1 (a) Write a short note on the following (any three):

- (i) SDS
    - SDS, or Sodium Dodecyl Sulfate, is a powerful anionic detergent frequently employed in biochemistry and molecular biology, particularly in the technique of SDS-Polyacrylamide Gel Electrophoresis (SDS-PAGE). Its primary function is to denature proteins by binding to them in a consistent ratio, causing them to unfold into linear chains and acquire a uniform negative charge-to-mass ratio. This property allows for the separation of proteins primarily based on their molecular weight during electrophoresis. Additionally, SDS is effective in lysing cells and solubilizing cellular membranes due to its strong detergent properties.
- (ii) Nuclear envelope
    - The nuclear envelope is a double membrane structure that encases the genetic material (DNA) within eukaryotic cells, thereby physically separating the nucleus from the cytoplasm. It consists of an outer and an inner membrane, with a perinuclear space in between that is continuous with the endoplasmic reticulum lumen. This envelope is punctuated by complex structures called nuclear pores, which meticulously regulate the bidirectional passage of molecules, including proteins and RNA, between the nucleus and cytoplasm. The outer membrane is often associated with ribosomes, while the inner membrane is supported by the nuclear lamina, a protein meshwork crucial for nuclear structure and chromatin organization.
- (iii) Golgi apparatus
    - The Golgi apparatus, also known as the Golgi complex, is a vital membrane-bound organelle in eukaryotic cells. It is primarily responsible for the modification, sorting, and packaging of proteins and lipids that are synthesized in the

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endoplasmic reticulum. Structurally, it is composed of a stack of flattened, membrane-bound sacs called cisternae, exhibiting distinct *cis* (receiving), medial, and *trans* (shipping) regions. The Golgi performs extensive glycosylation of proteins and lipids, sorts them into various vesicles destined for different cellular locations like the plasma membrane, lysosomes, or for secretion, and is also involved in the synthesis of certain polysaccharides.

- (iv) Hurler syndrome
    - Hurler syndrome, medically known as Mucopolysaccharidosis Type I H (MPS I H), is a rare, inherited lysosomal storage disorder. It arises from a genetic deficiency in the enzyme alpha-L-iduronidase, which is essential for breaking down specific complex carbohydrates called glycosaminoglycans (GAGs), namely dermatan sulfate and heparan sulfate, within lysosomes. The accumulation of these undigested GAGs within cells leads to progressive and widespread damage to various tissues and organs. Symptoms, typically appearing in infancy, include skeletal abnormalities (e.g., short stature, joint stiffness), progressive intellectual disability, enlarged liver and spleen, heart problems, and corneal clouding. It is an autosomal recessive condition, and treatments such as enzyme replacement therapy and hematopoietic stem cell transplantation can help manage its progression.

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1 (b) Differentiate between the following (any three):

- (i) Prokaryotic cell and eukaryotic cell
    - **Prokaryotic Cell:**
        - Lacks a true nucleus; genetic material is located in a nucleoid region within the cytoplasm.
        - No membrane-bound organelles (e.g., mitochondria, endoplasmic reticulum, Golgi apparatus).

- Generally much smaller in size, typically 0.1-5 micrometers in diameter.
- Contains a single, circular chromosome; may also possess plasmids.
- Ribosomes are 70S type.
- Cell wall is almost always present, primarily composed of peptidoglycan (in bacteria).
- Primarily reproduces by binary fission (asexual).

- **Eukaryotic Cell:**
  - Possesses a true nucleus enclosed by a nuclear envelope.
  - Contains various membrane-bound organelles (e.g., mitochondria, endoplasmic reticulum, Golgi apparatus, lysosomes, vacuoles).
  - Generally much larger in size, typically 10-100 micrometers in diameter.
  - Contains multiple, linear chromosomes complexed with histone proteins, located within the nucleus.
  - Ribosomes are 80S type in the cytoplasm (with 70S in mitochondria/chloroplasts).
  - Cell wall is present in plants (cellulose) and fungi (chitin), but absent in animal cells.
  - Reproduces by mitosis (asexual) and meiosis (sexual).

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- (ii) Desmosomes and hemidesmosomes
  - **Desmosomes (Macula Adherens):**
    - Mediate strong cell-to-cell adhesion, connecting adjacent cells.

        - Consist of dense cytoplasmic plaques on the plasma membranes of two adhering cells, to which intermediate filaments (e.g., keratin filaments) are anchored.
        - Transmembrane adhesion proteins called cadherins extend from the plaques into the extracellular space, linking the two cells.
        - Abundant in tissues subjected to mechanical stress, such as skin and cardiac muscle.
    - **Hemidesmosomes:**
        - Mediate strong cell-to-extracellular matrix (ECM) adhesion, anchoring cells to the underlying basal lamina.
        - Consist of a dense cytoplasmic plaque on the inner surface of the cell's plasma membrane, also anchoring intermediate filaments.
        - Transmembrane adhesion proteins called integrins extend from the plaque into the extracellular space, binding to components of the basal lamina (e.g., laminin, collagen).
        - Crucial for maintaining tissue integrity and preventing epithelial detachment from the basement membrane.
- (iii) NLS and NES
    - **NLS (Nuclear Localization Signal):**
        - A short amino acid sequence (typically rich in basic amino acids like lysine and arginine) that acts as a signal for proteins to be actively imported from the cytoplasm into the nucleus.
        - Recognized and bound by importin receptor proteins in the cytoplasm.

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        - Found on proteins that function within the nucleus (e.g., histones, DNA polymerase, transcription factors).
    - **NES (Nuclear Export Signal):**
        - A short amino acid sequence (typically rich in hydrophobic amino acids like leucine) that acts as a signal for proteins (and associated RNA) to be actively exported from the nucleus to the cytoplasm.
        - Recognized and bound by exportin receptor proteins in the nucleus.
        - Found on proteins that function in the cytoplasm but are synthesized or processed in the nucleus (e.g., ribosomal proteins, some RNA-binding proteins).

- (iv) Mitosis and meiosis
    - **Mitosis:**
        - Purpose: Asexual reproduction, growth, repair, and tissue regeneration.
        - Location: Occurs in somatic (body) cells.
        - Number of Divisions: One division.
        - Chromosome Number: Daughter cells maintain the same chromosome number as the parent cell (e.g., diploid parent produces diploid daughters).
        - Number of Daughter Cells: Produces two daughter cells.
        - Genetic Content: Daughter cells are genetically identical to the parent cell and to each other.
        - Crossing Over: Does not occur.
    - **Meiosis:**

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- Purpose: Sexual reproduction, production of gametes (in animals) or spores (in plants and fungi).
- Location: Occurs only in germ-line cells.
- Number of Divisions: Two sequential divisions (Meiosis I and Meiosis II).
- Chromosome Number: Daughter cells have half the chromosome number of the parent cell (e.g., diploid parent produces haploid daughters).
- Number of Daughter Cells: Produces four daughter cells.
- Genetic Content: Daughter cells are genetically different from the parent cell and from each other due to crossing over and independent assortment.
- Crossing Over: Occurs during prophase I, leading to genetic recombination.

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2 (a) Discuss the model of membrane structure as proposed by Singer and Nicolson. How is this model different from earlier models of membrane structure?

- **Singer and Nicolson's Fluid Mosaic Model (1972):**
  - This model, the most widely accepted for biological membranes, proposes that the cell membrane is a dynamic, fluid structure rather than a rigid one.
  - **Key Features:**
    - **Fluid Lipid Bilayer:** The fundamental framework is a double layer of amphipathic phospholipids. Their hydrophilic heads face the aqueous environment, and their hydrophobic tails form the interior. The "fluid" aspect emphasizes the constant lateral movement, rotation, and flexion of these lipid molecules.

        - **Mosaic of Proteins:** Various proteins are embedded within, span across, or are loosely associated with this lipid bilayer, resembling a mosaic.
            - **Integral proteins:** Tightly associated, often spanning the entire membrane (transmembrane proteins).
            - **Peripheral proteins:** Loosely attached to the surface.
        - **Asymmetry:** The distribution of lipids, proteins, and carbohydrates (glycocalyx on the outer surface) across the two leaflets of the bilayer is asymmetrical.
        - **Cholesterol:** In animal cells, cholesterol molecules are interspersed within the bilayer, modulating fluidity and stability.
    - The model depicts the membrane as a "sea of lipids" in which "icebergs of proteins" are floating and able to move.

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- **Difference from Earlier Models (e.g., Davson-Danielli "Sandwich" Model, 1935):**
    - Earlier models, like the Davson-Danielli model, proposed a more static "sandwich" structure where a lipid bilayer was flanked by continuous, rigid layers of globular proteins on both inner and outer surfaces.
    - **Key Distinctions:**
        - **Protein Placement:** Singer and Nicolson showed that proteins are not just superficial layers but are integral, often penetrating or spanning the lipid bilayer. Earlier models suggested proteins formed continuous sheets.
        - **Fluidity:** The fluid mosaic model explicitly emphasized the dynamic, fluid nature of the membrane, allowing for

lateral diffusion of components, a concept largely absent in the more rigid earlier models.

- **Mosaic Arrangement:** It highlighted the discrete, non-continuous "mosaic" arrangement of proteins, contrasting with the continuous protein layers proposed before.
- **Asymmetry:** The fluid mosaic model incorporated and explained membrane asymmetry, which was not adequately addressed by previous models.
- **Functional Explanation:** The fluidity and mosaic arrangement better explained observed membrane functions such as selective permeability, cell-cell recognition, and receptor function, which were challenging for earlier models to reconcile.

2 (b) Discuss the term membrane asymmetry and membrane fluidity. Also discuss the experiment performed by Frye and Edidin to prove the mobility of membrane proteins.

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- **Membrane Asymmetry:**
  - Membrane asymmetry refers to the uneven distribution of lipids, proteins, and carbohydrates between the two leaflets (halves) of the lipid bilayer. This non-random arrangement is crucial for membrane function.
  - **Lipid Asymmetry:** Specific phospholipids (e.g., phosphatidylserine, phosphatidylethanolamine) are preferentially located on the inner leaflet, while others (e.g., phosphatidylcholine, sphingomyelin) are more abundant on the outer leaflet. Glycolipids are exclusively found on the outer leaflet.
  - **Protein Asymmetry:** Integral membrane proteins have a defined, fixed orientation within the membrane, and peripheral proteins are associated with either the inner or outer surface.

This ensures that specific protein domains are exposed to their appropriate environment (e.g., receptor binding sites extracellularly, signaling domains intracellularly).

- **Carbohydrate Asymmetry:** Carbohydrate chains are found almost exclusively on the outer surface, forming the glycocalyx, which is vital for cell-cell recognition and adhesion.
- **Significance:** Asymmetry is critical for signal transduction, membrane fusion events, maintaining ionic gradients, and distinguishing the cell's internal and external environments.

- **Membrane Fluidity:**
  - Membrane fluidity describes the viscosity of the lipid bilayer, reflecting how easily its constituent lipids and proteins can move laterally within the membrane plane. The membrane is a dynamic, rather than static, structure.
  - **Factors Influencing Fluidity:** Temperature (higher temperature = more fluid), degree of fatty acid saturation (unsaturated = more fluid due to kinks), fatty acid chain length (shorter chains = more fluid), and cholesterol content (modulates fluidity in animal cells).
  - **Significance:** Proper fluidity is essential for diverse membrane functions, including protein movement and interaction, membrane fusion (e.g., endocytosis, exocytosis), cell growth and movement, and regulating membrane permeability.
- **Frye and Edidin Experiment (1970) on Protein Mobility:**
  - **Purpose:** To provide direct experimental evidence for the lateral mobility of proteins within the cell membrane, supporting the fluid mosaic model.
  - **Methodology:**
    - They took human cells and mouse cells.

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- Surface proteins of human cells were labeled with a green fluorescent antibody.
- Surface proteins of mouse cells were labeled with a red fluorescent antibody.
- The human and mouse cells were then fused using Sendai virus to form hybrid cells (heterokaryons).
- **Observation:**
  - **Immediately after fusion:** The hybrid cells showed a distinct segregation of colors, with one half glowing green and the other half red. This indicated that the proteins from each cell remained in their original membrane halves.
  - **After incubation at 37°C:** Within approximately 40 minutes to an hour of incubation at physiological temperature, the green and red fluorescent labels completely intermixed and were uniformly distributed across the entire surface of the hybrid cell.

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- **Conclusion:** This rapid intermixing of membrane proteins unequivocally demonstrated that proteins are not rigidly fixed but are free to diffuse laterally within the fluid lipid bilayer, thus providing strong evidence for the "fluid" aspect of the fluid mosaic model.

3 (a) On what basis can we say that mitochondria and chloroplasts originated from bacteria that were engulfed by the precursor of eukaryotic cells? How are the two organelles different from each other?

- The **Endosymbiotic Theory** posits that mitochondria and chloroplasts originated from free-living bacteria that were engulfed by a host cell (the precursor to eukaryotic cells) and subsequently evolved into obligate endosymbionts.

- **Basis for Endosymbiotic Origin (Evidence):**
    - **Size and Morphology:** Both organelles are similar in size (1-10 µm) and shape (rod-like/oval) to typical bacteria.
    - **Double Membranes:** They possess two membranes: the inner membrane is thought to be the original bacterial plasma membrane, and the outer membrane is derived from the host's phagosomal membrane.
    - **Circular DNA:** Both contain their own circular DNA molecule, similar to bacterial chromosomes, and distinct from the linear DNA in the host nucleus. Their DNA lacks introns and is not associated with histones, characteristic of prokaryotic DNA.
    - **Ribosomes:** They possess 70S ribosomes, which are similar in size and structure to bacterial ribosomes and differ from the 80S ribosomes in the eukaryotic cytoplasm.
    - **Reproduction by Binary Fission:** Both organelles reproduce by a process similar to binary fission, dividing independently of the host cell's mitotic cycle.
    - **Genetic Homology:** DNA sequencing reveals strong genetic similarities between mitochondrial DNA and alpha-proteobacteria, and between chloroplast DNA and cyanobacteria, indicating a common evolutionary ancestry.
    - **Similarities in Metabolic Pathways:** They retain bacterial-like metabolic pathways (e.g., electron transport chain in mitochondria, photosynthesis in chloroplasts).
    - **Antibiotic Sensitivity:** Their protein synthesis is inhibited by antibiotics that target bacterial ribosomes, but not by those that affect eukaryotic cytoplasmic ribosomes.
- **Differences between Mitochondria and Chloroplasts:**
    - **Function:**

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        - **Mitochondria:** Primarily perform cellular respiration, generating ATP (energy) from organic molecules.
        - **Chloroplasts:** Primarily perform photosynthesis, converting light energy into chemical energy (sugars).
    - **Internal Structure:**
        - **Mitochondria:** Characterized by inner membrane folds called cristae, and an internal matrix.
        - **Chloroplasts:** Characterized by stacks of thylakoids (grana) within the stroma.
    - **Presence:**
        - **Mitochondria:** Present in nearly all eukaryotic cells (animal, plant, fungal, protist).
        - **Chloroplasts:** Present only in photosynthetic eukaryotic cells (plants and algae).
    - **Originating Bacteria:**
        - **Mitochondria:** Believed to have originated from an aerobic alpha-proteobacterium.
        - **Chloroplasts:** Believed to have originated from a photosynthetic cyanobacterium.
    - **Pigments:**
        - **Mitochondria:** Lack photosynthetic pigments.
        - **Chloroplasts:** Contain chlorophylls and carotenoids.

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3 (b) Explain in detail the process of co-translational transport of proteins in the ER.

- Co-translational transport is the primary pathway for proteins destined for the ER lumen, ER membrane, Golgi, lysosomes, endosomes, or

secretion, where they are moved into or across the ER membrane *as they are being synthesized*.

- **Detailed Process:**
    - **1. Synthesis Initiation in Cytoplasm:** Protein synthesis begins on free ribosomes in the cytoplasm.
    - **2. Signal Peptide Emergence:** The mRNA sequence for proteins destined for the ER contains a specific **ER signal peptide** (typically 15-30 hydrophobic amino acids) usually at the N-terminus. As this signal peptide emerges from the ribosome, it becomes exposed.
    - **3. SRP Binding and Translational Arrest:** A cytosolic ribonucleoprotein complex called the **Signal Recognition Particle (SRP)** recognizes and binds to the exposed signal peptide and the ribosome. This binding event temporarily halts protein synthesis (translational arrest).
    - **4. Docking at ER Membrane:** The entire SRP-ribosome-mRNA complex then diffuses to the surface of the Endoplasmic Reticulum membrane. Here, it binds to the **SRP receptor**, an integral protein embedded in the ER membrane. This binding step is regulated by GTP hydrolysis.
    - **5. Transfer to Translocon:** The SRP-ribosome complex is then handed over from the SRP receptor to a protein translocator channel in the ER membrane, known as the **translocon (Sec61 complex)**. Following this transfer, the SRP is released (requiring more GTP hydrolysis) and can be recycled for subsequent rounds of transport.
    - **6. Resumption of Synthesis and Translocation:** Protein synthesis resumes, but now the growing polypeptide chain is directly threaded through the aqueous pore of the translocon channel into the ER lumen.

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    - **7. Signal Peptide Cleavage and Folding:** As the polypeptide enters the ER lumen, the signal peptide is usually cleaved off by an enzyme called **signal peptidase**, located on the ER lumenal side of the membrane. Within the ER lumen, the nascent protein begins to fold into its correct 3D structure, often assisted by chaperone proteins (e.g., BiP) and undergoing modifications like N-linked glycosylation and disulfide bond formation.
    - **8. Final Destination:** For soluble proteins, synthesis completes, and the protein is released into the ER lumen. For transmembrane proteins, specific hydrophobic "stop-transfer" or "start-transfer" sequences within the polypeptide interact with the translocon, causing segments of the protein to be integrated into the ER membrane, establishing its correct orientation within the bilayer.
- This co-translational mechanism ensures efficient and accurate targeting of proteins to the ER, preventing them from being released into the cytoplasm where they might misfold or aggregate.

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4 (a) What are the distinguishing features between Gap junctions and plasmodesmata?

- Both Gap junctions and plasmodesmata facilitate direct intercellular communication, but they are found in different types of organisms and have distinct structures.
- **Gap Junctions:**
    - **Organisms:** Found exclusively in **animal cells**.
    - **Structure:** Formed by protein channels called **connexons**, each composed of six transmembrane protein subunits (connexins). Two aligned connexons from adjacent cells create a continuous aqueous channel. There is a small extracellular gap (2-4 nm) between the connected cell membranes.

- **Permeability:** Allow the passage of small molecules and ions (e.g., ATP, cAMP, sugars, amino acids) up to about 1,000 Daltons.
- **Regulation:** Their opening and closing can be regulated by intracellular calcium concentration and pH.
- **Function:** Facilitate rapid electrical and chemical communication between cells, crucial for coordinated activity in tissues like cardiac and smooth muscle, and for metabolic coupling in various epithelia.

- **Plasmodesmata:**
  - **Organisms:** Found exclusively in **plant cells**.
  - **Structure:** Are cytoplasmic channels that traverse the rigid plant cell wall, directly connecting the cytoplasm of adjacent cells. The plasma membrane of one cell is continuous with the plasma membrane of the next. A central tubular structure, the **desmotubule**, derived from the ER, typically runs through the center of the channel.
  - **Permeability:** Allow the passage of small molecules, ions, and even macromolecules like proteins and RNA up to about 50,000-100,000 Daltons.
  - **Regulation:** Their permeability can be regulated in response to developmental cues or stress.
  - **Function:** Essential for intercellular transport of water, nutrients, signaling molecules, and even transcription factors and viruses, crucial for plant growth, development, and defense. They represent the symplastic pathway for transport within plant tissues.

4 (b) Describe the two types of ER present in the cell. Illustrate any two functions of SER.

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- The Endoplasmic Reticulum (ER) is an extensive, interconnected network of membranes found in eukaryotic cells, forming a continuous lumen. It exists in two main forms: Rough ER (RER) and Smooth ER (SER).
- **1. Rough Endoplasmic Reticulum (RER):**
    - **Structure:** Characterized by the presence of ribosomes on its cytosolic surface, giving it a "rough" appearance. It typically forms flattened sacs (cisternae) and is continuous with the outer nuclear membrane.
    - **Main Functions:** Primarily involved in the synthesis, folding, modification, and quality control of proteins destined for secretion, insertion into membranes, or delivery to other organelles of the endomembrane system. Initial N-linked glycosylation and disulfide bond formation also occur here.
- **2. Smooth Endoplasmic Reticulum (SER):**
    - **Structure:** Lacks ribosomes on its surface, appearing "smooth." It is typically a network of interconnected tubules that are continuous with the RER. The extent of SER varies greatly depending on the cell type and its specific metabolic activities.
    - **Illustrative Functions (any two):**
        - **(i) Lipid Synthesis:** The SER is a major site for the synthesis of various lipids. This includes phospholipids, which are crucial components of all cellular membranes, and cholesterol, a vital steroid. In specialized cells, such as those in the adrenal cortex and gonads, the SER is highly developed for the synthesis of steroid hormones (e.g., testosterone, estrogen).
        - **(ii) Detoxification of Drugs and Poisons:** Particularly abundant in liver cells (hepatocytes), the SER contains a rich array of enzymes (e.g., cytochrome P450 enzymes).

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These enzymes are responsible for metabolizing and detoxifying a wide range of lipid-soluble drugs, pesticides, and harmful compounds. They convert these hydrophobic substances into more water-soluble forms, facilitating their excretion from the body.

- **(iii) Calcium Ion Storage and Release:** The SER plays a critical role in regulating intracellular calcium (Ca^{2+}) levels. It actively pumps Ca^{2+} from the cytosol into its lumen for storage. In muscle cells, a specialized SER called the sarcoplasmic reticulum (SR) is highly developed for the rapid release of stored Ca^{2+}, which is essential for initiating muscle contraction. In other cells, this controlled Ca^{2+} release acts as a crucial second messenger in various cellular signaling pathways.

5 (a) Discuss the structure, classification, and functions of intermediate filaments. How do they contribute to cellular structural integrity?

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- Intermediate filaments (IFs) are a key component of the cytoskeleton in eukaryotic cells, renowned for their exceptional mechanical strength and resilience. They have an approximate diameter of 10 nm, placing them "intermediate" between actin filaments and microtubules.
- **Structure:**
  - IFs are robust, rope-like polymers formed from various fibrous protein subunits.
  - **Monomer:** Each protein monomer has a central alpha-helical rod domain flanked by globular N-terminal head and C-terminal tail domains.
  - **Dimer:** Two monomers associate in a parallel coiled-coil formation.

    - **Tetramer:** Two such dimers then associate in an anti-parallel, staggered arrangement to form a staggered tetramer, which is the fundamental building block.
    - **Filament Assembly:** These tetramers then assemble end-to-end and laterally to form the final 10 nm wide filament. Unlike actin and microtubules, IF assembly does not require ATP or GTP hydrolysis, relying instead on hydrophobic interactions.
- **Classification:** IFs are classified into six main types based on their protein composition and cell/tissue distribution:
    - **Type I & II:** Keratins (acidic and basic/neutral) - found in epithelial cells (skin, hair, nails).
    - **Type III:** Vimentin (mesenchymal cells, fibroblasts, leukocytes), Desmin (muscle cells), GFAP (astrocytes, glial cells), Peripherin (peripheral neurons).
    - **Type IV:** Neurofilaments (L, M, H subunits) - found in neurons, particularly in axons.
    - **Type V:** Lamins - form the nuclear lamina, supporting the inner nuclear membrane in most eukaryotic cells.
    - **Type VI:** Nestin (neural stem cells).
- **Functions:**
    - **Mechanical Strength:** The primary function is to provide tensile strength, protecting cells from mechanical stress and rupture.
    - **Structural Support:** They help maintain cell shape and the spatial organization of organelles within the cytoplasm.
    - **Nuclear Integrity:** Nuclear lamins provide structural support to the nuclear envelope and play a role in chromatin organization.

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- **Tissue Cohesion:** In epithelial cells, keratins link to desmosomes and hemidesmosomes, connecting cells to each other and to the extracellular matrix, thereby strengthening tissues.
- **Axonal Stability:** Neurofilaments are abundant in the axons of neurons, providing structural support to maintain their shape and integrity over long distances.

- **Contribution to Cellular Structural Integrity:**
  - Intermediate filaments are crucial for cellular structural integrity due to their remarkable tensile strength and resistance to stretching. They form a resilient, flexible network throughout the cytoplasm, acting as an internal scaffolding that withstands mechanical forces. By linking to specialized cell junctions (desmosomes and hemidesmosomes), IFs effectively distribute these forces across multiple cells and anchor cells to their surrounding extracellular matrix. This interconnected "rope-like" system prevents individual cells from tearing apart under mechanical stress and provides overall structural stability to tissues, especially those subjected to significant physical forces like the skin, muscles, and nerves. Genetic defects in IF proteins can lead to severe blistering diseases (e.g., Epidermolysis bullosa simplex), highlighting their critical role in maintaining tissue cohesion.

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5 (b) Describe the role of extracellular and intracellular signaling molecules and receptors in cellular communication. Give an example of any extracellular receptor and its mechanisms of action.

- Cellular communication, or cell signaling, enables cells to receive, process, and respond to information from their environment and other cells. This process relies on specific signaling molecules and their corresponding receptors.
- **Role of Signaling Molecules:**

- **Extracellular Signaling Molecules (First Messengers/Ligands):**
    - These are the initial messengers that transmit information between cells. They are diverse in nature and include hormones (long-distance), neurotransmitters (short-distance, synaptic), growth factors (local, paracrine), and cytokines.
    - Their primary role is to carry the signal from the signaling cell to the target cell, binding to specific receptors on or within the target cell.
- **Intracellular Signaling Molecules (Second Messengers):**
    - These are small, non-protein molecules or ions generated or released *within* the cytoplasm in response to the activation of an extracellular receptor. They amplify and relay the signal from the cell membrane to various intracellular targets.
    - Examples include Cyclic AMP (cAMP), Calcium ions (Ca^{2+}), Inositol Trisphosphate (IP_3), and Diacylglycerol (DAG).
    - Their role is to propagate and amplify the signal inside the cell, triggering a cascade of events that ultimately leads to the desired cellular response.

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- **Role of Receptors:**
    - **Extracellular Receptors (Cell-Surface Receptors):**
        - These are transmembrane proteins embedded in the plasma membrane. They bind to hydrophilic extracellular signaling molecules that cannot readily cross the cell membrane.

        - They act as signal transducers, converting the extracellular signal into an intracellular signal, initiating a cascade of events inside the cell.
        - Examples: G protein-coupled receptors (GPCRs), enzyme-linked receptors (like Receptor Tyrosine Kinases), and ion-channel-linked receptors.
    - **Intracellular Receptors:**
        - These are located in the cytoplasm or nucleus. They bind to small, hydrophobic signaling molecules (e.g., steroid hormones) that can diffuse directly across the plasma membrane.
        - Upon ligand binding, these receptors often function as transcription factors, directly regulating gene expression in the nucleus.

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- **Example of an Extracellular Receptor: Receptor Tyrosine Kinase (RTK)**
    - **Structure:** RTKs are enzyme-linked receptors typically composed of an extracellular ligand-binding domain, a single transmembrane helix, and an intracellular domain possessing intrinsic tyrosine kinase enzyme activity.
    - **Mechanism of Action (e.g., Epidermal Growth Factor Receptor - EGFR):**
        - **1. Ligand Binding and Dimerization:** When a specific extracellular signaling molecule (e.g., Epidermal Growth Factor, EGF) binds to the extracellular domains of two RTK monomers, it induces them to associate and form a dimer.
        - **2. Autophosphorylation:** The dimerization brings the intracellular tyrosine kinase domains of the two receptors into close proximity, enabling them to cross-

phosphorylate each other. They phosphorylate specific tyrosine residues on each other's cytoplasmic tails, utilizing ATP.

- **3. Creation of Binding Sites:** These newly phosphorylated tyrosine residues act as high-affinity docking sites for various intracellular signaling proteins. These signaling proteins typically contain specialized domains (e.g., SH2 domains) that specifically recognize and bind to phosphorylated tyrosines.
- **4. Downstream Signaling Cascade:** The binding of these intracellular signaling proteins activates them, initiating a cascade of events. A common pathway involves the activation of the **Ras-MAP kinase pathway**. An adaptor protein (e.g., Grb2) binds to the phosphorylated RTK, recruiting a guanine nucleotide exchange factor (GEF, e.g., Sos). Sos then activates a small G protein called Ras by promoting the exchange of GDP for GTP. Activated Ras then triggers a phosphorylation cascade involving a series of protein kinases (e.g., Raf, MEK, ERK/MAP kinase).
- **5. Cellular Response:** Ultimately, the activated terminal kinase (e.g., ERK) translocates to the nucleus and phosphorylates transcription factors, leading to changes in gene expression that mediate the specific cellular response, such as cell proliferation, differentiation, or survival.

- RTKs are vital for regulating cell growth, division, differentiation, and survival, and their dysregulation is frequently implicated in various diseases, including cancer.

6 (a) Role of centrioles in cell division.

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- Centrioles are small, cylindrical organelles typically found in pairs within the centrosome, the primary microtubule-organizing center (MTOC) in most animal cells and some lower plant cells. While not universally essential for cell division (some eukaryotes can divide without them), they play significant roles in organizing the spindle poles during animal cell division.
- **Key Roles in Mitosis (Animal Cells):**
    - **Centrosome Duplication and Separation:** During interphase, the two centrioles within the centrosome duplicate. At the onset of mitosis (prophase), these two centrosomes migrate to opposite poles of the cell.
    - **Microtubule Nucleation:** The centrosomes, with their associated pericentriolar material (PCM), act as the main sites for nucleating and organizing the microtubules that form the **mitotic spindle fibers**. These microtubules extend outwards from the poles.
    - **Spindle Pole Establishment:** The movement of the centrosomes to opposite poles establishes the two poles of the mitotic spindle, which is crucial for the bipolar organization required for accurate chromosome segregation.
    - **Asters Formation:** Around each centrosome, a radial array of short microtubules called the aster forms, contributing to the positioning and orientation of the spindle within the cell.
    - **Chromosome Segregation:** While centrioles themselves do not directly attach to chromosomes, the microtubules nucleated from the centrosomes (kinetochore microtubules) attach to the kinetochores of sister chromatids. The forces generated by these microtubules, along with interpolar microtubules, ensure the precise separation of sister chromatids to opposite poles during anaphase.

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    - **Cytokinesis:** The position of the mitotic spindle, determined by the centrosomes, often dictates the plane of cell division during cytokinesis.

- In essence, centrioles, as part of the centrosome, are crucial for assembling and organizing the mitotic spindle apparatus that ensures accurate chromosome distribution to daughter cells during cell division in many eukaryotic organisms.

6 (b) cAMP as a second messenger in the signal transduction pathway.

- Cyclic Adenosine Monophosphate (cAMP) is a critical **second messenger** molecule in numerous intracellular signal transduction pathways. It acts as an intermediary, relaying and amplifying signals initiated by extracellular signaling molecules (first messengers) binding to cell-surface receptors.
- **Generation of cAMP:**
    - The most common pathway for cAMP generation involves **G protein-coupled receptors (GPCRs)**.
    - When a ligand (e.g., hormone, neurotransmitter) binds to and activates a GPCR, it leads to the activation of an associated heterotrimeric G protein.
    - Specifically, the Gα subunit of the G protein exchanges GDP for GTP and dissociates from the Gβγ subunits.
    - The activated Gα subunit (GTP-bound) then activates a membrane-bound enzyme called **adenylyl cyclase**.
    - Adenylyl cyclase catalyzes the conversion of ATP into cAMP, releasing pyrophosphate.
- **Mechanism of Action (Signal Relay and Amplification):**
    - Once generated, cAMP diffuses rapidly through the cytoplasm. Its primary target is usually **cAMP-dependent protein kinase (PKA)**, also known as protein kinase A.

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- PKA exists in an inactive tetrameric form, comprising two regulatory (R) subunits and two catalytic (C) subunits.
- When cAMP binds to the regulatory subunits of PKA, it causes a conformational change that leads to the dissociation and activation of the two catalytic subunits.
- The now active catalytic subunits of PKA proceed to phosphorylate specific serine or threonine residues on various target proteins (enzymes, ion channels, transcription factors) within the cell, using ATP. This phosphorylation alters the activity of these target proteins, ultimately leading to a diverse range of cellular responses.

- **Amplification and Diversity:**
  - A single activated GPCR can activate multiple G proteins.
  - Each activated G protein can activate multiple adenylyl cyclase enzymes.
  - Each adenylyl cyclase can produce a large number of cAMP molecules.
  - Each cAMP molecule can activate PKA.
  - Each activated PKA can phosphorylate numerous target proteins. This multi-step cascade results in significant **signal amplification**.
  - The same cAMP signal can elicit different responses in different cell types because PKA phosphorylates different target proteins in those cells, leading to varied physiological outcomes (e.g., epinephrine in liver cells promotes glycogen breakdown, while in heart cells it increases heart rate).
- **Signal Termination:** cAMP signaling is rapidly terminated by the enzyme **cAMP phosphodiesterase**, which hydrolyzes cAMP back

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into inactive 5'-AMP, ensuring that the cellular response is tightly controlled and transient.

6 (c) Importance of the G1/S checkpoint in the cell cycle.

- The cell cycle is precisely regulated by a series of checkpoints that ensure proper progression and prevent errors. The **G1/S checkpoint**, also known as the "restriction point" (in mammalian cells) or "start" (in yeast), is considered one of the most critical regulatory points. It occurs at the end of the G1 phase, just before the cell enters the S phase (DNA synthesis).
- **Importance:**
    - **Commitment to Cell Division:** This checkpoint represents a crucial decision point. Once a cell passes the G1/S checkpoint, it is generally committed to completing the rest of the cell cycle (S, G2, and M phases), even if external growth signals are later withdrawn. Cells that do not receive the necessary signals or fail to meet the criteria typically exit the cycle and enter a quiescent state called G0.
    - **Integration of External Signals:** The cell assesses its external environment at this checkpoint. It checks for the presence of sufficient growth factors (mitogens) that stimulate cell division and adequate nutrient availability. Without these, cell cycle progression is halted.
    - **DNA Integrity Surveillance:** A vital function of the G1/S checkpoint is to monitor the integrity of the cell's DNA. If any DNA damage is detected during the G1 phase, the cell cycle will be arrested at this point. This pause provides critical time for DNA repair mechanisms to fix the damage before DNA replication commences in S phase. This prevents the replication of damaged DNA, which could lead to harmful mutations or chromosomal aberrations in daughter cells.

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- **Cell Size and Resource Sufficiency:** The checkpoint also ensures that the cell has grown to an adequate size and has accumulated sufficient resources (e.g., proteins, nucleotides) required for DNA replication and subsequent cell division.
- **Prevention of Uncontrolled Proliferation (Tumor Suppression):** The G1/S checkpoint acts as a crucial barrier against uncontrolled cell growth and division. Key regulatory proteins involved in this checkpoint, such as the tumor suppressor proteins p53 and Rb (retinoblastoma protein), play a central role in preventing cells with damage or inappropriate signals from dividing. Dysregulation or mutations in these checkpoint components are frequently observed in various cancers, highlighting its critical role in maintaining genomic stability and preventing tumor formation.

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