-BP provides positive feedback to the enzyme activity	**(3)**

Question Number	Answer	Additional guidance	Mark
6(c)	An answer the makes reference to four of the following: • use pH buffers at a range of pH values below 7 (1) • provide an excess of ATP (1) • (use) F-6-P at an appropriate concentration (1) • suitable variable controlled (1) • measure quantity of F-2,6-BP produced per unit time (1)	e.g. 2 mmol dm^{-3} (values between 1 and 2.5 mmol dm^{-3}) e.g. {enzyme / phosphofructokinase } concentration / temperature ALLOW measure change in concentration of F-2,6-BP / phosphate incorporated	**(4)**

8 L-dopa forms a colourless solution in water. Dopa oxidase is an enzyme that converts L-dopa into dopachrome, which is red.

A colorimeter can be used to study this reaction. As the red colour appears, the amount of light absorbed by the solution increases.

(a) The graph shows the course of a reaction in which there was an enzyme concentration of 20 (arbitrary units) of reaction mixture.



Calculate the initial rate of reaction for this concentration of enzyme.

(3)

Answer .....

(b) In another study, a student used this procedure with a range of enzyme concentrations. The results are shown in the table below.

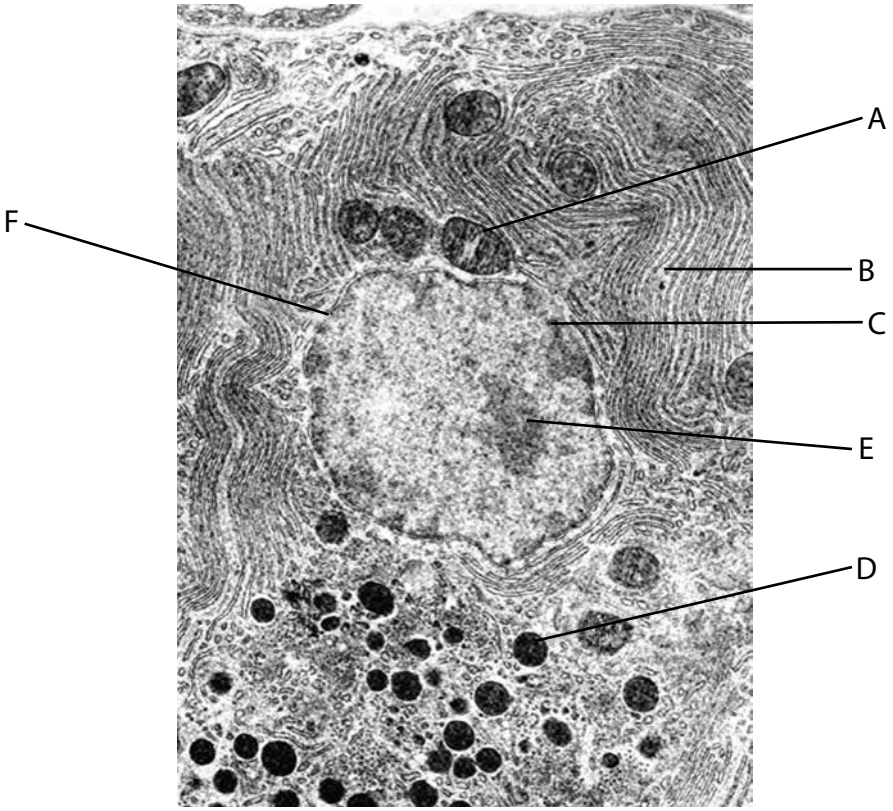
Enzyme concentration / arbitrary units	Initial rate of reaction / absorbance s^{-1}
0	0.0
10	2.5
30	6.1
50	9.0
70	11.0
90	11.0

Explain the effect of enzyme concentration on the initial rate of this reaction.

(3)

(c) Enzymes are made, modified, stored and used in cells.

The photograph shows an electron micrograph of a cell from the pancreas.



(i) Which structure in the photograph uses enzymes in respiration?

(1)

- ☒ **A**
- ☒ **B**
- ☒ **C**
- ☒ **D**

(ii) Which structure in the photograph is the nucleolus?

(1)

- ☒ **B**
- ☒ **C**
- ☒ **E**
- ☒ **F**

(Total for Question 8 = 8 marks)

Question Number	Acceptable Answer	Additional guidance	Mark
8(a)	• suitable time interval chosen (in range 0 to 70 s, must be on straight line portion) (1) • absorbance change calculated (1) • ans. 0.053 au s^{-1} (or as appropriate for part of graph chosen) (1)	Example: at 0 s abs = 0.4, at 60 s abs = 3.6 (1) so change is 3.6 − 0.4 = 3.2 (1) over 60 s, make rate 3.2 ÷ 60 = 0.053 au s^{-1} (1)	**(3)**

Question Number	Acceptable Answer	Additional guidance	Mark
8(b)	An explanation that makes reference to three of the following: • as enzyme concentration increases the rate of reaction increases and levels off (1) • because number of active sites of the enzyme molecules is increasing (1) • because enzyme concentration is the limiting factor (1) • it levels off because the substrate concentration is limiting (1)		**(3)**

Question Number	Acceptable Answer	Additional guidance	Mark
8(c)(i)	A		**(1)**

Question Number	Acceptable Answer	Additional guidance	Mark
8(c)(ii)	E		**(1)**

(Total for Question 8 = 8 marks)

Do not write outside the box

0 7 . 1 Explain how the active site of an enzyme causes a high rate of reaction.

[3 marks]

Do not write outside the box

The action of the enzyme catalase is shown below.

$$\text{hydrogen peroxide} \xrightarrow{\text{catalase}} \text{water} + \text{oxygen}$$

A student investigated the effect of hydrogen peroxide concentration on the rate of this reaction. He used catalase from potato tissue.

The student:

- put five potato chips in a flask
- added 20 cm^3 of 0.5 mol dm^{-3} hydrogen peroxide solution to the flask
- measured the time in seconds for production of 10 cm^3 of oxygen gas
- repeated this procedure with four different concentrations of hydrogen peroxide solution.

His results are shown in **Table 5**.

Table 5

Hydrogen peroxide concentration / mol dm^{-3}	**Time for production of 10 cm^3 of oxygen gas / seconds**	**Rate of reaction / arbitrary units**
0.5	18	
1.0	10	
1.5	7	
2.0	6	
2.5	6	

0 7 . 2 Other than those stated, give **one** factor the student would have controlled in his investigation.

[1 mark]

Do not write outside the box

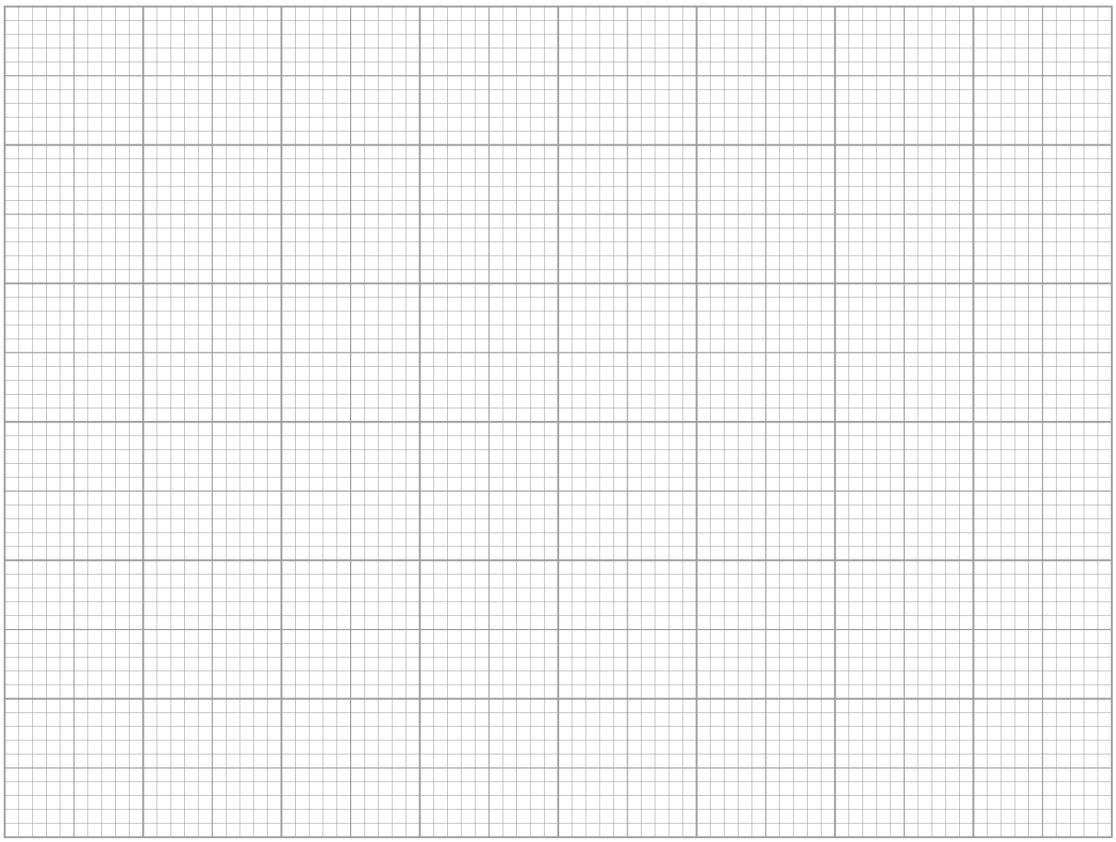
0 7 . 3 The student gave the maximum rate of reaction a value of 1.0 arbitrary units.

Complete **Table 5** by calculating the rate of reaction in arbitrary units at each hydrogen peroxide concentration. Record the rates using an appropriate number of significant figures.

[2 marks]

0 7 . 4 Plot a suitable graph of your processed data shown in **Table 5**.

[3 marks]



0 7 . 5 Suggest a change the student could make to his procedure so that 10 cm^3 of oxygen would be produced in less than 6 seconds.

[1 mark]

10

Question	Marking Guidance	Mark	Comments
07.1	1. Lowers activation energy; 2. Induced fit **causes** active site (of enzyme) to change shape; 3. (So) enzyme-substrate complex **causes** bonds to form/break;	3	3. Accept: description, of induced fit 3. Accept: enzyme-substrate complex causes stress/strain on bonds.
07.2	Size/dimensions /mass/variety of potato **OR** Temperature (of solution/flask) **OR** pH (of solution);	1	Accept : weight of potato Ignore : amount of potato Ignore concentration/ volume of catalase
07.3	0.33, 0.60, 0.86, 1.0, 1.0 = 2 marks;; $\frac{6}{time}$ 2 significant figures If answer incorrect accept for 1 mark, Correct values but incorrect number of significant figures **OR** 1.0 written on row for hydrogen peroxide 2.0/2.5 in Table 5 **OR** Answers showing correct division, eg 0.3, 0.6, 0.9 **OR** Answers showing correct significant figures using incorrect calculation (÷18) 1.0, 0.56, 0.39, 0.33, 0.33	2	
07.4	1. Hydrogen peroxide concentration on x axis **and**	3	1. Graph should cover

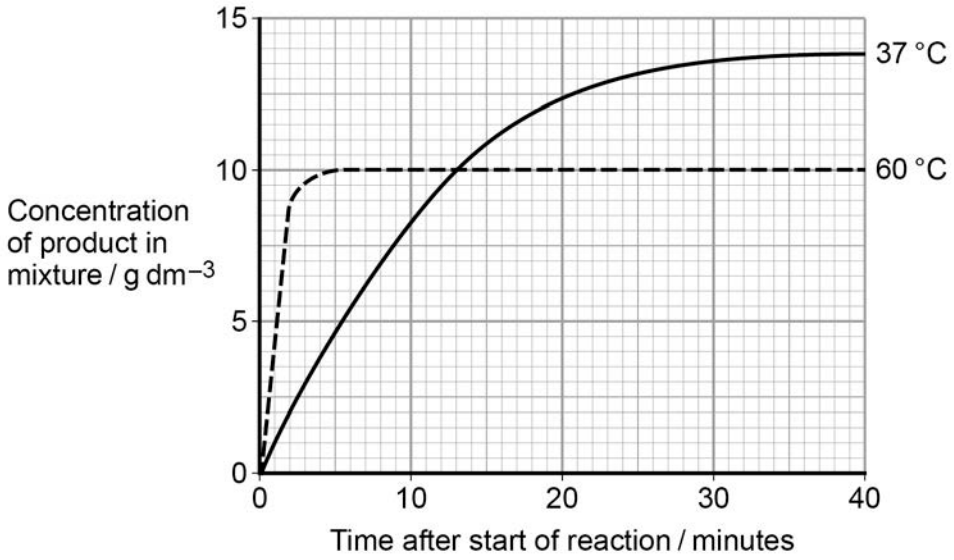
	rate of reaction on Y axis, linear number sequence **and** appropriate scale; 2. Correct units /mol dm^{-3} **and** /arbitrary units/au; 3. All co-ordinates plotted accurately **with** point-to-point or smooth curve;		half or more of the grid; eg reject if Y axis covers only three big squares 2. Accept brackets instead of solidus 3. Accept accurate plotting of co-ordinates given in 07.3 3. Reject: bar chart 3. Reject: if ruled straight line of best fit 3. Accept: if x axis starts at 0.5 3. Accept: if line is extended to (0,0) Plot coordinates must be processed data, hydrogen peroxide vs time = 0
07.5	Cut up/use discs/homogenise/increase surface area (of potato chips) **OR** Use bigger chips **OR** Increase temperature **OR** Change pH;	1	Reject answer if the temperature is above 40°C Ignore: more/increase heat
TOTAL		**10**	

Answer **all** questions in the spaces provided.

1 A technician investigated the effect of temperature on the rate of an enzyme-controlled reaction. At each temperature, he started the reaction using the same concentration of substrate.

Figure 1 shows his results.

Figure 1



0 1 . 1 Give **two** other factors the technician would have controlled.

[1 mark]

1 _____

2 _____

0 1 . 2 Draw a tangent on each curve to find the initial rates of reaction.
Use these values to calculate the ratio of the initial rates of reaction at 60 °C : 37 °C.
Show your working.

[2 marks]

Ratio = _____ :1

0 1 . 3 Explain the difference in the initial rate of reaction at 60 °C and 37 °C.

[2 marks]

0 1 . 4 Explain the difference in the rates of reaction at 60 °C and 37 °C between 20 and 40 minutes.

[4 marks]

[Extra space]

Question	Marking Guidance	Mark	Comments
01.1	Any **two** of the following; Concentration of enzyme Volume of substrate solution pH	1	Allow same concentration of substrate
01.2	Ratio between 4:1 and 5:1;;	2	Initial rates incorrect but correctly used = 1 mark
01.3	At 60 °C: 1. More kinetic energy; 2. More E–S complexes formed;	2	Allow converse for 37 °C
01.4	Different times: 1. Higher temperature / 60 °C causes denaturation of all of enzyme; 2. Reaction stops (sooner) because shape of active site changed; Different concentrations of product (at 60 °C) 3. Substrate still available (when enzyme denatured); 4. But not converted to product;	4	Accept converse for 37 °C 2. Reject if active site on substrate