Cambridge Assessment International Education

Skills Pack

Investigating the effect of copper sulfate concentration on catalase activity using paper discs

Cambridge International AS & A Level Biology 9700

Version 0.1

Cambridge Pathway

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Introduction

This pack will help you to develop your learners' experimental skills as defined by assessment objective 3 (AO3 Experimental skills and investigations) in the course syllabus.

Important note
Our *Skills Packs* have been written by **classroom teachers** to help you deliver topics and skills that can be challenging. Use these materials to supplement your teaching and engage your learners. You can also use them to help you create lesson plans for other experiments.

This content is designed to give you and your learners the chance to explore practical skills. It is not intended as specific practice for Paper 3 (Advanced Practical Skills) or Paper 5 (Planning, Analysis and Evaluation).

This is one of a range of *Skills Packs* and each pack is based on one experiment. The packs can be used in any order to suit your teaching sequence.

The structure is as follows:

Briefing lesson (1 hour*)
This lesson reinforces the key concepts, relevant skills, knowledge and understanding linked to the experiment.

↓

Planning lesson (1 hour*)
This lesson focuses on planning an experiment.
It ends with a demonstration video of the experiment.

↓

Lab lesson (1 hour*)
This lesson focuses on carrying out the experiment including the collection and recording of observations, measurements and estimates.

↓

Debriefing lesson (1 hour*)
This lesson focuses on the analysis and interpretation of data.
This includes making conclusions, evaluating methods and the quality of data and how improvements could be made.

** the timings are a guide only; you may need to adapt the lessons to suit your circumstances.*

In this pack, you will find lesson plans, worksheets and teacher resource sheets.

Experiment: Investigating the effect of copper sulfate concentration on catalase activity using paper discs

This *Skills Pack* focuses on an investigation into the effect of copper sulfate concentration on catalase activity using paper discs.

This investigation shows how varying the concentration of an inhibitor solution affects the rate of an enzyme-catalysed reaction. The results of this investigation can be used to calculate the value of the Michaelis-Menten constant (K_m) and determine what type of inhibitor is involved: competitive or non-competitive.

This experiment has links to the following syllabus content (2025–2027 syllabus):

- **3.2.2.** explain that the maximum rate of reaction (Vmax) is used to derive the Michaelis–Menten constant (K_m), which is used to compare the affinity of different enzymes for their substrates.
- **3.2.3.** explain the effects of reversible inhibitors, both competitive and non-competitive, on enzyme activity.

The experiment covers the following experimental skills, as listed in **AO3: Experimental skills and investigations:**

- plan experiments and investigations
- collect, record and present observations, measurements and estimates.
- analyse and interpret data to reach conclusions.
- evaluate methods and quality of data and suggest improvements.

Prior knowledge

Knowledge from the following syllabus topics is useful for this experiment.

- **3.1.2.** explain the mode of action of enzymes in terms of an active site, enzyme–substrate complex, lowering of activation energy and enzyme specificity, including the lock-and-key hypothesis and the induced-fit hypothesis.
- **3.1.3.** investigate the progress of enzyme-catalysed reactions by measuring rates of formation of products using catalase and rates of disappearance of substrate using amylase.

Briefing lesson: Slowing enzymes down

Resources	• Worksheets A and B • Teacher instructions • Adhesive tape or putty • Marker pens • Poster paper • Modelling clay • Sticky notes • String
Learning objectives	By the end of the lesson: • **all** learners should be able to outline the effect of inhibitors on the rate of an enzyme-catalysed reaction. • **most** learners should be able to describe the effect of competitive and non-competitive inhibitors on the rate of an enzyme-catalysed reaction. • **some** learners will be able to explain the effect of competitive and non-competitive inhibitors on the rate of an enzyme-catalysed reaction.

Timings	Activity
15 minutes	**Starter/Introduction** Engage learners with the starter activity shown in the **Teacher Instructions**. This is called 'what's the question,' and can be set as an individual task or as a paired pursuit. Follow up with learners by engaging them in a 'think, pair, share' activity with their partner to consider why particular factors influence enzyme activity. Encourage discussion by pointing out that maintaining these factors at nearly constant levels is important in the human body. Examples include temperature and pH. In the discussion that follows, ensure that learners have a good understanding of how the terms 'kinetic energy,' 'initial rate' and 'denature' relate to this concept.
40 minutes	**Main lesson** Inform learners that in the upcoming practical lesson, they will investigate the effect of changing copper sulfate concentration on catalase activity. Divide the class board into groups of 2-3. Provide each group with **Worksheet A**. These questions build learners' confidence in using key terms in the correct context. After 10 minutes, pair learners at random and ask them to read each other's work and discuss points on which they disagree, to promote a common class understanding. Provide **Worksheet B** to learners. This challenges learners to collaborate in small groups to prepare a poster to illustrate the different types of enzyme inhibition: competitive and non-competitive. Some guidance (such as keeping text to a minimum and filling available space) will be necessary. Some could add a three-dimensional aspect to their posters by including models of enzymes, enzyme-substrate complexes, inhibitors and enzyme-inhibitor complexes in the form of modelling clay. As you circulate around the room, ask learners to describe and explain how their posters illustrate the activity of an enzyme and formatively assess their understanding. Questions to ask include, 'why is this described as complementary binding?,' 'how does your model show that the enzyme is specific?' and 'what happens during the period of time that the enzyme–substrate complex exists?' Learners' models can be retained for use in subsequent lessons as a visual aid.

	At the end of the activity, host a 'marketplace' activity in which one member of each group stands by their poster and offers an explanation to other groups as they move around the room. Alternatively, learners could be asked to stick their work on the wall or hang it from a 'washing line' to display to others.
5 minutes	**Plenary** Host a short activity in which learners are challenged to compete with others to compare and contrast a series of terms relevant to this lesson. An example could be 'competitive inhibitor, non-competitive inhibitor, substrate.' All these molecules can bind to enzymes; however, only one of them attaches at a site other than the active site (the non-competitive inhibitor). or Encourage students to construct Venn diagrams to summarise the key features of inhibitors. For example, students could draw a circle labelled 'competitive inhibitors' overlapping with another circle labelled 'non-competitive inhibitors. Properties that these inhibitors have in common (e.g. both reduce the ability for an enzyme to bind to its substrate, both reduce the rate of reaction) can be listed in the overlapping area. Properties that are unique (e.g. effect on K_m, how they achieve their inhibition in terms of molecular binding) can be listed separately.

Planning lesson: Initial rates of reaction

Resources	• Worksheets C and D • 5 % (5 vol.) hydrogen peroxide (H_2O_2), 50 cm3, in labelled beaker • cutting board • eye protection • filter paper • hole punch • knife • large beaker • large conical flask • marker pen or labels • pair of forceps • paper towels • piece of black card • stick of celery • test tube rack • test tubes • timer
Learning objectives	By the end of the lesson: • **all** learners will be able to outline how to determine the initial rate of an enzyme-catalysed reaction by measuring the rate of formation of products. • **most** learners will be able to describe some of the problems associated with determining the initial rate of an enzyme-catalysed reaction by measuring the rate of formation of products. • **some** learners will be able to suggest how to overcome some of the problems encountered when the initial rate of an enzyme-catalysed reaction is determined by measuring the rate of formation of products.

Timings	Activity
20 minutes	**Starter/Introduction** Ask learners to gather around the teacher's table. Remind learners that they are going to plan an experiment to investigate the effect of copper sulfate concentration on catalase activity. An appropriate method to do this is required. Show learners an eye-catching demonstration. When some freshly-cut cubes of celery stem are added to hydrogen peroxide in a conical flask, bubbles of oxygen form to produce a foam. As the demonstration proceeds, explain that the celery tissue contains an enzyme called catalase (be sure to distinguish this word from 'catalyst'). Explain how this protein breaks down hydrogen peroxide, a toxic by-product of cellular metabolism, to water and oxygen. You could conclude by asking learners to suggest how this reaction can be represented in the form of a word equation and why, without enzymes, metabolic reactions would not take place fast enough to sustain life. Be sure to use the terms 'substrate' and 'products' during this discussion. Discuss with learners how they could use the basis of this demonstration to investigate the rate of the reaction catalysed by catalase. Elicit from them that the rate of oxygen release is one possible approach. Help them to understand the basis of method that will be used – determining the time taken for the disc to begin rising from the bottom of the substrate solution in the test tube.

	With this demonstration fresh in their mind, host a discussion focusing on the question: “why would a cell, or a biotechnologist, want to slow down the activity of an enzyme-catalysed reaction?” Elicit an understanding of why enzyme inhibitors are important in many contexts and remind learners that they will explore this concept in their investigation. It will be helpful for learners to have sentence stems to use when evaluating a practical experiment. For example, provide the statement ‘an investigation into the effect of X and Y, while keeping A, B and C the same’. Provide a writing frame to help learners write a conclusion and evaluation for this activity. This should have a series of model sentences but with key words removed, which the learners should complete. Share **Worksheet C** with learners. Challenge them to spend 5-10 minutes to prepare a brief outline of the basis of this investigation, which uses a novel method to determine the initial rate of reaction. They should use as many of the provided key words as possible.
30 minutes	**Main lesson** Instruct learners to engage in an ‘ideas hothouse’ to consider the information that can be obtained from a graph showing the change in the initial rate of reaction as the concentration of substrate increases. They are encouraged to work together in pairs to describe and explain an appropriate graph drawn on the class board. After one to two minutes of discussion, the pairs are to join into groups of four and then eight to discuss this further and come up with an agreed list of points. Host a class discussion to prepare a final list of statements on the board. Highlight the difference between the key command words, ‘describe’ and ‘explain’. Ensure that the descriptions outline in words what is shown by the graph without giving reasons, whereas explanations use biological knowledge and understanding to give reasons for the shape of the graph. Hand out **Worksheet D**, which provides learners with a series of prompts regarding the planning of this investigation. Provide 10 minutes for learners to complete the exercise, and then challenge them to compare their choices with a neighbour and decide if any of their choices have changed considering their discussions. Share with learners the answers to Worksheet D and discuss the method.
10 minutes	**Plenary** Ask learners to line along the wall of the classroom/ outside the classroom in the corridor. Assuming that there are an equal or greater number of learners than the number of steps in the method, walk past each learner in the line and as you do, each learner should, call out the step of the method or a safety precaution. Learners should be instructed to keep Worksheet D safe and bring it to the upcoming *Lab lesson*.

Lab lesson: Slowing enzymes down

Resources

- 10 cm^3 measuring cylinder x1
- 100 cm^3 beakers x2
- 1 cm^3 syringes x3
- 10 cm^3 syringes x2
- 250 cm^3 beaker of distilled water
- 500 cm^3 celery extract (labelled in a beaker) as a source of catalase
- Substrate solutions:
  - 1 % (1 vol.) hydrogen peroxide (H_2O_2), 50 cm3, in labelled beaker
  - 2 % (2 vol.) hydrogen peroxide (H_2O_2), 50 cm3, in labelled beaker
  - 3 % (3 vol.) hydrogen peroxide (H_2O_2), 50 cm3, in labelled beaker
  - 4 % (4 vol.) hydrogen peroxide (H_2O_2), 50 cm3, in labelled beaker
  - 5 % (5 vol.) hydrogen peroxide (H_2O_2), 50 cm3, in labelled beaker
- Inhibitor solutions:
  - 50 cm3 1.0 $moldm^{-3}$ copper sulfate (CuSO4) solution
  - 50 cm3 0.1 $moldm^{-3}$ copper sulfate (CuSO4) solution
- test tube rack
- test tubes
- eye protection
- filter paper
- hole punch
- large beaker
- marker pen or labels
- pair of forceps
- paper towels
- piece of black card
- timer

Learning objectives

By the end of the lesson:

- **all** learners will be able to investigate the effect of copper sulfate concentration on the initial rate of an enzyme-catalysed reaction by measuring the rate of formation of products.
- **most** learners will be able to explain why specific steps are taken during an investigation into the effect of copper sulfate concentration on the initial rate of an enzyme-catalysed reaction by measuring the rate of formation of products.
- **some** learners will be able to make suggestions to improve an investigation into the effect of copper sulfate concentration on the initial rate of an enzyme-catalysed reaction by measuring the rate of formation of products.

Timings	Activity
5 minutes	**Starter/Introduction** Check that learners have brought with them Worksheets D and E and reiterate some of the key messages from the previous lesson regarding best practice in this activity.
45 minutes	**Main lesson** Explain that as they conduct the investigation, they need to write down three problems they encounter and how they overcame them and to consider what kind of graph they should plot of their data. **Safety** Always circulate the classroom during the experiment so that you can make sure that your learners are safe and that the data they are collecting is accurate.

10 minutes	**Plenary** Learners will be at different stages of the practical activity towards the end of the lesson, with some likely to need the full hour to finish. Provide graph paper and elicit that as the data is continuous, they should draw a line graph. Ask if the points should be joined by a series of straight lines, a smooth curve, or a line/curve of best fit and why. Some learners may need to undertake this section of the task for homework.

Teacher notes

Watch the video showing the investigation into the effect of changing copper sulfate concentration on catalase activity using paper discs (teacher version) and read these notes.

Each group will require:

- 10 cm^3 measuring cylinder x1
- 100 cm^3 beakers x2
- 1 cm^3 syringes x3
- 10 cm^3 syringes x2
- 250 cm^3 beaker of distilled water
- 500 cm^3 celery extract (labelled in a beaker) as a source of catalase
- Substrate solutions:
  - 1 % (1 vol.) hydrogen peroxide (H_2O_2), 50 cm^3, in labelled beaker
  - 2 % (2 vol.) hydrogen peroxide (H_2O_2), 50 cm^3, in labelled beaker
  - 3 % (3 vol.) hydrogen peroxide (H_2O_2), 50 cm^3, in labelled beaker
  - 4 % (4 vol.) hydrogen peroxide (H_2O_2), 50 cm^3, in labelled beaker
  - 5 % (5 vol.) hydrogen peroxide (H_2O_2), 50 cm^3, in labelled beaker
- Inhibitor solutions:
  - 50 cm^3 1.0 $moldm^{-3}$ copper sulfate ($CuSO_4$) solution
  - 50 cm^3 0.1 $moldm^{-3}$ copper sulfate (CuSO4) solution
- test tube rack
- test tubes
- eye protection
- filter paper
- hole punch
- large beaker
- marker pen or labels
- pair of forceps
- paper towels
- piece of black card
- timer

Safety

The information in the table below is a summary of the key points you should consider before undertaking this experiment with your learners.

It is your responsibility to carry out an appropriate risk assessment for this experiment.

Substance	Hazard	First aid
hydrogen peroxide (5 % / vol.)	oxidising agent, corrosive	If hydrogen peroxide encounters the skin, immediately rinse the affected area with running water. Use cool or lukewarm water and gently wash the skin to remove any residual hydrogen peroxide. Avoid using hot water, as it can cause further skin irritation. If hydrogen peroxide splashes into the eyes, flush the eyes with copious amounts of

Substance	Hazard	First aid
		clean, lukewarm water for at least 15 minutes. Hold the affected eye open and allow the water to flow over it to wash out the chemical. If the person wears contact lenses, they should be removed before flushing the eyes. Seek medical attention.
copper sulfate solution ($1.0\ moldm^{-3}$)	irritant	If copper(II) sulfate contacts the skin, immediately remove any contaminated clothing and rinse the affected area with plenty of water. Use mild soap to wash the skin thoroughly, and gently pat the area dry with a clean towel. If there are any signs of irritation, redness, or other symptoms, seek medical attention. If copper(II) sulfate splashes into the eyes, rinse the eyes immediately with gentle, continuous, and low-pressure flowing water for at least 15 minutes. Hold the affected eye open while rinsing to ensure the chemical is completely flushed out. Remove contact lenses, if applicable, to avoid trapping the chemical between the lens and the eye. Seek immediate medical attention after rinsing the eyes.
celery (extract)	allergen	Handle the celery extract with gloves.

How to make the celery extract

- Break or cut one or two large stalks of celery into several pieces and place in an electric blender. Add approximately 400 cm^3 of distilled water.

- Switch on the blender to make a suspension of celery extract in water.

- Place some muslin in a filter funnel and support the funnel over a beaker. Pour the celery extract into the funnel and leave it so the liquid part of the extract passes through the muslin. You can squeeze it gently to speed up this process.

- Any biological material will contain catalase. If celery is not available, try other plant material, such as potato, carrot, apple or other fruit or vegetable. Animal tissues, such as liver, can also be used, but the catalase in these is often so active that it is difficult to measure the rate of reaction.

How to make the stock solutions of copper sulfate

The stock solution of 1.0 mol dm^{-3} copper sulfate solution can be prepared by putting 16.0 g of copper sulfate in 50 cm^3 of distilled water and making up to 100 cm^3 with distilled water.

This can then be diluted by a factor of ten to produce the solution of 0.1 mol dm^{-3} copper sulfate solution by mixing 10 cm^3 of this solution with 90 cm^3 distilled water.

How to make the solutions of hydrogen peroxide

Pre-prepare concentrations of 1, 2, 3, 4, 5 and 6 vol hydrogen peroxide (H_2O_2) (approximately 50 cm^3 each) in labelled beakers.

Teacher method

This is your version of the method for this experiment that accompanies the *Teacher walkthrough* video.

Do not share this method with learners.

Before you begin

Plan how you will group your learners during the experiment session.

Think about:

- whether there is enough time for all learners to conduct the full investigation, or if different learners could investigate different concentrations of hydrogen peroxide, or inhibitor, and then collate the class results.
- the number of groups you will need (group size 2–4 learners)
- the amount of equipment/chemicals required.

Experiment

Walk around the learners during the experiment in case they encounter any difficulties.

Steps	Notes
1. To prepare the first reaction mixture, 9 cm^3 celery extract is put into a test tube using a syringe.	*Rationale for step 1: N/A*
2. Next, 1 cm^3 of the first copper sulfate stock solution is transferred from the stock beaker into the same test tube using a smaller syringe. This solution, which is a concentration of 0.1 $moldm^{-3}$ copper sulfate mixed with celery extract containing catalase, is inverted to mix.	*Rationale for step 2:* *Note that the concentration of the copper sulfate has been reduced by a tenth compared with the stock solution.*
3. To prepare the second reaction mixture, 9 cm^3 celery extract is put into a test tube using the same 10 cm^3 syringe.	*Rationale for step 3: N/A*
4. Next, 1 cm^3 of the second, more dilute, copper sulfate stock solution is transferred from the stock beaker into the same test tube using a different 1 cm^3 syringe to the one that was used before. This is to prevent contamination of one inhibitor solution with the other. This solution, which now contains a concentration of 0.01 $moldm^{-3}$ copper sulfate, is inverted to mix	*Rationale for step 4:* *Note that again the concentration of the copper sulfate has been reduced by a tenth compared with the stock solution*
5. A final solution is prepared in a third test-tube by adding 9 cm^3 celery extract using the same 10 cm^3 syringe.	*Rationale for step 5: N/A*
6. Next, 1 cm^3 of distilled water is transferred from the stock beaker into the same test tube using a different 1 cm^3 syringe to the one that was used	*Rationale for step 6:* *Adding an equal volume of distilled water means that this test-tube will act as a control experiment*

before. This is to prevent contamination of this syringe with inhibitor. This solution, which contains no inhibitor, is inverted to mix.	*in this investigation.* *These steps have prepared three solutions that will kept stored at room temperature for use later in the investigation.*
7. A holepunch must be used to prepare some small discs of filter paper of an identical size.	*Rationale for step 7:* *5 discs will be needed in total, but it is useful to have 5 spare.*
8. One by one, these filter paper discs can be used to determine the rate of the enzyme-catalysed reaction in the three test-tubes. In this first experiment, the test tube containing no inhibitor will be used.	*Rationale for step 8:* *Note that after using them it is important to wash the forceps, so that they do not contaminate the next experiment.* *It is important to start the timer as soon as the substrate, hydrogen peroxide, is added to the test-tube. This is done using a new syringe that has not been used to transfer any other solution.* *Gently squeeze the hydrogen peroxide down the side of the test tube to avoid disturbing the filter paper disc too much.*
9. Very soon, bubbles begin to form on the surface of the disc. Not long afterwards, the disc begins to rise. It is at this moment that the timer is stopped.	*Rationale for step 9: N/A*
10. This is repeated another two times to obtain three readings for the time, which can be used to calculate a mean value.	*Rationale for step 10: N/A*
11. This procedure is then repeated three times for the next concentration of inhibitor, and then again three times for the final concentration of inhibitor.	*Rationale for step 11:* *In this investigation, five concentrations of substrate, hydrogen peroxide, are used. The procedure described is carried out for each of the concentrations of hydrogen peroxide.*

Clean-up

After the experiment learners should:

- clean all glassware.
- tidy up their workspace.
- ensure any spillages have been mopped up.
- return all equipment and any unused chemicals to you.

Debriefing lesson: Unanswered questions

Resources	• Worksheet E
Learning objectives	By the end of the lesson: • **all** learners will be able to describe the results of their investigation into the effect of copper sulfate concentration on catalase activity using paper discs. • **most** learners will be able to explain the results of their investigation into the effect of copper sulfate concentration on catalase activity using paper discs. • **some** learners will be able to suggest how their investigation into the effect of copper sulfate concentration on catalase activity using paper discs could be extended.

Timings	Activity
20 minutes	**Starter/Introduction** Ask learners to retrieve their graphs and to compare their work with that of their partner. Encourage them to first *describe* the trend for all three plotted lines, and then *explain* the relationship they see. Ask for a few pairs of learners to offer their contributions to the class. Next, provide learners with **Worksheet E**. Encourage them to review their responses considering this information.
35 minutes	**Main lesson** A technique called 'rainbow grouping' can be used to help spread ideas around the class. Give each learner a number or colour. Learners with the same number or colour then join up, making new groups of representatives of each original group. In their new group, learners take turns to: • describe/explain qualitative data (observations) they made • describe/explain quantitative data they collected • describe/explain sources of error in the investigation and how these could be avoided As they work, circulate around the room to offer prompts and guidance.
10 minutes	**Plenary** Remind learners of the importance of their investigation by providing additional context. Ask students to think back to their work in the Briefing lesson to consider specific examples of situations that involve enzyme inhibition in addition to the example of coppoer sulfate and catalase, to illustrate the importance of enzyme inhibition. Discuss examples, such as: ○ administering ethanol after the accidental ingestion of ethylene glycol (antifreeze), due to the competitive inhibition of alcohol dehydrogenase (ADH). ○ competitive inhibition of rubisco by oxygen during photosynthesis. Can they plan investigations that could investigate these situations?

Worksheets and answers

	Worksheet	Answers
For use in *Briefing lesson*:		
Teacher Instructions: What's the question?	**18**	**N/A**
A: Enzyme review	**19**	**28**
B: Poster presentations	**21**	**N/A**
For use in *Planning lesson*:		
C: Measuring the initial rate of a reaction	**22**	**N/A**
D: Justifying choices	**23**	**29**
For use in *Evaluation lesson*:		
F: Appropriate graphing	**26**	**N/A**

Teacher instructions: What's the question?

In this activity, pose questions 'in reverse' to learners. Give them a series of answers and then challenge them to suggest a question for which the answers could be given. This engages learners in higher-order thinking skills. To add an extra degree of challenge, ask learners to decide on the most appropriate command term (taken from the Syllabus) for each of their responses.

Examples should focus on the topics relevant to the upcoming topic. Three examples are provided below.

answer to provide to learners.	suggested question	command word
inhibitor	*'what is the name of a substance that reduces the rate of an enzyme-catalysed reaction?'*	***state***
at high concentrations of substrate, the initial rate of reaction is the same.	*'how can the effects of competitive and non-competitive inhibitors on the initial rate of an enzyme-catalysed reaction be distinguished?'*	***describe***
the inhibitor binds to a part of the enzyme away from the active site and changes the shape of the enzyme's active site, reducing the rate of formation of enzyme-substrate complexes.	*'what happens when a non-competitive inhibitor encounters its enzyme?'*	***explain***

Worksheet A: Enzyme review

Question 1

What determines the specificity of an enzyme?

1 the covalent and other bonding between R groups of the polypeptide

2 the optimum pH of the enzyme

3 the covalent peptide bonds between amino acids of the polypeptide

4 the shape of the substrate molecule

A 1, 2, 3 and 4 **B** 1 and 3 only **C** 1 only **D** 2, 3 and 4 only

Question 2

A student investigated the effect of enzyme concentration on the rate of hydrolysis of protein in milk.

When the enzyme and milk were mixed, the protein was hydrolysed and the mixture changed from cloudy to clear.

The student investigated five different enzyme concentrations and recorded the time taken for the mixture to become clear.

What is an appropriate control for this investigation?

A Carrying out an experiment where the enzyme solution is replaced with water.

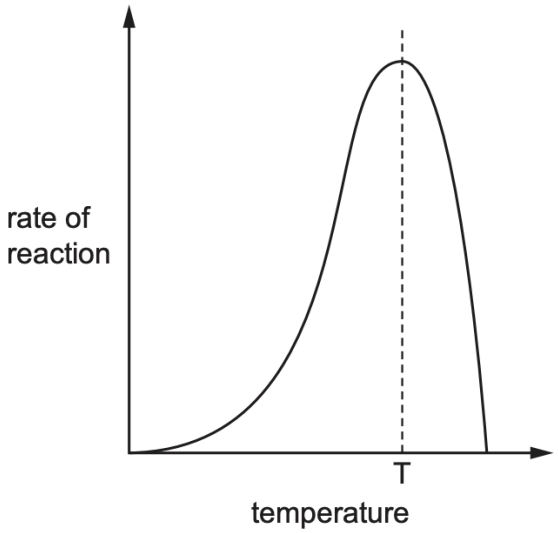
B Carrying out each experiment in a thermostatically controlled water-bath at 35 °C.

C Repeating each experiment three times for each of the five enzyme concentrations.

D Using the same volume of enzyme solution for each of the five experiments.

Question 3

The graph shows the effect of temperature on the rate at which the enzyme in a biological washing powder digests and removes fruit juice stains.



Which statements explain the shape of the graph at temperatures higher than T?

1 Bonds are broken between the R groups of the amino acids in the polypeptide chains of the enzyme.

2 There are more collisions between the enzyme and its substrate.

3 The tertiary structure of the enzyme is altered.

4 The shapes of the active site and the substrate are no longer complementary.

A 1, 2 and 3 **B** 1, 2 and 4 **C** 1, 3 and 4 **D** 2, 3 and 4

Question 4

When developing an enzyme-catalysed reaction for use in industry, the progress of the reaction is studied.

Outline how the progress of an enzyme-catalysed reaction can be investigated experimentally.

.....

.....

.....

.....

.....

.....

..... [3]

Worksheet B: Poster presentations

In this activity, you will work as a group to produce a poster. This poster should illustrate the difference in action between competitive and non-competitive inhibitors on the initial rate of an enzyme-catalysed reaction.

You have been provided with the following resources:

- Glue
- Marker pens
- Modelling clay
- Poster paper
- Scissors

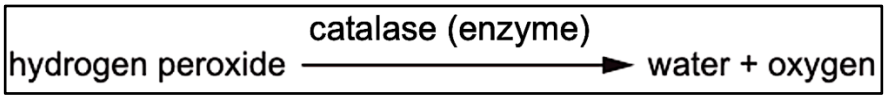
Ensure that you include in your poster reference to:

- Enzyme-substrate and enzyme-inhibitor complexes.
- Active sites and allosteric sites.
- Products.

Your teacher will inform you how long you must complete this activity. When this time has passed, you will be invited to display your work on the wall and receive feedback from other groups in your class.

Worksheet C: Measuring the initial rate of a reaction

The initial rate of an enzyme-catalysed reaction can be measured by either measuring the rate of disappearance of the substrate, or the rate of product formation. Here, you will consider the rate of formation of one of the products of hydrogen peroxide decomposition: oxygen gas.



Outline how you could investigate the effect of changing the copper sulfate concentration on catalase activity using paper discs.

You should use as many of the following 6 terms as you can in your summary.

- begins
- gas
- rate
- start
- stop
- time taken

Worksheet D: Justifying choices

For each step of the method in the table below, two options are provided. Cross out and tick the options you feel are best. Justify your choice.
The first step has been done for you,

Step	Option 1	Option 2	Justify your choice
1	Cut discs of filter paper using a holepunch. ✗	Cut discs of filter paper using scissors. ✓	The dimensions of a filter paper discs will be more difficult to determine
2	Use serial dilution to prepare several hydrogen peroxide solutions of different concentration.	Use proportional (simple) dilution to prepare several hydrogen peroxide solutions of different concentration.	
3	Start the timer when the hydrogen peroxide is added to the test tube.	Start the timer when the filter paper disc begins to rise from the bottom of the test tube.	
4	Start the timer when the filter paper disc begins to rise from the bottom of the test tube.	Stop the timer when the filter paper disc reaches the top of the test tube.	

Safety precautions

To conclude your plan, list three safety precautions you will follow to minimise risk in this investigation.

1.

2.

3.

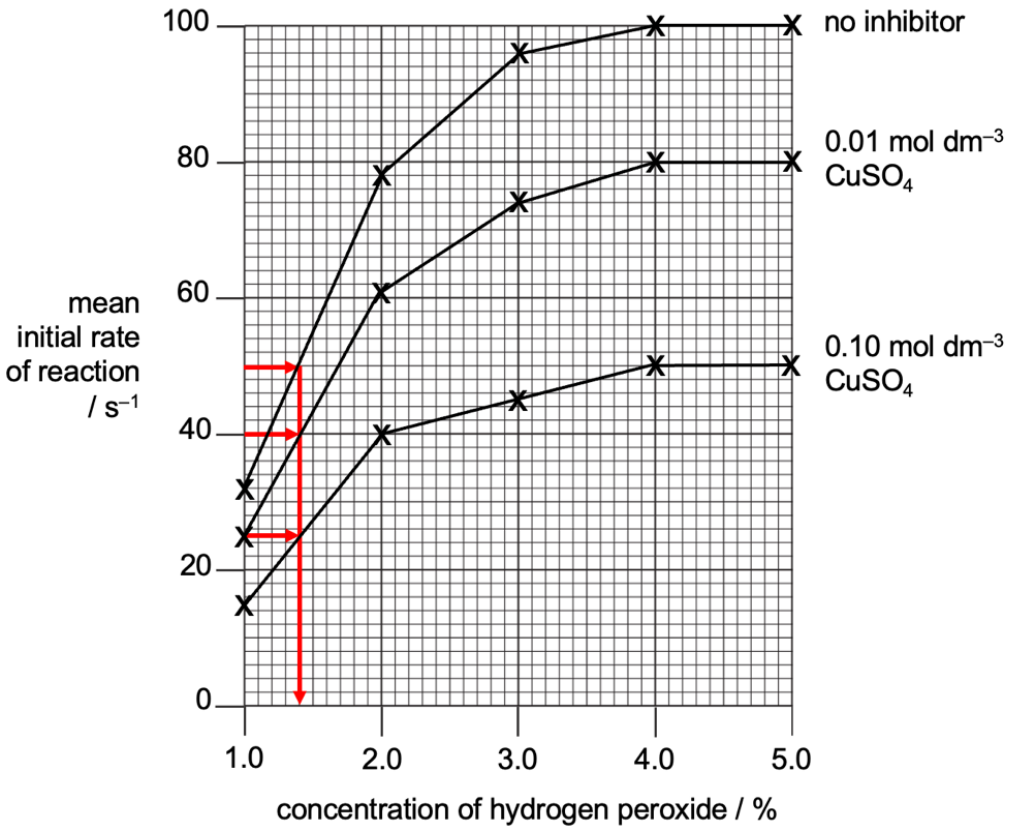
Worksheet E: Appropriate graphing

Use the following guidance to **plot a line graph to show how the mean initial rate of reaction (s $^{-1}$) changes as the concentration of hydrogen peroxide increases, in the absence of coppoer sulfate and in the presence of two different concentrations of copper sulfate**.

When **drawing** your graph, remember to:

- display the independent variable on the x-axis and the dependent variable on the y-axis.
- use a small cross to mark each data point.
- make sure the intersection of the crosses are exactly on the required point.
- make sure the plotted points are connected with a clear, sharp straight line passing through each point (assuming that there are no anomalous points, which should be excluded)
- avoid extrapolating (extending) the curve beyond the plotted points.
- draw the peak where it naturally falls on the curve rather than at the highest point.

Example graph:



If you finish plotting your graph and still have time, consider the following points:

When **describing** a graph, remember to:

- avoid using the term ‘because.’

- Identify specific sections of the plotted line to talk about by looking for changes in the gradient. Use terms such as ‘however’ to make comparisons.
- avoid referring to terms related to time, e.g. ‘slow increase’ and ‘fast decrease.’

When **explaining** the results in a graph, remember to:

- refer to the scientific basis of the changes you have described, by using terms such as ‘because’ and ‘due to.’

Worksheet A: Answers

Question 1
C

Question 2
A

Question 3
C

Question 4

1 ref. to controlled variables ;
e.g. constant, pH / temperature

2 take samples at timed intervals ;
A regular intervals

3 determine, substrate / lactose, concentration
or
determine, product / glucose / galactose, concentration ;

4 plot graph of, substrate concentration / product concentration, against time ;

5 ref. to rate of disappearance of substrate
or
ref. to rate of appearance of product ;

6 determine initial rate ;

Worksheet D: Answers

Step	Option 1	Option 2	Justify your choice
1	Cut discs of filter paper using a holepunch. ✗	Cut discs of filter paper using scissors. ✓	The dimensions of a filter paper discs will be more difficult to determine
2	Use serial dilution to prepare several hydrogen peroxide solutions of different concentration. ✓	Use proportional (simple) dilution to prepare several hydrogen peroxide solutions of different concentration.	A serial dilution is a step-wise series of dilutions, where the dilution factor is the same for each step
3	Start the timer when the hydrogen peroxide is added to the test tube. ✓	Start the timer when the filter paper disc begins to rise from the bottom of the test tube.	The reaction catalysed by catalase will begin from the moment that it encounters hydrogen peroxide
4	Start the timer when the filter paper disc begins to rise from the bottom of the test tube.	Stop the timer when the filter paper disc reaches the top of the test tube. ✓	It is important to measure the total time taken for enough gas to be evolved to cause the disc to rise

Safety precautions

Three safety precautions you should follow to minimise risk in this investigation:

1. Wear gloves.

2. Wear protective eyewear.

3. Pour solutions down the sink with plenty of running water.

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