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## **1.3 Cells: Membrane Structure & Transport**



# **IB Biology - Revision Notes**

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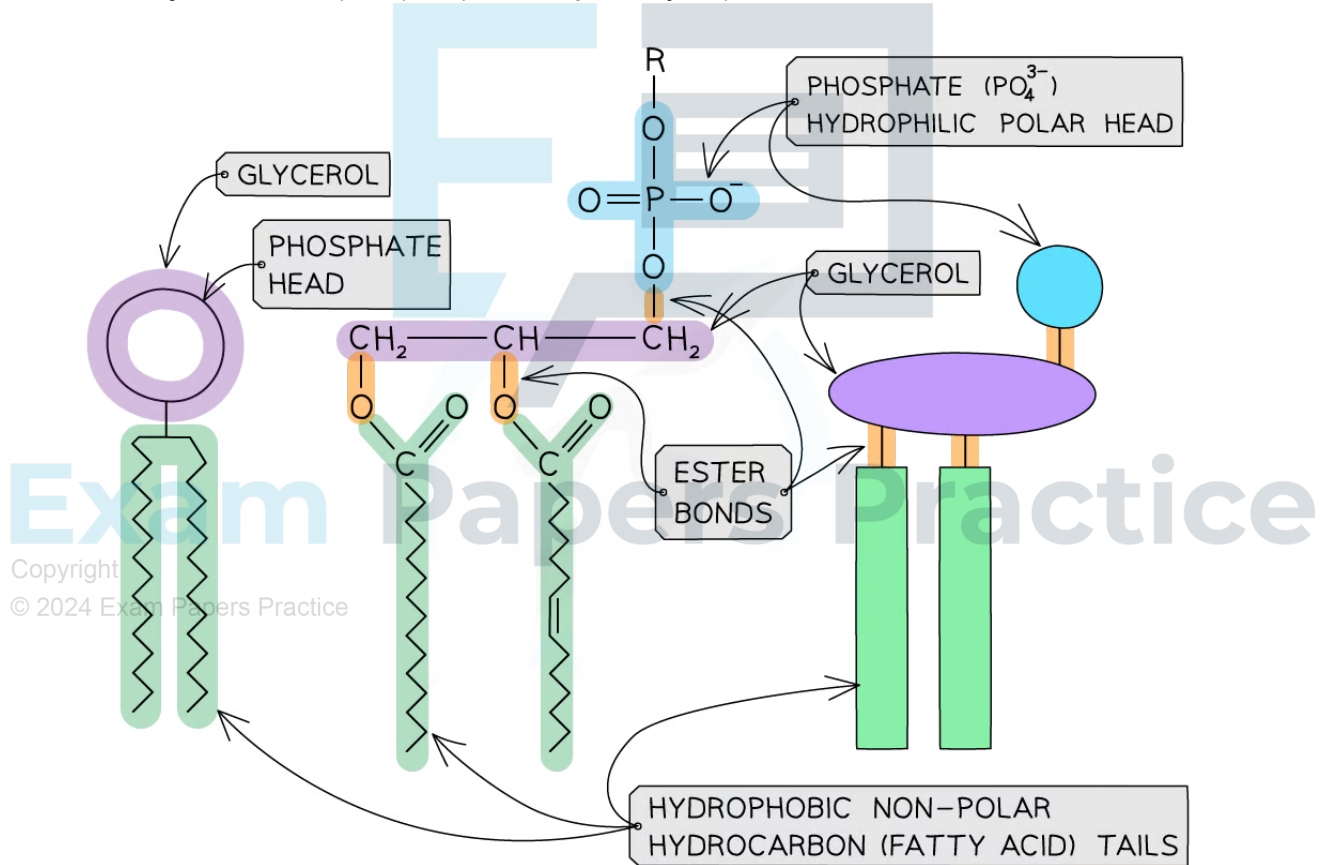
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## 1.3.1 Phospholipid Bilayer Properties

### Amphipathic Properties

#### Phospholipids

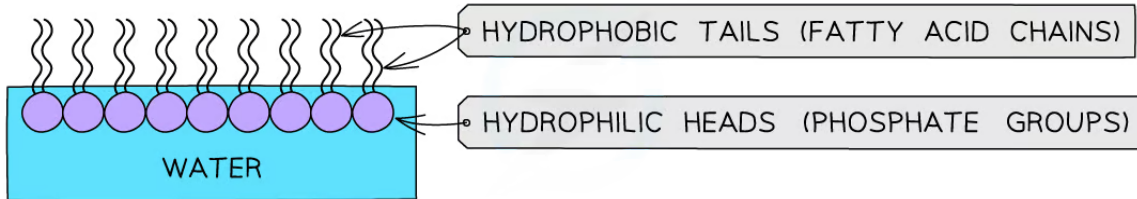
- Phospholipids form the basic structure of the membrane (the phospholipid bilayer)
- They are formed by a hydrophilic **phosphate head** bonding with two hydrophobic **hydrocarbon (fatty acid) tails**
- As phospholipids have a **hydrophobic** and **hydrophilic** part they are known as **amphipathic**
- 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



*The generalised molecular structure of a phospholipid*

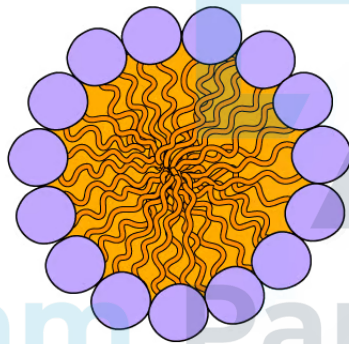
- Due to their **amphipathic** properties, phospholipids display an **emergent property** when placed into water

- The **hydrophilic** phosphate heads orientate towards the water and the **hydrophobic** hydro carbon tails orientate inwards (away from the water)
  - They form a **phospholipid monolayer**

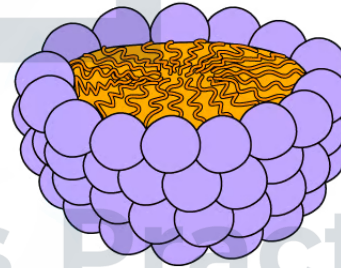


*A phospholipid monolayer*

- 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**



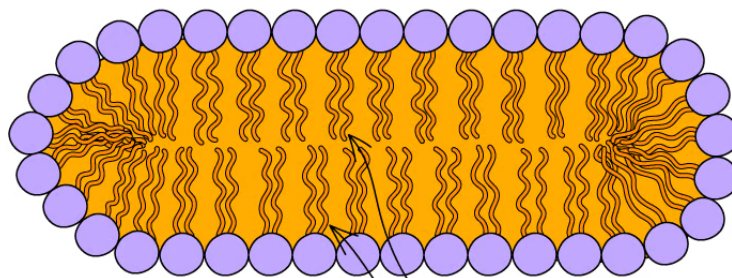
CROSS SECTION OF A SPHERICAL MICELLE



MICELLE IN THREE DIMENSIONS

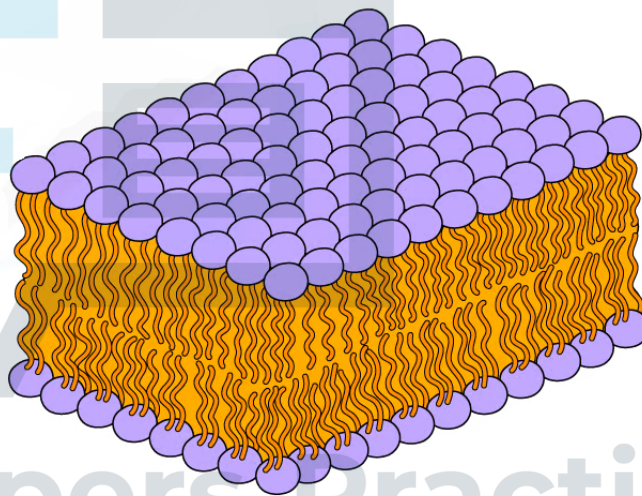
*A micelle*

- 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



TWO LAYERS OF PHOSPHOLIPID MOLECULES (BILAYER)

SHEET-LIKE STRUCTURE OF A BILAYER SEEN IN THREE DIMENSIONS



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*A phospholipid bilayer is composed of two layers of phospholipids; their hydrophobic tails facing inwards and hydrophilic heads outwards*

- The two layers of phospholipids are loosely held together by **weak hydrophobic interactions** between the hydrocarbon tails allowing some membrane fluidity
- The amphipathic properties result in the phospholipid bilayer acting as a **barrier to most water-soluble substances** (the non-polar fatty acid tails prevent polar molecules or ions from passing across the 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

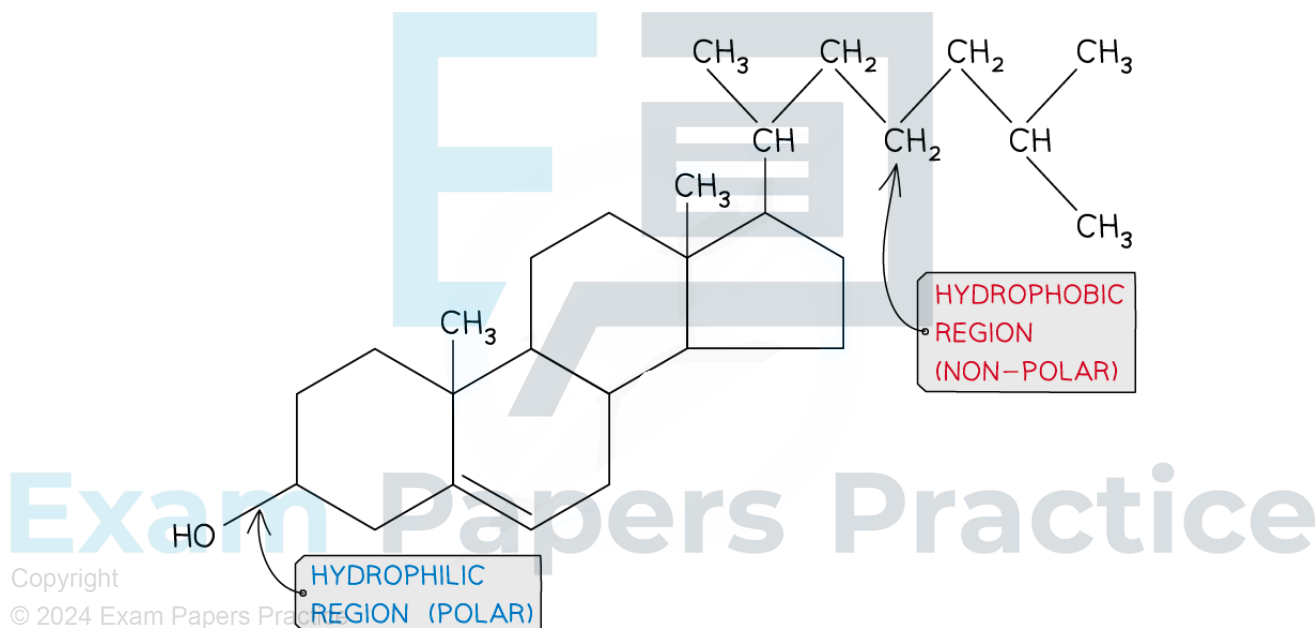


## Animal Cell Membranes: Cholesterol

- **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.

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## 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

### 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)



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### 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

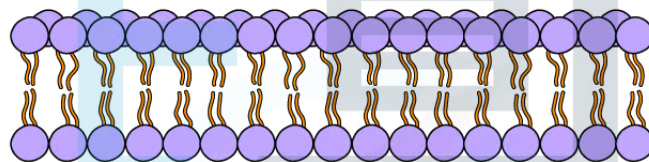
- **Davson and Danielli's** model of the membrane suggested that the **proteins** were arranged in **layers above and below the phospholipid bilayer**
- **Evidence** for this model:
  - Membranes were effective at controlling the movement of substances in and out of cells
  - Electron micrographs showed 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 **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

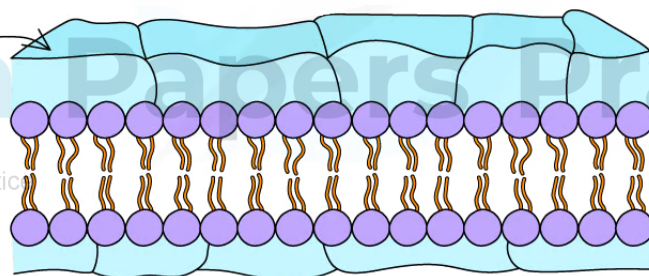
1920's GROTER-GRENDEL MODEL



B

1930's DAVSON-DANIELLI MODEL

GLOBULAR PROTEIN



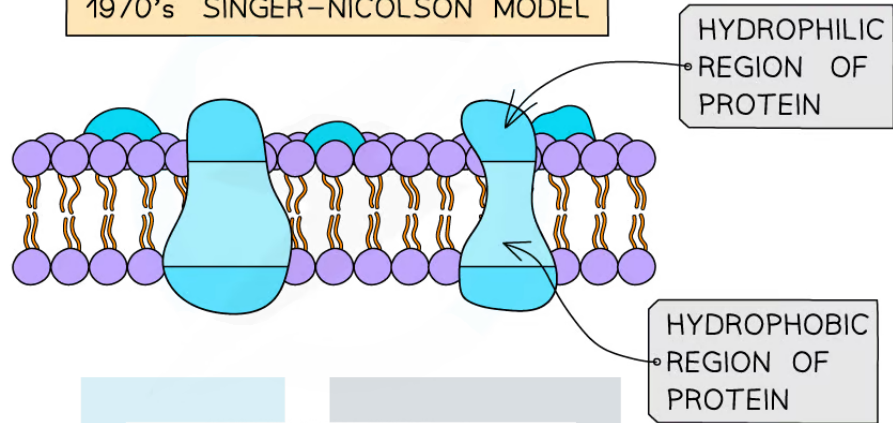
PHOSPHOLIPID BILAYER

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C

1970's SINGER-NICOLSON MODEL



*Three models of membrane structure*

**Future models**

- With further developments in technology more is still to be discovered about the plasma membrane and so the model we use to represent it continues to evolve
  - e.g. the presence of the cellular cytoskeleton on the inside and the extracellular matrix on the outside makes the membrane less fluid than suggested by the fluid mosaic model

**Exam Tip**

You will need to learn the difference between the Davson-Danielli and Singer-Nicolson model of membrane structure and the reasons why they proposed their models.

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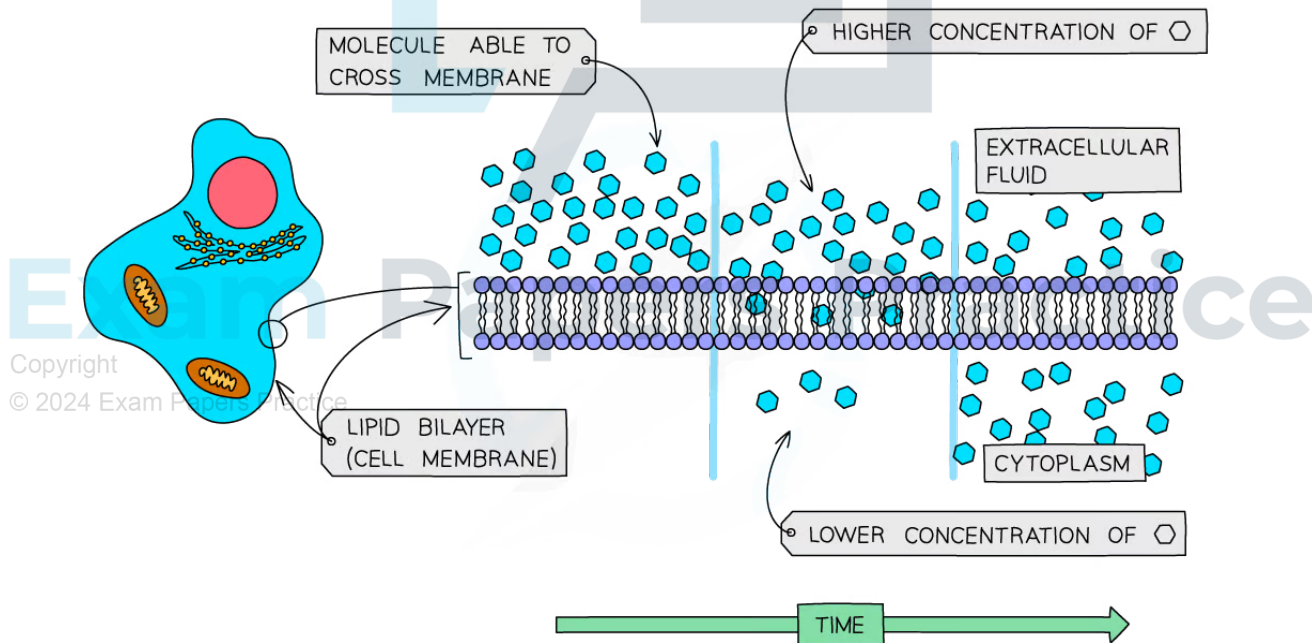
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## 1.3.4 Membrane Transport

### 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



*Diffusion across the cell membrane*



- The **rate** at which a substance diffuses across a membrane depends on several factors:
  - '**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

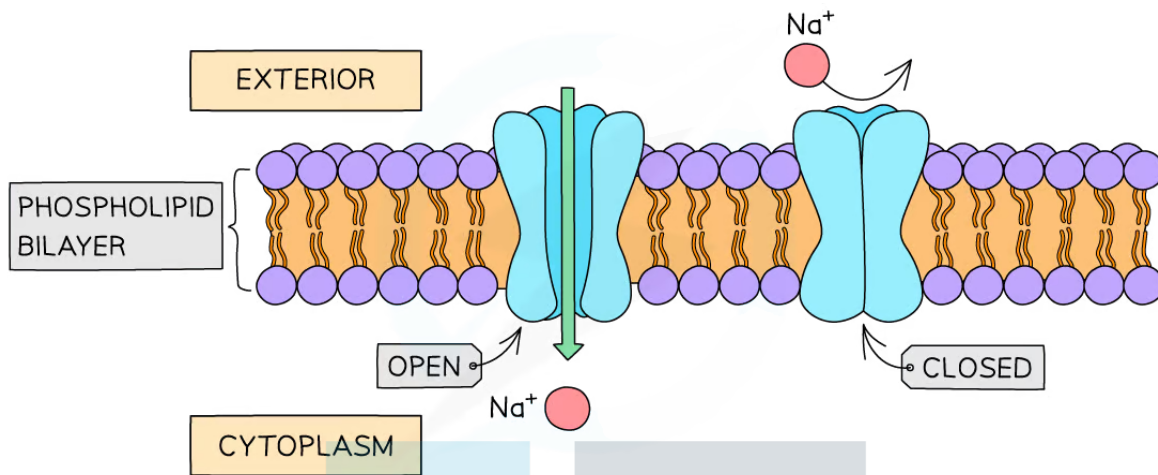
- Certain substances cannot diffuse through the phospholipid bilayer of cell membranes. These include:
  - **Large polar molecules** such as glucose and amino acids
  - **Ions** such as sodium ions ( $\text{Na}^+$ ) and chloride ions ( $\text{Cl}^-$ )
- These substances can only cross the phospholipid 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

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- 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



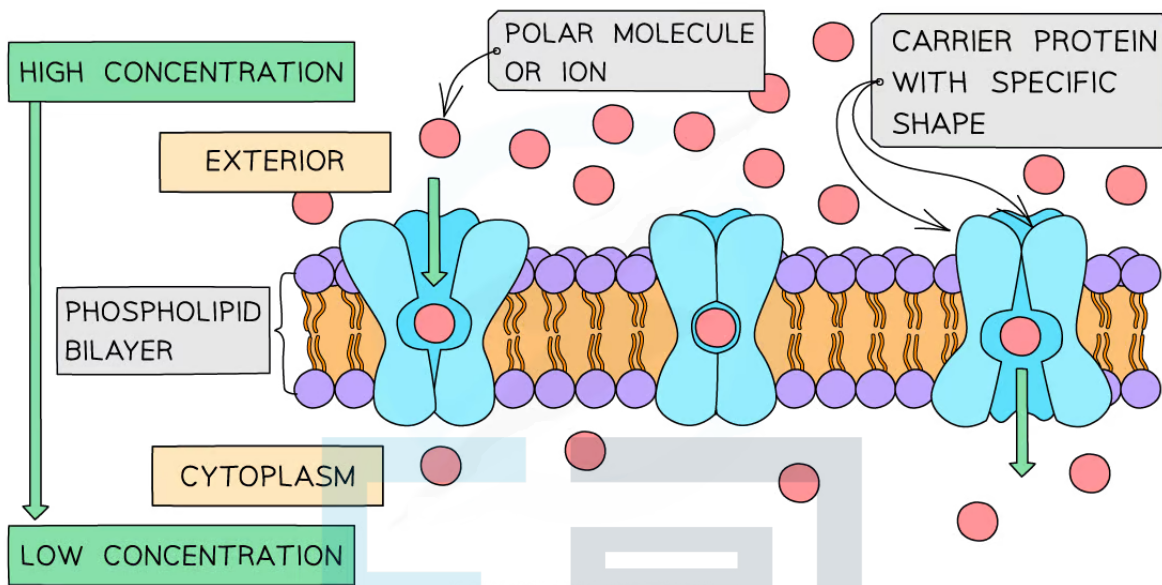
*A channel protein (open and closed)*

### 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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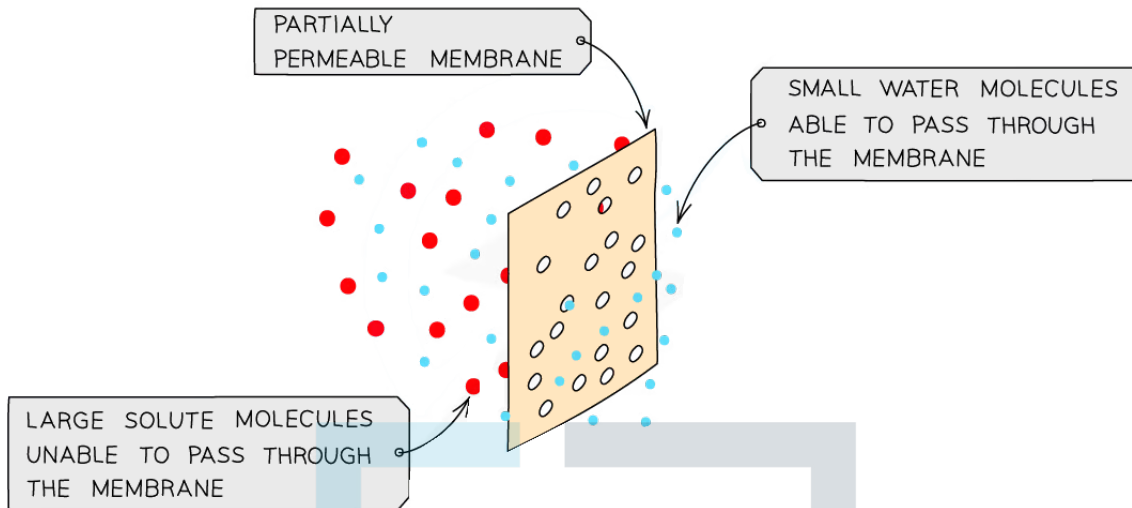


*A carrier protein changing shape during facilitated diffusion*

### 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)

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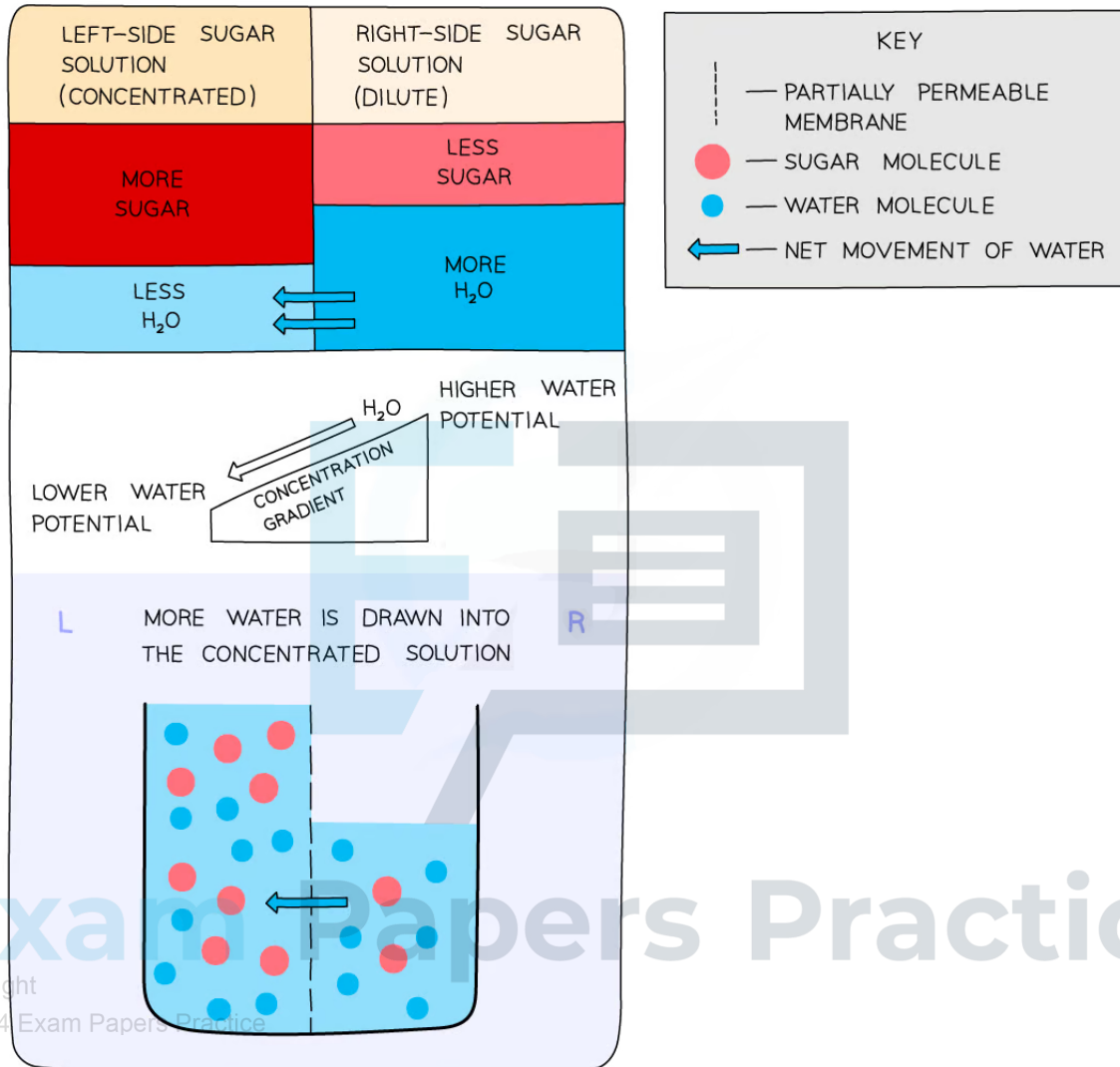


### *Osmosis and the partially permeable membrane*

- 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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*How osmosis works. The water moves from the region of higher water potential (dilute solution) to the region of lower water potential (concentrated solution).*

## 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**

HYPERTONIC SOLUTION	ISOTONIC SOLUTION	HYPOTONIC SOLUTION
<ul style="list-style-type: none"> <li>— RED BLOOD CELLS HAVE HIGHER WATER POTENTIAL THAN SOLUTION</li> <li>— NET MOVEMENT OF WATER OUT</li> <li>— SHRIVELLED CELLS</li> </ul>	<ul style="list-style-type: none"> <li>— WATER POTENTIAL EQUAL BETWEEN RED BLOOD CELL AND SOLUTION</li> <li>— NO NET MOVEMENT OF WATER</li> <li>— NORMAL CELLS</li> </ul>	<ul style="list-style-type: none"> <li>— RED BLOOD CELLS HAVE LOWER WATER POTENTIAL THAN SOLUTION</li> <li>— NET MOVEMENT OF WATER IN</li> <li>— CELLS SWELL, MAY LYSE (BURST)</li> </ul>

<p><b>KEY</b></p> <p> = MOVEMENT OF WATER BY OSMOSIS</p> <p> = SOLUTE</p>
---------------------------------------------------------------------------

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## *Effect of osmosis on animal cells*

### 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 1L 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:
  - 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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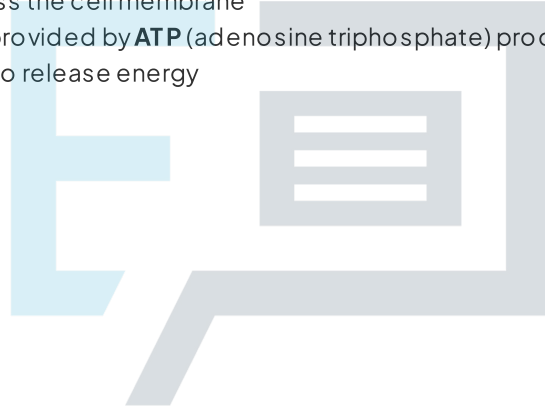
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## 1.3.5 Active Transport & Bulk Transport

### 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



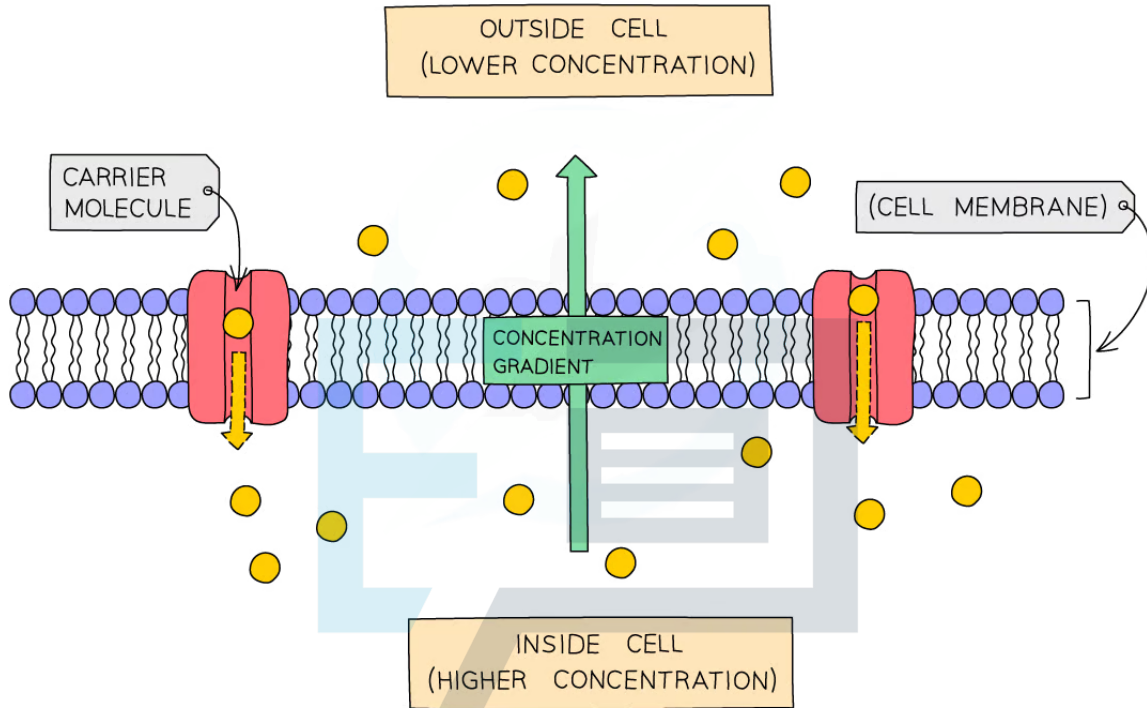
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ACTIVE TRANSPORT ACROSS THE CELL MEMBRANE



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*Active transport across the cell membrane*

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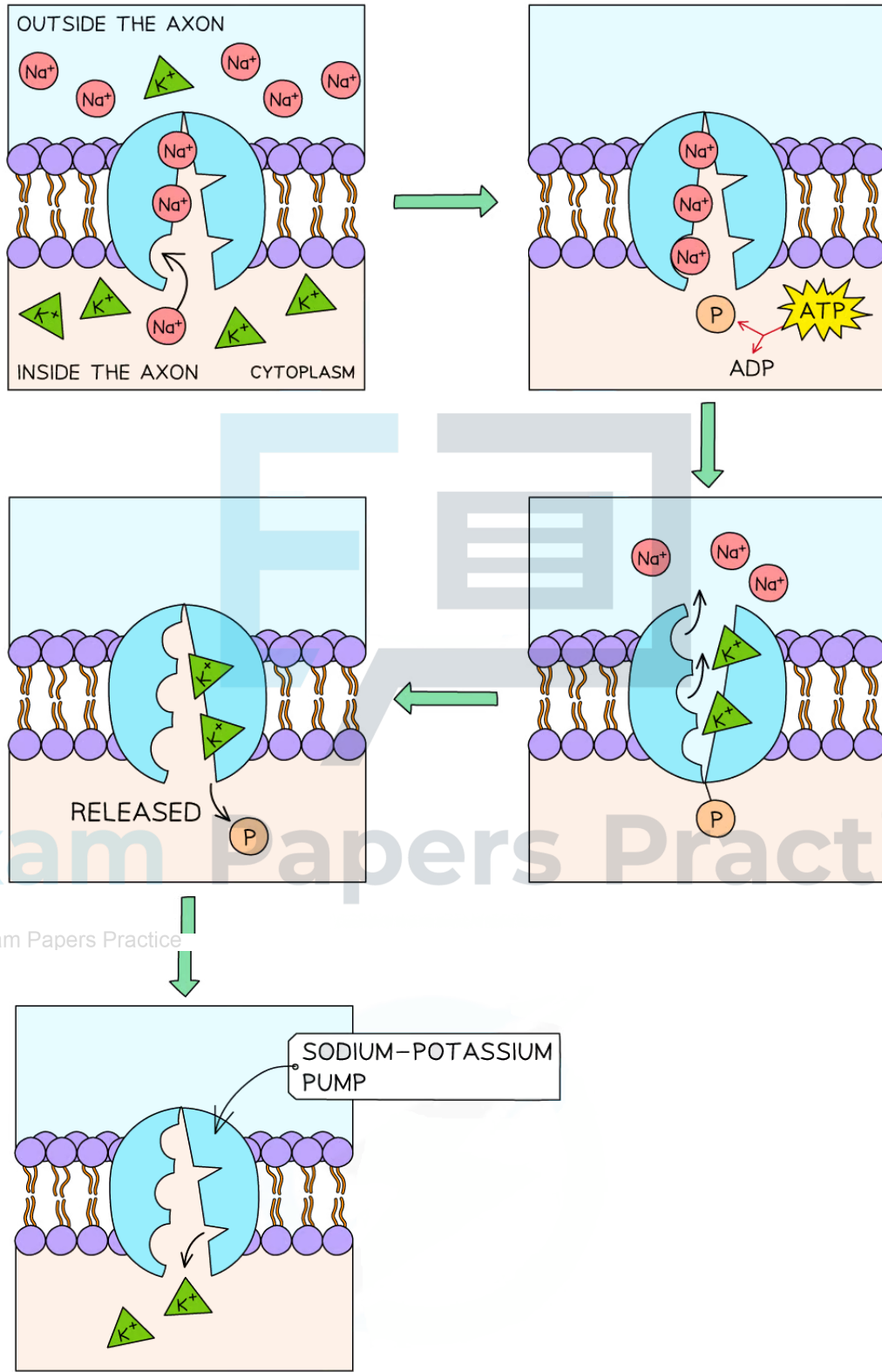
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## Active Transport: Example

### 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:
  - **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

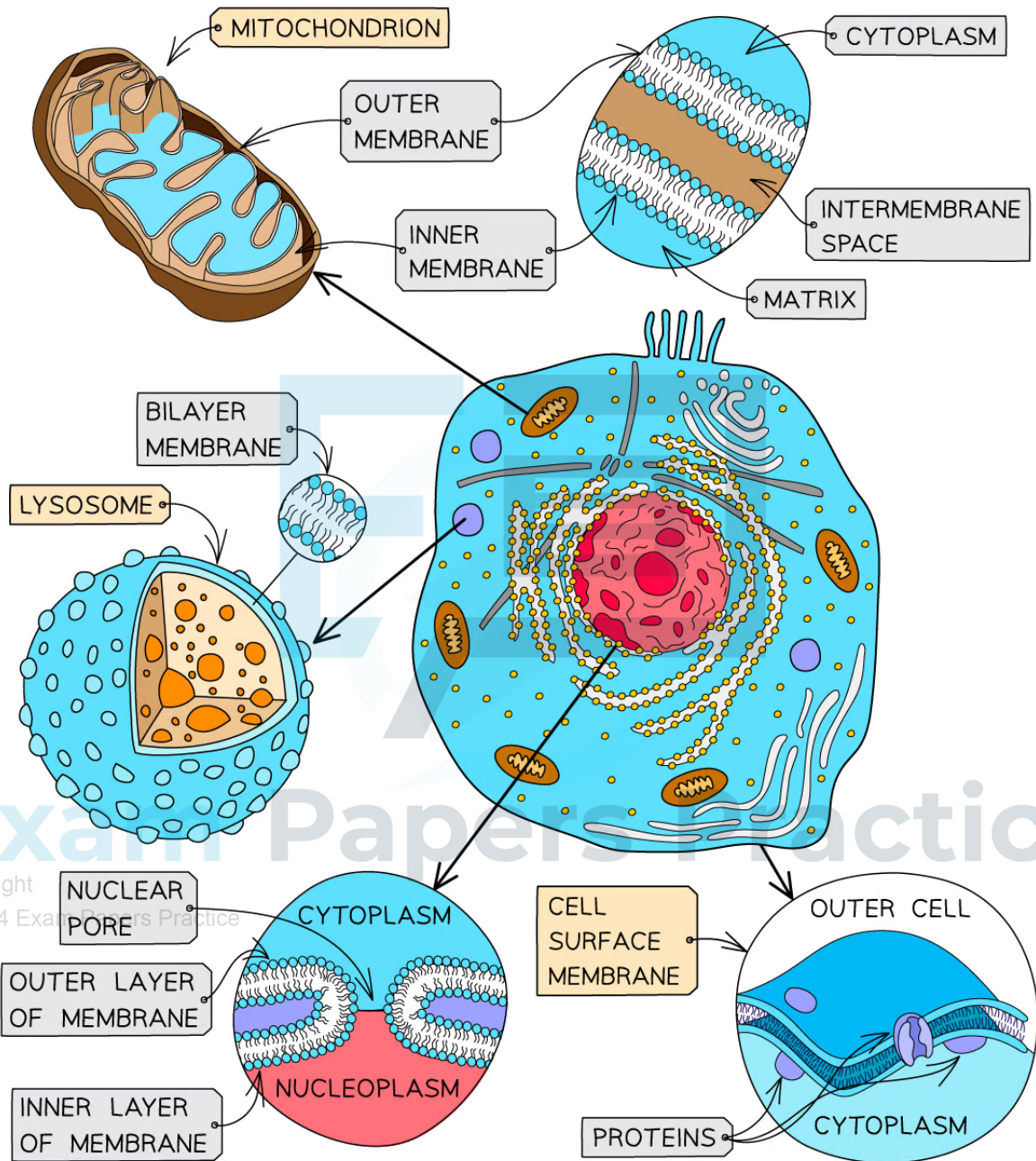
#### Membranes

- 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**

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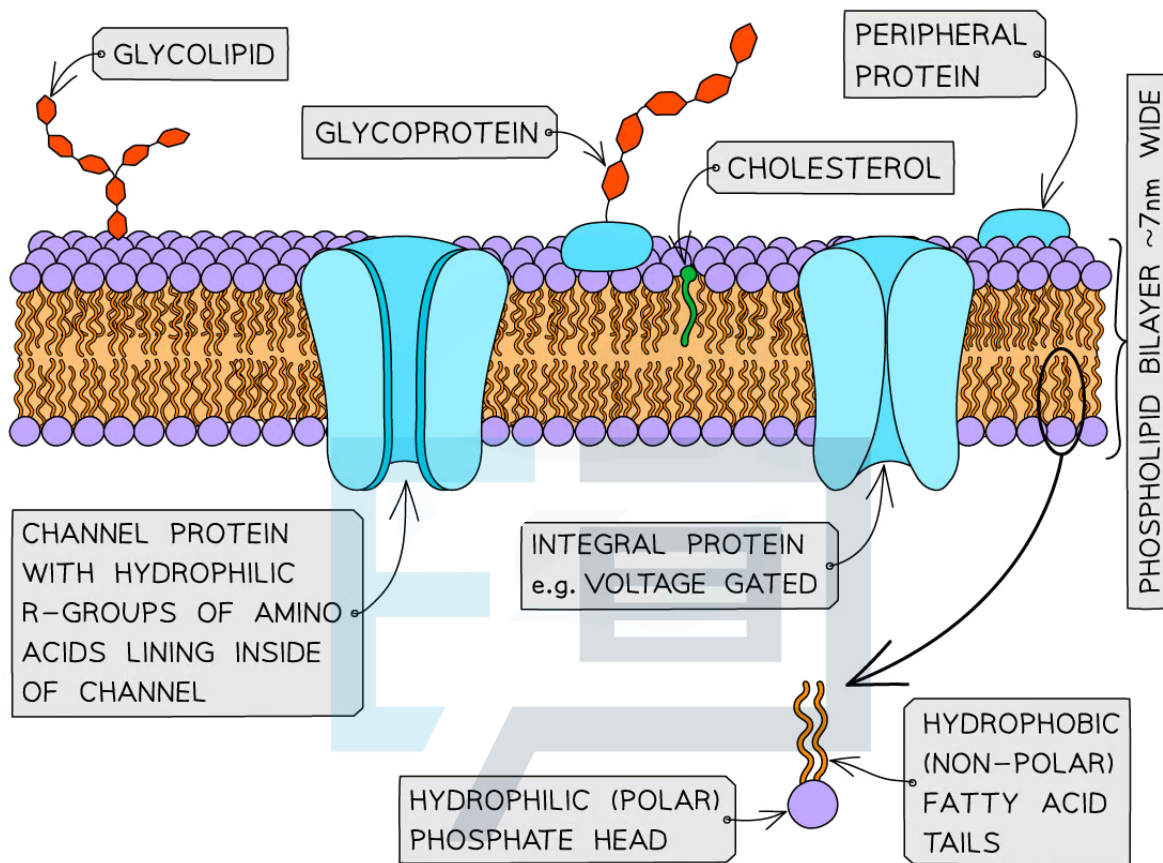
*Membranes formed from phospholipid bilayers help to compartmentalise different regions within the cell, as well as forming the cell surface membrane*

### 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
- 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



*The main components of cell membranes. The distribution of the proteins within the membrane gives a mosaic appearance and the structure of the proteins determines their position in the membrane.*

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### Exam Tip

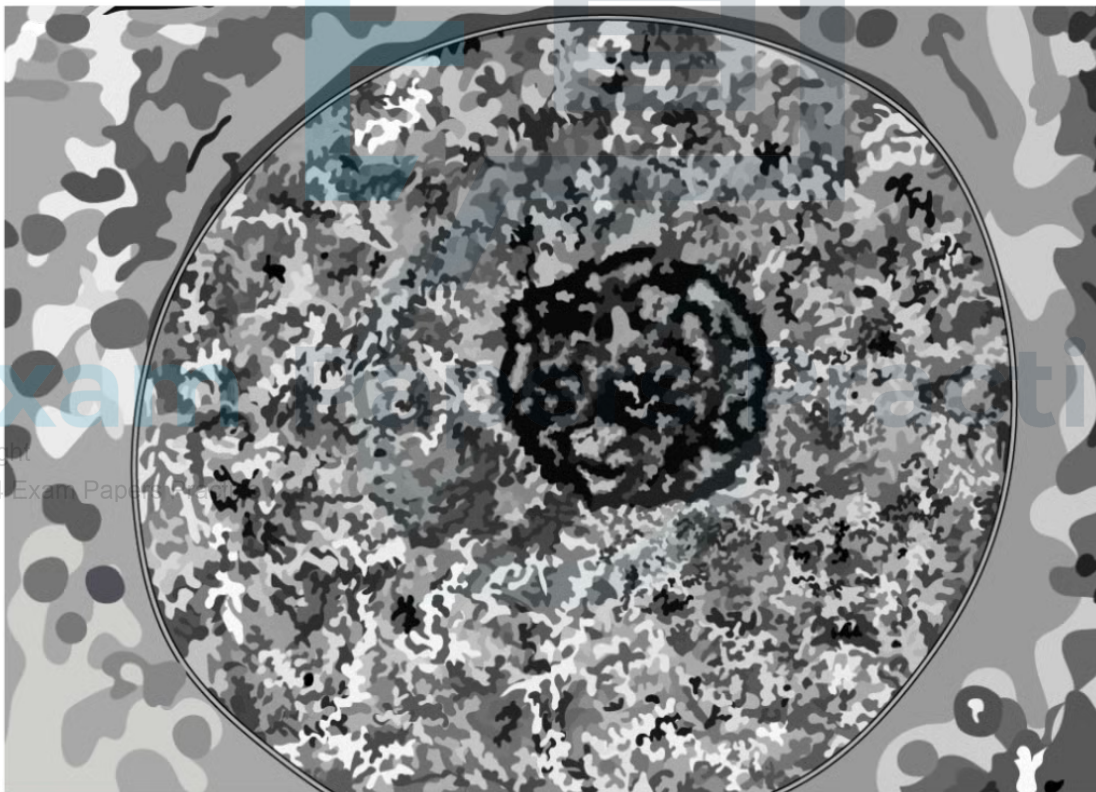
When drawing the fluid mosaic model remember to include (and label) the **phospholipid bilayer** (making it clear which part is the phosphate head and which parts are the hydrocarbon tails), the **thickness of the membrane (7 - 10 nm)**, **integral proteins** (show them embedded in the phospholipid bilayer 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).

## 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

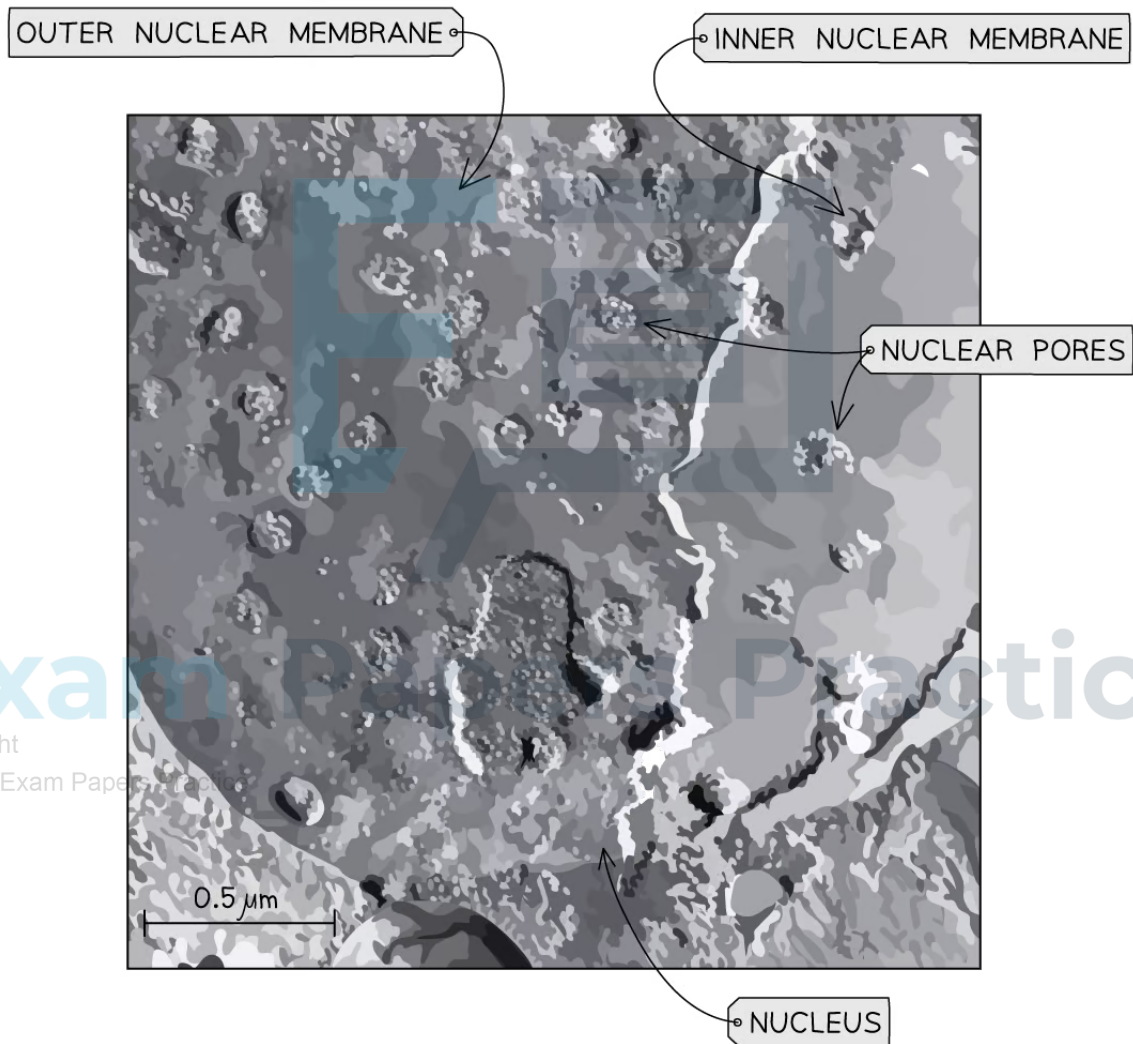
- When analysing transmission electron micrographs comment on:
  - How the membrane has **two darker layers** surrounding a **lighter** line
  - **Proteins** were known to appear **darker** in electron micrographs
- These were the observations that **supported** Davson–Danielli's model



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

### Freeze-etched electron micrographs

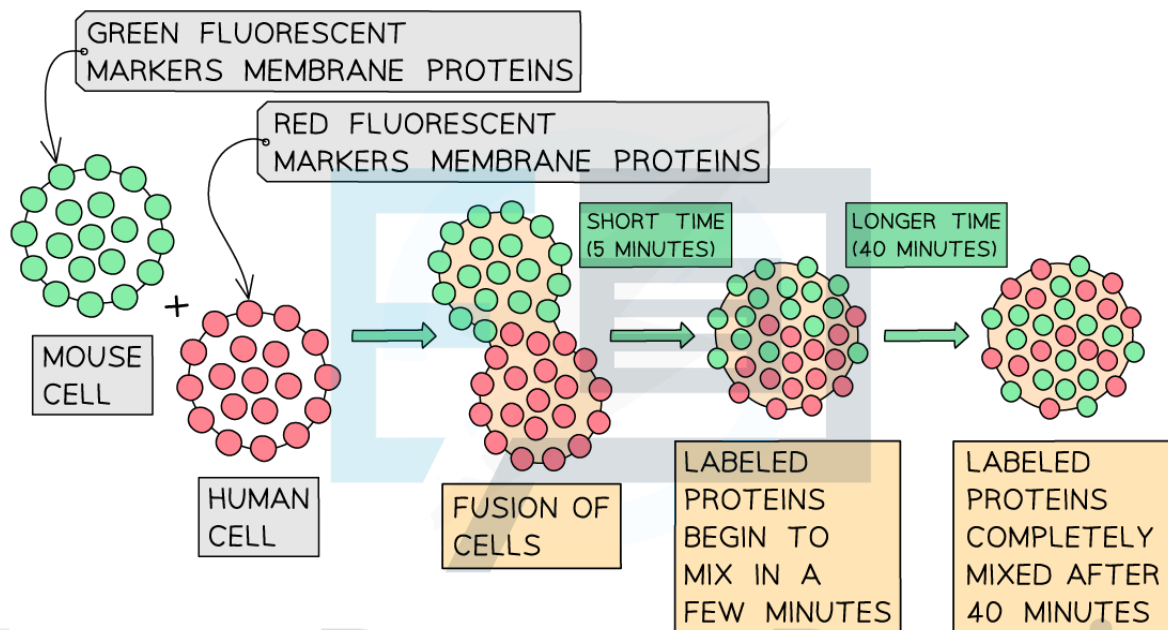
- When asked to analyse freeze-etched electron micrographs note that the very **small bumps** seen on the membranes are the **integral proteins**
- This provided evidence **against** Davson-Danielli's model as it showed proteins extending into the **centre** of the membrane
- Be careful if the image is of a nuclear membrane as the larger circles represent the nuclear pores



*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



*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.

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## 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 1L 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

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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)
- 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

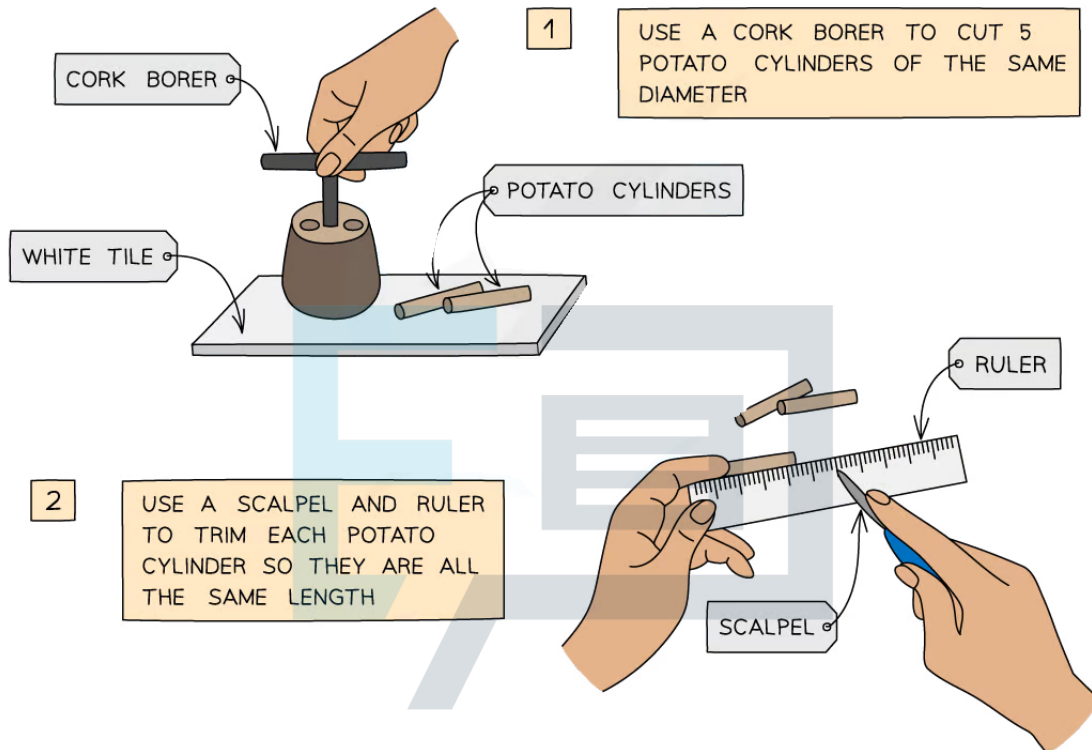
- Potato x2 (same variety)
- Cork borer (e.g. 5mm)
- White tile
- Scalpel
- 10cm ruler or vernier calipers
- Weighing balance (2dp)
- 10 cm<sup>3</sup> sucrose solution (0 mol/dm<sup>3</sup>, 0.25 mol/dm<sup>3</sup>, 0.5 mol/dm<sup>3</sup>, 0.75 mol/dm<sup>3</sup>, 1.00 mol/dm<sup>3</sup>)
- 5 test tubes (in test tube rack)
- 10 cm<sup>3</sup> 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<sup>o</sup>)
  - 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



OSMOSIS METHOD



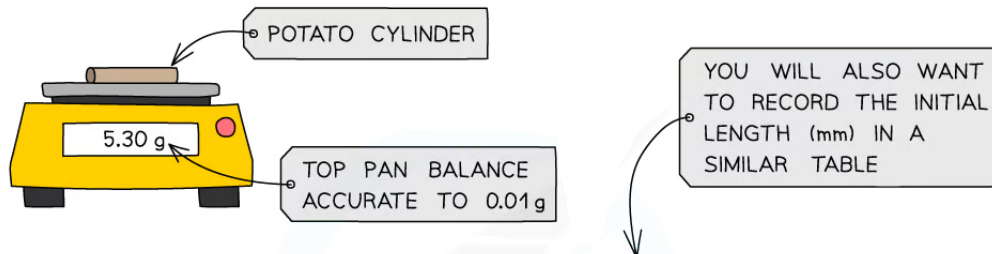
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3 MEASURE THE MASS OF EACH POTATO CYLINDER AND RECORD IN A TABLE OF RESULTS



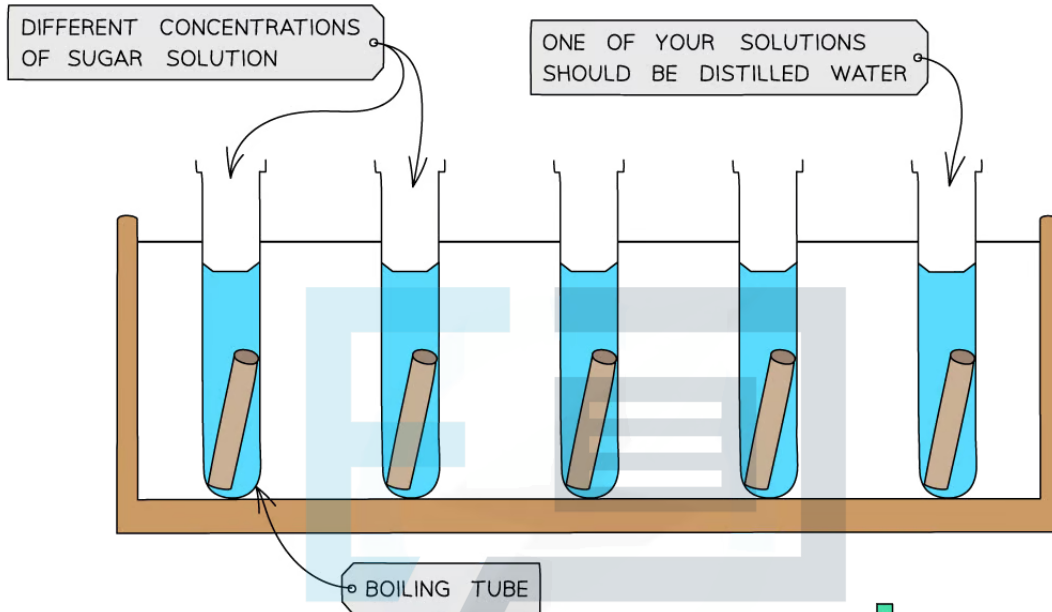
Concentration of sucrose solution mol/dm <sup>3</sup>	Initial mass (g)	Final mass (g)	Change in mass (g)	% change in mass
0 (distilled water)	5.30			
0.25	5.32			
0.50	5.29			
0.75	5.31			
1.00	5.29			

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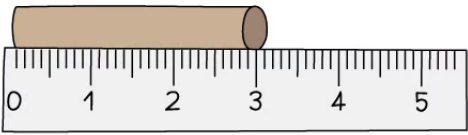
4 MEASURE  $10\text{cm}^3$  OF EACH SUGAR OR SALT SOLUTION AND POUR INTO EACH BOILING TUBE. LABEL EACH BOILING TUBE CLEARLY



5 ADD ONE POTATO CYLINDER TO EACH BOILING TUBE AND LEAVE FOR A SPECIFIED AMOUNT OF TIME

AFTER A SET TIME

6 REMOVE THE POTATOES. BLOT DRY AND RECORD THE FINAL MASS AND LENGTH OF EACH



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



OSMOSIS ANALYSIS

Concentration of sucrose solution mol/dm <sup>3</sup>	Initial mass (g)	Final mass (g)	Change in mass (g)	% change in mass
0 (distilled water)	5.30	5.80	+0.50	9.4
0.25	5.32	5.42	+0.10	?
0.50	5.29	5.24	-0.05	-1.0
0.75	5.31	5.11	-0.20	-3.8
1.00	5.29	5.02	-0.27	-5.1

1

CALCULATE THE PERCENTAGE CHANGE IN MASS FOR EACH CYLINDER

$$\frac{(\text{FINAL MASS} - \text{INITIAL MASS})}{\text{INITIAL MASS}} \times 100$$

e.g. FOR 0.25 mol/dm<sup>3</sup>

$$= \frac{(5.42 - 5.32)}{5.32} \times 100$$

$$= 1.9\%$$

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To find the percentage change in mass, the change in mass must be divided by the initial mass and then multiplied by 100



*A positive percentage change in mass indicates that the potato has gained water by osmosis*

- A **positive** percentage change in mass indicates that the potato has gained water by osmosis (net movement of water from the solution into the potato) meaning the **solution** had a **lower osmolarity** than the potato
  - The gain of water makes the potato cells **turgid**, as the water exerts turgor pressure (or hydrostatic pressure) on the cell walls – the potatoes will feel hard
- A **negative** percentage change suggests the opposite, that is, the solution had a **higher osmolarity** than the potato
  - The potato cylinder in the **strongest sucrose concentration** will have **decreased in mass** the most as there is the **greatest concentration gradient** in this tube between the potato cells



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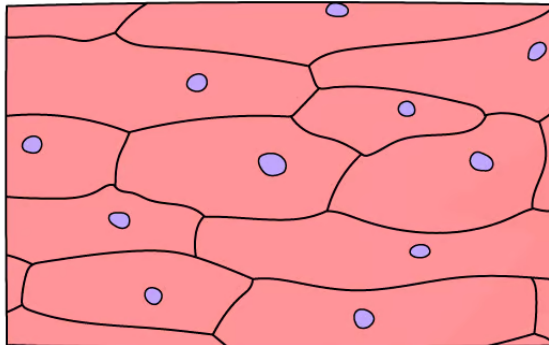
- (lower osmolarity) and the sucrose solution (higher osmolarity)
- More water molecules will move out of the potato cells by **osmosis**, making them **flaccid** and decreasing the mass of the potato cylinder – the potato cylinders will feel floppy
  - If looked at underneath the microscope, cells from 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

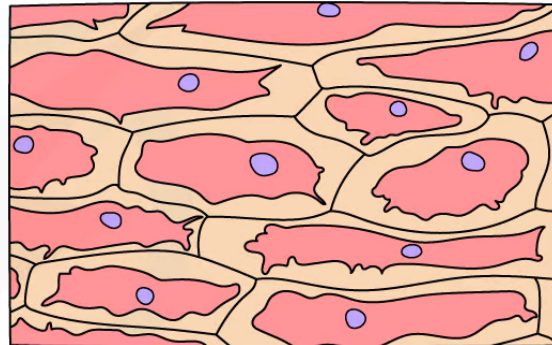
- Evidence of osmosis occurring in plant cells can be shown when the cells undergo **plasmolysis**:
  - If a plant cell is placed in a solution with a **higher osmolarity** than the cell (such as a concentrated sucrose solution), water will **leave** the cell through its partially permeable cell surface 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

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NORMAL RED ONION CELLS



PLASMOLYSED RED ONION CELLS

*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  $1.0 \text{ mol dm}^{-3}$  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.