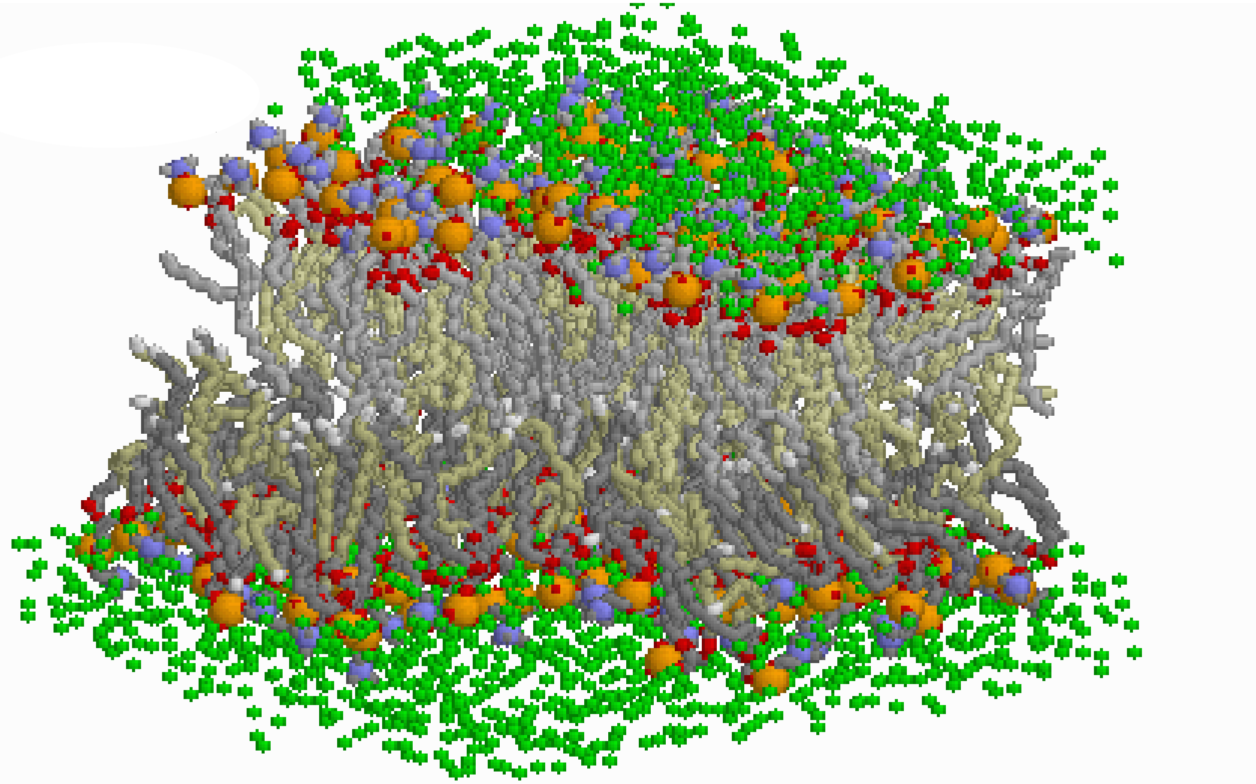
# Cell membrane – Structure and Functions



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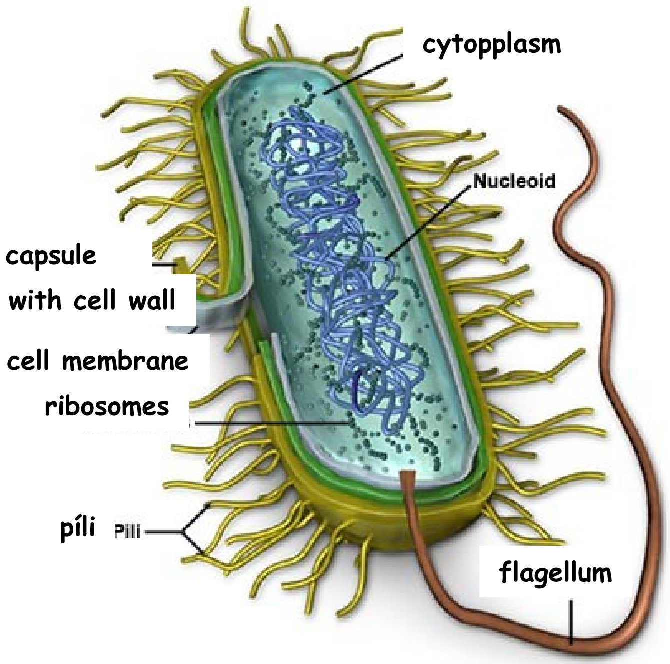
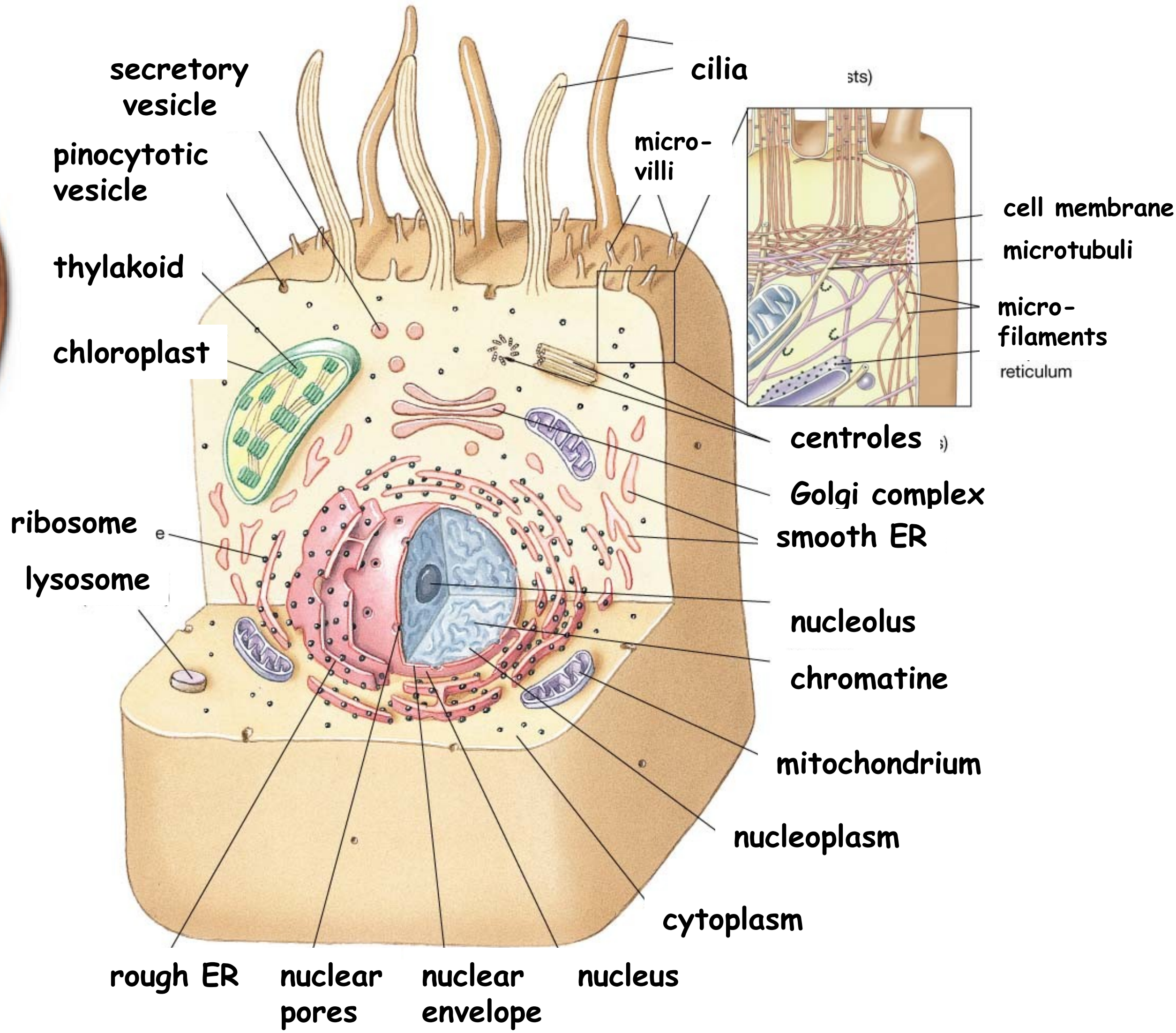
2023.

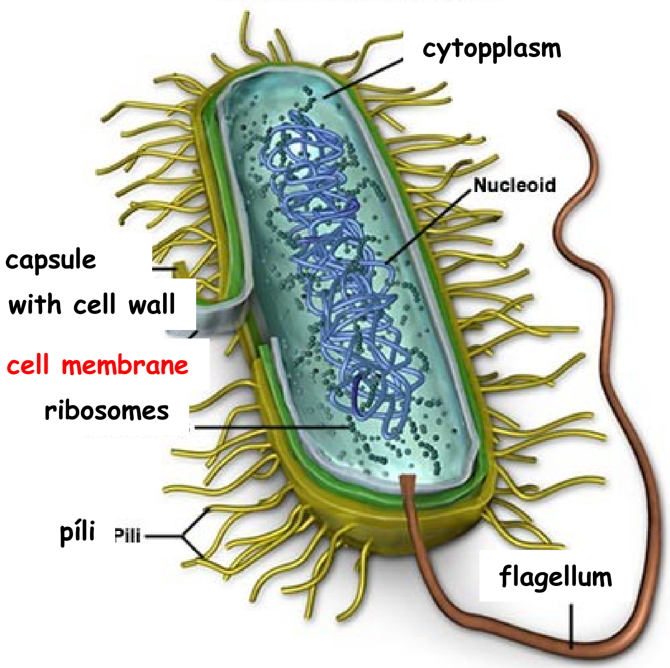
# Prokaryotic cell



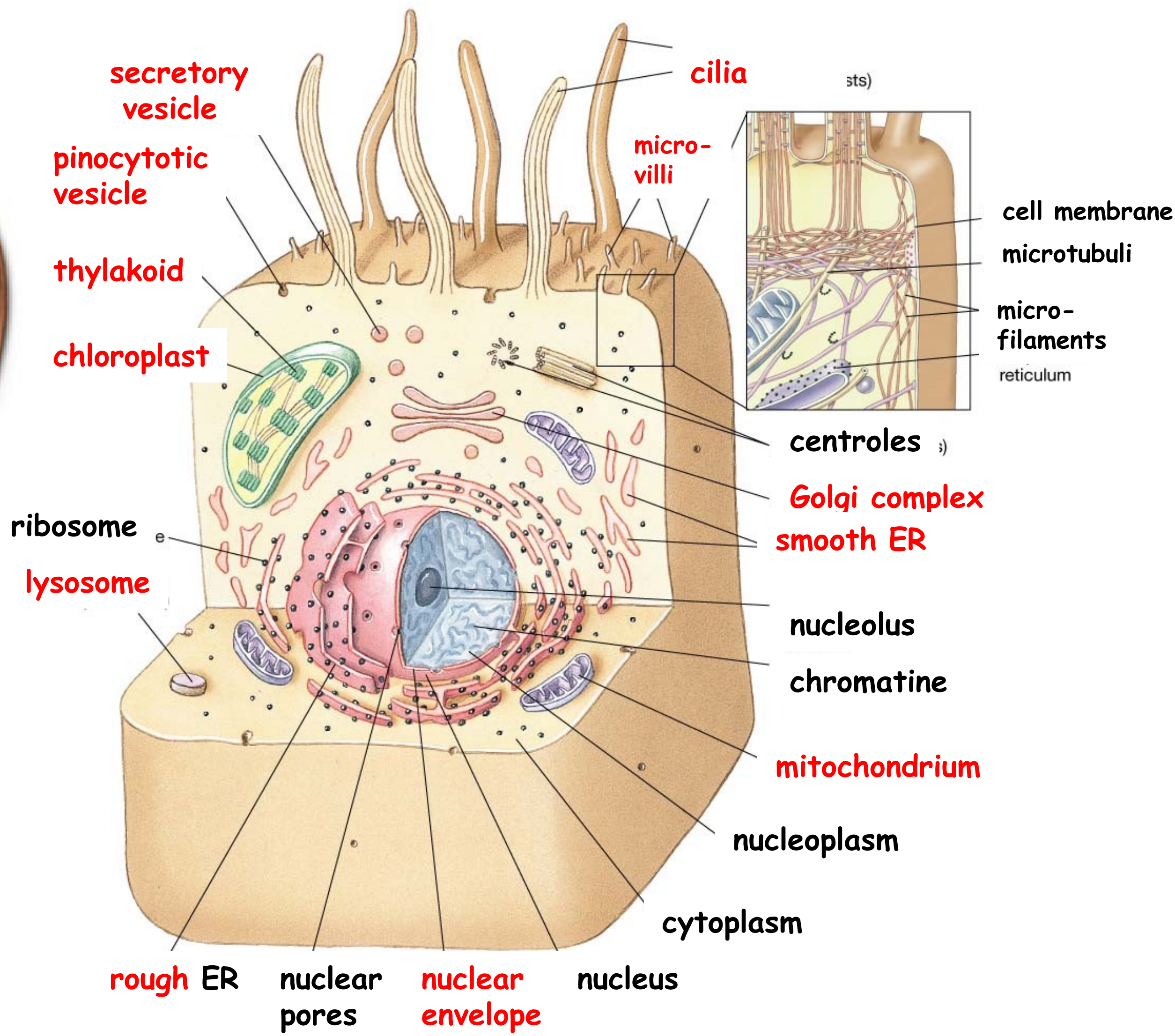

# Eukaryotic cell



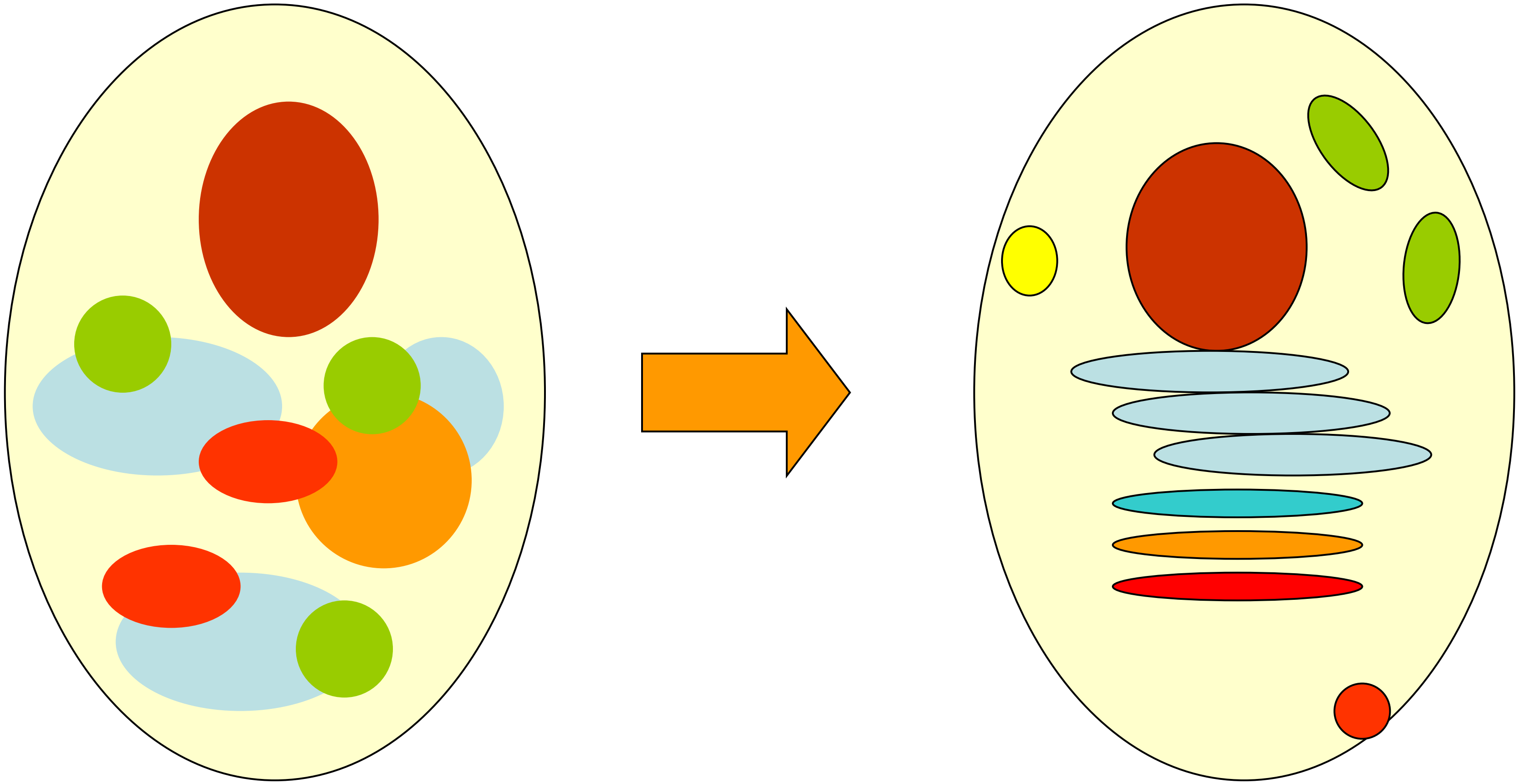
# Prokaryotic cell




# Eukaryotic cell

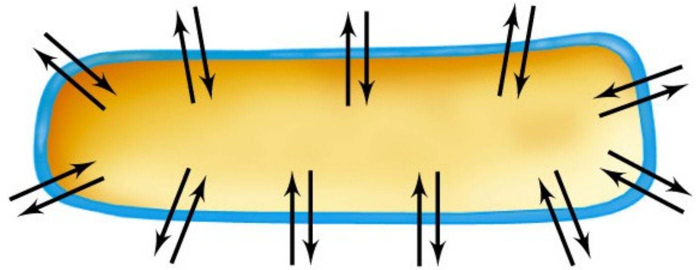


# Compartmentalization

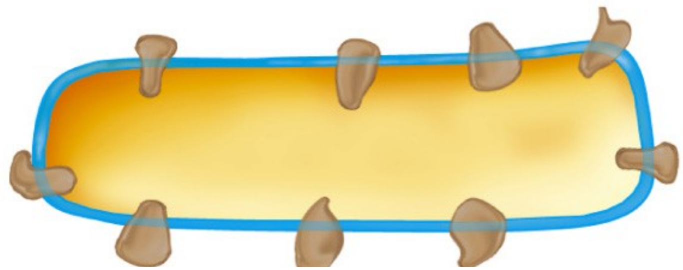


Biochemical pathways are running in isolated, membrane bounded spaces – This provides the optimal environmental conditions to the processes.

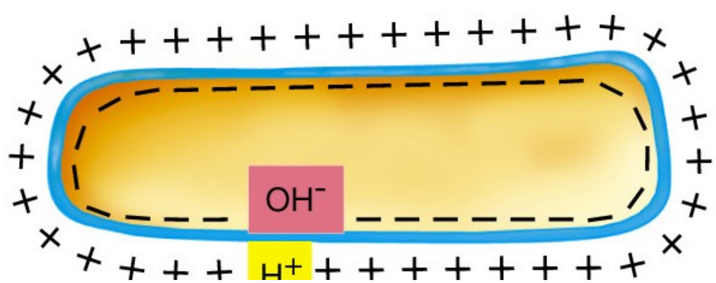
# Main functions of cytoplasmatic membranes



**Boundaries** – facilitates in- and outward transport of nutrients



**Anchoring proteins** – trasport, bioenergetical processes, sensing signals




**Generating and storage of energy** – generation of proton gradients

# Functions of membranes

•**1. Boundaries and selectively permeable** – cell membrane/plasma membrane and intracellular membranes – cell organells

•**2. Specific functions** – they are distinguished upon proteins associated (integrant or periferial proteins). Glucose phosphatase – rER-membrane associated enzyme and its marker, too.

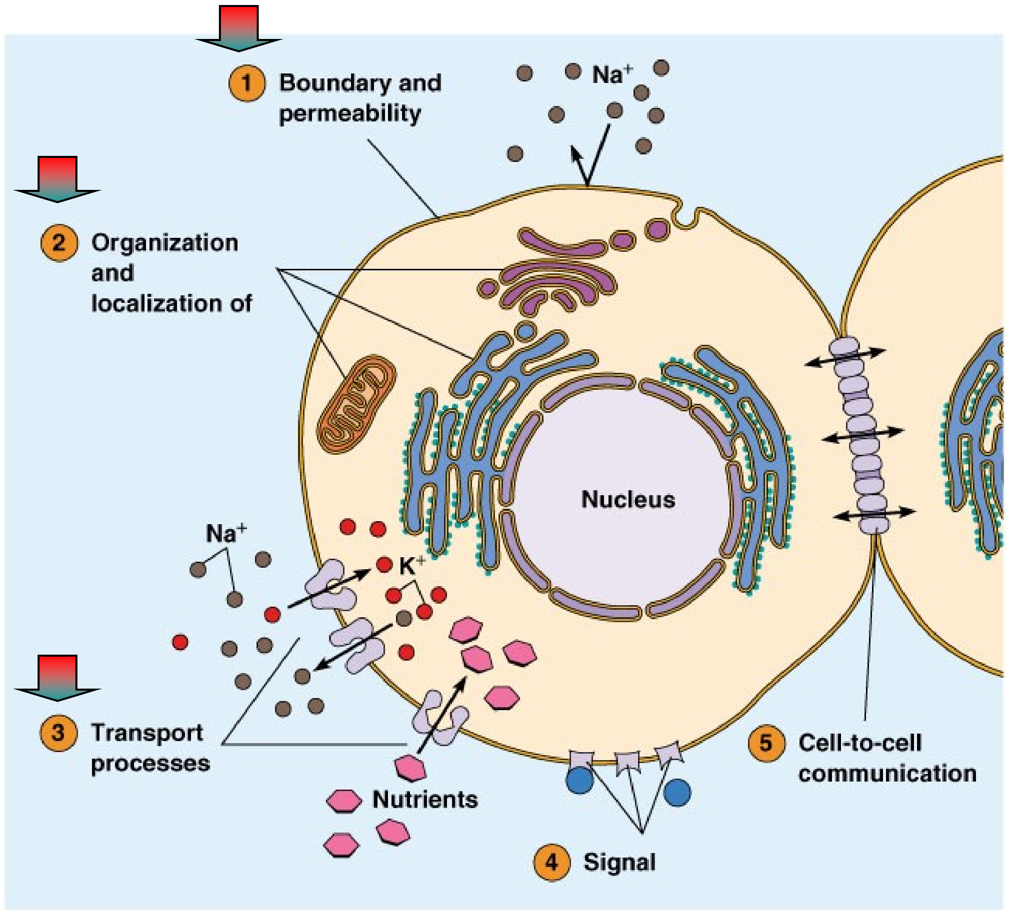
•**3. Transports are regulated by membranes** –

a. Simple **diffusion** (high cc.->low cc.): $H_2O$, $O_2$, $CO_2$, ethanol

b. **Facilitated** diffusion (high cc.->low cc.) by the help of transport proteines: saccharides, amino acids

c. Proteins of "**pump mechanisms**", they are essential in ion-gradients: low cc.-> higy cc. Intake of saccharides, amino acids against concentration gradient.

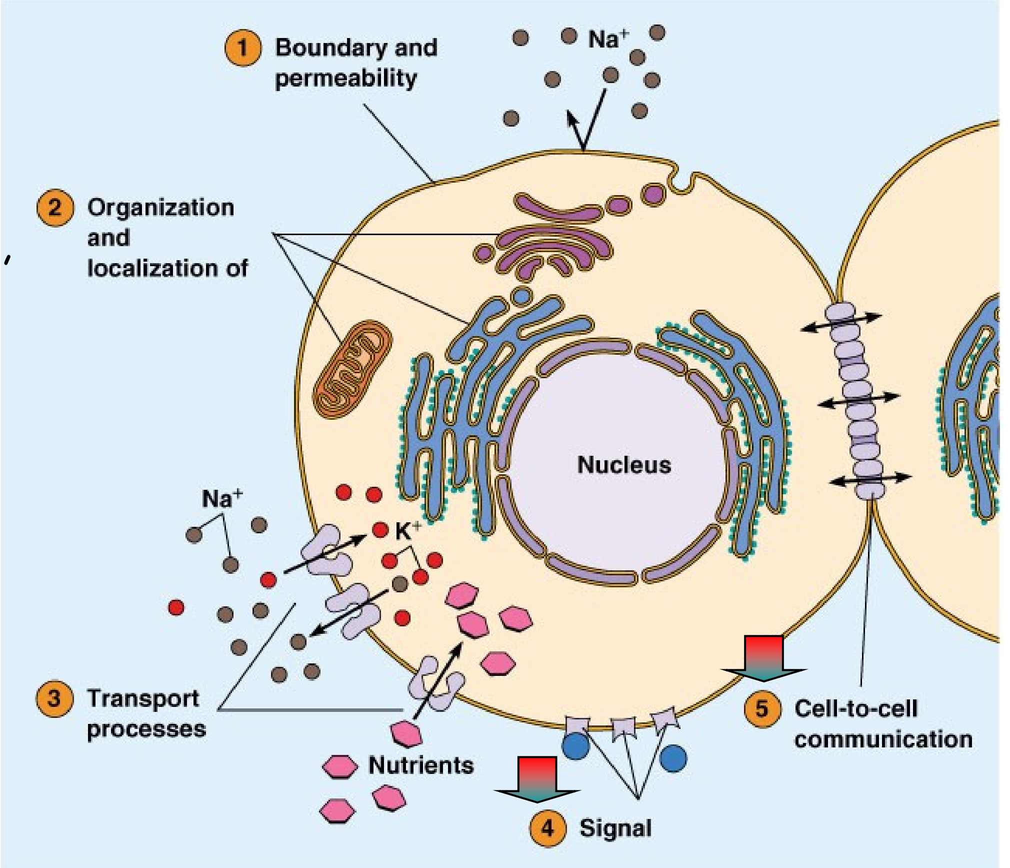
d. Endocytosis and exocytosis, etc.



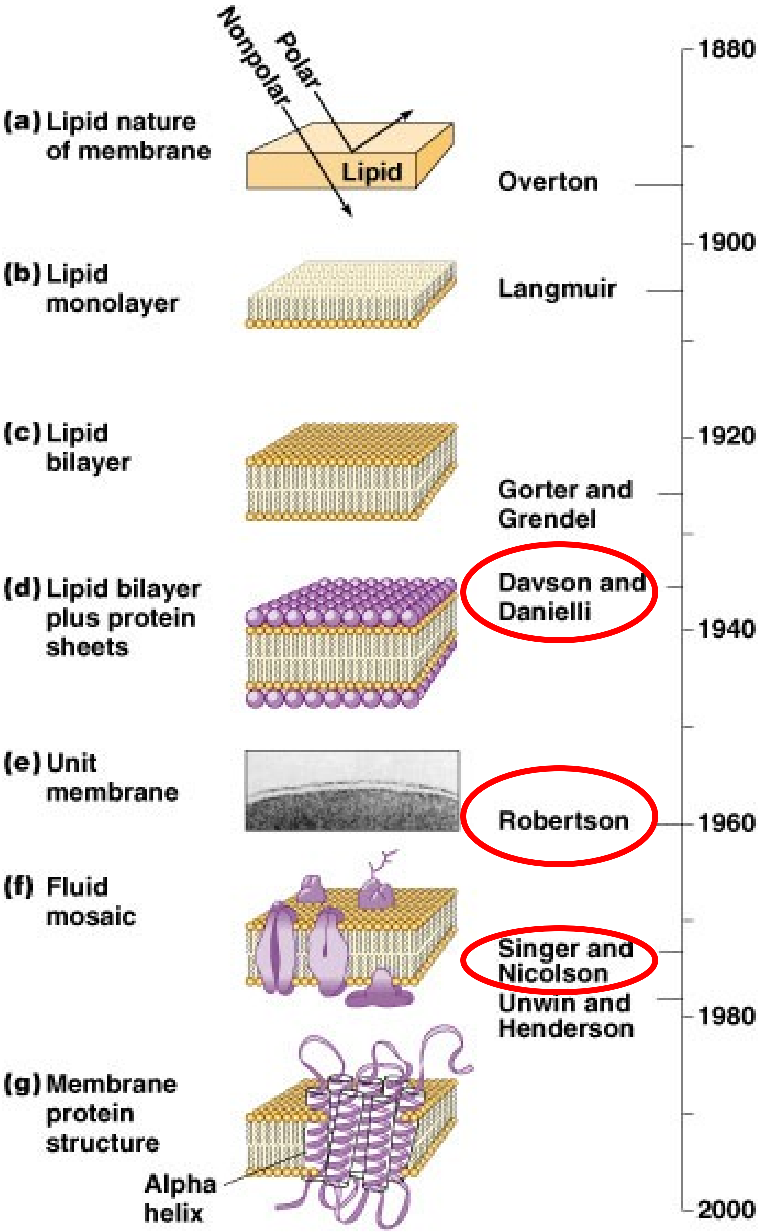
# Functions of membranes

•4. **Sensing and transmission of electric and chemical signals** – signal transduction: receiving and transmitting external signals
E.g.: Estrogen – can cross membranes due to its hydrophobic character; other molecules require receptors which process generates secondary messengers. The signal can reach even the nucleus and can influence levels of gene expression.

•5. **Cell-cell communication**
**The communication mediator components are** – gap junction (in animal cells) and plasmodesmata (in plant cells) allow direct, intercellular communication.



# Membrane-models





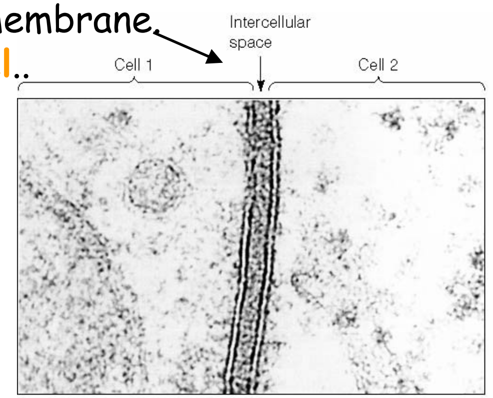

Lipid soluble substances enter the cell rapidly

Benzine-lipid mixture, after evaporation of benzine, a **molecular lipid film** is formed

Benzine-soluble lipids form a **bilayer** film on the surface of water

**Proteins are integral parts** of the cell membrane. The lipid bilayer is covered by a layer of proteins inside and outside. Partially explains the rapid passage of proteins, sugars, ions and other hydrophilic substances.

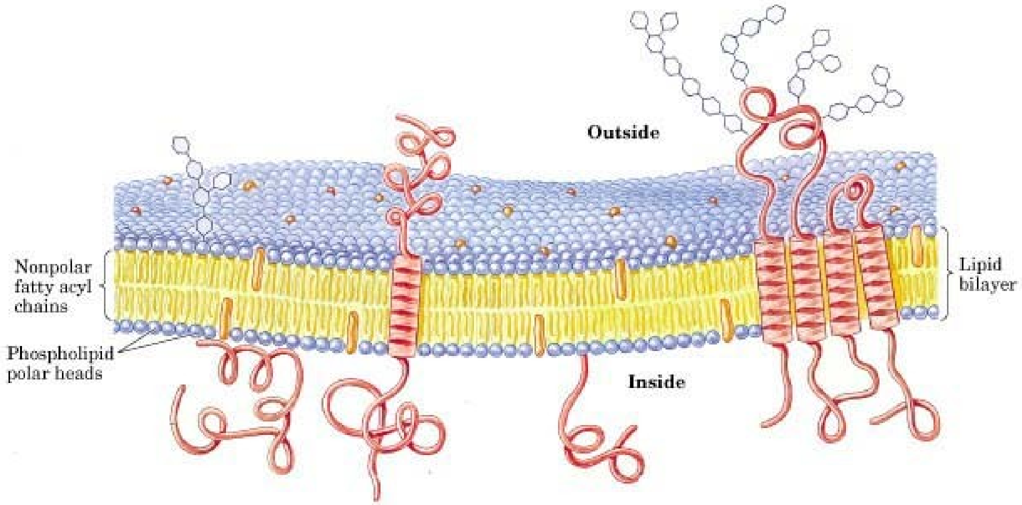
Discovery of electron microscope. The cells are covered with a plasma membrane. **"Unit-membrane" model**..



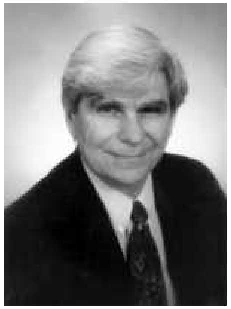

**Mosaic** arrangement of proteins in the membrane.

Some proteins cross the membrane, **transmembrane proteins**

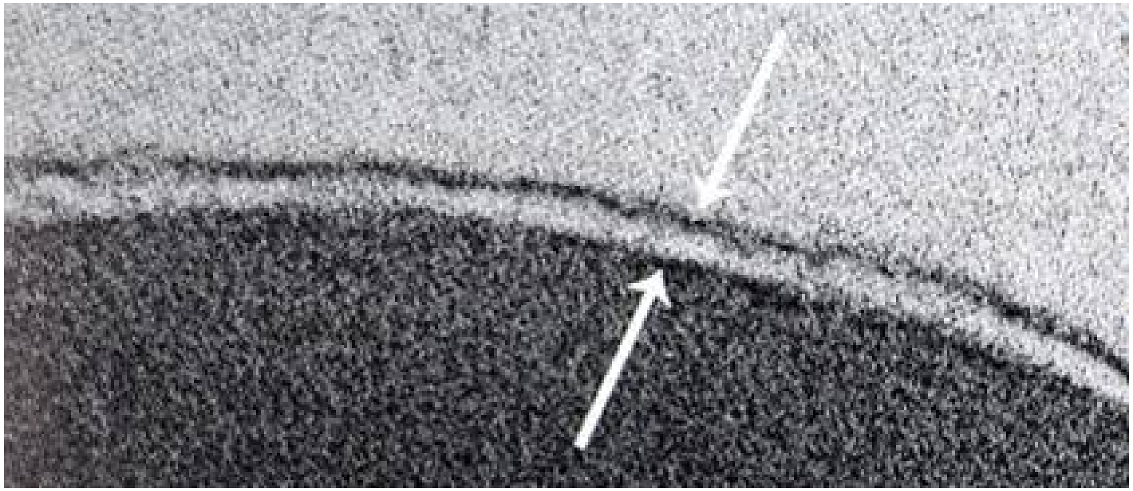
# Fluid mosaic membrane model




Singer – Nicolson
1972



RBC membrane



# Membrane models – Fluid mosaic model:

Proteins arranged mosaic-like, scattered and embedded within the liquid lipid bilayer

**It is the first protein to be described by the transmembrane domain and with its retinol-like and also energy-binding moiety.**
**The energy of sunlight after binding causes a structural change that results in the release of protons from the cell. The resulting proton gradient serves as a source of energy.**

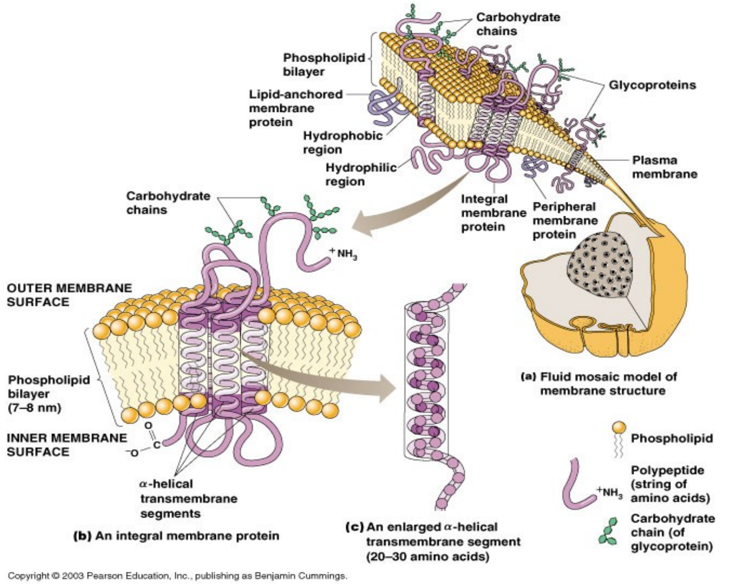


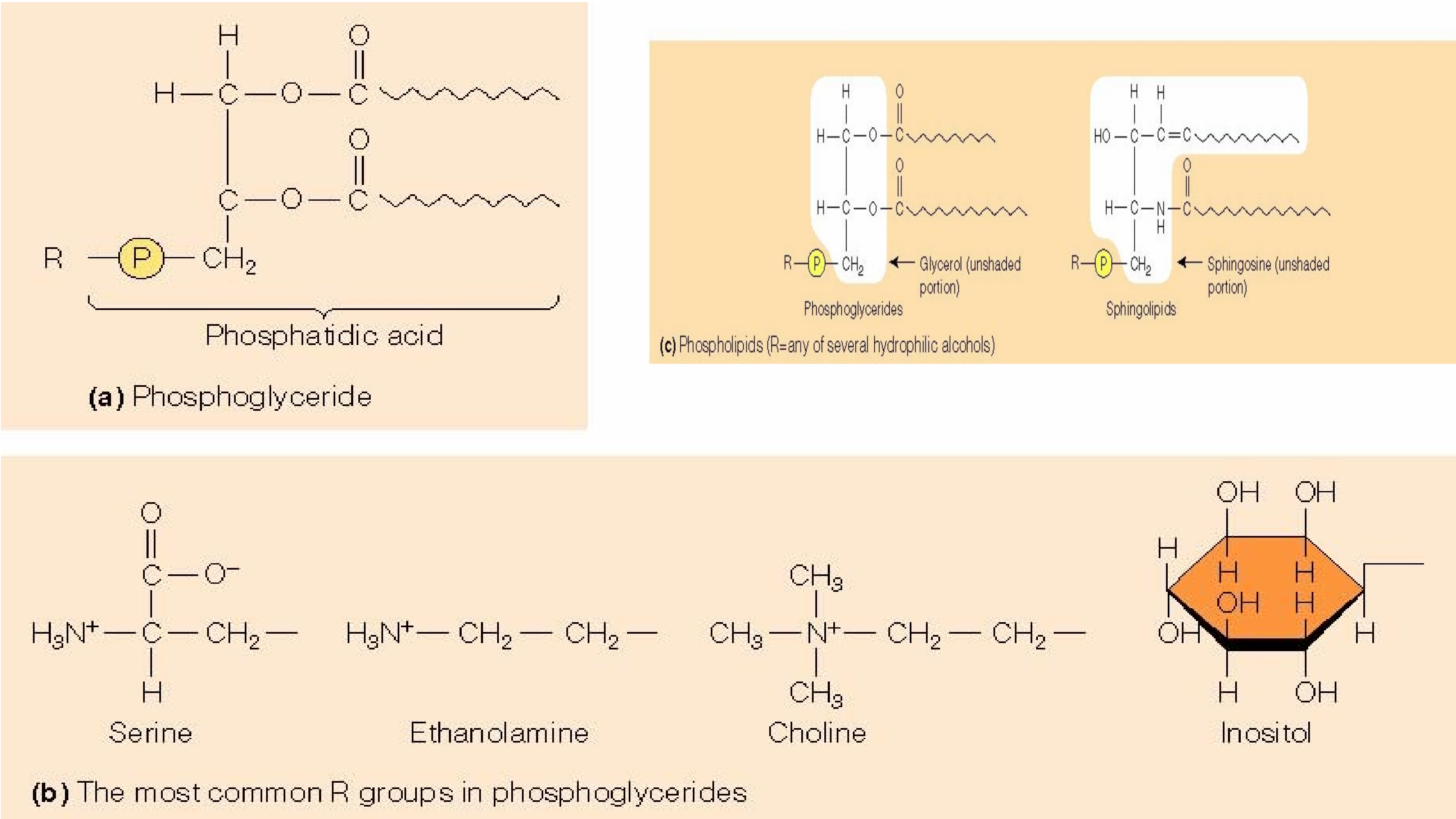

**Table 7-1 Protein, Lipid, and Carbohydrate Content of Biological Membranes**

| Membrane | Approximate Percent by Weight | | | Protein/Lipid Ratio |
|---|---|---|---|---|
| | Protein | Lipid | Carbohydrate | |
| Plasma membrane | | | | |
| Human erythrocyte | 49 | 43 | 8 | 1.14 |
| Mammalian liver cell | 54 | 36 | 10 | 1.50 |
| Amoeba | 54 | 42 | 4 | 1.29 |
| Myelin sheath of nerve axon | 18 | 79 | 3 | 0.23 |
| Nuclear envelope | 66 | 32 | 2 | 2.06 |
| Endoplasmic reticulum | 63 | 27 | 10 | 2.33 |
| Golgi complex | 64 | 26 | 10 | 2.46 |
| Chloroplast thylakoids | 70 | 30 | 0 | 2.33 |
| Mitochondrial outer membrane | 55 | 45 | 0 | 1.22 |
| Mitochondrial inner membrane | 78 | 22 | 0 | 3.54 |
| Gram-positive bacterium | 75 | 25 | 0 | 3.00 |



**Lipids, proteins, and carbohydrates are also involved in the construction of most membranes.**

# General structure of phospholipids




(a) Phosphoglyceride

(c) Phospholipids (R=any of several hydrophilic alcohols)

(b) The most common R groups in phosphoglycerides

## The main lipid components that make up the membrane

Phospholipides
Glycolipides

## Other membrane lipids (not phospholipids)

Cholesterol

Cerebrosides
Sphingolipides
Ceramide

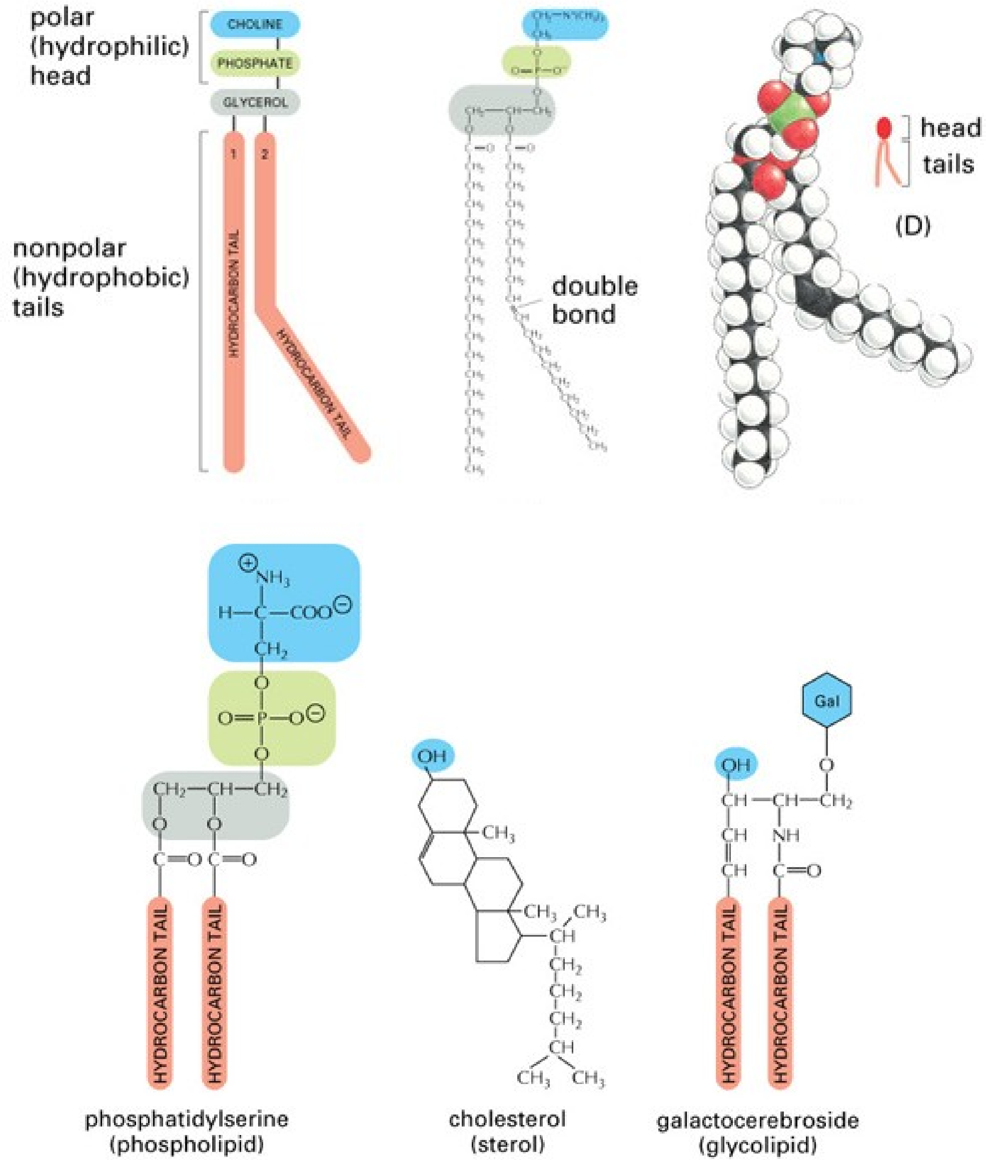


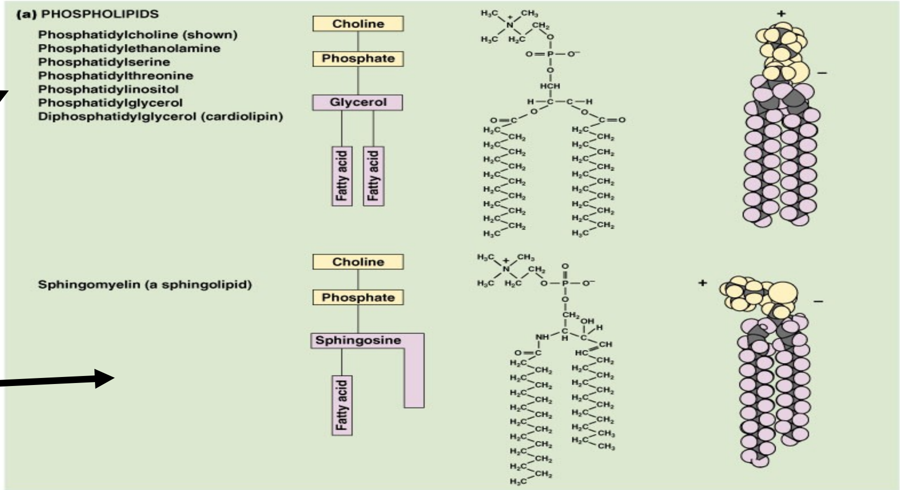

Figure 11-7 Essential Cell Biology, 2/e. (© 2004 Garland Science)

# Membrane lipids: elements responsible for the fluidity of the model

Several types of lipids are involved in the formation of membranes
- Phospholipids

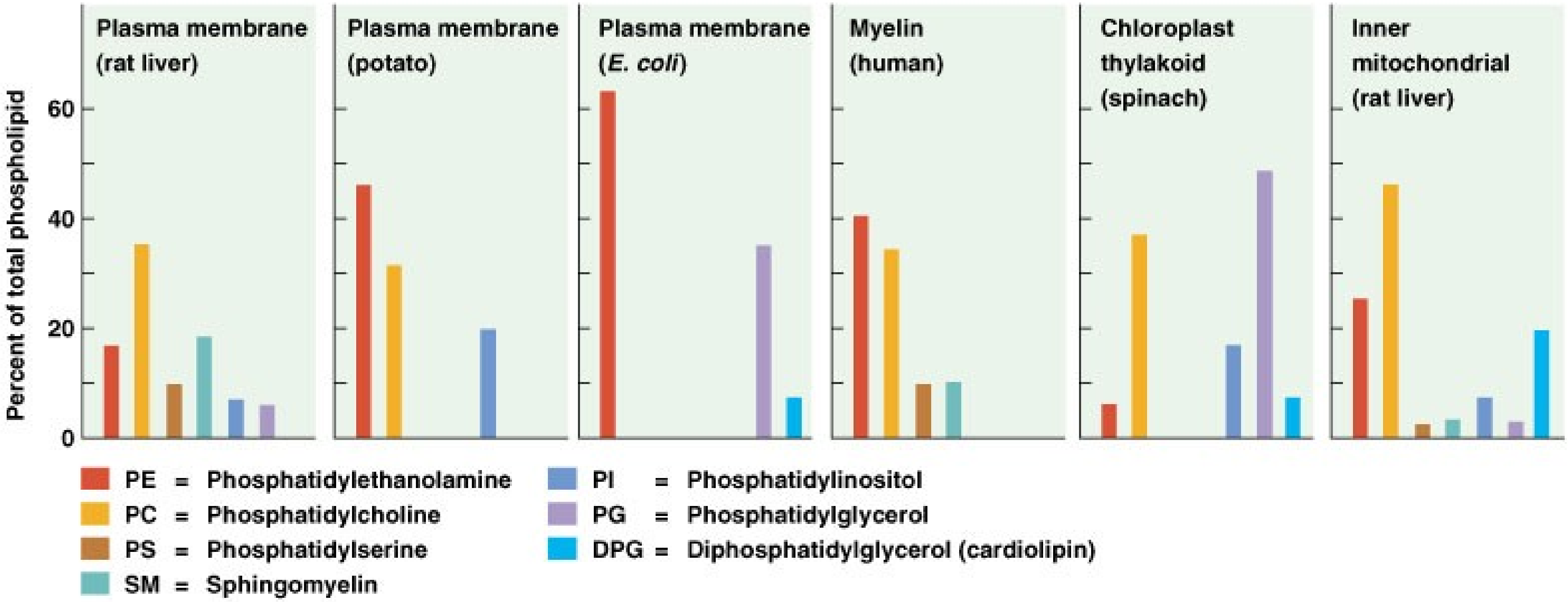
Phosphoglycerides

Sphingolipids





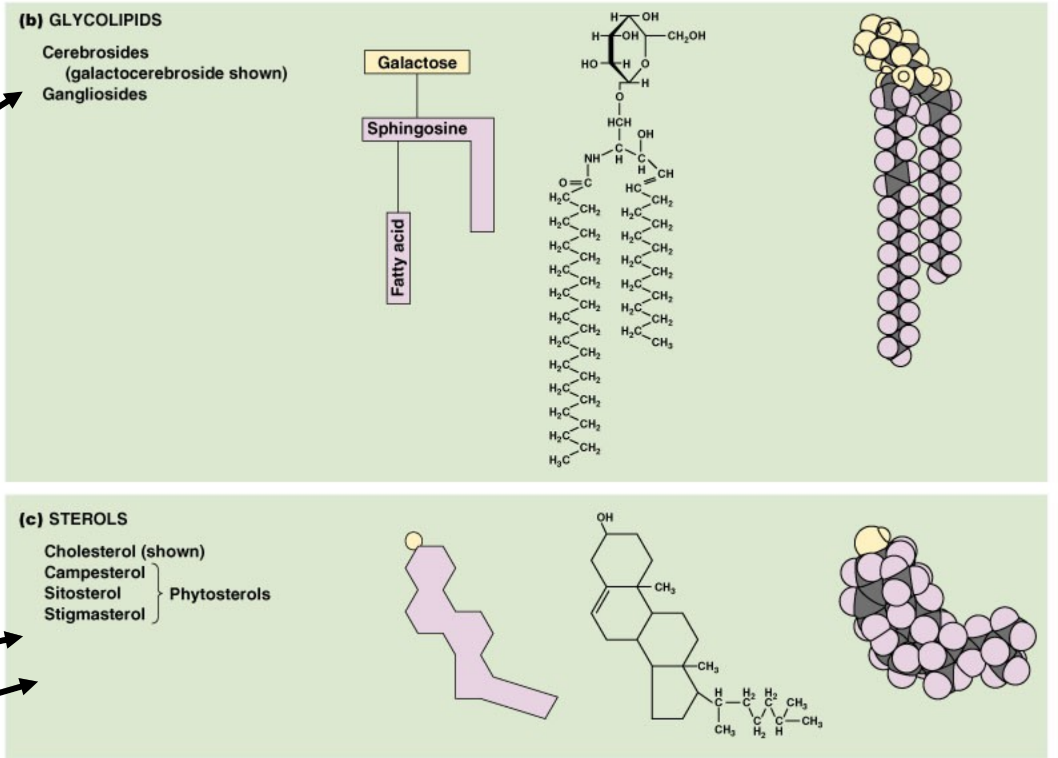

Phospholipid composition of different membranes



- **Glycolipids**

  Cerebrosides and gangliosides

  Tay-Sachs disease:
  Lysosomes are deficient in β-N-acetilhexoseaminidase – degradation of gangliyosides is impaired; ganglyoside accumulation in CNS.

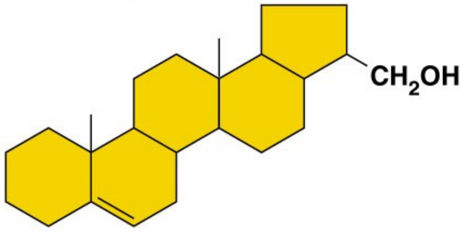




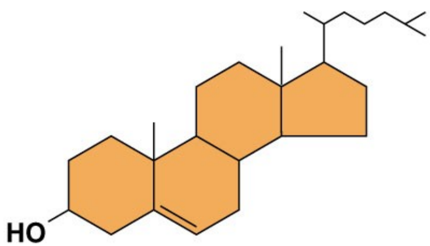

- **Steroids** (they are not found in prokaryotes or in the inner membranes of the mitochondria, the color body)

  Cholesterol

  Phytosterol




**(a)** A hopanoid




**(b)** Cholesterol

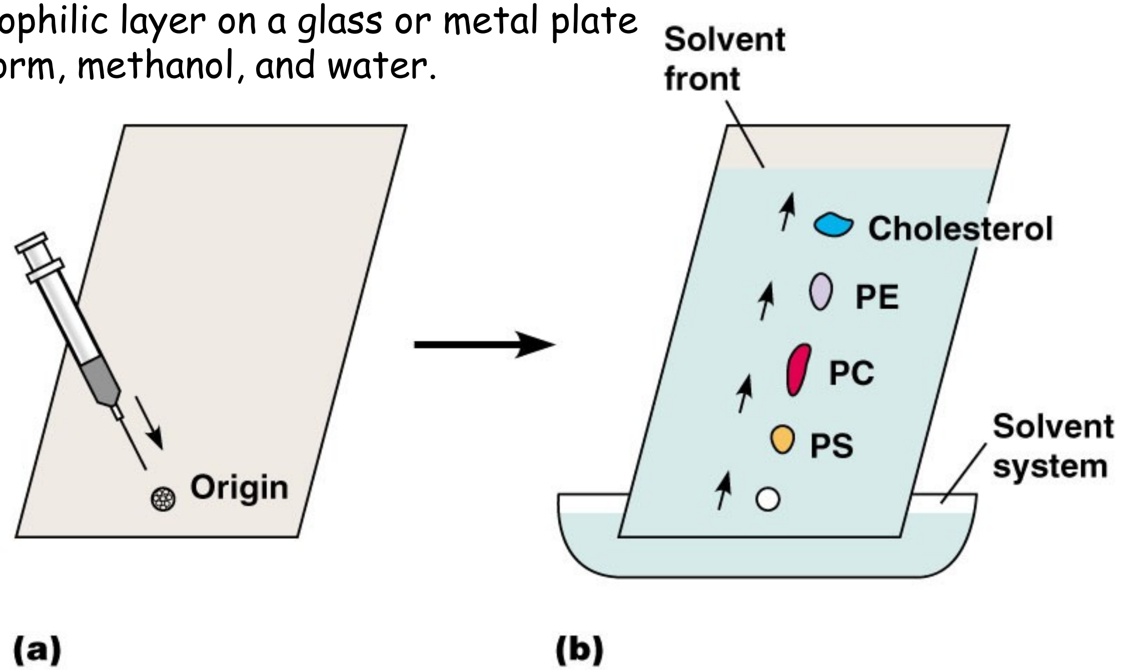


The membranes of bacteria and cyanobacteria contain steroid-like molecules - haponoid (rich in petroleum deposits; membrane components of early prokaryotes?)

# Thin layer chromatography - an important tool for lipid analysis:

Stationary phase - thin silica gel with a hydrophilic layer on a glass or metal plate
Mobile phase - a mixture of solvent, chloroform, methanol, and water.

| | |
|---|---|
| Low affinity | Cholesterol |
| ↑ | Phosphatidylethonolamine (PE) |
| | Phosphatidylcholine (PC) |
| High affinity | Phosphatidylserine (PS) |





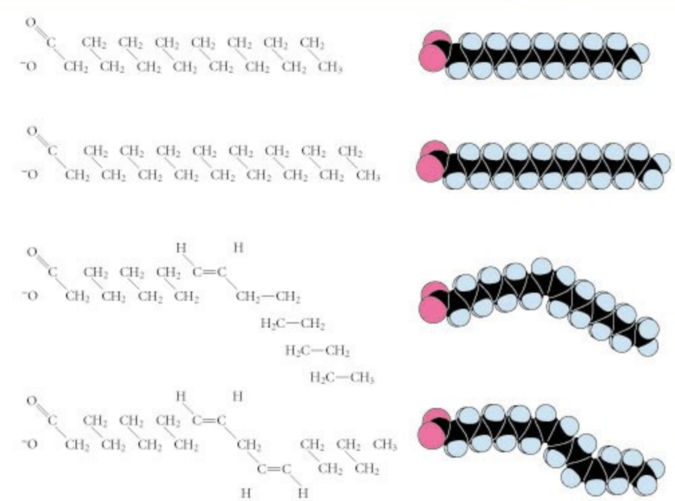

**Fatty acids are also important for membrane structure and function**

Fatty acids are components of all membrane lipids except steroids.
Hydrophobic hydrocarbons are generally 16- or 18-C long and provide stability to the lipid bilayer..

Saturated Fatty Acids: Palmitic (16C) and Stearic (18C)
Unsaturated Fatty Acids: Oleic and Linoleic Acids (both are 18C)

Table 7-2 Structures of Some Common Fatty Acids

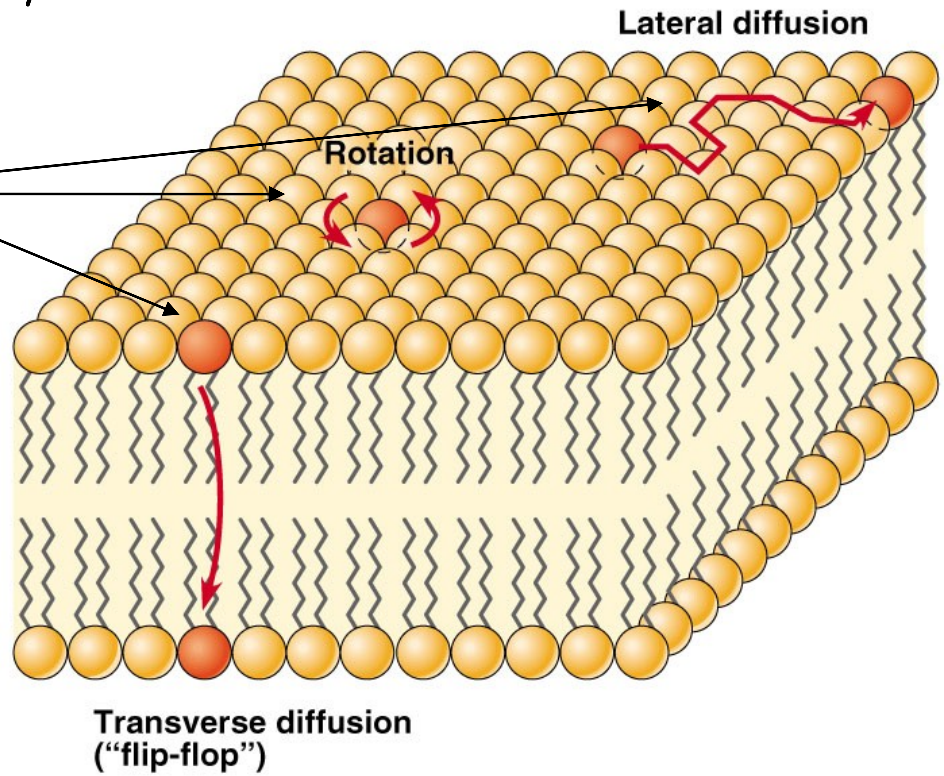
| Name of Fatty Acid | Number of Carbon Atoms | Number of Double Bonds | Structural Formula | Space-Filling Model |
|---|---|---|---|---|
| Saturated | | | | |
| Palmitate | 16 | 0 | | |
| Stearate | 18 | 0 | | |
| Unsaturated | | | | |
| Oleate | 18 | 1 | | |
| Linoleate | 18 | 2 | | |



## Membrane asymmetry: the majority of lipids are unequally distributed in the two lipid monolayers - differences in quantity and quality

Asymmetry is embodied in differences in lipid types and degrees of saturation.
Asymmetry occurs during the biogenesis of membranes.
The resulting asymmetry, for thermodynamic reasons, prevents the exchange of lipids between the two surfaces.
External Membrane: Glycolipids - Recognition processes and signaling
Internal membrane: important for signaling to the cell's interior - phosphatidylethanolamine, phosphatidylinositol, phosphatidylserine
Membrane asymmetry: the majority of lipids are unequally distributed in the two lipid monolayers - differences in quantity and quality

Movement of lipids in the membrane:
- "Flip-flop" or inverse diffusion
- rotation
- lateral diffusion

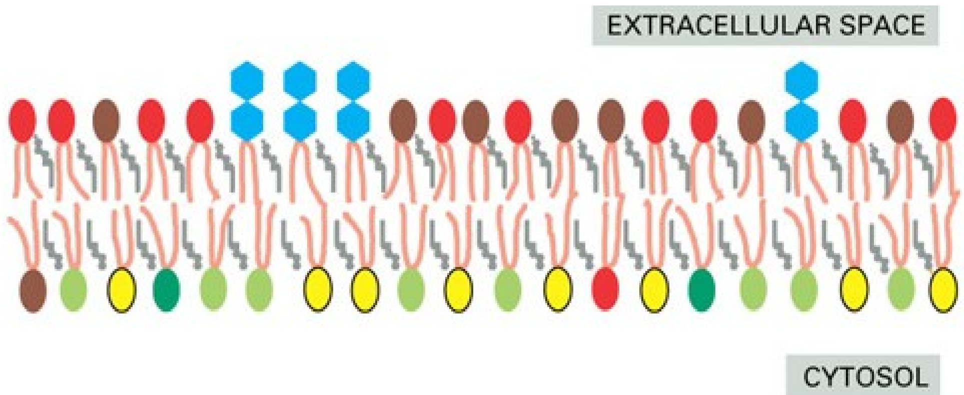




Flip-flop movement is rare, but is promoted in biological membranes by phospholipid translocators or flippases.

Therefore, phospholipid translocation in biological membranes is more common than in artificial lipid bilayers.

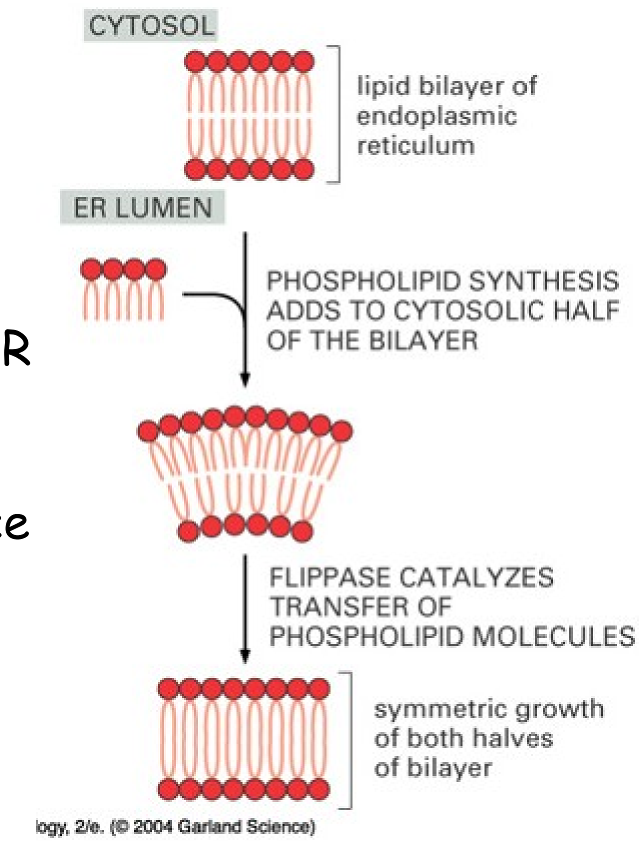
# Development and maintenance of membrane asymmetry :




- Phospholipids are synthesized bound to the ER membrane.

- Phospholipids bind to the cytoplasmic surface of the ER

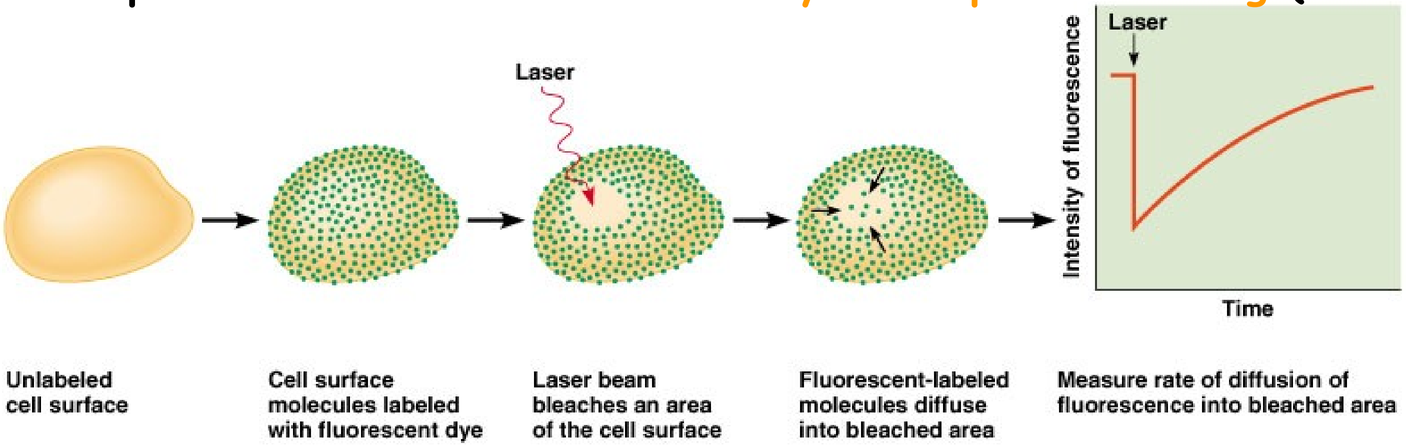
- Flippase transfers each lipid to the other side of the bilayer.



# Investigation of the fluidity of the lipid bilayer

By lateral diffusion, lipids and proteins, can move a few millimeters (equal to the length of a bacterium) of a lipid of 800 M.S. in 1 second! Due to their large size, proteins move more slowly.

**Test procedure: fluorescence recovery after photobleaching (FRAP) technique**






**The membrane only functions properly in its "fluid" state**

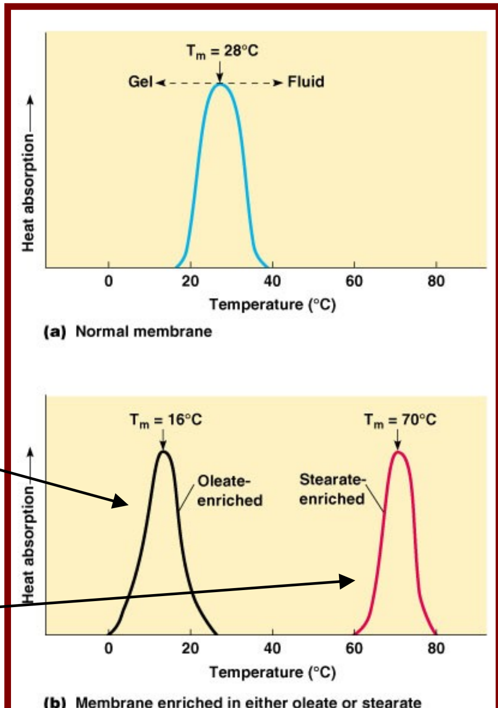
Phase transition temperature (Tm):
Phase transition: gel-like state -> fluid

Unsatuirated:
Low Tm

Saturated:
High Tm

## Differential scanning calorimetry

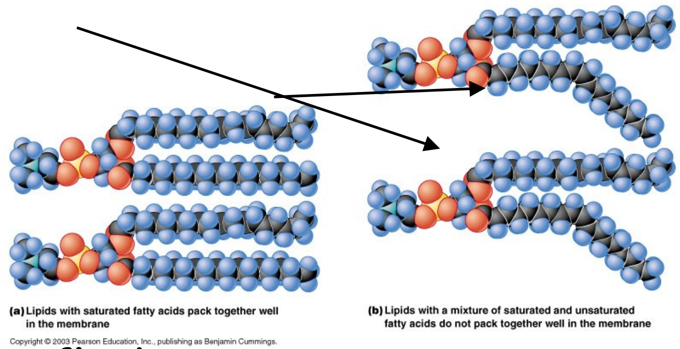
Calorimeter: chamber for measuring heat absorption.
The location of the peak of the curve indicates Tm.

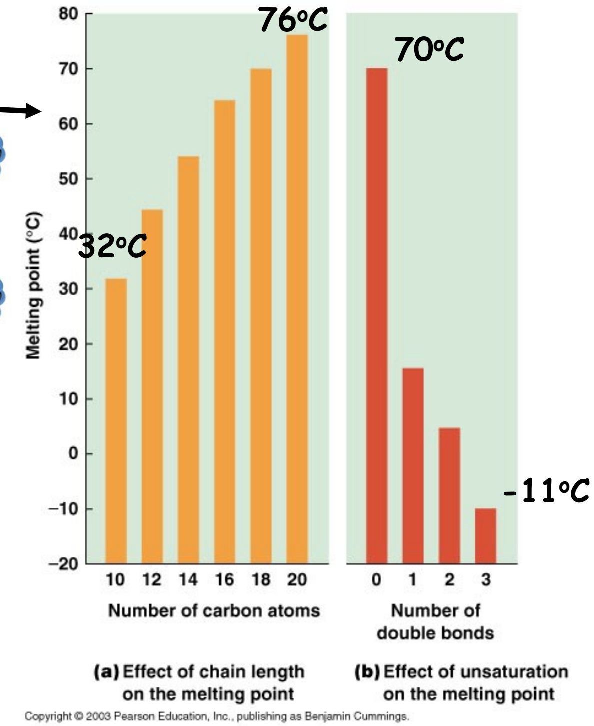


## Membrane-fluidity and fatty acid composition

There are two main aspects: chain length and degree of saturation

Membrane-lipids often consist of 1 saturated and 1 unsaturated fatty acid




18-C lipids

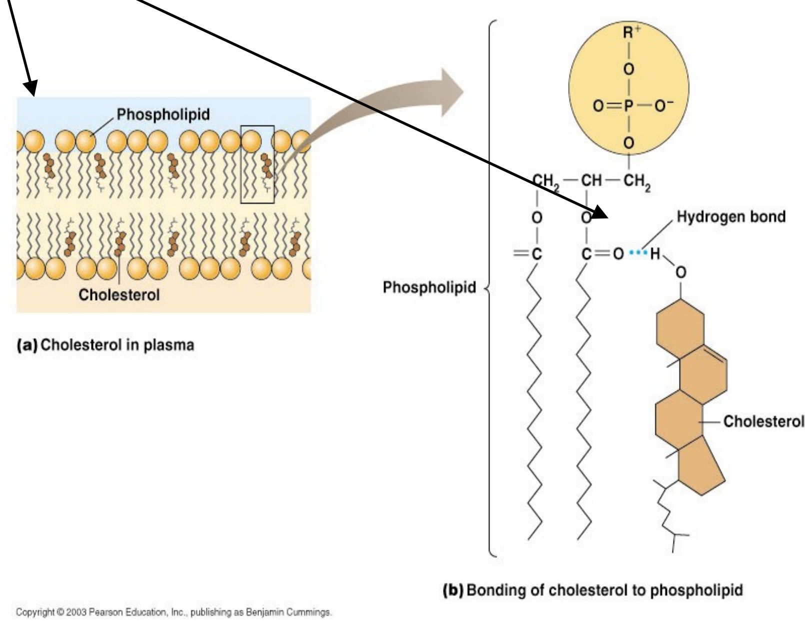
## Effect of steroids on membrane fluidity

Steroids - cholesterol and phytosterol. Steroids may represent up to 50% of the total membrane lipid content

Cholesterol - forms hydrogen bonds with the oxygen atoms of neighboring phospholipids.

**Paradoxical effect of cholesterol**: decreases membrane fluidity at high temperatures (effect of rigid rings) and increases fluidity at low temperatures (inhibiting the natural arrangement of adjacent hydrocarbon chains)

HOWEVER, steroids reduce the permeability of the lipid bilayer - filling the space between phospholipids.




## The organisms can control membrane fluidity

**Poikelotherms** (bacteria, fungi, plants, amphibians, etc.) **vs.** **homeotherms** (warm-blooded animals)

**Homeoviscopic adaptation** - membrane viscosity retained despite changing temperature - shortening or reduction of fatty acid chains by removal of terminal C's. de-saturation (e.g., E. coli desaturase)

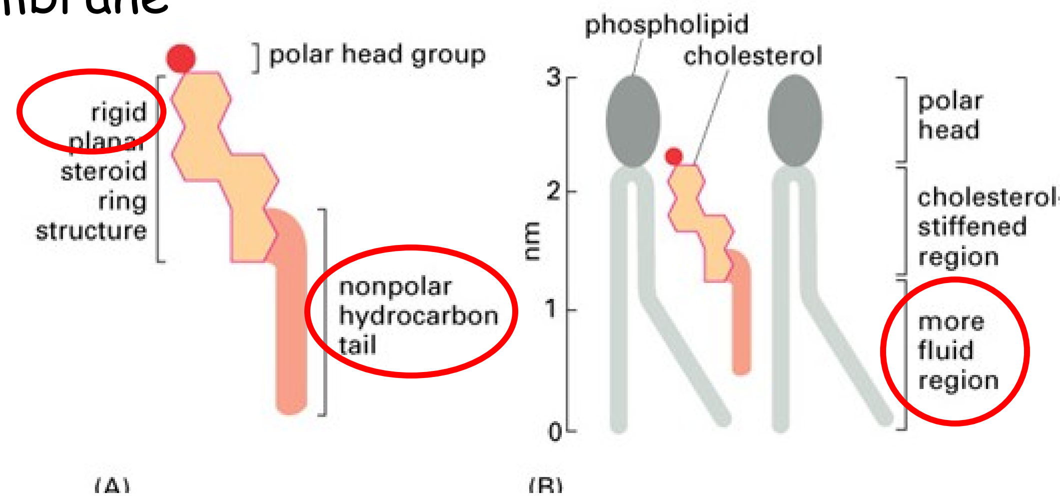
The fluidity of the membrane is influenced by:

phospholipids:

chain length

saturation

Cholesterol content



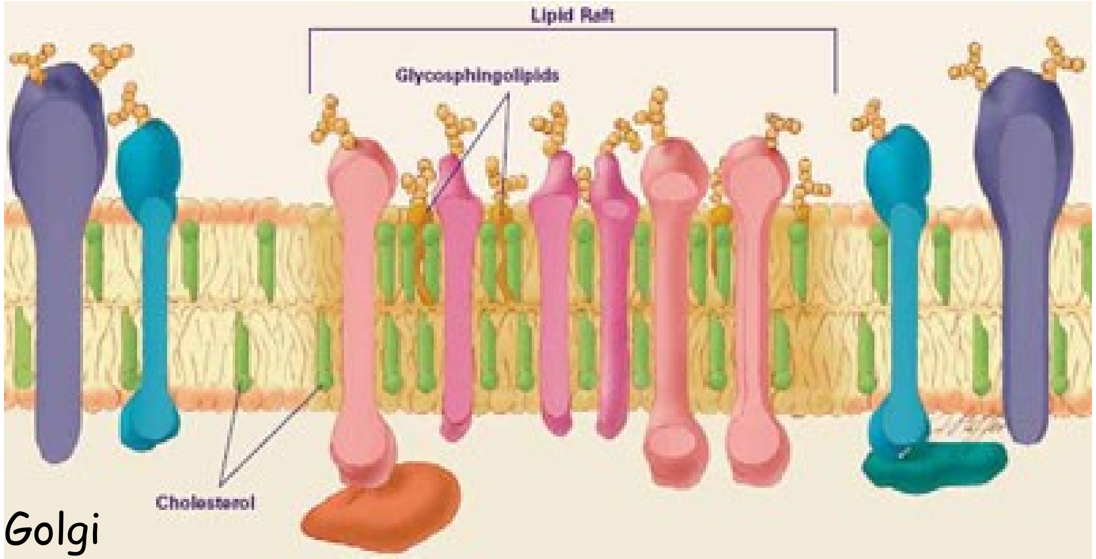
# Lipid rafts:

- High in cholesterol

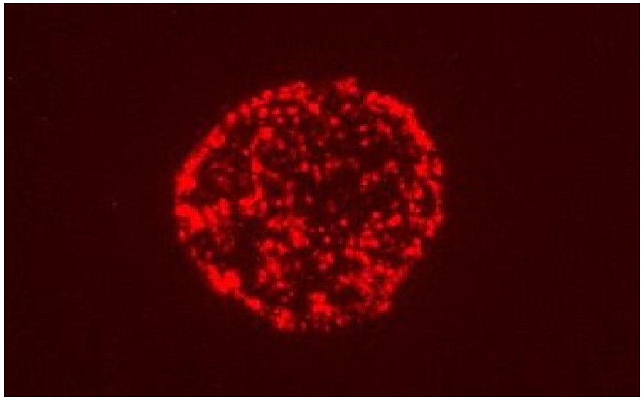
- Micro-domains

-Glycolipids + sphingolipids

- Rafts are present in the cell surface membrane and on membranes attached to the Golgi




-Their importance: to reduce the **difference in free energy** between each cell membrane layer membrane bound transport processes



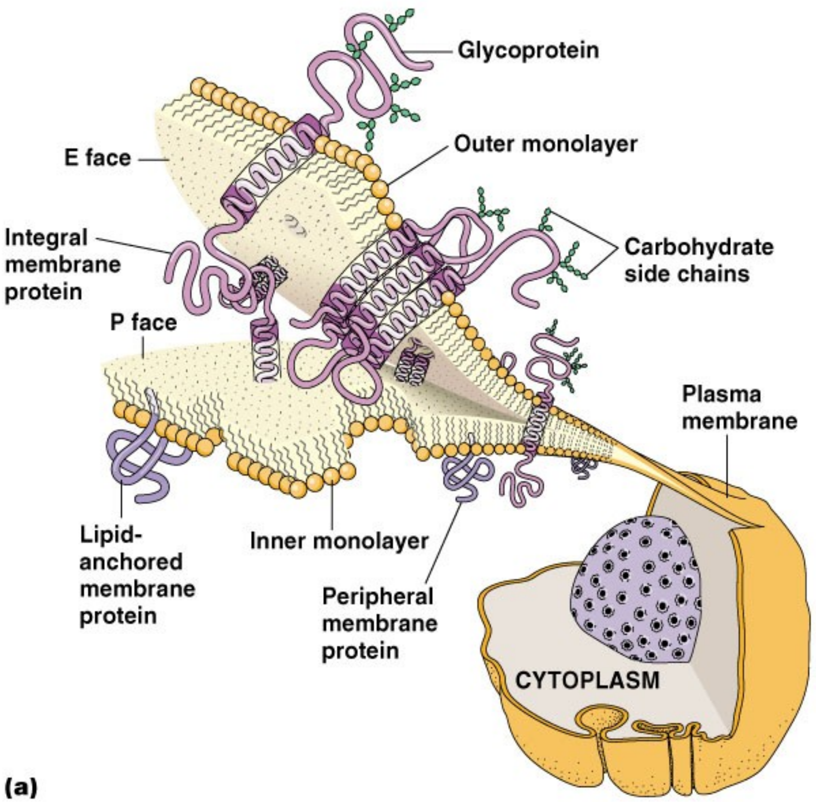
Other names in the literature:
- detergent-insoluble glycolipid-enriched complexes (GEM or DIG)
- detergent resistant membranes (DRMs)

Detection: fluorescence microscopy - GM1 gangliosides; AFM

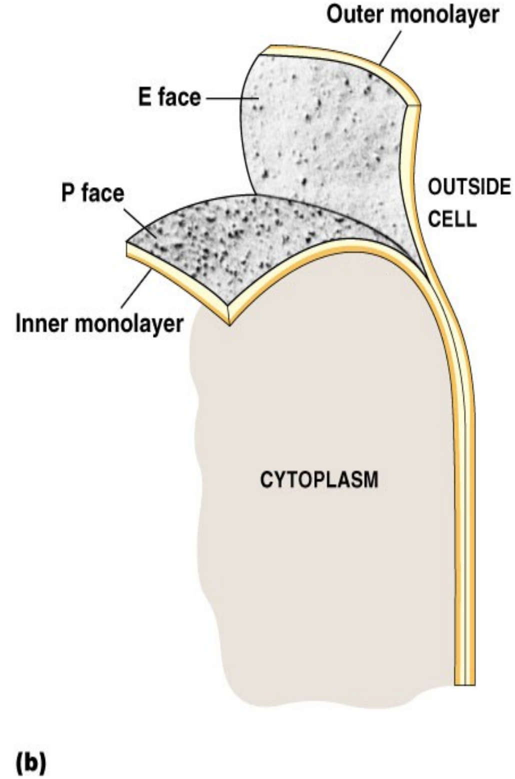
# Membrane Proteins: The "mosaic" character of the model

Freezing fracturing: rapid freezing (in liquid nitrogen) then diamond blade exploratin after

**E face (exoplasmatic face) and P face (protoplasmatic face)**





# Membrane proteins:

- integrant
- peripheric
- anchored by lipids

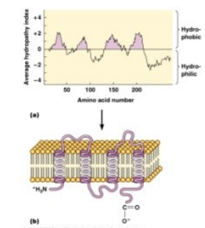
**Integrant membrane proteins:** ~20 amino acids, transmembrane segment. Hard to isolate, extraction requires treatment with detergents
.

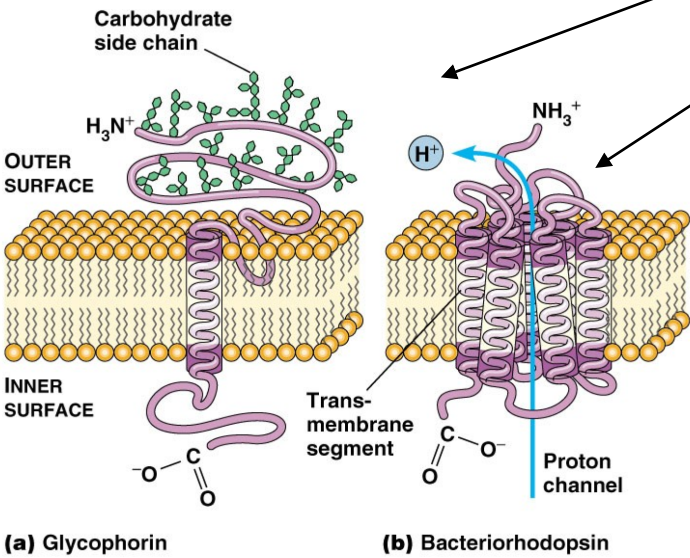
Intergrant, monotopic proteins
Transmembrane proteins:
singlepass
multipass
multi-subunit

Glycophorin (vvt.)

Bacteriorhodopsin

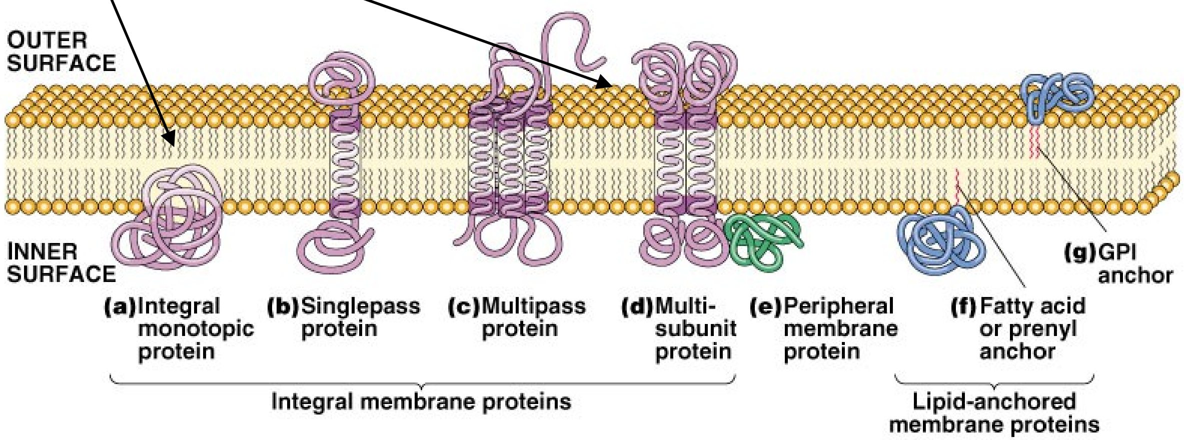
Hydropathy plot

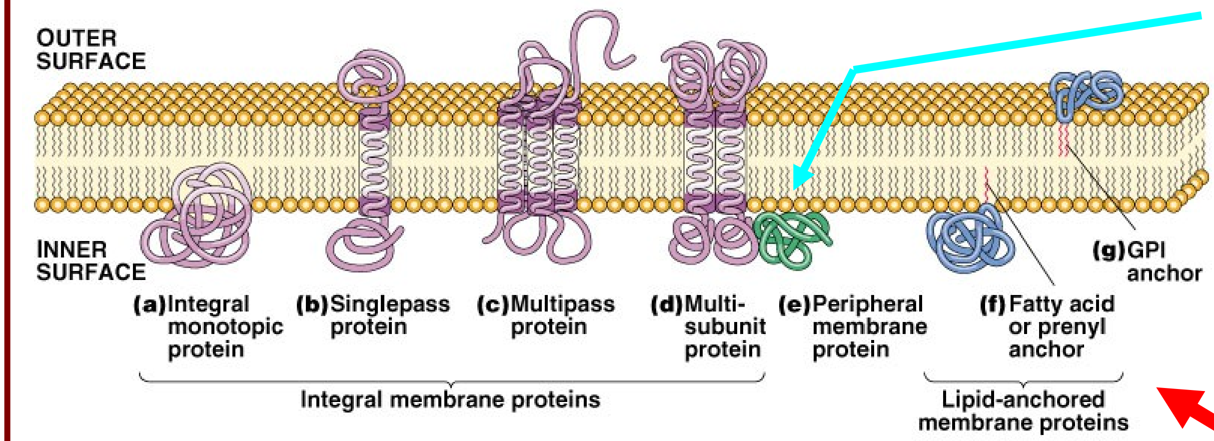




(a) Glycophorin (b) Bacteriorhodopsin










**Peripheral membrane proteins** are attached to membrane surfaces by weak electrostatic forces or hydrogen bonds. Connection with hydrophilic moieties of integrant proteins or polar head portions of membrane lipids. E.g.: spectrin and ankyrin in RBC plasma membrane.

**Lipid-bound membrane proteins**: covalent bond with lipid.

- Fatty acid or prenyl as an anchor
- GPI-switching (glycosylphosphatidyinositol, GPI)

**The proteins can be separated by SDS-polyacrylamide gel electrophoresis**

**Isolation of membrane proteins**

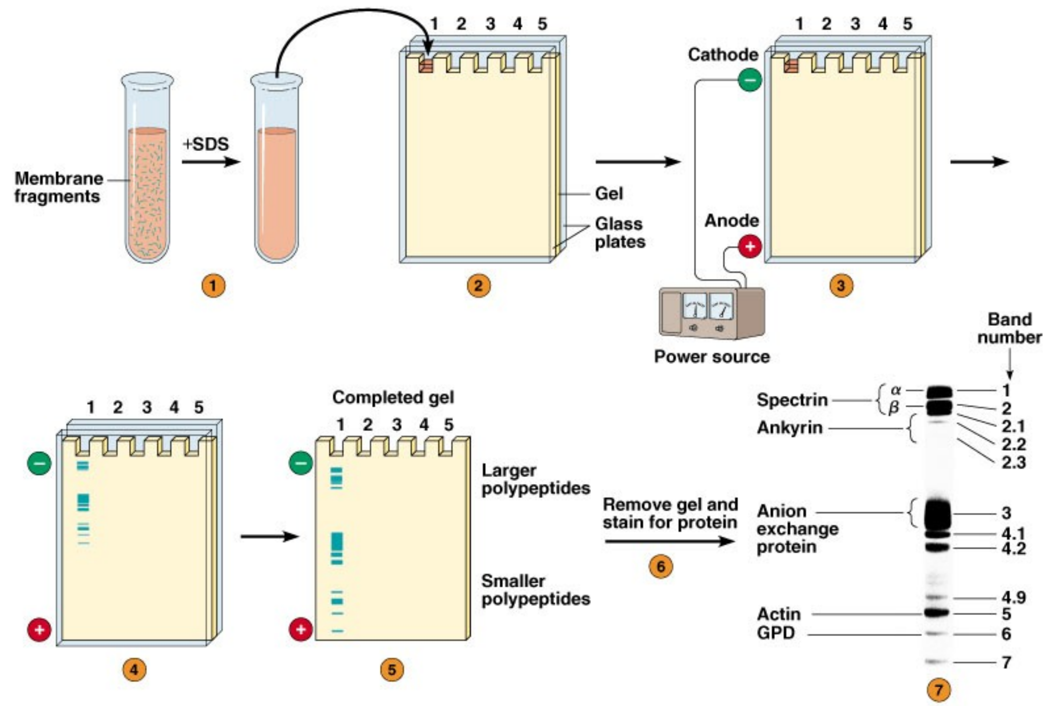
Peripheral membrane proteins: altering **pH** or **ionic bonds**, removing **calcium** -chelating agents (**cationic bonds**), or urea (breaking down **hydrogen bonds**).

**Lipid-bound proteins** can be isolated in the same way as peripheral proteins, since **covalent bonds must also be cleaved..**

Extraction of **integral membrane proteins** is a more difficult, **detergent-intensive task**. The detergent breaks down the **hydrophobic bonds** and interacts with the lipid bilayer. SDS (sodium dodecyl sulfate) treatment

**SDS-polyacrylamide gel electrophoresis**: The use of an electrophoretic electric field to separate electrically charged molecules. The migration speed depends on the charge and size.

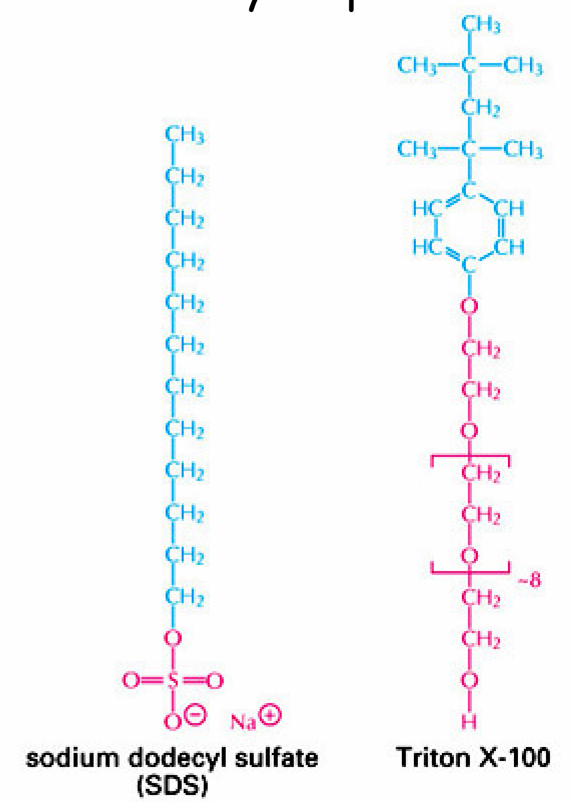
SDS digestion (proteins coated with negatively charged SDS) -> electrophoresis -> staining with Coomassie brilliant blue.



# Isolation and assay of membrane proteins:

The detergents used mimics the phospholipid structure around proteins

hydrophilic

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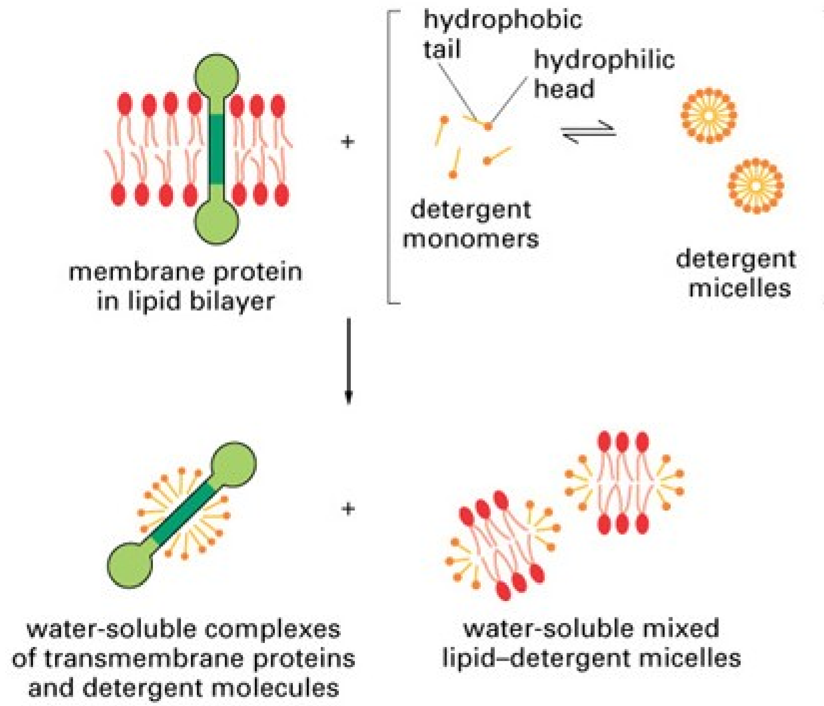
hydrophobic

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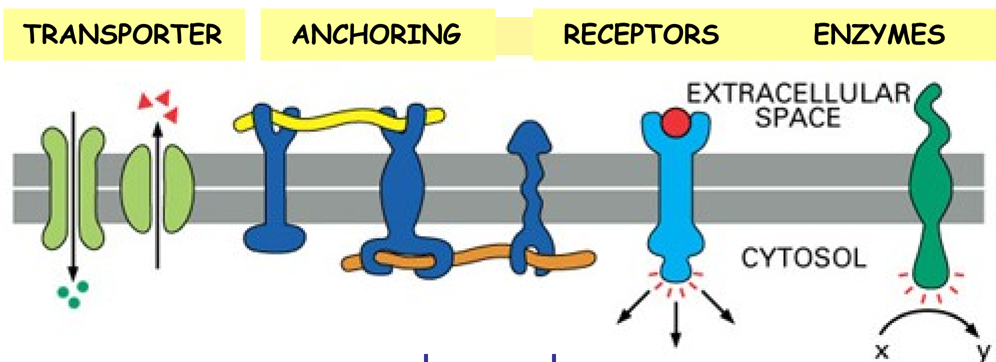
# Membrane proteins can perform many functions

**<u>Enzyme:</u>** E.g.: glucose phsphatase (ER-assocated) and glycerolaldehyde-3-phosphate dehydrogenase (GPD), peripheral proteins of RBCs invoolved in the glucose metablism of blood.

**<u>Transport proteins</u>** e.g. transport ATPases use energy of ATP to transfer ions across membrane.

**<u>Receptors:</u>** e.g. hormones and growth factors

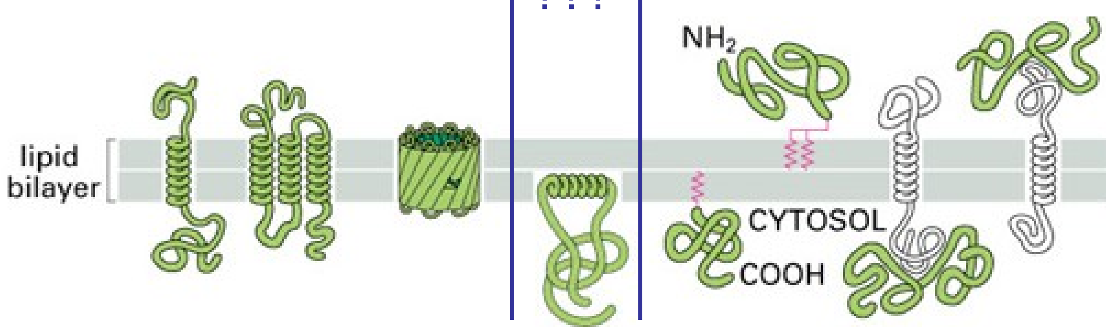
**<u>Other:</u>** endocytosis and exocytosis; targeting, sorting, modification of proteins in ER or Golgi; light perception; stabilization of cell membrane structure and determination of shape (ankyrin, spectrin).

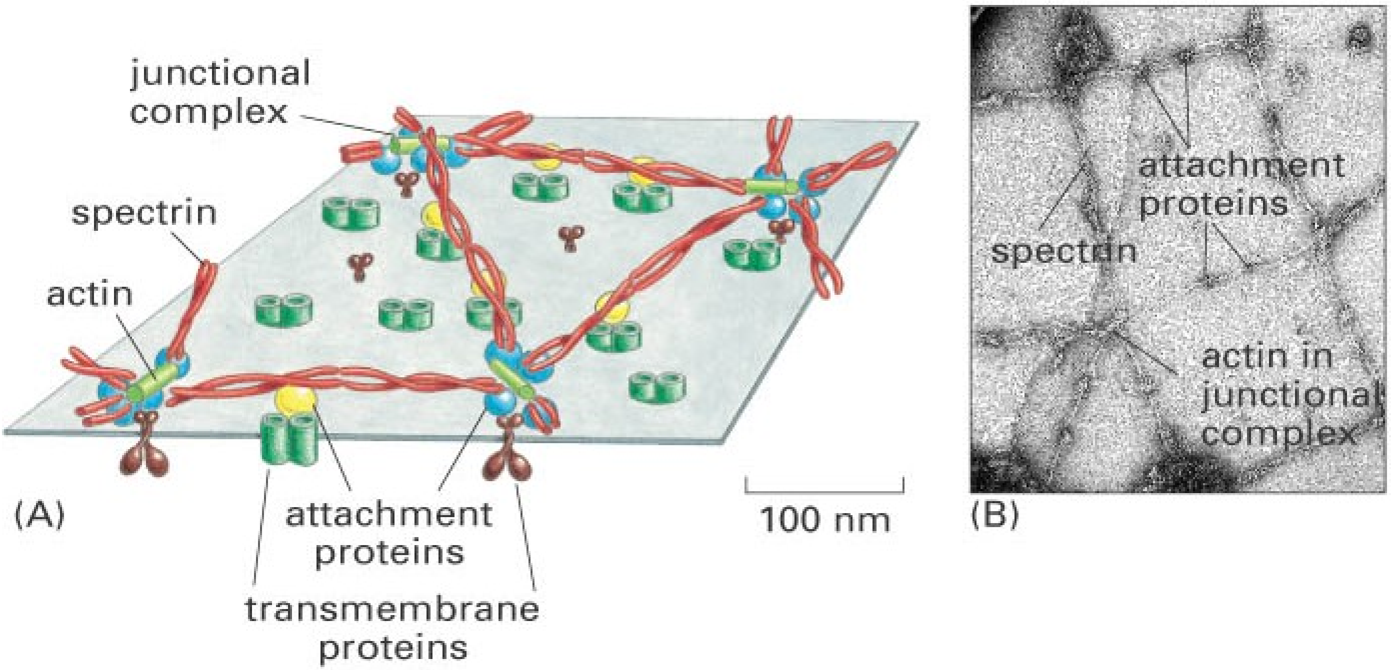



**Transmembrane (intergrant)**
**Single-pass Multi-pass**

???

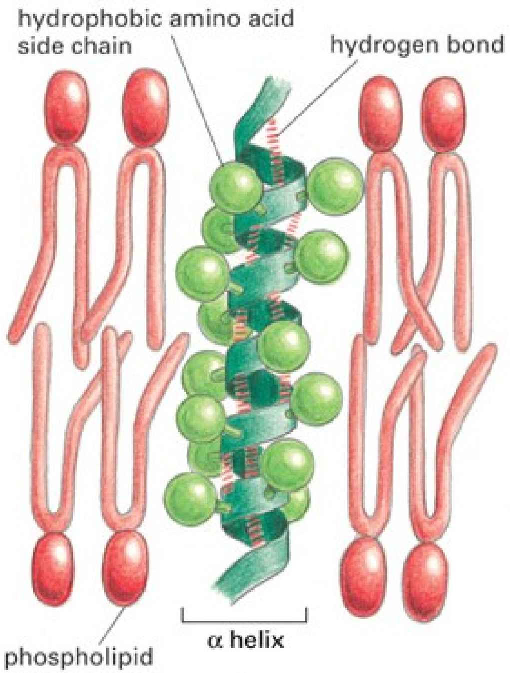
**Membrán-associated (periferial)**
**Covalently bound vs non-covalent**



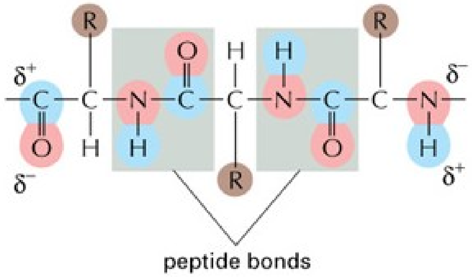

junctional
complex
spectrin
actin
(A)
attachment
proteins
transmembrane
proteins
100 nm
attachment
proteins
spectrin
actin in
junctional
complex
(B)

# Embedding of membrane proteins in bilayer

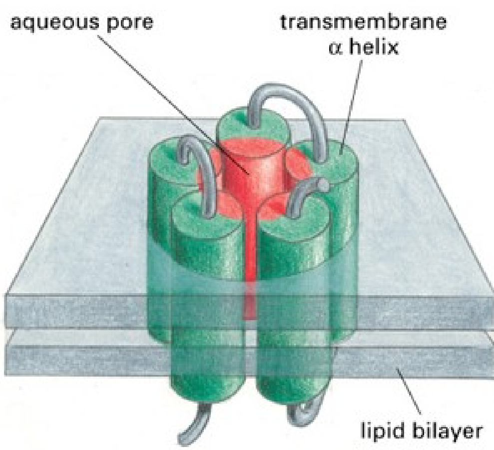
The **a-helical** structure is very common.
This is because the expression of the **hydrophobic amino acids** facilitates the anchoring of proteins to the hydrophobic layer of the membranes expressed.




logy, 2/e. (© 2004 Garland Science)




The a-helical structures **hydrophilic** components a are important in **pore formation**



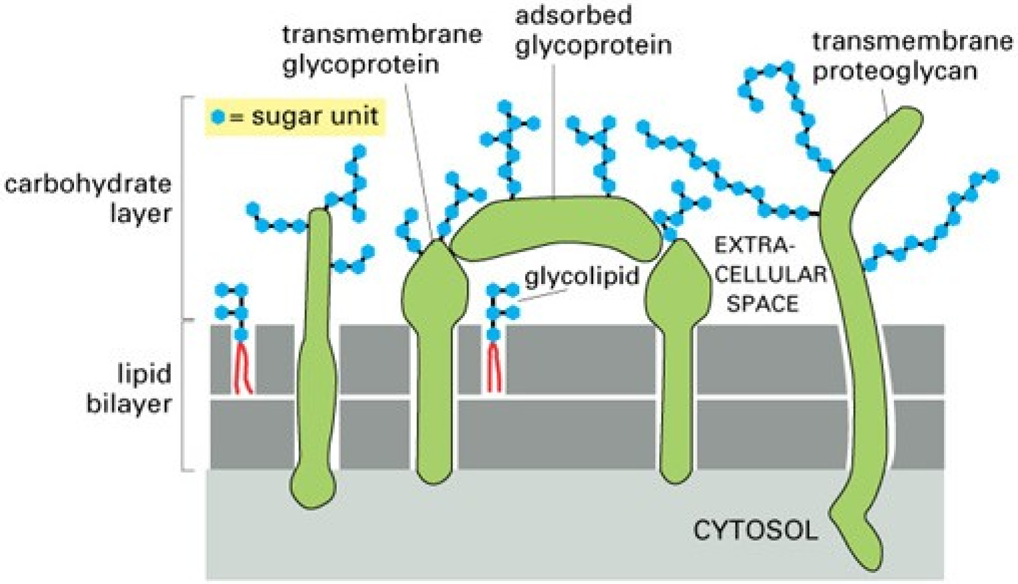
# The carbohydrate components of the cell membrane

<u>Functions:</u>

Surface protection

Recognition

Cell adhesion -
- extracellular matrix



**Most of the membrane proteins are glycosylated**

Human RBC membrane: 52% proteins, 40% lipids and 8% carbohydrates.

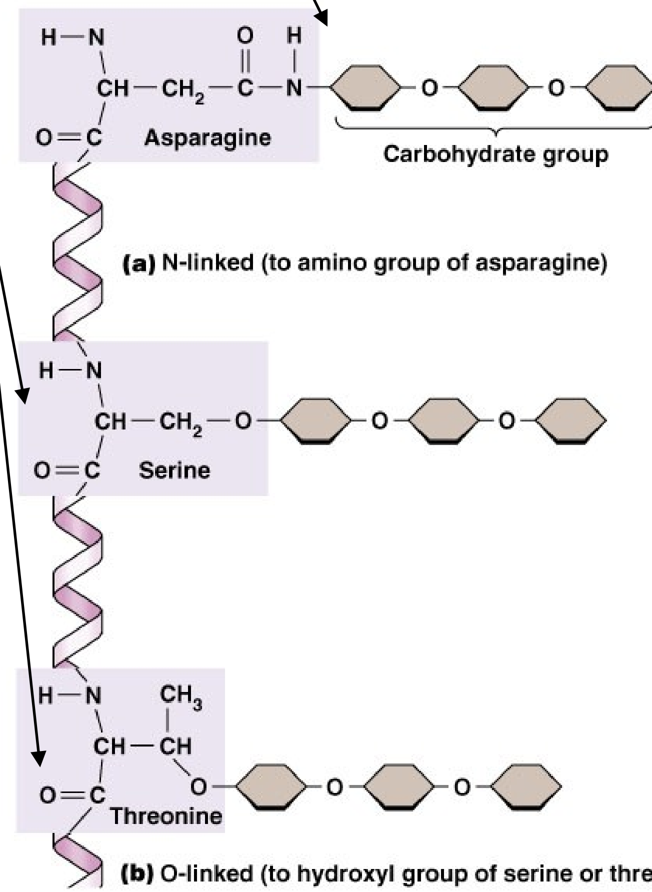
Carbohydrates are found in glycolipid (rare) and glycoprotein (common) in the membrane

**Glycoproteins:**

They are N-linked through the amino group of the Asp side chain

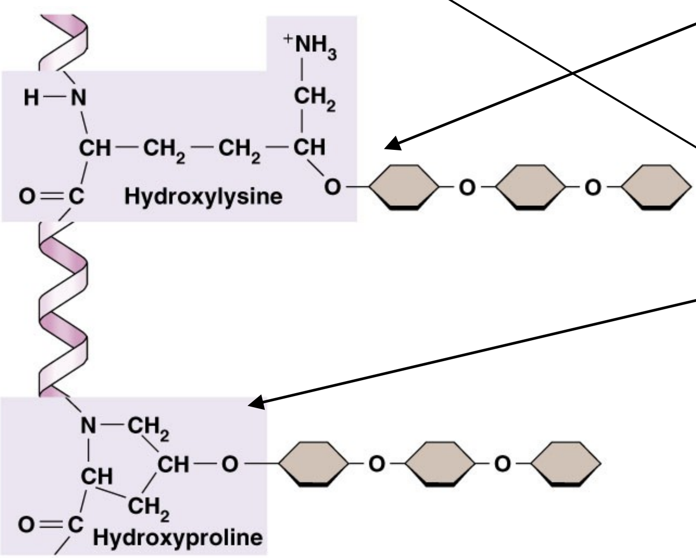
O-linked - through hydroxyl grup of Ser orThr

Some O-linked carbohydrates are linked via the hydroxyl group of hydroxy-Lys or hydroxy-Pro




(b) O-linked (to hydroxyl group of serine or threonine)



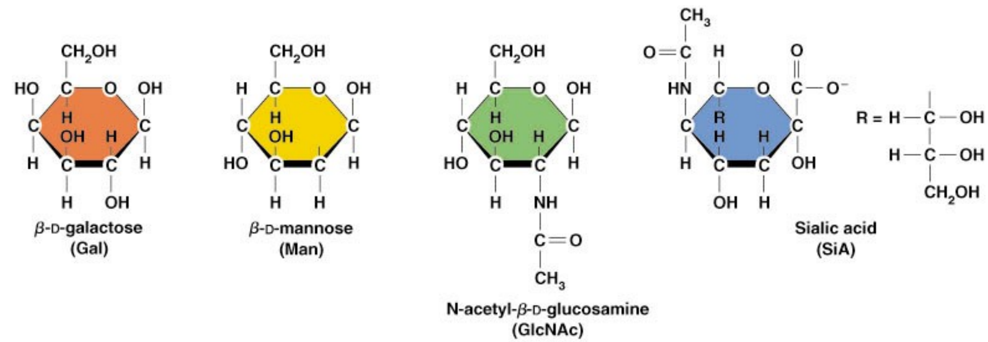



(c) O-linked (to hydroxyl group of hydroxylysine or hydroxyproline)

Length of protein-bound carbohydrates: **2-60 units**, straight v. linear

Common building blocks: **galactose, mannose, N-acetylglucosamine** and **sialic acid**

The integral protein (glycophorin) of RBC has a negatively charged **sialic acid** at its terminal position (15 O-linked and 1 N-linked), which reduces the tendency of RBCs to adhere and to **reduce blood viscosity**.



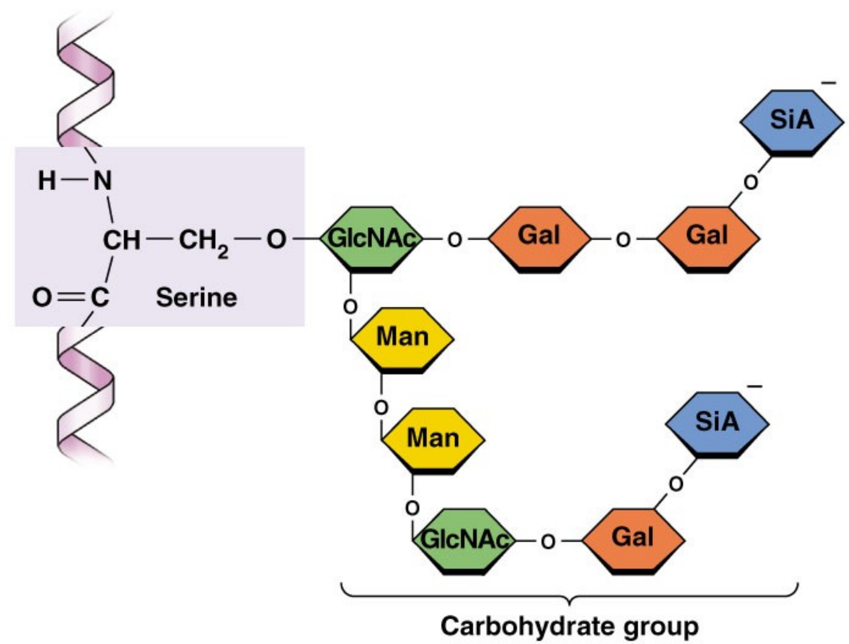

**(a)** Common sugars found in glycoproteins



Glycoproteins play a primary role in the **cell-cell recognition** function of cell membranes

**Experiment:**

**Lectins:** plant (animal) proteins with sugar-specific bonds. E.g.: wheat germ agglutinin (wheat) binds oligsaccharides with terminal N-acetylglucoseamine; the **concanavalin** is isolated from „jack bean" and binds mannoses of oligosaccharides. **Ferritin** is an iron cntaining protein which is bound to glycoproteins – it is easy to detect its binding due to its high electronedensity.

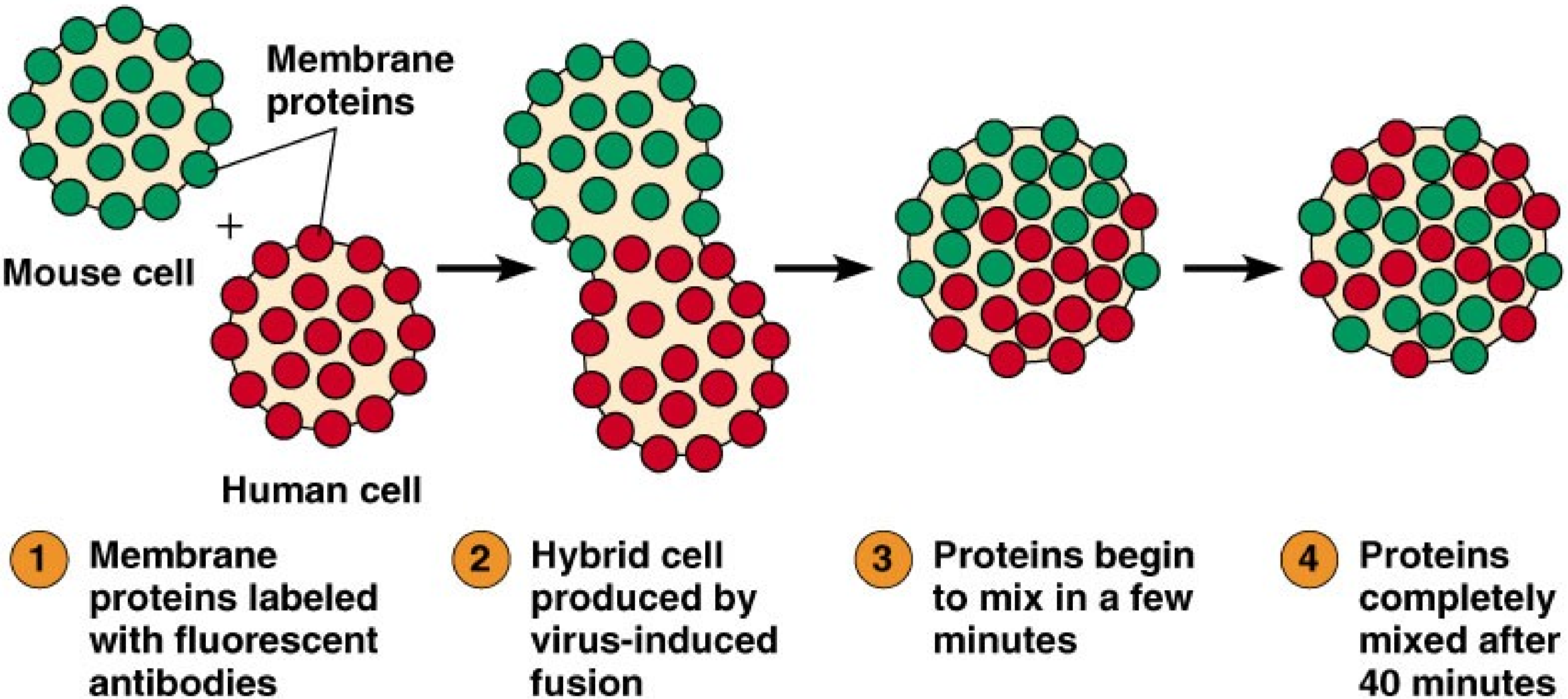



**(b)** The carbohydrate group of glycophorin

# Mobility of membrane proteins can vary widely

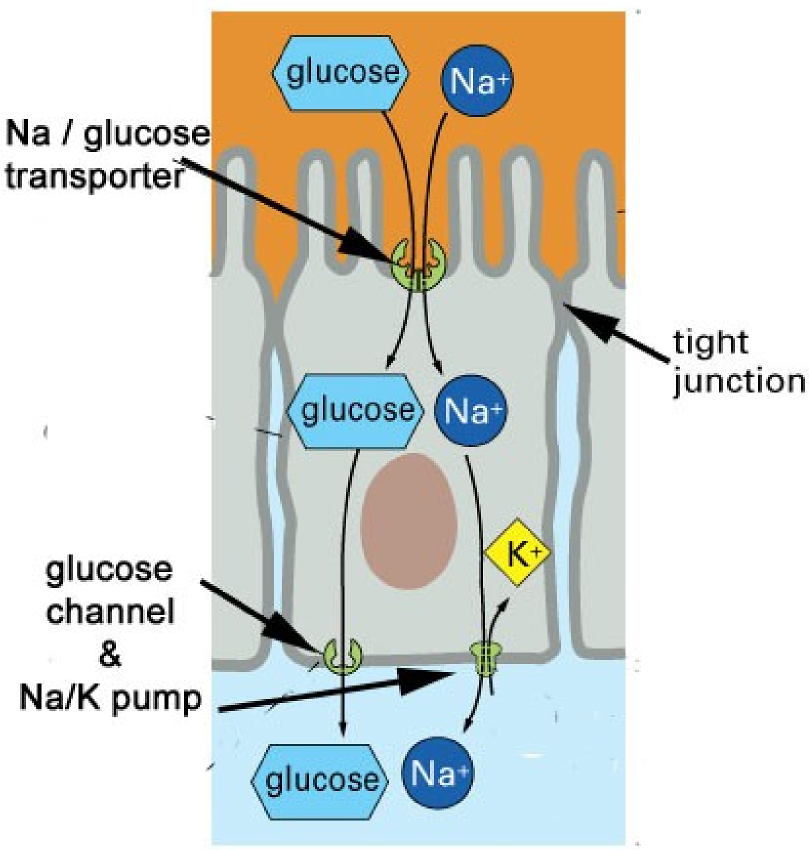
Some proteins move freely while others bind.

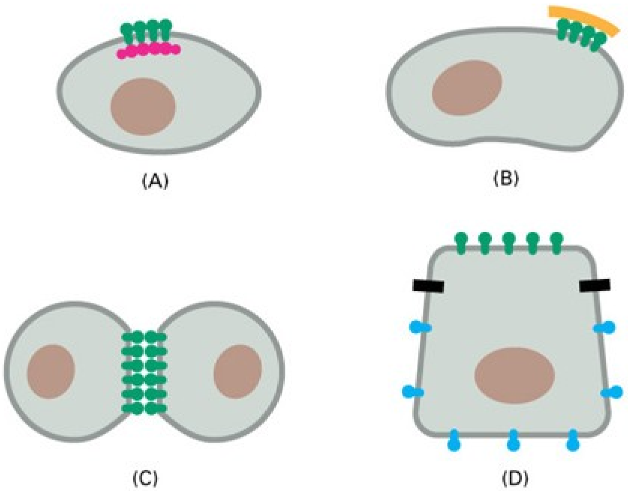
Technique used to detect mobility: cell fusion

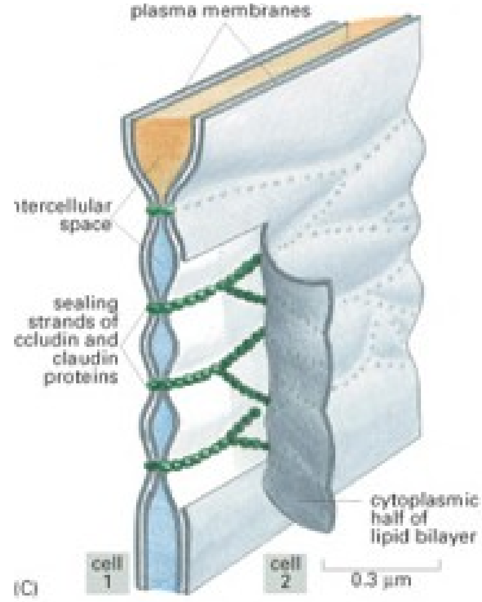


# Cell membrane protein movement is restricted :

E.g. polarized surface of intestinal epithelial cells or glandular epithelium

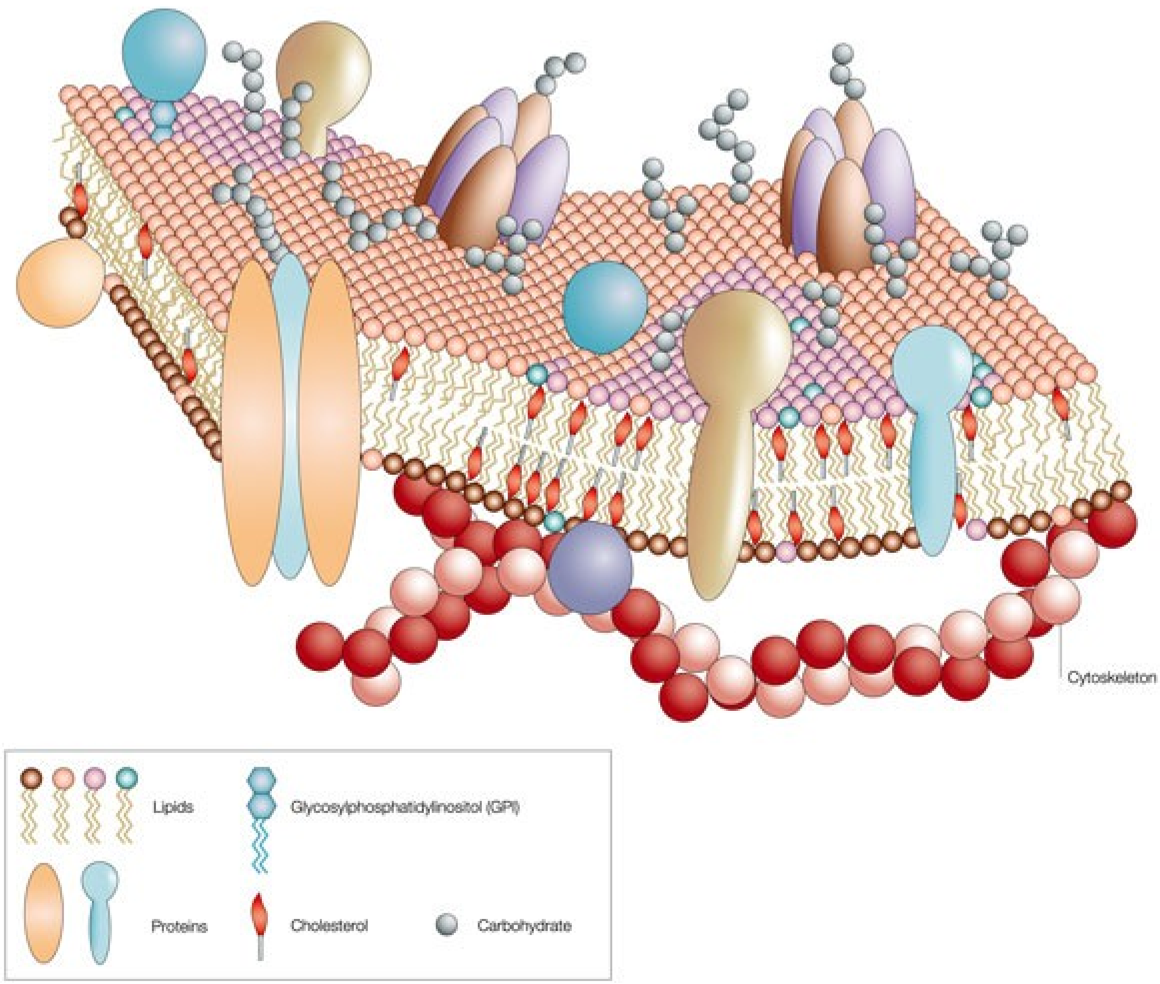


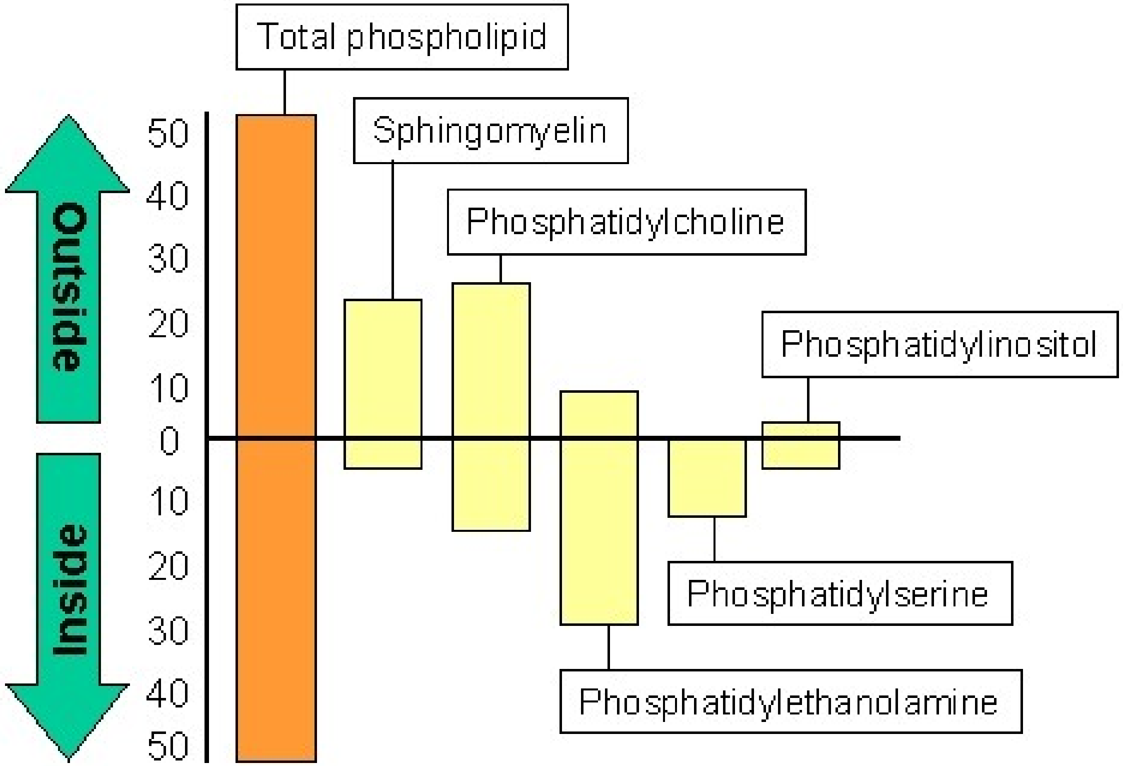



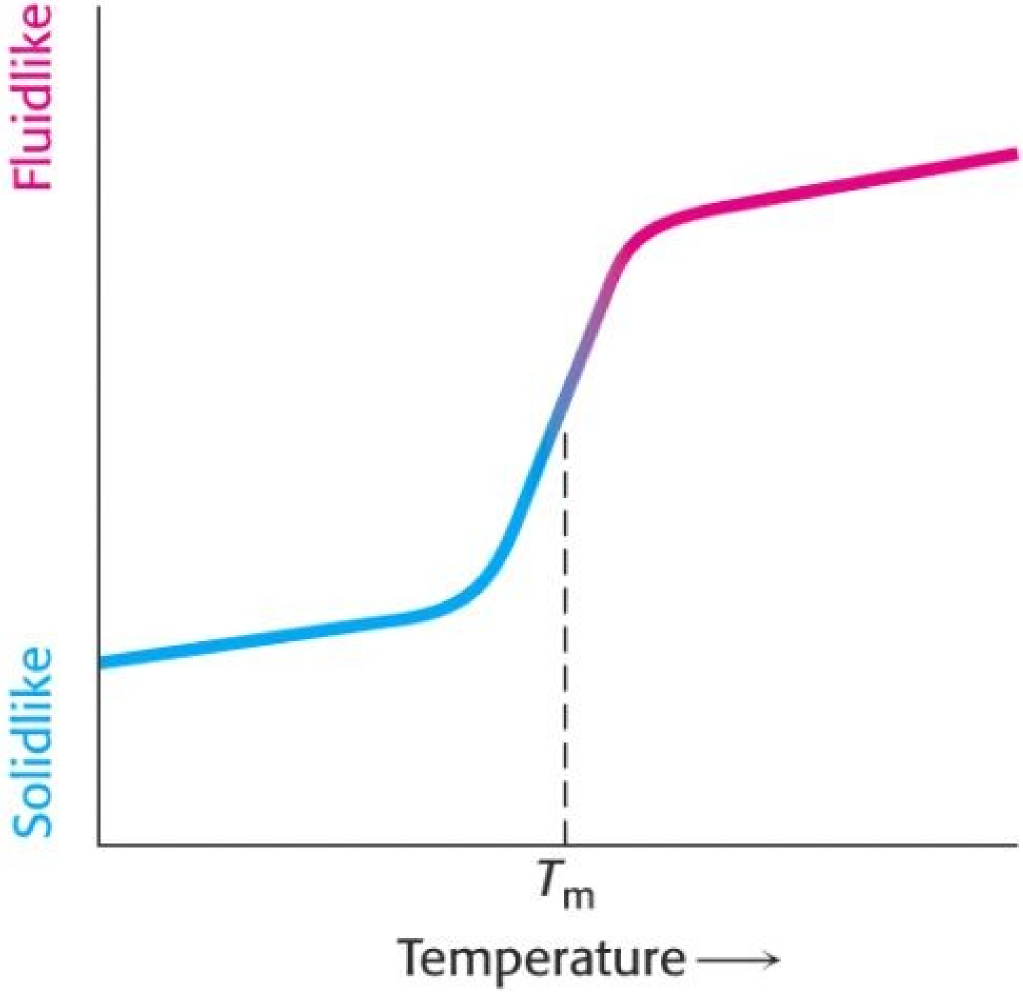

Polarized location, appearance of receptors, transporters, cell coupling structures .

# Take home message



Cytoskeleton
Lipids
Glycosylphosphatidylinositol (GPI)
Proteins
Cholesterol
Carbohydrate

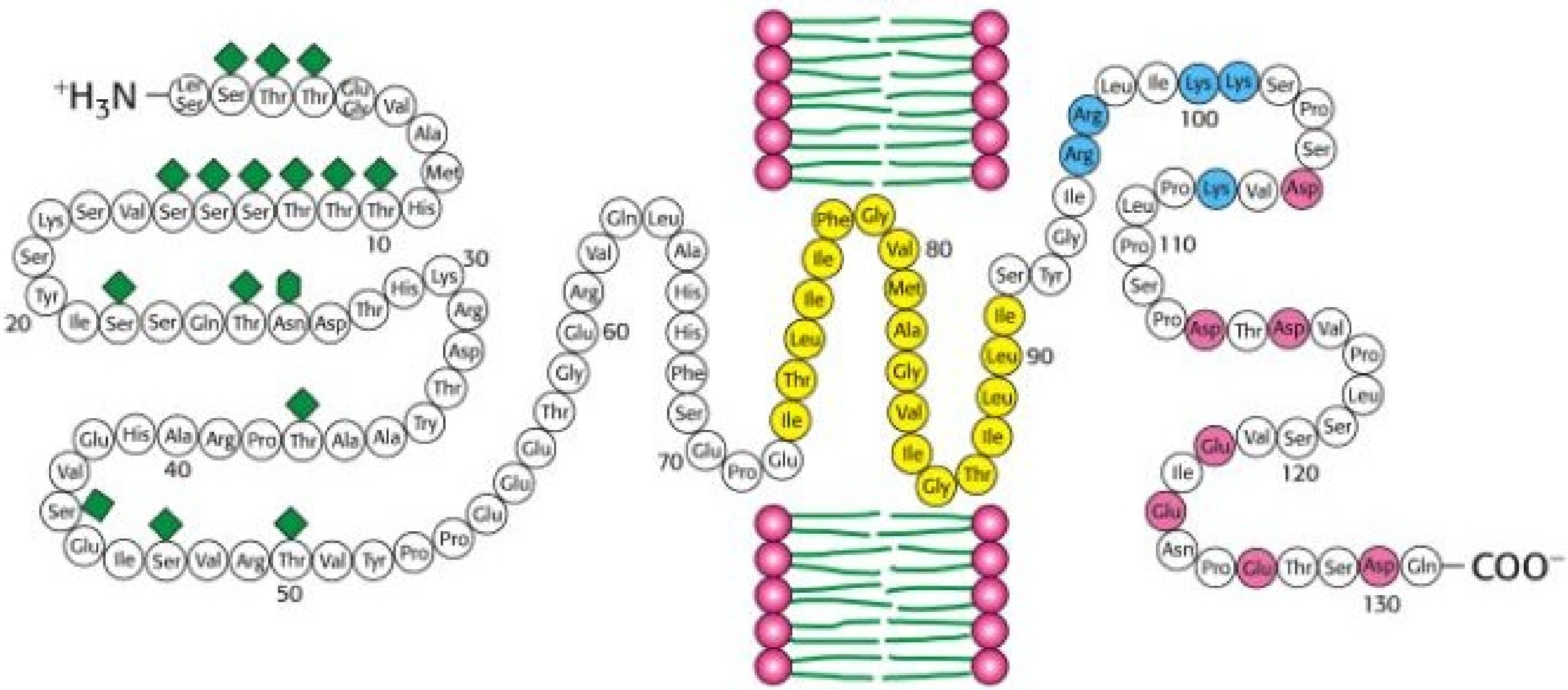

Total phospholipid
Sphingomyelin
Phosphatidylcholine
Phosphatidylinositol
Phosphatidylserine
Phosphatidylethanolamine
Outside
Inside
50
40
30
20
10
0
10
20
30
40
50

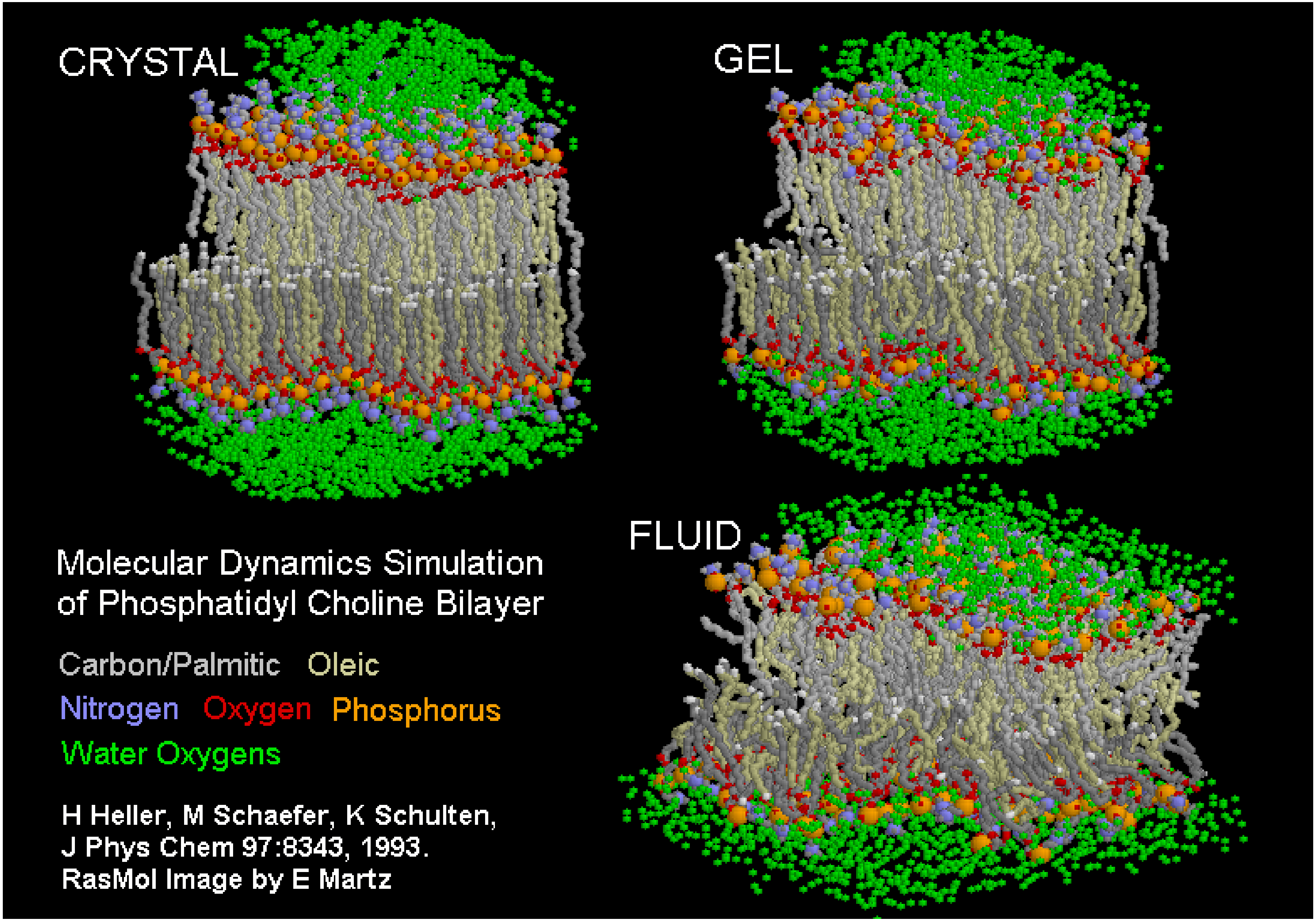


Fluidlike
Solidlike
$T_m$
Temperature ⟶

Extracellular

Intracellular




CRYSTAL
GEL
FLUID
Molecular Dynamics Simulation
of Phosphatidyl Choline Bilayer
Carbon/Palmitic Oleic
Nitrogen Oxygen Phosphorus
Water Oxygens
H Heller, M Schaefer, K Schulten,
J Phys Chem 97:8343, 1993.
RasMol Image by E Martz

**Reference:**

**Hardin, J., Kleinsmith, L., Becker, W.: World of the cell with free solutions, (Benjamin Cummings, 2002)**

**Alberts, B., Johnson, A., Lew, J.: Essential Cell Biology, (Garland, 2004)**