

This unit should fill approximately **27 periods** of teaching time.

Learning competencies for Unit 3

By the end of this unit students should be able to:

- Define enzymes and explain the properties of enzymes.
- Explain how enzymes are named and then classify them according to their structure.
- Conduct an experiment to show the specificity of an enzyme.
- Appreciate the importance of enzymes in industries and local products.
- Explain how enzymes lower activation energy.
- Explain the mechanism of enzyme action.
- Discuss the action of apo- and coenzymes.
- Give examples of vitamins and minerals in food that act as cofactors.
- Explain factors that affect enzyme activity.
- Investigate the destruction of an enzyme by heat.
- Show how temperature, pH, substrate concentration and enzyme concentration affect enzyme activity.
- Explain allosteric regulation and feedback control mechanism of enzyme activity.
- Appreciate the role of enzymes in controlling our metabolic activities.

This section should fill approximately **7 periods** of teaching time.

3.1 Nature of enzymes

Learning competencies

By the end of this section students should be able to:

- Define enzymes and explain the properties of enzymes.
- Explain how enzymes are named and then classify them according to their structure.
- Conduct an experiment to show the specificity of an enzyme.
- Appreciate the importance of enzymes in industries and local products.

Starting off

Introduce the topic by explaining to students that nearly all enzymes are proteins. There are a few RNA molecules that have catalytic properties but, effectively, all enzymes are proteins.



Discuss with them the structure of lipase (figure 3.1 in the Students' Book) and identify the regions suggested. Get them to appreciate that no other enzyme will have a tertiary structure quite the same as this.

From there, familiarise the students with the term and concept 'substrate' as it will be used a lot in this unit. Also, introduce the concept of an 'active site' within the enzyme molecule as the region of the enzyme that is chiefly responsible for the specificity of the enzyme.

Use figure 3.2 in the Students' Book as a model for the cycle of events that take place in an enzyme-controlled reaction and introduce the terms:

- enzyme–substrate complex (stage 2 in figure 3.2)
- enzyme–product complex (stage 3 in figure 3.2)

By the end of the introduction, the definition of an enzyme given in the Students' Book:

An enzyme is a globular protein with a uniquely shaped active site; it acts as a biological catalyst for a specific reaction, but remains unaltered by the reaction.

should be meaningful to the students.

Teaching notes

Revise briefly with the students the nature of enzymes. Stress again that (for all practical purposes) they are all proteins. Discuss with them the specificity of enzymes and the implication of this for control of all the different chemical reactions that take place in living cells.

Mention that they can be inactivated by extremes of pH and by high temperatures and that their activity is influenced by their concentration and the concentration of their substrate. However, this is only an introduction as these will be discussed in more detail later.

The nature of catalysis is an important concept for them to grasp. It should not be new as they will likely have encountered it before in studies in chemistry. Stress that catalysts only alter the pathway of the reaction, not the nature of the products or the overall energy change in the reaction. Stress also that the catalyst remains in its original form at the end of the reaction.

The naming of enzymes is a mystery to many students. They will probably have noticed that the names of many enzymes end in –ase. Start from here and ask them if they have noticed anything about the rest of the name. Ask them to name the enzymes that act on proteins (they should know protease) and lipids (lipase). Explain that another common way of naming enzymes is by the type of reaction they catalyse (polymerase, dehydrogenase, oxidase are examples). Discuss the varied ways of naming enzymes before moving towards the formal method of classifying enzymes approved by the Enzyme Commission.

The students should find the initial classification into six divisions/classes straightforward, even if some of the names are a little difficult to comprehend, but they will need help with the other levels of classification. Point out the benefits of such a classification; it is accessible to all biologists everywhere and is not really language dependent.

The uses of enzymes is a huge topic and lends itself to a display by the students. It is as well to start by giving the students a historical perspective on this, explaining that man was using enzymes before he ever knew what enzymes were! The



examples of brewing and baking illustrate this point. Further examples are best chosen to illustrate local use of enzymes.

Point out to the students that, because enzymes allow reactions to proceed at moderate temperatures, there are benefits beyond the cost-saving to the manufacturer. If less heat is required, then less fuel need be burned, which is of benefit to the environment by reducing the amount of CO₂ released into the atmosphere. Figure 3.7 in the Students' Book shows some of the savings in CO₂ emissions per ton of product in some industrial processes.

SA = starter activity MA = main activity CA = concluding activity	
The nature of enzymes	
SA	Students examine the idea that all enzymes are globular proteins.
MA	Students discuss the consequences of enzymes being globular proteins: • unique tertiary structure (and therefore shape) • regions with an α-helix and a β-pleated sheet • specificity of action. Students examine the concepts of active site, substrate and enzyme-substrate complex and discuss the properties of enzymes (including those of a catalyst).
CA	Students copy the definition of an enzyme in the text and make notes on the properties of enzymes, explaining why enzymes are specific and why they are affected by temperature.
Naming and classifying enzymes	
SA	Students give the names of some enzymes and to find the pattern in the names – that they frequently end in -ase.
MA	Students discuss with the teacher the origin of the first part of the name of an enzyme such as: • lipase • oxidase to establish different ways of naming enzymes. Class to discuss briefly the principles of the Enzyme Commission classification and the benefits of systematic naming of enzymes.
CA	Give the students examples of enzymes and ask them to deduce how they have been named (i.e. by the substrate they act on, by the type of reaction they catalyse, common naming, etc.). Students can do Activity 3.2 in class if there is time, or as homework. Review their answers quickly at the beginning of the next lesson.
Uses of enzymes (1)	
SA	Students learn the origin of the word 'enzyme' – point out that it means 'in yeast'. Students learn that yeast was the first source of enzymes to be used.
MA	Students discuss the role of yeast in brewing and baking. Point out that fermentation of sugars happens in both processes.
CA	Students make a display on brewing tella and baking injera.
Uses of enzymes (2)	
SA	Students give one or two examples of processes in which enzymes are used. Students discuss the benefits of using enzymes in these processes.
MA	Students use table 3.3 and their own sources to make a display of how enzymes are used in different processes.
CA	Class reviews some of the uses of enzymes.
Uses of enzymes (3)	
SA	Students begin discussion on when and where enzymes are used in local manufacturing – write their ideas on the board.

MA	Students conduct the debate as described in activity 3.3: you will need to chair it, and ensure that all students get a turn to speak and respect each other's opinions.
CA	Students conclude the debate; make notes of the main points in preparation for the next lesson.
Uses of enzymes (4)	
SA	Students recall the main ideas and opinions from the debate in the previous lesson and discuss whether they have changed their minds at all since – if so, why? You may have to prompt them using your notes.
MA	Students write a summary report of the discussion or present it as a diagram or poster.
CA	Students study figure 3.7.
Uses of enzymes (5)	
SA	Students recall that enzymes increase the rate of reactions of biological processes at moderate temperatures.
MA	Students discuss the benefits of using moderate temperatures, including saving on energy bills, reduced emissions of carbon dioxide from burning fossil fuels. Students can study the 'Did you know' box for ideas.
CA	Students make notes on the benefits of using enzymes in industrial processes, quoting examples.

Answers to review questions

1. B 6. D
2. C 7. D
3. A 8. C
4. D 9. B
5. A 10. D

3.2 Functions of enzymes

Learning competencies

By the end of this section students should be able to:

- Explain how enzymes lower activation energy.
- Explain the mechanism of enzyme action.
- Discuss the action of apo- and coenzymes.
- Give examples of vitamins and minerals in food that act as cofactors.

This section should fill approximately **9 periods** of teaching time.

Starting off

Begin by reminding the students of the nature of enzymes, that they are:

- proteins
- biological catalysts
- specific
- affected by pH and temperature

- affected by the concentration of their substrate and the presence of certain substances that act as inhibitors.

Recap also the nature of catalysts, remind them that catalysts:

- speed up a chemical reaction with no overall change to:
 - the nature of the products
 - the energy change that takes place during the reaction
 - the catalyst itself.

Teaching notes

Introduce the nature of catalysis by enzymes with a discussion of energy change during the reaction. Stress again that there is no overall change in the energy levels of reactants and products, but that the pattern of the reaction alters. Introduce the concept of activation energy as the energy required to initiate a reaction. Use figure 3.8 in the Students' Book to compare the activation energy of a catalysed and uncatalysed reaction.

Discuss the consequences of a lower activation energy being required in terms of molecules having the necessary kinetic energy. Explain that not all molecules have the same kinetic energy and that only those with sufficient kinetic energy will be able to 'climb the activation energy hill'. If this 'hill' is lower, a larger proportion of the molecules will have the necessary energy. The diagram below may be helpful when discussing this concept.

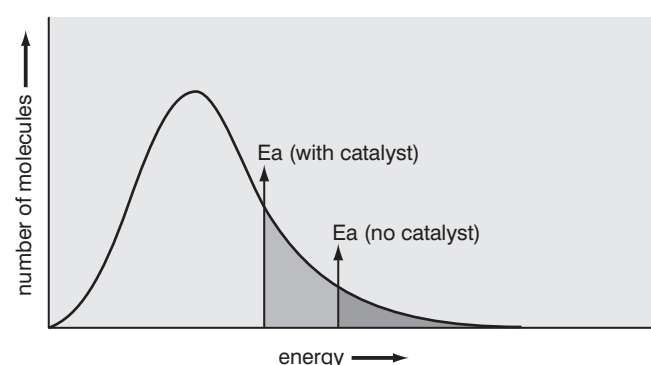


Figure 3.1 Activation energy with and without catalyst

The two models of enzyme action, lock-and-key and induced-fit models, are described in the Students' Book. Explain to the students reasons for preferring the induced-fit model, including:

- it explains how the transition state might lower activation energy (by putting bonds in the reactant under 'tension' so that they are more likely to break and re-combine; this is a rather simplistic and physical explanation, but it gives the students some idea of how activation energy might be lowered)
- it better explains the action of non-competitive inhibitors, whose action requires the re-shaping of the active site.

Table 3.4 gives some data on the effects of enzymes on increasing the rate of reaction. Some of the concepts in the table are not essential to understanding how enzymes work, but the turnover number is a useful idea for them to be able to carry around. Ask students to write out some of the rate enhancements longhand to give a better appreciation of the effect of the enzyme.

Finally, we look at the question of cofactors and how they are sometimes necessary for enzyme action. Figure 3.12 in the Students' Book illustrates why this might be. The apoenzyme provides the basic structure, but the active site is not truly complementary in shape to the substrate without the cofactor. This is not the only mode of action of cofactors, but it does give the students a physical model to build the concept around.

Table 3.5 gives some examples of cofactors together with the enzyme that they activate and the role of the active enzyme. Table 3.6 gives similar information about some mineral ions and their role in activating enzymes.

SA = starter activity MA = main activity CA = concluding activity	
How do enzymes act as catalysts?	
SA	Students discuss what a catalyst does.
MA	Students examine the concepts of: • energy levels of reactants and products • activation energy • exergonic (exothermic) reactions • endergonic (endothermic) reactions. Explain that by reducing the activation energy needed, the energy input to the system is reduced and so more product is formed for the same energy input – i.e. the reaction proceeds faster.
CA	Students make notes on these concepts.
Enzyme model	
SA	Students brainstorm the characteristics and function of an enzyme.
MA	Students complete activity 3.5, making their model if possible.
CA	Students present either their plan for a model or actual model to the class and explain their design.
How do enzymes lower activation energy (1) – the lock-and-key model	
SA	Students discuss the concept of complementary shapes and enzyme–substrate complex (ES complex).
MA	Students examine that forming an ES complex can lead to: • a transition state • an alternative pathway for the reaction with a lower energy demand. Explain also that a weakness of the model is that it does not explain how the transition state is formed, nor how it reduces the energy demand.
CA	Students make notes on this.
How do enzymes lower activation energy (2) – the induced-fit model	
SA	Students discuss the concept of complementary shapes and enzyme–substrate complex (ES complex).
MA	Students discuss the concepts of the induced-fit model paying particular attention to the conformational change that takes place as enzyme and substrate bind. Students discuss the concept of 'turnover rate' as a measure of rate of reaction of an enzyme-controlled reaction.
CA	Students summarise the similarities and differences between this model and the lock-and-key model.

Industrial visit (1)	
SA	Tell the students about the industrial location to which you have chosen to take them on a visit, for activity 3.4.
MA	Preparation for visit: students plan what they will need to find out, questions to ask, and do some personal library/Internet research into the industry.
CA	Students draft a structure for their eventual report on their visit.
Industrial visit (2)	
SA	Students complete industrial visit (will take whole lesson and perhaps longer).
MA	Students continue industrial visit.
CA	Students continue industrial visit and write up their report as homework.
Research project (1)	
SA	Students brainstorm different industrial uses of enzymes. Push them to think creatively about different situations where enzymes may be used.
MA	Students select one use of enzymes (not the one already covered in the industrial visit) to write a full research project on). Ensure that there is a good variety of topics across the class, and that each one chosen provides sufficient scope for a detailed report. Students plan the structure of their project and scope of work, and identify the information sources they will need, including independent research visits and interviews where possible and relevant.
CA	Students begin work on their projects and continue their research outside of the classroom as homework.
Research project (2)	
SA	Students show evidence of progress on their projects to you, and continue work.
MA	Students complete research projects.
CA	Students present a brief summary of their research projects to the rest of the class.
Cofactors	
SA	Students examine the idea that active enzymes may not be single entities.
MA	Students examine the distinction between cofactors as a generic group of substances that activate apoenzymes and the different types of cofactors. In particular, students examine coenzymes and NAD and FAD as examples of coenzymes in the respiratory pathway. Students be aware of some mineral ions that act as cofactors.
CA	Students describe the role of cofactors.

Answers to review questions

1. B 6. A
2. C 7. B
3. B 8. A
4. D 9. B
5. C 10. C

3.3 Factors affecting the functions of enzymes

Learning competencies

By the end of this section students should be able to:

- Explain factors that affect enzyme activity.
- Investigate the destruction of an enzyme by heat.
- Show how temperature, pH, substrate concentration and enzyme concentration affect enzyme activity.
- Explain allosteric regulation and feedback control mechanism of enzyme activity.
- Appreciate the role of enzymes in controlling our metabolic activities.

This section should fill approximately **11 periods** of teaching time.

Starting off

Begin by reminding the students of the basic pattern of an enzyme-controlled reaction described in figure 3.2 and reproduced again here for convenience. Discuss with the students how different factors might influence the pattern of this reaction. Get them to think about the influence of:

- more substrate molecules (substrate concentration)
- more kinetic energy (higher temperature)
- more enzyme molecules (enzyme concentration)

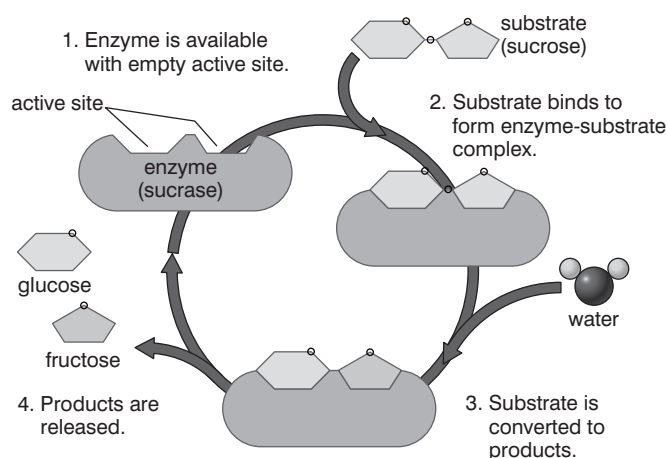


Figure 3.2 The hydrolysis of sucrose by sucrase

Teaching notes

Begin discussing the effect of temperature on enzyme action by establishing the fact that heat gives particles kinetic energy with the result that the particles will either:

- move around more quickly if they are free to do so (in this case they are more likely to meet other particles and react)
- vibrate more vigorously if they are held in place as part of a molecule (in this case the increased vibration will put extra strain on the bonds holding the particles in position).

Introduce the concept of denaturation by considering the second of the above effects in a little more detail. Stress to them that denaturation is normally non-reversible *in vitro* but can often be reversed *in vivo*, provided the temperatures haven't been too extreme. The concept of non-reversible denaturation can be

illustrated by thinking of boiling an egg. The liquid albumen protein becomes white and solid because it has been heat denatured. It does not revert to a translucent liquid on cooling. The effect is not reversible.

Stress to students that the activity of an enzyme at a given temperature is a 'trade-off' between the effect of increased collisions and the effect of denaturation, both of which increase with temperature. Get them to realise that there will be some denaturation at the optimum temperature, and to think of the optimum temperature as:

the highest temperature at which the effects of increased collisions still outweigh the effect of denaturation.

At temperatures higher than the optimum, denaturation occurs much more rapidly and activity soon reduces to zero. Ask the students to recognise this in the pattern of the activity/temperature graph. Also, get them to realise that not all enzymes have an optimum of 37 °C! Ectothermic animals living under the polar ice need enzymes to function efficiently at temperatures much lower than that.

With the effects of denaturation established, the effects of extreme pHs are straightforward to explain, although the mechanism of inactivation is different to that for temperature. Also, students must appreciate that some enzymes do have optimum pHs that are not neutral. This is easily demonstrated by reference to the enzymes from different regions of the human gut.

At one level, the effect of substrate concentration is straightforward for the students to understand; they readily appreciate that more substrate molecules means more enzyme–substrate complexes and, so, a faster rate of reaction. However, it requires an appreciation of 'turnover number' to fully explain the shape of the graph in figure 3.17 in the Students' Book, reproduced here again for reference. At high substrate concentrations, $V^{\max}$ is approached as nearly all the active sites are occupied at any given time.

Discussing the effect of enzyme concentration allows a distinction to be made between enzyme activity (the turnover rate of each individual molecule of enzyme) and reaction rate (the overall rate at which product is formed).

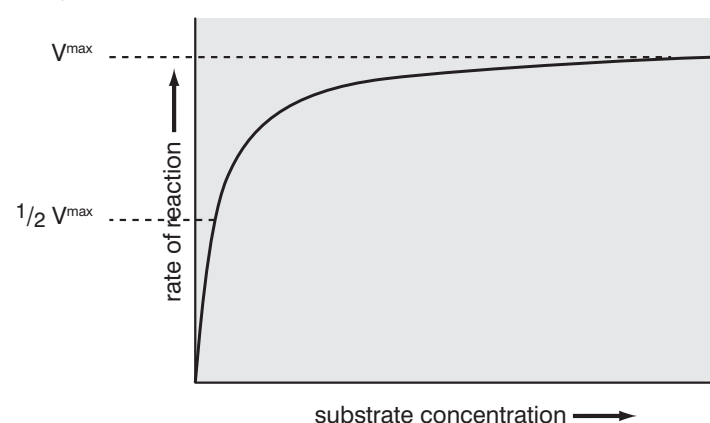


Figure 3.3 The effect of substrate concentration on enzyme activity

There are so many opportunities for practical work with enzymes that it is difficult to know which to suggest. However, a number are suggested in the Students' Book that are based on the decomposition of hydrogen peroxide by catalase; this is often a convenient practical as there are many sources of catalase. One method of measuring the volume of oxygen produced is suggested in the Students' Book, but there are clearly others.

At the most basic level, numbers of bubbles can be counted (students could then in their evaluations point out that the bubbles are unlikely to all be of the same volume).

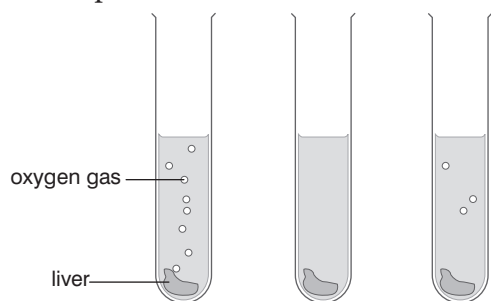


Figure 3.4

Alternatively, the volume of oxygen gas produced can be measured by collecting the gas in an inverted measuring cylinder.

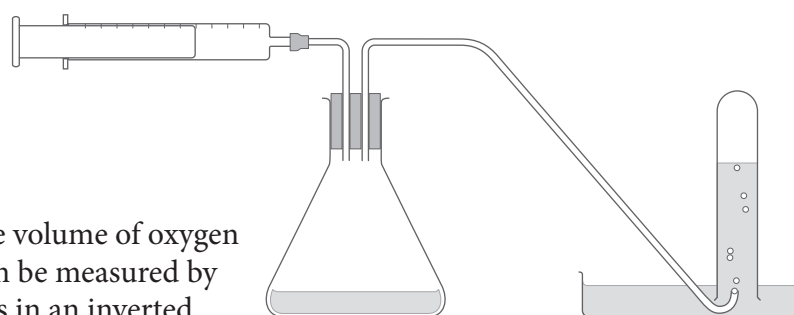


Figure 3.5

Whatever method is chosen to measure the volume of gas produced, there is considerable scope to discuss with the students:

- issues relating to the validity of the experiment; these could include:
 - does the experiment really measure what is intended to be measured or could there be other factors that could be influencing the results?
 - is the range of values for the IV appropriate to draw valid conclusions?
- issues relating to the reliability of the experiment; these could include:
 - have repeats been carried out to ensure reproducibility of the results?
 - were changes in the DV (volume of gas produced) measured to an appropriate degree of accuracy?
 - have changes to the IV (temperature, pH) been maintained to an appropriate degree of accuracy?

If it is not practicable for some reason to carry out these, or other, investigations then sample results are provided for analysis.

In each case, students should:

- calculate mean values from the raw data supplied
- plot graphs of mean values of the DV against the IV
- describe any patterns evident in the graphs
- try to explain these patterns using biological knowledge of enzyme action
- suggest how the investigation might be improved to give more reliable and valid results.

The effect of competitive inhibitors on enzyme action is relatively easy for student to grasp; they can appreciate that if another substance has a similar shape to that of the substrate, then it could enter the active site and prevent substrate molecules from entering.

The Institute of Biology recommends that for graphs such as these:

- mean data are always plotted
- points are joined by ruled lines or, where appropriate, a line of best fit is drawn through the points.

What is more difficult for them is to appreciate the proportionality of this effect. They need to appreciate that in a mix of 80% substrate and 20% inhibitor, 80% of the collisions with the enzyme will be substrate molecules and 20% will be inhibitor molecules. Put another way, each active site will be occupied with substrate for 80% of the time and with inhibitor for 20% of the time, reducing enzyme activity to 80%.

The effect of non-competitive inhibition is a very visual concept. Figure 3.22 in the Students' Book should help the students to understand how binding to the allosteric site can influence the active site. This will be easier to grasp if they have fully understood the induced-fit model of enzyme action.

The role of non-competitive inhibitors in feedback control of metabolic activity is the final concept in this unit. Students need to appreciate that some kind of control mechanism must exist in cells otherwise they would just continue making product that could not be used. This would be at least wasteful of resources and at worst lethal to the cell if the product were toxic in high concentrations.

The concept of feedback inhibition is usually quite readily appreciated if the idea of non-competitive inhibition is fully understood. Explain to the students not just the switching off of the pathway but also that, when concentrations of the end-product fall again, the inhibition of the key enzyme will be removed and the pathway will be reinstated.

SA = starter activity MA = main activity CA = concluding activity	
The effect of temperature on enzyme activity	
SA	Students examine the distinction between heat (a form of energy) and temperature (a measure of the heat content of a system).
MA	Students discuss how heat might affect: • the kinetic energy of the molecules involved in a reaction • the kinetic energy of the atoms making up each molecule. From these ideas develop the ideas that increasing temperature will lead to: • increased kinetic energy of enzyme and substrate molecules, making collision and binding more likely • increased rate of movement of atoms within a molecule – including the enzyme molecules. The increased rate of movement of atoms will lead to denaturation – leading, in turn, to a change in shape of the active site, making binding with the substrate less likely the activity of an enzyme will depend on a trade-off between the two effects.
CA	Students copy figure 3.14 and explain the shape of the graph.
The effect of pH on enzyme activity	
SA	Students revisit the idea that anything that makes binding between active site and substrate will reduce the activity of the enzyme.
MA	Students examine the dual effects of extreme pHs in: • changing the charge structure of the active site • breaking ionic bonds leading to denaturation of the enzyme molecule.
CA	Students copy and explain the shape of the graphs in figure 3.15 and figure 3.16.
The effect of substrate concentration	
SA	Students remember what happens in an enzyme-controlled reaction – in both models of enzyme action, the active site binds with the substrate.

MA	Students remember the concept of 'turnover rate'. Go on to discuss the conditions for maximum rate of reaction – all the active sites must be operating at their maximum turnover rate. Students sketch figure 3.17 and explain the shape of the graph.																																																
CA	Students read and discuss the key idea of changing substrate concentration over the duration of a reaction and to sketch a graph for the rate of reaction as the reaction proceeds.																																																
Catalase practical to measure enzyme activity (1)																																																	
SA	Students discuss with the teacher the two generic ways of measuring the rate of an enzyme-controlled reaction: • measuring the rate at which the substrate is used up • measuring the rate at which (one of) the product(s) is formed. Discuss the reasons for choosing the second of these in the case of the catalase reaction.																																																
MA	Students carry out the experiment (activity 3.6). This is a good, basic experiment to show enzyme activity. Detailed information on this is given in the teaching notes.																																																
CA	Students write up and plot a graph of their results.																																																
Catalase practical to measure enzyme activity (2) – improving the investigation																																																	
SA	Students discuss with the teacher reasons why the reliability and validity of the experiment in its original form are limited (lack of controls limit validity and lack of repeats limit reliability).																																																
MA	Students discuss with the teacher realistic ways of improving the reliability and validity of the experiment.																																																
CA	Students write up ways of improving the experiment, using table 3.7 as a basis.																																																
Catalase practical to measure enzyme activity (3) – planning further investigations																																																	
SA	The students are to design their own investigations. Students recall the importance of choosing: • a suitable way of changing the independent variable • a suitable range of values for the independent variable • a suitable method of measuring the dependent variable • suitable ways of controlling other variables.																																																
MA	Students design experiments to investigate the effect of: • temperature • pH • substrate concentration • enzyme concentration on the activity of catalase. This could be in the form of a table such as: <table><tr><th>IV</th><th>How varied</th><th>DV</th><th>How measured</th><th>Controlled variables</th><th>How controlled</th></tr><tr><td></td><td></td><td></td><td></td><td></td><td></td></tr><tr><td></td><td></td><td></td><td></td><td></td><td></td></tr><tr><td></td><td></td><td></td><td></td><td></td><td></td></tr><tr><td></td><td></td><td></td><td></td><td></td><td></td></tr><tr><td></td><td></td><td></td><td></td><td></td><td></td></tr><tr><td></td><td></td><td></td><td></td><td></td><td></td></tr><tr><td></td><td></td><td></td><td></td><td></td><td></td></tr></table>	IV	How varied	DV	How measured	Controlled variables	How controlled																																										
IV	How varied	DV	How measured	Controlled variables	How controlled																																												
CA	Students write up their plans.																																																

Catalase practical to measure enzyme activity (4) – optional – students carry out planned experiments	
SA	Students recall safety requirements and modify their plans if: • there are safety issues • there are issues of reliability or practicability.
MA	Students carry out their planned experiments (this may take several sessions).
CA	Students write up the results of their investigations.
Catalase practical to measure enzyme activity (5) – analysing the results	
SA	Discuss with the students the reasons for: • calculating and using mean values • plotting graphs of their results.
MA	Students calculate means and plot graphs of their results or of the data supplied in the textbook.
CA	Students write up conclusions from their graphs.
Types of enzyme inhibitors	
SA	Students discuss the concept of an inhibitor and suggest how another substance might reduce the turnover rate of an enzyme. Students focus on turnover rate.
MA	Students examine that there are different types of inhibitors: • irreversible • reversible • competitive • non-competitive and explain how they work. Students make notes on this.
CA	Students explain the action of the painkillers: • aspirin • ibuprofen in terms of enzyme inhibition.
Enzyme inhibitors in living cells	
SA	Students examine the concept of a metabolic pathway.
MA	Students discuss why metabolic pathways need to be ‘switched on and off’. Students examine that the end-product of a metabolic pathway often inhibits one of the earlier reactions, thus limiting the rate of the production of the end-product.
CA	Students make notes on the importance of: • end-product inhibition • activators in controlling metabolic pathways.

Answers to review questions

- | | |
|------|-------|
| 1. D | 6. D |
| 2. B | 7. A |
| 3. B | 8. D |
| 4. C | 9. D |
| 5. C | 10. D |

Answers to end of unit questions

1. a) Non-protein substance that binds with apoenzyme to form active enzyme
b)

Type of group	Organic	Protein	Binds tightly
Apoenzyme	✓	✓	✓
Coenzyme	✓	✗	✓
Ion	✗	✗	✗

One mark per correct row

If no correct rows, allow one mark per correct column

2.

Factor controlled	How controlled	Reason for controlling factor
Temperature	Water bath	Temperature increases/ decreases rate of reaction High temperature may denature enzyme
pH	pH buffer	Changes in pH can alter charge on amino acids in the active site
Substrate concentration	Equal strength solutions	Different concentrations influence rate of reaction by affecting number of active sites in use

3. a) Enzyme A 36–38 °C
Enzyme B 76–78 °C
These temperatures show peak enzyme activity.

- b) Enzyme B
high optimum temperature
thermophilic means 'heat-loving'

- c) Rate of reaction increases
increased temperature gives more kinetic energy
increased collisions between enzyme and substrate
more enzyme–substrate complexes formed

4. a) i) Reaction velocity increases
increased number of substrate molecules
increased number of collisions with active sites
ii) Reaction velocity levels off
all active sites occupied at any time
extra substrate molecules have no effect

- b) Line increases to same maximum
but maximum reached at a lower concentration
- 5. a) Initial energy of reactants is the same
final energy of products is the same
- b) *Three of:*
X and Y represent activation energies
higher for uncatalysed reaction
because catalyst/enzyme allows transition state
bonds in reactants put under strain
- c) *Three of:*
Lower activation energy
so more reactant molecules have sufficient kinetic energy
to 'climb activation energy hill'/to achieve activation energy levels
so more reactions per second
- 6. a) Any three suitable examples such as:
washing powders, leather industry, brewing, baking
- b) Reduce heating costs
reduce emissions
- c) Lower temperatures due to lower energy requirements
reduce amount of fuel burned
reduce CO₂ emissions
- 7. a) i) 19 °C
ii) May have been quicker just lower or just higher than this
need more temperatures to be sure
- b) Enzyme is a catalyst
only speeds up the reaction, does not cause it to happen
- c) i) Increased temperature increases kinetic energy
more collisions
more enzyme-substrate complexes
- ii) Increased temperature causes increased vibration of atoms in the
enzyme molecule
begins to denature/active site changes shape
substrate molecules will no longer fit/bind with active site
- 8. a) *Four of:*
Inhibitor binds with allosteric site/site away from active site
enzyme undergoes conformational change
active site changes shape
substrate molecules cannot bind
extent of inhibition does not depend on amount of substrate/extent of
inhibition only depends on amount of inhibitor



- b) (i) Substance D is the end product
each reaction dependent on the one before it
if the chain of reactions is broken, end product will not be formed
end product inhibits E1
by non-competitive inhibition
- (ii) End product can inhibit an enzyme in a pathway
so pathway is 'switched off'/cannot take place
so concentration of end product decreases
as it is used up by the cell
as concentration decreases, inhibition of enzyme decreases
enzyme becomes active and pathway active again

Answers to end of unit crossword puzzle

Across

- 6 specific
13 ASE
15 non-competitive inhibitor
16 induced fit
17 hydrolysis
18 temperature
20 cofactor

Down

- 1 biological catalysts
2 enzyme-substrate complex
3 washing powder
4 optimum
5 activation energy
7 catalysts
8 lock and key
9 active site
10 competitive inhibitor
11 concentration
12 globular protein
14 apoenzyme
19 PH

Further resources

http://www.biochemistry.org/Portals/0/Education/Docs/BASC03_full.pdf – research and further activities related to enzymes

http://www.biology.arizona.edu/biochemistry/problem_sets/energy_enzymes_catalysis/energy_enzymes_catalysis.html – problems/exercises on enzymes

<http://www.practicalbiology.org/areas/advanced/bio-molecules/enzyme-catalysed-reactions/> – example experiments

<http://www.practicalbiology.org/areas/advanced/bio-molecules/quantitative-food-tests/> – example food test experiments

