

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ
(وَفَوْقَ كُلِّ ذِي عِلْمٍ عَلِيمٌ)



Cytology & Molecular Biology | Lecture 1

Introduction & Endoplasmic Reticulum



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Me!

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- Come in groups

Course outline (1) **Cell Biology Part**

Focus on diseases

- Introduction and biomembranes
- Endoplasmic reticulum and protein sorting
- Golgi apparatus
- Vesicular network
- Mitochondria and mitochondrial diseases
- Peroxisomes
- The nucleus
- Cytoskeletal networks
- The extracellular network
- Cell signaling, proliferation, differentiation, and death
- The biology of cancer

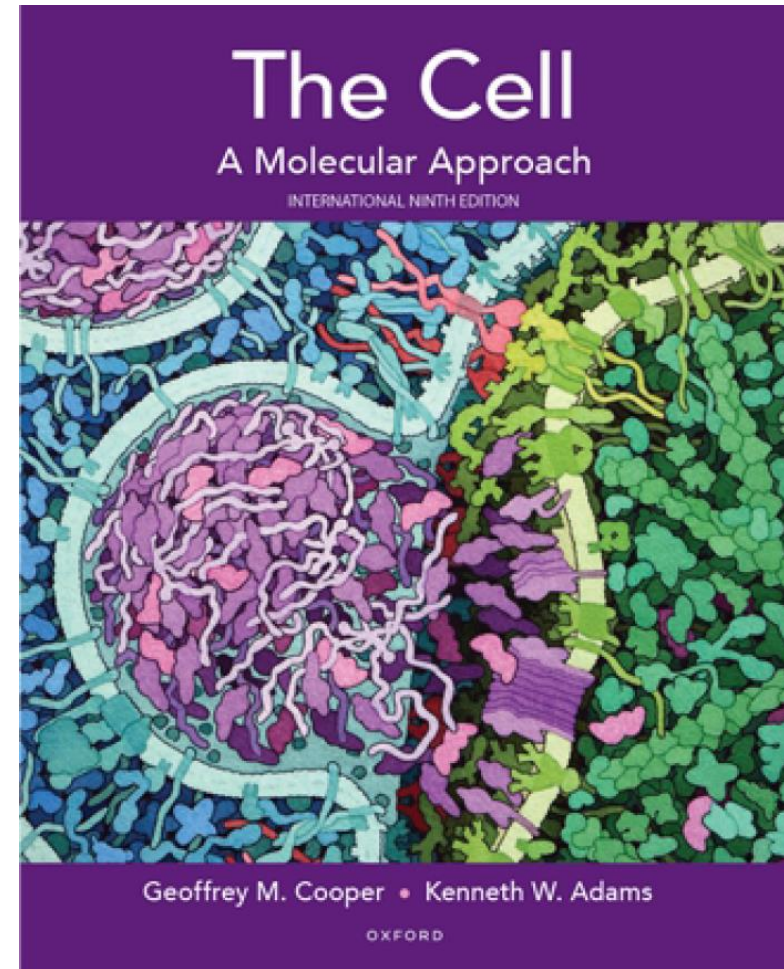
Course outline (2) **Molecular Biology Part**

**Focus on
processes
and
techniques**

- Overview of nucleic acids and gel electrophoresis
- The concept and utilization of the (de/re)naturaton concepts
 - Dot blotting and Southern blotting
- Restriction endonucleases, recombinant DNA technology, DNA cloning, and RFLP
- The central dogma of molecular biology DNA replication
- PCR and DNA sequencing
- The human genome
- Transcription, mechanisms of regulation, and epigenetics
 - Coding and non-coding RNAs
- RNA detection, quantification, and detection
- Translation
- Yeast two-hybrid system
- DNA mutations
- DNA repair and CRISPR-Cas9

The textbook

- **The Cell: A Molecular Approach**, [Geoffrey M. Cooper](#) and Kenneth W. Adams, 9th edition, Sinauer Associates, 2023.



The cell

➤ we classify living creatures to three kingdoms :

1) **Eukaryotes** (true nucleus and nuclear membrane).

2) **Prokaryotes** (simply bacteria , don't have nucleus or nuclear membrane or any membrane bound organelle).

3) **Archaea** (something in between , they have prokaryotic and eukaryotic features , some of them exist in the dead sea).

We will focus on **eukaryotic cells and its organelles** .

Mitochondria : plural
Mitochondrion : single

➤ In addition to nucleus , Eukaryotic cells has :

1) Mitochondrion.

2) Lysosomes.

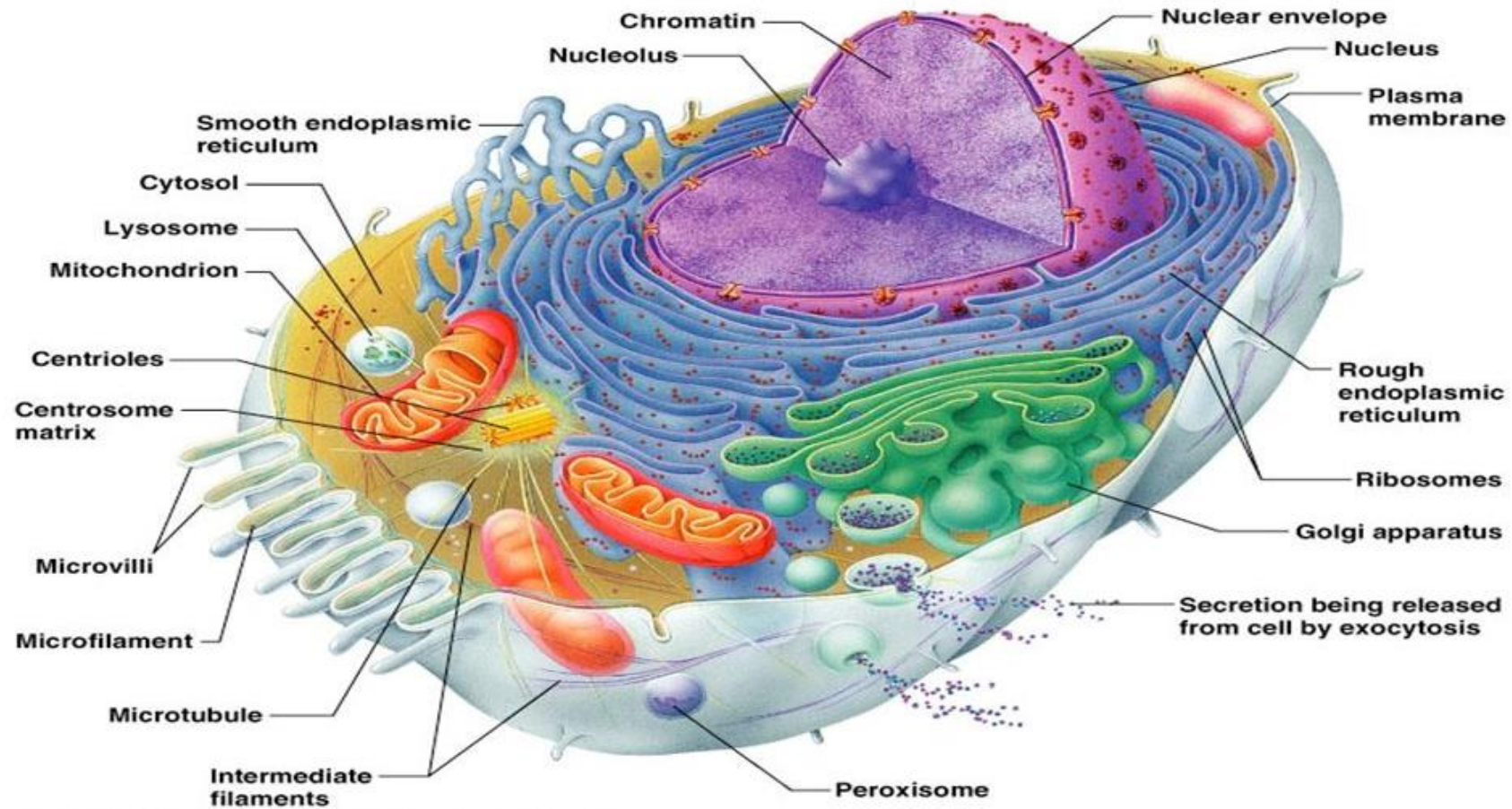
3) Golgi apparatus.

4) Endoplasmic reticulum.

5) Peroxisomes.

And many other structures (as shown in the next slide).

The cell



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What organisms do we use to study cells?

The idea is that we want to study human cells, but we can't do experiments on human because it is not ethical.

We use models (which are organisms) to study and understand cells.

- Escherichia coli (E. coli)

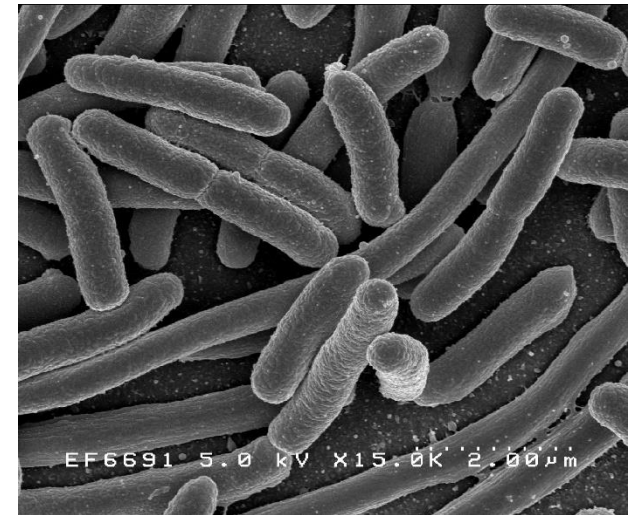
- Bacteria, why? Because :

- 1) its **molecular mechanisms are similar to molecular mechanisms of human cells.**

- 2) **unicellular.**

So by simple cells I can understand complex systems like human cells.

- Exist in rotten meat it can be fatal.

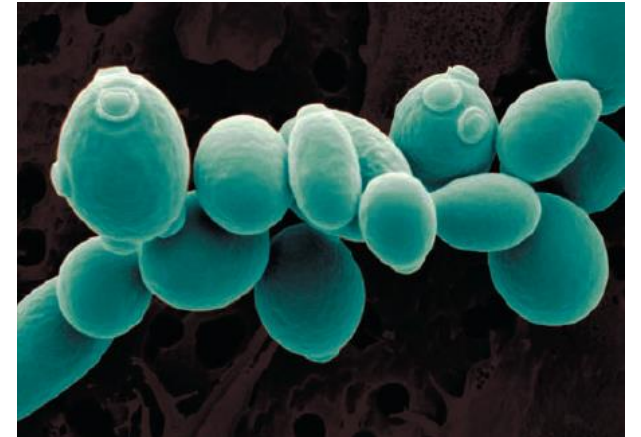


- Yeast (*Saccharomyces cerevisiae*)

What makes Yeast good as a model for understanding human cells that :

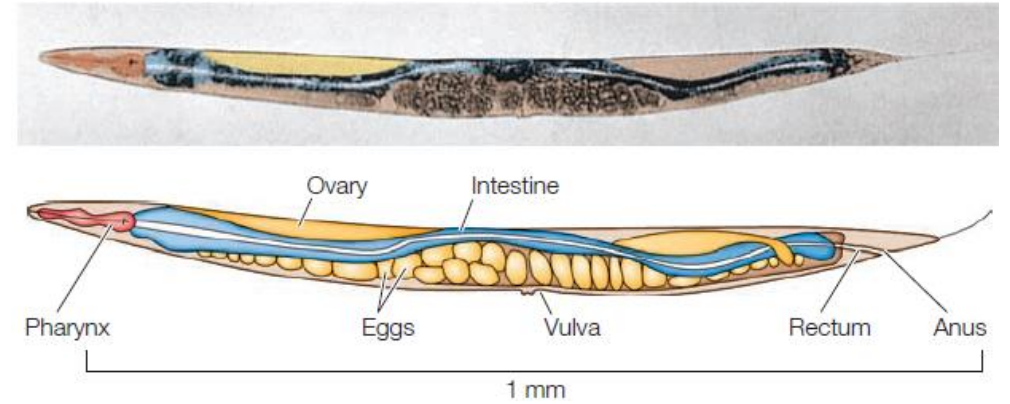
- They are **eukaryotes cells** : have nucleus and nuclear membrane.
- their **molecular system is complex** like humans (the same cellular mechanism in both).
- they have **multiple and linear chromosomes** (like humans) but in bacteria it is single and circular.
- **unicellular** (so easier to study).
- as an example (*saccharomyces cerevisiae*).

But there is problem that they **aren't differentiable** so there is not nervous or digestive system etc.



- **Caenorhabditis elegans (C-Elegans)**

- it is a microscopic worm
- it is complex **multicellular systems** (opposite to yeast, its cells are **differentiable**).
- It has (1079) cells (we know every single cell and its function).
- when it matures they become (952) cells.
The decline of cells because of **cell programmed death** which use **the same proteins genes and mechanisms** in human body and then could be easily studied by this worm.



- **Drosophila melanogaster**: (ذبابة الفاكهة)

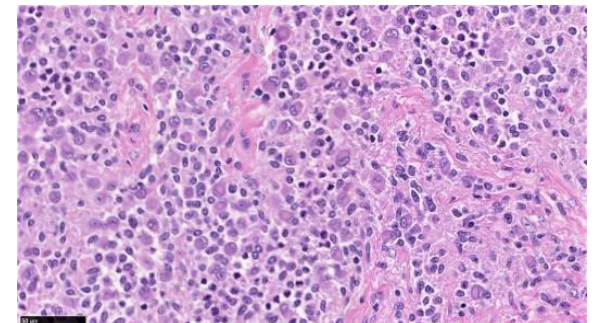
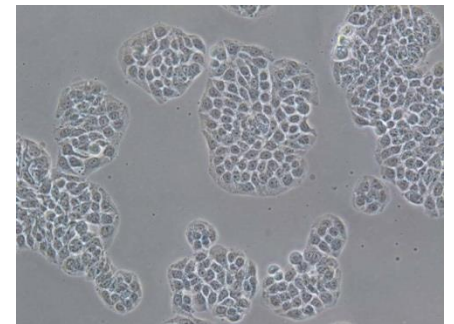
- **Short live** (few days).
- **Really complex** (it has ears ,eyes), but still easily to study compare to other big creatures.
- The bases of genetics science (inheritance pattern) based on this insect.



- Mice (more complicated)
 - Mice better than monkeys, rabbits and other animals because they reproduce very fast.
 - Mice have the same number of genes as in humans but different regulation mechanisms gives completely different creatures.



- Cultured cells and tissues (specifically human cells)
 - We isolate these cells from a human and then grow them. The problem here is they do not exist in a real system to work effectively (isolated from other kinds of cells).
 - Also we could use tissue sections like in histology using a lot of methods like immunohistochemistry.



So we have understood the human cells by these models.

Nucleus

Nuclear envelope: membrane enclosing the nucleus. Protein-lined pores allow material to move in and out.

Chromatin: DNA plus associated proteins.

Nucleolus: condensed region where ribosomes are formed.

Peroxisome: metabolizes waste

Endoplasmic reticulum

Rough: associated with ribosomes; makes secretory and membrane proteins.

Smooth: makes lipids.

Cytoskeleton

Microtubules: form the mitotic spindle and maintain cell shape.

Centrosome: microtubule-organizing center.

Intermediate filaments: fibrous proteins that hold organelles in place.

Microfilaments: fibrous proteins; form the cellular cortex.

Plasma membrane

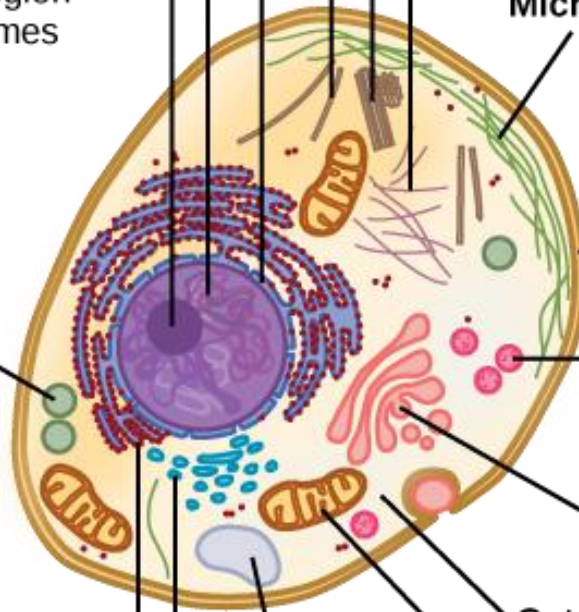
Lysosome: digests food and waste materials.

Golgi apparatus: modifies proteins.

Cytoplasm

Mitochondria: produce energy.

Vacuole



For more understanding :

[Cell Cross Section | BioDigital Human Platform](#)

Organelles

TABLE 4-2 *Organelles*

Organelle	Function
Mitochondrion	transfers energy from organic compounds to ATP
Ribosome	organizes the synthesis of proteins
Endoplasmic reticulum (ER)	prepares proteins for export (rough ER); synthesizes steroids, regulates calcium levels, breaks down toxic substances (smooth ER)
Golgi apparatus	processes and packages substances produced by the cell
Lysosome	digests molecules, old organelles, and foreign substances
Microfilaments and microtubules	contribute to the support, movement, and division of cells
Cilia and flagella	propel cells through the environment; move materials over the cell surface
Nucleus	stores hereditary information in DNA; synthesizes RNA and ribosomes
Cell wall*	supports and protects the cell
Vacuole*	stores enzymes and waste products
Plastid*	stores food or pigments; one type (chloroplast) transfers energy from light to organic compounds

*Cell walls, large vacuoles, and plastids are found in the cells of plants and some other eukaryotes, but not in the cells of animals.

Major molecular components of cells

➤ Macromolecules :

1. Nucleic acids
 2. Carbohydrates
 3. Proteins
 4. Lipids (50% of mass of plasma membranes, **40% proteins and about 10% sugar molecules**. 30% of mitochondrial membranes)
- Molecules function by interacting with each non-covalently.

Which governs the biochemical life of the cell. Ex : interaction between ligand and its receptor is non covalent interaction (electrostatic , hydrophobic, Hydrogen bond ..)

How do we study cell components?

Cell and protein detection

1. Microscopy
2. Cell Fractionation

Microscopy

Light Microscopy :

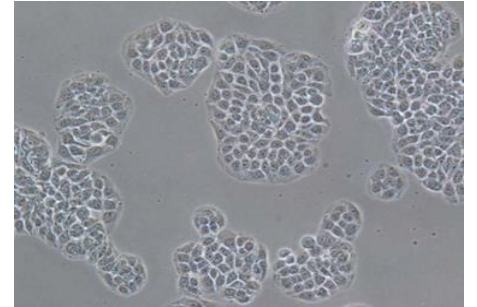
we can see the cell with different magnifications.

Immunofluorescence: antibodies (have specific tag molecule which makes the antibody fluorescent) that target specific proteins inside the cell so the protein lights up (this light comes from antibodies not proteins) **Electron:** zooming into specific organelles (peroxisomes, mitochondria..) with a high magnification. **Scanning electron:** 3D image that determine the cell and organelles shapes

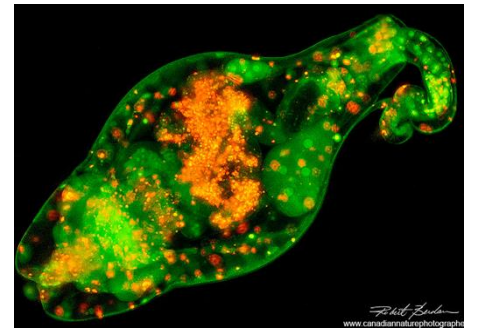
Electron :

it has high resolution so I can look at specific molecules like glycogen and DNA but not nucleic acids.

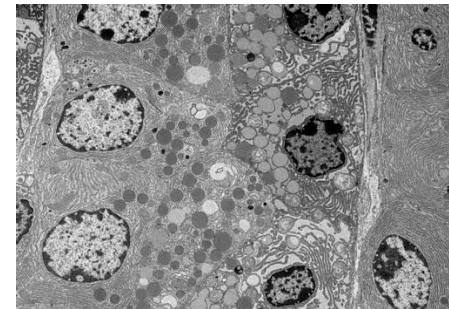
scanning electron microscopy which make 3D images of the cell and organelles.



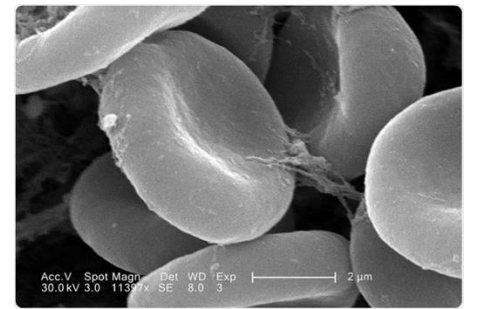
Light Microscopy Image



Immunofluorescence image



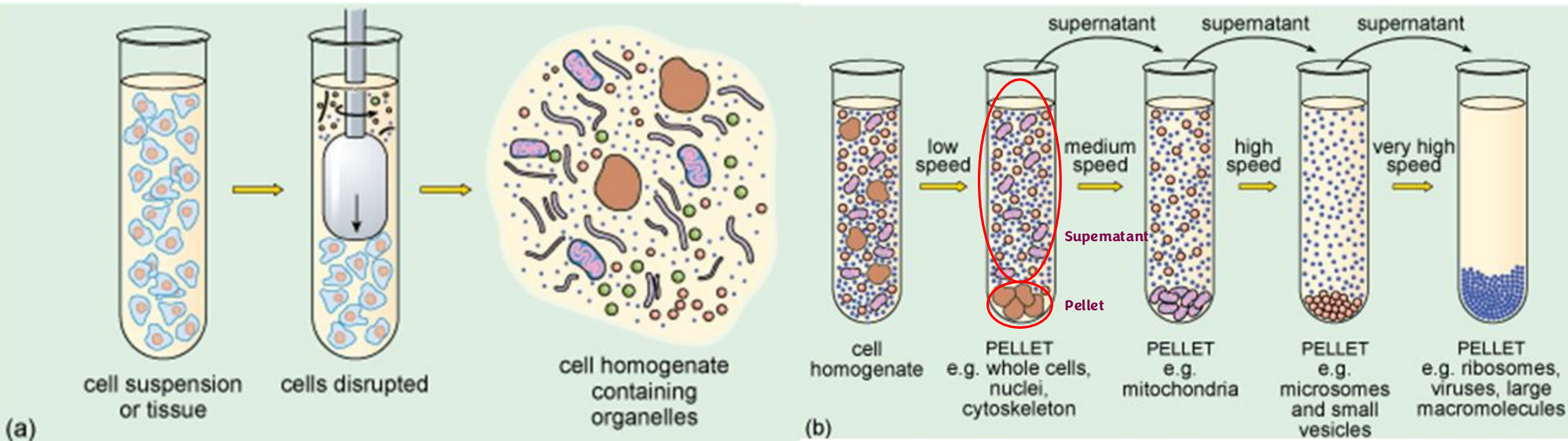
Electron microscope image.



Scanning electron microscope image.

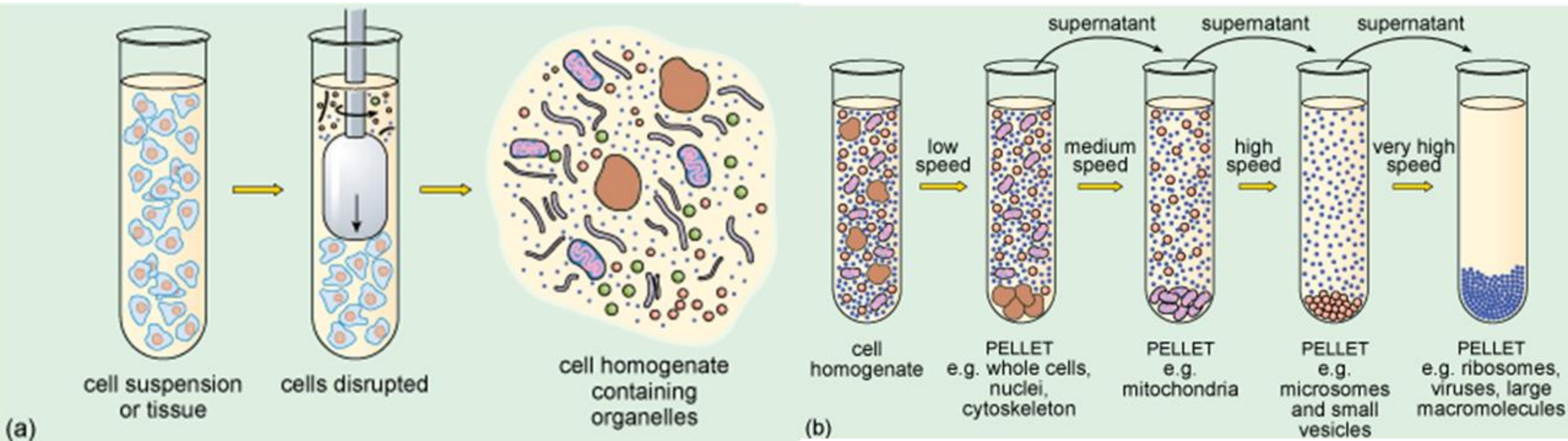
Cell Fractionation

When I want to study specific protein how I can isolate it? we break the cells and so far we have thousands of protein (from nucleus, mitochondria ...etc) , preproteins, lipids , and carbohydrates these sample now called **homogenate** then we do more processing to have simpler sample (proteins from the same organelles) so now **we use fractionation by centrifugation** the sample spin at different speeds by this **heavy things sink (pellet)** and **light things will be in liquid above and we call this liquid (supernatant)**.



Cell Fractionation

The speed is being increased progressively at low speed the whole unbroken cells sink and nuclei and cytoskeleton then we take the liquid (supernatant) to other tube and increase the speed (medium 10000 revolution per second) there will be new pellet consist of mitochondria by this now I have mitochondrial protein alone then I return the last step but with high speed the pellet will have vesicle organelles like lysosome and endosomes the return the step and use really high speed 100000 revolutions smaller things will make the pellet like ribosomes, macromolecule for example glycogen in this step is in the supernatant will have nutrients, like glucose fructose , small proteins.



<https://www.facebook.com/reel/792265814706206>



Biochemical composition of plasma membranes

Membrane	Protein (%)	Lipid (%)	Carbohydrate (%)	Weight fraction of protein	Ratio of protein to lipid
Plasma membranes					
Myelin	18	79	3	0.18	0.23
Blood platelets	33—42	51—58	7.5	0.4	0.7
Mouse liver cells	46	54	2—4	0.46	0.85
Human erythrocytes	49	43	8	0.49	1.1
Amoeba	54	42	4	0.54	1.3
Rat liver cells	58	42	(5—10)*	0.58	1.4
HeLa cells	60	40	2.4	0.6	1.5
Nuclear envelope of rat liver cells	59	35	2.9	0.59	1.6
Retinal rods, bovine	51	49	4	0.51	1.0
Mitochondrial outer membrane	52	48	(2—4)*	0.52	1.1
Sarcoplasmic reticulum	67	33	—	0.67	2.0
Chloroplast lamellae, spinach	70	30	(6)*	0.7	2.3
Mitochondrial inner membrane	76	24	(1—2)*	0.76	3.2
Gram-positive bacteria	75	25	(10)*	0.75	3.0
<i>Halobacterium</i> purple membrane	75	25		0.75	3.0

Biochemical composition of plasma membranes

There are different type of membranes like plasma ,nuclear ,and mitochondrial membranes which is inner and outer and *there are different type of cells like immune cells, red blood cells , brain cells so we don't expect that they have the same composition of the membranes* for example with regard to function epithelial cells structure should be **solid, rough , strong** for protecting, while we expect that the rod cells or retinal cells membranes are viscous to have very fast signal go to last slide and look at **mitochondrial inner membrane** look at **proteins percent it should be high** because of **electron transport protein (76%)** but the **outer membrane (52%)** look at **erythrocytes also (50%)** look at **myeline lipids percent it is(79%)** the numbers not required but you have to know the principle.

Myelin	18	<u>79</u>	3	0.18	<u>0.23</u>
Mitochondrial inner membrane	<u>76</u>	24	(1—2)*	0.76	32
Retinal rods, bovine	51	49	4	0.51	1.0
Mitochondrial outer membrane	<u>52</u>	48	(2—4)*	0.52	<u>1.1</u>

Lipid composition of organelles

Table 1: Head group composition of the membranes of some mammalian liver cells, erythrocytes , and nerve cells in weight percent. Adapted from Jamieson and Robinson (1977). Abbreviations: PC = phosphatidylcholines, PE = phosphatidylethanolamines, PS = phosphatidylserines, PI = phosphatidylinositols, SM = sphingomyelin, CL = cardiolipin.

Membrane	PC	PE	PS	PI	SM	CL	Glycolipid	Cholesterol	Others
Erythrocyte (human)	20	18	7	3	18	–	3	20	11
Plasma (rat liver)	18	12	7	3	12	–	8	19	21
ER	48	19	4	8	5	–	tr	6	10
Golgi	25	9	3	5	7	–	0	8	43
Lysosome	23	13	–	6	23	≈ 5	–	14	16
Nuclear membrane	44	17	4	6	3	1	tr	10	15
Mitochondria	38	29	0	3	0	14	tr	3	13
Neurons	48	21	5	7	4	–	3	11	1
Myelin	11	17	9	1	8	–	20	28	6

Myelin a high amount of lipids (especially glycolipids for cell interaction & cholesterol for rigidity)

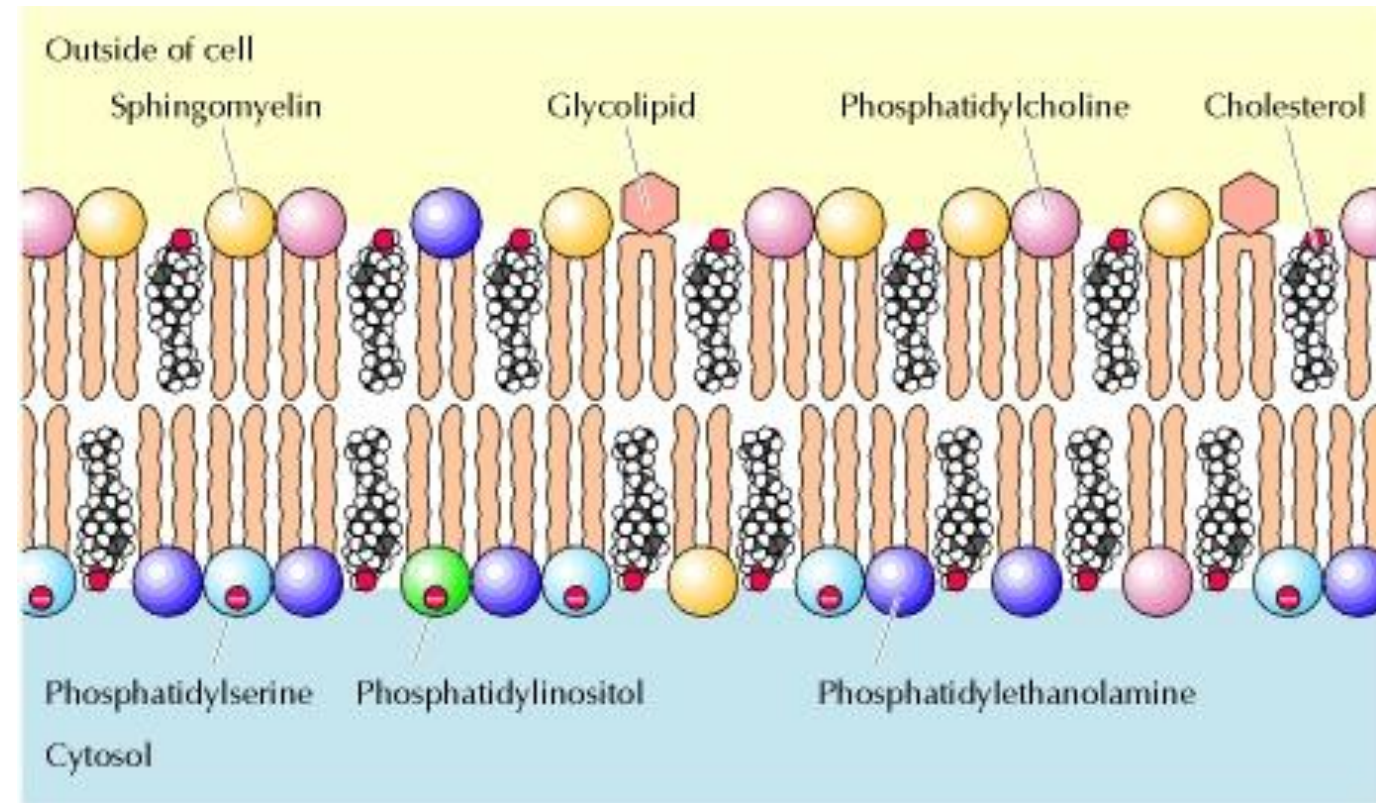
Look at myeline again it has high percent of glycolipids (20%) compare with other cells ,look at cholesterol present in myeline too , it is nearly (20%) , the idea is that even the percent of different types of lipids differ in different cells in order to fit function.

Cholesterol is an essential component of animal plasma membranes. It is not present in bacteria and plant cells, but the latter cells contain sterols.

Composition and properties of plasma membranes

- The phospholipids are asymmetrically distributed between the two halves of the membrane bilayer.
 - The outer leaflet: Ⓟcholine, **sphingomyelin**
 - The inner leaflet: Ⓟethanolamine, Ⓟserine, **Ⓟinositol (minor)**
 - Ⓟinositol has **a role in cell signaling. Secondary messenger**
 - Glycolipids are found exclusively on the outer membrane. Because of their function in cell signaling.

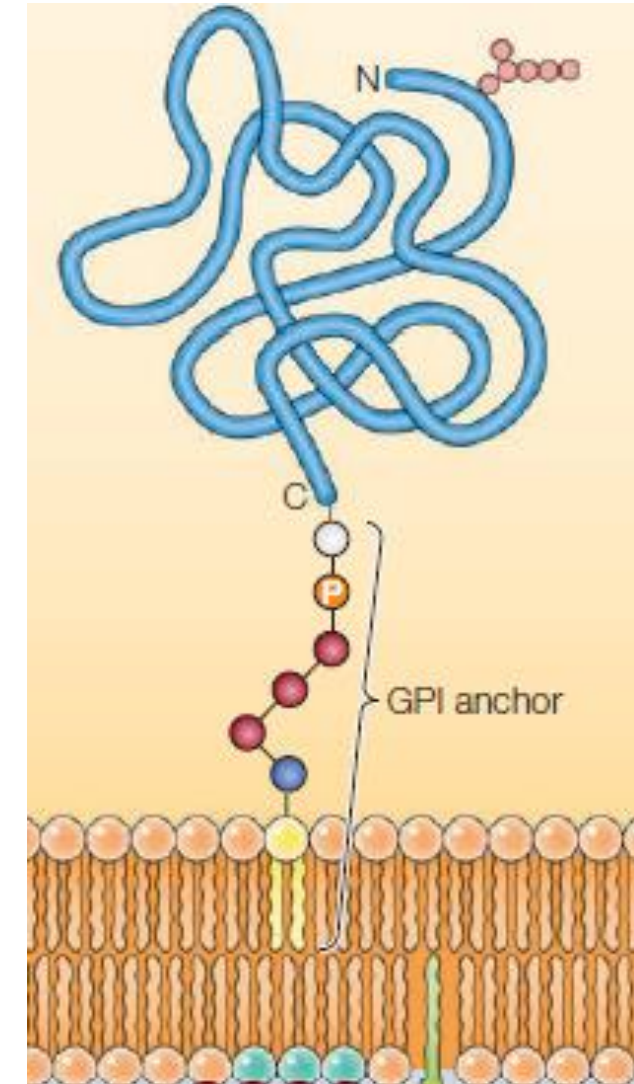
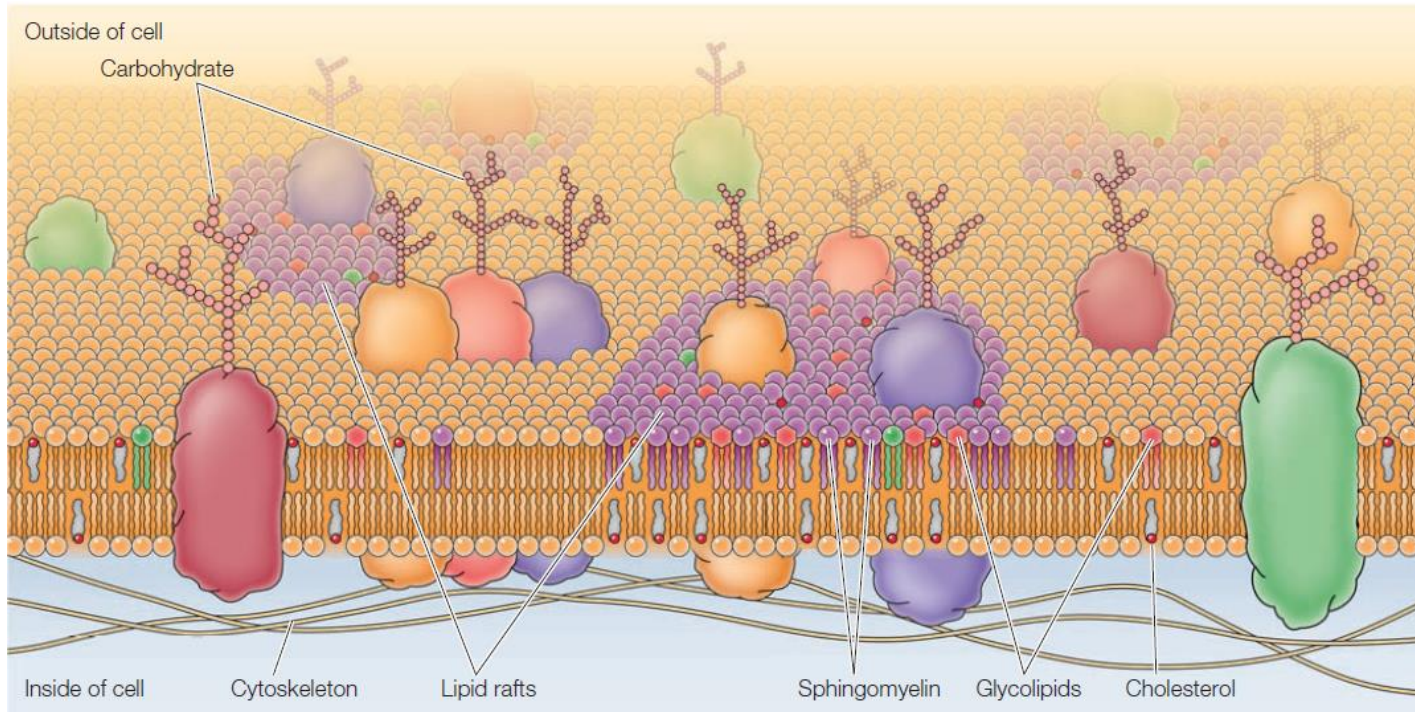
The cholesterol is equally distributed between outer leaflet and inner leaflet.



Even the phospholipids could differ by the head group that connected to phosphate group, it could be choline, ethanolamine, serine, inositol, sphingomyelin.

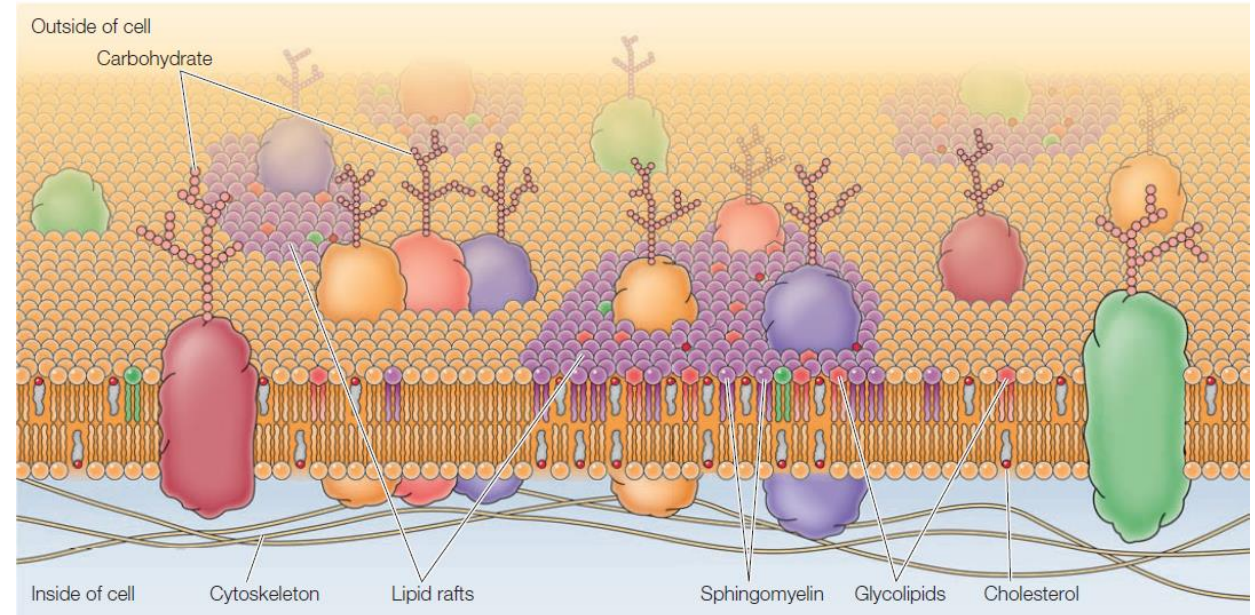
Lipid rafts

- Specialized membrane regions with clusters of cholesterol and the sphingolipids (sphingomyelin and glycolipids).
- Rafts are enriched in glycosylphosphatidylinositol (GPI)-anchored proteins, and proteins involved in signal transduction and intracellular vesicular trafficking (transport).



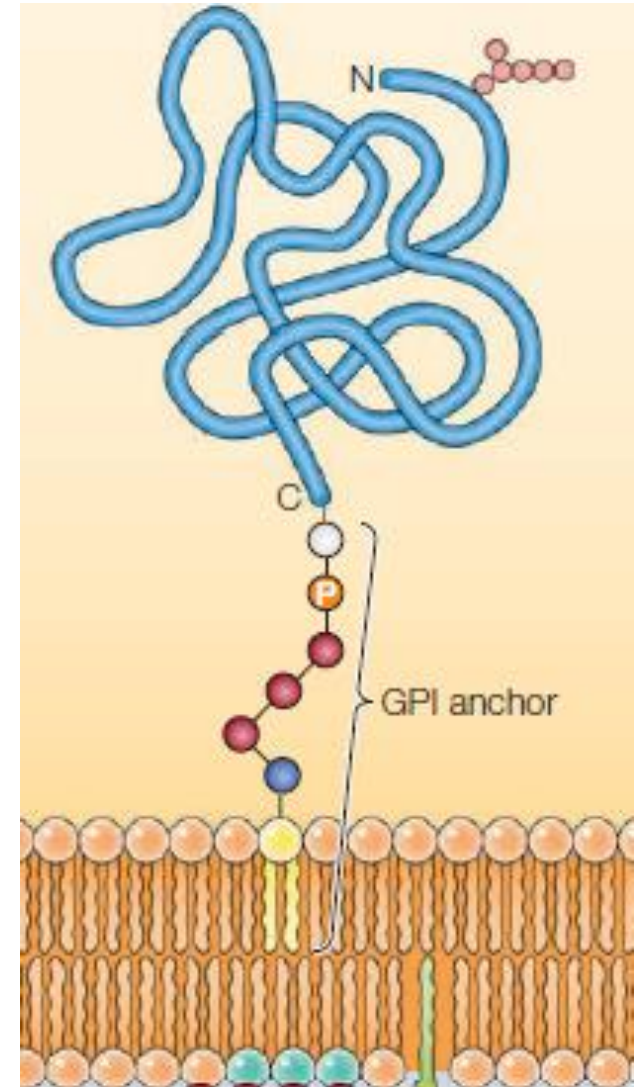
Lipid rafts

- If we look at the plasma membrane we will see that the signaling molecules or signaling proteins (the receptors) like the insulin receptor or epidermal growth factor receptor, and so on, are not all over the place on the plasma membrane.
- We will see that they are clustered, aggregated, in specific regions of the membrane one of these regions called the lipid raft
- **Lipid rafts** are small, specialized regions of the plasma membrane characterized by a high concentration of **cholesterol and sphingolipids, especially glycosphingolipids**. These regions also contain a high number of signaling molecules, which are clustered together within the raft.



Lipid rafts

- Also, the lipid rafts has an associated membrane proteins through a glycolipid by called a Glycosylphosphotidyl inositol.
- Glycosylphosphotidyl inositol is a **lipid molecule** (inserted inside the membrane) covalently bounded to a sugar molecule that bind to a protein.
- These proteins are called (GIP-anchored proteins).



Caveolae (Latin for “little caves”)

- They are a subset of lipid rafts that require cholesterol for their formation.
- They are formed the membrane protein **caveolin**, which **interacts with cholesterol and the cytoplasmic protein cavin**.
- They are important for several cellular activities, including endocytosis, cell signaling, regulation of lipid transport, and protection of the plasma membrane against mechanical stress.

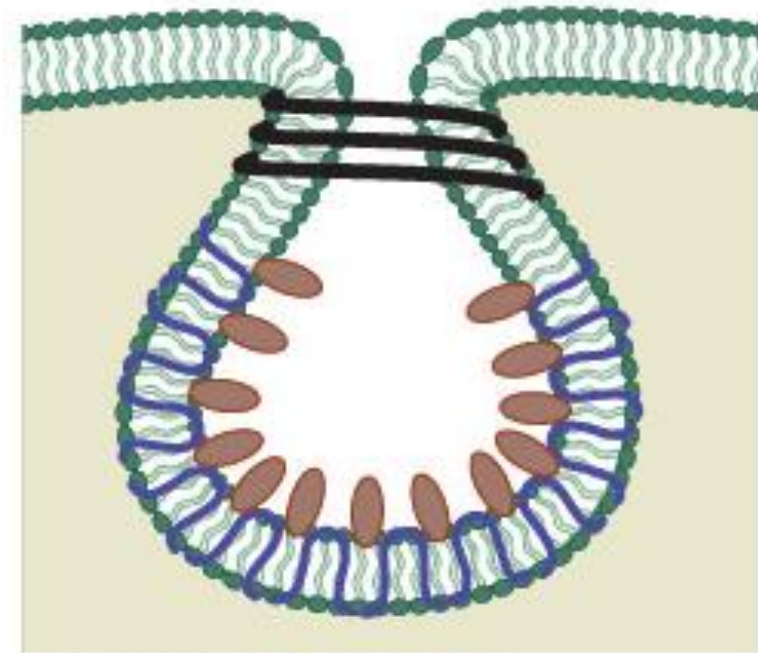
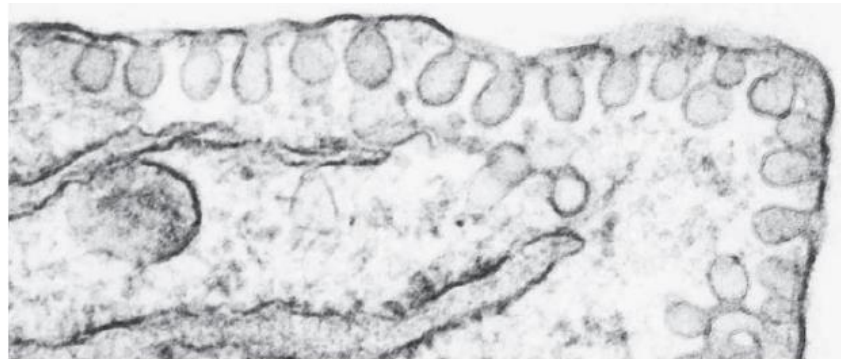
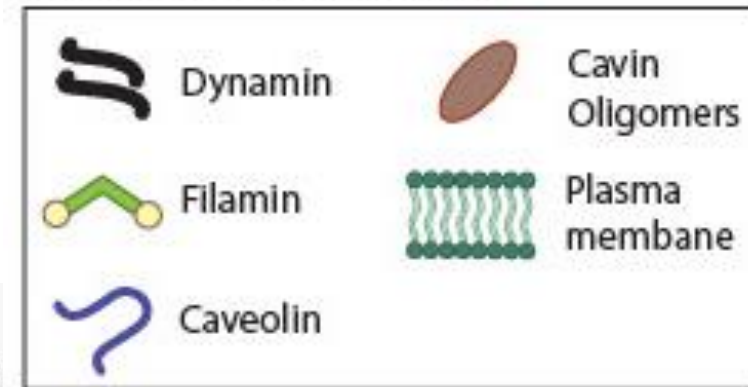


Fig. Simplified model of the Caveolae

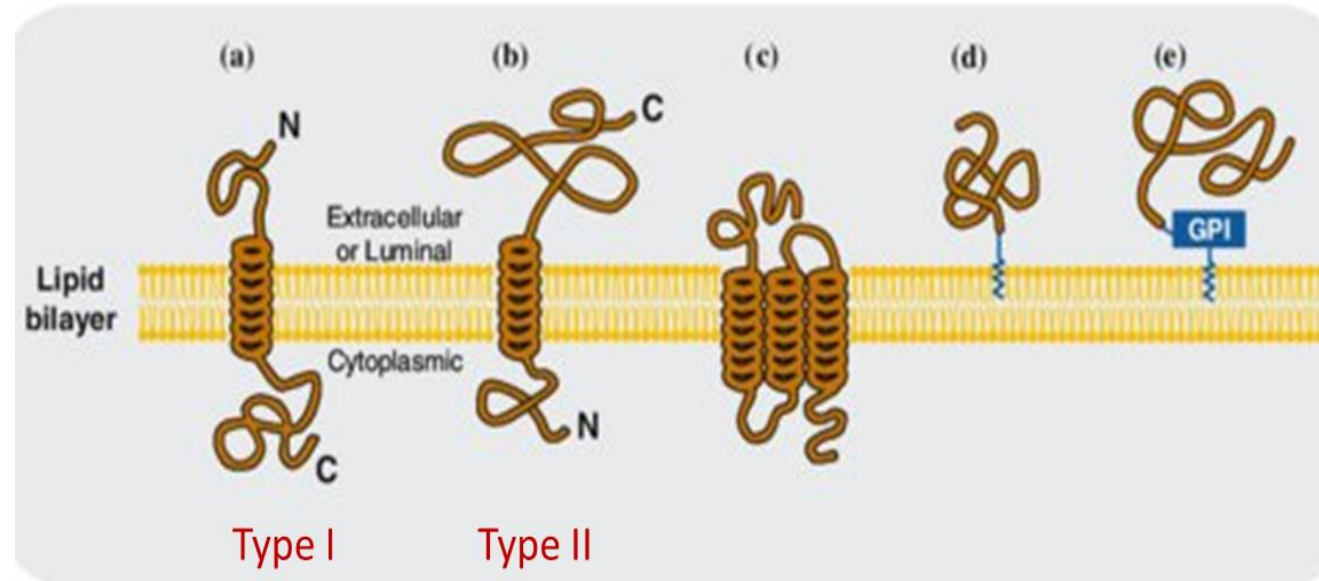


Caveolae

- They are a specific type of a raft known as caveola.
- Caveola have specific types of proteins.
- Caveola are formed by a protein known as caveolin. This caveolin has an interaction with cholesterol and cytolitic protein cavin. the caveolin contains a cluster of signaling molecules (receptors).
- Caveolae are also very important for **transport processes like (endocytosis)** often happens in the caveolae regions. This shows that the **plasma membrane is not homogeneous**, but rather **contains specialized regions, such as lipid rafts and caveolae**, each playing specific roles in cell signaling and transport.

Membrane proteins

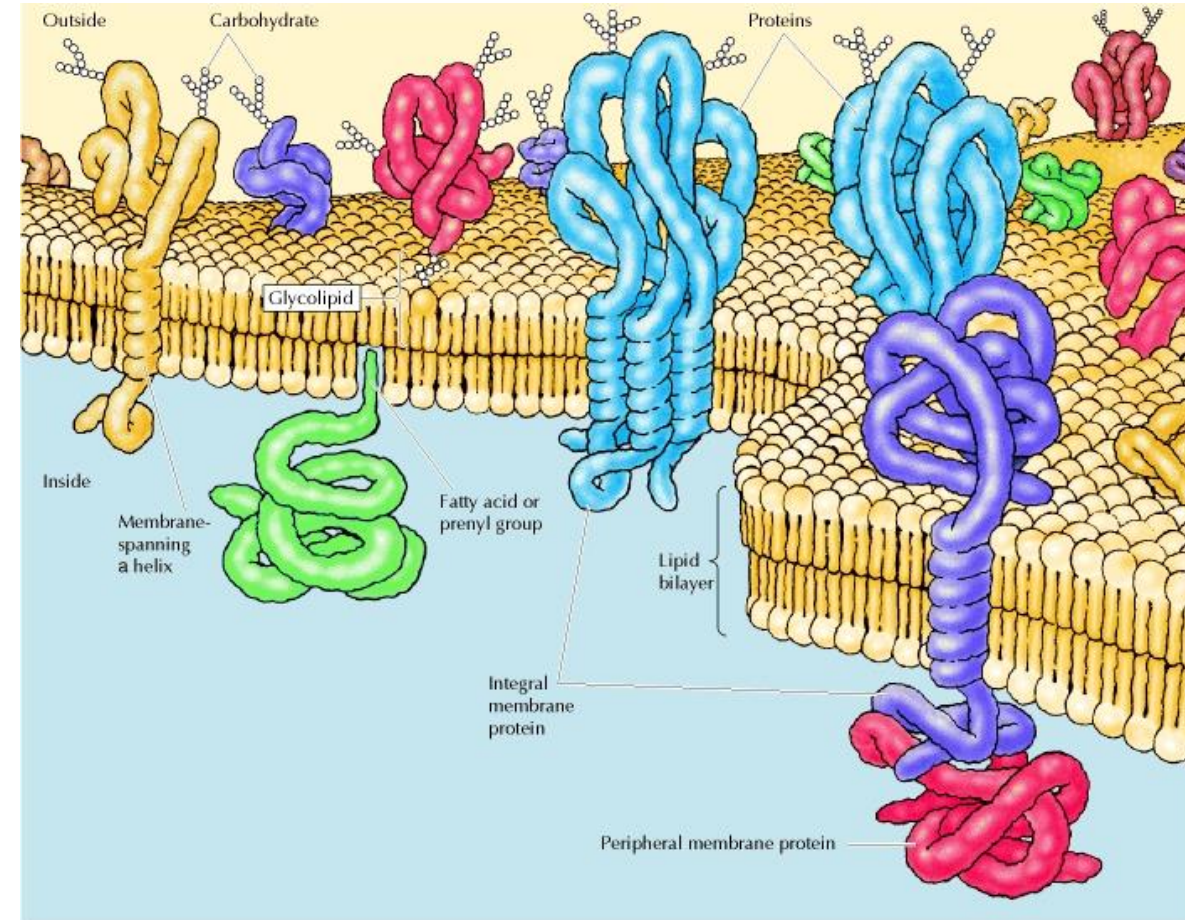
- **Membrane Proteins:** There are different types of proteins associated with the plasma membrane, and they interact with it in various ways. One major category is **integral membrane proteins**. The term integral means that **these proteins are embedded within or inserted into the lipid bilayer** – The region of the protein that spans the membrane is **made up of hydrophobic amino acids**, allowing it to **interact with the hydrophobic core of the lipid bilayer**.



- An integral membrane protein **can cross the membrane once (single-pass), twice (double-pass) these two cases are the most popular , or multiple times** – for example, 7 times (like G-protein-coupled receptors) or even more. This means the protein may enter and exit the membrane several times, forming transmembrane segments.
- Even a protein with a single transmembrane domain differs in its organization (e.g the N terminus can be outside (type I) or inside (type II)) therefore, we have different mechanisms of inserting proteins inside the cell.

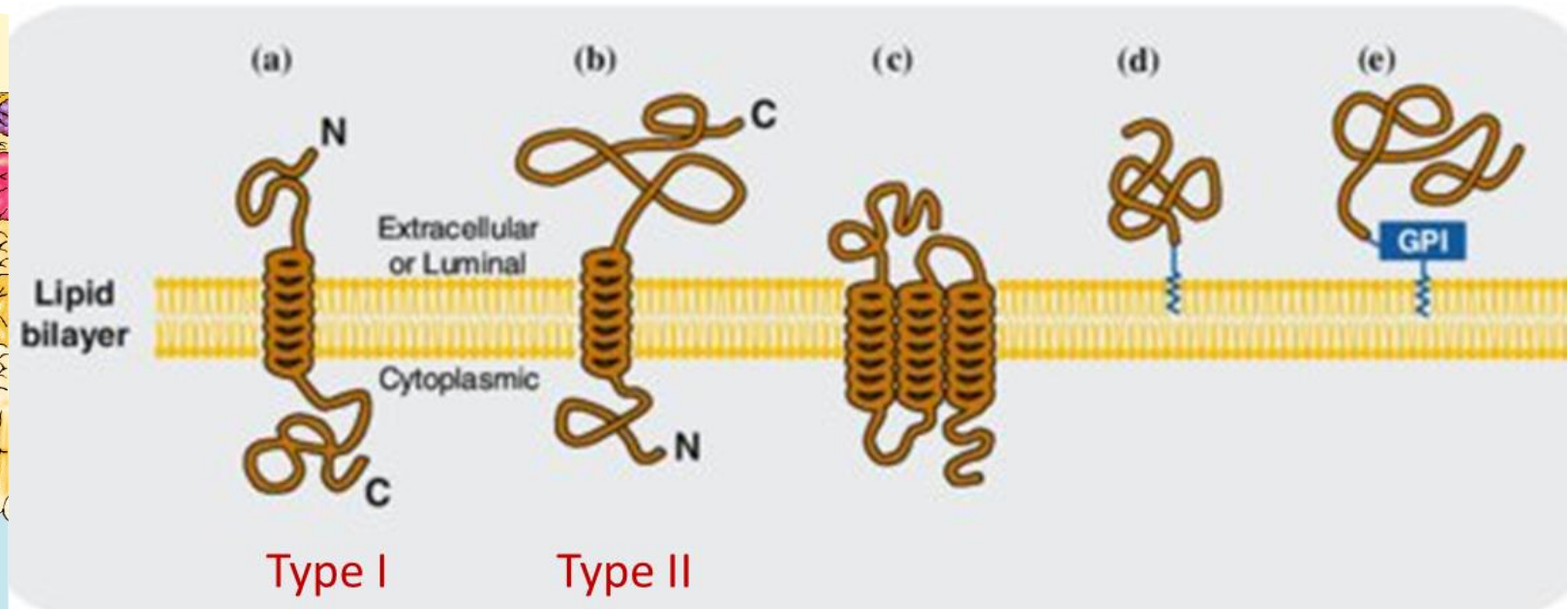
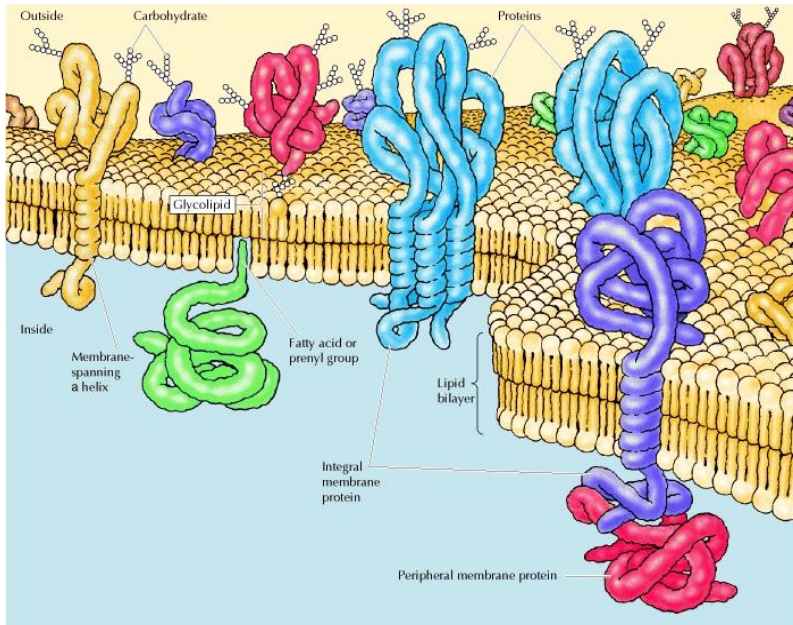
Membrane proteins

- Another type of membrane protein is known as **peripheral membrane proteins**. These proteins **do not penetrate the lipid bilayer**, but rather **associate with one side of the membrane** – either the **inner or outer surface** – by (for example) **electrostatic interactions between charged amino acid residues of the protein and the phosphate head groups of membrane lipids**.
- In addition, some proteins can attach to the membrane through a **lipid molecule**, such as a **fatty acid** (covalently bond to protein and the fatty acid inserted in the membrane).
- or GPI anchor, and these are called **lipid-anchored proteins**. An example is the **GPI-anchored protein**.



Types of membrane proteins

- Peripheral membrane proteins are indirectly and loosely associated with membranes through protein-protein interactions, mainly ionic bonds.
- Integral membrane proteins have some of their **helical** parts inserted into the lipid bilayer.
 - Single-pass (type I or II) or multi-pass proteins.
- Lipid-anchored membrane proteins (myristoylation, palmitoylation, glycosylphosphatidylinositol)



Additional Resources:

رسالة من الفريق العلمي:

Reference Used:

1. 3D human | BioDigital website for 3D cell model : [Cell | BioDigital Human Platform](#)
2. Dopamine (Doctor 2023 JUMedicine modified slides)



اللهم أعزّ الإسلام والمسلمين

For any feedback, scan the code or click on it.



Corrections from previous versions:

Versions	Slide # and Place of Error	Before Correction	After Correction
V0 → V1	Slide 11; the extra gray information	Mice have the same number of chromosomes s in humans but different regulation mechanisms gives completely different creatures.	Mice have the same number of genes as in humans but different regulation mechanisms gives completely different creatures.
V1 → V2			