Exercise walk-through

Factors that affect enzyme action ▪ 3.2

eXercise

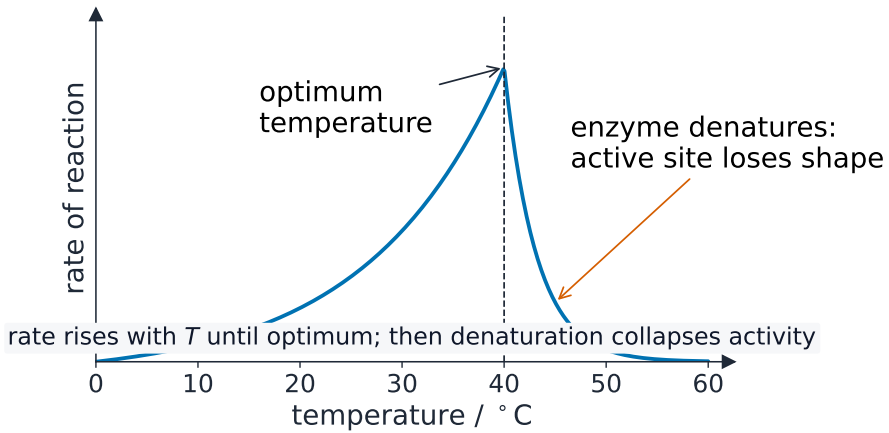
You must be able to

- explain the effect of temperature, pH, enzyme concentration and substrate concentration on rate
- explain V_{max} and K_m and use K_m to compare affinity
- explain competitive and non-competitive inhibition, and state the advantages of immobilised enzymes

Method — Temperature and pH

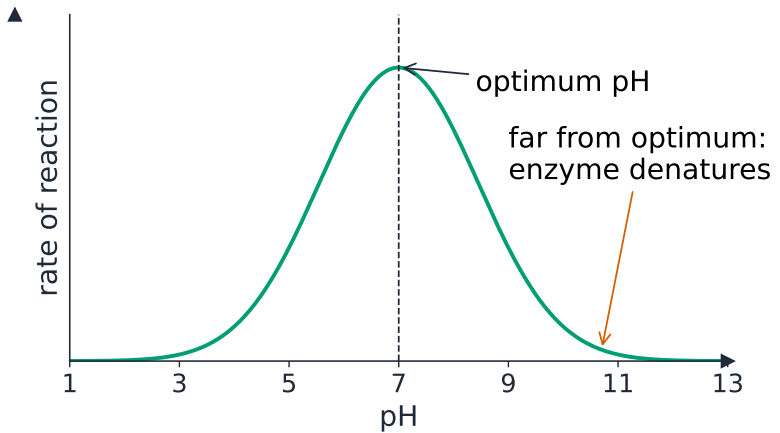
Rate rises with temperature (more **kinetic energy 动能**, more collisions) up to the **optimum**; above it the enzyme **denatures 变性** — bonds holding its shape break, the active site changes, and rate falls fast. Each enzyme also has an optimum pH; far from it the enzyme denatures.

Method — Temperature and pH



A graph of rate against temperature rising to a peak then falling steeply

Method — Temperature and pH



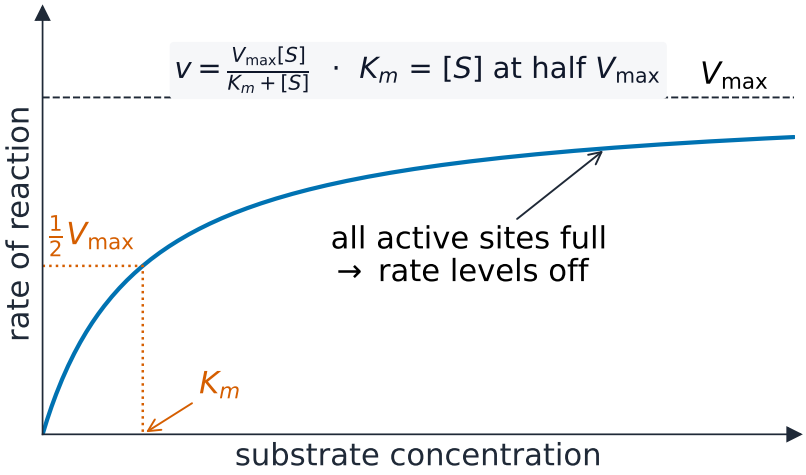
A bell-shaped graph of rate against pH peaking at the optimum

Method — Substrate concentration, V_max and K_m

Adding substrate speeds the reaction until every active site is busy; then rate levels off at the maximum, $V_{\max}$. The **Michaelis–Menten constant** K_{m} is the substrate concentration that gives **half** of $V_{\max}$. A **low** K_{m} = **high affinity 亲和力** (reaches half-speed at low substrate).

K_m is the substrate concentration giving half V_{max} — a lower K_m means higher affinity

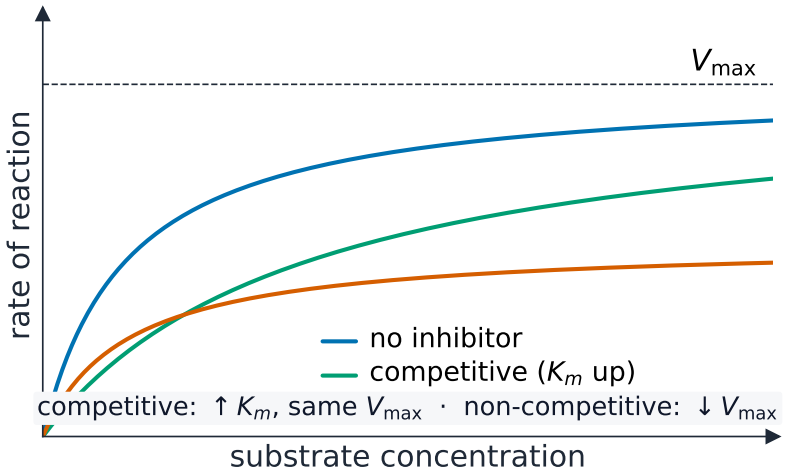
Method — Substrate concentration, V_max and K_m



Method — Reversible inhibitors

- **competitive 竞争性**: binds the active site (similar shape to substrate); more substrate overcomes it $\rightarrow$ V_{max} unchanged, K_m rises.
- **non-competitive 非竞争性**: binds elsewhere, changing the active site; more substrate does not help $\rightarrow$ V_{max} falls.

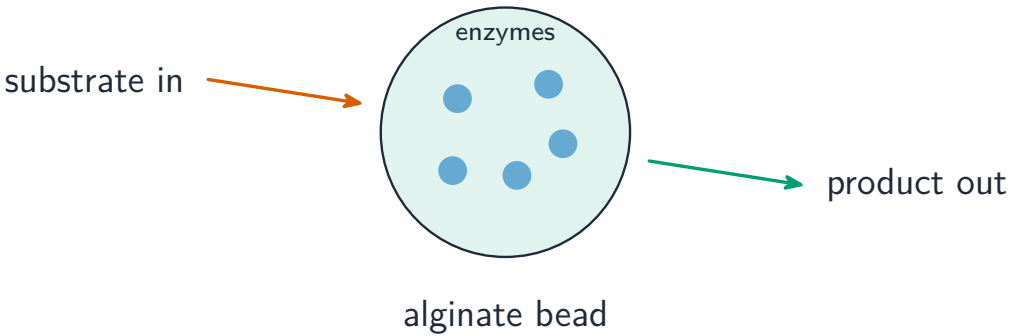
Method — Reversible inhibitors



Method — Immobilised enzymes

An **immobilised enzyme** is fixed in place (e.g. trapped in **alginate** 海藻酸盐 beads). Advantages: re-used many times; pure product; more stable to temperature/pH; can run continuously.

Method — Immobilised enzymes



A bead with substrate flowing in and product flowing out, the enzymes trapped inside

Practice 2.1 ▪ 3 marks

Explain why the rate of an enzyme reaction falls above the optimum temperature.

Solution 2.1

Method: Temperature and pH

Worked steps

heat breaks the bonds (hydrogen/ionic) holding the enzyme's shape **M1**; the active site changes shape / denatures **M1**; so the substrate no longer fits and fewer enzyme–substrate complexes form **A1**.

Practice 2.2 ▪ 2 marks

State **two** advantages of using an immobilised enzyme rather than a free one.

Solution 2.2

Method: Immobilised enzymes

Worked steps

Any two: re-used many times / product is pure (no enzyme) / more stable to temperature and pH / allows continuous flow B1 *each, max 2.*

Exam-style 3.1 ▪ 1 mark

A competitive inhibitor is added to an enzyme reaction. Compared with no inhibitor, what happens to V_{max} and K_m?

A V_{max} unchanged; K_m increases

B V_{max} decreases; K_m unchanged

C both increase

D both decrease

Solution 3.1

Method: Reversible inhibitors

A V_{max} unchanged; K_m increases ✓

B V_{max} decreases; K_m unchanged

C both increase

D both decrease

Worked steps

A. A competitive inhibitor raises K_m but leaves V_{max} unchanged (more substrate overcomes it).

Exam-style 3.2 ▪ 1 mark

Enzyme Y has a lower K_m than enzyme Z for the same substrate. This means that enzyme Y:

A has a higher affinity for the substrate

B has a higher V_{max}

C is denatured more easily

D works at a higher optimum pH

Solution 3.2

A has a higher affinity for the substrate ✓

B has a higher V_{max}

C is denatured more easily

D works at a higher optimum pH

Worked steps

A. A lower K_m means half V_{max} is reached at lower substrate concentration — higher affinity.

Exam-style 3.3 ▪ 2 marks

A student investigates how temperature affects the rate at which catalase breaks down hydrogen peroxide. She measures the volume of oxygen produced in the first minute at five temperatures.

(a) State **two** variables she must control to make this a fair test.

(b) Describe and explain the results she would expect between 10 °C and 60 °C.

(c) Explain why a buffer solution would be used in a similar experiment that varied pH.

Solution 3.3

Worked steps

(a) any two: substrate (hydrogen peroxide) concentration / volume / enzyme concentration / pH / time **B1** *each*.
(b) rate rises from 10 °C towards the optimum as molecules gain kinetic energy and collide more often **M1** **A1**; above the optimum the enzyme denatures, so the rate falls **M1** **A1**.
(c) a buffer keeps pH constant **M1** so that pH is not an uncontrolled variable affecting the rate **A1**.

Exam-style 3.4 ▪ 2 marks

The graph of rate against substrate concentration for an enzyme levels off at high substrate concentration. Explain why.

Solution 3.4

Method: Substrate concentration, V_max and K_m

Worked steps

at high substrate concentration every active site is occupied / saturated M1, so adding more substrate cannot increase the rate further — it is at $V_{\max}$ A1.