CLASS: I B.Sc Biotech **COURSE NAME: Cell Biology**
COURSE CODE: 18BTU102 **BATCH: 2018-2021**

SEMESTER I

18BTU102	**CELL BIOLOGY**	**4H - 4C**
Total hours/week: L:4 T:0 P:0	**Marks: Internal: 40 External: 60**	**Total: 100**

Scope: Cell biology deals with the morphological and physiological properties of cells, their behaviours, interactions and their environment.

Objective: This paper will enable the students to learn the basics and lay strong foundation in understanding the composition of cells, how cells works is fundamental to living systems.

UNIT- I

Cell: Introduction and classification of organisms by cell structure, cytosol, compartmentalization of eukaryotic cells, cell fractionation. Cell Membrane and Permeability: Chemical components of biological membranes, organization and Fluid Mosaic Model, membrane as a dynamic entity, cell recognition and membrane transport.

UNIT- II

Extracellular Matrix: Composition, molecules that mediate cell adhesion, membrane receptors for extra cellular matrix, macromolecules, regulation of receptor expression and function. Signal transduction.

UNIT- III

Membrane Vacuolar system, cytoskeleton and cell motility: Structure and function of microtubules, Microfilaments, Intermediate filaments. Endoplasmic reticulum: Structure, function including role in protein segregation. Golgi complex: Structure, biogenesis and functions including role in protein secretion.

UNIT-IV

Cell organelles: Lysosomes, Vacuoles and micro bodies: Structure and functions Ribosomes: Structures and function including role in protein synthesis. Mitochondria: Structure and function, Genomes, biogenesis. Chloroplasts: Structure and function, genomes, biogenesis Nucleus: Structure and function, chromosomes and their structure.

UNIT-V

Cell abnormalities: Carcinogenesis, agents promoting carcinogenesis, characteristics and molecular basis of cancer.

References

1. Karp, G. (2013). *Cell and Molecular Biology: Concepts and Experiments* (7th ed.). Hoboken, US: John Wiley & Sons. Inc.
2. Cooper, G.M., & Hausman, R.E. (2013). *The Cell: A Molecular Approach* (6th ed.). Washington, USA: ASM Press & Sunderland, D.C., Sinauer Associates.
3. Becker, W.M., Kleinsmith, L.J., Hardin. J., & Bertoni, G. P. (2009). *The World of the Cell* (7th ed.). San Francisco: Pearson Benjamin Cummings Publishing.
4. De Robertis, E.D.P., & De Robertis, E.M.F. (2006). *Cell and Molecular Biology* (8th ed.). Lippincott Williams and Wilkins, Philadelphia.

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LECTURE PLAN

S.No	Lecture Duration (hr)	Topics to be covered	Support materials
Unit I			**10 hr**
1	1	Introduction to cell biology	T1: 2-19
2	1	Classification of Organisms by cell structure	T2: 42-45
3	1	Cytosol, Compartmentalization of Eukaryotic cells,	T1: 117-324
4	1	Cell fractionation	T1: 118-119
5	1	Cell Membrane and Permeability	T2: 56-59
6	1	Chemical Components of Biological Membrane	T2: 76-110
7	1	Organization and Fluid Mosaic Model	T2: 116-120
8	1	Membrane as a dynamic entity	T2: 121-135
9	1	Cell recognition and Membrane Transport	T1: 143-144
10		Revision	
Unit II			**9 hr**
1	1	Composition of Extracellular Matrix	T3: 273- 275
2	1	Molecules that Mediate Cell adhesion	T3: 275-280
3	1	Membrane Receptor for Extracellular Matrix	T3: 275-280
4	1	Macromolecules	T3: 280-289
5	1	Regulation of Receptor Expression and function	T3: 583-600
6	1	Signal Transduction	T3:600-629
7	1	Macromolecules in Signal Transduction	T3:600-629
8	1	Revision	
9	1	Unit test	
Unit III			**8 hr**

1	1	Membrane Vascular System, Cytoskeleton and Cell motility	T2: 293-309
2	1	Structure and Function of Microtubules, Microfilaments	T2: 293-309
3	1	Intermediate Filaments	T2: 303
4	1	Endoplasmic Reticulum: Structure and Function	T3: 69-70
5	1	Endoplasmic Reticulum role in Protein Segregation	T3: 71-72
6	1	Golgi Complex: Structure, and function	T2: 166-170
7	1	Biogenesis role in Protein Secretion	T2: 171-174
8	1	Revisions	
Unit IV			**13 hr**
1	1	Cell Organells: Lysosomes	T2: 175-180
2	1	Vacuoles	T2: 180-181
3	1	Microbodies- Structure and Function	T2: 182-183
4	1	Ribosomes – Structure	T3: 68
5	1	Ribosomes – Function	T3: 69
6	1	Mitochondira: Structure	T2: 191-195
7	1	Mitochondira: Function	T2: 195-200
8	1	Mitochondira: Genome & biogenesis	T2: 200-219
9	1	Chloroplasts: Structure and Function	T2: 220-230
10	1	Genome & biogenesis	T2: 231-242
11	1	Nucleus: Structure and Function	T2: 243-256
12	1	Chromosome: Structure and Function	T2: 257-279
13	1	Revisions	
Unit V			**8hr**

1	1	Cell Abnormalities	T4: 1-5
2	1	Carcinogensis	T4: 7,8
3	1	Agents Promoting Carcinogenesis	T4: 10-12
4	1	Cancer -Characteristics	T4: 14
5	1	Molecular Basis of Cancer	T4:14-15
6	1	Revisions	
7	1	ESE question paper revision	
8	1	ESE question paper revision	
Total Hours = (Unit I+ II + III + IV + V) = 48 hr			

UNIT-1

SYLLABUS

Cell: Introduction and classification of organism by cell structure, cytosol, compartmentalization of eukaryotic cells, cell fractionation. Cell membrane and permeability, chemical components of biological membrane, organization and fluid mosaic model, membrane as a dynamic entity, cell recognition and membrane transport.

Cell

The cell is the basic unit of organization or structure of all living matter.

History

- The cell was discovered by Robert Hooke in 1665.
- He examined very thin slices of cork and saw a multitude of tiny pores.
- He remarked that it looked like the walled compartments of a honeycomb, so he called them cells.
- However, Hooke did not know their real structure or function.
- His cell observations gave no indication of the nucleus and other organelles found in most living cells.

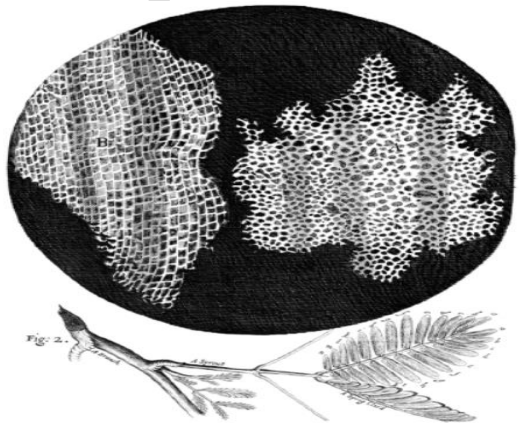

Fig: Drawing of the structure of cork by Robert Hooke that appeared in Micrographia.

"The cell is the fundamental element of organization"

The bservations of Hooke, Leeuwenhoek, Schleiden, Schwann, Virchow, and others led to the development of the cell theory.

The cell theory states:

- All living things or organisms are made of cells.
- New cells are created by old cells dividing into two.
- Cells are the basic building units of life.

Modern interpretation:

The generally accepted parts of modern cell theory include:

1. The cell is the fundamental unit of structure and function in living organisms.
2. All cells arise from pre-existing cells by division.
3. Energy flow (metabolism and biochemistry) occurs within cells.
4. Cells contain hereditary information (DNA) which is passed from cell to cell during cell division.
5. All cells are basically the same in chemical composition in organisms of similar species.
6. All known living things are made up of one or more cells.
7. Some organisms are made up of only one cell and are known as unicellular organisms.
8. Others are multicellular, composed of a number of cells.
9. The activity of an organism depends on the total activity of independent cells.

Exceptions

1. Viruses are considered alive by some, yet they are not made up of cells. Viruses have many features of life, but by definition of the cell theory, they are not alive.
2. The first cell did not originate from a pre-existing cell. There was no exact first cell since the definition of cell is
3. imprecise.
4. Mitochondria and chloroplasts have their own genetic material, and reproduce independently from the rest of the cell.

Classification of cell types

1. Prokaryotes :

- The prokaryotic *(Greek; pro = primitive or before; karyon = nucleus)* are small, simple and most primitive.

- Prokaryotes lack a nucleus *(though they do have circular DNA)* and other membrane-bound organelles *(though they do contain ribosomes)*.
- Bacteria and Archaea are two domains of prokaryotes.

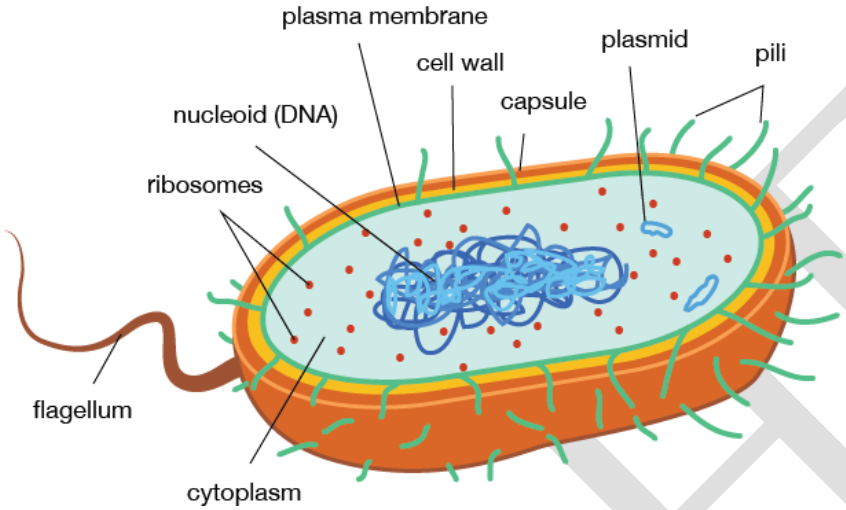

Fig: Schematic diagram of a prokaryotic cell.

Flagella:

- Long, whip-like protrusion found in most prokaryotes that aids in cellular locomotion.
- It also often functions as a sensory organelle, being sensitive to chemicals and temperatures outside the cell.

Capsule:

- It is found in some bacterial cells.
- This additional outer covering protects the cell when it is engulfed by phagocytes and by viruses.
- Assists in retaining moisture, and helps the cell stick to to surfaces and nutrients.
- The capsule is found most commonly among Gram-negative bacteria.
- Examples- *Escherichia coli (E.coli), Salmonella* etc.
- Examples of Gram positive bacteria -*Streptococcus pneumoniae, Streptococcus pyogenes* etc.

Cell wall:

- It is the outermost layer - protects the bacterial cell and gives it shape.
- One exception - Mycoplasma lacks cell wall.
- Bacterial cell walls are made of peptidoglycan which is made from polysaccharide chains cross-linked by unusual peptides containing D-amino acids.
- The antibiotic penicillin is able to kill bacteria by preventing the cross-linking of peptidoglycan and this causes the cell wall to weaken.

- There are two different types of cell wall in bacteria, called Gram-positive and Gram-negative. The names originate from the reaction of cells to the Gram stain, a test long-employed for the classification of bacterial species.
- Gram-positive bacteria possess a thick cell wall containing many layers of peptidoglycan and teichoic acids.
- Gram-negative bacteria have a relatively thin cell wall consisting of a few layers of peptidoglycan surrounded by a second lipid membrane containing lipopolysaccharides and lipoproteins.

Cell membrane:

Cell membrane surrounds the cell's cytoplasm and regulates the flow of substances in and out of the cell.

Cytoplasm:

The cytoplasm of a cell is a fluid in nature that fills the cell and is composed mainly of 80% water that also contains enzymes, salts, cell organelles, and various organic molecules.

Cytosol:

(Gel like fluid other than nucleoid)

- The plasma membrane is followed by the colloidal organic fluid called ***matrix*** or***cytosol***.
- The cytosol is the aqueous portion of the ***cytoplasm***(the extra-nuclear protoplasm) and of the***nucleoplasm***(the nuclear protoplasm).
- It fills all the spaces of the cell and constitutes its true ***internal milieu***.
- Cytosol is particularly rich in differentiating cells and many fundamental properties of cell arebecause of this part of the cytoplasm.
- The cytosol serves to dissolve or suspend the great variety of small molecules concerned with cellular metabolism, *e.g.*, glucose, amino acids, nucleotides, vitamins, minerals, oxygen and ions.

Ribosomes:

Ribosomes are the organelles of the cell responsible for protein synthesis.

Nucleiod Region:

- The nucleoid region is possessed by a prokaryotic bacterial cell.
- It is the area of the cytoplasm that contains the bacterial DNA molecule.

Plasmids:

*(The term plasmid was first introduced by the American molecular biologist **Joshua Lederberg** in **1952**.)*

- Many species of bacteria also may carry extrachromosomal genetic elements in the form of small, circular and closed DNA molecules

- Plasmids usually occur naturally in bacteria, but are sometimes found in eukaryotic organisms. Their sizes vary from 1 to over 1,000 kbp.

2. Eukaryotes:

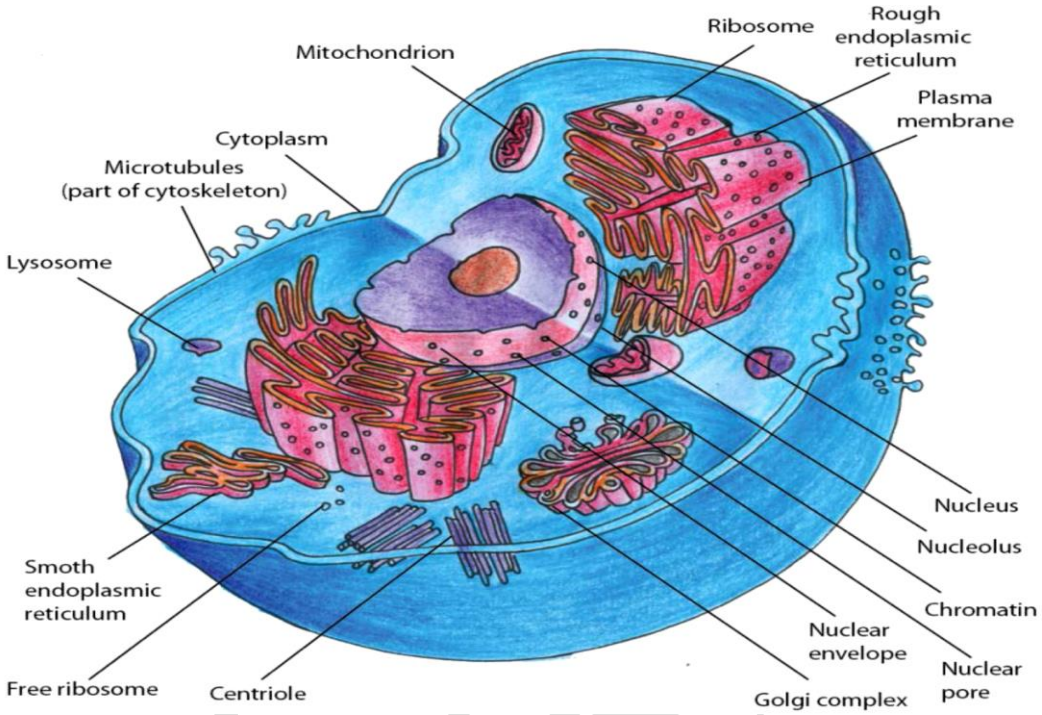

Fig: Eukaryotic cell.

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- The eukaryotic cells *(Greek; eu=good, karyotic=nucleated).*
- Eukaryotes, on the other hand, have distinct nuclei bound by a nuclear membrane and membrane-bound organelles *(mitochondria, chloroplasts, lysosomes, rough and smooth endoplasmic reticulum, vacuoles).*
- In addition, they possess organized chromosomes which store genetic material.

Difference between prokaryotes and eukaryotes:

Characteristic	Prokaryotes	Eukaryotes
Size of cell	Typically 0.2-2.0 m m in diameter	Typically 10-100 m m in diameter

Nucleus	No nuclear membrane or nucleoli (nucleoid)	True nucleus, consisting of nuclear membrane & nucleoli
Membrane-enclosed organelles	Absent	Present; examples include lysosomes, Golgi complex, endoplasmic reticulum, mitochondria & chloroplasts.
Flagella	Consist of two protein building blocks	Complex; consist of multiple microtubules
Glycocalyx	Present as a capsule or slime layer	Present in some cells that lack a cell wall
Cell wall	Usually present; chemically complex (typical bacterial cell wall includes peptidoglycan)	When present, chemically simple
Plasma membrane	No carbohydrates and generally lacks sterols	Sterols and carbohydrates that serve as receptors present
Cytoplasm	No cytosketeton or cytoplasmic streaming	Cytoskeleton; cytoplasmic streaming
Ribosomes	Smaller size (70S)	Larger size (80S); smaller size (70S) in organelles
Chromosome (DNA) arrangement	Single circular chromosome; lacks histones	Multiple linear chromosomes with histones
Cell division	Binary fission	Mitosis
Sexual reproduction	No meiosis; transfer of DNA fragments only (conjugation)	Involves Meiosis

Compartmentalization of Eukaryotes:

- In Eukaryotes, cells are arranged into compartments *(as it is bound on all the sides by a cell membrane).*

- It separates the protoplasm within the cell from the surrounding environment.
- Intracellular membrane systems, creats enclosed compartments that are separate from Cytosol.
- As a result, the cell is able to retain specific molecules and cartry out certain reactions in oderly manner.
- Prokaryotes evolved to form Eukaryotes, in the process *Cytosol compartmentalised* to form *Cytoplasm*.

(The cell cytoplasm contains *cytoplasm, cell organelles,* and *fluids - Cytosol*).

Plant cells

Plant cells are eukaryotic cells that differ in several key aspects from the cells of other eukaryotic organisms. Their distinctive features include the following organelles:

1. Vacuole:

- It is present at the centre and is water-filled volume enclosed by a membrane known as the tonoplast.
- The function is to maintain the cell's turgor, pressure by controlling movement of molecules between the cytosol and sap, stores useful material and digests waste proteins and organelles.

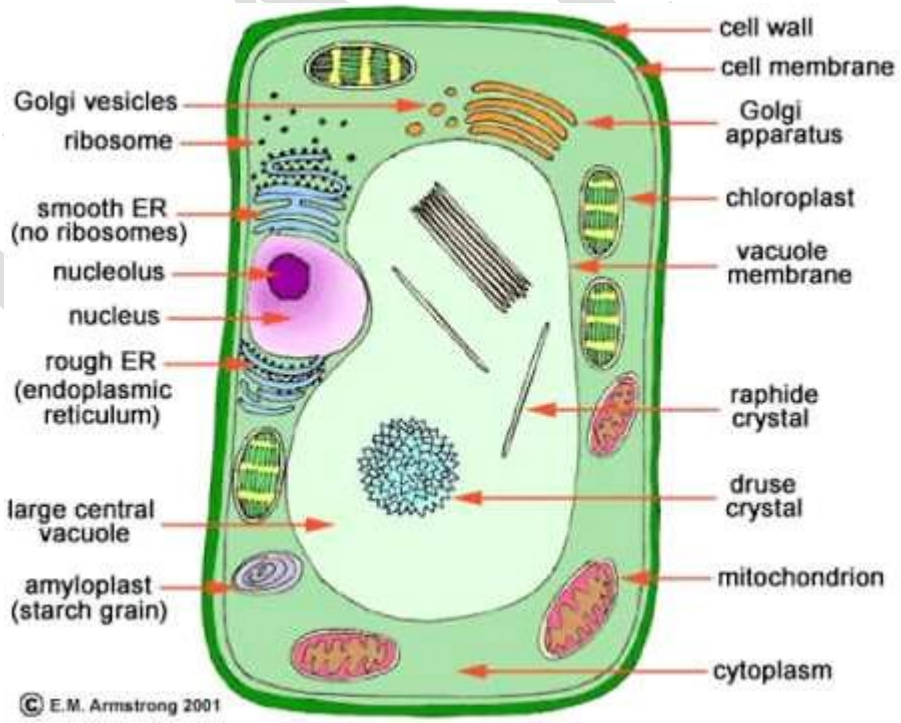


Fig:*Anatomy of Plant Cell.*

2. Cell Wall:

- It is the extracellular structure surrounding plasma membrane.
- The cell wall is composed of cellulose, hemicellulose, pectin and in many cases lignin, is secreted by the protoplast on the outside of the cell membrane.
- This contrasts with the cell walls of fungi (which are made of chitin), and of bacteria, which are made of peptidoglycan.

3. Plasmodesmata:

Pores in the primary cell wall through which the plasmalemma and endoplasmic reticulum of adjacent cells are continuous.

4. Plastids:

- The plastids are chloroplasts, which contain chlorophyll and the biochemical systems for light harvesting and photosynthesis.
- A typical plant cell (e.g., in the palisade layer of a leaf) might contain as many as 50 chloroplasts.

Plant cell types :

Parenchyma cells:

These are living cells that have diverse functions ranging from storage and support to photosynthesis and phloem loading (transfer cells).

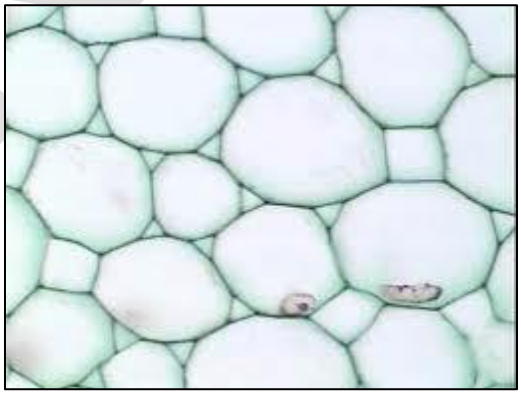


Fig:*Parenchyma cells which have thin primary cell wall.*

Collenchyma cells:

- Collenchyma cells are alive at maturity and have only a primary wall.
- These cells mature from meristem derivatives that initially resemble parenchyma, but differences quickly become apparent.

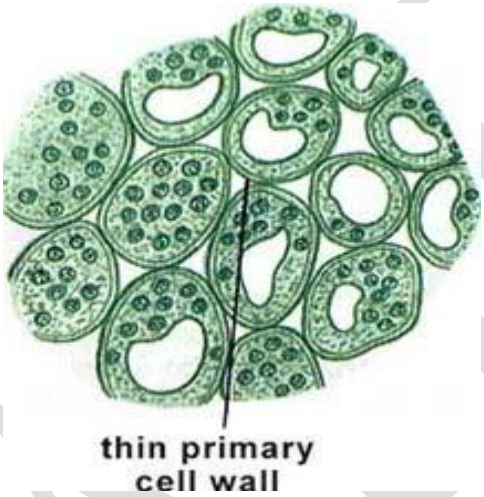


Fig:Typical collenchyma cell.

Sclerenchyma cells:

Sclerenchyma cells (from the Greek **skleros**, *hard*) are hard and tough cells with a function in mechanical support. They are of two broad types – sclereids or stone cells and fibres.

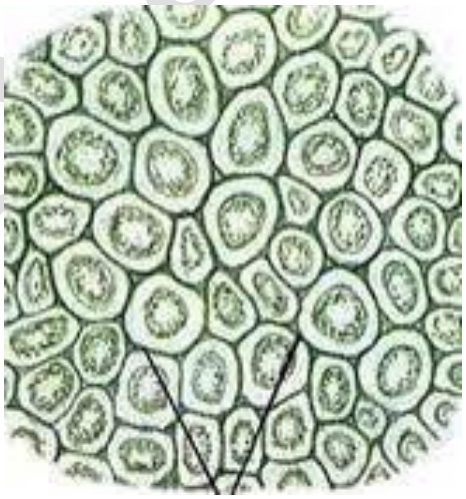


Fig:Sclerenchyma cells with irregularly thickened cell wall.

Animal cells:

- An animal cell is a form of eukaryotic cell that makes up many tissues in animals.
- The animal cell is different from plant cells, as they lack cell walls and chloroplasts, and they have smaller vacuoles.
- Due to the lack of a rigid cell wall, animal cells can adopt a variety of shapes, and a phagocytic cell can even engulf other structures.

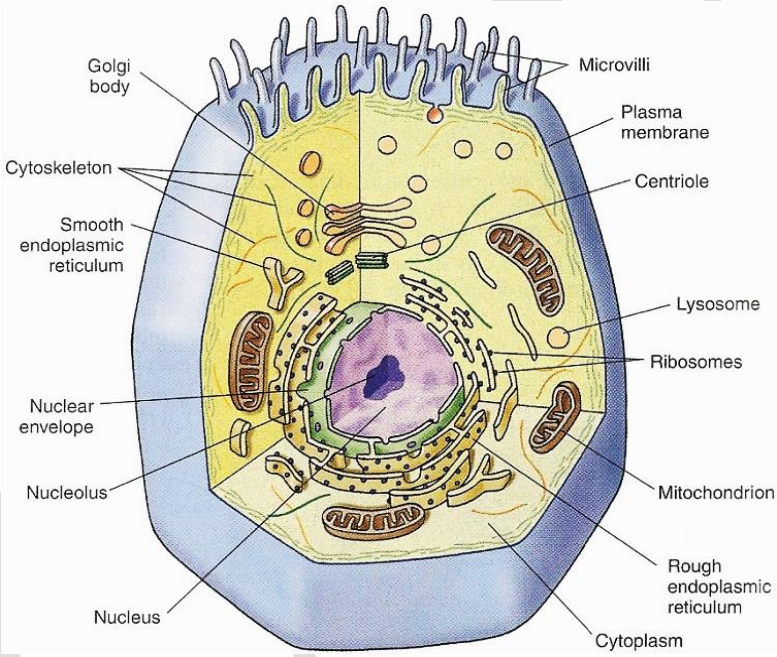


Fig:Schematic representation of a typical animal cell.

Cell organelles in animal cell:

1. Cell membrane:

- Plasma membrane is the thin layer of protein and fat that surrounds the cell (inside the cell wall – for plant cells).
- The cell membrane is semipermeable, allowing selective substances to pass into the cell and blocking others.

2. Nucleus:

- They are spherical body containing many organelles, including the nucleolus.
- The nucleus controls many of the functions of the cell (by controlling protein synthesis) and contains DNA (in chromosomes).
- The nucleus is surrounded by the nuclear membrane and possesses the nucleolus which is an organelle within the nucleus - it is where ribosomal RNA is produced.

3. Golgi apparatus:

It is a flattened, layered, sac-like organelle involved in packaging proteins and carbohydrates into membrane-bound vesicles for export from the cell.

4. Ribosome and Endoplasmic reticulum (ER) :

(Around the *Nucleus* there is ***densely granulated region*** – called ***Endoplasm***. ER is called so because it is present in the Endoplasm region **).**

Ribosomes are small organelles composed of RNA-rich cytoplasmic granules that are sites of protein synthesis and Endoplasmic reticulum are the sites of protein maturation and they can be divided into the following types:

a. Rough endoplasmic reticulum:

- These are a vast system of interconnected, membranous, infolded and convoluted sacks that are located in the cell's cytoplasm (the ER is continuous with the outer nuclear membrane).
- Rough ER is covered with ribosomes that give it a rough appearance.

b. Smooth endoplasmic reticulum:

- These are a vast system of interconnected, membranous, infolded and convoluted tubes that are located in the cell's cytoplasm (the ER is continuous with the outer nuclear membrane).
- The space within the ER is called the ER lumen. Smooth ER transport materials through the cell.

5. Mitochondria:

- These are spherical to rod-shaped organelles with a double membrane.
- The inner membrane is infolded many times, forming a series of projections (called cristae).

- The mitochondrion converts the energy stored in glucose into ATP (adenosine triphosphate) for the cell.

6. Lysosome:

- Lysosomes are cellular organelles that contain the hydrolase enzymes which breaks down waste materials and cellular debris.
- They can be described as the stomach of the cell.

- They are found in animal cells, while in yeast and plants the same roles are performed by lytic vacuoles.
- The membrane around a lysosome allows the digestive enzymes to work at the 4.5 pH they require.
- This means if enzymes escape from the lysosome they will be inactivated by the nuteral pH of the cell cytosol.
- Lysosomes digest excess or worn-out organelles, food particles, and engulf viruses or bacteria.

7. Centrosome:

- They are small body located near the nucleus and has a dense center and radiating tubules.
- The centrosomes are the destination where microtubules are made.
- During mitosis, the centrosome divides and the two parts move to opposite sides of the dividing cell. Unlike the centrosomes in animal cells, plant cell centrosomes do not have centrioles.

8.Peroxisome :

- Peroxisomes are organelles that contain oxidative enzymes, such as D-amino acid oxidase, ureate oxidase, and catalase.
- Peroxisomes function to rid the body of toxic substances like hydrogen peroxide, or other metabolites.

- They are a major site of oxygen utilization and are numerous in the liver where toxic byproducts accumulate.

9.Vacuoles and vesicles :

- Vacuoles are single-membrane organelles that are essentially part of the outside that is located within the cell.
- The single membrane is known in plant cells as a tonoplast.
- Many organisms will use vacuoles as storage areas.
- Vesicles are much smaller than vacuoles and function in transporting materials both within and to the outside of the cell.

Differences between Animal and Plant cell :

Sr. No.	Plant Cell	Animal Cell
01.	**Larger**	**Smaller** (Comparatively)
02.	**Cell wall is present** (made up of cellulose).	**Cell wall is absent.**
03.	**Plastid is present.**	**Absent.**
04.	**Large Vacuole** (occupies 90% of cell space).	**Vacuole is absent or *very small*.**
05.	**Absent**	**Lysosomes are present.**
06.	**Absent**	**Centriols are present.**
07.	**Plasmodesmata** (cytoplasmic strands connecting protoplast of adjacent cells through cell walls) **is present.**	**Absent** (tight junction between cells).
08.	**Absent**	**Desmosome is present.**
09.	**Ability to synthesis *amino acid, coenzymes, and vitamins* required by them.**	**Can not synthesis *amino acid, coenzymes, and vitamins* required by them.**

10.	**Nucleus is near cell wall** (because vacuole occupies large space)**.**	**Nucleus lies in the center.**
11.	**Glyoscysomes present.**	**Absent**
12.	**Food stored in the form of *Starch*.**	**Food stored in the form of *Glycogen.*** *(It is animal starch- which is profusely branched)*
13.	**Spindel fibres are *Anastral.***	**Spindel fibres are *Astral.***
14.	**Cytokinesis occurs in *Plate method.***	**Cytokinesis occurs in *Burrowing method.***
15.	**Plant cells do not burst in *Hypotonic solution.*** (It is due to the presence of ***Cell wall***.)	**Animal cells do not burst in *Hypotonic solution.*** (It is due to the absence of ***Cell wall***.)

Cell Fractionation: Extraction, Homogenization and Centrifugation

Cell fractionation:

Cell fractionation is a procedure for rupturing cells, separation and suspension of cell constituents in isotonic medium in order to study their structure, chemical composition and function.

Cell fractionation involves 3 steps: Extraction, Homogenization and Centrifugation.

1. Extraction:

It is the first step toward isolating any sub-cellular structures. In order to maintain the biological activity of organelles and bio-molecules, they must be extracted in mild conditions called cell-free systems. For these, the cells or tissues are suspended in a solution of appropriate pH and salt content, usually isotonic sucrose (0.25 mol/L) at0-40°C.

2. Homogenization:

The suspended cells are then disrupted by the process of homogenization.

It is usually done by:

(i) Grinding

(ii) High Pressure (French Press or Nitrogen Bomb),

(iii) Osmotic shock,

(iv) Sonication (ultrasonic vibrations). Grinding is done by pestle and mortar or potter homogenizer (a high-speed blender). The later consists of two cylinders separated by a narrow gap.

The shearing force produced by the movement of cylinders causes the rupture of ceils. Ultrasonic waves are produced by piezoelectric crystal. They are transmitted to a steel rod placed in the suspension containing cells. Ultrasonic waves produce vibrations which rupture the cells. The liquid containing suspension of cell organelles and ether constituents is called homogenate. Sugar or sucrose solution preserves the cell organelles and prevents their clumping.

3. Centrifugation:

The separation (fractionation) of various components of the homogenate is carried out by a series of cemrifugations in an instrument called preparative ultracentrifuge. The ultracentrifuge has a metal rotor containing cylindrical holes to accommodate centrifuge tubes and a motor that spin the rotor at high speed to generate centrifugal forces. Theodor Svedberg (1926) first developed die ultracentrifuge which he used to estimate the molecular weight of hemoglobin.

Present day ultracentrifuge rotate at speeds up to 80,000 rpm (rpm= rotations per minute) and generates a gravitational pull of about 500,000 g, so that even small molecules like t-RNA, enzymes can sediment and separate from other components.

The chamber of ultracentrifuge is kept in a high vacuum to reduce friction, prevent heating and maintain the sample at 0-4°C.

During centrifugation, the rate at which each component settle down depends on its size and shape and described in terms of sedimentation coefficient or Svedberg unit or S-value, where IS = 1 x 10^{-13} second.

The standard cell fractionation technique involves following methods:

(a) Differential velocity centrifugation [Velocity sedimentation or Rate zonal centrifugation):

It is the first step of cell fractionation by which various sub-cellular organelles are separated based on differences in their size. The homogenate in first filtered to remove unbroken cell clumps and collected in a centrifuge tube. The filtered homogenate when centrifuged in a series of steps at successively greater speeds, each step yields a pellet and a supernatant

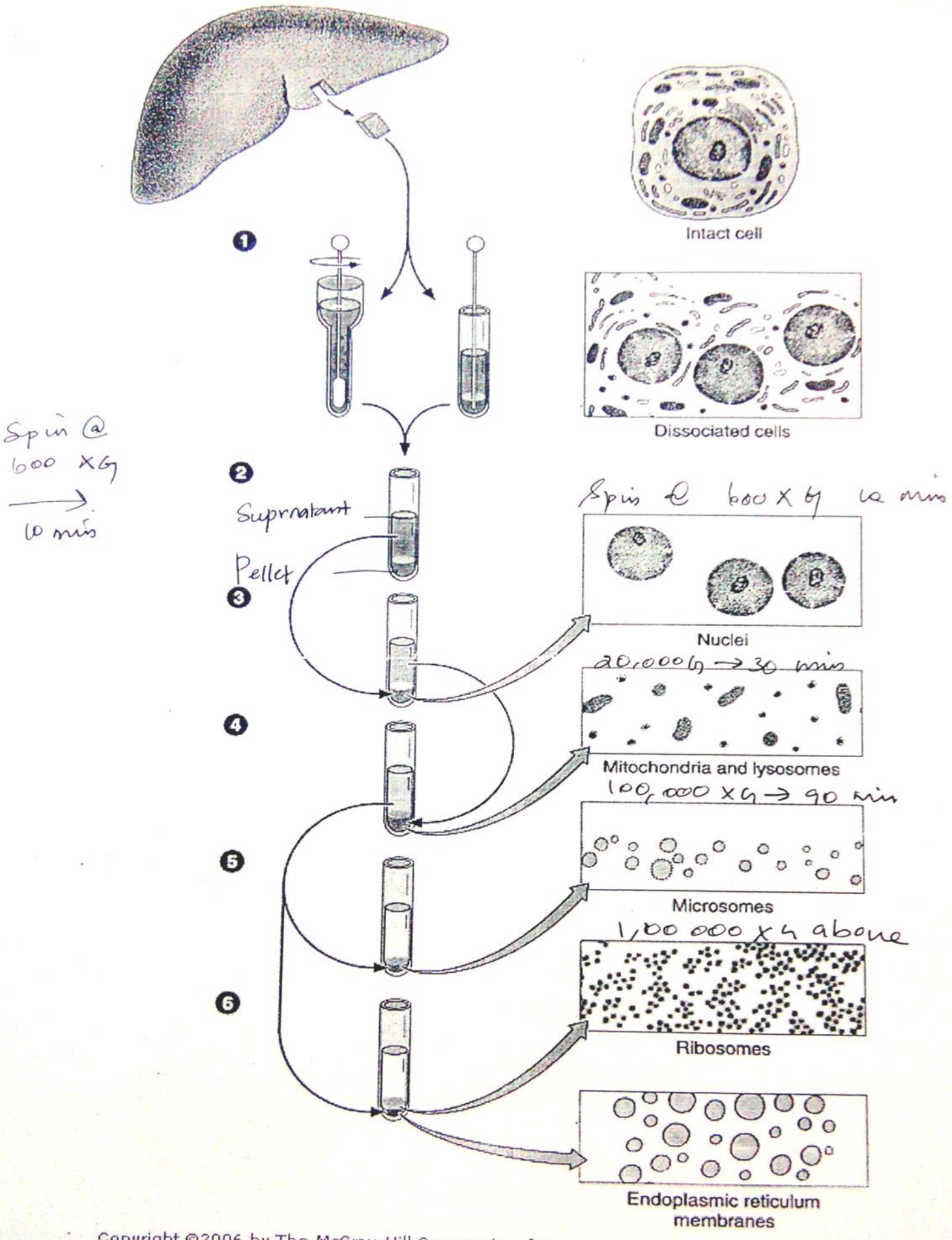
The supernant of each step is removed to a fresh tube for centrifugation. For instance, at low speed (600g. for: 10 min) nuclear fraction or pellet will sediment at medium speed (15,000g x 5 min) mitochondria fraction sediment and at high speed (80,000 g. x 5 min.) micro-somal fraction sediment. The final supernant is soluble fraction or cytosol.

(b) Equilibrium Density-gradient centrifugation (Equilibrium sedimentation):

The organelle fractions (pallets) obtained in velocity centrifugation is purified by equilibrium density-gradient centrifugation. In this method organelles are separated by their density not by their size.

The impure organelle fraction is layered on the top of a gradient solution, e.g., sucrose solution or glycerol solution. The solution is more concentrated (dense) at the bottom of the centrifuge tube, and decreases in concentration gradually towards the top. The tube when centrifuged at high speed the various organelles migrate to an equilibrium position where their density is equal to the density of the medium. Meselson, Stahl and Vinograd (1957) used denser cesium chloride gradient for separation of a heavy DNA with ^{15}N from DNA with ^{14}N to provide evidence for semi-conservative DNA replication.

In conclusion, we may say that what one can learn about cells, depends on the tools at one's disposed and, in fact, major advances in cell biology have frequently taken place with the introduction of new too is and techniques to the study of cell. Thus, to gain different types of information regarding cell, cell biologists have developed and employed various instruments and techniques. A basic knowledge of some of these methods is earnestly required.



Cell Membrane and Transport Mechanisms Across The Cell Membrane:

Cell membrane:

- Cell membrane or plasma membrane mainly regulates the cellular entry and exit of molecules and ions.
- This function of cell membrane is called cell permeability.
- Plant cells have a thick cell wall that covers plasma membrane and protects it.
- Animal cells have a cell coat or external laminae.
- Isolation of membranes from erythrocytes is relatively easy.
- Hypotonic solutions cause swelling of membranes forcing out hemoglobin from red blood cells forming a red cell ghost and hemolysis.

Components of cell membrane :

- The membrane is made up of a lipid bilayer embedded with proteins some of them protruding out from the membrane.
- Red blood cell membrane has 52% protein, 40% lipids and 8% carbohydrates.
- There is a wide variation in this composition among various membranes.
- Phospholipids, cholesterol and galactolipids generally constitute the major lipid portion of membranes but their composition varies among different membranes.
- Major phospholipids include the neutral phospholipids (no net charge at neutral pH) such as phosphatidyl choline, phophatidyl ethanolamine and sphingomyelin.
- Acidic phospholipids (5-20%) are negatively charged that include phosphatidyl inositol, phosphatidyl serine, phosphatidyl glycerol, cardiolipin and sulfolipids.

Membrane proteins

- Proteins are not only structural components but also serve as carriers or channels.
- They also serve as receptors for various signaling ligands in addition being enzymes and antigens.
- Membrane proteins are generally classified into extrinsic (peripheral) or intrinsic (integral) proteins.
- Peripheral proteins are soluble in water/aqueous solutions and do not contain much of lipids. Eg: Spectrin, cytochrome c.
- The majority of membrane proteins are integral proteins that are insoluble in water.
- They are in strong association with lipids and carbohydrates. Membrane bound enzymes, histocompatibility antigens and various drug and hormone receptors belong to this category.
- Hydrophilic or polar /hydrophilic amino acids are mainly present near the surface in the peripheral proteins whereas the nonpolar/hydrophobic amino acids are buried inside
- In the case of integral proteins the nonpolar amino acids are more exposed to the surface.

- Outer surface of some membranes contain acetyl choline esterase, nicotinamide dinunucleotide-adenine dinucleotidase and the ouabain binding site of Na+ K+ ATPase.
- Inner surface contains NADH-diaphorase, adenylatecyclase, protein kinase and Mg++ ATPase.

Table :*Composition of different membrane lipids.*

Type	Composition	Example/ Remarks
Phosphoglycerides	esters of phosphoric acid and a trifunctional alcohol-glycerol	Phosphatidate four common substituents for phosphatidate; Serine, ethanolamine, choline and inositol.
Sphingolipids	Phosphoglycerides where glycerol is substituted with sphingosine.	Sphingomyelin, Glycosphingolipid Found in particularly nerve cells and brain tissues

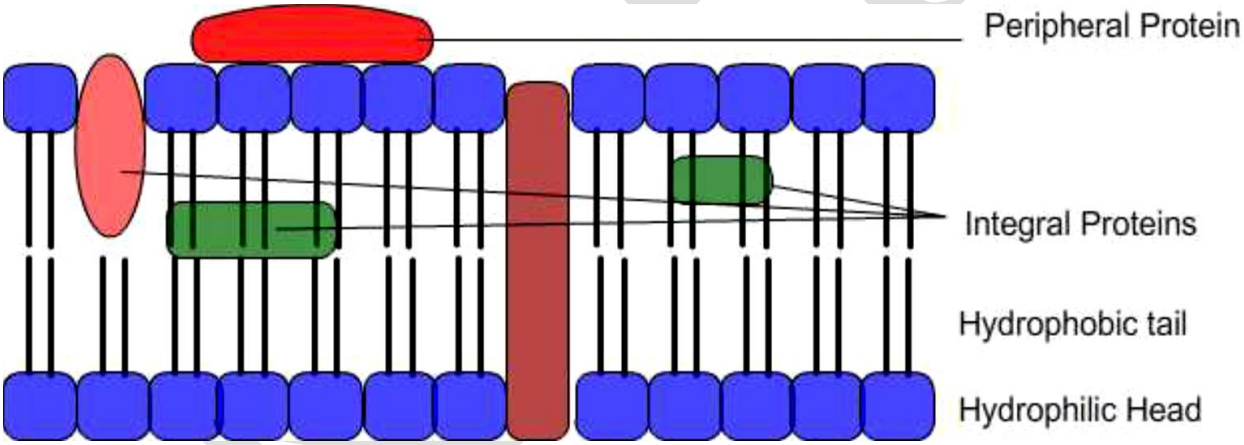
Table :*Cellular Architecture and function of Cell Membrane.*

Organism	Lipid composition	Membrane properties	Functionalities
Theories on Membrane Structures			
Bacteria	Phosphatidylethanolamine and Phosphatidylglycerol	Robust Different shapes	Membrane protein incorporation
Yeast	Sphingolipids, Glycerophospholipids and Sterols	Robust Different shapes Complex organelle morphology	Membrane protein incorporation Membrane budding Vesicular trafficking
Higher Eukaryotes	Glycerophospholipids, sterols, and tissue-specific Sphingolipids	Robust Different shapes Complex organelle morphology Complex and specific cellular architecture	Membrane protein incorporation Membrane budding Vesicular trafficking Specific functions depending on the cell type

- In 1902 it was thought that the membranes had only lipids (Overton).
- In 1926 *Gorter and Grendell* proposed that lipids are capable of forming a double layer.
- In 1935 *Danielli* and *Davson* proposed the lipid bilayer model that includes proteins adhering to both lipid-aqueous interfaces
- Artificial model systems such as the liposomes supported the idea of *Danielli* and *Devson.*
- A droplet of lipid made soluble in an organic solvent can be spread over a small hole on a septum that divides two chambers containing water.
- This set up is useful to study biophysical properties of a bilayer such as permeability and electrical resistance.
- Channels for ions can be formed by adding certain proteins or polypeptides.
- Liposomes act as excellent carriers for different molecules such as chemotherapeutic compounds, insulin and antibodies.

Fluid mosaic model

- Fluid mosaic model proposed by S.J. Singer and G.L. Nicolson (1972) was finally acceptable to most biologists
- This model recognizes that lipids and proteins are in a mosaic arrangement.



- It also recognizes that there is translational movement of lipids and proteins within the lipid bilayer.
- Non covalent interactions ensure a fluid like state for the membranes.
- Integral proteins are intercalated into the continuous lipid bilayer.
- Polar/hydrophilic regions of proteins protrude from the surface while the nonpolar/hydrophobic regions are embedded inside.
- The concept of fluidity is attractive as it explains the considerable freedom of lateral movement for proteins and lipids observed within the bilayer.

Fluid mosaic model Permeability

- Permeability is an important property of the plasma membrane and other membranes in a living cell.
- This is important for maintaining the required intracellular conditions.
- Basically this determines as to what substances should enter or leave the cell and in turn this is essential to maintain life.
- The composition of important body fluids depends on the permeability.
- Osmotic pressure of intra and extra cellular fluids depends upon the permeability.

Electrical and ionic gradients

- Between the extracellular and intracellular compartments ionic and electrical gradients exist.
- Their interdependence is known since the distribution of ions on both sides of the membrane contributes to the electrical potential.
- Intracellular fluid contains more of K+ ions and organic anions
- Interstitial fluid contains more of Na+ and Cl- ions.
- The resting/steady potential is usually negative inside a cell and varies between -20 and -100 mV.
- The diffusion of ions depends both on the concentration and electrical gradients across the membrane.

Passive permeability:

- Membrane acts as a barrier to the passage of water soluble molecules.
- Lipid soluble substances more easily pass through the membrane.
- Size and solubility of molecules are important factors affecting their permeability.
- P=KD/t where P is permeability, K is partition coefficient, D is diffusion coefficient, and t is thickness of membrane.
- If two molecules have the same size, the one with higher solubility in lipids will penetrate the membrane faster.
- If two molecules have equal solubility in lipids, the smaller molecule will penetrate the membrane faster.

Membrane Transport

Types of passive transport:

1. Diffusion:

- The process of the net movement of solutes from a region of high concentration to a region of low concentration is known as diffusion.
- The differences of concentration between the two regions are termed as concentration gradient and the diffusion continues till the gradient has been vanished.
- Diffusion occurs down the concentration gradient.

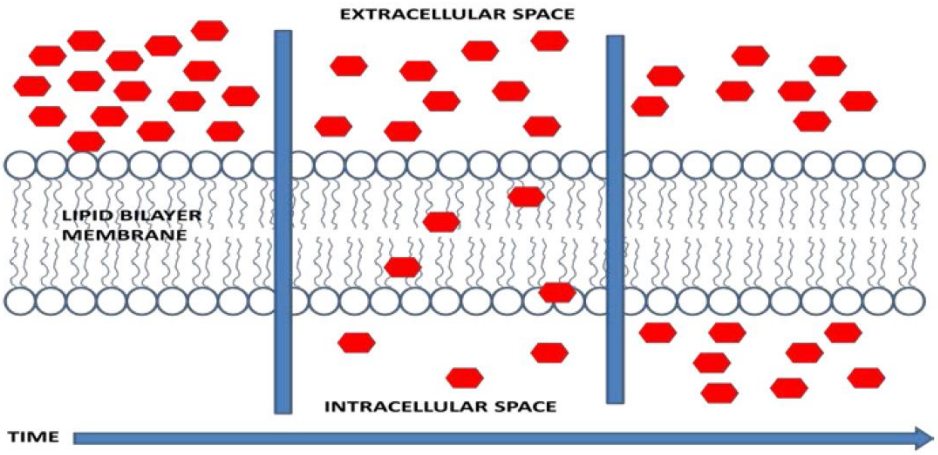


Figure 3: Diffusion.Extracellular space contains high concentration of solutes than intracellular space and hence the solutes move from extracellular space to intracellular space till there is no concentration gradient between the spaces.

2. Osmosis:

- Osmosis is the type of diffusion of water molecules across a semi- permeable membrane, from a solution of high water potential to a region of low water potential.
- A cell with a less negative water potential will draw in water but this depends on other factors as well such as solute potential (pressure in the cell e.g. solute molecules) and pressure potential (external pressure e.g. cell wall).

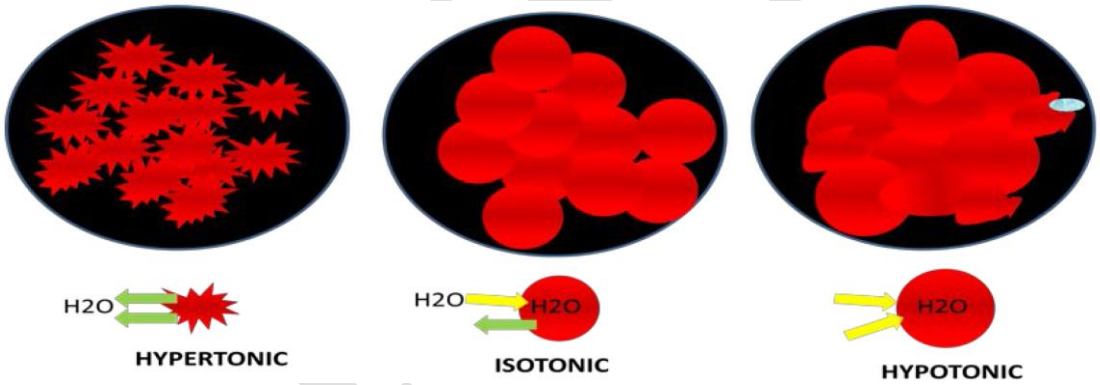


Fig:Osmosis.(A) In hypertonic solution, there are more solute molecules outside the cell, which causes the water to be sucked in that direction which leads to the shrinkage of cells. **(B)** In isotonic solution, there is equal concentration of solute on both sides, henceforth the water with move back in forth. **(C)** In hypotonic solution, there are less solute molecules outside the cell, since salt sucks and water will move inside the cell. The cell will gain water and grow larger, and finally burst.

Active Transport:

- If only passive diffusion operates in cells any increase or decrease in membrane potential would result in asymmetric ionic distribution.
- Many experiments have shown that indeed there are active transport mechanisms in living cells that require energy.
- ATP provides energy for such active transport processes.
- Thus oxygen consumption is required when an ion is transported against the electrochemical gradient.
- Active transport is also needed to maintain the resting potential
- The cells are able to keep a constant osmotic pressure by regulating the ionic transport across the cell membrane.
- Potassium ions are present in higher concentrations inside the cells through a pumping mechanism that requires energy to work against the concentration gradient.

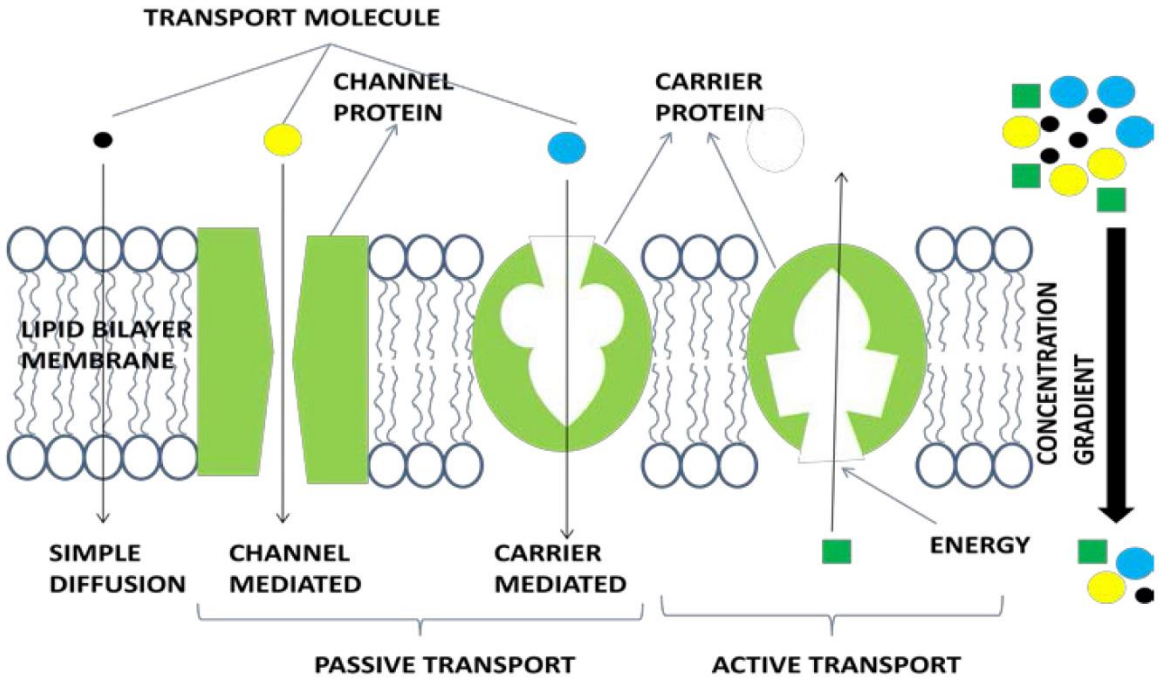


Figure 2: Mediated transport. (A) Passive transport and (B) Active transport

Sodium Pump

- Although Na+ does not have a higher mol. wt. in comparison to K+ and Cl- ions its ionic radius in the hydrated conditions is higher and thus it cannot enter the cell easily.
- Sodium pump throws out Na+ ions together water from inside to the outside of the cell by an active transport mechanism.
- Na+ K+ ATPase is an enzyme that couples the hydrolysis of ATP with the elimination of Na+ ions from the cytoplasm against electrochemical gradient.

- Hydrolysis of one ATP can give energy to transport two K+ ions towards inside and three Na+ ions towards outside.
- Thus both these ions can activate ATPase and ouabain can inhibit it by binding to it on the extracellular surface of the enzyme.
- Vanadate also inhibits this enzyme but this acts from the cytoplasmic side of the enzyme.
- The first step for this enzyme reaction is the formation of a covalent phosphoenzyme intermediate.
- This happens on the inner side of the membrane in the presence of Na+ ions but Ca++ ions inhibit this reaction
- In the second step the intermediate complex is hydrolyzed forming the free enzyme and phosphate ions and this requires K+ but is inhibited by ouabain.
- Other substances such as glucose and amino acids may use the sodium pump for their transport.

Sodium PumpTransport proteins

- Selective transport of molecules across the membranes is also achieved by means of carriers/permeases/transport proteins.
- High degree of specificity by this mechanism is related to the chemical structures of molecules being transported.
- For example the structures of glucose and galactose are very similar except for the position of OH group at carbon 4, but these two molecules cross the membranes by using different carriers.
- Permeases help in achieving this specificity related to the structure and in the process the permeases do not change and get recycled for another round.
- Some permeases work under a favorable concentration gradient in a mode of passive diffusion called facilitated diffusion.
- Some others work against the concentration gradient employing an active transport mechanism.

Mechanisms

- The **carrier mechanism** works by first binding of the molecule with the carrier protein at the outer surface of the cell.
- Then this complex translocates into the cytoplasm by a rotatory movement.
- However, this kind of a mechanism is not thermodynamically favorable as rotation and translocation across the bilayer may not be easy.
- **Fixed pore mechanism** suggests that the carriers are actually integral proteins and they undergo conformational change once a molecule to be transported gets attached with the carrier.
- In the above mechanism, the carrier proteins are suggested to be oligomers forming a channel or pore that has a hydrophilic lining in the middle.
- This mechanism can account for the sodium pump action and also for the transport of glucose and amino acids.

Cell Recognition

Also called-

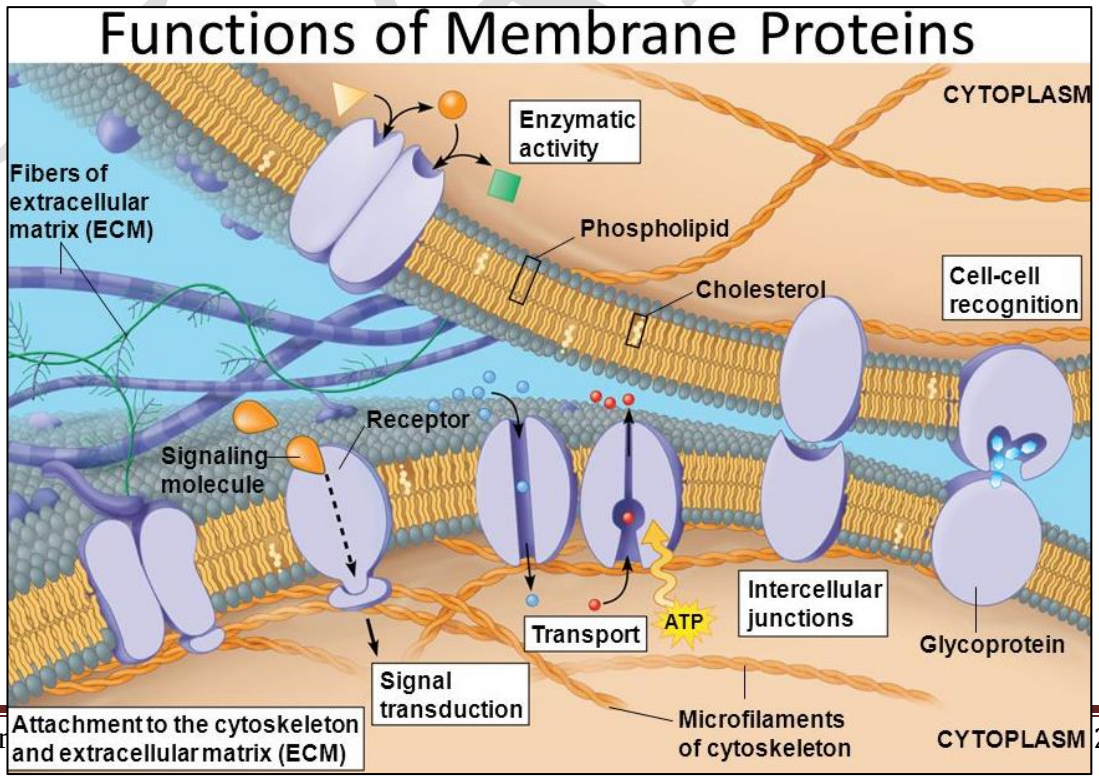
- *Cellular recognition*
- *cell-cell recognition*
- *cell to cell recognition*
- It is the mutual recogniton between ***cells***, usually by specific complementary interaction between their respective surface molecules or membrane ***glycoproteins***.
- The process in which cell recognises or identifies its environment.
- It is possible through specific cellular ***adhesion molecules*** on the surface of the ***cell***.

Cell Adhesion,*the binding of a* ***cell*** *to another* ***cell*** *or to an* ***extrcellular matrix component,*** *essential in organ formation during* ***embryonic*** *development and in confering structural framework and* ***tissue*** *maintenance.*

Cell adhesion may be through :

- ***Homophilic adhesion*** *– where cell adhesion is faciliated by binding of similar adhesion molecules.*
- ***Heterophilic sdhesion*** *– where cell adhesion is faciliated by binding of unlike adhesion molecules.*

<u>Cell recognition and Adhesion involve proteins at the cell surface.</u>



- ***Complex carbohydrates***coat the surfaces of the cells and have the potential to carry the information necessary for ***cell-cell recognition***.

- ***Sugar-specific receptors***(***lectins***) are also present on the cells, and can interact with ***sugars on***apposing cells. This may result in the adhesion of the two cells via ***carbohydrates*** and ***specific cell-receptors.***

- Such ***carbohydrate-directed*** cell adhesion appears to be important in many ***inter cellular activities*** including *infection by bacteria and viruses, communication among cells of lower eukaryotes, specific binding of sperm to egg; and re-circulation oflymphocytes*, among others.

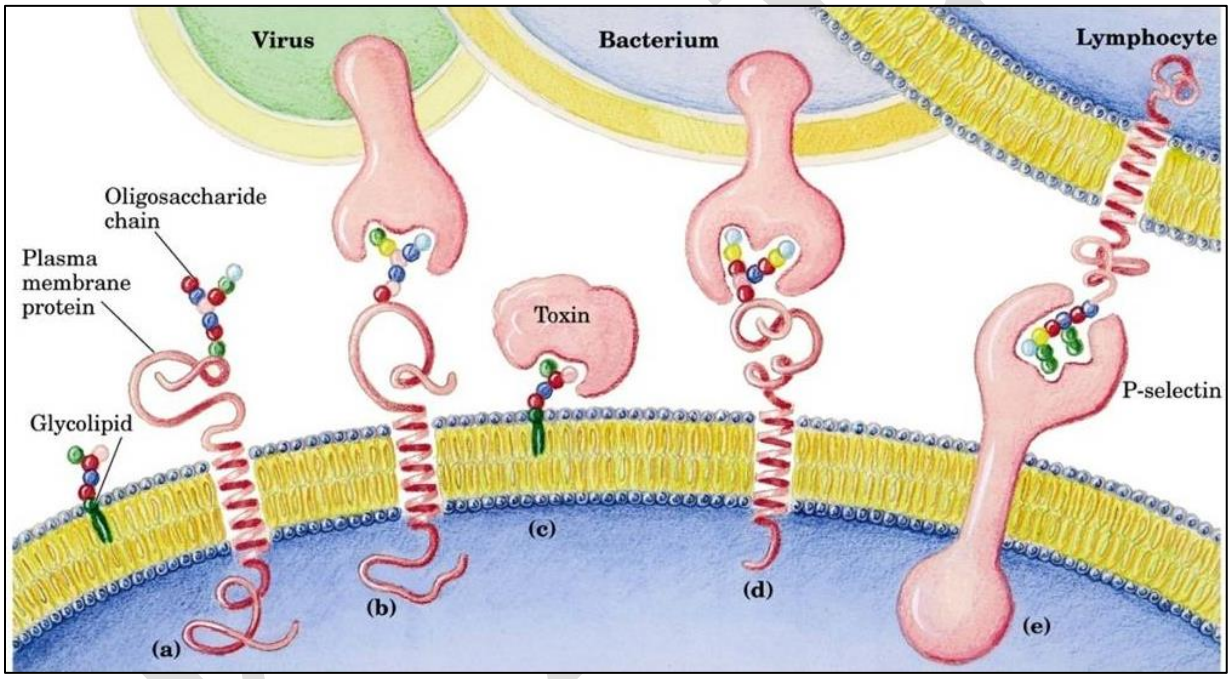


Fig :*(a) Oligo-saccharide chain linked with Plasma membrane; Bond with (b) Virus, (c) Toxin, (d) Bacterium, (e) Lymphocyte.*

Possible Questions

2 Marks

1. Comment on the cell theory.
2. Explain about the cell.
3. Define a) adherin b) cytosol.
4. Write about Cell recognition.
5. Define passive transport.
6. Define active transport.

8 Marks

1. Write about the cell compartmentalization.
2. Explain in detail Cell fractionation.
3. Write about the thrust Cell membrane and permeability.
4. Explain the fluid mosaic model.
5. Briefly explain about Cell membrane transport.
6. Draw the structure of eukaryotic cells.
7. Distinguish prokaryotic and eukaryotic cells.

Unit: II

Extracellular Matrix: Composition, molecules that mediate cell adhesion, membrane receptors for extracellular matrix, macromolecules, regulation of receptor expression and function. Signal transduction.

Extracellular matrix

- Animal cells are surrounded by extracellular matrix beyond the immediate vicinity of their plasma membrane, filling spaces between cells and adhering cells together.
- Extracellular matrices are of various types consisting of secreted proteins and polysaccharides and are most abundant in connective tissues.
- One of the examples of extracellular matrix is the basal laminae. It is a continuous sheet of 50 to 200 nm thickness and on top of which a thin layer of epithelial cells rest.
- Such basal laminae surround muscle cells, adipose cells, and peripheral nerves.
- The differences between various types of extracellular matrices result from both quantitative variations in the types or amounts of these different constituents and from modifications in their organization.
- The three major components of extracellular matrix are matrix proteins, matrix polysaccharides and the matrix adhesion proteins.

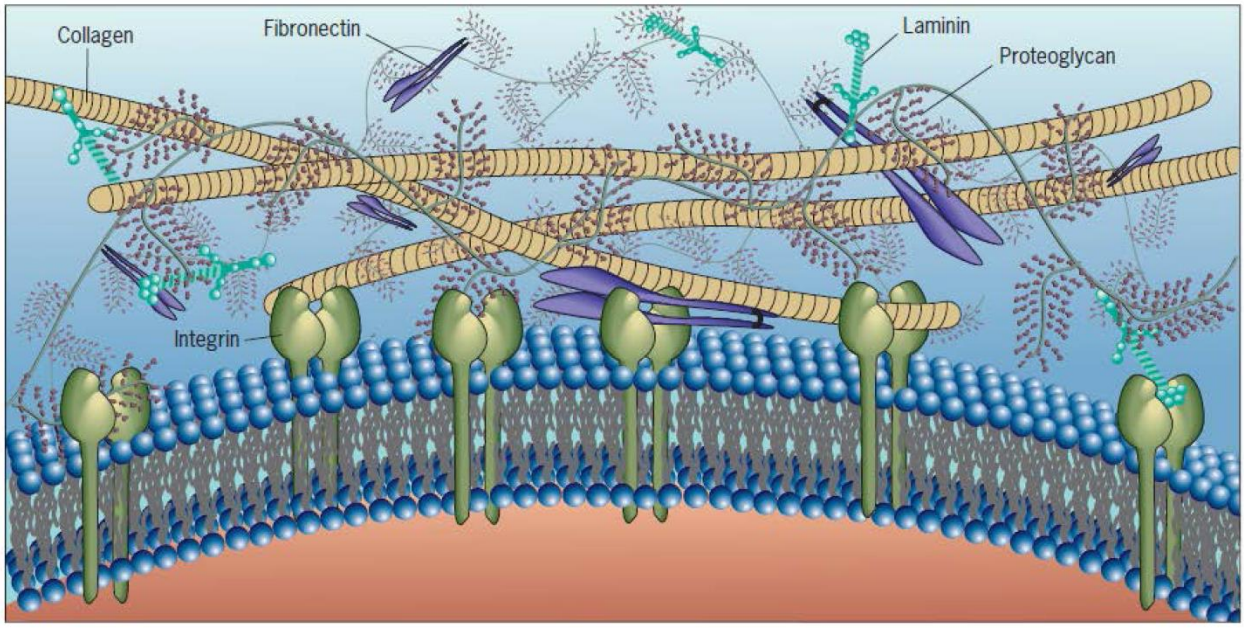


Fig : An overview of the extracellular matrix molecular organization. The proteins; fibronectin, collagen, and laminin contain binding sites for one another, as well as binding sites for receptors like integrins that are located at the cell surface. The proteoglycans are huge protein polysaccharide complexes that occupy much of the volume of the extracellular space. This figure has been adapted from Cell and Molecular Biology Concepts and Experiments by Karp, 2010.

Structural proteins of Matrix

- Matrix proteins are fibrous in nature.
- The major structural protein is collagen whose secondary structure is a triple helix.
- The collagens belong to large family of proteins and are characterized by the formation of triple helices in which three polypeptide chains are wound tightly around one another in a ropelike manner.
- The different collagen polypeptides can assemble into 42 different trimers.
- The triple helix domains of the collagens consist of repeats of the amino acid sequence Gly-X-Y.

- The most abundant type is collagen type I and is one of the fibril forming collagens that are the basic structural components of connective tissues**.**
- Elastin is another matrix protein, which gives elasticity to tissues, allowing them to stretch when needed and then return to their original state.
- They are present in blood vessels, the lungs, in skin, and the ligaments. Elastins are synthesized by fibroblasts and smooth muscle cells.

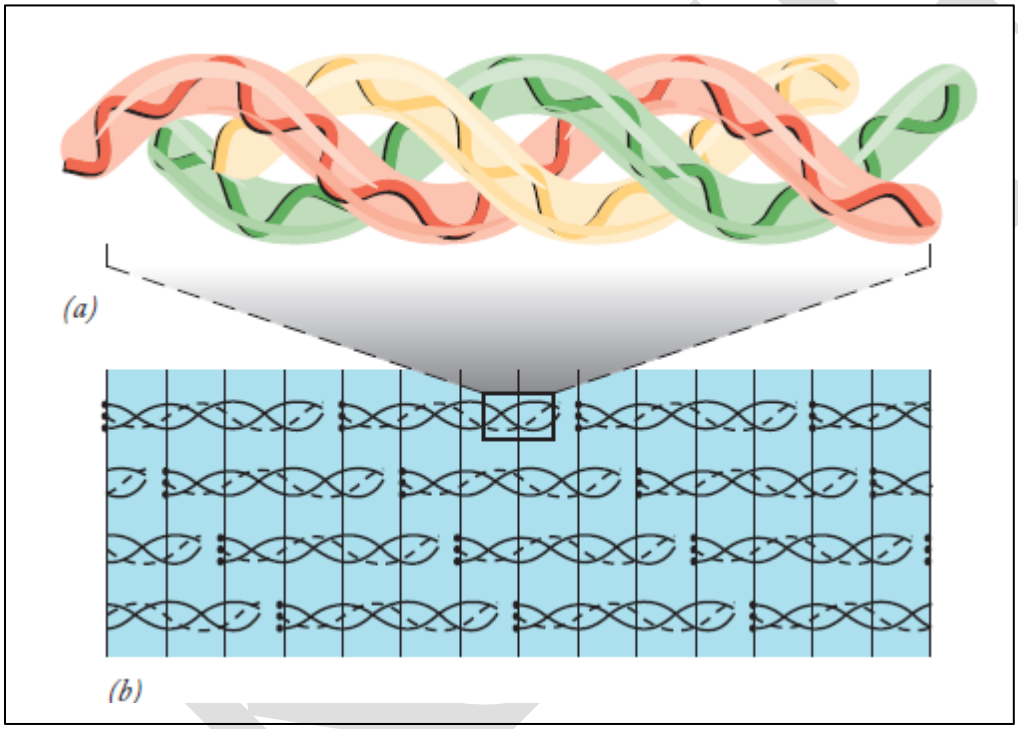


Fig :*The structure of collagen I. (a) The monomer of collagen. (b) Collagen I molecules become aligned in and a bundle of collagen I molecules, such as that shown here, form a collagen fibril.*

Polysaccharides of Matrix

- The structural proteins of the extracellular matrix are rooted in polysaccharides called glycosaminoglycans (GAGs).

- One sugar of the disaccharide is either N-acetylglucosamine or N-acetylgalactosamine and the second is usually glucuronic acid or iduronic acid.
- They can also be sulfated like the chondroitin sulfate, dermatan sulfate, heparan sulfate, and keratan sulfate.
- These polysaccharides are highly negative in charge and bind positively charged ions and water molecules to form hydrated gels.
- The function of such gels is to provide support to the matrix. Hyaluronan is the only GAG that occurs as a single long polysaccharide chain.
- GAGs also attach with proteins through Serine residues and are known as proteoglycans.
- A number of proteoglycans interact with hyaluronan to form large complexes in the extracellular matrix e.g., aggrecan which is the major protein of the cartilage.
- Proteoglycans also interact with collagen and other matrix proteins to form gel-like networks in which the fibrous structural proteins of the extracellular matrix remain rooted.

Adhesion proteins of Matrix

- Matrix adhesion proteins are accountable for connecting the components of the matrix to one another and to the surfaces of cells.
- They act together with collagen and proteoglycans to direct matrix organization and bind to integrins.
- The first of its kind is fibronectin, which is the main adhesion protein of connective tissues.

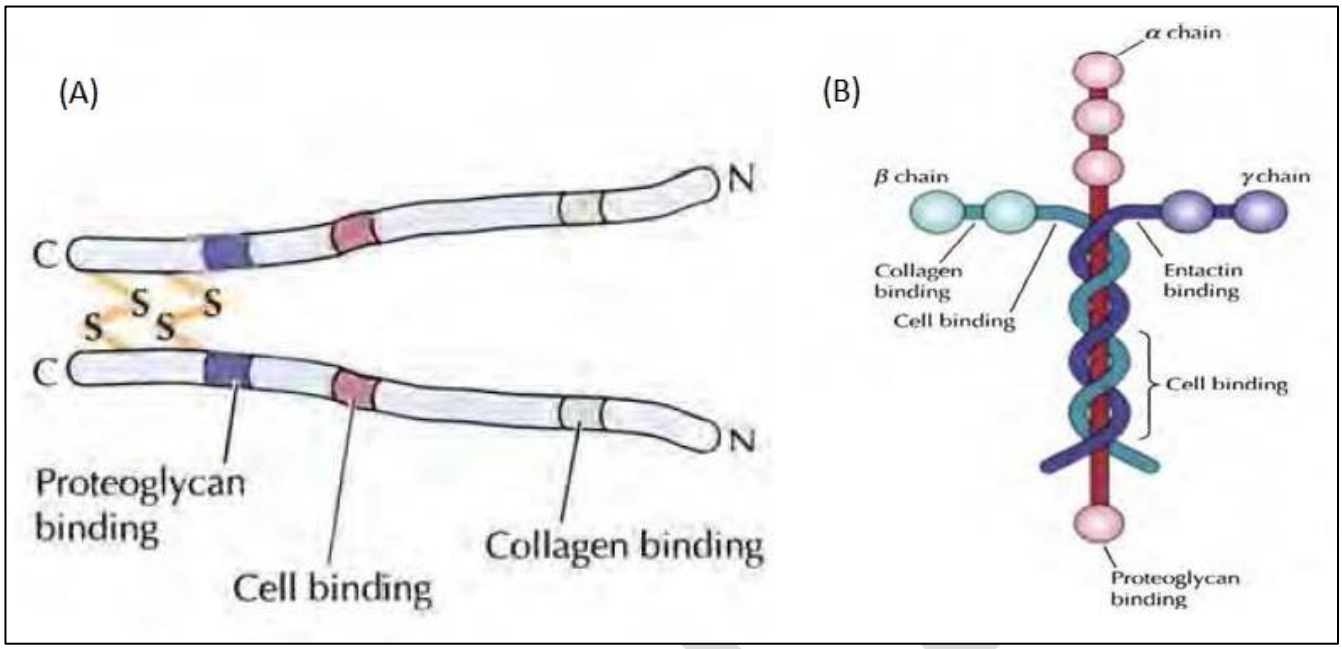


Fig :*An illustration of matrix associated proteins. (A). Fibronectin. (B). Laminin.*

- Fibronectin is a glycoprotein with two polypeptide chains, of 2500 amino acids.
- Additionally fibronectin possess binding sites for both collagen and GAGs thus crosslinking these matrix.
- A specific site on the fibronectin molecule is responsible for recognizing cell surface receptors like integrins attaching of cells to the extracellular matrix.
- Prototype of adhesion proteins belong to the laminin family with the property of self assembly into mesh like networks.

Cell matrix interaction

- Cells remain attached to the extracellular matrix through the aid of cell surface receptors such as integrins.
- The integrins belong to the family of transmembrane proteins consisting of one α and one β subunits.

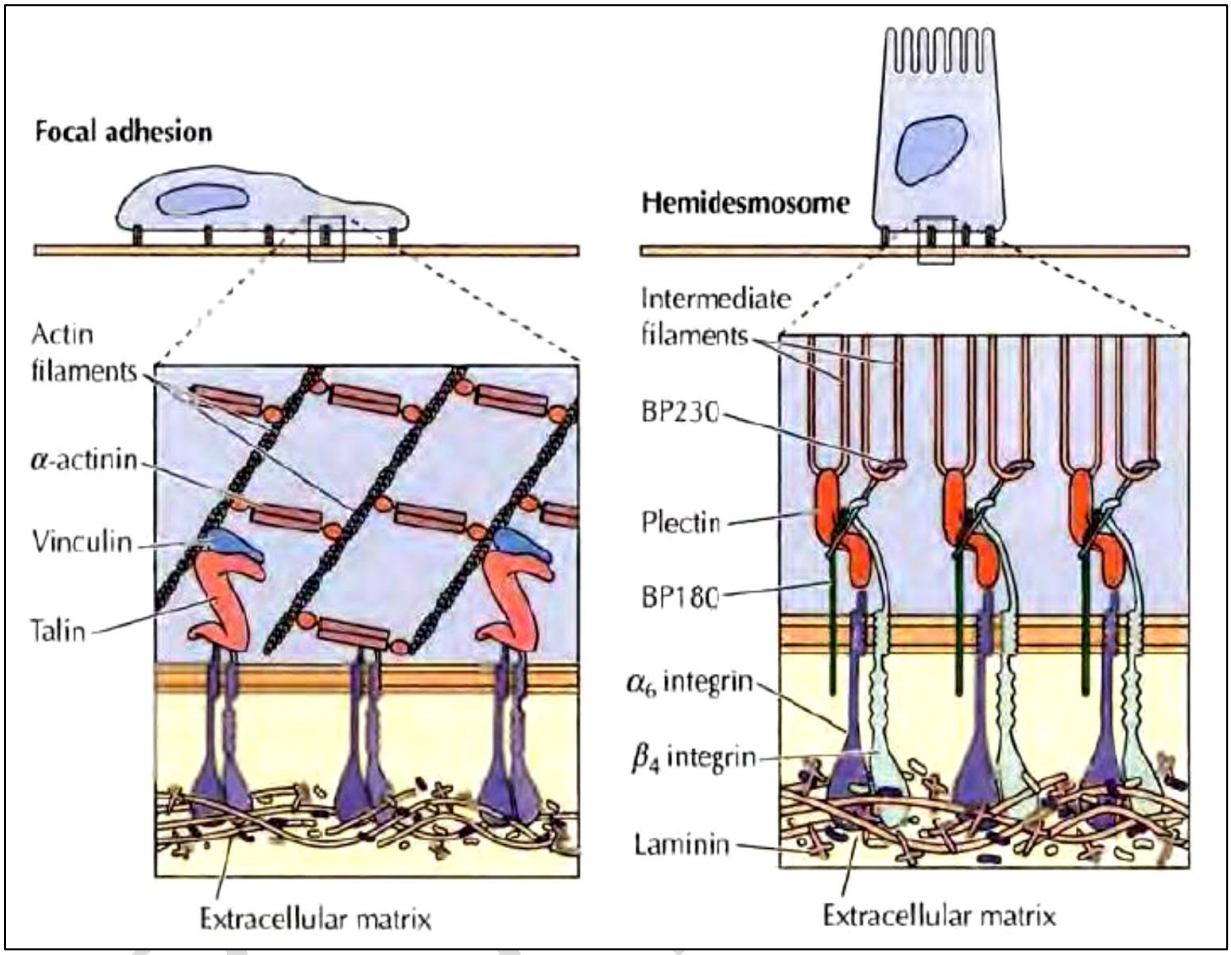


Fig:*Cell-matrix junctions mediated by integrins. Integrins mediate two types of stable junctions the focal adhesions where bundles of actin filaments are anchored to integrins through associations with a number of other proteins, including α-actinin, talin, and vinculin. In hemidesmosomes, integrin links the basal lamina to intermediate filaments via plectin and BP230. BP180 functions in hemidesmosome assembly and stability.*

- The integrins bind to short amino acid sequences present in multiple components of the extracellular matrix, including collagen, fibronectin, and laminin.
- In addition to attaching cells to the extracellular matrix the integrins also provide anchors for the cytoskeleton resulting in stability of the cell matrix junctions.

- Integrins interact with the cytoskeleton at two junctions of the extracellular matrix known as the focal adhesions and hemidesmosomes.
- Focal adhesions attach a variety of cells, including fibroblasts, to the extracellular matrix and hemidesmosomes mediate epithelial cell attachments at with a specific integrin.
- Cell-matrix interaction is a step wise process and occurs through recruitment of specific junctional molecules.
- Focal adhesions develop from a small cluster of integrins, termed focal complexes, by the sequential recruitment of talin, vinculin, and α-actinin.
- This follows recruitment of formin, which initiates actin bundle formation.
- Myosin II then comes leads the development of tension at the point of adhesion resulting in cell signaling.

Cell to cell integration

- Direct interactions between cells, as well as between cells and the extracellular matrix, are critical to the development and function of multicellular organisms.
- Some cell-cell interactions are transient, such as the interactions between cells of the immune system and the interactions that direct white blood cells to sites of tissue inflammation.
- In other cases, stable cell-cell junctions play a key role in the organization of cells in tissues.
- For example, several different types of stable cell-cell junctions are critical to the maintenance and function of epithelial cell sheets.
- Plant cells also associate with their neighbors not only by interactions between their cell walls, but also by specialized junctions between their plasma membranes called plasmodesmata.

Plasmodesmata

Plant cells, surrounded as they are by cell walls, don't contact one another through wide stretches of plasma membrane the way animal cells can. However, they do have specialized junctions

called **plasmodesmata** (singular, **plasmodesma**), places where a hole is punched in the cell wall to allow direct cytoplasmic exchange between two cells.

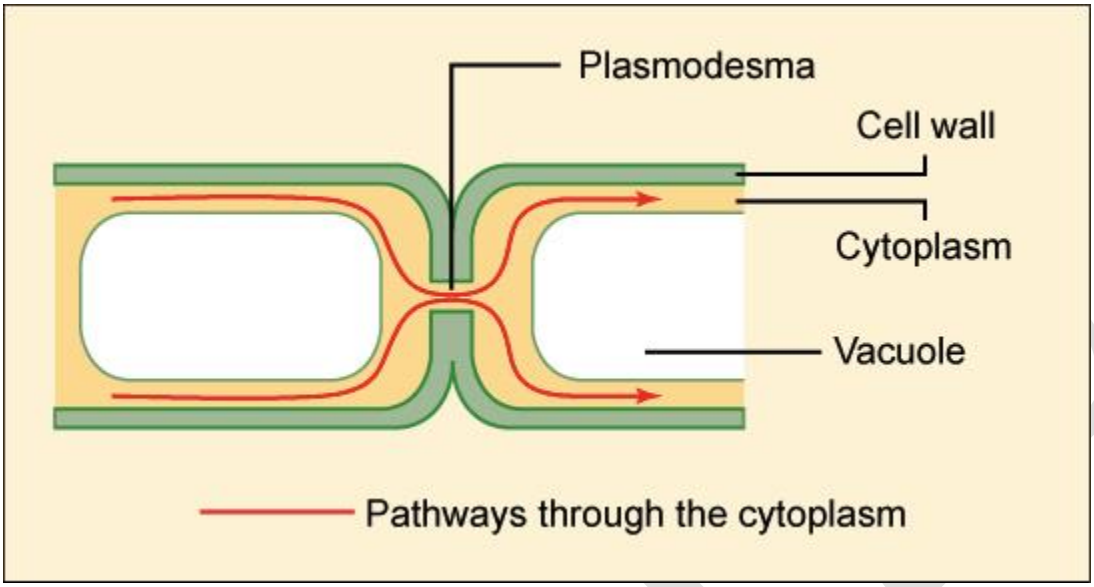


Image of two cells connected by a plasmodesma, showing how materials can travel from the cytoplasm of one cell to the next via the plasmodesma.

Plasmodesmata are lined with plasma membrane that is continuous with the membranes of the two cells. Each plasmodesma has a thread of cytoplasm extending through it, containing an even thinner thread of endoplasmic reticulum (not shown in the diagram above).

Molecules below a certain size (the size exclusion limit) move freely through the plasmodesmal channel by passive diffusion. The size exclusion limit varies among plants, and even among cell types within a plant. Plasmodesmata may selectively dilate (expand) to allow the passage of certain large molecules, such as proteins, although this process is poorly understood.

Gap junctions

Functionally, **gap junctions** in animal cells are a lot like plasmodesmata in plant cells: they are channels between neighboring cells that allow for the transport of ions, water, and other substances. Structurally, however, gap junctions and plasmodesmata are quite different.

In vertebrates, gap junctions develop when a set of six membrane proteins called **connexins** form an elongated, donut-like structure called a **connexon**. When the pores, or "doughnut holes," of connexons in adjacent animal cells align, a channel forms between the cells. (Invertebrates also form gap junctions in a similar way, but use a different set of proteins called innexins.).

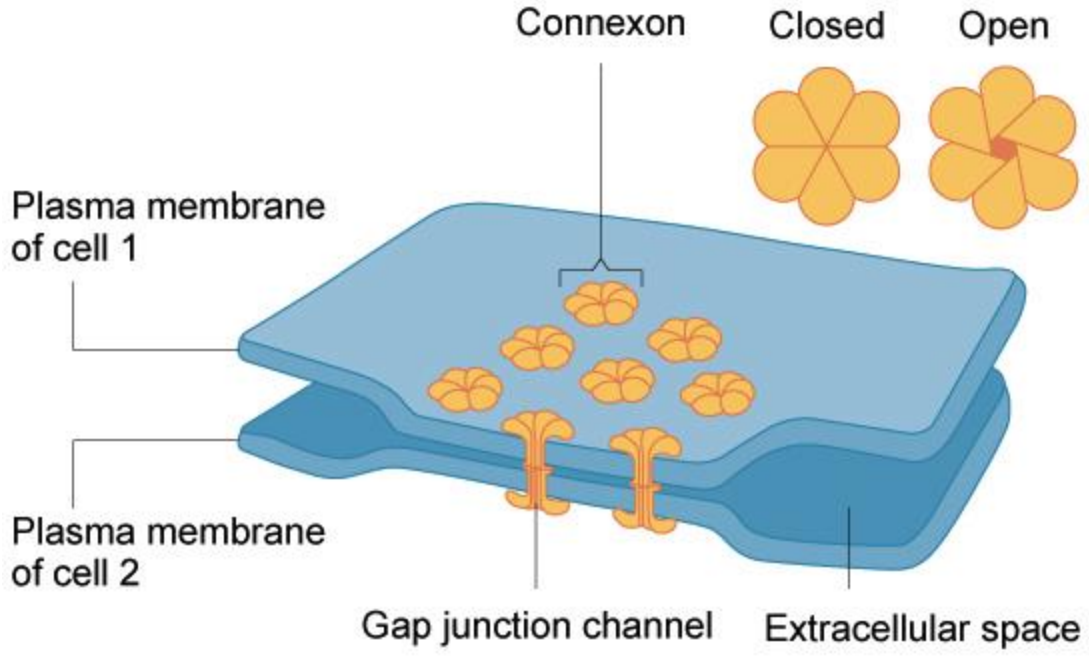


Image of the plasma membranes of two cells held together by gap junctions. Where two connexons from the different cells meet, they can form a channel leading from one cell into the next.

Gap junctions are particularly important in cardiac muscle: the electrical signal to contract spreads rapidly between heart muscle cells as ions pass through gap junctions, allowing the cells to contract in tandem.

Tight junctions

Not all junctions between cells produce cytoplasmic connections. Instead, **tight junctions** create a watertight seal between two adjacent animal cells.

At the site of a tight junction, cells are held tightly against each other by many individual groups of tight junction proteins called **claudins**, each of which interacts with a partner group on the opposite cell membrane. The groups are arranged into strands that form a branching network, with larger numbers of strands making for a tighter seal.

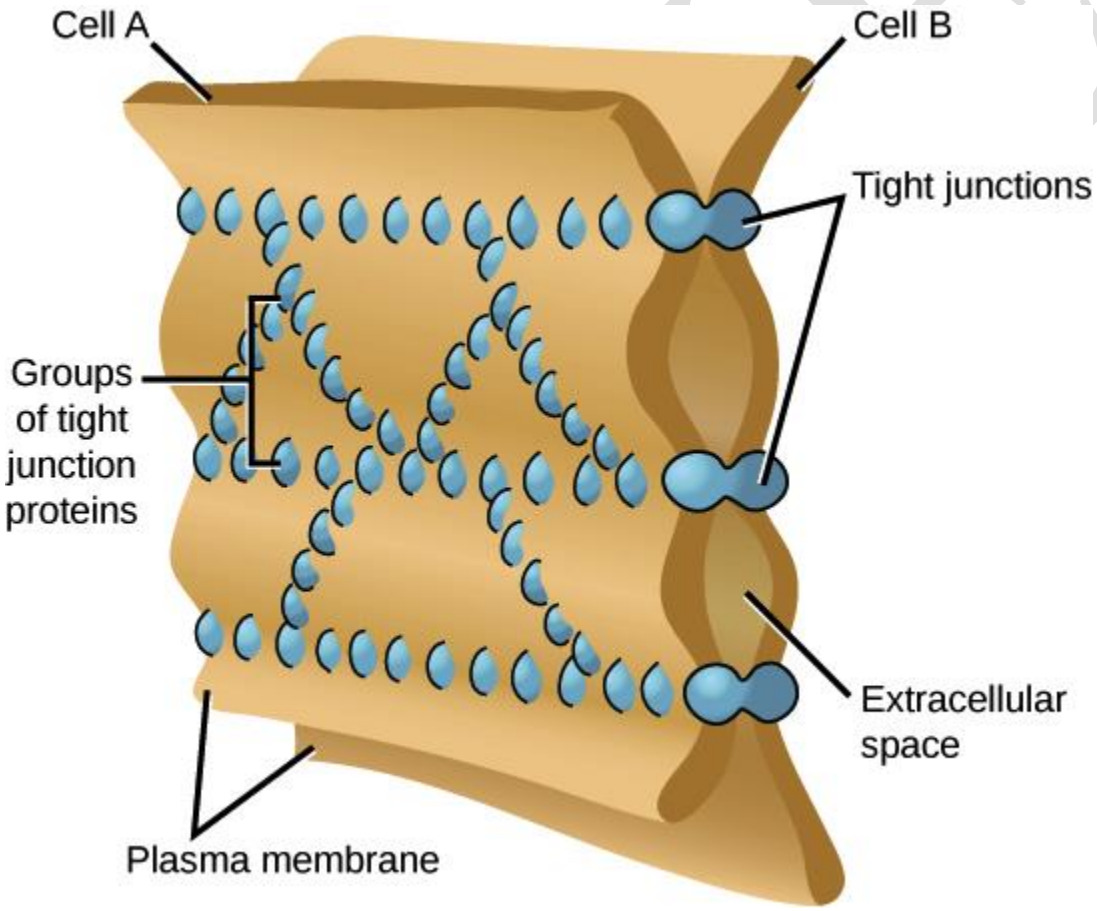


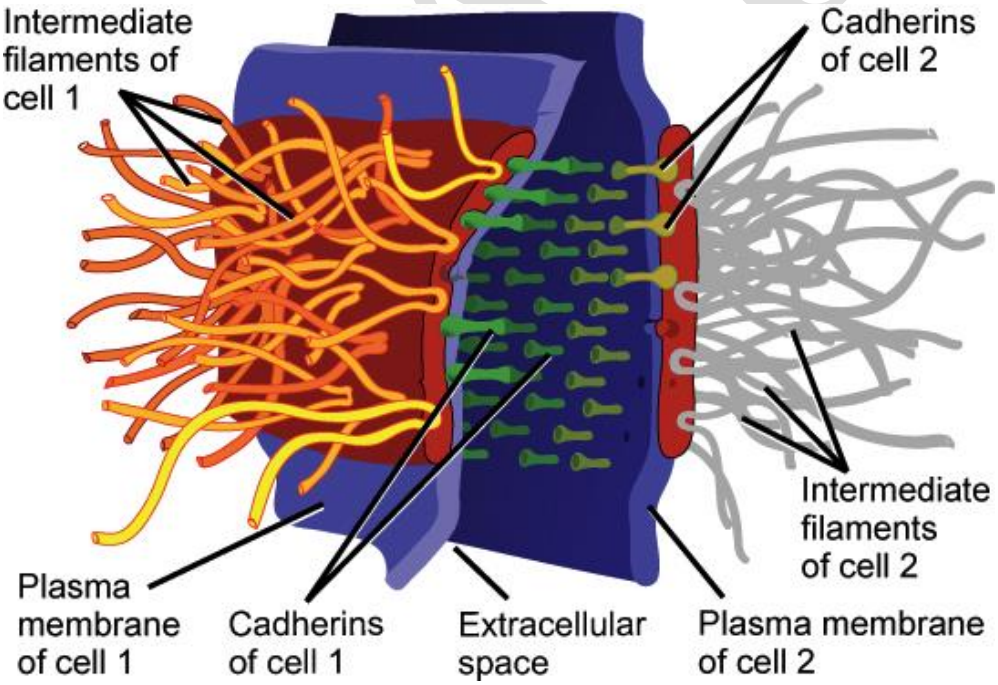
Image of the membranes of two cells held together by tight junctions. The tight junctions are like rivets, and they are arranged in multiple strands that form lines and triangles.

The purpose of tight junctions is to keep liquid from escaping between cells, allowing a layer of cells (for instance, those lining an organ) to act as an impermeable barrier. For example, the tight junctions between the epithelial cells lining your bladder prevent urine from leaking out into the extracellular space.

Desmosomes

Animal cells may also contain junctions called **desmosomes**, which act like spot welds between adjacent epithelial cells. A desmosome involves a complex of proteins. Some of these proteins extend across the membrane, while others anchor the junction within the cell.

Cadherins, specialized adhesion proteins, are found on the membranes of both cells and interact in the space between them, holding the membranes together. Inside the cell, the cadherins attach to a structure called the cytoplasmic plaque (red in the image at right), which connects to the intermediate filaments and helps anchor the junction.



Desmosomes pin adjacent cells together, ensuring that cells in organs and tissues that stretch, such as skin and cardiac muscle, remain connected in an unbroken sheet.

Cell adhesion proteins

- Cell-cell adhesion is a selective process, such that cells adhere only to other cells of specific types.
- This is accomplished with the aid of the selectin and integrin proteins.
- The selectins mediate the initial adhesion this is followed by the formation of more stable adhesions, in which integrins on the surface of leukocytes bind to intercellular adhesion molecules (ICAMs), which are members of the Ig superfamily expressed on the surface of endothelial cells.
- The fourth group of cell adhesion molecules, are the cadherins.
- They are not only involved in selective adhesion between embryonic cells but are also primarily responsible for the formation of stable junctions between cells in tissues.
- The cell-cell interactions mediated by the selectins, integrins, and members of the Ig superfamily are transient adhesions in which the cytoskeletons of adjacent cells are not linked to one another.
- Stable adhesion junctions involving the cytoskeletons of adjacent cells are instead mediated by the cadherins.

Adhesion between plant cells is mediated by their cell walls rather than by transmembrane proteins. In particular, a specialized pectin-rich region of the cell wall called the middle lamella acts as a glue to hold adjacent cells together. Because of the rigidity of plant cell walls, stable associations between plant cells do not require the formation of cytoskeletal links, such as those provided by the desmosomes and adherens junctions of animal cells.

Signal transduction

• Responding to a signal from the environment (intracellular or extra cellular) is a fundamental character of all living cells or organisms.

• Bacteria – respond to changes in oxygen, nutrients, pH, toxins or even other organisms attacking them.

• Plants – respond to light, touch, hormones etc.

• Animals – Developmental signals and others.

• Cells do not do anything without a cue or signal. Hence all the normal functions of a cell require signals/cues.

• From this view point all the pathological conditions/diseases are due to deregulation of one or more signaling pathways.

• Understanding of signaling pathways is essential so that we can target them for therapeutic interventions.

Signals, ligands and receptors

• Signals in biology refer to the molecules, ions, gases, sound, mechanical touch, stress, temperature, heat and physical factors that elicit a response when they communicate/contact with a cell or when they bind or diffuse into a cell.

• If the signal comes from outside the cell – extracellular signal.

• If the signal is generated inside the cell - intracellular signal.

• If the signaling molecule binds with a receptor - call it a Ligand/agonist/first messenger (with a message).

• Receptors bind with ligands and can be located on the cell surface, cytosol or nucleus.

Ligands/ First messengers

• Lipophilic molecules can enter the membrane and bind to intracellular receptors: steroids, thyroxine, and retinoic acids.

• Lipophilic molecules binding to cell surface receptors: prostaglandins.

• Hydrophilic molecules that are not able to cross the membrane bind to cell surface receptors:

• peptides: growth hormones, cytokines, gastrin, glucagon, insulin, TSH, LH, FSH, Parathyroid hormone, secretin

• b) amino acid derivatives : epinephrine, norepinephrine, serotonin, dopamine, histamine.

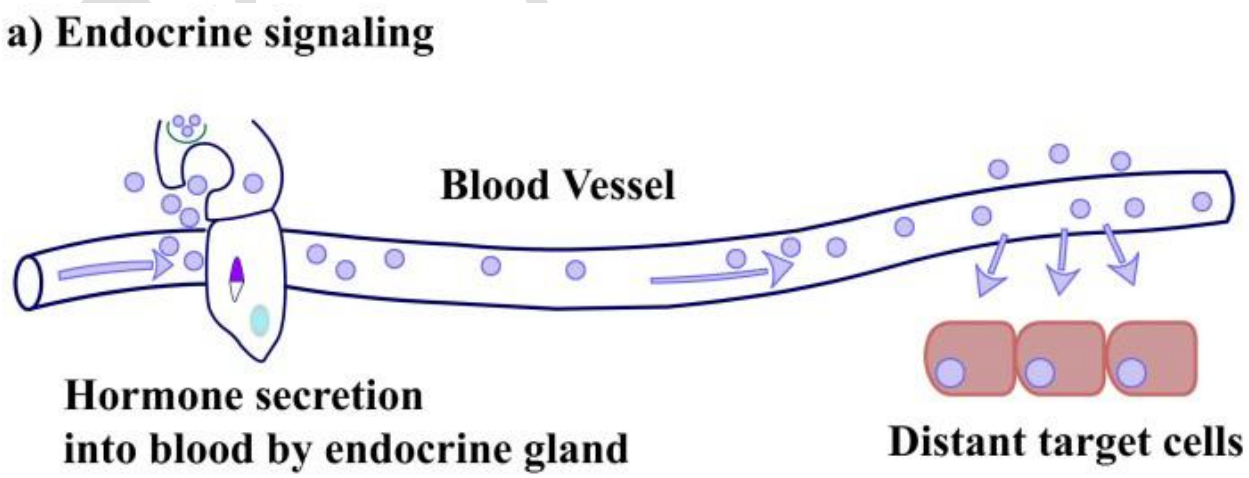
• Some ligands are initially synthesized as trans membrane inactive proteins and then cleaved by enzymes and secreted out into the extracellular compartment Growth factors

• The word "signal transduction" started to appear in the biological literature in the 1970s.

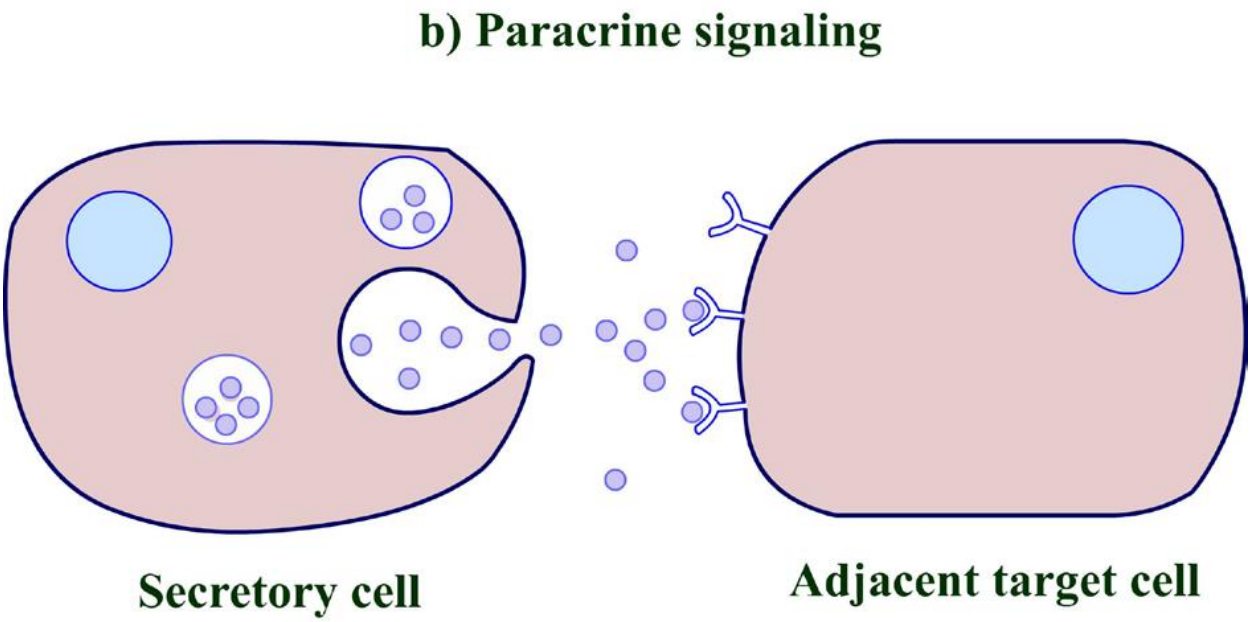
• GTP and GTP-binding proteins in metabolic regulation were described by Alfred G. Gilman and Martin Rodbell

• They were awarded the Nobel Prize in 1994 for their discovery of G-proteins and their role in signal transduction in cells

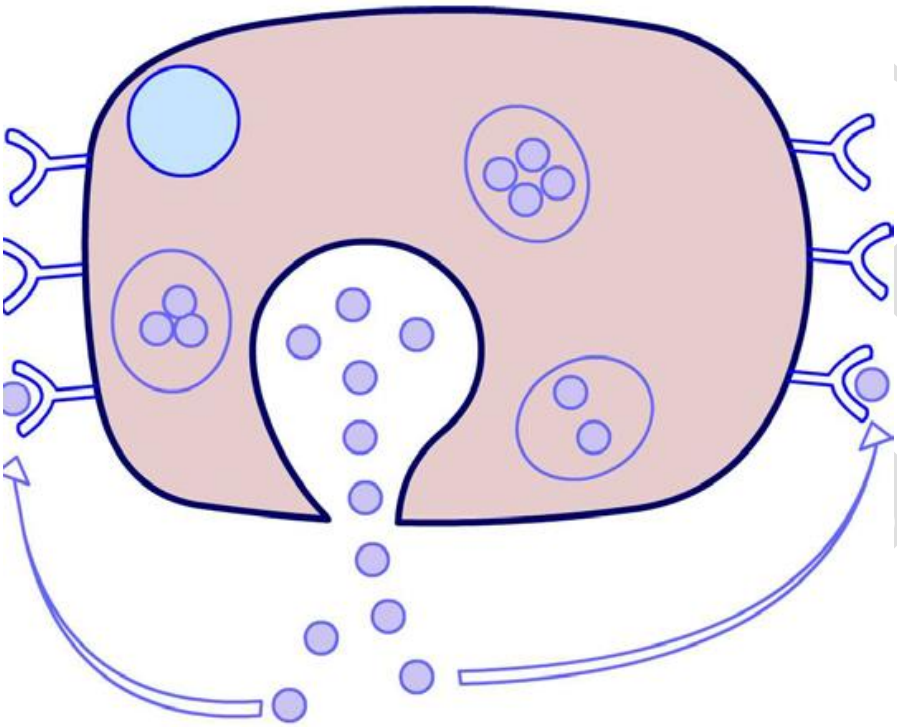
Signaling Molecules Operate Over Various Distances



b) Paracrine signaling



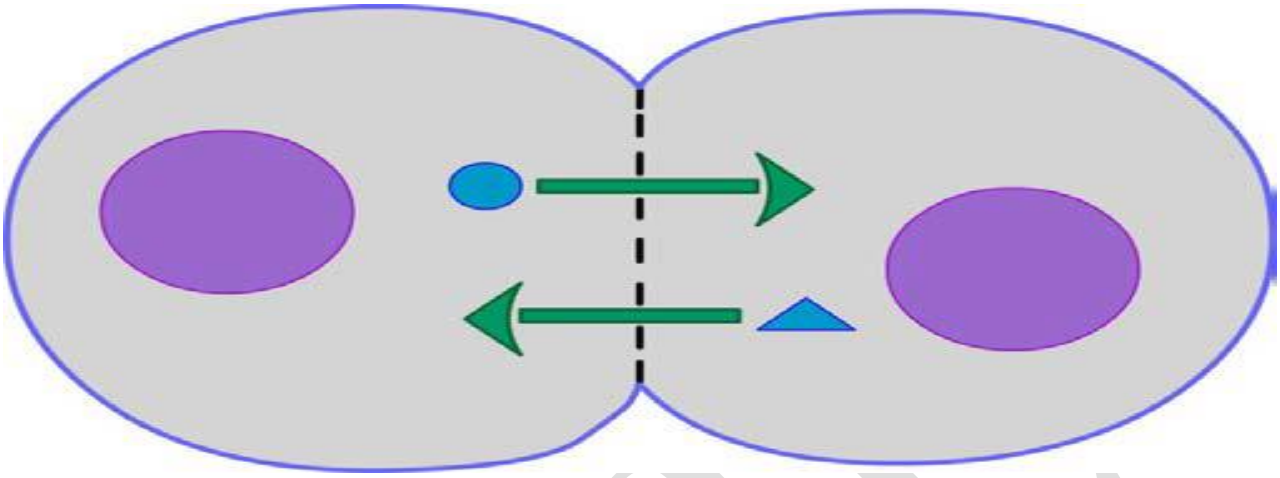
c) Autocrine signaling



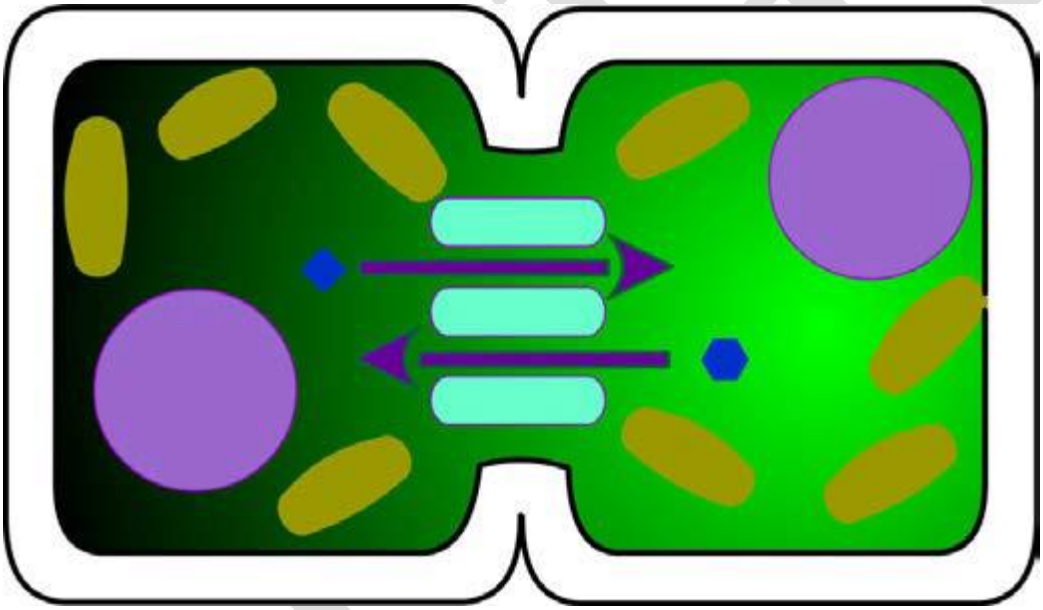
Target sites on same cell

Animal and plant cells

• Have cell junctions that directly connect the cytoplasm of adjacent cells



Gap junctions between animal cells

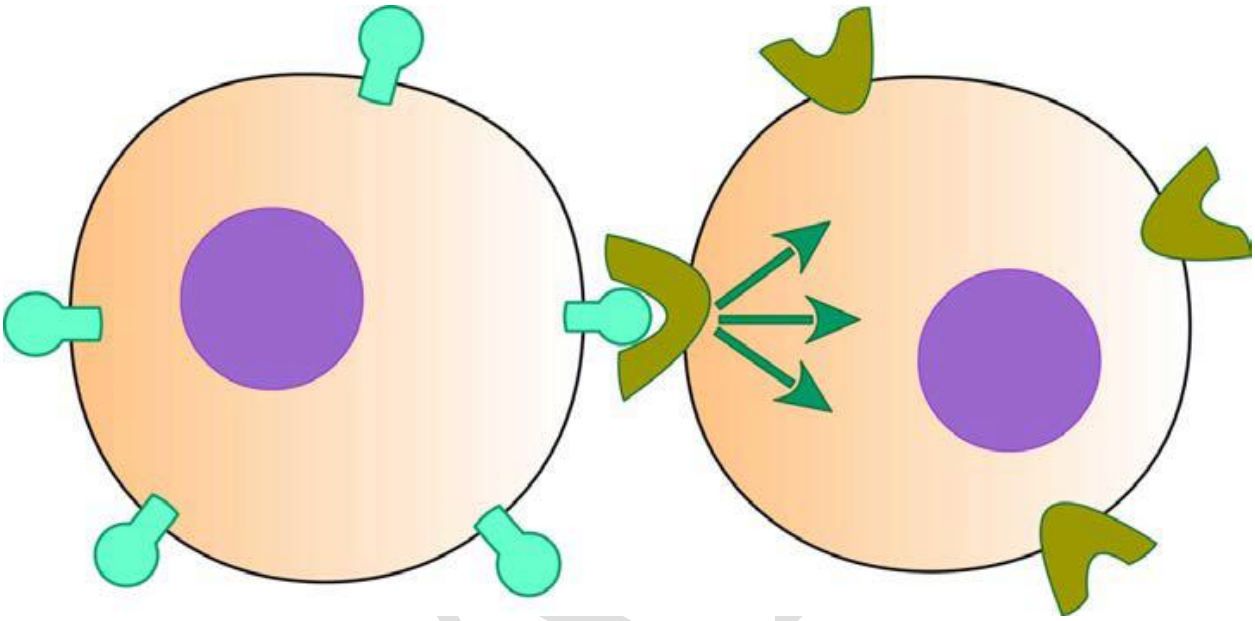


Plasmodesmata between plant cells

Cell junctions: Both animals and plants have cell junctions that allow molecules to pass readily between adjacent cells without crossing plasma membranes

In local signaling, animal cells

• May communicate via direct contact



Specificity

• Specificity is an important criterion in ligand-receptor interactions. High specificity means the ligand binds with a specific receptor that is expressed only in certain cell types.

• Receptor and ligand generally have complementary structural features.

• Non-covalent interactions similar to antigen-antibody reactions and enzyme-substrate reactions with high affinity (low Kd values) co-operativity.

Amplification

• The actual cell concentrations of ligands may be very low and are often transient and short-lived but the effects they ellicit are not small and may be some major changes.

• Ligand-receptor binding may result in the activation of a cascade of enzymes.

• Amplification is achieved through multiple enzymes and scaffolding proteins.

Signal integration

• Cells often receive multiple signals and they can be conflicting signals from reciprocal pathways

• Parallel signaling pathways may be activated

• Several signaling pathways may also work for a common goal

• Therefore the signals, pathways and systems have to be coordinated to provide an integrated cellular response

Termination of signal/Desensitization

• A transient signal emanating from a ligand must produce a rapid and major cellular response.

• Sometimes the signal persists and the sensitivity to the ligand decreases.

• Desensitization can be ahieved by a feedback loop, dephosphorylation, endocytosis.

• The receptor may now have less affinity, less activity or less expression.

Cell-surface and nuclear receptors

• G Protein Coupled receptors

• Ion-channel receptors

• Receptor tyrosine kinases

• Receptor serine/threonine kinases

• Receptors with phosphatase activity

• Receptors lacking enzyme activity but associate with intracellular enzymes

• Cell adhesion receptors

• Nuclear receptors

Transduction

• **Transduction** means the action of leading across or bringing across

• The action of transferring the signal from one compartment of the cell to another or to transmit the signal from outside to inside

• Conveying the message from the signal/ligand into a response that is essentially a chemical change

• This is mediated by **receptors** and **signaling intermediates**

• Intracellular signaling is often initiated with second messengers such as cAMP

• Earl W. Sutherland was awarded the Nobel Prize in 1971 for his discoveries concerning the mechanisms of action of hormones.

• Identification of cyclic AMP (cAMP) as the second messenger to adrenaline or glucagon was a great discovery as it became possible to link activation of specific classes of receptors with specific biochemical responses.

Signaling intermediates

• Cellular signaling intermediates include kinases, phosphatases, GTPases and adapter proteins with interaction domains forming a scaffold.

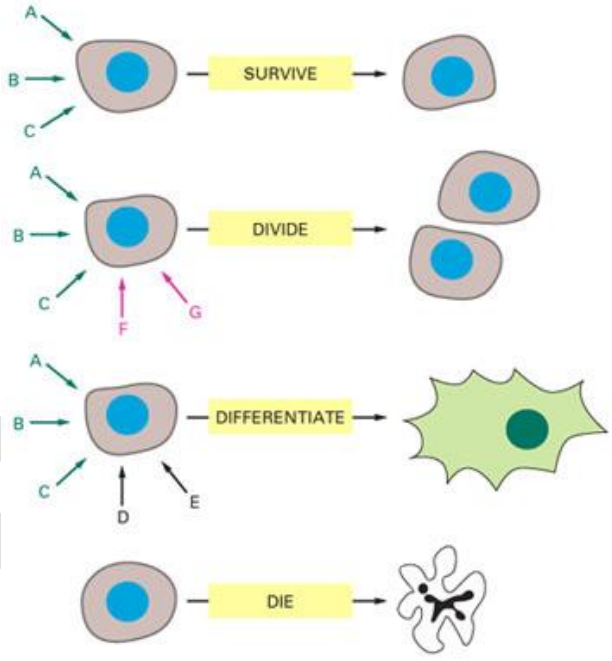
• They help in transmitting the signal and its amplification

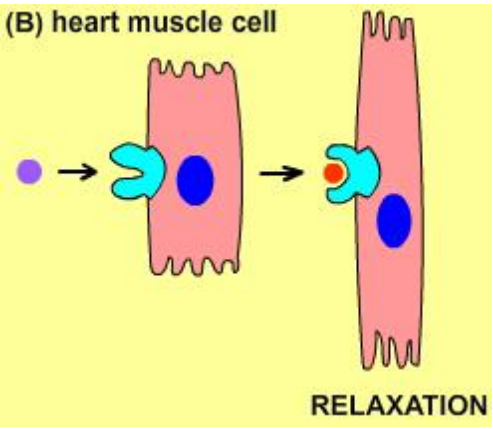
Responses

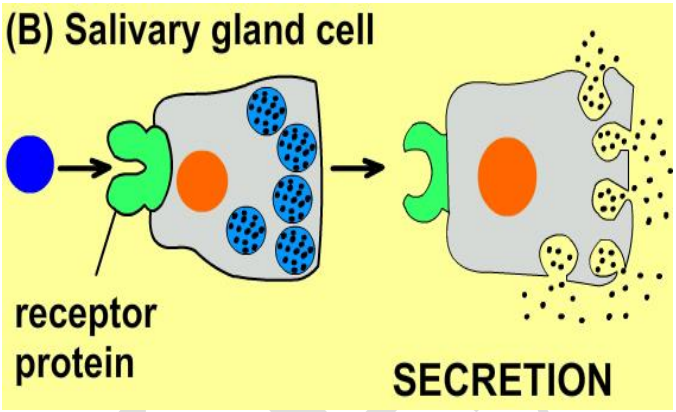
• Cells need **cues or signals** to carry out their functions or physiological processes.

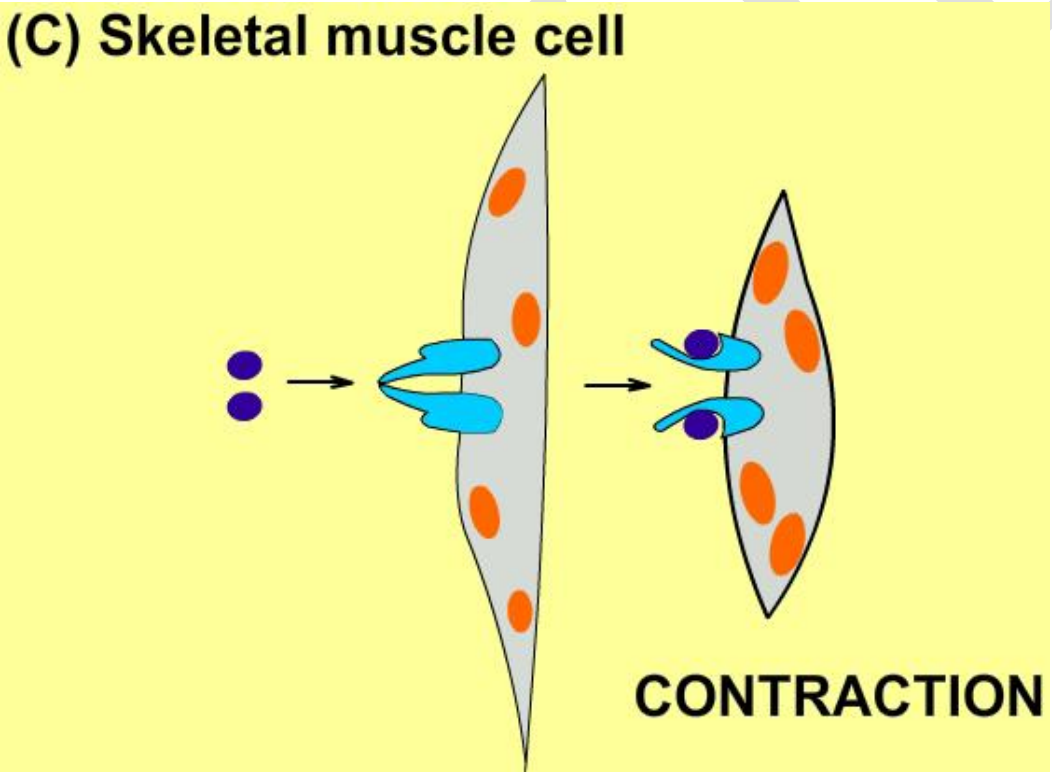
• What are the physiological processes?

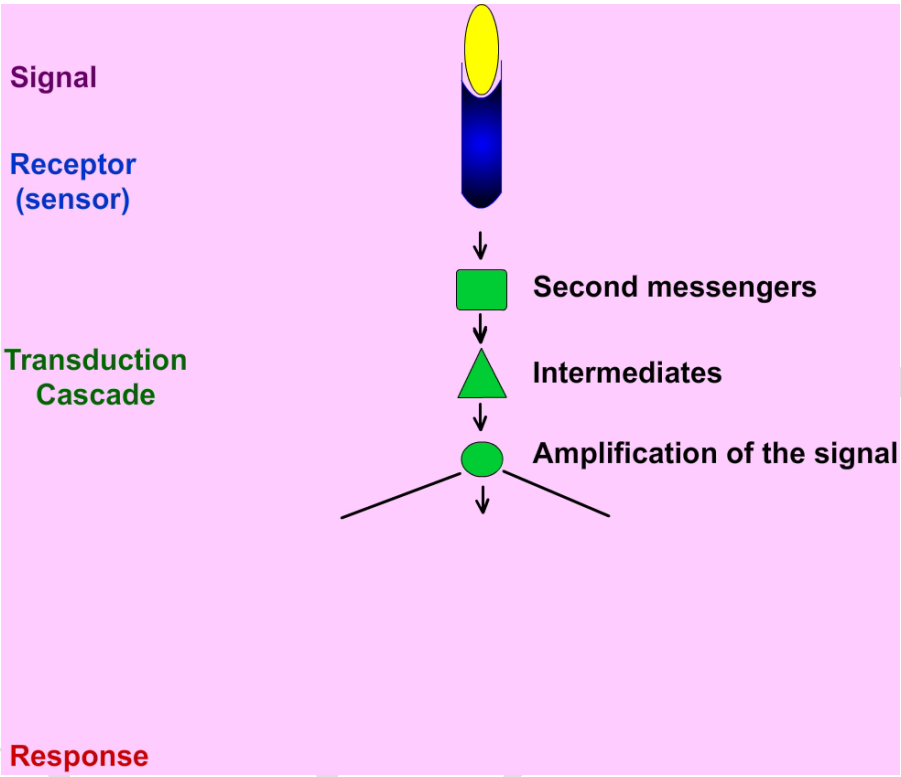
• Development, proliferation, differentiation, migration, cell death, survival etc need signals.




(B) heart muscle cell
RELAXATION


(B) Salivary gland cell
receptor protein
SECRETION


(C) Skeletal muscle cell
CONTRACTION



Major events of Signal transduction

Generation of signals

Ligands

Recognition of signals

Receptors

Transduction and amplification

Change of external signal into intracellular message

Effects or responses

Modification of cell behavior, gene expression, function etc

Termination/Desensitization of signals

Receptor endocytosis, Dephosphorylation

Dimerization

• Homodimers – formed between two identical receptors

• EGFR-EGFR

• Heterodimers – formed between two different receptors

• EGFR-ErbB2

• What are the advantages of dimerization?

Dimerization is an important mechanism for signal transduction

• Dimers have certain advantages in signaling than a monomer

• Relative distance and orientation of dimers (apart from ligands some antibodies

• can also activate some RTKs by their ability to cross bridge the dimers)

• Specificity can be generated for the ligand in a heterodimer

• Only one receptor may have the binding site for the ligand

• Even if one partner is inactive in its enzyme activity the other one can bind

• Increased affinity may be achieved through multiple contact points

• Two receptors may bring in several ligands for the given signaling pathway

• Multiple intracellular interaction partners

• Cross phosphorylation or transphosphorylation is achieved by dimers.

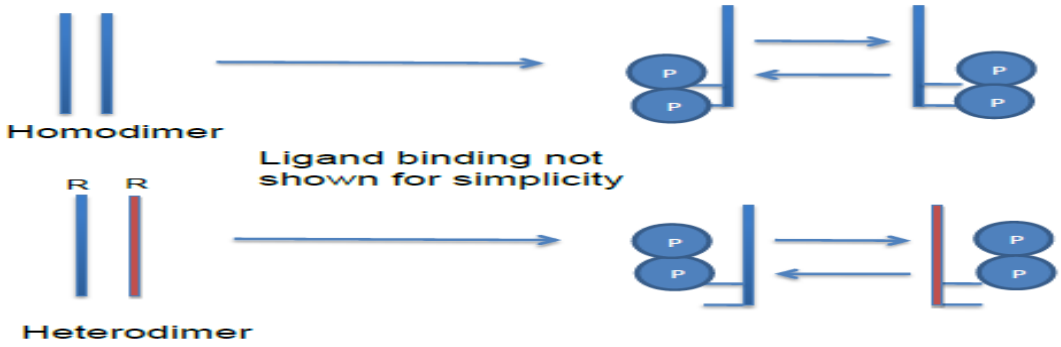
Phosphorylation

• Phosphorylation is an important mechanism of receptor activation.

• More than 99% of the phosphoamino acids in normal cells are phosphothronine and phosphoserine.

• Phosphotyrosine constitutes only 0.05 – 0.1 % of total

• phosphoamino acids (but very important!)



Possible Questions

2 Marks

1. Define extracellular matrix.
2. What is focal adhesion.
3. Define plasmadesmata.
4. Define gap junction.
5. Define tight junction.

8 Marks

1. Explain the concept of Extracellular matrix
2. Give a detailed account on Cell adhesion
3. Explain membrane receptors
4. Explain the Adhesion proteins
5. Write about types of cadherins and their features
6. Explain Selectins
7. Explain G-protein coupled receptor
8. Write in detail signal transduction
9. Explain Cell Adhesion Molecules (CAM)

Unit III

Membrane Vacuolar system, Cytoskeleton and Cell Motility: Structure and function of Microtubules, Microfilaments, Intermediate filaments. Endoplasmic Reticulum: Structure and function including role in protein segregation. Golgi complex: Structure biogenesis and function including role in protein secretion.

Membrane Vascular System:

Cytoskeletal elements and architecture :

- The cytoskeleton can be defined as a cytoplasmic system of fibers which is critical to cell motility.
- It is dynamic three-dimensional scaffolding contained within a cell's cytoplasm and is made of protein.
- The ability of eukaryotic cells to adopt a variety of shapes and to carry out coordinated and directed movements depends on the cytoskeleton.
- The cytoskeleton was known to be unique to eukaryotic cells.
- The cytoskeleton can also be referred to as cytomusculature, because :
  - (i) it is directly involved in movements such as crawling of cells on a substratum,
  - (ii) muscle contraction and the various changes in the shape of a developing vertebrate embryo;
  - (iii) it also provides the machinery for cyctosis in cytoplasm.

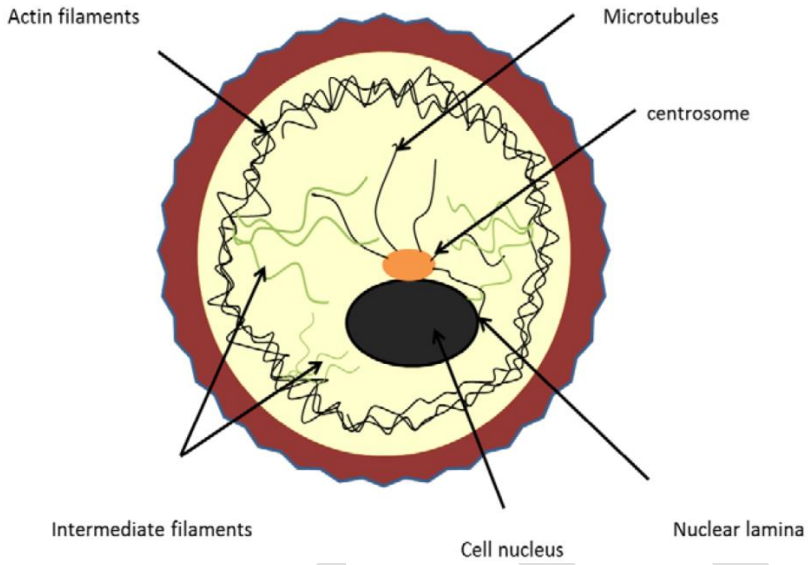


Fig:The cytoskeletal system

- The main proteins that are present in the cytoskeleton are *tubulin* (in the microtubules), *actin, myosin, tropomyosin* and other (in the microfilaments) and *keratins, vimentin, desmin, lamin* and others (in intermediate filaments).
- Tubulin and actin are globular proteins, while subunits of intermediate filaments are fibrous proteins.
- The primary types of fibers comprising the cytoskeleton are:
  (i) Microfilaments
  (ii) Intermediate filaments
  (iii) Microtubules

Table: Differences among cytoskeletal elements

Microfilaments	Intermediate filaments	Microtubules
Depolymerize into their soluble subunits	Extremely stable	Depolymerize into their soluble subunits
7 nm in diameter	10 nm in diameter	24 nm in diameter
Beaded structure	α-helical rods that assemble into ropelike filaments	Hollow tubules

Require nucleotide hydrolysis for polymerization G-actin	Subunits do not require nucleotide hydrolysis for polymerization	Require nucleotide hydrolysis for polymerization of αβ-tubulin

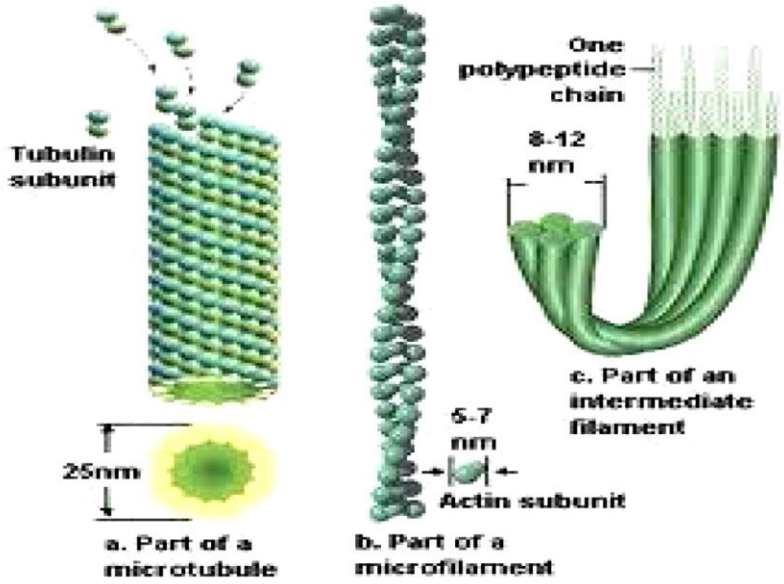


Fig :The difference among the various cytoskeletal systems

a. Microfilaments :

Structure :

- Microfilaments are involved in cell locomotion.
- Microfilaments also extend into cell processes, especially where there is movement.
- Thus, they are found in the microvilli of the brush border of intestinal epitheliun and in cell types where amoeboid movement and cytoplasmic streaming are prominent.
- Microfilaments are powered by actin cytoskeleton which is a medium sized protein of 375 amino acid residues which is encoded by a highly conserved gene family.
- Actin proteins are localized in cytoplasm, nucleus and in the muscles.
- However the richest area of actin filaments in a cell lies in a narrow zone just beneath the plasma membrane known as the cortex.

- Actin protein is structurally globular composed of G-actin and F-actin; which in turn is a linear chain of G-actin subunits.
- Each actin molecule contains an Mg2+ ion co-factor bound ATP or ADP.
- Thus there are four states of actin: ATP – G-actin, ADP – G-actin, ATP – F-actin, and ADP – F-actin.
- The assembly of G-actin into F-actin is accompanied by the hydrolysis of ATP to ADP and Pi.
- In F-filament all actin moieties point toward the same filament end.
- ATP-binding cleft of an actin subunit is exposed to the surrounding solution.
- Finally actin filaments form bundles and networks which provide a framework that supports the plasma membrane.
- Long, flexible cross-linking proteins are able to adapt to any arrangement of actin filaments and tether orthogonally oriented actin filaments in networks.
- Membrane microfilament binding proteins join membrane to the cytoskeleton framework.
- The simplest connections entail binding of integral membrane proteins directly to actin filaments.

Function :

1. An important function of actin microfilament is that it can produce movement in the absence of motor proteins. At the cell membrane microfilament assembly protrudes the membrane forward producing the ruffling membranes in actively moving cells.

2. Microfilaments can also play a passive structural role by providing the internal stiffening rods in microvilli, maintaining cell shape, and anchoring cytoskeletal proteins.

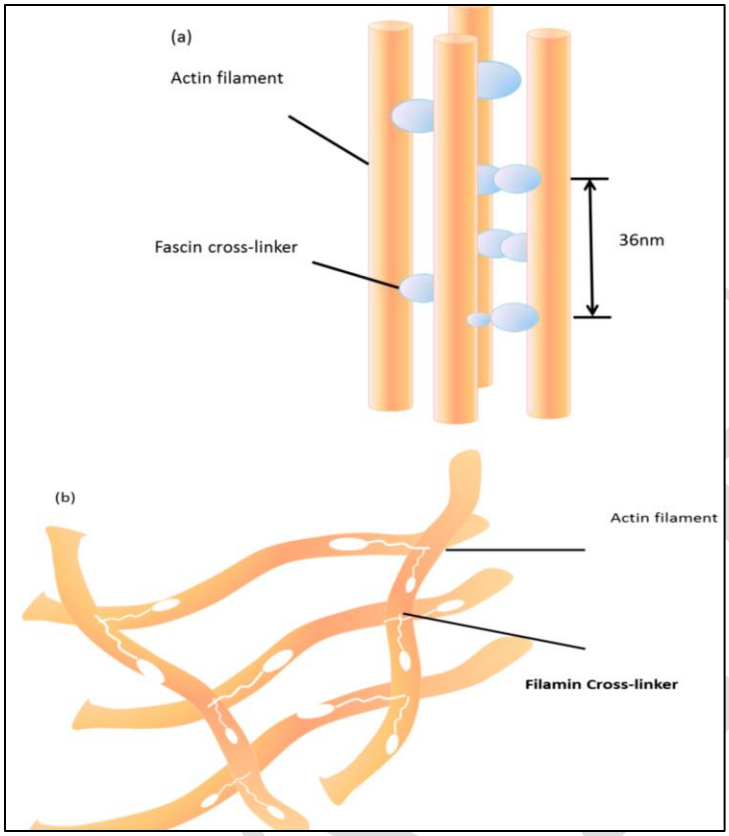


Fig : Actin cross-linking proteins bridging pairs of actin filaments.

b. Intermediate filaments :

- Intermediate filaments (IFs) are tough, durable protein fibres in the cytoplasm of most higher eukaryotic cells typically between 8 nm to 10 nm in diameter.
- They are particularly prominent where cells are subjected to mechanical stress, such as in epithelia, where they are linked from cell to cell at desmosomal junctions, along the length of axons, and throughout the cytoplasm of smooth muscle cells.
- Intermediate filaments are typically organized in the cytosol as an extended system that stretches from the nuclear envelope to the plasma membrane.
- Some intermediate filaments run parallel to the cell surface, while others traverse the cytosol.

- They also form the nuclear lamina. In cross-section, intermediate filaments have a tubular appearance. Each tubule appears to be made up of 4 or 5 protofilaments arranged in parallel fashion (Figure 2).
- IFs are composed of polypeptides of a surprisingly wide range of sizes (from about 40,000 to 130,000 daltons).
- Protein subunits from the family of α-helical proteins make the intermediate filaments and these protein subunits can be divided into six major classes which are widely divergent in sequence and vary greatly in molecular weight.
- *The keratins are the most diverse classes of IF proteins and can be divided into two groups:*
  (i) keratins specific for tough epithelial tissues, which give rise to nails, hair, and wool and cytokeratins which are more generally found in the epithelia that line internal body cavities.
  (ii) Each type of epithelium always expresses a characteristic combination of type I and type II keratins which associate in a 1:1 ratio to form heterodimers, which assemble into heteropolymeric keratin filaments.
  (iii) Apart from keratins most widely distributed of all IF class III proteins is vimentin, which is typically expressed inleukocytes, blood vessel endothelial cells, some epithelial cells, and mesenchymal cells such as fibroblasts.
  (iv) Vimentin filaments help support cellular membranes.

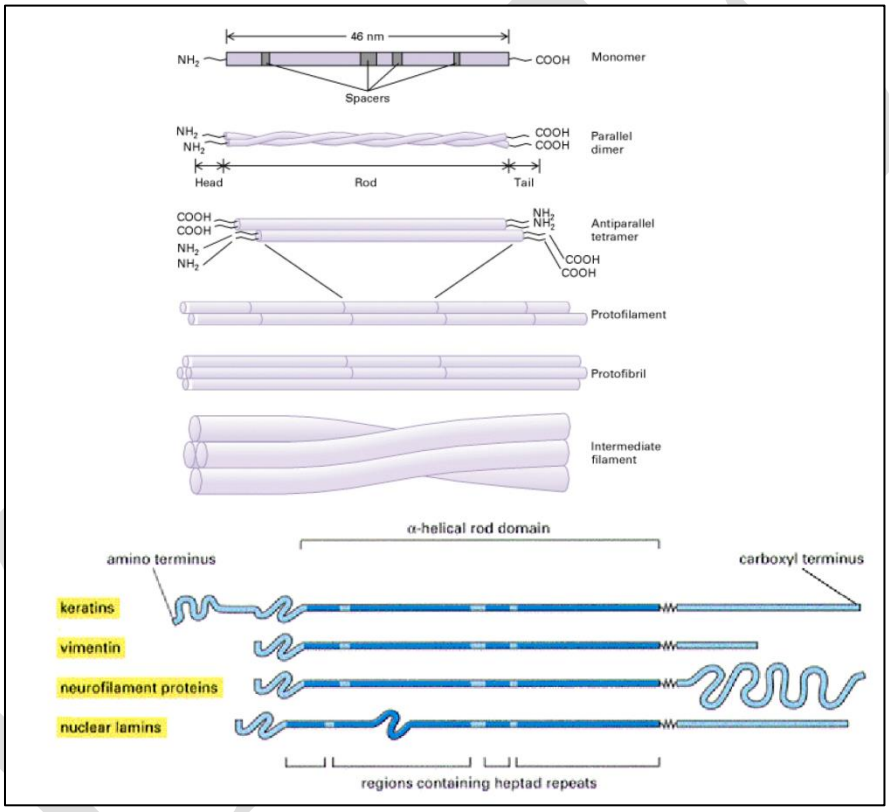
Structure :

- Intermediate filament proteins are 10 nm in diameter, a central α-helical conserved core flanked by globular N- and C-terminal domains which vary in different IF proteins.
- The core helical domain is conserved among all IF proteins.
- It consists of four α-helices separated by three spacer regions.

The polypeptide chains are parallel in a dimer. A pair of dimers associate laterally into a tetramer.

- Proteins cross-link intermediate filaments with one another, forming a bundle (a tonofilament) or a network, and withother cell structures, including the plasma membrane.

Classes of proteins making the intermediate filaments. The values just indicates the molecular mass range of different proteins.



IF protein	MW (10^{-3})	Tissue distribution
Type I		
Acidic keratins	40-57	Epithelia
Type II		
Basic keratins	53-67	Epithelia
Type III		
Vimentin	57	Mesenchyme
Desmin	53	Muscle
Glial fibrillay acidic protein	50	Glial cells and astrocytes
Peripherin	57	Neurons
Type IV		
NF-L	62	Mature neurons
NF-M	102	Mature neurons
NF-H	110	Mature neurons
Internexins	66	Developing central nervous system
Non standard type IV		
Filensin	83	Lens fibre cells
Phakinin	45	
Type V		Cell nucleus
Lamin A	70	
Lamin B	67	
Lamin C	67	

Function:

1. The main function of Intermediate filament is mechanical support.

- The best example is the nuclear lamina along the inner surface of the nuclear membrane.
- IFs in epithelia form a transcellular network that resists external forces.

- The neurofilaments in the nerve cell axons resist stresses caused by the motion of the animal, which would otherwise break these long, thin cylinders of cytoplasm.
- Desmin filaments provide mechanical support for the sarcomeres in muscle cells, and vimentin filaments surround and probably support the large fat droplets in the fat cells.

2. They form an internal framework that helps support the shape of the cell.

- In vitro binding experiments suggest that at the plasma membrane, vimentin filaments bind two proteins:
  - ankyrin, the actinbinding protein associated with the Na+/K+ ATPase in nonerythroid cells,
  - and plectin.

b. Microtubules :

- A microtubule is a polymer of globular tubulin subunits, which are arranged in a cylindrical tube measuring 24 nm in diameter which is more than twice the width of an intermediate filament and three times the width of a microfilament.
- Microtubules are also much stiffer than either microfilaments or intermediate filaments because of their tubelike construction.
- The building block of a microtubule is the tubulin subunit, a heterodimer of α- and β-tubulin.
- Both of these 55,000-MW monomers are found in all eukaryotes, and their sequences are highly conserved. Although a third tubulin, γ-tubulin, is not part of the tubulin subunit, it probably nucleates the polymerization of subunits to form αβ-microtubules.
- The interactions holding α-tubulin and β-tubulin in a heterodimeric complex and are strong enough ensuring rare dissociation of a tubulin subunit under normal conditions.
- Each tubulin subunit binds two molecules of GTP.
- One GTP-binding site is located in α-tubuli and binds GTP irreversibly and does not hydrolyze it, whereas the second site, located on β-tubulin, binds GTP reversibly and hydrolyzes it to GDP.

- In a microtubule, lateral and longitudinal interactions between the tubulin subunits are responsible for maintaining the tubular form.
- Longitudinal contacts between the ends of adjacent subunits link the subunits head to tail into a linear protofilament.
- Every microtubule in a cell is a simple tube or a singlet microtubule, built from 13 protofilaments.
- In addition to the simple singlet structure, doublet or triplet microtubules are found in specialized structures such as cilia and flagella (doublet microtubules) and centrioles and basal bodies (triplet microtubules).

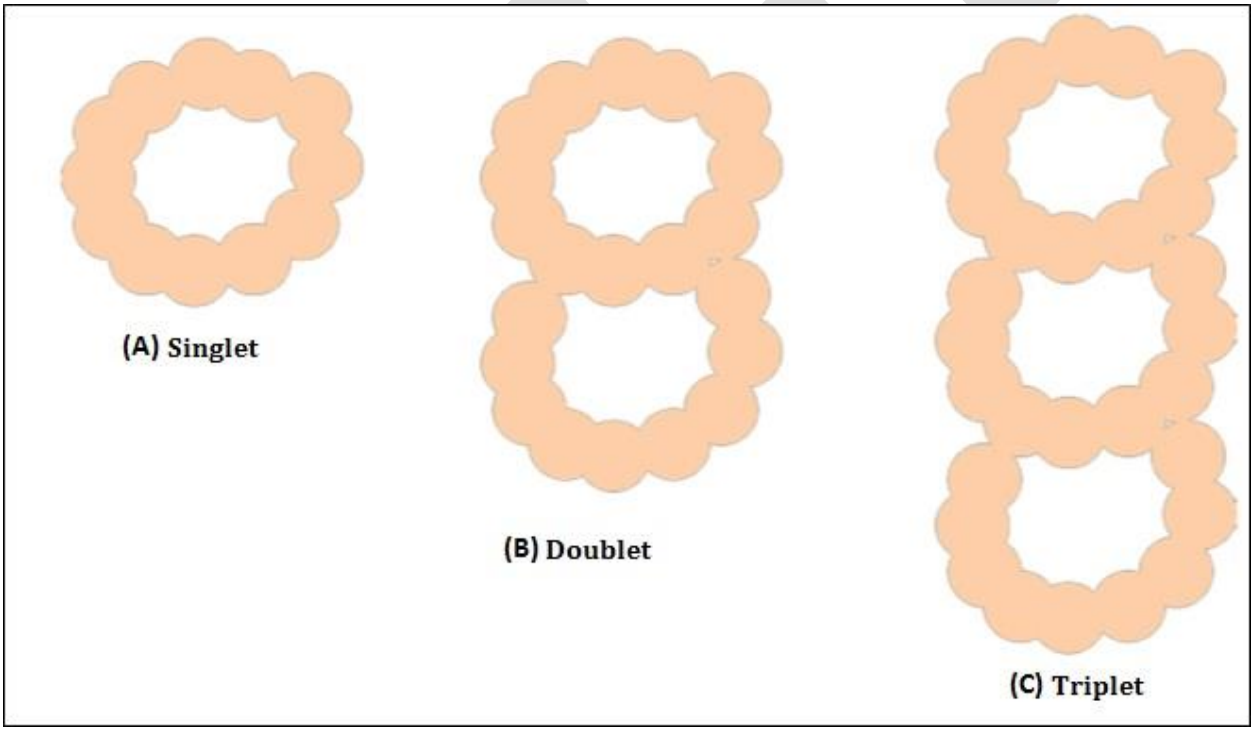


Fig :*In cross section, a typical microtubule, a singlet, is a simple tube built from 13 protofilaments. In a doublet microtubule, an additional set of 10 protofilaments forms a second tubule 'B' by fusing to the wall of a singlet 'A' microtubule. Attachment of another 10*

protofilaments to the 'B' tubule of a doublet microtubule creates a 'C' tubule and a triplet structure.

Functions :

1. Mechanical function:

The shape of the cell (red blood cells of non-mammalian vertebrates) and cells such as axons and dendrites of neurons, microvilli, etc., have been correlated to the orientation and distribution of microtubules.

2. Morphogenesis:

- During cell differentiation, the mechanical function of microtubules is used todetermine the shape of the developing cells.
- The enormous elongation in the nucleus of the spermatid during spermiogenesis is accompanied by the production of an orderly array of microtubules that are wrapped around the nucleus in a double helical arrangement.
- Similarly, the elongation of the cells during induction of the lens placode in the eye is also accompanied by the appearance of numerous microtubules.

3. Cellular polarity and motility:

- The determination of the intrinsic polarity of certain cells is governed by the microtubules.
- Directional gliding of cultured cells is depended on the microtubules.

4. Contraction:

- Microtubules play a role in the contraction of the spindle and movement of
- chromosomes and centrioles as well as in ciliary and flagellar motion.

5. Circulation and transport:

- Microtubules are involved in the transport of macromol-ecules, granules and vesicles within the cell.
- The protozoan *Actinosphaerium* (Heliozoa) sends out long, thin pseudopodia within which cytoplasmic particles migrate back and forth.
- These pseudopodia contain as many as 500 microtubules disposed in a helical configuration.

6. The Microtubule Organizing Centre (MTOC):

- MTOC is the major organizing structure in a cell and helps determine the organization of microtubule-associated structures and organelles *(e.g., mitochondria, the Golgi complex, and the endoplasmic reticulum).*
- In a nonpolarized animal cell such as a fibroblast, an MTOC is perinuclear and strikingly at the center of the cell.
- Because microtubules assemble from the MTOC, microtubule polarity becomes fixed in a characteristic orientation.

- In most animal cells, for instance, the (−) ends of microtubules are closest to the MTOC or basal body (refere diagram - down).
- During mitosis, the centrosome duplicates and migrates to new positions flanking the nucleus.
- There the centrosome becomes the organizing center for microtubules forming the mitotic apparatus, which will separate the chromosomes into the daughter cells during mitosis.

7. The microtubules in the axon of a nerve cell are all oriented in the same direction and help

stabilize the long process of nerve conduction.

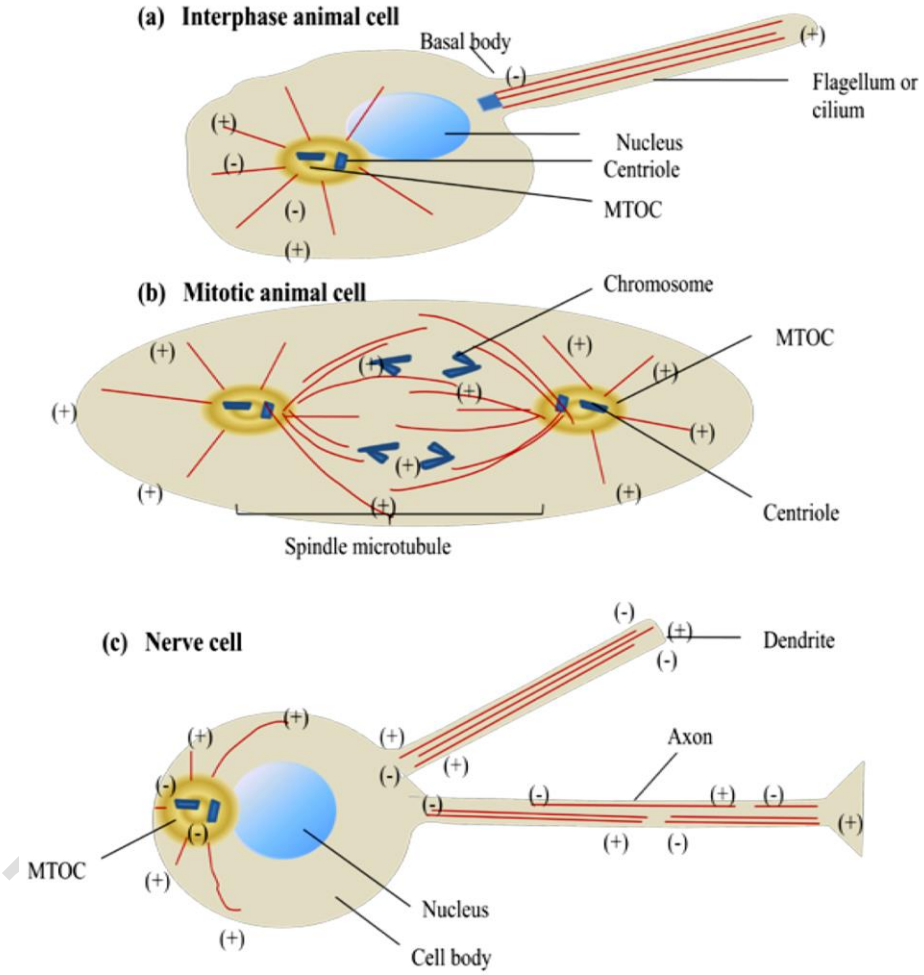


Fig :*(a) In interphase animal cells, the (−) ends of most microtubules areproximal to the MTOC. Similarly, the microtubules in flagella and cilia havetheir (−) ends continuous with the basal body, which acts as the MTOC inthese structures.*

(b) As cells enter mitosis, the microtubule networkrearranges, forming a mitotic spindle. The (−) ends of all spindlemicrotubules point toward one of the two MTOCs, or poles, as they arecalled in mitotic cells.

(c) In nerve cells, the (−) ends of axonal microtubules areoriented toward the base of the axon. However, dendritic microtubules havemixed polarities.

Endoplasmic reticulum:

- Endoplasmic reticulum is a network of interconnected internal membranes generally, the largest membrane in a eukaryotic cell—an extensive network of closed, flattened membrane-bounded sacs called cisternae.
- The endoplasmic reticulum has a number of functions in the cell but is particularly important in the synthesis of lipids, membrane proteins, and secreted proteins.

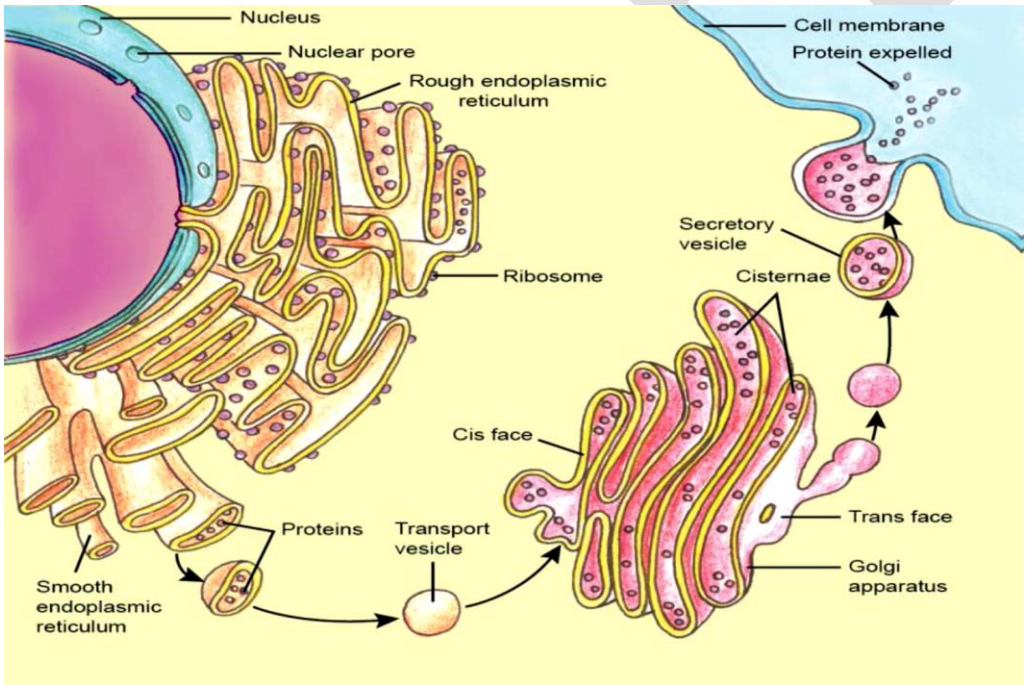


Fig :*The Endoplasmic reticulum.*

Occurrence:

- The occurrence of the endoplasmic reticulum is in eukaryotic cells with variation in its position from cell to cell.
- The erythrocytes (RBC), egg and embryonic cells lack in endoplasmic reticulum.
- ER is poorly developed in certain cells as the RBC which produces only proteins to be retained in the cytoplasmic matrix (haemoglobin), although the cell may contain many ribosomes).
- The spermatocytes also have poorly developed endoplasmic reticulum.

Morphology:

The endoplasmic reticulum occurs in three forms:

(i) Lamellar form or cisternae which is a closed, fluid-filled sac, vesicle or cavity is called ***cisternae***;

(ii) vesicular form or ***vesicle***and

(iii) tubular form or ***tubules***.

1. Cisternae:

- The cisternae are long, flattened, sac-like, unbranched tubules having diameter of 40 to 50 μm.
- They remain arranged parallely in bundles or stakes.
- RER mostly exists as cisternae which occur in those cells which have synthetic roles as the cells of pancreas, notochord and brain.

2. Vesicles:

- The vesicles are oval, membrane-bound vacuolar structures having diameter of 25 to 500 μm.
- They often remain isolated in the cytoplasm and occur in most cells but especially abundant in the SER.

3. Tubules:

- The tubules are branched structures forming the reticular system along with the cisternae and vesicles. They usually have the diameter from 50 to 190 μm and occur almost in all the cells.
- Tubular form of ER is often found in SER and is dynamic in nature, *i.e.*, it is associated with membrane movements, fission and fusion between membranes of cytocavity network.

Types of endoplasmic reticulum:

Smooth Endoplasmic Reticulum:

- ER with no embossed ribosomes makes it smooth in appearance.
- The adipose tissues, brown fat cells and adrenocortical cells, interstitial cells of testes and cells of corpus luteum of ovaries, sebaceous cells and retinal pigment cells contain only smooth endoplasmic reticulum (SER).
- The synthesis of fatty acids and phospholipids takes place in the smooth ER.
- It is abundant in hepatocytes. Enzymes in the smooth ER of the liver modify or detoxify hydrophobic chemicals such as pesticides and carcinogens by chemically converting them into more water-soluble, conjugated products that can be excreted from the body.
- High doses of such compounds result in a large proliferation of the smooth ER in liver cells.

Rough Endoplasmic Reticulum:

- Ribosomes bound to the endoplasmic reticulum make it appear rough.
- The rough ER synthesizes certain membrane and organelle proteins and virtually all proteins to be secreted from the cell.
- A ribosome that fabricates such a protein is bound to the rough ER by the nascent polypeptide chain of the protein.
- As the growing polypeptide emerges from the ribosome, it passes through the rough ER membrane, with the help of specific proteins in the membrane.
- Newly made membrane proteins remain associated with the rough ER membrane, and proteins to be secreted accumulate in the lumen of the organelle.
- All eukaryotic cells contain a discernible amount of rough ER because it is needed for the synthesis of plasma membrane proteins and proteins of the extracellular matrix.
- Rough ER is particularly abundant in specialized cells that produce an abundance of specific proteins to be secreted.

- The cells of those organs which are actively engaged in the synthesis of proteins such as acinar cells of pancreas, plasma cells, goblet cells and cells of some endocrine glands are found to contain rough endoplasmic reticulum (RER) which is highly developed.

Rough endoplasmic reticulum and protein secretion:

- The defined pathway taken by secreted protein is: Rough ER - Golgi - secretory vesicles- cell exterior.
- In mammalian cells most proteins are transferred into the ER while they are being translated on membrane bound ribosomes.
- Proteins that are destined for secretion are then targeted to the endoplasmic reticulum by a signal sequence (short stretch of hydrophobic amino acid residues) at the amino terminus of the growing polypeptide chain.
- The signal sequence is K/HDEL which is Lys/His-Asp-Glu-Leu.
- This signal peptide is recognized by a signal recognition particle consisting of six polypeptides and srpRNA. The SRP binds the ribosome as well as the signal sequence, inhibiting further translation and targeting the entire complex (the SRP, ribosome, and growing polypeptide chain) to the rough ER by binding to the SRP receptor on the ER membrane.
- Binding to the receptor releases the SRP from both the ribosome and the signal sequence of the growing polypeptide chain.

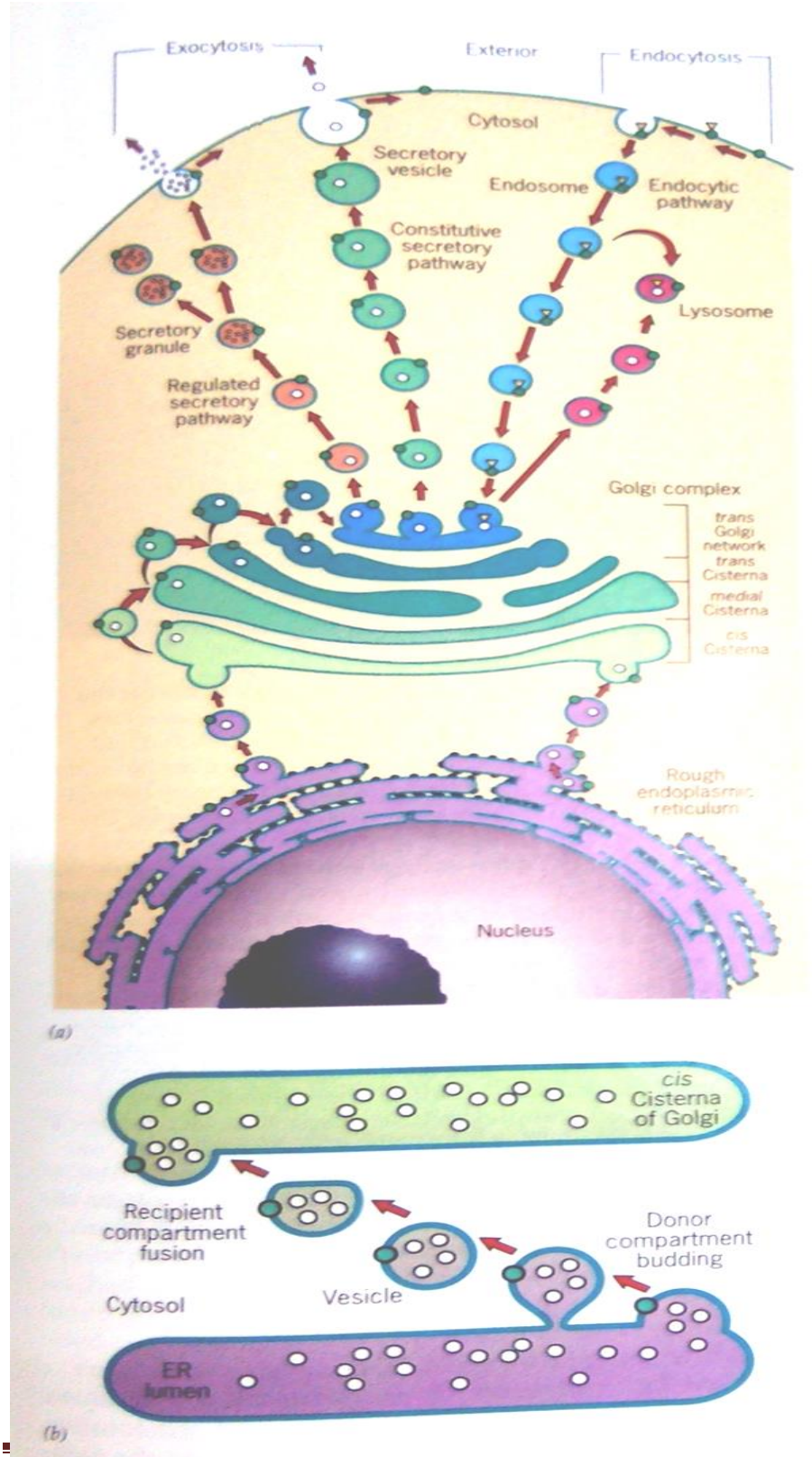


Fig :*Biosynthetic and endocytic pathways unite the Endomembranes into a dynamic, interconnected framework.*

(a) The biosythetic pathways descibes the flow of materials (especially proteins) from the ER through the Golgi complex, and out to various locations including lysosomes, endosomes, secretory vesicles, vacuoles and plasma membrane.

(b) Illustrates the process of vesicle transport by wich materials are transported from a donar compartment to recipient compartment . vesicle form by membrane budding during which membrane proteins of the donor membrane can be incorporated into the vesicle membrane and soluble proteins in the donar compartment can be enclosed in the lumen of the vesicles. When the transport vesicle subsequently fuses, the proteins of the vesicle membrane become the part of the recipient membrane and the soluble proteins become sequestered within

- The ribosome then binds to a protein translocation complex in the ER membrane, and the signal sequence is inserted into a membrane channel or translocon with the aid of GTP.
- Transfer of the ribosome mRNA complex from the SRP to the translocon opens the gate on the translocon and allows translation to resume, and the growing polypeptide chain is transferred directly into the translocon channel and across the ER membrane as translation proceeds.
- As translocation proceeds, the signal sequence is cleaved by signal peptidase and the polypeptide is released into the lumen of the ER.

Smooth endoplasmic reticulum and lipid synthesis:

- Hydrophobic lipids are synthesized in the ER and then they are then transported from the ER to their ultimate destinations either in vesicles or by carrier proteins.
- Phospholipids are synthesized in the cytosolic side of the ER membrane from water-soluble cytosolic precursors.
- Other lipids that are synthesized in the ER are cholesterol and ceramide which is further converted to either glycolipids or sphingomyelin in the golgi apparatus.
- Smooth ER are also the site for the synthesis of the steroid hormones from cholesterol.
- Thus steroid producing cells in the testis and ovaries are abundant in smooth ER.

Common functions of SER and RER:

1. The endoplasmic reticulum provides an ultrastructural skeletal framework to the cell andgivesmechanical support to the colloidal cytoplasmic martix.
2. The exchange of molecules by the process of osmosis, diffusion and active transport occurs through the membranes of endoplasmic reticulum. The ER membrane has permeases and carriers.
3. The endoplasmic membranes contain many enzymes which perform various synthetic andmetabolic activities and provides increased surface for various enzymatic reactions.
4. The endoplasmic reticulum acts as an intracellular circulatory or transporting system. Various secretory products of granular endoplasmic reticulum are transported to various

organelles as follows: Granular ER− agranular ER − Golgi membrane−lysosomes, transport vesicles or secretory granules. Membrane flow may also be an important mechanism for carrying particles, molecules and ions into and out of the cells. Export of RNA and nucleoproteins from nucleus to cytoplasm may also occur by this type of flow.

5. The ER membranes are found to conduct intra-cellular impulses. For example, the sarcoplasmic reticulum transmits impulses from the surface membrane into the deep region of the muscle fibres.
6. The sarcoplasmic reticulum plays a role in releasing calcium when the muscle is stimulated and actively transporting calcium back into the sarcoplasmic reticulum when the stimulation stops and the muscle must be relaxed.

The Golgi Complex:

Processes and Sorts Secreted and Membrane Proteins :

The golgi complex was discovered by Camillo Golgi during an investigation of the nervous system and he named it the "internal reticular apparatus".

Functionally it is also known as the post office of the cell.

Certain important cellular functions such as biosynthesis of polysaccharides, packaging (compartmentalizing) of cellular synthetic products (proteins), production of exocytotic (secretory) vesicles and differentiation of cellular membranes, occurs in the Golgi complex or Golgi apparatus located in the cytoplasm of animal and plant cells.

Occurrence:

- The Golgi apparatus occurs in all eukaryotic cells.
- The exceptions are the prokaryotic cells (mycoplasmas, bacteria and blue green algae) and eukaryotic cells of certain fungi, sperm cells of bryophytes and pteridiophytes, cells of mature sieve tubes of plants and mature sperm and red blood cells of animals.
- Their number per plant cell can vary from several hundred as in tissues of corn root and algal rhizoids (*i.e.*, more than 25,000 in algal rhizoids, Sievers,1965), to a single organelle in some algae.

- In higher plants, Golgi apparatuses are particularly common in secretory cells and in young rapidly growing cells. In animal cells, there usually occurs a single Golgi apparatus, however, its number may vary from animal to animal and from cell to cell.
- *Paramoeba* species has two golgi apparatuses and nerve cells, liver cells and chordate oocytes have multiple golgi apparatuses, there being about 50 of them in the liver cells.

Morphology :

- The Golgi apparatus is morphologically very similar in both plant and animal cells.
- However, it is extremely pleomorphic: in some cell types it appears compact and limited, in others spread out and reticular (net-like). Its shape and form may vary depending on cell type.
- It appears as a complex array of interconnecting tubules, vesicles and cisternae.
- There has been much debate concerning the terminology of the Golgi's parts.
- The simplest unit of the Golgi apparatus is the cisterna.
- This is a membrane bound space in which various materials and secretions may accumulate.
- Numerous cisternae are associated with each other and appear in a stack-like (lamellar) aggregation.
- A group of these cisternae is called the dictyosome, and a group of dictyosomes makes up the cell's Golgi apparatus.

All dictyosomes of a cell have a common function. The detailed structure of three basic components of the Golgi apparatus are as follows:

1. **Flattened Sac or Cisternae :**

- Cisternae of the golgi apparatus are about 1 μm in diameter, flattened, plate-like or saucer-like closed compartments which are held in parallel bundles or stacks one above the other.
- In each stack, cisternae are separated by a space of 20 to 30 nm which may contain rod-like elements or fibres.
- Each stack of cisternae forms a dictyosome which may contain 5 to 6 Golgi cisternae in animal cells or 20 or more cisternae in plant cells.

- Each cisterna is bounded by a smooth unit membrane (7.5 nm thick), having a lumen varying in width from about 500 to 1000 nm.
- The margins of each cisterna are gently curved so that the entire dictyosome of Golgi apparatus takes on a bow-like appearance.

- The cisternae at the convex end of the dictyosome comprise proximal, forming or cis-face and the cisternae at the concave end of the dictyosome comprise the distal, maturing or trans-face.
- The forming or cis face of Golgi is located next to either the nucleus or a specialized portion of rough ER that lacks bound ribosomes and is called "transitional" ER.
- Trans face of Golgi is located near the plasma membrane.
- This polarization is called cis-trans axis of the Golgi apparatus.

2. Tubules

A complex array of associated vesicles and tubules (30 to 50 nm diameter) surround the dictyosome and radiate from it. The peripheral area of dictyosome is fenestrated or lace-like in structure.

3. Vesicles

The vesicles are 60 nm in diameter and are of three types :

(i) Transitional vesicles are small membrane limited vesicles which are form as blebs from the transitional ER to migrate and converge to cis face of Golgi, where they coalasce to form new cisternae.

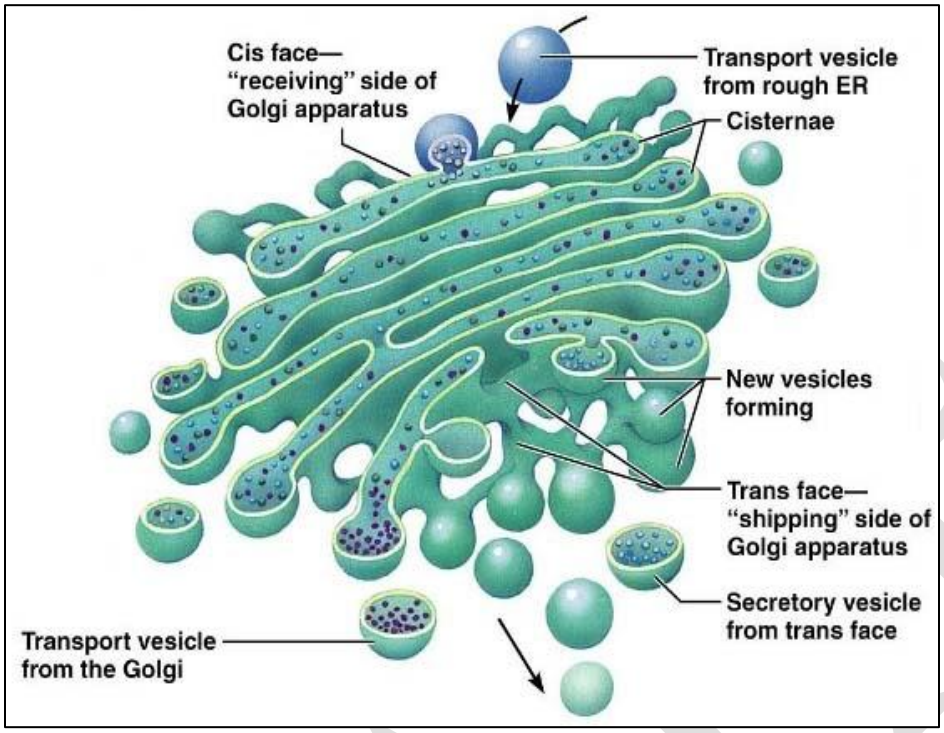


Fig :The Golgi complex.

(ii) Secretory vesicles are varied-sized membrane-limited vesicles which discharge from margins of cisternae of Golgi. They, often, occur between the maturing face of Golgi and the plasma membrane.

(iii) Clathrin-coated vesicles are spherical protuberances, about 50 μm in diameter and with a rough surface. They are found at the periphery of the organelle, usually at the ends of single tubules, and are morphologically quite distinct from the secretory vesicles. Theclathrin-coated vesicles are known to play a role in intra-cellular traffic of membranes and of secretory products.

Origin or biogenesis :

- Origin of Golgi apparatus involves the formation of new cisternae and there is great variation in
- shape, number and size of cisternae in each stack (dictyosome).
- *The process of formation of newcisternae may be performed by any of the following methods:*

Individual stacks of cisternae may arisefrom the pre-existing stacks by division or fragmentation.

The alternative method of origin of Golgiis based on *de novo* formation.

- In fact, various cytological and biochemical evidences have establishedthat the membranes of the Golgi apparatus are originated from the membranes of the smooth ER whichin turn have originated from the rough ER.
- The proximal Golgi saccules are formed by fusion of ERderivedvesicles, while distal saccules "give their all" to vesicle formation and disappear.
- Thus, Golgisaccules are constantly and rapidly renewed.
- The cells of dormant seeds of higher plants generally lack Golgi apparatuses but they do display zone of exclusion having aggregation of small transition vesicles.
- Photomicrographs of cells in earlystages of germination suggest progressive development of Golgi bodies in these zones of exclusion;and the development of Golgi apparatuses coincides with the disappearance of the aggregation of vesicles.

Functions:

1. Modifying, sorting, and packaging of macromolecules for cell secretion:

- The golgi complex is involved in the transport of lipids around the cell, and the creation of lysosomes.
- Proteins are modified by enzymes in cisternae by glycosylation and phosphorylation by identifying the signal sequence of the protein in question.
- For example, the Golgi apparatus adds a mannose-6-phosphate label to proteins destined for lysosomes.
- One molecule that is phosphorylated in the Golgi is Apolipoprotein, which forms a molecule known as VLDL that is a constituent of blood serum.
- The phosphorylation of these molecules is important to help aid in their sorting for secretion into the blood serum.

2. Proteoglycans and carbohydrate synthesis:

This includes the production of glycosaminoglycans (GAGs), long unbranched polysaccharides which the Golgi then attaches to a protein synthesised in the endoplasmic reticulum to form proteoglycans.

1. Golgi Functions in Animals:

- ***In animals, Golgi apparatus is involved in the packaging and exocytosis of the following:***
  1. Zymogen of exocrine pancreatic cells; Mucus (a glycoprotein) secretion by goblet cells of intestine;
  2. Lactoprotein (casein) secretion by mammary gland cells (Merocrine secretion);
  3. Secretion of compounds (thyroglobulins) of thyroxine hormone by thyroid cells;
  4. Secretion of tropocollagen and collagen;
  5. Formation of melanin granules and other pigments;
  6. and Formation of yolk and vitelline membrane of growing primary oocytes.
- It is also involved in the formation of certain cellular organelles such as plasma membrane, lysosomes, acrosome of spermatozoa and cortical granules of a variety of oocytes.

2. Golgi Functions in Plants:

- In plants, Golgi apparatus is mainly involved in the secretion of materials of primary and secondary cell walls (formation and export of glycoproteins, lipids, pectins and monomers for hemicellulose, cellulose, lignin).
- During cytokinesis of mitosis or meiosis, the vesicles originating from the periphery of Golgi apparatus, coalesce in the phragmoplast area to form a semisolid layer, called cell plate.
- The unit membrane of Golgi vesicles fuses during cell plate formation and becomes part of plasma membrane of daughter.

Possible Questions

2 Marks

1. Define microtubules.
2. Define microfilaments.
3. What is cisternae.
4. Write about cisterna.
5. What is protein segregation?

8 Marks

1. Write about the structure and functions of Microtubules
2. Brief on Microfilaments
3. Write a short note on Intermediate Filaments
4. Explain in detail structure and functions of endoplasmic reticulum
5. Describe in detail golgi complex
6. Explain ribosome's
7. Write about the segregation

Unit IV

Cell Organelles: Lysosomes, Vacuoles and Micro bodies: Structure and functions Ribosomes: Structure and function including role in protein synthesis: Mitochondria: Structure and function, Genomes, Biogenesis. Chloroplasts: Structure and function: Genome, Biogenesis. Nucleus – structure and function. Chromosomes – structure and function.

Lysosomes

- Lysosomes is an organelle which provides an excellent example of the ability of intracellular membranes to form closed compartments in which the composition of the lumen (the aqueous interior of the compartment) differs substantially from that of the surrounding cytosol.
- Found exclusively in animal cells, lysosomes are responsible for degrading certain components that have become obsolete for the cell or organism.
- Lysosomes are often budded from the membrane of the Golgi apparatus, but in some cases they develop gradually from late endosomes, which are vesicles that carry materials brought into the cell by a process known as endocytosis.
- The biogenesis of the lysosomes requires the synthesis of specialized lysosomal hydrolases and membrane proteins.
- Both classes of proteins are synthesized in the ER and transported through the Golgi apparatus, then transported from the trans Golgi network to an intermediate compartment (an endolysosome) by means of transport vesicles (which are coated by clathrin protein).

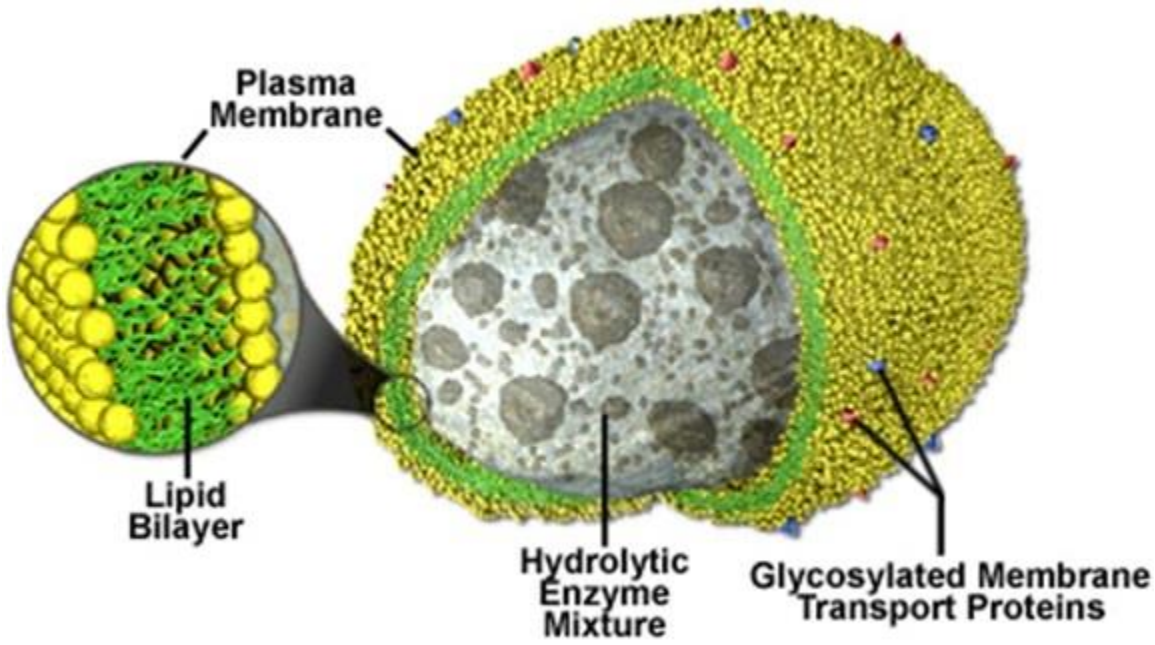


Fig :*Anatomy of the Lysosome.*

Occurrence:

- The lysosomes occur in most animal and few plant cells.
- They are absent in bacteria and mature mammalian erythrocytes.
- Few lysosomes occur in muscle cells or in acinar cells of the pancreas.
- Leucocytes, especially granulocytes are a particularly rich source of lysosomes.
- Their lysosomes are so large-sized that they can be observed under the light microscope.
- They are also numerous in epithelial cells of absorptive, secretory and excretory organs (intestine, liver, and kidney).
- They occur in abundance in the epithelial cells of lungs and uterus.
- Phagocytic cells and cells of reticuloendothelial system (bone marrow, spleen and liver) are also rich in lysosomes.

Structure:

- The lysosomes are round vacuolar structures bounded by single unit membrane. Their shape and density vary greatly. Lysosomes are 0.2 to 0.5μm in size.

- Since, size and shape of lysosomes vary from cell to cell and time to time (they are polymorphic), their identification becomes difficult.

Isolation and chemical composition:

- Lysosomes are very delicate and fragile organelles.
- Lysosomal fractions have been isolated by sucrose-density centrifugation (Isopycnic centrifugation) after mild methods of homogenization.
- The location of the lysosomes in the cell can also be pinpointed by various histochemical or cytochemical methods. For example, lysosomes give a positive test for acid Schiff reaction.
- Certain lysosomal enzymes are good histochemical markers. For example, acid phosphatase is the principal enzyme which is used as a marker for the lysosomes by the use of Gomori'staining technique. Specific stains are also used for other lysosomal enzymes such as B- glucuronidase, aryl sulphatatase, N-acetyl-B-glucosaminidase and 5-bromo-4-chloroindolacetate esterase.
- A lysosome may contain up to 40 types of hydrolytic enzymes.
- They include proteases (cathepsin for protein digestion), nucleases, glycosidases (for digestion of polysaccharides and glycosides), lipases, phospholipases, phosphatases and sulphatases.
- All lysosomal enzymes are acid hydrolases, optimally active at the pH5. The membrane of the lysosome normally keeps the enzymes latent and out of the cytoplasmic matrix or cytosol (pH is ~7.2), but the acid dependency of lysosomal enzymes protects the contents of the cytosol (cytoplasmic matrix) against any damage even if leakage of lysosomal enzymes occur.
- The latency of the lysosomal enzymes is due to the presence of the membrane which is resistant to the enzymes that it encloses.
- Most probably this is due to the fact that most lysosomal hydrolases are membrane-bound, which may prevent the active centres of enzymes to gain access to
- susceptible groups in the membrane.

Lysosomal Membrane:

- The lysosomal membrane is slightly thicker than that of mitochondria.

- It contains substantial amounts of carbohydrate material, particularly sialic acid.
- In fact, most lysosomal membrane proteins are unusually highly glycosylated, which may help protect them from the lysosomal proteases in the lumen.
- The lysosomal membrane has another unique property of fusing with other membranes of the cell.
- This property of fusion has been attributed to the high proportion of membrane lipids present in the micellar configuration.
- Surface active agents such as liposoluble vitamins (A,K,D and E) and steroid sex hormones have a destabilizing influence, causing release of lysosomal enzymes due to rupture of lysosomal membranes. Drugs like cortisone, hydrocortisone and others tend to stabilize the lysosomal membrane and have an anti-inflammatory effect on the tissue.
- The entire process of digestion is carried out within the lysosome.
- Most lysosomal enzymes act in an acid medium.
- Acidification of lysosomal contents depends on an ATP-dependent proton pump which is present in the membrane of the lysosome and accumulates H+ inside the organelle.
- Lysosomal membrane also contains transport proteins that allow the final products of digestion of macromolecules to escape so that they can be either excreted or reutilized by the cell.

Functions:

1. Lysosomes serve as digestion compartments for cellular materials that have exceeded their lifetime or are otherwise no longer useful by autophagy. When a cell dies, the lysosome membrane ruptures and enzymes are liberated. These enzymes digest the dead cells. In the process of metamorphosis of amphibians and tunicates many embryonic tissues,

e.g., gills, fins, tail, etc., are digested by the lysosomes and utilized by the other cells.

2. Lysosomes break down cellular waste products, fats, carbohydrates, proteins, and other macromolecules into simple compounds, which are then transferred back into the cytoplasm as new cell-building materials. To accomplish the tasks associated with digestion, the lysosomes

utilize about 40 different types of hydrolytic enzymes, all of which are manufactured in the endoplasmic reticulum and modified in the Golgi apparatus.

3. Digestion of large extracellular particles: The lysosomes digest the food contents of the phagosomes or pinosomes. The lysosomes of leucocytes enable the latter to devour the foreign proteins, bacteria and viruses.

4. Extracellular digestion: The lysosomes of certain cells such as sperms discharge their enzymes outside the cell during the process of fertilization. The lysosomal enzymes digest the limiting membranes of the ovum and form penetra path in ovum for the sperms. Acid hydrolases are released from osteoclasts and break down bone for the reabsorption; these cells also secrete lactic acid which makes the local pH enough for optimal enzyme activity. Likewise, preceding ossification (bone formation), fibroblasts release cathepsin D enzyme to break down the connective tissue.

Ribosomes

- Ribosomes are the protein synthesis units of a cell described by G.E. Palade in 1952. They are complex of ribosomal RNA and various proteins.
- Ribosomes are small, dense, rounded and granular particles of the ribo-nucleoprotein.
- They occur either freely in the matrix of mitochondria, chloroplast and cytoplasm or remain attached with the membranes of the endoplasmic reticulum.
- They occur in most prokaryotic and eukaryotic cells and provide a scaffold for the ordered interaction of all the molecules involved in protein synthesis.
- They are the most abundant RNA-protein complex in the cell, which directs elongation of a polypeptide.

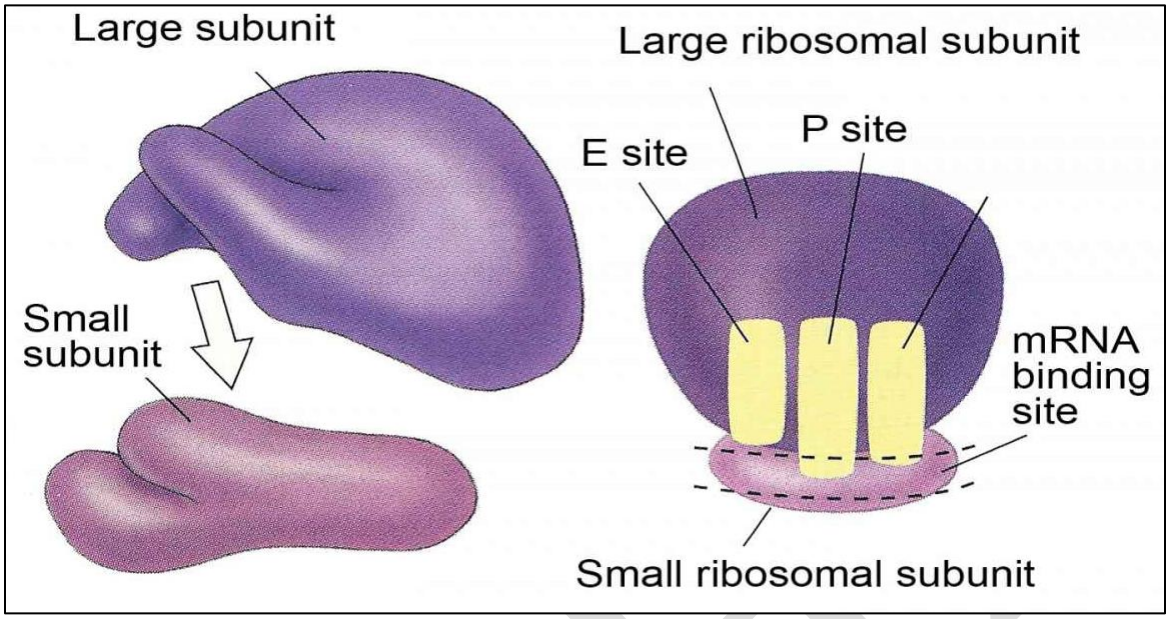


Fig :*Structure of Ribosome.*

Occurrence and distribution:

- The ribosomes occur in both prokaryotic and eukaryotic cells. In prokaryotic cells the ribosomes often occur freely in the cytoplasm or sometimes as polyribosome.
- In eukaryotic cells the ribosomes either occur freely in the cytoplasm or remain attached to the outer surface of the membrane of endoplasmic reticulum.
- The yeast cells, reticulocytes or lymphocytes, meristamatic plant tissues, embryonic nerve cells and cancerous cells contain large number of ribosomes which often occur freely in the cytoplasmic matrix.
- Cells like the erythroblasts, developing muscle cells, skin and hair which synthesize specific proteins for the intracellular utilization and storage also contain large number of free ribosomes.
- In cells with active protein synthesis, the ribosomes remain attached with the membranes of the endoplasmic reticulum.

- ***Examples:***are the pancreatic cells, plasma cells, hepatic parenchymal cells, Nissls bodies, osteoblasts, serous cells or the submaxillary gland, thyroid cells and mammary gland cells.

Types of ribosomes:

- Ribosomes are classified into two types based on their sedimentation coefficient, 70S and 80S.
- S stands for *Svedberg unit* and related to sedimentation rate (sedimentation depends on mass and size). Thus, the value before S indicates size of ribosome.
- ***70S Ribosomes :***Prokaryotes have 70S ribosomes. The 70S ribosomes are comparatively smaller in size and have sedimentation coefficient 70S with molecular weight 2.7×10^6daltons.
- ***80S Ribosomes:***Eukaryotes have 80S ribosomes. The 80S ribosomes have sedimentation coefficient of 80S and molecular weight 40×10^6daltons.

Number of ribosomes:

- An *E. coli* cell contains 10,000 ribosomes, forming 25 per cent of the total mass of the bacterialcell.
- Whereas, mammalian cultured cells contain 10 million ribosomes per cell.

Chemical composition:

- The ribosomes are chemically composed of RNA and proteins as their major constituents; both occurring approximately in equal proportions in smaller as well as larger subunit.
- The 70S ribosomes contain more RNA (60 to 40%) than the proteins (36 to 37%). The ribosomes of *E. coli* contain 63% rRNA and 37% protein.
- While the 80S ribosomes contain less RNA (40 to 44%) than the proteins (60 to 56%), yeast ribosomes have 40 to 44% RNA and 60 to 56% proteins; ribosomes of pea seedling contain 40% RNA and 60% proteins. There is no lipid content in ribosomes.

Ribosomal Proteins:

- A ribosome is composed of three (in bacteria) or four (in eukaryotes) different rRNA molecules and as many as 83 proteins, organized into a large subunit and a small subunit.
- When both 50S and 30S ribosomal subunits are dissociated by centrifuging both of them in a gradient of 5 M cesium chloride, then there are two inactive core particles (40S and 23S,

respectively) which contain the RNA and some proteins called core proteins (CP) at the same time several other proteins—the so-called split proteins (SP) are released from each particle.

- There are SP50 and SP30 proteins which may reconstitute the functional ribosomal subunit when added to their corresponding core.
- Some of the split proteins are apparently specific for each ribosomal subunit.
- The split proteins have been further fractionated and divided into acidic (A) and basic (B) proteins.
- According to Nomura (1968, 1973) and Garett and Wittmann (1973) each 70S ribosome of *E. coli* is composed of about 55 ribosomal proteins.
- Out of these 55 proteins, about 21 different molecules have been isolated from the 30S ribosomal subunit, and some 32 to 34 proteins from the 50S ribosomal subunit.
- Similar organization of ribosomal proteins and RNA is found in 80S Ribosomes. Different rRNA molecules evidently play a central role in the catalytic activities of ribosomes in the process of protein synthesis.

Metallic Ions:

The most important low molecular weight components of ribosomes are the ***divalent metallic ions*** such as Mg++, Ca++ and Mn++.

Structure:

- The ribosomes are oblate spheroid structures of 150 to 250A^{o} in diameter.
- Each ribosome is porous, hydrated and composed of two subunits.
- One ribosomal subunit is large in size and has a domelike shape, while the other ribosomal subunit is smaller in size, occurring above the larger subunit and forming a cap-like structure.
- The small ribosomal subunit contains a single rRNA molecule, referred to as small rRNA. The large subunit contains a molecule of large rRNA and one molecule of 5S rRNA, plus an additional molecule of 5.8S rRNA in vertebrates.
- The lengths of the rRNA molecules, the quantity of proteins in each subunit, and consequently the sizes of the subunits differ in bacterial and eukaryotic cells.

- The assembled ribosome is 70S in bacteria and 80S in vertebrates.
- Both the subunits remain separated by a narrow cleft.
- The two ribosomal subunits remain united with each other due to high concentration of the Mg++ (.001M) ions.
- When the concentration of Mg++ ions reduces in the matrix, both ribosomal subunits get separated.
- Actually in bacterial cells the two subunits are found to occur freely in the cytoplasm and they unite only during the process of protein synthesis.
- At high concentration of Mg++ ions in the matrix, the two ribosomes (monosomes) become associated with each other and known as the dimer.
- Further, during protein synthesis many ribosomes are aggregated due to common messenger RNA and form the polyribosomes or polysomes.

Mitochondria

Structure and Function:

- The mitochondria were first observed by Kolliker in 1850 as granular structures in the striated muscles.
- Mitochondria are called the 'powerhouse of the cell'.
- They are intracellular organelles found in almost all eukaryotic cells having bilayered membranes.

• Most eukaryotic cells contain many mitochondria, which occupy up to 25 percent of the volume of the cytoplasm.

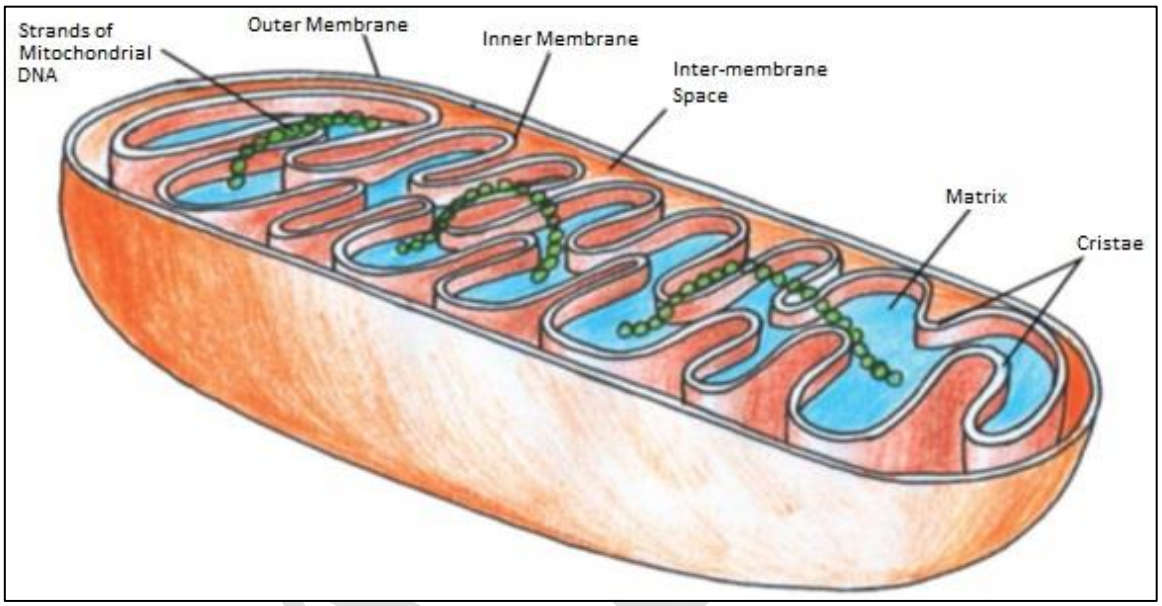


Fig : *Structure of Mitochondrion.*

• These crucial organelles, the main sites of ATP production during aerobic metabolism, are generally exceeded in size only by the nucleus, vacuoles, and chloroplasts.

• They are responsible for aerobic metabolism through oxidative phosphorylation, which leads to energy production in the form of adenosine triphosphate (ATP).

• Mitochondria contain a number of enzymes and proteins that help in processing carbohydrates and fats obtained from food we eat to release energy.

• Each human cell contains on average hundreds to thousands of mitochondria.

• The exception is mature red blood cells, which rely exclusively on anaerobic metabolism and contain no mitochondria.

Localisation:

- Mitochondria are present in all eukaryotic cells. They move autonomously in the cytoplasm, so they generally have uniform distribution in the cytoplasm, but in many cells their distribution is restricted.
- The distribution and number of mitochondria can be correlated with type of function the cell performs.
- Typically mitochondria with many cristae are associated with mechanical and osmotic work situations, where there are sustained demands for ATP *e.g.*, between muscle fibres, in the basal infolding of kidney tubule cells, and in a portion of inner segment of rod and cone cells of retina.
- Myocardial muscle cells have numerous large mitochondria called sarcosomes that reflect the great amount of work done by these cells.
- Mitochondria are particularly numerous in regions where ATP-driven osmotic work occurs, *e.g.*, brush border of kidney proximal tubules, the infolding of the plasma membrane of dogfish salt glands and Malpighian tubules of insects, the contractile vacuoles of some protozoans as Paramecium.
- Non-myelinated axons contain many mitochondria that are poor ATP factories, since each has only single cristae.
- In this case, there is a great requirement for monoamine oxidase, an enzyme present in outer mitochondrial membrane that oxidatively deaminates monoamines including neurotransmitters (acetylcholine).

Orientation:

- The mitochondria have definite orientation. For example, in cylindrical cells the mitochondria
- usually remain orientated in basal apical direction and lie parallel to the main axis.
- In leucocytes, the mitochondria remain arranged radially with respect to the centrioles.

- As they move about in the mitochondria form long moving filaments or chains, while in others they remain fixed in one position where they provide ATP directly to a site of high ATP utilization,
- ***e.g.,*** they are packed between adjacent myofibrils in a cardiac muscle cell or wrapped tightly around the flagellum of sperm.

Structure:

- Each mitochondrion is bound by two highly specialized membranes that play a crucial role in its activities. Each of the mitochondrial membrane is 6 nm in thickness and fluidmosaic in ultrastructure.
- The membranes are made up of phospholipids and proteins. The space in between the two membranes is called the inter-membrane space which has the same composition as the cytoplasm of the cell.
- Inner and the outer membrane is separated by a 6–8 nm wide space.

Outer Membrane

- The two membranes that bound a mitochondrion differ in composition and function. The outer membrane, composed of about half lipid and half protein, contains porins that render the membrane permeable to molecules having molecular weights as high as 10,000 dalton.
- In this respect, the outer membrane of mitochondria is similar to the outer membrane of gram-negative bacteria.
- The outer membrane is smooth unlike the inner membrane and has almost the same amount of phospholipids as proteins.
- It has a large number of special proteins called porins that allow molecules of 5000 daltons or less in weight to pass through it.
- It is completely permeable to nutrient molecules, ions, ATP and ADP molecules.

Inner Membrane

- The inner membrane is much less permeable, than the outer membrane. It has about 20 percent lipid and 80 percent protein.

- The surface area of the inner membrane is greatly increased by a large number of infoldings, or finger like projections called cristae, that protrude into the matrix, or central space, increasing the surface area for the complexes.
- It contains the complexes of the electron transport chain and the ATP synthetase complex, they also serve to separate the matrix from the space that will contain the hydrogen ions, allowing the gradient needed to drive the pump.
- It is permeable only to oxygen, carbon dioxide and water and is made up of a large number of proteins that play an important role in producing ATP, and also helps in regulating transfer of metabolites across the membrane.
- In general, the cristae of plant mitochondria are tubular, while those of animal mitochondria are lamellar or plate-like. Some mitochondria, particularly those from heart, kidney and skeletal muscles have more extensive cristae arrangements than liver mitochondria.
- In comparison to these, other mitochondria (from fibroblasts, nerve axons and most plant tissues) have relatively few cristae.
- Attached to matrix face of inner mitochondrial membrane are repeated units of stalked particles, called elementary particles, inner membrane subunits or oxysomes.

Matrix

The matrix is a complex mixture of enzymes that are important for the synthesis of ATP molecules, special mitochondrial ribosomes, tRNAs and the mitochondrial DNA. Besides these, it has oxygen, carbon dioxide and other recyclable intermediates.

Function of mitochondria

1.The most important function of the mitochondria is to produce energy. The food that we eat is broken into simpler molecules like carbohydrates, fats, etc., in our bodies. These are sent to the mitochondrion where they are further processed to produce charged molecules that combine with oxygen and produce ATP molecules. This entire process is known as oxidative phosphorylation.

2.It is important to maintain proper concentration of calcium ions within the various compartments of the cell. Mitochondria help the cells to achieve this goal by serving as storage tanks of calcium ions.

3.Mitochondria help in the building of certain parts of the blood, and hormones like testosterone and estrogen.

4.Mitochondria in the liver cells have enzymes that detoxify ammonia.

Although most of the genetic material of a cell is contained within the nucleus, the mitochondria have their own DNA. They have their own machinery for protein synthesis and reproduce by the process of fission like bacteria do. Due to their independence from the nuclear DNA and similarities with bacteria, it is believed that mitochondria have originated from bacteria by endosymbiosis.

Chloroplasts

The chloroplast (*chlor*=green; *plast*=living) is most widely occurring chromoplast of the plants. It occurs mostly in the green algae and higher plants. The chloroplast contains the pigment chlorophyll 'a' and chlorophyll 'b' and DNA and RNA.

Chloroplasts were described as early as seventeenth century by Nehemiah Grew and Antonie van Leeuwenhoek.

Distribution:

The chloroplasts remain distributed homogeneously in the cytoplasm of plant cells. But in certaincells, the chloroplasts become concentrated around the nucleus or just beneath the plasma membrane.

The chloroplasts have a definite orientation in the cell cytoplasm. Chloroplasts are motile organelles, and show passive and active movements.

Morphology:

Shape:Higher plant chloroplasts are generally biconvex or plano-convex. However, in different plant cells, chloroplasts may have various shapes, *viz*., filamentous, saucer-shaped, spheroid, ovoid, discoid or club-shaped. They are vesicular and have a colourless centre.

Size:The size of the chloroplasts varies from species to species. They generally measure 2–3μm in thickness and 5–10μm in diameter (*Chlamydomonas*). The chloroplasts of polyploid plant cells are comparatively larger than those of the diploid counterparts. Generally, chloroplasts of plants grown in the shade are larger and contain more chlorophyll than those of plants grown in sunlight.

Isolation:

Chloroplasts are routinely isolated from plant tissues by differential centrifugation following the disruption of the cells.

Ultrastructure:

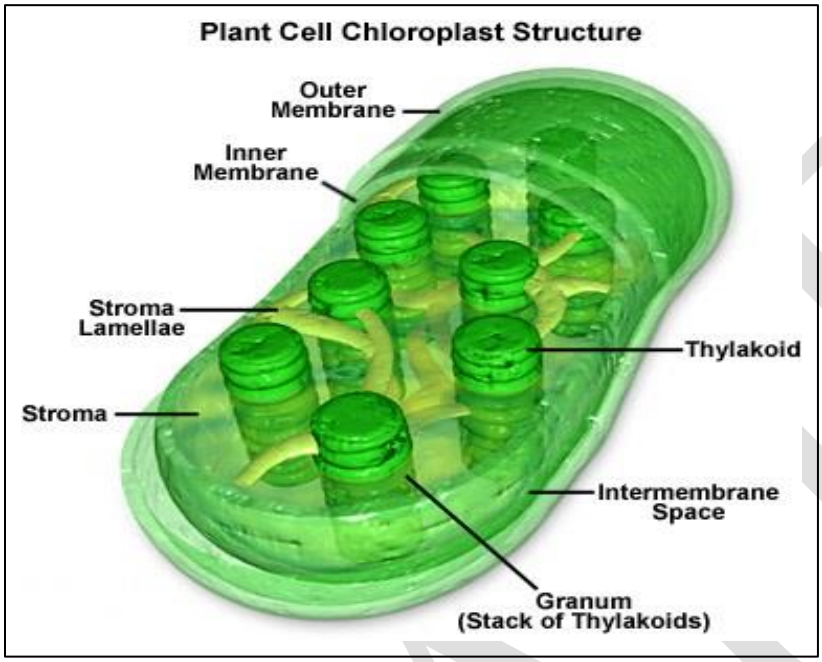
Chloroplast comprises of three main components:

1. Envelope :

- The entire chloroplast is bounded by a double unit membrane. Across this double membrane envelope occurs exchange of molecules between chloroplast and cytosol.
- Isolated membranes of envelope of chloroplast lack chlorophyll pigment and cytochromes but have a yellow colour due to the presence of small amounts of carotenoids. They contain only 1 to 2 per cent of the total protein of the chloroplast.

2. Stroma :

- The matrix or stroma fills most of the volume of the chloroplasts and is a kind of gel-fluid phasethat surrounds the thylakoids (grana).
- It contains about 50 per cent of the proteins of the chloroplast, most of which are soluble type.
- The stroma also contains ribosomes and DNA molecules both of which are involved in the synthesis of some of the structural proteins of the chloroplast.
- The stroma is the place where CO2 fixation occurs and where the synthesis of sugars, starch, fatty acids and some proteins takes place.



3. Thylakoids :

- The thylakoids (thylakoid = sac-like) consists of flattened and closed vesicles arranged as a membranous network.
- The outer surface of the thylakoid is in contact with the stroma, and its inner surface encloses an intrathylakoid space.
- Thylakoids get stacked forming grana. There may be 40 to 80 grana in the matrix of a chloroplast. The number of thylakoids per granum may vary from 1 to 50 or more.
- For example, there may be single thylakoid (red alga), paired thylakoids (Chrysophyta), triple thylakoids and multiple thylakoids (green algae and higher plants).
- Like the mitochondria, the chloroplasts have their own DNA, RNAs and protein synthetic machinery and are semiautonomous in nature.

- Chloroplasts are the largest and the most prominent organelles in the cells of plants and green algae.
- Chloroplasts and mitochondria have other features in common: both often migrate from place to place within cells, and they contain their own DNA, which encodes some of the key organellar proteins.
- Though most of the proteins in each organelle are encoded by nuclear DNA and are synthesized in the cytosol, the proteins encoded by mitochondrial or chloroplast DNA is synthesized on ribosomes within the organelles.
- Chloroplasts have a highly permeable outer membrane; a much less permeable inner membrane, in which membrane transport proteins are embedded; and a narrow intermembrane space in between.

- Together, these membranes form the chloroplast envelope. The inner membrane surrounds a large space called the stroma, and contains many metabolic enzymes.
- The electron-transport chains, photosynthetic light-capturing systems, and ATP synthase are all contained in the thylakoid membrane, a third distinct membrane that forms a set of flattened disclike sacs, the thylakoids**.**
- The lumen of each thylakoid is connected with the lumen of other thylakoids, defining a third internal compartment called the thylakoid space, which is separated by the thylakoid membrane from the stroma that surrounds it.

Photosynthesis

The many reactions that occur during photosynthesis in plants can be grouped into two broad categories:

1.Electron-transfer reactions or the light reactions: In the choloroplast, energy derived from sunlight energizes an electron of chlorophyll, enabling the electron to move along an electron-transport chain in the thylakoid membrane in much the same way that an electron moves along the respiratory chain in mitochondria.

The chlorophyll obtains its electrons from water (H2O), producing O2 as a by-product.

During the electron-transport process, H+ is pumped across the thylakoid membrane, and the resulting electrochemical proton gradient drives the synthesis of ATP in the stroma.

As the final step in this series of reactions, high-energy electrons are loaded onto NADP+, converting it to NADPH. All of these reactions are confined to the chloroplast.

2.Carbon-fixation reactions or the dark reactions wherein the ATP and the NADPH produced by the photosynthetic electron-transfer reactions serve as the source of energy and reducing power, respectively, to drive the conversion of CO2 to carbohydrate.

The carbon-fixation reactions, which begin in the chloroplast stroma and continue in the cytosol, produce sucrose and many other organic molecules in the leaves of the plant.

The sucrose is exported to other tissues as a source of both organic molecules and energy for growth.

Thus, the formation of ATP, NADPH, and O2 and the conversion of CO2 to carbohydrate are separate processes, although elaborate feedback mechanisms interconnect the two. Several of the chloroplast enzymes required for carbon fixation, for example, are inactivated in the dark and reactivated by light-stimulated electron-transport processes.

Nucleus

- Nucleus means kernel and was the first organelle to be discovered. It was discovered and named by Robert Brown in 1833 in the plant cells and is recognized as a constant feature of all animal and plant cells.
- Certain eukaryotic cells such as the mature sieve tubes of higher plants and mammalian erythrocytes contain no nucleus. It is the largest cellular organelle in eukaryotes. Prokaryotic cells lack nucleus and is complemented by nucleoid.

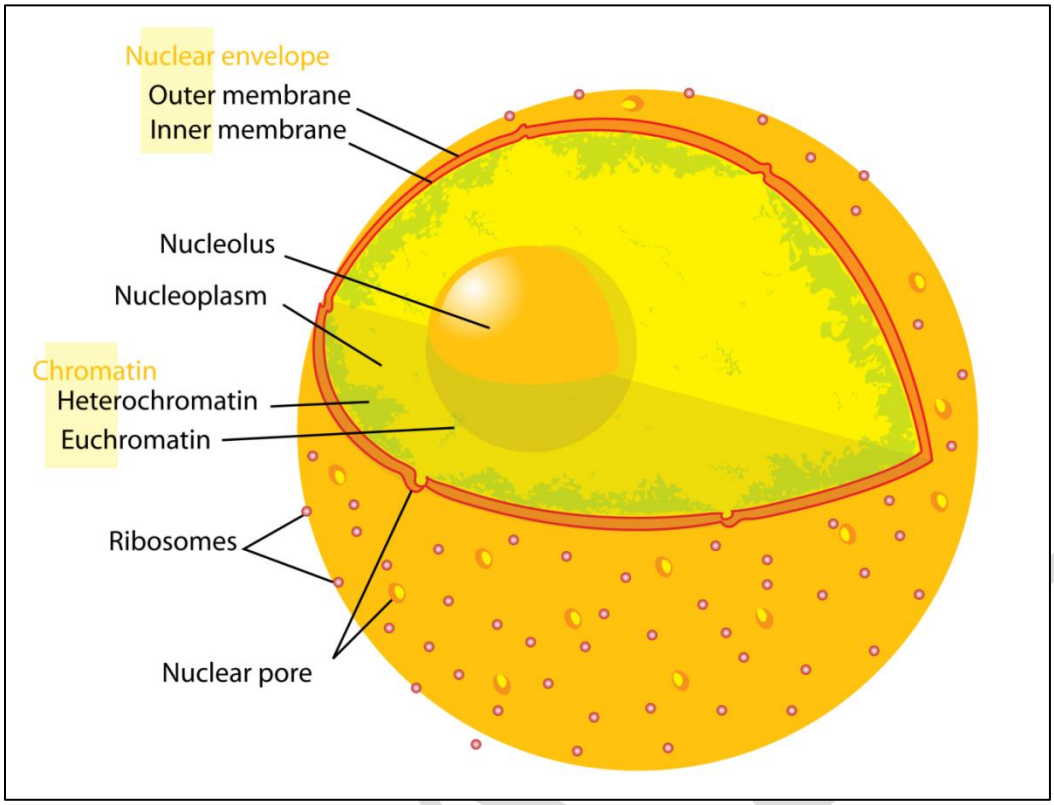


Fig :The schematic representation of nucleus.

- In mammalian cells, the average diameter of the nucleus is approximately 6 micrometers (μm), occupying about 10% of the total cell volume.
- The contents of the nucleus are DNA genome, RNA synthetic apparatus, and a fibrous matrix. It is surrounded by two membranes, each one a phospholipid bilayer containing many different types of proteins.
- The inner nuclear membrane defines the nucleus itself. In most cells, the outer nuclear membrane is continuous with the rough endoplasmic reticulum, and the space between the inner and outer nuclear membranes is continuous with the lumen of the rough endoplasmic reticulum.
- The two nuclear membranes appear to fuse at nuclear pores, the ringlike complexes composed of specific membrane proteins through which material moves between the nucleus and the

cytosol. It contains cell's genetic material, organized as multiple long linear DNA molecules in complex with histones, to form chromosomes.

- The genes within these chromosomes are the cell's nuclear genome. The function is to maintain the integrity of the genes that controls the activities of the cell by regulating gene expression.
- In a growing or differentiating cell, the nucleus is metabolically active, replicating DNA and synthesizing rRNA, tRNA, and mRNA. Within the nucleus mRNA binds to specific proteins, forming ribonucleoprotein particles.
- Most of the cell's ribosomal RNA is synthesized in the nucleolus, a subcompartment of the nucleus that is not bounded by a phospholipid membrane. Some ribosomal proteins are added to ribosomal RNAs within the nucleolus as well.
- The finished or partly finished ribosomal subunits, as well as tRNAs and mRNA-containing particles, pass through a nuclear pore into the cytosol for use in protein synthesis.
- In a nucleus that is not dividing, the chromosomes are dispersed and not dense enough to be observed in the light microscope.
- Only during cell division are individual chromosomes visible by light microscopy. In the electron microscope, the nonnucleolar regions of the nucleus, called the nucleoplasm, can be seen to have dark and light staining areas.
- The dark areas, which are often closely associated with the nuclear membrane, contain condensed concentrated DNA, called heterochromatin.
- Fibrous proteins called lamins form a two-dimensional network along the inner surface of the inner membrane, giving it shape and apparently binding DNA to it. The breakdown of this network occurs early in cell division.

Cell Nucleus: Ultrastructure

The structure of a cell nucleus consists of a nuclear membrane (nuclear envelope), nucleoplasm, nucleolus, and chromosomes. Nucleoplasm, also known as karyoplasm, is the matrix present inside the nucleus.

Nuclear Membrane :

- It is a double-membrane structure each 5–10 nm thick . Numerous pores occur in the envelope, allowing RNA and other chemicals to pass, but not the DNA.
- Because the nuclear membrane is impermeable to most molecules, nuclear pores are required to allow movement of molecules across the envelope.
- These pores cross both of the membranes, providing a channel that allows free movement of small molecules and ions.
- The movement of larger molecules such as protein requires active transport regulated by carrier proteins.
- The nuclear envelope (or perinuclear cisterna) encloses the DNA and defines the nuclear compartment of interphase and prophase nuclei.
- The spherical inner nuclearmembrane contains specific proteins that act as binding sites for the supporting fibrous sheath ofintermediate filaments (IF), called nuclear lamina.
- Nuclear lamina has contact with the chromatin (or chromosomes) and nuclear RNAs.
- The inner nuclear membrane is surrounded by the outer nuclear membrane, which closely resembles the membrane of the endoplasmic reticulum, that is continuous with it.
- Like the membrane of the rough ER, the outer surface of outer nuclear membrane is generally studded with ribosomes engaged in protein synthesis.
- The proteins made on these ribosomes are transported into space between the inner and outer nuclear membrane, called perinuclear space.
- The perinuclear space is a 10 to 50 nm wide fluid-filled compartment which is continuous with the ER lumen and may contain fibres, crystalline deposits, lipid droplets or electrondense material.
- Nuclear pores and nucleocytoplasmic traffic. The nuclear envelope in all eukaryotic forms, from yeasts to humans, is perforated by nuclear pores.

Nucleo-cytoplasmic traffic:

- Quite evidently there is considerable trafficking across the nuclear envelope during interphase. Ions, nucleotides and structural, catalytic and regulatory proteins are imported from the cytosol (cytoplasmic matrix); mRNA, tRNA are exported to the cytosol (cytoplasmic matrix).
- However, one of the main functions of the nuclear envelope is to prevent the entrance of active ribosomes into the nucleus.

Nucleoplasm:

- The space between the nuclear envelope and the nucleolus is filled by a transparent, semi-solid,
- granular and slightly acidophilic ground substance or the matrix known as the nuclear sap or
- nucleoplasm or karyolymph.
- The nuclear components such as the chromatin threads and thenucleolus remain suspended in the nucleoplasm which is composed mainly of nucleoproteinsbut it also contains otherinorganic and organic substances, namely nucleic acids, proteins, enzymes and minerals.
- The most common nucleic acids of the nucleoplasm are the DNA and RNA.
- The nucleoplasm contains many types of complex proteinscategorized into:

(i) Basic proteins. The proteins which take basic stain are known as the basic proteins. The most important basic proteins of the nucleus are nucleoprotamines and the nucleohistones.

(ii) Non-histone or Acidic proteins. The acidic proteins either occur in the nucleoplasm or in the chromatin. The most abundant acidic proteins of the euchromatin (a type of chromatin) are the phosphoproteins. The nucleoplasm contains many enzymes which are necessary for the synthesis of the DNA and RNA. Most of the nuclear enzymes are composed of non-histone (acidic) proteins.

- The most important nuclear enzymes are the DNA polymerase, RNA polymerase, NAD synthetase, nucleoside triphosphatase, adenosine diaminase, nucleoside phosphorylase, guanase, aldolase, enolase, 3-phosphoglyceraldehyde dehydrogenase and pyruvate kinase. The nucleoplasm also contains certain cofactors and coenzymes such as ATP and acetyl CoA. The nucleoplasm has small lipid content.

- The nucleoplasm also contains several inorganic compounds such as phosphorus, potassium, sodium, calcium and magnesium. The chromatin comparatively contains large amount of these minerals than the nucleoplasm.
- The nucleoplasm contains many thread-like, coiled and much elongated structures which take readily the basic stains such as the basic fuchsin. These thread-like structures are known as the chromatin (*chrome*=colour) substance or chromatin fibres. Chromosome will be discussed in detail in the next module.

Nucleolus:

- Most cells contain in their nuclei one or more prominent spherical colloidal acidophilic bodies, called nucleoli. However, cells of bacteria and yeast lack nucleolus.
- The nucleolus is mainly involved in the assembly of ribosomes.
- After being produced in the nucleolus, ribosomes are exported to the cytoplasm where they translate mRNA.
- Some of the eukaryotic organisms have nucleus that contains up to four nucleoli.
- The nucleolus plays an indirect role in protein synthesis by producing ribosomes.
- Nucleolus disappears when a cell undergoes division and is reformed after the completion of cell-division.
- The size of the nucleolus is found to be related with the synthetic activity of the cell.
- Therefore, the cells with little or no synthetic activities, sperm cells, blastomeres, muscle cell, etc., are found to contain smaller or no nucleoli, while the oocytes, neurons and secretory cells which synthesize the proteins or other substances contain comparatively large-sized nucleoli.
- The number of the nucleoli in the nucleus depends on the species and the number of the chromosomes. The number of the nucleoli in the cells may be one, two or four.
- A nucleolus is often associated with the nucleolar organizer (NO) which represents the secondary constriction of the nucleolar organizing chromosomes, and are 10 in number in human beings. Nucleolar organizer consists of the genes for 18S, 5.8S and 28S rRNAs. The genes for fourth type of r RNA, i.e., 5S rRNA occur outside the nucleolar organizer.

- Nucleolus is not bounded by any limiting membrane; calcium ions are supposed to maintain its intact organization. Nucleolus also contains some enzymes such as acid phosphatase, nucleoside phosphorylase and NAD+ synthesizing enzymes for the synthesis of some coenzymes, nucleotides and ribosomal RNA.
- RNA methylase enzyme which transfers methyl groups to the nitrogen bases occurs in the nucleolus of some cells. Functionally nucleolus is the site where biogenesis of ribosomal subunits (40S and 60S) takes place.
- In it three types of rRNAs, namely 18S, 5.8S and 28S rRNAs, are transcribed as parts of a much longer precursor molecule (45S transcript) which undergoes processing (RNA splicing) by the help of two types of proteins such as nucleolin and U3 sn RNP (U3 is a 250 nucleotide containing RNA, sn RNP represents small nuclear ribonucleoprotein).
- The 5S r RNA is transcribed on the chromosome existing outside the nucleolus and the 70S types of ribosomal proteins are synthesized in the cytoplasm.
- All of these components of the ribosomes migrate to the nucleolus, where they are assembled into two types of ribosomal subunits which are transported back to the cytoplasm.
- The smaller (40S) ribosomal subunits are formed and migrate to the cytoplasm much earlier than larger (60S) ribosomal subunits; therefore, nucleolus contains many more incomplete 60S ribosomal subunits than the 40S ribosomal subunits.
- Such a time lag in the migration of 60S and 40S ribosomal subunits, prevents functional ribosomes from gaining access to the incompletely processed heterogeneous RNA (hn RNA; the precursor of m RNA) molecule inside the nucleus.

Functions of the nucleus

- Speaking about the functions of a cell nucleus, it controls the hereditary characteristics of an organism.
- This organelle is also responsible for the protein synthesis, cell division, growth, and differentiation.

- Some important functions carried out by a cell nucleus are:

1.Storage of hereditary material, the genes in the form of long and thin DNA (deoxyribonucleic acid) strands, referred to as chromatins.

2.Storage of proteins and RNA (ribonucleic acid) in the nucleolus.

3.Nucleus is a site for transcription in which messenger RNA (mRNA) are produced for the protein synthesis.

4.Exchange of hereditary molecules (DNA and RNA) between the nucleus and rest of the cell.

5.During the cell division, chromatins are arranged into chromosomes in the nucleus.

6.Production of ribosomes (protein factories) in the nucleolus.

7.Selective transportation of regulatory factors and energy molecules through nuclear pores.

As the nucleus regulates the integrity of genes and gene expression, it is also referred to as the control center of a cell. Overall, the cell nucleus stores all the chromosomal DNA of an organism.

Chromosome:

- German biologist Walter Flemming in the early 1880s revealed that during cell division the nuclear material organize themselves into visible thread like structures which were named as chromosomes which stains deep with basic dyes.
- The term chromosome was coined by W. Waldeyer in 1888. ***'Chrome'*** means coloured and ***'soma'*** means body, hence they mean "colored bodies" and can be defined as higher order organized arrangement of DNA and proteins.
- It contains many genes or the hereditary units, regulatory elements and other nucleotide sequences.
- Chromosomes also contain DNA-bound proteins, which serve in packaging the DNA and control its functions.

- Chromosomes vary both in number and structure among organisms and the number of chromosomes is characteristic of every species.
- Benden and Bovery in 1887 reported that the number of chromosomes in each species is constant.
- W.S. Sutton and T. Boveri in 1902 suggested that chromosomes are the physical structures which acted as messengers of heredity.
- Chromosomes are tightly coiled DNA around basic histone proteins, which help in the tight packing of DNA.
- During interphase, the DNA is not tightly coiled into chromosomes, but exists as chromatin.
- In eukaryotes to fit the entire length of DNA in the nucleus it undergoes condensation and the degree to which DNA is condensed is expressed as its packing ratio which is the length of DNA divided by the length into which it is packaged into chromatin along with proteins.

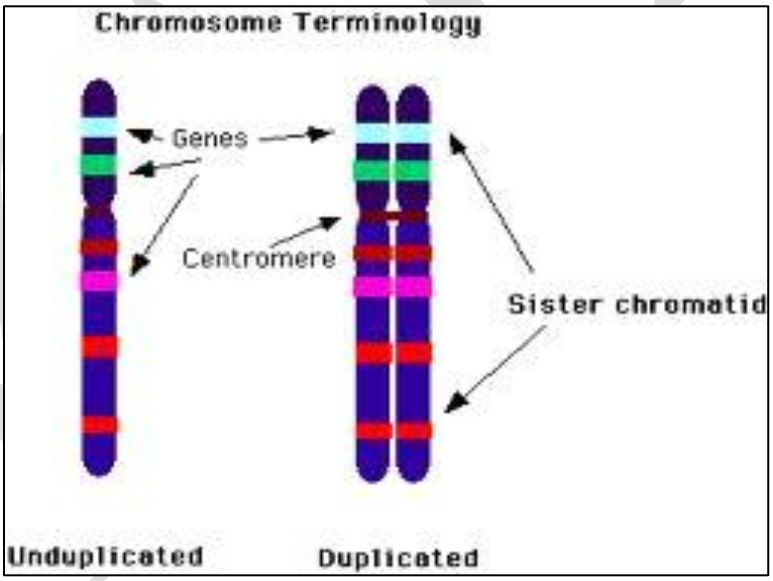


Fig:*Eukaryotic chromosome*

- The shortest human chromosome contains 4.6 x 107 bp of DNA. This is equivalent to 14,000 μm of extended DNA.

- In its most condensed state during mitosis, the chromosome is about 2 μm long.
- This gives a packing ratio of 7000 (14,000/2). The DNA is packaged stepwise into the higher order chromatin structure and this is known as "hierarchies of chromosomal organization".

Chromosome number:

- There are normally two copies of each chromosome present in every somatic cell.
- The number of unique chromosomes (N) in such a cell is known as its haploid number, and the total number of chromosomes (2N) is its diploid number.
- The suffix 'ploid' refers to chromosome 'sets'. The haploid set of the chromosome is also known as the genome.
- Structurally, eukaryotes possess large linear chromosomes unlike prokaryotes which have circular chromosomes.
- In Eukaryotes other than the nucleus chromosomes are present in mitochondria and chloroplast too.
- The number of chromosomes in each somatic cell is same for all members of a given species.
- The organism with lowest number of chromosome is the nematode, *Ascaris megalocephalusunivalens* which has only two chromosomes in the somatic cells (2n=2).

Organism	No. of chromosomes
Arabidopsis thaliana (diploid)	10
Maize (diploid)	20
Wheat (hexaploid)	42
Common fruit fly (diploid)	8
Earthworm (diploid)	36
Mouse (diploid)	40
Human (diploid)	46
Elephants (diploid)	56
Donkey (diploid)	62
Dog (diploid)	78
Gold Fish (diploid)	100-104
Tobacco(tetraloid)	48
Oat (hexaploid)	42

Table:*Number of chromosomes in different organisms.*

Autosomes and sex chromosomes:

- In a diploid cell, there are two of each kind of chromosome (termed homologus chromosomes) except the sex chromosomes.
- In humans one of the sex has two of the same kind of sex chromosomes and the other has one of each kind. In humans there are 23 pairs of homologous chromosomes (2n=46).
- The human female has 44 non sex chromosomes, termed autosomes and one pair of homomorphic sex chromosomes given the designation XX.
- The human male has 44 autosomes and one pair of heteromorphic sex chromosomes, one X and one Y chromosome.

Structure of Chromosome:

- A chromosome at mitotic metaphase consists of two symmetrical structures called chromatids.
- Each chromatid contains a single DNA molecule and both chromatids are attached to each other by centromere and become separated at the beginning of anaphase.

- The chromomeres are bead like accumulations of chromatin material that are sometimes visible along interphase chromosomes.
- The chromomere bearing chromatin has an appearance of a necklace in which several beads occur on a string.
- Chromomeres are regions of tightly folded DNA and become especially prominent in polytene chromosomes.
- Centromere in a chromosome contain specific DNA sequences with special proteins bound to them, forming a disc shaped structure, called kinetochore.
- In electron microscope the kinetochore appears as a plate or cup like disc, 0.20-0.25 nm, in diameter situated upon the primary constriction or centromere.
- The chromosomes of most organisms contain only one centromere and are known asmonocentric chromosomes.
- Some species have diffused centromeres, with microtubules attached along the length of the chromosomes and are termed holocentric chromosomes.
- Chromosomes of *Ascaris megalocephala* are examples of diffused centromeric chromosomes.
- Telomere is the chromosomal ends which prevents other chromosomal segments to be fused with it.
- Besides the primary constrictions or centromeres, chromosomes also posses secondary constriction at any point of the chromosome and are constant in their position and extent.
- These constrictions are helpful in identifying particular chromosomes in a set.
- Chromosomes also contain nucleolar organizers which are certain secondary constrictions that contain the genes coding for 5.8S, 18S and 28S ribosomal RNA and induce the formation of nucleoli.
- Sometimes the chromosomes bear round, elongated or knob like appendages known as satellites. The satellite remains connected with the rest of the chromosomes by a thin chromatin filament.

Possible Questions

2 Marks

1. What is microbodies.
2. Define lysosomes.
3. Write about functions of vacuoles.
4. Describe about various type of ribosomes subunits.

8 Marks

1. Explain lysosomes structure and its function.
2. Brief about the vacuoles.
3. Write about the microbodies.
4. Describe in detail structure and functions of mitochondria.
5. Explain chloroplasts.
6. Brief a note on Ribosomes.
7. Explain Nucleus.
8. Give a detailed account on Nucleoplasm.
9. Explain Chromosomes in detail.
10. Write about the various types of chromosomes.

Unit V

Cell abnormalities: Carcinogenesis, agents promoting carcinogenesis, characteristics and molecular basis of Cancer.

Cancer

- Cancer was first described by Hippocrates, the Greek physician who lived during 460–370 BC.
- He coined the term carcinoma. In Greek *karcinos means "crab"* .This is because tumors often have a central cell mass with extensions radiating outward that mimic the shape of a Crab.

- The Roman physician, Celsus, who lived during 28-50 BC, later translated the Greek term into *cancer, the Latin word for crab.*
- Galen (130-200 AD), another Roman physician, used the word *oncos (Greek for* swelling) to describe cancers.

Study of cancer - *oncology*; cancer specialists - *oncologists*.

- Cancer is a family of about 200 complex diseases capable of affecting any of our tissues
- Cancer is a group of diseases of higher multicellular organisms
- Cancers of different organs or different origins have different causes, varied course and varied response to treatment
- Each tumour is different
- The common phenomenon observed is the deregulation of growth processes caused by multiple changes in gene expression
- Deregulation occurs not only in cell proliferation but also in cell death
- Evolution of a population of cells that can invade tissues and metastasize to distant sites, causing significant morbidity
- If left untreated, this disease is fatal.

Types of cancer :

Solid cancers:

- **Carcinomas** are solid cancers/solid tumors) (90%) that arise from the epithelial cells
- **Sarcomas** are solid cancers that arise from the connective tissue cells such as muscle and bone.

Liquid cancers:

- It arises from the bone marrow or lymphatic system, which carries fluids throughout the body.
- **Leukemia** (cancer of the blood or bone marrow).
- **Lymphoma**(cancer in the lymphatic system).
- **Multiple myeloma** (cancer of the plasma cells in the blood).

Tumors:

- A tumor is any abnormal mass or swelling in the body
- It can be a **benign (noncancerous) tumor. It is generally harmless but a large tumor in certain locations like brain can be harmful or a malignant (cancerous)** tumor
- All tumors are believed to begin with mutations in a single cell that alters its DNA and that of its offspring thus initiating functional aberrations upon accumulation of these changes.

Mutations:

- **Sporadic mutations** occur spontaneously during the lifespan of a cell
- It may be **due to errors in cell cycle** when a cell copies its DNA prior to dividing the **incorrect repair** of a damaged DNA molecule or **chemical modification** of the DNA each of this interferes with expression of the genetic information.
- **Inherited mutations are present in the** DNA contributed by the sperm and/or egg at the moment of conception.
- To date, 90–95% of diagnosed cancers appear to be sporadic in nature and thus have no heredity basis.

Causes :

- Carcinogenesis – is the process of cancer development. It is a multistep process thriving with mutations, epigenetic changes and a conducive microenvironment.
- The factors/agents that induce/cause cancer are called carcinogens. Most carcinogenic agents cause **genetic damage** and induce **neoplastic transformation** of cells.
- They include chemicals, radiation and microbes aided by a host of risk factors. Thus this list includes external/environmental and internal factors which may act together.

- Hereditary – Inherited cancer syndromes o Retinoblastoma, Li-Fraumeni syndrome, Multiple Endocrine Neoplasia (MEN) type 1 and 2, Hereditary Non-Polyposis Colon Cancer (HNPCC).

Risk factors :

- Many Risk Factors have been identified that increase the risk
- Diet and Tobacco play a major role
- Sexual behavior
- Occupational hazards
- Alcohol
- Pollution
- Industrial products
- Medical products/procedures
- Sedentary living

Chemical Carcinogens :

- ***Sir Percival Pott***– in the 18th century, related the high incidence of scrotal skin cancer in chimney sweeps to chronic exposure to soot – bathing daily controlled the cancer! – First known correlation between a cancer and a chemical / environmental agent.

- Mutagenic potential of chemicals assessed by Ames test – Ability of the chemical to induce mutations in Salmonella typhimurium. Most carcinogens are mutagens.

Major Chemical Carcinogens :

Alkylating agents	B-propiolactone, Dimethyl sulfate, Diepoxy butane. (Cyclophosphamide, Chlorambucil,(Nitro-sources,etc.)

Acylating agents	1-Acetyl-imidazole Dimethyl carbamyl chloride
polycyclic and heterocyclic Aromatic hydrocarbons	Benz(a)anthracene Benzo(a)pyrene 3-methyl chloranthrene
Aromatic amines, Amides, Azo dyes	2-Naphthylamine Benzidine Dimethyl aminoazo benzene (butter yellow)
Others	Others Nitrosamine and amides Vinyl chloride Nickel Chromium Insecticides fungicides
Plant and microbial Products	Aflatoxin B1 Griseofulvin Cycasin Safrole Betel nuts

Chemical Carcinogens :

- **Cigarette smoke** contains **43 known carcinogens**, including PAHs, Polonium 210, N'-Nitrosonornicotine(NNN).
- A major cause of cancer of lung, larynx, oral cavity.

Radiation :

- ***Hiroshima and Nagasaki***– Increased incidence of Breast and thyroid cancer years after the bombings.
- ***Chernobyl disaster***– Thyroid carcinoma .
- ***Marshall and Rongelap Islands***– Near nuclear weapons testing sites high incidence of Thyroid carcinoma.

Radiation Types –

- UV rays – from the sun; Skin cancers.
- UVC – more mutagenic but filtered out by Ozone.
- UVB – Pyrimidine dimers (corrected by Nucleotide Excision Repair XerodermaPigmentosum a case in point).
- Ionizing radiation o X-rays ;

1. γ rays
2. Particulate (α, β, protons, neutrons).

Microbes :

Viruses :

- Human Papilloma Virus (HPV) – cervical cancer, oral cancer.
- Epstein Barr Virus (EBV) – Burkitt Lymphoma.
- Hepatitis B Virus – Liver cancer.
- Human T-Cell Leukemia Virus Type 1 (HTLV-1) – Leukemia.

Bacteria :

- Helicobacter pylori – stomach cancer

Other non-microbial pathogens :

- Schistosoma (bladder fluke)
- Inflammation
- Bladder cancer

The Neoplastic Phenotype :

Terms used:

1. **Anaplasia:**

   Lack of differentiation – a hallmark of malignant transformation.

2. **Dysplasia:**

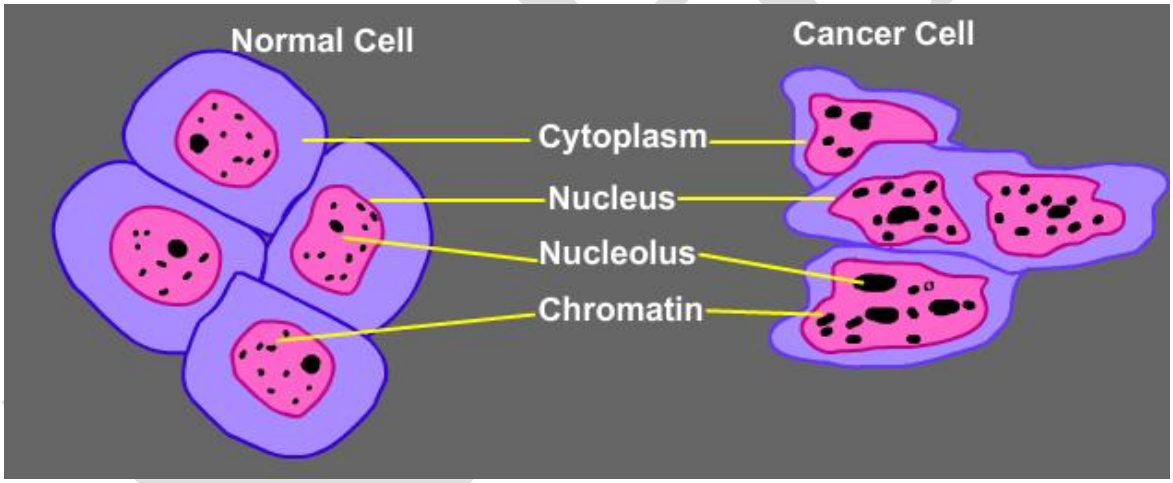
   A loss in uniformity of the individual cells as well as a loss in architectural orientation (abnormal structural change).

3. **Hyperplasia:**

   Increase in cell number; regulated and physiological.

4. **Hypertrophy:**

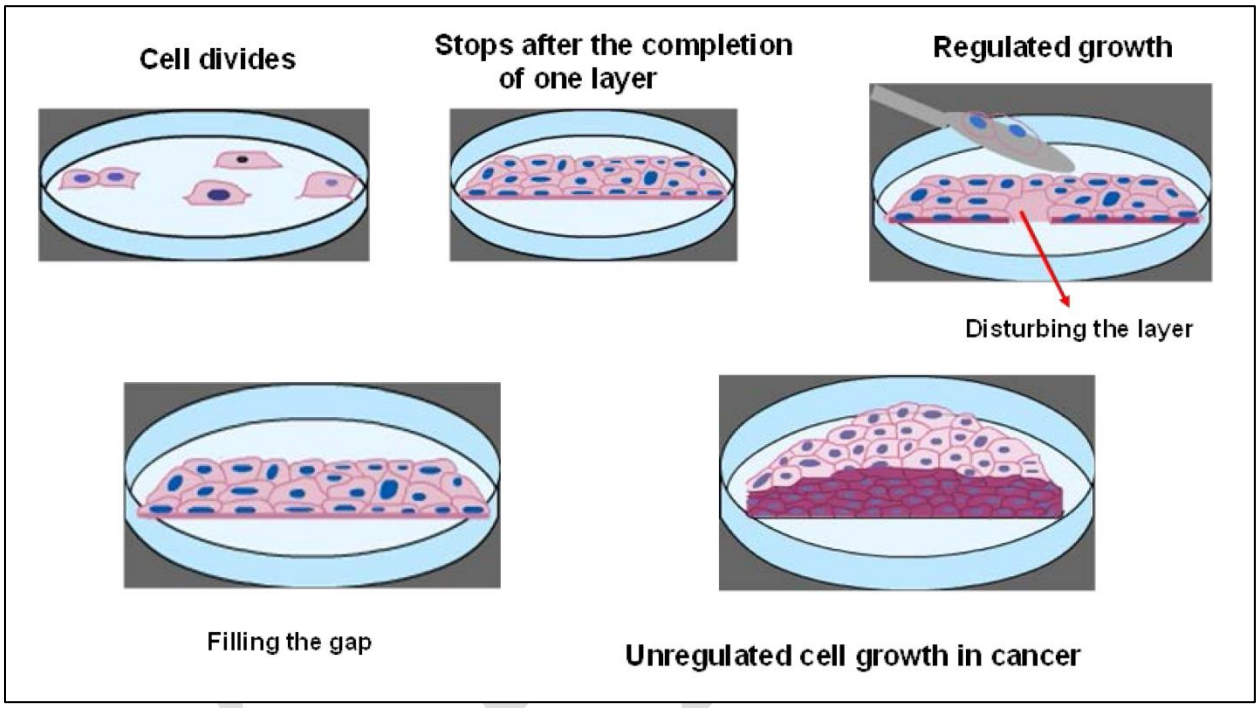
   Increase in cell size.



Metastasis:

- It is the condition in which the cells from the original primary tumor migrate to either local or distant locations in the body where they will divide and form secondary tumors.
- Malignant tumors that do not metastasize are very rare; e.g.: Gliomas, basal cell carcinomas of skin.

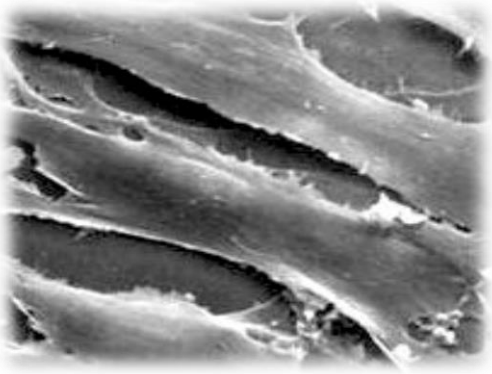
- ***How does metastasis affect the outcome for the patient?***Significantly worsens prognosis 90% of all cancer deaths are due to metastases, only 10% due to the primary tumors.



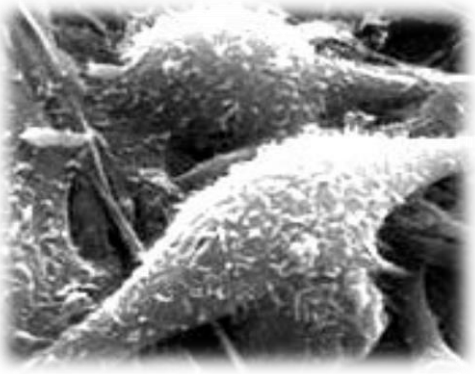
Development of a malignant tumor:

- Human body has 1014 cells descended from a single fertilized egg cell. This can partly explain why each tumor is different.
- Cancer cells have mutations that encourage mutation – flawed DNA repair mechanisms.
- Conversion of a normal cell into a malignant cell is now known to require multiple mutations. At least 6 to 13 genes must be disabled to have a full-fledged malignant cancer.
- Many mutations occur in cells over your lifetime.

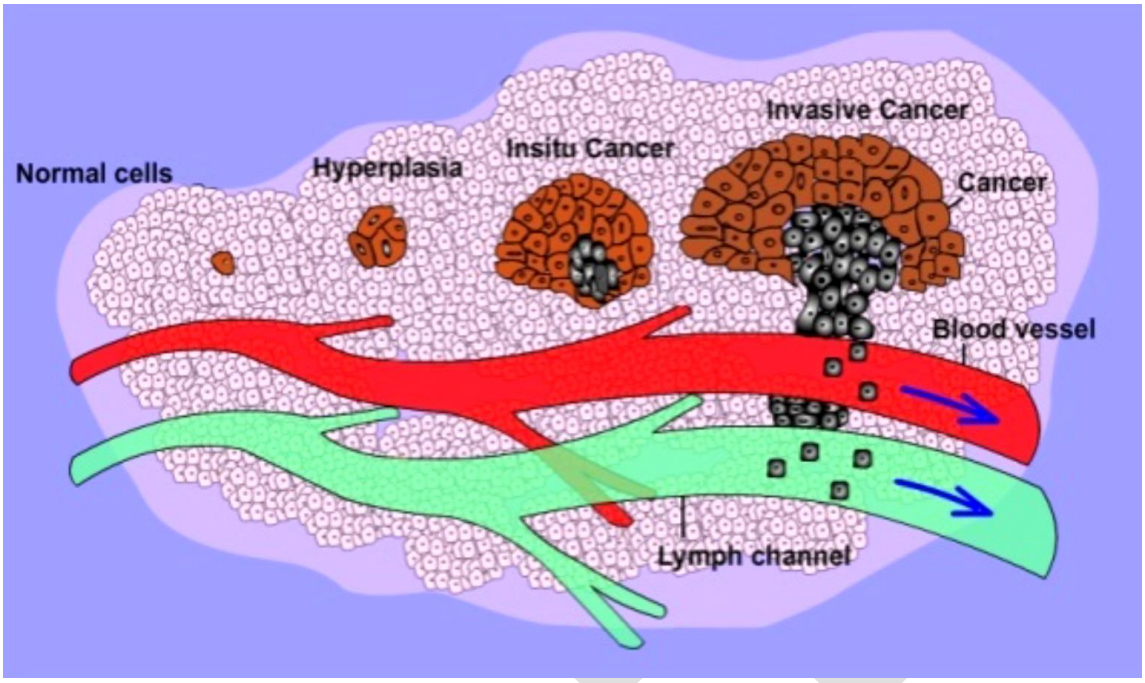
- Chances of developing cancer increase as a person gets older because more time has been available for mutations to accumulate.
- Evolutionary journey to develop cancer also requires the epigenetic changes and a facilitating microenvironment.
- Most cancers occur in highly proliferating cells (epithelial, stem cells) or cells that can come in contact with chemicals/viruses/environment.



Large cytoplasm
Single nucleus
Single nucleolus
Fine chromatin

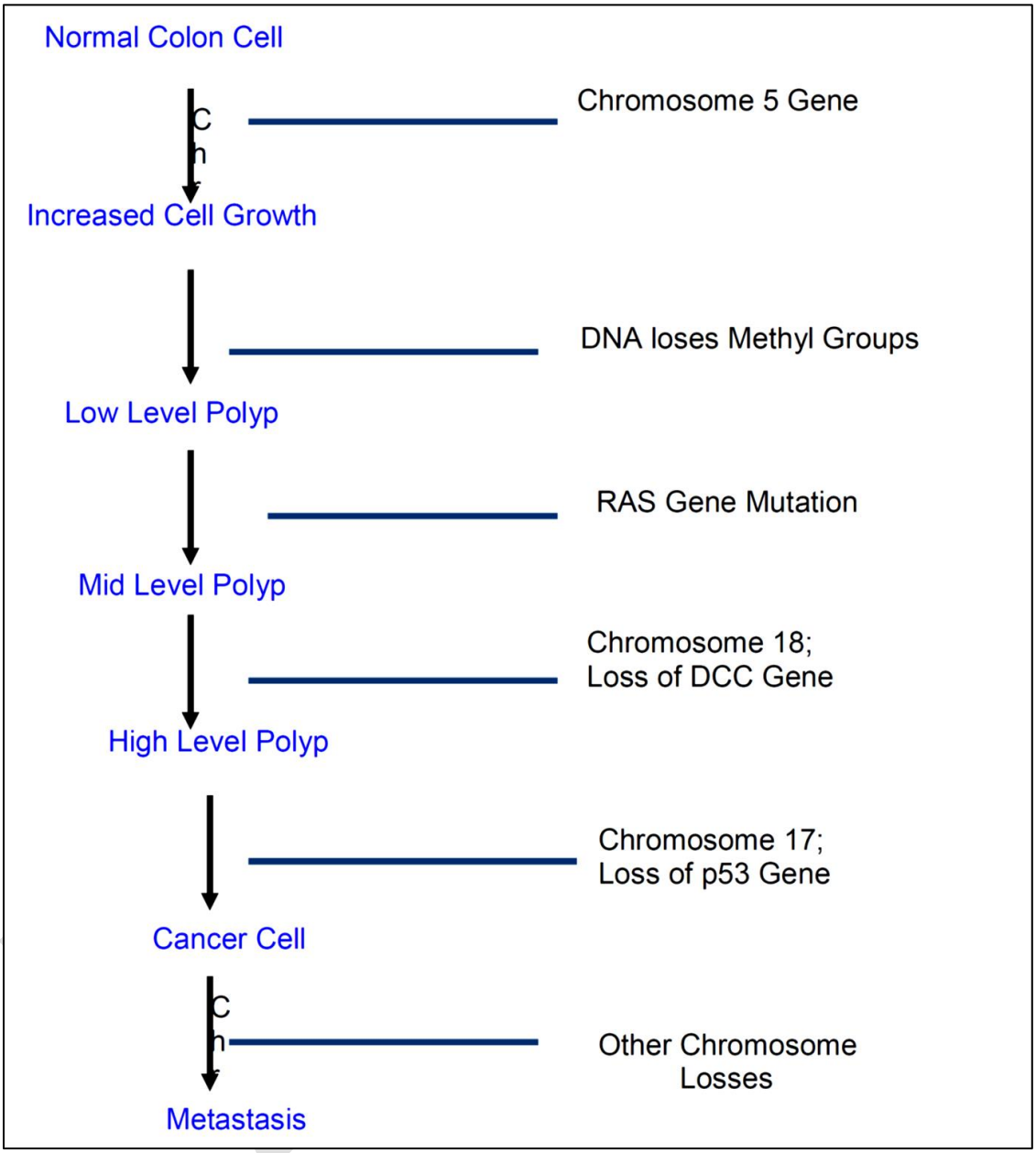


Small cytoplasm
Multiple nucleus
Multiple nucleolus
Coarse chromatin

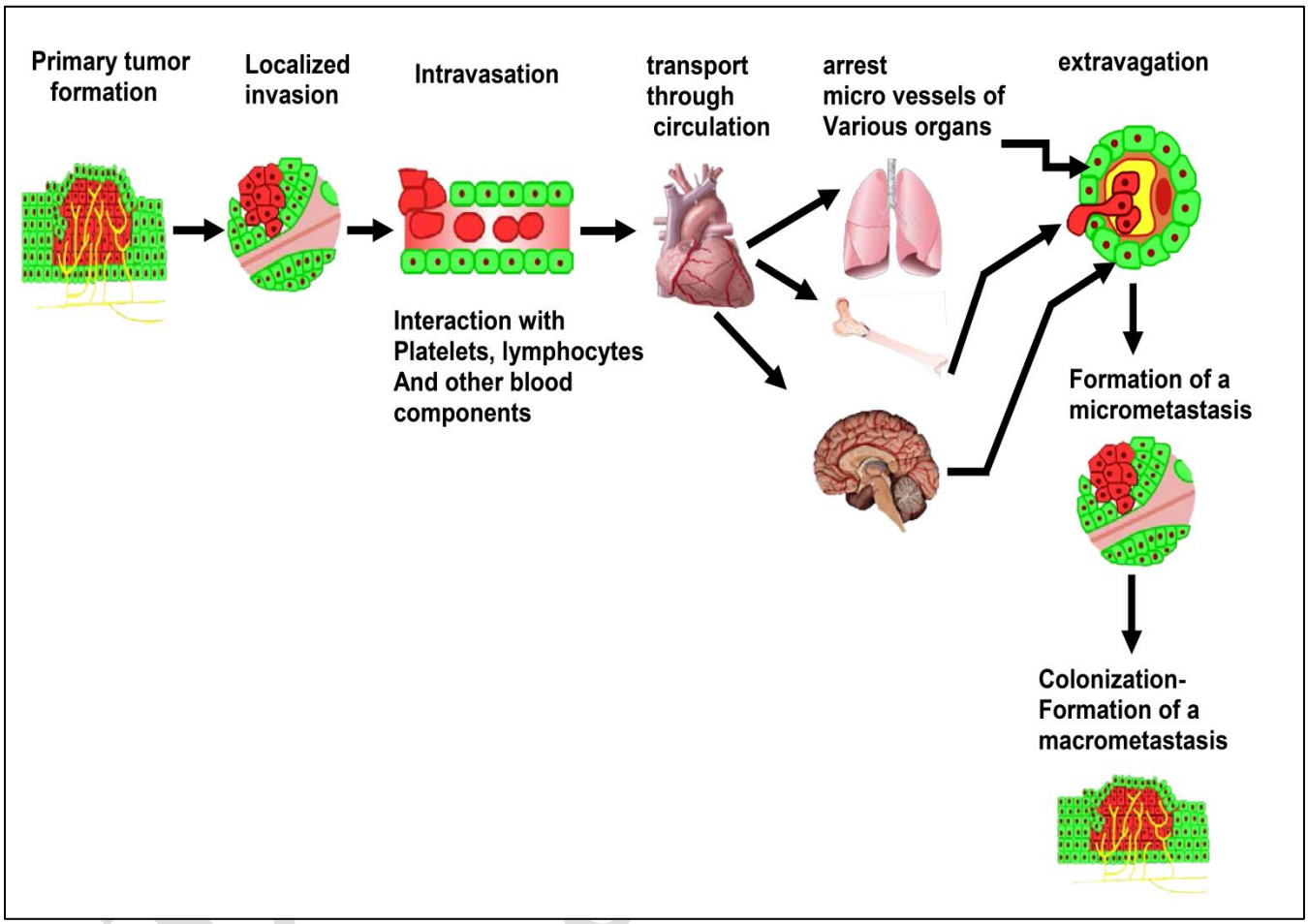


- Human body has 1014 cells descended from a single fertilized egg cell. This can partly explain why each tumor is different.
- Cancer cells have mutations that encourage mutation – flawed DNA repair mechanisms.
- Conversion of a normal cell into a malignant cell is now known to require multiple mutations. At least 6 to 13 genes must be disabled to have a full fledged malignant cancer.
- Many mutations occur in cells over your lifetime.
- Chances of developing cancer increase as a person gets older because more time has been available for mutations to accumulate.
- Evolutionary journey to develop cancer also requires the epigenetic changes and a facilitating microenvironment.

Development of a Colon Cancer Cell :


Normal Colon Cell
Chromosome 5 Gene
Increased Cell Growth
DNA loses Methyl Groups
Low Level Polyp
RAS Gene Mutation
Mid Level Polyp
Chromosome 18;
Loss of DCC Gene
High Level Polyp
Chromosome 17;
Loss of p53 Gene
Cancer Cell
Other Chromosome
Losses
Metastasis

The Invasion-Metastasis Cascade :



Six steps to metastasis:

- Breach of basement membrane
- Intravasation
- Transport to distant tissues
- Arrest at target organs and extravasation
- Formation of micrometastases
- Colonization at distant sites

Routes:

Lymphatic spread:

Although tumors have no lymphatic supply, surrounding areas have lymphatic vessels

Most common way of spread

Eg: Breast cancer and axillary lymph nodes.

Routes :

Hematogenous spread :

Liver, lungs, bone marrow most commonly involved, because of extensive blood supply.

Veins more easily penetrated by tumor than arteries (because of thinner walls).

Cancer cells are much larger than RBCs and WBCs and not easily deformable; they get **trapped in capillary beds, esp. in the lungs.**

Theories of carcinogenesis :

Early theories :

Humoral origin – "black bile" (Hippocrates, Galen)

Lymphatic origin (John Hunter)

Humoral theory: Hippocrates believed that the body had 4 *humors* (body fluids) : blood, phlegm, yellow bile, and black bile. When the humors were balanced, a person was thought to be healthy. Too much or too little of any of the humors caused disease. An excess of black bile in various body sites was thought to cause cancer. This theory of cancer was passed on by the Romans and was accepted by Galen. This remained the unchallenged through the Middle Ages for over 1,300 years.

Lymph theory: Stahl and Hoffman theorized that cancer was composed of fermenting and degenerating lymph varying in density, acidity, and alkalinity. The lymph theory gained rapid support. The eminent Scottish surgeon John Hunter (1728−1793) agreed that tumors grow from lymph constantly thrown out by the blood.

Modern theories :

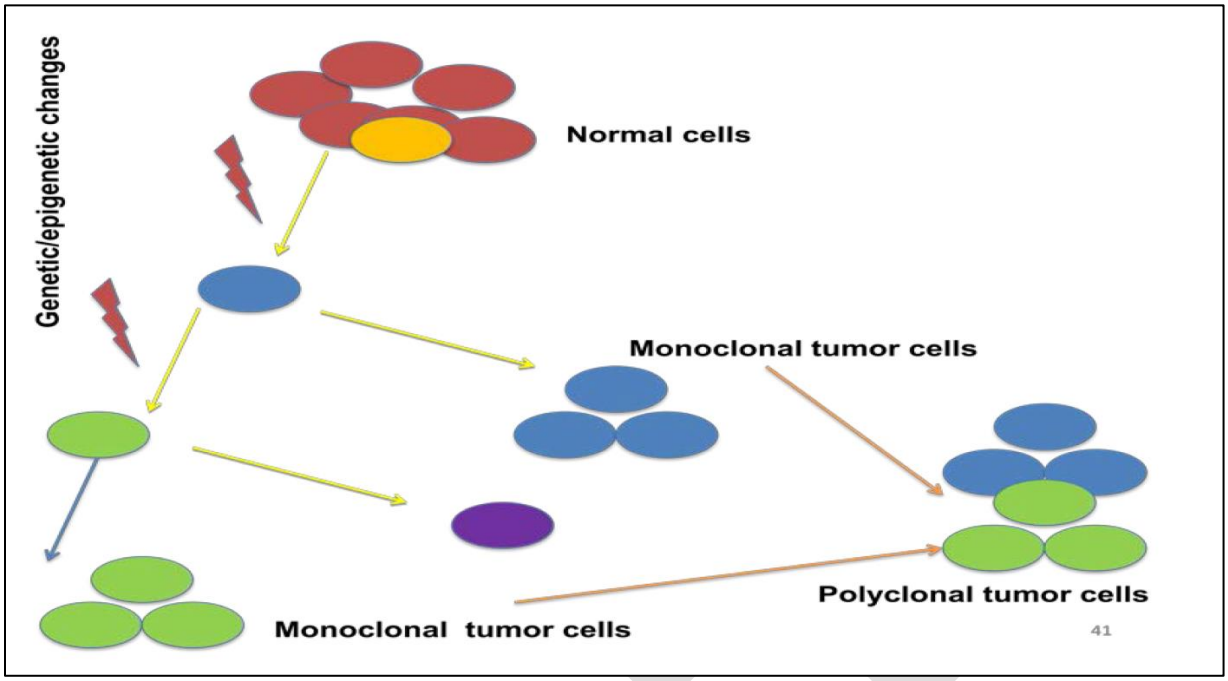
Somatic Mutation Theory:

Somatic mutation theory was first enunciated in 1914 by Theodor Boveri in his book entitled The Origin of Malignant Tumors claimed that ‘‘the problem of tumors is a cell problem’’ and that cancer was due to ‘‘a certain permanent change in the chromatin complex’’ which, ‘‘without necessitating an external stimulus, forces the cell, as soon as it is mature, to divide again.’’

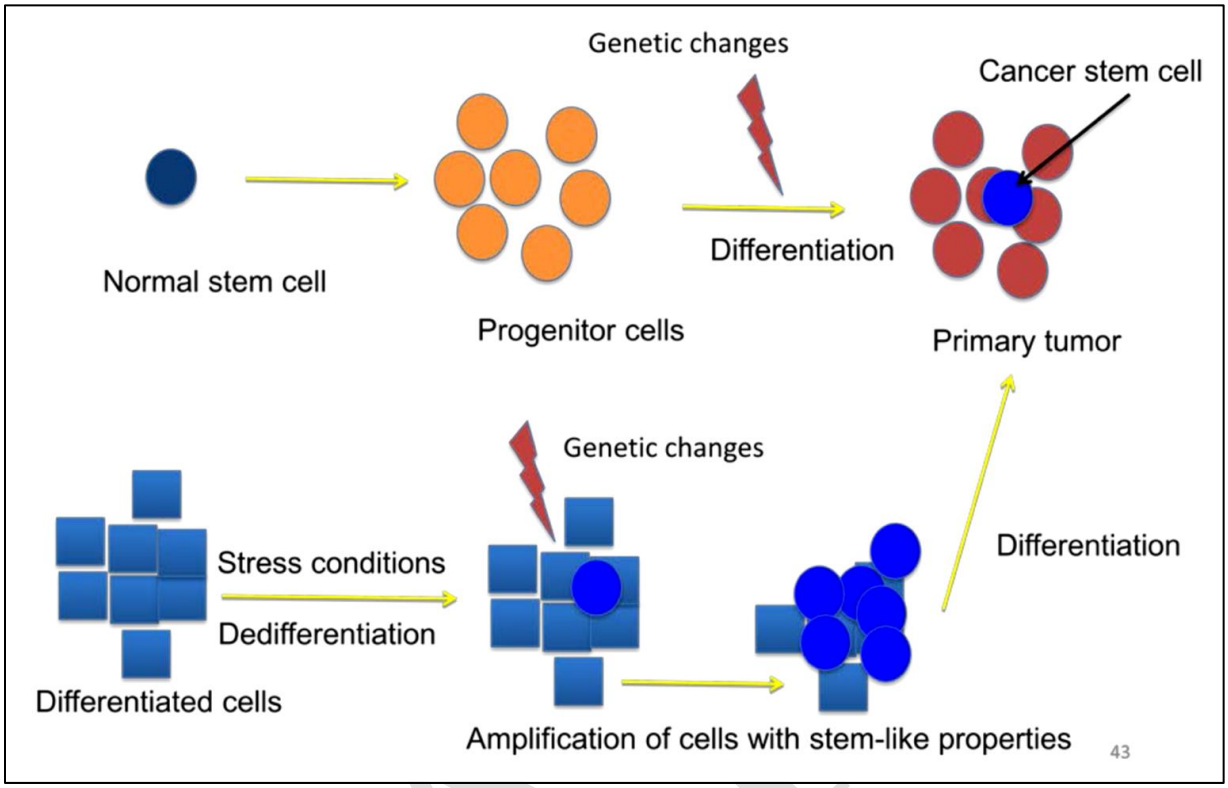
Tissue Organization Field Theory :

“Carcinogenesis is a problem of tissue organization, comparable to organ formation during early development”

Clonal hypothesis :



Cancer Stem cell theory :



Tissue organization field theory (TOFT) states that carcinogenesis takes place at the tissue level of biological organization, as does normal morphogenesis. A single or multiple carcinogenic exposure acts disturbing the reciprocal biophysical and biomechanical communication between the parenchyma and the mesenchyme/stroma. This results in miscues that manifest morphologically in both the stroma and the epithelium. The proliferation and motility restraints imposed by normal tissue architecture loosen and as a consequence, hyperplasia of the epithelium may occur. Further alteration of the reciprocal interactions between tissue compartments will induce metaplasia, dysplasia, and carcinoma. The stroma also may show alterations.

Theories on metastasis :

- The Anatomic Model.
- Secondary tumors occur in the organs which they come across first during their journey into the blood/lymphatic circulation from the primary tumor.
- Tumor cells from colon first pass through the capillary bed of the liver that provides a suitable environment for the growth of these secondary tumors.
- Theories on metastasis.
- The Seed and Soil Hypothesis.

In 1889 Stephen Paget observed that breast cancer patients often had secondary metastatic tumors in the liver. For him selective accessibility of the liver by the blood supply was not acceptable as many other organs are equally accessible for the blood supply. He hypothesized that tumor cells (seeds) can only grow in selective organs (soil) that provided suitable growth environment.

Modern theories on metastasis :

Chemo-attraction theory :

- Organ-specific attractant molecules (chemokines) enter the circulation
- They attract the circulating tumour cells containing the corresponding chemokine receptors
- This stimulates them to invade the walls of blood vessels and enter the organs.
- Higher expression of CXCR4 receptor in breast-cancer tissue than normal breast tissue
- CXCL12 chemokine is expressed in many metastatic sites/organs of breast cancer, such as lymph nodes, bone marrow and lungs.

Therapy :

- Conventional therapies include

- Surgery
- Radiation
- Chemotherapy
- No conventional drug is without side effect
- With our understanding on the molecular biology of cancer, now researchers have developed many targeted therapies

Cell Signal Transduction

As living organisms we are constantly receiving and interpreting signals from our environment. These signals can be light, heat, odours, touch or sound. The cells of our bodies are also constantly receiving signals from other cells.

In animals, rapid responses to the changes in the environment are mediated primarily by the nervous system and by hormones including small peptides, small non-peptide molecules such as the catecholamines (Dopamine, epinephrine, norepinephrine).

Various secondary messengers are involved in signaling process. These signaling molecules are released from the cells and they travel through the blood to their specific target cells. Some molecules are transported long distances by the blood while others have more of local effects. Certain membrane-bound proteins on one cell can directly signal an adjacent cell.

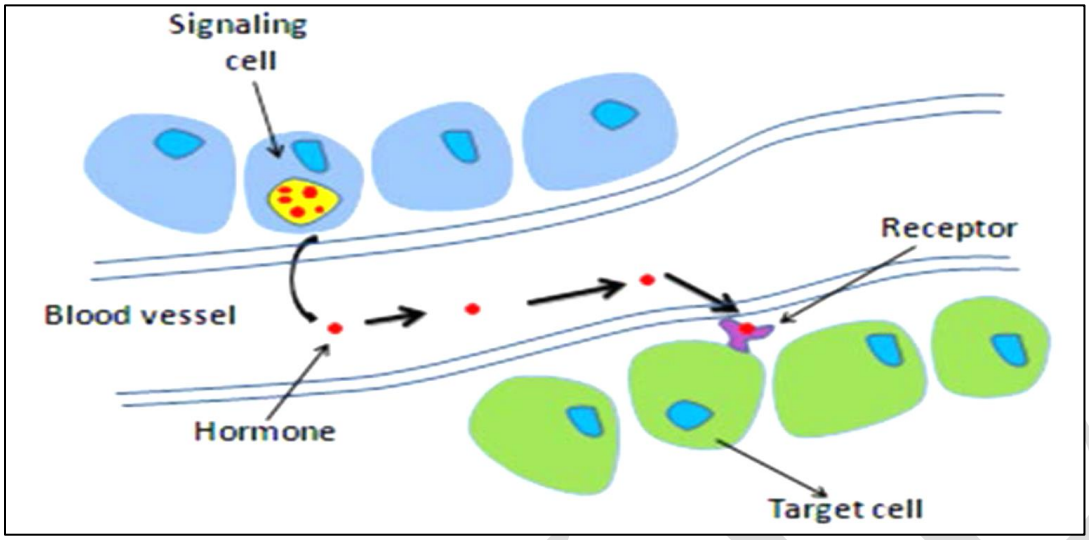


Fig :Signaling molecules released from cell and transported by the blood to the target cell.

Cell signaling can be divided into 3 stages:

1. Reception:

A cell detects a signaling molecule from the outside of the cell. A signal is detected when the ligand binds to a receptor protein on the surface of the cell or inside the cell.

2. Transduction:

When the signaling molecule binds to the receptor, it changes the receptor protein. This change initiates the process of transduction. Each relay molecule in the signal transduction pathway changes the next molecule in the pathway.

3. Response:

Finally, the signal triggers a specific cellular response.

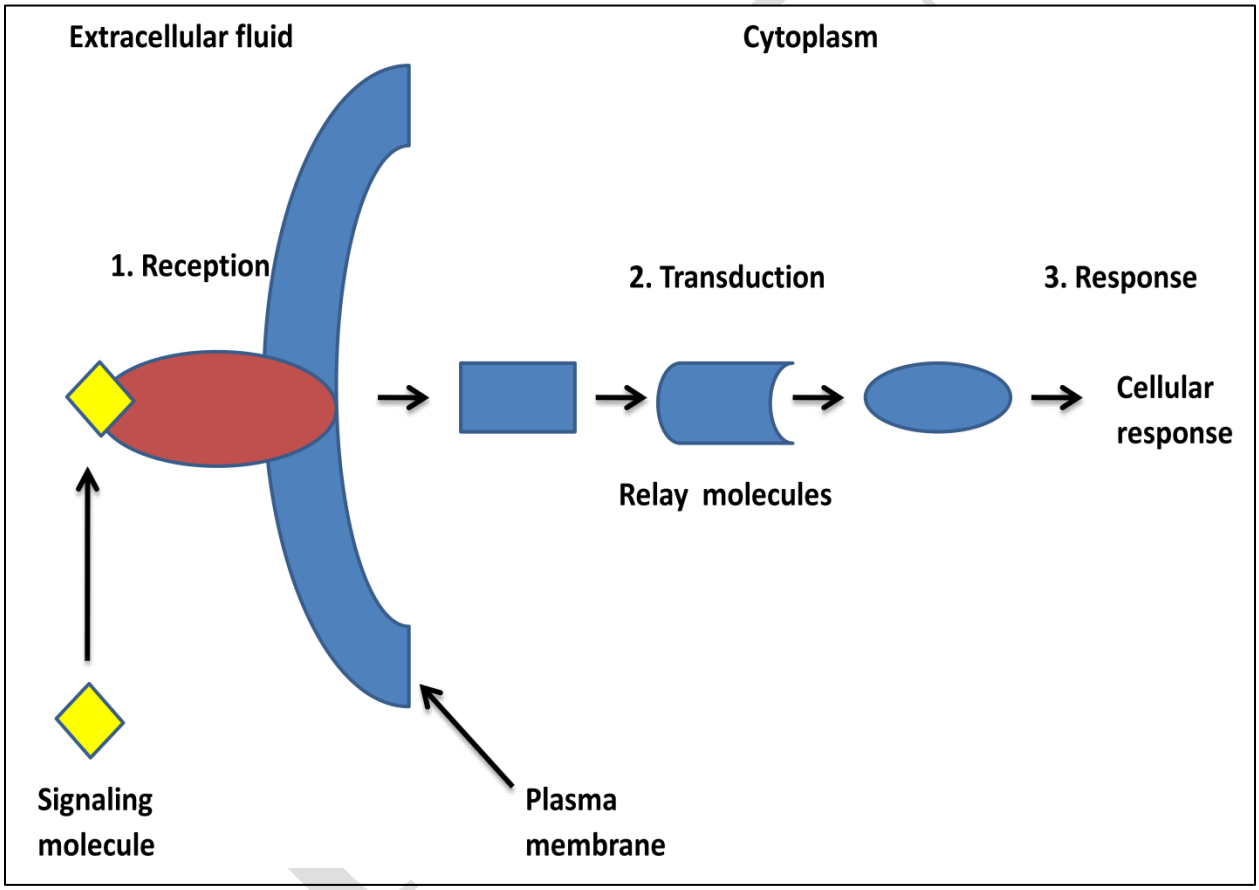


Fig :Cell signaling stages.

Signal Transduction:

- Signal transduction is phenomenon which involves in the transfer of signal from extracellular to intracellular environment through the cell surface receptor protein that stimulate intracellular target enzymes, which may be either directly linked or indirectly coupled to receptors by G proteins.

- These intracellular enzymes serve as downstream signalling elements that propagate and amplify the signal initiated by ligand binding. Thus, signal transduction pathway allows cellsto respond to extracellular environmental signals. These signals can be physical and chemical such as light, oxygen, nutrient, hormones.

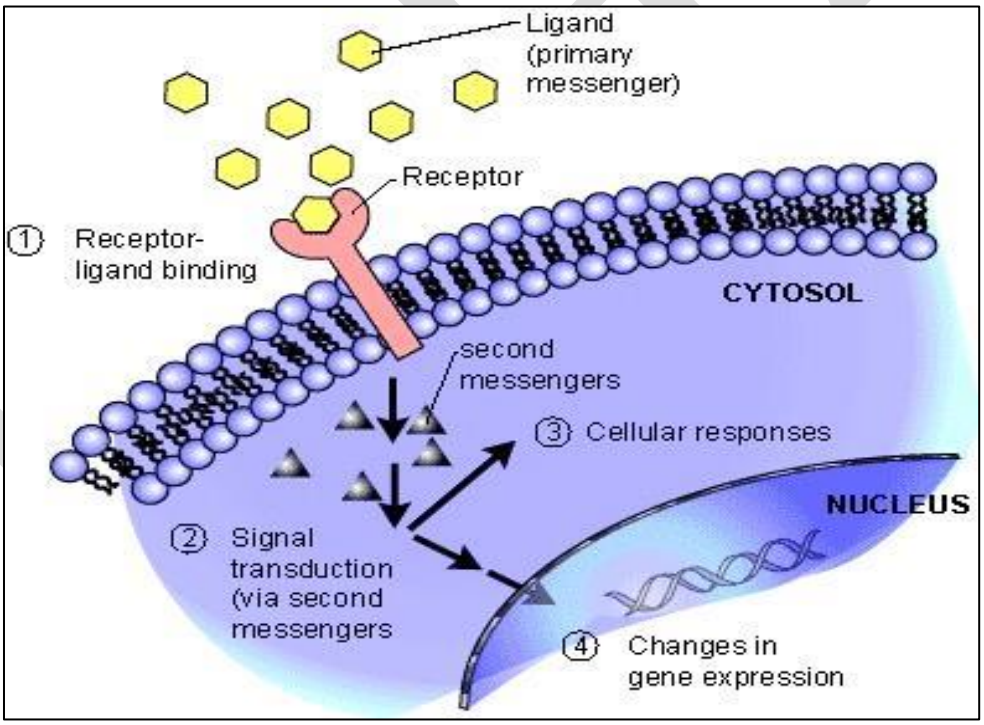


Fig :*Transduction.*

- Thus signal transduction begins with receiving signal to the cell receptor and end with a change in cellular function.
- The cell receptor can be of various types- ***G-protein coupled receptor***, ***tyrosin kinase receptor*** etc.
- The transduction process is typically mediated via a cascade of some important second messengers including ***cAMP***, ***cGMP***, ***calcium ion***, ***inositol 1, 4, 5-trisphosphate, (IP3)***, and ***diacylglycerol (DAG)***.
- Second messengers are intracellular molecules that change in concentration in response to environmental signals and involve in conveying information inside the cell.
- Most of signalling molecules are too large and too polar so they are unable to cross the membrane, hence there is no appropriate transport system. In this case these signaling molecules transmit signals through cell surface receptor protein without crossing the cell membrane.
- These receptors are intrinsic membrane protein which consist both extracellular and intracellular domain.
- A binding site present in extracellular domain specifically recognizes the signaling molecule (i.e. well known as ligand). Such binding sites are analogous to enzyme active sites except that no catalysis takes place within them.
- When these signal molecules comes and bind to binding site on receptor protein in extracellular region then some conformational change occurs in tertiary and quaternary structure of the receptor which results in the drastic change in the intracellular domain of the receptor.
- These structural changes are not sufficient to yield an appropriate response, because they are restricted to a small number of receptor molecules in the cell membrane. The information embodied by the presence of the ligand, often called the primary messenger, must be transduced into other forms that can alter the biochemistry of the cell.

Possible Questions

2 Marks

1. What is cancer.
2. Define Metastasis.
3. Give some examples of chemical carcinogens.
4. Give some examples of biological carcinogens.
5. Define Carcinoma.
6. What is mutation.

8 Marks

1. Explain the term Oncology.
2. Explain the characteristics of tumor.
3. Explain in detail about the causative agents for tumor
4. Comment on apoptosis
5. Explain DNA repair mechanism
6. Describe in detail oncogene
7. Explain proto-onco gene
8. Describe Carcinogenesis with suitable examples.

S.no.	Questions	Opt 1	Opt 2	Opt 3	Opt 4	Answer
	Unit-I					
1	--- ----------is the structural and functional unit of all organisms	cell	DNA	RNA	Gene	cell
2	The largest known cell is an---------	Ostrich Egg	Nerve cell	Macrophage	Bone cells	Ostrich Egg
3	Bacteria is Classified in to gram positive and gram negative on the basis of----------	Cell wall	Nucleus	Mitochondria	Shape	Cell wall
4	The heredity unit of cell is	Genes	Cell membrane	Ribosomes	Nucleus	Genes
5	---------- is a special type of nuclear division which segregates one copy of each homologous chromosome into each new "gamete".	Mitosis	Meiosis	Both	None of the above	Meiosis
6	Long, whip like micro fibrils that facilitate movement by cells are known as ___.	cilia	flagella	leather	pseudopodia	flagella
7	Simple non-nucleated cells are ---------	Prokaryotic cells	Eukaryotic cells	Stem cells	Blood cells	Prokaryotic cells
8	The contribution of Robert Hook cointed the term--------	Karyokinesis	cell membrane	cell	Inverted microscope.	cell
9	Cell wall is absent in	Mycoplasma	Gram positive bacteria	Gram-negative Bacteria.	All	Mycoplasma
10	Peptidoglycons found in which organism.	Fungi	Algae	Protozoa	Bacteria	Bacteria
11	The junctions between nerve cells are known as ___.	gap junctions	synapses	tight junctions	villi	synapses
12	The term cell was coined by	Schwann	Robert Hooke	de Bary	Tatum	Robert Hooke

13	Cell theory was proposed by	Beadle and Tatum	Robert Hooke	Schleiden and Schwann	Leenuwenhoeck	Schleiden and Schwann
14	The cell theory is not applicable to	Bacteria	algae	Virus	fungi	virus
15	Semiautonomous organelle in the cell is	Peroxisomes	Chloroplast	reticulam	Golgibodies	Chloroplast
16	The membrane around the vacuole is called	cytoplast	tonoplast	amyloplast	elaioplast	tonoplast
17	Identify the non -membraneous organelle from the following	Ribosome	Endoplasmic reticulam	Nucleus	Chloroplast	Ribosome
18	Plant cell wall mainly composed of	cellulose	starch	protein	lipid	cellulose
19	Cell wall of plant cell contains pit fields known as	Mitochondria	Golgi bodies	Plasma membrane	Plasmodesmata	Plasmodesmata
20	The prokaryotic cell contain	70s ribosome	60s ribosome	50s ribosome	80s ribosome	50s ribosome
21	The space between the nuclear envelop and nucleolus in filled by	Nucleic acids	Nucleoplasm	Nucleotides	Nucleoprotein	Nucleoplasm
22	Rough endoplasmic reticulum is embedded with	RNA	Ribosome	Protein	All the above	Ribosome
23	Prokaryotic cell Contain	Centriole	DNA	Nuclear	Ucleolus	DNA
24	The organism that look like bunches of grapes	Staphylococcus	Streptococcus	Micrococcus	Diplococcus	Staphylococcus
25	Plasma membrane is also called as	Plasmalemma	Periplasm	Cytoplasm	Cell wall	Plasmalemma
26	The fluid model was introduced by	S.J Singer and J.D Robertson	J.D Robertson	Jacob and Monad	S.J Singer and	S.J Singer and G.Nicolson.
27	--------- is the barrier, able to regulate what enters and exits the cell	cell membrane	Plasma menbrane	Lipid bilayer	All the above	All the above
28	The cell membrane consists of ----------of amphipathic lipids	three classes	four classes	five classes	six classes	three classes
29	---------- the type of lipid found in cell membrane	phospholipids	glycolipids	sterods	all the above	all the above
30	The majority of cases lipid in cell membrane is	phospholipids	glycolipids	sterods	all the above	phospholipids

31	The entire membrane is held together by---------- interaction of hydrophobic tails	co valent	non covalent	dipeptide	hydroxide	non covalent
32	The amount of fluid protein in cell membrane is	25%	50%	75%	100%	50%
33	The large amount of ---------- is responsible for various activities of cell membrane.	protein	lipids	amino acids	fatty acids	protein
34	How many types of protein are found in cell membrane	1	2	3	4	2
35	Bacteria is Classified in to gram positive and gram negative on the basis of----------	Cell wall	Nucleus	Mitochondria	Shape	Cell wall
36	Bacteria is Classified in to gram positive and gram negative on the basis of----------	Genes	Cell membrane	Ribosomes	Nucleus	Genes
37	---------- is a special type of nuclear	Mitosis	Meiosis	Both	Conjugation	Meiosis
38	---------- are example for	enzymes	hormones	both	none	both
39	The cell membrane contains ----------"tail" regions	hydrophobic	hydrophilic	thermophilic	mesophilic	hydrophobic
40	The cell membrane contains ----------"head" regions	hydrophobic	hydrophilic	thermophilic	mesophilic	hydrophilic
41	Lipid bilayer contains the cellular component approximately ---------- in	5nm	7nm	9nm	11nm	7nm
42	Cellular components of lipid bilayer is visualized by	SEM	TEM	DFM	EM	TEM
43	Which of the following is not the plasma membranes domains	protein-protein complexes	lipid rafts	Desmosomes	All	All

44	---------- is present in cell membrane to increase the nutrient uptake	cilia	flagella	microvilli	all the above	microvilli
45	Key role of cell membrane is to	cell function	cell potential	cell organels	cell structure	cell potential
46	---------- provides the shape of the cell	cytoskeleton	matrix	cytoplasm	organells	cytoskeleton
47	Receptor present in the cell membrane	electrical	chemical message	physical	thermal	chemical message
48	The mechanical tension in the membrane has effect the	rate of exchange	fuction	both	none of the above	both
49	---------- are similar to fats, but have only two fatty acids rather than	phospholipids	glycolipids	sterods	all the above	phospholipids
50	---------- are described as amphipathic in plasma membrane	lipids	proteins	steroids	all the above	proteins
51	---------- is a common component of animal cell membranes and functions to help stabilize the membrane	Glycerol	Cholesterol	triglycirides	Fat	Cholesterol
52	---------- Affects the fluidity of the membrane.	Temperature	PH	Osmolality	Pressure	Temperature
53	---------- Embedded in the fluid matrix of the lipid bilayer	Proteins	Fat	Carbohydrates	All	Proteins
54	---------- Of proteins have been found in the plasma membrane of red blood	20 kinds	30 kinds	40 kinds	50 kinds	50 kinds
55	----------is the carrier protein mediated transport of biochemicals	diffusion	facilitated diffusion	both a and b	passivetransport	facilitated diffusion
56	---------- requires cellular energy to transport biochemicals	active transpot	passive transport	both a and b	none of the above	active transpot
57	which type of transpot requires ATP	active transpot	passive transport	both a and b	none of the	active transpot

58	freeze - etching experiment demonstrate the	lipid bilayer	embeded proteins	both lipid layer and embeded	structure	both lipid layer and embeded proteins
59	Form active transport, molecules move against either an electrical or concentration gradient collectively	isoelectric gradient	isopycric gradient	electrochemical gradient	electromolecular gradient	electrochemical gradient
60	The active transport of ---------- across a cell membrane is generally carried out by transport proteins that are found in	small molecule	ions	both	large molecules	both
	Unit-II					
1	Microfilaments are composed mainly of a protein called	actin	tubulin	myosin	chitin	actin
2	Which of the following is associated with the golgi complex	Cristae	Cisternae	Annuli	Quatasomes	Cisternae
3	Smooth endoplasmic reticulam is the site of	protein synthesis	carbohydrate synthesis	amino acid synthesis	Lipid synthesis	Lipid synthesis
4	In higher plants, the shape of the chloroplast is	Discoid	cup shaped	girdle shaped	ribbon shaped	Discoid
5	Which of the following is possessed only by some bacteria, not by eukaryotes	Cilia	Flagella	Capsule	Centriole	Capsule
6	A major function of Golgi bodies or Dictyosomes is	modification of proteins	in fermentation	independent photosynthesis	in electron transport	in modification of proteins
7	Which of the following does not surrounded by a double membrane in eukaryotes	nucleus	mitochondria	chloroplasts	the cell	the cell
8	The organelles involved in protein transport	ER and Golgi	ER and mitochondria	Golgi and mitochondria	lysosomes and golgi	ER and Golgi
9	Transmembrane proteis are also known as	integral protein	Peripheral protein	Lipid anchord Protein	All the above	integral protein

10	G- protein is the example for ----------	integral protein	Peripheral protein	Lipid anchord	All the above	Lipid anchord Protein
11	.---------- are attached to integral membrane protein	integral protein	Peripheral protein	Lipid anchord Protein	All the above	Peripheral protein
12	Pore- like connections between adjacent cells is an example of	gap junction	desmosomes	tight junction	cell junction	gap junction
13	Desmosomes is composed of	fibers	soft tissue	hard tissue	Bones	fibers
14	Giulio Bizzozero, was an Italian pathologist who discovered	gap junction	desmosomes	tight junction	cell junction	desmosomes
15	Junction that prevents two cell compartments from mixing is	gap junction	desmosomes	tight junction	cell junction	tight junction
16	Gap junction allows the exchange of	solutions	solutes	solvent	Water only	solutes
17	Gap junctions are characteristic of	Axodendritic synapses	Electrical synapses	Dendrodendritic synapses	Axosomatic synapses	Electrical synapses
18	Synaptic signaling involves	endocrine signals	paracrine signals	autocrine signals	neurotransmitters	neurotransmitters
19	Cell surface receptors may be any of the following except	G protein linked	enzymic receptors	single-pass transmembrane proteins for neurotransmitte	chemically-gated ion channels	single-pass transmembrane proteins for neurotransmitters
20	In the cAMP pathway, the G protein stimulates	phospholipase C	adenylyl cyclase	the endoplasmic	calmodulin	adenylyl cyclase
21	The ____ surrounds the cell like a belt, preventing the passage of substances between the cells	gap junction	desmosome	hemidesmosome	tight junction	tight junction
22	The receptor for nitric oxide (NO) is	intercellular	intracellular	extracellular	ultracellular	intracellular
23	In desmosomes, cadherins link to _____ of an adjacent cell	integrins	connexons	intermediate filaments	ras proteins	intermediate filaments

24	Characteristics of intracellular receptors that regulate gene transcription include all of the following except:	a DNA binding site	an extracellular binding site	a transcription activating domain	may be signaled by lipid soluble	an extracellular binding site
25	In most cases protein kinases	hydrolyze proteins	bind cGMP	adenylyl cyclase	phosphate groups to	add phosphate groups to proteins
26	_____ junctions may protect a damaged cell through chemical gating	tight	Gap	adherens	occluding	gap junction
27	In paracrine signalling, the signalling molecules affect only	target cell is close to the	target cell is away from the signal	target cell and signal secreted	all the above	target cell is close to the signal secreted cel
28	Simple nerve reflexes use signaling molecules called	endocrine signals	paracrine signals	autocrine signals	neurotransmitters	neurotransmitters
29	Which of the folloeing is not a type of signalling molecules?	Harmones	Insulin	tyroxin	Adenylate cyclase	Adenylate cyclase
30	When a cell releases a signal molecule into the environment and a number of cells in the immediate vicinity respond, this type of signaling is	autocrine signaling	paracrine signaling	endocrine signaling	synaptic signaling	paracrine signaling
31	Synaptic signaling between adjacent neurons is like hormone signaling in which of the following ways?	it sends its signal molecules	it sends its signal molecules quite a distance.	It requires calcium ions.	it requires binding of a signalingmole	it requires binding of a signalingmolecule to a recepto
32	A small molecule that specifically binds to another molecule, usually a larger one	is called a signal	is called a ligand	is called a polymer	seldom is involved in	is called a ligand
33	Of the following, a receptor protein in a membrane that recognizes a chemical signal is most similar to	of an allosteric enzyme in the cytoplasm that binds to a specific substrate	RNA specifying the amino acids in a polypeptide	metabolic pathway operating within a specific organelle.	genes making up a chromosome	the active site of an allosteric enzyme in the cytoplasm that binds to a specific substrate
34	Thyroid hormones bind to _____ receptors	G-protein-linked	tyrosine-kinase	plasma membrane ion-channel	intracellular	intracellular

35	Membrane receptors that attach phosphates to specific animo acids in proteins are	not found in humans	called receptor tyrosine-kinases.	G-protein signal receptors.	with several bacterial diseases in	called receptor tyrosine-kinases.
36	Up to 60% of all medicines used today exert their effects by influencing what structures in the cell membrane?	tyrosine-kinases receptors	ligand-gated ion channel receptors	G proteins	cholesterol	G proteins
37	Which of the following are chemical messengers that pass through the plasma membrane of cells & have receptor molecules in the cytoplasm?	Insulin	Testosterone	cAMP	Ephinephrine	Testosterone
38	The receptors for a group of signaling molecules known as growth factors are often	igand-gated ion channels	igand-gated ion channels	cyclic AMP	receptor tyrosine kinases	receptor tyrosine kinases
39	In general, a signal transmitted via phosphorylation of a series of proteins	brings a conformational change to	requires binding of a hormone to a cytosol recepto	requires phosphorylase activity.	allows target cells to change their	brings a conformational change to eachprotein
40	The general name for an enzyme that transfers phosphate groups from ATP to a protein is	phosphorylase	protein kinase	protease	ATPase.	protein kinase
41	The toxin of Vibrio cholerae causes profuse diarrhea because it	protein involved in	adenylyl cyclase and triggers the	trisphosphate to become a	calmodulin and activates	involved in regulating salt and water secretion
42	Which of the following would be inhibited by a drug that specifically blocks the addition of phosphate groups	G-protein-linked receptor	ligand-gated ion channel signaling	phosphatase activity	receptor tyrosine kinase	receptor tyrosine kinase activity
43	Which of the following most likely would be an immediate result of growth factor binding to its receptor?	G-protein-linked receptor	ligand-gated ion channel signaling	protein kinase activity	phosphatase activity	protein kinase activity
44	Adenylyl cyclase has the opposite effect of which of the following?	protein kinase	protein phosphatase	phosphodiesterase	phosphorylase	phosphodiesterase

45	phosphodiesterase. Therefore, the cells of a person who has recently consumed	phosphorylated proteins	GTP	cAMP	adenylyl cyclase	cAMP
46	Synaptic signaling between adjacent neurons is like hormone signaling in which of the following ways?	It sends its signal molecules	It sends its signal molecules quite a distance	It requires calcium ions.	It requires binding of a signalingmole	It requires binding of a signalingmolecule to a receptor
47	From the perspective of the cell receiving the message, the three stages of cell signaling are	the paracrine, local, and synaptic stages.	signal reception, signal transduction, and cellular response.	signal reception, nucleus disintegration,	signal reception, cellular response, and	signal reception, signal transduction, and cellular response.
48	The process of transduction usually begins	chemical signal is	molecule changes the receptor	after the target cell divide	hormone is released from	molecule changes the receptor protein in
49	Which of the protein has carbohydrate attached to it	Glycoprotein	Lipoprotein	Cannel protein	carrier protein	Glycoprotein
50	The ABO glycoproteins expressed on	Glycoprotein	Lipoprotein	Cannel protein	Receptor	Receptor protein
51	Cystic fibrosis can result from a failure of a _____ protein	Cannel	carrier	transport	enzymatic	Cannel
52	A protein that combines with a substance and helps to move it across the memberane is called	Glycoprotein	Lipoprotein	Cannel protein	carrier protein	carrier protein
53	the most abudant membrane lipids are	phospholipids	cholestrol	glycolipid	spingolipid	phospholipids
54	membrane _____ serve as self-identity markers that play important roles in recognition of self and in cell to cell interactions	lipids	Protein	Carbohydrates	lipo proteins	Carbohydrates
55	A base is changed by the repositioning of a hydrogen atom.	tautomerism	depurination	deamination	transition	transition
56	Loss of a purine base (A or G)	tautomerism	depurination	deamination	transition	depurination
57	A purine changes to another purine, or a pyrimidine to a pyrimidine	tautomerism	depurination	deamination	transition	transition

58	which of the following is an alkylating agent	NTG	BU	NH2OH	HA	NTG
59	Size of plasmids varies from	1 to over	1 to 400 kb	1 to 600kb	1 to 1000kb	1 to 400 kb
60	plasmids are associated with	transducfermat	conjugation	reproduction	all the above	all the above
	Unit -III					
1	The subunit of prokaryotic ribosomes are	60 S+40 S	70 S+30 S	60 S+30 S	50 S+30 S	50 S+30 S
2	The main function of Centrosome is	Secretion	osmoregulation	Protein synthesis	Formation of spindle fibre	Formation of spindle fibre
3	Assembly of two subunits 40 S and 60 S of the ribosome is	100 S unit	80 S unit	70 S unit	90 S unit	80 S unit
4	Which of the following does not occur within mitochondria	Krebs cycle	Glycolysis	transport system	ATP synthesis	Glycolysis
5	The "brain" of cell is	Cytoplasm	Nucleus	Mitochondria	membrane	Nucleus
6	"Zone of exclusion" is associated wit	Nucleus	Golgi bodies	Mitochondria	membrane	Golgi bodies
7	Glyoxysomes occur in	Plant cells	Animal cells	Both A and B	None of these	Plant cells
8	RNA molecules that exhibit catalytic activity are called	M RNA	Ribosomes	Ribozymes	Ribonucleotides	Ribozymes
9	The term gene was introduced by-	Sutton	Muller	Johenson	Bridges	Johenson
10	Chemically gene is formed by a	DNA	RNA	Both	None of the	DNA
11	Virus –mediated transfer of cellular genetic material from one bacterial cell to another by means of virus particles is called	Induction	Transfection	Transformation	Transduction.	Transduction.
12	Introns are present in the genes of-	Prokaryote	Eukaryotes	Both	None of the	Eukaryotes
13	In the classical model of transcriptional control described by Jacob and Monod, a repressor protein binds to	Enhancer	AUG sequence	Operator	Ribosome binding site	Operator
14	Phage can multiply only in a	Bacterium	Viruses	Metabolizing bacterium	Non-Metabolizing	Bacterium

15	Which of the following best describe the function of the sigma subunit in the RNA Polymerase of E.Coli?	Essential for elongation of the RNA transcript	Recognition and binding to the promoter sequence	For transcription termination	Increase RNA Polymerase binding to	Recognition and binding to the promoter sequence
16	Which of the following is not involved in the processing of mRNA precursors in eukaryotic cells	Capping of the 5'end	Splicing of exons	Addition of Pol A	Excision of introns.	Splicing of exons
17	130.Genes mainly produce ------	Enzymes	Elements	Vitamins	Essential	Enzymes
18	Single standered circular DNA is present in	j/174 bacteriophages	all viruses	Animal viruses	Plant viruses	j/174 bacteriophages
19	The ribosome is involved in all of the following expect	Peptide bond formation	Aminoacylation of t RNA	Binding of aminoacyl t	Binding of rho facter	Binding of rho facter
20	An E.Coli strain lacking DNA polymerase I would be deficient in DNA	Repair	Splicing	Methylation	Transcripition	Repair
21	According to central concept of genetics genes are called as	Biological information	Hereditary vehicles	Chromosomes	All the above	All the above
22	Maleness in bacteria is called donors	F- strain	F+ strain	HFr	None of the above	F+ strain
23	Recombination of genes in bacteria involves	Transfer of genetic material from one bacteria cell to another	With in bacteria	With one plasmid to another plasmid	None of the above	Transfer of genetic material from one bacteria cell to another
24	Lederberg's experiment depicting	Three different method it recombination in bacteria	Transformation	Transduction	Conjugation	Three different method it recombination in bacteria
25	The term nucleoid otherwise called as	Bacterial chromosome	Viral chromosome	Human chromosome	All the above	Bacterial chromosome

26	In rolling circle replication single stranded viral DNA otherwise called as	Positive strand	Negative strand	Replicative strand	Rolling strand	Positive strand
27	The DNA replication is semi conservative. It was proved by	Messelson-stalks	Okazaki	Lehniger	Lorunberg	Messelson-stalks
28	The cap m RNA is formed by	Methyl cytosine	7-methyl guanosine	5- methyl guanosine	None of the above.	Methyl cytosine
29	The backbone of nucleic acid structure	Hydrogen bond	Phosphodiester bond	Ionic bond	None of the above	Hydrogen bond
30	The planes of base pairs are fitted at 23 with respect to its helix is	A –DNA	B-DNA	C DNA	2-DNA	A –DNA
31	Z-DNA form was discovered by	Watson and Crick	Hoogsteen	Chargaft	Wang and Rick	Wang and Rick
32	DNA Polymerase I has	5-3 Exonuclease	Proof reading activity	Polymerising activity	All the above	Polymerising activity
33	Okazaki fragment are the repeating prime in	Leading strand	Lagging strand	both leading and lagging	polypeptides	Lagging strand
34	Function of helicases	Excise introns from the RNA	Polymerize nucleotide to form RNA	Protect bacteria from foreign DNA	Unwinding the DNA.	Unwinding the DNA.
35	Which of the following catalize the formation of negative coils during DNA replication?	helicases	gyrases	primosome	polymerase	gyrases
36	The base not found in RNA is	Adenine	Guanine	Cytosine	Thymine	Adenine
37	In Hershey and Chase experiment S^{35} is incorporated in formation of----------	DNA	Proteins	Cytoplasm	All the above	Proteins
38	RNA contain Uracil in place of	A	C	G	T	T
39	The proteins associated with eukaryotic DNA are	Tryptones	Histones	Deoxyribose	None	Histones
40	The enzyme DNA photolyase is involved	Park repair	SOS repair	Photo reactivation	Recombination	Photo reactivation

41	The Lac Z gene encodes the enzyme	B-Galactosidase	B-Galactosidase	Protease	Galactose	B-Galactosidase transacetylase
42	An enzyme that contain 7 different polypeptides	RNA pol	Reverse transcriptase	DNA pol I	DNA pol III	Reverse transcriptase
43	A segment of mRNA before the region encoding the first polypeptide chain in	Leader	Spacer	Intron	Exon	Spacer
44	In all plants the mono hybrid ration in	9:3:3:1	1:2:1	15:1	9:7	1:2:1
45	---------- Subunit of RNA Polymerase in required for promoter binding	α Subunit	β Subunit	σ Subunit	μ Subunit	σ Subunit
46	RNA has the following bases	ATG	AUG	APG	ATT	AUG
47	The non-coding sequence found in chromosomes	Introns	Exons	Amber	All	Introns
48	Kornberg enzyme is	DNA Pol I	DNA pol II	DNA pol III	RNA	DNA Pol I
49	RNA is made up of	Triple helical structure	Double helical structure	Single helical structure	None of the above	None of the above
50	Thymine is the characteristic base of	RNA	DNA	Vitamin	None of the	DNA
51	Hershey and Chase experiment on----------	T2-Phages	Salmonella	Amoebae	Bacterium	T2-Phages
52	In Hershey and Chase experiment P^{35} is incorporated in formation of----------	DNA	Amino Acids	Cytoplasm	All the above	DNA
53	Conjugation is initiated, via a mating signal, a complex of proteins called	relaxosome	peroxisomes	cell signaling	none of the above	relaxosome
54	Transfer of genetic material between bacteria through cell-to-cell contact is	transduction	Conjugation	Transfermation	Budding	Conjugation
55	Structural genes that are required for transcription initiation in prokaryotes	Pribnow box	Hogness box	TAAAT box	CC AAT box	Pribnow box
56	In E coli conjugation transfer of the entire bacterial chromosome takes	100 minutes	150 minutes	200 minutes	250 minutes	100 minutes

57	The simplest unit of DNA in	Recon	Muton	Both a and b	Cistron.	Recon
58	The genetic material of phages are	ssRNA	dsRNA	ssDNA	all the above	ssDNA
59	A sequence of nucleotide triplets that lacks a termination codon	Open reading frame	Operator	Oncogene	Mutagen	Operator
60	The synthesis of RNA primers catalyzed by a	helicases	primases	polymerases	gyrases	primases
	Unit-V					
1	Cancer cells are…	a) BHK	b) Veo	c) HL-8	d) Hela cells	d) Hela cells
2	Cancer is caused by….	uncontrolled mitosis	b) uncontrolled meiosis	c) rupturing of cells	immunity of the cells	a) uncontrolled mitosis
3	Cancer cells can easily be destroyed by radiations due….	a) fast mutation	b) rapid cell division	c) lack of mutation	d) lack of oxygen	b) rapid cell division
4	Reason of lung cancer is	a) coal mining	b) cement factory	fluoride	mining	a) coal mining
5	Which on of the following is used in treatment of thyroid cancer?	a) U-238	b) I-131	c) C-14	d) rA-240	b) I-131
6	Migration of cancerous cells from the site of origin to other part of the body forming secondary tumours is called	a) diapedesis	b) metastasis	c) proliferation	d) none of these	b) metastasis
7	A patient is suspicious of having breast cancer. What type of test will a physician conduct to diagnose the cancer	a) blood test	b) pap test	c) CT scan	d) mammography	d) mammography
8	Which one of the following genes is involved in the conversion of proto-oncogenes into oncogenes causing cancer?	a) metastasis genes	b) angiogenesis genes	c) transposons	d) tumour suppressor genes	d) tumour suppressor genes

9	Which one of the following therapies will involve only the cancerous cells not the normal cells in treatment	a) immunotherapy	b) surgery	c) aromatherapy	d) chemotherapy	a) immunotherapy
10	Which one of the following cancers does not form a solid neoplasm	a) leukemia	b) lymphoma	c) lipoma	d) sarcoma	a) leukemia
11	Cancer may start when	number of chromosomes become defective	b) changes occur in genes controlling cell divisions	c) when loci start to change in chromosomes	d)All of above	b) changes occur in genes controlling cell divisions
12	Carcinogens	A. may not occur naturally	B. are all man made	C. are all caused due to ionic radiation	caused due to tar of tobacco smoke	D. may be caused due to tar of tobacco smoke
13	Any agent that causes cancer is called	A. mutagen	B. carcinogen	C. oncogene	above	B. carcinogen
14	Cancer is caused due to	A. controlled mitosis	B. uncontrolled mitosis	C. controlled meiosis	D. exposure to stress	
15	Naturally occur carcinogens include	A. asbestos	B. certain dioxins	C. aflatoxin	dyes	
16	Recombination termed as	Linkage	Crossing over	Alleles	All the above	Crossing over
17	---------- is mRNA from DNA	Translation	Transcription	Regulation	None of the above	Transcription
18	---------Amino acid initiate the polypeptide synthesis	Methionine	Valine	Alanine	Aginine	Methionine
19	Which of the following is an initiation codon	AUG	ATT	AAA	TAA	AUG
20	Termination of polypeptide chain occur by ---- codon	UAA	UAG	UGA	All	All
21	Bacterial transformation is ----------	Griffith effect	Averys effect	Hershey effect	All	Griffith effect

22	Genetic recombination in bacteria discovered by -----	Lederberg and tautum	Hershey and chase	Avery and Mccarthy	Griffith	Lederberg and tautum
23	---------- create variation in gene pool	replication	Mutation	DNA repair	None of the above	Mutation
24	The ----------is abiological assay to assess the mutagenic potential of chemical compounds	Ames test	Luria bertani test	Averys test	Mc Cornkeys test	Ames test
25	In histidine free media ---------- organish can synthesis histodine	Mutated	tester	Sensitive	Recessive	Mutated
26	Ames test is a----------	Biochemical assay	Biological assay	Immunological assay	None of the above	Biological assay
27	Luria-Delbruck experiment also called as	Fluctuation Test	Fraction test	Biochemical test	None of the above	Fluctuation Test
28	---------- mend most changes before they become permanent mutations	replication	Mutation	DNA repair	None of the above	DNA repair
29	Exchange of a single nucleotide (A ↔ G) or (C ↔ T).is ----------	transition	transversion	both	none of the above	transition
30	An example of a transversion is ---------	(A ↔C)	(A ↔ G)	(C ↔ T).	all the above	(A ↔C)
31	Point mutation which code for the same amino acid is	missense mutation	nonsense mutation	silent mutation	all the above	silent mutation
32	Point mutation which code for the differnt amino acid is	missense mutation	nonsense mutation	silent mutation	all the above	missense mutation
33	Point mutation which code for a stop and truncate the protein is	missense mutation	nonsense mutation	Silent mutation	all the above	nonsense mutation
34	Mutation occur in non coding region is ----------	missense mutation	nonsense mutation	Silent mutation	all the above	Silent mutation

35	Poly A tail sequence involved in eukaryotic translation is	100 nucleotide long	200 nucleotide long	300 nucleotide long	500 nucleotide long	200 nucleotide long
36	How many nucleotide require for a	One	two	three	four	three
37	Thymine dimmer formation as result of	UV radiation	Ionizing radiation	Base` analog	Alkylating	UV radiation
38	In which type of repair involves sequence of enzyme catalyzed steps	Excision repair	Photo reactivation	Dark repair	All the above	Excision repair
39	Direct evidence for the spontaneous origin of T1 –r mutation in E.Coli cells not exposed to phage is called	Replica plating	Fluctuation test	Mutation rate	All the above	Fluctuation test
40	Which of the following is a base analog	Nitrous acid	5 – Bromo uracil	EES	EMS	5 – Bromo uracil
41	Ethyl methane sulfonate in a potent ----------	Alkglating agent	Base analogue	Intercalating agent	All the above.	Alkglating agent
42	Which is not a chemical mutagen	HNO2	EMS	HA	GC	GC
43	The process in which wild –type phenotype is regained	Back mutation	Reverse mutation	Reversion	All the above.	Reverse mutation
44	The common form of thymine is	enol	keto	Amino	Imino	keto
45	The enzyme photolyase involved in an enzymatic reaction is called	Photo reactivation	Photosynthesis	Phosphorylation	Reverse mutation	Reverse mutation
46	Liquid –holding recovery in now Known to be a manifestation of a general phenomenon called	Dark repair	Light repair	Both	None of the above	Dark repair
47	The biochemical mechanism tolerance of the damage is	SOS repair	Excision repair	Recombination	All the above.	SOS repair
48	-------- Includes a bypass system that allows DNA chain growth across damaged segment at the cost of fidelity of replication	SOS repair	Excision repair	Recombination Excision repair	Incision repair	SOS repair

49	The test for reversion as a means of defecting mutagens and carcinogens is	Ames test	VPRL test	Fluctuation test	All the above.	Ames test
50	DNA that carries information necessary for protein synthesis from the DNA to the Ribosome	mRNA	tRNA	rRNA	All the above	mRNA
51	In genetics, suppression of a mutation refers to	Restriction of the original phenotype due to a second	Restriction of the original DNA sequence by mutation	Prevention of expression of the mutant gene by	Appearance of the recessive phenotype in	Restriction of the original DNA sequence by mutation
52	Common lesions found in DNA after exposure to ultraviolet light	Pyrimidine dimers	Single strand breaks	Base deletion	Purine dimers.	Pyrimidine dimers
53	During mutation substitution of Adenine by Guanine is known as	Transition	Transversion	Deletion	Transposition s	Transition
54	---------- Mutation leads to the death of cell	Lethal	Point	Frame shift	Missense	Lethal
55	Which of the following leads to frame shift mutation	base analog	ionization	uv radiation	5 BU	uv radiation
56	Which one is an mutagen	EMS	EES	NTG	All the	EMS
57	Mutation can be caused by---------	Virus	Mutagens	Radiation	All the above	All the above
58	AT repeats are the examples of	insertion	deletion	transversion	translocation	insertion
59	A base is changed by the repositioning of a hydrogen atom.	tautomerism	depurination	deamination	transition	transition
60	Loss of a purine base (A or G)	tautomerism	depurination	deamination	transition	depurination