SYLLABUS

B.Sc II SEM

SUBJECT: CELL BIOLOGY AND METABOLISM

Unit I	**Topic** **Discovery of cell and cell theory** **Comparison of Prokaryotic and eukaryotic cells** **Cell division an cell cycle** **Anomalies in cell Division** **Animal cell and plant cell**
Unit II	**Cell synchrony and its application** **Cell- cell interaction** **Eukaryotic genome- chromosomal organisation** **chromosomal structure** **Euchromatin, Heterochromatin, Centromere** **Telomere, Definition of- introns/exons** **Chromatin structure(nucleosomes)- histon, non-histon proteins** **Regulatory sequences** **Promoters, enhensers**
Unit III	**Strcture of prokaryotic gene** **Prokaryotic transcription and regulation** **Eukaryotic transcription** **Transcription factors and regulation** **Post -transcriptional modification- 5'-cap formation**

	3'- end processing **Polyadenylation** **Gene splicing**
Unit IV	**Translation in prokaryotes and Eukaryotes** **Mechenisms of initiation** **Elongation and Termination** **Co and post Translation modification** **Regulation of gene expression in prokaryotes** **Induction** **Repression** **Operon models- lac operon** **his and ara operon** **trp operon, Catabolite repression**
Unit V	**DNA damage and repair** **Mutation, Genetic code** **Post-transcriptional modification and transport of proteins** **Sprotein synthesis** **Insertion elements and transposons** **IS elements, Tn3 elements** **Yeast TY family** **P Extra chromosomal DNAin prokaryotes-** **Plasmids**

UNIT-I

Q.1 Cell Theory: Cell theory refers to the idea that cells are the basic unit of structure in every living thing. Development of this theory during the mid 17th century was made possible by advances in microscopy. This theory is one of the foundations of biology. The theory says that new cells are formed from other existing cells, and that the cell is a fundamental unit of structure, function and organization in all living organisms.

History Drawing of the structure of cork by Robert Hooke that appeared in Micrographia The cell was discovered by Robert Hooke in 1665. He examined (under a coarse, compound microscope) very thin slices of cork and saw a multitude of tiny pores that he remarked looked like the walled compartments of a honeycomb. Because of this association, Hooke called them cells, the name they still bear. However, Hooke did not know their real structure or function.[1] Hooke's description of these cells (which were actually non-living cell walls) was published in Micrographia. [2] . His cell observations gave no indication of the nucleus and other organelles found in most living cells. The first man to witness a live cell under a microscope was Antony van Leeuwenhoek (although the first man to make a compound microscope was Zacharias Janssen), who in 1674 described the algae Spirogyra and named the moving organisms animalcules, meaning "little animals".[3] . Leeuwenhoek probably also saw bacteria. [4] Cell theory was in contrast to the vitalism theories proposed before the discovery of cells.

Classical interpretation

1. All living organisms are made up of one or more cells. 2. Cells are the basic unit of life. 3. All cells arise from pre-existing cells. (omni cellulae e cellula) 4. The cell is the unit of structure, physiology, and organization in living things. 5. The cell retains a dual existence as a distinct entity and a building block in the construction of organisms. Modern interpretation.

The generally accepted parts of modern cell theory include:

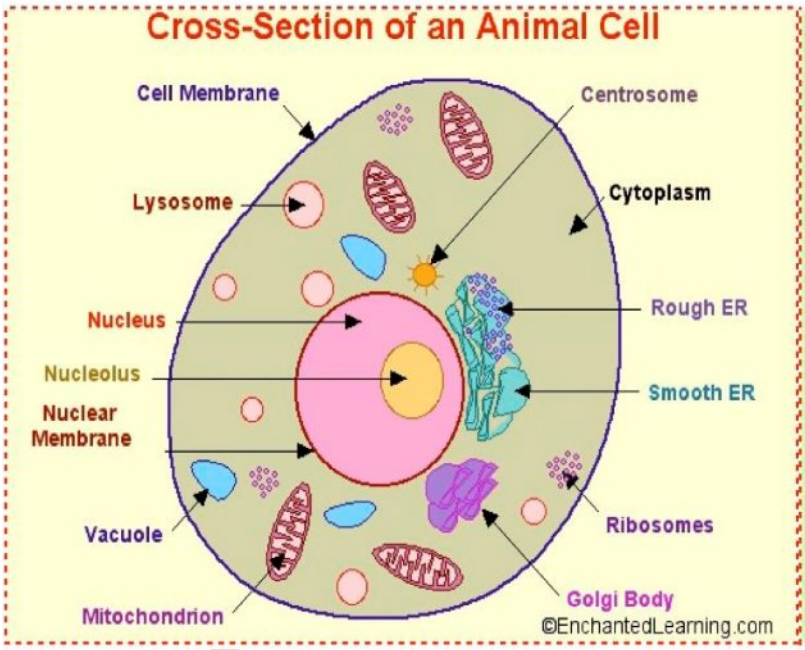
1. The cell is the fundamental unit of structure and function in living organisms. 2. All cells arise from pre-existing cells by division. 3. Energy flow (metabolism and biochemistry) occurs within cells. 4. Cells contain hereditary information (DNA) which is passed from cell to cell during cell division. 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.

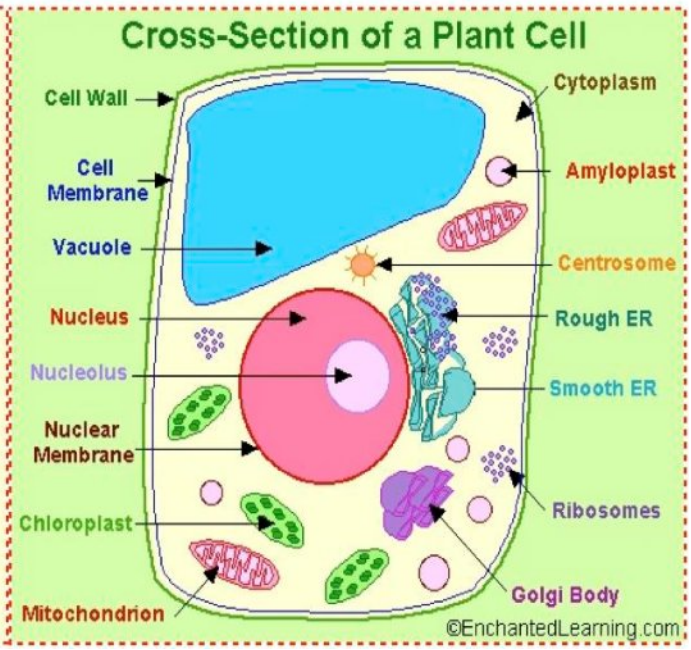
Exceptions

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 imprecise. 3. Mitochondria and chloroplasts have their own genetic material, and reproduce independently from the rest of the cell.

Q.2Difference between Animal cell and Plant cell:

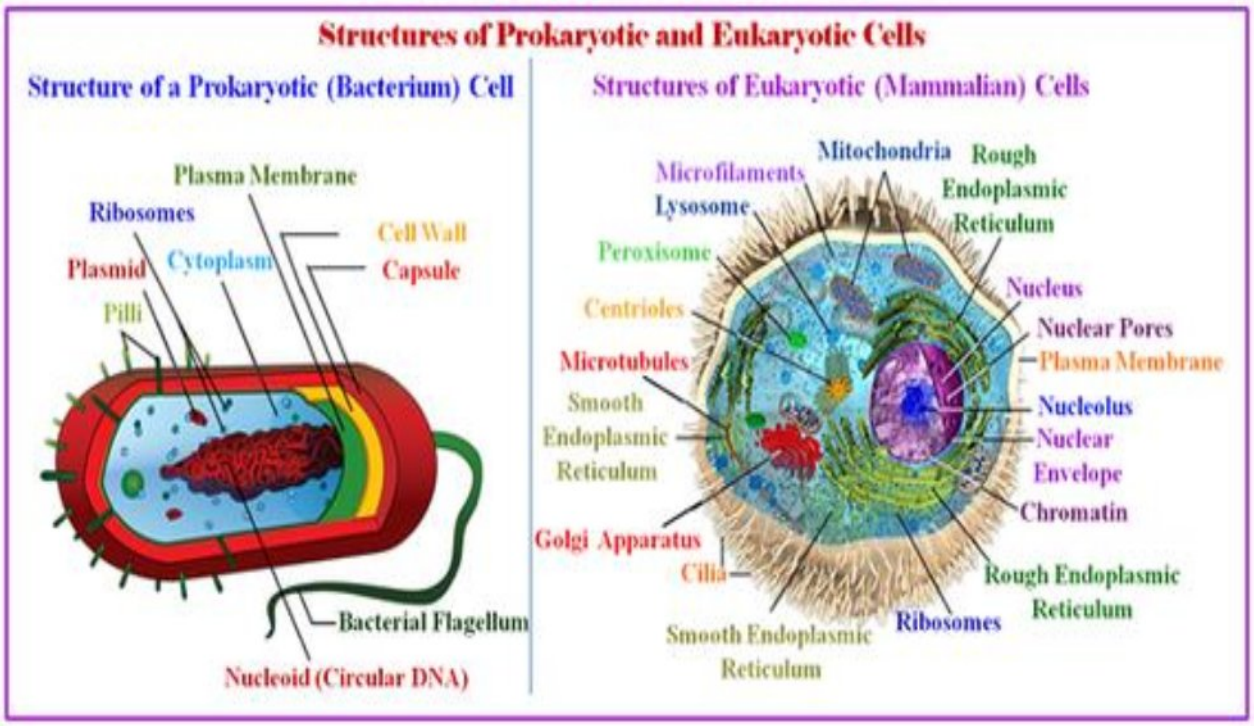
Animal Cell	Plant Cell
1. Cell wall absent. Cellulose in any form is also absent.	Cellulose cell wall is present in plant cells.
2. Cytoplasm is denser, more granular and occupies most of the space in the cell.	Cytoplasm is pushed to the periphery and forms a thin lining against the cell wall.
3. Vacuoles absent. If present, they are small, temporary and concerned with excretion or secretion.	Vacuoles are large and prominent. Maybe one or more.
4. Plastids are absent.	Plastids are generally present.
5. Centrosome is present with one or two centrioles.	Centrosome is absent but two small clear areas called polar caps are present. These participate in cell division.
6. Prominent and highly complex Golgi bodies present near the nucleus.	Several subunits of Golgi apparatus called dictyosomes present.
7. Reserve food stored in the form of **glycogen.**	Reserve food stored in the form of **starch.**





Q.3 Difference between Prokaryotic cell and Eukaryotic cell:

Prokaryotic cell	Eukaryotic cell
Most prokaryotic cells are unicellular.	Most eukaryotic cells are multicellular.
Size of the cell is generally small (0.5-5 µm).	Size of the cell is generally large (50- 100 µm).
Nuclear region is poorly defined due to the absence of a nuclear membrane or the cell lacks true nucleus.	Nuclear region is well-defined and is surrounded by a nuclear membrane, or true nucleus bound by a nuclear membrane is present in the cell.
It contains a single chromosome.	It contains more than one chromosome.
Nucleolus is absent.	Nucleolus is present.
Membrane-bound cell organelles such as plastids, mitochondria, endoplasmic reticulum, Golgi apparatus, etc. are absent.	Cell organelles such as mitochondria, plastids, endoplasmic reticulum, Golgi apparatus, lysosomes, etc. are present.
Cell division occurs only by mitosis.	Cell division occurs by mitosis and meiosis.
Prokaryotic cells are found in bacteria and blue-green algae.	Eukaryotic cells are found in fungi, plants, and animal cells.



Q.4 Cell Division and cell cycle:

Phases of Cell Cycle

There are four distinct phases in the cell cycle - G_1 phase, S phase (synthesis), G_2phase and M phase. The G_1, S phase and G_2 phase together are known as interphase. The M phase or the mitotic phase is of two processes, one where the chromosomes of the cell is divided between two sister cells and the other is cytokinesis where the cell's cytoplasm divides into half forming two distinct cells.

The cells that have stopped dividing temporarily or reversibly are said to be in the state of quiescence called G_0 phase. Progression from phase to another depends on the proper completion of the previous one. After the process of cell division, the daughter cells begin the interphase of the new cycle. The stages of interphase are not morphologically distinguishable, yet each phase has a distinct biochemical process that prepares the cell for initiation of cell division.

G_0 Phase

Sometimes the cells in the quiescent and senescent cells are referred to as post mitotic. The cells which are indivisible in multicellular eukaryotes generally enter the quiescent G_0 state from G_1. They may remain in the quiescent state for long periods of time. This state can be for indefinite like in neurons and is very common in cells that are fully differentiated. Death of the cells in response to damage of DNA or degradation would make the progeny of the cells nonviable. Some cells like the cells of liver and kidney enter the G_0 phase semi-permanently.

Interphase

Earlier to to the cell division process, the cell needs to accumulate nutrients. During the interphase all the preparations are done. In interphase of a newly formed cell, a series of changes takes place in the cell and the nucleus, before it is capable of division. This phase is also known as intermitosis. Earlier this stage was known as resting stage because no remarkable activiyt realated to cell division takes place here. Interphase proceeds in a series of three stages, G_1,S, and G_2. Division of cell operates in a cycle, hence the interphase of the cycle is preceded by the previous cycle of M phase and cytokinesis. Interphase is also called the preparatory phase. In the interphase stage the division of nucleus and cytosol does not occur. The cell prepares for division. This is a stage between the end of mitosis and start of the next phase. Many events occur in this stage and most significant event that occurs is the replication of genetic material.

G_1 Phase

This is the first phase in the interphase. From the end of the previous M phase till the beginning of the DNA synthesis in the next cycle is called the G_1 phase, here G indicates gap. This phase is also called growth phase. In this phase the biosynthetic activities of the cell, which shows a considerable slow down during the M phase of resumes it activities at a high rate. In this phase there is a marked production of proteins by the use of 20 amino acids. Also enzymes that are required in S phase needed during DNA replication. The duration of the G_1 phase is highly variable, also among different cells of the same species. The G_1 phase is under the control of the p53 gene.

S phase

The start of the S phase is when the DNA replication commences. When the phase completes all the chromosomes have been replicated. Each chromosome has two sister chromatids. During this phase, the amount of DNA in the cell is doubled but the ploidy of the cell remains unchanged. In this phase the synthesis is completed as soon as possible as the exposed base pairs are sensitive to external factors like drugs or mutagens.

G_2 phase

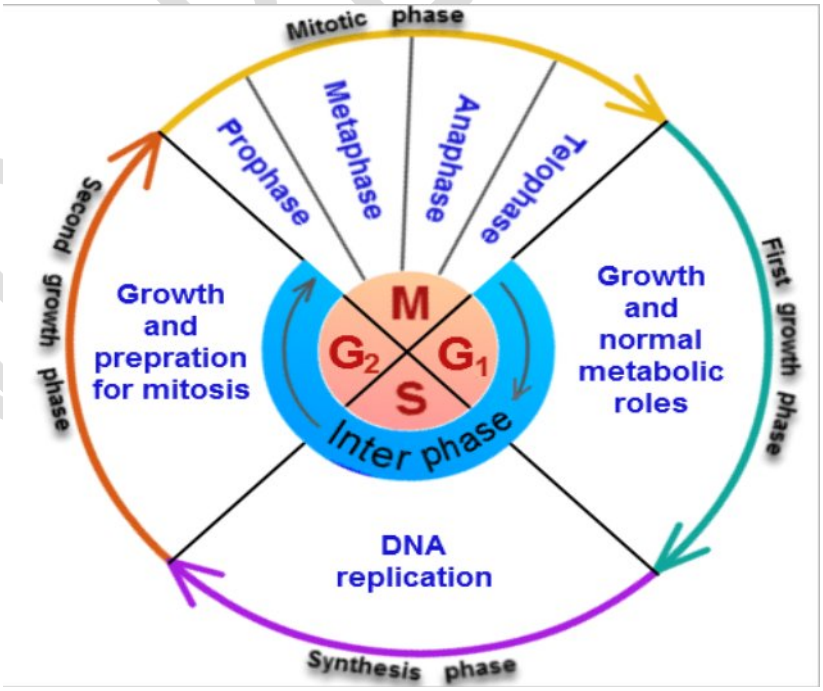
It is again the gap phase which happens during the gap between the DNA synthesis and mitosis. During this phase the cell will continue to grow. The G_2 checkpoint mechanism controls to ensure that the cell is ready to enter the M (mitosis) phase and divides.

Mitosis or M phase

The M phase consists of karyokinesis - nuclear division. The M phase is of several distinct phases, known as

Prophase,
Metaphase,
Anaphase,
Telophase,
Cytokinesis.

The process of mitosis takes place only in eukaryotes, the chromosomes in the nucleus of the cell into two identical nucleus. This stage is followed by cytokinesis. In cytokinesis the cell, nuclei, cytoplasm, organelles and cell membrane is divided into two equal shares. Mitosis and cytokinesis together make the mitotic (M) phase of the cell cycle. The mother cell divides into two daughter cells that are genetically identical to each other. Mitosis is seen only in eukaryotic cells, but it occurs in different ways in different species. The process of mitosis is a sequence of events divided into three stages - prophase, metaphase, anaphase and telophase. During this process of mitosis the chromosome pairs condense and they attach to fibres that pull sister chromatids to opposite sides of the cell. The cell with the process of cytokinesis produces two identical daughter cells.



Q.5 Anomalies in cell division:

Principal Meiotic Events and Outcomes of Their Failures

Stage	Meiotic events	Results of unsuccessful completion of meiotic events
Leptotene	Chromosomes become visible; lateral elements begin to form	Germ cell degeneration; sometimes nondisjunction
Zygotene	Chromosomes form bouquet; each chromosome pairs with its lateral element; homologous lateral elements unite into a synaptonemal complex, which completes the pairing	Germ cell degeneration; sometimes nondisjunction
Pachytene, early	Recombination nodules attach to the central elements	No crossing over; chromosomes remain univalent
Pachytene, late	During crossing over, recombination nodules change into bars	Because of lack of chiasmata, bivalents fall into univalents
Diplotene	Homologues repel each other until they are held together only at the chiasmata	More univalents visible than in earlier stages
Metaphase I, Anaphase I, Metaphase II, Anaphase II	Orderly segregation of chromosomes is prerequisite for regular gametogenesis	Univalents may undergo nondisjunction, loss, or misdivision, spindle abnormalities interfere with chromosome segregation

UNIT-II

Q.1 Cell synchronization

Cell Synchronization is a process by which cells at different stages or the cell cycle in a culture are brought to the same phase. "Cell synchrony" is required to study the progression of cells through the cell cycle.

The types of synchronizations arc broadly categorized into two groups:

I) "Physical Fractionation"

2) "Chemical Blockade"

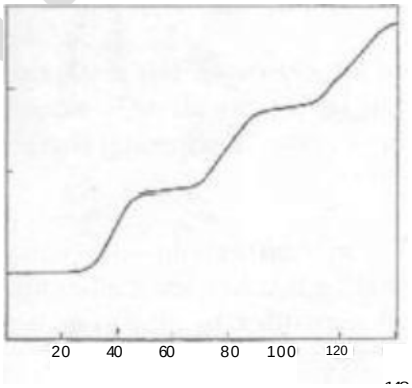
Cell separation by physical means

Physical fractionation or cell separation techniques, based on the following characteristics are in use.

- Cell density
- Cell size
- Affinity of antibodies on cell surface epitopes.
- Light scatter or fluorescent emission by labeled cells.

The two commonly used techniques arc:

Centrifugal separation



The physical characteristics — cell size and sedimentation velocity are operative in the technique of centrifugal elutriation. Centrifugal elutriator (from Beckman) is an advanced device lir increasing the sedimentation rate so that the yield and resolution of cells is better.

The cell separation is carried out in a specially designed centrifuge and rotor.

Fluorescence-activated cell sorting

Fluorescence-activated cell sorting (FACS) is a technique for sorting out the cells based on the differences that can he detected by light scatter (e.g. cell size) or fluorescence emission thy penetrated DNA, RNA, proteins, and antigens). The procedure involves passing of a 2 single stream of cells through a laser beam so that the scattered light from the cells can be detected and recorded. There are two instruments in use based on its principle:

a) Flow cytometer

h) Fluorescence-activated cell sorter

Cell separation by chemical blockade

The cells can be separated by blocking metabolic reactions. Two types of metabolic blockades are in use:

Inhibition of DNA synthesis

During the S phase of cell cycle, DNA synthesis can be inhibited by using inhibitors such as thymidine, amino protein, and hydroxyurea and cytosine arabinoside. The effects of these inhibitors ace variable. The cell cycle is predominantly blocked in S phase that results in viable cells. •

Nutritional deprivation

Elimination of serum from the culture medium for about 24 hours results in the accumulation of cells at GI phase. This effect of nutritional deprivation can be restored by their addition by which time the cell synchrony

Occurs

Example of cell synchrony

Synchronous Growth of Bacteria

Studying the growth of bacterial populations in batch or continuous cultures does not permit any conclusions about the growth behavior of individual cells, because the distribution of cell site (and hence cell age) among the members of the population is completely random. Information about the growth behavior of individual bacteria can, however be obtained by the study of synchronous cultures. Synchronized cultures must be composed of cells which are all at the same stage of the bacterial cell cycle. Measurements made on synchronized cultures are equivalent to measurements made on individual cells.

A number of clever techniques have been devised to obtain bacterial populations at the same stage in the cell cycle. Theoretically, the smallest cells in a bacterial population arc those that have just completed the process

of cell division. Synchronous growth of a population of bacterial cells is illustrated in I figure . Synchronous cultures rapidly lose synchrony because not all cells in the population divide at exactly the same size. age or time.

Application of cell synchrony

1. It is used and important to knots the regulation between various cell cycle phases
2. It provides methodology to study a side variety of cell cycle processes.
3. It is used for cell separation.
4. It is helpful in industrial application as to obtain synchronous cultures.
5. It is helpful in enumeration and estimation of bacteria in media and brink
6. It helps in measurement of total cell yield from dense culture.
7. It is used in microbiological assays.

Q.2 Cell - cell interaction

Direct interactions between cells, as cell 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 shire blood cells $t_{o\ s}it_{es}$ 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.

For a coordinated function of cells in a tissue, tissues in an organ. organs in a system and systems in the body. cells need to he able to communicate with each other. Each cell should be capable of sending chemical signals to other cells and of 'wt.' it).t chemical signals from other cells as well as signals (chemical or other) from its immediate environment

Cell Adhesion Protein.

Cell-cell adhesion selective process. such that cells adhere only to other cells of specific types. this

selectivity was first demonstrated in classical studies of embryo development. which showed that cells from one tissue (e.g.. liver) specifically adhere to cells of the same tissue rather than to cells of a different tissue (e.g.. brain). Such selective cell-cell adhesion is

mediated by transmembrane proteins called cell-

adhesion molecules, which can he divided into

four major groups:

1) Selectins
2) Integrins
3) Inummoglobulin (Ig)superfamily (so named because they contain structural domains similar)
4) Cadherins

Cell adhesion mediated by the seletins.integrins. andcadherins requires ('a or %hi'. so many adhesive interactions between cells are Ca^{2+} or Mg^{2+} dependent.

Cell Adhesive on Molecules:

Family	Ligands recognized	Stable cell junctions
Selectins	Carbohydrates	no
Integrins	Extracellular matrix	Focal adhesion and hemidesmosomes
	Members of Ig superfamily	No
Ig superfamily	Integrins	No
	Homophilic interactions	No
Cadherins	Homophilic interactions	Adherens junctions and desmosomes

The selectins mediate transient interactions between leukocytes and endothelial cells or blood platelets. 'theselectins recognize cell surface carbohydrates. 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 binding of ICAMs to integrins is an example of a heterophilic interaction. in which an adhesion
molecule on the surface of one cell (e.g.. an ICAM) recognizes a different molecule on the surface of another

Cell (eg., an integrin). Other members of the Ig superfamily mediate homophilic interactions, in which an adhesion molecule on the surface of one cell hinds to the same molecule on the surface of another cell.

the fourth group is cell adhesion molecules. The cadherins. also display homophilic binding specificities. 'theyare not only involved in selective adhesion between embryonic cells but are also primarily responsible for the formation of stable junctions between cells in tissues.

Tight Junctions

It is a specialized cell-cell junction play key roles in animal tissues. Tight junctions usually associated with adherens junctions and desmosomes in a junctional complex are critically important to the function of epithelial cell sheets as barriers between fluid compartments.

For example.the intestinal epithelium separates the lumen of the intestine from the underlying connective tissue, which contains blood capillaries.

Tight junctions play two roles in allowing epithelia to fulfill such barrier functions.

1) Tight junctions form seals that prevent the free passage of molecules (including ions) between the cells of epithelial sheets.

2) Tight junctions separate the apical and hasolateral domains ofthe plasma membrane by preventing the free diffusion of lipids and membrane proteins between them.

(A)Electron micrograph of epithelial cells joined by a junctional complex. Including a tight junction, an adherens junction, and a desmosome. (13) Tight junctions are formed by interactions Between strands of transmembrane proteins (occludin and claudins) on adjacent cells

junctions are the closest known contacts between adjacent cells. They were originally described as sites of apparent fusion between the outer leaflets of the plasma membranes. although it is now clear that the membranes do not fuse. Instead, tight junctions appear to be formed by a network of protein strands that continues around the entire circumference of the cell.

Gap Junctions: Gap junctions, fouund in most animal tissues, serve as direct connections between the cytoplasms of adjacent cells. They provide open channels through the plasma membrane, allowing ions and small molecules to diffuse freely between neighboring cells, but preventing the passage of proteins and nucleic acids.

Consequently.gap junctions couple both the metabolic activities and the electric responses of the cells they connect. Most cells in animal tissues including epithelial cells. endothelial cells. andthe cells of cardiac and smooth muscle communicate by gap junctions. In electrically excitable cells.such as heart

muscle cells. thedirect passage of ions through gap junctions couples and synchronizes the contractions of neighboring cells. Gap junctions also allow the passage of sonic intracellular signaling molecules. such as cAMP and Ca^{2+}. bemcen adjacent cells. potentially coordinating the responses of cells in tissues.

Gap junctions are constructed of transmembrane proteins called connexins

Six **connexins** assemble to form a cylindrical with an open aqueous pore in its center. Such an assembly of connexins in the plasma membrane of one cell then aligns with the connexins of an adjacent cell. forming an open channel between the Iwo cytoplasms. the plasma membranes of the nut cells are separated by a gap corresponding to the space occupied by the comes in extracellular domains--hence the term ˜ gap junction˜.

Gap iunctions•

(A) electron micrograph of a gap junction (arrows) between two liver cells

(B) Gap junctions consist of assemblies of six connexins. which form open channels through the plasma membranes of adjacent cells.

Plant Cell Adhesion and Plasmodesmata

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 celldonotrequire the formation of cytoskeletal links such as those provided by the the desmosomes and adherens junctions of

animal cells however adjacent plant cells communicate with each other through cytoplasmic connections called plasmodesmata. Which function analogously to animal cell Gap junctions.

Despite their similarities in function, plasmodesmata are structurally unrelated to gap junctions. A each plasmodesma the plasma membrane of one cell is continuous with that of its neighbor, creating an open channel between the two cytosols . An extension of the smooth endoplasmic reticulum passes through the port• leaving a ring of surrounding cytoplasm through which ions and small molecules are able to pass freely between the cells. In addition.plasmodesmatacan expand in response to appropriate stimuli, permitting the regulated passage of macromolecules between adjacent cells. Plasmodesmata may thus play a key role in plant development by controlling the trafficking of regulatory molecules. such as transcription factors or RNAs. between cells.

Plasmodesmata. (A) Electron micrograph of plasmodesmata (arrows). (B) At plasmodesmata, the plasma membranes of neighboring cells are continuous. forming cytoplasmic channels through the adjacent cell walls.

Q.3 Cell signaling

Cell signaling is part of a complex syst*em* of communication that **governs** basic cellular activities and coordinates cell actions. The ability of cells to perceive and correctly respond to their microenvironment is the basis of development. tissue repair. and immunity **as well** as normal tissue homeostasis.

Many different kinds of molecules transmit information between the cells of multicellular organisms. Although all these molecules act as ligands that hind to receptors expressed by their target

cells, there is considerable variation in the structure and function of the different byes of molecule; that Nene ***as si***gnal transmitters.

Modes of cell signaling

A cell can communicate signals to other cells in various ways:

1) **Autocrine signaling** is a way for a cell to alter its own extracellular environment. **which**in turn affects the was the cell functions. the cell secretes chemicals outside of its membrane and the presence of those chemicals on the outside modifies the behavior of that same cell. 'Ibis process is important for growth.
2) **Paracrine signaling** is a was for a cell to affect the behavior of neighboring cells by secreting chemicals into the common intercellular space. this is an important process during embryonic development.
3) **Endocrine signaling** utilizes hormones. A cell secretes chemicals into the bloodstream. those chemicals affect the behavior of distant target cells. We will go into more details of autocrine. paracrine and endocrine signaling later on. when we tackle the human endocrine system

Cell signaling can result either from the direct interaction of a cell with its neighbor or from the action of secreted signaling molecules. Signaling by direct cell-cell (or cell-matrix) interactions plays a critical role in regulating the behavior of cells in animal tissues.

For example the integrins and cadherins function not only as cell adhesion molecules but also as signaling molecules that regulate cell proliferation and survival in response to cell-cell and cell-matrix contacts. In addition, cells express a variety of cell surface receptors that interact with signaling molecules on the surface of neighboring cells. Signaling via

such direct cell-cell interactions plays a critical role in regulating the many interactions between different types of cells that take place during embryonic development, as well as in the maintenance of adult tissues.

Cell receiving a signal

Some small molecules are capable of entering the cell through the plasma membrane. Nitrous oxide is one example. Upon entering the cell, it activates an enzyme.

Some small hormones also enter the cell directly, by passing through the membrane. samples are steroid hormones, thyroid hormones and melatonin. Once inside the cell, they hind cytoplasmic or nuclear receptors. The hormone-receptors complex enters the nucleus and binds to a particular sequence on the DNA. Thus. a new protein appears in the cell and assumes its normal function within it for gets secreted). The action of nuclear receptors is slow, as it takes some hours for the whole process to occur. The effect is long-lasting (or even permanent) and changes the properties the cell. This type of process is

important in development. Differentiation and maturation of cells. e.g...Gametes (eggs and sperm cells).

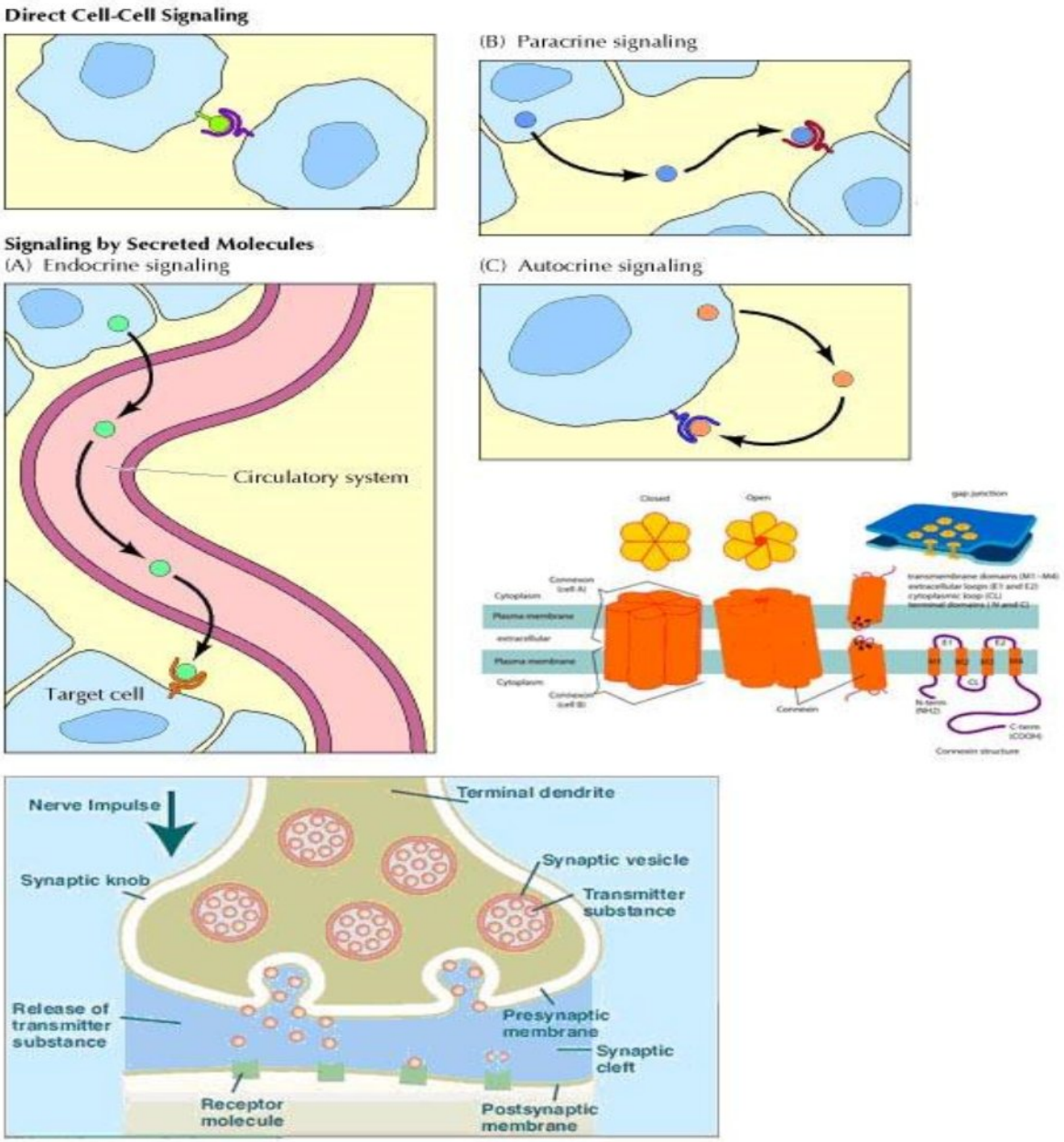


Fig :- Mode of Cell Signaling

Thereare three types of cell surface receptors:

1) Membrane enzymes
2) Ion channels
3) Transmembrane receptors

When a signaling chemical binds to the **membrane enzymes** protein on the outside of the cell, triggers a change in the 3D conformation of that protein, which in turn, triggers a chemical reaction on the inside of the cell.

When a signaling molecule binds to an **ion channel** on the outside of the 3D conformation of the protein and the channel opens, allowing the ions to move in or out of the cell following their electrical gradients and thus altering the polarization of the cell membrane. Some ion channels respond to non-chemical stimuli in the same way, including changes in electrical charge or mechanical disturbance of the membrane.

G protein-linked receptors: are seven-pass transmembrane proteins. This means that the poly peptide chain traverses the membrane seen times. When a chemical - a hormone or a pharmaceutical agent - binds to the receptor on the outside of the cell, this triggers a series of chemical reactions. including the movement and binding of the G protein. transformation of GTP into GDP **and** activation of second messengers. Second messengers(e.g.. cyclic AMP) start a cascade of enzymatic reactions leading to the cellular response. this signaling method isquite fast and. more importantly. it amplifies the signal.Binding of a single hormone molecule quickly results in thousInds of molecules of second messengers acting on even more molecules of enzymes and so on.

Q.4 Cell Locomotion

It is the cell migration or cell crawling, a central process in the development and maintenance of organisms.

Cell may migrate using different structures, proteins and towards chemicals.

The Prokaryotic Flagellum

The bacterial flagellum is a helical structure that drives the cell through the media like a propeller. The structure is rigid and turned by a rotator motor at the base where it connects to the bacteria's body. The rotator motor consists of several wheel- like discs one of which the M-ring interacts with the c-ring and studs to rotate the whole structure. The flagella is composed of a protein called flagella which is synthesized in the cell body and transported through the narrow lumen of the growing flagella itself to polymerize at the tip.

This system has evolved into a syringe -like mechanism to inject toxins into the cells of vertebrates during infection. The bacterial flagellum is driven by a proton motive force resulting from a gradient of protons. Bacterial chemotaxis brought about by alterations in the direction that the motor rotates in: this in turn is controlled by phosphorylation.

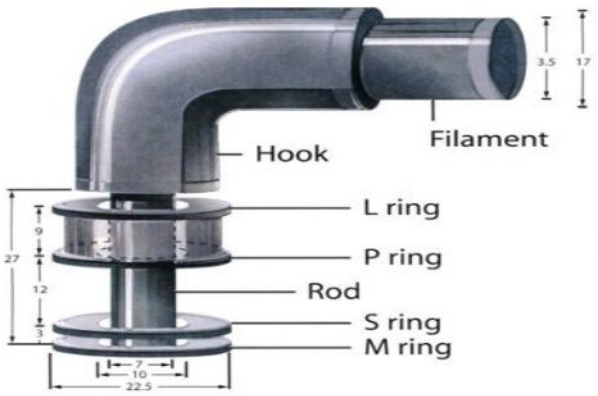


Fig:- The prokaryotic Flagellum

The Eukaryotic Flagellum

Although at first sight the flagella of eukaryotes are similar to the flagella of prokaryotes. our flagella are completely dissimilar in structure. function and in the genes that encode their components. The principle component of the eukaryotic flagella is the microtubule. a tubular array of proteins of the tubulin family. Instead of rotating as the prokaryotic flagella do, the eukaryotic flagella produce contortions in shape that travel around the structure like a Mexican wave.

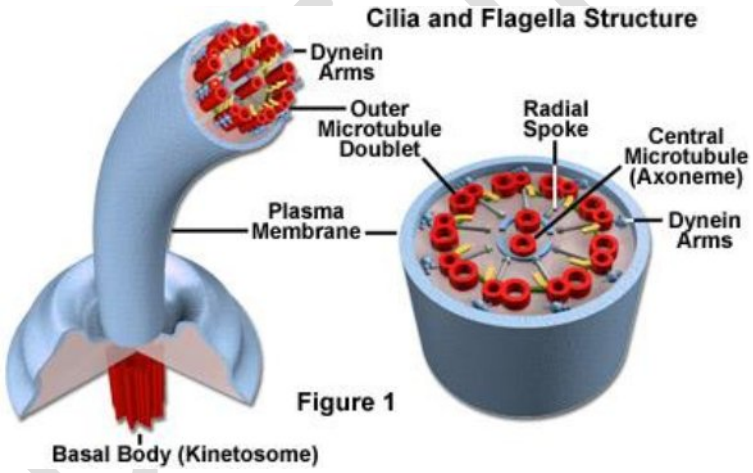


Fig: - The Eukaryotic Flagellum

The term cilia is generally used to describe small grouped structures less than 10mm. and flagella lend to be single structures about -10mm and flagella tend to be single structures about 40mm .In the human body they are used in mucus membranes to driven mucus around (out of the lungs), to drive sperm cells. but bizarrely in development a single cilium is responsible for setting up the asymmetry of our internal organs. The outer doublets are composed of microtubules and the outer and inner arms are dynein. Dynein is a motor protein that works with microtubules much like myosin works on actin so that the whole flagellum is sent into spiral motions as each set of arms (dynein) walks up the microtubules.

Crawling locomotion of cells

Observations on the amoeba revealed that there were two convertible states of cytoplasm. endoplasm and ectoplasm. Endoplasm (seen at the cell centre, thus the name) was fluid. while ectoplasm under the cell membrane is gellated and comparatively static. During active locomotion. endoplasm flows forwards faster than the speed of the cell. As the fluid endoplasm reaches the "hyaloplasm".a special optically clear form of ectoplasm. the flow diverted toward the membrane whereupon the endoplasm Gelled to firm ectoplasm. Vesicles.crystals, and other visible cytoplasmic inclusions are seen to become suddenly immobile having previously been seen to vibrate in Browning motion.

These transformations also take place in other cell types but are less visible because of the much smaller scale and because they take place over a much longer time scale. However these transformations are quite clearly visible in small amoeba such as *Acomthamoeho. Dicytostelium*and Naegleria.and also in highly motile human cells such as macrophages and neutrophils.

Cellular motors

They are biological molecular machines that are the essential agents of movement in living organisms. In general terms, a motor may he defined as a device that consumes energy in one form and converts it into n in or mechanical work: for example. many protein-based molecular motors harness the chemical free energy released by the hydrolysis of ATP in order to perform mechanical work.

Motor proteins utilizing the cytoskeleton for movement fall into two categories based on their substrates: Actin motors such as myosin move along microfilaments through interaction with actin. Microtubule motors such as dynein and kinesinmove along microtubules through interaction with tubulin. There are two basic types of microtubule motors: plus-end motors and minus-end motors. depending on the direction in which they "walk" along the microtubule cables within the cell.

- **Actin motors**

Myosin

myosins are actin motors and form myosin complexes consisting of two heavy chains wills motor heads and \ light chains. Derived from the Greek word for muscle. myosin is the protein responsible for generating muscle contraction. By non-processively walking along actin filaments.many molecules of myosin generate enough force to contract muscle tissue. Myosins are also vital in the process of cell division. They are also involved in cytoplasmic streaming. wherein movement along microfilament *networks* in the cell allows organdies and cytoplasm to stream in a particular direction.

The Myosins — Key motor proteins in Cell Motility and Locomotion.

litemyosins are a group of motor proteins capable of transforming chemical energy in the form of ATP to movement via the amplification (by levers) of conformational changes within the ATP hydrolysis head group. Although there are a huge number of myosin members we will be discussing myosin II. this is the myosin that is the major myosin in muscle and is responsible for two-headed contractile functions (such as cytokinesis) in the vast majority of non-muscle cells.

Myosin II: Each catalytic head group is controlled by two light chains. a regulatory and an essential light chain. Light chains are calmodulin-like proteins and wrap around a helical neck region. The heavy chain tail regions wrap round each other too. Each myosin II molecule self associates in an anti-parallel manner regulated by phosphorylation at the C-terminus (P).

- **Microtubule Motors**

Kinesin

kinesis are a group of related motor proteins that use a microtubule track along which to "walk." They are vital to movement of chromosomes during mitosis and are also responsible Mr shuttling mitochondria. Golgi bodies.and vesicles within eukaryotic cells. Kinesins typically contain two heavy chains with motor heads which move along microtuhules via a pseudo-processive asymmetric walking motion. which can he towards the plus-end or the minus-end. depending on the type of kinesin.

Dynein

Dyneins are microtubule motors capable of a sliding movement. Dynein complexes are much larger and more complex than kinesin and myosin motors. Dynein facilitates the movement of cilia and flagella.

Q.5 Cell Senescence and cell death

'Senescence' or 'biological aging' is the change in the bioology of an organism as it ages after its maturity. Such changes range from those affecting its cells and their function to those affecting the whole organism. There are a number of theories as to why senescence occurs: for example. some posit it is programmed by gene expression changes. others that it is the cumulative damage caused by biological processes. Senescence is not the inevitable fate of all organisms. A variety of organisms, including some cold-blooded animals, have negligible senescence.

The word *senescence* is derived from the Latin word *wont* meaning *old man,* old age, or *advanced in age.*

Cellular senescence

Cellular senescence is the phenomenon by which normal diploid cells lose the ability to divide. normally alter about 50 cell divisions in vitro (artificially). Some cells become senescent after fewer replication cycles as a result of DNA double strand breaks, toxins. etc. This phenomenon is also known as "replicative senescence", the "Hayflick phenomenon", or the Hayflick limit in honour of Dr. Leonard Hayflick, co-author with Paul Moorhead, of the first paper describing it in 1961.

In response to DNA damage (including shortened telomeres), cells either age or self-destruct (apoptosis. programmed cell death) if the damage cannot he easily repaired. In this 'cellular suicide', the death Of one cell or more, may benefit the organism as a whole.

It is deteriorative process which naturally slows down and terminates the functional life of a cell.

Senescence indicated by decline in functional efficiency of specialized nun-dividing cells. E.g.: Nerve cells. muscle cells. Decline in division capacity of actively dividing cells (Replicative senescence/ I Heyflick effect) e.g.: Lymphocytes, Epithelial cells.

It involves two processes:

I) Cell aging

2) Cell senescence

Hay flick Limit and Effect

In 1961 . Leonard Hay flick and Paul M**oorhead** discovered that human cells derived from embryonic tissues could only divide a finite number of times in culture. They divided the stages of cell culture into Phases I, II.

Phase I is the primary culture. when cells from the explains multiply to cover the surface of the culture flask--most cell types grow in the lab attached to a solid surface.

Phase II represents the period when cells divide in culture. Briefly, once cells cover a flask's surface. they stop multiplying. For cell growth to continue, the cells must be subcultivated. To do so one removes the culture's medium and adds a digestiv*e* enzyme called trypsin that dissolves the substances binding cells together. After adding growth medium and pipetting one obtains the cells in a homogeneous suspension that are then divided by two--or more--new• flasks. Cells then attach to the new flasks' surface and start dividing once again until a new sub cultivation is required. Most cells divide vigorously and can often be subcultivated in a matter of a few days.

After several months, however, cells start dividing slower, which marks the beginning of Phase Ill. Eventually cells stop dividing at all. though they may or may not die. Hay flick and Moorhead noticed that cultures stopped dividing after an average of 50 cumulative population doublings (CPDs)--splitting one flask of cells into two new flasks of the same size increases CPDs by *one,* splitting by four flasks increases the CPIs by two and so on. This phenomenon of growth arrest alter a period of apparently normal cell proliferation is known as the Hayflick limit. Phase III phenomenon. or. as it will be called herein, replicative senescence (RS).

Cellular changes during senescence

1) Morphological:

- Change in cell size and shape — Atrophy.
- Accumulation of Lipofuscin pigment.

- Nuclear pyknosis.
- Lipid vacuole formation.

2) Physiological changes:

- Accumulation of calcium ions.
- Loss of ribosomal RNA.
- Decline in transcription.
- Decline in protein synthesis.
- Stiffening o collagen.
- Decline in energy production.

3) Sub-cellular changes:

- Change in fluidity and permeability of plasma Membrane.
- Decrease in granular ER.
- Degeneration of mitochondria.
- Degeneration of chloroplasts (plant cells).

Theories of Senescence: The process of senescence is complex. And may derive from a variety of different mechanisms and exist for a variety of different reasons. However senescence 15 not universal and scientific evidence suggests that cellular senescence evolved in certain species because it prevents the onset of cancer. In a few, simple species .such as Hydra senescence is negligible and cannot be detected.

I. DNA Damage theories

- Accumulated damage to DNA inhibits cell functions and division.
- Decline in DNA repair capacity • • Gene expression inhibited.
- Damage to mitochondria! DNA - Decrease in energy production.

2. Built---in Breakdown theories (Genetic theories)

- Genetically programmed process. Senescence associated genes express after a fixed life span of cells.
- Expression of genes coding for hydrolyticenzymes.
- Formation of DNA synthesis inhibiting proteins.

3. Telomere Theory

- Gradual shortening or telomeres with each division / Reduction in telomerase activity.
- Division stops \ hen telomere becomes very short.
- Replicative cell senescence

4. Free radical damage theory

- Damage by highly reactive free radicals like OH, H2O2. etc.
- Disrupts cell and nuclear membranes.
- Alters membrane permeability. Damage DNA / Proteins.

- Antioxidants reduce free radical damage.

5. Other Theories

- Glycation theory - Glucose forms cross-links between proteins and stiffens them.
- Immune theory - Antibodies against body's my n cells - autoimmune responses.
- Endocrine theory. - A pituitary hormone triggers senescence.

Q.6 Programmed Cell Death (PCD) or Apoptosis

Cells are committing suicide by the activation of an intra-cellular death program.

Apoptosis means Falling 1)111 like the leaves aa tree in autumn.

It is a neat and silent death of cells.

Apoptosis is the process of programmed cell death (PCD) that may occur in multicellular organisms. Biochemical events lead to characteristic cell changes (morphology) and death. These changes include blebbing, cell shrinkage, nuclear fragmentation, chromatin condensation, and chromosomal DNA fragmentation.

For example, the differentiation of fingers and toes in a developing human embryo occurs because cells between the fingers apoptose; the result is that the digits arc separate.

Mechanism of PCD (Programmed cell death)

I. Reception of death or apoptotic signals (Internal / external).

2. Activation of Inactive Procaspases to active Caspases (Cascade Reaction).
3. Degradation of cytoplasmic and nuclear proteins.
4. Fragmentation of DNA/Nucleus. Shrinkage and blebbing of cells.
5. Formation of smaller membrane bound structures called apoptotic bodies.
6. Digestion of apoptotic bodies by macrophages Phagocytosis.
7. Recycling of the macro-molecules.

Apoptosis Mainly mediated by a group of proteolytic enzymes called Caspaces. Present in all cells in an inactive term called Procaspaces. Activated to kill cells by death signals, The Poison pills of Cells!

Significance of PCD/Apoptosis

1. Normal process during development.
2. Responsible for sculpturing of body parts during development of animals.
3. Development of animal paws. Metamorphosis of tadpole into frog. insect larva into pupa.
4. Balancing number of cells in developing Tissues (homeostasis).

5. Eliminating unwanted/ improperly formed/ functionless and dangerous cells.

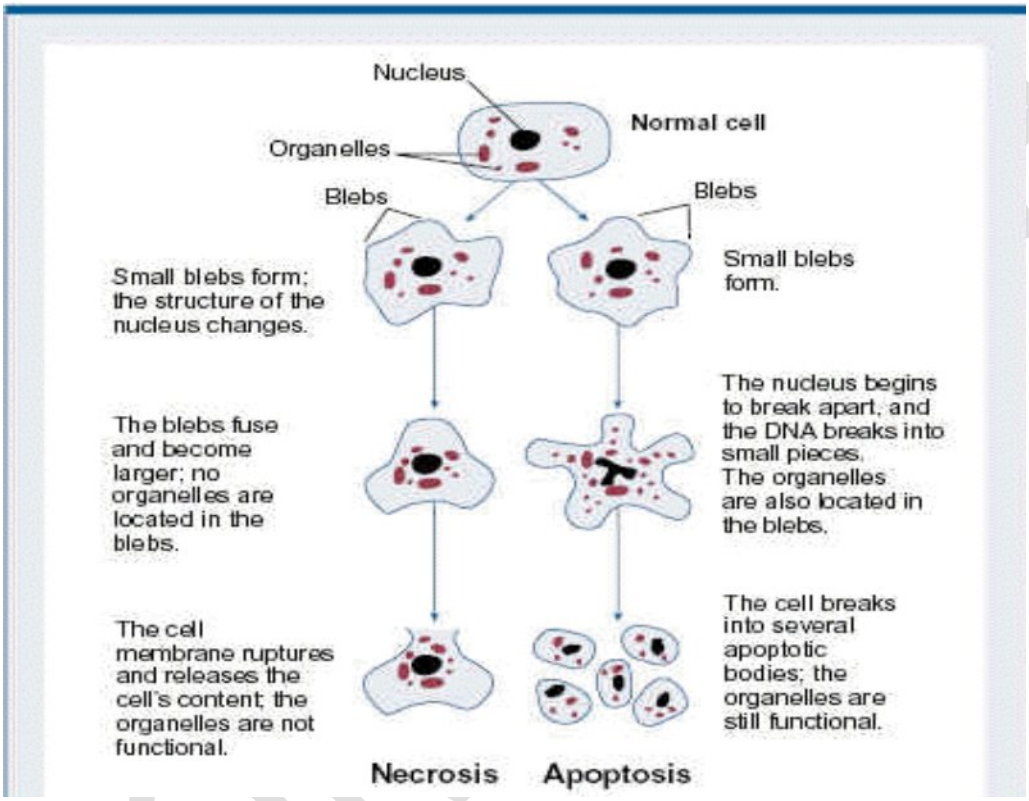


Fig :- The Programmed Cell Death (Apoptosis)

Q.7 Cellular Differentiation

Cellular differentiation is the process by which a less specialized cell becomes a more specialized cell type. Differentiation occurs numerous times during the development of a multicellular organism as the organism changes from a simple zygote in a complex system of tissues and cell types

Differentiation is a common process in adults as well: adult stein cells divide and create fully differentiated daughter cells during, tissue repair and during normal cell turnover.

Differentiation dramatically changes a cell's size, shape, membrane potential, metabolic activity and responsiveness to signals. These changes are largely due to highly controlled modifications in gene

expression. A cell that is able to differentiate into all cell types of the adult organism is known as *pluripotent.* Such cells are called embryonic stem cells in animals and meristematic cells in higher plants.

A cell that is able to differentiate into all cell types, including the placental tissue, is known as *Totipotent*. In mammals, only the zygote and subsequent blastomeres are totipotent, while in plants many differentiated cells can become totipotent with simple laboratory techniques.

Mammalian cell types

Three basic categories of cells make up the mammalian body:

1. Germ cells: Germ line cells are any line of cells that give rise to gametes —eggs and sperm— and thus are continuous through the generations. Stem cells, on the other kind, have the ability to divide for indefinite periods and to give rise to specialized cells. They are best described in the context of normal human development.
2. Somatic cells: Each of the approximately 100 trillion 10^{14} cells in an adult human has its own copy or copies of the genome except certain cell types, such as red blood cells, that lack nuclei in their fully differentiated state. Most cells are diploid they have two copies of each chromosome. Such cells, called somatic cells, make up most of the human body, such as skin and muscle cells. Cells differentiate to specialize for different functions.
3. Stem cells: Development begins when a sperm fertilizes an egg and creates a single cell that has the potential to form an entire organism. In the first hours alter fertilization; this cell divides into identical cells. In humans. Approximately four days after fertilization and after several cycles of cell division, these cells begin to specialize. forming a hollow sphere of cells, called a blastocyst. The blastocyst has an outer layer of cells, and inside this hollow sphere. there is a cluster of cells called the inner cell muss. The cells of the inner cell mass go on to form virtually all of the tissues of the human body. Although the cells of the inner cell mass can form virtually every type of cell found in the human body. they cannot form an organism. These cells are referred to as pluripotent.

Stages of cell differentiation

A cell that is able to diffrentiate into all cell types is known as totipotent. In mammals, only the zygote and the products of the first few cell divisions (cleavage) are totipotent, while in plants, many differentiated cells can become totipotent with simple laboratory techniques.

A cell that is capable of differentiating into man cell types is known as pluripotent these cells are called stem cells in animals and meristematic cells in higher plants. The pluripotent cells can divide to produce differentiated descendants. yet also retain the ability to divide to maintain the stem cell population. They are the most versatile stem cells.

Pluripotent stem cells undergo further specialization into stein cells that are committed to give rise to cells that have a particular function. Examples include blood stein cells that give rise to red blood cells, white blood

Cells, and platelets, and skin stem cells that give rise to the various types of skin cells. These more specialized stem cells are called multipotent; multipotent cells are capable of giving rise to several kinds of cells, tissues or structures.

As cells undergo different union, they change from being totipotent Io pluripotent to multipotent to, finally. Specialized cells.

Mechanisms of differentiation

Each specialized cell type in an organism expresses a subset of all the genes that constitute the genome of that species.

Loch cell type is defined by its particular pattern of regulated gene expression. cell differentiation is thus a transition of a cell from one cell type to another and it involves a switch from one pattern of gene expression to another.

Cellular differentiation during development can be understood as the result of a gene regulatory network. Regulatory *gene and its* regulatory modules are nodes in a gene regulatory network: they receive input and create output elsewhere in the network.

A few evolutionary conserved types of molecular process are often involved in the cellular mechanism that control these switches. The major types of molecular processes that control cellular differentiation involve cell signaling.

Many of the signal molecules that convey information from cell to cell during the control of cellular differentiation *are* called growth factors. Although the details of specific signal transduction pathway s vary these pathways often share the following general steps. A ligand produce by one cell binds to a receptor in the extra cellular region of another cell, inducing a conformational change in the receptor. The shape of the cytoplasmic domain of the *receptor* changes, and the receptor acquires enzymatic activity. The receptor then catalyzes reactions that phosphorylate other proteins, activating them. A cascade of phosphorylation reactions eventually activates a dormant transcription factor or cytoskeletal protein, thus contributing to the differentiation process in the target cell.

Induction refers to cascades of signaling events, during which a cell or tissue signals to another cell or tissue to influence its developmental fate.

Other important mechanisms fall under the category of asymmetric cell divisions, divisions that give rise to daughter cells with distinct developmental fates.

Asymmetric cell divisions can occur because of segregation of cytoplasmic determinants or because of signaling. In the former mechanism, distinct daughter cells are created during cytokinesis because of an uneven distribution of regulatory molecules in the parent cell. The distinct cytoplasm that each daughter cell inherits results in a distinct pattern of differentiation liar each daughter cell.

For e.g., a well-studied example of pattern formation by asymmetric divisions is body axis patterning in Drosophila. RNA molecules are an important type of intracellular differentiation control signal.

The molecular and genetic basis of asymmetric cell divisions has also been studied in green algae of the genus *volvox*, a model system Air studying how unicellular organisms can evolve into multicellular organisms.