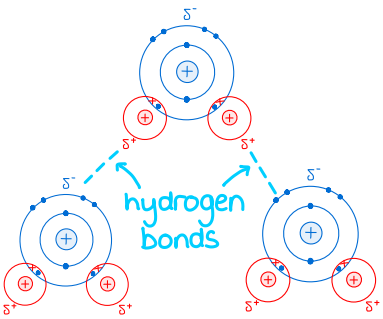
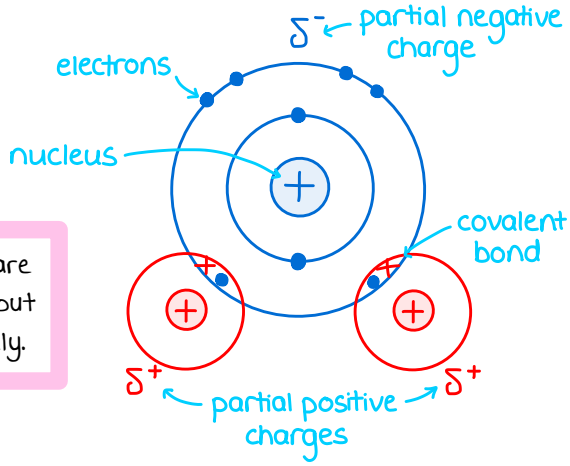
WATER

Structure

- Two hydrogen atoms share electrons with one oxygen atom (covalent bonds)
- Electrons pulled towards oxygen → hydrogens have a partial positive charge
- Oxygen has two lone (unshared) electron pairs → oxygen has a partial negative charge
- A polar molecule
- Partial charges attract other water molecules
  → this forms hydrogen bonds

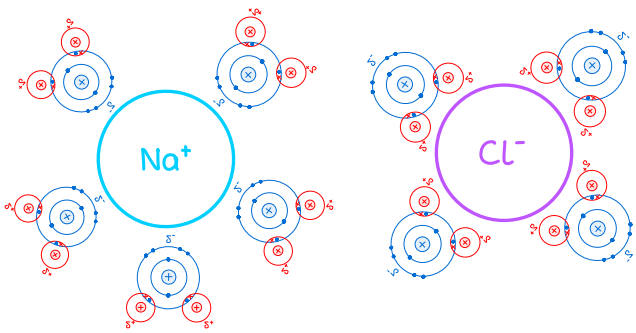


Hydrogen bonds are weak individually but strong collectively.



Properties

- Good solvent → polar water molecules surround and are attracted to ions or other polar molecules, so they can dissolve and be transported e.g. glucose dissolves in blood plasma



If a molecule has polar OH groups it will dissolve in water because it can form hydrogen bonds with water. More free OH groups = more soluble.

- Strong cohesion → polar water molecules attracted to each other with hydrogen bonds
  → water flows well e.g. it forms an unbroken chain in the xylem vessels
  → has a high surface tension so forms droplets, and can support small organisms
- Adhesion → water molecules can stick to other surfaces e.g. the walls of the xylem vessels
- Useful metabolite → used in metabolic reactions e.g. condensation and hydrolysis reactions
- High latent heat of vaporisation → lots of energy needed to break hydrogen bonds
  → uses lots of heat energy to evaporate so it has a cooling effect
- High specific heat capacity → hydrogen bonds can absorb lots of energy
  → can buffer changes in temperature because it can lose or gain a lot of energy without changing temperature (good for aquatic organisms)
- Ice is less dense than liquid water → ice floats and forms an insulating layer on top of water so aquatic organisms can survive
- Transparent → allows light to pass through to reach aquatic plants
- Density allows organisms to float → gives buoyancy
- Water is a good habitat because of its properties

See Module 3 for more about water transport in plants.

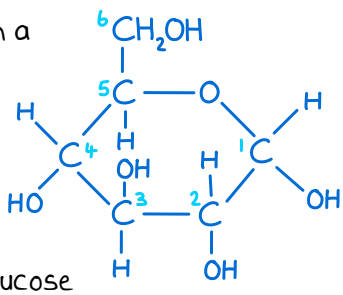
CARBOHYDRATES

- Carbohydrates contain carbon, hydrogen and oxygen → many monosaccharides have the general formula $C_nH_{2n}O_n$

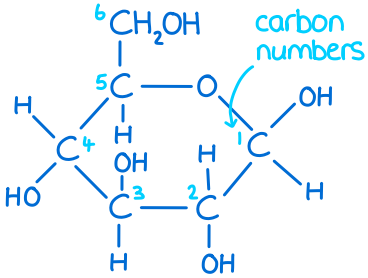
Glucose

- A monosaccharide with the formula $C_6H_{12}O_6$
- A hexose monosaccharide (six carbon atoms) in a ring structure
- Soluble in water → easily transported
- Main energy source for animals and plants → chemical bonds store lots of energy
- Two isomers: α-glucose and β-glucose → H and OH groups on carbon 1 inverted in β-glucose

α-glucose



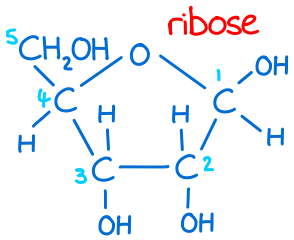
β-glucose



Ribose

- A monosaccharide with the formula $C_5H_{10}O_5$
- A pentose monosaccharide (five carbon atoms) in a ring structure
- A component of RNA nucleotides and ATP

ribose

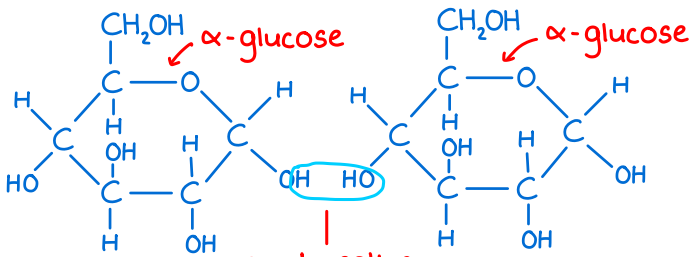


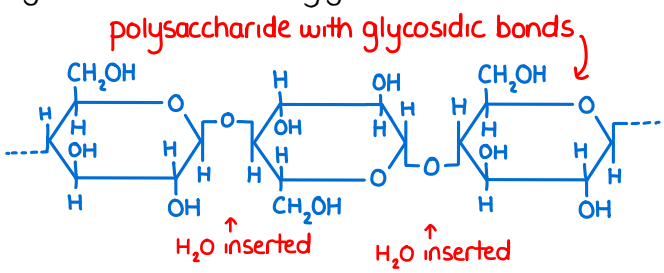
Monosaccharides are small soluble carbohydrate monomers. They also include fructose and galactose.

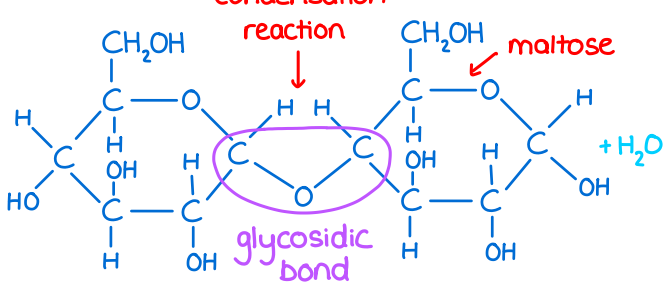
Glycosidic bonds, condensation and hydrolysis reactions

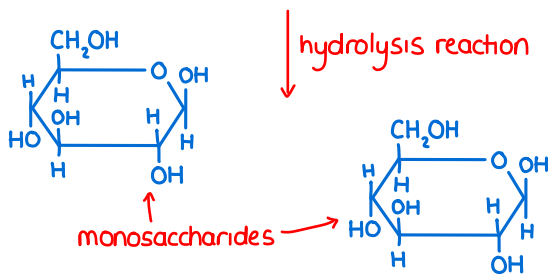
- Condensation reaction → two molecules join to form a new chemical bond and a water molecule is eliminated
- Hydrolysis reaction → a water molecule is inserted and the chemical bond is broken (reverse of a condensation reaction)
- Condensation reactions form glycosidic bonds between monosaccharides to create disaccharides and polysaccharides, and hydrolysis reactions break glycosidic bonds

Monosaccharides and disaccharides are sugars.









Disaccharides

- Two monosaccharides joined together with a glycosidic bond in a condensation reaction
- Soluble in water → disaccharides and monosaccharides have free OH groups
- Maltose = α-glucose + α-glucose joined with an α1,4 glycosidic bond
- Sucrose = α-glucose + fructose joined with an α1,2 glycosidic bond
- Lactose = β-glucose + galactose joined with a β1,4 glycosidic bond

CARBOHYDRATES

Polysaccharides

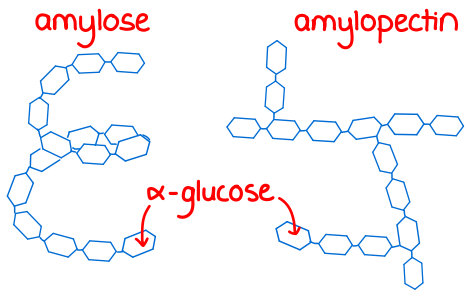
- Large insoluble polymers of monosaccharides joined with glycosidic bonds
- Starch and glycogen are large metabolically inactive energy storage molecules which cannot leave cells

Starch

- Glucose storage in plant cells → hydrolysed when glucose is needed
- Insoluble in water → does not affect the water potential of cells so water is not drawn in by osmosis
- Amylose → unbranched α-glucose polysaccharide (α1,4 glycosidic bonds)
  → coiled structure so is compact
- Amylopectin → branched α-glucose polysaccharide (α1,4 and α1,6 glycosidic bonds) so is compact
  → branches mean enzymes can easily access more glycosidic bonds = faster glucose release

Iodine test for starch

1) Add iodine in potassium iodide solution to sample.
2) If starch is present: goes from browny-orange to blue-black.

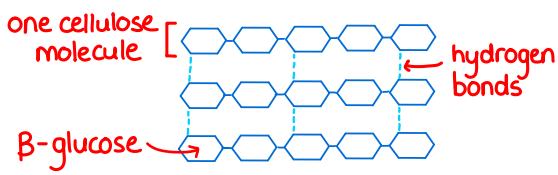


Glycogen

- Excess glucose storage in animal cells
  → hydrolysed when glucose is needed
- Insoluble in water → does not affect the water potential of cells so water is not drawn in by osmosis
- Highly branched α-glucose polysaccharide (α1,4 and α1,6 glycosidic bonds) → more ends so glucose can be released quickly by enzymes, and more energy can be stored in a small space
- More branched and compact than amylopectin

Cellulose

- Found in plant cell walls to give strength
- Unbranched long and straight β-glucose polymers (β1,4 glycosidic bonds)
- Chains linked with many hydrogen bonds to form inflexible microfibrils with high tensile strength

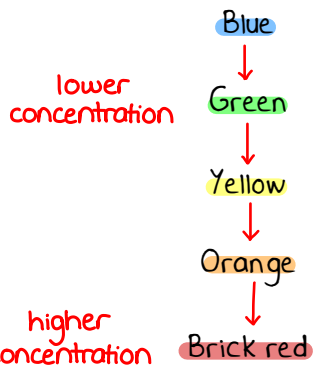


Benedict's test for sugars

- Monosaccharides, maltose, and lactose are reducing sugars
- Sucrose is a non-reducing sugar

1) Add an excess of blue Benedict's reagent to liquid food sample in a test tube.
2) Heat the tube in a water bath set to boil.
3) If reducing sugars are present: coloured precipitate forms. End test here.
4) If no reducing sugars are present: solution stays blue. Go to step 5.
5) Hydrolyse non-reducing sugars to monosaccharides by adding dilute HCl to new tube of sample and heat in a water bath set to boil.
6) Neutralise by adding sodium hydrogencarbonate.
7) Repeat steps 1 and 2.
8) If coloured precipitate now forms, non-reducing sugars are present in the sample.
9) If the solution is still blue, neither type of sugars are present.

Colour of precipitate depends on the sugar concentration:



You could filter, dry, and weigh the precipitate to make more accurate comparisons.

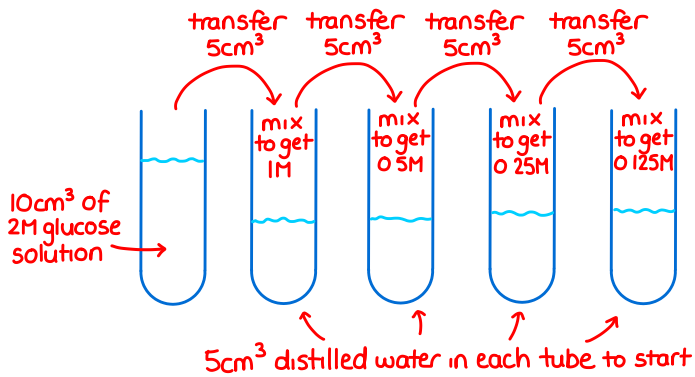
CARBOHYDRATES

Using a colorimeter

- A more accurate and quantitative way to measure glucose concentration after the Benedict's test
  - → judging colour is subjective and more affected by human error
- A colorimeter measures absorbance of light
  - → a lower absorbance reading means more light is transmitted through the sample
- Higher glucose concentration in a sample results in more precipitate being formed in the Benedict's test
  - → the precipitate is filtered out to leave a solution which will be less blue if more precipitate was formed
  - → a less blue solution absorbs less light, so gives a lower absorbance reading
  - → the lower the absorbance, the higher the glucose concentration
- Can quantify the concentration of glucose by producing a calibration curve:

1) Start with a solution of known glucose concentration (e.g. 2M) and create a serial dilution. In this case we are diluting the glucose solution by a factor of two each time.

Mix well at each stage and use a clean pipette for each transfer.



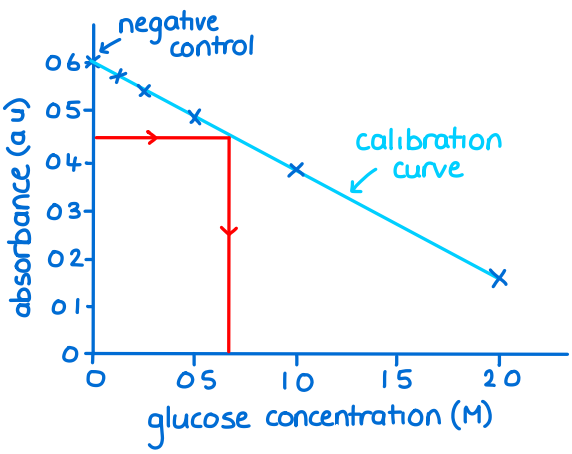
2) Set up the colorimeter by selecting the red filter and zeroing to a distilled water. This ensures the absorbance values are comparable. (A filter of a complementary colour to the solution is chosen because that wavelength is absorbed most strongly and transmitted least.)

3) Carry out the Benedict's test (detailed on previous page) on all serial dilution tubes, samples, and a negative control (distilled water). Control the volume and concentration of Benedict's solution used and control the time in the boiling water bath.

4) Filter the precipitate out of the solution so you are left with tubes containing the remaining solution (varying shades of blue). Transfer these to cuvettes.

5) Measure the absorbance of each using the colorimeter. Produce a calibration curve as below from the serial dilution.

6) For the samples with unknown glucose concentration, use the calibration curve to read across from the absorbance value and down to find the concentration. This is called interpolation.



This could be done for other reducing sugars as well as glucose.

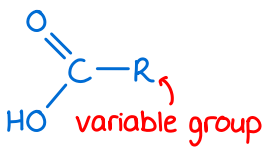
If the precipitate was not filtered out of the solution, the highest glucose concentration would give the highest absorbance reading.

LIPIDS

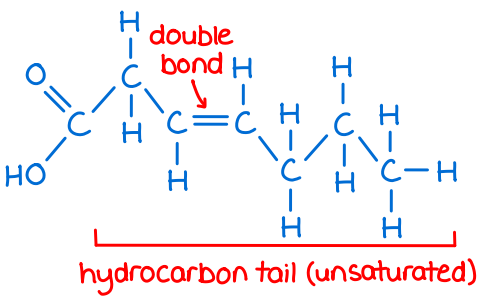
- Lipids contain carbon, hydrogen and oxygen

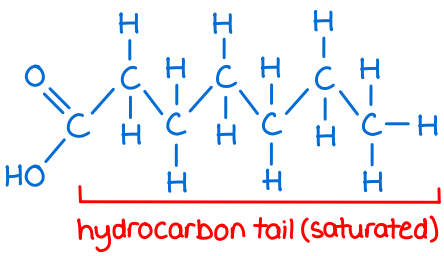
Fatty acids

- Have a carboxyl group and a variable R group (the hydrocarbon tail)
- Saturated fatty acids
  - → no double C=C bonds in the hydrocarbon tail
  - → more tightly packed together so higher melting point e.g. butter (solid at room temperature)
- Unsaturated fatty acids
  - → one or more double C=C bonds in the hydrocarbon tail so the chain kinks
  - → kinks mean molecules are less tightly packed together so lower melting point e.g. vegetable oils (liquid at room temperature)
  - → monounsaturated fatty acids have one double C=C bond
  - → polyunsaturated fatty acids have more than one double C=C bond
- Hydrocarbon tails are hydrophobic (insoluble in water)



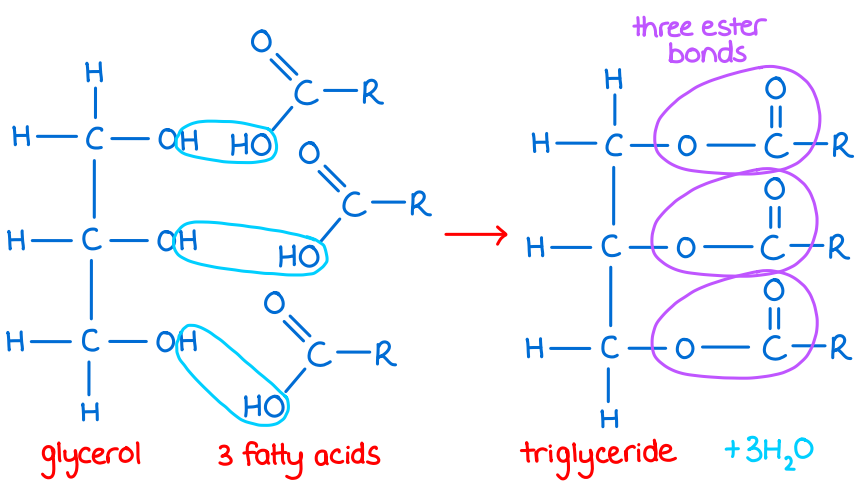
Saturated fatty acid general formula = $C_nH_{(2n+1)}COOH$



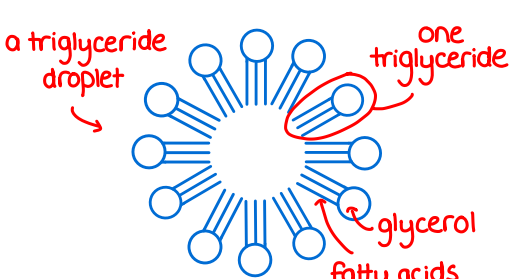


Triglycerides

- One molecule of glycerol bound to three fatty acids
- Fatty acids join to glycerol in a condensation reaction
  - → an ester bond is formed and a water molecule is released
- Three water molecules released and three ester bonds formed for each triglyceride
- Ester bonds are broken by a hydrolysis reaction
- Energy storage molecules → hydrocarbon tails release a lot of energy when broken down
- In animals, fat stored as triglycerides provides thermal insulation and protects organs
- Insoluble in water → do not affect the water potential of cells so water is not drawn in by osmosis
- Clump together in droplets with the hydrophobic hydrocarbon tails facing inwards
  - → do not form a bilayer because there is no hydrophilic region



Know how to relate structure to function.



LIPIDS

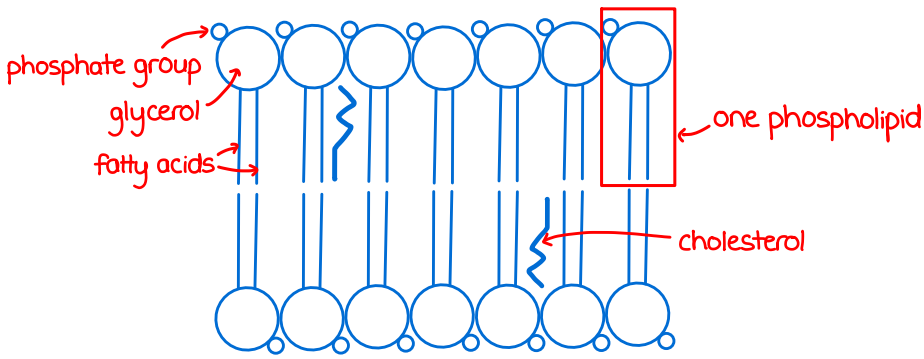
Phospholipids

- One molecule of glycerol bound to two fatty acids and a phosphate group
- Phosphate groups are hydrophilic, fatty acids are hydrophobic
- Form a bilayer in cell membranes → water soluble (polar) substances cannot pass through the hydrophobic centre of the bilayer
- Fatty acids can be saturated or unsaturated
  → more saturated fatty acids in the phospholipid bilayer decreases fluidity because they pack together more closely than unsaturated fatty acids

Cholesterol

- Hydrocarbon ring (with a polar hydroxyl group) and a hydrocarbon tail
- Small and flat shape → fits between phospholipids and binds to hydrophobic tails
- Affects fluidity of eukaryotic cell membranes
  → at higher temperatures (including body temperature) cholesterol increases packing of the membrane to decrease membrane fluidity
  → at low temperatures cholesterol increases membrane fluidity to stop the membrane solidifying

See the cell membrane notes for more on this.



Emulsion test for lipids

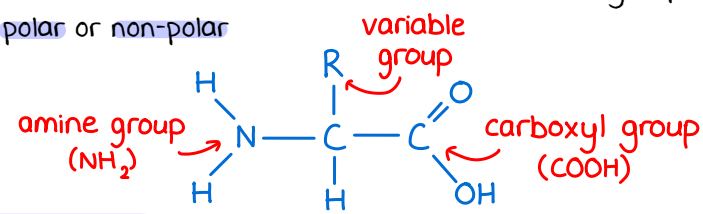
1) Add ethanol to sample and shake.
2) Add water and mix.
3) If lipid is present → milky emulsion forms.
4) If no lipid is present → stays clear.

PROTEINS

Amino acids

- The monomers of proteins
- Contain carbon, hydrogen, nitrogen, oxygen and sometimes sulfur
- Have a carboxyl group, an amine group, and a variable R group
- There are 20 different amino acids, each with a different R group
- R groups can be polar or non-polar

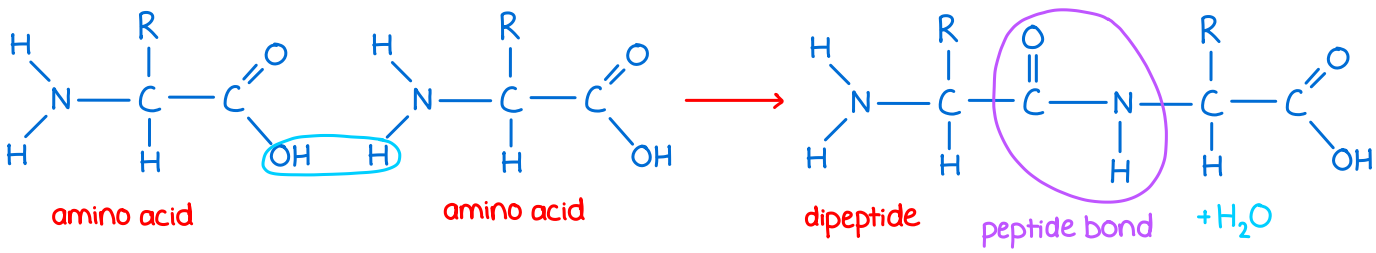
A monomer is a smaller unit from which larger polymers are made.



Amino acids can be joined in any order and length.

Peptide bonds and dipeptides

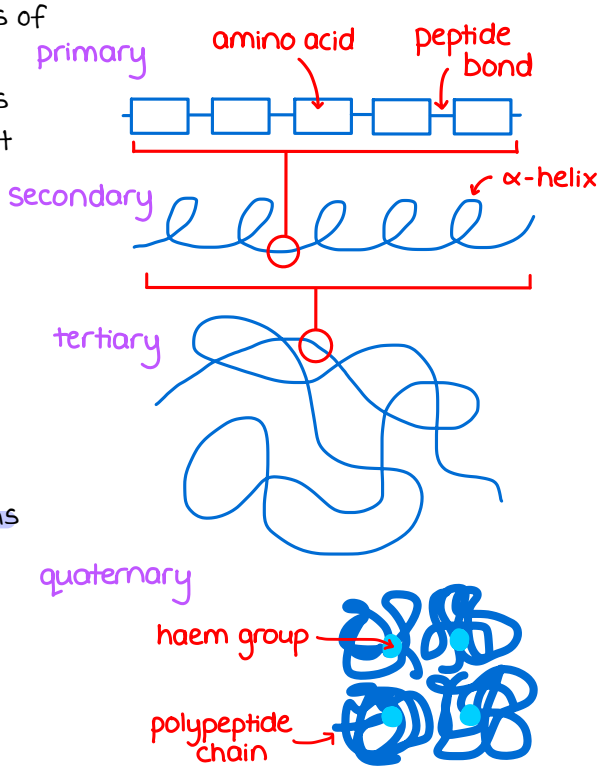
- A condensation reaction joins two amino acids with a peptide bond
  - → produces a dipeptide and a molecule of water
- A hydrolysis reaction breaks a peptide bond by adding a molecule of water
- A polypeptide is a polymer of amino acids (a long chain of amino acids joined with peptide bonds)



Protein structure

- Primary structure → order of amino acids in the polypeptide chain
  - → amino acids are joined with peptide bonds
- Secondary structure
  - → hydrogen bonds form between partial charges on parts of amino acids in the polypeptide
  - → alpha helix (coiled) or beta pleated sheet (folded) forms
  - → can be areas of both alpha helix and beta pleated sheet within the same polypeptide
- Tertiary structure
  - → further folding and coiling (bonds form between R groups of amino acids in the polypeptide)
  - → more hydrogen bonds
  - → ionic bonds between positively and negatively charged R groups
  - → disulfide bridges between cysteine amino acids
  - → hydrophobic regions clump together, hydrophilic regions turn outwards
  - → final structure for proteins made from one polypeptide
- Quaternary structure
  - → some proteins have more than one polypeptide chain
  - → polypeptides held together with hydrogen bonds, ionic bonds, and disulfide bridges

Cysteine contains a sulfur atom in the R group.



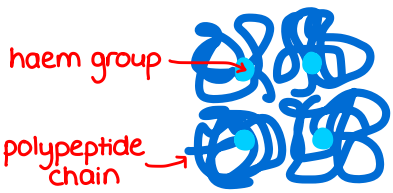
PROTEINS

Globular and fibrous proteins

- Globular → soluble, spherical, often complementary to another molecule
  - → hydrophilic regions (with polar R groups) face outwards and hydrophobic regions face inwards which them soluble
  - → easily transported

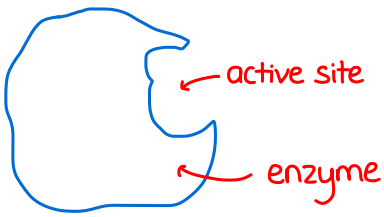
Haemoglobin

carries oxygen in red blood cells



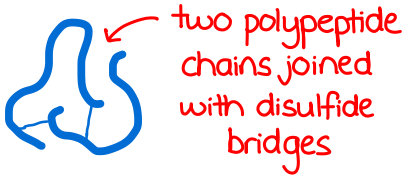
Enzymes

e.g. amylase



Hormones

e.g. insulin



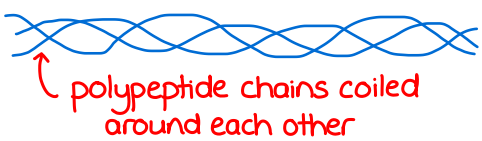
Haemoglobin is a conjugated protein: a globular protein with a non-protein prosthetic group (haem) attached with covalent, ionic, or hydrogen bonds.

Primary structure determines the tertiary structure and function.

- Fibrous → long chains of amino acids, insoluble (many non-polar hydrophobic R groups), flexible, strong
  - → little tertiary structure and mostly unreactive
  - → chains parallel to each other or twisted round each other into a rope shape
  - → chains held together with many crosslinks e.g. disulfide bridges
  - → provide strength, support, and flexibility (structural functions)

Collagen

in tissue that needs high tensile strength, flexibility, and stretch resistance
e.g. ligaments, connective tissue



Elastin

in elastic tissue to allow stretch and recoil
e.g. walls of arteries, elastic cartilage

Keratin

in skin and other external structures for strength and protection
e.g. hair, nails, horns

Biuret test for proteins

1) Add a few drops of Biuret reagent.
2) If protein is present: solution turns purple.
3) If no protein is present: solution stays blue.

PROTEINS

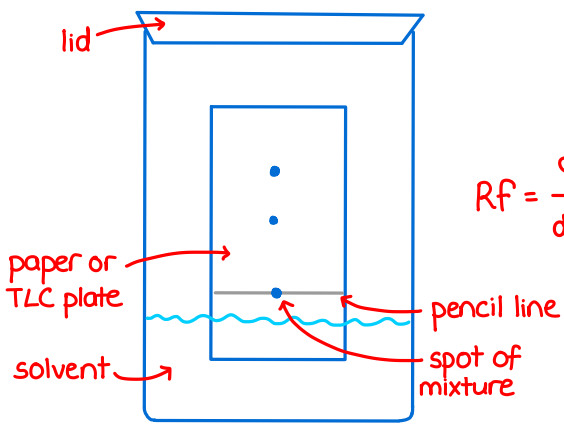
Separating and identifying amino acids using chromatography

- Chromatography separates molecules based on their different affinities for the stationary phase vs the mobile phase
  - → molecules can adsorb to stationary phase with different strengths
  - → molecules have different solubilities in the mobile phase
- Paper chromatography → stationary phase is paper, mobile phase can be water or an organic solvent
- Thin layer chromatography (TLC)
  - → stationary phase is silica gel, mobile phase is normally an organic solvent e.g. butanol
- Only R groups differ in amino acids
  - → charge and polarity of R groups determines interaction with the stationary and mobile phases
  - → amino acids with non-polar R groups are hydrophobic and will not dissolve in water (a polar solvent)
- An amino acid which is more soluble in the solvent will travel further because it spends more time dissolved in the solvent → solvents carry dissolved solutes through the stationary phase

Method:

1) Draw a pencil line near the bottom of the paper or TLC plate
   - → do not use pen because it will dissolve in the solvent
2) Put concentrated spots of your amino acid mixtures onto the pencil line
   - → make sure the spots are spread out so they do not merge
   - → let the spot dry and then repeat to build up a concentrated spot
3) Place the paper or TLC plate into the solvent
   - → make sure the pencil line is above the solvent so the spots do not dissolve into the solvent
   - → make sure the paper or TLC plate is supported to be kept level and at a constant height
4) Put a lid on the container to prevent the solvent from evaporating
5) Allow the solvent to move up until it is nearly at the top, then remove paper or TLC plate
   - → mark the solvent front with pencil before the solvent evaporates
   - → stain the paper or TLC plate with ninhydrin to visualise the amino acids
6) Calculate the Rf values
   - → repeat the experiment and find the average Rf value to increase accuracy
7) Look up Rf values in standard reference tables to find out which amino acids you have in the mixture
   - → Rf values are specific to a solvent so you have to look at the table relevant to your solvent

Molecules adsorb to the stationary phase, not absorb.



$$Rf = \frac{\text{distance moved by solute}}{\text{distance moved by solvent}}$$

measure from origin to centre of spot

measure from origin to solvent front

This process cannot be automated easily.

Chromatography can also be used to separate carbohydrates, vitamins and nucleic acids.

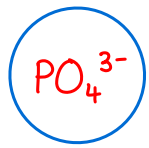
- Safety considerations
  - → wear gloves to protect skin and to prevent contamination (hands might have amino acids on them)
  - → work in a fume cupboard if using organic solvents or ninhydrin

INORGANIC IONS

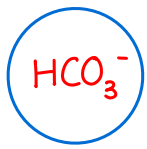
- Ions have electric charge → anions have negative charge
  → cations have positive charge
- Inorganic ions are soluble → dissolved in the fluids of an organism and in the cytoplasm

Examples

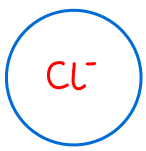
- Phosphate ions → attached to other molecules to become a phosphate group (phosphorylation)
  → found in ATP, DNA, RNA, phospholipids and calcium phosphate (in bones)
  → give DNA and RNA a negative charge
  → used in photosynthesis and respiration



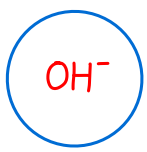
- Nitrate ions → source of nitrogen for plants (absorbed from the soil)



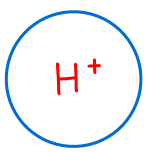
- Hydrogencarbonate ions → help to maintain pH of the blood by acting as a buffer



- Chloride ions → a cofactor for amylase
  → help to maintain pH of the blood (chloride shift)

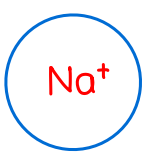


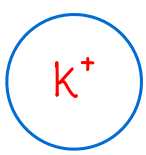
- Hydroxide ions → higher concentration of OH^- ions means a higher (more alkaline) pH
  → neutralise stomach acid
  → enzyme activity is affected by pH (and therefore OH^- concentration)



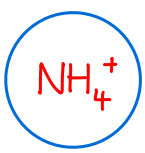
- Hydrogen ions → higher concentration of H^+ ions present means a lower (more acidic) pH
  → involved in chemiosmosis in respiration and photosynthesis
  → enzyme activity is affected by pH (and therefore H^+ concentration)

- Sodium ions → used in co-transport to help molecules cross membranes e.g. glucose absorption in the small intestine
  → needed to create an action potential in neurones
  → involved in regulation of fluid balance



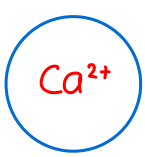


- Potassium ions → needed to create an action potential in neurones
  → involved in muscle contraction and regulation of fluid balance
  → activate some enzymes in photosynthesis



- Ammonium ions → source of nitrogen for plants (absorbed from the soil)

- Calcium ions → important role at synapses between neurones and in muscle contraction
  → a cofactor for some enzymes involved in coagulation (blood clotting)
  → important for the secretion of insulin from the pancreas



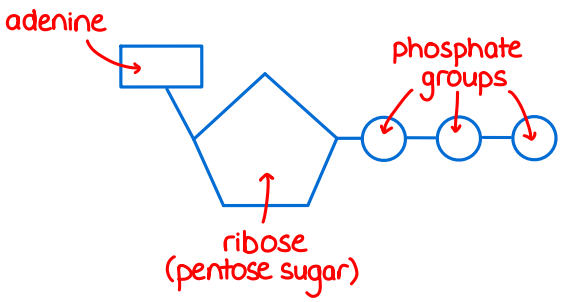
Inorganic normally means no carbon, but HCO_3^- is an exception.

You will come across the role of these ions throughout all modules.

ATP

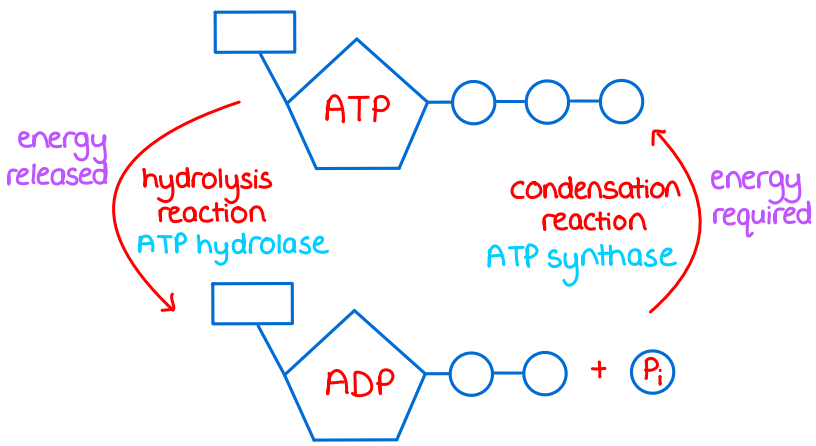
Structure

- ATP = adenosine triphosphate
- Contains carbon, hydrogen, oxygen, phosphorus and nitrogen
- A phosphorylated nucleotide
- A ribose sugar bound to adenine (a purine base) and three phosphate groups



Function

- Diffuses to areas of cells where energy is needed and does not leave the cell
- Hydrolysed to ADP (adenosine diphosphate) and P_i (inorganic phosphate)
  → energy is released for use in metabolic reactions
- Immediate energy supply → not stored long term
- Rapidly re-synthesised by phosphorylating ADP with P_i
  → requires energy released from glucose in respiration



Remember that water is produced in condensation and used in hydrolysis.

ATP and ADP can be referred to as phosphorylated nucleotides because they have a base, pentose sugar, and phosphate groups.

DNA AND RNA

- DNA = deoxyribonucleic acid, RNA = ribonucleic acid
- Contain carbon, hydrogen, phosphorous, oxygen and nitrogen

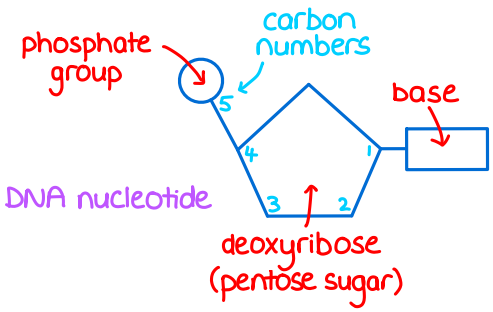
DNA nucleotides

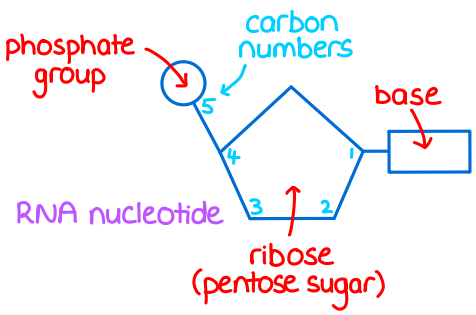
- The monomers of DNA
- Consist of a pentose sugar (deoxyribose), a nitrogenous base, and a phosphate group
- The base can be adenine (A), thymine (T), guanine (G) or cytosine (C)

RNA nucleotides

- The monomers of RNA
- Consist of a pentose sugar (ribose), a nitrogenous base, and a phosphate group
- The base can be adenine (A), uracil (U), guanine (G) or cytosine (C)

- Nucleotides join in condensation reactions to form polynucleotides
  - → phosphodiester bonds form between the phosphate group of one nucleotide and the pentose sugar (deoxyribose for DNA, ribose for RNA) of the next nucleotide
- Adenine and guanine are purine bases → contain two carbon-nitrogen rings
- Thymine, uracil and cytosine are smaller pyrimidine bases → contain one carbon-nitrogen ring

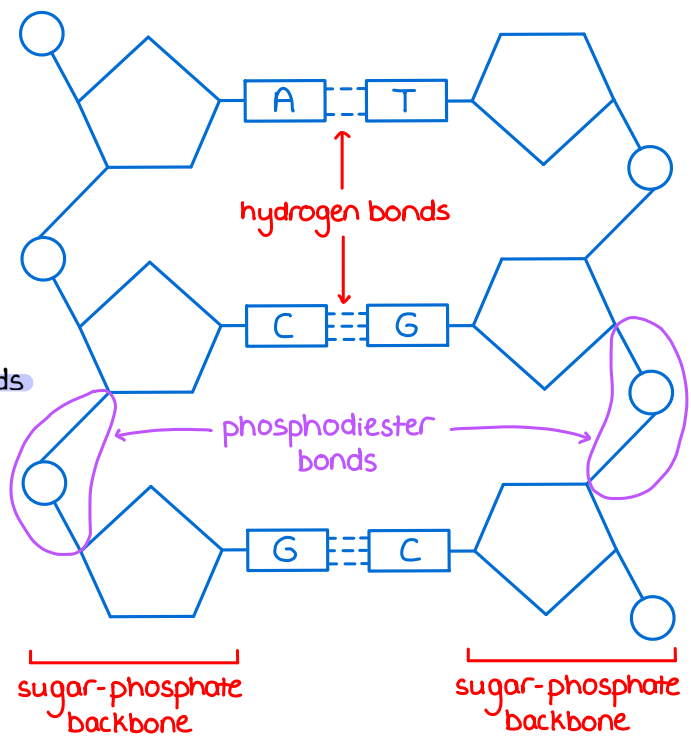




A pentose sugar has five carbon atoms.

DNA

- Stores genetic information
- Double-stranded polymer of DNA nucleotides
- Two antiparallel polynucleotide chains twisted into a double-helix structure
- The phosphate groups and pentose sugars form the sugar-phosphate backbone
- The bases join by complementary base pairing
  - → adenine pairs thymine with two hydrogen bonds
  - → cytosine pairs guanine with three hydrogen bonds
  - → holds the two strands together in the double-helix
  - → always the same amount of adenine and thymine and the same amount of cytosine and guanine in DNA



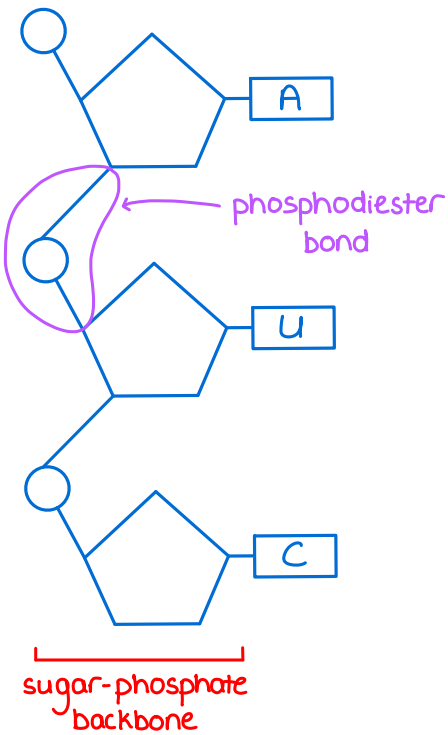
Antiparallel means the strands run in opposite directions: one is 5' to 3', the other is 3' to 5'. These numbers relate to the position of the carbon atoms in the pentose sugar.

The double-helix was discovered by Watson and Crick in 1953.

DNA AND RNA

RNA

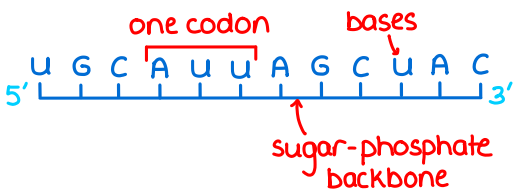
- A single polynucleotide chain with different functions
- Single-stranded polymer of RNA nucleotides
- The phosphate groups and pentose sugars form the sugar-phosphate backbone
- Complementary base pairing happens in transcription and translation
  → adenine pairs with uracil, cytosine pairs with guanine
- Messenger RNA (mRNA) transfers genetic information from DNA to the ribosomes
- Transfer RNA (tRNA) brings specific amino acids to ribosomes
- Ribosomal RNA (rRNA) is part of the structure of ribosomes along with proteins



Phosphodiester bonds are two ester bonds with a phosphate in the middle.

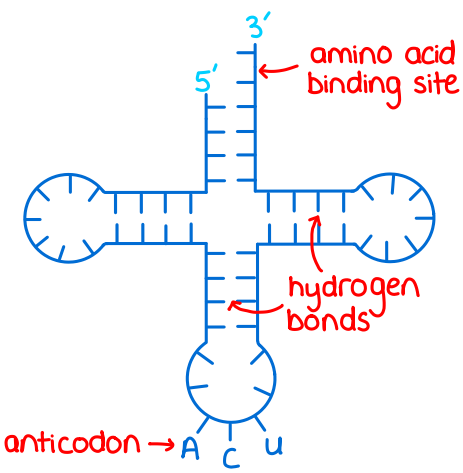
Messenger RNA

- A single linear polynucleotide strand made during transcription
- Can be different lengths
- Much shorter than DNA → can fit through the nuclear pores
- A three base sequence is a codon



Transfer RNA

- A single polynucleotide strand folded into a cloverleaf shape
- Hydrogen bonds between complementary bases hold the shape
- Contains an amino acid binding site and an anticodon
- Found in the cytoplasm



DNA AND RNA

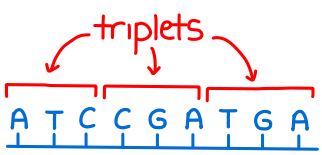
DNA purification

- A method to extract pure DNA from cells

1) If using plant cells, crush and grind the piece of plant using a pestle and mortar or blender
   → this breaks the cell walls
2) Mix with a solution of detergent and salt, and incubate in a 60°C water bath for about 15 mins
   → detergent breaks cell membranes to release the contents of the cells and nuclei
   → salt makes DNA less soluble in water
3) Transfer to an ice bath to cool then filter the solution
   → low temperature reduces the activity of enzymes which could break down DNA
4) Add protease and RNAse enzymes to the filtered solution, then add ice cold ethanol
   → protease hydrolyses the histone proteins associated with DNA
   → RNAse hydrolyses any RNA present
   → ethanol precipitates the DNA from solution and it will be visible as a white layer which can be removed carefully

The genetic code

- Triplet code → a triplet of bases codes for one amino acid
  → sequence of bases determines primary protein structure
- Non-overlapping → each nucleotide is only part of one triplet of bases, there is never overlap
- Degenerate → more than one triplet codes for each specific amino acid
- Universal → the same triplet codes for the same amino acid in all species
- A gene is a sequence of DNA that codes for a polypeptide

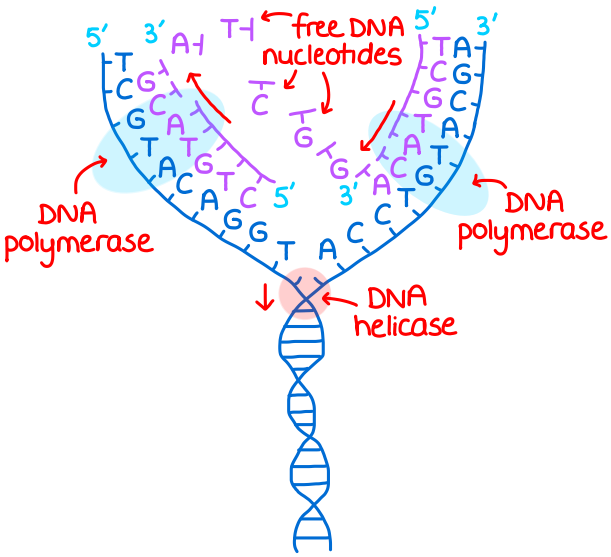


CTT CTG
CTC TTA
CTA TTG
} all code for leucine (an amino acid)

DNA REPLICATION

Semi-conservative replication

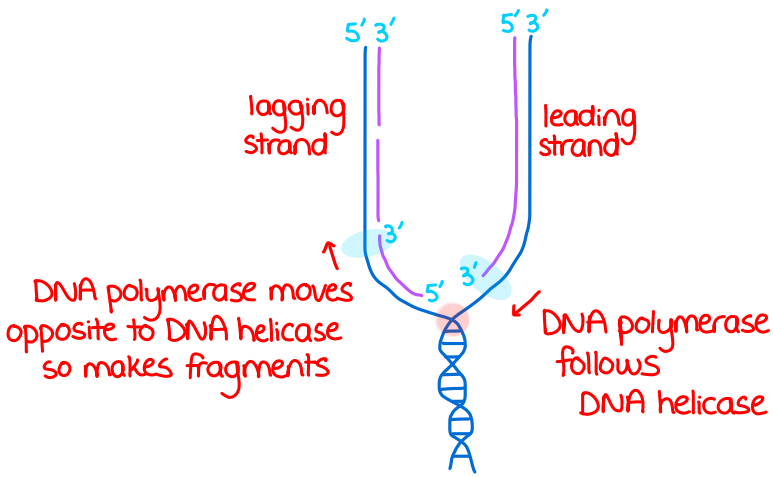
- The Watson-Crick model shows that DNA replication is semi-conservative
  - → each new DNA molecule has one strand from the original DNA molecule and one newly synthesised strand
- Both strands of the DNA act as template strands and determine the order of bases



1) DNA helicase breaks weak hydrogen bonds between complementary bases so the double-helix unzips and the two strands separate. Both strands will act as template strands.
2) Free DNA nucleotides are attracted to exposed bases on the two template strands and pair up by complementary base pairing.
3) DNA polymerase joins the adjacent nucleotides with phosphodiester bonds in condensation reactions to form the sugar-phosphate backbone.
4) The DNA winds into the double-helix shape.

- Complementary base pairing makes sure that DNA replication is normally very accurate
  - → a purine must pair with a pyrimidine because they are different sizes
  - → the right number of hydrogen bonds must be able to form, so each base only has one option
- Mutations sometimes occur spontaneously and randomly during DNA replication
  - → the sequence of DNA bases changes due to wrong complementary pairs being matched
  - → mutagenic agents (e.g. UV light) can increase the rate of mutations
  - → a change to the sequence of DNA bases could alter the structure and function of the protein
- DNA polymerase can only form phosphodiester bonds in the 5' to 3' direction of the new strands
  - → it can only add nucleotides at the 3' end due to enzyme specificity
  - → the leading strand is made in one piece
  - → the lagging strand is made in fragments
  - → fragments are joined with phosphodiester bonds by DNA ligase

The fragments are called Okazaki fragments.

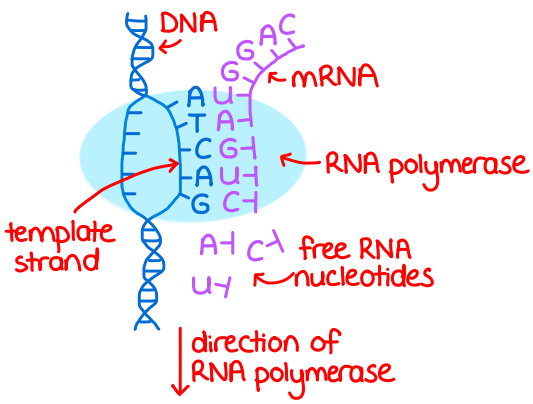


PROTEIN SYNTHESIS

Transcription

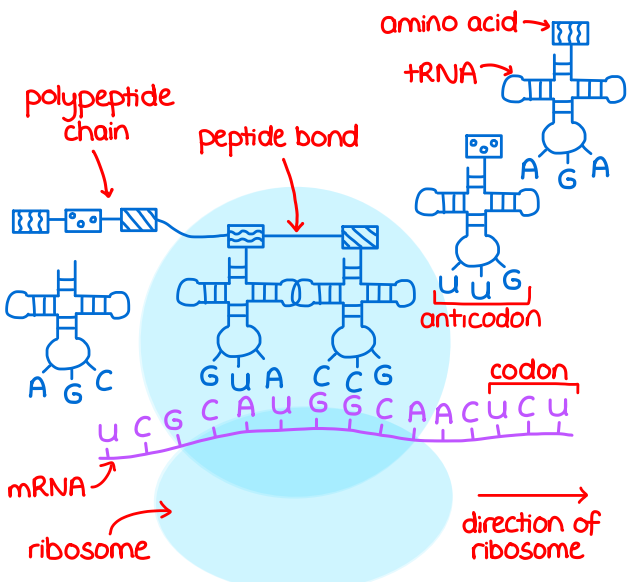
- Transcribing DNA to mRNA
- Happens in the nucleus in eukaryotic cells and the cytoplasm in prokaryotic cells
- DNA is too large to fit through the nuclear pores → mRNA is shorter so can travel to the ribosomes

1) RNA polymerase attaches to DNA at the start of a gene.
2) Hydrogen bonds between the DNA strands break leaving exposed bases on the template strand.
3) Free RNA nucleotides complementary base pair with the exposed bases.
4) RNA polymerase joins the RNA nucleotides together with phosphodiester bonds in condensation reactions.
5) RNA polymerase moves along the DNA until it reaches the end of the gene, then detaches.



Translation

- Translating mRNA into a polypeptide
- Happens at the ribosomes



1) The mRNA from the nucleus travels to the ribosomes and the ribosome attaches at the start codon.
2) tRNA molecules bring specific amino acids to the ribosomes → joining of an amino acid to tRNA requires energy from ATP.
3) The anticodon on the tRNA binds to the codon on the mRNA with complementary base pairing.
4) A second tRNA molecule lines up next to the first and a peptide bond forms between the amino acids (in a condensation reaction catalysed by rRNA).
5) The ribosome moves along the mRNA until it reaches the stop codon, then detaches.

mRNA codons are complementary to DNA triplets:

DNA triplet ATC
mRNA codon UAG

tRNA anticodons are complementary to mRNA codons:

mRNA codon UAG
tRNA anticodon AUC

Ribosomes are made of ribosomal RNA (rRNA) and protein.

ENZYMES

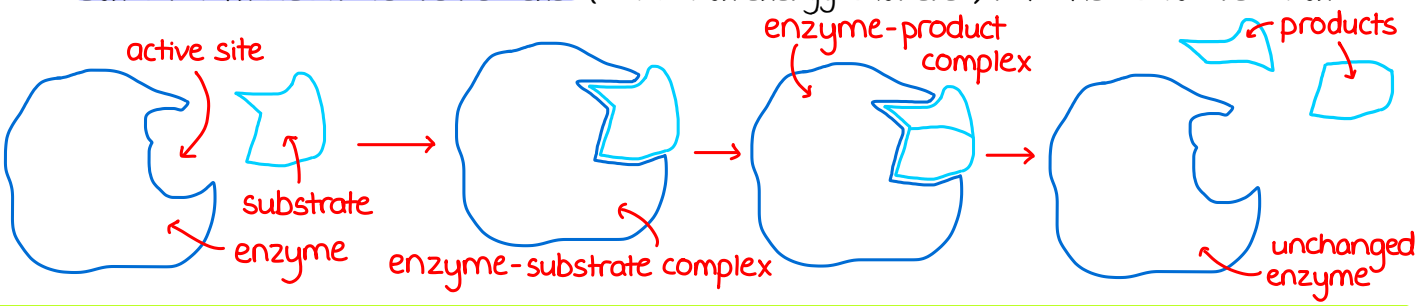
Function of enzymes

- Enzymes are globular proteins with a specific tertiary structure
- The active site is complementary to one specific substrate
- Biological catalysts → speed up a reaction by lowering the activation energy
- Can be intracellular (act inside cells) or extracellular (act outside cells)
- Determine structure and function of cells and whole organisms

Very few substrates would have enough energy to react without an enzyme.

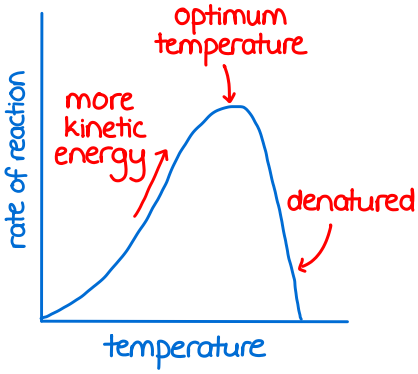
Induced fit model

- Replaced the lock and key model (where the enzyme and substrate are exactly complementary shapes)
- The active site is not fully complementary before reaction
  → shape of the active site changes as substrate binds to form the enzyme-substrate complex
- Formation of an enzyme-substrate complex forms stronger bonds between enzyme and substrate
  → bonds within substrate are weakened (activation energy is lowered) which leads to a reaction



Temperature

- As temperature increases, enzyme and substrate have more kinetic energy so there are more frequent successful collisions between substrate and active site, and more enzyme-substrate complexes form
- After the optimum temperature, the enzyme becomes denatured
  → too much kinetic energy breaks the hydrogen bonds and ionic bonds between amino acid R groups
  → the tertiary structure of the enzyme changes, and the active site changes shape so it is no longer complementary to the substrate
  → enzyme-substrate complexes cannot form
- Effects of low temperature are reversible, high temperature are not
- The temperature coefficient (Q_{10}) tells you by what factor the rate of reaction changes when you increase the temperature by 10°C

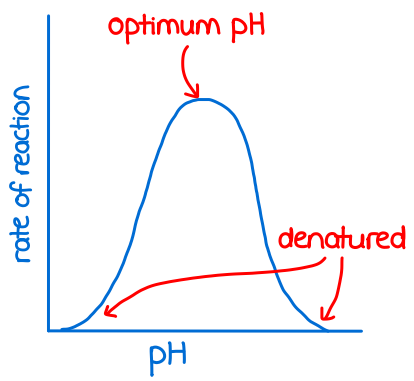


$$Q_{10} = \frac{R_2 \leftarrow \text{rate at temperature + 10°C}}{R_1 \leftarrow \text{rate at temperature}}$$

For many enzyme-controlled reactions, $Q_{10} = 2$ (rate doubles every 10°C increase).

pH

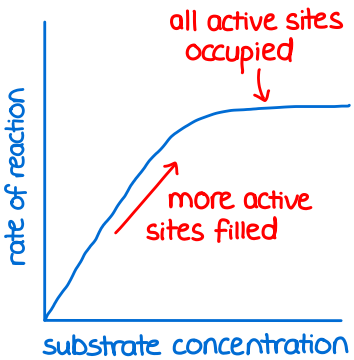
- Enzymes are denatured above and below the optimum pH
  → H^+ (acidic) or OH^- (alkali) ions interfere with the hydrogen bonds and ionic bonds between amino acid R groups
  → the tertiary structure of the enzyme changes, and the active site changes shape so it is no longer complementary to the substrate
  → enzyme-substrate complexes cannot form
- Hydrogen bonds can reform if pH returns to the optimum pH
  → effects of small pH changes are sometimes reversible

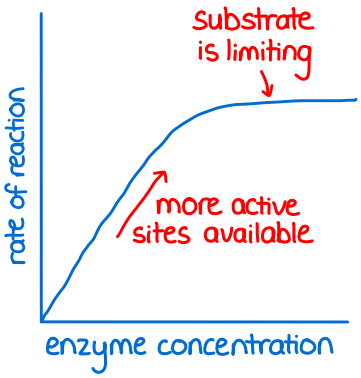


ENZYMES

Enzyme or substrate concentration

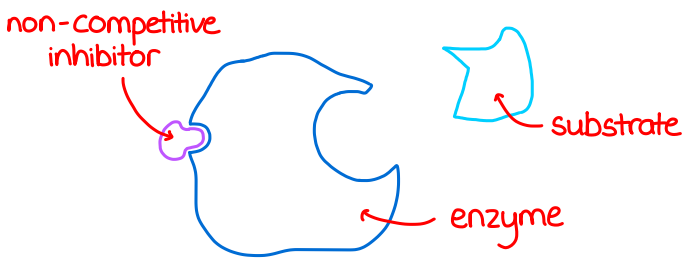
- Increasing enzyme or substrate concentration increases the frequency of collisions between the active site and the substrate, so more enzyme-substrate complexes form
- Eventually all available active sites are filled, or substrate concentration becomes limiting, so rate of reaction plateaus

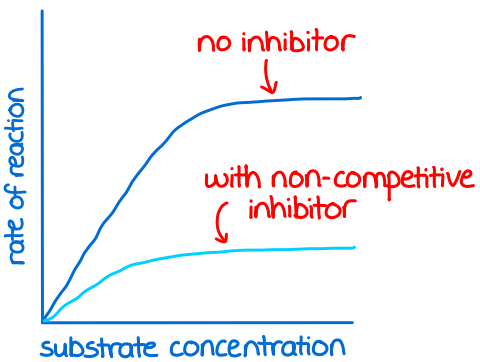




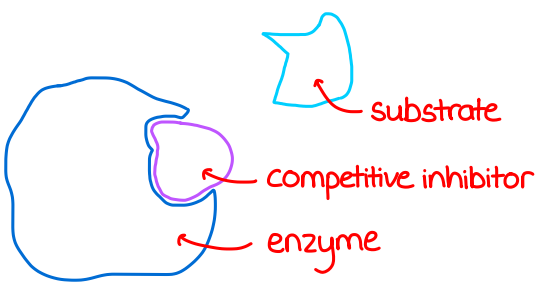
Inhibitors

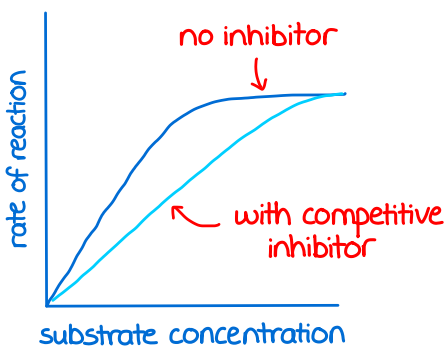
- Non-competitive inhibitors
  - → bind to an allosteric site on the enzyme (not the active site)
  - → alter the tertiary structure so the active site changes shape
  - → active site no longer complementary to substrate, so less enzyme-substrate complexes form
  - → increasing substrate concentration does not increase the rate of reaction





- Competitive inhibitors
  - → have a very similar shape to the substrate
  - → bind to the active site in place of the substrate so less enzyme-substrate complexes form
  - → increasing substrate concentration will increase the rate of reaction (because the substrate "wins" the competition more often)



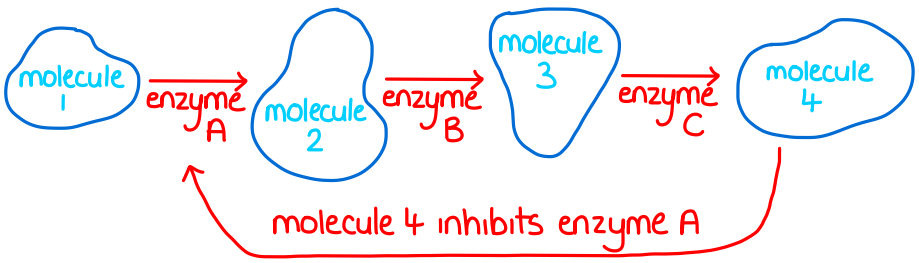


- Non-reversible inhibitors bind to the enzyme with strong covalent bonds
- Reversible inhibitors bind to the enzyme with weak hydrogen bonds

ENZYMES

End-product inhibition

- When the final product in a metabolic pathway inhibits an enzyme further up the pathway
- Helps to regulate the pathway and control the amount of products
- A type of reversible inhibition



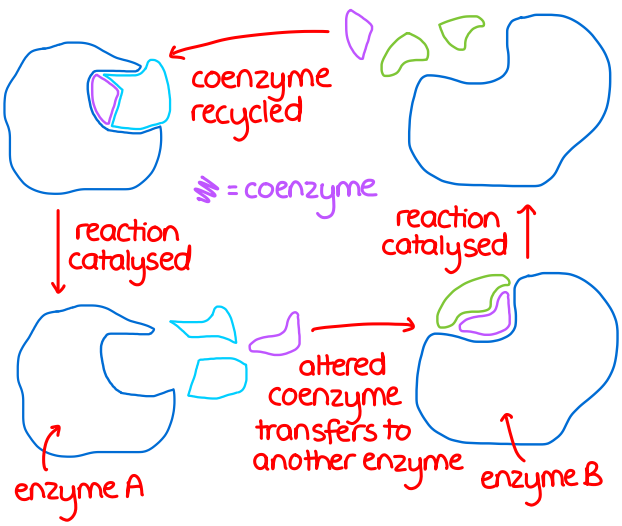
A metabolic pathway is a linked series of reactions happening in a cell.

Cofactors

- Non-protein inorganic molecules or ions which bind to enzymes
- Help the substrate to bind to the enzyme and increases catalytic activity
- Called a prosthetic group if tightly bound to the enzyme with strong bonds e.g. covalent bonds
- Not used up or changed in the reaction
- Chloride ions (Cl^-) → cofactor for amylase

Coenzymes

- Non-protein organic molecules which bind to enzymes but are not permanently attached
- Assist an enzyme to catalyse the reaction e.g. allow the substrate to bind to the enzyme more easily
- Transfer chemical groups between enzymes and are continually recycled
- Many vitamins are coenzymes



You will come across coenzymes NAD, FAD, and NADP in Module 5.

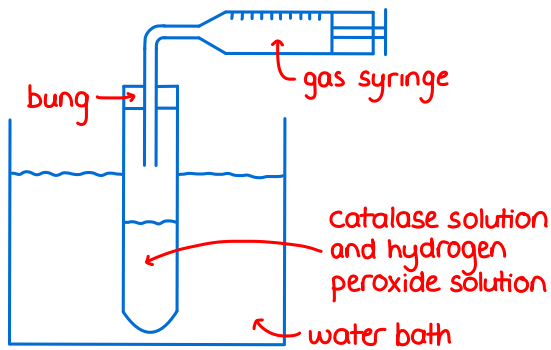
Specific enzyme examples

- Catalase → an intracellular enzyme
  → catalyses the conversion of toxic hydrogen peroxide to non-toxic oxygen and water
- Amylase → an extracellular enzyme secreted by the salivary glands and pancreas
  → hydrolyses starch to maltose in digestion (hydrolyses glycosidic bonds)

ENZYMES

Investigating enzyme-controlled reactions

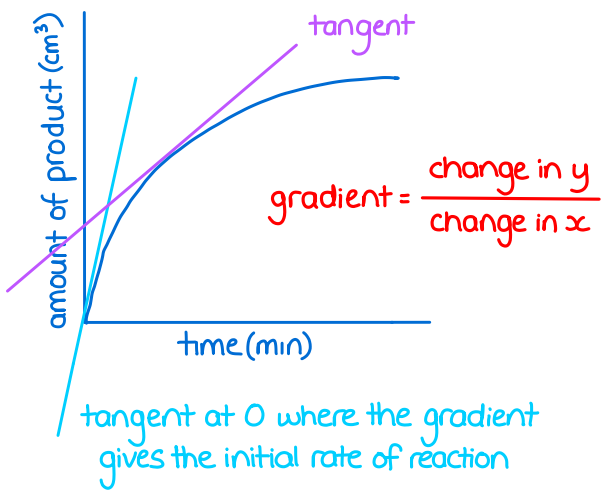
- Many different possibilities for what to investigate and which reaction to use
- Only change one variable at a time and control all other variables which could affect enzyme activity
- Example: the enzyme catalase converts toxic hydrogen peroxide to harmless oxygen and water
  - → could use a gas syringe to record the volume of oxygen gas produced over time and repeat at a range of different temperatures
  - → have a negative control experiment with the same volume and concentration of denatured catalase to show no oxygen is produced without the active enzymes
- Possible control variables
  - → volume of the substrate solution
  - → concentration of the substrate solution
  - → volume of the enzyme solution
  - → concentration of the enzyme solution
  - → temperature of the solutions (use a water bath)
  - → pH of the solutions (use a buffer solution)



Control variables depend on the investigation. Be specific to the question.

Always repeat the experiment and calculate a mean.

- Rate of reaction can be calculated by finding the gradient of a line
  - → draw a tangent at zero to find the initial rate of reaction
  - → draw a tangent at any part of the curve to find the rate at that specific point
- Initial rate of reaction is highest because plenty of substrate is available
  - → initially very frequent collisions between enzyme and substrate and many enzyme-substrate complexes form
  - → rate decreases as the substrate concentration decreases and there are less frequent collisions
  - → the reaction stops when there is no substrate left



In this example the units of rate would be $cm^3\ min^{-1}$.