

1.3 Cells: Membrane Structure & Transport

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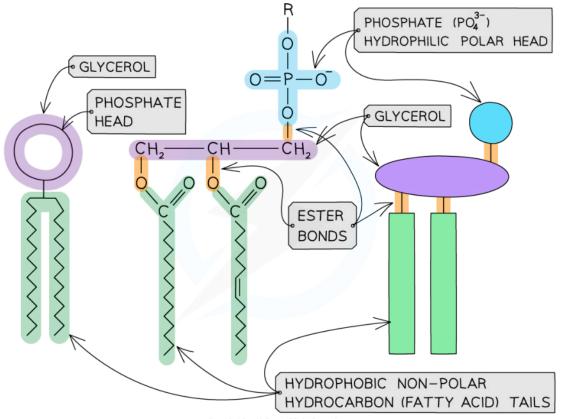


1.3.1 Phospholipid Bilayer Properties

Amphipathic Properties

Phospholipids

- Phospholipids form the basic structure of the membrane (the phospholipid bilayer)
- They are formed by a hydrophilic **phosphate head** bonding with two hydrophobic **hydrocarbon (fatty acid) tails**
- As phospholipids have a **hydrophobic** and **hydrophilic** part they are known as **amphipathic**
- The **phosphate head** of a phospholipid is **polar** (hydrophilic) and therefore **soluble** in water
- The fatty acid tail of a phospholipid is nonpolar (hydrophobic) and therefore insoluble in water



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The generalised molecular structure of a phospholipid

- Due to their amphipathic properties, phospholipids display an emergent property when placed into water
- The **hydrophilic** phosphate heads orientate towards the water and the **hydrophobic** hydrocarbon tails orientate inwards (away from the water)

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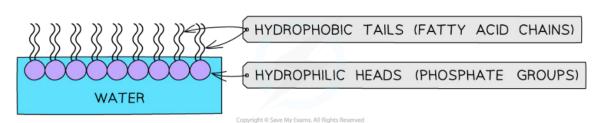


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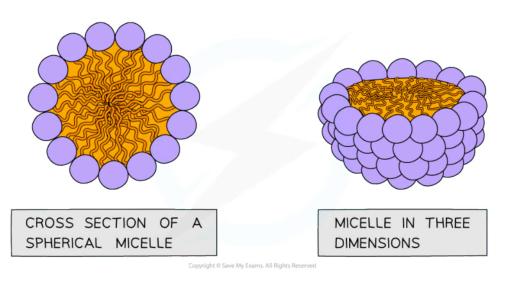
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• They form a **phospholipid monolayer**



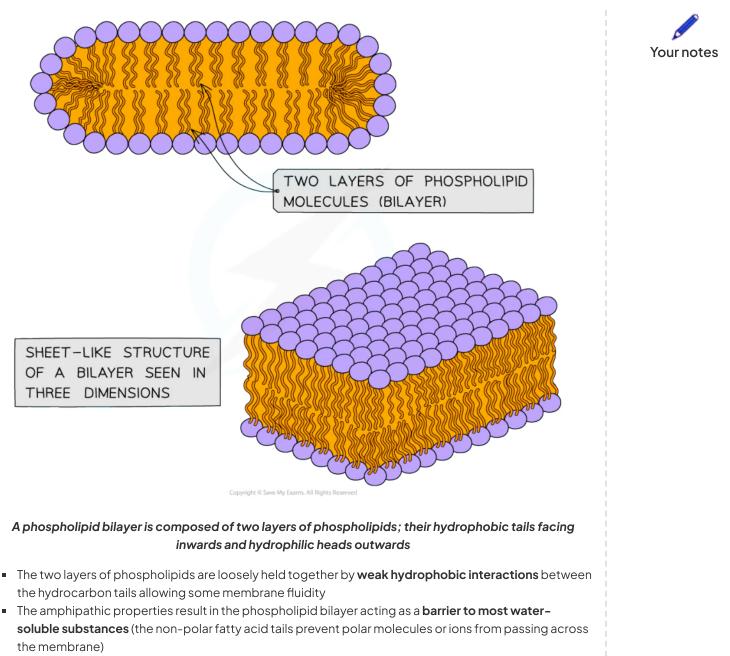
A phospholipid monolayer

- If phospholipids are mixed/shaken with water they form spheres with the hydrophilic phosphate heads facing out towards the water and the hydrophobic fatty acid tails facing inwards
 - This is called a **micelle**



A micelle

- Alternatively, when there is a sufficient concentration of phospholipids present then two-layered structures may form
- These sheets are called **phospholipid bilayers** this is the basic structure of the cell membrane



This ensures water-soluble molecules such as sugars, amino acids and proteins cannot leak out of the cell and unwanted water-soluble molecules cannot get in

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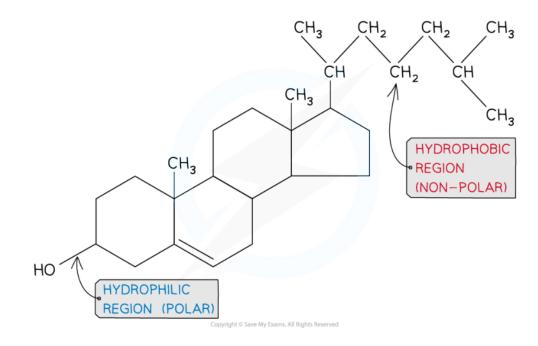
Your notes

Animal Cell Membranes: Cholesterol

 Phospholipids and cholesterol are the two main components of animal cell plasma membranes (cholesterol is absent in plant membranes)

Cholesterol

- Cholesterol is a **lipid** that belongs to the **steroid** group
- It is amphipathic, with the majority of the cholesterol molecule being hydrophobic and therefore attracted to the hydrophobic hydrocarbon tails of the phospholipid
- The **hydroxyl** group of the cholesterol molecule is **hydrophilic**. It is attracted to the **phosphate heads** of the phospholipid
- Therefore in the plasma membrane cholesterol is positioned between phospholipids



The molecular structure of cholesterol

Mammalian Membranes: Role of Cholesterol

- The **plasma membrane** is **fluid**, meaning the components are free to move
- The fluidity of the membrane needs to be controlled:
 - If it was too fluid the cell could not regulate what moved in and out
 - If it was not fluid enough then the cell would not be able to move and substances would be unable to move into or out of the cell
- Cholesterol helps with the regulation of the membrane fluidity and permeability
 - Interaction between cholesterol and phospholipid tails stabilises the plasma membrane at higher temperatures by stopping the membrane from becoming too fluid
 - Cholesterol molecules bind to the hydrophobic tails of phospholipids, stabilising them and causing phospholipids to pack more closely together
 - At **colder temperatures** cholesterol **increases the fluidity of the membrane**, stopping it crystallizing and becoming too rigid
 - This occurs because cholesterol **stops the phospholipid tails packing too closely together**
 - The impermeability of the membrane to hydrophilic ions (e.g. sodium and hydrogen) is also reduced by cholesterol
- Cholesterol increases the mechanical strength and stability of membranes (without it membranes would break down causing cells to burst)

💽 Exam Tip

It is important to remember that cholesterol affects membrane fluidity and the permeability of hydrophilic ions (e.g. sodium and hydrogen) in mammal membranes.



1.3.2 Membrane Proteins

Membrane Proteins

- The phospholipid bilayer carries out the main function of the plasma membrane to control the movement of substances into and out of the cell
- The other functions are carried out by **proteins** in the membrane
- Plasma membranes are **globular** proteins
- These proteins are grouped into two categories:
 - Integral these are partially hydrophobic and therefore are embedded in the phospholipid bilayer (either in both layers or just one)
 - **Peripheral** these are **hydrophilic** and so are temporarily attached to either the surface of integral proteins (inside or outside the cell) or connected to the plasma membrane via a hydrocarbon chain
- The protein content of membranes can vary depending on the function. Membranes of the mitochondria and chloroplasts have the highest protein content with their many electron carriers

Membrane protein functions

 Membrane proteins carry out many functions: transport, receptors, cell adhesion, cell-to-cell recognition and immobilized enzymes

Transport

- Transport proteins create hydrophilic channels to allow ions and polar molecules to travel through the membrane
- There are two types:
 - Channel (pore) proteins
 - Carrier proteins
 - Carrier proteins change shape to transport a substance across the membrane e.g. protein pumps and electron carriers
- Each transport protein is specific to a particular ion or molecule
- Transport proteins allow the cell to **control** which substances enter or leave

Receptors

- Receptors are for the binding of peptide hormones (e.g. insulin), neurotransmitters or antibodies
- The binding generates a signal that triggers a series of reactions

Immobilized enzymes

 Immobilized enzymes are integral proteins with the active site exposed on the surface of the membrane (can be inside or outside the cell)

Cell adhesion

• Cell adhesion allows **tight junctions** to be formed between cells

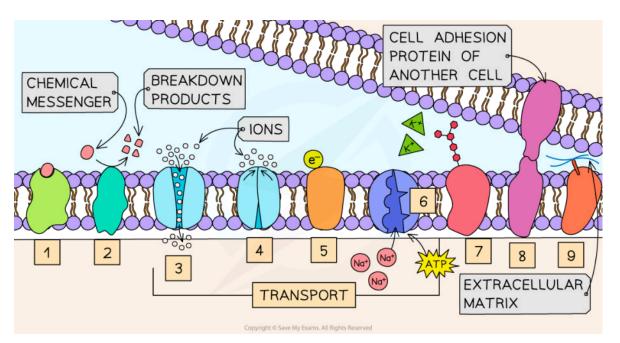
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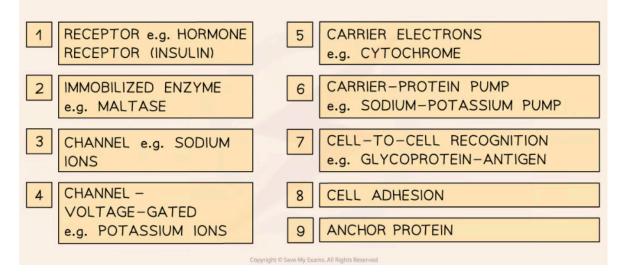


Cell-to-cell recognition

• Glycoproteins act as cell markers or **antigens**, for **cell-to-cell recognition** (eg. the ABO blood group antigens are glycolipids and glycoproteins that differ slightly in their carbohydrate chains)

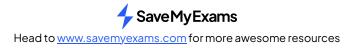






Examples of the functions of membrane proteins

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As you go through the Biology course you will learn specific examples of how membrane proteins are used so try to make the links, this will help you remember in the exams.



1.3.3 History of Fluid Mosaic Model

History of Fluid Mosaic Model

NOS: Using models as representations of the real world; there are alternative models of membrane structure

- Scientists use models to represent real world ideas, organisms, processes and systems that cannot be easily investigated. Scientists can experiment on the models enabling them to test predictions and develop explanations for observations made
- Over time as technological developments have been made the models used to represent the structure of membranes of cells and organelles have changed

1920's Gorter and Grendel

- The Gorter and Grendel model showed that the phospholipids in the membrane of cells were arranged into a bilayer
- Evidence for this model:
 - The number of phospholipids extracted from red blood cell membranes was double the area of the plasma membrane if it was arranged as a monolayer
- **Problems** with this model:
 - Their model did not explain the location of proteins or how molecules that were insoluble in lipids moved into and out of the cell

1930's Davson and Danielli

- Davson and Danielli's model of the membrane suggested that the proteins were arranged in layers above and below the phospholipid bilayer
- Evidence for this model:
 - Membranes were effective at controlling the movement of substances in and out of cells
 - Electron micrographs showed the membrane had two dark lines with a lighter band between. In electron micrographs, proteins appear darker than phospholipids
- **Problems** with this model:
 - Freeze-etched electron micrographs of the centre of the membrane showed globular structures scattered throughout
 - Improvements in technology used to analyse the proteins in the membranes showed that proteins were globular, varied in size and had parts that were hydrophobic
 - These problems suggested it was **unlikely** that the proteins would **form continuous layers**

1970's Singer and Nicolson

• Singer and Nicolson proposed the fluid mosaic model which stated that membranes were fluid and that the globular proteins were both peripheral and integral (with some crossing both membranes) and

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dispersed throughout the membrane

• **Evidence** for this model:

- Analysis of freeze-etched electron micrographs showed proteins extending into the centre of membranes
- Biochemical analysis of the plasma membrane components
- The use of coloured fluorescent markers of antibodies. Antibodies were tagged with red and green fluorescent markers. These antibodies were bound to membrane proteins on different cells. Forty minutes after these cells were fused together the markers were seen to have mixed throughout the fused cells membrane showing that membrane proteins are free to move within the layer

A 1920'S GROTER-GRENDEL MODEL 1920'S OAVSON-DANIELLI MODEL GLOBULAR PROTEIN PHOSPHOLIPID BILAYER CARACTERINER

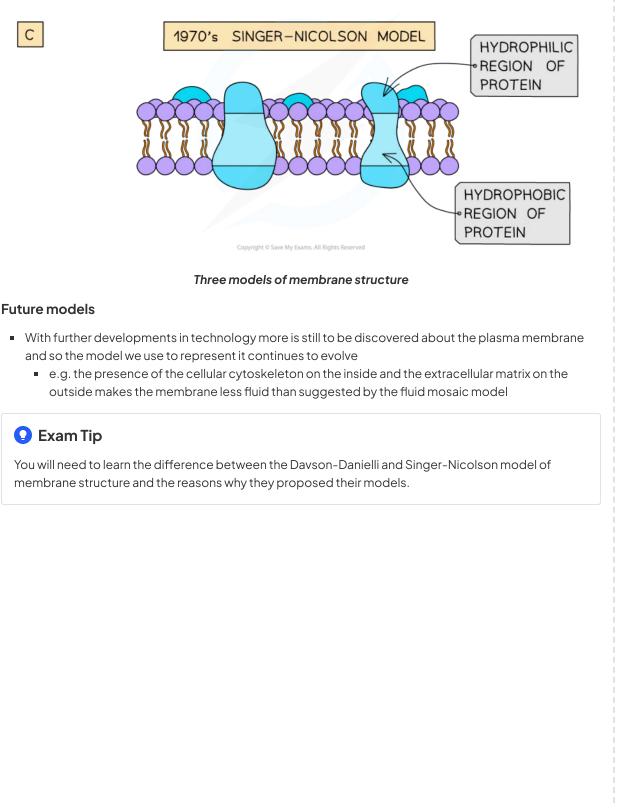


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Your notes

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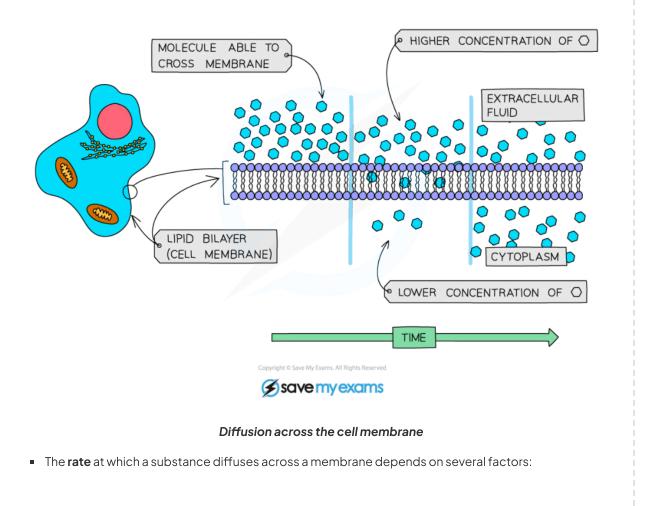


1.3.4 Membrane Transport

Passive Transport

Simple diffusion

- Simple diffusion is a type of **transportation** that involves particles passing between phospholipids in **the plasma membrane**
- It can be defined as:
 - The net movement, as a result of the random motion of its molecules or ions, of a substance from a region of its higher concentration to a region of its lower concentration
- The molecules or ions move **down a concentration gradient**
- The random movement is caused by the natural **kinetic energy** of the molecules or ions
- As a result of diffusion, molecules or ions tend to reach an equilibrium (given sufficient time), where they are evenly spread within a given volume of space





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- 'Steepness' of the concentration gradient the greater the difference the higher the rate of diffusion
- **Temperature** the higher the temperature the higher the rate of diffusion
- Surface area the greater the surface area the higher the rate of diffusion
- Properties of the molecules or ions
 - Large molecules diffuse more slowly as they require more energy to move
 - **Uncharged** molecules (e.g. oxygen) diffuse faster as they move directly across the phospholipid bilayer
 - Non-polar molecules diffuse more quickly as they are soluble in the non-polar phospholipid bilayer
 - Although polar molecules cannot easily pass through the hydrophobic part of the membrane, smaller polar molecules (e.g. urea) can diffuse at low rates

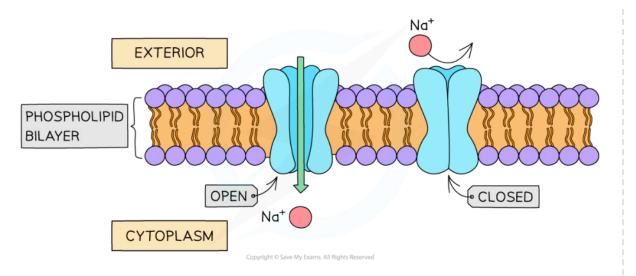
Facilitated diffusion

- Certain substances cannot diffuse through the phospholipid bilayer of cell membranes. These include:
 - Large polar molecules such as glucose and amino acids
 - lons such as sodium ions (Na⁺) and chloride ions (Cl⁻)
- These substances can only cross the phospholipid bilayer with the help of certain proteins
- This form of diffusion is known as **facilitated diffusion**
- There are two types of proteins that enable facilitated diffusion:
 - Channel proteins
 - Carrier proteins (these can also be used during active transport)
- They are **highly specific** (they only allow one type of molecule or ion to pass through)

Channel proteins

- Channel proteins are water-filled **pores**
- They allow **charged substances** (eg. ions) to diffuse through the cell membrane
- The diffusion of these ions does not occur freely, most channel proteins are '**gated**', meaning that part of the channel protein on the inside surface of the membrane can move in order to close or open the pore
- This allows the channel protein to **control** the exchange of ions

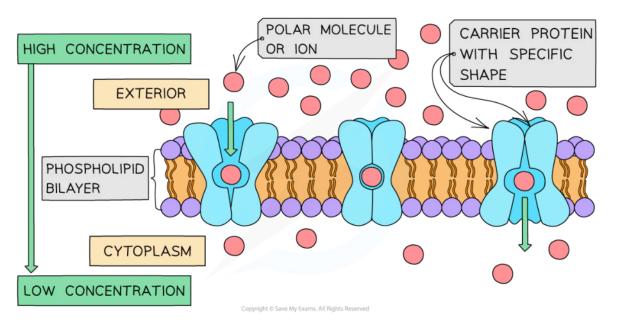
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A channel protein (open and closed)

Carrier proteins

- Unlike channel proteins which have a fixed shape, carrier proteins can switch between two shapes
- Initially, the binding site of the carrier protein is open to one side of the membrane
- When the carrier protein switches shape it opens to the other side of the membrane
- The direction of movement of molecules diffusing across the membrane depends on their relative concentration on each side of the membrane
- During facilitated diffusion, the net diffusion of molecules or ions into or out of a cell will occur down a concentration gradient (from an area containing many of that specific molecule to an area containing less of that molecule)

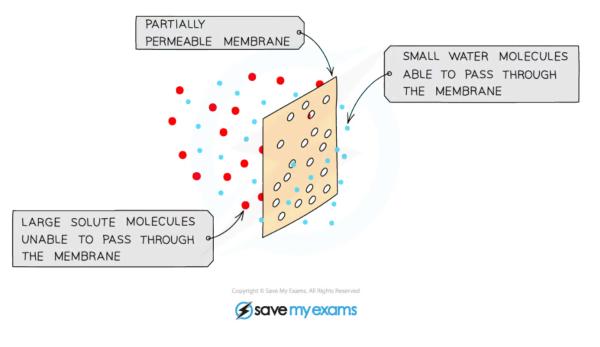


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A carrier protein changing shape during facilitated diffusion

Osmosis

- All cells are surrounded by a cell membrane which is partially permeable
- Water can move in and out of cells by **osmosis**
- Osmosis is the diffusion of water molecules from a dilute solution to a more concentrated solution across a partially permeable membrane
 - In doing this, water is moving down its **concentration gradient**
 - A dilute solution has a high concentration of water molecules and a concentrated solution has a low concentration of water molecules
- The cell membrane is partially permeable which means it **allows small molecules (like water) through** but not larger molecules (like solute molecules)



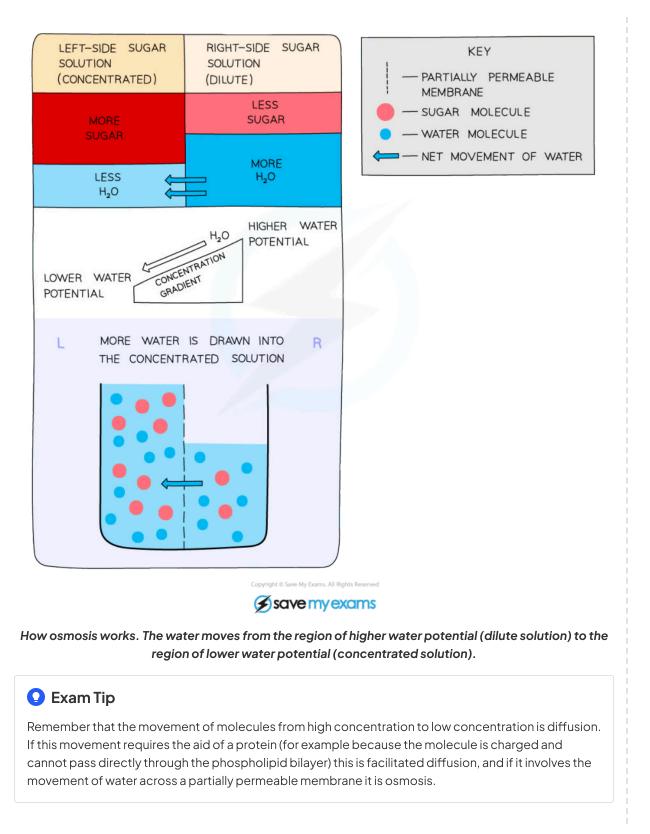
Osmosis and the partially permeable membrane

- The term **osmolarity** can be used to describe the solute concentration of a solution; a solution with high osmolarity has a high solute concentration and a solution with low osmolarity has a low solute concentration.
 - Water will move from a solution of low osmolarity to a solution of high osmolarity across a partially permeable membrane
- Osmosis can also be described as the net movement of water molecules from a region of higher water potential to a region of lower water potential, through a partially permeable membrane
 - Water potential describes the tendency of water to move out of a solution; this term is used to avoid confusion between water concentration and solute concentration of a solution

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Facilitated Diffusion: Example

Axons

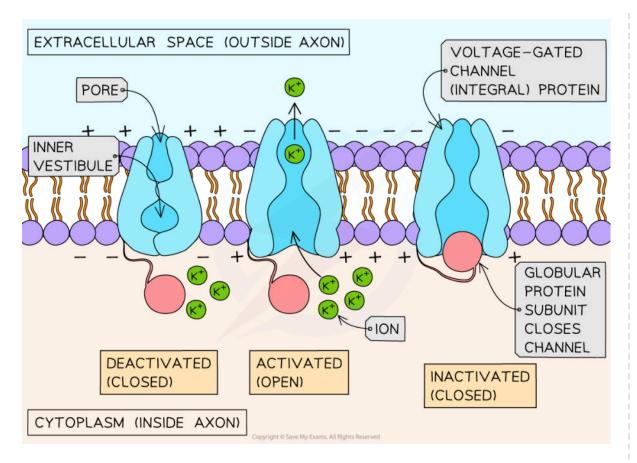
- The **axon** is part of a **nerve cell** (neuron)
- It is a long, narrow tube (it can be one metre in length with a diameter of one micron) containing cytoplasm surrounded by a membrane
- Axons transmit **nerve impulses**
- These nerve impulses occur because sodium and potassium ions are being moved across the axon membrane creating a voltage difference

Facilitated diffusion of potassium

- Voltage-gated channel proteins (enabling facilitated diffusion) allow the movement of the potassium ions
- Potassium ions **require channel proteins** to diffuse across the axon as they are **charged** and bond with water when they dissolve in the cytoplasm
- Potassium channels are **integral proteins** and **allow only potassium ions** through because:
 - Other positively charged ions are too large to move through the channel
 - Other ions are too small to form bonds with the amino acids located in the channel so they remain attached to water molecules
- The potassium channels in axons are **voltaged gated**
- The channel proteins will **open** (to allow potassium ions to diffuse out) when the charge inside the axon is relatively more **positive** than outside
- However, the channel proteins rapidly close due to the presence of an extra globular protein subunit. This subunit fits inside the open channel within milliseconds of the channel opening blocking any further diffusion out of the potassium ions
- The subunit remains in place until the potassium channel closes



Your notes



Voltage-gated potassium channels facilitate the diffusion of potassium ions

Prevention of Osmosis in Medical Procedures

- Animal cells can lose and gain water as a result of osmosis
- As animal cells **do not have a supporting cell wall** (unlike plant cells), the results of this loss or gain of water on the cell are **severe**
- This is why a constant water potential **must** be maintained inside the bodies of animals

Animal cells losing water

- If an animal cell is placed in a solution with a lower water potential than the cell, water will leave the cell through its partially permeable cell surface membrane by osmosis and the cell will shrink and shrivel up
 - This is **crenation** (the cell has become **crenated**), which is usually **fatal** for the cell
- Crenation occurs when the cell is in a hypertonic environment (the solution outside of the cell has a higher solute concentration than the inside of the cell)

Animal cells gaining water

- If an animal cell is placed in pure water or a dilute solution, water will enter the cell through its partially
 permeable cell surface membrane by osmosis, as the pure water or dilute solution has a higher water
 potential
- The cell will continue to gain water by osmosis until the cell membrane is stretched too far and the cell bursts (cytolysis), as it has no cell wall to withstand the increased pressure created
 - This is **fatal** for the cell
- Lysis occurs when the cell is in a hypotonic environment (the solution outside of the cell has a lower solute concentration than the inside of the cell)

Animal cells in isotonic environments

- If an animal cell is in an isotonic environment (the solution outside of the cell has the same solute concentration as the inside of the cell)
- The movement of water molecules into and out of the cell occurs at the same rate (no net movement of water) and there is no change to the cells



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| RED BLOOD CELLS HAVE HIGHER WATER POTENTIAL THAN SOLUTION NET MOVEMENT OF WATER OUT SHRIVELLED CELLS | WATER POTENTIAL EQUAL BETWEEN RED BLOOD CELL AND SOLUTION NO NET MOVEMENT OF WATER NORMAL CELLS | RED BLOOD CELLS HAVE LOWER WATER POTENTIAL THAN SOLUTION NET MOVEMENT OF WATER IN CELLS SWELL, MAY LYSE (BURST) |
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| Osmolarity of solutions used in medical procedures | | |
| Tissues and organs that are to be used in medical procedures must be kept in solution to prevent damage to the cells The osmolarity of the solution is key The osmolarity of a solution measures the number of solute particles (that can form bonds with water per 1 L of solvent Osmolarity is expressed as osmoles or milliosmoles per litre of solution (Osm/L or mOsm/L) Human tissue is normally 306 mOsm/L A solution with the same osmolarity = isotonic A solution with a higher osmolarity = hypertonic A solution with a lower osmolarity = hypotonic | | |
| Isotonic sodium chloride solutions (normal saline) are generally used as they can be: | | |

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- Frozen to create a slush used to pack donor organs for transportation
- Injected into a patient's blood system
- Used to sterilise wounds
- Used as eye drops



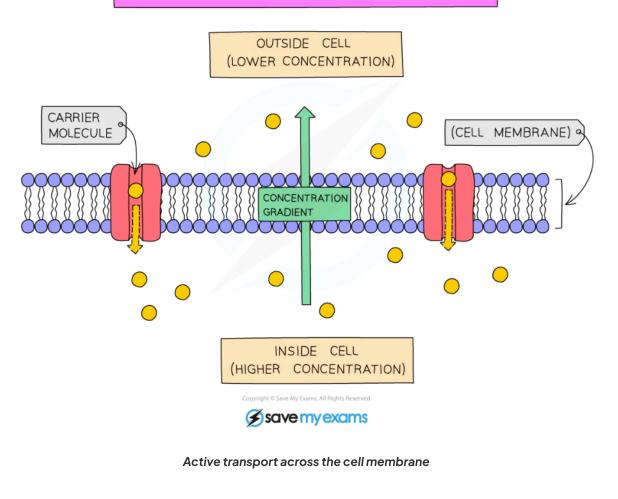
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1.3.5 Active Transport & Bulk Transport

Active Transport

- Active transport is the **movement of molecules and ions through a cell membrane from a region of lower concentration to a region of higher concentration using energy from respiration**
- Active transport requires carrier proteins (each carrier protein being specific for a particular type of molecule or ion)
- Although facilitated diffusion also uses carrier proteins, active transport is different as it requires energy
- The energy is required to make the carrier protein **change shape**, allowing it to transfer the molecules or ions across the cell membrane
- The energy required is provided by **ATP** (adenosine triphosphate) produced during **respiration**. The ATP is **hydrolysed** to release energy

ACTIVE TRANSPORT ACROSS THE CELL MEMBRANE





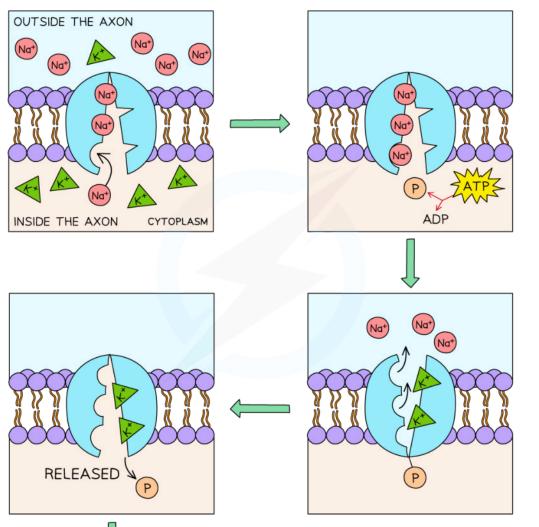
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Active Transport: Example

Sodium-potassium pumps in axons

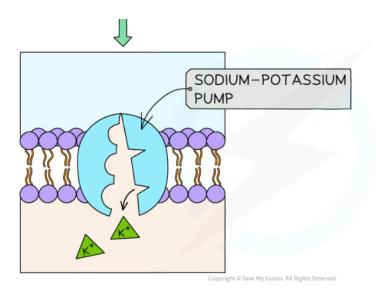
- Sodium-potassium carrier pump proteins are integral proteins that enable an electrochemical gradient (resting membrane potential) to be maintained between the inside and outside of the axon
- Nerve impulses that travel along axons are dependent on sodium and potassium ions being moved across the axon membrane to create this gradient
- The sodium-potassium pumps move three sodium ions out of the axon and two potassium ions into the axon using one ATP molecule per cycle
- The pumps are always moving the ions against their concentration gradient via active transport
- The cycle continues until the resting membrane potential is reached
- The steps to this cycle are:
 - Three sodium ions from the inside of the axon bind to the pump
 - **ATP attaches** to the **pump** and **transfers** a **phosphate** to the pump (phosphorylation) causing it to change shape, resulting in the pump opening to the outside of the axon
 - The three **sodium ions** are **released** out of the axon
 - Two potassium ions from outside the axon enter and bind to their sites
 - The attached phosphate is released altering the shape of the pump again
 - The change in shape causes the **potassium ions** to be **released inside** the axon







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Active transport of sodium and potassium ions in axons using sodium-potassium pump carrier proteins

Bulk Transport

Bulk transport

- The processes of diffusion, osmosis and active transport are responsible for the transport of individual molecules or ions across cell membranes
- However, the bulk transport of larger quantities of materials into or out of cells is also possible
- Examples of these larger quantities of materials that might need to cross the membrane include:
 - Large molecules such as proteins or polysaccharides
 - Parts of cells
 - Whole cells eg. bacteria
- Bulk transport **into** cells = **endocytosis**
- Bulk transport **out** of cells = **exocytosis**
- These two processes **require energy** and are therefore forms of active transport
- They also require the formation of **vesicles** which is dependent on the fluidity of membranes

Fluidity of membranes

- The phospholipid bilayer is loosely held together by weak hydrophobic interactions between the hydrocarbon tails
- These weak interactions allow for some degree of membrane fluidity
- The membrane fluidity allows larger substances to move in and out of the cell in vesicles formed when proteins and ATP are used to pinch off small regions of the plasma membrane

Vesicles

- Vesicles are small spherical sacs of plasma membrane containing water and solutes
- They will often contain larger molecules that cannot pass across the plasma membrane (e.g. proteins)
- The formation of vesicles is an **active** process requiring **ATP** and **proteins** and involves a small region of the plasma membrane being pinched off
- Vesicles are normally present in eukaryotic cells
- Vesicles move materials within cells. These materials may be required by other organelles or may be required outside the cell
- An example of materials moved by vesicles out of the cell is digestive enzymes
 - In exocrine pancreatic gland cells, proteins synthesised by ribosomes on the rough endoplasmic reticulum are packaged into vesicles that move them to Golgi apparatus. Here the vesicles fuse with the membrane of the Golgi apparatus and the proteins are modified. New vesicles then pinch off and move to the plasma membrane to secrete the digestive enzymes into the pancreatic ducts
- Vesicles can also be used to move membrane proteins and phospholipids to the plasma membrane so cells can grow or to organelles in the cytoplasm so they can increase in size

Endocytosis

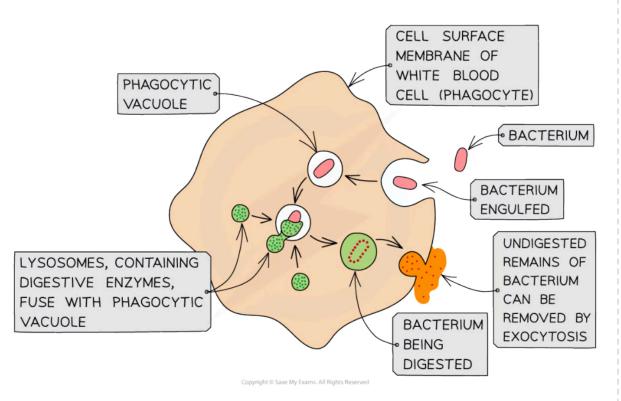
- Endocytosis is the process by which the plasma membrane engulfs material, forming a small sac (or 'endocytic vacuole') around it
- There are two forms of endocytosis:

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- Phagocytosis:
 - This is the bulk intake of solid material by a cell
 - Cells that specialise in this process are called phagocytes
 - The vacuoles formed are called phagocytic vacuoles
 - An example is the engulfing of bacteria by phagocytic white blood cells
- Pinocytosis:
 - This is the bulk intake of liquids
 - If the vacuole (or vesicle) that is formed is extremely small then the process is called micropinocytosis



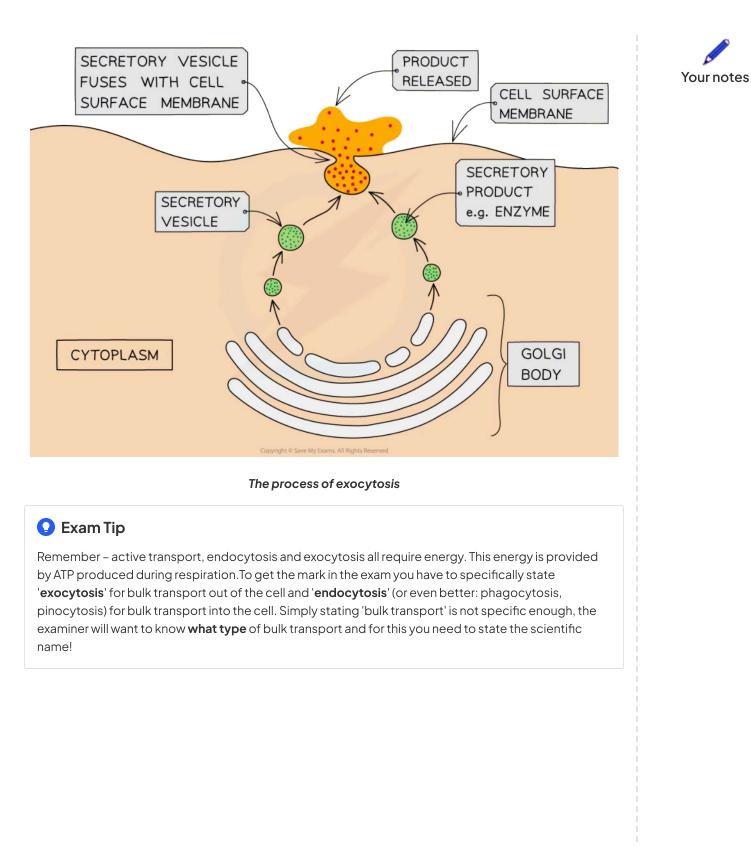


Exocytosis

- Exocytosis is the process by which materials are removed from, or transported out of, cells (the reverse of endocytosis)
- The substances to be released (such as enzymes, hormones or cell wall building materials) are packaged into secretory vesicles formed from the Golgi body
- These vesicles then travel to the cell surface membrane
- Here they fuse with the cell membrane and release their contents outside of the cell
- An example is the secretion of digestive enzymes from pancreatic cells

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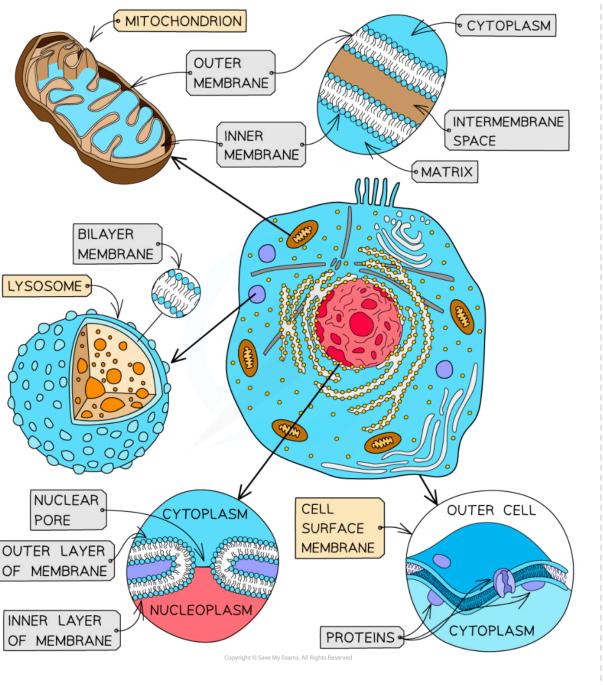
1.3.6 Skills: Membrane Structure & Transport

Drawing the Fluid Mosaic Model

Membranes

- Membranes are vital structures found in all cells
- The **cell surface membrane** creates an enclosed space separating the internal cell environment from the external environment
- Intracellular membranes (internal membranes) form compartments within the cell, such as organelles (including the nucleus, mitochondria and RER) and vacuoles
- Membranes not only separate different areas but also control the exchange of materials passing through them; they are partially permeable
- Membranes form partially permeable barriers between the cell and its environment, between cytoplasm and organelles and also within organelles
- Substances can cross membranes by diffusion, facilitated diffusion, osmosis and active transport
- Membranes play a role in **cell signaling** by acting as an **interface** for **communication between cells**





Your notes

Membranes formed from phospholipid bilayers help to compartmentalise different regions within the cell, as well as forming the cell surface membrane

Fluid Mosaic Model

• The **fluid mosaic model** of membranes was first outlined in 1972 by **Singer and Nicolson** and it explains how biological molecules are arranged to form cell membranes

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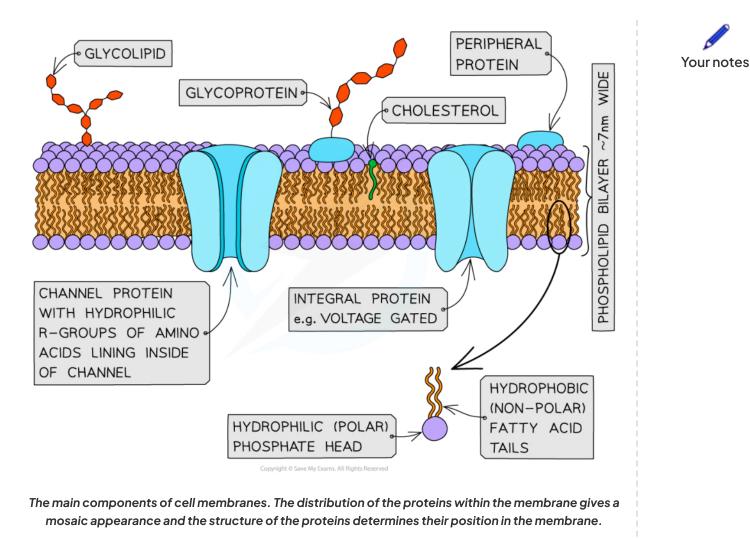
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- The fluid mosaic model also helps to explain:
 - Passive and active movement between cells and their surroundings
 - Cell-to-cell interactions
 - Cell signalling
- The fluid mosaic model describes cell membranes as '**fluid**' because:
 - The **phospholipids** and **proteins** can **move around** via diffusion
 - The phospholipids mainly move sideways, within their own layers
 - The many different types of proteins interspersed throughout the bilayer move about within it (a bit like icebergs in the sea) although **some may be fixed** in position
- The fluid mosaic model describes cell membranes as **'mosaics'** because:
 - The **scattered pattern** produced by the **proteins** within the phospholipid bilayer looks somewhat like a mosaic when viewed from above
- The **fluid mosaic model** of membranes includes four main components:
 - Phospholipids
 - Cholesterol
 - Glycoproteins and glycolipids
 - Transport proteins



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When drawing the fluid mosaic model remember to include (and label) the **phospholipid bilaye**r (making it clear which part is the phosphate head and which parts are the hydrocarbon tails), the **thickness of the membrane (7 – 10 nm)**, **integral proteins** (show then embedded in the phospholipid bilayer and include a couple of different types e.g. channel/carrier), **peripheral proteins (do not** extend the protein into the hydrophobic region), **glycoprotein** (with a carbohydrate attached) and finally **cholesterol** (ensure the orientation is correct, OH group next to the phosphate heads and the rest positioned next to the tails).

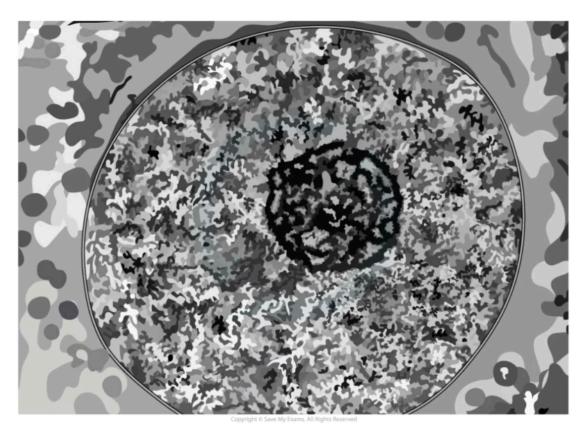
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Analysis of Evidence: Davson-Danielli Model

- Analysis of evidence from **electron microscopy** led to the proposal of the Davson-Danielli model
- Other methods were then used to further investigate the model and suggested evidence against the model
 - Freeze-etchings
 - Fluorescent markers of membrane proteins

Transmission electron micrograph (TEM) of the plasma membrane

- When analysing transmission electron micrographs comment on:
 - How the membrane has two darker layers surrounding a lighter line
 - **Proteins** were known to appear **darker** in electron micrographs
- These were the observations that **supported** Davson-Danielli's model



TEM of a plasma membrane suggests evidence for Davson-Danielli's model

Freeze-etched electron micrographs

• When asked to analyse freeze-etched electron micrographs note that the very **small bumps** seen on the membranes are the **integral proteins**

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- This provided evidence **against** Davson-Danielli's model as it showed proteins extending into the **centre** of the membrane
- Be careful if the image is of a nuclear membrane as the larger circles represent the nuclear pores

OUTER NUCLEAR MEMBRANE INNER NUCLEAR MEMBRANE NUCLEAR PORES • NUCLEUS

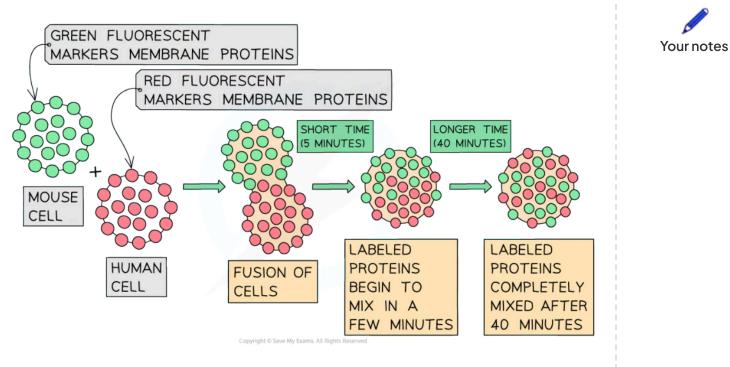
Freeze-etching of a nucleus suggests evidence against Davson-Danielli's model

Fluorescent markers on membrane proteins

- When analysing data on the use of red and green fluorescent markers attached to membrane proteins, the key evidence to note, is that **as time progresses** after the fusion of the different cells with the different markers has occurred, **more mixing** of the markers is observed
- This evidence **did not support Davson-Danielli's model** that the proteins were a uniform layer above and below the phospholipids
- It supported the 'fluid' part of Singer & Nicolson's fluid mosaic model as it suggested that membrane proteins can move

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Fluorescent markers on membrane proteins suggest evidence against Davson-Danielli's model

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Falsification of Davson-Danielli Model

NOS: Falsification of theories with one theory being superseded by another; evidence falsified the Davson-Danielli model

- For about 30 years the technology available to scientists supported the Davson-Danielli model of membrane structure
- From the 1950's an advancement in technology led to the accumulation of evidence which resulted in the Davson-Danielli model being superseded by the Singer-Nicolson 'fluid mosaic model'
- Analysis of freeze-etched electron micrographs showed proteins extending into the centre of membranes
- Biochemical analysis of membranes suggested that it was unlikely proteins formed continuous layers because it showed:
 - Proteins were globular
 - Varied in sizes
 - They had parts that were **hydrophobic**
- The use of **coloured fluorescent markers** of antibodies showed that within forty minutes of fusing cells with different coloured fluorescent markers the markers had mixed
 - This suggested that membrane proteins were **free to move** within the layer

💽 Exam Tip

It is important to be able to provide the reasons why the evidence collected falsified the Davson-Danielli model.



Your notes

1.3.7 Skills: Estimation of Osmolarity

Practical 2: Estimation of Osmolarity

NOS: Experimental design; accurate quantitative measurements in osmosis experiments are essential

- Planning is an essential part of experimental biology, it will help ensure that valid conclusions can be made
- Preliminary (meaning "to come before") research must be completed to ensure the experiment design considers:
 - The **results** that will be collected
 - Quantitative data allows more valid conclusions to be made
 - Qualitative data (descriptive) can be useful to support the conclusions
 - How measurements will be made so they are as precise and as accurate as possible
 - The choice of apparatus and techniques should be based on the science surrounding the issue being investigated
 - How many **repeats** will be undertaken to ensure the data collected is reliable
 - The variables that will be tested and need to be controlled
- Once the preliminary research has been completed then **preliminary studies** can be conducted to further aid the experimental design
- These studies are very important for:
 - Identifying additional variables that affect the experiment
 - Finding the best way to control these variables
 - Deciding on the quantities and volumes of substances that are needed so that you do not run out of reactants/reagents
- Any experiment conducted without preliminary research or studies is likely to be invalid as the other variables that affect the results in the experiment will not have been identified and controlled

Practical 2: Estimation of osmolarity in tissues by bathing samples in hypotonic and hypertonic solutions

- The osmolarity of a solution measures the number of solute particles (that can form bonds with water) per 1 L of solvent
- Osmolarity is expressed as [popover id="XqIR9B3GzVySI6JG" label = "osmoles"] or milliosmoles per litre of solution (Osm/L or mOsm/L)
- A hypotonic solution has a lower osmolarity than the tissue being bathed in it (so the tissue will increase in mass or length) whereas a hypertonic solution has a higher osmolarity (so the tissue will decrease in mass or length)
- An **isotonic solution** will have the **same osmolarity** as the tissue (so the mass or length will remain unchanged)

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Your notes

- It is possible to investigate the effects of immersing plant tissue in solutions of different osmotic concentrations (osmolarity) and to use the results to estimate the osmolarity of the plant tissue itself
- The most common osmosis practical of this kind involves cutting **cylinders of potato** and placing them into solutions with a **range of different osmotic concentrations**
 - Usually sucrose solutions of increasing concentration at least 5 different concentrations are usually required

Apparatus

- Potato x 2 (same variety)
- Cork borer (e.g. 5mm)
- White tile
- Scalpel
- 10cm ruler or vernier calipers
- Weighing balance (2dp)
- 10 cm³ sucrose solution (0 mol/dm³, 0.25 mol/dm³, 0.5 mol/dm³, 0.75 mol/dm³, 1.00 mol/dm³)
- 5 test tubes (in test tube rack)
- 10 cm³ measuring cylinder
- Paper towels

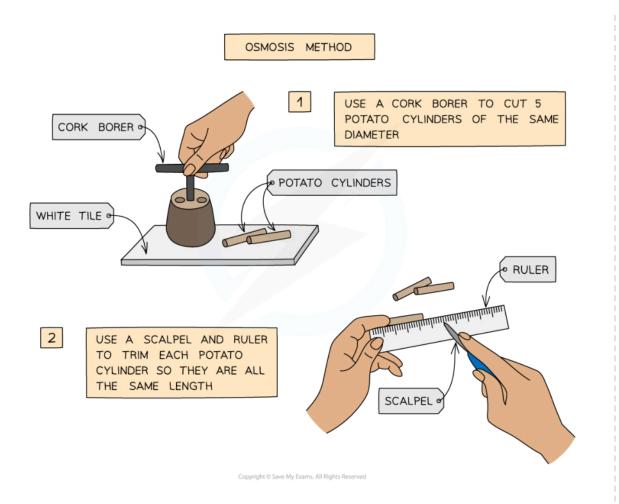
Method

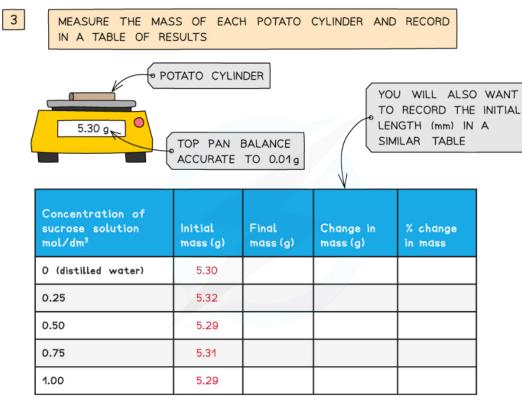
- The required number of potato cylinders are cut
 - At least 5 for each of the solutions you are testing to ensure you have sufficient repeats
- They are all cut to the same length and, once blotted dry to remove any excess moisture, their initial mass is measured and recorded before placing into the solutions
- The potato cylinders are left in the solutions for a set amount of time (eg. 30 minutes), usually in a water bath (set at around 30°)
 - The solutions are prepared by serial dilutions of a specific solute concentration determined during the preliminary research/trials)
- The cylinders are then removed and dried
 - This is done to **remove excess liquid**
- The final length and mass of each potato cylinder is then measured and recorded

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Your notes

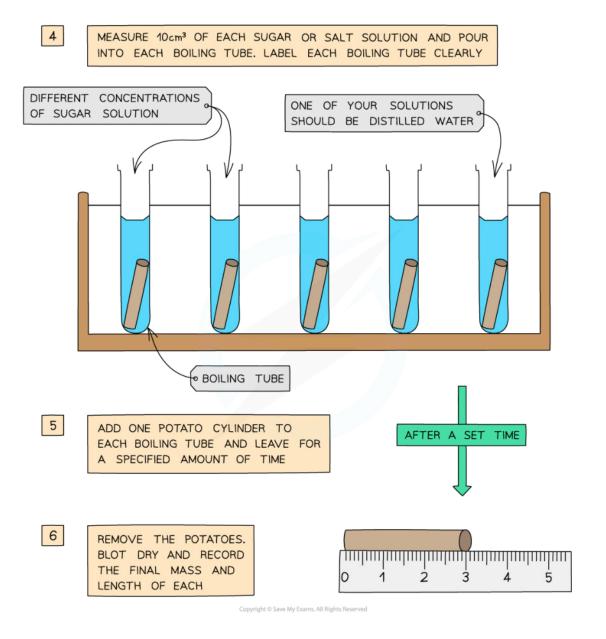




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Your notes

Your notes



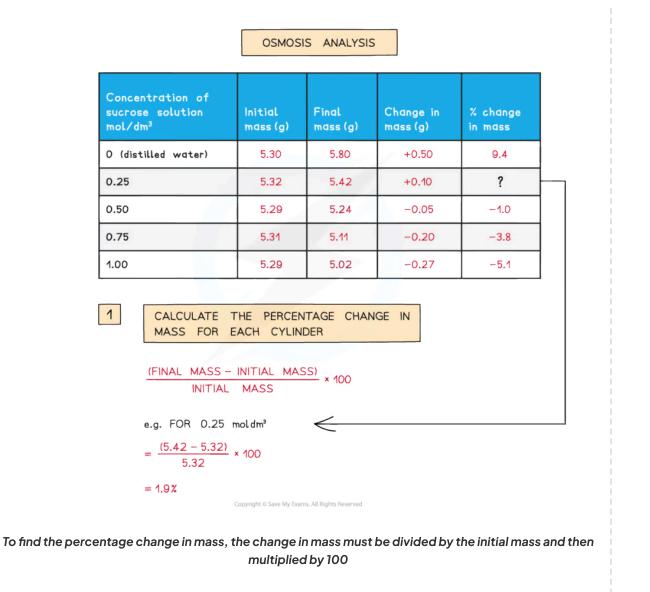
You will need to use apparatus appropriately to measure out the volumes of your solutions and record your measurements

Analysis

• The percentage change in mass for each potato cylinder is calculated and then plotted

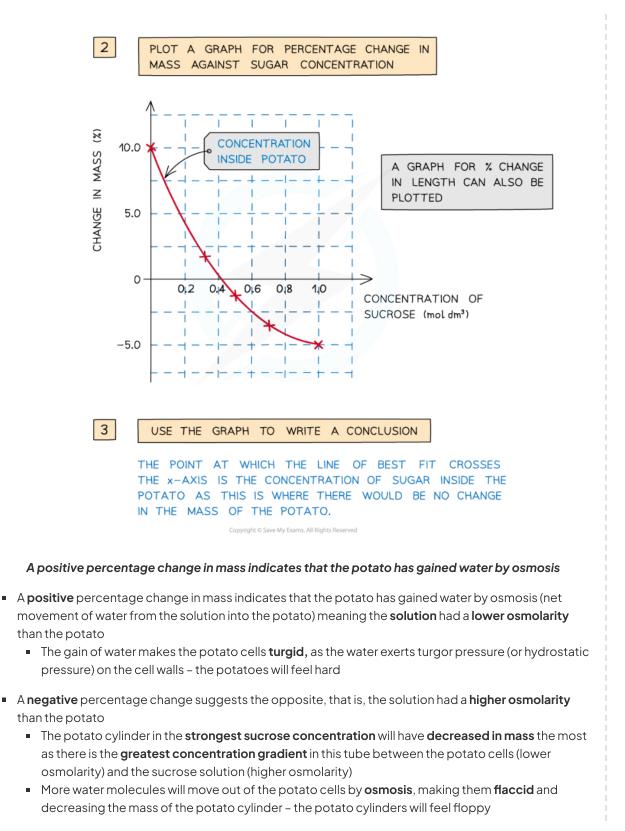
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Your notes



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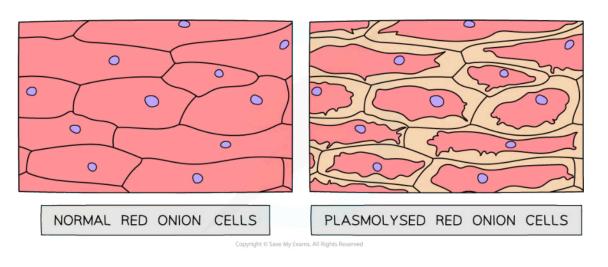
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- If looked at underneath the microscope, cells from this potato cylinder might be **plasmolysed**, meaning the cell membrane has pulled away from the cell wall
- If there is a potato cylinder that has neither increased nor decreased in mass, it means there was no overall net movement of water into or out of the potato cells
- The solution that this particular potato cylinder was in had the same osmolarity as the solution found in the cytoplasm of the potato cells, so there was no concentration gradient and therefore no net movement of water into or out of the potato cells
- The concentration of sucrose inside the potato cylinders can be found if a graph is drawn showing how the percentage change in mass changes with the concentration of sucrose solution
- The point at which the line of best fit **crosses the x-axis** is the concentration of sucrose inside the potato cylinders

Investigating osmolarity using onion cells

- Evidence of osmosis occurring in plant cells can be shown when the cells undergo **plasmolysis**:
 - If a plant cell is placed in a solution with a higher osmolarity than the cell (such as a concentrated sucrose solution), water will leave the cell through its partially permeable cell surface membrane by osmosis
 - As water leaves the **vacuole** of the plant cell, the volume of the cell **decreases**
 - The protoplast (living part of the cell inside the cell wall) gradually shrinks and no longer exerts pressure on the cell wall
 - As the protoplast continues to shrink, it begins to pull away from the cell wall
 - This process is known as **plasmolysis** the plant cell is **plasmolysed**
- This process can be observed using epidermal strips (sections of the very thin outer layer of tissue in plants)
 - Plants with coloured sap (such as red onion bulbs, rhubarb petioles and red cabbage) make observations easier
- The epidermal strips are placed in a range of molarities of sucrose solution or sodium chloride solutions, of gradually decreasing water potential
- The strips are then viewed under a light microscope and the **total number** or **percentage** of **onion cells** that have undergone **plasmolysis** can be counted
 - Plasmolysis may take several minutes to occur





Light micrographs of normal red onion cells alongside those that have plasmolysed (artistic impression). The cells on the left are epidermal cells that have been immersed in distilled water, whilst the cells on the right are epidermal cells that have been immersed in 1.0 mol dm³ sucrose solution.

💽 Exam Tip

Questions involving experiments investigating osmolarity and osmosis are common and you should be able to use your knowledge of osmosis to explain the results obtained. Don't worry if it is an experiment you haven't done – simply figure out where the higher concentration of water molecules is – this is the solution with the lower osmolarity – and explain which way the molecules move due to the differences in osmolarity.



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