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Section 1 - ① Biological Molecules

1.1 - Introduction to Biological Molecules

Biological Molecules - particular group of chemicals that are found in living organisms

Different Bondings that form molecules:

- **Covalent Bonding**; shared pair of electrons in outer shells, which fills it, forming a stable compound (molecule)

- **Ionic Bonding**; strong electrostatic forces of attraction between oppositely charged ions (weaker than covalent)

- **Hydrogen Bonding**: electrons in a molecule are not evenly distributed and spend more time in one position, making the region more negatively charged - unevenly distributed charge in a molecule makes it polarised (polar molecule)
  - two oppositely charged regions of polar molecules attract, forming a weak electrostatic bond (collectively can be important forces that alter physical properties)

Monomers → smaller units from which larger molecules are made

↓ linked together to form long chains - polymerisation

Polymers → molecules made from large numbers of monomer sub-units joined together

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examples of monomers and their polymers:

- monosaccharides
  - ↳ polysaccharides

- amino acids
  - ↳ proteins / poly-peptides

- nucleotides
  - ↳ polynucleotides

Condensation and Hydrolysis Reactions:

condensation reaction → joins two molecules together with the formation of a chemical bond and involves the elimination of a water molecule

hydrolisis reaction → breaks the chemical bonds, in a polymer, between two molecules by the addition of a water molecule, splitting molecule into its constituent parts

Metabolism → all the chemical processes that take place in a living organism collectively

Moles and Molar Solution

mole (mol) is the SI unit for measuring the amount of a substance
- ↳ contains 6.022×10^{23} particles (Avogadro Constant)

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molar solution (M) is a solution that contains one mole of solute in one litre of solution

1 mole = molecular mass (g) = Mr of 1 molecule

1.2 - Carbohydrates (Monosaccharides)

Carbohydrates - carbon molecules combined with water

carbon - consisting molecules → organic molecules

- carbon readily forms bonds with other carbons

carbohydrate = polymer
- basic monomer unit is a sugar (saccharide)
- single monomer is a monosaccharide
  - a pair forms a disaccharide
- can be combined into larger numbers to form polysaccharides

Monosaccharides

- sweet-tasting, soluble substances
- general formula: $(CH_2)_n$ → n is a number from 3 to 7

Examples of monosaccharides: glucose, fructose, galactose

glucose is a hexose (6-carbon) sugar
- $C_6H_{12}O_6$

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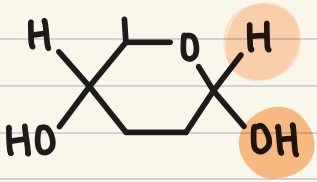
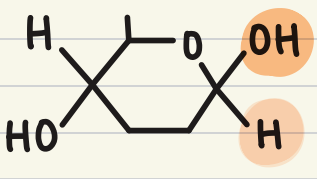
→ atoms of carbon, hydrogen and oxygen can be re-arranged

Glucose has 2 isomers - α and β glucose;



α - glucose



β - glucose

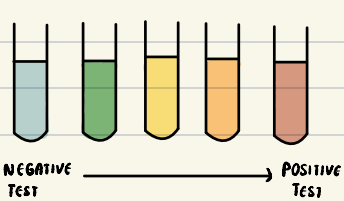
Test for reducing sugar:

reducing sugar - sugar that can (reduce or) donate electrons to another chemical (ie. Benedict's reagant)

TEST: Benedict's test

→ benedict's reagant is an alkaline solution of copper (II) sulfate - when reducing sugar is heated and reacted with, an insoluble red precipitate (copper (I) oxide) forms

- $2cm^3$ of food sample put in test tube, if not already in liquid form, mix with water
- add equal volume of benedict's reagant
- heat mixture gently in water bath



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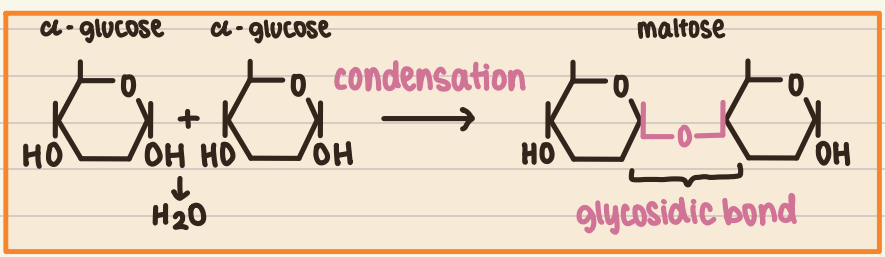
1.3 - Carbohydrates - Disaccharides and Polysaccharides

Disaccharides

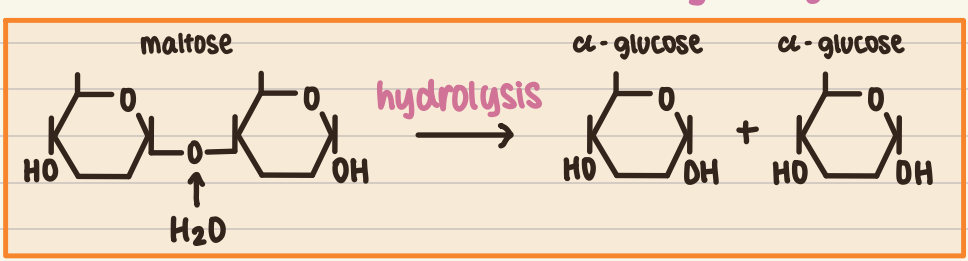
└ formed from a condensation reaction between two monosaccharides;

- glucose + glucose = maltose
- glucose + fructose = sucrose
- glucose + galactose = lactose

Monosaccharides joining is a condensation reaction and it forms a glycosidic bond
└ molecule of water lost



Adding water to a disaccharide under suitable conditions breaks glycosidic bond and splits into the constituent monosaccharides - hydrolysis reaction



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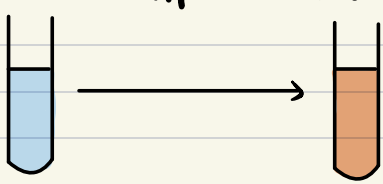
Testing for non-reducing sugars:

Some disaccharides are **non-reducing sugars** (ie. sucrose)
↳ do not change colour of Benedict's reagant when heated with it

TEST: Must be hydrolysed into its monosaccharides first

→ first carry out a **Benedict's test**, if **no colour change**, it **isn't** a **reducing sugar**
- add **2cm³** of **food sample** to **2cm³** of **HCl (dilute)** in a test tube placed in a **boiling water bath** for **5 mins**
  ↳ will hydrolyse disaccharide
- slowly add some **sodium hydrogencarbonate** solution to **neutralise acid (HCl)** - test with **pH paper** to check if it is alkaline - Benedict's reagant doesn't work in acidic conditions
- Retest with **2cm³** of Benedict's reagant; if there was a non-reducing sugar in original sample - solution will turn **orange-brown**



Polysaccharides

↳ **polymers** formed by **combining** together **many monosaccharide molecules**
- form **glycosidic bonds** from **condensation reactions**
- **large molecules** → **insoluble** (suitable for storage)
- when **hydrolysed** break down into **mono/di-saccharides**
- some polysaccharides, ie. cellulose, **give structural support**

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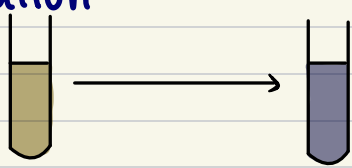
for plant cells
- starch is a polysaccharide that is found in many parts of plants in the form of small granules / grains
  - formed by a condensation reaction between 200-100000 α-glucose molecules

Test for starch:

→ Starch is easily detected by its ability to change the colour of the iodine in potassium iodide solution from yellow / orange to blue / black
- place 2cm³ of food sample into a test tube / 2 drops into depression of spotting tile
- add two drops of iodine solution and shake or stir
- presence of starch → blue / black colouration



1.4 - Starch, Glycogen, and Cellulose

Starch
- polysaccharide that is found in many parts of a plant in the form of small grains
- large amounts occur in seeds and storage organs
- forms an important component of food and is the major energy source
- made up of chains of α-glucose monosaccharides linked by glycosidic bonds that are formed by condensation reactions
  - can be branched or unbranched
- unbranched chain is wound up into a tight coil that makes the molecule very compact

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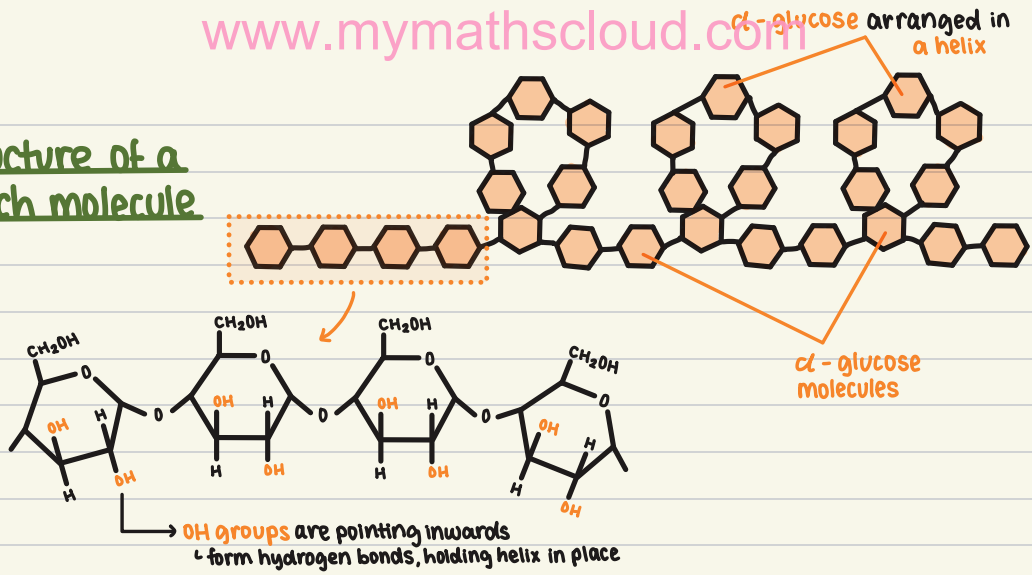
Structure of a starch molecule



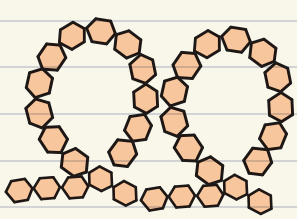
Main role of starch is **energy storage**, its structure is suited for this function, because:

- it is **insoluble** and therefore **doesn't affect water potential**, so water is not drawn into the cells by **osmosis**
- large + insoluble, doesn't diffuse out of cells
- it is **compact**, so a lot of it can be stored in **a small space**
- when **hydrolysed** it forms **α-glucose**, which is both easily transported and readily used in respiration
- the **branched form** has **many ends**, each of which can be acted on by **enzymes simultaneously** meaning that glucose monomers are **released rapidly**

Two structures of starch:

Amylose: formed by **a series of condensation reactions** that bond **α-glucose molecules** together into a long chain forming **many glycosidic bonds** - once chain formed, coils into a helix

- **1-4 α-glycosidic bonds**, (unbranched chain wound into a **helix**)



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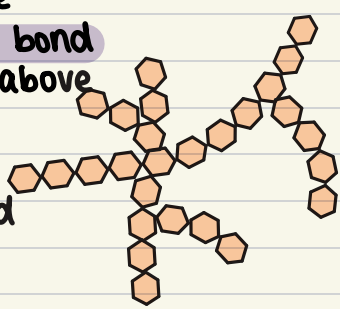
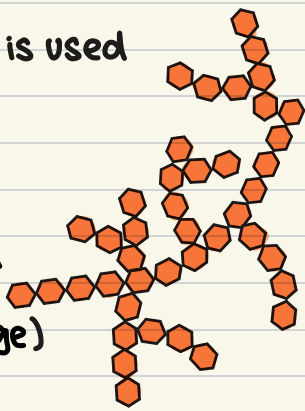
Amylopectin: consists of a straight chain of α-glucose units with branch points occuring at approximately every twelvth glucose unit along the straight chain
- ↳ branch points form when carbon 6, of a glucose molecule in the straight chain, forms a glycosidic bond with a carbon 1 of a glucose molecule positioned above the chain
- 1-4 and 1-6 α-glycosidic bonds (highly branched molecule amylopectin is wrapped around amylose to make up starch)



Glycogen

- Starch not found in animal cells; instead glycogen is used
  - ↳ found in animals and bacteria
- similar structure to starch, but has shorter chains and is more highly branched + major carbohydrate storage product for animals ('animal starch')
- stored as small granules mainly in the muscles and liver
  - ↳ mass relatively small compared to fat (main storage)



Structure suited to storage, because:

- it is insoluble and therefore doesn't tend to draw water into cells by osmosis
- being insoluble, it doesn't diffuse out of cells
- it is compact, so lot can be stored in a small space
- more highly branched than starch, so has more ends for enzymes to act on simultaneously, therefore more rapidly broken down to form glucose monomers, used in respiration
  - ↳ important as animals have a higher metabolic rate than plant → 1-4 and 1-6 α-glycosidic bonds

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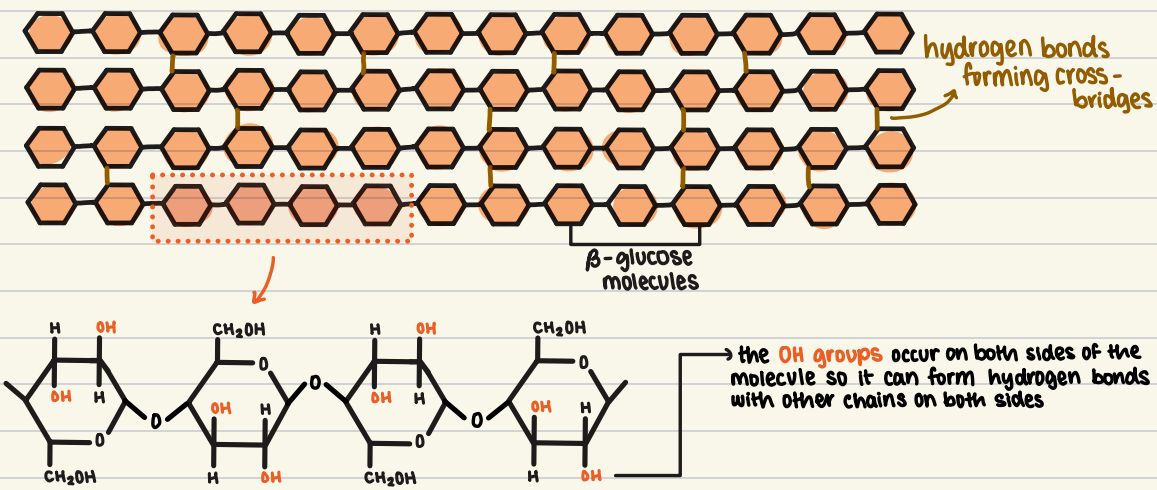
Cellulose

- made up of **β-glucose monomers** - has **straight, unbranched chains**
- chains **run parallel** to one another, allowing **hydrogen bonds** to form **cross-linkages** between **adjacent chains**
- **each, individual hydrogen bond** adds very little to the strength of the molecule, the sheer overall number of them makes a considerable contribution to **strengthening cellulose** - makes a **valuable structural material**



- the **cellulose molecules** are grouped together to form **microfibrils**, which are arranged in **parallel groups** called **fibres**
- cellulose is a major component of **plant cell walls** and provides **rigidity** to the plant cell
  - cellulose cell walls **prevents the cell from bursting** as water **enters** by **osmosis**
- it does this by **exerting an inward pressure** that stops any further **influx of water**

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- living plant cells are turgid and push against one another, making non-woody parts of the plant semi-rigid
  - especially important in maintaining stems and leaves in a turgid state so that they can provide the maximum surface area for photosynthesis

Structure suited to function, because:

- cellulose molecules are made up of β-glucose and so form long straight, unbranched chains
- these cellulose molecular chains run parallel to each other and are crossed linked by hydrogen bonds which add collective strength
- these molecules are grouped to form microfibrils which in turn are grouped to form fibres all of which provides yet more strength

1.5 - Lipids

common characteristics:

- contain carbon, hydrogen, oxygen
- proportion of oxygen to carbon and hydrogen is smaller than carbohydrates
- they are insoluble in water
- soluble in organic solvents such as alcohols and acetone

Main groups of lipids are triglycerides (fats and oils) and phospholipids

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Roles of Lipids:

- have many roles, one role is in the cell membranes (cell-surface membranes / membranes around organelles)
- phospholipids contribute to the flexibility of membranes and the transfer of lipid soluble substances accross them

Other roles of lipids;

- source of energy - when oxidised, lipids provide more than twice the energy as the same mass of carbohydrate and release valuable water
- waterproofing - lipids are insoluble in water and therefore useful as a waterproofing (e.g. plant's waxy cuticle and mammals produce an oily secretion from sebaceous glands in the skin
- insulation - fats are slow conductors of heat and when stored beneath the body surface help to retain body heat
  - act as electrical insulators (e.g. myelin sheath around nerve cells)
- protection - stored around delicate organs (e.g. kidney)

- fats solid at room temperature (10-20°C) whereas oils are liquids

Tryglycerides

- three fatty acids 'tri-' combined with glycerol (-glyceride)
- each fatty acid forms an ester bond with glycerol in a condensation reaction
- hydrolysis converts triglycerides back into its individual components

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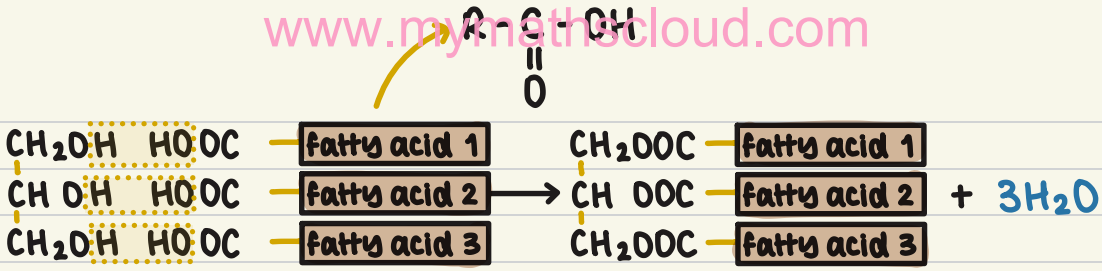

glycerol + 3 fatty acids ⟶ tryglyceride + 3 waters

↳ glycerol in all tryglycerides is the same, the differences in properties of different fats and oils comes from variation in the fatty acids - 70 different fatty acids and all have carboxyl group (-COOH) with a hydrocarbon chain attached; if chain as no C=C double bonds, it is saturated as all carbon atoms are linked to maximum possible number of hydrogen atoms

- single C=C → mono-unsaturated
- multiple C=C double bonds → polyunsaturated

↳ double bond allow molecules to bend, therefore cannot be packed closely together, so they are liquid at room temperature
↳ ie. Oils

Structure of tryglycerides related to their property;

- have a high ratio of energy-storing carbon-hydrogen bonds to carbon atoms, therefore an excellent source of energy
- have low mass to energy ratio, making them good storage molecules because much energy can be stored in a small volume
- being large non-polar molecules, tryglycerides are insoluble in water, therefore their storage doesn't affect osmosis in cells or their water potential
- high ratio of hydrogen to oxygen atoms, so triglycerides release

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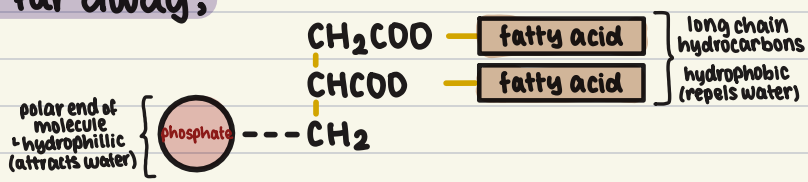
water when oxidised and therefore provide an important source of water (e.g. for animals in dry deserts

Phospholipids

↳ similar to lipids, but one fatty acid molecule is replaced by a phosphate molecule - fatty acid molecules are hydrophobic (repel water) and phosphate molecules are hydrophillic (attract water)
Two parts of a phospholipid:
- hydrophyllic 'head' - interacts with water but not with fat
- hydrophobic 'tail' - orients itself away from water but mixes readily with fat

Molecules that have two ends (poles) that behave differently in this way are said to be polar
↳ when polar phospholipid molecules are placed in water they position themselves so 'head' is as close to water as possible and tails as far away;



↳ structure of phospholipid related to their properties;

- phospholipids are polar molecules, having a hydrophyllic phosphate head and a hydrophobic tail of two fatty acids, therefore in an aqueous environment, phospholipid molecules form a bilayer within cell-surface membranes, creating a hydrophobic barrier is formed between inside and outside of a cell

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- hydrophillic phosphate 'heads' of phospholipid molecules help to hold at the surface of the cell-surface membrane
- phospholipid structure allows them to surface to form glycolipids by combining with carbohydrates within the cell-surface membrane
  - glycolipids important in cell recognition

Test for Lipids:
- emulsion test

1. Take completely dry and grease-free test tube
2. Add $5cm^3$ ethanol to $2cm^3$ of the sample
3. Shake tube thoroughly to dissolve any lipid into the sample
4. Add $5cm^3$ of water and shake gently
5. Milky-white emulsion indicates the presence of a lipid
6. As a control, repeat the procedures using water instead of the sample, final solution should be clear

- cloudy colour is due to any lipid in the sample being finely dispersed in the water to form an emulsion - light passing through this emulsion is refracted as it passes from oil droplets to water droplets, making it appear cloudy

1.6 - Proteins

Proteins are usually very large molecules

Structure of an amino acid:

- amino acids are basic monomer units which combine up to make

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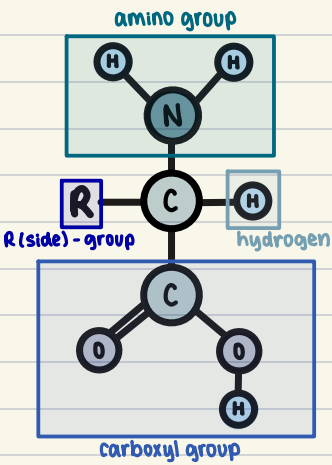
up a polymer, called a polypeptide
- polypeptides can be combined to form proteins

→ 100 identified amino acids of which 20 are naturally occurring in proteins - same that occur in all living organisms (indirect evidence for evolution)

→ Every amino acid has a central carbon atom to which are attached 4 different chemical groups;



- amino group ($-NH_2$) - basic group from which the name 'amino' acids is derived from
- carboxyl group ($-COOH$) - an acidic group from which the name amino 'acids' is derived from
- hydrogen atom ($-H$)
- R (side) group - variety of different chemical groups and each amino acid has a different R group - the 20 naturally occuring amino acids differ only in their R (side) group

Formation of a peptide bond:

2 amino acid monomers form a dipeptide
- made by the removal of a water molecule - condensation reaction
  - → water molecule made by combining an $-OH$ from the carboxyl group of one amino acid with an $-H$ from the amino group of another amino acid
  - these two amino acids become linked by a new peptide bond between the carbon atom of one amino acid and the nitrogen atom

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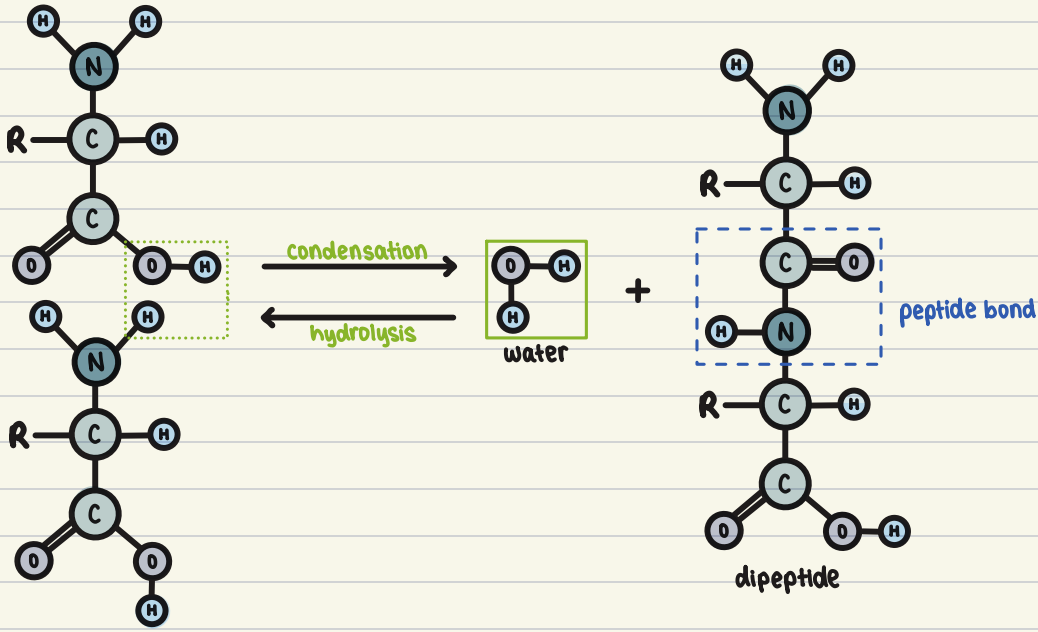
Of the other - the dipeptide can be broken down into its two constituent amino acids by the addition of water (hydrolysis)



Primary structure of proteins - polypeptides

- through a series of condensation reactions, many amino acid monomers can be joined together in a process called polymerisation
- resulting chain of many hundreds of amino acids is called a polypeptide
- sequence of amino acids in a polypeptide chain forms the primary structure of any protein (determined by DNA)
- almost limitless number of possible combinations of arranging the 20 amino acids, there are therefore many possible primary protein structure

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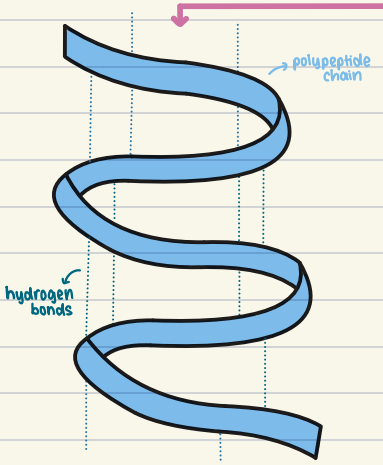
- primary structure of a protein that determines its ultimate shape and hence its function
  - a change in a single amino acid in the primary sequence can lead to a change in the shape of the protein and may stop it carrying out its function - protein's shape is very specific to its function
- a simple protein may consist of a single polypeptide chain
  - however commonly a protein is made up of a number of polypeptide chains

Secondary structure of proteins

- the linked amino acids that make up a polypeptide possess both $-NH$ and $-C=O$ groups on either side of every peptide bond
  - the hydrogen of the $-NH$ group has an overall positive charge while the O of the $-C=O$ group has an overall negative charge
- these two groups therefore readily form weak bonds, called hydrogen bonds
  - causes the long polypeptide chain to be twisted into a 3D shape such as the coil known as an α-helix or can be a β-pleated sheet



Tertiary structure of proteins

- α-helices of the secondary protein structure can be twisted and folded even more to give the complex, and often specific, 3D-structure of each protein
  - maintained by a number of different bonds (between R group)

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• where the bonds of the tertiary structure occur, depends on the primary structure of the protein, these bonds include:

→ disulfide bridges - fairly strong and not easily broken
→ ionic bonds - formed between any carboxyl and amino groups that are not involved in forming peptide bonds, they are weaker than disulfide bonds and are easily broken by changes in pH
→ hydrogen bonds - numerous but easily broken
+ hydrophobic interactions

↳ it is the 3D shape of a protein that is important when it comes to how it functions, it makes each protein distinctive and allows it to recognise, and be recognised by, other molecules
- can then interact with them in a very specific way

Quaternary Structure of proteins

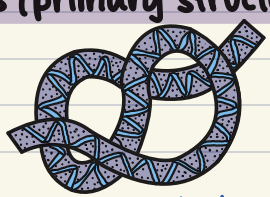
↳ large proteins often form complex molecules containing a number of individual polypeptide chains that are linked in various ways
- may also be non-protein (prosthetic) groups associated with the molecules
- 3D structure is important is important to how a protein functions, it is the sequence of amino acids (primary structure) that determines the 3D shape in the first place



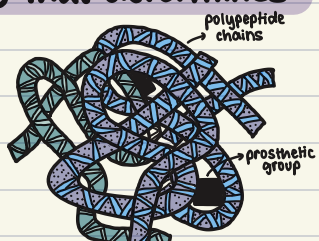
primary structure = amino acids in a polypeptide chain
- sequence determines its properties and shape



secondary structure = shape of the polypeptide chain after it forms hydrogen bonds - most often α-helix



tertiary structure = bending and twisting of polypeptide helix into a compact structure
- different bonds contribute to maintenance of tertiary structure



quaternary structure = combination of polypeptide chains and associated non-proteins (prosthetic) groups into a large, complex molecules protein

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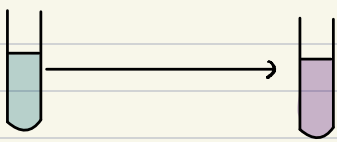
Types of Proteins:

Fibrous proteins - structural functions - elongated shape + insoluble
- e.g. collagen (3 polypeptide chains)

Globular proteins - metabolic functions - spherical shape + soluble
- e.g. haemoglobin (4 polypeptide chains + iron prosthetic group)

Test for proteins:

- biuret test, which detects peptide bonds;



- place a sample of the solution to be tested in a test tube and add an equal volume of sodium hydroxide solution at a random temperature
- add a few drops of very dilute (0.05%) copper (II) sulfate solution and mix gently
- a purple colouration indicates the presence of peptide bonds and hence a protein, if no protein is present, the solution remains blue

1.7 - Enzyme Action

Enzymes are globular proteins that act as catalysts
- alter the rate of chemical reaction without undergoing any permanent changes themselves
- they can be reused repeatedly and are therefore effective in small amounts
- speed up reactions

intracellular - work inside cells

extracellular - secreted by cells and work outside cell

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Enzymes as catalysts lowering activation energy

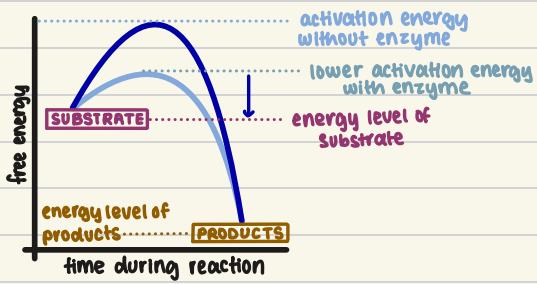
(substrates) ⟶ (products)
↳ for reaction to take place, a number of conditions must be met:

- substrates / reactants must collide with sufficient energy to alter the arrangement of atoms
- free energy of the products must be less than that of the substrates
- many reaction require an initial amount of energy to start
  - minimum amount is called activation energy

Activation energy

↳ level / energy barrier which must be overcome before the reaction can proceed

- Enzymes work by lowering the activation energy level
  ↳ allow reactions to take place at lower temperatures



Enzyme Structure

↳ globular protein, with a specific 3D-shape that is the result of their sequence of amino acids

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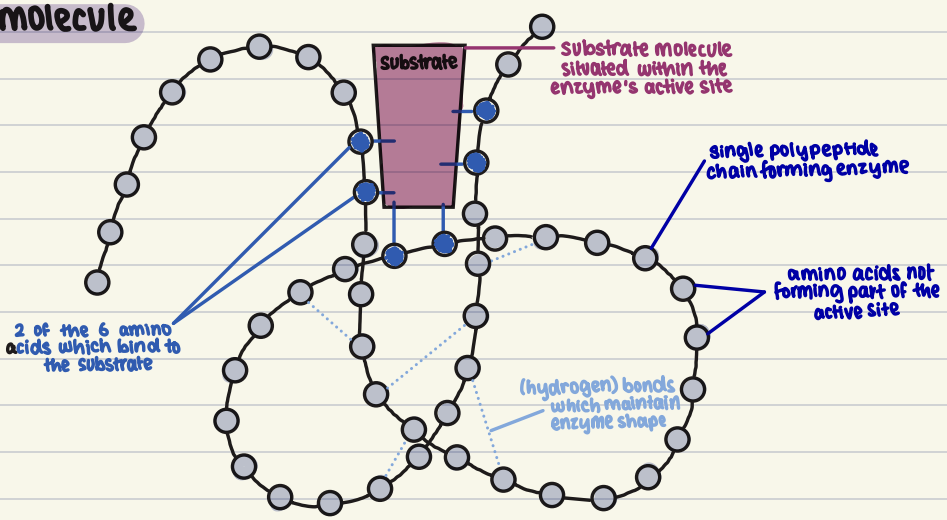
A specific region of the enzyme is functional, this is known as the active site - made up of a relatively small number of amino acids
└ forms a small depression within larger enzyme molecule

Substrate - molecule enzyme acts on
└ fits into depression (active site) to form an enzyme substrate complex
- held within active site by bonds that temporarily form between certain amino acids of the active site and groups on the substrate molecule



Induced fit model of enzyme action

└ active site forms as the enzyme and substrate interact
- proximity of the substrate (a change in the environment of the enzyme) leads to a change in the enzyme that forms the functional active site
- enzyme is flexible and can mould itself around the substrate
- enzyme has a general shape but alters in the presence of the

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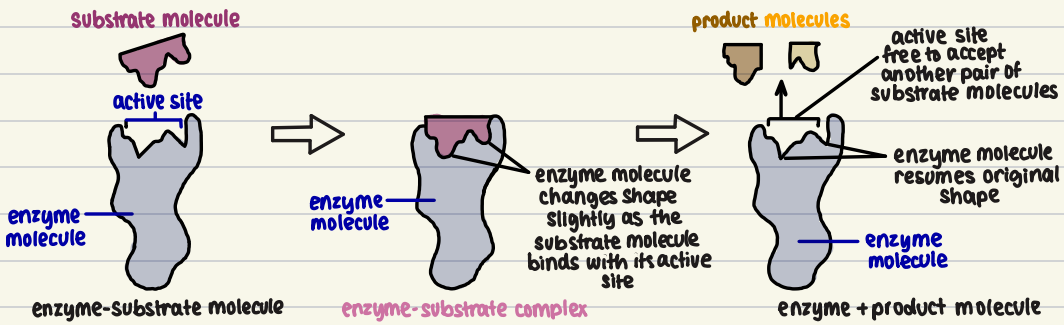
Substrate

- as it changes shape, the enzyme puts strain on the substrate molecule
  - strain distorts a particular bond / bonds in the substrate and consequently lowers activation energy needed to break bond

- any change in enzyme's environment is likely to change its shape (e.g. colliding with its substrate → shape changes - induced fit)



1.8 - Factors affecting enzyme action

For an enzyme to work, it must:

- come into physical contact with its substrate
- have an active site which fits the substrate

Measuring enzyme - catalysed reactions

- to find progress of an enzyme-catalysed reaction we usually measure its time-course, that is how long it takes for a particular event to run its course

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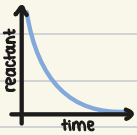
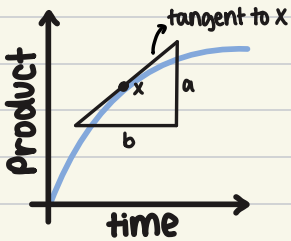
Two changes most frequently measured are:

- the formation of the products of the reaction
- the disappearance of the substrate





Measuring rate of change

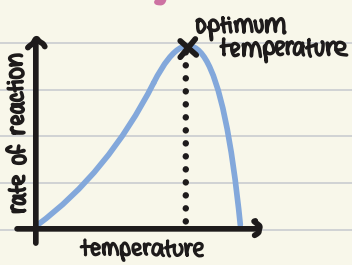


$$\text{rate of change} = \frac{a}{b}$$

Factors affecting enzyme action

- **temperature**

- rise in temperature increases the Kinetic energy of molecules, so the molecules collide successfully more often with enzymes, forming more enzyme - substrate complexes, so rate of reaction increases



- as temperature gets higher, the hydrogen and other bonds to break, causing changes to the shape of the active site, slowly preventing the substrate from binding

- after a certain temperature, enzymes denature, as the tertiary structure is permanently disrupted

- enzymes have different optimum temperatures

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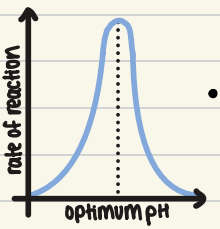
• pH

- measure of a solutions hydrogen ion concentration
- each enzyme has an optimum pH (works fastest)

$pH = -\log_{10}[H^+]$

└ $[H^+] \rightarrow 1 \times 10^{-x} = pH$ of x



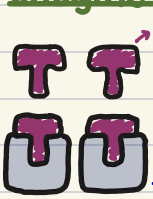
• An increase or decrease in pH reduces the rate of enzyme
└ if the change in pH is more extreme, the enzyme denatures

pH's effect on an enzyme's function:

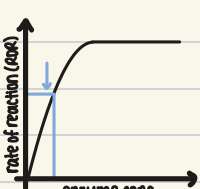
- a change in pH alters the charges on the amino acids that make up the active site of the enzyme
└ as a result substrate can no longer bind to form complex
- extreme pH changes may cause bonds maintaining enzyme's tertiary structure to break, changing shape of active site

• arrangement of active site is partly determined by the hydrogen and ionic bonds between $-NH_2$ and $-COOH$ groups of the polypeptides that make up the enzyme
- change in H^+ ions affect this bonding, causing shape change in the active site

• enzyme concentration



Low Enzyme Concentration



→ too few enzymes molecules for no. of substrate, so ROR is half of its max

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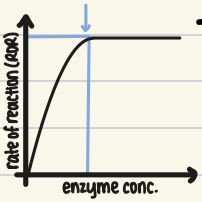
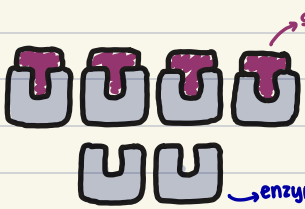
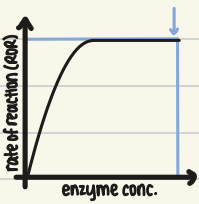
Intermediate Enzyme Concentration



→ all the substrates can occupy an active site, so ROR doubled to max.

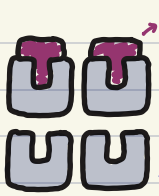


High Enzyme Concentration



→ addition of enzymes has no effect to ROR, as all substrates already have an available enzyme

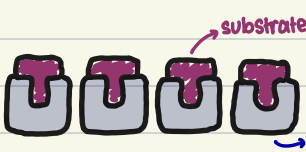
- Substrate concentration



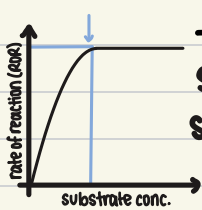
Low Substrate Concentration



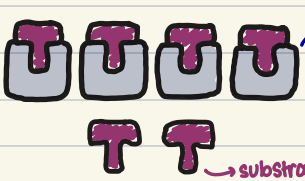
→ too few substrate molecules for no. of enzymes, so ROR is half of its max



Intermediate Substrate Concentration



→ twice as many substrate, all active sites full, ROR is at its max. (V_{max})



High Substrate Concentration



→ addition of more substrates has no effect as all achve sites are occupied, so no increase in ROR

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1.9 - Enzyme Inhibition

Enzyme inhibitors are substances that directly or indirectly interfere with the functioning of the active site and so reduce its activity:

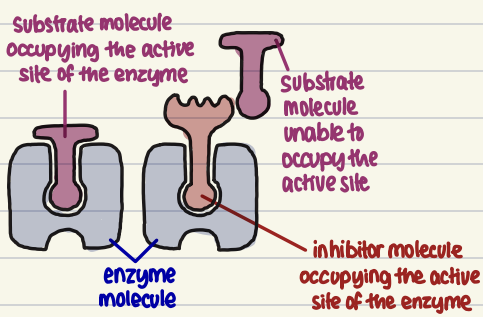
- competitive inhibitors → bind to active site of enzyme
- non-competitive inhibitors → bind to enzyme at a position other than the active site

Competitive Inhibitors

└ have a similar molecular shape to substrate molecule so they can occupy the active site of an enzyme - compete for available active sites

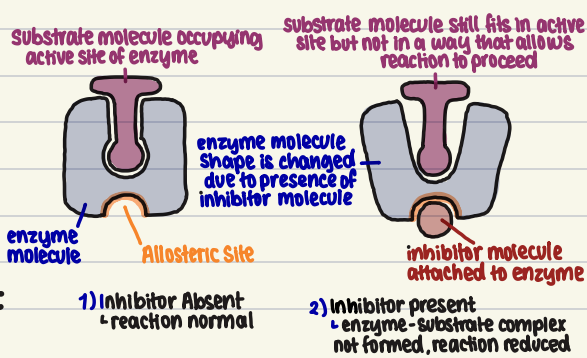
- difference in their concentration determines impact on enzyme activity
  └ [inhibitors] > [substrate] = ↑ effect on enzyme activity

- inhibitors aren't permanent bound to active site, so another molecule can bind to enzyme
- substrates all eventually bind to an active site, inhibitors just slow the process down



Non-Competitive Inhibitors

└ attach themselves to the enzyme at a binding site which is not the active site, altering shape of enzyme and its active site, so substrate can't bind to it



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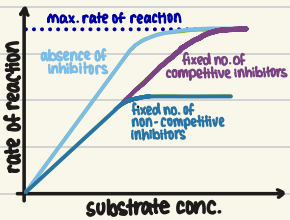
↳ therefore enzyme cannot function
- not competing, so increase in substrate concentration doesn't reduce the effect of the inhibitors



End-Product Inhibition

↳ enzyme inhibition is important in regulating metabolic pathways
- the final (end) product often acts as a regulator

• when end product too high, it binds non-competitively to an enzyme in the pathway, blocking further production of itself