Enzymes

Model Answers 1

Level	A Level
Subject	Biology
Exam Board	OCR
Module	Foundations in Biology
Topic	Enzymes
Booklet	Model Answers 1

Time allowed: **66 minutes**

Score: **/49**

Percentage: **/100**

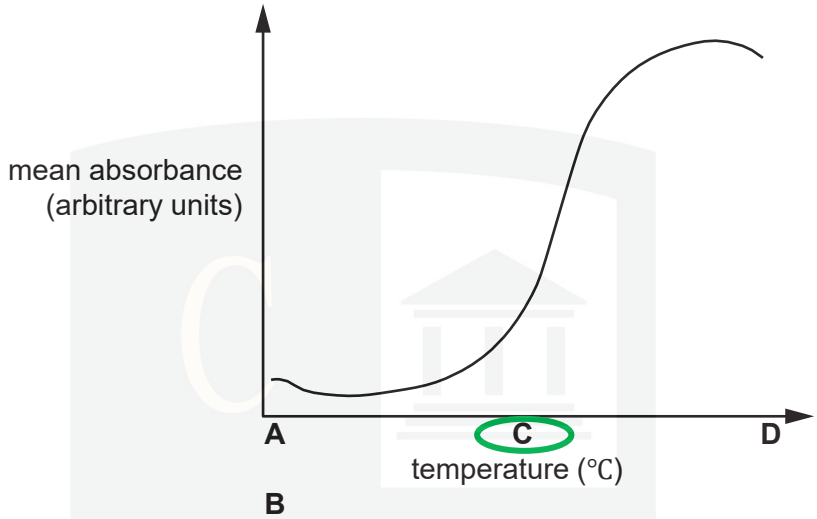
Grade Boundaries:

A*	A	B	C	D	E
>69%	56%	50%	42%	34%	26%

Question 1

Swiss chard is a leafy green vegetable related to spinach. Some varieties have yellow stalks that have vacuoles containing yellow betaxanthin pigments.

The graph below shows the effect of temperature on the release of these pigments recorded as mean absorbance, when measured with a colorimeter.



It was deduced that the betaxanthins were released from the vacuole due to the denaturing of proteins in the tonoplast (vacuolar membrane).

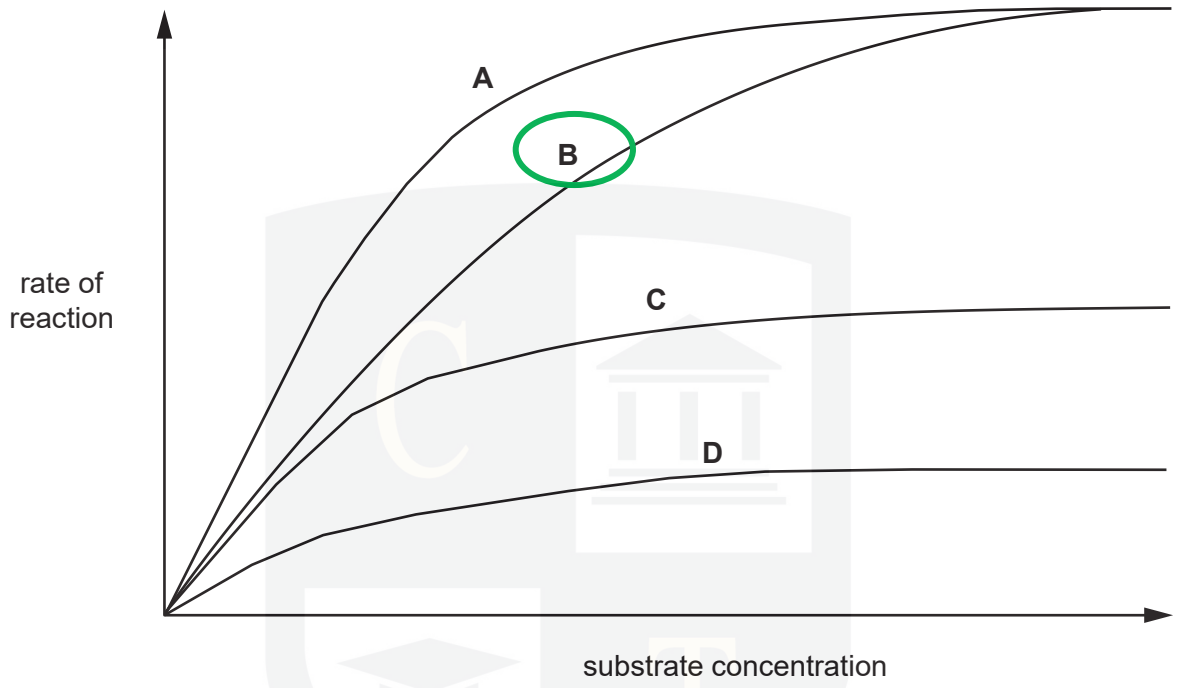
Which letter, **A** to **D**, shows the temperature at which the proteins denature?

[1]

As the proteins in the membrane denature they leave large gaps allowing more of the betaxanthins to leave the vacuole causing the absorbance to dramatically increase

Question 2

The following graph shows the rate of reaction of an enzyme in different substrate concentrations.



Which letter, **A** to **D**, shows the rate of reaction with a fixed quantity of competitive inhibitor?

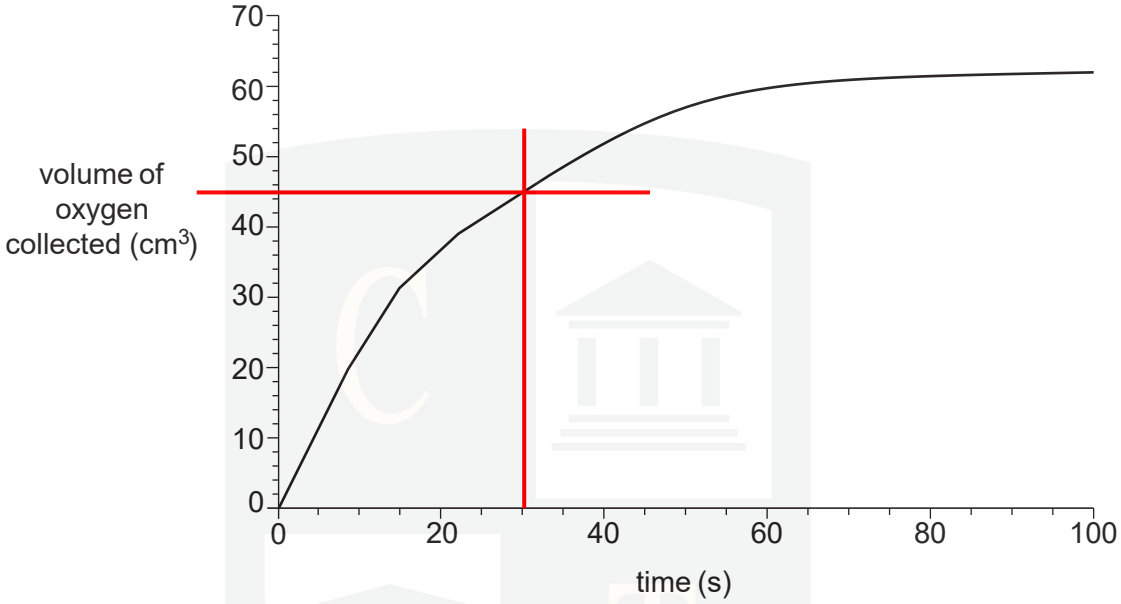
[1]

Competitive inhibitors have a similar shape to the substrate so they can bind to the active site. This will lead to a slower rate of product formation but eventually it will become saturated at the same rate but at a higher substrate concentration

Question 3

Celery contains the enzyme catalase, which breaks down hydrogen peroxide into oxygen and water.

A student added liquidised celery to a solution of hydrogen peroxide and collected the oxygen given off by the reaction. The results are shown in the graph below.



Which of the following shows the rate of reaction **at** 30s?

A 0.85 cm^3 s^{-1}

B 1.00 cm^3 s^{-1}

C 1.15 cm^3 s^{-1}

D 1.50 cm^3 s^{-1}

[1]

At 30 seconds the volume of oxygen given off is 45 cm^3

Per second just divide this by 30 which is 1.5cm^3 s^{-1}

Question 4

The protease enzyme bromelain can be extracted from pineapples. A student investigated the effect of changing the concentration of the enzyme and measured the time taken to break down the protein gelatine.

(a) State **three** variables that the student would need to control in order to make the results of this investigation **valid**. **[3]**

- Temperature
- pH
- Concentration of protein
- Volume of protein
- Volume of enzyme / bromelain
- Same source of enzyme

These are all control variables and affect the validity of the results.

The term to avoid here is 'amount'.

'Amount of enzyme', 'amount of protein'

Instead use volume and concentration. In this case 'amount scores nothing but volume and concentration scores 3!

(b) The data from the student's experiment is shown in Table 26.

Concentration of bromelain (%)	Rate of protein digestion (s^{-1})	Standard deviation
0.010	0.0037	0.00014
0.025	0.0090	0.00034
0.050	0.0155	0.00260
0.075	0.0184	0.00371
0.100	0.0198	0.00340

Table 26

(i) Describe how the rate of reaction was calculated. **[1]**

1 ÷ time taken

Units are per sec or better still is (s^{-1})

(ii) Explain what the standard deviation shows in Table 26. **[2]**

- Standard deviation shows the spread of the data around the mean
- All the values in the table have a small SD
- There is little variation so there is a high repeatability
- As concentration increases the SD increases
- As concentration increases the repeatability decreases

If you plot the standard deviations on a graph you get error bars. If the error bars overlap then you cannot claim that the results are valid

(c) Fig. 26 shows the results plotted on a graph with the standard deviations as error bars.

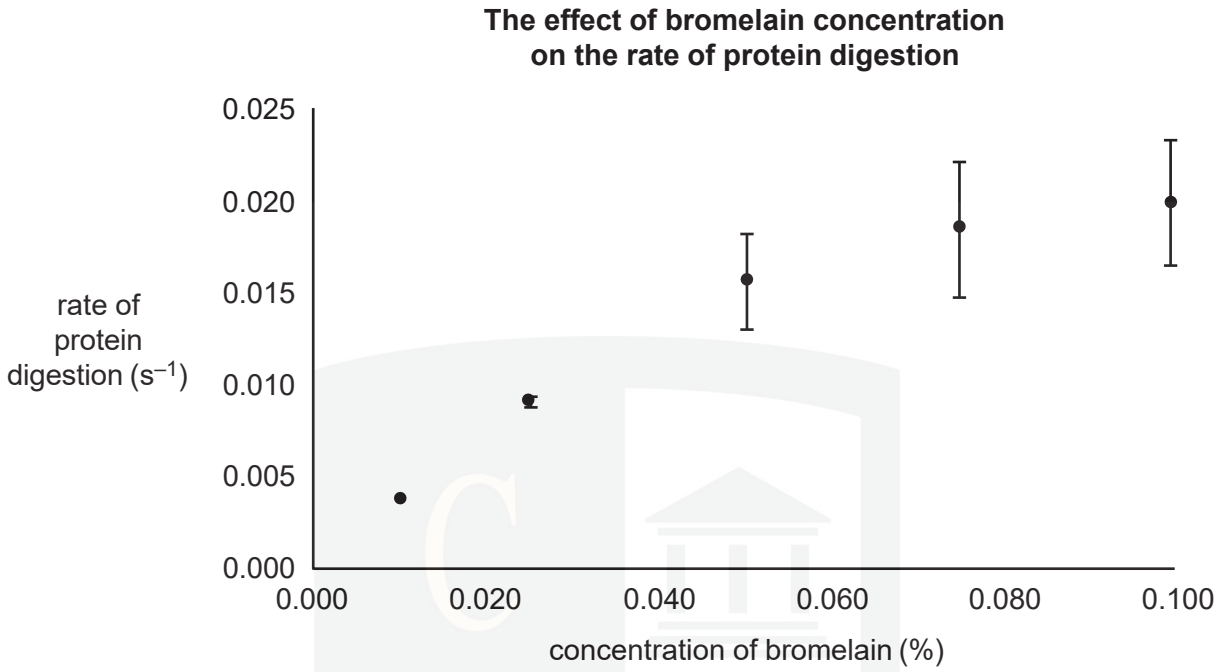


Fig. 26

Explain the pattern shown in the data using Table 26 and Fig. 26.

- As concentration of bromelain increases the rate of digestion increases
- As concentration of bromelain increases the substrate concentration stays the same
- The rate eventually levels off when there are lots of active sites remaining empty
- Substrate concentration is a limiting factor
- At higher concentrations the error bars start to overlap so any difference in data might be uncertain

As substrate concentration increases, the number of active sites stays the same and eventually they are forming E/S complexes as fast as they can convert them to product. At this point the enzymes are saturated and this also known as the Vmax.

[Total: 9]

Question 5

(a) Fungi produce enzymes to digest complex food substances. Amylase is an enzyme that catalyses the conversion of starch to maltose.

- A sample of the fungus *Amanita citrina* was placed on agar in a petri dish.
- The agar contained starch.
- The dish was incubated until the thread-like hyphae had grown a few centimetres.
- Iodine solution was then poured onto the surface of the agar.

A diagram representing the results is shown in Fig. 4.

Fig. 4

(i) To which genus does this fungus belong? **[1]**

Amantina

In the binomial system the genus is the first word and always has a capital letter, the species is next and has a lower case letter

(ii) The region of yellow staining shown in Fig. 4 includes part of the agar where the fungus had not yet grown.

What does this pattern indicate about the action of the fungal enzymes? **[1]**

- Starch has been digested where the hyphae have not yet reached
- Enzymes must have been secreted and released by the fungus then moved away

This is typical of saprophytic or saprotrophic feeding. Enzymes are secreted by the fungus which digest large molecules outside the hyphae to smaller, soluble molecules which can then be absorbed

(b) Lipase is an enzyme that catalyses the breakdown of lipids.

An investigation was carried out to see the effect of temperature on the activity of a lipase.

- 5 cm^3 of an alkaline solution of lipid was poured into a test tube.
- The test tube was placed into a water bath maintained at 20 °C and left to equilibrate.
- A few drops of an indicator were added to the wells of a white spotting tile. The indicator is pink above pH values of 8.3 and turns colourless at pH values below 8.3.
- Once the lipid solution had equilibrated, 1 cm^3 of 0.5% lipase solution at the same temperature was then added to the test tube.
- For five minutes, at 30 second intervals, the solution was stirred and a few drops were removed from the test tube and placed in a well on the white spotting tile.
- The time was recorded when the solution and indicator did not remain pink.
- The procedure was repeated four more times at 20 °C and then again at a further six temperatures.

The results are shown in Table 4.1 below.

Temperature (°C)	**Time when solution did not remain pink**				
	Replicate 1	**Replicate 2**	**Replicate 3**	**Replicate 4**	**Replicate 5**
20	210	270	240	300	270
25	90	120	210	180	120
30	60	60	90	90	60
35	60	60	60	90	60
40	210	120	210	180	210
45	240	300	300	–	270
50	–	–	–	–	–

Table 4.1

(i) Why is pH **not** a controlled variable in this investigation? **[1]**

The change in pH is the dependent variable or the one which is being measured

When lipids are hydrolysed they breakdown to produce fatty acids, hence the drop in pH

(ii) Identify **one** variable that has been controlled in this procedure. **[1]**

Volume of lipid solution or volume of enzyme or concentration of enzyme solution

or temperature or time intervals

Remember control variables make it valid

(iii) Identify **one** variable, other than pH, that has **not** been controlled in this procedure. **[1]**

Concentration of lipid or volume / number of drops of indicator solution / volume of

sample removed

(iv) The procedure required the solution to be stirred and then drops of solution to be placed on a white spotting tile.

Suggest why this procedure was followed rather than simply adding indicator to the test tube, stirring the solution and looking for the colour change in the test tube. **[1]**

Placing a small volume on a white tile allows you to better judge the colour change

or the addition of indicator solution may affect the enzyme involved

Judging the colour change by eye is subjective and depends upon judgement

(v) What can be concluded from the results in Table 4.1 about the optimum temperature for lipase activity? **[1]**

The optimum is between 30 and 35° C

Remember units here and you must state the range

(vi) Describe **two different** ways in which the procedure could be modified to obtain a more accurate value for the optimum temperature for lipase activity.

- Repeat the experiment using narrower temperature intervals **[4]**
- In the 30 to 35° C region
- Take samples more often
- For example every 15° C
- Use a colorimeter to measure
- Colour change would not be subjective
- Use a pH meter to measure change in pH

(c)* There are two models for the mechanism of enzyme action. Outline how changes in temperature can affect these mechanisms of lipase action. **[6]**

- Increasing temperature gives the molecules more kinetic energy
- This gives more successful collisions
- More enzyme / substrate complexes form
- Enzymes have an optimum temperature
- Above this the hydrogen bonds that hold the tertiary structure are affected
- The 3D shape changes and the active site is distorted
- The change in shape prevents the substrate from binding
- The effect of high temperature is known as denaturing
- This is irreversible

The two models for the mechanism of enzyme action must be included in your answer. Both the above, traditional model and 'induced fit' should be described. Remember the induced fit involves the substrate binding by creating a few weak bonds which then allows the enzyme and substrate to form stronger bonds and in doing so changes the 3D shape of the enzyme which, in turn, puts a strain on the bonds in the substrate which are broken

[Total: 17]

Question 6

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**.

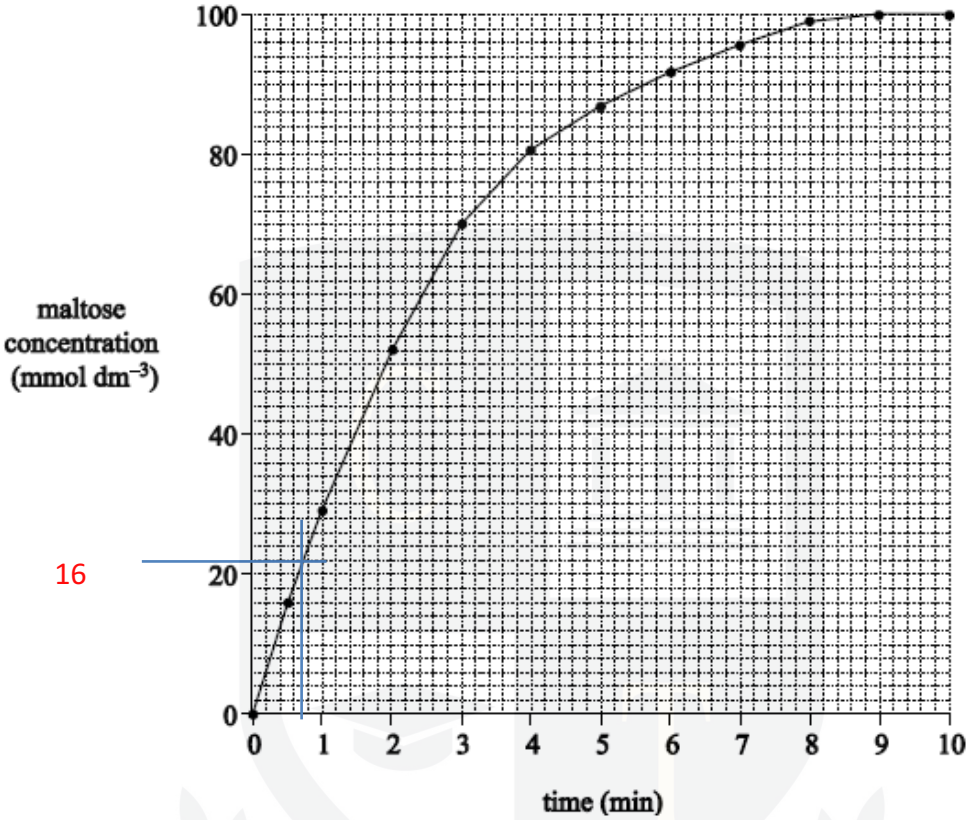


Fig. 2.1

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

Use appropriate units. **[2]**

- 32
- mmol dm^{-3} min^{-1}

16 mmol in 30 seconds so 32 mmol in one minute. Use appropriate units is also requested

(ii) How would this calculated rate differ from the 'true' initial rate of reaction? Explain your answer. **[3]**

- The initial rate will be faster
- There is more substrate present at the start
- More chance of enzyme / substrate collisions

The initial rate is always used to compare the effects of changing variables on the rate of enzyme reactions. This is when the enzyme is saturated and the rate is not affected by lack of substrate. Any later in the reaction and it will begin to be affected by lack of substrate.

(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.

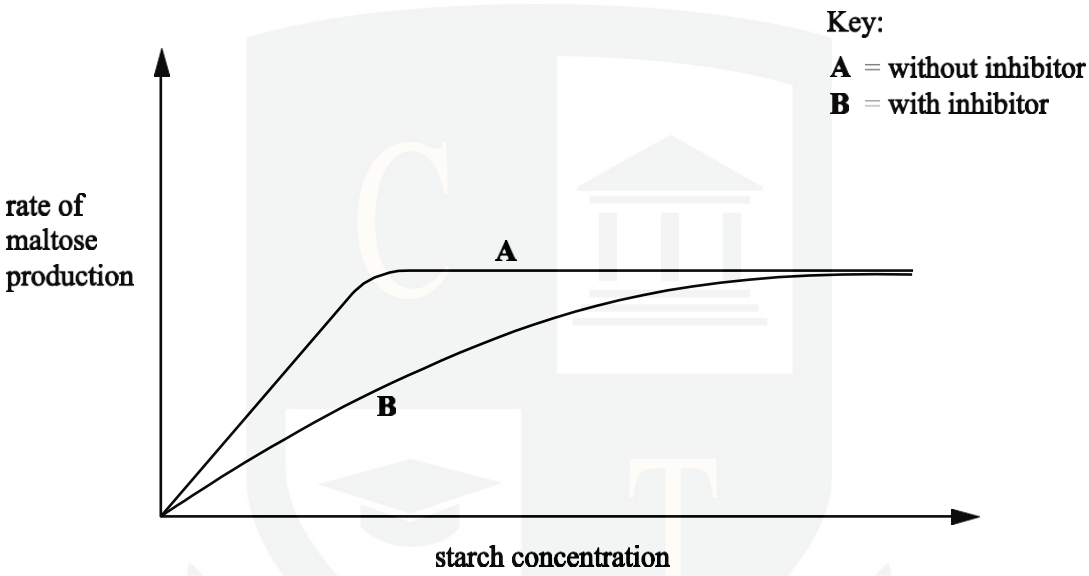


Fig. 2.2

(i) Explain the mechanism by which the extract from the kidney bean inhibited the amylase. **[3]**

- Competitive inhibition
- The inhibitor has a similar shape to the substrate so combines with the active site
- The inhibitor blocks the active site and prevents the substrate binding the effect is temporary

(ii) What evidence from the graph supports your answer to part **(i)**? **[1]**

- At high starch concentrations the rate approaches the rate without the inhibitor

When there is more substrate, the frequency of collisions with the normal substrate is increased.

Non competitive inhibitors combine with the allosteric site and distort the shape of the active site

(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]**

- Use the same volume of starch and amylase
- Carry out at the same temperature
- Remove the same volume of the sample
- Test for starch with iodine instead of pH

Remember never to use the dreaded word……..'Amount'

It's volume or concentration, never 'amount!'

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

- Hydrogen or ionic bonds are broken
- At low pHs there is a high concentration of hydrogen ions
- This changes the 3D or tertiary structure of the enzyme
- Substrate no longer fits the active site
- The enzyme is denatured

This type of question is there to score marks. Don't rush it, take it step by step, state the obvious and follow points through to completion. And don't forget they are denatured! This point is so obvious many students forget to include it!!

(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 **[3]**

- The optimum pH could be anywhere between 6 and 8
- Only one value was tested between 6 and 8
- The data suggests that the optimum pH is lower than 7

Improvement

- Repeat the experiment using more intervals between 6 and 8

(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]**

cofactor

[Total: 20]