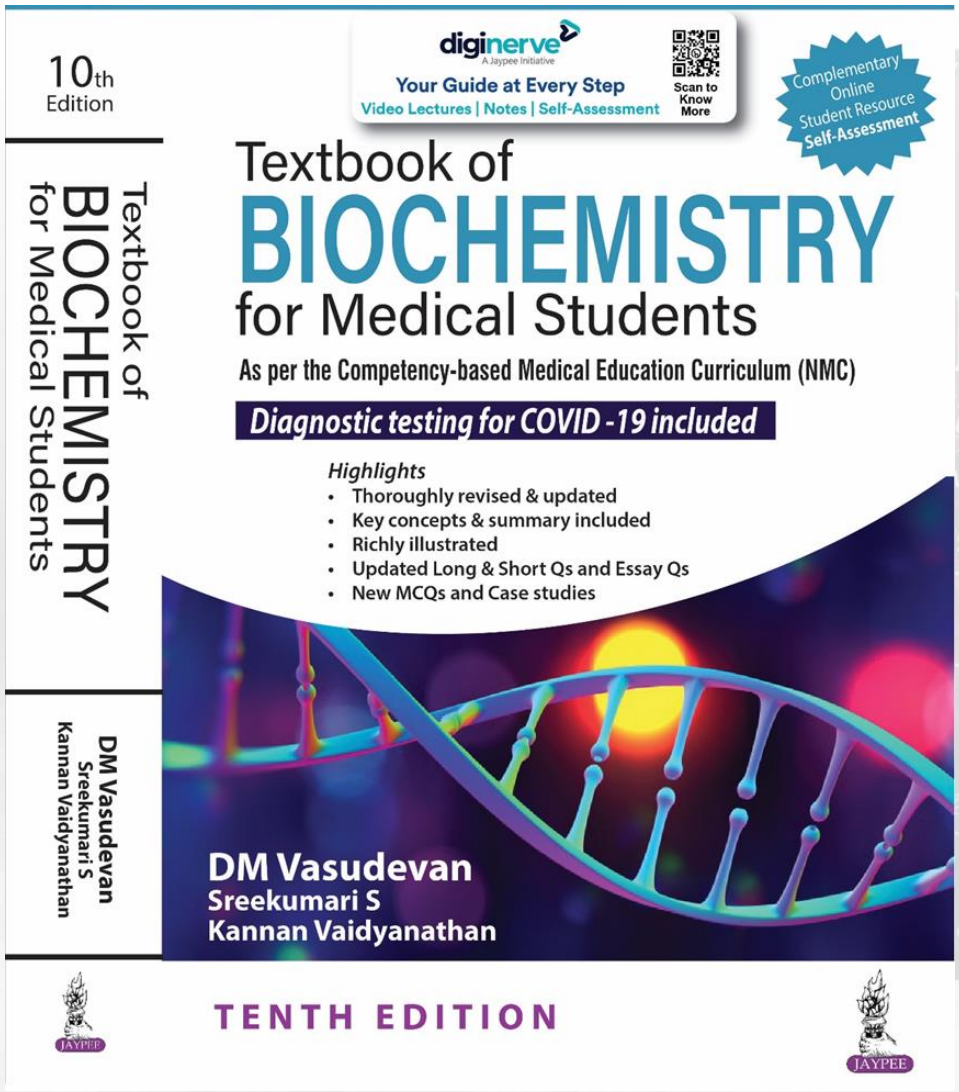


Chapter 2A:

The Cell and Subcellular Organelles

Textbook of

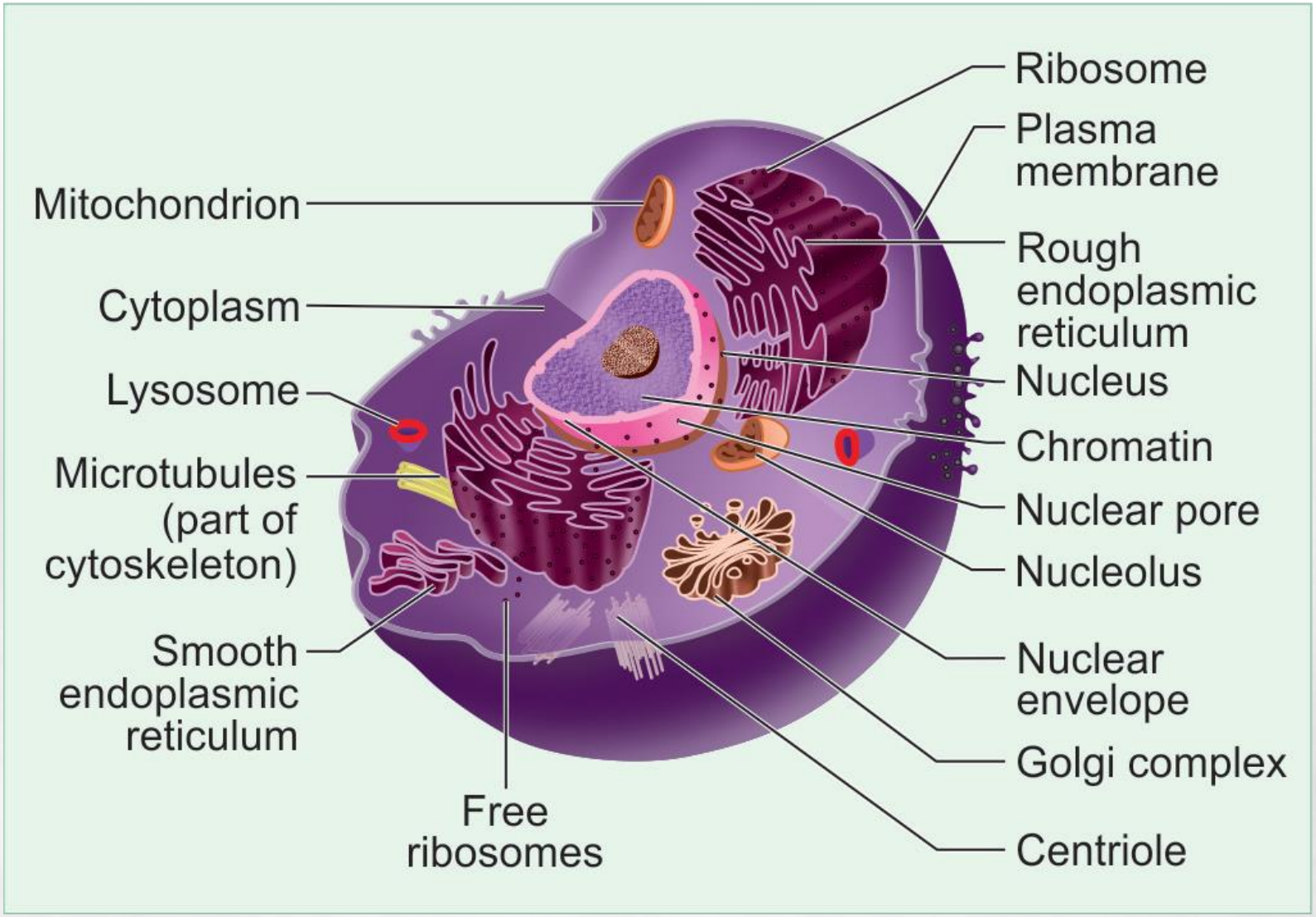
BIOCHEMISTRY

for Medical Students

By DM Vasudevan, *et al.*

TENTH EDITION

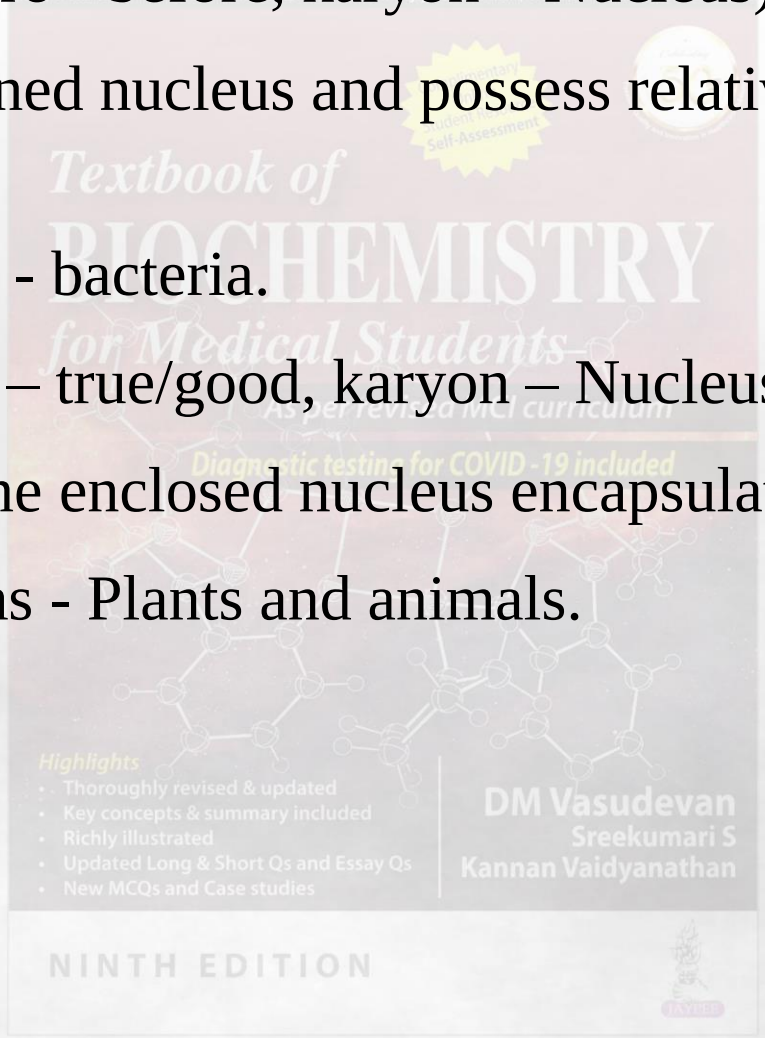
A Typical Cell



- **Prokaryotes** (Pro - before, karyon – Nucleus).
- Lack a well defined nucleus and possess relatively simple structure.
- Lower organism - bacteria.

Eukaryotes (Eu – true/good, karyon – Nucleus)

- Have a membrane enclosed nucleus encapsulating their DNA.
- Higher organisms - Plants and animals.

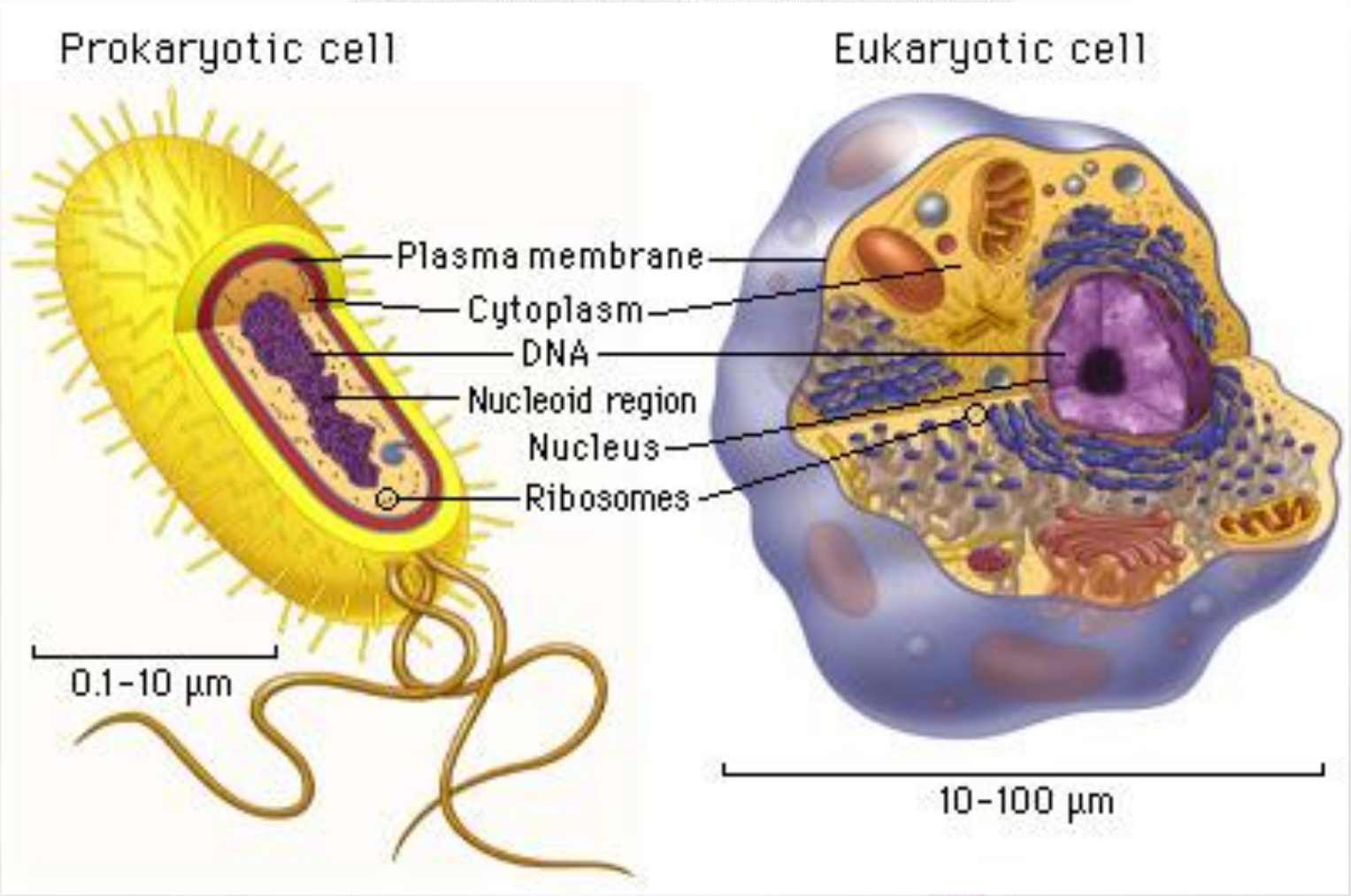


Differences Between Eukaryotic and Prokaryotic Cell

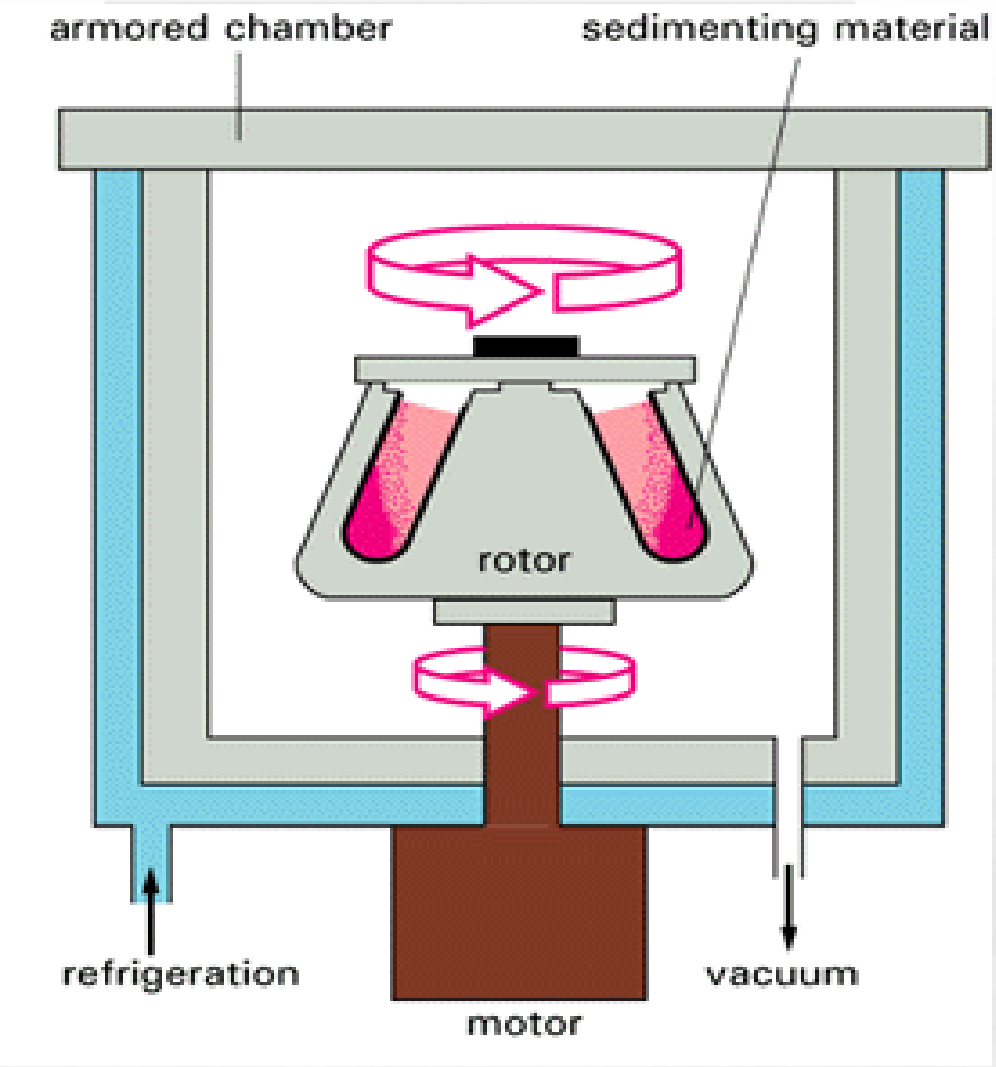
Characteristics	Prokaryotes	Eukaryotes
1. Size	0.1-10 μm	10-100 μm
2. Cell membrane	Rigid cell wall	Plasma membrane
3. Cytoplasm	Viscous, No subcellular organelle	Subcellular organelle present

Difference Between Eukaryotes and Prokaryotes

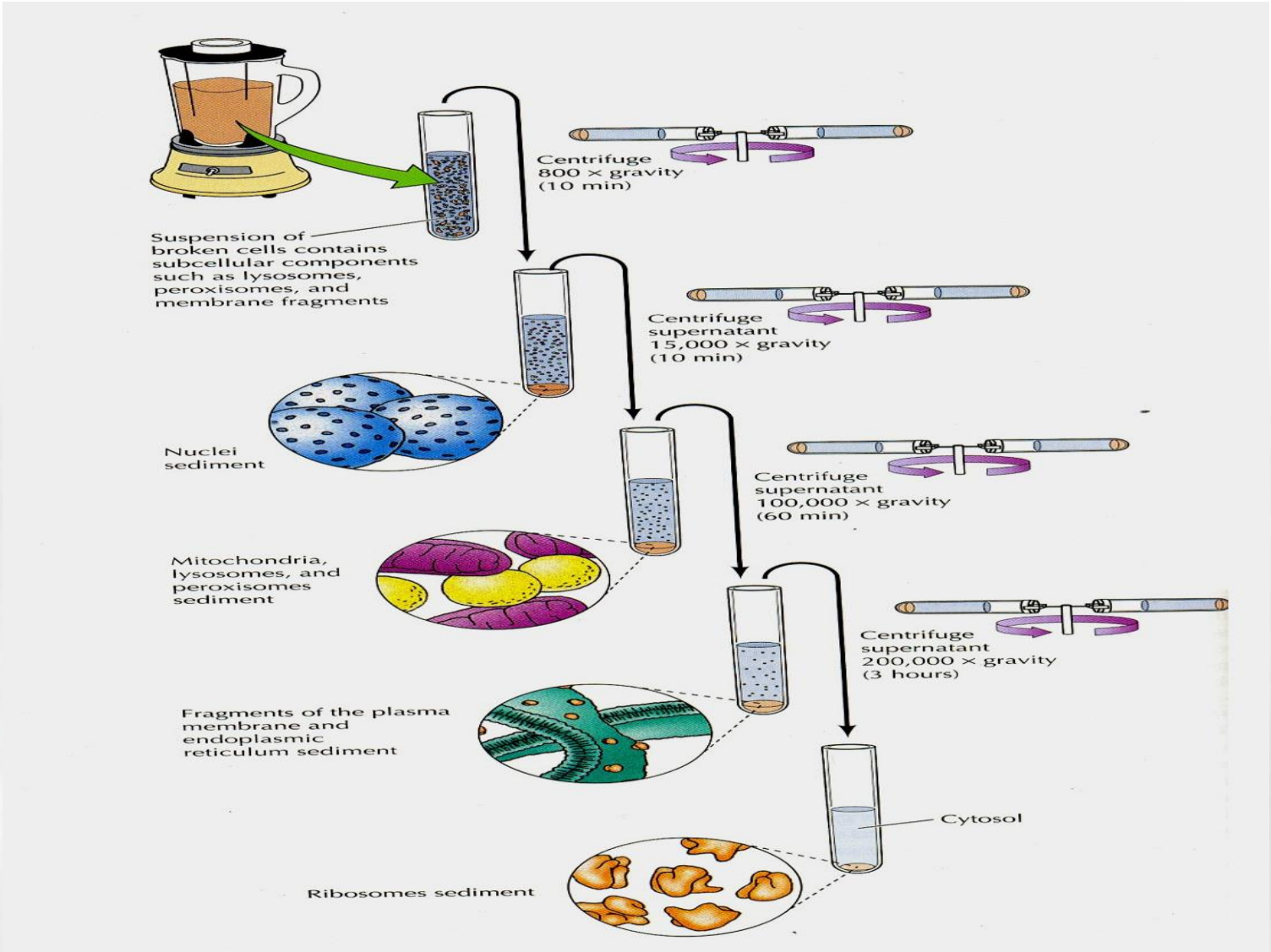
Characteristics	Prokaryotes	Eukaryotes
4. Nucleus	A nuclear zone with DNA	Well defined Nucleus
5. Nucleolus	Absent	Present
6. Mitochondria	Absent	Present
7. Golgi	Absent	Present
8. Cytoskeleton	Absent	Present
9. Cell division	Fission	Mitosis


Prokaryotic cell
Eukaryotic cell
Plasma membrane
Cytoplasm
DNA
Nucleoid region
Nucleus
Ribosomes
0.1–10 µm
10–100 µm

Cell Organelles can be Separated by Ultracentrifugation



Separation of Organelles

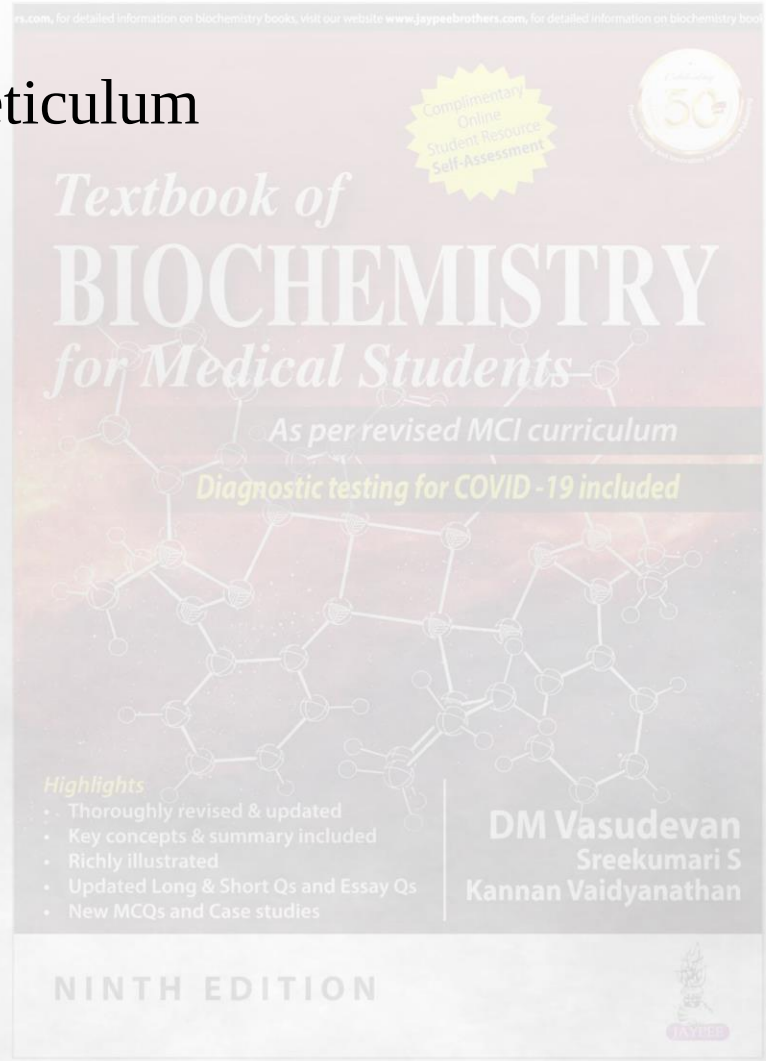


Separation of Subcellular Organelles.

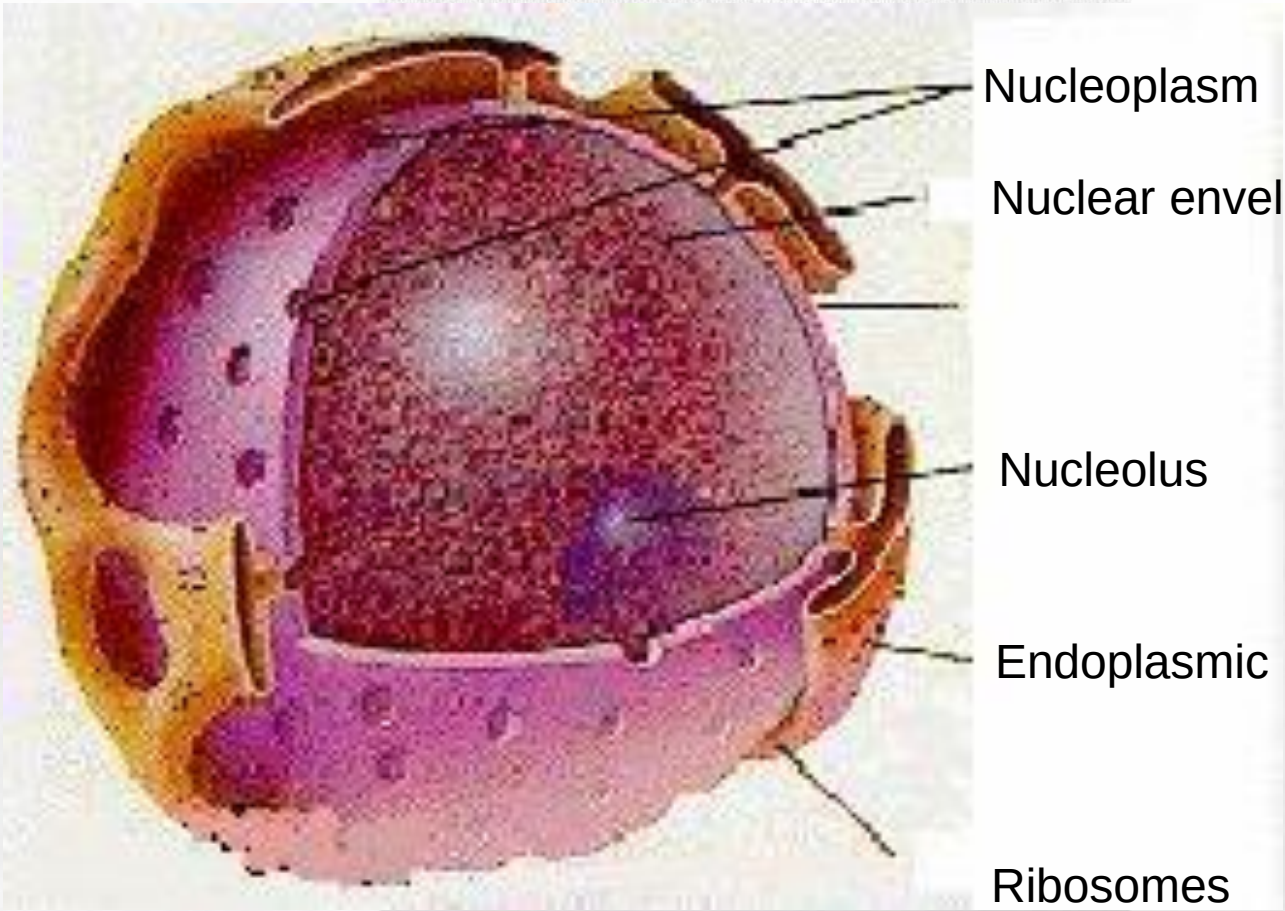
Subcellular organelle Marker enzyme	*Pellet formed at the centrifugal force of*	*Marker*
Nucleus	600–750 x g, 10 minutes	DNA
Nucleus	600–750 x g, 10 minutes Mitochondria 10,000–15,000 x g, 10 minutes	Inner membrane, ATP synthasse
Lysosome	18,000–25,000 x g, 10 minutes	Cathepsin, Acid phosphatase
Golgi complex	35,000–40,000 x g, 30 minutes	Galactosyltransferase
Microsomes	75,000–100,000 x g, 100 minutes	Glucose-6-phosphatase
Cytoplasm	Supernatant	Lactate dehydrogenase

Sub Cellular Organelles

- Nucleus
- Endoplasmic Reticulum
- Golgi apparatus
- Mitochondria
- Lysosomes
- Peroxisomes

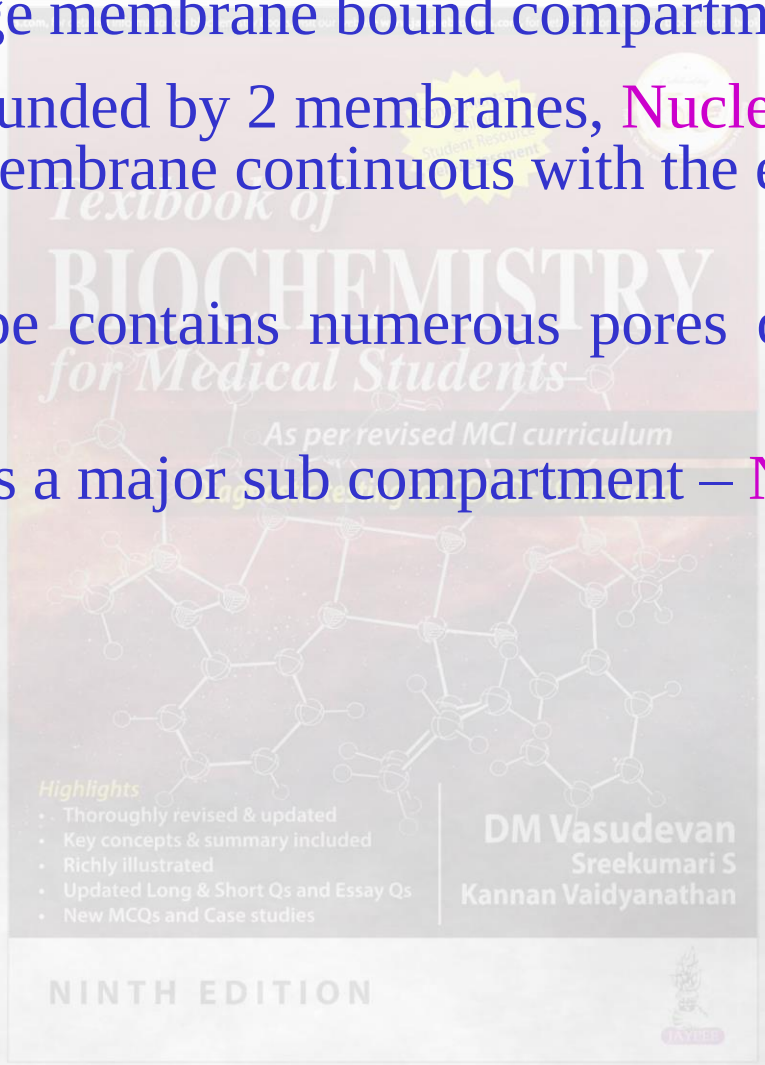


Nucleus is the Site of DNA and RNA Synthesis



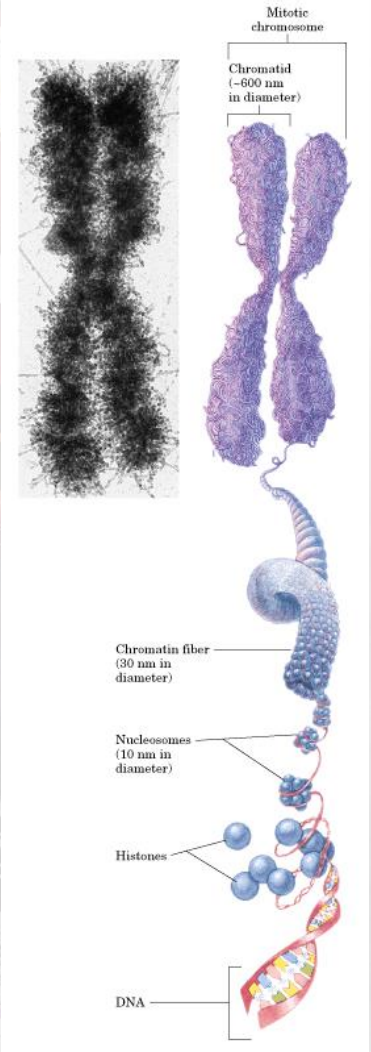
Absent in RBC

- Nucleus is a large membrane bound compartment .
- Nucleus is surrounded by 2 membranes, Nuclear envelope, with the outer membrane continuous with the endoplasmic reticulum.
- Nuclear envelope contains numerous pores of about 90 nm in diameter.
- Nucleus contains a major sub compartment – Nucleolus.



Four Main Types of Carbon-based Molecules are Found in Living Things

Deoxy ribonucleic acid (DNA), the repository of genetic information is located in the nucleus as DNA-Protein complex known as chromatin that is organized into chromosomes during cell division.

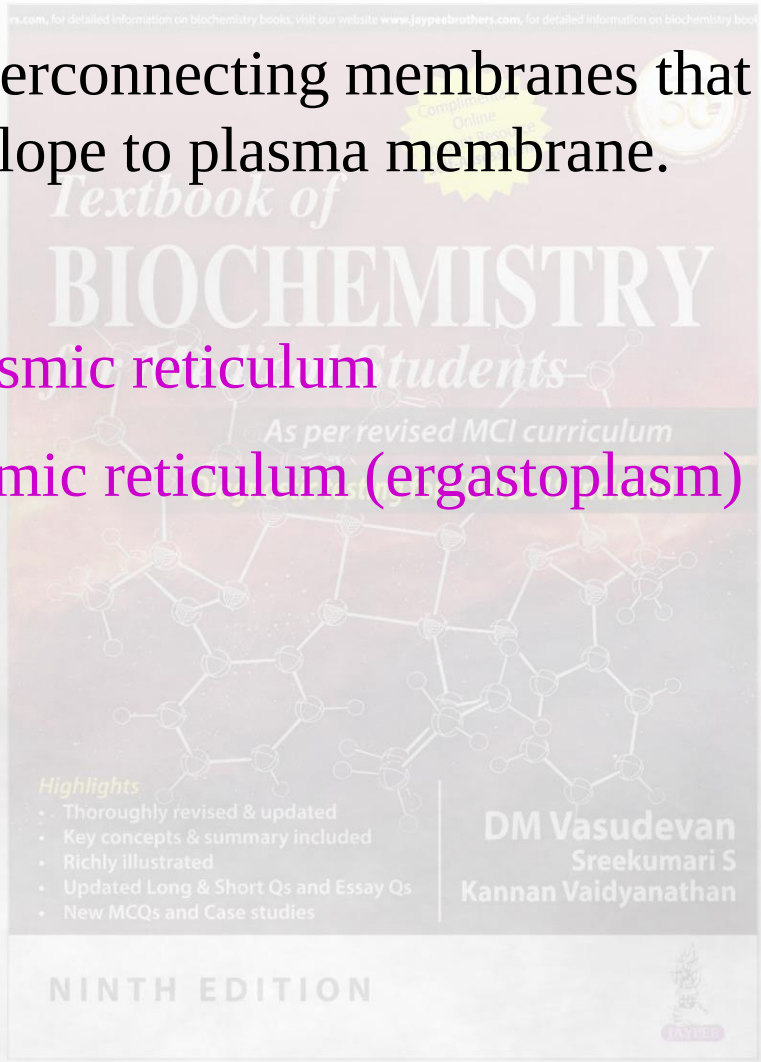


Endoplasmic Reticulum has a Role in Protein Synthesis and Many Synthetic Pathways.

- A network of interconnecting membranes that thread from the nuclear envelope to plasma membrane.

2 Types

- Smooth endoplasmic reticulum
- Rough endoplasmic reticulum (ergastoplasm)



Smooth Endoplasmic reticulum – Microsomes.

- Lipid synthesis
- Hydroxylation reactions. (Cytochrome P450)

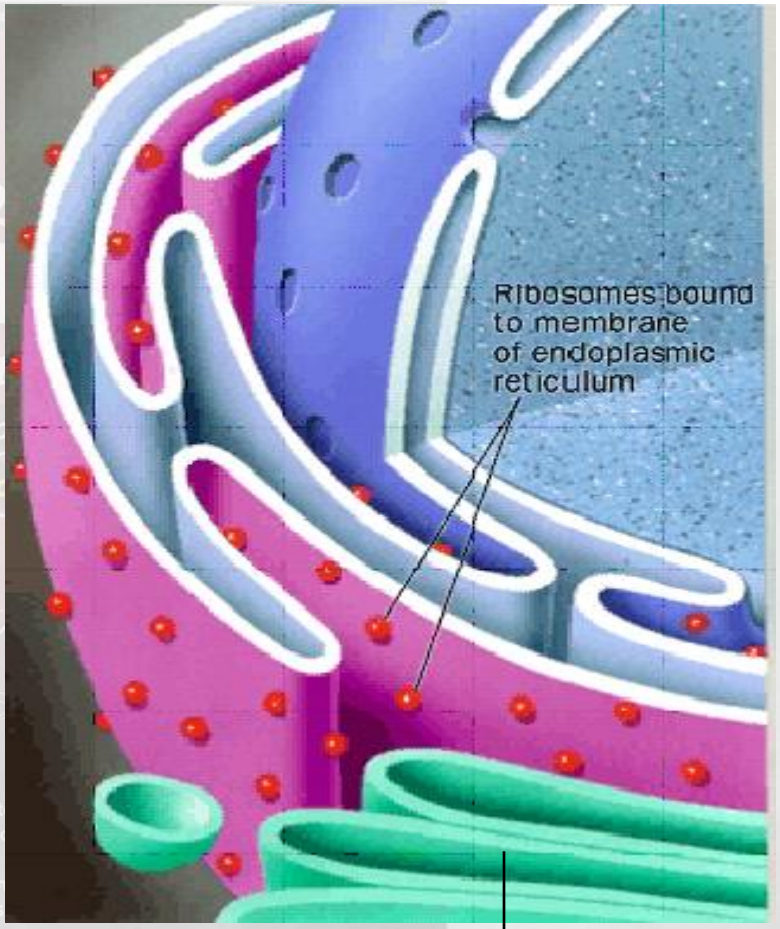
Marker enzyme

Glucose 6 Phosphatase

Rough endoplasmic reticulum

ER studded with ribosomes

- Protein synthesis.

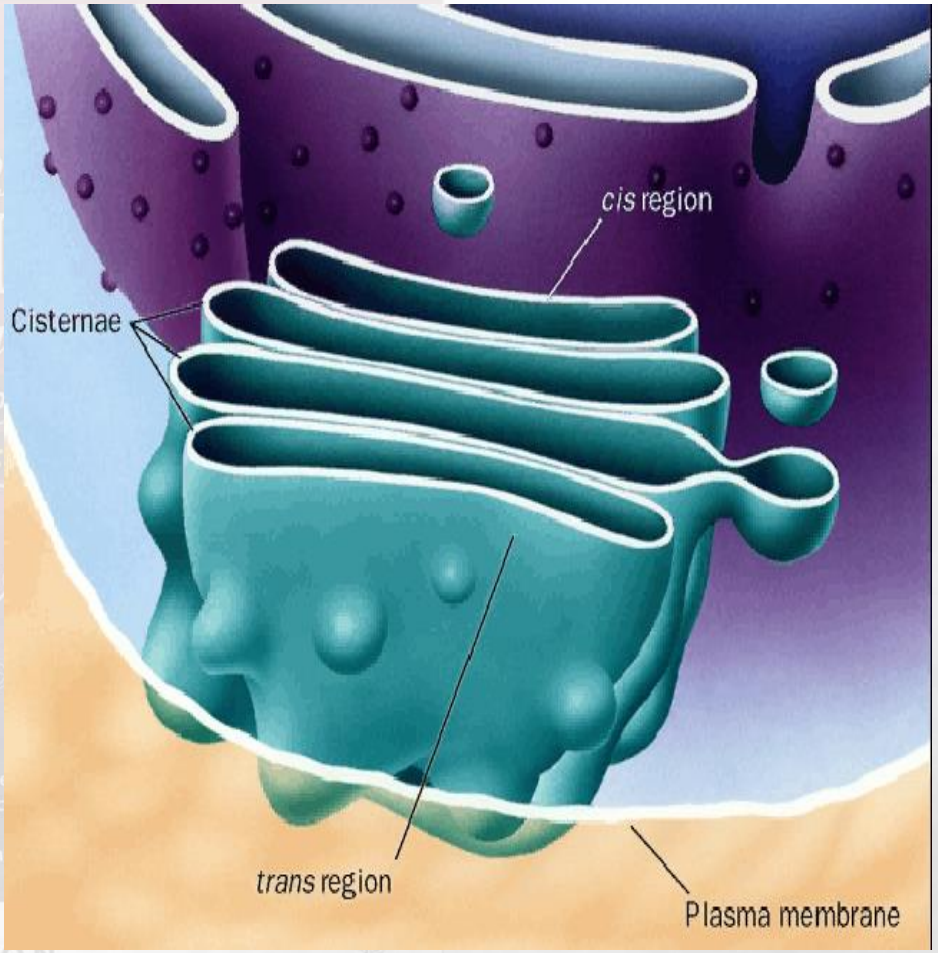


Golgi Complex (Dictyosomes)

Golgi complex is a network of flattened smooth membrane sacs- Cisternae.

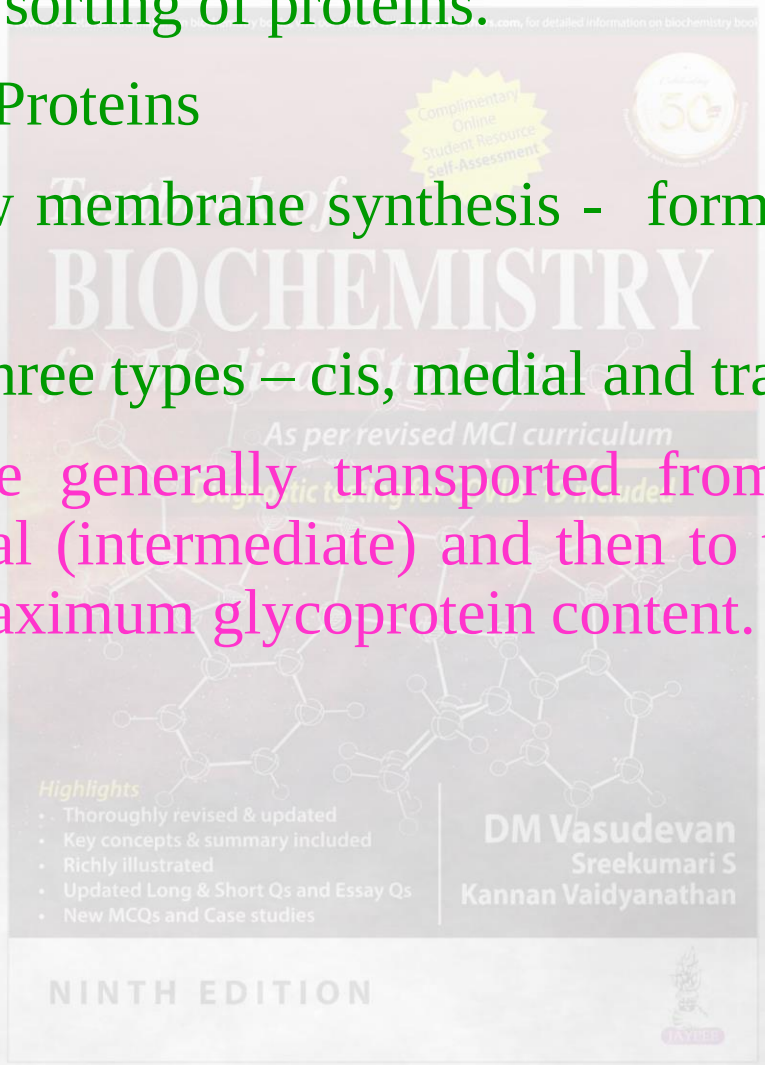
- Marker enzyme

Galactosyl transferase



Golgi Complex is Involved in Secretion of Proteins

- Modification and sorting of proteins.
- Glycosylation of Proteins
- Major site of new membrane synthesis - formation of lysosomes and peroxisomes.
- Cisternae are of three types – cis, medial and trans.
- Glycoproteins are generally transported from ER to cis golgi (proximal), medial (intermediate) and then to trans (distal) golgi. Trans Golgi as maximum glycoprotein content.



The finished products may have the **following destinations:**

1. They may pass through plasma membrane to the surrounding medium. This forms continuous secretion, e.g. secretion of immunoglobulins by plasma cells.
2. They reach plasma membrane and form an integral part of it, but not secreted.
3. They are formed into a secretory vesicle, where these products are stored for a longer time. Under appropriate stimuli, the contents are secreted. Release of trypsinogen by pancreatic acinar cells and release of insulin by beta cells of Langerhans are cited as examples.
4. The synthesized materials may be collected into lysosome packets.

Lysosomes

- Vesicular structures
- Size 0.4 μm
- Surrounded by lipoprotein membrane
- Acidic pH (~5)
- Contain a group of hydrolytic enzymes (>40)
- Marker enzyme :

Acid Phosphatase

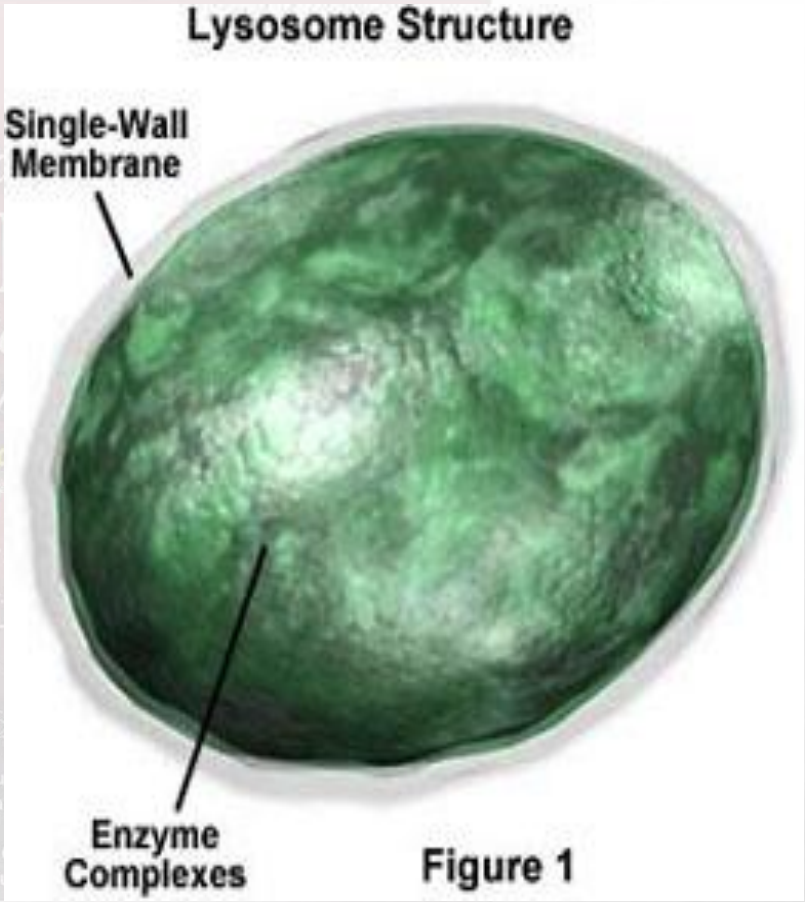
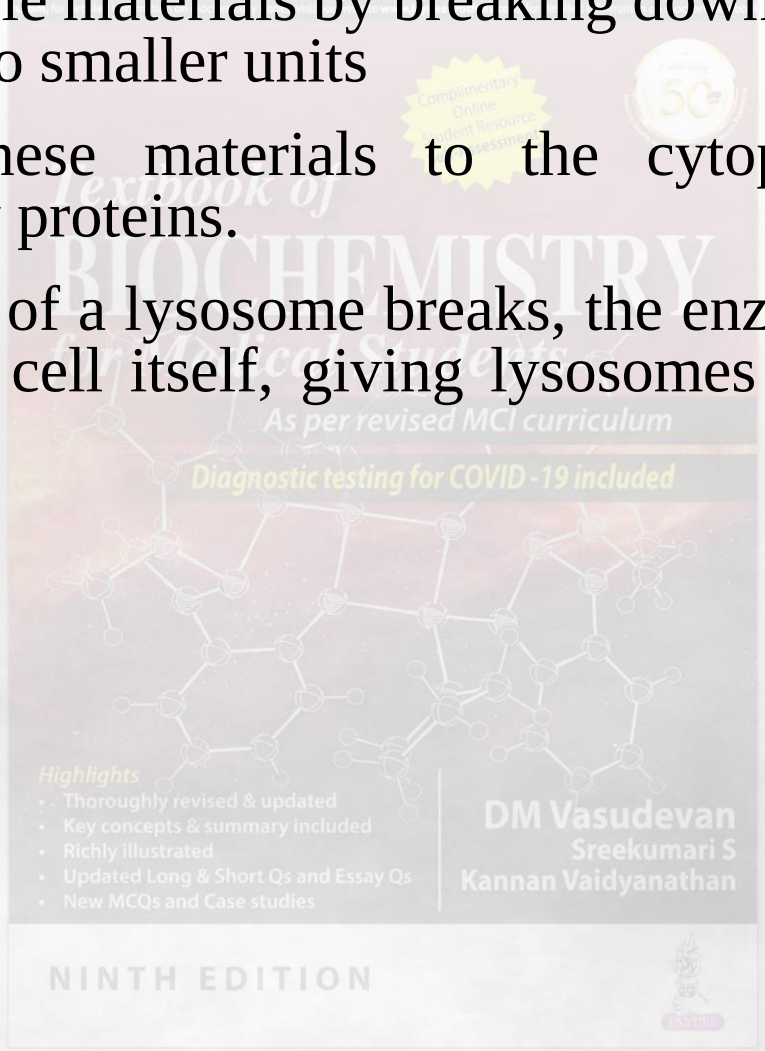


Figure 1

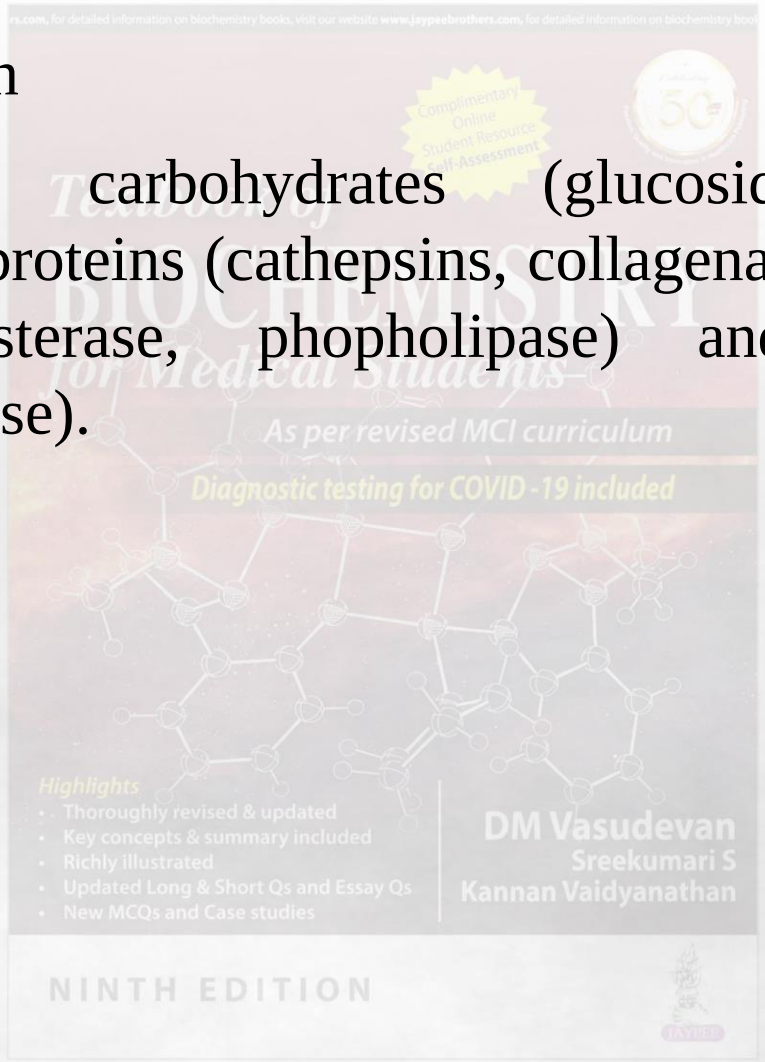
Lysosomes are the Cells' Garbage Disposal System

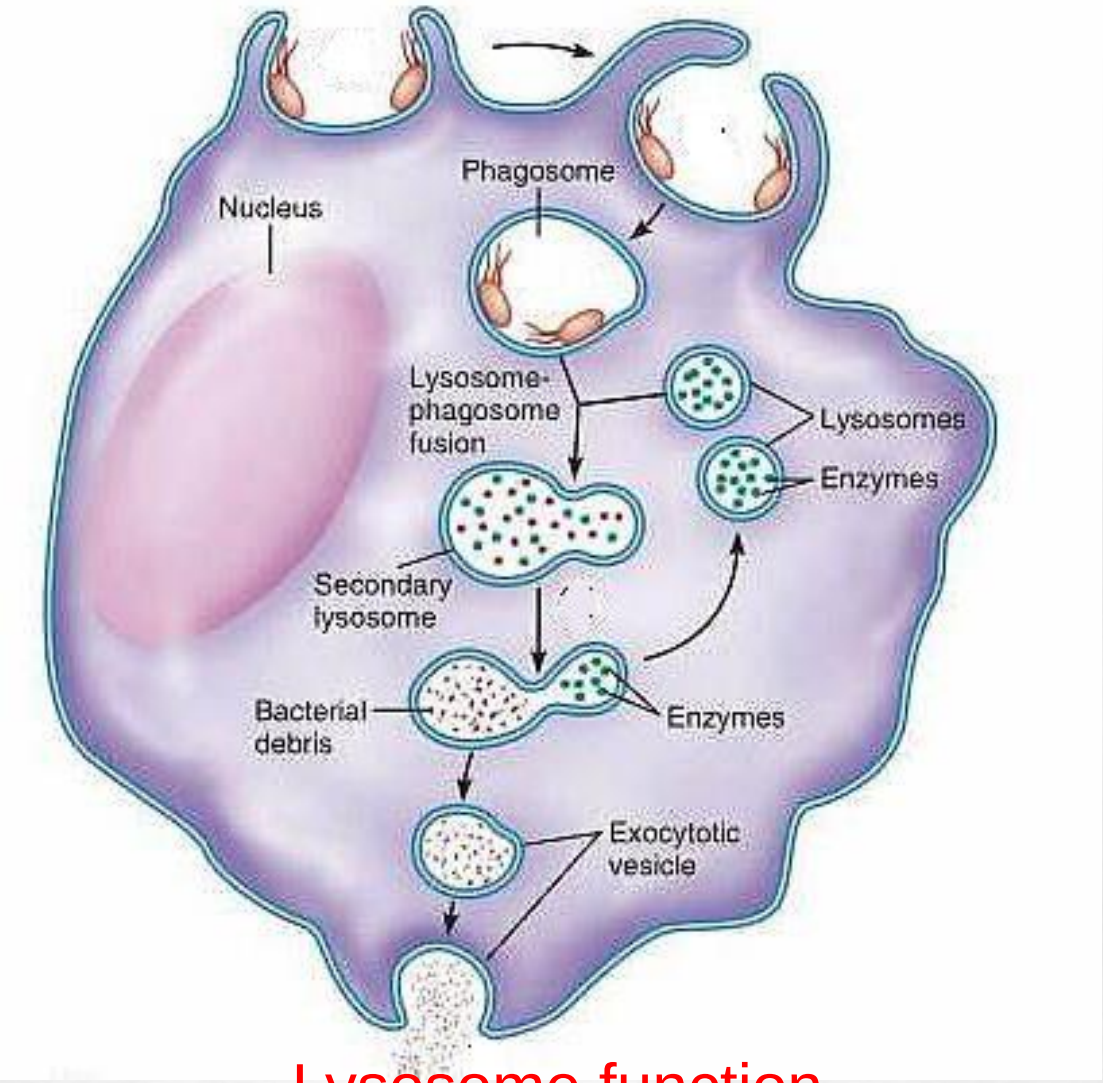
- Lysosomes recycle materials by breaking down worn-out parts of a cell into smaller units
- They deliver these materials to the cytoplasm for use in constructing new proteins.
- If the membrane of a lysosome breaks, the enzymes released may also destroy the cell itself, giving lysosomes the name "suicide bag".



Lysosomes are Required for Intracellular Digestion of Biopolymers

- Cellular digestion
- Hydrolysis of carbohydrates (glucosidase, fucosidase, hyaluronidase); proteins (cathepsins, collagenase, elastase); lipids (fatty acyl esterase, phopholipase) and nucleic acids (phosphodiesterase).





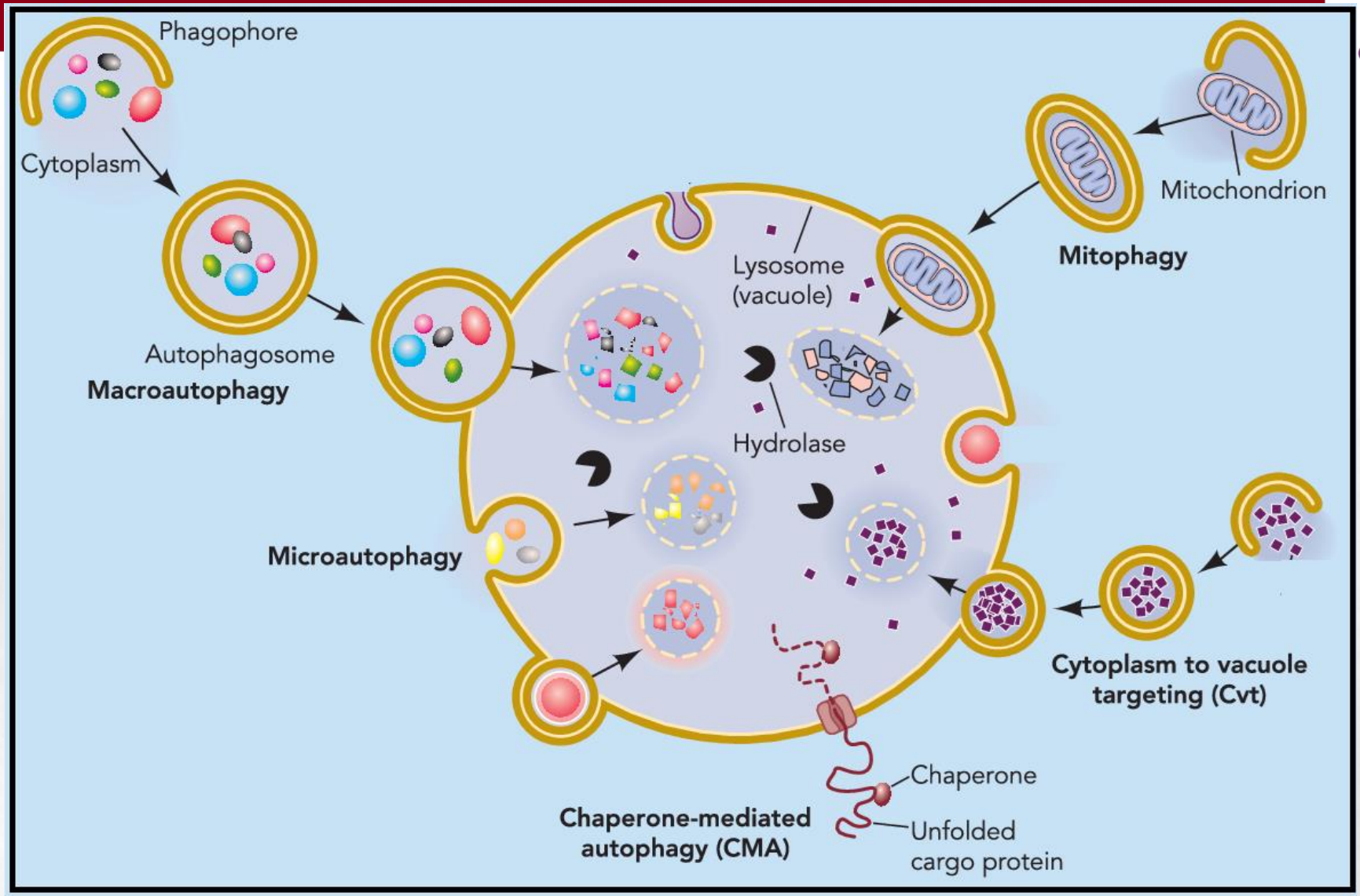
Lysosome function

Lysosomes – Clinical Aspects

- Lysosomal storage disorders
- Inclusion cell disease – a protein targeting defect – accumulation of undegraded molecules – inclusion bodies.
- Gout – Urate crystals are phagocytosed and cause physical damage of lysosomes releasing enzymes producing inflammation and arthritis.
- Cathepsins are implicated in tumor metastasis. They degrade basal lamina of cells.
- Silicosis – Lysosomal enzymes are released stimulating fibroblast proliferation.

Autophagy

Lysosomes play a central role in autophagy. Autophagy is a self-digesting mechanism responsible for the removal of damaged organelles, malformed proteins during biosynthesis, and nonfunctional proteins. There are three general types of autophagy, namely, microautophagy, chaperone-mediated autophagy (CMA), and macroautophagy. In microautophagy, cytoplasmic proteins are taken up by direct invagination to the lysosomal membrane. In the second mechanism of CMA, proteins flagged with markers were selectively taken up by the lysosome and are degraded. Macroautophagy involves the formation of subcellular structures called autophagosomes that deliver the unwanted materials into lysosomes for breakdown. Without autophagy, toxic protein aggregates are not discarded and accumulate causing cancer, neuromuscular disorders, Alzheimer's disease and other age related disorders.



Different types of autophagy.

Peroxisomes

Diverse group of organelles found in eukaryotic cell. In most human cells , their abundance ranges from less than hundred to more than thousand per cell.

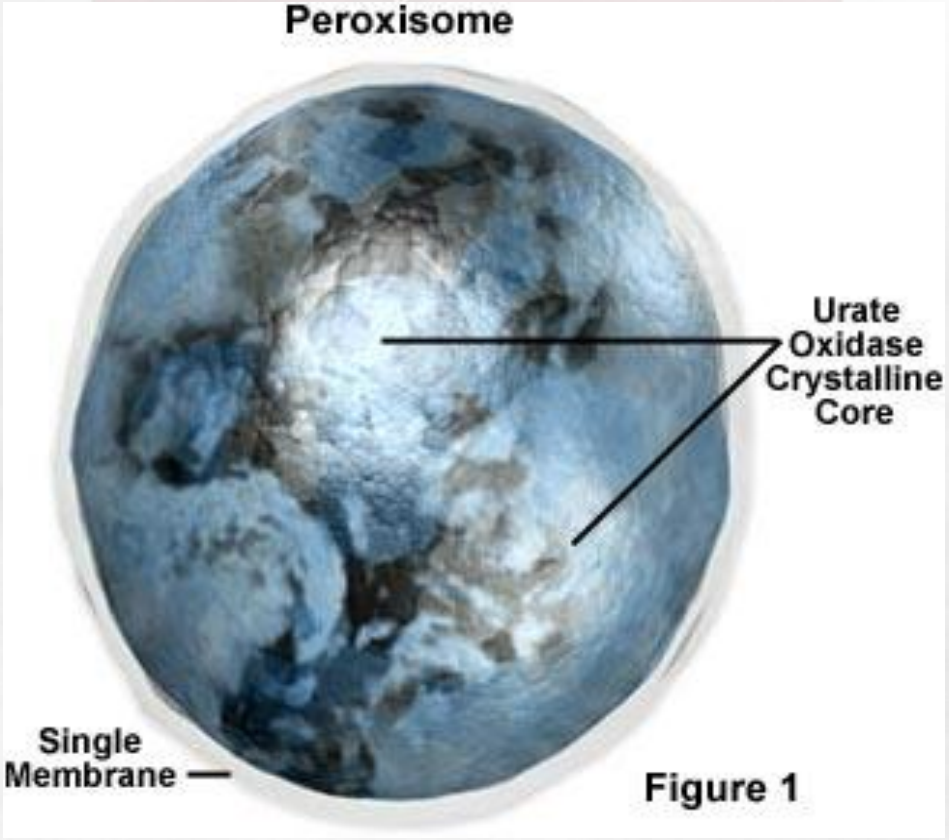
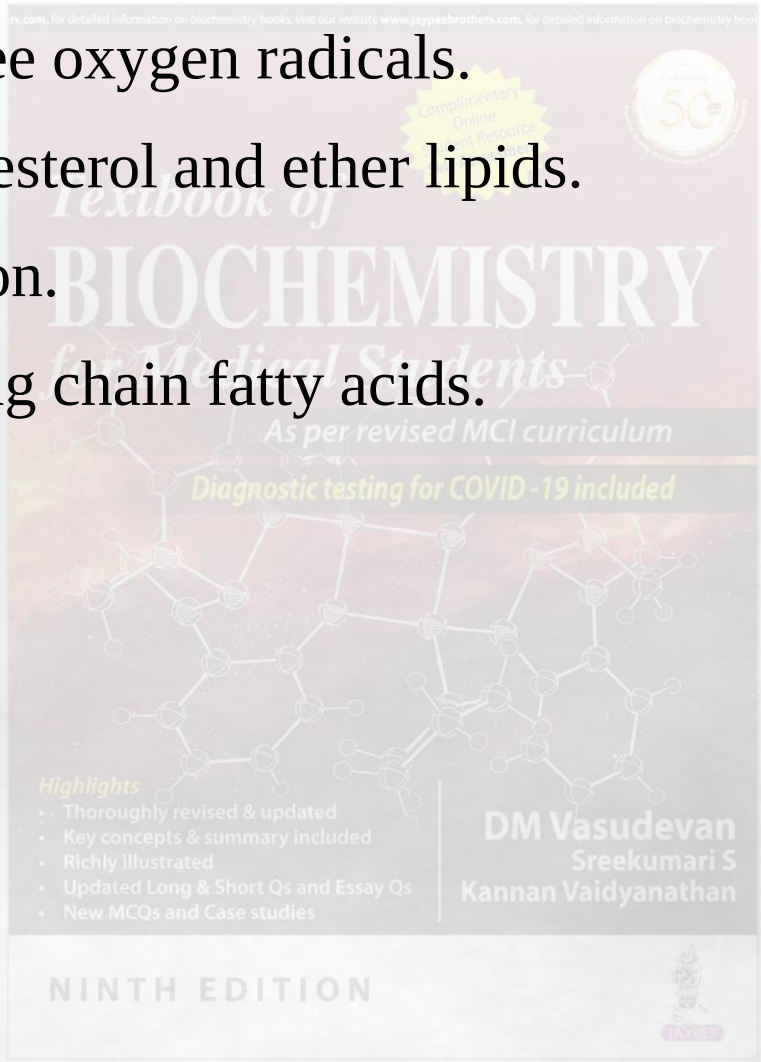


Figure 1

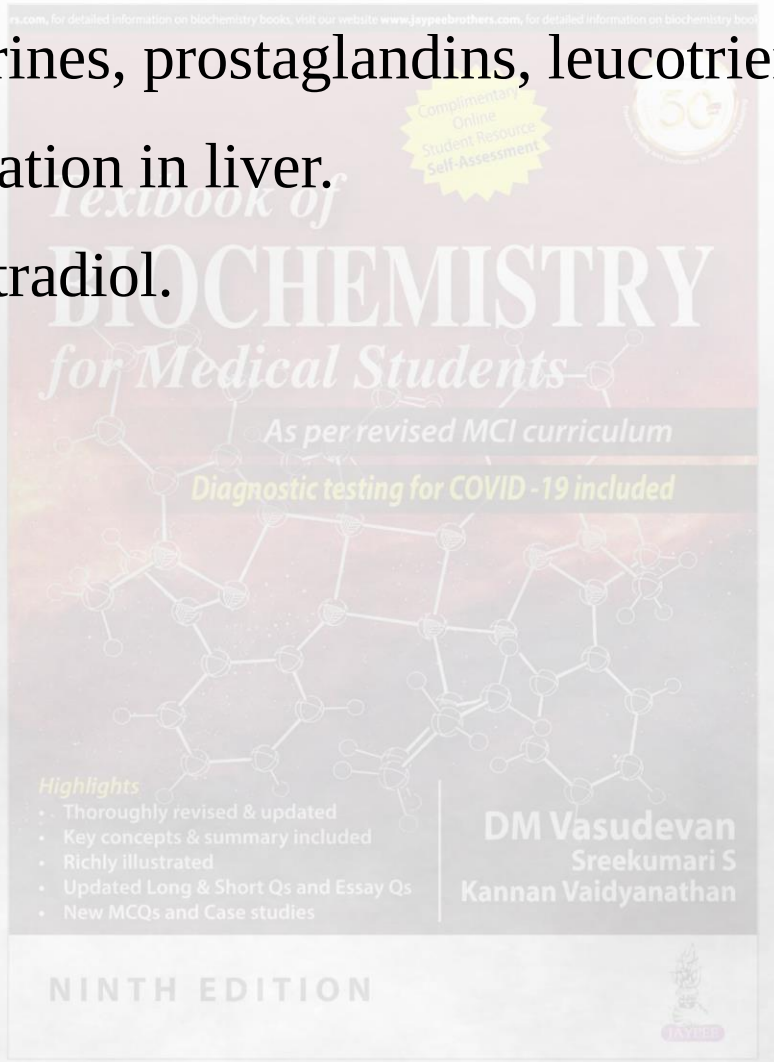
Peroxisomes have got an Important Role in Lipid Metabolism

- Metabolism of free oxygen radicals.
- Synthesis of cholesterol and ether lipids.
- Bile acid formation.
- Catabolism of long chain fatty acids.



Peroxisomes has got an Important Role in Lipid Metabolism

- Catabolism of purines, prostaglandins, leucotrienes.
- Alcohol detoxification in liver.
- Metabolism of estradiol.



Peroxisomes are named so because they play unique role in Hydrogen Peroxide metabolism.

- Contain enzymes that use molecular oxygen to remove hydrogen atoms from specific substrates

$$RH_2 + O_2 \rightarrow R + H_2O_2$$

- Hydrogen peroxide is used up or detoxified.
- Catalases in peroxisome convert this hydrogen peroxide to water.

$$2H_2O_2 \rightarrow 2\ H_2O + O_2$$

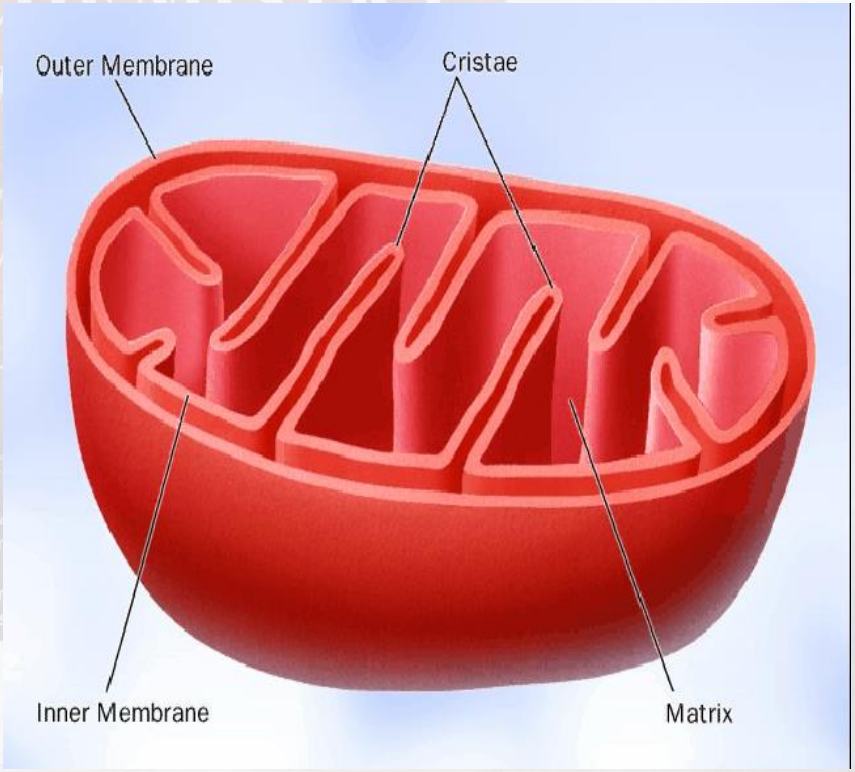
Clinical Significance of peroxisomes

Peroxisomal biogenesis disorders

1. Deficiency of peroxisomal matrix proteins can lead to **adrenoleukodystrophy (ALD)** (Brown-Schilder's disease) characterized by progressive degeneration of liver, kidney and brain. It is a rare autosomal recessive condition. The defect is due to insufficient oxidation of very long chain fatty acids (VLCFA) by peroxisomes.
2. In **Zellweger syndrome,** proteins are not transported into the peroxisomes. This leads to formation of empty peroxisomes or peroxisomal ghosts inside the cells.
3. **Primary hyperoxaluria** is due to the defective peroxisomal metabolism of glyoxalate derived from glycine

Mitochondria

- Spherical, oval or rod like bodies
- Size: 0.2- 0.8 μm
- Number of mitochondria in a cell varies.
  Erythrocytes no mitochondrion
  Liver cell 800-2500



Mitochondria

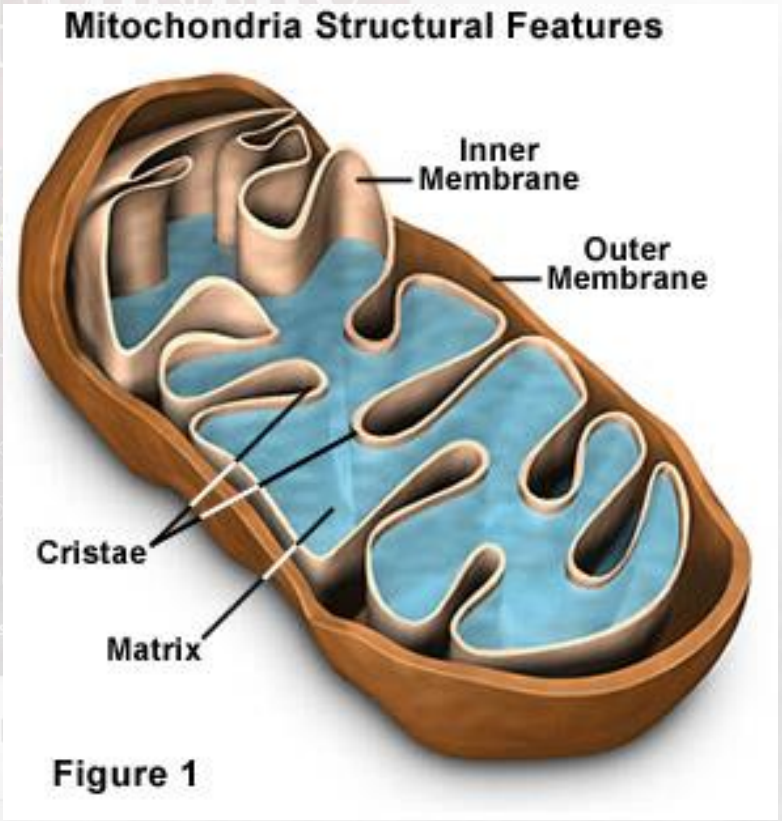
- Bilayered

OMM – Smooth

IMM – convolutes into folds - Cristae

2 Compartments

- Inter membrane space
- Matrix

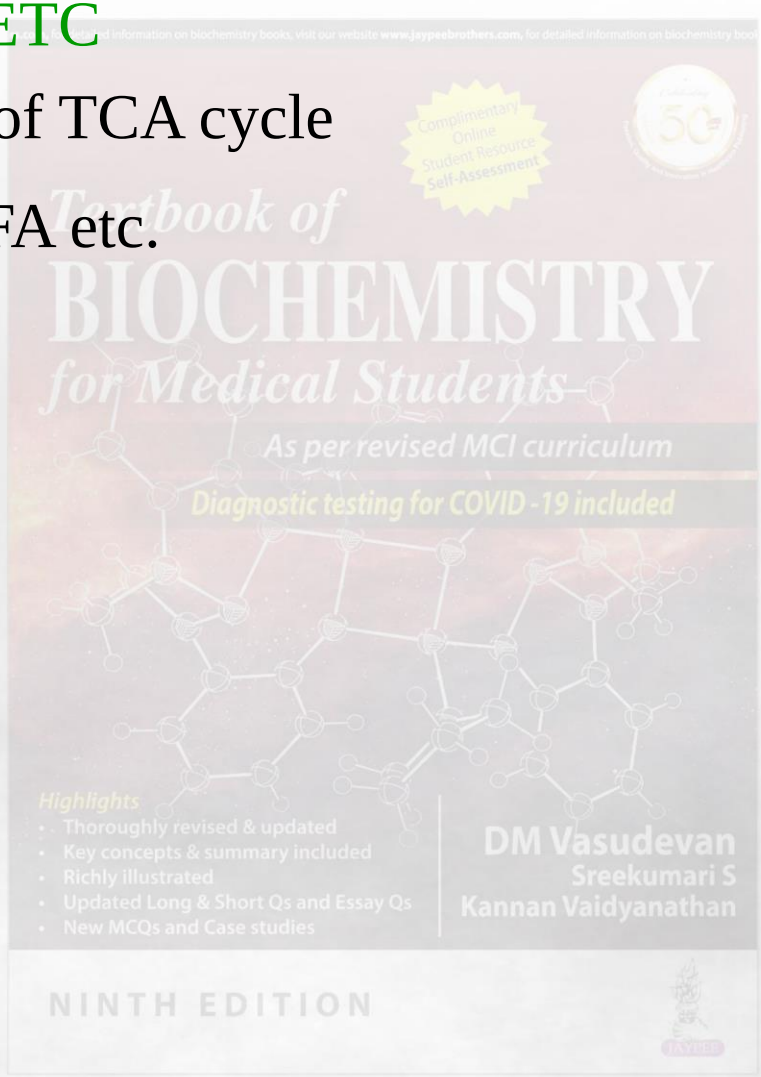


Mitochondria are the 'Power House' of the Cell

IMM contains the ETC

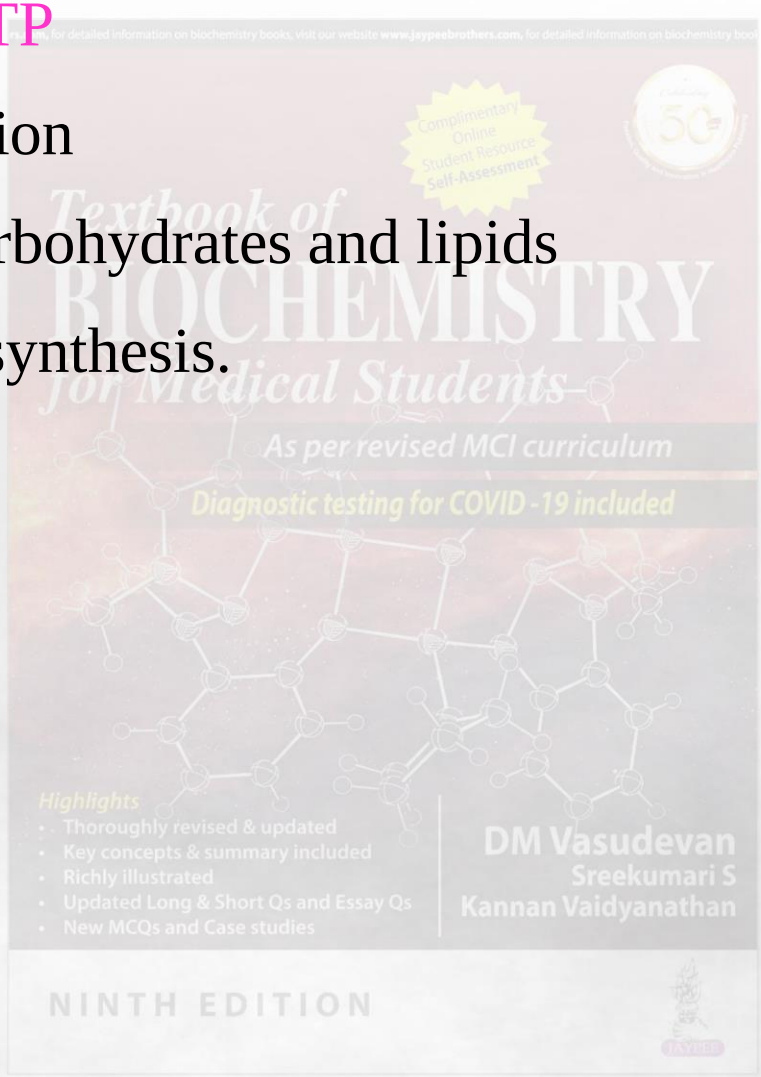
- Matrix- Enzymes of TCA cycle

Beta Oxidation of FA etc.



Mitochondria Supply Most of the Cell's Need for ATP

- Production of ATP
- Cellular respiration
- Oxidation of Carbohydrates and lipids
- Urea and heme synthesis.



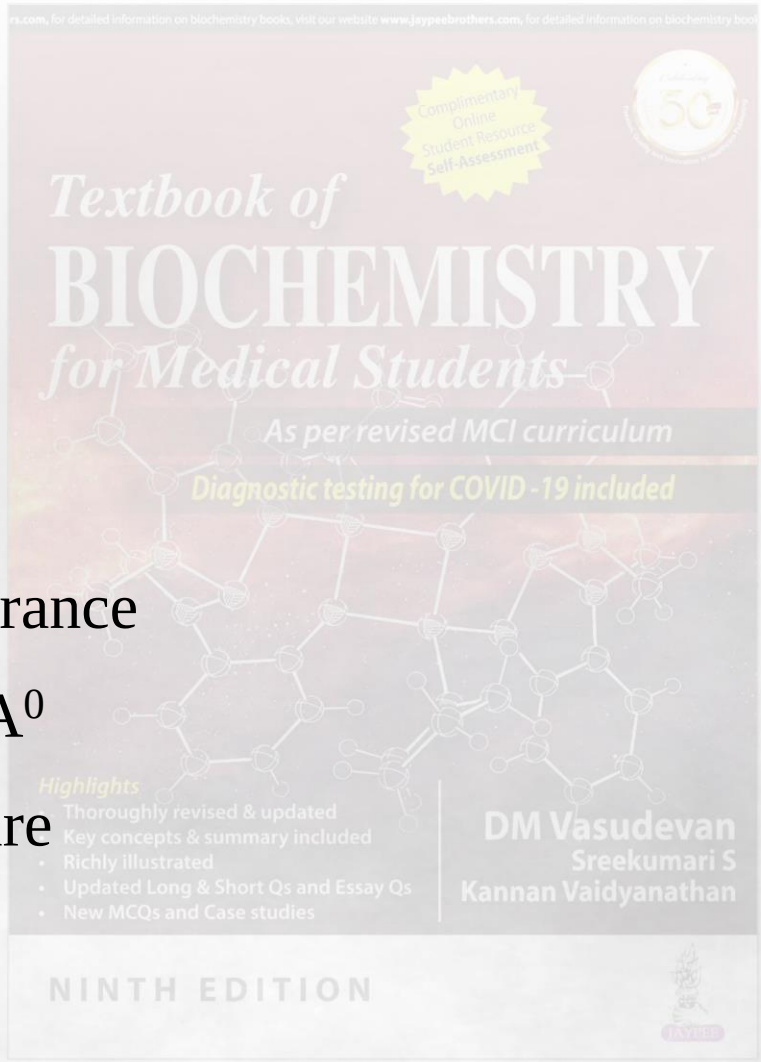
Clinical Significance

- Luft's disease – Defective energy transduction
- Mt. Myopathies – OXPHOS diseases

  Due to mutation in Mt DNA
- Parkinson's, Cardiomyopathies – age related degenerative diseases.
- Antibiotics inhibiting bacterial protein synthesis do not affect cellular processes, but inhibits mitochondrial protein synthesis.
- Mitochondria are considered 'parasites' which entered the cell during the course of evolution !

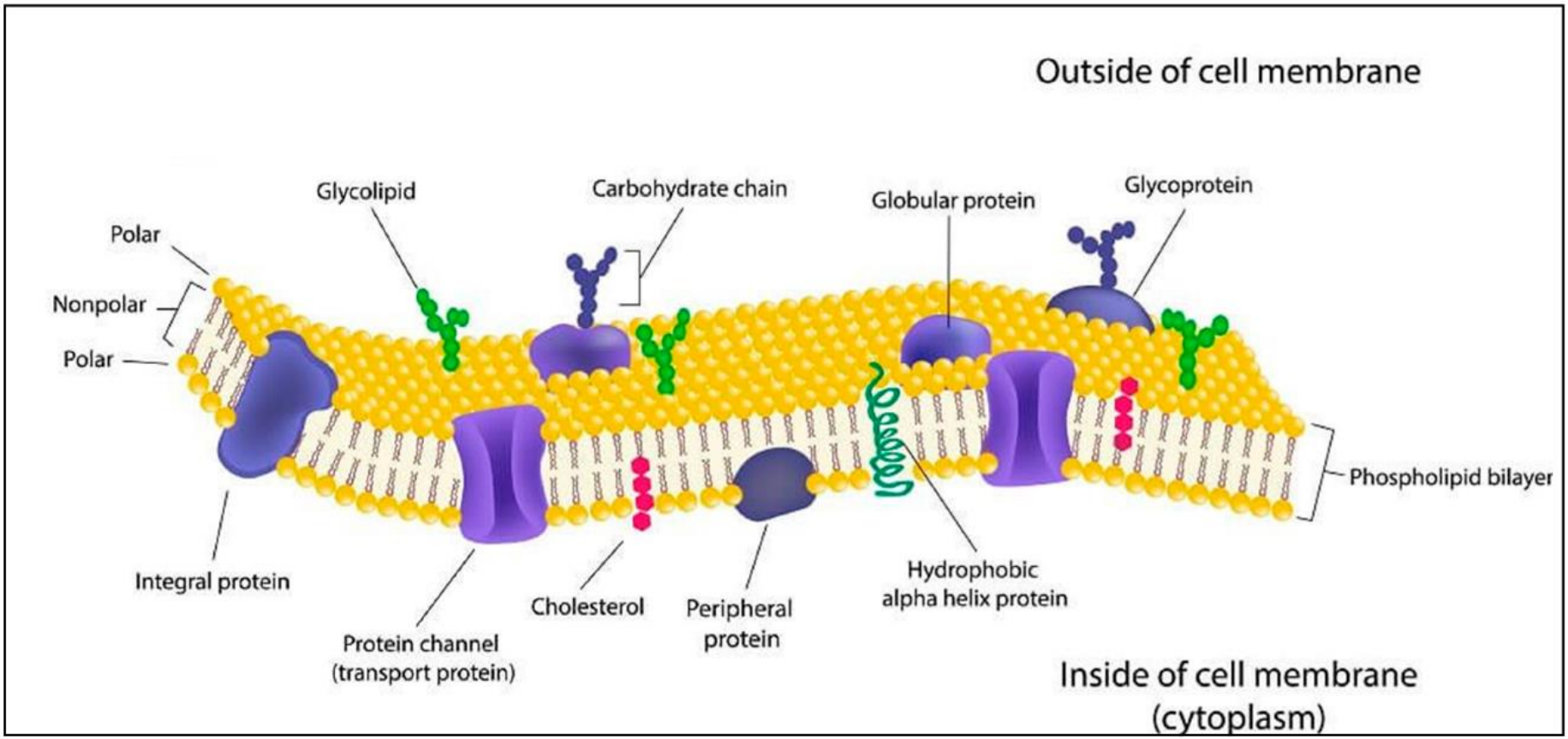
Biological Membranes

- Composition
- Lipids: 50%
- Proteins: 50%
- Carbohydrates

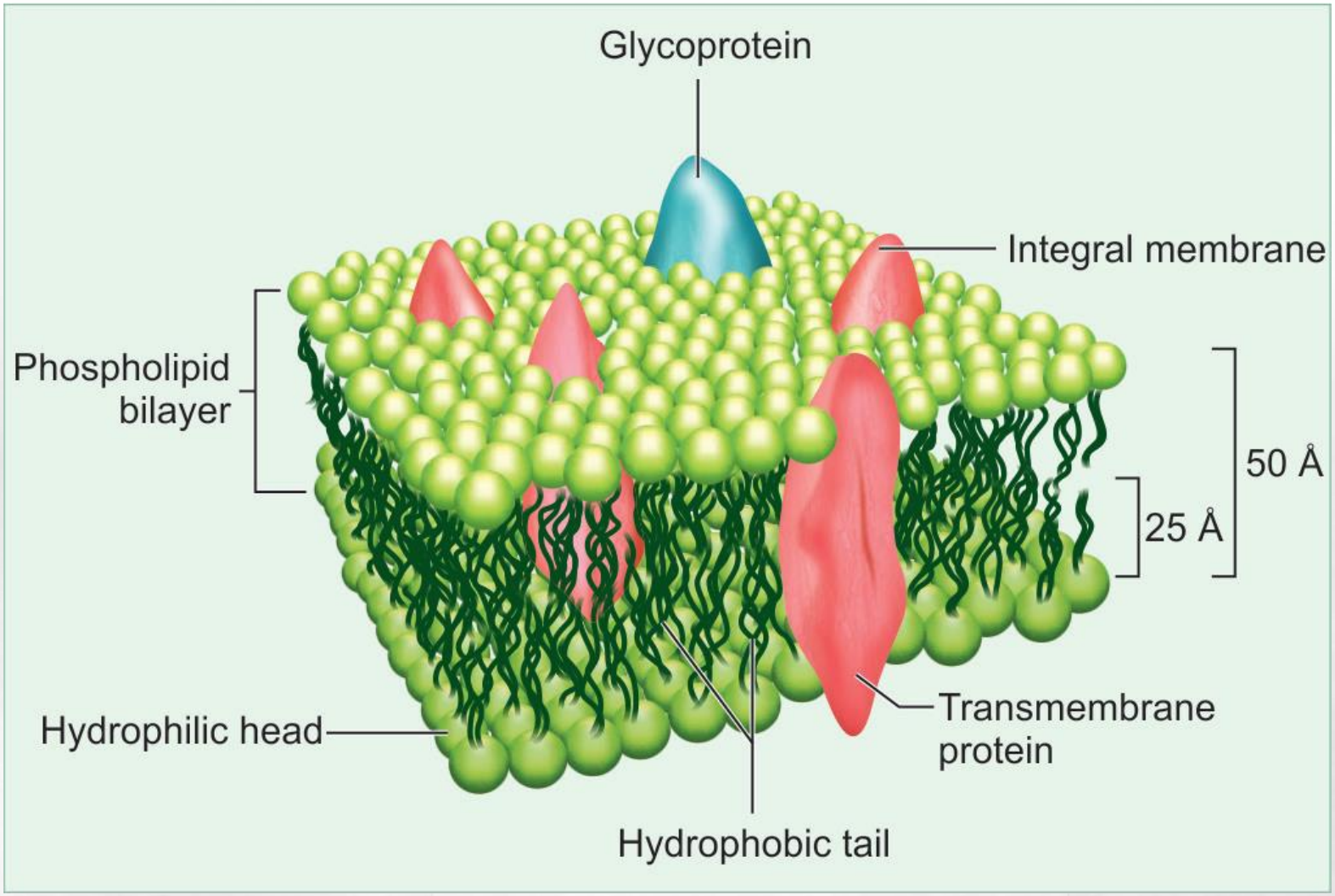


- Trilamilar appearance
- Width: 50 - 80 A^0
- Dynamic structure
- Amphipathic

Composition

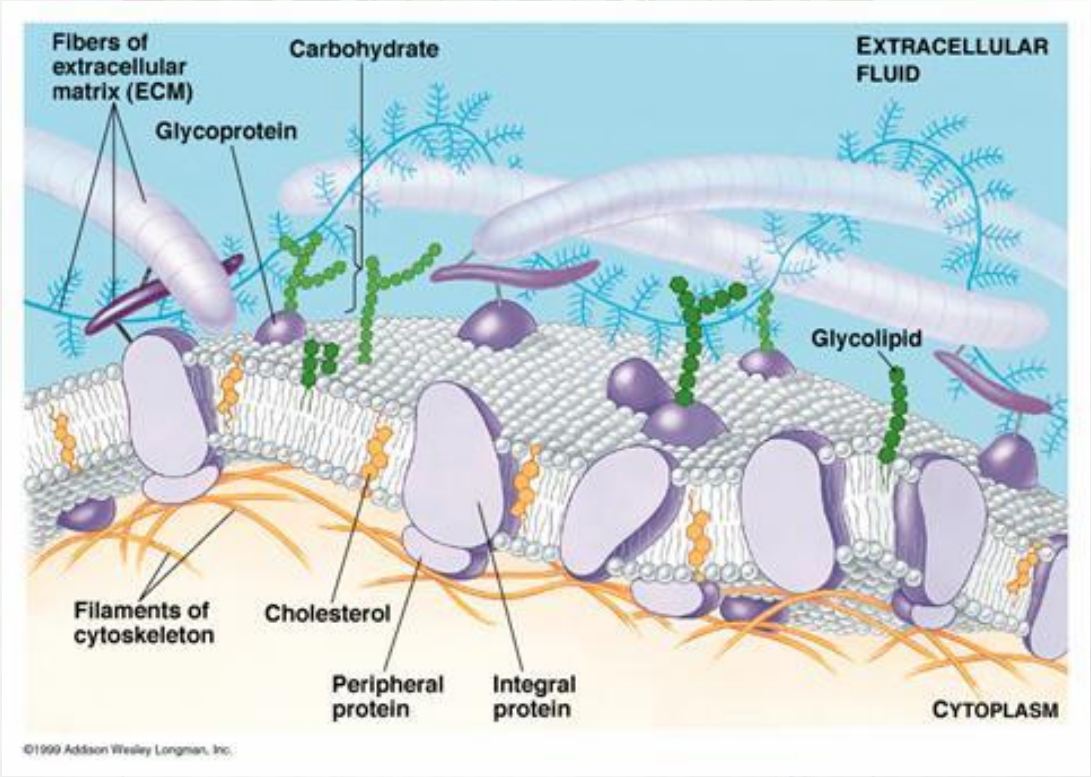


The fluid mosaic model of the membrane.

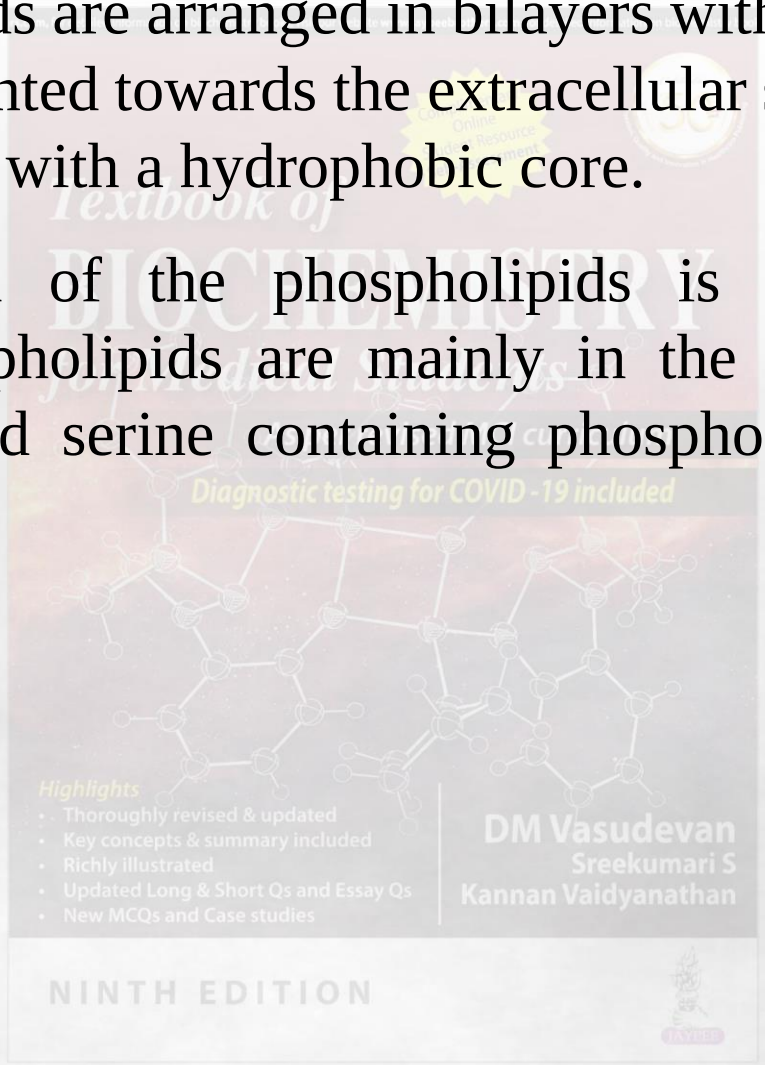

Glycoprotein
Integral membrane
Phospholipid bilayer
50 Å
25 Å
Hydrophilic head
Transmembrane protein
Hydrophobic tail

Plasma Membrane- Nicolson Singer Fluid Mosaic Model

The concept that membranes are fluid with proteins floating in them like icebergs was proposed by Sanger and Nicolson in their "fluid mosaic model".

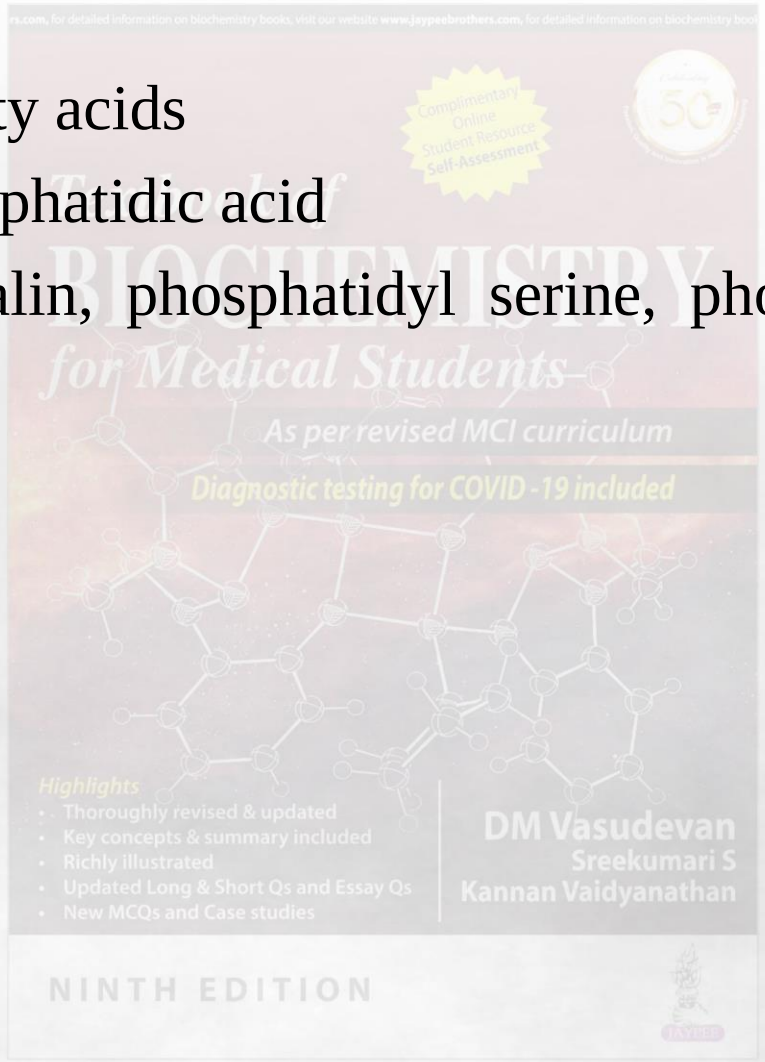


- The phospholipids are arranged in bilayers with the polar head groups oriented towards the extracellular side and the cytoplasmic side with a hydrophobic core.
- The distribution of the phospholipids is such that choline containing phospholipids are mainly in the external layer and ethanolamine and serine containing phospholipids in the inner layer.

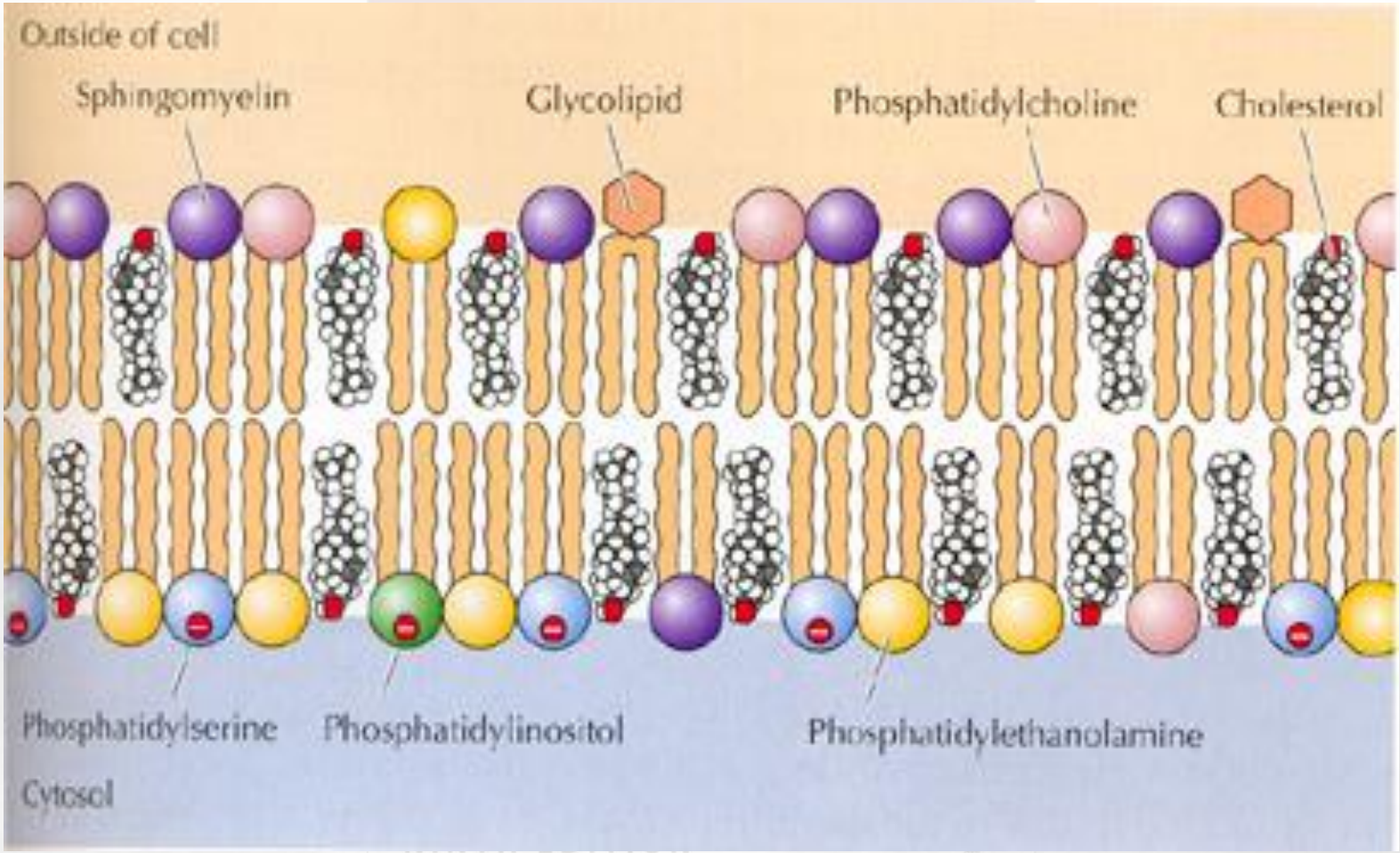


Phospholipids

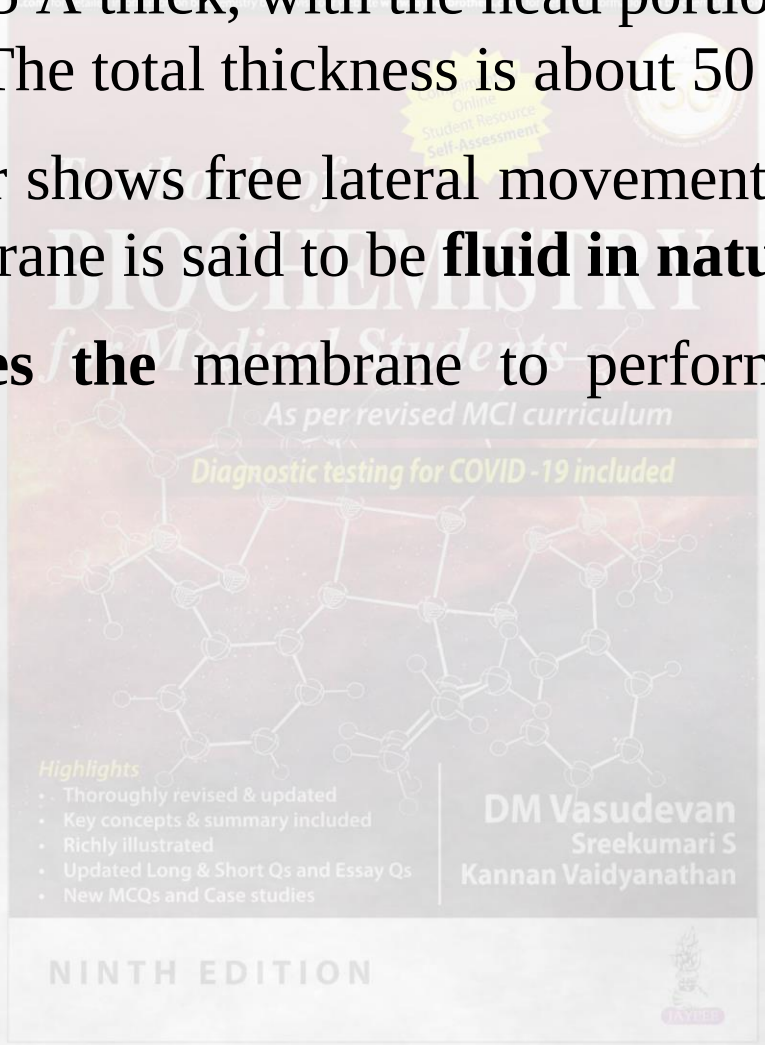
- Most common
- Long chain fatty acids
- Simplest - phosphatidic acid
- Lecithin, cephalin, phosphatidyl serine, phosphatidyl inositol, cardiolipin



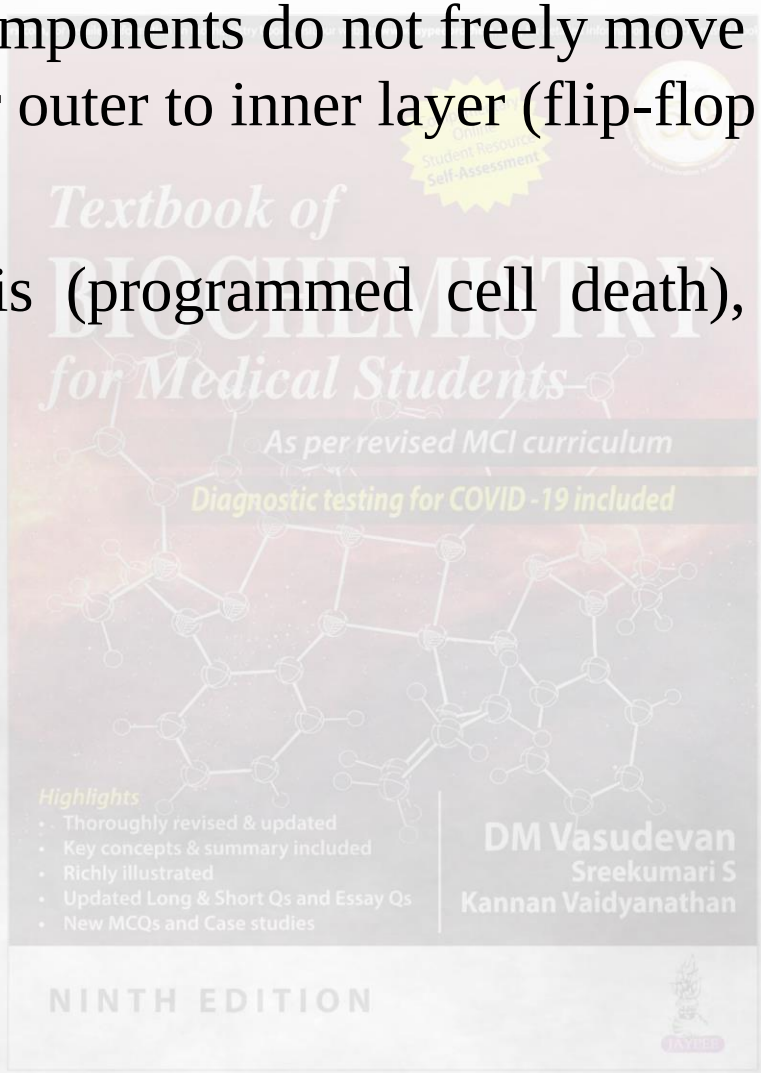
Lipids


Outside of cell
Sphingomyelin
Glycolipid
Phosphatidylcholine
Cholesterol
Phosphatidylserine
Phosphatidylinositol
Phosphatidylethanolamine
Cytosol

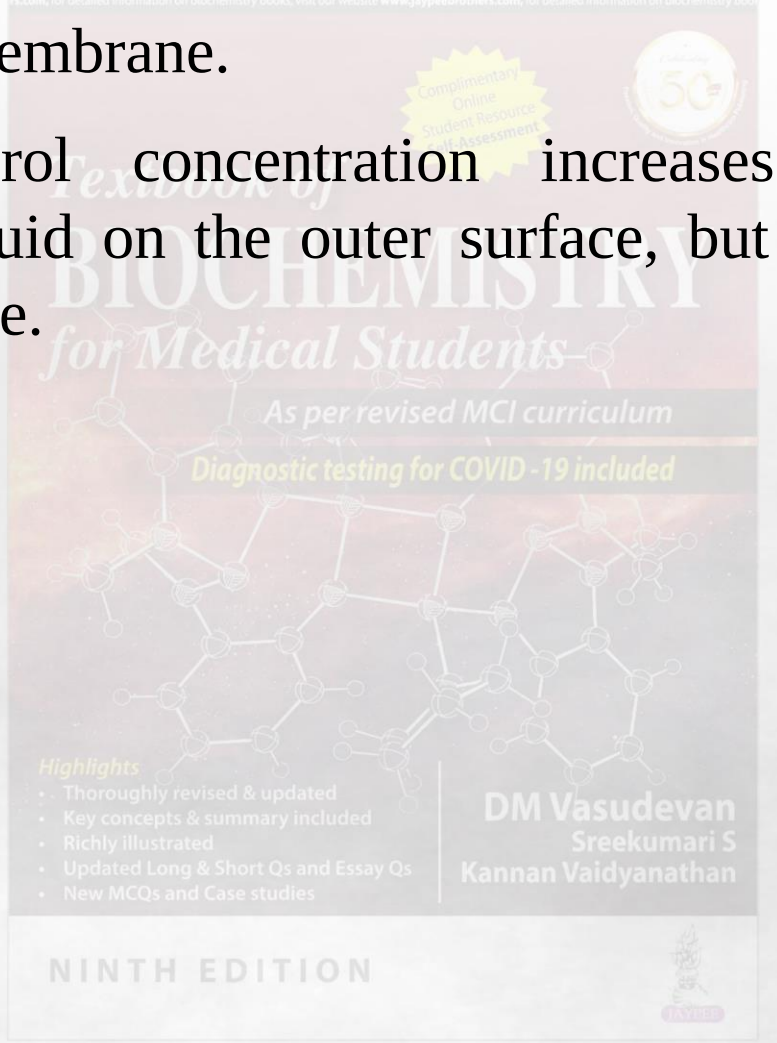
- Each leaflet is 25 Å thick, with the head portion 10 Å and tail 15 Å thick. The total thickness is about 50 to 80 Å.
- The lipid bilayer shows free lateral movement of its components, hence the membrane is said to be **fluid in nature.**
- **Fluidity enables the** membrane to perform endocytosis and exocytosis.



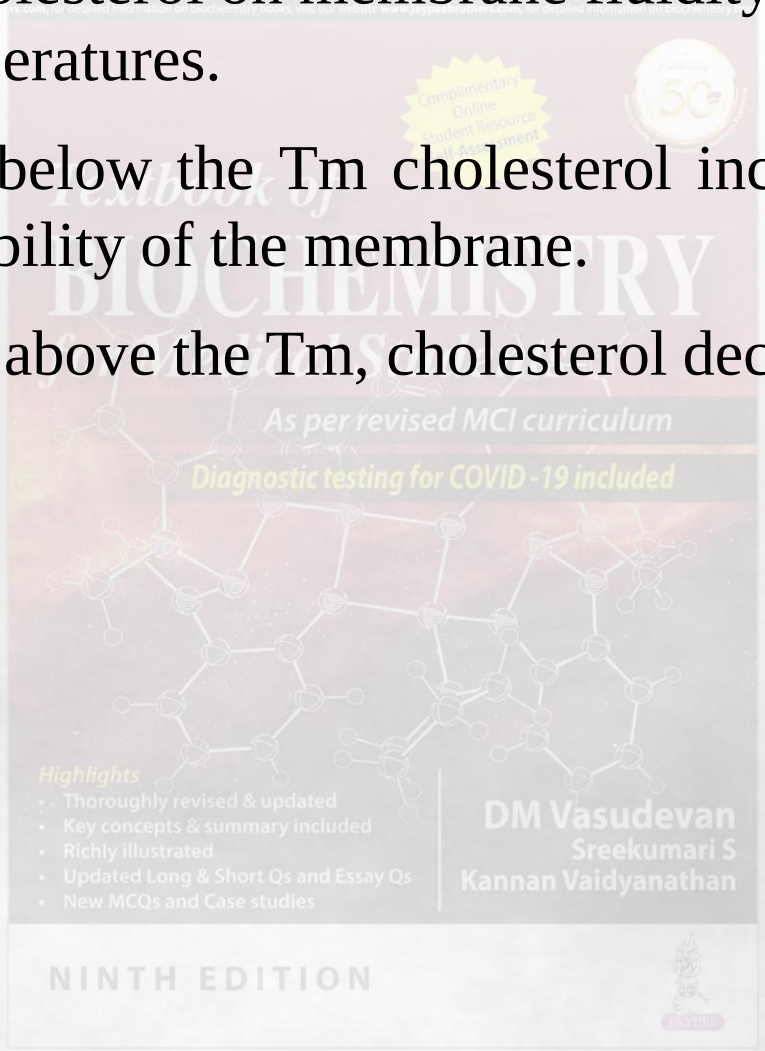
- However, the components do not freely move from inner to outer layer, or outer to inner layer (flip-flop movement is restricted).
- During apoptosis (programmed cell death), flip-flop movement occurs.



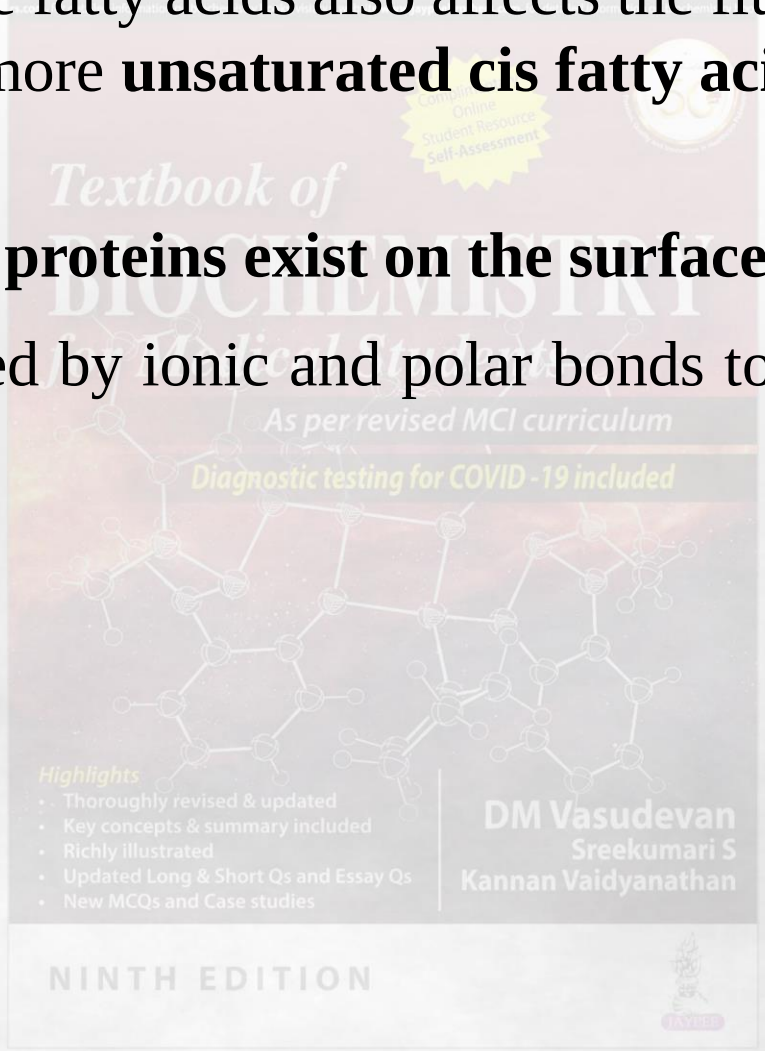
- The **cholesterol content of the membrane** alters the fluidity of the membrane.
- When cholesterol concentration increases, the membrane becomes less fluid on the outer surface, but more fluid in the hydrophobic core.



- The effect of cholesterol on membrane fluidity is different at different temperatures.
- At temperature below the Tm cholesterol increases fluidity and there by permeability of the membrane.
- At temperatures above the Tm, cholesterol decreases fluidity.



- The nature of the fatty acids also affects the fluidity of the membrane, the more **unsaturated cis fatty acids increase the fluidity.**
- **The peripheral proteins exist on the surfaces** of the bilayer.
- They are attached by ionic and polar bonds to polar heads of the lipids.

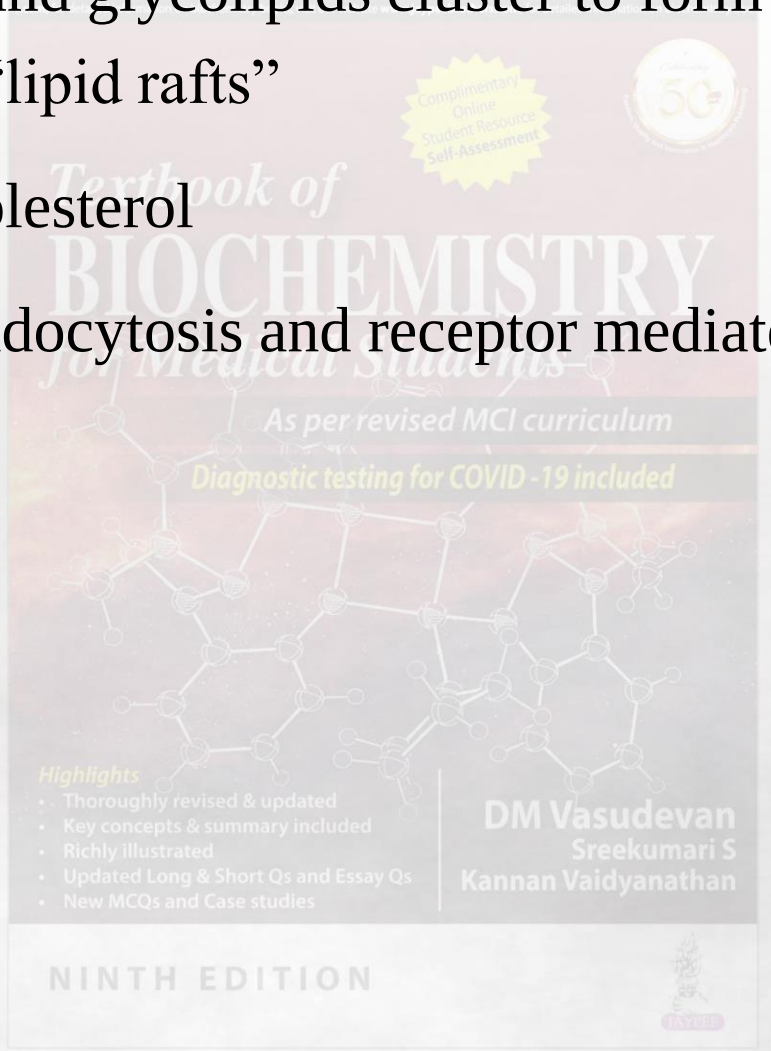


Composition of Different Membranes: Different Lipids as Percentage of Total Lipids

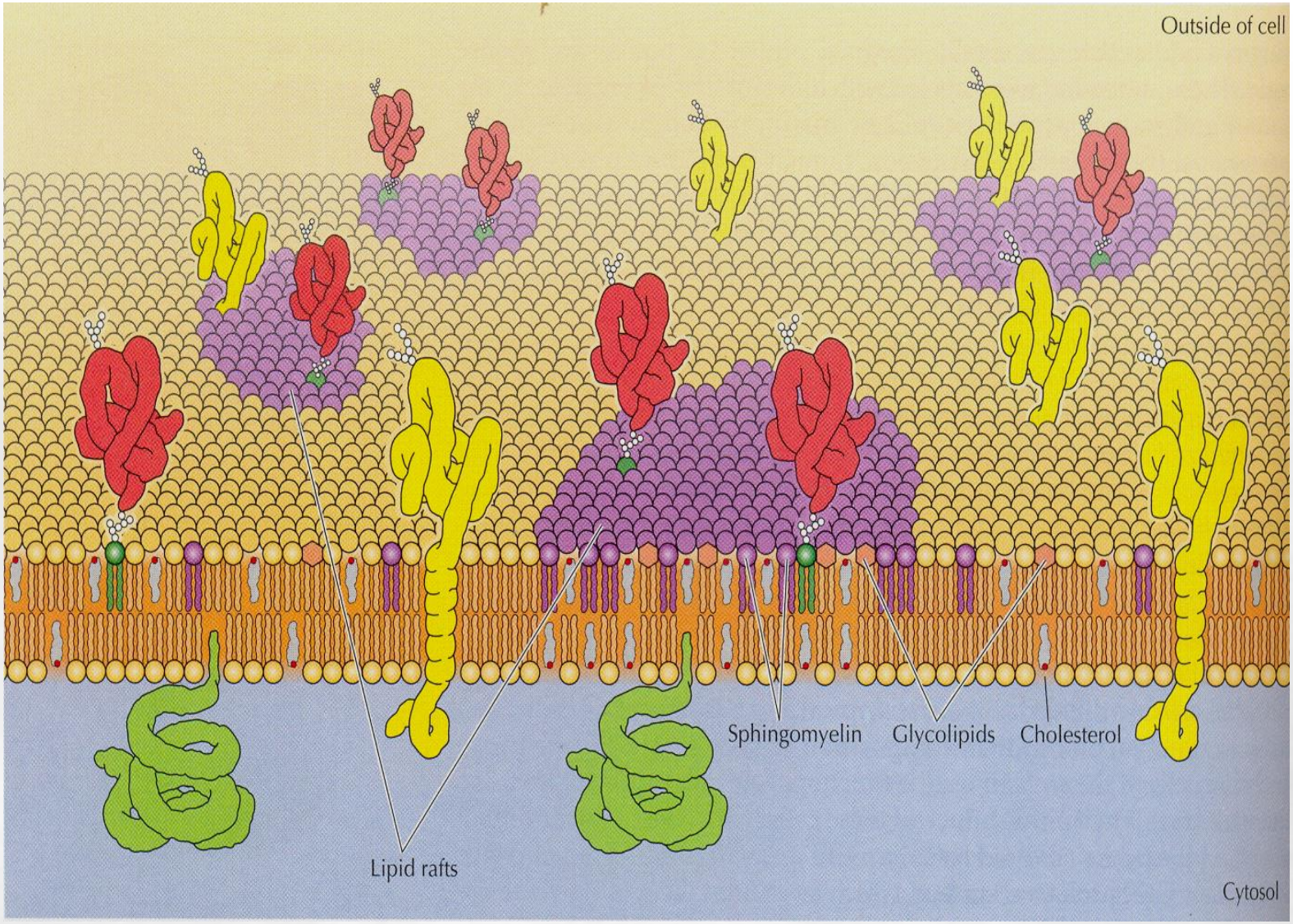
Type of membrane	Various types of lipids					
	Cholesterol	Lecithin	Cephalin	Phosphatidyl Serine	Sphingomyelin	Glycolipid
Plasma membrane	20	19	12	7	12	10
Nuclear membrane	3	45	20	3	2	0
Outer mitochondrial membrane	8	45	20	2	4	0
Inner mitochondrial membrane	0	35	25	0	3	0
Endoplasmic reticulum	5	48	19	4	5	0
Myelin	28	11	17	6	7	29

Lipid Rafts

- Sphingomylin and glycolipids cluster to form semisolid patches called “lipid rafts”
- Enriched in cholesterol
- Specific role endocytosis and receptor mediated signaling

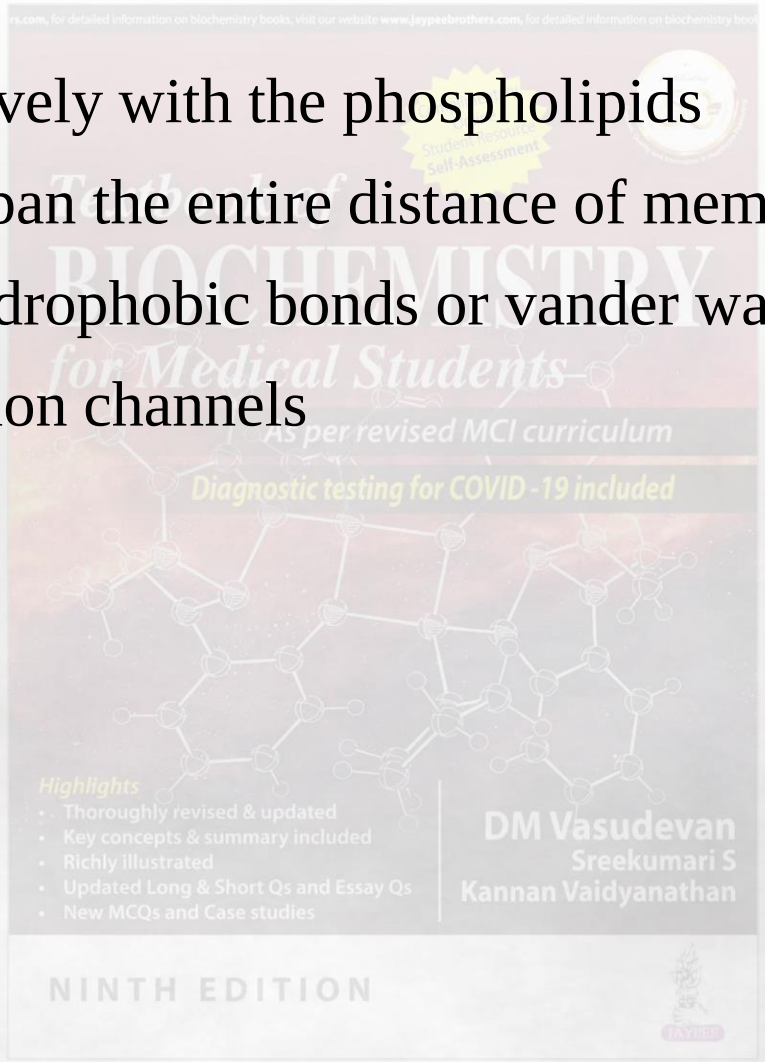


Lipid Rafts

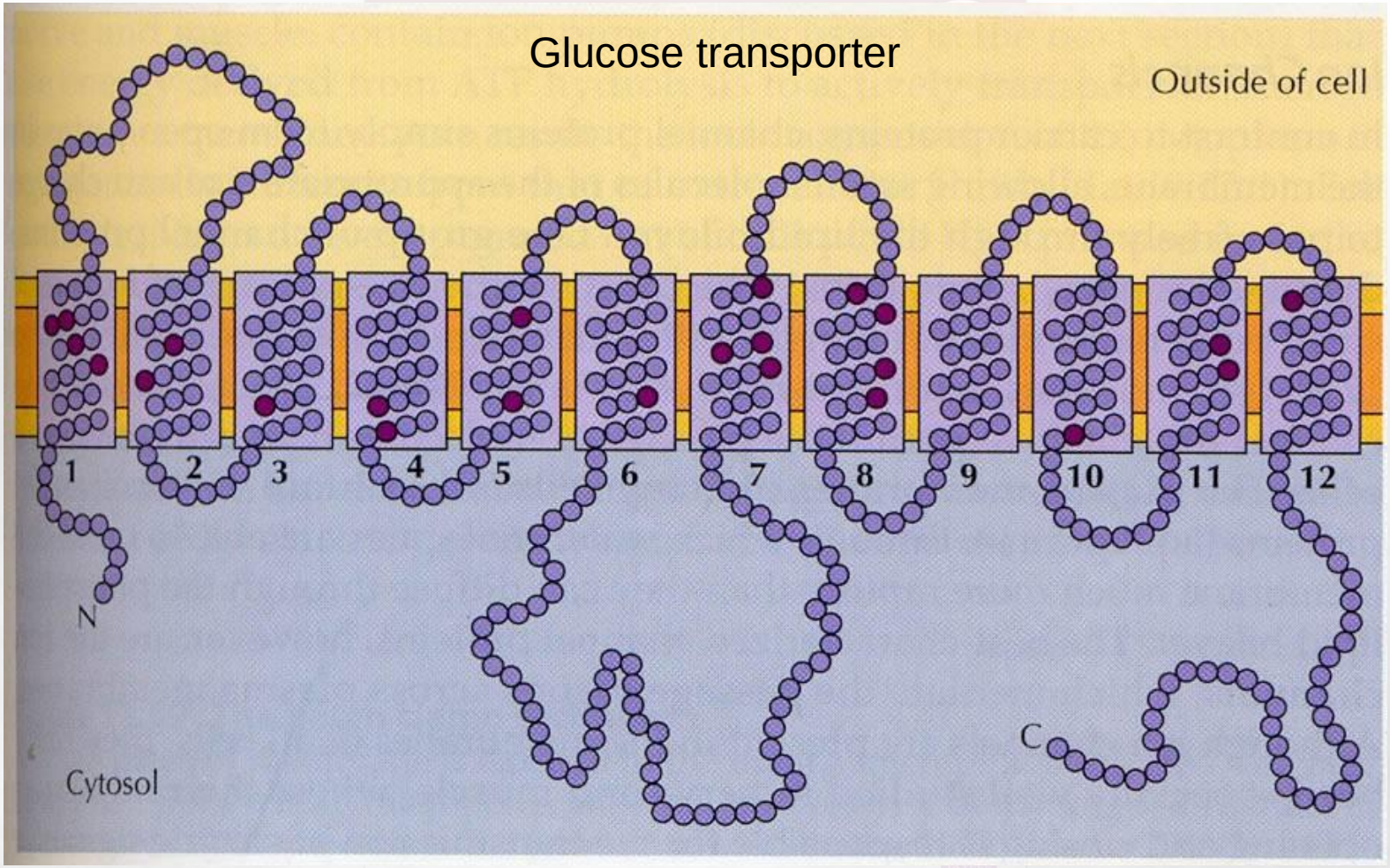

Outside of cell
Sphingomyelin
Glycolipids
Cholesterol
Lipid rafts
Cytosol

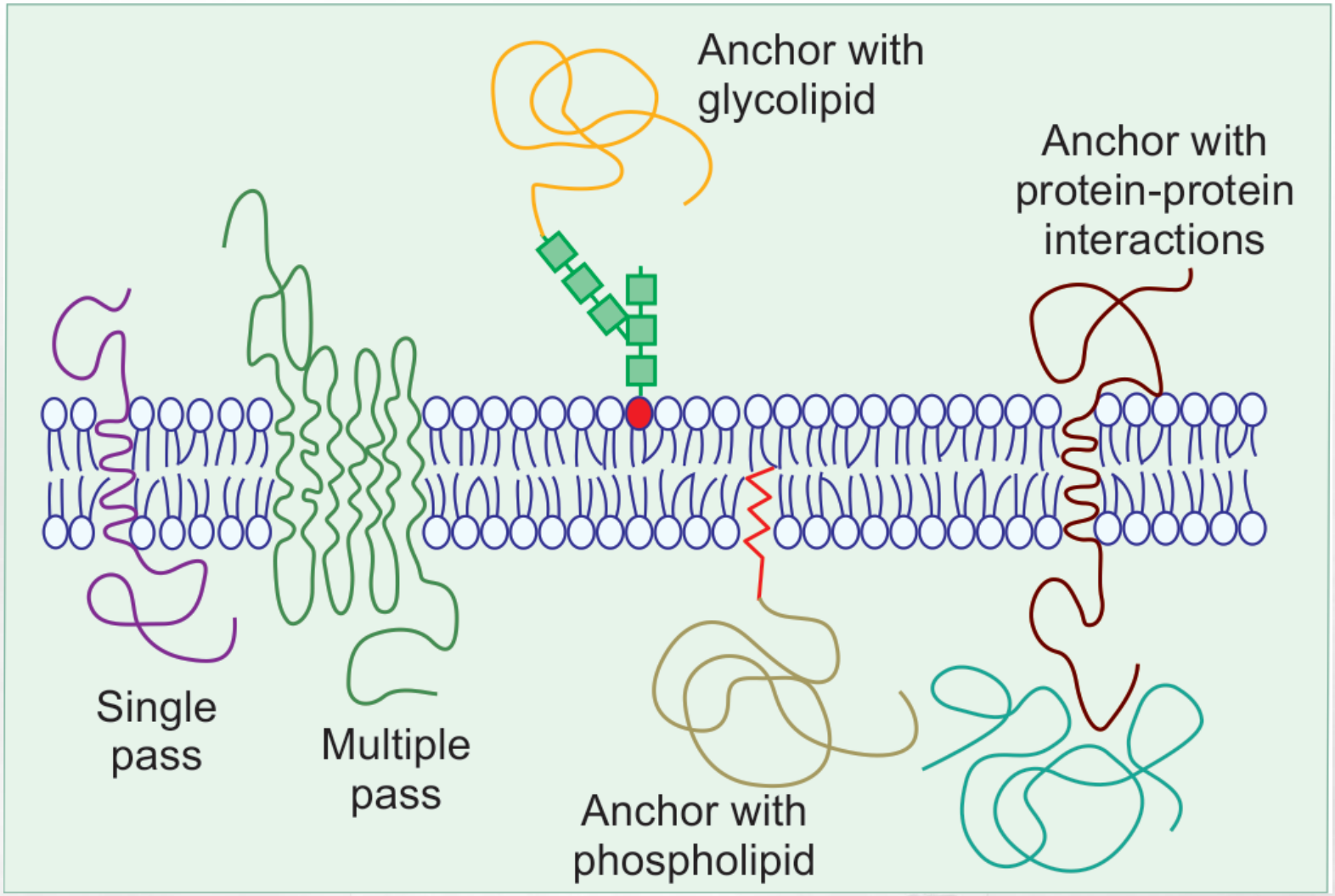
Integral Proteins

- Commonest
- Interact extensively with the phospholipids
- Embedded or span the entire distance of membrane
- Attached by hydrophobic bonds or vander waals forces
- E.g. receptors, ion channels



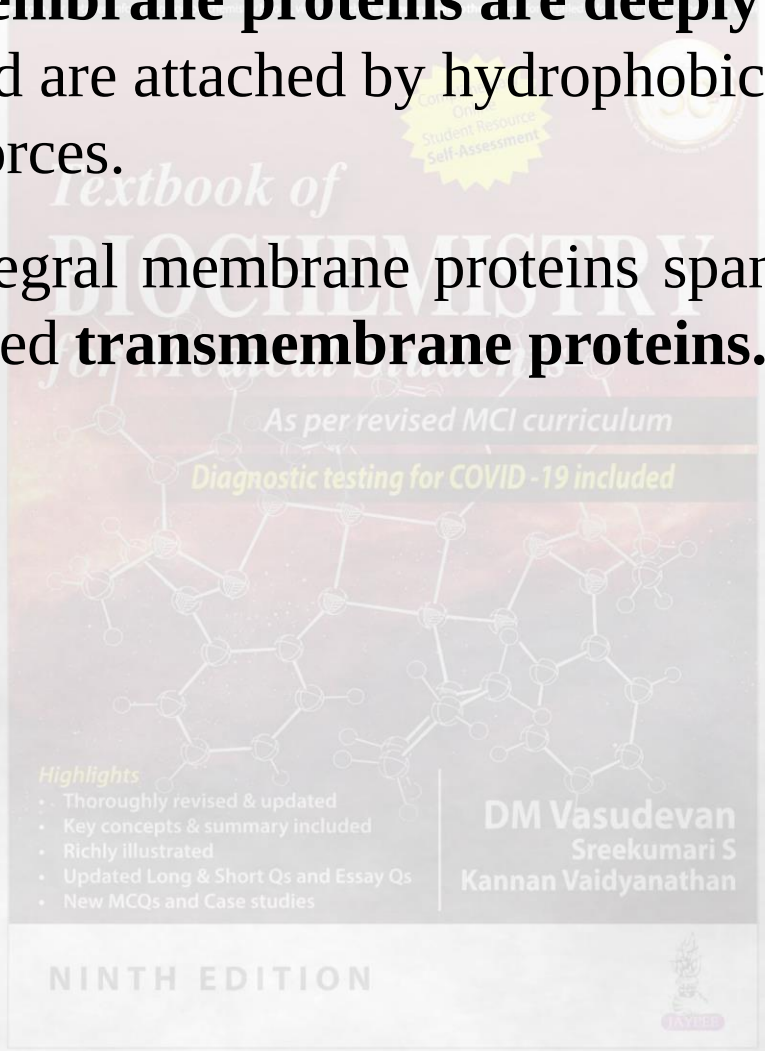
Integral Proteins



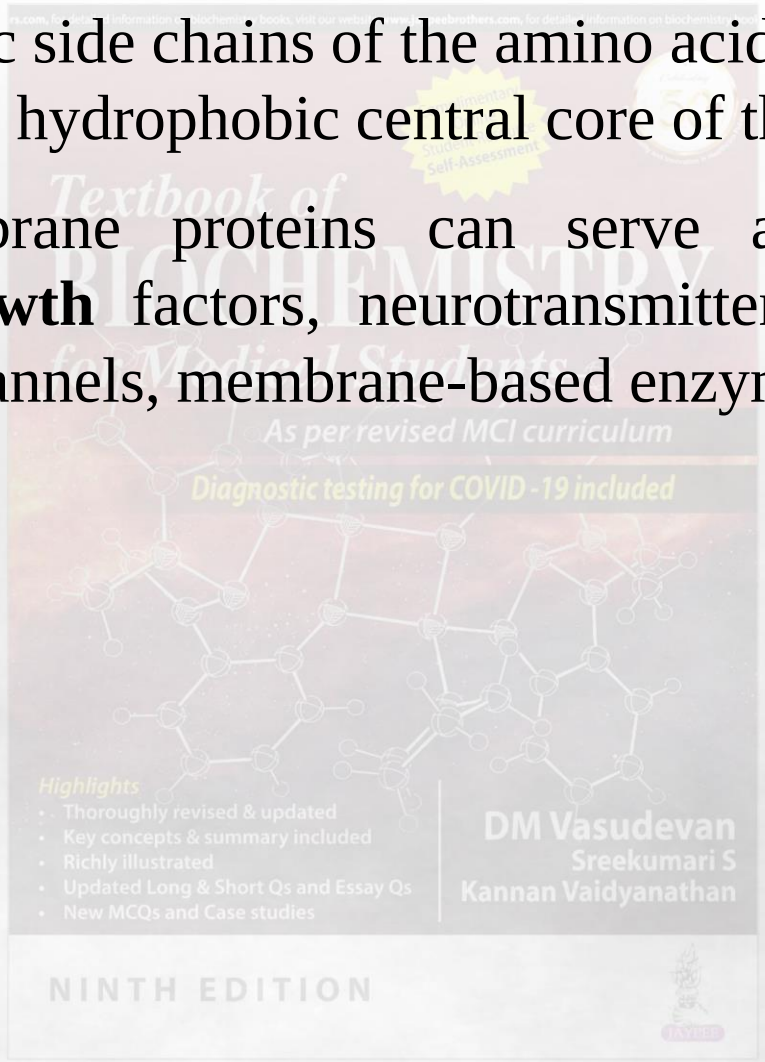


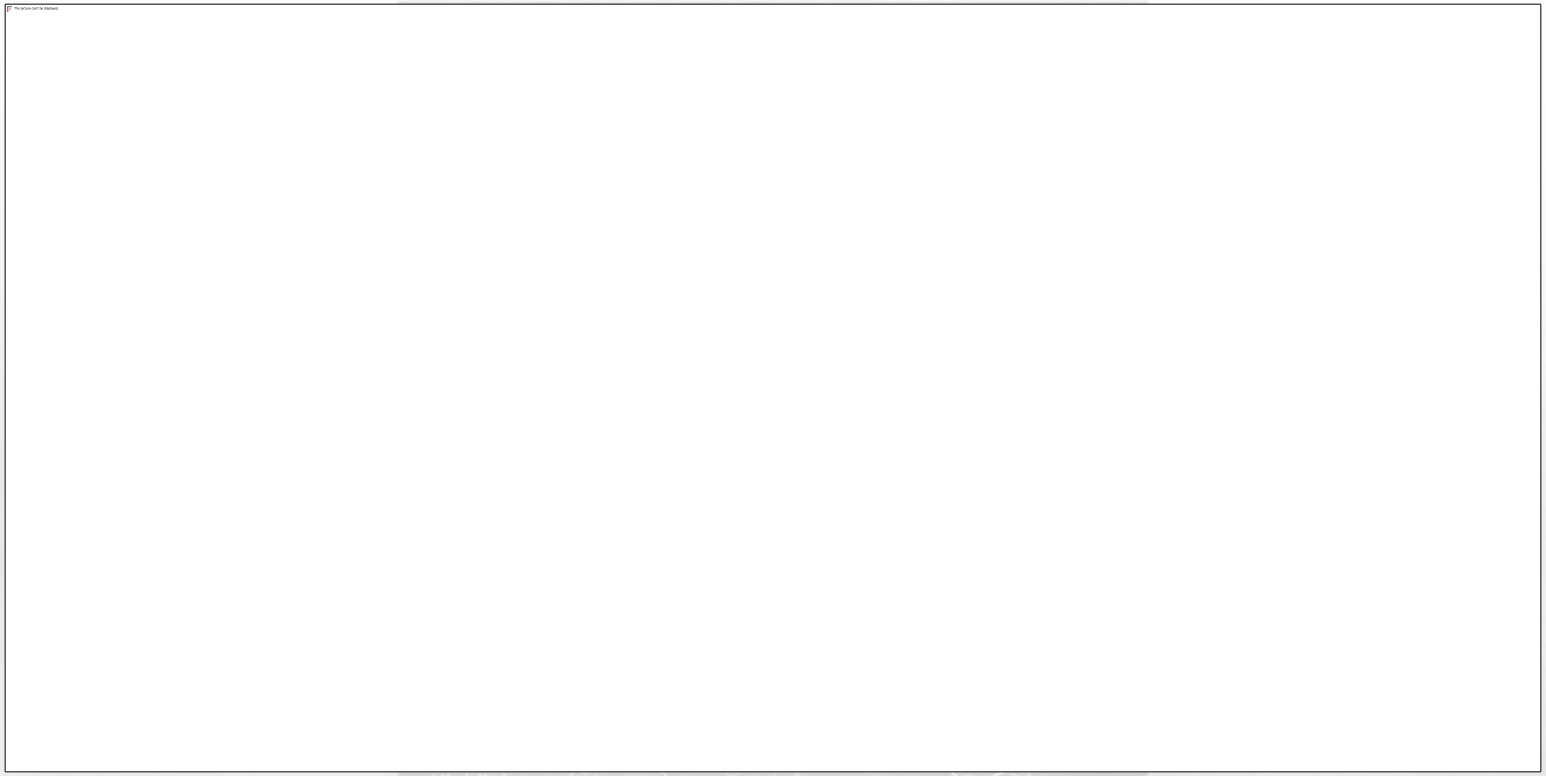
Proteins are anchored in the membrane by different mechanisms

- **The integral membrane proteins are deeply** embedded in the bilayer and are attached by hydrophobic bonds or van der Waals forces.
- Some of the integral membrane proteins span the whole bilayer and they are called **transmembrane proteins.**

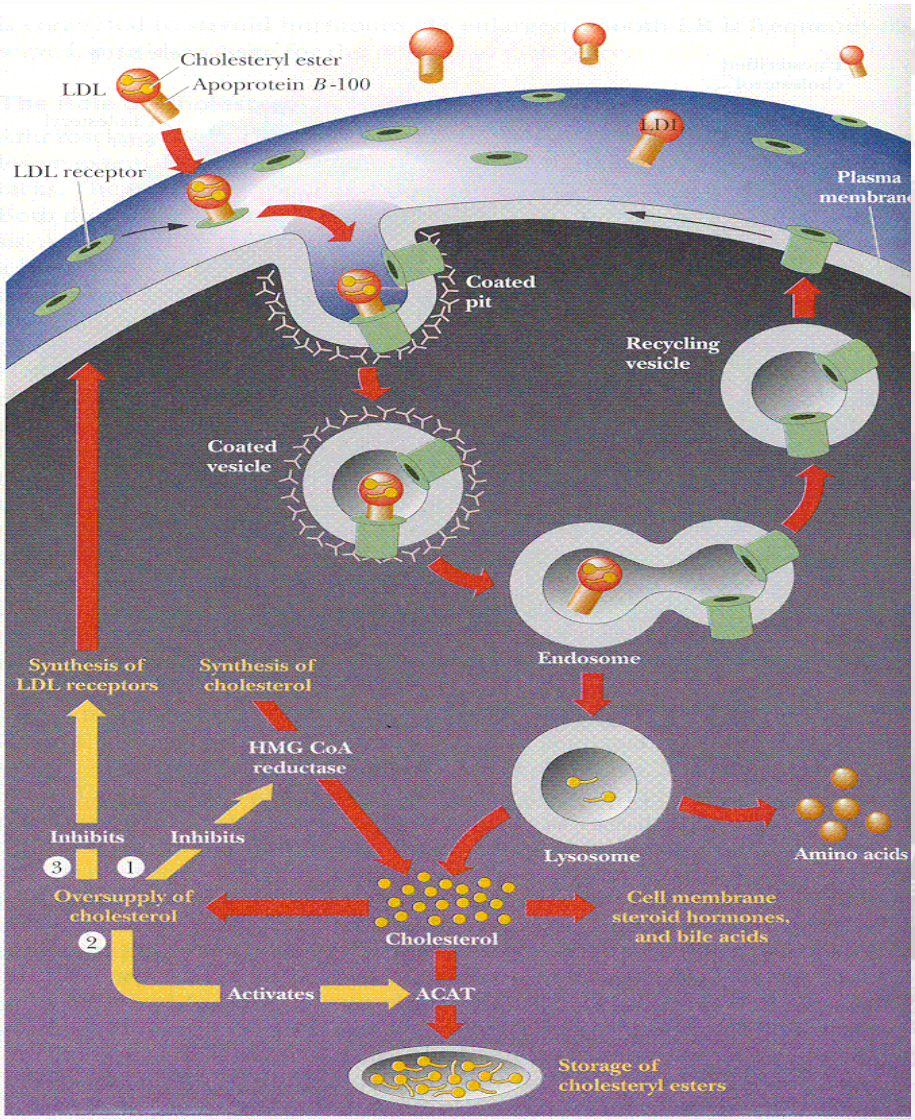


- The hydrophobic side chains of the amino acids are embedded in the hydrophobic central core of the membrane.
- The transmembrane proteins can serve as **receptors (for hormones, growth** factors, neurotransmitters), tissue specific antigens, ion channels, membrane-based enzymes, etc.





- Peripheral membrane proteins
- Integral membrane proteins
- Transmembrane proteins



LDL receptor

The Receptors for Most Growth Factors are Transmembrane Tyrosine-specific Protein Kinases.

These include the receptors for

platelet-derived growth factor (PDGF),

fibroblast growth factors (FGFs),

hepatocyte growth factor (HGF),

insulin,

insulinlike growth factor-1 (IGF-1),

nerve growth factor (NGF),

vascular endothelial growth factor (VEGF),

and *macrophage colony stimulating factor (M-CSF),*

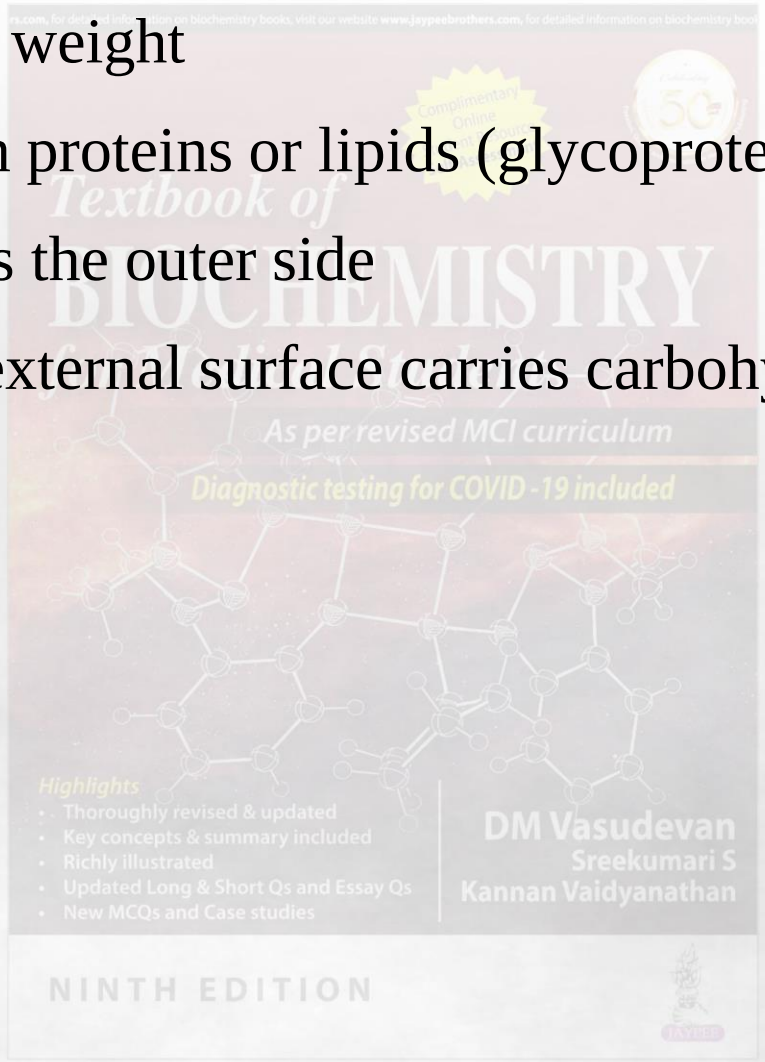
Receptor tyrosine kinases are divided into six families.

Ligand binding causes the EGF receptor to assemble into dimers, which enables the two cytoplasmic domains to cross-phosphorylate each other on multiple tyrosine residues.

autophosphorylation.

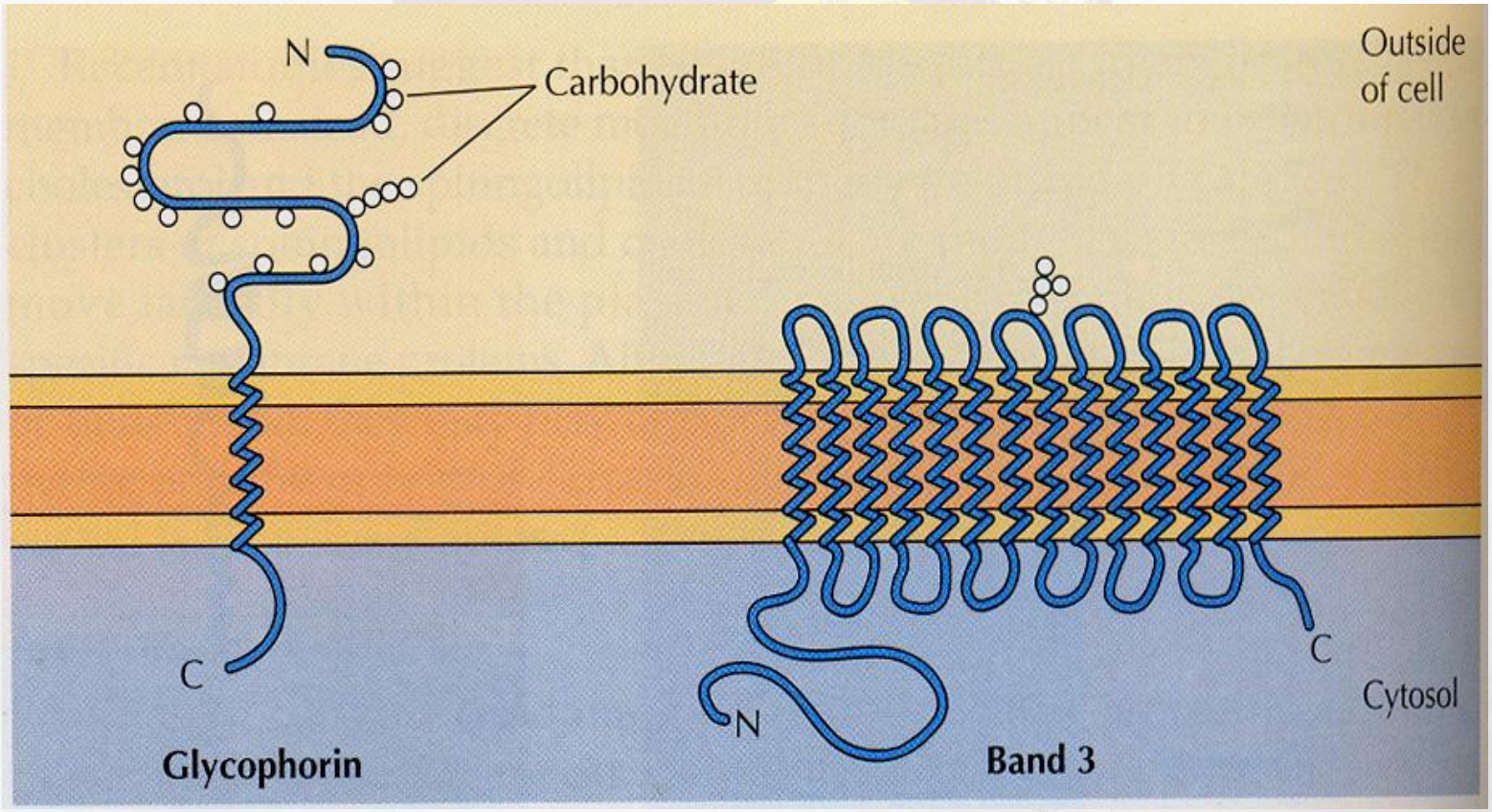
Carbohydrates

- Only 2 - 8% by weight
- Associated with proteins or lipids (glycoproteins or glycolipids)
- Always towards the outer side
- N-terminal on external surface carries carbohydrates
- Glycophorin



Carbohydrates

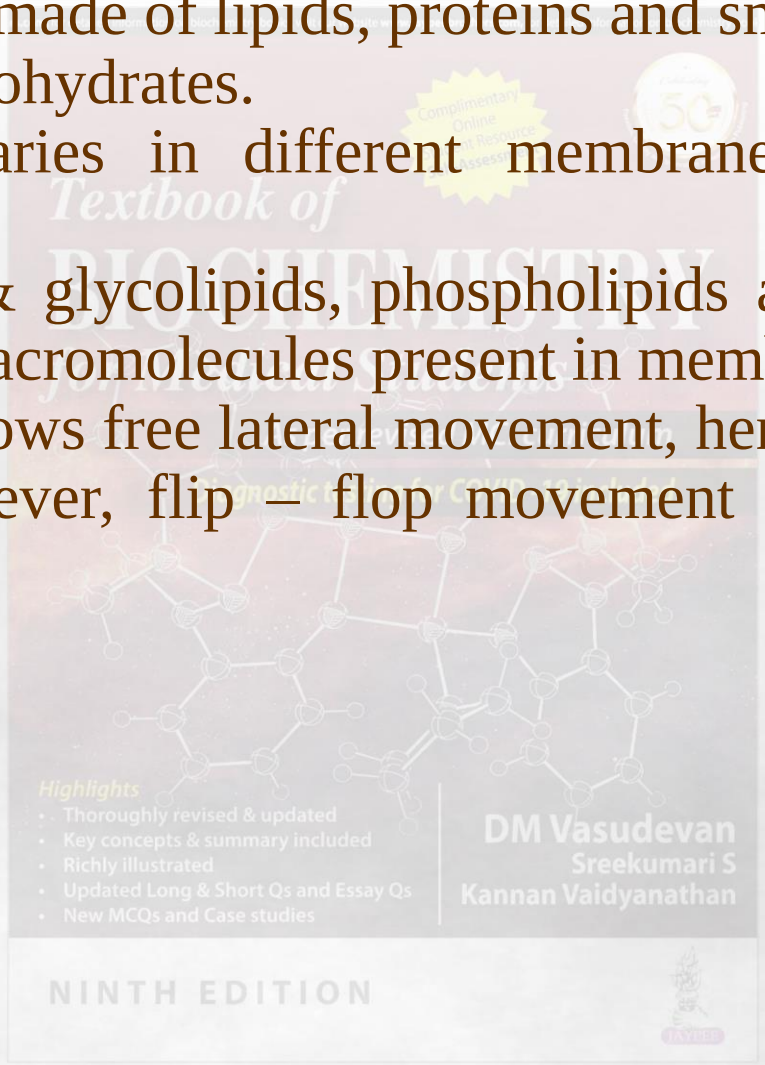
RBC membrane



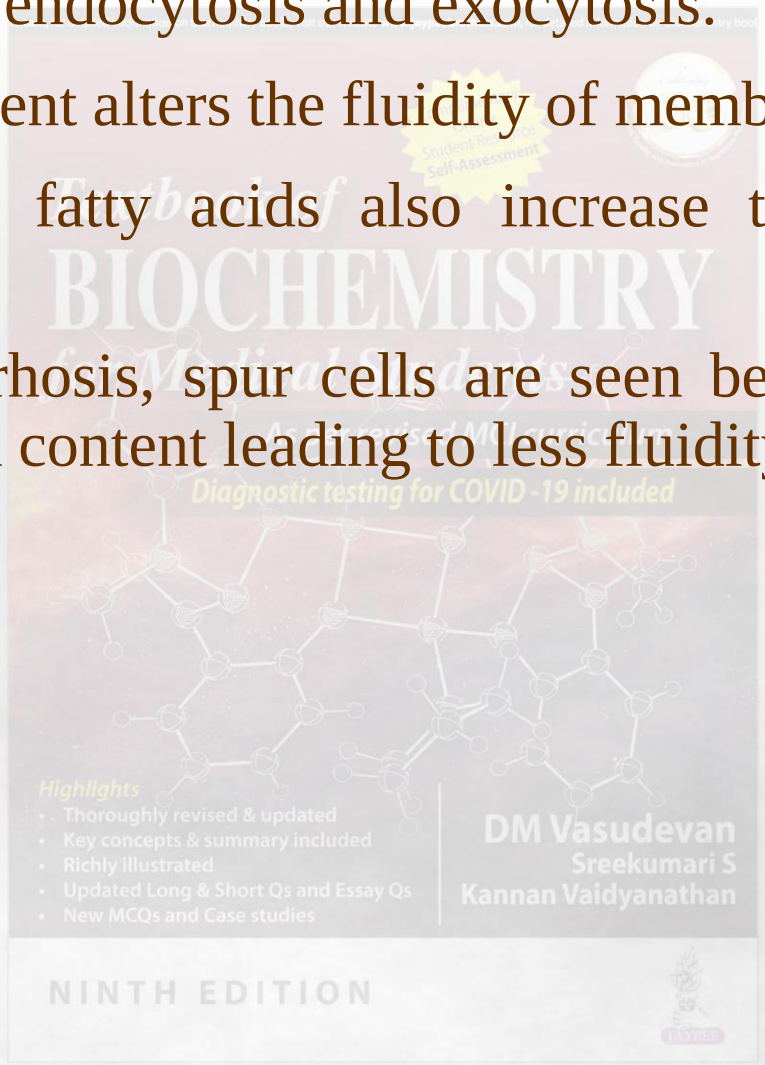
Functions of Plasma Membranes

- Protective sheath
- Selective transport of ions and molecules.
- Recognition of various stimuli.
- Contains receptors for bio molecules like hormones, neurotransmitters etc.
- Cell morphology and movement.
- Compartmentalization
- Membrane is very active metabolically
- Contains the ecto-enzymes 5’ nucleotidase (nucleotide phosphatase) and alkaline phosphatase on the OUTER membrane

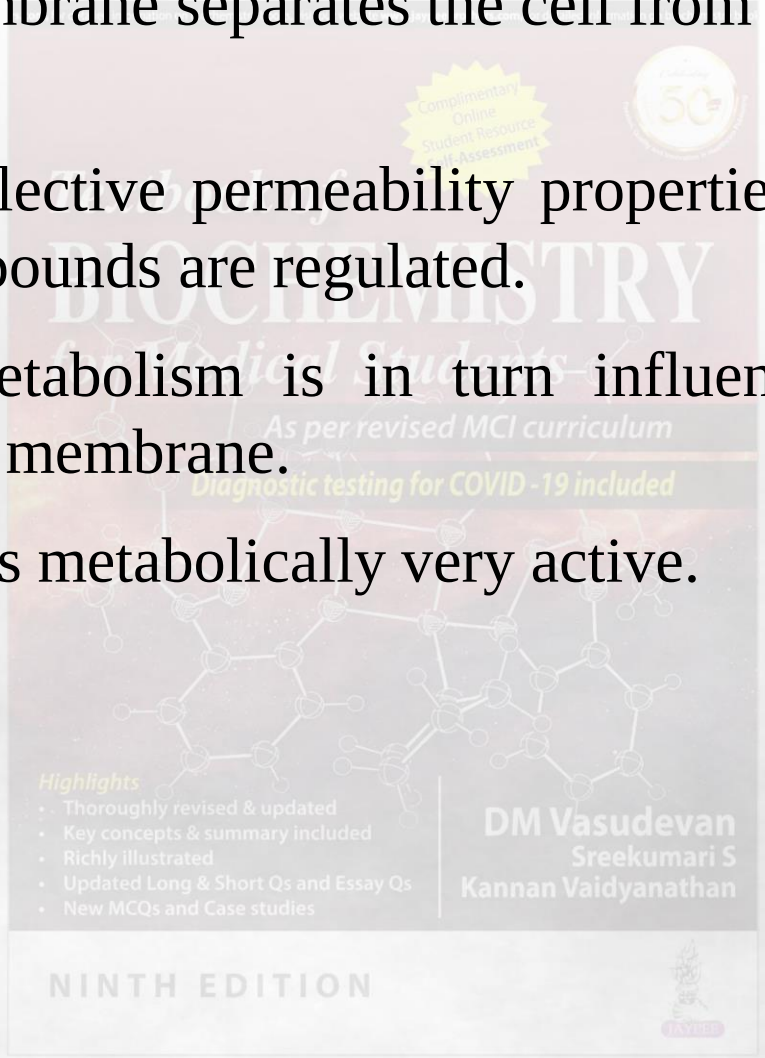
- Membranes are made of lipids, proteins and small amounts of carbohydrates.
- Composition varies in different membranes based on their functions.
- Glycoproteins & glycolipids, phospholipids and cholesterol are the important macromolecules present in membranes.
- Lipid bilayer shows free lateral movement, hence it is called fluid in nature. However, flip – flop movement (inner to outer) is restricted.



- Fluidity enables endocytosis and exocytosis.
- Cholesterol content alters the fluidity of membranes.
- Unsaturated cis fatty acids also increase the fluidity of the membranes.
- In alcoholic cirrhosis, spur cells are seen because of increased RBC cholesterol content leading to less fluidity.

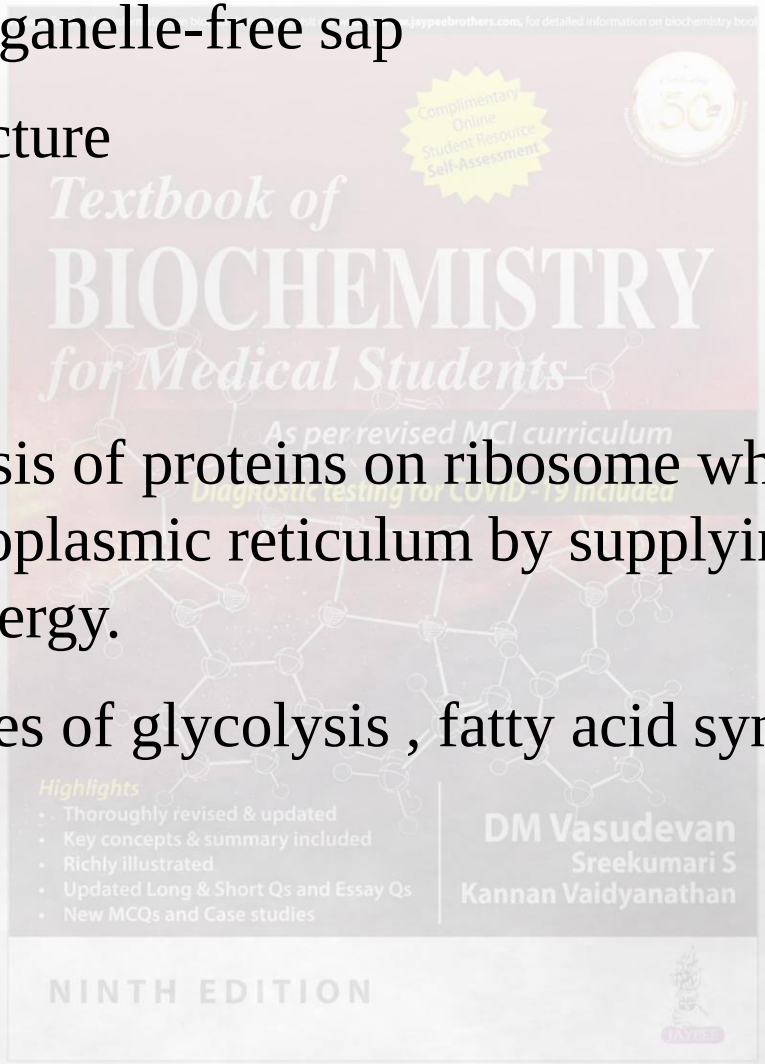


- The plasma membrane separates the cell from the external environment.
- It has highly selective permeability properties so that the entry and exit of compounds are regulated.
- The cellular metabolism is in turn influenced and probably regulated by the membrane.
- The membrane is metabolically very active.



Cytosol Contains Soluble Cellular Components.

- Cytosol is the organelle-free sap
- No specific structure
- Rich in proteins



- Supports synthesis of proteins on ribosome which are free or bound to endoplasmic reticulum by supplying the cofactors and energy.
- Contains enzymes of glycolysis , fatty acid synthesis etc.

Metabolic Functions of Subcellular Organelles

Organelle	*Functions*
Nucleus	DNA replication, transcription
Endoplasmic reticulum	Biosynthesis of proteins, glycoproteins, lipoproteins, drug metabolism, ethanol oxidation, synthesis of cholesterol (partial)
Lysosome	Degradation of proteins, carbohydrates, lipids and nucleotides
Mitochondria	Electron transport chain, ATP generation, TCA cycle, beta oxidation of fatty acids, ketone body production, urea synthesis (part), heme synthesis (part), gluconeogenesis (part), pyrimidine synthesis (part)
Cytosol	Protein synthesis, glycolysis, glycogen metabolism, HMP shunt pathway, transamination, fatty acid synthesis, cholesterol synthesis, heme synthesis (part), urea synthesis (part), pyrimidine synthesis (part), purine synthesis

Comparison of Cell with a Factory

Plasma membrane	Fence with gates; gates open when a message is received
Nucleus	Manager's office
Endoplasmic reticulum	Conveyer belt of production units
Golgi apparatus	Packing units
Lysosomes	Incinerators
Vacuoles	Lorries carrying finished products
Mitochondria	Power generating units