Cell membrane structures 2

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Schedule of the present Lecture

1. **The Plasma Membrane, the Lipid Bilayer**

- *Chemical structure of cell membranes*
- *Membrane proteins*

2. **Principles of Membrane Transport**

- *Transporters, passive and active membrane transport*
- *Ion channels and the electrical properties of membranes*

2. Principles of Membrane Transport

Transporters, passive and active membrane transport

Membrane transport function

Because of its hydrophobic interior, the lipid bilayer of cell membranes prevents the passage of most polar molecules

This barrier function allows the cell to maintain concentrations of solutes in its cytosol that differ from those in the extracellular fluid and in each of the intracellular enclosed compartments

The ion concentrations in and out

Typical mammalian cell

COMPONENT	INTRACELLULAR CONCENTRATION (mM)	EXTRACELLULAR CONCENTRATION (mM)
Cations		
Na^+	5–15	145
K^+	140	5
Mg^{2+}	0.5	1–2
Ca^{2+}	10^{-4}	1–2
H^+	7×10^{-5} ($10^{-7.2}$ M or pH 7.2)	4×10^{-5} ($10^{-7.4}$ M or pH 7.4)
Anions*		
Cl^-	5–15	110

*The cell must contain equal quantities of positive and negative charges (that is, it must be electrically neutral). Thus, in addition to Cl^-, the cell contains many other anions not listed in this table; in fact, most cell constituents are negatively charged (HCO_3^-, PO_4^{3-}, proteins, nucleic acids, metabolites carrying phosphate and carboxyl groups, etc.). The concentrations of Ca^{2+} and Mg^{2+} given are for the free ions. There is a total of about 20 mM Mg^{2+} and 1–2 mM Ca^{2+} in cells, but both are mostly bound to proteins and other substances and, for Ca^{2+}, stored within various organelles.

Diffusion

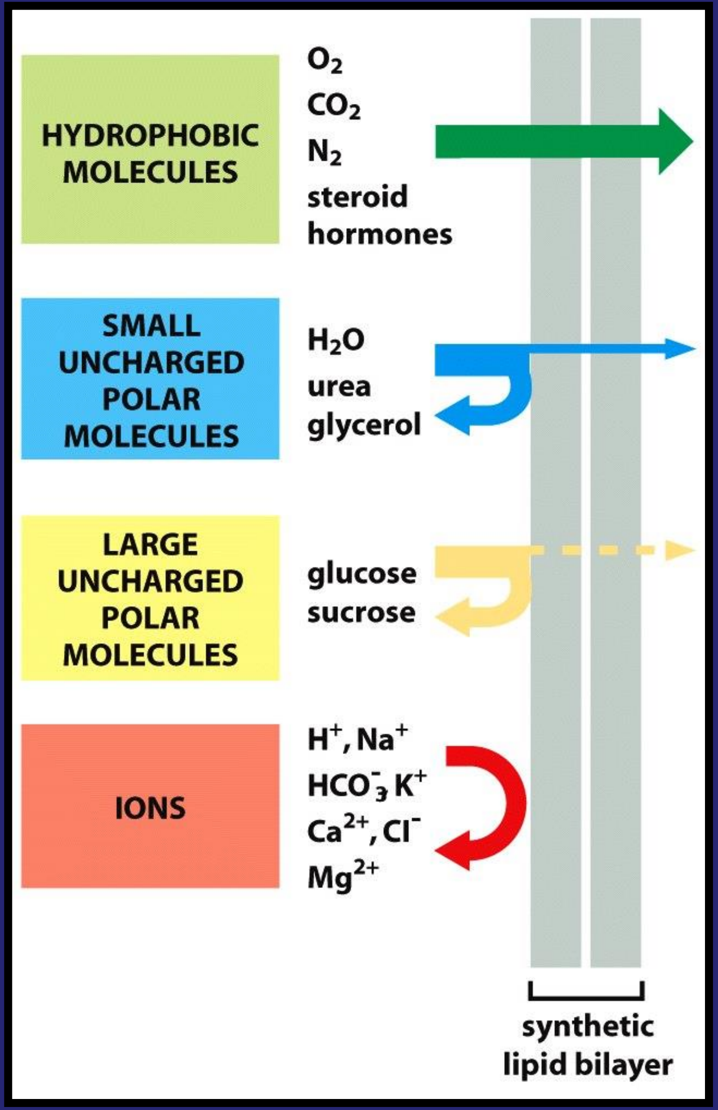
Only for

- molecules dissolvable in lipids
- small molecules without any charge

Any other small organic molecule = some membrane transport molecules must be used

- the transporters are proteins
- collection of the protein differ according to the membrane type

Relative permeability



- the smaller the molecule and, more importantly, the less strongly it associates with water, the more rapidly the molecule diffuses across the bilayer

Membrane transport proteins

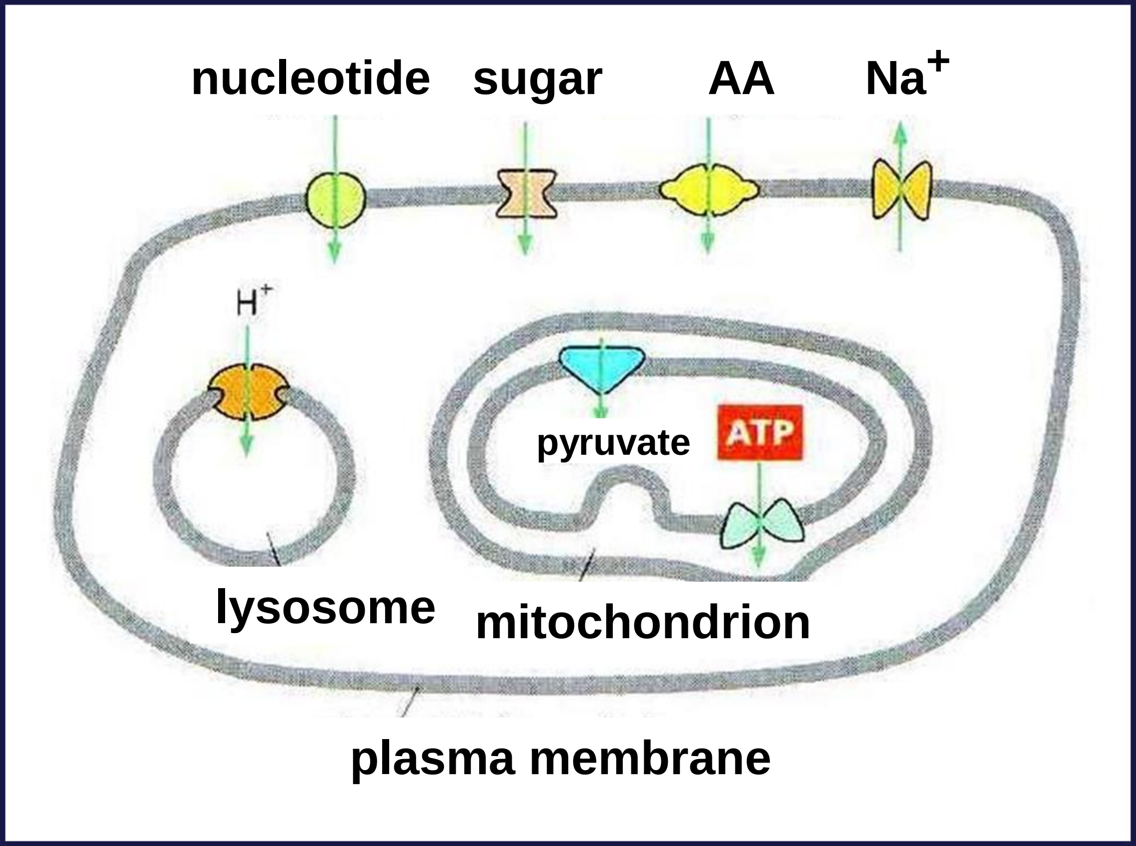
- cross membranes (plasmatic or intracellular) and form channels (narrow hydrophilic pore) allowing transport of specific molecules
- Are responsible for presence of certain molecules in cell or in its compartments

There are two main classes of MTP

- transporters (carriers, permeases)
- channels (for inorganic ions)

Examples

Each membrane has its own set of transporter molecules



Channel proteins

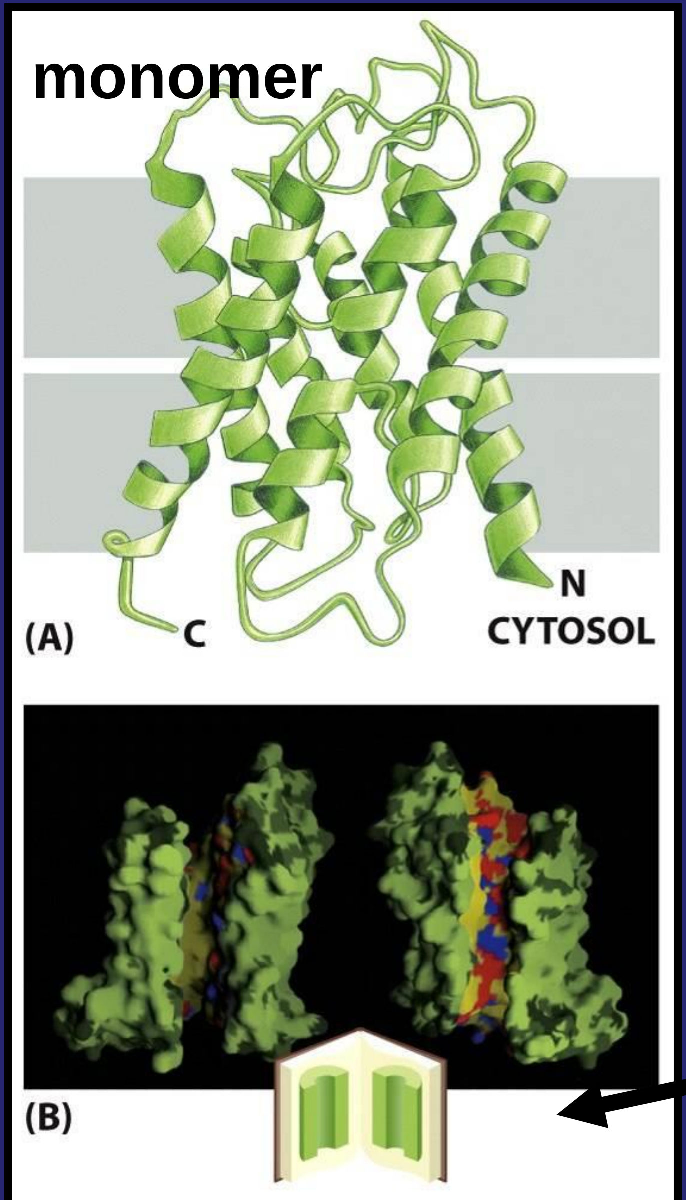
They form

- **Porins and aquaporins** (permanently open, wider, only for water molecules)
- **Ion channels** (narrow channels, may be closed, ion selective = ions are coming through according to their size and electric charge)

If channel is open only small molecules with suitable charge is coming through …

… ions are transported very quickly (more than 1 million ions per second)

Structure of aquaporins



In the membrane, aquaporins form tetramers

Each monomer contains a pore in its centre (not shown)

Space-filling model (cut and opened as a book)

Transporters

- highly selective
- mostly transport only one type of molecule
- select molecules which fit to its binding site

Transport speed is about 1 000 molecules per second

Three types of transporter movement

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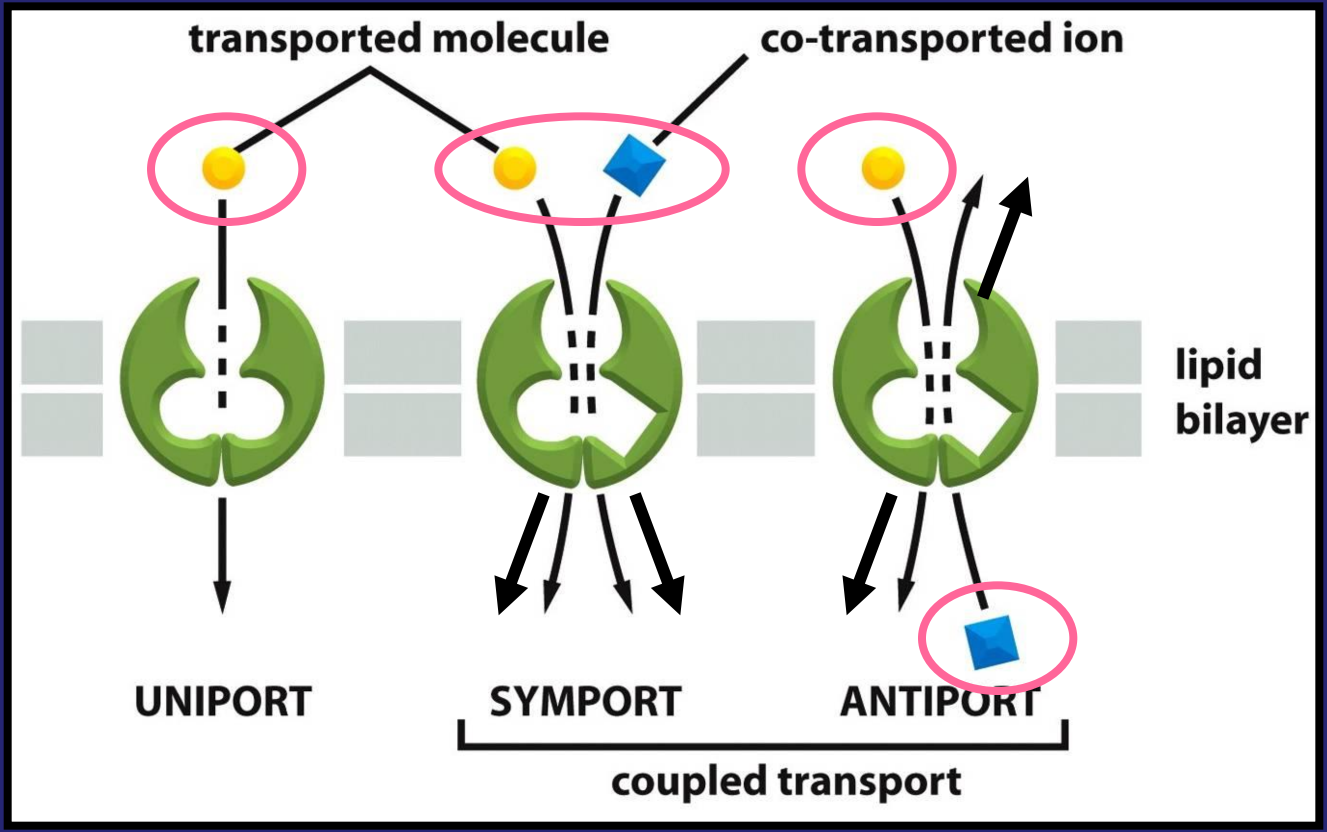
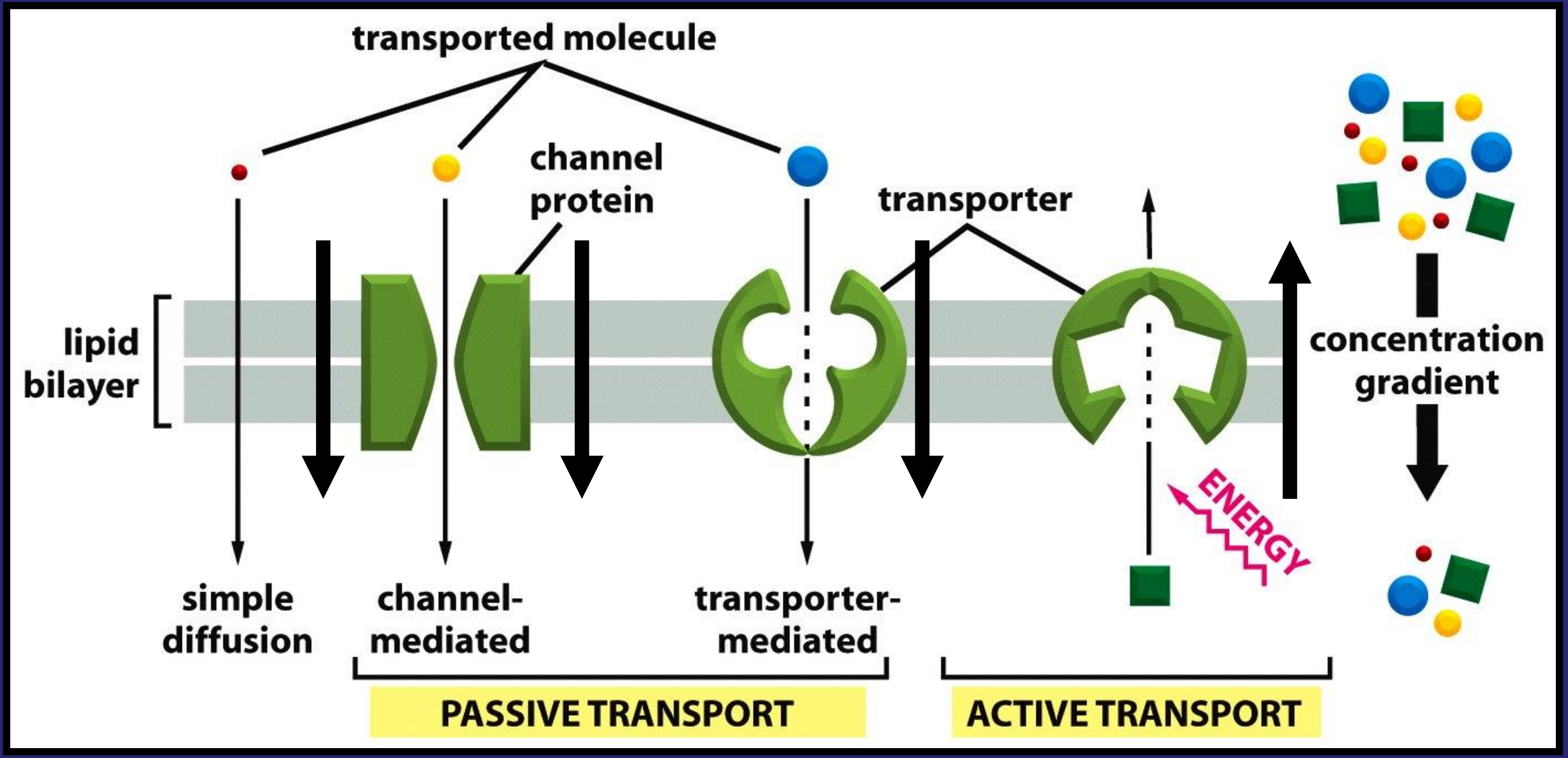


Passive and active transport



Does not require energy
- simple diffusion or facilitated diffusion

Require energy
- it is against electrochemical gradient

Passive transport

Does not require an input of metabolic energy

- **molecules come across the membrane thanks their electrochemical gradients**
- **simple diffusion through the lipid bilayer**
- **facilitated diffusion through channels and passive transporters**

Active transport

Requires an input of metabolic energy

- actively pump certain molecules across the membrane against their electrochemical gradients
- transporters are named pumps
- transport is coupled with ATP hydrolysis

Three types of active transport

- **Coupled transport**

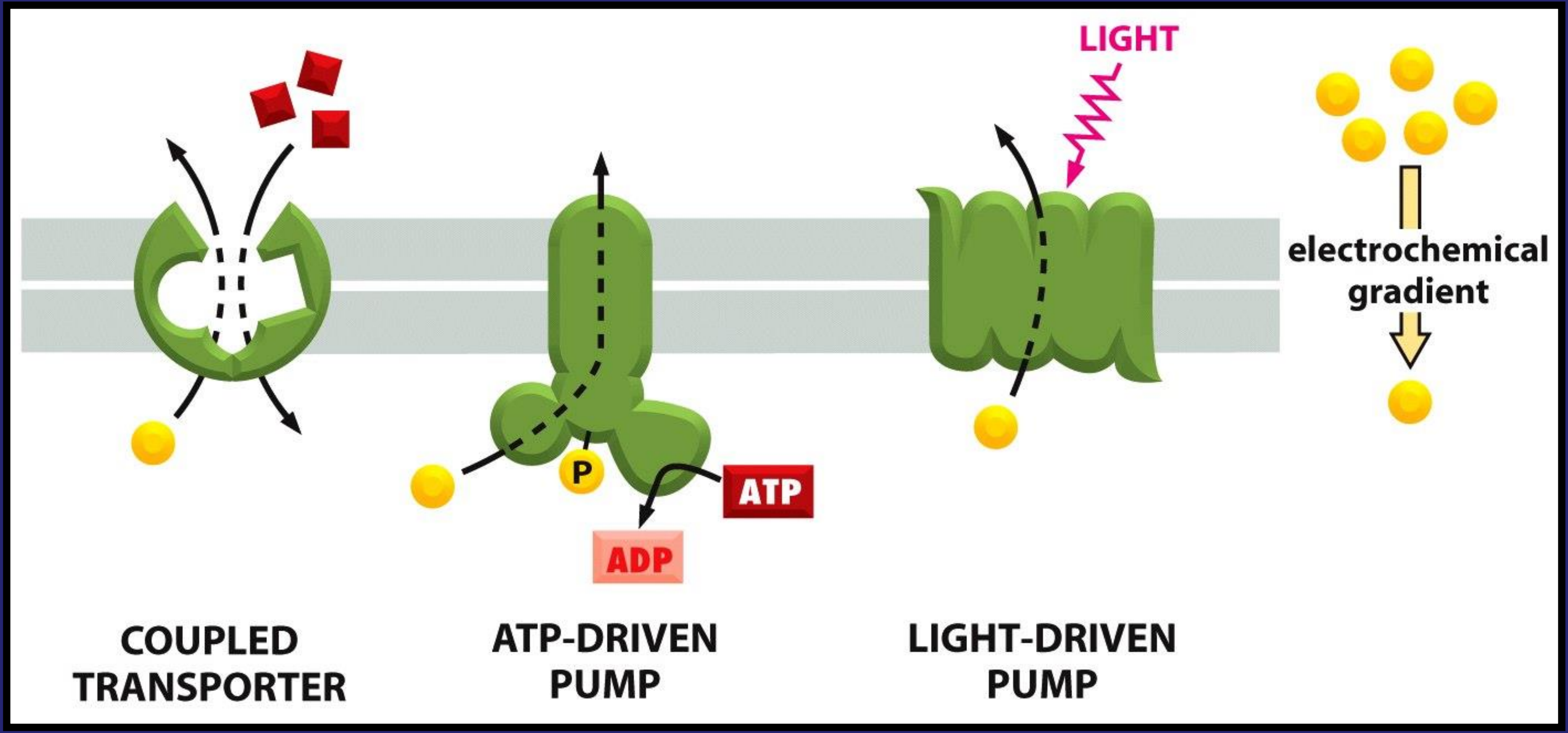
  One molecule is transported by ion gradient (down the hill) and another against it (up the hill)

- **ATP-driven pump**

  The energy for up the hill transport is coupled with ATP hydrolysis

- **Light-driven pump** (only halobacteria)

Three types of active transport



Examples of coupled transport

- **Animal cells**
  use to support the active transport Na^{+} ions gradient

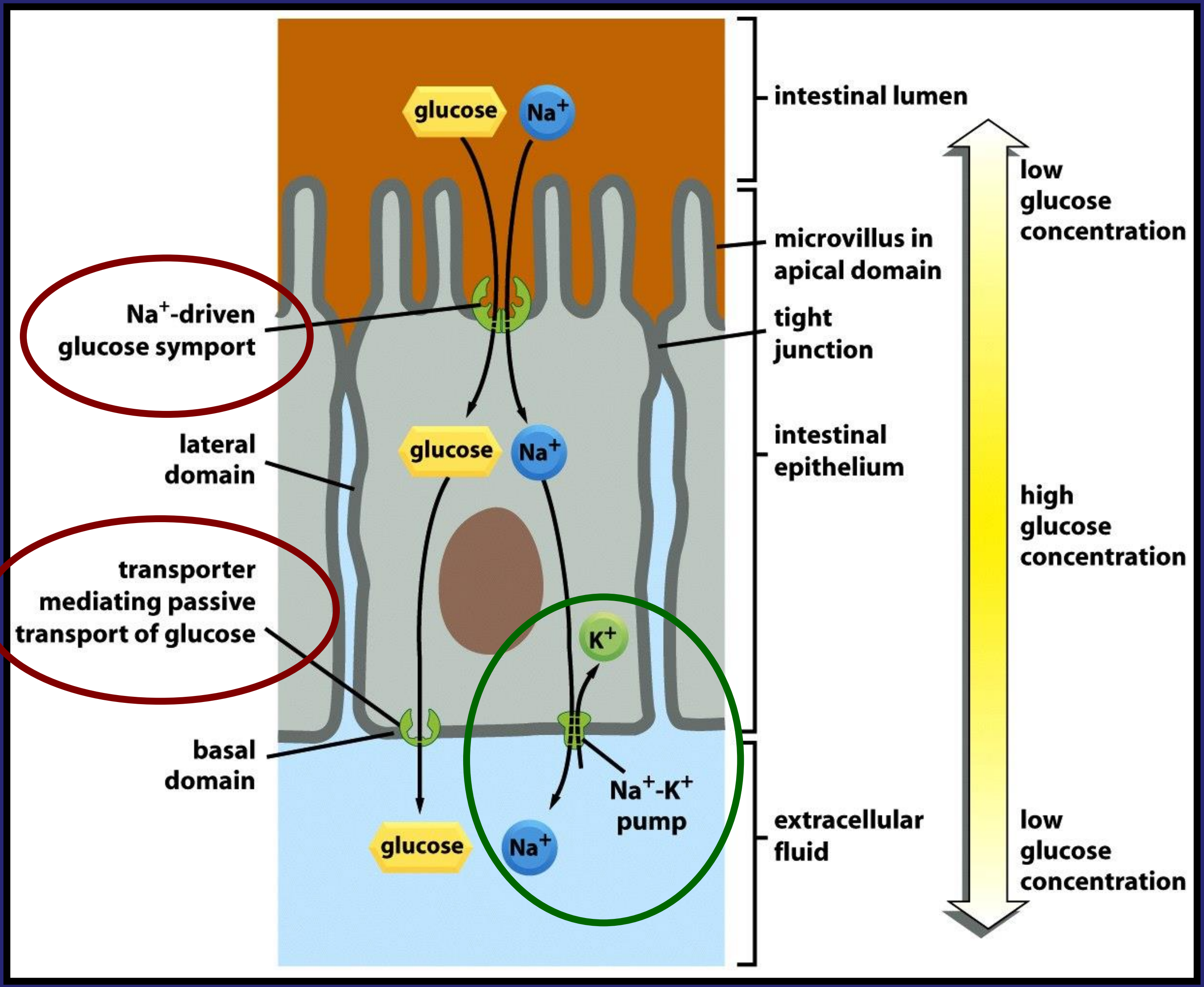
- **Plant cells, bacteria, fungi, and yeast**
  use to support the active transport H^{+} ions gradient

Examples of protein transporters

Transporter	Energy source	Function
glc transporter	Na^+ gradient	Active transport of glc
Na^+ and H^+ antiport	ATP hydrolysis	Active export of H^+, pH regulation
Na-K ATPase	ATP hydrolysis	Active export of Na^+ and import K^+, osmotic balancy
Ca ATPase	ATP hydrolysis	Active export Ca^{2+} from cytosole
H ATPase	ATP hydrolysis	Active export of H^+ from cell
bacteriorhodopsin	light	Active export of H^+ from cell

Glucose can be transported by active or passive transport

Transcellular transport of glucose

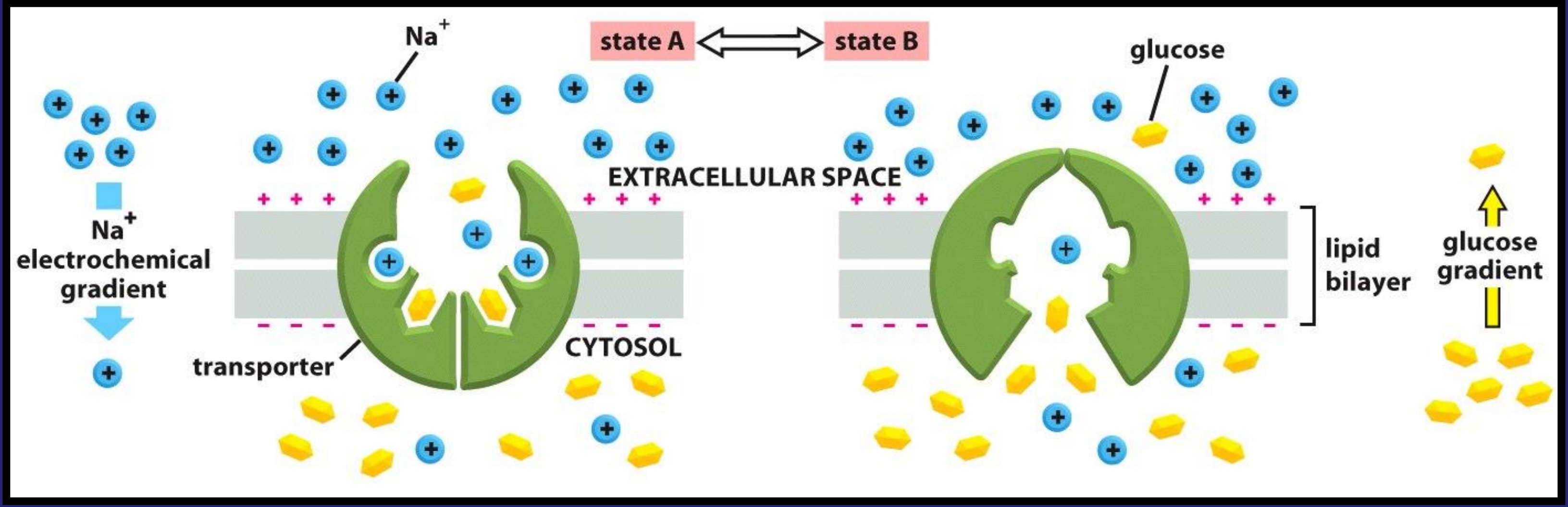


Glucose transporter can be driven by a Na^+ gradient

Transporter oscillates between two alternate states

state A: the protein is open to extracellular space

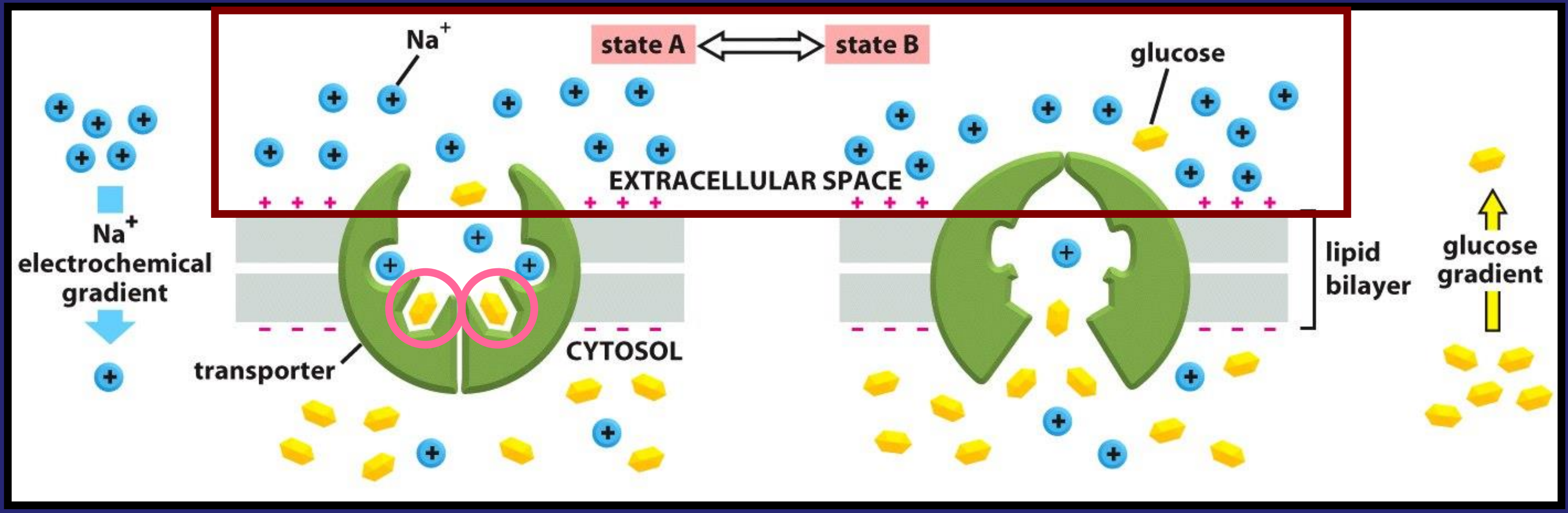
state B: it is open to the cytosol



Binding Na^+ and glc is cooperative – ligand induces the conformational change that increase the protein's affinity for the other ligands

Glucose transporter can be driven by a Na^+ gradient

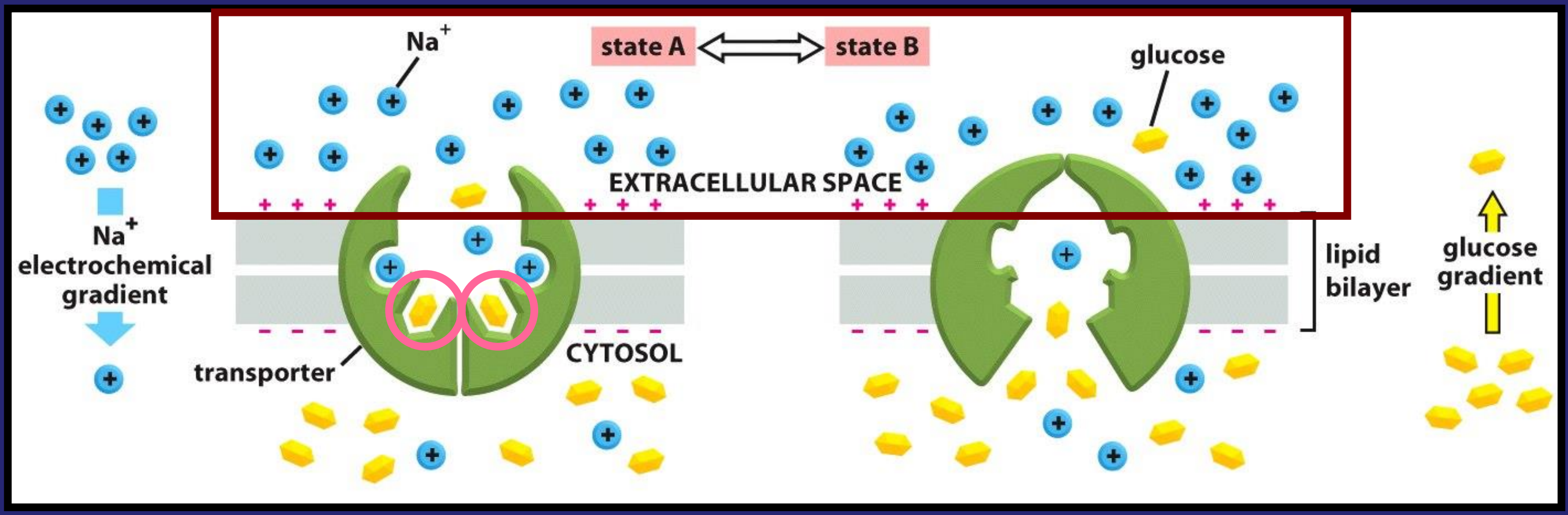
Since Na^+ concentration is much higher in the extracellular space …..



….. glucose is more likely to bind to the transporter in the A state

Glucose transporter can be driven by a Na^+ gradient

Therefore, both Na^+ and glc enter the cell (via A→B) much more often than they leave it (via B→A)



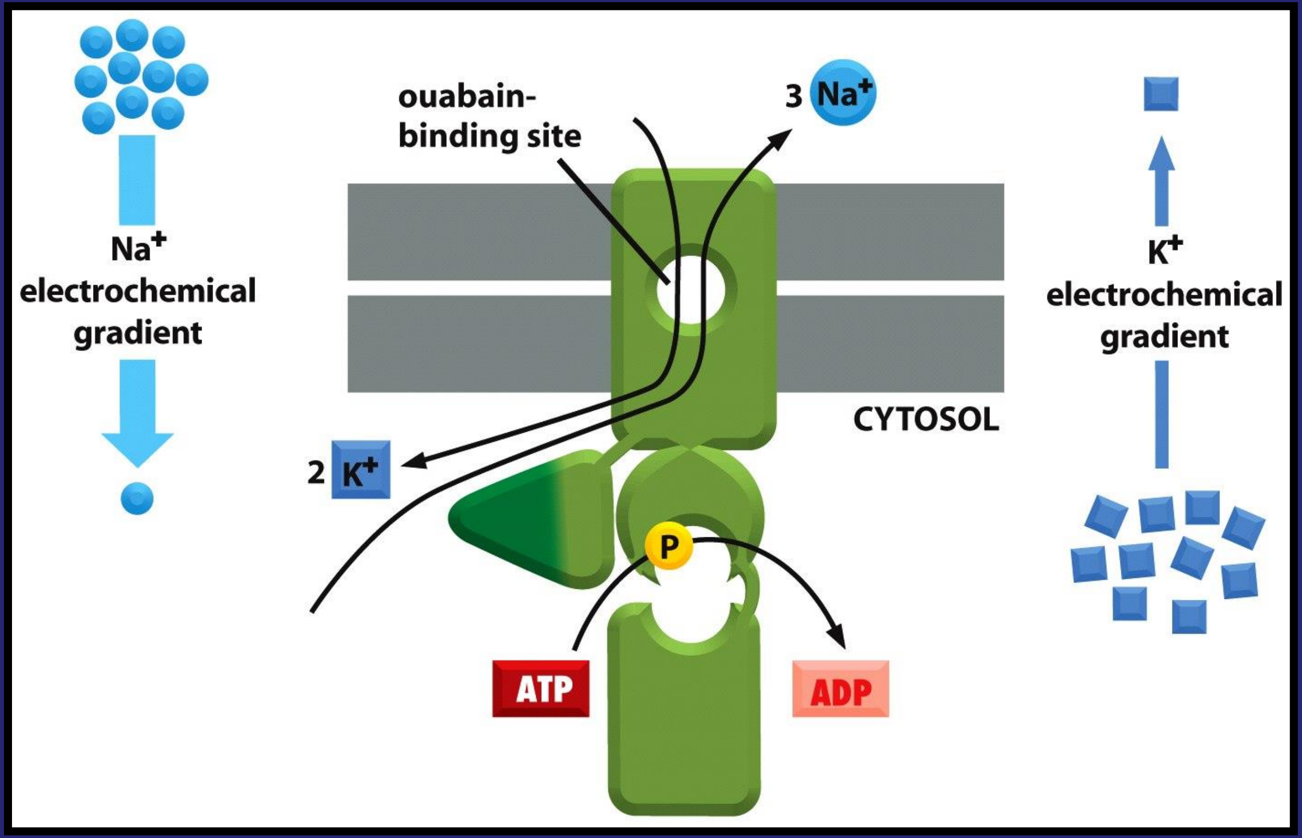
The overall result is the net transport of both Na^+ and glc into the cell

Glucose transporter can be driven by a Na^+ gradient

Because the binding is cooperative, if one of the two solutes is missing, the other fails to bind to the transporter

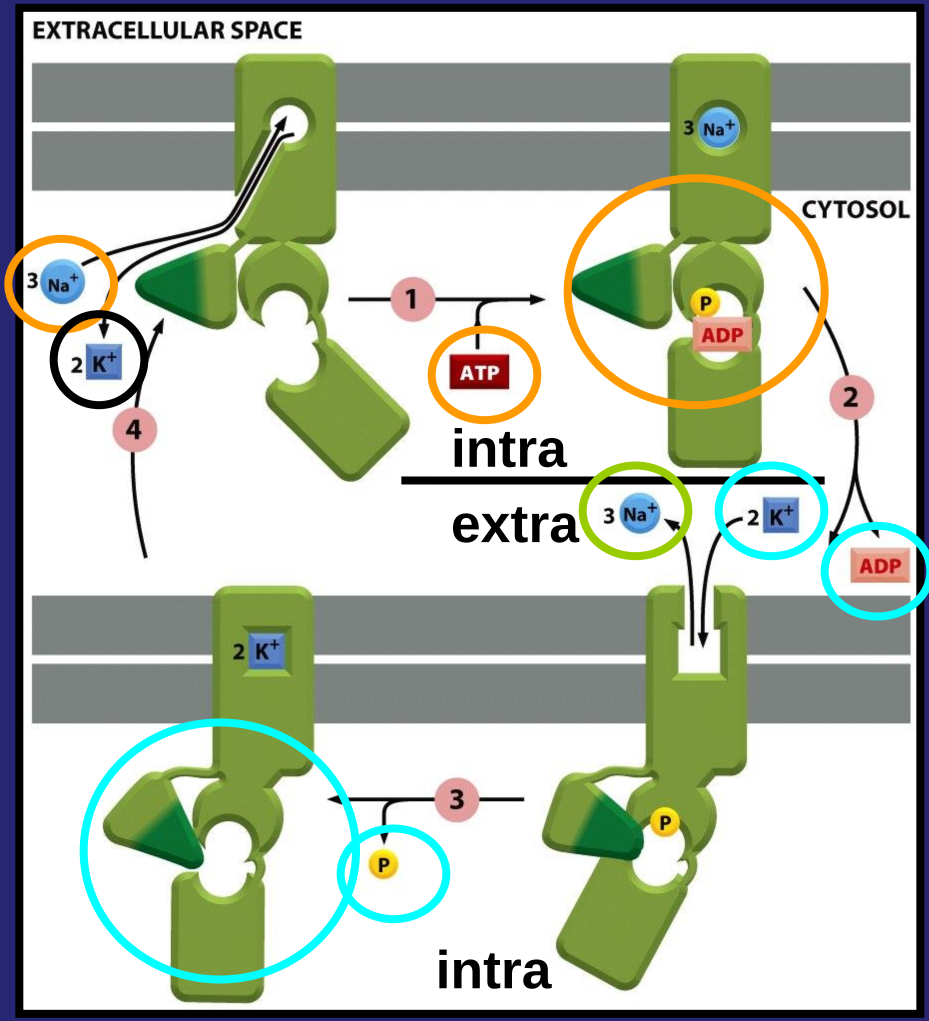
Thus, the transporter undergoes a conformational switch between the two states only if both solutes or neither are bound

ATP-driven pumps



This transporter actively pumps Na^+ out of and K^+ into a cell against their electrochemical gradients

ATP-driven pumps



1) Binding intracellular Na^+ and phosphorylation by ATP induce conformational change

2) Transfer Na^+ across the membrane out

3) Binding of K^+ and dephosphorylation return the protein to original stage

4) Transfer of K^+ into cytosol

Three classes of ATP-driven pumps

1) P-type pumps

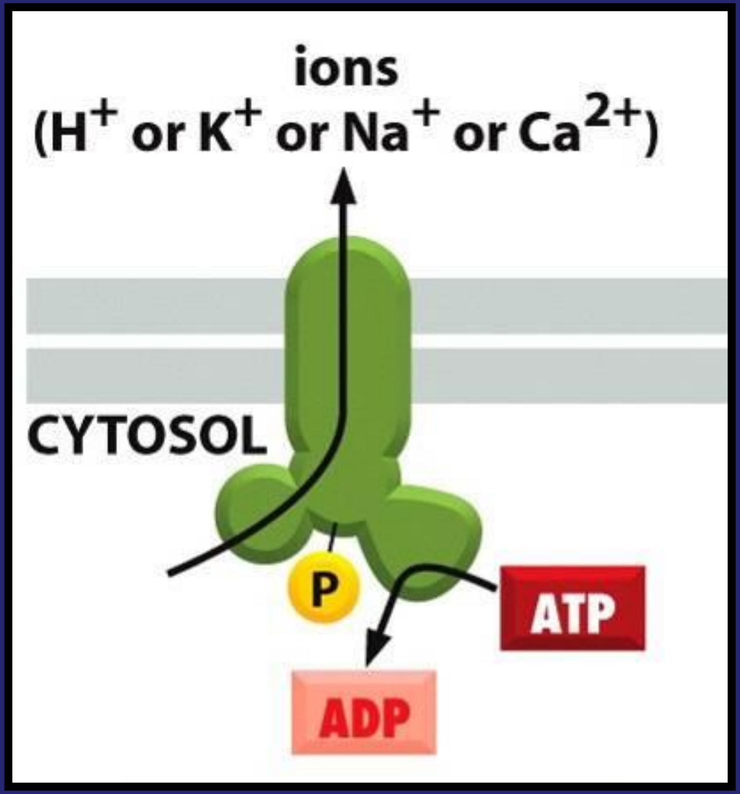
2) F-type pumps

3) ABC transporters

- the source of energy for the active transport is hydrolysis of ATP
- ATP-driven pumps can transport through membrane
  - ions
  - small molecules

P-type pumps

Structurally and functionally related multipass transmembrane proteins

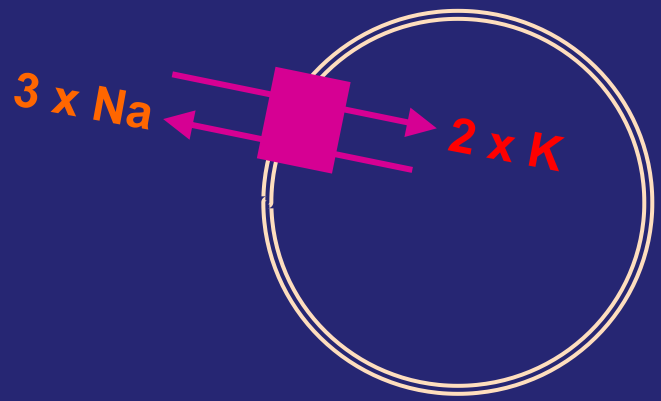


- „P-type“, because phosphorylate themselves during the pumping cycle

- This class include many of the ion pumps that are responsible for setting up and maintaining gradients of Na^+, K^+, H^+, and Ca^{2+} across cell membrane

Na-K ATPase pump

Important for membrane potential and osmotic balance



Ca^{2+} ATPase pump

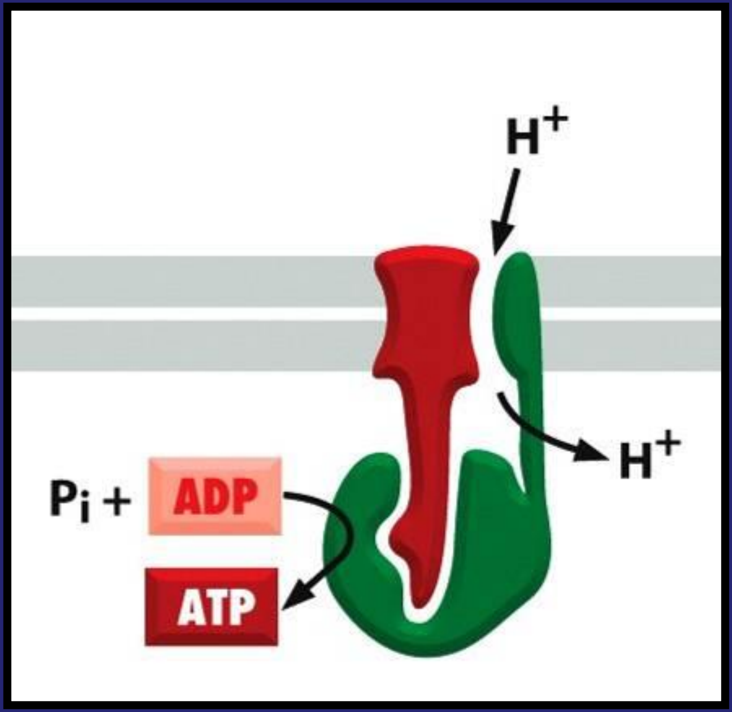
Strict regulating of Ca^{2+} concentration in cell

Growing concentration of Ca^{2+} ions is a trigger a lot of biological processes (mediator secretion to synaptic aperture, contraction of muscles, secretion, etc.)

The effects of calcium ions are indirect and are mediated by binding to Ca^{2+} dependent protein (for example calmodulin which is activated by proteinkinase C, CaM kinase, etc.)

F-type pumps

Turbine-like proteins constructed from multiple different subunits



- **Are found in the plasma membrane of bacteria, the inner membrane of mitochondria, and the thylakoid membrane of chloroplasts**
- **Called ATP synthases**

- **Structurally related are V-type ATPases that normally pump H^+ rather than synthesise ATP**
- **They pump H^+ to organelles to acidify their interior**

F-type pumps

To acidify organelles interior?

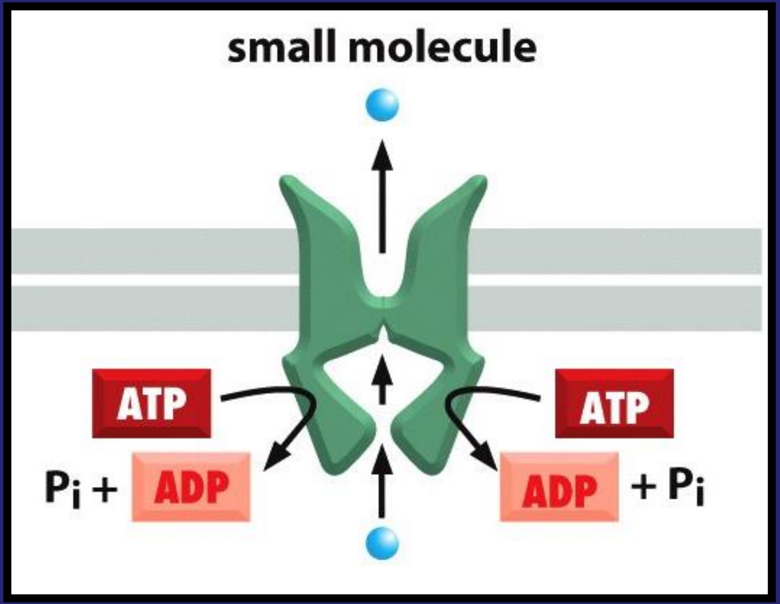
WHY?

F-type pumps

Acidify interior of lysosomes, synaptic vesicles, and plant vacuoles

ABC transporters

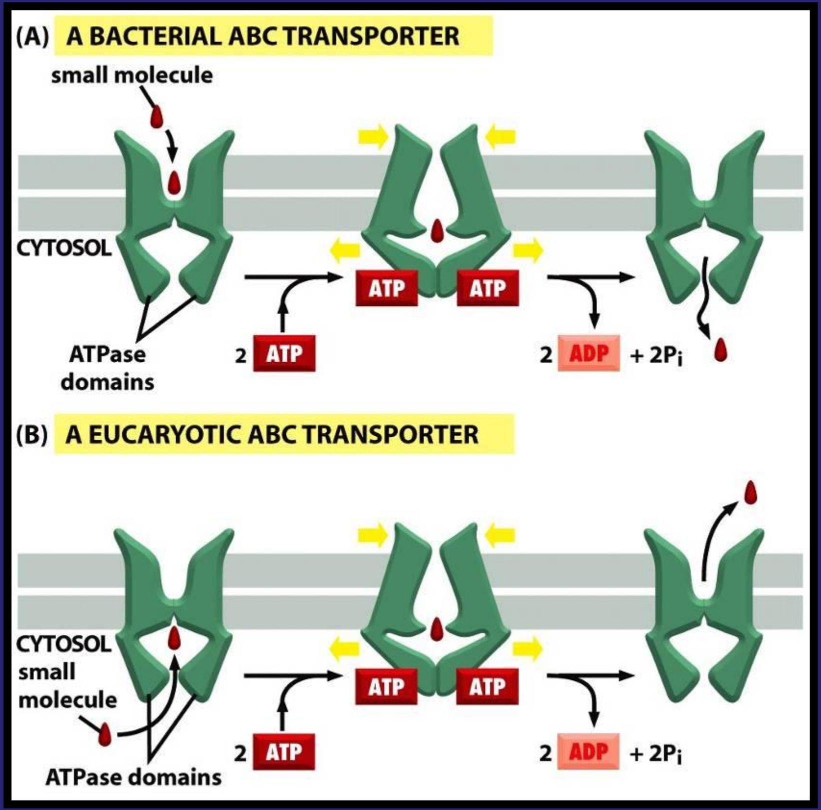
Primarily pump small molecules across cell membranes, in contrast to P-type and F- or V-type ATPases, which exclusively transport ions



- ABC transporters are also on ER membrane where help to transport molecules from cytosol to ER lumen
- Includes also MDR proteins (multidrug resistance) or CFTR (cystic fibrosis transmembrane regulator) proteins

ABC in prokaryotes and eukaryotes

Multiple domains, two hydrophobic (six membrane-spanning segments) which provide substrate specificity and two ATPase domains protruding into cytosol



- **Prokaryotic are used for export and import**
- **Eukaryotic only for export**
- **Eukaryotic are able to pump hydrophobic drugs out of the cytosole (resistency to anticancer drugs)**

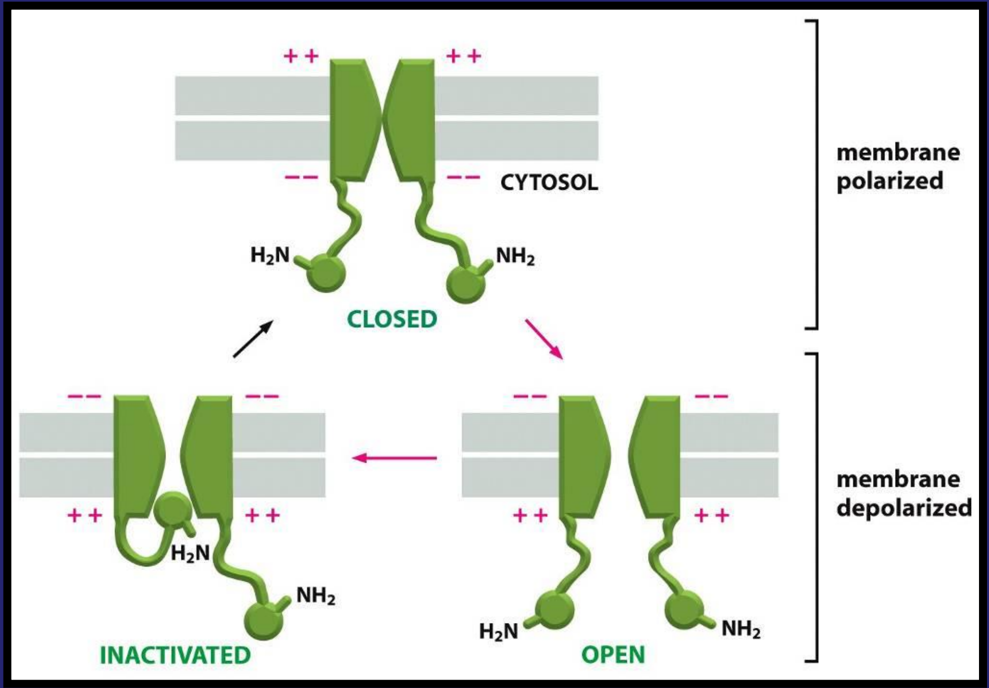
2. Principles of Membrane Transport

Ion channels and the electrical properties of membranes

Ion channels

Ions transport through membrane has crucial importance in biology. It helps to keep internal ion composition, which is different from extracellular space.

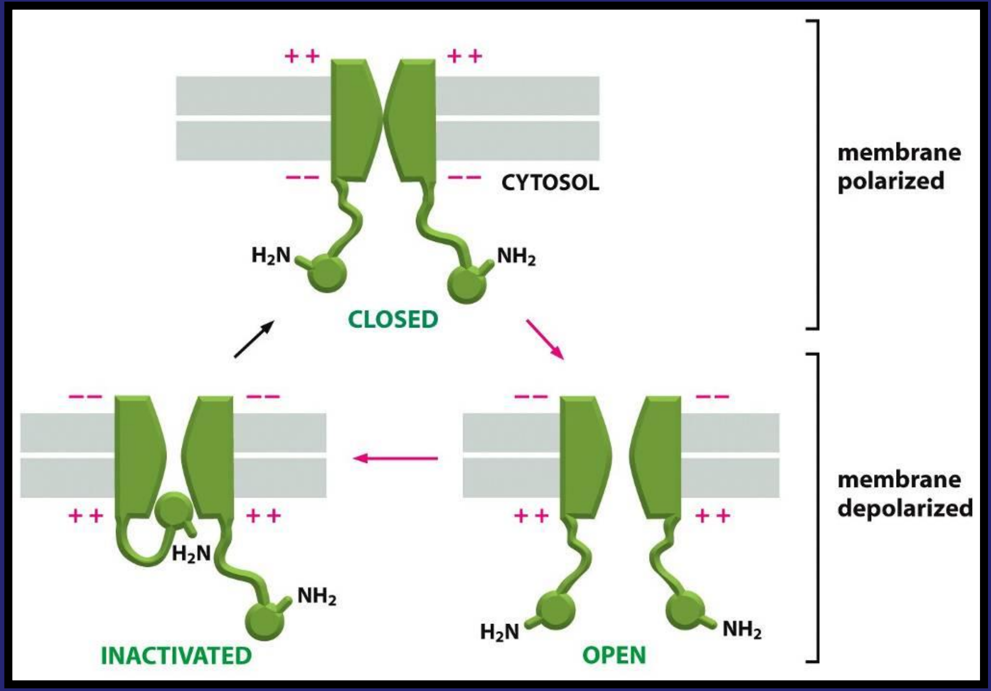
Ion channels are closable. They have two or three conformations (*closed – open, inactivated*).



Ion channels

When the membrane is at rest (highly polarized), the closed conformation has the lowest free energy and is therefore most stable

When the membrane is depolarized, the energy of the open conformation is lower, so the channel has a high probability of opening

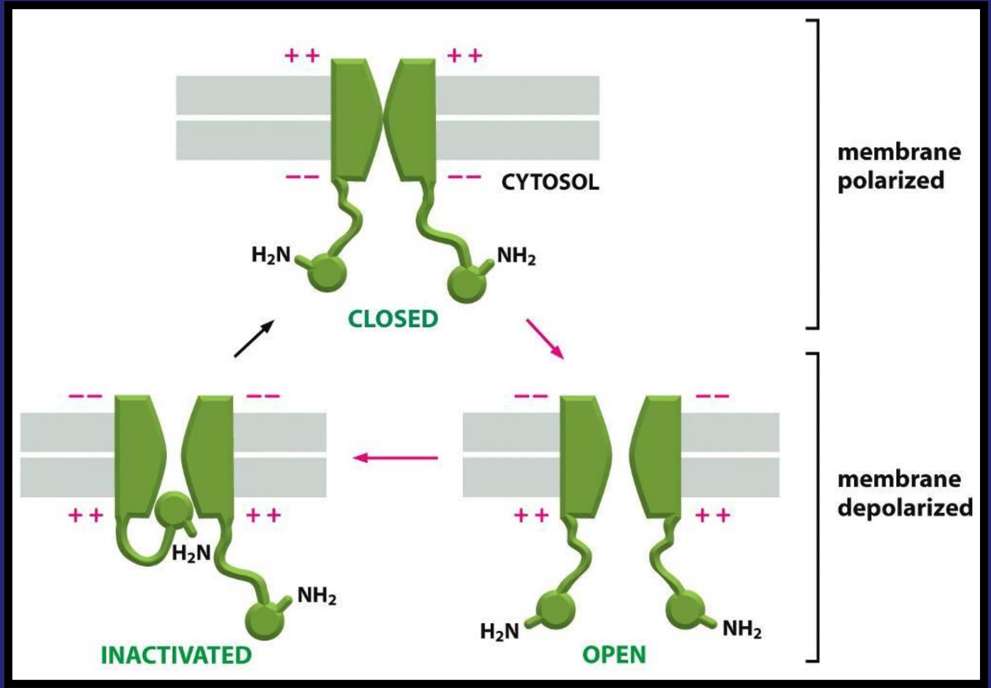


Ion channels

However in the depolarized membrane the inactivated conformation is the most probably state

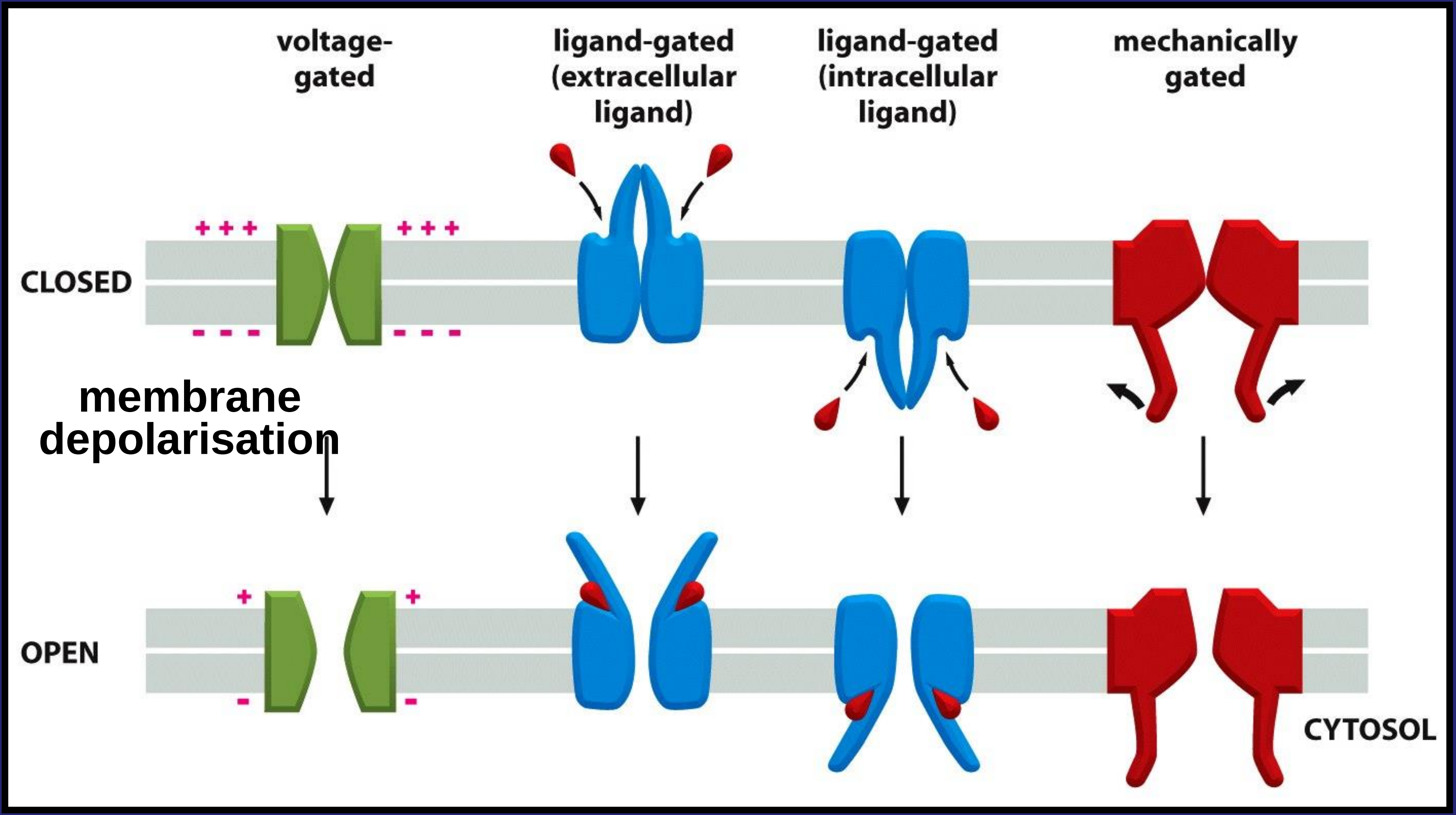
So after a variable period spent in the open state the channel becomes inactivated

The red arrows = sequences that follows a sudden depolarization



The black arrow = return to the original conformation as the lowest energy state

The types of ion channels



Voltage-gated ion channels

This type of the ion channels play the main role in the tissues which are excitable (neurons, muscle and heart cells)

They are <u>regulated by membrane potential</u>

The membrane potential is directed by permeability of membrane for specific ions

Voltage-dated ion channels are Na^+ channels, K^+ channels, Ca^{2+} channels of N type (Neuron) and T type (Transient)

Ligand-gated ion channels

The channel opening is regulated by binding of some ligand molecule into receptor (extra or intracellular)

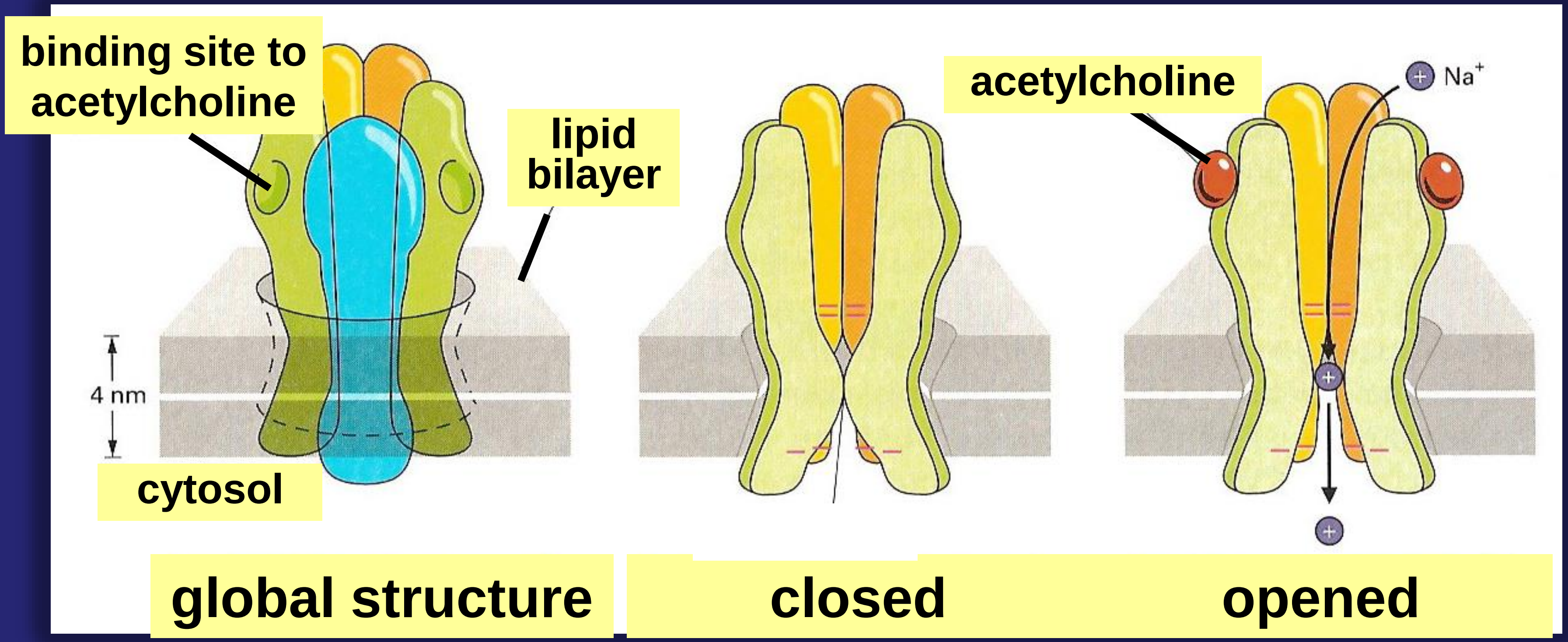
This channels mediate very quick responses to stimuli = in milliseconds

They are usually part of membrane receptors

- ***Acetylcholine-nicotine receptor*** ***connected to*** Na^+/K^+ ***or*** Ca^{2+} ***channel (neuro-muscle plate or vegetative ganglia)***
- ***Glutamate receptor*** ***connected to*** Na^+/K^+ ***or*** Ca^{2+} ***channel***
- ***GABAA –receptor*** ***and*** ***Glycine receptor*** ***connected to*** Cl^- ***channel***
- ***Muscarine receptor*** ***M2 connected to*** K^+ ***channel***
- ***Serotonin receptor*** ***– part of cations channel***

Na^+ channel with acetycholine

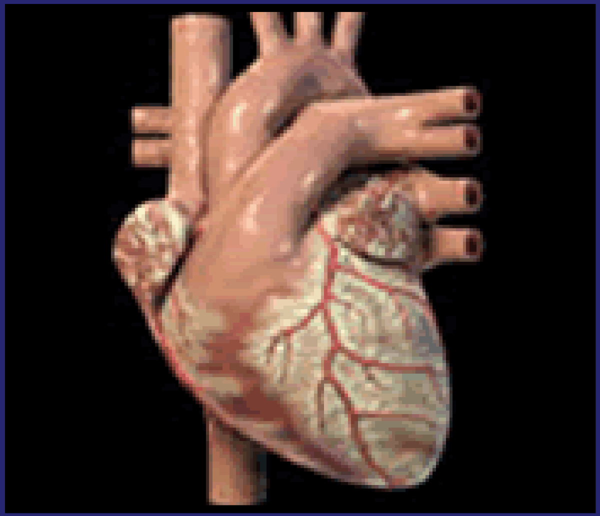
The channel opening is regulated by binding of acetylcholine molecule into receptor (extra or intracellular)

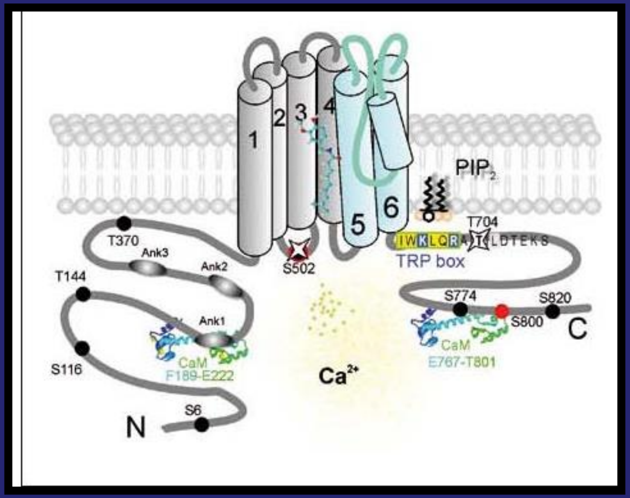


Voltage and ligand-gated

They are open by membrane depolarisation, but probability of opening depends on ligand binding

Examples are so named slow channels for Na^+ (type L) and Ca^{2+} (type L) in the cell of myocard





Mechanically-gated ion channels

Channel opening is regulated by mechanical force, which affects the channel

Mechanical connection between the channel and membrane is mediated by <u>microfibril</u>, which open the channel only than the membrane is taut

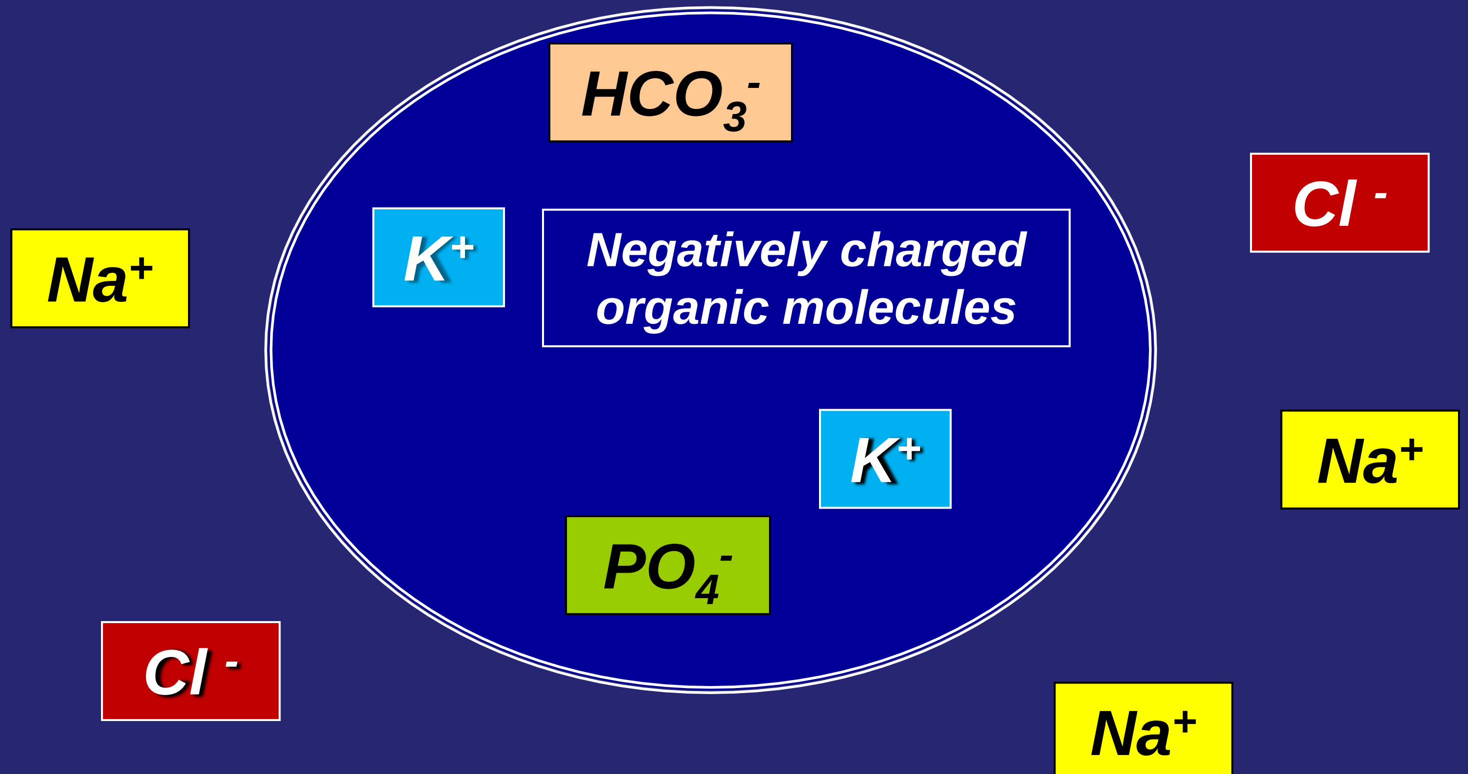
Example: K^+ channels in the <u>vestibular apparatus</u>

The vibration of sound mechanically open the K^+ channels in hair cells of internal ear which generate <u>neuronal excitement</u>

The electricity of membrane

The plasma membrane of all electrically excitable cells contains voltage-gated cation channels

They are responsible for generating the action potential



Ions concentration

Comparison of ions concentration inside and outside a typical mammalian cell

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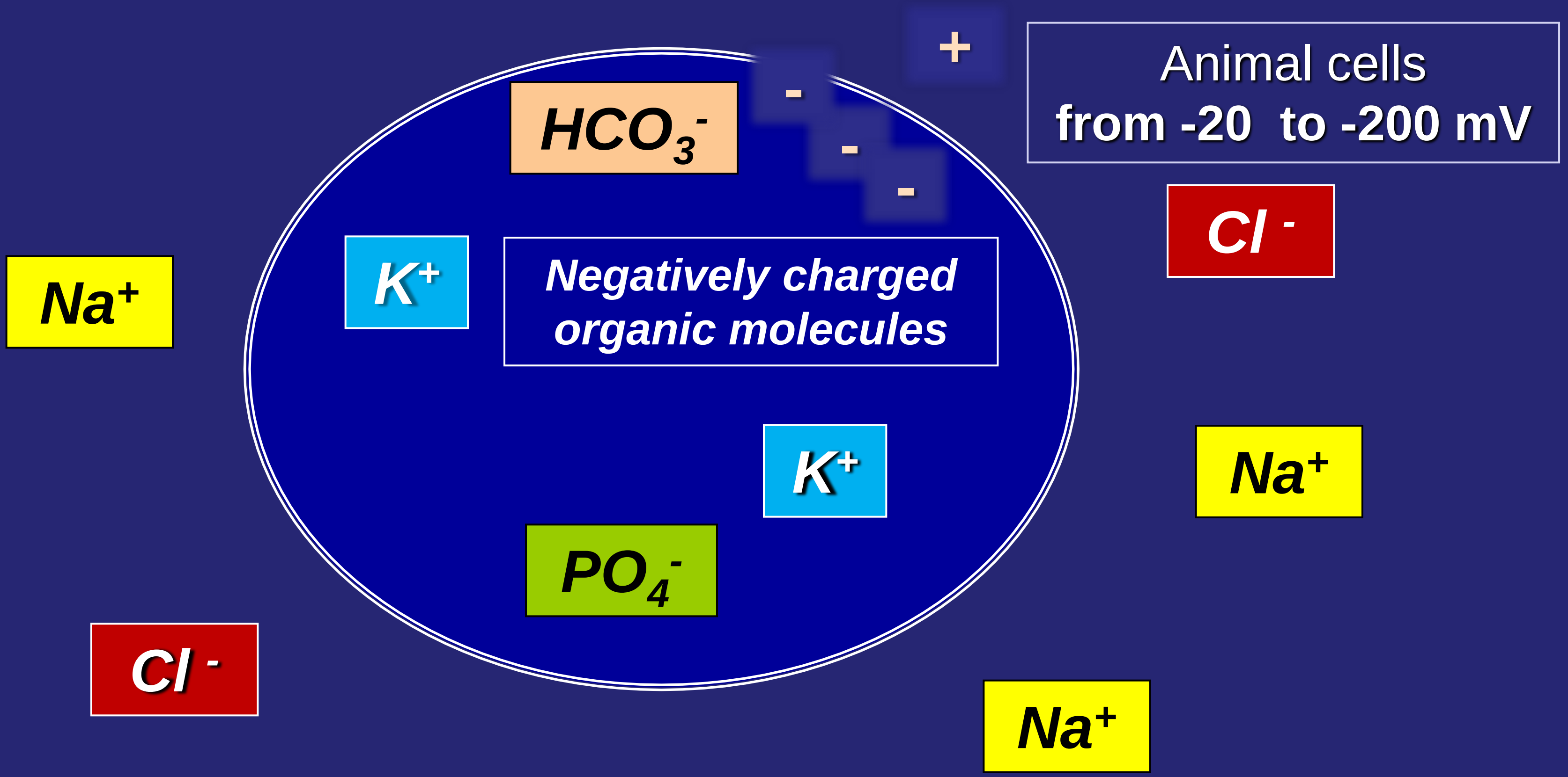
Anions*		
Cl^-	5–15	110

Polarization of membrane

A difference between electrical potential on external and internal side of membrane exist = the membrane potential. It is NEGATIVE



THE MEMBRANE IS POLARIZED

Description of membrane polarization

Inside the cell there are in the high concentration of negative ions of organic molecules

This negative charge is balanced by K^+ ions that are the most important ions in the cell

- Cell actively pump K^+ inside (against gradient of concentration)
- K^+ ions escape by channels out of the cell

- The ions imbalance is produced by this process
- The negative charge stay inside, which the cell is not able to equilibrate
- Steady state is set up = RESTING MEMBRANE POTENTIAL

This potential protect another K^+ movement out of the cell (electrochemical gradient is ZERO)

Two forces

The force which drives ion through the membrane has two parts

- **electrical potential the membrane**
- **concentration gradient of this ion**

resting membrane potential varies in mammalian cells from –50 to –90mV according to the cell type. It has negative charge because intracellular space is negative in regard to extracellular environment (negative ions are in the cell in excess to positive ions)

Action potential

Any change in membrane permeability for ions (especially cations) activates change of ion potential and can disrupt the resting membrane potential and cause action potential

This is used in electrical signalisation of cells in excitable tissues (neuronal, muscle or heart cells)

Activation of action potential – 1/2

1. Sudden supply of positive ions to the cell → change of resting membrane potential → **MEMBRANE DEPOLARIZATION**

2. Sufficiently large depolarization leads to **opening of voltage-gated Na^+ channels**

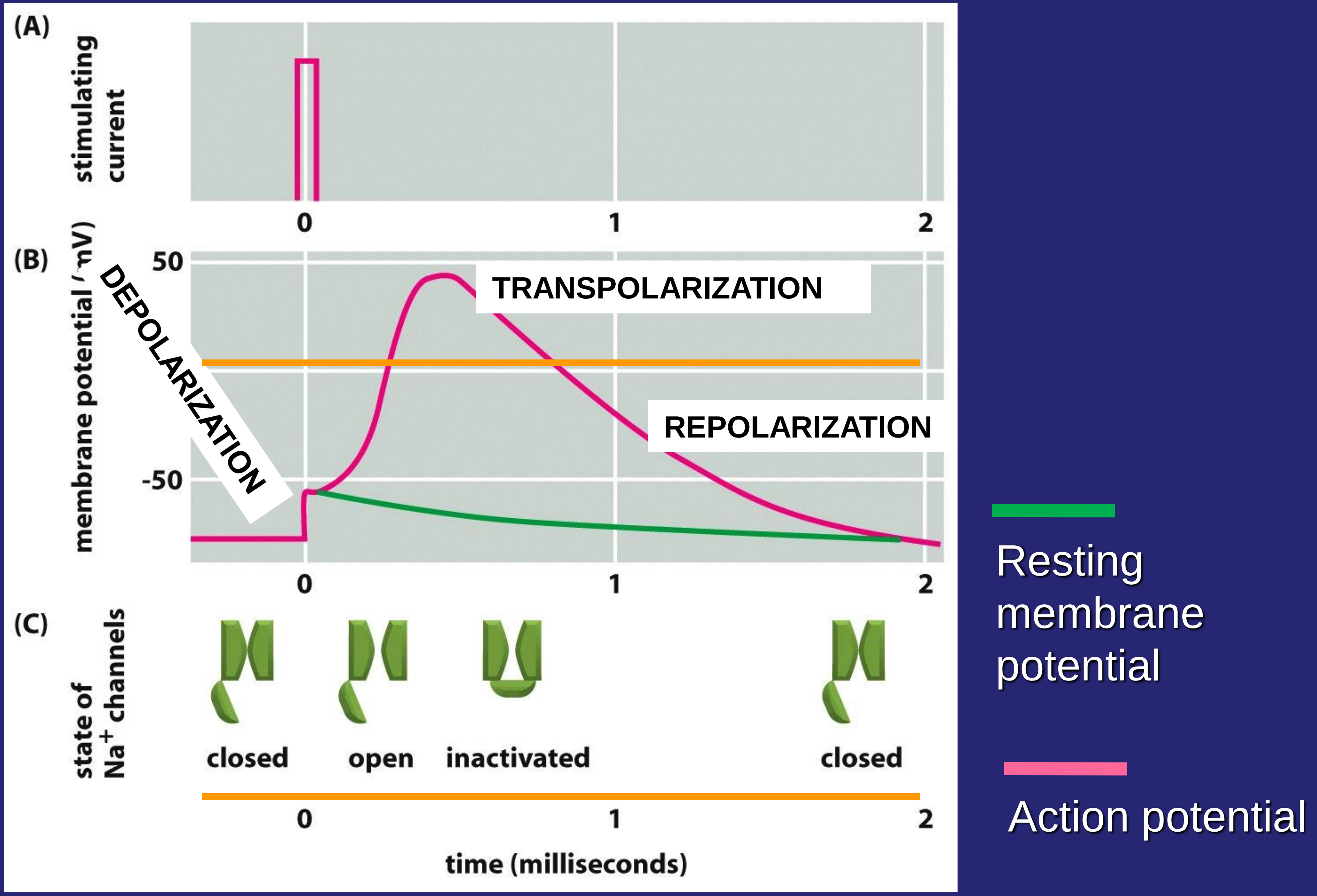
Local depolarization of plasma membrane started the **ACTION POTENTIAL**

Activation of action potential – 2/2

3. Na$^+$ ions enter very quickly to the cell, the depolarisation is intensify, and polarity is convert to +30 up to +40 mV = **MEMBRANE TRANSPOLARIZATION.** In this moment the ion gradient for Na$^+$ is equilibrated. No more ions enter to the cell.

4. After it the leaving K$^+$ channels are open and Na/K pump returns the ions ration to initial stage (Na$^+$ ionts out, K$^+$ ionts in). **MEMBRANE REPOLARISATION** is coming (return to resting membrane potential)

Activation of action potential



Cell communication by electric signalisation – 1/2

Action potential is able to *depolarise also neighbouring parts of the membrane and extend itself as a wave (tight junctions)*

Gradually more and more Na^+ channels are opened and whole process is accelerated

During several miliseconds the resting membrane potential changes from –60mV to +40mV and again back

Cell communication by electric signalisation – 2/2

The action potentials contribute for quick cell communication on long distance

They administer in neurons during neuronal signals transportation, during muscle cells contractions, including myocardium

The electric activity of the cells is usually mediated by voltage-gated Na^{+} channels (in myocardium Ca^{2+} ions are used)