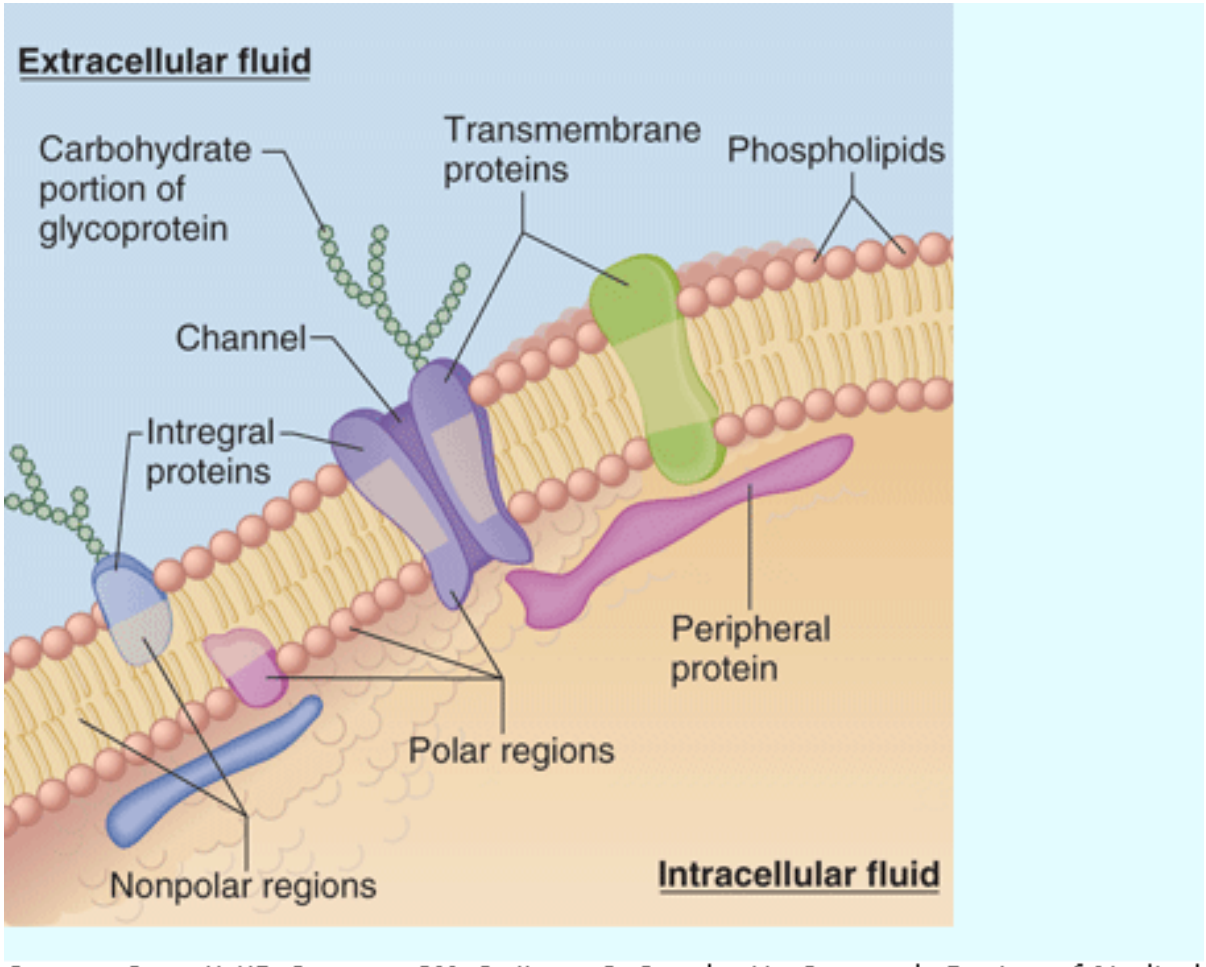
# CELL

Kenneth Alonso, MD, FACP

# Cell membrane



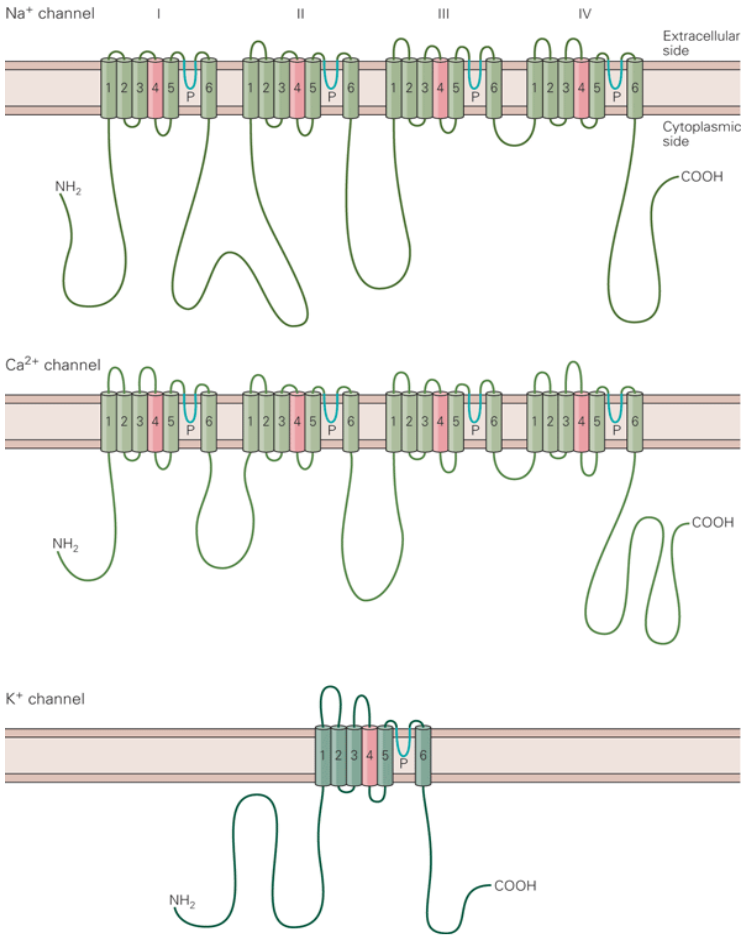

Source: Barrett KE, Barman SM, Boitano S, Brooks H: *Ganong's Review of Medical Physiology, 23rd Edition*: http://www.accessmedicine.com



Reproduced with permission from Widmaier EP, Raff H, Strang K: Vander's Human Physiology: The Mechanisms of Body Function, 11th ed. McGraw-Hill, 2008.)

Fig. 2-2 Accessed 07/01/2010

# Pore forming units






(Reproduced with permission from Kandel ER, Schwartz JH, Jessell TM [editors]: *Principles of Neural Science*, 4th ed. McGraw-Hill, 2000.)

The subunit of the $Na^+$ and $Ca^{2+}$ channels traverse the membrane 24 times in four repeats of six membrane-spanning units.

Each repeat has a "P" loop between membrane spans 5 and 6 that does not traverse the membrane.

<u>These P loops are thought to form the pore.</u>

Note that span 4 of each repeat is colored in red, representing its net "+" charge.

The $K^+$ channel has only a single repeat of the six spanning regions and P loop. Four $K^+$ subunits are assembled for a functional $K^+$ channel.

# Movement across the cell membrane

- The membrane is a bi-layer of phospholipid in which are interspersed large globular protein molecules that protrude through the service.
- Channels through the structures of the protein molecules serve as pores.
- Diffusion across the membrane occurs as molecules migrate from a region of high concentration to one of low concentration as a result of random motion.
- The diffusion of water is called “osmosis”.

# Movement across the cell membrane

- The rate of diffusion is related to:
- The difference in electrical potential and chemical concentration across the membrane
- The permeability of the membrane (lipid soluble substances favored)
- The surface area of the membrane.

# Movement across the cell membrane

- Diffusion is inversely related to the size of the solute.
- Large molecules such as glucose, water, and solvated ions pass through the membrane channels formed by glycoprotein molecules that extend through the membrane.

# Movement across the cell membrane

- Facilitated diffusion
- Glucose is transported from the lumen through the cell membrane through a symport by combining chemically with a carrier protein, the glucose transporter (glut), that penetrates through the membrane.
- $Na^+$ enters with the glucose.
- Glucose is then released into the cytosol.
- Glucose leaves the cell for the blood through a uniport.

# Movement across the cell membrane

- A $Na^+$-$K^+$-ATPase dependent pump at the blood interface maintains the cellular concentration of $Na^+$.
- ATP on cytoplasmic side.
- This “facilitated” diffusion is a form of secondary active transport.
- While the rate of diffusion increases proportionally with the concentration of the diffusing substance, a transport maximum is reached in facilitated diffusion.

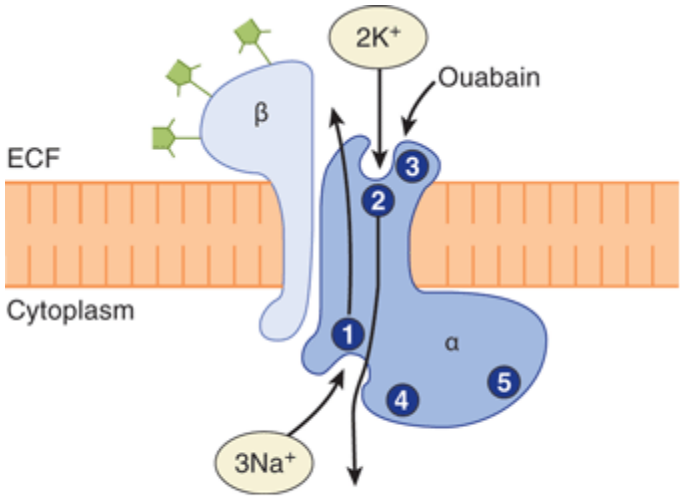
# Movement across the cell membrane

- There are four transporters in the blood brain barrier
- Acidic, basic, neutral, branched amino acids.
- Aquaporin-1 is a pore whose size only permits the entrance of hydronium ion (water)
- Responsive to antidiuretic hormone (ADH).

# Movement across the cell membrane

- Active transport differs from facilitated diffusion in that it can transport the substance even in the absence of or against an electrochemical gradient.
- High energy phosphate compounds provide the energy to drive that transport.
- The $Na^+$-$K^+$ transport pump causes the concentration of $Na^+$ inside the cell to become very low while simultaneously greatly increasing the intracellular $K^+$ concentration.
- Cardiac glycosides bind and inhibit $Na^+$-$K^+$-ATPase on the extracellular side of the pump.

# $Na^+$–$K^+$ ATPase




Source: Barrett KE, Barman SM, Boitano S, Brooks H: *Ganong's Review of Medical Physiology, 23rd Edition*: http://www.accessmedicine.com



The intracellular portion of the α subunit has:
(1) A $Na^+$-binding site
(4) A phosphorylation site
(5) An ATP-binding site.
The extracellular portion has:
(2) A $K^+$-binding site
(3) A ouabain-binding site.

(From Horisberger J-D et al: Structure–function relationship of Na–K-ATPase. Annu Rev Physiol 1991;53:565. Reproduced with permission from the *Annual Review of Physiology*, vol. 53. Copyright © 1991 by Annual Reviews) Fig. 2-18 Accessed 07/01/2010

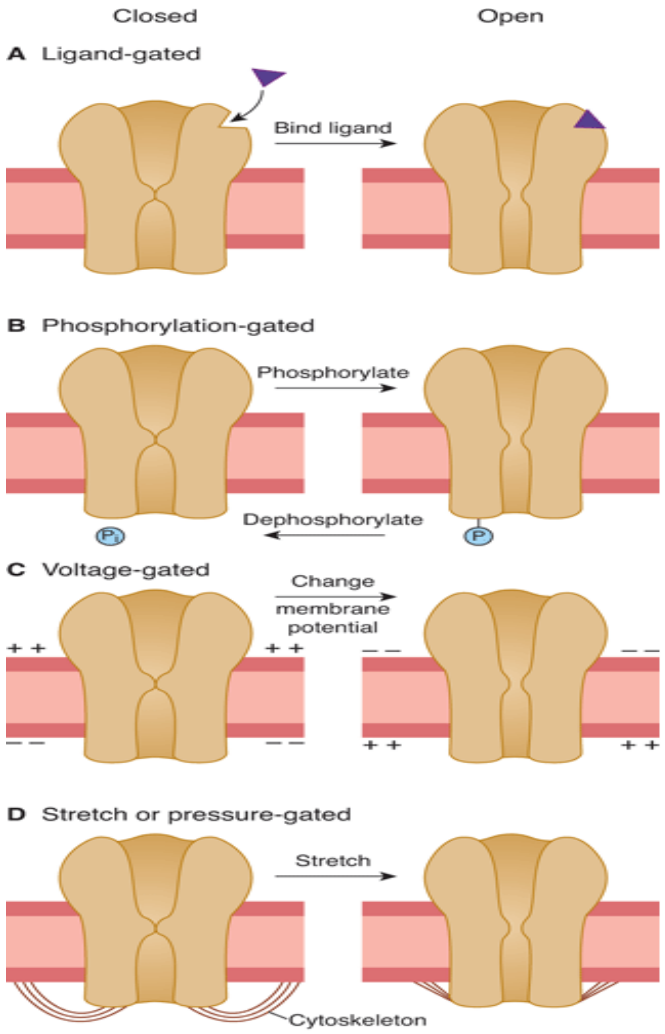
# Movement across the cell membrane

- Counter-transport is seen with $Na^{+}$-$Ca^{2+}$ and $Na^{+}$-$H^{+}$.
- Both utilize the same carrier protein.
- The $Ca^{2+}$ pump transports $Ca^{2+}$ outside of the cell membrane as well as into the organelles of the cell, maintaining a very low intracellular $Ca^{2+}$ concentration.
- It is a P type pump in that the transporter undergoes covalent phosphorylation during the transport cycle.
- It is ATP dependent.

# Movement across the cell membrane

- The $H^+$ F type pump is found in mitochondria where the pump couples oxidation with ATP generation.
- The $H^+$ V type pump is found in lysozymes where protons are introduced.
- It is an ATP dependent pump.
- The multiple drug resistant (MDR1) or ABC pump is also ATP dependent and is found in the cell membrane.

# Gates




Source: Barrett KE, Barman SM, Boitano S, Brooks H: *Ganong's Review of Medical Physiology, 23rd Edition*: http://www.accessmedicine.com



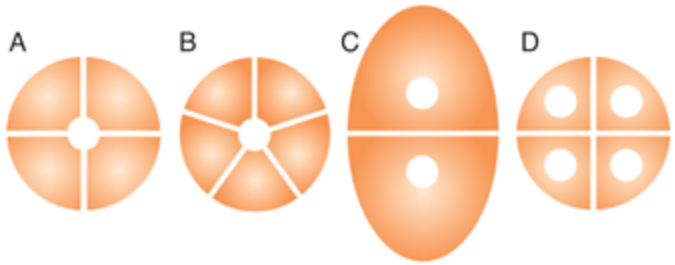
(Reproduced with permission from Kandel ER, Schwartz JH, Jessell TM [editors]: Principles of Neural Science, 4th ed. McGraw-Hill, 2000.)

Fig. 2-15 Accessed 07/01/2010

# Movement across the cell membrane

- Voltage gated ion channels.
- $Na^+$ is the most important one.
- Maintains polarization.
- $K^+$ and $Ca^{2+}$ channels are dependent upon potential generated when cell depolarized.
- Ligand gated ion channels.
- Receptor binding induces conformational change in ion channel.
- Nicotine receptor for acetylcholine as an example ($Na^+$-$K^+$).

# Ion channels






(Reproduced with permission from Jentsch TJ: Chloride channels are different. Nature 2002;415:276.) Fig. 2-16 Accessed 07/01/2010

Many $K^+$ channels are tetramers (A), with each protein subunit forming part of the channel.

In ligand-gated cation and anion channels (B) such as the acetylcholine receptor, five identical or very similar subunits form the channel.

$Cl^-$ channels from the ClC family are dimers (C), with an intracellular pore in each subunit.

Aquaporin water channels (D) are tetramers with an intracellular channel in each subunit.

# Endoplasmic reticulum

- The rough endoplasmic reticulum (RER) is continuous with the plasma membrane.
- It contains ribosomes and is the site of synthesis of secretory proteins and of N-linked oligosaccharide addition to many proteins.
- In neurons these are the Nissl bodies.

# Endoplasmic reticulum

- The smooth endoplasmic reticulum (SER) is continuous with the RER, but contains no ribosomes
- Involved in steroid production in the adrenals and gonads
- Involved in excitation-contraction mechanisms of muscle
- Involved in fat absorption in the intestine
- Involved in cholesterol and lipid metabolism and drug detoxification in the liver.
- The UV resistant associated gene protein (UVRAG) controls movement between the endoplasmic reticulum and the Golgi apparatus.

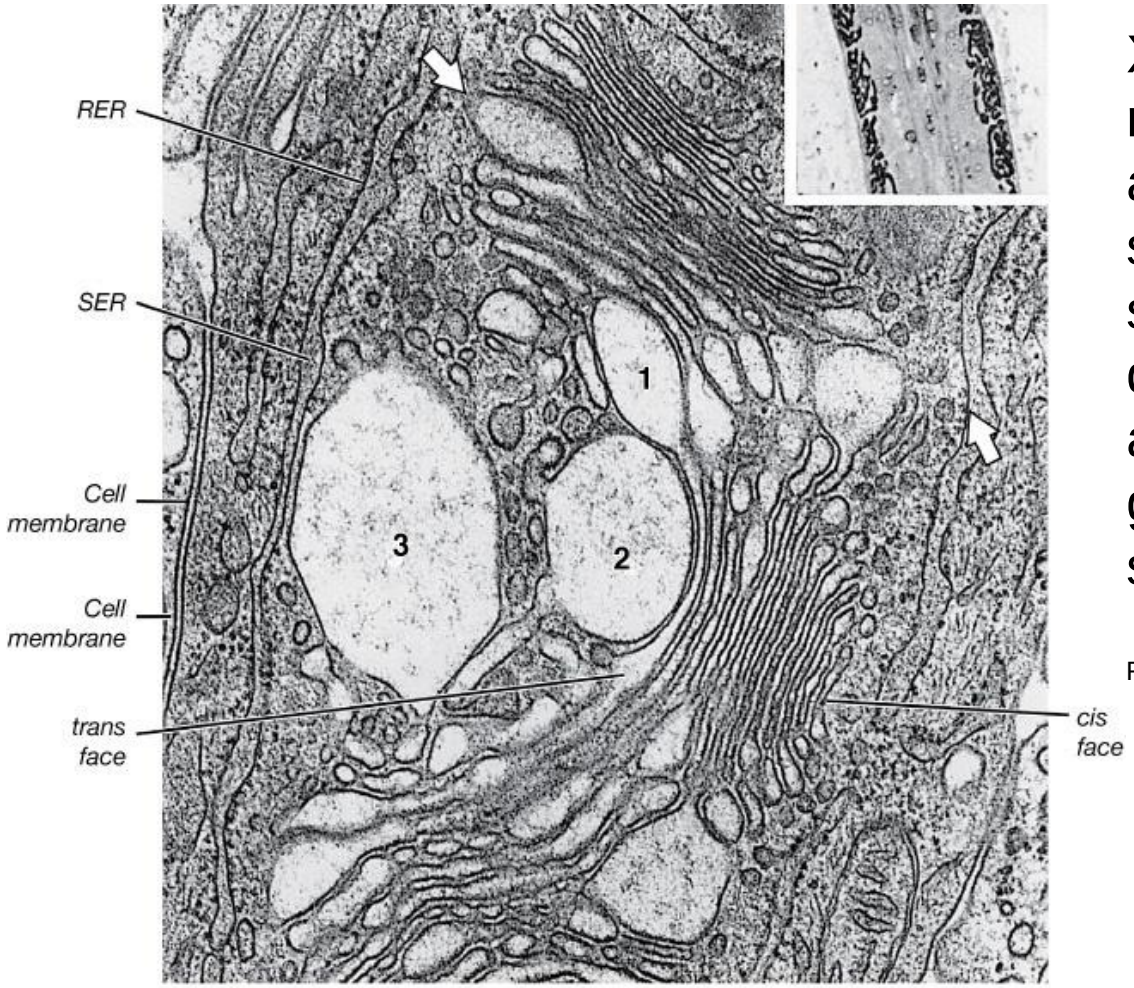
# Golgi apparatus

- The cis-Golgi network receives from the endoplasmic reticulum.
- COP I is the vesicular transport protein that moves vesicles from the smooth endoplasmic reticulum to the Golgi
- COP II moves vesicles from rough endoplasmic reticulum to the Golgi.

# Golgi apparatus

- In the Golgi stack (cis, medial, and trans cisterna), proteoglycans are prepared.
- Phosphorylation, sulfation, and later modification may also add targeting signals.
- In the trans-Golgi network, sorting and release follows.
- Clathrin moves vesicles from trans-Golgi
- Associated with receptor mediated endocytosis.

# Golgi apparatus




X30,000. Inset: a small region of a Golgi apparatus in a 1- m section impregnated with silver, which demonstrates the abundance of glycoproteins within some cisternae. X1200.

Fig. 12-21 Accessed 08/01/2010

# Organelles

- Many of the membrane-less organelles observed in cells are formed by liquid-liquid phase separation (LLPS) caused by interactions between proteins and nucleic acids.
- LLPS defines distinct compartments to efficiently organize cellular processes by concentrating certain factors in their proper place without interfering with one another in the complex and heterogeneous environment within a cell.
- Cajal bodies, P(rocessing) bodies, speckles, and stress granules as examples.

# Organelles

- These cellular bodies are dissolved during mitosis and reformed in the next round of the cell cycle
- They are also reversible, unlike aggregates, and appear to be in a viscoelastic-dynamic fluid state, which gives them plasticity and flexibility.
- A disordered region of proteins termed the intrinsically disordered region (IDR), or the low-complexity (LC) domain, facilitates assembly.

# Organelles

- RNA also serves as a seed in defining the location of the phase-separated compartment.
- The largest nuclear structure for ribosome biogenesis, the nucleolus, is formed near ribosomal RNA (rRNA) transcription sites.
- When rRNAs are artificially transcribed elsewhere in the chromosome, a new nucleolus-like condensate is formed at that site
- Nucleolar component assemblies at random nuclear positions are observed in inhibiting rRNA transcription or deletion of ribosomal DNA

# Organelles

- Stress granules are formed in the cytosol in order to suppress translation.
- Save limited resources and energy for essential functions
- Avoid increasing defective ribosomal products (RBPs) because of misfolding or premature termination
- Self-assembly of RBPs
- Parse multiple cellular systems
- NIR group of proteins as chaperones

# Organelles

- Abnormal aggregates associated with prion-like formations
- Prominent in Amyotrophic lateral sclerosis and Frontotemporal dementia

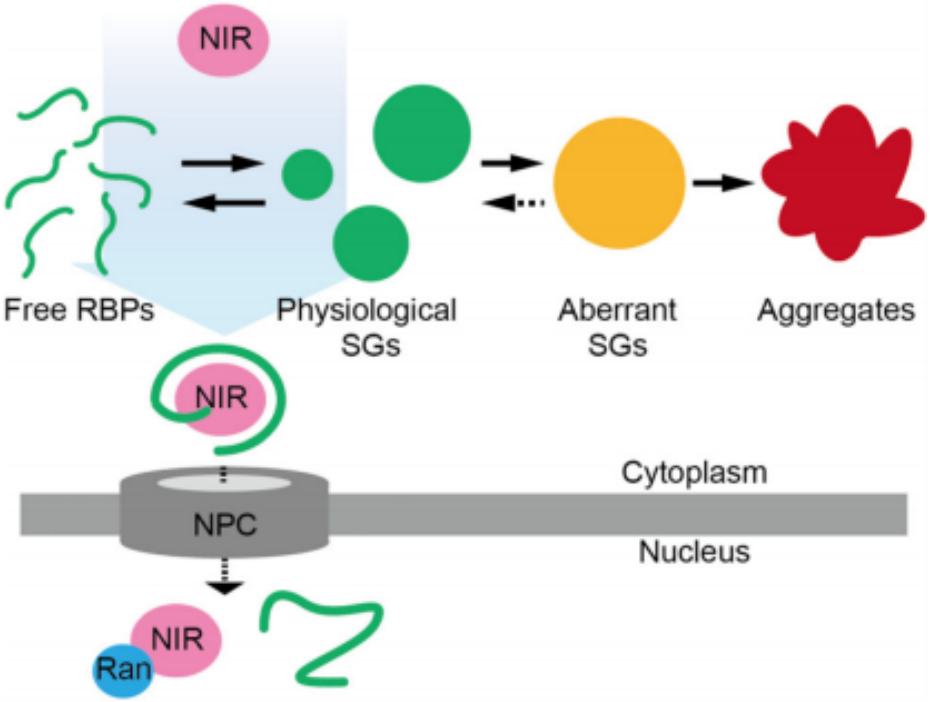



**Fig. 2** Model of chaperone function of NIRs for RBPs in cells. NIRs (pink) actively carry nascently translated proteins (green lines) or proteins in physiological SGs (green circles) into the nucleus, preventing LLPS from occurring in the cytoplasm. Small GTPase Ran (blue) displaces the proteins and binds NIR in the nucleus. In the absence of this regulatory system, physiological SGs could then subsequently transform into aberrant SGs (yellow) and irreversible aggregates (red) in the cytoplasm

Biophysical Reviews (2020) 12:519–539 https://doi.org/10.1007/s12551-020-00680-x

# Organelles

- The fundamental structural unit of chromatin is the nucleosome, which consists of negatively charged DNA containing a phosphate backbone wrapped around an octameric assembly of positively charged histone proteins
- Free cations promote self-association of chromatin
- LLPS is involved in organizing functionally distinct but physically adjacent chromatin domains in the nucleus.
- May provide chromatin with platform plasticity
- Phosphorylation involved in compaction and decompaction of heterochromatin

# Targeting signals

- Signal peptide sequence targets the endoplasmic reticulum membrane
- The amino terminal sequence lysine-aspartate-glutamine-leucine targets the luminal surface of the endoplasmic reticulum.
- The amino terminal sequence of a 20-80 residue protein targets the mitochondrial matrix.
- The amino terminal sequence proline-proline-lysine-lysine-alanine-lysine-valine (as an example) targets the nucleus.
- The amino terminal sequence serine-lysine-leucine (as an example) targets the peroxisome and matrix.
- Mannose-6-phosphate targets the lysosome.

# Microtubule

- A helical array of polymerized dimers of α- and β-tubulin.
- Each dimer has 2 GTP bound.
- Incorporated into centrioles, mitotic spindles.
- Grow slowly; collapse quickly.
- Incorporated into flagella.
- Cilia have a 9+2 arrangement of microtubles.
- ATPase links peripheral 9 couplets and leads to bending of cilium by differential sliding of doublets.
- Also involved in slow axoplasmic transport in neurons.

# Microtubule

- Molecular motor proteins transport cellular material toward opposite ends of microtuble tracks:
- Dynein, retrograde to microtubule
- Kinesin, antegrade.

# Microtubule abnormalities

- Chediak-Hagashi syndrome results from a microtubule polymerization defect.
- Phagocytosis is impaired.
- Peripheral neuropathy identified.
- Often seen with partial albinism.
- Kartagener's syndrome results from a defect in the dynein arm of the cilia.
- Sperm are immotile.
- Bronchiectasis and recurrent sinusitis common as bacteria not cleared. Associated with situs inversus.
- The azoles, taxols, vinca alakaloids, griseofulvin, and colchicine all act on microtubules.

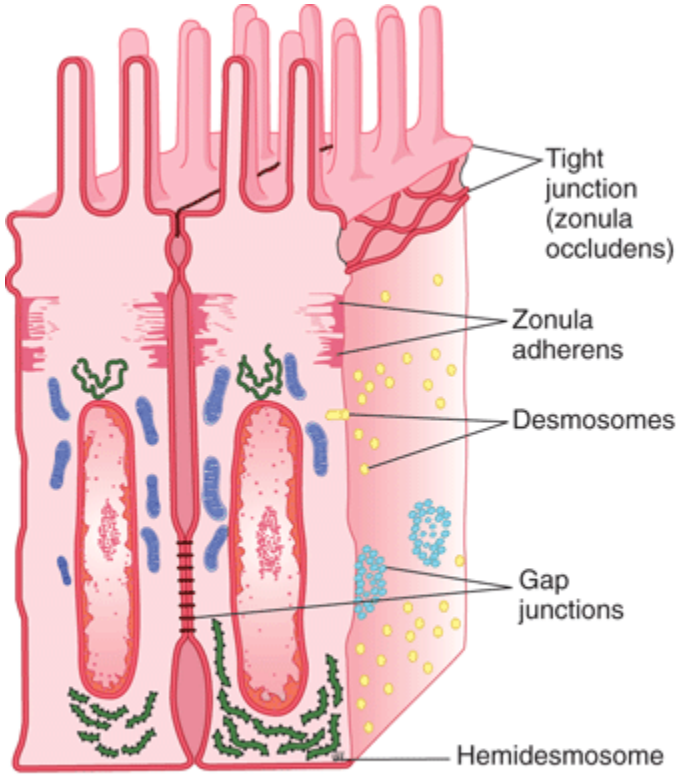
# Desmosomes

- Desmosomes are molecular complexes of cell adhesion proteins and linking proteins that attach the cell surface adhesion proteins to intracellular keratin cytoskeletal filaments.
- The cell adhesion proteins of the desmosome are members of the cadherin family of cell adhesion molecules.
- Cadherins are transmembrane proteins that bridge the space between adjacent epithelial cells by way of homophilic binding of their extracellular domains to other desmosomal cadherins on the adjacent cell.

# Tight junction

- Tight junctions are composed of a branching network of strands.
- Each strand acts independently of the others.
- Each strand is formed from a row of trans-membrane proteins embedded in both plasma membranes, with extracellular domains joining one another directly.
- The strands are anchored to the actin cytoskeleton and join the cyoskeletons of adjacent cells.
- They seal the cell.

# Intercellular connections




Source: Barrett KE, Barman SM, Boitano S, Brooks H: *Ganong's Review of Medical Physiology, 23rd Edition*: http://www.accessmedicine.com



Tight junctions (zonula occludens), adherens junctions (zonula adherens), desmosomes, gap junctions, and hemidesmosomes are all shown in relative positions in a polarized epithelial cell.

Fig. 2-8 Accessed 07/01/2010

# Gap junction

- A gap junction is a specialized intercellular connection between cells that  directly connects the cytoplasm of two cells.
- This allows molecules and ions to pass freely between cells.

# Gap junction

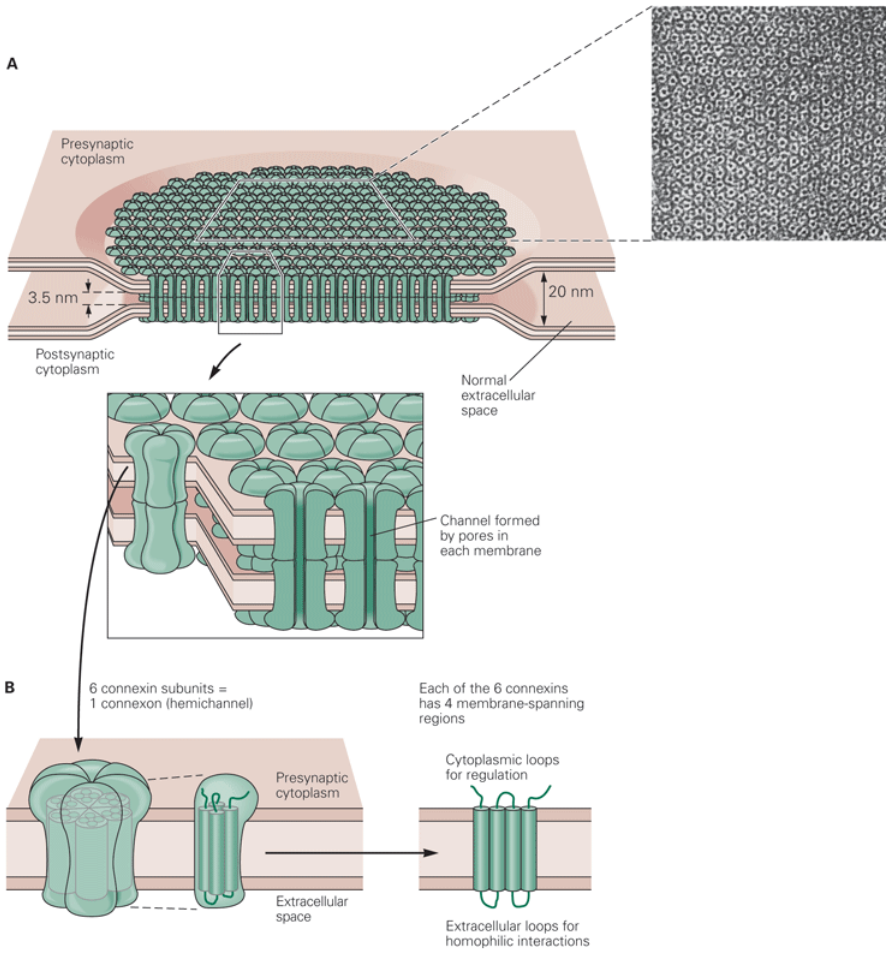




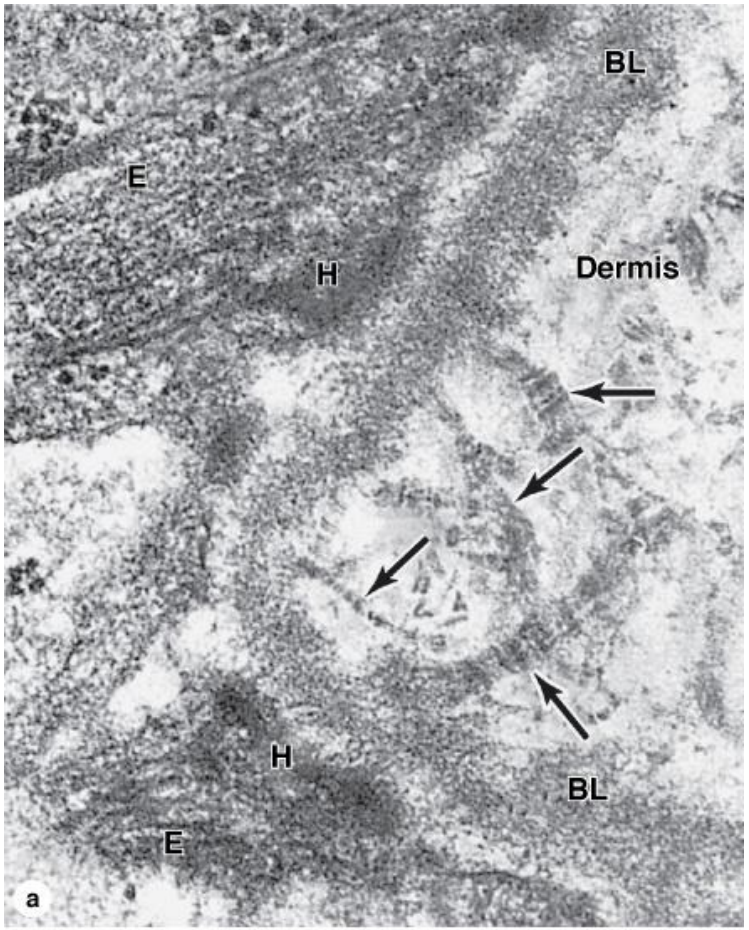

Fig. 2-9 Accessed 07/01/2010

A) A gap junction plaque, or collection of individual gap junctions, is shown to form multiple pores between cells that allow for the transfer of small molecules. Inset is electron micrograph from rat liver (N. Gilula).

B) Topographical depiction of individual connexon and corresponding 6 connexin proteins that traverse the membrane. Note that each connexin traverses the membrane four times.

(Reproduced with permission from Kandel ER, Schwartz JH, Jessell TM [editors]: *Principles of Neural Science,* 4th ed. McGraw-Hill, 2000.)

# Basal lamina






The basal lamina is a layer of extracellular matrix on which epithelium sits. It is secreted by epithelial cells.
The basal lamina (BL) is shown to have a dense layer with a clear layer on each side.
The underlying dermis contains anchoring fibrils (arrows) of collagen which help anchor the epithelium to the underlying connective tissue.
Hemidesmosomes (H) occur at the epithelial–connective tissue junction.
X54,000.

Fig. 4-2 Accessed 07/01/2010