Biomembranes, transport through membranes

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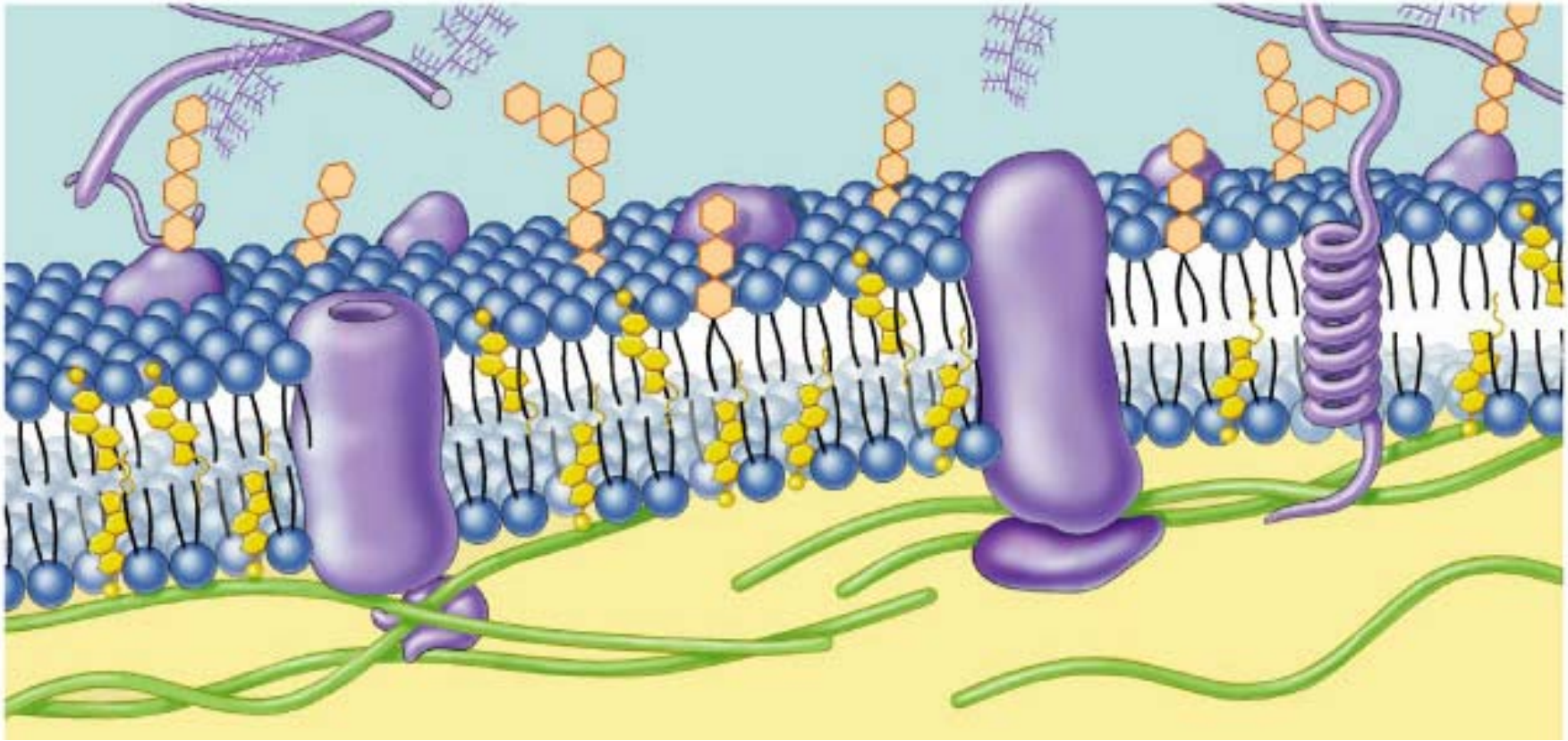
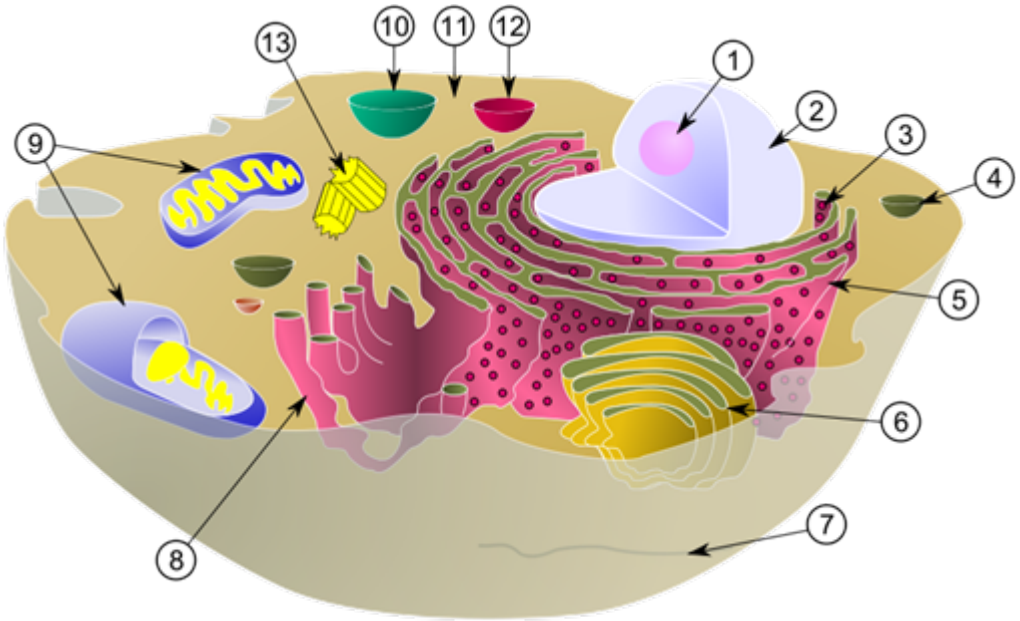
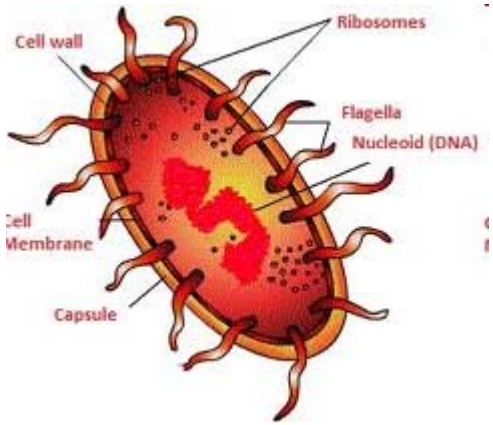


Cell membrane and cell organelle membranes - compartments





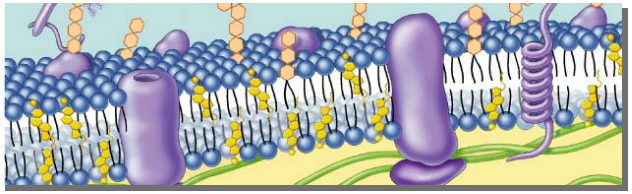
1-2. nucleolus- nucleus
3-5. ribosome, rER
4. vesicle
6. Golgi apparatus
(7. Cytoskeleton under the cell membrane)
8. sER
9. mitokondrions
10. vesicle
11. cytoplasm
12. lysosome
(13. centriolum)

- 80% of the dry mass of eukorytic cell is biomembrane
- selective advantage towards prokaryotes during evolution

--- **importance of compartmentalisation**

- same thickness: 6-10 nm
  (except: nucleus memb., mitochondrion m.)
- same content: *„unit membrane”*
  - lipids (40-60 %)
  - proteins (60-40 %)
  - carbohydrates (2-10 %) and water (!)

Functions of biomembranes (A)



- **semipermeable membrane** (near impermeable)
  - separation membrane
    (permeability: e.g. SCFA, steroid hormones)
  - **selective (regulated) transport (transporters)**

- **Regulated információáramlás/communication**
  - **metabolic effects:**
    receptors of soluble ligands /cell membrane/
    (hormones, GFs, neurotransmitters – metabotropic receptors)
    membrane proteins in signaling pathways (e.g. G-proteins)
  - **irritability:**
    (ionotropic neurotransmitter receptors; ionchannels, ionpumps)
  - **adhesion receptors** /cell membrane- cytoskeleton/
    (cell-cell adhesion, cell-ECM adhesion)
  - **mediation of antigenicity („selfidentity”)**/cell membrane/

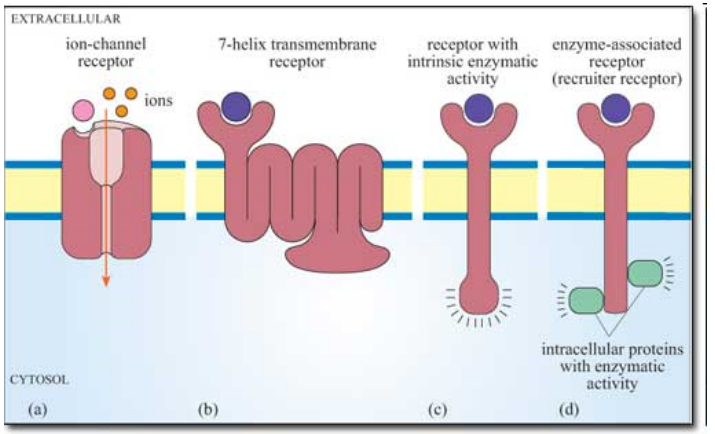
- **Enzyme function**
  - metabolic enzymes (e.g. sER – some steps of cholesterol synthesis)
  - role in signals (e.g. PLA2, PLC – cell membrane)
  - producing energy (mitochondrion)
  - special cells: NADPH oxidase (respiratory burst) … … ...

Functions of biomembranes (B)
endo- and exocytosis, phagocytosis

- Endo-, exocytosis; phagocytosis, pinocytosis
- Receptor mediated endocytosis (e.g. LDL-R; coated vesicle, clathrin)
- Cell polarity, cell shape, cell motility: chemotaxis, cell division, cell fusion

Function of cell membrane proteins

- transporters
- enzymes
- receptors
  (ionotropic, 7-TM, Tyr-kinase, enzyme-associated R)
- antigen
- cell-cell adhesion
- cytoskeleton binding



Membrane lipids:

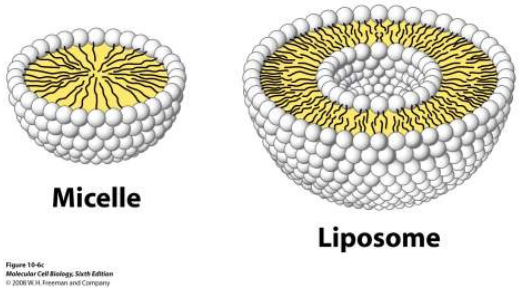
1. Double layer of the phospholipids:

Types of phosphoglycerides: *phosphatidylcholine*, *-ethanolamine*, *-serine*, *-inositol* (and cardiolipin)

Polar groups (water coat): phosphate, N-containing group or carbonhydrate (inositol), /glycerol/

Phospholipid double layer or micelles - liposomes

- kialakulás vizes közegben: spontanous, free energy on the minimum level
- **self-closing layers** (hole is energetically energetikailag disadvantagous)
- **dense layers**: displace water from the apolar lipid environment
- **mobile, non rigid system**
- weak binding forces:
  - „hydrophobic interaction”
  - electrostatic interactions
  - H bridges
  - van der Waals forces
- importance of micelles:
  - e.g. fat digestion: bile acidic micelles
  - taking drogs



2. Sphingolipids

backbone: sphingosine, ceramide -> sphingomyelin; A cerebroside, A ganglioside (glycolipids)

3. Cholesterol

- polar 3-OH group: attaches to the polar group of phospholipids
- apolar ring and chain: attaches to the FA sidechain of phopholipids
- in the membrane there is non-esterified cholesterol

(the uptake of cholesterol from membrane: HDL!)

Lipid-anchored membrane proteins

Types of covalent binding (anchoring):

A/ **fatty acid acyl group** – N-terminal Gly
(e.g. /C14 saturated/ miristilation: Ras (p21)

B/ **prenilation:**
attachment of farnesyl or geranyl-
to C-terminal or Cys
(e.g. Ras, Rab – small G proteins)

C/ **GPI- anchoring:**
through **membrane glycosyl-phosphatidylinositol, carbohydrates, and phosphoethanolamine**
to C-terminal
(e.g. acetylcholinesterase)

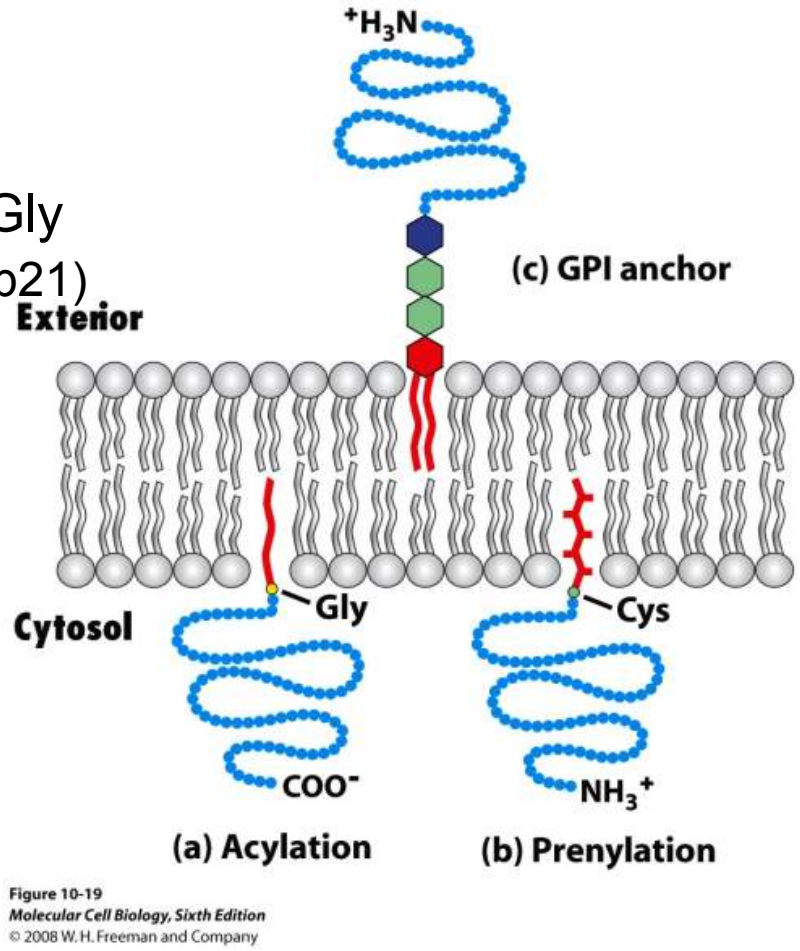
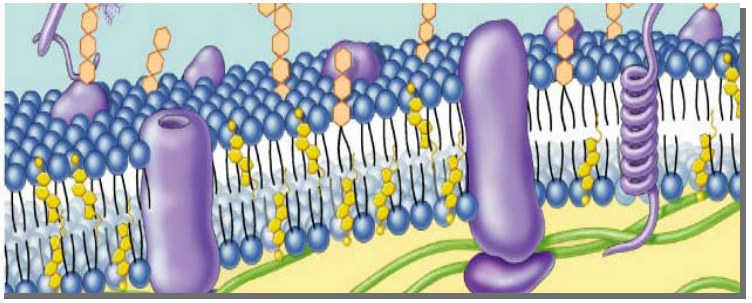


Figure 10-19
Molecular Cell Biology, Sixth Edition

Structure of biomembranes: mosaic nature

(fluid mosaic model)



- **dinamic mosaic structure of protein and lipid**

- localisation of proteins:

  - **integrant (intrinsic) membrane proteins (mainly glycoproteins)**
  - transmembrane region/domain: apolar, globular middle
    α-helical structure (25 amino acid is enough) or multiple β-turns (hydrophobic amino acids: Ala, Val, Leu, Ile)
  - inside the membrane interaction with the fatty acid chains
  - they can be extracted only with potent erős handling (e.g. detergents, organic solvents, which ruin the membrane)

  - **periferial (extrinsic) membrane proteins**
  - water-soluble
  - attach to integrant membrane proteins or lipids with electrostatic interactions

  - **lipid-anchored periferial membrane proteins**
  - covalent binding to membrane lipids

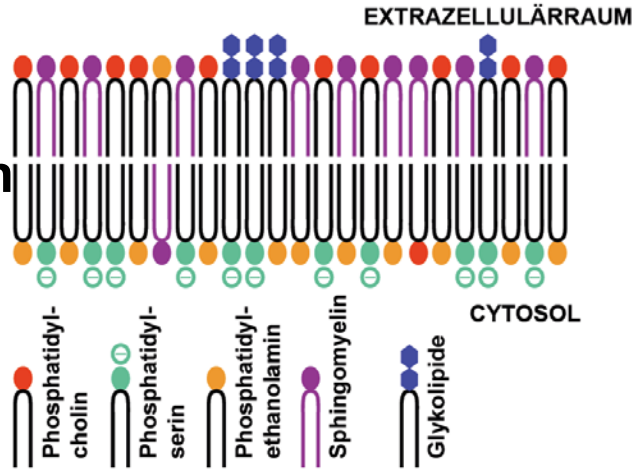
Structure of biomembranes: membrane asymmetry

➢ the compound of the two layer are not identical

- different periferial membrane proteins, different bonding
- different integrant membrane protein domains
- different density of glycosylation (cell membrane, ER, Golgi)
- different lipidcompound

➢ **phospholipid asymmetry**

- outer layer:
phosphatidylcholine, sphingomylein
(+ charge)
- inner layer:
phosphatidylethanolamine,
-serine, -inositol

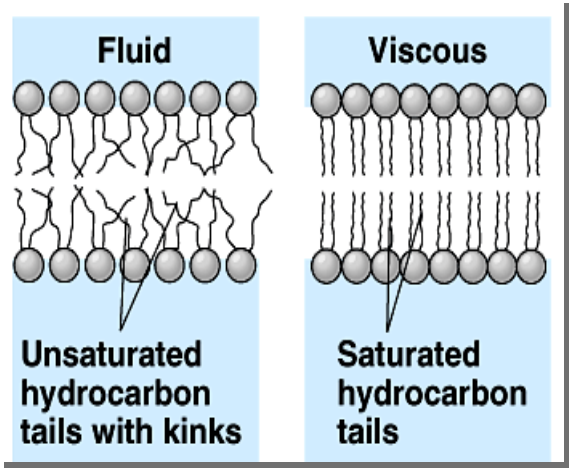


Asymmetry change:

❖ platelet activation
❖ apoptosis (phosphatidylserine on the outer surface)

Structure of biomembranes: fluid nature

- fluidity on body temperature: like olive oil
- fluidity rate influances the function of membrane proteins
- **influental factors:**
  - length of fatty acid chain (length: viscosity ↑)
  - rate of saturated fatty acids (viscosity ↑)
    /van der Waals interactions – „more dense" membrane/
  - rate of unsaturation, trans-cis configuration (trans: viscosity ↑)
    /cold-adaptation: more unsaturated fatty acid/
  - incidence rate of cholesterol (body temperature: viscosity ↑)
    /hinders sterically the move of fatty acid chains,
    interaction with them/
    /characteristic of eukaryotes
  - increase of temperature:
    slow phase transition
    viscous „gel"---- „fluid"
    /increased fatty acid chain motility/

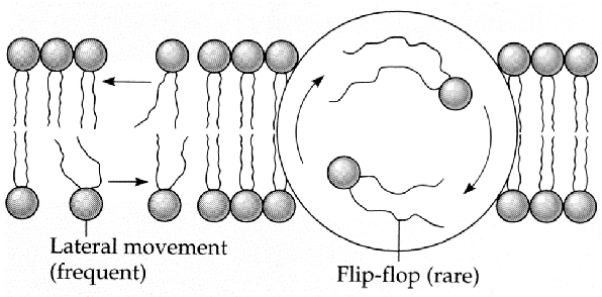


Dinamics of biomembranes:

1. lipid movements in the membrane

Phospholipids

- dislocation of fatty acid sidechains (vibration)
- rotation (around an axis)
- lateral diffusion (in the plane of the membrane)
  /mean: 2μ/s/
- flip-flop (from the one layer of the membrane to the other)
  /this is energetically not beneficial/
  /one lecithin molecule only 1x in more hours!
  /10^9x slower, than lateral diffusion/



2. protein movements in the membrane

- rotation (around an axis)
- lateral diffusion – big difference between proteins
  e.g. very mobile: rhodopsin (DHA!)
  meanly mobile: adhesion receptors
  (capping, clustering: group of receptors – with the help of actin-cytoskeleton)

Microdomains in the membrane

special lipidenvironment around proteins

-**„bulk lipid”** – ordinary membrane lipid compound

vs

- **„annular lipid”** – the integrant membrane proteins surrounding,

relatively permanent „lipidring”

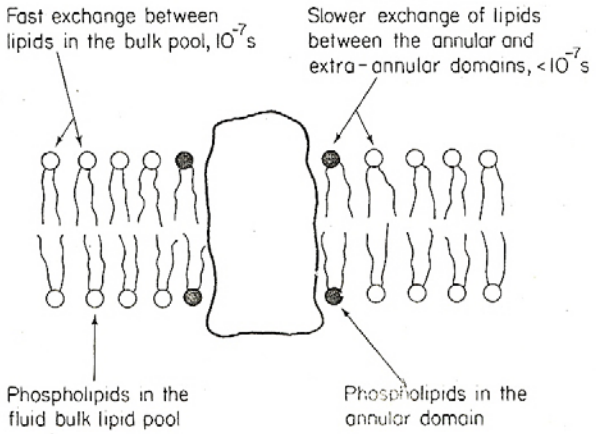


Figure 3.1 Annular and bulk lipid domains. The immediate ring of lipids that provide the interface between the protein and the bulk lipid pool has been termed the annular lipid domain. By virtue of the interaction of annular lipid with the protein, the rate of exchange of annular lipid with lipid in the bulk lipid pool is rather slower than the exchange between adjacent lipids within the bulk lipid pool

Microdomains in the membrane: lipidrafts

- **lipid components: cholesterol and sphingomyelin**
- **they contain many type of cell membrane receptors, signaling proteins → signaling complexes**
- **proof: hystochemical marking for tracking localization**

Membrane carbohydrates: glycocalyx

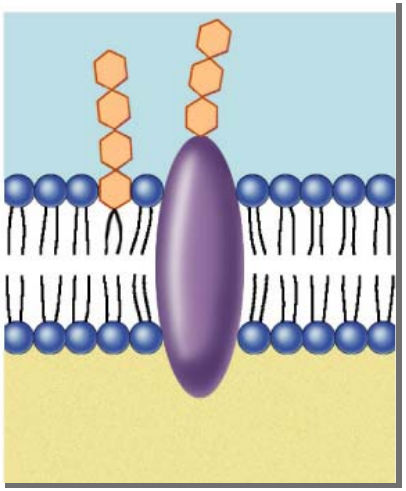
e.g. ABO blood group

- Essential role in antigenity

/basis of the function of immune system

/changes on the surface of malignant tumor cells and dead/apoptotic cells!

- Essential role in cell adhesion and receptor function
- Essential role in embriogenesis



- carriers of oligosaccharides:
  - glycoproteins
    (O- and N-glycosides bind)
  - glycolipids

The oligosaccharide-sequence can be very specific.

Membrane transport: channels and carriers

Passive transport

Passive transport is a transport through membranes which

-requires no energy

-molecules are transported down cc. gradient

Passive transport depends on:

- Concentration gradient
- Lipophilicity of the molecules
- Size of the molecules
- Charge of the molecules

Simple passive diffusion through biological membranes:

- water
- Small lipid-soluble substances
- gases
- cholesterol, fatty acids

Selective permeability: integrant membrane proteins allows selectivity of substances transported through the membrane

Channel proteins allow getting through by constituting a polar inner surface

Carrier proteins bind specific molucules and help their getting through the membrane

With th help of carrier proteins:

Facilitated diffusion transport of substances from the low concentration to the low concentration area

-specific

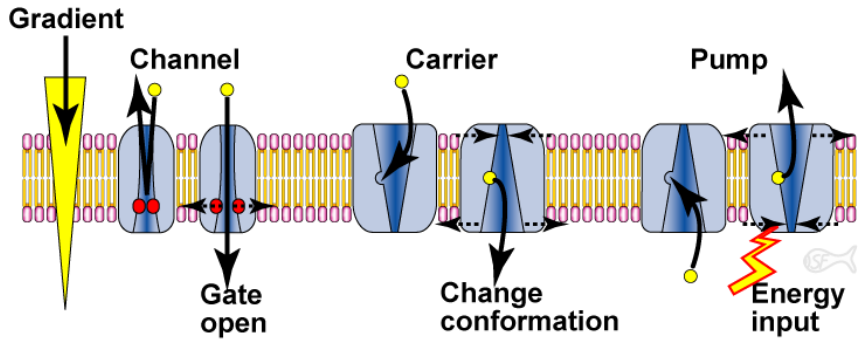
-passive

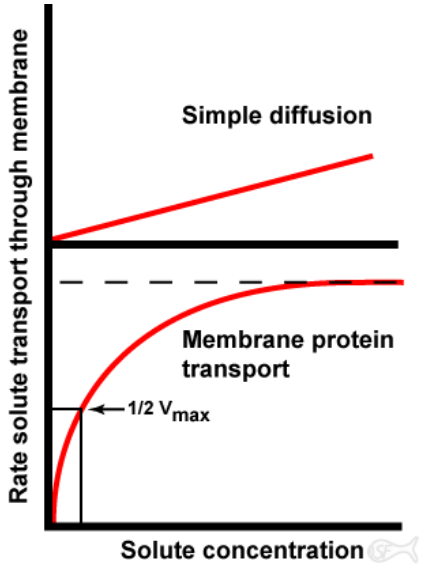
-saturable, if all the channel proteins bind ligand

Membrane protein transporter types

Channels: promote diffusion through a aqueous, polar pore, which induce change of the conformation and opens the channel

Carriers: one group allows facilitated diffusion according to cc. gradient, other group works like a pump and use energy and transports against cc. gradient





- **Simple diffusion**
  - **Limitated amount of molecules can be transported**
  - **slow, shows linear kinetics**
- **Membrane protein transport**
  - **not limitated**
  - **Specific for the transported molecule**
  - **fast, shows saturation kinetics**

Active transport

-requires energy – direct or indirect use of ATP

-molecules move against concentration gradient

-it requires carrier proteins to its action

Grouping of carrier proteins:

-uniporters – one molecule at once

-symporters – two molecules in one direction

-antiporters – two molecules in different directions

Secondary active transport

Co-transport

-uses the energy of an other transport for transporting a substance against concentration gradient

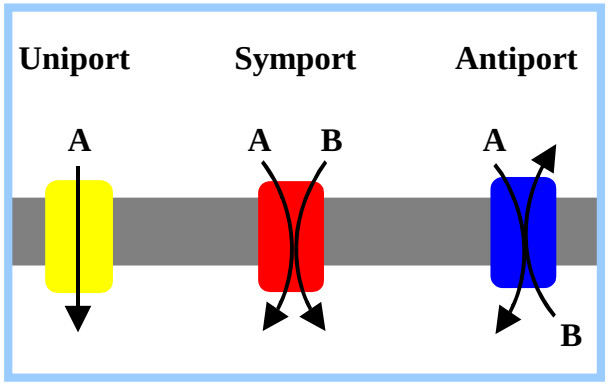
-symporter or antiporter mechanism

-glucose-Na^+ symporter (cholera!!)

-Na-H antiporter (intracellular pH regulation)

-Na-Ca antiporter

Classification of carrier proteins:



1. Uniport (facilitated diffusion) one way transport of one substance

e.g. **GLUT1** glucose transporter

Ionophor **valinomycin**

2. Symport (co-transport): the carrier binds two substrates at the same time and transports them to the other side of the membrane together

The transport of the two molecules is necessarily co-transport.

The transport of a molecule (ion) down gradient may allow the transport of an other molecule against gradient: **secondary active transport**.

E.g. ◆ **glucose-Na^+ symport**, in the cell membrane of epithel cells

◆ bacterial **lactose permease**, a **H^+ symport** carrier.

3. Antiport (exchange transport) transport two molecules in the opposite direction (e.g. Band3 – RBC Cl-/HCO3)

◆ Antiporters show "ping pong" kinetics

Substrate 1 is binded and transported

Substrate 2 is binded and transported in the opposite direction

Exchange transport may occur only

The conformation change may not occur without the binding of the substrate

Pumps

F-type H^+-ATPases

inner mitochondrial membrane

-The proton pumping electron transport chain uses redoxpotential for making pmf

-pmf drives H^+-flow through F-type ATPase → ATP is synthesized

P-type H^+-ATPases

mushroom PM H^+-ATPase

plant PM H^+-ATPase

Na^+ / K^+ ATPase (animal cells): pumps 3 Na^+ ions out & 2 K^+ ions in; Na^+ & K^+, regulate the cell membrane gradient

Ca^{2+}-ATPases (plant and animal PM and endomembranes: pump Ca^{2+} out of the cytosol (e.g. SERCA)

H^+ / K^+ changer ATPase (mammalian stomach mucusa layer): pumps H+ into the lumen of the stomach (pH=0,8)

Common features:

•Can be inhibited with orthovanadate ($H_2VO_4^-$)

•Domain structure identical (mainly ATP-binding site is conservated among pumps)

V-type H^+-ATPases (tonoplast, ER, Golgi, membrane of coated vesicles)

Function: acidification of the vacuolar space (circa to pH 5,5)

it energizes the membrane for carriers and the pH optimum of many vacuolar enzymes (proteases, glycosidases, phosphatases, nucleotidases) is in acidic range

Vacuolar proton pirophosphatase (H^+-PPase)

ABC-type pumps: ATP binding casette (pl. MDR1, CFTR)

Catalyse the transport of amphipathic molecules through vacuolar membranes

e.g. flavonoids, antocianinok, degradation secondary product of chlorophyll, xenobiotics (herbicides).

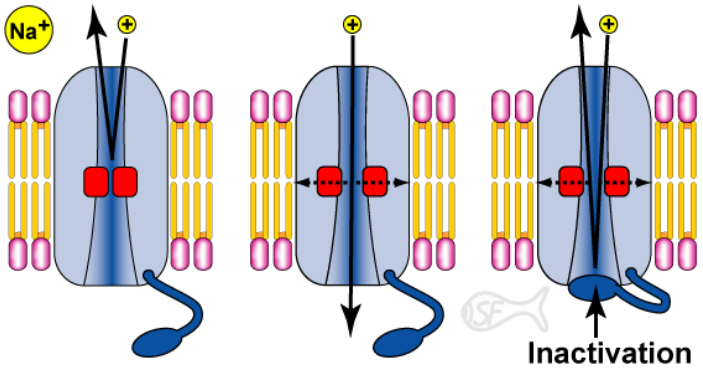
Ion channels

- They are selective for ions in different rate (specificity depends on the size and the charge of the ion)

1. Channels working with gate mechanism (opened and closed states alter)

- Ligand-gated: binding of the ligand to the receptor leads to change of conformation of the channel
- Voltage-gated: the voltage between the two sides of membrane
- mechanical: hair cells of inner ear
- others

Different ionchannels inactivates on different ways.



2. Leaky channels (always open): e.g. K+-channel, through this K+ leaves the cell

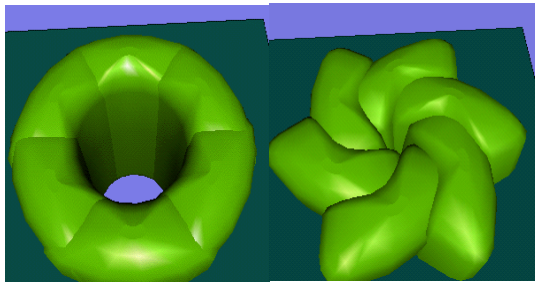
Porins

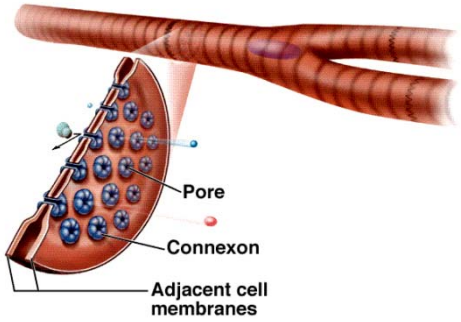
- Form pore in the membrane, through this specific molecules may move through the membrane with passive diffusion
- Gram– and Gram+ bacteria
  mitochondria
  chloroplast

Junctions

- Adhesion junction
- Tight junction: tight connection between cells, which inhibits passive diffusion between cells
- Gap junction: communication channel between cells

Structure of the gap junction





- embriogenesis-morphogenesis
- synchronization of heart contractions
- regulation of cell proliferation and differentiation (tumor supression)
- nutrition of avascular tissues (eye-lense)