

Membranes, Proteins, DNA and Gene Expression

revision notes

Membranes, Proteins, DNA and Gene Expression is topic 2 of Pearson Edexcel International AS / A Level Biology (XBI11 / YBI11) and is examined in Unit 1. It covers gas exchange surfaces and the lung, membrane structure and transport, proteins and enzymes, DNA and protein synthesis, and inheritance, cystic fibrosis and genetic screening. It includes core practical 3 (membrane permeability) and core practical 4 (the initial rate of enzyme-catalysed reactions).

For Pearson Edexcel International AS Biology XBI11 and International A Level Biology YBI11. **A Level only** is IA2 content (Units 4 to 6), not needed for the International AS.

1 Gas exchange, membranes and transport **Spec 2.1-2.5**

Gas exchange surfaces and Fick's law **Spec 2.1**

Every living organism must exchange gases with its environment: oxygen in and carbon dioxide out for aerobic respiration, and the reverse in photosynthesis. The gases cross a **gas exchange surface** by diffusion, so the surface has to be built to make diffusion fast.

A small, single-celled organism has a very large **surface area to volume ratio**, so diffusion across its outer surface is enough. A large organism has a much smaller surface area to volume ratio, so it needs a specialised exchange surface (such as the lungs of a mammal) and a transport system.

Properties of a good gas exchange surface

The three properties and why each speeds up diffusion

Property	Effect on diffusion
Large surface area to volume ratio	More area for molecules to cross at the same time, so more gas is exchanged each second relative to the volume of the organism that needs it.
Thin surface (short diffusion distance)	Molecules have only a short distance to travel, so each crosses more quickly. Exchange surfaces are often one cell thick.
Large difference in concentration (steep concentration gradient)	Diffusion is the net movement down a concentration gradient. The steeper the gradient, the faster the net movement. The gradient is kept steep by removing gas on one side (blood flow) and bringing fresh supplies to the other (ventilation).

Fick's law of diffusion

Fick's law links the rate of diffusion to all three properties at once.

rate of diffusion $\propto$ (surface area $\times$ difference in concentration) $\div$ thickness of the exchange surface

- Doubling the surface area doubles the rate (all else equal).
- Doubling the concentration difference doubles the rate.
- Doubling the thickness of the surface halves the rate.

Using Fick's law to compare two surfaces

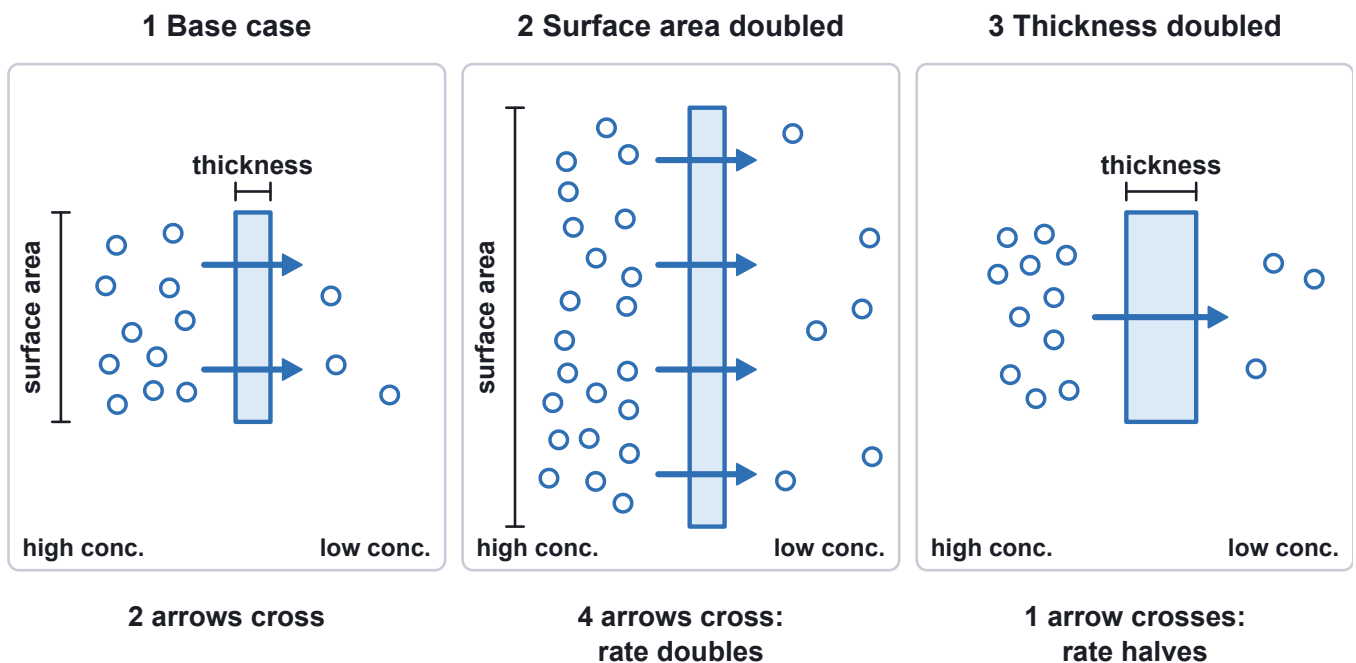
Surface A has an area of 50 mm², a concentration difference of 12 arbitrary units and a thickness of 0.5 µm. Surface B has the same area and concentration difference but is 2.0 µm thick. Calculate the relative rate of diffusion across each and state how many times faster diffusion is across A.

1. Relative rate = (area × concentration difference) ÷ thickness.
2. A: (50 × 12) ÷ 0.5 = 600 ÷ 0.5 = 1200 (arbitrary units).
3. B: (50 × 12) ÷ 2.0 = 600 ÷ 2.0 = 300 (arbitrary units).
4. Compare: 1200 ÷ 300 = 4.

Answer: A: 1200, B: 300. Diffusion is 4 times faster across A, because A is one quarter as thick.

Maths skill: Fick's law questions give you numbers and ask for a rate or a comparison. Use the same units for both surfaces, show the substitution, and give the answer in the units asked for or as a ratio.

Exam tip: Do not write 'large surface area' on its own for an organism. Edexcel asks about surface area to volume ratio, and about thin surfaces and concentration gradients as three separate marking points.



$$\text{rate of diffusion} \propto (\text{surface area} \times \text{concentration difference}) \div \text{thickness}$$

Each blue arrow is one net movement of a gas molecule across the exchange surface.

Fick's law: rate of diffusion $\propto$ (surface area × concentration difference) ÷ thickness. Doubling the area doubles the rate; doubling the thickness halves it.

The mammalian lung Spec 2.1

Air enters the lungs by the **trachea**, which divides into two **bronchi**, one to each lung. Each bronchus branches repeatedly into smaller **bronchioles**, which end in clusters of tiny air sacs called **alveoli**. Gas exchange happens across the walls of the alveoli.

1. Air passes down the trachea, held open by rings of cartilage.
2. It moves into a bronchus, then through the many branching bronchioles.
3. It reaches the alveoli, where oxygen diffuses into the blood and carbon dioxide diffuses out.

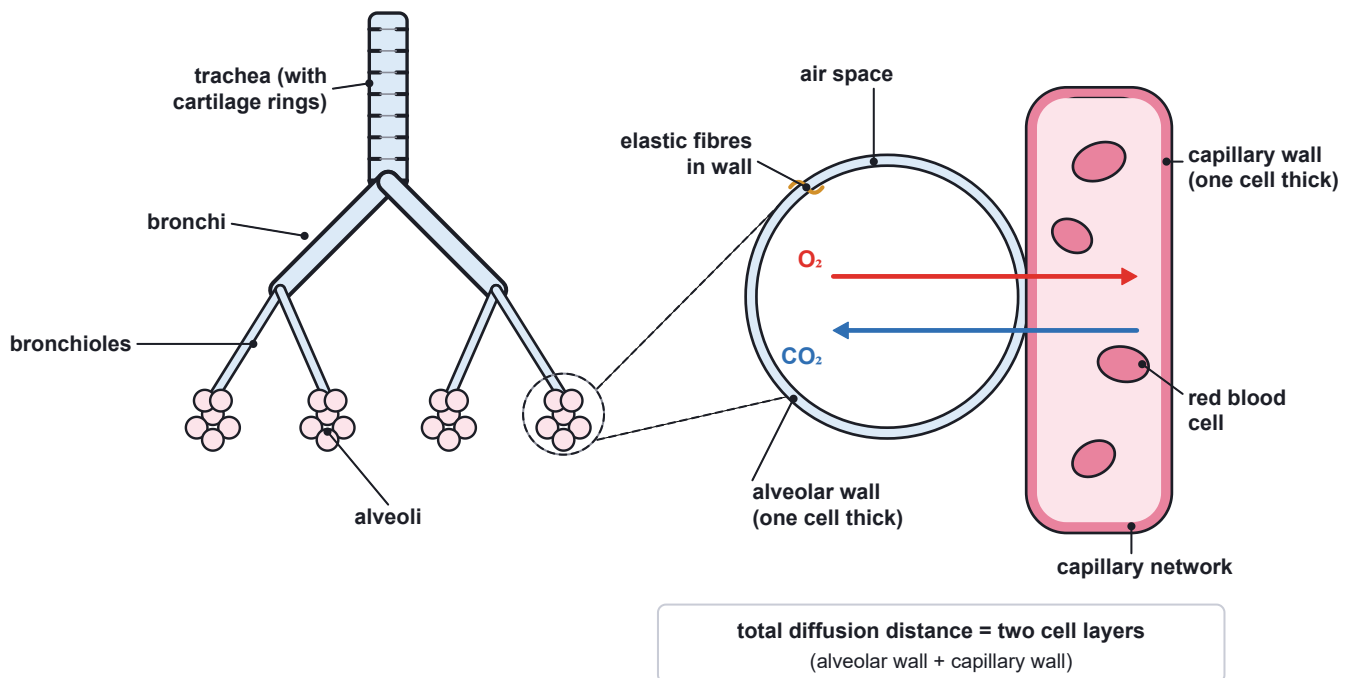
The alveolus and its blood supply

The wall of an alveolus is a single layer of flattened (squamous) epithelial cells. Next to it is the wall of a capillary, which is a single layer of flat endothelial cells. Oxygen therefore diffuses across only two very thin cells (a distance of well under 1 μm in total), from the air in the alveolus into the red blood cells in the capillary.

How the lung is adapted for rapid gas exchange	
Feature	How it helps
About 300 to 500 million alveoli in the two human lungs together (a very large number of small sacs)	Gives a very large surface area to volume ratio for gas exchange.
Alveolar wall and capillary wall each only one cell thick	Short diffusion distance, so diffusion is rapid (Fick's law: thickness is in the denominator).
Dense network of capillaries around each alveolus	Brings a large volume of blood close to the air, and carries oxygen away quickly, which keeps the oxygen concentration gradient steep.
Ventilation (breathing in and out)	Brings in fresh air with a high oxygen concentration and removes air with a high carbon dioxide concentration, maintaining both concentration gradients.
Blood flow through the capillaries	Continually removes oxygenated blood and delivers blood low in oxygen and high in carbon dioxide, maintaining the gradients.
Elastic fibres in the alveolar walls	Stretch as the alveoli fill and recoil when air is breathed out, helping to push air out.
Red blood cells squeezed through narrow capillaries	Slows their flow and brings them close to the capillary wall, giving more time for oxygen to be taken up.

Exam tip: In an 'explain how the lung is adapted' question, link every feature to a consequence for the rate of diffusion. A feature named without its effect ('so diffusion is faster because ...') may not earn the mark.

Common mistake: Ventilation does not make gas diffuse faster by itself. It maintains the concentration gradient by replacing the air in the alveoli.



The gas exchange system, and an enlarged **alveolus** beside its capillary: only **two cell layers** separate the air from the blood.

The structure and properties of cell membranes Spec 2.2

All cells are surrounded by a **cell-surface membrane**, and eukaryotic cells also have membranes around organelles. They all share the same basic structure, which is described by the **fluid mosaic model**.

The phospholipid bilayer

A **phospholipid** has a hydrophilic (water-loving) phosphate head and two hydrophobic (water-hating) fatty acid tails. In water, phospholipids form a **bilayer**: the heads face outwards towards the water inside and outside the cell, and the tails face inwards, away from water. The bilayer is about 7 nm thick. Phospholipids are not fixed in place, so the layer is fluid: they move sideways within their own layer.

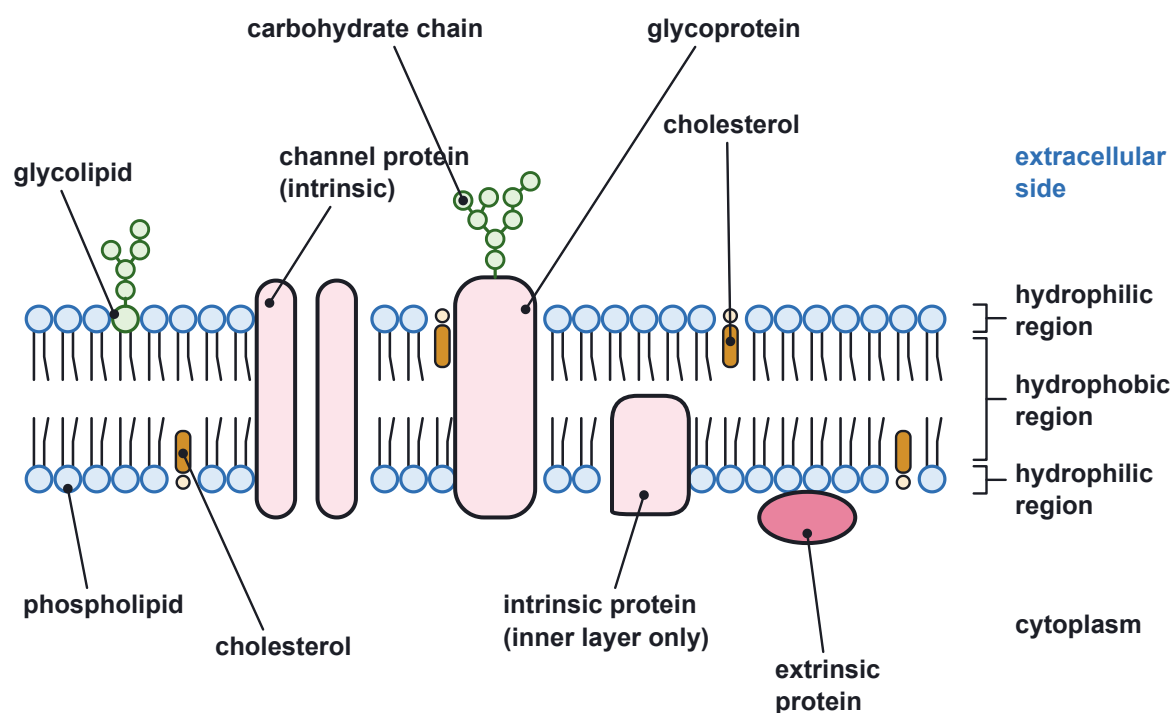
Other components

Components of the membrane and their roles		
Component	Position	Role
Phospholipids	Form the bilayer	Form a barrier to most water-soluble substances; the hydrophobic centre blocks ions and large polar molecules.
Cholesterol	Between the phospholipid tails (animal cells)	Regulates fluidity: it stops the membrane becoming too fluid at higher temperatures and stops the tails packing too closely at lower temperatures. Also makes the membrane more stable.
Intrinsic (integral) proteins	Span the whole bilayer	Form channels and carriers for transport; some are enzymes or receptors.
Extrinsic (peripheral) proteins	On one surface only	Act as enzymes, receptors or recognition sites, or link to the cell's internal framework.
Glycoproteins and glycolipids	On the outer surface, with carbohydrate chains	Act as receptors for hormones and other signals, and as cell recognition sites (antigens).

Properties of the membrane

- **Partially permeable**: small non-polar molecules such as oxygen and carbon dioxide dissolve in the lipid and pass through the bilayer. Water also crosses, in small amounts, by diffusing between phospholipids. Ions and large polar molecules cannot cross the hydrophobic core and need channel or carrier proteins.
- Fluid and flexible: the membrane can change shape and fuse with other membranes (endocytosis and exocytosis).
- Separates the cell from its surroundings and divides the cell into compartments, so that different conditions can be kept in different places.
- Contains receptors and recognition sites, so cells can respond to signals and recognise one another.

Exam tip: Use the word 'hydrophobic' for the tails and 'hydrophilic' for the heads, and say where they face. Use these scientific terms rather than 'water-hating' and 'water-loving' alone.



The **fluid mosaic model** of the cell-surface membrane, about 7 nm thick.

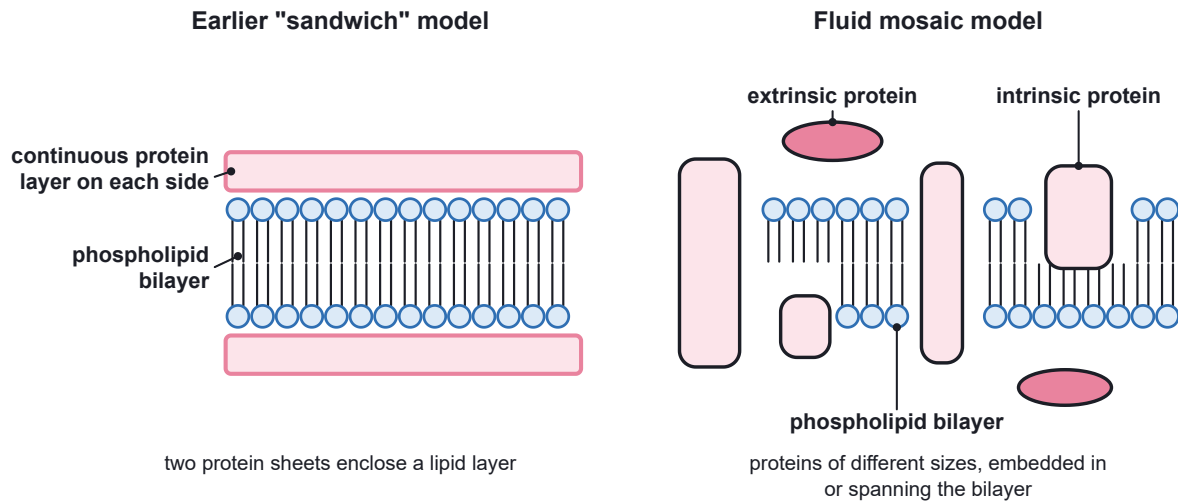
Models of membrane structure and the evidence for them Spec 2.2

Scientists cannot see molecules in a membrane directly. A **model** is an explanation built from the data available. When new data do not fit, the model is changed or replaced. This is how the fluid mosaic model came to be accepted.

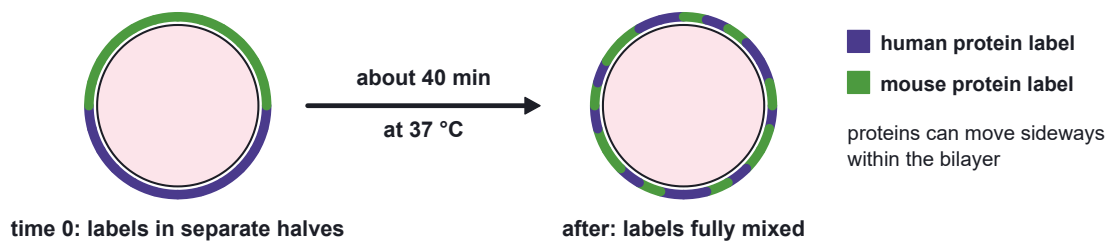
How data have been interpreted		
Evidence	What it showed	How it was interpreted
Early electron micrographs of membranes	Two dark lines with a lighter band between them.	Taken to show a layer of protein on each side of a lipid layer, an early 'sandwich' model with continuous protein layers.
Freeze-fracture electron microscopy	Bumps (protein molecules) scattered within the lipid layers, which split apart between the two layers.	Proteins are embedded within the bilayer, and not in continuous layers on the surface. This supports the mosaic.
Biochemical analysis of membrane proteins	Many proteins have large hydrophobic regions and are not soluble in water.	Hard to explain if all proteins lay on the surface; consistent with proteins spanning the hydrophobic interior.
Cell fusion experiment: a mouse cell and a human cell fused and their surface proteins labelled with different fluorescent markers	At 37 °C the two labels mixed completely over about 40 minutes. At low temperature, mixing was greatly slowed.	Proteins can move sideways in the membrane, so the membrane is fluid. Slower movement at low temperature fits with the lipid becoming more rigid.

The model is named for these two ideas: the bilayer is **fluid** because the phospholipids and many proteins move, and **mosaic** because the proteins are scattered through the bilayer in a pattern like the tiles of a mosaic.

Exam tip: In 'how does the data support the model' questions: first state what the data show (with a value if the data allow), then say what that means about the membrane, then name the feature of the model it supports.



Cell-fusion experiment (human and mouse cells fused, proteins labelled with different fluorescent dyes)



The **sandwich model** against the **fluid mosaic model**. In the cell-fusion experiment the labels mix, so proteins can move within the bilayer.

Core practical 3: membrane permeability, alcohol and temperature Spec 2.3

Core practical: Core practical 3: investigate membrane properties including the effect of alcohol and temperature on membrane permeability.

Beetroot cells contain a red-purple pigment, **betalain**, inside the vacuole. While the vacuole membrane (tonoplast) and the cell-surface membrane are intact, the pigment cannot leave the cell. If the membranes are damaged and become more permeable, pigment leaks out. The more colour in the surrounding liquid, the greater the permeability.

Method (temperature version)

1. Use a cork borer to cut cylinders of beetroot of the same diameter. Cut them to the same length with a scalpel on a white tile. Rinse them in distilled water until the water runs clear, to remove pigment released from cut cells.
2. Set up water baths at a range of temperatures (for example 0, 10, 20, 30, 40, 50, 60, 70 °C). Check each temperature with a thermometer.
3. Put the same volume of distilled water (for example 10 cm³) into each of a set of test tubes, and bring them to temperature in the water baths.
4. Add one beetroot cylinder to each tube and leave for the same time (for example 10 minutes).
5. Remove the cylinders. Shake each tube gently to mix the liquid, and pour it into a cuvette.
6. Measure the absorbance of each liquid in a colorimeter with a blue-green filter, after zeroing it on distilled water. (Or compare colours with a set of colour standards.)
7. Repeat at least three times at each temperature and calculate a mean.

For the alcohol version, keep the temperature constant (for example 20 °C) and instead use a set of ethanol concentrations (for example 0, 10, 20, 30, 40, 50 %), made by diluting ethanol with distilled water to the same total volume.

Variables		
Type	Temperature version	Alcohol version
Independent	Temperature of the water	Concentration of ethanol
Dependent	Absorbance of the liquid (a measure of pigment released)	Absorbance of the liquid
Controlled	Size and shape (surface area) of beetroot piece, same beetroot, volume of liquid, time in liquid, colorimeter filter, rinsing	As temperature version, plus a constant temperature

Risk management

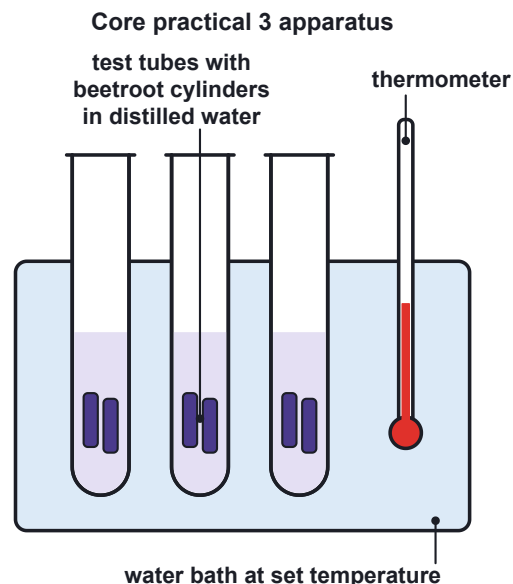
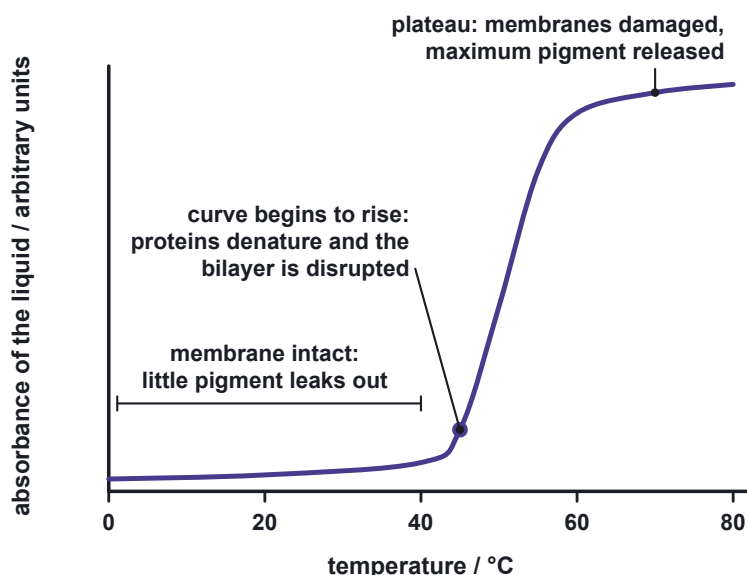
- Ethanol is flammable: keep it away from flames and wear eye protection.
- Hot water can scald: use a thermostatically controlled bath where possible and do not carry hot water across the room.
- Scalpels and cork borers are sharp: cut on a tile, away from the hand holding the beetroot.
- Beetroot juice stains: wear a lab coat.

Processing and explaining results

- Calculate the mean absorbance at each value of the independent variable. Plot a graph with the independent variable on the x axis and mean absorbance on the y axis. Include units.
- Temperature: little leakage at low temperatures (the membrane is intact and its proteins are not damaged). Above about 40 °C the absorbance rises sharply, because proteins in the membrane denature, which opens gaps, and the phospholipids move more, so the bilayer becomes more fluid and leaky.
- Alcohol: ethanol dissolves lipids and disrupts the phospholipid bilayer (and at higher concentrations denatures membrane proteins), so the membrane becomes more permeable and absorbance rises as the concentration rises.
- Compare with a control (beetroot in distilled water at room temperature) to show that the pigment is not just leaking out anyway.

Maths skill: Calculate a mean of repeats, and ignore anomalous results (explain why you did). When comparing treatments, you can work out the percentage change in absorbance relative to the control: $(\text{value} - \text{control}) \div \text{control} \times 100$.

Common mistake: Do not say the membrane 'melts' or that the pigment 'breaks down'. Say that the proteins denature and the phospholipid bilayer is disrupted, so permeability increases.



after a fixed time, measure the absorbance of the liquid with a colorimeter

Core practical 3. **Illustrative data:** above about 40 °C the membranes are damaged and more pigment leaks out, until the curve plateaus.

Osmosis and water potential Spec 2.4

Water potential (symbol ψ , the Greek letter psi) is a measure of the tendency of water molecules to move from one place to another. It is measured in **kilopascals (kPa)**. Pure water has the highest water potential, which is defined as zero. Dissolved solutes attach to some of the water molecules, so fewer water molecules are **free** to move. Adding solute lowers the water potential, so the water potential of any solution is negative.

Osmosis is the net movement of free water molecules through a partially permeable membrane, down a water potential gradient. Water molecules move in both directions across the membrane, but more move from the region of higher (less negative) water potential to the region of lower (more negative) water potential. Osmosis is a passive process and needs no ATP.

What happens to cells in solutions of different water potential

Solution compared with the cell	Net movement of water	Animal cell (no cell wall)	Plant cell (cellulose cell wall)
Higher water potential (more dilute, such as pure water)	Into the cell	Swells and may burst (lysis).	Swells until the wall resists: turgid.
Same water potential	No net movement	Stays the same.	Stays the same.
Lower water potential (more concentrated)	Out of the cell	Shrinks and the surface looks shrivelled (crenation).	Cytoplasm shrinks and pulls away from the cell wall: plasmolysed.

Recommended practical: tissue water potential using plant tissue

The specification recommends an investigation of tissue water potentials using plant tissue and graded concentrations of a solute. Cylinders of potato are cut to the same size, blotted, weighed, left in sucrose solutions of different concentrations for the same time, then blotted and weighed again. In dilute solutions the cylinders gain mass; in concentrated solutions they lose mass.

$$\text{percentage change in mass} = (\text{final mass} - \text{initial mass}) \div \text{initial mass} \times 100$$

Percentage change in mass

A potato cylinder has a mass of 2.40 g before and 2.70 g after 24 hours in a sucrose solution. Calculate the percentage change in mass.

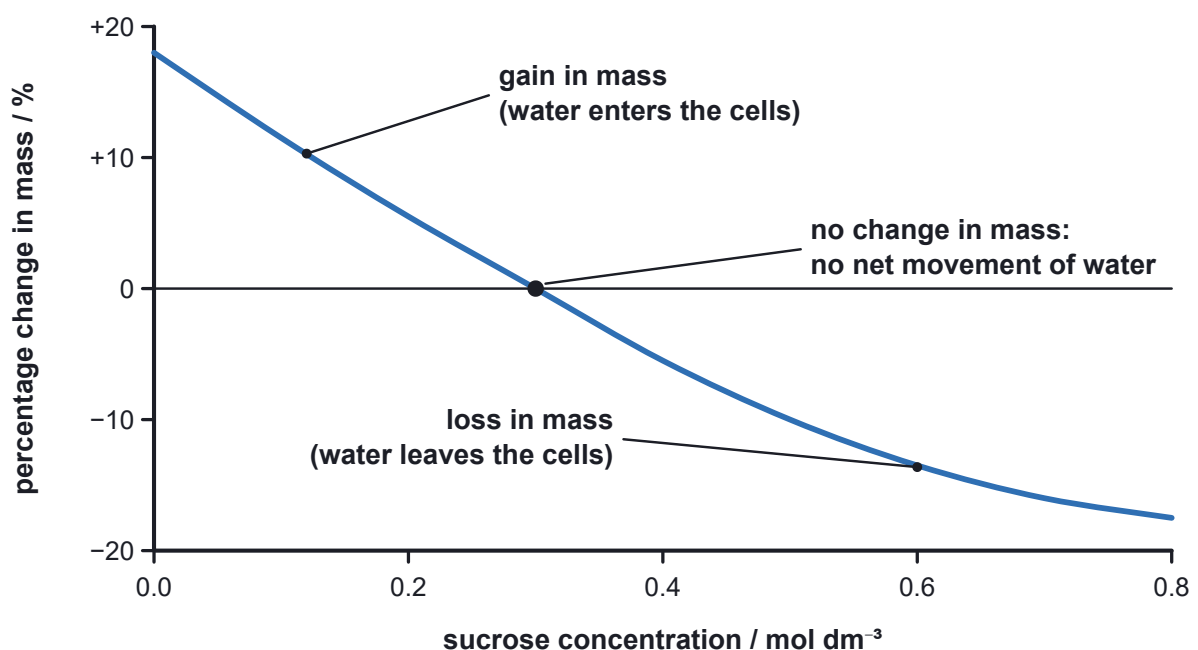
1. Change in mass = $2.70 - 2.40 = +0.30$ g.
2. Divide by the initial mass: $0.30 \div 2.40 = 0.125$.
3. Multiply by 100: 12.5 %.

Answer: +12.5 % (an increase, so water entered by osmosis).

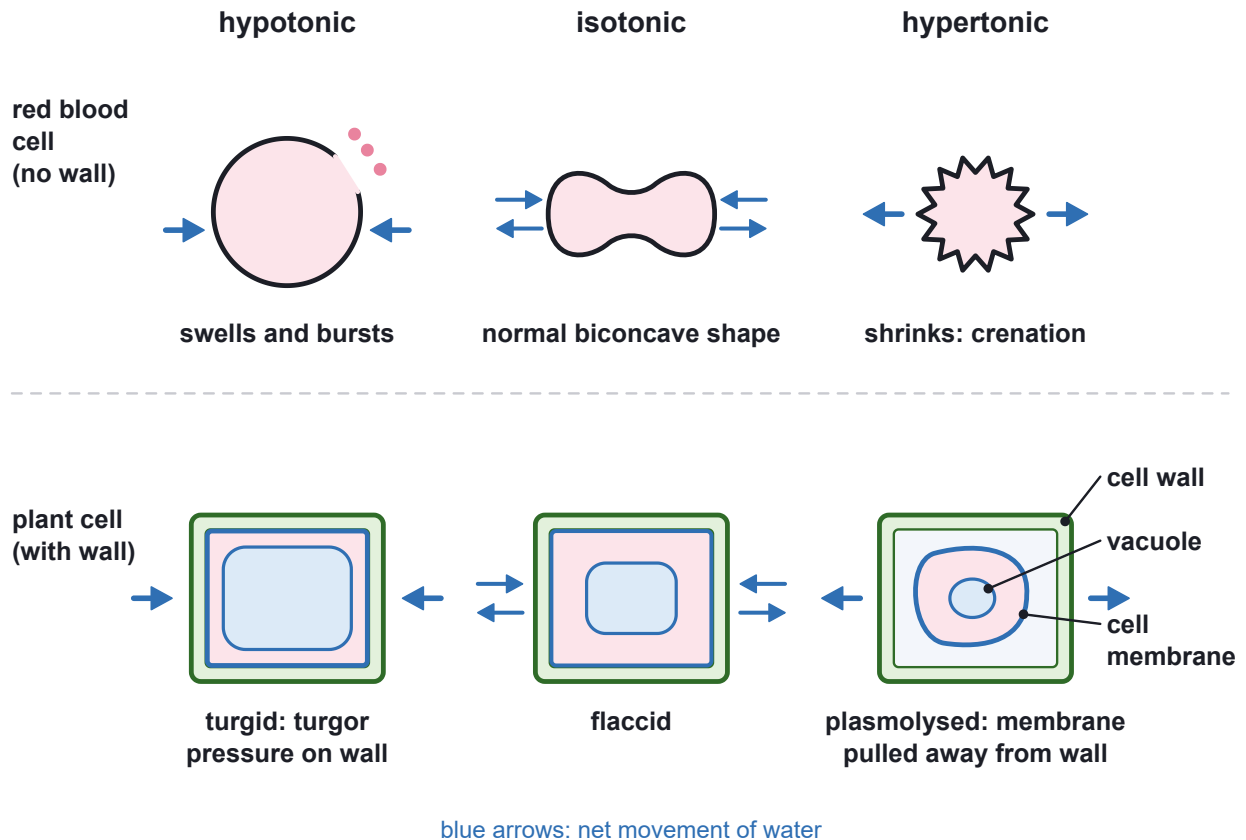
Plot percentage change in mass (y axis) against sucrose concentration (x axis). Where the line crosses zero, there is no net movement of water, so the water potential of the potato tissue equals that of the solution. Read off the concentration at the intercept, then use a calibration table to find the water potential.

Practical skill: Control the size of the cylinders, the volume of solution, the time, the temperature and the way the pieces are blotted. Use percentage change rather than change in mass, so that pieces of slightly different starting mass can be compared.

Common mistake: Do not say water moves 'from a high concentration of water to a low concentration of water'. Say 'from a higher (less negative) water potential to a lower (more negative) water potential'. And keep the sign: -200 kPa is a higher water potential than -500 kPa.



Where the line crosses zero, **no net movement of water** occurs: the water potential of the tissue equals that of the solution.
Illustrative data.



Plant cells in different solutions: **turgid**, flaccid and **plasmolysed**.

Passive transport: diffusion, facilitated diffusion, channel and carrier proteins

Spec 2.5

Passive transport is the movement of substances across a membrane without the cell supplying metabolic energy (no ATP is used). The substance moves down its concentration (or electrochemical) gradient, using the kinetic energy of its own molecules. There are two kinds: diffusion and facilitated diffusion.

Diffusion

Diffusion is the net movement of molecules or ions from a region of higher concentration to a region of lower concentration, down a concentration gradient. Small non-polar molecules such as oxygen and carbon dioxide diffuse directly through the phospholipid bilayer.

Facilitated diffusion

Ions and polar molecules (such as glucose) cannot cross the hydrophobic centre of the bilayer.

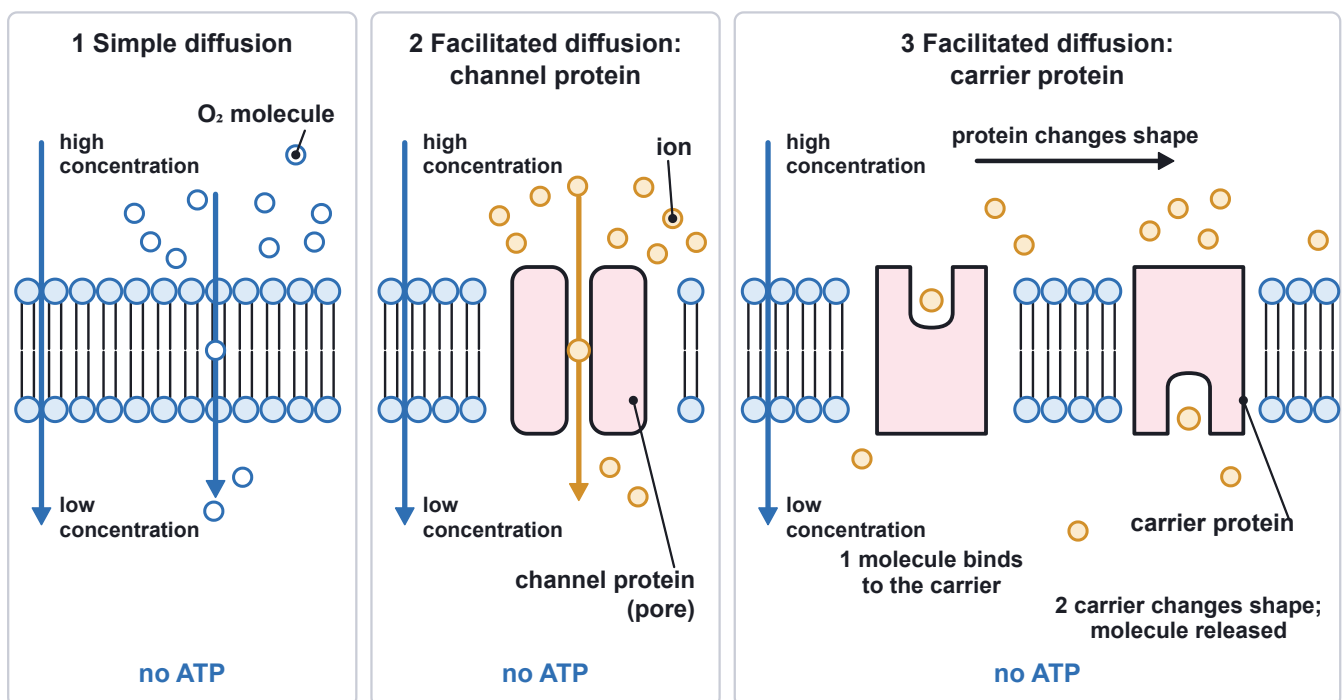
Facilitated diffusion is passive transport through the membrane with the help of transport proteins. There are two types of protein.

Channel proteins compared with carrier proteins

Feature	Channel protein	Carrier protein
Structure	Water-filled pore through the membrane, lined with hydrophilic amino acids; often gated (opens and closes).	Has a binding site specific to one molecule or ion, on one side of the membrane.
How it works	Ions or small polar molecules of the right size and charge pass through the pore.	The molecule binds, the protein changes shape (conformation) and releases the molecule on the other side.
Specificity	Specific to particular ions by pore size and charge.	Specific to a particular molecule by the shape of the binding site.
Energy	Passive: no ATP.	Passive in facilitated diffusion: no ATP.

The rate of facilitated diffusion rises with the concentration gradient, but only up to a point. When all the carrier proteins are working at their maximum rate (saturated), adding more of the substance does not increase the rate. The rate also depends on the number of transport proteins in the membrane.

Exam tip: 'Facilitated diffusion' still needs a concentration gradient and still uses no ATP. Say 'down a concentration gradient through a channel or carrier protein' to earn the marks.



Passive transport: all three move down a concentration gradient and use no ATP.

Active transport, endocytosis and exocytosis Spec 2.5

Active transport

Active transport is the movement of molecules or ions across a membrane against a concentration gradient, using energy from ATP and a carrier protein. **ATP** is the immediate source of energy for the cell. It is hydrolysed to ADP and inorganic phosphate (P_i), which releases energy.

1. The molecule or ion to be moved binds to a specific site on a carrier protein.
2. ATP binds to the carrier protein and is hydrolysed to ADP + P_i , which releases energy.
3. The protein changes shape and releases the molecule on the other side of the membrane, against the gradient.

4. The protein returns to its original shape, ready to bind again.

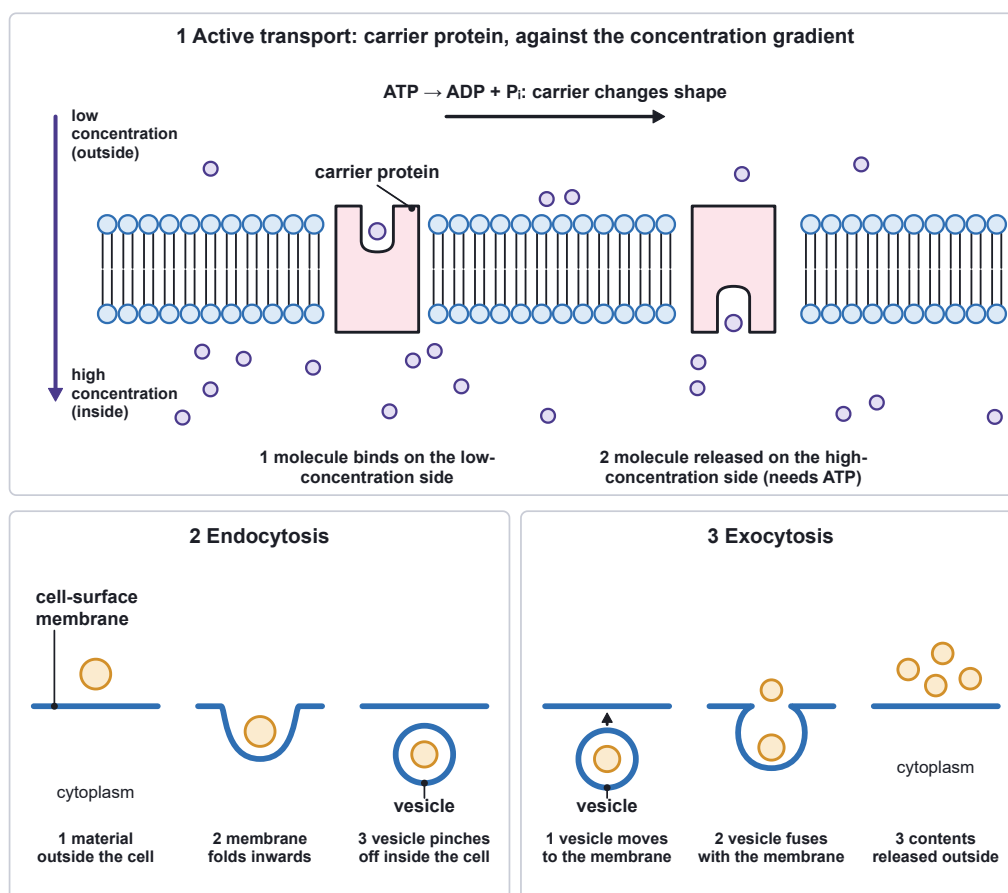
Endocytosis and exocytosis

- **Endocytosis** is the bulk transport of material into a cell. A region of the cell-surface membrane folds inwards around the material and pinches off to form a vesicle inside the cell. Cells use it to take in large particles or fluid droplets; ATP is needed.
- **Exocytosis** is the bulk transport of material out of a cell. A vesicle (often from the Golgi apparatus) moves to the cell-surface membrane, fuses with it and releases its contents outside. Cells use it to secrete substances such as enzymes and hormones; ATP is needed.

Comparing the transport processes			
Feature	Diffusion	Facilitated diffusion	Active transport
Direction relative to gradient	Down	Down	Against
ATP needed	No	No	Yes
Protein needed	No	Yes (channel or carrier)	Yes (carrier)
Specific	No	Yes	Yes

Exam tip: A good mark point for active transport is 'ATP is hydrolysed to ADP and P_i, which releases energy'. An answer that only says 'uses energy' is not enough. Cells with a lot of active transport have many mitochondria.

Common mistake: Endocytosis and exocytosis are not diffusion or osmosis, and they do not use channel or carrier proteins. They move bulk material in vesicles and they need ATP.



Active transport needs ATP; **endocytosis** and **exocytosis** move bulk material in vesicles and also need ATP.

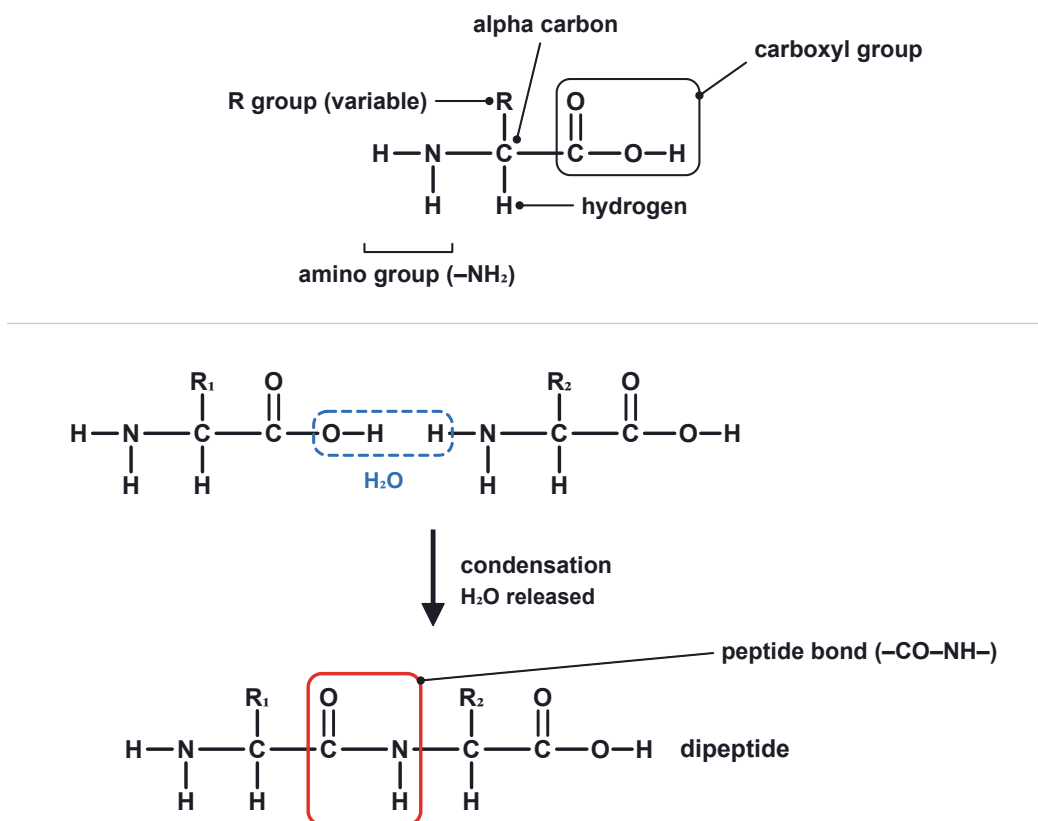
Quick check

- 1 . State the three properties of a gas exchange surface.
- 2 . What happens to the rate of diffusion if the thickness of the exchange surface doubles?
- 3 . Why can oxygen cross the phospholipid bilayer, but sodium ions cannot?
- 4 . Define osmosis.
- 5 . What is the role of ATP in active transport?

2 Proteins and enzymes Spec 2.6-2.8

Amino acids, peptide bonds and polypeptides Spec 2.6

Proteins are polymers made from **amino acid** monomers. All amino acids share the same basic structure around a central carbon atom: an **amino group** ($-\text{NH}_2$), a **carboxyl group** ($-\text{COOH}$), a hydrogen atom, and a variable **R group**. About 20 different amino acids are used to make proteins, and they differ only in their R group. (You do not need to learn the structures of individual amino acids.)



A **peptide bond** forms in a **condensation** reaction between the carboxyl group of one amino acid and the amino group of the next.

Condensation and hydrolysis

Two amino acids join in a **condensation reaction**. The carboxyl group of one amino acid reacts with the amino group of the next. A molecule of water is released, and a covalent **peptide bond** ($\text{C}-\text{N}$) forms between them, to give a **dipeptide**. Repeating this builds a chain of many amino acids, called a **polypeptide**. The reverse reaction, hydrolysis, uses water to break peptide bonds.

amino acid + amino acid $\rightarrow$ dipeptide + water (condensation, forms a peptide bond)

A **protein** is made of one or more polypeptide chains, folded into a precise three-dimensional shape. The sequence of amino acids in each chain is determined by the sequence of bases in a gene.

Practical skill: Recommended additional practical: use a semi-quantitative method to estimate protein concentration with biuret reagent and colour standards. Add sodium hydroxide, then copper(II) sulfate. A purple (lilac) colour shows peptide bonds; the more protein, the deeper the colour. Compare with standards of known concentration or use a colorimeter. Take care: sodium hydroxide is corrosive, so wear eye protection.

Common mistake: The peptide bond is between the carbon of one amino acid's carboxyl group and the nitrogen of the next amino group. A condensation reaction releases water, and does not use it up.

Levels of protein structure and the bonds involved Spec 2.6

Levels of protein structure		
Level	Description	Bonds involved
Primary	The sequence of amino acids in the polypeptide chain.	Peptide bonds (covalent).
Secondary	Regular coiling or folding of the chain: the alpha helix and the beta pleated sheet.	Hydrogen bonds between the C=O and N-H groups of the polypeptide backbone, between amino acids near each other.
Tertiary	Further folding of the whole chain into a specific three-dimensional shape.	Hydrogen bonds, ionic bonds, disulfide bonds (covalent, between sulfur atoms of cysteine residues), and hydrophobic interactions between non-polar R groups.
Quaternary	Two or more polypeptide chains (and sometimes a non-protein group) together in one protein.	The same types of bond and interaction between the chains.

The **primary structure** decides everything else. The sequence of R groups determines where hydrogen bonds, ionic bonds and disulfide bonds can form, so it determines how the chain folds, and therefore the final shape and properties of the protein. Changing even one amino acid can change the shape, and so the function.

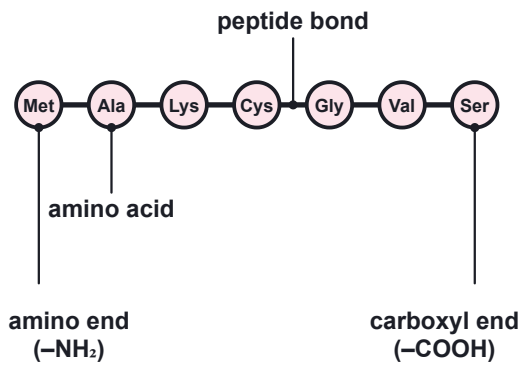
The bonds in the tertiary structure

- **Hydrogen bonds**: weak, but numerous. They form between polar R groups. They are broken by changes in temperature and pH.
- **Ionic bonds**: between positively and negatively charged R groups (for example -NH_3^+ and -COO^-). Stronger than hydrogen bonds but broken by changes in pH.
- **Disulfide bonds**: covalent bonds between the sulfur atoms of two cysteine R groups. Strong, and not easily broken.
- Hydrophobic interactions: non-polar R groups cluster in the centre of a globular protein, away from water.

Exam tip: Do not say the primary structure is 'held together by hydrogen bonds'. Peptide bonds hold the primary structure; hydrogen bonds hold the secondary structure and help hold the tertiary structure.

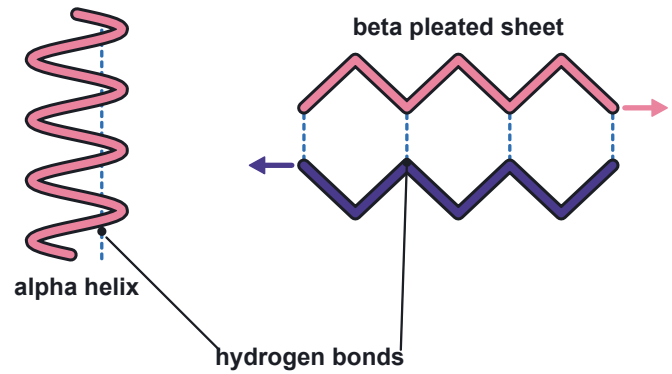
1 Primary structure

order of amino acids in a polypeptide



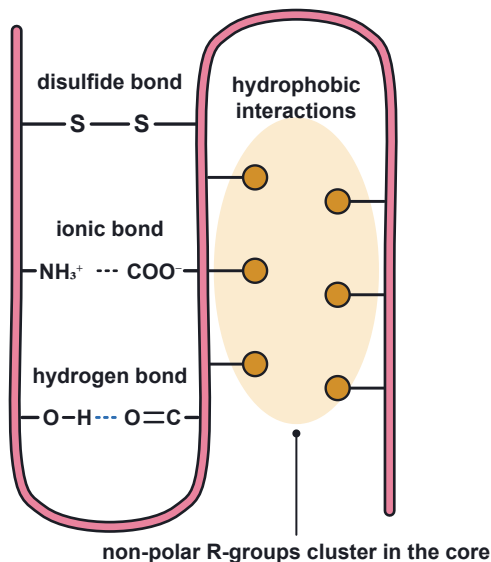
2 Secondary structure

folding of the chain held by hydrogen bonds



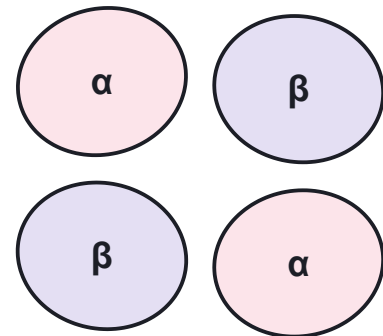
Primary structure is the amino acid sequence; **secondary structure** is the alpha helix or beta pleated sheet held by **hydrogen bonds**.

3 Tertiary structure



4 Quaternary structure

more than one polypeptide chain



four polypeptide chains packed together (e.g. haemoglobin: two α and two β)

held together by the same types of bond as in the tertiary structure

Tertiary structure is held by disulfide bonds, ionic bonds, hydrogen bonds and hydrophobic interactions; **quaternary structure** is more than one chain.

Globular and fibrous proteins: haemoglobin and collagen Spec 2.6

Globular and fibrous proteins compared

Feature	Globular (for example haemoglobin)	Fibrous (for example collagen)
Shape	Compact, rounded, folded.	Long, rope-like fibres.
Solubility	Soluble in water: hydrophilic R groups face outwards, hydrophobic R groups face inwards.	Insoluble: many hydrophobic R groups on the outside.
Structure	Complex tertiary (and often quaternary) structure.	Repetitive amino acid sequence; little tertiary folding; chains wound together.
Role	Metabolic: transport, enzymes, hormones, antibodies.	Structural: strength and support.

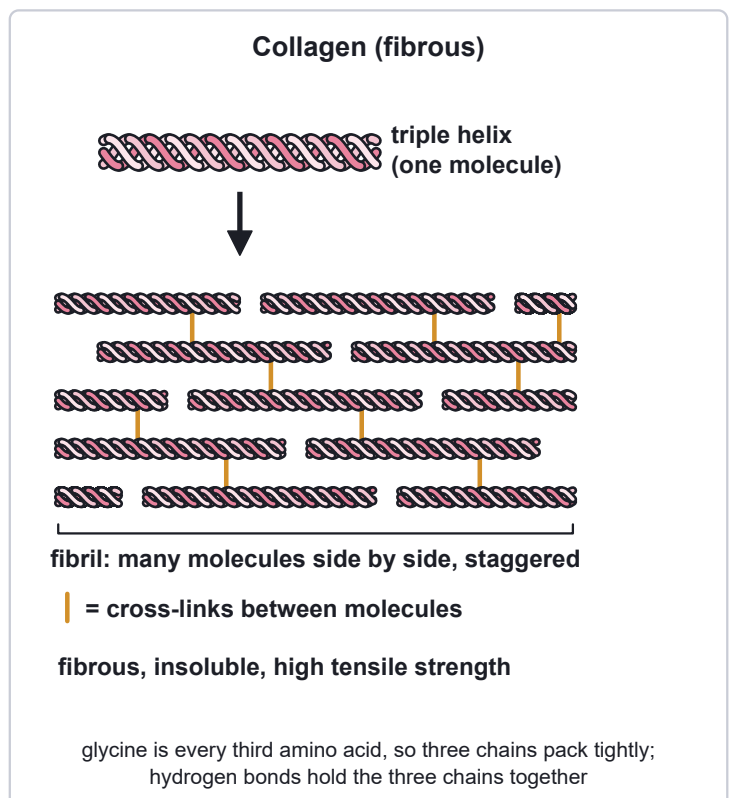
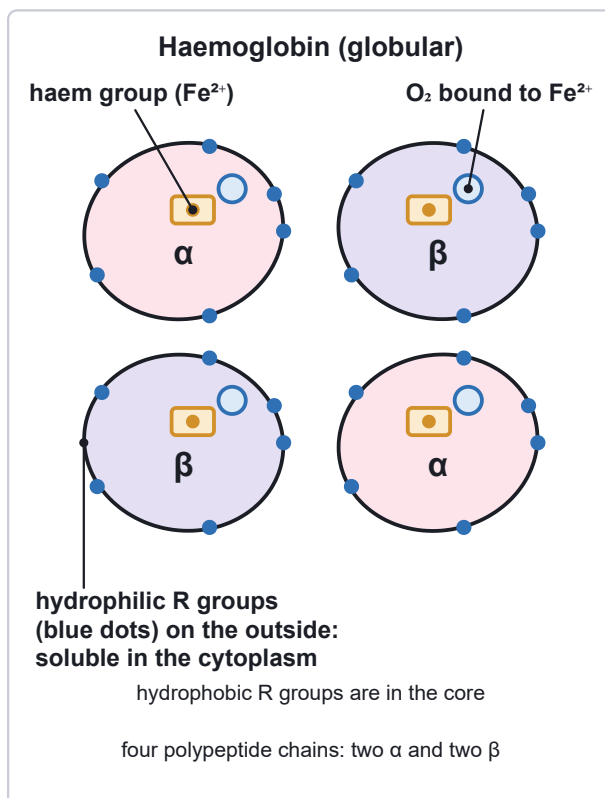
Haemoglobin

Haemoglobin is a globular protein with quaternary structure: four polypeptide chains (two alpha and two beta chains). Each chain is folded around a **haem group** that contains an iron ion (Fe^{2+}), which binds one oxygen molecule, so each haemoglobin molecule can carry four oxygen molecules. The hydrophilic R groups on the outside make haemoglobin soluble in the cytoplasm of red blood cells, and the precise shape allows it to bind and release oxygen.

Collagen

Collagen is a fibrous protein found in tendons, skin, bones, the walls of arteries and cartilage. Three polypeptide chains, each a left-handed helix, are wound together into a right-handed triple helix. Every third amino acid is glycine, the smallest amino acid, which allows the three chains to pack tightly together. The chains are held together by many hydrogen bonds, and neighbouring collagen molecules are linked by covalent cross-links, making fibrils and fibres. This gives collagen a very high tensile strength and makes it insoluble, which suits its structural function.

Exam tip: Always link structure to function: 'haemoglobin is soluble because hydrophilic R groups are on the outside, so it can be carried in the cytoplasm of red blood cells'; 'collagen has high tensile strength because of hydrogen bonds between three chains and cross-links between molecules'.



Haemoglobin is globular and soluble; **collagen** is fibrous with high tensile strength.

Enzymes: mode of action and specificity Spec 2.7

An **enzyme** is a globular protein that acts as a **biological catalyst**: it speeds up a chemical reaction without being used up. It does this by lowering the **activation energy**, the energy needed to start the reaction. The enzyme is not changed overall and can be used again.

The active site and specificity

An enzyme has an **active site**, a small region of the molecule that has a particular three-dimensional shape and charge pattern, set by the enzyme's tertiary structure. The substrate has a complementary

shape and binds to the active site, forming an **enzyme-substrate complex**. The products leave and the enzyme can bind another substrate.

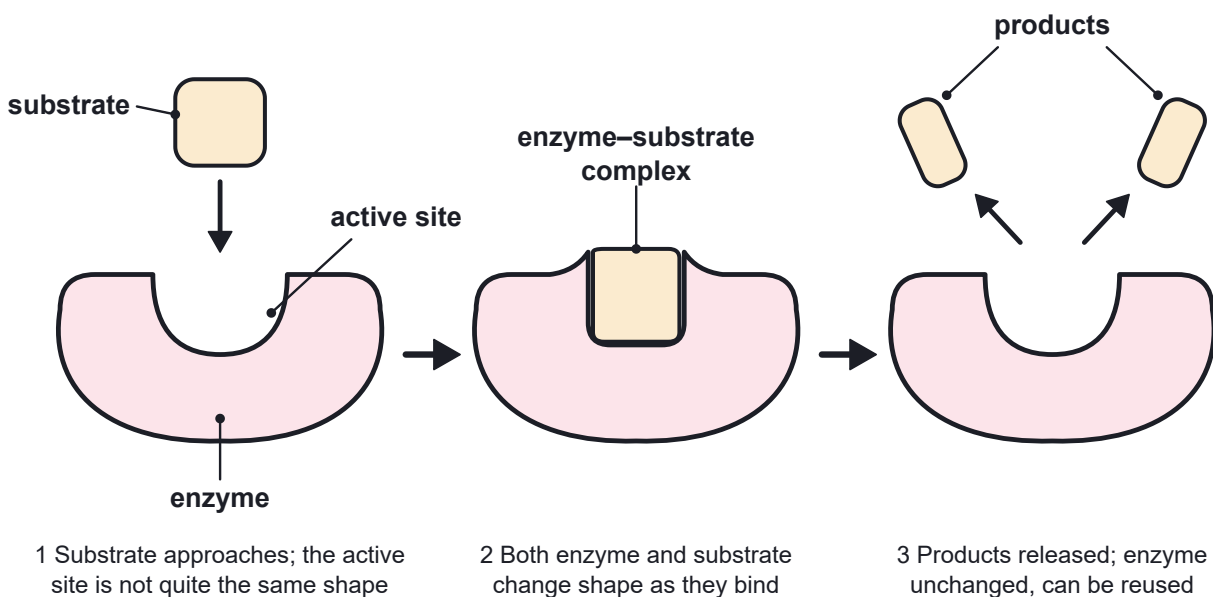
1. The substrate collides with the active site and binds, forming an enzyme-substrate complex.
2. The active site changes shape slightly to fit the substrate more closely (induced fit), which puts strain on bonds in the substrate.
3. This lowers the activation energy, so the substrate is converted to products.
4. The products leave the active site, which returns to its original shape.

Enzymes are **specific**: the shape of the active site only fits a particular substrate (or a small group of similar substrates). A different substrate cannot bind, so only one reaction is catalysed. The tertiary structure is determined by the primary structure, so a change in amino acid sequence can alter the active site and affect the enzyme's activity.

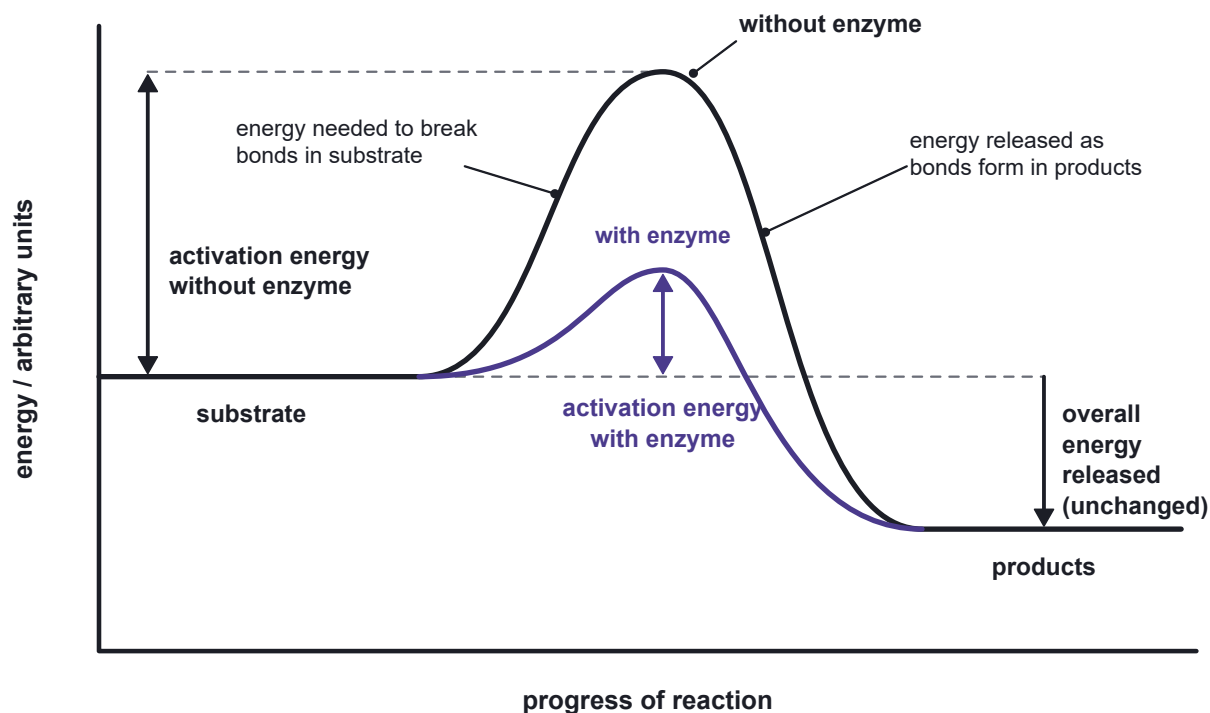
Intracellular and extracellular enzymes

- **Intracellular enzymes** catalyse reactions inside cells, for example catalase, which breaks down hydrogen peroxide inside cells, and the enzymes of respiration in the mitochondria.
- **Extracellular enzymes** are made in cells and secreted to catalyse reactions outside the cells, for example amylase and trypsin, which are secreted into the digestive system to digest starch and protein.

Exam tip: 'Lowers the activation energy' is the key mark point for how an enzyme works. Say that the substrate fits the active site because of its complementary shape, and that the shape is due to the tertiary structure.



The **enzyme-substrate complex**: the substrate fits the active site, and the enzyme is unchanged afterwards.



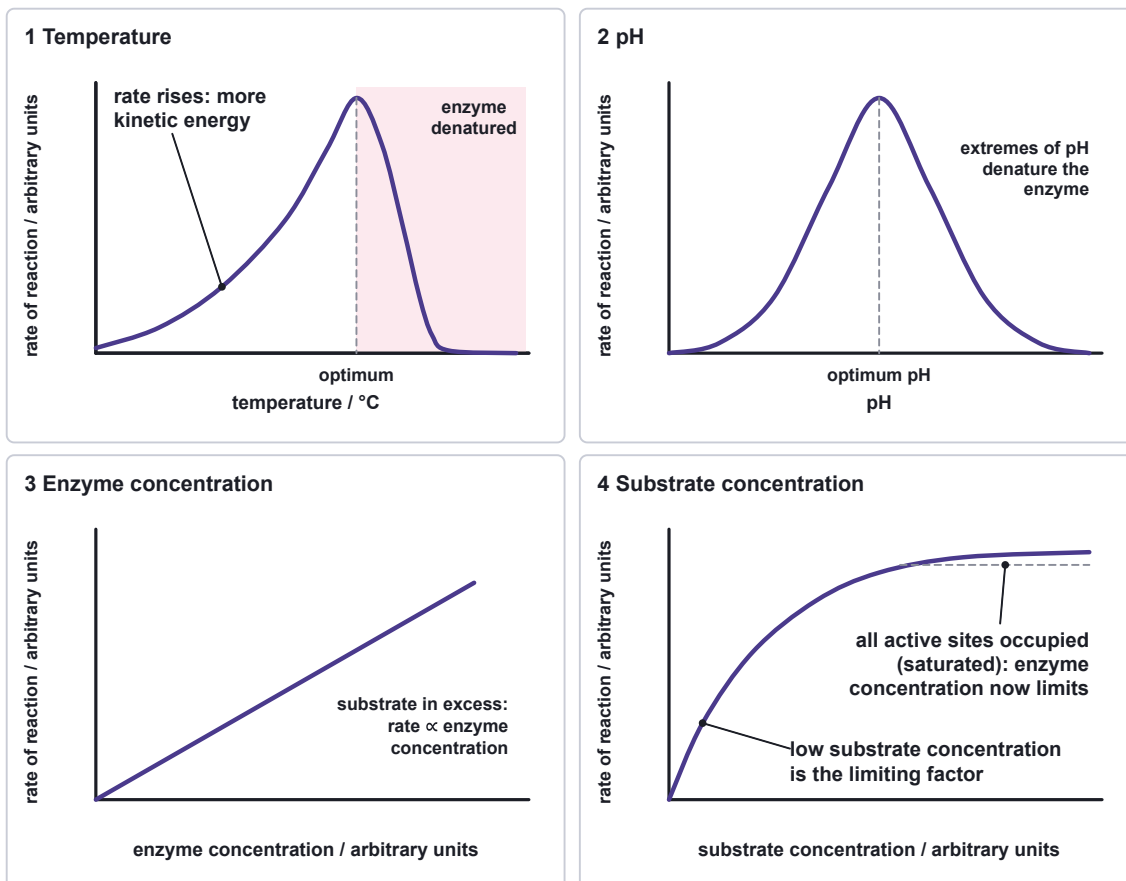
Enzymes **lower the activation energy** of a reaction.

Factors affecting the rate of enzyme-catalysed reactions Spec 2.8

The **initial rate** of reaction is the rate at the very start, when the substrate concentration is highest and little product has built up. Measuring the initial rate makes comparisons fair, because the rate falls later as substrate is used up.

Effect of each factor		
Factor	Effect on rate	Explanation
Temperature	Rate rises to an optimum, then falls rapidly.	Below the optimum, molecules have less kinetic energy, so there are fewer collisions and fewer enzyme-substrate complexes form. Above the optimum, hydrogen and ionic bonds in the tertiary structure break, the active site changes shape and the enzyme is denatured, and substrate no longer fits.
pH	Rate is highest at the optimum pH and falls on either side.	Changes in H^+ concentration change the charges on R groups and disrupt ionic and hydrogen bonds, so the active site changes shape and fewer enzyme-substrate complexes form. Far from the optimum pH the enzyme is denatured.
Enzyme concentration	Rate is proportional to enzyme concentration, if substrate is in excess.	More active sites are available, so more enzyme-substrate complexes form per second. At high enzyme concentration, substrate becomes limiting and the rate levels off.
Substrate concentration	Rate rises, then levels off at a maximum.	More substrate gives more collisions with active sites. When all the active sites are occupied (saturated), enzyme concentration is the limiting factor and rate stops rising.

Common mistake: Enzymes at low temperature are inactive, not dead: the active site is not changed and activity returns on warming. Above the optimum, denaturation is permanent. Do not say enzymes are 'killed' or 'destroyed', because enzymes are not alive.



Factors affecting the rate of an enzyme-controlled reaction. **Illustrative curves.**

Core practical 4: the initial rate of enzyme-catalysed reactions Spec 2.8

Core practical: Core practical 4: investigate the effect of temperature, pH, enzyme concentration and substrate concentration on the initial rate of enzyme-catalysed reactions.

A suitable system is catalase breaking down hydrogen peroxide into water and oxygen. The oxygen is collected, and the volume is measured over time. (Other systems also work, for example amylase and starch, where the time for the iodine colour to disappear is measured, or the breakdown of milk protein by a protease.)

1. Place a fixed volume of hydrogen peroxide solution, adjusted to the chosen pH with a buffer, in a boiling tube and bring it to the chosen temperature in a water bath.
2. Add a fixed volume of catalase solution (for example from yeast or liver) and immediately connect a delivery tube to a gas syringe or an upturned measuring cylinder over water.
3. Start a stopwatch. Record the volume of oxygen at regular intervals (for example every 10 seconds) for several minutes.
4. Repeat at least three times for each value of the independent variable, and calculate a mean.

Variables for each version of the investigation

Independent variable	How to change it	Keep constant
Temperature	Water baths at, for example, 10, 20, 30, 40, 50 and 60 °C.	pH (buffer), enzyme concentration and volume, substrate concentration and volume.
pH	Buffer solutions of different pH, for example pH 3 to 10.	Temperature, enzyme and substrate concentrations and volumes.
Enzyme concentration	Dilute the enzyme stock to give a range of concentrations (serial dilution).	Temperature, pH, substrate concentration and volume.
Substrate concentration	Dilute hydrogen peroxide stock to a range of concentrations.	Temperature, pH, enzyme concentration and volume.

Risk management

- Hydrogen peroxide is an irritant (and stronger solutions are corrosive): wear eye protection and gloves, and wash splashes off skin.
- Do not use a very high concentration of enzyme with high hydrogen peroxide, because the gas is evolved quickly: use a gas syringe or a measuring cylinder that is big enough.
- Hot water baths can scald.

Finding the initial rate

Plot the volume of product (y axis) against time (x axis). The curve is steepest at the start, then levels off as substrate is used up. Draw a **tangent** to the curve at time zero, and calculate its gradient. The gradient is the initial rate.

initial rate = change in volume of product ÷ change in time (gradient of the tangent at t = 0)

Initial rate from a graph

A tangent drawn to the curve at time zero passes through the points (0 s, 0 cm³) and (20 s, 14 cm³). Calculate the initial rate of the reaction.

1. Initial rate = change in volume ÷ change in time.
2. = (14 – 0) cm³ ÷ (20 – 0) s.
3. = 0.70 cm³ s⁻¹.

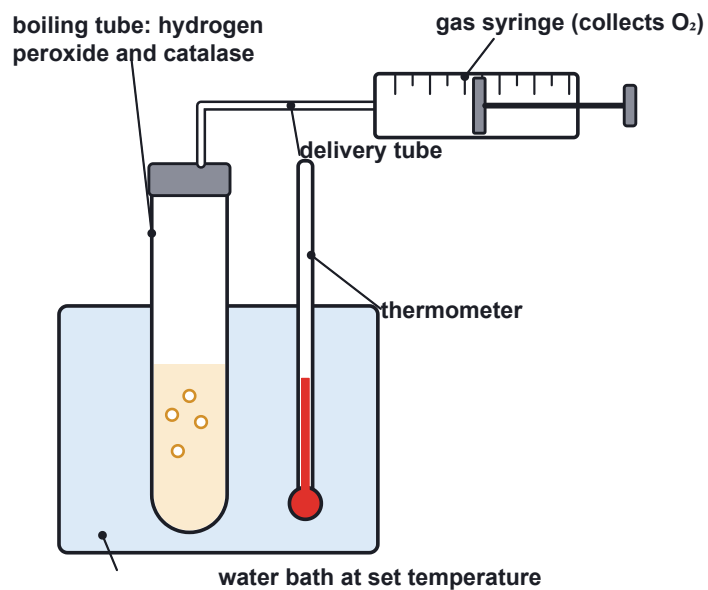
Answer: 0.70 cm³ s⁻¹ (the unit is cm³ per second).

Maths skill: Use a large triangle on the tangent to find the gradient. If the rate is taken from a data table instead, use the first, steepest interval. Give the unit (for example cm³ s⁻¹ or mm³ min⁻¹).

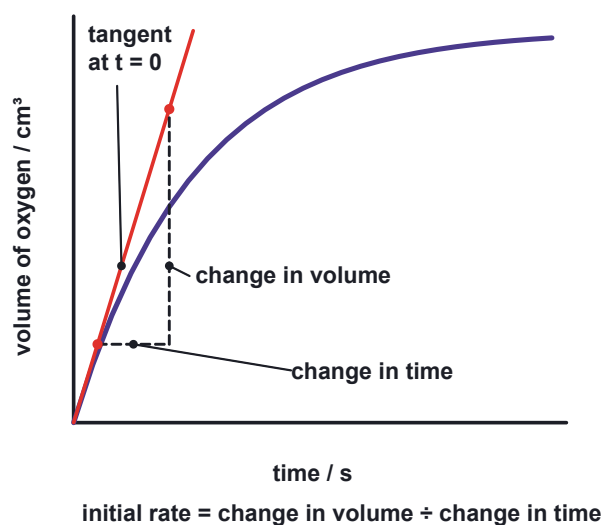
Percentage change is also used to compare rates between treatments: (new – original) ÷ original × 100.

Exam tip: In an evaluation, enzymes at different temperatures must be brought to temperature before they are mixed. Otherwise the reaction starts at the wrong temperature and the initial rate is not valid.

Core practical 4 apparatus



Initial rate from the graph



Core practical 4: find the **initial rate** from the **gradient of a tangent** at $t = 0$. Illustrative graph.

Quick check

1. Name the bond that links two amino acids.
2. Which bonds hold the tertiary structure of a protein?
3. Why is haemoglobin soluble?
4. What does an enzyme do to the activation energy of a reaction?
5. How is the initial rate of reaction found from a graph?

3 DNA, the genetic code and gene expression Spec 2.9-2.14

The structure of mononucleotides, DNA and RNA Spec 2.9

A **mononucleotide** has three parts: a pentose sugar, a phosphate group and a nitrogen-containing base. In DNA the sugar is **deoxyribose**; in RNA it is **ribose**. DNA has four bases: adenine (A), thymine (T), cytosine (C) and guanine (G). In RNA, thymine is replaced by **uracil (U)**.

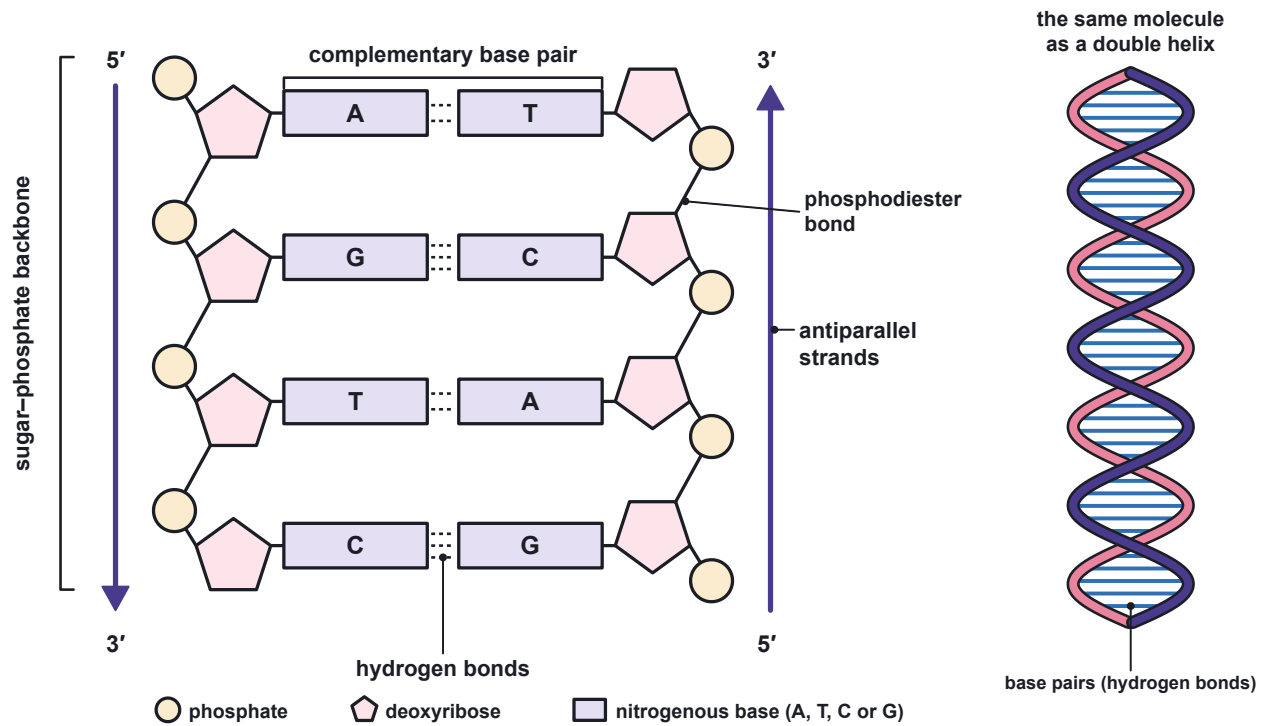
Mononucleotides join in condensation reactions. A **phosphodiester bond** forms between the phosphate group of one nucleotide and the sugar of the next, releasing water. Many nucleotides in a chain make a **polynucleotide**, with a sugar-phosphate backbone and bases sticking out to the side.

The DNA double helix

- A DNA molecule is made of two polynucleotide strands wound into a double helix.
- The strands are **antiparallel**: they run in opposite directions (one runs 5' to 3', the other 3' to 5').
- **Complementary base pairing**: adenine pairs with thymine (two hydrogen bonds) and cytosine pairs with guanine (three hydrogen bonds). The base pairs lie across the middle of the helix.
- Many **hydrogen bonds** hold the two strands together. Each bond is weak, so the strands can be separated for replication and transcription, but together they make DNA stable.
- The sugar-phosphate backbones protect the bases on the inside.

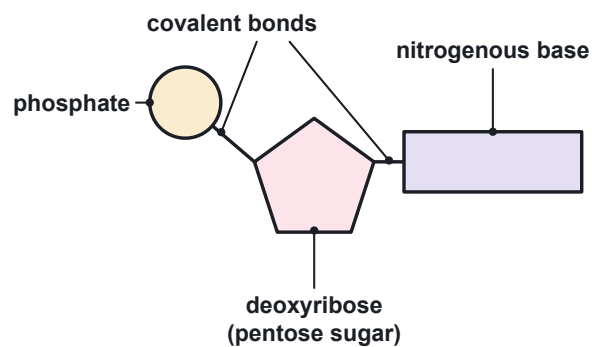
Comparing DNA and RNA		
Feature	DNA	RNA
Sugar	Deoxyribose	Ribose
Bases	A, T, C, G	A, U, C, G
Strands	Two polynucleotide strands, double helix	One polynucleotide strand
Size	Very long	Relatively short
Types	—	Messenger RNA (mRNA), transfer RNA (tRNA), ribosomal RNA (rRNA)

Common mistake: Phosphodiester bonds join nucleotides along one strand; hydrogen bonds join bases between two strands. Do not mix them up, and do not say hydrogen bonds link A with G.

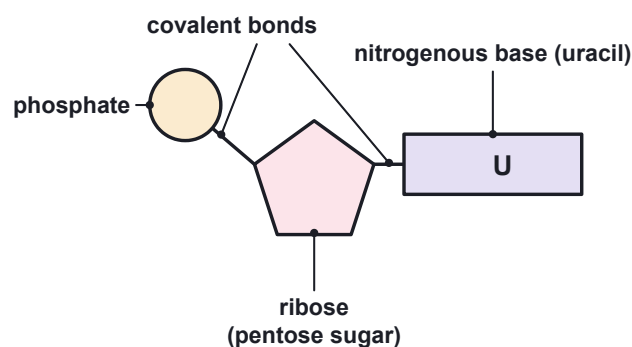


The **double helix**: antiparallel strands joined by **hydrogen bonds** between complementary bases.

DNA mononucleotide



RNA mononucleotide



A DNA nucleotide contains **deoxyribose**; an RNA nucleotide contains **ribose** and the base **uracil**.

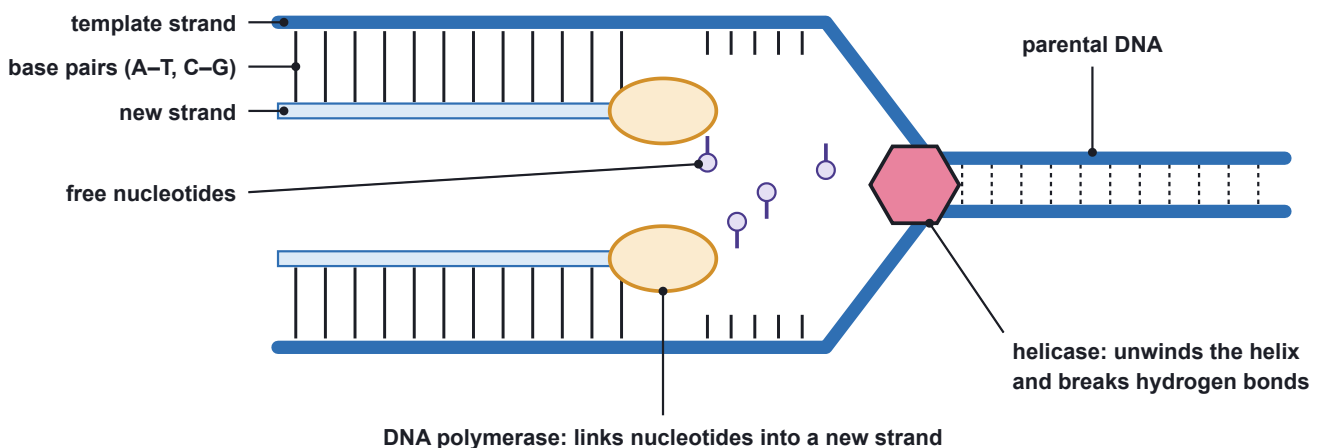
DNA replication and the role of DNA polymerase Spec 2.10

Before a cell divides, its DNA must be copied. In **semi-conservative replication**, each of the two new DNA molecules is made of one original (old) strand and one newly synthesised strand.

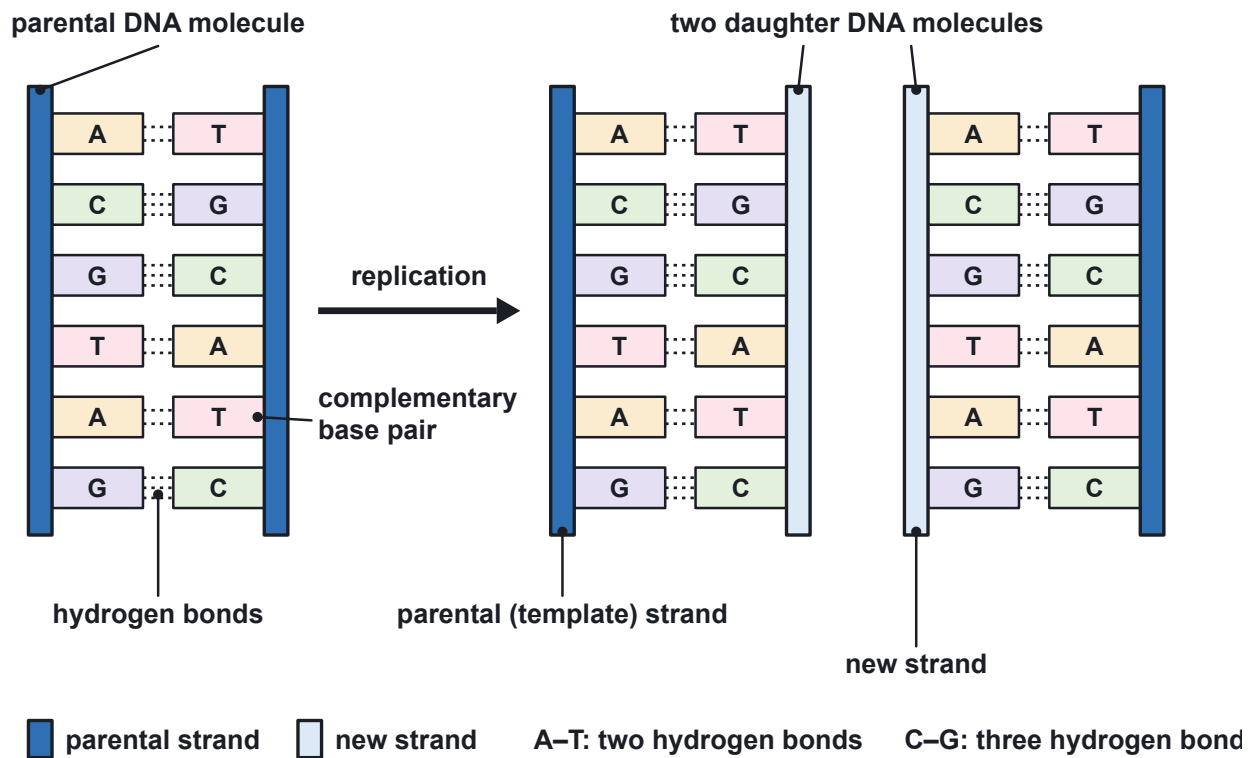
1. The enzyme DNA helicase unwinds the double helix and breaks the hydrogen bonds between the complementary bases, exposing the bases of the two template strands.
2. Free DNA nucleotides in the nucleus pair up with their complementary bases on each template strand: A with T, C with G.
3. **DNA polymerase** joins the nucleotides next to each other, forming phosphodiester bonds between them, which makes a new sugar-phosphate backbone. It adds nucleotides to the growing strand in one direction only.
4. Hydrogen bonds form between the bases of the template and new strands, and each pair twists into a double helix.
5. Two identical DNA molecules result, each with one old strand and one new strand.

The base sequence of the new strand is determined by the template strand, so the two new molecules have identical base sequences to the parent molecule (unless a mistake occurs, see gene mutations). This ensures that daughter cells receive the same genetic information.

Exam tip: A full-mark replication answer names DNA helicase (unwinding, breaking hydrogen bonds), complementary base pairing, DNA polymerase (joining nucleotides, forming phosphodiester bonds) and says 'semi-conservative'.



At the replication fork, **DNA helicase** unwinds the helix and **DNA polymerase** joins nucleotides to the new strand.



Semi-conservative: each daughter molecule keeps one old strand and one new strand.

The Meselson-Stahl experiment Spec 2.10

Three models were proposed for how DNA could be copied. In the **conservative** model the original molecule stays intact and a completely new molecule is made. In the **semi-conservative** model each new molecule has one old and one new strand. In the **dispersive** model, each strand is a mixture of old and new DNA.

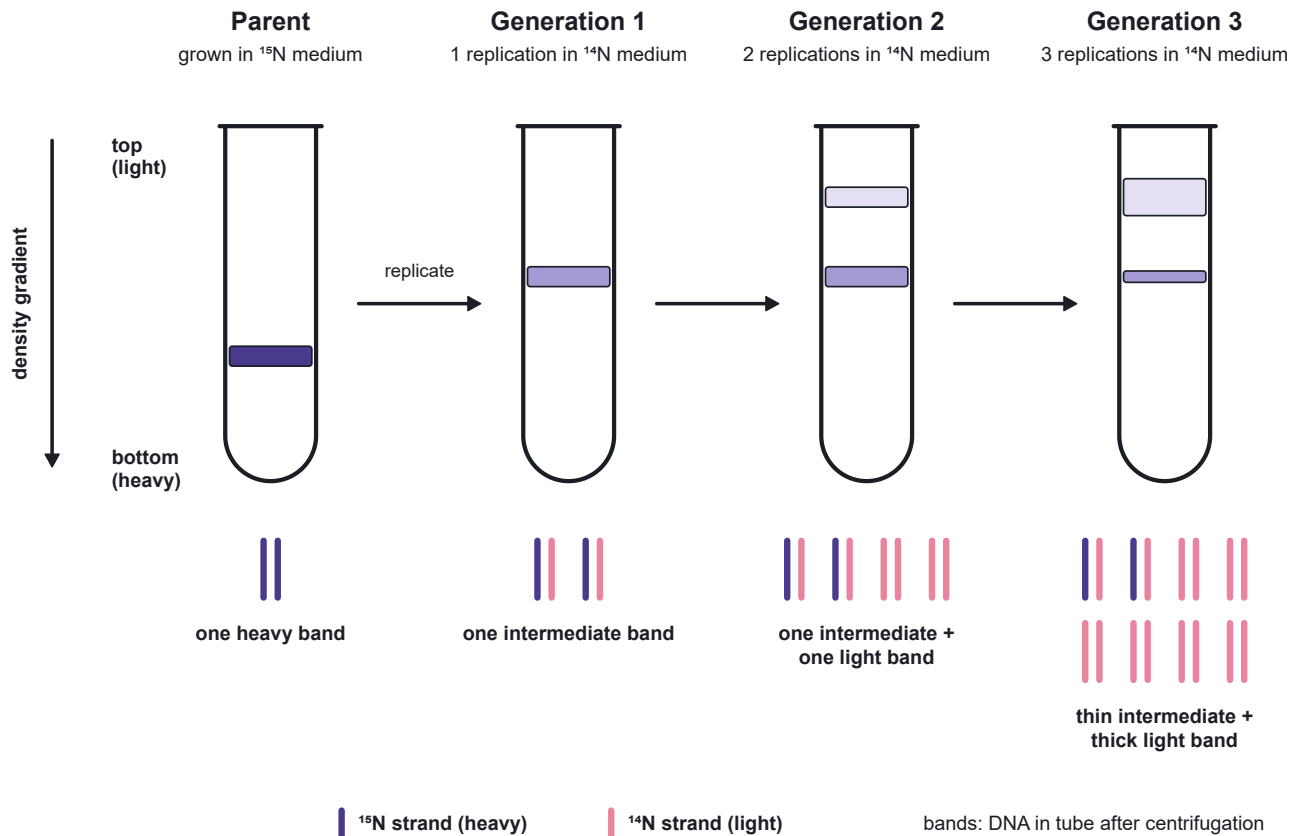
The experiment

1. Bacteria (*E. coli*) were grown for many generations in a medium with heavy nitrogen, ^{15}N , so all their DNA contained ^{15}N (heavy DNA).
2. The bacteria were transferred to a medium containing only normal, lighter nitrogen, ^{14}N , and allowed to divide. New DNA could only be made using ^{14}N .
3. Samples were taken after each generation. The DNA was extracted and spun in a centrifuge in a caesium chloride solution, which separates DNA by density into bands.

Results and what each model predicted				
Sample	Result observed	Conservative model	Semi-conservative model	Dispersive model
Start (all ^{15}N)	One band, heavy	One heavy band	One heavy band	One heavy band
After 1 generation	One band, intermediate density	Two bands: heavy and light (not matching)	One intermediate band (matches)	One intermediate band (matches)
After 2 generations	Two bands: intermediate and light	Heavy and light bands (not matching)	Intermediate and light bands (matches)	One band, lighter than intermediate (not matching)

After one generation, a single band of intermediate density rules out the conservative model, which would give separate heavy and light bands. After two generations, the presence of both an intermediate and a light band rules out the dispersive model, which would give a single band of lower density. Only the semi-conservative model fits both results. The new data therefore **supported** the accepted theory and **refuted** the competing theories.

Exam tip: In a data question, state what the band positions mean in terms of nitrogen (heavy = both strands ^{15}N , intermediate = one ^{15}N strand and one ^{14}N strand, light = both strands ^{14}N), then compare with each model's prediction.



The **Meselson-Stahl** results support **semi-conservative replication**.

The genetic code and the gene Spec 2.11, 2.12

A **gene** is a sequence of bases on a DNA molecule that codes for a sequence of amino acids in a polypeptide chain. The order of bases in the gene therefore determines the order of amino acids in the polypeptide, and so the protein's structure and function.

Features of the genetic code

- Triplet code:** a sequence of three bases (a triplet in DNA; a **codon** in mRNA) codes for one amino acid. There are four bases, so there are $4^3 = 64$ possible triplets, and these are enough to code for the 20 amino acids. (A doublet code would give only $4^2 = 16$ combinations, too few.)
- Non-overlapping:** each base is part of only one triplet, so the sequence is read in separate groups of three, one after another.
- Degenerate:** most amino acids are coded for by more than one triplet. For example, several codons code for glycine. This means that some changes in the base sequence do not change the amino acid.
- Some triplets are not amino acids: the codon AUG is a start codon (it also codes for methionine) and three codons (UAA, UAG, UGA) are stop codons, which end translation.

Reading a sequence in triplets

A section of the template strand of DNA reads TAC GGG TTT. State the number of amino acids coded for by these nine bases, and write the sequence of the mRNA transcribed from it.

1. The code is a non-overlapping triplet code, so divide the bases into groups of three: $9 \div 3 = 3$ triplets.
2. Each triplet codes for one amino acid: 3 amino acids.
3. mRNA bases are complementary to the template: A-U, T-A, C-G, G-C.
4. TAC → AUG; GGG → CCC; TTT → AAA.

Answer: 3 amino acids. mRNA: AUG CCC AAA.

Common mistake: 'Degenerate' does not mean the code is damaged. It means there is more than one codon for an amino acid. 'Non-overlapping' does not mean there are gaps between triplets.

Transcription Spec 2.13

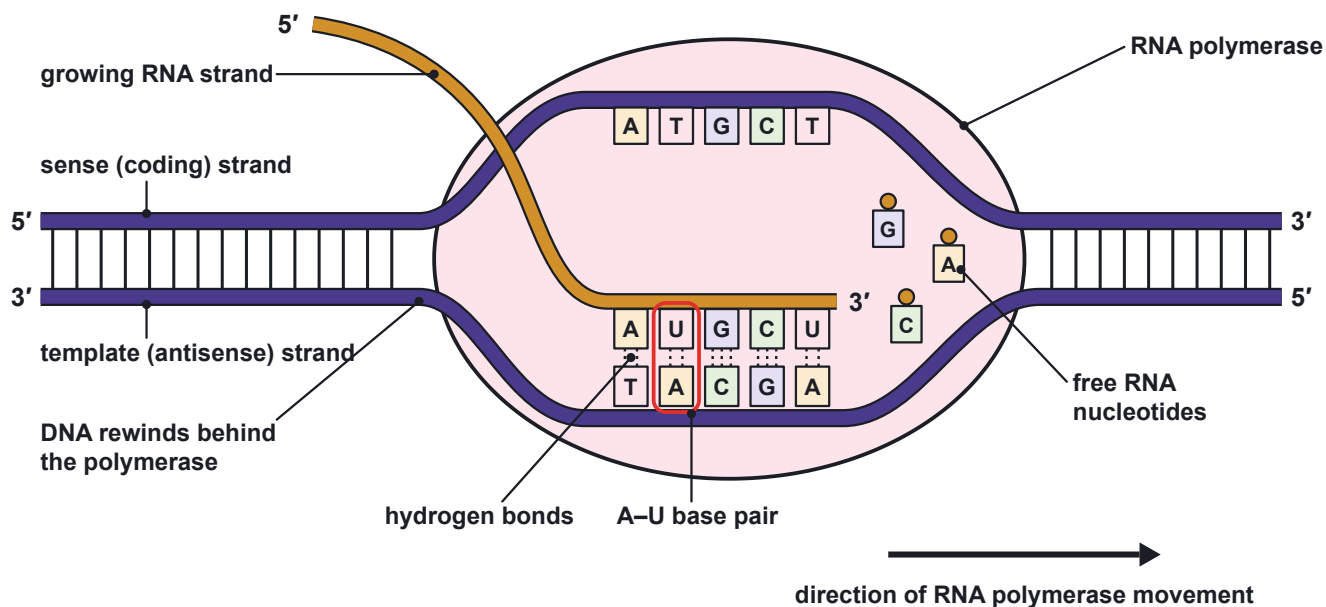
Protein synthesis has two stages: **transcription** (making mRNA from DNA in the nucleus) and **translation** (making a polypeptide from mRNA at a ribosome). DNA stays in the nucleus, so a copy of the gene, **messenger RNA (mRNA)**, carries the instructions to the cytoplasm.

1. RNA polymerase attaches to the DNA at the start of the gene. The two strands separate as the enzyme breaks the hydrogen bonds between bases.
2. One strand, the **template (antisense) strand**, is used to make the mRNA. The other strand, the sense (coding) strand, has the same base sequence as the mRNA (but with T instead of U).
3. Free RNA nucleotides pair with the complementary bases on the template strand: A-U, T-A, C-G, G-C.
4. RNA polymerase joins the RNA nucleotides together by phosphodiester bonds, to make a strand of mRNA.
5. The DNA strands re-join behind the enzyme. When RNA polymerase reaches a stop signal at the end of the gene, mRNA is released and leaves the nucleus through a nuclear pore.

Template and mRNA: an example

Strand	Base sequence
Template (antisense) strand of DNA	T A C G C A T T T
mRNA transcribed	A U G C G U A A A
Sense (coding) strand of DNA	A T G C G T A A A

Exam tip: Questions on the 'role of the template strand' want: it is the strand copied by RNA polymerase, and the mRNA base sequence is complementary to it. A common error is to transcribe the sense strand.



Transcription: RNA polymerase builds mRNA complementary to the **template strand**.

Translation, tRNA and ribosomes Spec 2.13

Translation takes place at a **ribosome** in the cytoplasm. A ribosome is made of ribosomal RNA (rRNA) and protein. **Transfer RNA (tRNA)** is a single strand of RNA folded into a clover-leaf shape. At one end it carries a specific amino acid; at the other end it has an **anticodon**, three bases that are complementary to a **codon** on mRNA.

1. The ribosome attaches to the mRNA and moves along it to the **start codon** (AUG). The tRNA with the complementary anticodon (UAC) and its amino acid (methionine) binds to the start codon.
2. A second tRNA, with the anticodon complementary to the next codon, binds. The ribosome holds two tRNAs at a time.
3. A peptide bond forms between the two amino acids, by a condensation reaction, and the first tRNA is released to pick up another amino acid.
4. The ribosome moves along the mRNA by one codon (three bases) and the next tRNA binds. The polypeptide chain gets longer one amino acid at a time.
5. This continues until the ribosome reaches a **stop codon** (UAA, UAG or UGA). No tRNA matches a stop codon, so translation ends and the polypeptide is released.

The molecules compared

Molecule	Structure	Role
mRNA	Single strand; sequence of codons.	Carries the genetic code from DNA to the ribosome.
tRNA	Folded single strand with an anticodon and an amino acid binding site.	Brings a specific amino acid to the ribosome and matches its anticodon with the mRNA codon.
Ribosome	rRNA and protein, two subunits.	Site of translation; holds mRNA and tRNA in place and helps form peptide bonds.

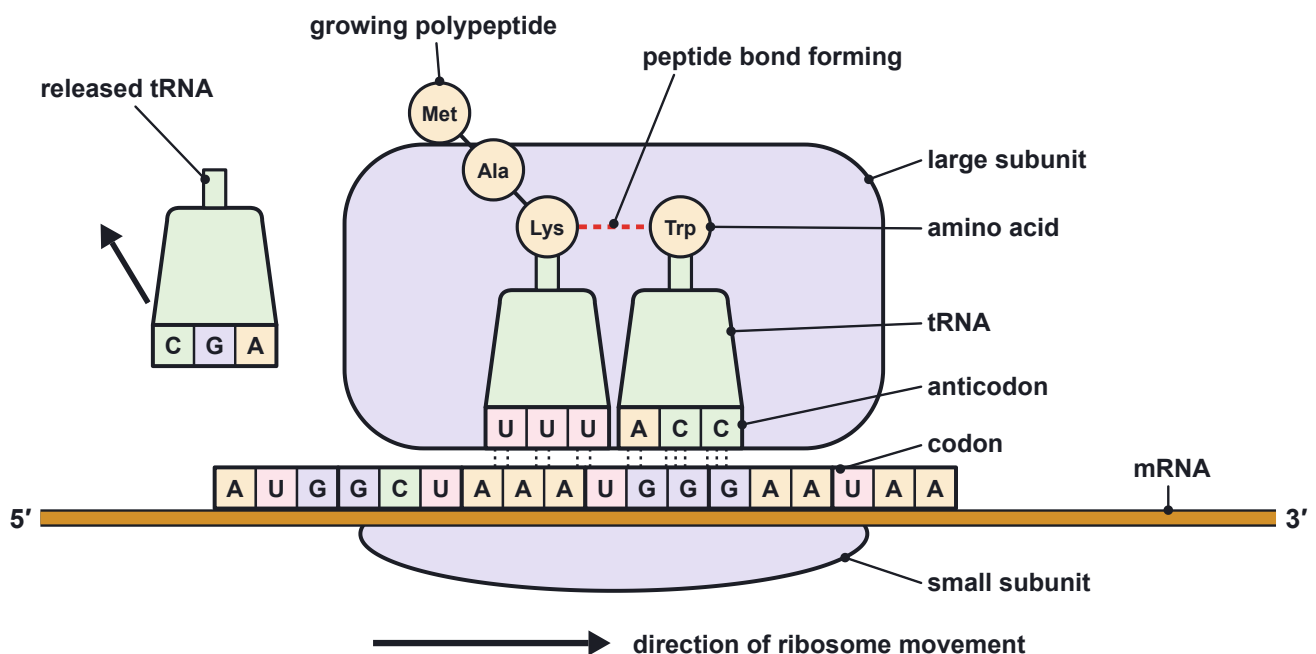
From DNA to polypeptide

A gene has the template strand sequence TAC TTT CCC ATT. Using the table of codons (AUG = methionine, AAA = lysine, GGG = glycine, UAA = stop), state the mRNA, the tRNA anticodons and the amino acid sequence.

1. mRNA is complementary to the template: TAC → AUG; TTT → AAA; CCC → GGG; ATT → UAA.
2. tRNA anticodons are complementary to the mRNA codons: AUG → UAC; AAA → UUU; GGG → CCC (no tRNA for UAA).
3. Translate each codon: AUG = methionine; AAA = lysine; GGG = glycine; UAA = stop.

Answer: mRNA: AUG AAA GGG UAA. Anticodons: UAC, UUU, CCC. Polypeptide: methionine - lysine - glycine.

Common mistake: The anticodon is on the tRNA; the codon is on the mRNA. The anticodon is complementary to the mRNA codon, which means it has the same base sequence as the template strand but with U instead of T.



Translation at the ribosome: tRNA anticodons pair with mRNA codons and a **peptide bond** forms between amino acids.

Gene mutations Spec 2.14

A **mutation** is a change in the sequence of bases in DNA. Mutations can arise from errors in DNA replication, when the wrong nucleotide is inserted, or when a base is added or missed out.

Types of gene mutation

Type	What happens	Possible effect
Substitution	One base is replaced by another.	Only one codon changes. The amino acid may stay the same (code is degenerate), change to a different amino acid, or become a stop codon (shorter polypeptide).
Insertion	An extra base is added.	Reading frame shifts (frameshift): every codon after the insertion changes, so a completely different sequence of amino acids is likely.
Deletion	A base is lost.	Frameshift: every codon after the deletion changes. (Adding or deleting three bases would add or remove one amino acid without shifting the rest.)

Effect of a deletion

The mRNA sequence AUG GAA CCU UUA is read in codons. The first G after AUG is deleted, giving AUG AAC CUU UA. State the effect on the codons.

1. Original codons: AUG GAA CCU UUA (four codons).
2. After the deletion the bases are AUGAACCUUUA, read in threes: AUG AAC CUU UA.
3. Every codon after the deletion has changed (GAA → AAC, CCU → CUU), so the amino acid sequence after the mutation is changed.

Answer: A frameshift: all the codons after the deletion are changed, giving a different amino acid sequence.

Consequences of mutations

- Some mutations cause **cancer**, if they occur in genes that control cell division, leading to uncontrolled cell division and a tumour.
- Some cause **genetic disorders**, if the protein made is faulty, for example the mutation that causes cystic fibrosis.
- Many mutations have **no observable effect**: the code is degenerate so the amino acid may not change; the change may be in a part of DNA that does not code for a protein; or the new amino acid may not alter the shape of the protein.

Exam tip: In a question about the effect of a mutation, work through the chain: base sequence → mRNA codon → amino acid sequence → tertiary structure → function. Say whether it is a substitution or a frameshift.

mRNA codons, and the amino acids they code for

original



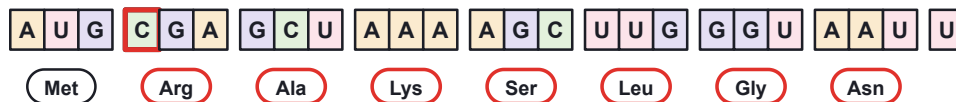
reading frame: triplets (codons)

substitution



A → U: one amino acid changes (Glu → Val)

insertion



extra C: frameshift, every codon after it changes

deletion



first G of GAG lost: frameshift, early STOP codon

A **substitution** changes at most one amino acid; an **insertion** or **deletion** causes a **frameshift**.

Quick check

- 1 . Name the bond that joins nucleotides in one DNA strand.
- 2 . Which bases pair together in DNA?
- 3 . What does semi-conservative mean?
- 4 . What is the role of RNA polymerase in transcription?
- 5 . What is an anticodon?

4 Inheritance, cystic fibrosis and genetic screening Spec 2.15-2.18

Genetic terms Spec 2.15

Gene	A sequence of bases on a DNA molecule that codes for a sequence of amino acids in a polypeptide chain, found at a particular locus on a chromosome.
Allele	One of the different forms of a gene.
Genotype	The alleles that an organism has for a particular gene (or genes).
Phenotype	The observable characteristics of an organism, resulting from its genotype and the environment.
Dominant	An allele that is expressed in the phenotype when present in only one copy (in the heterozygote).
Recessive	An allele that is expressed in the phenotype only when two copies are present (in the homozygote).
Codominance	Both alleles are expressed in the phenotype of the heterozygote, because neither is dominant over the other.
Homozygote	An organism with two identical alleles of a gene.
Heterozygote	An organism with two different alleles of a gene.

A person has two alleles of each gene, one inherited from each parent. A **dominant** allele is shown by a capital letter (for example B) and a **recessive** allele by a lower-case letter (b). The **genotype** BB is homozygous dominant, bb is homozygous recessive, and Bb is heterozygous.

Codominance

In **codominance**, two alleles are both expressed. For example, the human ABO blood group gene has alleles I^A , I^B and I^O . I^A and I^B are codominant, so a person with genotype $I^A I^B$ has blood group AB. Both are dominant over I^O , so $I^A I^O$ has group A and $I^B I^O$ has group B, and only $I^O I^O$ has group O. When codominant alleles are used, the capital letter is the gene and the superscripts are the alleles.

Common mistake: A dominant allele is not necessarily common or 'stronger'. It just shows in the phenotype in the heterozygote. Also, a 'carrier' has one copy of a recessive allele and does not show the condition.

Monohybrid inheritance and pedigree diagrams Spec 2.15

A **monohybrid cross** looks at the inheritance of one gene. Each parent makes gametes containing one allele of the gene (as a result of meiosis), and the alleles combine at random when gametes fuse at fertilisation.

A cross between two heterozygotes

Cystic fibrosis is caused by a recessive allele, f. Two parents are both heterozygous (Ff). Draw a genetic diagram, and find the probability that a child has cystic fibrosis.

1. Parent genotypes: Ff × Ff.
2. Gametes: F or f from each parent.
3. Possible offspring: FF, Ff, Ff, ff (Punnett square).
4. Phenotypes: FF and Ff do not have the condition (3), ff has cystic fibrosis (1).

Answer: Ratio 3 : 1 (unaffected : affected). Probability of a child with cystic fibrosis is 1 in 4 (25 %).

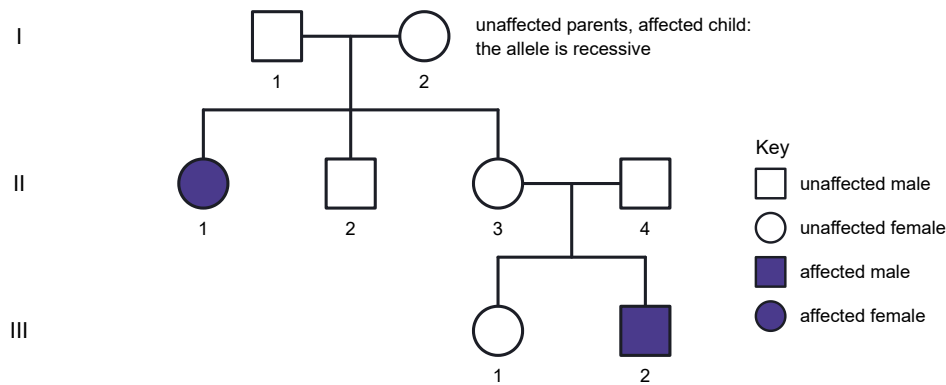
Pedigree diagrams

A **pedigree diagram** shows the phenotypes of family members across generations. Males are usually drawn as squares and females as circles; shaded symbols show people with the condition; horizontal lines join parents and vertical lines lead to their children.

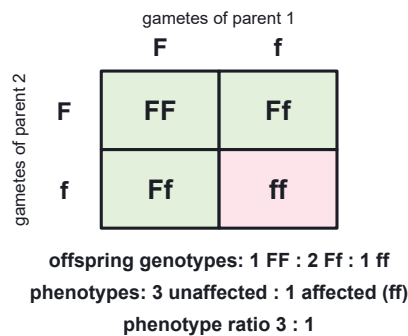
Clues for working out the type of inheritance

Clue in the pedigree	Conclusion
Two unaffected parents have an affected child	The condition is caused by a recessive allele, and both parents are heterozygous carriers.
Two affected parents have an unaffected child	The condition is caused by a dominant allele, and both parents are heterozygous.
Every affected person has an affected parent	Suggests a dominant allele.
Condition skips generations	Suggests a recessive allele.

Exam tip: To prove that an allele is recessive, find a pair of unaffected parents with an affected child, and say so, naming the individuals. To prove dominant, find affected parents with an unaffected child. Give the genotypes of the individuals concerned.



Punnett square for the cross $Ff \times Ff$ (F = unaffected allele, f = recessive allele)



Unaffected parents with an affected child show that the allele is **recessive**. $Ff \times Ff$ gives **3 : 1**.

Sex linkage and red-green colour blindness Spec 2.15

Human females have two X chromosomes (XX) and males have one X and one Y (XY). The Y chromosome is shorter and carries few of the genes found on the X chromosome. A **sex-linked** characteristic is controlled by a gene on a sex chromosome, and here we consider genes on the **X chromosome**.

Red-green colour blindness is caused by a recessive allele on the X chromosome. The alleles are written as superscripts on the X: X^B (normal colour vision, dominant) and X^b (colour blind, recessive).

Genotypes and phenotypes	
Genotype	Phenotype
$X^B X^B$	Female, normal colour vision
$X^B X^b$	Female, normal colour vision, carrier of the allele
$X^b X^b$	Female, colour blind
$X^B Y$	Male, normal colour vision
$X^b Y$	Male, colour blind

A cross involving a carrier mother

A woman who is a carrier of red-green colour blindness ($X^B X^b$) has children with a man with normal colour vision ($X^B Y$). Give the possible genotypes and phenotypes of their children.

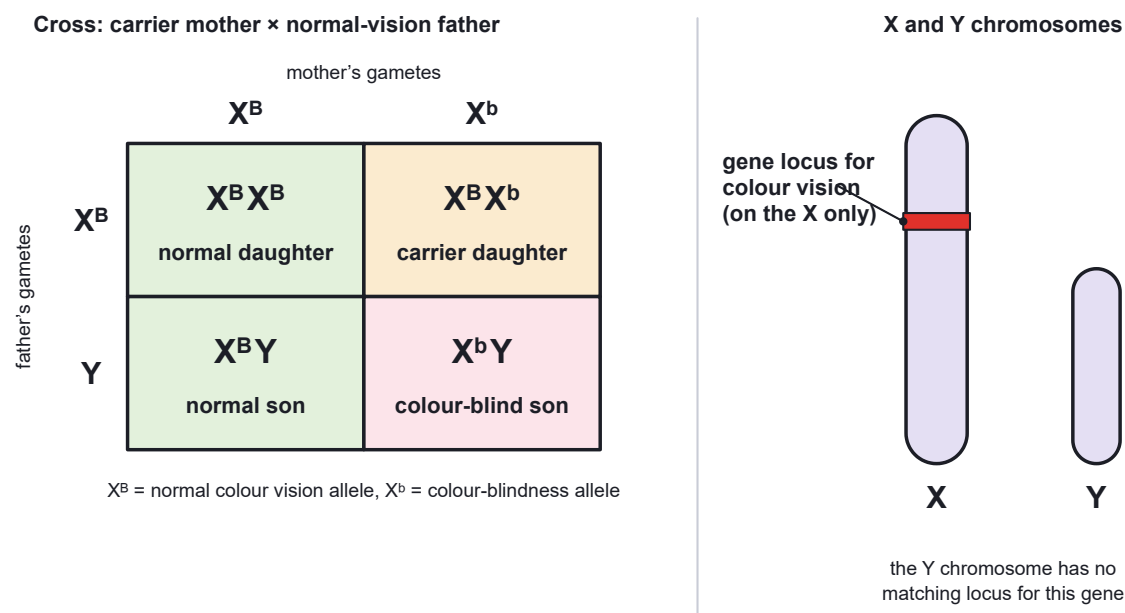
1. Gametes from the mother: X^B or X^b . Gametes from the father: X^B or Y .
2. Daughters: $X^B X^B$ (normal) or $X^B X^b$ (carrier, normal vision).
3. Sons: $X^B Y$ (normal) or $X^b Y$ (colour blind).

Answer: Half of the sons (so 1 in 4 of all children) are colour blind; no daughters are colour blind, but half the daughters are carriers.

Why the condition is more common in males

- A male has only one X chromosome, so a single recessive allele on it is always expressed (there is no second allele to mask it).
- A female must inherit the recessive allele on both of her X chromosomes to show the condition.
- A male inherits his X chromosome from his mother, never from his father. A colour-blind man passes the allele to all of his daughters (who become carriers) and to none of his sons.

Exam tip: Write sex-linked genotypes with the X, and the allele as a superscript, and always include the Y for males. Marks are lost for writing the alleles without the sex chromosomes.



Colour blindness is **sex-linked**: the gene is on the **X chromosome** only.

Cystic fibrosis Spec 2.16

Cystic fibrosis is a genetic disorder caused by a recessive allele of the CFTR gene, on chromosome 7. Someone with the condition has two copies of the faulty allele; carriers (heterozygous) have one copy and do not have symptoms. The commonest mutation is a deletion of three bases, which removes one amino acid from the protein.

From mutation to symptoms

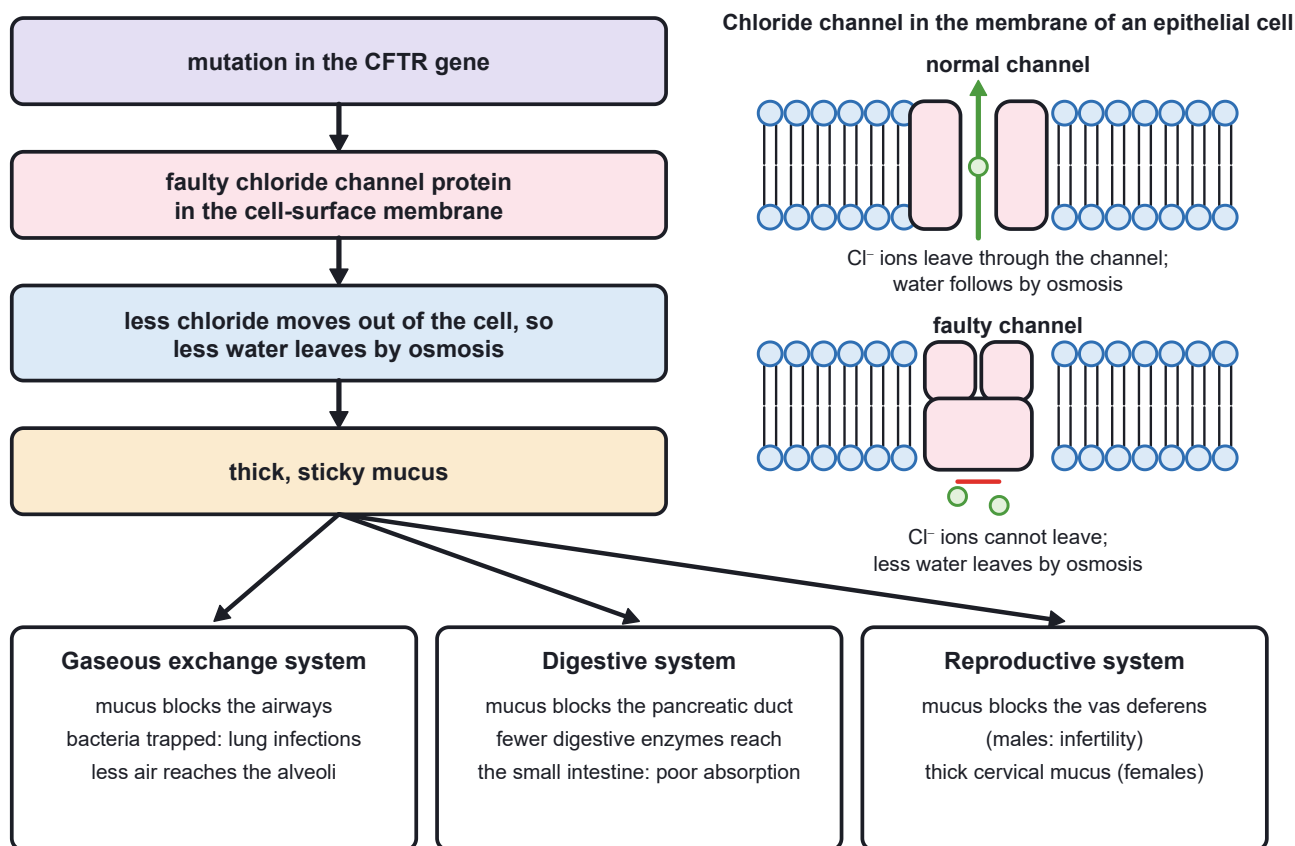
1. The gene mutation changes the primary structure of the CFTR protein, so it folds into the wrong tertiary structure.
2. The faulty protein does not work as a chloride ion channel in the cell-surface membrane (or does not reach the membrane).

- Chloride ions are not secreted out of the epithelial cells, so water does not move out by osmosis (the water potential gradient is reduced).
- The mucus on the surface of the epithelium is therefore thick and sticky, instead of thin and watery.

Effects of cystic fibrosis on body systems

System	Effect
Gaseous exchange	Thick mucus blocks the airways and traps bacteria, which cannot be removed by cilia. This causes repeated lung infections, inflammation and damage, reducing the surface area for gas exchange and making breathing difficult. Less oxygen reaches the blood.
Digestive	Thick mucus blocks the pancreatic duct, so digestive enzymes (amylase, lipase and protease) cannot reach the small intestine. Food is poorly digested and absorbed, which causes poor growth and malnutrition. Mucus may also block the intestine.
Reproductive	In males, the tubes (vas deferens) that carry sperm may be blocked or missing, causing infertility. In females, thick mucus in the cervix can prevent sperm from reaching the egg, reducing fertility.

Exam tip: To score full marks on 'how does a mutation cause these symptoms', follow the chain: **gene mutation** → **faulty channel protein** → **less chloride and water secretion** → **thick mucus** → **blockage of the specific system**.



How a **CFTR mutation** leads to **thick sticky mucus** and the symptoms of cystic fibrosis.

Genetic screening: carriers, PGD and prenatal testing Spec 2.17

Identifying carriers

A DNA sample (from blood or a cheek swab) can be tested for the presence of a particular recessive allele. People with a family history of a condition such as cystic fibrosis can find out if they are **carriers**,

and so assess the risk of having an affected child.

Pre-implantation genetic diagnosis (PGD)

1. Eggs are fertilised in vitro (IVF) and the embryos develop to an early stage (about 8 cells; many clinics now wait until the blastocyst stage at about 5 days).
2. One or two cells (or, at the blastocyst stage, a few cells from the outer layer) are removed from each embryo, and their DNA is tested for the faulty allele.
3. Only embryos that do not carry the allele (or that are not affected) are implanted into the uterus.

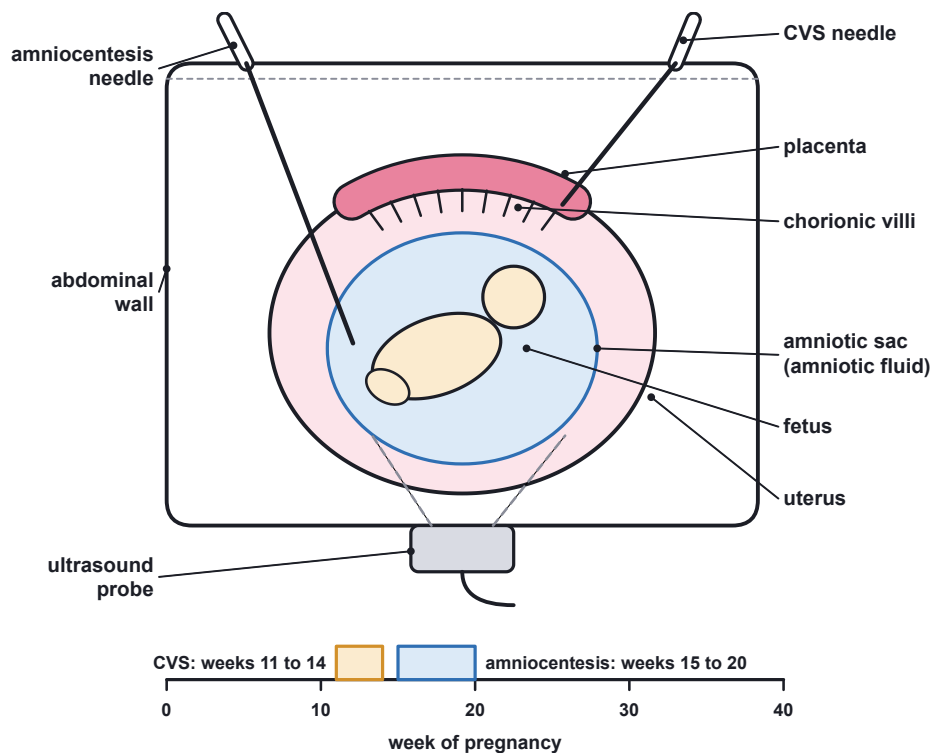
Prenatal testing

Amniocentesis and chorionic villus sampling		
Feature	Amniocentesis	Chorionic villus sampling (CVS)
Sample	Amniotic fluid containing fetal cells.	Cells from the chorionic villi (part of the placenta).
Method	A needle is passed through the abdominal wall and uterus wall, guided by ultrasound, to remove fluid.	A needle is passed through the abdomen (or a tube through the cervix), guided by ultrasound, to remove a small sample of tissue.
Timing	From about 15 weeks of pregnancy.	From about 11 weeks of pregnancy (earlier).
Risk	Small risk of miscarriage.	Small risk of miscarriage (slightly higher than amniocentesis).
After	Fetal cells cultured; DNA or chromosomes analysed.	Fetal cells analysed directly, so results sooner.

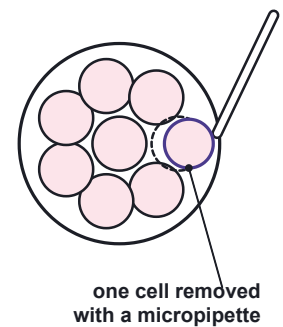
Implications of prenatal screening

- A positive result may lead parents to consider termination of the pregnancy, or to prepare for the birth of a child with the condition.
- Both tests carry a risk of miscarriage, including of an unaffected fetus.
- A result can cause anxiety, and an incorrect result (false positive or false negative) could lead to a wrong decision.
- Results show only that a fetus has a particular allele. They do not show how severe the condition will be, and for many conditions there is no cure.
- Test results about a fetus also give information about the genes of the parents and other relatives.

Exam tip: Do not describe these tests as treatment. They are screening tests: they give information. Be specific about risks, such as miscarriage, rather than saying 'it is dangerous'.



PGD
embryo from IVF, 8-cell stage



the removed cell is tested for
the faulty allele; only unaffected
embryos are put in the uterus

CVS is done at 11-14 weeks and **amniocentesis** at 15-20 weeks; **PGD** tests an IVF embryo before it is transferred.

Ethical and social issues of genetic screening Spec 2.18

To **discuss** the ethical and social issues, identify the issue, set out arguments on each side from different viewpoints, and reach a reasoned conclusion. The issues come from the fact that screening gives information about people, and about embryos and fetuses that cannot consent.

Viewpoints on genetic screening		
Viewpoint	In favour of screening	Against, or concerns
Religious	Some traditions allow screening to prepare for, or prevent, severe suffering, and to allow informed choices.	Many traditions hold that life is sacred from conception, so discarding embryos in PGD or terminating a pregnancy is wrong. Views differ between and within religions.
Moral	Respect for autonomy: parents have the right to information and to choose. Avoiding the suffering of a child with a severe disorder.	Every life has equal value, including that of a person with a disorder. Embryos may have a right to life. Screening could lead to selection of embryos for other traits (so-called designer babies).
Social	Reduces the number of children born with serious conditions, and reduces demand on families and health services.	Pressure on parents to screen and terminate; stigma towards people living with the condition; cost and unequal access; discrimination by insurers or employers using genetic information; privacy of genetic data.

- Who should have access to the results (the individual, family members, employers, insurers)?
- Should screening be offered for all disorders, including those with late onset or with mild effects?
- How should genetic counselling be provided to help people understand the results and the options?

Exam tip: A discuss answer needs both sides and the named viewpoints (religious, moral and social). Avoid giving a personal opinion without reasons. A balanced conclusion earns credit, such as 'screening is justified for severe disorders, provided counselling and consent are given'.

Common mistake: Do not assume that all religious people share one view. Say 'some religious groups' or 'many religious traditions' to stay accurate.

Quick check

- 1 . What is the difference between genotype and phenotype?
- 2 . What is codominance?
- 3 . Why is red-green colour blindness more common in males?
- 4 . Name the protein that is faulty in cystic fibrosis.
- 5 . Name two methods of prenatal testing.

Quick check answers

Gas exchange, membranes and transport

1. A large surface area to volume ratio, a thin surface (short diffusion distance) and a large concentration difference.
2. It halves, because the rate is inversely proportional to the thickness (Fick's law).
3. Oxygen is small and non-polar, so it dissolves in the hydrophobic centre. Ions are charged and are repelled by it.
4. The net movement of free water molecules through a partially permeable membrane, down a water potential gradient.
5. It is hydrolysed to ADP and P_i , releasing the energy that changes the shape of the carrier protein.

Proteins and enzymes

1. A peptide bond (covalent), formed in a condensation reaction that releases water.
2. Hydrogen bonds, ionic bonds and disulfide bonds (plus hydrophobic interactions).
3. Hydrophilic R groups face outwards, so it dissolves in the cytoplasm of red blood cells.
4. It lowers it.
5. From the gradient of the tangent drawn at time zero on a graph of product against time.

DNA, the genetic code and gene expression

1. A phosphodiester bond, formed by a condensation reaction.
2. Adenine with thymine (two hydrogen bonds), cytosine with guanine (three hydrogen bonds).
3. Each new DNA molecule has one original strand and one newly made strand.
4. It breaks the hydrogen bonds, and joins RNA nucleotides complementary to the template strand by phosphodiester bonds.
5. Three bases on a tRNA molecule that are complementary to a codon on mRNA.

Inheritance, cystic fibrosis and genetic screening

1. Genotype is the alleles an organism has; phenotype is its observable characteristics.
2. Both alleles are expressed in the phenotype of a heterozygote.
3. The allele is recessive and on the X chromosome; males have only one X, so one copy is expressed.
4. The CFTR protein, a chloride ion channel in cell-surface membranes.
5. Amniocentesis and chorionic villus sampling (CVS).