

Enzymes revision notes

Enzymes are proteins that speed up the chemical reactions that keep organisms alive. This topic covers what enzymes are, how the shape of the active site explains what they do, and how temperature and pH change their activity. It also covers how to investigate these effects in the laboratory.

For Cambridge IGCSE Combined Science 0653. **Supplement** is Extended content, only examined on Papers 2 and 4.

1 Enzymes Spec B5.1

Enzymes as biological catalysts Spec B5.1.1

A **catalyst** is a substance that increases the rate of a chemical reaction and is not changed by the reaction. Because a catalyst is not changed, it is not used up, so a small amount can be used again and again.

Enzymes are proteins that are involved in all metabolic reactions, where they function as **biological catalysts**. Metabolic reactions are the chemical reactions that happen inside living organisms, such as breaking down food and building new molecules.

Catalyst	A substance that increases the rate of a chemical reaction and is not changed by the reaction.
Enzyme	A protein that functions as a biological catalyst.

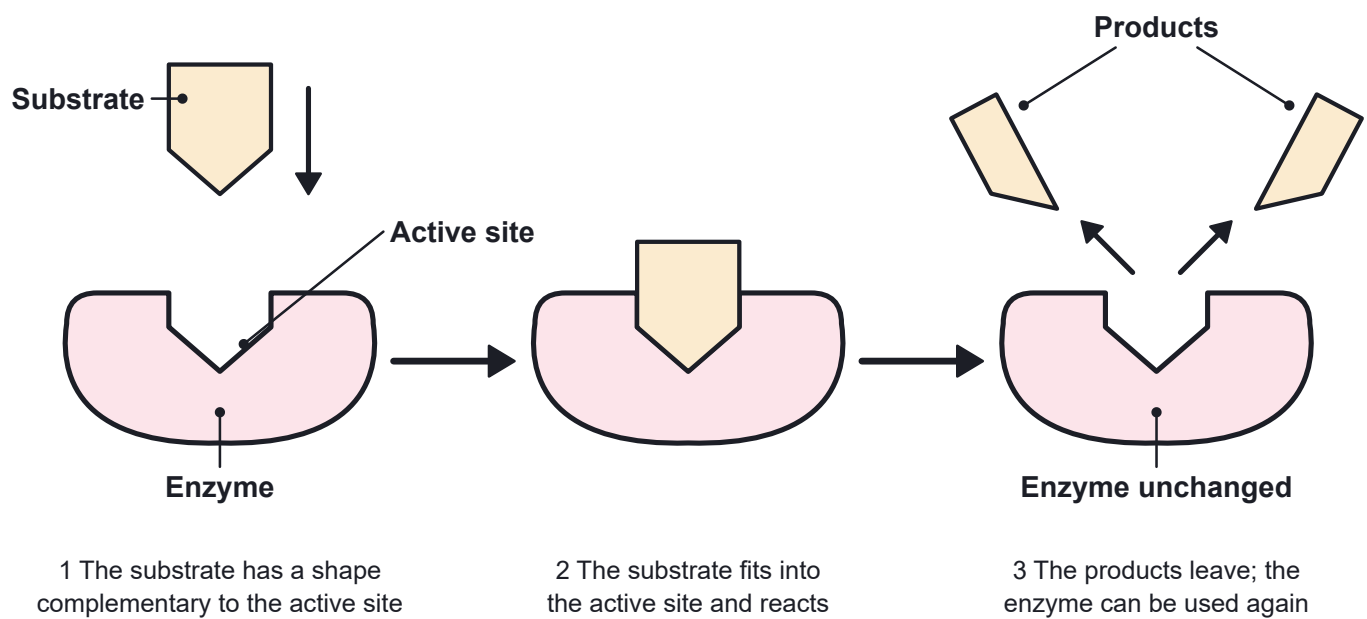
- Enzymes are proteins.
- They speed up reactions but are not changed or used up.
- They are needed for all metabolic reactions, so every living organism has them.

Common mistake: Do not say that enzymes are 'alive' or that they are 'killed' by heat. Enzymes are proteins, so they are denatured, not killed.

How enzymes work Spec B5.1.3, B5.1.4 Supplement

Each enzyme has a region called the **active site**. The substance the enzyme acts on is the **substrate**. The shape of the active site is **complementary** to the shape of its substrate, so the substrate fits into it.

1. The substrate fits into the active site of the enzyme, because their shapes are complementary.
2. The reaction takes place and the substrate is changed into the product or products.
3. The products leave the active site. The enzyme is unchanged and can be used again.

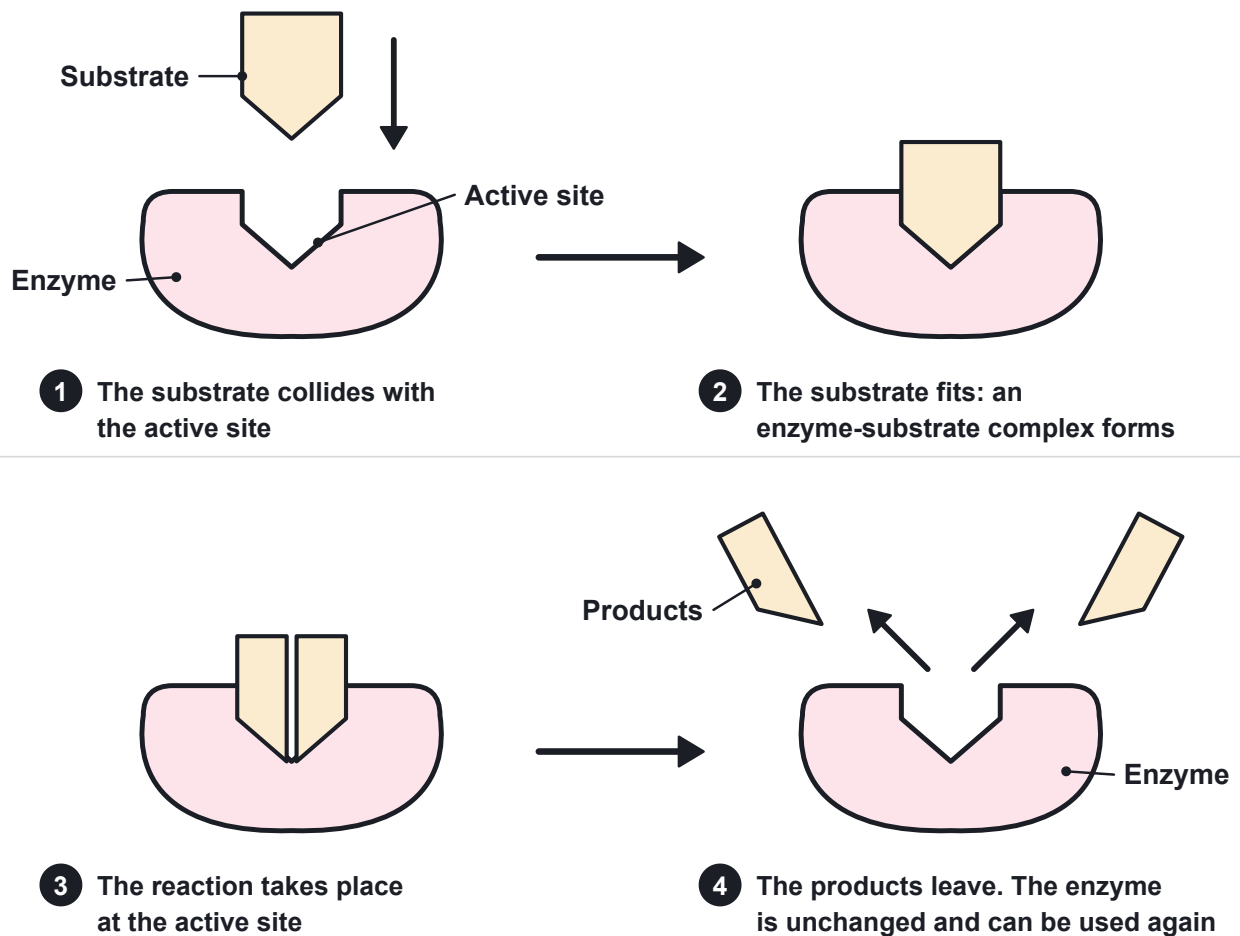


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The substrate fits the **active site** because their shapes are **complementary**. The enzyme is unchanged.

*Exam tip: Use the word **complementary** for the shapes of the active site and substrate. Do not say the shapes are 'the same'.*

Supplement



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The lock and key model, showing the enzyme-substrate complex. The enzyme is not used up.

Supplement

In more detail, the substrate molecule collides with the active site and binds to it, forming an **enzyme-substrate complex**: the temporary structure formed when the substrate is bound to the active site. The complex forms only when the shape and fit are right. The substrate is then changed into products, which leave the active site.

Supplement

Enzymes are **specific**. This means each enzyme acts on only one substrate (or a very small group of similar substrates). Only a substrate with a shape complementary to the active site can fit into it and form an enzyme-substrate complex. A substrate with a different shape does not fit, so no reaction takes place.

Supplement

Key terms for enzyme action

Term	Meaning
Active site	The region of the enzyme where the substrate binds
Substrate	The substance the enzyme acts on
Enzyme-substrate complex	The enzyme with its substrate bound in the active site
Product	The substance made by the reaction

Supplement

Exam tip: When asked to explain enzyme action, use all four terms: active site, substrate, enzyme-substrate complex and product.

Temperature, pH and enzyme activity Spec B5.1.5, B5.1.6 Supplement

Each enzyme has an optimum temperature and an optimum pH, at which its activity is highest. The effects are explained using kinetic energy, shape and fit, the frequency of effective collisions, and denaturation.

Temperature

- Below the optimum: enzyme and substrate molecules have less kinetic energy, so they move more slowly. The frequency of effective collisions between substrate and active site is low, so few enzyme-substrate complexes form and the rate is low.
- As temperature rises towards the optimum: kinetic energy increases, so the frequency of effective collisions increases. More enzyme-substrate complexes form per second and the rate rises.
- Above the optimum: the enzyme molecules vibrate more and the bonds holding the enzyme in shape break. The shape of the active site changes, so the substrate no longer fits. The enzyme is denatured, fewer enzyme-substrate complexes form, and the rate falls steeply, reaching zero.

pH

A pH above or below the optimum changes the bonds that hold the enzyme in shape. The shape of the active site changes, so the substrate no longer fits and fewer enzyme-substrate complexes form. At extreme pH the enzyme is denatured. Different enzymes have different optimum pH values.

A denatured enzyme cannot form enzyme-substrate complexes, so it can no longer act as a catalyst.

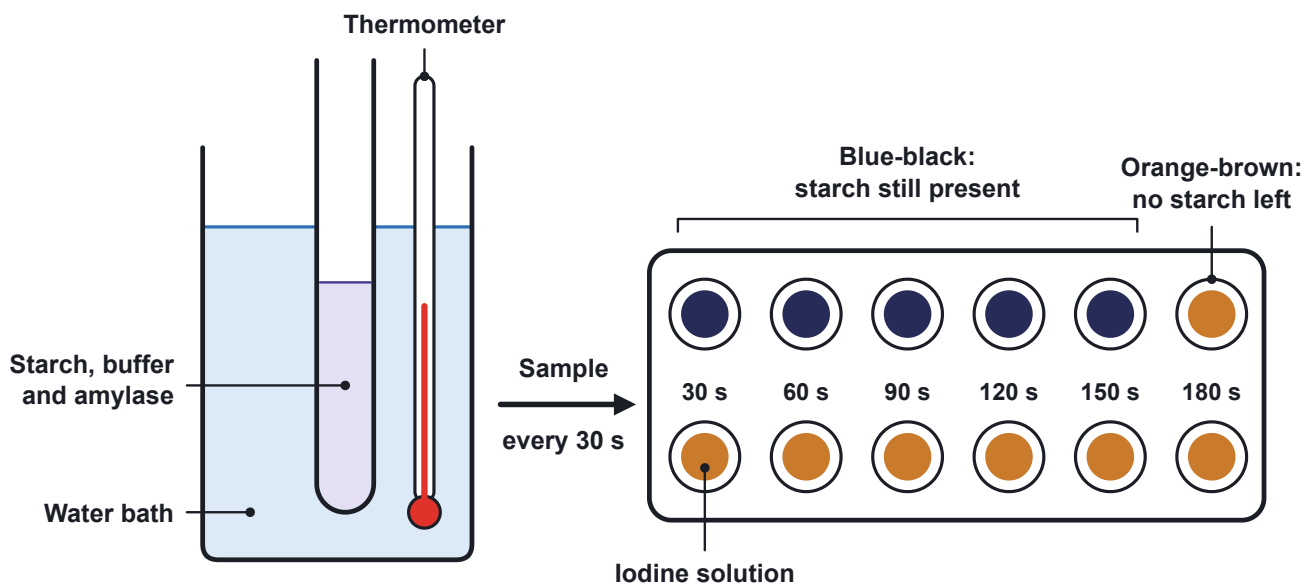
Exam tip: Explanations of temperature need the syllabus ideas: kinetic energy, frequency of effective collisions, shape and fit, and denaturation. Say 'denatured' above the optimum, not 'killed'.

Investigating temperature and pH Spec B5.1.2

You need to be able to investigate and describe the effect of changes in temperature and pH on enzyme activity. A common investigation uses the enzyme amylase, which breaks down starch to maltose. Starch gives a blue-black colour with iodine solution. When all the starch has been broken down, the iodine stays orange-brown. The first sample that stays orange-brown marks the end point.

- Place drops of iodine solution in the wells of a spotting tile.
- Mix starch solution and buffer solution (to set the pH) in a test tube. Put this tube, and a separate tube of amylase solution, in a water bath at the chosen temperature for 5 minutes.
- Add the amylase to the starch, mix and start a stop-clock.

- Every 30 seconds, take a drop of the mixture and add it to a new drop of iodine solution.
- Record the time at which iodine stays orange-brown (no starch left).
- Repeat at different pH values (keeping the temperature constant) or at different temperatures (keeping pH constant).



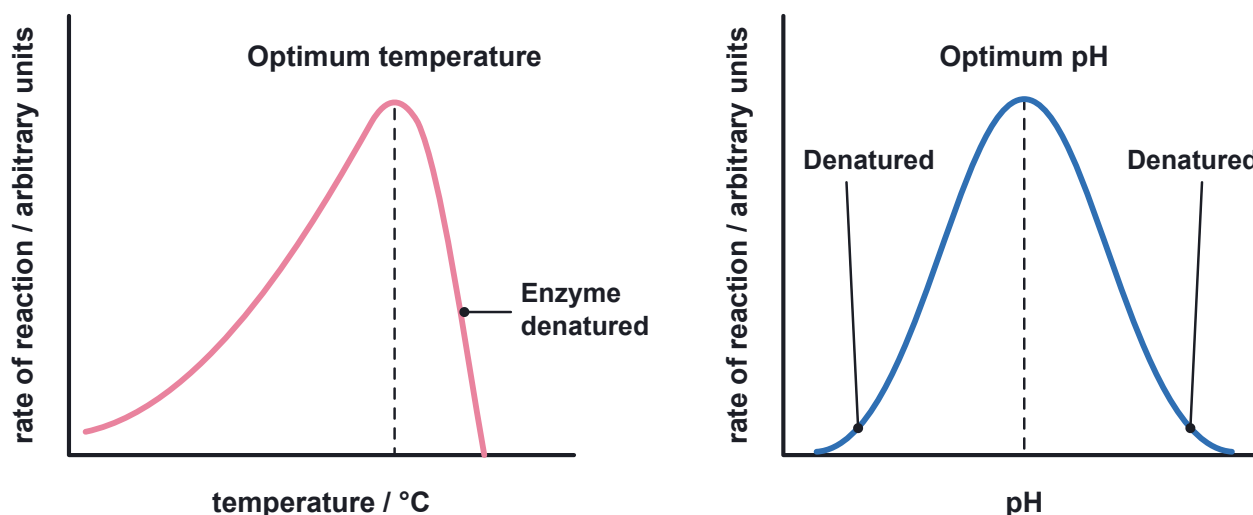
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Testing samples with iodine solution to find when all the starch has gone.

Variables in the investigation		
Variable	Effect of temperature	Effect of pH
Independent (changed)	Temperature of the water bath / °C	pH of the buffer
Dependent (measured)	Time for starch to disappear / s	Time for starch to disappear / s
Controlled (kept the same)	pH, volume and concentration of starch and amylase	Temperature, volume and concentration of starch and amylase

Describing the results

- Temperature:** as the temperature increases, enzyme activity increases up to the **optimum temperature**, where activity is highest. Above the optimum, activity decreases quickly because the enzyme is **denatured**.
- pH:** activity is highest at the optimum pH. Above or below the optimum, activity decreases. At extreme pH the enzyme is denatured.
- When the enzyme is completely denatured, the starch is not broken down at all and every sample stays blue-black.



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Activity is highest at the **optimum**. Above the optimum temperature, or at extreme pH, the enzyme is **denatured**.

Denaturation is a permanent change in the shape of an enzyme's active site, so the substrate no longer fits. A denatured enzyme no longer works, even if the temperature or pH is returned to the optimum.

Common mistake: Low temperature does not denature an enzyme. The enzyme is just slow, and its activity increases again when it is warmed.

Rate of reaction = $1 \div \text{time taken}$, so a shorter time means a faster rate. Because $1 \div \text{time}$ gives very small numbers, questions often use rate = $1000 \div \text{time}$ instead. Plot rate against temperature or pH to find the optimum.

Calculating a rate

Starch was broken down in 50 s at 35 °C. Calculate the rate of reaction as $1000 \div \text{time}$.

1. rate = $1000 \div \text{time}$
2. rate = $1000 \div 50$

Answer: 20 (arbitrary units)

Practical skill: Keep the enzyme and substrate at the water bath temperature before mixing them, and use a thermometer to check the temperature. Use a fresh drop of iodine for each sample, and repeat the experiment to improve reliability. Wear eye protection.

Quick check

1. Define a catalyst.
2. What are enzymes?
3. **Supplement** What word describes the shape of an active site compared with its substrate?
4. **Supplement** What happens to an enzyme above its optimum temperature?
5. Which solution is used to test for starch in the amylase investigation?

Quick check answers

Enzymes

1. A substance that increases the rate of a chemical reaction and is not changed by the reaction.
2. Proteins that function as biological catalysts in metabolic reactions.
3. Complementary.
4. The shape of the active site changes and the enzyme is denatured.
5. Iodine solution.